Washington Department of Fish and Wildlife Plan for Rebuilding Olympia Oyster (*Ostrea lurida*) Populations in Puget Sound with a Historical and Contemporary Overview

Brady Blake and Alex Bradbury
Washington Department Fish & Wildlife
Point Whitney Shellfish Laboratory
1000 Point Whitney Road
Brinnon, WA 98320

I. Introduction

The Olympia oyster (*Ostrea lurida:* Carpenter 1864; = *Ostreola conchaphila*) is Washington's only native oyster. It currently exists throughout almost the entirety of its historic (circa 1850) range in Puget Sound, but its overall overall abundance has diminished since that time. Native oysters are rarely found in large, naturally occurring expanses of biogenic habitat commonly referred to as "oyster beds." Prior to the initiation of commercial exploitation around 1850, native oysters occurred widely along intertidal shorelines of Puget Sound's interior, but only at a limited number of locations as "oyster beds." These beds occurred where the specific physical conditions were amenable to the formation and persistence of that particular type of habitat structure (Galtsoff 1929; Gillespie 1999). In some rare circumstances, subtidal beds were present in association with large intertidal populations.

Exploitation of those rich intertidal deposits of oysters from 1850 to the early 1900s resulted in near extirpation of large natural beds, both by harvest and conversion to intensive cultivation. Both harvest and cultivation eliminated decades of natural shell accumulation and living oysters that formed oyster beds which in some cases were described as being up to a foot thick in depth. The resulting conditions for wild stocks of native oysters were further exacerbated beginning in the latter portion of commercial exploitation by additional stressors: Further habitat alterations, water pollution, invasive predators, alternative uses and values for historic habitat footprints (i.e. aquaculture and eelgrass). Shoreline and watershed developments further diminished the presence of natural beds and the overall abundance of the oyster in Puget Sound, along with restricting its ability to naturally rebuild beds and abundances.

Since 1997, Olympia oysters have been listed by the Washington Department of Fish and Wildlife (WDFW) as a State Candidate species. State Candidate species are those that WDFW may review for possible future listing as State Endangered, Threatened, or Sensitive species. The Olympia oyster is also included in WDFW's Priority Habitats and Species (PHS) List, a catalog of habitats and species considered a priority for conservation and management; all State Candidate, Endangered, Threatened and Sensitive species are automatically included in the PHS List. Puget Sound wild stocks of *O. lurida* are not at this time exploited for commercial or recreational harvest, although they may be subject to treaty tribe ceremonial and subsistence harvest. For management purposes, *O. lurida* is statutorily classified by WDFW as "shellfish" (WAC 220-12-020) and is passively protected from recreational harvest on public tidelands by a rule that prohibits recreational harvest of any oyster less than 2.5 inches in shell size (WAC 220-56-310). Annual Bivalve Management Plans between the State and Puget Sound treaty Tribes also indirectly protect native oysters on public tidelands from tribal commercial harvest with the same 2.5 inches minimum shell size restriction. Mortality incidental to commercial harvest of Pacific oysters (*Crassostrea gigas*) on both public and private tidelands does occur, since Olympia oysters frequently set as larval spat on Pacific oyster shell.

Restoration efforts in Puget Sound were initiated following the development of WDFW's 1998 Olympia Oyster Stock Rebuilding Plan (Cook *et al*. 1998). The key actions cited in the 1998 Rebuilding Plan included development of survey methods, population inventories, natural restoration techniques, site selection criteria, genetic integrity investigations, water quality improvement, and habitat protection. The 1998 Rebuilding Plan was not funded as a WDFW project, however, and many aspects of the plan have been undertaken by the non-profit Puget Sound Restoration Fund (PSRF). The past 14 years of native oyster restoration in Puget Sound have been a collaborative partnership effort facilitated by PSRF involving government agencies, treaty tribes, shellfish growers, other non-profit organizations, private tideland owners, and volunteers. WDFW's primary role during this period has been to provide oversight for planning, implementation, research and as a source of historical and current information regarding *O. lurida*. During that time, a vast amount of knowledge has been gained through research, as well as the successes and failures of trial and error associated with a relatively youthful restoration effort.

II. Purpose Statement

This document is an updated revision of the Puget Sound portions of WDFW's Olympia Oyster Stock Rebuilding Plan (Cook *et al.* 1998). Revisions are based upon research and knowledge gained during the past 14 years of rebuilding *O. lurida* stocks. The purpose of this document is to inform and guide WDFW and partner organizations in efforts to restore native oysters in Puget Sound. This plan includes an historical review for native oysters and current information on restoration projects carried out since the 1998 Plan. As such, this document will continue to be expanded by advances in science and the results of on-going and future restoration projects.

WDFW co-manages shellfish resources, including Olympia oysters, with treaty tribes and the Washington Department of Natural Resources (DNR). This plan will provide valuable information, insights and guidelines for WDFW's shellfish co-managers -- as well as other groups collaborating in Olympia oyster restoration.

WDFW recommends a focused restoration strategy at 19 sites within Puget Sound by 2022. If successful, this strategy will result in the re-establishment of self-sustaining, large-scale, dense native oyster assemblages able to function as source populations. This strategy is recommended for both the biological conservation of the species and as a foundation for continued rebuilding of the species by natural or artificial means.

The primary objectives of this restoration strategy are the biological conservation of the species and its associated habitat. Those objectives are achieved by re-establishing, rebuilding and enhancing natural native oyster assemblages, ensuring the species' long -term persistence in the face of changing sea levels, temperatures, chemistry and the competing uses of Puget Sound marine waters and tidelands. The term "restoration" as used in this document should not be confused to mean "restoring shellfish beds for harvest" or "restoring water quality to make shellfish safe for human consumption."

Ecosystem services (e.g., increased fish use, increased species diversity) are not a primary objective of the restoration strategy, but are an expected benefit that will likely occur at each restoration site over time.

Re-establishing a wild stock fishery for Olympia oysters, if achievable, will likely be limited to provisioning historic tribal cultural needs. Re-establishing recreational wild stock fisheries on public tidelands is not an objective of this plan, and would conflict with achieving and maintaining biological conservation and ecological restoration objectives (Galtsoff 1929; Elsey 1933; Gillespie 1999; Gillespie

2009; Jackson *et al.* 2001; Kirby 2004; Powers *et al.* 2009; Schulte *et al.* 2009). A limited treaty commercial harvest may eventually be feasible in some cases, but such harvests are likely to be sustainable only under very conservative exploitation rates.

Restoring the full historical extent of Olympia oyster "beds" and abundances is of doubtful benefit and is probably not feasible. The estimated historical (circa 1850) footprints of *O. lurida* beds were often extensive (e.g.,≈ 500 acres in Lynch Cove and ≥ 2,000 acres in Samish Bay), although in recent years competing uses in many locations have reduced the restoration potential over such large tracts. Tidelands devoted to aquaculture and those containing eelgrass (*Zostera marina*) limit the potential for restoration in many areas. Additionally, the costs required for restoration -- sometimes as high as $50,000 per acre -- need to be considered when prioritizing and selecting restoration sites.

III. Background

Prior to the exploitation that began around1850, *O. lurida* was widely distributed throughout the interior of Puget Sound, and extended westward at least to Sequim Bay in the Strait of Juan de Fuca; these were part of the larger trans-boundary metapopulation that extended northward across the U.S.-Canadian border (Figure 1). Within that broad distribution range, the oyster occurred in dense biogenic assemblages, often described as "beds." Such beds occurred at a limited number of locations where specific physical conditions allowed formation and persistence of those large three-dimensional structured biogenic habitats (Meeker 1921; Prosch 1904; Galtsoff 1929; Gillespie 1999). Scattered widely throughout the extent of that historic metapopulation was a more common presence of individuals and small aggregations, wherever habitat conditions allowed. An exhaustive search and review of available information indicates that this historic metapopulation was predominantly located in the intertidal zone but there were rare subtidal occurrences where unique physical and biological conditions were present

Prior to European settlement, native oyster beds were utilized as an important local food source and for commerce by various tribes located near large intertidal beds. In several locations, tribes may have practiced methods of cultivation to make the oysters more readily available (Barsh and Murphy 2008; Dinnel 2011; Elmendorf 1992; Gunter and McKee 1960; Hatch *et al.* 2005; Suttles 1974; Steele 1957). In particular, tribal winter villages appeared to have been strongly associated with large, dense intertidal oyster beds. Tribes provided this local source of wild food to the new European immigrants in Puget Sound, and as those pioneers began commercially exploiting the native oyster resources themselves, those same natives often provided a source of knowledge and a labor force for harvest, cultivation, and transport. In some instances, tribes commercially exploited the natural beds themselves for rapidly developing local markets (Meeker 1905; Prosch 1906; Galtsoff 1929).

Commercial exploitation of the native oyster resources of Puget Sound by early pioneers began about 1850 with the discovery of large oyster beds. One reference cites 1852 as the first year of commercial harvest (Hines 1894), while two others mention 1851 as the initial harvest year (Bonnot 1935; Barret 1963). Six hundred bushels of oysters were shipped to San Francisco to meet the burgeoning demand by gold-seeking immigrants, a demand that could not be met by the limited -- and in some cases unsatisfactory -- local oyster resources available to them locally (Ingersoll 1881; Babalis 2009; Babalis

2011). Simple exploitation of the wild stocks, with little if any restraint, continued until around 1870, followed by the addition of rudimentary cultivation efforts and transfers of stocks within and between sub-basins. Marketing of fresh oysters in the shell increased within Puget Sound, east of the Cascades, and to ports in California, Oregon and British Columbia. By 1890, depletion of the available wild Olympia oyster stocks, plus market competition from both imported and cultivated Atlantic oysters (*Crassostrea virginica*), had resulted in a diminished oyster economy in Puget Sound.

Advanced cultivation practices (dikes) were introduced in about 1890, resulting in a revitalization and expansion of the Puget Sound oyster industry. This was driven by converting the remaining natural beds to artificial beds contained within diked impoundments, and maintained by intensive seed collection, supplemented by inter- and intra-basin seed transfers. As the oyster industry further expanded in scope, a growing desire for additional economic opportunities saw imports of *C. virginica* to Puget Sound (circa 1900) for cultivation on depleted native oyster beds, and trial attempts at *C. gigas* cultivation using imported adult oysters from Japan.

As the Puget Sound native oyster industry peaked in the 1920s, two critical events occurred almost simultaneously that would shape the future of the native oyster industry and transform oyster species as a component of the Puget Sound ecosystem. The first event was the 1927 startup of a Kraft (sulphite process) pulp mill located in Shelton, in the southern Puget Sound basin. The vast majority of the cultivation -based native oyster industry had consolidated itself in this region, due to overharvest elsewhere and the availability of rail transportation linking the industry to its markets. The sulphite waste liquor (SWL) released by this pulp mill into Hammersly Inlet immediately began affecting *O. lurida* throughout the southern Puget Sound basin inlets. Over the next thirty years, SWL pollution in varying degrees diminished annual reproduction, recruitment, abundance, growth, and oyster condition; in some locations, severe mortalities occurred (Steele 1957; Gunter and McKee 1960). SWL may also have led to increased blooms of the chain diatom *Melosira sp.* in several embayments. *Melosira* was reported to smother oyster beds and may have contributed to siltation of oyster habitat. The input of SWL in southern Puget Sound ceased in 1957, and an immediate resurgence in native oyster productivity was reported in the previously-affected embayments.

The second concurrent event was the initiation of large-scale cultivation of *C. gigas* in Puget Sound, which proved increasingly successful. Initially, *C. gigas* was seen as a viable option for "oyster" cultivation in northern Puget Sound, where *O. lurida* was no longer a feasible economic choice due to the absence of sufficient reproductive potential in local wild and cultivated stock remnants. Importation of *C. gigas* from Japan into Puget Sound, and inter-basin transfers of oyster stocks within Puget Sound brought new stressors for the native oyster: The predatory drill *Ocinebrellus inornatus (= Ceratostoma inornata)* and the flatworm *Koinostylochus ostreophagus* (= *Pseudostylochus ostreophagus*), along with the parasitic copepod pest *Myticola orientalis.* These non-native invasive species joined the predatory drill *Urosalpinx cinerea* and the pest slipper shell *Crepidula fornicata* , both of which had arrived with the earlier imports of *C. virginica* as stressors for cultivated native oysters and remnant wild stocks at several locations throughout Puget Sound. It should be noted that these invasive predators and pests were not distributed across all native oyster occurrences, and where they were present, their effects varied considerably.

The combined effects of SWL in southern Puget Sound, the non-native pests and predators imported from Asia, and economic factors related to the cultivated oyster industry all led to a wholesale switch to *C. gigas* cultivation. Only a handful of native oyster cultivation operations remain today, and no extant wild-stock harvest occurs at this time (Cheney and Mumford 1987). Much of the tidelands altered for native oyster cultivation were converted to commercial *C. gigas* production, while other tidelands were converted to the production of the Manila clam *Venerupis philippinarum* during more recent decades.

Of the large biogenic structured habitats ("beds") formed by native oysters circa 1850, it has been coarsely estimated that less than 5% currently remain in Puget Sound. A more rigorous definition of what constitutes a native oyster "bed" would likely result in a determination that this specific habitat has been functionally extirpated in Puget Sound. At this time, only one representative large-scale natural bed located in Case Inlet has been identified, although several smaller occurrences and at least two restoration sites may also be considered as having "oyster beds." Nevertheless, *O. lurida* as a species occurs widely and persistently throughout nearly all of its historic range in Puget Sound. Most of these oysters are found as dispersed individuals and assemblages in three of the five Puget Sound sub-basins. These oysters cumulatively appear to meet the definition of *source populations,* despite the fact that "oyster beds" are rarely present. In the central reach of Hood Canal and portions of southern and central Puget Sound, the native oyster is a commonly encountered intertidal species, and is apparently increasing in abundance in several locations based on our observations.

Restoration efforts in Puget Sound since 1998 have successfully increased native oyster extent, abundance, and reproduction in Liberty Bay/Dogfish Bay through the use of habitat enhancements. Prior to that, the distribution of approximately 12 million generic hatchery seed at some 80 locations throughout Puget Sound, including several large out-plantings, has not been shown to directly contribute to re-establishment or restoration of reproducing oyster populations. Nor have several habitat enhancement efforts substantially contributed to re-establishing or enhancing reproducing populations (e.g., Woodward Bay and Frye Cove). One large restoration site located in Fidalgo Bay that utilized both habitat enhancement and generic hatchery seed has experienced a significant increase in oyster recruitment and abundance; but genetic analyses of two sample groups showed that increase was due to reproduction by pre-existing wild oysters, with no observed contribution from hatchery seeding (David Stick, Oregon State University, Dept. of Fisheries and Wildlife, Hatfield Marine Science Center, personal communication). In addition to seeding and habitat enhancement efforts, a large body of knowledge has developed since 1998 regarding restoration methodologies, current and historic occurrences, habitat preferences, and the genetic structure of *O. lurida* populations in Puget Sound.

Significant lessons learned since 1998 have resulted in several changes in the approach to restoration: (1) An emphasis on habitat enhancement rather than a reliance on hatchery seed; (2) The development and use of genetic conservation protocols for hatchery seed; (3) Better-informed decisions on where and how to conduct restoration, and; (4) More effective collaboration between resource managers and restoration proponents.

Current stressors of native oysters in Puget Sound are limited to the following factors: (1) Localized occurrences of the non-native predators *Ocinebrellus inornatus* and *Koinostylochus ostreaophagus*; (2) Shoreline and tideland modifications, including nearshore or estuarine restoration projects; (3) By-catch mortality from Pacific oyster commercial harvest and other uses of tidelands; (4) Siltation from upland practices and nutrient inputs, and; (5) Genetic fitness impacts from unrestricted distribution of generic hatchery-origin native oysters.

Pollutants have not been shown to be a significant stressor of native oysters in recent years. The often-cited water pollution effects of SWL released into southern Puget Sound ceased in 1957, and although some concerns have been expressed regarding PAHs, anti-foulants, pesticides and other chemicals, no specific pollutants have been documented at this time as active stressors for native oysters in Puget Sound.

Likewise, the harvest of wild native oyster stocks is no longer a significant stressor. That being said, any return to commercial harvesting would pose a threat to the sustainability of the wild stock resource, for the following reasons: (1) Wild stocks are very slow to build up; (2) Exploitation can quickly deplete them, as happened in the past; and (3) Local populations can be subject to extreme decreases in abundance due to storm and freezing events (Galtsoff 1929).

Factors restricting restoration efforts and natural re-establishment of oyster beds vary somewhat between sub-basins, but are generally dominated by a lack of suitable habitat conditions where historic natural beds occurred. These factors include: (1) Alterations by other uses, such as occupancy of tidelands by eelgrass or shellfish cultivation; (2) Loss of intertidal extents due to fill, diking and siltation, and; (3) Diminished or functionally extirpated reproductive potential.

Additional factors restricting restoration efforts include tideland ownership issues and management conflicts, alternative or higher-valued uses of those tidelands, human health considerations, and at some sites, the presence of predators.

Some factors that have been suggested as limiting factors throughout Puget Sound remain undocumented, or occur only at certain sites. For example, competition with *C. gigas* has not been documented in cases where both species overlap as reproductive populations in Puget Sound. It has also been suggested that *C. gigas* accumulations in the mid- and high intertidal may function as a “larval sink” for native oysters, since spat attracted to these tidal heights will not survive; but we have not observed this occurring in Puget Sound. Predation by the non-native drill *O. inornatus* has not been observed as a limiting factor except in several locations. In other locations, both non-native drills and native oysters appear to co-occur in abundance, and at one of these sites (Potlatch State Park), native oysters have been increasing in abundance.

Potential stressors or restrictive factors such as ocean acidification and rising sea levels are not addressed in this plan. Those subjects, while of great concern, cannot be addressed directly through native oyster restoration efforts, and including them as a component of restoration would diminish the availability of staff and funding resources. It is assumed that re-establishment and enhancement of native oyster source populations at multiple locations (and enhanced gene flow between locations) is a prudent choice for conservation of native oysters in anticipation of large environmental changes.

IV. Restoration Strategy and Products

In order to achieve the primary objectives of native oyster restoration in Puget Sound by 2022, WDFW recommends strategically focused efforts towards re-establishing or enhancing the presence of viable, self-sustaining source populations. These source populations should be in the form of natural assemblages that will trend towards the formation of historic natural beds in areas where such beds existed prior to the period of exploitation and decline. Nineteen primary restoration locations have been identified by WDFW and partner organizations for focused actions meeting that primary objective (Figure 2). Achieving objectives at these 19 primary sites -- coupled with enhanced gene flow as described below -- will complement the existing remnants of *O. lurida* still present in Puget Sound.

Secondary to the primary objective -- but necessary to ensure genetic health and persistence of a restored native oyster metapopulation -- is the re-establishment and enhancement of genetic diversity. Re-establishing source populations will greatly facilitate the export of genetic material, but additional linkages may be necessary in all sub-basins to provide the needed pathways for gene flow within and between sub-basins. Secondary site actions aimed at enhancing gene flow should only be initiated in any sub-basin after restoration actions aimed at primary objectives have been fully implemented; gene flow is not a primary restoration objective, nor would it achieve restoration objectives absent the presence of an enhanced source population.

While restoration at historic locations remains preferable, the small-scale cultivation of genetically appropriate stocks may also function to provide gene flow pathways; limited opportunities for small-scale cultivation should therefore be explored. At the very least, cultivation would provide some ecosystem services benefits, although the continued use of hatchery seed carries genetic conservation risks, and preference should be given to using seed that has been collected locally from wild stocks.

By 2022, the expected result of these strategies is a restored self-sustaining presence of dense natural oyster assemblages as well as gene flow pathways that approximate the probable metapopulation structure present prior to exploitation. These strategies should allow for the continued self-rebuilding of local oyster abundances, re-colonization of former oyster extents, and local ecosystem services which increasingly trend towards historic conditions (Table 1).

With regard to ecosystem services, restoration proponents should recognize that native oyster restoration is unlikely to have immediate positive influences upon water quality or nutrient cycling, and by itself should not be viewed as a “quick fix” for these issues. Even in the long term, oyster restoration is unlikely to mitigate in any substantial way the increasing effects of human inputs of nitrogen and other nutrients into Puget Sound. With regard to water quality and nutrient cycling, the native oyster was at best only a local services provider, one contributor amongst a large multi-species guild of filter-feeding bivalves that still exists today. The most likely immediate benefits would be derived from increased local habitat structure and complexity, especially in those embayments where eelgrass and/or macroalgae have been substantially reduced as three-dimensional habitat structure providers.

Beyond 2022, selective habitat enhancement plus the use of wild stock seed and adult transfers may be pursued to increase the velocity of rebuilding. At that point, large-scale restoration efforts -- including hatchery seeding -- should not be necessary in most locales. The expectation after 2022 is that the re-established native oyster populations will be able to continue increasing in abundance and extent, with minimal or no direct assistance. After 2022, a decision to continue expanding native oyster populations significantly would require the dedication of tidelands that are currently more valued for aquaculture and as essential fish habitat (EFH).

V. Implementation

Success in reaching the objectives of native oyster restoration at the identified 19 locations in Puget Sound by 2022 is dependent upon: (1) A focused strategy that minimizes diversions which do not provide meaningful direct results in terms of biological conservation and ecosystem services; (2) A strong partnership of government, tribes, non-profits, industry, academia, and citizens, and; (3) Adequate funding and support.

WDFW will continue in its role as the primary state agency responsible for conservation, restoration, enhancement and management of the native oyster in Puget Sound. Within budgetary and staffing constraints, WDFW will continue to provide planning, oversight and guidance, site selection, restoration methodology training, site mapping, and research. WDFW will review project reports for determination of restoration status for each location and achievement of objectives.

Achieving objectives at each restoration site is dependent upon individuals and partner organizations outside of WDFW assuming lead roles as project proponents. Native oyster restoration project partners will coordinate with the department on site selection, appropriate methodologies, work plans and timelines for actions proposed for public tidelands. The department will review those project proposals for appropriateness in meeting restoration objectives, and will provide technical and field assistance as necessary. Partner organizations will also be responsible for acquiring all necessary permits and access permissions from public tideland owners. Evaluating the progress and success in meeting the restoration objectives of this plan will require continued research and monitoring into historic and current occurrences, genetic stock composition, hatchery propagation, habitat requirements, and enhancement methodologies. To achieve these objectives, sampling protocols and analysis will be conducted, utilizing the WDFW metrics in sections VI and VII below and reports provided to the department and partners for review. Research regarding ecosystem services or benefits derived from restored native oysters is currently lacking; specifically, their role in nutrient cycling and habitat values. Each restoration project action should include components of the research necessary for achieving restoration objectives as well as post-restoration sampling and evaluation reports. The department will continue to assist restoration proponents in developing their research priorities, plans, data collection and analysis as a component of each restoration proposal, and also as separate concurrent research efforts.

Restoration Methodologies

Due to the wide range of existing physical, biological, and management conditions between and within each primary restoration location, restoration actions and appropriate methods are to be determined by site-specific conditions, feasibility, and an evaluation of the potential benefits. Restoration methodologies for each specific site will be evaluated and selected by WDFW staff in consultation with project leads for public tidelands. WDFW will only serve in an advisory role for those efforts proposed for and conducted on tribal and private tidelands. The Olympia Oyster Restoration Methods Tool-kit (Table 2) summarizes the existing methodologies available to proponents.

As prioritized in Table 2, the preferred restoration methodology involves re-establishment or enhancement of habitat structure required for successful larval settlement and formation of natural beds; ideally, this would require no further maintenance applications of materials. Currently, the use of relict *C. gigas* shell is the standard method of habitat enhancement, although shell from *O. lurida* may be useful in certain applications, if available. Shell from clams and mussels might also be explored as an alternative to the limited supply of suitable materials. Applications of shell for habitat restoration should minimize, as much as feasible, the depth and continuity of those applications, emphasizing “patchiness” to maximize habitat complexity and species diversity.

Rocky material such as gravel has been proposed for habitat enhancement, and might be useful on a very limited basis. But use of rocky materials also runs the risk of leaving a persistent artificial feature if restoration fails, and may also result in colonization by non-target species such as hardshell clams; these species would likely attract intensive harvest, an outcome that would not be compatible with native oyster restoration. Likewise, persistent artificial constructs made of concrete, cement, plastics and other anthropogenic materials are not appropriate for natural oyster beds. Short-term use of some materials -- such as plastic mesh bags and rebar -- may be necessary for grow-out and conditioning of seed oysters prior to out-planting, but they should not be used as permanent fixtures at primary restoration sites. Where secondary sites involving cultivation are developed for gene flow enhancement and harvest, the use of aquaculture technology and materials including plastics would be appropriate.

Wherever enhancement with oyster “seed” is proposed, the collection and use of wild-stock seed within sub-basins is preferable to the use of hatchery seed, if feasible. Additionally, collection and transfers of adult oysters may be necessary in some cases to establish a presence of congeners or cues that enhance larval attraction and settlement. It is assumed that as local populations increase in abundance due to restoration efforts, some of the resulting oysters could be utilized for transfers within the sub-basin for restoration at additional primary and secondary locations.

The use of hatchery-origin seed as a restoration method should only be selected where the preferred action -- habitat re-establishment -- is determined in itself to be inadequate for achieving restoration goals. To date, the use of generic hatchery seed for restoration has not been proven to result in reproducing populations, raising questions regarding fitness. Current seed production for Olympia oyster restoration projects, unlike generic hatchery production, uses strict genetic conservation protocols that theoretically ensure genetic diversity so far as it is currently measured. *Appendix A* provides interim guidelines for hatchery production of seed for restoration projects that were utilized in an on-going Olympia oyster restoration effort in Budd Inlet.

While these genetic protocols outlined in *Appendix A* may prove successful in contributing to restoration, concerns regarding the fitness of hatchery seed remain. Hatchery seed should therefore remain a cautiously used methodology, and even then only as a short-term application. The distribution and use of generic hatchery seed for native oyster restoration is discouraged due to genetic conservation concerns.

Active control of predators such as *O. inornatus* on primary restoration sites should not be necessary for achieving successful restoration. If predation is determined to be a barrier to success at any primary site, those specific projects should be reviewed by WDFW and project proponents to determine if there are alternative non-maintenance actions that may be taken, or if continued restoration remains feasible at all. Alternative primary locations can be substituted through consultation with WDFW if necessary to maintain the Year 2022 objective of 19 restored populations. Predator and pest control on secondary sites used for cultivation may be necessary, and may be conducted as appropriate.

It will also be necessary to dedicate tidelands to native oyster restoration to facilitate the feasibility and success of habitat restoration and/or seeding. Approximately 70% of tidelands in Puget Sound are in non-public ownership (including tribal) over which WDFW has no authority with regard to native oyster restoration. The remaining 30% of tidelands that are in public ownership, while predominantly managed by WDNR also include a mixture of ownership by the federal government, other state agencies, counties, cities, ports and other public entities. Both public and non-public tidelands may have existing uses or conditions that would not be compatible with native oyster restoration and conservation. In several instances, restoration actions would require altering existing uses (such as aquaculture) or habitat conditions (such as tidelands dominated by eelgrass) in order for the primary objectives in this plan to be achieved. With regard to eelgrass, considerable research will be required to validate the restoration of historic native oyster habitat extents now occupied by this highly valued essential fish habitat (EFH).

The use of *ad hoc* "shellfish gardens" as a restoration tool for native oysters in Puget Sound is not supported or advocated in this plan, at least when they rely on generic hatchery seed. While "shellfish gardens" are popularly seen as having restoration benefits, there is no evidence that they contribute to re-populating native oyster beds or measurably improving water quality; they may also pose a genetic conservation risk to remnant and restored wild stocks. A strict adherence to appropriate genetic protocols would be necessary for shellfish gardening using native oyster hatchery seed to be considered a best management practice.

Commercial cultivation or farming of native oysters, while not directly restoring natural native oyster beds or populations, may contribute indirectly via the use of either hatchery seed that meets genetic conservation protocols or via the use of locally caught wild-origin spat. Those cultivation sites would then assist in enhancing gene flow and colonization of available habitat.

VI. Restoration Monitoring and Metrics of Success

Measureable benchmarks or metrics, including the collection of baseline data and long-term monitoring, are a prerequisite for determining success or the need for adaptive changes to restoration actions. Due to differences in annual reproduction and recruitment between sub-basins -- and variation annually within sub-basins – the determination of success may require monitoring periods of 10 years following the start of restoration actions. In particular, reproductive success in northern Puget Sound has historically been sporadic, and recent work at the Fidalgo site supports those observations (Steele 1957; Dinnel *et al.* 2011).

The minimum threshold for determining successful restoration is the observation of significant reproduction, recruitment, survival, utilization, expansion and colonization as described in *Measurable Benchmarks* (Table 3) during any three years within a 10-year period. This threshold should be achievable in all sub-basins, although the northern Puget Sound basin may experience sporadic or irregular spawning and setting due to lower water temperature (Galtsoff 1929; Steele 1957). In primary restoration locations (such as Dyes Inlet) where there are multiple sub-inlets, or in the case of a large geographic area (such as the Point Jefferson-Orchard Point complex of passages and inlets), efforts may be directed at multiple sites or sub-inlets; consequently, only one site/sub-inlet within each location needs to meet the threshold for determination of success. It is expected, however, that over the 10-year period of this plan that multiple sites within each location would reach that threshold. Taken together, these metrics should indicate the restored presence -- or lack of presence -- of a naturally functioning source population. Such a population is self-sustaining, exporting competent larvae, and is assumed to be trending towards historic (pre-1850) conditions, increasingly providing local ecosystem services benefits.

No minimum thresholds are provided here with regard to ecosystem services. Those services are assumed to increase as biological conservation objectives are achieved; they are also difficult to quantify, being innately subjective and dependent upon a reviewer's perspective. Monitoring metrics are offered in brief, however, as measures of desired benefits resulting over time from restoration. Monitoring for ecosystem services -- due to cost considerations, the lack of standardized methodologies, and the requisite supporting data -- may not be feasible in each of the 19 primary restoration locations. Data specific to *O. lurida* in Puget Sound regarding key metrics -- such as oyster dimensions, water temperature, salinity, immersion and exposure regimes for intertidal oyster occurrences, seston load and composition – are generally lacking, and will, by necessity, require acquisition over the 10-year period of this plan.

Genetic monitoring is included as a metric in order to determine if the use of hatchery-origin oysters is contributing to achieving restoration objectives Existing analytical methods that have been used for examining Puget Sound native oysters for population structure and gene flow may not be adequate for truly determining contribution, and the application of new methodologies will need to be developed concurrent with restoration efforts. Additionally, there remain concerns regarding genetic fitness of hatchery oysters and interactions with wild stocks that have not been examined. For hatchery seed to be an appropriate and useful tool for restoration their use must: (1) Cause no harm, and; (2) Contribute to establishment of viable self-sustaining natural populations. Additional genetic examinations of extant wild stocks should also continue as they are encountered to increase the knowledge base regarding structure and gene flow.

Measureable benchmarks such as “acreage” and “quantities of oysters seeded” are not included in this plan as metrics of success, although in principle the “acreage restored” is a desirable long-term result. “Acreage restored” by seeding or habitat enhancement is an often-used deliverable for grants that is highly subjective, and which by itself provides no biological metrics of restoration success. A “natural bed,” while subjectively definable both physically and biologically, is a desired state at all locations; but “natural beds” in the historical sense are not a realistic goal at any site by 2022, and are not included as a defined restoration goal in this plan. Long term viability, self-sustainability, source population functions, and ecosystem services are all achievable conditions at oyster densities lower than those which might be classified as constituting an “oyster bed.” The primary goal of restoration is met with all established populations measurably trending towards ‘bed” formation by 2022.

VII. Site Selection, Prioritization and Descriptions

The selection and prioritization of sites for restoration should focus on known or probable historic natural oyster beds that would have functioned as source populations and provided local ecosystem services prior to exploitation. Due to habitat alterations and conflicting current uses at several historic beds, there may be several locations -- including entire embayments -- that are of little value or feasibility as primary restoration sites. Additionally, the degree of influence provided by existing multi-species guilds of native and non-native bivalves (including commercially cultivated bivalves) may make the contribution of additional *O. lurida* insignificant with regard to water quality ecosystem services.

WDFW has conducted an extensive review of historical records to identify locations where large natural *O. lurida* “beds” were present in Puget Sound prior to exploitation. In many cases those locations can only be identified to the broad scale of embayments (e.g., Bellingham Bay, Padilla Bay or Drayton Harbor). In such cases, further refinement will be required, based upon both historical information and more recent observations from locations where more detail is available, or where native oysters are currently abundant. Records providing extent and abundance for historical native oyster beds are lacking for all locations, although at several sites there exists some information that allows estimates of minimum extents (e.g., ≥ 2,000 acres for Samish Bay).

The North Puget Sound sub-basin historically had large native oyster beds located in or at Drayton Harbor, Portage Island, Bellingham Bay, Chuckanut Bay, Samish Bay, Padilla Bay, Fidalgo Bay, and Similk Bay. Smaller beds or occurrences were known in Dugalla Bay, Penn Cove and throughout the San Juan archipelago. Native oyster beds were also found immediately north of the United States/Canadian border in Boundary Bay. Current known natural presence is limited to Drayton Harbor, Samish Bay, Fidalgo Bay and Shoal Bay Lagoon on Lopez Island. Fidalgo Bay is the location of a recent resurgence of natural larval sets. Location descriptions for North Sound primary restoration locations are as follows:

Drayton Harbor: The purpose of restoration here is biological conservation. Ownership is a mixture of public and private tidelands. Barriers to restoration actions are eelgrass, oyster drills, human health concerns, lack of substrate and commercial aquaculture. We recommend initially establishing several small intertidal sites using hatchery seed with the goal of at least a persistent population exhibiting larval production. If this strategy is successful, we then recommend habitat enhancement to expand that population.

Bellingham Bay (South) Shoreline, Portage Island and Chuckanut Bay: The purpose of restoration here is biological conservation. Ownership is mixed public/tribal/private. Barriers to restoration actions are human health concerns. We recommend initially establishing several small intertidal sites using hatchery seed with the goal of establishing a reproducing self-sustaining population. If this is successful, we then recommend habitat enhancement to expand that population.

Samish Bay: The purpose of restoration here is biological conservation. Ownership is predominantly private with limited public tidelands. Barriers to restoration actions are eelgrass, commercial aquaculture, oyster drills, lack of substrate, and ownership. We recommend initially establishing several small sites using hatchery seed with the goal of establishing a reproducing self-sustaining population. If this proves successful, we then recommend habitat enhancement to expand that population. Acquisition of large tideland tracts may be needed here.

Padilla Bay: The purpose of restoration here is biological conservation. Ownership is public. Barriers to restoration include eelgrass, oyster drills, management issues, and human health concerns. We recommend initially establishing a single small site using hatchery seed with the goal of establishing a self-sustaining population. If this proves successful, further seeding and habitat enhancement is unlikely to be needed. Transfers of recruits to other suitable locations within the bay to facilitate larval recruitment and colonization might be explored.

Fidalgo Bay: The purpose of restoration here includes both biological conservation and ecosystem services. Ownership is a mixture of private and public tidelands. Barriers to restoration are eelgrass, oyster drills, land management issues, lack of habitat, and human health concerns. We recommend enhancing and expanding habitat within and adjacent to the current footprint of an expanding natural population at the Trestle restoration site. Trial ouplants of hatchery seed at Cap Sante Marina and transfers of recruits to other adjacent locations should also be explored.

Similk Bay: The purpose of restoration here includes both biological conservation and ecosystem services. Ownership is a mixture of tribal, public,and private tidelands. Barriers to restoration are eelgrass, oyster drills, land management issues, human health concerns, ownership and lack of habitat. We recommend initially establishing several small sites using hatchery seed with the goal of establishing a self-sustaining population. Initial sites at Lone Tree Point Lagoon and Kiket Lagoon are intended to establish self-sustaining populations in the vicinity. If these are successful, further seeding and habitat enhancement in Similk and Turner Bays will be needed. Transfers of recruits to other suitable locations to facilitate larval recruitment should be explored. Natural-set seed collection inside both lagoons should be considered for transfers to other sites in the vicinity.

The Strait/Admiralty Inlet sub-basin historically had moderate- to small-sized native oyster beds located in Sequim Bay, Discovery Bay, Kilisut Harbor and Port Gamble Bay. Naturally occurring oysters are currently present in Discovery Bay, Kilisut Harbor, and Port Gamble and all these beds appear to be self-sustaining although oyster densities are low. Location descriptions for Strait/Admiralty primary restoration locations are as follows:

Sequim Bay: The purpose of restoration here includes both biological conservation and ecosystem services. Ownership is a mixture of tribal, public, and private tidelands. Barriers to restoration are eelgrass, land management issues, human health concerns, ownership and lack of habitat. We recommend initially establishing several small sites using hatchery seed with the goal of establishing a self-sustaining population. The initial site at Jimmy Come Lately Creek is intended to establish a reproducing population that can be used for relays to establish additional populations in Washington Harbor and Paradise Cove.

Discovery Bay: The purpose of restoration here includes both biological conservation and ecosystem services. Ownership is a mixture of tribal, public, and private tidelands. Barriers to restoration are eelgrass, land management issues, human health concerns, ownership and lack of habitat. We recommend initially establishing one or more small sites at the head of the bay using habitat enhancement with the goal of increasing the density and abundance of the population so as to function as a source population.

Kilisut Harbor: The purpose of restoration here includes both biological conservation and ecosystem services. Ownership is a mixture of military, public, and private tidelands. Barriers to restoration are eelgrass, land management issues, human health concerns, ownership and lack of habitat. We recommend initially establishing several small sites using hatchery seed and habitat enhancement in lower Scow Bay, with the goal of increasing the abundance and density of the population, so as to function as a source population. If this proves successful, the resulting adults may be transferred to additional sites in Mystery Bay.

Port Gamble Bay: The purpose of restoration here includes both biological conservation and ecosystem services. Ownership is a mixture of tribal, public, and private tidelands. Barriers to restoration are eelgrass, land management issues, human health concerns, ownership and lack of habitat. We recommend initially establishing several small sites using hatchery seed and habitat enhancement with the goal of increasing the abundance and density of the populations so as to function as a source population.

The Hood Canal sub-basin historically had very large native oyster beds located in Quilcene Bay, at the Seal Rock/ north Dosewallips tidelands, and on the Union River/ Big and Little Mission Creek(s) deltas. Smaller beds, aggregates and individuals occurred throughout the entirety of Hood Canal. Naturally occurring oysters are currently present across all shorelines but small and large beds are absent. Source population function appears to be at least present within the middle reach of Hood Canal, but absent or diminished in the northern and southern reaches. Following primary site restoration in the southern reach of Hood Canal, proponents should consider establishing secondary sites for gene flow enhancement south of Ayock Point. Location descriptions and strategies for Hood Canal primary restoration locations are as follows:

Quilcene Bay: The purpose of restoration here includes both biological conservation and ecosystem services. Ownership is a mixture of public and private tidelands. Barriers to restoration are eelgrass, predation, land management issues, human health concerns, ownership and lack of habitat. We recommend initially establishing several small sites using hatchery seed and habitat enhancement with the goal of increasing abundance and density of the population, and enhancing its function as a source population.

Union River/ Big and Little Mission Creek(s) deltas: The purpose of restoration here includes both biological conservation and ecosystem services. Ownership is a mixture of publicand private tidelands. Barriers to restoration are eelgrass, predation, land management issues human health concerns, ownership and lack of habitat. We recommend initially establishing several small sites using hatchery seed and habitat enhancement with the goal of increasing abundance and density of the population, and enhancing its function as a source population.

The Central Puget Sound sub-basin historically had large native oyster beds located in Liberty Bay, Dyes Inlet and Sinclair Inlet. Smaller beds, aggregates and individuals occurred throughout the entirety of Central Sound. Naturally occurring oysters are present in Liberty Bay, Dyes Inlet, Port Madison, Manchester, Brownsville and several other small inlets. Presence in Sinclair Inlet is unknown. As primary location objectives are met, secondary sites should be considered for Gig Harbor, Quartermaster Harbor, Yukon Harbor, Commencement Bay and the small inlets along the eastern side of Bainbridge Island to further enhance gene flow within the basin. Location descriptions and strategies for Central Puget Sound primary restoration locations are as follows:

Liberty Bay and sub-inlets: The purpose of restoration here includes both biological conservation and ecosystem services. Ownership is a mixture of public and private tidelands. Barriers to restoration are oyster drills, lack of habitat, human health concerns, tideland ownership and management issues. Prior and ongoing restoration efforts have resulted in significant increases in oyster extent and abundance. Additional efforts may be directed at expanding habitat extent. We recommend evaluation of location for determination of success.

Dyes Inlet and sub-inlets: The purpose of restoration here includes both biological conservation and ecosystem services. Ownership is a mixture of public, tribal, and private tidelands. Barriers to restoration are oyster drills, land management issues, human health concerns, ownership and lack of habitat. A larval dispersion gap may be present here that limits the source population to the southern half of the inlet, with the northern portion functioning as a larval sink. We recommend using habitat enhancement in the southern inlets with the goal of increasing the abundance and density of the population, and enhancing its function as a source population.

Sinclair Inlet: The purpose of restoration here includes both biological conservation and ecosystem services. Ownership is a mixture of public and private tidelands. Barriers to restoration are human health concerns, lack of habitat, tideland ownership and management issues. We recommend initially establishing several small sites using hatchery seed and habitat enhancement with the goal of increasing the abundance and density of the population, and enhancing its function as a source population.

Point Jefferson-Orchard Point complex of passages and inlets: The purpose of restoration here includes biological conservation, enhanced ecosystem services, and re-establishment of gene flow. Ownership is a mixture of public, tribal, and private tidelands. Barriers to restoration are eelgrass, oyster drill predation, human health concerns, lack of habitat, tideland ownership and management issues. We recommend initially establishing several small sites using hatchery seed and habitat enhancement with the goal of increasing the abundance and density of the population, and enhancing gene flow between source populations.

The South Puget Sound sub-basin historically had large native oyster beds located in Budd Inlet, Henderson Inlet, Totten Inlet, Big Skookum Inlet, Eld Inlet, Oakland Bay, North Bay, Carr Inlet and on Squaxin Island. Smaller beds, aggregates and individuals occurred throughout the entirety of South Sound. Naturally occurring oysters are present in all historic locations in -South Sound. -In particular, the North Bay population is Puget Sound's single example of a naturally self-sustaining native oyster bed. Albeit bed structure is not present, the abundance of oysters in Totten Inlet continues to support the commercial collection of wild-origin spat. Location descriptions and strategies for Central Puget Sound primary restoration locations are as follows:

Budd Inlet: The purpose of restoration here includes both biological conservation and enhanced ecosystem services. Ownership is a mixture of public and private tidelands. Barriers to restoration are human health concerns, lack of habitat, tideland ownership and management issues. A dispersion gap may separate the southern portion of the inlet from the northern portion. Restoration action and research has already been initiated at Priest Point. We recommend establishing several small sites using wild-caught or hatchery seed and habitat enhancement in the vicinity of the Port of Olympia and southwestern shoreline of the inlet, with the goal of increasing the abundance and density of the population, and enhancing its function as a source population. As primary objectives are met, secondary sites might be pursued at Gull Harbor in the northern section.

Henderson Inlet: The purpose of restoration here includes both biological conservation and enhanced ecosystem services. Ownership is a mixture of public, tribal and private tidelands. Barriers to restoration are oyster drill predation, human health concerns, lack of habitat, tideland ownership and management issues. We recommend establishing several small sites using wild caught seed and habitat enhancement in the lower portion of the inlet south of Woodward Bay with the goal of increasing the abundance and density of the population, and enhancing its function as a source population. As primary objectives are met, secondary sites should be considered in the vicinity of Woodward Bay.

Harstine/Squaxin Islands complex of passages and inlets: The purpose of restoration here includes both biological conservation and enhanced ecosystem services. Ownership is a mixture of public, tribal and private tidelands. Barriers to restoration are oyster drill predation, human health concerns, lack of habitat, tideland ownership and management issues. We recommend establishing several small sites using wild caught seed and habitat enhancement on Squaxin Island and along Peale Passage, with the goal of increasing the abundance and density of the population, and enhancing its function as a source population.

Figure 1. Historic distribution of Olympia oysters in Puget Sound. (circa 1850)

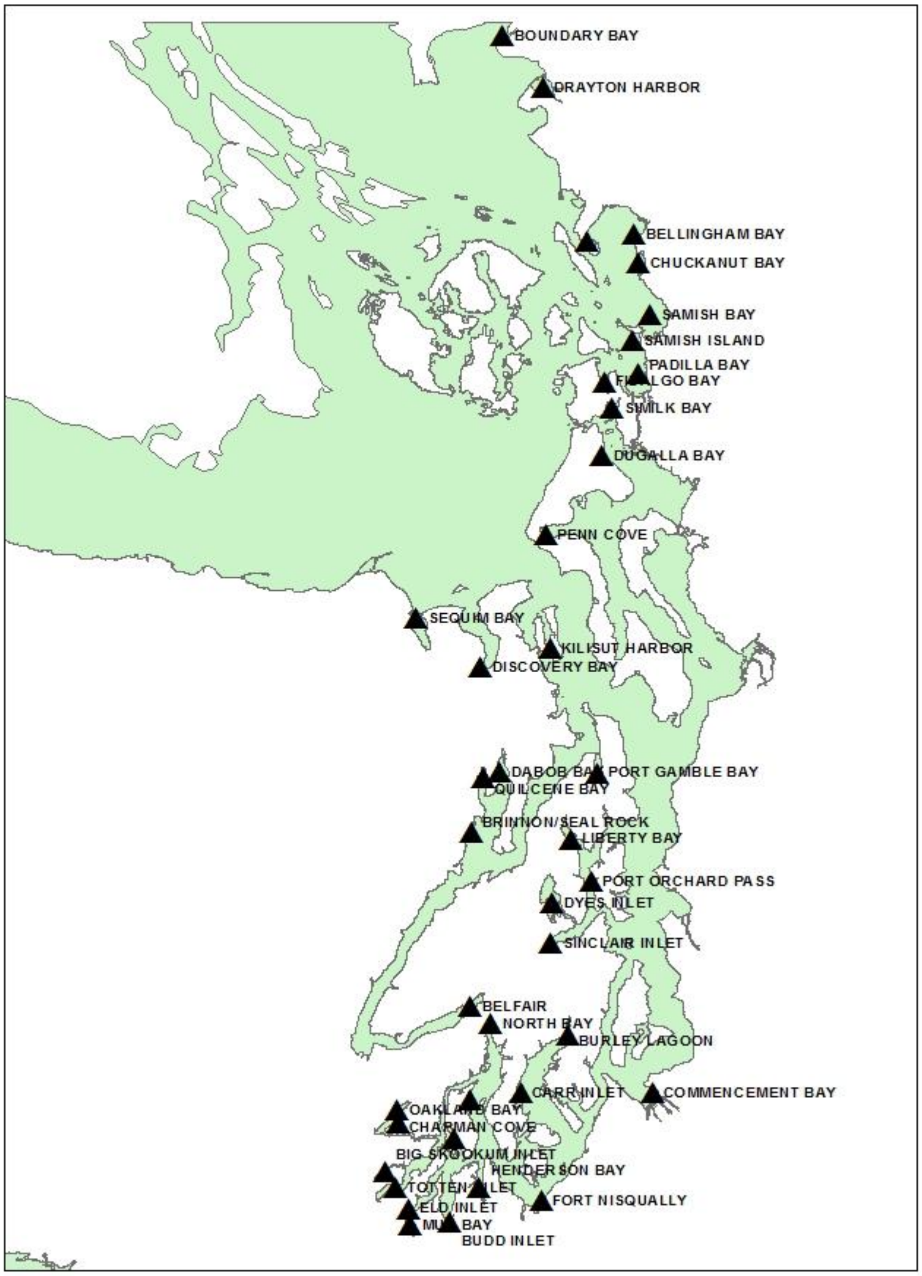

Figure 2. Nineteen proposed Olympia oyster restoration sites in Puget Sound.

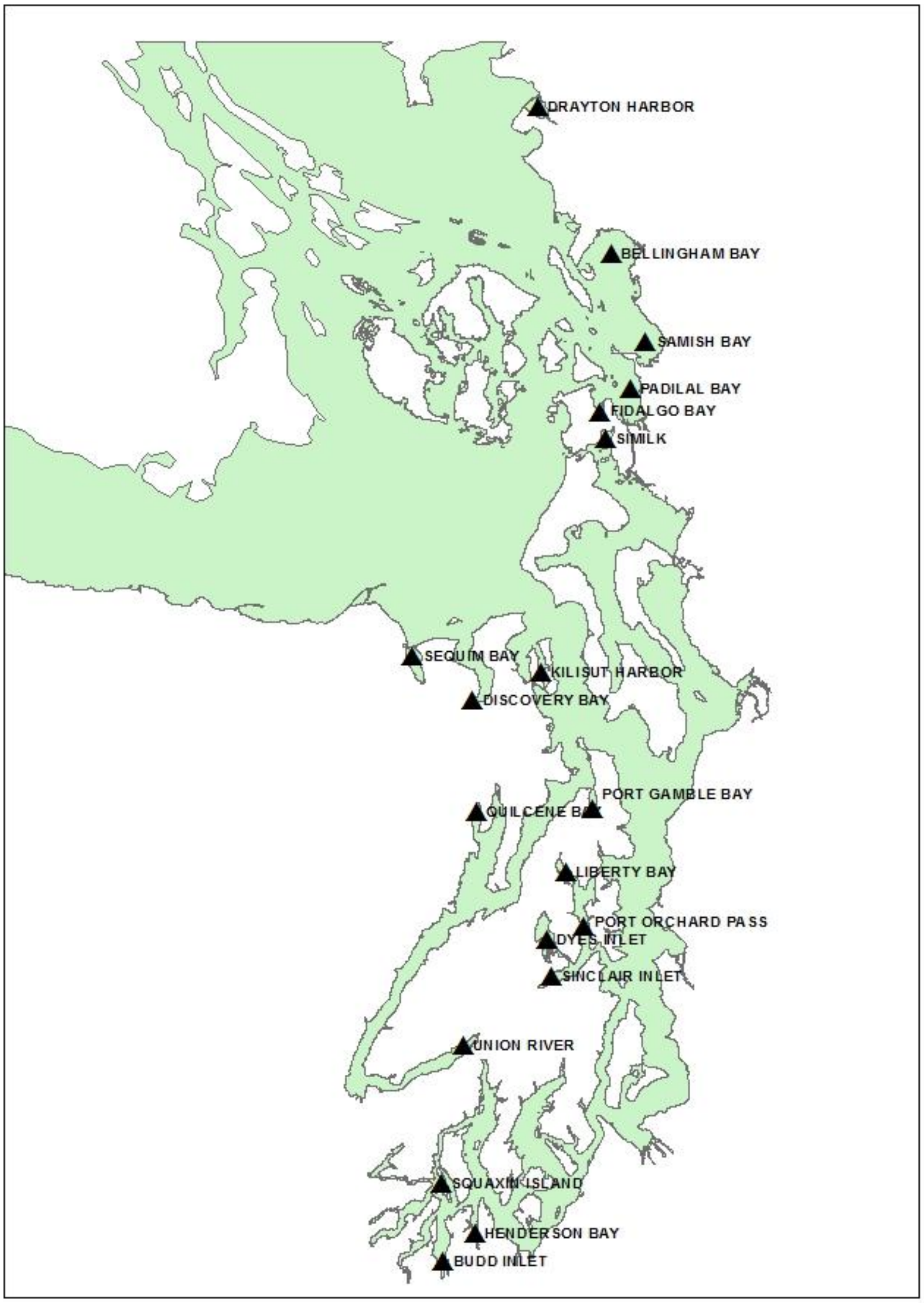

Table 1. Expected Products of Native Oyster Restoration efforts in Puget Sound

1. Naturally self-sustaining, viable, and persistent populations of native oysters and the ecosystem habitat they provide.Enhanced gene flow and decreasing risk of isolation, drift and inbreeding depression.
2. Decreased risk of further declines in distribution and abundance.
3. Localized regulating services through the maintenance of water quality and stabilization of tidelands by sediment trapping. Oysters can maintain and improve water quality by filtering large quantities of particulate matter (both organic and inorganic) and nutrients from the overlying water column. Additionally, oyster reefs alter hydrodynamic conditions, further increasing the removal of particulate matter from the water column (Nelson *et al.* 2004).
4. Supportive services through nutrient cycling (denitrification and remineralization), the provision of nursery habitat and forage for finfish and crustaceans, increased biodiversity, and three-dimensional habitat structure.
5. Provisioning services for tribal ceremonial and subsistence needs.

Table 2. Olympia Oyster Restoration Methods Tool-kit

1. Habitat structure re-establishment and enhancement, using relict shell or other appropriate hard substrate to provide a foundation for native oyster bed formation. Habitat enhancement should result in natural structure following recruitment. Artificial structures such as concrete shapes and structures that do not recreate or result in natural conditions typical for *O. lurida* are an inappropriate choice for restoration. Site selection for habitat restoration should not occur in locations that would require perpetual maintenance in order for habitat structure to persist. Methods should avoid use of materials that may create commercially attractive hardshell clam beds that could create potential fishery conflicts.
2. Reproductive potential re-establishment using hatchery-origin seed or natural set seed to provide a base population for reproduction and recruitment. Use of hatchery-origin seed represents a risk to extant natural oyster populations and achieving restoration objectives. Strict genetic conservation guidelines are required for use of seeding as a restoration tool and seed application should be minimized and then cease once measureable biological benchmarks for determining re-establishment success are achieved. Analysis and monitoring tools should be developed and utilized to identify the biological and economic value of hatchery seed as a restoration tool. See Appendix A for genetic conservation guidelines for native oyster restoration.
3. Collection and transfers of wild stock adults to restoration sites to enhance larval setting. Transfer of natural origin oysters may be given preference over the use of hatchery seed where such resources are available, and should eventually make the continued use of hatchery origin seed an unnecessary genetic conservation risk.
4. Tideland dedication for native oyster restoration and conservation purposes. Methods include planning, conversion of tideland uses, agreement, donation, easement, lease, purchase or other actions facilitating native oyster restoration and conservation. In some instances, alteration of existing habitat conditions such as eelgrass may be required.

Table 3. Measureable Benchmarks for describing baseline conditions, monitoring, and determining restoration status.

1. Reproduction, recruitment and survival resulting in multiple year classes.

    - Reproduction metric is observed sexual maturity, fertilization, larval development, brooding and larval release. H_0: *No significant reproduction occurred,* $\alpha=0.05$

    - Recruitment metric is determined by early spring quantitative surveys of each preceding year's larval set or year class. H_0: *No significant larval recruitment occurred,* $\alpha=0.05$

    - Survival metric is a quantitative estimate of total population abundance determined from early spring surveys. H_o: *Abundance in current year does not significantly differ from previous years abundance,* $\alpha=0.05$

    - Annual length-frequency analysis from early spring quantitative surveys for survival. H_0: *Length frequency distribution does not indicate bi-modal or multi-modal distribution.*

2. Utilization of available appropriate habitat structure and colonization.

    - Measure of increased oyster coverage per m^2 within initial footprint acquired during early spring survival and recruitment surveys. H_0: *Live oyster coverage in current year is not significantly different from previous years coverage,* $\alpha=0.05$

    - Colonization expanding from initial footprint. H_0: *Live oyster abundance does not significantly differ in current year from previous year,* $\alpha=0.05$

    - Measure of population expansion distance from initial footprint. H_0: *Live oyster extent not significantly different in current year from previous year.*

3. Observable ecosystem services.

    - Increased fish use. H_0: *Fish utilization of restoration area does not significantly differ in current year from baseline year,* $\alpha=0.05$.

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Appendix A: Interim Guidelines for Hatchery Production of Native Oyster Seed for Restoration

Minimize among-population effects

Use locally-derived broodstocks. Locally-derived is defined as the geographically closest and/or ecologically most similar and viable source population.

Minimize within-population effects

Maximize the ratio of genetically effective numbers of breeders (N_b) to broodstock census size (N) and maximize genetic diversity.

Maximize ratio of N_b:N

(1) Use as many broodstock as possible in full or partial factorial matrices.
(2) Target a 1:1 gender ratio in brood animals.
(3) Minimize variance in family size for outplants.

Maximize genetic diversity

(1) Procure new wild broodstock for each cohort; avoid repeat spawns for each breeder.
(2) Minimize inbreeding by using a large geographically proximate broodstock population. The number of broodstock used will depend on local abundance and the ability to integrate genetic diversity over successive years or cohorts.
(3) Avoid collection of broodstock where hatchery derived animals are present.

Archive tissue samples of every brood animal used (non-lethally when possible) and a subsample of at least 100 seed from each cohort for genetic analyses.

Avoid disease transfers

(1) Certify brood animals and seed by a qualified histopathologist for disease free status according to accepted OIE standards.
(2) Sample seed animals in numbers sufficient to provide 95% confidence of detecting a pathogen with prevalence as low as 5% and compare to wild population.
(3) Return brood animals to place of origin after use when disease-free status of the broodstock population is known.