

## Attachment 2

### Aquatic Invertebrate Monitoring Study Plan

#### Interagency Study Proposal

June 15, 2003

### Evaluation of Rotenone Use in Silver King Creek Basin on Aquatic Macroinvertebrates, 2003-2007

#### Background

The California Department of Fish and Game (Department) proposes to treat Silver King Creek basin with rotenone during the late summer of 2003, 2004, and possibly 2005. The goal of this project is to restore Paiute cutthroat trout (*Oncorhynchus clarki seleniris*), a federally listed threatened species, to its historic habitat. The Department anticipates that successful reintroduction of Paiute cutthroat trout will lead to delisting of the fish as federally threatened, as well as the creation of a Fish and Game Commission designated Heritage Trout Fishery.

The Lahontan Regional Water Quality Control Board has expressed concern regarding possible impacts of rotenone within the project area to non-target organisms, specifically aquatic macroinvertebrates. The Department conducted studies in Silver King Creek (Trumbo et al 2000a) and nearby Silver Creek (Trumbo et. al. 2000b) to evaluate rotenone impacts to aquatic macroinvertebrates during past projects. Both studies found that rotenone use did not affect species abundance, and that there was evidence of short-term impacts. No evidence of long-term impacts were found in either study.

The Department of Fish and Game proposes to evaluate the response of aquatic macroinvertebrates to the chemical treatment of Silver King Creek. This study will be funded through multi-agency cooperation of the Humboldt-Toiyabe National Forest, U.S. Fish and Wildlife Service, and the Department.

#### Objective

The primary objective of this study is to determine if rotenone use will significantly impact the biological condition of Silver King Creek. A number of metrics will be analyzed to examine measures of taxa richness, composition and function, including: 1) taxa richness; 2) abundance; 3) Ephemeroptera, Plecoptera, Trichoptera (EPT) richness; 4) EPT Index; 5) number of families; 6) percent dominant taxon; 7) Shannon Diversity Index; 8) mean tolerance value, and 9) Community Similarity Indices (Jaccard and/or Brillouin Index).

#### Study Design

A basin approach study design has been selected by the cooperating agencies. Sampling will be conducted pre-treatment at all sites during mid-August 2003, and post-treatment at all sites during mid-August 2005 and 2006. Should the project require a third year of rotenone treatment in 2005, post-treatment sampling would be postponed until 2006 and 2007.

The aquatic macroinvertebrate study will have four pairs of control and treatment sites on the main stem of Silver King Creek (Figure 1). Four of these eight sites have already been located as follows: Site 1 – Upper Fish Valley upstream of Bull Canyon Ck; Site 2 – Upper Fish Valley downstream of Bull Canyon Ck; Site 3 – Lower Fish Valley; Site 4 – Long Valley. Sites 1 and 2 would serve as paired controls for sites 3 and 4 which are situated within the rotenone project area. The two pairs of additional sites (two treatments and two controls) on the main stem of Silver King Creek will be located during summer of 2003.

Additional sites will be identified during early summer 2003 to evaluate the response of aquatic macroinvertebrates to rotenone in first order streams. Treatment sites will be located in the rotenone project area of Tamarack Creek. Control sites upstream of the project area will be situated in stringer meadows of Corral Valley Creek downstream of the main Corral Valley. The additional sites would be situated as follows: Site 5 – upper main stem Tamarack Ck; Site 6 – lower main stem Tamarack Ck; Site 7 – Upper Corral Valley Creek in stringer meadows below the main valley; Site 8 – in stringer meadows downstream of Site 7. One additional pair of control sites will be determined summer of 2003 and will be located in Coyote Valley Creek or another suitable first order tributary stream of Silver King Creek.

### **Sample Collection and Processing**

Samples will be collected from study site riffles by randomly selecting 3 locations out of all possible 0.09 m<sup>2</sup> areas within the site boundary. Aquatic macroinvertebrates will be collected using a D-frame kick net sampler with a 0.5 mm mesh net. Samples will be collected from the lowermost portion of the riffle, working upstream. After sampling, contents of the net will be emptied into a bucket, concentrated with a 500  $\mu$ m sieve, and preserved in 95% ethanol. Any organisms clinging to the sample net or sieve, or remaining in the bottom of the bucket will be put into the sample.

Samples will be processed and keyed by the National Aquatic Monitoring Center (The BugLab) at Utah State University, Logan, UT. The BugLab will provide a taxa list and summary metrics for each sample. Laboratory techniques, taxonomic levels, and metrics analyzed by the BugLab can be reviewed at the website: [www.usu.edu/buglab/](http://www.usu.edu/buglab/). The Buglab website also reviews quality control and assurance measures. The Buglab will archive midges (Chironomidae) and mites (Hydracarina) in separate vials for possible later analysis.

## **Physical and Chemical Habitat Characteristics**

Selected physical and chemical habitat characteristics will be measured to describe sample site characteristics, and assure similarity in habitat. For each sampling site, the following physical parameters will be measured: length, width (top and bottom), gradient, depth, % canopy, % substrate composition, embeddedness. The following physicochemical parameters will be measured for each sampling site: temperature, conductivity, alkalinity, and pH.

Rotenone concentration and duration will be determined as part of water quality monitoring during the rotenone treatment. Although water samples will not be collected at specific aquatic macroinvertebrate sampling stations, selected reaches within the basin will be sampled to evaluate rotenone efficacy during the application.

Latitude and longitude of each sampling site will be recorded with hand-held GPS units, and sample sites will be identified with rebar stakes. Digital photographs will be used to record sample site locations.

## **Data Analysis/Statistics**

Biological metrics will be analyzed for conformity to normality and evaluated for the appropriate transformation. Biological metrics may then be tested for statistical significance using fixed effect two or three factor ANOVA (site vs. year vs. season). The Bonferroni approximation can be applied to ANOVA significance levels to account for dependence among measured metrics (Ramsey 1980). If data deviate from the assumptions of normality, homoscedasticity and linearity, non-parametric analyses will be used such as the Mann-Whitney or Kruskal Wallis tests for testing hypotheses (Zar 1984).

## **Study Design Cost Analysis**

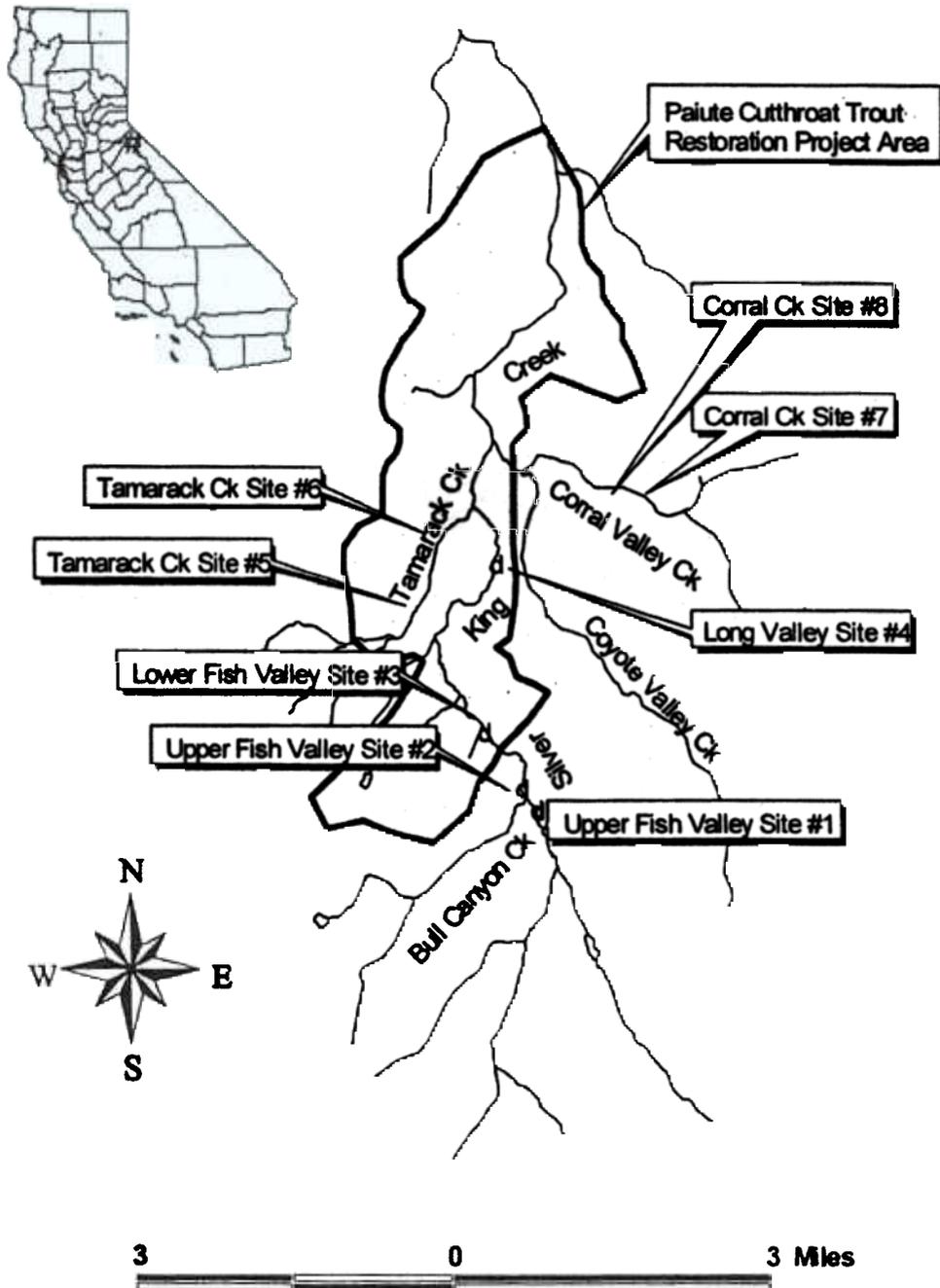
This study will be performed by the Humboldt-Toiyabe National Forest, U.S. Fish and Wildlife Service, and the Department. The Buglab estimates sample processing to cost \$200/ sample. Sample processing costs for the study designs are as follows: 3 samples/site x 14 sites = 42 samples/year; one year = \$8,400; three years (whole study) = \$25,200.

Sample processing costs will be shared by the Humboldt-Toiyabe National Forest and the U.S. Fish and Wildlife Service. Costs associated with personnel, per diem, and equipment will be borne and shared by the participating agencies.

[This outdated invertebrate monitoring site map is superseded by a revised invertebrate monitoring site map included as Attachment 5 to the Monitoring and Reporting Program. Please refer to Attachment 5 for the current map of invertebrate monitoring sites.]

Figure 1.

## Aquatic Invertebrate Sample Site Locations Silver King Creek, Alpine Co., CA



## **References**

- Ramsey, P.H. 1980. Choosing the most powerful pair wise multiple comparison procedure in multivariate analysis of variance. J. Applied Psych. 65(3):317-326.**
- Trumbo, J., S. Siepmann, and B. Finlayson. 2000a. Impacts of rotenone on benthic macroinvertebrate populations in Silver King Creek, 1990 through 1996. Office of Spill Prevention and Response, Administrative Report 00-5, March 2000. Pesticide Investigations Unit, Office of Spill Prevention and Response, California Department of Fish and Game. 40 p.**
- Trumbo, J., S. Siepmann, and B. Finlayson. 2000b. Impacts of rotenone on benthic macroinvertebrate populations in Silver Creek, 1994 through 1998. Office of Spill Prevention and Response, Administrative Report 00-7, December 2000. Pesticide Investigations Unit, Office of Spill Prevention and Response, California Department of Fish and Game. 37 p.**
- Zar, J. H. 1984. Biostatistical Analysis, 2nd ed. Prentice Hall, Englewood Cliffs, New Jersey.**

Appendix 1. *Normal taxonomic resolution of the Buglab*

Taxon or Taxa group	Buglab's Current Standard Taxonomic Level	Northwest Bioassessment Work Group Minimum Standard Taxonomic Effort
Annelida		
	Genus	Genus
Oligochaeta	Order	Family
Arthropoda		
Hydracarina	Order	Order
Crustacea		
Anostraca	Genus/species	Genus/species
	Genus/species	
Copepoda	Genus/species	
Decapoda	Genus/species	Genus
Ostracoda	Order/Family/Genus	
Amphipoda	Genus/species	Genus
Isopoda	Genus	Genus
Collembola	Order	
Insecta		
Coleoptera	Genus/species	Genus
Except Curculionidae, Heteroceridae, Ptiliidae	Family	Family
Diptera		
Atherceridae	Genus	Genus
Blephariceridae	Genus/species	Genus
Ceratopogonidae	Genus	Subfamily
Chaoboridae	Genus/species	
Chironomidae	Subfamily	Genus
Culicidae	Genus	
Deuterophlebiidae	Genus/species	Genus
Dixidae	Genus	Genus
	Family	

Empididae	Genus	Genus
Ephydriidae	Family	Family
Muscidae	Family	Family
Pelecorhynchidae	Genus	Genus
Psychodidae	Genus	Genus
Ptychopteridae	Genus	Genus
Sciomyzidae	Family	
Simuliidae	Genus	Genus
Stratiomyidae	Genus	Genus
Tabanidae	Genus	Family
Tanyderidae	Genus	Genus
Thaumaleidae	Genus	Genus
Tipulidae	Genus	Genus
Ephemeroptera	Genus	Genus
Ephemerellidae	Genus/species	species
Hemiptera	Genus/species	Genus
Lepidoptera	Genus	Genus
Megaloptera	Genus/species	Genus
Odonata	Genus/species	Genus
Plecoptera	Genus/species	Genus
Pteronarcys	Genusspecies	species
Taeniopterygidae	Family/Genus	Family
Trichoptera	Genus/species	
Coelenterata	Class	Class/Order
Mollusca		
Gastropoda	Family/Genus/species	Genus
Pelecypoda	Order/Family/Genus/species	Genus
Sphaeriidae	Family Genus	Family/Genus
Nematoda	Phylum	Phylum
Nematophora	Phylum	Phylum
Porifera	Phylum	Phylum
Turbellaria	Class	Class