Brachypodium Control

Experimental Treatments to Control *Brachypodium*
An Adaptive Approach for Conserving Endemic Species
San Diego County, California

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Executive Summary

Brachypodium distachyon is an emerging invasive species with potentially widespread ecological implications for native species, habitats, and ecosystem processes. It has increased in extent and dominance in recent years in San Diego County, possibly in response to repeated fires and climatic conditions. *Brachypodium* decreases native species diversity and may alter soil ecology, vegetation community structure and composition, and natural fire regimes. This species is particularly dense on restricted soils and, thus, threatens edaphic endemic plants such as *Acanthomintha ilicifolia, Bloomeria clevelandii, Brodiaea filifolia, Brodiaea orcuttii, Deinandra conjugens, Dudleya variegata, Nolina interrata,* and *Tetracoccus dioicus*, as well as native grassland and coastal sage scrub communities. These plants and habitats are conservation targets under the Natural Community Conservation Planning programs in San Diego County, California. The conserved areas selected for treatment─Crestridge Ecological Reserve and South Crest─form a central core area for linking populations of both plants and animals between north and south San Diego County preserves.

Conceptual Models

We used results from previous studies and developed conceptual models to:

1. Document our understanding of life history traits that influence persistence and dispersal of *Brachypodium*.
2. Identify observed or potential ecological effects, based on environmental correlates.
3. Identify those variables that may respond to control treatments and be used for developing restoration strategies.
4. Predict areas at risk of invasion now and under future climate regimes.

Based on the models, we developed mechanical and chemical treatment and restoration strategies that focused on reducing or eliminating *Brachypodium* while creating conditions under which native species could germinate, establish, and persist (Figure ES-1). Our objectives were:

1. Reduce *Brachypodium* biomass (thatch) and cover to allow for native species germination.
2. Reduce and prevent further input to the *Brachypodium* seed bank.
3. Restrict seed dispersal through Best Management Practices to avoid inadvertently moving seed between sites.
4. Establish native species that are functionally similar to invaders, thereby increasing both habitat resistance to future invasions and potentially suitable habitat for conservation target species, specifically *Acanthomintha ilicifolia, Nolina interrata,* and *Dudleya variegata.*

Figure ES-1

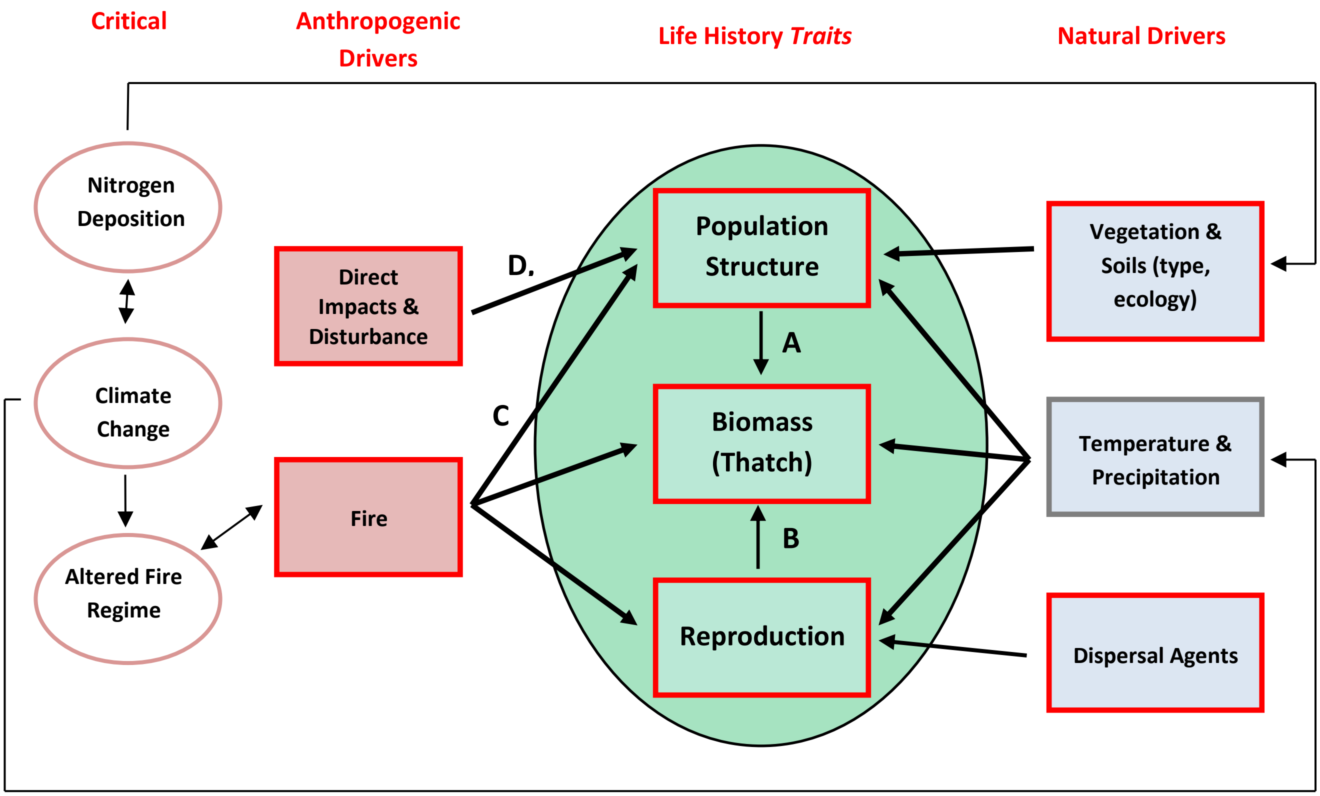
Brachypodium Conceptual Management Model

Brachypodium distachyon

Goal: Reduce or eliminate *Brachypodium* where it forms dense stands on conserved lands and threatens sensitive species and habitat persistence, and restore habitat to prevent future invasions by this species.

Monitoring

Monitor response of *Brachypodium* and native species to invasives control, access control, and habitat restoration.



Uncertainties

- BMPs for *Brachypodium* control
- Types of disturbances that are most problematic
- Soil affinities
- Seed bank longevity

Others: visualized in white bubbles

Management

A) Reduce/eliminate above-ground population
B) Minimize inputs to seed bank
C) Restore habitat to reduce gaps
D) Reroute roads/trails; prohibit/restrict recreational activities that promote seed dispersal
E) Exclude (excessive fire)

Experimental Design

We used relatively small experimental treatment plots on clay and gabbro-derived soils to assess the relative effectiveness of potential management strategies for *Brachypodium* and to develop restoration methods for augmenting native species populations. We conducted standardized habitat assessments in the field to establish pre-treatment site conditions and against which to compare our treatment results. The experiment used elements of both blocked and split-plot designs, paired with adjacent controls.

We designed treatment and restoration plans to address the following questions:

1. Are there significant differences in species cover and richness with the different treatment combinations?
    - Dethatch-Herbicide (Fusilade–glyphosate)-Seeding
    - Dethatch-Mechanical (mowing)-herbicide (glyphosate)-Seeding
    - Herbicide (Fusilade-glyphosate)
    - Control
2. Does dethatching improve treatment effectiveness or enhance native species richness?
3. Are there significant differences in native species cover/richness between seeded and non-seeded (natural recruitment) plots?

We developed site-specific restoration plans for 14 polygons and conducted experimental treatments for 2 years within 8 of those polygons. Dethatching was conducted in Fall 2012 in polygons with low native species diversity, and was followed by mechanical or herbicide treatments in 2013 and 2014 and seeding in 2013. Herbicide-only treatment polygons were treated in 2013 and 2014. Working with volunteers, we collected seed onsite and from the South San Diego County region and either seeded directly into restoration sites or bulked seed, by growing plants to increase the amount of seed available for restoration, using a nursery in South San Diego County. We installed educational and informational signage and fencing to protect the sites from outside variables and introduced seed.

Results

We monitored cover and species richness pre- and post-treatment, using a 0.5 x 1 m quadrat in each plot, randomly sited initially and stationary thereafter. Our major findings over this period were:

- Control of *Brachypodium* can be achieved with one of several chemical (herbicide) treatment combinations. A single Fusilade application per year provided effective control when applied uniformly and timed appropriately relative to rainfall events; an additional application may be required where late rainfall stimulates additional *Brachypodium* germination. Results of mechanical treatments (mowing) were intermediate between herbicide and controls; thus, mechanical treatment may be used in lieu of herbicide where the latter is not feasible or practical.

- Dethatching substantially reduces litter and may increase suitable sites for native species germination, although we did not see a significant increase in native species diversity in dethatched areas. Several native species present onsite appeared to benefit from thatch removal as indicated by increased growth or germination.
- Observationally, the dethatch-herbicide-seeding combination consistently had the highest number of native species present, probably due to increased seed-soil contact. The dethatch-mechanical-herbicide-seeding combination was almost identical to the herbicide-only combination with respect to number of native species present. Thatch left in place in the dethatch-mechanical-herbicide-seeding treatment may have limited seed-soil contact.
- We did not see a significant increase in native species diversity, which may be a result of small sample plots, low rainfall conditions, or short timeframe of the study. Estimates of species richness in quantitative plots were low and idiosyncratic. Observationally, species richness appeared higher in seeded versus control plots.
- Because of high seed output, high seed viability, and minimal seed dormancy, there is the potential for *Brachypodium* to rebound in treated areas if control measures are discontinued prematurely.

The relatively low cover of native species may have been related, at least in part, to drought conditions. We observed good initial germination following seeding and a rainfall event, but the majority of plants did not persist to flowering or fruiting, presumably due to lack of water following germination. It appeared that germination was limited compared to the amount of seed introduced into the soil seed bank. The bulk of the introduced seed may still be present in the seed bank and available for germination with adequate rainfall conditions, particularly if *Brachypodium* cover and thatch are maintained at low levels.

Recommendations

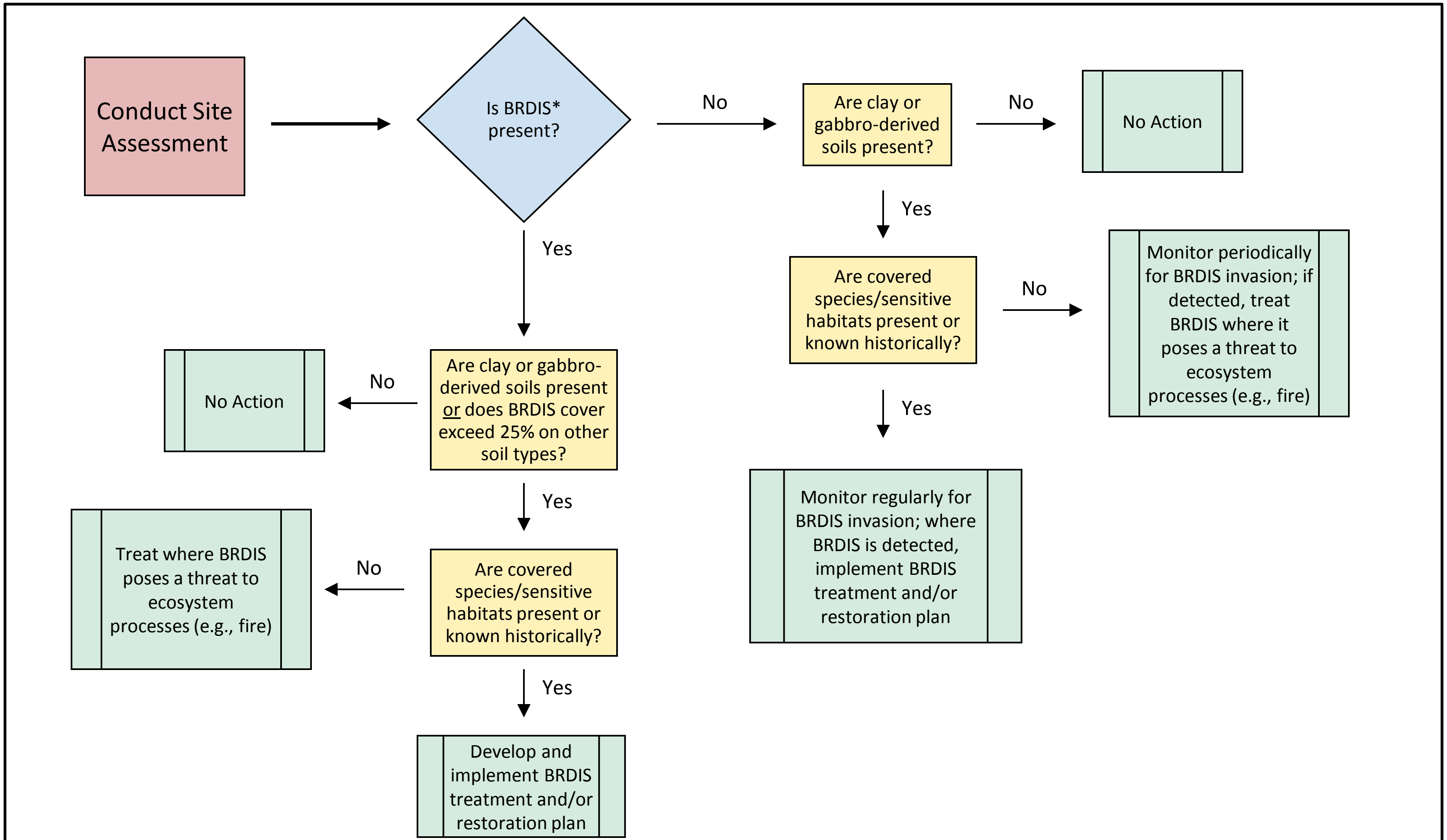
This experiment provides an important baseline of data, and adding further years of treatment and monitoring will only increase their value. However, the real utility of these methods for management depends on how they can be scaled up.

- Pre-treatment cover estimates can be eliminated without losing information or power.
- In future seeding efforts, incorporate watering as a contingency measure, where feasible.
- Estimate species richness and composition from larger belts or areas to provide more precise information about changes in community composition.

- Continue monitoring seeded plots (5-7 years) to separate trend from inter-annual fluctuations.
- Continue treating seeded plots, as necessary, to maintain the low cover of nonnative species achieved in this study and provide suitable conditions for germination of native species.
- Dethatch treatment areas prior to herbicide or mechanical control to improve native species germination and growth, particularly where native propagules are introduced into the site.

Figure ES-2 provides a decision tree for treatment. Scale up future treatments by using large mowers or cooperative mules. Test additional methods to determine their effectiveness in controlling *Brachypodium*, such as grazing and burning. The topographic heterogeneity of many conserved areas in San Diego County limits the feasibility of some of these methods.

Figure ES-2
Brachypodium Treatment Decision Tree



*BRDIS = *Brachypodium distachyon*

Cost Analysis

Table ES-1 summarizes relative costs and treatment effectiveness for the Crestridge Ecological Reserve and South Crest.

Table ES-1

Brachypodium Treatment Costs and Effectiveness

Treatment (year)	Crestridge		South Crest	
	Cost/Acre[1]	Control[2]	Cost/Acre[1]	Control[2]
Dethatching[3] (2013)	$1,600	NA[4]	$1936-2,058	NA[4]
Fusilade (2013)	$445	93%	$306	99.5%
Fusilade (2014)	$843	97%	NA[4]	NA[4]
Glyphosate[5] (2013)	$112	NA[4]	$255	NA[4]
Glyphosate[5] (2014)	$178	NA[4]	$511	NA[4]
Mowing (2013)	$350[6]	99%	---	NA[4]
Mowing (2014)	$1,150	92%[7]	---	NA[4]

[1] Approximate costs/acre = treatment costs. Costs were averaged where >1 treatment occurred per year. Costs include labor and field-associated expenses.

[2] Control = Effectiveness of *Brachypodium* control treatment in experimental treatment plots.

[3] Dethatching occurred in combination with other treatments and is included only for costs/acre. Refer to other treatments for overall effectiveness.

[4] NA = not applicable.

[5] Glyphosate does not affect *Brachypodium* cover, but is included in the table for approximate treatment costs/acre.

[6] The 2013 mowing event followed dethatching, which greatly reduced the amount of standing biomass and dethatching effort.

[7] Lower *Brachypodium* control in 2014 versus 2013 is believed to be due to a post-mowing germination event; differences are not statistically significant.

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Appendices

1. Introduction

Under an Environmental Mitigation Program grant from the San Diego Association of Governments (SANDAG), the Conservation Biology Institute (CBI) worked with a number of project partners in the San Diego region (Endangered Habitats Conservancy [EHC], Earth Discovery Institute [EDI], City of San Diego, San Diego Management and Monitoring Program [SDMMP], San Diego State University's Institute for Ecological Monitoring and Management [IEMM], Soil Ecology and Restoration Group [SERG], RECON Environmental, Inc. [RECON], and RECON Native Plant Nursery [RNP]), to conduct a comprehensive review of the nonnative invasive grass, *Brachypodium distachyon* (*Brachypodium*), and test experimental *Brachypodium* control treatments. Experimental treatments were conducted on the Crestridge Ecological Reserve (CER) and the South Crest properties (South Crest) in Management Unit (MU) 3 of the Management Strategic Planning Area (MSPA) (SDMMP 2013) in San Diego County, California (Figure 1).

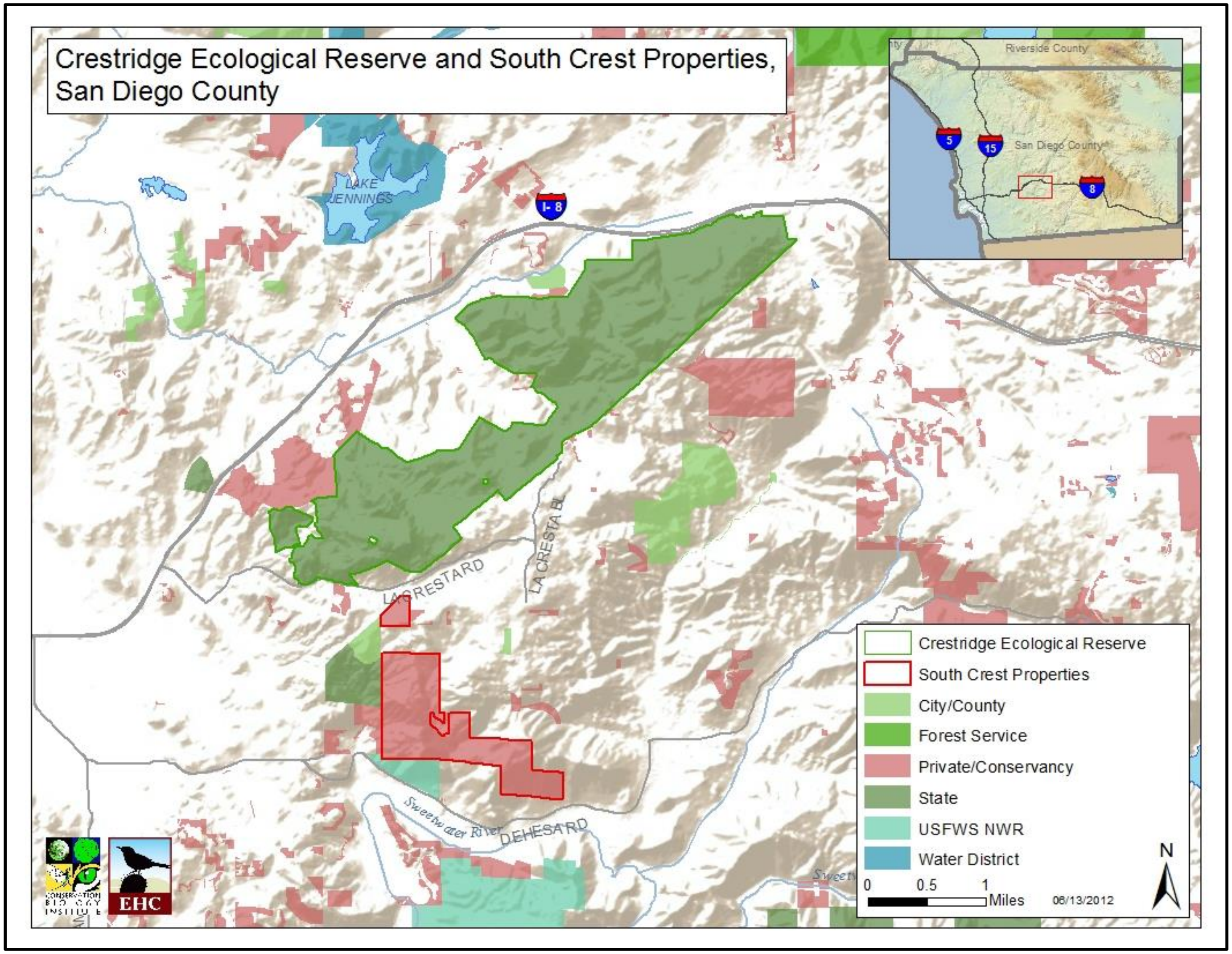
1.1 Purpose and Approach

Brachypodium is an emerging invasive species with potentially widespread ecological implications for native species, habitats, and ecosystem processes. In southern California, *Brachypodium* has increased in extent and dominance in recent years, possibly in response to fires and climatic conditions. The species can form nearly monotypic stands characterized by a thick and persistent thatch layer that suppresses germination of annual species and may affect recruitment (and thus, long-term persistence) of perennials and geophytes. *Brachypodium* appears to out-compete native and other non-native species for resources, potentially alters soil ecology and vegetation community structure and composition, and may contribute to a grass/fire cycle and habitat-type conversions. This species is particularly dense on restricted soils and, thus, threatens edaphic endemic plants such as *Acanthomintha ilicifolia*, *Bloomeria clevelandii*, *Brodiaea filifolia, Brodiaea orcuttii, Deinandra conjugens*, *Dudleya variegata*, *Nolina interrata*, and *Tetracoccus dioicus*. Covered species and focal habitats addressed in this project include *Acanthomintha ilicifolia*, *Nolina interrata*, *Dudleya variegata*, native grassland, and coastal sage scrub.

This project represents a continuum in management and monitoring efforts on the subject properties. CBI conducted baseline covered and invasive species mapping on CER and South Crest between 2009 and 2012 (CBI 2009, 2011a,b, 2012a); these studies mapped the location of covered species, identified *Brachypodium* as a potential threat, and mapped the extent of the *Brachypodium* invasion on the subject properties. In 2012, CBI conducted a pilot experimental control project for *Brachypodium* on CER (CBI 2012a). The current project builds off these earlier efforts by using data and results from those studies, along with information collected in the current study, to develop control strategies and treatment plans. In addition, the City of San Diego contributed data on *Brachypodium* presence and cover at covered species monitoring sites

Figure 1

Study Area and Subject Properties



throughout the region; these data, along with data contributed by CBI, were used by the San Diego Management and Monitoring Program (SDMMP) in developing a *Brachypodium* habitat suitability model.

The approach used in the *Brachypodium* project included the following components:

- Develop models to guide restoration plan development, identify monitoring targets, and provide predictive tools for early detection.
- Conduct habitat assessments to prioritize areas for treatment and restoration.
- Develop and implement site-specific restoration plans, including an experimental design to test the effectiveness of different treatment strategies.
- Collect, bulk, and purchase seed for restoration.
- Assess success rates and costs per acre of alternative *Brachypodium* control and restoration methods.
- Develop BMPs for *Brachypodium* control, based on results to date.
- Identify next steps for *Brachypodium* control on subject properties.
- Identify next steps for refining *Brachypodium* BMPs, including key research questions.

Appendices A – H provide detailed results or supporting documentation for many of the project elements discussed in this document. In addition, maps and documents can be viewed and downloaded from CBI's Data Basin website (http://databasin.org/); refer to the San Diego Conservation Group, *Brachypodium* and Supporting Documents folders.

1.2 Relationship to Regional Plans

The effort to control *Brachypodium* on conserved lands in San Diego County has a direct relationship to two regional plans: the Management Strategic Plan (MSP) (SDMMP 2013) and the Invasive Plant Strategic Plan (IPSP) (CBI et al. 2012). The former plan provides specific objectives for management of covered species, including (for many species) invasive species control. This project develops and refines Best Management Practices (BMPs) for *Brachypodium*, which was identified as a threat to a number of covered species in the MSP. The IPSP identified *Brachypodium* as a Management Level 4 species of particular concern because of its impacts to covered species and, particularly, narrow endemic species. Management Level 4 species require directed management at the sub-management unit or preserve-level, and control efforts are for the benefit of NCCP resources (CBI et al. 2012). The IPSP identified *Brachypodium* as a top tier stressor, or stressor with the potential to exert the most detrimental effects on narrow endemic species or their habitats (CBI et al. 2012). This project incorporates several IPSP recommendations for *Brachypodium*, including:

- Eliminate the species from invaded habitat or reduce species' cover so that it becomes a subdominant component of the vegetation.
- Incorporate experimental design into treatments to test alternative control methods and applications.
- Document effective control methods for replication at other sites.
- Restore native habitat components subsequent to treatment to minimize invasion pathways.

2. *Brachypodium* Biology, Threats, and Invasion History

2.1 Biology

Brachypodium consists of three distinct cytotypes (2n=10, 2n=20, 2n=30). A recent, comprehensive systematic study of the *Brachypodium distachyon* complex supports the description of two novel species, *B. stacei* (2n=20) and *B. hybridum* (2n=30), while retaining *B. distachyon* for the 2n=10 lineage (Catalan et al. 2012). Based on ploidy level, California plants may fall under *B. hybridum* (Bakker et al. 2009); however, we retain the specific epithet used in the Jepson Manual (Baldwin et al. 2010) until formal recognition.

Brachypodium is a small, fast-growing annual grass that is native to southern Europe and Eurasia (Piep 2013, Bakker et al 2009). The species is characterized by a short life cycle and small genome (Schwartz et al. 2010, Bakker et al. 2009, Opanowicz et al. 2008, Draper et al. 2001). Because of these traits, *Brachypodium* has been identified as a model grass for crop genetics (Mur et al. 2011, Vogel and Bragg 2009, Watt et al. 2009, Garvin et al. 2008, Olsen et al. 2006, Hasterok et al. 2004). The same traits that make it an ideal model species are also attributes of a successful invader (Bakker et al. 2009). For example, a short life cycle combined with rapid growth provides a competitive advantage by allowing for multiple life cycles during a growing season (Basu et al. 2004). Species genetics can also contribute to invasion success (Bakker et al. 2009). Some of the most successful weed species are polyploids (Bakker et al. 2009, Soltis and Soltis 1999, Soltis and Soltis 2000, Lee 2002), which have the potential to increase their genetic diversity through recombination of multiple chromosome sets (Bakker et al. 2009). California populations of *Brachypodium* appear to be tetraploids (2n=30), whereas the species exhibits diploid and tetraploid races in its native range in Eurasia (Bakker et al. 2009).

As an annual species, *Brachypodium* reproduces primarily by seed. It is self-fertile (Schwartz et al. 2010, Bakker et al. 2009, Opanowicz et al. 2008, Draper et al. 2001), with a typical life cycle of less than 4 months (Opanowicz 2008, Draper et al. 2001). Throughout its natural and introduced range, flowering time has been reported as between 3-4 weeks without a vernalization requirement, to more than 8 weeks following 6 weeks or more of vernalization. Tetraploids generally lack vernalization requirements (Opanowicz et al. 2008), and the southern California population may additionally represent an early flowering phenotype (Bakker et al. 2009). In studies on diploid accessions of *Brachypodium* from the Middle East, germination of fresh seed was strongly inhibited by blue light (found at the soil surface), while red light (found in the soil layer immediately below the surface) strongly promoted germination. This controlling effect of light on dormancy eventually faded in after-ripened seed (Barrero et al. 2011).

Florets are primarily gravity-dispersed, falling near the parental plant, but can be dispersed greater distances by animals, vehicle tires, mountain bikes, and other human activities (Bakker et al. 2009, DiTomaso and Healy 2007, Carr et al. 1992, Gordon-Reedy pers. obs.). Some researchers consider vertebrates to be the main dispersal agent of *Brachypodium* seed (Crossman et al. 2011). Seed bank persistence is presumed to be short (e.g., less than one year), although stored seed shows little loss of viability over four years (Gordon-Reedy pers. obs.). Individual plants are killed by fire (Brown and Bettink 2010), but the species appears to be able to recolonize quickly and spread in extent post-fire.

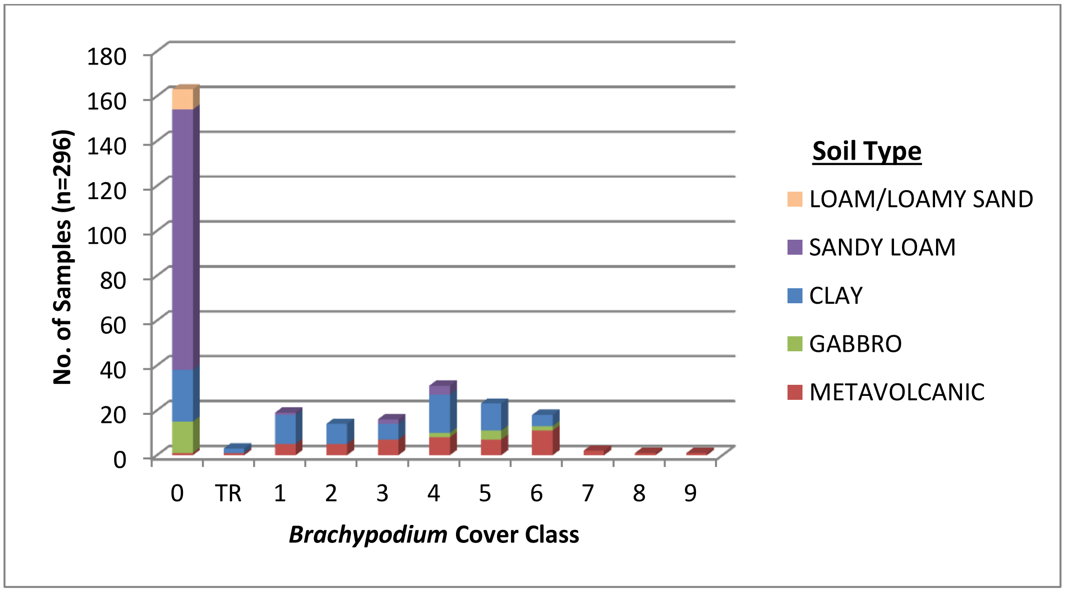
2.2 Threats

Brachypodium can become dominant in grasslands and the understory of shrubs and oak woodland, forming monospecific stands that limit establishment of native species and outcompete and exclude native herbs and grasses (Brown and Bettink 2010, Gordon-Reedy pers. obs.). The species forms a thick litter layer and thus, has the potential to alter fire regimes (Brown and Bettink 2010, D'Antonio and Vitousek 1992), as well as nutrient cycles. In studies on the Sweetwater National Wildlife Refuge in southern San Diego County, Wolkovich et al. (2010) found that invasive grasses (including *Brachypodium*) greatly increased carbon (C) and nitrogen (N) storage pools in the soil, acting as sinks for these elements, while the added litter increased above-ground native and non-native biomass due to greater inputs (invasive grasses), slower decomposition rates of grass versus shrub litter, and shading effects of grass litter which reduced decomposition rates of both non-native and native litter. Changes in C and N storage were linked to increases in the soil fungi:bacteria ratio, increased plant inputs, and decreased litter loss. Wolkovich et al. (2009) demonstrated that litter addition facilitated non-native grass growth, suggesting a positive feedback mechanism for invasion success. This study also demonstrated that invasive grass litter may benefit native shrubs by altering soil moisture, but did not examine the effects of shrub regeneration (e.g., seedling germination and growth) under conditions of high grass litter.

Brachypodium density may be related, at least in part, to soil type. In San Diego County, dense stands often occur on restricted soil types, such as clay and gabbro-derived soils (CBI et al. 2012), which also support rare plant species. CBI and partners collected *Brachypodium* cover data at multiple sites in San Diego County, in conjunction with rare plant or habitat assessments (Miller pers. comm., CBI 2012a,b), and assessed these data with respect to soils (Figures 2, 3). Although data are not comprehensive and represent only a 'snapshot' in time, they support initial observations that (1) *Brachypodium* currently forms dense stands on clay and gabbro-derived

Figure 2

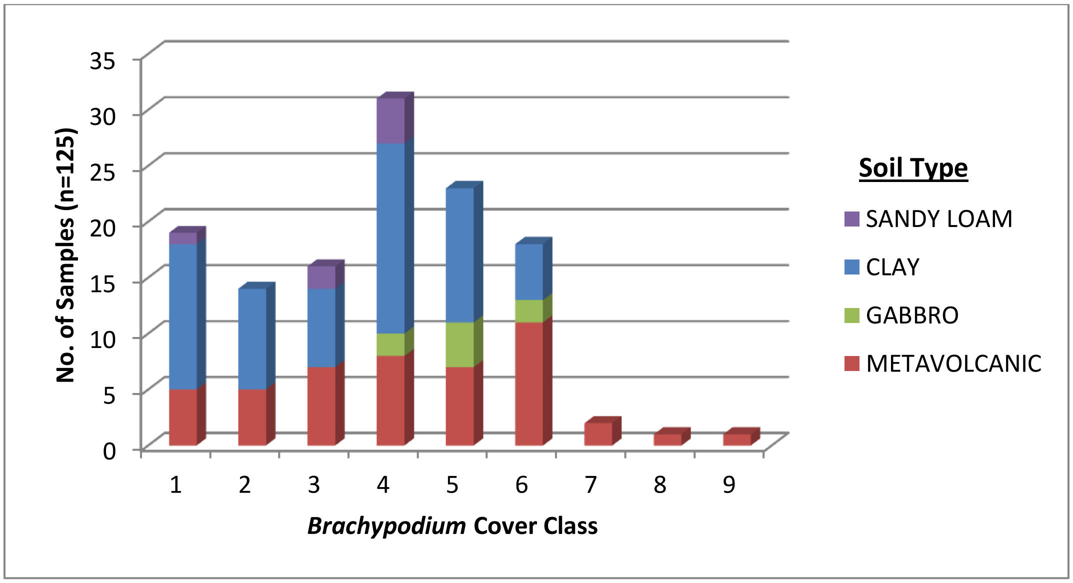
Brachypodium on All Sampled Soil Types[1]



[1] *Brachypodium* cover classes: 0 = absent; TR (>0-<1%), 1 (1-5%); 2 (5-10%); 3 (10-25%); 4 (25-50%); 5 (50-75%); 6 (75-90%); 7 (90-95%); 8 (95-99%); 9 (99-100%).

Figure 3

Brachypodium on Selected Sampled Soil Types[1,2]



[1] *Brachypodium* cover classes: 0 = absent; TR (>0-<1%), 1 (1-5%); 2 (5-10%); 3 (10-25%); 4 (25-50%); 5 (50-75%); 6 (75-90%); 7 (90-95%); 8 (95-99%); 9 (99-100%).

[2] Includes only sites with ≥1% cover of *Brachypodium*.

soils, (2) *Brachypodium* density on sandy or loam soils is generally low, and (3) there are areas with the potential to support high densities of *Brachypodium* (e.g., clays, gabbros) that have not yet been invaded. Dense stands of *Brachypodium* were also observed on soils derived from metavolcanic rock (e.g., San Miguel-Exchequer series), which can have an acidic clay subsoil (USDA 1973). The relationship between *Brachypodium* and sensitive plant species on metavolcanic-derived soils warrants further investigation. This information is preliminary and included only to guide monitoring and management efforts. Additional studies that assess soil properties and refine soil mapping would be valuable in assessing *Brachypodium* invasion risk.

Although work to date has focused on plant species, it is probable that *Brachypodium* adversely affects some animal species, as well (e.g., insects, reptiles, small mammals, and possibly, birds) through habitat degradation and loss of food sources.

2.3 Invasion History

Brachypodium was first documented in California in 1929 (Alameda County) and was first reported in San Diego County in 1950, when it was collected in a canyon in Carlsbad (CCH 2014).[1] The second county collection was in 1952 at Sweetwater Lake, and the species was collected just south of Torrey Pines in 1958 (CCH 2014). By the 1970s, Munz (1974) described the distribution of *Brachypodium* in southern California as ‘becoming established occasionally as at Santa Catalina Island and near Torrey Pines Park.’ The next county collections occurred near Peñasquitos High School in 1977 and Mission Bay in 1978 (CCH 2014). *Brachypodium* was collected only occasionally in San Diego County in the early 1980s. Beauchamp (1986) reported it as ‘uncommon in disturbed areas in Escondido, Carlsbad, Peñasquitos Canyon, Mission Bay, and Torrey Pines Mesa.’ Collection locations expanded in the 1990s to Camp Pendleton, Mission Trails Regional Park, Cowles Mountain, and Marine Corps Air Station (MCAS) Miramar (CCH 2014).

Collections of this species in the county increased in the 2000s due, in part, to intensified collection efforts in 2003, under the direction of Dr. Jon Rebman at the San Diego Natural History Museum (SDNHM 2014), and in 2009, as part of the SANDAG-funded vegetation mapping project, conducted by the California Department of Fish and Wildlife (CDFW) and AECOM. Several local botanists reported becoming aware of this species in the late 1990s-early 2000s (e.g., Vinje pers. obs., Lacy pers. comm., Spiegelberg pers. comm., Gordon-Reedy pers. obs.). In addition, *Brachypodium* was not mentioned as an associate of clay-endemic rare plant species in CNDDB records or reports from the 1980s and 1990s (e.g., CNDDB 2013, Bauder et al. 1994, Bauder and Sakrison 1997, Bauder and Sakrison 1999), but was regularly noted as an associate or dominant species at some of these same sites by the mid-2000s (e.g., City of

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[1] It is interesting to note that *Brachypodium* was not included in the 1949 annotated list of San Diego County plants (Higgins 1949).

Diego 2006, USFWS 2009). The species may have reached a threshold density during this time period where it became more noticeable among other nonnative grasses and forbs. *Brachypodium* superficially resembles some brome grasses (e.g., *Bromus hordeaceus*), as indicated by its common name, and it is conceivable that early and sparse infestations were overlooked or misidentified. Roberts (2008) also suggests that *Brachypodium* became widely established in Orange and San Diego counties during the last two decades.

Based on field observations and aerial imagery,[2] *Brachypodium* appears to have increased dramatically in extent in some San Diego County wildland areas after the large 2003 and 2007 wildfires, likely in response to post-fire gaps in vegetation and reduced competition. Sproul et al. (2012) recognized both *Brachypodium distachyon* and *Bromus* (*diandrus*, *hordeaceus*)-*Brachypodium distachyon* Semi-Natural Stands in San Diego County. The species' progression from 'uncommon' in the 1980s to identifiable vegetation types by 2010 is further indication of its increasing dominance in the region. Sproul et al. (2012) and others also recognized that this species was most dominant on clay soils.

Based on evidence to date, we believe *Brachypodium* has been present in San Diego County for over 60 years, and likely followed a typical invasion curve wherein it persisted at fairly low levels for decades before increasing in wildland areas. Figure 4 illustrates the collection history of *Brachypodium* in San Diego County (based on herbarium and Calflora records), which may or may not approximate the distribution of this species on the landscape.

3. Brachypodium Modeling

In developing *Brachypodium* control and management strategies, we conducted a comprehensive literature review and assembled conceptual life history, ecological, and management models for this species. These models synthesized information from a variety of sources and were intended to identify:

- Life history parameters conducive to manipulation and that may explain the invasion success of this species
- Observed or potential routes of establishment
- Observed or potential impacts to native species and ecosystem processes
- Monitoring targets
- Potential management actions
- Critical uncertainties with respect to both species persistence and effective management

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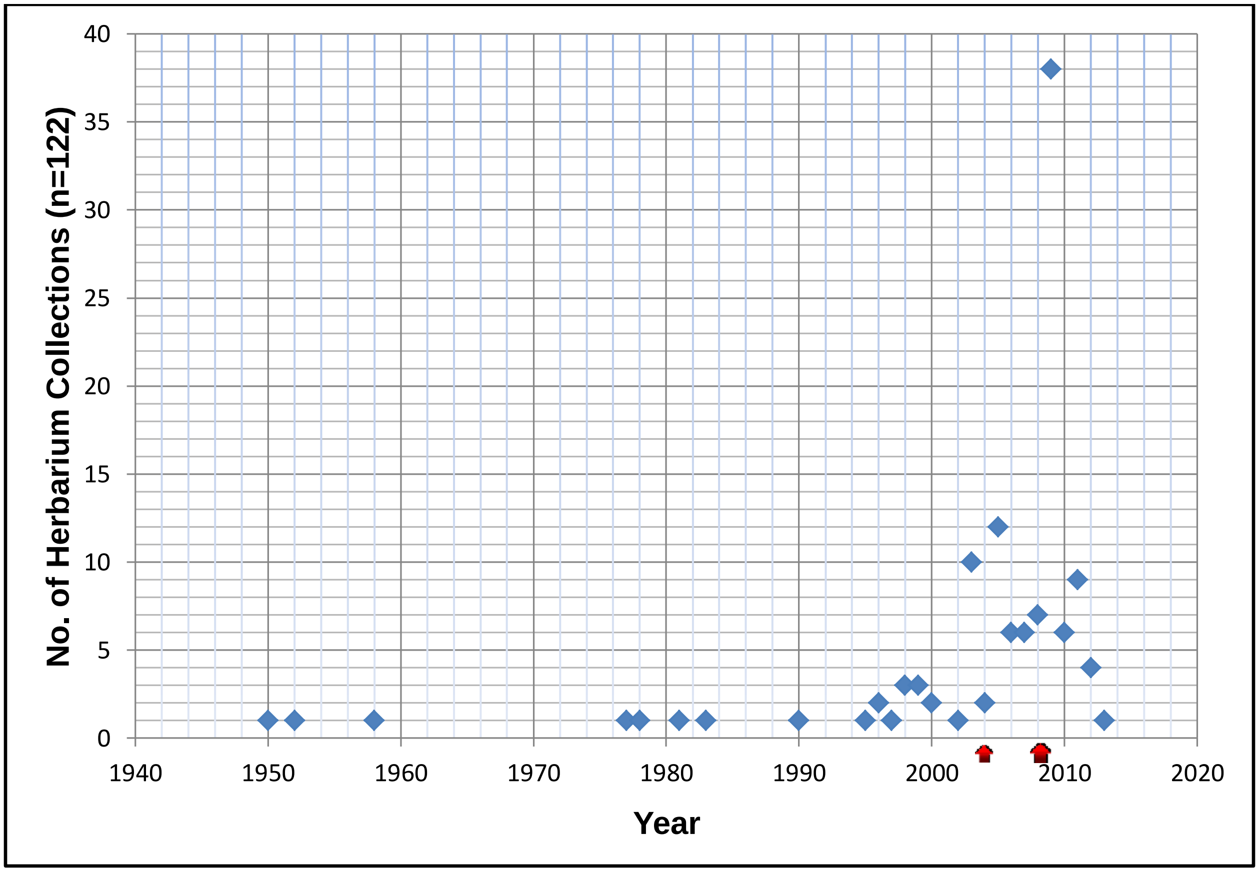
[2] *Brachypodium* is lime-green in spring when plants are actively growing. This makes it relatively easy to identify from aerial imagery when general infestation boundaries and species composition at a site are known; it is unknown whether this signature can be reliably identified in the absence of site-specific information.

Figure 4

Brachypodium distachyon Collections in San Diego County (1950-2012)[1,2,3,4,5]



[1] Sources: CCH (2014), San Diego County Plant Atlas, Calflora (2014).

[2] In 2002 (red arrow), the San Diego Natural History Museum launched the Plant Atlas Program, which likely contributed to an increased number of collections.

[3] In 2009 (red arrow), the California Department of Fish and Wildlife and AECOM conducted vegetation mapping in San Diego County; 35 of 38 (92%) of 2009 records are associated with that project.

[4] Records for 2012-2013 may be incomplete due to processing time.

[5] Duplicate herbarium collections were not included in total number of collections.

In addition, CBI worked with the SDMMP and the City of San Diego to develop a habitat suitability model for *Brachypodium* as a predictive tool for land managers. All models are discussed below with respect to characteristics and use in formulating management hypotheses or actions.

3.1 Conceptual Life History Model

Brachypodium is an annual species with a life history that follows a simple trajectory common to all annual plants. It is the details of the growth cycle, however, which provide insights into the competitive advantage and invasion success of this species, and identify potential points within the cycle that may be conducive to control. The life history model is presented in Figure 5; refer to Appendix A for supporting documentation.

From this model and supporting documentation, the following, key issues were identified:

- *Brachypodium* is self-fertile and produces copious amounts of highly viable seed; thus, it has the potential to increase rapidly under optimal conditions.
- *Brachypodium* seed exhibits little to no dormancy and germinates quickly; therefore, the species may be able to use and/or monopolize resources to the detriment of other native and nonnative species.
- *Brachypodium* has a short-life cycle and seed may be asynchronous, i.e., the species has the potential to produce more than one cohort per season under optimal conditions.
- *Brachypodium* produces a thick, persistent thatch layer that suppresses germination of other native and nonnative species.
- Fresh *Brachypodium* seed exhibits highest germination rates in the dark. Thus, it may be self-perpetuating by creating conditions that are detrimental to other species (thatch), but favorable to its own persistence.
- The *Brachypodium* seed bank may be transient and concentrated largely on the soil surface and uppermost soil layers; thus, seed bank management may be an important control strategy for this species.

Based on the life history model, the following, potential management strategies were identified:

- Early treatment of new infestations, with eradication as the goal, will be the most cost-effective control option.
- Where eradication is not feasible, continuous management will be necessary to keep *Brachypodium* populations at levels where they do not outcompete or suppress germination or growth of other species.
- Repeated, consecutive treatments (within and between seasons) will be necessary to reduce or limit inputs to the seed bank.
- Thatch removal may reduce the competitive advantage of *Brachypodium*.

It is not known whether this species is self-limiting or experiences episodic pulses based on climatic conditions and/or the availability of gaps for colonization or spread.

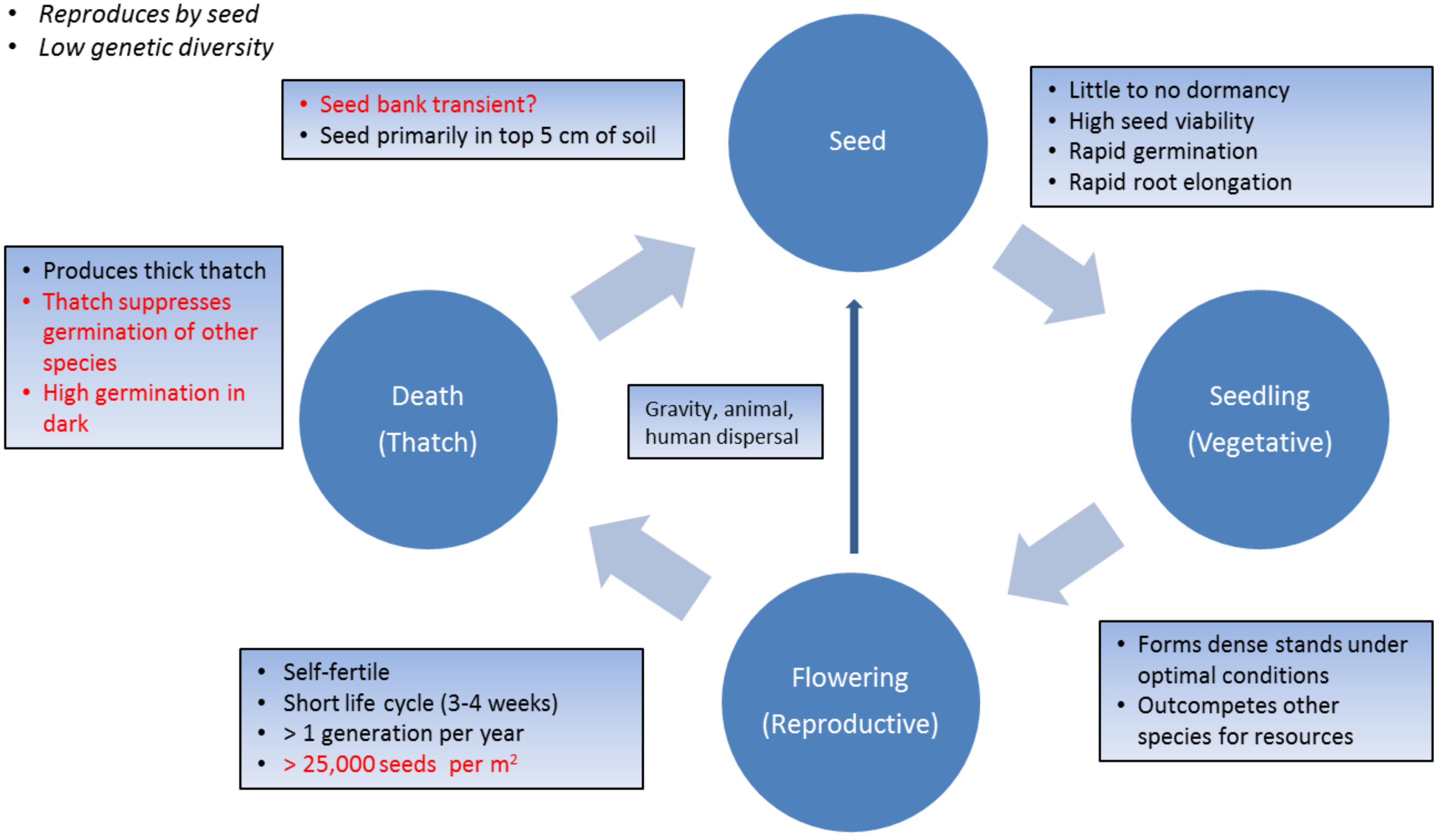
3.2 Conceptual Ecological Model

The conceptual ecological model focuses on anthropogenic and natural drivers of the ecosystem that contribute to *Brachypodium* establishment and spread, and presents observed or potential

Figure 5

Brachypodium distachyon Conceptual Life History Model

- *Annual*
- *Reproduces by seed*
- *Low genetic diversity*



- Seed bank transient?
- Seed primarily in top 5 cm of soil

Seed

- Little to no dormancy
- High seed viability
- Rapid germination
- Rapid root elongation

- Produces thick thatch
- Thatch suppresses germination of other species
- High germination in dark

Death
(Thatch)

Gravity, animal, human dispersal

Seedling
(Vegetative)

- Self-fertile
- Short life cycle (3-4 weeks)
- > 1 generation per year
- > 25,000 seeds per m^2

Flowering
(Reproductive)

- Forms dense stands under optimal conditions
- Outcompetes other species for resources

*Red = potential management opportunity

consequences of invasion (Figure 6). Refer to Appendix A for supporting documentation; information on environmental correlates is also included in Sections 2.2 and 3.4.

Based on the ecological model, the following issues were identified:

- *Brachypodium* invasion appears tied to disturbance that creates gaps in the vegetation matrix and presents opportunities for establishment.
- *Brachypodium* establishment may be influenced by soil type (see Figures 2, 3) and water availability.
- *Brachypodium* may alter soil ecology and utilize water resources to the detriment of other species.
- Dispersal agents (particularly, mammals) may contribute to the spread of *Brachypodium*.
- *Brachypodium* thatch may contribute to the grass-fire cycle.
- Dense stands of *Brachypodium* may alter native plant communities, reduce biodiversity, and reduce or eliminate habitat for wildlife or native plant pollinators.

Based on the model, the following, potential management strategies were identified:

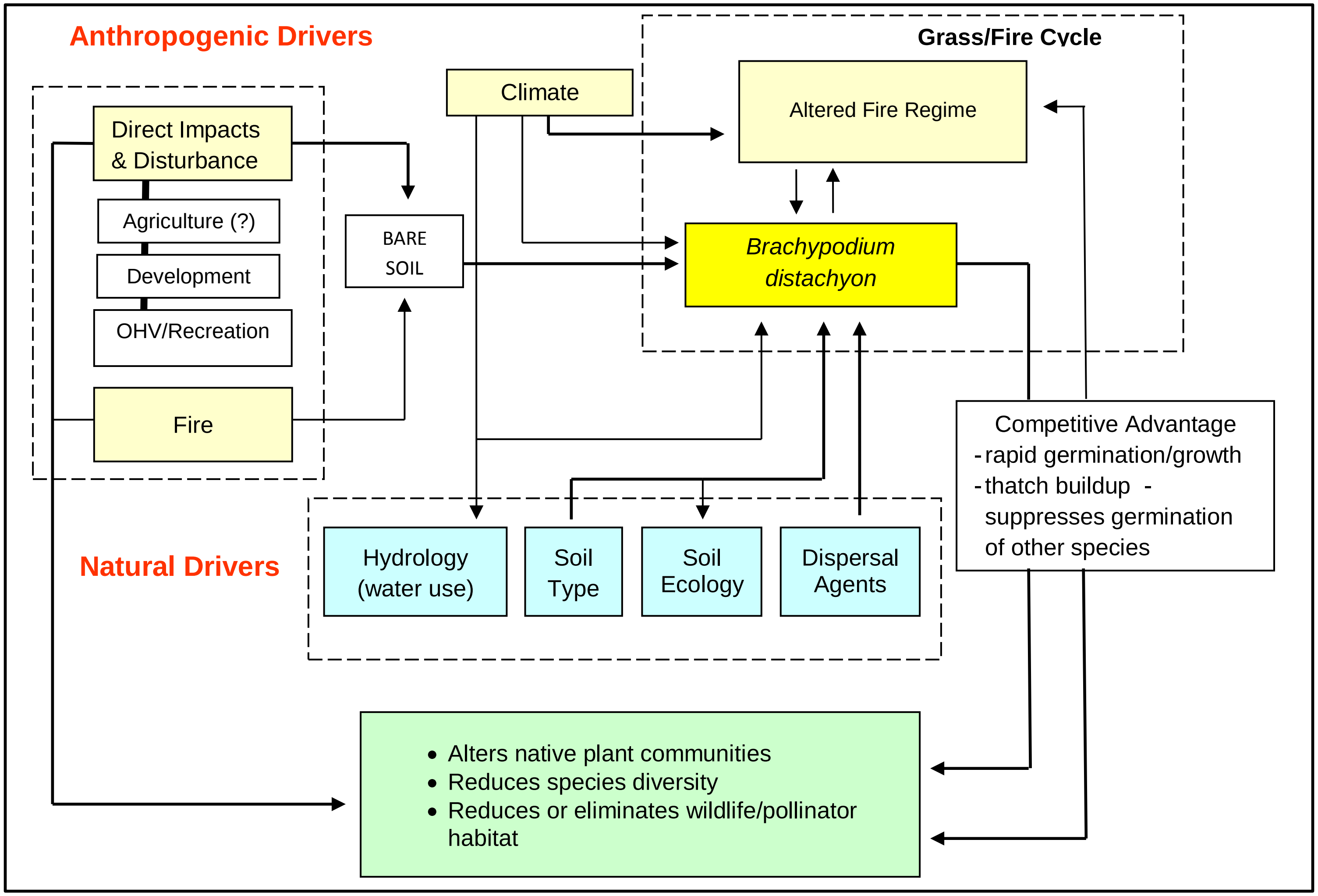
- Minimize disturbance or restore disturbed habitat on clay and gabbro-derived soils to reduce opportunities for *Brachypodium* establishment.
- Increase *Brachypodium* management following a disturbance event (e.g., fire), when the species might be present in low levels but has the potential to expand rapidly due to gaps and species' biology.
- Focus *Brachypodium* management in areas where the species might form dense stands (e.g., clay soils).
- Remove *Brachypodium* thatch to reduce biomass inputs to soil and fine fuel for fires, and increase habitat for wildlife or native plant pollinators.

3.3 Conceptual Management Model

The life history and ecological models were distilled into a simple *management* model to focus on those components most conducive to management and monitoring. This model also includes uncertainties suspected to drive invasion success, regardless of whether or not control actions are available. Development of the conceptual management model follows principles and format elucidated in Hierl et al. 2007 and refined by the Institute for Ecological Monitoring and Management (IEMM) in a conceptual model workshop (IEMM 2012) and species-specific models (Strahm 2012, Strahm et al. 2012). Per these sources, the following principles were incorporated into model development:

Figure 6

Brachypodium Conceptual Ecological Model



- Simpler models that represent the current state of knowledge and are supported by data are preferable to complex models with a high degree of uncertainty.
- Putative or secondary relationships should be differentiated from data-based primary relationships.
- The model should clearly identify management and monitoring goals.
- The model should include life history traits (species variables) that influence persistence, and focus on variables that may respond to monitoring and management.
- Proposed management actions should support the management goal; proposed monitoring should measure the effectiveness of management actions.

Also per the sources cited above, the following format was used to promote consistency among species conceptual models in the region:

- Management and monitoring goals are displayed at the top of the model (green and brown boxes, respectively).
- Anthropogenic drivers (change agents or stressors) are shown in pink boxes; natural drivers are in blue boxes.
- Elements outlined in red may be monitored to assess population status and effectiveness of management actions. Elements outlined in gray contribute to population status, but are not influenced by management actions.
- Elements in the green circle are *Brachypodium* life history traits (species variables) that can be measured to assess the response to management actions.
- Relationships between model elements are depicted with arrows. Black arrows depict direct or primary relationships; blue arrows depict secondary or putative relationships. The model focuses on primary relationships that are expected to affect population status and that may be influenced by management and monitoring.

The conceptual management model identifies general management and monitoring goals, and *Brachypodium* life history traits that contribute the most to detrimental effects and for which management actions may be available (Figure 7). Refer to Appendix A for supporting details. The model should be updated as additional data become available through research or monitoring. The model indicates that management actions should focus on:

- Reducing *Brachypodium* cover and increasing native species cover and richness.
- Reducing *Brachypodium* biomass (thatch).
- Reducing the *Brachypodium* seed bank and preventing further inputs to the seed bank.
- Restricting seed dispersal through BMPs to avoid inadvertently moving seed between sites.

Figure 7

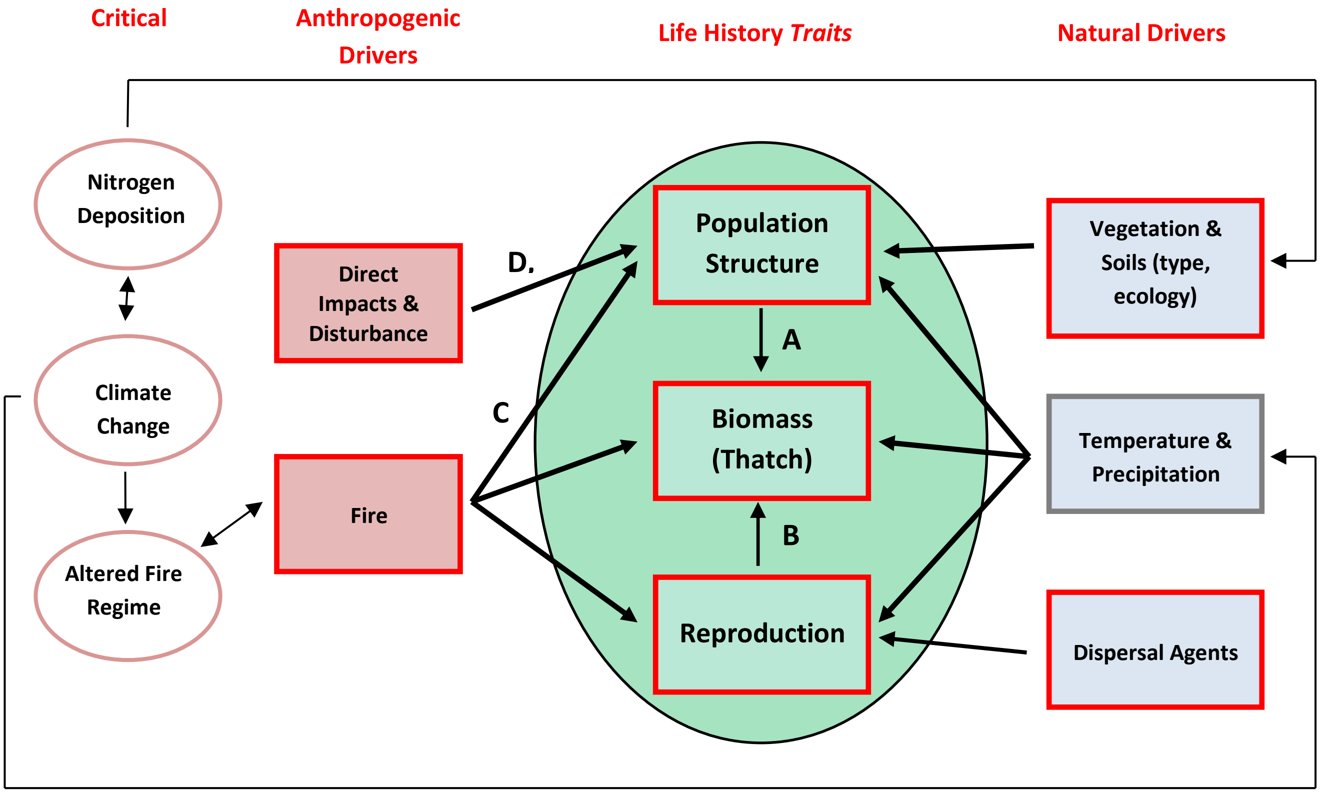
Brachypodium Conceptual Management Model

Brachypodium distachyon

Goal: Reduce or eliminate *Brachypodium* where it forms dense stands on conserved lands and threatens sensitive species and habitat persistence, and restore habitat to prevent future invasions by this species.

Monitoring

Monitor response of *Brachypodium* and native species to invasives control, access control, and habitat restoration.



Uncertainties

- BMPs for *Brachypodium* control
- Types of disturbances that are most problematic
- Soil affinities
- Seed bank longevity

Others: visualized in white bubbles

Management

A) Reduce/eliminate above-ground population
B) Minimize inputs to seed bank
C) Restore habitat to reduce gaps
D) Reroute roads/trails; prohibit/restrict recreational activities that promote seed dispersal
E) Exclude (excessive fire)

3.4 Habitat Suitability Model

The SDMMP (CBI 2014) developed a *Brachypodium* habitat suitability model using locational data from a variety of sources, including this project. Over 20 models were constructed and evaluated. The best-performing model (Figure 8) had a median validation Habitat Suitability Index (HSI) of 0.805 and median calibration HSI of 0.636. Based on available data, suitable habitat for this species in San Diego County occurs primarily west of the mountains, and overlaps with habitat for at least one covered species, *Acanthomintha ilicifolia* (CBI 2014). Environmental variables associated with *Brachypodium* are related to winter climate conditions, slope, and clay soils. Refer to CBI 2014 for a full description of the modeling process and results.

The habitat suitability model for *Brachypodium* over-predicts suitable habitat, which indicates the species has not yet saturated all available habitat niches in the county and is likely still expanding its distribution. The model may be refined as additional data are collected. Currently, it can be used as a predictive tool by land managers to (1) identify conserved lands at risk for *Brachypodium* invasion and (2) implement early detection programs or management measures for eradication or containment.

3.5 Climate Change Model

The California Invasive Plant Council (Cal-IPC) developed a climate change model for the invasive grass, *Brachypodium distachyon*, which has been identified as a threat to San Diego thornmint. Model results for 2050 predict that the range of this species in San Diego County will largely remain stable or expand to the east, with some range reductions in coastal and central areas (Figure 9) (Cal-IPC 2012). This suggests that the species may continue to be a management issue in many areas of the MSP for the foreseeable future.

4. *Brachypodium* Control Program

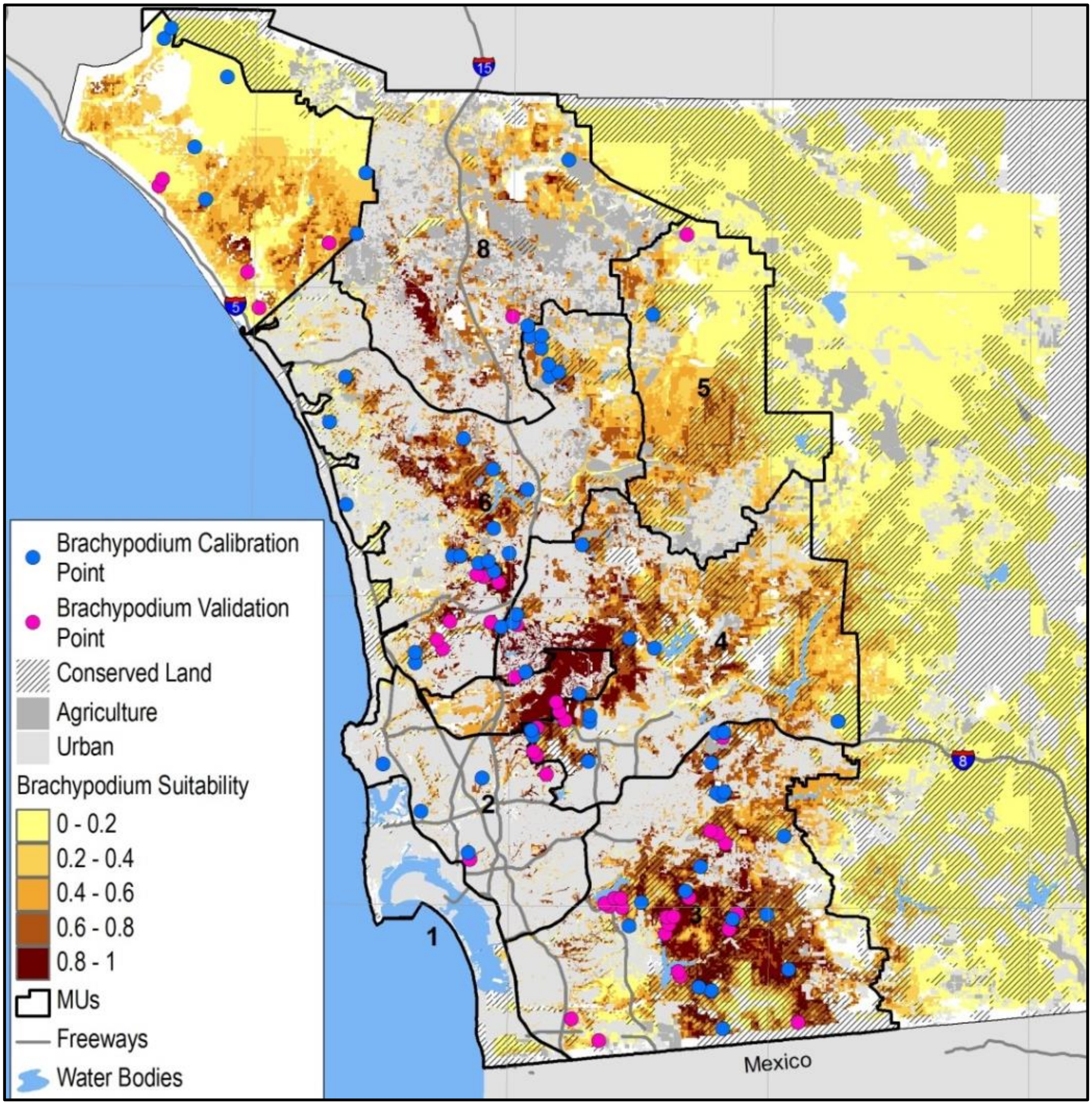
The *Brachypodium* control program included pre-restoration site assessments to establish baseline conditions for potential restoration areas and prioritize areas for treatment, and site restoration, including site preparation, invasive control treatments, reintroduction of native species, and site protection.

4.1 Site Assessment

CBI conducted qualitative and standardized habitat assessments on CER and South Crest to document existing habitat conditions and level of *Brachypodium* infestation. We used these data to prioritize areas for treatment and restoration, based on habitat suitability for both target resources and restoration sites.

Figure 8

Brachypodium Habitat Suitability on Conserved Lands in San Diego County[1,2]



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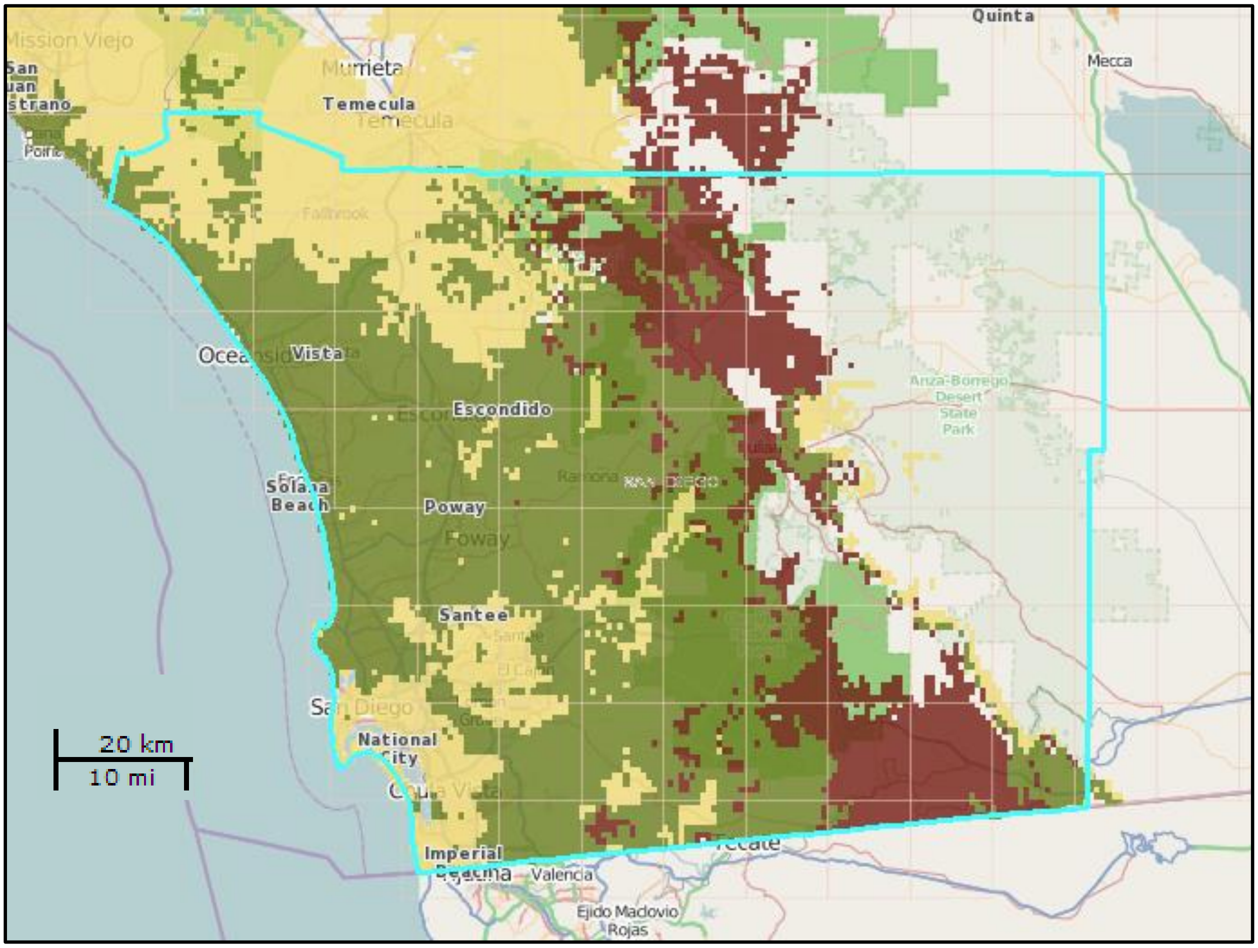
[1] White areas were not modeled due to a lack of soil data.

[2] Source: SDMMP *in* CBI 2014.

Figure 9

Brachypodium distachyon Change Prediction Model (2010-2050)[1]



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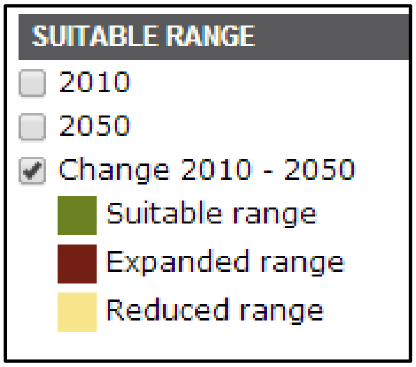
[1] Source: Cal-IPC 2012.



SUITABLE RANGE

☐ 2010

☐ 2050

☑ Change 2010 - 2050

🟩 Suitable range

🟥 Expanded range

🟨 Reduced range

4.1.1 Methods

Habitat assessment methods used in this project were developed by The Nature Conservancy (TNC) and refined by CBI, TNC, and San Diego State University (SDSU) for the South County grasslands project in southern San Diego County (CBI 2012b). The habitat assessment process collects information on biotic, abiotic, and management variables to determine both ecological suitability and management feasibility for restoration purposes. Prior to conducting habitat assessments, CBI reviewed soil maps, aerial photographs, results of previous vegetation mapping, and species occurrence data in the project areas and vicinity.

Habitat assessments were focused in areas of CER and South Crest that supported dense stands of *Brachypodium* and which supported or had the potential to support sensitive species and habitats. Not all *Brachypodium*-infested lands were included in these assessments, particularly on CER. The assessments were conducted in stands mapped in the field as discrete polygons. Stand size ranged from 0.38 acre to 4.4 acres. Following vegetation mapping protocols set forth by the California Native Plant Society (CNPS) and California Department of Fish and Wildlife (CDFW), (CNPS and CDFW 2011), we defined stands by both compositional integrity (i.e., similar species) and structural integrity (i.e., similar site history and environmental conditions). Visually, this combination of factors results in stand homogeneity. For analysis purposes, each stand included in the assessment process was maintained as a discrete polygon on maps, regardless of vegetation classification.

During the assessment process, CBI biologists systematically walked each assessment area to characterize and map vegetative condition and assess *Brachypodium* cover and presence of sensitive resources. For each polygon, biologists documented the attributes listed on the field assessment form (Appendix B-1). Copies of all habitat assessment forms and accompanying photodocumentation are maintained at CBI. In addition, data from all habitat assessment forms were entered into an Excel database (Appendix B-2) and used to map existing conditions and identify potentially suitable restoration sites.

4.1.2 Results

A total of 14 habitat assessments were completed, including 6 on CER and 8 on South Crest (Figures 10, 11). Table 1 provides a summary of polygon attributes for CER, and Table 2 provides a summary of attributes for South Crest.

4.1.3 Prioritization

We conducted habitat assessments over 11.5 acres on CER and 15.8 acres on South Crest. We prioritized approximately 20 acres of habitat for treatment and restoration where they (1) currently or historically supported covered species, (2) were adjacent to historic covered species localities and possessed many of the same habitat attributes, (3) were upslope from prioritized

Figure 10

Habitat Assessments, Crestridge Ecological Reserve

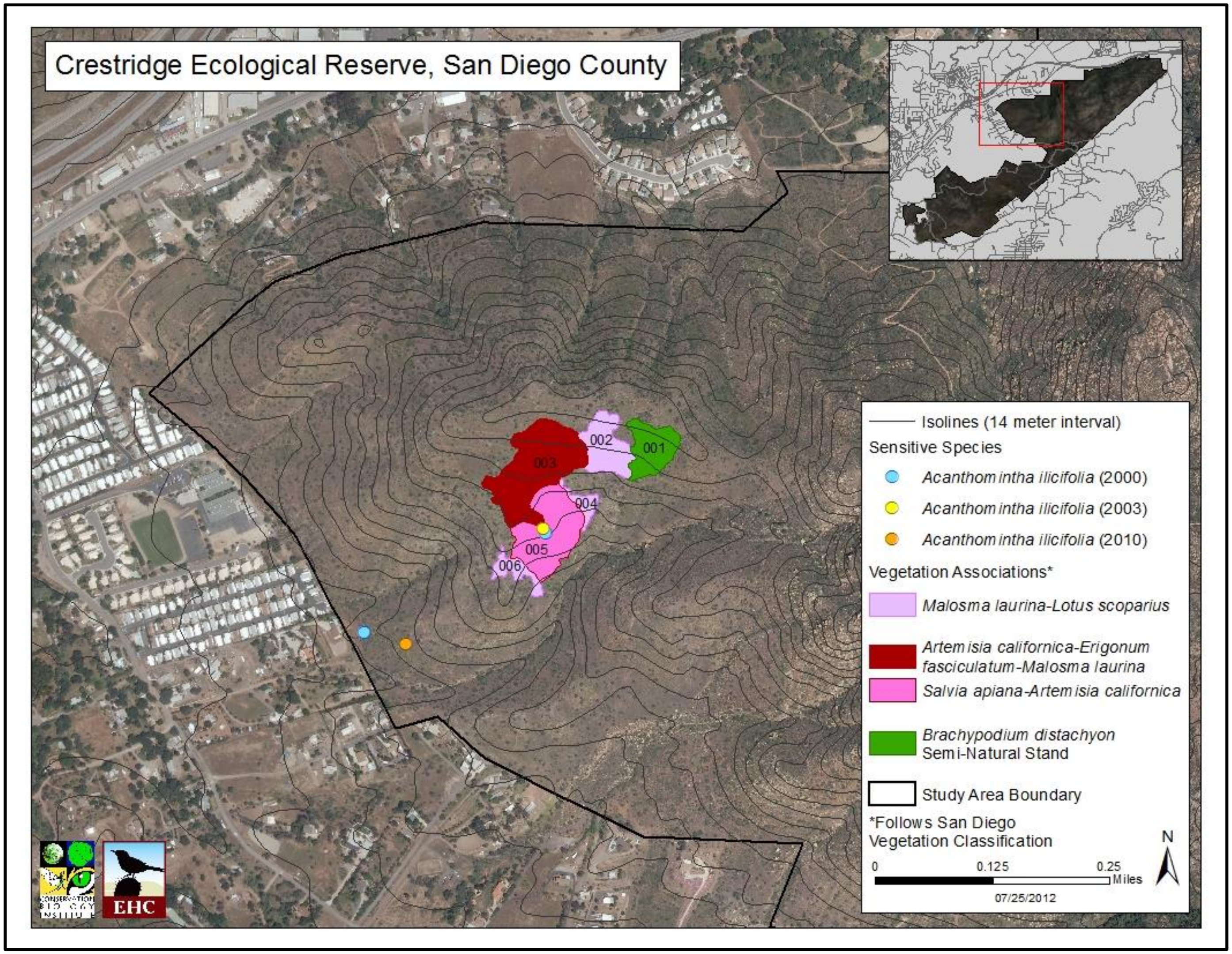


Figure 11

Habitat Assessments, South Crest Properties

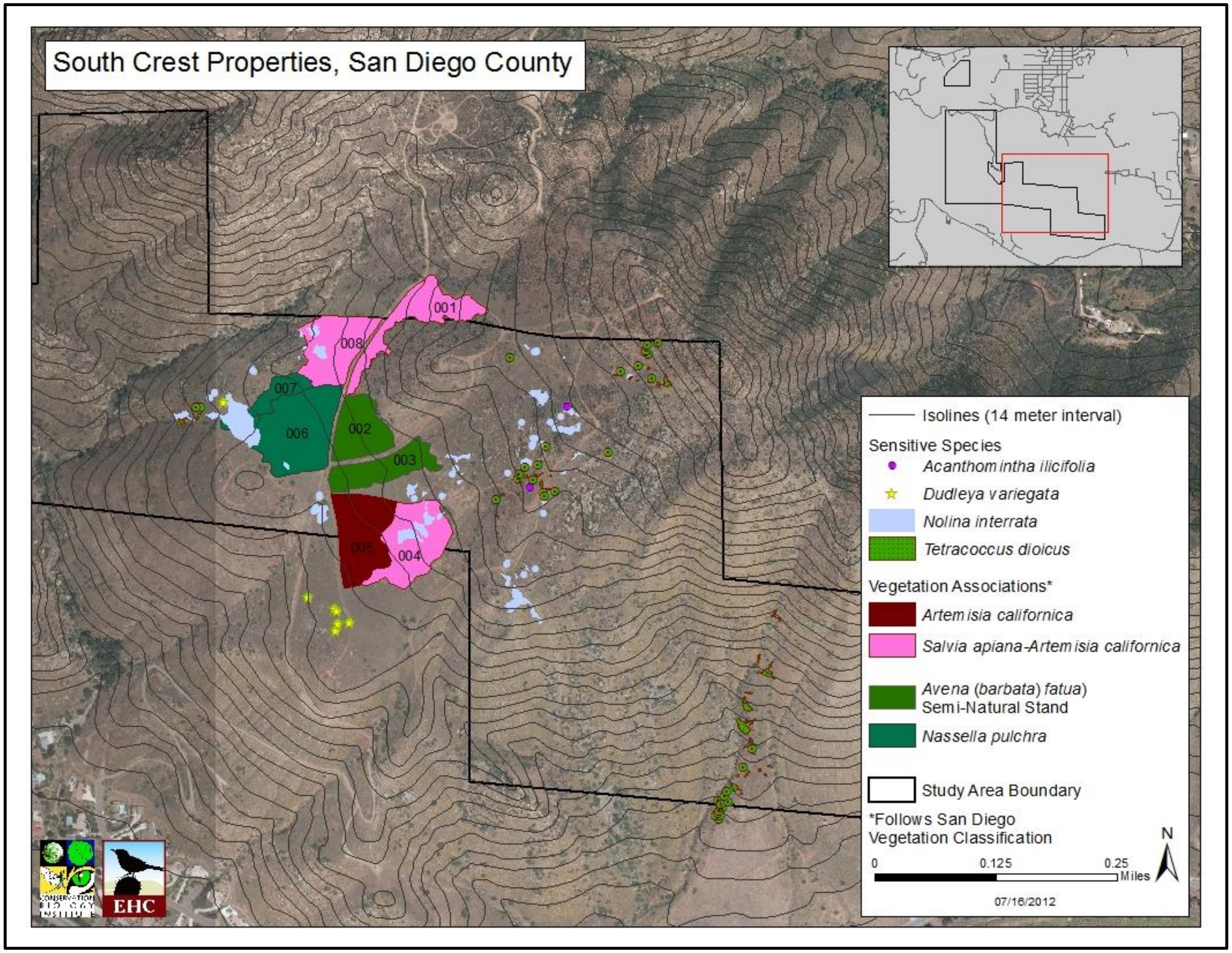


Table 1

Habitat Assessment Summary, Crestridge Ecological Reserve

Polygon	Attribute				
	Size (acres)	Slope	Soil Type[1]	Vegetation Association[2]	Target Species[3]
1	1.56	South, Southeast	Gabbro	*Brachypodium distachyon* Semi-Natural Stand	---
2	1.66	South, Southeast	Gabbro	*Malosma laurina-Lotus scoparius*	---
3	4.20	South, Southeast	Gabbro	*Artemisia californica-Eriogonum fasciculatum-Malosma laurina*	---
4[4]	0.38	Southwest	Gabbro	*Malosma laurina-Lotus scoparius*	---
5	3.09	South, Southwest	Gabbro	*Salvia apiana-Artemisia californica*	*Acanthomintha ilicifolia*
6[4]	0.59	South	Gabbro	*Malosma laurina-Lotus scoparius*	---

[1] Gabbro-derived soils are in the Las Posas series (USDA-SCS 1973).

[2] Vegetation associations follow Sproul et al. 2011.

[3] Target species indicates focus of habitat restoration effort; species may or may not be present in polygon.

[4] No management actions are planned in these polygons during this project; however, actions are specified in the restoration plan (Appendix B) in the event that funding becomes available to extend treatments in the future.

polygons and functioned as a source of invasive seed propagules, or (4) were highly disturbed, thus allowing for the full spectrum of treatment and restoration.

Areas prioritized for treatment in this project included CER polygons 1, 3, and 5 and South Crest polygons 1, 2, 3, 4, 5, and 8. The additional polygons (CER 2, 4, 6 and South Crest 6, 7) should be treated as funding becomes available.

4.2 Site Restoration

We developed site-specific restoration plans, including treatment strategies, management goals and objectives, and restoration specifications based on conceptual models and habitat assessment results (detailed in Appendix C). Although the plans include specifications for all assessed polygons, we implemented restoration (including site preparation, invasive control treatments, and selected seeding) for only a subset of the polygons based on available funding. Areas were prioritized for this project as discussed in Section 4.1.3.

The overarching goal of restoration was to allow plant communities to shift in a favorable direction, with the realization that 100% control of *Brachypodium* and other invasive species was

Table 2

Habitat Assessment Summary, South Crest

Polygon	Attribute				
	Size (acres)	Slope	Soil Type[1]	Vegetation Association[2]	Target Species[3]
1	0.75[4]	West	Clay	*Salvia apiana-Artemisia californica*	*Nolina interrata*
2	2.00	West	Clay	*Avena (barbata) fatua)* Semi-Natural Stand	---
3	1.86	West	Clay; Gabbro	*Avena (barbata) fatua)* Semi-Natural Stand	*Acanthomintha ilicifolia, Dudleya variegata*
4	1.74[4]	Southwest, West	Clay; Gabbro	*Salvia apiana-Artemisia californica* Association	*Nolina interrata, Acanthomintha ilicifolia, Dudleya variegata*
5	1.63[4]	South, Southwest	Clay	*Artemisia californica* Association	*Dudleya variegata*
6	4.40	West, Flat	Clay	*Nassella pulchra*	*Nolina interrata*
7	0.78	Southwest	Clay	*Nassella pulchra*	*Nolina interrata*
8	2.62	Northwest, West	Gabbro	*Salvia apiana-Artemisia californica*	*Nolina interrata*

[1] Clay soils are in the Auld series; gabbro-derived soils are in the Las Posas series (USDA-NRCS 2007).
[2] Vegetation associations follow Sproul et al. 2011.
[3] Target species indicates focus of habitat restoration effort; species may or may not be present in polygon.
[4] Acreage onsite; polygon extends offsite.

unlikely within the 2-year timeframe of this project. The following principles were followed in implementing this shift:

- Remove nonnative, invasive plants to create conditions under which native species can flourish; minimize potential for reinvasion of restored habitat; and increase *potential* habitat for covered species and other native plant species.
    - Decrease growth, propagule production, and frequency of dispersal of invasive species.
    - Manage seed bank of invasive species.
- Establish desirable (native) species that are functionally similar to the invader species (*Brachypodium*).
    - Increase germination, propagule production, and frequency of dispersal of native species.

- Enhance native plant seed bank through seeding.

Restoration plan components include site preparation, invasive control treatments, seed procurement and seeding, and site protection. Refer to Appendix C for the schedule of restoration activities.

4.2.1 Site Preparation

We delineated treatment areas by staking eight polygons. An estimated 5.4 acres of habitat was dethatched on CER (polygon 1) and South Crest (polygons 2 and 3). Dethatched polygons supported few native species; dethatching removed *Brachypodium* and other nonnative grass biomass (thatch). Although earlier experimental studies demonstrated no significant differences in *Brachypodium* control between dethatched and control (no dethatch) plots (CBI 2012a), dethatching was conducted where seeding was a restoration component. In these cases, thatch removal was hypothesized to enhance native species germination by improving contact between soil and seed and possibly, decreasing *Brachypodium* germination by increasing light conditions at the soil surface. We used line trimmers to dethatch these areas in November and December 2012. At CER, cut thatch was left in place. At South Crest, dethatched material was raked, removed from polygons, and placed in piles adjacent to restoration sites for composting. Refer to Appendix D for photodocumentation of the dethatching process.

4.2.2 Invasives Control

Invasives control included mechanical (mowing) and herbicide treatments, as discussed below. Refer to Table 3 for treatment combinations in each treated polygon.

<u>Mechanical Treatment</u>

Mechanical treatment consisted of mowing nonnative grasses in CER polygon 1 with a line trimmer prior to seed set, when *Brachypodium* was approximately 6 inches high. Litter was left in place. Mowing was conducted by SERG in March 2013 and by RECON in April 2014.

<u>Herbicide Treatment</u>

Herbicide treatments included application of both a grass-specific herbicide (e.g., Fusilade II) and spot treatments for nonnative forbs using a glyphosate-based herbicide (referred to in this document as glyphosate). The latter was in recognition that removing the nonnative grasses might ‘release’ nonnative forbs for germination, as has been observed with similar restoration projects (e.g., Cox and Allen 2011). Herbicide treatments varied between polygons with respect to number of applications per year (Table 3).

SERG applied herbicide at both sites in 2013 using backpack sprayers. The first Fusilade application was in February and the second was in March. Glyphosate was applied at both sites

Table 3

Restoration Treatments[1,2]

Polygon[2]	2012	2013				2014		
	Dethatch	Mechanical	Fusilade	Glyphosate	Seed	Mechanical	Fusilade	Glyphosate
CER_1	1x	1x	---	2x	1x	1x	---	2x
CER_3	---	---	1x	2x	---	---	1x	2x
CER_5	---	---	2x	2x	---	---	1x	2x
SC_1	---	---	1x	2x	---	---	1x	2x
SC_2	1x	---	2x	2x	1x	---	---	2x
SC_3	1x	---	2x	2x	1x	---	---	2x
SC_4	---	---	1x	2x	---	---	1x	2x
SC_5	---	---	1x	2x	---	---	1x	2x
SC_8	---	---	1x	2x	---	---	1x	2x

[1] Treatment combinations = Dethatch/Mechanical/Glyphosate/Seed; Fusilade (1x)/Glyphosate; Fusilade (2x)/Glyphosate; Dethatch/Fusilade/Glyphosate/Seed.

[2] CER = Crestridge Ecological Reserve; SC = South Crest.

in mid- to late March, after the second Fusilade application. Refer to Appendix E.1 for application dates, rates, area treated, and target species.

In 2014, RECON applied herbicide treatments at CER and selected areas of South Crest using backpack sprayers. RECON treated CER polygons 3 and 5 with Fusilade in mid-February and polygons 1, 3, and 5 with glyphosate in mid-March. On South Crest, RECON treated polygon 8 (exclusive of treatment plots) with Fusilade and polygons 2, 3, 4, 5, and 8 with glyphosate in early March. Treatments were applied using backpack sprayers. Refer to Appendix E.2 for application dates, rates, area treated, and target species.

Carl Bell of the University of California Cooperative Extension treated nonnative grasses in South Crest polygons 4 and 5, and polygon 8 treatment plots on February 14, 2014 using a ‘Cooperative Mule’ to test the cost and treatment effectiveness of this method versus backpack sprayers. The Cooperative Mule is an all-terrain vehicle with an herbicide spray tank with either booms or boomless spray nozzles (http://ucanr.edu/blogs/socalinvasives/index.cfm?start=6). Using the mule, Mr. Bell applied Fusilade DX at a rate of 24 ounces per acre in a spray volume of 10.5 gallons of water per acre. The mule was driven at about 5 mph and sprayed a swath of 30 feet. Refer to Appendix E.2 for application dates, rates, area treated, and target species.

4.2.3 Seed Procurement

At the time the restoration plans were developed, both CER and South Crest were inaccessible to vehicles and lacked a water source. Thus, seeding by hand was the only feasible option for introducing native plant propagules into restoration sites. Seed palettes were developed for each site (Appendix F-1) and local seed collected for bulking and out-planting, as described below. Additional seed was purchased from commercial suppliers to fill shortages in seed production.

Seed Collection

Seed was collected in 2012 and 2013 by CBI biologists, as well as citizen volunteers under the direction of Cathy Chadwick of EDI. Several volunteer seed cleaning events were held at Rancho Jamul Ecological Reserve (RJER) in 2012 and 2013. Collected seed was bulked at Recon Native Plant Nursery (RNP) to increase the amount of seed available for restoration or sown directly into restoration sites in Fall 2013. Refer to Appendix F-2 for a list of volunteer-collected seed; Figure 12 presents photos of seed collecting and seed cleaning events.

Seed Bulking

Seed bulking was conducted at RNP in southern San Diego County to increase the amount of local seed available for restoration. Seed was bulked from collections made on CER, South Crest, and other conserved lands in south San Diego County. CBI delivered field-collected seed to RNP in Fall 2012. Upon receipt, RNP cleaned (if necessary), stored, and propagated seed of seven native plant species (Figure 13): *Stipa pulchra, Stipa lepida, Aristida adscensionis, Corethrogyne filaginifolia, Cryptantha intermedia, Plantago erecta,* and *Salvia columbariae.*

Seed was sown into the ground or propagation plug trays or flats in December (*S. pulchra, S. lepida*) or January to mid-February 2013 (all other species). Mudflats and plugs containing seed were maintained under optimal growing conditions. After sowing, RNP staff monitored development of each species to assess germination rates and plant growth, and determine optimal timing for transplanting.

All species except *P. erecta* germinated and presented well with uniform development in general. *Plantago erecta* was re-sown on February 20, 2013 due to field-planting problems and thereafter demonstrated uniform germination rates and development. Initial germination rates were 90% for *C. filaginifolia*, 80% for *S. pulchra*, *S. lepida*, *C. intermedia* and *S. columbariae*, 75% for *P. erecta*, and 40% for *A. adscensionis*. Certain species grew quickly (*S. lepida, S. pulchra, C. intermedia*), while others grew more slowly (*A. adscensionis, S. columbariae*). All species were transplanted the first two weeks of April except *P. erecta*, which was sown directly in the ground in February and *A. adscensionis*, which presented difficulties on the rooting stage and was transplanted to 1 gallon containers on June 17, 2013. Seed was harvested as follows:

Figure 12

Volunteer Seed Collection and Seed Cleaning Events

A.

B.

C.

D.

A. Seed collecting on Crestridge Ecological Reserve, B-D. Volunteer seed cleaning event at Rancho Jamul Ecological Reserve. Photos provided by Cathy Chadwick, Earth Discovery Institute.

Figure 13

Target Species for Bulking

A. B. C.

D. E. F.

A,B. *Corethrogyne filaginifolia*, C. *Aristida adscensionis*, ready to harvest, D. *Cryptantha intermedia*, full bloom, E. *Plantago erecta*, F. *Stipa pulchra* and *Stipa lepida*. Photos provided by RECON Native Plant Nursery.

- *Salvia columbariae* completed its flowering cycle and seed was collected on May 21, 2013.
- *Cryptantha intermedia* and *P. erecta* were collected from late June to early July 2013.
- *Stipa pulchra and S. lepida* were collected over many events from early August to late November 2013 as the plants continued flowering after each harvest.

Corethrogyne filaginifolia did not perform to expectations and exhibited only vegetative growth during the 2013 season. Plants will be maintained at RNP off-contract for harvest in Fall 2014. Under the direction of CBI representatives, RNP staff collected *C. filaginifolia* seed from CER in mid-October 2013 to fulfill the required quantities for this contract.

On May 30, 2013, due to low initial seed availability, CBI provided RNP with more *C. intermedia* seed for this project. The seed was sown and managed as discussed above for earlier lots, and harvest quantities are included in the total (Table 4).

Seed production was on target for the native grasses (*S. pulchra*, *S. lepida*), *P. erecta*, and *S. columbariae*. With additional wild-collection of seeds, the amount of seed needed for *C. filaginifolia* also met target goals. Seed production of *A. adscensionis* and *C. intermedia* fell short of target goals. For both species, the small amount of seed available for bulking likely contributed to final results. *Cryptantha intermedia* exhibited relatively high germination rates, and vigorous growth and reproduction. Conversely, *A. adscensionis* had a relatively low germination rate and growth problems that contributed to the low seed bulking results.

CBI requested testing of bulked seed for germination and viability. Due to low inventory quantities and the relatively large amounts needed for testing, germination results were provided for only 5 species for which seed was bulked or purchased for this project (Table 5). Note that no pre-treatments were conducted to enhance germination, nor were any post-germination tests run to assess viability. For some species, dormancy mechanisms may exist that preclude germination unless dormancy is relieved; thus, lack of germination does not necessarily equate to low viability.

Seed Purchase

To augment field-collected seed, we purchased additional seed for restoration from both RNP and S & S Seeds, Inc. Refer to Table 6 for species, vendor, amounts purchased, and source.

4.2.4 Seeding

Seeding was accomplished using a modified version of the 'DiSimone' strip seeding method, which consisted of seeding in long rows or strips that extended along slope contours.

Table 4

Bulk Seed Production

Species	Initial Seed Quantity (lbs)	Seed Goal (lbs)	# of Plants Grown	Growing Method	Seed Produced (lbs)
Stipa pulchra	0.61	17.58[1]	300	Field grown	17.55[1]
Stipa lepida	0.03	See above	144	Field grown	See above
Aristida adscensionis	0.12	3.12	66	1 gallon containers	0.08
Corethrogyne filaginifolia	0.44	2.93	429	Field grown/wild-collected	2.90
Cryptantha intermedia	0.05	5.16	6 flats @ ca. 200/flat	Flats	1.48
Plantago erecta	4.00	10.00	30,000 ft^2	Field grown	10.00
Salvia columbariae	0.06	1.65	576	1 gallon containers	1.68

[1] Includes *S. lepida*.

Table 5

Seed Purity and Germination

Species	Purity (%)[1]	Germination (%)
Corethrogyne filaginifolia	Not tested	41
Deinandra fasciculata	69	14
Plantago erecta	74	85
Salvia columbariae	Not tested	75
Stipa spp.	72	54

[1] Purity is the composition by weight of pure seed in a sample (% purity = [weight of pure seed/weight of sample] x 100). Percent (%) purity may be lowered by inclusions such as non-seed plant material.

Table 6

Purchased Seed

Species	Vendor[1]	Source	Amount Purchased (lbs)
Bahiopsis laciniata	RNP	Otay	3.12
Deinandra fasciculata	RNP	South San Diego	4.68
Plantago erecta	RNP	Otay and Marron Valley	4.00
Acmispon glaber	S&S Seeds, Inc.	San Diego	3.12
Artemisia californica	S&S Seeds, Inc.	Camp Pendleton	24.24
Bahiopsis laciniata	S&S Seeds, Inc.	Otay Mesa	5.62
Eriogonum fasciculatum	S&S Seeds, Inc.	Baja California	48.48
Eriophyllum confertiflorum	S&S Seeds, Inc.	Baja California	2.50
Isocoma menziesii	S&S Seeds, Inc.	Baja California	26.50
Lasthenia californica	S&S Seeds, Inc.	Commercial	3.25
Layia platyglossa	S&S Seeds, Inc.	Commercial	3.25
Lupinus bicolor	S&S Seeds, Inc.	Commercial	13.00
Salvia apiana	S&S Seeds, Inc.	Ramona	5.62
Sisyrinchium bellum	S&S Seeds, Inc.	Commercial	13.00

[1] RNP = RECON Native Plant Nursery.

Establishment of native species in strips serves as a seed source for unplanted, intervening habitat; thus, combining active and passive restoration and reducing seed costs (DiSimone no date). The advantages of this method include cost efficiencies by (1) concentrating seed in a smaller area to maximize germination success and bolster the seed bank and (2) focusing nonnative species control in intervening areas where native species are not as dense initially. Long-term monitoring on Audubon Starr Ranch in Orange County, CA indicates that although this process can be relatively slow, native cover does increase outward from seeded strips.

Elements of the strip seeding method used in this project included:

- Establishment of 1-meter (m) wide strips along slope contours; each strip was separated by a 5-m wide buffer. Strip boundaries were marked with pin flags.
- Ripping the soil to a depth of 3-6 inches, raking out ripped soil, and breaking large soil clumps within the 1-m strips.

- Seeding of strips in November 2013, prior to the onset of winter rains. Seed was measured out and then strips were hand-seeded and raked to distribute seed evenly.
- Post-seeding tamping of soil, using a hand tamper, to maximize seed-soil contact.

Strip installation and seeding was conducted by RECON in November 2013, under the direction of CBI biologist Jessie Vinje. On Crestridge, 16 strips were installed in polygon 1; on South Crest, 22 strips were installed in polygon 2 and 20 strips were installed in polygon 3. Strip length varied depending on polygon shape, but generally ran the width of the polygon. Approximately 56 pounds of native seed mix were hand-broadcast evenly into strips on CER on November 15, and 199 pounds of native seed mixes were hand-broadcast into strips on South Crest on November 20. Photodocumentation of the seeding process is presented in Figure 14. A rain event occurred within a week of seeding; photodocumentation of initial germination in strips is presented in Figure 15.

Seeding success was variable and survivorship was adversely impacted by low rainfall. Early germinating species in the strips included *P. erecta*, *C. intermedia*, and *Lupinus bicolor*. By January, *Plantago* and *Lupinus* seedlings were showing signs of stress. On Crestridge, there was relatively good germination of *S. apiana*, *S. columbariae*, *P. erecta*, *C. intermedia*, and *Deinandra fasciculata*. On South Crest, there was relatively good germination of *L. bicolor*, *P. erecta*, *C. intermedia*, *D. fasciculata*, and *Layia platyglossa*, with fewer *C. filaginifolia* and *Grindelia camporum* seedlings. *Salvia mellifera* seedlings were observed only in the east end of polygon 3.

4.3 Site Protection and Education

Fencing and signage were included as project components for both protective and educational purposes. The South Crest property, in particular, has been subjected repeatedly to unauthorized off-road vehicle traffic in or near *Brachypodium* restoration sites. In addition, the surrounding community uses the South Crest site for hiking, mountain biking, and dog-walking.

4.3.1 Fencing

Fencing was installed by Alpine Fence, Inc. on South Crest in January 2014, subsequent to seeding of restoration sites (Figure 16). The primary purpose of this fencing was to protect sites from unauthorized vehicle use. Installation included 2,200 feet of 42-inch high, 2-strand barbless wire fencing with 6-foot metal T-posts. This design allows for wildlife movement while inhibiting vehicular traffic. Galvanized steel posts were installed at fence termini using mechanized equipment and all T-posts were installed with a post pounder. Fencing was installed in two locations on Skeleton Flats (polygons 2 and 3), and did not include any gates. The fencing subcontractor worked with CBI and EHC regarding fence placement and avoidance of sensitive biological areas (including rare plants) during installation and staging. The fencing

Figure 14

Strip Seeding Process

A.

B.

C.

D.

E.

F.

A. Installation of seed strips: soil scarification, B. Seeding, C. Seeding and raking, D. Tamping seed, E. Tamping (close-up), F. Seeded and tamped strip.

Figure 15

Post-Seeding Germination

A. Native forbs and nonnative grasses emerging: *Lupinus bicolor*, *Cryptantha intermedia*, *Chlorogalum parviflorum*, and *Brachypodium distachyon*, B. Non-seeded area, C. Native forbs: *Lupinus bicolor, Cryptantha intermedia*, D. Native forbs: *Plantago erecta*.

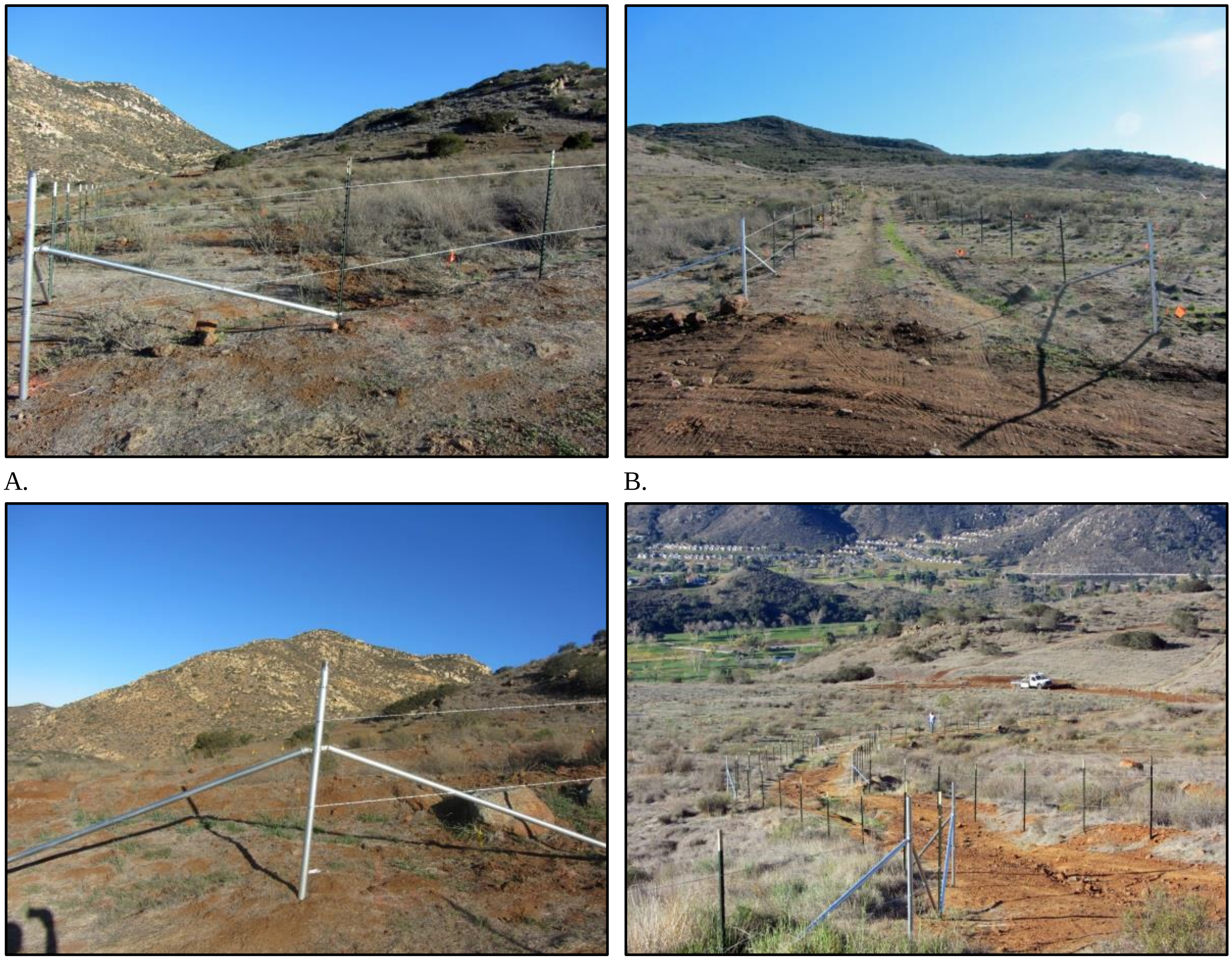
subcontractor will remove fencing one year after installation, unless alternative arrangements are made with the land owner, EHC. Due to steep terrain and general inaccessibility to the public, no fencing was installed on CER.

4.3.2 Signage

Signage was installed on South Crest in 2013 and 2014. Signage consisted of interpretive signs designed to educate the community on the biological importance of the site and the restoration process and informational signs designed to direct traffic around sensitive areas. Figure 17 depicts an interpretive sign that was created by CBI and EDI, and installed by EDI and

Figure 16

Fencing on the South Crest Property



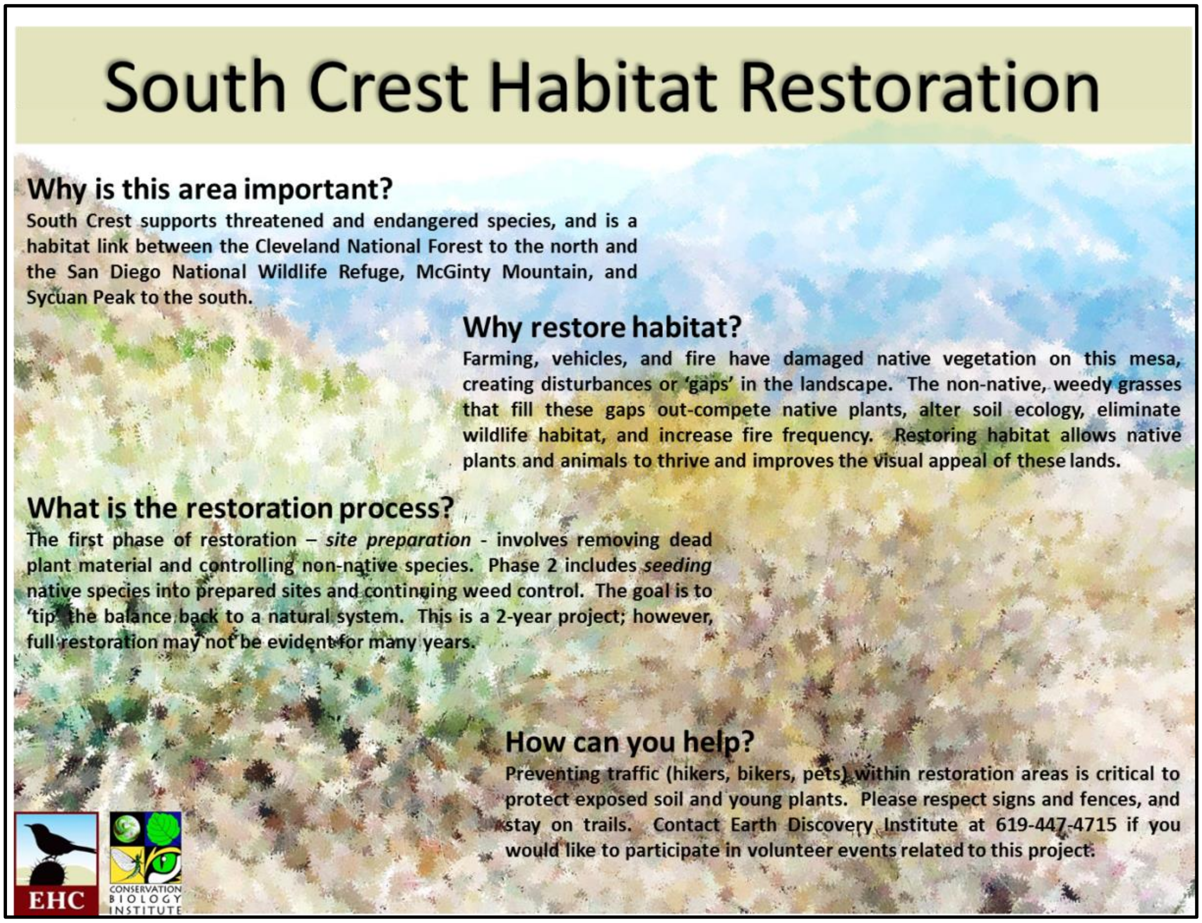
A. B. C. D.

A. 2-strand barbed wire fencing with galvanized steel posts, B. Fencing along boundary of polygon 002 (left) and 003 (right), C. Galvanized steel post at corner of fencing, D. Overview of fencing (view to southwest).

volunteers at the north and south ends of Skeleton Flats on South Crest in December 2012. Figure 18 presents information signs installed by EHC, EDI, and volunteers in May 2014. A total of 49 signs were installed at restoration sites:

Figure 17

Interpretive Signage on South Crest



South Crest Habitat Restoration

Why is this area important?

South Crest supports threatened and endangered species, and is a habitat link between the Cleveland National Forest to the north and the San Diego National Wildlife Refuge, McGinty Mountain, and Sycuan Peak to the south.

Why restore habitat?

Farming, vehicles, and fire have damaged native vegetation on this mesa, creating disturbances or 'gaps' in the landscape. The non-native, weedy grasses that fill these gaps out-compete native plants, alter soil ecology, eliminate wildlife habitat, and increase fire frequency. Restoring habitat allows native plants and animals to thrive and improves the visual appeal of these lands.

What is the restoration process?

The first phase of restoration – *site preparation* - involves removing dead plant material and controlling non-native species. Phase 2 includes *seeding* native species into prepared sites and continuing weed control. The goal is to 'tip' the balance back to a natural system. This is a 2-year project; however, full restoration may not be evident for many years.

How can you help?

Preventing traffic (hikers, bikers, pets) within restoration areas is critical to protect exposed soil and young plants. Please respect signs and fences, and stay on trails. Contact Earth Discovery Institute at 619-447-4715 if you would like to participate in volunteer events related to this project.

EHC

CONSERVATION BIOLOGY INSTITUTE

Figure 18

Informational Signage on South Crest

A. B. C. D.

A-D. Informational signs installed on South Crest to protect restoration areas.

- Interpretive signs – 2 signs along main access road at north and south ends of Skeleton Flats
- Habitat Restoration In Progress – 19 signs on fencing
- Closed Area, No Trespassing – 21 signs on fencing
- Off-road Activity Prohibited (with San Diego County vehicle code reference) – 2 signs on T-posts along main access road at north and south ends of Skeleton Flats
- Ecological Reserve, Dogs Must Be on Leash – 2 signs on T-posts along main access road at north and south ends of Skeleton Flats
- Trail Closed – 3 signs on T-posts at significant trails

5. Experimental Design and Monitoring

The project included an experimental component to test the relative effectiveness of different *Brachypodium* treatment and restoration methods. This section describes goals and objectives, research questions, experimental design, quantitative monitoring, data analysis, and results.

Two restoration strategies were used in developing site- and polygon-specific treatment and restoration plans: invasive species control and native species augmentation. The objective of invasive species control was to reduce or eliminate nonnative, invasive plants to create conditions under which native species could germinate, establish, and persist. The objective of native species augmentation was to establish desirable (native) species that are functionally similar to invaders, thereby increasing both (1) habitat resistance to future invasions and (2) *potentially* suitable habitat for covered species, including *Acanthomintha ilicifolia* on CER and *Nolina interrata* and *Dudleya variegata* on South Crest. Specific actions to achieve these objectives included:

- Dethatching, mowing, and/or herbicide applications to decrease the growth, propagule production, and frequency of dispersal of target invasive species.
- Introducing site- and habitat appropriate native seeds into selected treatment polygons to increase native plant propagule production and dispersal.

Treatment and restoration plans for both sites were designed to assess the following questions:

- Are there significant differences in species cover and richness with different treatment ‘combinations’? (e.g., Fusilade + glyphosate versus mechanical + glyphosate).
- Does dethatching improve treatment effectiveness or enhance native species richness?
- Are there significant differences in native species cover/richness between seeded and non-seeded (natural recruitment) plots?

5.1 Experimental Design

Dr. Douglas Deutschman at the Institute for Ecological Monitoring and Management (IEMM) at San Diego State University provided assistance with the experimental design. The experiment used elements of both blocked and split-plot designs (Figure 19) at CER and South Crest. These types of designs are common in agriculture and ecology/conservation because they allow managers to measure the impact of the treatment despite significant spatial heterogeneity. In addition, the design used a pre- and post- treatment survey (related to BACI designs: Before, After, Control, Intervention).

The design included polygons, blocks, and paired plots to test the effectiveness of management actions while minimizing the amount of untreated (control) habitat. Treatment polygons corresponded to habitat assessment polygons, and included CER polygons 1, 3, and 5 and South Crest polygons 2, 3, 4, 5, and 8.[3] Each treatment polygon was divided into three roughly equal-sized segments or blocks, which served as treatment replicates. Each block contained a set of paired plots. Paired plots were adjacent to each other to minimize variability due to habitat or topography, and sited by randomly locating the first plot, then placing the second plot approximately 3 meters (m) away.

Polygon and block sizes were variable; plot dimensions were 5 m^2. Within paired plots, the control (no treatment) and treatment were assigned randomly. Control plots were staked with 1 m lengths of rebar and pvc pipe at all four corners, while treatment plots were staked with rebar and pvc only at the northwest corner. Prior to treatment, all four corners of control plots were flagged to facilitate identification and alert subcontractors to avoid treatment within these plots. Although the entire plot was treated, quantitative monitoring occurred only in the innermost 4 m^2 to accommodate a 0.5 m outer buffer that received the heaviest foot traffic.

Five types of treatments were implemented within the project area in various combinations: dethatching, herbicide, mechanical (mowing), seeding, and a control (Table 7). As discussed in previous sections, dethatching was conducted in Fall 2012, herbicide treatments were initiated in February 2013 and continued through Spring 2014, mechanical treatments were conducted in Spring 2013 and 2014, and seeding occurred in Fall 2013.

5.2 Quantitative Monitoring

In 2013 and 2014, cover and species richness data were collected using a 0.5 x 1 m quadrat in each plot. Pre-treatment data were collected in January 2013; post-treatment data were collected in May 2013 and 2014. Quadrat placement in plots was random initially, and stationary thereafter. Cover measurements were taken at 36 points within the quadrat at the intersection of

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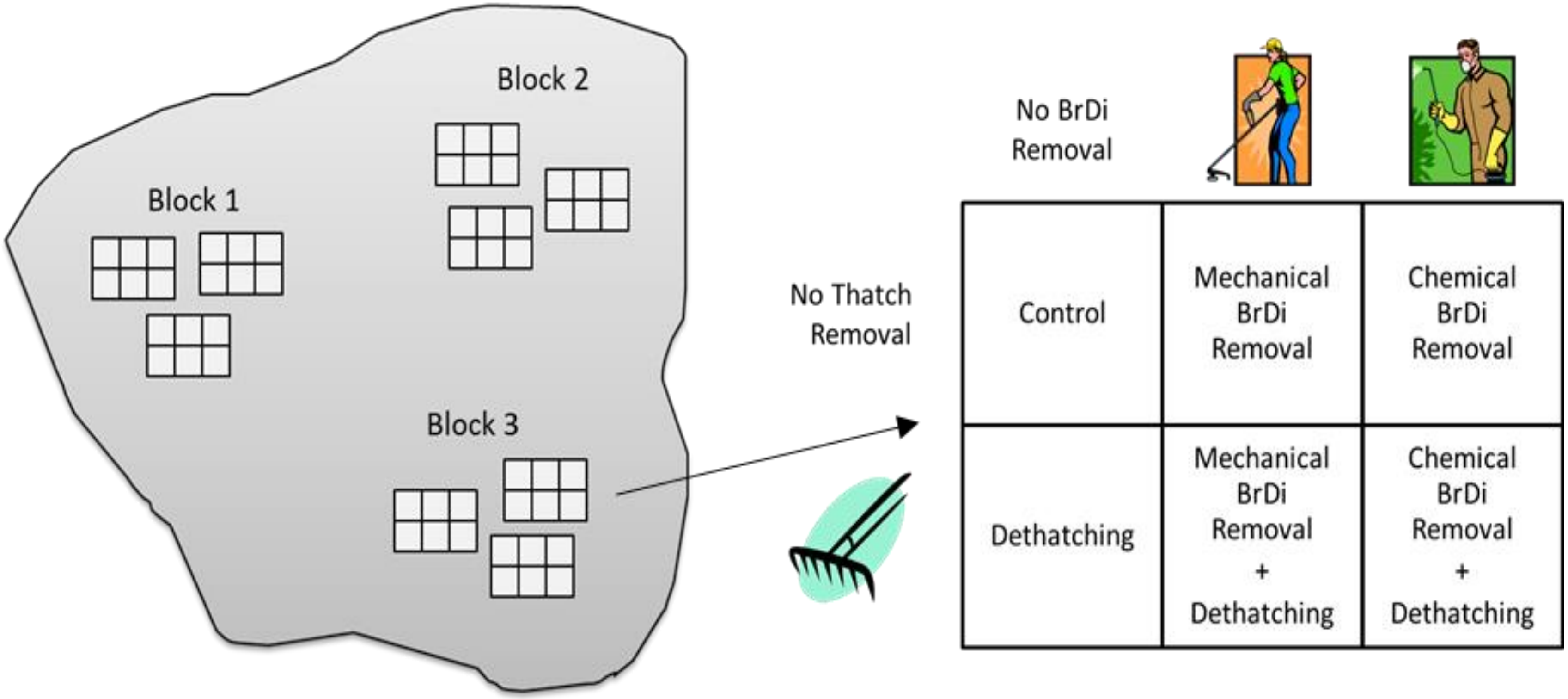
[3] South Crest polygon 1 was treated but not included in the experimental design.

Figure 19

Schematic of Experimental Design



	No BrDi Removal		
No Thatch Removal	Control	Mechanical BrDi Removal	Chemical BrDi Removal
	Dethatching	Mechanical BrDi Removal + Dethatching	Chemical BrDi Removal + Dethatching

Source: Dr. Doug Deutschman

Table 7

Brachypodium Treatment Combinations

Treatment Combination	No. of Polygons	No of Replicates[1]
Dethatch-Fusilade -Glyphosate-Seeding	2	6
Dethatch-Mechanical-Glyphosate-Seeding	1	3
Fusilade (1x)-Glyphosate	4	12
Fusilade (2x)-Glyphosate	1	3
Control	9	27

[1] Each polygon had 3 paired experimental plots; each paired plot represented a replicate for the purpose of statistical analysis.

a wire grid. Species richness data were collected within the entire quadrat. Refer to Appendix G for sampling data.

5.3 Quantitative Data Analysis

Quantitative data analyses and interpretation of results in this section were provided by Dr. Douglas Deutschmann, San Diego State University. Refer to Attachment G for Dr. Deutschman's full report. The statistical analysis of pre-post and split-plot designs can be complex because the model must include terms for the spatial structure as well as the paired values (pre and post) measured from the same plot. A repeated-measures ANOVA was used for all initial analyses. In many cases, analyses could be simplified to more common ANOVA and paired t-tests. When possible, the simpler analysis is presented to make interpretation easier.

Major Results: *Brachypodium* Control

In general, all treatments were effective at reducing the cover of *Brachypodium* (Table 8). In most cases, *Brachypodium* cover was reduced to zero or nearly zero for all treated plots (Figure 20). There was some evidence of polygon to polygon variability but no consistent difference between CER and South Crest. The treatment effect was the dominant statistical signal in both years.

In 2013, several plots at South Crest were not treated completely by the contractor (i.e., less than uniform herbicide application) leading to some residual *Brachypodium* (Gordon-Reedy, pers. comm.). In 2014, modest amounts of *Brachypodium* cover reflected new growth after an unseasonably late spring rain (Gordon-Reedy, pers. comm.). Refer to Figure 20 for *Brachypodium* cover in 2013 and 2014. Each polygon is a complete block of the experiment (three at CER and five at South Crest).

Table 8

General Linear Model (GLM) of *Brachypodium* Cover in 2013 and 2014[1]

2013	**SSQ**	**df**	**MSQ**	**F-ratio**	**P-value**
Between Blocks					
Site	3.90	1	3.90	0.11	0.742
Polygons within Sites	643.9	6	107.3	3.09	0.033
Error	555.7	16	34.7		
Within Blocks					
Treatment	7847.4	1	7847.4	177.9	<.001
Treatment * Site	3.07	1	3.07	0.07	0.795
Treatment * Polygons	413.2	6	68.9	1.56	0.222
Error	705.7	16	44.1		

2014	**SSQ**	**df**	**MSQ**	**F-ratio**	**P-value**
Between Blocks					
Site	185.0	1	185.0	1.41	0.252
Polygons within Sites	2657.4	6	442.9	3.38	0.024
Error	2095.0	16	130.9		
Within Blocks					
Treatment	23655.7	1	23655.7	244.0	<.001
Treatment * Site	1261.4	1	1261.4	13.0	0.002
Treatment * Polygons	1107.6	6	184.6	1.90	0.142
Error	1551.0	16	96.9		

[1] Note that the treatment effect is much larger than any differences among polygons or between years.

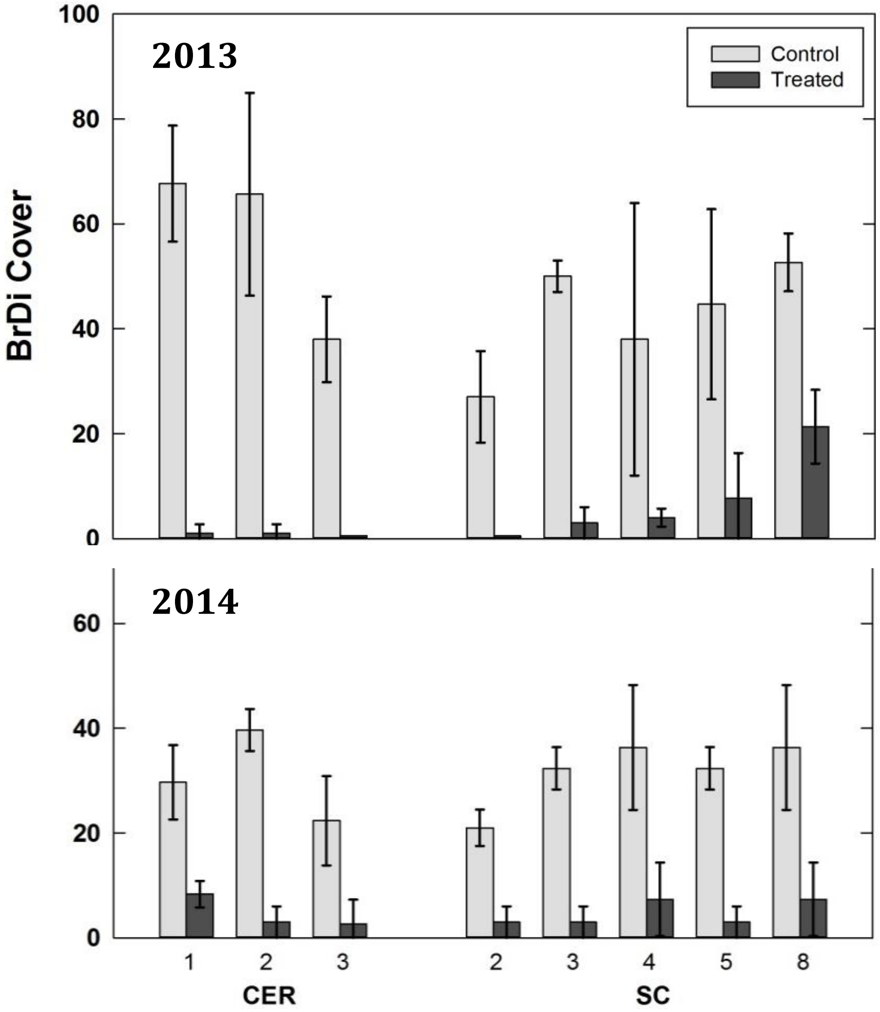
There was little difference among the different control methods used, as shown for 2014 data in Figure 21. Although there was some evidence that Fusilade + glyphosate was more effective than mechanical removal + glyphosate at CER, the addition of Fusilade at South Crest did not appear to improve control (note: the 2014 glyphosate + seed treatments at South Crest occurred in plots that had been dethatched and treated twice with Fusilade in 2013). The differences observed among the treatments were small compared to the difference between all the treated plots compared to the untreated controls.

Functional Groups and Richness Data

Cover of exotic grass was significantly higher on untreated plots in 2013 compared to 2014 (Figure 22). Inter-annual variation in grass is highly variable and often driven by the amount and

Figure 20

Brachypodium Cover in Experimental Plots in 2013 and 2014[1]



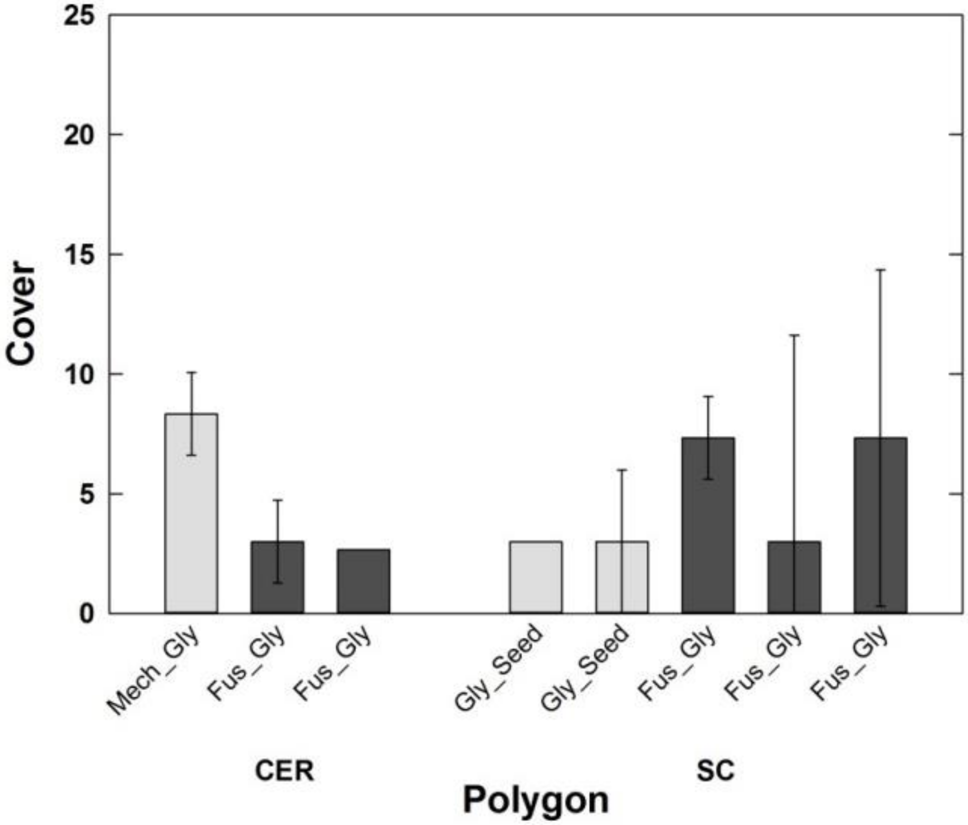
timing of rainfall. It is important to note that control of *Brachypodium* was achieved in both years.

Cover of native forbs and grasses was low and variable (Figure 23, left). Average cover of native plants was never greater than 10%. A similar pattern was observed on treated plots (Figure 23, right). There is no evidence that treatment altered native cover. It is important to remember that native cover was low and patchy.

There is some evidence that total species richness is higher in treated plots relative to controls (Figure 24). There is also some evidence that South Crest has higher species richness than CER. Species richness is low and these effects are fairly small. Detecting meaningful change in species richness probably requires scaling the experiment up to larger plots. Refer to Section 5.5 for additional, qualitative observations regarding species richness.

Figure 21

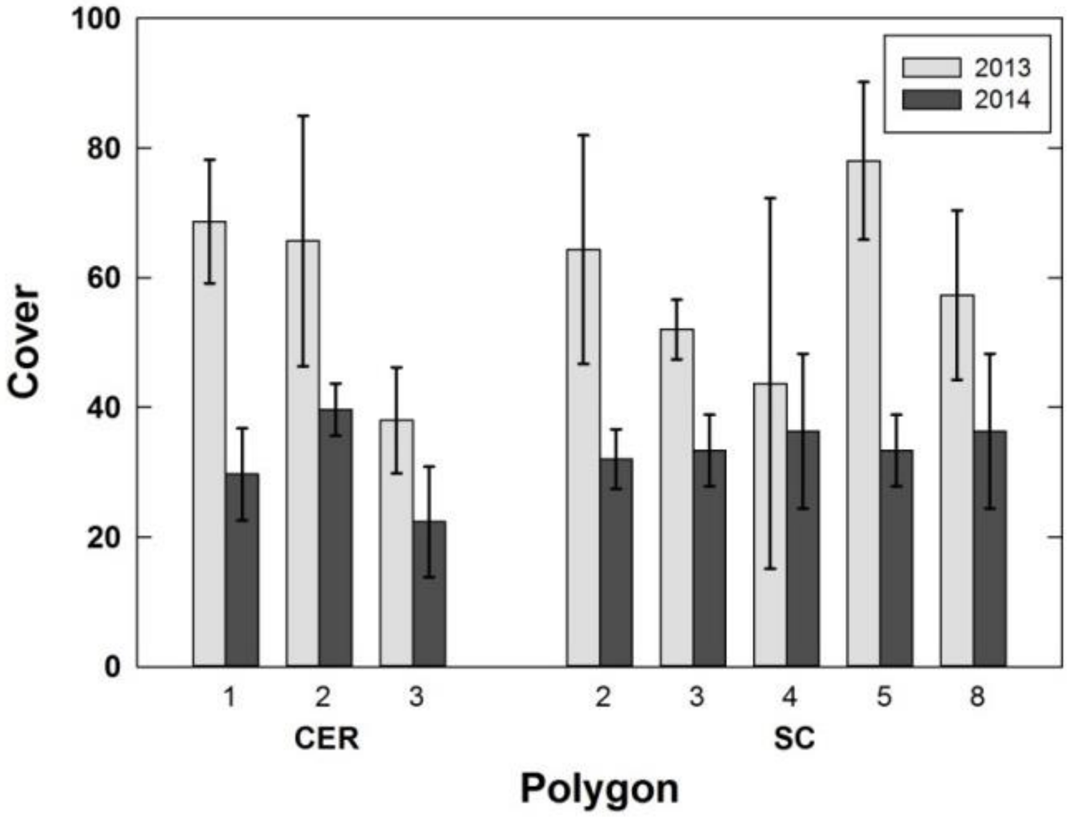
Brachypodium Cover as a Function of Treatment



	SSQ	df	MSQ	F-Ratio	P-Value
CER					
Treatment	60.5	1	60.5	5.76	0.047
Error	73.5	7	10.5		
SC					
Treatment	30.0	1	30.0	1.35	0.266
Error	288.9	13	22.2		

Figure 22

Exotic Grass Cover in Control Plots in 2013 and 2014[1]



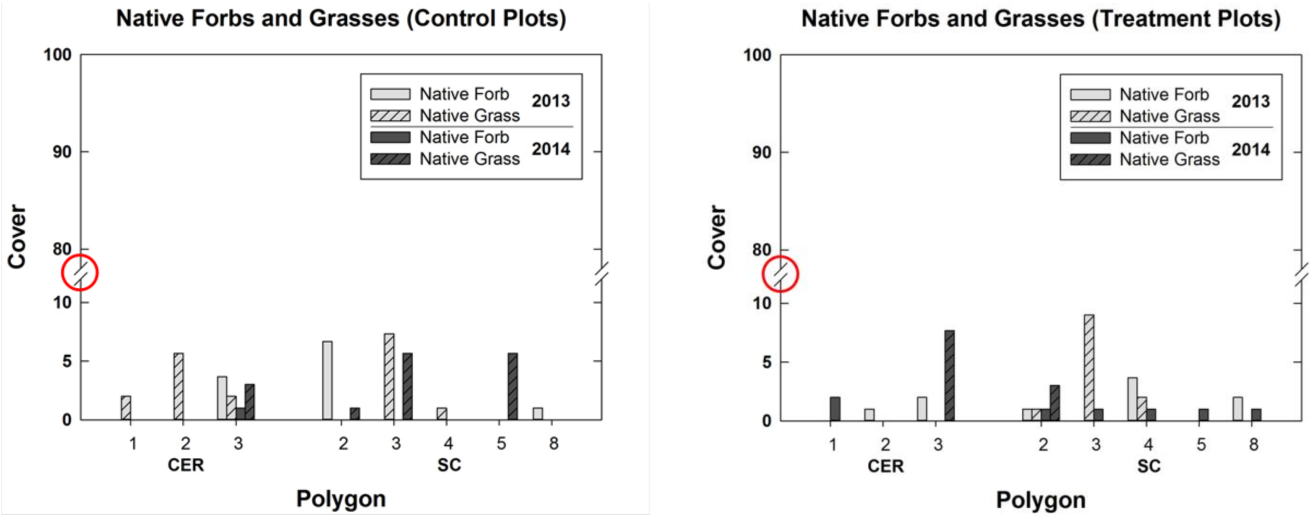
Source	SS	df	MS	F-Ratio	P-Value
Between Subjects					
Site	80.0	1	80.0	0.371	0.549
Error	4,746	22	215.7		
Within Subjects					
Year	7,514.3	1	7,514.3	36.73	0.000
Year * Site	12.27	1	12.27	0.060	0.809
Error	4,501	22	204.58		

---

[1] CER polygons 2 and 3 shown in Figure 22 = CER treatment polygons 3 and 5, respectively, as discussed in text.

Figure 23

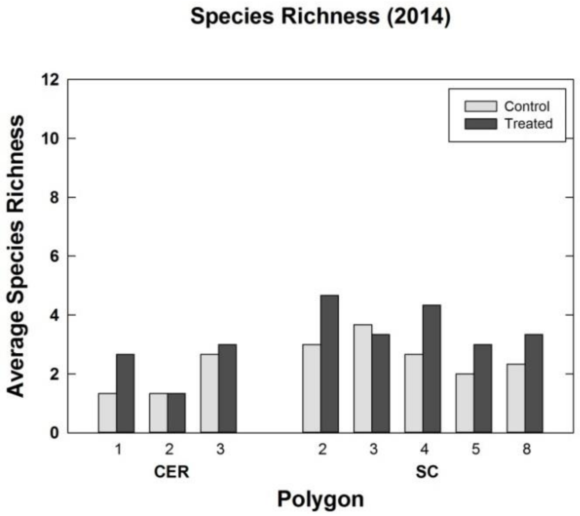
Native Grass and Forb Cover from Control and Treatment Plots in 2013-2014[1]



[1] CER polygons 2 and 3 shown in Figure 23 = CER treatment polygons 3 and 5, respectively, as discussed in text.

Figure 24

Total Species Richness[1,2]



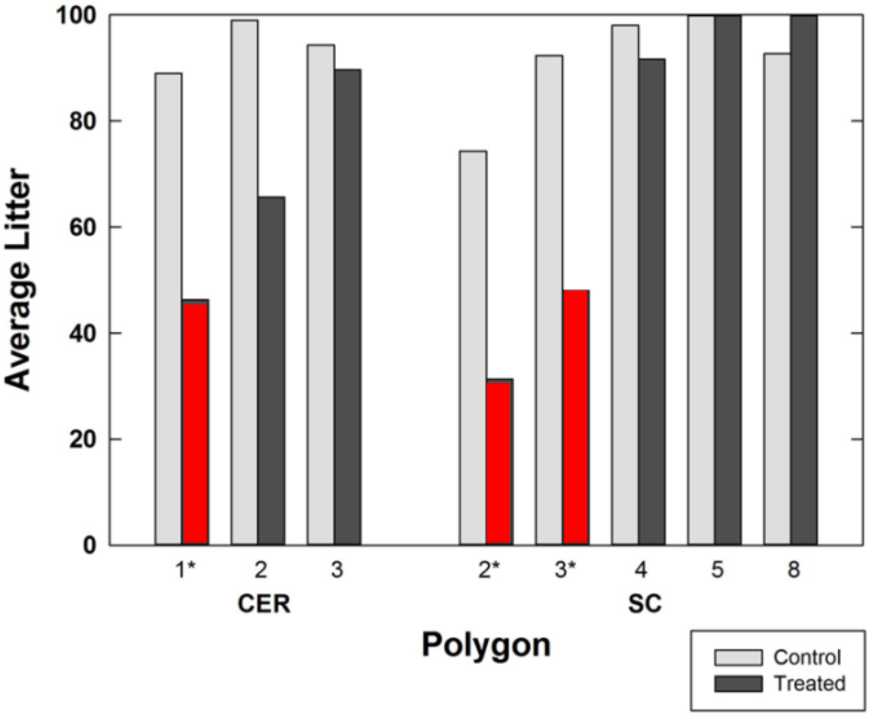
Source	SS	df	MS	F-Ratio	P-Value
Between Subjects					
Site	5.20	1	5.20	6.699	0.041
Error	4.66	6	0.77		
Within Subjects					
Treatment	2.27	1	2.27	7.500	0.034
Trt * Site	0.19	1	0.19	0.612	0.464
Error	1.82	6	0.30		

[1] CER polygons 2 and 3 shown in Figure 24 = CER treatment polygons 3 and 5, respectively, as discussed in text.
[2] Values are averages of blocks within each polygon.

There is strong evidence that the dethatching treatment reduces litter (Figure 25, red bars). Control of *Brachypodium* without dethatching did not reduce litter on this time scale.

Figure 25

Litter in Control and Treated Plots[1,2]



Source	SS	df	MS	F-Ratio	P-Value
Between Subjects					
Site	64.44	1	64.44	0.50	0.511
Dethatch	3320.1	1	3320.1	25.81	0.004
Error	643.1	5	128.6		
Within Subjects					
Treatment	180.4	1	180.4	2.64	0.165
Trt * Site	136.0	1	136.0	1.99	0.217
Trt * Dethatch	1259.7	1	1259.7	18.46	0.008
Error	341.3	5	68.2		

---

[1] CER polygons 2 and 3 shown in Figure 25 = CER treatment polygons 3 and 5, respectively, as discussed in text.

[2] Values are averages of blocks within each polygon. The three dethatched plots (red bars) have substantially lower litter than all others.

5.4 Results

Results from this adaptive management experiment are encouraging. Control of *Brachypodium* can be achieved with one of several chemical (herbicide) regimes. Further, dethatching reduces litter substantially. Despite these successes, the long-term success of the experiment is uncertain. The control of *Brachypodium* did not lead to substantial increases in the cover of native species. It is possible that controlling *Brachypodium* increased species richness, but the signal was small due to the scale of the plots.

- *Brachypodium* was reduced to low levels across the plots and in both years. As a result, measuring pre-treatment (before) cover values does not improve the analysis. Thus, the pre-treatment cover estimates can be eliminated without losing information or power.
- The cover estimates were very precise, but estimates of species richness were low and idiosyncratic. Species richness and composition should be estimated from larger belts or areas. This will provide more precise information about changes in community composition.
- There is significant inter-annual variability in the cover of *Brachypodium* and other species. Understanding the success of any control program requires measurement over a fairly long time period (perhaps 5 to 7 years?) in order to separate trend from inter-annual fluctuations.
- This experiment provides an important baseline of data and adding further years of treatment and/or monitoring will only increase their value.
- The utility of these methods for management depend on how they can be scaled up. If the experiment is continued, larger-scale plots should be pilot tested.

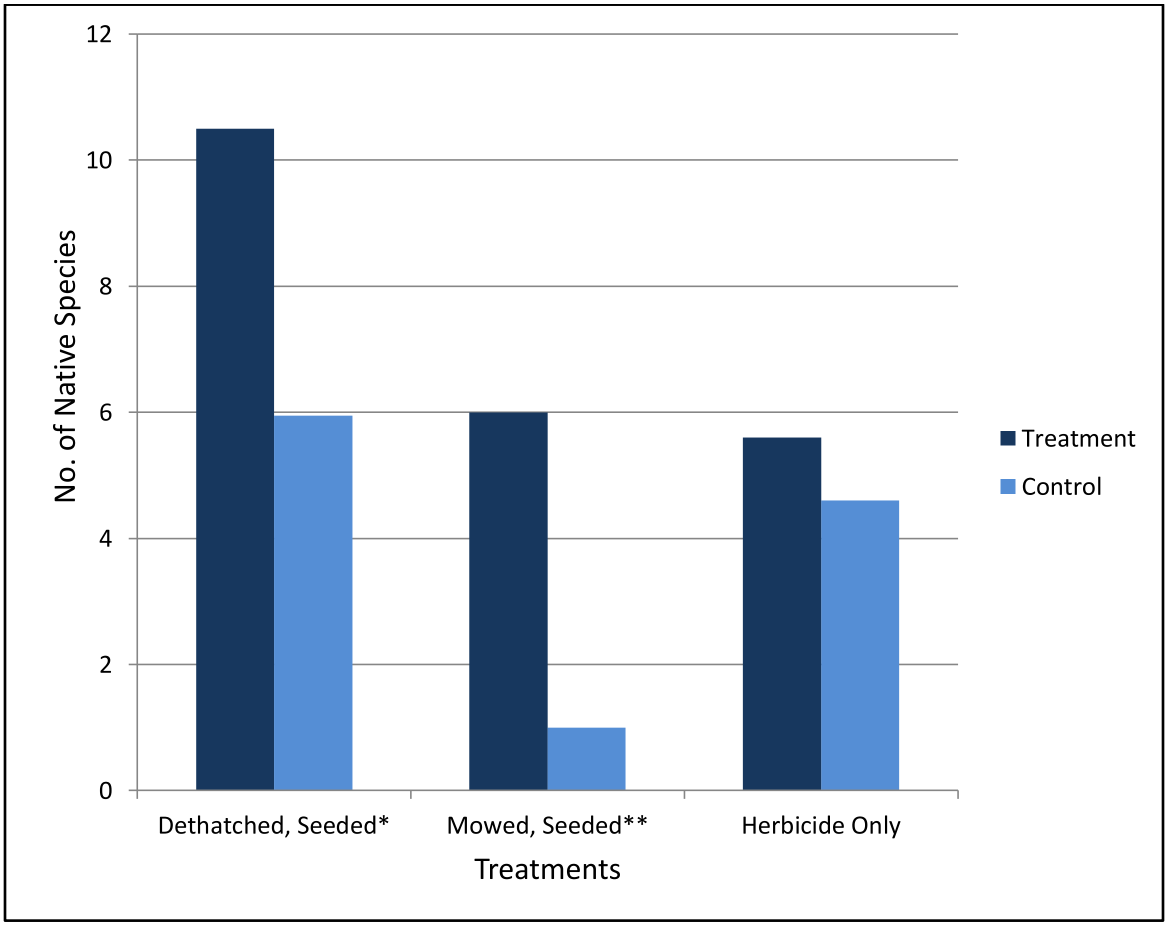
5.5 Qualitative Observations

Although quantitative data did not detect a significant increase in native species richness with seeding or other treatments, qualitative observations suggested that number of native species present was higher in treatment versus control plots. It may be that native species do not yet occur in sufficient numbers to be detected through quantitative sampling (or plot size used in this study). Figure 26 depicts the mean number of native species in treatment versus control plots for different treatments. Note that the dethatched-seeded treatment was treated with Fusilade twice in 2013, and seeding was conducted in Fall 2013.

The dethatched-seeded combination consistently had the highest number of native species present, and we suspect this was due to increased seed-soil contact. The other seeding treatment (mowed-seeded) was almost identical to the herbicide-only treatment with respect to number of native species present. Thatch was left in place in the mowed-seeded treatment, and may have limited seed-soil contact.

Figure 26

Native Species in Treatment versus Control Plots in 2014



* Treatment included herbicide application (Fusilade in 2013 and glyphosate in 2013 and 2014); treatment and control plots were dethatched, but only treatment plots were seeded.
**Treatment included herbicide application (glyphosate-based herbicide only).

The relatively low cover of native species may have been related, at least in part, to drought conditions. We observed good initial germination following seeding and a rainfall event, but the majority of plants did not persist to flowering or fruiting, presumably due to lack of water following germination. In addition, it appeared that germination was limited compared to the amount of seed introduced into the soil seed bank. The bulk of the introduced seed may still be present in the seed bank and available for release (germination) with adequate rainfall conditions, particularly if *Brachypodium* cover (including thatch) is maintained at low levels.

<u>Recommendations</u>:

- Native seed germinated in seeded plots but had relatively low survival due to below-average rainfall. Future seeding should incorporate watering events as a contingency measure, where feasible.
- Continue monitoring seeded plots to assess success beyond one year; this will be particularly important in an adequate rainfall year.
- Continue treating seeded plots, as necessary, to maintain the low cover of nonnative species achieved in this study and provide suitable conditions for germination of native species.

6. Cost Analysis

While the primary objective of this project was to determine effective treatment strategies for eradicating or controlling *Brachypodium* on conserved lands, a secondary objective was to provide land managers with a summary of treatment costs to assist in decision-making. In some cases, higher costs/unit may result in lower overall costs if a crew is more efficient or a method is more effective and requires fewer treatments. We expect some economy of scale with larger treatment areas. For example, there is often a minimum fee per day to field a restoration crew. Labor, travel, and equipment costs are higher in small treatment areas, particularly where crews finish applications in less than a full day. Use of mechanized equipment (dethatching, mowing, herbicide application) on large sites may also result in lower treatment costs/acre. An analysis of treatment costs is provided for the following project elements:

- Dethatching
- Mowing
- Herbicide
- Seeding

Table 9 summarizes costs and treatment effectiveness; refer to the sections below for additional analyses. In compiling costs for Table 9, it became apparent that many of the costs are not directly comparable due to changes in personnel, method, labor rates, and site conditions. Nonetheless, these costs may provide a relative ‘scale of effort’ for project planning.

Table 9

Brachypodium Treatment Costs and Effectiveness

Treatment (year)	Crestridge		South Crest	
	Cost/Acre[1]	Control[2]	Cost/Acre[1]	Control[2]
Dethatching[3] (2013)	$1,600	NA[4]	$1936-2,058	NA[4]
Fusilade (2013)	$445	93%	$306	99.5%
Fusilade (2014)	$843	97%	NA[4]	NA[4]
Glyphosate[5] (2013)	$112	NA[4]	$255	NA[4]
Glyphosate[5] (2014)	$178	NA[4]	$511	NA[4]
Mowing (2013)	$350[6]	99%	---	NA[4]
Mowing (2014)	$1,150	92%[7]	---	NA[4]

[1] Approximate costs/acre = treatment costs. Costs were averaged where >1 treatment occurred per year. Costs include labor and field-associated expenses.

[2] Control = Effectiveness of *Brachypodium* control treatment in experimental treatment plots.

[3] Dethatching occurred in combination with other treatments and is included only for costs/acre. Refer to other treatments for overall effectiveness.

[4] NA = not applicable.

[5] Glyphosate does not affect *Brachypodium* cover, but is included in the table for approximate treatment costs/acre.

[6] The 2013 mowing event followed dethatching, which greatly reduced the amount of standing biomass and dethatching effort.

[7] Lower *Brachypodium* control in 2014 versus 2013 is believed to be due to a post-mowing germination event; differences are not statistically significant.

6.1 Dethatching

Dethatching was conducted in polygons scheduled for seeding. Prior experiments demonstrated that dethatching did not significantly increase herbicide effectiveness (CBI 2012a). However, dethatching was hypothesized to be beneficial when followed by active restoration (seeding), as removal of biomass (and effects of shading) would provide bare soil for native forb germination while potentially exacting a small inhibitory effect on *Brachypodium* germination. While quantitative analyses did not detect a significant increase in native species germination or growth in dethatched plots, observational (qualitative) data did detect an increase in native species richness and growth. Examples include increased size of existing species, such *Calystegia macrostegia* and *S. pulchra*, following dethatching. We believe that dethatching is beneficial, but the effects on native species germination and growth may not be apparent immediately, particularly in years of below-average rainfall and in smaller plots where low species richness is difficult to detect.

The cost/acre for dethatching on CER was $1,600/acre, while dethatching on SC varied from $1,936-$2,058 acre. Dethatching took less time in grass-dominated habitat versus a grass-shrub matrix, and where thatch was not dense. Dethatching costs presented here include costs for field labor and expenses (equipment, travel) only, and do not include management or overhead expenses, which can vary considerably between contractors. Also, dethatching was conducted by SERG, which uses laborers presumed to be less experienced than professional field crews.

Dethatching can ‘jump-start’ passive restoration, but should be used in conjunction with other treatments (e.g., herbicide) to control nonnative grasses and forbs that may germinate following thatch removal. Dethatching is particularly important with active restoration (e.g., native species augmentation), since the bare soil surface that results from thatch removal provides a seed bed for germination.

6.2 Mechanical Treatment (Mowing)

Mowing was included as a treatment to provide land managers with options where they might not have access to herbicide or might prefer not to apply herbicide due to potential adverse effects to other resources. Previous work indicted that (1) mowing was intermediate in effectiveness between herbicide and no treatment in terms of *Brachypodium* control and (2) mowing released fewer nonnative forbs than herbicide application (CBI 2012a).

In this study, results indicate that appropriately-timed mowing can be an effective control for *Brachypodium*; we suspect it must be applied in consecutive seasons (and possibly, more than one time/season) to control the *Brachypodium* seed bank. Because mowed *Brachypodium* thatch was left to decompose in place in this study, little native or nonnative forb germination was observed. Low native species germination may have been influenced by low rainfall, as well. Forb germination may increase as thatch decomposes. Under this scenario, mowing might prove to be a cost-effective, but slower (passive) restoration process than herbicide treatment.

The cost/acre for mowing was approximately $350/acre in 2013 and $1,150/acre in 2014. Different crews were used in 2013 and 2014. The cost difference between the years is related to both the level of effort and billing rates (more experienced crews were used in 2014, at a higher billing rate than 2013 crews). The 2013 mowing occurred a few months after dethatching, which had greatly reduced the amount of standing biomass. The 2014 mowing removed both residual thatch from 2013 and 2014 growth.

Mowing was conducted only on CER, which was accessed by foot with an approximately 600-foot elevation gain. Costs are expected to be lower on more accessible sites. In both years, post-treatment *Brachypodium* cover in mowed plots averaged <10%. Post-treatment cover in 2014 was slightly higher than 2013 due to a late rainfall event that resulted in additional *Brachypodium* germination.

Mowing may be an acceptable *Brachypodium* treatment where immediate results in terms of native species richness are not required and where alternative treatments are not available or feasible. As with other treatments, timing and number of applications are keys to controlling the *Brachypodium* seed bank. We recommend more than one mechanical treatment per year, if needed (e.g., high rainfall or late rains), as well as the ability to defer treatment, if warranted by climatic conditions (e.g., drought with little germination).

6.3 Herbicide Treatment

The project assessed different herbicide combinations, as well as different methods of application. This assessment focuses on Fusilade application costs and effectiveness, but also includes costs for glyphosate treatments.

In 2013, selected polygons were treated either once or twice with Fusilade on both CER and South Crest. On CER, 2013 Fusilade applications averaged $445/acre, with virtually no difference in cost between the first and second application. Fusilade-treated polygons on CER had not been dethatched. On South Crest, 2013 Fusilade costs averaged $306/acre, with some cost differences between applications ($353/acre for the first application; $259/acre for the second application). In this case, a greater percentage of acreage in the second round had been dethatched, which facilitated application. The 2013 cost differences between CER and South Crest are due to site accessibility.

In 2014, Fusilade was applied once to 2013 Fusilade-treated polygons on CER. Treatment costs were significantly higher in 2014 ($843/acre) due to higher billing rates for the 2014 crew. Application time was slightly less in 2014 (129 hours versus 136 hours), but the application was more uniform and comprehensive than in 2013. The 2014 Fusilade application on South Crest was conducted using a different method and is discussed in the next section.

Fusilade application resulted in the greatest level of *Brachypodium* control in this study. Treatment costs varied by site and by contractor. Although results are not yet conclusive, there may be an advantage to treating sites 2x/year initially in terms of managing the *Brachypodium* seed bank, depending on *Brachypodium* density and rainfall. We recommend budgeting for more than one Fusilade application per year, if needed (e.g., high rainfall or late rains), as well as the ability to defer treatment, if warranted by climatic conditions (e.g., drought with little germination). Treatments are most effective when applied uniformly and timed appropriately, and land managers should consider contractor experience when developing a budget/treatment plan for *Brachypodium.*

Glyphosate was applied to all treatment polygons on an as-needed basis in 2013 and 2014. As indicated in Table 9, treatment costs varied by site and between years, due to site accessibility and nonnative forb diversity. As expected, the need for nonnative forb control increased as Brachypodium cover decreased and we expect this trend to continue in the short-term. We recommend budgeting for more than one glyphosate treatment per year, if needed (e.g., high rainfall or late rains), as well as the ability to defer treatment, if warranted by climatic conditions (e.g., drought with little germination).

Alternative Application Methods

For this project, herbicide was applied primarily using a backpack sprayer. In 2014, we assessed the use of a Cooperative Mule versus the backpack sprayer. Labor and equipment costs for using the Cooperative Mule are not included in Table 9 because this work was accomplished using volunteer time and grant funding. Instead, we assess level of effort (time) and treatment effectiveness to allow for comparisons with other methods.

The Cooperative Mule treated an estimated 3.5 acres of habitat in 1.5 hours, which is equivalent to a treatment time of about 26 minutes per acre. Based on previous work (Bell no date), a best-case scenario for treatment time using a backpack sprayer is 80 minutes per acre, which does not include time to stop and refill the backpack (10 tank loads per acre). Clearly, the Cooperative Mule is more efficient with respect to labor and herbicide usage than backpack sprayers. However, the cost of the mule is currently estimated at about $17,000 (Bell no date). This equipment could be cost-effective for large-scale nonnative grass control, particularly if shared between multiple land managers.

Brachypodium control was slightly more effective in areas treated with the Cooperative Mule, although the differences were not significant. Observationally, there were some areas within the Cooperative Mule treatment boundary that were missed (similar to observations regarding backpack spraying). Adjustments to the spraying regime that reduce these ‘gaps’ would likely increase the effectiveness of this method.

6.4 Seeding

This project used a strip-seeding method whereby a portion of the treatment area (rather than the entire treatment area) was seeded at a higher rate than would be possible if the entire treatment area were seeded. Exclusive of the cost of seed, which would be comparable to a strategy that seeded the entire area, costs included preparation of strips, seeding, and post-seeding tamping to ensure good seed-soil contact. Costs for strip-seeding were approximately $3,000/acre, which was higher than estimated costs per acre for seeding the entire area (average estimated cost = $1,885/acre). The benefits of this method are not entirely clear at this point. The method allows for continued weed control in non-seeded areas with minimal risk of damage to native species, and presumably an enhanced seed bank in a concentrated area. Where native species establish in strips, they are expected to act as a seed source for dispersal/colonization into the surrounding area. Quantitative monitoring results did not detect a significant increase in native species richness or cover in seeded plots in the first year (2014). Observationally, we did see a higher number of native species in seeded plots versus control plots (Figure 26). Plots were seeded in

Fall 2013 and monitoring conducted in Spring 2014. It is important to note that the 2013-2014 winter/spring rainfall totals were well below normal, which may account for the relatively low cover of native plants in seeded areas.

7. Recommendations

7.1 General Recommendations

Based on project results, we provide general recommendations to identify and assess the threat that *Brachypodium* poses to target resources (covered species, sensitive habitats) and ecosystem processes and to implement appropriate control measures to protect those resources/processes on conserved lands in San Diego County. Refer to Appendix H for a summary of recommended *Brachypodium* control BMPs, as well as alternative control methods that should be tested for this species.

1. Survey sites for the presence of *Brachypodium* and threats to covered species. *Brachypodium* has been identified on numerous soil types within San Diego County. Available data suggest that *Brachypodium* forms the densest stands on clay and gabbro-derived soils and adversely impacts covered species and native grasslands on these soils. Dense stands are also found on some metavolcanic soils, although the threat to covered species and sensitive habitats on these soils has not yet been established.

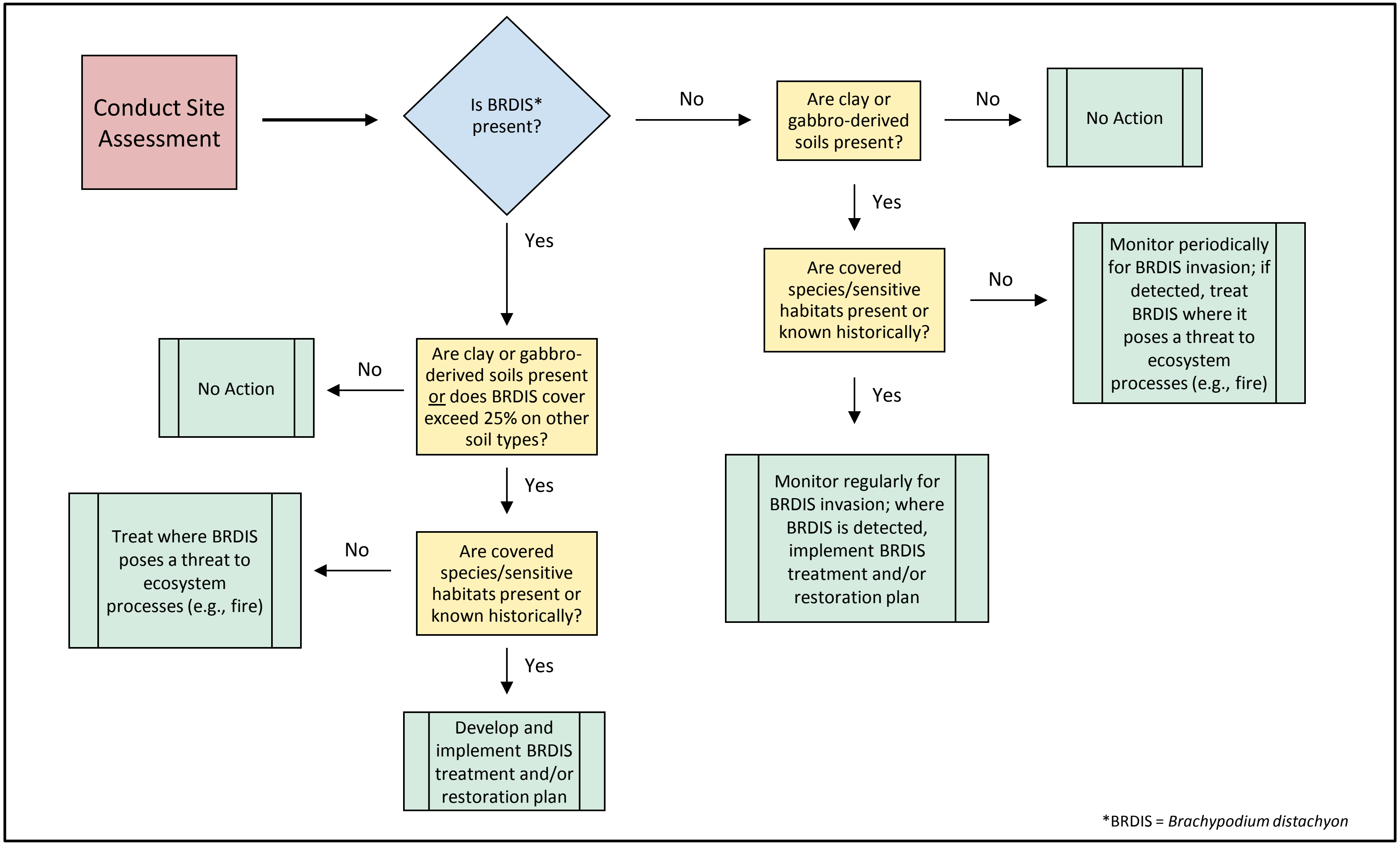
   Where *Brachypodium* is not detected on these soil types, additional tools (e.g., predictive modeling) should be consulted to determine the potential for occurrence. Until this species' distribution is better understood, land managers should survey annually for *Brachypodium* so that it can be detected and treated in the early stages of invasion. *Brachypodium* surveys may be conducted in conjunction with other surveys or monitoring efforts, e.g., for covered plant species that occur on clay and gabbro-derived soils. Figure 27 presents a proposed decision-tree for implementing *Brachypodium* control, which should be refined as more information on distribution and soil and species correlates become available.

2. Collect Baseline Data. Where *Brachypodium* is detected, determine threats to target resources and collect baseline data for restoration efforts including vegetation composition and cover, thatch cover and depth, percent bare ground, and presence or potential for target resources. Appendix B provides suggested *Brachypodium* habitat assessment forms. Review literature and spatial data for additional information on target resource occurrence (e.g., Master Occurrence Matrix [MOM], CNDDB or BIOS records).

3. Prioritize Treatment Areas. Prioritize *Brachypodium* treatment areas within a given site based on (a) threat to target resources, (b) topography (e.g., where feasible, treatment should proceed from upslope to downslope to minimize re-invasion due to gravity-dispersed seed), and (c) disturbance history (e.g., *Brachypodium* appears to colonize gaps in vegetation with

Figure 27

Brachypodium Treatment Decision Tree



*BRDIS = *Brachypodium distachyon*

4. evidence of soil surface disturbance). Note that some *Brachypodium* invasions (e.g., sparse occurrences on sandy soils) may not be a priority for treatment, particularly where they do not impact target resources.

5. Identify Restoration Strategy. Restoration is an integral part of the *Brachypodium* control strategy. Where a native species component is extant (as determined through habitat assessments), invasive species control may be sufficient to release the native seed bank and promote growth of existing native shrubs and grasses (passive restoration). Where the native component is absent or severely limited, active restoration should include seed, plugs, or container plantings. Supplemental watering during the first and second years may be necessary in seeded areas depending on the amount of rainfall, and will be necessary for plugs and container plantings.

6. Develop and Implement Treatment Plan(s). Focus on (a) removing existing, above-ground biomass, (b) preventing additional inputs to the soil seed bank, and (c) conducting passive or active restoration to minimize gaps for colonization and increase native species diversity. Treatment plans should include management and monitoring goals, objectives, implementation tasks, timeline, and funding and coordinate with regional or preserve-level goals and objectives for covered species and habitats.

   Where the *Brachypodium* infestation is large, the treatment plan may need to be phased. Focus on areas that support or formerly supported target resources, as well as adjacent areas that function as conduits for dispersal of *Brachypodium* seed into treatment areas (roads, trails) in the first treatment phase. Subsequent treatment phases should expand outward from initial treatment areas.

   Brachypodium stands likely can be reduced but not eliminated in 2 years. Therefore, we recommend a minimum 3-5 year treatment plan, recognizing that the level of treatment effort may decrease after year 2 and periodic follow-up treatments may be necessary beyond 5 years.

   Control methods

   The most effective control for *Brachypodium*-infested sites is a combination of a grass-specific herbicide (Fusilade) to treat *Brachypodium* and other nonnative grasses[4]and glyphosate to treat nonnative forbs. Mechanical methods are less effective than herbicide (but more effective than no treatment and are a suitable option where herbicide is too expensive or not appropriate for other reasons.

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[4] Some nonnative grasses and forbs may be more effectively treated with other herbicides.

Dethatching did not significantly improve *Brachypodium* control where herbicide application was uniform, but may be important for promoting native species establishment or reducing biomass that may adversely affect ecosystem processes. Observational data from this study and other restoration projects in the area (e.g., McMillan pers. comm, Dodero pers. comm.) suggest that many native species present in the soil seed bank respond positively to dethatching. Dethatching also increases bare ground and thus, is likely to improve plant-soil contact when introducing plant propagules (seed, plugs, plants). Dethatching prior to herbicide treatment is recommended, where feasible. However, dethatching will add to treatments costs and may not be feasible over large areas using methods tested in this study. In the absence of dethatching, native species richness might increase over a longer timeframe once thatch breaks down, assuming *Brachypodium* is actively controlled (e.g., mowing, herbicide).

While this study utilized line trimmers for mechanical control, selective and appropriately timed grazing or large (mechanized) mowers may provide similar levels of control and prove cost-effective over large landscapes. Neither grazing nor large mowers as control methods for *Brachypodium* were tested as part of this project. In some situations (e.g., rocky soils), large mowers may not effectively control *Brachypodium* if plants are small (Brooks pers. comm.).

Timing

Treat *Brachypodium* when it is approximately 2-6 inches high and prior to flower formation. In some cases, a second treatment will be necessary, depending on rainfall events. We treated *Brachypodium* in February in both a pilot study (CBI 2012a) and this project, which was sufficient for control in 2011-2013. In 2014, rainfall subsequent to the February treatment resulted in a post-treatment germination event and an increase in *Brachypodium* cover.

We also recommend multiple spot-treatment (glyphosate) events per year to accommodate variable nonnative forb phenology. Other studies have shown an inverse relationship between nonnative grass and nonnative forb cover (e.g., Cox and Allen 2011, Cox and Allen 2008a,b, Allen et al. 2005); therefore, the need for nonnative forb control may increase as *Brachypodium* cover decreases. The length of time necessary for ‘intensive’ nonnative forb control will depend on the diversity and longevity of nonnative forb seeds at a given site and, possibly, the degree of site colonization by native species.

Both treatment and post-treatment monitoring may be particularly valuable when climatic conditions promote optimal nonnative grass germination and survival (e.g., a ‘good’ grass year).

7. <u>Monitor Treatment Areas</u>. Monitor treatment areas annually in late spring (following winter-early spring treatments) during the 3-5 year treatment period. Include a qualitative assessment of vegetation composition and cover, *Brachypodium* cover), percent bare ground, and degree of thatch (Appendix B). Adjust treatment frequency based on monitoring results.

   Conduct post-treatment monitoring to detect *Brachypodium* re-invasion in its earliest stages. Treating *Brachypodium* before it establishes a seed bank is more cost-effective than treating infestations with a well-established seed bank. Post-restoration monitoring should be conducted annually until *Brachypodium* has been maintained at low levels (<10% cover) or is absent from the site for 3 consecutive years. Thereafter, monitoring should be conducted with covered species monitoring or every 3-5 years in the absence of covered species. Additional treatments are warranted when *Brachypodium* reaches a cover threshold of ≥10% in previously treated areas.

8. <u>Protect Treatment Areas</u>. Protect treated areas and minimize opportunities for *Brachypodium* re-invasion by installing fencing and/or signage to discourage human incursions (including vehicular traffic), and eliminating or restoring trails through or adjacent to treated habitat. In addition, biologists or restoration contractors working within treatment areas should ensure they are not moving *Brachypodium* seed between sites by cleaning shoes, clothing, equipment, or vehicles between site visits.

9. <u>Equipment Investment</u>. Invest in a Cooperative Mule or similar herbicide-delivery system to facilitate application at a landscape-scale. Due to the cost, land managers within a region or management unit might consider investing in equipment that can be shared among multiple land owners/properties.

7.2 Preserve-specific Recommendations

7.2.1 Crestridge Ecological Reserve

Control efforts on CER resulted in a significant decrease in *Brachypodium* cover in treated areas. Due to high seed viability, productivity, and longevity, the species has the ability to rebound quickly given optimal climatic conditions. Thus, we recommend (a) continuing treatments in treatment areas to ensure the species is either eliminated or maintained at low levels, and (b) expanding treatment areas as funding becomes available.

- Continue treating *Brachypodium* and nonnative forbs, as necessary, within treatment polygons for 3 years. Continue monitoring treatment plots for cover and species richness as outlined in this document.
- If funding for additional treatments is not available, continue monitoring treatment plots for 3 years to determine the longevity of the treatment effect.

- As funding allows, extend herbicide treatments into Phases 2 and 3, respectively, following methods described in this report. In this context, 'phase' refers to the extent of treatment areas; phases can be implemented concurrently or at different times. Refer to Figure 28 for a map of prioritized treatment areas.

7.2.2 South Crest

Control efforts on South Crest were similar to those described above for CER. Here, too, we recommend (1) continuing treatments in these areas to ensure the species is either eliminated or maintained at low levels, and (2) expanding treatments to additional areas as funding becomes available.

- Continue treating *Brachypodium* and nonnative forbs, as necessary, within treatment polygons for 3 years. Monitor treatment plots for cover and species richness as outlined in this document.
- If funding for additional treatments is not available, continue monitoring treatment plots for 3 years to determine the longevity of the treatment effect.
- As funding allows, dethatch additional areas and extend herbicide treatments into Phase 2 treatment areas, following methods described in this report. Refer to Figure 29 for a map of prioritized treatment areas.
- As funding allows, include Phase 3 treatment areas (Figure 29) in a burn treatment; monitor and assess effectiveness of burn + herbicide on *Brachypodium* control.
- Consider selective grazing as a treatment for long-term *Brachypodium* control; time grazing to maximize removal of *Brachypodium* and other nonnative grasses while minimizing impacts to clay soils.
- Retain fencing and signage for at least 5 years to allow establishment of native vegetation.
- At the end of the 5-year fencing and signage period, assess existing trails through or adjacent to restored habitat to determine the need for trail closures/restoration

7.3 Research Recommendations

Refine *Brachypodium* Control Strategies and BMPs

- Continue monitoring treated areas to determine treatment longevity and appropriate intervals for re-treatments (as necessary).
- Continue *Brachypodium* seed studies to inform management of the soil seed bank. Studies may include *Brachypodium* seed longevity/viability, seed depth in the seed bank (e.g., primarily surface versus buried), and seed susceptibility to fire.

Figure 28

Prioritized *Brachypodium* Treatment Areas, Crestridge Ecological Reserve

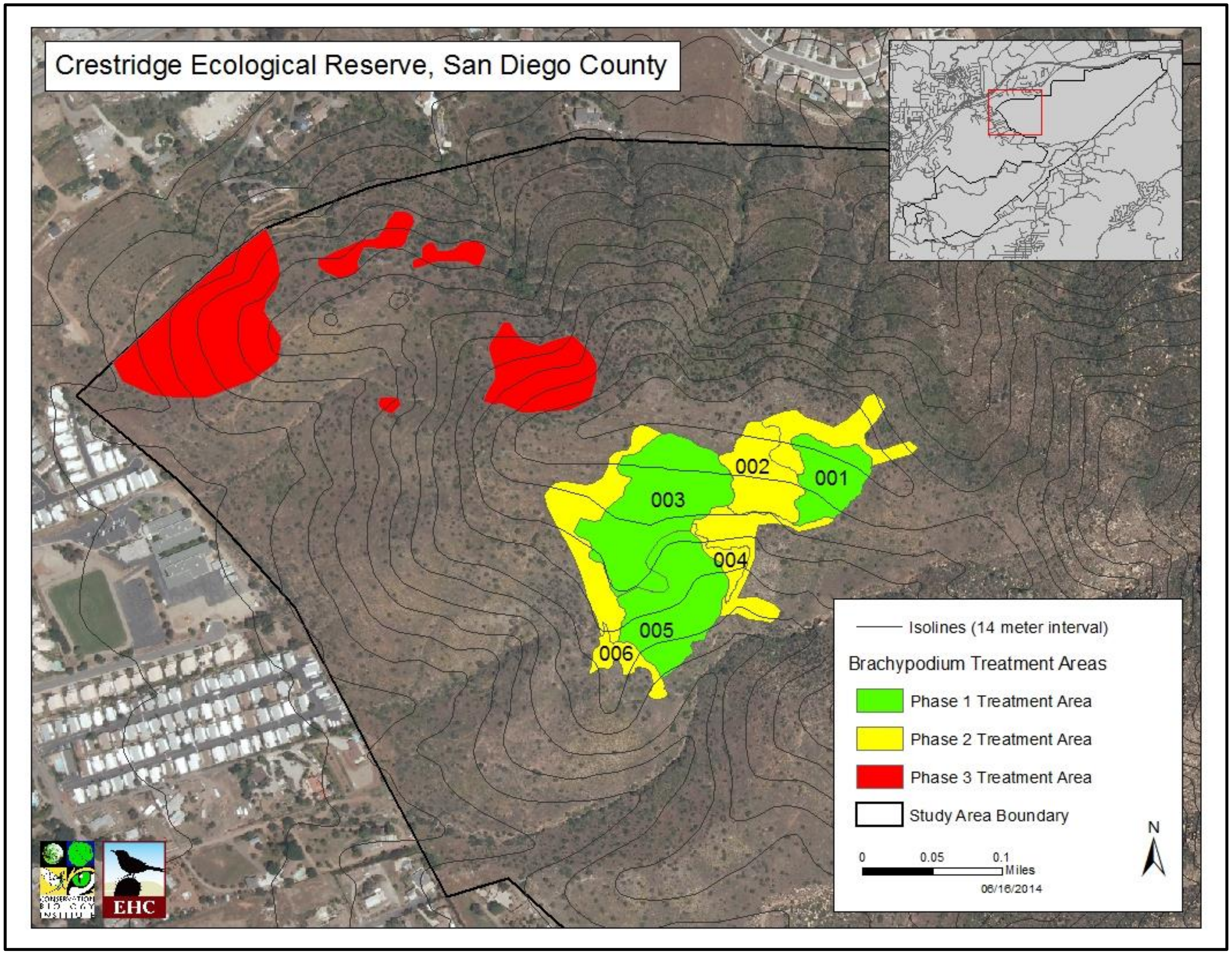
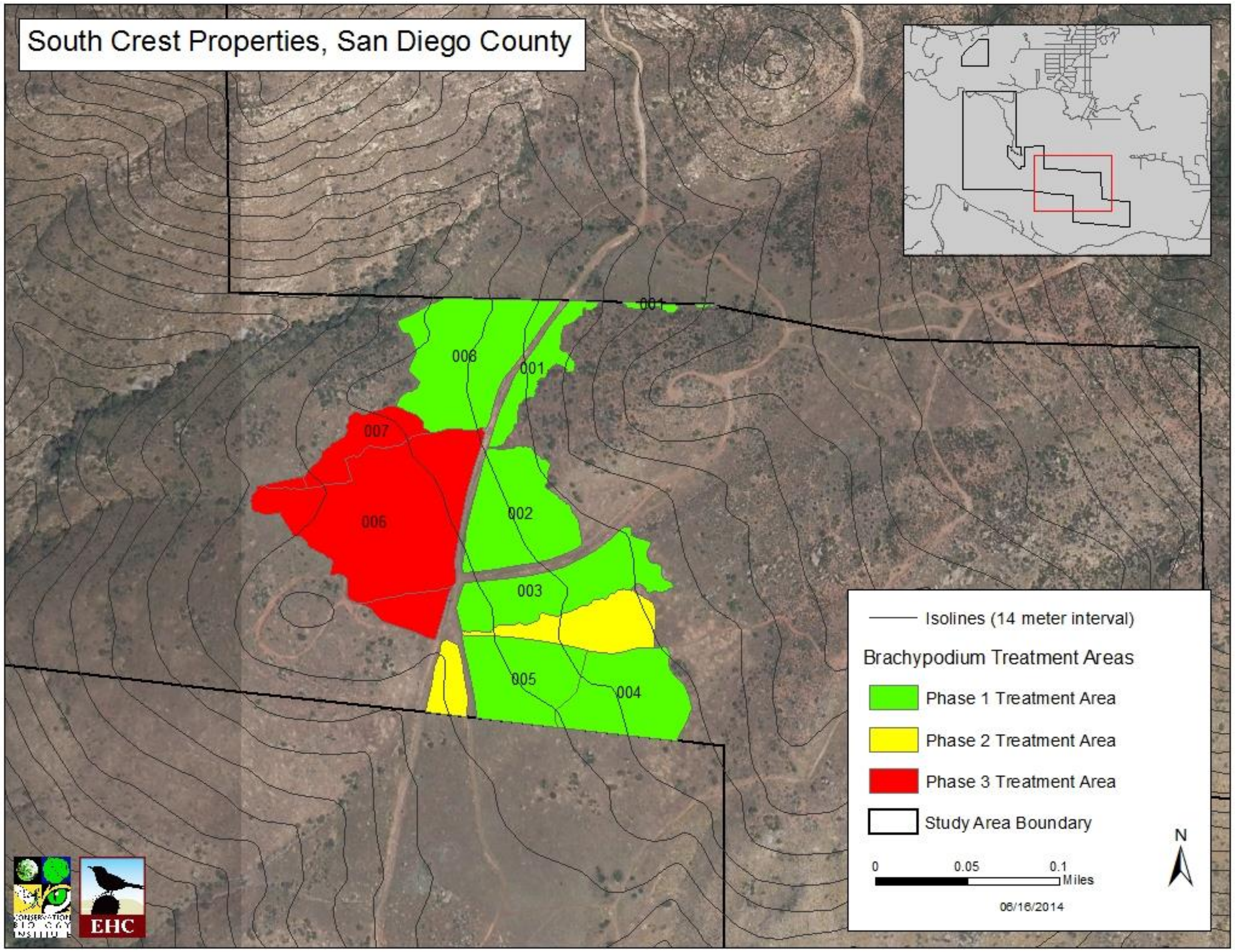


Figure 29

Prioritized *Brachypodium* Treatment Areas, South Crest



- Test additional treatment strategies for *Brachypodium* control that can be scaled up, including grazing and burning.

Develop/Refine Predictive Tools to Enhance Management

- Conduct site-specific soil sampling to refine soil mapping in areas of *Brachypodium* invasion and assess soil properties conductive to invasion. Use results to inform and refine conceptual models and habitat suitability modeling for early detection of *Brachypodium* invasion.
- Determine whether aerial photography and other imagery are useful tools for mapping *Brachypodium* and delineating areas requiring control.

Identify *Brachypodium* Ecosystem Effects that may Influence Management

- Conduct soil ecology studies to determine effects of *Brachypodium* thatch on nutrient cycling and soil fauna; studies should consider residual effects subsequent to thatch removal and effects of altered soil ecology on the native plant seed bank. Use results to modify conceptual models and management practices.
- Monitor burns on clay and gabbro soils for post-fire *Brachypodium* invasion. Use results to refine post-disturbance BMPs.

Identify Additional Species that may be Impacted by *Brachypodium*

- Investigate effects of *Brachypodium* invasion on fauna, including insects, reptiles, small mammals, and birds.

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