An Assessment of the Native Freshwater Mussels of the Middle Columbia Slough in Portland, Oregon

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Introduction

The purpose of this Masters of Environmental Management project is to examine the current health or status of the native freshwater mussel species (genus Anodonta) population in the Middle Columbia Slough, within the City of Portland. In order to accomplish this objective, it was necessary to establish a methodology that accurately collects data for location density and population characteristics. The data gathered in this project will be used by the City of Portland’s Bureau of Environmental Services (BES) as baseline characteristics for future studies, and as a possible metric with which to gauge the health of the Columbia Slough Watershed.

The Columbia Slough

The Columbia Slough is a watershed located in the north portion of Portland, Oregon (Figure 1). It parallels the Columbia River and in the past was linked through open channels to the river. There was flooding during high water events and some channels were refreshed by groundwater recharge during low water. During the early 1900’s landowners began draining the wet areas and cutting off seasonal flooding by creating levees. To manage drainage and water level, the Multnomah County Drainage District (MCDD) was created in 1917. With pumps, floodgates, and levees, the MCDD controls the water levels within the upper two thirds of the Slough. This portion is known
Figure 1 – The survey area
as the Upper and Middle reaches of the Slough are divided by a levee at 142nd Street in North Portland (Figure 2).

The Middle Columbia Slough has four named sloughs, Buffalo Slough (BS), Whitaker Slough (WS), Prison Pond (PP), and the Main Slough (MS) (Figure 1). The Main Slough is the longest slough in the complex of sloughs. It travels west to east, the entire length of the Middle Slough from NE 142nd St. to NE 14th St. in Portland. The other three sub sloughs or “southern arm sloughs” also drain from east to west. The westernmost slough is the Buffalo Slough, which starts at NE 42nd and travels to NE 26th going through a culvert underneath NE 33rd street. The next slough to the east is the longest of the southern sloughs and is known as the Whitaker Slough. This slough runs from NE 110th to NE 42nd travelling under I-205 in a culvert and provides an outlet for Johnson Lake and Whitaker Ponds. The easternmost sub slough in the Middle Slough is Prison Pond. It starts at NE 129th and ties into the Main Slough at NE 112th. These three southern sloughs were probably one channel historically, but have been bisected by bridges and fill and then turned to drain directly into the Main Slough. In the past 50 years they have been widened and occasionally dredged to create the slow water system that is in place today.

The Columbia Slough is the catchment for a watershed that is almost entirely urbanized. This results in contamination of sediment and water with the normal suite of toxics found in city environments - copper, lead, hydrocarbons, etc. There are also legacy contaminants left from agricultural and heavy industry activities within the watershed (ODEQ, BES, 2005). In the last 30 years, the Oregon Department of Environmental Quality (ODEQ) and BES has entered into an effort to locate and reduce the inflow of
Figure 2 – Localized map of the Middle Columbia Slough with sub-sloughs and sample sites.
toxics into the water body as well as identify and remediate areas of high concentrations of toxic sediment (BES, 2008). In some locations the contamination of sediment is at an intensity high enough to disrupt aquatic community assemblages through bioaccumulation and continuous exposure (ODEQ, BES, 2005).

With relatively long lives and sedentary life, freshwater mussels are well suited to serve as monitors of toxics at this interface between water and sediment. These characteristics provide an opportunity to collect data on entire watersheds in a spatially condensed location (Strayer and Smith, 2003). Accurately surveying freshwater mussels for density is the beginning step in using them as a biomonitor. These data can then be used to correlate positive and negative watershed conditions with mussel density and population trends (Bales and Foster, 1978; Balas and Nicklin, 2007). Given the historical pollution of the Columbia Slough and efforts to monitor and reduce toxics (BES, 2008), and a desire to account for biological communities within the city (Portland City Council, 2006), a regular count of freshwater mussels can provide data on watershed health and information on these little studied organisms.

Freshwater mussels

Freshwater mussels are at a nexus of several systems within a watershed. Their life cycle links them to fish, their flesh and waste support numerous aquatic and terrestrial species and they are found in the benthos and feed from the water column – exposing them to any toxics found in a system. West of the continental divide, freshwater mussels are not as well studied as their eastern counterparts (Nedeau,
Smith, and Stone, 2004). It is also clear that as a Family (Unionoid) they are in decline (Metcalf-Smith et al. 1998; Master et al. 2000; Lydeard et al. 2004).

A Unionid’s unique life cycle starts with a specialized larva, called a glochidium, which attaches to a specific host fish’s gills or fins for the completion of larval development. It then falls off and settles into the benthos, where it acquires a shell and continues to filter water for nutrients (McMahon and Bogan, 2001). This unique quality provides an ecological link to piscine issues within watersheds as a decline in mussel numbers may indicate a loss of host fishes (Nedeau, Smith, and Stone 2004).

Freshwater mussels are important members of the food web in waterways. They enrich the area surrounding them for other invertebrates by concentrating nutrients into waste pellets and creating microhabitats (Gutierrez et al. 2003, Spooner and Vaughn 2006). Freshwater mussels provide a nutrient rich food source for predators; the juveniles and glochidia are consumed by fish and other macro invertebrates, and the larger, thicker shelled adults are eaten by river otters, mink, raccoons, and muskrats (Nedeau, Smith, and Stone, 2004; Spooner, Vaughn, and Nichols, 2008).

**Anodonta**

There are seven recognized species of freshwater mussels found west of the Continental divide. Of these seven, one is of the Genus *Margaritifera*; one is of the Genus *Gonidea* and the other five of the Genus *Anodonta*. The three documented species found in the Columbia Slough are - *A. nuttalliana*—Winged Floater, *A. californiensis*—California Floater, and *A. oregonensis*—Oregon Floater (Smith pers.).
The identification of most Unionids involves the shape, number and orientation of pseudocardinal teeth, the small ridges of inner shell next to the hinge. Anodonta have no pseudocardinal teeth (*Ano* – without, *Donta* – teeth), and the determination of species was historically described based on morphological traits. The genesis of its common name stems from two habits. The mussel “floats” in the hyper saturated sediment at the bottom of the channel, and when these mussels die, the gasses from decomposition of the flesh make it buoyant, so that the mussel floats on the surface, shell and all. These floaters are known habitat generalists, found mainly in low energy hydrology and nutrient rich/fine sediment locations. They are short lived for mussels (~15y), and reach reproductive status quickly (Nedeau, Smith and Stone, 2004).

During the course of this survey, there was a reappraisal of species determination in the *Anodonta* genus. New DNA evidence found that the basis of species by shape, size and color, only partially aligned with genetic information. Instead of five species, there are 3 distinct clades, California/Winged Floaters (clade 1), Oregon/Western Floater (clade 2) and the Yukon Floater, which shares a clade with a Siberian Anodontid (Chong, Box, Howard, Wolf, Myers, Mock and Toy, 2008). These clade divisions equate a common ancestor within clades, but the determination of whether the previous species divisions are genetically distinct has not yet occurred.

In this study, mussels sampled are identified to species level with the understanding that classification may change. If morphological traits are found amongst DNA divided clades, it is hoped that this information will aid in keeping this data viable.

For this survey, all of the mussels were identified to a species level. However, in the analysis of the population characteristics the mussels sampled were lumped
together into the Genus *Anodonta*. This was done to create a large enough sample to effectively analyze and to create a body of data that will remain relevant – even if species divisions change in the near future.

**Methods**

Many of the standardized methods in the literature for accurately measuring mussel populations are designed for portions of waterways accessible by foot. In deeper habitats, SCUBA or snorkel gear is used to conduct surveys underwater. The Columbia Slough is an atypical waterway and defies many of the standardized sampling methods described in these studies. Due to its managed nature, the system is more lentic than lotic and this low energy coupled with urban sediment inputs creates an interesting phenomenon where super fine sediments never fully settle out but create a super saturated flocculate medium along the channel. In what appears to be shallow water (<1m), a surveyor walking on the channel bottom can sink another meter in depth. When disturbed, this sediment efficiently clouds the water column, taking 30-40 minutes to settle out. Finally, while the water quality has improved over the last 20 years, there are still several toxics found in the sediment of the Middle and Upper Slough (BES, 2007).

These conditions caused consternation when considering which survey instruments to use for gathering data in this survey. A walking survey would not allow the surveyor to sample the gradient across the channel. The slough was historically a
dumping area for refuse and for some residents continues to be so. From a safety perspective, this has placed metal, glass, and other hazards to harm the unwary surveyor. In considering a SCUBA or snorkel survey, there were multiple safety and practical barriers: even if a full body suit was used to limit exposure to toxics, the turbidity created by fanning the substrate or excavating for in situ mussels would hinder viewing conditions and location accuracy. In order to best use the resources and time allowed, as well as reduce incidence of harm, the following two phase survey was designed and implemented. For qualitative information of the area a visual search was performed at selected points. After each area was visually searched, a benthic sampling of the sediment followed with quantitative data gathered for further analysis.

**Phase 1 - Qualitative Survey**

Selection of Points

A series of 17 points was selected throughout the waterways of the Middle and Upper Columbia Slough. These points were randomly selected from the centerline of the channel by a sampling method known as General Randomized Tessellation Stratified (GRTS). This method creates a hierarchical grid with hierarchical addressing. This is best understood as dividing the area that bounds the sample line into 2x2 quadrants. Each quadrant is subdivided into 2x2 again, and again, etc. With each subdivision, there is a random assignment of independent numbers in each sub-region. When the scale of a pixel or point is reached, there is a unique hierarchical address for that point. The addresses are placed in numeric
order along a line and given a length of one. A random start is placed on this line and a systematic sample (in this case an equal unit) selects 17 points from the line. The 17 sampled addresses are then put in Reverse Hierarchical Order to maintain spatial balance (Olson and Stevens, 2004).

The centerline used for selecting the points is a polyline layer in Arc Map from the City of Portland’s database (CGIS). This centerline was corrected for isolated segments and out-of-channel placement of segments as compared to the aerial layer. Where points fell onto bridges, culverts, or other non-habitat, the point was moved upstream/against flow to the first point in open water habitat.

Search methods

Most of the Middle Slough is a simple channel, so that a line drawn perpendicular to flow would be approximately $90^\circ$ to the channel edge. Using the 17 randomly selected points as the guide for the area to be surveyed, a survey site area was determined by bisecting that survey point across the waterway (Figure 3, w). Next, the width of the bisecting line from wetted perimeter to wetted perimeter was doubled. This
measure was used to describe the length of the box (Figure 3, 2w). These sites each have an individual site name incorporating the initials for the slough it was found in and then numerically up stream. For example, the second site from the mouth of the Buffalo Slough is noted as “BS2”, the fourth site along the main stem of the slough is “MS4”. In areas where the survey box fell across an area that could not be surveyed, such as an earthen bridge or culvert, the area of the obstacle was subtracted, and then added onto the portion of the box in the upstream direction.

This methodology was implemented to encompass the same channel conditions throughout the Middle Slough. By keeping the search area proportional to the width of the stream, there were some cases where the area searched was quite large, while other areas were small. This allowed for searching upstream in wider channels, a method that captures the species richness that increases with stream size (Strayer, 1983; Watters, 1992).
The visual search of these areas required a 10 ft flat-bottomed boat, two surveyors (one for rowing and one for surveying), and an Aquascope- a tube with a tilted lens on the immersed end that aids in seeing below the surface. A series of transects parallel to channel length (Figure 3, 2w) were slowly rowed. These transects were approximately the width of the boat, so that by sweeping the Aquascope back and forth along the stern, a visual transect was created. In this way the entire area of the visual search area was visually searched.

Living and dead mussels were counted on the channel bottom, along with whole shells and fragments on the banks. In the case of expired mussels, shell fragments were included, with hinge pairs and fragments totaling a whole individual tallied as one. Also noted was the piling of mussels in one area (area ≤ 1m). These “middens” are where predators of the mussels (e.g. river otters, raccoons, muskrats), sit and consume them. While they are not good for quantitative values, these middens can indicate whether mussels are found nearby and to develop species lists (Strayer and Smith, 2003).

Phase 2 - Quantitative surveys

At the survey point a transect sample from bank to bank/perpendicular to flow was collected (Figure 4). This transect is known as “A” and was excavated to 10cm-15cm below the surface and 0.5 m wide. A depth of 10 centimeters corresponds with excavation samples in other systems with other freshwater mussels (Sheehan, Neves, and Kitchel 1989, Haag and Warren, 2007) and it was expected that all of the mussels
below the immediate surface in this sample will be accounted for. The tool for this excavation is a constructed D-net, like those used for benthic sampling, with this model having a reinforced frame and 1/4 inch wire mesh netting (Appendix A). This sampling net was drawn through the sediment in sweeps, the start of the next sweep occurring where the previous sweep ended. Each sweep was counted and direction of transect noted. All mussels were gathered from the sampling net, but only live mussels were counted and identified to species by the classification standard of Turgeon et al (1998) and Nedeau, Smith, and Stone (2004). Then the length, width, and height to the nearest millimeter of each mussel was measured and the mussels returned to the water.

**Figure 4** - The orange dot on the left is the randomly generated sample point, with the sample transect bounded by the black line to the right. The double transects capture the largest number of mussels visible on the surface. The arrows indicate the direction of sweep along the transect. The far right tan line is the upstream bound of the sight survey and where the quantitative transect would occur if no mussels were found in the visual survey.

During the qualitative or “visual” survey, if a grouping of live mussels was noted in the search area, a transect was drawn perpendicular to flow and passing through the largest collection. This transect, called “B”, was 0.5 meters wide, with the
same depth as the initial transect. Then, a transect “C” of the same width and depth as the others, located parallel to the previous transect, and close to the visual grouping was sampled. The B and C transects were located so that sampling within the same area would not occur, yet be close enough to sample a possible cluster of mussels. If no mussels are seen during the qualitative sample, than transects B and C were located at a distance two width of the channel, or the upstream bound of the visual survey box described in Figure 4.

As of 2008, the Oregon Department of Fish and Wildlife regulates the collection of freshwater mussels with scientific take permits (ODFW, 2008). A detailed description of the project, including the amount of mortality expected was submitted and reviewed by local and state level biologists to determine negative effects on mussel populations. To reduce the mortality of sampled mussels in this project, time out of water for measurements was kept brief and individuals were replaced in the sediment in the correct orientation for feeding. No mortality was observed during this survey.

**Analysis Methods**

After data collection, the information gathered from the above two methods was organized and corrected from the inevitable data entry errors. It was then divided and treated to three different analyses (Figure 5).
The length measurements of each mussel’s shell found in the benthic survey is compiled and used as an age measure. This data is then used to assess the population characteristics of the population of mussels. The count of mussels in each transect of the benthic survey is used to create a density of mussels for each site. These densities are used to create mean densities for each slough and for the entire Middle Slough. The site densities and the visual data are converted into categorical variables and compared in contingency tables to assess the predictive qualities of the visual survey data.
Population dynamics

Mussel assemblages with a stable population show positively skewed, unimodal frequency distributions (Miller and Payne, 1993), of age (x-axis) related to amount (y-axis) as seen in Figure 6. A distribution of this shape corresponds to a moderately long-lived unionid community whose growth slows with age (Miller and Payne, 1993). In a mussel community of this type there are few juveniles, grading into a large portion of the population that is non-growing and sexually mature, which tapers off to a few large, old individuals (Miller and Payne, 1993). Like other r-strategist species, freshwater mussels have large numbers of young. In a population density curve this would represent as a large number just beyond zero, a steep drop to maturity, and then a sowing of loss of the older individuals. The reason that the curve sweeps up towards a unimodal peak from the zero in Figure 6 is the under sampling of the very small individuals in the population (Christian, Harris, Posey, Hockmuth, and Harp, 2005; Payne and Miller, 2000). Glochidia and newly metamorphosed individuals pass through the smallest of mesh sieves and the higher incidence of mortality found in this portion of their lives, both contributing to the lack of shell lengths found on the left hand portion of this curve.

To determine age of mussels, growth rings found in the cross section of a shell or the hinge ligaments are used, but evidence is conflicting.
as to its validity (Anthony, Kesler, Downing, & Downing, 2001; Haag & Commens-Carson, 2008). The most accurate way to determine age is to take morphological measurements over time. In studies of this type, surveyors mark mussels and then resample them over a measured time period producing an average growth/time rate (Villella, Smith, and Lemarie, 2004). Shell length is most often used as the morphological trait to base growth on, and age length ratios exist for some species of freshwater mussels (Chojnacki, Lewandowska, and Rosinska, 2007; Müller and Patzner, 1996). Lacking ratios specific to the *Anodonta* in this survey, the histograms of length frequency of mussels can be substituted to indicate trends in populations (Christian, et.al. 2005; Payne and Miller, 2000; Haag and Warren, 2007). Using shell measurement histograms in this way is a method of comparing the population’s age relative to itself, rather than using absolute age.

This survey uses length of shell as a measure of age. This is the longest measurement of the shell, usually parallel to the hinge and passing through the center of the oblique view of the shell (Figure 7). Data for this analysis comes from every individual mussel found in the quantitative survey.

**Density estimate**

Density estimates for each site were determined by using the unequal transect area method as described in Stehman and Salzer (2000). This method seeks to maintain the population characteristics found at each site and not those created by the sampling method by using averages of counts. The number of mussels found in each transects A, B, and C for each site were divided by the total area of the three transects
for each site for a result of mussels/m² (Equation 1). These density amounts were appended to the information generated in the GRTS sample and analyzed as continuous data within the spatial survey program developed by the Environmental Protection Agency. The functions of this program were specifically written for the analysis of data generated by the GRTS sampling design. For continuous data, estimates of the population mean, total, variance, and standard deviation can be obtained (Kincaid, 2005). This analysis uses the mean of the densities as an indicator of mussel population characteristics.

\[
M_x = \text{number of mussels in transect} \\
A_x = \text{area of each transect} \\
\frac{M_A + M_B + M_C}{A_A + A_B + A_C} = \text{density of site}
\]

**Equation 1**

**Predictive qualities of the visual survey**

The visual survey was used as a way to become familiar with the sites and to gather qualitative data. This data is not an accurate assessment of density, but because it is an easy survey to implement, it is hoped that it can be used as a general measure of mussel population when used by volunteer groups. A multivariate analysis incorporating all of the observed variables in predicting the benthic density results was initially attempted, but due to the small sample size (N=17), this method was not used. Instead, the predictive ability of the visual survey was limited to whether
presence/absence can be determined in the benthic survey. The small sample area (sites) compared to the entire sample area (The Middle Columbia Slough), along with the ability to convert the results of both surveys to categorical variables, and the randomness of the sample points all made a presence/absence easy and accurate to use.

The predictor variables for the analysis came from the visual survey - mussels found on the bank (VB), mussels seen alive in the water (VL), and mussels seen dead in the water (VD). Visual variable counts and benthic densities greater than zero were converted to ones and zeros remained zeros. Each visual variable was individually compared to the benthic response. A simple contingency table is created to compare the counts of each possible outcome (Table 1).

**Table 1** – The ability of the visual survey to predict presence in the benthic survey

<table>
<thead>
<tr>
<th>Visual survey method found <strong>zero</strong> mussels (either VL, VB, or VB).</th>
<th>Benthic survey found <strong>zero</strong> mussels in the transects</th>
<th>Benthic survey found <strong>one or greater</strong> mussels in the transects</th>
</tr>
</thead>
<tbody>
<tr>
<td>Visual survey predicts zero mussels and zero mussels are found in the benthic survey</td>
<td>Visual survey predicts zero mussels and one or greater mussels are found in the benthic survey</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Visual survey method found <strong>one or greater</strong> mussels (either VL, VB, or ...)</th>
<th>Benthic survey found <strong>one or greater</strong> mussels in the transects</th>
<th>Benthic survey predicts one or greater mussels and one or greater mussels are found</th>
</tr>
</thead>
<tbody>
<tr>
<td>Visual survey predicts one or greater mussels and no mussels are found in the benthic survey</td>
<td>Visual survey predicts one or greater mussels and one or greater mussels are found</td>
<td></td>
</tr>
</tbody>
</table>
The chi square test for independence of variables is also included to determine whether there is a significant relationship between the individual visual responses and the categorical benthic values.

RESULTS

Population dynamics

The frequency of shell length as seen in the Middle Slough graph in Figure 8 shows a bimodal distribution. The higher of the two peaks is toward the larger shell size/older mussel age portion of the x axis. To investigate where this bimodal
distribution comes from, it is important to examine the distribution of the sub-sloughs composing the Middle Slough. The shell length distributions for Main Slough, Whitaker Slough and Buffalo Slough are also shown in Figure 8.

The peak density of 4cm for the Main Slough and 9cm for the Whitaker Slough corresponds directly with the two frequency peaks in the Middle Slough histogram. The Buffalo Slough does influence the right peak, but considering the number of observations (n=10), this influence is minor. The sampling of Prison Pond did not find mussels and therefore exerts no influence on this data.

The frequency histograms of shell length for each site were examined to determine if there were any sites with a strong influence on the frequency distribution of the individual sub-sloughs and concurrently the composite Middle Slough as a
whole. In the Whitaker Slough, the two sites with the most mussels and therefore the most influence are Whitaker Slough 2 (WS2) and Whitaker Slough 3 (WS3). The histograms in Figure 9 show that shell lengths between 7-10 cm have a strong influence at these two sites. There are 112 mussels of this length found at these two sites and account for 55% of the total lengths measured in the Whitaker Slough. The histograms of the other sites within the Whitaker slough are close to the “ideal” distribution seen in Figure 6.

Unlike the Whitaker Slough, the site data within the Main Slough shows no overriding influence from any one site. The shell length distribution of each site is close to the positive skew, unimodal curve. An investigation of the mean and median shell length for each site of the Main Slough shows similar values (Table 2). This is indicative of normal distribution and unbiased samples. These attributes indicate a density curve that represents the individual sites and the Main Slough as a whole.

**Figure 9** – The two sites with the greatest influence on shell length in the Whitaker Slough.
Density estimate

The results of the mean density estimates determined for the sub-sloughs and all of the sites together are shown in Table 3. Mussel density has a range of 0.2-0.9 mussels/m$^2$ across all divisions. Standard errors (StdErr) are larger in the Main and Whitaker Slough (0.18, 0.19) while the Buffalo Slough and all sloughs combined have less variance (0.12 for both). LCB and UCB 95Pct in Table 3 mark the upper and lower bounds of the confidence interval. Prison Pond is not included in this estimate because its one site did not have enough variables for an analysis.

<table>
<thead>
<tr>
<th>Main Slough</th>
<th>Site</th>
<th>Median shell length</th>
<th>Mean shell length</th>
<th>Count of mussels</th>
</tr>
</thead>
<tbody>
<tr>
<td>Buffalo Slough</td>
<td>MS1</td>
<td>3.89cm</td>
<td>3.5cm</td>
<td>42</td>
</tr>
<tr>
<td>Buffalo Slough</td>
<td>MS2</td>
<td>4.13cm</td>
<td>4.2cm</td>
<td>29</td>
</tr>
<tr>
<td>Buffalo Slough</td>
<td>MS3</td>
<td>4.13cm</td>
<td>3.7cm</td>
<td>7</td>
</tr>
<tr>
<td>Buffalo Slough</td>
<td>MS4</td>
<td>6.7cm</td>
<td>6.75cm</td>
<td>38</td>
</tr>
<tr>
<td>Buffalo Slough</td>
<td>MS5</td>
<td>6.83cm</td>
<td>6.85cm</td>
<td>46</td>
</tr>
<tr>
<td>Buffalo Slough</td>
<td>MS6</td>
<td>6.5cm</td>
<td>6.2cm</td>
<td>3</td>
</tr>
</tbody>
</table>

Table 3 – Results of cont.analysis from the spsurvey package

<table>
<thead>
<tr>
<th>Type</th>
<th>Subpopulation</th>
<th>Indicator</th>
<th>Statistic</th>
<th># of Sites</th>
<th>Estimate of mean density (mussels/m$^2$)</th>
<th>StdErr</th>
<th>LCB95Pct</th>
<th>UCB95Pct</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sub slough</td>
<td>Buffalo Slough</td>
<td>Density</td>
<td>Mean</td>
<td>3</td>
<td>0.1957</td>
<td>0.1233</td>
<td>0.0460</td>
<td>0.4373</td>
</tr>
<tr>
<td>Sub slough</td>
<td>Main Slough</td>
<td>Density</td>
<td>Mean</td>
<td>6</td>
<td>0.9250</td>
<td>0.1786</td>
<td>0.5749</td>
<td>1.2751</td>
</tr>
<tr>
<td>Sub slough</td>
<td>Whitaker Slough</td>
<td>Density</td>
<td>Mean</td>
<td>7</td>
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<td>0.1947</td>
<td>0.1959</td>
<td>0.9591</td>
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<tr>
<td>all</td>
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<td>Density</td>
<td>Mean</td>
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<td>0.5988</td>
<td>0.1232</td>
<td>0.3573</td>
<td>0.8425</td>
</tr>
</tbody>
</table>
Predictive quality of the visual survey

When interpreting the results of these contingency tables, it is important to remember that the ideal results would be zeros in lower left and upper right of each table. This result would indicate that in every instance the visual survey variable matched the benthic response. If the visual result indicated mussels, then the benthic method found mussels, conversely, if the visual method found no mussels, then the benthic variable indicated no mussels. The comparison results provided in Table 4 show that dead mussels seen during the qualitative survey might provide good information on mussel presence, but that the other two visual counts are less indicative. The portion of the visual survey that recorded live mussels (VL) seen with the Aquascope does not accurately predict mussels present in the following benthic survey. Of the 17 sites, only 8 of the visual sighting yielded the correct response (4+4=8). The more common result was not seeing live mussels when in fact there were mussels found with the dredge net.

Finding dead mussels appears to be the best measure of finding mussels in the benthic survey. 12 of the 17 sites were correctly identified as having a presence or

<table>
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<th></th>
<th>Benthic Density = 0</th>
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<tr>
<td><strong>VB = 0</strong></td>
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<td>3.66</td>
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</tr>
<tr>
<td><strong>VB &gt; 0</strong></td>
<td>0</td>
<td>7</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
absence of mussels utilizing the VD results, with only 5 of the sites having a presence not predicted by the visual survey.

Finally, the observation of mussels on the banks (VB) comes in a close second in predicting benthic results, trading only one site from the accurate prediction of mussels found on bank, presence in the sampling net to not seen in the visual survey but found in the benthic survey.

The result of a false positive, or that there were mussels counted in the visual survey, but no mussels found in the benthic survey, did not occur. This indicates that the visual survey method predicts presence of mussels.

Discussion

Slough Results

Buffalo Slough – Three sites were sampled within this smaller slough and low densities, a total sample of 10 mussels, and older skewed ages, points towards a population that is not faring well. An explanation for this result is the lack of flow in this water body. Water is constrained in the eastern portion of Buffalo Slough by a culvert under 33rd street. This may create a barrier to host fish and impound water during summer months. In the western portion of the Buffalo Slough, water depth becomes shallow in summer (≈ 0.4m), increasing water temperatures which would normally reduce available oxygen in the water (ODEQ, 2005). In the Middle Slough
however, the influx of groundwater during the summer months increases oxygen saturation in the water (Hendrickson, pers.). What is not known is the oxygen level at the interface between water and sediment. This area is where organic matter collects and decomposes, which reduces oxygen at the very place mussels live. There is also no data on the temperature range that these mussels survive or reproduce at. As with many answers, this data generates more questions, both for this slough and the species as a whole. Further study is needed to answer them.

Whitaker Slough – This slough shows a trend of shell lengths increasing towards the older segment of the population. The age that the six to ten centimeter shell length represents is likely the slow growing, adult reproductive population that is mentioned in Miller and Payne (1993). This older skewed population could indicate a population that is having trouble recruiting juveniles.

A possible explanation is the loss of host fish. Without the conversion of glochidia to juveniles, the population would show reduced numbers on the smaller shell length side of the graph. It is also possible that the consumption of juveniles and glochidium by other fish is creating the density curve for the Whitaker Slough seen in Figure 8.

The sites WS2 and WS3 are especially influential on this distribution of shell length (Figure 9). Both of these sites are adjacent to the Whitaker Ponds Natural Area and the NE 47th street bridge. There are three events that happened within the age range of this population and encompass the area these sites fall into. The area around the ponds was rehabilitated from residential / junkyard to a more natural system about
fifteen years ago. The 47th street bridge was changed from culverts to a free flowing channel about ten years ago. The third change for this area is the increase in groundwater created by the drawdown of water in the channel during the summer by MCDD. It is curious that since these three assumedly beneficial events that something has occurred to limit the amount of juvenile mussels entering the population. One would expect that a return to a more “natural” condition for this water body would increase the amount of juveniles seen in the sample.

How the increase in bank side habitat and cooler, clearer water might reduce host fish for these freshwater mussels is unknown. Why these two sites and not the other sites of this slough are unknown, clearly more investigation is needed.

Main Slough – With the highest density estimate and younger age grouping the population characteristics in this slough are promising. The density curve is skewed to the shorter shell length, and this fits the positively skewed definition of a healthy population discussed in Population Dynamics portion of the Methods section. An explanation for this result is possibly the dredging activities of the MCDD. In 2000 the Main Slough was dredged from NE 12th to NE148th to increase channel depth. This dredging removes benthic sediment and places it onto the banks of the slough, which both buries mussels in the dredged sediment or exposes them to the surface. Considering an event of this nature, the left skew of the data likely indicates a repopulation of this slough. Even with a density of less than one mussel per m², the shell length peak at 4cm (Figure 8) indicates a population with reproductive age.
mussels that have access to the host fish their progeny need to successfully complete the change from glochidium to juvenile mussels.

Prison Pond – There are no results for this slough. The one sample area in this slough returned no responses during the visual survey or the benthic survey. This water body has a strong groundwater recharge element and is unusually clear compared to the rest of the slough(s). It is possible that there is some unrecognized factor that limits host fish or mussel survival here, more sample sites would help determine this factor.

Shell Length

When using shell length as an estimate of age, one must be careful in attributing too much weight to the population trends. As an overview, there are some interpretations of the density of shell length graphs. In the sloughs that have enough individuals in the sample to determine population development, two have an older population trend (Whitaker Slough and Buffalo Slough), and the other (Main Slough) a younger trend.

Determining an accurate age to shell length ratio would point towards what occurred in the 6-10 cm time span on the Whitaker slough to create the mussel age class that one sees, especially at WS2 and WS3. Concurrently, the same metric could give a time frame for positive conditions that might indicate what occurred during the 2-6 cm length in the Main Slough to create the population rise.

Density
This study found an overall density of 0.6 mussels/meter$^2$. Historic density estimates for Anodonta are not available for this area but there are three density estimates found in the literature with which to compare this study’s results. The first estimate is from the Middle Fork of the John Day River in Oregon, with ~525 mussels/m$^2$ (Brim Box, Howard, Wolf, O’Brien, Nez, and Close, 2006). This survey was limited to known mussel beds, and is likely overrepresentative. The second estimate has Anodonta densities downstream from the Hanford site on the Columbia River of 0.66 and 0.1 mussels/m$^2$ (Helmstetler and Cowles, 2008). Finally, a study involving three lakes on Vancouver Island, British Columbia has estimates of 10-20/m$^2$ (Martel and Lauzon-Guay, 2005). These estimates are quite varied.

Considering that the Martel and Lauzon-Guay study is the least effected by urbanization and has lentic conditions like those found in the Middle Slough, it most likely approximates historic conditions. The Hanford site study’s sample sites were in the portion of the Columbia River that flowed through the Tri-city area of Richland, Pasco and Kennewick, Washington. While this area is not as heavily impacted watershed as the Columbia Slough is, it is possible that these numbers are a closer estimate to densities found in urbanized areas. In the balance, the Hanford (and this studies) results are what to expect, but the Martel study is what could be if conditions were improved.

With the inclusion of density errors, the upper bound of the confidence interval for any of the sloughs provides a density of 1.3 mussels/m2. In comparing this to the previously cited literature, this does not appear particularly dense for Anodonta. It is possible that the random selection of points used here misses the spatial clustering that
one sees with many of the Unionids. A method that corrects for this spatial congregation is known as adaptive cluster sampling (Strayer and Smith, 2003). This method relies on finding congregations of mussels and then systematically sampling within the cluster. If the Anodontids in the Middle Columbia Slough do exhibit this behavior, then using this method might more accurately determine densities of mussels across the watershed.

As a first try for sampling in the unique conditions found in the Columbia Slough, the method described in this document provides useful densities of mussels for further analysis.

**Predictive qualities of the qualitative survey**

The visual survey could be useful in determining the presence of mussels for the benthic survey. The data that shows the most promise is the amount of mussels seen dead on the benthos with the Aquascope. This is a surprising result. As previously described in the “Quantitative Surveys” portion of “Methods”, the benthic sampling specifically focused on portions of the sample area with congregations of live mussels seen in the visual survey. It was expected that the results of the B and C transects would have helped predict presence in the comparison. However, with these sites, this was not the case.

There are some factors that play a role in these results. The qualitative survey for most of these sites occurred in the summer of 2008, with the benthic sampling occurring over the spring of 2009. While planning a two-stage survey over two
seasons is common practice (Smith, Villella, and Lemarie; 2001), perhaps flow regimes or predation over the winter affected benthic results from early 2009.

A possible explanation of the dead mussels being a better predictor of presence may stem from live mussels pulling themselves below the sediment surface for protection (Nedeau, Smith and Stone; 2004). This might remove live mussels from the visual count. Dead mussels would likely rise to the surface of the benthos and be counted, indicating that there are enough mussels at the site to create a “graveyard”. This also assumes the mussels aren’t earning their name as “floaters” and being transported off site. Viewing conditions are not always in the surveyor’s favor - overcast days, water depth, turbidity, and plant growth all serve to hinder sighting mussels on the benthos.

Midden piles and shells on bank were included in the survey because when planning this survey, these indicators of mussels in the area were thought to represent the local population. If this connection between bank side mussels and mussel presence could be strengthened, it would prove valuable for volunteer groups. They’re easy to see, no special instruments are needed, and if piled, indicative of the predators that are using mussels as a food source. This variable’s predictive power falls just short of significance, but it is possible, as with all of these variables, that more data will make mussel shells seen on the bank an important part of determining the presence of mussels.

Ideally the data needed to confirm these speculations would come from an area with similar conditions. The intention is to sample the randomly generated points in
the Upper Slough, the data of which will be added to this in a final document generated for the City of Portland’s Bureau of Environmental Services.

**Future Use of this information**

Determining what events occurred on a slough wide area or in the local drainage area that influence these population trends would help project managers’ work towards mussel recovery throughout the whole watershed. This data is a “snapshot” of what mussel densities are found currently. A monitoring of landscape scale changes within the watershed - such as creation of impervious surfaces, or increase in streamside habitat - and comparing the results of future studies of mussel density would be incredibly beneficial in increasing the knowledge of how these changes affect mussel populations.

It would also be useful to use the current data to determine if there were some watershed level attribute that correlated with the density amounts. For example, outfall locations, current impervious area within sub watersheds, amount of industry, and any other landscape change. Utilizing Arc View to perform a spatial analysis to determine whether one or a combination of these watershed attributes effects mussel densities would be another way to help managers guide conditions for native mussel population viability.

If there is a spatial clustering of these mussels in the Middle Columbia Slough, preservation of the “patches” of the slough that host these clusters becomes more important for watershed management. Using this GRTS sampling method has provided the necessary density estimates for determining a level of sampling effort in
areas that have these possible clusters of mussels. Using volunteers to gather locations of mussel shells on the bank and/or dead mussels seen below the surface and then using an adaptive or random systematic sampling method would provide better density estimates and answer this question of whether the Anodontids of the Columbia Slough are clustered or not.

**Conclusion**

This survey of Middle Columbia Slough has provided the City of Portland with useful data in determining the status of native freshwater mussels within the watershed. The density estimates and population characteristics show promise for recovering mussel communities in some reaches, and that there are some as yet unknown limiting factors that hinder the mussel population in other sloughs. The visual survey has the propensity for being a useful volunteer tool in measuring mussel presence, but needs more data to be accurate. These methods and the resultant information are a good start to using native freshwater mussels for watershed health indicators in the Middle Columbia Slough.

**Acknowledgements**

This survey would not have been possible without the help and support of the following groups -

- City of Portland’s Bureau of Environmental Services- specifically the Columbia Slough Watershed Team, Field Operations, and the Revegetation group.
• The Columbia Slough Watershed Council
• The Xerces Society
• The Pacific Northwest Native Freshwater Mussel Workgroup
• Thanks again to the review committee – Nancy Hendrickson, Al Smith, and of course my advisor Dr. Joe Maser.

**Thank you**

To the many volunteers who braved the elements and a madman in a boat, my deepest appreciation go to you – Katie Lethe, Jennifer Bildersee, Henry Clark, Janet Johnson (where are you?), Chelsea Hansen, Nick Glass, David Belcher, Nick David, Molly Swanson, Kathy Garris, Jenny Grant, Brian Monnin, and all the volunteers who made this possible.

**and finally…**

To Erin, James, and little baby superman: Thank you for waiting, I’m pretty much done now.

**Bibliography**


Appendix A – Sampling net (DK1000)
The sturdy net detailed above was attached to an eight foot long, one and one half inches in diameter pole. It was able to withstand vigorous shaking, sampling at depth, picking up rocks and sticks, and general abuse.
Appendix B – Upper Slough data

This portion of the report describes the data and areas searched in the Upper Slough of the Columbia Slough, Portland Oregon. It is an addendum to the “Assessment of Native Freshwater Mussel Populations in the Middle Columbia Slough” found associated with this document. The Upper Slough portion of the Columbia Slough has the same physical characteristics, history, and mussel species as described in the Middle Slough document. This addendum will use the same methods, techniques, and analysis as the former document and limit its bulk to describing the areas surveyed and looking at the three questions from the previous document. These questions are:

1) what is the density of mussels in the Upper Slough

2) what are the population characteristics of the Upper Slough

3) What is the predictive power of the visual survey to predict presence of mussels in the Upper Slough?

An interpretation of these results will follow.

The data found herein will then be combined to determine a Middle and Upper Slough analysis of the results. Again, the three analyses of Middle and Upper Sloughs - density, age characteristics, and visual survey’s determination of presence will be showcased and interpreted.

The Upper Slough
The Upper Slough waterway starts at Fairview Lake and travels west to the NE 142nd street levee which separates it from the Middle Slough. The sampling points generated by the GRTS program are found in three distinct waterways - the Main Upper Slough, Four Corners Slough, and West Upper Slough.

The West Upper Slough is part of the “Big Four Corners” complex. In this area, the Main Upper Slough channel intersects with an old channel that was historically connected to the Columbia River. The West Upper Slough is a western arm of this old channel. This channel is characterized by development on both sides of its channel, low, slow moving water, and divided by roads.

The Four Corners Slough channel would probably be better named as the Wyndmarr channel. The channel is narrow and cuts through an open field. Yellow flag iris (*Iris pseudacorus*) is found throughout this channel and is slowly reducing the amount of open water.

The Main Upper Slough is as the name implies the “main channel in this portion of the Columbia Slough. Like the Main Slough in the Middle Slough, all sub sloughs drain into this channel.

There are eight sites within the Upper Slough, four are in the Main Upper Slough, and two each in the Four Corners and the West Upper sloughs. These sites are eight of the original selection of 25 GRTS points placed within the Middle and Upper Slough.

**Densities**
The methods used to determine these densities were the same as those used for the Middle Slough survey. The Four Corner Slough and West Upper Slough have two sites each and the sample size is too small to accurately determine mussel density (Table 1). The Main Upper slough has four sites and while somewhat more robust, there are also not enough mussels in the sample to make an assertion as to density. This leaves the combining of all sites for a mean density within the Upper Slough of 0.19 mussels/m$^2$ with a standard error of 0.1 mussels/m$^2$.

Table 1

<table>
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<th>Subpopulation</th>
<th>Indicator</th>
<th>Statistic</th>
<th># of sites</th>
<th>Estimate of mean density (mussels/m$^2$)</th>
<th>StdError</th>
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<td>0.230</td>
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<tr>
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<td>0.19</td>
<td>0.100</td>
<td>-0.011</td>
<td>0.381</td>
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Population Characteristics
There were few mussels collected during sampling in this portion of the slough. Of the mussels collected, the typical shell length is longer, ranging from 5cm to 14cm. In fact, 14cm is the largest mussel sampled in either the Middle or Upper Slough. The histogram of shell lengths (Figure 1) has a sharp peak at 10cm, indicating a grouping of older mussels. The lack of samples leads to caution when analyzing the population characteristics of the mussels in the Upper Slough. The largest grouping of age occurs in the 9-11cm shell length, characteristically old for Anodontids. The method of looking at each site’s histogram to further analyze the characteristics did not provide any clearer results. This was due to the small amount of individuals in the sample.

**Predictive qualities of the visual survey**

The amount of sites available (N=7) were not enough to get a reliable significant relationship between the two variables.

**Discussion**

It is difficult to determine an overarching analysis of the Upper Slough. The sample size is small, and the number of mussels found within this sample set is also
too small to make good determinations. This can be dealt with in two ways. One is that there were not enough samples, a more concerted effort in this portion of the Columbia Slough is needed to speak definitively as to the population characteristics, and densities found here. The other is that the results found here are indicating a small enough and old enough population of mussels that regardless of sample effort, you can’t find what isn’t there. Obviously, more sampling would be helpful in determining how this population is doing.

The information found in the Upper Slough is not good for the analysis how the population is faring in this area. Using these results from the Upper Slough to determine a level of sampling effort would not be advised. Where this data has the most merit is in its inclusion with the data for the Middle Slough for a Middle and Upper Slough (MUS) analysis of the native mussel population.

The Middle and Upper Slough

Densities of the MUS

The methods used for these results are the same as the Middle and Upper Slough portion of this document. With the inclusion of the Upper Slough data, the densities for all subpopulations are slightly altered compared to the results of the spsurvey utilizing only the data from Middle and Upper sloughs individually. This is likely due to the increased amount of spatial data in the analysis.
Table 2

<table>
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Population Dynamics of the MUS

With the combination of the two sloughs, the resulting histogram is similar to what is observed when the

Figure 2

N = 407
data is exclusively from the Middle Slough (Figure 8 – main document). There is a bimodal histogram with peaks surrounding 4cm and 8cm (Figure 3). Even with that similarity, it appears that the peak at 8cm is broader and the 4cm peak more finite. This is probably due to the widening of the data that occurs when adding more samples. The 8cm shell length peak benefits from the longer shell lengths found in the Upper Slough and isolates the 4cm peak. Remembering that the peak at the 4cm shell length comes from Main Slough sites, it is possible that this peak is more of an anomaly when compared to the entire Middle and Upper Slough.

**Predictive quality of the visual survey**

<table>
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| VL = 0  | 7                   | 8                   | $x^2 = 6.48$  
| VL > 0  | 0                   | 10                  | $p = 0.01$    
| VL = 0  | 7                   | 8                   | $x^2 = 6.48$  
| VL > 0  | 0                   | 10                  | $p = 0.01$   |

Much like the results of the population dynamics above, the results of the predictive quality of the visual survey look similar to the same analysis of the Middle Slough. Once again the presence or absence of mussels seen dead in the channel and the shells seen on the bank have greater predictive power of the results in the benthic survey. The chi-squared and p-values show a greater significance between these results and the ones seen in the Middle Slough. This is further validation of the predictive power of these
visual variables to indicate presence or absence. The live mussels seen during the visual survey continue to be poor choice for indicating presence and absence during the benthic survey.

**Discussion**

The results of the population and predictive are similar to the results of the Middle Slough analysis, this could be due to the “drop in the bucket” effect that the Upper Slough has on the larger data trends found in the Middle Slough. When examining the data on freshwater mussels across the entire Middle and Upper Slough, we find a density of 0.63 mussels/m² and a population that trend towards the longer shell length/older age.

The Upper Slough, specifically the Four Corner Slough and West Upper Slough had sample points that were isolated and close to completely drying out during summer months. These conditions create low oxygen and high temperatures, as well as increasing predation. If mussels are found here, it is for brief periods and not representative of the population. In retrospect, these areas would have been taken out of the survey.

However, there are bright spots in this data. The Main Slough in the Middle Slough has indicated strong levels of reproductive success in the past two years. There is something in those sample areas that makes it possible for juvenile mussels to survive. While the Whitaker Slough shows a trend of older shell lengths, the count of
young mussels is still high. It is possible that the mussels in this slough are only recently finding conditions favorable for reproduction.

Many of the same analyses and conclusions are drawn from the combination of the Middle and Upper Sloughs data as are found in the report on the Middle Slough. What associations of green space, impervious area, outfall contents, and other factors are indicated by these bright spots of higher density and viable shell length curves? Conversely, can a relationship be drawn between these factors and areas of low density, or when some localized event impacted the population dynamics of a sample area? This data will help determine these associations for future management decisions and ultimately lead to a better understanding of native freshwater mussels and the valuable role they play in the watershed.
Appendix C – Location of data

The location of the data used for this analysis is found with three groups, and includes:

- A GIS ArcView map document file (.mxd) that details
  - the spatial location of the GRTS generated points
  - the location, size measurements, and species name of individual mussels sampled in this survey
  - the polygons that encompass the visual search area
- Two MS excel files with
  - the location, size measurements, and species name of individual mussels sampled in this survey
  - a worksheet with the count of mussels at each search area by method of search (VL, VB, VD, and benthic)
- A .pdf copy of the field notes

The three repositories of this information are:

1) The City of Portland - Bureau of Environmental Services - Columbia Slough Watershed Group - attn: Dave Helzer

For city employees
Group 104 - S:\Columbia Slough\Fish & Wildlife\Mussels

2) The Pacific Northwest Native Freshwater Mussel Group – attn: Sarina Jepson
pnwmussel@googlegroups.com

3) The author, David Kennedy -
P3atr33@gmail.com