

Alternative Barging Strategies to Improve Survival of Transported Juvenile Salmonids – 2005



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Final Report

September 2006

Prepared for the U.S. Army Corps of Engineers
Walla Walla District, Walla Walla, Washington
Under Contract DACW57-00-D-0009 #11

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Executive Summary

In spring and summer 2005, Battelle-Pacific Northwest Division, NOAA Fisheries, and the U.S. Geological Survey Western Fisheries Research Center conducted a study for the U.S. Army Corps of Engineers, Walla Walla District, to determine whether alternative barging strategies might improve the survival of salmon smolts traveling down the Columbia River toward the Pacific Ocean in the northwestern United States. To increase understanding of whether transporting steelhead smolts an additional 204 km down river would increase their survival to ocean entry, a study was designed and implemented to examine the travel time, distribution, survival, and avian predation rates on Snake River-origin steelhead released at the traditional release location (Skamania Landing, rkm 226.9) and an experimental release location closer to the ocean (Astoria Bridge, rkm 22.5). In addition, a new technique to collect pathogen data using non-lethal methodologies was used to determine what pathogen loads were in the hatchery steelhead we tagged and whether there were fish performance differences that could be related to the pathogen loads in individual fish.

Hatchery steelhead of Snake River origin were captured from fish transport barges between John Day and Bonneville dams and then tagged with either a passive integrated transponder (PIT) and an acoustic transmitter (AT) or just a PIT tag. At the time of tagging, non-lethal gill clip samples were collected from all AT-tagged fish for pathogen (*Renibacterium salmoninarum* and *Nucleospora salmonis*) analyses. A total of 1,002 hatchery steelhead, in four AT (and PIT) -tagged groups were released at the Skamania landing and Astoria Bridge sites between May 7 and 23, 2005. In addition, 2,475 hatchery steelhead with PIT tags (no AT) were released at these sites between May 16 and 23, 2005. The PIT-tag-only groups were released to determine whether the AT increased the susceptibility of the steelhead to avian predation.

Detection arrays, consisting of autonomous and cabled receivers were deployed near the mouth of the Columbia River in two arrays. The primary array was located at river kilometer (rkm) 8.4, near the piscivorous bird colonies on East Sand Island. The secondary array was located at rkm 2.6. Data from both arrays were processed and analyzed to determine the travel times, cross-channel distribution, and survival of AT-tagged fish. In addition, to estimate the number of fish in each release group that were eaten by piscivorous birds, the bird colonies were scanned for PIT tags to detect tags from fish released in this study. Gill filament samples were analyzed for pathogen detection by polymerase chain reaction (PCR) tests. A non-quantitative nested PCR (nPCR) test was used for detection of *N. salmonis*, and both nPCR and real-time quantitative PCR (qPCR) were used for detection of *R. salmoninarum*.

About 40% of the AT-tagged steelhead were detected near the mouth of the Columbia River. Transporting steelhead the additional distance from Skamania Landing to the Astoria Bridge resulted in different cross-channel distributions, different time-of-day passage through the area of the mouth of the Columbia River adjacent to the large piscivorous bird colonies on East Sand Island, and different avian predation rates. These changes would be expected to produce a juvenile survival advantage from point of release to ocean entry for fish released at the Astoria Bridge. However, the survival estimates for the Skamania and Astoria release groups in this study did not show a significant survival advantage for fish released at the Astoria Bridge site. The survival estimates in this study were impacted by highly variable detection probability values between and within release groups (dates).

Pathogen analyses by PCR detected *R. salmoninarum* in 41% of the AT-tagged steelhead and *N. salmonis* in 25% of these fish. Comparisons of proportions of AT-tagged fish that were detected by

the primary or secondary array or both arrays showed no significant differences between fish that were uninfected and those that were infected with one or both pathogens. Similarly, this study showed no detectable influence of the presence or absence of these pathogens on avian predation rates. The *R. salmoninarum* infection levels in all but three fish were very low, however, and only one of the three fish with higher infection levels was detected by the primary receiving array. None of the three fish were detected by the secondary receiving array or on the bird colonies. Because a qPCR test was not available for *N. salmonis*, the infection levels of this pathogen in the fish were unknown. Although steelhead can be infected by both *R. salmoninarum* and *N. salmonis*, they are less susceptible to clinical disease than certain other species such as Chinook salmon.

Most of the evidence collected in 2005 suggests that a greater proportion of the fish released at the Astoria Bridge would be expected to enter the Pacific Ocean in comparison to those released at the Skamania Landing site 204 km upstream. However, to understand the biologically significant implications of this would require survival studies that estimate smolt-to-adult return (SAR). Use of larger experimental groups of PIT-tagged fish could allow for these types of estimates.

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1.0 Introduction

In spring and summer 2005, Battelle-Pacific Northwest Division, the National Oceanic and Atmospheric Administration National Marine Fisheries Service (NOAA Fisheries), and the U.S. Geological Survey Western Fisheries Research Center conducted a study for the U.S. Army Corps of Engineers, Walla Walla District, to determine whether alternative barging strategies might improve the survival of salmon smolts traveling down the Columbia River toward the Pacific Ocean in the northwestern United States. To increase understanding of whether transporting steelhead smolts an additional 204 km down river would increase their survival to ocean entry, a study was designed and implemented to examine the travel time, distribution, survival, and avian predation rates on Snake River-origin steelhead released at the traditional release location (Skamania Landing rkm 226.9) and an experimental release location closer to the ocean (Astoria Bridge). Data on fish movement, timing, and survival were collected for comparison with previous release data for preliminary evaluation of the concept. In addition, a new technique to collect pathogen data using non-lethal methodologies was used to determine what pathogen loads were in the hatchery steelhead we tagged and whether there were fish performance differences that could be related to the pathogen loads in individual fish.

Fish passage around the hydroelectric facilities along the Snake and Columbia rivers has been facilitated by placing screens in the top portion of the turbine intakes to guide downstream migrating smolts away from the turbines and into juvenile bypass systems through which they are returned to the river or collected for barging or trucking downstream. This collection process reduces the number of smolts passing directly through turbines as they migrate downstream. In addition, screening provides the option of collection and transport of juvenile salmonids by either truck or barge to a release site downstream of Bonneville Dam to enhance their downstream survival.

Transported smolts have survived to return as maturing adults at a different rate than smolts that migrate downstream in-river. The transport to in-river ratio of smolts is currently being examined to determine the factors that account for the difference in post-Bonneville Dam passage survival. The average survival to adult of transported fish has been variable and relationships may be different for different fish stocks and/or times of year. The difference between the survival of in-river migrants and transported fish has been termed differential delayed mortality and an increased understanding of this issue was the endeavor of this work. This research was part of an ongoing effort by the Anadromous Fish Evaluation Program (AFEP) to discern changes that can be implemented to the existing fish transportation program to improve post-Bonneville release survival.

Fish condition has been assessed in previous years prior to and after transportation (Schreck et al. 2005; Congleton et al. 2000; Pascho and Elliott 1989; Elliott and Pascho 1991, 1993, 2004; Elliott et al. 1997). Although stress and stressors have been examined in detail in previous years, the correlation with a particular parameter to develop a solution to the issue has been elusive. This project was directed at acquiring information leading to a better understanding of the differential delayed mortality experienced by transported smolts.

The primary goal of the 2005 alternate barge release site pilot study was to determine whether releasing barged hatchery steelhead smolts at the Astoria Bridge rather than at the typical release location, Skamania Landing, which is 200 rkm upstream, improved survival of the fish to ocean entry. The strategy was to minimize the time spent moving into and through the estuary and to document fish condition, which provided insight into the vulnerability of a smolt to predators. Previous research by Schreck et al. 2005 had recommended exploring a strategy to minimize exposure to avian predators. The

general approach was to tag transported smolts with microacoustic tags, collect pathogen samples, and release fish at the current barge release site downstream of Bonneville Dam (Skamania Landing; river kilometer (rkm) = 226.9) and at the Astoria Bridge (Oregon channel; rkm = 22.5).

Objectives for this study were as follows:

Objective 1. Determine hatchery steelhead smolt survival to ocean entry (at the Sand Islands at rkm 8.4) for groups released at Skamania Landing (downstream of Bonneville Dam) and Astoria Bridge.

Objective 2. Compare survival between hatchery steelhead groups released at Skamania Landing and Astoria Bridge.

Objective 3. Determine the prevalence of *Renibacterium salmoninarum* and *Nucleospora salmonis* for each release group and document relationships between survival rates and the prevalence of these two pathogens.

Objective 4. Determine whether hatchery steelhead smolts tagged with acoustic tags are more likely to be subjected to avian predation than hatchery steelhead smolts tagged with passive integrated transponder (PIT) tags (based on recoveries on East Sand Island).

2.0 Methods

2.1 Test Fish Acquisition, Tagging, and Release

2.1.1 Fish Acquisition

To ensure that all test fish were of similar life histories we used only hatchery steelhead that were diverted into the juvenile bypass system at Lower Granite Dam and then loaded onto a transportation barge. To obtain these fish, we boarded transportation barges at John Day Dam and collected steelhead from barge holds that had only received fish at Lower Granite Dam. Since the barge holds contain a combination of Chinook salmon and steelhead, we developed a sampling device, termed a lift-net grader, (Figure 1) to minimize handling of non-target ESA-listed fishes while acquiring sufficient numbers of steelhead for our tagging purposes. The lift-net grader consisted of a square bottom (86 cm on a side) made of size separation bars used at Columbia River Dams (17 mm, McComas et al. 2003) sewn to netting 122 cm long. At the top of the netting, there was an open stainless steel square the same dimension as the bottom to keep the net open. To capture the steelhead, the grader was lowered into the hold, and the entire contraption was allowed to rest on the bottom of the hold. Then, after a short duration (several minutes), the grader was quickly raised through the water column with guide ropes until the separation bars were in a few centimeters of water. The bottom of the grader was then tilted from horizontal toward vertical, which encouraged the smaller Chinook salmon to exit through the separation bars. This device worked very well for the intended purpose of capturing the hatchery steelhead without having to handle the smaller Chinook salmon and wild steelhead. Only one of the 1,002 live steelhead implanted with acoustic tags was less than 180 mm FL (it was 177 mm FL).

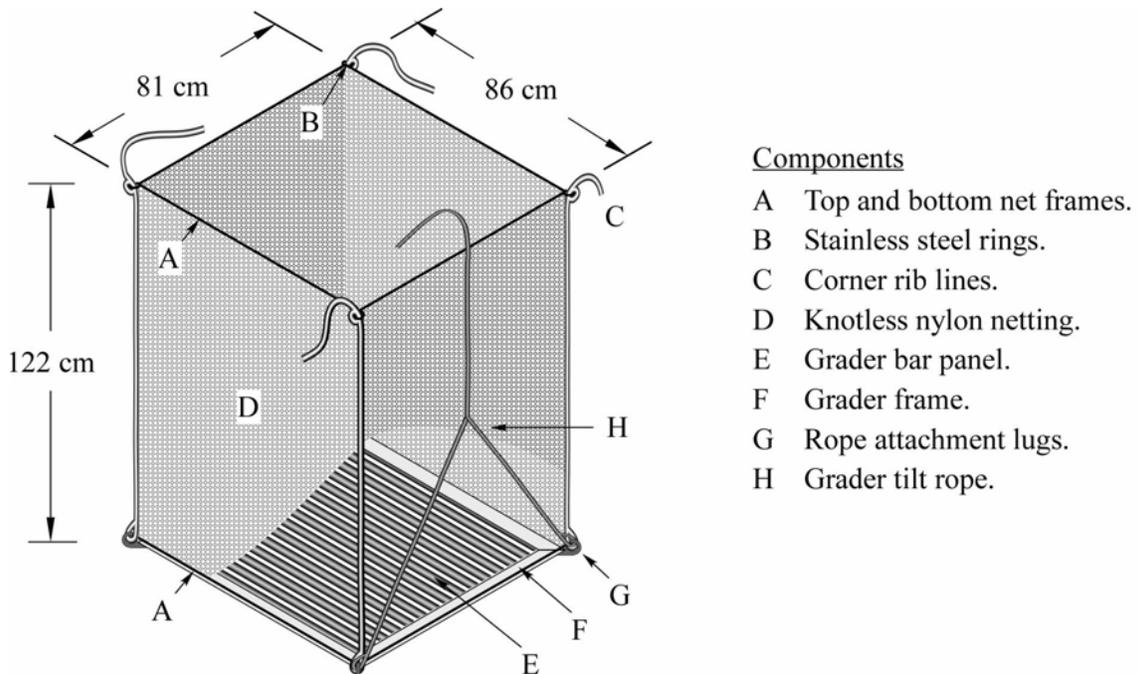


Figure 1. Lift-Net Grader Device used to Acquire Juvenile Steelhead from Barge Holds

Fish were transferred from the lift-net grader to an anesthetic container using sanctuary type dip nets containing about 7 L of water. Fish were anesthetized using tricaine methanesulfonate at a concentration of about 50 mg/L. We tabulated the catch by species and presence or absence of a clipped adipose fin, selecting non-injured, adipose-fin clipped steelhead for use in the study. Other species were allowed to recover from the effects of anesthetic and then returned to barge holds that we were not sampling. Selected fish were then placed in the barge pen (described below). We captured a total of 3,933 steelhead for our tests, out of a group of 5,692 fish; thus, 69% of the fish processed aboard the barge were used in the study.

We designed and constructed a fish containment pen adequate for the entire complement of fish for each sampling day. The empty barge pen was loaded onto the barge at John Day Dam with the use of a crane and placed in the center starboard barge hold. The pen was designed such that the side plates could be removed, leaving perforated plates that allowed water to flow through. Upon arrival at Bonneville Dam, the side plates were slid back into place and the entire pen was lifted from the barge and placed on the deck of the dam using a 28-ton crane (Figure 2). Fish were supplied with flow-through water by a submersible pump. Fish were placed on a pumped river-water supply and monitored until tagging on the following day. Oxygen levels and water temperature were examined hourly.



Figure 2. Barge Pen Being Lifted from Fish Transport Barge to Tagging Area at Bonneville Dam

2.1.2 Tag Implantation

Surgeries on juvenile steelhead to implant acoustic tags took place in a 2.7-m x 6.1-m covered trailer specifically modified for conducting surgical procedures on juvenile fish. This trailer was equipped with aluminum counter tops allowing surgeons to comfortably stand while performing surgeries along with plenty of electrical outlets to supply power to the necessary electronics. The trailer was located on the deck of Bonneville Dam near the entrance of the navigation lock.

Prior to surgery, fish were placed in an anesthetic bucket containing a solution of 80 to 100 mg/L tricaine methanesulfonate (MS-222). After a fish lost equilibrium, a gill sample was taken for pathogen detection (see below), and the fish was weighed, measured, and placed on the surgery table. During surgeries, a maintenance dose of approximately 40 mg/L solution (MS-222) was administered via a tube inserted into the fish’s mouth. Fish were tagged using procedures similar to Anglea et al. (2004). With the fish facing ventral side up, an incision approximately 8 mm in length was made 2 to 5 mm from and parallel to the mid-ventral line anterior to the pelvic girdle. A PIT tag (Biomark model TX1411ST 12.5 mm x 2 mm) was inserted into the peritoneal cavity followed by an acoustic transmitter (Sonic Concepts model E101 19 mm x 5.5 mm; 0.63 g in air; 0.39 g in water; pulse rate interval was one transmission every five seconds; battery life was 30 days). Both tags were positioned parallel to the length of the fish. The incision was closed using two simple, interrupted sutures (Ethicon 5-0 absorbable coated vicryl sutures). Post surgery, fish were placed in a recovery bucket with fresh oxygenated river water and monitored to ensure that they recovered equilibrium before they were transferred to the holding/release tanks.

Of the 3,477 total live fish releases, 2,475 were PIT tagged only, and 1,002 received both a PIT tag and an acoustic tag. Steelhead that were only receiving a PIT tag were tagged according to protocols and standards outlined in the *PIT Tag Marking Procedures Manual* (CBFWA 1999), using mass marking and simple PIT-tag injectors. The PIT-tagging equipment and holding equipment were set up on folding tables under a temporary awning on the deck of Bonneville Dam near the entrance of the navigation lock. Collected fish that were not used in the study were held and released with the Skamania Landing groups.

Table 1 shows the average size, by release date, for those hatchery steelhead that were tagged with both a microacoustic tag and a PIT tag and for those fish that were tagged with a PIT tag only.

Table 1. Average Fork Length (mm (SD)) Information for Hatchery Steelhead Smolts Tagged with only a PIT Tag and with Microacoustic Tags plus a PIT Tag for the Skamania Landing and Astoria Bridge Release Sites Combined in 2005.

Release Date	PIT tag only	PIT + Acoustic tag
5/7/2005		241.0 (14.5)
5/15/2005	244.8 (18.1)	243.3 (16.2)
5/19/2005	239.8 (18.6)	241.6 (18.4)
5/21/2005	240.6 (20.0)	241.1 (18.6)

2.1.3 Fish Releases

After tagging, steelhead were placed in Achord tanks on the deck of the dam near the entrance to the navigation lock. Achord tanks are 1.8 m long x 0.6 m wide x 0.9 m deep (on deep end – tapered up to 0.8 m deep on the end away from the release valve). Loading density and water volume replacement in all Achord tanks were set not to exceed the specifications of the two newer versions of the transport program’s barges (Series 4000 and 8000). The barge specifications shown in Table 2 indicate that a total of 495 fish could be transported in one Achord tank.

Table 2. Loading Density Data for U.S. Army Corps of Engineers Barges and for the Achord Tanks to be used in the Alternate Barge Release Location Study in 2005

Barge Style	Pounds	Gallons	Inflow	lbs/gal	Replacement Rate
2000	23000	85000	4600	0.27	18.48
4000	50000	100000	10000	0.50	10.00
8000	75000	150000	15000	0.50	10.00
Achord Tank	120	246	25	0.49	9.84
number of fish/tank	495				

Acoustic- and PIT-tagged steelhead destined for transport to Astoria, Oregon, were held in a single Achord tank on the deck of the dam until they were scheduled to be transported, generally about 24 hours after tagging. At the scheduled departure time, the tagged steelhead were transferred via a flexible hose approximately 10 cm in diameter to a second Achord tank onboard the NOAA Fisheries research vessel (RV) *Siliqua*, a 12.5-m motor vessel.

The travel speed and course of the RV *Siliqua* emulated the speed and course of a full-size transport barge to the Astoria Bridge release location. We estimated that at a speed of 15 km per hour, it would take a barge an additional 14 to 15 hours per trip downstream to reach Astoria. To allow time for negotiating the navigation locks at Bonneville Dam and inclement weather, the RV *Siliqua* departed Bonneville Dam 16 to 17 hours prior to the desired release time. The release times were set to occur after dark on outgoing tides on four separate dates throughout May 2005. Upon arrival at Astoria, the RV *Siliqua* was positioned in the middle of the shipping channel directly below the Astoria Bridge and released the fish from the Achord tank via a flexible hose.

As part of the Astoria releases, 31 dead fish were implanted with microacoustic tags and released with the live fish to test the assumption that dead fish would not drift from the Astoria Bridge release site to the primary array (a distance of 13.4 km in the navigation channel) (see Table 3). We assumed that dead fish from the Skamania Landing release site would not drift the approximately 220 km to the primary array, so no dead fish were released there.

Steelhead to be released at the normal barge release site near Skamania Landing were held in two Achord tanks on the deck of the dam with the number of acoustic- and PIT-tagged fish divided evenly between the two tanks. The day following tagging, the tanks were positioned using a forklift so that a flexible pipe, approximately 10 cm in diameter, could be used to transfer the tagged fish to one of the holds on the next available transport barge passing through the locks. These fish were then released at Skamania Landing with the other fish in the transport barge. Table 3 shows the release schedule for steelhead released at the Astoria Bridge and at Skamania Landing.

Table 3. Steelhead Released in the Columbia River in 2005 that were Implanted with Microacoustic Transmitters and PIT Tags

Release Date	Release Location	Live Fish Released		Dead Fish Released
		Acoustic Tag	PIT tag only	Acoustic Tag
5/7/2005	Astoria	90	0	10
5/7/2005	Skamania Landing	160	0	0
5/16/2005	Astoria	92	361	5
5/17/2005	Skamania Landing	160	641	0
5/21/2005	Astoria	90	163	10
5/21/2005	Skamania Landing	160	640	0
5/23/2005	Astoria	90	0	6
5/23/2005	Skamania Landing	160	670	0
Total		1002	2475	31

2.2 Survival and Behavior

2.2.1 Receiving Arrays

A total of 65 receiving nodes, arranged in primary and secondary arrays, were deployed to detect and record the presence of passing fish bearing the microacoustic transmitters. The primary array was deployed at rkm 8.4 and consisted of large stationary cabled receivers deployed by NOAA Fisheries and an ocean engineering firm. The secondary array, located at about rkm 2.6, was composed of autonomous receiving nodes (Figure 3). Autonomous nodes included on-board power (30-day battery life) and data storage (256 MB Compact Flash). The autonomous nodes were attached to 68-kg anchors with bungee moorings. The moorings were 3.7 m long and attached the acoustic release (InterOcean Systems, Inc., San Diego, CA; model 111) to the anchor. The acoustic releases had a tag line canister filled with 45 m of 4.7-mm-diameter Samson line, which allowed the nodes to surface when the acoustic release was activated. The lead from the acoustic release to the autonomous node was 0.9 m long and was made of 9.5-mm-diameter Samson Tenex line with a clear Samthane coating. The node was attached to the line by a 0.61-m-long bridle made of vinyl-coated 2.4-mm stainless steel cable that was terminated in stainless steel thimbles on the node end and in a milled UHMW swivel block on the rope lead end. All rope leads were terminated with a braided splice around a 9.5-mm SeaDog nylon thimble and were professionally braided (by a vendor for West Marine). From the node bridle to the surface ran a 9.5-mm-diameter Samson Tenex line with a subsurface buoy (yellow, Spongex CB6, 4.8 kg of buoyancy, 15.2 cm in diameter, and 35.6 cm long) placed approximately 5.5 m above the node. Three additional yellow CB6 buoys were placed on the line at the surface. The length of the rigging was designed to be approximately two times the depth at each deployment location at high tide. Figure 3 shows several autonomous nodes in the process of rigging with the associated radio buoys prior to deployment.

Some nodes on the secondary array (N=14) were initially deployed with a radio communication system buoy attached. The radio system was deployed as a feasibility effort to transmit data to researchers in near real time and to send global positioning system (GPS)-derived time signals out to the nodes to synchronize the clocks within the autonomous nodes (Figure 4). These nodes used a Freewave radio inside a custom buoy that was linked to the autonomous node by an RS-485 cable. The RS-485 cable was attached to the mooring rope by cable ties and electrical tape. The nodes transmitted to a base station that was located in the Lewis and Clark Interpretive Center (at the top of Cape Disappointment).

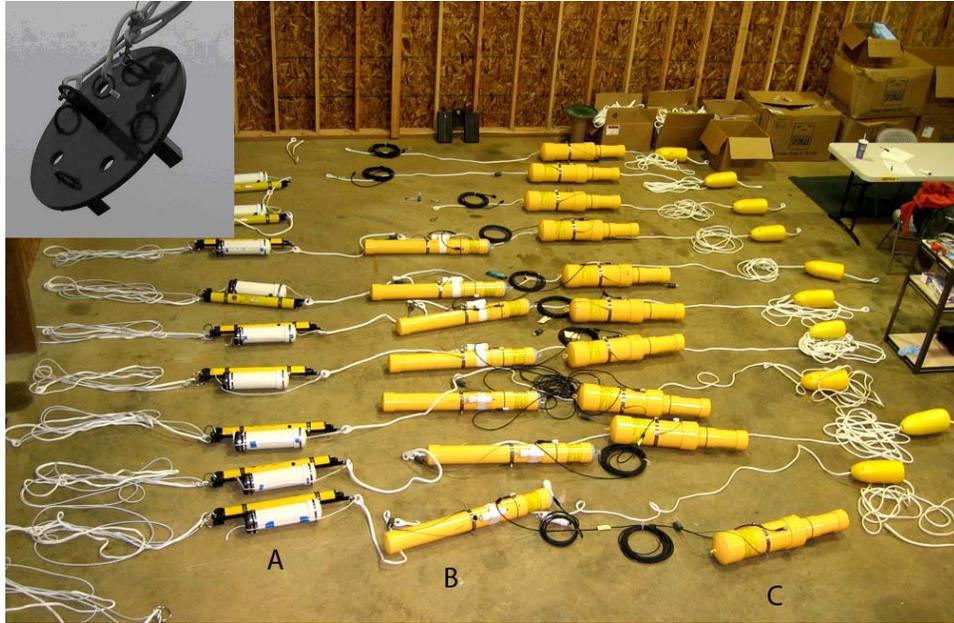


Figure 3. Autonomous Nodes in the Process of Rigging for Deployment on the Columbia River Bar. The 68-kg anchor (inset) was attached to the leads at the left of the photo below the acoustic release (A), the autonomous node (B), and the radio buoy (C).

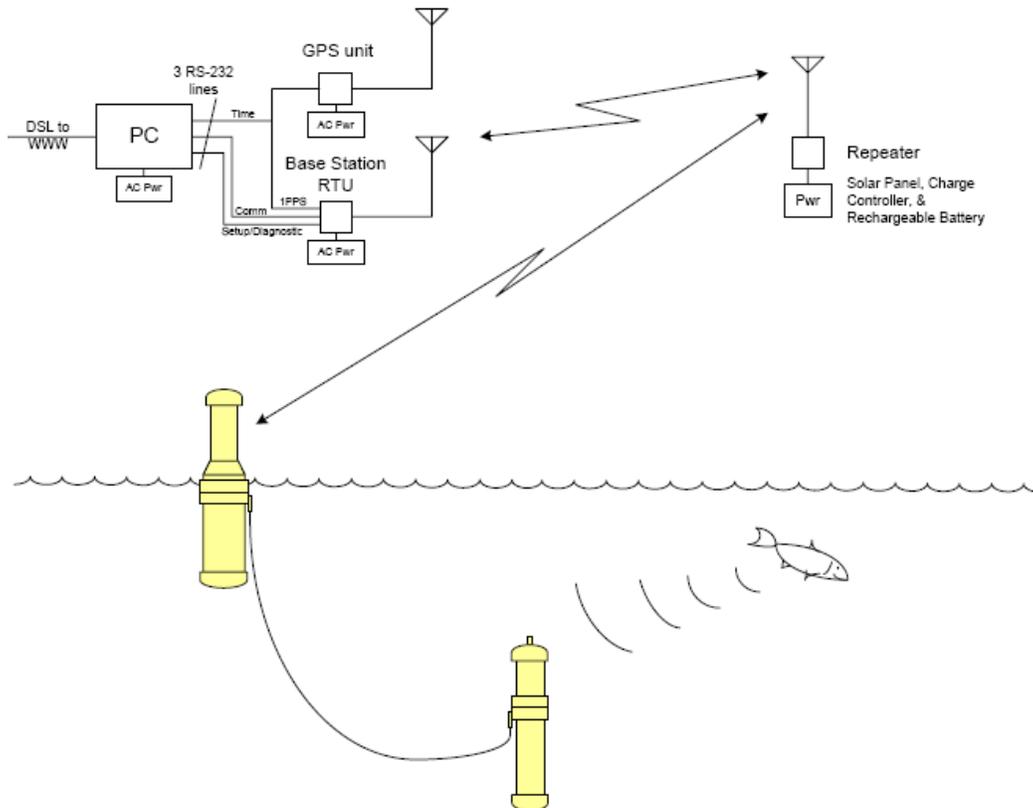


Figure 4. Block Diagram of the Radio Communication System Tested on the Columbia River Bar in Spring 2005

Table 4. Dates of Autonomous Node Deployment, Servicing, and Removal

	Initial Deployment	Servicing	Removal
Secondary array	April 4 - 9	May 3 - 6 June 7 - 9 July 6 - 10	August 16 - 18
Temp. primary array	April 25 - 26 & May 2	May 23 - 25 June 21 - 23	July 6 - 10

2.2.2 Pathogen Sampling

Fish were analyzed for the presence of two salmonid pathogens known to occur in the Snake and Columbia River basins: *Renibacterium salmoninarum*, the causative agent of bacterial kidney disease (BKD), and *Nucleospora salmonis*, an intranuclear microsporidian parasite that primarily infects lymphoblast cells and can cause a chronic, severe lymphoblastosis and a leukemic-like condition. Fish clinically infected with either pathogen may exhibit exophthalmos and ascites and fish infected with *Nucleospora salmonis* may show extremely pale gills associated with anemia, but most infections are sub-clinical. Gill filament samples for determining the presence and levels of *R. salmoninarum* and the presence of *N. salmonis* in tagged fish were collected from fish in every release group at the time of microacoustic tag and PIT tag implantation (see above). These samples were taken from all individual fish in which both tag types were implanted; a total of 1,002 fish were sampled for pathogen analysis. Sample collection methodology followed the protocol for non-lethal gill filament sampling described by Schrock et al. (1994). Briefly, a 2-mm x 3-mm gill sample (approximately 10 mg) was removed from each fish by use of surgical scissors. Samples were placed in individual labeled tubes containing 250 μ L of 95% ethanol at room temperature (below 21°C) and transported to the USGS Western Fisheries Research Center for analysis. The microacoustic tag and PIT-tag IDs associated with each gill filament sample number were recorded. For consistency in treatment, a gill filament sample was also taken from each fish that was PIT-tagged only, but these samples were discarded.

A sub-sample of 60 steelhead from each barge group used for tagging was sacrificed for analysis of both kidney and gill tissue from each fish for both *R. salmoninarum* and *N. salmonis* analysis. This sample was necessary because the relative sensitivity of *R. salmoninarum* and *N. salmonis* detection by polymerase chain reaction (PCR) from gill and kidney tissue of steelhead is unknown; kidney is the tissue normally tested. This sample included 240 fish total from the four release groups. A 25-mg kidney tissue sample and a 10-mg gill filament sample were removed and processed as described above.

Gill and kidney samples were processed and tested for *R. salmoninarum* by a nested PCR (nPCR) according to the procedures of Chase and Pascho (1998) and a real-time quantitative PCR (qPCR) according to the procedures of Chase et al. (in press). Although the sensitivity of the nPCR may be somewhat higher than that of the qPCR (Elliott and Pascho 2004), only the qPCR can provide a measure of the infection levels in fish. Thus, testing a single sample by both PCR techniques was desirable to provide the most information. For detection of *N. salmonis* in gill and kidney samples, the nested PCR method of Barlough et al. (1995) was used, with the following modifications: the use of gelatin and tetramethylammonium chloride (TMAC) was omitted, and 5 μ l of extracted DNA was used in the first round of PCR amplification and 2 μ l of the first round product was used in the nested round of amplification.

2.2.3 Bird Colony Sampling

To evaluate whether steelhead tagged with an acoustic tag suffered different rates of avian predation than steelhead tagged only with a PIT tag, we tagged a separate group of steelhead with only PIT tags and released them concurrently with steelhead tagged with the microacoustic transmitters. The PIT-tagged steelhead were treated exactly the same as the acoustic-tagged fish with the exception of the tag implantation method (i.e., surgical implantation of acoustic tag versus injection of PIT tag).

Having PIT tags in both the acoustic group and the PIT-tag-only group allowed us to use the avian predation data gathered from the NOAA Fisheries avian predation project to facilitate our estimates of predation rates of the fish released in this study. The NOAA Fisheries avian predation project evaluates the impacts of predation by Caspian terns and double-crested cormorants on juvenile salmonids by using electronic detection methods to detect PIT tags in the guano at piscivorous water bird colony locations in the Columbia River Basin (Ryan et al. 2001, 2003). Comparing the rates of predation of PIT-tagged versus acoustic-tagged fish allowed us to determine whether the acoustic-tagged fish were more susceptible to predation by piscivorous birds.

To evaluate if there was a significant difference between the proportion of PIT-tagged and acoustic-tagged steelhead detected on the East Sand Island tern colony, based on release location, release date, or tagging type, we used a two-factor ANOVA (release location and tagging type) with release date included as a blocking factor. The first date, May 7, was omitted from the analysis due to a lack of detected fish available to produce a PIT-tagged-only group. We first added an interaction term between the factors, which was omitted in a secondary ANOVA if the p-value was greater than 0.10. We visually examined a normal probability plot to assess whether these percentage data could be assumed to be approximately normally distributed.

While we are aware of the large cormorant population in the estuary, and the impact it has on predation rates, we did not use these data for statistical analysis due to the low PIT-tag recovery numbers on the cormorant colonies. Because of low detection efficiency on the cormorant colony, we do not feel that, with small release numbers, the fish would be properly represented.

2.3 Data Processing and Analyses

Data collected by the autonomous nodes were recorded as text files on Compact Flash cards. These text files, containing the full tag ID message (consisting of 31 bits comprised of a preamble, ID, and cyclic redundancy check (CRC)), date/time of each signal detection, and a signal strength indication) were transferred to a laptop computer when the nodes were serviced during the season or when recovered at the end of the season. Physical data were also written to file every 15 seconds. Physical data recorded included date, time, pressure, water temperature, tilt, and battery voltage. Detections of acoustic transmitters were recorded in real time as they were received. They were written to media with Tag ID (individual code of transmitter), date/time stamp, receive signal strength indicator (RSSI), and RxThreshold (a calculated measure of noise). Data files from all nodes were coded with the node location and stored in a database that was developed specifically for storing and processing acoustic telemetry data (TagViz©). To filter out “false positives” (detections of Tag IDs that did not meet criteria to be considered a valid detection), a post-processing program was implemented. This program comprised a sequence of steps that involved comparing each detection to a list of tags that were released (only tag IDs that we released were retained in the database), then comparing the detection date to the release date (only tag IDs detected after they were released were kept), then analyzing the RSSI/RxThreshold (essentially signal-to-noise ratio) and only tags that had an RSSI that was 0.75 times higher than the RxThreshold

were kept. Finally, the time spacing between detections was analyzed and only the detections with the correct time spacing were kept in the valid detection file.

Once the valid detection file was created, the detection histories of each release group were analyzed to determine the travel times, travel rates, arrival times, cross-channel distribution, and residence times for each group of steelhead released at the Astoria Bridge and at Skamania Landing at the primary array, as well as the area 5.8 km downstream where the secondary array was located. Travel time between the primary and secondary array was calculated for fish from each group that were observed on both arrays. Fish that were first observed on the secondary array and later observed on the primary array (purportedly due to predation) were excluded from these analyses. The rates of travel from the point of release, either Skamania Landing or the Astoria Bridge, to the primary and secondary array were also calculated for detected fish in each group. To evaluate arrival times, the database was queried for the first observation of each fish observed at the primary and secondary arrays. A count of fish arriving at each array for each hour (independent of day) was then plotted. Day was considered to begin half an hour before sunrise and end half an hour after sunset. An hourly count of fish, based on first observations only, was also plotted against tide elevation. To determine cross-channel distribution, the database was queried to get a count of distinct tags that were observed at each node location for each release group. From this, the percentage of fish observed at each location was calculated. Residence time was calculated for each fish on both the primary and secondary arrays by finding the difference between the times of the first and last observations on an array.

2.3.1 Survival Calculations

Survival estimates were derived from conventional statistical models for mark-recapture data from a single group of marked animals (Cormack 1964; Jolly 1965; and Seber 1965). This model is known by various names, including CJS Model and Single-Release (SR) Model. The model is simple when there are only two detection opportunities for each marked animal. For purposes of survival estimation, detection data are summarized as the “detection history” for each marked fish. With only two opportunities, the possible histories are:

“00” – tagged fish never detected

“10” – tagged fish detected on primary detector array but not on secondary

“01” – tagged fish detected on secondary detector array but not on primary

“11” – tagged fish detected on both arrays.

To estimate survival for a group of tagged fish released at a certain time (a “release group”), the counts of fish in the group with each of the detection histories are used, denoted n_{00} , n_{01} , n_{10} , and n_{11} , along with the total number of fish released, denoted R .

The proportion of fish released that were detected on the primary array $[(n_{10} + n_{11})/R]$ is an estimate of the combined, or joint probability that a fish survived from release to the primary array (S) and that the fish was detected given that it survived (P). Assuming that survival to the array and detection on the array are independent events, the joint probability of both events occurring is the simple product of the two probabilities. Thus, the proportion detected on the primary array is an estimate of survival probability (SP).

To separate the two probabilities in the product requires a method to estimate either of the probabilities individually. The estimate of the second probability can then be obtained by dividing the joint estimate by the estimate of the first. The probability of detection on the primary array can be estimated independently by making the assumption that fish that survived to the secondary array and were detected

there ($n_{01} + n_{11}$) represent a random sample of all fish from the group that were alive as they passed the primary array. The estimated detection probability on the primary array is then the proportion of the sample that were detected on the primary array [$n_{11}/(n_{01} + n_{11})$].

Survival between the primary and secondary arrays cannot be estimated separately from the detection probability on the secondary array because, without a third detection opportunity, there is no way to construct the sample from which to estimate detection separately. Thus, we can estimate only the joint probability of surviving between the two arrays and detection on the secondary array.

The calculations can be summarized with the following equations:

$$\hat{S}\hat{P} = \frac{(n_{10} + n_{11})}{R}$$

$$\hat{P} = \frac{n_{11}}{(n_{11} + n_{01})}$$

$$\hat{S} = \frac{\hat{S}\hat{P}}{\hat{P}}$$

Formulas for the standard errors of these estimates are more complex and are not reproduced here. They can be found in Cormack (1964).

2.3.2 Pathogen Analyses

Contingency tables were used to compare the relative proportions of uninfected fish and fish infected with *R. salmoninarum*, *N. salmonis*, or either or both pathogens in groups of steelhead released at both sites (Astoria Bridge and Skamania Landing), in groups of fish released on different days, in groups of fish that were detected or not detected by one or more receiving arrays, and in groups of fish with PIT tags detected or not detected on the bird colonies. Fisher's exact test (Motulsky 1995) was used for analysis of 2×2 tables, and the chi-square test was used to analyze larger contingency tables.

For each sample date, lengths and weights of fish that were infected with either *R. salmoninarum*, *N. salmonis*, or both pathogens were compared with lengths and weights of fish that were not infected with either pathogen. Because Kolmogorov-Smirnov testing indicated that some of the length and weight data were not sampled from populations that followed Gaussian distributions, the nonparametric Kruskal-Wallis test was used for these analyses.

3.0 Results

3.1 Acoustic Telemetry

A total of 390 of the 1,002 fish (39%) implanted with microacoustic transmitters for this study were detected on the arrays near the mouth of the Columbia River in 2005 (Appendix). The following sections provide information on the survival and general behavior of these juvenile steelhead as they passed through the last few kilometers of the Columbia River estuary and entered the Pacific Ocean. Specific sections present detailed information on the travel time, time of arrival, cross-channel distribution, residence time, survival rates, avian predation, and pathogen levels of fish implanted for this study.

3.1.1 Travel Time and Rates of Travel

The travel time of steelhead released at the Skamania Landing site to the primary array near the mouth of the Columbia River was about 3 days, while steelhead released at the Astoria Bridge were detected at the primary array very soon after release (mean = 0.16 day (3.8 hours), Table 5). Steelhead that migrated downstream from Skamania Landing encountered multiple tidal exchanges during the course of their travels, while most of the steelhead released at the Astoria Bridge passed the primary array on a single ebb tide.

Table 5. Mean Travel Time Information for Acoustic-Tagged Steelhead Released at Skamania Landing (rkm = 227) and Astoria Bridge (rkm = 22.5) and Detected on the Primary Array (rkm = 8.4) near the Mouth of the Columbia River in 2005. Number released and sample size detected (N) used to calculate travel time are shown.

Number Released	Release Location	Release Date	Average Travel Time (days) - release to primary array	N
90	Astoria	5/7/2005 1:30	0.16	15
92	Astoria	5/16/2005 22:45	0.18	91
90	Astoria	5/21/2005 1:00	0.12	22
90	Astoria	5/23/2005 2:40	0.08	15
		Astoria Season Mean	0.16	143
160	Skamania Landing	5/7/2005 1:45	3.79	49
160	Skamania Landing	5/17/2005 4:00	2.66	60
160	Skamania Landing	5/21/2005 2:45	2.62	53
160	Skamania Landing	5/23/2005 0:10	2.65	52
		Skamania Season Mean	2.91	214

The rate of travel from point of release to the primary array for steelhead released at Astoria Bridge was significantly faster than for steelhead released at Skamania Landing (Figure 6). The fish released at the Astoria Bridge were typically traveling at a rate of about 90 km per day (about 2.3 mi per hour) with the maximum rate for the group released on May 23 (175 km/day; 4.5 mi per hour) between the release site and the primary array 14 km downstream while fish released at Skamania Landing were traveling at a rate of about 75 km/day (2.1 mi per hour). The maximum travel rate for groups released at Skamania Landing was for the group released on May 21 (83 km/day; 2.2 mi per hour).

There was not a significant difference in the rate of travel between the primary and secondary arrays for the groups released at the different locations; however, sample sizes were lower on the secondary array (Figure 7). Rate of travel from release to the secondary array were similar for steelhead released at both release locations. Although sample sizes were generally low, three of the four groups had faster mean rates of travel from release to the secondary array when released at the Astoria Bridge compared to fish that migrated in the river from the release site at Skamania Landing (Figure 8). One dead steelhead released at the Astoria Bridge (3.2% of the 31 dead fish released) was detected on the Oregon shore portion of the primary array. This dead fish was detected 0.18 days (4.2 h) after it was released.

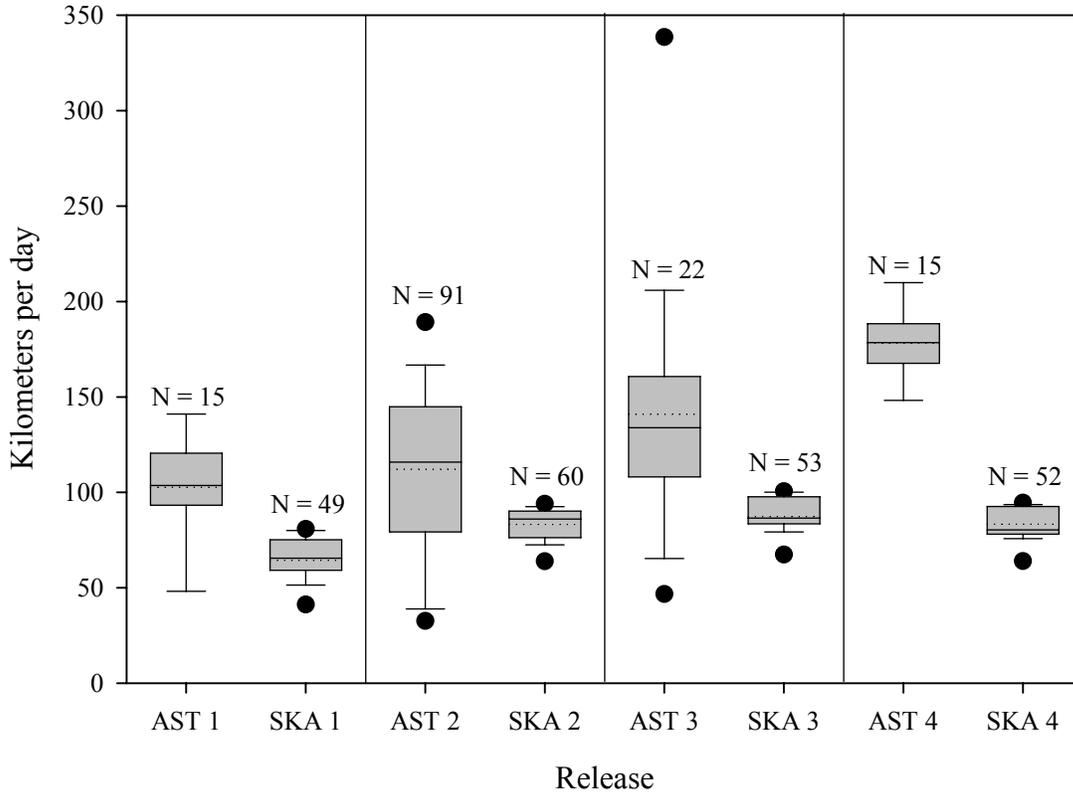


Figure 6. Migration Speed (km/day) of Juvenile Steelhead Released at the Astoria (AST) and Skamania Landing (SKA) Release Sites on Four Release Days and Detected on the Estuary Primary Array. Release dates are separated by solid vertical lines. Dotted horizontal lines within box plots represent means, solid horizontal lines represent medians, upper and lower limits of the boxes represent the 25th and 75th percentiles, whiskers represent the 10th and 90th percentiles, and dots indicate the 5th and 95th percentiles.

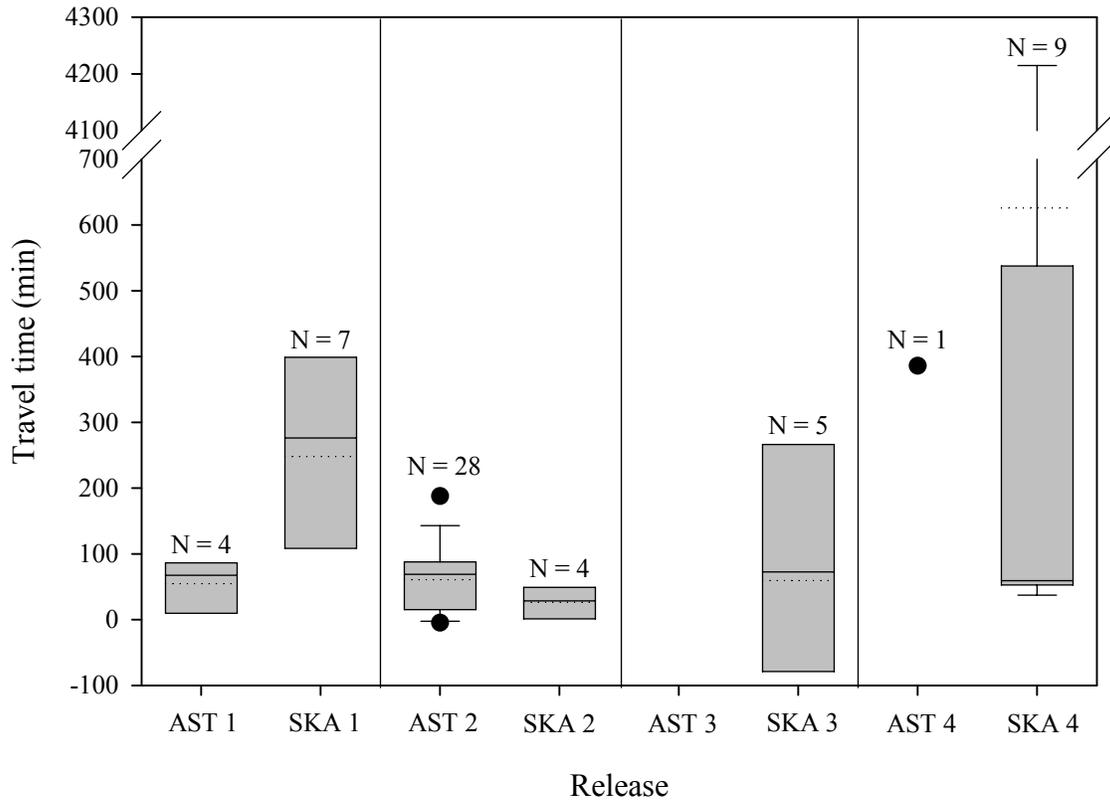


Figure 7. Travel time (minutes) between the Primary and Secondary Arrays for Juvenile Steelhead Released at the Astoria (AST) and Skamania Landing (SKA) Release Sites on Four Release Days. Release dates are separated by solid vertical lines. Dotted horizontal lines within box plots represent means, solid horizontal lines represent medians, upper and lower limits of the boxes represent the 25th and 75th percentiles, whiskers represent the 10th and 90th percentiles, and dots indicate the 5th and 95th percentiles. The dot for AST 4 represents the actual value.

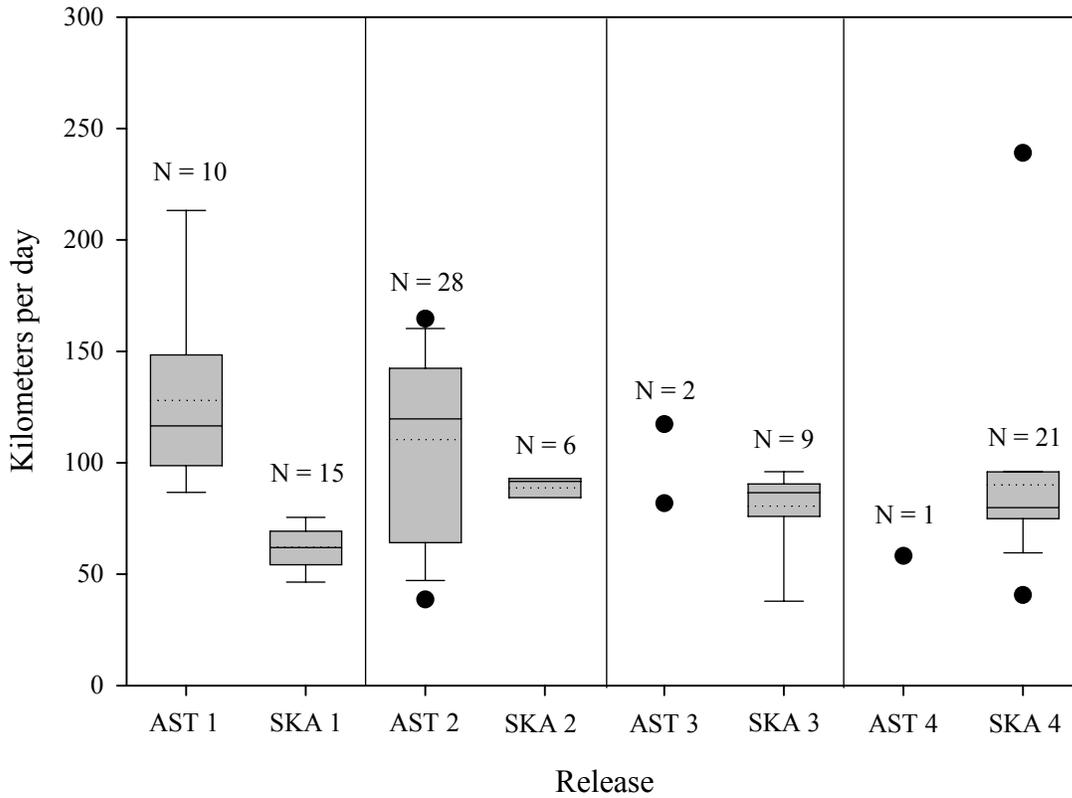


Figure 8. Migration Speed (km/day) of Juvenile Steelhead Released at the Astoria (AST) and Skamania Landing (SKA) Release Sites on Four Release Days and Detected on the Estuary Secondary Array. Release dates are separated by solid vertical lines. Dotted horizontal lines within box plots represent means, solid horizontal lines represent medians, upper and lower limits of the boxes represent the 25th and 75th percentiles, whiskers represent the 10th and 90th percentiles, and dots indicate the 5th and 95th percentiles. Dots for release times with fewer than three detections (i.e., AST 3 and AST 4) represent actual values and not percentiles.

3.1.2 Time of Arrival

Most steelhead released at the Astoria Bridge (65%) arrived at the primary array during hours of darkness. There was no clear pattern in the time of arrival for steelhead released at Skamania Landing; only 14% arrived at the primary array during hours of darkness (Figure 9). Similar patterns of arrival times occurred at the secondary array (Figure 10). Fish released at the Astoria Bridge generally arrived at the primary array shortly after their release (1.7 to 15 hours). Steelhead released at Skamania Landing arrived over a longer and wider time span (53 to 101 hours) (Figure 11).

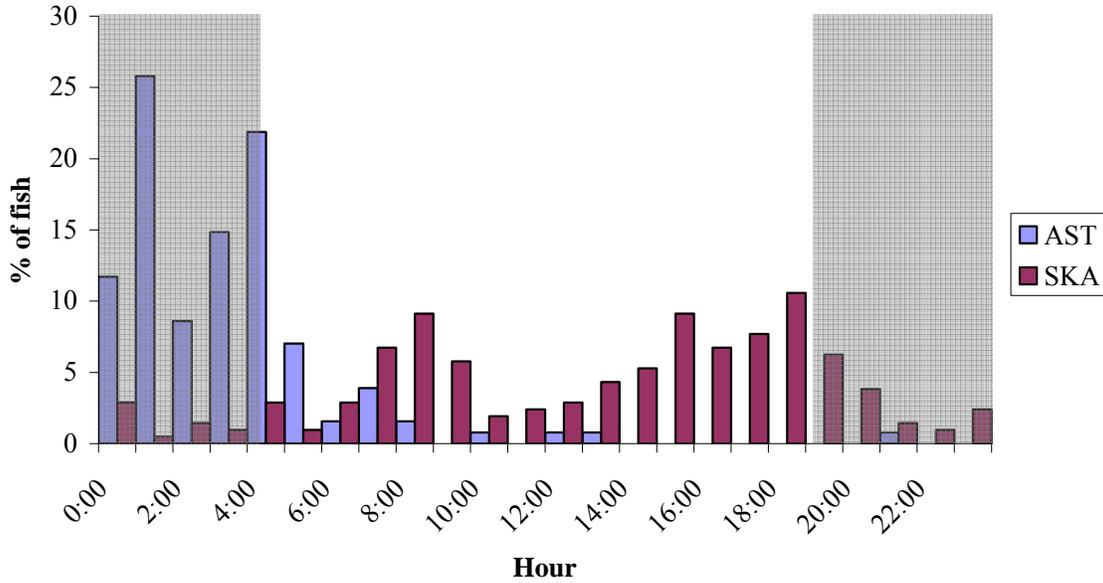


Figure 9. Percent of Fish Released at Astoria (AST) and Skamania Landing (SKA) that were Detected on the Primary Array by Hour. Shaded areas represent hours of darkness.

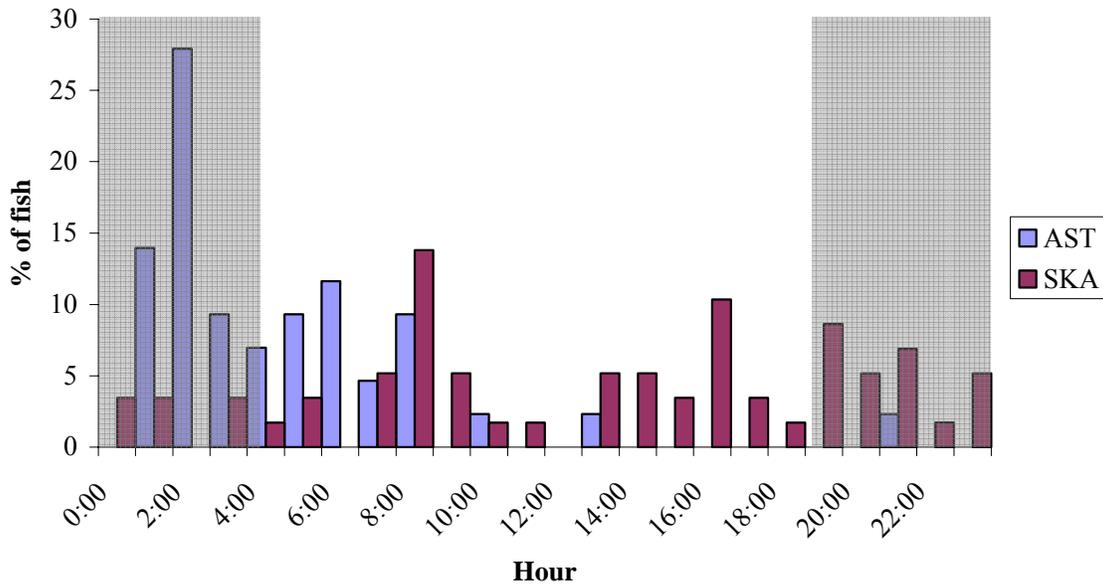


Figure 10. Percent of Fish Released at Astoria (AST) and Skamania Landing (SKA) that were Detected on the Secondary Array by Hour. Shaded areas represent hours of darkness.

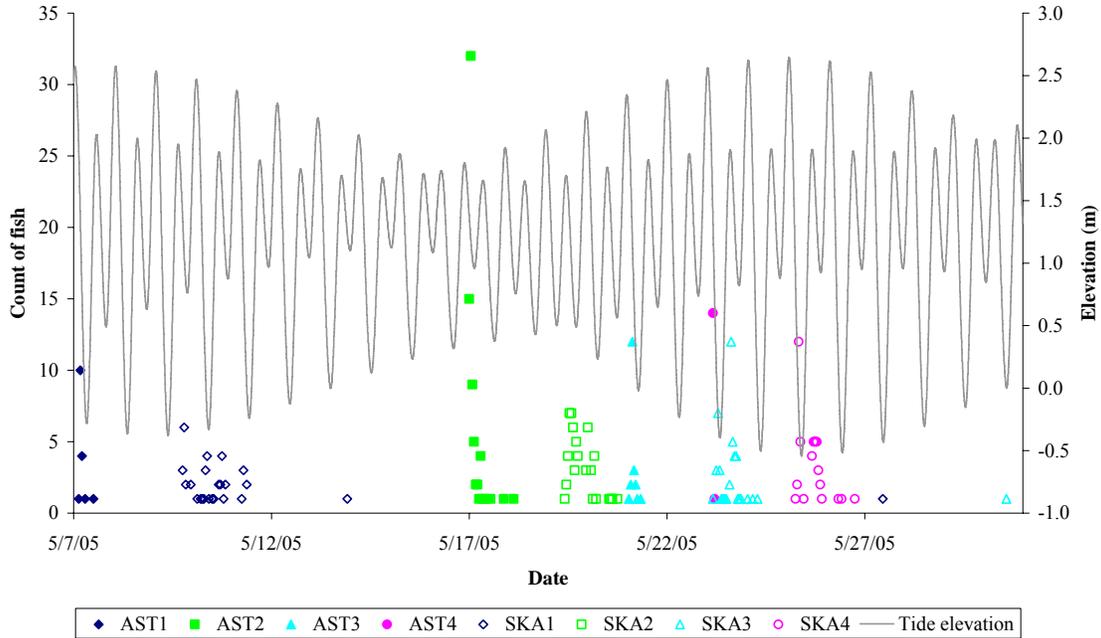


Figure 11. Count of Fish Observed per Hour on the Primary Array versus Tide Elevation (secondary y axis) at the Primary Array for Steelhead Released at the Astoria Bridge (AST) and Skamania Landing (SKA). Table 5 provides dates and times of these releases.

3.1.3 Cross-Channel Distribution

Most steelhead released at the Astoria Bridge were observed on the Oregon side of the navigation channel on the primary array. Steelhead released at Skamania Landing tended to be distributed across the channel at the primary array, with only a slightly higher proportion near the navigation channel (Figure 12). At the secondary array, there was no clear pattern of distribution for fish relative to release location. Fish from both release locations were detected along the North Jetty (Figure 13).

3.1.4 Residence Time

The mean residence time within the range of the primary array nodes for steelhead released at the Astoria Bridge was 3.9 minutes. This was significantly lower ($t=-2.665$, $df=355$, $p=0.008$) than the 112 minutes observed for steelhead released at Skamania Landing (Figure 14). The mean residence time on the secondary array was 15.2 minutes for the Astoria Bridge fish and 11.7 minutes for the Skamania Landing fish, and these were not significantly different ($t=0.451$, $df=90$, $p=0.653$) (Figure 15).

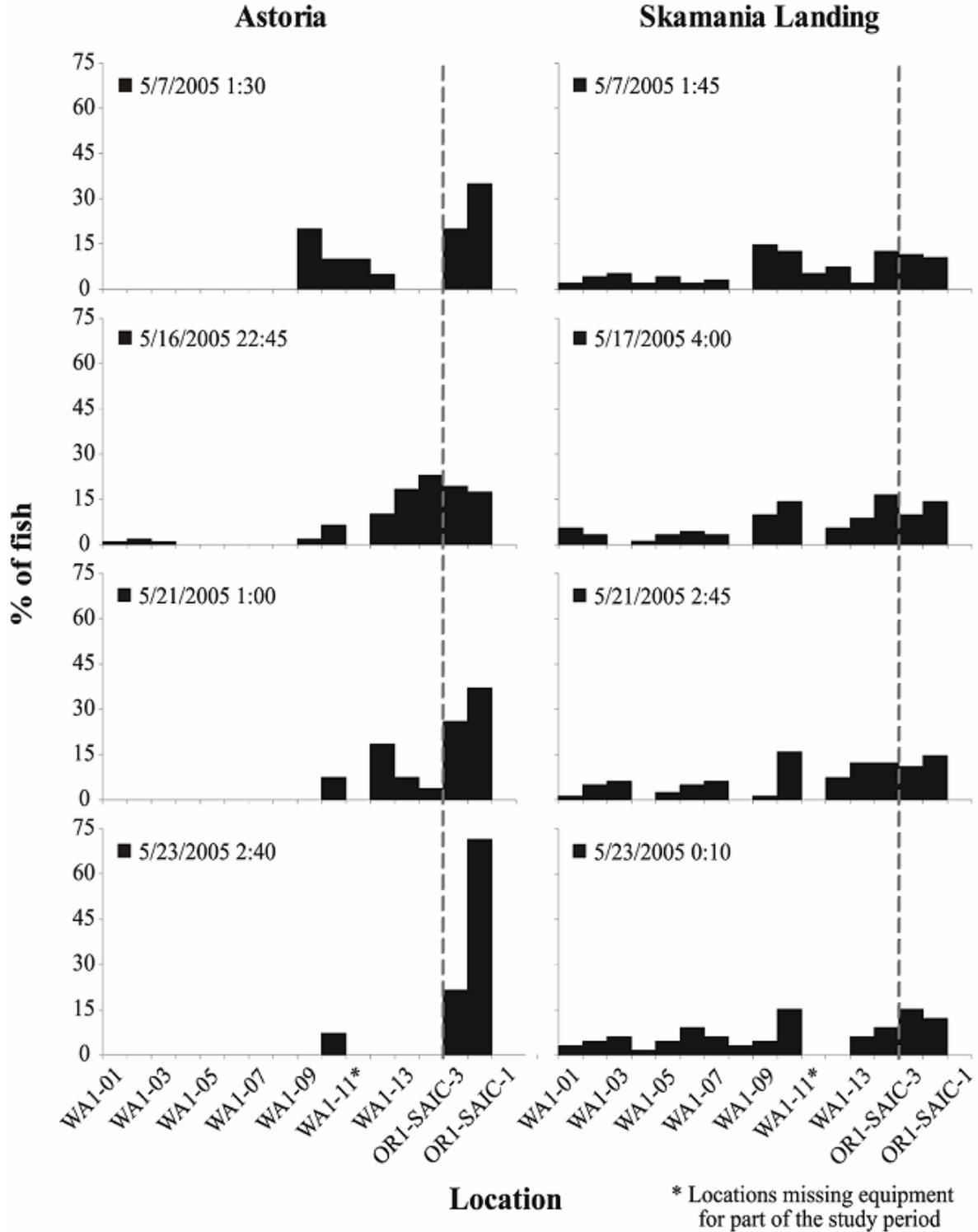


Figure 12. Cross-Channel Distribution of Steelhead Released at Astoria Bridge and Skamania Landing that were Detected on the Primary Array. The dashed lines indicate the navigation channel (where no receivers were deployed).

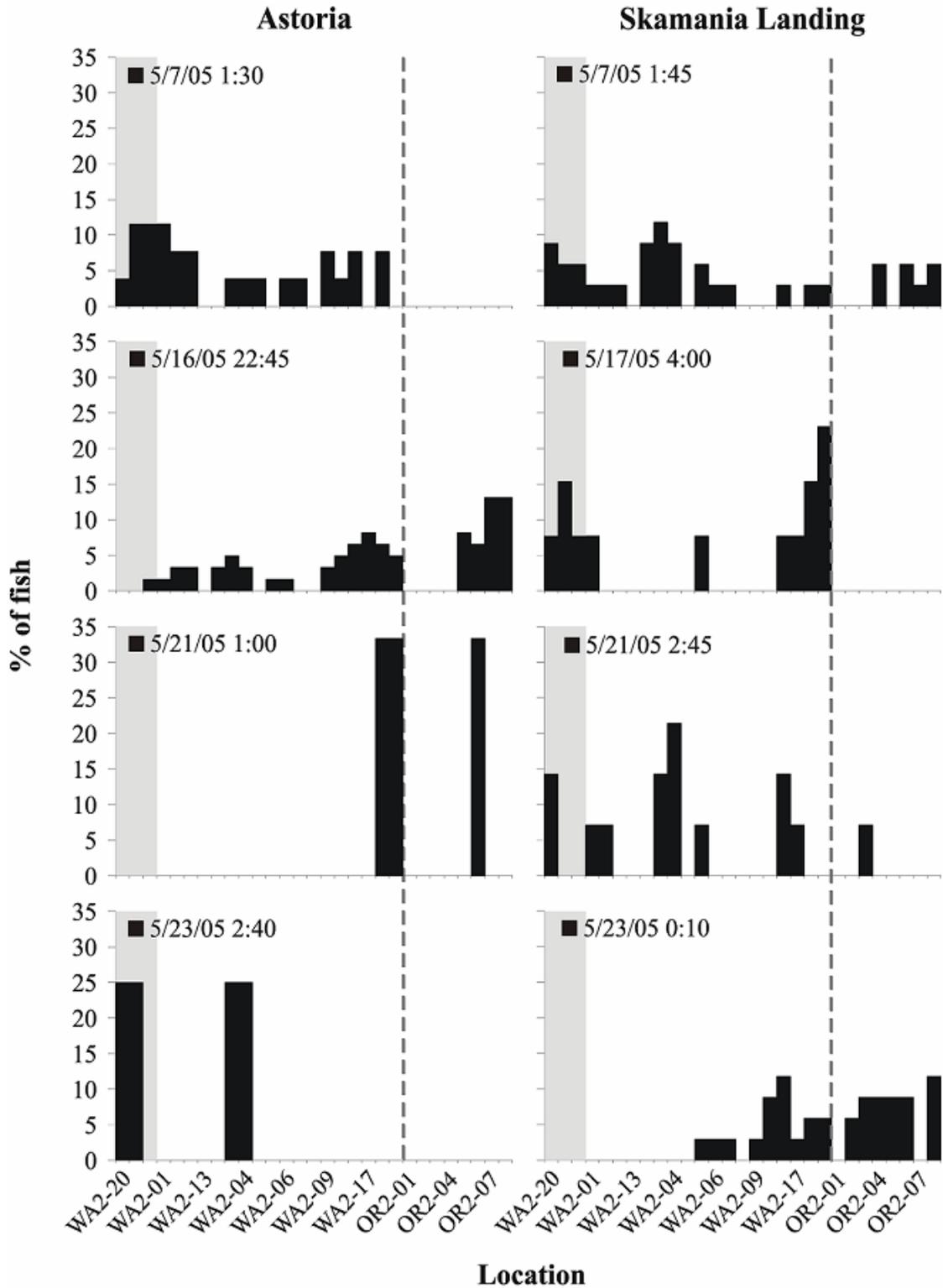


Figure 13. Cross-Channel Distribution of Steelhead Released at Astoria Bridge and Skamania Landing that were Detected on the Secondary Array. The dashed lines indicate the navigation channel (where no receivers were deployed). The shaded areas indicated nodes in the dumping ground near the North Jetty.

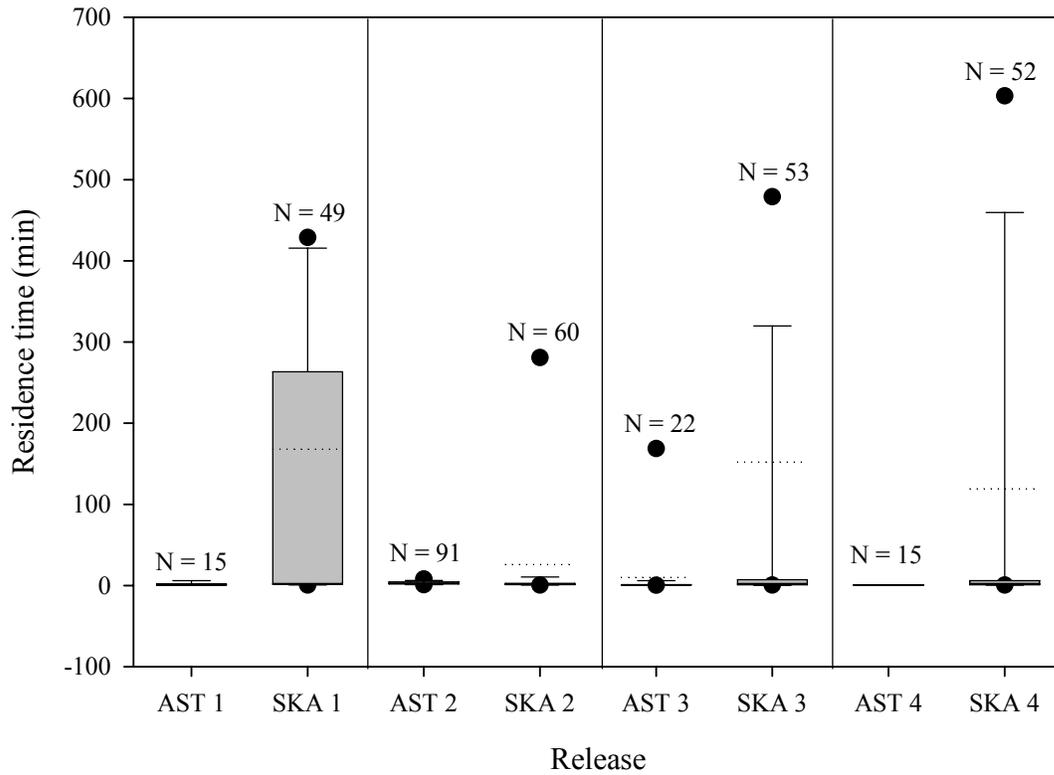


Figure 14. Residence Time (minutes) at the Primary Array of Juvenile Steelhead Released at the Astoria (AST) and Skamania Landing (SKA) Release Sites on Four Release Days. Release dates are separated by solid vertical lines. Dotted horizontal lines within box plots represent means, solid horizontal lines represent medians, upper and lower limits of the boxes represent the 25th and 75th percentiles, whiskers represent the 10th and 90th percentiles, and dots indicate the 5th and 95th percentiles.

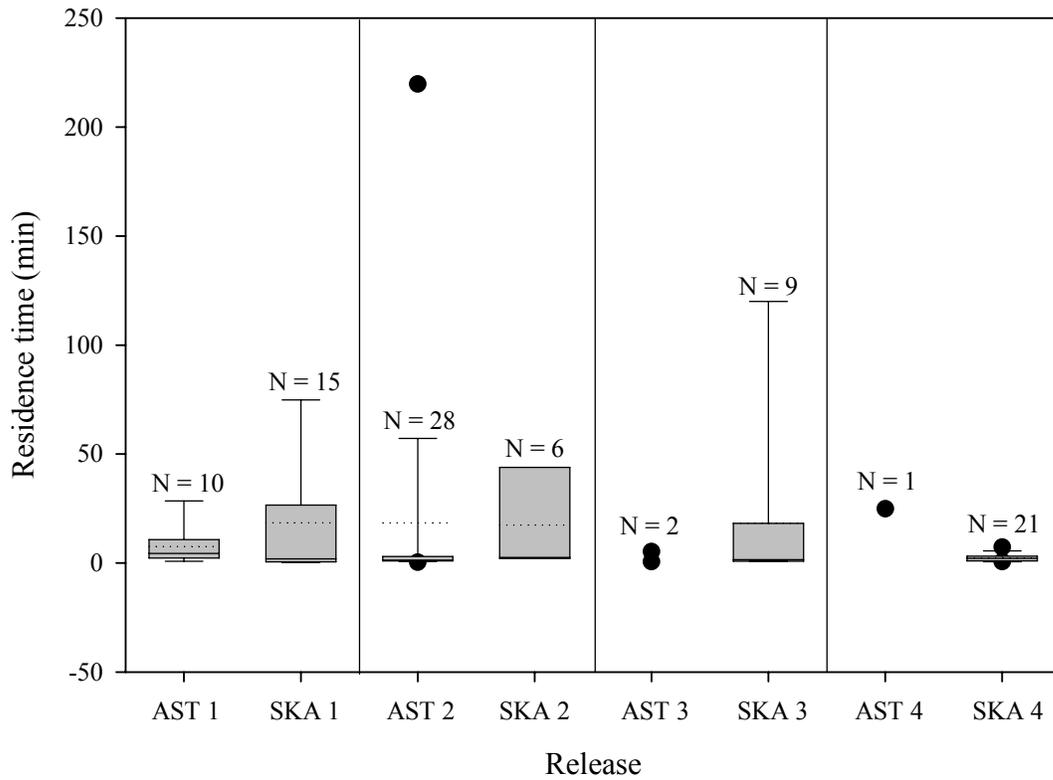


Figure 15. Residence time (minutes) at the secondary array of juvenile steelhead released at the Astoria (AST) and Skamania Landing (SKA) release sites on four release days. Release dates are separated by solid vertical lines. Dotted horizontal lines within box plots represent means, solid horizontal lines represent medians, upper and lower limits of the boxes represent the 25th and 75th percentiles, whiskers represent the 10th and 90th percentiles, and dots indicate the 5th and 95th percentiles. Dots for release times with fewer than three detections (i.e., AST 3 and AST 4) represent actual values and not percentiles.

3.1.5 Survival

It was not possible to make survival estimates for three of the four individual comparisons, due to extremes in the detection probability (Table 6). When we used a seasonal average across all four releases, the survival estimates for steelhead released at Astoria averaged 0.69 over the four releases, compared to an average of 0.63 for the Skamania releases (Table 6). These differences were not significant based on 95% confidence interval.

Table 6. Survival Estimates (S) and Detection Probability (P) for Acoustic-Tagged Steelhead Transported to the Skamania Landing Release Site and the Astoria Release Site. The standard error (S.E.) and 95% confidence intervals (C.I.) are supplied for each release date and the season total when detection probabilities are not equal to zero or one.

Release Date	Release Site	(P)	(S)	S.E.	95% C.I.	
5/7/2005	Astoria	0.40	0.39	0.14	0.12	0.66
5/7/2005	Skamania	0.47	0.62	0.16	0.30	0.93
5/16/2005	Astoria	1.00	0.82		-----NA-----	
5/17/2005	Skamania	0.67	0.56	0.16	0.25	0.88
5/21/2005	Astoria	0.00	---NA---		-----NA-----	
5/21/2005	Skamania	0.67	0.49	0.12	0.26	0.71
5/23/2005	Astoria	1.00	0.14		-----NA-----	
5/23/2005	Skamania	0.43	0.69	0.16	0.37	1.00
Total	Astoria	0.43	0.69	0.04	0.61	0.76
Total	Skamania	0.51	0.63	0.08	0.47	0.79

3.1.6 Avian Predation

The addition of an acoustic tag did not significantly influence the susceptibility of steelhead smolts to avian predation in the Columbia River estuary. However, steelhead released at the Astoria Bridge at night on an ebbing tide experienced lower avian predation rates than steelhead released much farther upstream. On the East Sand Island tern colony, 8.4% of the tags from PIT-tagged fish were recovered, while 6.6% were recovered from acoustic-tagged fish. Fewer tags were recovered from the cormorant colony, with 2.3% of the PIT-tagged fish and 1.6% of the acoustic-tagged fish detected.

Acoustic-tagged fish (which also received a PIT tag) were released on four separate occasions in May at both the Skamania and Astoria release sites (Table 7). Steelhead that were only PIT-tagged were released on three occasions in May from Skamania Landing and two occasions from Astoria Bridge (Table 7). The proportion of acoustic- and PIT-tagged and PIT-tag-only fish detected at the East Sand Island tern colony were not significantly different for the Astoria or Skamania release sites (paired t-test; $p=0.40$ and $p=0.43$), respectively (Table 7). The Astoria release groups generally resulted in lower tag proportions detected on both tern and cormorant colonies than did the Skamania release groups. However, this difference was only significant for acoustic-tagged steelhead preyed on by terns (paired t-test; $p = 0.026$).

Table 7. Percentage of Tags Recovered on the East Sand Island Caspian Tern and Double-Crested Cormorant Colonies

Acoustic-Tagged Fish				
Date Release	Caspian Tern		Double-Crested Cormorant	
	Astoria	Skamania	Astoria	Skamania
5/7/2005	0.0	13.8	1.0	0.0
5/17/2005	2.0	6.3	0.0	1.3
5/21/2005	1.1	11.9	0.0	4.4
5/23/2005	1.1	7.5	0.0	3.8
PIT-Tagged Fish				
5/7/2005				
5/17/2005	2.8	7.5	0.8	0.9
5/21/2005	1.2	10.9	0.0	4.5
5/23/2005		11.5		2.8

3.2 Pathogen Analyses

A summary of the results of PCR comparison of gill and kidney tissues for detection of *R. salmoninarum* DNA by lethal sampling of 60 steelhead smolts from each of the four barge groups used for tagging is shown in Table 8. The proportion of *R. salmoninarum*-positive fish detected by the nested PCR (nPCR) was slightly higher for gill samples than for kidney samples, but the difference was not significant ($p=0.38$). A similar trend was observed for gill and kidney samples tested from the same fish by the real-time quantitative PCR (qPCR), but this difference also was not significant ($p=0.66$). However, the proportion of *R. salmoninarum*-positive fish detected by nPCR was significantly higher than that detected by qPCR for both the gill samples ($p=0.0003$) and kidney samples ($p=0.0015$).

Among the lethally sampled fish testing positive for *R. salmoninarum*, only 14% of the kidney and gill samples from the same individual were positive by nPCR, and only 4% were positive by qPCR. Additionally, only 16% of the *R. salmoninarum*-positive gill samples were positive by both nPCR and qPCR, and only 14% of the positive kidney samples were positive by both tests. Analysis of the samples by qPCR revealed that the *R. salmoninarum* levels in the majority of samples were very low; only 4 of the 28 qPCR-positive gill samples and 1 of 24 qPCR-positive kidney samples had *R. salmoninarum* levels at or above the concentration required for consistent detection of the bacterium by qPCR (5 *R. salmoninarum* per qPCR reaction). The highest *R. salmoninarum* level detected in a gill sample was 10 bacteria per reaction (about 40 bacteria per mg of tissue), and the highest *R. salmoninarum* level detected in a kidney sample was 5 bacteria per reaction (about 20 bacteria per mg of tissue).

Testing of lethally sampled fish by nPCR showed a higher prevalence of *Nucleospora salmonis* in kidney samples than in gill samples ($p=0.0078$; Table 8). Nevertheless, among the fish testing positive for DNA of this parasite, 40% of fish with positive kidney samples also had positive gill samples. No qPCR is available for this pathogen, so infection levels could not be quantified.

Table 8. Pathogen Detection in Lethally Sampled Fish. Comparison of detection of *Renibacterium salmoninarum* by nested PCR and quantitative PCR, and detection of *Nucleospora salmonis* by nested PCR, in gill and kidney tissues from steelhead sampled lethally from the four barge groups at the time of tagging.

Test	No. <i>R. salmoninarum</i> -Positive Fish of 240 Total (%)	
	Gill	Kidney
Nested PCR	59 (25%)	50 (21%)
Quantitative PCR	28 (12%)	24 (10%)
	No. <i>N. salmonis</i> -Positive Fish of 240 Total (%)	
Nested PCR	40 (17%)	65 (27%)

Non-lethally sampled gill tissues from all of the 1,002 fish marked with microacoustic tags were analyzed for *R. salmoninarum* and *N. salmonis* by PCR (Table 9; Appendix), and one or both pathogens were detected in 550 (55%) of these fish. *R. salmoninarum* was detected by nPCR, qPCR, or both in 414 (41%) of the fish. The prevalence of *R. salmoninarum* detected by nPCR was higher ($p < 0.0001$) than that detected by qPCR. Among the fish testing positive by qPCR, *R. salmoninarum* levels were very low; only 3 of the 137 fish had levels at or above the concentration required for consistent detection of the bacterium by qPCR (5 *R. salmoninarum* per reaction, equivalent to 200 bacteria per 10 mg gill sample). *R. salmoninarum* was detected in the three highest-level fish by nPCR as well as by qPCR. The *R. salmoninarum* levels in these three fish ranged from 7 to 169 bacteria per reaction (i.e., from about 27 to 677 bacteria per mg of tissue, or from 269 to 6,774 bacteria per 10 mg gill sample) (Table 10). *N. salmonis* was detected by nPCR in 25% of the fish, and 133 (13%) of the fish were infected with both *N. salmonis* and *R. salmoninarum*.

Table 9. Pathogen Detection in Non-Lethally Sampled Fish. Detection of *Renibacterium salmoninarum* by nested PCR and quantitative PCR and detection of *Nucleospora salmonis* by nested PCR in gill tissues from steelhead sampled non-lethally at the time of tagging from the four release groups of fish marked with microacoustic tags.

Test	No. Positive Fish of 1002 Total (%)
	<i>Renibacterium salmoninarum</i>
Nested PCR	334 (33%)
Quantitative PCR	137 (14%)
	<i>Nucleospora salmonis</i>
Nested PCR	256 (25%)

There was no evidence that the presence of *R. salmoninarum* or *N. salmonis* had affected the size of the sampled steelhead. For each sample date, fish that were infected with either *R. salmoninarum* or *N. salmonis* or both pathogens did not differ significantly in length ($P \geq 0.28$) or weight ($P \geq 0.13$) from fish that were not infected with either pathogen.

Table 10. Mean Levels of *Renibacterium salmoninarum* Detected by qPCR in Non-Lethally Sampled Gill Tissue from each Release Group. The Astoria and Skamania releases are combined for each sample date. Only three of the qPCR-positive fish had *R. salmoninarum* levels above the threshold for consistent detection by this assay (200 bacteria per 10 mg gill sample).

Release Date(s) ^a	No. Positive Fish by qPCR	Geometric Mean No. <i>R. salmoninarum</i> per 10 mg gill sample (±SD)
May 7, 2005	30	51 (±2)
May 16-17, 2005	33	60 (±3) ^b
May 21, 2005	31	68 (±2) ^c
May 23, 2005	43	62 (±2) ^d

(a) Pathogen sampling dates were 5/05/2005 for the 5/07/2005 release, 5/15/2005 for the 5/16/2005 and 5/17/2005 release dates, 5/19/2005 for the 5/21/2005 release date, and 5/21/2005 for the 5/23/2005 release date.

(b) One fish in the 5/17/05 Skamania release group had a calculated concentration of 6,774 bacteria in the 10 mg gill sample.

(c) One fish in the 5/21/05 Astoria release group had a calculated concentration of 369 bacteria in the 10 mg gill sample.

(d) One fish in the 5/23/05 Skamania release group had a calculated concentration of 269 bacteria in the 10 mg gill sample.

To determine if there were differences (before transport) in pathogen prevalence between the fish groups released at the Skamania Landing and Astoria Bridge sites, we compared the proportions of pathogen-infected and uninfected fish destined for release at each site on a given date. No differences were detected between fish groups to be released at the two sites in the proportions of fish infected or not infected with *R. salmoninarum* (Table 11), with *N. salmonis* (Table 12), or in fish infected with either or both pathogens (Table 13). In addition, the proportions of fish infected with *R. salmoninarum*, *N. salmonis*, or either or both pathogens in the transport groups did not change between the first and last transport dates (chi-square test, $p \geq 0.029$). These data indicated that there was no unintentional bias in pathogen prevalence in any of the release groups; i.e., there was no difference at the time of release.

Table 11. Proportions of Fish Infected with *Renibacterium salmoninarum* in Microacoustically Tagged Groups of Steelhead Released at Astoria Bridge or Skamania Landing on Each Release Date and for all Releases during 2005. At Bonneville Dam, gill tissues were sampled non-lethally from each fish for pathogen testing before the fish were transported to the release sites. Data were analyzed by the Fisher exact test. The probability (P), odds ratio, and 95% confidence interval (95% C.I.) are given for each comparison.

Release Date ^(a)	Release Site	Percent <i>R. salmoninarum</i> -Positive ^(b)	(P)	Odds Ratio	95% C.I.
5/07/2005	Astoria	40%	0.60	0.86	0.51 – 1.45
5/07/2005	Skamania	44%			
5/16/2005	Astoria	39%	1.00	0.96	0.57 – 1.63
5/17/2005	Skamania	40%			
5/21/2005	Astoria	44%	0.17	1.49	0.88 – 2.52
5/21/2005	Skamania	35%			
5/23/2005	Astoria	39%	0.19	0.69	0.41 – 1.16
5/23/2005	Skamania	48%			
All releases	Astoria	41%	0.74	0.95	0.73 – 1.24
All releases	Skamania	42%			

(a) Pathogen sampling dates were 5/05/2005 for the 5/07/2005 release, 5/15/2005 for the 5/16/2005 and 5/17/2005 release dates, 5/19/2005 for the 5/21/2005 release date, and 5/21/2005 for the 5/23/2005 release date.

(b) Sample sizes: 90 fish tested from each Astoria release group except 92 fish sampled on 5/16/2005; 160 fish tested from each Skamania release group.

Table 12. Proportions of Fish Infected with *Nucleospora-salmonis* in Microacoustically Tagged Groups of Steelhead Released at Astoria Bridge or Skamania Landing on Each Release Date during 2005. At Bonneville Dam, gill tissues were sampled non-lethally from each fish for pathogen testing before the fish were transported to the release sites. Data were analyzed by the Fisher exact test. The probability (P), odds ratio, and 95% confidence interval (95% C.I.) are given for each comparison.

Release Date ^(a)	Release Site	Percent <i>N. salmonis</i> -Positive ^(b)	(P)	Odds Ratio	95% C.I.
5/07/2005	Astoria	20%	0.28	0.70	0.38 – 1.31
5/07/2005	Skamania	26%			
5/16/2005	Astoria	30%	1.00	1.02	0.58 – 1.78
5/17/2005	Skamania	30%			
5/21/2005	Astoria	24%	1.00	1.04	0.57 – 1.90
5/21/2005	Skamania	24%			
5/23/2005	Astoria	27%	0.54	1.25	0.69 – 2.27
5/23/2005	Skamania	23%			
All releases	Astoria	25%	1.00	0.99	0.74 – 1.33
All releases	Skamania	26%			

(a) Pathogen sampling dates were 5/05/2005 for the 5/07/2005 release, 5/15/2005 for the 5/16/2005 and 5/17/2005 releases, 5/19/2005 for the 5/21/2005 release date, and 5/21/2005 for the 5/23/2005 release date.

(b) Sample sizes: 90 fish tested from each Astoria release group except 92 fish sampled on 5/16/2005; 160 fish tested from each Skamania release group.

Table 13. Comparisons of Proportions of Pathogen-Infected Fish (fish infected with either *Renibacterium salmoninarum* or *Nucleospora salmonis*, or both pathogens) and Uninfected Fish in Microacoustically Tagged Groups of Steelhead Released at Astoria Bridge or Skamania Landing on Each Release Date during 2005. At Bonneville Dam, gill tissues were sampled non-lethally from each fish for pathogen testing before the fish were transported to the release sites. Data were analyzed by the Fisher exact test. The probability (P), odds ratio, and 95% confidence interval (95% C.I.) are given for each comparison.

Release Date ^(a)	Release Site	Percent Pathogen-Positive ^(b,c)	(P)	Odds Ratio	95% C.I.
5/07/2005	Astoria	52%	0.43	0.81	0.48 - 1.36
5/07/2005	Skamania	57%			
5/16/2005	Astoria	53%	0.60	0.86	0.52 – 1.45
5/17/2005	Skamania	57%			
5/21/2005	Astoria	56%	0.24	1.38	0.82 – 2.32
5/21/2005	Skamania	48%			
5/23/2005	Astoria	53%	0.29	0.74	0.44 – 1.25
5/23/2005	Skamania	61%			
All releases	Astoria	54%	0.55	0.92	0.71 – 1.19
All releases	Skamania	56%			

(a) Pathogen sampling dates were 5/05/2005 for the 5/07/2005 release, 5/15/2005 for the 5/16/2005 and 5/17/2005 release dates, 5/19/2005 for the 5/21/2005 release date, and 5/21/2005 for the 5/23/2005 release date.

(b) Pathogen-positive fish tested positive for either *R. salmoninarum* or *N. salmonis* or both by PCR.

(c) Sample sizes: 90 fish tested from each Astoria release group except 92 fish sampled on 5/16/2005; 160 fish tested from each Skamania release group.

Comparisons of proportions of microacoustically tagged fish that were detected by the primary or secondary array or both receiving arrays showed no differences between fish that were infected or uninfected with *R. salmoninarum* (Table 14), *N. salmonis* (Table 15), or either or both pathogens (Table 16). For these analyses, all of the release groups were combined to achieve greater numbers (i.e., greater statistical power). The fish with the highest *R. salmoninarum* infection level (Table 10) as determined by qPCR (Skamania release May 17, 2005) was detected by the primary array but not the secondary array. The other two fish with *R. salmoninarum* levels above the threshold for consistent qPCR detection (≥ 200 bacteria per gill sample, Table 10) were not detected by any of the arrays. One of these fish was in the May 21, 2005, Astoria release group and the other was in the May 23, 2005, Skamania release group.

Table 14. Comparisons of Proportions of *Renibacterium salmoninarum*-Infected Fish and Fish not Infected with this Pathogen Detected by the Primary Array, the Secondary Array, or Both Arrays after Release in 2005. Data were analyzed by the Fisher exact test. The probability (P), odds ratio, and 95% confidence interval (95% C.I.) are given for each comparison. (Sample size = 1002 fish)

Detection by Array(s)	Percent <i>R. salmoninarum</i> - Positive Fish Detected	Percent <i>R. salmoninarum</i> - Negative Fish Detected	(P)	Odds Ratio	95% C.I.
Detected by primary array	35%	32%	0.37	1.13	0.87 – 1.48
Detected by secondary array	9%	10%	0.66	0.89	0.57 – 1.37
Detected by both arrays	6%	6%	1.00	1.01	0.60 – 1.72

Table 15. Comparisons of Proportions of *Nucleospora salmonis*-Infected Fish and Fish not Infected with this Pathogen Detected by the Primary Array, the Secondary Array, or Both Arrays after Release in 2005. Data were analyzed by the Fisher exact test. The probability (P), odds ratio, and 95% confidence interval (95% C.I.) are given for each comparison. (Sample size = 1002 fish)

Detection by Array(s)	Percent <i>N. salmonis</i> - Positive Fish Detected	Percent <i>N. salmonis</i> - Negative Fish Detected	(P)	Odds Ratio	95% C.I.
Detected by primary array	35%	32%	0.48	1.12	0.83 – 1.51
Detected by secondary array	11%	9%	0.32	1.29	0.81 – 2.05
Detected by both arrays	6%	6%	0.88	1.06	0.59 – 1.92

Table 16. Comparisons of Proportions of Pathogen-Infected Fish (fish infected with either *Renibacterium salmoninarum* or *Nucleospora salmonis*, or both pathogens) and Uninfected Fish Detected by the Primary Array, the Secondary Array, or Both Arrays after Release in 2005. Data were analyzed by the Fisher exact test. The probability (P), odds ratio, and 95% confidence interval (95% C.I.) are given for each comparison. (Sample size = 1002 fish)

Detection by Array(s)	Percent Pathogen- Positive Fish Detected	Percent Pathogen- Negative Fish Detected	(P)	Odds Ratio	95% C.I.
Detected by primary array	33%	32%	0.74	1.05	0.81 – 1.37
Detected by secondary array	10%	9%	0.74	1.10	0.71 – 1.1.69
Detected by both arrays	6%	6%	0.79	1.08	0.64 – 1.83

The presence of *Renibacterium salmoninarum*, *Nucleospora salmonis*, or both pathogens did not influence the susceptibility of steelhead to avian predation by Caspian terns or double-crested cormorants on East Sand Island. Comparisons of proportions of double-tagged fish (microacoustic and PIT tags) with PIT tags found on the bird colonies showed no differences between fish that were infected with *R. salmoninarum*, *N. salmonis*, or with both pathogens: 8% tested negative and 9% tested positive ($P = 0.65$, odds ratio 1.14 with a 95% CI of 0.72 to 1.1.79). For these analyses, data from all of the pathogen analyses, all release groups, and from both the tern and cormorant colonies were combined to achieve sufficient numbers. The PIT tags from the three fish with the highest *R. salmoninarum* infection levels as determined by qPCR were not detected on East Sand Island.

4.0 Discussion

The primary goal of this pilot study was to assess whether survival of steelhead smolts to ocean entry could be increased by utilizing the Astoria Bridge release site which is 200 km downstream of the traditional release site at Skamania Landing. The fish were also released at night on an outgoing tide to further decrease exposure to avian predators in the lower Columbia River estuary.

Our findings showed that tags from steelhead released at Astoria Bridge were found in significantly lower proportions on avian bird colonies in the Columbia River estuary than their Skamania Landing counterparts, thus validating our assumption that avian predation in the estuary would be reduced by releasing steelhead smolts at the Astoria Bridge at night on an outgoing tide. However, even though it appears that losses to avian predators were significantly reduced, we did not find a statistically significant difference in survival to ocean entry between the Astoria Bridge and Skamania Landing release locations.

There are several possible reasons for this result including small sample size and high variability in results and lack of test sensitivity to detect differences. In addition, the detectability of steelhead released at the Astoria Bridge site may have been lower than for fish released at Skamania Landing, thus violating the assumption of equal detectability required by the survival model. The fish released at the Astoria Bridge Site moved very rapidly and tended to be distributed in and around the navigation channel more so than that Skamania Landing fish. Because no receivers had been placed in the navigation channel, the detection efficiency of acoustically tagged fish was lower there than in areas outside of the navigation channel.

No statistical difference was found in predation rates between PIT-tagged and acoustic-tagged fish. There are several factors that could explain the lack of a significant difference between the proportions of acoustic-tagged and PIT-tagged steelhead detected on piscivorous bird colonies. First, the acoustic tag used in this study was designed for juvenile salmonids to a minimum of 100 mm in length and, in this study, the steelhead used were considerably larger (mean length >240 mm). Due to proportional size differences between the acoustic tag and the steelhead, we did not expect the effect of the tag to be significant. In addition, due to the limited number of steelhead available to tag, we were able to detect only large differences in predation rates, allowing small differences to go undetected.

This study demonstrated the potential utility of non-lethal sampling of salmonids for detection of pathogens such as the BKD agent *Renibacterium salmoninarum* and the microsporidian parasite *Nucleospora salmonis*.

Although the nPCR detected up to twice as many *R. salmoninarum*-positive fish as the qPCR in gill and kidney tissue samples in this study, only the qPCR could provide information on *R. salmoninarum* infection levels in the fish. Therefore, both tests are recommended for obtaining maximum information on the prevalence and levels of the pathogen in a population. The proportion of *R. salmoninarum*-positive fish detected in the steelhead population did not differ significantly regardless of whether the test tissue was gill or kidney, but the bacterium was detected in gill and kidney tissues from the same individual by nPCR in only 14% of the fish and by qPCR in only 4% of the fish. The low overall *R. salmoninarum* infection levels likely influenced this result; only 5 of the 240 fish tested in the lethal sampling groups and 3 of the 1,002 fish tested in the non-lethal sampling groups had *R. salmoninarum* levels at or above the concentration required for consistent detection of the bacterium by qPCR (Chase et al. in press). Because the tissue samples used for PCR are small (10 to 25 mg), detecting bacteria that are present at very low concentrations in these samples may be a “hit-or-miss” situation. Contamination of tissues that

are exposed to the environment (e.g., gill) by *R. salmoninarum* present in the water might also influence results and should be investigated.

Comparisons of proportions of microacoustically tagged fish that were detected by the primary or secondary array or both arrays showed no differences between fish that were uninfected and those that were infected with one or both pathogens. Similarly, this research showed no detectable influence of the presence or absence of these pathogens on avian predation rates.

These results for the *R. salmoninarum*-infected fish are not surprising, considering the extremely low infection levels detected in most of the fish. The only three fish with infection levels above the threshold for consistent qPCR detection were not detected by any of the arrays, nor were PIT tags from these fish detected on the bird colonies. These three fish had infection levels equivalent to low-to-medium *R. salmoninarum* antigen levels as detected by the enzyme-linked immunosorbent assay (ELISA) (Chase et al. in press) and likely would not have had clinical lesions. Although steelhead and rainbow trout are susceptible to bacterial kidney disease (BKD), they are less susceptible to the disease than some other salmonids such as Chinook and sockeye salmon (Sanders et al. 1978; Starliper et al. 1997). In previous studies in which Columbia and Snake River steelhead out-migrants were tested for *R. salmoninarum* by ELISA at Lower Granite, Priest Rapids, and McNary dams, the majority of hatchery steelhead testing positive for *R. salmoninarum* showed low antigen levels (Pascho and Elliott 1989; Elliott and Pascho 1991, 1992). In contrast, wild steelhead sampled at these dams at the same time consistently showed higher levels of *R. salmoninarum* antigen than the hatchery fish, although it was not determined whether any of the fish had active infections.

This research did not demonstrate a relation between *N. salmonis* infection and detection of fish by one or more arrays or susceptibility of fish to bird predation, but the *N. salmonis* infection levels could not be determined. Natural infections with this intranuclear parasite have been associated with acute or chronic mortality, particularly in Chinook salmon (Elston et al. 1987; Hedrick et al. 1990; Morrison et al. 1990). Chronic disease can result in poor growth, secondary infections, and low-grade mortality in Chinook salmon (Hedrick et al. 1990), but the impact on steelhead is largely unknown.

5.0 Conclusions and Recommendations

Based on our study of survival differences in barged steelhead smolt survival from the traditional release site at Skamania Landing (rkm 226.9) and at an alternative site 200 km downstream at Astoria Bridge (rkm 22.5) 2005, we conclude the following:

- Most steelhead released at the Astoria Bridge passed the primary array on a single ebb tide, while the fish released at the Skamania Landing site arrived over a period of days and encountered several ebb and flood tides.
- Most steelhead released at the Astoria Bridge (65%) arrived at the primary array during hours of darkness, while there was no clear pattern in the time of arrival for steelhead released at Skamania Landing; only 14% arrived at the primary array during hours of darkness.
- Most steelhead released at the Astoria Bridge were observed on the Oregon side of the navigation channel on the primary array. Steelhead released at Skamania Landing tended to be distributed across the channel at the primary array, with only a slightly higher proportion near the navigation channel.
- The mean residence time within the range of the primary array nodes for steelhead released at the Astoria Bridge was 3.9 minutes. This was significantly lower than the 112 minutes observed for steelhead released at Skamania Landing.
- It was not possible to make survival estimates for three of the four individual comparisons, due to extremes in the detection probability.
- When we used a seasonal average across all four releases, the survival estimates for steelhead released at Astoria averaged 0.69 over the four releases, compared to an average of 0.63 for the Skamania releases.
- The survival difference to ocean entry between the Astoria Bridge and Skamania Landing release locations was not statistically significant.
- The addition of an acoustic tag did not significantly influence the susceptibility of steelhead smolts to avian predation in the Columbia River estuary.
- Steelhead released at the Astoria Bridge at night on an ebbing tide experienced lower avian predation rates than steelhead released at Skamania Landing.
- There was no statistical difference in survival to the primary or secondary array for fish infected or not infected with either the BKD agent *Renibacterium salmoninarum*, the microsporidian parasite *Nucleospora salmonis*, or both pathogens.
- There was no detectable influence of *Renibacterium salmoninarum* or *Nucleospora salmonis* on avian predation rates.
- The *R. salmoninarum* infection levels in the majority (99%) of positive steelhead were very low; the *N. salmonis* infection levels could not be determined.

While we did not detect significant differences in survival of steelhead smolts released at the different locations, it may be that the more important question is which release site will provide the greatest smolt to adult return ratio (SAR). It is conceivable that survival to ocean entry could be essentially the same between groups but that one group might return as adults at a higher rate than the

other due to latent effects of passage history and pathogen exposure on ocean survival. If the overall goal is to increase adult salmonid returns, the only way to be certain that this is achievable is to measure SARs using a long-term tag, such as a PIT tag or coded wire tag. While the microacoustic tag provides useful information for identifying behavior patterns, migration rates, time of passage through the avian predation areas, areas of loss, and survival from release to ocean entry, its life is not yet long enough for adult return studies.

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Appendix

Detection and Pathogen Sample Histories for All Fish Released

(data on attached CD)

Appendix

Detection and Pathogen Sample Histories for All Fish Released

The data shown here are only a small portion to illustrate format and content. The full data file is included on the CD included with the report (File name: Final Acoustic + Pathogens All data.csv).

Appendix column heading descriptions:

Project ID: The project for which the data were collected.

Release Date: Date and time (military time PDT) the fish were released.

Release Location: Location of fish release.

Species: Code used by the PTAGIS system to identify fish; 32H is the code for hatchery-origin steelhead.

Dead: Indicates whether the fish was alive at the time of release (0 = alive, 1 = dead).

Acoustic_ID: Unique acoustic-tag code that identifies each fish to the acoustic receivers.

Length: Fork length in mm

Weight: weight in g

WA 1: Indicates whether the fish was detected on the northern (Washington) portion of the primary array located in the Columbia River estuary (0 = not detected, 1 = detected).

OR 1: Indicates whether the fish was detected on the southern (Oregon) portion of the primary array (0 = not detected, 1 = detected).

WA 2: Indicates whether the fish was detected on the northern (Washington) portion of the secondary array located at the mouth of the Columbia River (0 = not detected, 1 = detected).

OR 2: Indicates whether the fish was detected on the southern (Oregon) portion of the secondary array (0 = not detected, 1 = detected).

1° Array Detection: Indicates whether the fish was detected anywhere on the primary array (0 = not detected, 1 = detected).

2° Array Detection: Indicates whether the fish was detected anywhere on the secondary array (0 = not detected, 1 = detected).

Both Arrays: Indicates whether the fish was detected on both primary and secondary arrays (0 = not detected on both arrays, 1 = detected on both arrays).

Time of First 1° Array Detection: Date and time of first detection of the fish on the primary array.

Time of First 2° Array Detection: Date and time of first detection of the fish on the secondary array.

PIT_ID: Unique PIT-tag code that identifies each fish to the PIT-tag detectors.

Tagger: Name of surgeon who performed the acoustic-tag implantation.

E. Sand Terns: Indicates whether the PIT- and/or acoustic-tag from the fish was detected/found on the tern colony on East Sand Island.

E. Sand Corm: Indicates whether the PIT- and/or acoustic-tag from the fish was detected/found on the cormorant colony on East Sand Island.

Gill_Sample: Number used to identify the gill tissue sample taken from the fish for pathogen analyses.

nPCR Rs: Indicates whether the fish tested positive (1) or negative (0) for *Renibacterium salmoninarum* by nested PCR.

qPCR Rs: Indicates whether the fish tested positive (1) or negative (0) for *Renibacterium salmoninarum* by real-time quantitative PCR.

No. Rs/Gill Snip: Number of *Renibacterium salmoninarum* detected per gill sample by real-time quantitative PCR (gill tissue samples were assumed to be about 10 mg in weight).

nPCR Ns: Indicates whether the fish tested positive (1) or negative (0) for *Nucleospora salmonis* by nested PCR.

ProjectID	Release Date	Release Location	Species	Dead	Acoustic_ID	Length(mm)	Wt(gm)	WA 1	OR 1	WA 2	OR 2	1 st Array Detection	2 nd Array Detection	Both Arrays	Time of first 1 st Array Detection	Time of first 2 nd Array Detection	PIT_ID	Tagger	E. Sand Terns	E. Sand Corm	Gill_Sam ple	nPCR Rs	qPCR Rs	No. Rs/Gill snip	nPCR Ns	
BARGE	5/7/05 1:30	Astoria	32H	0	G72478F26	245	125.7	0	0	0	0	0	0	0			3D9_1BF22CD8RICH		0	0	0.0254	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G724788A5	242	116.3	0	0	1	0	0	0	0		5/7/2005 6:31	3D9_1BF22CC4KATE		0	0	0.0197	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72478406	242	118.5	0	0	0	0	0	0	0			3D9_1BF22A7RICH		0	0	0.0162	1	0	0	1	
BARGE	5/7/05 1:30	Astoria	32H	0	G7247A966	254	146.7	0	1	0	0	0	1	0	0	5/7/2005 5:08	3D9_1BF22BE4KATE		0	0	0.0251	1	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72475C2D	244	108.4	0	0	0	0	0	0	0			3D9_1BF22CE8RICH		0	0	0.0172	1	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72475BAE	242	111.7	0	0	0	0	0	0	0			3D9_1BF229F7KATE		0	0	0.0169	0	0	0	1	
BARGE	5/7/05 1:30	Astoria	32H	0	G7247570D	247	122.6	0	1	0	0	0	1	0	0	5/7/2005 4:40	3D9_1BF22F70BRAD		0	0	0.0220	1	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72475653	252	126.5	0	0	0	0	0	0	0			3D9_1BF229FFRICH		0	0	0.0248	1	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G7247430C	258	151.5	0	0	0	0	0	0	0			3D9_1BF22F49KATE		0	0	0.0177	1	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G724742AF	249	125.9	0	0	0	0	0	0	0			3D9_1BF22A88RICH		0	0	0.0250	0	0	0	1	
BARGE	5/7/05 1:30	Astoria	32H	0	G7245C531	261	134.6	0	1	0	0	0	1	0	0	5/7/2005 5:08	3D9_1BF22CC4KATE		0	0	0.0245	1	0	0	1	
BARGE	5/7/05 1:30	Astoria	32H	0	G7245C00E	238	98.1	0	0	0	0	0	0	0			3D9_1BF22F4E8RICH		0	0	0.0178	1	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G7245B5C9	247	138.1	0	0	0	0	0	0	0			3D9_1BF22F41RICH		0	0	0.0252	0	1	23	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G7245B497	243	115.7	0	0	0	0	0	0	0			3D9_1BF22F33RICH		0	0	0.0182	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G7245AA15	274	168.9	0	0	0	0	0	0	0			3D9_1BF22CCHKATE		0	0	0.0215	0	1	88	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G7245A6B6	233	109.4	0	0	1	0	0	1	0	0	5/7/2005 4:28	3D9_1BF22CB7BRAD		0	0	0.0208	1	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G724595EA	229	91.7	0	0	0	0	0	0	0			3D9_1BF22A05BRAD		0	0	0.0206	1	0	0	1	
BARGE	5/7/05 1:30	Astoria	32H	0	G72459269	253	124.7	0	0	0	0	0	0	0			3D9_1BF225F4BRAD		0	0	0.0238	0	0	0	1	
BARGE	5/7/05 1:30	Astoria	32H	0	G72458E57	233	105.5	0	0	0	0	0	0	0			3D9_1BF22F46RICH		0	0	0.0168	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72455DE2	270	192.5	0	0	0	0	0	0	0			3D9_1BF22F69KATE		0	0	0.0227	1	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G724544E3	234	97.8	0	0	0	0	0	0	0			3D9_1BF22CE8RICH		0	0	0.0204	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72446B45	251	144.1	0	0	1	0	0	1	0	0	5/7/2005 4:49	3D9_1BF22CB7KATE		0	0	0.0249	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72446404	264	153.2	0	0	0	0	0	0	0			3D9_1BF22F39KATE		0	0	0.0192	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G724459FB	259	151.6	0	0	0	0	0	0	0			3D9_1BF22F39RICH		0	0	0.0200	0	0	0	1	
BARGE	5/7/05 1:30	Astoria	32H	0	G72445AA5	229	102.5	0	0	0	0	0	0	0			3D9_1BF22A05KATE		0	0	0.0184	1	1	68	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72445947	229	86.7	0	0	0	0	0	0	0			3D9_1BF22CF6RICH		0	0	0.0185	1	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72444118	247	129.7	0	0	0	0	0	0	0			3D9_1BF22CE6BRAD		0	0	0.0226	1	0	0	1	
BARGE	5/7/05 1:30	Astoria	32H	0	G724434DF	246	121.2	0	0	0	0	0	0	0			3D9_1BF22CCRICH		0	0	0.0213	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G724431E0	242	110.5	0	0	0	0	0	0	0			3D9_1BF22CBBRICH		0	0	0.0244	1	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72442DDE	254	138	1	0	0	0	0	1	0	0	5/7/2005 4:46	3D9_1BF22CB3RICH		0	0	0.0222	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G724427A0	235	106.5	0	0	0	0	0	0	0			3D9_1BF22CC8RICH		0	0	0.0198	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G7244229F	239	113.8	0	0	0	0	0	0	0			3D9_1BF22CF8RICH		0	0	0.0188	0	0	0	1	
BARGE	5/7/05 1:30	Astoria	32H	0	G72442023	208	74.7	0	0	0	0	0	0	0			3D9_1BF229FBBRAD		0	0	0.0232	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72441BB0	244	124.8	0	0	0	0	0	0	0			3D9_1BF22CB8KATE		0	0	0.0209	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G724415A2	229	102.2	0	0	0	0	0	0	0			3D9_1BF22CD0BRAD		0	0	0.0201	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G724414FC	226	98.1	0	0	0	0	0	0	0			3D9_1BF22F68RICH		0	0	0.0228	1	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72440DFD	246	117.1	0	0	0	0	0	0	0			3D9_1BF22D5F8RICH		0	0	0.0256	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G7243E23B	244	115.3	0	0	0	0	0	0	0			3D9_1BF22CC8KATE		0	0	0.0205	1	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72430E26	252	125.9	0	0	0	0	0	0	0			3D9_1BF22CE8KATE		0	0	0.0171	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72430E45	242	114.5	0	0	0	0	0	0	0			3D9_1BF22CC3BRAD		0	0	0.0203	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G7243BE43	246	117.5	0	0	0	0	0	0	0			3D9_1BF22CD0BRAD		0	0	0.0235	0	0	0	1	
BARGE	5/7/05 1:30	Astoria	32H	0	G7243B102	218	104.6	0	0	0	0	0	0	0			3D9_1BF22CCRICH		0	0	0.0166	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G7243AA40	244	110.6	1	0	0	0	0	1	0	0	5/7/2005 4:18	3D9_1BF22A05BRAD		0	0	0.0223	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G7243A0C1	238	115.2	0	0	0	0	0	0	0			3D9_1BF22A93RICH		0	0	0.0210	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G724399E3	236	107.2	0	0	0	0	0	0	0			3D9_1BF22CE4KATE		0	0	0.0165	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72438BC2	230	100.5	0	0	0	0	0	0	0			3D9_1BF22F48KATE		0	0	0.0195	1	0	0	1	
BARGE	5/7/05 1:30	Astoria	32H	0	G724380E2	241	118.4	1	0	0	0	1	1	0	0	5/7/2005 5:01	5/7/2005 5:51	3D9_1BF22CE7RICH		0	0	0.0194	0	1	48	0
BARGE	5/7/05 1:30	Astoria	32H	0	G72436534	238	113.9	0	0	0	0	0	0	0			3D9_1BF22CF4KATE		0	0	0.0255	1	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G7243646A	201	64.2	0	0	0	0	0	0	0			3D9_1BF22D40RICH		0	0	0.0260	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G724354D4	242	108.9	0	0	0	0	0	0	0			3D9_1BF22F63RICH		0	0	0.0239	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72434F69	249	142.4	0	0	0	0	0	0	0			3D9_1BF22F70RICH		0	0	0.0242	0	1	83	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72434A56	243	131.5	0	0	0	0	0	0	0			3D9_1BF22CF7BRAD		0	0	0.0229	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72434517	236	103.2	0	0	0	0	0	0	0			3D9_1BF22CB3KATE		0	0	0.0234	1	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72433812	225	90.2	0	0	1	0	0	1	0	0	5/7/2005 5:23	3D9_1BF22CD8RICH		0	0	0.0236	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G7243360D	245	125.2	0	0	0	0	0	0	0			3D9_1BF22CF4BRAD		0	0	0.0190	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72433FOC	234	100.7	0	0	0	0	0	0	0			3D9_1BF22CBBRICH		0	0	0.0225	0	1	34	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G72432690	232	104.2	0	0	0	0	0	0	0			3D9_1BF22A01RICH		0	0	0.0174	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G7243204D	237	116.4	0	1	0	0	0	1	0	0	5/7/2005 4:19	3D9_1BF22A05KATE		0	0	0.0189	0	0	0	1	
BARGE	5/7/05 1:30	Astoria	32H	0	G7242EDBE	252	119.8	0	0	0	0	0	0	0			3D9_1BF22F59KATE		0	0	0.0181	0	0	0	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G7242E57C	234	113.7	0	0	0	0	0	0	0			3D9_1BF22CE8KATE		0	0	0.0221	0	1	38	0	
BARGE	5/7/05 1:30	Astoria	32H	0	G7242DBDD																					

