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Intertidal Habitat Utilization by Endangered Green Sturgeon (Acipenser medirostris), Willapa Bay, Washington

Luke Stilwater
Central Washington University, lukewater22@hotmail.com

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INTERTIDAL HABITAT UTILIZATION BY ENDANGERED GREEN STURGEON (*ACIPENSER MEDIROSTRIS*), WILLAPA BAY, WASHINGTON

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by

Luke Joseph Stilwater

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CENTRAL WASHINGTON UNIVERSITY

Graduate Studies

We hereby approve the thesis of

Luke Joseph Stilwater

Candidate for the degree of Master of Science

APPROVED FOR THE GRADUATE FACULTY

Dr. Anthony Gabriel, Committee Chair

Dr. Megan Walsh

Dr. Karl Lillquist

Dean of Graduate Studies

ABSTRACT

INTERTIDAL HABITAT UTILIZATION BY ENDANGERED GREEN STURGEON (*ACIPENSER MEDIROSTRIS*) WILLAPA BAY, WASHINGTON.

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This study looks at a portion of the designated critical habitat for the threatened southern distinct population segment of green sturgeon (*Acipenser medirostris*) in Willapa Bay, Washington. Willapa Bay is an intermediate size (258.7mi^2) estuary on the southwest coast of Washington State, approximately 30 miles north from the mouth of the Columbia River. Recent studies completed by the National Marine Fisheries Service have shown that significant aggregations of green sturgeon in Willapa Bay occur during the late summer months, and foraging activity for burrowing ghost shrimp (*Neotrypaea californiensis*) is evidenced by small round feeding pits (30-60 cm diameter) in the intertidal substrate. The environmental factors of feeding sites were compared to non-feeding sites nearby. The most prevalent foraging areas of the estuary were identified and associations between feeding sites and environmental factors may present themselves. Our findings show that green sturgeon are feeding in areas with fine-grained sediment (>2.0 phi). Feeding pit observations declined when surveying areas with thick eelgrass beds and increased in bare areas. No feeding pit activity was observed within aquaculture parcels adjacent to feeding sites. The majority of feeding pits were found between +0.25 m and +1.5 m MLLW, in terms of tidal elevation.

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CHAPTER I

INTRODUCTION

Statement of Problem

The green sturgeon (*Acipenser medirostris*) is an anadromous fish that frequents West Coast estuaries such as Willapa Bay, Washington, to hunt for one of their primary prey species, ghost shrimp (*Neotrypaea californiensis*) (Moser et al. 2009.) In April 2006, the National Oceanic and Atmospheric Administration (NOAA) listed the Southern Distinct Population Segment (DPS) of green sturgeon as “threatened” under the Endangered Species Act and the northern segment as a “species of concern”. In 2009, NOAA released a description of critical habitat for the species that included the Willapa Bay estuary in Washington State (NOAA 2015).

Efforts to preserve the habitat for green sturgeon, however, may clash with the interests of the shellfish farmers that work the tidelands in Willapa Bay, the single most productive oyster aquaculture area in Washington (PSI 2013). Shellfish growers claim they must control the populations of ghost shrimp to maintain the viability of their oyster beds. This may inadvertently change the habitat of the sturgeon (Dumbauld et al. 2008).

The interaction between fish and eelgrass beds is also of interest to policy-makers and agencies tasked with the health of this resource. Both the native *Zostera marina* (*Z. marina*) and non-native *Zostera japonica* (*Z. japonica*) species of eelgrass found in Willapa Bay provide certain habitat functions for intertidal species. The goal of “no net loss” of native eelgrass recently set by the Puget Sound Partnership in conjunction with regulatory agencies (WSDNR 2015). Eelgrass beds or meadows have high inter-annual variability and more information is needed about both long-term and short-term factors that could influence the marine environment for eelgrass (WSDNR 2015).

Numerous studies completed by NOAA document the habitat and distribution of green sturgeon in Willapa Bay using techniques such as fish tag telemetry, acoustics mapping and low-tide surveys to show the distributions of sturgeon and their prey throughout the estuary (Corbett et al. 2011). Sturgeons foraging in the intertidal zone excavate small (10-30 cm diameter) feeding pits that remain in the sediment, providing evidence that the fish has been there. More information about the location where these pits tend to be found, the density of pits distributed throughout Willapa Bay, and the environmental characteristics of the foraging sites used by the sturgeon is essential to make informed tideland management decisions and to develop best management practices that can maintain the area as an important source of food for humans as well as sturgeon.

Goals and Objectives

Green sturgeon are an important part of the Willapa Bay ecosystem and represent a valuable natural and cultural resource to the region. The Washington State Department of Natural Resources (WSDNR) wants to continue to lease profitable public-trust tidelands to aquaculture operations while avoiding or minimizing harm to aquatic resources (WSDNR 2012). They are also interested in clarifying the relationships between *Z. marina* eelgrass beds and use by higher trophic species (fish and birds) and monitor the extent of eelgrass (WSDNR 2015). This study investigates green sturgeon foraging activity at several sites with native and non-native eelgrass species. With the listing of the southern DPS of green sturgeon and designation of Willapa Bay as critical habitat, federal agencies are required to ensure their actions do not destroy or adversely modify critical habitat (NMFS 2009).

The purpose of this research is to document feeding activity of green sturgeon at multiple sites throughout Willapa Bay, Washington, focusing on determining the environmental factors that influence sturgeon feeding habitat. Objectives include:

1. mapping spatial distribution of feeding pits at several sites with native *Z.marina* and non-native *Z. japonica* eelgrass species;
2. sampling burrowing shrimp populations to determine potential differences in forage availability and feeding impacts; and
3. comparing other environmental factors such as elevation, sediment characteristics and distance to eelgrass and aquaculture to determine potential controls of feeding patterns.

Significance

This study is significant for state and tribal agencies already studying the green sturgeon in Willapa Bay. By furthering our understanding of the habitat characteristics sturgeon utilize most we can direct development of tidelands away from those areas or devise strategies for improving the habitat for this threatened species. Analysis of the sediment with highest sturgeon pit densities (an indicator of feeding sites) offers valuable insight into the substrate conditions preferred for foraging by green sturgeon. Furthermore, GIS analysis of other intertidal features such as eelgrass cover, tidal elevation, and other features identifying the portion of tidelands in Willapa Bay that have the highest value as sturgeon habitat. Management decisions by regulatory agencies such as the WSDNR based on this research could have significance for the local shellfish farmers who utilize the same tidelands as the green sturgeon to provide their livelihoods.

According to the International Union for the Conservation of Nature (2010), 85% of sturgeon species are at risk of extinction, more than any other group of fish. Sturgeon have an extremely long lineage stretching back over 250 million years and have adapted to many changes in their environment over that time, but the hurdles posed by overfishing, habitat fragmentation, and pollution from anthropogenic activity have wreaked havoc on populations worldwide. A better understanding of green sturgeon habitat in Willapa Bay could be applied to similar estuaries around the globe where other anadromous species are at risk.

CHAPTER II

THE STUDY AREA

Geographic Location

Willapa Bay is an intermediate size (258.7mi^2) estuary on the southwest coast of Washington State, approximately 30 miles north from the mouth of the Columbia River (Fig. 1.).

Fig. 1. General map of study area and watershed (NRCS 2006).

It is separated from the Pacific Ocean by the Long Beach Peninsula, a spit formed by the outflow and sediment load of the Columbia River (Banas et al. 2004).

This estuary resulted from rising sea levels after the last ice age, which inundated the numerous small river valleys (NOAA 2008). The shape of Willapa Bay is basically divided by the two channels stemming from the Willapa River to the east and the Naselle River to the south (Hedgpeth et al. 1981). Willapa Bay has freshwater inputs from the Bone, Palix, North, Feather, Bear, Niawiakum, Willapa and Naselle rivers with the Willapa and Naselle being the largest contributors at the northern and southern extent of the estuary respectively (Fig. 1)(Banas et al. 2004, Moser et al. 2007). These rivers have variable flows due to seasonal fluctuations in rainfall which can have significant impact on the salinity of the estuary, especially in the inner bay away from the opening to the ocean, though the estuary overall tends to remain well-mixed, with over 65% of the water pulled out and replaced with ocean water during each tidal cycle (Banas et al. 2004). The total area of the basin that drains into Willapa Bay is approximately 1,865km^2. The watershed highlands are densely forested with conifers that were readily exploited for lumber throughout the last century. Parts of the high marshland were also converted to pasture through diking and filling (Hedgpeth et al. 1981).

Weather and Climate

Willapa Bay is classified as a temperate marine climate which receives most precipitation as rain. Willapa Bay receives an annual rainfall of about 85 inches, most of which falls during the winter months (Banas et al. 2004). The prevailing wind at Willapa Bay blows in off the ocean towards the southeast. Winds at Willapa Bay can influence local wave conditions and during storm events at high tide they can move sediment between shoaling areas and intertidal flats (Andrews 1965). The sheltered nature of Willapa Bay and large intertidal area make for warmer conditions. The temperature averages 7^{o}-9^{o} C in the winter and 14^{o}-20^{o} C in the summer (Chin and Hill 1978). The salinity and temperature of Willapa Bay is highly dependent on the seasonal fluctuations of river flows and is an important factor to understanding the ideal conditions for summer aggregations of green sturgeon (Moser 2007).

Intertidal Characteristics

The intertidal zone is defined as the area exposed at low tide and submerged at high tide. The average tidal range of Willapa Bay is 2.7 meters and depth of the main channels ranges from 8 to 20 meters (Fig. 2). The seasonal cycle of tides, wind, and river inputs create a net increase in sediment and tidal flats within the bay (Banas et al. 2004). At the northern end of Willapa Bay sites such as Stony Point and Ellen Sands receive their sediment from erosion of Cape Shoalwater on the northern side of the estuary opening (Andrews 1965). Sources of sediment at the south end of the bay are almost exclusively river input. Wind also transports sediment into Willapa Bay from the dunes and beaches of the peninsula.

Sediment composition in Willapa Bay is primarily muddy in the upper intertidal and subtidal zones with some muddy-sandy areas in the middle intertidal zone (Gingras et al. 2001). The firmness of substrate affects the distribution of burrowing shrimp and other intertidal fauna, with shrimp favoring softer substrates and bivalves preferring firmer areas (Dumbauld et al. 1996). In general, there are two sedimentary environments within Willapa Bay; the tidal flats exposed frequently at low tide and inundated during high tide (i.e. intertidal zone), and the channels (Stanley, Nahcotta and Willapa) and distributaries within the estuary that remain submerged (i.e. subtidal zone) (Fig. 2). The river channels are characterized by fine sand and silt, with areas of higher current accumulating larger grain sizes (Gingras et al. 2001). The tidal flats of Willapa Bay are composed of well-sorted fine sand, with silt and clay size particles accumulating in the southern end of the bay where they are deposited by several rivers that enter the estuary (Andrews 1965). A relationship exists between grain size and organic content/organic nitrogen; both of these values increase with a decrease in grain-size of tidal flat sediment (Andrews 1965). Andrews (1965) also observed that waves generated within Willapa Bay are quite capable of moving bottom sediment up onto the flats. He also notes a particular storm at high tide on October 28th 1962 that moved enough sediment to bury entire oyster beds in fine material.

Oyster Aquaculture

More than half of the estuary surface area is in the intertidal zone (Fig. 2). This makes it an optimal location for commercial aquaculture operations that rely on extensive tide flats to grow and harvest their crop (Hedgpeth et al. 1981, Moser et al. 2007). In the past, native Olympia oyster (*Ostrea lurida*) beds inhabited the estuary naturally and were simply harvested and sold, but growers soon replaced these with the larger and more resilient Pacific oyster (*Crassostrea gigas*) that thrives in aquaculture operations and has higher export value (PSI 2013). Oyster aquaculture operations are concentrated in the tidal range of +1.2 to -1.1 m MLLW (Hiss et al. 1986). Some of the factors that can affect oyster rearing include the salinity, temperature, exposure time, and type of substrate

(Sanford 2012). The most commonly reared shellfish is the Pacific oyster (*Crassotrea gigas*). These oysters are "seeded" into the growing bed by dispersing the small half inch juvenile oysters attached to pieces of old oyster shell. Oysters are placed in growing areas closer to the tributaries of the bay to reach minimum size, they are then moved to fattening beds closer to the ocean or channels where they have better access to their food source, phytoplankton (Hiss et al. 1986 and Sanford 2012). Oysters typically spend a year on the fattening beds before being harvested.

Eighty percent of the areas of bivalve aquaculture production in Pacific County overlap with recorded eelgrass beds (PSI 2013). Areas of dredged or hand-picked shellfish beds contained higher densities of eelgrass than mudflats in the same area without underlying structure (The Watershed Company 2014). The filtering effect of bivalves creates a more conducive environment for eelgrass growth, but the disturbance associated with maintaining and harvesting these species can limit the extent of eelgrass (Dumbauld 2009). Other vegetation found in the intertidal zone includes red and green algae, which is prevalent all along Washington's coast.

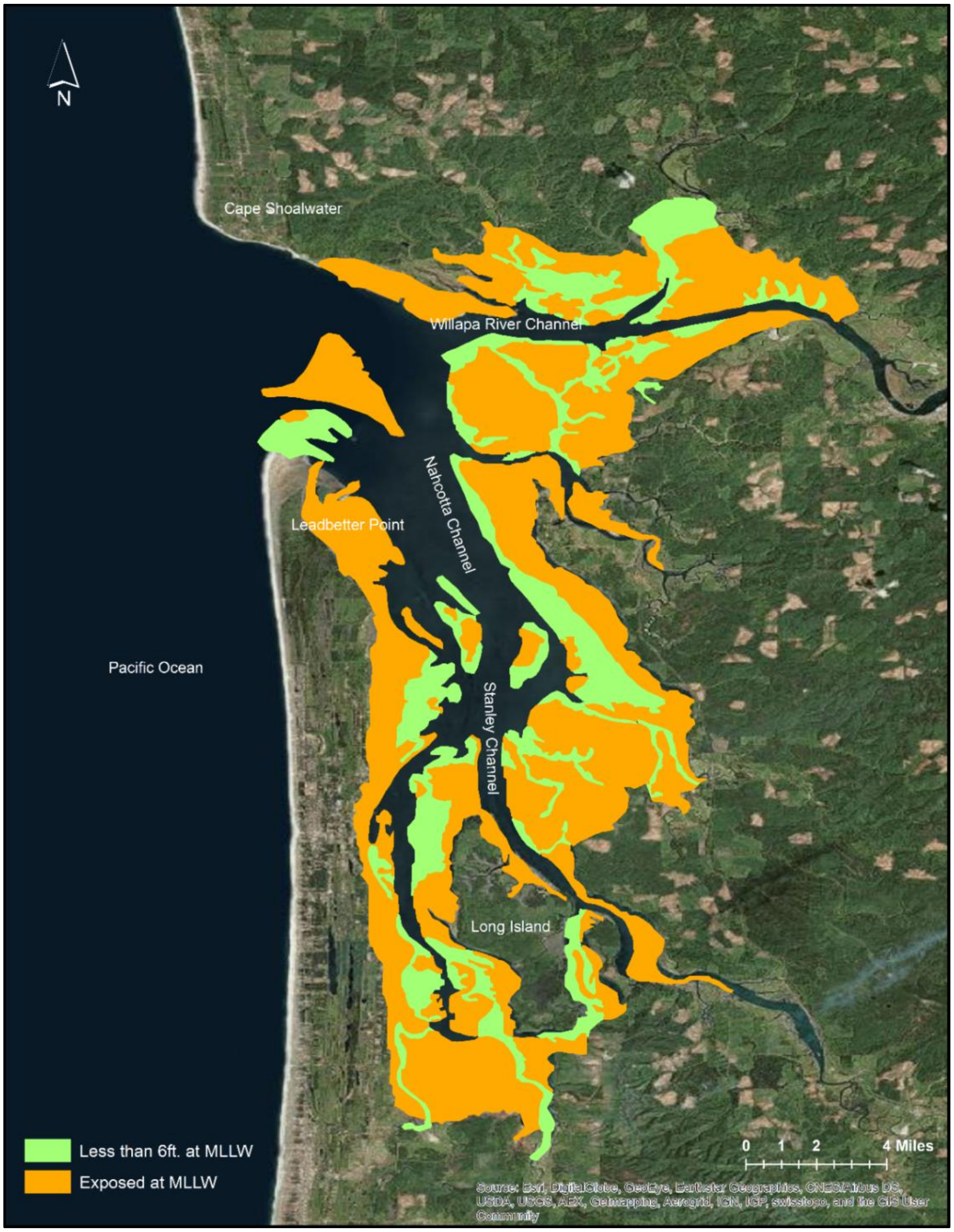

Fig. 2. Willapa Bay, exposure at low tide and main navigation channels. Areas in yellow are exposed at Mean Low Low Water (MLLW) while the areas in green are at depths of less than six feet at the same tidal level (Modified from Hedgpeth et al. 1981).

The State of Washington through the Puget Sound Partnership has set a policy of "no net loss" of native eelgrass. This policy, although focused on Pacific salmon restoration and eelgrass as nursery habitat, can also benefit other resident species if the habitat associations are known. Research on juvenile salmon has shown they spend a limited amount of time in sea grass and there are no significant eelgrass beds within the Columbia River system, which receives the highest annual return of Pacific salmon (SMA 2009). Even though salmon does not seem to lend weight to the importance of sea grasses, there are several other commercial species that are commonly found as juveniles within sea grass meadows, including herring, Dungeness crab (*Cancer magister*), English sole, and rockfish, whose populations within eelgrass beds have already been quantified (Armstrong et al. 2003). Additional use by non-commercial species such as green sturgeon is currently un-documented, and may be an important concern for eelgrass meadow management within the state.

Research has shown that eelgrasses such as *Z. marina* modify the habitat, and can be a valuable indicator species when assessing overall coastal conditions (Waycott et al. 2009). Characteristics and habitat value of eelgrass beds cannot be determined by only looking at one aspect such as density (Van Horne 1983). Other factors must be taken into consideration including the complexity, spatial pattern, and relative location of beds, which can all contribute to variations in site conditions (Beck et al. 2001). More information is needed to determine the degree to which fish and bird communities differ inside and outside eelgrass meadows, and how predation pressures change around the edges of these meadows.

If green sturgeon habitat can be linked to *Z. marina* eelgrass beds, then it would also be useful to know the factors that shape eelgrass distribution within Willapa Bay specifically. Eelgrass beds respond to physical and chemical stressors with changes in the extent, density and morphology (Thom et al. 2003). Eelgrass distributions are concentrated between 0 and -1.5 MLLW, with upper limits determined by the desiccation of shoots during low tide, and corresponding lower limits controlled by the availability of light for photosynthesis (Thom et al. 2003, Hedgpeth and Obrebski 1981, Phillips 1984). A recent study concluded that shoot density is also positively linked with increasing summer salinity in the bay, and negatively with water temperature at the monitoring sites (Thom et al. 2003). Eelgrass beds are also sensitive to changes in water level and circulation in terms of its distribution. Water level affects the dessication time of eelgrass exposed at low tide and the gradient at which *Z. marina* extends from deeper water into the intertidal flats (Thom et al. 2003). Circulation of water in the estuary can influence the distribution of eelgrass by affecting the transport and deposition of seeds (Borde et al. 2003). Due to the influence of El Niño and La Niña events, the annual change in overall eelgrass abundance within Willapa Bay can be as high as 700% (Thom et al. 2003), as occurred during a five-fold increase in eelgrass from 1998-2000. El Niño and La Niña events are different stages of the El Niño Southern Oscillation which affect ocean temperature. El Niño is the warmer period while La Niña is typically colder. This extreme annual variability makes planning and protection efforts difficult. Climate variation is a certainty in Pacific Northwest estuaries, either aiding or hindering the growth and flowering ability of eelgrass.

The effects of erosion and turbidity also need to be considered in determining the controls of eelgrass distribution. As coastal estuaries face the threat of changing sea levels, an analysis of how those changes will affect eelgrass beds is essential to protecting desirable habitat and for long-term planning of any restoration efforts (Thom 2000). Information on distribution of habitat types within estuaries can help refine models of the ecosystem interactions in our Northwest estuaries. While eelgrass meadows change tidelands in terms of structure, they also contribute to the chemical composition, organic content, and other attributes of the water body.

Tideland Management in Willapa Bay

A study by Hedgpeth and Obrebski (1981) indicated a large reduction (-35.5%) in tidal marsh area in Willapa Bay, between U.S. Geodetic surveys in 1905 and 1974. The major cause of this reduction was dike and fill operations. Dredging activities that have influenced other Pacific Northwest estuaries are not as apparent in Willapa Bay, which did not require the extensive maintenance of navigation channels for large vessels because of the nature of the regional economy (Borde et al. 2003). Additional habitat changes have been caused by the introduction of non-native species. One species of non-native cord grass, *Spartina alterniflora*, was introduced around 1880 as a packing material used to ship Atlantic oysters (Borde et al. 2003). By 1980, the cord grass was producing its own viable seed populations within Willapa Bay, and has continually increased its range on previously un-vegetated tidal flats (Borde et al. 2003). The extent of non-native eelgrass, *Z. japonica*, is of special interest to aquaculture operations that have begun spraying to control this species to protect their oyster beds. According to a 2013 *Z. japonica* research project, the non-native eelgrass is extending its range within

Willapa Bay and Gig Harbor. This increase in *Z. japonica* has been accompanied by an increase of fine sediment settling in previously sandy areas, which creates a tidal flat substrate consistency that is unsuitable for aquaculture. The range increase measured was from a previous survey of eelgrass beds conducted by the USDA in 2006 and 2007 (WSDNR/USDA 2013).

During Washington's induction to the union in 1889, the state government asserted ownership of all beds and shores of navigable waters up to the high water line (WSDNR 2010). The state legislature of 1889-1890 authorized sale of public tidelands to private parties and as a result 60% of state beaches were sold before discontinuing this practice in 1971 (WSDNR 2010). The remaining publicly owned aquatic lands allowed access to waters for transportation, fish and shellfish propagation, and other "water-oriented" industries. Currently, there are approximately 1,300 miles of tideland shoreline in state ownership, managed by various state agencies such as Washington State Parks and Recreation, Washington State Department of Fish and Wildlife, and the Washington State Department of Natural Resources (WSDNR 2012).

Management of these shoreline resources are principally authorized through the Shoreline Management Act (SMA), which was passed in 1971 with an overall purpose: "to prevent the inherent harm in an uncoordinated and piecemeal development of the state's shorelines." Some applicable components of the SMA include:

1. shoreline use restrictions to control pollution and prevent damage to environment;

2. protecting the land, water and associated wildlife from adverse effects by requiring mitigation for allowed uses;
3. providing public access to publicly owned areas, as well as preserving or enlarging recreational areas; and
4. public ownership of all state waters are not invalidated by private ownership of the underlying land.

The WSDNR is steward to over 2.6 million acres of state-owned aquatic lands. This was established when Washington became a state in 1889 under the "equal footing doctrine", article 17 of the U.S. Constitution and Washington State RCW 79.105 to ensure access to the navigable waters for fishing, transportation and trade (WSDNR 2012). One of the main goals of the Aquatic Resources division is to "Encourage direct public use and access, foster water-dependent uses, ensure environmental protection, promote continuing production of renewable resources, allow for suitable lands to be used for mineral and material production, generate income from the use of aquatic lands" (WSDNR 2012). Income is generated from selling rights to harvest wild geoducks and shellfish, and from leasing/licensing state-owned aquatic lands, which is used to fund other public services such as education (Fig. 3) (WSDNR 2010).

There is a strict protocol for leasing State-owned aquatic lands. The process is detailed briefly below and relates to the multitude of aquaculture operations in Willapa Bay (Fig. 3):

1. call DNR Aquatics to determine if your project is on state-owned lands;

2. file a Joint Aquatic Resources Permit Application;

3. preliminary review of land available, and proposed use;

4. review of application for potential environmental impacts, denial of application or authorization to obtain permits;

5. Coordination between WSDNR land-manager and lessee to obtain all licenses, easements, and rights-of-entry; terms and conditions of authorization including rent, survey requirements, insurance, are reviewed;

6. Review by WSDNR of application to ensure completion of step 5, and, if appropriate, offers an authorization to use state-owned aquatic lands.

Any efforts to use spraying or mechanical mitigation techniques to deter burrowing shrimp and non-native eelgrass have to be assessed by the agency in respect to these protocols.

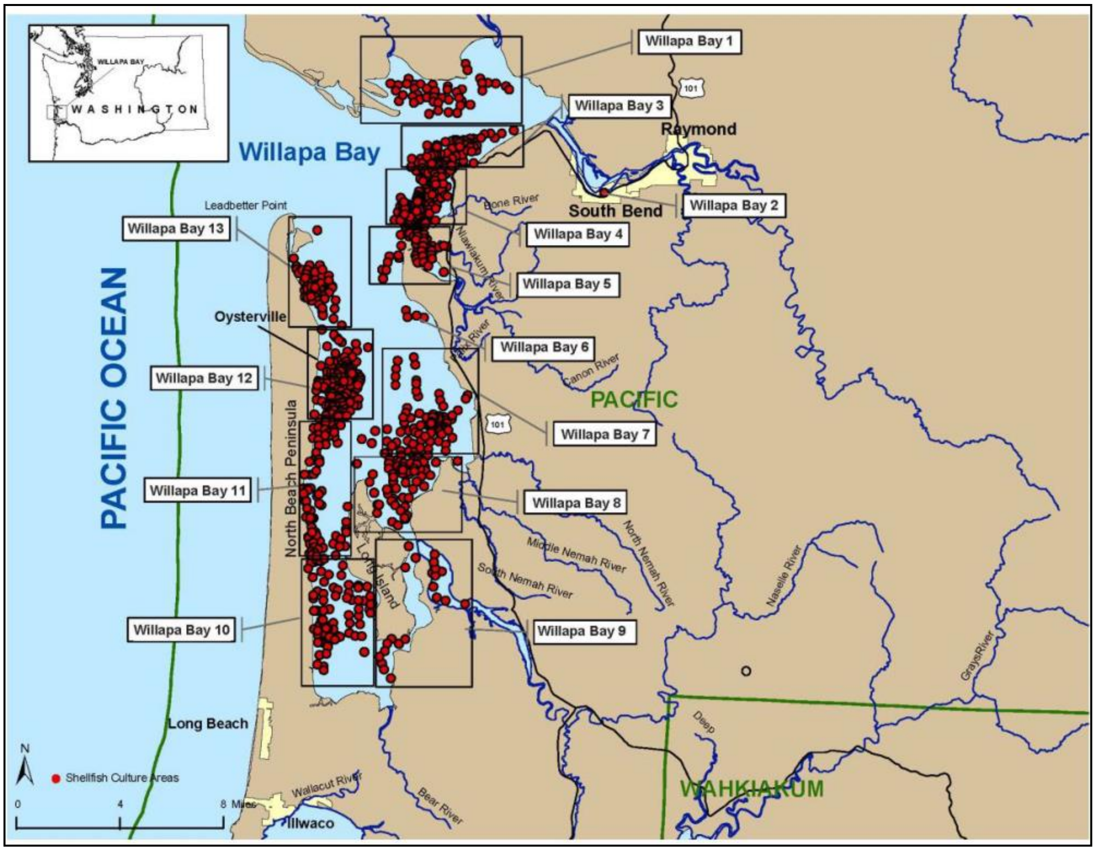

Fig. 3. Willapa Bay Shellfish Aquaculture Operations (The Watershed Co. et al. 2014).

CHAPTER III

LITERATURE REVIEW

Green Sturgeon Biology and Ecology

The North American green sturgeon is a long-lived (up to 70 years) anadromous species of bony fish that frequents west coast estuaries such as Willapa Bay, Washington (Nakamoto et al. 1995, Corbett et al. 2011). Anadromous species reproduce in freshwater but spend most of their lives in the ocean. For the green sturgeon, this means they spend the first 1-3 years in the freshwater stream of their birth before becoming increasingly marine dwellers (Nakamoto et al. 1995, Moser et al. 2007) During their sub-adult phase, the sturgeon occupy the sub-littoral zone off the coast at depths less than 100 meters through the winter and spring (Erickson et al. 2007). There are significant aggregations of these fish during the late summer and fall in estuaries such as those of the Columbia River, Willapa Bay, and Grays Harbor, Washington (Moser et al. 2007, Dumbauld et al. 2008). Green sturgeon do not reach reproductive maturity until they are 15-20 years old and return to the stream of their own birth to spawn (Nakamoto et al. 1995, Adams et al. 2007). Green sturgeon are broadcast spawners that deposit their eggs over cobble beds. They produce fewer eggs than white sturgeon (Acipenser transmontanus), but their eggs are larger, providing more nourishment to the larvae after hatching (Adams et al. 2006).

West Coast spawning populations of green sturgeon have been documented only in the Klamath (Oregon/California), Rogue (California) and Sacramento (California) rivers (NMFS 2009). The difference between northern and southern population segments is based on which stream the fish spawn in. Sacramento River fish represent the southern DPS while the Klamath and Rogue make up the northern DPS. In April 2006, the National Marine Fisheries Service (NMFS) listed the southern DPS of green sturgeon as threatened under the Endangered Species Act (ESA); the northern DPS is currently a species of concern (NMFS 2009).

Green sturgeon have been found to migrate rapidly between Washington state estuaries, across state and national boundaries and throughout varying salinity in their constant search for prey species and periodic returns to their natal spawning streams (Moser 2007). Research suggests that eighty percent of the green sturgeon that gather in Willapa Bay come from the southern DPS, making this estuary an important part of the habitat for this threatened species (Dumbauld et al. 2008). Some of the activities that may threaten this population while in the bay include shrimp control activities, inadvertent by-catch in the salmon fishery, tideland modification, and non-point source pollution. Aggregations of green sturgeon take place during late spring into early fall when the water temperature in estuaries exceeds that of the ocean by 2° Celsius (Moser et al. 2007). The current hypothesis for this activity is that the sturgeons are maximizing their growth potential during this period by feeding in the warmer waters of estuary systems (Moser et al. 2007). Green sturgeon movements within Willapa Bay tend to follow the highest distributions of burrowing shrimp (Moser et al. 2009).

The feeding habits of green sturgeon have been studied in relation to their morphology and potential for aquaculture. These fish have a unique combination of electroreceptors, barbels (whisker-like sensory organ), and a protruding mouth located on the ventral side of the head that lend to the opportunistic feeding on mollusks, bivalves, and even small fish to obtain their dietary requirements (Miller 2006). The anchovy, for example, is an important species for the white sturgeon in the Columbia River estuary and the green sturgeon has the same adaptations that would allow for the feeding on small fishes (Miller 2006). Studies have shown that sturgeon have poor eyesight that does not appear to be of use in feeding activity (Miller 1987). Sturgeons detect their prey by constantly roving across the substrate, using scent, electroreceptors and sensitive barbels to hone in on prey (Erickson and Hightower 2007). Once prey has been detected, sturgeon can force their jaws downward from the bottom of the head, extending their reach and using a powerful suction force to pull the prey item into their mouth. The length of their jaw protrusion determines in part the suction force generated within the mouth and palate as negative pressure is created and explains how the sturgeon can capture so many types of prey despite not being an exceptionally fast, chase predator (Goldsworthy 2007). The array of sensory systems utilized by the green sturgeon allows them to feed in daylight or darkness, in clear or turbid water, engaging in opportunistic feeding of benthic and pelagic prey (Miller 2006). These adaptations have given the sturgeon an ability to survive in various conditions and to take advantage of prey species that are inaccessible to other fish species. A study on rearing green sturgeon larvae in captivity found that the juveniles exhibited limited mobility and activity during daylight hours, and more vigorous feeding and movement during the night (Van Eeneennaam 2001). This may reflect a predisposition of the species to feed in a nocturnal pattern, as light also does not affect their prey detecting senses.

One indicator of feeding in shallow areas can be the sight of a sturgeon caudal fin rapidly moving back and forth while the fish is positioned with its head down in the substrate (Moser 2016). Another study of green sturgeon feeding activity used tagged individuals to track feeding movements of fish actively pursuing northern anchovies in Humboldt Bay (Goldsworthy et al. 2007). The fish were found to frequent higher tidal elevation areas when the tide permitted, using small channels adjacent to aquaculture beds to facilitate this movement. The incursions into these higher elevation areas occur relatively rapidly and opportunistically as green sturgeon searched for or pursued prey that inhabited the intertidal area, or were seeking refuge in the eelgrass/aquaculture beds (Goldsworthy et al. 2007).

A recent study conducted by Moser (2017) has illuminated some more aspects of green sturgeon feeding activity in Willapa Bay. This study found that the highest density of feeding pits were located in shallow, intertidal muddy areas with no eelgrass and relatively small grain sized substrate, and were completely absent within oyster beds. They also found that the presence of *Z. japonica* greatly reduced the prevalence of feeding pits, though some pits were found to occur within the beds of native *Z. marina* beds. Further examination of sub-tidal pits found that the lowest density feeding areas were also the deepest, perhaps relating to a reduction of burrowing shrimp density at those locations, as approximately 75% of the pit variability was linked to burrowing shrimp densities.

Eelgrass Life Cycle and Habitat Function

Z. marina in the Pacific Northwest typically experience flowering from March to July. Seed germination is most prevalent from April to July, although this can take place at any time of the year. They have a burst in vegetative growth in July and then produce seeds from July to October. Those seed can be dispersed from the middle of August to October and by November the plant has produced winter leaves. The non-native species of eelgrass, *Z. japonica*, experiences seed germination from the middle of March to August, vegetative growth from April to January and can flower at any time, although the maximum growth and flowering both occur in August (Phillips 1984). *Z. marina* beds are an important coastal resource as they provide food both directly and through decomposition. The standing submerged leaves also act as a refuge area for fish and invertabrates and many types of fish important to Pacific Northwest fisheries use this environment as nursery habitat (Dumbauld et al. 2003). *Z. japonica* beds are shorter and provide less cover for animals although this species of eelgrass is a favorite food of some migratory waterfowl such as Brandt geese (Phillips 1984).

Burrowing Shrimp Biology, Ecology and Management

The primary prey of green sturgeon in Willapa Bay are the mud shrimp (*Upogebia pugettensis*), and ghost shrimp (*Neotrypaea californiensis*), the latter being most prevalent (Dumbauld et al. 2008). Both species of burrowing shrimp are native to Pacific Coast estuaries, and have been actively surveyed in Willapa Bay since 1989 (Dumbauld et al. 2008). These burrowing shrimp species dig extensive networks of tunnels in the sediment approximately 9-21 mm in diameter and 40-60 cm deep that can affect the habitat composition for other intertidal fauna (Dumbauld et al. 1996). Shrimp densities have been quantified by counting the number of burrow openings, and through coring and sieving of sediment to obtain shrimp counts. Foraging activity on the tide flats of Willapa Bay has been quantified by the number and distribution of small feeding pits (30-60 cm in diameter) created when sturgeon excavate sediment in search of their quarry. These pits have been surveyed previously by acoustic methods in the sub-tidal zone and visual survey in the intertidal areas (Moser et al. 2009, Corbett et al. 2011). Feeding pits are a valuable indicator of the areas of Willapa Bay most frequented by green sturgeon, but are also highly ephemeral features that may be washed away during times of high turbulence (Moser 2016).

Ghost shrimp are also utilized by the sport fishing industry as bait for salmon, but harvests for this use have not been sufficient to maintain the quality of tidelands for aquaculture. The aquaculture industry employs pest control methods to limit the numbers of burrowing shrimp near their operations, because the excavating activity of these invertebrates can cause mature bivalves to sink into the substrate or the larvae to suffocate by covering them with a fine layer of sediment (Dumbauld et al. 2008).

Ghost shrimp have been most commonly controlled in Willapa Bay through the application of Carbyl (made by Sevin), a pesticide that has been sprayed on the burrows during low tide since the 1960s (Frew 2013). However, the use of Carbyl has been limited by regulation so new pest management techniques are needed (Felsot et al. 2002). Several other chemical insecticides have been proposed for use in Willapa Bay tidelands to control shrimp populations. For example, one recent study proposed the use of a neonicotinoid called imidacloprid (Felsot et al. 2002). The results showed that the correct application of this chemical can cull shrimp effectively while remaining well below the EPA threshold of acute toxicity exposure for endangered species, and below lethal concentrations during all tidal conditions when tested on juvenile white sturgeon (Frew 2013 and Felsot et al. 2002).

Alternative treatments for burrowing shrimp control are born out of the concern for non-target species in affected tidelands. The study of shrimp as a food source for endangered species is a common concern in both the Puget Sound and Willapa Bay. Shrimp populations have increased dramatically since the 1950s and a decrease in predation by species such as green sturgeon could be a contributing factor (Dumbauld et al. 2008), along with warming ocean temperatures. Ghost shrimp have been making headlines in Washington State as a keystone species in the Puget Sound for whales that bring tourists to the area and are also harvested for bait in the sport fishing industry. Surveys conducted in Puget Sound concluded that the biomass of shrimp available was more than enough to support whale feeding activities and commercial harvest for bait (Thompson 2016). An analysis of the number of shrimp taken by green sturgeon during forage activities is a useful tool for assessing the effect these fish have on shrimp densities. If populations of large predators such as the green sturgeon became higher as they were in the past, fewer pest control methods for burrowing shrimp might be necessary (Dumbauld et al. 2008).

CHAPTER IV

METHODS

Sample Site Selection

Ten sample sites were selected within Willapa Bay with the guidance of both WSDNR Aquatics personnel and local researchers from the University of Washington. WSDNR has been conducting burrowing shrimp surveys prior to beginning this study and were helpful in selecting sample sites. Sites were selected to get a representative distribution throughout the bay. Paired sites were selected based on their proximity to each other so that the feeding and non-feeding sites were in the same general part of Willapa Bay. Alan Trimble and Jennifer Ruesink from University of Washington’s Marine Ecology Department were invaluable resources for selecting similar intertidal areas that had no recent history of visible sturgeon feeding activity, which were used to select five non-feeding sites for comparison. These were also spread out through Willapa Bay to get a representation of multiple geographies within the estuary and paired with the nearest feeding site, respectively, for comparison of environmental factors. The size of sampling areas was 250 m alongshore by 100 m shoreward. Final sites elected for this study are shown below (Fig. 4).

Sample Site Descriptions

Feeding Sample Sites:

Stony Point (STP) – This site was the northernmost of the study, located about 0.8 miles south from the launch at Tokeland (Fig. 4). It is a sand bar along the channel of the Willapa River heading west towards the bay opening. This sandy bar had limited features other than several navigation pilings and some algae in depressions in the intertidal zone. This area is also a popular trolling spot for salmon and there are dense beds of *Z. marina* present, albeit over 200m away from the boundary of the sampling site and a small patch near the landing zone.

Rhodesia Beach (RHB) – This sandy intertidal flat was located on the eastern side of the bay approximately 7 miles northeast from Nahcotta. There are several small tidal creeks draining off the flats into the Nahcotta Channel. The area is at the waterward side of flats that extend about half a mile further to the east before reaching the bluffs with residential development. There was no eelgrass present at this site but there were patches of green algae.

North Long Island (NLI) – This site was part of an extensive tidal flat at the northern tip of Long Island. There is a small tidal channel to the east of the sampling area, with oyster beds on the other side. To the south, the flats extend on for at least 500 meters before reaching the higher ground of the island. This site was accessed by traveling northeast from Nahcotta Marina and then turning south down the Stanley Channel. There was *Z. marina* and *Z. japonica* intermixed at this site.

Middle Sands (MSD) – This site was located on the western side of the bay, approximately 600 meters south of the Nachotta boat launch. This site is part of a large sandy bar exposed at low tide, yet still separated from the mainland by a small but deep channel between the bar and the western shore. There is an active staked oyster bed at the southern edge of the sampling boundary, and a large *Z. japonica* bed about 70-100 meters north of the aquaculture operation.

South Mill Channel (SMC) – This was the southernmost site in the study and was located about 2.5 miles south of Nahcotta on the western side of the bay. This site also had oyster beds on the southern edge and patchy *Z. japonica* throughout the site. The conditions were very muddy at SMC and the landing beach dropped off steeply into the deeper channel right around the upper limit of *Z. marina* shown on the corresponding map.

Non-Feeding Sample Sites:

Ellen Sands (ESD) – Paired with Stony Point this northernmost non-feeding sample site was located directly east of Grassy Island on the other side of the bay. It is approximately 0.5 miles north of Bay Center on another sandy tidal flat with a rippled surface created by wave action. The sand here was quite firm and there was no eelgrass nearby. This site was also a favorite hangout of local harbor seals that would slide into a deep channel running through the flat. This site was only accessible by boat and required a considerable run from Tokeland.

Grassy Island (GIL) – Paired with Rhodesia Beach and located at the northern end of Long Beach Peninsula on the eastside of Leadbetter Point, this site was one of the hardest to access both by boat and foot. It was a bare sandy flat with no eelgrass present nearby, though the vegetated shoreline was within 50 meters of the sampling area. The flats here contained small wave depressions and polykete burrows, but no sturgeon feeding pits.

Parcel A (PAR) – This non-feeding site, paired with North Long Islnad, is located 200 meters south from the jetty at Nahcotta Marina, also accessible from the western shore at a WDFW public access site for shellfish collection. The site was muddy, scattered with oyster shell, and interspersed with numerous beds of both *Z. marina* and *Z. japonica*. There were also several oyster beds surrounding the sampling area with pine saplings marking the corners of each parcel.

Shovel Middle Sands (SMS) – Located on the same bar as its paired site Middle Sands, but approximately 0.5 miles to the south, this site featured a thick *Z. japonica* bed on the flat and *Z. marina* on the water-ward side. There was more mounds and depressions at this site and a deep channel which bisected the bar to the south of the site.

Little Middle Sands (LMS) – This southernmost non-feeding site was located on the opposite side of a deep channel from Shovel Middle Sands on a sandy bar that extended several hundred meters to the western shore. It is paired with the South Mill Channel feeding site. There was only *Z. marina* present at this site, extending from the sub-tidal environment into the sampling area. The slope from this deep channel and extent of *Z. marina* seem to follow a similar path at this site while there was no *Z. japonica* in close proximity to the site.

Fig. 4. Feeding and non-feeding sample site locations within Willapa Bay, WA.

Sampling Periods

All sampling was conducted during the summer of 2016. Two sampling periods were used in order to capture potential changes in feeding activity between early (7/3 – 7/21) and late summer, (8/2 – 8/19). Feeding pit locations and related shrimp densities were collected during both sampling periods, with tidal elevations ranging between -0.6 and 3.7 m MLLW (Table 1).

Table 1. Dates and tidal ranges of sampling days at each feeding site.

	Early Summer		Late Summer	
Site	Date	Tidal Range meters MLLW (High)(Low)	Date	Tidal Range meters MLLW (High)(Low)
MSD	7/3/2016	(+3.7)(-0.5)	8/3/2016	(+3.5)(-0.4)
SMC	7/4/2016	(+3.7)(-0.6)	8/2/2016	(+3.5)(-0.4)
STP	7/5/2016	(+3.6)(-0.6)	8/19/2016	(+3.4)(-0.3)
RHB	7/6/2016	(+3.5)(-0.5)	8/18/2016	(+3.4)(-0.3)
NLI	7/19/2016	(+3.3)(-0.2)	8/4/2016	(+3.4)(-0.3)

Feeding Pit Identification and Documentation

Green sturgeon feeding pits were previously identified and characterized in Willapa Bay by Moser (2016), Dumbauld (2008), and WSDNR. Using these studies and further training by the WSDNR Aquatics team, a general methodology was developed for determining feeding pits from other inter-tidal depressions based on its size (30-60 cm diameter), roundness (fairly uniform circles), depth (5-10 cm), and the perceived “roughness” of the pit edges (evidence of recent excavation). Pits which had been excavated more recently had a clear ring of broken organic matting around the edge that had not been smoothed by tidal action.

Pit identification began approximately an hour before low tide which ranged from -0.2 m MLLW to -0.6 m MLLW, at each feeding site, daylight permitting, beginning with a walking visual survey for feeding pits in the exposed inter-tidal area within the confines of each 250 m wide sample site boundary. When a pit was found, a marker flag was placed at the location; two field researchers continued walking the area, marking any pits encountered until the entire area has been surveyed. After placing all the flags at the visible feeding pits, handheld Garmin eTrex GPS units were used to georeference the location of all pits (Fig. 5). A subset of 30 pits was also selected for additional core sampling for ghost shrimp. The 30 pits to be sampled were selected randomly but tended to be the more defined pits that both field researchers agreed were characteristic of green sturgeon feeding activity. Sampling of this pit subset occurred after marking all pit locations.

Related elevation profiles were developed for all sites. This was performed by collecting elevation data along transects perpendicular to the water’s edge using a Topcon GPS with Real-Time Correction. Transects were systematically spaced every 25 m in a 250 m area and elevation points were automatically collected every 1 m. This data allowed for the calculation of beach slope and the determination of feeding pit elevations.

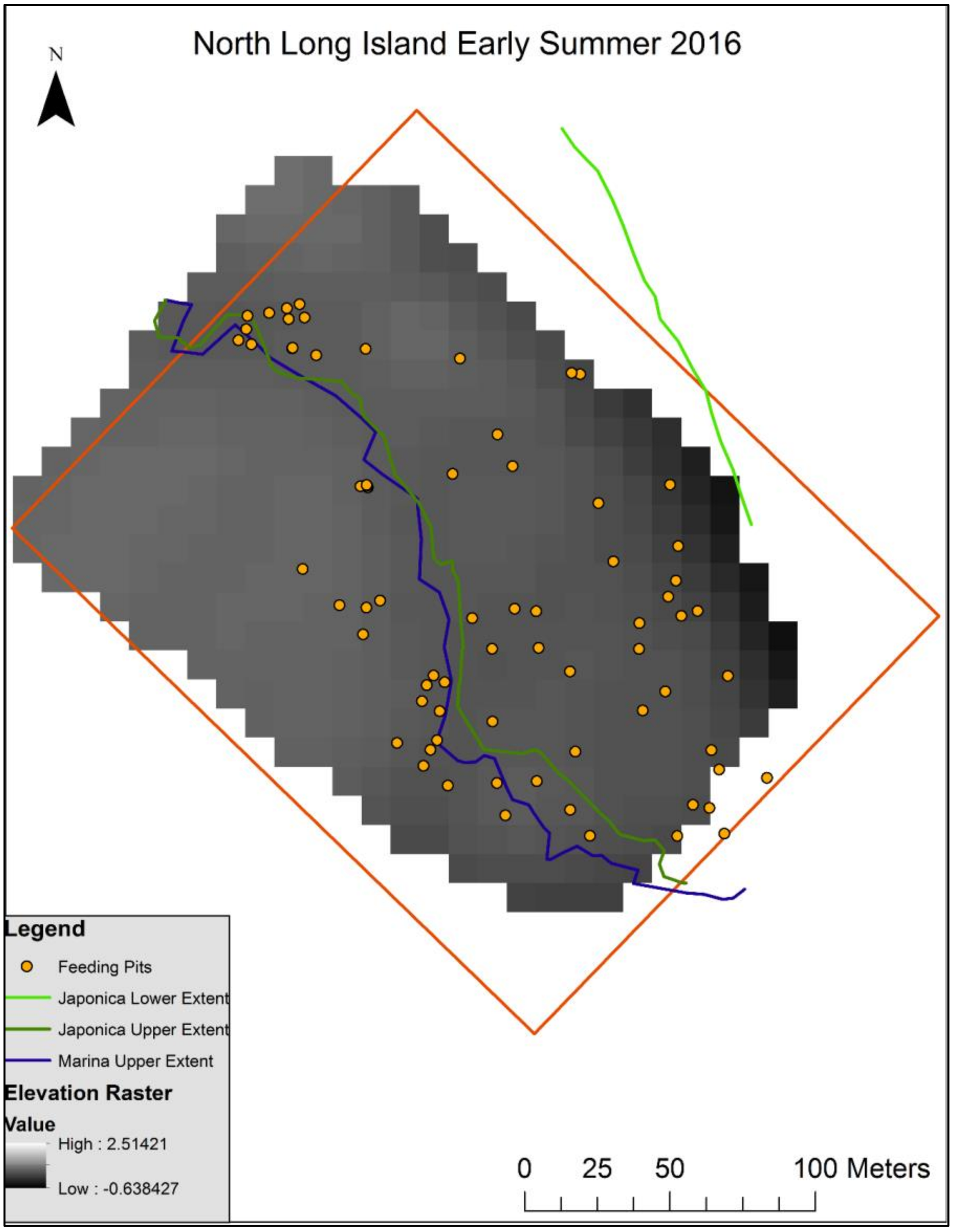

Fig. 5. Pit mapping at North Long Island early summer 2016.

Shrimp Sampling Procedures

Clam guns (30 cm long with a 10 cm diameter opening) made of PVC were used to sample shrimp numbers inside and outside 30 feeding pits at each sample site to see if sturgeon predation significantly affected the local shrimp density (Fig. 6). Burrowing shrimp were sampled at all feeding sites by taking two cores within and outside each feeding pit, spreading out the sediment by hand, counting and measuring each shrimp length in millimeters, and recording the information on data sheets. Finer sediment types with less pore space and higher water content were more effectively excavated with the clam guns than dry sand or rocky substrates, but the research team tried to control for this by excavating an equivalent amount of sediment (depth of clam gun) from pits at each site, through additional coring of the same initial hole.

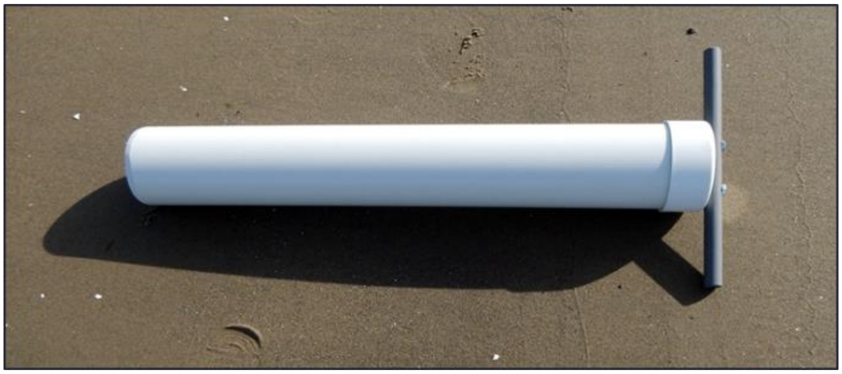

Fig. 6. Clam gun used for shrimp sampling.

Transects were also conducted (7/5/16-7/7/16) at seven of the sampling sites to assess the numbers and length of shrimp found at each. These transects consisted of five clam gun cores distributed within a meter quadrat taken every 10 meters for an average total of 13 quadrats within the site boundary. Using sampling techniques only suitable for adult shrimp (i.e. no use of sieves and screens), counts of shrimp recruits (<10 mm) were undoubtedly under-represented by each method used. These transects were performed at sites where WSDNR were also assessing shrimp populations. Shrimp transect information was useful for comparing the non-feeding sites that did not have core sampling data.

Sediment Sampling

A total of 75 sediment samples were collected from all sites by taking random samples at five locations inside the feeding area and five outside at feeding sites, while the non-feeding sites required only one set of five. Sample locations were determined randomly by throwing a quarter meter quadrat and taking the sample where it landed. Samples were collected using a small garden trowel and stored in labeled zip-lock bags in an iced cooler to reduce decomposition of organics. Sediment samples were analyzed in the CWU Hydrology Lab. Subsamples of each sample were taken and frozen for later analysis of organics. The remainder of the sample was put in a drying oven at 55 degrees Celsius for 48 hours, to remove moisture. Samples were then processed for grain size by putting the sample through a standard set of sediment sieves using a Roto-Tap motorized sieve shaker for 5 minutes and determining the substrate composition by weight. Sieves ranged from Phi size -5 to 5 as most of the sediment consisted of sand and fine silts. This data was entered into Excel spreadsheets as Phi sizes, and graphs were produced to calculate the mean sediment size and sorting at each site.

The organic samples were dried, weighed, and placed in a Gilson MF-6010 muffle oven for two hours at approximately 550^{o} Celsius. The samples were then weighed again to determine the amount of organic content loss-on-ignition. This value was then divided by the original weight of the sample before burning and an average organic content for the sample was determined.

Eelgrass Mapping and Characterization

Using a handheld Garmin eTrex GPS unit, we identified and mapped the upper and lower extents of *Z. marina* and *Z. japonica* at each site, along with the edges of aquaculture beds that were in close proximity (<50m) to the feeding pits (Fig. 5). Aquaculture beds were defined by the numerous stakes used to suspend oysters or by large mounds of active oyster beds within a staked parcel of tideland. We measured the shoot density and length of both *Z. marina* and *Z. japonica* at five random points within the beds bordering or containing feeding pits by throwing a quarter meter quadrat and recording the total number of shoots and the shoot lengths of three random shoots of each species contained therein. Mature specimens of each species can be identified by the leaf length and width but small *Z. marina* cannot be differentiated without examining the sheath at the bottom of the shoot. *Z. marina* sheaths completely enclosed the leaf while *Z. japonica* has two overlapping leaves (PIBC 2004).

Video Recording of Feeding Behavior

To record video of green sturgeon feeding behavior, we used an underwater array of 12 Go-Pro cameras fitted with intervolometers to record two minutes of video every ten minutes. This time interval allowed for extended battery life and less maintenance between tidal cycles. Cameras are equipped with interval meters that record two minutes of video every ten minutes (extending the battery life from the original two hours of continuous video to about eight hours of intermittent capture). The cameras were mounted on PVC stands in the intertidal zone facing towards a bait canister filled with burrowing shrimp to attract sturgeon. Cameras were set out six times during sampling.

Cameras were set at Middle Sands primarily for ease of retrieval the next day en route to another sampling site. The array was set as the tide was coming in after the morning low tide so they could record during daylight hours. Footage was reviewed later for evidence of sturgeon feeding.

GIS and Statistical Analysis

ArcGIS 10.2 was used to compile the spatial data collected, including site boundaries, feeding pit locations, eelgrass limits and elevation profiles. This software was used to process the data collected with the various GPS units into the same datum (WGS Conic) and projection (NAD83), as well as performing clustering and proximity analyses. We were able to show the boundaries of each site and the locations of our sampling sites in relation to one another and create map products for each site that give a visual representation of the features and location of feeding pits mapped. Raster layers of elevation data were produced by personnel of WSDNR Aquatic Resources who used the raw elevation data points to produce a continuous raster for each site, from which pit elevations and slopes were extracted. Data of eelgrass extents and aquaculture edges were used to calculate the distance between feeding pits and these intertidal features.

Some statistics were also performed within this software such as nearest neighbor analysis and Ripley’s K function, to calculate the clustering or dispersion of pits at each site. Average nearest neighbor area was based on the minimum enclosing rectangle that encompassed all feeding pits. Pit density was analyzed using the calculate density tool in ArcGIS. This measures the number of point features within a defined area and gives a density per unit area (meters) output. These areas were generally smaller than the original study site boundaries and represent only the area of the site where pits were found. The number of pits was divided by the total area contained within these polygons to calculate the density within each feeding area.

Additional statistical analysis was conducted after the sampling season with Microsoft Excel and Statistix10. Analysis software was provided by the CWU Geography Department. Non-parametric tests were used due to small sample size, as some sites did not have adequate sample sizes to properly test for the normality assumption required by parametric methods. The Wilcoxon rank sum and signed rank tests were used to compare results obtained inside and outside of feeding pits, early summer versus late summer season, and feeding versus non-feeding sites. Kruskal-Wallis analysis of variance was used to analyze different relationshinps between all sites. Spearman rank correlation and Chi-square were used to find links between different factors such as pit density and shrimp counts, average shrimp length, sediment size, and pit number by tidal elevation class.

CHAPTER V

RESULTS

Feeding Pit Data

When comparing the feeding pit elevations by site irrespective of season, a significant difference was found (Kruskall Wallis, $p<0.05$) (Tables 2, 3, Fig. 7). The highest average feeding pit elevations were found at Stony Point (+0.93 m MLLW) and Middle Sands (+0.90 m MLLW), while the lowest were found at Rhodesia Beach (-0.24 m MLLW). This difference is representative of the different tidal elevations of feeding sites across Willapa Bay, indicating that green sturgeon feed at wide variety of tidal elevations.

No significant differences were found in pit elevations between sampling periods for the majority of feeding sites (Mann Whitney U, $p>0.05$) (Fig. 7), with the exception of North Long Island, where average pit elevations were +0.53 m MLLW in early summer and slightly higher at +0.56 m MLLW in late summer (Mann Whitney U, $p<0.05$) (Table 2).

Table 2. Feeding pit characteristics, including pit density, elevation, average slope, and nearest neighbor analysis (NNA) with corresponding p-values.

Feeding Pit Characteristics								
Site	Density (pits/ha)	Elevation (MLLW)		Slope (degrees)	Nearest Neighbor Analysis (meters)			
Early Summer		Median	IQR	Mean	Observed Mean	Expected Mean	NN-Ratio	P-Value
North Long Island	41	0.53	0.94	0.22	9.07	10.01	0.91	0.13
Middle Sands	69	0.93	0.07	0.23	4.93	9.21	0.53	<0.01
South Mill Channel	217	0.81	0.32	0.33	1.75	4.62	0.38	<0.01
Stony Point	32	0.96	0.11	0.26	3.38	4.79	0.71	<0.01
Rhodesia Beach	99	-0.30	-0.20	0.43	6.44	7.22	0.89	0.26
Late Summer								
North Long Island	107	0.56	0.07	0.22	4.76	8.03	0.59	<0.01
Middle Sands	86	0.91	0.16	0.23	6.56	7.81	0.83	0.03
South Mill Channel	62	0.81	0.28	0.33	6.47	8.29	0.78	<0.01
Stony Point	16	1.00	0.23	0.26	23.39	22.56	0.71	0.76

The general trend in feeding pits related to tidal elevation is a significant increase in the percentage of feeding pits as the tidal elevation decreased, generally moving shoreward from the deeper channels that stayed submerged during most tidal cycles (Table 3). This overall trend is generally evident at all the feeding sites, with the exception of Rhodesia Beach, where all of the feeding pits were found at lower tidal elevations (-0.54 to 0.25 m MLLW).

Table 3. Proportion of feeding pits by tidal elevation (%)

Elevation Class (meters, MLLW)				
Site	-0.54 to 0.25	0.26 to 0.50	0.51 to 0.75	0.76 to 1.50
MSD	0	0	1.0	
NLI	0.3	5.6	15.9	12.3
SMC	0.9	5.5	15.3	0
RHB	4.0	0	0	29.9
STP	0.1	0.3	1.0	0
Total %	4.1	11.4	33.3	7.7

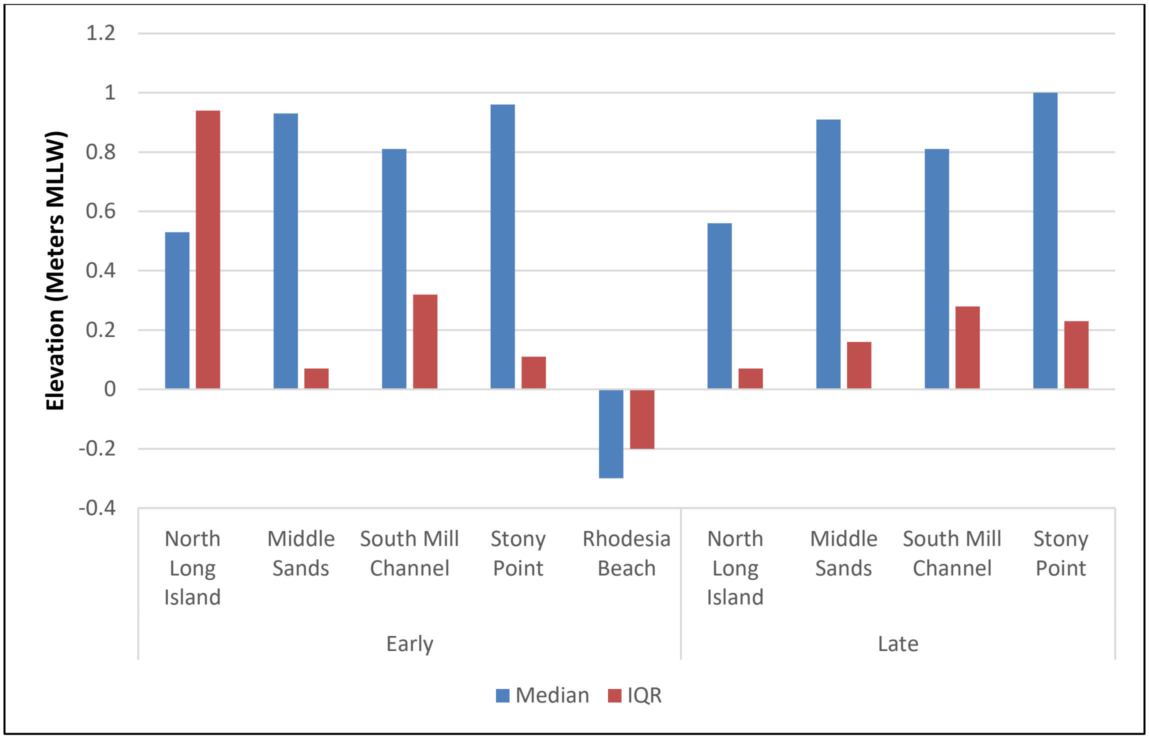

Fig. 7. Feeding pit elevations (median and interquartile range) early and late summer, 2016. (note: No visible pits found at Rhodesia Beach during the late summer.)

The frequency of feeding pits was significantly related to tidal elevation and sample site (chi-square test, $p<0.05$), with a related Cramer's V coefficient of 61%. While most of observed frequencies within each class were generally close to expected, there are several notable exceptions. A much larger number of feeding pits were found than expected at higher tidal elevation classes at both Middle Sands and North Long Island (12.3-15.9% of the total), and at lower tidal elevations at Rhodesia Beach (4.13% of the total).

Differences in mean slope between neighboring feeding and non-feeding sites were insignificant (Wilcoxon signed rank, $p>0.05$). The mean slope at feeding sites was 0.29 degrees declination, ranging between 0.22 to 0.43 degrees (Table 2, Fig. 8). Rhodesia Beach had a higher slope than the other feeding sites owing to its proximity to a steep sided, deep channel. This site also exhibited the least defined pits, also likely due to this proximity to the channel, as more intense currents and a longer inundation period between tidal cycles likely increased erosion of feeding pits. Non-feeding sites had a mean slope of 0.34 degrees, ranging between 0.18 and 0.5 degrees.

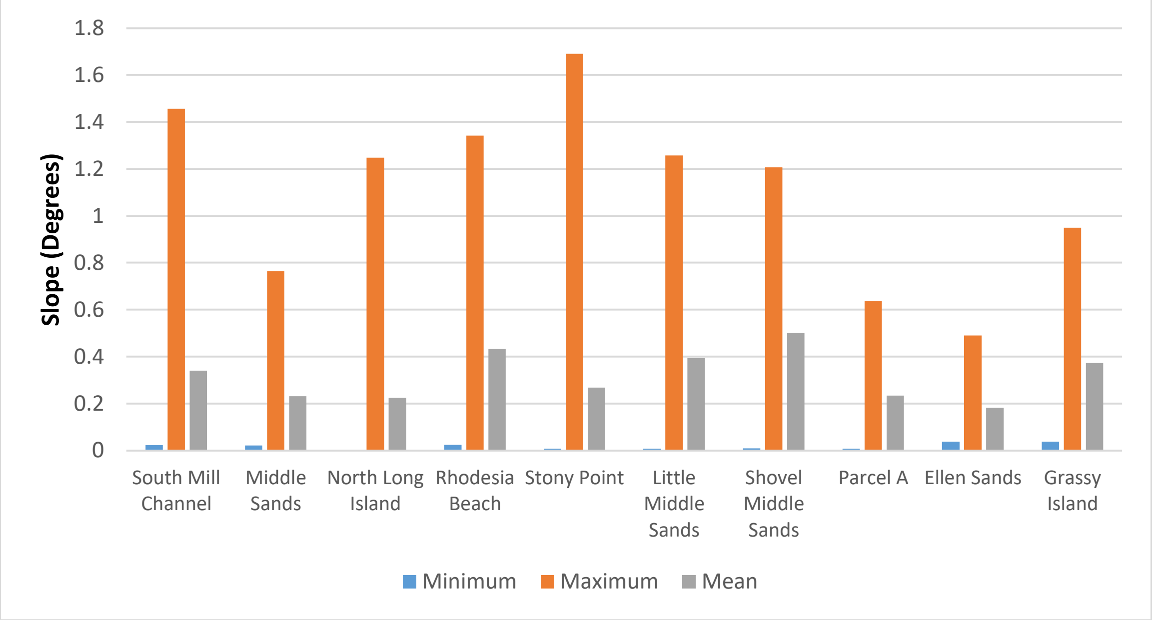

Fig. 8. Slope characteristics of sample sites.

Mean pit densities were highest at South Mill Channel (62 - 217 pits/ha), Middle Sands (69 – 85 pits/ha), and North Long Island (41 – 107 pits/ha) (Table 2, Figs. 9-12). While feeding activity varied between sites, pit densities were not significantly different between early and late summer (Wilcoxon signed rank, $p>0.05$), indicating consistent feeding activity at each site throughout the summer (with the exception of Rhodesia Beach (Fig. 13), which had no visible feeding pits in late summer).

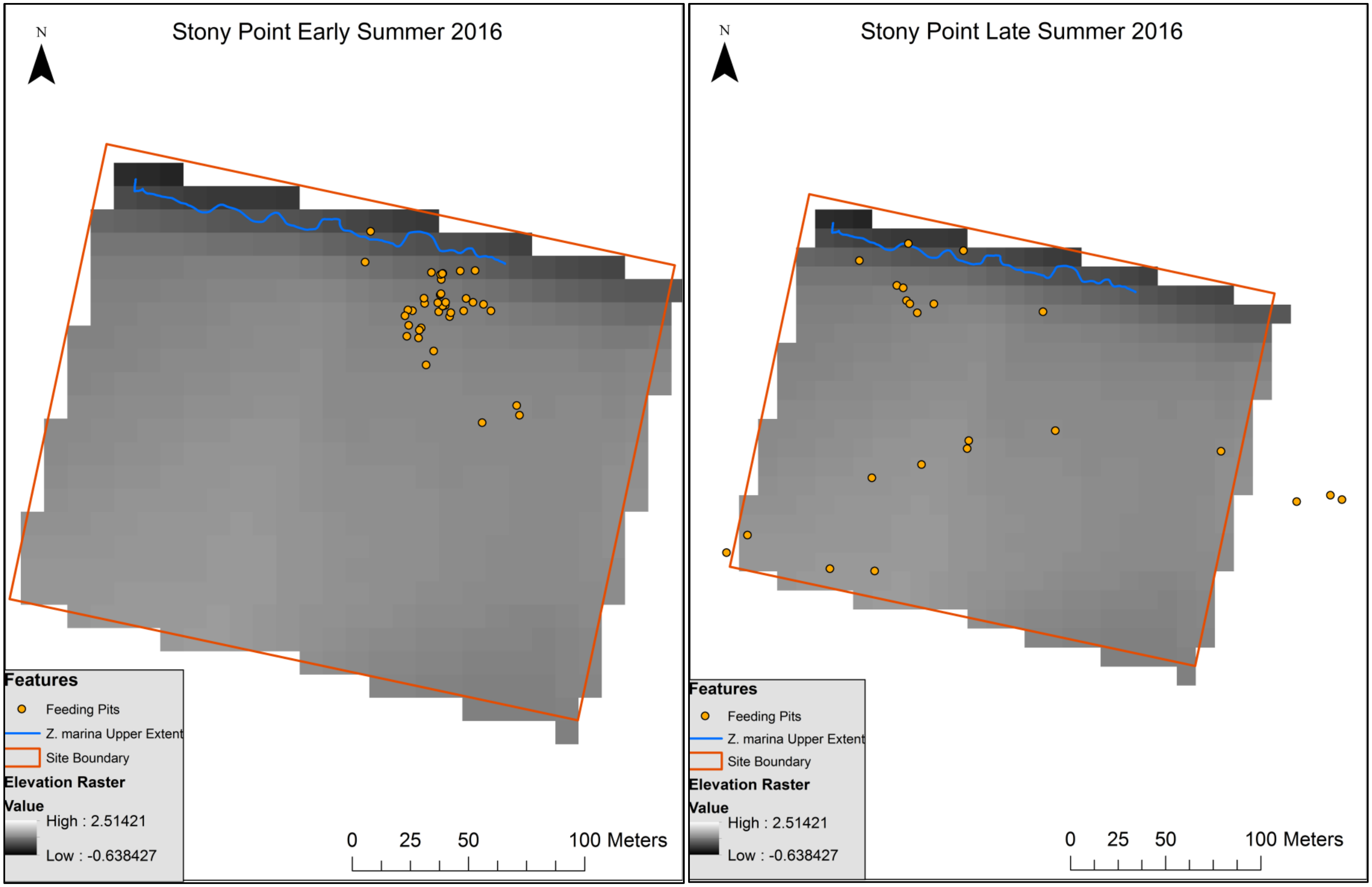

Figure 9. Pit Locations at Stony Point Summer 2016.

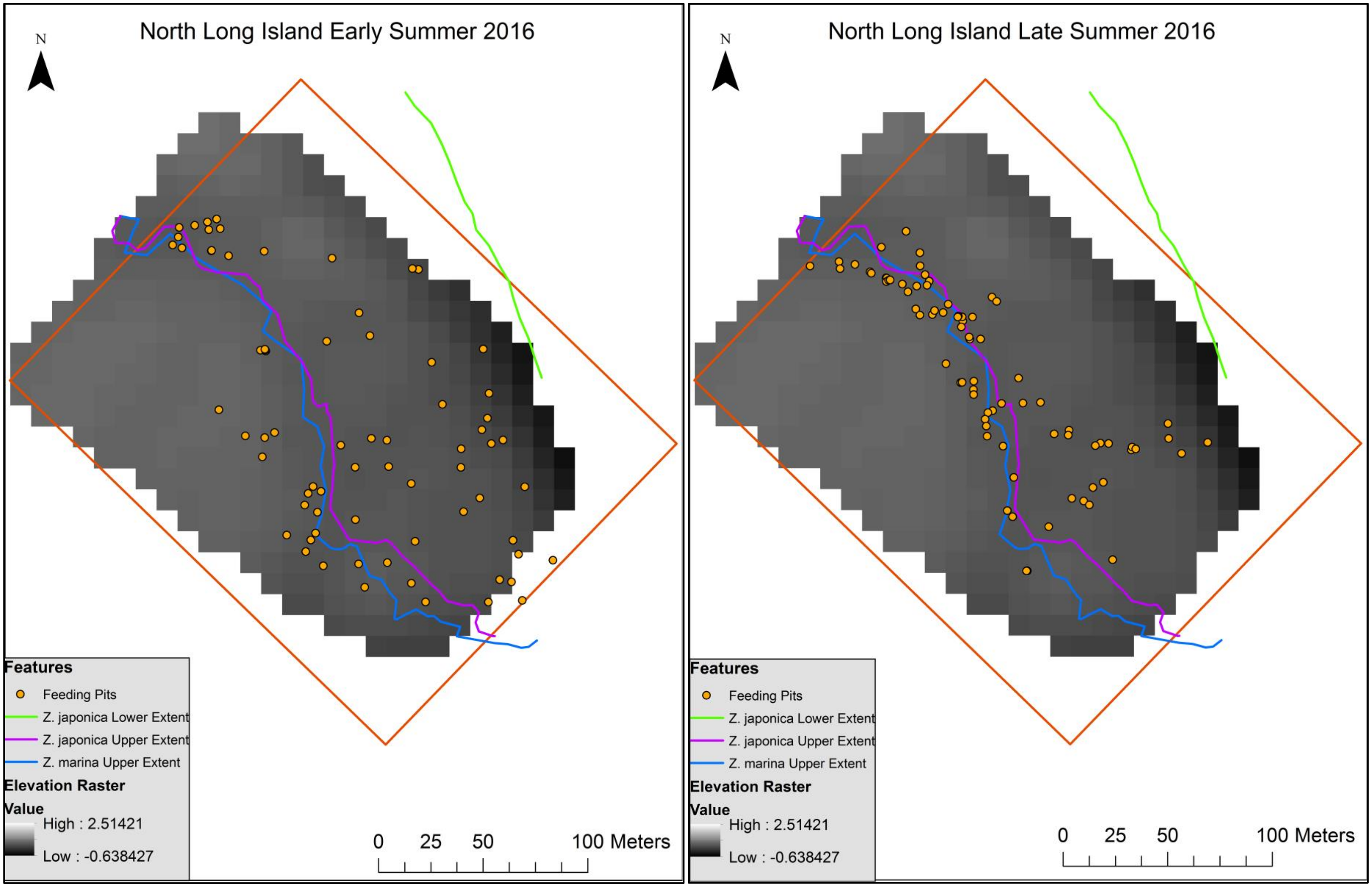

Fig. 10. Feeding pits and eelgrass limits at North Long Island.

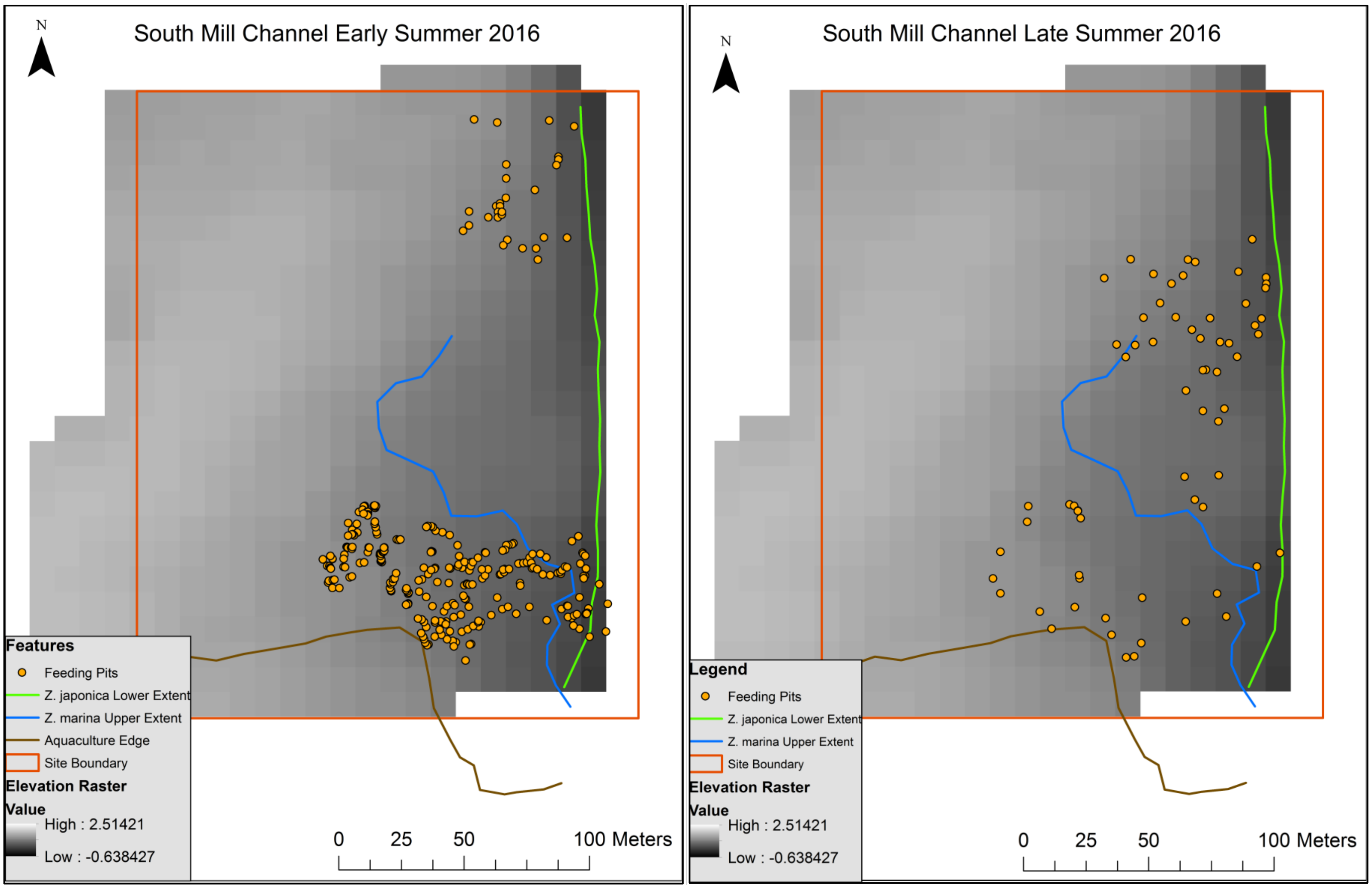

Fig. 11. South Mill Channel feeding pit locations relative to eelgrass limits and aquaculture, early and late summer.

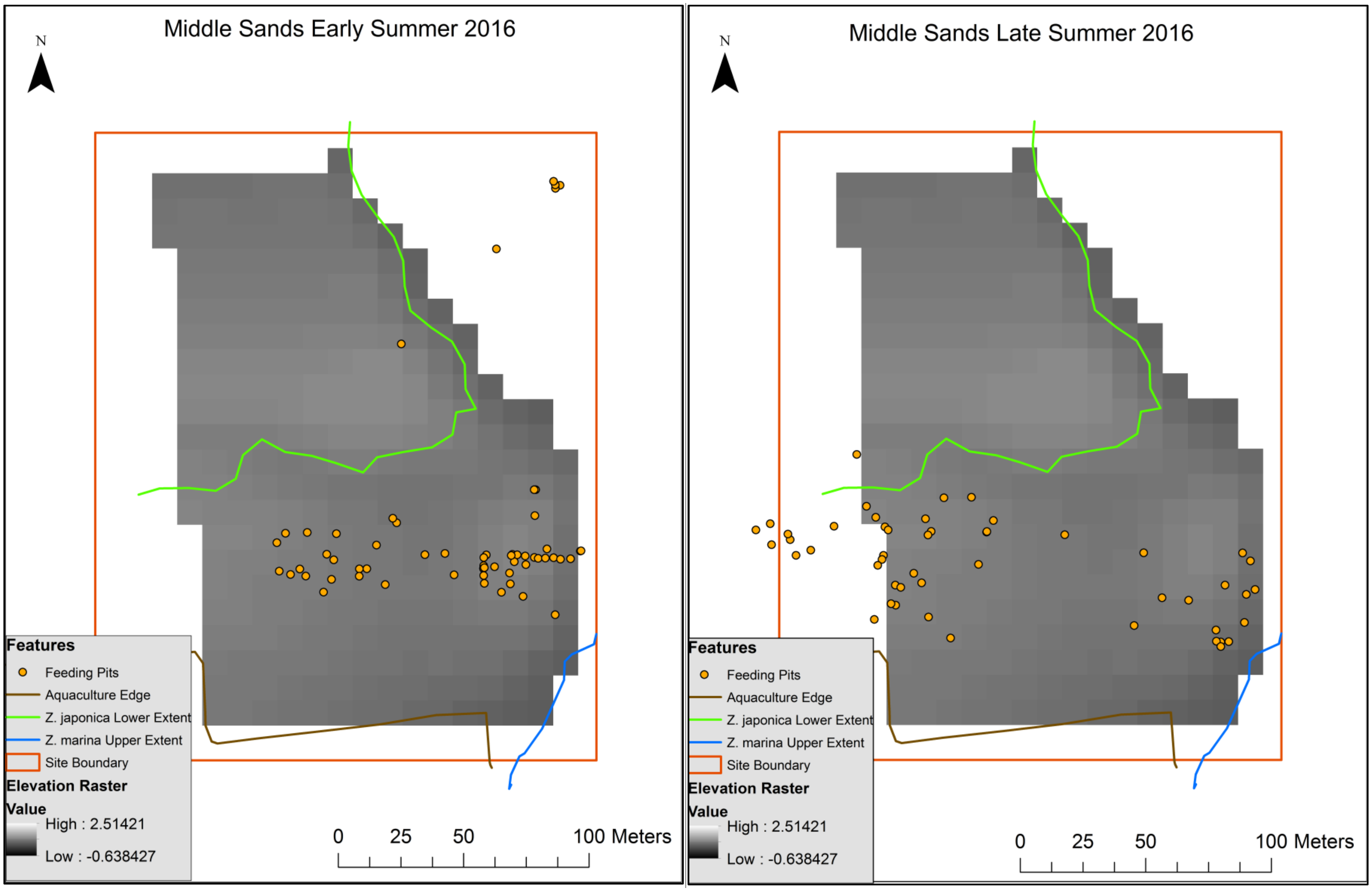

Fig. 12. Middle Sands feeding pit locations relative to eelgrass beds and aquaculture.

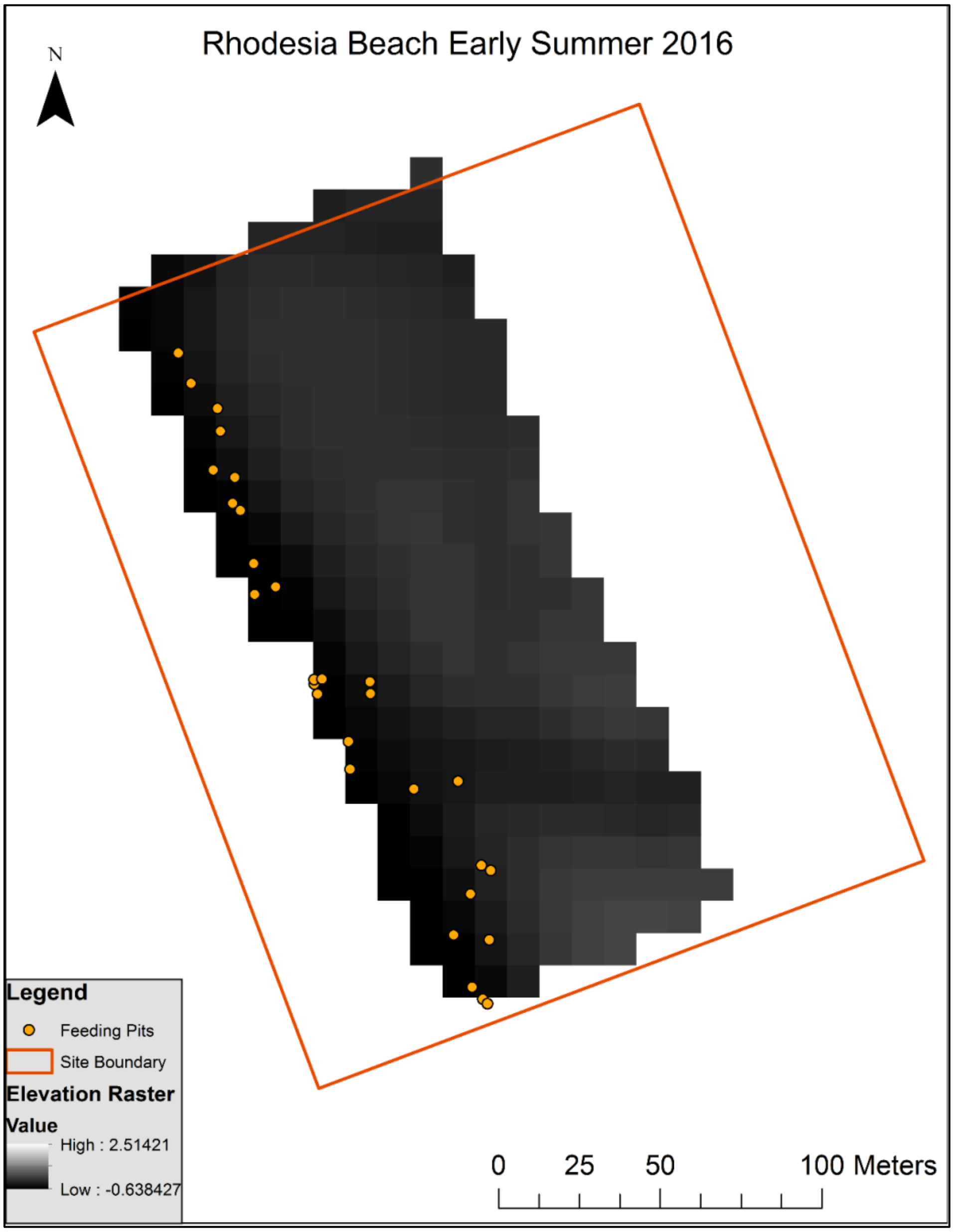

Fig. 13. Pit locations and elevation raster collected from Rhodesia Beach.

The lowest pit densities were found at Stony Point (Table 2, Fig. 14). This site is also the nearest to the outlet of the estuary and does not have any aquaculture or eelgrass beds in close proximity to the sampling site. There was no significant correlation found between mean slope and mean pit density or between mean pit elevation and mean pit density (Spearman rank correlation, p>0.05).

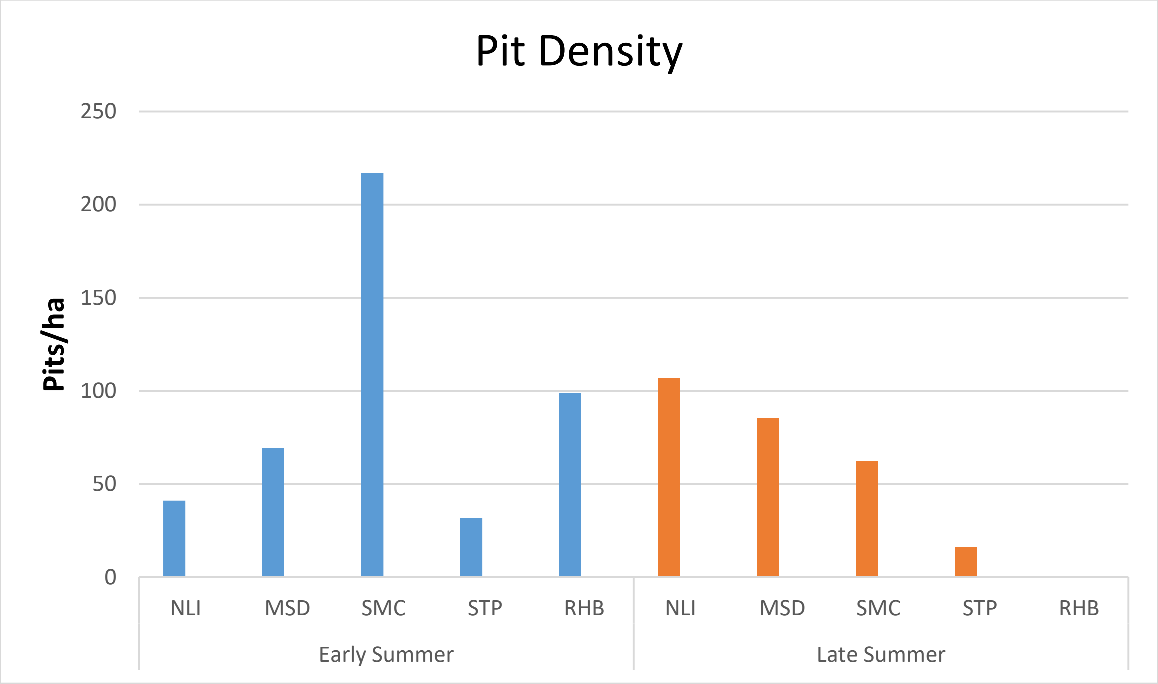

Fig. 14. Mean feeding pit density (pits per hectare) at feeding sites in early and late summer, 2016.

Significant clustering of feeding pit sites, relative to a random distribution of the same number of points in the same size area, is illustrated by an observed mean well below the expected mean and a nearest neighbor ratio of less than 1 ($p < 0.05$). There was significant clustering at all sites, ranging between 4.28 to 1.25 m below the expected distance, except during the late summer season at Stony Point where the observed was 0.83m above the expected distance (Table 2, Fig. 15). The greatest clustering was found at South Mill Channel, where the feeding area was constricted by staked oyster beds to the south and thick eelgrass beds to the north; the predominantly bare area in between these features is where almost all of the feeding activity was evident. This pattern held true at other feeding sites such as North Long Island and Middle Sands where pits declined sharply within eelgrass beds or aquaculture areas.

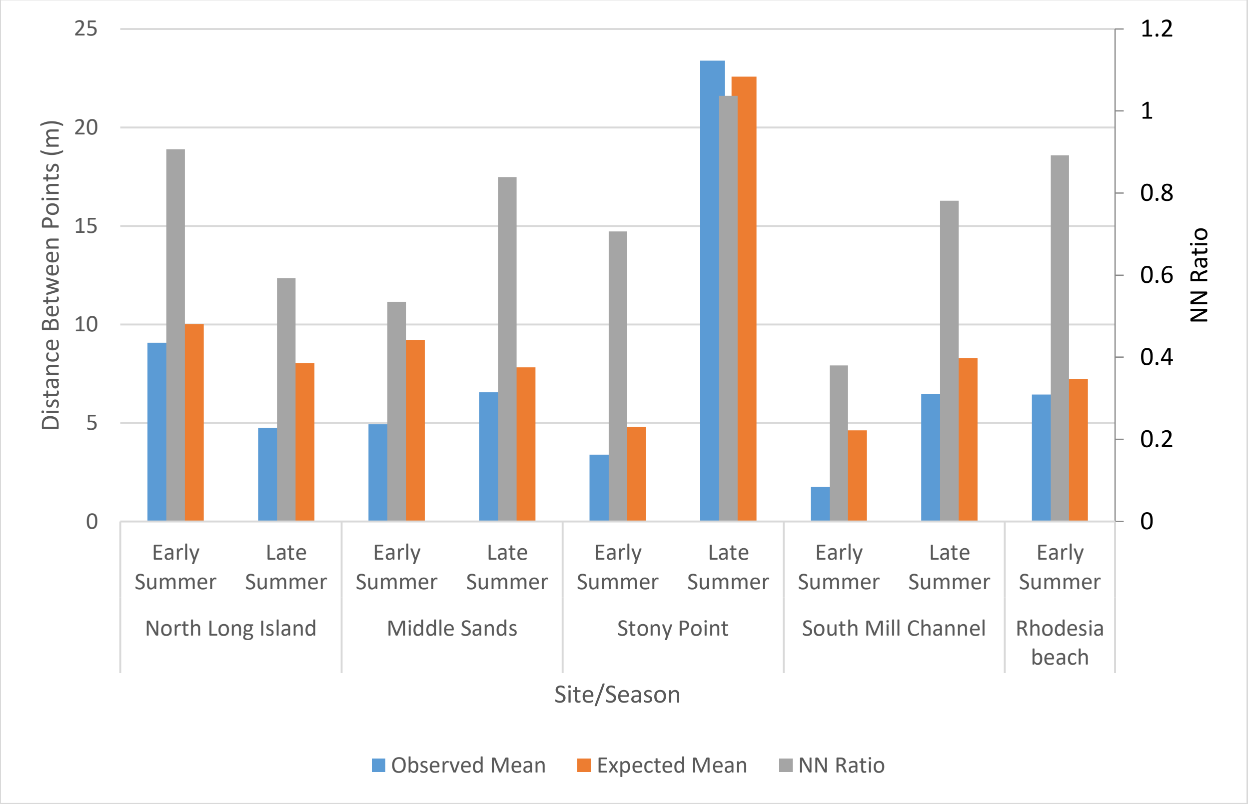

Fig. 15. Nearest neighbor analysis for feeding pits at all feeding sites by season (note: Rhodesia Beach did not have any visible pits during the late summer sampling.)

Sediment Characteristics

All sample sites within the study area were primarily composed of relatively fine-grained sand with varying levels of silt and clay likely due to differences in various environmental controls (e.g., wave energy, aspect, slope, tidal elevation, sediment inputs from rivers). Median sediment size was significantly smaller at feeding than non-feeding sites (Wilcoxon rank sum, $p<0.05$). The median sediment size for feeding sites was 2.0

phi, ranging from 1.92 to 2.08 phi (Table 4, Fig. 16). Non-feeding sites had a median sediment size of 1.93 phi, ranging between 1.86 to 1.98 phi. The highest median phi values were found at South Mill Channel (2.58), showing that the sediment there is composed of more fine-grained material such as silt, while the lowest values occurred at Little Middle Sands and Parcel A, each with a coarser median of 1.87 phi.

Feeding Site	**Sediment Size (phi)**			**Organic Content (%)**		
	Median	Max	Min	Median	Max	Min
North Long Island	2.00	2.23	1.98	1.4	3.1	1.1
South Mill Channel	2.00	2.38	1.98	1.6	2.3	1.0
Stony Point	2.00	2.01	2.00	1.0	1.1	0.9
Rhodesia beach	1.97	1.98	1.91	1.1	1.3	1.1
Middle Sands	2.00	2.00	1.98	1.2	1.7	1.1
Non-Feeding Site						
Little Middle Sands	1.98	2.01	1.93	1.1	1.3	1.0
Parcel A	1.85	1.93	1.81	1.4	1.7	1.1
Ellen Sands	1.98	2.00	1.96	1.0	1.0	0.8
Shovel Middle Sands	1.93	2.10	1.90	1.4	2.0	1.1
Grassy Island	1.98	2.00	1.93	1.0	1.1	0.9

Table 4. Sediment characteristics from feeding and non-feeding sample sites.

The median organic content of substrate at feeding sites was 1.7%, ranging from 1.0% to 2.6%. Non-feeding sites had a median organic content of 1.3%, ranging from

1.0% to 1.5% (Fig. 16). The highest organic content was again found at South Mill Channel, while the lowest organic content was from Ellen Sands, a non-feeding site with no eelgrass present. When comparing only feeding sites, significant differences in substrate organic content were again found between the five sites (Kruskall Wallis, $p<0.05$) with South Mill Channel and North Long Island having the highest amounts (1.64% and 1.35%), while Stony Point had the lowest (0.97%).

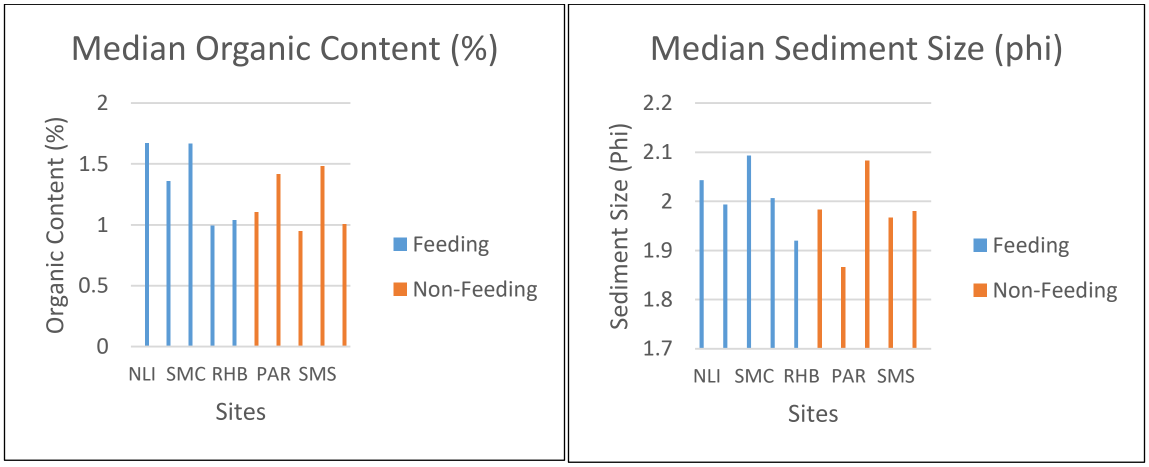

Fig. 16. Median sediment size, and organic content for feeding versus non-feeding sites.

Overall, no significant difference was found between either the mean sediment size or organic content of substrate inside and outside of feeding pit areas (Wilcoxon signed rank, $p>0.05$) (Fig. 17). However, several slight, though significant differences were found at several individual feedings sites when comparing the organic content and sediment size inside and outside each feeding area (Wilcoxon rank sum, $p<0.05$). Both Rhodesia Beach and Middle Sands had finer sediment inside feeding areas (median phi size of 1.92-2.0) than outside (median phi size of 1.96-1.99), while both Stony Point and South Mill Channel had higher median substrate organic content inside feeding areas (1.2-2.6%) than outside (1.0-1.7%). Higher median sediment sizes were found at sites with more exposure to wave energy. Distance from the deeper channels was also a factor as the higher currents there can strip away finer material and leave larger grains behind (Andrews 1965).

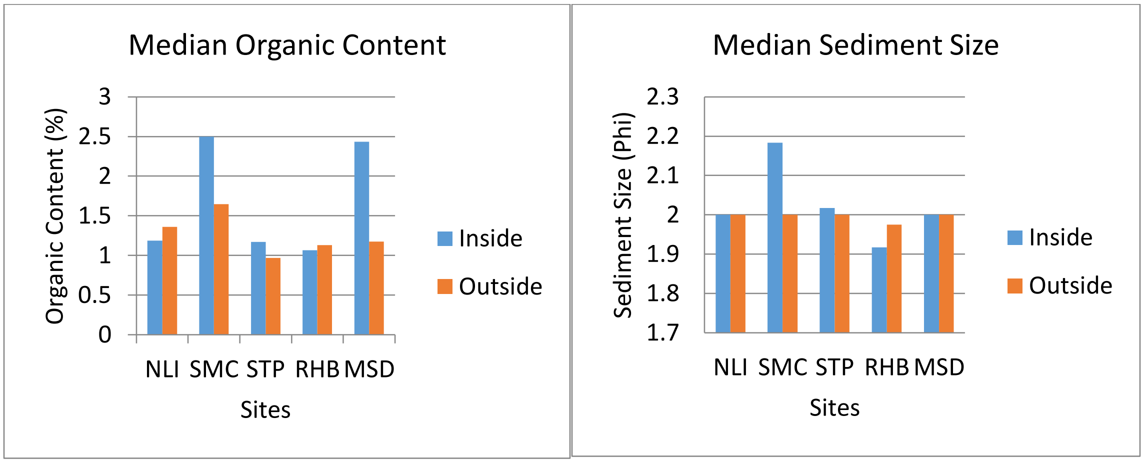

Fig. 17. Comparison of median sediment size and organic content for sediment inside and outside feeding pit areas at each feeding site.

There was no significant correlation between organic content and pit density overall (Spearman rank p>0.05). However, there is an interesting link between higher organic content and higher feeding pit density evident at South Mill Channel which had a pit density of 217 pits/ha and a median organic content of 1.6% (Tables, 2 and 4; Figs. 9 and 16). Higher organic substrate content is typically indicative of lower wave energy environments and active bio-accumulation zones, mechanisms that contribute to this organic build-up include intertidal vegetation, proximity to turbulent channels and input of marine organic matter. Common examples of bio-accumulation zones include saltwater marshes, eddies, slack water areas, and areas protected from erosion by jetties or natural coastal configuration. The pits may also have been more easily identified at this site because of organic matting present around the pits due to the high amount of decomposing organic material and fine grained sediment. This extends their longevity between tidal cycles, while also increasing their visibility.

Sites were paired based on their presence/absence of feeding pits combined with proximity between geographic locations to determine any significant differences in substrate characteristics (Wilcoxon rank sum, $p<0.05$). South Mill Channel and Middle Sands had significant higher percentages of organic content and finer average sediment grains sizes (2 to 2.08 phi) than their paired non-feeding sites (Fig. 18) and were the highest density feeding sites. Organic content between paired sites were higher at feeding sites with the exception of North Long Island (feeding 1.0-2.6% vs. non-feeding 1.0-1.5%). Mean sediment grain size was also finer among paired feeding sites in all cases except one (Rhodesia Beach) where the sediment was coarser (1.92 phi) than its counterpart Grassy Island (1.98 phi). This site was also a low density feeding site and there were no pits found during the late summer sampling period. This analysis also illustrates the higher variability of organic content between paired sites within Willapa Bay, as compared to limited variability in mean sediment grain sizes which ranged between 1.81 to 2.38 phi.

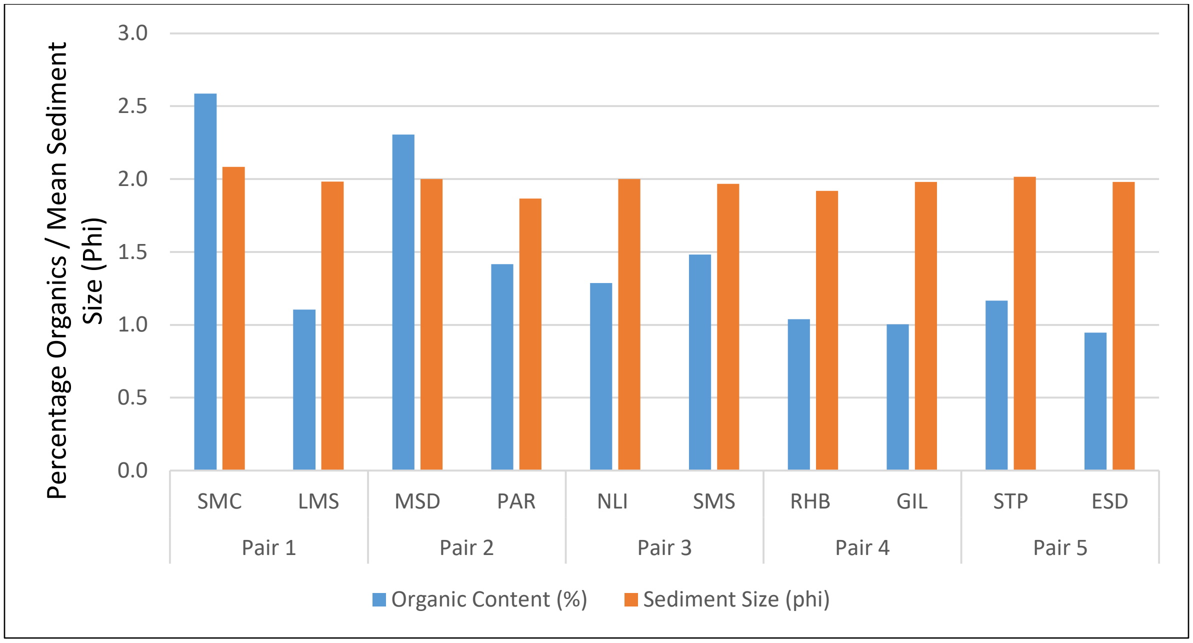

Fig. 18. Sediment characteristics of paired feeding/non-feeding sites.

Shrimp Cores

The number of shrimp collected in cores from feeding sites showed significantly high variability between sites in both early and late summer, ranging from 0.7 to 5.2 mean numbers of shrimp (Table 5, Fig. 19)(Kruskal-Wallis, $p<0.05$). However, differences between shrimp collected inside and outside of pits were statistically insignificant (Wilcoxon Rank Sum $p>0.05$) within each sampled site. Shrimp counts outside of feeding pits were slightly higher at most sites with the exception of Stony Point in early summer. The median shrimp count inside cores among feeding sites was 2.2 while the median count outside was 2.7.

	Early Summer				**Late Summer**			
Site	Inside		Outside		Inside		Outside	
	Count	Length (mm)	Count	Length (mm)	Count	Length (mm)	Count	Length (mm)
NLI	2.2 (1.7)	67.6 (12.4)	3 (1.4)	69.0 (16.5)	2.1 (1.4)	64.7 (14.6)	2.5 (1.8)	67.8 (13.9)
SMC	0.8 (0.7)	61.9 (7.8)	1.0 (1.0)	64.8 (7.4)	0.7 (1.1)	69.0 (5.08)	0.9 (1.2)	68.5 (5.8)
MSD	2.5 (1.6)	67.4 (16.5)	2.9 (1.4)	68.7 (12.5)	2.7 (1.6)	59.8 (22.0)	2.8 (1.5)	56.6 (24.0)
STP	4.1 (1.8)	64.5 (19.6)	4 (2.0)	60.4 (22.2)	4.8 (2.0)	65.2 (18.3)	5.3 (2.4)	68.1 (17.1)
RHB	1.4 (1.6)	53.7 (18.1)	1.8 (1.9)	50.2 (18.2)	NA	NA	NA	NA

Table 5. Characteristics [mean (SD)] of ghost shrimp at green sturgeon feeding sites, Willapa Bay, summer 2016.

Differences in shrimp counts between sites did not correlate to the number of pits found at each site (Spearman rank, $p > 0.05$). For example, South Mill Channel (SMC) had the lowest median shrimp counts (0.7-1.0) but had the highest concentrations of feeding pits (62-102 pits/ha). Similarly, Stony Point (SP) had the highest shrimp counts (4.0-5.3) but also a very low pit densities in both early and late summer (16-32 pits/ha). These results may also indicate some localized effects of predation where higher levels of predation, as indicated by feeding pit densities, may limit numbers, and vice versa.

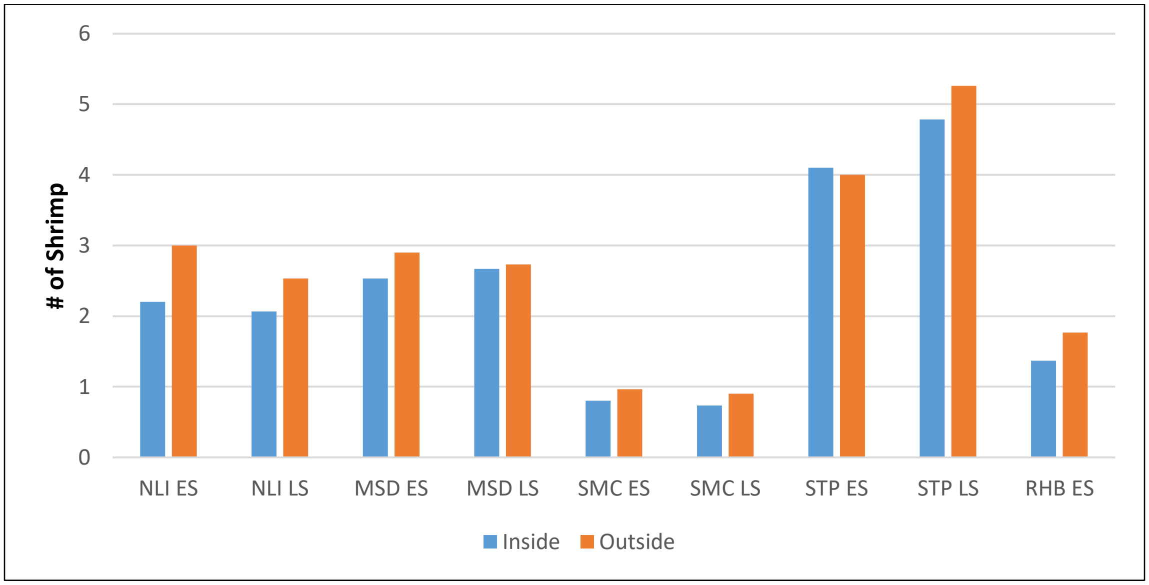

Fig. 19. Mean shrimp counts inside and outside feeding pits in early summer (ES) and late summer (LS).

The mean lengths of ghost shrimp collected from cores at feeding sites were predominately between 50 and 70 mm, and again showed significant variability between sites, both in early and late summer (Kruskall Wallis, $p < 0.05$) (Table 5, Fig. 20). Significant differences in ghost shrimp lengths were found inside and outside of feeding pits between sites in the early summer (mean lengths inside ranged from 53.7 to 67.6 mm and outside from 59.8 to 67.6 mm in the early summer), as well as outside the feeding pits between sites in the late summer (mean lengths outside ranged from 50.2 to 68.9 mm) (Kruskall Wallis, $p<0.05$). This shows that shrimp lengths were varied between sites in both seasons and all sites are home to adult ghost shrimp populations, although no correlation was found between shrimp length and feeding pit density (Spearman Rank $p>0.05$). Mean length inside and outside feeding pits had a low variability of between 1 to 3 mm showing that the shrimp at each site had similar growth and maturity characteristics that were not being significantly affected by sturgeon feeding behavior (Wilcoxon rank sum, $p>0.05$).

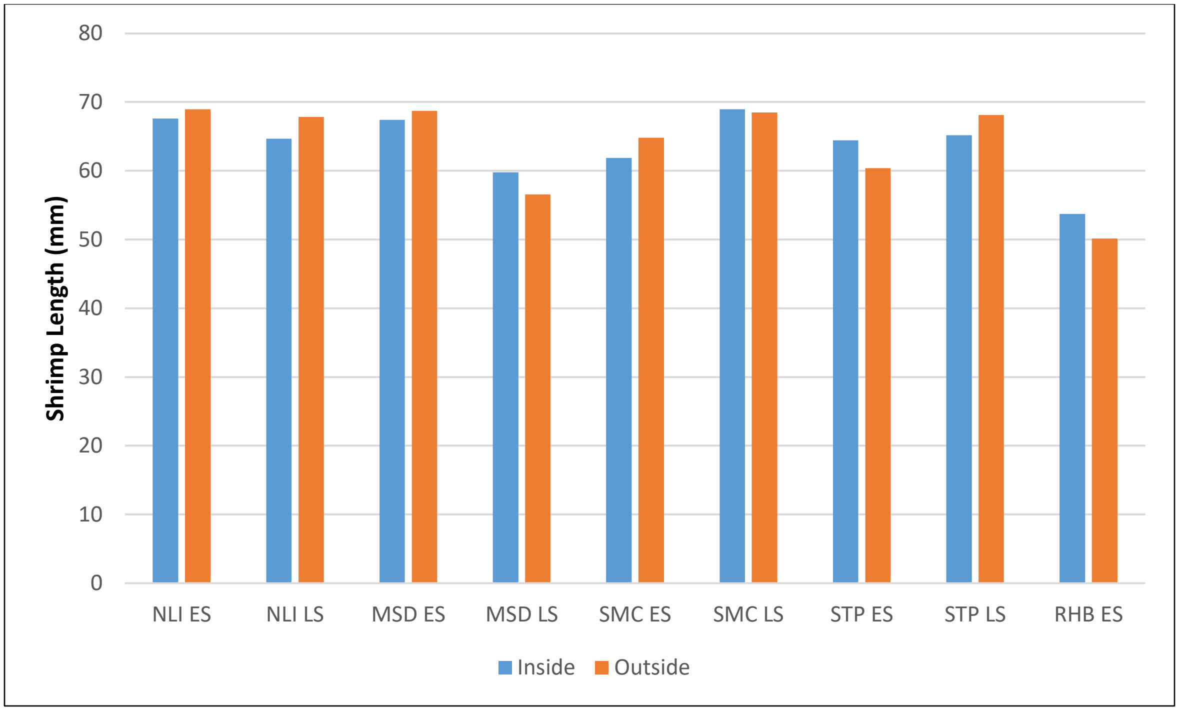

Fig. 20. Mean lengths of ghost shrimp collected by coring at feeding sites (ES = Early Summer. LS = Late Summer)

Shrimp Transects

Based on the available transect data, feeding sites did have significantly higher median shrimp counts (13.8) than the non-feeding sites (3.7) (Wilcoxon rank sum, $p<0.05$) (Table 6, Fig. 21). Median ghost shrimp lengths again ranged between 50 and 70 mm, though were significantly larger at feeding (62.9 mm) than non-feeding sites (59.2 mm)(Wilcoxon rank sum, $p<0.05$). This shows that the feeding sites, according to the transect data, have a higher average concentration of burrowing shrimp and they are on average slightly larger than those found at the non-feeding sites. However, only two feeding sites were surveyed using this method due to time constraints, limiting comparable results.

Feeding Sites					**Non-Feeding Sites**				
	Count		Length			Count		Length	
	Median	IQR	Median	IQR		Median	IQR	Median	IQR
Rhodesia Beach	12	5	65	13	Little Middle Sands	1	1	63	10
Stony Point	17	6.5	68	13	Shovel Middle Sands	2	2	60.5	14.25
					Parcel A	4	4.5	64	11.25
					Ellen Sands	6	7	55	26

Table 6. Shrimp characteristics from available sampling transect data for a subset of non-feeding and feeding sites.

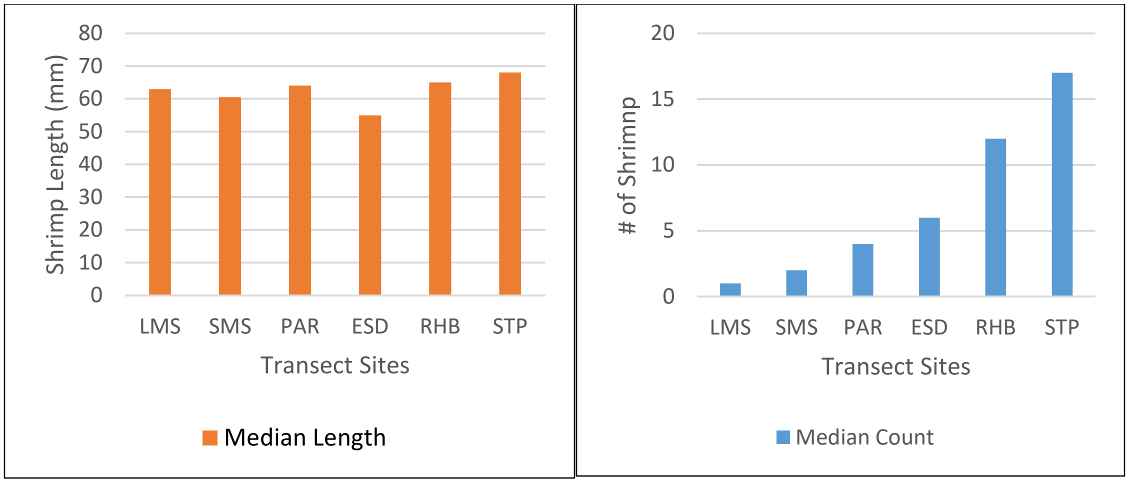

Fig. 21. Median ghost shrimp lengths and counts collected from transect sampling.

Eelgrass Data

The median shoot densities and lengths of *Z. marina* varied significantly between the sites (Kruskall Wallis, $p<0.05$) (Fig. 22). The median shoot density was 1.8 shoots per quarter meter, ranging between 0 to 10, while overall median shoot length was 21.6 cm, ranging from 19 to 24.1 cm, *Z. marina* had the highest counts at Parcel A (median 3.0 shoots per quarter meter), when excluding the results from Stony Point, which had very dense beds of *Z. marina* over 200 m away from the nearest feeding pit. The lowest counts were found at North Long Island. Parcel A had the longest *Z. marina* shoots (median 24.1 cm) while Little Middle Sands had the shortest shoots (median 21.0 cm).

Z. japonica density did not vary significantly between sample sites, with an overall median density of 22 shoots per quarter meter, ranging from 17 to 77 (Fig. 22). Little Middle Sands (LMS) had the highest density of *Z. japonica*, with a median count of approximately 77 shoots per quarter meter while the lowest density of *Z. japonica* shoots was found at Shovel Middle Sands, with a median of 17, ranging from 0 to over 100 shoots per quarter meter. By comparison, shoot lengths of *Z. japonica* varied significantly between the sites, with an overall median of 6.1 cm, ranging between 5 and 10 cm (Kruskall Wallis, $p<0.05$). The longest *Z. japonica* shoots were found at Parcel A (median 9.1 cm) while the shortest shoots were recorded at Shovel Middle Sands (median 5.0 cm).

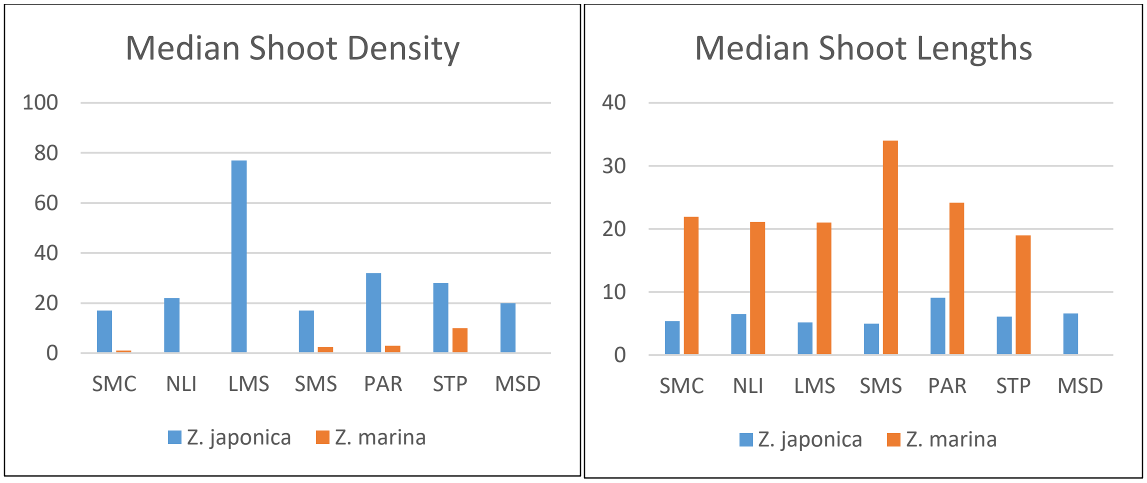

Fig. 22. Eelgrass (*Zostera marina* and *Zostera japonica*) characteristics at sample sites (median shoot density and length).

Distance from Pits to Eelgrass Beds and Aquaculture

Feeding pits were often found above the upper limit of *Z. marina* and the lower limit of *Z. japonica* (Table 7, Figs. 2 -24). All feeding sites had some *Z. marina* encroaching into the tidal flat area from deeper channels that remained submerged during most low tides (Figs. 10-14). Median feeding pit distances from the upper edge of *Z. marina* in the early summer averaged 34.8 m, ranging between 13.4 m and 56.0 m (Table 7, Figs. 23-24). . In the late summer, median feeding pit distances from the upper edge of

Z. marina averaged 84.8 m, ranging between 8.3 m and 116.5 m. North Long Island had pits closest to the upper edge of *Z. marina* (8.3-13.4 m) while Middle Sands was furthest away (56.0-116.5 m). Only two sites had *Z. marina* beds present shoreward of feeding pits, and feeding pit distances varied significantly between the two (Wilcoxon rank sum, p<0.05). Of these two sites, the median feeding pit distance at North Long Island was 25.4-46.5 m from the upper *Z. marina* edge, while the median distance of feeding pits was higher at South Middle Channel, ranging between 40.8 and 57.4 m during the summer.

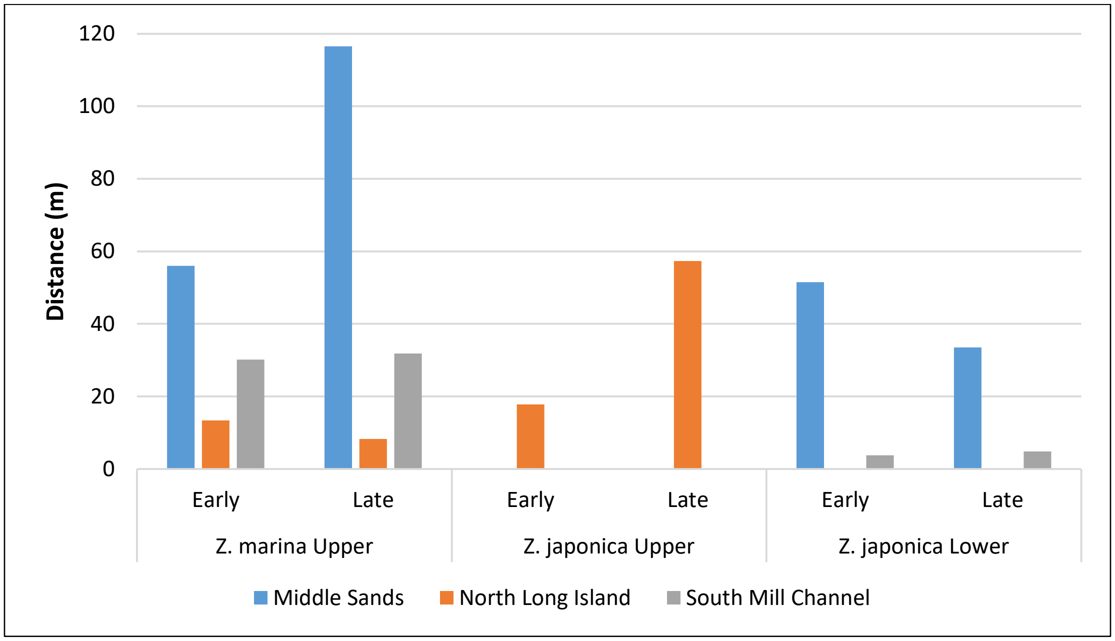

Fig. 23. Median distance of feeding pits outside eelgrass limits, summer 2016.

From Outside Edge (m)	Z. marina Upper Edge		Z. japonica Upper Edge		Z. japonica Lower Edge	
Early Summer	*Median*	*IQR*	*Median*	*IQR*	*Median*	*IQR*
MSD	56.0	60.9			51.5	19.5
NLI	13.4	6.4	17.7	27.2		
SMC	30.2	26.9			3.7	5.3
STP	23.3	11.2				
Late Summer						
MSD	116.5	112.5			33.5	42.8
NLI	8.3	16.9	9.6	11.9		
SMC	31.8	23.2				
STP	92.6	127.0				
From Inside Edge (m)						
Early Summer						
NLI	46.5	52.9	57.3	36.1	110.0	50.6
SMC	57.4	48.1			53.6	51.7
Late Summer						
NLI	25.4	33.6	18.4	34.2	111.2	31.8
SMC	40.8	44.7			39.2	42.0
Aquaculture Limit	Early Summer		Late Summer			
	Median	*IQR*	*Median*	*IQR*		
MSD	63.9	13.3	50.9	22.1		
SMC	36.1	29.8	103.1	89.9		

Table 7. Distance of feeding pits to different types of eelgrass bed and aquaculture edges.

When present, *Z. japonica* was generally found landward of feeding pit areas at three out of four sites, as it typically inhabits a higher tidal elevation zone than *Z. marina* (Table 7, Figs. 23-24). The median distance of feeding pits above the lower limit of *Z. japonica* beds ranged between 39.2-53.6 m (South Mill Channel) to 110.0-111.2 m (North Long Island) during the summer, with the notable exception of Stony Point, where the feeding area was over 380 m away from the nearest *Z. japonica* bed and therefore not included in either the summary table or graph (Figs. 10-14). While many pits were found within sparse beds of Z. japonica at North Long Island, the highest concentrations of pits observed at North Long Island were located in bare areas between the denser beds of eelgrass, with median distances of 18.4-37.3 m below the upper edge of *Z. japonica* (Figs. 12 and 24). North Long Island was also the only site with feeding pits above the upper limit of *Z. japonica*, with median distances ranging between 9.6-17.7 m. Almost all the pits at Middle Sands were similarly found in bare areas waterward of *Z. japonica* beds (Fig. 11). Middle Sands was also the principal site where feeding pits were found seaward of the lower limit of *Z. japonica* with median feeding pit distances from the lower edge ranging 33.5-51.5 m throughout the summer. There were also large eelgrass beds at several of the non-feeding sites which are displayed in maps found in the appendix (B-C).

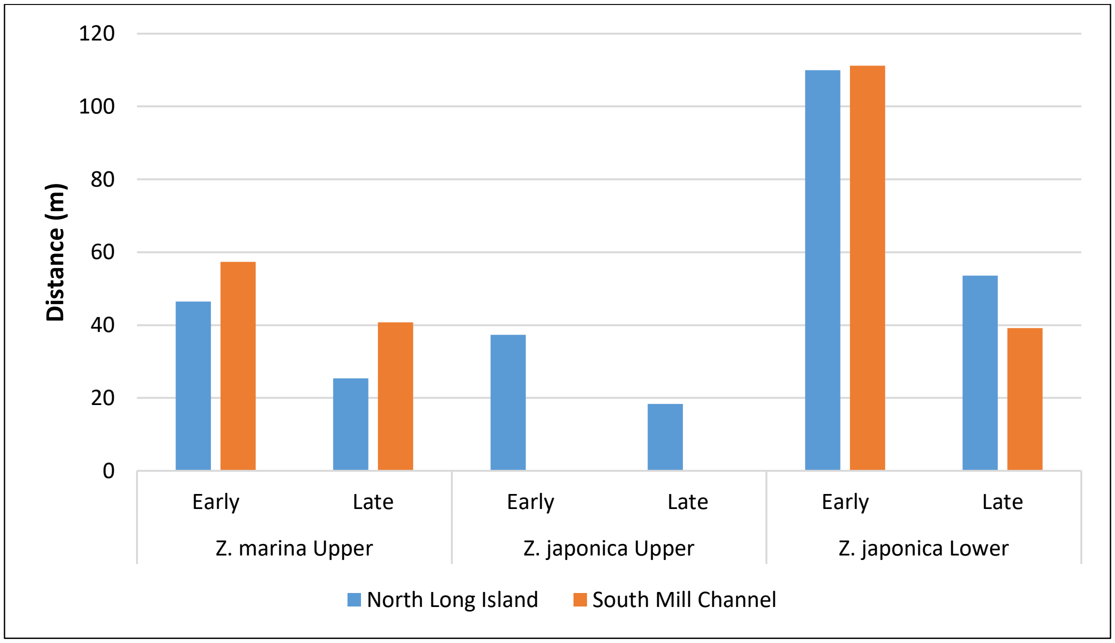

Fig. 24. Median distance of feeding pits inside eelgrass limits, summer 2016.

Distance to aquaculture operations was measured at Middle Sands and South Mill Channel, both of which had oyster beds adjacent to the south of the sturgeon feeding area (Figs. 10-11). Middle Sands had some of the pits closest to aquaculture during the early summer (median 13.3 m) and late summer (median 22.1 m) (Fig. 25). The distance of feeding pits from aquaculture at South Mill Channel was similar in the early summer (median 29.8 m) but pits were much further away during the late summer period (median 103.1 m). (Wilcoxon rank sum $p<0.05$). The close proximity of pits to eelgrass beds at South Mill Channel is further evidence of the constriction to feeding areas created by dense eelgrass beds and aquaculture operations (Fig. 10). No feeding pits were observed within the borders of staked oyster beds visible from the two sample sites, likely due to the obstructions offered by the stakes and lines, as well as the hard/sharp oyster shells littering the substrate. Rhodesia Beach and Stony Point were the two feeding sites farthest away from aquaculture operations due to their location in the northeastern part of the bay. Non-feeding sites with aquaculture nearby included: Parcel A, Little Middle Sands, and Shovel Middle Sands (App. B-C).

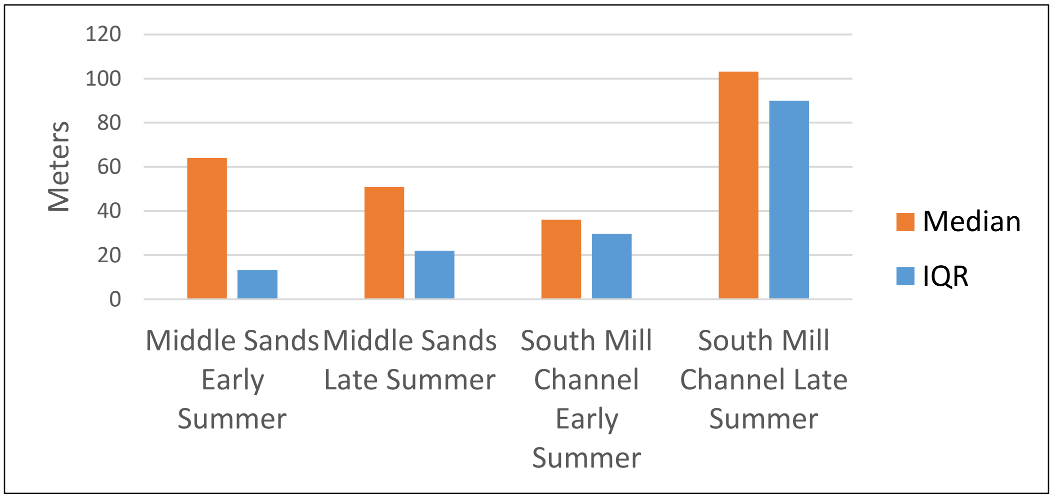

Fig. 25. Feeding pit distance to aquaculture, 2016.

Video Capture Results

Cameras were set out six times during the study, recording over 78 hours of footage. This footage was reviewed and no green sturgeon were observed by the camera arrays. Bait cannisters attracted large numbers of crabs and sculpins but did not entice any of the target fish into frame. As no recording of green sturgeon feeding has been captured before, there was limited guidance on ideal timing, but new pits were observed in close proximity to the cameras upon retrieval. Since no sturgeon appear in the video, they had to enter and leave within eight minutes before the cameras recorded another two minute interval or they visited the site later after the maximum recording time of the array.

CHAPTER VI

CONCLUSION

At the outset of this research there were several objectives in mind:

1) mapping the spatial distribution of green sturgeon feeding pits at several sites in relation to native and non-native eelgrass species;
2) sampling of burrowing shrimp populations to determine potential difference in forage availability and feeding impacts; and
3) comparing other environmental factors such as elevation, sediment characteristics and proximity to eelgrass and aquaculture to determine potential controls of feeding patterns.

Sturgeon aggregations are occurring in Willapa Bay on an annual basis and their feeding and migration patterns are observable as stated in previous research (Erickson and Hightower, 2007, Moser et al. 2016). Green sturgeon are feeding in certain areas of Willapa Bay while completely avoiding others, despite the consistent presence of burrowing shrimp (Dumbauld et al. 2008; Moser et al. 2007). The results of this study confirm several of the conclusions drawn by the NOAA study on green sturgeon in Willapa Bay (Moser et al. 2017). First of all, it does appear that these fish prefer to feed in finer substrate and areas lacking dense *Z. japonica* or *Z. marina* beds. Although a few feeding pits were found within these beds, they were usually in sparsely colonized areas of the beds. Less than 30% of the overall pits found were within mapped eelgrass boundaries. We also did not observe any feeding activity within the oyster beds around active feeding sites, showing that they seem to avoid these environments completely when searching for prey but do feed prevalently in areas adjacent to these beds. Although less than 30% of feeding pits were found within eelgrass beds, all feeding sites with the exception of Stony Point had eelgrass beds in close proximity to the feeding area. The production of organic material and the potential refuge function eelgrass beds provide to intertidal organisms, likely contribute to the increased shrimp numbers and sturgeon feeding activity at these sites.

Sediment distributions found throughout Willapa Bay, are dependent on the proximity to accreting river deltas from the many freshwater rivers that empty into the estuary as well as the tidal depth which the sample was collected. Tidal flats have the potential to accumulate the finer-grained sediments preferred by green sturgeon as feeding habitat. This may be a contributing factor in their selection as feeding sites and the exclusion of deeper sub-tidal areas (Moser et al. 2017). Feeding sites had slightly smaller average sediment grain sizes than non-feeding sites (feeding 1.99 phi and non-feeding 1.94 phi). This was especially distinct at the less exposed, southernmost site in the estuary, South Mill Channel, which had the highest silt content (max 2.38 phi) and the highest concentration of feeding pit activity recorded during this research. Green sturgeon are also feeding in substrates with higher organic content, as four out of five feeding sites had slightly higher organic content percentages (median 1.3%) than their non-feeding counterparts (median 1.2%). The sturgeon seem to be drawn to particularly fertile tide flats, where plant and animal material have built up in varying states of decay. Since ghost shrimp are known to feed on organic detritus, this factor could be drawing a higher corresponding amount of feeding activity. The turbidity caused by burrowing shrimp can also limit eelgrass beds by reducing light for photosynthesis and burial of young shoots (Dumbauld et al. 2003).

Goldsworthy (2007) asserted that feeding by green sturgeon in inter-tidal areas occurred rapidly and opportunistically and this seems to coincide with the findings of this study in that new pits would appear after just one tidal cycle and old pits were washed out rapidly by wave and tidal action. In Willapa Bay, sturgeon feeding pits declined with tidal elevation with few feeding pits extended into neighboring deeper channels that remained submerged during low-tide conditions (Moser et al. 2017). Most of the recorded pit locations had an average tidal elevation between 0.5 and 1.0 m MLLW. This assessment could not be confirmed with the use of sub-tidal acoustic imaging such as was used by NOAA, but the pits did taper off visually before being unobservable below the turbid waters. This trend can also be seen in Table 2 where feeding pits increased with shallower tidal elevations at four out of five feeding sites. This trend coincides with the findings of previous studies that mapped feeding locations within Willapa Bay (Dumbauld 2008; Moser et al. 2017).

Moser (2017) determined that feeding pit densities were highest at their sampling sites during these months was also confirmed by this study. Moser (2017) concluded that temperature and prey availability both affect the seasonal use of estuaries such as Willapa Bay. The prevalence of pits at sites closer to the southern end of Willapa Bay during July and August is also represented in both studies. Average monthly pit densities collected by Moser (2017) ranged from a low of 7-36 pits/ha at Nahcotta North site to a high of 95-1042 pits/ha at Mill Channel. The location of these sample sites are most comparable to North Long Island (41-106 pits/ha) which is at approximately the same latitudinal position as Nahcotta North. Data collected at South Mill Channel were also the highest pit densities collected in this research (62-217 pits/ha).

Moser (2017) found that the highest concentrations of feeding pits occurred in soft muddy substrates with no eelgrass and that pit densities declined rapidly in *Z. japonica* beds. Their findings coincide with the results of this study which also recorded more pits in bare areas or where *Z. japonica* beds became patchy. Moser (2017) also did not find any evidence of sturgeon feeding pits in nearby oyster beds. Several explanations for the lack of pits in aquaculture areas include: hardening of the bottom, frequent disturbance of the site, and gravelling of beds which restricts access to their prey (Moser et al. 2017, Dumbauld et al. 2001). The sediment stabilization created by dense beds of *Z. japonica* deters feeding activity, as shown by Moser (2017) where experimental removal of Z. japonica was conducted and pit densities increased in these treated areas. Seasonal variations in feeding activity was also linked to this aversion to Z. japonica beds as pit densities declined at sites as non-native eelgrass grew and expanded over the summer.

An interesting similarity between shrimp size was apparent in the coring results. Shrimp from all feeding and non-feeding sites averaged between 50 to 70 mm in length, showing that conditions at all of these sites are conducive to ghost shrimp growth and reproduction, and the presence or absence of feeding pits could not be predicted by the presence or absence of the prey species. As noted by Moser (2017), the removal of shrimp did not significantly impact pit formation. Therefore the observed differences in feeding pit activity are likely linked to other site conditions.

One conclusion by Moser (2017) that does not align with the findings of this study are that sturgeon feeding areas followed the areas of highest ghost shrimp density, and that this factor could account for 75% of the variability in pit density. The presence of burrowing shrimp is not necessarily an indicator of sturgeon feeding habitat. Transect data shows that all of the non-feeding sites also had shrimp in significant numbers when sampled, though lower than feeding sites. Some of the highest feeding pit densities were found at sites which did not have the highest shrimp counts (e.g. SMC median shrimp count 0.85, NLI median shrimp count 2.1), but did possess fine-grained substrate and an area bereft of eelgrass where feeding activity was concentrated. Stony Point is an interesting example where shrimp numbers were very high, and there was no eelgrass present but the pit numbers were still very low. The substrate at this site was sand, but a similar substrate was found at Middle Sands where a large number of pits were located. The northernmost, exposed location of Stony Point and the lack of nearby aquaculture may have had an effect on the low number of pits, since the highest density feeding sites were at the south end of the bay and in close proximity to oyster beds. This could have affected the survey because Stony Point is in the direct path of higher energy wind-driven waves coming in off the Pacific Ocean and entering the bay, thereby removing evidence of feeding activity quickly, while the aquaculture or eelgrass beds that could attenuate wave energy are also largely absent.

Our results suggest that sturgeon feeding behavior does not significantly affect the number of ghost shrimp present at feeding sites. Larger numbers of these predators may have had a slightly larger impact in the past, though the dramatic rise in ghost shrimp populations throughout the bay are less related to the decline of this species but more likely due to the change in temperatures cited by previous studies (Dumbauld 2008).

Goldsworthy (2007) concluded through telemetry data that the fish were using small channels adjacent to aquaculture beds to access higher intertidal areas. We found the highest concentrations of feeding pits at sites such as South Mill Channel, Middle Sands, and North Long Island, all of which have aquaculture beds immediately adjacent to the study area. These oyster rearing areas function to obstruct the flow of water off the tide flats at low tide and increase development of small channels in the substrate at the margins of these beds.

The findings by Van Eeneennaan (2001) that green sturgeon show increased activity during hours of darkness could provide some justification for the lack of sturgeon feeding caught on our camera arrays, as they were only viable during daylight and did not have the capability of nocturnal observation. Future efforts to capture green sturgeon feeding activity on video should employ night vision cameras and operate during hours of darkness if the cameras are sufficient to detect sturgeon through the turbid waters using this technology.

Recommendations

Our major conclusions are that areas of fine-grained substrate with no eelgrass seem to be of particular value as feeding habitat for green sturgeon, and should be allowed to remain in their current state. Areas with relatively larger grain-size material might be of less value, especially if inhabited by thick beds of *Z. marina*. Areas with finer average sediment sizes are more problematic for the operation of oyster aquaculture which means there may be continued opportunities for feeding habitat conservation in these locations. Some oyster growers are also abandoning certain plots that have become too muddy for their purposes and these may become prime feeding areas in the future as the shells breakdown and the shrimp populations continue to grow, if they are not colonized by non-native eelgrass beds.

In terms of the tidal elevation conservation and habitat improvement efforts, these should be focused between 0.5 and 1.0 m MLLW where the vast majority of feeding pits were observed. This may be a source of conflict within Willapa Bay, as that same tidal elevation band is also prime habitat for oyster rearing which is one of the main economic activities of the area.

As illustrated in this study and previous literature (Moser et al. 2016) the absence of *Z. japonica* enhances the feeding habitat for green sturgeon, and spraying efforts by aquaculture operators could therefore reduce the prevalence of this non-native eelgrass species. The 10 m buffer currently used to protect native *Z. marina* should be maintained, but could be difficult to implement in areas where the two species are sporadically intermixed. Removal of non-native eelgrass beds could proceed not only in aquaculture beds, but in the surrounding tidal flats. Since sturgeon feeding activity has not been observed within aquaculture beds there would be limited impact to sturgeon feeding habitat from the removal of *Z. japonica* in these zones. Rather, areas directly adjacent to aquaculture seem to be prime feeding habitat if they remain un-vegetated.

Spraying of pesticides to remove ghost shrimp from areas deemed desirable for aquaculture does not seem to currently limit available forage for green sturgeon. Spraying in areas with observable feeding pits should however be avoided, because the sturgeon would be present immediately after application during the high tide and potentially ingesting the affected shrimp. Preferred conditions for spray application would be during spring, before the sturgeon aggregation period, or at tidal elevations above 0.26 m MLLW, where feeding activity predominates (Tables 2-3) especially near the small tidal channels used by green sturgeon to access feeding areas. The spring months would also be best for reducing burrowing shrimp populations as they have not yet released their eggs (Dumbauld et al. 2008).

Green sturgeon will continue to aggregate in Willapa Bay during summer months to feed on burrowing shrimp in the tide-flat environment. Efforts to maintain this habitat must address the colonization of non-native seagrass and the careful management of aquaculture beds to allow foraging opportunities to continue in the areas where feeding pits have been documented.

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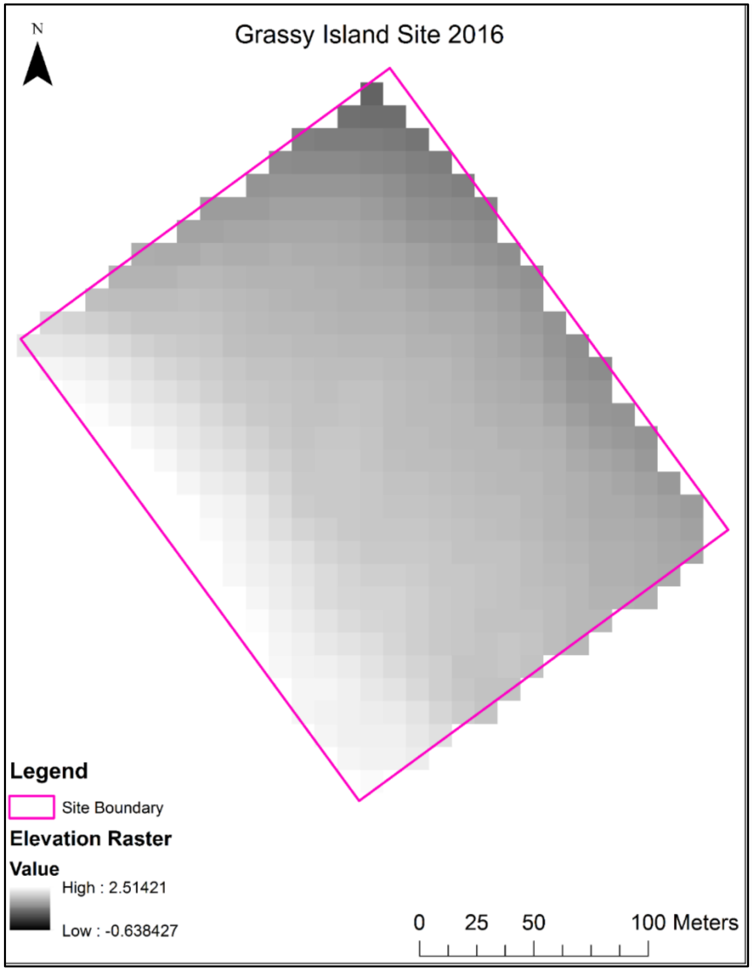
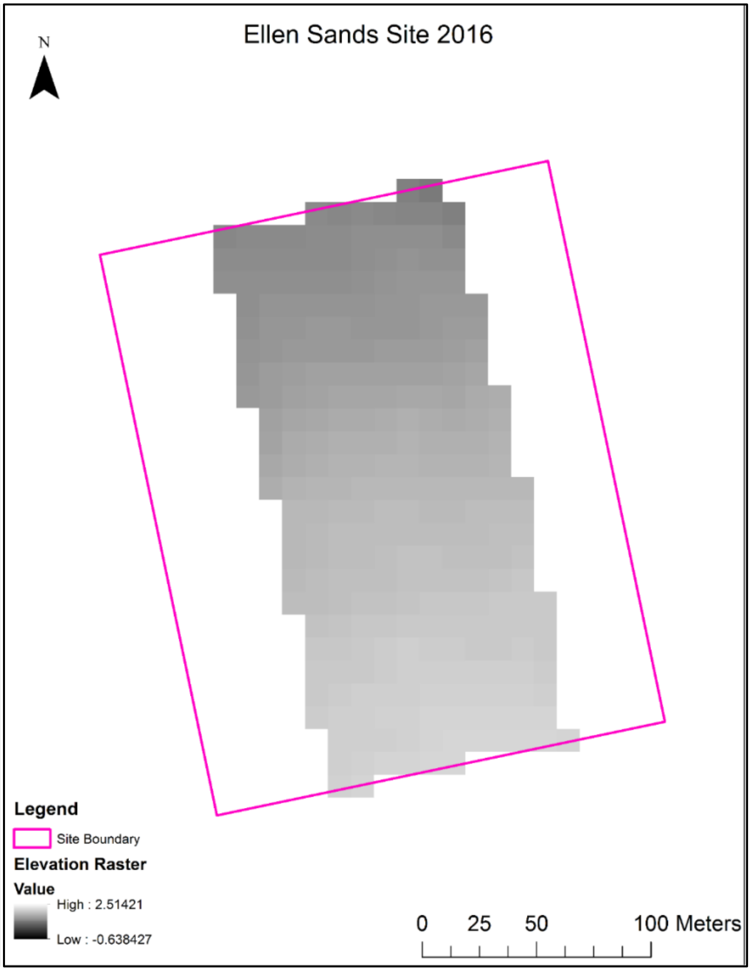
APPENDIXES

APPENDIX A

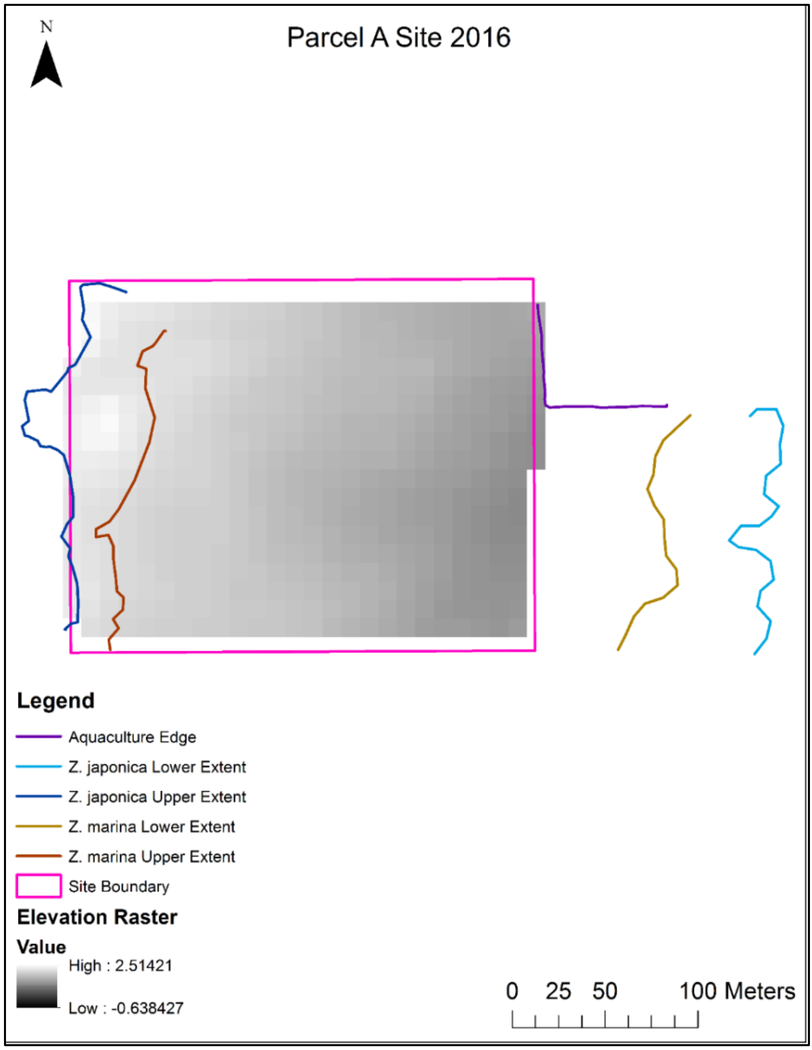
GRASSY ISLAND AND ELLEN SANDS SITE MAPS

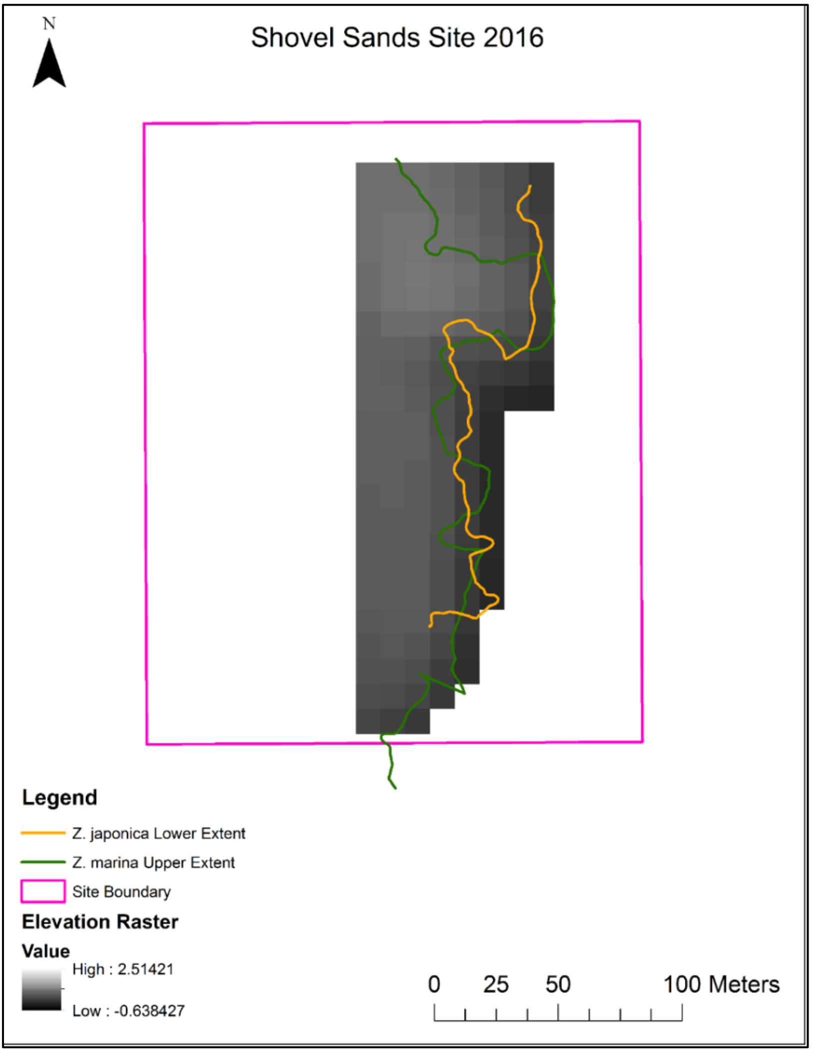




APPENDIX B

PARCEL A AND SHOVEL MIDDLE SANDS SITE MAPS





APPENDIX C

LITTLE MIDDLE SANDS SITE MAP