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## IMPLEMENTING A RESTORATION PROGRAM FOR THE ENDANGERED WHITE ABALONE (*HALIOTIS SORENSENI*) IN CALIFORNIA

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**ABSTRACT** A restoration program including wild population surveys, captive breeding, health monitoring, recovery site preparation, and recovery modeling has been implemented to restore white abalone (*Haliotis sorenseni*) populations in California. White abalone once supported a lucrative fishery and are now endangered, nearing extinction at less than 1% of baseline abundances. Recent deep water surveys indicate that populations continue to decline with no signs of recruitment, despite the closure of the fishery in 1996. Four sites with artificial reefs ( $n = 12/\text{site}$ ) in optimal white abalone habitat were established. No wild white abalone have been found at these sites. Captive abalone were spawned in the spring of each year from 2012 to 2015. Each year, the production of 1-y-old abalone has increased in the captive breeding program from approximately 20 in 2012, to 150 in 2013 and an estimated 2,000 in 2014. In 2015, the breeding program reached two milestones: (1) most successful spawning season to date and (2) the hatchery distributed 200 captive-reared abalone to 4 partner institutions within the White Abalone Recovery Consortium (WARC). The WARC is made up of federal and state agencies, universities, public aquaria, and aquaculture organizations, all committed to white abalone restoration. The next steps for the program include expanding the captive breeding program to increase production, monitoring abalone health and genetic diversity, and conducting stocking studies to enhance growth and survival in the ocean. The goal of the stocking program is to create a reproductive population in the wild to bring white abalone back from the brink of extinction.

**KEY WORDS:** captive propagation, endangered species, marine conservation, partnership, recovery

The more we take,  
The more they make  
In deep sea matrimony  
Race suicide can not betide  
The fertile abalone  
(Jack London & George Sterling, 1907)

### INTRODUCTION

The white abalone *Haliotis sorenseni* was the first marine invertebrate on the U.S. federal endangered species list [National Oceanic and Atmospheric Administration (NOAA 2001); Federal Register 65 FR 2616, and 66 FR 29046]. Despite conservation measures, such as closing the fishery in 1996, the species is on the edge of extinction (Davis et al. 1998, Lafferty et al. 2004). Fishing has been identified as the primary cause of the decline (Hobday & Tegner 2000), despite the belief held for decades that as abalone are so fertile fishing could never lead to extinction (Lundy 1997). Remnant white abalone populations at offshore banks are now declining at an alarming rate (Stierhoff et al. 2012), consistent with natural mortality estimated for similar species (Leaf et al. 2007, Button & Rogers-Bennett 2011). Population estimates at the most abundant site have declined from 15,000 in 2002 to 3,000 in 2010 (Stierhoff et al. 2012). Estimates of baseline abundances suggest legal-sized white

abalone once numbered greater than 360,000 in California (Rogers-Bennett et al. 2002). In Mexico, populations are also thought to have declined dramatically from 1972, when density estimates showed low populations in Baja California (Guzmán del Próo 1992). Recovery plans by both the State of California (Abalone Recovery and Management Plan 2005) and the federal NOAA Fisheries highlight the need for a captive breeding program to produce healthy animals for stocking into the ocean as the number one strategy for restoration.

Today, threats to the species in the wild are 2-fold: (1) natural mortality due to an aging wild population and (2) reproductive failure due to extremely low population densities. In the white abalone status review, Hobday and Tegner (2000) warned that existing white abalone were old (large) and that the population would disappear as a result of natural mortality without human intervention. Evidence from bomb carbon research shows the life span for white abalone is just 28–30 y, underscoring the concern that the population is now dying of old age. There have been no signs of successful white abalone recruitment in the wild for over 30 y with the exception of one juvenile found in an artificial reef at Santa Cruz Island in 2001 (Rogers-Bennett et al. 2004).

Low population densities (<1–2 abalone/ha) translate into large distances between spawning individuals (>20 m), which is considered as the primary mechanism responsible for reproductive failure (Haaker et al. 1994, Davis et al. 1996, Davis et al. 1998, Behrens & Lafferty 2005). Abalone are broadcast spawners, requiring males and females to be less than 4 m apart for eggs to be fertilized (Babcock & Keesing 1999). This type of Allee

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effect impacts populations at low densities, which can suffer recruitment failure below minimum viable population (MVP) densities (Shepherd & Brown 1993). Without human intervention in the form of a recovery program including a captive breeding program and stocking juveniles into suitable habitats, the wild population is predicted to go extinct in less than 20 y (Catton et al. 2016). Therefore, a captive white abalone recovery program was established in California. Initial efforts to produce white abalone in captivity were successful but plagued by mortalities due to the abalone disease withering syndrome, which was subsequently treated successfully with an antibiotic regimen (Friedman et al. 2007). Today, most of the broodstock in the captive breeding program came from this early success, and they are maintained in filtered, ultraviolet (UV)-sterilized, cool (14°C) seawater conditions along with rigorous health monitoring.

White abalone, including first- and second-generation captive bred animals, were spawned in the spring of 2012 to 2015 at five partner facilities. Fertilized embryos were transported to the hatchery at UC Davis Bodega Marine Laboratory, and the larvae were cultured and then settled onto diatom-covered substrates. Juveniles were cultured to increase the number of individuals held in captivity and for future stocking in the ocean. Artificial reefs were established along the mainland and Channel Islands to survey for wild white abalone and prepare for stocking in southern California. The next steps for the captive white abalone program in California are discussed as well as the exceptional partnership developed between federal,

state, academic, and private institutions, including aquaria and abalone aquaculturists, all dedicated to saving this endangered species.

## MATERIALS AND METHODS

### Study Sites

Four sites were established in deep water in southern California to monitor white abalone and prepare for future restoration actions (Fig. 1). The first site was established at a depth of 23 m on Santa Cruz Island (33° 59' N 119° 32' W) in 2009. The Santa Cruz Island site consists of low-relief cobble and boulder substrate interspersed with sand channel and rock ledges and can have dramatic surface currents. Extensive giant kelp (*Macrocystis pyrifera*) beds dominate the canopy, with genera such as *Eisenia*, *Laminaria*, and *Pterygophora* forming the subcanopy and a moderate cover of crustose coralline algae forming the benthos. The second site was established at Santa Catalina Island (N 33° 26' W 118° 29') at a depth of 20 m. The rocky reef substrate at this site is variable, ranging from high to low relief with boulders and cobbles. Giant kelp canopies dominate the area, with subcanopy, foliose, and turf algae present (*Eisenia*, *Laminaria*, *Pterygophora*), as well as a high abundance of crustose coralline algae. The San Diego study area (N 32° 49' W 117° 17'), at a depth of 20–22 m, is characterized by a low-relief sandstone and mudstone substrate with scattered boulders and extensive ledge systems and is also dominated by

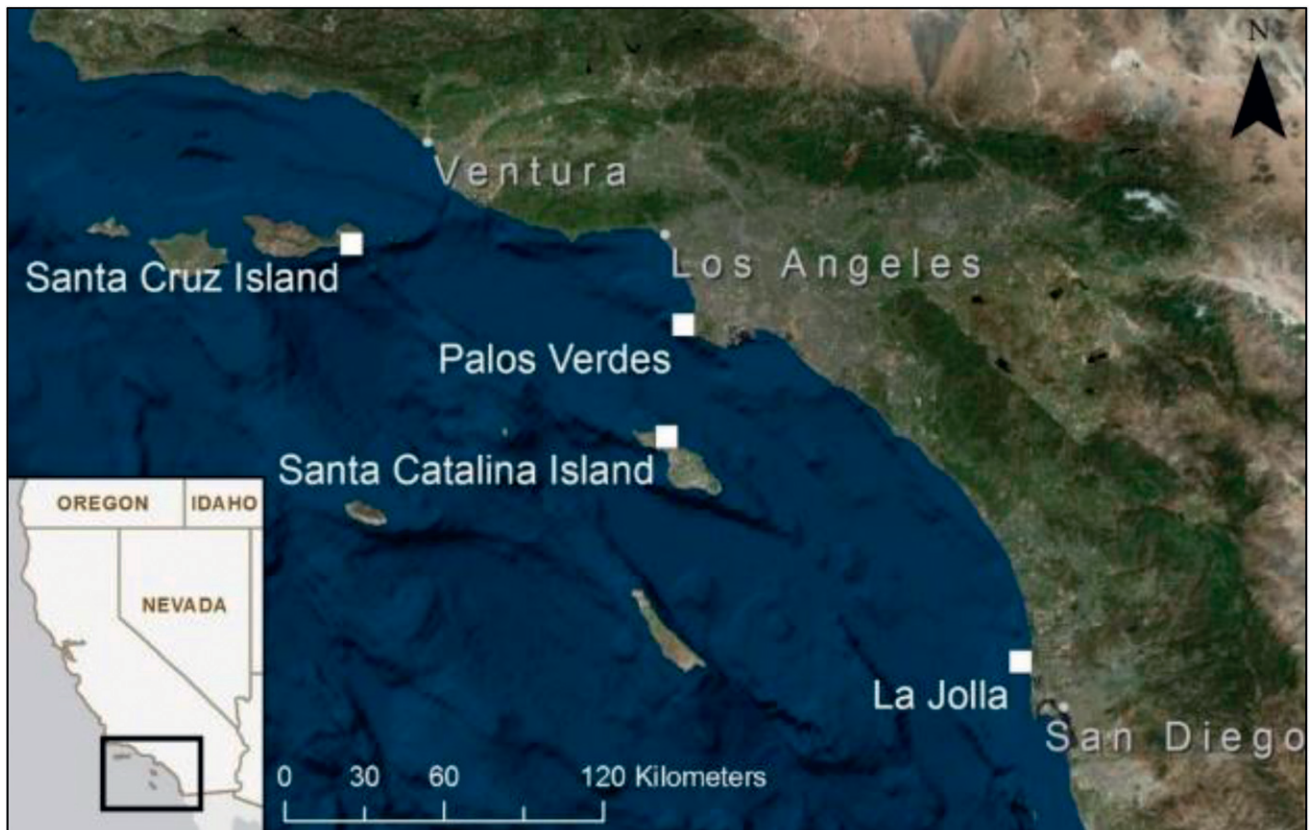


Figure 1. Map of southern California with juvenile abalone recruitment traps shown at Santa Cruz Island, Santa Catalina Island, Los Angeles, and San Diego.

giant kelp, with holdfasts often measuring over a meter in diameter. Clearings in the giant kelp are dominated by understory kelp species (*Laminaria*, *Pterygophora*, and *Eisenia*). The fourth site along the mainland of Los Angeles (N 33° 44 W 118° 25) in 20 m consists of primarily low-to-moderate relief rocky reef substrate with sand channels separating reef systems, and boulders and cobbles usually at reef and sand interfaces. Giant kelp canopies dominate the area, whereas other subcanopy, folios, and turf species are limited (*Eisenia*, *Laminaria*, *Cystoseira*, erect coralline, other fleshy red algae).

#### White Abalone Dive Surveys

Artificial reefs were established at these four sites in southern California. At each site, 12 artificial reefs were deployed in 3 groups of 4 reefs. The reefs were made of modified lobster traps filled with cinder blocks cut into 4 parts, for a total of 32 quarter blocks per reef. The surface area within each reef was estimated to be 4.6 m<sup>2</sup>. Reefs were placed in the ocean for 1 y prior to its first survey. Reefs were surveyed once a year (weather and funding permitting) by removing the lids and taking out the quarter cinder blocks. Each of the blocks was examined by a diver for the presence of abalone.

#### Spawning and Larval Culturing in the Hatchery

Spawning of adults was conducted using standard hydrogen peroxide protocols (Morse et al. 1977). Briefly, adult white abalone were placed into individual buckets with hydrogen peroxide sea water solution. Eggs were fertilized with sperm and the embryos were cultured at low densities (<20/ml) until they were competent to settle. Competent larvae were induced to settle onto diatom-covered tiles using 1 μM gamma-aminobutyric acid solution. Newly settled abalone consumed diatoms and were then weaned onto macroalgal diets at 4–5 mo (Aquilino et al. in prep.). The growth and survival of the juvenile white abalone were determined annually.

#### Abalone Health Monitoring

Abalone were maintained in 5-μm filtered, UV-sterilized seawater at 14°C to maintain abalone free from the bacteria that can cause withering syndrome in warm water conditions. In addition to water quality measures, the shells of the adult abalone were waxed approximately twice yearly to suffocate shell boring organisms such as the polychaete *Polydora* spp.

#### Restoration Modeling

Deterministic population models were constructed to quantitatively assess the status of and potential risks to the wild white abalone populations.

#### Larval Temperature Experiments

To explore the relationship between temperature and development, white abalone larvae were grown in larval chambers in four different temperature treatments. Temperature treatments for red abalone ranged from 9°C to 16°C. Four replicate static chambers were filled with 1,500 ml of sterilized UV-treated seawater filtered to 1 μm. The white abalone larvae used in this experiment were fertilized on March 2, 2013. Approximately 4,500 larval abalone or 3 abalone/ml were stocked into the

TABLE 1.  
Number of white abalone found inside artificial reefs in southern California from 2010 to 2015.

Year	Santa Cruz	Catalina	La Jolla	Los Angeles
2010	0	–	–	–
2011	0	–	–	–
2012	ND	0	–	–
2013	ND	0	0	–
2014	0	0	0	–
2015	0	0	0	0

Twelve artificial reefs are at each of the four sites. ND, no data for that site for that year; a dash - indicates the site was not yet established.

culture jars on day after 3 days of development at 12°C. Clean jars and water replacement occurred once per day. Larvae were retained with a 100-μm sieve, being careful not to pin larvae on the mesh. Developmental stage was assessed for 10 larvae per replicate using a dissecting microscope.

## RESULTS

#### White Abalone Reef Surveys

No white abalone were found in the artificial reefs during the annual dive surveys (Tables 1 and 2, Fig. 2). Four other species of abalone were found in the reefs, including red (*Haliotis rufescens*), pink (*Haliotis corrugata*), threaded (*Haliotis kamtschatkana assimilis*), and green abalone (*Haliotis fulgens*). Sea urchins were common in the reefs, including Coronado urchin (*Centrostephanus coronatus*), red urchin (*Mesocentrotus franciscanus*), and purple sea urchin (*Strongylocentrotus purpuratus*), along with numerous other marine invertebrates.

#### Spawning, Larval, and Juvenile Culturing

During all 4 y, adult white abalone spawned successfully with embryos surviving the larval stage to settle and grow to juveniles (Fig. 3). White abalone showed a strong degree of synchrony in spawning readiness during the spring, as has been seen in the past in wild populations (Tutschulte & Connell 1981). It was found that white abalone larvae reared in warmer temperatures developed more quickly and became competent to settle earlier than larvae reared at the cooler temperatures (Fig. 4), as has been observed previously for this and other species (Leighton 1972). The development of eye spots was a good indicator of normal development and cephalic tentacles developed when larvae were competent to settle. At 9°C, larvae survived but did not develop and did not reach competency within 10 days (Fig. 4). The captive breeding program successfully increased the production of 1-y-olds each year (Fig. 5).

#### Abalone Health

The program's focus on disease prevention has been successful in maintaining abalone health. In the hatchery, sterilized seawater coupled with temperature control (14°C) has been used to maintain adults and juveniles free from Withering Syndrome. The waxing of the shell of broodstock across

TABLE 2.  
Descriptions of the sites selected for artificial reefs in southern California.

Site	Depth (m)		Macroalgal assemblage		
			Canopy	Sub-Canopy	Benthos
Santa Cruz	23	Low-relief cobbles and boulders sand channels rock ledges	Extensive giant kelp ( <i>Macrocystis pyrifera</i> )	Moderate <i>Eisenia</i> , <i>Laminaria</i> , <i>Pterygophora</i>	Moderate cover of crustose coralline algae
Catalina	20	High-to-low relief boulders cobbles	Extensive giant kelp ( <i>M. pyrifera</i> )	Moderate <i>Eisenia</i> , <i>Laminaria</i> , <i>Pterygophora</i>	High cover of crustose coralline algae
San Diego	20–22	Low-relief sandstone mudstone; few boulders; extensive ledges	Extensive giant kelp ( <i>M. pyrifera</i> )	Moderate <i>Eisenia</i> , <i>Laminaria</i> , <i>Pterygophora</i>	Limited cover of crustose coralline algae
Los Angeles	20–22	Low-relief sandstone; low-relief reef; boulders; extensive ledges	Extensive giant kelp ( <i>M. pyrifera</i> )	Limited <i>Eisenia</i> , <i>Laminaria</i> , <i>Pterygophora</i>	Limited cover of crustose coralline algae

multiple facilities has also helped reduce mortality associated with shell boring organisms.

#### Restoration Modeling

Population models were constructed to aid in planning for restoration stocking efforts. The results of the models inform multiple aspects of the restoration program, including juvenile production goals, site selection, stocking performance measures, and expected timeframes for monitoring. The inclusion of stocked juveniles in the model enhances the population and in some model scenarios can lead to sustained population growth (Catton et al. 2016).

#### White Abalone Recovery Consortium

The White Abalone Recovery Program has developed a consortium of partners dedicated to white abalone recovery efforts in California. The White Abalone Recovery Consortium (WARC) is composed of federal NOAA National Marine Fisheries Service Protected Resources and Southwest Fisheries Science Center; state entities, including the California Department of Fish and Wildlife, Marine Region and Fisheries Branch as well as the University of California Davis and University of California Santa Barbara; public aquaria, including the Santa Barbara Museum of Natural History Sea Center, Cabrillo Marine Aquarium, and Aquarium of the Pacific; and California's aquaculture industry, including The Cultured Abalone and The Abalone Farm (Fig. 6). The WARC has partnered to share expertise, hold captive white abalone, conduct joint spawning events, and raise juvenile abalone. In September of 2015 the WARC reached an important landmark in the recovery program, with the hatchery producing enough abalone to share white abalone ( $n = 200$ ) among partner facilities for grow out and expansion of the breeding program.

## DISCUSSION

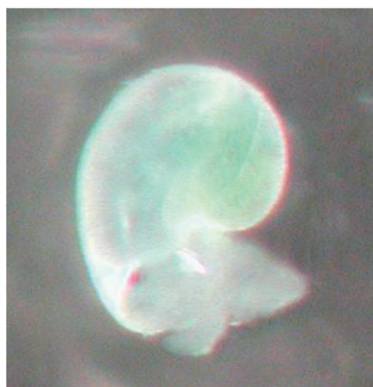
#### Status of White Abalone Populations

White abalone populations in the wild are declining at an alarming rate with populations so low that no successful

reproduction has been observed in decades. Artificial reefs designed to facilitate quantifying the abundance of small abalone were installed. No white abalone were found in or near artificial reefs established at the four sites in southern California (Table 1). Because other species of abalone were detected in the artificial reefs, it is suggested that these artificial reefs provide suitable juvenile abalone habitat. Furthermore, no white



Figure 2. Photograph of diver D. Stein surveying the juvenile abalone recruitment traps for abalone at Santa Catalina Island, May 2015.



A



B



C

**Figure 3. Photographs showing cultured white abalone (A) larvae (B) newly settled juvenile and (C) 1-y-old juveniles.**

abalone have been found in similar artificial reefs at six other sites (>72 reefs) monitored each year by the Kelp Forest Monitoring Program (D. Kushner, personal communication). The one exception was a single small white abalone observed in artificial reefs at Santa Cruz Island in 2001 suggesting the depth of the artificial reefs is appropriate for white abalone juveniles (Rogers-Bennett et al. 2004). This observation led to the selection of Santa Cruz Island for the current work. This is the only small white abalone known, which has been observed in the past 25 y in the wild on natural or artificial reefs. In summary, the results from this subtidal dive work, and those of other scuba monitoring programs in California, indicate that there is total recruitment failure of white abalone in the wild.

This result makes the captive breeding program all the more important.

Recruitment failure is coupled with very low numbers of adult white abalone detected using both remote operated vehicles (ROV) and diver searches in southern California. Population estimates made using ROV surveys of the site with the highest population, a remote offshore bank, have declined from 15,000 in 2002 to 3,000 in 2010 (Stierhoff et al. 2012). This decline is approximately 14% per year (Catton et al. 2016), which is comparable to natural mortality rates estimated for similar species (Leaf et al. 2007, Button & Rogers-Bennett 2011). This supports the hypothesis that there is total recruitment failure in this species. Dive surveys over the past 5 y

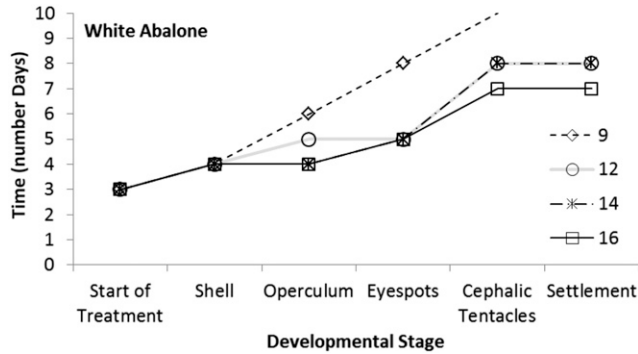


Figure 4. Graph showing the developmental stage of white abalone larvae at four different temperatures ranging from 9°C to 16°C starting the temperature treatment at day 3 of development. Please note the arrested development of the larvae at 9°C.

targeting white abalone habitat have revealed 30 observations of adult abalone (D. Witting and M. Neuman, unpublished data). This may be as many as 30 individuals, many of which may be single animals suitable for inclusion in the breeding program. The speed of the decline from 2002 to 2010 with the lowest numbers estimated from 2014 surveys (Stierhoff et al. 2014) makes restoration work all the more critical.

Low population densities in broadcast spawners, such as abalone, can have dire population consequences leading to fertilization failure. Experimental work has shown that male and female abalone need to be closer than 3 m for fertilization to occur (Babcock & Keesing 1999). Allee effects such as these, where low population densities impair reproduction, can occur below MVP densities (Shepherd & Brown 1993). Data from many species of abalone suggest that populations decline when densities fall below 2,000/ha (Karpov et al. 2000, Tomascik & Holmes 2003, Rothaus et al. 2008). White abalone density estimates generated using ROV at the two sites thought to have the most white abalone in California, revealed densities two orders of magnitude below MVP: an offshore bank <25/ha (Stierhoff et al. 2012) and a remote Island <2/ha (Stierhoff et al. 2014). When populations are at these extremely low density, two mechanisms may act synergistically resulting in reproduction failure (1) large distances between nearest neighbors and (2) small aggregation sizes such that there is a low probability of both males and females being in the aggregation (Button 2008).

#### Captive Breeding Program

The dire state of the wild population with declining adult stocks and recruitment failure lends urgency to the captive breeding program for white abalone. The captive breeding program has had increased success in each of the past four spawning seasons with production increasing each year (Fig. 5). The program is working with small numbers of adults and relatively small numbers of gametes due to the endangered status of the species. Primary broodstock in the program is F1 generation, from spawnings in 2001 to 2003 (T. McCormick Channel Islands Marine Resource Institute), and only one of the original wild caught abalone remains in the breeding program today. A modification to the current white abalone permit has been submitted (April 2013) requesting permission

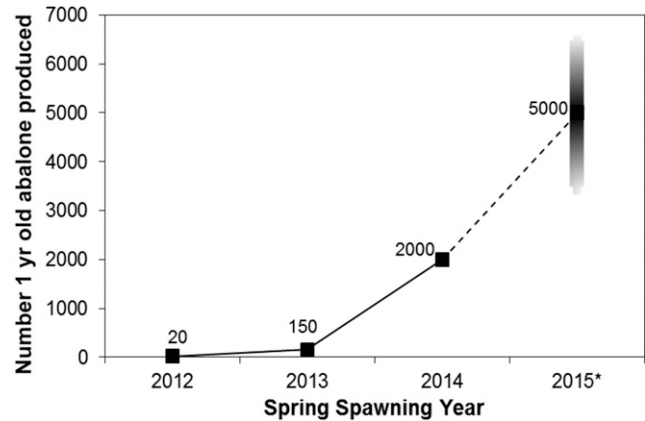


Figure 5. Graph of the number of juvenile white abalone cultured in the Captive Breeding Program since 2012. Figures for the 2015 cohort are estimates based on survival of previous cohorts in the program.

to collect additional wild animals to bolster the captive breeding program (request pending, October 2015). The addition of UV sterilization and temperature control (14°C) has helped the program move beyond disease issues, which plagued culturing efforts early on. Despite this, mortality events still occur in the broodstock, making disease and health monitoring an important feature of the captive breeding program. Population modeling of potential stocking scenarios highlights which options increase model population growth rates the most. With the success of both the captive breeding, disease prevention, and stocking models, the program can now start to move into the active ocean stocking phase of the work to help restore white abalone populations to the wild.

#### NEXT STEPS IN RESTORATION

The recovery program reached an important goal in 2015. The program has now produced enough small abalone to conduct stocking trials in the ocean to enhance the wild population. The next steps in the program will be to use a non-endangered congener, the red abalone (*Haliotis rufescens*) to conduct and optimize stocking strategies. A trial red abalone stocking experiment will be conducted to inform planning for a white abalone stocking. The stocking strategy includes (1) stocking in the winter when some predators are less abundant, (2) stocking inside and outside of artificial reefs to determine which promote survival, (3) predator relocation for sea stars and octopus, (4) tagging juveniles to quantify growth and survival, and (5) conducting both disease and genetic assessments of the cohort destined for stocking. Once small white abalone have been certified as healthy and genetic samples have been collected, they will be stocked using methods that promote abalone growth and survival. Sites will be examined to determine which have optimal temperature conditions promoting (1) health within withering syndrome (WS-RLP) endemic water, (2) development of gonad in stocked animals, and (3) survival of larvae produced by stocked animals.

The white abalone was the first marine invertebrate to be listed as an endangered species in the United States. Despite the challenges of restoring abalone, such as taking into account the importance of density and Allee effects as well as high mortality rates in the larval and newly settled life history stages, there is



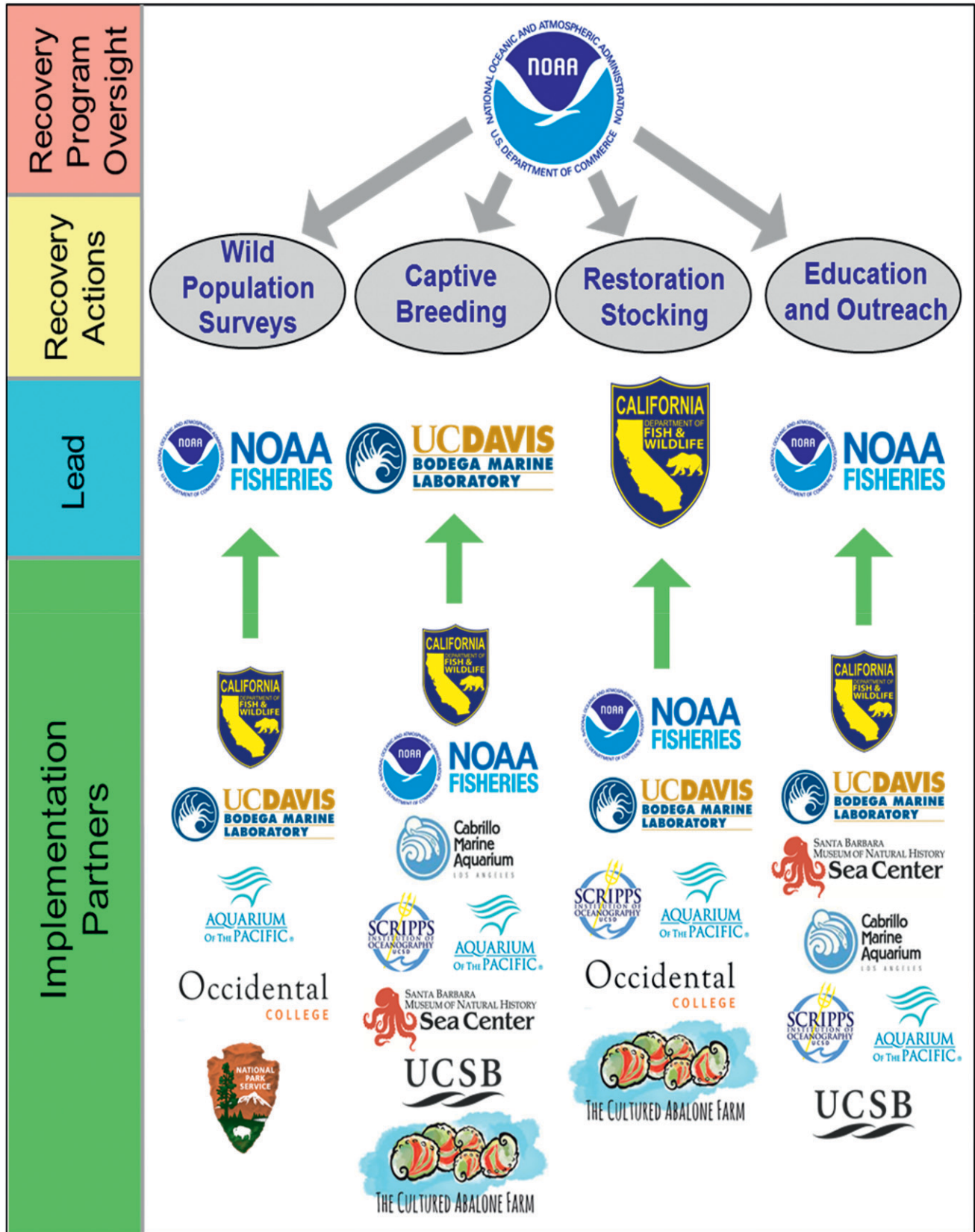


Figure 6. Diagram showing the White Abalone Recovery Consortium (WARC) partnership: National Oceanic and Atmospheric Administration (NOAA), NOAA Fisheries, Channel Islands National Park Service (CINPS), California Department of Fish and Wildlife (CDFW), University of California, Davis, Bodega Marine Laboratory (BML), UC San Diego Scripps Institution of Oceanography (SIO), UC Santa Barbara (UCSB), Aquarium of the Pacific (AoP), Cabrillo Marine Aquarium (CMA), Santa Barbara Natural History Museum Sea Center (SBNHM Sea Center), The Cultured Abalone, Occidental College.

hope. The wealth of knowledge within the abalone aquaculture industry suggests we have the technical expertise to generate enough individuals to stock into the wild. More work will need to be done to develop the science needed to enhance wild populations through successful stocking. The WARC partnership is poised to contribute to the restoration of this species both through culturing animals and tracking their success once stocked in the ocean. The WARC is working to attract funding to meet the program's needs, by drafting federal and state grant proposals as well as approaching nongovernmental groups for support. Education and outreach are an integral part of the program, as our public aquaria partners are able to show this endangered species to thousands of visitors each year and educate them about the problems and potential solutions faced in restoration of this species. Together we are working to save this iconic species that has been part of California's rich natural history and fishing heritage.

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