COUNTY: Skagit

Grant Number: SEANWS-2019-skCoPW-00003

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

TASK NUMBER: 3.2: Pinto Abalone Recovery Final Project Report

PERIOD COVERED: October 1, 2019 – September 30, 2020

DATE SUBMITTED: September 30, 2020

This project has been funded wholly or in part by the United States Environmental Protection Agency. The contents of this document do not necessarily reflect the views and policies of the Environmental Protection Agency under Assistance Agreement [CE-01J65401]. The contents of this document do not necessarily reflect the views and policies of the Environmental Protection Agency, nor does mention of trade names or commercial products constitute endorsement or recommendation for use.

Pinto Abalone Recovery Project
2020 Final Report to the Skagit MRC
Josh Bouma, Puget Sound Restoration Fund
Paul Dinnel, Skagit MRC
September 30th, 2020

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in the San Juan Archipelago (SJA) even after the closure of the recreational fishery in 1994 (Rothaus et al. 2008, WDFW unpublished data).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have been observed and the average size of abalone continues to increase (Rothaus et al. 2008, Bouma et al. 2012, WDFW unpublished data).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 of these 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, and 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 Kessing (1999) estimated that the minimum density is 0.15 abalone/m^2 in order for successful

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

Steps are being taken in an effort 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, managers, students 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 WWU SEA Discovery Center in Poulsbo; the University of Washington, School of Aquatic & Fishery Sciences (UW) and others. Annual funding to PSRF from the Skagit County Marine Resources Committee (Skagit MRC-Skagit County contract #C20190432) supports abalone conservation aquaculture and restoration activities in Skagit County. This funding support for abalone restoration in 2019-2020 was supplemented by funding from WDFW and the Benjamin & Margaret Hall Family Foundation.

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, 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, 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 a hatchery, 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. Almost 13,700 of these healthy, genetically diverse hatchery produced juvenile abalone have been outplanted to eight rocky reef sites in Skagit County waters since 2009. An additional 11,300 abalone have also been outplanted at ten sites in San Juan 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 released into the wild (Carson et al. 2019).

Methods and results of surveys prior to 2020 can be found in annual WDFW, PSRF and Skagit MRC summary reports (Bouma and Dinnel 2017, 2018, and 2019) 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 time period from October 1st, 2019-September 30th, 2020.

Nursery Husbandry & Hatchery Production

Culture and husbandry of the 2018 juvenile and 2019 post-set cohorts was the primary activity within the abalone nursery from fall of 2019 to early spring 2020 in preparation for spring restoration outplants. Weekly care of these two year class populations in the nursery included gentle tank cleaning, feeding post-set abalone with three diatom species cultured in the hatchery and feeding juveniles macroalgae (*Palmaria mollis* cultured at the lab and *Nereocystis luetkeana* collected in the wild). Regular inventory data of nursery abalone were taken (numbers and shell lengths) to track survival and growth during the culture process. In 2019, a concerted effort was made to size sort and consolidate within families to keep similar-sized juvenile animals within tanks. The goal of this effort was to decrease within-tank competition, a possible contributor to low survival within tanks, and to stimulate faster growth rates. Bi-monthly inventories (numbers and shell lengths) were taken to track survival and growth during the culture process (Figure 1).

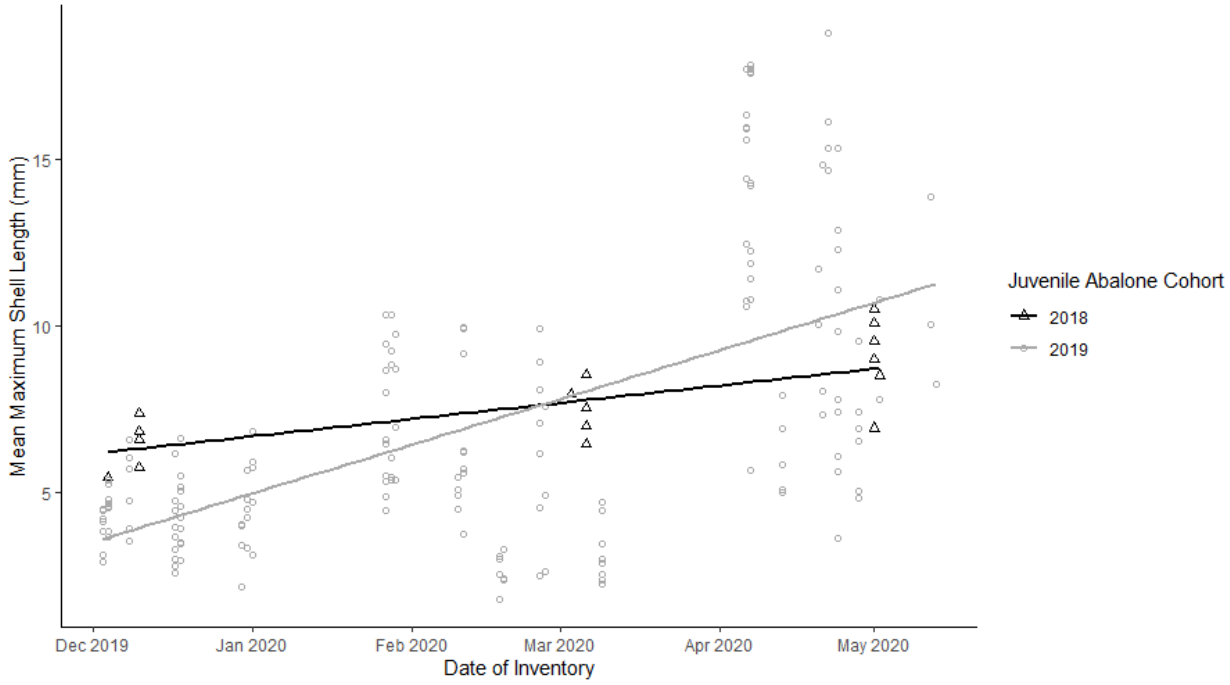


Figure 1. Mean SL (maximum shell length) of 2018 and 2019 cohorts of juvenile pinto abalone from December 2019 to May 2020. Each point represents N=30 individual measurements per unique family.

A significant portion of the 2019 cohort of juvenile abalone being reared at the Chew Center was not outplanted as expected during 2020 fieldwork for several reasons. Primarily, COVID-19 pandemic related hatchery staffing reductions and restrictions/limitations in the field inhibited site reconnaissance, necessary surveying and hatchery preparation to conduct a large-scale outplant. Additionally, results from monitoring efforts on recovery sites that were seeded with both the one year old and two year old cohorts were not conclusive that the younger year class can survive equally well and an additional year of this type of outplanting is necessary before outplanting the younger cohort at a broader scale.

From the 2019 abalone spawning efforts, almost 5600 juveniles from 17 families continue to be reared in the PSRF nursery (mean SL 13.7 mm), these will be available to be combined with the new 2020 cohort for outplanting in spring 2021. Also, and similar to efforts in the previous two years, PSRF delivered a batch of 2019s (n=1000) that will be reared for one year at the Port Townsend Marine Science Center (Table 1).

2019 Juvenile Abalone Remaining

Location	Family	Number of Animals		Mean Length (mm)
Moved to satellite facility for rearing				
	OR94 X GR09	118		20.53
	OR94 X OR85	92		20.06
	OR94 X OR97	82		10.72
	GR14 X OR29	363		14.93
	GR14 X OR50	198		17.73
	GR14 X OR55	217		18.49
		1,070		17.07
Remain at the Chew Center Shellfish Nursery				
	Joyride (mixed family)	103		13.49
	GR03 X GR11	93		21.32
	OR88 X GR05	246		14.86
	OR98 X OR87	50		11.19
	OR65 X GR08	242		11.25
	OR67 X GR08	776		8.85
	OR96 X GR08	782		10.07
	OR94 X GR12	185		11.97
	GR14 X OR45	1,403		7.79
	GR14 X OR57	97		8.26
	OR68 X OR57	544		10.81
		4,521		11.81
	Total 2019 cohort remaining	5,591	Average length	13.67

Table 1. Remainder of the 2019 juvenile abalone cohort not outplanted in 2020. About 1,000 juvenile abalone were sent to Port Townsend Marine Science Center and 4,521 remain at the Chew Center Shellfish Nursery for rearing until outplanting in spring 2021.

The 2020 pinto abalone spawning production season at the Chew Center began in early June and concluded with the last batch of larval settlement in mid-August. All five induced spawning attempts were successful and produced larvae for settlement in the nursery. Twenty-eight distinct novel families were created from seven females and 19 males and 1.6 million larvae were settled into 70 culture tanks (Table 2). An additional 1.1 million competent larvae from 2020 spawning events were not utilized for settlement and grow-out to avoid flooding the nursery with genetically homogeneous F1s along with lack of settlement tank space. PSRF aimed to produce as many new families as possible early in the 2020 season to ensure that nursery grow-out occurred successfully during optimal summer growing conditions with the end goal of having this cohort ready for outplanting in spring 2021 at an age under one year old.

Larvae set in 2020	*Female*								
Male	GR02	GR16	GR18	GR23	GR26	GR27	GR28	Grand Total	% of Grand Total
GR07		8,000				92,000		100,000	6.23%
GR09						91,800		91,800	5.72%
GR11						30,400		30,400	1.89%
GR12						70,398		70,398	4.38%
GR13			100,000		57,600			157,600	9.82%
GR17			75,000		48,600			123,600	7.70%
GR20	2,400							2,400	0.15%
GR24			75,000		40,800			115,800	7.21%
GR25				37,600				37,600	2.34%
OR14					43,200			43,200	2.69%
OR44				33,600	68,400			102,000	6.35%
OR46				75,000				75,000	4.67%
OR48			79,000		48,000			127,000	7.91%
OR50						85,500	9,600	95,100	5.92%
OR85				50,000				50,000	3.11%
OR86				50,000				50,000	3.11%
OR87			121,600	50,000				171,600	10.69%
OR91	2,000		60,800					62,800	3.91%
OR95			99,200					99,200	6.18%
Grand Total	**4,400**	**8,000**	**610,600**	**296,200**	**306,600**	**370,098**	**9,600**	**1,605,498**	
% of Grand Total	0.27%	0.50%	38.03%	18.45%	19.10%	23.05%	0.60%		100.00%

Table 2. Pinto abalone families produced at the Chew Center during the 2020 spawning season. Female broodstock ID is on the horizontal axis; male broodstock ID is on the vertical axis. Number of competent larvae per cross settled into nursery culture tanks is shown within each square.

Nursery Management

In addition to the general abalone husbandry routine, nursery system management for optimal animal health and grow-out also included regular filter changes, pump replacement, buffering system management, water chemistry probe cleaning/calibration/replacement and water quality data monitoring. During the late winter 2019, PSRF added an additional 300 gallon seawater reservoir in the nursery to support broodstock husbandry. This reservoir is connected via a venturi mixing valve and temperature controller to the primary heated seawater reservoir and incoming cold seawater supply allowing for separate manipulation of temperature to the broodstock tanks in the nursery. The new reservoir also has an independent pH controller, dosing pump and sodium carbonate reservoir for buffering capacity. This seawater supply upgrade allows PSRF to heat and buffer seawater during broodstock conditioning and gametogenesis, while simultaneously lowering the temperature and pH of the post-set tanks in preparation for juvenile outplanting in the spring (Figure 2).

r in the nursery to support broodstock husbandry.

Health & Disease

In January 2020, PSRF collected the annual hatchery health sample for disease screening from both the Chew Center nursery and the PTMSC satellite nursery: 130 live juveniles from each site representative of all the families produced during the 2019 spawning season and destined for outplanting during spring 2020. The sample from each location included 65 live individuals for histology and 65 live individuals for PCR analysis for withering syndrome. The resulting pathology reports from Dr. Ralph Elston, Aquatechnics Inc. concluded that no disease-causing pathogens were present in the samples and gave the hatchery abalone seed group a clean bill of health for introduction to outplant sites in the wild. Following the abalone hatchery health analysis and report, a new annual transfer permit for moving abalone from the Chew Center into the field was obtained from WDFW.

Abalone Broodstock

As of the end of the spawning season in 2020, PSRF has 40 broodstock animals (11 females, 29 males) at the Chew Center. Regular inventory, health and maintenance checks were conducted

Figure 3. A newly collected pinto abalone broodstock aboard the WDFW R/V Caliper.

on all broodstock, tanks were fed and cleaned weekly, and twice-yearly animals were measured, weighed and re-tagged as necessary. Due to COVID-19 restrictions, the annual broodstock collections were fewer and later in the season than normal. Generally occurring in early spring, the annual broodstock collections conducted by PSRF and WDFW divers in the San Juan Islands happened on two occasions, May 14 and May 16, 2020. Fourteen new animals (9 females, 5 males) were brought to the Chew Center in 2020 (Figure 3). One of the new males died prior to being used in any spawning events. This animal had visible lesions on its epipodium from handling and injury during collection. All incoming broodstock underwent the standard intake protocol: they were measured, weighed, ranked by gonad index, fouling sponges removed from the shell, animals tagged with vinyl disc tags and PIT tags, and general observation of overall health noted. Non-lethal genetic samples were taken using epipodial tentacle clips; this was done in early September upon completion of the spawning season to avoid additional stress on the animals prior to induced spawning attempts.

Juvenile Abalone Outplant Site Monitoring

Between February-May, 2020, PSRF divers participated with WDFW in surveys investigating survival, growth and emergence of hatchery reared pinto abalone at six of the eight restoration sites in Skagit County, located along the shorelines of Cypress, Burrows and Allan Islands. Two of the original Skagit County outplant sites were not surveyed this year; due to low survival at these two sites over multiple outplant years it was determined that they would not be overseeded in 2020. Two of the sites, newly established in 2019 along the shorelines of Washington Park were monitored for the first time since being outplanted one year ago.

Survey set-up included locating the four permanently marked plot corners, extending a survey tape measure around the plot to establish a perimeter, and installing weighted lines to distinguish 2 m survey lanes across the plot. Surveys also included the addition of the 2 m perimeter lane around the entire outside of each plot. This additional perimeter lane roughly doubles the amount of area surveyed at each site during a standard survey and provides informative emigration data. Divers meticulously conducted non-invasive surveys of each lane, including the full perimeter sweep. Dive lights were used to investigate cracks, crevices and

overhangs. The shell length and presence/absence of tags, if identifiable, were recorded for all abalone observed. Notes were also taken on where each observation was made within the plot lane (deep, mid or shallow) and how the animal was oriented within the substrate (cryptic, semi-cryptic or emergent). Empty abalone shells from mortalities were collected, measured and observed for tags when encountered and then removed from the plot.

The monitoring surveys at the six Cypress, Burrows and Allan Island sites in 2020 found a total of 314 abalone of which 29% were found outside of the plot boundaries (Table 3). The largest number of abalone (n=116 combined on and off plot) were found at Gold and the plot density at this site was 0.89 abalone/m^2. Omaha has typically been the site with the highest density but conditions and circumstances limited monitoring at this site to a partial survey. COVID-19 pandemic quarantine delayed the survey at Omaha until late May, at which point the site was completely covered in both understory and canopy kelp, severely decreasing survey efficiency. Mean shell length of observed abalone for all six plots combined was 91.1 mm. The overall mean density of observed abalone on-plot was 0.49/m^2 with individual plots ranging from 0.23 to 0.89 abalone/m^2 (Table 3). The four longer-term sites (Gold, Utah, Omaha and Baytown) had not been overseeded since 2017 as is evidenced by the larger mean shell lengths between 91-109 mm at these sites. The number of abalone observed at Switchback and Vitality (newly established and seeded for the first time in 2019) was lower than the anticipated average one year survival of 10%; this was true for both the younger and older cohorts outplanted to these sites. Surveys will be conducted again at Switchback and Vitality in early 2021 two years post-outplant to determine if surviving juveniles were cryptic or if site selection was not ideal at these two locations.

Site	Plot Area (m^2)	On Plot (n)	Plot Density (Ab/m^2)	Perimeter Area (m^2)	Off Plot (n)	Perimeter Density (Ab/m^2)	Mean SL (mm)
Gold	98.2	87	0.89	96.8	29	0.30	97.9
Utah	102.5	51	0.50	97.2	14	0.14	108.8
Omaha*	40.0	18	0.45	18.0	17	0.94	100.3
Baytown	79.3	31	0.39	162.6	24	0.15	91.1
Switchback**	73.0	20	0.27	73.0	5	0.07	37.9
Vitality**	64.0	15	0.23	67.0	3	0.04	40.1
Total	**457.0**	**222**	**0.49**	**514.6**	**92**	**0.18**	**91.1**

Table 3. Juvenile abalone outplant survey data collected at six sites in Skagit County from February-May 2020. SL=maximum shell length measurement. *The Omaha survey was a subsample due to conditions and only 2 of 5 lanes within the plot and a portion of one perimeter lane were surveyed. **Switchback and Vitality are newly established sites that were outplanted for the first time in 2019.

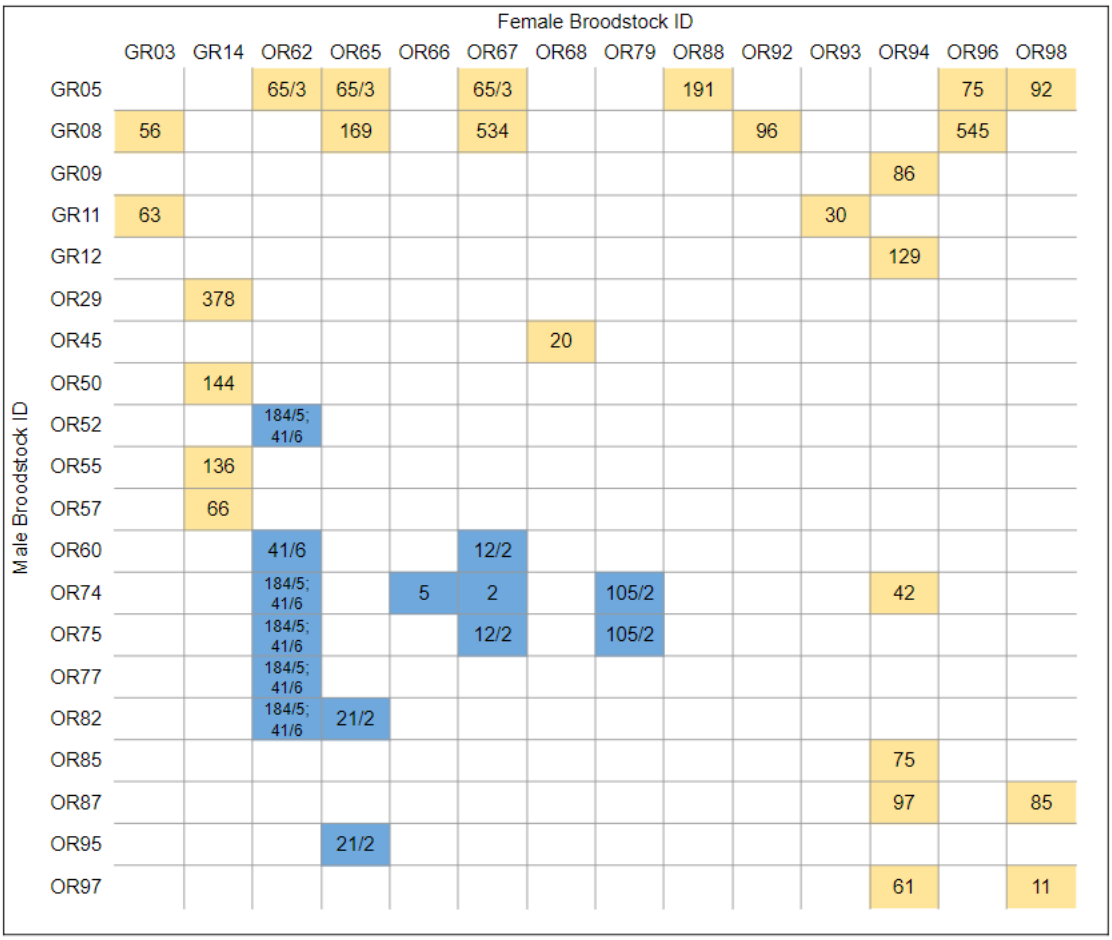
Juvenile Abalone Outplanting

In May and June 2020 and in collaboration with WDFW subtidal shellfish biologists, the PSRF team completed the 10th juvenile abalone outplant over the past 12 years at recovery sites in the San Juan Archipelago. 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, nearly 26,000 pinto abalone have been outplanted to 18 different restoration sites in the San Juan Islands; 13,700 of these healthy, genetically diverse hatchery produced juvenile abalone have been outplanted to eight rocky reef sites in Skagit County waters.

Description of juvenile outplant groups

3454 juvenile abalone representing 31 genetically distinct families from the 2018 and 2019 Chew Center cohorts were seeded to six existing sites in the San Juan Islands; this included 2302 abalone that were seeded to four sites in Skagit County and 1152 abalone that were seeded to two sites in San Juan County (Table 3). The Skagit County restoration sites seeded in 2020 included Gold, Omaha, Utah and Baytown. The average number of abalone outplanted per site and mean shell length for these four Skagit County sites was 576 and 14.7 mm respectively. The newest two sites created in Skagit County in 2019 were not overseeded in 2020 due to disappointing yet inconclusive survival results observed during 2020 monitoring efforts.

In preparation for outplanting, PSRF staff conducted an inventory, collected shell length data and sorted families into outplant groups. Additionally, all juveniles from the 2018 cohort were marked with a colored glue dot before outplanting to identify them from the younger group (Coraffix cyanoacrylate adhesive mixed with Eye Candy mica powder "Baku Red" pigment) using the same methods as the previous year.



Male Broodstock ID \ Female Broodstock ID	GR03	GR14	OR62	OR65	OR66	OR67	OR68	OR79	OR88	OR92	OR93	OR94	OR96	OR98
GR05			65/3	65/3		65/3			191				75	92
GR08	56			169		534				96			545	
GR09												86		
GR11	63										30			
GR12												129		
OR29		378												
OR45							20							
OR50		144												
OR52			184/5; 41/6											
OR55		136												
OR57		66												
OR60			41/6			12/2								
OR74			184/5; 41/6		5	2		105/2				42		
OR75			184/5; 41/6			12/2		105/2						
OR77			184/5; 41/6											
OR82			184/5; 41/6	21/2										
OR85												75		
OR87												97		85
OR95				21/2										
OR97												61		11

Table 3. Pinto abalone families produced at the Chew Center and outplanted in 2020. Female broodstock ID on horizontal axis; males on vertical axis. Number of individuals outplanted per cross is shown within each square (blue = 2018 progeny; yellow = 2019 progeny). Numbers with a slash represent pooled groups of multiple crosses. Total number of seed outplanted: 3,454 (3,616-162 mortalities post-sort).

The 2020 outplant effort was similar to 2019 in that both a younger and older year class were overseeded together onto the same sites. Mean shell length of the 2018 and 2019 cohorts at all six sites was 10.8 mm and 15.4 mm respectively. Following a 2.5-year study completed in February 2019 by PSRF and WDFW to investigate survival and growth of younger-aged outplants, future outplant strategies were to shift towards releasing hatchery juveniles before they reach one year in age. This could substantially improve annual production cycles in the Chew Center. The 2020 outplants represent another opportunity to test and monitor the two cohorts over the next year to confirm that survivorship of the younger group is similar to the older group.

Outplant Modules, Loading and Transport

Outplant modules were made from PVC pipe (6" ID, repurposed from a wetlands project) cut into approximately 18" long sections. Most of the tubes this year were recovered from previous outplants and reused. Each section of tube was power washed and scrubbed. Tubes were numbered by site destination for convenience once in the field. All tubes were conditioned in flow-through tanks at the Manchester lab for several weeks prior to being loaded with animals.

One day prior to outplant, abalone were transferred from their holding tanks at Manchester into the PVC outplant tubes. All the tubes destined for a particular site were positioned upright in a large seawater-filled fish cooler with one layer of fiberglass window screen (2 mm mesh size) secured with rubber bands covering the bottom end. Approximately 100 abalone were loaded into each tube. The open end of the tube was then closed with one layer of window screen and secured with a rubber band. Modules were then placed back into a flow-through seawater tank at the Chew Center for holding overnight.

The following morning, modules were transferred into a seawater filled 4'x 4' fish tote in the back of a 5'x 9' U-Haul towable trailer (Figure 4) and transported from Manchester to Port Townsend. The large fish tote in the trailer was aerated during transport using a 12V battery, DC to AC inverter, an aquarium pump and two large air stones placed in the tote. The trailer was drained at the marina and abalone modules were transferred into a waiting seawater filled, aerated fish tote on board the WDFW R/V Caliper.

Once at outplant sites, tubes were bound together with webbing straps and quick release buckles in bundles of three and carried to depth by divers. Tubes were placed within the delineated outplant plots in areas that appeared to have suitable substrate and cryptic habitat onto which juvenile abalone could exit. Tubes were wedged amongst cobble and boulders to secure them against current and surge. Once the tubes were secured within the substrate, divers immediately removed the mesh from the tubes and the abalone were free to move from the modules out onto the surrounding

Figure 4. PSRF personnel send juvenile abalone on their way to restoration outplants in spring 2020.

substrate. PVC outplant modules will be collected during site surveys in early 2021, cleaned and reused during future outplant efforts.

During the first outplant of 2020, to test the stress of transport to restoration sites, a group of about 100 animals from the 2019 cohort were exposed to all the same stressors of the outplanting process in a separate module. This included sorting, transport via the fish tote in a U-haul, boat ride and even being carried down to depth by divers. This test module was then returned to the Chew Center nursery to be monitored for any deleterious effects. Two animals suffered crushed shells when trapped between mesh screens secured to the outplant tube during transport. However, after two months of monitoring post-outplant, no additional mortalities have been reported from this control group.

During two outplant efforts, temperature loggers, one placed inside the tote, and the other placed inside the outplant tube, tracked temperature for the duration of the outplant day. Logger data from both outplant efforts in 2020 (Fig. 5) show that juvenile abalone experienced a 2-3^{o} C temperature drop over the course of eight hours, from the time they were removed from nursery tanks at the Chew Center to the time the modules were secured within the rocky reef substrate at an outplant site. Temperature changes occurred when the abalone were transferred from the U-haul tote into fresh seawater in the dive vessel tote and also when the abalone were moved from the tote and into the ocean by divers. These temperature stressors do not appear to be detrimental to abalone health during the outplant process.

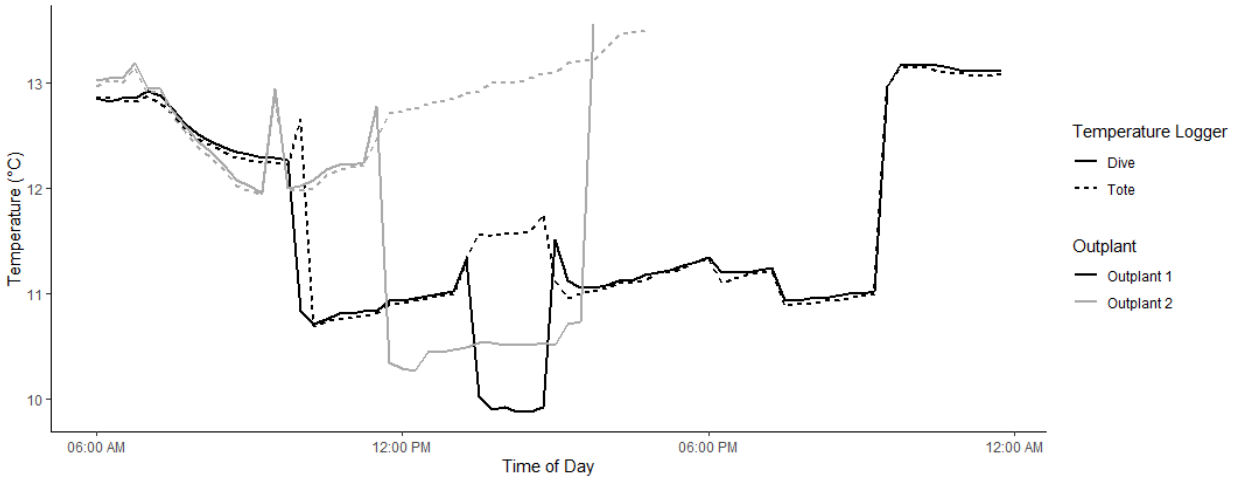


Figure 5. Temperature logger data during two juvenile outplanting events, spring 2020. During each event, one temperature logger (tote) stayed on the boat during the entire outplant process and the other logger (dive) travelled with a diver to depth at the restoration site. Outplant 1 loggers tracked temperature from start of trip at the hatchery, to outplant site, and back to hatchery. Outplant 2 loggers tracked temperature from start of trip at hatchery and concluded at the end of the outplant dive.

Outreach

PSRF focused on the following pinto abalone related outreach, funding support and collaboration building efforts during this annual contract period:

- PSRF composed several quarterly newsletters that highlighted abalone related accomplishments. This newsletter is distributed to PSRF supporters, board members, project partners and other collaborators.

- PSRF organized a session for the Salish Sea Ecosystems Conference, which before being cancelled due to COVID-19 was to be held in Vancouver, Canada in April 2020. The session was titled "Pinto abalone research and recovery: use of conservation aquaculture and other strategies to give the Salish Sea's largest rocky-reef-cleaning native marine snail a population boost". PSRF had recruited six presentations for this pinto abalone session including topics on genetics, ocean acidification, sperm cryopreservation, larval outplanting, juvenile outplant survival, the State's Recovery Plan and satellite rearing partnerships.

- Met with NOAA and WDFW collaborators in November 2019 to share updates on the pinto abalone genetics project developing new markers using RADseq in order to compare genetic diversity of outplant sites to wild populations and the historic broodstock population.

- Met with Dr. Jackie Padillo-Gamino from UW and student Eileen Bates in November 2019 to discuss next steps in Eileen's research on effects of ocean acidification on early life stage pinto abalone.

- Josh Bouma gave a presentation at the Puget Sound Partnership Leadership Council meeting in Bremerton in December 2019 highlighting pinto abalone restoration and research efforts in Washington.

- Seattle Aquarium and PTMSC satellite rearing partnership development.

Acknowledgments

This final report summarizes work accomplished by PSRF in coordination with Paul Dinnel, Skagit MRC's Project Lead, and Tracy Alker, Skagit MRC Administrative Coordinator, through September 30th, 2020. This project has been funded wholly or in part by the United States Environmental Protection Agency via the Puget Sound Partnership and the Northwest Straits Commission under assistance agreement Grant SEANWS-2019-SkCoPW-00003 to Skagit County. The contents of this document do not necessarily reflect the views and policies of the United States Environmental Protection Agency, nor does mention of trade names or commercial products constitute endorsement or recommendation for use.

References

Allee, W.C., A.E. Emerson, O. Park, T. Park and K.P. Schmidt. 1949. Principles of Animal Ecology. Saunders Publishing Co., Philadelphia, PA.

Babcock, R. and J. Keesing. 1999. Fertilization biology of the abalone *Haliotis laevigata*: laboratory and field studies. Can. J. Fish. Aq. Sci. 56:1668-1678.

Bergman, D. 2009. Pilot out planting of the abalone, *Haliotis kamtschtkana*, and the effects of predation on hatchery tagged individuals. Final Report for the 2009 Research

Experience for Undergraduates program, Shannon Point Marine Center, Western Washington University. 26 pp.

Benolkin, A., A. Thomson, P. Dinnel and N. Schwarck. 2012. Survey of previously out planted pinto abalone (*Haliotis kamtschatkana*) and an exploration of an optimal weaning diet. Final Report for the 2012 Research Experience for Undergraduates program, Shannon Point Marine Center, Western Washington University. 20 pp.

Bouma, J.V. 2007. Early life history dynamics of pinto abalone (*Haliotis kamtschatkana*) and implications for recovery in the San Juan Archipelago, Washington State. MS Thesis, School of Aquatic and Fishery Sciences, Univ. Wash., Seattle, WA.

Bouma, J.V., D.P. Rothaus, K.M. Straus, B. Vadopalas and C.S. Friedman. 2012. Low juvenile pinto abalone (*Haliotis kamtschatkana*) abundance in the San Juan Archipelago, Washington state. Transactions of the American Fisheries Society 141:76-83.

Bouma, J.V. and P.A. Dinnel. 2017. Pinto abalone monitoring and restoration: Pinto abalone final report. Final Report for Skagit County Marine Resources Committee, Mount Vernon, WA by Puget Sound Restoration Fund, Bainbridge Island, WA. 19 pp.

Bouma, J.V. and P.A. Dinnel. 2018. Pinto abalone monitoring and restoration: Pinto abalone final report. Final Report for Skagit County Marine Resources Committee, Mount Vernon, WA by Puget Sound Restoration Fund, Bainbridge Island, WA. 20 pp.

Bouma, J.V. and P.A. Dinnel. 2019. Pinto abalone monitoring and restoration: Pinto abalone final report. Final Report for Skagit County Marine Resources Committee, Mount Vernon, WA by Puget Sound Restoration Fund, Bainbridge Island, WA. 17 pp.

Carson, H.S., D.J. Morin, J.V. Bouma, M. Ulrich and B. Sizemore. 2019. The survival of hatchery-origin pinto abalone *Haliotis kamtschatkana* released into Washington waters. Aquatic Conservation: Mar. Freshwater Ecosyst. 2019:1-18.

Ebert, T.B. and E.E. Ebert. 1988. An innovative technique for seeding abalone and preliminary results of laboratory and field trials. Calif. Fish. Game 74(12):68-81.

Hester, J.B., J.M. Walker, P.A. Dinnel and N.T. Schwarck. 2011. Survey of previously out planted pinto (northern) abalone (*Haliotis kamtschatkana*) in the San Juan Archipelago, Washington State. Pp. 22-28 in: Diving for Science 2011, Proceedings of the American Academy of Underwater Sciences 30th Symposium, Dauphin Island, AL. (Pollock, N.W.,

editor). Also, Final Report for the Research Experience for Undergraduates (REU) program, Shannon Point Marine Center, Western Washington University, Anacortes, WA. 9 pp. + Appendix.

NOAA (National Oceanographic and Atmospheric Administration). 2007. Species of concern: Pinto abalone. NOAA, National Marine Fisheries Service. http://www.nmfs.noaa.gov/pr/species/concern.

Pratt, P. and P. Dinnel. 2010. Survey of previously out planted abalone, *Haliotis kamtschatkana*, and effects of weaning diets on growth rates of hatchery individuals. Final Report for the 2010 Research Experience for Undergraduates program, Shannon Point Marine Center, Western Washington University. 19 pp.

PSRF (Puget Sound Restoration Fund). 2014. Recovery plan for pinto abalone (*Haliotis kamtschatkana*) in Washington State. Final Report, Puget Sound Restoration Fund, Bainbridge Island, WA. 50 pp.

Rothaus, D., B. Vadopalas and C. Friedman. 2008. Precipitous declines in pinto abalone (*Haliotis kamtschatkana kamtschatkana*) abundance in the San Juan Archipelago, Washington, USA, despite statewide fishery closure. Canadian Journal of Fisheries and Aquatic Sciences 65: 2703-2711.

Walker, J., N. Schwarck, V. Hodges, T. Tymon, A. Thomson, K. Gabrian-Voorhees and P. Dinnel. 2013. Survey of previously out planted abalone (*Haliotis kamtschatkana*) at the West Allan Island out plant site, August, 2013. Final Report by Shannon Point Marine Center, Western Washington University for Washington Department of Fish and Wildlife, Olympia.