



NATURAL RESOURCES DEFENSE COUNCIL

*By Electronic Submittal and Overnight Mail*

November 28, 2008

Public Comment Processing  
Attention: RIN 1018-AW37  
Division of Policy and Directives Management  
U.S. Fish and Wildlife Service  
4401 North Fairfax Drive, Suite 222  
Arlington, VA 22203

**Re: Designation of the Northern Rocky Mountain Population of Gray Wolf as a Distinct Population Segment and Removing This Distinct Population Segment From the Federal List of Endangered and Threatened Wildlife**

Dear Madam/Sir:

On behalf of the Natural Resources Defense Council (“NRDC”) and our more than 475,000 members, we submit the following comments on the U.S. Fish and Wildlife Service’s (“Service”) Proposed Rule to designate the Northern Rocky Mountain population of gray wolf (*Canis lupus*) as a Distinct Population Segment (“DPS”) and to remove it from the list of threatened and endangered species. See “Designation of the Northern Rocky Mountain Population of Gray Wolf as a Distinct Population Segment and Removing This Distinct Population Segment From the Federal List of Endangered and Threatened Wildlife,” 73 Fed. Reg. 63,926 (Oct. 28, 2008) (“Proposed Rule”). These comments supplement the comments submitted to you on our behalf by Earthjustice.

We urge the Service to abandon the Proposed Rule. The delisting of the Northern Rocky Mountain gray wolf population is premature, and supported neither by currently available scientific information nor law. In particular, the Proposed Rule relies on recovery criteria that were not scientifically supportable when they were developed and are badly out of date today. Rather than a meta-population of at least 30 breeding pairs comprising 300+ wolves, the best available science indicates that a population in the thousands is required for wolves in the region to be deemed fully recovered. Further, the Service’s proposed interpretation of genetics belies well-

established scientific principles. Additionally, the Service may not, as the Proposed Rule contemplates, delist wolves in Montana and Idaho while simultaneously concluding that wolves in Wyoming are not yet recovered and ready for delisting. The Endangered Species Act (“ESA”), 16 U.S.C. §§ 1532, et seq., does not permit the Service to delist a portion of a species while that species remains endangered in a significant portion of its range. Nor can the Service maintain the Wyoming wolf population’s designation as a “nonessential experimental” population under section 10(j) of ESA, as Wyoming wolves are not “wholly separate geographically” from other wolf populations. 16 U.S.C. § 1539(j)(2).

Finally, we question the timing of the Proposed Rule. The Proposed Rule was published just months after a federal court struck down the Service’s previous delisting of this population. Defenders of Wildlife v. Hall, 565 F.Supp.2d 1160 (D. Mont., 2008). The current proposal seems more intent on rushing a delisting decision rather than an attempt to cure the legal defects found by the court and seriously reevaluate the agency’s approach to conserving wolves in the region. This is particularly true given that, as the Proposed Rule itself acknowledges, “[t]he only ‘official’ annual wolf population statistics are provided in the interagency annual report, which is normally available in March each year.” 73 Fed. Reg. at 63,928. Official statistics are generally compiled in the spring to allow scientists to take into account the toll that winter may have taken on wolf population levels. Indeed, this year has seen an unusually high level of mortality among the region’s wolves.<sup>1</sup> The Service should therefore withdraw the Proposed Rule and if it insists on proposing delisting, respond to the 2008 year-end official annual wolf population statistics in a new Proposed Rule.

## DISCUSSION

### **I. The Service Should Revise Its Recovery Goals for the Northern Rocky Mountain Population and Prepare a National Recovery Plan for the Gray Wolf**

As the Proposed Rule acknowledges, one of the key issues that must be addressed before the Northern Rocky Mountains population can be delisted is the appropriateness of continuing to rely on the Service’s “recovery goals” for the population. 73 Fed. Reg. at 63,929-30. While the Proposed Rule attempts to limit this question to exploring options for “maintaining or increasing genetic diversity in the NRM wolf population,” *id.* in fact, the ESA requires the Service to reassess its recovery goals for the population in its entirety.

---

<sup>1</sup> See Brett French, “Wolf pup numbers down; disease may be responsible,” *Billings Gazette* (Oct 23, 2008) (available at: <http://www.billingsgazette.net/articles/2008/10/23/news/state/30-wolf.txt>) (quoting Douglas Smith, National Park Service wolf biologist that the mortality level “is a cause for concern, especially with delisting. Wolf numbers region wide will be down this year.”); *Wyoming Wolf Report* (Nov. 10 - Nov. 21, 2008) (“Mange has been documented in >8 wolves from four different packs (Oxbow Creek, Mollies, Leopold, and one unnamed group of 4 wolves).”).

Section 4 of the ESA is clear that the Service may only add or remove a species from the list of threatened and endangered species based “solely on the basis of the best scientific and commercial data available to [it] after conducting a review of the status of the species.” 16 U.S.C. § 1533(b)(1)(A). Here, the Service is proposing to both list a new “species” (the Northern Rocky Mountains DPS) and then remove that species from the Act’s protections.<sup>2</sup> These determinations therefore must be solely justified by the best available science. In addition, Section 4 of ESA requires the Service to prepare a recovery plan for all “endangered species and threatened species” protected by the Act. 16 U.S.C. § 1533(f)(A).<sup>3</sup>

The Proposed Rule bases its proposed delisting on “recovery criteria” that call for establishment of at least 30 breeding pairs of wolves in three areas comprising a meta-population of at least 300 animals with genetic exchange between subpopulations. The core of these numeric targets—the requirement of 30 breeding pairs of wolves—originate with the Service’s 1987 recovery plan for the formally recognized wolf subspecies, *Canis lupus irremotus* (which roughly corresponds to the Northern Rocky Mountains population), as subsequently reevaluated by the Service in a 1994 Environmental Impact Statement on wolf reintroduction. Thus, the Service’s recovery criteria, largely relies on a recovery plan that is over twenty years old and an EIS that is nearly fifteen years old. The scientific understanding of minimum population viability, conservation genetics, and wolf ecology has significantly advanced since these criteria were established. Moreover, even setting aside these advances, the recovery criteria themselves were reached through a methodologically flawed process and were not supported by the scientific literature or opinion available to the Service at the time of their creation. Accordingly, if the Service wishes to delist the Northern Rocky Mountains population of gray wolves, it must completely reexamine its recovery criteria in light of the best scientific information now available.

---

<sup>2</sup> It is not clear that the Service has the authority to list a DPS solely for the purpose of delisting it. Recently, a court held that such authority is not plain from the face of the ESA and the Service has not done an adequate job explaining its belief that the statute gives it such power. Humane Society of the United States v. Kempthorne, Civil Action No. 07-0677 (PLF) (Sept. 29, 2008).

<sup>3</sup> Recovery plans prepared under the Act must contain the following elements: (1) a description of “site-specific management actions that may be necessary to achieve the plan’s goal” for the recovery of the species; (2) “objective, measurable criteria” which, when met, would result in an initial determination that delisting of the species may be appropriate; and (3) an estimate of the “time required and cost to carry out those measures” needed to achieve the plan’s recovery goals. Id. at §1533(f)(B). Unless the Service finds that preparing such a plan “will not promote the conservation of the species,” the obligation to prepare a recovery plan is mandatory. 16 U.S.C. § 1533(f). See also Southwest Center for Biological Diversity v. Bartel, 470 F.Supp.2d 1118 (S.D. Cal. 2006); Environmental Defense Center v. United States Department of the Interior, Case No. 99-9042, at 9 (C.D. Cal. May 20, 2001); Sierra Club v. Lujan, 1993 WL 151353, \*11 (W.D. Tex. 1993).

That science shows that a recovery goal of at least 300 wolves in 30 breeding pairs is grossly inadequate to reach minimum population viability for this population.<sup>4</sup>

A. The Listing of the Gray Wolf and the NRM Recovery Planning Process.

In January of 1974, shortly after the Endangered Species Act was overwhelmingly passed by Congress, two subspecies of gray wolves were listed as endangered: the eastern timber wolf (*C. l. lycaon*) and the Northern Rocky Mountain wolf (*C. l. irremotus*). See “Endangered Wildlife Lists,” 39 Fed. Reg. 1,171 (January 4, 1974). Two additional subspecies, the Mexican gray wolf (*C. l. baileyi*), and the Texas gray wolf (*C. l. monstrabilis*), were listed as endangered in April and June of 1976. See “Determination that Two Species of Butterflies are Threatened Species and Two Species of Mammals are Endangered Species (Schaus swallowtail; Bahama swallowtail; Mexican wolf, *Canis lupus baileyi*; gray bat, *Myotis grisescens*),” 41 Fed. Reg. 17,742 (April 28, 1976); “Endangered Status for 159 Taxa of Animals,” 41 Fed. Reg. 24,062 (June 16, 1976).

In 1978, due in part to uncertainty of these subspecies designations, the Service listed the gray wolf as endangered at the species level (*Canis lupus*) throughout the conterminous 48 states and Mexico, except for Minnesota, where the gray wolf was classified as threatened. See “Reclassification of the gray wolf in the United States and Mexico, with determination of critical habitat in Michigan and Minnesota,” 43 Fed. Reg. 9,607 (March 9, 1978). Despite the listing of the species as a whole, the Service has continued to manage the original subspecies separately, among other things developing recovery plans for individual subspecies. In 1978, the Service completed a recovery plan for the eastern timber wolf in the Northeast/Great Lakes region (USFWS 1978) and in 1982 the Service developed a recovery plan for the Mexican wolf in the Southwest (USFWS 1982).

In 1980, the Service established a recovery plan for *Canis lupus irremotus* and then revised that plan in 1987. The plan called for establishing a minimum of 10 breeding pairs, in each of three separate areas for three successive years (USFWS 1987), thus comprising a total of 30 breeding pairs in the region. On November 22, 1994, areas of Idaho, Montana and Wyoming were designated as nonessential experimental population areas for the gray wolf under section 10(j) of the ESA and wolves were reintroduced to the region in two separate areas. See “Establishment of a Nonessential Experimental Population of Gray Wolves in Yellowstone National Park in Wyoming, Idaho and Montana,” 59 Fed. Reg. 60,252 (November 22, 1994); “Establishment of a Nonessential Experimental Population of Gray Wolves in Central Idaho and Southwestern Montana,” 59 Fed. Reg. 60,266 (November 22, 1994).

---

<sup>4</sup> Much of the information in these comments is taken from a Petition, filed by NRDC and the Defenders of Wildlife with the Service, to prepare a Recovery Plan for the Gray Wolf. See NRDC and Defenders of Wildlife, “A Petition to Prepare A Recovery Plan Under the Endangered Species Act for the Gray Wolf,” (February 2007). A copy of the Petition is provided with this comment letter.

As part of the reintroduction process, the Service evaluated the recovery goals set out in its 1987 recovery plan in a 1994 Environmental Impact Statement (EIS) and again in 2001.<sup>5</sup> The Service concluded that 30 breeding pairs, comprising at least 300 wolves in a metapopulation with genetic exchange between subpopulations, for three successive years, are needed for recovery. See 72 Fed. Reg. 6,106. These recovery targets do not form an adequate scientific basis for the recovery of the Northern Rocky Mountains wolf population.

B. Multiple Connected Populations of Wolves With Thousands of Individuals Are Required to Achieve Regional Recovery

It is a well-established principle of conservation biology that populations of organisms need substantial and robust numbers of individuals to maintain viability. An often cited estimate for minimum population viability (MPV) is an effective population size ( $N_e$ ) of 500 individuals to avoid the effects of genetic loss due to drift (Soule and Wilcox 1980, Frankel and Soule 1981, Soule 1986, Franklin and Frankham 1998). For these reasons, Soule and Simberloff (1986) concluded that “estimates of MVPs for many animal species are rarely lower than an *effective size* of a few hundred.” Since effective population sizes are generally only 10-20% of the census population, this lower limit translates into a total population count of 2,500-5,000 individuals (Frankham 1995, Palstra and Ruzzante 2008).

Other estimates have predicted that viable population numbers should be even higher. For example, Lande (1988) criticized the application of a blanket number like  $N_e=500$  because it fails to consider critical species-specific demographic data. Lande then outlined examples in which demographic parameters, such as an allee effect, stochasticity, edge effects or local extinctions in a patchy habitat, could require populations to have even larger numbers than an effective population of 500. Lande (1995) further explored this topic in the context of genetic variation and mutation and concluded that effective populations should number in the 5,000s. C. D. Thomas (1990) also estimated that MVPs should number in the thousands – ideally, 10,000 individuals for populations that experience fluctuations. Similarly, in 2004, Reed and Hobbs examined the population viability of 2,387 populations of 203 species and found that vertebrates need to number in the thousands for effective conservation.

Recently, a number of studies have been published that examine population viability based on empirical data and gray wolves specifically. Brook et al. (2006) estimated the MVP for 1,198 species including the gray wolf and found that the median overall estimate was 1,377 individuals. Traill et al. (2007) conducted a meta-analysis of MVPs for 212 species including gray wolves and concluded that the MVP for most species will exceed a few thousand individuals. Finally, Reed et al. (2003) estimated

---

<sup>5</sup> Bangs, E.E., *Wolf population viability peer review – draft summary*, U.S. Fish and Wildlife Service, Ecological Services (2002), 9 pp.

the minimum viable population size for over 100 vertebrate organisms, including the gray wolf. The MVP for *adult* gray wolves was estimated at 1,403. When Reed et al (2003) corrected for 40 generations worth of data, the MVP for gray wolves was estimated to be 6,332.

Moreover, genetic data shows that wolves in the United States historically numbered in the several hundreds of thousands (Leonard et al. 2005) and that the genetic diversity of the extirpated North American gray wolves was twice that of the current population. Therefore, the current assemblage of gray wolves in the lower-48 states is a profound under-representation both numerically and genetically of the original gray wolves that once occupied this landscape.

Current scientific literature is thus clear that a population of 300 wolves, including 30 breeding pairs, is simply inadequate to achieve minimum population viability. That was true even based on the published scientific literature available to the Service at the time these targets were set, and is even clearer today. They therefore cannot be relied upon by the Proposed Rule as the basis for delisting.

C. The Service's 1987 Recovery Plan's goals lacked scientific basis and justification, and the Service's 1994 evaluation of these goals ignored the best available science.

As discussed above, the demographic recovery goal established by the 1987 recovery plan was 10 breeding pairs living in each of three separate areas for at least three consecutive years (USFWS 1987). This basic paradigm—30 breeding pairs of wolves distributed throughout three locations—still forms the fulcrum of the Service's recovery criteria today. Unfortunately, the 1987 Recovery Plan provides little justification for these goals, merely stating that they “were developed based on the most current information and the opinions of recovery team members, other ‘experts’ on the species, and the Fish and Wildlife Service” (USFWS 1987, p.19). However, the plan does not include any presentation of scientific literature to support the proposed recovery goal, nor does it outline which “experts” were consulted or the process by which the recovery goals were established.

The recovery goals set by the 1987 NRM Recovery Plan were reevaluated by the Service in 1994 for the EIS prepared in connection with the introduction of an experimental population of wolves into the region (USFWS 1994: Appendix 9). After this evaluation, the Service reaffirmed its recovery criteria for the region: a minimum of 30 breeding pairs in three separate locations over a three-year period. This evaluation was also published as an article.<sup>6</sup> The 1994 reevaluation included three elements: (1) a review of the scientific literature on minimum viable population size; (2) the results of a survey, or opinion poll, of wolf biologists on proposed recovery

---

<sup>6</sup> Fritts, S. H. and L. N. Carbyn. 1995. Population viability, nature reserves and the outlook for gray wolf conservation in North America. *Restoration Ecology* 3: 26-38.

goals; and (3) a presentation of case histories of small, isolated wolf populations that appear to be self-sustaining. A close examination reveals, however, that each part of this reevaluation either provided little support for the Service's recovery goal or clearly indicated that a minimum population size far above those established by the 1987 recovery plan was needed.

## 1. Review of literature

First, a review of the literature available in 1994 clearly demonstrated that a far larger population was needed in order to achieve recovery. Indeed, as Fritts' (USFWS 1994: Appendix 9) review itself acknowledged, then-current scientific literature showed that long-term viability for wolf populations would require an effective population size ( $N_e$ ) of at least 500 and up to the low thousands of individuals (p.38-39). Yet Fritts dismissed this finding as unachievable stating, "Clearly, finding an area to support  $N_e = 500$  of wolves in the lower 48 states is very unlikely, as this would equate to a total population in the low thousands" (USFWS 1994: Appendix 9, pg. 38). Similarly, in their subsequent article, Fritts and Carbyn (1995) conclude, "it is clear that finding any totally protected reserves that could support an  $N_e$  of 500 wolves in the lower 48 states or Canada would be very difficult – much less any additional such reserves." They further state, "If (thousands of wolves) were the case, long-term conservation for many wolf populations in a specific regional setting would be impossible."

The Service's own evaluation of the recovery goal identified in the 1987 recovery plan thus clearly finds that the plan's target population (10 breeding pairs in three locations, for a total of 30 breeding pairs) was simply not supported by the available scientific literature, yet the Service simply discounted this science believing (erroneously) that maintaining a population of thousands of wolves in the region would be unachievable.

## 2. Survey of biologists

Second, the Service's survey of wolf biologists also provided little support for the proposed recovery goal. Fritts (USFWS 1994: Summarized in Appendix 9) surveyed the opinions of wolf biologists to determine whether they thought that the Service's recovery goal constituted a viable population (see also Fritts and Carbyn 1995). The survey did not provide a definition of viability, leaving a critical element to the discretion of the evaluator. Additionally, the survey presented the biologists with the Service's arbitrary recovery goals, rather than soliciting the biologist's own definition of recovery. Furthermore, the survey was designed in a way that likely biased support for the Service's pre-established goal.

The survey began by asking if even *fewer* wolves than proposed in the 1987 recovery goals would constitute a viable population. That is, the survey first asked whether one group of 10 breeding pairs would constitute a viable population. This question was followed by asking whether three groups of 10 breeding pairs acting in a

metapopulation would constitute a viable population. Predictably, many of the answers indicated that the second definition of three groups would be “more viable” (Mike Phillips, Kyran Kunkel), “more defensible” (John Weaver), or would have “more chance of constituting a viable population” (John Theberge), than the first definition. Several of these same respondents, however, were critical of either definition, indicating that many responses were comparative and did not necessarily indicate agreement that the definition constitutes a viable population.

Further, while some of the biologists did agree with one or the other of the definitions, many were also careful to warn that their response represented their opinion only, which was subjective since none of the definitions were based on explicit data. For example, Bob Stephenson wrote, “Unless someone has done a study of minimum viable population (MVP) of wolves from a genetic standpoint there would be no way to know for sure whether this population would sustain itself in the long term.” John Weaver responded, “In lieu of a formal PVA [population viability analysis] for gray wolves in the Northern Rocky Mountains, I can only respond subjectively to the proposed definitions.” Mark Boyce cautioned, “A definition for a viable population is arbitrary, and we do not know enough to say how many is sufficient.” Lu Carbyn advised, “I would not split hairs over what is viable or not – make sure you have large enough areas with suitable prey base....then let nature seek its own level.” Finally, Kyran Kunkel concludes, “When any of the above definitions are finally made, I think it is essential for us to realize and state that these definitions are not based on any true knowledge of what a population or viable population for wolves is but rather, mostly a guess based upon the best information available. We should be willing to change our definition as new information is obtained. These definitions should not make or break wolf recovery or reintroductions.” (USFWS 1994a) (emphasis added).

Some biologists’ responses not only were subjective, but were affected by their perception of agency motivations. For example, the third and final question of the survey addressed the definition of a wolf population (not a viable population). Mark Boyce, who favored allowing natural dispersers to colonize the area, questioned whether the agency’s definition of a wolf population was designed to allow the 10(j) experimental population designation to move forward. He writes, “if accepting your definition would imply that a population doesn’t exist and therefore you have free reign to carry on with a release program, I would reject your definition.” By contrast, Mike Phillips indicates that he would be likely to agree to any definition of a viable population if it meant that the recovery process would move forward. After agreeing with the Service’s first definition, Phillips writes, “As a matter of fact, I’d support a revised Plan that presented smaller numbers for recovery goals...if such a revision increased the odds of getting wolves ‘on the ground.’” However, Phillips also indicates that he believes greater numbers of wolves than represented by the second definition would constitute “an even more viable population (higher probability of persistence).”

Other reviewers disagreed with either of the definitions. Jim Peek responded, “Aren’t these numbers a bit *low* and the *time interval too short?*” (emphasis in original). John Theberge wrote, “30 breeding pairs is still well below the 1% rule which I believe is overly low itself. I think this population size is still too marginal to be considered viable. These definitions are inadequate.” Mike Nelson responded, “There also appears to be agreement that ‘several hundreds’ of breeders are needed to ensure long-term evolutionary potential. The common value in the literature is  $N_e = 500$  and that translates into the low thousands for a population size in wolves. By this criterion, the individual wolf populations as well as their metapopulation would not be evolutionarily viable.”

### 3. Case histories

Third, the “case histories” used by the Service to support the recovery plan’s goal were flawed. In their review, Fritts and Carbyn (1995) present a series of examples of small, isolated populations of wolves in order to make the case that a wolf population below that supported by the existing scientific literature could, in fact, persist.<sup>7</sup> As discussed in more detail below, many of the populations presented as case studies had not been isolated for more than 20-30 years and some of them experienced wide fluctuations in numbers, including an extinction event on Coronation Island, Alaska, and a population crash of 75% on Isle Royale, Michigan in the early 1990s. Furthermore, most of the populations evaluated are either free of human persecution or occur in culturally and geographically different settings than the Rocky Mountains. None of the case histories lend any support to the notion that a population of 30 breeding pairs of wolves in the Rocky Mountain region would be viable over a significant time period.

In short, the Service’s 1994 evaluation provided little scientific justification for setting recovery criteria of 30 breeding pairs of wolves within a meta-population of 300 individuals. Indeed, if anything, the 1994 evaluation identified the need for thousands of wolves in order for recovery to be achieved.

D. The Service’s 2001 survey of its recovery goals identified significant scientific objections to its recovery criteria and do not provide any scientific justification for the criteria.

The Service evaluated the recovery goals for a second time in 2001 through another opinion survey of biologists. See Bangs, E.E., *Wolf population viability peer review – draft summary*, U.S. Fish and Wildlife Service, Ecological Services (2002), 9 pp. While the Service referred to this evaluation as a “peer review,” the survey, or opinion poll, again identified significant scientific disagreement with variations of the

---

<sup>7</sup> These case histories were: Isle Royale National Park, Michigan; Riding Mountain National Park, Manitoba; Jasper National Park, Alberta; Kenai Peninsula, Alaska; Alexander Archipelago, Alaska; Coronation Island, Alaska; various European populations in Italy, Spain, Poland, and Scandinavia.

Service's recovery goal of 30 breeding pairs in three separate recovery areas for three successive years. The one modification to the recovery criteria presented in the 2001 survey and ultimately embraced by the Service, was to require that the 30 breeding pairs of wolves be embedded within a total meta-population of 300 wolves in three locations, with genetic exchange between those locations. Id.

In its 2001 survey, biologists were presented with three alternative definitions of a viable population and asked to rank the definitions in order of viability. The three definitions were all slight variations of the Service's goal of 30 breeding pairs in three separate areas for three successive years. Reviewers were also offered a fourth possibility of creating their own definition. A number of reviewers were highly critical of the survey's methodology. One reviewer described the presentation as "artificial and misleading" (Reed Noss). Another reviewer noted, "By limiting the choices to those 3 options approved by the Service, plus a category of 'other', it may unfairly bias the results" (Brian Miller). As other reviewers pointed out, viability and introduction are not the same as recovery.

The survey also identified considerable dissent from a number of scientific experts. A common criticism was that the proposed recovery goals had no scientific basis. In fact, Brian Kelly, a U.S. Fish and Wildlife Service employee, was asked to review the recovery plans and concluded that "a definition of viability without quantify-able [sic] data to back it up is problematic and will be difficult to defend because it is subjective...Some reasonable attempt to model the dynamics of the N. Rockies population showing that 30 (breeding pairs)/300 (wolves) has a reasonable expectation to persist, is needed." Kelly continued by pointing out, "Survival/mortality rates, age at first breeding, fluctuations in prey numbers, among other factors, should be incorporated into the determination of whether a population is viable." He concluded that, "in the absence of such a quantitative assessment, it is subjective and conjectural to simply interpret 30/300 as meeting...population viability."

This sentiment was reflected in a number of other reviews. Mark Shaffer and Martin Smith noted that, "Despite the intense study wolves have received in this region, and the wealth of population data that must be available to the Service, the Service has presented no quantitative modeling of the dynamics of the existing populations...Such a modeling effort is essential to gauge the relative worth, from a population viability perspective, of the various definitions you have asked us to consider." Another reviewer, Robert Taylor, wrote, "The fact that the Fish and Wildlife Service has not had the vision to support such a (spatially explicit, individual-based) modeling exercise is not sufficient reason to force me to make wild guesses about the parameters of viability." Brian Miller noted, "None of the definitions offered by the Service is calibrated from the probability, length of time, or specific conditions of survival by 30 breeding pairs of wolves. Unless we are given such information, we are being asked to choose among three 'black boxes.'" Reed Noss wrote, "Viability is relative, not strictly yes or no...One must consider population growth rates, spatial distribution, and source-sink dynamics, among other factors...The recovery area and population goals need to be expanded." Yet another reviewer noted that, "It may be

generally inappropriate to conduct an opinion poll, even from experts, when no quantitative analyses have been conducted to assess the issues at hand” (John Vucetich).

In addition to excluding life history data, a number of reviewers noted that genetic problems were likely to become an issue without greater attention to connectivity. Gordon Haber noted that the proposed definitions “ignore underlying qualitative – behavioral and genetic – aspects of population biology.” Fred Allendorf reviewed the recovery plans with his conservation genetics class and concluded that “the recovery goal of at least 300 wolves is too small to avoid genetic problems in the foreseeable future....Therefore, a population of this size should not be considered [sic] to be ‘recovered’....Thus, the recovery criteria need to require some gene flow into this population.” Dan Pletscher ranked the proposed plans, but added, “Without connectivity to Canada, this is unlikely to be viable.”

Dale Seip, a wildlife ecologist, noted during his review that, “Presumably, delisting is not going to result in some rampant slaughter of wolves. It would be useful to state the management consequences of delisting the species....If conditions have been suitable for wolves to increase over the past few years, so long as those conditions do not drastically change, there is no major risk in delisting. However, if delisting would lead to drastic changes then there would be concern” (USFWS 2002). And finally, the Service’s western gray wolf recovery coordinator, Ed Bangs, who implemented and evaluated the 2001 survey recently said about the recovery goal, “I, personally, think it is too low” (Morell 2008). Like the Service’s 1994 survey, or opinion polls, its 2001 survey thus identified significant scientific dissent and objection to the recovery criteria now being used in the Proposed Rule.

Clearly, the science of population viability estimates has advanced since the development of the Northern Rocky Mountain gray wolf recovery plan in 1987 and its reevaluation in 1994 and 2001. Although estimates at the time the recovery plans were crafted already pointed to the need for a larger number of wolves, developments since this time solidify the scientific conclusion that recovery goals for wolves should number in the thousands rather than the hundreds. Therefore, the Service’s recovery criteria for the Northern Rocky Mountain wolf is not consistent with the best available science and needs to be revised before this population may be delisted. In the Proposed Rule the Service notes that one option it will be considering includes “maintaining higher rather than lower overall wolf numbers in all or select recovery areas.” 73 Fed. Reg. at 63,930. Current science indicates that, at a minimum, a viable population size of at least 2,500 - 5,000 individuals in at least three interconnected populations is required in the Northern Rocky Mountains. Independently viable or connected populations should also be established in the Colorado/Utah area and in Oregon and Washington before this region can be considered to be recovered.

## **II. The Proposed Rule’s Assessment of “genetics relative to recovery criteria” Dismisses Fundamental and Well-Established Scientific Principles**

The Proposed Rule dismisses “future theoretical inbreeding” as a possible threat to wolves in the Northern Rocky Mountains. Yet the isolation of small groups of wolves, leading to the loss of genetic diversity and/or an increase in inbreeding, is a well-documented phenomenon (e.g. Liberg et al. 2005, Bensch et al. 2006, Raikonen et al. 2006, Raikonen et al. In review). Despite this established science, along with specific predictions that inbreeding is likely to become a problem with wolves in Yellowstone National Park (vonHoldt et al. 2007) without further recovery measures, the Proposed Rule states that it is the Service’s “professional judgment” that even in the event of complete isolation of a subpopulation of wolves within Northern Rocky Mountains, any loss of genetic diversity would not threaten the population. 73 Fed. Reg. at 63,927. The Proposed Rule makes three arguments to support this judgment: (a) other small, isolated populations of wolves have survived in the past; (b) wolves practice inbreeding avoidance and would recruit dispersers as breeders; and (c) the introduction of just one or two new genetic lines can save a severely inbred small wolf population. All of these arguments over-simplify complicated issues and disregard evidence that either contradicts or fails to support the Service’s claims.

### **A. Small populations**

The Proposed Rule’s claim that the existence of numerous small wolf populations provides support for its contention that isolated populations of wolves are viable is largely based on a review by Fritts and Carbyn (1995). In their review, Fritts and Carbyn (1995) present a series of examples of small, isolated populations of wolves in order to argue that wolf populations in the low hundreds or less can persist in the wild.<sup>8</sup> It should be noted that, despite discussing these examples, Fritts and Carbyn (1995)’s review actually formed the basis for the Service’s modification of its recovery criteria to emphasize the importance of genetic connectivity between subpopulations (USFWS 1994). Additionally, the case studies include examples of populations that have either gone extinct, demonstrated instability in population size, or have subsequently expressed severe inbreeding.

For example, the Scandinavian wolf population has high inbreeding coefficients and significantly elevated occurrences of vertebral deformities (Liberg et al. 2005, Bensch et al. 2006, Raikonen et al. 2006). These same types of deformities have recently been documented in the wolves on Isle Royale (Raikonen et al. In review) and are likely to have fitness consequences based on the symptoms documented in domestic dogs with these deformities (Morgan et al. 1993, Morgan 2000).

---

<sup>8</sup> These case histories were: Isle Royale National Park, Michigan; Riding Mountain National Park, Manitoba; Jasper National Park, Alberta; Kenai Peninsula, Alaska; Alexander Archipelago, Alaska; Coronation Island, Alaska; various European populations in Italy, Spain, Poland, and Scandinavia.

The mere existence of wolves in small populations does not suggest that they are ecologically and evolutionarily viable. The fitness consequences of inbreeding and loss of genetic diversity in wolves (such as reduced litter size (Liberg et al. 2005) and vertebral deformities (Raikkonen et al. 2006, In review) make small populations more susceptible to demographic or environmental stochasticity (such as disease, weather, changes in prey base, survival, etc.) (Keller and Waller 2002). The interaction between inbreeding or loss of genetic diversity and these stochastic events is what leads to the endangerment of small populations of any species. In fact, the relationship between small population size and extinction risk (with or without the consideration of genetic effects) is a basic ecological principle (Ricklefs and Miller 2000). The scientific literature thus does not justify the Proposed Rule's reliance on the existence of these small populations as a basis for concluding that a small, isolated, population of wolves in the Northern Rocky Mountains is not threatened by the loss of genetic diversity.

#### B. Wolves and inbreeding avoidance

The Proposed Rule also argues that “wolves have a strong tendency to avoid inbreeding by selecting breeders based on genetic difference” (citing vonHoldt 2007) and that “future dispersers into a system experiencing some level of inbreeding would be much more likely to be selected for breeding” (citing Bensch et al. 2006). 73 Fed. Reg. at 63,929. While the vonHoldt et al. 2007 study does document inbreeding avoidance, the study also observed that heterozygosity of the Yellowstone National Park population of wolves was still within the range of a population that exhibits random mating.<sup>9</sup> Similarly, in a study of Scandinavian wolves, Bensch et al. (2006) found that heterozygous individuals were more likely to become breeders, but the authors detected only weak or no indication that the pairs that did reproduce had offspring with higher heterozygosity than if mating was random (Bensch et al. 2006, pg. 5). This suggests that while the wolves avoid mating with genetically similar individuals, their overall genetic diversity may not be much different than a population that mates randomly (i.e. does not actively avoid inbreeding) because, given a certain amount of genetic variation, random mating is, on average, sufficient to maintain heterozygosity and inbreeding avoidance and selection for heterozygous breeders does not confer much, if any, additional advantage.

Moreover, while wolves in the Scandinavian population selected heterozygous individuals as breeders, wolves in Yellowstone only chose non-relatives, but not necessarily the most heterozygous individuals (vonHoldt et al. 2007, p. 8). In fact, the authors report that their findings do “not support a bias toward matings of individuals within higher heterozygosity” as found in Bensch et al. 2006.

Finally, despite the fact that more heterozygous individuals were selected as breeders in the Scandinavian population, inbreeding continued in this population and even increased over time (Bensch et al. 2006, pg. 2, 5). In fact, because of the

---

<sup>9</sup> vonHoldt et al. 2007, Figure 2 (where the “open” population equates to random mating).

extensive inbreeding, the Scandinavian population also exhibits extensive linkage disequilibrium (when alleles at multiple loci are associated with each other more or less frequently than predicted by their individual frequencies) – a condition that facilitates heterozygosity. Therefore, the results from the Bensch et al. (2006) study may not be generally applicable to other populations of wolves (see attached statement from Staffan Bensch). Taken together, whether inbreeding avoidance in wolves actually slows or prevents the loss of diversity more than random mating is not well established and does not support the Proposed Rule’s conclusion that wolves in the Northern Rocky Mountains have recovered sufficiently to support delisting.

### C. Genetic rescue by single individuals

Finally, the Proposed Rule argues that the “introduction of just one or two new genetic lines can save a severely inbred small wolf population.” 73 Fed. Reg. at 63,929. This is incorrect. The Service’s argument here confuses the relationship between multilocus heterozygosity (MLH) and inbreeding coefficient ( $f$ ). In fact, the very examples the Service cites to support their claim (Vila et al. 2003, Liberg et al. 2005, Fredrickson et al. 2007) represent highly inbred populations whose inbreeding has not been relieved by the addition of single or few individuals.

Multilocus heterozygosity (MLH) is a measure of neutral, non-coding regions of DNA located throughout an organism’s genome. MLH has been shown in some cases to be related to fitness measures, and the assumption has been that MLH acts as a surrogate measure for inbreeding (reviewed in Balloux et al. 2004). However, research within the last several years has clearly demonstrated that multi-locus heterozygosity (MLH) is very poorly, if at all, correlated with the inbreeding coefficient ( $f$ ) (Slate et al. 2004, Balloux et al. 2004, Pemberton 2004, Bensch et al. 2006). This same research has also demonstrated that there are mechanisms other than inbreeding that would cause MLH to correlate to fitness measurements (Slate et al. 2004, Balloux et al. 2004). Consequently, MLH is not generally reflective of inbreeding. More importantly, even populations that have relatively high heterozygosity (MLH) levels and are considered diverse, may in fact be inbred (Balloux et al. 2004, Slate et al. 2004, Bensch et al. 2006).

The case of the Scandinavian wolf population provides a useful example. While the introduction of a single immigrant into this population did increase the population’s heterozygosity (MLH) (Vila et al. 2003), and the inbreeding coefficient dropped initially, it began to increase again with time, ultimately reaching greater levels than before the immigrant arrived (Liberg et al. 2005, Bensch et al. 2006). Additionally, Bensch et al. (2006) measured what they refer to as “breeding-failure equivalents” which is similar to “litter-reducing equivalents,” a measure of the reduction of litter size due to inbreeding, and they found that the value was similar to what they calculated before the immigrant arrived (Liberg et al. 2005). That is, despite the increase in heterozygosity, the Scandinavian population remained inbred with a fitness consequence of reduced reproduction.

In response to the immigration of this one individual, the population also demonstrated increased population growth (Vila et al. 2003, Mills 2007, pp. 196-197). Again, however, the observed population growth did not relieve the inbreeding depression in the population. In fact, Liberg et al. (2005) explain that the population was not “rescued” by the single immigrant. They write:

Our interpretation is that before this male arrived there was no population but just a strongly inbred family. The arrival of this newcomer allowed young wolves to find partners outside of their own family, and this sparked off a rapid initial increase, but has not prevented the succeeding inbreeding.

(Liberg et al. 2005, p. 19.) This interpretation is supported by Vila himself, who additionally emphasizes that the addition of one individual will not resolve the genetic problems of an inbred population (see attached statement by Carles Vila).

This same population of Scandinavian wolves has been shown to have a significant occurrence of vertebral deformities compared to two outbred populations of wolves (Raikonen et al. 2006). Fifty-two wolves from the small Scandinavian population that died between 1983 and 2003 were examined for vertebral deformities. 10.2% of the wolves examined showed cervical deformities compared to 0-1.3% of outbred populations. These types of deformities are likely to have fitness consequences for the wolves.

In domestic dogs, these same deformities are known to cause clinical symptoms including paresis (partial loss of movement or impaired movement) and paralysis (Raikonen et al. 2006). While Raikonen et al. 2006 did not directly relate heterozygosity or inbreeding coefficients with the individuals expressing deformities, a study of Icelandic sheepdogs found that, while the inbreeding coefficient was significantly correlated with the frequency of hip dysplasia, stMLH (standardized multi-locus heterozygosity) was not (Olafsdottir and Kristjansson 2008). Similarly, a data-intensive study that explored the relationship between MLH and the inbreeding coefficient ( $f$ ) in sheep found that  $f$ , but not MLH, detected evidence of inbreeding depression for morphological traits (Slate et al. 2004). These studies provide further support that the Scandinavian population of wolves not only was not “rescued” by a single immigrant, but continues to suffer from significant inbreeding depression.

The Proposed Rule’s other example of genetic rescue documents separate highly inbred lineages of wolves that have increased litter size when they are crossed with each other (F1 X F1). 73 Fed. Reg. at 63,929 (citing Fredrickson et al. 2007). This phenomenon—that crosses between different lines result in increased fitness—is called ‘heterosis’ and reflects the fact that different populations tend to fix different subsets of deleterious alleles. It does not, however, reflect the elimination of inbreeding. That is, heterosis is not the inverse of inbreeding depression (Keller and Waller 2002). In the Fredrickson et al. (2007) study, subsequent crossings with the ‘rescued’ offspring (F1 X cross-lineage) resulted in reduced litter size again and a

positive inbreeding coefficient (f), “revealing a high genetic load, the accumulation of deleterious or lethal alleles” (Fredrickson et al. 2007, p. 2368, Figure1, S1). The Proposed Rule in presenting these examples, thus confuse a demographic response to the introduction of a new immigrant with the elimination of inbreeding when, in fact, each of the populations remains severely inbred. Thus, contrary to the Service’s assertions, the scientific evidence does not support the idea that an inbred population of Northern Rocky Mountain wolves can be “saved” by the introduction of one or two individuals.

Finally, the Proposed Rule argues that “natural connectivity is not and has never been required to achieve our recovery goal. Human intervention in maintaining recovered populations is necessary for many conservation-reliant species and a well-accepted practice in dealing with population concerns (Scott et al., 2005).” 73 Fed. Reg. at 63,930. The particular citation given for this statement, however, actually identifies the gray wolf specifically as being a species that is not conservation-reliant.

The term “conservation-reliant species” was developed to describe species “that are at risk from threats so persistent that they require continuous management intervention” (Scott et al. 2005). Examples include species that occur only in captivity or are only sustained through releases of captive-bred individuals or continued modification to their habitat, for example. In their overview of “the recovery continuum” Scott et al. (2005) identify the gray wolf as a species that possesses the qualities that would make it independent of the need for continued management and one that “can maintain viable populations...without further direct intervention” (Id. p.385).

By proposing that wolves in the Northern Rocky Mountains can be considered “recovered” while requiring artificial measures to ensure genetic exchange sufficient to ensure their survival, the Service has acknowledged that they do not intend to recover a self-sustaining population of wolves. This is contrary to both the letter and spirit of ESA, which calls for the recovery of species rather than the creation of a perpetually managed and manipulated group of organisms. Furthermore, while the Service has suggested that they will take action in the future, if necessary, to address any genetic problems that occur, they have not formally established a plan to do so. For example, the Memorandum of Understanding (“MOU”) drafted as an agreement between the states of Wyoming, Montana, Idaho and the Service, see U.S. Fish and Wildlife Service, Draft Memorandum of Understanding, Maintenance and Enhancement of Gray Wolf Recovery in the Northern Rocky Mountains (available at: <http://westerngraywolf.fws.gov>), does not identify any system to document or monitor genetic exchange between subpopulations of wolves nor does it establish a genetic measure or a coordinated process for determining when intervention would be necessary or how it would be carried out. Overall, the draft MOU is vague, contains no quantifiable goals, and does not even define its terms (such as “adaptive management” or “genetic connectivity”). It also is completely non-binding:

All signatories recognize that each has statutory responsibilities that cannot be delegated and that this MOU does not and is not considered to abrogate any signatory's statutory responsibilities. This MOU is subject to and is intended to be consistent with all appropriate federal and state laws. This MOU does not obligate any of the undersigned agencies to the expenditure of funds except in accordance with lawful appropriations by the respective agencies. The undersigned agencies will take appropriate steps to seek funding to implement this MOU.

(Draft MOU at ¶ 8.)

In short, the best available scientific information does not support the Service's contention that Northern Rocky Mountain wolves are not threatened by genetic isolation, or that these threats can be easily cured by the natural or artificial introduction of one or two individuals into an inbred population. Rather than developing a plan to fully recover the gray wolf by facilitating the natural genetic exchange the Service itself deems necessary for the wolves' recovery, the Service has chosen to abnegate its responsibility.

### **III. The Endangered Species Act Does Not Allow the Service to Delist Wolves in Montana and Idaho while Wolves in Wyoming Remain Endangered**

The Proposed Rules states that the Service is "considering a final rule that would . . . continue[] to protect wolves under the Act and retain[] their nonessential experimental status in the significant portion of the range in northwestern Wyoming, outside the National Parks, while removing the Act's protections in the remainder of the DPS." 73 Fed. Reg. at 63,927. The Service, however, may not delist wolf populations in Montana and Idaho while wolves in Wyoming remain in danger. Even if such a partial delisting were possible, the Service would not be able to continue to classify Wyoming wolves as a "nonessential experimental" portion of the Northern Rocky Mountains DPS.

As an initial matter, the plain text of the ESA simply does not allow the Services to list or delist only a portion of a distinct population segment. Under the Act, the only listable entities are species, which the Act defines as including "any subspecies of fish or wildlife or plants, and any distinct population segment of any species of vertebrate fish or wildlife which interbreeds when mature." 16 U.S.C. § 1532(16). Likewise, the ESA defines an "endangered species" as "any species which is in danger of extinction throughout all or a significant portion of its range . . . ." 16 U.S.C. § 1532(6) (emphasis added). Thus, when a DPS, which for purposes of the Act is a species, is in danger of extinction throughout a significant portion of its range, it is an endangered species pursuant to the definitions in the ESA. Citing a recent Solicitor's opinion, the Service implies that it may list only the portion of the DPS that constitutes a significant portion of the range and delist the remainder of the DPS. 73 Fed. Reg. at 63,930. This is contrary to the definition of "endangered species" described above. If the Service concludes that the portion of the Northern Rocky

Mountain DPS in northwestern Wyoming outside the National Parks is a significant portion of the range of the DPS, and that wolves in Wyoming are in danger of extinction, it must list the entire DPS pursuant to the Act's definitions of species and endangered species.<sup>10</sup>

Further, an interpretation of the ESA that allows listing only those portions of species, subspecies, and distinct population segments that are a "significant portion of the range," renders the Service's distinct population segment definition of species meaningless. In 1978, Congress amended the definition of "species" to include "any distinct population segment of any species or vertebrate fish or wildlife which interbreeds when mature." 16 U.S.C. § 1532(16). The Services further defined this term to mean populations that are (1) discrete and (2) significant to the taxon to which they belong. See "Policy Regarding the Recognition of Distinct Vertebrate Population Segments Under the Endangered Species Act," 61 Fed. Reg. 4,722 (February 7, 1996). An interpretation of the statute that allows the Service to list or delist a population that is a significant portion of the range renders the concept of a DPS unnecessary, because most any population that qualifies as a DPS, and is thus significant to the taxon to which it belongs, will likely also occupy a significant portion of the range of a species or subspecies. Thus, the Service does not have the option of selectively delisting portions of the Northern Rocky Mountain DPSs' range. If the gray wolf remains endangered in Wyoming, then it cannot be delisted in Montana and Idaho.

Even if the Service could delist wolves in Montana and Idaho while they remain in danger in Wyoming, it certainly cannot continue to classify that population as a "nonessential experimental" population pursuant to Section 10(j) of the Act. 16 U.S.C § 1539(j)(2). A population may only be deemed an "experimental population" under 10(j) of the Act if it is "wholly separate geographically from nonexperimental populations of the same species." 16 U.S.C. § 1539(j)(2). The Wyoming population that lives outside of the National Parks is not geographically separate from other wolves within Wyoming or even the greater Yellowstone area. Therefore, this population is not "wholly separate geographically" from other members of the Northern Rocky Mountain DPS and cannot be designated an experimental population under section 10(j).<sup>11</sup>

---

<sup>10</sup> Wyoming constitutes a significant portion of the range of the Northern Rocky Mountain DPS, both in terms of geographic area and total population. According to *Wyoming Wolf Weekly* (Oct 27 to Nov 8, 2008); "the Wyoming wolf population (outside Yellowstone National Park)" contained ">20 confirmed packs, ~16-17 breeding pairs, and ~180 wolves in 2008."

<sup>11</sup> In addition, even if the Wyoming population could be considered an experimental population, it could not be classified as a "nonessential" experimental population. An essential experimental population is one that is determined to be "essential to the continued existence of an endangered species or a threatened species." *Id.* at § 1539(j)(2)(B). Wolves within Wyoming and outside of the National Park represent a significant portion of the Northern Rocky Mountain DPS's total population and, as the Service itself has found, maintaining healthy wolf populations in Wyoming is essential to the DPS's survival. Indeed, it makes little sense that a population could be both "nonessential" to the continued existence of the DPS while constituting a "significant" portion of the range of that same DPS. In proposing to designate the entire Northern Rocky Mountain wolf population a DPS and then delist all

The Service's proposal to designate the Northern Rocky Mountain population of the gray wolf a distinct population segment of a species and then delist most of that species while maintaining the nonessential experimental population status of the remaining DPS violates the ESA and should not be adopted.

## CONCLUSION

The Service's renewed proposal to delist wolves in the Northern Rocky Mountains is based on flawed recovery criteria that are not supported by the best available scientific information. Moreover, the Service's conclusion that this population is not threatened by genetic isolation and that inbreeding can be cured by the introduction of a few individual immigrants is also contradicted by the best available science. Finally, the ESA does not allow the Service to delist wolves in Montana and Idaho, while concluding that they are still threatened in Wyoming. We therefore urge the Services to withdraw its Proposed Rule and to retain protections for the Northern Rocky Mountains gray wolf until it establishes scientifically defensible recovery criteria, prepared in the context of a national wolf recovery plan.

Very truly yours,



Sylvia Fallon, Ph.D  
Rebecca Riley  
Andrew Wetzler

---

but "a significant portion of the range" of that DPS, the Service has thus conceded that the remaining population is significant to the DPS as a whole. It is inconsistent to designate this population both significant and nonessential to the same DPS.

## LITERATURE CITED

- Balloux, F., W. Amos and T. Coulson. 2004. Does heterozygosity estimate inbreeding in real populations? *Molecular Ecology* 13: 3021-3031.
- Bangs, E.E. 2002. Wolf population viability peer review – draft summary. U.S. Fish and Wildlife Service, Ecological Services, 100 N. Park, Suite 320, Helena, Montana. Unpublished report. 9 pp.
- Bensch, S., H. Andren, B. Hansson, H. Pedersen, H. Sand, D. Sejberg, P. Wabakken, M. Akesson and O. Liberg. 2006. Selection for heterozygosity gives hope to a wild population of inbred wolves. *PLoS ONE* 1: e72.  
doi:10.1371/journal.pone.0000072.
- Brook, B. W., L. W. Traill, C. J. A. Bradshaw. 2006. Minimum viable population sizes and global extinction risk are unrelated. *Ecology Letters* 9: 375-382.
- Frankel, O. H. and M. E. Soule. 1981. *Conservation and Evolution*. Cambridge University Press, New York, NY.
- Frankham R. 1995. Effective population size/adult population size ratios in wildlife: a review. *Genetical Research* 66: 95-107.
- Franklin, I. R. and R. Frankham. 1998. How large must a population be to retain evolutionary potential? *Animal Conservation* 1: 69-70.
- Fredrickson, R. J., P. Siminski, M. Woolf and P. W. Hedrick. 2007. Genetic rescue and inbreeding depression in Mexican wolves. *Proceedings of the Royal Society B* 274: 2365-2371.
- Fritts, S. H. and L. N. Carbyn. 1995. Population viability, nature reserves and the outlook for gray wolf conservation in North America. *Restoration Ecology* 3: 26-38.
- Keller, L. F. and D. M. Waller. 2002. Inbreeding effects in wild populations. *Trends in Ecology and Evolution* 17: 230-241.
- Lande, R. 1988. Genetics and demography in biological conservation. *Science* 241: 1455-1460.
- Lande, R. 1995. Mutation and conservation. *Conservation Biology* 9: 782-791.

- Liberg, O., Andren, H., Pedersen, H.C., et al. 2005. Severe inbreeding depression in a wild wolf (*Canis lupus*) population. *Biology Letters* 1: 17-20.
- Leonard, J. A., C. Vila, R. K. Wayne. 2005. Legacy lost: genetic variability and population size of extirpated US gray wolves. *Molecular Ecology* 14: 9-17.
- Morgan, J. P., A. Bahr, C. E. Franti, C. S. Bailey. 1993. Lumbosacral transitional vertebrae as a predisposing cause of cauda equina syndrome in German Shepherd Dogs: 161 cases (1987–1990). *Journal of the American Veterinary Medical Association* 202: 1877–1882.
- Morgan, J.P. 2000. Lumbosacral disease. In J.P. Morgan, A. Wind, and A.P. Davidson (Eds.) *Hereditary Bone and Joint Disease in the Dog*. Hannover: Schlutersche. Pp.209–229.
- Olafsdottir, G. A. and T. Kristjansson. 2008. Correlated pedigree and molecular estimates of inbreeding and their ability to detect inbreeding depression in the Icelandic sheepdog, a recently bottlenecked population of domestic dogs. *Conservation Genetics* 9: 1639-1641.
- Palstra, F. P. and D. E. Ruzzante. 2008. Genetic estimates of contemporary effective population size: what can they tell us about the importance of genetic stochasticity for wild population persistence? *Molecular Ecology* 17: 3428-3447.
- Pemberton, J. 2004. Measuring inbreeding depression in the wild: the old ways are the best. *Trends in Ecology and Evolution* 19: 613-615.
- Raikkonen, J., A. Bignert, P. Mortensen and B. Fernholm. 2006. Congenital defects in a highly inbred wild wolf population (*Canis lupus*). *Mammalian Biology* 71: 65-73.
- Raikkonen, J., J. A. Vucetich and R. O. Peterson. In review. Congenital bone deformities and the inbred wolves of Isle Royale.
- Reed, D. H, J. J. O’Grady, B. W. Brook, J. D. Ballou and R. Frankham. 2003. Estimates of minimum viable population sizes for vertebrates and factors influencing those estimates. *Biological Conservation* 113: 23-34.
- Reed, D. H. and G. R. Hobbs. 2004. The relationship between population size and temporal variability in population size. *Animal Conservation* 7: 1-8.
- Ricklefs, R. E. and G. L. Miller. 2000. *Ecology* (4<sup>th</sup> edition). W. H. Freeman and Company. New York, NY.

- Scott, J.M., Goble, D.D., Wiens, J.A., Wilcove, D.S., Bean, M., Male, T. 2005. Recovery of imperiled species under the Endangered Species Act: The need for a new approach. *Frontiers in Ecology and the Environment* 3: 383-389.
- Slate, J., P. David, K. G. Dodds, B. A. Veenvliet, B. C. Glass, T. E. Broad and J. C. McEwan. 2004. Understanding the relationship between the inbreeding coefficient and multilocus heterozygosity: theoretical expectations and empirical data. *Heredity* 93: 255-265.
- Soule, M. E. 1986. (Ed.) *Conservation Biology, the science of scarcity and diversity*. Sinauer, Sunderland, MA.
- Soule, M. E. and B. A. Wilcox. (Eds.) 1980. *Conservation Biology, an evolutionary-ecological perspective*. Sinauer, Sunderland, MA.
- Soule, M. E. and D. Simberloff. 1986. What do genetics and ecology tell us about the design of nature reserves? *Biological Conservation* 35: 19-40.
- Thomas, C. D. 1990. What do real population dynamics tell us about minimum viable population sizes? *Conservation Biology* 4: 324-327.
- Traill, L. W., C. J. A. Bradshaw, B. W. Brook. 2007. Minimum viable population size: A meta-analysis of 30 years of published estimates. *Biological Conservation* 139: 159-166.
- U.S. Fish and Wildlife Service. 1978. Recovery plan for the eastern timber wolf. U.S. Fish and Wildlife Service. Twin Cities, Minnesota.
- U.S. Fish and Wildlife Service. 1982. Mexican gray wolf recovery plan. U. S. Fish and Wildlife Service, Albuquerque, New Mexico. 103 pp.
- U.S. Fish and Wildlife Service. 1987. Northern Rocky Mountain Wolf Recovery Plan. U.S. Fish and Wildlife Service, Denver, Colorado. 119 pp.
- U.S. Fish and Wildlife Service. 1994. Final Environmental Impact Statement: The Reintroduction of Gray Wolves to Yellowstone National Park and Central Idaho. Appendix 9. US Fish and Wildlife Service, Helena, MT. Pp. 8-37 – 8-44.
- U.S. Fish and Wildlife Service. 1994a. Survey of biologists for Environmental Impact Statement. On file with U.S. Fish and Wildlife Service, Denver, Colorado.
- Vila, C. A. Sundquist, O. Flagstad, J. Seddon, S. Bjornerfeldt, I. Kojola, A. Casulli, H. Sand, P. Wabakken and H. Ellegren. 2003. Rescue of a severely bottlenecked wolf (*Canis lupus*) population by a single immigrant. *Proceedings of the Royal Society B* 270: 91-97.

vonHoldt, B. M., D. R. Stahler, D. W. Smith, D. A. Earl, J. P. Pollinger and R. K. Wayne. 2007. The genealogy and genetic viability of reintroduced Yellowstone gray wolves. *Molecular Ecology* 17: 252-274.

Attachments: Statement by Staffan Bensch  
Statement by Carles Vila

## ATTACHMENT 1

### Statement by Staffan Bensch

(submitted via: <http://www.regulations.gov>)

It has been brought to my attention that a study I conducted is being cited in your proposal to remove endangered species protections from gray wolves in the northern Rocky Mountains (Bensch et al. 2006). Specifically, the proposal states that “dispersers into a system experiencing some level of inbreeding would be much more likely to be selected for breeding and have their genes incorporated into the inbred population (Bensch et al. 2006....)” While my study demonstrated that more heterozygous individuals were more likely to become breeders, the research also revealed that inbreeding continued to persist. In fact, we found that heterozygosity and inbreeding were poorly correlated and over the timeframe of the study, the inbreeding coefficient for this population actually increased. Before extrapolating the results from our study to managing actions on other wolf or small populations of other species, one should be aware of that some of the aspects of the Scandinavian wolf population are rather unique and may have contributed to the strong effects we observed on heterozygosity selection. First, the whole population was founded by only three individuals, and the average level of the inbreeding coefficient is very high ( $>0.25$ , i.e. corresponding to full sib matings) compared to most populations in the wild. Second, the number of generations since founding by the unrelated and outbred immigrants are few (2-6) resulting in unusually strong genome wide linkage disequilibrium. Strong linkage disequilibrium will facilitate selection on overall heterozygosity. Hence, it is not clear whether less inbred populations with lower linkage disequilibrium will respond in the same way as we observed among the Scandinavian wolves.

Staffan Bensch  
Professor  
Molecular Population Biology Lab  
Department of Ecology  
Lund University  
S-223 62 Lund  
Sweden  
Phone Office +46(46) 2224292; Home +46(46) 209650  
<http://www.biol.lu.se/zoekologi/molekol/index.html>

## ATTACHMENT 2

### Statement by Carles Vila

(submitted via: <http://www.regulations.gov>)

November 24, 2008

To US Fish and Wildlife Service,

I am aware that the recent proposal to remove endangered species protections from gray wolves in the northern Rocky Mountains references one of my studies as an example in which “one or two new genetic lines can save a severely inbred small wolf population” (73 FR 63929). The study (Vila et al. 2003) documented an increase in the multilocus heterozygosity and population size of a small population of inbred wolves after the addition of a single immigrant.

However, while my study shows an increase of heterozygosity, the increase in the population size was mainly due to behavioral aspects and not to an increase in heterozygosity. We believe that the arrival of one (third) individual allowed the formation of a second pack and then pups born in the two packs would not see each other as relatives and would easily mate and form new packs. Until then all matings had to take place between members of the same pack.

My study actually shows the importance of facilitating gene flow between populations, and in the extreme case of the Scandinavian wolves the arrival of one individual allowed a population growth, but this does not mean that there are not inbreeding problems. In fact, follow up studies to my publication found that while MLH increased in this population of wolves after the introduction of a single immigrant, the inbreeding coefficient did not go down – it continued to rise (Liberg et al. 2005, Bensch et al. 2006). Additionally, vertebral deformities associated with inbreeding have been documented for this population (Raikkonen et al. 2006).

Migrants can help to reduce inbreeding, but my study does not imply that just one individual will solve all genetic problems.

Sincerely,

Carles Vilà  
Estación Biológica de Doñana-CSIC  
Avd. María Luisa s/n  
41013 Sevilla, Spain  
[carles.vila@ebd.csic.es](mailto:carles.vila@ebd.csic.es)