Study Plan for Population Size Estimates of
O. mykiss in the lower Tuolumne River

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1   BACKGROUND AND PURPOSE

Fisheries monitoring for the Don Pedro Project (FERC Project No. 2299) by the Turlock Irrigation District (TID) and Modesto Irrigation District (MID) has long documented the presence of *Oncorhynchus mykiss (O. mykiss)* in the lower Tuolumne River (TID/MID 2005). On March 19, 1998 the National Marine Fisheries Service (NMFS) first listed the Central Valley steelhead as threatened under the Endangered Species Act (ESA). After several court challenges, NMFS issued a new final rule relisting the Central Valley steelhead on January 5, 2006 (71 FR 834). In a separate process regarding terms of the 1996 FERC license amendments for the Project, NMFS staff provided input to a draft limiting factors analysis for Tuolumne River salmonids (Mesick et al 2007) and included recommendations for developing abundance estimates, habitat use surveys and anadromy determination of resident *O. mykiss*. These recommendations were conceptually used to develop the Districts FERC Study Plan (TID/MID 2007) which was the subject of an April 3, 2008 FERC Order. As part of the Order, the Districts are required to conduct population estimate surveys in summer (June/July) and winter (February/March), starting in summer 2008 to determine *O. mykiss* population abundance by habitat type.

The purpose of the proposed *O. mykiss* population surveys is to provide population size estimates over several sampling seasons of differing environmental conditions to determine habitat use and needs within the lower Tuolumne River. The surveys will be used to examine the following hypotheses:

> Hypothesis 1: Summertime distribution of suitable habitat by observed life stages of *O. mykiss* is related to ambient river water temperature.

> Hypothesis 2: Habitat use by *O. mykiss* juveniles and adults observed in the Tuolumne River occurs at the same density in both restored and nearby reference sites.

As recommended by Stillwater Sciences (Stillwater), the surveys will employ a two-phase sampling approach of potential *O. mykiss* habitat using snorkel surveys for the development of a "bounded count" population estimate (Hankin and Mohr 2001). Although the methodology presented below discusses both repeated dive counts and calibration by depletion electrofishing, current ESA permit restrictions for both NMFS Section 10(a)(1)(A) permit No's 1280 (TID) and 1282 (Stillwater) do not allow sufficient incidental take to conduct the second phase surveys at this time using electrofishing. Discussions with NMFS permitting staff and Stillwater have occurred since submittal of the 2007 FERC Study Plan, resulting in a pending formal request to NMFS by Stillwater for modification of Permit 1282 (see Section 6 below). The Section 10 Permit 1280 issued to TID in 2005 authorized only up to 5 juvenile *O. mykiss* annually by electrofishing that was further restricted to River Mile 25–30 during September to November. Thus that permit is not applicable or adequate to the season, location, and fish numbers needed to conduct the electrofishing for this population estimate study. Consequently, the July 2008 survey was conducted using snorkel surveys only as provided for in the 2007 study plan. It is not anticipated that the pending permit amendment request will be resolved prior to the winter 2009 survey, as such this will be conducted using snorkel surveys. If the pending amendment request is resolved prior to July 2008, then summer 2009 surveys will be conducted using the combined method presented below.

2   FIELD SAMPLING AND DATA COLLECTION

The two-phase stratified sampling design involves snorkeling pre-selected habitat units (e.g., riffle, run, pool, etc.) multiple times in order to quantify the variance associated with density and

subsequent population estimates. Habitat units are selected using stratified random sampling where the habitat types possess a pre-determined probability of occurrence within areas where *O. mykiss* have been frequently observed during the summer in the lower Tuolumne River, extending from approximately river mile (RM) 52–40 during summers and potentially extending to near the city of Waterford (RM 30) during colder winter conditions.

In a typical Phase 1 sampling approach, primary snorkel surveys (Edmundson et al. 1968, Hankin and Reeves 1998, McCain 1992, Dolloff et al. 1996) will be conducted across a subset of all habitat units. In Phase 2, approximately 20–70% of each habitat type sampled will be randomly selected for replicated surveys by either repeated dive counts or depletion electrofishing (Reynolds 1996). Although the bounded counts methodology was developed for use in smaller stream systems (Hankin and Mohr 2001), applying the methodology to a larger system such as the Tuolumne River is feasible provided key assumptions are satisfied. A critical assumption of the bounded counts approach is that all individuals have a chance of being observed. This may not be practically attainable due to the depths of some of the in-channel mining pits and also potentially due to low visibility conditions occurring at downstream locations or due to winter-time sediment inputs during rain events. Hankin and Mohr (2001) found that their survey designs were suitable for coho salmon (*O. kisutch*), but they were less confident about applying the methodology to *O. mykiss* juveniles because the fish's furtive nature may violate the assumption that all fish have an observation probability >0. Sampling sites and methods may be modified following initial surveys because local conditions cannot be anticipated and may dictate the use of other schedules, locations, or techniques. Stillwater Sciences will notify TID, FERC, and permitting authorities if substantive changes in the study design, methods or schedule are anticipated.

2.1 Habitat Typing

On-the-ground mapping of potential habitat for *O. mykiss* will be delineated on digital ortho-rectified aerial photographs and information from previous habitat mapping efforts. Appendices A and B shows preliminary habitat units from RM 52–30 based upon habitat mapping conducted by Stillwater Sciences (2008) between La Grange Dam (RM 52) and Roberts Ferry Bridge (RM 40) (Appendix A) as well as preliminary habitat units from RM 40 to Waterford (RM 30) based upon mapping conducted by McBain & Trush (2004) and EA Engineering (1997) shown in Appendix B. The Appendix B habitat maps will be updated for flow and morphological characteristics in the field in late February and late June in each year. The final habitat maps will delineate all potential *O. mykiss* habitats according to the major types listed in Table 1, as well as transitional habitats that may be preferentially used by various size classes (i.e., pool heads, pool bodies, pool tails, run heads, run bodies, run tails, and riffles).

Table 1. Coarse scale habitat types to be used during snorkel surveys

Habitat Type	Description[a]	Approximate Depth
Riffle	Shallow with swift flowing, turbulent water. Partially exposed substrate dominated by cobble or boulder. Gradient moderate (less than 4%).	0–4 ft
Run	Fairly smooth water surface, low gradient, and few flow obstructions. Mean column velocity generally greater than one foot per second (fts^{-1}).	4–10 ft
Pool	Slow flowing, tranquil water with mean column water velocity less than 1 fts^{-1}.	>10 ft

[a]Major habitat types determined based upon observed hydraulic conditions (McCain 1992, Thomas and Bovee 1993, Cannon and Kennedy 2003)

A Geographic Information System (GIS) will be used to update and refine habitat maps prior to thorough field verification of flow, depth, and habitat conditions in the river. Within each reach, individual habitat units will be digitized as two-dimensional features of varying shapes, or polygons, where each unit is a discrete functional habitat, as defined above. This approach is consistent with the general techniques of McCain (1992), Thomas and Bovee (1993), and Cannon and Kennedy (2003) and allows a flexible approach to evaluating habitat and habitat use patterns at a scale that can be easily delineated given available data, readily depicted, and is ecologically meaningful for aquatic species.

Habitat units will be assigned a natural sequence order (NSO), starting at one which is the first unit at the upstream end of the site, and a habitat type unit number (1…N pools, runs and riffles). The maximum depth, length and width (usually at 1/3 and 2/3 of the units length) will be recorded and flagging tied at both upstream and downstream ends of units to be surveyed. Pertinent information such as date, unit number, and type is included on the flag. Lastly, the upper and lower end of each unit will be located by GPS and mapping from previous efforts will be verified or updated.

2.2 Sample Site Selection

After all potential habitat units are typed and all pertinent information recorded, a subset of each habitat unit type will be selected for single-pass snorkel surveys. Although additional units may be selected at gravel augmentation and other in-channel restoration sites (See Hypothesis 2), selection for sampling proceeds by random selection of the starting sampling unit in the upper survey section, followed by a systematic uniform sampling of the remaining units in the survey reach. For example, every 3rd, 4th or larger selection interval will be used to distribute the selected units uniformly across the survey reach.

Because the total length of river sampled affects the confidence bounds of the resulting *O. mykiss* population estimates, at least 10% of the total length of a given habitat type and a minimum of 5 units of each type will be sampled. Based upon preliminary habitat mapping and median unit lengths of various habitat types, Table 2 shows that 63 sampling units for the winter surveys will be selected from representative locations between RM 52–30 to meet the minimums above. This estimate further assumes that, since detailed habitat type mapping has not been conducted from RM 40–30, habitat type distribution and median length from RM 40–30 are similar to RM 52–40, as determined by summer 2008 habitat type mapping (Stillwater Sciences 2008). The exact number sampled will be determined after random selection of the habitat units prior to study implementation.

During summer, an estimated 35 units will be selected for single-pass snorkel survey from representative locations between RM 52–40 (Table 2). For both winter and summer surveys, the number and location of habitat units may be adjusted if initial systematic sampling does not allow the study to adequately to test Hypothesis 2.

Table 2. Estimated number of sampling units that will meet study design assumption of sampling at least 10% of the total length of a given habitat type.

Habitat Type	Total length (ft) RM 52-40[a]	Estimated total length (ft) RM 40-30[b]	Estimated total length (ft) RM 52-30	Median length (ft)[c]	# of units to be sampled Winter 2009 RM 52-30[d]	Estimated sampled Length Winter 2009	# of units to be sampled Summer 2009 RM 52-40[d]	Estimated sampled Length Summer 2009
Riffle	14,320	13,590	27,910	322	9	10%	5	11%
Pool head	619	618	1,237	106	9	77%	5	86%
Pool body	6,741	6,795	13,536	393	9	26%	5	29%
Pool tail	781	618	1,399	124	9	80%	5	79%
Run head	2,067	1,853	3,920	51	9	12%	5	12%
Run body	37,350	35,829	73,179	843	9	10%	5	11%
Run tail	2,393	2,471	4,864	54	9	10%	5	11%
Total	**64,271**	**61,775**[e]	**126,046**		**63**		**35**	

[a]From Stillwater Sciences (2008)
[b]Assumes same proportion of habitat types as from RM 52-40
[c]Assumes median habitat unit lengths from RM52-40 are proportional to median lengths along RM 40-30.
[d]Assumes at least 10% of the total length of each habitat type will be sampled; Estimates based upon 10% of the total length of a habitat type by median habitat unit length to determine a minimum number of units
[e]Actual river length from RM 40-30

2.3 Sampling Period

Winter sampling will begin in late February with systematic random selection of habitat units from RM 52-30, based upon summer 2008 maps (Appendix A) and previous habitat typing between RM 40–30 (Appendix B). Following habitat selection, Stillwater will use single-pass snorkel surveys and second phase calibration surveys within units of each type to develop uncertainty and bias estimates. Second phase sampling will be conducted using multi-pass snorkel surveys and/or depletion electrofishing methods as allowed under applicable permits (See Section 6).

Summer sampling will use habitat maps from RM 52–40 developed in summer 2008 (Appendix A). Although no additional habitat mapping is anticipated following winter 2009 surveys, habitat unit flagging will be established in advance of each snorkel survey effort and seasonal changes in habitat distribution may force revision of habitat type maps, specifically the upper and lower boundaries of habitat units and/or channel margins, prior to summer 2009 surveys.

2.4 Measurement Parameters and Sampling Methods

Multiple parameters will be measured in order to meet the objectives for this study (Table 3). Photos and GPS locations will be taken at each site, and site locations identified on GIS maps corresponding to mapped aquatic habitat units. General site information recorded at fish sampling locations will include site name, GPS coordinates, time, date, and crew member names. *In situ* water quality parameters (Temperature, dissolved oxygen, and conductivity) will be collected using a pre-calibrated multi-probe (YSI 85, Yellow Springs Instruments, Yellow Springs, OH). Underwater visibility will also be estimated into the sun and away from the sun using a Secchi disk to monitor any changes in visibility. Dissolved oxygen probes will be recalibrated at each site and checked for accuracy against concentrations measured in Winkler titrations (Grasshoff et al 1983) at the beginning and end of the sampling effort using a dissolved oxygen test kit.

Table 3. Measurement parameters and methods for snorkel surveys

Parameter	Method	Metric/Descriptor	Method Reporting Limit
Habitat Typing Attributes			
Natural sequence order (Reach ID – Habitat unit #)	N/A	A-1, A-2, A-3, …	N/A
Latitude/Longitude	Handheld GPS receiver	UTM	N/A
Habitat type	Visual estimation	See Table 1	N/A
Average unit width	Horizontal distance	meters (feet) (measured at multiple transects)	3 ft (1 m)
Average unit length	Horizontal distance	meters (feet)	3 ft (1 m)
Maximum/minimum depth	Vertical distance	meters (feet)	1 ft (0.3 m)
Bed substrate composition	Visual estimation	bedrock, boulder, cobble, gravel, organic, sand, silt	10%
Cover type	Visual estimation	none, boulder, cobble, IWM, bedrock ledges, overhead vegetation, aquatic vegetation	10%
Field Data During Snorkel Surveys			
Temperature	EPA 170.1	°C	0.1 °C
Dissolved Oxygen	SM 4500-O	mg/L	0.0 mg/L
Conductivity	SM 2510A	umhos/cm	1.0 umhos/cm
Visibility	Secchi depth	meters (feet)	0.01 m (0.1 ft)
Date/Start time/End time	N/A	Day/month/year	N/A
Number of Individuals	Visual estimation	Number	1
Fish length – snorkeling	Visual estimation	millimeter	50 mm
Fish length – electrofishing	Fork length	millimeter	1 mm
Weight - electrofishing	Electronic balance	gram	0.1 g

2.4.1   Snorkel Surveys

Snorkel surveys will be conducted during daylight hours (7:00am–5:00pm winter; 6:00am–8:00pm summer). A two phase survey design will be used to survey the seven different strata (Table 4). At the first phase, single-pass dive surveys will be conducted by a four to five person crew depending upon river flows and underwater visibility. Sampling units will generally be sampled from downstream to upstream in dive lanes using a zigzag pattern, passing fish and allowing them to escape downstream of the diver. If fish are observed to escape upstream, the diver will take care to avoid counting these fish twice. Divers will record their observations of pertinent attributes (Table 3) and numbers of *O. mykiss* and Chinook salmon (*O. tshawtscha*) observed; with fish lengths to be estimated in 50 mm size ranges using a scale model or markings on the slates to correct for underwater size distortion. After the first dive pass is completed a tab is then pulled to determine if the unit is included in the second phase of sampling.

Table 4. Preliminary sample unit selection and survey count.

	Winter 2009				Summer 2009			
	Phase I Dives		Phase II Survey		Phase I Dives		Phase II Survey	
Habitat	Initial Units	Passes	Repeat Units	Passes	Initial Units	Passes	Repeat Units	Passes
Riffle	9	1	2	2	5	1	2	2
Pool head	9	1	2	2	5	1	2	2
Pool body	9	1	2	2	5	1	2	2
Pool tail	9	1	2	2	5	1	2	2
Run head	9	1	2	2	5	1	2	2
Run body	9	1	2	2	5	1	2	2
Run tail	9	1	2	2	5	1	2	2
	Total	63	Total	28	Total	35	Total	28

The second phase of sampling collects data that will later be used to extrapolate dive counts to total population estimates by three passes of either repeated dive counts or depletion electrofishing. Ideally, if the count of *O. mykiss* from the Phase 1 snorkel survey is less than or equal to 20 individuals then three additional dive passes are made. If electrofishing is permitted, all units with a count of juvenile *O. mykiss* counts greater than 20 individuals will be surveyed by electrofishing. Lastly, occurrence of other native and non-native fish species will be recorded as presence/absence.

2.4.2   Electrofishing at Riverine Sites

If employed during the summer 2009 survey, electrofishing will be conducted by a 4 person crew during the daylight hours (6:00am-8pm) following the dive surveys. Ideally, 3-pass electrofishing will be used on all second phase dive units where the first dive pass exceeded 20 *O. mykiss*. Dive units that require electrofishing for dive calibration will be completed as soon as possible after the dive survey.

Shallow water habitat may be sampled using back pack electrofishing units while deep water habitat may be sampled using a boat electrofishing unit. Back pack electrofishing in shallow waters less than 3–4 ft depth will be conducted using two or more Smith-Root back pack electrofishers (Model LR-24 or Model 12 with 11-inch anode rings and standard "rat-tail" cathodes). Boat electrofishing may be used in deeper riverine habitats using a boat mounted Smith Root 1.5 KVA electrofishing unit. To ensure the health of all fish captured during electrofishing, all electrofishing will be conducted in accordance with NMFS (2000) electrofishing guidelines and an electrofishing logbook will be maintained and updated at each sampling site.

Depending upon river flows and depth, electrofishing will use block nets placed at the upstream and downstream ends of the unit to be fished, taking care to avoid disturbance of the unit during net set-up. Block nets will be set up where possible to prevent fish from moving out of the unit. If block nets are not feasible, then a snorkeler may be stationed at the upstream end of a unit to observe any fish moving out of the unit.

First pass electrofishing will proceed slowly and deliberately upstream from the downstream end of the unit; members of an electrofishing crew will move to the top and back down to the bottom working closely together. To maintain equal effort on subsequent passes, electrofishing time (seconds) will be recorded to allow for any adjustments in sampling effort. A fourth pass will be conducted if one of the following applies:

1. The number of *O. mykiss* caught on the 2nd pass exceeds the number of *O. mykiss* caught on the 1st pass.
2. The number of *O. mykiss* caught on the 3rd pass is greater than or equal to 25 percent of number caught on the 2nd pass.

The procedure may be modified in riffle habitats to facilitate capture of shocked fish in fast water. In the riffle strata, a pass consists of a sweep from the top to the bottom of the unit. Depending on the water velocity, block nets may or may not be set at the upstream end of riffle units.

2.4.3   Fish Handling Protocols

Any fish captured during electrofishing surveys will be processed, and information collected regarding species identification, fork length (FL, mm), weight (g), and, if applicable, notes on general condition. All fish will be rapidly retrieved using dip nets and placed immediately into aerated live wells or buckets with water. Large fish will be kept separate from juvenile fish to avoid confinement predation. Fish will be identified to species and origin (hatchery or wild stock) where possible. Fish that are weighed and measured will be anesthetized using clove oil to minimize handling stress. After all fish are identified, counted, and measured, fish will be held for approximately 10 minutes, until they show signs of "normal" swimming patterns and behavior.

2.5 Hypothesis Testing

The purpose of the proposed *O. mykiss* population surveys is to provide population size estimates over several sampling seasons of differing environmental conditions to determine habitat use and needs within the lower Tuolumne River. The surveys will be used to examine the following hypotheses:

> Hypothesis 1: Summertime distribution of suitable habitat by observed life stages of *O. mykiss* is related to ambient river water temperature.

> Hypothesis 2: Habitat use by *O. mykiss* juveniles and adults observed in the Tuolumne River occurs at the same density in both restored and nearby reference sites.

While the selection for sampling proceeds by random selection of the starting sampling unit in the upper survey section, followed by a systematic uniform sampling of the remaining units in the survey reach, additional units adjacent to or near restoration sites may be non-randomly selected to provide treatment and control locations to test Hypothesis 2, especially during winter 2009 surveys when low ambient river water temperatures obviate the need to test Hypothesis 1.

2.6 Field Work Notification

To ensure field staff safety and to satisfy scientific collecting permit requirements, the parties listed in Table 5 will be notified in advance of the proposed sampling in as required to confirm sampling dates.

Table 5. Field Work Notification

Contact	Affiliation	Address	Phone and Email
Tim Ford	TID	333 East Canal Dr. Turlock, CA 95380	209.883.8275 tjford@tid.org
Tim Heyne	CDFG	P.O. Box 10 La Grange, CA 95329	209.853.2533 x1# theyne@dfg.ca.gov
Jeffery Jahn	NMFS	777 Sonoma Ave. Rm 325 Santa Rosa, CA 95404	707.575.6097 Jeffrey.Jahn@noaa.gov

Prior to mobilization, planned river operations by the Districts will be checked to determine if fish sampling would be safe under the anticipated flow and all parties will be notified of any delay or modification to the sampling schedule.

3   QUALITY ASSURANCE

The objective of data collection for this Project is to produce data that represent as closely as possible, *in situ* conditions of the Tuolumne River with respect to river flow conditions, water quality, abundance and habitat use by *O. mykiss*. To meet this objective, field sampling, sample preparation, and analysis will follow general guidelines outlined in USEPA (2002) by ensuring that:

- the project's objectives, hypotheses and data quality objectives are identified and agreed upon,
- the intended measurements and methods are consistent with project objectives,
- the assessment procedures are sufficient for determining if data of the type and quality needed and expected are obtained, and
- any potential limitations on the use of the data can be identified and documented.

Aquatic environments are inherently variable, but management decisions must be based on a data from a limited number of locations and often collected in short time periods. How well the information collected represent the reach or river-wide fish population depends upon a systematic approach to quality assurance.

3.1 Data Quality Objectives for Measurement Data

The data quality parameters used to assess the acceptability of the data are precision, accuracy, representativeness, comparability, and completeness. Precision measures the reproducibility of measurements under a given set of conditions. Analytical precision is limited to water quality and physical habitat characteristics (Table 6). Accuracy is an expression of the degree to which a measured or computed value represents the true value. Field accuracy is controlled by adherence to sample collection procedures.

Table 6. Data quality objectives for field parameters

Parameter	Units	Accuracy	Precision	Completeness
Dissolved Oxygen	mg/L	$\pm$ 0.5	10%	90%
Temperature	^{o}C	$\pm$ 0.5	5%	90%
Conductivity	umhos/cm	$\pm$ 5%	$\pm$ 5%	90%
Depth	meters	$\pm$ 0.2	N/A	N/A
Visibility (Secchi)	meters	$\pm$ 0.05	N/A	N/A

- Representativeness expresses the degree to which data accurately and precisely represent an environmental condition. For this study, monitoring site selection will be conducted based on physical habitat attributes. Additionally, specific measurement parameters have been identified as relevant based on numerous studies indicating factors associated with species distribution.

- Comparability expresses the confidence with which one data set can be evaluated in relation to another data set. For this biological assessment, comparability of data will be established through the use of standard analytical methodologies and reporting formats.

- The project goal for completeness, a measure of the amount of data that is determined to be valid in proportion to the amount of data collected, will be 90% for analytical water quality parameters. The data quality objective for completeness for all components of this study is 90%.

3.2 Training Requirements/Certification

Specialized training is required for the proposed sampling activities, however none of the sampling activities require outside certification from an agency or another entity. Required permits for biological sampling are discussed in Section 5. Field crews will be staffed by a variety of qualified personnel, which due to the nature of extended field activities, will necessarily be rotated in and out of the field.

3.3 Instrument/Equipment Testing, Inspection, and Maintenance Requirements

To ensure proper equipment performance in the field, maintenance and operational procedures, including preventative maintenance, will be performed on all YSI multiprobes (temperature, dissolved oxygen, and conductivity). YSI maintenance will be recorded in a logbook with the date the maintenance was performed and the initials of the technician. When the instruments are not deployed, the calibration or storage cup will be used to protect sensors from damage and desiccation.

3.4 Instrument Calibration and Frequency

Field probes used for field sampling will be calibrated prior to use, midway through each sampling event, and at the end of each sampling event. Measurement devices for conductivity will be checked against a standard whose source is different than that selected for calibration. Dissolved oxygen will be checked against aerated water whose oxygen content is established by the Winkler method (Grashoff et al 1983). Temperature does not require calibration because of the unvarying nature of the temperature sensor and its conditioning circuitry.

3.5 Reconciliation with Data Quality Objectives

If data do not meet the project's specifications, the following actions will be taken. First, the task leaders working with the field crew leaders (in some cases they will be the same person) will review the errors and determine if the problem is equipment failure, calibration/maintenance techniques, or monitoring/sampling techniques. They will suggest corrective action. If the problem cannot be corrected by training, revision of techniques, or replacement of supplies/equipment, then the task leaders will review the data quality objectives (DQOs) and determine if the DQOs are feasible. If the

specific DQOs are not achievable, they will determine whether the specific DQO can be relaxed, or if the parameter should be eliminated from the monitoring program.

3.6 Data Management

All field data will be amassed in a quality-checked database and summarized. QA checks will be applied to all data before data entry and data will be stored on Stillwater Sciences servers. Full backup of data from all offices is done on a weekly basis, while differential backup (files that have changed since the last full backup) is done on a nightly basis. The backup process is accomplished with a Fast Tape Library and backup processes are completed during off-peak hours. Two sets of tapes are taken offsite by two Information Technology (IT) staff members on a weekly basis to ensure recovery in case of failure or catastrophe.

4   DATA ANALYSIS

Data analysis will be conducted to summarize *in situ* water quality and fish counts in each sampling strata. Bounded counts or depletion estimators will be used to determine populations and linear density for each sampled unit, together with estimates of uncertainty. In addition to comparisons of fish density between sampling strata, the density estimates and uncertainties will be propagated across the unsampled areas for an overall population estimate. Exploratory multiple regression analysis will also be used to determine relationships between fish density and recorded habitat variables.

5   REPORTING

A data report will be prepared for use with permitting authorities that includes: date, time, and location of sampling activities; species and number of species collected; and a copy of field data sheets. Results of the winter 2009 surveys will be transmitted to TID electronically within three weeks of the survey completion (April/May 2009). A client review draft of the technical report covering the results of both winter and summer 2009 surveys will be submitted to TID by August 24, 2009. Assuming an internal and Agency review comments are received within one and three weeks of issuance of the client review and Agency review drafts, respectively, the Agency review draft will be available by September 8, 2009 and final report will be complete by October 16, 2009.

6   PERMITTING REQUIREMENTS

Stillwater Sciences will maintain the following permits to sample fish populations that may be present:

- NMFS Section 10(a)(1)(A) permit 1282
- California Department of Fish and Game individual Scientific Collection Permits.

A NMFS Section 10(a)(1)(A) permit 1282 has been obtained and all NMFS guidelines (e.g., notification, data gathering, preservation) will be followed if any Central Valley steelhead are captured. Under that existing NMFS permit, electrofishing is limited to an authorized incidental take of 40 juvenile *O. mykiss* and the <5% unintentional mortality limit, and no adults. An amendment to the sampling description was submitted to NMFS on June 2, 2008 with increased take limits for handling electrofishing of 100 adults and 200 juveniles at an unintentional mortality rate of <10%. Mr. Jeffrey Jahn of NMFS will be notified at least two weeks prior to applicable sampling to confirm

sampling dates and locations. Electrofishing under an amended permit will be suspended in the event that the authorized incidental take limits were exceeded and all subsequent calibration surveys would be made by repeat dive surveys. Annual reporting will be provided to Mr. Jeffrey Jahn of NMFS by March 1, of each year.

CDFG Scientific Collecting Permits (SCPs) will be maintained for species potentially present in the project area. CDFG guidelines (e.g., notification, data gathering, and preservation) will be followed if special-status species are captured and the CDFG 24-hr dispatch (916.446.0045) will be notified should unrelated events result in fish kills.

No intentional mortality or removal of special-status species from the wild is included in this study plan. In the event unintentional mortality occurs beyond the take permit limits, NMFS staff will be contacted within 24 hrs and a fin-clip will be provided to the Salmonid Genetic Repository. CDFG will also be contacted to determine the disposition of the individual specimen and whether the individual may be retained for otolith analysis.

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Appendix A
Lower Tuolumne River Habitat Mapping and Habitat Types from RM 52-40 (Stillwater Sciences 2008)

Appendix B
Preliminary Habitat Mapping and Habitat Types in the lower
Tuolumne River from RM 40-30

Data sources: EA Engineering 1997,
                 McBain & Trush 2004,
                 USDA NAIP Imagery