Revised 12-Month Finding on a Petition to List the Upper Missouri River Distinct Population Segment of Arctic Grayling

SUMMARY: We, the U.S. Fish and Wildlife Service (Service), announce a revised 12-month finding on a petition to list Upper Missouri River distinct population segment (Upper Missouri River DPS) of Arctic grayling (*Thymallus arcticus*) as an endangered or threatened species under the Endangered Species Act of 1973, as amended (Act). After a thorough review of the best available scientific and commercial information, we find that it is not warranted at this time to list the Upper Missouri River DPS of arctic grayling. However, we ask the public to submit to us at any time any new information relevant to the status of the DPS or its habitat.

SUPPLEMENTARY INFORMATION:

Background

Under section 4(b)(3)(B) of the Act (16 U.S.C. 1531 *et seq.*), we are required to make a finding whether or not a petitioned action is warranted within 12 months after receiving any petition that we have determined contained substantial scientific or commercial information indicating that the petitioned action may be warranted (‘‘12-month finding’’). We must make a finding that the petitioned action is: (1) Not warranted; (2) warranted; or (3) warranted but precluded. ‘‘Warranted but precluded’’ means that (a) the petitioned action is warranted, but the immediate proposal of a regulation implementing the petitioned action is precluded by other pending proposals to determine whether species are endangered or threatened species, and (b) expeditious progress is being made to add qualified species to the Lists of Endangered and Threatened Wildlife and Plants (Lists) and to remove from the Lists species for which the protections of the Act are no longer necessary. Section 4(b)(3)(C) of the Act requires that we treat a petition for which the requested action is found to be warranted but precluded as though resubmitted on the date of such finding, that is, requiring that a subsequent finding be made within 12 months of that date.

Previous Federal Actions

We have published a number of documents on Arctic grayling since 1982, and have been involved in litigation over previous findings. We describe previous federal actions that are relevant to this document below.

We published our first status review for the Montana Arctic grayling (*Thymallus arcticus montanus*), then thought to be a subspecies of Arctic grayling, in a **Federal Register** document on December 30, 1982 (47 FR 58454). In that document, we designated the purported subspecies, Montana Arctic grayling, as a Category 2 species. At that time, we designated a species as Category 2 if a listing as endangered or threatened was possibly appropriate, but we did not have sufficient data to support a proposed rule to list the species.

On October 9, 1991, the Biodiversity Legal Foundation and George Wuerthner petitioned us to list the fluvial (riverine) populations of Arctic grayling in the Upper Missouri River basin as an endangered species throughout its historical range in the coterminous United States. We published a notice of a 90-day finding in the January 19, 1993, **Federal Register** (58 FR 4975), concluding the petitioners presented substantial information indicating that listing the fluvial Arctic grayling of the Upper Missouri River in Montana and northwestern Wyoming may be warranted. This finding also noted that taxonomic recognition of the Montana Arctic grayling (*Thymallus arcticus montanus*) as a subspecies (previously designated as a category 2 species) was not widely accepted, and that the scientific community generally considered this population a geographically isolated member of the wider species (*T. arcticus*).

On July 25, 1994, we published a notice of a 12–month finding in the **Federal Register** (59 FR 37738), concluding that listing the DPS of fluvial Arctic grayling in the Upper Missouri River was warranted but precluded by other higher priority listing actions. This DPS determination predated our DPS policy (61 FR 4722, February 7, 1996), so the entity did not undergo a DPS analysis as described in the policy. The 1994 finding placed fluvial Arctic grayling of the Upper Missouri River on the candidate list and assigned it a listing priority of 9, indicating that the threats were imminent but of moderate to low magnitude.

On May 31, 2003, the Center for Biological Diversity and Western Watersheds Project (Plaintiffs) filed a complaint in U.S. District Court in Washington, D.C., challenging our 1994 "warranted but precluded" determination for the DPS of fluvial Arctic grayling in the Upper Missouri River basin. On May 4, 2004, we elevated the listing priority number of the fluvial Arctic grayling to 3 (69 FR 24881), indicating threats that were imminent and of high magnitude. On July 22, 2004, the Plaintiffs amended their complaint to challenge our failure to emergency list this population. We settled with the Plaintiffs in August 2005, and we agreed to submit a revised determination on whether this population warranted listing as endangered or threatened to the **Federal Register** on or before April 16, 2007.

On April 24, 2007, we published a revised 12-month finding on the petition to list the Upper Missouri River DPS of fluvial Arctic grayling (72 FR 20305) ("2007 finding"). In this finding, we determined that fluvial Arctic grayling of the upper Missouri River did not constitute a species, subspecies, or DPS under the Act. Therefore, we found that the upper Missouri River population of fluvial Arctic grayling was not a listable entity under the Act, and, as a result, listing was not warranted. With that document, we withdrew the fluvial Arctic grayling from our candidate list.

On November 15, 2007, the Center for Biological Diversity, Federation of Fly Fishers, Western Watersheds Project, George Wuerthner, and Pat Munday filed a complaint (CV-07-152, in the District Court of Montana) to challenge our 2007 finding. We settled this litigation on October 5, 2009. In the stipulated settlement, we agreed to: (a) Publish, on or before December 31, 2009, a document in the **Federal Register** soliciting information on the status of the upper Missouri River Arctic grayling; and (b) submit, on or before August 30, 2010, a new 12-month finding for the upper Missouri River Arctic grayling to the **Federal Register**.

On October 28, 2009, we published in the **Federal Register** a notice of intent to conduct a status review of Arctic grayling (*Thymallus arcticus*) in the upper Missouri River system (74 FR 55524). To ensure the status review was based on the best available scientific and commercial data, we requested information on the taxonomy, biology, ecology, genetics, and population status of the Arctic grayling of the upper Missouri River system; information relevant to consideration of the potential DPS status of Arctic grayling of the upper Missouri River system; threats to the species; and conservation actions being implemented to reduce those threats in the upper Missouri River system. That document further specified that the status review might consider various DPS designations that include different life histories of Arctic grayling in the upper Missouri River system and different DPS configurations, including fluvial, adfluvial (lake populations), or all life histories of Arctic grayling in the upper Missouri River system.

On September 8, 2010, we published a revised 12-month finding on the petition to list the Upper Missouri River DPS of Arctic grayling (75 FR 54708) ("2010 finding"). In this finding, we determined that fluvial and adfluvial Arctic grayling of the upper Missouri River did constitute a DPS under the Act. Further, we found that a DPS configuration including both adfluvial and fluvial life histories was the most appropriate for the long-term conservation of Arctic grayling because genetic evidence indicated that fluvial and adfluvial life-history forms did not represent distinct evolutionary lineages. We concluded by finding that the Upper Missouri River DPS of Arctic grayling was warranted for listing under the Act, but precluded by other higher priority listing actions.

On September 9, 2011, we reached an agreement with plaintiffs in Endangered Species Act Section 4 Deadline Litig., Misc. Action No. 10–377 (EGS), MDL Docket No. 2165 (D. DC) (known as the "MDL case") on a schedule to publish proposed listing rules or not-warranted findings for the species on our candidate list. This agreement stipulated that we would submit for publication in the **Federal Register** either a proposed listing rule for the Upper Missouri River DPS of Arctic grayling, or a not-warranted finding, no later than the end of Fiscal Year 2014.

On November 26, 2013, we published a document in the **Federal Register** (78 FR 70525) notifying the public that we were initiating a status review of the Upper Missouri River DPS of Arctic grayling to determine whether the entity meets the definition of an endangered or threatened species under the Act. That document requested general information (taxonomy, biology, ecology, genetics, and status) on the Arctic grayling of the upper Missouri River system, as well as information on the conservation status of, threats to, planned and ongoing conservation actions for, habitat selection of, habitat requirements of, and considerations concerning the possible designation of critical habitat for the Arctic grayling of the upper Missouri River system.

On August 20, 2014, we published a revised 12-month finding on the petition to list the Upper Missouri River DPS of Arctic grayling (79 FR 49384), fulfilling our commitments under the MDL case. In this document, we announced our finding that listing the DPS was not warranted, and removed the DPS from the candidate list. We concluded that habitat-related threats previously identified, including habitat fragmentation, dewatering, thermal stress, entrainment, riparian habitat loss, and effects from climate change, had been sufficiently ameliorated and that 19 of 20 populations of Arctic grayling were either stable or increasing.

On February 5, 2015, the Center for Biological Diversity (CBD), Western Watersheds Project, and two individuals filed a complaint against the Department of the Interior (DOI) and the Service challenging our August 20, 2014, revised 12-month finding that the Upper Missouri River DPS of Arctic grayling did not warrant listing as a threatened species or endangered species (*Center for Biological Diversity v. Jewell*, No. 2:15-cv-00004-SEH, 2016 WL 4592199 (D. Mont. 2016)). Plaintiffs also brought a facial challenge to the Service's Final Policy on Significant Portion of its Range (SPR Policy; 79 FR 37578), arguing that the SPR Policy was contrary to case law in defining a species' range to only include current range and not historical range. The district court found for the government on all claims, and the plaintiffs appealed.

On August 17, 2018, the Court of Appeals affirmed in part and reversed in part (*Center for Biological Diversity v. Zinke*, No. 16-35866, 900 F. 3d 1053 (9th Cir. 2018)). The court agreed with the district court that we permissibly defined "range" as current range in the SPR Policy. But that court found that we erred in the listing finding in four ways: (1) we should not have concluded that the Big Hole River grayling population was increasing when available biological information showed that the population was declining; (2) we should not have relied on cold water refugia in the Big Hole River, because information showed that the River will experience low stream flows and high water temperatures, and we did not adequately address it; (3) we did not adequately explain why the uncertainty presented by climate change with regard to low stream flows and higher water temperatures did not weigh in favor of listing the grayling; and (4) we arbitrarily relied on the Ruby River grayling population to provide redundancy for the grayling outside of the Big Hole River. The court upheld the finding in all other respects, including our analysis of cold water refugia other than in the Big Hole River, and our conclusion that small population size did not pose a risk to genetic viability of the grayling.

The court vacated the finding and remanded it to us to reconsider in light of the court's opinion, and ordered that we make one of the findings set forth in 16 U.S.C. § 1533(b)(3)(B)(i)-(iii) for the Upper Missouri River Valley DPS. Further, we are required to submit such finding to the Office of the Federal Register no later than July 1, 2020. Our revised finding is published in the Federal Register, and explained in further detail in this document.

We note that this document is intended to mirror the 2014 finding in structure and format, and retains some language from the 2014 finding where still applicable. In this document we also update our evaluation with new scientific information that has become available since 2014, and present our reevaluation of the status of the DPS under the Act. While using this format (i.e., a five-factor analysis) is no longer the Service's typical practice in listing determinations, this format was used in order to clearly convey to readers how we have addressed the four points raised by the Court regarding the 2014 finding. The Service's current practice is to compile scientific information regarding species in a separate Species Status Assessment (Smith et al. 2018). However, in this case, for continuity with all previous review documents for this species, all scientific information and analysis regarding the Upper Missouri River DPS of arctic grayling is summarized within this document, using a five-factor analysis structure, as was done in the 2014 finding.

Summary of Changes and New Information

Biological information on Arctic grayling in the Upper Missouri River basin is continually being collected and updated. We have included the best available information in this status review. This updated biological information includes demographic and genetic information that helps characterize the current status and future adaptive potential of some of the 19 populations of Arctic grayling currently included in the DPS. In addition, new biological observations have increased our understanding of the varied life history of Arctic grayling and demonstrated that life history behaviors occur along a spectrum, instead of the discrete life history types we have described in the past. We also provide more detailed explanation of information about the Big Hole River population (see Population Status and Trends in the Upper Missouri River DPS), Big Hole River water temperature regimes (see Dewatering from Irrigation and Increased Water Temperatures), Ruby River population (see Population Status and Trends in the Upper Missouri River DPS), and the cumulative effects of climate change (see Climate Change/Dewatering/Water Temperature Increases) in response to a recent court remand.

Species Information

Taxonomy and Species Description

The Arctic grayling (*Thymallus arcticus*) is a fish belonging to the family Salmonidae (salmon, trout, charr, whitefishes), subfamily *Thymallinae* (graylings), and it is represented by a single genus, *Thymallus*. Arctic grayling have elongate, laterally compressed, trout-like bodies with deeply forked tails, and adults typically average 300-380 millimeters (mm) (12-15 inches (in.)) in length. Coloration can be striking, and varies from silvery or iridescent blue and lavender, to dark blue (Behnke 2002, pp. 327–328). The sides are marked with a varying number of V-shaped or diamond-shaped spots (Scott and Crossman 1988, p. 301). During the spawning period, the colors darken and the males become more brilliantly colored than the females. A prominent morphological feature of Arctic grayling is the sail-like dorsal fin, which is large and vividly colored with rows of orange to bright green spots, and often has an orange border (Behnke 2002, pp. 327–328).

Distribution

Arctic grayling are native to Arctic Ocean drainages of Alaska and northwestern Canada, as far east as Hudson's Bay, and westward across northern Eurasia to the Ural Mountains (Scott and Crossman 1998, pp. 301–302; Froufe *et al.* 2005, pp. 106–107; Weiss *et al.* 2006, pp. 511–512). In North America, they are native to northern Pacific Ocean drainages as far south as the Stikine River in British Columbia (Nelson and Paetz 1991, pp. 253–256; Behnke 2002, pp. 327–331).

Distribution in the Conterminous United States

Two disjunct groups of Arctic grayling were native to the conterminous United States: One in the upper Missouri River basin in Montana and Wyoming; and another in Michigan that was extirpated in the late 1930s (Hubbs and Lagler 1949, p. 44), and has not been detected since.

During the status review process, the Service received information indicating that Arctic grayling may have also been native to areas outside the Upper Missouri River basin in Montana and Wyoming. This information included multiple historical newspaper clippings and several reports from early Army expeditions purporting that Arctic grayling were captured in the Yellowstone River drainage in Montana and the Snake River drainage in Idaho (Shea 2014, entire). Some of these reports even included descriptions of captured fish. However, none of the descriptions mentions the colorful, sail-like dorsal fin of Arctic grayling, a prominent feature that clearly distinguishes Arctic grayling from other salmonids. In addition, a similar species resembling Arctic grayling (i.e., mountain whitefish) is native to both the Yellowstone River drainage and Snake River drainage. Mountain whitefish were sometimes referred to as "grayling" in some areas of the West (Ellis 1914, p. 75). Thus, it is likely that early reports of Arctic grayling occurring outside the upper Missouri River basin were mountain whitefish misidentified as Arctic grayling. Therefore, without information to the contrary, we consider Arctic grayling to be native only to the upper Missouri River basin in Montana and Wyoming and to Michigan.

Native Distribution of Arctic Grayling in the Upper Missouri River Basin

The first Euro-American "discovery" of Arctic grayling in North America is attributed to members of the Lewis and Clark Expedition, who encountered the species in the Beaverhead River in August 1805 (Nell and Taylor 1996, p. 133). Vincent (1962, p. 11) and Kaya (1992, pp. 47–51) synthesized accounts of Arctic grayling occurrence and abundance from historical surveys and contemporary monitoring to estimate the historical distribution of the species in the upper Missouri River system (Figure 1). We base our conclusions on the historical distribution of Arctic grayling in the upper Missouri River basin on these two reviews.

Arctic grayling were widely but irregularly distributed in the upper Missouri River system above the Great Falls in Montana and in northwest Wyoming within the present-day boundaries of Yellowstone National Park (Vincent 1962, p. 11). They may have inhabited up to 2,000 kilometers (km) (1,250 miles (mi)) of stream habitat until the early 20th century, based on early observations (Kaya 1992, pp. 47–51); however, extrapolation used to produce this estimate is based on multiple assumptions with unknown validity (Kaya 1992, p. 51). Arctic grayling were reported in the mainstem Missouri River, as well as in the Smith, Sun, Jefferson, Madison, Gallatin, Big Hole, Beaverhead, and Red Rock Rivers (Vincent 1962, p. 11; Kaya 1992, pp. 47–51; USFWS 2007; 72 FR 20307, April 24, 2007). Anecdotal accounts report that the species may have been present in the Ruby River, at least seasonally (Magee 2005, pers. comm.), and were observed there as recently as the early 1970s (Holton, undated). Arctic grayling also occurred in multiple lakes in the upper Missouri River basin. For example, Arctic grayling are native to Red Rock Lake in the Centennial Valley (Vincent 1962, pp. 112–121; Kaya 1992, p. 47). Vincent (1962, p. 120) stated that Red Rock Lakes were the only natural lakes in the upper Missouri River basin accessible to colonization by Arctic grayling. However, Arctic grayling appear to also be native to Elk Lake (in the Centennial Valley; Kaya 1990, p. 44) and a few lakes (Miner and Mussigbrod lakes) in the upper Big Hole River drainage, based on recent genetic information (Peterson and Ardren 2009, p. 1768).

The distribution of native Arctic grayling in the upper Missouri River has been reduced since the late 19th century, particularly in riverine habitats (Vincent 1962, pp. 86–90, 97–122, 127–129; Kaya 1992, pp. 47–53). The populations that formerly resided in the Smith, Sun, Jefferson, Beaverhead, Gallatin, and mainstem Missouri Rivers are considered extirpated; populations (or parts of populations) that remain in riverine habitats include those found in the Big Hole River and its tributaries, the Centennial Valley, the Madison River, and the Ruby River (Gander et al. 2019, p. 4; Cayer 2014a, pers. comm.; MFISH 2014b, unpublished data; Kaya 1992, pp. 51–53). Other populations in the upper Missouri River that occur within known historically occupied habitat reside in Miner and Mussigbrod lakes in the upper Big Hole River system (Peterson and Ardren 2009, pp. 1762, 1768).

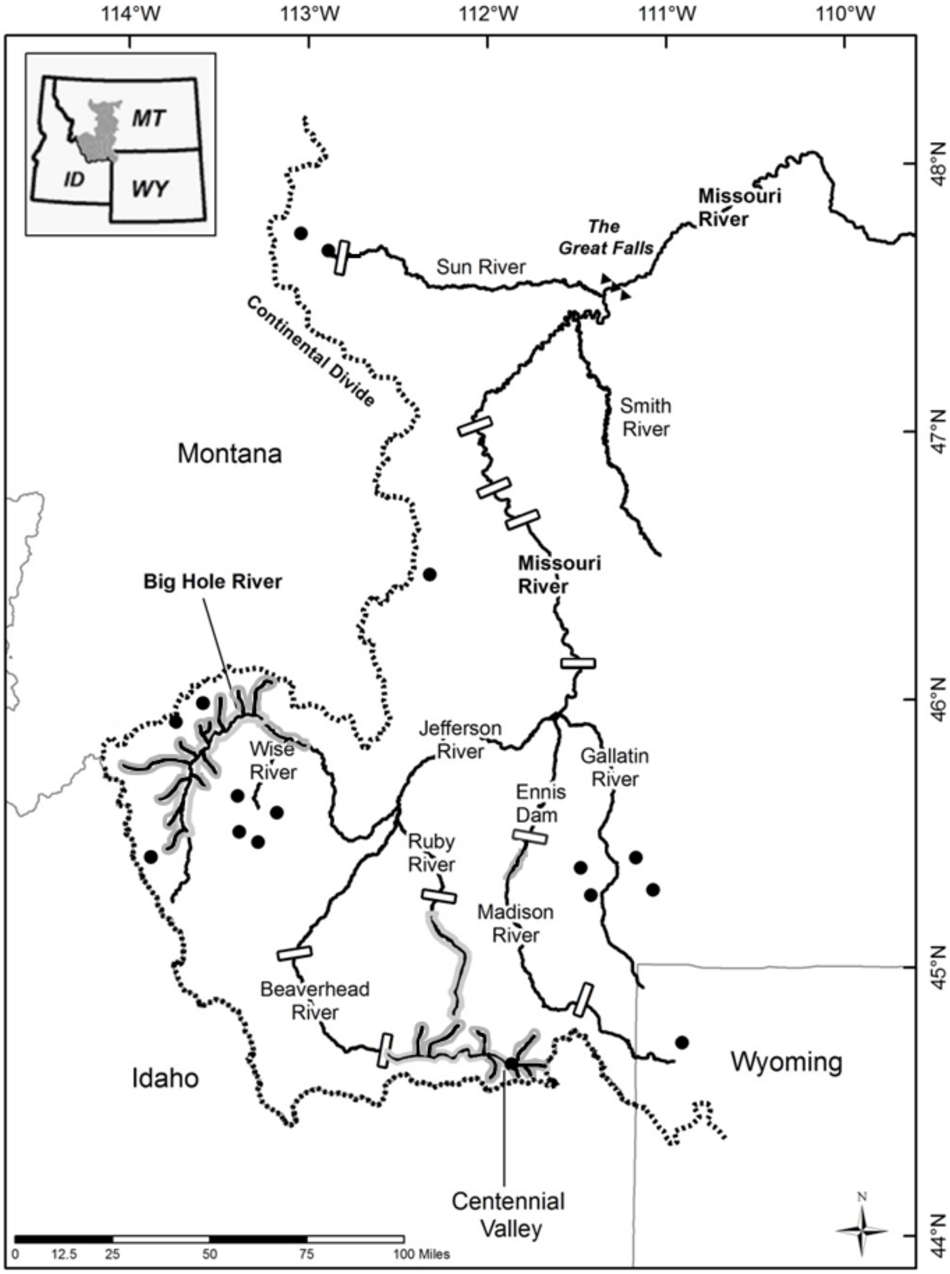


Figure 1. Approximate current distribution [gray outlines (rivers/streams) and black circles (lakes)] of Arctic grayling in the upper Missouri River basin. Small rectangles denote locations of mainstem river dams. Some smaller occupied tributaries in the Big Hole River and Centennial Valley are not shown due to the broad scale of the map.

Introduced Arctic Grayling in the Upper Missouri River Basin

From 1898 through the 1960s, an estimated 100 million Arctic grayling were stocked across Montana and other western States. The sources of these stockings varied through time as different State, Federal, and private hatchery operations were created, but the ultimate source for all hatcheries in Montana appears to be stock from two Montana populations: Centennial Valley and Madison River (Peterson and Ardren 2009, p. 1767; Leary 2014, unpublished data; MFISH 2014a). Arctic grayling derived from these two sources were stocked on top of every known native Arctic grayling population in the upper Missouri River basin. In addition, Arctic grayling were stocked in multiple high elevation lakes, some of which likely were historically fishless.

There are 19 known, introduced Arctic grayling populations that exist in the upper Missouri River basin that meet our criteria for inclusion in the DPS. Criteria for inclusion in the DPS are that populations must: (1) occupy natural habitat; (2) reproduce naturally, and (3) not be part of the captive brood (genetic) reserve program. In the 2014 Finding, one of the criteria we used to determine whether a population was considered part of the DPS or not was whether the population was "self-sustaining". Our intent with this term was to make the distinction between a population that reproduces naturally and has natural spawning habitat available versus one that is the result of a stocking effort where no natural reproduction occurs (e.g., Tunnel Lake). However, an unintended consequence of using the term "self-sustaining" was that it could appear we were prejudging the status of a population. To correct this, yet maintain the original intent of distinction, we have used the term "naturally reproducing" in this status review in place of "self-sustaining" when referring to criteria for inclusion into the DPS. Also in 2014, we stated that there were 20 populations of Arctic grayling in the DPS; one being the population in Diversion Lake, based in part on discussions from our expert elicitation workshop in 2014. However, we are currently not aware of any evidence of natural reproduction in Diversion Lake, thus we have determined that we erroneously included that population in the DPS in 2014. We do not include the Diversion Lake population in the current DPS as part of this status review for that reason. In this status review, we primarily focus our analysis on the 19 populations that meet our criteria for inclusion in the DPS.

Many other populations of Arctic grayling exist in the Upper Missouri River basin but are not included in the DPS because they do not meet one or more of these same criteria (Table 1). These populations could be considered for inclusion in the DPS in the future if monitoring indicated that they met all three of our inclusion criteria.

Table 1. Arctic grayling populations not currently considered part of the Upper Missouri River Distinct Population Segment of Arctic grayling and criteria not met for inclusion.

Population	Criteria not met
Axolotl Lake	Captive brood
Green Hollow Lake	Captive brood
Elk Lake	Captive brood
Handkerchief Lake	Captive brood
Sunnyslope Canal	Unnatural habitat (irrigation canal)
Diversion Lake	No known natural reproduction
Tunnel Lake	No known natural reproduction
South Fork Sun River	No known natural reproduction
Grayling Creek	No known natural reproduction
Cherry Creek	No known natural reproduction
Goose Lake	No known natural reproduction
Wolf Lake	No known natural reproduction
Ice Lake	No known natural reproduction
Spanish Creek/Gallatin River	No known natural reproduction
Gibbon River/Upper Madison River	No known natural reproduction
Odell Creek, tributary to Madison River	No known natural reproduction

Of the 19 populations of Arctic grayling in the Upper Missouri River DPS, there are 13 populations that are likely the result of historical stocking (Table 2). In our previous finding in 2010, we considered and discussed the conservation value of these populations. Based on the information available at that time, we considered these introduced populations to not have conservation value for multiple reasons. Below, we list each of the reasons for this conclusion in italics (discussed in both our 2010 and 2014 findings) and provide an updated assessment and conclusion (in non-italic text) about the potential conservation value of these populations, based on new information.

1. *The Service interprets the Act to provide a statutory directive to conserve species in their native ecosystems (49 FR 33885, August 27, 1984) and to conserve genetic resources and biodiversity over a representative portion of a taxon's historical occurrence (61 FR 4722, February 7, 1996). Since most of the introduced populations of Arctic grayling were of unknown genetic origin and in lakes that were likely historically fishless, these populations were considered in 2010 to be outside the species' native range, and we concluded that they did not appear to add conservation value to the DPS.*

Since 2010, new genetic information from 7 of the 14 introduced populations indicates there are moderate to high levels of genetic diversity within and among these populations, and indicates these populations were derived from native sources within the upper Missouri River basin (Leary 2014, unpublished data; Table 2). In addition, stocking records show common stocking sources for introduced populations that were genotyped (as described previously) and the two populations that were not genotyped (the remaining 2 populations were reintroductions of known Montana origin sources; Table 2). Thus, it appears that all 13 introduced Arctic grayling populations contain moderate to high levels of genetic diversity of Arctic grayling from the upper Missouri River

Table 2. Geographic distribution, genetic status, and source of introduced Arctic grayling populations in the upper Missouri River basin.

Population	Drainage	Genetic analysis completed?	Source[a]	Citation
Agnes Lake	Big Hole	No	Madison/Centennial	MFISH 2014a
Odell Lake	Big Hole	Yes	Centennial	Peterson and Ardren 2009, p. 1766; Leary 2014, unpublished data
Bobcat Lake	Big Hole	Yes	Centennial	Peterson and Ardren 2009, p. 1766; Leary 2014, unpublished data
Schwinegar Lake	Big Hole	No	Madison/Centennial[c]	-
Pintlar Lake	Big Hole	Yes	Madison/Centennial	Leary 2014, unpublished data
Deer Lake	Gallatin	Yes	Madison/Centennial	Leary 2014, unpublished data
Emerald Lake	Gallatin	Yes	Madison/Centennial	Leary 2014, unpublished data
Grayling Lake	Gallatin	Yes	Madison/Centennial	Leary 2014, unpublished data
Hyalite Lake	Gallatin	Yes	Madison/Centennial	Leary 2014, unpublished data
Gibson Reservoir	Sun	Yes[b]	Big Hole	Horton 2014a, pers. comm.; Magee 2014, pers. comm.
Lake Levale	Sun	Yes[b]	Big Hole	Horton 2014a, pers. comm.; Magee 2014, pers. comm.
Park Lake	Missouri	No	Madison/Centennial[d]	-
Grebe Lake	Madison	Yes	Centennial	Peterson and Ardren 2009, p. 1766; Varley 1981, p. 11

[a]Origin of source stock was determined by genetic analysis and through analysis of historical stocking records and scientific literature, in some cases. Where multiple sources are cited, fish from each population were known to be stocked, although the genetic contribution of each donor population to the current population structure is unknown.
[b]These populations are the result of reintroductions using known sources of Montana origin.
[c]Schwinegar Lake Arctic grayling population is likely from Montana-origin sources due to proximity to other lakes with known Montana origin; however, definitive evidence is lacking.
[d]Park Lake stocking records show Montana-origin stockings, despite genetic analysis not being complete yet.

basin that was not captured within the DPS designation in the 2010 finding.

The Service's interpretation of the Act is consistent with that in the 2010 finding; we believe it is important to conserve species in their native ecosystems and to conserve genetic resources and biodiversity over a representative portion of a taxon's historical occurrence. In light of the new genetics information gained since 2010 (Leary 2014, unpublished data), we also believe it is important to acknowledge the moderate to high levels of genetic diversity within the introduced populations in the upper Missouri River basin and the potential adaptive capabilities represented by this diversity. All Arctic grayling populations (introduced or not), currently within the upper Missouri River basin are derived from a common ancestor and have a distinct evolutionary trajectory relative to the historical founding populations in Canada and Alaska. Thus, Arctic grayling originating from and currently within the upper Missouri River basin represent the southernmost assemblage of the species, facing similar selection pressures and evolving independent of more northern populations.

The introduced Arctic grayling populations in the upper Missouri River basin occupy, for the most part, high-elevation habitats that are high-quality because of intact riparian areas and a consistent supply of cool water. Given the predicted effects of climate change in the West (see discussion under "Climate Change" in Factor A below), these types of habitats are the same habitats that the Service would explore for long-term conservation of Arctic grayling, if needed, because they may serve as thermal refugia as temperatures rise and provide greater redundancy in case of catastrophic events.

2. *In 2010, the Service concluded there did not appear to be any formally recognized conservation value for the introduced populations of Arctic grayling in the upper*

Missouri River basin because they were not being used in conservation or restoration programs.

Until recently, the genetic structure and source of these introduced populations were unknown. Populations with a high likelihood of being Montana origin were used for conservation purposes (e.g., reintroductions) as a precautionary approach to Arctic grayling conservation. Now that the amount of genetic diversity within and among the introduced Arctic grayling populations and their source(s) are known, it is probable these introduced populations could be used in future conservation actions as source stock, if needed.

3. *In 2010, the Service indicated there were concerns that introduced Arctic grayling populations could pose genetic risks to the native population (i.e., Big Hole Population) as cited in the Montana Fluvial Arctic Grayling Restoration Plan (Montana Fluvial Arctic Grayling Restoration Plan 1995, p. 15). In this Plan, Arctic grayling populations in Agnes, Schwinegar, Odell, Miner and Mussigbrod lakes were identified as potential threats to the genetic integrity of the Big Hole River population because of hydrologic connectivity between these lakes and the Big Hole River and the potential for genetic mixing.*

Recently, genetic analyses have confirmed reproductive isolation among extant Arctic grayling populations in the upper Missouri River basin and within the Big Hole River watershed (Peterson and Ardren 2009, p. 1770; Leary 2014, unpublished data). In addition, multiple historical stockings have occurred in the Big Hole River from other sources within the upper Missouri River basin. Recent genetic analysis found no evidence of a significant genetic contribution from historical stocking on the current genetic structure of Arctic grayling in the Big Hole River (Peterson and Ardren 2009, p. 1768). Thus, we concluded in 2014 and continue to conclude that the concern that populations occupying lakes within the Big Hole River watershed could pose genetic risks to the Big Hole River population appears unfounded.

4. *In 2010, the Service concluded that introduced populations of Arctic grayling in the upper Missouri River basin had no conservation value because these populations apparently had been isolated from their original source stock for decades without any supplementation from the wild and were established without any formal genetic consideration to selecting and mating broodstock.*

It is now apparent from our review of historical stocking records that many of these introduced populations received multiple stockings from the same source or multiple stockings from several different sources over a wide range of years (MFISH 2014a, unpublished data). Additionally, most individual stockings involved a large number of eggs or fry (up to 1 million for some stockings). Cumulatively, this information suggests several points. First, stockings that used a large number of eggs or fry necessitate that gametes from multiple brood fish were used per stocking, given the physical constraints of number of eggs per unit body size of female Arctic grayling. Second, stockings in most of the introduced populations occurred over many years (up to 60 years in some cases). This indicates different cohorts of Arctic grayling had to be used, since the generation time of Arctic grayling is approximately 3.5 years in the upper Missouri River basin (references *in* DeHaan *et al*. 2014, p. 10). Lastly, the new genetic analyses from seven of the introduced Arctic grayling populations indicate moderate to high levels of genetic diversity within the populations. This result could only be obtained from the founding of these populations using large numbers of brood fish and gametes over multiple years. Mutation is unlikely to have accounted for these levels of genetic diversity over a relatively short time period of isolation (Freeman and Herron 2001, p. 143).

For perspective, Montana Fish, Wildlife, and Parks has developed guidelines for the establishment and maintenance of Arctic grayling broodstock. To adequately capture most of the genetic variation in a source population, the crossing of a minimum of 25 male and 25 female Arctic grayling is currently recommended (Leary 1991, p. 2151). It is likely that the historical stockings used to found the introduced Arctic grayling populations in the upper Missouri River basin equaled or exceeded this through stocking large numbers of eggs or fry over multiple years.

5. *In 2010, the Service concluded that the source populations used to found the introduced Arctic grayling populations in the upper Missouri River drainage were not well documented (Peterson and Ardren 2009, p. 1767), so we could not be certain of whether these Arctic grayling were of local origin.*

Since 2010, new genetic information (Leary 2014, unpublished data) and review of historical stocking records (MFISH 2014a, unpublished data) indicate the founding populations used for stocking are from the Upper Missouri River DPS of Arctic grayling, and contain moderate to high levels of genetic diversity.

6. *In 2010, the Service concluded the primary intent of culturing and introducing Arctic grayling populations within the upper Missouri River basin was to provide recreational fishing opportunities in high mountain lakes, and that, therefore, these introduced populations had no conservation value.*

Review of the historical literature indicates Arctic grayling populations were stocked both for recreational fishing and conservation purposes (Brown 1943, pp. 26-27; Nelson 1954, p. 341; Vincent 1962, p. 151). Following the drought in the 1930s, conservation stockings of Arctic grayling were advocated because most rivers and streams were dewatered, prompting fish managers to introduce Arctic grayling into habitats with a more consistent supply of cool water (e.g., high-elevation mountain lakes; Brown 1943, pp. 26-27; Nelson 1954, p. 341; Vincent 1962, p. 151).

In conclusion, introduced populations of Arctic grayling established within the upper Missouri River basin, whether they were originally established for recreational fishing or conservation purposes, captured moderate to high levels of genetic diversity of upper Missouri River basin Arctic grayling. The potential adaptive capabilities represented by this genetic diversity have conservation value, particularly in a changing climate. These populations reside in high-quality habitat, the same habitat the Service would look to for long-term conservation, if needed. Thus, the introduced populations of Arctic grayling within the upper Missouri River basin have conservation value, and, therefore, we include them in our analysis of a potential DPS of Arctic grayling.

Origins, Biogeography, and Genetics of Arctic Grayling in North America

North American Arctic grayling are most likely descended from Eurasian *Thymallus* that crossed the Bering land bridge during or before the Pleistocene glacial period (Stamford and Taylor 2004, pp. 1533, 1546). There were multiple opportunities for freshwater faunal exchange between North America and Asia during the Pleistocene, but genetic divergence between North American and Eurasian Arctic grayling suggests that the species could have colonized North America as early as the mid-late Pliocene (more than 3 million years ago) (Stamford and Taylor 2004, p. 1546). Genetic studies of Arctic grayling using mitochondrial DNA (mtDNA, maternally inherited DNA located in cellular organelles called mitochondria) and microsatellite DNA (repeating sequences of nuclear DNA) have shown that North American Arctic grayling consist of at least three major lineages that originated in distinct Pleistocene glacial refugia (Stamford and Taylor 2004, p. 1533). These three groups include a South Beringia lineage found in western Alaska to northern British Columbia, Canada; a North Beringia lineage found on the North Slope of Alaska, the lower Mackenzie River, and to eastern Saskatchewan; and a Nahanni lineage found in the lower Liard River and the upper Mackenzie River drainage in northeastern British Columbia and southeastern Yukon (Stamford and Taylor 2004, pp. 1533, 1540). Arctic grayling from the upper Missouri River basin were tentatively placed in the North Beringia lineage because a small sample (three individuals) of Montana Arctic grayling shared a mtDNA haplotype (form of the mtDNA) with populations in Saskatchewan and the lower Peace River, British Columbia (Stamford and Taylor 2004, p. 1538).

The existing mtDNA data suggest that Missouri River Arctic grayling share a common ancestry with the North Beringia lineage, but other genetic markers (e.g., allozymes, microsatellites) and biogeographic history indicate that Missouri River Arctic grayling have been physically and reproductively isolated from northern populations for millennia. Pre-glacial colonization of the Missouri River basin by Arctic grayling was possible because the river flowed to the north and drained into the Arctic-Hudson Bay prior to the last glacial cycle (Cross *et al.* 1986, pp. 374–375; Pielou 1991, pp. 194–195).

Low mtDNA diversity observed in a small number of Montana Arctic grayling samples and a shared ancestry with Arctic grayling from the North Beringia lineage suggest a more recent, post-glacial colonization of the upper Missouri River basin. In contrast, microsatellite DNA show substantial divergence between Montana and Saskatchewan (i.e., same putative mtDNA lineage) (Peterson and Ardren 2009, entire). Differences in the frequency and size distribution of microsatellite alleles between Montana populations and two Saskatchewan populations indicate that Montana Arctic grayling have been isolated long enough for mutations (i.e., evolution) to be responsible for the observed genetic differences.

Additional comparison of 21 Arctic grayling populations from Alaska, Canada, and the Missouri River basin using 9 of the same microsatellite loci as Peterson and Ardren (2009, entire) further supports the distinction of Missouri River Arctic grayling relative to populations elsewhere in North America (USFWS 2010, unpublished data). Analyses of these data using two different methods clearly separates sample fish from 21 populations into two clusters: one cluster representing populations from the upper Missouri River basin, and another cluster representing populations from Canada and Alaska (USFWS 2010, unpublished data). This data, support the interpretation that the previous analyses of Stamford and Taylor (2004, entire) underestimated the distinctiveness of Missouri River Arctic grayling relative to other sample populations, likely because of the combined effect of small sample sizes and the lack of variation observed in the Missouri River for the markers used in that study (Stamford and Taylor 2004, pp. 1537–1538). Thus, these recent microsatellite DNA data suggest that Arctic grayling may have colonized the Missouri River before the onset of Wisconsin glaciation (more than 80,000 years ago).

Genetic relationships among native and introduced populations of Arctic grayling in Montana have recently been investigated (Peterson and Ardren 2009, entire). Introduced populations of Arctic grayling occupying lakes trace some of their original ancestry to the Centennial Valley (Peterson and Ardren 2009, p. 1767), and stocking of hatchery Arctic grayling did not have a large effect on the genetic composition of the extant native populations (Peterson and Ardren 2009, p. 1768). Differences between native populations of Arctic grayling across the life history spectrum (i.e., formerly referred to as adfluvial and fluvial) are not as large as differences resulting from geography (i.e., drainage of origin). For a full discussion on Arctic grayling life history, see the section Life History Diversity in Arctic Grayling in the Upper Missouri River Basin in this status review on p. 23).

Habitat

Arctic grayling occupy a variety of habitats including small streams, large rivers, lakes, and bogs (Northcote 1995, pp. 152–153; Scott and Crossman 1998, p. 303). They may even enter brackish water (less than or equal to 4 parts per thousand salt content) when migrating between adjacent river systems (West *et al.* 1992, pp. 713–714). Native populations are found at elevations ranging from near sea level, such as in Bristol Bay, Alaska, to high-elevation montane valleys (more than 1,830 meters (m) or 6,000 feet (ft)), such as the Big Hole River and Centennial Valley in southwestern Montana.

Arctic grayling have defined thermal tolerances. We reported in our 2014 Finding that water temperatures become unsuitable for Arctic grayling above 20 ºC (68 ºF) (Hubert *et al.* 1985, p. 24). However, this value was derived from a Habitat Suitability Index model not intended to predict actual presence or absence of Arctic grayling in relation to water temperature (Hubert et al. 1985, p. 13). Further the model reliability was expected to vary among different geographical areas (Hubert et al. 1985, pp. iii, 7) and there was no field testing of modelled relationships (Hubert et al. 1985, p. 7). Other studies, including some more recent studies investigating the thermal tolerances of Arctic grayling in laboratory and field conditions (McCullough 2017, pp. 16-17; Vatland 2015, entire; Feldmeth and Eriksen 1978, p. 2041; Lohr et al. 1996, pp. 935, 937) clearly show adult Arctic grayling can indeed survive at water temperatures above 20°C (Feldmeth and Eriksen 1978, p. 2041; Lohr et al. 1996, pp. 934, 937; Chadwick et al. 2015, pp. 5-8; references *in* Vatland 2015, pp. 33-34;), including two studies used in the development of the Habitat Suitability Index models (see LaPerrier and Carlson 1973, pp. 29-30; Feldmeth and Eriksen 1978, p. 2041 *in* Hubert et al. 1985, pp. 5, 12). Currently, the water temperature threshold of 21°C (70°F) is believed to be the temperature at which salmonids (including Arctic grayling) *begin* to experience physiological stress (Chadwick et al. 2015, pp. 5-8; references *in* Vatland 2015, pp. 33-34). Water temperatures which can cause instantaneous death in Arctic grayling vary by testing methodology, acclimation temperature and grayling life stage (among other factors) but have been reported as high as 29.3°C (84.7°F) for juvenile grayling (Lohr et al. 1996, pp. 935, 937). For an in-depth discussion on water temperature thresholds for

Arctic grayling, see Dewatering From Irrigation and Increased Water Temperatures (later in this review).

Channel gradient is another important habitat variable influencing local distributions of Arctic grayling. Arctic grayling are typically found in low-to-moderate gradient (less than 4 percent) streams and rivers with low-to-moderate water velocities (less than 2 feet/sec (60 centimeters/sec)) and are not generally found in swift, high-gradient streams (Vincent 1962, p. 36–37, 41–43). Within streams and rivers, juvenile and adult Arctic grayling spend much of their time in pool habitat (Kaya 1990 and references therein, p. 20; Lamothe and Magee 2003, pp. 13–14). In general, Arctic grayling appear to be able to occupy varying sizes of ponds/lakes, as long as water temperatures are within their defined limits and there is an inlet or outlet stream for spawning.

Breeding

Arctic grayling typically spawn in the spring or early summer, depending on latitude and elevation (Northcote 1995, p. 149). In Montana, Arctic grayling generally spawn from late April to mid-May by depositing adhesive eggs over gravel substrate without excavating a nest (Kaya 1990, p. 13; Northcote 1995, p. 151). In general, the reproductive ecology of Arctic grayling differs from other salmonid species (trout and salmon) in that Arctic grayling eggs tend to be comparatively small; thus, they have higher relative fecundity (females have more eggs per unit body size). Males establish and defend spawning territories rather than defending access to females (Northcote 1995, pp. 146, 150–151). The time required for development of eggs from embryo until they emerge from stream gravel and become swim-up fry depends on water temperature (Northcote 1995, p. 151). In the upper Missouri River basin, development from embryo to fry averages about 3 weeks (Kaya 1990, pp. 16–17). Small, weakly swimming fry (typically 1–1.5 centimeters (cm) (0.4–0.6 in.) at emergence prefer low-velocity stream habitats (Armstrong 1986, p. 6; Kaya 1990, pp. 23–24; Northcote 1995, p. 151).

Diet

Arctic grayling of all ages feed primarily on aquatic and terrestrial invertebrates captured on or near the water surface, but also will feed opportunistically on fish and fish eggs (Northcote 1995, pp. 153–154; Behnke 2002, p. 328). Feeding locations for individual fish are typically established and maintained through size-mediated dominance hierarchies where larger individuals defend favorable feeding positions (Hughes 1992, p. 1996).

General Life History Diversity

Migratory behavior is a common life-history trait in salmonid fishes such as Arctic grayling (Armstrong 1986, pp. 7-8; Northcote 1995, pp. 156–158; 1997, pp. 1029, 1031–1032, 1034). In general, migratory behavior in Arctic grayling and other salmonids results in cyclic patterns of movement between refuge, rearing-feeding, and spawning habitats (Northcote 1997, p. 1029).

Arctic grayling may move to refuge habitat as part of a regular seasonal migration (e.g., in winter), or in response to episodic environmental stressors (e.g., high summer water temperatures). In Alaska, Arctic grayling in rivers typically migrate downstream in the fall, moving into larger streams or mainstem rivers that do not completely freeze (Armstrong 1986, p. 7). In Arctic rivers, fish often seek overwintering habitat influenced by groundwater (Armstrong 1986, p. 7). In some drainages, individual fish may migrate considerable distances (greater than 150 km or 90 mi) to overwintering habitats (Armstrong 1986, p. 7). In the Big Hole River, Montana, similar downstream and long-distance movement to overwintering habitat has been observed in Arctic grayling (Shepard and Oswald 1989, pp. 18–21, 27). In addition, Arctic grayling in the Big Hole River move downstream in proximity to colder tributary streams in summer when thermal conditions in the mainstem river become stressful (Lamothe and Magee 2003, p. 17).

In spring, mature Arctic grayling leave overwintering areas and migrate to suitable spawning sites. In river systems, this typically involves an upstream migration to tributary streams or shallow riffles within the mainstem (Armstrong 1986, p. 8; Shepard and Oswald 1989; p. 18). Arctic grayling in lakes typically migrate to either the inlet or outlet to spawn (Armstrong 1986, p. 8; Kaya 1989, p. 474; Northcote 1995 p. 148). In some situations, Arctic grayling exhibit natal homing, whereby individuals spawn in or near the location where they were born (Northcote 1995 pp. 157–160; Boltz and Kaeding 2002, p. 22); however, it is unclear what factors may be influencing the extent of this phenomenon.

Fry from river-dwelling populations typically seek feeding and rearing habitats in the vicinity of where they were spawned (Armstrong 1986, pp. 6–7; Kaya and Jeanes 1995, p. 455; Northcote 1995, p. 156), while those from lake populations migrate downstream (inlet spawners) or upstream (outlet spawners) to the adjacent lake.

Following spawning, adults move to appropriate feeding areas if they are not adjacent to spawning habitat (Armstrong 1986, pp. 7–8; Shepard and Oswald 1989; p. 18). Juvenile Arctic grayling may undertake seasonal migrations between feeding and overwintering habitats until they reach maturity and add the spawning migration to this cycle (Northcote 1995, pp. 156–157).

Life History Diversity in Arctic Grayling in the Upper Missouri River Basin

Arctic grayling in the Upper Missouri River basin exhibit a range of life histories. Some Arctic grayling spend their entire lives in flowing water (often referred to as fluvial), some primarily reside in lakes and only use flowing water for spawning (often referred to as adfluvial) and others appear to use some combination of both strategies. In previous findings, we characterized populations as being primarily fluvial or adfluvial; however, we now note that there appears to be a spectrum of behaviors that Arctic grayling are using, rather than the two distinct strategies that were formerly classified as fluvial and adfluvial (Arctic Grayling Workgroup 1995; p. 1; Cayer 2014a, pers. comm.; MFISH 2014b, unpublished data; Gander et al. 2019, pp. 16, 19-21). For example, Arctic grayling from the Centennial Valley (Long Creek and Red Rock Creek) and Ennis Reservoir/Madison River (mainstem Madison River) have been documented in riverine habitats well past the spawning period through autumn. These occurrences do not appear to be linked to individual Arctic grayling seeking thermal refugia during summer (Arctic Grayling Workgroup 1995; p. 1; Cayer 2014a, pers. comm.; MFISH 2014b, unpublished data; Gander et al. 2019, pp. 16, 19-21). These occurrences include multiple age classes (Age-1 to Age-3) of Arctic grayling in both Long Creek and the Madison River and are located in stream reaches that are considerable distances (up to 15 miles in the Madison River) from lake habitats (Cayer 2014a, pers. comm.; MFISH 2014b, unpublished data; Gander et al. 2019, pp. 16, 19-21; MFWP 2019, unpublished data). The presence of life history strategies from across the behavioral spectrum is consistent with how other salmonid populations are structured, where populations utilize different strategies, depending on habitat conditions and other environmental factors, to maximize their chances of survival and ultimately reproduction (Willson 1997, entire; Thorpe et al. 1998, pp. 581-586; Riemann and Dunham 2000, pp. 53-60; Koskinen 2002, pp. 826-829). In this status review, we still use the terms "fluvial" or "adfluvial" in some instances; however, these instances are typically where historical document titles explicitly use these terms and we are citing or referring to these specific documents or the populations they referred to historically.

The different life-history forms of Arctic grayling in the upper Missouri River do not appear to represent distinct evolutionary lineages. Instead, they appear to represent an example of adaptive radiation (Schluter 2000, p. 1), whereby the forms differentiated from a common ancestor and developed traits that allowed them to exploit different habitats. The primary evidence for this conclusion is genetic data that indicate that within the Missouri River basin the different forms are more closely related to each other than they are to the same form elsewhere in North America (Redenbach and Taylor 1999, pp. 27–28; Stamford and Taylor 2004, p. 1538; Peterson and Ardren 2009, p. 1766). Historically, most Arctic grayling in the Missouri River basin above the Great Falls resided in flowing water, perhaps because there were only a few lakes accessible to natural colonization of Arctic grayling that would permit variability in life history expression (Kaya 1992, p. 47).

There are differences in genetic characteristics among the different forms of Arctic grayling, but also some plasticity in behavior where individuals from a population can exhibit a range of behaviors. For example, Arctic grayling fry in Montana can exhibit heritable, genetically-based differences in swimming behavior between life forms (Kaya 1991, pp. 53, 56–58; Kaya and Jeanes 1995, pp. 454, 456). Progeny of Arctic grayling from riverine habitats exhibited a greater tendency to hold their position in flowing water relative to progeny from lake-reared populations (Kaya 1991, pp. 53, 56–58; Kaya and Jeanes 1995, pp. 454, 456). Similarly, young Arctic grayling from inlet and outlet spawning lake-reared forms exhibited an innate tendency to move downstream and upstream, respectively (Kaya 1989, pp. 478–480). All three studies (Kaya 1989, entire; 1991, entire; Kaya and Jeanes 1995, entire) demonstrate that the response of fry to flowing water depended strongly on the life-history form of the source population, and that this behavior has a genetic basis. However, behavioral responses also were mediated by environmental conditions (light—Kaya 1991, pp. 56–57; light and water temperature—Kaya 1989, pp. 477–479), and some progeny exhibited a range of behavioral characteristics. For example, some progeny of Arctic grayling from riverine habitats moved downstream rather than holding position, and some individuals from an inlet-spawning, lake-dwelling form held position or moved upstream (Kaya 1991, p. 58). These observations indicate that some plasticity across a range of behaviors exists, at least for very young Arctic grayling, despite the predominate life history form of adult fish producing the progeny.

The ability of different life forms of Arctic grayling to give rise to other populations is unclear. While most extant Arctic grayling populations occupying lakes in the Upper Missouri River originated from sources from riverine habitats (see Table 2; Kaya 1992, p. 53; Jeanes 1996, pp. 54), the ability of individuals from lakes to give rise to a functional population in flowing water is less certain. Circumstantial support for reduced plasticity in Arctic grayling occupying lakes comes from observations that these fish stocked in river habitats almost never establish populations (Kaya 1990, pp. 31–34). However, these observations may be confounded by stocking method, because most historical attempts to establish Arctic grayling in riverine environments included stocking of juvenile or adult Arctic grayling, which is not effective even when using fish from riverine sources. For example, Arctic grayling from the Big Hole River brood reserve were stocked into the Ruby River early in the reintroduction process and did not result in an established population, yet use of remote site incubators (a different stocking method) with Big Hole River grayling eggs later in the Ruby River reintroduction process was effective at establishing the population (Gander et al. 2019, pp. 2, 4). We note that Arctic grayling occupying lakes retain some life-history flexibility—at least in lake environments—as naturalized populations derived from inlet-spawning stocks have established outlet-spawning demes (a deme is a local population that shares a distinct gene pool) in Montana and in Yellowstone National Park (Kruse 1959, p. 318; Kaya 1989, p. 480). In addition, a small percentage of young Arctic grayling occupying lakes exposed to flow exhibited characteristics of fish reared in flowing water (e.g., station-holding or upstream movement) in a laboratory experiment (Kaya 1991, p. 56). These results indicate some plasticity exists in Arctic grayling occupying lakes, but the frequent failure of introductions of Arctic grayling occupying lakes into flowing water habitats suggest preservation of the breadth of the known Arctic grayling life history spectrum is warranted.

Age and Growth

Age at maturity and longevity in Arctic grayling varies regionally and is probably related to growth rate, with populations in colder, northern latitudes maturing at later ages and having a greater lifespan (Kruse 1959, pp. 340–341; Northcote 1995 and references therein, pp. 155–157). Arctic grayling in the upper Missouri River typically mature at age 2 (males) or age 3 (females), and individuals greater than age 6 are rare (Kaya 1990, p. 18; Magee and Lamothe 2003, pp. 16–17). The majority of the Arctic grayling spawning in two tributaries in the Centennial Valley, Montana, were age 3, and the oldest individuals aged from a larger sample were age 6 (Nelson 1954, pp. 333–334). Arctic grayling spawning in Red Rock Creek were mostly ages 2 to 5, but some individuals were age 7 (Mogen 1996, pp. 32–34).

Generally, growth rates of Arctic grayling are greatest during the first years of life then slow dramatically after maturity. Within that general pattern, there is substantial variation among populations from different regions. Arctic grayling populations in Montana (Big Hole River and Red Rock Lakes) have very high growth rates relative to those from British Columbia, Asia, and the interior and North Slope of Alaska (Carl *et al.* 1992, p. 240; Northcote 1995, pp. 155–157; Neyme 2005, p. 28).

Distinct Population Segment

Pursuant to the Act, we must consider for listing any species, subspecies, or, for vertebrates, any distinct population segment (DPS) of these taxa, if there is sufficient information to indicate that such action may be warranted. To interpret and implement the DPS provision of the Act and Congressional guidance, the Service and the National Marine Fisheries Service published, on February 7, 1996, an interagency Policy Regarding the Recognition of Distinct Vertebrate Population Segments under the Act (61 FR 4722; February 7, 1996). This policy addresses the recognition of DPSs for potential listing actions. The policy allows for more refined application of the Act that better reflects the biological needs of the taxon being considered, and avoids the inclusion of entities that do not require its protective measures.

Under our DPS policy, three elements are considered in a decision regarding the status of a possible DPS as endangered or threatened under the Act. These are applied similarly for additions to the list of endangered and threatened species, reclassification, and removal from the list. They are: (1) Discreteness of the population segment in relation to the remainder of the taxon; (2) the biological or ecological significance of the population segment to the taxon to which it belongs; and (3) the population segment's conservation status in relation to the Act's standards for listing (i.e., whether the population segment is, when treated as if it were a species or subspecies, an endangered or threatened species). Discreteness refers to the degree of isolation of a population from other members of the species, and we evaluate this factor based on specific criteria. If a population segment is considered discrete, we must consider whether the discrete segment is "significant" to the taxon to which it belongs by using the best available scientific and commercial information. When determining if a potential DPS is significant, our policy directs us to sparingly list DPS's while encouraging the conservation of genetic diversity. If we determine that a population segment is both discrete and significant, we then evaluate it for endangered or threatened species status based on the Act's standards.

Below we provide our updated evaluation of discreteness and significance under the DPS policy of the segment of the arctic grayling occurring in the upper Missouri River basin.

Distinct Population Segment Analysis for Arctic Grayling in the Upper Missouri River Basin.

Analysis of Discreteness

Under our DPS Policy, a population segment of a vertebrate species may be considered discrete if it satisfies either one of the following conditions: (1) It is markedly separated from other populations of the same taxon as a consequence of physical, physiological, ecological, or behavioral factors (quantitative measures of genetic or morphological discontinuity may provide evidence of this separation); or (2) it is delimited by international governmental boundaries within which differences in control of exploitation, management of habitat, conservation status, or regulatory mechanisms exist that are significant in light of section 4(a)(1)(D) of the Act (inadequacy of existing regulatory mechanisms).

Arctic grayling native to the upper Missouri River are isolated from all other populations of the species, which inhabit the Arctic Ocean, Hudson Bay, and north

Pacific Ocean drainages in Asia and North America. Arctic grayling native to the upper Missouri River occur as a disjunct group of populations approximately 800 km (500 mi) to the south of the next-nearest Arctic grayling population in central Alberta, Canada. Missouri River Arctic grayling have been isolated from other populations for at least 10,000 years based on historical reconstruction of river flows at or near the end of the Pleistocene (Cross *et al.* 1986, p. 375; Pileou 1991, pp. 10–11). Genetic data confirm Arctic grayling in the Missouri River basin have been reproductively isolated from populations to the north for millennia (Everett 1986, pp. 79–80; Redenbach and Taylor 1999, p. 23; Stamford and Taylor 2004, p. 1538; Peterson and Ardren 2009, pp. 1764–1766; USFWS, unpublished data). Consequently, we conclude that Arctic grayling native to the upper Missouri River are markedly separated from other native populations of the taxon as a result of physical factors (isolation), and therefore meet the first criterion of discreteness under the DPS policy. As a result, Arctic grayling native to the upper Missouri River are considered a discrete population according to the DPS policy. Because the entity meets the first criterion (markedly separated), an evaluation with respect to the second criterion (international boundaries) is not needed.

Analysis of Significance

If a population segment is considered discrete under one or more of the conditions described in the Service's DPS policy, its biological and ecological significance will be considered in light of Congressional guidance that the authority to list DPSs be used "sparingly" while encouraging the conservation of genetic diversity. In making this determination, we consider available scientific evidence of the discrete population segment's importance to the taxon to which it belongs. Since precise circumstances are likely to vary considerably from case to case, the DPS policy does not describe all the classes of information that might be used in determining the biological and ecological importance of a discrete population. However, the DPS policy describes four possible classes of information that provide evidence of a population segment's biological and ecological importance to the taxon to which it belongs. As specified in the DPS policy (61 FR 4722), this consideration of the population segment's significance may include, but is not limited to, the following:

(1) Persistence of the discrete population segment in an ecological setting unusual or unique to the taxon;

(2) Evidence that loss of the discrete population segment would result in a significant gap in the range of a taxon;

(3) Evidence that the discrete population segment represents the only surviving natural occurrence of a taxon that may be more abundant elsewhere as an introduced population outside its historical range; or

(4) Evidence that the discrete population segment differs markedly from other populations of the species in its genetic characteristics.

A population segment needs to satisfy only one of these conditions to be considered significant. Furthermore, other information may be used as appropriate to provide evidence for significance.

Unique Ecological Setting

Water temperature is a key factor influencing the ecology and physiology of ectothermic (body temperature regulated by ambient environmental conditions) salmonid fishes, and can dictate reproductive timing, growth and development, and life-history strategies. Groundwater temperatures can be related to air temperatures (Meisner 1990, p. 282), and thus reflect the regional climatic conditions. Warmer groundwater influences ecological factors such as food availability, the efficiency with which food is converted into energy for growth and reproduction, and ultimately growth rates of aquatic organisms (Allan 1995, pp. 73–79). Aquifer structure and groundwater temperature is important to salmonid fishes because groundwater can strongly influence stream temperature, and consequently egg incubation and fry growth rates, which are strongly temperature-dependent (Coutant 1999, pp. 32–52; Quinn 2005, pp. 143–150).

Missouri River Arctic grayling occur within the 4 to 7 ºC (39 to 45 ºF) ground water isotherm (see Heath 1983, p. 71; an isotherm is a line connecting bands of similar temperatures on the earth's surface), whereas most other North American Arctic grayling are found in isotherms less than 4 ºC, and much of the species' range is found in areas with discontinuous or continuous permafrost (Meisner *et al.* 1988, p. 5; Table 2). Much of the historical range of Arctic grayling in the upper Missouri River is encompassed by mean annual air temperature isotherms of 5 to 10 ºC (41 to 50 ºF) (USGS 2009), with the colder areas being in the headwaters of the Madison River in Yellowstone National Park. In contrast, Arctic grayling in Canada, Alaska, and Asia are located in regions encompassed by air temperature isotherms 5 ºC and colder (41 ºF and colder), with much of the species distributed within the 0 to -10 ºC isolines (32 to 14 ºF). This difference is significant because Arctic grayling in the Missouri River basin have evolved in isolation for millennia in a generally warmer climate than other populations. The potential for thermal adaptations makes Missouri River Arctic grayling a significant biological resource for the species under expected climate change scenarios.

Table 2. Differences between the ecological setting of the Upper Missouri River and elsewhere in the species' range of Arctic grayling.

Ecological Setting Variable	Missouri River	Rest of Taxon
Bailey's Ecoregion	Dry Domain: Temperate Steppe	Polar Domain: Tundra & Subarctic Humid Temperate: Marine, Prairie, Warm Continental Mountains
Air temperature (isotherm)	5 to 10 °C (41 to 50 °F)	-15 to 5 °C (5 to 41 °F)
Groundwater temperature (isotherm)	4 to 7 °C (39 to 45 °F)	Less than 4 °C (Less than 39 °F)

Arctic grayling in the upper Missouri River basin occur in a temperate ecoregion distinct from all other Arctic grayling populations worldwide, which occur in Arctic or sub-Arctic ecoregions dominated by Arctic flora and fauna. An ecoregion is a continuous geographic area within which there are associations of interacting biotic and abiotic features (Bailey 2005, pp. S14, S23). These ecoregions delimit large areas within which local ecosystems recur more or less in a predictable fashion on similar sites (Bailey 2005, p. S14). Ecoregional classification is hierarchical, and based on the study of spatial coincidences, patterning, and relationships of climate, vegetation, soil, and landform (Bailey 2005, p. S23). The largest ecoregion categories are domains, which represent subcontinental areas of similar climate (e.g., polar, humid temperate, dry, and humid tropical) (Bailey 1994; 2005, p. S17). Domains are divided into divisions that contain areas of similar vegetation and regional climates. Arctic grayling in the upper Missouri River basin are the only example of the species naturally occurring in a dry domain (temperate steppe division; Table 2). The vast majority of the species' range is found in the polar domain (all of Asia, most of North America), with small portions of the range occurring in the humid temperate domain (northern British Columbia and southeast Alaska). Occupancy of Missouri River Arctic grayling in a temperate ecoregion is significant for two primary reasons. First, an ecoregion represents a suite of factors (climate, vegetation, landform) influencing, or potentially influencing, the evolution of species within that ecoregion. Since Missouri River Arctic grayling have existed for thousands of years in an ecoregion quite different from the majority of the taxon, they have likely developed adaptations during these evolutionary timescales that distinguish them from the rest of the taxon, even if we have yet to conduct the proper studies to measure these adaptations. Second, the occurrence of Missouri River Arctic grayling in a unique ecoregion helps reduce the risk of species-level extinction, as the different regions may respond differently to environmental change.

Arctic grayling in the upper Missouri River basin have existed for at least 10,000 years in an ecological setting quite different from that experienced by Arctic grayling elsewhere in the species' range. The most salient aspects of this different setting relate to temperature and climate, which can strongly and directly influence the biology of ectothermic species (like Arctic grayling). Arctic grayling in the upper Missouri River have experienced warmer temperatures than most other populations. Physiological and life-history adaptation to local temperature regimes are regularly documented in salmonid fishes (Taylor 1991, pp. 191–193), but experimental evidence for adaptations to temperature, such as unusually high temperature tolerance or lower tolerance to colder temperatures, is lacking for Missouri River Arctic grayling because the appropriate studies have not been conducted. Lohr *et al.* (1996, p. 934) studied the upper thermal tolerances of Arctic grayling from the Big Hole River, but their research design did not include other populations from different thermal regimes, so it was not possible to make between-population contrasts under a common set of conditions. Arctic grayling from the upper Missouri River demonstrate very high growth rates relative to other populations (Northcote 1995, p. 157). Experimental evidence obtained by growing fish from populations under similar conditions would be needed to measure the relative influence of genetics (local adaptation) versus environment.

We conclude that the occurrence of Arctic grayling in the upper Missouri River is biogeographically important to the species, that grayling there have occupied a warmer and more temperate setting that is distinctly different from the ecological settings relative to the rest of the species (see Table 2, above), and that they have been on a different evolutionary trajectory for at least 10,000 years. We conclude that these differences are significant because they may provide the species with additional evolutionary resiliency in the future in light of the changing climate. Consequently, we believe that Arctic grayling in the upper Missouri River occupy a unique ecological setting for the species.

Gap in the Range

Arctic grayling in Montana (southern extent is approximately 44º36’23”N latitude) represent the southern-most extant population of the species’ distribution since the Pleistocene glaciation. The next-closest native Arctic grayling population outside the

Missouri River basin is found in the Pembina River (approximately 52°55'6.77"N latitude) in central Alberta, Canada, west of Edmonton (Blackburn and Johnson 2004, pp. ii, 17; ASRD 2005, p. 6). The Pembina River drains into Hudson Bay and is thus disconnected from the Missouri River basin. Loss of the native Arctic grayling of the upper Missouri River would shift the southern distribution of Arctic grayling by more than 8º latitude (about 500 miles). Such a dramatic range constriction would constitute a significant geographic gap in the species' range and would eliminate a genetically distinct group of Arctic grayling, which may limit the species' ability to cope with future environmental change.

Marginal populations, defined as those on the periphery of the species' range, are believed to have high conservation significance (Mitikka *et al*. 2008; Gibson *et al*. 2009, entire; Haak *et al*. 2010, entire; Osborne *et al*. 2012). Peripheral populations may occur in suboptimal habitats and thus be subjected to very strong selective pressures (Fraser 2000, p. 50). Consequently, individuals from these populations may contain adaptations that may be important to the taxon in the future. Lomolino and Channell (1998, p. 482) hypothesize that because peripheral populations should be adapted to a greater variety of environmental conditions, then they may be better suited to deal with anthropogenic (human-caused) disturbances than populations in the central part of a species' range. Arctic grayling in the upper Missouri River have, for millennia, existed in a climate warmer than that experienced by the rest of the taxon. If this selective pressure has resulted in adaptations to cope with increased water temperatures, then the population segment may contain genetic resources important to the taxon. For example, if northern populations of Arctic grayling are less suited to cope with increased water temperatures expected under climate warming, then Missouri River Arctic grayling might represent an important population for reintroduction in those northern regions. We believe that Arctic grayling's occurrence at the southernmost extreme of the range in the upper Missouri River contributes to the resilience of the overall taxon because these peripheral populations may possess increased adaptability relative to the rest of the taxon.

Only Surviving Natural Occurrence of the Taxon that May Be More Abundant Elsewhere as an Introduced Population Outside of Its Historical Range

This criterion does not directly apply to the Arctic grayling in the upper Missouri River because it is not the only surviving natural occurrence of the taxon; there are native Arctic grayling populations in Canada, Alaska, and Asia.

Differs Markedly in Its Genetic Characteristics

Differences in genetic characteristics can be measured at the molecular, genetic, or phenotypic level. Three different types of molecular markers (allozymes, mtDNA, and microsatellites) demonstrate that Arctic grayling from the upper Missouri River are genetically different from those in Canada, Alaska, and Asia (Everett 1986, pp. 79-80; Redenbach and Taylor 1999, p. 23; Stamford and Taylor 2004, p. 1538; Peterson and Ardren 2009, pp. 1764–1766; USFWS, unpublished data). These data confirm the reproductive isolation among populations that establishes the discreteness of Missouri River Arctic grayling under the DPS policy. Here, we speak to whether these data also establish significance.

Allozymes

Using allozyme data, Everett (1986, entire) found marked genetic differences among Arctic grayling collected from the Chena River in Alaska; those descended from fish native to the Athabasca River drainage in the Northwest Territories, Canada; and native upper Missouri River drainage populations or populations descended from them (see Leary 2005, pp. 1–2). The Canadian population had a high frequency of two unique alleles (forms of a gene), which strongly differentiated them from all the other samples (Everett 1986, p. 44). With the exception of one introduced population in an irrigation canal (Sunnyslope canal) in Montana that is believed to have experienced extreme genetic bottlenecks, the Chena River (Alaskan) fish were highly divergent from all the other samples as they possessed an unusually low frequency of a specific allele (Everett 1986, p. 60; Leary 2005, p. 1), and contained a unique variant of another allele (Leary 2005, p. 1). Overall, each of the four native Missouri River populations examined (Big Hole, Miner, Mussigbrod, and Centennial Valley) exhibited statistically significant differences in allele frequencies relative to both the Chena River (Alaska) and Athabasca River (Canada) populations (Everett 1986, pp. 15, 67).

Combining the data of Everett (1986, entire), Hop and Gharrett (1989, entire), and Leary (1990, entire) provides information from 21 allozyme loci (genes) from five native upper Missouri River drainage populations, five native populations in the Yukon River drainage in Alaska, and the one population descended from the Athabasca River drainage in Canada (Leary 2005, pp. 1–2). Examination of the genetic variation in these samples indicated that most of the genetic divergence is due to differences among drainages (29 percent) and comparatively little (5 percent) results from differences among populations within a drainage (Leary 2005, p. 1).

Mitochondrial DNA

Analysis using mtDNA indicates that Arctic grayling in North America represent at least three evolutionary lineages that are associated with distinct glacial refugia (Redenbach and Taylor 1999, entire; Stamford and Taylor 2004, entire). Arctic grayling in the upper Missouri River basin belong to the so-called North Beringia lineage (Redenbach and Taylor 1999, pp. 27–28; Samford and Taylor 2004, pp. 1538-1540) because they possess a form of mtDNA that was generally absent from populations collected from other locations within the species' range in North America (Redenbach and Taylor 1999, pp. 27-28; Stamford and Taylor 2004, p. 1538). The notable exceptions were that some fish from the lower Peace River drainage in British Columbia, Canada, and all sampled individuals from the Saskatchewan River drainage Saskatchewan, Canada, also possessed this form of mtDNA (Stamford and Taylor 2004, p. 1538).

A form of mtDNA common in upper Missouri River Arctic grayling, which occurs at lower frequencies in other populations, indicates that Arctic grayling native to the upper Missouri River drainage probably originated from a glacial refuge in the drainage and subsequently migrated northwards when the Missouri River temporarily flowed into the Saskatchewan River and was linked to an Arctic drainage (Cross *et al.* 1986, pp. 374–375; Pielou 1991, p. 195). When the Missouri River began to flow southwards because of the advance of the Laurentide ice sheet (Cross *et al.* 1986, p. 375; Pileou 1991, p. 10), the Arctic grayling in the drainage became physically and reproductively isolated from the rest of the species' range (Leary 2005, p. 2; Campton 2004, p. 6), which would have included those populations in Saskatchewan. Alternatively, the Missouri River Arctic grayling could have potentially colonized Saskatchewan or the Lower Peace River (in British Columbia) or both post-glacially (Stamford 2001, p. 49) via a gap in the Cordilleran and Laurentide ice sheets (Pielou 1991, pp. 10-11), which also might explain the low frequency "Missouri River" mtDNA in Arctic grayling in the Lower Peace River and Upper Yukon River.

We do not interpret the observation that Arctic grayling in Montana and Saskatchewan, and to lesser extent those from the Lower Peace and Upper Yukon River systems, share a mtDNA haplotype to mean that these groups of fish are genetically identical. Rather, we interpret it to mean that these fish shared a common ancestor tens to hundreds of thousands of years ago.

Microsatellite DNA

Recent analysis of microsatellite DNA (highly variable portions of nuclear DNA) showed substantial divergence between Arctic grayling in Missouri River and Saskatchewan populations (Peterson and Ardren 2009, entire). This divergence between populations was measured in terms of allele frequencies, using a metric called F_{st} (Allendorf and Luikart 2007, pp. 52–54, 198–199). An analogous metric, named R_{st}, also measures genetic differentiation between populations based on microsatellite DNA, but differs from F_{st} in that it also considers the size differences between alleles (Hardy *et al.* 2003, p. 1468). An F_{st} or R_{st} of 0 indicates that populations are the same genetically, whereas a value of 1 indicates the populations share no genetic material at the markers being surveyed. F_{st} values range from 0.13 to 0.31 (average 0.18) between Missouri River and Saskatchewan populations (Peterson and Ardren 2009, pp. 1758, 1764-1765), whereas R_{st} values range from 0.47 to 0.71 (average 0.54) for the same comparisons (Peterson and Ardren 2009, pp. 1758, 1764-1765). These values indicate that the two populations differ significantly in allele frequency and also in the size of those alleles. This outcome indicates that the observed genetic differences are due to mutational differences, which suggests the groups may have been separated for millennia (Peterson and Ardren 2009, pp. 1767-1768).

Analysis of Arctic grayling populations from Alaska, Canada, and the Missouri River basin using nine of the same microsatellite loci as Peterson and Ardren (2009, entire) further supports the distinction of Missouri River Arctic grayling relative to populations elsewhere in North America (USFWS, unpublished data). This analysis clearly separated sample fish from 21 populations into two clusters: one cluster representing populations from the upper Missouri River basin, and another cluster representing populations from across Canada and Alaska (USFWS, unpublished data). Divergence in size among these alleles further supports the distinction between Missouri River Arctic grayling and those in Canada and Alaska (USFWS, unpublished data). The interpretation of these data is that the Missouri River populations and the Canada/Alaska populations are highly genetically distinct at the microsatellite loci considered.

Phenotypic Characteristics Influenced by Genetics—Meristics

Phenotypic variation can be evaluated by counts of body parts (i.e., meristic counts of the number of gill rakers, fin rays, and vertebrae characteristics of a population) that can vary within and among species. These meristic traits are influenced by both genetics and the environment (Allendorf and Luikart 2007, pp. 258–259). When the traits are controlled primarily by genetic factors, then meristic characteristics can indicate significant genetic differences among groups. Arctic grayling north of the Brooks Range in Alaska and in northern Canada had lower lateral line scale counts than those in southern Alaska and Canada (McCart and Pepper 1971, entire). These two scale-size phenotypes are thought to correspond to fish from the North and South Beringia glacial refuges, respectively (Stamford and Taylor 2004, p. 1545). Arctic grayling from the Centennial Valley had a phenotype intermediate to the large- and small-scale types (McCart and Pepper 1971, pp. 749, 754). Arctic grayling populations from the Missouri River (and one each from Canada and Alaska) could be correctly assigned to their group 60 percent of the time using a suite of seven meristic traits (Everett 1986, pp. 32–35). Those native Missouri River populations that had high genetic similarity also tended to have similar meristic characteristics (Everett 1986, pp. 80, 83).

Arctic grayling from the Big Hole River showed marked differences in meristic characteristics relative to two populations from Siberia, and were correctly assigned to their population of origin 100 percent of the time (Weiss *et al.* 2006, pp. 512, 515-516, 518). The populations that were significantly different in terms of their meristic characteristics also exhibited differences in molecular genetic markers (Weiss *et al.* 2006, p. 518).

Inference Concerning Genetic Differences in Arctic Grayling of the Missouri River Relative to Other Examples of the Taxon

We believe the differences between Arctic grayling in the Missouri River and sample populations from Alaska and Canada measured using allozymes (Everett 1986, entire; Leary 2005, entire), mitochondrial DNA (Redenbach and Taylor 1999, entire; Stamford and Taylor 2004, entire), and microsatellite DNA markers (Peterson and Ardren 2009, pp. 1764–1766; USFWS, unpublished data) represent "marked genetic differences" in terms of the extent of differentiation (e.g., F_{st}, R_{st}) and the importance of that genetic legacy to the rest of the taxon. The presence of morphological characteristics separating Missouri River Arctic grayling from other populations also likely indicates genetic differences, although this conclusion is based on a limited number of populations (Everett 1986, pp. 32–35; Weiss *et al.* 2006, entire), and we cannot entirely rule out the influence of environmental variation.

The intent of the DPS policy and the Act is to preserve important elements of biological and genetic diversity, not necessarily to preserve the occurrence of unique alleles in particular populations. In Arctic grayling of the Missouri River, the microsatellite DNA data indicate that the group is evolving independently from the rest of the species. The extirpation of this group would mean the loss of the genetic variation in one of the two most distinct groups identified in the microsatellite DNA analysis, and the loss of the future evolutionary potential that goes with it. Thus, the genetic data support the conclusion that Arctic grayling of the upper Missouri River represent a unique and irreplaceable biological resource of the type the Act was intended to preserve. Thus, we conclude that Missouri River Arctic grayling differ markedly in their genetic characteristics relative to the rest of the taxon.

Upper Missouri River Arctic grayling satisfy the significance criteria outlined in the Services' DPS policy because they occur in a unique ecological setting, are separated from other Arctic grayling populations by a large gap in their range, and differ markedly in their genetic characteristics relative to other Arctic grayling populations. Therefore, we consider the Arctic grayling in the upper Missouri River basin significant to the taxon to which it belongs under the Service's DPS policy.

DPS Conclusion

We find that a population segment of Arctic grayling in the upper Missouri River basin satisfies the discreteness standard of the DPS policy. The segment is physically isolated, and genetic data indicate that Arctic grayling in the Missouri River basin have been separated from other populations for thousands of years. The population segment occurs in an isolated geographic area far south of all other Arctic grayling populations worldwide, and we find that loss of this population segment would create a significant gap in the species' range. Molecular genetic data clearly differentiate Missouri River Arctic grayling from other Arctic grayling populations, including those in Canada and Alaska.

Based on the best scientific and commercial information available, as described above, we find that, under the Service's DPS policy, upper Missouri River Arctic grayling are discrete and are significant to the taxon to which they belong. Because the upper Missouri River population of Arctic grayling is both discrete and significant, it qualifies as a DPS under the Act.

As we described above, we are including introduced Arctic grayling populations that occur in lakes in the upper Missouri River basin as part of the DPS. The Service has interpreted the Act to provide a statutory directive to conserve species in their native ecosystems (49 FR 33885; August 27, 1984) and to conserve genetic resources and biodiversity over a representative portion of a taxon's historical occurrence (61 FR 4722; February 7, 1996). The introduced Arctic grayling populations occur within the boundaries of the upper Missouri River basin and represent moderate to high levels of genetic diversity from within the basin. The future adaptive capabilities represented by this genetic diversity have conservation value, particularly given a changing climate.

We define the historical range of this DPS to include the major rivers, and some tributary streams of the upper Missouri River (mainstem Missouri, Smith, Sun, Beaverhead, Jefferson, Big Hole, and Madison Rivers), as well as a few lakes where Arctic grayling are or were believed to be native (Elk Lake, Red Rock Lakes in the Centennial Valley, Miner Lake, and Mussigbrod Lake, all in Beaverhead County, Montana). We define the current range of the DPS to consist of extant native populations in the Big Hole River, Ruby River, Miner Lake, Mussigbrod Lake, Madison River–Ennis Reservoir, and Centennial Valley, as well as all known introduced populations within the upper Missouri River basin that meet the criteria of (1) occupy natural habitat; (2) reproduce naturally, and (3) are not part of the captive brood (genetic) reserve program. We refer to this entity as the Upper Missouri River DPS of Arctic grayling. The remainder of this finding will thus focus on the population status of and potential threats to this entity.

Population Status and Trends in the Upper Missouri River DPS

The Upper Missouri River DPS of Arctic grayling is comprised of 19 populations (Table 1). Occupied habitat (habitat that is currently used by Arctic grayling for some portion of their life history) is estimated at 314 miles (505 kilometers) of rivers and streams and 6,045 hectares (14,938 acres) of lakes and reservoirs. Fifteen of the 19 populations occur solely on Federal land, two mostly on Federal land (~70% of the Ruby River and ~90% of the Centennial Valley) and two (Big Hole River and Ennis Reservoir/Madison River) occur primarily (>90%) on private land (Table 1).

Big Hole River

Multiple monitoring datasets exist that characterize the Big Hole River population of Arctic grayling (Table 4, Figure 2). Montana Fish, Wildlife and Parks has compiled an electrofishing dataset from 1991 to 2016 that indexes abundance of Arctic grayling as catch-per-unit effort (e.g., fish/mile). A second dataset was analyzed by DeHaan et al. 2014 that estimated genetic diversity (with a suite of metrics), expected rate of loss of diversity (N_e) and the number of breeding adult Arctic grayling (N_b) at four different points in time; 1987-88, 1995-96, 2005-06, and 2011-2012. These four different points in time include samples from two consecutive years, thus they are reported as a range of years; however, these year ranges actually refer to biological dates that precede them by 3-5 years. For example, the year range 2007/08 refers to data that characterizes what was happening in the Big Hole River grayling population in 2002 – 2005.

Table 3. Characteristics of populations within the Upper Missouri River DPS of Arctic grayling.

Population	Drainage	Primary Ownership	Extent[a] **Stream miles** / lake hectares	N_e (95% CI)[b]	N_b (95% CI or **range**)[c]	Annual census population size[d]	Qualitative Descriptor[e]	Biological date of population data[f]
Big Hole River (+11 tributaries)	Big Hole	Private	**199**	see Population Status and Trends	see Population Status and Trends	~500-3000		census (2007-2018)
Ennis Reservoir / Madison River	Madison	Private	**15** / 1469	162 (76 - ∞)		697 – 2,317		1991-1993
Centennial Valley	Red Rock	Federal	**78** / 3756	see Population Status and Trends	see Population Status and Trends			see Population Status and Trends
Mussigbrod Lake	Big Hole	Federal	42	1497 (262 - ∞)		6,437 – 21,407		2001-2003
Miner Lake	Big Hole	Federal	27	286 (143 – 4,692)		1,230 – 4,090		2001-2003
Ruby River	Ruby	Federal	**40**		see Population Status and Trends			see Population Status and Trends
Agnes Lake	Big Hole	Federal	44			~24,000		1972
Odell Lake	Big Hole	Federal	13	577 (222 - ∞)		2,481 - 8251		2001-2003
Bobcat Lake	Big Hole	Federal	2	252 (114 - ∞)		1084 - 3604		2001-2003
Schwinegar Lake	Big Hole	Federal	2				Common	
Pintlar Lake	Big Hole	Federal	16				Common	
Deer Lake	Gallatin	Federal	5			800 – 1,100		1989-2002
Emerald Lake	Gallatin	Federal	6				Abundant	
Grayling Lake	Gallatin	Federal	1				Rare	
Hyalite Lake	Gallatin	Federal	64			499[**]		1998-2012
Gibson Reservoir	Sun	Federal	521				Rare	
Lake Levale	Sun	Federal	5				Abundant	
Park Lake	Missouri	Federal	13				Common	
Grebe Lake	Madison	Federal	59	Infinite[*]		~27,000		1999-2003 (N_e) 1954 (census)

[a]Habitat extent is the amount of habitat currently being used by Arctic grayling for some portion of their life history. It does not mean the amount specified is occupied continuously.
[b]N_e denotes effective population size; a parameter that dictates the rate at which genetic variation is lost and inbreeding accumulates within a population. For more information, see discussion of effective population size in Population Status and Trends in the Upper Missouri River DPS.
[c]N_b denotes the number of reproductively successful adults contributing genetic variation to a given cohort. For more information, see discussion of effective population size in Population Status and Trends in the Upper Missouri River DPS.
[d]Population size of reproductively mature individuals (not to be confused with total annual census population size which includes adults and juveniles) estimated from N_e assuming N_e / N_c = .07 (minimum estimate) and .23 (maximum estimate) for all populations, except the Big Hole River. These two values represent the range of median N_e / N_c ratios for salmonids cited in Palstra and Fraser 2012. Annual census population size for the Big Hole River is taken from Kovach et al. 2019, p. 9 and is different than the range of values reported in the 2014 Finding. This difference is attributable to updated data used in the more recent calculation and does not imply a reduction in number of breeding Arctic grayling in the Big Hole River from 2014 to present.
[e]Qualitative descriptors are from Montana Fish, Wildlife, and Parks MFISH database and are based on biomass estimates where available, or biologist observations and professional biological judgment.

[f]Approximate date to which the N_e, N_b, or annual census population size refers. Biological dates for N_e or N_b estimates refer to the generation of breeders that produced the sample of offspring that were genotyped.
[*]Point estimate for Grebe Lake N_e was negative, indicating no evidence for any disequilibrium caused by genetic drift due to finite number of parents (Perterson and Arden 2009, p. 1767). This population was recently removed as part of a project targeting removal of non-native rainbow trout. About 120,000 Arctic grayling were subsequently reintroduced into Grebe Lake following non-native removal (for more information see Population Status and Trends in the Upper Missouri River DPS).
[**]The N_b estimate for Hyalite Lake is reported as the mean number of adult spawning individuals observed in the spawning run in Hyalite Creek from 1998-2012.

A third dataset was analyzed by Leary 2014 that estimated genetic diversity (with a suite of metrics) and the number of breeding adult Arctic grayling (N_b) on an annual basis from 2007 to 2013. A fourth dataset was analyzed by Whiteley et al. 2018 and included annual N_b estimates from 2007 to 2017, including all N_b data used in Leary 2014. A fifth dataset analyzed by Kovach 2019 includes all N_b data used in Whiteley et al 2018, but includes updated confidence intervals (calculated slightly different due to new information) for the 2007- 2017 N_b estimates reported in Whiteley et al. 2018 and N_b estimates for 2018 and 2019. Overall, these datasets are complimentary and show very similar patterns in abundance and trend of Big Hole River Arctic grayling, which we discuss below.

Table 4. Population monitoring information for the Big Hole River population of Arctic grayling in Montana.

Source	Metric estimated[b]	Grayling Age	Year(s)	Limitations
MFWP 2019, unpublished data	Abundance[c]	Age-0 and Age 1+	1991-2016[d]	Detection probability unknown among years, which can limit inference if changes in abundance are small
DeHaan et al. 2014	N_b, N_e, A_t, A_m, A_r, H_{exp}, H_{obs}	Age-1+	1987-88, 1995-96, 2005-06, 2011-12[e]	Authors combined fish samples from multiple years and multiple cohorts, which results in ambiguous estimates, thus data more appropriate for broad trend analysis
Leary 2014	N_b, A_r, H_e	Age-0	2007-2013	No confidence intervals associated with estimates, however intervals were provided by Kovach et al. 2019
Whiteley et al. 2018[a]	N_b	Age-0	2007-2017	
Kovach et al. 2019	N_b, N_e, A_r H_e	Age-0	2007-2019	

[a]Whiteley et al. 2018 incorporates all of Leary's 2014 data, as well as more recent data collected and analyzed since 2014.

[b] N_b- the number of reproductively successful adults contributing genetic variation to a given cohort. (e.g., age-0 or age-1); N_e- dictates the rate at which genetic variation is lost and inbreeding accumulates within a population; A_t- total number of alleles observed; A_m- mean number of alleles per locus; A_r- allelic richness; H_{exp}- expected

heterozygosity; H_{obs}- observed heterozygosity. A_m was reported by DeHaan et al. 2014, but was not central to the conclusions drawn from the study, thus we omit mention of it in the status review.
[c]Abundance is expressed as catch-per-unit effort (e.g., fish/mile) from electrofishing surveys.
[d]Years that catch per unit effort data were collected for the Big Hole River varied by river segment.
[e]Estimates of effective population size (N_e) were made using different combinations of these years.

Catch per unit effort data indicates grayling abundance in the Big Hole River decreased to historical lows (~1991 to 2006), then recently peaked in 2012, followed by a subsequent decrease and period of relative stability from 2013-2016. Based on data from three long-term monitoring reaches (1991-2006) and newer monitoring in the Candidate Conservation Agreement with Assurances (CCAA; for more information see Water Management in the Upper Missouri River Basin) Project Area (2006-2016), abundance of Age 1+ Arctic grayling in the Big Hole River declined from past levels to historical lows by 2006 (MFWP 2019, unpublished data). Since 2006, grayling abundance has increased and peaked in 2012, after which abundance declined and was relatively stable from 2013-2016 (MFWP 2019, unpublished data). Catch per unit effort estimates and their reliability for trend analysis are influenced by two factors: (1) the abundance (or density) of the species being monitored, and (2) the probability of detecting the species with the sampling gear; neither of these parameters are known nor can be estimated. Detection probability with electrofishing gear, as was used to sample Arctic grayling in the Big Hole River, can be influenced by a variety of factors, including water levels or temperatures, which can make trend analysis difficult when changes in trend are small. However, the magnitude of the historical (1991-2006) decline shown by this data is fairly large, increasing our confidence that the decline did indeed occur and was not wholly attributable to variability in electrofishing detection probability. Thus, these data indicate a historical decline of age 1+ Arctic grayling in the Big Hole River from 1991 to 2006, followed by a temporary peak around 2012, then a decline to a period of relative stability from 2013-2016.

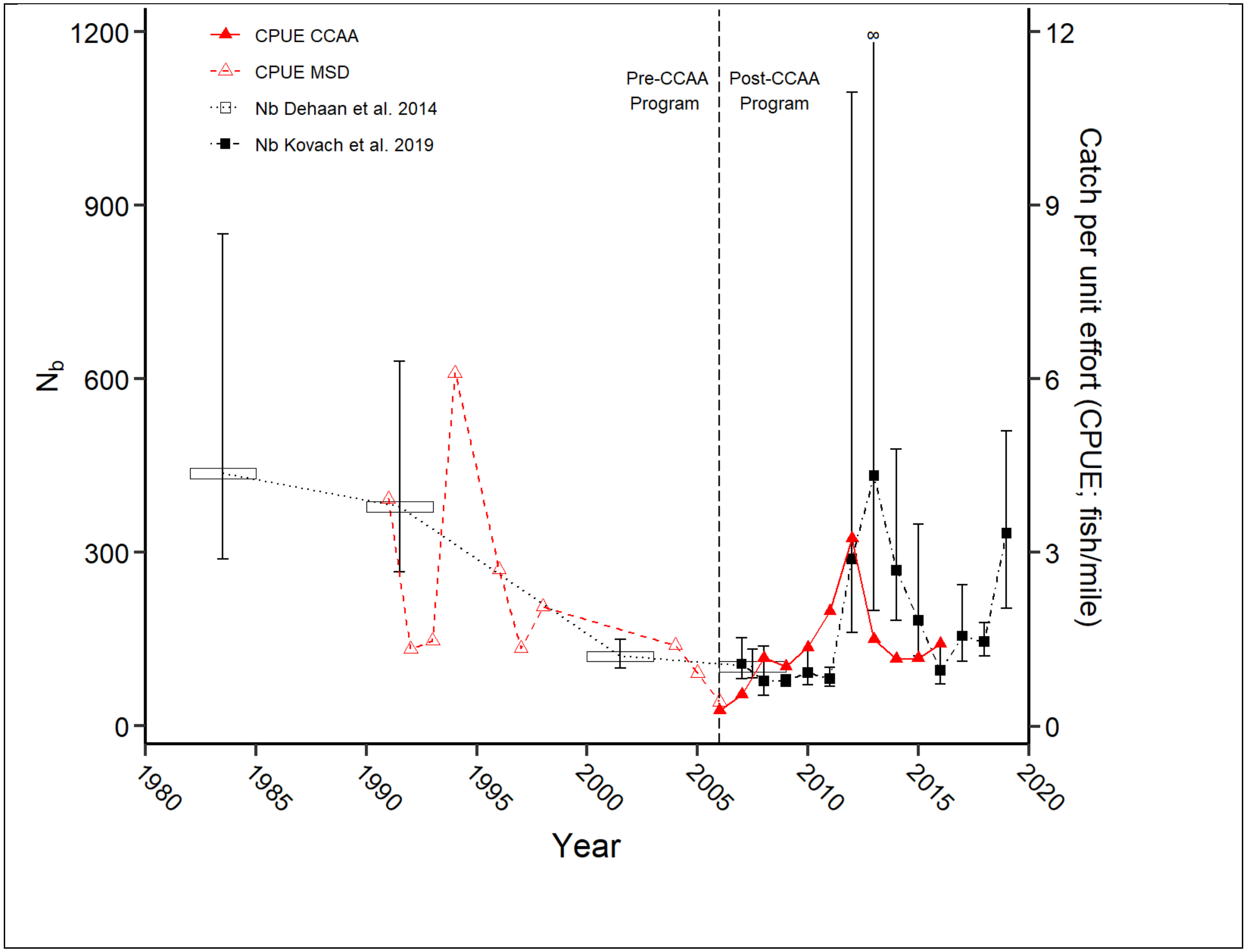


Figure 2. Trends in abundance (indexed as catch-per-unit effort; CPUE) and effective number of breeders (N_b) of Arctic grayling in the Big Hole River population from multiple datasets through time. Effective number of breeders is the number of reproductively successful adults contributing genetic variation to a given cohort. Catch per unit effort data are from three long-term monitoring sections of the Big Hole River (1991-2006; Catch per unit effort MSD) before the inception of the CCAA in 2006 (dashed vertical line) and from standardized sections in the Big Hole CCAA Project Area (2006-2016; Catch per unit effort CCAA). N_b estimates from DeHaan et al. 2014 correspond to the biological range of years that the data represent (i.e., a sample taken in 1987/1988 actually represents individuals that were spawned 3-5 years prior to collection because a mixed age sample was taken). N_b estimates from Kovach et al. 2019 represent annual estimates of effective number of breeders because a single age sample (e.g., age-0) was taken.

Given the challenges of estimating numerical abundance using catch per unit effort mentioned above, Montana Fish, Wildlife and Parks (MFWP) shifted from monitoring catch per unit effort in 2016 to monitoring effective number of breeders (N_b) to characterize both the evolutionary and demographic status of the Big Hole Arctic grayling population. The number of effective breeders (N_b) is the number of reproductively successful adults contributing genetic variation to a given cohort. The N_b represents a powerful tool for genetic monitoring and methodological approaches to estimate N_b have rapidly advanced in the last decade (Waples et al. 2013, entire; 2014, entire). Prior to these advances, DeHaan et al. 2014 attempted to estimate N_b of Arctic grayling in the Big Hole River using archived genetic data from multiple cohorts of grayling (DeHaan et al. 2014, entire). Those data suggested there was a decline in N_b from 1978/88 to 2005/06 (DeHaan et al. 2014, p. 39). While the estimate of N_b was also lower in 2011/12 than in 2005/06 in DeHaan et al. 2014, there was substantial overlap of the confidence intervals, meaning the two estimates (2005/06 and 2011/12) were statistically similar, indicating N_b stabilized during that time period. Unfortunately, by combining genetics data from multiple cohorts, it is challenging to accurately estimate the true number of effective breeders producing a cohort or generation (Waples 2006, entire) but trends in these estimates are informative (DeHaan et al. 2014, pp. 23, 25). To incorporate the most up to date methodological approaches, MFWP began using single cohort estimates of N_b for population monitoring (Leary 2014, entire; Whiteley 2018, entire; Kovach 2019, entire) in 2007. Annual N_b estimates for Arctic grayling in the Big Hole River from 2007 to 2011 were relatively consistent (mean = ~87 breeders across years), but larger and more variable from 2012-2019 (mean = ~237 breeders across years); a 111% increase (and statistically significant) relative to mean N_b for 2007-2011 (Kovach et al. 2019, pp. 8-9, 20, 26). Cumulatively, the trends in N_b mirror the trends in catch-per-effort for the years they overlap. The most recent N_b data suggests the spawning grayling population in the Big Hole River has stabilized from the decline shown by DeHaan et al. 2014 and recently increased to a higher mean number of spawners in the past 7 years, relative to the 2007-2011 time period when MFWP initiated genetic monitoring.

The increase in mean number of effective breeding Arctic grayling in the Big Hole River from the 2007-2011 time period to the 2012-2019 time period could have been influenced by several factors, including use of remote site incubators from 2010-2016, improved spawning and rearing habitat, favorable environmental conditions, or some combination of these factors (Whiteley et al. 2018, pp. 1, 3-6; Kovach et al. 2019, pp. 7-10). However, we note that the 2019 N_b estimate is not expected to be influenced by remote site incubators because they were not being used in or near the tributaries where most of the age-0 grayling were sampled to derive the N_b estimate (Jaeger 2019, pers. comm.). Whatever the reason(s), it is clear that conservation measures are working, either singularly or in concert with one another, at a level high enough to increase the number of effective breeding Arctic grayling, on average, since 2006. Cumulatively, increases in N_b (number of effective breeding Arctic grayling) are contributing to a more robust population of Arctic grayling in the Big Hole River and adding resiliency to the population.

Estimates of the effective number of breeders can be used to further estimate the genetic effective population size (N_e), a statistic often used to inform population and evolutionary viability (Kovach et al. 2019, pp. 4-6). Specifically, N_e is a critical parameter in conservation genetic theory that dictates the rate at which genetic variation is lost and inbreeding accumulates within a population (Franklin 1980, entire). In the Big Hole River, a conservative estimate of N_e, based on the number of effective breeders between 2007 and 2019 was 306 (Kovach et al. 2019, p. 9). DeHaan et al. 2014 used an alternative method and estimated the effective population size for the period spanning 1987-2012 at 371 in the Big Hole River (DeHaan et al 2013, pp. 17, 40). Thus, multiple independent methods suggest that N_e of the Big Hole Arctic grayling population is approximately 300 or larger. Population genetic theory predicts the Big Hole River population of Arctic grayling would lose less than 10 percent of its heterozygosity over the next 50 generations (~200 years) if this effective population size (~300) is maintained (Kovach et al 2019, p. 11).

Heterozygosity and allelic richness (other measures of genetic diversity) represent the future adaptive capacity for Arctic grayling to respond to environmental changes and have been relatively high and stable in Big Hole River Arctic grayling through time (Leary 2014, pp. 5-6; DeHaan et al. 2014, pp. 16, 22, 39; Kovach et al. 2019, pp. 8, 10, 24). For example in the Big Hole River, minor variations in both allelic richness and heterozygosity indicated relatively stable genetic diversity through time (1987-88 to 2011-12; DeHaan et al. 2014, pp. 22, 39). Similarly, allelic richness and heterozygosity in Arctic grayling in the Big Hole River were stable from 2007-2013 (Leary 2014, pp. 5-6), although a decline in allelic richness in Arctic grayling in the Big Hole River was documented by Kovach et al. 2019 (pp. 8, 10, 24)). However, it appears the majority of the decline in allelic richness was from the 1990s to early 2000s; a trend we would expect to see given the magnitude of demographic decline (in the aforementioned catch per unit effort data) that happened during that time. No trend in allelic richness was detected in more recent years (2000s to 2010s; Kovach et al. 2019, pp. 8, 10, 24)). Furthermore, heterozygosity was temporally stable throughout the time series, as expected when the effective population size is large (Kovach et al. 2019, pp. 8-10). Thus, despite large declines in abundance (~1991-2006) and number of effective breeding Arctic grayling (1987-88 through 2005-06; DeHaan et al. 2014, p. 39) in the past, genetic diversity of Arctic grayling in the Big Hole River has remained relatively high and stable.

Multiple lines of evidence indicate the Big Hole River Arctic grayling population has more effective breeders on average than when the CCAA started, stable genetic diversity and is more robust and resilient than in the past. The catch per unit effort data from MTFWP and N_b estimates from DeHaan et al. 2014 indicate population declines in abundance of Arctic grayling and number of effective breeding Arctic grayling in the Big Hole River from the 1990s to 2006. However, data from DeHaan et al. 2014, Leary 2014, Whiteley et al. 2018 and Kovach et al. 2019 indicate N_b estimates stabilized in the Big Hole River from 2003 to 2011. From 2012-2019, N_b estimates increased 111%, on average, compared to estimates from 2007-2011. Despite historical declines in both abundance and number of breeding adults, data from DeHaan et al. 2014, Leary 2014; Whiteley et al. 2018 and Kovach et al. 2019 indicate that little genetic variation has been lost in the Big Hole River population, and current rates of expected loss are very low (<10 percent over 200 years). The general pattern of historical decline in abundance and number of breeders due to historical threats, followed by a stabilization of abundance and N_b as conservation actions began, next followed by an increase in N_b as more conservation actions were implemented, is how we would expect a natural population to respond as the benefits of conservation actions accrue. The population of Arctic grayling in the Big Hole River is demographically and genetically stable and appears to be responding favorably to conservation actions that have been implemented over the past several decades.

Centennial Valley

The number of spawning adult Arctic grayling in a portion of Red Rock Creek used to monitor the Centennial Valley population has fluctuated through time (Figure 3; Gander et al. 2019, pp. 16-17). Spawning adults occurred at lower abundance in the mid-late 1990s, followed by a period of greater abundance in the early 2000s to 2015, then more recently at lower abundance in 2016 through 2019 (Figure 3; Gander et al. 2019, pp. 16-17). A management goal of 1000 spawning adult fish has been set for this population (Gander et al. 2019, p. 2), with the most recent abundance estimates being below that level (2016 to 2019 mean = 230 fish; Warren et al. 2019, p. 1; for more information on management actions to reach the management goal, see Conservation Efforts to Reduce Habitat Destruction, Modification, or Curtailment of Its Range). Although spawning adult estimates from 2016 to 2019 have fluctuated, confidence intervals associated with those estimates overlap substantially, indicating the annual estimates are not statistically different from one another (Warren et al. 2019, p. 1).

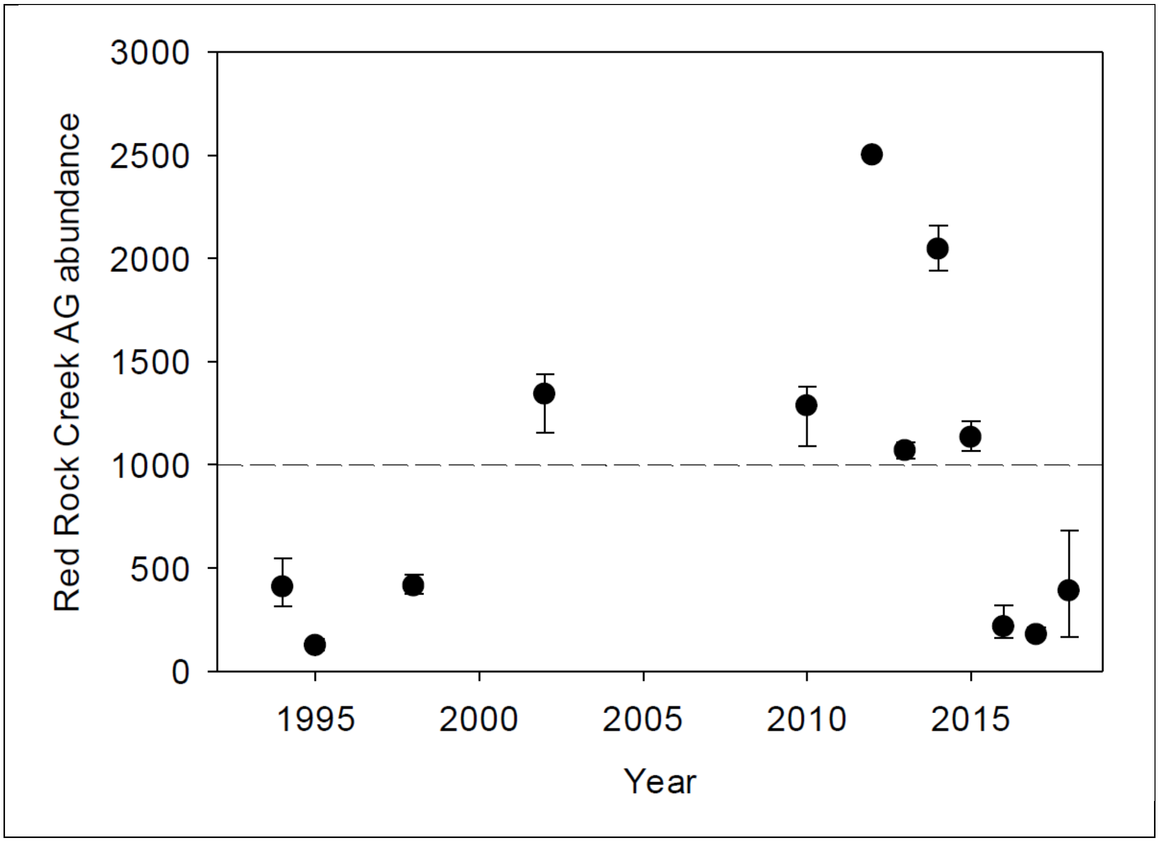


Figure 3. Abundance of spawning adult grayling in Red Rock Creek in the Centennial Valley through time. Dashed horizontal line indicates the management goal (1,000 spawners) for this population. Not shown on this figure is the estimate of number of adult spawners from 2019, which is similar to and not statistically different from any of the estimates in 2016-2018.

Monitoring of effective number of breeders (N_b) to characterize both the evolutionary and demographic status of the Centennial Valley Arctic grayling population has largely indicated stability through time (DeHaan et al. 2014, p. 39; Gander et al. 2019, p. 6), with a recent decline in 2015 (Kovach et al. 2019, pp. 9, 11, 22). The number of effective breeders (N_b) is the number of reproductively successful adults contributing genetic variation to a given cohort. The N_b represents a powerful tool for genetic monitoring and methodological approaches to estimate N_b have rapidly advanced in the last decade (Waples et al. 2013,entire; 2014, entire). Prior to these advances, DeHaan et al. 2014 attempted to estimate N_b of Arctic grayling in the Centennial Valley using archived genetic data from multiple cohorts of grayling (DeHaan et al. 2014, entire). Those data suggest there was an increase in N_b from 1998 to 2012 (DeHaan et al. 2014, p. 39). Unfortunately by combining genetics data from multiple cohorts, it is challenging to accurately estimate the true number of effective breeders producing a cohort or generation (Waples 2005, entire), but trends in these estimates are informative (DeHaan et al. 2014, pp. 23, 25). To incorporate the most up to date methodological approaches, MFWP began using single cohort estimates of N_b for population monitoring (Leary 2014, entire; Whiteley 2018, entire; Kovach 2019, entire). Annual N_b estimates for Arctic grayling in the Centennial Valley from 2010 to 2014 were relatively consistent ranging from 207 to 406 (Gander et al. 2019, p. 6; Kovach et al. 2019, pp. 11, 22, 26), with N_b decreasing to about 40 in 2015 (Kovach et al. 2019, pp. 11, 22, 26). The 2015 N_b estimate should be interpreted cautiously due to a smaller sample size, but is consistent with the recent decline in the number of spawning adult Arctic grayling in Red Rock Creek (Kovach et al. 2019, pp. 11, 21, 22).

Estimates of the effective number of breeders can be used to further estimate the genetic effective population size, a statistic often used to inform population and evolutionary viability (Kovach et al. 2019, pp. 3, 7, 8). Specifically, N_e is a critical parameter in conservation genetic theory that dictates the rate at which genetic variation is lost and inbreeding accumulates within a population (Franklin 1980, entire). In the Centennial Valley an estimate of N_e, based on the number of effective breeders between 2010 and 2014 was 190 (Kovach et al. 2019, pp. 9, 22). DeHaan et al. 2014 used an alternative method and estimated a range of effective population sizes of 166 to 291 for the period spanning 1998-2012 in the Centennial Valley (DeHaan et al. 2014, pp. 17, 40), which encompasses the estimate from Kovach et al. 2019. Thus, multiple independent methods suggest that N_e of the Centennial Valley Arctic grayling population is large enough to avoid immediate effects of inbreeding, but the population may lose genetic diversity in the long-term if unfavorable overwinter conditions persist or are not mitigated (Kovach et al 2019, p. 12).

Allelic richness, a measure of genetic diversity sensitive to rapid demographic change, has declined in Centennial Valley Arctic grayling though time (Kovach et al. 2019, pp. 8, 25). This is consistent with known declines in abundance of spawning Arctic grayling in Red Rock Creek (Kovach et al. 2019, p. 10). In contrast to allelic richness, heterozygosity, a critical measure of genetic diversity in population genetic theory, has remained relatively stable through time, despite known demographic declines (Kovach et al. 2019, pp. 5, 8, 25). This result is expected in populations with relatively large effective population sizes and suggests that past demographic declines likely followed a pattern of short duration and moderate intensity (Kovach et al. 2019, p. 10).

Multiple lines of evidence indicate the Centennial Valley grayling population has a stable, but lower number of adult spawners than in the recent past, yet relatively high genetic diversity with a relatively robust effective population size. Little genetic variation has been lost in the population, despite the recent decline in adult spawners. Rate of expected loss of genetic diversity is low if current effective population size is maintained.

Ruby River

Arctic grayling were reintroduced into the Ruby River using stocking and remote site incubators from 1997 to 2008 to establish a stable, naturally reproducing population and provide redundancy of the fluvial ecotype with the historical range of Arctic grayling (Gander et al. 2019, p. 2). As part of the original Montana Fluvial Arctic Grayling Restoration Plan, any reintroduced population would be considered stable and viable in a stream when monitoring confirms that, for at least 10 years, successful stock recruitment exceeds mortality of reproductive adults to successfully compensate for stochastic factors and perpetuate the species within suitable habitats (Montana Fluvial Arctic Grayling Restoration Plan 1995, p. 1). One way to measure whether this goal has been achieved is to monitor natural reproduction of Arctic grayling. Most historical reintroduction attempts in riverine environments have resulted in Arctic grayling being present near the reintroduction area for several years (Kaya 1990, pp. 31-35), but ultimately disappearing, due in part to lack of natural reproduction. Without natural reproduction, Arctic grayling are not able to persist for more than one generation (~3.5 years). Thus, evidence of natural reproduction is critical to ensuring Arctic grayling are able to fulfill their entire life history in the streams or rivers where they were reintroduced.

Recently, we became aware of more specific objectives (relative to the Montana Fluvial Arctic Grayling Restoration Plan) that were also outlined for the Ruby River Arctic grayling reintroduction in 1996 (Byorth 1996, entire). This document was recently digitized and was submitted as part of this status review. These objectives were created to aid in achieving the overarching goal of the Montana Fluvial Arctic Grayling Restoration Plan and included: (1) Monitoring survival, movements, and densities of introduced grayling to determine factors affecting success of reintroduction, (2) Through monitoring, document natural reproduction by 2002, and (3) Attain stable to increasing population densities in sampling sections where natural reproduction equals or exceeds annual mortality for three consecutive years (Byorth 1996, p. 2).

Monitoring of the Arctic grayling population within the Ruby River from 2009-2018 has indicated natural reproduction for 10 consecutive years (Gander et al. 2019, p. 4), thus the Ruby River population has met the criteria for a viable population outlined in the Montana Fluvial Arctic Grayling Restoration Plan. This is also consistent with our assertion in the 2010 Finding for Arctic grayling that 5-10 more years of monitoring would be needed to deem this population viable (75 FR 54743); we now have 8 additional consecutive years of monitoring documenting natural reproduction since 2010 (Gander et al. 2019, p. 4). The three objectives in the Upper Ruby River Fluvial Arctic Grayling Reintroduction Plan have also been met. Monitoring of natural recruitment (survival part of the objective), spatial distribution (where fish have moved in the system; movement part of the objective) and abundance (density part of the objective) of Arctic grayling have been conducted every year since 2009 to determine factors affecting success (Gander et al. 2019, p. 4), thus this objective has been met. Prior to 2009, natural reproduction was not documented because fish produced by remote site incubators could not be differentiated from naturally-reproduced fish and high mortality of stocked fish in the early years of the reintroduction effort (Gander et al. 2019, p. 2). However, as a result, reintroduction methods were changed to using remote site incubators, and natural reproduction has now been documented for 10 consecutive years (Gander et al. 2019, p. 4), thus meeting this objective. In addition, stable population densities have been achieved in sampling sections where natural reproduction equals or exceeds annual mortality for 3 consecutive years (MTFWP 2019, unpublished data). We note that population densities of Arctic grayling in the Ruby River experienced large declines from 2009 to 2010, which is likely an artifact of overshooting the actual carrying capacity of the river with artificially high numbers of Arctic grayling produced by remote site incubators. From 2010 to 2018, density of Arctic grayling has fluctuated up and down, as would be expected in a wild population affected by natural factors, but otherwise appears stable.

In addition to the monitoring outlined in the original Montana Fluvial Arctic Grayling Restoration Plan and Upper Ruby River Fluvial Arctic Grayling Reintroduction Plan, MFWP has also monitored genetic metrics in the Ruby River population of Arctic grayling. The genetic metrics of heterozygosity (H_e) and allelic richness (A_r) provide information on how genetically diverse the population is and represent the future adaptive capacity for Arctic grayling to respond to environmental changes. The number of effective breeders (N_b) provides information on how many breeding individuals contributed genetics to each cohort (year class) of grayling. Since 2010, heterozygosity and allelic richness have been relatively high and stable, indicating a relatively diverse, stable population (Gander et al. 2019, p. 5). Allelic richness was determined to be lower than the founding population (i.e., Big Hole River stock from broodstock ponds), and is likely an artifact of the small number of grayling families that founded the original population (Leary et al. 2015 *in* Gander et al. 2019, p. 5). The N_b estimates from the Ruby River increased from 2010 through 2012 and have since declined through 2017 (Gander et al. 2019, p. 5). This data indicates a recent decline in the number of breeding adults contributing their genetics to individual cohorts of Arctic grayling. However, if N_b

continues to decline or does not increase in the next several generations (i.e., ~7-10 years), losses of genetic diversity may occur (Gander et al. 2019, p. 5). Future monitoring is warranted to track genetic diversity of the population (Gander et al. 2019, pp. 5-6).

Overall, Ruby River Arctic grayling have naturally reproduced for 10 consecutive years. Current genetic data indicate a diverse, stable population and all objectives and goals of the Montana Fluvial Arctic Grayling Restoration Plan and Upper Ruby River Fluvial Arctic Grayling Reintroduction Plan have been met, indicating the Ruby River population of Arctic grayling is viable. Monitoring will continue in the future to track genetic metrics characterizing the genetic health of the population. Therefore, this population contributes to the redundancy and representation of the varied life history of Arctic grayling in the Upper Missouri River basin.

Madison River

No recent standardized surveys have been conducted to assess the status of Arctic grayling in the Madison River. Multiple captures of angled grayling in the Madison River drainage have been reported since 2016 (MFWP 2019 unpublished data). No inference of trend can be made from these observations, but it appears Arctic grayling still persist and reproduce at presumably low levels in the system. Past surveys of young Arctic grayling in Ennis Reservoir and spawning adults in the Madison River both indicate a declining population (Clancey and Lohrenz 2009, pp. 30, 74). Our interpretation of this data is that the population of Arctic grayling in the Madison River continues to persist at presumably low abundance. However, reports of angled grayling in the headwaters of the Madison River (e.g., Gibbon River, Grayling Creek) are encouraging.

Other Lake Populations

The 15 other populations of Arctic grayling reside in higher elevation mountain lakes, many of which are remote and difficult to access. As such, monitoring of some of these populations is infrequent and is typically in the form of quantitative genetics monitoring, qualitative surveys during spawning, disease testing and angler reports (MFWP 2019, unpublished data; Moser 2019, pers. comm.; Olsen 2019a, pers. comm.; Vivian 2019, pers. comm.). Despite infrequent monitoring, the recent monitoring we are aware of indicate persistence of grayling populations and/or robust spawning runs in many of the lakes (MFWP 2019, unpublished data; Moser 2019, pers. comm., Vivian 2019, pers. comm.). Most of these lakes have continuously supported Arctic grayling populations since the 1930s, with no stocking of grayling in recent years (Gander 2020, pers. comm). Thus, the presence of grayling in these lakes currently indicates that successful natural reproduction is occurring. No threats to Arctic grayling or their habitat are known or predicted in these lakes. Thus, despite infrequent monitoring, the continued persistence of these populations is occurring and expected to continue, given the long-term track record of these lakes supporting Arctic grayling populations over the last 80 years or more.

A recent non-native fish removal project at Grebe Lake removed all fish from the lake, including Arctic grayling, to establish a headwater population of cutthroat trout and Arctic grayling (Koel et al. 2019, p. 6-8). Approximately 120,000 Arctic grayling were subsequently reintroduced to Grebe Lake once the fish removal project was complete. We have no direct evidence of natural reproduction in Grebe Lake since the reintroduction because the reintroduced grayling are not old enough to spawn yet. However, we have no evidence that natural reproduction will not take place based on observations of Arctic grayling using inlet and outlet streams to spawn in Grebe Lake prior to the fish removal project (Kruse 1959, pp. 318-322), the longevity of the former grayling population prior to removal (almost 100 years; Kruse 1959, p. 312) and that no stream or spawning habitat was altered during the fish removal project.

Summary of Information Pertaining to the Five Factors

Since the Arctic grayling in the upper Missouri River basin qualifies as a DPS, we will now evaluate its status with regard to its potential for listing as endangered or threatened based on the five factors enumerated in section 4(a) of the Act.

Section 4 of the Act (16 U.S.C. 1533) and the implementing regulations at part 424 of title 50 of the Code of Federal Regulations (50 CFR part 424) set forth procedures for adding species to, removing species from, or reclassifying species on the Lists. The Act defines ‘‘endangered species’’ as any species that is in danger of extinction throughout all or a significant portion of its range (16 U.S.C. 1532(6)), and ‘‘threatened species’’ as any species that is likely to become an endangered species within the foreseeable future throughout all or a significant portion of its range (16 U.S.C. 1532(20)). Under section 4(a)(1) of the Act, a species may be determined to be an endangered species or a threatened species because of any of the following five factors:

(A) The present or threatened destruction, modification, or curtailment of its habitat or range;

(B) Overutilization for commercial, recreational, scientific, or educational purposes;

(C) Disease or predation;

(D) The inadequacy of existing regulatory mechanisms; or

(E) Other natural or manmade factors affecting its continued existence.

In considering whether a species may meet the definition of an endangered species or a threatened species because of any of the five factors, we must look beyond the mere exposure of the species to the threat to determine whether the species responds to the threat in a way that causes actual impacts to the species. If there is exposure to a threat, but no response, or only a positive response, that threat does not cause a species to meet the definition of an endangered species or a threatened species. If there is exposure and the species responds negatively, we determine whether that threat drives or contributes to the risk of extinction of the species such that the species warrants listing as an endangered or threatened species. The mere identification of threats that could affect a species negatively is not sufficient to compel a finding that listing is or remains warranted. For a species to be listed or remain listed, we require evidence that these threats are operative threats to the species and its habitat, either singly or in combination, to the point that the species meets the definition of an endangered or a threatened species under the Act.

In conducting our evaluation of the five factors provided in section 4(a)(1) of the Act to determine whether the Upper Missouri River DPS of arctic grayling meets the definition of “endangered species” or “threatened species,” we considered and thoroughly evaluated the best scientific and commercial information available regarding the past, present, and future threats. We reviewed the petition, information available in our files, and other available published and unpublished information. This evaluation may include information from recognized experts; Federal, State, and tribal governments; academic institutions; foreign governments; private entities; and other members of the public.

Our evaluation of the Upper Missouri River DPS of Arctic grayling follows. In making our revised 12-month finding on the petition, we consider and evaluate the best available scientific and commercial information. This evaluation includes all factors we considered in our previous findings and, at the end of this analysis, explains whether and how the Services’ conclusions differ now.

Factor A. The Present or Threatened Destruction, Modification, or Curtailment of Its Habitat or Range

Curtailment of Range and Distribution

The range and distribution of Arctic grayling in the upper Missouri River basin was reduced over the past 100 years (Kaya 1992, p. 51), primarily due to historical habitat fragmentation by dams and irrigation diversions and by habitat degradation or modification from unregulated land use (Vincent 1962, pp. 97-121). Arctic grayling typically need large expanses of connected habitat to fulfill their life-history stages (Armstrong 1986, p. 8). For example, Arctic grayling in the Big Hole River have been documented migrating over 60 miles (97 km) between overwintering, spawning, and foraging habitats (Shepard and Oswald 1989, pp. 18-21, 27). These past reductions in range and distribution may have reproductively isolated Arctic grayling populations within the basin (Peterson and Ardren 2009, p. 1770).

Although the range and distribution of Arctic grayling has contracted in rivers and streams from historical levels, expression of the full range of known life histories is represented in the DPS and distribution of Arctic grayling occupying lakes has increased, relative to historical levels. Arctic grayling occupying flowing water occur in the Big Hole and Ruby rivers, Centennial Valley (Long and Red Rock creeks) and Ennis Reservoir/Madison River (mainstem Madison River). Large reaches of connected riverine habitat remain in these areas and still permit the expression of this life history, despite the presence of mainstem dams in three of four watersheds (Kaya 1992, entire; see Figure 1). Arctic grayling are known to occupy all lakes that were historically occupied (although they are not reproducing in Elk Lake currently because of low spring flows in the primary spawning tributary), as well as 13 additional lakes in the Upper Missouri River basin (Table 3). Arctic grayling populations are still present in 7 of 10 historically occupied watersheds in the upper Missouri River basin (see "Drainage" column in Table 3). Thus, despite historical curtailment of range in rivers and streams, the full range of known life history behaviors is represented in the DPS and there has been a range expansion for Arctic grayling populations occupying lakes.

Dams on Mainstem Rivers

Much of the historical range of the Upper Missouri River DPS of Arctic grayling has been altered by the construction of dams and reservoirs (Kaya 1990, pp. 51–52; Kaya

1992, p. 57). The construction of large dams on mainstem river habitats throughout the upper Missouri River system fragmented river corridors necessary for the expression of Arctic grayling migratory life histories in some systems. Construction of dams that obstructed upstream fish passage on the mainstem Missouri River (Hauser, Holter, Canyon Ferry, and Toston dams), Madison River (Madison–Ennis, Hebgen dams), Beaverhead River and its tributary Red Rock River (Clark Canyon, Lima dams), Ruby River (Ruby dam), and Sun River (Gibson dam) all likely contributed to the historical decline of Arctic grayling in the DPS (Vincent 1962, pp. 127–128; Kaya 1992, p. 57). Lack of upstream fish passage at these dams contributed to the extirpation of Arctic grayling from some waters by blocking migratory corridors (Vincent 1962, p. 128), curtailing access to important spawning and rearing habitats, and impounding water over former spawning locations (Vincent 1962, p. 128). Most dams within the upper Missouri River basin were constructed between 1905 and 1960 (Kaya 1990, entire).

Despite the construction of multiple dams throughout the historical range of Arctic grayling, multiple populations exhibiting the full range of known life histories are represented in the current DPS. These populations reside in areas where sufficient quantity and quality of habitat exist and permit the expression of the full range of life histories. In some cases, dams may be providing a benefit, because currently many of the dams that historically affected Arctic grayling populations are now precluding invasion by nonnative fish from downstream sources. For example, Lima Dam in the Centennial Valley is currently precluding brown trout invasion from downstream sources (Mogen 2014, pers. comm). Currently, there are four Arctic grayling populations within the current DPS that occur above mainstem dams (Centennial Valley, Ruby River, Hyalite

Lake, and Gibson Reservoir) with at least one nonnative fish species occurring downstream of each of these dams (MFISH 2014d, unpublished data).

Some reservoirs created by dams are currently being used by Arctic grayling as overwintering, rearing and foraging areas. Both adult and juvenile Arctic grayling use Ennis Reservoir for overwintering, rearing, and foraging (Byorth and Shepard 1990, entire). In the Centennial Valley, Arctic grayling have recently been detected in Lima Reservoir (MFISH 2014e, unpublished data). The movements of Arctic grayling within and out of Lima Reservoir are unknown; however, Lima Reservoir is a large reservoir and, as such, is likely used for overwintering purposes.

Arctic grayling have been documented in stream and river reaches below some dams, most likely indicating downstream passage of fish over or through dams. These fish are essentially "lost" to the population residing above the dam, because none of the mainstem river dams in the upper Missouri River basin provides upstream fish passage. Substantial losses from a population resulting from downstream entrainment of fish through dams could cause declines in reproductive potential and abundance in the reservoir population above the dam (Kimmerer 2008, entire). However, it is unknown what entrainment rates currently are in populations residing near dams. Rate of entrainment is dependent on a number of factors, including dam operations, season, water conditions in the reservoir, initial population size above the dam, etc. Recent monitoring data and angler reports of Arctic grayling observed downstream of reservoirs supporting Arctic grayling populations are sporadic (Horton 2014c, pers. comm.; Service 2014).

Historically, operational practices at Madison Dam have likely affected the Arctic grayling population in Ennis Reservoir/Madison River. A population decline in Arctic grayling appeared to coincide with a reservoir drawdown in the winter of 1982–1983 (Byorth and Shepard 1990, pp. 52–53). This drawdown likely affected the forage base, rearing habitat, and spawning cycle of Arctic grayling in the reservoir. However, under a new licensing agreement dated September 27, 2000, between the Federal Energy Regulatory Commission and Ennis Dam operators, such substantial drawdowns in elevation of Ennis Reservoir are no longer permitted (Clancey 2014, pers. comm.).

Given the above information, mainstem dams historically impacted Arctic grayling populations in the upper Missouri River basin and likely still preclude the expression of the full range of life histories in some areas of historical range. However, large, connected habitats remain intact currently, despite the presence of mainstem dams on some rivers. These intact habitats are large enough to allow Arctic grayling to express their full range of life histories. Dams still impact individuals, because some Arctic grayling are likely being entrained and lost from their source population; however, we have no evidence that mainstem river dams are affecting extant Arctic grayling populations at the population or DPS level.

Water Management in the Upper Missouri River Basin

The predominant use of private lands in the upper Missouri River basin is irrigated agriculture and ranching. These activities have historically had significant effects on aquatic habitats, primarily changes in water availability and alteration of the structure and function of aquatic habitats. Changes in water availability can affect Arctic grayling reproduction, survival, and movements among habitat types (Kaya 1990, entire).

In contrast to most of the Arctic grayling populations in the Upper Missouri River DPS that occur on Federal land, the population of Arctic grayling in the Big Hole River occurs on primarily (~90 percent) private land. Thus, any conservation efforts conducted in the Big Hole River Valley need support from involved agencies and private landowners. While intermittent conservation actions to benefit Arctic grayling started in the 1990s in the Big Hole Valley, a candidate conservation agreement with assurances (CCAA; Montana Fish, Wildlife, and Parks *et al*. 2006, entire) was developed in 2006 as a more comprehensive conservation approach for Arctic grayling in the Big Hole River. The conservation goal of this CCAA was to secure and enhance the fluvial population of Arctic grayling in the upper Big Hole River drainage. Conservation projects conducted under the CCAA are prioritized and guided by the Big Hole Arctic Grayling Strategic Habitat Conservation Plan (hereafter Strategic Habitat Conservation Plan) (for more specific information, see "Conservation Efforts to Reduce Habitat Destruction, Modification, or Curtailment of Its Range," below).

Since 2006, many conservation and restoration projects have been completed in the upper Big Hole River under the direction of the CCAA and Strategic Habitat Conservation Plan (Table 5). Below, we describe and evaluate the implementation and effectiveness of these projects relative to the potential stressors analyzed under Factor A for the Big Hole River population. We also analyze the effects of potential stressors under Factor A for the other Arctic grayling populations in the DPS.

Table 5. Conservation projects and results, and Arctic grayling response in the Big Hole River since 1999, including implementation of the Big Hole CCAA in 2006. All information on conservation projects and conservation results cited from unpublished database, Montana Fish, Wildlife, and Parks 2019.

Threat factor	Stressor	Conservation projects[a]	Conservation result	Arctic grayling response
A	Dams/ habitat fragmentation	Fish ladders: **61** Bridges : **18** Grade control structures: **9** Siphons**: 1**	Stream miles (%) accessible to grayling[a]: • Tier I- 82(**98%; pre-CCAA=87%**) • Tier II- 61(**67%; pre-CCAA=27%**) • Tier III- 32(**20%: pre-CCAA=6%**)	Arctic grayling distribution has increased 18 miles in the Big Hole River and its tributaries -Rock Creek (young-of-year and Age 1+) -Big Lake Creek (Age 1+; Strategic Habitat Conservation Plan 2013, p. 12) -McVey Creek (Age 0 and 1+; Olsen 2019a, pers. comm.) -Warm Springs Creek (Age 1+; MFWP 2019, unpublished data); -Upper Big Hole River near Jackson (MFWP 2019, unpublished data). Average number of effective breeders has increased from **~87** (2007-2011) to ~**237** (2012-2019) (Kovach et al. 2019)[b] Stable genetic diversity as measured by heterozygosity and allelic richness (Leary 2014, pp. 5-6; DeHaan et al. 2014, pp. 16, 22, 39; Kovach et al. 2019, pp. 8, 10, 24). Effective population size of approximately 300 (less than 10% loss of genetic diversity over the next 200 years at current effective population size; Kovach et al. 2019, pp. 9-12).
	Dewatering/ Thermal stress	Points of diversion: **425** of **504** with signed site specific plans Irrigation Efficiency and Flow Measuring: **194** Stockwater Projects: **70** Stream Restoration Projects: **29** Rock Creek restoration	• Achievement of instream flow goals increased from 50% (pre-CCAA) to 80% (post-CCAA) • Landowner contributions to streamflow increasing, on average, as # of points of diversion with signed site specific plans increase **[landowner contribution to instream flows in Big Hole River (pre-2006 = 0 cfs; 2019 = 220 cfs)]**[c] • Temperature reductions in tributaries (see Rock Creek example below) Pre-restoration (2007): • **36 days max. temp >70 °F** • **16 days max. temp >77 °F** Post-restoration (2013): • **0 days max temp. >70 °F**	
		Instream Flow Contributions	• Decrease duration of water temperatures in mainstem Big Hole River stressful or lethal to Arctic grayling (MFWP 2019, unpublished data)	
		Riparian restoration	• Decrease duration of water temperatures in tributaries stressful or lethal to Arctic grayling (MFWP 2019, unpublished data; McCullough 2019, entire)	
	Entrainment	Fish screens: **2**		

	Prioritized monitoring protocol	• Observed low entrainment rates in unscreened ditches from 2014 (252 fry) to 2018 (0 fry)
Riparian habitat loss	Stream restoration: **29** Riparian enhancement: **66** Stock water: **70** Grazing mgmt. plans: **21** landowners (**85,000** ac.) Noxious weed management Willow planting (**72,200** planted)	• 79% increase in Sustainable enrolled acres (pre-CCAA = 61 miles, 2019 = 110 miles) • 27% decrease in At Risk enrolled acres) pre-CCAA = 128 miles, 2019 = 93 miles) • 76% decrease in Unsustainable enrolled acres (pre-CCAA = 17 miles, 2019 = 4 miles) • Adaptive management in place to address non-improving areas

[a]Tier I is core spawning, rearing and adult habitat that is currently occupied by Arctic grayling, Tier II is periphery habitat intermittently used by Arctic grayling, Tier III is suitable, but currently unoccupied historical habitat; [b]The estimates of number of breeding adults in the Big Hole River differs from the 2014 Finding because of newer data and newer statistical methodologies in estimating N_b; [c]Landowner contributions to instream flows do not always increase annually because the contributions are influenced by factors such as weather (i.e., a year with more precipitation and higher flows will not require as much instream flow contributions from landowners due to flow targets being exceeded more often).

Habitat Fragmentation/Smaller Seasonal Barriers

Big Hole River: Smaller dams or diversions associated with irrigation structures historically posed a threat to Arctic grayling migratory behavior, especially in the Big Hole River drainage. In the Big Hole River, numerous diversion structures have been identified as putative fish migration barriers (Petersen and Lamothe 2006, pp. 8, 12–13, 29) that may limit the ability of Arctic grayling to migrate to spawning, rearing, or sheltering habitats under certain conditions. As with the larger dams, these smaller fish passage barriers can reduce reproduction (access to spawning habitat is blocked), reduce growth (access to feeding habitat is blocked), and increase mortality (access to refuge habitat is blocked). Historically, these types of barriers were numerous and widespread across the Big Hole River drainage.

Currently, habitat fragmentation due to irrigation diversion structures in the Big Hole is being systematically reduced under the CCAA (for more specific information, see "Conservation Efforts to Reduce Habitat Destruction, Modification, or Curtailment of Its Range") and Strategic Habitat Conservation Plan. Since 1999, 61 fish ladders have been installed in the mainstem Big Hole River and tributaries (Table 4). Multiple culverts have been replaced with bridges and several grade control structures have been installed (Table 4). As a result, no fish barriers now exist in the mainstem upper Big Hole River. Almost all (98 percent) of tier I habitat and the majority (68 percent) of tier II habitat is connected and accessible to Arctic grayling (Table 4): 67 miles of stream have been reconnected in the Big Hole River system since 1999 (MFWP 2014a, unpublished data; for more specific information, see Water Management in the Upper Missouri River

Basin). This is important because these projects have connected habitats that Arctic grayling have been documented to use for spawning, rearing and thermal refuge. Arctic grayling now have the option and ability to move around freely within almost all Tier 1 habitat and avoid water temperatures that may be stressful or lethal by using thermal refugia in the mainstem river or tributaries. This connectivity has increased the resilience of the Big Hole River population by providing Arctic grayling a connected, diverse suite of habitats to complete their life cycle.

Other populations: Smaller fish passage barriers also have been noted to affect Arctic grayling in the Centennial Valley (Unthank 1989, p. 9). Historically, spawning Arctic grayling migrated from the Jefferson River system, through the Beaverhead River and Red Rock River through the Red Rock Lakes and into the upper drainage, and many likely returned downstream after spawning (Henshall 1907, p. 5). The construction of a water control structure (sill) at the outlet of Lower Red Rock Lake in 1930 (and reconstruction in 1957 (USFWS 2009, p. 74)) created an upstream migration barrier that blocked these migrations (Unthank 1989, p. 10; Gillin 2001, p. 4-4). However, recent changes in water management at the Red Rock Lakes National Wildlife Refuge (NWR) have resulted in year-round fish passage through the control structure at the outlet of Lower Red Rock Lake (West 2013, pers. comm.).

In Mussigbrod Lake, Arctic grayling occasionally pass downstream over a diversion structure at the lake outlet, and become trapped in an isolated pool (Olsen 2014, pers. comm.). During high-snowpack years, Arctic grayling likely can swim back up to the lake from the pool, but in low snowpack years, some Arctic grayling perish when the isolated pool dries up (Olsen 2014, pers. comm.). However, this phenomenon has occurred periodically in recent history and has had no discernible impacts on Arctic grayling abundance in Mussigbrod Lake (Olsen 2014, pers. comm.).

The remaining 16 Arctic grayling populations in the upper Missouri River basin are not influenced by smaller seasonal barriers because none are known to be present. The effect of a barrier at the outlet of Mussigbrod Lake is likely impacting individuals, but not the population because of the robust population size in Mussigbrod Lake and historical stability of that population since the outlet structure was created.

Degradation of Riparian Habitat

Riparian corridors are important for maintaining habitat for Arctic grayling in the upper Missouri River basin, and in general are critical for the ecological function of aquatic systems (Gregory *et al.* 1991, entire). Riparian zones are important for Arctic grayling because of their effect on water quality and water temperature, and their role in maintaining natural ecological processes responsible for creating and maintaining necessary physical habitat features (i.e., pools, riffles, and scour areas) used by the species to meet its life-history requirements.

Big Hole: Arctic grayling abundance in the upper Big Hole River is positively related to the presence of overhanging vegetation, primarily willows (*Salix* spp.), that is associated with pool habitat (Lamothe and Magee 2004, pp. 21–22). Removal of willows and riparian clearing concurrent with livestock and water management along the upper Big Hole River has led to a shift in channel form (i.e., braided channels becoming a single wide channel), increased erosion rates, reduced cover, increased water temperatures, and reduced recruitment of large wood debris into the active stream channel (Confluence Consulting *et al.* 2003, pp. 24–26). These factors combine to reduce the suitability of the habitat for species like Arctic grayling (Hubert 1985, entire).

Currently, restoration of riparian areas in the upper Big Hole River system is a priority under the CCAA (for more specific information, see "Conservation Efforts to Reduce Habitat Destruction, Modification, or Curtailment of Its Range," below). Since 2006, efforts to restore and conserve riparian habitats have been numerous and multi-faceted (see Table 4). Riparian health is an important component of the Big Hole CCAA because riparian health scores are correlated with water temperature degree-days (i.e., change in water temperature), where healthier riparian areas were related to less change in water temperature (i.e., less warming from solar inputs; McCullough 2019, p. 7-8).

Riparian areas enrolled in the Big Hole CCAA are monitored every 5 years using a standardized riparian protocol (NRCS 2004, entire) and have shown considerable improvement through time. Riparian health is binned into three categories (from high health to low health); Sustainable, At Risk and Unsustainable (for scoring metrics, see NRCS 2004). About 207 miles (333 km; 61%) of riparian habitat are currently enrolled in the Big Hole CCAA, out of a total of about 340 miles (547 km) of total riparian habitat in the CCAA Management Area. Of the 207 miles of enrolled riparian habitat, there are currently 110 miles (177 km; 53 percent) functioning as sustainable; a 78% increase since initial riparian surveys were conducted (MFWP 2019, unpublished data). Initial riparian surveys were conducted in varying years for each enrolled property, depending on when the site-specific plans were finalized. Another 93 miles (150 km; 45 percent) of riparian habitat are functioning at risk; a 27 percent decrease (i.e., improved habitat conditions) since initial riparian surveys were conducted (MFWP 2019, unpublished data). About 4 miles (6 km; 2 percent) of riparian habitat are functioning as unsustainable; a 76 percent decrease (i.e., improved habitat conditions) since initial riparian surveys were conducted (MFWP 2019, unpublished data). These data show improvement in riparian area health trends associated with the implementation of the Big Hole River CCAA. In addition, adaptive management within the CCAA framework will allow for reevaluation of conservation measures being implemented in riparian habitats that have not improved.

Other populations: In the Centennial Valley, historical livestock grazing both within the Red Rock Lakes NWR and on adjacent private lands negatively affected the condition of riparian habitats on tributaries to the Red Rock Lakes (Mogen 1996, pp. 75–77; Gillin 2001, pp. 3-12, 3-14). In general, degraded riparian habitat limits the creation and maintenance of aquatic habitats, especially pools, which are preferred habitats for adult Arctic grayling (Lamothe and Magee 2004, pp. 21–22; Hughes 1992, entire), although some spawning adult Arctic grayling in Red Rock Creek outmigrate soon after spawning and likely do not use available pool habitat (Jordan 2014, pers. comm.). Loss of riparian vegetation increases bank erosion, which can lead to siltation of spawning gravels, which may in turn harm Arctic grayling by reducing the extent of suitable spawning habitat and reducing survival of Arctic grayling embryos already present in the stream gravels.

In 2010, the Red Rock Lakes NWR acquired land on Red Rock Creek, upstream of the refuge boundary (West 2014a, pers. comm.). Much of this parcel was riparian habitat that was historically heavily grazed; thus, the refuge implemented a rest-rotation grazing system where more durable lands are grazed while more sensitive lands (e.g., riparian areas) are rested for up to 4 years. On average, grazing intensities on the refuge have decreased from 20,000 Animal Unit Months (AUMs, number of cow/calf pairs multiplied by the number of months grazed) to about 5,000 AUMs. As a result of these changes, riparian habitat within the refuge has dramatically improved (West 2014b, pers. comm.) and is expected to continue improving under the new grazing regime. Given the riparian improvements within Red Rock Lakes NWR, and that the refuge represents the vast majority of current Arctic grayling habitat in the Centennial Valley, we do not expect riparian health to be limiting the Centennial Valley population of Arctic grayling on the Red Rock Lakes NWR.

In the Centennial Valley on non-Federal lands, riparian areas are degraded in some areas. One priority of the Centennial Valley CCAA (for more in-depth discussion, see Conservation Efforts to Reduce Habitat Destruction, Modification or Curtailment of Range) is to restore riparian areas and improve Arctic grayling habitat through similar conservation actions that have been successfully implemented in the Big Hole CCAA (e.g., grazing management, fencing, active restoration). Given the Centennial Valley CCAA has the same conservation measures as the Big Hole CCAA, we expect improvements to riparian habitat similar to what we have observed in the Big Hole River. However, since the Centennial Valley CCAA has only recently been implemented, we are not basing any conclusion about the status of the Arctic grayling population on any anticipated future benefits from the Centennial Valley CCAA in this review.

Most of the riparian habitat surrounding high-elevation lakes on Federal land where the remaining populations are found is intact and of high quality (MFISH 2014a, unpublished data; MFWP 2014e, unpublished data; USFS 2014, p. 2). These habitats are in remote locations, national parks or wilderness areas with little anthropogenic disturbance. Riparian habitat is expected to remain intact on Federal land because of existing regulatory mechanisms (see in Factor D discussion, below). Riparian habitat in the Big Hole River is expected to continue improving because of the proven track record of conservation evidenced by the current upward trend in riparian habitat quality.

Dewatering From Irrigation and Increased Water Temperatures

Demand for irrigation water in the semi-arid upper Missouri River basin historically dewatered many rivers formerly or currently occupied by Arctic grayling. The primary effect of this dewatering is increased water temperatures. In ectothermic species like salmonid fishes, water temperature sets basic constraints on species' distribution and physiological performance, such as activity and growth (Coutant 1999, pp. 32–52). Increased water temperatures can reduce the growth and survival of Arctic grayling (physiological stressor). Reduced habitat capacity can concentrate fishes and thereby increase competition and predation (ecological stressor). Below we discuss the potential effects of increased water temperature on the Upper Missouri River DPS of Arctic grayling.

Three water temperature thresholds are biologically important for Arctic grayling. First, 21°C (70°F) is the temperature at which salmonids, including adult Arctic grayling, begin to experience physiological stress (Table 6; Chadwick et al. 2015, pp. 5-8; references *in* Vatland 2015, pp. 33-34). Physiological stress is sublethal (does not kill the fish), but can manifest as increased metabolism (Cho et al. 1982, entire) and reduced growth (Al-Chokhachy et al. 2013, p. 3073), among other factors. Second, 25°C (77°F) is the temperature at which 50% of juvenile Arctic grayling are expected to die if exposed to this water temperature constantly for 7 days (Table 6; upper incipient lethal temperature; Lohr et al. 1996, p. 934). Third, 26.9°C (80.4°F) is the water temperature at which adult Arctic grayling lose equilibrium and cannot escape conditions that will promptly lead to death (Table 6; critical thermal maximum); Feldmeth and Eriksen 1978, p. 2041]. In general, juvenile Arctic grayling can tolerate warmer water temperatures than adults (Feldmeth and Eriksen, p. 2041; Lohr et al. 1996, pp. 935-937), which is particularly relevant because much of the upper Big Hole River is an important rearing area for fry and juveniles (Magee and Lamothe 2003, pp. 13, 18-21).

Table 6. Temperature tolerances for three different life stages (adult, juvenile, fry) of Arctic grayling determined by different methods and varying acclimation temperatures.

Life stage	Onset of physiological stress	Upper incipient lethal temperature (acclimation temperature)	Critical thermal maximum (acclimation temperature)	Reference
Adult	21°C			Chadwick et al. 2015; references *in* Vatland 2015
Adult			26.9°C (13°C)	Feldmeth and Eriksen 1978
Juvenile		23.0°C (8.4°C)		Lohr et al. 1996
Juvenile		23.0°C (16.0°C)		Lohr et al. 1996
Juvenile		25.0°C (20.0°C)		Lohr et al. 1996
Juvenile			29.3°C (20.0°C)	Lohr et al. 1996
Juvenile			28.5°C (16.0°C)	Lohr et al. 1996
Juvenile			26.4°C (8.4°C)	Lohr et al. 1996
Fry			28.7°C (13.0°C)	Feldmeth and Eriksen 1978

The upper incipient lethal temperatures and critical thermal maximums are typically determined in a laboratory and use a range of methods and water temperatures (critical thermal maximums = slowly increasing, and upper incipient lethal temperatures=constant) to elicit stress or death in fish. However, neither methodology accounts for daily temperature fluctuations or durations of time at specific water temperatures during those fluctuations that fish experience in the wild (Johnstone and Rahel 2003, pp. 96-98; Hokansen et al. 1977, pp. 643-647). Consideration of daily temperature fluctuations is important because fish exposed to these fluctuations can survive higher water temperatures than those determined for fish exposed to constant or slowly increasing water temperatures (critical thermal maximums and upper incipient lethal temperatures; Johnstone and Rahel 2003, pp. 96-98). This phenomenon occurs because wild fish are typically only exposed to otherwise lethal water temperatures (upper incipient lethal temperatures or critical thermal maximums) for a portion of a day (minutes or hours), not constantly for consecutive days (Johnstone and Rahel 2003, pp. 96-98; Dickerson and Vinyard 1999; pp. 518-520; Lohr et al. 1996, p. 936), which allows for some level of physiological recovery during the cooler portions of the water temperature cycle (Johnstone and Rahel 2003, pp. 97-98; Dickerson and Vinyard 1999; p. 520). Thus, the upper incipient lethal temperatures and critical thermal maximum water temperature thresholds need to be considered in the appropriate context (i.e., some consideration of temperature duration) and should be viewed as conservative estimates when analyzing potential effects of water temperature on wild Arctic grayling experiencing daily temperature fluctuations in natural habitats, such as the Big Hole River.

It is well-established that acclimation temperature (the water temperature at which fish are acclimated to before thermal threshold testing) can affect thermal thresholds of fish (Brett 1952, entire; Cherry et al. 1977, pp. 241-246; Lohr 1996, pp. 935-938;

Beitinger et al. 2000, pp. 242-263). In general, higher acclimation temperatures result in higher water temperature thresholds for species, up to a point (Brett 1952, entire; Cherry et al. 1977, pp. 241-246; Lohr 1996, pp. 935-938; Beitinger et al. 2000, pp. 242-263). The two upper water temperature thresholds reported for Arctic grayling (Table 6; 25°C and 26.9°C) were determined using Arctic grayling acclimated to 20°C (68°F) and 13°C (55°F), respectively (Lohr et al. 1996, pp. 934-937; Feldmeth and Eriksen, pp. 2040-2041), which are water temperatures consistently observed in the spring and summer in the Big Hole River (MTFWP 2019, unpublished data). In addition, Arctic grayling used in determining these thermal tolerances were offspring from Big Hole River Arctic grayling (Lohr et al. 1996, p. 934). Thus, these water temperature thresholds, although conservative, are meaningful for assessing effects on wild adult Arctic grayling in the Big Hole River and elsewhere in the upper Missouri River basin.

Despite the limitations of applying laboratory-derived thermal thresholds to wild fish experiencing daily water temperature fluctuations, the upper incipient lethal temperature is similar to field-based occupancy thresholds for some fish (Vatland 2015, pp. 34-35). For example, in the Big Hole River, abundance of age 1+ Arctic grayling (measured as catch-per-unit-effort) was significantly lower (<5 fish per kilometer) when water temperatures exceeded 25°C, relative to when water temperatures were below 25°C (McCullough 2017, pp. 16-17, 49). While it was outside the scope of the McCullough 2017 study to identify the mechanism(s) for this relationship (i.e., were fish dying or were they moving out of these areas and avoiding detection), multiple lines of evidence suggests that water temperatures around 25°C may be prompting Arctic grayling to emigrate out of an area in search of cooler water. This evidence includes; (1) Arctic grayling in the Big Hole River are experiencing water temperatures >25°C for only portions of day, not constantly for consecutive days (MFWP 2019, unpublished data) as they would be if tested under the upper incipient lethal temperature methodology (and thus are not expected to be dying), (2) no fish kills have been observed in the Big Hole River when daily water temperatures have been greater than 25°C but below 26.9°C (MFWP 2019, unpublished data), (3) the documentation of salmonids, including Arctic grayling in the Big Hole River, actively seeking thermal refuge during warm periods (for references, see full discussion of this topic later in this section), and (4) fish experiencing fluctuating water temperatures have higher thermal tolerances and longer resistance times than those determined using constant water temperatures in a laboratory (Hokansen et al. 1977, pp. 642-647; Feminella and Matthews 1984, pp. 458-460; Johnstone and Rahel 2003, pp. 96-98).

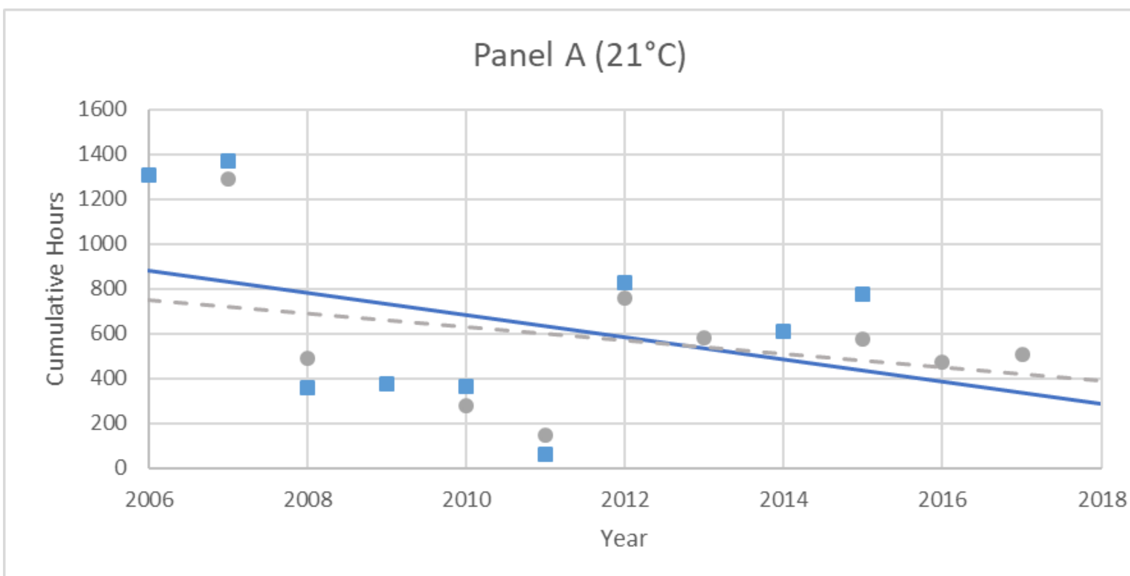
Historical summer water temperatures in the Big Hole River and its primary tributaries (within the CCAA boundary) were assessed to determine how often water temperature thresholds of 21°C (70°F) and 25°C (77°F) were met or exceeded in the past [decades 1980-89 (hereafter 1980s) and 2000-09 (hereafter 2000s); Vatland 2015, pp. 26-59]. Water temperature thresholds were determined to have been met when the weekly mean of the daily maximum (WMDM) water temperature was observed to equal or exceed the threshold value (21°C or 25°C, respectively). A simplified way to visualize this metric is as a running seven-day average of the daily maximum temperatures. It is important to note that a WMDM is an average, thus for a given threshold (21°C or 25°C), daily maximum temperatures within any given 7 day period are going to be higher and lower than the mean. Both the proportion of weeks WMDMs exceeded both 21°C and

25°C thresholds and amount of total stream network exceeding these same thresholds increased from the 1980s to 2000s in the Big Hole River and tributaries (Vatland 2015, pp. 48, 50-51). Cumulatively, these data indicate past warming in the Big Hole River and tributaries, both in terms of frequency (more frequent exceedances of water temperature thresholds through time) and space (increases in amount of stream network exceeding water temperature thresholds).

Vatland 2015 also modelled projected exceedance of the 21°C and 25°C water temperature thresholds for the future decades 2040-49 (hereafter 2040s) and 2060-69 (hereafter 2060s; Vatland pp. 47-51). Given the specific focus of Vatland 2015 on the effects of climate change and water temperatures on Arctic grayling, we present Vatland's modeling of future water temperatures in the 2040s and 2060s and analyze the potential effects of these scenarios in the "Climate Change/Dewatering/Water Temperature Increases" section of the Finding.

Since the end of Vatland's study in 2010, more empirical water temperature data has been collected in the Big Hole River and its tributaries and shows stressful water temperature conditions for Arctic grayling have decreased in duration within the CCAA Project Area (Figure 4; MFWP 2019, unpublished data). Ten thermographs (water temperature recorders) were deployed to monitor water temperatures in the mainstem Big Hole River and one tributary in each of the five Management Sections of the CCAA Area. In some years, data were lacking because one or more of the thermographs were lost or malfunctioned; these years were excluded from analysis due to incomplete data. However, on average, cumulative number of hours that maximum daily water temperatures reached or exceeded the 21°C and 25°C water temperature thresholds in the mainstem Big Hole River and tributaries has trended downward since the inception of the CCAA (Figure 4). These data indicate that stressful temperature conditions for Arctic grayling in the Big Hole River and its tributaries, although still present, have decreased in duration since the inception of the CCAA.

In addition, considerable heterogeneity in water temperatures (changes in water temperatures over short distances) exists in the Big Hole River and its tributaries (Vatland et al. 2015, pp. 31, 38-40, 43-44; McCullough 2019, pp. 6-7, 14). This heterogeneity is often in the form of groundwater mixing or tributary mouths (Magee and



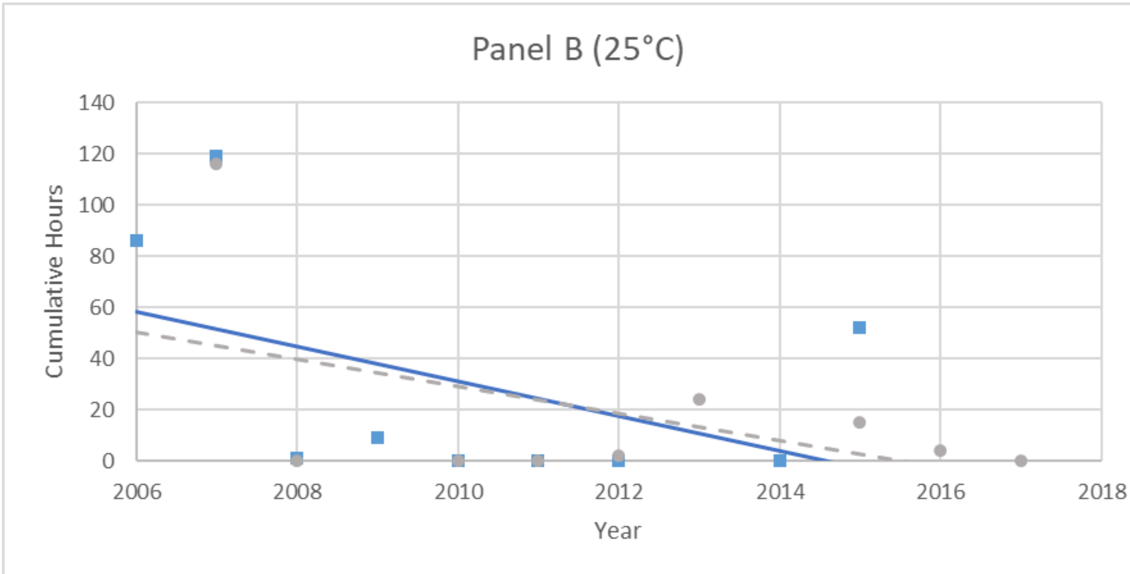


Figure 4. Cumulative hours maximum daily water temperatures equaled or exceeded water temperature thresholds of 21°C (Panel A) and 25°C (Panel B) at monitoring sites in the mainstem Big Hole River (squares; both panels) and tributaries (circles; both panels). Trendlines for cumulative hours of water temperature threshold exceedance at mainstem water temperature monitoring sites (solid; both panels) and tributary water temperature monitoring sites (dashed; both panels) were added to show trends through time.

Lamothe 2003, pp. 13, 30; Vatland 2015, pp. 53-54) and is important because it provides thermal refugia for Arctic grayling (Magee and Lamothe 2003, pp. 13, 30; Vatland 2015, pp. 53-54) when water temperatures elsewhere in the system become stressful. The extent of water temperature heterogeneity in the Big Hole River and its tributaries is typically not captured by using fixed site water temperature monitoring stations (Vatland et al. 2015, pp. 31, 38-40, 43-44), as is currently being done in the Big Hole River and its tributaries (MFWP 2019, unpublished data). Water temperature data from these fixed site monitoring stations is intended to capture broad trends in water temperature across a large landscape, not characterize fine scale water temperatures within each tributary or section of mainstem river.

Most of the water temperature recorders on tributaries to the Big Hole River are placed at locations near their mouth (where the tributary meets the Big Hole River) because of ease of deployment and retrieval. As such, water temperatures recorded at these sites are often the warmest water temperatures in the entire tributary (MFWP 2019, unpublished data). This pattern of cooler water upstream and warmer water downstream is fairly consistent in the Big Hole tributaries (McCullough 2019, pp. 6-7, 14), mainstem Big Hole River (Vatland et al. 2015, pp. 34-35, 38-39) and elsewhere in the West (Isaak et al. 2015, pp. 2541; Isaak et al. 2016, p. 2). Despite water temperatures exceeding the 21°C or 25°C thermal thresholds for Arctic grayling at some of the downstream tributary temperature recording sites for portions of some days, water temperatures cool relatively quickly in the upstream direction and cooler water refugia are typically in close proximity upstream of the monitoring sites (MFWP 2019, unpublished data; McCullough 2019, pp. 6-7, 14).

In the Big Hole River, Arctic grayling have demonstrated their ability to seek, find, and use thermal refugia in tributaries when water temperatures in the mainstem become warm (Magee and Lamothe 2003, pp. 13, 24, 30; Lamothe and Magee 2003, p. 17; Olsen 2019b, pers. comm.). Fish response to changing water temperatures are a well-studied phenomenon (Berman and Quinn 1991, pp. 307-310; Baird and Krueger 2003, pp. 1198-1199, 1202-1204; Nielsen et al. 1994, pp. 620-624; Thorpe 1994, p. 607; Beitinger et al. 2000, p. 238-239; Magoulick and Kobza 2003, entire; Schaefer et al. 2003, pp. 394-395; 398-399; Breau et al. 2007, pp. 1183-1186, 1188-1189; Breau et al. 2011, pp. 850-851; Dugdale et al. 2015, pp. 7-8, 10-14). A common theme in these studies is that fishes have the ability to detect changing water temperatures [documented changes as small as 0.5°C(0.9°F); Bardach and Bjorklund 1957, pp. 238-244] and change their behavior to avoid or escape thermally hostile areas (Neill and Magnuson 1974, p. 706; Coutant 1975, pp. 575-581, 591; Richards et al. 1977, entire; Breau et al. 2011, pp. 850-851; Dugdale et al. 2015, pp. 7-8, 10-14). Salmonids, a cool-water group of fish including Arctic grayling, use these same behavioral adaptations to avoid unfavorable water temperatures and find thermal refugia (Wojcik 1955, pp. 25-26; LaPerriere and Carlson 1973 pp. 32-33; Berman and Quinn 1991, pp. 307-310; Baird and Krueger 2003, pp. 1198-1199, 1202-1204; Nielsen et al. 1994, pp. 620-624; Matthews and Berg 1997, pp. 61, 63; Beitinger et al. 2000, p. 239; Quigley and Hinch 2006, p. 429; Breau et al. 2007, pp. 1183-1186, 1188-1189; Vatland et al. 2009, pp. 10-15; Breau et al. 2011, pp. 850-851; references in Chadwick et al. 2015, p. 9; Dugdale et al. 2015, pp. 7-8, 10-14). In the Big Hole River, Arctic grayling are seeking, finding, and using thermal refugia areas in the lower part of the CCAA Project Area (e.g., Fishtrap, LaMarche creeks; Magee and Lamothe 2003, pp. 13, 24, 30; Lamothe and Magee 2003, p. 17) or parts of the mainstem Big Hole River downstream of the CCAA area that have high quality pool habitats and cooler water temperatures (Olsen 2019b, pers. comm.), when other areas of the mainstem Big Hole River or tributaries become warm.

Overall, Arctic grayling have defined thermal tolerances, but these tolerances are typically laboratory-derived and do not fully account for the effects of natural, cyclic water temperatures on wild fish. Many considerations are needed to provide the appropriate context when assessing effects of water temperature on Arctic grayling, including but not limited to, the magnitude of the water temperature, duration of exposure to that water temperature, daily fluctuations in water temperature, magnitude of daily fluctuations, acclimation temperatures, presence of groundwater or tributary mouths providing thermal refugia, and limitations of fixed-site water temperature sampling designs. In the Big Hole River, historical average water temperatures have increased from the 1980s to 2000s. However, more recent empirical data indicate water temperatures considered stressful or lethal to Arctic grayling have trended downward in both the mainstem Big Hole River and tributaries and considerable heterogeneity exists in the Big Hole River system in the form of groundwater inputs and tributaries. Arctic grayling have been documented seeking, finding and using this heterogeneity when water temperatures become stressful or potentially lethal elsewhere in the system.

Since 2006, water conservation and restoration projects associated with the Big Hole Arctic grayling CCAA (for more specific information, see "Conservation Efforts to Reduce Habitat Destruction, Modification, or Curtailment of Its Range," below) have been implemented to increase instream flows and reduce water temperatures in the Big Hole River and tributaries. Varying flow targets for different management segments of the Big Hole River were outlined in the CCAA, based on the wetted perimeter method, a biologically-based method for determining instream flow requirements to provide necessary resources for all life stages of Arctic grayling. Over 400 irrigation diversions are operated under flow agreements within finalized site-specific plans (Table 5; MDNRC 2019, unpublished data). Although we are aware of the future potential of more points of diversion being managed under signed site plans to contribute to Arctic grayling conservation, these anticipated future efforts do not contribute to Arctic grayling conservation currently, and as such, we have not considered them as part of this status review or our listing determination for this DPS. Multiple additional other projects designed to decrease dewatering and thermal stress have been implemented since 2006 (Table 5). The collective result of these efforts are increasing streamflows, increased access to cold-water refugia via fish ladders, and marked temperature reductions, particularly in some tributaries (Table 5).

Specific flow targets were developed for the different Management Segments in the CCAA Management Area (see MFWP *et al.* 2006, pp. 7, 9, 13, for more information on CCAA Management Segments). The goal for increasing instream flow was to achieve flow targets 75 percent of days in each Management Segment during years of average or greater snowpack. This goal was based on a comparison between minimum flow targets and historical streamflows recorded in Management Segments C and D. Achieving flow targets 75 percent of days in each Management Segment was intended to be a general goal because many other factors influence instream flows in the Big Hole River that are outside the control of landowners (e.g., snowpack, precipitation). Before implementation of the CCAA (2000-2005), average flow targets were met among all Management Segments 50 percent of the time, and since implementation of the CCAA (2006-2018), they have been met 80 percent of the time (MFWP 2019, unpublished data). Thus, the CCAA flow targets are currently being met.

Consistently since 2006, one management area, known as Management Segment C, has exhibited the lowest instream flows among all Management Segments. In part, instream flows in Management Segment C are influenced by several large diversions immediately upstream of the flow measuring device at the downstream boundary of Management Segment C (Robert 2014, pers. comm.). Some of this diverted water is returned to the Big Hole River downstream of the flow measuring device (Robert 2014, pers. comm.). As such, instream flows in Management Segment C represent the "worst case" scenario among all Management Segments. The Montana Department of Natural Resources and Conservation conducted an analysis of this "worst case" scenario, to explore how instream flows in Management Segment C have changed since the inception of the Big Hole CCAA. Given that natural factors such as summer precipitation and annual snowpack influence instream flows in the Big Hole River, the analysis of instream flows in Management Segment C included comparisons among several years of similar (but below average) snow pack and similar summer precipitation, both before and after CCAA implementation (Table 7).

Flows in Management Segment C were less than the Spring flow target of 160 cfs (14.5 cubic meters/second (m^3/s)) for 52 percent fewer days post-CCAA (28 days) than pre-CCAA (58 days; Table 7), on average. Similarly, number of days where flows were less than the Summer/Fall flow target of 60 cfs (1.7m^3/s) decreased 33 percent pre- to post-CCAA (average of 123 days (pre-CCAA) to 83 days (post-CCAA)). Number of days instream flows were below 20 and 10 cfs (0.6 and 0.3 m^3/s, respectively) (these flows represent common, historical low water levels in Management Segment C) were reduced 73 percent, and 100 percent, respectively, from pre- to post-CCAA implementation (Table 7). In brief, there has been an observed 317 percent increase in average discharge from July through September (from 14 to 45 cfs (0.4 and 1.3 m^3/s, respectively)) as a result of achieving flow targets in Management Area C for a higher percentage of days post-CCAA, relative to pre-CCAA (Roberts 2014, 2019, unpublished data). Landowner contributions to instream flow from reducing irrigation withdrawals appears to be the primary factor increasing instream flows in the Big Hole River in late summer (Table 7). Increasing discharge in larger rivers such as the Big Hole River is the primary mechanism found to best mitigate water temperature increases in summer (Poole and Berman 2001, pp. 790-792; Allan 1995, entire); a critical thermal period for Arctic grayling.

Table 7. Comparison of number of days varying flow targets were achieved among similar years of below average snowpack in the Big Hole River CCAA Management Segment C, pre- and post CCAA. All information in this table cited from Roberts 2014, unpublished data and Roberts 2019, unpublished data.

	Pre-CCAA		Post-CCAA	
	1988	2003	2012	2015
Peak snowpack (percent of average)	73[a]	108	81	75[a]
May – Aug. precipitation (in.)	4.14	3.85	4.74	4.08
July – Aug. temps (degrees F; departure from normal)	-1.3	8.0	1.4	-1.0
Signed site specific plans	0	0	12	20
Landowner contributions[b] (cfs)	0	0	252	220
Days <160[c] cfs	50	8	11	17
Days <60[c] cfs	123	123	87	78
Days <20 cfs	79	68	0	17
Days <10 cfs	65	7	0	0
Mean discharge (cfs; July – Sept.)	8.4	19.7	45	44
Mean discharge (cfs; Aug.)	1.1	14.2	34	27
Total Days <Streamflow target	173	131	98	95

[a]Normalized to base period 1971-2000; [b]Landowner contributions to instream flows do not always increase annually because the contributions are influenced by factors such as weather (i.e., a year with more precipitation and higher flows will not require as much instream flow contributions from landowners due to flow targets being exceeded more often); [c]160 cfs = flow target for Spring (April – June), 60 cfs = flow target for Summer and Fall (July – October) in CCAA Management Area C; Cfs = cubic feet per second.

Despite Management Segment C exhibiting the lowest rate of instream flow achievement relative to the other Management Segments, we note that the proportion of Tier I habitat encompassed by Management Segment C is 12 percent; the remainder of Tier I habitat (88 percent) is located in Management Segments D and E (MFWP 2014c, unpublished data). Since the initiation of the Big Hole Arctic grayling CCAA in 2006 to 2013, average achievement rate of instream flow goals in Management Segments D and E during the spring is 96 percent and 99 percent, respectively. Average achievement rate of instream flow goals in Management Segments D and E during the summer/fall is 84 percent and 76 percent, respectively. More recent data from 2017 indicate similar achievement rates; 100 percent for the spring target in both Management Segments D and C and 76 and 78 percent achievement of the summer flow target in those Segments, respectively (MFWP 2019, unpublished data). Thus, flow targets are being met.

Other populations:

Centennial Valley

Summer water temperatures that are stressful to grayling may exist in Upper Red Rock Lake, given its large surface area, yet relatively shallow depth. However, Arctic grayling in the lake appear to be able to cope with these temperatures by using cooler tributaries and spring sources as thermal refugia (Jaeger 2014b, pers. comm.). For example, the presence of Arctic grayling in the lower 100 m (328 ft) of East Shambow Creek in 1994 was attributed to fish seeking refuge from high water temperatures in the lake (Mogen 1996, p. 44). The Centennial Valley, in particular, appears to have many cool spring-fed tributaries that are accessible to Arctic grayling and are used intermittently (Mogen 1996, p. 44). Mean summer water temperatures in Red Rock Creek can occasionally exceed 21ºC (70ºF) during drought conditions (Mogen 1996, pp. 19, 45); however, on average, water temperatures are relatively cool and well within the thermal tolerances for Arctic grayling (USFWS 2012, unpublished data; USGS 2019, unpublished data) and could provide thermal refugia for Arctic grayling. For example, from 2016 to 2019, maximum water temperatures in Red Rock Creek did not exceed 21°C at any time (USGS 2019, unpublished data). Other tributaries in the Centennial Valley are similarly cool; maximum water temperatures in three of five tributaries monitored from 2016-2019 did not exceed 21°C (70ºF) during those years (MFWP 2019, unpublished data). One monitored tributary that did exceed 21°C (70ºF; Tom Creek) did so for about 19 hours a year, on average, from 2016-2019 (MFWP 2019, unpublished data). Biologically, this means that Arctic grayling may have been subjected to water temperatures that would have caused stress for 19 hours a year, on average. However, this is a low amount of time relative to the total duration of time during summer and even if all 19 hours of stressful water temperatures occurred on a single day, effects would be expected to be temporary and non-lethal (see Dewatering From Irrigation and Increased Water Temperatures). In addition, the above scenario also assumes that Arctic grayling did not move out of the area and seek thermal refugia when water temperatures became stressful, which is unknown in this case, but certainly within the capability of Arctic grayling and has been documented in other systems (see Dewatering From Irrigation and Increased Water Temperatures). The last monitored tributary, Metzel Creek, did exceed 21°C (70ºF) and 25°C (77ºF) multiple times throughout 2016 and 2017, but the measurements were influenced by moving the temperature recorder multiple times and the presence of several geothermal warm springs in the area (Brummond 2019, pers. comm).

Ruby River

The most recent water temperature data we are aware of indicate maximum daily water temperatures exceeded 21°C (70°F) in certain sections of the Ruby River on some years (e.g., 2014 and 2015); however, did not exceed 21°C (70°F) in other sections during those same years (MFWP 2015, unpublished data). Duration of time maximum daily water temperature exceeded 21°C in the some sections of the Ruby River was relatively short; less than 104 hours per year in 2015 and less than 54 hours per year in 2014 (excluding one site that is influenced by geothermal activity; MFWP 2015, unpublished data). Biologically, this means that Arctic grayling may have been subjected to water temperatures that would have caused stress for 104 hours a year and 54 hours a year, respectively. However, both these durations are low relative to the total duration of time during summer and even if all the hours of stressful water temperatures occurred sequentially, effects would be expected to be temporary and non-lethal (see Dewatering From Irrigation and Increased Water Temperatures). In addition, the above scenario also assumes that Arctic grayling did not move out of the area and seek thermal refugia when water temperatures became stressful, which is unknown in this case, but certainly within the capability of Arctic grayling and has been documented in other systems (see Dewatering From Irrigation and Increased Water Temperatures). Maximum daily water temperatures did not exceed 25°C (77°F) in either 2014 or 2015 in any section of the

Ruby River (MFWP 2015, unpublished data), thus no mortality of Arctic grayling would be expected (see Dewatering From Irrigation and Increased Water Temperatures). Cumulatively, these data suggest maximum daily water temperatures occur that can be stressful to Arctic grayling (i.e., >21°C), but durations of time water temperatures are over this threshold are relatively short and other sections of river remain under these thresholds. Similar to the Big Hole River, water temperatures in the Ruby River are monitored at fixed sites, which do not document the heterogeneity of water temperatures often present in natural river systems or cool water inputs in the form of groundwater or tributaries (for a more in-depth discussion on these limitations, see Dewatering From Irrigation and Increased Water Temperatures). Regardless of this limitation, the duration of time water temperatures were above the 21°C threshold to initiate stress is low.

Madison River

Mean and maximum summer water temperatures can exceed 21 ºC (70 ºF) in the Madison River below Ennis Reservoir (U.S. Geological Survey (USGS) 2019, unpublished data), and have exceeded 22 ºC (72 ºF) in the reservoir, and 24 ºC (75 ºF) in the reservoir inlet (Clancey and Lohrenz 2005, p. 34). However, water temperatures in the Madison River upstream of Ennis Reservoir are often cooler and available to Arctic grayling for thermal refugia, if needed. For example in 2017 and 2018, water temperatures in the Madison River rarely exceeded 21°C and would have provided Arctic grayling cooler water temperatures (USGS 2019, unpublished data). Additionally in 2017 and 2018, duration of time water temperatures were greater than 21°C was relatively short (i.e., hours; USGS 2019, unpublished data).

Lake populations

Increased water temperatures do not appear to be prevalent in most other populations, likely due to the high elevation of these habitats and the intact nature of riparian areas bordering inlet tributaries. No thermal fish kills have ever been reported in any of the high mountain lakes occupied by Arctic grayling. Although water temperatures may increase with climate change in the future, the high elevation and existing cold water are not expected to rise outside of the range of thermal tolerances for Arctic grayling.

Entrainment

Entrainment results when fish are transported along with the flow of water out of their natural habitat (e.g., river, lake or reservoir) into unnatural habitat, typically in the form of irrigation ditches. Entrainment can permanently remove individual fish from a natural population and strand them in a habitat that lacks the required characteristics for reproduction and survival. Entrainment in Arctic grayling habitat results when irrigation headgates are closed and ditches subsequently are dewatered, resulting in mortality of entrained Arctic grayling.

Big Hole: Entrainment of individual Arctic grayling in irrigation ditches historically occurred and currently occurs in the Big Hole River (Skarr 1989, p. 19; Streu 1990, pp. 24–25; MFWP *et al.* 2006, p. 49; Lamothe 2008, p. 22; MFWP 2013b, unpublished data; MFWP 2019, unpublished data). Over 1,000 unscreened diversion structures occur in the upper Big Hole River watershed, and more than 300 of these are located in or near occupied Arctic grayling habitat (MFWP *et al.* 2006, pp. 48–49).

Recent entrainment surveys in irrigation ditches along the mainstem Big Hole River and tributaries indicate low levels of juvenile Arctic grayling entrainment (MFWP 2019, unpublished data). From 2014 to 2018, 283 juvenile Arctic grayling have been documented being entrained (average of 57 per year) in irrigation diversions, most of which were young-of-year (i.e., fry; MFWP 2019, unpublished data). Although this number is higher than we reported in the 2014 finding, it is still very low relative to the size of the population and relative to the high fecundity of Arctic grayling of average length (i.e., each female can have, on average, 2,300eggs; Kruse 1959, p. 327). Biologically, 57 entrained fry represent 1.4 percent of the total estimated fry production for a given year in the Big Hole River, which would be expected to be a very conservative estimate and biologically low effect (USFWS 2019, unpublished data). Most of the documented entrainment has occurred in several irrigation ditches, and does not appear to be widespread. No entrainment of Arctic grayling was documented in 2018, despite multiple surveys (MFWP 2019, unpublished data). We do note that sampling typically does not occur during the larval stage for Arctic grayling. Larval losses into irrigation ditches could be substantial and go undetected under the current sampling protocol. However, observations of young of year Arctic grayling in the Big Hole River indicate that many, but not likely all, newly emerged fry stay relatively close to the area where they were hatched (Skaar 1989, p. 51; Streu 1990, p. 28; McMichael 1990, p. 38), thus reducing the risk of entrainment because of minimal instream movements during their first summer.

Irrigation ditches are prioritized and systematically monitored based on the ditch location relative to known Arctic grayling distribution, additive maximum flow rate, and distance from the mainstem Big Hole River (MFWP *et al*. 2006, p. 116). In addition, electrofishing efficiency in simple habitats (such as irrigation ditches) is high (Kruse *et al*. 1998, pp. 942-943); thus, we have high confidence that these surveys have been accurate and that entrainment in the Big Hole River system is currently low.

Other populations: We have no evidence that entrainment is occurring in the Ruby River or Madison River. Entrainment was likely a historical threat for Arctic grayling at some locations within the Centennial Valley (Unthank 1989, p. 10; Gillin 2001, pp. 2-4, 3-18, 3-25), particularly outside of the Red Rock Lakes NWR (Boltz 2010, pers. comm.). Recent, repeat monitoring of multiple irrigation ditches on private lands in the Centennial Valley have not documented entrainment of Arctic grayling in 2016 and 2017 (Gander et al. 2019, p. 22). Currently, one irrigation ditch is present near the core of the Centennial Valley population within the Red Rock Lakes NWR. This ditch conveys water from Red Rock Creek to a waterfowl slough for a portion of the year; however, it is not operated by the Refuge when Arctic grayling fry are expected to be in Red Rock Creek (Bill West 2014a, pers. comm.). Other irrigation ditches are present upstream and downstream of the NWR boundary; however, Arctic grayling densities in these areas are low, and any mortality associated with entrainment in these areas is expected to be negligible at the population level. We also have no evidence that entrainment is occurring in any of the populations occupying high mountain lakes because we are not aware of any irrigation canals being fed directly by these lakes.

Entrainment of Arctic grayling in the Big Hole River is low and has not been documented in the Centennial Valley. We have no evidence that entrainment is occurring in the Ruby River or Madison River. Habitats occupied by the remaining 15 Arctic grayling populations occupying lakes in the upper Missouri River basin are not subjected to irrigation withdrawals because there are no known irrigation diversions present. We expect irrigation withdrawal volume to remain similar to current levels, particularly in the Big Hole River, in the future as more flow agreements are signed under the CCAA.

Sedimentation

Sedimentation has been proposed as a mechanism behind the decline of Arctic grayling and its habitat in the Centennial Valley (Unthank 1989, p. 10; Mogen 1996, p. 76), which includes Upper and Lower Red Rock Lakes. Historically, livestock grazing upstream likely led to accelerated sediment transport in tributary streams, and deposition of silt in both stream and lakes, thus modifying and reducing fish habitat by filling in pools, covering spawning gravels, and reducing water depth in Odell and Red Rock Creeks, where Arctic grayling spawn (MFWP 1981, p. 105; Mogen 1996, pp. 73–76). Sedimentation in the Upper and Lower Red Rock Lakes is believed to affect Arctic grayling in winter by reducing habitat volume and promoting hypoxia (low oxygen) in winter, which generally concentrates fish in specific locations, thus increasing the probability of competition and predation. In summer, reduced habitat volume could contribute to increased warming.

It has been reported that depths in the Red Rock Lakes have decreased significantly, with a decline in maximum depth from 7.6 to 5.0 m (25 to 16.4 ft) to less than 2 m (6.5 ft) noted in Upper Red Rock Lake over the past century (Mogen 1996, p. 76). This conclusion is prevalent among historical accounts of the Centennial Valley. However, a more recent analysis of sedimentation entering Upper Red Rock Lake indicated modest rates of sediment accumulation in Upper Red Rock Lake over the last century and that the rate of infilling in Upper Red Rock Lake has been relatively constant, based on lead and cesium analysis in lake bottom cores (Allison 1996, unpublished data). Thus, it appears historical accounts of rapid infilling of Upper Red Rock Lake were invalid, sedimentation in Upper Red Rock Lake is occurring at modest rates and likely not on a time scale that would be expected to influence habitat quality for Arctic grayling.

Sedimentation in tributary streams due to unregulated land use may have contributed to historical Arctic grayling declines in the Centennial Valley (Vincent 1962, p. 114). Now, land use is regulated, particularly on Federal land, which comprises the majority of ownership in the Centennial Valley. However, some of the tributary streams in the Centennial Valley on non-Federal lands are still affected by sediment. The effect of these levels of sediment on Arctic grayling in the Centennial Valley is currently unclear. However, currently the majority of Arctic grayling in the Centennial Valley do not reside within systems on non-Federal land, thus any potential effects of sedimentation in these streams on Arctic grayling would only be affecting a very small percent of the population.

The effects of erosion and sedimentation on spawning gravels in Red Rock Creek and reduction of habitat volume in Upper and Lower Red Rock Lakes are being mitigated because improved grazing practices appear to be reducing erosion rates upstream of Red

Rock Lakes NWR (USFWS 2009, pp. 75–76; Korb 2010, pers. comm.; West 2014; pers. comm.). Natural infilling of Upper Red Rock Lake is occurring (Allison 1996, unpublished data), but is occurring at a low rate.

Overwinter conditions (low oxygen)

We have no information indicating low dissolved oxygen from unfavorable winter conditions is affecting any population of Arctic grayling in the DPS, except for the Centennial Valley population. Recent results from the Centennial Valley Adaptive Management Plan indicate overwinter conditions (low dissolved oxygen) is the most supported driver of Arctic grayling dynamics in Upper Red Rock Lake in the Centennial Valley. In winter, deep snow can cover the ice on Upper Red Rock Lake and block sunlight that is used by aquatic plants to photosynthesize. The effect of blocked sunlight can reduce dissolved oxygen levels in the lake in two ways; (1) the aquatic plants die and no longer add oxygen to the water through photosynthesis, and (2) the decomposition process of the dead aquatic plants consumes dissolved oxygen from the water. The result of both these processes is less dissolved oxygen in the water that is available for Arctic grayling and other aquatic life.

Dissolved oxygen levels in Upper Red Rock Lake during winter can drop below levels typically considered lethal for Arctic grayling (Gangloff 1996, pp. 41–42, 72; Warren et al. 2019, p. 2). As a result, winter kill of invertebrates and fishes (e.g., suckers (*Catostomus* spp.)) has been recorded in Upper Red Rock Lake (Gangloff 1996, pp. 39–40). Recent modelling and analyses indicate that years of low abundance of spawning Arctic grayling in Red Rock Creek are typically preceded by low oxygen conditions during winter in Upper Red Rock Lake (Warren et al. 2019, pp. 2-3). However, we note that despite periods of low oxygen, there appears to be refugia areas of higher oxygen for Arctic grayling near inlet streams of Upper Red Rock Lake (Gangloff 1996, pp. 78-79; Cutting 2019, unpublished data; Davis et al. 2019, pp. 850-851) and in the inlet streams. Despite some winters where low oxygen conditions exist in the majority of Upper Red Rock Lake, some Arctic grayling are surviving, likely due to the presence and use of these refugia areas.

Climate Change

Our analyses under the Endangered Species Act include consideration of ongoing and projected changes in climate. The terms "climate" and "climate change" are defined by the Intergovernmental Panel on Climate Change (IPCC). The term "climate" refers to the mean and variability of different types of weather conditions over time, with 30 years being a typical period for such measurements, although shorter or longer periods also may be used (IPCC 2018, p. 4). The term "climate change" thus refers to a change in the mean or variability of one or more measures of climate (e.g., temperature or precipitation) that persists for an extended period, typically decades or longer, whether the change is due to natural variability, human activity, or both (IPCC 2018, p. 4).

Water temperature and hydrology (stream flow) are sensitive to climate change, and influence many of the basic physical and biological processes in aquatic systems. For ectothermic organisms like fish, temperature sets basic constraints on species' distribution and physiological performance, such as activity and growth (Coutant 1999, pp. 32–52). Stream hydrology not only affects the structure of aquatic systems across space and time, but influences the life history and phenology (timing of life-cycle events) of aquatic organisms such as fishes. For example, the timing of snowmelt runoff can be an environmental cue that triggers spawning migrations in salmonid fishes (Brenkman *et al.* 2001, pp. 981, 984), and the timing of floods relative to spawning and emergence can strongly affect population establishment and persistence (Fausch *et al.* 2001, pp. 1438, 1450). Significant trends in water temperature and stream flow have been observed in the western United States (Kaushal *et al.* 2010, entire; Isaak et al. 2012, entire; Null et al. 2013, entire), and climatic forcing (the energy difference between incoming solar radiation and outgoing radiation from Earth) caused by increased air temperatures and changes in precipitation are partially responsible.

Observations on flow timing in the Big Hole River, upper Madison River, and Red Rock Creek in the Centennial Valley indicate a tendency toward earlier snowmelt runoff (Wenger *et al*. 2011, entire; Towler *et al*. 2013, entire; De Haan *et al*. 2014, p. 41). These hydrologic alterations may be biologically significant for Arctic grayling in the Missouri River basin because they typically spawn prior to the peak of snowmelt runoff (Shepard and Oswald 1989, p. 7; Mogen 1996, pp. 22–23; Rens and Magee 2007, pp. 6–7). A trend toward earlier snowmelt runoff has resulted in earlier spawning in European grayling in Switzerland (Wedekind and Kung 2010, pp. 1419-1420). The effects of altered timing of spawn on Arctic grayling demographics are unknown. However, it has been hypothesized that the timing of fry emergence in salmonids is synchronized with when food resources are available (Crozier *et al*. 2008). Given that many ecological processes in aquatic environments are water temperature dependent (Durance and Ormerod 2007, entire; Brittain 2008, entire), it is likely that any alterations in timing of salmonid fry emergence would be synchronous with alterations in the timing of emergence and availability of prey species (Brittain 2008, entire).

Recent climate analyses in the Big Hole River Valley and Centennial Valley indicate rising air temperatures (1.8 – 3.2 °F (1.0 – 1.8 °C) / decade) from the 1980s to mid-2000s (De Haan *et al*. 2014, p. 29). During that time, the number of breeding Arctic grayling in the Big Hole River declined while the number of breeding Arctic grayling in the Centennial Valley increased (DeHaan *et al*. 2014, p. 17), despite a coherent climate signal between both drainages. Since about 2012, the number of breeding Arctic grayling has increased in the Big Hole River, on average, relative to 2007-2011 (Kovach et al. 2019, p. 8). The recent decline in number of spawning adults in Red Rock Creek is attributed to overwinter conditions, not warming air temperatures from climate change (Warren et al. 2019, entire). Thus, we have no information to conclude that increasing air temperatures have had a significant effect on number of breeding Arctic grayling in these systems in recent years.

The effect of warming water from increased air temperatures would be similar to that described for increased temperatures associated with stream dewatering (see discussion under Factor A), namely there has been an increased frequency of high water temperatures that have the potential to affect survival or optimal growth for Arctic grayling, which is considered a cold-water (stenothermic) species (Selong *et al.* 2001, p. 1032). However, the transfer of heat from air to water (i.e., convection) is a relatively small proportion of the energy exchange that occurs (Johnson 2003, p. 497). The more important factor influencing water temperature is likely to be solar radiation input (Johnson 2003, p. 497; Cassie 2006, p. 1393). Thus, the changes in ambient air temperature predicted to occur as the climate changes are not likely to have as large an effect on water temperatures as solar radiation. Changes in channel morphology (reducing width-to-depth ratios), increased instream flows and riparian vegetation (shading) resulting from the conservation actions being implemented for Arctic grayling have been shown to be correlated with reducing water temperatures by blocking some solar radiation and reducing surface area that solar radiation can interact with (McCullough 2019, entire). In tributaries to the Big Hole River, where riparian areas are improving, substantial reductions in water temperature have been observed (see Table 4, Rock Creek restoration for example and see Dewatering From Irrigation and Increased Water Temperatures; McCullough 2019, entire). We have empirical data showing the restoration of riparian areas, increased instream flows and concomitant channel morphology changes have occurred and are effective at mitigating the effects of climate change.

In the Centennial Valley, intact riparian areas are expected to minimize the effects of climate change, through similar processes as in the Big Hole River because tributary sizes are similar among the two watersheds and the past performance of riparian restoration of lowering water temperatures in the Big Hole River tributaries (McCullough 2019, entire). In addition, mean and maximum water temperatures are much lower in the Centennial Valley tributaries, on average, than those in the Big Hole River tributaries (MFWP 2019, unpublished data; USGS 2019, unpublished data), thus the potential for effects to Arctic grayling from rising water temperatures, even if only partially mitigated by riparian restoration, is low.

In the Ruby River, riparian areas along the Federal portion of the mainstem river and many of the tributaries are intact and most are functioning appropriately (Moran 2019, pers. comm.; USFS 2019, unpublished data). Although riparian areas along several tributaries to the Ruby River are classified as "functioning at risk"; the trends in riparian recovery are either static or improving (USFS 2019, unpublished data) and these tributaries are not known to be occupied by Arctic grayling.

The portion of the Madison River known to be occupied by Arctic grayling is much wider than would be expected to benefit from riparian shading. Similar to the mainstem Big Hole River, it is expected that increased flows in the mainstem river would have a larger effect on keeping water temperatures lower than riparian restoration. In 2017 and 2018, water temperatures reached the 21°C (70°F) stress threshold for Arctic grayling for several hours each summer (USGS 2019, unpublished data), but the overall time water temperatures were stressful to Arctic grayling was low.

Warming patterns in the western United States are not limited to streams. In California and Nevada, lake water surface temperatures have increased by an average of 0.11 ºC (0.2 ºF) per year since 1992, and at a rate twice that of the average minimum air surface temperature (Schneider *et al.* 2009, p. L22402). This suggests lake habitats are not immune to the predicted effects of climate change. Shallow lakes with a large surface area, such as Upper Red Rock Lake and Ennis Reservoir, would be expected to warm faster than deeper lakes. However, all 15 Arctic grayling populations in the upper Missouri River drainage occurring in lake habitats are expected to have thermal regimes well below upper thermal tolerances for Arctic grayling because of high elevation, bathymetry (underwater topography), and cool inputs from shaded inlet streams.

The land area of the upper Missouri River basin is predicted to warm through the end of the century (Ray *et al.* 2010, p. 23), although currently occupied Arctic grayling habitat tends to be in colder areas of moderate-to-high elevation. Most of the Arctic grayling populations are at approximately 1,775 to 2,125 m (5,860 to 9,000 ft) elevation (Peterson and Ardren 2009, p. 1761; MFISH 2014a, unpublished data). Alterations to instream flow and timing of runoff are already documented. However, Arctic grayling are likely to persist in the upper Missouri River drainage because of what appears to be an inherent ability of Arctic grayling to adjust spawn timing with changing water temperature regimes (Wedekind and Kung 2010, pp. 1419-1420). In addition, it has been demonstrated in the Big Hole River and Centennial Valley that Arctic grayling are capable of increasing in abundance and distribution, despite a warming climate (DeHaan *et al*. 2014, p. 17; Leary 2014, unpublished data; Kovach et al. 2019, pp. 10-11). It appears Arctic grayling within the upper Missouri River basin are responding favorably to increasing quality of habitat based on increasing abundance and distribution in systems with large-scale, ongoing habitat improvements (Big Hole River). Riparian restoration, particularly in the Big Hole River, has been empirically shown to minimize the effects of increasing water temperatures due to climate change (McCullough 2019, entire). Fifteen other populations are currently in habitats that will likely not be affected significantly by climate change due to their high elevation, intact riparian areas, and cool inputs of tributary water. Further, observed water temperature reductions following riparian restoration projects indicate that intact riparian areas can mitigate for many of the anticipated effects of climate change in the future.

Conservation Efforts to Reduce Habitat Destruction, Modification, or Curtailment of Its Range

Big Hole River CCAA

In 2006, a CCAA was developed for Arctic grayling in the Big Hole River. The conservation goal of this CCAA is to secure and enhance the population of Arctic grayling in the upper Big Hole River drainage. The CCAA Management Area encompasses about 382,000 acres and is divided into five management segments to make the conservation guidelines more spatially meaningful to property owners enrolled in the CCAA and to allow the involved agencies to track the progress of the conservation measures both temporally and spatially.

Site-specific plans are developed with each enrolled landowner; these plans identify conservation actions needed (or already completed) to meet the conservation goals of the CCAA. The conservation guidelines of the CCAA are met by implementing conservation measures that:

(1) Remove barriers to Arctic grayling migration;

(2) Improve streamflows;

(3) Identify and reduce or eliminate entrainment threats for Arctic grayling; and

(4) Improve and protect the function of riparian habitats.

Currently, 31 landowners have enrolled 158,000 acres (~52 percent total enrollable land) in the Big Hole CCAA Management Area and have signed (finalized) site-specific plans.

Restoration and conservation efforts outlined in site-specific plans are guided by the Big Hole Strategic Habitat Conservation Plan (hereafter, Strategic Habitat Conservation Plan), a science-based framework for making management decisions and prioritizing where and how to deliver conservation efficiently to achieve specific biological outcomes for Arctic grayling. The Strategic Habitat Conservation Plan delineates four spatial "tiers" that help prioritize where conservation will most benefit Arctic grayling:

(1) Tier I is 84 miles (23 percent) of core spawning, rearing and adult habitat that is currently occupied by Arctic grayling;

(2) Tier II is 91 miles (25 percent) of periphery habitat intermittently used by Arctic grayling;

(3) Tier III is 161 miles (43 percent) of suitable, but currently unoccupied, historical habitat; and

(4) Tier IV is 33 miles (9 percent) of potentially suitable habitat with unknown historical occupancy.

For reference, lands currently enrolled in the CCAA include 86 percent of tier I, 73 percent of tier II, 42 percent of tier III, and 24 percent of tier IV habitats. Given that the conservation measures outlined in the CCAA directly address known threats to Arctic grayling and their habitat in the Big Hole River, and that all conservation actions are strategically prioritized through the Strategic Habitat Conservation Plan, the Service is encouraged by the positive habitat and Arctic grayling response to the conservation actions in the Big Hole River.

Centennial Valley CCAA

In late 2018, a CCAA was finalized for Arctic grayling in the Centennial Valley. This CCAA identified the same potential threats to Arctic grayling in the Centennial Valley that were identified in the Big Hole CCAA: reduced streamflows, degraded riparian areas, fish barriers, and entrainment. Currently, three landowners have enrolled about 4,000 acres (1618 hectares) of land in the CCAA and work is ongoing to collect baseline data and begin to develop site-specific plans for the enrolled landowners. This represents about 5% of the total land eligible to be enrolled under the CCAA. Although we are aware of the future potential of this CCAA to contribute to Arctic grayling conservation, these anticipated future efforts do not contribute to Arctic grayling conservation currently, and as such, we have not considered them as part of this status review or our listing determination for this DPS.

Centennial Valley Adaptive Management Plan

The Centennial Valley Adaptive Management Plan identifies limiting factor(s) to Arctic grayling in the upper Centennial Valley. Three long-standing hypotheses that limit Arctic grayling (non-native hybrid cutthroat trout, spawning habitat, and overwinter habitat) are currently being tested to see the relative influence of each factor on abundance of spawning adult Arctic grayling in Red Rock Creek. Recent results indicate that overwinter conditions (low oxygen within Upper Red Rock Lake) is likely the primary factor negatively affecting abundance of spawning adult Arctic grayling in Red Rock Creek (Warren et al. 2019, p. 2). In response to this new information, Federal and state stakeholders recently initiated an alternatives analysis, the purpose of which was to identify management alternatives that would help alleviate low oxygen conditions during the winter in Upper Red Rock Lake. The top three alternatives that resulted from this analysis were mechanical aeration, tributary outflow diversion, and dredging. Currently, a solar aerator is being pilot-tested in Upper Red Rock Lake and oxygen sensors are deployed to characterize its effectiveness during the 2019/20 winter (Jaeger 2019, pers. comm.).

Private Landowner Conservation Efforts in the Big Hole Not Enrolled in the CCAA

Since 2006, twelve landowners in the Big Hole Valley who are not enrolled in the Big Hole CCAA have implemented voluntary conservation measures to benefit Arctic grayling. These conservation measures are similar to the conservation measures outlined in the site specific plans of landowners enrolled in the CCAA, including irrigation withdrawal reductions, installation of fish passage ladders, riparian fencing, stream restoration, and installation of stockwater tanks (MFWP 2014f, unpublished data). In addition, several of these landowners have informal flow agreements where the landowners have agreed to not utilize water returned to the stream by upstream enrollees in the Big Hole CCAA (MFWP 2014f, unpublished data). Although the majority of conservation projects in the Big Hole are completed through the CCAA, the Service is very encouraged by the participation of non-enrolled landowners to further grayling conservation in the Big Hole River and its tributaries.

Big Hole River Drought Management Plan

The purpose of the Drought Management Plan is to mitigate the effects of low stream flows and lethal water temperatures for fisheries through a voluntary effort among participants including agriculture, municipalities, business, conservation groups, anglers, and affected government agencies (Big Hole Watershed Committee 2014, p.1). The Drought Management Plan outlines flow triggers that, when met, initiate specific voluntary actions to conserve water. The flow triggers in the Drought Management Plan are the same as the flow targets outlined in the Big Hole CCAA. The Drought Management Plan has been in effect since 1999.

One key difference between the Drought Management Plan and the CCAA is that the Drought Management Plan is in effect for the entire Big Hole River, not just the upper Big Hole River, like the CCAA. Arctic grayling occur outside of the CCAA Management Area; thus, any conservation efforts occurring in these areas still likely benefit Arctic grayling, although Arctic grayling densities outside the CCAA Management Area are low and represent a small fraction of the total population inside the CCAA Management Area (MFWP 2013d, unpublished data). Another key difference is that the Drought Management Plan is structured to disseminate flow and water temperature information to all users of the Big Hole River, not just private landowners in the CCAA. This structuring allows for near real-time information sharing that helps inform users when voluntary conservation actions are needed. Such actions include reductions in irrigation withdrawal (for downstream users not in the CCAA); reductions in municipal, industrial, and personal water use; and reductions in recreation (e.g., angling).

The extent and magnitude of beneficial effects to Arctic grayling from the voluntary conservation measures recommended in the Drought Management Plan are unclear. However, the Drought Management Plan appears to have broad-based support. Most participants reduce irrigation withdrawals in response to observed low flows on nearby USGS gauges, before phone calls are made to request irrigation reductions (Downing 2014, pers. comm.). Increases in instream flow attributable to efforts under the Drought Management Plan have been observed as "bumps" in the hydrograph in the middle and lower reaches of the Big Hole River (Downing 2014, pers. comm.). Although difficult to quantify, these "bumps" typically result in instream flows rising above low flow triggers (Downing 2014, pers. comm.). In addition, the inherent value of information sharing among diverse stakeholder groups about the potential effects of dewatering and thermal stress on the Big Hole fishery is likely significant. An increased understanding of conservation efforts needed to benefit Arctic grayling, and aquatic habitat in general, has been demonstrated to be a necessary precursor for more formalized conservation actions, such as the creation and implementation of the Big Hole CCAA.

Native Arctic Grayling Genetic Reserves and Translocation

Given concern over the status of Arctic grayling, the Montana Arctic Grayling Recovery Program was formed in 1987, to address conservation concerns for primarily the Big Hole River population, and to a lesser extent the population in the Centennial Valley (Memorandum of Understanding (MOU 2007, p. 2). The Arctic Grayling Workgroup was established as an ad hoc technical workgroup of the Arctic Grayling Recovery Program, composed of Federal and state officials tasked with Arctic grayling conservation. In 1995, the Arctic Grayling Workgroup finalized a restoration plan that outlined an agenda of restoration tasks and research, including management actions to secure the Big Hole River population, brood stock development, and a program to re-establish four additional populations (Arctic Grayling Workgroup 1995, pp. 7–17). Due to the large amount of new information that has become available since 1995, the Arctic Grayling Workgroup is currently updating the Montana Arctic Grayling Restoration Plan.

Consequently, Montana Fish, Wildlife and Parks established genetic reserves of Big Hole River and Centennial Valley Arctic grayling (Leary 1991, entire). Currently, brood (genetic) reserves of Big Hole River Arctic grayling are held in two closed-basin lakes in south-central Montana (Rens and Magee 2007, p. 22). These fish are manually spawned to provide gametes for translocation efforts in Montana (e.g., Ruby River population) (Rens and Magee 2007, p. 22). Two brood reserves of Centennial Valley Arctic grayling have recently been established in Elk Lake and Handkerchief Lake; Instream flows in the sole spawning tributary (Narrows Creek) to Elk Lake have been low in recent years, likely as a result of low snowpack in some years and seismic activity that altered the hydrology of Narrows Creek (Jaeger 2014c, pers. comm.), resulting in no documented natural reproduction to date. Future conservation actions on Narrows Creek include securing a more consistent water supply during the Arctic grayling spawning season through a water rights exchange; however, at this time, these conservation actions and the future viability of the Elk Lake population are too uncertain to warrant consideration in this finding. Spawning has not been documented in Handkerchief Lake due to the brood reserve being recently introduced, but Handkerchief Lake previously had a grayling population (with non-native genetics that were removed from the lake) that spawned naturally in inlet streams.

A reintroduction effort in the upper Ruby River, where Arctic grayling were previously extirpated, using Big Hole River genetic reserves concluded in 2008. Arctic grayling eggs from the Big Hole River reserves were hatched on-site in incubators, and fry were allowed to drift into the reintroduction area. Since 2008, natural reproduction has been documented in the upper Ruby River for 10 consecutive years (Gander et al. 2019, pp. 4-6). Recent genetic analyses of the Ruby River population indicate high levels of genetic diversity (heterozygosity and allelic richness), albeit low estimate of effective number of breeders (Gander et al. 2019, pp. 4-6). Despite low numbers of breeding adults currently, genetic diversity of the Ruby River population remains high and stable.

Another recent conservation effort using Big Hole River genetic reserves involves an assisted recolonization effort of Arctic grayling in Rock Creek, a historically occupied tributary of the upper Big Hole River. From 2010-2014, incubators were used to reintroduce young-of-year Arctic grayling into Rock Creek. Encouragingly, young-of-year and older Arctic grayling have been documented in 4 miles of Rock Creek following reintroduction. This increase of Arctic grayling abundance and distribution in Rock Creek is likely due, at least in part, to the introduction of thousands of fry via the onsite incubators. Habitat improvement projects on Rock Creek have occurred simultaneously with fry reintroduction, so it is difficult to distinguish the relative effects of fry reintroduction and habitat improvement on the resulting increase in distribution and abundance of young Arctic grayling. Likely, both factors have played a role in reestablishing Arctic grayling in Rock Creek. Regardless, both conservation actions are having their intended effect: increasing Arctic grayling abundance and distribution in historically occupied habitat.

In 2013, a marked increase in the number of breeding Arctic grayling was observed in the Big Hole River (Leary 2014, unpublished data; Kovach et al. 2019, p. 26). Given that fry were being reintroduced into the Big Hole River (and Rock Creek) beginning in 2010, there was initial uncertainty about the relative contribution of fish produced by remote site incubators to the observed increase in breeding adults. Genetic analysis of a sample of young Arctic grayling obtained in 2013 indicated a low level of relatedness (<10 percent of sample were half- or full siblings) among individuals within the sample (Leary 2014, unpublished data). These results indicate that in 2010, fish produced by remote site incubators contributed very little to the increase in breeding adults in 2013, as we would have expected a high degree of relatedness within the 2013 sample due to a small number of grayling spawned to produce eggs for the remote site incubator reintroduction effort. Thus, these data suggest that factors other than the influence of remote site incubators were responsible for increasing abundance of adult spawners in the Big Hole River in 2013.

Similar reintroductions to the Rock Creek effort are also underway in several other tributaries and lakes within the upper Big Hole drainage and elsewhere, including the Wise River, Trail Creek, Twin Lakes and the Madison River. This suite of reintroductions is ongoing and monitoring has been initiated to document any changes in distribution of Arctic grayling..

In the Centennial Valley, remote site incubators have been used fairly extensively to try to establish spawning runs of adult Arctic grayling in multiple tributaries to Upper

Red Rock Lake (Boltz and Kaeding 2002, entire; Jaeger 2014d, pers. comm.). In 2017, Elk Springs Creek was reconnected to Upper Red Rock Lake and remote site incubators were used to introduce young Arctic grayling. Encouragingly, several sub-adult Arctic grayling have been observed in Elk Springs Creek, indicating Arctic grayling are now using Elk Springs Creek. The effect of Arctic grayling fry produced by remote site incubators on establishment of Arctic grayling in Elk Springs Creek is unknown, but the Service hopes that using remote site incubators as a conservation tool will result in fish produced by remote site incubators recruiting to the Centennial Valley population.

Another Arctic grayling reintroduction project has been completed in Grayling Creek within Yellowstone National Park in Wyoming. Arctic grayling were reintroduced to approximately 30 miles of historically occupied stream habitat. Current monitoring indicates grayling are present in Grayling Creek, although natural reproduction has not been documented yet (Puchany 2019, pers. comm.). Thus, although we are aware of the future potential of this project to contribute to Arctic grayling conservation, we do not consider this project to contribute to Arctic grayling conservation currently, and have not considered it as part of this status review or our listing determination for this DPS.

Multiple other Arctic grayling introductions have occurred recently in Yellowstone National Park. Arctic grayling have been introduced into Ice, Wolf and Goose lakes and the upper Gibbon River (Koel et al. 2019, pp. 7-8). Natural reproduction has not been documented in these populations, due to the Arctic grayling not being sexually mature yet. Thus, although we are aware of the future potential of these reintroductions to contribute to Arctic grayling conservation, they do not contribute to Arctic grayling conservation currently, and as such, are not considered as part of this status review or our listing determination for this DPS.

Summary of Factor A

Based on the best available information, we find that the historical range of the Missouri River DPS of Arctic grayling has been reduced particularly by large-scale habitat fragmentation by dams. However, despite fragmentation, sufficient habitat remains intact and is currently supporting multiple, viable, Arctic grayling populations representing the full range of known life histories. Historical threats to habitat quantity and quality in the Big Hole River are systematically being eliminated or minimized by the CCAA and Strategic Habitat Conservation Plan through conservation projects designed to directly address the four conservation criteria outlined in the CCAA. Large-scale habitat improvements are occurring; quality of riparian areas has improved in both the Big Hole River and Centennial Valley through riparian restoration projects, and these projects have been shown to minimize effects of climate change through blocking of some solar radiation and channel morphology changes. In addition, Arctic grayling populations are responding favorably to habitat improvements in the Big Hole River. The Centennial Valley population is currently below the management goal for the population, but is stable and poor overwinter conditions have now been identified as the likely mechanism influencing Arctic grayling dynamics in the Centennial Valley. In the future, we expect habitat to remain suitable in the Big Hole River because of the proven past performance of CCAA projects. These protections are expected to persist into the future and maintain the integrity of the habitat. Most of the other populations of Arctic grayling reside in high-quality habitats on Federal land.

B. Overutilization for Commercial, Recreational, Scientific, or Educational Purposes

Arctic grayling of the upper Missouri River are handled for recreational angling and for scientific/population monitoring, and restoration purposes.

Recreational Angling

Arctic grayling are highly susceptible to capture by angling (ASRD 2005, pp. 19–20), and intense angling pressure can reduce densities and influence the demography of exploited populations (Northcote 1995, pp. 171–172). Historically, overfishing likely contributed to the rangewide decline of Arctic grayling in the upper Missouri River system (Vincent 1962, pp. 49–52, 55; Kaya 1992, pp. 54–55). In 1994, concern over the effects of angling on Arctic grayling led the State of Montana to implement catch-and-release regulations for Arctic grayling captured in streams and rivers within its native range, and those regulations remain in effect today (MFWP 2019, p. 57). Catch-and-release regulations also are in effect for Ennis Reservoir on the Madison River (MFWP 2019, p. 65). Angling is not permitted in either of the Red Rock Lakes in the Centennial Valley (USFWS 2009, p. 147), and catch-and-release regulations remain in effect for any Arctic grayling captured in streams (e.g., Odell Creek or Red Rock Creek) in the Centennial Valley (MFWP 2019, p. 57). Additionally, angling is closed in Red Rock Creek during the Arctic grayling spawning period (May 15 to June 14; MFWP 20119, p. 73).

In all other populations, anglers can keep up to 5 Arctic grayling per day and have up to 10 in possession, in accordance with standard daily and possession limits for that angling management district (MFWP 2019, p. 57). The population trends of Arctic grayling in many of the lakes (see Table 3, above) suggest that present angling exploitation rates are low, even though harvest is allowed on most of these populations.

Repeated catch-and-release angling may harm individual fish, causing physiological stress and injury (i.e., hooking wounds). Catch-and-release angling also can result in mortality at a rate dependent on hooking location, hooking duration, fish size, water quality, and water temperature (Faragher *et al.* 2004, entire; Bartholomew and Bohnsack 2005, p. 140; Boyd *et al*. 2010, entire). Repeated hooking (up to five times) of Arctic grayling in Alaska did not result in significant additional mortality (rates 0 to 1.4 percent; Clark 1991, pp. 1, 25–26). In Michigan, hooking mortality of Arctic grayling in lakes averaged 1.7 percent per capture event, based on 355 individuals captured with artificial flies and lures (Nuhfer 1992, pp. 11, 29). Higher mortality rates (5 percent) have been reported for Arctic grayling populations in the Great Slave Lake area, Canada (Falk and Gillman 1975, cited in Casselman 2005, p. 23). Comparatively high catch rates for Arctic grayling have been observed in the Big Hole River, Montana (Byorth 1993, pp. 26–27, 36), and average hooking wound rates ranged from 15 to 30 percent among study sections (Byorth 1993, p. 28). However, overall hooking mortality from single capture events was low (1.4 percent), which led Byorth to conclude that the Big Hole River population was not limited by angling (Byorth 1994b, entire).

Compared to the average catch-and-release mortality rates of 4.2 to 4.5 percent in salmonids as reported by Schill and Scarpella (1997, p. 873), and the mean and median catch-and-release mortality rates of 18 percent and 11 percent from a meta-analysis of 274 studies (Bartholomew and Bohnsack 2005, pp. 136–137), the catch-and-release mortality rates for Arctic grayling are comparatively low (Clark 1991, pp. 1, 25–26; Nuhfer 1992, pp. 11, 29; Byorth 1994b, entire). We are uncertain whether these lower observed rates reflect an innate resistance to effects of catch-and-release angling in Arctic grayling or whether they reflect differences among particular populations or study designs used to estimate mortality. Even if catch-and-release angling mortality is low (e.g., 1.4 percent as reported in Byorth 1994b, entire), the high catchability of Arctic grayling (ASRD 2005, pp. 19–20) raises some concern about the cumulative mortality of repeated catch-and-release captures. For example, based on the Arctic grayling catch rates and angler pressure reported by Byorth (1993, pp. 25–26) and the population estimate for the Big Hole River reported in Byorth (1994a, p. ii), a simple calculation suggests that age 1 and older Arctic grayling susceptible to recreational angling may be captured and released 3 to 6 times per year.

In conclusion, angling harvest may have significantly reduced the abundance and distribution of the Upper Missouri River DPS of Arctic grayling during the past 50 to 100 years, but current catch-and-release fishing regulations (or angling closures) in most waters occupied by extant populations have likely ameliorated the past threat of overharvest. Although we do note the potential for cumulative mortality caused by repeated catch-and-release of individual Arctic grayling in the Big Hole River, we have no evidence indicating that repeated capture of Arctic grayling under catch-and-release regulations is currently limiting that population or the DPS. Moreover, fishing is restricted in the Big Hole River, an important recreational fishing destination in southwestern Montana, when streamflow and temperature conditions are likely to increase stress to captured Arctic grayling. Anglers can still capture and keep Arctic grayling in most lake populations in accordance with State fishing regulations, but we have no evidence that current levels of angling are affecting these populations. We have no information at this time to indicate that future fishing regulations are likely to change in a way that would be detrimental to Arctic grayling.

Scientific/Population Monitoring

Montana Fish, Wildlife and Parks consistently monitors the Big Hole River Arctic grayling populations (MFWP 2019, unpublished data). Electrofishing (use of electrical current to temporarily and non-lethally immobilize a fish for capture) is a primary sampling method to monitor Arctic grayling in these populations (Rens and Magee 2007, pp. 13, 17, 20). A number of studies have investigated the effects of electrofishing on various life stages of Arctic grayling. Dwyer and White (1997, p. 174) found that electrofishing reduced the growth of juvenile Arctic grayling and concluded that long-term, sublethal effects of electrofishing were possible. Hughes (1998, pp. 1072, 1074–1075) found evidence that electrofishing and tagging affected the growth rate and movement behavior of Arctic grayling in the Chena River, Alaska. Roach (1999, p. 923) studied the effects of electrofishing on fertilized Arctic grayling eggs and found that while electrofishing could result in egg mortality, the population-level effects of such mortality were not likely to be significant. Lamothe and Magee (2003, pp. 16, 18–19) noted mortality of Arctic grayling in the Big Hole River during a radio-telemetry study, and concluded that handling stress or predation were possible causes of mortality.

However, population monitoring activities in the Big Hole River are curtailed when environmental conditions become unsuitable (Big Hole Watershed Committee 1997, entire), and recent monitoring reports (Cayer and McCullough 2012, 2013, entire) provide no evidence that electrofishing is harming the Arctic grayling population in the Big Hole River.

Traps, electrofishing, and radio telemetry have been used to monitor and study Arctic grayling in the Centennial Valley (Gangloff 1996, pp. 13–14; Mogen 1996, pp. 10–13, 15; Kaeding and Boltz 1999, p. 4; Rens and Magee 2007, p. 17) and Ruby River (Gander et al. 2019, entire); however, there are no data to indicate these monitoring activities reduce the growth and survival of individual Arctic grayling (Gander et al. 2019, entire).

The Arctic grayling population in the Madison River–Ennis Reservoir is monitored sporadically (Rens and Magee 2007, pp. 20–21). When electrofishing surveys targeting Arctic grayling in the Madison River occur, they are conducted during the spawning run for that population (Clancey 1996, p. 6). Capture and handling during spawning migrations or during actual spawning could affect the reproductive success of individual Arctic grayling. However, under recent monitoring frequencies, any population-level effect of these activities is likely negligible, and we have no data to indicate these monitoring activities reduce the reproductive success of individual Arctic grayling.

Most of the populations of Arctic grayling occupying high mountain lakes are infrequently monitored (MFISH 2014a, unpublished data). Many of these surveys are qualitative surveys of spawner abundance and are not expected to have any effects on the

Arctic grayling populations because no capture of individuals is involved, only observations. Some handling of Arctic grayling does occur when genetic samples are taken; however these are typically non-lethal and any handling effects from this are expected to be negligible, given the non-invasive procedures used.

The intensity and type of monitoring and scientific investigation varies among the different populations in the DPS, but we have no evidence suggesting that monitoring or scientific study is having population level effects on Arctic grayling in the Missouri River basin. We expect similar levels of population monitoring and scientific study in the future.

Restoration Efforts

Attempts to re-establish native populations of Arctic grayling may result in the mortality of some embryos and young fish. Currently, gametes (eggs and sperm) used to re-establish Arctic grayling in suitable habitat within the Upper Missouri River basin come from captive brood reserves of Big Hole River Arctic grayling maintained in Axolotl and Green Hollow II Lakes (Rens and Magee 2007, pp. 22–24). Removal of gametes from the wild Big Hole River population was necessary to establish this brood reserve (Leary 1991, entire) and will likely continue intermittently in the future to ensure the genetic representation of the brood reserve. The previous removal of gametes for conservation purposes could hypothetically reduce the abundance of the wild population if the population was unable to compensate for this effective mortality by increased survival of remaining individuals. However, in this case, the Big Hole River population is genetically diverse with an increasing number of effective breeders and fairly large effective population size (Kovach et al. 2019, entire), thus it does not appear the previous removal of gametes has affected the population in any measurable way. In addition, the establishment of a brood reserve provides a conservation benefit from the standpoint that gametes from the reserve can be harvested to use for translocation efforts to benefit the species. Ultimately, we conclude that past gamete collection from the Big Hole River population has not harmed the wild population or that collection in the future will harm the population.

Efforts to re-establish native, genetically representative populations of Arctic grayling in the Centennial Valley and to maintain a brood reserve for that population have resulted in the direct collection of eggs from Arctic grayling spawning runs in Red Rock Creek. During 2000–2002, an estimated 315,000 Arctic grayling eggs were collected from females captured in Red Rock Creek (Boltz and Kaeding 2002, pp. v, 8). The Service placed over 180,000 of these eggs in remote site incubators in streams within the Red Rock Lakes NWR that historically supported Arctic grayling spawning runs (Boltz and Kaeding 2002, pp. v, 10). However, recent declines in abundance of Arctic grayling in the spawning run in Red Rock Creek are attributable to harsh overwinter conditions in Upper Red Rock Lake (Warren et al. 2019, p. 2) , and are likely not associated with prior gamete removal from the population.

Montana Fish, Wildlife and Parks and the Service are currently collaborating on an effort to re-establish an Arctic grayling spawning run in Elk Springs Creek and a replicate of the Centennial population in Handkerchief Lake (West 2014a, pers. comm., Gander et al. 2019, pp. 6-10). These actions required the collection of gametes (approximately 370,000 eggs) from Arctic grayling captured in Red Rock Creek (Jaeger

2014f, pers. comm.). Approximately 10 percent of these eggs were returned to Red Rock Creek and incubated in that stream (using a method resulting in high survivorship of embryos) (Kaeding and Boltz 2004, entire) to mitigate for collection of gametes from the wild spawning population. We infer that past gamete collection in Red Rock Creek has not significantly influenced recruitment in Red Rock Creek, as abundance of returning spawners to Red Rock Creek has been found to be primarily influenced by harsh overwinter conditions in Upper Red Rock Lake (Warren et al. 2019, p. 2).

Overall, we conclude that collection of gametes from the wild populations in the Big Hole River and Centennial Valley systems has not contributed to population-level declines in those populations, or that the previous collections represent overexploitation. Future plans to collect gametes from Arctic grayling should be evaluated in light of the status of those populations at the anticipated time of the collections. We encourage the agencies involved to coordinate their efforts and develop a strategy for broodstock development and conservation efforts that minimizes any potential impacts to wild native populations. However, at present, we do not have any data indicating collection of gametes for conservation purposes has been detrimental to the Big Hole River and Centennial Valley populations. We have no evidence to indicate that gamete collection will increase in the future.

Conservation Efforts to Reduce Recreational Overutilization

The MFWP closes recreational angling in specific reaches of the Big Hole River when environmental conditions are considered stressful. Specific streamflow and temperature thresholds initiate mandatory closure of the fishery (Big Hole Watershed

Committee 1997, entire). Such closures have been implemented as recently as 2019. These closures are expected to be effective at reducing mortality of angled Arctic grayling, given that similar species (mountain whitefish, rainbow trout, brown trout) do incur some post-release mortality after being angled during periods of high water temperature (Boyd et al. 2010, entire).

Summary of Factor B

Based on the best information available, we conclude that overexploitation by angling may have contributed to the historical decline of the Upper Missouri River DPS of Arctic grayling, but we have no evidence to indicate that current or future levels of recreational angling, population monitoring, scientific study, or conservation actions constitute overexploitation. We expect similar or decreased levels of these activities to continue in the future.

Factor C. Disease or Predation

Disease

Arctic grayling are resistant to whirling disease, which is responsible for population-level declines of other stream salmonids (Hedrick *et al.* 1999, pp. 330, 333). However, Arctic grayling are susceptible to bacterial kidney disease, a bacterial disease causing reduced immune response and mortality in some fish species (Meyers *et al*. 1993, p. 181). Some wild populations in pristine habitats test positive for bacterial kidney disease (Meyers *et al.* 1993, pp. 186–187), but clinical effects of the disease are more likely to be evident in captive populations (Meyers *et al.* 1993, entire; Peterson 1997, entire). To preclude transmission of bacterial kidney disease between Arctic grayling during brood reserve, hatchery, and wild Arctic grayling translocation efforts, MFWP tests kidney tissue and ovarian fluid for the causative agent for bacterial kidney disease as well as other pathogens in brood populations (Rens and Magee 2007, pp. 22–24).

Since 2010, testing for seven pathogens (3 bacterial, causing furunculosis, enteric redmouth, and bacterial kidney disease; one parasite, causing whirling disease; and 3 viruses, causing infectious pancreatic necrosis, infectious hematopoietic, and viral hemorrhagic septicemia) has been conducted on over 6,000 Arctic grayling in the upper Missouri River basin (MFWP 2019, unpublished data). No Arctic grayling have tested positive for any of the pathogens that were tested for (MFWP 2019, unpublished data). Most of the pathogen testing has been conducted on Arctic grayling from the multiple brood reserves, because clinical effects of the disease are more likely to be evident in captive populations (Meyers *et al.* 1993, entire; Peterson 1997, entire). However, some testing of wild fish has also occurred (MFWP 2019, unpublished data). Consequently, the best available evidence at this time does not indicate that disease threatens native Arctic grayling of the upper Missouri River.

Predation By and Competition With Nonnative Trout

Brook trout (*Salvelinus fontinalis*), brown trout (*Salmo trutta*), and rainbow trout are widely distributed and abundant in the western United States, including the upper Missouri River system (Schade and Bonar 2005, p. 1386; Table 8). One or more of these nonnative trout species co-occur with 10 of the 19 Arctic grayling population in the basin. The remaining nine Arctic grayling populations occur with other native species or no other fish species (Table 8).

Table 8. Nonnative species composition for each Arctic grayling population within the upper Missouri River DPS. Rows highlighted in gray indicate populations occurring in native habitat. All data presented in this table are from MFISH 2019, unpublished data.

Population	Nonnative species present*
Big Hole River (+11 tributaries)	Rainbow, Brook, Brown
Ennis Reservoir / Madison River	Rainbow, Brown, Utah chub
Centennial Valley	Brook, Yellowstone cutthroat hybrid
Mussigbrod Lake	Brook
Miner Lake	Rainbow, Brook
Ruby River	Rainbow hybrid, Brook, Brown
Agnes Lake	None
Odell Lake	None
Bobcat Lake	None
Schwinegar Lake	None
Pintlar Lake	Rainbow, Brook
Deer Lake	None
Emerald Lake	None
Grayling Lake	None
Hyalite Lake	Brook, Yellowstone cutthroat
Gibson Reservoir	Rainbow, Brook
Lake Levale	None
Park Lake	Yellowstone cutthroat
Grebe Lake	None

*All listed nonnative fish species are trout species, unless otherwise noted.

Ecological interactions (predation and competition) with the brook trout, brown trout, and rainbow trout are among the long-standing hypotheses to explain the historical decline of Arctic grayling in the upper Missouri River system and the extirpation of some populations from specific waters (Nelson 1954, p. 327; Vincent 1962, pp. 81–96; Kaya 1992, pp. 55–56). Strength of competition and predation can be very difficult to measure in wild trout populations (Fausch 1988, pp. 2238, 2243; 1998, pp. 220, 227). Predation on Arctic grayling eggs and fry by brook trout has been observed in both the Big Hole River and the Centennial Valley (Nelson 1954, entire; Streu 1990, p. 17; Katzman 1998, pp. 35, 47, 114), but such observations have not been definitively linked to population declines of Arctic grayling. To our knowledge, no studies have investigated or attempted to measure predation by brown trout or rainbow trout on Arctic grayling in Montana. Brook trout do not appear to negatively affect habitat use or growth of juvenile, hatchery-reared Arctic grayling (Byorth and Magee 1998, p. 921), but further studies are necessary to determine whether competition or predation occur at other life stages or with brown or rainbow trout (Byorth and Magee 1998, p. 929). Predation represents direct mortality that can limit populations, and young-of-year Arctic grayling may be particularly susceptible to predation by other fishes because they are smaller and weaker swimmers than trout fry (Kaya 1990, pp. 52–53).

The evidence for predation and competition by nonnative trout on Arctic grayling in the upper Missouri River basin is largely circumstantial or correlational, and inferred from the reduced historical abundance and distribution of Arctic grayling following encroachment by nonnative trout (Kaya 1990, pp. 52–54; Kaya 1992, p. 56; Magee and Byorth 1995, p. 54; McCullough 2017, pp. 16, 22). In addition, the historical difficulty in establishing Arctic grayling populations in waters already occupied by nonnative trout, especially brown trout (Kaya 2000, pp. 14–15) may suggest competition and predation play a role. However, the often-cited case histories where nonnative trout were implicated in the decline of Arctic grayling also involved prior or concurrent habitat modification or degradation, thus confounding the two factors (Kaya 1990, pp. 52–54; Kaya 1992, p. 56; Magee and Byorth 1995, p. 54) and making it difficult to pinpoint the cause of the decline. Where past habitat degradation has not been a factor (e.g., many of the high-elevation lake populations), successful coexistence between brook trout and rainbow trout and Arctic grayling has occurred over long durations, greater than 100 years in some populations (Jaeger 2014, unpublished data; MFISH 2014a, unpublished data). Despite past habitat degradation in the Big Hole River, Arctic grayling have coexisted with brook, rainbow and brown trout for at least 60 years (Liknes 1981, p. 34).

In the Big Hole River, brook trout, rainbow trout, and brown trout are more abundant than Arctic grayling (Rens and Magee 2007, p. 42). In general, brook trout is the most abundant nonnative trout species in the Big Hole River upstream from Wisdom, Montana (Rens and Magee 2007, pp. 7, 42; Lamothe *et al.* 2007, pp. 35–38). Recent analyses indicated that streams with higher abundance (expressed as catch per unit effort) of brook trout also had higher abundance of age 1+ Arctic grayling (McCullough 2017, pp. 16, 22), indicating brook trout are likely not affecting abundance of older (age-1+) Arctic grayling. Rainbow trout and brown trout are comparatively more abundant in the downstream reaches (Kaya 1992, p. 56; Oswald 2005, pp. 22–29; Lamothe *et al.* 2007, pp. 35–38; Rens and Magee 2007, p. 10) and streams with higher abundance (expressed as catch per unit effort) of rainbow and brown trout had lower abundance of young (age-0) Arctic grayling (McCullough 2017, pp. 16, 22), indicating rainbow and brown trout may be affecting abundance of young (age-0) Arctic grayling. Recently, brown trout abundance has increased in the upper Big Hole River upstream of Wisdom (MFWP 2013e, unpublished data). In the reach of the upper Big Hole River where Arctic grayling densities are highest, nonnative trout abundances are lower than upstream or downstream reaches, and appear to have been stable since at least 2006 (Cayer 2013, unpublished data).

The potential effects of nonnative trout species (rainbow, brown, brook, and Yellowstone cutthroat trout) on Arctic grayling recruitment are largely unknown. Arctic grayling experts from Montana convened to explore such effects predicted a less than 12 percent reduction in Arctic grayling recruitment when nonnative trout densities for any species were 500 fish/mile or fewer, on average (Service 2014, p. 2). Predicted reduction in Arctic grayling recruitment when any of the nonnative species were present at 1,000 fish/mile was higher and similar among species (20 to 25 percent; Service 2014, p. 2). These estimates were derived with the assumption that habitat was not a limiting factor.

Currently, densities of nonnative trout (brook, brown, rainbow) are fewer than 20 fish/mile (per species) in the mainstem Big Hole River where Arctic grayling densities are highest (Cayer 2013, unpublished data). Densities of brown and rainbow trout are fewer than 20 fish/mile in Big Hole River tributaries, while brook trout density in tributaries is higher (~80 fish/mile). Brook trout density estimates only include fish greater than 10 inches, thus it is unknown how many total brook trout reside in these areas. At current densities of rainbow and brown trout, effects on Arctic grayling recruitment would be expected to be small, based on the predictions of recruitment reduction from nonnatives from the expert meeting.

In the Madison River in and near Ennis Reservoir, brown trout and rainbow trout are abundant and are the foundation of an important recreational fishery (e.g., Byorth and Shepard 1990, p. 1). Nonnative rainbow trout and brown trout densities in the Madison River near Ennis Reservoir are about 3,500 to 4,000 fish per mile (both species included). These densities are substantially higher than those observed in other systems occupied by Arctic grayling, and we did not ask Arctic grayling experts to predict effects on Arctic grayling recruitment at these higher densities. Arctic grayling abundance in the Ennis Reservoir/Madison River population appears to be suppressed and declining (MFWP 2013f, unpublished data). The relationship between the higher densities of nonnative trout and the low and declining abundance of Arctic grayling in this population is unclear. However, the densities of nonnative trout observed in the Madison River are not representative of densities of nonnatives in any of the other 18 populations of Arctic grayling in the DPS. Thus, the effect of nonnatives on Arctic grayling recruitment is a concern in the Madison River.

In the Centennial Valley, brook trout and hybrid cutthroat trout (Yellowstone cutthroat trout (*Oncorhynchus clarkii bouvieri*) crossed with rainbow trout; Mogen 1996, p. 42) have a well-established population and dominate the abundance and biomass of the salmonid community (Katzman 1998, pp. 2–3; Boltz 2010, pers. comm.). In Upper Red Rock Lake, diet overlap among hybrid cutthroat trout, brook trout and Arctic grayling was documented (Cutting et al. 2016, p. 7), although food may not be a limiting factor in this system, given the eutrophic, highly productive nature of Upper Red Rock Lake (Cutting et al. 2016, p. 15; Jaeger 2014g, pers. comm.). In addition, hybrid Yellowstone cutthroat trout in Upper Red Rock Lake may occupy a similar ecological niche once occupied by native westslope cutthroat trout, a species with which Arctic grayling co-evolved. Thus, the adaptations Arctic grayling developed over thousands of years to coexist with westslope cutthroat trout may be equally advantageous when coexisting with hybrid Yellowstone cutthroat trout.

Predation of Arctic grayling by brook trout and hybrid cutthroat trout occurs in Upper Red Rock Lake (Nelson 1954, entire; Katzman 1998, pp. 35, 47, 114). In 2013, the Service and MFWP initiated a removal effort to suppress hybrid cutthroat trout in Red Rock Creek and Upper Red

Rock Lake as part of the Centennial Valley Adaptive Management Plan. This effort occurred for 5 years; however, the models developed as part of the Adaptive Management Plan indicate that the hybrid cutthroat trout are likely not a primary driver of Arctic grayling dynamics in Upper Red Rock Lake (Warren et al. 2019, p. 2). It is plausible that extensive macrophyte beds present in Upper Red Rock Lake (Katzman1998, p. 81) provide complex hiding and rearing cover for juvenile Arctic grayling and minimize interactions between young Arctic grayling and nonnative fishes (Almany 2004, entire).

In the upper Missouri River basin, it appears that the extent and magnitude of competition and predation between nonnative trout and Arctic grayling likely depends on environmental context (e.g., habitat type and quality, environmental conditions such as temperature, etc.) in most populations. High-quality habitats likely provide more food resources and complexity (rearing areas) than lower quality habitats (MacArthur and Levins 1967, entire). These features of high-quality habitats may lessen competition (MacArthur and Levins 1967, entire) and reduce predation (Almany 2004, p. 107) by providing complex rearing areas for the vulnerable young life stages of Arctic grayling. For these reasons historically, when many of the riverine habitats were degraded, competition and predation likely had a larger effect on Arctic grayling populations than they currently do. Competition and predation are likely still occurring in habitats occupied by both nonnatives and Arctic grayling. Increases in rainbow and brown trout abundance correlate with lower abundance of age-0 Arctic grayling in the Big Hole River; however, correlations do not definitively implicate competition/predation as casual mechanisms. The increase in habitat quality observed in recent years, particularly in the Big Hole River and Centennial Valley, appear to have minimized effects of competition and predation on respective Arctic grayling populations. The primary evidence of this is recent trends showing increasing numbers of both nonnatives and Arctic grayling in systems with high-quality habitat, including increasing brown trout and number of spawning adult Arctic grayling in the Big Hole River. Other populations of Arctic grayling have coexisted with brook, rainbow, and Yellowstone cutthroat trout for extended periods of time (>60 years) with no observed declines in grayling abundance.

Predation By Birds And Mammals

In general, the frequency and magnitude of predation by birds and mammals on Arctic grayling is not well understood because few detailed studies have been completed (Northcote 1995, p. 163). Black bear (*Ursus americanus*), mink (*Neovison vison*), and river otter (*Lontra canadensis*) are present in southwestern Montana, but direct evidence of predatory activity by these species is often lacking (Kruse 1959, p. 348). Osprey (*Pandion haliaetus*) can capture Arctic grayling during the summer (Kruse 1959, p. 348). In the Big Hole River, Byorth and Magee (1998, p. 926) attributed the loss of Arctic grayling from artificial enclosures used in a competition experiment to predation by minks, belted kingfisher (*Ceryle alcyon*), osprey, and great blue heron (*Ardea herodia*). In addition, American white pelican (*Pelecanus erythrorhynchos*) are seasonally present in the Big Hole River, and they also may feed on Arctic grayling. The aforementioned mammals and birds can be effective fish predators; however, Arctic grayling evolved with these native predator species and have developed life-history and reproductive strategies to mitigate for predation losses. We have no data demonstrating any of these species historically or currently consume Arctic grayling at levels sufficient to exert a measureable, population-level impact on native Arctic grayling in the upper Missouri River system. We expect the current situation to continue.

Summary of Factor C

Predation and competition can influence the distribution, abundance, and diversity of species in ecological communities. Predation by and competition with nonnative species can negatively affect native species, particularly those that are stressed or occurring at low densities due to unfavorable environmental conditions. Historically, the impact of predation and competition from nonnatives was likely greater because many of the habitats used by Arctic grayling were degraded. Thus, predation and competition likely played a role historically in decreasing the abundance and distribution of Arctic grayling. Currently, habitat conditions have improved markedly for those Arctic grayling populations primarily on Federal land (17 of 19 populations) and for the Big Hole River population on primarily private land. Predation and competition with nonnative species are still likely occurring in these systems, although the extent and magnitude of these effects appears to be mediated by habitat quality. Abundance of Arctic grayling is negatively correlated with increasing abundance of rainbow and brown trout in the Big Hole River; however, causal mechanisms remain unclear. Based on empirical evidence, number of nonnative cutthroat trout hybrids in the Centennial Valley are not influencing number of spawning adult Arctic grayling. We acknowledge nonnative trout densities are high in the Madison River and may be contributing to the decline of that Arctic grayling population; however, most other populations appear to have stable abundance of Arctic grayling and nonnatives. Further, Arctic grayling experts project only a small effect of predicted nonnative trout densities on Arctic grayling recruitment in the future.

Little is known about the effect of predation on Arctic grayling by birds and mammals. Such predation likely does occur, but we are not aware of any situation where an increase in fish-eating birds or mammals has coincided with the decline of Arctic grayling.

Factor D. The Inadequacy of Existing Regulatory Mechanisms

Section 4(b)(1)(A) of the Act requires the Service to take into account “those efforts, if any, being made by any State or foreign nation, or any political subdivision of a State or foreign nation, to protect such species...” We consider relevant Federal, State, and Tribal laws, and regulations when evaluating the status of the species. Regulatory mechanisms, if they exist, may preclude the need for listing if we determine that such mechanisms adequately address the threats to the species such that listing is not warranted. Only existing ordinances, regulations, and laws, that have a direct connection to a law, are enforceable and permitted are discussed in this section. All other measures are discussed under the specific relevant factor.

<u>U.S. Federal Laws and Regulations</u>

No Federal laws in the United States specifically address the Arctic grayling, but several, in their implementation, may affect the species’ habitat.

National Environmental Policy Act

All Federal agencies are required to adhere to the National Environmental Policy Act (NEPA) of 1970 (42 U.S.C. 4321 *et seq*.) for projects they fund, authorize, or carry out. The Council on Environmental Quality’s regulations for implementing NEPA (40 CFR 1500–1518) state that, when preparing environmental impact statements, agencies shall include a discussion on the environmental impacts of the various project alternatives, any adverse environmental effects which cannot be avoided, and any irreversible or irretrievable commitments of resources involved (40 CFR 1502). The NEPA itself is a disclosure law, and does not require subsequent minimization or mitigation measures by the Federal agency involved. Although Federal agencies may include conservation measures for Arctic grayling as a result of the NEPA process, any such measures are typically voluntary in nature and are not required by NEPA.

Federal Land Policy and Management Act

The Federal Land Policy and Management Act (FLPMA) of 1976 (43 U.S.C. 1701 *et seq*.), as amended, states that the public lands shall be managed in a manner that will protect the quality of scientific, scenic, historical, ecological, environmental, air and atmospheric, water resource, and archeological values. This statute protects lands within the range of the Arctic grayling managed by the Bureau of Land Management (BLM).

The BLM considers the Arctic grayling a sensitive species requiring special management consideration for planning and environmental analysis (BLM 2009a, entire, BLM 2009b, entire). The BLM has developed a resource management plan (RMP) for the Dillon Field Office Area that provides guidance for the management of over 900,000 acres of public land administered by BLM in southwest Montana (BLM 2006a, p. 2). The Dillon RMP area thus includes the geographic area that contains the Big Hole, Miner, Mussigbrod, Madison River, and Centennial Valley populations of Arctic grayling. A RMP planning area encompasses all private, State, and Federal lands within a designated geographic area (BLM 2006a, p. 2), but the actual implementation of the RMP focuses on lands administered by the BLM that typically represent only a fraction of the total land area within that planning area (BLM 2006b, entire). Restoring Arctic grayling habitat and ensuring the long-term persistence of grayling populations are among the RMP’s goals (BLM 2006a, pp. 30–31). However, there is little actual overlap between the specific parcels of BLM land managed by the Dillon RMP and the current distribution of Arctic grayling (BLM 2006b, entire).

The BLM also has a RMP for the Butte Field Office Area, which includes more than 300,000 acres in south-central Montana (BLM 2008, entire), including portions of the Big Hole River in Deerlodge and Silver Bow counties (BLM 2008, p. 8; 2009c, entire). The Butte RMP considers conservation and management strategies and agreements for Arctic grayling in its planning process and includes a goal to opportunistically enhance or restore habitat for Arctic grayling (BLM 2008, pp. 10, 30, 36). However, the Butte RMP does not mandate specific actions to improve habitat for Arctic grayling in the Big Hole River and little overlap exists between BLM-managed lands and Arctic grayling occupancy in this planning area.

National Forest Management Act

Under the U.S. Forest Service (USFS) National Forest Management Act (NFMA) of 1976, as amended (16 U.S.C. 1600 *et seq*.), the USFS strives to provide for a diversity of plant and animal communities when managing national forest lands. Individual national forests may identify species of concern that are significant to each forest's biodiversity. The USFS considers Arctic grayling a sensitive species (USFS 2004, entire) for which population viability is a concern. However, this designation provides no special regulatory protections.

Most of the upper Missouri River grayling populations occur on National Forest land; all 15 populations occupying lakes and the Ruby River population (majority on National Forest) occur on USFS-managed lands. These populations occur across four different National Forests; consequently the riparian habitats surrounding the lakes and tributaries are managed according to the standards and guidelines outlined in each National Forest Plan. All Forest Plans do not contain the same standards and guidelines; however, each Plan has standards and guidelines for protecting riparian areas around perennial water sources. In the Beaverhead-Deerlodge and Helena National

Forest Plans, the Inland Native Fish Strategy (INFS) standards and guidelines have been incorporated. The INFS, in part, defines widths of riparian buffer zones adequate to protect streams and lakes from non-channelized sediment inputs and contribute to other riparian functions, such as stream shading and bank stability. These protections have been incorporated into the Beaverhead-Deerlodge and Helena National Forest Plans through amendments and are currently preserving intact riparian areas around most, if not all, Arctic grayling habitats. Exceptions to the riparian protections outlined in INFS are occasionally granted; however, these exceptions require an analysis of potential effects and review by a USFS fish biologist.

On the Gallatin National Forest, standards and guidelines in the Forest Plan include using "best management practices" to protect water sources and riparian areas. Similar to INFS, best management practices outline buffer strips along watercourses where disturbance and activity is minimized to protect riparian areas and water quality. On the Lewis and Clark National Forest, standards and guidelines are in place to leave timbered buffer strips adjacent to waterbodies to protect riparian areas. Grayling habitat on the Gallatin and Lewis and Clark National Forests consists of seven high-elevation mountain lakes.

National Park Service (NPS) Organic Act

The NPS Organic Act of 1916 (16 U.S.C. 1 *et seq*.), as amended, states that the NPS "shall promote and regulate the use of the Federal areas known as national parks, monuments, and reservations ... to conserve the scenery and the national and historic objects and the wild life therein and to provide for the enjoyment of the same in such manner and by such means as will leave them unimpaired for the enjoyment of future generations." Arctic grayling are native to the western part of Yellowstone National Park and habitats are managed accordingly for the species under the

Native Species Management Plan (NPS 2010, entire; Koel et al. 2019, pp. 6-8). One Arctic grayling population, Grebe Lake, currently occurs in Yellowstone National Park. Other populations of Arctic grayling are currently attempting to be established in YNP in Goose, Wolf and Ice lakes, as well as the Gibbon River (Koel 2019, pers. comm.; Koel et al. 2019, pp. 6-8). The habitat in Grebe Lake and the other reintroduction sites is managed for conservation (Koel et al. 2019, pp. 6-8; NPS 2010, p. 44). Further, it is expected that these habitats will be managed for conservation in the future, based on provisions in the Organic Act and guidance outlined in the Native Species Management Plan.

National Wildlife Refuge System Improvement Act of 1997

The National Wildlife Refuge Systems Improvement Act (NWRSIA) of 1997 (Pub. L. 105-57) amends the National Wildlife Refuge System Administration Act of 1966 (16 U.S.C. 668dd *et seq*.). The NWRSIA directs the Service to manage the Refuge System's lands and waters for conservation. The NWRSIA also requires monitoring of the status and trends of refuge fish, wildlife, and plants. The NWRSIA requires development of a Comprehensive Conservation Plan for each refuge and management of each refuge consistent with its plan.

The Service has developed a final Comprehensive Conservation Plan to provide a foundation for the management and use of Red Rock Lakes NWR (USFWS 2009, entire) in the Centennial Valley. Since the development of the Comprehensive Conservation Plan, Refuge staff have conducted numerous habitat conservation/restoration projects to benefit Arctic grayling, including: removal of an earthen dam whose reservoir inundated several hundred meters of historical Arctic grayling spawning habitat in Elk Springs Creek, reconnection of Elk Springs Creek to Upper Red Rock Lake and subsequent reintroductions and tracking of young-of-year

Arctic grayling in Elk Springs Creek (West 2014a, pers. comm.). However to date, the reintroductions in Elk Springs Creek have not established a spawning run. Other conservation projects conducted on the Refuge include the acquisition of new land and decreases in grazing intensities from 20,000 AUMs to about 5,000 AUMs. The Refuge has implemented a rest-rotation grazing system where more durable lands are grazed while more sensitive lands (e.g., riparian areas) are rested for up to 4 years (West 2014a, pers. comm.). Some active riparian restoration has also occurred, including a project to reconnect Red Rock Creek to a historical channel and replacement of four culverts to allow for natural tributary migration across alluvial fans (West 2014a, pers. comm.). The Refuge is also actively engaged in supporting ongoing graduate research efforts to explore potential limiting factors for Arctic grayling in the Centennial Valley.

Other conservation projects under the Comprehensive Conservation Plan have been focused on potential nonnative species effects on Arctic grayling, namely a 5-year project removing hybrid cutthroat trout captured during their upstream spawning run (as outlined in the Adaptive Management Plan) and a study of dietary overlap between Arctic grayling and Yellowstone cutthroat trout (West 2014a, pers. comm.). The Refuge also operates a sill dam to provide upstream fish passage. The sill dam was previously a barrier to upstream fish movement, but is no longer a barrier due to changes in how the Refuge operates the sill dam (West 2014a, pers. comm.) The Refuge also operates one irrigation ditch, but only when snowpack is average or above (i.e, adequate flows exist in Red Rock Creek) and when young Arctic grayling are not present near the diversion (West 2014a, pers. comm.).

The proven track record of completed conservation projects on the refuge indicate that the continued implementation of the Comprehensive Conservation Plan during the next 5 years will continue to improve habitat conditions on the refuge.

Federal Power Act (FPA)

The Federal Power Act of 1920 (16 U.S.C. 791 *et seq*., as amended) provides the legal authority for the Federal Energy Regulatory Commission (FERC), as an independent agency, to regulate hydropower projects. In deciding whether to issue a license, FERC is required to give equal consideration to mitigation of damage to, and enhancement of, fish and wildlife (16 U.S.C. 797(e)). A number of FERC-licensed dams exist in the Missouri River basin in current (i.e., Ennis Dam on the Madison River) and historical Arctic grayling habitat (e.g., Hebgen Dam on the Madison River; Hauser, Holter, and Toston dams on the mainstem Missouri River; and Clark Canyon Dam on the Beaverhead River). The FERC license expiration dates for these dams range from 2024 (Toston) to 2059 (Clark Canyon) (FERC 2010, entire). None of these structures provides upstream passage of fish, and such dams are believed to be one of the primary factors that led to the historical decline of Arctic grayling in the Missouri River basin (see discussion under Factor A, above). However, recent monitoring data indicate multiple stable Arctic grayling populations occurring above mainstem dams, with the exception of the Ennis Reservoir/Madison River population. The drawdowns in reservoir water level believed to have historically affected the Ennis Reservoir/Madison River Arctic grayling population are not permitted under a new licensing agreement between the Federal Energy Regulatory Commission and Madison Dam operators, as we described previously in this finding (Clancey 2014, pers. comm.).

Clean Water Act

The Clean Water Act (CWA) of 1972 (33 U.S.C. 1251 *et seq*.) establishes the basic structure for regulating discharges of pollutants into the waters of the United States and regulating quality standards for surface waters. The CWA's general goal is to "restore and maintain the chemical, physical, and biological integrity of the Nation's waters" (33 U.S.C. 1251(a)). The CWA requires States to adopt standards for the protection of surface water quality and establishment of total maximum daily load (TMDL) guidelines for rivers. The Big Hole River has approved TMDL plans for its various reaches (MDEQ 2009a, entire; 2009b, entire); thus, complete implementation of this plan should improve water quality (by reducing water temperatures, and reducing sediment and nutrient inputs) in the Big Hole River in the future. As of October 2017, a TMDL was being developed for the Madison River watershed, but no significant TMDL plan development activity in the Red Rock watershed in the Centennial Valley (see MDEQ 2017). Currently, TMDL documents have been approved for the Ruby River. All planning areas containing other Arctic grayling populations in the upper Missouri River basin have approved TMDLs, including the Gallatin, Lake Helena, and Sun watersheds (see MDEQ 2014).

State Laws

Montana Environmental Policy Act

The legislature of Montana enacted the Montana Environmental Policy Act (MEPA) as a policy statement to encourage productive and enjoyable harmony between humans and their environment, to protect the right to use and enjoy private property free of undue government regulation, to promote efforts that will prevent or eliminate damage to the environment and biosphere and stimulate the health and welfare of humans, to enrich the understanding of the ecological systems and natural resources important to the State, and to establish an environmental quality council (MCA 75-1-102). Part 1 of the MEPA establishes and declares Montana's environmental policy. Part 1 has no legal requirements, but the policy and purpose provide guidance in interpreting and applying statutes. Part 2 requires State agencies to carry out the policies in Part 1 through the use of systematic, interdisciplinary analysis of State actions that have an impact on the human environment. This is accomplished through the use of a deliberative, written environmental review. In practice, MEPA provides a basis for the adequate review of State actions in order to ensure that environmental concerns are fully considered (MCA 75-1-102). Similar to NEPA, the MEPA is largely a disclosure law and a decision-making tool that does not specifically require subsequent minimization or mitigation measures.

Laws Affecting Physical Aquatic Habitats

A number of Montana State laws have a permitting process applicable to projects that may affect stream beds, river banks, or floodplains. These include the Montana Stream Protection Act, the Streamside Management Zone Law, and the Montana Natural Streambed and Land Preservation Act (Montana Department of Natural Resources (MDNRC) 2001, pp. 7.1–7.2). The Montana Stream Protection Act requires that a permit be obtained for any project that may affect the natural and existing shape and form of any stream or its banks or tributaries (MDNRC 2001, p. 7.1). The Montana Natural Streambed and Land Preservation Act requires private, nongovernmental entities to obtain a permit for any activity that physically alters or modifies the bed or banks of a perennially flowing stream (MDNRC 2001, p. 7.1). The Montana Stream Protection Act and Montana Natural Streambed and Land Preservation Act do not mandate any special recognition for species of concern, but in practice, biologists that review projects permitted under these laws usually stipulate restrictions to avoid harming such species (Horton 2010, pers. comm.). The Streamside Management Zone Law regulates forest practices near streams (MDNRC

2001, p. 7.2). The Montana Pollutant Discharge Elimination System Stormwater Permit applies to all discharges to surface water or groundwater, including those related to construction, dewatering, suction dredges, and placer mining, as well as to construction that will disturb more than 1 acre within 100 ft (30.5 m) of streams, rivers, or lakes (MDNRC 2001, p. 7.2).

Review of applications by Montana Fish, Wildlife and Parks, Montana Department of Environmental Quality and Montana Department of Natural Resources Conservation is required prior to issuance of permits under the above regulatory mechanisms (MDNRC 2001, pp. 7.1–7.2). These regulatory mechanisms are expected to limit impacts to aquatic habitats in general.

Montana Water Use Act

The purpose of the Montana Water Use Act (Title 85: Chapter 2, Montana Codes Annotated) is to provide water for existing and future beneficial use and to maintain minimum flows and water quality in Montana's streams. The Missouri River system is generally believed to be overappropriated, and water for additional consumptive uses is only available for a few months during very wet years (MDNRC 1997, p. 12). However, the upper Missouri River basin and Madison River basin have been closed to new water appropriations because of water availability problems, overappropriation, and a concern for protecting existing water rights (MDNRC 2009, p. 45). In addition, recent compacts (a legal agreement between Montana, a Federal agency, or an Indian tribe determining the quantification of federally or tribally claimed water rights) have been signed that close appropriations in specific waters in or adjacent to Arctic grayling habitats. For example, the USFWS–Red Rock Lakes–Montana Compact includes a closure of appropriations for consumptive use in the drainage basins upstream of the most downstream point on the Red Rock Lakes NWR and the Red Rock Lakes Wilderness Area (MDNRC 2009, pp. 18, 47). The NPS–

Montana Compact specifies that certain waters will be closed to new appropriations when the total appropriations reach a specified level, and it applies to Big Hole National Battlefield and adjacent waters (North Fork of the Big Hole River and its tributaries including Ruby and Trail Creeks), and the portion of Yellowstone National Park that is in Montana (MDNRC 2009, p. 48).

The State of Montana is currently engaged in a Statewide effort to adjudicate (finalize) water rights claimed before July 1, 1973. The final product of adjudication in a river basin is a final decree. To reach completion, a decree progresses through several stages: (1) Examination, (2) temporary preliminary decree, (3) preliminary decree, (4) public notice, (5) hearings, and (6) final decree (MDNRC 2009, pp. 9–14). As of May 2019, the Centennial Valley has a preliminary decree, and the Big Hole and Madison Rivers have temporary preliminary decrees (MDNRC 2019, entire). It is unclear when final adjudication of all the river basins in Montana that currently contain native Arctic grayling will be completed, and we do not know if this process will eliminate the potential overallocation of water rights. We note that the overallocation of water in some systems within the upper Missouri river basin is of general concern to Arctic grayling because of the species' need for adequate quantity and quality of water for all life stages.

Angling Regulations

Arctic grayling are considered a game fish, but are subject to special catch-and-release regulations in streams and rivers within its native range, as was described under Factor B, above (MFWP 2019, p. 57). Catch-and-release regulations also are in effect for Ennis Reservoir on the Madison River (MFWP 2019, p. 65). Arctic grayling in other populations are subject to more liberal regulations; anglers can keep up to 5 per day and have up to 10 in possession in accordance with standard daily and possession limits for that angling management district (MFWP 2019, p. 57).

Summary of Factor D

Current Federal and State regulatory mechanisms are adequate to protect Arctic grayling of the upper Missouri River. We conclude this because the majority of populations are on Federal land where regulatory mechanisms are in place to preserve intact habitats and are expected to remain in place. In the Big Hole River, Arctic grayling generally occupy waters adjacent to private lands (MFWP *et al.* 2006, p. 13; Lamothe *et al*. 2007, p. 4), so Federal regulations may have limited ability to protect that population. However, other measures are in place to promote conservation on private lands. Some Federal regulations (e.g., CWA, FPA, NMFA, NWRSIA, NPS Organic Act) are adequate to sustain and improve habitat conditions for Arctic grayling. In addition, we did not identify other threats to the DPS that would require regulatory protections.

Factor E. Other Natural or Manmade Factors Affecting Its Continued Existence

Drought

Drought is a natural occurrence in the interior western United States (see National Drought Mitigation Center 2010). The duration and severity of drought in Montana appears to have increased during the last 50 years, and precipitation has tended to be lower than average in the last 20 years (National Climatic Data Center 2010). Drought can affect fish populations by reducing stream flow volumes. This leads to dewatering and high temperatures that can limit connectivity among spawning, rearing, and sheltering habitats. Drought can also reduce the volume of thermally suitable habitat and increase the frequency of water temperatures above the physiological limits for optimum growth and survival in Arctic grayling. In addition, drought can interact with human-caused stressors (e.g., irrigation withdrawals, riparian habitat degradation) to further reduce stream flows and increase water temperatures.

Reduced stream flows and elevated water temperatures during drought have been most apparent in the Big Hole River system (Magee and Lamothe 2003, pp. 10-14; Magee *et al.* 2005, pp. 23-25; Rens and Magee 2007, pp. 11-12, 14). In the Big Hole River, evidence for the detrimental effects of drought on Arctic grayling populations is primarily inferential; observed declines in Arctic grayling and nonnative trout abundances in the Big Hole River coincide with periods of drought (Magee and Lamothe 2003, pp. 22–23, 28) and fish kills (Byorth 1995, pp. 10–11, 31).

Although the response of stream and river habitats to drought is expected to be most pronounced because of the strong seasonality of flows in those habitats, effects in lake environments can occur. For example, both the Upper and Lower Red Rock Lakes are shallow (Mogen 1996, p. 7). Increased frequency or duration of drought could lead to increased warming in shallower lakes, such as Upper Red Rock Lake. However, the Centennial Valley has many spring sources (Mogen 1996, p. 82) and multiple tributaries (USFWS 2012, unpublished data) that could, at least in part, mitigate for increases in water temperature due to increased drought frequency and magnitude. Other potential effects from drought could include a reduction in overall lake depth, which could in turn affect summer or overwintering habitat. Arctic grayling populations in high mountain lakes would likely not be affected significantly by drought because air (and thus water) temperatures in these habitats are relatively cool due to the greater distance from sea level at high elevations (~ a 3.6 °F (6.5 °C) decrease in air temperature for every 3,200 ft.

(1 kilometer) above sea level; Physics 2014). In addition, most of these habitats are relatively large bodies of water volumetrically, thus are resistant to warming, given the high specific heat of water (USGS 2014). Further, intact riparian areas in these habitats buffer against water temperature increases in tributaries by blocking incoming solar radiation (Sridhar *et al*. 2004, entire; Cassie 2006, p. 1393).

Given the climate of the intermountain West, we conclude that drought has been and will continue to be a natural occurrence. We assume that negative effects of drought on Arctic grayling populations, such as reduced connectivity among habitats or increased water temperatures at or above physiological thresholds for growth and survival, are more frequent in stream and river environments and in very shallow lakes relative to larger, deeper lakes. As discussed under Factor A, the implementation of the Big Hole Arctic grayling CCAA is likely to minimize some of the effects of drought in the Big Hole River, by reducing the likelihood that human-influenced actions or outcomes (irrigation withdrawals, destruction of riparian habitats, and fish passage barriers) will interact with the natural effects of drought (reduced stream flows and increased water temperatures). We expect the impact of drought may act at the individual level, but not at the population or DPS level because most grayling populations reside in drought-resistant habitats in high mountain lakes. Some populations will likely be affected by drought, but implemented conservation measures and natural spring sources and cooler tributaries are expected to minimize the impact in both the Big Hole River and Centennial Valley. Drought is expected to increase in both duration and severity in the future due to climate change; however, resiliency currently being incorporated into riparian and aquatic habitats through conservation projects will likely buffer many of the predicted effects of drought.

Stochastic (Random) Threats, Genetic Diversity and Small Population Size

A principle of conservation biology is that the presence of larger and more productive (resilient) populations can reduce overall extinction risk. To minimize extinction risk due to stochastic (random) threats, life-history diversity should be maintained, populations should not all share common catastrophic risks (i.e. sufficient redundancy), and both widespread and spatially close populations are needed (Fausch *et al.* 2006, p. 23; Allendorf *et al.* 1997, entire).

The Upper Missouri River DPS of Arctic grayling exists largely as a collection of isolated populations (Peterson and Ardren 2009, entire), with little to no gene flow among populations. While the inability of fish to move between populations limits genetic exchange and demographic support (Hilderbrand 2003, p. 257), large population sizes coupled with adequate number of breeding individuals minimize the effects of isolation. For example, Mussigbrod Lake, a large population, receives no genetic infusion from any other population in the upper Missouri River basin, yet has a very large estimated effective population size (see Table 3, above). Loss of genetic diversity from genetic drift is not a concern for this population, despite it being reproductively isolated.

Abundance varies widely among the 19 Arctic grayling populations (see Table 3, above). Individually, small populations like Ruby River need to maintain enough adults to minimize loss of variability through genetic drift and inbreeding (Rieman and McIntyre 1993, pp. 10–11). The Ruby River population exhibits a low effective number of breeders, but contains the second highest genetic diversity among all populations (Leary 2014, unpublished data) and has shown stable genetic diversity through time (Gander et al. 2019, pp. 5-6). Future monitoring is warranted to track genetic diversity of the population (Gander et al. 2019, pp. 5-6).

Known effective population size estimates for other Arctic grayling populations vary from 162 to 1,497, although estimates have not been calculated for some of the populations (see Table 3, above). There has been considerable debate about what effective population size is adequate to conserve genetic diversity and long-term adaptive potential (see Jamieson and Allendorf 2012 for review, p. 579). However, loss of genetic diversity is typically not an immediate threat even in isolated populations with an N_e <100 (Palstra and Ruzzante 2008, p. 3441), but rather is a symptom of deterministic processes acting on the population (Jamieson and Allendorf 2012, p. 580). In other words, loss of genetic diversity due to small effective population size typically does not drive species to extinction (Jamieson and Allendorf 2012, entire); other processes, such as habitat degradation, have a more immediate and greater impact on species persistence (Jamieson and Allendorf 2012). We acknowledge that loss of genetic diversity can occur in small populations; however, in this case, it appears that there are adequate numbers of breeding adults to minimize loss of genetic diversity in many of the populations.

The full range of life histories of Arctic grayling is being expressed in the current DPS. Conservation of life-history diversity (i.e., a form of representation) is important to the persistence of species confronted by habitat change and environmental perturbations (Beechie *et al.* 2006, entire). Reintroductions of Arctic grayling into the upper Ruby River have provided additional redundancy and representation of life history diversity. From multiple populations that occupy rivers or streams for all of their life history to others that primarily occupy lakes, all are representative of life history diversity of historical Arctic grayling in the upper Missouri River basin. Thus, conservation of the full range of life history diversity is being achieved in the DPS.

Populations of Arctic grayling in the Upper Missouri River DPS are for the most part widely separated from one another, occupying 7 of 10 historically occupied watersheds (see Table

3, above). Thus, risk of extirpation by a rare, high-magnitude environmental disturbance (i.e., catastrophe) is relatively low. In addition, multiple spawning locations exist for 11 of the 19 populations in the Upper Missouri River DPS (USFWS 2019, unpublished data). The 11 populations with access to multiple spawning tributaries include all the largest populations in terms of abundance, except Mussigbrod Lake (see Table 3). Abundance and number of breeding individuals is adequate in most populations to sustain moderate to high levels of genetic diversity currently observed.

Summary of Factor E

Overall, we conclude that the Upper Missouri River DPS of Arctic grayling has faced historical threats from drought, loss of genetic diversity, and small population size. However, the DPS currently exists as multiple, isolated populations across a representative portion of its historical range. While reproductive isolation can lead to detrimental genetic effects, the current effective population size of most Arctic grayling populations, suggest these effects will be minimal. In instances where effective number of breeders is low (e.g., Ruby River), no measurable loss of genetic diversity has occurred, although future monitoring is warranted. Redundancies within and among populations are present: multiple spawning tributaries, geographic separation, range of life-history diversity and replication.

Cumulative Effects from Factors A through E

We focus our discussion of cumulative effects from Factors A through E on interactions involving climate change. Our rationale for this is that climate change has the highest level of uncertainty among other factors considered, and likely has the most potential to affect Arctic grayling populations when interacting with other factors.

Climate Change and Nonnative Species Interactions

Changes in water temperature due to climate change may influence the distribution of nonnative trout species (Rahel and Olden 2008, p. 524) and the outcome of competitive interactions between those species and Arctic grayling. Brown trout are generally considered to be more tolerant of warm water than many salmonid species common in western North America (Coutant 1999, pp. 52–53; Selong *et al.* 2001, p. 1032), and higher water temperatures may favor brown trout where they compete against salmonids with lower thermal tolerances (Rahel and Olden 2008, p. 524). Recently, observed increases in the abundance and distribution of brown trout in the upper reaches of the Big Hole River (MFWP 2013, unpublished data) may be consistent with the hypothesis that stream warming is facilitating encroachment. In addition, recent analyses indicate increasing abundance of brown trout is negatively correlated with age-0 Arctic grayling abundance in the Big Hole River (McCullough 2017, pp. 16, 22).

Currently, brown trout are at relatively low densities (<20 fish/mile) in the upper Big Hole River, where Arctic grayling densities are highest (MFWP 2013e, unpublished data). At densities of 100 brown trout per mile (a plausible future scenario), Arctic grayling experts predicted a 5 percent reduction in Arctic grayling recruitment in the Big Hole River, due to competition and predation (Service 2014, p. 2).

Climate Change/Dewatering/Water Temperature Increases

In 2014, we stated that synergistic interactions were possible between climate change and other potential stressors such as dewatering, but that those potential interactions were too speculative to characterize at that time. Since 2014, more climate change studies have emerged that provide more information about these potential interactions. One theme in several of these studies is that projected effects of climate change on water temperature and water availability are not expected to be synergistic (i.e., that the collective sum of effects is greater than the sum of individual effects), but rather cumulative (i.e., increasing by successive additions). For example, climate change may influence water availability (Schewe et al. 2014, p. 3246) which may in turn increase water temperatures (Vatland et al. 2015, p. 49; Isaak et al. 2016, pp. 3-4), but these predicted effects are a series of direct and indirect effects that may accumulate through time and become cumulative. Thus, our discussion below focuses on the potential cumulative effects of climate change on the properties of water.

Warming air temperatures due to future climate change are expected globally (IPCC 2018, p. 4-6) and regionally in the upper Missouri River basin (Ray et al. 2010, p. 23). As air temperatures increase, regional water temperatures in some streams or portions of streams are also expected to increase (Isaak et al. 2015, pp. 2545-2546, 2549-2551; Vatland et al. 2015, p. 49; Isaak et al. 2016, pp. 2-6). Recent analyses indicate that past warming of regional water temperatures (including most of the upper Missouri River basin) has progressed slower than previously modeled, and future predicted warming is likely to continue at a similar, slow rate (Isaak et al. 2016, pp. 3-6). For example, streams with mean August temperatures >10°C (such as the Big Hole River and its tributaries in Montana), warmed in the past, on average, of 0.105°C per decade (Isaak et al. 2016, p. 3).

Warming water temperatures due to climate change are predicted to decrease available habitat for cold-water fishes; however, habitats capable of supporting cold-water fishes are predicted to remain at least into the 2080s. For example, habitats capable of supporting bull trout and westslope cutthroat trout, two salmonids with cooler thermal tolerances than Arctic grayling, are expected to remain into the 2080s in the Pacific Northwest (including most of the upper Missouri River basin), even under the most extreme climate scenarios modeled (Isaak et al. 2015, pp. 2542, 2548-2549). The Big Hole River and some of its tributaries were included in this study, thus we would expect similar trends in Arctic grayling habitat, where some warming is predicted to occur, but areas of thermal refugia remain intact. In support of this, modeled water temperatures in the Big Hole River indicate that cooler water sites used by Arctic grayling in the 2000s were expected to remain relatively cool through at least the 2060s (Vatland 2015, pp. 49, 65). Thus, while decreases in available habitat for cold-water fish are predicted to occur from climate change, many habitats are expected to be more climate-resilient.

Recently, another study modeled the future effects of climate change on salmonids and their habitat in the Big Hole River in Montana. In the Big Hole River, Vatland modeled exceedance of the 21°C and 25°C water temperature thresholds for the future decades 2040s and 2060s (Vatland pp. 47-51). These water temperature thresholds were calculated as weekly mean of daily maximum temperatures. It is important to note that a weekly mean of daily maximum temperature is an average, thus for a given threshold (21°C or 25°C), daily maximum temperatures within any given 7 day period are going to be higher and lower than the mean. In addition, water temperatures fluctuate widely on a daily basis, with fish potentially being exposed to water temperatures greater than either threshold for portions of day, not constantly for entire days as in the upper incipient lethal temperature methodology. In the Big Hole River, weekly mean of daily maximum water temperatures were modeled to exceed both 21°C and 25°C thresholds more often in the 2040s and 2060s (Table 9) than they do currently. In addition, the amount of total stream network exceeding these same thresholds was modeled to increase in the 2040s and 2060s (Table 9; Vatland 2015, pp. 48, 50-51). Cumulatively, these data indicate predicted future warming in the Big Hole River and tributaries as a result of climate change, both in terms of frequency (more frequent exceedances of water temperature thresholds through time) and space (increases in amount of stream network exceeding water temperature thresholds).

Table 9. Percent of summer weeks and water temperature measurement sites that exceeded (for decades 1980s and 2000s) or were modelled to exceed (for 2040s and 2060s) the chronic (21°C) and acute (25°C) water temperature thresholds for Arctic grayling in the upper Big Hole River and tributaries at least once in the stated decade. Water temperature thresholds are calculated as weekly mean of daily maximum water temperatures. Asterisks denote estimates derived from Figures 4 (p. 65), 6 (p. 67) and 7 (p. 68) in Vatland 2015.

		1980s	**2000s**	**2040s**	**2060s**
	Threshold				
% summer weeks exceeding:	21°C	20	26*	35*	44
	25°C	<1	1*	3*	5
% water temperature measurement sites exceeding:	21°C	80	92*	99*	100
	25°C	0	10*	32*	64

All modeling studies rely on certain assumptions and therefore have inherent limitations with respect to their output. We note several key assumptions used in the Vatland 2015 study that provide important context. First, two sites with groundwater influence were specifically excluded from this study (Vatland 2015, pp. 47, 53, 63). This likely resulted in the model runs underestimating the importance of variation in water temperature in space and time (Vatland 2015, pp. 53-54), especially given the documented biological importance of such features to provide thermal refugia and salmonids ability to seek out and use these features (Wojcik 1955, pp. 25-26; LaPerriere and Carlson 1973 pp. 32-33; Berman and Quinn 1991, pp. 307-310; Baird and Krueger 2003, pp. 1198-1199, 1202-1204; Nielsen et al. 1994, pp. 620-624; Matthews and Berg 1997, pp. 61, 63; Beitinger et al. 2000, p. 239; Quigley and Hinch 2006, p. 429; Breau et al. 2007, pp. 1183-1186, 1188-1189; Vatland et al. 2009, pp. 10-15; Breau et al. 2011, pp. 850-851; references in Chadwick et al. 2015, p. 9; Dugdale et al. 2015, pp. 7-8, 10-14). Second, the analysis in this study was based on a static landscape (Vatland 2015, pp. 55-56), with no accounting for any conservation measures implemented after the conclusion of the study (i.e., 2010; Vatland 2015, p. 35). This is important to consider, especially given the extensive conservation efforts and focus in the Big Hole River valley that have occurred under the CCAA since the Vatland 2015 study concluded (i.e., 2010). Thus, the Vatland 2015 study represents an incomplete assessment of effects of climate change on Arctic grayling and their habitat in the Big Hole River and its tributaries, due to the associated assumptions and limitations of the study design.

Since the conclusion of the Vatland study, multiple lines of more recent empirical evidence indicate that riparian area restoration along tributaries as part of the Big Hole CCAA has been successful at mitigating warming water temperatures due to climate change. McCullough 2019 documented a significant correlation between riparian health and water temperature in tributaries to the Big Hole River, where greater riparian health correlated with lower mean daily water temperatures (expressed as lower difference in degree-days; McCullough 2019, pp. 5, 7-8). Similarly, shorter durations of water temperatures that are stressful to Arctic grayling in tributaries have been observed since the inception of the CCAA, despite a warming climate (see Figure 4). Further evidence of the effectiveness of riparian restoration at mitigating water temperature increases in smaller streams was shown in another study in Montana, where water temperatures in some streams with restored riparian vegetation had 7°C lower summer maximum water temperatures than prior to riparian restoration (Pierce et al. 2013, pp. 72, 78). In Oregon, simulations in a forested setting indicated riparian shading could result in stream temperatures that were cooler than current conditions, despite a projected increase of 4°C in air temperature due to climate change (Wondzell et al. 2019, pp. 123, 128), further emphasizing the ability of riparian areas to mitigate rising water temperatures. All these findings are also consistent with Vatland 2015, who also found that riparian health was a significant predictor of summer water temperature in Big Hole River tributaries (Vatland 2015, pp. 49-50, 56) and recommended riparian restoration as a way to mitigate the effects of climate change on water temperature. Cumulatively, multiple lines of independent, primarily empirical evidence all indicate that riparian vegetation restoration, a core focus of the Big Hole CCAA, is an effective way to mitigate the effects of climate change on water temperature in tributaries.

In larger streams/rivers that are wider than primary tributaries, such as the mainstem Big Hole River, maintaining or increasing instream flows have been shown to have a larger impact than riparian vegetation/shading on keeping water temperatures cooler (Poole and Berman 2001, pp. 790-792; Allan 1995, entire). More instream flow increases the volume of water that heat (e.g., solar, etc.) interacts with, thus the water stays cooler relative to if the same amount of heat interacted with a lesser volume of water (Poole and Berman 2001, p. 789), which would result in warmer water. In the mainstem Big Hole River, water temperatures considered stressful or lethal to Arctic grayling have declined in cumulative duration since the inception of the CCAA (see Figure 4), which places primary emphasis of conservation actions on maintaining biologically-based flow targets in the mainstem river. Reductions in duration of stressful or lethal water temperatures, especially during times of drought or unseasonably warm air temperatures, is likely a result, at least in part, of the Big Hole CCAA's focus on maintaining or increasing instream flows (for a more in-depth discussion on instream flow contributions from enrolled landowners in the Big Hole CCAA, see Tables 5 and 7).

Overall, warming stream temperatures are predicted in the Big Hole River due to future climate change, but have been shown to be occurring at a slower rate than previously modeled. Thermal refugia, in the form of groundwater inputs and tributaries, are expected to be present and available to Arctic grayling in the future. Modeling of water temperatures in the Big Hole River project increases in frequency of temperature thresholds being exceeded over greater areas of stream, although the assumptions and limitations of this modeling study do not incorporate the empirically-documented heterogeneous water temperatures or extensive conservation efforts that have been conducted under the Big Hole CCAA since the study ended in 2010. Recent empirical evidence from multiple sources clearly show that riparian vegetation restoration is effective at mitigating water temperature increases from warming air temperatures due to climate change in tributaries to the Big Hole River. Coupled with a focus of the Big Hole CCAA on maintaining adequate flows for Arctic grayling in the mainstem river, water temperatures considered stressful or lethal to Arctic grayling are trending downward since the inception of the CCAA. Thus, while small cumulative impacts of warming water temperatures due to climate change are expected, they are expected to be less than modeling studies suggest and mitigated in large part by restoring riparian areas and restoring more flow to the mainstem Big Hole River, both of which are central tenets of the Big Hole CCAA.

Summary

Recent genetic analyses have concluded that many of the introduced populations of Arctic grayling in the upper Missouri River basin contain moderate to high levels of genetic diversity and that these populations were created from local sources within the basin. These introduced populations currently occur within the confines of the upper Missouri River basin and occupy high quality habitats on Federal land, the same places the Service would look to for long-term conservation of the species, if needed. As such, these populations and their future adaptive potential have conservation value and are included in the Upper Missouri River DPS of Arctic grayling.

Currently, we recognize 19 populations of Arctic grayling in the Upper Missouri River DPS, 17 of which occur primarily on Federal land. Adequate regulatory mechanisms exist to ensure the conservation of habitat on Federal land for these populations. Historical habitat degradation on private land has affected the Big Hole River population; however, habitat conditions have been improving since the implementation of the Big Hole CCAA in 2006. Conservation actions associated with the Big Hole CCAA and Strategic Habitat Conservation Plan have reduced water temperatures in tributaries, increased instream flows in tributaries and the mainstem Big Hole River, decreased the duration of stressful or lethal water temperatures for Arctic grayling, connected almost all core habitat so Arctic grayling can access thermal refugia if water temperatures become too warm in parts of the Big Hole River system, and improved riparian health. Arctic grayling have responded favorably to these improvements because distribution and number of breeding adults have increased throughout the upper Big Hole River. The Service is encouraged by the successful track record of conservation actions implemented under the Big Hole CCAA and Strategic Habitat Conservation Plan over the past 13 years and the recent implementation of the Centennial Valley CCAA and Adaptive Management Plan.

Riparian restoration and increasing flow contribution efforts in the Big Hole River and Centennial Valley are ongoing and will continue to be key in mitigating the anticipated effects of drought and climate change. Increased shading of tributaries can effectively minimize effects from increasing air temperatures and drought and instream flow contributions have been effective at reducing the duration of stressful or lethal water temperatures conditions in the mainstem Big Hole River. In addition, these changes to habitat can alter predation and competition potential where both nonnative species and Arctic grayling coexist, as they have for over 100 years in some populations.

We acknowledge the uncertainty regarding the current status of the Madison River population and probable declining trend in abundance. The factors influencing the current demographics of this population are unclear. However, we are encouraged by the recent FERC relicensing agreement precluding reservoir drawdowns that likely affected this population and its habitat in the past and the recent reintroduction efforts in Odell Creek and Yellowstone National Park.

In conclusion, we find viable populations representing the full range of life history diversity in the DPS, the majority of which occur on Federal land and are protected by Federal land management measures. Effective number of breeding adults are currently increasing in the Big Hole River, stable in the Centennial Valley, and decreasing in the Ruby River. High-quality habitat is present for most populations or is improving where it is not optimal (e.g., Big Hole River). Health of riparian areas is trending upward and will be key to minimizing effects of climate change and drought. All Arctic grayling populations are relatively genetically diverse, are of Montana-origin, and occur in 7 of 10 historically occupied watersheds.

In 2010, we identified multiple threats as acting on the Upper Missouri River DPS of Arctic grayling. At that time, we determined that habitat-related threats included habitat fragmentation, dewatering, thermal stress, entrainment, riparian habitat loss, and effects from climate change. Since 2010, we have 9 additional years of monitoring data and have gained new insight. It is now apparent that these threats are being effectively mitigated on private land (Big Hole River) by conservation actions under the Big Hole CCAA and do not appear to be present or acting at a level to warrant concern on most of the other populations. Almost all (98 percent) of core habitat in the Big Hole River is now connected, allowing Arctic grayling access to thermal refugia, if needed. Recent riparian restoration activities have appreciably reduced water temperatures and improved riparian habitat in tributaries to the Big Hole River and are expected to buffer the effects of climate change. Entrainment of Arctic grayling into irrigation canals in the Big Hole system is low. Habitats on Federal land are largely intact and these populations are not subject to many of the stressors historically identified for other populations because no irrigation diversions are present, habitats are primarily high-elevation lakes that have cool water temperatures, and riparian areas are largely intact.

In 2010, another threat identified as acting on the Upper Missouri River DPS of Arctic grayling was the presence of nonnative trout. We considered nonnative trout a threat at that time because we were aware of several instances where Arctic grayling declines had occurred following nonnative trout introductions. Currently, we have a better understanding of the interactions between nonnative trout and Arctic grayling. Our review of these interactions and case histories suggests that habitat degradation, concurrent with nonnative trout introductions, likely contributed to historical declines in Arctic grayling in those instances. Further, it appears the effect of nonnative trout on Arctic grayling are likely habitat-mediated; nonnative trout affect Arctic grayling disproportionately when habitat conditions are degraded, but both Arctic grayling and nonnatives can coexist at viable levels when habitat conditions are improved. The primary evidence supporting this assertion is the increasing abundance and distribution of both Arctic grayling and nonnatives in the Big Hole River (brown trout) and Centennial Valley (Yellowstone cutthroat trout before suppression began). Another line of evidence to support this assertion is observed spatial segregation between nonnatives and Arctic grayling in the core Arctic grayling areas in the Big Hole River, especially spawning and rearing areas (Service 2014). In addition, Arctic grayling in adfluvial habitats have maintained stable or increasing population levels in the presence of brook, rainbow, and Yellowstone cutthroat trout for over 100 years in many instances in the upper Missouri River basin, where habitat degradation has not occurred or been extensive.

In 2010, we stated that existing regulatory mechanisms were inadequate to protect the Upper Missouri River DPS of Arctic grayling. The primary reason for this assertion was that Arctic grayling populations were reported as declining; thus existing regulatory mechanisms were believed to be inadequate because they had failed to halt or reverse this decline. Currently, we have updated information indicating that most populations of Arctic grayling are either stable or increasing. Existing regulatory mechanisms have precluded riparian habitat destruction on Federal lands or mandated restoration of impaired areas and are expected to provide similar protections in the future.

In 2010, we identified reduced genetic diversity, low abundance, random events, drought, and lack of a fluvial replicate as threats to the Upper Missouri River DPS of Arctic grayling. Updated genetic information that was not available in 2010 indicates moderate to high levels of genetic diversity within most Arctic grayling populations in the DPS. Further, estimates of number of effective breeders and effective population size derived from this updated genetic information indicate more robust Arctic grayling populations than previously thought. Adequate redundancy exists within the DPS to minimize the effects of random events and drought; most lake habitats occupied by most Arctic grayling populations are relatively drought-resistant. Lastly, there are now 10 years of documented natural reproduction in the Ruby River and acknowledgement of more life history diversity in the Madison River and Centennial Valley populations than previously known, which increases the redundancy and representation of life history diversity in the DPS.

Determination of Species Status

Section 4 of the Act (16 U.S.C. 1533) and its implementing regulations (50 CFR part 424) set forth the procedures for determining whether a species meets the definition of “endangered species” or “threatened species.” The Act defines an “endangered species” as a species that is “in danger of extinction throughout all or a significant portion of its range,” and a “threatened species” as a species that is “likely to become an endangered species within the foreseeable future throughout all or a significant portion of its range.” The Act requires that we determine whether a species meets the definition of “endangered species” or “threatened species” because of any of the following factors: (A) The present or threatened destruction, modification, or curtailment of its habitat or range; (B) Overutilization for commercial, recreational, scientific, or educational purposes; (C) Disease or predation; (D) The inadequacy of existing regulatory mechanisms; or (E) Other natural or manmade factors affecting its continued existence.

Status Throughout All of Its Range

We have evaluated potential threats to the Upper Missouri River DPS of arctic grayling, and assessed the cumulative effect of the threats under the section 4(a)(1) factors, including curtailment of range and distribution (Factor A), dams (Factor A), water management (Factor A), habitat fragmentation/smaller seasonal barriers (Factor A), degradation of riparian habitat (Factor A), dewatering and increased water temperatures (Factor A), entrainment (Factor A), sedimentation (Factor A), overwinter conditions (Factor A), climate change (Factor A), recreational angling (Factor B), monitoring and scientific study (Factor B), restoration efforts (Factor B), disease (Factor C), predation and competition (Factor C), drought (Factor E), stochastic threats, genetic diversity and small population size (Factor E). We also evaluated whether existing regulatory mechanisms are adequate (Factor D), and evaluated existing conservation efforts to reduce the impact of these stressors.

Overall, we found that the potential threats we evaluated are having minimal impacts in most arctic grayling populations within the DPS. Fifteen out of the 19 populations occur in high-elevation lakes primarily on high quality habitats on Federal land, and are considered stable and have minimal to no impacts from stressors. The other four populations have a fluvial component, and of these, the Big Hole River represents 60 percent of the total riverine miles within the DPS. Within the Big Hole River, many years of management, including 13 years of implementation of the Big Hole CCAA, have addressed many past threats, and resulted in both improvements in habitat conditions, and increases in the number of effective breeders. The best available information indicates that most populations of arctic grayling are diverse and stable both demographically and genetically. Therefore, there is currently a high level of resilience in most populations within the DPS.

The fact that the species still occupies seven out of 10 historical watersheds, and is spread across 19 populations, provides a high level of redundancy in the case of a catastrophic event. The full spectrum of life-history diversity is represented, including populations that are primarily adfluvial, and those that are primarily fluvial. While there are fewer (four) populations that have a fluvial component, there is also a high level of within-system redundancy in the Big Hole River, which includes 199 river miles of both mainstem and tributary habitat for arctic grayling, such that no single catastrophic event would be expected to impact the entire Big Hole River population; and in addition, the other three primarily fluvial systems provide additional redundancy. The presence of populations from the full spectrum of life histories, as well as the presence of moderate to high levels of genetic diversity within many populations, provides representation, or the ability to adapt to changing conditions. Therefore, after assessing the best available information, given its current levels of resiliency, redundancy, and representation, we determine that the Upper Missouri River DPS of arctic grayling is not in danger of extinction throughout all of its range.

We also considered the viability of the DPS into the foreseeable future. With the potential exception of climate change and drought, for most of the potential threats we evaluated, there is no information to indicate that the magnitude of the threat or impacts on arctic grayling will increase in the future. With respect to climate change, as discussed above, we have information projecting trends in future global average surface temperature change from simulations using multiple climate models (IPCC 2014, p. 11). These predictions begin to diverge rapidly around the year 2050, as evidenced by the bounds of uncertainty not overlapping the estimates from the most conservative and least conservative emissions pathways (IPCC 2014, p. 11). We also have predictions of future regional (Northwest U.S., including most of the Upper Missouri River basin) water temperatures in the 2080s (Isaak et al. 2015) and more specifically, water temperature predictions for the Big Hole River in the 2040s and 2060s (Vatland 2015). We have also seen a slow increase in non-native brown trout moving upstream through time, although we have no specific projections or timeframes associated with this stressor into the future.

Despite projected increases in temperature and frequency of drought, as described above (see *Factor E. Other Natural or Manmade Factors Affecting Its Continued Existence*), fifteen out of nineteen populations in the DPS are currently in lake habitats that will likely not be affected significantly by climate change due to their high elevation, intact riparian areas, and cool inputs of tributary water. Riparian restoration, particularly in the Big Hole River, has been empirically shown to minimize the effects of increasing water temperatures due to climate change. In the future, we do not expect habitat to decline in the Big Hole River because of the proven track record of CCAA projects. These protections are expected to persist into the future and maintain the integrity of the habitat. Most of the other populations of Arctic grayling reside in high-quality habitats on Federal land where mechanisms exist to conserve that habitat. With respect to non-native fish, we expect that impacts to arctic grayling populations will be low, as non-natives have co-existed with some lake populations for many decades. Brown trout have also entered the Big Hole River CCAA area, despite which, record numbers of Arctic grayling young-of-the-year have been recorded and effective number of breeders has increased. Given the lack of stressors that are projected to occur in the future, as well as the projected continued resilience of most populations within the DPS, we expect that levels of redundancy and representation will also be maintained into the future. Therefore, we find that the Upper Missouri River DPS of Arctic grayling is not likely to become an endangered species within the foreseeable future throughout all of its range.

Thus, after assessing the best available information, we conclude that the Upper Missouri River DPS of arctic grayling is not in danger of extinction throughout all of its range nor likely to become so within the foreseeable future.

Status Throughout a Significant Portion of Its Range

Under the Act and our implementing regulations, a species may warrant listing if it is in danger of extinction or likely to become so in the foreseeable future throughout all or a significant portion of its range. Having determined that the Upper Missouri River DPS of arctic grayling is not in danger of extinction or likely to become so in the foreseeable future throughout all of its range, we now consider whether it may be in danger of extinction or likely to become so in the foreseeable future in a significant portion of its range. The range of a species can theoretically be divided into portions in an infinite number of ways, so we first screen the potential portions of the species' range to determine if there are any portions that warrant further consideration. To do the "screening" analysis, we ask whether there are portions of the species' range for which there is substantial information indicating that: (1) the portion may be significant; and, (2) the species may be, in that portion, either in danger of extinction or likely to become so in the foreseeable future. For a particular portion, if we cannot answer both questions in the affirmative, then that portion does not warrant further consideration and the species does not warrant listing because of its status in that portion of its range. Conversely, we emphasize that answering both of these questions in the affirmative is not a determination that the species is in danger of extinction or likely to become so in the foreseeable future throughout a significant portion of its range—rather, it is a step in determining whether a more detailed analysis of the issue is required.

If we answer these questions in the affirmative, we then conduct a more thorough analysis to determine whether the portion does indeed meet both of the "significant portion of its range" prongs: (1) the portion is significant and (2) the species is, in that portion, either in danger of extinction or likely to become so in the foreseeable future. Confirmation that a portion does indeed meet one of these prongs does not create a presumption, prejudgment, or other determination as to whether the species is an endangered species or threatened species. Rather, we must then undertake a more detailed analysis of the other prong to make that determination. Only if the portion does indeed meet both prongs would the species warrant listing because of its status in a significant portion of its range.

At both stages in this process—the stage of screening potential portions to identify any portions that warrant further consideration and the stage of undertaking the more detailed analysis of any portions that do warrant further consideration—it might be more efficient for us to address the "significance" question or the "status" question first. Our selection of which question to address first for a particular portion depends on the biology of the species, its range, and the threats it faces. Regardless of which question we address first, if we reach a negative answer with respect to the first question that we address, we do not need to evaluate the second question for that portion of the species' range.

For the Upper Missouri River DPS of arctic grayling, we identified two potential portions of the range to screen to see if they warranted further consideration as potential significant portions of the range. First, we look for any portions of the range that could potentially be considered "significant". We identified a grouping of the four populations with a fluvial component (Big Hole, Centennial, Madison, and Ruby Rivers) as a potential portion to consider. Given the contributions of these populations as a group to representation (as they contain the only populations with a fluvial component on the spectrum of life histories within the DPS), they may be an important source of adaptive capacity for the DPS as a whole. Therefore, this portion of the range may potentially be considered significant. However, we then consider whether Arctic grayling may be, in that portion, either in danger of extinction or likely to become so in the foreseeable future. We found that there is sufficient redundancy within the primarily fluvial systems to protect from catastrophic events, as there are four river systems within this group that are separated by tens to hundreds of miles. There is also substantial redundancy within the Big Hole River population and Centennial Valley population, in the form of multiple, occupied tributaries and mainstem habitats. The Big Hole River and Centennial Valley have higher levels of resiliency than the other two populations, and also collectively represent 83 percent of total river miles. The Big Hole River tributaries have been empirically shown to be resilient to climate change as the result of riparian shading and stream restoration [Vatland 2015, pp. 49-50, 56; McCullough 2019, pp. 5, 7-8; for a full discussion, see Climate change (under Factor A) and *Cumulative Effects from Factors A through E*]. Durations of stressful water temperatures in the mainstem Big Hole River have been empirically shown to decline since the initiation of the CCAA (MFWP 2019, unpublished data; for a full discussion, see Dewatering From Irrigation and Increased Water Temperatures). The tributaries in the Centennial Valley and the mainstem Ruby River and its tributaries are similar in size to some of the tributaries in the Big Hole River and intact riparian areas in these systems are expected to have similar effects on buffering against climate change [for a full discussion, see Climate change (under Factor A) and *Cumulative Effects from Factors A through E]*. Indeed, empirical evidence shows that these systems have cooler water temperatures that rarely exceed the stress threshold for Arctic grayling (Mogen 1996, p. 44; MFWP 2015, unpublished data; MFWP 2019, unpublished data; USGS 2019, unpublished data; for a full discussion, see Dewatering From Irrigation and Increased Water Temperatures)). Within the Big Hole, Centennial Valley, and Ruby River populations, habitat has been stable or improving, and there are few threats of concern. While the Madison River population continues to have a low abundance of Arctic grayling, and higher densities of nonnative trout than the other populations with a fluvial component, the Madison River represents only five percent of the total miles of occupied Arctic grayling river habitat. Therefore, the populations with a fluvial component, as a group, are not in danger of extinction or likely to become so in the foreseeable future, and thus do not warrant further consideration as a significant portion of the range.

We also look for any portions of the range that may be in danger of extinction or likely to become so in the foreseeable future. To conduct this screening, we consider whether the threats are geographically concentrated in any portion of the species' range at a biologically meaningful scale. We identified the Madison River as a potential portion to consider. Past surveys have indicated a declining population in the Madison River (Clancey and Lohrenz 2009, pp. 30, 74), and Arctic grayling in the Madison River continue to persist at a low abundance. The factors influencing the current demographics of this population are unclear, although historical reservoir drawdowns likely affected this population and its habitat in the past. In addition, densities of nonnative trout in the Madison River are substantially higher than in other systems occupied by Arctic grayling, so the potential effects of nonnatives on Arctic grayling recruitment are a concern, and may be contributing to the decline of this population. Therefore, this portion of the range may potentially be considered in danger of extinction or likely to become so in the foreseeable future. However, we then consider whether arctic grayling may be, in that portion, considered significant. The Madison River contains only 15 miles of habitat occupied by Arctic grayling, representing five percent of the total miles of occupied arctic grayling river habitat. Given its low abundance, this population likely has low resiliency, and while it is one of four primarily fluvial populations, it is such a small proportion of fluvial habitat for the species that its contributions to redundancy and representation are small. Therefore, there is not substantial information that the Madison River population may be significant, and thus this portion does not warrant further consideration as a significant portion of the range.

Therefore, for the Upper Missouri River DPS of arctic grayling, we conclude, based on this screening analysis, that no portions warrant further consideration through a more detailed analysis, and the species is not in danger of extinction or likely to become so in the foreseeable future in any significant portion of its range. Our approach to analyzing significant portions of the species' range in this determination is consistent with the courts' holdings in *Desert Survivors* v. *Department of the Interior*, No. 16-cv-01165-JCS, 2018 WL 4053447 (N.D. Cal. Aug. 24, 2018); *Center for Biological Diversity v. Jewell*, 248 F. Supp. 3d , 946, 959 (D. Ariz. 2017); and *Center for Biological Diversity v. Everson*, 2020 WL 437289 (D.D.C. Jan. 28, 2020).

Determination of Status

Our review of the best available scientific and commercial information indicates that the Upper Missouri River DPS of arctic grayling does not meet the definition of an endangered species or a threatened species in accordance with sections 3(6) and 3(20) of the Act. Therefore, we find that listing the Upper Missouri River DPS of arctic grayling is not warranted at this time.

References Cited

A complete list of references cited in this document are available on the Internet at *http://www.regulations.gov* at Docket No. FWS–R6–ES–XXXX–XXXX and upon request from the Montana Ecological Services Office (see **FOR FURTHER INFORMATION CONTACT**).

Authors

The primary authors of this document are the staff members of the Montana Ecological Services Office and the Mountain-Prairie Regional Office.

Authority

The authority for this action is the Endangered Species Act of 1973, as amended (16 U.S.C. 1531 *et seq.*).

APPROVAL/CONCURRENCE: Lead Regions must obtain written concurrence from all other Regions within the range of the species before recommending changes, including elevations or removals from candidate status and listing priority changes; the Regional Director must approve all such recommendations. The Director must concur on all resubmitted 12-month petition findings, additions or removal of species from candidate status, and listing priority changes.

Approve: ____________________________________
Regional Director, Fish and Wildlife Service Date

Concur: ____________________________
Director, Fish and Wildlife Service Date

Do not concur: ____________________________
Director, Fish and Wildlife Service Date

Director's Remarks:

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APPROVAL/CONCURRENCE: Lead Regions must obtain written concurrence from all other Regions within the range of the species before recommending changes, including elevations or removals from candidate status and listing priority changes; the Regional Director must approve all such recommendations. The Director must concur on all resubmitted 12-month petition findings, additions or removal of species from candidate status, and listing priority changes.

Approve: ______________________________ March 5, 2020
Regional Director, Fish and Wildlife Service Date

Concur: ______________________________ June 29, 2020
Director, Fish and Wildlife Service Date

Do not concur: ______________________________
Director, Fish and Wildlife Service Date

Director's Remarks: