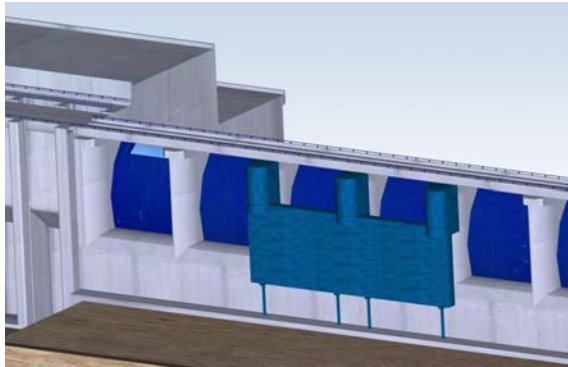


**Study Plan for Using Acoustic Tags to
Track the Fine-Scale Movements of
Juvenile Salmonids in Relationship to a
Top-Spill Bulkhead at Priest Rapids Dam and the
Future Unit By-Pass at Wanapum Dam in 2008**



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Appendix A LGL Limited Standard Field Operating Procedures A-1

1.0 Introduction

The Public Utility District No. 2 of Grant County (Grant PUD) has devoted considerable effort to increasing the survival of juvenile Pacific salmon and steelhead (*Oncorhynchus spp.*) at Wanapum and Priest Rapids dams. Grant PUD initiated consideration of non-turbine passage (top spill fish passage) prior to the issuance of the interim Biological Opinion for the Priest Rapids Project (NOAA 2004). Alternative passage routes (non-turbine) are presented in Voskuilen et al. 2003. Hydraulic modeling, Computational Fluid Dynamics (CFD) modeling, mechanical engineering, and evaluation of previous fish studies were all necessary in the development of the Wanapum Future Unit Fish Bypass (WFUFB) for Wanapum Dam and Top-Spill concept for Priest Rapids Dam. The Fish Passage Design Team determined the information acquired from a behavior/approach study was needed for validation of the WFUFB concept and final production design and is further needed for the development of the Top-Spill fish bypass at Priest Rapids Dam.

On May 3, 2004, the National Marine Fisheries Service (NMFS - then referred to as NOAA Fisheries) issued its Biological Opinion of the effects (Biological Opinion) of the proposed action on listed species, in accordance with Section 7 of the Endangered Species Act of 1973 as amended (16 USC 1531 et seq.), regarding the Federal Energy Regulatory Commission's proposed action amending Grant PUD's existing license for the Priest Rapids Hydroelectric Project (Project) to authorize implementation of an Interim Protection Plan for listed anadromous salmonids.

Reasonable and Prudent Alternatives (RPAs) No. 3 and 13 of the Biological Opinion discusses Alternative Top-Spill Concepts for fish passage at Wanapum and Priest Rapids dams; respectively. The completion of the WFUFB in early 2008 will be the final phase of fulfilling RPA 3 (Top-Spill through Future Units). In fulfilling RPA 13 of the Biological Opinion (Alternative Top Spill Concepts for Priest Rapids Dam), the Fish Passage Design Team has determined that similar information regarding fish behavior, including approach paths toward the dam (Priest Rapids), will be useful for evaluating design considerations relative to a top-spill fish passage route at Priest Rapids Dam

The proposed study will provide the information on fish behavior and location required to address the following questions associated with top-spill fish passage at Priest Rapids Dam:

- Will the addition of bottom spill increase the effectiveness of the surface spill;
- Will migrating smolts reject an opening (surface spill) where acceleration upstream of the opening is uncontrolled from the ambient velocities in the forebay, up to velocities that trap the fish;
- Do different species of smolts approach and pass the dam in similar or different patterns.

This study plan will also provide approach, behavioral and relative survival estimates for juvenile salmonids as they approach the WFUFB, during the first season of 20 kcfs operation.

The study plan approach for 2008 is to undertake an evaluation of the WFUFB and the Priest Rapids Dam prototype surface spill passage route (a top-spill bulkhead located at spillbays 19 & 20, with the addition of bottom-spill at spillbay 21), to monitor fish behavior and location in the

forebay to address unknown aspects of fish passage. Information collected from this evaluation will guide the design process for a non-turbine passage route at Priest Rapids Dam. This field study is one component of a comprehensive assessment of design alternatives for non-turbine fish passage at Priest Rapids Dam. Other components include hydraulic and CFD modeling, and mechanical engineering.

2.0 Study Objectives

In order to obtain data that will advance the design of a fish bypass at Priest Rapids Dam, the following specific objectives have been defined for the 2008 studies:

1. Determine how yearling Chinook approach Priest Rapids Dam and are distributed in passage across the powerhouse and spillway under different operational configurations;
2. Determine the fish passage efficiency (FPE) and fish collection efficiency (FCE) of the Priest Rapids top-spill with the addition of bottom spill;
3. Compare three-dimensional time-stamped records of the approach and passage at Priest Rapids Dam from acoustic tagged steelhead and sockeye that will be passing Priest Rapids Dam as a result of Public Utility No. 1 of Chelan County (Chelan PUD) fish studies upstream of the Priest Rapids Project;
4. Evaluate migrating smolt behavior as they approach Wanapum Dam.

The goal of the above objectives is to answer critical questions regarding the placement and design of the proposed future fish bypass at Priest Rapids Dam. The potential FPE for a bypass would be estimated through a review of the three-dimensional behavior and approach data. Forebay entrance design details will be developed based on an analysis of rejection rates observed at the top-spill opening.

To test study objectives, the top-spill bulkhead will be located at spill bays 19 and 20 of the Priest Rapids spillway. The top-spill will consist of a free surface overflow through a notch in a bulkhead (Figure 1). The top-spill will pass 16 kcfs through an 80 ft opening, nominally 13-1/2 ft deep (sill elevation 473 ft). In addition to this 16 kcfs surface flow, 7 kcfs of bottom-spill will be added via the opening of spillbay 21. This addition of bottom-spill is expected to increase the FPE and FCE of the top spill bulkhead, based on hydraulic modeling work that was completed by the Fish Passage Design Team at the Iowa Institute of Hydraulic Research (IIHR) in Iowa City, IA.

The primary method to achieve the study objectives will include acoustic-tagging 1,000 yearling Chinook smolts of run-of-river origin and tracking them through the forebay of Wanapum Dam, with the WFUFB (Figure 2) in operation, and on through the Priest Rapids reservoir to the Priest Rapids forebay, where they will pass Priest Rapids Dam via top-spill, bottom-spill, or powerhouse passage routes.

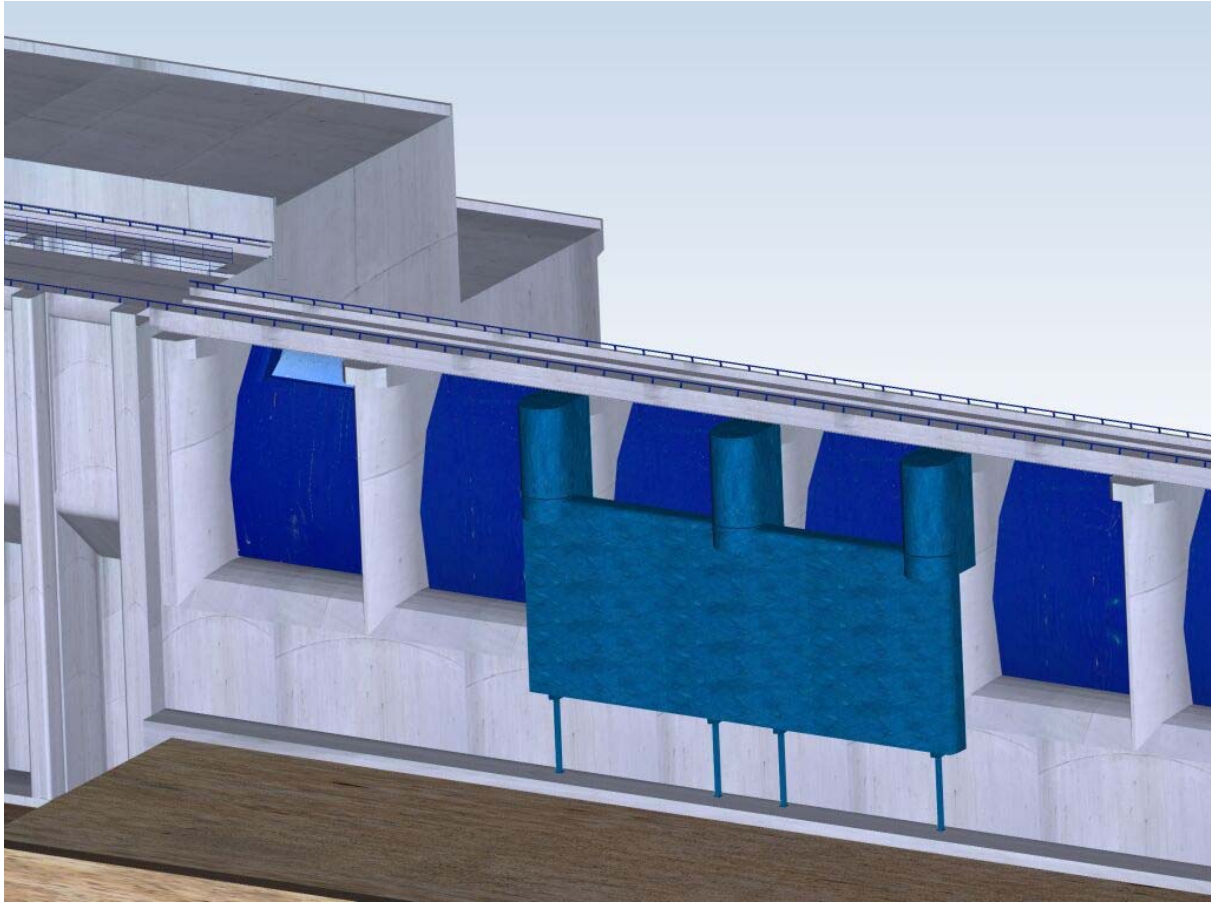


Figure 1 Conceptual drawing of Priest Rapids top-spill bulkhead looking downstream. Spillbay 21 is to the left of the bulkhead.

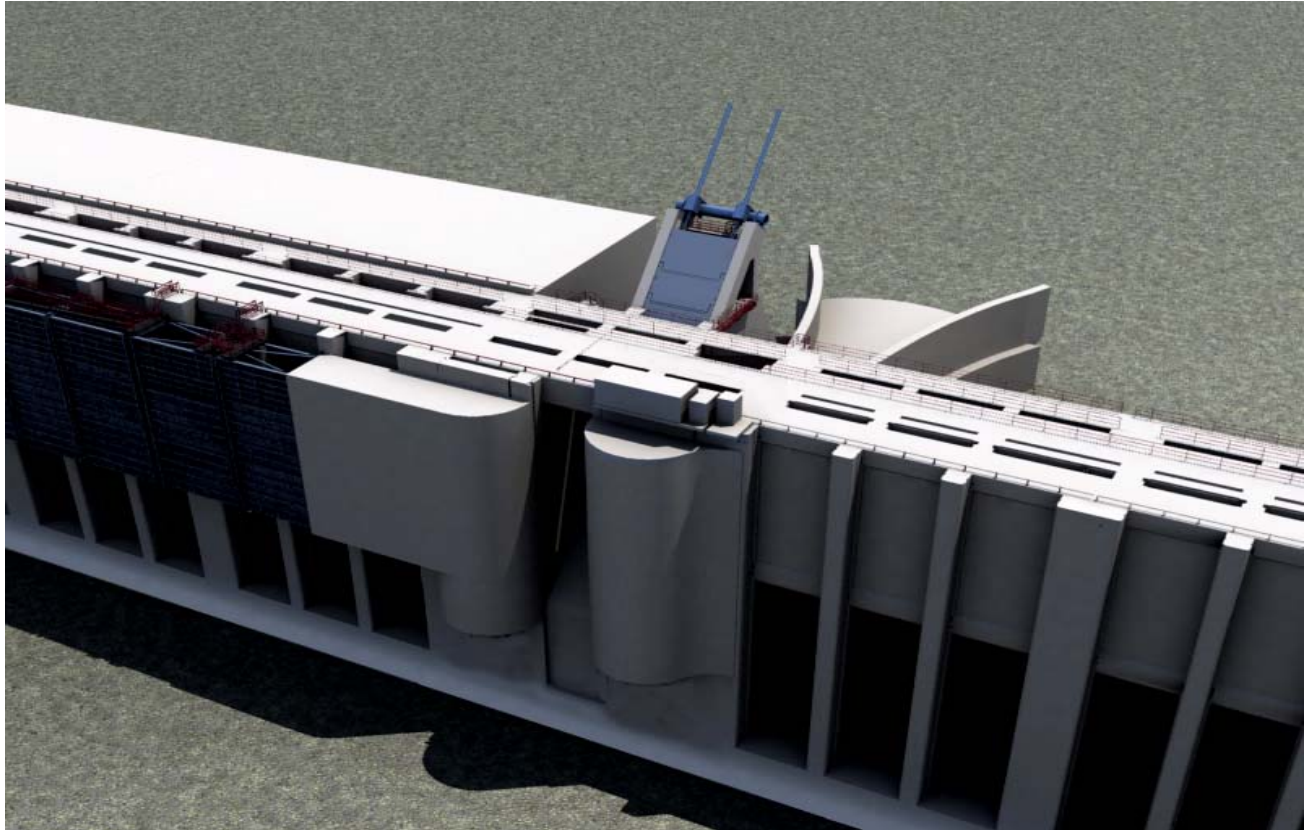


Figure 2 Conceptual drawing of the WFUFB at Wanapum Dam looking downstream.

3.0 Site Description

Priest Rapids Dam (Figures 3 and 4) is located on the Columbia River at RM 397, 18 miles downstream of Wanapum Dam and approximately 70 miles upstream of the confluence of the Snake and Columbia rivers. Normal maximum forebay elevation is 488 ft above mean sea level (MSL), and the normal operating head is 78 ft.

The dam has a 1,025 foot-long powerhouse located at the northeast end, oriented approximately perpendicular to river flow, and a 1,152 foot-long spillway on the southwest end of the dam. The Priest Rapids development contains ten vertical shaft turbines, with a total capacity of 855 MW (85.5 MW each) and connecting to ten generators having a total rated capacity of 955.6 MW. Each turbine unit has three intake slots.

The spillway contains 22 Tainter gates, each 40-ft wide and 50-ft tall. These gates rest on the ogee at 50 ft in depth (elevation 436 ft), and when opened they produce a submerged bottom-spill at a depth of 60 ft. Each spill gate passes an average of 6-10 kcfs under normal operating conditions. A surface sluiceway is located between the powerhouse and spillway (TG-22) and provides opportunity to pass water downstream via a surface opening. The sluiceway opens from the top, as opposed to the spillway Tainter gates, and the discharge of the sluiceway is approximately 2 kcfs.

In 2006, a surface flow prototype top-spill bulkhead was installed for testing of fish behavior and passage efficiency at Spillbays 19 and 20 (**Error! Reference source not found.** and 5). The

prototype is 100-ft wide and typically spills up to 16 kcfs (8 kcfs per spill bay) of surface water. Similar to the sluiceway, the top-spill gates open from the top to release water downstream.

Wanapum Dam (Figure 3) is located on the Columbia River at RM 416, 18 miles upstream of Priest Rapids Dam and 38 miles downstream of Rock Island Dam. Typical operational elevation of the forebay is 570 ft above MSL and the depth of the forebay at the powerhouse is approximately 110 ft (460 ft relative to elevation 570 ft). Normal operating head is 80 ft. The dam has a 1,540 foot-long powerhouse, oriented approximately perpendicular to river flow, and an 832 foot-long spillway at approximately a 45° angle to flow. Wanapum Dam has two fish ladders, one located at the east end of the powerhouse and the other located at the west end of the spillway.

The powerhouse has 10 Kaplan turbine units, numbered from north to south. Three of the original turbines have been replaced with a new advanced turbine design, referred to as the “advanced turbine.” Both original and advanced are Kaplan designs. To date, turbine units W4, W8 and W10 have been replaced with advanced turbines and turbine unit W9 is scheduled to be replaced. Upon completion of the turbine upgrades, estimated to be in 2012, the Wanapum development will include of ten Advanced Turbine Units with a total rated capacity of 1,125 MW (112.5 MW each), connecting to ten generators with a total rated capacity of 1,038 MW (103.8 MW each); The purpose of replacing the original turbines with the advance design turbines is to improve fish passage survival and increased turbine efficiency.

There are additional intake structures for six potential future units in an area between the powerhouse and spillway. One of the six potential future units, Future Unit 11, has been modified into the Wanapum Future Unit Fish Bypass (WFUFB), a surface flow bypass for the passage of downstream migrant salmonids (Figure 6). The bypass opening, positioned in the forebay directly south of turbine unit W-10, is 18.5 ft wide and has an adjustable depth of up to 75.8 ft. The total surface spill is adjustable and was designed at four increments of 5 kcfs, 10 kcfs, 15 kcfs, and 20 kcfs. The total length of the bypass design is 290 ft and the exit chute width increases to 90 ft. The exit chute is elevated and curved to evenly spread the discharge flow and minimize total dissolved gas in the tailrace.

The spillway contains 12 Tainter gates, each 65-ft tall by 50-ft wide. These gates open at the bottom to produce submerged spill at a depth of 65 ft. A surface flow spillway bulkhead is in place at Spillbay 12. A surface-skimming sluiceway is located at the north end of the spillway. It is 20-ft wide and normally spills up to 10 ft of surface water per second (1.9 kcfs).

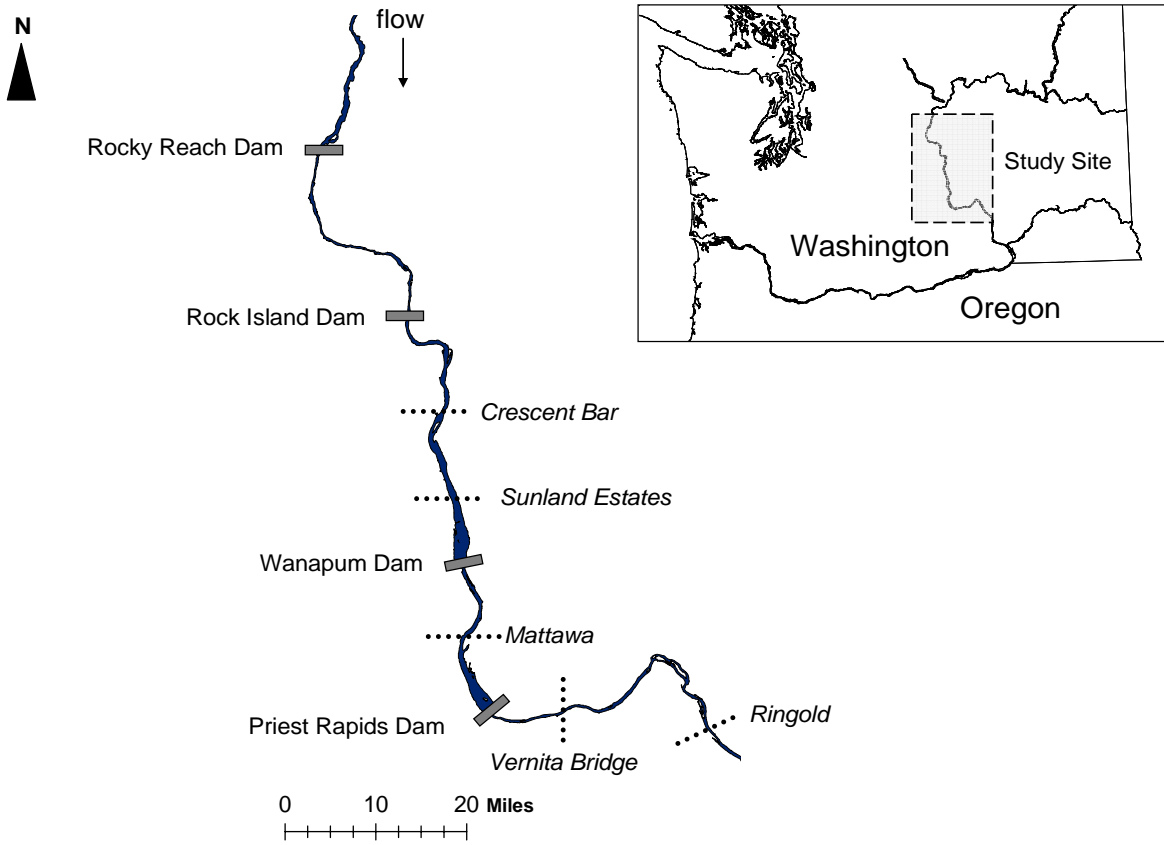


Figure 3 Plan view of Wanapum and Priest Rapids dams located at RM 416 and 397, respectively.

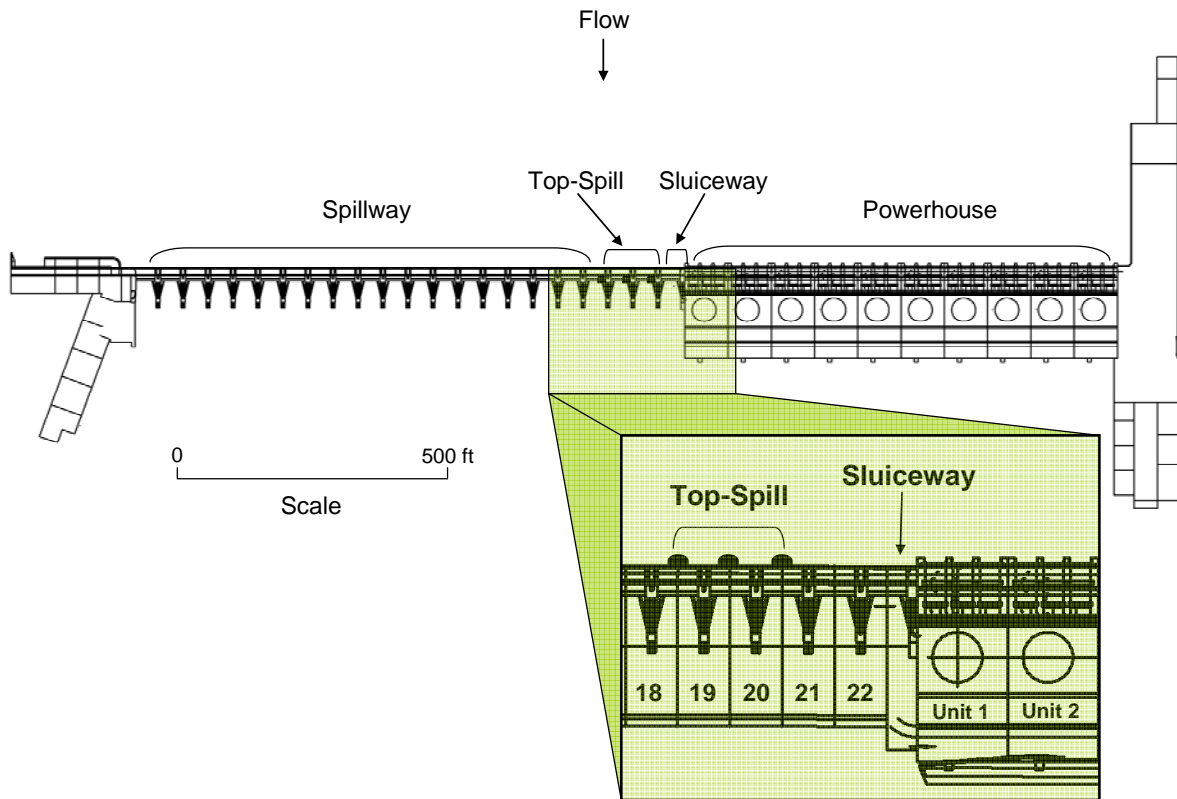


Figure 4 Plan view of Priest Rapids Dam. The prototype top-spill bulkhead at Priest Rapids Dam located at Spill Bay 19 and 20. The sluiceway is located between the junction of Turbine Unit 1 and the spillway.

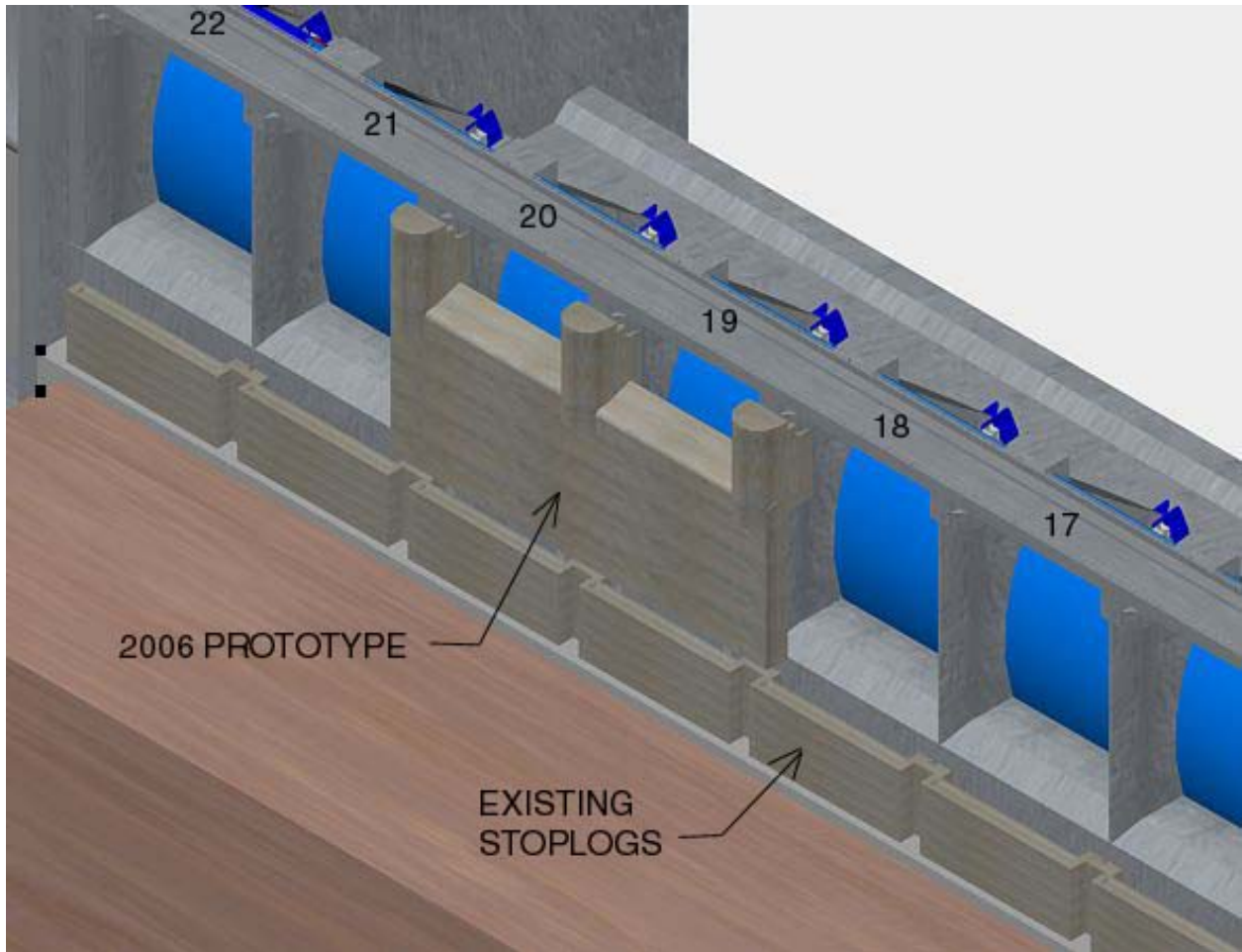


Figure 5 Schematic drawing of the Priest Rapids Dam top-spill prototype bulkhead, installed at Spill Bays 19 and 20, near the junction of the spillway and powerhouse.

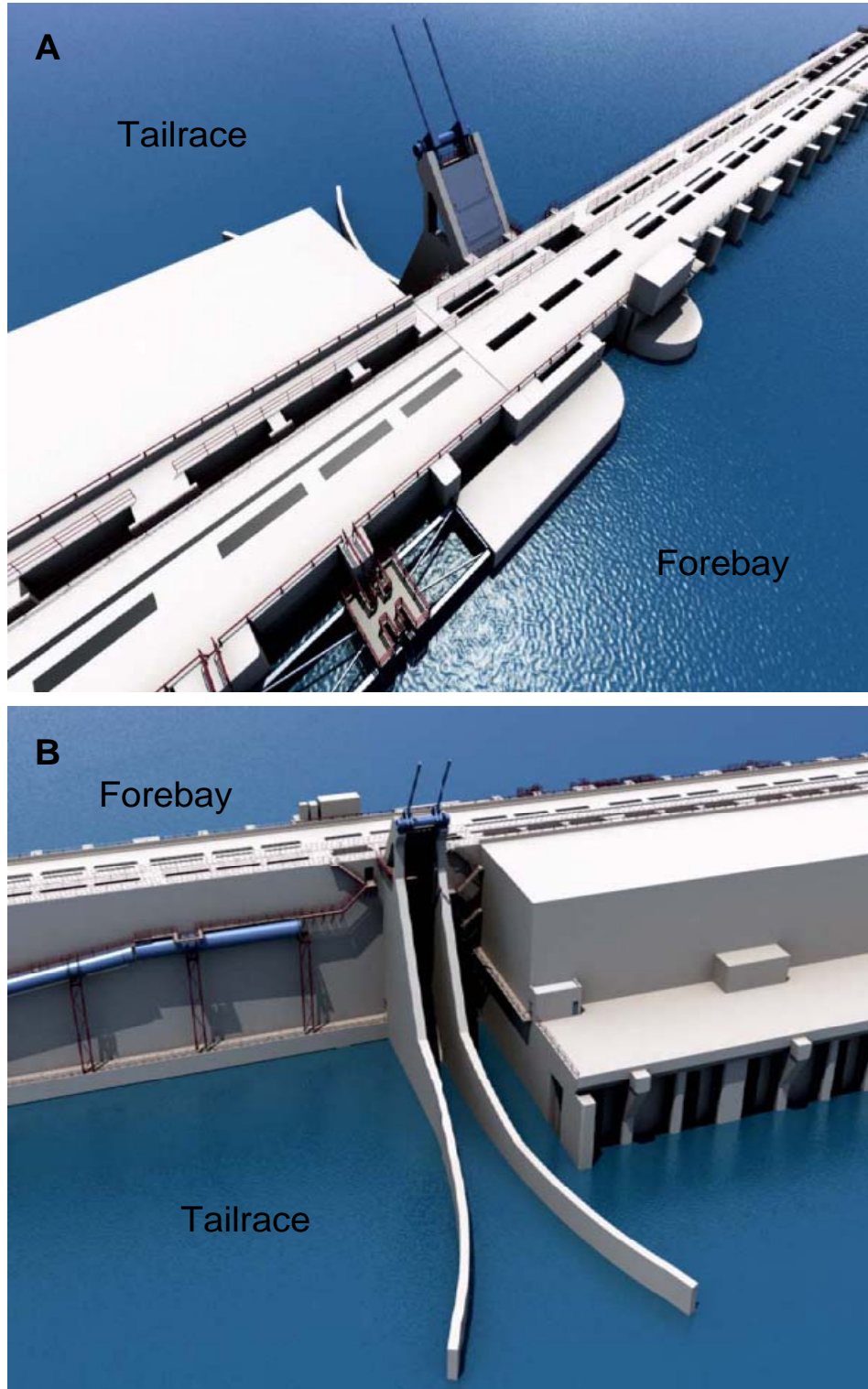


Figure 6 Wanapum Future Unit Fish Bypass Project (WFUFB) upstream view from the forebay (A) and downstream view from the tailrace (B).

4.0 Methods

In 2004 HTI conducted acoustic tracking tag studies to assess the effectiveness of top-spill for passing juvenile Chinook, steelhead, and sockeye at Wanapum Dam. These studies and similar studies conducted 1998-2004 at Rocky Reach and Rock Island dams and other dams (Table 1) tracked tagged juvenile salmonids in three dimensions with sub-meter resolution.

Table 1 Northwest studies conducted with HTI Model 290 Acoustic Tag Tracking Systems and Model 795 Acoustic Tags.

Name	Year(s)	Study Type(s)
Bonneville Dam	1999	Three-dimensional smolt tracking
Chittenden Lock and Dam	2002, 2003	Three-dimensional adult Chinook tracking
Cowlitz Falls Dam	2000, 2002	Three-dimensional smolt tracking
Grand Coulee Dam	2000, 2001, 2002, 2003, 2004	Three-dimensional smolt tracking
Lower Granite Dam forebay	1999, 2000, 2001, 2002, 2003	Three-dimensional smolt tracking
Lower Granite Dam tailrace	2002	Three-dimensional smolt tracking
Mayfield Dam	2001	Three-dimensional smolt tracking
North Fork Dam	2001	Three-dimensional smolt tracking
Priest Rapids Dam	2004	Presence/absence tagged smolts
Rock Island Dam	2001, 2002, 2003, 2004	Three-dimensional smolt tracking; smolt survival
Rocky Reach Dam	1998, 1999, 2000, 2002, 2004	Three-dimensional smolt tracking, smolt survival
Swift Dam	2002	Three-dimensional smolt tracking
The Dalles Dam	2004	Three-dimensional smolt tracking
Upper Baker Dam	2002	Three-dimensional smolt tracking
Wanapum Dam	2004 (2)	Three-dimensional smolt tracking

The acoustic-tag methods used to monitor tagged fish at the various detection sites in 2008 will be very similar to those employed at Priest Rapids Dam in 2006 and 2007 (Timko et al. 2006, Timko et al. 2007 *draft*) and at Wanapum Dam in 2004 (Robichaud et al. 2005), as well as at Rocky Reach Dam (HTI 1997, Steig 1999a, Steig 1999b, Steig et al. 1999, Steig 2000, Steig and Timko 2000, Steig et al. 2001), and at Rock Island Dam (Steig et al. 2002, Skalski et al. 2003a, Skalski et al. 2003b, Skalski et al. 2005, Steig et al. 2005).

4.1 Study Design

One thousand (1,000) acoustic-tagged yearling Chinook will be released at the Vantage Bridge, approximately 5 miles upstream from Wanapum Dam. Information acquired from these fish will be used for fine-scale three-dimensional tracking of movement near the WFUFB, and approach, behavior, FPE and FCE information as it relates to the Priest Rapids top-spill.

Detection histories for the WFUFB will be collected at Wanapum Dam (Figure 7) and at two sites downstream of the dam that were used during the 2006 and 2007 studies (Mattawa at RM 409; Figure 3), and the other at Priest Rapids Dam (Figure 8).

Detection histories for the Priest Rapids Top-Spill Bulkhead will be monitored as during the 2006 and 2007 studies (Figure 8) for forebay information and also at two sites downstream of the dam, south of Priest Rapids Dam, at the Vernita Bridge (RM 388) and in the Hanford Reach near the Ringold Hatchery, at RM 384 (Figure 3). To verify the operating life of the tags, a life test of 50 tags will be conducted. These tags will be surgically implanted into test fish and held (tested) in continuously cycled river water, similar to the fish holding tanks.

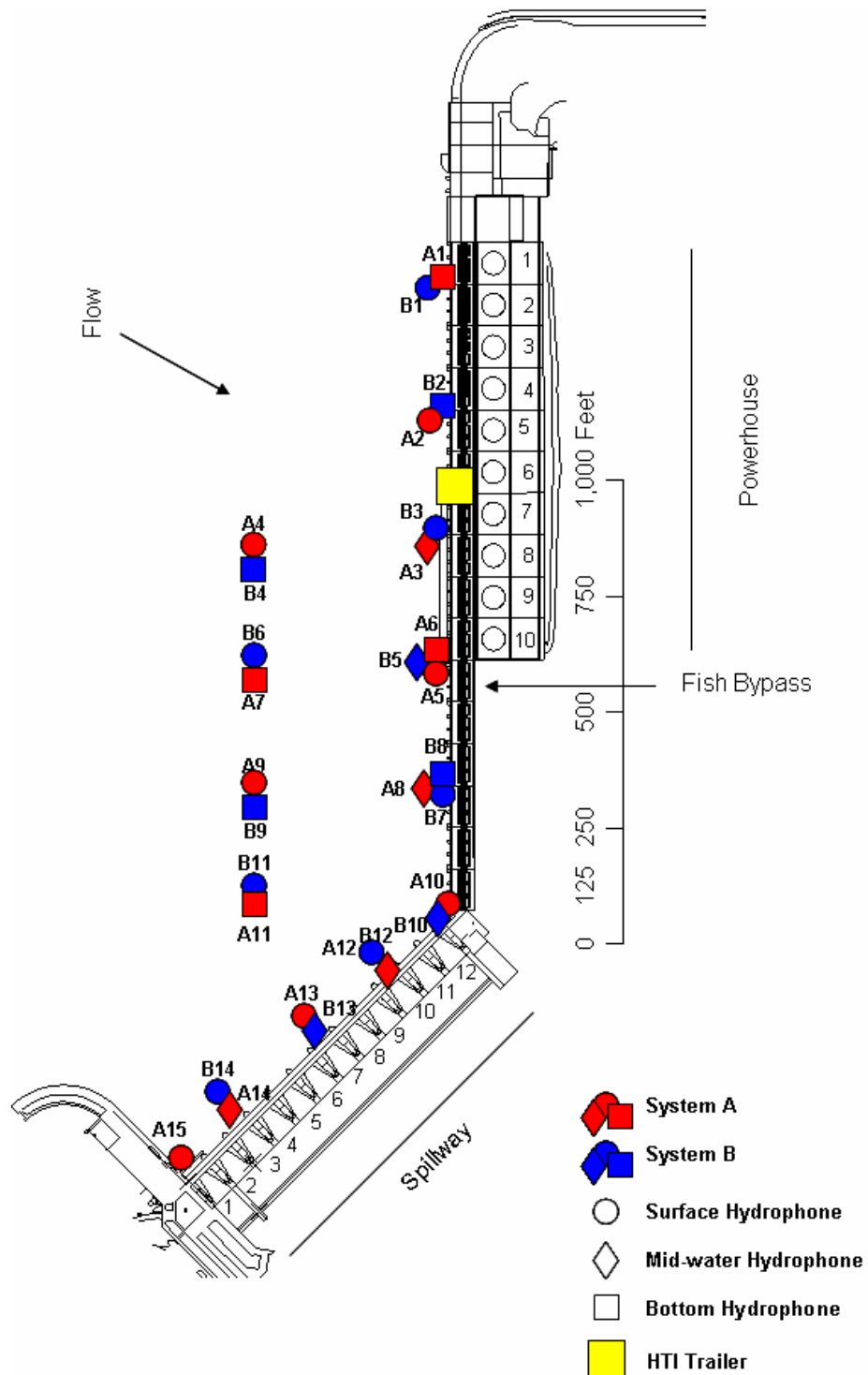


Figure 7 Wanapum Dam (RM 416) showing the proposed hydrophone locations for the spring 2008 acoustic-tag studies.

Priest Rapids – Hydrophone Array

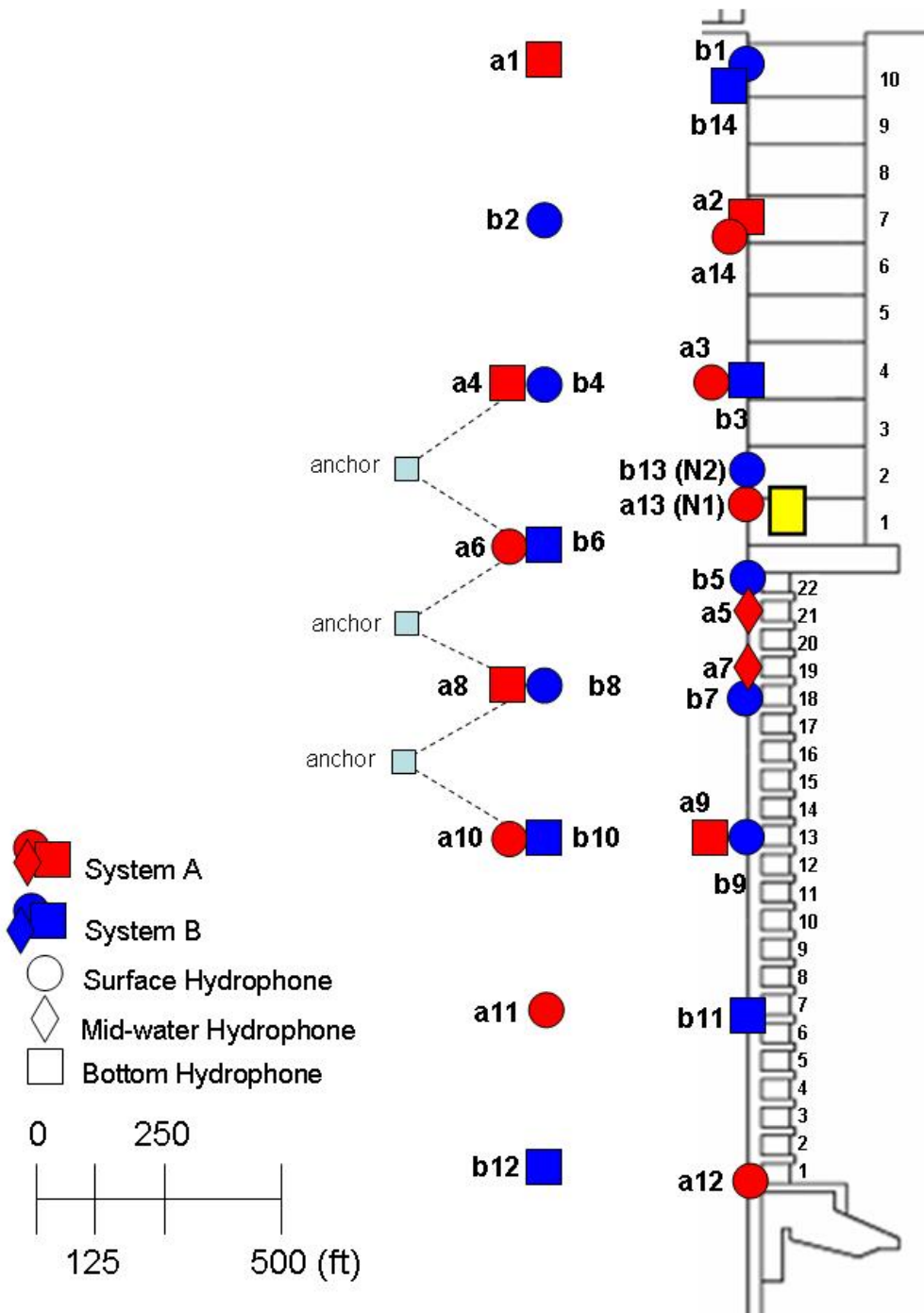


Figure 8 Priest Rapids Dam (RM 397) showing the proposed hydrophone locations for the spring 2008 acoustic-tag studies.

- 4.1.1 Task 1: Estimate the fish passage efficiency (FPE) and fish collection efficiency (FCE) of the Priest Rapids top-spill with the addition of bottom-spill

Acoustic-tagged and released (at the Vantage Bridge) yearling Chinook will be tracked in three dimensions as they approach Priest Rapids Dam, using the same acoustic-tag tracking techniques used at Priest Rapids Dam in 2006 and 2007 (Timko et al. 2006 and Timko et al. 2007 *draft*). In addition to the 16 kcfs surface-drawn spill that is generated by the Priest Rapids Top-Spill Bulkhead, 7 kcfs of bottom-drawn spill will be generated via the opening of spill bay 21 for fish passage.

- 4.1.2 Task 2: Compare three-dimensional time-stamped records of the approach, behavior and passage at Priest Rapids Dam from acoustic tagged steelhead and sockeye that will be passing Priest Rapids Dam as a result of Chelan PUD fish studies upstream of the Priest Rapids Project and Grant PUD's steelhead survival study.

Chelan PUD-tagged and released sockeye smolts and acoustic-tagged steelhead from Grant PUD's concurrently conducted survival study approaching Wanapum and Priest Rapids dams will be tracked in three dimensions (3-D) as they near the WFUFB and Priest Rapids Top-Spill Bulkhead using the same acoustic-tag tracking techniques used at Wanapum Dam in spring 2004 (Robichaud et al. 2005) and Priest Rapids Dam in 2006 and 2007 (Timko et al. 2006 and Timko et al. 2007 *draft*).

- 4.1.3 Task 3: Evaluate migrating smolt behavior as they approach Wanapum Dam.

Same as above: Section 4.1.2

4.2 Acoustic-Tag Receiver System Design

The HTI Model 290 Acoustic Tag Tracking System is comprised of five components: the system receiver, hydrophone, hydrophone cable, acoustic tag, and a personal computer. The Model 290 System detects the transmitted signal of an acoustic tag as each acoustic tagged fish migrates downstream through the detection arrays. Once the tag is detected, the arrival time is recorded for each hydrophone as it approaches and passes through each detection array. At the 3-D hydrophone array located at Priest Rapids and Wanapum dams the differences in arrival times based on signals from an individual tag on each hydrophone are used to calculate a 3-D position following the methods of Ehrenberg and Steig (2002, 2003).

4.2.1 Priest Rapids Dam

A total of 29 hydrophones will be deployed at Priest Rapids Dam to detect the presence, 3-D swimming pathway, and exit location of acoustic tagged smolts. The primary focus of 3-D positioning at Priest Rapids Dam will be at the prototype top-spill bulkhead. Therefore, more hydrophones will be deployed in this vicinity. Each independent system used 14 (System A) to 15 (System B) hydrophones, with the location of hydrophones for both systems overlapping (Figure 8).

4.2.2 Wanapum Dam

A total of 29 hydrophones will be deployed in the forebay of Wanapum Dam to detect the presence and exit location of acoustic tagged smolts. Each independent system used 14 or 15 hydrophones, and the location of hydrophones for both systems was overlapping (Figure 7).

4.2.3 Acoustic-Tags

For this 2008 study, the HTI *Model 795E Acoustic-tag* will be used. These tags will be approximately 6.8 mm in diameter by 18 mm long, and weigh 1.5 g in air, with an average operating life of 32–36 days, based on 2003-2006 tag life tests reported by Skalski et al. (2003b, 2005, 2006) and Steig et al. (2005). Ping rate, pulse width, and individual tag ID will be programmed in the field. Up to 50,000 individual tags can be uniquely coded. For *Model 795E Acoustic Tags* the transmission rate (i.e. ping rate) can be set from 50 pings/sec to 1 ping every 16 sec, with pulse widths from 0.5 msec to 5 msec, with either a standard CW pulse or an encoded pulse. For this 2008 study, the ping rate will be set at 1 ping per second and a pulse width of 1.0 msec. The ping rates associated with acoustic-tagged fish from other on-going studies (i.e. Chelan PUD sockeye and Grant PUD steelhead survival study) will be set at 1 ping every 4-8 sec.

4.3 System Deployment

The ATR systems and equipment require installation and testing in the field at each site. HTI will be responsible for these installations with the assistance of Grant PUD personnel. The installation of some of the hydrophones will require the assistance of Grant PUD mechanics, and possibly divers.

4.4 Fish Handling

4.4.1 Fish Collection and Holding

Grant PUD will contract separately with LGL, Limited to handle, tag, hold and release tagged smolts. Fish collection, handling and tagging will be follow the procedures employed during the 2007 (Timko et al. 2007. draft) acoustic-tag tracking study conducted at Wanapum and Priest Rapids dams. **[The source of fish (yearling Chinook) for conducting this behavior study is still pending, waiting for final review of the 2007 Fish Source Comparison Study that was conducted to evaluate a source of test fish for Grant PUD's studies.]**

4.4.2 Tagging

One day prior to the surgical implantation of transmitters, the tags were programmed by HTI personnel and verified functional before being received by LGL. After tags were verified functional, they were soaked in a diluted germicide (Germiphene, a bactericidal, fungicidal and virucidal diluted at 16 ml of Germiphene to 1 L of water) for 20 minutes the evening before surgical implantation (Appendix A).

The surgical implantation procedures used will be based on the methods outlined in Adams et al. (1998), and Martinelli et al. (1998), and used during the acoustic-tag studies at Wanapum and Priest Rapids dams conducted in 2007 (Timko et al. 2007, draft).

Tagging will take place inside the tagging trailer next to the sanctuary boxes at Wanapum Dam on the right bank. On tagging days, smolts will be dipped from the sanctuary boxes, and

temporarily transferred to a large plastic container (supplied with a continuous flow of river water) located near the surgical tables.

Individual fish will be transferred to a light anesthetic bath containing tricaine methanesulfonate (MS222; 13.9 mg/L) and held for approximately 3 min. After the light anesthetic, fish will be transferred to a stronger anesthetic bath (MS222; 72 mg/L) and held for approximately 3 min or until the fish losses equilibrium. Anesthetized fish will be measured to the nearest millimeter, and weighed to the nearest tenth of a gram. If a fish is smaller than 147 mm, or larger than 200 mm, or if there are visible signs of injury or disease, the fish will not tagged. Anesthetized fish will be placed on a Plexiglas surgical table in order to stabilize the fish's body during surgery. The surgical table will be soaked with Stress Coat (Aquarium Pharmaceuticals, Inc. Chalfont, PA) to minimize scale loss and maintain the exterior mucous coat. Fish will be placed ventral side up on the surgical pad, and their gills will be continuously flushed with anesthetic solution (MS222, 16.7 mg/L) fed through a tube placed in the mouth of the fish. About 1 min prior to completion of surgery, the flow of anesthetic solution will be replaced with fresh river water to start the recovery process. River water supplied from within the fish ladder entrance will serve as the water source during tagging operations.

To implant the transmitter in the fish, a 1 cm incision will be made 3 mm away from and parallel to the mid-ventral line starting about 3 mm anterior to the pelvic girdle. The incision will be only deep enough to penetrate the peritoneum (Adams et al. 1998). The transmitter will be implanted with the transducer (the ceramic tip of the tags) pointing towards the head of the fish. Tags will be positioned horizontally directly under the incision. An intraperitoneal antibiotic will be pipetted (30 μ L) into the incision to prevent infection. The incision will be closed with three interrupted, absorbable sutures evenly spaced across the incision. A small amount of a cyanoadhesive compound (Vetbond) will be applied to the incision to secure the sutures in place. Surgical equipment will be disinfected with a diluted germicidal solution after each fish (Appendix A, *Guidelines for Surgical Tag Implantation Procedures*).

4.4.3 Post-Tagging Recovery

Immediately after surgery, tagged fish (1 fish per bucket) will be moved to 20 L oxygenated recovery buckets where they will be monitored until they gain equilibrium and begin regular swimming and breathing movements. Recovery buckets will then be placed on shelves in a recovery room, adjacent to the tagging trailer. Each bucket will have an opening in the lid that allowed for continuous water flow, and a PVC overflow spout. Dissolved oxygen (DO) and water temperature will be measured twice a day. The recovery room will be supplied with two redundant pumps in case of pipe blockage in one. Water flow to the recovery buckets will be checked at least twice a day.

Two days after tagging, the tagged fish will be moved into fly tanks (45 fish per tank). The fly-tanks are similar to the sanctuary boxes except that there is no Plexiglas window, there is an external "fin," and the bottom of the tank sloped towards an outflow hole (in order to ensure that fish slide from the tank upon release). Fish will be held in the fly tanks for several hours, up until their release into the river via helicopter.

4.4.4 Release Procedures

Tagged yearling Chinook will be released by helicopter, similar to the procedures used in 2004, 2006 and 2007 (Robichaud et al. 2005, Timko et al. 2006, and Timko et al. 2007, draft). Approximately 3 hr prior to departure from the Wanapum Dam holding site, DO and water temperature will be measured in the fly tanks. Measurements will be taken immediately before and immediately after the tagged fish are transferred into the fly tanks.

For all releases, water flow to the fly tank will be shut off 10 min prior to departure. An oxygen tank, attached to each fly tank will provide oxygenation to the water during transport. Fly tanks will be attached to a helicopter by a long cable, and will be transported to their release site. The helicopter will lower the fly tank to within about 1 m of the water, and will trigger a release mechanism that will open a valve near the bottom of the fly tank, causing the fish to pour out into the river (Figure 9). It will take approximately 60 sec for the fly tank to be emptied into the river. After each release tank is emptied, the release site will be examined for dead or moribund fish and the tank will be inspected for residual fish upon return to Wanapum Dam. Dissolved oxygen and water temperature in the river will be recorded, along with release times.



Figure 9 Helicopter releasing fish into Columbia River.

4.5 Data Collection

The time for all received signals will be synchronized in order to determine time of arrival for each detected pulse. The individual echoes will be entered in digital format into a database using the HTI software package *MarkTags*.

4.6 Data Analysis

There are three phases associated with preparing the databases required for the survival analyses: I) data detection during the field season; II) data marking and tracking during and after the field

season; and III) data quality control during and after the field season. The specific data processing steps included in each phase are listed below:

4.6.1 Phase I: Data Detection during the Field Season

1. Detection of acoustic-tag codes for each fish in all release groups.
2. Verification and data input of all the fish and tag release data.
3. Continuous verification of the operation of all of the ATR systems.
4. Collection and backup of all raw acoustic-tag data.
5. Phase II: Data Marking and Tracking during and after the Field Season
6. All acoustic-tag data files will be tracked using the *MarkTags* tracking software. Lists of all detected fish will be determined daily.
7. Phase III: Data Quality Control during and after the Field Season
8. The data will be constantly verified during and after the field season to ensure the integrity of the acoustic-tag database.
9. The Project Leaders in charge of each of the study tasks will review the data on a daily basis.

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Appendix A
LGL Limited Standard Field Operating Procedures:
Guidelines for Surgical Tag Implementation Procedures



environmental research associates

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LGL LIMITED STANDARD FIELD OPERATING PROCEDURES

Guidelines for Surgical Tag Implantation Procedures

*-Updated June 2007 based on SOP from the USGS – Columbia Research Lab in Cook, WA-
and the JSATS meeting in Portland, OR*

Surgical Equipment and Materials Needed:

- dissolved oxygen (DO) meters which include temperature readings
- Total dissolved gas (TDG) meter
- Autoclave
- Tags (radio or acoustic), receivers or other equipment
- Tricaine methanesulfonate (MS 222; 55.5g/L stock solution)
- Sodium bicarbonate solution (buffer; 100g/L)
- Stress coat – (stock solution 15 ml in 2L of water)
- 20L buckets marked at 10L and clearly labeled “HEAVY” (referring to anesthetic buckets)
- 20L numbered recovery buckets with lids
- Pair of 20L buckets (gravity feed) marked at 10L connected by rubber tubing with in-line shut-off valves – one labeled “MS222” and one labeled “H₂O”
- Syringes for measuring anesthetic (60cc)
- Oxygen delivery system
- Small and large dip nets
- Sanctuary nets
- Latex or non-latex gloves
- Waterproof scale measuring to the nearest 0.1 g
- Large plastic weigh boats

- Surgery table – Plexiglas box with a built in v-shaped trough (trough is covered with plastic cupboard lining to avoid mucus and scale loss). Plastic hoses are also connected to the surgery table for outtake and intake of water. The water from outtake hose is emptied into a 20L bucket.
- Trays for holding solutions used to clean surgical tools
- Needle drivers
- Forceps
- disposable scalpel (#15's - #10's can be used for larger fish)
- Germiphene (bactericidal, fungicidal and virucidal; diluted 16 ml of Germiphene to 1 L of water)
- Liquamycin LA 200 (antibiotic – oxytetracycline injection)
- Pipette (20-100 microliter (μL) volume) and tips
- Sutures (5-0 [4-0 or 3-0 can be used for larger fish] Vicryl coated and tapered RB-1 needle)
- Catheters (shielded needle – 14 G or 16G)
- Triple antibiotic ointment
- Cotton-tipped swabs
- 2 L bottles to make up stock solutions for stress coat, Germiphene and MS222
- Timepiece
- Sharps container and Ziploc Bags
- Data sheets, writing tools, computer

Procedures:

1) Collection and Pre-Tag Holding

- a. Verify that proper collection and transport permits have been obtained and are in possession at the time of collection and tagging. All staff involved in the tagging procedures must be aware of permit restrictions. Copies of the permits are to be visible at the collection facility.
- b. All species (i.e. Chinook and steelhead) should be transported (both pre and post surgery) separately; especially with respect to the collection and transport from the Rocky Reach bypass facility.
- c. Before putting fish in pre-tagging holding tanks, the tanks are scrubbed, flushed, and re-filled. The DO and temperature of the water are checked to ensure adequate water quality. Pre-holding tanks should have no light access (i.e. no windows) as darkness is thought to reduce Frustrated Smolt Syndrome (FSS).
- d. Pre-tagging holding densities is approximately 300 fish per tank (however, the maximum density of each tank is 700 fish). Holding tanks must have flowing, untreated river water supplied at all times during holding. The upper pre-holding density should not exceed 50 g/L and fish should be separated by species. The

maximum holding density in 2007 was 100 smolts in 825 L of water, or 12 g/L (assuming 100 g per smolt).

- e. The pre-tag holding period begins once fish are placed in holding tanks (approximately 24 hours). Pre-holding times for study fish should be between 15 to 24 hours and not to exceed 36 hours. For example, if LGL is delivered fish at noon, and tagging the following day runs from 8 AM until 4 PM, then our pre-tagging holding times will range from 20-28 hours, and any fish tagged after 12 PM would be over the recommended holding time. All fish (including gatewell dipped fish) would have to be delivered by 5 PM in order for our pre-holding times to range from 15-23 hours. In 2007, all fish were delivered to tagging site prior to 3 PM.
- f. Each species collected is held in a separate holding tank to reduce stress. Collection dates and holding densities are recorded on each pre-tag holding container using erasable whiteboards.
- g. Pre-tag holding containers are monitored for DO and temperature twice daily until tagging begins.

2) Fish Size Criteria

- a. Size of fish tagged is dependent on the type of tag being used. The size criteria for juvenile salmonids was between 150 and 210mm. The corresponding weight of this size class of fish is 35.6 to 94.5 g for Chinook and 30.3 to 88.1 g for steelhead. Given the 1.5 g acoustic transmitters used in 2007, the maximum tag weight to body weight ratio for Chinook and steelhead were 4.2% and 5%, respectively.

3) Pre-Tag Preparations

- a. Environmental conditions – All staff must be trained on all water quality measuring equipment.
 - i. Dissolved oxygen (DO): will be measured in mg/L in a pre- and post-tag holding tanks/buckets during each tag session.
 - 1. Measurements will be taken using a DO meter.
 - 2. DO concentrations in pre- and post-tag holding tanks/buckets should be between 7mg/L and 13 mg/L. If readings are outside of this range, inform your field manager and check for limited water flow to the tank which may cause low oxygen levels. Add supplemental oxygen if necessary.
 - ii. Water temperature: will be measured in °C in a pre- and post-tag holding tank during each tag session. Temperature may be taken with a DO meter. Temperature of the pre- and post-tag holding tanks/buckets should be within 2°C above or below ambient water temperature. If readings are outside this range inform your field manager and change the water.
 - iii. Air temperature: within the surgical area should be controlled to ensure that the fish on the surgical table are not too hot or too cold. Typically, the tagging trailer and post-op recovery trailer are equipped with swamp coolers.
 - iv. Total dissolved gas (TDG): will be measured as percent saturation twice a day in the headbox and in a recovery bucket. Measure once at the start of the day and again at the end of tagging for the day.
 - 1. Measurements will be taken using a Common Sensing TDG meter (various models)

2. Gas super saturation (TDG > 110%) may lead to gas bubble disease and must be avoided. Contact your field manager if TDG approaches 110%. Check that water inflows are being off-gassed before use.
- b. Setup of equipment
- i. Tags should be activated, tested, and prepared for implantation (procedure depends on tag type, either radio or acoustic).
 - ii. Disinfect all tags in diluted Germiphene solution. The tags remain in disinfectant for up to an hour and are then transferred to a distilled water rinse bath to soak overnight. The tags are taken out of the rinse bath and actively rinsed under running distilled water. The tag will be placed, using forceps, in a vessel of distilled water just prior to implant. Note that some chemicals in certain disinfectants may adversely affect the coating on the transmitters.
 - iii. Prepare surgical table and equipment for use.
 - iv. Setup measuring board and scale. All staff must be trained on the use of all weight and length measuring equipment.
 1. Ensure the scale is functioning properly. Scales should be calibrated at the start of the season and recalibrated as necessary.
 2. Put approximately 1-2mL of diluted stress coat on the surgical table.
- c. Recovery buckets must be filled with untreated river water and supplied with oxygen if necessary. Dissolved oxygen levels should be consistent with that of the river as the recovery buckets are filled with water just prior to the fish being placed in the bucket. If there is a delay (due to rejecting fish) it is the surgeons call to add oxygen to the recovery bucket.
- d. Administration of anesthetic: The effectiveness of MS-222, as an anesthetic, varies with factors such as temperature, fish of size, species, exposure, and fish density. Adjustment of the anesthesia concentration should be based on the amount of time it takes for a fish to lose equilibrium. However, the anesthesiologist should target between 60-80 mg/L MS 222 concentration level. Communication among the tagging crew is very important to ensure that MS-222 is not administered to the anesthetic buckets and gravity feed buckets more than once or not at all. Never administer MS-222 into a bucket until you confirm that no one else has.
- i. Fill the "HEAVY" anesthetic bucket with 10L of untreated river water. Add approximately 10 ml of MS-222 stock solution to yield a concentration of 56 mg/L.
 - ii. Fill both gravity feed buckets with 10L of untreated river water. Add 3 ml of MS-222 stock solution to the bucket marked "MS222" for a light dose. Do NOT add MS-222 to the freshwater bucket.
 - iii. Splash some dilute Stress-Coat solution (helps maintain the slime coat on the fish, which helps prevent infection and reduce stress) to all buckets (anesthetic, gravity feed, surgery table, and recovery bucket).
 - iv. Use a pH meter to monitor pH levels of the anesthetic buckets (HEAVY and gravity feed maintenance buckets). If the MS222 solution changes the pH level by more than 1 point, or to less than 7, add a sodium bicarbonate solution (buffer) to any bucket that has anesthetic. Add 1 ml of buffer solution for every ml of anesthetic added.

- v. Water in all buckets (anesthetic and gravity feed) should be changed and monitored periodically to minimize dilution of anesthetic water and temperature changes (a temperature change more than 2°C above or below ambient water temperature) and to ensure you do not run out of water in your gravity feed buckets during a surgery. Typically, water in the anesthetic and gravity feed buckets is discarded, and refilled after about 10 fish were tagged. During warm days water may need to be changed more frequently due to rapid increases in water temperature (this did not occur in 2007).
- vi. All anesthetic and gravity-feed freshwater maintenance buckets will be monitored for dissolved oxygen. Oxygen levels should be maintained near saturation. It is the surgeons or anesthesiologists call to add oxygen to the necessary buckets.
- vii. Anesthetic and freshwater buckets should be filled and prepared just prior to tagging.
- viii. Transfer fish (no more than 30) using a sanctuary net and two 20L buckets from pre-tag holding tank to a large container (garbage bucket) which is plumbed with untreated river water. The large container is covered, and nothing happens for 20 minutes (so the fish calm down).
- ix. Transfer individual fish from the large container into “HEAVY” anesthetic buckets. Although the USGS recommends the use of sanctuary nets for this transfer, aquarium nets were used in 2007. Use of sanctuary nets caused the dilution of the HEAVY sedation solution, making fish anesthetization problematic. The large container was close enough to the “HEAVY” anesthetic bucket to minimize “out of water transfer” stress. Moreover, it was much easier to catch fish in the large container using an aquarium net, which meant less fish-chasing, and hence less stress to the fish in the container.

4) Implantation of tags

a. Anesthetizing fish

- i. A fish is dip-netted (using sanctuary net – see recommendation 3.d.ix) from the large container (garbage can) and placed in “HEAVY” anesthetic bucket of MS-222. Secure the lid of the anesthetic bucket as soon as the fish is in the bucket. After one minute the fish will have slowed down enough to quickly measure fork length and check for abnormalities. If the fish is rejected after one minute then it is transferred to a freshwater recovery tank and released into the river.
- ii. If the fish is not rejected after one minute it remains in the “HEAVY” anesthetic bucket. The fish should take an additional 1-3 minutes to loose equilibrium and then it is ready for tagging. If after sedating a few fish, you notice that time required for fish to lose equilibrium is more or less than normal, then adjust the concentration of the anesthetic up or down. If loss of equilibrium takes less than one minute or greater than 5 minutes, reject the fish. Countdown timers should be either attached or beside the “HEAVY” buckets in order to monitor timing of each fish in “HEAVY” anesthetic.

- iii. Once the fish loses equilibrium, the surgeon will visually screen the fish for tags, fin clips, fungus, disease, descaling, bloated belly, or any obvious abnormalities. Relay any necessary information to the data recorder.
 - iv. Rejects – If the fish is unacceptable for tagging, transfer the fish to a freshwater tank and then release it into the river.
 - v. Starting at in May 2006, in response to some acute mortality events, LGL was asked to strengthen cull criteria. Fish were rejected if they had mild descaling, discoloration, or had tattered or missing fins. As of 12 May 2007, by request of Curt Dotson (GCPUD), LGL Limited returned to the typical, more relaxed cull criteria for both steelhead and Chinook. LGL Limited taggers resumed the tagging of fish with frayed or damaged fins, and with scale loss less than 20%. LGL Limited taggers continued to cull fish with injuries, parasites, missing fins and >20% scale loss. LGL Limited taggers also continued to cull fish based on size restrictions (150mm - 210mm).
- b. Recording fish length and weight
- i. Transfer (using sanctuary net) the anesthetized fish to the scale and weigh the fish to the nearest 0.1g.
 - ii. Transfer the fish to the measuring board (on the surgery table) and measure the fork length to the nearest millimeter (mm). Data must be vocally relayed to the data recorder to avoid data errors. The data recorder should then record this information and repeat numbers back to avoid any miscommunication.
 - iii. Any fish dropped on the floor before tagging must be rejected. A fish that flops from the tagging trough into the surgical table during tagging can still be tagged. If a fish is dropped on the floor after it is tagged, remove the tag and reject the fish.
- c. Surgery
- i. Place the fish on the surgery table ventral side up. Anesthetic should be administered through the gravity feed tubing as soon as the fish is on the surgery table. The tubing must be placed just inside the mouth or as close as possible so the water flows across the gills. The flow of water should be just enough to cover the gills. If the flow is too low, the fish will flare its gills and become agitated. Adjust to a flow that keeps the respiration of the fish normal throughout the surgery. Use the in-line valve to control the flow of anesthetic, fresh water, or a mixture of both. Start with a constant flow of anesthetic and monitor the condition of the fish.
 - ii. Using a scalpel, make an incision, approximately 5 mm in length (dependent on tag size), about 3 mm away from and parallel to the mid-ventral line. Start your incision a few millimeters anterior to the pelvic girdle. The incision should be just deep enough to penetrate the peritoneum (the thin membrane separating the gut cavity from the musculature), avoiding the internal organs. The spleen is close to the incision position so pay close attention to the depth of the incision (Figure A1).



Figure A1 Ventral view of a juvenile salmonid showing the location and proper placement of incision.

- iii.
 1. There is no exact specification for what size scalpel blade to use for each fish. We use a veterinary purchased #15 disposable scalpel. Since fish come in all sizes, there will be fish for which a #10 scalpel is more appropriate (i.e. adult bull trout).
 2. One scalpel blade can be used on about 7-10 fish before it becomes dull. If the blade is pulling roughly or making jagged incisions, it needs to be changed prior to tagging the next fish.
 3. If you believe you cut an internal organ, do not implant the tag, stitch the incision, and reject that fish. Excessive bleeding should be noted on the datasheet.
- iv. If the tag does not have an antenna, skip to section 4.c.vi. If the tag has an antenna, make an antenna exit site in the body wall using a shielded needle (needle and catheter; 16 gauge for juvenile salmonids and 14 gauge for adult bull trout). Holding the shielded needle between thumb and forefinger, guide it through the body cavity, keeping the needle tip covered by the catheter to protect the organs. Using your thumb and forefinger, push the end of the needle through the catheter to puncture the body wall. The exit hole should be posterior between the pelvic girdle and viscera, to a point 5-10mm off-center from the mid-ventral line and posterior to the origin of the pelvic fins (Figure A2). Figure A3 is a photograph of the lateral view of an adult bull trout showing proper placement of the antenna exit hole in relation to the incision. Note the incision for the bull trout is closer to the pelvic girdle due to the length limitation of the catheter
- v. The tip of the needle should be against the body wall as it is pushed through the muscle. Make certain the needle punctures the body wall, not the plastic

catheter. The catheter should remain in the body wall while the needle is extracted from the exit site.

1. There is no exact specification for what size catheter to use for each fish. For juvenile salmonids we typically use 16 gauge catheter size.
2. Route the antenna through the catheter sheath and gently pull the sheath out of the body wall and off the end of the antenna.



Figure A2 Lateral view of a juvenile salmonid showing proper placement of the antenna exit hole in relation to the incision.



Figure A3 Lateral view of an adult bull trout showing proper placement of the antenna exit hole in relation to the incision. Note the incision for the bull trout is closer to the pelvic girdle due to the limited length of the catheter.

- vi. Gently push the tag into the body cavity. It should lie directly under the incision and will aid in keeping the organs from protruding through the incision. If the tag has an antenna, you can gently pull on the antenna to help position the tag within the body cavity.
- vii. Use a pipette to administer Liquamycin LA 200 (oxytetracycline) in the incision at a dosage of (30 μ L). Change the pipette tip after each fish.
- viii. Begin suturing the incision. Three interrupted stitches are usually used to close the incision, depending on the size of the tag and incision. For adult fish (i.e. bull trout) you may need up to 5 stitches to close the incision.
 1. To make a stitch, lock the needle (at the end of the suture) in the needle drivers so the needle point faces you. Enter the outside edge of the incision on the side farthest from you and exit through the other edge of the incision, pulling the suture perpendicular through the two edges. The needle should enter and exit the skin as close to the edge of the incision as possible without tearing the skin (~ 2 mm from edge of incision). Pull the needle and suture through the skin to leave a tag end of about 2-3 cm of suture material protruding from the needle entrance location, then release the needle from the needle drivers. With your non-dominant hand, grasp the long end of the suture material (usually with thumb and forefinger) at or below the needle, and make three forward wraps (i.e., away from your body) around the tip of the needle driver, which should be held in your dominant hand.

With the three wraps still around the needle driver, grasp the short tag end of suture material with the needle driver and tighten the stitch by pulling the wraps off the needle driver and pulling both ends of suture material perpendicular to the incision. On the first knot, the dominant hand holding the needle driver should pull toward your body and the non-dominant hand should pull away from your body. Tighten the suture lightly, just so the edges of the incision meet, but do not overlap, pucker, or bulge the edges of the incision. The second knot is the same as the first, but in reverse order. On the second knot, grasp the long end of suture material with your non-dominant hand, make three reverse wraps (i.e., toward you body) around the end of the needle driver, grasp the short end of suture with the needle driver, and tighten the stitch. This time, the knot should be tightened by pulling your dominant hand (holding the needle drivers) away from you and your non-dominant hand toward you. The second knot should be tightened snug to prevent the stitch from coming loose. This completes one stitch. Cut the suture with the needle drivers, leaving ends approximately 5 mm in length.

- a. When making wraps around the needle driver, it is easiest to make the wraps closer to your hand holding the suture, rather than closer to the fish. After making wraps around the needle driver, be sure to grab (with the needle drivers) the short end of the suture close to the end of the suture material. If you grab the suture material closer to the fish than to the end of the suture, then the tag end will fold onto itself, get tightened into the knot, and leave two strands of suture material in the knot. This is a common mistake.
 - b. When pulling a knot tight, be sure the knot lays flat and does not twist onto itself into a “balled-up” knot. If the knot begins to “ball-up” when pulling tight, it can sometimes be coaxed to lay flat by twisting the suture material between your thumb and forefinger. With experience, each surgeon develops their own “tricks” for making sure the knot lays flat. Although not every knot is perfect, you should strive for perfection on every stitch, rather than settling for an imperfect stitch. An imperfect stitch will be more likely to come untied, possibly resulting in slower wound healing or tag loss, which could ultimately affect the survival of the fish.
2. There is no exact specification for what size suture to use. Generally, 5-0 suture is used for Chinook and steelhead smolts. For larger smolts, a 4-0 suture may be used. The 3-0 suture is used for bull trout. Since fish come in all sizes, there will be some overlap in these approximate parameters.
 3. Generally, a good time to switch the in-line valve on the gravity feed buckets to untreated river water is just prior to the last stitch (approximately one minute before the surgery is complete). This

initiates recovery from anesthesia as early as possible. However, if the fish appears to be inadequately gilling, provide a mixture or all fresh water as soon as possible. If the fish is too lively to finish the surgery do not switch to fresh water.

4. If a radio tag has been used, attach the antenna to the side of the fish with a single suture in the caudal peduncle about 5-6 mm posterior to the antenna exit site.
 5. Because sutures are long, each individual suture (one packet) can be used on 2-4 fish.
- ix. To keep the process flowing, the surgeon should indicate when the next fish should be sedated. This assures that time is being utilized to its fullest. Typically, a good time to start sedation on the next fish is when the last stitch is started. Faster surgeons may start sedation of the next fish immediately after taking one out of the sedation bucket.
 - x. Once the incision is closed, dry the incision area and gently roll a cotton-tipped swab covered with triple antibiotic ointment over the incision and sutures.
 - xi. Transfer the fish from the surgery table directly to a labeled recovery bucket using your gloved hands. The data recorder should record the label of the bucket as the location for that fish. There should be no more than 2 fish per recovery bucket. Recovery buckets are hooked up to river-water in post-tagging recovery facility.
 - xii. Between surgeries, the surgeon should prepare their tools for the next surgery. Disinfect the tools in Germiphene solution and ensure that the scalpel blade and suture are acceptable to use on the next fish. If necessary, replace the scalpel blade and catheter. All surgical equipment must be disinfected between each fish to avoid transmitting disease among individuals. Surgical instruments must be soaked in the disinfectant for several minutes (see directions on bottle, typically at least 10 minutes) for the disinfectant to be effective. It may be necessary to have 3-4 sets of surgical equipment instruments which are rotated in order to increase soak time between uses.
 - xiii. Tagged fish are checked at various times throughout the day. We wait approximately ½ hour after the last fish of day is tagged for a final assessment of all tagged fish. If a tagged fish does not exhibit signs of proper swimming behaviour or has not gained equilibrium it is up to the surgeon to decide if the fish should be rejected. If the fish is rejected then the tag is pulled and put into another fish. The rejected fish is re-sutured and released into the river.
- 5) Cleanup at the end of the tagging day
- a. Wipe down all counter tops, scales and measuring boards with the Germiphene solution to disinfect. Germiphene solution should not be made of river water, but rather distilled water.
 - b. Soak scalpels, catheters, forceps, and scissors in Germiphene solution and thoroughly dry to prevent rusting.
 - c. Scrub needle drivers with a small brush or scour sponge.

- d. All autoclavable surgical equipment should be autoclaved.
 - e. Buckets should be rinsed thoroughly with untreated river water and placed upside down to dry. Buckets that contained fish that died need to be scrubbed with disinfectant and rinsed before being placed upside down to dry for 24 hours.
 - f. Store electronics in proper cases.
 - g. Extra fish in the pre-tagging holding tanks are released back into the river. Empty holding tanks are scrubbed, flushed, and either re-filled (for use the next day), or left empty.
- 6) Datasheets
- a. The appropriate tagging and water quality data sheets for your project will be filled out before and during tagging. At the end of the tagging day, the field manager should review these datasheets to ensure proper collection procedures were followed.
- 7) Post-Op Recovery
- a. Optimal post-op recovery time should be 15-24 hours, and should not exceed 36 hours. Post-op holding time should start when the last fish is tagged. Therefore, the first fish of the day could be held about 8 hours longer than the last fish of the day. Currently, GCPUD study fish are held up to 48 hours after surgery.

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