SAN FRANCISCO ESTUARY INVASIVE SPARTINA PROJECT

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Preserving native wetlands

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Website:
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Date: July 1, 2011

To: Jennifer Krebs, SFEP

From: Peggy Olofson

Subject: Report of Work Completed Under Estuary 2100 Grant #X7-00T04701

The State Coastal Conservancy received an Estuary 2100 Grant for $172,325 to use for control of non-native invasive *Spartina*. Conservancy distributed the funds through sub-grants to four Invasive *Spartina* Project (ISP) partners, including California Wildlife Foundation, San Mateo Mosquito Abatement District, Friends of Corte Madera Creek Watershed, and State Parks and Recreation. These four ISP partners collectively treated approximately 90 net acres of invasive *Spartina* for two consecutive years, furthering the baywide eradication of invasive *Spartina* restoring and protecting many hundreds of acres of tidal marsh (Figure 1, Table 1). In addition to treatment work, the grant funds also provided laboratory analysis of water samples collected from treatment sites where herbicide was applied, to confirm that water quality was not degraded by the treatments.

ISP Partners and contractors conducted treatment work in accordance with Site Specific Plans prepared by ISP (Grijalva et al. 2008; www.spartina.org/project_documents/2008-2010_site_plans_doc_list.htm), and reported in the 2008-2009 Treatment Report (Grijalva & Kerr, 2011; www.spartina.org/project_documents/2008-2009_treatment_rept_list.htm).

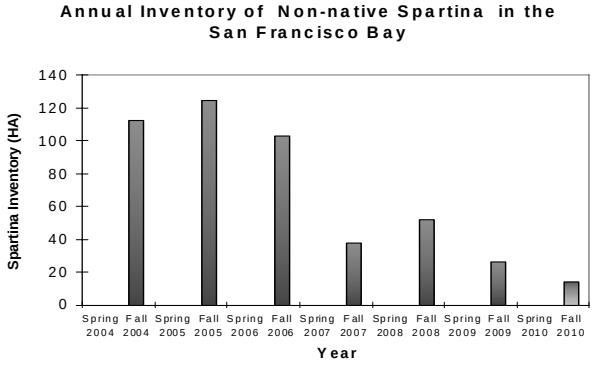
Water quality samples were collected and analyzed in accordance with the Aquatic Pesticide Application Plan and Addendum (Kerr 2009; www.spartina.org/project_documents/2009_APAP_Updated_dec09.pdf, www.spartina.org/project_documents/2009_APAP_Addendum.pdf).

The results of the water quality monitoring are presented in the Water Quality Report for 2007-2010 (Kerr 2011; www.spartina.org/project_documents/2007-10_WQMonRept.htm).

Inventory and efficacy monitoring were conducted in accordance with the Monitoring Program Quality Assurance Document ("QAD"; Hogle et al. 2008, updated 2009; www.spartina.org/project_documents/QAD_2009_Update_All(032410).pdf).

The efficacy of treatment and the change in *Spartina* coverage over time was reported in the 2008-2009 Monitoring Report (Hogle, 2011; www.spartina.org/project_documents/2008-09_MonReport.htm). The text, tables, and figures of the monitoring report are provided as Attachment 1 of this report. Pages 73-80 of the report provide excellent illustration of the successful restoration of pre-invasion marsh conditions following *Spartina* eradication.

Outcomes



Since its first full-scale control season in 2005 (pilot projects assessing various treatment approaches on selected sites were done in 2004), the ISP has reduced the net area of non-native Spartina by more than 90%, and is believed to be on on-track for successful eradication within the next decade. Most remaining sites are at levels well below 1% of their historic peak infestation levels, and correspondingly, most of the remaining infestations cover less than 1% of the total site area. It is anticipated that by 2013 up to 90% of the 170 sites currently infested with non-native Spartina will have achieved the first year of 'zero- detect' of non-native Spartina; on track for 3 years of 'zero-detect' constituting eradication on those sites.

The status of treated marshes following removal of the vegetative cover provided by non-native Spartina has been a large-scale return to a native-plant dominated condition at suitable elevations and a return to the native mudflat condition at lower elevations. An example of significant passive revegetation of treated areas can be seen at the Eden Landing marsh complex in Union City, where treatment on the central channel of Old Alameda Creek has been ongoing since 2005. Non-native Spartina has been nearly extirpated from the Creek, and as of 2011, the banks of the Creek that were formerly infested are dominated (in many cases with 100% cover) with native tidal marsh plant species like *Sarcocornia* spp, *Jaumea carnosa*, *Frankenia salina*, *Distichlis spicata*, and others. In contrast, at the Colma Creek complex in South San Francisco, the pre-invasion condition of the majority of the area was mudflat. Subsequent to control work there, the area has transitioned back to mudflat-dominated habitat. The interruption and reversal of the ongoing expansion of the non-native Spartina in these marshes has, in many cases, significantly altered the vegetative structure from a dense, invaded condition to a more open native condition. As the marshes are allowed to recover from the Spartina invasion, it is anticipated that historical vegetative complexity and density will be passively reestablished in most marshes.

The change in vegetative structure following treatment has had anticipated effects on several animal species in infested marshes, especially the California Clapper Rail. At most locations where clapper rail were present within the non-native Spartina, local populations decreased during the first 4-6 years of treatment, sometimes notably, likely due to removal of protective cover provided by the cordgrass. At many of these locations, especially those where treatment efforts were begun earlier, clapper rail populations have shown signs of stabilizing or slowly increasing by 2010 and 2011 (citation) as native marsh vegetative structure has re-established. A few sites showed significant clapper rail declines, as had been predicted at the outset of the control program, as they had both a high density and cover of non-native Spartina hybrids and high rail populations. Prior to the establishment of the non-native Spartina, these ar-

eas previously had no, or little, tall vegetative cover, and the clapper rail populations had appeared or increased rapidly in response to the new, introduced habitat. Where an established native marsh was subsequently invaded by non-native Spartina, clapper rail numbers show greater resilience to the removal of the vegetative structure provided by non-native Spartina. Although this has not been quantitatively studied, it is possibly due to underlying channel and hydrological complexity, native plant presence (even at a low level beneath the non-native Spartina canopy) and consequent availability to recolonize the treated site and a developed food web that could rebound after the removal of the non-native Spartina stressor .

Since the vast majority of the non-native Spartina historically present in the Estuary has been removed as of the 2010 Treatment Season, and most remaining stands are disparate, stunted and represent fractional percentages of the marshlands where they are found, almost all impacts associated with the removal of the vegetative cover provided by non-native Spartina have been realized. Treated marshes will continue on a trajectory of passive restoration of native marsh structure and composition. Over the medium to longer term, it is anticipated that rail populations will closely track pre-Spartina invasion levels as marshes revert to native conditions. Impacts associated with ISP activities going forward will predominantly be related to the potential to harass clapper rail as a result of monitoring, restoration or treatment activities, rather than habitat removal.

Achievement of Objectives

The Invasive *Spartina* Project initiated an aggressive *Spartina* treatment effort in 2005, and has successfully reduced the net area of invasive *Spartina* from greater than 800 acres, to less than 100 acres in six years. This is an exceptional accomplishment by any weed eradication standards, and it is made more significant by the number of environmental, physical, financial, and other challenges that have been overcome. The success has been at a cost of $1.5-2M per year, but it has helped to protect billions of dollars of past, present, and future tidal marsh restoration projects, and preserve the priceless balance of the San Francisco Estuary ecosystem. The treatment work conducted by the four grantees under the Estuary 2100 Grant funding played a significant role in helping to achieve the long term, bay wide objective of invasive Spartina eradication and preservation of native wetlands.

Challenges

Most weed eradication programs fail because they end too soon – either because of loss of funding or because of loss of institutional memory of the reason the effort was initiated to begin with. The easiest work is removing the large meadows of easily discernable plants. The hard work is keeping up rigorous monitoring over an extended period of time, until all plants have been found and eradicated and the seed bank exhausted. For the *Spartina* eradication, the "end game" challenge is further complicated by the presence of "cryptic" hybrids, which look similar to native foliosa but may contain genetic coding that will cause them to behave invasively given the right environmental conditions. If long term eradication of invasive *Spartina* is to be successful, all stakeholders in the tidal marsh ecosystem restoration of San Francisco Estuary must commit to staying informed and alert, potentially for decades to come, to assure the hybrid *Spartina* does not reemerge as a problem in the marsh.

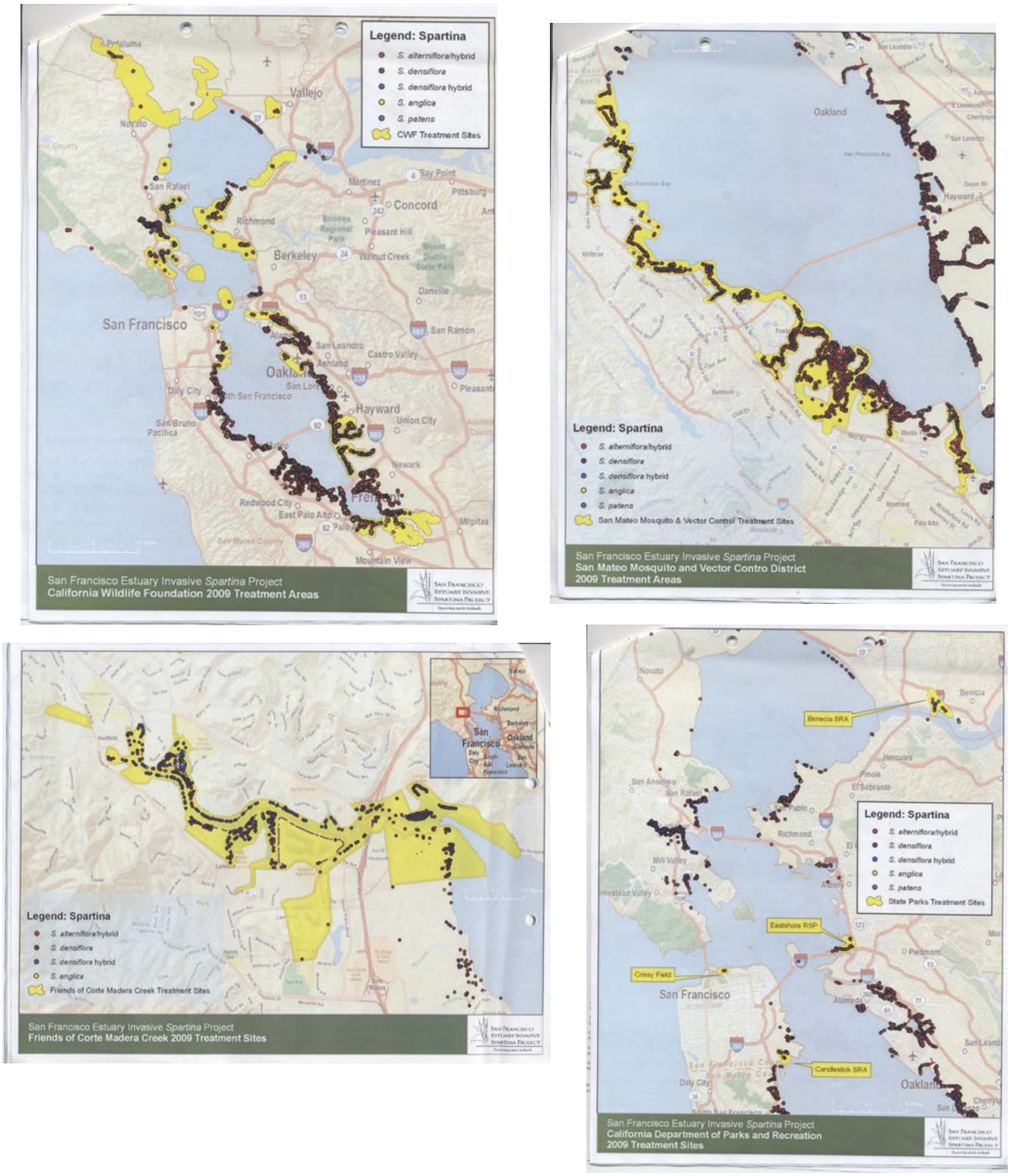


Figure 1. Location of Marshes Treated by Estuary 2100 Grantees to Eradicate Invasive Spartina. From top left: California Wildlife Foundation, San Mateo Mosquito and Vector Control District, Friends of Corte Madera Creek Watershed, California Parks and Recreation

Table 1. List of Sites Treated by Estuary 2100 Grantees to Eradicate Invasive Spartina.
Sub-Area numbers correlate with site numbers used in ISP treatment and monitoring reports. (www.spartina.org/project.htm).

Grantee	*ISP Sub-Area #*	Name of Sub-Area/Marsh
CWF	03a	Blackie's Creek (above bridge)
CWF	03b	Blackie's Creek Mouth
CWF	05g	Cargill Pond (W Suites Hotel)
CWF	9	Pickleweed Park
CWF	12b	Pier 98/Heron's Head
CWF	12c	India Basin
CWF	12d	Hunters Point Naval Reserve
CWF	12h	Yerba Buena Island
CWF	12i	Mission Creek
CWF	13d	Whale's Tail North Fluke
CWF	13e	Whale's Tail South Fluke
CWF	13f	Cargill Mitigation Marsh
CWF	13i	Eden Landing-Pond 10
CWF	13j	Eden Landing-Mt Eden Creek
CWF	13k	Eden Landing Reserve South - North Creek Marsh
CWF	13l	Eden Landing Reserve North- Eden Creek Marsh
CWF	15a	South Bay Marshes - Santa Clara County
CWF	15b	Faber/Laumeister Marsh
CWF	15c	Shoreline Regional Park at Mountain View
CWF	16	Cooley Landing (Ravenswood Open Space Preserve)
CWF	17f	Oakland Inner Harbor
CWF	17g	Coast Guard Island
CWF	17j	Fan Marsh
CWF	20b	Oakland Metropolitan Golf Links (formerly Lew Galbraith)
CWF	20r	Oakland Airport Shoreline and Channels
CWF	22a	Wildcat Marsh (Chevron Marsh)
CWF	22b	San Pablo Marsh
CWF	22c	Rheem Creek Area
CWF	22d	Stege Marsh
CWF	22e	Hoffman Marsh
CWF	22f	Richmond/Albany Shoreline
CWF	23a	Brickyard Cove
CWF	23b	Beach Drive
CWF	23c	Loch Lomond Marina
CWF	23d	San Rafael Canal Mouth North
CWF	23e	Muzzi & Martas Marsh
CWF	23f	Paradise Cay
CWF	23g	Greenwood Beach Road/Harbor
CWF	23h	Strawberry Point
CWF	23i	Strawberry Cove
CWF	23j	Bothin Marsh
CWF	23k	Sausalito
CWF	23l	Starkweather Park
CWF	23m	Novato
CWF	23n	Triangle Marsh
CWF	24a	Upper Petaluma River- Upstream of Grey's Field
CWF	24b	Grey's Field
CWF	24c	Petaluma Marsh
CWF	24d	Lower Petaluma River-Downstream of San Antonio Cr
CWF	25a	Tom's Point, Tomales
CWF	26a	White Slough/Napa River
CWF	26c	Sonoma Creek
CWF	26d	Sonoma Baylands
FCMCW	04a	Corte Madera Ecological Reserve

Grantee	*ISP Sub-Area #*	**Name of Sub-Area/Marsh**
FCMCW	04b	College of Marin Ecological Study Area
FCMCW	04c	Piper Park East
FCMCW	04d	Piper Park West
FCMCW	04e	Larkspur Ferry Landing Area
FCMCW	04f	Riviera Circle (Larkspur Marina)
FCMCW	04g	Creekside Park
FCMCW	04h	Upper Corte Madera Creek (Above Bon Air Rd)
FCMCW	04i	Lower Corte Madera Creek (Bon Air Rd to HWY 101)
FCMCW	04j	Corte Madera Creek Mouth (Below HWY 101)
FCMCW	04k	Boardwalk No. 1 (Arkites)
FCMCW	04l	Murphy Creek
SMCMAD	18a	Colma Creek
SMCMAD	18b	Navigable Slough
SMCMAD	18c	"Old Marina"
SMCMAD	18d	"Inner Harbor"
SMCMAD	18e	Sam Trans Peninsula
SMCMAD	18f	"Confluence Marsh"
SMCMAD	18g	San Bruno Marsh
SMCMAD	18h	San Bruno Creek
SMCMAD	19a	Brisbane Lagoon
SMCMAD	19b	Sierra Point
SMCMAD	19c	Oyster Cove
SMCMAD	19d	Oyster Point Marina
SMCMAD	19e	Oyster Point Park
SMCMAD	19f	Point San Bruno
SMCMAD	19g	Seaplane Harbor
SMCMAD	19h	SFO
SMCMAD	19i	Mills Creek Mouth
SMCMAD	19j	Easton Creek Mouth
SMCMAD	19k	Sanchez Marsh
SMCMAD	19l	Burlingame Lagoon
SMCMAD	19m	Fisherman's Park
SMCMAD	19n	Coyote Point Marina/Marsh
SMCMAD	19o	San Mateo Creek /Ryder Park
SMCMAD	19p	Seal Slough Mouth
SMCMAD	19q	Foster City
SMCMAD	19r	Anza Lagoon
State Parks	06a	Emeryville Crescent East
State Parks	11	Southampton Marsh
State Parks	12e	Yosemite Channel
State Parks	12f	Candlestick Cove

San Francisco Estuary
Invasive Spartina Project
2008 - 2009
Monitoring Report

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Part I

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The UC Davis Spartina Lab (Dr. Don Strong, Dr. Debra Ayres, Dr. Christina Sloop, and PhD candidate Laura Feinstein) and independent consultant Dr. Emma Jack informed our understanding of the issues surrounding the genetic analysis of *Spartina*, as reported in this document.

ISP Project Director Peggy Olofson and Control Program Managers Erik Grijalva and Drew Kerr assisted greatly with the review of this monitoring data and with the expanded collaboration between the Monitoring and Control Programs.

The hard work and dedication of all involved has enabled the ISP to succeed in coordinating a large and complex regional monitoring and eradication effort.

Part II

Assistant Monitoring Program Manager Tripp McCandlish manages the data collection and processing of photos for the project's permanent plot photo monitoring.

OEI Biologist Jeff Lewis performed the statistical analysis and write-up of treatment efficacy monitoring data in collaboration with Ingrid Hogle.

Cover Photos: Views of Oyster Point Park in 2007 and in 2009 after invasive Spartina eradication. *Spartina foliosa* marsh in the San Pablo Baylands in background photo.

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California Wildlife Conservation Board (MOU #99-054-01 and subsequent)

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INTRODUCTION

The San Francisco Estuary Invasive *Spartina* Project was established by the California Coastal Conservancy in 2000 in response to the invasion of hybridized non-native *Spartina* into the marshes and mudflats of the San Francisco Estuary (referred to as Estuary or Bay throughout this report).

In the last several decades, four non-native cordgrasses, including *Spartina alterniflora* (Atlantic cordgrass), *S. densiflora* (Chilean cordgrass), *S. anglica* (English cordgrass), and *S. patens* (saltmeadow cordgrass), were introduced to the Estuary. Each of these species is known to be an aggressive invader outside of its native range, and each has demonstrated varying degrees of invasiveness since establishing in the Estuary. The Army Corps of Engineers introduced *S. alterniflora* in Pond 3A near the Alameda Flood Control Channel in the early 1970s with the intention of restoring marsh vegetation. The introduced cordgrass established successfully at this site and was subsequently transplanted into other restoration sites around the Bay. *Spartina densiflora* and *S. anglica* were introduced at Creekside Park in Corte Madera, where they were intentionally planted in a park design. The history of the introduction of *Spartina patens* to the Estuary is unknown. To date it has been found at only one site – Benicia State Recreation Area's Southampton Marsh.

Both *S. alterniflora* and *S. densiflora* hybridized with native *S. foliosa* (Daehler and Strong 1996, Ayres et al. 2003, Ayres et al. 2008a). Offspring of *S. alterniflora x foliosa* hybrids backcrossed with the parent species and with one another, producing an extremely robust and fertile "hybrid swarm," which has invaded habitat throughout the Estuary, threatening the ecological integrity of the Estuary's existing and potential future restored tidal wetlands and mudflats (Daehler and Strong 1996, Goals Project 1999, Ayres et al. 2003, Conservancy 2003, Ayres et al. 2004b, Ayres et al. 2008a).

The purpose of the ISP is to implement a coordinated, region-wide program to control and eventually eradicate *S. alterniflora* and their hybrids as well as other non-native *Spartina* species from the Estuary.

As part of its regional program, the ISP conducts annual monitoring to track and map the extent and rate of spread of nonnative *Spartina*, to inform the ISP's Control Program, and to monitor the efficacy of treatment efforts. Part I of this report presents the results of region wide inventory monitoring conducted by the ISP in 2008 and 2009, with Baywide monitoring results presented in **Tables 1** and **2**.

Since its inception, the ISP has collaborated with researchers at the UC Davis Spartina Lab (Dr. Don Strong, Dr. Debra Ayres and colleagues). The Spartina Lab conducts research regarding the hybridization of introduced *Spartina* species with the native *S. foliosa* and developed genetic markers for such work, including RAPD and microsatellite markers. Until 2008, the State Coastal Conservancy contracted with the UC Davis Spartina Lab to analyze *Spartina* samples for species identification using Random Amplified Polymorphic DNA (RAPD) markers. In 2009 the ISP contracted with a commercial lab to perform genetic testing. Information regarding genetic testing results are integrated within Part I of this report.

Treatment efforts began with small-scale manual removal in 2002 and 2003. In 2004, the herbicide glyphosate was applied at most treated sites, and testing of aerial applications of the herbicide imazapyr began at a few trial locations. The ISP Control Program has coordinated annual region-wide *Spartina* control efforts using the highly effective herbicide imazapyr in aerial and ground-based applications from 2005 to present, with full-scale treatment beginning in 2006. Treatment methods are generally described in the ISP's Programmatic EIS/R (Conservancy 2003). Specific treatment approaches are described in site-specific control plan prepared for each site (ISP 2004, 2005, Grijalva et al. 2008, 2011). *Spartina* treatment operations are reported annually by the ISP Control Program (Grijalva 2004, Grijalva and Kerr 2006, Grijalva et al. 2008).

Part II presents the results of the photo point and permanent plot monitoring data collected by the ISP through 2008 to assess the efficacy of control efforts.

PART I: INVENTORY MONITORING

BACKGROUND

Inventory Monitoring

The ISP began Estuary-wide inventory monitoring in 2000, with annual monitoring of all sites beginning in 2004. The original geographic scope of monitoring efforts was limited to the bayward side of most major highways due to staff constraints (see QAD). Since 2006 all potential invasive *Spartina* habitat identified within the San Francisco Estuary, Bolinas, Point Reyes and Tomales Bay has been surveyed by the ISP or its partners. This includes annual surveys of over 50,000 acres of tidal marsh and mudflat throughout the Estuary and Outer Coast areas.

Inventory monitoring is conducted for two purposes: to track change in the extent and net cover of the infestation over time for purposes of analysis and reporting, and to locate and map patches of invasive *Spartina* to inform management and coordination of field operations by the ISP Control Program.

Genetic Testing

From 2000 to 2008, the ISP has contracted with the lab of Dr. Don Strong at the University of California, Davis (also referred to as the UC Davis Spartina Lab) to conduct RAPD-based genetic testing to determine hybridity of collected *Spartina* samples. The RAPD markers used by the ISP for identification of *S. foliosa*, *S. alterniflora S. densiflora*, *S. anglica* and their hybrids were developed within Dr. Strong's lab (Daehler and Strong 1997, Ayres et al. 1999, Ayres and Strong 2001, Ayres et al. 2008a). The ISP relies on these RAPD tests to confirm taxonomic field identification and to test for hybridity of those plants that are difficult to identify based on field characteristics.

In 2009, the ISP coordinated with researchers from the UC Davis Spartina Lab to select microsatellite markers, also referred to as simple sequence repeat (SSR) markers, for genetic testing to help determine the species composition of individual *Spartina* samples. With support from Dr. Sloop, ten SSR markers were chosen by L. Feinstein from the suite of available SSR markers previously developed by UC Davis researchers (Blum et al. 2004, Sloop et al. 2006). These markers were selected based on their power to distinguish native from hybrid plants, based on SSR screens of *S. foliosa* and *S. alterniflora x foliosa* hybrids performed by Drs. Sloop and Blum during initial marker development. Markers were additionally validated by subsequent genetic analysis of San Francisco Bay hybrid *Spartina* (Sloop et al. 2009, 2011), and by unpublished genetic screenings of additional *S. foliosa* sampled from Baja, Mexico, and from Northern California populations by Dr. Bando in 2007.

Samples collected in 2009 were sent to a commercial lab in Colorado (STA Labs) for DNA extraction and subsequent testing of microsatellite and RAPD markers. STA Labs was unsuccessful in their attempts at RAPD testing, so the ISP contracted with Dr. Ayres to perform RAPD testing on a subset of samples. The ISP contracted with L. Feinstein at UC Davis to perform duplicate microsatellite testing on a subset of samples to allow comparison of consistency of microsatellite results between the two labs (STA and UC Davis) (Feinstein and Hogle in preparation).

The ISP analyzed the microsatellite data with the assistance of plant geneticist Dr. Emma Jack, with whom the ISP contracted to consult on methodologies and data analysis. Dr. Jack worked with the ISP to perform the analysis of microsatellite results from STA Labs and to interpret these results.

Treatment Surveys

The ISP initiated a pilot project in 2009 to conduct monitoring of treatment activities, termed "treatment surveys". The purpose of these surveys was to map treated patches of *Spartina* so as to help inform future years' monitoring efforts. Because regrowth of treated *Spartina* is often stunted and can be difficult to identify or distinguish from native *Spartina*, knowledge of patch-level treatment activities provides additional evidence which is helpful for the identification of *Spartina* within a patch. Treatment surveys had the added benefit of allowing monitoring staff to assist treatment crews in the identification of invasive *Spartina* and relocation of previously mapped patches. There were concerns regarding the potential for monitoring staff presence to slow down or impede the work of the treatment crews, but generally this did not occur.

INVENTORY METHODS

ISP field biologists conducted inventory monitoring between May and December in 2004, 2005, 2006, 2007, 2008 and 2009, with the majority of monitoring completed between mid-June and mid-October. Mapping-grade global positioning system (GPS) units (Trimble GeoXT 2003 model) were used to collect point, line and polygon data containing *Spartina* species and percent cover data using ArcPad software. Field sites were accessed using the least destructive, most efficient, thorough, and cost-effective methods possible for each site. Access methods included walking, boating, kayaking and helicopter. Binoculars were used to help identify plants at a distance. GPS features were offset when necessary using a laser rangefinder and compass to determine distance and direction from observer. Details of inventory monitoring methods are described in the ISP Quality Assurance Document (QAD) (Hogle et al. 2008).

Species Identification

Species mapped included *S. alterniflora x foliosa* hybrids ("hybrids"), *S. densiflora*, *S. densiflora* x *foliosa*, *S. anglica*, and *S. patens*. Although some pure *S. alterniflora* plants may still exist in the Estuary, the project assumed that this is unlikely due to the documented pollen swamping and superior invasiveness of the *S. alterniflora x foliosa* hybrids compared to the *S. alterniflora* parents (Anttila et al. 1998, Ayres et al. 2004a, Ayres et al. 2004b). Project biologists did not attempt to distinguish between pure *S. alterniflora* and *S. alterniflora x foliosa* hybrids, but lumped these together for the purposes of monitoring and treatment, referring to them as *S. alterniflora x foliosa*, *S. alterniflora*/hybrids or simply "hybrids".

Species were identified based on a number of considerations including morphology, location, phenology, and/or past years' lab results. Field staff used drop-down menus in their GPS data forms to record their species identification (based on any of the above factors) as well as their level of confidence associated with that identification for each feature. Confidence level choices included: lower, moderate or high confidence. Ambiguous plants were either identified to species and given a lower field-identification confidence, or identified as "unknown alterniflora or foliosa", "unknown densiflora/hybrid" or "unknown anglica/alterniflora hybrid".

Samples were collected for genetic testing, to compare field-identification with lab-identification. Where logistically possible, samples were collected for genetic testing of ambiguous individuals. In 2008 and 2009, 382 and 359 ambiguous samples were collected, respectively.

Samples of plants identified by field staff with moderate to high confidence were also sampled for genetic testing throughout each season. Plants field-identified as *S. alterniflora x foliosa* hybrids with moderate to high confidence were sampled in 2008 (250 samples) and 2009 (245 samples). Plants field-identified as *S. foliosa* with moderate to high confidence were sampled in 2008 (375 samples) and 2009 (470 samples). In 2008, these samples were collected with the intention of testing the field identification skills of the biologist, using genetic sampling to confirm species identification. Such samples were taken at sites with a mixture of native and hybrid *Spartina*, as well as at sites believed to contain only the native *S. foliosa*. In 2009, these samples were primarily collected with the same intention, but a few samples of high confidence field identification were collected to be used as controls for the genetic testing.

Treatment Area

Beginning in 2008 the ISP began recording a "treatment cover" value for each GPS feature recorded (see *Reporting *Spartina* Area* box). Capturing this value allowed calculation of an estimate of the area requiring treatment, thereby allowing the ISP Control Program to more accurately plan for treatment activities. Cover classes were used for estimation of the percent of each patch requiring treatment, so the resulting sum in "treatment area" for a site has a minimum, mean, and maximum value based on the minimum, mean, and maximum of the cover class category.

Treatment Surveys

Beginning in 2009, the ISP monitoring staff began conducting treatment surveys at a small subset of sites (approximately 12 sites). Monitoring data was "checked out" onto GPS units using ArcPad software, and customized forms were used to record treatment survey information, primarily whether and how a patch was treated. Treatment methods recorded included sprayed (with imazapyr herbicide), dug, tarped, or other.

During treatment surveys, monitoring staff kept up with, guided, or lagged behind treatment crews. When lagging behind, staff were able to record what was or was not treated based on observance of the blue dye (Blazon Blue) added to the herbicide used at sites treated with herbicide. This method was necessary when there were fewer monitoring staff than treatment crews, or when treatment crews were moving more quickly than data could be recorded. At sites treated with manual control methods, treatment was generally slower and monitoring staff could more easily keep up with treatment crews.

Mapping Methods

Past monitoring methods have included field-based monitoring using GPS as well as digitizing of aerial imagery in a GIS. Field-based monitoring is conducted by using GPS units with customized software to map point, line and polygon features and associated attributes to document the location, extent and density of individual patches of invasive *Spartina*. All three feature types are used during surveys, so that the resulting GIS data includes point, line and polygon features which must be all be viewed together for a complete representation of field-mapped *Spartina*. The minimum size for a feature is a 5 cm-radius point; there is no maximum size for a feature.

Interpretation of aerial imagery followed the "heads-up" digitizing methods described in the ISP Quality Assurance Document (Hogle et al. 2008) While the invasive *Spartina* population was largely untreated in 2004 and 2005, interpretation was relatively simple and accurate, as assessed by ground truthing of digitized areas. From 2004 through 2007, many large and/or inaccessible sites were inventoried via heads-up digitizing of custom-flown 30 cm resolution, orthorectified color infrared aerial photography. As control progressed, interpretation of aerial imagery became less effective as a way to accurately map remaining patches of invasive *Spartina*. Digitizers found identification of *Spartina* patches in the aerial imagery difficult in 2006, and ground truthing efforts in 2006 indicated some inaccuracies in aerial image interpretation. In 2007, even the acquisition of higher (up to 16 cm resolution) digital color IR aerial imagery was not effective in allowing accurate identification of remaining patches of invasive *Spartina* within areas where treatment had successfully reduced *Spartina* cover. Ground truthing efforts in 2007 indicated presence of live, non-native *Spartina* regrowth in many areas where aerial imagery interpretation indicated absence of any remaining living *Spartina*. The project returned to exclusively field-based inventory monitoring in 2008.

*** Reporting *Spartina* Area***

Two methods are used to measure and report area of non-native *Spartina*, "**net area**" and "**treatment area**". "Net area" refers to the actual amount of the *Spartina*, and is calculated to represent the coverage as if all non-native *Spartina* plants were contiguous (i.e., compacted onto one discrete area). Net area is not very useful for planning and management purposes, as it does not give an accurate picture of the marsh area that will need to be treated or monitored. For this purpose, "treatment area" is used, which is defined as the area requiring treatment. Gross area is the area of all GIS features recorded, and is calculated using GIS and a point/line-buffering strategy. Cover class categories are used to define the net and treatment area of *Spartina* within gross areas. Treatment area is a new concept developed by the ISP in 2008, and has proved extremely useful for planning and management purposes.

We replaced the use of aerial image interpretation with the use of helicopters or "inventory grids" to map *Spartina* in 2008. In 2008, after an extremely successful trial, we contracted with a company specializing in helicopter monitoring to provide a small, low-flying helicopter, pilot and support staff to allow inventory and mapping of *Spartina* in large, remote sites where inventory by foot or boat is either impractical or would have a significant destructive impact on the marsh. The helicopter flew high enough to minimize rotor wash effects on the vegetation, and low enough to allow accurate identification of *Spartina* plants. As the passenger, the ISP biologist was able to record a GPS feature while the pilot hovered or flew slowly over each patch. The helicopter was able to briefly land in the marsh as needed for the biologist to collect plant samples. Both field staff and the pilot were trained to be acutely aware of bird responses, and the pilot flew away from any birds that appeared to be disturbed by the helicopter. If possible and necessary, the pilot returned to the inventory location once the birds had flown away. (This strict avoidance of birds is important both to minimize impact to wildlife and to avoid interference with the helicopter.) Helicopter monitoring was expanded in 2009, leading to more efficient monitoring of several sites.

In areas which could be accessed by foot or by boat, but where *Spartina* distribution was relatively even (i.e. not clustered into discernable point, line or polygon GPS features), we created site-specific "inventory grids" in a GIS to facilitate field mapping of *Spartina* cover. Grid sizes were based on ease of access and level of precision required by the Control Program, and included 10x10 meter grids (for mapping *Spartina densiflora* at Creekside Park), 25x25 meter grids (at smaller sites), 50x50 meter grids (at larger sites), and 100x100 meter grids (at one site, Cooley Landing, where access was very difficult). Field staff used GPS to navigate to the center of each grid cell, and then recorded *Spartina* cover information for the grid cell.

When used throughout this report, the term "feature" may refer to a GPS-collected point, line or polygon feature, a digitized polygon, or a polygon "grid" feature, as described above.

GPS Field Data

In response to the increased difficulty in identifying hybrid *Spartina* in successfully treated areas, we took steps to be able to bring past years' data into the field as GPS layer files. This allowed us to identify and navigate to past years' patches to look for and help identify any regrowth of invasive *Spartina* in these areas. This did not replace, but rather augmented our full inventory of all new and old patches of invasive *Spartina*.

To facilitate the visualization of past years' data in the field, we transitioned to use of ArcPad (ESRI trademark) software on our GPS units. In past years, monitoring staff used the software TerraSync (Trimble trademark) to map and record information about invasive *Spartina* in the field. The TerraSync software allowed use of data dictionaries (electronic forms filled in with information for each recorded feature), but it did not facilitate "copy out" of past years' data by site and with associated attributes and symbols onto GPS units. With ArcPad, we were able to efficiently "copy out" all relevant GIS data for a site to the GPS file to be used in the field. Thus staff were able to see and navigate to the exact locations where invasive *Spartina* had been mapped in the past and query the information associated with these locations while in the field. ArcPad also allowed us to check out other data from our GIS to bring into the field as well, such as inventory boundary layers indicating where to look for *Spartina*. This GPS data augmented paper maps with the same data which were also taken out into the field as navigational tools for every survey. Improvements to the ArcPad data entry forms were implemented for the 2009 season and assisted with improving the efficiency of data collection.

Data Processing and Editing

Inventory data was checked in from ArcPad to an ArcGIS geodatabase, then checked for accuracy and edited for location and attribute accuracy in a GIS. All GPS features were checked for positional and attribute accuracy in ArcMap by the same individuals who collected the data.

Genetic results from RAPD DNA tests were linked to the point layer indicating where genetic samples were collected then overlaid onto the inventory data. All lab-based species identifications were recorded in a "lab identification" column. Final species determinations for features associated with specific DNA samples were based on a review of lab identification results and a cross-check of field photos and attributes recorded

by the biologist who collected the data. Through this method, biologists have the opportunity to review the lab data for potential false positives, and the lab is contacted to review specific results when such concerns arise.

In 2008, as in prior years, RAPD results with high lab ID confidence were generally considered more reliable than field-identification, and were rarely refuted. (Confidence of lab ID is explained below.) In 2009, genetic results were viewed with greater caution due to the greater lack of correspondence between field identification and genetic evidence at many sites.

Collaborator Data

All data were collected by ISP field biologists with the exception of the 2008 dataset for the South Bay which was submitted by Santa Clara Valley Water District (SCVWD). The water district has mapped and treated invasive *Spartina* in tidally-influenced marshes within their jurisdiction since 2003. They have contributed their GPS location data and associated attribute information (patch size and treatment notes) annually since 2003. Although 2007 was their final year mapping a full inventory of identified and treated *Spartina*, SCVWD submitted partially-mapped data in 2008. ISP field biologists conducted independent inventory monitoring of all South Bay areas in 2008. Data from SCVWD and ISP were combined and edited in ArcGIS to remove redundant features.

INVENTORY ANALYSIS METHODS

Summary statistics of *Spartina* inventory monitoring data were calculated by converting all data to polygon data. This was done by buffering lines by width and points by diameter values recorded in the field at time of data collection.

For regional analysis, models were created using ArcGIS Model Builder to clip these summary polygons by regional boundaries, calculate minimum, mean and maximum net and treatment area per clipped feature, then sum net area and treatment area (see Box in Section 1.1 above) by species within each region. Minimum, average (mean) and maximum area values were calculated based on the cover class ranges used to record net and treatment cover during inventory monitoring (<1%, 1-5%, 5-9%, 10-19%, 20-29%...90-99%, 100%).

Designation of mapped locations as new populations was determined in ArcGIS by selecting those patches of invasive *Spartina* mapped in 2008 or 2009 that were greater 500 m – 1 km from any invasive *Spartina* mapped in past years. Other "Sites of Concern" included those with confusing morphologies and/or lab results.

Results are reported in acres and/or square meters. This combination of standard and metric units is an artifact of the units of measurement used by the ISP Control Program and past monitoring reports (acres) and the units of measurement used during patch-level monitoring efforts (square meters). Summary data are presented in standard units for large areas and square meters for small areas so as to maintain consistency in presentation from past years and for the ease of those who collect and use this data on a regular basis.

GENETIC METHODS

Genetic samples were collected by ISP field biologists in the field to confirm field identification of some plants, and to test for hybridity of other plants. Samples were collected on an as-needed basis, and were primarily collected to test the hybridity of plants which were difficult to identify as native or hybrid in the field. Samples were collected from around the project area, as shown in **Figures 1.4** through **1.7.**

2008 Genetic Testing Methods

A total of 1,063 samples were collected and tested in 2008. Of these, twenty-six were specifically tested for *S. anglica* hybridity, thirty-three were tested for *S. densiflora* hybridity, and the remainder were solely tested for hybridity between *S. foliosa* and *S. alterniflora*.

Genetic testing was performed by the UC Davis Spartina Lab using their RAPD method protocol, as documented in detail for the ISP by L. Feinstein (2009) and as briefly described in other publications (Daehler and Strong 1997, Ayres et al. 1999, Daehler et al. 1999, Ayres and Strong 2001).

Primers and species-specific fragments ("markers") amplified by each primer were selected from those developed by the UC Davis Spartina Lab and effectively included four alterniflora-specific markers, seven anglica-specific markers, five densiflora-specific markers and four foliosa-specific markers (**Table 3 RAPD markers used**). Presence of marker B7 800 is indicative of either of foliosa or densiflora (Ayres, unpublished data), and for the preceding list is counted in both the foliosa and densiflora marker counts. Markers B7 550 and B7 650 are described as a single marker (B7 550/650), as presence of either or both markers is described as indicative of *S. alterniflora* (Ayres, unpublished data).

Testing of 2008 samples was performed by lab technicians under the direction of Dr. Debra Ayres of the UC Davis Spartina Lab. PCR plates did not contain controls, but were scored based on expected banding positions relative to ubiquitous, unscored bands. Gels were visually scored for presence or absence of species-specific markers and results were input into a Microsoft Excel spreadsheet by technicians. Data were checked by Dr. Ayres and reported to the ISP electronically via a standardized Excel spreadsheet containing cumulative results to date, including results from reruns of samples for which PCR failed to amplify during the initial run. When samples failed to amplify, up to six replicate PCR trials were attempted. Photographs of each gel are stored in a laboratory notebook at the UC Davis lab, and are referenced to check scoring when questions arise regarding RAPD results.

2009 Genetic Testing Methods

A total of 1132 samples were collected and processed in 2009. Leaf samples were collected in the field following the collection protocol specified in the contract with UC Davis (see QAD Appendix 5) and were shipped to STA Labs in Colorado, where DNA was extracted for genetic testing. Samples were kept refrigerated prior to shipment and were shipped overnight with blue ice in small coolers on a weekly basis beginning in July 2009.

STA Labs conducted microsatellite testing for all samples received. STA Labs used their own (proprietary) DNA extraction methods, and used the microsatellite primer sequences selected for the ISP by Christina Sloop and Laura Feinstein (Feinstein 2009). A list of these markers is presented in **Table 4 2009 SSR Primers-SE+ih.doc**.

Samples from four *Spartina* patches of presumed known species, based on field-identification, were used as internal controls in the testing by STA labs. These samples included one *S. alterniflora*/hybrid from Blackie's Pasture in Marin County, one *S. alterniflora* sample from South Freeport, Maine, one presumed *S. foliosa* American Canyon, and one S. densiflora from Burlingame Lagoon. (Note: the *S. alterniflora*/hybrid from Blackie's Pasture was located in a previously unsurveyed and untreated side channel.) As internal controls, samples of DNA taken from each of these four individuals were run on each of the 11 plates analyzed by STA Labs.

STA Labs was unable to successfully perform RAPD testing on any samples, and the ISP contracted with Dr. Debra Ayres to conduct RAPD testing on a subset of the 2009 genetic samples. In January 2010, STA Labs shipped ten sealed, 96-well trays of frozen, extracted DNA with an inventory chart of the samples in each of the wells to UC Davis. The wells contained DNA remaining following STA's microsatellite testing. Dr. Ayres purified then performed RAPD testing on over 300 of these samples to allow comparison of RAPD results from UC Davis and microsatellite results from STA Labs. Two primers, X9 and X18, which had been used and tested within the UC Davis lab but had not been used for ISP sample testing in past years were added to the 2009 RAPD analysis and one primer previously used (A17) was excluded from the RAPD analysis in 2009 (**Table 3 RAPD markers used.xls**). As described in the report by Dr. Ayres (2010), gels were scored within an excel spreadsheet using a photo-gauge made from the images of species standards. However, controls were not run on the gels and no molecular ladder was run alongside the samples to check the accuracy of the banding size. Gels were scored at least twice, and the accuracy of data entry was verified each time. Duplicate reactions were performed for about 13% of the sample x primer

combinations. The accuracy of band evaluation was compared between the original and duplicate sample x primer combinations.

GENETIC DATA ANALYSIS METHODS

RAPD Results Analysis

Genetic results from RAPDs were analyzed by the ISP using binary matrix formulas created by the ISP and the UC Davis Spartina Lab to calculate species identification and identification confidence level for each sample. The UC Davis Spartina Lab reported results via standardized Excel spreadsheets, using a binary matrix (columns to indicate presence or absence) of species-specific markers. These data were analyzed using algorithms in Excel which were developed by UC Davis, expanded by the ISP, and reviewed for accuracy by an independent consultant. These algorithms use the presence or absence of species-specific RAPD markers to determine if a sample contains markers indicative of S. densiflora, *S. foliosa*, *S. alterniflora* or their hybrids.

Degree of hybridity is calculated by a "hybridity index" formula which calculates the percentage of RAPD marker results indicative of hybridity between *S. foliosa* and *S. alterniflora* (Ayres et al. 1999, Ayres et al. 2004a, Ayres et al. 2008b). The presence of *S. alterniflora*-specific markers and the absence of *S. foliosa*-specific markers weigh equally in their contribution to this hybridity index score. Both presence and absence of species-specific markers were classified as informative because markers were chosen to be both unique and ubiquitous within parent species (Ayres, personal communication).

Lab identification confidence was calculated by an additional formula which was developed by the ISP in collaboration with Dr. Debra Ayres. This formula applies Dr. Ayre's expertise to the analysis of lab identification confidence, and is based on number of markers amplified, number of *S. alterniflora*-specific markers present, and number of *S. foliosa* markers absent. This formula is based on the logic that confidence in the evidence of hybridity increases as presence of *S. alterniflora*-specific markers and absence of *S. foliosa*-specific markers increase. All of these formulas are based on analysis of up to four *S. foliosa*-specific markers and up to four *S. alterniflora*-specific markers.

Very low confidence evidence of hybridity is defined as the absence of a single *S. foliosa*-specific marker, and the absence of any *S. alterniflora*-specific markers among reactions that successfully amplified.

Low confidence evidence of hybridity is defined as the (a) absence of at least two *S. foliosa*-specific markers, (b) absence of one *S. foliosa*-specific marker and absence of at least one *S. alterniflora*-specific marker, or (c) absence of one *S. foliosa*-specific marker, presence of at least one *S. foliosa*-specific marker, and absence of all *S. alterniflora*-specific markers among reactions that successfully amplified.

Medium confidence evidence of hybridity is defined as the (a) presence of only one *S. alterniflora*-specific marker and presence of all *S. foliosa*-specific markers, or (b) absence of two *S. foliosa*-specific markers and absence of all *S. alterniflora*-specific markers among reactions that successfully amplified.

High confidence evidence of hybridity is defined as the (a) presence of at least one *S. alterniflora*-specific marker and the absence of at least one *S. foliosa*-specific marker, or (b) the presence of at least two *S. alterniflora*-specific markers among reactions that successfully amplified.

Very high confidence evidence of hybridity is defined by the presence of all four *S. alterniflora*-specific markers and the absence of any *S. foliosa* and any *S. anglica*-specific markers among reactions that successfully amplified. (Note: *S. anglica* contains all *S. alterniflora*-specific markers in addition to *S. anglica*-specific markers. Thus presence of *S. anglica*-specific marker would indicate that the sampled plant was *S. anglica*, not *S. alterniflora*.)

Microsatellite Results Analysis

Microsatellite results were reviewed for consistency of internal controls run on each plate. These results were not completely consistent, but varied as shown in **Table 5 SSR Internal Control Results**. One set of controls was selected for use during analysis.

Microsatellite results were analyzed using the free software package *structure* which uses a clustering method to investigate population structure in genetic data. This software can be used to assign individuals to populations and to study hybrid zones (Pritchard et al. 2000). This software is used by the UC Davis for microsatellite analysis, is used frequently by other North American population biologists for such analyses.

In our *structure* analysis we follow the assumptions used by the UC Davis Spartina Lab in their analyses. Although *S. alterniflora* and *S. foliosa* are hexaploid species, we follow the assumption that the selected microsatellites behave as diploid markers (Blum et al. 2004); thus we did not modify the ploidy parameter in *structure*. By running *structure* with this diploid model, we did not allow for recessive or null alleles in our *structure* analyses. The structure model assumes that, within populations, loci are at Hardy-Weinberg equilibrium and linkage equilibrium (Pritchard et al. 2009). Because ISP was unable to identify any other models which did not include or allowed for violation of this assumption, the *structure* model was applied although Blum et al. (2004) and Sloop et al. (2006) show this assumption to be violated in many of the microsatellite loci used in 2009.

We tested the sensitivity of results to different parameters and settings in our *structure* analyses, such as number of populations (K), assumption of admixture or no admixture, inclusion of population data or no population data, identification of species controls in the model, and input of the full Bay dataset versus local subsets of the data. We compared a total of 6 sets of results from *structure* after applying multiple permutations of these parameters and settings. We found that the model was most sensitive to the dataset entered (full Bay dataset versus local subsets of the full dataset) and to the number of populations (K) assumed. Results changed only slightly when we added information regarding source location or field identification and field id confidence (using the optional columns LOCDATA and/or POPDATA in the model). (The $q^{(17)}$ values changed by 0.02 or less for 87% of the samples, and the maximum change was 0.07 for one sample.)

We performed 2009 data analysis using an admixture model with no population prior and including all samples collected in 2009.

We set K=2 set to model two clusters, corresponding to *S. foliosa* or *S. alterniflora*. We were able to determine the species corresponding to cluster 1 and cluster 2 based on the values of the three control samples (one *S. foliosa* sample and two *S. alterniflora* samples) which *structure* consistently assigned to cluster 1 or cluster 2. We also performed informal checks for correspondence between high confidence field ID samples with their assigned clusters.

We set an arbitrary inferred proportion of 0.75 *S. foliosa* ancestry as the minimum value defining "pure" *S. foliosa*. We set an arbitrary inferred proportion of 0.75 *S. alterniflora* ancestry as the minimum value defining a genetically discernable *S. alterniflora x foliosa* hybrid. Since there are only two clusters, the proportions for each of the two clusters sum to 1. Samples with values of < 0.75 for both parent species are inferred to have intermediate proportions of ancestry from both parental species, based on the eight microsatellite markers analyzed.

Up to eight microsatellite markers were analyzed, and data from all samples (including those with missing data) were included in the *structure* analysis. Results from primers Spar 20 and Spar 25 were not included in the analysis. Use of Spar 25 was discontinued after STA Labs advised the ISP to discontinue its use, as the first three plates of results (with 96 samples per plate) yielded identical alleles (245 and 253) for all samples resulting from this primer. Use of Spar 20 was discontinued following the advice of our genetic consultant who found the low diversity of alleles among samples to yield this primer suspect and/or uninformative (see **Table 6 2009 SSR Allele Frequencies**).

RESULTS

Bay Area Counties All Affected

As shown in **Map 1 2009 subarea centroids for axf.jpg**, which displays 2009 net acres of hybrids by ISP subarea, *S. alterniflora x foliosa* hybrids are now found throughout the project area in all nine Bay Area counties (Alameda, Contra Costa, Marin, Napa, San Francisco, San Mateo, Santa Clara, Solano and Sonoma), but are primarily concentrated in those subareas located in San Mateo and Alameda counties.

S. densiflora was primarily found in those subareas located in Marin County (**Map 2 2009 subarea centroids for den.jpg**), where it was originally planted. Hybrids between S. densiflora and *S. foliosa* were identified in many of the subareas where S. densiflora was found (**Map 3 2009 subarea centroids for ang dxf pat.jpg**). *S. anglica* remained restricted to one location, Creekside Park, in Marin County, where it was originally planted. *S. patens* remained restricted to Benicia State Recreation Area in Solano County.

Baywide Populations in Decline

The approximate 158 net acres of invasive *Spartina* remaining in 2009 represents a Baywide decline of 42% since 2008 and an 80% reduction since full-scale treatment began in 2005 (**Map 4 Baywide Net Acres.jpg**). Graphs showing the trajectory of net acres over time are presented for S. densiflora, *S. alterniflora x foliosa* hybrids and all invasive *Spartina* in **Figures 1.1, 1.2 and 1.3 Bay and Outer den axf all graphs.xls**.

At the height of the infestation in 2005, we estimated just over 800 net acres of invasive *Spartina*. Our inventory results from 2006 indicated a decline of approximately 25% to just fewer than 600 net acres of invasive *Spartina*. In 2007 our inventory results indicated a decline of an additional 65%; however 2007 data was artificially low for the many large areas mapped by digitizing aerial imagery. During the 2008 field season we mapped approximately 275 net acres of invasive *Spartina*, and we mapped just less than 160 acres in 2009.

The increase in net acres mapped between 2007 and 2008 was seen throughout the Bay, but was especially pronounced in the Southern South Bay region (south of the Dumbarton Bridge), where the majority of 2007 mapping efforts were performed via digitizing aerial images. As explained in the discussion section below, aerial images in 2007 were difficult to interpret due to highly successful treatment of invasive *Spartina* in 2005 and 2006. Thus, 2007 monitoring data are consistently, artificially low for all sites that were digitized in 2007.

We estimate a 25% decline in area requiring treatment between 2008 and 2009. A total of 322 acres (between 276 – 358 acres) Baywide were estimated as requiring treatment for invasive *Spartina* in 2009. These numbers were down from 431 acres (between 368 – 482 acres) in 2008. These estimates of acres requiring treatment are based on the mean value of the treatment cover class ranges recorded in the field by staff for individual *Spartina* features during inventory monitoring, and include all species of invasive *Spartina*. These numbers differ from net area, as explained in the box *Reporting *Spartina* Area* above.

New Populations

Within ISP Bay Regions, annual trends have mirrored those of the Baywide decline except where populations are small and discovery of new populations has led to increases. ISP Bay Regions are defined by the Subtidal Habitat Goals Report (Goals Project 1999) and are further subdivided into Northern South Bay and Southern South Bay by the Highway 84 Dumbarton Bridge, as shown in **Map 5 All Spartina 04-09 by ISP Bay Region.jpg**. Within the Central Bay and Northern South Bay, where the largest infestations are located, treatment has led to substantial population declines. In the Southern South Bay, declines are not as dramatic, a result of difficulty in plant identification during monitoring efforts (as discussed later) and less effective treatment (as described in ISP Treatment Reports) in this region compared to other regions. Total net acres of invasive *Spartina* have remained stable or increased in the Outer Coast, North Bay and Suisun Bay regions as a result of the discovery of new populations of invasive *Spartina* in these regions. Discovery

of new populations of hybrids in Suisun Bay and of both hybrids and S. densiflora in the North Bay led to increases in net area of invasive *Spartina* in these regions (**Map 6 ISP Bay Regions 2004-09 axf bar graph symbols.jpg, Map 7 ISP Bay Regions 2004-09 den bar graph symbols.jpg**). The Outer Coast, North Bay and Suisun Bay have had very few populations of invasive *Spartina* and thus annual summary data in these more remote regions is especially sensitive to discoveries of even small new populations.

Trends Vary By Subarea

The ISP Control Program divides its 25 treatment sites into 173 subareas for logistical and reporting purposes. Net area of invasive *Spartina* for 2001 and 2004-2009 within these sites and subareas is summarized in **Table 7 Acres By Subarea By Year.xls**. Change in net area of invasive *Spartina* within sites and subareas between 2008 and 2009, and between the height of the infestation (2005) and 2009 is presented in **Table 8 Change in Acres 09 since 05 and 08.xls**. Estimated area requiring treatment for 2008 and 2009 as calculated by treatment area (see box *Reporting *Spartina* Area*) are summarized by subarea in **Table 9 Trtmnt Acres By Subarea By Year.xls**. Change varies greatly by subarea. While overall Baywide net acres were reduced by 42% between 2008 and 2009, reductions reached up to 90% in some subareas. Net area of invasive *Spartina* did not decline but even increased in other subareas. Extensive descriptions of treatment activities at each subarea can be found in the Annual ISP Treatment Reports written and compiled by the ISP Control Program, and can serve to explain some of this variability in trends by subarea.

Genetic vs Phenotypic Evidence of Hybridity

2008 Results

Baywide in 2008, 990 samples were successfully tested with RAPDs and linked to DNA sampling locations in our GIS. Of these, 492 samples were identified as *S. foliosa* in the field and successfully tested with RAPDs.

Results showed no evidence of hybridity for 81% of these samples, supporting the *S. foliosa* field identifications for these 395 samples.

Results suggested evidence of hybridity with *S. alterniflora* for 19% of the samples (97 samples). Of these, the majority (33) were samples that were field-identified as *S. foliosa* with moderate field id confidence and having "medium confidence" evidence of hybridity. Only five samples were field-identified as *S. foliosa* with high field id confidence and had "high confidence" evidence of hybridity.

Of the 427 samples identified as *S. alterniflora* / hybrids in the field and successfully tested with RAPDs in 2008, 48% of the samples were found to have evidence of hybridity and an additional 6% were identified as possible pure *S. alterniflora* plants based on the presence of all *S. alterniflora*-specific markers and the absence of all *S. foliosa*-specific markers.

A full 45% of the field-identified hybrid plants (192 samples) were found to have no evidence of hybridity based on RAPD testing. Of these, 49% (95 samples) were field-identified as hybrids with low field id confidence and lab-identified as *S. foliosa* with high to medium lab id confidence. Only fourteen samples were field-identified as *S. alterniflora* / hybrids with high field id confidence and lab-identified as *S. foliosa* with high to medium lab id confidence; these represent plants that appear to be hybrids based on the consideration of all field characteristics, but for which RAPD results find all *S. foliosa* alleles present and all *S. alterniflora* alleles absent.

2009 Results

Baywide in 2009, 1032 samples were successfully tested with microsatellite primers, 340 samples were successfully tested with RAPD primers, and both results were linked to DNA sampling locations in our GIS.

Both RAPD and microsatellite results from 2009 provided evidence of hybridity for greater numbers and greater percentages of field-identified *S. foliosa* samples than in any past years. In past years (2004-2008), up to 23% of samples (up to 70 samples per year) identified as *S. foliosa* in the field with moderate to high confidence, based on morphology, location and any other phenotypic characteristics, were identified as hybrids based on RAPD testing. In 2009, well over 100 samples of moderate to high confidence field identified *S. foliosa* samples were found to have genetic evidence of hybridity. Of the 2009 samples identified as *S. foliosa* in the field with moderate to high confidence, 42% were found to have evidence of >75% *S. alterniflora* ancestry based on microsatellites results, and of the subset tested with RAPDs, 61% were found to have evidence of hybridity (**Figure 1.8**).

Genetic results in 2009 for field-identified hybrids were in line with expectations based on previous years (**Figure 1.9**).

Species and Site Updates

Spartina patens

Spartina patens remained restricted to Southampton Marsh at Benicia State Recreation Area (**Map 8 patens at Southampton.jpg**). Its population continued to decrease, down from a height of approximately 2,625 net m^2 in 2005, to 102 net m^2 in 2008 and 68 net m^2 in 2009.

Spartina anglica

Spartina anglica remains restricted to Corte Madera's Creekside Park, where the population has declined from a height of approximately 439 net m^2 in 2004 to 311 net m^2 in 2009 (**Map 9 anglica at Creekside.jpg**).

Data from 2008 indicated very low abundance of *S. anglica* at Creekside Park (20 net m^2) due to difficulty in identification of *S. anglica* in late July when inventory monitoring was conducted. In 2009, inventory monitoring for *S. anglica* was conducted in early June, when *S. anglica* is more readily distinguished from *S. foliosa* by its inflorescence, leading to better mapping of this species.

Spartina densiflora

Spartina densiflora is the second most abundant and widespread species of invasive *Spartina* in the Bay area. In 2008, 2.8 acres required treatment of *S. densiflora* populations. These were located in the Outer Coast (Tomales Bay), San Pablo Bay (Mare Island and Point Pinole), Southeast Marin (multiple sites), and the San Francisco Peninsula (Sanchez Marsh in Burlingame), (**Map 2 2009 subarea centroids for den.jpg**). Populations have been declining steadily in Southeast Marin and the Outer Coast as a result of annual treatment.

No new populations of *S. densiflora* were found in 2008 or 2009, but new patches, located >500 m from patches in past years, were located in the northwestern corner of Sanchez Marsh and west of the population discovered on Mare Island in 2007. In 2009, one new patch 3 meters in diameter was discovered in an urbanized area of Corte Madera Channel, behind a house on the south side of Hickory Avenue.

Spartina densiflora x foliosa

Spartina densiflora x foliosa hybrids were found in Southeast Marin and in Burlingame's Sanchez Marsh prior to 2008. In 2008, new locations were identified based on genetic and field identification *S. densiflora x foliosa* hybrids. RAPD results confirmed field ID of samples in Creekside Park and in the new location of Muzzi Marsh. Other new locations for 2008 included Starkweather Park, Larkspur Ferry Landing, Corte Madera Creek, Corte Madera Ecological Reserve, and Blackie's Pasture and Creek (**Map 10 new 2008 dxf locations.jpg**).

New *S. densiflora x foliosa* hybrids were found in 2009 at Hog Island Oyster Company in Tomales Bay, Mare Island in Suisun Bay, and in various Southeast Marin locations: San Rafael Creek Mouth North Bank, Piper

Park, Starkweather Park, Triangle Marsh, Greenwood Cove and Strawberry Point (**Map 11 new 2009 dxf locations.jpg**).

S. alterniflora x densiflora x foliosa

Field characteristics and genetic results indicated possible formation *S. alterniflora x densiflora x foliosa* hybrid plants at Muzzi Marsh (adjacent to the *S. densiflora x foliosa* plants identified in 2008), Corte Madera Creek under the highway ramp east of Corte Madera Rowing Club, and at Sanchez Marsh in 2008 (**Map 12 axdxf extent 2008.jpg**). These plants were mapped as *S. alterniflora x foliosa* hybrid, Possible *S. anglica*, and *S. densiflora x foliosa* hybrid, respectively, in 2009, and no *S. alterniflora x densiflora x foliosa* hybrids were mapped in 2009. All plants were treated as non-natives for control purposes, regardless of the exact genetic make-up of the plants as indicated by RAPD results.

A patch with intermediate morphology between *S. densiflora* and *S. foliosa* was identified as an *S. densiflora x foliosa* plant at Sanchez Marsh, both through genetic testing and field identification, in 2007. This patch had *S. densiflora* features such as curled leaves, but did not have the bunchgrass morphology indicative of *S. densiflora*. RAPD testing in 2008 found markers indicative of *S. alterniflora* (D11 575) and *S. foliosa* (A17 725, B7 800, D5 1100). There were no results for the *S. densiflora* markers. Based on the genetic results indicating presence of both *S. alterniflora* and *S. foliosa* alleles, and the curled leaf morphology indicative of *S. densiflora*, two plants at this site were classified by ISP biologists as *S. alterniflora x densiflora x foliosa* in 2008. These two patches were classified as *S. densiflora x foliosa* in 2009. Regardless of exact genetic identity, both patches were treated.

Spartina alterniflora x foliosa

Net area of hybrids declined by approximately 42% between 2008 and 2009 in response to coordinated treatment efforts, corresponding to a 26% decline in area requiring treatment (**Tables 1 and 2**). In 2008, hybrids covered 427 of the total 431 acres of invasive *Spartina* (of any kind) requiring treatment throughout the Bay, and made up 294 out of the total 295 net acres of invasive *Spartina*. In 2009, hybrids covered 318 of the total 322 acres requiring treatment throughout the Bay, and made up 156 out of the total 158 net acres of invasive *Spartina*. See **Map 1 2009 subarea centroids for axf .jpg**.

Morphologies, phenologies, growth characteristics and other phenotypic characteristics can vary widely between individual *Spartina alterniflora x foliosa* hybrids (Callaway and Josselyn 1992, Anttila et al. 1998, Daehler et al. 1999, Ayres et al. 2004a, Ayres et al. 2008b). Some plants are easy to identify, while others can be extremely difficult to distinguish from *S. foliosa*. Due to this difficulty, level of confidence in field identification is recorded during monitoring.

The vast majority of the *S. alterniflora x foliosa* acres mapped contained plants identified in the field with high confidence in both 2008 and 2009 (91% and 93% respectively). In both years, about 4% of the plants were identified as *S. alterniflora x foliosa* with moderate confidence, around 1% were identified with low confidence, and a few plants (< 0.1% in terms of acreage) were mapped in the field as unknowns (possible *S. alterniflora x foliosa*). The plants in this latter category could not be definitively identified by genetic testing of associated DNA samples; these features were noted for follow-up surveys and genetic testing in future years.

Evaluation of Species Identifications

Multiple Lines of Evidence

Identification of hybrids is especially difficult when and where the hybrid plants are not dramatically distinct from the native plants. The timing of inventory has a significant impact on the ability to distinguish hybrids from natives. Hybrids may become green earlier or stay green later than natives, and hybrids may have longer inflorescences bearing more seeds than natives (Anttila et al. 1998, Ayres et al. 2008b). Waiting until all *Spartina* is at full growth and is flowering is not possible at all sites, however, due to staffing limitations and timing considerations; data is required for treatment during the growing season, sometimes prior to flowering time.

Multiple lines of evidence augment field identification, including past year's inventory data and lab results brought into the field via GPS and maps. Using morphology, phenology, environment and these additional lines of evidence from past years, *Spartina* is mapped as accurately as possible in the field, with the understanding that data may be subject to later evaluation.

Evaluation of past and subsequent years' inventory data and lab results in GIS can lead to the re-evaluation of the final determination of species. During data QC, ISP biologists use these multiple lines of evidence to finalize their species designations.

Most edits performed during QC have little impact on Baywide summary data, but may have significant impacts within a site. Re-evaluation of final species designations for several very large features collected using the new helicopter monitoring method in 2008 did lead to a significant impact on both summary data and site data, as described below for Calaveras Point and Greco Island North.

Lab Results Inform Monitoring Results

Lab results from 2008 informed the final species designation of 22 acres worth of *Spartina* (measured in "treatment area"), as mapped by 1079 features (points, lines and polygons) in 2008. (As mentioned above, each genetic sample collected is uniquely linked to a corresponding GPS point, line or polygon feature.) Biologists chose to question lab results when determining their final species designations for only 56 of these mapped *Spartina* features, accounting for 4 acres.

One plant with a *S. alterniflora x foliosa* lab result was determined by the biologist to be *S. alterniflora* x densiflora x foliosa, and two plants with *S. foliosa* lab results were determined by the biologist to be *S. densiflora x foliosa*, based on the morphology of these plants. Biologists chose to designate as *S. alterniflora x foliosa* 39 features (accounting for 0.4 acres) from which lab results indicated *S. foliosa.*

Biologists chose to designate as *S. foliosa* four features (accounting for 3.8 acres) from which DNA results indicated evidence of hybridity. One of these features was a 3.6 acre polygon, and is discussed under the Greco Island North heading below.

Microsatellite and RAPD results were consulted to inform the final species designations of associated features collected in 2009. Of the 416 features identified as hybrids in the field and for which a DNA sample was collected, 20 were given a final species designation of "Possible *S. alterniflora*/hybrid" and 82 were given a final species designation of *S. foliosa.* Of the 543 features field-identified as *S. foliosa* and for which a DNA sample was collected, 41 were changed to "Possible *S. alterniflora*" and 63 were given final species designations as hybrids. Remaining features were given final species designations matching field identifications. Of those features for which final species designation was changed from the original field ID, changes were based on both 2009 genetic results and 2010 field observations. (This was possible because of the lag time in finalization of 2009 genetic results analysis, after completion of the 2010 field season.)

New Hybrid Populations Detected

New populations of *S. alterniflora x foliosa* ("hybrids") were detected in several locations around the Bay in 2008 and 2009 (**Map 113 new axf and possible sites in 2008.jpg, Map 14 new axf and possible sites in 2009.jpg**). New populations are defined as patches of invasive *Spartina* further than one kilometer away from invasive *Spartina* mapped in any previous year, and/or in sites never before surveyed by the ISP.

Four new populations were discovered in 2008 on property of the San Pablo National Wildlife Refuge on the shoreline of Mare Island, along the Benicia and Southampton Marsh shorelines, and in the recently restored area of Eden Landing. Genetic results from RAPD testing indicated the potential presence of *S. alterniflora*-specific RAPD alleles in Tomales Bay and Gallinas Creek in 2008. These sites are discussed below.

Three new hybrid populations were discovered in 2009 in the area of Limantour Estero, on the shoreline of Point Pinole, and within Plummer Mitigation Marsh. An additional four sites were identified as containing possible hybrids based on genetic evidence of potential *S. alterniflora* ancestry detected through microsatellite

and/or RAPD testing in 2009. These included locations along the Benicia shoreline, Grey's Field in Petaluma, Carquinez Straight Regional Shoreline, and Point Richmond.

Limantour Estero

A relatively large (4 m radius) patch of field-identified *S. alterniflora x foliosa*, or possibly pure *S. alterniflora*, was discovered between Drakes and Limantour Esteros during a kayak survey in November 2009 (**Map 15 2009 Limantour Estero Results.jpg**. This patch was obviously not native based on its morphology. Flowers were removed at time of discovery. ISP staff returned to tarp this patch in January 2010, with follow-up tarping conducted in June 2010.

Grey's Field

Grey's Field in Petaluma is a passive restoration area resulting from an unrepaired accidental levee breach. *Spartina* is colonizing the site, and the ISP collected samples of *Spartina* for genetic testing in and around Grey's Field beginning in 2006. (See **Maps 16-18 Grey's Field 2007-2009.jpg**.)

Results of genetic sampling provided weak evidence of hybridity at this location. Eleven samples were collected in 2008, and of these two had evidence of hybridity based on the presence of a single *S. alterniflora*-specific RAPD marker (B7 550/650) (**Map 17 Grey's Field 2008.jpg**).

Analysis of seven to eight microsatellite markers inferred high levels of *S. foliosa* ancestry (97%) for six samples and suggested extremely low levels of *S. alterniflora* ancestry (29% and 35%, shown in grey in **Map 18 Grey's Field 2009 Results.jpg**) for two samples in 2009. For two locations within the site, our data seem to indicate different lab results for samples collected from the same distinct patches of colonizing *Spartina* in multiple years. The sample on the inboard side of the levee north of the breach contained a single *S. alterniflora*-specific marker (B7 550) in the 2008 RAPD results, but a sample collected approximately 2 meters away within the same putative patch of *Spartina* contained only *S. foliosa*-specific RAPD markers when tested in 2009. The 2009 sample collected here did have evidence of hybridity based on the microsatellite results (inferred 35% *S. alterniflora* ancestry, as mentioned above). The 2008 sample in the southeastern corner of the marsh contained two *S. alterniflora*-specific RAPD markers (B7 550/650). A sample collected in 2009 just 3 meters away contained only *S. foliosa*-specific RAPD markers. No RAPD testing was performed on the other three 2009 samples in this vicinity. The sample inferred to be of marginal (29%) *S. alterniflora* ancestry was located nearby, approximately eight meters away, lending weak evidence for the potential of hybridity at this location.

The plant on the inboard levee north of the breach was treated in 2009, as it had some discernable morphological characteristics indicative of hybrids and had consistent genetic evidence of hybridity in 2008 and 2009. The plants in the southeast corner of Grey's Field were relocated by the Control Program in 2009 but were deemed to be native and were thus not treated. The ISP will continue to monitor and collect samples for genetic analysis of the plants in the southeast corner and in the rest of the marsh.

San Pablo National Wildlife Refuge: Mare Island Shoreline

In 2007, *S. densiflora* and *S. alterniflora x foliosa* were found along the bayfront of Mare Island on the property of San Pablo Bay National Wildlife Refuge. In 2008 the entire bayfront of Mare Island was surveyed by foot, leading to the detection of more plants and a new area of *S. densiflora* plants (**Map 19 2008 SPBNWR.jpg**). (Note: all *S. densiflora* inflorescences were cut off and removed from the site at time of survey to prevent further spread by seed prior to digging at a later date.)

A previously undetected *S. alterniflora x foliosa* hybrid clone was found along Sonoma Creek just north of Highway 37 while surveying by boat in 2008. A new ISP subarea was created (26c: Sonoma Creek) and the ISP Control Program coordinated treatment at this new site.

Benicia and Southampton Marsh Shoreline

Along the shoreline of Benicia State Recreation Area's Southampton marsh, hybrids were discovered for the first time in 2008. These plants were identified by biologists as possible hybrids, and DNA samples were

collected. Results indicated presence of *S. alterniflora*-specific RAPD markers in the five samples tested (**Map 20 Benicia 2008 DNA.jgp**). Four samples contained three of the four *S. alterniflora*-specific markers (with C10 470 lacking), and one sample contained all four of the *S. alterniflora*-specific markers. All of the *S. foliosa*-specific markers among reactions that successfully amplified were absent in these five samples.

The patches along the shoreline from which these samples were collected were distinguishable from the native plants based on morphology, and were treated.

In the interior of Southampton Marsh, two samples of field-identified *S. foliosa* were collected in 2008. All 2008 RAPD results found only *S. foliosa*-specific markers to be present. In 2009, six samples were collected in the interior of the marsh; analysis of eight microsatellite markers provided evidence of 85% *S. foliosa* ancestry for one of these samples, and evidence of 40%, 60%, 91% and 78% *S. alterniflora* ancestry for the other five individuals (**Map 21 Benicia 2009 DNA.jpg**). The samples inferred to have 40% and 78% *S. alterniflora* ancestry based on microsatellite data also had evidence of hybridity via RAPD testing. This evidence was based on presence of one *S. alterniflora*-specific RAPD marker (X18 950) for the former sample, and both the presence of one *S. alterniflora*-specific RAPD marker (X18 450) and the absence of one *S. foliosa*-specific RAPD marker (B7 800) for the latter sample. The sample inferred to have 60% *S. alterniflora* ancestry based on microsatellite data contained no evidence of hybridity based on testing of all nine species-specific RAPD markers.

The patches from which these interior marsh samples were collected were not distinguishable from surrounding *Spartina*. These patches were not treated, but were noted for follow-up monitoring.

Southeast of Southampton marsh, at Benicia's Matthew Turner Shipyard Park (at the bay shore end of 12th Street), a patch mapped as low field id confidence hybrid in 2007 was tested with RAPDs in 2008, with results suggesting the plant could be pure *S. alterniflora* based on the presence of all four *S. alterniflora*-specific markers tested and the absence of the one *S. foliosa*-specific markers tested (**Map 22 Matthew Turner Shipyard DNA.jpg**). This 2.5 meter diameter patch and an adjacent, 4 meter diameter patch, were mapped in 2008.

In 2009, another sample was taken at this site, 60 meters northwest of the 2008 sample. This sample was field-identified as *S. foliosa*. Analysis of eight microsatellite markers provided evidence for 94% *S. alterniflora* ancestry.

Two samples of field-identified *S. foliosa* were collected in 2009 along the shoreline between Matthew Turner Shipyard Park and Southampton Marsh, and both had genetic results indicating evidence of hybridity. Analysis of eight microsatellite markers provided evidence for 87% and 76% *S. alterniflora* ancestry for northern and southern individuals, respectively. The southern sample was also tested with microsatellites, which indicated hybridity based on the presence of one of four *S. alterniflora*-specific markers (X18 450) and the presence of three *S. foliosa*-specific markers. (Note: RAPD primer B7 did not amplify for this sample.)

Based on the morphological evidence, these samples along the Benecia shoreline were not treated but were noted for continued monitoring.

Carquinez Strait

Field ID and lab results indicated presence of a possible hybrid patch at the Carquinez Strait Regional Shoreline pier in 2009 (**Map 23 Carquinez Pier 2009.jpg**). Analysis of eight microsatellite markers suggest this individual to be of 70% *S. alterniflora* ancestry. RAPDs provided evidence of hybridity based on the presence of a single *S. alterniflora*-specific marker (D5 600). The Control Program made plans to treat this patch in 2010.

Genetic results for samples taken along the shoreline of Carquinez Strait between Crocket and Martinez (**Map 24 Carquinez Strait DNA 2009.jpg**) provided evidence of weak hybridity, but plants were identified as natives through field identification. One sample's RAPD results indicated hybridity based on the presence of a single *S. alterniflora* marker (B7 650), and had no clear ancestry based on microsatellite data (inferred 36% *S. alterniflora* ancestry based on eight markers). Further east, another sample's RAPD results

indicated hybridity base on the presence of a different single *S. alterniflora*-specific marker (X18 450), but was inferred to be of 96% *S. foliosa* ancestry based on the analysis of eight microsatellites. Further east still, at Waterfront Park in Martinez, two samples were inferred to be of 97% *S. foliosa* ancestry based on eight microsatellites each. One of these samples was also tested with RAPDs which showed evidence of hybridity based on the presence of a single *S. alterniflora*-specific marker (B7 650). As the evidence of hybridity is considered weak, these areas will continue be monitored but no control actions have been taken at this point.

Point Pinole & Shoreline

On the eastern tip of Point Pinole, a 3 meter diameter patch of field-identified hybrid was mapped for the first time in 2009. A genetic sample was collected, and analysis of 7 microsatellite markers indicated 95% *S. alterniflora* ancestry.

Patches of hybrid were discovered based on both morphology and genetic testing along the shoreline east of Point Pinole (**Map 25 Point Pinole Results 2009**). In past years this area was monitored by boat, which was faster but required monitoring from a greater distance. This area was walked for the first time in 2009, at which time these hybrid plants were discovered and five samples were collected.

Analysis of microsatellite results inferred four of the five individuals to be of >90% *S. alterniflora* ancestry based on analysis of eight microsatellite markers. The fifth sample was inferred to be of 38% *S. alterniflora* ancestry based on analysis of seven microsatellite markers.

RAPD testing was performed on four of these samples and provided evidence of hybridity for all four. The three plants identified as hybrids based on morphology contained three *S. alterniflora*-specific RAPD markers (D11 575, X18 450, X18 950) and all *S. foliosa*-specific RAPD markers. The plant identified as *S. foliosa* in the field contained the single *S. alterniflora*-specific RAPD marker X18 450 and all *S. foliosa*-specific RAPD markers. (Note: RAPD primer B7 did not amplify in this latter sample, so B7 markers were not scored.)

The ISP Control Program is coordinating treatment of these newly discovered patches of hybrid along the Pinole Shoreline.

Point Richmond

In 2001, two samples collected on the northern shoreline of Point Richmond between Point San Pablo Yacht Harbor and Wildcat Marsh were identified as hybrids based on RAPD testing. No hybrid plants were discovered in this vicinity again until 2009, when two small (1 meter diameter) patches of field-identified hybrid were observed at Point San Pablo Yacht Harbor. Samples were collected at these two patches and from nearby patches that appeared to be *S. foliosa* in 2009. Analysis of eight microsatellite markers suggested >90% *S. alterniflora* ancestry for nine of the twelve samples collected between Point San Pablo Yacht Harbor and Wildcat Marsh. (See **Map 26 San Pablo Wildcat Results 2001.jpg**, **Maps 27-31 San Pablo Wildcat Results 2004-8.jpg, & Map 32 Point San Pablo Results 2009.jpg**)

Eden Landing

Eden Landing, part of the South Bay Salt Pond Restoration Project, was restored to tidal action in fall 2006. This site was monitored by the ISP for the first time in late 2007, at which time the discovered infestation of hybrid *Spartina* was mapped and treated. Treatment of the approximately 2,000 net m^2 mapped in this subarea (13h) in 2007 led to a decline to just under 500 net m^2 in 2008(**Map 33 Eden Landing Restoration 2008 Results.jpg**).

Plummer Creek Mitigation Marsh

The mitigation marsh at the north end of Plummer Creek has had “suspicious” plants sampled for genetic testing annually since 2003 (with the exception of 2007). DNA results were returned as native until, in 2008, one of eight samples collected at this marsh was identified with low confidence as a hybrid based on the absence of a single *S. foliosa*-specific RAPD marker (A2 575).

In 2009, several patches of hybrids were mapped based on morphology, and four DNA samples were collected. Two of these were inferred to be of 87 and 89% *S. alterniflora* ancestry based on analysis of eight and seven microsatellite markers, respectively. The other two were inferred to be of 97% *S. foliosa* ancestry based on analysis of eight microsatellite markers (**Maps 34 - 39 Plummer Creek Mitigation Marsh 2003-2009**).

A patch of potential hybrid was noted in a difficult-to-access location from across a channel and was mapped as a possible hybrid, to be tested in 2010.

Species Evaluation Case Studies

Tomales Bay

Since 2002, the ISP has surveyed and taken genetic samples in Tomales Bay to test for the presence of hybrids. In 2008, for the first time, a sample collected on a channel edge approximately 370 meters north of Waldo's Dike contained evidence of hybridity based on RAPD testing. (**Map 40 Tomales 2008 DNA.jpg**). Only two of the RAPD primers that yield *S. foliosa*-specific markers successfully amplified for this sample, but both expected *S. foliosa*-specific markers were present. All four of the RAPD primers that yield *S. alterniflora*-specific markers amplified successfully, and a single *S. alterniflora*-specific marker (D11 575) was found to be present. This sample was one of nine samples collected, with the other eight samples containing no evidence of hybridity.

Taking into consideration the potential for error in lab identification based on a single marker, and the fact that the patch sampled had no observable differences in growth, morphology or phenology from the surrounding acres of *S. foliosa*, the ISP determined that more genetic sampling in the 2009 season was prudent prior to finalizing the species determination of this plant and/or initiating any control actions.

This same patch was resampled in 2009 and assigned to the *S. foliosa* cluster based on *structure* analysis of microsatellite results (with no missing microsatellite data).

A total of 35 samples were tested from Tomales Bay in 2009. (See **Map 41 Tomales 2009 DNA map.jpg**) For one of these, just six meters away from the 2008 RAPD-identified hybrid, 2009 RAPD results indicating hybridity based on a single, but different, *S. alterniflora* marker (B7 550). This same sample was inferred to have a high proportion of *S. foliosa* ancestry using microsatellite markers (with only 11% missing data).

Analysis of microsatellite results for one sample collected on the northern bank of Walker Creek inferred an 83% proportion of *S. alterniflora* ancestry. This location was noted for revisit and evaluation in 2010.

The remaining 27 samples from Tomales Bay in 2009 were lab-identified as *S. foliosa*, with the majority of these (26) having inferred proportions of 94% or greater *S. foliosa* ancestry.

Based on the extent of native lab results, the lack of detectable hybrid traits in the samples collected, and the lack of any change in extent or density of the patch identified by RAPDs as hybrid in 2008, the ISP believes the weight of evidence indicates that the samples identified as hybrids by RAPDs in the area north of Waldo's Dike are actually native.

Tolay Creek

A genetic sample collected along Tolay Creek in 2008 that was identified in the field as *S. foliosa* was found to have evidence of hybridity based on a single *S. alterniflora*-specific RAPD marker (D11 575). (**Map 19 2008 SPBNWR.jpg**.) The site was re-visited later in the season upon receipt of lab results, and the *Spartina* in the vicinity of the sample was re-assessed for any visible hybrid characteristics. In addition, past year's aerial imagery was viewed to assess the rate of spread of *Spartina* at this site. Neither the field visit nor the review of aerial imagery indicated any reasonable suspicion of hybridity or invasive characteristics. Based on

the extent of the *Spartina* at this site, and the lack of evidence for hybrid characteristics, the lab results were considered to be potentially erroneous and the site was not treated by the ISP Control Program.

The same patch within Tolay Creek was resampled in 2009 and analysis of eight microsatellite markers inferred a high proportion of *S. foliosa* ancestry (97%). Additional samples collected within Tolay creek were also all inferred to be of 97% *S. foliosa* ancestry based on eight microsatellite markers. The ISP biologists who have monitored this site believe these plants to be native, and no treatment has been undertaken at this location.

White Slough

White Slough, in Vallejo, has been a confusing site since 2007 RAPD results indicated presence of *S. alterniflora x foliosa* hybrids. Eight of thirteen samples collected during initial monitoring in November 2007 were field-identified as hybrids based on morphology. Of all thirteen November samples, RAPD results only indicated one of these samples to be a hybrid, based on presence of the C10 470 alterniflora marker.

Based on the concerns of the field staff in November, Control Program staff returned to this site in December 2007 and sampled all unsampled patches, for a total of 72 samples collected at this site in 2007 (**Map 42 White Slough 2007 DNA.jpg**). Thirteen samples were field-identified as *S. alterniflora x foliosa* and 46 were field-identified as *S. foliosa*. Of all 59 December samples, 19 were identified by RAPDs as hybrids. (Of these, only two were field-identified as hybrids.) Of the 19 RAPD-identified hybrids at White Slough, 17 were identified based on presence of the C10 470 marker. One sample was identified based on the D5 600 alterniflora marker, and one sample (for which the C10 primer did not work) was identified based on absence of the D5 1100 foliosa marker.

Follow-up monitoring and genetic testing at this site in 2008 continued to cause concern by identifying all samples as native, including samples from patches that had been lab-identified as hybrids in 2007. Seven samples were collected in 2008, with four field-identified as *S. alterniflora x foliosa* based on 2007 lab ID and/or morphology. Lab results in 2008 indicated that all seven samples were *S. foliosa* (**Map 43 White Slough 2008 DNA.jpg**). (Note that amplification was successful for C10 for all samples, but failed for D5 for 1 sample. Foliosa marker D5 1100 was not analyzed for any samples in 2008 due to concerns regarding species specificity.)

In 2009, microsatellite and RAPD results gave variable results once again (**Map 44 White Slough 2009 DNA.jpg**). RAPDs in 2009 found presence of additional alterniflora markers (B7 550 and 650, D5 600, D11 575 and X18 450). Primer C10 was not used in 2009 RAPD analysis.

Gallinas Creek

Gallinas Creek in Marin County was surveyed by the ISP for the first time in 2008. Based solely on the genetic testing of what was characterized in the field as *S. foliosa*, two patches of hybrids totaling 22 square meters (treatment area) were mapped at the southern end of Gallinas Creek in 2008. These lab results were based on the presence of one *S. alterniflora*-specific RAPD marker each (B7 550 for one sample and D11 575 for the other sample) (**Map 45 Gallinas 2008 DNA.jpg**).

Two samples collected in 2009 adjacent to those identified as hybrids at the southern end of Gallinas Creek in 2008 were both lab-identified as hybrids again in 2009. Analysis of eight microsatellite markers indicated high (>85%) proportions of *S. alterniflora* ancestry for both samples. The southern sample was also tested with RAPDs and was found to contain four of the five *S. alterniflora*-specific markers (B7 550/650, D5 600, D11 575, X18 450).

An additional eight samples were collected at the northern end of Gallinas Creek in 2009, seven of which were field-identified as *S. foliosa* and one of which was a resampling of a 2008 lab-identified hybrid. The 2008 and 2009 genetic results both indicated evidence of hybridity for the resampled patch. Of the seven samples from field-identified *S. foliosa*, only one sample had evidence of hybridity based on RAPD and microsatellite results. These were based on the presence of a single *S. alterniflora*-specific RAPD marker (X18 450) and inference of 76% *S. alterniflora* ancestry based on the analysis of eight microsatellite markers. Upon weighing the genetic evidence against the morphological evidence, the ISP biologist who collected this

sample identified the patch from which it came as *S. foliosa* and noted this location for additional future genetic testing. (**Map 46 Gallinas 2009 DNA.jpg**)

Wildcat Marsh

Concerns arose over the possible hybridity of plants that appeared to be *S. foliosa* when 2007 RAPD results indicated hybridity of a plant identified as *S. foliosa* with high confidence in the field. Additional samples were collected in 2008, and in 2009 even more samples were collected with the hope of relating lab results to morphological characteristics of collected samples. The pattern that emerged was one of conflicting evidence of hybridity based on morphological characteristics, RAPD results and microsatellite results (**Map 31 San Pablo Wildcat Results 2008 & 47 Wildcat Results 2009.jpg**).

Goodman's Lumber

Following up on reports that the *Spartina* planted at Crissy Field and Stege Marsh (in the 1990s) was collected from the small, remnant marsh behind Goodman's Lumber in Mill Valley (775 Redwood Hwy), the ISP conducted genetic sampling at this marsh in 2008. Although the majority of the *Spartina* in this marsh appears to be *S. foliosa* based on other morphological characteristics, many of the *Spartina* stems in this marsh contain a pink to reddish tinge, generally considered characteristic of *S. alterniflora x foliosa* hybrids (Callaway and Josselyn 1992, Ayres et al. 2004a). The results of these genetic tests are presented in **Maps 48-49 Goodmans Lumber DNA 2008-2009.jpg**.

Strawberry Point School

Genetic sampling was conducted in the lagoon behind Mill Valley's Strawberry Point School on June 30, 2008 in response to concerns by local biologists involved in the restoration of this marsh. The five samples collected within the lagoon were tested with RAPDs and were found to contain no evidence of hybridity based on seven species-specific RAPD markers (**Map 50 Strawberry Pt School DNA 2008.jpg**). In follow-up email to the ISP, one biologist expressed concerns that the *Spartina* growing in the interior tidal pond/pan is accreting sediment, "which is converting the pond into a pan with black, sulfidic, anoxic mud." He noted that, "the 'ambiguous' *Spartina* in the anoxic mud at the pan edge has wide culm bases and rigid, wide leaf blades (green late in fall) and persistent attached…leaf litter. RAPD test results aside, this doesn't strike me as ambiguous for hybrid morphology or ecology." (P. Baye, personal communication 12/19/08) Based on rapid colonization of the tidal channel, also noted by these biologists, plants were treated at the mouth of this channel, at the northwestern end of the footbridge (see 2005-09 ISP Treatment Report).

ISP biologists subsequently collected two samples within the tidal pond/pan area in 2009 (**Map 51 Strawberry Pt School DNA 2009.jgp**). Analysis of eight microsatellite markers inferred one of these to be of 97% *S. foliosa* ancestry; no RAPD testing was performed on this sample. The sample collected in the northwest corner of the interior marsh, had evidence of hybridity based on lack of one *S. foliosa*-specific RAPD marker (A2 575), presence of one *S. alterniflora*-specific RAPD marker (X18 950A), and analysis of eight microsatellite markers indicating low levels (54%) *S. alterniflora* ancestry.

Samples collected along the edge of the channel due east of the Strawberry Point School marsh levee contained evidence of hybridity based on RAPD testing in 2008 and 2009. The 2008 result was based on the presence of a single *S. alterniflora*-specific RAPD marker (D5 600). A sample collected in 2009 at this same location also contained evidence of hybridity, but was based on the presence of a different single *S. alterniflora*-specific RAPD marker (B7 550) and analysis of eight microsatellite markers which inferred 92% *S. alterniflora* ancestry.

The 2008 results informed the monitoring program biologist who surveyed this eastern channel to initially designate as hybrid the *Spartina* within a 100 x 1.5 meter linear area along the channel edge. As with the Calaveras and Greco Island North examples above, this line was considered for editing, to be shortened with the lower portion changed to a final species designation of *S. foliosa* based on subsequent years of field observations at this site. However, two samples taken along this linear patch in 2009 led to further confusion; these two samples are designated as "grey area" SSR results in **Map 51 Strawberry Pt School DNA 2009.jgp**. The southern of these two samples was inferred to contain a small (46%) proportion *S.*

alterniflora ancestry based on analysis of seven microsatellite markers. The northern of these two samples had evidence of hybridity based on presence of a single *S. alterniflora*-specific RAPD marker (X18 450) and based on analysis of seven microsatellite markers which inferred 52% *S. alterniflora* ancestry. This linear patch was deemed to be *S. foliosa* at time of treatment and thus was not treated.

Crissy Field

Genetic results at Golden Gate Recreation Area's Crissy Field have shown some evidence of hybridity in plants that appear to be native. Fourteen samples were collected from Crissy Field for RAPD testing in 2008, all of which were field-identified as *S. foliosa* with moderate confidence. Of these, RAPD results for three samples on the south side of the lagoon indicated hybridity based on the presence of all *S. foliosa* markers tested plus the presence of one *S. alterniflora*-specific marker each (B7 550 for one sample, C10 470 for the two other samples).

Despite past evidence of hybridity in Crissy Field, these 2008 results were not expected. (See **Maps 52 – 57 Crissy Field 2003-8 DNA.jpgs**.) Several seedlings had been identified as hybrids in 2003 based on RAPD results, but these had been pulled and disposed of, and were located in a different portion of the marsh than the patches with evidence of hybridity based on 2008 RAPDs. Samples had been collected in the vicinity of these 2008 "hybrids" in 2006 and 2007, and RAPD results in both years indicated that these patches were native. None of the plants identified by RAPDs as hybrids were morphologically distinguishable from adjacent plants identified as *S. foliosa* in 2008. Testing in 2009 found no evidence of hybridity from any of these three locations, based on RAPD and microsatellite testing.

However, microsatellite results and morphology-based field identifications indicated presence of more hybrid plants than previously mapped in the southeastern quadrant of Crissy Field in 2009. These patches are within 200 meters of a field-identified and genetically confirmed hybrid plant first identified in 2005 and mapped again in 2009. (See **Map 58 Crissy Field 2009 DNA.jgp**.)

The ISP Monitoring Program will continue to monitor this site closely in future years and the ISP Control Program will continue to communicate with GGNRA regarding any control measures to be taken at this site.

Greco Island North

Greco Island North was mapped by helicopter on August 30, 2008. In a situation similar to that at Calaveras Point, a much greater area was mapped as *S. alterniflora x foliosa* than in past or future years. Of the features mapped at this site, two long lines and one large polygon comprising 2.5 net acres occurred in areas where future, ground-based mapping efforts found no hybrids. These features were changed to a final species determination of *S. foliosa*, resulting in a decrease in the estimated 2008 net acres at this site from 12.5 to 10 acres.

Faber and Laumeister Marshes

Prior to 2008, *Spartina* at Faber and Laumeister Marshes was believed to be native, with the exception of one single patch of hybrid 10-12 meters in diameter in each marsh. These two patches were discovered during the 2005 survey of the perimeter of these marshes. No new patches were detected in 2006 or 2007. All surveys were conducted from the perimeter of the marshes and no DNA samples were collected at Laumeister Marsh until 2008.

In 2008, an ISP biologist who had walked extensively throughout both Faber and Laumeister Marshes while conducting USGS clapper rail telemetry studies raised concerns about the potential hybridity of a number of patches in the marshes. In response, samples from Faber and Laumeister were tested for hybridity in 2008 and 2009 (**Maps 59-62 Faber and Laumeister 2008-2009 DNA.jpg**).

Of the 44 samples collected in 2008, 15 were found to contain *S. alterniflora* RAPD markers. Of these 15, only one sample, from the northeastern shoreline of Faber was missing any *S. foliosa* markers. This same sample, which was missing two *S. foliosa* markers, also contained all four of the *S. alterniflora* markers, and thus had extensive evidence of being a hybrid plant based on RAPD markers. (It is notable that this plant

was growing sparsely on the mudflat and was identified as a hybrid with low confidence based on morphology.) The other 14 samples identified as hybrids contained all *S. foliosa* markers for which they were tested, and nine samples had only a single *S. alterniflora* marker present (**Map 60 Laumeister 2008 DNA.jpg**). Samples with only one of the four possible *S. alterniflora* RAPD markers are presumed to be hybrids, but have less evidence of hybridity than those plants with more *S. alterniflora* markers and/or missing *S. foliosa* markers.

In 2009, twenty-four samples were collected from Laumeister Marsh for microsatellite testing. Of these, six samples were also tested by Debra Ayres using RAPDs.

Microsatellite and RAPD tests revealed different results for two of the six samples (**Map 62 Laumeister 2009 DNA.jpg**): RAPDs indicated hybridity; microsatellite *structure* analysis estimated 97% *S. foliosa* ancestry. Of these, one sample was collected along a channel in the southwest corner of the marsh and was field-identified as a hybrid with high confidence; results indicated presence of all *S. alterniflora* and all *S. foliosa* RAPD markers for this sample. The other sample was field-identified as "unknown"; RAPDs indicated hybridity based on the presence of *S. alterniflora* marker X18 450A in addition to all *S. foliosa* markers. Two samples fell into the "grey area" of structure results (inferred to contain between 25-75% *S. alterniflora* ancestry). Of these, one sample was identified by RAPDs as a possible *S. alterniflora* (based on one "sketchy call" on the presence of *S. alterniflora* marker X18 950A) and had a 64% chance of being of *S. alterniflora* ancestry according to *structure*. The other was identified by RAPDs as *S. foliosa*, with *structure* results estimating a 77% proportion of *S. foliosa* ancestry.

Two samples were identified as hybrids by both microsatellite and RAPD results. One was mapped as a low confidence native and the other as an unknown when collected in the field in November. Field identification was based on morphology and location, and neither of these two samples had inflorescences to assist with identification.

Calaveras Point

In 2008, the ISP biologist mapping Calaveras Point mapped significantly greater amounts of hybrid mixed in with *S. foliosa* than in previous or subsequent years. In previous years, monitoring at this site was conducted by boat by SCVWD in conjunction with ISP biologists, primarily by boat but with some walking in the marsh. Interior portions of the marsh were largely inaccessible by boat, and the majority of patches mapped and treated were hybrids of the tall variety which were visible from channels accessed by boat. Prior to 2008, monitoring was conducted between late October and late December at Calaveras Point.

Monitoring was conducted by helicopter on July 30, 2008 with some follow-up monitoring on August 31, 2008. All *Spartina* in the South Bay is notably taller and more robust than *Spartina* in the Central and North Bay, making differentiation between native and hybrid *Spartina* more difficult in the South Bay due to the robust nature of the South Bay native. Inflorescence length and width is extremely helpful for differentiating the hybrid from the native in such areas. In July, South Bay populations are not yet flowering, so there were no inflorescences to assist in identification of hybrids. The southern quarter of the marsh was identified by the biologist as an extensive hybrid swarm, based primarily on variability in heights of stands in the marsh, and on stem and leaf width.

Based on this original helicopter-based mapping, approximately 16 net acres of *Spartina* spread out over 93 acres within Calaveras Point were identified as hybrids. Of these, approximately 12 net acres, spread out over 45 acres, were changed to a final determination of *S. foliosa* after two subsequent years of on-the-ground monitoring during flowering season resulted in no discernable hybrid plants found in these areas. As a result of this change in final determination, the acreage at this site was reduced to approximately 4 net acres of hybrid *Spartina* spread out over 48 acres within Calaveras Point.

(Please note that Calaveras Point is within subarea 5a: Mowry and Calaveras Marshes; thus the acreage reported for this larger subarea in **Tables 7 and 9** includes and is greater than the values described above which are for only Calaveras Point.)

DISCUSSION

Improved Detection Methods

As described in the inventory monitoring background section above, monitoring methods were adapted and improved in 2008 to address difficulties in detection of invasive *Spartina*. Most notable were (1) the transition from digitizing aerial imagery to conducting all surveys in the field by land, water or air (helicopter), and (2) the transition to use of ArcPad software to allow navigation to and query of GIS data in the field.

The ISP hired four additional field biologists in 2008 to assist with the increased on-the-ground efforts required. The additional staff allowed survey to be conducted using methods that allowed for greater thoroughness. Examples of more time-intensive but thorough survey methods include conducting walking surveys rather than boat surveys along shorelines, and conducting kayak surveys so as to penetrate small channels and shallow areas inaccessible by motor boat. The increase in thoroughness has paid off with the discovery of new populations and newly discovered potential habitats (such as side channels and ditches not previously noticed). The more detailed mapping of *Spartina* also assisted the ISP Control Program, as discussed below.

Monitoring became much more challenging in 2007 as a result of the decline in *Spartina* populations following effective, regional treatment in 2006. As the density, height and vigor of treated patches declined, the remaining live biomass of invasive *Spartina* within treated patches became much more difficult to detect both on-the-ground and, most notably, in aerial imagery.

While the increase in net *Spartina* mapped between 2007 and 2008 is largely artificial due to the combination of improved methods of detection used in 2008, increases in sites that were not digitized may be accurate due to new findings and spread of invasive plants. Additionally, ISP field biologists surveyed the entire Southern South Bay area for the first time in 2008. In prior years, much of the Southern South Bay area was mapped exclusively by Santa Clara Valley Water District, with data provided to the ISP for inclusion in our datasets. The expanded populations of invasive *Spartina* mapped by the ISP in this region in 2008 may be due to the greater specialization of ISP biologists in the detection of hybrid *Spartina*.

Dramatic Reduction Since 2005

At the height of the infestation, in 2005, the ISP mapped 809 net acres (327 hectares) of invasive *Spartina* throughout the Bay and outer coast areas. By 2008, net area was down to 274 acres (111 hectares) and by 2009 only 158 net acres (63 net hectares) remained (**Table 1 Baywide Acres 01to09**).

These survey results showing dramatic declines in Baywide net acreage of invasive *Spartina* following the 2005 field season confirm our subjective observations that invasive *Spartina* populations declined significantly since the ISP began using the highly effective herbicide imazapyr at most sites. Photo monitoring (described in Part II of this report) has also documented this visible decline (for example, see **Figures 2.2 through 2.8**). The reduction in net acres in 2006 was a result of use of the highly effective herbicide imazapyr, which was used on most populations for the first time in 2005. Follow-up treatment in future years was less dramatic but no less important, and both treatment and mapping became more difficult with each year of successful treatment.

The notable decline in *Spartina* as a result of treatment is especially impressive given observations of the continued expansion of new patches even while treatment reduced overall cover. Field observations in 2008 indicated that while there was observable decline in *Spartina* density and cover in treated patches of mature *Spartina*, the late timing of treatment at most sites allowed invasive *Spartina* to successfully set and drop seeds. These seeds, and vegetative propagules from remaining live hybrid plants, were able to spread and form new patches and populations.

Artificially High/Low Cover

Within mapped features, detectability and percent cover estimates can vary depending on the bias of the observer, month and/or mapping method. When these variations are consistent they can lead to artificially low or high estimates of invasive *Spartina* acreage by year within a site. While we take efforts to reduce all three sources of error, we are aware of consistently low cover bias in 2007 digitization efforts at low density sites and consistently high cover bias in 2008 helicopter monitoring at high density sites. We are also aware of issues of detectability related to plant phenology.

In 2007, all sites mapped via digitizing of aerial imagery can be assumed to have artificially low invasive *Spartina* cover estimates. This is because visual detection of live *Spartina* regrowth within treated stands with low cover was difficult to impossible even using very high (16 cm) resolution, digital aerial imagery acquired in 2007. Use of aerial imagery for accurately identifying vegetation and estimating cover class is known to be especially difficult for low or high density vegetation (van Klinken et al. 2007, Andujar et al. 2010). Ground truthing was conducted to assist with digitizing efforts, which helped to alert staff to the difficulty of identifying low density *Spartina* via aerial imagery but did not mitigate the difficulty of the task.

In 2008 and, to a lesser extent in 2009, sites with high densities of invasive *Spartina* that were mapped via helicopter monitoring are suspect for having artificially high invasive *Spartina* cover estimates. This is because helicopter monitoring was a new and unfamiliar method for the project in 2007, and was generally used for sites with which staff were relatively unfamiliar (as they were primarily sites that were formerly digitized). Also, while helicopter monitoring is very good for detecting new populations, consistent estimation of weed cover by helicopter is known to difficult (van Klinken et al. 2007).

The phenology of plants also led to consistent bias in acreage at some sites. Those sites mapped prior to flowering may be under-mapped due to a combination of low cover bias (plants are smaller) and difficulty in identification of hybrid individuals that are not flowering. This latter bias due to difficulty in identification extends to those plants that remain vegetative (no sexual reproduction), such as can happen with regrowth of stems within treated patches.

For on-the-ground mapping, field biologists train for consistency in cover estimation at the beginning of the season and periodically throughout the season. During such trainings, and based on occasional duplicate mapping of features, we estimate that cover estimates typically vary by up to one or two cover classes between observers. Observer calibration serves to reduce this error as much as possible, and any errors in accuracy are expected to be relatively consistent from year-to-year due to this annual training and due to the low turnover of field staff at the ISP.

Identification Increasingly Difficult

Changes in confidence levels of *Spartina* field identification over time reflect the increasing difficulty of differentiating hybrid seedlings and hybrid regrowth weakened by treatment from native *S. foliosa*, which is typically less robust and vigorous than hybrids. Differentiation between colonization of adjacent, native cordgrass versus regrowth of weakened hybrid within a treated patch is particularly challenging.

Identification of hybrids may also be becoming more difficult as control of hybrids with distinct morphologies (unlike those found in *S. foliosa*) leads to the artificial selection of those hybrids with less distinct morphologies. Even in the absence of such selective pressure, the diminished presence of "obvious" hybrids after successful treatment can lead to greater awareness of and initiative to identify hybrid *Spartina* with less obvious hybrid morphologies.

As a result of these challenges, the ISP began collecting many more genetic samples in 2006 than in past years. As numbers of samples collected increased, number of samples with lab identifications that did not match field identifications also increased (**Figures 1.8** and **1.9**). Despite increases in the number of samples with genetic evidence of hybridity that had been identified as *S. foliosa* in the field, the proportion of such samples never rose over 23%. In 2009, of those samples identified as *S. foliosa* in the field, 60% of those tested with RAPDs and 42% of those tested with microsatellites were found to have evidence of hybridity.

Regardless of whether this increase in proportion of morphologically "cryptic hybrids" in 2009 is real or is simply an artifact of changes in genetic testing techniques in 2009, the challenge of distinguishing morphologically indistinct hybrids from pure native *S. foliosa* is undeniable.

Detection Increasingly Difficult

As vast meadows of invasive *Spartina* are reduced to small patches or even single stems of regrowth within treated areas, detection becomes increasingly difficult and time-consuming. Standing dead plant material often further complicates detection, as live stems can be difficult to see under such cover. Regrowth is often stunted and does not flower, making it even more difficult to detect or identify.

As a result of these challenges, the levels of effort and expertise required to detect and identify patches of invasive *Spartina* has increased considerably as the size and density of invasive *Spartina* patches has decreased.

Increased Evidence of Hybridity Troubling

The significant increase in genetic evidence of hybridity in 2009 for those samples field-identified as *S. foliosa* (**Figure 1.8**) was not easily explained. A number of theories arose, including the potential for errors due to using a new lab using its own proprietary DNA extraction protocol, potential reduction in the quality of the extracted DNA tested with RAPDs at UC Davis after testing in and shipment from Colorado, or potential problems with the new genetic methodologies employed in 2009 (microsatellites and additional RAPD primers).

The increased evidence of hybridity of samples believed to be native could of course reflect a true increase in genetically detectable hybridity in plants without obvious hybrid morphologies. Such results could be attributable to poor field identification, increased sampling of hybrids during 2009 sampling efforts, or increased ability of 2009 techniques (microsatellites and additional RAPD primers) to detect hybridity.

Because of the low turnover in staff at the ISP between 2007 and 2009, a decrease in ability to accurately identify *S. foliosa* due to reduced sensitivity of ISP biologists is not likely. (In fact, with increasing experience, one would expect an increase in ability to accurately identify *S. foliosa*.) In 2007, ISP biologists tended to be perhaps overly sensitive to possible hybridity in collected samples, as seen in large numbers of field-identified hybrid results with no RAPD-based evidence of hybridity that year (**Figure 1.9**). The ISP hired several additional biologists in 2008, and hired no new staff in 2009. While it is possible that experienced ISP biologists may have tended to be less sensitive to morphological evidence of hybridity in 2009 than in past years, it is more likely that the samples collected in 2009 were simply less visibly discernable as hybrids in 2009.

It is also entirely likely that the increased evidence of hybridity in field-identified *S. foliosa* samples is an artifact of the new genetic techniques employed in 2009, and may not be directly comparable to past years' data. The reliability of the genetic techniques used in 2009 are being explored by the ISP in consultation with the UC Davis Spartina Lab and plant geneticist Dr. Jack.

Improved Collaboration with Control Program

Improved efficiencies in mapping techniques and collection of additional information by the Monitoring Program have allowed for greater collaboration between the Control and the Monitoring Programs during the treatment season. The invention of the concept of "treatment cover" (see box *Recording *Spartina* Area*) and collection of this attribute beginning in 2008 led to summary data of much greater practical use to the Control Program than summary of net area had been. This new treatment area value was recorded so as to assist with Control Program estimations of required herbicide and treatment effort at a site, and was the result of discussions with and training by the ISP field operations managers.

Greater collaboration during the field season has improved the ability of the ISP to quickly respond to new infestations, and has allowed the Control Program to use current-year monitoring data to inform treatment at many locations.

The treatment surveys conducted on a trial basis in 2009 proved extremely valuable at the sites where they were conducted. These surveys allowed treatment crews to efficiently plan for and navigate to locations for treatment, and allowed monitoring staff to map additional patches of invasive *Spartina* noticed during treatment. Because of the increasing difficulty in distinguishing hybrid from native *Spartina*, collaboration in the field, combining the expertise of both ISP biologists and treatment crews, has proven helpful to both parties.

Acres Mapped Versus Acres Treated

Because inventory mapping typically does not take place during treatment events, estimates of acres requiring treatment as calculated by the Monitoring Program data do not necessarily represent the acreage actually treated or deemed to require treatment by the ISP Control Program or partners performing treatment. (For information on acres actually treated, see the ISP Control Program 2008 and 2009 Reports.) Discrepancies can result from the following scenarios.

(1) Inventory mapping may take place significantly earlier in the year than treatment. Identification of hybrid plants can be more difficult earlier in the season (prior to flowering). Thus, monitoring staff may under-map an area. In such a circumstance, treatment crews will likely notice and treat more invasive *Spartina* than was mapped by the Monitoring Program.

(2) The difficulty of identifying invasive *Spartina* with less obvious hybrid morphologies can lead to differences in those patches identified as invasive *Spartina* and treated by partners and their contractors versus those patches mapped as invasive *Spartina* by the ISP Monitoring Program. Even in situations where there are no concerns of hybrid identification, differences in the ability to accurately detect, identify and treat or map plants can differ among individuals. It is thus not surprising if differences arise in this situation, where differentiation between native and hybrid plants adds additional challenge to consistency in patch evaluation between individuals.

(3) Inventory mapping may be conducted by field staff unfamiliar with the morphology of *Spartina foliosa* at a particular site, resulting in the misidentification of *Spartina* species at that site. This can lead to either the over- or under-mapping of invasive *Spartina* at a site. Although steps are taken to avoid this situation, including the checkout of past years' data onto GPS units for each site beginning in 2008 and extensive training and pairing of new staff with experienced staff, this situation can happen, especially at difficult sites (such as those in the Southern South Bay where *S. foliosa* is often extremely robust).

Treatment surveys, conducted as a pilot project in 2009, helped to reduce discrepancies between mapped versus treated acres by better integrating monitoring and treatment efforts.

CONCLUSIONS

Monitoring Surveys

Improvements to the efficiency of data collection and processing has made monitoring survey information more accessible to the Control Program. The increased difficulty in detection of hybrid plants has made monitoring more time-consuming and difficult than in past years. The Monitoring Program will continue to use and improve customizations to the software used on GPS units to bring all past years' information into the field so as to inform inventory monitoring navigation and decision-making.

ISP biologists will continue to search diligently for previously undetected potential habitat, both in the field and using aerial imagery in the office. Prior to the initiation of future monitoring seasons, a comprehensive effort to update and identify new potential habitat boundaries will be undertaken using high resolution aerial imagery in a GIS.

Treatment Surveys

Treatment surveys proved valuable to both the Monitoring and Control Programs and will be expanded in the future. Additional ISP staff will be hired to conduct treatment surveys. The cost of the required increase in monitoring staff should ultimately be offset in the long term by the increased thoroughness of treatment activities facilitated by this integrated approach to monitoring and treatment. More thorough annual treatment effort at a site should lead to a reduction in the time to eradication of invasive *Spartina* at that site.

Discernment of Hybrids

The ISP continues to grapple with the difficulties of discerning hybrid from native *Spartina* both in the field and with the evidence provided by genetic testing using RAPD or microsatellite markers.

Through review of scientific literature, participation in relevant workshops and conferences, and consultations with plant geneticists and others, ISP managers are increasing their awareness of some of the potential shortcomings of currently available genetic testing methods for conducting purity assessments of *Spartina*, the most notable of which is the number of markers available. Up to six *S. alterniflora*-specific and five *S. foliosa*-specific RAPD markers have been used to date on samples collected by the ISP. Ten microsatellite markers were used in 2009, eight of which were ultimately analyzed in 2009. It is now our understand that, for hexaploid species whose hybrids have undergone multiple generations of backcrossing and introgression, the evidence provided by this limited number markers may be insufficient to accurately distinguish complex hybrids from natives (Boecklen and Howard 1997).

The ISP has determined that use of microsatellite testing to help distinguish native from hybrid individuals is more practical than the continued use of RAPD methods. The UC Davis Spartina Lab is the only lab that has successfully tested *Spartina* samples using RAPD techniques. The UC Davis Spartina Lab had difficulties processing the large volume of samples (>1000) sent in recent years, however, and is designed for research, not for large throughput like a commercial lab. The ISP was able to locate only one commercial lab (STA Labs in Colorado) willing to perform RAPD testing in 2009, and this lab was unable to successfully perform these tests after months of attempts. The ISP was able to locate and obtain competitive bids from several commercial labs able to perform microsatellite testing, and scientific literature suggests that results are much more reproducible between labs for microsatellite than RAPD testing (Jones et al. 1997).

The ISP will expand the number of microsatellite markers analyzed and will collect many more putatively pure *S. foliosa* samples and "obvious" field-identified hybrid individuals to assist with the analysis of microsatellite results in the future.

The ISP will continue to endeavor to understand and use all available, affordable and practical information to assist in the discernment between hybrid and native individuals.

PART II: EFFICACY MONITORING

PHOTO POINT MONITORING

Background

Photos were taken at 146 permanent photo point locations within 21 control program areas starting in 2006 to assess and document efficacy twice per season. Sites for photo point monitoring were selected to encompass a range of marsh types and treatment methods, and were located within 70 control program subareas (**Figure 2.1**). Photos were taken early enough in the spring to allow the Control Program to assess the upcoming effort required at each site, and were taken again late enough in the summer to assess initial impact of current-year control efforts.

Methods

Photo point monitoring took place beginning in late May following the protocol described in the QAD. Photos were taken at all photo point locations, with exact points relocated using GPS and compass directions, and with print-outs of past year's photos used to ensure the correct angle and horizon level in the camera view-frame. Two images were photographed at each location. Final images were selected and cropped as necessary in the office to allow for the best match of horizon and view extent so as to allow for ease in comparability between years.

Photo point images were shared with ISP Control Program Field Operations Managers within several days of data acquisition and used to inform field operations planning on an as-needed basis.

Results

Photo point monitoring was used by the ISP Control Program to quickly assess and plan for the upcoming treatment season. Photo monitoring was completed at 57 sites, with a total of 136 photos taken at each of two rounds. Digital photos are saved to the ISP server, where they are organized by photo point to allow ease of viewing change over time using photo-viewing software.

Examples of photos from 2006 through 2009, and maps illustrating 2009 *Spartina* inventory data overlaid with photo point locations are presented for six locations in this report. These locations were selected to show a diversity of sites (**Figure 2.1**) and a diversity of treatment efficacy as evidenced by the photos. These include Subarea 2g: Bunker Marsh (**Figure 2.2**), Subarea 8: Palo Alto Baylands (**Figure 2.3**), Subarea 13e: Whale's Tail South Fluke (**Figure 2.4**), Subarea 17d: MLK Regional Shoreline (**Figure 2.5**), Subarea 18a: Colma Creek (**Figure 2.6**), Subarea 19h: SFO (**Figure 2.7**), and Subarea 19l: Burlingame Lagoon (**Figure 2.8**). Note: Corresponding net area, treatment area and treatment efficacy for these subareas can be looked up in **Tables 7, 8** and **9**.

Discussion

The information on site-specific efficacy collected through photo point monitoring was used by the ISP Control Program to plan for field operations. This photo monitoring effort is also strengthening the documentation of observed efficacy of *Spartina* treatment efforts throughout the Bay, and is documenting the resulting restoration of marsh vegetation or open mudflat at many sites.

Conclusion

Photo point monitoring has proven to be an efficient and effective method of monitoring and capturing information regarding the results of treatment, including passive restoration of tidal marsh and mudflat. Photo point monitoring will be continued into the future by the ISP Monitoring Program.

PERMANENT PLOT MONITORING

Background

Permanent plot monitoring assesses treatment efficacy through the annual sampling of permanent monitoring plots which were established and sampled prior to the first year of treatment.

Monitoring plots were initiated in 2004 at the subareas treated in 2003 and at subareas slated for treatment in 2004, to provide baseline information for the evaluation of future treatment actions. These plots and additional plots were monitored until 2008.

Sites for monitoring were selected to encompass a range of marsh types and treatment methods.

Field Methods

Monitoring of permanent plots was conducted at 22 subareas in 2004, 44 subareas in 2005, and 56 subareas in 2006, 2007 and 2008.

At each sampled subarea, percent cover of all vegetation and density of invasive *Spartina* was recorded at up to 30 plots and a central quadrat with each plot. The methodology for sampling quadrat data has not changed since plot establishment. The methodology of sampling plot data changed between 2005 and 2006 due to the high efficacy in treating invasive *Spartina*. Prior to 2006, treatment plot size was based on the size of the *Spartina* clone within which the permanent quadrat was located. In 2006, contiguous clones of *Spartina* were no longer identifiable due to the impact of treatment, and so plot sizes could no longer be based on clone size. The methods for collecting treatment efficacy data are fully described in the QAD. As described in the QAD, plot size was standardized to a 3 m radius circle centered on the permanent quadrat starting in 2006.

Analysis Methods

Data from the treatment monitoring plots was analyzed to determine the effects of treatment and imazapyr application method on *Spartina* stem counts and plant biodiversity within the permanent plots. For each year that each permanent plot was monitored, a Shannon-Weaver index of plant biodiversity was calculated. Mean *Spartina* stem counts and Shannon-Weaver diversity indices for each year that each site was monitored were used in analysis rather than the individual plot data in order to avoid pseudoreplication. The mean *Spartina* stem counts were natural log transformed as this transformation yielded the regression models distributions of residuals that most closely approximate the normal distribution. Permanent plot data for each site and year was then combined with a table of data containing the cumulative number of years each site has been treated, the cumulative number of years each site has been treated with imazapyr, the cumulative number of years each site has been aerially treated with imazapyr, and the cumulative number of years the site has been treated from the ground with imazapyr (including by backpack, amphibious vehicle, truck, and airboat)

To analyze the general effect of treatment of *S. alterniflora x foliosa* and *S. densiflora* on plant biodiversity, a standard least squares regression model was created using Shannon-Weaver index as the response variable and cumulative years of treatment and site as the model effects. The resulting model was based on a linear equation: Shannon-Weaver Diversity Index = (constant intercept) + (constant slope) * (number of years treated) + (a variable that is held constant for each site). The last of these terms acts as a correction factor to take into account natural variation in diversity between sites. To examine the effects of treatment on *S. alterniflora x foliosa* and *S. densiflora* stem counts, similar least squares regression models were created for these subsets of the permanent plot data using natural log of stem count as the response variable and cumulative years of treatment as the model effects. In order to compare the efficacy of aerial *S. alterniflora x foliosa* imazapyr treatment and ground-based *S. alterniflora x foliosa* imazapyr treatment, only those sites which had received aerial imazapyr treatment but no ground-based imazapyr treatment or the sites which had received ground-based imazapyr treatment but no aerial imazapyr treatment were included in the analysis. Least squares regression models were created using natural log of *Spartina* stem counts as the response variable

and cumulative years of either aerial imazapyr treatment or ground-based imazapyr treatment as the model effects. Similar least squares regression models were then created using Shannon-Weaver diversity index as the response variable and cumulative years of either aerial imazapyr treatment or ground-based imazapyr treatment as the model effects. Sample sizes were too low to examine effects of various treatment methods of *S. densiflora* on stem counts and diversity.

Results

The regression model examining the effect of cumulative years of treatment of *S. alterniflora x foliosa* on stem count found a negative correlation between cumulative years of treatment and stem counts (slope=-0.69, SE=0.06, $p<0.0001$, n=162) with an R square of 0.62 for the entire model (**Figure 2.9**). The model examining the effect of cumulative years of treatment on Shannon-Weaver diversity index found a positive correlation between number of years of treatment and diversity (slope=0.22, SE=0.03, $p<0.0001$, n=162) with an R square of 0.71 for the entire model (**Figure 2.10**).

The regression model examining the effect of cumulative years of treatment of *S. densiflora* on *Spartina* stem count found a negative correlation between cumulative years of treatment and stem count (slope=-0.75, SE=0.12, $p<0.0001$, n=30) with an R square of 0.81 for the entire model (**Figure 2.11**). The model examining the effect of cumulative years of treatment of *S. densiflora* on Shannon-Weaver diversity index found a positive correlation between cumulative years of treatment and diversity (slope=0.32, SE=0.07, $p=0.0002$, n=29) with an R square of 0.54 for the entire model (**Figure 2.12**).

The regression model examining the effect of aerial treatment of *S. alterniflora x foliosa* on *Spartina* stem count found a negative correlation between cumulative years of aerial treatment and stem count (slope=-0.73, SE=0.23, $p=0.0046$, n=37) with a R square of 0.55 for the entire model (**Figure 2.13**). The regression model examining the effect of ground-based treatment of *S. alterniflora x foliosa* on *Spartina* stem count found negative correlation between cumulative years of ground-based treatment and *Spartina* stem count (slope=-0.93, SE=0.175, $p<0.0001$, n=42) with a R square of 0.64 for the entire model (**Figure 2.14**).

The regression model examining the effect of aerial treatment of *S. alterniflora x foliosa* on diversity did not find a significant correlation between cumulative years of aerial treatment and Shannon-Weaver diversity index. The regression model examining the effect of ground-based imazapyr treatment of *S. alterniflora x foliosa* on diversity found a positive correlation between cumulative years of ground-based treatment and Shannon-Weaver diversity index (slope=0.32, SE=0.09, $p=0.0016$, n=42) with an R square of 0.82 for the entire model (**Figure 2.15**).

Discussion

Permanent plot monitoring showed evidence that treatment of *S. alterniflora x foliosa* and *S. densiflora* resulted in a significant decrease in *Spartina* stem counts and a significant increase in Shannon-Weaver diversity. Increases in plant biodiversity with cumulative years of treatment are thought to be due to release of other marsh plants from competition with invasive *Spartina*.

While the permanent plot monitoring suggests that aerial and ground-based treatment had approximately equal efficacy in reducing *Spartina* stem counts, these methods differed in their effects on diversity. Ground-based imazapyr treatment was shown to have a strong positive effect on plant biodiversity, but aerial imazapyr treatment had no significant effect on plant biodiversity. There are two possible explanations for this difference in the effects of different treatment methods. The first possibility is that sites chosen for aerial treatment had some barrier to recovery of plant biodiversity that is not present in sites chosen for ground-based treatment. This barrier could involve a variety of factors including recruitment limitation, degree of *Spartina* infestation, existing biodiversity, and marsh hydrology. The second possibility is that the greater precision with which ground-based treatment may target invasive *Spartina* allows treatment crews to avoid incidental treatment of non-target plant species. This would cause there to be a larger number of plant species left in closer proximity to *Spartina* treatment areas, allowing for a more rapid increase in biodiversity.

These two possibilities are not mutually exclusive and it is very likely that a variety of factors affect the correlation between diversity and cumulative years of imazapyr treatment.

Conclusion

Permanent plot monitoring documented the efficacy of imazapyr on reduction of invasive *Spartina* density and also documented an increase in plant diversity within imazapyr-treated plots over time.

When permanent plot monitoring was initiated in 2004, expected efficacy of treatment was an unknown. Permanent plot monitoring was originally designed as an attempt to detect even subtle, sublethal effects of treatment, and was funded through 2008. Due to the dramatic success of imazapyr in reducing populations of invasive *Spartina,* efficacy was equally discernable through annual inventory monitoring as discussed in Part I and through analysis of stem reduction in monitoring plots as discussed in Part II. Continuation of permanent plot monitoring was deemed unnecessary and further funding for such efforts was not pursued.

Permanent plot monitoring and photo point monitoring are the only methods used by the ISP to document marsh recovery following treatment, with permanent plot monitoring yielding statistically sound data documenting this recovery. As noted in the analysis above, biodiversity within plots increased with cumulative years of ground-based imazapyr treatment, reflecting the recovery of tidal marsh vegetation concurrent with removal of invasive *Spartina.*

These results indicate that any non-target effects of imazapyr on adjacent vegetation are insignificant to overall recovery of tidal marsh biodiversity, even in the short term. This documented increase in biodiversity concurrent with 2-3 years of annual treatment of invasive *Spartina* with imazapyr is very encouraging.

Follow-up surveys of permanent plots in future years would be valuable, as such efforts could document long-term changes in biodiversity observed in marshes following treatment of invasive *Spartina.*

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TABLES

Table 1. Estimated net acres baywide.

Spartina Species	Estimated Net Acres						
	2001	2004	2005	2006	2007	2008	2009
All invasive spartina	480	759	809	590	212	274	158
S. alterniflora x foliosa	472	756	804	585	208	272	156
S. densiflora	7.7	2.5	4.2	3.8	2.6	1.4	1.3
S. anglica	0.09	0.08	0.06	0.11	0.09	0.01	0.08
S. patens	0.54	0.55	0.65	0.21	0.05	0.03	0.02

Table 2. Estimated acres requiring treatment baywide.

Spartina Species	Estimated Acres Requiring Treatment		
	2008	2009	Change
All invasive spartina	431	322	25% decline
S. alterniflora x foliosa	427	318	26% decline
S. densiflora	2.84	2.27	20% decline
S. anglica	0.012	0.144	increase
S. patens	0.053	0.046	13% decline

Table 3. RAPD markers used.

Specific to Spartina species	Primer	Fragment	2008	2009
S. anglica	B10	750	x	
	C10	3100	x	
	C12	575	x	
	F10	900	x	
	G2	1050	x	
	H7	650	x	
	H7	750	x	
	H7	1500	x	
S. densiflora	B7	1200	x	
	B7	700	x	
	D5	550	x	
	D5	800	x	
S. foliosa &/or S. *densiflora*	B7	800	x	x
S. foliosa	A17	725	x	
	A2	575	x	x
	D5*	1100	x	x
	X18	750		x
S. alterniflora	B7	550/650	x	x
	C10	470	x	
	D11	575	x	x
	D5	650	x	x
	X9*	1000		x
	X18	450		x
	X18	950		x

* Markers D5 1100 and X9 1000 considered unreliable by UCD lab upon further testing.

Table 4. Primers for 2009 STA Labs SSR Analysis

Primer ID	*Forward sequence* (5'-3')	*Reverse sequence* (5'-3')	*S. foliosa alleles*	*Alleles occurring only in hybrids*	T_a* (℃)	$MgCl_2$ (mM)	$MgCl_2$ (µL)
Spar.02	GAAGGACGAGTCTCATTTGG	GGCTGCCCCTGTTTCACG	193	201, 213	56	2.50	0.20
Spar.08	CTAAGGTCCCAAACGACGAC	GCGACGAGCGAGGATTTAC	193	180, 188	58	2.50	0.20
Spar.09	GTGGCCTAGCCTATCGACCT	TGAATGGAAAGGGGAAATGA	279, 285, 294	273, 277, 292, 296	58	2.50	0.20
Spar.15	ATTTGCTGCTTTTGGTAGAC	GTAGAACAATGGAAGAATGC	266-268	266-285	51	2.83	0.39
Spar.20	ACCGTGCCTCAGCTACTG	GGTGTTTCCTCGCATAGATC	171	173, 175, 177, 179, 181	52	2.17	0.02
Spar.23	GGGAAGTGAAATCTGGTTGC	GCTTGCTTGTCTCAGTCC	262, 264, 266, 268	248, 250, 274	55	2.17	0.02
Spar 25	CGGTAGAGACGGAGTTGTGG	GCTTGGGAGATGAGACTGGAC	245, 249	253	69	3.00	0.48
Spar 26	TTCAACTGGCGTAGTGATTCC	AACATTTCCGACTGGTAGAGC	263	263-291	58	2.00	0.58
Spar 27	CATCAAAAGCAAGAGGA	GACACCAACGGAACTG	314	304-331	50	2.16	0.02

Table 5. Microsatellite results from internal control samples.

Species	From	Plate #	Spar 02	Spar 08	Spar 09	Spar 15	Spar 20	Spar 23	Spar 25	Spar 26	Spar 27	Spar 28
S. alterniflora	Maine	1										
		2	193-213	188-193	277-296	269-271	171-173	268-272	244-253	263-269	314-323	416-420
		3	193-196	188-193	285-292	269-271	173-181	262-274	245-253	263-281		
		4	193-206	185-193	285-292	269-271	171-177	264-274	245-253	263-275	314-323	416-476
		5	193-213	188-193	285-292	266-271	171-179			263	304-314	
		6	193-201	180-193	285-292	266-271	171-177	250-266		263-285		416-419
		7	193-201		285-292	266-271	171-173	264-274		266-277	314-304	416-419
		8	193-201	180-193	285-273	266-269	171-173	266-274			314-321	416-419
		9	193-201	180-193	285-292	266-269	171-175	264-277		263-268	304-314	416-419
		10	193-213	180-193	285-292	266-271	171-179	250-268		263-281	304-314	416-419
		11	193-201	188-193	277-285	266-271	171-173	262-250		263-277	304-314	416-419
		12			285-296	266-273						
S. alterniflora/hybrid	Tiburon	1										
		2	193-201	180-193	277-285	266-269	171-177	248-262	245-253	263-281	314-328	416-476
		3	193-201	180-193	277-285	266-269	171-177	248-262	245-253	263-281	304-314	416-420
		4	193-201	180-193	277-285	266-275	171-177	248-264	245-253	263-281	314-321	416-419
		5	193-201	180-193	277-285	266-269	171-173	248-266		263-283	304-314	416-419
		6				266-269	171-173	248-264		263-277	314-323	
		7	193-201	193-180	279-273	266-269	171-177	248-264		263-291	314-304	416-419
		8	193-201	180-193	277-279	266-271	171-177	248-264		263-285	314-321	416-419
		9	193-201	188-193	277-285	266-273	171-173	248-262		263-281	314-328	416-419
		10	193-213	180-193	277-279	266-269	171-177	248-262		263-281	305-314	416-419
		11	193-201	180-193	277-285	266-269	171-177	262-250		263-275	304-314	416
		12	193-201	180-193	277-285	266-271	171-177	248-264		263-275	314-323	416-419
S. foliosa	American Canyon	1										
		2	193	193	285	266	171	264	245-253	263	314	416
		3	193	193	285	266	171	264	245-253	263	314	416
		4	193	193	285	266-271	171	264	245-253	263	314	416
		5	193	193	285	266	171	266		263	314	416
		6	193	193	285	266	171	264		263	314	416
		7	193	193	285	266	171	264		263	314	416
		8	193	193	285	266	171	264		285	314	416
		9	193	193	285	266	171	264		263	314	416
		10	193	193	285	266	171	264		263	314	416
		11	193	193	285	266	171	266		263	314	416
		12	193	193	285	266	171	264		263	314	416
S. densiflora	Burlingame	1										
		2	193-215	191-193	279-285	266-296			245-253	263-275	314-321	
		3		185-193	285-296	266-269			245-253	263-275		
		4	193-213	185-193	277-285	271	168-174		245-253	258-274	314-336	416-440
		5	191-193				171-177	248-266		258-264	308-314	
		6	193-201	188-193	271-285							416-419
		7	193-225			266-296	171-73	248-273		266-275	314-336	422-441
		8	193-198				169-173			247-253	293-301	
		9					171					409-417
		10				226-296	167-171	248-262		246-263		
		11	189-240				166-171					
		12	193-201	176-193	277-285	270-275	171-173	248-265		263-277	314	411

Notes: Results are from control samples used by STA Labs on each of twelve plates. Plate 1 was not run. Blank cells represent missing data. Use of primer spar 25 was discontinued after plate 4.

Table 6. Observed frequency of SSR markers resulting from ISP 2009 microsatellite testing.

Primer	Allele	Frequency	Primer	Allele	Frequency	Primer	Allele	Frequency
Spar 02	180	1	Spar 15	75	1	Spar 26	209	2
	189	1		171	2		246	1
	191	1		226	1		247	1
	193*	1545		266*	1290		253	1
	196	1		267	2		258	2
	198	1		269	211		263*	1587
	201	312		270	1		264	4
	203	4		271	164		266	8
	206	1		273	76		268	1
	210	5		274	1		269	51
	213	103		275	85		271	5
	214	1		277	6		273	3
	215	2		279	35		274	1
	216	1		283	1		275	57
	225	1		285	3		277	53
	240	1		292	1		281	25
	314	5		296	4		282	2
	416	2		.	228		283	11
	.	124	Spar 20	166	1		285	55
Spar 08	176	1		167	1		286	12
	179	4		168	1		291	47
	180	137		169	1		295	1
	185	4		170	4		.	182
	188	189		171*	1557	Spar 27	23	1
	191	1		172	2		28	1
	193*	1591		173	280		293	1
	194	1		174	1		298	1
	195	1		175	25		301	1
	204	5		176	1		304	51
	205	2		177	116		305	5
	276	4		178	1		308	4
	.	172		179	30		309	5
Spar 09	173	4		181	11		314*	1475
	185	3		.	80		321	91
	193	2	Spar 23	243	1		322	2
	243	1		248	236		323	66
	271	2		249	2		324	1
	272	1		250	108		326	1
	273	44		252	1		328	122
	275	1		253	1		331	52
	276	1		254	2		336	2
	277	194		259	1		362	1
	279*	146		262*	270		363	1
	284	30		263	5		.	228
	285*	1321		264*	936	Spar 28	314	2
	290	1		265	1		409	1
	292	64		266*	298		411	2
	294	4		267	1		413	1
	295	1		268*	13		416*	1486
	296	32		269	4		417	1
	299	1		271	2		419	275
	301	1		272	1		420	37
	304	2		273	1		422	1
	.	256		274	40		427	1
				276	1		440	1
				277	1		441	1
				278	4		475	5
				280	1		476	52
				281	1		479	4
				.	180		.	242

Note: Markers with an * asterisk denote those identified as found by the UC Davis Spartina Lab to be present in Spartina foliosa. Rows marked as "." indicate missing data for each primer.

Table 7. Acres by subarea by year.

				Net acres						
Subarea	alterniflora x foliosa	densiflora	densiflora x foliosa	2001	2004	2005	2006	2007	2008	2009
1a: Channel Mouth	x			4.91	16.25	10.66	6.3	1.26	0.64	0.27
1b: Lower Channel	x			29.27	72.3	62.12	37.79	2.73	5.61	5.48
1c: Upper Channel	x			13.4	22.36	18.75	10.98	1.2	1	0.54
1d: Upper Channel - Union City Blvd to I-880	x				10.89	16.88	2.86	0.59	<¼ acre (746 m²)	<¼ acre (41 m²)
1e: Strip Marsh No. of Channel Mouth	x			2.21	9.62	9.18	4.06	<¼ acre (81 m²)	<¼ acre (207 m²)	<¼ acre (106 m²)
1f: Pond 3-AFCC	x			12.68	3.87	8.05	4.87	0.33	0.49	<¼ acre (419 m²)
1: Alameda Flood Control Channel Subarea Total	x			62.46	135.29	125.65	66.85	6.12	7.99	6.44
2a: Belmont Slough/Island, North Point, Bird Isl, Steinberger Sl/Redwood Shores	x			2.97	23.2	10.83	2.11	7.39	4.63	4.74
2b: Steinberger Sl South, Corkscrew Sl, Redwood Cr North	x			2.45	10.46	7.88	5.78	4.27	3.29	4.1
2c: B2 North Quadrant	x			23.6	35.29	43.31	36.9	10.26	21.97	9.07
2d: B2 South Quadrant - Rookery	x			28.15	30.79	18.14	9.32	1.15	1.01	3.14
2e: West Point Slough NW	x			<¼ acre (379 m²)	<¼ acre (895 m²)	0.57	0.32	0.37	<¼ acre (408 m²)	<¼ acre (529 m²)
2f: Greco Island North	x			0.5	3.19	8.07	5.68	4.86	9.97	4.23
2g: West Point Slough SW and East	x			<¼ acre (979 m²)	0.48	5.24	1.15	0.89	2.54	3.37
2h: Greco Island South	x			8.79	7.98	15.75	10.63	3.4	5.34	5.08
2i: Ravenswood Slough & Mouth	x			5.7	17.07	12.96	14.2	3.3	3.72	2.34
2j: Ravenswood Open Space Preserve	x			1.18	0.95	0.98	1.93	0.78	0.27	<¼ acre (587 m²)
2k: Redwood Creek and Deepwater Slough Restoration	x			<¼ acre (353 m²)	1.59	1.9	2.01	1.88	0.85	2.97
2l: Inner Bair	x				<¼ acre (215 m²)	<¼ acre (373 m²)	0.51	<¼ acre (200 m²)	<¼ acre (752 m²)	<¼ acre (485 m²)
2: Bair/Greco Islands Subarea Total	x			73.76	131.26	125.72	90.55	38.60	53.87	39.43
3a: Blackie's Creek (above bridge)	x	x	x	<¼ acre (133 m²)	<¼ acre (609 m²)	0.29	<¼ acre (476 m²)	<¼ acre (47 m²)	<¼ acre (46 m²)	<¼ acre (150 m²)
3b: Blackie's Creek Mouth	x	x	x	<¼ acre (160 m²)	<¼ acre (484 m²)	0.27	<¼ acre (846 m²)	<¼ acre (117 m²)	<¼ acre (209 m²)	<¼ acre (296 m²)
3: Blackie's Pasture Subarea Total	x	x	x	0.07	0.27	0.56	0.33	0.04	0.06	0.11
4a: Corte Madera Marsh Reserve	x	x	x	<¼ acre (46 m²)	<¼ acre (457 m²)	<¼ acre (631 m²)	<¼ acre (36 m²)	<¼ acre (100 m²)	<¼ acre (40 m²)	<¼ acre (151 m²)

				Net acres						
Subarea	alterniflora x foliosa	densiflora	densiflora x foliosa	2001	2004	2005	2006	2007	2008	2009
4b: College of Marin Study Area	x	x	x		<¼ acre (265 m²)	<¼ acre (132 m²)	<¼ acre (47 m²)	<¼ acre (73 m²)	<¼ acre (68 m²)	<¼ acre (14 m²)
4c: Piper Park East		x	x	<¼ acre (92 m²)	<¼ acre (37 m²)	<¼ acre (44 m²)	<¼ acre (25 m²)	<¼ acre (1 m²)	<¼ acre (3 m²)	<¼ acre (14 m²)
4d: Piper Park West	x	x		<¼ acre (59 m²)	<¼ acre (32 m²)	<¼ acre (44 m²)	<¼ acre (4 m²)	<¼ acre (13 m²)	<¼ acre (4 m²)	<¼ acre (14 m²)
4e: Larkspur Ferry Landing Area	x	x	x	<¼ acre (120 m²)	<¼ acre (127 m²)	<¼ acre (397 m²)	<¼ acre (256 m²)	<¼ acre (315 m²)	<¼ acre (10 m²)	<¼ acre (55 m²)
4f: Riviera Circle	x	x	x	0.36	<¼ acre (797 m²)	0.42	0.38	<¼ acre (316 m²)	<¼ acre (399 m²)	<¼ acre (533 m²)
4g: Creekside Park *	x	x	x	5.61	1.52	2.35	2.17	1.44	0.98	0.94
4h: Upper Corte Madera Creek (Above Bon Air Rd)	x	x	x	0.44	<¼ acre (716 m²)	0.51	0.74	0.65	<¼ acre (43 m²)	<¼ acre (111 m²)
4i: Lower Corte Madera Creek (Bon Air Rd to HWY 101)	x	x	x	0.37	0.29	0.39	0.27	<¼ acre (480 m²)	<¼ acre (811 m²)	<¼ acre (604 m²)
4j: Corte Madera Creek Mouth (Below HWY 101)	x	x	x	0.44	0.53	0.52	<¼ acre (319 m²)	1.02	<¼ acre (709 m²)	0.62
4k: Boardwalk Number One (Arkites)	x	x		<¼ acre (46 m²)	<¼ acre (93 m²)	<¼ acre (130 m²)	<¼ acre (380 m²)	<¼ acre (148 m²)	<¼ acre (32 m²)	<¼ acre (52 m²)
4l: Murphy Creek		x						<¼ acre (38 m²)		
4: Corte Madera Creek Complex Subarea Total	x	x	x	7.31	2.97	4.52	3.82	3.47	1.51	1.94
5a: Mowry Marsh-Newark Slough to Calaveras Point	x			3.98	2.25	4.19	8.57	9.46	9.36	2.72
5b: Dumbarton/Audubon	x			2.65	6.73	3.38	7.51	1.94	4.6	2.93
5c: Newark Slough	x			0.87	3.15	3.2	0.48	0.67	1.45	0.79
5d: LaRiviere Marsh	x			0.63	2.41	4.33	7.74	4.27	7.25	3.33
5e: Mayhew's Landing	x				1.43	1.52	0.26	<¼ acre (573 m²)	0.71	<¼ acre (668 m²)
5f: Coyote Creek- Alameda County	x					<¼ acre (36 m²)	<¼ acre (106 m²)	<¼ acre (127 m²)	<¼ acre (165 m²)	<¼ acre (5 m²)
5g: Cargill Pond (W Hotel)	x					<¼ acre (473 m²)	<¼ acre (511 m²)	<¼ acre (442 m²)	0.47	<¼ acre (267 m²)
5h: Plummer Creek Mitigation	x									<¼ acre (24 m²)
5: Coyote Creek/ Mowry Complex Subarea Total	x			8.13	15.96	16.75	24.70	16.62	23.88	10.02
6a: Emeryville Crescent East	x			<¼ acre (538 m²)	0.75	0.74	0.27	0.34	<¼ acre (205 m²)	<¼ acre (1000 m²)
6b: Emeryville Crescent West	x			<¼ acre (272 m²)	2.02	0.59	0.48	0.43	<¼ acre (519 m²)	<¼ acre (468 m²)
6: Emeryville Crescent Subarea Total	x			0.20	2.77	1.33	0.75	0.77	0.18	0.36
7a: Oro Loma Marsh-east	x			7.54	2.37	4.99	3.29	0.47	7.95	1.5

				Net acres						
Subarea	alterniflora x foliosa	densiflora	densiflora x foliosa	2001	2004	2005	2006	2007	2008	2009
7b: Oro Loma Marsh-west	x			6.29	15.64	36.27	15.39	0.88	8.11	2.9
7: Oro Loma Marsh Subarea Total	x			13.83	18.01	41.26	18.68	1.35	16.06	4.40
8: Palo Alto Baylands	x			<¼ acre (455 m²)	0.72	0.59	0.77	0.43	0.3	1.24
8: Palo Alto Baylands Subarea Total	x			0.11	0.72	0.59	0.77	0.43	0.30	1.24
9: Pickleweed Park	x	x	x	0.46	1.07	<¼ acre (102 m²)	<¼ acre (60 m²)	<¼ acre (44 m²)	<¼ acre (26 m²)	<¼ acre (104 m²)
9: Pickleweed Park Subarea Total	x	x	x	0.46	1.07	0.03	0.01	0.01	0.01	0.03
10a: Whittel Marsh	x	x		<¼ acre (9 m²)	<¼ acre (15 m²)	<¼ acre (697 m²)	<¼ acre (1 m²)	<¼ acre (4 m²)	<¼ acre (1 m²)	<¼ acre (10 m²)
10b: Southern Marsh	x	x		<¼ acre (12 m²)	<¼ acre (88 m²)	<¼ acre (75 m²)	<¼ acre (75 m²)	<¼ acre (306 m²)	<¼ acre (109 m²)	<¼ acre (90 m²)
10c: Giant Marsh	x	x				<¼ acre (297 m²)	<¼ acre (71 m²)	<¼ acre (39 m²)	<¼ acre (105 m²)	<¼ acre (185 m²)
10: Point Pinole Marshes Subarea Total	x	x		0.01	0.03	0.26	0.04	0.09	0.05	0.07
11: Southampton Marsh **	x			0.54	0.55	0.65	<¼ acre (866 m²)	<¼ acre (221 m²)	<¼ acre (322 m²)	<¼ acre (576 m²)
11: Southampton Marsh Subarea Total	x			0.54	0.55	0.65	0.21	0.05	0.08	0.14
12a: Pier 94	x			<¼ acre (163 m²)	<¼ acre (21 m²)	<¼ acre (119 m²)	<¼ acre (14 m²)	<¼ acre (10 m²)	<¼ acre (27 m²)	<¼ acre (18 m²)
12b: Pier 98/Heron's Head	x			<¼ acre (9 m²)	<¼ acre (23 m²)	<¼ acre (122 m²)	<¼ acre (237 m²)	<¼ acre (201 m²)	<¼ acre (261 m²)	<¼ acre (140 m²)
12c: India Basin	x			<¼ acre (388 m²)	0.31	0.68	<¼ acre (965 m²)	<¼ acre (546 m²)	<¼ acre (45 m²)	<¼ acre (163 m²)
12d: Hunters Point Naval Reserve	x				<¼ acre (530 m²)	0.56	0.43	0.43	0.61	<¼ acre (949 m²)
12e: Yosemite Channel	x			<¼ acre (842 m²)	2.05	1.49	0.32	<¼ acre (269 m²)	<¼ acre (51 m²)	<¼ acre (385 m²)
12f: Candlestick Cove	x			0.36	<¼ acre (392 m²)	0.28	0.91	0.58	0.56	<¼ acre (726 m²)
12g: Crissy Field	x				<¼ acre (17 m²)	<¼ acre (1 m²)	<¼ acre (2 m²)		<¼ acre (29 m²)	<¼ acre (1 m²)
12h: Yerba Buena Island	x					<¼ acre (3 m²)	<¼ acre (12 m²)	<¼ acre (9 m²)	<¼ acre (11 m²)	<¼ acre (2 m²)
12i: Mission Creek	x						<¼ acre (11 m²)	<¼ acre (4 m²)	<¼ acre (9 m²)	<¼ acre (13 m²)
12: Southeast San Franciso Subarea Total	x			0.70	2.61	3.08	1.96	1.27	1.27	0.59
13a: Old Alameda Creek North Bank	x			<¼ acre (217 m²)	4.61	6.84	3.02	<¼ acre (327 m²)	0.77	<¼ acre (391 m²)
13b: Old Alameda Creek Island	x			21.77	13.59	16.03	10.21	0.63	2.57	<¼ acre (862 m²)
13c: Old Alameda Creek South Bank	x			20.02	1.36	4.02	1.43	0.55	0.94	<¼ acre (40 m²)

				Net acres						
Subarea	alterniflora x foliosa	densiflora	densiflora x foliosa	2001	2004	2005	2006	2007	2008	2009
13d: Whale's Tail North Fluke	x			2.68	11.4	13.73	6.39	<¼ acre (890 m²)	0.67	0.38
13e: Whale's Tail South Fluke	x			29.73	10.99	16.14	8.68	1.04	1.02	<¼ acre (542 m²)
13f: Cargill Mitigation Marsh	x			2.59	24.65	22.16	8	1.05	1.99	<¼ acre (55 m²)
13g: Upstream of 20 Tide Gates	x			0.76	0.26	0.65	<¼ acre (867 m²)	<¼ acre (491 m²)	<¼ acre (425 m²)	<¼ acre (6 m²)
13h: Eden Landing-North Creek	x						<¼ acre (151 m²)	0.56	<¼ acre (492 m²)	<¼ acre (99 m²)
13i: Eden Landing-Pond 10	x						<¼ acre (91 m²)	<¼ acre (33 m²)	<¼ acre (118 m²)	<¼ acre (53 m²)
13j: Eden Landing-Mt Eden Creek	x			<¼ acre (168 m²)			<¼ acre (19 m²)	<¼ acre (1 m²)	<¼ acre (229 m²)	<¼ acre (584 m²)
13k: Eden Landing Reserve South	x								<¼ acre (321 m²)	<¼ acre (433 m²)
13: Whale's Tail Complex Subarea Total	x			77.64	66.86	79.56	38.01	4.26	8.35	1.14
15a: South Bay Marshes - Santa Clara County	x			0.72	6.12	1.31	2.29	3.73	3.85	1.1
15b: Faber/Laumeister Marsh	x				<¼ acre (61 m²)	<¼ acre (193 m²)	<¼ acre (157 m²)	<¼ acre (279 m²)	0.47	0.32
15c: Shoreline Regional Park at Mountain View	x			<¼ acre (55 m²)	<¼ acre (49 m²)	0.66	0.88	0.64	0.44	<¼ acre (821 m²)
15: South Bay Marshes Subarea Total	x			0.73	6.15	2.02	3.20	4.44	4.75	1.62
16: Cooley Landing (Ravenswood Open Space Preserve)	x			<¼ acre (233 m²)	4.81	5.98	5.52	3.29	11.87	8.62
16: Cooley Landing Salt Pond Restoration Subarea Total	x			0.06	4.81	5.98	5.52	3.29	11.87	8.62
17a: Alameda Island South (Elsie, Crown, Crab Cove)	x			10.1	11.97	11.98	12.56	2.86	4.51	2.53
17b: Bay Farm	x			0.87	1.97	4.58	3.04	1.33	0.3	<¼ acre (336 m²)
17c: Arrowhead Marsh	x			9.46	20.27	22.83	26.8	12.51	18.65	10.69
17d: MLK Regional Shoreline/Garretson Point	x			8.49	16.26	15.87	18.23	6.8	4.04	2.46
17e: San Leandro Creek	x			2.14	0.82	2.94	0.57	1.09	0.28	<¼ acre (712 m²)
17f: Oakland Inner Harbor	x			0.89	1.36	1.35	1.04	1.58	0.54	0.42
17g: Coast Guard Island	x			1.92	1.15	2.6	2.11	3.09	0.47	<¼ acre (452 m²)
17h: MLK Marsh	x			<¼ acre (687 m²)	1.76	4.76	7.39	3.1	3.86	3.86
17i: Coliseum Channels	x				1.16	2.63	0.56	1.56	0.25	<¼ acre (689 m²)
17j: Fan Marsh	x			1.79	6.99	6.85	7.29	1.34	4.04	2.68

				Net acres						
Subarea	alterniflora x foliosa	densiflora	densiflora x foliosa	2001	2004	2005	2006	2007	2008	2009
17k: Airport Channel	x			0.38	1.36	1.91	0.51	<¼ acre (520 m²)	<¼ acre (593 m²)	<¼ acre (632 m²)
17l: Doolittle Pond	x			<¼ acre (424 m²)	0.49	0.42	<¼ acre (1001 m²)	0.33	0.33	<¼ acre (516 m²)
17m: Alameda Island East (Aeolian Club & East Shore)	x			4.06	3.7	3.77	4.25	3.05	2.44	1.39
17: Alameda/San Leandro Bay Complex Subarea Total	x			40.39	69.25	82.50	84.60	38.77	39.88	24.86
18a: Colma Creek	x			5.49	5.31	6.29	5.03	2.48	1.01	0.58
18b: Navigable Slough	x			1.96	2.69	3.23	4.31	3.36	1.75	0.93
18c: Old Marina	x			2.22	3.34	3.4	3.52	2.6	1.51	1.34
18d: Inner Harbor	x			5.97	4.98	5.49	5.92	<¼ acre (828 m²)	2.98	0.59
18e: Sam Trans Peninsula	x			11.54	11.4	8.5	9.81	4.51	6.74	2.26
18f: Confluence Marsh	x			4.46	4.73	4.65	4.71	0.28	1.57	1.04
18g: San Bruno Marsh	x			20.04	20.66	17.9	19.24	6.25	14.42	3.44
18h: San Bruno Creek	x			1.92	0.57	1.78	1.86	0.59	<¼ acre (708 m²)	0.41
18: Colma Creek San Bruno Marsh Complex Subarea Total	x			53.60	53.68	51.23	54.41	20.27	30.15	10.60
19a: Brisbane Lagoon	x			1.91	1.95	2.65	3.04	0.95	0.53	0.65
19b: Sierra Point	x			0.36	1.33	0.46	0.72	0.97	<¼ acre (248 m²)	0.43
19c: Oyster Cove	x			0.96	0.89	1.09	1.73	0.33	0.32	<¼ acre (510 m²)
19d: Oyster Point Marina	x			<¼ acre (921 m²)	0.54	0.31	0.75	<¼ acre (48 m²)	<¼ acre (359 m²)	<¼ acre (248 m²)
19e: Oyster Point Park	x			0.46	1	1.34	1.2	0.94	0.32	<¼ acre (456 m²)
19f: Point San Bruno	x			1	1.62	1.96	1.21	1	0.42	<¼ acre (215 m²)
19g: Seaplane Harbor	x			1.14	1.76	1.39	1.21	1.51	0.39	0.47
19h: SFO	x			0.42	10.02	5.61	4.13	5	1.92	1.2
19i: Mills Creek Mouth	x			0.51	0.3	1.49	0.56	1.1	<¼ acre (796 m²)	0.3
19j: Easton Creek Mouth	x			<¼ acre (594 m²)	1.7	1.17	1.32	0.66	0.48	1.53
19k: Sanchez Marsh	x	x	x	<¼ acre (242 m²)	1.33	0.68	0.72	0.87	<¼ acre (556 m²)	0.34
19l: Burlingame Lagoon	x	x	x	<¼ acre (526 m²)	2.44	1.18	2.07	<¼ acre (912 m²)	0.3	0.43

				Net acres						
Subarea	alterniflora x foliosa	densiflora	densiflora x foliosa	2001	2004	2005	2006	2007	2008	2009
19m: Fisherman's Park	x			<¼ acre (459 m²)	<¼ acre (294 m²)	0.51	<¼ acre (143 m²)	<¼ acre (49 m²)	<¼ acre (7 m²)	<¼ acre (18 m²)
19n: Coyote Point Marina/Marsh	x			0.33	7.99	3.06	2.27	0.8	<¼ acre (937 m²)	0.35
19o: San Mateo Creek/Ryder Park	x			<¼ acre (827 m²)	0.75	1.07	1.37	0.54	<¼ acre (456 m²)	<¼ acre (356 m²)
19p: Seal Slough Mouth	x			13.16	34.8	31.79	18.04	4.53	9.56	3.49
19q: Foster City	x			0.84	4.51	1.6	0.88	<¼ acre (685 m²)	<¼ acre (760 m²)	<¼ acre (127 m²)
19r: Anza Lagoon	x			0.42	0.69	0.53	0.4	<¼ acre (845 m²)	<¼ acre (17 m²)	<¼ acre (11 m²)
19: West San Francisco Bay Subarea Total	x	x	x	22.38	73.70	57.88	41.67	19.84	15.27	9.68
20a: Oyster Bay Regional Shoreline	x			1.78	7.01	3.69	5.27	1.49	1.16	1.34
20b: Oakland Metropolitan Golf Links	x				0.38	0.4	0.54	0.44	0.62	0.63
20c: Dog Bone Marsh	x			0.67	2.44	2.89	3.43	0.32	0.46	<¼ acre (200 m²)
20d: Citation Marsh	x			0.6	9.79	9.77	13.51	5.92	1.76	3.32
20e: East Marsh	x			<¼ acre (770 m²)	0.48	0.38	0.46	0.26	<¼ acre (48 m²)	<¼ acre (36 m²)
20f: North Marsh	x			0.6	1.47	7.38	17.87	1.1	2.17	5.36
20g: Bunker Marsh	x			13.73	13.99	12.72	11.56	3.96	0.75	3.58
20h: San Lorenzo Creek & Mouth	x			10.65	20.72	17.08	10.82	1.48	0.93	1.09
20i: Bockmann Channel	x			<¼ acre (644 m²)	0.65	0.3	0.37	<¼ acre (30 m²)	<¼ acre (87 m²)	<¼ acre (42 m²)
20j: Sulphur Creek	x			0.4	<¼ acre (81 m²)		<¼ acre (149 m²)	<¼ acre (115 m²)	<¼ acre (1 m²)	<¼ acre (1 m²)
20l: Johnson's Landing	x			0.27	0.59	1.55	0.27	<¼ acre (400 m²)	<¼ acre (648 m²)	<¼ acre (9 m²)
20m: Cogswell Marsh, Quadrant A	x			6.28	7.84	18.46	6.55	<¼ acre (853 m²)	0.3	1.44
20n: Cogswell Marsh, Quadrant B	x			44.59	60.29	66.04	36.75	28.32	33.81	7.95
20o: Cogswell Marsh, Quadrant C	x			9.4	10.05	22.78	4.87	0.36	2.07	1.81
20p: Hayward Shoreline Outliers	x			1.08	0.84	<¼ acre (634 m²)	0.77	<¼ acre (370 m²)	2.38	<¼ acre (49 m²)
20q: San Leandro Shoreline Outliers	x			<¼ acre (956 m²)	1.04	0.73	1.27	0.38	0.85	0.31
20r: Oakland Airport Shoreline and Channels	x			<¼ acre (906 m²)	0.74	1.08	1.22	1.02	1.11	0.69
20s: H.A.R.D. Marsh	x					<¼ acre (202 m²)	<¼ acre (216 m²)	<¼ acre (110 m²)	<¼ acre (54 m²)	<¼ acre (30 m²)

				Net acres						
Subarea	alterniflora x foliosa	densiflora	densiflora x foliosa	2001	2004	2005	2006	2007	2008	2009
20t: San Leandro Marina	x				<¼ acre (3 m²)	<¼ acre (12 m²)	<¼ acre (4 m²)	<¼ acre (18 m²)	<¼ acre (26 m²)	<¼ acre (39 m²)
20u: Estudillo Creek Channel	x				<¼ acre (499 m²)	<¼ acre (642 m²)	<¼ acre (560 m²)	<¼ acre (431 m²)	<¼ acre (378 m²)	<¼ acre (82 m²)
20v: Howard Landing Canal	x			<¼ acre (626 m²)	0.93	19.02	1.36	0.72	<¼ acre (213 m²)	<¼ acre (134 m²)
20w: Triangle marsh	x					<¼ acre (1 m²)		<¼ acre (13 m²)	<¼ acre (1 m²)	<¼ acre (1 m²)
20: San Leandro/Hayward Shoreline Subarea Total	x			91.01	139.42	184.65	117.10	46.35	48.72	27.66
21a: Ideal Marsh North	x			19.39	14.22	16.7	5.07	0.33	0.54	0.92
21b: Ideal Marsh South	x			6.87	16.71	6.36	24.72	0.9	3.68	1.25
21: Ideal Marsh Subarea Total	x			26.26	30.92	23.06	29.79	1.23	4.22	2.17
22a: Wildcat Marsh	x			<¼ acre (76 m²)		<¼ acre (154 m²)	0.26	0.29	<¼ acre (61 m²)	0.36
22b: San Pablo Marsh	x			<¼ acre (849 m²)	<¼ acre (623 m²)	0.49	4.5	2.55	4.7	4.93
22c: Rheem Creek Area	x			<¼ acre (6 m²)	<¼ acre (6 m²)		<¼ acre (868 m²)	<¼ acre (901 m²)	<¼ acre (148 m²)	0.49
22d: Stege Marsh	x			<¼ acre (31 m²)	<¼ acre (28 m²)	<¼ acre (30 m²)	<¼ acre (142 m²)	<¼ acre (112 m²)	<¼ acre (93 m²)	<¼ acre (219 m²)
22e: Hoffman Marsh	x				<¼ acre (107 m²)	<¼ acre (75 m²)		<¼ acre (1 m²)	<¼ acre (1 m²)	<¼ acre (8 m²)
22f: Richmond/Albany Shoreline	x			<¼ acre (19 m²)	0.47	0.5	<¼ acre (241 m²)	<¼ acre (113 m²)	<¼ acre (499 m²)	<¼ acre (890 m²)
22: Two Points Complex Subarea Total	x			0.24	0.66	1.05	5.07	3.12	4.90	6.05
23a: Brickyard Cove	x	x	x	<¼ acre (0 m²)	<¼ acre (133 m²)	<¼ acre (42 m²)	<¼ acre (96 m²)	<¼ acre (2 m²)	<¼ acre (184 m²)	<¼ acre (9 m²)
23b: Beach Drive	x				<¼ acre (492 m²)	0.33	0.4	<¼ acre (459 m²)	<¼ acre (702 m²)	<¼ acre (1002 m²)
23c: Loch Lomond Marina	x			<¼ acre (19 m²)	<¼ acre (593 m²)	<¼ acre (196 m²)	<¼ acre (240 m²)	<¼ acre (37 m²)	<¼ acre (331 m²)	<¼ acre (251 m²)
23d: San Rafael Canal Mouth North	x	x	x	<¼ acre (1 m²)	<¼ acre (789 m²)	<¼ acre (7 m²)	<¼ acre (63 m²)	<¼ acre (287 m²)	<¼ acre (32 m²)	<¼ acre (276 m²)
23e: Muzzi & Martas Marsh	x	x	x	<¼ acre (21 m²)	<¼ acre (20 m²)	<¼ acre (45 m²)	<¼ acre (348 m²)	<¼ acre (556 m²)	<¼ acre (100 m²)	<¼ acre (152 m²)
23f: Paradise Cay	x	x		<¼ acre (13 m²)	0.42	0.41	<¼ acre (371 m²)	<¼ acre (773 m²)	<¼ acre (67 m²)	<¼ acre (58 m²)
23g: Greenwood Beach Road/Harbor	x	x	x	<¼ acre (97 m²)	<¼ acre (82 m²)	<¼ acre (64 m²)	<¼ acre (143 m²)	<¼ acre (123 m²)	<¼ acre (142 m²)	<¼ acre (90 m²)
23h: Strawberry Point	x	x	x	<¼ acre (31 m²)	<¼ acre (85 m²)	<¼ acre (286 m²)	<¼ acre (150 m²)	<¼ acre (38 m²)	<¼ acre (35 m²)	<¼ acre (103 m²)
23i: Strawberry Cove	x			<¼ acre (19 m²)				<¼ acre (601 m²)	<¼ acre (30 m²)	<¼ acre (136 m²)
23j: Bothin Marsh	x				0.27		0.55	0.42	<¼ acre (516 m²)	<¼ acre (84 m²)

				Net acres						
Subarea	alterniflora x foliosa	densiflora	densiflora x foliosa	2001	2004	2005	2006	2007	2008	2009
23k: Sausalito	x				0.34	<¼ acre (299 m²)	<¼ acre (16 m²)	<¼ acre (454 m²)	<¼ acre (470 m²)	<¼ acre (121 m²)
23l: Starkweather Park	x	x	x				<¼ acre (149 m²)	<¼ acre (86 m²)	<¼ acre (4 m²)	<¼ acre (27 m²)
23m: Novato	x					<¼ acre (31 m²)	<¼ acre (181 m²)	<¼ acre (97 m²)	<¼ acre (14 m²)	<¼ acre (37 m²)
23n: Triangle Marsh	x	x	x		<¼ acre (12 m²)	<¼ acre (38 m²)	<¼ acre (74 m²)	<¼ acre (106 m²)	<¼ acre (13 m²)	<¼ acre (44 m²)
23: Marin Outliers Subarea Total	x	x	x	0.05	1.57	1.00	1.41	1.31	0.65	0.59
24a: Upper Petaluma River- Upstream of Grey's Field	x						<¼ acre (210 m²)	<¼ acre (607 m²)	<¼ acre (30 m²)	<¼ acre (59 m²)
24b: Grey's Field	x									<¼ acre (8 m²)
24c: Petaluma Marsh	x						<¼ acre (13 m²)	<¼ acre (13 m²)	<¼ acre (0 m²)	<¼ acre (1 m²)
24: Petaluma River Subarea Total	x			0.00	0.00	0.00	0.06	0.15	0.01	0.02
25a: Tom's Point, Tomales		x	x	<¼ acre (2 m²)	<¼ acre (1 m²)	<¼ acre (0 m²)	<¼ acre (3 m²)	<¼ acre (3 m²)	<¼ acre (2 m²)	<¼ acre (3 m²)
25b: Limantour Estero	x				<¼ acre (12 m²)		<¼ acre (15 m²)	<¼ acre (63 m²)		<¼ acre (33 m²)
25c: Drakes Estero	x						<¼ acre (108 m²)	<¼ acre (129 m²)	<¼ acre (0 m²)	<¼ acre (0 m²)
25d: Bolinas Lagoon, North	x			<¼ acre (7 m²)	<¼ acre (7 m²)				<¼ acre (10 m²)	<¼ acre (1 m²)
25e: Bolinas Lagoon, South	x				<¼ acre (37 m²)				<¼ acre (27 m²)	<¼ acre (1 m²)
25: Outer Coast Subarea Total	x	x	x	0.00	0.01	0.00	0.03	0.05	0.01	0.01
26a: White Slough/Napa River	x							<¼ acre (52 m²)	<¼ acre (74 m²)	<¼ acre (2 m²)
26b: San Pablo Bay NWR and Mare Island	x	x	x					<¼ acre (100 m²)	<¼ acre (87 m²)	0.31
26c: Sonoma Creek	x								<¼ acre (0 m²)	<¼ acre (40 m²)
26d: Sonoma Baylands	x								<¼ acre (107 m²)	
26: North San Pablo Bay Subarea Total	x	x	x	0.00	0.00	0.00	0.00	0.04	0.07	0.32

* Creekside Park also contains *S. anglica.*

**Southhampton Marsh also contains *S. patens.*

Table 8. Change in acres 2009 since 2005 and 2008.

Subarea	2005-2009 Decrease	2008-2009 Decrease
1a: Channel Mouth	97%	58%
1b: Lower Channel	91%	2%
1c: Upper Channel	97%	46%
1d: Upper Channel - Union City Blvd to I-880	100%	94%
1e: Strip Marsh No. of Channel Mouth	100%	49%
1f: Pond 3-AFCC	99%	79%
1: Alameda Flood Control Channel Subarea Total	**95%**	**19%**
2a: Belmont Slough/Island, North Point, Bird Isl, Steinberger Sl/Redwood Shores	56%	increase (0.11 ac)
2b: Steinberger Sl South, Corkscrew Sl, Redwood Cr North	48%	increase (1.07 ac)
2c: B2 North Quadrant	79%	59%
2d: B2 South Quadrant - Rookery	83%	increase (2.13 ac)
2e: West Point Slough NW	77%	increase (0.03 ac)
2f: Greco Island North	48%	58%
2g: West Point Slough SW and East	36%	increase (0.83 ac)
2h: Greco Island South	68%	5%
2i: Ravenswood Slough & Mouth	82%	37%
2j: Ravenswood Open Space Preserve	85%	46%
2k: Redwood Creek and Deepwater Slough Restoration	increase (1.07 ac)	increase (2.12 ac)
2l: Inner Bair	increase (0.03 ac)	36%
2: Bair/Greco Islands Subarea Total	**69%**	**27%**
3a: Blackie's Creek (above bridge)	87%	increase (0.03 ac)
3b: Blackie's Creek Mouth	73%	increase (0.02 ac)
3: Blackie's Pasture Subarea Total	**80%**	**increase (0.05 ac)**
4a: Corte Madera Marsh Reserve	76%	increase (0.03 ac)
4b: College of Marin Study Area	89%	79%
4c: Piper Park East	67%	increase (<0.01 ac)
4d: Piper Park West	69%	increase (<0.01 ac)
4e: Larkspur Ferry Landing Area	86%	increase (0.01 ac)
4f: Riviera Circle	68%	increase (0.03 ac)
4g: Creekside Park	60%	5%
4h: Upper Corte Madera Creek (Above Bon Air Rd)	95%	increase (0.02 ac)
4i: Lower Corte Madera Creek (Bon Air Rd to HWY 101)	61%	26%
4j: Corte Madera Creek Mouth (Below HWY 101)	increase (0.10 ac)	increase (0.44 ac)
4k: Boardwalk Number One (Arkites)	60%	increase (0.01 ac)
4: Corte Madera Creek Complex Subarea Total	**57%**	**increase (0.43 ac)**
5a: Mowry Marsh-Newark Slough to Calaveras Point	35%	71%
5b: Dumbarton/Audubon	13%	36%
5c: Newark Slough	75%	45%
5d: LaRiviere Marsh	23%	54%
5e: Mayhew's Landing	89%	77%
5f: Coyote Creek- Alameda County	85%	97%
5g: Cargill Pond (W Hotel)	44%	86%
5h: Plummer Creek Mitigation	n/a	n/a

Subarea	2005-2009 Decrease	2008-2009 Decrease
5: Coyote Creek/ Mowry Complex Subarea Total	**40%**	**58%**
6a: Emeryville Crescent East	67%	increase (0.20 ac)
6b: Emeryville Crescent West	80%	10%
6: Emeryville Crescent Subarea Total	**73%**	**increase (0.18 ac)**
7a: Oro Loma Marsh-east	70%	81%
7b: Oro Loma Marsh-west	92%	64%
7: Oro Loma Marsh Subarea Total	**89%**	**73%**
8: Palo Alto Baylands	increase (0.65 ac)	increase (0.95 ac)
8: Palo Alto Baylands Subarea Total	**increase (0.65 ac)**	**increase (0.95 ac)**
9: Pickleweed Park	0%	increase (0.02 ac)
9: Pickleweed Park Subarea Total	**0%**	**increase (0.02 ac)**
10a: Whittel Marsh	99%	increase (<0.01 ac)
10b: Southern Marsh	increase (<0.01 ac)	18%
10c: Giant Marsh	38%	increase (0.02 ac)
10: Point Pinole Marshes Subarea Total	**73%**	**increase (0.02 ac)**
11: Southampton Marsh	78%	increase (0.06 ac)
11: Southampton Marsh Subarea Total	**78%**	**increase (0.06 ac)**
12a: Pier 94	85%	33%
12b: Pier 98/Heron's Head	0%	46%
12c: India Basin	94%	increase (0.03 ac)
12d: Hunters Point Naval Reserve	58%	61%
12e: Yosemite Channel	94%	increase (0.08 ac)
12f: Candlestick Cove	36%	68%
12g: Crissy Field	increase (<0.01 ac)	95%
12h: Yerba Buena Island	17%	77%
12i: Mission Creek	n/a	increase (<0.01 ac)
12: Southeast San Franciso Subarea Total	**81%**	**53%**
13a: Old Alameda Creek North Bank	99%	87%
13b: Old Alameda Creek Island	99%	92%
13c: Old Alameda Creek South Bank	100%	99%
13d: Whale's Tail North Fluke	97%	43%
13e: Whale's Tail South Fluke	99%	87%
13f: Cargill Mitigation Marsh	100%	99%
13g: Upstream of 20 Tide Gates	100%	99%
13h: Eden Landing-North Creek	n/a	80%
13i: Eden Landing-Pond 10	n/a	55%
13j: Eden Landing-Mt Eden Creek	n/a	increase (0.09 ac)
13k: Eden Landing Reserve South	n/a	increase (0.03 ac)
13: Whale's Tail Complex Subarea Total	**99%**	**86%**
15a: South Bay Marshes - Santa Clara County	16%	71%
15b: Faber/Laumeister Marsh	increase (0.27 ac)	31%
15c: Shoreline Regional Park at Mountain View	69%	54%
15: South Bay Marshes Subarea Total	**20%**	**66%**
16: Cooley Landing (Ravenswood Open Space Preserve)	increase (2.64 ac)	27%
16: Cooley Landing Salt Pond Restoration Subarea Total	**increase (2.64 ac)**	**27%**

Subarea	2005-2009 Decrease	2008-2009 Decrease
17a: Alameda Island South (Elsie, Crown, Crab Cove)	79%	44%
17b: Bay Farm	98%	72%
17c: Arrowhead Marsh	53%	43%
17d: MLK Regional Shoreline/Garretson Point	85%	39%
17e: San Leandro Creek	94%	38%
17f: Oakland Inner Harbor	69%	22%
17g: Coast Guard Island	96%	76%
17h: MLK Marsh	19%	0%
17i: Coliseum Channels	94%	33%
17j: Fan Marsh	61%	34%
17k: Airport Channel	92%	increase (0.01 ac)
17l: Doolittle Pond	70%	61%
17m: Alameda Island East (Aeolian Club & East Shore)	63%	43%
17: Alameda/San Leandro Bay Complex Subarea Total	**70%**	**38%**
18a: Colma Creek	91%	42%
18b: Navigable Slough	71%	47%
18c: Old Marina	61%	12%
18d: Inner Harbor	89%	80%
18e: Sam Trans Peninsula	73%	67%
18f: Confluence Marsh	78%	34%
18g: San Bruno Marsh	81%	76%
18h: San Bruno Creek	77%	increase (0.24 ac)
18: Colma Creek San Bruno Marsh Complex Subarea Total	**79%**	**65%**
19a: Brisbane Lagoon	76%	increase (0.11 ac)
19b: Sierra Point	6%	increase (0.37 ac)
19c: Oyster Cove	88%	60%
19d: Oyster Point Marina	80%	31%
19e: Oyster Point Park	92%	65%
19f: Point San Bruno	97%	87%
19g: Seaplane Harbor	66%	increase (0.08 ac)
19h: SFO	79%	37%
19i: Mills Creek Mouth	80%	increase (0.10 ac)
19j: Easton Creek Mouth	increase (0.36 ac)	increase (1.05 ac)
19k: Sanchez Marsh	51%	increase (0.20 ac)
19l: Burlingame Lagoon	63%	increase (0.14 ac)
19m: Fisherman's Park	99%	increase (<0.01 ac)
19n: Coyote Point Marina/Marsh	89%	increase (0.12 ac)
19o: San Mateo Creek/Ryder Park	92%	22%
19p: Seal Slough Mouth	89%	64%
19q: Foster City	98%	83%
19r: Anza Lagoon	99%	33%
19: West San Francisco Bay Subarea Total	**83%**	**37%**
20a: Oyster Bay Regional Shoreline	64%	increase (0.18 ac)
20b: Oakland Metropolitan Golf Links	increase (0.23 ac)	increase (0.12 ac)
20c: Dog Bone Marsh	98%	89%
20d: Citation Marsh	66%	increase (0.12 ac)

Subarea	2005-2009 Decrease	2008-2009 Decrease
20e: East Marsh	98%	26%
20f: North Marsh	27%	increase (0.12 ac)
20g: Bunker Marsh	72%	increase (0.12 ac)
20h: San Lorenzo Creek & Mouth	94%	increase (0.12 ac)
20i: Bockmann Channel	97%	52%
20j: Sulphur Creek	n/a	increase (<0.01 ac)
20l: Johnson's Landing	100%	99%
20m: Cogswell Marsh, Quadrant A	92%	increase (0.12 ac)
20n: Cogswell Marsh, Quadrant B	88%	76%
20o: Cogswell Marsh, Quadrant C	92%	12%
20p: Hayward Shoreline Outliers	92%	99%
20q: San Leandro Shoreline Outliers	57%	63%
20r: Oakland Airport Shoreline and Channels	36%	38%
20s: H.A.R.D. Marsh	85%	44%
20t: San Leandro Marina	increase (0.01 ac)	increase (<0.01 ac)
20u: Estudillo Creek Channel	87%	78%
20v: Howard Landing Canal	100%	37%
20w: Triangle marsh	0%	increase (<0.01 ac)
20: San Leandro/Hayward Shoreline Subarea Total	**85%**	**43%**
21a: Ideal Marsh North	94%	71%
21b: Ideal Marsh South	80%	66%
21: Ideal Marsh Subarea Total	**91%**	**48%**
22a: Wildcat Marsh	increase (0.32 ac)	increase (0.34 ac)
22b: San Pablo Marsh	increase (4.44 ac)	increase (0.22 ac)
22c: Rheem Creek Area	n/a	increase (0.46 ac)
22d: Stege Marsh	increase (0.05 ac)	increase (0.03 ac)
22e: Hoffman Marsh	89%	increase (<0.01 ac)
22f: Richmond/Albany Shoreline	56%	increase (0.10 ac)
22: Two Points Complex Subarea Total	**increase (5.0 ac)**	**increase (1.15 ac)**
23a: Brickyard Cove	78%	95%
23b: Beach Drive	26%	increase (0.07 ac)
23c: Loch Lomond Marina	increase (0.01 ac)	24%
23d: San Rafael Canal Mouth North	increase (0.07 ac)	increase (0.06 ac)
23e: Muzzi & Martas Marsh	increase (0.03 ac)	increase (0.01 ac)
23f: Paradise Cay	97%	13%
23g: Greenwood Beach Road/Harbor	increase (0.01 ac)	37%
23h: Strawberry Point	64%	increase (0.02 ac)
23i: Strawberry Cove	n/a	increase (0.03 ac)
23j: Bothin Marsh	n/a	84%
23k: Sausalito	59%	74%
23l: Starkweather Park	n/a	increase (0.01 ac)
23m: Novato	increase (<0.01 ac)	increase (0.01 ac)
23n: Triangle Marsh	increase (<0.01 ac)	increase (0.01 ac)
23: Marin Outliers Subarea Total	**41%**	**9%**
24a: Upper Petaluma River- Upstream of Grey's Field	n/a	increase (0.01 ac)
24b: Grey's Field	n/a	n/a

Subarea	2005-2009 Decrease	2008-2009 Decrease
24c: Petaluma Marsh	n/a	increase (<0.01 ac)
24: Petaluma River Subarea Total	**n/a**	**increase (0.01 ac)**
25a: Tom's Point, Tomales	increase (0.01 ac)	increase (<0.01 ac)
25b: Limantour Estero	n/a	n/a
25c: Drakes Estero	n/a	increase (<0.01 ac)
25d: Bolinas Lagoon, North	n/a	94%
25e: Bolinas Lagoon, South	n/a	97%
25: Outer Coast Subarea Total	**increase (0.01 ac)**	**3%**
26a: White Slough/Napa River	n/a	97%
26b: San Pablo Bay NWR and Mare Island	n/a	increase (0.28 ac)
26c: Sonoma Creek	n/a	increase (0.01 ac)
26d: Sonoma Baylands	n/a	100%
26: North San Pablo Bay Subarea Total	**n/a**	**increase (0.25 ac)**

Table 9. Treatment Acres by Subarea by Year

	Spartina species					Net Treatment Acres	
Subarea	anglica	alterniflora x foliosa	densiflora	densiflora x foliosa	patens	**2008**	**2009**
1a: Channel Mouth		x				1.19	0.62
1b: Lower Channel		x				11.50	11.42
1c: Upper Channel		x				1.55	0.91
1d: Upper Channel - Union City Blvd to I-880		x				<¼ acre (970 m²)	<¼ acre (97 m²)
1e: Strip Marsh No. of Channel Mouth		x				<¼ acre (419 m²)	<¼ acre (164 m²)
1f: Pond 3-AFCC		x				0.64	<¼ acre (651 m²)
1: Alameda Flood Control Channel Subarea Total		x				**15.22**	**13.18**
2a: Belmont Slough/Island, North Point, Bird Isl, Steinberger Sl/Redwood Shores		x				9.35	10.58
2b: Steinberger Sl South, Corkscrew Sl, Redwood Cr North		x				7.05	9.84
2c: B2 North Quadrant		x				27.89	16.01
2d: B2 South Quadrant - Rookery		x				2.20	8.75
2e: West Point Slough NW		x				<¼ acre (651 m²)	0.32
2f: Greco Island North		x				16.19	12.26
2g: West Point Slough SW and East		x				3.64	4.89
2h: Greco Island South		x				8.63	10.52
2i: Ravenswood Slough & Mouth		x				7.71	4.53
2j: Ravenswood Open Space Preserve		x				0.37	0.34
2k: Redwood Creek and Deepwater Slough Restoration		x				1.30	5.94
2l: Inner Bair		x				0.49	0.30
2m: Pond B3 (Middle Bair)		x				<¼ acre (13 m²)	0
2: Bair/Greco Islands Subarea Total		x				**84.98**	**84.27**
3a: Blackie's Creek (above bridge)		x	x	x		<¼ acre (103 m²)	<¼ acre (203 m²)
3b: Blackie's Creek Mouth		x	x	x		<¼ acre (458 m²)	<¼ acre (504 m²)
3: Blackie's Pasture Subarea Total		x	x	x		**0.14**	**0.17**
4a: Corte Madera Marsh Reserve		x	x	x		<¼ acre (101 m²)	<¼ acre (305 m²)
4b: College of Marin Study Area		x	x	x		<¼ acre (119 m²)	<¼ acre (20 m²)
4c: Piper Park East			x	x		<¼ acre (5 m²)	<¼ acre (22 m²)
4d: Piper Park West		x	x			<¼ acre (37 m²)	<¼ acre (25 m²)
4e: Larkspur Ferry Landing Area		x	x	x		<¼ acre (17 m²)	<¼ acre (75 m²)
4f: Riviera Circle		x	x	x		0.29	<¼ acre (749 m²)
4g: Creekside Park	x	x	x	x		1.65	1.62
4h: Upper Corte Madera Creek (Above Bon Air Rd)		x	x	x		<¼ acre (331 m²)	<¼ acre (194 m²)
4i: Lower Corte Madera Creek (Bon Air Rd to HWY 101)		x	x	x		0.52	<¼ acre (960 m²)
4j: Corte Madera Creek Mouth (Below HWY 101)		x	x	x		0.45	0.99
4k: Boardwalk Number One (Arkites)		x	x			<¼ acre (60 m²)	<¼ acre (84 m²)

Subarea	Spartina species					Net Treatment Acres	
	anglica	alterniflora x foliosa	densiflora	densiflora x foliosa	patens	2008	2009
4l: Murphy Creek			x			<¼ acre (0 m²)	<¼ acre (0 m²)
4: Corte Madera Creek Complex Subarea Total	x	x	x	x		**3.07**	**3.21**
5a: Mowry Marsh-Newark Slough to Calaveras Point		x				18.36	5.98
5b: Dumbarton/Audubon		x				8.44	7.80
5c: Newark Slough		x				2.10	1.67
5d: LaRiviere Marsh		x				10.85	7.93
5e: Mayhew's Landing		x				1.22	0.39
5f: Coyote Creek- Alameda County		x				<¼ acre (305 m²)	<¼ acre (7 m²)
5g: Cargill Pond (W Hotel)		x				0.62	<¼ acre (654 m²)
5h: Plummer Creek Mitigation		x				<¼ acre (0 m²)	<¼ acre (52 m²)
5: Coyote Creek/ Mowry Complex Subarea Total		x				**41.67**	**23.95**
6a: Emeryville Crescent East		x				<¼ acre (656 m²)	0.51
6b: Emeryville Crescent West		x				0.28	<¼ acre (998 m²)
6: Emeryville Crescent Subarea Total		x				**0.44**	**0.76**
7a: Oro Loma Marsh-east		x				9.96	5.54
7b: Oro Loma Marsh-west		x				11.77	6.73
7: Oro Loma Marsh Subarea Total		x				**21.73**	**12.27**
8: Palo Alto Baylands		x				0.44	1.79
8: Palo Alto Baylands Subarea Total		x				**0.44**	**1.79**
9: Pickleweed Park		x	x	x		<¼ acre (39 m²)	<¼ acre (182 m²)
9: Pickleweed Park Subarea Total		x	x	x		**<¼ acre (39 m²)**	**<¼ acre (182 m²)**
10a: Whittel Marsh		x	x			<¼ acre (7 m²)	<¼ acre (20 m²)
10b: Southern Marsh		x	x			<¼ acre (246 m²)	<¼ acre (166 m²)
10c: Giant Marsh		x	x			<¼ acre (235 m²)	<¼ acre (315 m²)
10: Point Pinole Marshes Subarea Total		x	x			**0.12**	**0.12**
11: Southampton Marsh		x			x	<¼ acre (491 m²)	<¼ acre (810 m²)
11: Southampton Marsh Subarea Total		x			x	**0.12**	**0.2**
12a: Pier 94		x				<¼ acre (31 m²)	<¼ acre (34 m²)
12b: Pier 98/Heron's Head		x				<¼ acre (425 m²)	<¼ acre (304 m²)
12c: India Basin		x				<¼ acre (149 m²)	<¼ acre (215 m²)
12d: Hunters Point Naval Reserve		x				0.89	0.39
12e: Yosemite Channel		x				<¼ acre (177 m²)	<¼ acre (730 m²)
12f: Candlestick Cove		x				0.88	0.26
12g: Crissy Field		x				<¼ acre (52 m²)	<¼ acre (5 m²)
12h: Yerba Buena Island		x				<¼ acre (12 m²)	<¼ acre (3 m²)
12i: Mission Creek		x				<¼ acre (17 m²)	<¼ acre (20 m²)

Subarea	Spartina species					Net Treatment Acres	
	anglica	alterniflora x foliosa	densiflora	densiflora x foliosa	patens	2008	2009
12: Southeast San Franciso Subarea Total		x				**1.98**	**0.98**
13a: Old Alameda Creek North Bank		x				1.42	<¼ acre (701 m²)
13b: Old Alameda Creek Island		x				6.44	0.36
13c: Old Alameda Creek South Bank		x				2.14	<¼ acre (130 m²)
13d: Whale's Tail North Fluke		x				1.03	0.66
13e: Whale's Tail South Fluke		x				2.35	0.35
13f: Cargill Mitigation Marsh		x				2.87	<¼ acre (86 m²)
13g: Upstream of 20 Tide Gates		x				<¼ acre (868 m²)	<¼ acre (13 m²)
13h: Eden Landing-North Creek		x				<¼ acre (651 m²)	<¼ acre (240 m²)
13i: Eden Landing-Pond 10		x				<¼ acre (262 m²)	<¼ acre (82 m²)
13j: Eden Landing-Mt Eden Creek		x				<¼ acre (430 m²)	<¼ acre (756 m²)
13k: Eden Landing Reserve South		x				<¼ acre (937 m²)	0.34
13: Whale's Tail Complex Subarea Total		x				**17.03**	**2.20**
15a: South Bay Marshes - Santa Clara County		x				4.79	2.15
15b: Faber/Laumeister Marsh		x				1.03	0.83
15c: Shoreline Regional Park at Mountain View		x				0.53	0.57
15: South Bay Marshes Subarea Total		x				**6.35**	**3.55**
16: Cooley Landing (Ravenswood Open Space Preserve)		x				19.05	22.59
16: Cooley Landing Salt Pond Restoration Subarea Total		x				**19.05**	**22.59**
17a: Alameda Island South (Elsie, Crown, Crab Cove)		x				6.37	3.76
17b: Bay Farm		x				0.59	<¼ acre (719 m²)
17c: Arrowhead Marsh		x				26.27	16.73
17d: MLK Regional Shoreline/Garretson Point		x				8.91	3.82
17e: San Leandro Creek		x				0.44	0.31
17f: Oakland Inner Harbor		x				0.92	0.86
17g: Coast Guard Island		x				0.76	<¼ acre (792 m²)
17h: MLK Marsh		x				6.42	6.02
17i: Coliseum Channels		x				0.45	0.33
17j: Fan Marsh		x				5.91	3.57
17k: Airport Channel		x				0.26	0.27
17l: Doolittle Pond		x				0.81	<¼ acre (987 m²)
17m: Alameda Island East (Aeolian Club & East Shore)		x				3.74	2.68
17: Alameda/San Leandro Bay Complex Subarea Total		x				**61.86**	**38.96**
18a: Colma Creek		x				1.34	0.89
18b: Navigable Slough		x				2.57	1.88
18c: Old Marina		x				2.25	1.99

Subarea	Spartina species					Net Treatment Acres	
	anglica	alterniflora x foliosa	densiflora	densiflora x foliosa	patens	2008	2009
18d: Inner Harbor		x				4.23	1.03
18e: Sam Trans Peninsula		x				8.86	4.32
18f: Confluence Marsh		x				3.23	1.84
18g: San Bruno Marsh		x				17.99	5.51
18h: San Bruno Creek		x				0.33	0.57
18: Colma Creek San Bruno Marsh Complex Subarea Total		x				**40.79**	**18.02**
19a: Brisbane Lagoon		x				1.30	1.01
19b: Sierra Point		x				<¼ acre (822 m²)	0.56
19c: Oyster Cove		x				0.70	<¼ acre (955 m²)
19d: Oyster Point Marina		x				<¼ acre (752 m²)	<¼ acre (441 m²)
19e: Oyster Point Park		x				0.76	<¼ acre (851 m²)
19f: Point San Bruno		x				0.92	<¼ acre (528 m²)
19g: Seaplane Harbor		x				0.66	0.82
19h: SFO		x				5.41	2.19
19i: Mills Creek Mouth		x				0.49	0.61
19j: Easton Creek Mouth		x				0.93	2.53
19k: Sanchez Marsh		x	x	x		0.40	0.79
19l: Burlingame Lagoon		x	x	x		0.45	0.80
19m: Fisherman's Park		x				<¼ acre (12 m²)	<¼ acre (32 m²)
19n: Coyote Point Marina/Marsh		x				0.51	0.60
19o: San Mateo Creek/Ryder Park		x				<¼ acre (718 m²)	<¼ acre (573 m²)
19p: Seal Slough Mouth		x				17.39	7.51
19q: Foster City		x				0.48	<¼ acre (294 m²)
19r: Anza Lagoon		x				<¼ acre (31 m²)	<¼ acre (27 m²)
19: West San Francisco Bay Subarea Total		x	x	x		**30.96**	**18.34**
20a: Oyster Bay Regional Shoreline		x				1.80	1.81
20b: Oakland Metropolitan Golf Links		x				0.73	1.01
20c: Dog Bone Marsh		x				0.69	<¼ acre (417 m²)
20d: Citation Marsh		x				1.79	7.20
20e: East Marsh		x				<¼ acre (76 m²)	<¼ acre (68 m²)
20f: North Marsh		x				2.86	9.41
20g: Bunker Marsh		x				1.35	6.20
20h: San Lorenzo Creek & Mouth		x				1.85	2.30
20i: Bockmann Channel		x				<¼ acre (143 m²)	<¼ acre (68 m²)
20j: Sulphur Creek		x				<¼ acre (1 m²)	<¼ acre (2 m²)
20l: Johnson's Landing		x				<¼ acre (894 m²)	<¼ acre (29 m²)

Subarea	Spartina species					Net Treatment Acres	
	anglica	alterniflora x foliosa	densiflora	densiflora x foliosa	patens	2008	2009
20m: Cogswell Marsh, Quadrant A		x				0.54	2.63
20n: Cogswell Marsh, Quadrant B		x				48.70	22.13
20o: Cogswell Marsh, Quadrant C		x				3.71	4.16
20p: Hayward Shoreline Outliers		x				2.73	<¼ acre (135 m²)
20q: San Leandro Shoreline Outliers		x				1.08	0.63
20r: Oakland Airport Shoreline and Channels		x				1.33	1.23
20s: H.A.R.D. Marsh		x				<¼ acre (79 m²)	<¼ acre (47 m²)
20t: San Leandro Marina		x				<¼ acre (72 m²)	<¼ acre (70 m²)
20u: Estudillo Creek Channel		x				<¼ acre (606 m²)	<¼ acre (145 m²)
20v: Howard Landing Canal		x				<¼ acre (496 m²)	<¼ acre (360 m²)
20w: Triangle marsh		x				<¼ acre (1 m²)	<¼ acre (2 m²)
20: San Leandro/Hayward Shoreline Subarea Total		x				**69.76**	**59.04**
21a: Ideal Marsh North		x				1.54	1.76
21b: Ideal Marsh South		x				4.73	3.24
21: Ideal Marsh Subarea Total		x				**6.26**	**5.00**
22a: Wildcat Marsh		x				<¼ acre (143 m²)	0.53
22b: San Pablo Marsh		x				6.98	9.18
22c: Rheem Creek Area		x				<¼ acre (432 m²)	0.66
22d: Stege Marsh		x				<¼ acre (158 m²)	<¼ acre (156 m²)
22e: Hoffman Marsh		x				<¼ acre (2 m²)	<¼ acre (21 m²)
22f: Richmond/Albany Shoreline		x				<¼ acre (626 m²)	0.50
22: Two Points Complex Subarea Total		x				**7.32**	**10.91**
23a: Brickyard Cove		x	x	x		<¼ acre (475 m²)	<¼ acre (16 m²)
23b: Beach Drive		x				0.38	0.36
23c: Loch Lomond Marina		x				<¼ acre (792 m²)	<¼ acre (454 m²)
23d: San Rafael Canal Mouth North		x	x	x		<¼ acre (54 m²)	<¼ acre (714 m²)
23e: Muzzi & Martas Marsh		x	x	x		<¼ acre (153 m²)	<¼ acre (417 m²)
23f: Paradise Cay		x	x			<¼ acre (156 m²)	<¼ acre (129 m²)
23g: Greenwood Beach Road/Harbor		x	x	x		<¼ acre (224 m²)	<¼ acre (142 m²)
23h: Strawberry Point		x	x	x		<¼ acre (64 m²)	<¼ acre (228 m²)
23i: Strawberry Cove		x				<¼ acre (132 m²)	<¼ acre (323 m²)
23j: Bothin Marsh		x				0.40	<¼ acre (203 m²)
23k: Sausalito		x				<¼ acre (548 m²)	<¼ acre (300 m²)
23l: Starkweather Park		x	x	x		<¼ acre (8 m²)	<¼ acre (49 m²)
23m: Novato		x				<¼ acre (115 m²)	<¼ acre (58 m²)
23n: Triangle Marsh		x	x	x		<¼ acre (24 m²)	<¼ acre (71 m²)

Subarea	Spartina species					Net Treatment Acres	
	anglica	alterniflora x foliosa	densiflora	densiflora x foliosa	patens	2008	2009
23: Marin Outliers Subarea Total		x	x	x		**1.45**	**1.13**
24a: Upper Petaluma River- Upstream of Grey's Field		x				<¼ acre (96 m²)	<¼ acre (127 m²)
24b: Grey's Field		x				<¼ acre (0 m²)	<¼ acre (11 m²)
24c: Petaluma Marsh		x				<¼ acre (1 m²)	<¼ acre (2 m²)
24: Petaluma River Subarea Total		x				**0.02**	**0.03**
25a: Tom's Point, Tomales			x	x		<¼ acre (2 m²)	<¼ acre (17 m²)
25b: Limantour Estero		x				<¼ acre (0 m²)	<¼ acre (64 m²)
25c: Drakes Estero		x				<¼ acre (1 m²)	<¼ acre (12 m²)
25d: Bolinas Lagoon, North		x				<¼ acre (22 m²)	<¼ acre (2 m²)
25e: Bolinas Lagoon, South		x				<¼ acre (39 m²)	<¼ acre (16 m²)
25: Outer Coast Subarea Total		x	x	x		**0.02**	**0.03**
26a: White Slough/Napa River		x				<¼ acre (128 m²)	<¼ acre (13 m²)
26b: San Pablo Bay NWR and Mare Island		x	x	x		<¼ acre (110 m²)	0.91
26c: Sonoma Creek		x				<¼ acre (1 m²)	<¼ acre (187 m²)
26d: Sonoma Baylands		x				<¼ acre (130 m²)	0.00
26: North San Pablo Bay Subarea Total		x	x	x		**0.09**	**0.96**

Figures I

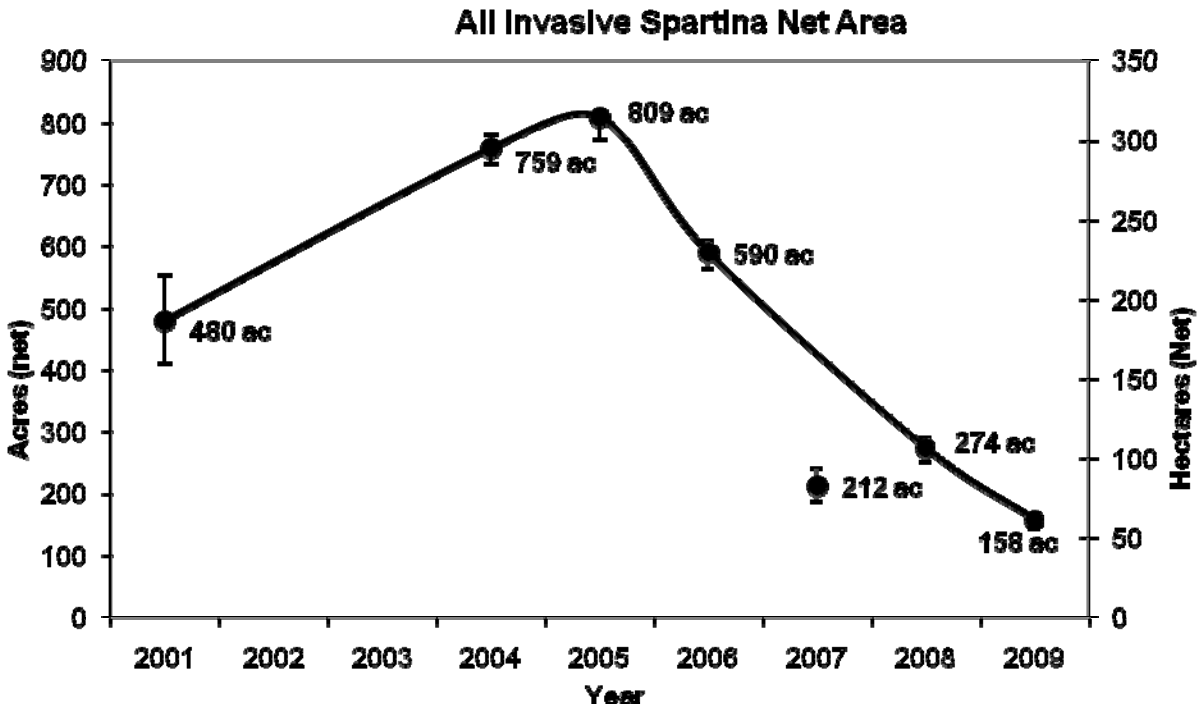


Figure 1.1. All Invasive Spartina Net Area

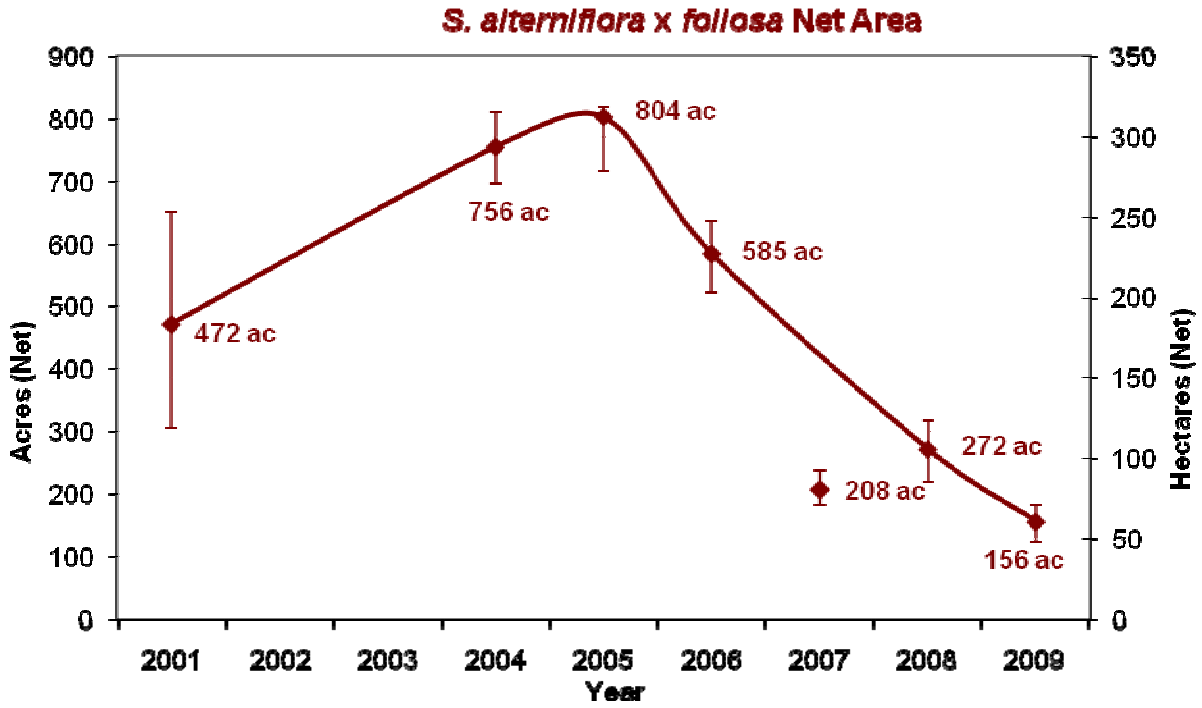


Figure 1.2. *Spartina alterniflora x foliosa* Net Area

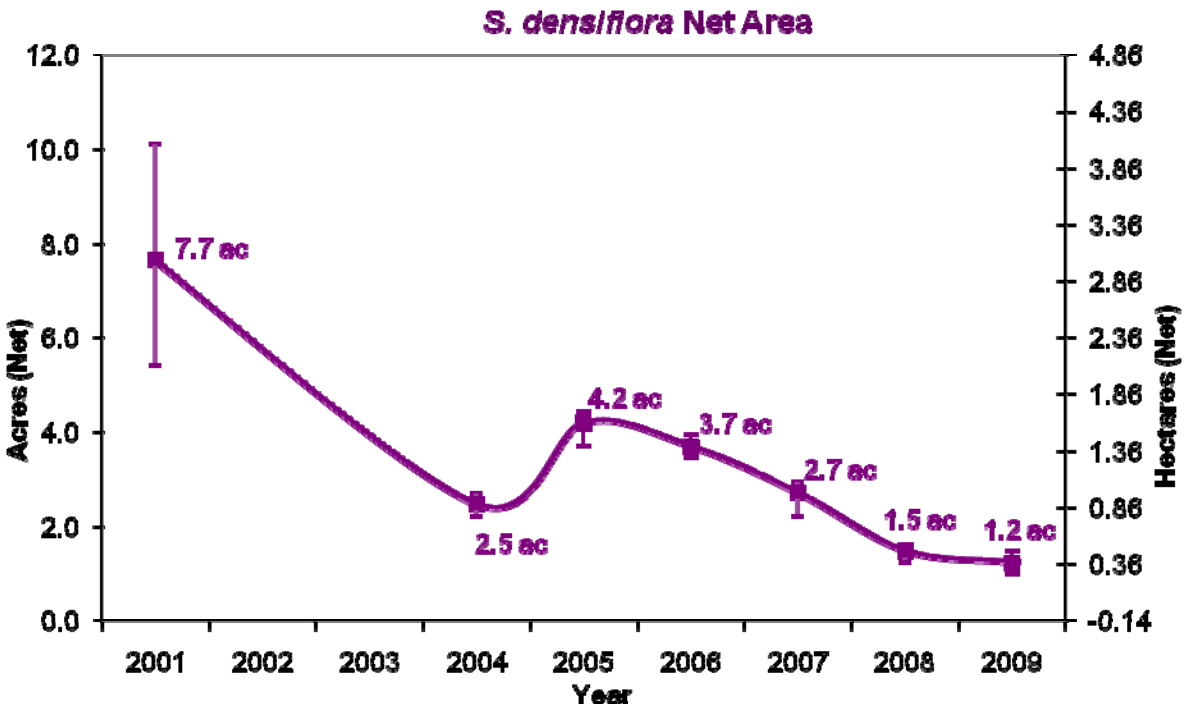


Figure 1.3. *Spartina densiflora* Net Area

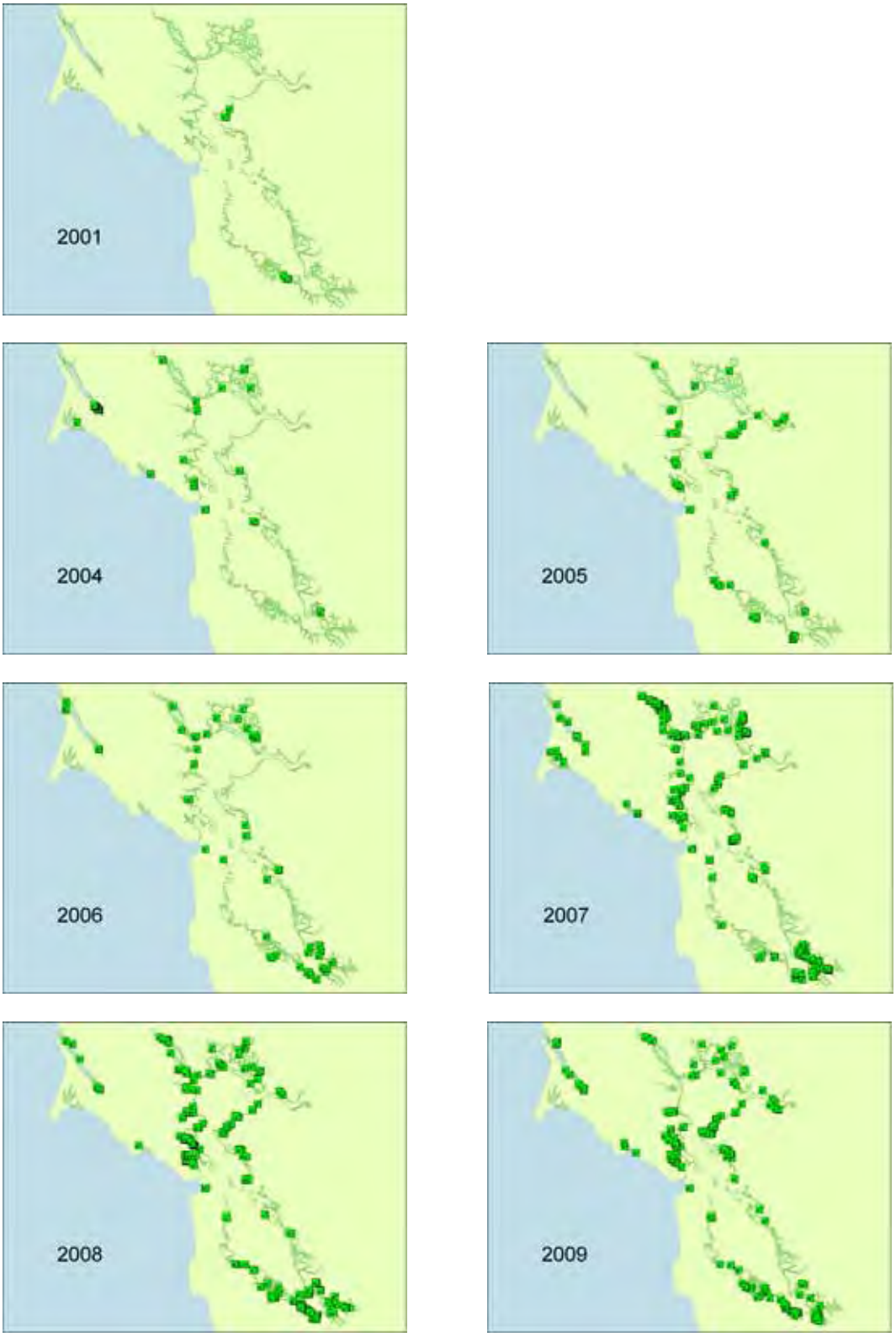


Figure 1.4. Locations of genetic samples field-identified by ISP biologists as *S. foliosa* and for which genetic test results diagnosed plants as pure *S. foliosa.*

Genetic results are based on RAPD testing from 2001-2008, and are based on microsatellite testing in 2009. Microsatellite results are analyzed with the software *structure* and those presented have evidence of ≥75% *S. foliosa* ancestry.

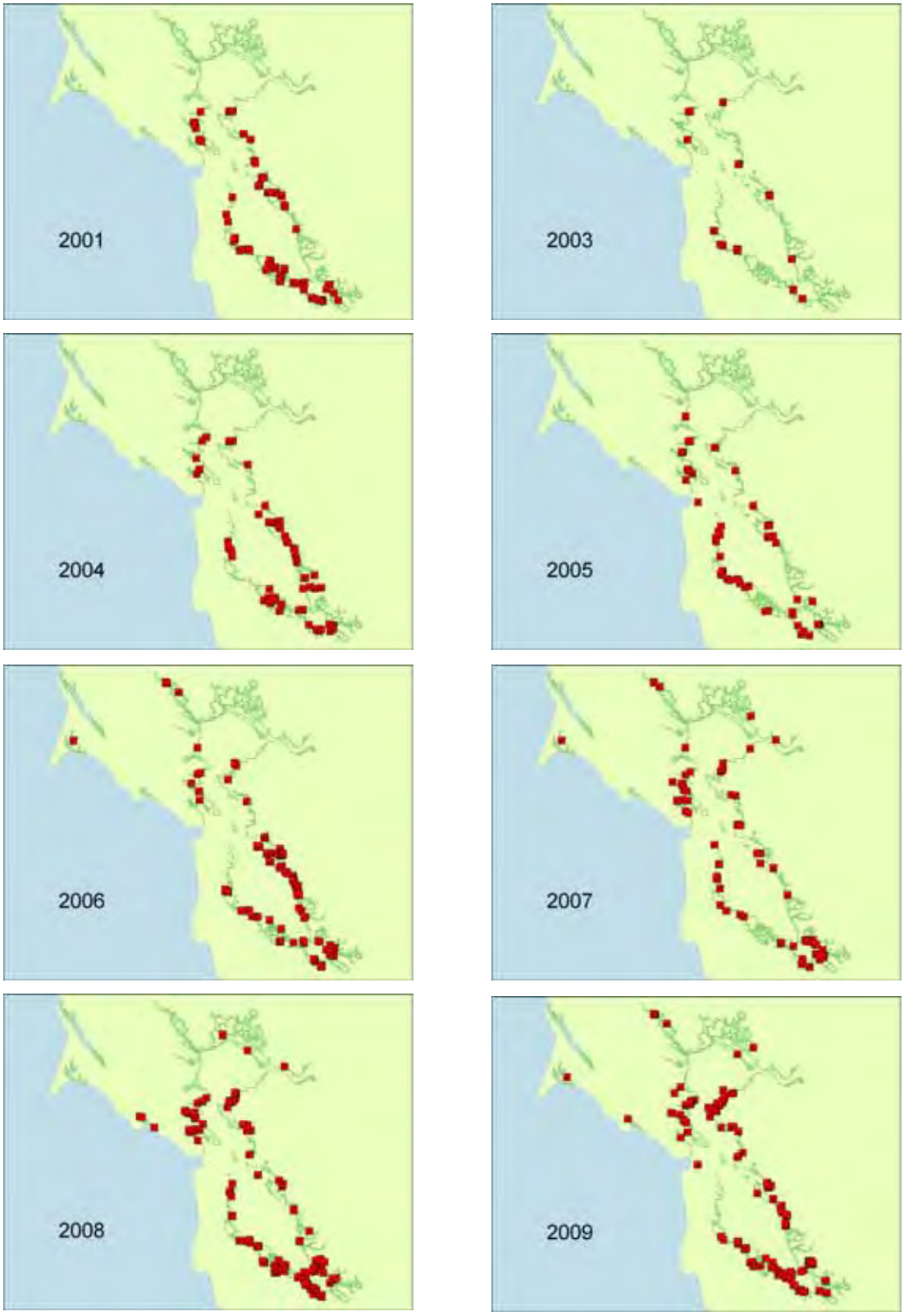


Figure 1.5. Locations of genetic samples field-identified by ISP biologists as hybrids that also had genetic evidence of hybridity.

Genetic results are based on RAPD testing from 2001-2008, and in 2009 are based on microsatellite results indicating >75% *S. alterniflora* ancestry.

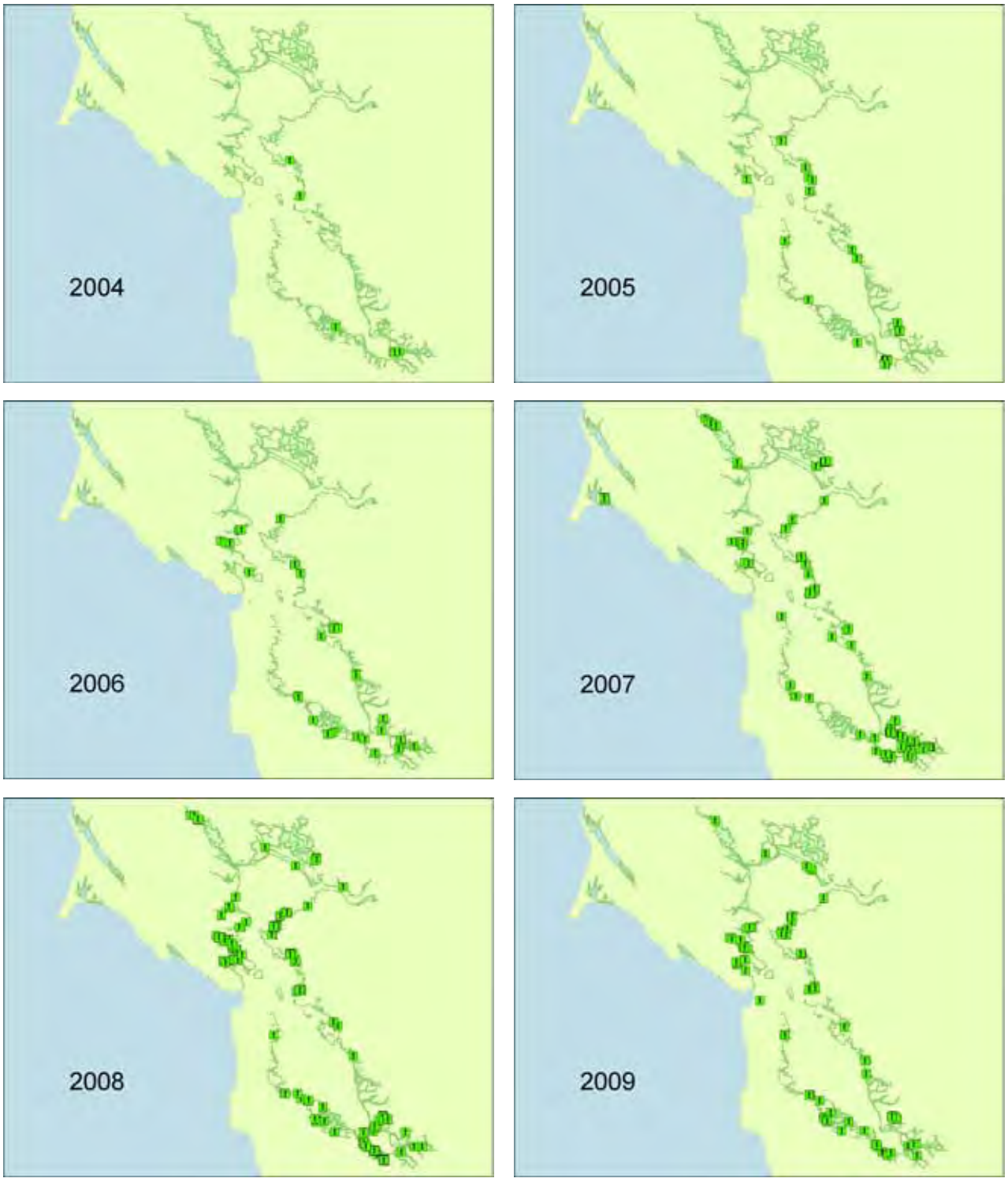


Figure 1.6. Locations of genetic samples field-identified by ISP biologists as hybrids for which genetic test results indicated no evidence of hybridity.

Genetic results are based on RAPD testing from 2004-2008, and are based on microsatellite testing in 2009. Microsatellite results are analyzed with the software structure and those presented have evidence of ≥75% *S. foliosa* ancestry.

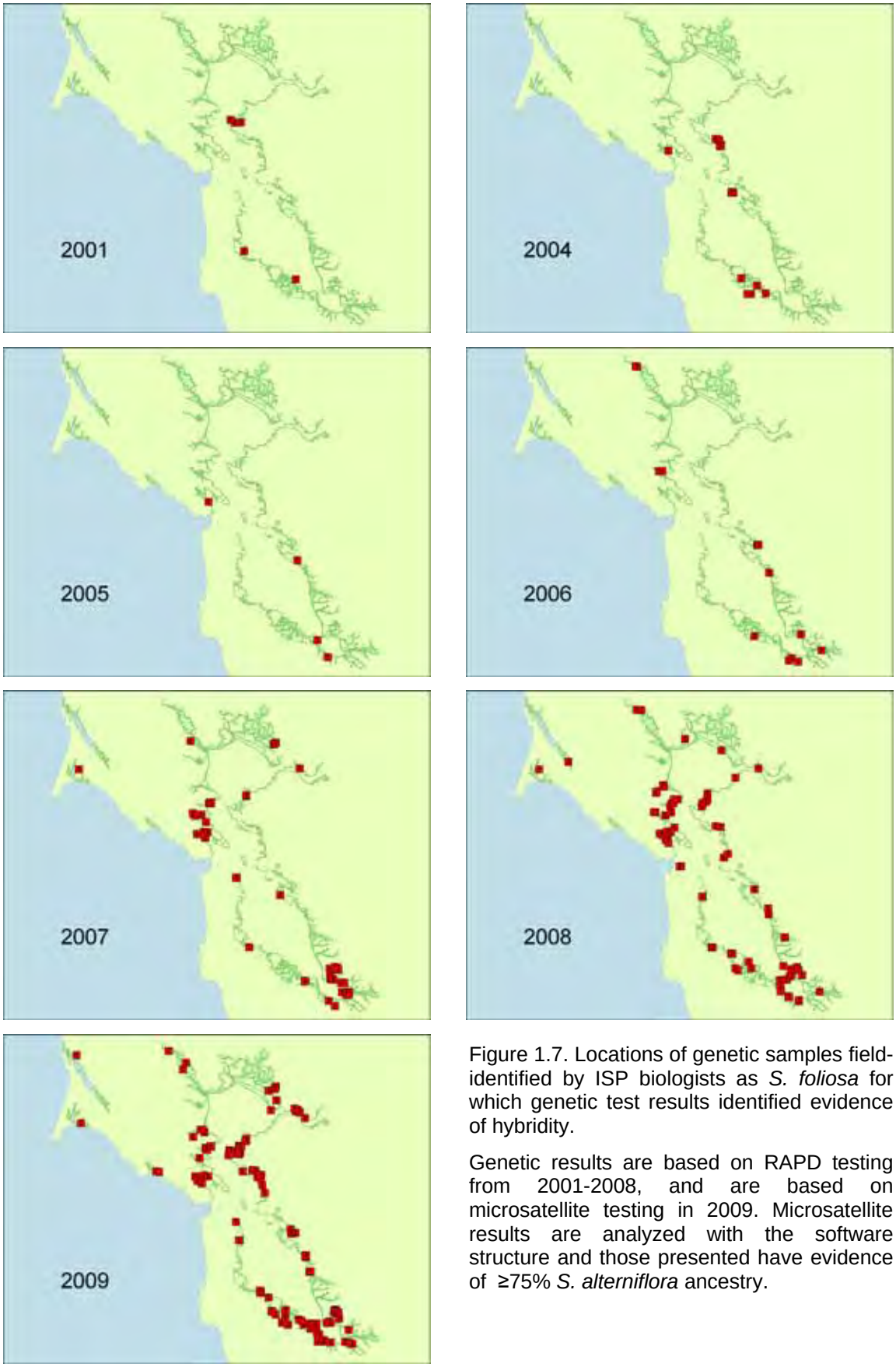
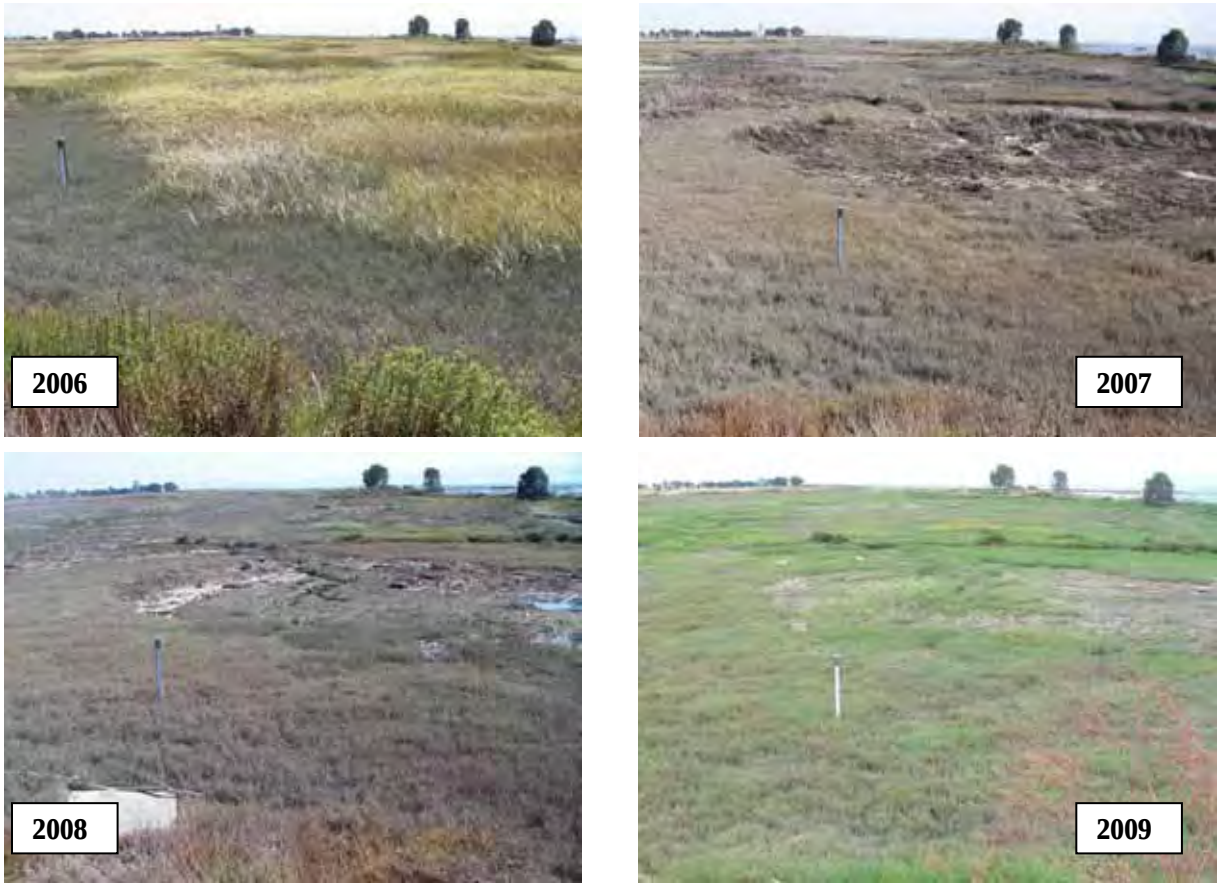


Figure 1.7. Locations of genetic samples field-identified by ISP biologists as *S. foliosa* for which genetic test results identified evidence of hybridity.

Genetic results are based on RAPD testing from 2001-2008, and are based on microsatellite testing in 2009. Microsatellite results are analyzed with the software structure and those presented have evidence of ≥75% *S. alterniflora* ancestry.

Figures II

Figure 2.1. Photo point locations. All photo point locations are displayed as black dots on the upper map. The lower map identifies those photo point locations highlighted in subsequent figures.



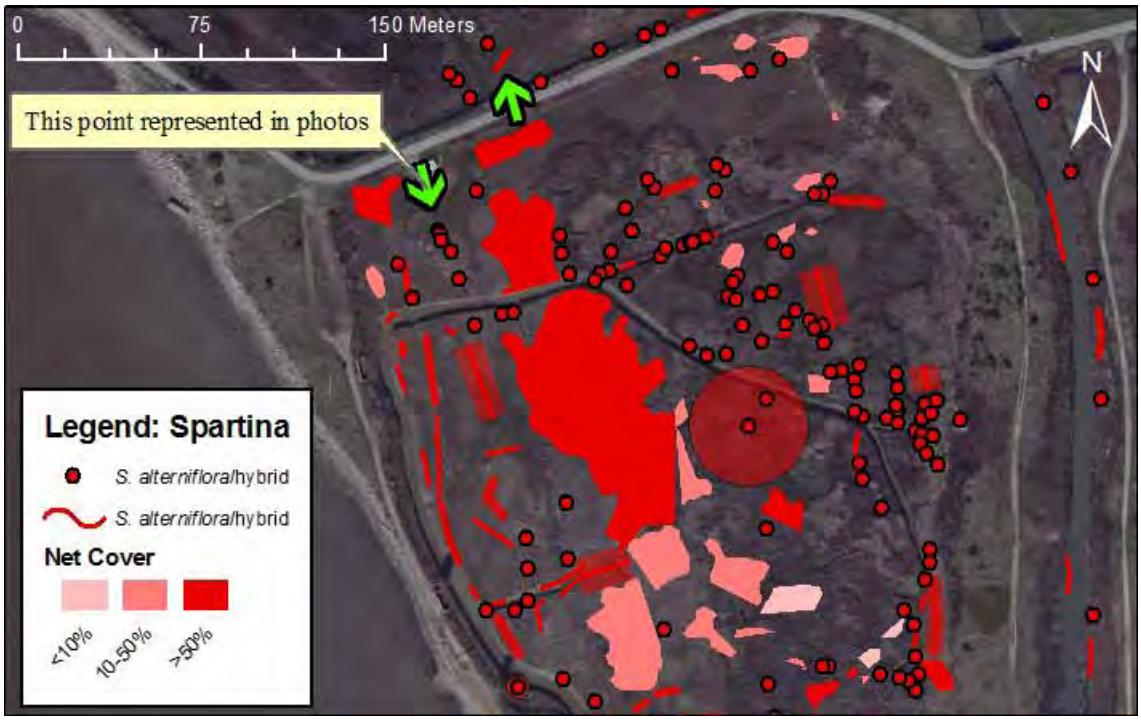


Figure 2.2. Subarea 20g: Bunker Marsh.

Annual photos and map of 2009 inventory results zoomed in to show location of hybrid Spartina in photos. Arrow represents photo point location and direction.

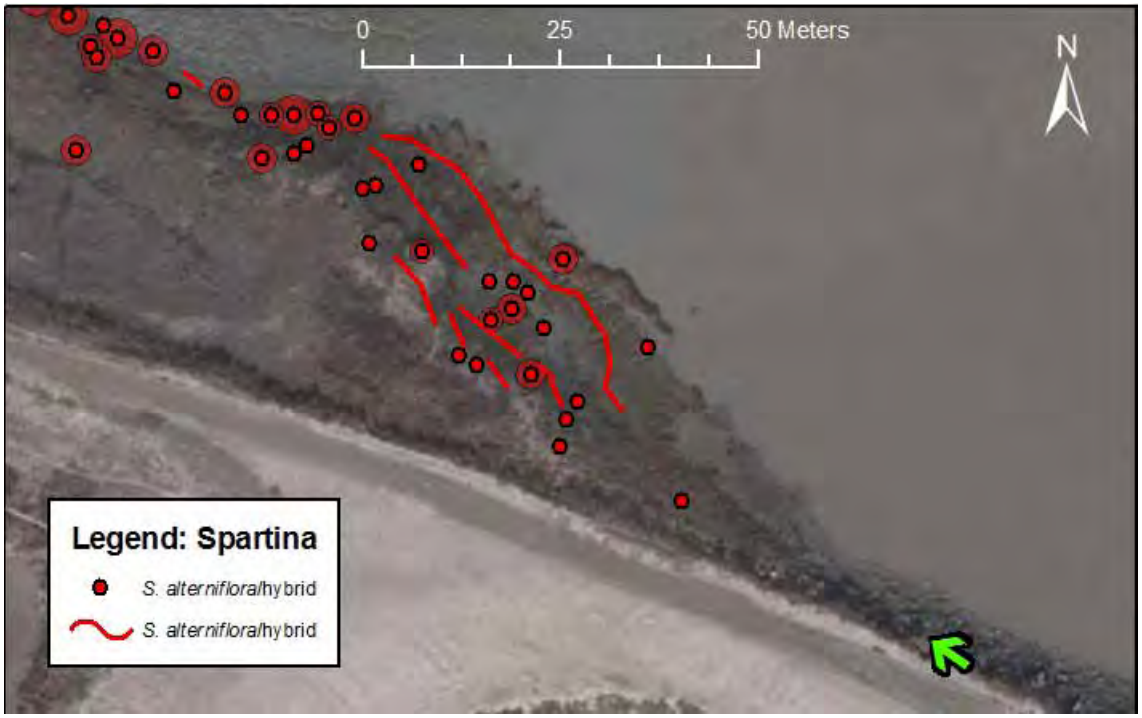


Figure 2.3. Subarea 8: Palo Alto Baylands.
Annual photos and map of 2009 inventory results zoomed in to show location of hybrid Spartina in photos just south of Hooks Island, along the Bay Trail. Arrow represents photo point location and direction.

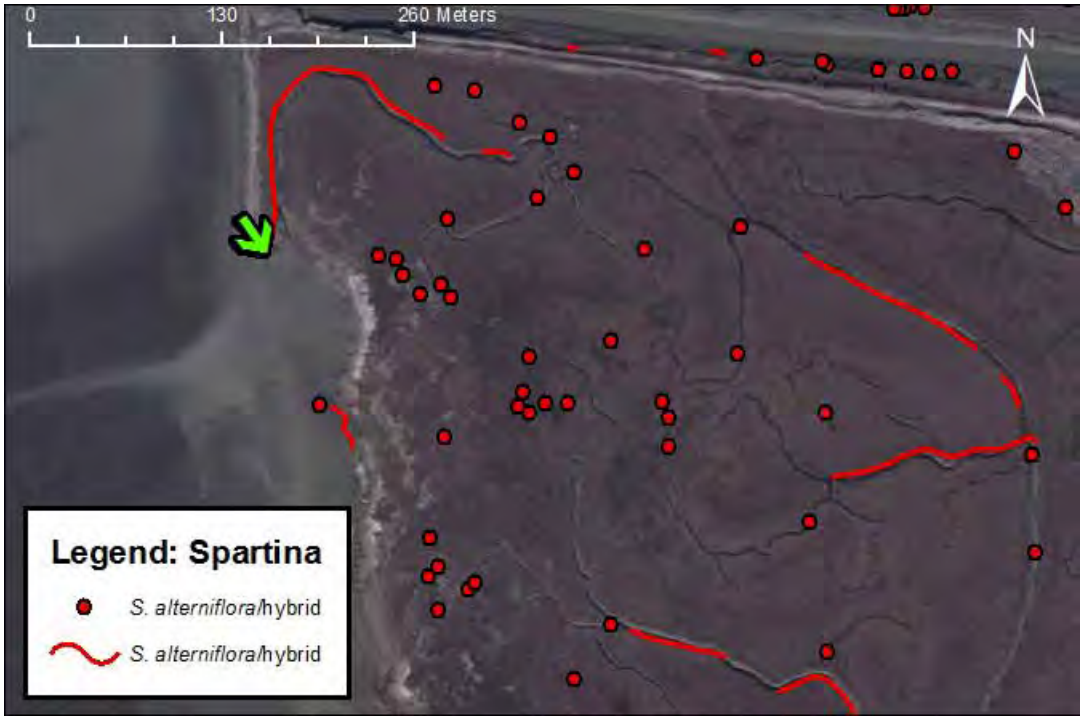


Figure 2.4. Subarea 13e:Whale's Tail South Fluke.

Annual photos and map of 2009 inventory results zoomed in to show location of hybrid Spartina in photos. Arrow represents photo point location and direction.

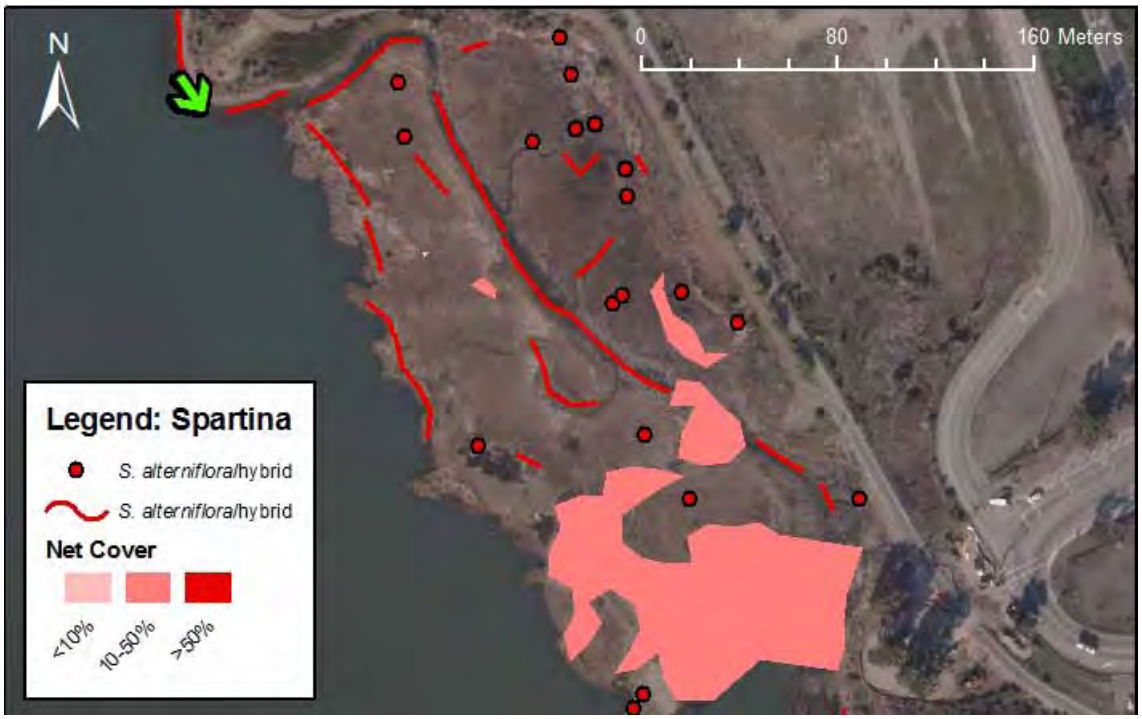


Figure 2.5. Subarea 17d: MLK Regional Shoreline.

Annual photos and map of 2009 inventory results zoomed in to show location of hybrid Spartina in photos. Arrow represents photo point location and direction.

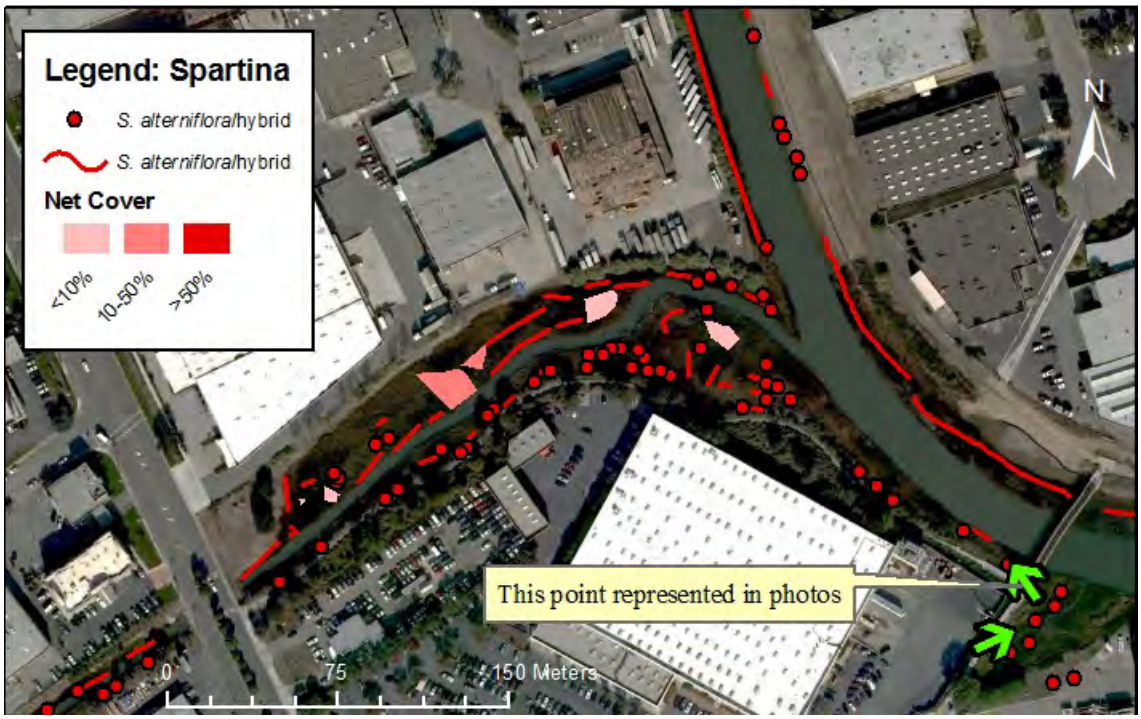


Figure 2.6. Subarea 18a: Colma Creek.

Annual photos and map of 2009 inventory results zoomed in to show location of hybrid Spartina in photos. Arrow represents photo point location and direction.

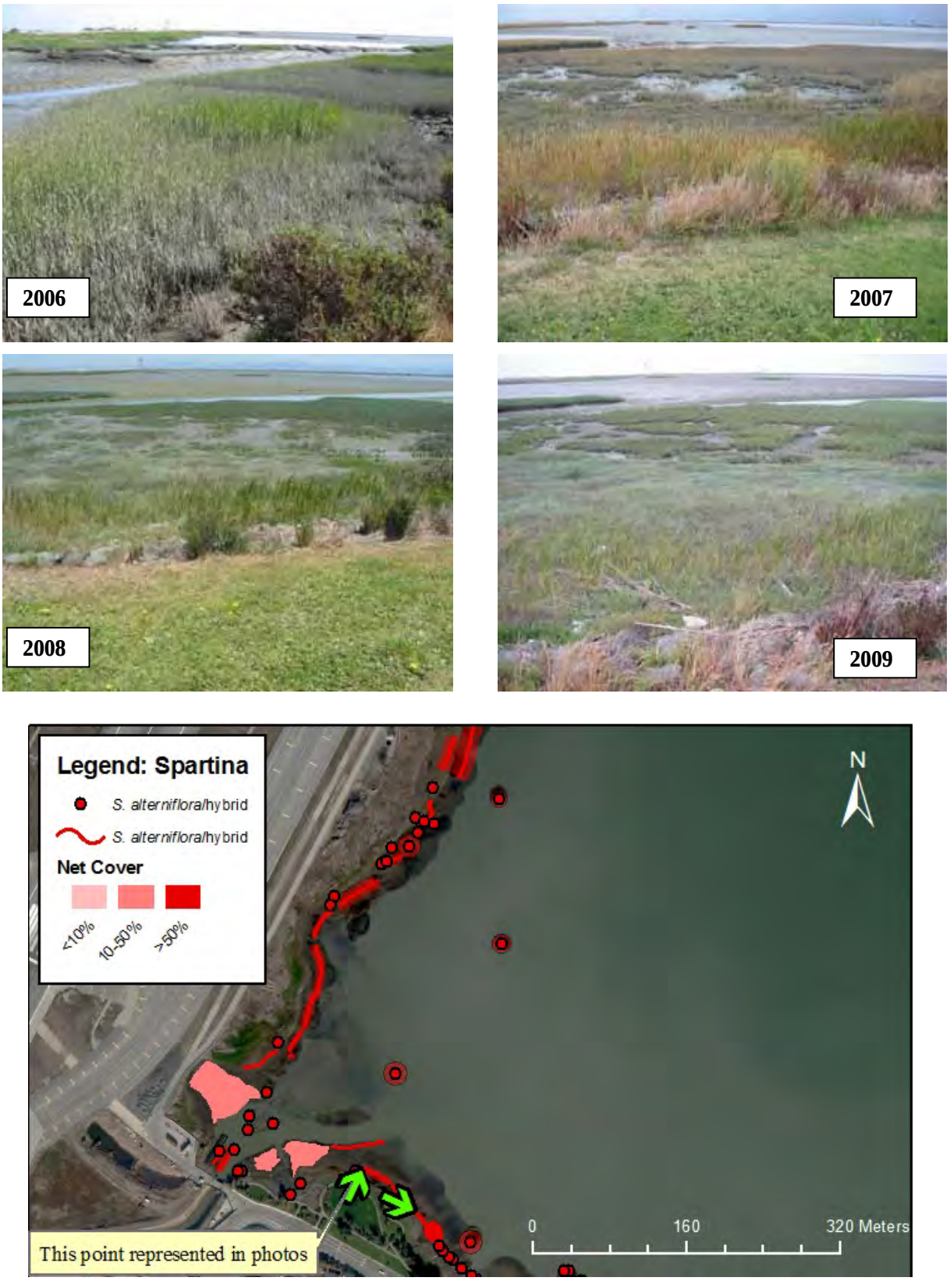


Figure 2.7. Subarea 19h: SFO Airport.

Annual photos and map of 2009 inventory results zoomed in to show location of hybrid Spartina in photos. Arrow represents photo point location and direction.

Figure 2.8. Subarea 19l: Burlingame Lagoon.
Annual photos and map of 2009 inventory results zoomed in to show location of hybrid Spartina in photos. Arrow represents photo point location and direction.