Summary Information

California State Coastal Conservancy

Monitoring for Invasive Spartina Control in the San Francisco Estuary

Amount sought: $1,651,396

Duration: 36 months

Lead investigator: Ms. Peggy Olofson, SF Estuary Invasive Spartina Project

Short Description

The proposed project includes annual regional surveys for non−native corgrasses (Spartina) in the San Francisco Estuary and outer coast marshes. It also will monitor marsh areas treated to control Spartina to determine if treatment was effective. A major research component of the proposal is the development of genetic markers to indentify particulaly invasive Spartina hybrid genotypes.

Executive Summary

The proposed project includes annual regional surveys for non−native corgrasses (Spartina) in the San Francisco Estuary and outer coast marshes. It also will monitor marsh areas treated to control Spartina to determine if treatment was effective. A major research component of the proposal is the development of genetic markers to indentify particulaly invasive Spartina hybrid genotypes.

All of this information will be used by the San Francisco Estuary Invasive Spartina Project Control Program to adapt it's control strategies and goals.

Proposal for the Ecosystem Restoration Program
2004 Monitoring and Evaluation Solicitation

Proposal Title:	**Monitoring for Invasive Spartina Control in the San Francisco Estuary**
Project Duration:	36 months
Lead Agency:	California Coastal Conservancy
Requested Funding:	$1,651,396

A. Project Description

1. Problem, Goals, and Objectives

The California Coastal Conservancy (Conservancy) initiated the San Francisco Estuary Invasive *Spartina* Project (ISP) with CALFED funding in 2000. The ISP is a regionally coordinated effort to arrest and reverse the spread of invasive cordgrasses (*Spartina* species) in the San Francisco Estuary, and to prevent their spread to outer-coast and upstream marshes. The geographic scope of the project currently includes the nine Bay-area counties, in those regions referred to as the South Bay, Central Bay, North Bay (a.k.a. San Pablo Bay), Suisun, and the Outer Coast (Figure 1). Under previous funding from CALFED and other sources[1], the ISP has completed the following:

- Established a strong program structure, including multiple partnerships with landowning and managing agencies
- Developed required programmatic NEPA and CEQA documents (Conservancy/USFWS 2003)
- Prepared for publication a detailed evaluation of the threat of non-native *Spartina* (Baye 2004)
- Initiated a proactive public education and outreach strategy, including presentations, brochures, and a bay-wide informational signage program
- Supported several research projects to provide necessary information for successful control work (Stralberg *et al.* 2004, Anderson 2004, Ayres *et al*. 2003, 2004, 2004),
- Initiated bay-wide inventory monitoring and mapping (Zaremba and McGowan 2004)
- Developed a regional strategy and site-specific plans for effectively controlling the *Spartina* invasion (Grijalva 2004)
- Coordinated permit acquisition for all projects, including programmatic and site-specific NPDES, Section 7, Section 106, and NEPA documents
- Planned and hosted a highly successful international conference focused on *Spartina* science and management (program and abstracts are available at www.the-conference.com/2004/spartina/).

[1] Funding for the ISP has also been provided by U.S. Fish and Wildlife Service (2001, $22,000), National Fish and Wildlife Foundation (2000, $101,500), National Oceanic and Atmospheric Administration (2000, $59,900), and the State Coastal Conservancy (ongoing, approximately $1,000,000 to date for program management and administration).

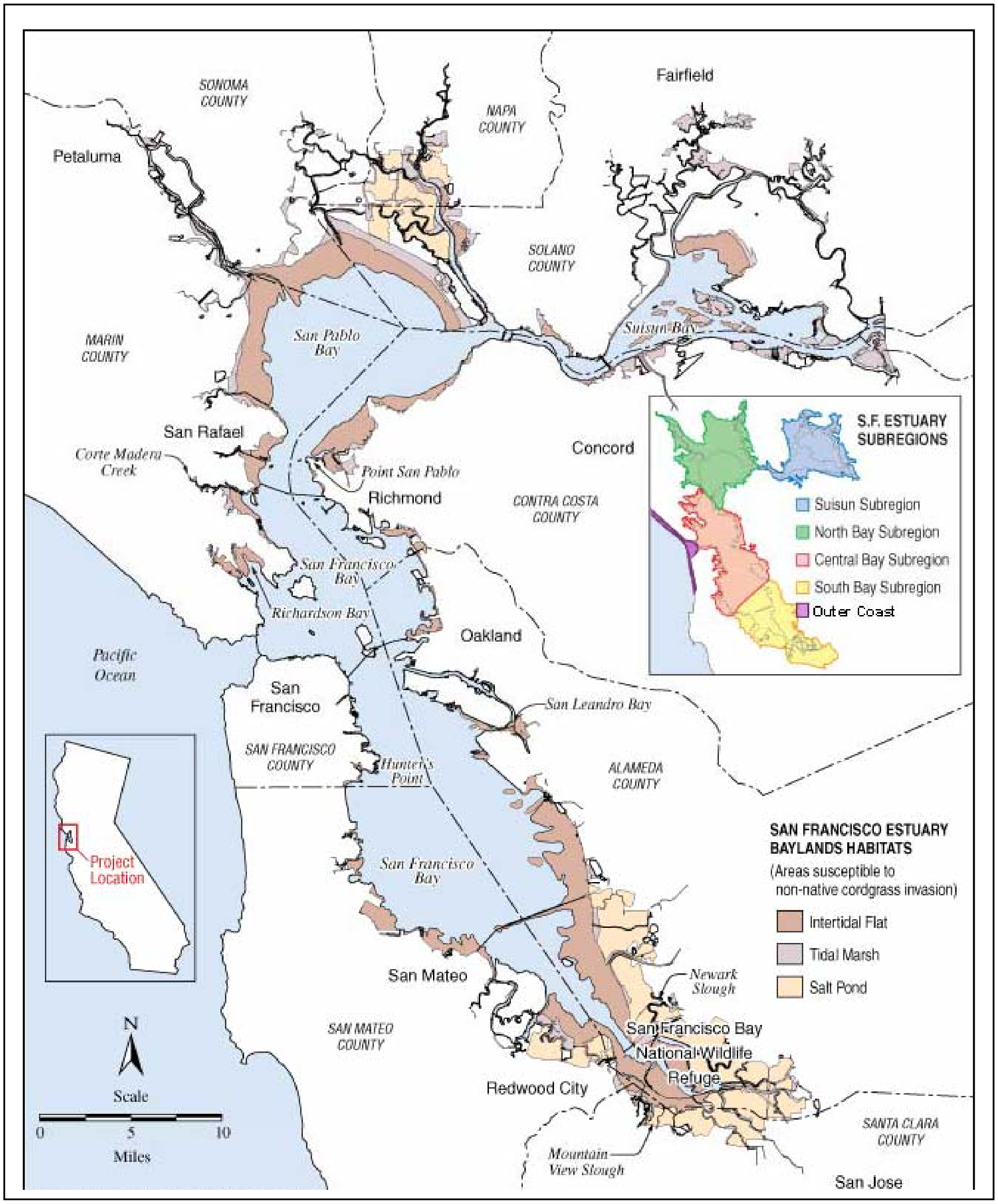

Figure 1. Geographic scope of San Francisco Estuary Invasive Spartina Project and the proposed monitoring program. Note: The scope of the *Spartina* Project may be extended further upstream of Suisun or along the Outer Coast if non-native Spartina is found to threaten those areas.

In late Summer 2004, the ISP launched full-scale implementation of its *Spartina* Control Program (SCP), with treatment of approximately 400 acres of non-native *Spartina* (25% of the estimated 2003 population). The SCP is now developing plans for the 2005, and expects to treat 2,000+ new acres, in addition to retreating, as needed, the sites treated in 2004.

During a special session at the international *Spartina* conference in November, the ISP received resounding positive peer-review of its strategy and methods by an international expert panel[2]. The panel urged immediate, rapid action to control *Spartina*.

Overview of Non-native Spartina in the San Francisco Estuary

Five species of *Spartina* are currently found in the San Francisco Estuary. Only one of these five species is native, *S. foliosa.* The four non-native species currently found in the Estuary are *S. alterniflora*, *S. densiflora*, *S. anglica*, and *S. patens*. One of the non-native *Spartina* species in particular, Atlantic smooth cordgrass or *S. alterniflora*, and its hybrids (formed when this species crosses with the native Pacific cordgrass, *S. foliosa*) are now threatening the ecological balance of the Estuary and are likely to eventually cause the extinction of native Pacific cordgrass, choke tidal creeks, dominate newly restored tidal marshes, and displace thousands of acres of existing shorebird habitat (Callaway and Josselyn, 1992; Cohen and Carleton, 1995; Daehler and Strong, 1996; Ayres *et al.* 2003; California Coastal Conservancy 2003; Ayres *et al.* 2004). Once established in this Estuary, invasive cordgrasses could rapidly spread to other estuaries along the California coast through seed dispersal on the tides (Ayres *et al.* 2003 and 2004). A detailed review of the potential impacts of the *S. alterniflora* hybrids is provided in the ISP's newly completed report, "*A Review and Assessment of Potential Long-term Ecological Consequences of the Introduced Cordgrass* <u>*Spartina alterniflora*</u> *in the San Francisco Estuary*" (Baye 2004).

A major issue complicating the control of non-native *Spartina* in the San Francisco Estuary is the reciprocal hybridization of *S. alterniflora* with the native *S. foliosa*. This has resulted in a phenomenon known as a "hybrid swarm," which has accelerated the spread of extremely robust, highly adaptable invasive plants to super-exponential rates (Zaremba 2004).

An extensive bay-wide survey in 2001 identified a total of 483 net acres of non-native *Spartina* distributed throughout nearly 40,000 acres of tidal marsh and 29,000 acres of tidal flats of the Estuary (Ayres *et al.* 2004).. This included 470 acres of *S. alterniflora*-hybrids; 13 acres of *S. densiflora;* 0.58 acres of *S. patens;* and 0.09 acres of *S. anglica*. In 2003, a limited survey of a subset of sites (Zaremba and McGowan 2004) determined that the net area of non-native *Spartina* had quadrupled, and there were a total of 2,000 acres, with the greatest rate of spread found in the S. *alterniflora* hybrids. Another bay-wide

[2] Including Dr. Alan Gray, Center for Ecology and Hydrology, U.K.; Dr. Donald Strong, University of California, Davis; Dr. Shuqing An, Nanjing University, China; Dr Mark Burtness, Brown University, Rhode Island; Dr. James Morris, University of South Carolina; Dr. Kim Patten, Washington State University; Dr. Paul Hedge, National Oceans Office, Australia; Dr. Malika Ainouche, Universite de Rennes Botanique, France, and Dr. Bo Li, Fudan University, China.

survey was completed in November 2004, but the data has not yet been analyzed to determine rate of spread.

Goals and Objectives of the ISP Control Program

The goals of the ISP Control Program are to eradicate all non-native *Spartina* species and hybrids from the San Francisco Bay and to prevent their spread to the outer coastline, Suisun, and the Delta. The over-arching objective of this work is to preserve and restore native habitat to support a functioning estuarine ecosystem. Underlying this objective is the critical need to eradicate non-native *Spartina* to prevent it from invading and dominating the planned tidal restoration areas of the South Bay Salt Ponds Restoration Project.

The goal of eradication is complicated in relation to the hybrids, which display a wide range of physical characteristics, some very similar to native *S. foliosa*. It is unclear at which point the hybrids can be considered effectively "eradicated," as genetic dilution of native *S. foliosa* will likely have occurred in some local *S. foliosa* populations, and there will likely continue to be latent genetic strains of *S. alterniflora* present throughout most invaded areas. In response to this, the ISP is proposing to remove 100% of *Spartina* at known hybrid-invaded sites, and then to continue to monitor for and remove *Spartina* clones and populations that display discernable hybrid characteristics. This proposal includes research to assist, and likely improve upon, this approach by developing genetic Invasive Marker Profiles (IMPs) for hybrid *Spartina*.

Based on our current understanding of the rate of spread of non-native *Spartina* (super-exponential, perhaps doubling every year), our available control techniques, and regulatory constraints, we estimate that we will have achieved some level of control at all known *Spartina*-invaded sites within two seasons (by the end of 2006), and should have approximated "eradication" within four seasons (by the end of 2008).

2. Justification

There are three primary components of the proposed monitoring and research program:

1. *Spartina* Monitoring - monitor various aspects of non-native *Spartina* populations, including rate of spread, area covered, effectiveness of treatment, and recruitment of seedlings;
2. Invasive Marker Profiling - To develop an approach for identifying particularly invasive *Spartina* hybrid genotypes
3. Clapper rail monitoring - To determine presence of the endangered California clapper rail at sites slated for treatment; and

Each of these is intended to provide critical information and tools to support development and refinement of the ISP's *Spartina* control strategies. The mechanisms for incorporating this information into are shown in Figure 2. The *Spartina* and clapper rail monitoring components are relatively simple visual surveys that determine the presence or absence of the subject. The theories related to the development of Invasive Marker Profiles are considerably more complex, stemming from the dynamics of invasive *Spartina* (Figure 3). Additional details of this work follow.

Figure 2

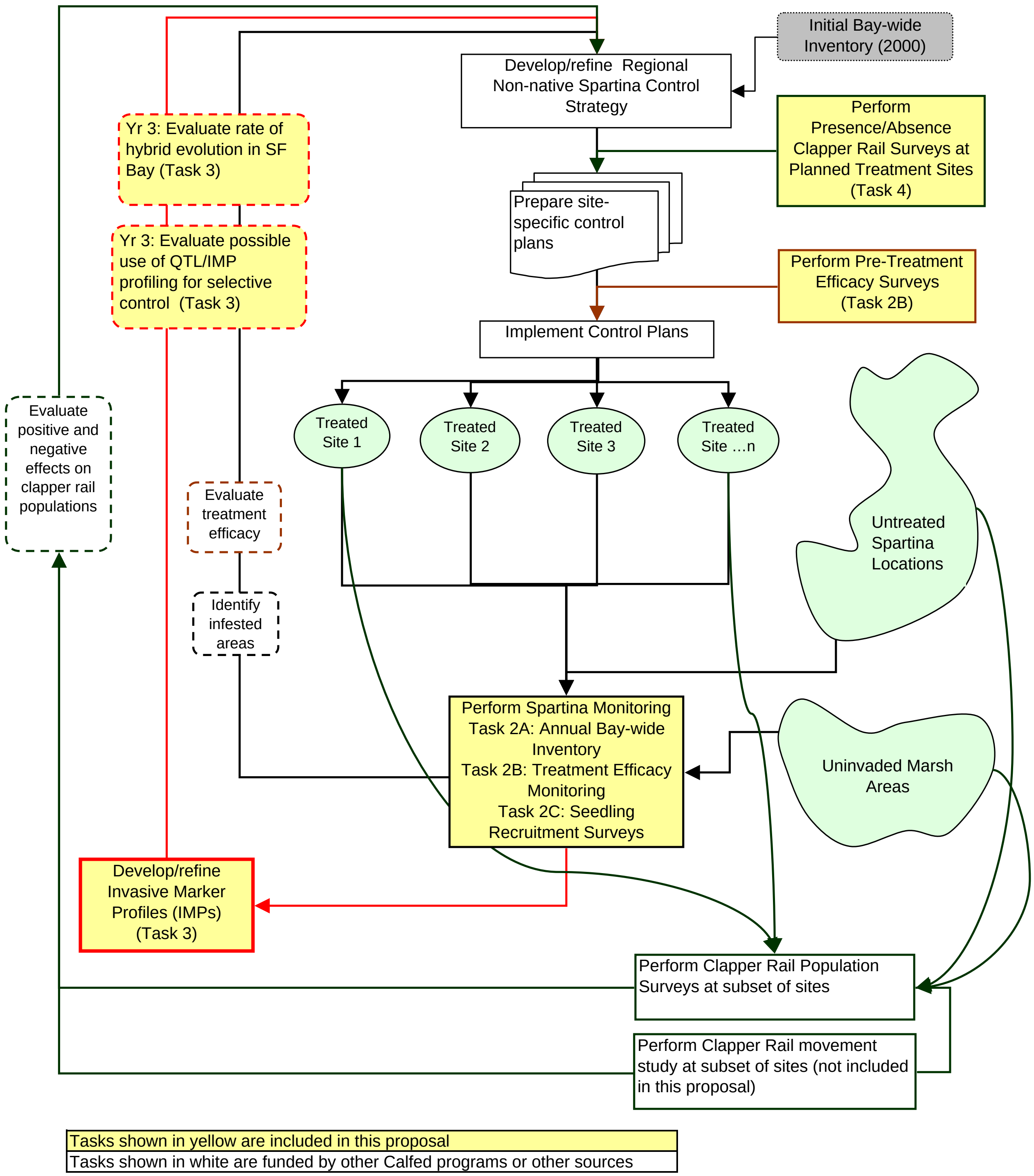

Figure 2. Adaptive Management Model for San Francisco Estuary Invasive Spartina Project Control Program

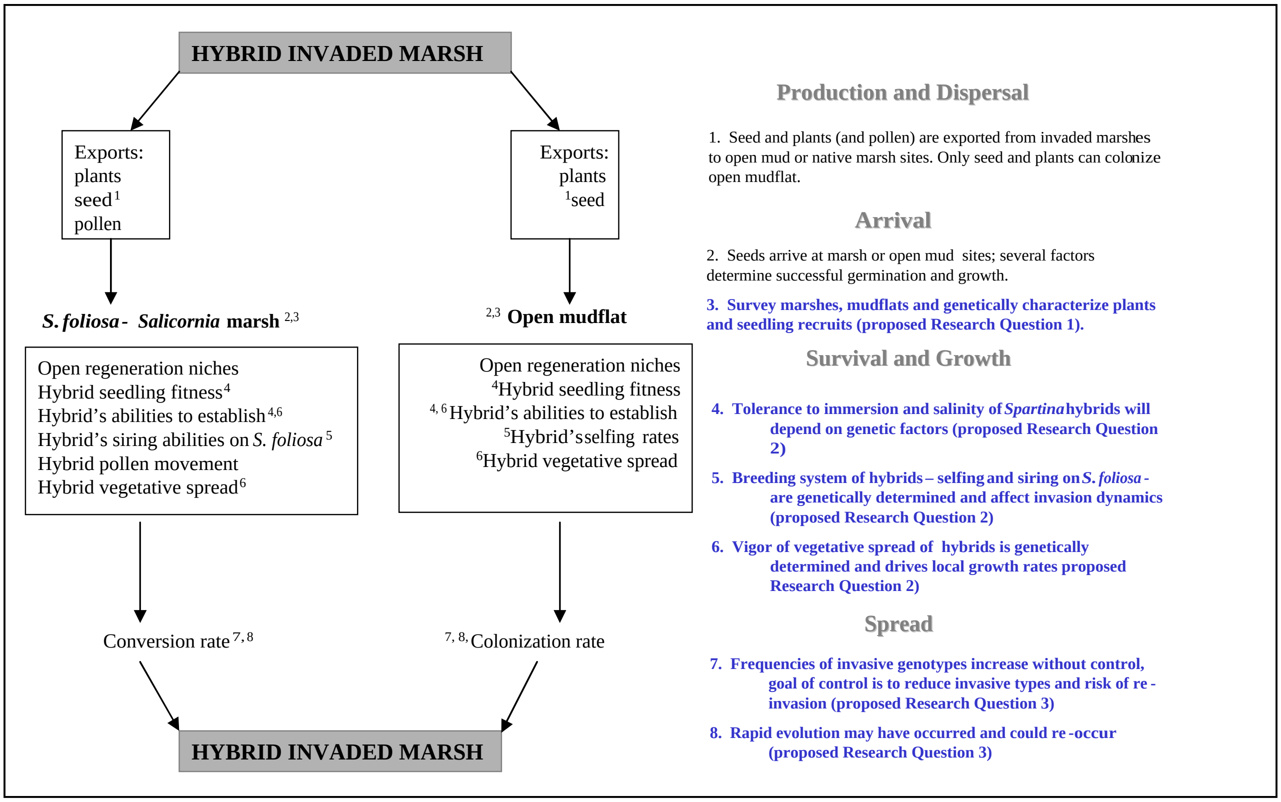

Figure 3. Conceptual model of invasive dynamics of hybrid cordgrass, from seed production and dispersal to marsh invasion and relationship to proposed research. Processes are numbered in superscripts. Proposed research questions are in bold (and blue). (Model developed by D. Ayres, UC Davis)

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Background for Task 3 – Development of Invasive Marker Profiles for S. alterniflora hybrids (written by Dr. Debra Ayres, UC Davis)

Spartina alterniflora, smooth cordgrass, endemic to the eastern U. S., was introduced into the range of a native congeneric species, *S. foliosa*, California cordgrass, in south San Francisco Bay ca 30 years ago (US Army Corps of Engineers 1978). In previous research, using Random Amplified Polymorphic DNA (RAPD) markers and chloroplast DNA sequences, we determined that extensive hybrid swarms have arisen through reciprocal hybridization (Ayres et al, 1999; Antilla 2000). The primeval condition of Pacific estuaries leaves vast expanses of open mud in intertidal habitats upon which animals native to the Pacific rely. Both *Spartina* hybrids and *S. alterniflora* can invade this open mud, modify the hydrology, and threaten the native biota. Hybrid *Spartina* and *S. alterniflora* also invade marshes dominated by *Salicornia virginica* (pickleweed) and *S. foliosa*. *Spartina foliosa* is virtually absent in salt marshes where *S. alterniflora* and their hybrids were deliberately planted; we found roughly equal numbers of *S. alterniflora* and hybrid individuals (Ayres et al. 1999).

Researchers from the University of California, Davis and Conservancy mapped the location and extent of all *Spartina* hybrids and *S. alterniflora* in the estuary in 2001 (Ayres et al. 2004). Radiating from sites of deliberate introduction, *S. alterniflora* and hybrids covered ca 190 ha, slightly less than 1% of the Bay's tidal mudflats and marshes, mainly in the South and Central Bay. Estimates of rate of areal increase over the past 25 years show an accelerating rate of increase in cover by cordgrass hybrids. Surveys in 2003-04 show that hybrid expansion is still accelerating (K. Zaremba, CCC, 2004) . This accelerating growth rate could be due to the proliferation of hybrid plants capable of rapid expansion, having superior seed set and siring abilities and/or having high tolerance to salinity and tidal innundation. We have found plants with these exceptional abilities growing in nature (Ayres et al, submitted; Pakenham- Walsh, 2003; Zaremba, 2000). *Spartina* seed floats on the tide, raising the possibility that this invasion will be spread throughout the San Francisco estuary, and exported to estuaries beyond the Golden Gate. We found isolated plants of *S. alterniflora* in outer coast estuaries north of the Bay suggesting the likelihood for the San Francisco Bay populations to found others on the Pacific coast.

The emerging view, based on our genetic and ecological research, is that plants of exceptional growth and reproductive characters have originated through hybridization and are being favored by natural selection. Through this ongoing positive feedback process ever more invasive plants will be generated and selected, which is in contrast to conventional views of weed science and management based on species with narrow evolutionary potentials spreading in predictable ways. The threats posed by hybrid cordgrass, therefore, far exceed those posed by *S. alterniflora* alone. This process has already created widespread risk to the ecosystem from structural changes due to conversion of open tidal flats and native marsh to elevated cordgrass meadows (Callaway, 1990; Callaway and Josselyn, 1992; Daehler and Strong, 1996). These structural changes may lead to severe reductions in shorebird feeding habitats (Strahlberg et al, 2004), and alterations of the habitats of federally listed California Clapper Rails and Salt-Marsh Harvest Mice (summarized in Baye, 2004). Further, we have predicted the eventual Bay-wide, and perhaps global extinction of native *S. foliosa* due to genetic assimilation and replacement of *S. foliosa* by competition with robust hybrid cordgrass (Ayres et al, 2003; Ayres and Strong, in press). As a result of our findings, the control strategy for these hybrid invaders has been changed from containment to possible eradication (California Coastal Conservancy, 2003).

We propose to monitor the genetic background of cordgrass plants and seedlings during active control efforts (Phase I) using molecular techniques we developed and used to identify, characterize, and track *Spartina* hybrids (Ayres et al, 1999; Ayres and Strong, 2001; Ayres et al 2003; Ayres et al 2004). Many potentially invasive hybrids have traits such as high seed production or salinity tolerance that may not be immediately apparent; that is, they do not display the visible hybrid traits of tall stems and wide lateral expansion. As well, it is generally impossible to distinguish hybrids in early seedling populations. Cryptic hybrids and seedlings are readily distinguished by our genetic techniques. We propose to provide genetic surveys of established and recruiting populations of cordgrass to CCC's Invasive Spartina Project to monitor the progress of the invasion and to evaluate the efficacy of control during Phase I control efforts (short-term monitoring).

We also propose to develop new molecular tools that would establish a link between microsatellite DNA markers (Blum et al 2004, Sloop et al, in preparation) and specific invasive traits to identify plants with Invasive Marker Profiles (IMPs). Marker-trait associations will allow us to identify and categorize hybrids as invasive (IMPs) or non-invasive (non-IMPs) genotypes for priority eradication during active Phase I control, to track the evolutionary dynamics of hybrid cordgrass pre- and post active control to determine the efficacy of control efforts in slowing the proliferation of invasive genotypes (IMPs), and to assess control success during active salt-marsh restoration in the coming years (Phase II control). The use of this new tool may allow us to reverse the positive feedback process described above by imposing stringent artificial selection on the invasive genotypes driving the invasion. As the total elimination of all hybrid cordgrass in the estuary would likely be difficult and expensive, this could represent a substantial cost savings, in addition to reducing the risk of re-invasion. Our data and evolutionary theory predict that the invasion cycle can begin again if highly invasive genotypes re-invade.

Conceptual Model of hybrid Spartina spread

The UC Davis researchers identified the hybridization of native California cordgrass with Atlantic salt marsh cordgrass (Daehler and Strong 1996) and have continued this research investigating the rates and mechanisms of hybrid spread. In our model (Figure 3), the hybrid invasion advances with the production of hybrid seed from invaded marshes which disperses to regeneration sites in the open mud of tidal flats or restoration sites, and established native marshes, where the survival and growth of some hybrids are superior to native *S. foliosa*.

Once hybrid seed arrives at a bare patch within a marsh or on open mudflat, its survival, establishment and spread will depend on the plant's ability to tolerate tidal immersion, ambient salinity, and biotic competition (#2, 4 - 6, Figure 3). The abiotic tolerances of a range of hybrid genotypes are being investigated in common-garden environments in ongoing research (Ayres and Strong, in progress) where we have found clear evidence of a subset of extremely fit hybrid genotypes. The key traits are high vegetative growth rate and lateral expansion in the mudflat environment and high tolerance to salinity in pickleweed terrace gardens.

Single isolated hybrids grow below the tidal range of the native cordgrass and some of these may be able to self-pollinate (#5, Figure 3). We have found striking differences between hybrids in ability to self-fertilize, with some hybrids far exceeding both parental species in self-pollinated seed production and the vigor of their inbred progeny (Sloop et al, in preparation). High self-fertilization ability confers great selective advantage on population founders.

In a native marsh, siring abundant seed in surrounding *S. foliosa* plants confers high fitness on invading hybrids (#5, Figure 3). Interestingly, we found few putative F1 hybrids in nature (Ayres and Strong unpublished) and F1 hybrids are almost impossible to make in the laboratory. This leads us to propose that the initial interspecific hybridization, the production of F1s from crosses of the two parental species, is rare in nature (Ayres et al submitted). We have also found little temporal overlap in flowering between *S. foliosa* and *S. alterniflora*, while hybrids overlap in flowering with both parental species. Our preliminary data shows that some hybrids can sire the lion's share of seed in surrounding *S. foliosa* plants (Ayres et al, in review). This observation is consistent with the results of our chloroplast DNA study where the ratio of *S. foliosa* cpDNA to *S. alterniflora* cpDNA in hybrids was 2:1 (Anttila et al. 2000) indicating that *S. foliosa* has excelled as a seed parent to hybrids.

Production of hybrid seed accelerates with time after marsh invasion. We found there were disproportionately large numbers of hybrid seedlings recruiting in the open mud at a minimally (5% hybrid cover) invaded marsh in San Lorenzo (23% hybrid seedlings) and a moderately (50% hybrid cover) invaded marsh at San Mateo (83% hybrid seedlings).

Our previous and ongoing research suggests a strong genetic component to potentially invasive traits: 1) seedling vigor, which favors survival and establishment of new plants; 2) height, which may confer competitive dominance over *S. foliosa* and is correlated to survival at lower tidal elevations; 3) rate of lateral spread, which anchors plants to the mud, and results in an overall increase in area covered and in flowering culms; 4) pollen production and vigor, which may result in greater backcrossing on *S. foliosa* and increased production of hybrid seed from native plants; 5) self-fertilization rate, which allows single plants to found new populations; and 6) salinity tolerance, which enables plants to survive and reproduce at higher elevations in the marsh, enlarging the potential ecological range of cordgrass hybrids. Some hybrids exceed both parental species in some or all of these traits and we infer that the proliferation of these invasive traits has been and will be driven by natural selection. On the other hand, some hybrids are inferior to the parental species and would likely have limited ability to invade.

The final steps in the dynamics of hybrid *Spartina* invasion, spread, and ultimately control is to monitor the frequencies of invasive and non-invasive genotypes in marshes and mudflats to assess control efficacy (#7, Figure 3) and to determine the rate of evolution in invasive hybrids over time to evaluate the potential for rapid re-invasion of hybrids should control efforts fail (#8, Figure 3).

We anticipate that Phase I genetic monitoring techniques (RAPD-based) will be supplanted by the more refined and predictive monitoring based on IMP-based genetic tools. As the focus in the Bay turns toward active marsh restoration in 2008 (e.g. South Bay Salt Pond Restoration Project), it is important to “determine the rates of invasion in newly restored marshes”; whether there is a “low - level” population size or distribution”.. *or genetic make-up of hybrid cordgrass*.. “that can be sustained without adverse impact on the natural environment”; whether there are “other mechanisms in restoration design that can limit invasion” and “biological controls that can be developed to effectively limit invasive species”, *such as stringent selection against invasive hybrid genotypes*; and to develop monitoring tools “to effectively detect invasive species prior to their becoming a problem in the environment” (quotes from South Bay Salt Pond Draft Science Plan, 2004, page 25; italics are ours; (http://www.southbayrestoration.org/pdf_files/national_sci_panel/03NSP_SciPlan_Oct0104.pdf). The proposed research thus directly addresses key questions and uncertainties affecting future tidal marsh restoration in the Bay.

Task Objectives

1. To use molecular genetic techniques to identify cryptic hybrids in minimally invaded marshes; to assess cover and presence of hybrids during Bay-wide surveys; and to identify hybrid seedlings colonizing seed safe sites in established and restored marshes. These assessments would be used to inform control efforts and to track control efficacy during Phase I control.
2. To determine whether there are specific quantitative traits (QTLs) that correlate with molecular markers to genetically characterize highly invasive individuals and build molecular tools to identify potentially invasive plants in natural popula-

tions in the Bay. Hybrid plants with Invasive Markers Profiles (IPMs) could be targeted for timely eradication. This technique would be available for use toward the end of Phase I control efforts to target invasive genotypes for immediate removal, and to screen seedlings colonizing marshes newly restored under the South Bay Salt Pond Restoration Program to determine whether these marshes will become dominated by native (or native-like) vegetation or cordgrass hybrids.

3. To assess whether the invasion can be controlled by artificially removing invasive genotypes. According to evolutionary theory, and our data on spread, we hypothesize that the accelerating rate of hybrid populations is due to natural selection favoring invasive genotypes. When plants with IMPs are removed, will the frequency of invasive plants, and thus the invasion rate, decline?
4. To evaluate the rate of evolution of hybrids in the Bay by comparing frequencies of IMPs in plant DNA collected in 1997-98 (pre-control), in 2005-08 (during active control), and after Phase I control is complete to evaluate trends in the evolution of hybrids, how quickly natural selection has favored plants with IMPs, and the effect of control on the frequencies of IMP- and non-IMP- hybrids in Bay marshes. We will then be able to address the question of future risk to the estuary should re-invasion by IMP-containing hybrids occur.

3. **Previously Funded Monitoring**

The Invasive *Spartina* spp. distribution and abundance monitoring began in 2000 with the establishment of the San Francisco Estuary Invasive Spartina Project (ISP). The first 2000-2001 Invasive Spartina Project monitoring program surveyed 69,403 acres of tidal marsh and mudflat of the San Francisco Bay Estuary with the goal to determine the distribution and abundance of the invasive non-native cordgrass, *Spartina* species present in the Estuary. Four non-native *Spartina* species were known to exist in the Estuary. These include *S. alterniflora* and its hybrids with native *S. foliosa*, *S. densiflora*, *S. anglica*, and *S. patens*. The ISP monitoring program includes *Spartina* treatment site monitoring to determine the control efficacy, and mapping of ongoing genetic surveys to confirm field identification of the *Spartina* hybrids. The results of the inventory (distribution and abundance), treatment efficacy and genetic survey monitoring are applied to the conceptual model used to guide, and to adapt accordingly, the ISP Control Program strategy.

2001 Monitoring Results

The 2001 inventory survey found 13 net acres of *S. densiflora*, less than acres of *S. anglica* and *S. patens* and 470 net acres of *S. alterniflora*-hybrids. The *S. alterniflora*-hybrids were the dominant *Spartina* invader with populations as far North as Pt. Pinole Regional Shoreline in Contra Costa County, as far South as Coyote Creek in Santa Clara County, and as far West as the outer coast marshes of Point Reyes National Seashore. The finding of the 2000-2001 monitoring program results were presented as final map entitled "Distribution of Invasive Species Populations by Species", summary graphs and tables included on the ISP webpage (www.spartina.org) and in a poster format presented at the CALFED Bay-Delta Science Conference.

2003 Monitoring Results

Following the labor-intensive estuary wide survey, the 2003 monitoring program inventory included a random selection of 28 sampling sites stratified across site type (marsh type) and bay region (latitude). The objective of the 2003 inventory was to determine the change in area over time, or spread, of non-native *Spartina* by species, marsh type and bay region. The non-native *Spartina* spp. spread an average 244% across all marsh types and bay regions. The *S. alterniflora*-hybrids spread the greatest with an average of 317% increase, suggesting a more than exponential rate of spread. The estuary wide population of *S. alterniflora*-hybrids was estimated to be as high as 1960 acres. The rate of spread was greatest for *S. alterniflora*-hybrids in the Central Bay, near the introduction sites, and in marsh types including mudflats, strip marshes and formerly diked baylands. *S. densiflora* spread an average 52% between 2001 and 2003, *S. patens* apparently decreased in cover but this is thought to be the result of mapping error. The finding of the 2003 monitoring program were presented in the annual 2003 Spartina Monitoring Report.

2004 Monitoring Results (final results pending)

The ISP is currently completing the 2004 estuary wide survey with the objective of determining the current distribution and abundance of the invasive non-native cordgrass, *Spartina* spp. in 2004. The 2004 inventory net abundance results will be analyzed relative to the totals predicted from the 2003 sampling. *Spartina* spread over time will be analyzed relative to species, marsh type, and region (latitude). The 2004 surveys included monitoring a sampling of the 350 acres of *Spartina* treatment that took place in 2004 to determine control efficacy. Any additional genetic surveys requested by landowners were mapped. A total of approximately 500 genetic samples were tested to confirm species identification of the *S. alterniflora* and *S. densiflora* hybrids.

4. **Approach and Scope of Work**

This proposal is comprised of three main tasks, monitoring *Spartina*, developing Invasive Profile Markers, and monitoring California clapper rails prior to treatment. Each of these tasks is described below, and their relationship to the overall project is illustrated in the model in Figure 1. Task 1 is project management, and it is not described herein.

Task 2: Spartina *Monitoring*

The distribution and abundance, or inventory monitoring surveys include both field and aerial photo interpretation methods to survey for and map the presence of the non-native cordgrass. Field based methods include surveying the estuary's tidal marsh, channels, and mudflats using boats, bikes, cars, and foot surveys to access the sites, and GPS (Global Positioning System) to map the species, location and area of the infestations. Species are identified in the field by trained biologists. Hybrid species that can not be confidently identified in the field using morphological characterists are sampled for genetic tests to confirm species identification. Field identification is also confirmed with 5 randomly selected plant samples per site of field identified as *a S. alterniflora-hybrid*. Identified non-native *Spartina* plants are mapped as points with a given diameter, lines with a given width and percent cover class, and polygons with a given percent cover class. Larger infestation sites are mapped with aerial photo interpretation methods using orthorectified infrared aerial photography. Using GIS, ArcView, polygons are digitized around infestations and given a percent cover class. Each estimated cover class is confirmed with a field visit for ground truthing. The monitoring program supports the further development of remote sensing techniques. The monitoring program will work to use the most accurate and efficient mapping and monitoring methods and will modify and adapt accordingly.

Treatment sites are monitored to determine control efficacy. *Spartina* Treatment sites are monitored at 30 randomly selected 0.25 m^2 sampling plots (unless 30 plants were not present, then the total number of plants were monitored) pre- and post-treatment. Data collected includes patch size (diameter or meadow), stem density, stem height, percent cover of native and non-native vegetation, percent flower of *Spartina, Spartina* vigor (high, medium or low), sediment type and tidal zone (high, middle or low). The treatment efficacy data gathered from the treatment monitoring is used to guide future treatments and further control strategy.

The monitoring program provides managers or concerned landowners with genetic surveys of *Spartina* species to confirm the presence of hybrid cordgrass. Individual *Spartina* plants may be sampled, or entire populations maybe sampled for genetic analyses. A transect is run the length of the marsh population sampling leaves every 10 meters. The lab randomly selects a subset of the transect samples for genetic analysis. The sample location points or transects are mapped using GPS.

Inventory, treatment efficacy, and genetic survey field data and aerial photo interpretation data collection is managed by the ISP Field Biologist/Monitoring Coordinator. Inventory and treatment site monitoring is scheduled around the Clapper rail breeding season (*Rallus longirostris*) and the low and high tides. Field data is most often collected by scheduling foot surveys during low tides. Where appropriate, high tide boat surveys are scheduled. Once the data is collected by field staff, they are responsible for downloading the GPS data and reviewing their data for quality control. The data is then transferred to the ISP Field Biologist/Monitoring Coordinator who organizes the data and provides further quality assurance, manages the data organization and compilation. Using Pathfinder Office the GPS data is differentially corrected and exported as ArcView shapefiles for further organization and GIS analysis. Aerial photos are scheduled and flown at low tide during the peak growing season for *Spartina*, ideally between August and September. As noted above, once the photos are orthorectified, the infested sites are digitized and given a cover class and total acreages per site are calculated and incorporated into the overall analysis. The data files are merged and summarized by total area by species, site, and bay region.

Data Analysis:

Statistical tools are used to examine data for outliers, and possible typographical errors for quality assurance. Inventory data analysis includes species spread over time relative to marsh type or bay region (latitude). Treatment monitoring plot data, including stem density or percent cover change pre-post treatment, are analyzed to determine the treatment efficacy. Treatment monitoring plot data will be further analyzed relative to parameters including treatment method, treatment date, flowering phenology (percent flower), marsh type, sediment type, tidal zone or species, using multivariate analyses. Accuracy of field identification using genetic confirmation of is examined. The final data presented in table, graph and map format.

Task 3: Development of Invasive Marker Profiles

1. Identify hybrids during Phase I control.

Genetic Analysis

We have developed and successfully used Random Amplified Polymorphic DNA (RAPD) technique in several studies investigating *Spartina* hybrids (Ayres et al, 1999; Ayres and Strong, ; Ayres et al 2003; Ayres et al 2004). In summary, we developed markers that amplified DNA fragments specific to each parental species, and then used these markers to identify hybrids; those being plants with mixtures of markers from both species. Extraction of DNA will follow the proteinase-K based method outlined in Daehler et al (1999) for RAPD analysis. Amplification conditions and temperatures are shown in Daehler et al (1999); gel scoring is described in Ayres et al (1999).

The use of RAPD markers is specifically addressed by the journal "Molecular Ecology": "The appropriateness of RAPD markers for population genetic inference is increasingly questioned by our reviewers and editors because of concerns about reproducibility, dominance, and homology. Given these worries, and the ready availability of other kinds of markers that do not suffer from all of these problems, studies based primarily on RAPDs only rarely pass the scrutiny of peer review in Molecular Ecology. Of course, there may be situations in which RAPDs are appropriate, such as in genetic mapping studies or in searches for *diagnostic markers for a given species or trait*. These latter kinds of studies will continue to be reviewed by the journal" (italics are ours). We interpret this policy as sanctioning our use of RAPD markers for hybrid identification. However, this method is not sanctioned for the population-genetic inferences we propose to make in proposed research #2-4; hence our advocacy of microsatellite markers for these research goals.

2. Build molecular tools to identify potentially invasive plants in natural populations in the Bay using marker-Quantitative Trait Loci associations.

In light of the accelerating spread of *Spartina* hybrids in San Francisco Bay (Ayres *et al* 2004) we propose that highly invasive individuals at the forefront of the invasion possess extreme phenotypes for traits important in hybrid cordgrass survival and spread. We posit these traits are i) self-compatibility, which enables single individuals to found new populations; ii) height, which confers the ability to grow low on the open mud of the intertidal plane; iii) lateral spread, which anchors plants into the shifting substrate and allows colonization of occupied or unoccupied neighboring patches; iv) tolerance to high (40 ppt) salinity which allows plants to growth higher on the intertidal plane in the range of *Salicornia virginica*, and v) the timing and abundance of flowering which is important in siring ability on early flowering native *S. foliosa* and in seed production. The complexity of phenotypic traits, particularly of those that are involved in adaptation, likely arises from segregation of alleles at many interacting genetic loci (quantitative trait loci or QTL), the effects of which are sensitive to the environment (Mauricio 2001). QTL analysis has been frequently used in marker-assisted selection of crop traits and in disease resistance research in agriculture (Frary et al 2000, Alpert & Tanksley 1996) and medicine (Lander and Schork 1994). Several QTL have been identified for seed size, fruit size, and seed number and flower morphology in *Arabidopsis thaliana* (Alonso-Blanco *et al* 1999, Juenger *et al* 2000), for example, and for reproductive and morphological traits in *Eucalyptus nitens* (Byrne *et al* 1997a, 1997b), Scots pine (Hurme et al 2000), and *Populus*

(Bradshaw and Stettler 1995, Frewen et al 2000). While the physical assessment of quantitative traits in a mature plant can take some years, ultimately having a suite of genetic markers that are linked to invasive traits will allow us to easily conduct hybrid population surveys and quickly identify especially vigorous invasive hybrids and slate them for immediate eradication. Using marker - quantitative trait loci (QTL) association analysis will allow us to determine the correlation of molecular markers and those quantitative trait loci (QTL) that underlie physical traits conferring invasive ability in *Spartina* hybrids. This will permit the specific genetic detection of highly invasive hybrid individuals via 'invasive marker profile (IMP)' surveys. This approach enables more focused eradication efforts by specifically targeting those hybrid plants that are likely driving the spread and continued invasion and that are thus most threatening to the ecosystem, restoration of tidal marshes, and persistence of the native *Spartina*.

<u>Methods:</u>

Quantitative trait analysis requires an adequately large number of molecular markers (~200) in order to ensure that these markers will be distributed throughout most of the genome, assuring the greatest probability of finding associations between gene loci and markers (Mackay 2001). Co-dominant molecular markers such as microsatellites are the most effective in successfully finding marker-trait associations (Erickson *et al* 2004). We already possess a *Spartina* sequence library (Blum *et al* 2004), and a large number (ca 120) of microsatellite markers (Sloop *et al* in preparation, Bando *et al* in preparation). We request funding to develop an additional set of 100 markers.

Sampling Population and Experimental Conditions. Using an inbred-line type of plant breeding, we are planning to grow 384 F2 or BC1 (back-cross 1) seedlings and measure fitness related traits (height, spread rate, self-compatibility, salinity tolerance, pollen and seed production). By using a relatively large number of individuals in the mapping population (384) we are balancing the need for adequate statistical power to detect quantitative trait loci (QTL) of small effect (Doerge 2002) with the per sample cost of the analysis and the rearing of such a sampling population. In order to grow the sampling population we will rent and develop experimental rice-growing fields at UC Davis, and manage the fields to mimic tidal conditions. We will not be doing any experimental plantings in tidal marshes.

We will evaluate tolerance to salinity by vegetatively propagating our sampling population, placing individual plants in small pots, and growing replicated sets of plants under high salinity in a greenhouse at UC Davis. At the end of several weeks, we will measure plant biomass to determine salinity tolerance (See Pakenham-Walsh, 2003, for complete methodology).

Microsatellite genotyping. Microsatellite genotyping requires the duplication of the specific section of the genome containing the region targeted by each marker primer pair via PCR (polymerase chain reaction). One of the primers is labeled with a fluorescent dye used for 'visualization' of the length of the fragment during the genotyping reaction in an ABI 3730xl 96-capillary DNA analyzer. The ABI 3730 output file using ABI GeneMapper 3.0 software then determines individual allele sizes. These allele size data will then be used in conjunction with the individual trait measurements to construct a QTL-microsatellite marker linkage map via Composite Interval Mapping (CIM) (Zeng 1994, Jansen & Stam 1994, Jiang & Zeng 1995) using QTL cartographer software (Basten *et al* 2001).

3. Can the invasion can be controlled by artificially removing invasive genotypes?

We hypothesize that hybrid individuals possessing a genetic profile that includes a majority of markers associated with high fitness traits are most invasive. In order to establish whether this is the case, and whether the invasion can be controlled by artificially removing these invasive genotypes, we will genetically survey the existing or remaining natural hybrid populations each year, using the suite of molecular markers comprising the 'invasive marker profile (IMP)'. This will further enable long-term monitoring of control success, so once the most obvious plants/populations are killed we can evaluate whether the IMP is still present in the remaining populations, and evaluate the potential for the invasive population to be re-created over time. The IMP baseline will allow us to carry out monitoring surveys every five years after current control plans are completed to ensure no future hybrid spread.

<u>Methods:</u>

We will conduct Bay-wide hybrid surveys and genotype a maximum of 288 DNA samples each year. We estimate we will need ~30 markers to comprise the IMP. We will recommend giving IMP *Spartina* hybrids the highest priority for eradication on a Bay-wide scale, but will also assess and monitor the progress of this method at a specific site by also surveying the spread of a control population of non-IMP or 'harmless' hybrids.

4. Evaluate the rate of evolution of hybrids in the Bay.

Using archival DNA samples from hybrid surveys conducted in 1999 and 2004 (by the UC Davis Strong lab), we will conduct population genetic assessments in order to determine the level of gene flow vs. inbreeding among hybrids. This analysis may be extended to later years depending on future funding. This will give us an estimate of the rate of adaptation of *Spartina* to the various growing environments. *Spartina* hybrids may follow a different evolutionary path in relatively protected marsh sites versus exposed open mudflat sites. This may affect the number and distribution of markers within IMPs and is of great importance relative to control efforts targeting IMP individuals.

<u>Methods:</u>

We plan to genotype at least 30 individuals from six hybrid populations collected in 1999 and 2004/5. We will assess the level of genetic variation in each group using Analysis of Molecular Variance (AMOVA) (Excoffier 1992) using Arlequin 2.001 software (Schneider *et al* 2000). We will determine the frequency of IMPs by genotyping all DNA samples with those markers correlated with invasive traits and will be using standard statistical methods to determine differences in marker frequencies among sites and years (4 x 4 contingency table).

Task 4: Clapper rail monitoring

Winter call counts to determine presence absence of California clapper rail (*Rallus longirostris*) and will be undertaken in sites where *Spartina* control has taken place, in 2004, or will take place in 2005. If California clapper rails are not detected, Spartina treatment can begin as soon possible. If herbicide is the selected treatment method, the treatment can begin as early as May, when the plants are actively growing and susceptible to the herbicide. If Clapper rail are detected, aerial herbicide treatments may begin July 15. If Clapper rail are detected and aerial herbicide application is not the appropriate control technique, treatments must wait until September 1, after the Clapper rail breeding season is complete.

Two survey methods will be used to detect the presence of Clapper rail. Narrow sites, including channels, creeks, and strip marshes will be surveyed using “Walking Transects”. Sites greater than 300 meters in width will be surveyed using “Stationary Survey”. "Walking Transect Surveys" will have listening stations 200 meters apart. Stations will be set up from the creek or channel mouths upstream. "Stationary Surveys" will be established as a grid pattern with stations 200 meters apart along the length of the grid line. Clapper rail recordings will be broadcast at each station and responding clapper rail call counts will be monitored to determine presence. Stations will be marked with PVC pipe and GPS coordinated will be collected. If sites have either some remnant burms or levees on which the stations could be established, this is preferred. The total number of stations is site dependent, and range from 2-5 depending on the size and dimensions of the site. Stations need to be relatively accessible. The PVC pipes used to mark the stations will be approximately 2 feet above ground and visible, marked with florescent flagging or paint. The PVC pipes will be placed just off the levees slightly, but not entirely out of view. In order to avoid creating any perches, the stakes will be kept 1-2 feet above ground (or vegetation). Surveys should be conducted at sunrise or sunset (at tides bellow 4.5 - 5 ft NGVD at the GG bridge), however given the winds that tend to pick up before the sunset surveys, the sunrise surveys will be preferred. Surveys conducted at sunrise should begin 30 minutes before sunrise and continue until 1 hour after sunrise. Surveys conducted at sunset should begin 1 hour before sunset and continue until 30 minutes after sunset. Each site will require 3-4 surveys over time.

Each treatment site will require a number people who are approved by USFWS to conduct the rail surveys. Partner agency staff and consultants will work together to pool their resources and work together on the Clapper rail surveys of treatment sites. The Invasive Spartina Project (ISP) has selected approximately 24 *Spartina* treatment sites that will require winter call counts in 2005. Each site will require approximately 100 hours of biologist time for set up and call counts. Approximately half of these hours will be matched by staff time from the partner agencies such as U.S. Fish and Wildlife Service (USFWS), California Department of Fish and Game (CDFG) and East Bay Regional Park District (EBRPD).

All the survey sites will be mapped and marked before the first week of March and the surveys to begin at least by the first week of March. The 3-4 follow up surveys will be scheduled in the weeks that follow the initial establishment of the listening stations.

5. **Feasibility**

The ISP has demonstrated success in previous years completing work similar to the proposed work as described in # 3 above. There have been no changes in the project structure since that time, and there is every reason to believe that we will be able to continue successful work.

6. Expected Outcomes and Products

1. Data on the rate and extent of non-native *Spartina* to guide ISP control efforts.
2. Data on the effectiveness of treatment methods.
3. Information regarding presence of Clapper rail at treatment sites.
4. Genetic markers to help identify aggressive invaders.

7. Data Handling, Storage, and Dissemination

This section documents the Quality Assurance and Quality Control (QA/QC) practices that were implemented for the ISP monitoring program for 2003. These practices were developed and followed to ensure that the resulting data were of the type and quality needed and expected for their intended use. The QA/QC practices generally follow guidance provided by the U.S. EPA at their website (www.epa.gov/quality) and at the website of the National Center for Environmental Research (www.es.epa.gov/ncr).

Although the geographic scope of the 2003 monitoring program was relatively large, its purposes were simple – to assess the rate of spread of specific grass species at a subset of sites, and to determine the efficacy of treatment methods at a number of locations that had been treated. The simplicity of the program allowed relatively simple QA/QC protocols and documentation, as well. The ISP monitoring program Quality Assurance Plan consists of the following elements: (1) statement of monitoring program purpose, (2) monitoring program design, (3) sample handling procedures, (4) equipment calibration, (5) data reduction and statistical methods, (6) program evaluation procedures, and (7) recommendations for improvements for next season. These elements are described below. Additional details regarding conduct of data collection and analysis may be found in the Methods section of this report. Additional details regarding the application and efficacy of the quality assurance and quality control in achieving the objectives of the monitoring program may be found in the Results and Discussion sections of this report.

1. Monitoring program purpose. The primary purpose of the 2003 ISP monitoring program was to estimate the rate(s) of change in area covered and percent covered by non-native cordgrass since 2000-1, based on monitoring at a limited number of sites. Additional sites were monitored to determine the efficacy of treatment methods that had been or were to be implemented at the site.
2. Monitoring program design. The monitoring program sample sites represented the geographic range and habitat variability of invaded marshes in the San Francisco Bay system. The monitoring program design consisted of a 28 sampling sites stratified by latitude that included three bay regions (North, Central, and South) and stratified by site or habitat type (four types with sub-types, described in detail in the Methods section of this report). At least six marshes of each site type were selected. The sites included at least two types within each of the three regions of the bay. The majority of the sampling sites were in the South Bay Region where the bulk of the non-native *Spartina* is currently distributed. A randomized, fully factorial sample was not possible to obtain due to restricted access time windows at some sites because of the presence of endangered clapper rails or tidal eleva-

tion. Within a sample site all non-native cordgrass was identified, mapped with hand-held GPS units, and leaf blades of S. alterniflora-hybrids were collected for genetic analysis to verify field based species identifications.

3. Sample handling procedures. Site and species data were entered into handheld GPS units in real time. Data were transferred from the GPS units into computer spreadsheets, corrected for GPS error, and then packed up onto CD-ROMs. All processing of the data from the GPS to data spreadsheet and GIS file was conducted by the project Field Manager. All field data collector personnel were previously trained by the Field Manager. Where needed to confirm field identification, cordgrass blade and leaf samples were collected in accordance with U.C. Davis protocols defined in Conservancy Agreement #02-024. All samples were labeled by indelible marker in the field with collection numbers and placed in ziploc bags. The bagged specimens were mailed to the U.C. Davis laboratory for genetic analysis, using methods defined in the Conservancy Agreement #02-024.

4. Instrument calibration. Calibration procedures for analytical instrumentation. GPS units were factory calibrated when acquired. GPS data were processed with commercially available software provided by the manufacturer and corrected using data downloaded from designated web sites. The GPS instruments used have a corrected accuracy of +/- 3 meters. Instrumentation for the genetic analyses (molecular biology) was calibrated according to standard laboratory procedures of Ayers et al. (1999). Aerial photography used for synoptic remote sensing surveys of non-native cordgrass was processed according to procedures described in the Guidelines to Monitor the Distribution, Abundance, and Treatment of Non-indigenous Species of Cordgrass in the San Francisco Estuary by Collins et al. (2001).

5. Data reduction and statistical methods. Data collection was similar to methods used in 2001 corresponding to guidelines presented in Collins et al. (2001). All field biologists were trained by K. Zaremba for uniformity and consistency in field methods. Data on species, location, and area covered were entered into handheld GPS units in the field. Supplemental notes were added as needed. Once in the office, data were downloaded to Pathfinder software, differentially corrected, and reviewed by the ISP Biologist and Monitoring Coordinator. After review, the data files were exported to ArcView GIS software. All files were backed up regularly to CDs. There were multiple editing checks of the data and frequent back-ups to CD of intermediate and final data files. Once all data files were collected for the season, the individual site files were merged into a single data set. Files were sorted by data collector and exported to Excel spreadsheets for another round of review by the data collectors. Files were edited as necessary and then combined and imported into ArcView for preliminary GIS analysis and imported into SYSTAT for further statistical analyses. The first statistical analysis was a cross-tabulation of categorical names to check for typographical errors and duplications. Summary statistics were then calculated for quantitative variables to check for unreasonable ranges and outliers. A few errors were detected in the SYSTAT screening tests and these were corrected in the GIS data base and the Excel spreadsheets. GIS plots and statistical comparisons were performed on the final edited files. Data are presented by maps of each sampled site in the Figures

1-31 of the report. Percent cover, percent field identification confirmed by genetic identification, and percent change in area covered were plotted in Figures 32-38.

6. Monitoring program evaluation procedure. The monitoring program will be evaluated by internal and external peer review. The ISP will form a review group of local wetlands and monitoring experts to review this report and provide recommendations for future program revisions. In addition, this report will be made available to the public via the ISP website, and the ISP will fully consider any comments that are provided. The criteria of success for the program will include the ability to quantify rates of change in area covered, to begin to identify and predict full-infestation levels, and to assess the effectiveness of treatment methods.

7. Recommendations for improvement. Based on this year's monitoring experiences and results, we recommend implementing some additional procedures to validate data and to measure the repeatability of, and hence the confidence in, field observations of non-native cordgrass. These procedures include scheduling repeat data collection events to a site by both the original monitoring biologist(s) and by another biologist(s). Data from all events should be compared to assess consistency and repeatability. We also recommend repeating monitoring events at multiple sites, or sub-areas within sites, to quantify inter-observer error (statistical error) and to learn which, if any, site types need more or special attention for better estimating non-native cordgrass cover.

Clapper rail data will be handled according to direction from US FWS.

UC Davis data will be handled according to university protocol, which will be defined in the contract prior to initiation of work.

8. Public Involvement and Outreach

After ISP monitoring data has been evaluated and quality checked, it is made available to the public on request. All of our monitoring results are published annually in the form of Annual Reports, and maps are distributed in hardcopy and electronically.

9. Work Schedule

The Conservancy is prepared to receive the CALFED grant and enter into contracts immediately to begin the work. Each task will run the duration of the 3 year proposed period, except clapper rail monitoring which will be completed at the end of 30 months.

Each of the proposed tasks is independent, although closely interrelated, as shown in Figure 1.

B. Applicability to CALFED Bay-Delta Program ERP Goals, the ERP Draft Stage 1 Implementation Plan, and CVPIA Priorities

ISP Objective	ERP Goal #	NIS Goal #	Calfed Identified Scientific Uncertainty Addressed
Continue regional effort to plan and implement control of invasive *Spartina* to prevent an invasion of San Pablo and Suisun Bays and significantly reduce invasive *Spartina* populations bay wide.	5. Non-native invasive species: Prevent the establishment of additional nonnative invasive species and reduce the negative ecological and economic impacts of established nonnative species in the Bay-Delta estuary and its watershed. **Objective 5:** Halt the introduction of nonnative invasive aquatic and terrestrial plants into the Bay-Delta estuary, its watershed, and other central California waters. **Objective 7:** Limit the spread or, when possible and appropriate, eradicate populations of nonnative invasive species through focused management efforts.	II. Limit spread or eliminate populations through management. III. Reduce harmful ecological, economic, social and public health impacts resulting from infestations of NIS through appropriate management.	
Contribute to the overall scientific understanding of how ecological engineers can physically alter the S.F. Bay ecosystem, how the process of introgression can potentially lead to extinction of native species, and how molecular tools can be used to assist in the management a hybrid invasion.			Non-native Invasive Species (PSP pg 29) ISP, if funded, will help provide data, maps and information regarding: 1) To what extent can *Spartina*, including its hybrid with the native, be eradicated/controlled? 2) To what extent will *Spartina* preclude achieving restoration objectives? 3) How to colonize native species post control? 4) Comprehensive surveys and mapping of *Spartina* populations. 5) Development of management and implementation plans and programs.
Build a bay-wide infrastructure to detect, prevent, and control future invasive species in the intertidal zone.	5. Non-native invasive species: Prevent the establishment of additional nonnative invasive species and reduce the negative ecological and economic impacts of established nonnative species in the Bay-Delta estuary and its watershed. **Objective 5:** Halt the introduction of nonnative invasive aquatic and terrestrial plants into the Bay-Delta estuary, its watershed, and other central California waters. **Objective 7:** Limit the spread or, when possible and appropriate, eradicate populations of nonnative invasivespecies through focused management efforts.	I. Prevent new introductions.	

C. Qualifications

1. Project participants

KATHERINE ZAREMBA

Katherine Zaremba has been working with the San Francisco Estuary Invasive Spartina Project (ISP) for 4 years. She began in 2000 as a Field Biologist assisting with the non-native *Spartina* mapping and monitoring of the Estuary and is currently acting as the Field Biologist/Monitoring Coordinator. As the Monitoring Coordinator her responsibilities currently include the management of the monitoring program's estuary wide distribution and abundance and treatment efficacy monitoring, coordination of annual endangered California clapper rail surveys, training of seasonal monitoring staff, coordination of *Spartina* samples collected for genetic analysis, coordination of annual aerial photography incorporated into the GIS based monitoring, production of GIS maps as needed, presentations at meetings and scientific conferences, production of outreach materials including non-native species identification brochures, and the production of the annual monitoring report. Before joining the ISP Katherine Zaremba completed her Masters Thesis, entitled "Hybridization and Control of a Native-Non Native Spartina Complex in San Francisco Bay" (2000), in Conservation Biology at San Francisco State University. Her research was based at the U.C. Davis Bodega Marine Lab in Dr. Don Strong's Spartina Lab where she also worked as a Graduate Assistant Researcher and Post Graduate Researcher. While working with Dr. Strong she received a position as a California Sea Grant Trainee and a Internship in Population Biology "A Graduate Training Program in Environmental Biology: From Genes to Communities" National Science Foundation grant. Before attending graduate school, Katherine Zaremba worked for 10 years as a naturalist and environmental educator on the San Francisco Bay and California coast working for organizations including Point Reyes National Seashore, Gulf of the Farallones National Marine Sanctuary, Friends of the Estuary and The Marine Mammal Center. She received her B.A. in Romance Languages at Colorado College in 1989.

DONALD R. STRONG

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Professional Preparation

B.S. University of California, Santa Barbara, 1966, BS Zoology, with Honors

M.S. Biology, University of California, Irvine, Biology, 1968.

Ph.D. Biology, University of Oregon, Ecology and Limnology, 1972.

Postdoctoral: University of Chicago, Ford Foundation Fellow in Population Biology, 1972-1973.

Appointments

Professor, Section of Evolution and Ecology & Bodega Marine Laboratory,
University of California, Davis, 1991-present.

Assistant Professor (1973), Associate (1977), then Professor (1983-1991),
Florida State University, Tallahassee, Florida, Department of Biological Science.

Visiting Professor. Swedish University of Agriculture, Uppsala,1989 & 1990.

Synergysitic Activities

2001- Present, Editor in Chief, Ecology and Ecological Monographs.

1997 – 2000, Editor-In-Chief, Oecologia,

1986-1990 Founder and First Editor: Special Features" section, Ecology,.

1993-1994 National Research Council, EMAP Review Committee, Member,

1996 – 1999 National Center for Ecological Analysis and Synthesis, Scientific Advisory Board Member.

1998 – 2000 Director, Center for Population Biology UC Davis.

Recent and Relevant Publications

In Press

Ayres DR, Zaremba K and DR Strong. In press. Extinction of a Common Native Species by Hybridization with an Invasive Congener. Weed Technology.

Jaffee, B. A. and Strong, D. R. in press. Strong bottom-up and weak top-down effects in soil: nematode-parasitized insects and nematode-trapping fungi. Soil Biology & Biochemistry

Strong, D. R. Evolving Weeds and Biological Control. In press. In: Proceedings of the XI International Symposium on Biological Control of Weeds (eds Cullen, J.M., Briese, D.T., Kriticos, D.J., Lonsdale, W.M., Morin, L. and Scott, J.K.) pp. n-n+x. CSIRO Entomology, Canberra, Australia.

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Ayres, D. R., D. R. Strong, and P. Baye. 2003. *Spartina foliosa* – a common species on the road to rarity? Madroño 50: 209-213.

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Grevstad, F. Strong, D. R., Garcia-Rossi, D, Switzer, R. and Wecker, M. 2003. Biological Control of *Spartina alterniflora* in Willapa Bay Washington: Agent specificity, introduction, and early results. Biological Control 27:32-42.

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Anttila CK, King AR, Ferris C, Ayres DR, Strong DR. 2000. Reciprocal hybrid formation of *Spartina* in San Francisco Bay. *Molecular Ecology* 9: 765-771.

Ayres DR, Garcia-Rossi D, Davis, HG, Strong DR. 1999. Extent and degree of hybridization between exotic (*Spartina alterniflora*) and native (*S. foliosa*) cordgrass (Poaceae) in California, USA determined by random amplified polymorphic DNA (PAPDs). *Molecular Ecology* 8: 1179-1186.

Daehler CC, Antilla, CK, Ayres DR, Strong DR. 1999. Evolution of a new ecotype of *Spartina alterniflora* (Poaceae) in San Francisco Bay, California, USA. *American Journal of Botany* 86: 543-546.

Wu M-Y, Hacker S, Ayres DR, Strong DR. 1999. Potential of *Prokelisia* spp. as biological control agents of English cordgrass, *Spartina anglica*. *Biological Control*. 16: 267-273.

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EDUCATION

Ph.D., Ecology, University of California, Davis. March 1997

Master of Science, Biology, University of Oregon, Eugene, June 1978

Bachelor of Arts, Botany, University of California, Berkeley, June 1976. GPA 3.6

RESEARCH EXPERIENCE

Staff Research Associate IV, Supervisor, Evolution and Ecology, UC Davis, 4/01 to present.

Continuation of *Spartina* research program. Dr. Donald Strong.

Post-doctoral Researcher, Bodega Marine Laboratory, 7/97 to 4/00, and UC Davis 4/00 to 4/01Dr. Donald Strong, Section of Evolution and Ecology, UC Davis, Davis, CA 95616

Doctoral Research, UC Davis, 3/92 to 3/97. Dr. Marcel Rejmánek, Section of Plant Biology, Section of Evolution and Ecology, UC Davis, and Dr. Frederick Ryan, USDA-ARS, Fresno, California.

Postgraduate Researcher, UC Davis, 6/95 to 9/96.

Surveyed allozyme and RAPD banding patterns of Russian Thistle (*Salsola tragus*) to characterize patterns of genetic variation in northern California and Europe. We have discovered a cryptic species, new to California, perhaps to science. Dr. Frederick Ryan, USDA-ARS.

PUBLICATIONS

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Blum MJ, Sloop CM, Ayres DR, Strong DR. 2004. Characterization of microsatellite loci in *Spartina* species (Poaceae). Molecular Ecology Notes 4: 39 - 42.

Ayres DR, Smith DL, Zaremba K, Klohr S, Strong DR. 2004. Spread of exotic cordgrasses and hybrids *(Spartina sp.)* in the tidal marshes of San Francisco Bay. Biological Invasions. 6: 221-231.

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Foin TC, Reilly SP, Pawley AL, Ayres DR, Carlson TM, Hodem PJ, and Switzer PV. 1998. Improving recovery planning for the conservation of threatened and endangered taxa. *Bioscience* 48(3): 177-184.

Ayres DR and Ryan FJ. 1997. The clonal and population structure of a rare endemic plant, *Wyethia reticulata* (Asteraceae): allozyme and RAPD analysis. *Molecular Ecology* 6:761-772.

<u>IN PRESS</u>

Ayres DR, Zaremba K and DR Strong. In press. Extinction of a Common Native Species by Hybridization with an Invasive Congener. Weed Technology.

2. Project structure

The ISP has been operating for four years, and has developed strong structures for implementing it's various programs. The work included under this proposal will be accomplished primarily through an Interagency Agreement between with Conservancy (Task 2 and 3), and contracts with the ISP's primary officers: Peggy Olofson (Olofson Environmental, Inc.), the ISP Director, and Katy Zaremba, the ISP Monitoring Program Manager. Additional tasks and contract work will be subcontracted from these primaries.

D. Cost

See attached budget

E. Compliance with Standard Terms and Conditions

The Conservancy has read and agrees to comply with all standard terms and conditions.

F. Literature Cited

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G. **Nonprofit Verification [not applicable]**

			TASK 1		TASK 2A		TASK 2B		TASK 2A & 2C		TASK 3		TASK 4		
			Project Management		Annual Bay-wide Spartina Inventory		Treatment Efficacy Monitoring		Genetic analysis and seedling survey		Invasive Marker Profiling		Clapper Rail Presence/Absence Surveys		
		Rate	*Hours*	*Cost*	*Inventory monitoring*	*Inventory monitoring cost*	*treatment monitoring*	*treatment monitoring costs*	*UCD genetic analysis and seedling survey*		*UCD DevelopQTL and IMPS*		*CACR P/A*	*CACR P/A costs*	*Total*
2005															
Staff	Grant Manager - Maxene Spellmar	$4,337/month+29% ben	240	$8,927											$8,927
	Program Manager - Nadine Hitchc	$5987/month+ 29% ben	24	$1,070											
	Attorney - Jack Judkins	$7,386/month+ 29% ben	120	$6,599											$6,599
2005 Staff Subtotal				$15,526		$0		$0		$0		$0		$0	$15,526
Contracts	contract - program manager (KTZ)	$65/hr			999	$64,935	665	$43,225					50	$3,250	$111,410
	contract - Project Manager (Peggy)	$85/hr	100	$8,500											$8,500
	contract - Clapper rail expert	$100/hour											480	$48,000	$48,000
	contract - crew (hours)	$25/hr			888	$22,200	499	$12,475						$0	$34,675
	contract - crew leader (hours)	$40/hr			600	$24,000	333	$13,320							$37,320
	contract - UC Davis	from 11/16/04 proposal				$110,339			Included in 2A			$139,950			$250,289
2005 Contracts Subtotal				$8,500		$221,474		$69,020		$0		$139,950		$51,250	$490,194
Direct Expenses	boat days (days)	$250/day			5	$1,250	3	$750						$0	$2,000
	travel (miles)	$0.36/mile			5580	$2,009	3120	$1,123						$0	$3,132
	Photos (each, including image and orthorectification)	$284/each			130	$36,920		$0						$0	$36,920
2005 Direct Subtotal				$0		$40,179		$1,873		$0		$0		$0	$42,052
	Total 2005			**$24,026**		**$261,653**		**$70,893**	**Included in 2A**			**$139,950**		**$51,250**	**$547,772**
							Task 2 total	*$332,546*							
2006															
Staff	Grant Manager - Maxene Spellmar	$4,337/month+29% ben	240	$8,927											$8,927
	Program Manager - Nadine Hitchc	$5987/month+ 29% ben	24	$1,070											
	Attorney - Jack Judkins	$7,386/month+ 29% ben	120	$6,599											$6,599
2006 Staff Subtotal				$15,526		$0		$0		$0		$0		$0	$15,526
Contracts	contract - program manager (KTZ)	$65/hr			999	$64,935	665	$43,225					50	$3,250	$111,410
	contract - Project Manager (Peggy)	$85/hr	100	$8,500											$8,500
	contract - Clapper rail expert	$100/hr											480	$48,000	$48,000
	contract - crew (hours)	$25/hr			888	$22,200	595	$14,875						$0	$37,075
	contract - crew leader (hours)	$40/hr			600	$24,000	397	$15,880						$0	$39,880
	contract - UC Davis	from 11/16/04 proposal				$110,339			Included in 2A			$139,950			$250,289
2006 Contracts Subtotal				$8,500		$221,474		$73,980		$0		$139,950		$51,250	$495,154
Direct Expenses	boat days (days)	$250/day			6	$1,500	4	$1,000						$0	$2,500
	travel (miles)	$0.36/mile			5580	$2,009	3720	$1,339						$0	$3,348
	Photos (each, including image and orthorectification)	$284/each			130	$36,920		$0						$0	$36,920
2006 Direct Expenses Subtotal				$0		$40,429		$2,339		$0		$0		$0	$42,768
Total 2006				**$24,026**		**$261,903**		**$76,319**	**Included in 2A**			**$139,950**		**$51,250**	**$553,448**
							Task 2 total	*$338,222*							

2007															$0
Staff	Grant Manager - Maxene Spellmar	$4,337/month+29% ben	240	$8,927											$8,927
	Program Manager - Nadine Hitchc	$5987/month+ 29% ben	24	$1,070											
	Attorney - Jack Judkins	$7,386/month+ 29% ben	120	$6,599											$6,599
2007 Staff Subtotal				$15,526		$0		$0		$0		$0		$0	$15,526
Contracts	contract - program manager (KTZ)	$65/hr			999	$64,935	665	$43,225					50	$3,250	$111,410
	contract - Project Manager (Peggy)	$85/hr	100	$8,500											$8,500
	contract - Clapper rail expert	$100/hr											480	$48,000	$48,000
	contract - crew (hours)	$25/hr			888	$22,200	643	$16,075						$0	$38,275
	contract - crew leader (hours)	$40/hr			600	$24,000	429	$17,160						$0	$41,160
	contract - UC Davis	from 11/16/04 proposal				$110,339			Included in 2A			$139,950			$250,289
2007 Contracts Subtotal				$8,500		$221,474		$76,460		$0		$139,950		$51,250	$497,634
Direct Expenses	boat days (days)	$250/day			9	$2,250	3	$750						$0	$3,000
	travel (miles)	$0.36/mile			5580	$2,009	4020	$1,447						$0	$3,456
	Photos (each, including image and orthorectification)	$284/each			130	$36,920		$0						$0	$36,920
2007 Direct Expenses Subtotal				$0		$41,179		$2,197		$0		$0		$0	$43,376
Total 2007				**$24,026**		**$262,653**		**$78,657**	**Included in 2A**			**$139,950**		**$51,250**	**$556,536**
							Task 2 total	*$341,310*							
	GRAND TOTAL			**$72,078**		**$786,208**		**$225,870**	Included in 2A			**$419,850**		**$153,750**	**$1,657,756**
							Task 2 total	$1,012,078							

Tasks And Deliverables

Monitoring for Invasive Spartina Control in the San Francisco Estuary

Task ID	Task Name	Start Month	End Month	Deliverables
1	Project Management	1	36	Semiannual and final reports. Periodic invoices.
2	Spartina Monitoring	1	36	Annual monitoring reports and inventory maps. Data and GIS available to public at end of year.
3	Invasive Marker Profiling	1	36	Semiannual reports. Peer reviewe report of findings.
4	Clapper Rail Presence/Absence Surveys	6	30	Semiannual reports.

Comments

If you have comments about budget justification that do not fit elsewhere, enter them here.

Budget Summary

Project Totals

Labor	Benefits	Travel	Supplies And Expendables	Services And Consultants	Equipment	Lands And Rights Of Way	Other Direct Costs	Direct Total	Indirect Costs	Total
$38,595	$10,473	$900	$0	$1,601,428	$0	$0	$0	$1,651,396	$0	$1,651,396

Do you have cost share partners already identified?
Yes.

If yes, list partners and amount contributed by each:

The Wildlife Conservation Board (WCB) has committed $3M for ISP management and Spartina control over the next 3−5 years (average $750K per year).

Do you have potential cost share partners?
Yes.

If yes, list partners and amount contributed by each:

The Moore Foundation has expressed interest in providing ISP funding. We have not yet worked out details, but we are hoping they may be able to match the WCB commitment.

Are you specifically seeking non−federal cost share funds through this solicitation?
No.

Monitoring for Invasive Spartina Control in the San Francisco Estuary

Monitoring for Invasive Spartina Control in the San Francisco Estuary

Year 1 (Months 1 To 12)

Task	Labor	Benefits	Travel	Supplies And Expendables	Services And Consultants	Equipment	Lands And Rights Of Way	Other Direct Costs	Direct Total	Indirect Costs	Total
1: project management (12 months)	12865	3491	300	0	8500	0	0	0	$25,156	0	$25,156
2: Spartina Monitoring (12 months)	0	0	0	0	332546	0	0	0	$332,546	0	$332,546
3: Invasive Marker Profiling (12 months)	0	0	0	0	139950	0	0	0	$139,950	0	$139,950
4: Clapper Rail Presence/Absence Surveys (7 months)	0	0	0	0	48000	0	0	0	$48,000	0	$48,000
Totals	**$12,865**	**$3,491**	**$300**	**$0**	**$528,996**	**$0**	**$0**	**$0**	**$545,652**	**$0**	**$545,652**

Year 2 (Months 13 To 24)

Task	Labor	Benefits	Travel	Supplies And Expendables	Services And Consultants	Equipment	Lands And Rights Of Way	Other Direct Costs	Direct Total	Indirect Costs	Total
1: project management (12 months)	12865	3491	300	0	8500	0	0	0	$25,156	0	$25,156

2: Spartina Monitoring (12 months)	0	0	0	0	338222	0	0	0	$338,222	0	$338,222
3: Invasive Marker Profiling (12 months)	0	0	0	0	139950	0	0	0	$139,950	0	$139,950
4: Clapper Rail Presence/Absence Surveys (12 months)	0	0	0	0	48000	0	0	0	$48,000	0	$48,000
Totals	**$12,865**	**$3,491**	**$300**	**$0**	**$534,672**	**$0**	**$0**	**$0**	**$551,328**	**$0**	**$551,328**

Year 3 (Months 25 To 36)

Task	Labor	Benefits	Travel	Supplies And Expendables	Services And Consultants	Equipment	Lands And Rights Of Way	Other Direct Costs	Direct Total	Indirect Costs	Total
1: project management (12 months)	12865	3491	300	0	8500	0	0	0	$25,156	0	$25,156
2: Spartina Monitoring (12 months)	0	0	0	0	341310	0	0	0	$341,310	0	$341,310
3: Invasive Marker Profiling (12 months)	0	0	0	0	139950	0	0	0	$139,950	0	$139,950
4: Clapper Rail Presence/Absence Surveys (6 months)	0	0	0	0	48000	0	0	0	$48,000	0	$48,000

Totals	**$12,865**	**$3,491**	**$300**	**$0**	**$537,760**	**$0**	**$0**	**$0**	**$554,416**	**$0**	**$554,416**

Budget Justification

Monitoring for Invasive Spartina Control in the San Francisco Estuary

Labor

Maxene Spellman, Project Manager, will be primarily responsible for management of this grant. Ms. Spellman has been the Project Manager for the ISP since its inception in 2000, and has facilitated movement of over $2M for the project. It is anticipated she will work approxmately 240 hours per year, at a rate of $4,337 per month plus 29% benefits, for an annual budget of $8,927.

Jack Judkins is staff attorney, and will review all legal documents. It is anticipated that he will work approxmately 120 hours per year, at a rate of $7,386 per month plus 29% benefits, for an annual budget of $6,599.

Nadine Hitchcock is the Manager of the Conservancy's Bay Program, and will oversee all grant and contract activities. It is anticipated she will work approxmately 24 hours per year, at a rate of $5,987 per month plus 29% benefits, for an annual budget of $1,070.

Benefits

Benefit rates for staff that will be retained by UC Davis for work on this project are described in the "Services and Consultants" section below.

Travel

Travel costs for consultants and universtity staff are described in the "Services and Consultants" section below.

Supplies And Expendables

No supplies or expendables are proposed for purchase under this grant, with the exception of laboratory materials, described in the "Services and Consultants" section, below, that will be included in the interagency agreement with UC Davis.

Services And Consultants

Task 1 – Project Management ($8,500/year)

Peggy Olofson (Olofson Environmental, Inc.), the Director of the Spartina Project, will be contracted at a rate of $85 per hour for an average of 8 hours per month (100 hours per year) to prepare draft reports and oversee and advise on subcontracts under this grant. Ms. Olofson has been Director of the Spartina Project for 2 ½ years, and typically oversees and directs all aspects of the project. The annual budget for this work is $8,500.

Tasks 2 and 3 – Spartina Monitoring ($338,000/year) and Invasive Marker Profiling ($139,940/year)

Katy Zaremba, the Spartina Project Monitoring Program Director, will be contracted at a rate of $65 per hour for and average of 139 hours per month (approximately ¾-time) to oversee and direct all Spartina inventory and efficacy monitoring work, including scheduling and supervising field crew and crew leaders, evaluating data, and preparing annual reports. Ms. Zaremba has four years experience planning and implementing the Spartina Monitoring Program. The annual budget for this work is $108,160.

Field crew and crew leaders have not yet been selected, and this will be done via standard State contracting protocols. Crew leaders will be biologists extensively trained in identification of non-native Spartina, collection of data using GPS, and work in tidal marshes. They will be contracted at a rate not expected to exceed $40 per hour. There will be two crew leaders, and each is expected to work approximately

12.5 weeks per year. Four crew members (biologists with sufficient training to work adequately under direct supervision of a crew leader) will be contracted at a rate not expected to exceed $25 per hour for approximately 9.5 weeks per year each. The crew will work with the Monitoring Program Director to perform all spartina surveys. The annual budget for crew and leaders is $75,000.

Dr. Don Strong and Dr. Debra Ayres will be retained through an interagency agreement with University of California, Davis. They designed the Task 2 research (developing Invasive Marker Profiles for S. alterniflora hybrids), and will participate in all phases of laboratory and research work. They will coordinate the activities, be the immediate supervisors of all personnel, and make all spending decisions associated with this work. They will implement the objectives of the proposed study and will serve as the liaison with the other agencies and individuals concerned with invasive cordgrasses in Pacific estuaries. Dr. Strong will dedicate the equivalent of one month of his time each year to this project, supervising and taking part in all aspects of the research. Dr. Strong's anticipated billing rate is $14,074 per month plus benefits (0.4%) and indirect (25%), for an annual budget of approximately $18,296 per year. Dr. Ayres will be the full-time coordinator of the genetics and ground truthing aspects of this research. Dr. Ayres has extensive (> 6 years) experience working in this system and is intimately familiar with the Bay, its marshes, and the nuances of Spartina population biology and genetics. She has published extensively on this subject, has represented the lab's research to regional, state, national, and international audiences, and is uniquely qualified to co-lead the proposed Task 2 research. Dr. Ayres' anticipated billing rate is $3,914 per month plus benefits (31%) and indirect (25%), for an annual budget of approximately $76,910 per year.

Christina Sloop, a PhD candidate in Strong's lab, was selected by Dr. Ayres to conduct the QTL-microsatellite work associated with this proposal, and will work full-time. Ms. Sloop has developed microsatellite markers for Spartina sp. in the past two years as part of her doctoral dissertation work. She is

highly qualified to continue Spartina microsatellite marker development in the context of quantitative trait analysis (one of her dissertation exam topics) and the assessment of population genetic structure of Spartina hybrids. Ms. Sloop will have completed her PhD before the start of the grant research; this research will be part of her post-doctoral training. Ms. Sloop’s anticipated billing rate is $2,639 per month plus benefits (25%), indirect (25%), and fee remission to the University ($9,270 per year), for an annual proposed budget of approximately $58,751.

A graduate student researcher (GSR VII) will be employed by the University to assist Ms. Sloop with molecular work. The GSR will work half-time during the school year and full-time during the summer, at a rate of $3,869 per month plus benefits (2-3%) and indirect (25%), for an annual budget of approximately $35,810.

A part time undergraduate laboratory assistant will be employed by the University to assist with processing DNA samples. The assistant will work 1/4-time during the school year and ½-time during the summer, at a rate of $2,056 per month plus benefits (5%) and indirect (25%), for an annual budget of approximately $10, 118. [Note: this budget includes work to be performed under Task 4 as well]

Laboratory costs associated with the genetics aspect of this research total $35,000 per year, and cover costs associated with extracting and purifying DNA, reagents and plastics consumed in PCRs (polymerase chain reactions), and the use of the ABI 3730 capillary sequencer in the Genome Center at UC Davis for microsatellite analysis.

Other research-related costs include the rental of experimental rice growing fields in the Davis area to grow the required sampling population. This will allow us to determine differences in trait measurements caused by environmental effects rather than genetic ones. The estimated cost of rice field rental is $3,000 the first year, and $1,500 the second year.

Other monitoring-related costs include the acquisition and ortho-rectification of aerial photographs for selected sites to assist in estimation of spread rates and treatment success ($36,920 per year, to be included as a direct expense in the contract with Katy Zaremba and/or Olofson Environmental, Inc).

Task 4 – Clapper Rail Presence/Absence Surveys ($51,250/yr)

Clapper rail expert(s) will be contracted to perform surveys to determine whether endangered California clapper rails are present in locations proposed for Spartina control. Surveys must be performed in winter (typically December-February) by highly trained and permitted (Section 10(a)(1)(a)) or certified biologists. The surveys are performed twice a day for up to five days over three weeks. A typical billing rate for a qualified biologist is $100 per hour. We anticipate that we will need to survey approximately 24 sites each year, with five days (two 4-hour surveys each day) required to complete each survey (960 hours). However, we anticipate that ISP partners, including USFWS, East Bay Regional Parks District, and California Department of Fish and Game, will provide qualified staff to complete half of these surveys, therefore the proposed budget for clapper rail surveys is $48,000 per year.

Katy Zaremba will work with the contractor and ISP partners to plan and coordinate clapper rail surveys with other work. The annual budget for this is $3,250 per year.

Travel will be included in the Interagency Agreement with the University for collection of clones and material for DNA analysis, and travel to regional and national meetings to present results (approximately $3,000 per year), and in contracts for field crew and crew leaders to access sites for survey work (approximately $3,300 per year).

Equipment

No equipment will be purchased under this grant. The ISP recently purchased three new GPS units (for a total of five units) for inventory work.

Lands And Rights Of Way

No lands or easements will be acquired for the proposed work.

Other Direct Costs

There are no other direct costs.

Indirect Costs/Overhead

Indirect costs of 25% of salaries and benefits will be charged by UC Davis under the planned interagency agreement. This will be $144,007 of the total grant amount.

Comments

Environmental Compliance

Monitoring for Invasive Spartina Control in the San Francisco Estuary

CEQA Compliance

Which type of CEQA documentation do you anticipate?
X none
− negative declaration or mitigated negative declaration
− EIR
− categorical exemption

If you are using a categorical exemption, choose all of the applicable classes below.
− Class 1. Operation, repair, maintenance, permitting, leasing, licensing, or minor alteration of existing public or private structures, facilities, mechanical equipment, or topographical features, involving negligible or no expansion of use beyond that existing at the time of the lead agency's determination. The types of "existing facilities" itemized above are not intended to be all−inclusive of the types of projects which might fall within Class 1. The key consideration is whether the project involves negligible or no expansion of an existing use.
− Class 2. Replacement or reconstruction of existing structures and facilities where the new structure will be located on the same site as the structure replaced and will have substantially the same purpose and capacity as the structure replaced.
− Class 3. Construction and location of limited numbers of new, small facilities or structures; installation of small new equipment and facilities in small structures; and the conversion of existing small structures from one use to another where only minor modifications are made in the exterior of the structure. The numbers of structures described in this section are the maximum allowable on any legal parcel, except where the project may impact on an environmental resource of hazardous or critical concern where designated, precisely mapped, and officially adopted pursuant to law by federal, state, or local agencies.
− Class 4. Minor public or private alterations in the condition of land, water, and/or vegetation which do not involve removal of healthy, mature, scenic trees except for forestry or agricultural purposes, except where the project may impact on an environmental resource of hazardous or critical concern where designated, precisely mapped, and officially adopted pursuant to law by federal, state, or local agencies.
− Class 6. Basic data collection, research, experimental management, and resource evaluation activities which do not result in a serious or major disturbance to an environmental resource, except where the project may impact on an environmental resource of hazardous or critical concern where designated, precisely mapped, and officially adopted pursuant to law by federal, state, or local agencies. These may be strictly for information gathering purposes, or as part of a study leading to an action which a public agency has not

yet approved, adopted, or funded.
– Class 11. Construction, or placement of minor structures accessory to (appurtenant to) existing commercial, industrial, or institutional facilities, except where the project may impact on an environmental resource of hazardous or critical concern where designated, precisely mapped, and officially adopted pursuant to law by federal, state, or local agencies.

Identify the lead agency.
California Coastal Conservancy

Is the CEQA environmental impact assessment complete?
Yes.

If the CEQA environmental impact assessment process is complete, provide the following information about the resulting document.

Document Name	**Final Programmatic EIS/EIR – San Francisco Estuary Project Spartina Control Program**
State Clearinghouse Number	**2001042058**

If the CEQA environmental impact assessment process is not complete, describe the plan for completing draft and/or final CEQA documents.

NEPA Compliance

Which type of NEPA documentation do you anticipate?
X none
– environmental assessment/FONSI
– EIS
– categorical exclusion

Identify the lead agency or agencies.

US Fish and Wildlife Service

If the NEPA environmental impact assessment process is complete, provide the name of the resulting document.
Final Programmatic EIS/EIR – San Francisco Estuary Project Spartina Control Program

If the NEPA environmental impact assessment process is not complete, describe the plan for completing draft and/or final NEPA documents.

Successful applicants must tier their project's permitting from the CALFED Record of Decision and attachments providing programmatic guidance on complying with the state and federal endangered species acts, the Coastal Zone Management Act, and sections 404 and 401 of the Clean Water Act.

Please indicate what permits or other approvals may be required for the activities contained in your proposal and also which have already been obtained. Please check all that apply. If a permit is *not* required, leave both Required? and Obtained? check boxes blank.

Local Permits And Approvals	**Required?**	**Obtained?**	**Permit Number (If Applicable)**
conditional Use Permit	–	–	
variance	–	–	
Subdivision Map Act	–	–	
grading Permit	–	–	
general Plan Amendment	–	–	
specific Plan Approval	–	–	
rezone	–	–	
Williamson Act Contract Cancellation	–	–	
other	–	–	

State Permits And Approvals	**Required?**	**Obtained?**	**Permit Number (If Applicable)**
scientific Collecting Permit	–	–	
CESA Compliance: 2081	–	–	
CESA Complance: NCCP	–	–	
1602	–	–	
CWA 401 Certification	–	–	
Bay Conservation And Development Commission Permit	–	–	

reclamation Board Approval	–	–	
Delta Protection Commission Notification	–	–	
state Lands Commission Lease Or Permit	–	–	
action Specific Implementation Plan	–	–	
other	–	–	

Federal Permits And Approvals	Required?	Obtained?	Permit Number (If Applicable)
ESA Compliance Section 7 Consultation	–	–	
ESA Compliance Section 10 Permit	X	X	
Rivers And Harbors Act	–	–	
CWA 404	–	–	
other Project Biologists May Work Under Special FWS Certification In Lieu Of 10(A)(1)(A)Permit	X	X	

Permission To Access Property	Required?	Obtained?	Permit Number (If Applicable)
permission To Access City, County Or Other Local Agency Land Agency Name Alameda County Flood Control District, City Of Palo Alto, Baylands Preserve, East Bay Regional Parks District, San Francisco Department Of Parks And Recreation, Santa Clara Valley Water District	X	X	
permission To Access State Land Agency Name	X	X	

California Department Of Fish And Game, California Department Of Parks And Recreation, State Lands Commission			
permission To Access Federal Land **Agency Name** US Fish And Wildlife Service, US Coast Guard, US Army Corps Of Engineers, US Department Of Navy	X	X	
permission To Access Private Land **Landowner Name** Tiscornia Trust, Port Of Oakland, Port Of San Francisco	X	X	

If you have comments about any of these questions, enter them here.

Land Use

Monitoring for Invasive Spartina Control in the San Francisco Estuary

Does the project involve land acquisition, either in fee or through easements, to secure sites for monitoring?
X No.
– Yes.

How many acres will be acquired by fee?

How many acres will be acquired by easement?

Describe the entity or organization that will manage the property and provide operations and maintenance services.

Is there an existing plan describing how the land and water will be managed?
– No.
– Yes.

Will the applicant require access across public or private property that the applicant does not own to accomplish the activities in the proposal?
– No.
X Yes.

Describe briefly the provisions made to secure this access.

The ISP has obtained letters of authorization from all public and private landowners for access to survey sites. These letters are reviewed and updated annually.

Do the actions in the proposal involve physical changes in the current land use?
X No.
– Yes.

Describe the current zoning, including the zoning designation and the principal permitted uses permitted in the zone.

Describe the general plan land use element designation, including the purpose and uses allowed in the designation.

Describe relevant provisions in other general plan elements affecting the site, if any.

Is the land mapped as Prime Farmland, Farmland of Statewide Importance, Unique Farmland, or Farmland of Local Importance under the California Department of Conservation's Farmland Mapping and Monitoring Program?

X No.
− Yes.

Land Designation	Acres	Currently In Production?
Prime Farmland		−
Farmland Of Statewide Importance		−
Unique Farmland		−
Farmland Of Local Importance		−

Is the land affected by the project currently in an agricultural preserve established under the Williamson Act?

X No.
− Yes.

Is the land affected by the project currently under a Williamson Act contract?

− No.
− Yes.

Why is the land use proposed consistent with the contract's terms?

Describe any additional comments you have about the projects land use.