Appendix E

Draft Segregation Protocol - August 28, 2015

June 2017

Protocol to Minimize Introgression between Spring-run and Fall-run Chinook Salmon for Permit 17781 under Section 10(a)(1)(A) of the Endangered Species Act

Background

As the SJRRP establishes populations of both SRC and FRC in the Restoration Area, the potential exists for these populations to interbreed or interfere with successful spawning through redd super-imposition. Several other river systems with constrained spawning habitat due to man-made passage barriers support FRC and SRC populations. In these systems, fall-run tend to numerically dominate although it is not clear to what extent the population size differences are due to hatchery practices.

Although, FRC and SRC have different spawning timing, enough overlap exists to allow the potential for Tomalty et al. (2014) hypothesize that in the absence of any management efforts to preclude hybridization, the Restoration Area would likely result with interbreeding between FRC and SRC. This hypothesis is supported by documentation of hybridization between Chinook salmon runs in other river systems such as the Trinity (Kinziger et al. 2008), Puntledge (Withler et al. 2012), and White Rivers (Smith et al. 2011). Of particular interest to the SJRRP, Kinziger et al. (2008) documented that interbreeding had occurred between spring-run and fall-run Chinook salmon on the Trinity River, but did not hypothesize on any negative consequences to the stocks or how the level of introgression compared on historic rates prior to dam construction. Conversely, on the Lewis River in Washington, FRC and SRC are believed to have enough temporal separation to preclude interbreeding between the two runs (refs). Stillwater et al. (2003) hypothesize that SRC and FRC may segregate spatially in the Restoration Area suggesting the two runs could co-exist without management efforts to separate two runs.

Although it is not clearly understood how much interbreeding occurred between FRC and SRC under unperturbed conditions, Tomalty et al. (2014) examined the causes and consequences of hybridization of Chinook salmon runs in the San Joaquin River and provided the following observations:

- Hybridization is defined as the mating and production of offspring by individuals from genetically distinct groups, be they species or genetically divergent populations within a single species.
- Central Valley Chinook salmon are an excellent example of a group that faces genetic diversity loss and population structure collapse, in part from hybridization.
- For the San Joaquin, overlap in migration spawn timing and lack of spatial separation between runs will likely create conditions that encourage introgression.
- Introgression will almost certainly lead to the loss of distinct spring-run and fall-run phenotypes and/or genotypes.

- Introgression may lead to a hybrid storm where phenotypes are lost and the population consists primarily of hybrids.
- Or, run-timing phenotypes may be preserved, but the genetic distinction between runs may be lost.

The Hatchery Scientific Review Group evaluated the impacts of introgression of hatchery stocks on wild stocks, and found that significant decreases would result with increasing introgression rates (HSRG 2014). Although these results don't directly relate to FRC introgression on SRC, they do illustrate the potential consequences of introgression between distinct runs.

The potential for FRC to disturb SRC redds during their spawning activity is a function of habitat overlap, available habitat, and relative population sizes. In the Restoration Area, we have identified the potential distance downstream from Friant Dam that will allow Chinook salmon spawning, and are further evaluating the quality of potential habitat based on hydrology and substrate composition. We do not have data on the habitat overlap since both runs have been extirpated form the Restoration area for over 60 years. For the Restoration area, we expect the risk of redd superimposition to increase as population sizes increase. Based on data from the capture of FRC over the past few years, we expect to have fewer than 100 FRC redds per year during the time frame of this protocol and permit. FRC superimposing on SRC redds does not necessarily mean a loss of the eggs in the SRC redd. Yuba River researchers found that 45 percent of the redds that experienced superimposition actually had egg pockets that were affected (YRDP 2013). Based on the amount of habitat and low number of FRC spawners, we predict a low risk for superimposition in the near term, but will continue testing and be prepared to deploy methods to protect SRC redds when both runs are in the system.

The 10(a)(1)(A) permit 17781 requires the US Fish and Wildlife Service to develop this Segregation Protocol to ensure to the greatest extent possible, the prevention of fall-run Chinook salmon (FRC) genetic introgression to the spring-run Chinook salmon (SRC) population and FRC superimposition on SRC redds. This protocol must be implemented within two years of issuance of this permit. The focus of concern is both 1) genetic introgression (the hybridization of FRC with Chinook salmon SRC fish) and 2) potential mortality to SRC eggs from FRC superimposition on SRC redds. These concerns are discussed in more detail in both the Hatchery and Genetics Management Plan (HGMP) and the Fisheries Management Plan. To summarize some of the key concepts from the HGMP:

1) It is high priority to reduce hybridization between SRC and FRC, especially if Feather River Fish Hatchery SRC are being used to re-establish SRC in the San Joaquin River due to the hybridization that has already occurred in the Feather River system.
2) A performance measure for genetic introgression should be established with data collected and reported.
3) Genetic analysis should be used to evaluate the degree of hybridization occurring in re-established SRC population.
4) Placement and use of a segregation weir should be evaluated.
5) Carrying capacity of the spawning habitat should be evaluated.
6) Flow management should be used to help segregate runs.
7) Marking and tagging techniques should be used to distinguish between runs.

Total "prevention" might not be possible because there may be fish with one genotype exhibiting the other phenotype. For example, in the Feather River, a few adult salmon with a FRC genotype migrate in spring and a few adult salmon with a SRC genotype migrate in the fall. If this occurs in the Restoration Area, it will not be possible to completely segregate the two runs. However, discussions with the Genetics subgroup have concluded that a low rate of genetic introgression ($\leq$ 2%) will not have serious consequences to SRC and actually may facilitate the adaptation of reintroduced SRC fish to environmental conditions in the Restoration Area. The March 12, 2014 recommendation of the Genetics Subgroup is to monitor the level of introgression to ensure there is no loss of the SRC phenotype due to genetic introgression. Following their recommendation, a segregation weir would be used only if monitoring determines that genetic introgression is likely to exceeds 2 percent, there is FRC superimposition on SRC redds, and it is determined that a segregation weir could be effective in the Restoration Area.

Overall Strategy

Many questions exist regarding the need for managing the interactions between FRC and SRC and the effectiveness of potential strategies. Therefore, this draft protocol will be focused on implementing an adaptive strategy and focuses on actions to be implemented during the initial 5-year time frame of the 10(a)(1)(A) permit 1778. These actions are designed to reduce the risk of genetic introgression and redd superimposition while informing the long-term strategy for managing the two runs in the Restoration Area.

The Program has identified several questions to evaluate during the first few years of implementation. These questions are generally encompassed below:

1. *Will FRC and SRC spatially segregate within the Restoration Area resulting in little risk of genetic introgression or superimposition?*

   One hypothesis for how SRC and FRC will interact within the Restoration Area is that the two runs will have very little interaction with each other. The two runs may segregate spatially if seasonal shifts in water temperature influence SRC to primarily use habitat closer to Friant Dam and FRC use habitat lower in the Restoration Area. They may also seek out different habitat across the stream channel. SRC may spawn in deeper water towards the center of the channel whereas FRC may be more likely to spawn in habitat towards the margins of the stream channel.

   The Program has been monitoring FRC spawning site selection and timing for the past few years, but we have not had SRC in the system to monitor spawning site selection and timing. Site selection is likely to vary on annual basis due to environmental conditions and population size. The first few years of SRC returns will provide some initial insights into how SRC will use the available habitat, but continual monitoring will be required as flows increase, in different water year types, and as the number of SRC returns increases.

2. *Will FRC and SRC segregate temporally in the Restoration Area resulting in little risk of genetic introgression?*

   It is possible that SRC spawning will be completed by the time FRC spawning begins so that there will be little chance of hybridization. If spawning timing is separated enough, the risk of genetic introgression will be minimal. However, the Program will still need to evaluate the need to protect SRC redds from superimposition from FRC.

3. *Can a segregation weir be effective for separating SRC and FRC during spawning?*

   Although segregation weirs have been used to successfully to separate the two runs in other systems, several factors must be addressed to determine if it can be an effective tool within the Restoration Area. If SRC spawn throughout the entire range of suitable spawning habitat, then a segregation weir would not be able to preclude SRC and FRC from having some habitat overlap, assuming both runs are allowed to spawn in the Restoration Area. If SRC do spawn throughout the entire range of spawning habitat, the Program would need to decide how much of the available habitat should be protected for SRC.

   The effectiveness of a weir is also dependent upon finding a suitable location for the construction and operation of a segregation weir.

As information addressing these and more specific questions becomes available to the Program annual strategies will be adjusted accordingly.

2016-2019 Implementation

It is anticipated that fish passage will mostly remain blocked under normal conditions during this time period, allowing the Program control over when and how many SRC and FRC are able to access the spawning area. We anticipate the need for migrating SRC adults to be detected, captured, and trucked to the spawning habitat during the spring months (est. Feb-June) beginning in 2016.

Tissue samples will be collected from all fish for genetic analysis, and the fish will be externally marked. Regular monitoring of these fish will occur over the summer to determine whether they find suitable holding habitat and hold successfully in cool pools over the summer. Beginning in late August and continuing into October, we expect the SRC adults to move into spawning habitat and spawn. Redd and carcass surveys would be conducted to recover post-spawn fish and document redd locations.

Beginning in October and continuing through December, we expect to see FRC trying to access the Restoration Area. If SRC are spawning in Reach 1, we would truck FRC only after SRC have completed spawning to minimize the potential for any introgression. If no SRC are spawning in Reach 1, trapping and trucking of FRC adults could take place as soon as they enter

the Restoration Area. We will also test our ability to protect SRC redds with physical barriers (redd "grates") and evaluate the likelihood of FRC superimposition. Since we expect a limited number of SRC redds during the first years of SRC returning, we will be able to deploy and monitor redd grates for all SRC redds.

This strategy will allow the Program to learn where SRF and FRC will choose to spawn and if the FRC might damage SRC redds. Studies of the effects of flow management on run segregation cannot occur during the initial 5-year time frame because adult passage and conveyance of the fall pulse flow throughout the Restoration Area are not expected to occur for several years.

During this same time frame, potential locations, designs, and operations of segregation weirs can be evaluated and tested. In addition, evaluations of spawning habitat carrying capacity, including both physical and biological study results, can be finalized. All passage facilities constructed in the Restoration Area will also be evaluated for potential designs to limit or block migrating adult FRC as such facilities may be the most cost effective means for managing SRC/FRC interactions when passage is restored rather than operating an independent segregation weir.

Prior to passage being restored and expiration of this time frame, a revised Segregation Protocol will be developed to encompass both the changes due to fish passage and also to capture the information and learning that has occurred during the initial period. This report will include relevant information such as population estimates, spawning locations and timing, genetic analysis results, effectiveness of physical barriers tested, and segregation weir evaluations.

Monitoring Elements

There are three primary monitoring elements: 1) genetic introgression; 2) redd superimposition; and 3) segregation weir.

Genetic Introgression

There are two monitoring elements for genetic introgression: 1) quantify the potential spatial and temporal overlap between SRC and FRC spawners and 2) develop performance measures to evaluate the degree of hybridization in the SRC population.

First, the potential for genetic introgression must be managed during the initial time period, because FRC may spawn at the same time and location as some SRC spawners. Monitoring will be needed to determine if more than 2 percent of the adult SRC population spawn at the same time and location as would be expected for FRC spawning. It is expected that most SRC will spawn in September and October, whereas most FRC would probably spawn in November and December. It is also anticipated that most SRC will spawn near Friant Dam where temperatures are most suitable; whereas FRC will likely spawn throughout the 10-mile spawning reach, as they did in fall 2013 and 2014.

Monitoring Element #1.

Monitor SRC spawning timing and distribution with weekly spawning surveys from mid-August until all SRC have spawned. The exact location of each SRC redd will be identified with GPS coordinates. The timing and distribution of SRC spawning will be compared to the timing and distribution of FRC spawning in Reach 1 that the Program began Monitoring in 2012.

Second, it will be necessary to develop a performance measure(s) to evaluate the degree of potential hybridization in the SRC population. This will require a genetic metric that can detect hybridization between the SRC and FRC populations and baseline data describing the genetic structure and phenotypic expression (i.e., run timing) of the SRC population prior to any potential hybridization. These monitoring elements may require more than this initial 5-year time frame to complete.

Monitoring Element #2.

Characterize the genetic structure of the SRC population each year during the initial 5-year period?

In conjunction with genetic monitoring of our captive broodstock program, the Program will develop a baseline of the genetic signature of adult SRC returning to the Restoration Area.

Monitoring Element #3.

Characterize the phenotypic expression (run timing) of the SRC population each year. During this initial period, when adult SRC would have to be trapped and transported to Reach 1, daily catch rates would be monitored from mid-February at least through mid-July or until adults are no longer caught.

Redd Superimposition

It is assumed that SRC redds may be superimposed by FRC spawners if both SRC and FRC are transported to Reach 1 in any year. Therefore, before FRC are transported to Reach 1, FRC redds built early in the spawning period will be protected with “redd grates” to study effectiveness of this strategy at preventing superimposition. Heavy metal redd grates placed over individual redds were effective at protecting FRC Chinook salmon eggs buried in artificial redds for the Knights Ferry Gravel Replenishment Project on the Stanislaus River in fall 2004. Each redd grate was constructed with 3-foot pieces of reinforcing bar connected with plastic ties to form a triangle that was anchored with cobble immediately over the egg pockets. The metal triangle and cobble did not interfere with hyporheic flow or fry emergence and successfully prevented superimposition during the Stanislaus River study. The Program evaluated the use of redd grates in the Fall of 2014 and Fall of 2015.

Monitoring Element #4.

Evaluate redd grate effectiveness during FRC spawning. If FRC are transported to Reach 1 during fall 2014 and fall 2015, redd grates would be placed over the tailspills of the early spawning fish. Weekly surveys would be conducted to inspect and if necessary

repair redd grates and evaluate whether the grates protect redds from late spawning FRC. Redd grates would be assumed to be effective, if late spawners do not remove more than 3 inches of gravel from a grate-protected tailspill or bury a grate-protected tailspill with fine sediment.

Monitoring Element #5.

Evaluate redd grate effectiveness during SRC spawning. If both SRC and FRC are to be transported to Reach 1 during the same year, redd grates would be placed over the tailspills of all SRC redds before any FRC are transported dependent on proper permitting being in place. Weekly surveys would be conducted to inspect and if necessary repair redd grates and evaluate whether the grates protect SRC redds from FRC spawners. Redd grates would be assumed to be effective, if FRC spawners do not remove more than 3 inches of gravel or deposit fine sediment onto a grate-protected SRC redd. All adult FRC would receive an external tag to distinguish them from SRC.

It will be assumed that SRC have completed spawning when no live SRC or recent redd activity is observed during two consecutive weekly spawning surveys.

Segregation Weir

A segregation weir may be needed to minimize genetic introgression and/or redd superimposition after adult passage has been restored. During this initial 5-year period, potential locations, designs, and operations for a segregation weir will be evaluated in the event that a weir is needed at that time.

Monitoring Element #6.

Evaluate designs for segregation weirs

The Program will continue to examine the use of segregation weirs in other systems and their potential for use in the Restoration Area. This element will be accomplished through literature review and discussions with individuals from other programs.

Monitoring Element #7.

Evaluate locations for segregation weirs

The Program will determine and evaluate potential locations for a segregation weir within the Restoration Area. This task will be accomplished through an evaluation of the physical parameters of potential locations, and information gained on habitat use by SRC and FRC.

References

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