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Evaluation of Larval Pacific Lamprey Occupancy of Habitat Restoration Sites in the Portland Harbor Superfund Area

2019 Annual Report

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On the cover: *Photograph of St. Johns Bridge over the Willamette River. (Photo by J. Barkstedt).*

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Abstract – Habitat restoration actions focused on the recovery of juvenile Chinook salmon *Oncorhynchus tshawytscha* are being implemented in the Portland Harbor Superfund area of the Willamette River. These actions may also have effects on co-occurring Pacific Lamprey *Entosphenus tridentatus*. Use of restored habitats by lampreys, particularly the larval life stage has not been extensively studied. As such, there is a need to monitor the effectiveness of these efforts, in part, relative to larval Pacific Lamprey. Determining the effects of habitat restoration actions on Pacific Lamprey requires evaluation of lamprey occurrence before and after project implementation. Currently, this study is focused on the occupancy of larval Pacific Lamprey and *Lampetra* spp. in shoreline, confluence, and tributary habitats at five restoration sites being constructed to provide compensation for injuries to natural resources as part of the Portland Harbor Natural Resource Damage Assessment (NRDA): Alder Point, Harborton, Linnton, Triangle Park, and Rinearson. In addition, the study is evaluating the occupancy of lamprey at a non-NRDA site, PGE 13.1, located in a reach of the Willamette River that bisects the city of Portland. In 2019, sampling at restoration sites was conducted at Alder Point and Rinearson. We also evaluated whether larval Pacific Lamprey occupied corresponding habitats at seven reference sites in the Portland Harbor Superfund area (Multnomah Channel, McCarthy Creek, Columbia Slough, Ross Island, Cemetery Creek, Oswego Creek, and Miller Creek). A generalized random tessellation-stratified approach was used to select random, spatially-balanced sample quadrats (30 m x 30 m square) across the lower Willamette River, Multnomah Channel as well as reaches (50 m) in wadeable tributaries. At the Alder Point site, two larval lamprey were detected at one of thirty confluence quadrats (detection probability (d) = 0.03) and zero of ten shoreline quadrats (d = 0.00). No lamprey were detected in any of the ten quadrats (d = 0.00) in the newly constructed Alder Slough. Quadrat-specific detection probability at Alder Point, post-implementation (2016- 2019) has been similar to that pre-implementation (2014). At the seven reference sites, larvae were detected at McCarthy Creek, Cemetery Creek, Oswego Creek, Oswego Creek and Miller Creek. A total of 38 larval lamprey were captured and quadrat-specific detection rates ranged from 0.00 to 0.71. This information is being used as part of a long-term evaluation of the effects of habitat restoration on occupancy and distribution of larval lamprey in the Portland Harbor Superfund area.

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Introduction

Pacific Lamprey *Entosphenus tridentatus* in many areas, such as the Columbia River Basin (CRB), appear to have experienced a decline in abundance (Close et al. 2002) and have been given protected status within Oregon (Kostow 2002). Lamprey are culturally important to Native American tribes, are ecologically important within the food web, and are an indicator species whose decline provides further insight into the impact of human actions on ecological function (Close et al. 2002). Much information is lacking on the basic biology, ecology, and population dynamics that is required for effective conservation and management.

Pacific Lampreys have a complex life history that includes a multiple year larval, migratory juvenile, and adult marine phase (Scott and Crossman 1973). Larvae are strongly associated with stream and river sediments. Larvae live burrowed in stream and river sediments for multiple years after hatching, where they filter feed detritus and organic material (Sutton and Bowen 1994). Larval metamorphosis into juveniles occurs from July to December (McGree et al. 2008) and major migrations of juveniles are made downstream to the Pacific Ocean in the fall and spring (Beamish and Levings 1991). The sympatric Western Brook Lamprey *Lampetra richardsoni* does not have a major migratory or marine life stage although adults may exhibit local upstream migrations before spawning (Renaud 1997). For both species, the majority of the information on distribution and habitat preference of larvae comes from CRB tributary systems (Moser and Close 2003; Torgersen and Close 2004; Stone and Barndt 2005; Stone 2006) and coastal basins (Farlinger and Beamish 1984; Russell et al. 1987; Gunckel et al. 2009).

Larval lamprey are known to occur in sediments of low-gradient streams (<5th order [1:100,000 scale]; Torgersen and Close 2004) but their use of relatively large river habitats and deep areas is not well understood. Downstream movement of larvae, whether passive or active, appears to occur year-round (Nursall and Buchwald 1972; Gadomski and Barfoot 1998; White and Harvey 2003). Larval Sea Lamprey *Petromyzon marinus* have been documented in deepwater habitats in tributaries of the Great Lakes, within the lakes in proximity to river mouths (Hansen and Hayne 1962; Wagner and Stauffer 1962; Lee and Weise 1989; Bergstedt and Genovese 1994; Fodale et al. 2003), and in large water bodies associated with the St. Marys River (Young et al. 1996). However, references to other species occurring in deepwater or lacustrine habitats are scarce (American Brook Lamprey *L. appendix*; Hansen and Hayne 1962). In the Pacific Northwest, observations of larval lamprey occurrence in large rivers have been made, for example during smolt monitoring operations at Columbia River hydropower facilities, impinged on screens associated with juvenile bypass systems (Moursund et al. 2003; CRITFC 2008), or through observation during dewatering events. Specific collections of larvae have been made in large river habitats in British Columbia, which are thought to be representative of larvae dispersing downstream (Beamish and Youson 1987; Beamish and Levings 1991). More recently, evaluations of larval Pacific Lamprey occupancy and distribution in mainstem river habitats have suggested widespread occurrence in certain areas of the Columbia River and Willamette River mainstem (Jolley et al. 2012; Harris and Jolley 2017).

A portion of the mainstem of the lower Willamette River, an area that is known to be occupied by larval Pacific and Western Brook Lamprey (Jolley et al. 2012), was declared a Superfund Site in 2000 by the U.S. Environmental Protection Agency. The Superfund study area extends

from river kilometer 3.2 to river kilometer 18.9 and has a broader focus area extending from the Columbia River to Willamette Falls (Figure 1). To compensate for past environmental damage being identified through the Natural Resource Damage Assessment (NRDA) process, this area is subject to various habitat restoration projects as well as assessments of the effectiveness of these projects. For context, restoration is being used as a broad and general term intended to capture efforts to restore, rehabilitate, replace, or acquire the equivalent (see Rosenzweig 2003; Roni et al. 2008) of those natural resources injured as the result of hazardous substance and oil releases from the Portland Harbor Superfund site (NOAA 2017). Presently, the projects in the aquatic environment are focused on recovering juvenile Chinook Salmon *Oncorhynchus tshawytscha*. It is unclear whether any of these projects will provide benefits to other co-occurring species including larval and juvenile Pacific Lamprey that may likewise occur in these areas. However, these projects provide an opportunity to understand the potential effects of habitat restoration on larval and juvenile lampreys. As such, there is interest in monitoring the effectiveness of the projects, in part, relative to larval Pacific Lamprey.

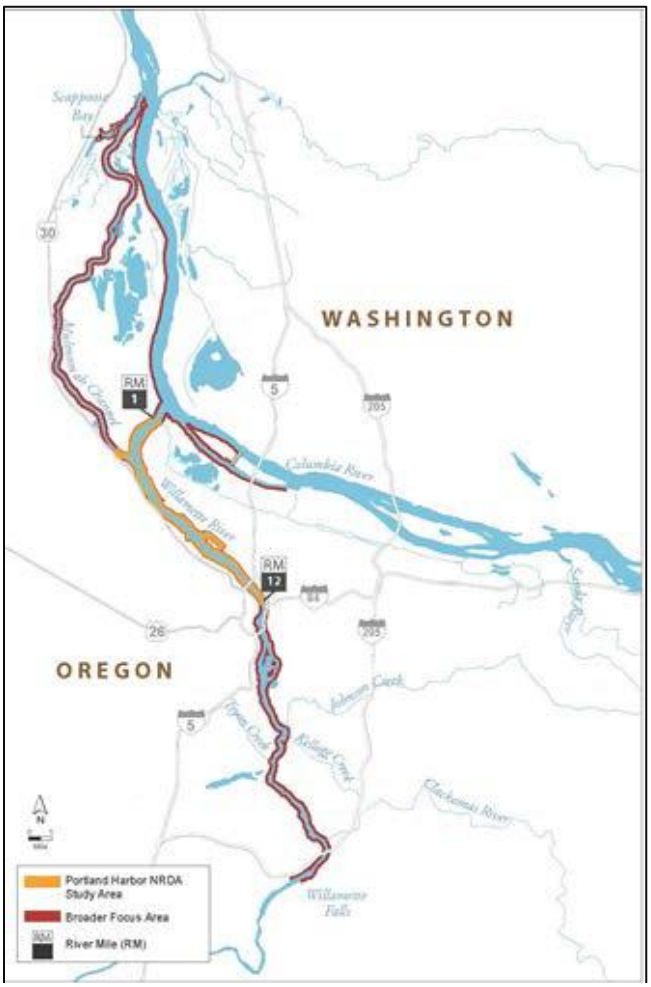

Figure 1. Portland Harbor Superfund study area (orange outline) and the broader focus area (red outline) on the lower Willamette River.

A lamprey monitoring plan (LMP) for habitat restoration projects in the Portland Harbor Superfund area was developed based on a set of monitoring goals and objectives that were identified by the Trustee Council and lamprey biologists over two workshops held in the fall of 2011. The LMP priorities included (i) monitoring the impact of projects on larval and juvenile lamprey populations and health in Portland Harbor, and (ii) gathering information about larval and juvenile lamprey life history, biology, and habitat requirements that could be used by the Trustee Council to inform future design and evaluation of habitat restoration projects targeting lamprey. Since lamprey biology and life history are different from many other aquatic biota, the overlap between the LMP and the general monitoring and stewardship plan is not extensive. The LMP differs from the general monitoring and stewardship plan, in part, because the lamprey monitoring is proposed to continue for a period of 20 years. In most cases, the metrics proposed for collection as part of the lamprey monitoring effort need to be co-located with lamprey sampling. To maximize efficiencies, the Trustee Council will, to the extent possible, use data collected as part of the LMP for general monitoring and stewardship. Biologists

recommended monitoring lamprey for 20 years after the completion of a habitat restoration project, with the goal of capturing data for one to two complete generations. Pre-implementation monitoring will be conducted to the extent practical at each project site. Lampreys have the ability to colonize habitats rapidly. Therefore, monitoring will be conducted on a yearly basis for the first five years after project implementation, and every five years thereafter. In general, the proposed work is guided by the LMP. However, due to site-specific conditions and constraints, the specific metrics and timing of monitoring proposed for any given site may differ slightly from those outlined in the LMP.

Understanding larval lamprey usage of areas in and adjacent to rehabilitated habitat is critical to gauging the effectiveness of these projects. At present, little specific information is available on whether lampreys will colonize rehabilitated habitats, which life stages may use these habitats, or how quickly and for how long they will use these habitats. A before-after control-impact (BACI) approach is being used to evaluate the effectiveness of habitat restoration projects. The BACI approach allows us to make inferences about whether observed changes in lamprey occupancy are the result of the habitat restoration projects. Our sampling in 2019 was part of an ongoing effort to determine whether larval Pacific Lamprey occupy restoration sites and reference sites both prior to and after project implementation. Our specific objectives for this phase of NRDA monitoring were as follows:

1. Determine whether lamprey occupy restoration sites in the lower Willamette River and Multnomah Channel.
2. Determine whether lamprey occupy reference sites in the lower Willamette River and Multnomah Channel.
3. Determine the types of habitat available at each site and in which habitat types lamprey are detected.
4. Characterize lamprey species and life history stage that occupy each site.
5. Evaluate the health of lamprey detected at each site.

Study Sites

Restoration Sites

Alder Point

The Alder Point site is located at the southern tip of Sauvie Island (Multnomah County, OR), and thus is bordered on the east side by the Willamette River (at approximately river km 6), and on the west side by Multnomah Channel (Figure 2). As part of a habitat restoration project, slough habitat (henceforth Alder Slough) was constructed through the site, connecting the Willamette River and Multnomah Channel. Unlike the typical confluence habitat in the Superfund area (a tributary or slough having a single confluence with the mainstem), Alder Slough (one water body) has three distinct confluence habitats, two in the main Willamette River and one in the

Multnomah Channel. Restoration of shoreline habitat (i.e., levee removal) also occurred along both the Willamette River and Multnomah Channel (Figure 3). Pre-implementation sampling was conducted in 2014. Post-implementation sampling began in 2016.

Harborton

The Harborton site is located on the southwest side of the Willamette River at river km 5.1, near the confluence of Multnomah Channel (Figure 2). The site contains the Harborton Wetlands, a remnant black cottonwood and ash floodplain forest wetland area that provides off-channel habitat, floodplain function, and habitat connectivity between the river and Forest Park. Currently the site has wadeable tributary or slough habitat (henceforth referred to as tributary habitat) as well as a confluence and shoreline habitat (Figure 4). Proposed actions include improvements to the tributary habitat. In the case of the Harborton site, monitoring needs include tributary habitat (since this habitat is currently believed to be inaccessible to fish, monitoring would be post-implementation only) and the confluence habitat (pre- and post-implementation). Pre-implementation sampling was conducted in 2017.

Linnton

The Linnton site is located on the southwest side of the Willamette River at river km 7.5 just upstream of Sauvie Island (Figure 2). It is an industrial property that contains an inactive plywood company. Currently the site has tributary habitat as well as a confluence habitat and associated shoreline habitat (Linnton Creek, Figure 5). The current tributary habitat runs through a pipe, underground and is not accessible to fish. Lampreys are not believed to occupy or have access to the underground tributary habitat being proposed for restoration. In the case of the Linnton site, monitoring needs include newly exposed tributary habitat (since this habitat is currently believed to be inaccessible to fish) post-implementation only, as well as the confluence and shoreline habitat (pre- and post-implementation). Pre-implementation monitoring was conducted at confluence and shoreline habitats in the Willamette River in 2017 (Skalicky et al. 2018). Post-implementation monitoring would consist of sampling in tributary habitats as well as shoreline and confluence habitats in the Willamette River.

Triangle Park

The Triangle Park site is located on the east side of the Willamette River, near the University of Portland, Oregon (Figure 2). There is a proposed action to improve shoreline and riparian habitat at and remove pilings from the site (Figure 6). In the case of the Triangle Park site, monitoring needs include shoreline habitat and habitat areas around the existing pilings (pre- and post-implementation). Pre-implementation monitoring was conducted at shoreline and piling habitats in the Willamette River in 2017 (Skalicky et al. 2018). Post-implementation monitoring would consist of sampling shoreline habitats in and areas where pilings had been removed from the Willamette River.

Rinearson Natural Area

The Rinearson Natural Area (RNA) site is located at river km 39. Rinearson Creek flows

through the RNA (Clackamas County, OR) and enters the Willamette River from the east, just downstream of the mouth of the Clackamas River (Figure 2). The site has tributary habitat that drains into the Willamette River, as well as associated confluence habitat in the mainstem Willamette River (Figure 7). A project has been proposed to improve and redirect tributary habitat at this site. Pre-implementation monitoring was conducted at confluence habitat in the Willamette River mainstem as well as wadeable depth tributary habitat in Rinearson Creek in 2015 (Silver et al. 2016). Post-implementation monitoring consisted of sampling for larval lamprey in tributary reaches in Rinearson Creek as well as confluence habitats in the mainstem Willamette River.

PGE 13.1

In addition to the NRDA restoration sites described above, Portland General Electric (PGE) requested that the FWS evaluate a similar, non-NRDA restoration action. The PGE 13.1 restoration site is near kilometer 21.1 of the Willamette River (Figure 2). Although restoration at PGE 13.1 (Figure 8) is not specifically related to the NRDA process and outside of the formal Superfund area, it is within the reach of the Willamette River that bisects the city of Portland. The PGE 13.1 restoration site and the NRDA restoration sites have many commonalities in regard to the types of habitats being restored and the biological questions being addressed. As such, the FWS is applying a similar lamprey monitoring approach at the PGE 13.1 and the NRDA restoration sites as well as including all results in this report.

The PGE 13.1 site is located on the east side of the Willamette River. Portland General Electric has proposed a project to rehabilitate the habitat on the east bank of the Willamette River at this site (Figure 2). The site has shoreline habitat with associated city effluents. It was unknown whether lamprey occupied the site (Figure 8). In the case of the PGE 13.1 site, monitoring needs include shoreline habitat (pre- and post-implementation). Pre-implementation monitoring was conducted at shoreline habitats in the Willamette River in 2017 (Skalicky et al. 2018). Unlike the design for the NRDA projects, post-implementation sampling for this PGE project was designed to occur in years 2, 4, 6, and 8 post-implementation. As such, post-implementation monitoring was conducted in 2019 and is scheduled to be conducted in 2021, 2023 and 2025.

Reference Sites

Seven reference sites have been identified throughout the lower Willamette River and Multnomah Channel (Figure 2). Initially, we attempted to monitor habitats from six of these reference sites. However, after various attempts to sample some of the habitats at these sites, it became clear that all the desired reference habitat was not available (e.g. barriers near tributary mouths, unwadeable conditions, private ownership with lack of access). Thus, in 2019 we began sampling tributary habitat at the Oswego Creek reference site and one additional reference site (Miller Creek). Currently, various habitat combinations from seven different sites are being used for reference. Reference sites were selected in locations that contained confluence, shoreline, or tributary habitats and in sites not proposed for habitat restoration in the immediate future. The reference sites were chosen in an attempt to provide confluence, shoreline, or tributary habitats that are similar to those which may exist at restoration sites following project implementation.

Multnomah Channel (Shoreline)

The Multnomah Channel site is located just downstream of the McCarthy Creek (near river km 24; Figure 2). The Multnomah Channel reference site contains shoreline habitat (Figure 9). The Multnomah Channel site is not currently paired with a specific restoration site, but currently serves as a reference site for shoreline habitat.

McCarthy Creek (Confluence and Shoreline)

The McCarthy Creek site is located where McCarthy Creek enters the Multnomah Channel from the southwest, downstream of the Sauvie Island Bridge (near river km 29; Figure 2). The McCarthy Creek reference site provides confluence and shoreline habitats in Multnomah Channel (Figure 10). Although the McCarthy Creek site has a tributary, this habitat is not conducive to sampling (e.g. poor flows, not wadeable) and was discontinued as a tributary reference. The McCarthy Creek site serves as a specific reference for the Linnton site.

Columbia Slough (Confluence)

The Columbia Slough site is located where the Columbia Slough enters the Willamette River from the east, near the confluence of the Willamette and Columbia Rivers (near river km 2; Figure 2). Confluence habitat occurs in the mainstem Willamette River associated with the mouth of Columbia Slough (Figure 11). The Columbia Slough site serves as a specific reference for the Harborton site.

Ross Island (Shoreline)

The Ross Island site, located just upstream of the Ross Island Bridge near downtown Portland (near river km 24; Figure 2), contains shoreline habitat (Figure 12). The Ross Island site serves as a specific reference for the Alder Point site.

Cemetery Creek (Confluence and Shoreline)

The Cemetery Creek site is located where enters the Willamette River from the west, upstream of Ross Island (near river km 27; Figure 2). The Cemetery Creek reference site has tributary habitat that drains into the Willamette River, as well as confluence and shoreline habitats in the mainstem Willamette River (Figure 13). The Cemetery Creek site, specifically the tributary and confluence habitat, was selected to serve as a specific reference for the RNA site. Although the Cemetery Creek site has a tributary, this habitat is not conducive to monitoring (e.g. lack of Type I habitat and a putative passage barrier) and was discontinued as a tributary reference. In place of the Cemetery Creek tributary habitat as a reference, in 2019 we began sampling the tributary habitat at the Oswego Creek site. The confluence habitat at the Cemetery Creek site continues to serve as a reference for the RNA site.

Oswego Creek (Confluence and Tributary)

The Oswego Creek site is located where Oswego Creek enters the Willamette River from the west, near the town of Lake Oswego (near river km 34; Figure 2). Confluence habitat occurs where the tributary habitat enters the mainstem Willamette River (Figure 14). Prior to 2019, sampling focused on the confluence habitat at the Oswego Creek site. In 2019, we began sampling the tributary habitat to supplement existing tributary reference sites. The tributary habitat associated with the Oswego Creek site (Cemetery Creek site replacement) was added to serve as a specific reference for the tributary habitat at the RNA restoration site. The confluence habitat associated with the Oswego Creek site is not currently paired with a specific restoration site but serves as a general reference site for confluence habitat.

Miller Creek (Tributary)

Miller Creek enters the Multnomah Channel (Figure 2) from the south and just opposite of the upstream end of Sauvie Island (Figure 15). In 2019, we began sampling this habitat to supplement tributary reference sites. The Miller Creek site was added to serve as a specific reference for the Linnton site (McCarthy Creek replacement).

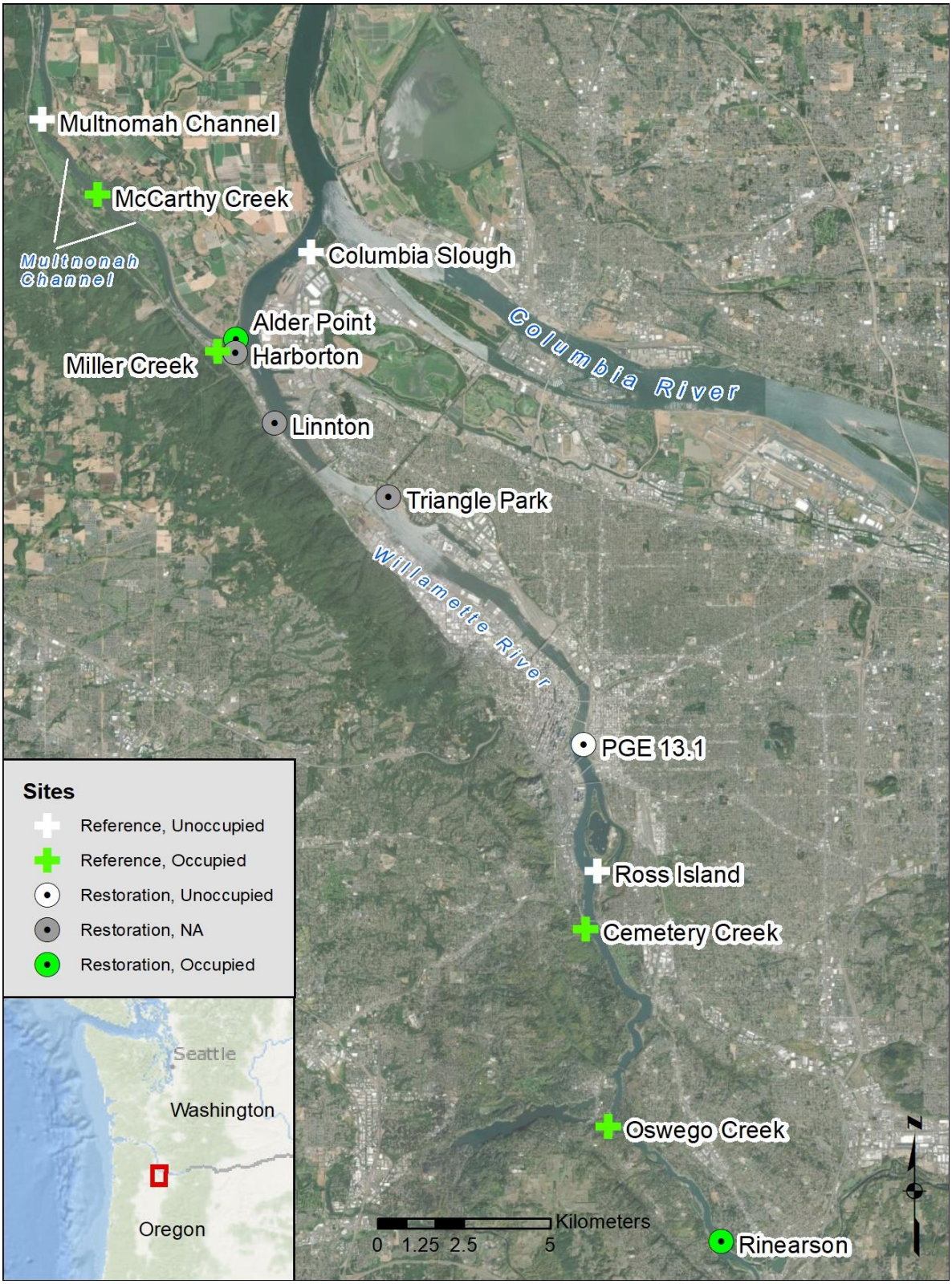

Figure 2. Locations of the restoration (circles) and reference (crosses) sites sampled in 2019. Occupied sites are green. Unoccupied sites are in white. Sites not sampled are in grey. *Note: PGE 13.1 is not a NRDA restoration site.*

Methods

Sample Framework

We evaluated occupancy of larval lamprey in the restoration and reference sites by adapting an approach that has been applied previously to studies of larval lamprey occupancy in the Columbia River basin in both mainstem and tributary habitats (Silver et al. 2010; Jolley et al. 2012; Jolley et al. 2013; Jolley et al. 2014). The approach has several requirements: 1) a unit- and gear-specific detection probability (assumed or estimated); 2) the probability of presence (given no detection) at a predetermined acceptably low level; and 3) random identification of spatially balanced sample units that allow estimation of presence and refinement of detection probabilities. A unit-specific probability of detection, d_{unit}, was calculated as the proportion of sample quadrats or reaches in which larvae were captured. The posterior probability of area occupancy, given a larval lamprey was not detected, was estimated as:

$$(1)\ \mathrm{P}(F|C_o) = \frac{\mathrm{P}(C_o|F)\cdot\mathrm{P}(F)}{\mathrm{P}(C_o|F)\cdot\mathrm{P}(F) + \mathrm{P}(C_o|\sim F)\cdot\mathrm{P}(\sim F)},$$

where P(F) is the prior probability of larval lamprey presence. Although in this case we knew the lower Willamette River was occupied with larval lamprey, a P(F) of 0.5 (uninformed) was used for future study design (i.e., P[$F|C_o$]) in areas where larval lamprey presence is unknown. P(~F), or 1 – P(F), is the prior probability of species absence, and P($C_o|F$), or 1 – d, is the probability of not detecting a species when it occurs (C_0 = no detection; Peterson and Dunham 2003). Random identification of spatially-balanced sample units was achieved by using a generalized random-tessellation stratified (GRTS) approach to delineate sample units in an ordered, unbiased manner (Stevens and Olsen 2004).

Tributary (Wadeable Water) Sample Framework

Evaluation of larval lamprey occupancy of wadeable depth tributary habitats was conducted at restoration sites pre- and post-restoration. For each tributary habitat longer than 400 m, we developed a layer of 50 m-long sample reaches for subsampling. The GRTS approach was again used to delineate sample reaches in a random, spatially-balanced order (Stevens and Olsen 2004). The GRTS method assigns a hierarchical order to the reaches within the creek which is used as an unbiased method of ranking the priority of reaches for sampling. Delineation of sample reaches that are unbiased, randomly selected, and spatially-balanced within a sample universe allows for calculation of unit-specific detection probabilities. In turn, unit-specific estimates of detection probability can be applied to determine sample effort necessary for achieving a desired level of certainty that a tributary is not occupied by lamprey when they are not detected. As they are selected in the GRTS approach, the lower numbered reaches are given highest priority for sampling. Here we used a subsampling effort (number of sample reaches) that would allow for at least 80% certainty that larval lampreys do not occupy at least 20% of a tributary habitat when they were not detected (see Bayley and Peterson 2001; Peterson and Dunham 2003). The amount of effort was based, in part, on estimates of reach-specific detection probabilities generated from previous work (Silver et al. 2010; USFWS unpublished data). Sample effort was also dependent, in part, on total area. For wadeable depth tributaries, if the area of interest was less than 400 m in length, we sampled all reaches (contiguous 50 m reaches). If the area of

interest was 400 m or longer, we sampled seven reaches.

Confluence and Shoreline (Deep Water) Sample Framework

Sample quadrats at confluence and shoreline habitats were derived from the work of Jolley et al. (2012). Quadrats were delineated using the generalized random-tessellation stratified (GRTS) approach scripted in Program R (Stevens and Olsen 2004; Jolley et al. 2012; R Core Team, 2013). The GRTS method assigns a hierarchical order to quadrats, which can be used as an unbiased method of ranking the priority of quadrats for sampling. Delineation of quadrats that are unbiased, randomly selected, and spatially-balanced within a sample universe allows for calculation of unit-specific detection probabilities. In turn, unit-specific estimates of detection probability can be applied to determine sample effort necessary for achieving a desired level of certainty that an area is not occupied by lamprey when they are not detected. Here we used a sampling effort (number of sample quadrats) that we estimated would allow for at least 80% certainty that larval lampreys do not occupy at least 20% of a confluence or shoreline habitat when they were not detected (see Bayley and Peterson 2001; Peterson and Dunham 2003). The amount of effort was based, in part, on estimates of quadrat-specific detection probabilities generated from previous work (Jolley et al. 2012). Sample effort was also dependent, in part, on total area. This sample effort corresponded to sampling of 10 quadrats at each confluence and/or shoreline habitat at both restoration and reference sites. In the case where slough habitat was deep and not wadeable, the sample framework described above was also applied to the slough (as a sample unit).

Restoration Sites

Alder Point (Slough, Confluence and Shoreline)

Confluence quadrats at the Alder Point site comprised a subset of quadrats filtered from the lower Willamette River and Multnomah Channel layers (described above). Quadrats were filtered from the larger layers according to the placement of a semicircular buffer of 100 m radius centered on each confluence of the Alder Slough and the Willamette River or Multnomah Channel (Figure 3). The three branches of Alder Slough each form a distinct confluence habitat at Alder Point, two occur on the Willamette River and one occurs on Multnomah Channel. In this case, the confluence quadrat selection process was duplicated at each of the three confluence habitats (Figure 3), resulting in 60 total sample quadrats at the three Alder Slough confluence habitats. At each of the three confluence locations, the 10 lowest numbered of each of the confluence quadrats as ordered by the GRTS method were assigned the highest priority for sampling.

Shoreline quadrats at the Alder Point site also comprised a subset of quadrats filtered from the lower Willamette River and Multnomah Channel layers (described above). Quadrats were filtered from the larger layers according to the placement of a 100 m-wide polygon, from the waterline perpendicular 100 m into the Willamette River or Multnomah Channel. The length of the shoreline polygon was determined by the project area boundaries (Figure 3). The shoreline quadrat selection process resulted in 117 total sample quadrats adjacent to restored shorelines at Alder Point. The 10 lowest numbered shoreline quadrats as ordered by the GRTS method were

assigned the highest priority for sampling.

To evaluate larval lamprey occupancy of Alder Slough, a layer of 30 m x 30 m quadrats was developed and overlaid on the newly constructed channel at Alder Point. Using the GRTS approach, quadrats in Alder Slough were delineated in a random spatially-balanced manner. The lowest 10 numbered quadrats were assigned the highest priority for sampling.

Harborton (Tributary and Confluence)

At the Harborton restoration site (Figure 4), we anticipate sample effort will correspond to 10 confluence quadrats in each of three confluence habitats (30 total quadrats, to be done pre and post restoration). We anticipate the sample effort will also correspond to 6-8, 50 m tributary reaches in each of two tributaries (post-restoration only).

Linnton (Tributary, Confluence and Shoreline)

At the Linnton restoration site (Figure 5), we anticipate the sample effort will correspond to 6-8, 50 m tributary reaches (post-restoration only). Pre- (and post-) restoration sample effort will also correspond to 10 confluence quadrats and 10 shoreline quadrats.

Triangle Park (Shoreline)

We propose to determine whether larval Pacific Lamprey occupy the restoration area both prior to and after piling removal. In this unique case, shoreline sample framework is being defined as the area 30 m around the line connecting the piling structures (see Figure 6). Pre- (and post-) restoration sample effort will correspond to 21 shoreline quadrats (25% of the total number of quadrats).

Rinearson Natural Area (Tributary and Confluence)

At the Rinearson Natural Area, Rinearson Creek forks into two distributary channels near the Willamette River creating two distinct confluence habitats in the restoration site. In this case, the confluence quadrat selection process was carried out as described above at Alder Point, and duplicated at each of the two distinct confluence habitats (Figure 7). The selection process resulted in 34 total sample quadrats at the two confluence habitats. At each of the two confluence locations, the lowest numbered quadrats as ordered by the GRTS method were assigned the highest priority for sampling. Evaluation of larval lamprey occupancy in Rinearson Creek post-restoration is proposed to occur over an approximately 1,200 m long segment of creek, spanning from the confluence with the Willamette River upstream to the crossing of River Road (Milwaukie, OR; Figure 7). Sample reaches were delineated at a rate of one 50 m reach for every 50 m of stream. Thus, within the approximately 1,200 m long study area in Rinearson Creek, 24 sample reaches were delineated, of which the lowest numbered reaches, as ordered by the GRTS method, were assigned the highest priority for sampling. Because the area of interest in Rinearson Creek was longer than 400 m, sampling effort will correspond to seven, 50 m-long reaches in the creek.

PGE 13.1 (Shoreline)

To evaluate larval lamprey occupancy of PGE 13.1, a layer of 30 m x 30 m quadrats was developed and overlaid on the restoration area (shoreline polygon) at PGE 13.1 (Figure 8). Using a GRTS approach, quadrats at PGE 13.1 were delineated in a random spatially-balanced manner. The lowest 10 numbered quadrats were assigned the highest priority for sampling.

Reference Sites

Multnomah Channel (Shoreline)

The Multnomah Channel reference site contains shoreline habitat (Figure 9). The quadrat selection process was carried out as described above for shorelines at Alder Point. The length of the shoreline was modeled after that of restoration sites. The 10 lowest numbered shoreline quadrats as ordered by the GRTS method were again assigned the highest priority for sampling.

McCarthy Creek (Confluence and Shoreline)

The McCarthy Creek reference site has tributary, confluence and shoreline habitats. Evaluation of larval lamprey occupancy in McCarthy Creek occurred over an approximately 1,350 m long segment of creek, spanning from the confluence with the Multnomah Channel upstream to the crossing of Highway 30 (near Burlington, OR). Because the area of interest in McCarthy Creek was longer than 400 m, we visited seven 50 m-long GRTS reaches in the creek. Sample reaches were delineated at a rate of one 50 m reach for every 50 m of stream. Thus, within the approximately 1,350 m long study area in, 27 sample reaches were delineated, of which the lowest numbered seven reaches as ordered by the GRTS method were assigned the highest priority for sampling (Figure 10). Upon visiting the site we discovered that the tributary did not contain a reasonable amount of Type I habitat. In McCarthy Creek confluence habitat within the Multnomah Channel, quadrat selection was carried out as described above for confluences at Alder Point. The 10 lowest numbered confluence quadrats as ordered by the GRTS method were again assigned the highest priority for sampling. In shoreline habitat within the mainstem Multnomah Channel, quadrat selection was carried out as described above for shoreline habitat at Alder Point. The 10 lowest numbered shoreline quadrats as ordered by the GRTS method were again assigned the highest priority for sampling (Figure 10).

Columbia Slough (Confluence)

The Columbia Slough reference site contains confluence habitat within the mainstem Willamette River. The confluence quadrat selection was carried out as described above for confluences at Alder Point. The 10 lowest numbered confluence quadrats as ordered by the GRTS method were again assigned the highest priority for sampling (Figure 11).

Ross Island (Shoreline)

The Ross Island reference site contains shoreline habitat. The quadrat selection process was carried out as described above for shorelines at Alder Point. The length of the shoreline was modeled after that of restoration sites. The 10 lowest numbered shoreline quadrats as ordered by the GRTS method were again assigned the highest priority for sampling (Figure 12).

Cemetery Creek (Confluence and Shoreline)

The Cemetery Creek reference site has tributary, confluence and shoreline habitats. In Cemetery Creek, the tributary area of interest was less than 400 m in length, spanning from the confluence with the Willamette River upstream approximately 300 m to a reach of very high gradient. Because the area of interest was less than 400 m in length, we attempted to sample all viable reaches (contiguous 50 m reaches) in Cemetery Creek up to a total of 350 m. For confluence habitat within the mainstem Willamette River, quadrat selection was carried out as described above for confluences at Alder Point. The 10 lowest numbered confluence quadrats as ordered by the GRTS method were again assigned the highest priority for sampling. For shoreline habitat within the mainstem Willamette River, quadrat selection was carried out as described above for shoreline habitat at Alder Point. The 10 lowest numbered shoreline quadrats as ordered by the GRTS method were again assigned the highest priority for sampling (Figure 13).

Oswego Creek (Confluence and Tributary)

The Oswego Creek reference site contains confluence habitat within the mainstem Willamette River. The confluence quadrat selection was carried out as described above for confluences at Alder Point. The 10 lowest numbered confluence quadrats as ordered by the GRTS method were again assigned the highest priority for sampling (Figure 14). The Oswego Creek reference site also has tributary habitat. In Oswego Creek, the tributary area of interest was less than 400 m in length. The area spanned from the confluence with the Willamette River upstream approximately 370 m to a reach with significant private ownership. Because the area of interest was not more than 400 m in length, we attempted to sample all viable reaches (contiguous 50 m reaches) in Oswego Creek up to a total of 350 m (the most downstream 350 m in Oswego Creek).

Miller Creek (Tributary)

Miller Creek has tributary habitat. In Miller Creek, the tributary area of interest was less than 400 m in length, spanning from the confluence with the Multnomah Channel upstream approximately 350 m to a high gradient reach with significant private ownership. Because the area of interest was not more than 400 m in length, we attempted to sample all viable reaches (contiguous 50 m reaches) in Miller Creek up to a total of 350 m (the most downstream 350 m in Miller Creek).

Reach Sampling in Wadeable Water Habitats

For wadeable depth tributary (or slough) habitats, each sampling event consisted of electrofishing 50 m reaches to determine if larval lamprey were present (Silver et al. 2010).

Sample reaches were accessed on foot using GPS units loaded with sample reach UTMs for navigation. When a reach could not be sampled due, for example, to dewatered conditions, excessive depth (e.g. > 2 m), or lack of access due to private property, they were eliminated and subsequent reaches were increased in priority. Once a sample reach was accessed, a 50 m segment was measured and flagged. Water temperature and conductivity were recorded in each reach. The reach was electrofished using an AbP-2 backpack electrofisher. Power output settings for the AbP-2 were adapted from Weisser and Klar (1990). Initially, the electrofisher delivered three DC pulses per second at 25% duty cycle, 125 V, with a 3:1 burst pulse train (i.e., three pulses on, one pulse off). This current is designed to stimulate burrowed larvae to enter the water column. Once a larva was observed in the water column, 30 pulses/second were occasionally applied to temporarily immobilize the larva for capture in a net. We spent relatively more time (approximately 30 seconds/m^2) within each reach electrofishing areas of preferred larval lamprey rearing habitat where depositional silt and sand substrates were dominant (henceforth Type I habitat, Slade et al. 2003). Relatively less time (approximately 5 seconds/m^2) was spent electrofishing areas with relatively large gravel, hard bedrock and boulder substrates (henceforth Type II and type III habitats, Slade et al. 2003). All larval lamprey observed were captured and placed in buckets containing stream water.

Quadrat Sampling in Deep Water Habitats

For deep water habitats, each sampling event consisted of a single drop with deepwater electrofishing equipment within the 30 x 30 m quadrat (Bergstedt and Genovese 1994; Jolley et al. 2012). Quadrats were accessed and sampled by boat, using quadrat center point in Universal Transverse Mercator (UTM) coordinates for navigation. When quadrats could not be sampled due, for example, to dewatered conditions, depth less than 0.3 m, excessive velocity, or excessive depth (>21 m) they were eliminated and subsequent quadrats were increased in priority (Table 1). The deepwater electrofisher was comprised of a modified AbP-2 electrofisher (ETS Engineering, Madison, WI) which delivered electrical stimulus to river bottom substrates at electrodes mounted to a fiberglass bell (or hood; 0.61 m^2 in area). The electrofisher delivered three pulses DC per second at 10% duty cycle, with a 2:2 pulse train (i.e., two pulses on, two pulses off). Output voltage was adjusted at each quadrat to maintain a peak voltage gradient between 0.6 and 0.8 V/cm across the electrodes. The electrofisher bell was coupled by a 76 mm vinyl suction hose to a gasoline-fueled hydraulic pump. The hydraulic pump was started approximately 5 seconds prior to shocking to purge air from the suction hose. Suction was produced by directing flow from the pump through a hydraulic eductor, which allows larvae to be collected in a mesh basket (27 x 62 x 25 cm; 2 mm wire mesh) while preventing them from passing through the pump. A 60 second pulse delivery was followed by an additional 60 seconds of pumping to further allow displaced larvae to cycle through the hose and into the collection basket. The sampling techniques are described in detail by Bergstedt and Genovese (1994) and were similar to those used in the Great Lakes region (Fodale et al. 2003) and the Willamette River (Jolley et al. 2012).

Biological Data Collection

Collected lamprey were anesthetized in a solution of buffered tricaine methanesulfonate (MS-222), measured for total length (TL in mm; total weight was not measured), classified according

to developmental stage (i.e., larvae, juvenile, or adult), and when possible (i.e., when larvae > 60 mm TL; Goodman et al. 2009) identified to genus (i.e., *Entosphenus* [Pacific Lamprey] or *Lampetra* [Western Brook or River Lamprey]) according to visual evaluations of caudal fin pigmentation patterns. Caudal fin tissue samples were also collected from select larvae for potential future assignment of genus genetically (Spice et al. 2011; Docker et al. 2016). Tissue samples are archived at the Columbia River Fish & Wildlife Conservation Office (CRFWCO) pending funding availability for genetic identification. Upon resuming active swimming behavior, larvae were released near the area of capture. Physical anomalies (lesions, suspected bird strikes, tumors, etc.) were recorded for all larvae. If abnormalities were observed on a larva, the individual was euthanized and preserved for potential evaluation at a later date. In addition, observations of juveniles, adults, or suspected Pacific Lamprey nests were also recorded.

Habitat Data Collection

Tributary

Within each sample reach, water temperature (°C) and conductivity (µS/cm) were measured, and visibility was qualitatively ranked as good, fair, or poor. The proportion (%) of Type 1 burrowing substrate within each reach was estimated. In general, larval lamprey habitats are classified as Type I, II, or III, and it is widely accepted that larvae appear to prefer Type I habitat the most and Type III the least (see Slade et al. 2003). Non-sediment habitat variables are presented as mean (± SE) unless otherwise noted. .

Confluence and Shoreline

Concurrent to each sampling event a sediment sample was taken (when possible) from each quadrat with a Ponar bottom sampler (16.5 cm x 16.5 cm). Each sample was mixed thoroughly and approximately two, 250-500 ml subsamples were transferred to containers provided by a contracted laboratory. Samples were labeled with the site number, replicate number and date, placed on ice, returned to the USFWS office, and subsequently handled per the instructions provided from the contracted laboratory. Sediment samples collected at each confluence quadrat were transferred to ALS Environmental Laboratory (Kelso, WA) for quantification of parameters such as grain size, grain type, and organic content. See Appendix 1 for information on sediment analyses. Water depth was measured at each quadrat are presented as mean (± SE) unless otherwise noted. Beginning in 2018, water temperature (°C) and conductivity (μS/cm) were recorded at each site on the sample day (in previous years, these data were recorded at each quadrat).

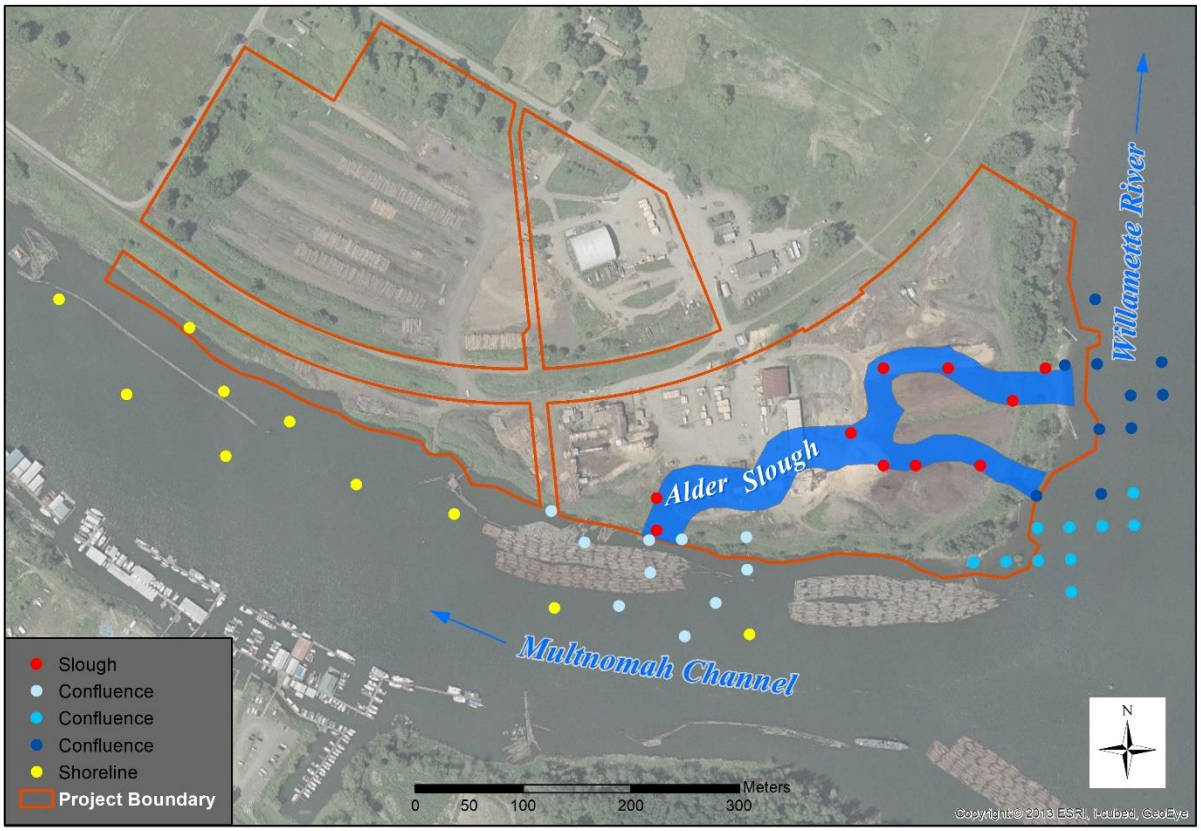

Figure 3. Alder Point restoration site sample sites. Habitats within the sites are confluence quadrats (blue points; each point represent a quadrat center point), shoreline quadrats (yellow points), and slough quadrats (red points).

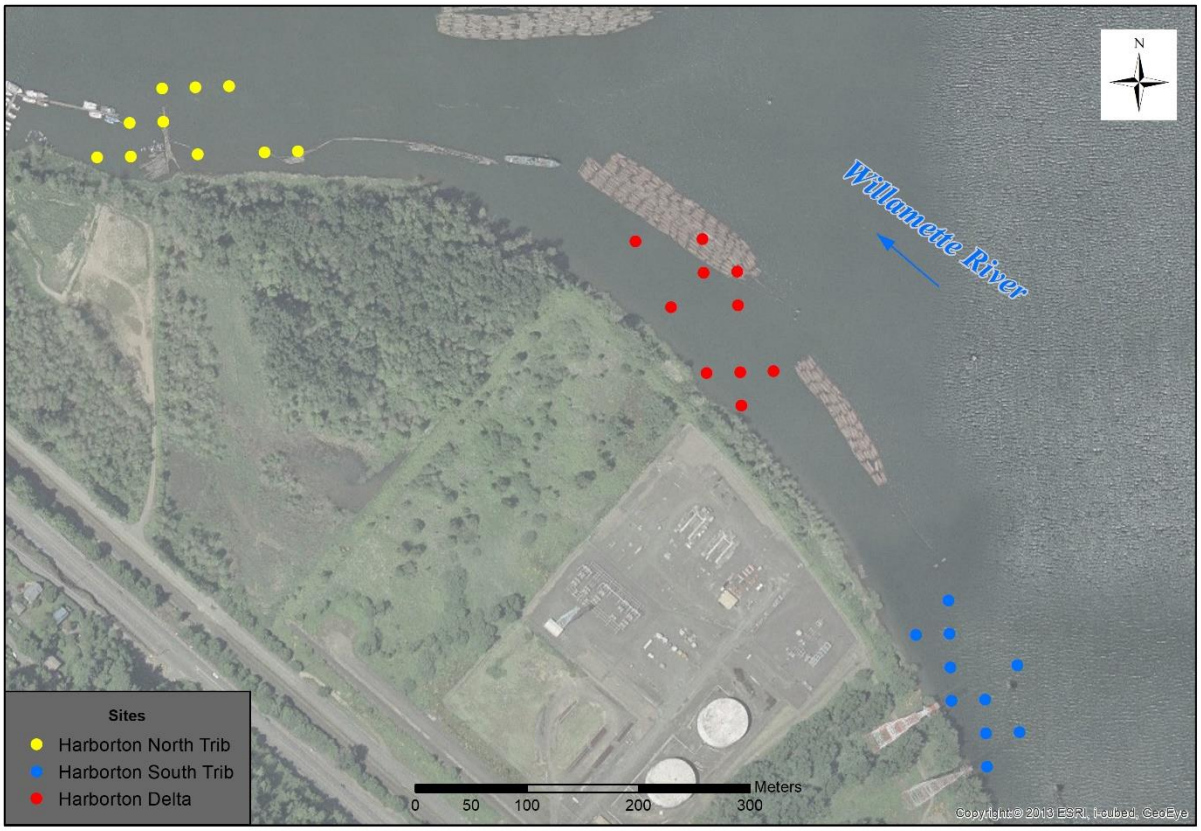

Figure 4. Harborton restoration sample sites with North Tributary quadrats (yellow points), South Tributary quadrats (blue points), and confluence quadrats (red points).

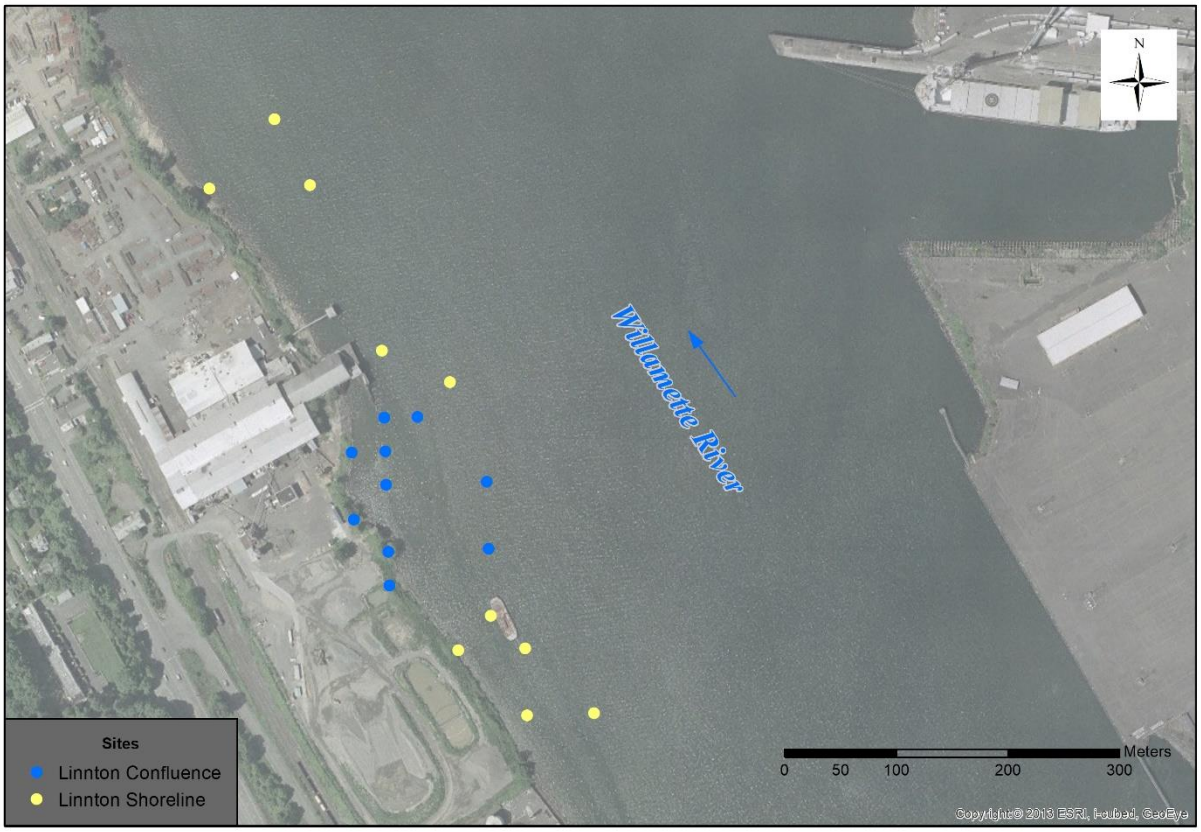

Figure 5. Linnton restoration site with shoreline quadrats (yellow points) and confluence quadrats (blue points).

Figure 6. Triangle Park restoration site with shoreline quadrats (yellow points).

Figure 7. Rinearson Natural Area restoration site with confluence quadrats (blue points) and tributary sample reaches (red points).

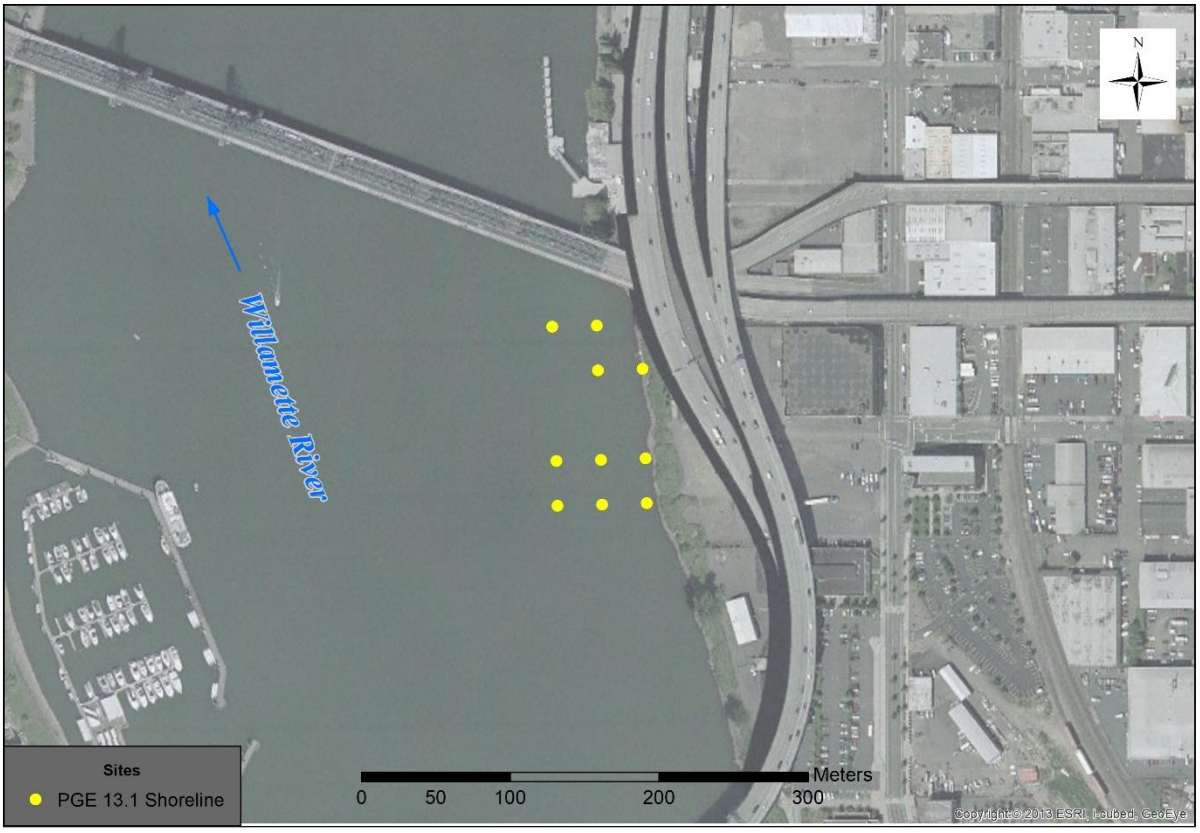

Figure 8. PGE 13.1 restoration site with shoreline quadrats (yellow points).

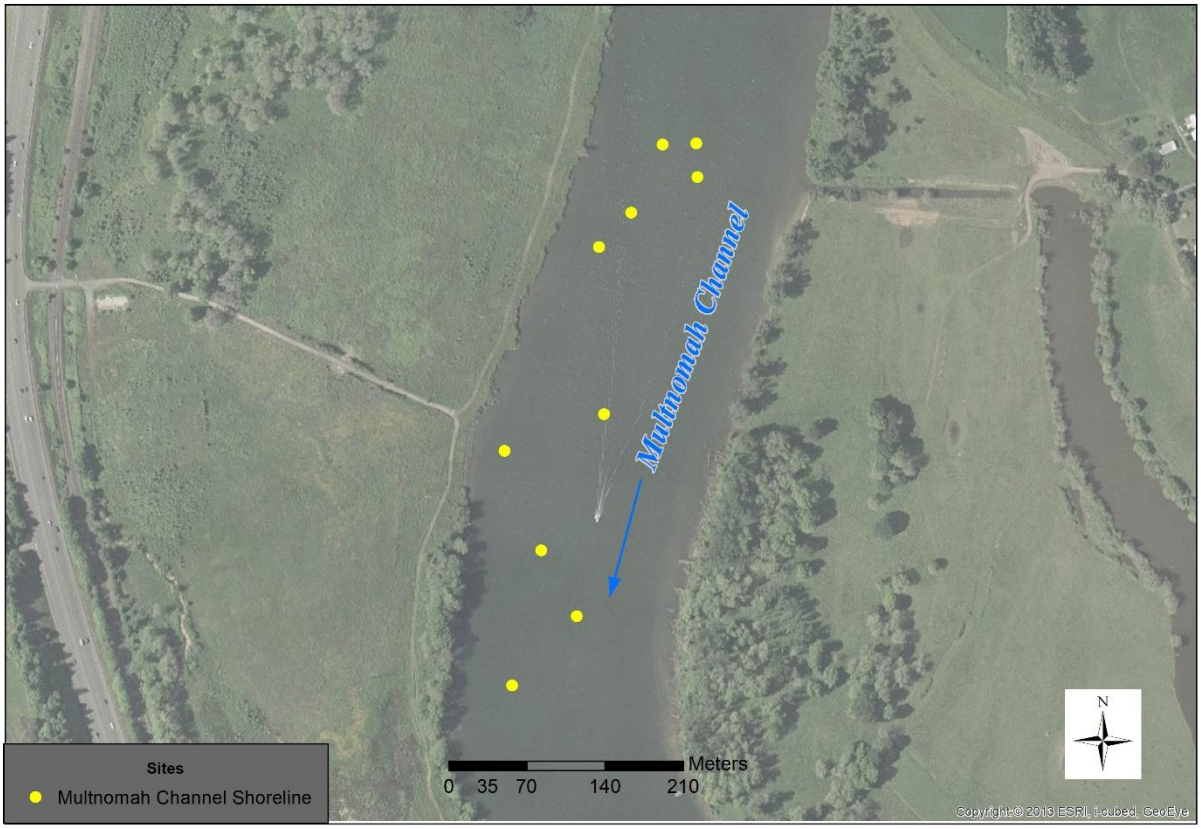

Figure 9. Multnomah Channel reference site with shoreline quadrats (yellow points).

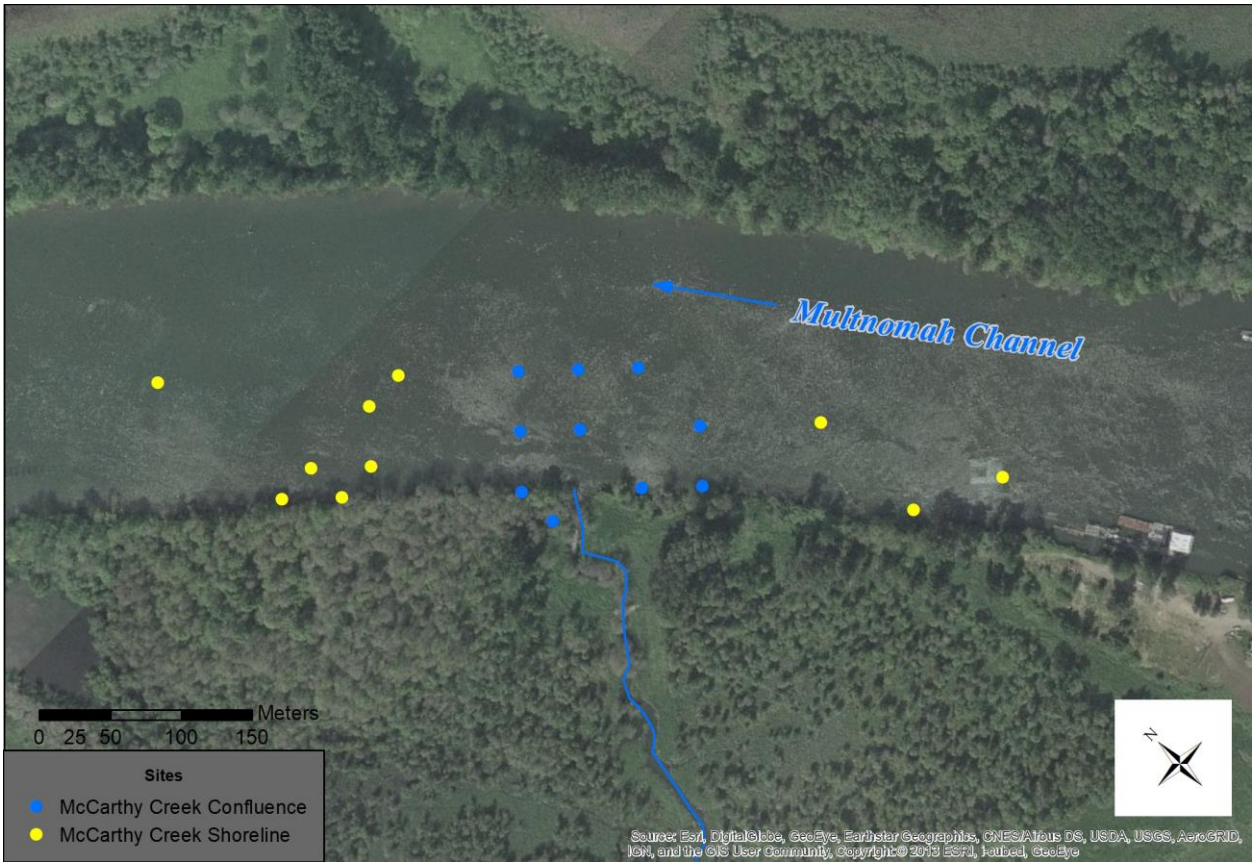

Figure 10. McCarthy Creek reference site with confluence quadrats (blue points) and shoreline quadrats (yellow points).

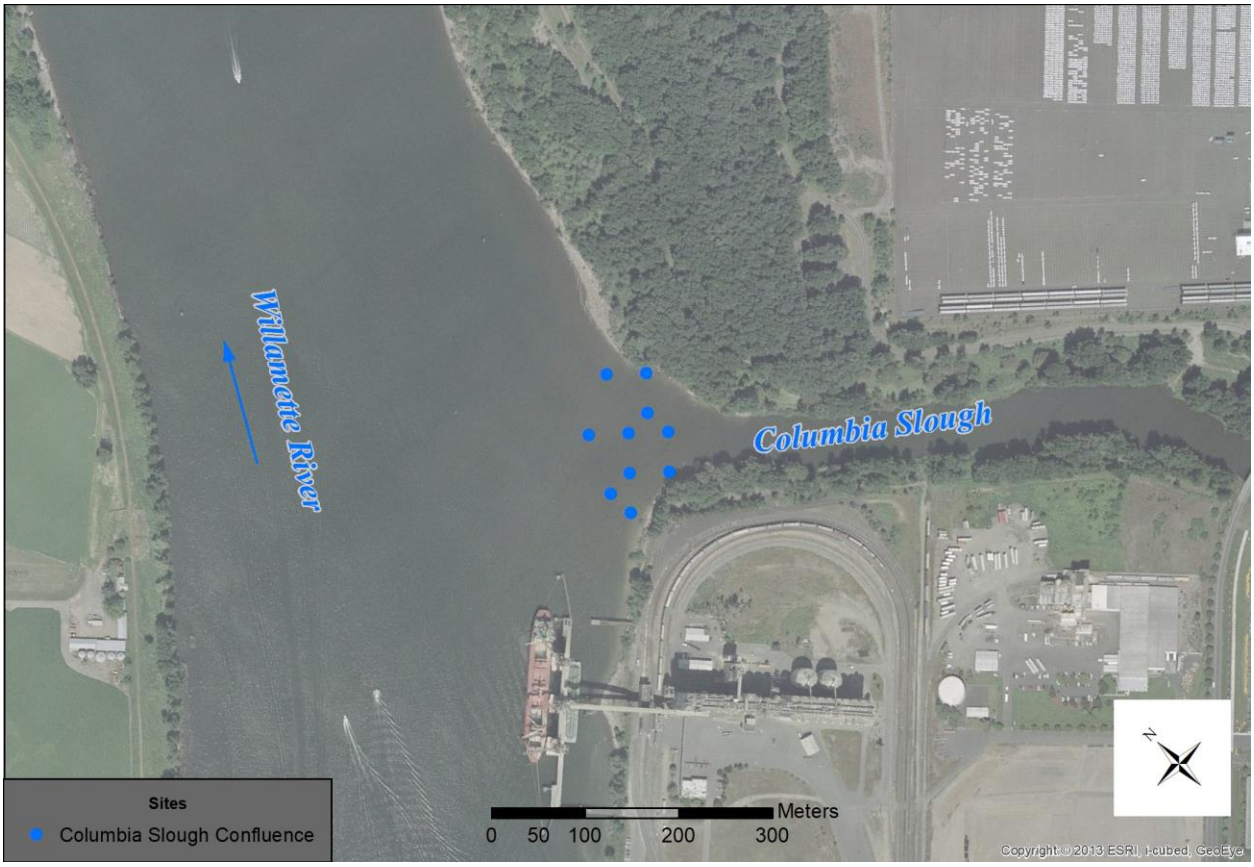

Figure 11. Columbia Slough reference site confluence quadrats (blue points).

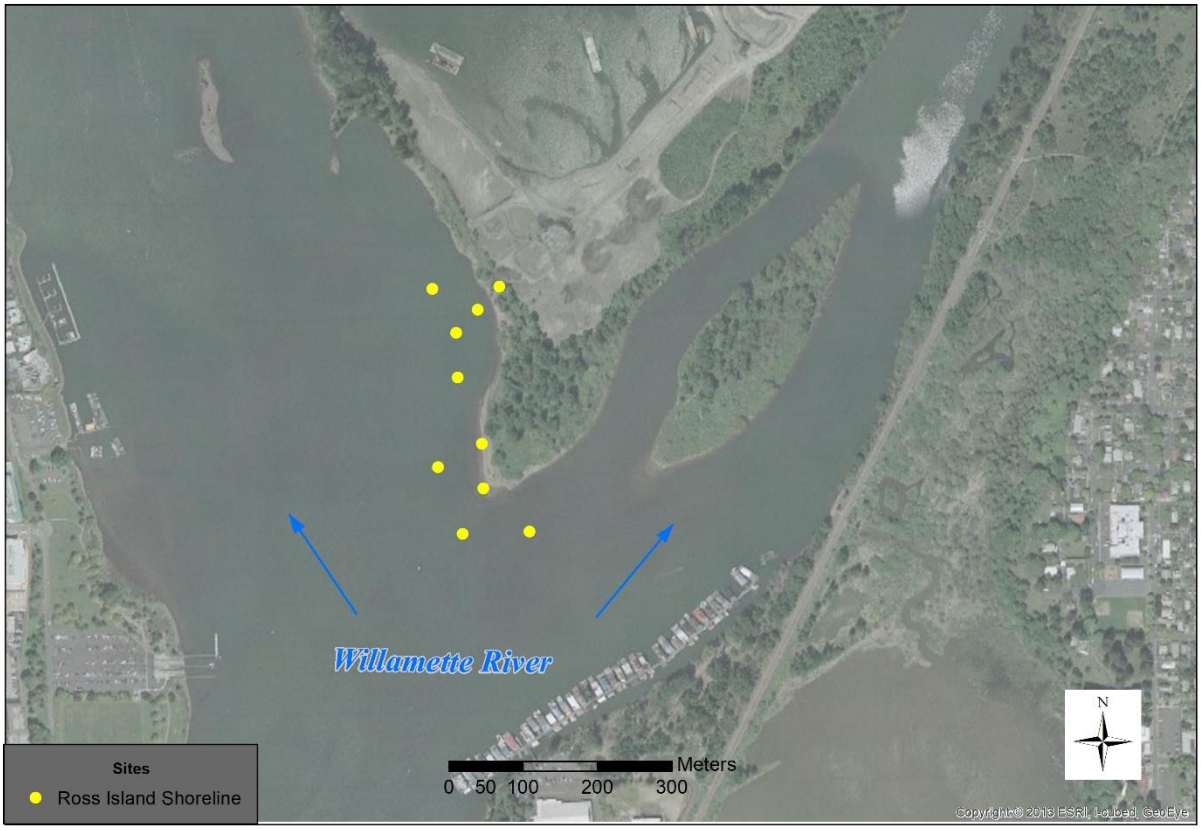

Figure 12. Ross Island reference site shoreline quadrats (yellow points).

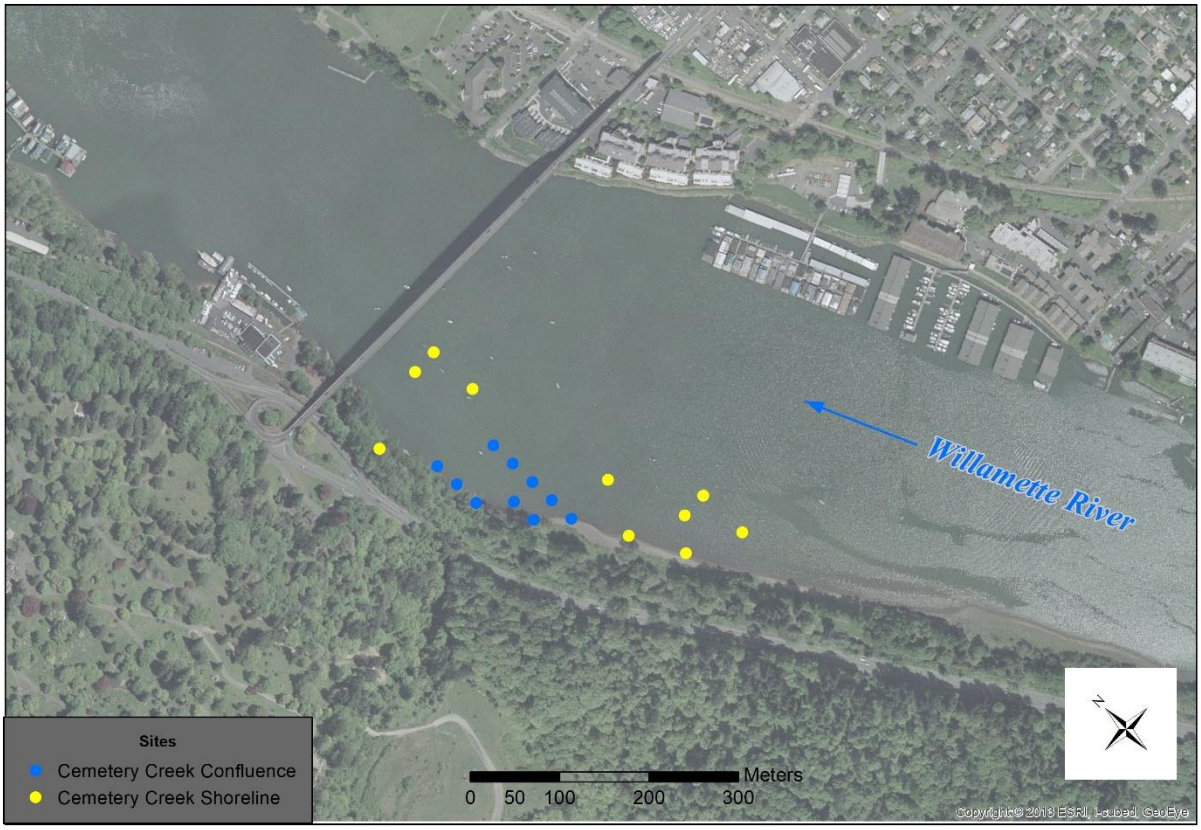

Figure 13. Cemetery Creek reference site with confluence (blue points) and shoreline (yellow points).

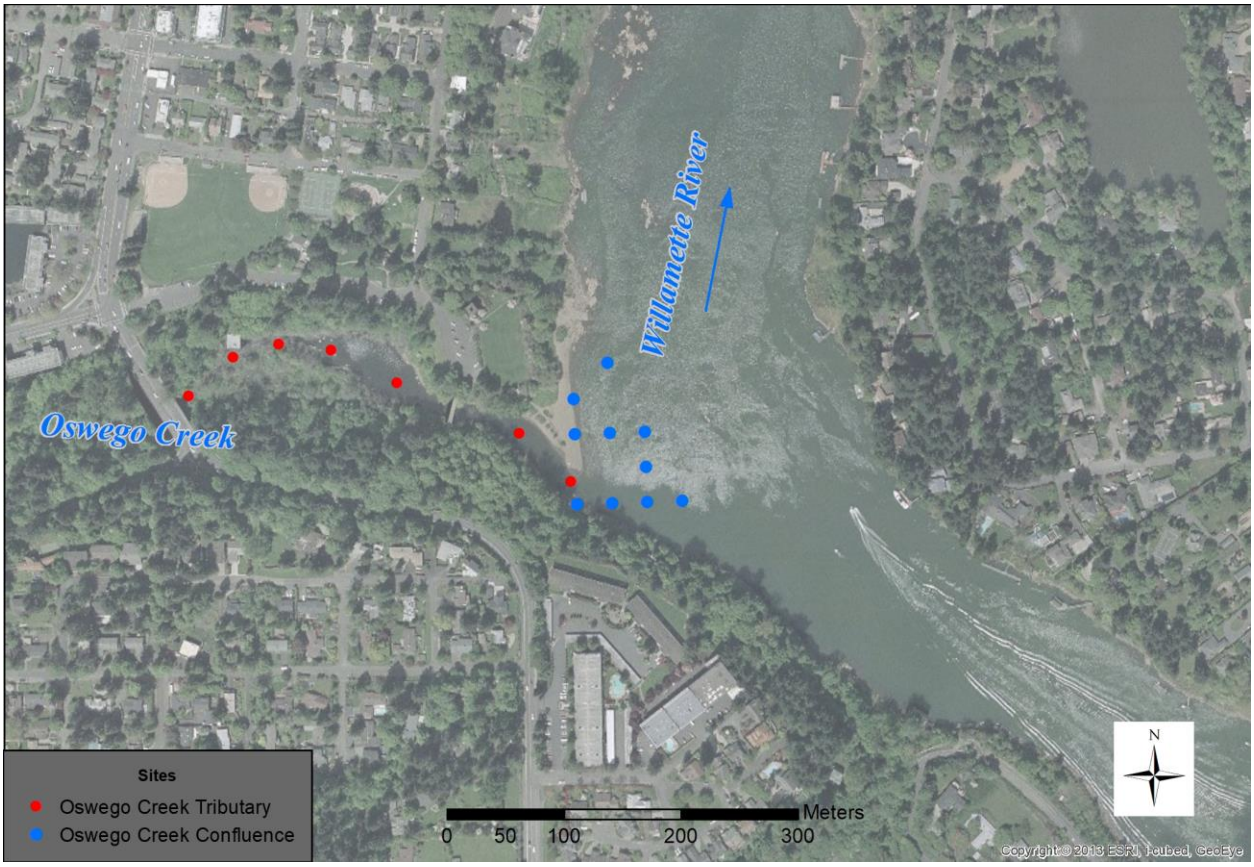

Figure 14. Oswego Creek reference site confluence quadrats (blue points) and tributary sample reaches (red).

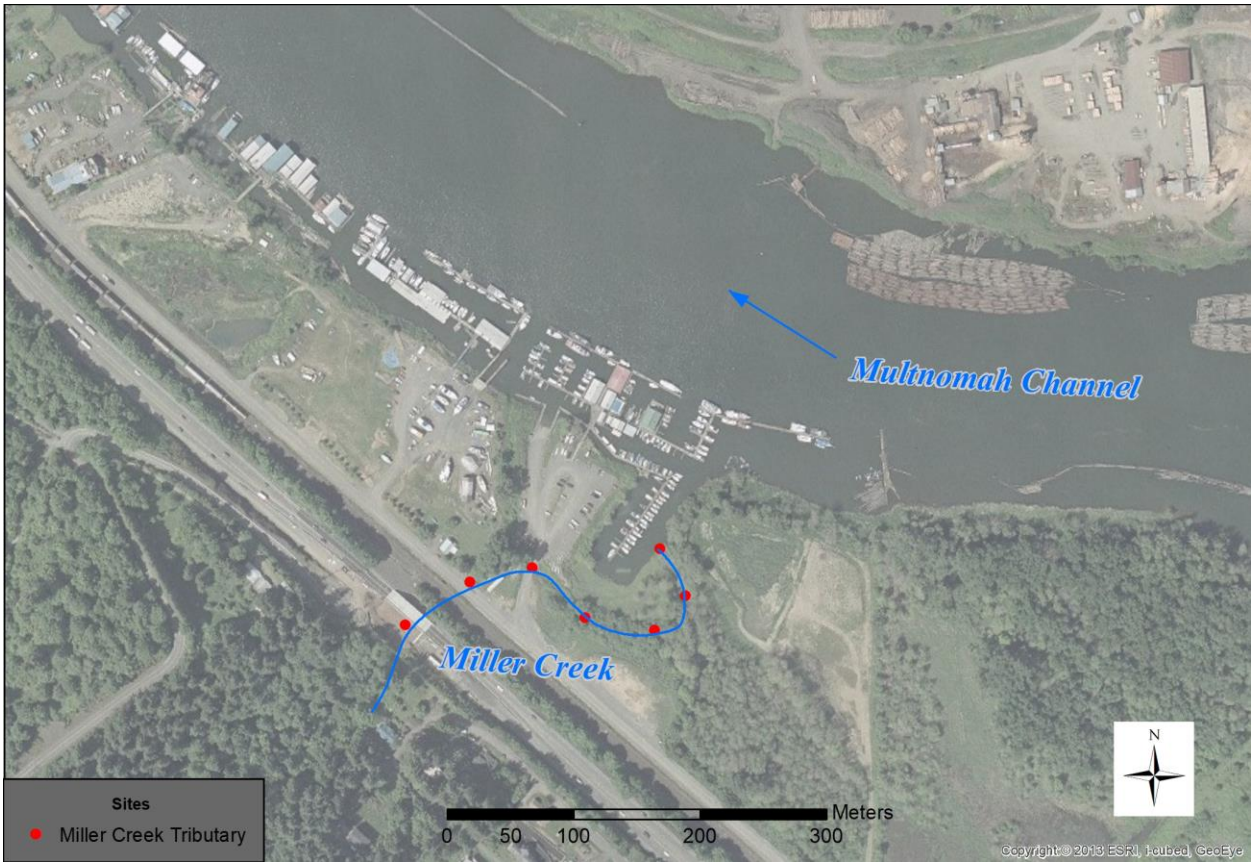

Figure 15. Miller Creek tributary sample reaches (red points).

Results

All lamprey collected at restoration sites were of the larval life stage. No detections of juveniles or evidence of adults (e.g., live adults, spawning nests) occurred. All larvae collected appeared healthy based on visual observation of external features, no abnormalities or indications of disease or poor health were observed. Lamprey identification and length measurements are summarized in Table 1 and site-specific depth, temperature, and conductivity are summarized in Table 2**.**

Restoration Sites

Alder Point Restoration Site

Larval lamprey (n = 2) were detected in 2 of 30 confluence quadrats (*d* = 0.07; Table 1). No lamprey were detected in the 10 shoreline quadrats or 10 quadrats in the newly constructed Alder Slough. The maximum number of larvae occupying any individual quadrat was one. Species composition included unidentifiable (less than 60 mm TL) lamprey (n = 1, TL = 21 mm) and *Lampetra spp.* (n = 1, TL = 81 mm). Sample depths ranged from 0.3 m to 12.9 m. Larvae were detected at depths of 0.3 m and 1.8 m (Table 2). Water temperature was 22.1°C and 24.1°C on 27 Aug 2019 and 29 Aug 2019, respectively. Conductivity ranged from 108.5-122.8 μS/cm across the five defined sites on 27-29 Aug 2019. Sediment samples collected at confluence quadrats were transferred to ALS Environmental Laboratory (Kelso, WA) in October 2019 for quantification of parameters such as grain size, grain type, and organic content. See Appendix 1 for information on sediment analyses.

Harborton Restoration Site

This site was not sampled in 2019.

Linnton Restoration Site

This site was not sampled in 2019.

Triangle Park Restoration Site

This site was not sampled in 2019.

Rinearson Natural Area

Larval lamprey (n = 3) were detected in 3 of 10 confluence quadrats (*d* = 0.30; Table 1). No lamprey were detected in the 4 of 6 tributary quadrats available to sample. The maximum number of larvae occupying any individual quadrat was one. Species composition included Pacific Lamprey (n = 1, TL = 76 mm) and unidentifiable (less than 60 mm TL) lamprey (n = 3, TL = 29-55 mm). Sample depths ranged from 0.4 m to 5.0 m. Larvae were detected at depths from 0.4 to 0.6 m (Table 2). Water temperature was 19.9°C on 11 Sept 2019 and conductivity

was 76 μS/cm.

PGE 13.1 Restoration Site

Larval lamprey were not detected in 10 confluence quadrats (d = 0.00; Table 1). Sample depths ranged from 0.7 m to 6.5 m. (Table 2). Water temperature was 20.5°C on 13 Sept 2019 and conductivity was 66.6 μS/cm.

Reference Sites

All lamprey collected at reference sites were of the larval life stage. No detections of juveniles or evidence of adults (e.g., live adults, spawning nests) occurred. All larvae collected appeared healthy based on visual observation of external features, no abnormalities or indications of disease or poor health were observed.

Multnomah Channel (Shoreline)

No lamprey were detected in any of the 10 quadrats sampled at Multnomah Channel (Table 1). Sample depths ranged from 4.1 to 9.8 m (Table 2). Water temperature was 23.0°C and conductivity was 148.2 μS/cm. Sampling occurred on 20 Sep 2019.

McCarthy Creek (Confluence and Shoreline)

Larval lamprey (n = 1) were detected at 1 of 10 shoreline quadrats sampled (d = 0.10) and no lamprey were detected in confluence quadrats (d = 0.00) (Table 1). Species were comprised of unidentified lamprey (n = 1, TL = 18 mm). Sample depths in the confluence and shoreline ranged from 1.8 to 9.4 m. Larvae were detected at a depth 7.1 m (Table 2). Water temperature was 21.9°C and conductivity was 134.2 μS/cm. Sampling occurred on 22 Sep 2019. In 2019, sampling of the tributary habitat in McCarthy Creek was discontinued due to flows and unwadeable conditions.

Columbia Slough (Confluence)

No lamprey were detected at the 10 confluence quadrats sampled (Table 1). Sample depths ranged from 0.3 to 3.6 m (Table 2). Water temperature was 22.1°C and conductivity was 141.4 μS/cm. Sampling occurred on 20 Aug 2019.

Ross Island (Shoreline)

No lamprey were detected at the 10 confluence quadrats sampled (Table 1). Sample depths ranged from 0.3 to 6.6 m (Table 2). Water temperature was 22.6°C and conductivity was 85.8 μS/cm. Sampling occurred on 28 Aug 2019.

Cemetery Creek (Confluence and Shoreline)

Larval lamprey (n = 12) were detected at 2 of 10 confluence quadrats sampled (d = 0.20), and 3

of 10 shoreline quadrats (*d* = 0.30; Table 1, Figure 14). The maximum number of larvae occupying any individual quadrat was three. Of the nine larvae collected at Cemetery Creek, all were too small to identify visually (TL = 15-44 mm). Sample depths ranged from 0.3 to 7.3 m. Larvae were detected at depths from 2.4 to 7.1 m (Table 2). Water temperature was 20.1°C and conductivity was 79.8 μS/cm. Sampling occurred on 26 Aug 2019. In 2019, sampling of the tributary habitat in Cemetery Creek was discontinued due to lack of Type I habitat and a putative passage barrier approximately 100 m upstream of the mouth.

Oswego Creek (Confluence and Tributary)

Larval lamprey (n = 13) were detected at 5 of 10 confluence quadrats sampled (*d* = 0.50; Table 1). The maximum number of larvae occupying any individual quadrat was five. All lamprey were relatively small (TL = 14 - 36 mm; Figure 12) and could not be identified to species. Sample depths ranged from 0.4 to 10.4 m. Larvae were detected at depths 0.7 m to 5.8 m (Table 2). Water temperature was 20.1°C and conductivity was 70.2 μS/cm on 26 Aug 2019. Larval lamprey (n = 1) were detected at 1 of 7 tributary reaches (*d* = 0.14; Table 1). The single lamprey sampled could not be identified to species (TL = 46 mm). Sample depths ranged from 0.2 - 1.1 m. Larvae were detected at depths 1.1 m (Table 2). Water temperature was 21.6°C and conductivity was 160.5 μS/cm. For the sites assessed, Type I habitat ranged from 15 to 95%. The single lamprey sampled was detected in a reach that was at 95% Type 1 habitat. Since 2019 was the first year we sampled in Oswego Creek, sediment samples were collected and transferred to ALS Environmental Laboratory (Kelso, WA) for quantification of parameters such as grain size, grain type, and organic content. See Appendix 1 for information on sediment analyses. Sampling occurred on 23 Sept 2019.

Miller Creek (Tributary)

Larval lamprey (n = 8) were detected at 5 of 7 tributary reaches sampled (*d* = 0.71; Table 1). The maximum number of larvae occupying any individual reach was five. Species composition included Western Brook Lamprey (n = 4, TL = 74-180 mm) and unidentifiable (less than 60 mm TL) lamprey (n = 4, TL = 49-59 mm). Sample depths ranged from 0.05 to 0.9 m. Larvae were detected at depths 0.1 m to 0.9 m (Table 2). Water temperature was 15.4°C and conductivity was 173.7 μS/cm. Type I habitat ranged from 5 to 50%. Lamprey were found in reaches containing 10 to 50% Type I habitat. Since 2019 was the first year we sampled in Miller Creek, sediment samples were collected and transferred to ALS Environmental Laboratory (Kelso, WA) for quantification of parameters such as grain size, grain type, and organic content. See Appendix 1 for information on sediment analyses. Sampling occurred on 13 Sep 2019.

Table 1. In 2019, total number of quadrats visited, sampled, occupied by larval lamprey, and corresponding larval lamprey detection probability (*d*). Small (i.e., less than 60 mm TL) larvae cannot be accurately identified and are classified as unidentified (UNID).

	Site		Quadrats				Pacific Lamprey		*Lampetra spp.*		UNID		
			Visited	Sampled	Occupied	*d*	*N*	TL (mm) Range	*N*	TL (mm)	*N*	TL (mm) Range	Total *N*
Restoration	Alder Point	Confluence	30	30	2	0.07	0	-	1	81	1	21	2
		Shoreline	10	10	0	0.00	0	-	0	-	0	-	0
		Slough	10	10	0	0.00	0	-	0	-	0	-	0
	13.1	Shoreline	10	10	0	0.00	0	-	0	-	0	-	0
	Rinearson	Confluence	10	10	3	0.30	1	76	0	-	3	29-55	4
		Tributary	6	4	0	0.00	0	-	0	-	0	-	0
Reference	Multnomah Channel	Shoreline	10	10	0	0.00	0	-	0	-	0	-	0
	McCarthy Creek	Confluence	10	10	0	0.00	0	-	0	-	0	-	0
		Shoreline	10	10	1	0.10	0	-	0	-	1	18	1
	Columbia Slough	Confluence	10	10	0	0.00	0	-	0	-	0	-	0
	Ross Island	Shoreline	10	10	0	0.00	0	-	0	-	0	-	0
	Cemetery Creek	Confluence	10	10	2	0.20	0	-	0	-	4	19-44	4
		Shoreline	10	10	3	0.30	0	-	0	-	5	15-29	5
	Oswego Creek	Confluence	10	10	5	0.50	0	-	0	-	13	14-36	13
	Oswego Creek	Tributary	7	7	1	0.14	0	-	0	-	1	46	1
	Miller Creek	Tributary	7	7	5	0.71	0	-	4	74-180	4	49-59	8

Table 2. In 2019, habitat variables measured at restoration and reference sites. Capture depth range is the minimum and maximum depths at which lamprey were captured. Sediment collection was transferred to ALS Environmental Laboratory (Kelso, WA).

Site Type	Site	2019 Date Sampled	Capture Depth (m), range	Sample Depth (m), range	Temperature (°C)	Conductivity (µS/cm)	Sediment Collected?
Restoration	Alder	27-29 Aug	0.3-1.8	0.3-12.9	22.1 - 24.1	108.5 - 122.8	Y
	13.1	13-Sep	-	0.7-6.5	20.5	66.6	N
	Rinearson	11-Sep	0.4-0.6	0.4-5.0	19.9	76.0	Y
	Rinearson Tributary	2-Sep	-	0.1-0.2	17.1	140.3	Y
Reference	Multnomah Channel	20-Sep	-	4.1-9.8	23.0	148.2	N
	McCarthy Creek	22-Sep	7.1	1.8-9.4	21.9	134.2	N
	Columbia Slough	30-Aug	-	0.3-3.6	22.1	141.4	Y
	Ross Island	28-Aug	-	0.3-6.6	22.6	85.8	Y
	Cemetery Creek	26-Aug	2.4-7.1	0.3-7.3	20.1	79.8	N
	Oswego Creek	26-Aug	0.7-5.8	0.4-10.4	20.1	70.2	Y
	Oswego Creek Tributary	13-Sep	1.1	0.2-1.1	21.6	160.5	Y
	Miller Creek Tributary	13-Sep	0.1-0.9	0.05-0.90	15.4	173.7	Y

Table 3. Occupancy results from sampling at restoration and reference sites across all sampling years (pre and post restoration actions). Total number of quadrats visited, sampled, occupied by larval lamprey, and corresponding larval lamprey detection probability (*d*). Small (i.e., less than 60 mm TL) larvae cannot be accurately identified and are classified as unidentified (UNID).

Site Type	Site Status	Sample Year	Site	Sample Area	Quadrats Visited	Quadrats Sampled	Quadrats Occupied	Quadrats d	Pacific Lamprey	*Lampetra* spp.	UNID	Total *N*
Restoration	Pre Yr 1	2014	Alder Point	Shoreline	30	29	2	0.07	0	3	0	3
		2015	Rinearson	Confluence	13	10	3	0.30	3	0	3	6
				Tributary		7	1	0.14	3	0	0	3
	Post Yr 1	2016	Alder Point	Confluence	30	30	1	0.03	0	0	1	1
				Shoreline	10	10	1	0.10	0	1	0	1
				Slough	10	10	0	0.00	0	0	0	0
	Post Yr 2	2017	Alder Point	Confluence	30	30	1	0.03	0	0	1	1
				Shoreline	10	10	2	0.20	1	0	1	2
				Slough	10	10	0	0.00	0	0	0	0
	Pre Yr 1		Harborton	Confluence	10	10	1	0.10	0	0	2	2
				North Trib.	10	10	1	0.10	0	1	0	1
				South Trib.	10	10	0	0.00	0	0	0	0
			Linnton	Confluence	10	10	0	0.00	0	0	0	0
				Shoreline	10	10	0	0.00	0	0	0	0
			Triangle Park	Shoreline	21	21	0	0.00	0	0	0	0
			PGE RM 13.1	Shoreline	10	10	0	0.00	0	0	0	0
	Post Yr 3	2018	Alder Point	Confluence	30	30	1	0.03	0	0	1	1
				Shoreline	10	10	0	0.00	0	0	0	0
				Slough	10	10	0	0.00	0	0	0	0
	Post Yr 4	2019	Alder Point	Confluence	30	30	2	0.07	0	1	1	2
				Shoreline	10	10	0	0.00	0	0	0	0
				Slough	10	10	0	0.00	0	0	0	0

Post Yr 2		13.1	Shoreline	10	10	0	0.00	0	0	0	0
Post Yr 1		Rinearson	Confluence	10	10	3	0.30	1	0	3	4
			Tributary	6	4	0	0.00	0	0	0	0
Reference	2014	Ross Island	Shoreline	28	26	5	0.19	0	6	0	6
	2015	Cemetery Creek	Confluence	10	10	5	0.50	2	0	6	8
			Tributary	2	2	0	0.00	0	0	0	0
	2016	Multnomah Channel	Shoreline	10	10	1	0.10	0	0	1	1
		McCarthy Creek	Confluence	10	10	0	0.00	0	0	0	0
			Shoreline	10	10	1	0.10	0	0	1	1
			Tributary	7	2	0	0.00	0	0	0	0
		Columbia Slough	Confluence	10	10	0	0.00	0	0	0	0
		Ross Island	Shoreline	10	10	2	0.20	2	0	0	2
		Cemetery Creek	Confluence	13	10	2	0.20	0	0	2	2
			Shoreline	10	10	3	0.30	1	0	2	3
		Oswego Creek	Confluence	10	10	4	0.40	2	1	3	6
	2017	Multnomah Channel	Shoreline	10	10	0	0.00	0	0	0	0
		McCarthy Creek	Confluence	10	10	1	0.10	1	1	0	2
			Shoreline	10	10	0	0.00	0	0	0	0
			Tributary	7	2	0	0.00	0	0	0	0
		Columbia Slough	Confluence	11	10	1	0.10	1	0	0	1
		Ross Island	Shoreline	13	10	3	0.30	5	0	1	6
		Cemetery Creek	Confluence	10	10	6	0.60	3	0	7	10
			Shoreline	10	10	4	0.40	3	0	3	6
		Oswego Creek	Confluence	10	10	5	0.50	3	0	6	9
	2018	Multnomah Channel	Shoreline	10	10	0	0.00	0	0	0	0
		McCarthy Creek	Confluence	10	10	2	0.20	1	0	1	2
			Shoreline	10	10	0	0.00	0	0	0	0
		Columbia Slough	Confluence	10	10	0	0.00	0	0	0	0
		Ross Island	Shoreline	10	10	3	0.30	3	2	1	6

	Cemetery Creek	Confluence	10	10	5	0.50	4	0	4	8
		Shoreline	10	10	3	0.30	2	0	4	6
	Oswego Creek	Confluence	10	10	2	0.20	2	0	2	4
2019	Multnomah Channel	Shoreline	10	10	0	0.00	0	0	0	0
	McCarthy Creek	Confluence	10	10	0	0.00	0	0	0	0
		Shoreline	10	10	1	0.10	0	0	1	1
	Columbia Slough	Confluence	10	10	0	0.00	0	0	0	0
	Ross Island	Shoreline	10	10	0	0.00	0	2	0	0
	Cemetery Creek	Confluence	10	10	2	0.20	0	0	4	4
		Shoreline	10	10	3	0.30	0	0	5	5
	Oswego Creek	Confluence	10	10	5	0.50	0	0	13	13
	Oswego Creek	Tributary	7	7	1	0.14	0	0	1	1
	Miller Creek	Tributary	7	7	5	0.71	0	4	4	8

Table 3. Continued.

Site Type	Site Status	Sample Year	Site	Sample Area	Quadrats Visited	Quadrats Sampled	Quadrats Occupied	*d*	Pacific Lamprey	*Lampetra* spp.	UNID	Total *N*
Reference		2014	Ross Island	Shoreline	28	26	5	0.19	0	6	0	6
		2015	Cemetery Creek	Confluence	10	10	5	0.50	2	0	6	8
				Tributary	2	2	0	0.00	0	0	0	0
		2016	Multnomah Channel	Shoreline	10	10	1	0.10	0	0	1	1
			McCarthy Creek	Confluence	10	10	0	0.00	0	0	0	0
				Shoreline	10	10	1	0.10	0	0	1	1
				Tributary	7	2	0	0.00	0	0	0	0
			Columbia Slough	Confluence	10	10	0	0.00	0	0	0	0
			Ross Island	Shoreline	10	10	2	0.20	2	0	0	2
			Cemetery Creek	Confluence	13	10	2	0.20	0	0	2	2
				Shoreline	10	10	3	0.30	1	0	2	3
			Oswego Creek	Confluence	10	10	4	0.40	2	1	3	6
		2017	Multnomah Channel	Shoreline	10	10	0	0.00	0	0	0	0
			McCarthy Creek	Confluence	10	10	1	0.10	1	1	0	2
				Shoreline	10	10	0	0.00	0	0	0	0
				Tributary	7	2	0	0.00	0	0	0	0
			Columbia Slough	Confluence	11	10	1	0.10	1	0	0	1
			Ross Island	Shoreline	13	10	3	0.30	5	0	1	6
			Cemetery Creek	Confluence	10	10	6	0.60	3	0	7	10
				Shoreline	10	10	4	0.40	3	0	3	6
			Oswego Creek	Confluence	10	10	5	0.50	3	0	6	9

Table 3. Continued.

Site Type	Site Status	Sample Year	Site	Sample Area	Quadrats Visited	Quadrats Sampled	Quadrats Occupied	Quadrats *d*	Pacific Lamprey	*Lampetra* spp.	UNID	Total *N*
Reference		2018	Multnomah Channel	Shoreline	10	10	0	0.00	0	0	0	0
			McCarthy Creek	Confluence	10	10	2	0.20	1	0	1	2
				Shoreline	10	10	0	0.00	0	0	0	0
			Columbia Slough	Confluence	10	10	0	0.00	0	0	0	0
			Ross Island	Shoreline	10	10	3	0.30	3	2	1	6
			Cemetery Creek	Confluence	10	10	5	0.50	4	0	4	8
				Shoreline	10	10	3	0.30	2	0	4	6
			Oswego Creek	Confluence	10	10	2	0.20	2	0	2	4

Findings

Restoration actions are complete at three sites, Alder Point, Rinearson and PGE 13.1. Monitoring at the Alder site can be directly compared to the reference site at Ross Island, which is sampled concurrently. Within site comparisons of pre- and post-restoration sampling show similarities in patterns of larval lamprey occupancy and rates of detection. To date, lamprey were found to occupy the confluence or shoreline habitats at both sites in most years of monitoring. In the newly constructed Alder Slough, no lamprey have been detected since the habitat was constructed. It is not known whether, and after how long, larval lamprey use of this newly-created habitat may occur. It remains unclear whether Alder Slough will provide useful habitat for lamprey. Thus, to date, lamprey appear to be using the post-restoration area of Alder Point in a manner similar to pre-restoration. Continued monitoring of larval lamprey occupancy at the Alder site and its paired reference site Ross Island is warranted and will provide a better understanding of larval lamprey colonization rates of newly available habitats. Monitoring at the Rinearson site can be directly compared to the reference site habitats at Cemetery and Oswego creeks, which are sampled concurrently, Larval lamprey have generally occupied confluence and shoreline habitat at both sites both pre and post restoration. Larval lamprey occupied tributary habitat within the RNA site pre restoration, but not immediately post restoration, as well as the tributary habitat at Oswego Creek post restoration. It is likely too soon after restoration to draw any conclusions about the long-term occupancy of the RNA tributary by lamprey.

Reference site monitoring is an important component of the lamprey monitoring program associated with the Portland Harbor Superfund project. Patterns of larval lamprey occupancy at reference sites will provide a baseline for evaluating changes in larval lamprey occupancy at restoration sites over time, and assessing the utility of restoration actions for larval lamprey. Monitoring at the reference sites is ongoing. After the 2017 sampling, we eliminated the wadeable portions of Cemetery Creek and McCarthy Creek from the group of reference sites (essentially, reference habitat was not available). In 2019, we assessed and added the Miller Creek tributary as a replacement for the McCarthy Creek tributary and added the Oswego Creek tributary as a replacement for Cemetery Creek tributary. Finding tributaries that are suitable as reference sites has proven to be a challenge.

Similar to the results of previous years' sampling (Jolley et al. 2015; Silver et al. 2016; Skalicky et al. 2018) we observed a combination of larval Pacific Lamprey and *Lampetra* spp. in the Portland Harbor Superfund area. Mainstem habitats associated with the Alder Point restoration, as well as habitats at many reference sites, continue to appear suitable to and available for colonization by larvae in the mainstem Willamette River and Multnomah channel. This was evidenced by the presence of larvae in shoreline and confluence habitats. The larvae detected at the Alder Point restoration site as well as at reference sites may have originated from tributaries that enter the Willamette River upstream of the study areas (for example, the Clackamas River subbasin) and gradually dispersed downstream to their location of capture. Given that the larvae detected included *Lampetra sp.*, and that brook lamprey would not be expected to spawn in relatively large and deep rivers, it is likely they originated from out of the area. Furthermore, evidence suggesting dispersal of larval lamprey out of tributaries and into mainstem habitats has been observed previously in the mainstem Columbia River and Willamette River basins (Jolley et al. 2012; Jolley et al. 2013; Jolley et al. 2014) and may occur over extensive distances

(Scribner and Jones 2002; Derosier et al. 2007). Pacific Lamprey spawning has not been documented in the Portland Harbor Superfund area. However, as observed in the lower mainstem of the Lewis River (J. Doyle, PacifiCorp, personal communication), Pacific Lamprey spawning in relatively large rivers may be plausible where suitable substrate and flow regimes occur. Thus, it is possible that the larval Pacific Lamprey detected at Alder point and reference sites originated from spawning within the Portland Harbor Superfund area.

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Appendix 1.

Results from sediment sampling have been provided to and can be obtained from:

Industrial Economics, Inc.
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