COUNTY: Skagit

Grant Number: SEANWS-2021-skCoPW-00003

PROJECT TITLE: Northwest Straits Project: Skagit Marine Resources Committee Operations

DELIVERABLE TASK NUMBER: 6.4 Pinto Abalone Recovery- Final Report

PERIOD COVERED: Oct 1, 2022 – Sept 30, 2023

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County: **Skagit County**

Grant No: **SEANWS-2021-SkCoPW-00003**

PROJECT TITLE: **Northwest Straits Project: Skagit County MRC Operations and Projects**

TASK NUMBER: **6: Pinto Abalone Restoration and Monitoring**

DELIVERABLE: **6.2: Pinto Abalone, Year 2 Final Report**

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Pinto Abalone Recovery Project
2023 Final Report to the Skagit MRC
Josh Bouma, Puget Sound Restoration Fund
Paul Dinnel, Skagit MRC
September 30th, 2023

Introduction

The pinto (northern) abalone, *Haliotis kamtschatkana*, is the primary abalone species indigenous to Washington waters, but populations are severely depleted and considered functionally extinct. The current number and distribution of reproductive wild abalone is too low and too widely distributed to maintain a sustainable population. The precipitous decline of abalone in Washington is largely due to anthropogenic factors, including overharvesting during the legal recreational fishery and poaching during the 1980-90s (Bouma 2007). Numbers in Washington state never supported a commercial fishery for abalone. Between 1992 and 2017, the density of pinto abalone declined by 97% at 10 index sites in the San Juan Archipelago (SJA) even after the closure of the recreational fishery in 1994 (Rothaus et al. 2008, WDFW unpublished data). Insignificant numbers of juvenile recruits have been observed and the average size of abalone continues to increase (Rothaus et al. 2008, Bouma et al. 2012, WDFW unpublished data). Both measures indicate likely recruitment failure of pinto abalone in areas of historical presence. They are now listed as a Washington State Endangered Species as of May 2019. They are also listed as a U.S. Federal Species of Concern and a Canadian Endangered Species (PSRF 2014).

Abalone are broadcast spawning invertebrates; gametes undergo fertilization in the water column. After a 7-10 day planktonic larval phase, the larvae go through metamorphosis and settle onto rocks encrusted with pink coralline algae. Juveniles prefer rocky reef and cobbled substrates with crack and crevice habitat to hide in. This large marine snail occurs primarily in the shallow subtidal zone, although they have been found in depths up to 100 ft (NOAA 2007). The abalone diet changes during different life stages; larval abalone are lecithotrophic while planktonic, small juvenile abalone primarily graze on the diatom and bacterial biofilm, while the adults feed on various species of macroalgae.

The apparent recruitment failure and complete lack of recovery for this species is thought to be largely due to the Allee effect (Allee et al. 1949) which can occur when existing animals, particularly broadcast spawning invertebrates like abalone, are not able to find each other and reproduce successfully. A low population density means less successful reproduction and a positive feedback loop that leads to eventual population extinction. Babcock and Keesing (1999) estimated that the minimum density is 0.15 abalone/m^2 for successful reproduction to

occur. Extensive sampling has shown that the remaining San Juan Archipelago (SJA) pinto abalone population is well below this threshold, unable to facilitate necessary reproduction for natural population recovery.

Steps are being taken to help restore the pinto abalone population in northern Puget Sound waters. The recovery project is a long-term collaboration between government agencies, NGOs, universities, tribes and more. This group includes researchers, resource managers, students, technicians and facilities support from the Washington Department of Fish & Wildlife (WDFW); the Puget Sound Restoration Fund (PSRF); Western Washington University's Shannon Point Marine Center (WWU SPMC); the NOAA Manchester Research Station; the Port Townsend Marine Science Center (PTMSC); the University of Washington, School of Aquatic & Fishery Sciences (UW); the Seattle Aquarium and others. Annual funding to PSRF from the Skagit County Marine Resources Committee (Skagit MRC-Skagit County contract #C20210470) supports abalone conservation aquaculture and restoration activities in Skagit County. This funding support for abalone restoration in 2022-2023 was supplemented by funding from WDFW, PSRF, WA Sea Grant, the Benjamin & Margaret Hall Family Foundation and other anonymous private donors.

Puget Sound Restoration Fund, with oversight from WDFW, has developed a conservation aquaculture program designed to supplement depleted wild stocks. The hatchery, wet laboratory and nursery facilities are located at the NOAA Marine Fisheries Research Station in Manchester, WA. This shellfish hatchery, named the Kenneth K. Chew Center for Shellfish Research and Restoration (Chew Center), is devoted to native shellfish and other marine species; work is ongoing on species such as pinto abalone, Olympia oysters, giant sea cucumbers, basket cockles, Dungeness crab, bull kelp and sugar kelp. The Chew Center has been operated by PSRF since 2013 through a Cooperative Research and Development Agreement (CRADA) with NOAA. PSRF began moving the abalone aquaculture program from the NOAA Mukilteo Research station to the Chew Center in 2016 and all pinto abalone hatchery production and research activities are now conducted at the NOAA Manchester Station.

Adult broodstock abalone are collected from the wild and brought into the Chew Center, these animals are spawned in the laboratory to produce larval and juvenile abalone for future outplanting and to provide early life stages for a variety of laboratory experiments. More than 19,500 of these healthy, genetically diverse hatchery produced juvenile abalone have now been outplanted to 10 (two of which were new in 2023) rocky reef sites in Skagit County waters since 2009. Approximately 31,100 abalone have also been outplanted at 17 sites in San Juan County and two sites in Island County, creating a recovery network across a significant portion of their geographical range in Washington. Surveys of some of these outplant sites are conducted each

year to monitor survival, growth and movement, which provide estimates of survival and growth of abalone populations released into the wild (Carson et al. 2019). Methods and results of surveys prior to 2023 can be found in annual WDFW, PSRF and Skagit MRC summary reports (Bouma and Dinnel 2017, 2018, 2019, 2020, 2021 and 2022) as well as project reports by Shannon Point Marine Center (SPMC) students (Bergman 2009, Pratt and Dinnel 2010, Hester et al. 2011, Benolkin et al. 2012, Walker et al. 2013). The following report summarizes PSRF project accomplishments related to the contract listed above during the period from October 1st, 2022-September 30th, 2023.

Chew Center: Facilities, Hatchery Production & Nursery Husbandry

The Chew Center nursery currently includes an 80-tank grow-out system plus an 18-tank research system supplied by a 1,000-gallon primary seawater reservoir tank. This reservoir provides for efficient temperature control by recirculation through a glycol heating loop during the winter and through a heat pump capable of seawater chilling during the summer. This reservoir also allows for sodium carbonate buffering to elevate pH of incoming seawater from 7.75 to 8.0 for optimal abalone culture conditions. An additional 300-gallon seawater reservoir in the nursery supports eight broodstock specific tanks. This reservoir is connected via a venturi mixing valve and temperature controller to the primary 1,000-gallon heated seawater reservoir and incoming cold seawater supply allowing for separate manipulation of temperature and buffering to the broodstock tanks in the nursery. General maintenance of these systems includes regular filter changes, pump replacement, buffering system management, seawater chemistry probe cleaning/calibration/replacement and water quality data monitoring. PSRF staff regularly monitors incoming seawater chemistry and quality, and also examines for potentially problematic nuisance species in the nursery water systems.

Figure 1. Stacked culture tanks in the Chew Center abalone nursery system.

During this contract period, significant plumbing changes were made to reduce the amount of heated seawater used in the nursery culture tank system where tanks are stacked either three or four high depending on the rack (Figure 1). Instead of each of the 98 culture tanks receiving single-pass seawater flow, supply and drain plumbing were altered so that each column of tanks receives new flow at the top with cascading flow moving vertically from the top tank to each subsequent tank below. The

advantage is a significant reduction in the amount of heated seawater used, and the amount of propane used to heat that seawater. Seawater usage in the nursery decreased from approximately 110 gpm to 35 gpm with this change. But there are three disadvantages: First, biosecurity becomes more challenging as fouling/nuisance organisms can travel from tank to tank. Second, maintenance of distinct genetic families may be compromised if small abalone are lost downstream. Third, consistent temperature control between upper and lower tanks may be more difficult. All of these disadvantages can be overcome with careful husbandry and tank maintenance practices.

During the 2023 induced spawning season, which began in May and concluded in August, PSRF successfully induced three out of four attempted spawns and produced 19 unique single-parent families using 4 females and 11 males. A total of 4 million abalone larvae were cultured to competence and 1.157 million of these larvae were settled into nursery grow-out tanks (2.84 million healthy larvae were culled due to lack of settlement space). Ninety-five grow-out tanks were settled with 885,000 larvae at the NOAA Manchester Chew Center at a density of 9,300 larvae per tank on average with a range of 2,400 - 11,500 larvae per tank. Additionally, 2023 was the first year PSRF and the Seattle Aquarium settled larvae into culture tanks at their new Off-Site Animal Care Center (SA ACC). By the end of August, all 32 grow-out tanks available at the SA ACC were settled with 272,000 larvae from 16 genetically distinct families produced during this spawning season, at a density of roughly 8,500 larvae per tank on average. Depending on growth and survival, these new post-set cohorts at both the Chew Center and the SA ACC will be reared until outplanting during spring field seasons in 2024 and 2025.

Figure 2. Post-set abalone (mean shell length 5 mm) clustered around the standpipe in a nursery culture tank.

Maintenance and husbandry of post-set animals (Figure 2) was one of the primary activities within the abalone nursery during the fall, winter, and spring. Weekly care of this post-set population in the nursery included gentle tank cleaning and feeding with three diatom species cultured at the Chew Center (*Amphora salina, Navicula incerta, Cylindrotheca closterium*).

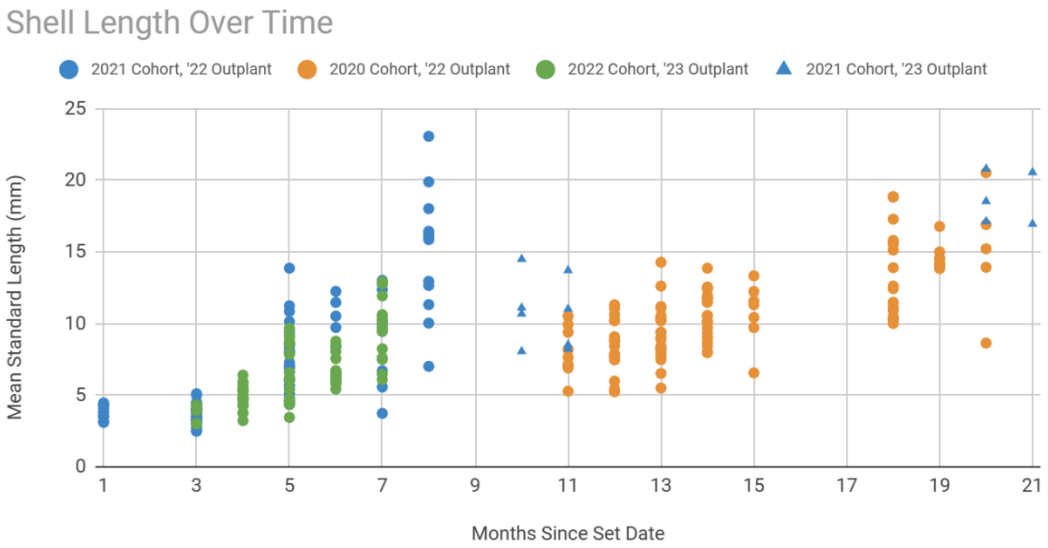

Figure 3: Juvenile abalone shell length over time of production cohorts between 2021-2023.

Once post-set abalone reached 5 mm in shell length, they were weaned onto dulse (*Palmaria mollis)* by blending it into small flakes and introducing it into the grow-out tanks. At six months post-settlement, hatchery staff continued to size sort and consolidate within families to keep similar-sized juvenile animals within a tank, aiming to decrease within-tank competition. Inventory and shell length data were recorded to track growth and survival (Figure 3).

Health & Disease

To confirm no disease-causing pathogens were present in hatchery-reared animals prior to moving abalone between facilities and outplanting animals into the wild, an annual hatchery health assessment was conducted by pathologist Dr. Ralph Elston, AquaTechnics Inc. Live samples were sent for histology to detect known infectious diseases and PCR analysis to determine if withering syndrome was present. On January 28th, 2023, health screening results from all three facilities (PSRF Chew Center, Seattle Aquarium, PTMSC) showed no disease-causing pathogens present. A total of 360 live abalone were sent for sampling, with each facility sending 60 live juveniles for histology and 60 live juveniles for PCR (Figure 4). Upon this confirmation of abalone hatchery health, new transfer permits were received from WDFW to move abalone from the nursey facilities for outplanting.

Figure 4. Chew Center hatchery staff collect abalone for health screening.

Abalone Broodstock

Regular inventory, health and maintenance checks were conducted on all broodstock; tanks were fed and cleaned weekly, and animals were measured, weighed and re-tagged as necessary. Broodstock collections were conducted by PSRF and WDFW divers in the San Juan Islands prior to the spawning season in May. All new broodstock underwent the standard intake protocol: they were measured, weighed, ranked by gonad index, fouling sponges removed from the shell, animals were tagged with vinyl disc tags and PIT tags, non-lethal genetic samples were taken and archived using epipodial tentacle clips, and general observation of overall health was noted. At the end of this reporting period, PSRF had 44 live broodstock animals (11 females, 33 males) at the Chew Center. Eleven new broodstock were collected in 2023 (9 females, 2 males) (Figure 5). Broodstock abalone were fed two types of macroalgae species: dulse and bull kelp (*Nereocystis luetkeana).* PSRF staff culture the dulse in tumble culture and bull kelp is wild collected as needed to sustain hatchery needs.

Juvenile Abalone Outplant Site Monitoring

Between February-March 2023, WDFW divers surveyed for survival, growth and emergence of hatchery reared pinto abalone at eight sites outplanted in 2022. This included six of which are

Figure 5. Divers prepare to scout for broodstock, and to look for ideal locations for new outplant sites.

in San Juan County and two in Island County. No Skagit County restoration sites were created or overseeded in 2022 and therefore no outplant site monitoring was done in Skagit County in 2023. Following guidelines adopted by WDFW and described in the Recovery Plan, when restoration sites are established, they are outplanted in the first and second year and then subsequently overseeded every three to four years to maintain aggregations and boost genetic diversity. The eight existing Skagit County sites were not due to receive juvenile abalone in 2022 according to this rotation, and instead, the abalone recovery team decided to prioritize development of new sites in northwestern areas of the SJA and at Smith Island.

Juvenile Abalone Outplanting

In April and May 2023, in collaboration with WDFW subtidal shellfish biologists, the PSRF team completed the 13th juvenile abalone outplant over the past 14 years at recovery sites in the San Juan Archipelago and surrounding areas. A fundamental objective of the pinto abalone conservation aquaculture program is to "do no harm" to existing wild stocks of abalone and therefore extreme care was taken during the restoration effort described here to outplant a genetically diverse and disease-free cohort of abalone. Since 2009, more than 50,000 pinto abalone have been outplanted to 29 different restoration sites in Skagit, San Juan and Island counties; 19,500 of these hatchery produced juvenile abalone have been outplanted to 10 rocky reef sites in Skagit County waters, two of which were newly established in 2023.

In 2023, 4893 juvenile abalone were outplanted, representing 21 genetically distinct families produced at the Chew Center in 2021 and 2022 and reared at the Chew Center, Seattle Aquarium and PTMSC (Figure 6). These animals were seeded to sites in Skagit (2 sites, newly established in 2023), Island (1 site) and San Juan (3 sites) Counties. Reconnaissance dives were conducted in Skagit County during the monitoring season to follow up on work done in 2022 and several sites for outplants were identified in areas surrounding Deception Pass. Two sites were selected and set up with permanent corner markers; site maps were created including site features, depths, compass headings between corner markers and GPS coordinates. These sites were named Goodenough and Torch. Code names are used to define sites in place of more geographically descriptive site names to maintain anonymity of outplant locations.

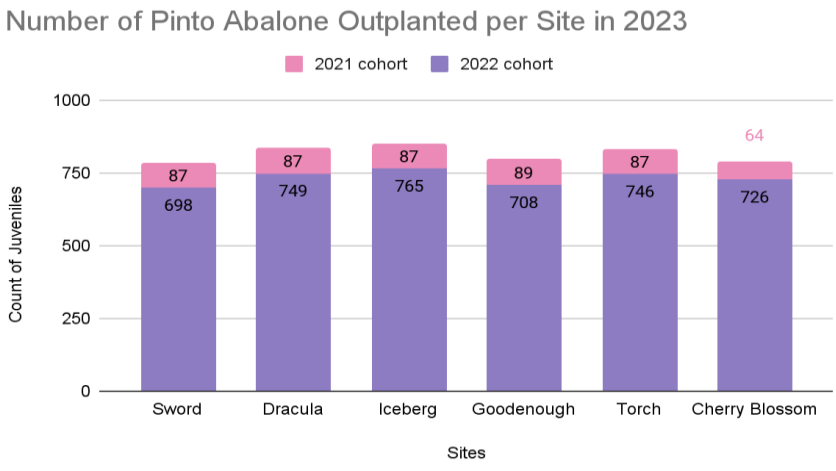

Figure 6: Number of pinto abalone outplanted per restoration site in 2023. Juveniles were evenly distributed among the six outplant sites.

In preparation for outplanting, PSRF staff conducted an inventory, collected shell length data, and sorted families into outplant groups. Additionally, all juveniles from the 2021 cohort were marked with an orange-colored glue dot (Coraffix cyanoacrylate adhesive mixed with Eye Candy mica powder pigment) before outplanting to identify them from the younger group using the same methods as the previous two years (Figure 7). The 2023 outplant effort was similar to the previous three years in that both a younger and older year class were overseeded together onto the same sites representing the fourth opportunity to test and monitor the two age cohorts. The mean shell length of outplanted juveniles across all six sites in 2023 was 10.1 mm

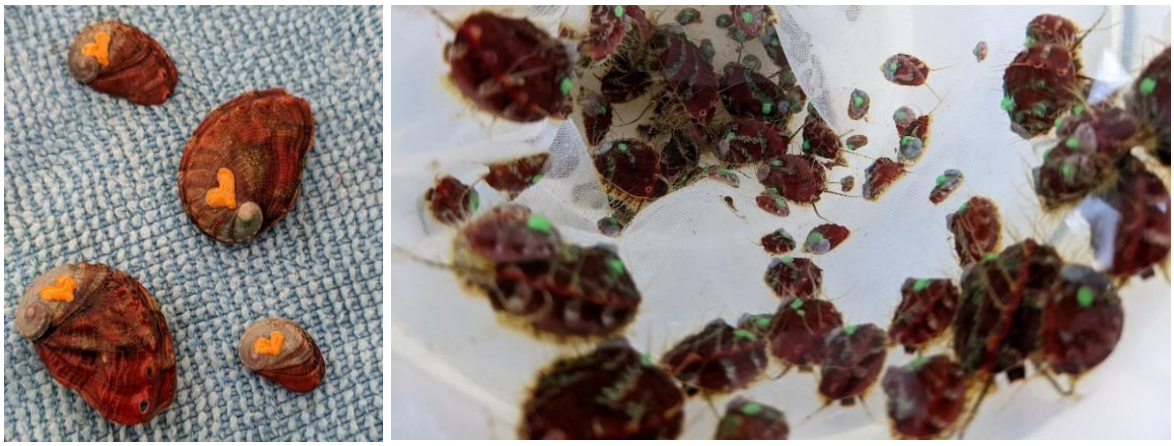

Figure 7. Two-year-old juvenile abalone tagged with an orange dot to indicate 2021 cohort for outplanting in 2023. A green dot indicated 2020 cohort for outplanting the year prior.

for the younger cohort and 18.5 mm for the older cohort.

Oceanographic sensors were installed at both new Skagit County outplant sites (Torch and Goodenough) in April 2023. These arrays, constructed by UW PhD candidate Eileen Bates, gather data on pH, temperature, salinity, dissolved oxygen, light penetration and current flow and have been installed on 18 outplant sites over the past three years. Sensor retrieval, data downloads, probe calibration and redeployment by UW, PSRF and WDFW divers will occur every two months until March, 2024.

Outreach & the Pinto Abalone Recovery Plan

The 11th International Abalone Symposium, a conference that occurs every three years and brings together leading abalone researchers, managers and aquaculturists, was held in Auckland, New Zealand from February 27th-March 2nd, 2023. Much of the pinto abalone research and restoration conducted here in the Pacific Northwest in recent years was presented at this conference:

Eileen Bates, University of Washington. “Can settlement on coralline algae ameliorate negative effects of ocean acidification and temperature increase on pinto abalone early life stages?”

Joshua Bouma, Puget Sound Restoration Fund. “Recovering pinto abalone: use of conservation aquaculture to give Washington State’s largest rocky-reef grazing snail a population boost.”

James Dimond, Western Washington University. “Population genomics of wild and hatchery-raised pinto abalone (*Haliotis kamtschatkana*).”

Katie Sowul, Washington Department of Fish & Wildlife. “Comparing survival of hatchery-reared pinto abalone (*Haliotis kamtshcatkana*) released in mixed-aged cohorts in Washington State.”

As a state listed endangered species, conservation aquaculture and field restoration activities to bring abalone back from the brink of local extirpation are guided by the Washington State Pinto Abalone Recovery Plan (Sowul et al. 2021) approved by state, tribal, university and NGO researchers. In addition to thorough review from collaborators, the Plan was open to public comment and was also presented to the Washington Fish & Wildlife Commission. WDFW, PSRF and Northwest Indian Fisheries Commission biologists finalized the Recovery Plan in 2021 and this document continues to guide restoration strategies. It can be found on both the WDFW and PSRF websites listed below.

https://wdfw.wa.gov/publications/02284
https://restorationfund.org/programs/pintoabalone

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