Companion planting as an understory restoration strategy along the American River riparian corridor

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Abstract

The purpose of this project is to restore a degraded site located in the Lower American River riparian corridor while conducting an in-situ experiment to evaluate whether the strategy of companion planting with *Artemisia douglasiana* has an effect on other native understory vegetation species (*Leymus triticoides* and *Carex barbarae*). All three of these plants were specifically selected for the ecological and ethnobotanic benefits of adding fire resiliency and cultural value to the site being restored. The experiments conducted did not reveal any significant difference in either the growth or survivorship of *Leymus triticoides* or *Carex barbarae* between the control and companion treatments, but were successful in establishing these species. The experiments did produce a finding of statistical significance that the strategy of companion planting with *Artemisia douglasiana* substantially reduces the colonization of the restoration effort by nonplanted species. More research on the potential of *Artemisia douglasiana* to prevent invasive species takeover of active restoration efforts is recommended.

Introduction

Riparian habitat continues to be lost or degraded around the world. As the buffer between our urban cities and our waterways, riparian corridors also serve a critical role as rare instances of continuous habitat for communities of native organisms to inhabit. Acting as the few remaining refuges for animals like coyotes, deer, river otters, and turtles, riparian corridors like the American River Parkway are sensitive to many forms of disturbance. As the foundation of an ecological community, it is critical to restore understory vegetation and conserve habitat in order to maintain ecosystem functions. Vegetation in the understory consists of ground cover plants, grasses, brush, and even young trees. Understory vegetation in a riparian corridor like the American River Parkway provides grazing for herbivorous species, habitat for small animals and
insects, and bank stabilization against the eroding force of streaming water (Xu & Zhou, 2011). Riparian restoration along the American River provides habitat for wildlife species such as the Valley Elderberry Longhorn Beetle (*Desmocerus californicus dimorphus*), an endangered species, and the Western Pond Turtle (*Actinemys marmorata*), a species of special concern in California (Golet, 2008; Stevens, 2017).

The purpose of this experiment is to observe the effect that the restoration strategy of companion planting with *Artemisia douglasiana* has on the plant growth and survival of selected native understory vegetation species (*Leymus triticoides, Carex barbarae*). Creeping Wild Rye (*Leymus triticoides*) and Santa Barbara Sedge (*Carex barbarae*) are two understory plants native to the Sacramento valley. As rhizomatous plants, these species have coevolved with the natural disturbance regimes historically present in the Sacramento Valley and are fire resilient and drought tolerant species (Lacey, 1974). Mugwort (*Artemisia douglasiana*) is an understory rhizomatous plant native to the Sacramento Valley that is used by the indigenous Nisenan, Maidu, and Miwok tribes as a medicinal and ceremonial plant.

**Background**

**The Lower American River**

The American River is a 120-mile long river that flows downstream from its headwaters in the Sierra Nevada mountain range, ending at the City of Sacramento; the American River meets its confluence with and flows into the Sacramento River at Sacramento (County of Sacramento, 2008). The American River is divided into the Upper and Lower American River at Folsom Dam, with everything downstream of Folsom Dam categorized as the Lower American River (Sacramento River Watershed Program, 2010a). The long-term accumulated deposition of sediments historically resulted in a Lower American River floodplain consisting of rich, alluvial
soils (Stevens, 2015). The Lower American River floodplain has been altered from its historic state by multiple large-scale disturbances that have degraded the riparian habitat along the river corridor.

One of the most significant lasting human disturbances to the American River floodplain occurred as a result of hydraulic mining during the California Gold Rush (Sacramento Area Flood Control Agency, 2015). The massive erosion of the landscape surrounding the Upper American River caused by hydraulic mining from the 1850s-1900s resulted in a massive downstream release of sediment that permanently altered the geomorphology of the Lower American River, burying the historic floodplain (SAFCA, 2015). The sediment eroded from hydraulic mining raised the historic river channel and reduced the soil quality along the Lower American River riparian corridor (SAFCA, 2015). Additional dredge mining of the Lower American River has resulted in changes to the floodplain topography and left large deposits of dredge material which further raise the river channel (SAFCA, 2015). These effects negatively impact both aquatic species and terrestrial vegetation along the riparian corridor by reducing the quality of spawning sediment and soil respectively.

Like most rivers in California, a leading human disturbance to the American River is that it is dammed. The Folsom and Nimbus dams were constructed on the American River in 1955 for the primary purpose of flood control downstream throughout California as part of the Central Valley Project (SAFCA, 2015). As it is defined as the section of the river that is downstream of Folsom Dam, the flow of the Lower American River is significantly altered by human management and dependent on release from both the Folsom and Nimbus dams (SRWA, 2010a). In addition to impacting hydrology, the dams along the American River significantly reduce the flow of naturally eroding sediment. Sediment is trapped at both the Folsom and Nimbus dams,
preventing gradual replenishment of the sediment bed of the Lower American River (SAFCA, 2015).

The construction of levees along the American River riparian corridor severely constrict its potential to flood into its historic floodplain (County of Sacramento, 2008). The historic floodplain of the American River extends far beyond the constructed levees into what is today the City of Sacramento, but was once constituted by an extended riparian forest corridor (SAFCA, 2015). The narrowing of the river channel by levees results in concentration of waterflow and increased erosion of the channel sediment bed, or channel incision (SAFCA, 2015). Combined with the trapping of sediment that would replenish this loss by dams, the Lower American River channel bed’s incision leaves it at a sinking height relative to its banks (SAFCA, 2015). The disparity between bank height and water table height reduces the quality of riparian habitat along the Lower American River corridor (SAFCA, 2015), whereas an undisturbed floodplain would have more even inundation and interaction between the water and the river’s banks.

The Lower American River riparian corridor faces more disturbances than just hydrogeologic changes. The riparian corridor along the Lower American River is designated the “American River Parkway”, for which paved bike trails reside on both banks of the river (California State Government, 2010). The trails of the American River Parkway attract many people to the riparian corridor daily for recreation, transportation, and fishing (County of Sacramento, 2008). Along with foot and bicycle traffic as a disturbance, people bring waste that pollutes the riparian habitat. Another disturbance is that colonization by invasive and non-native species persists all along the riparian corridor, especially with understory vegetation (County of Sacramento, 2008).
The Sacramento River Watershed Program (2010b) identifies aquatic and riparian habitat in the Lower American River as a key management issue as part of the greater Sacramento River Basin. Multiple organizations have been established in order to alleviate the many disturbances the Lower American River riparian corridor faces. Both governmental entities as well as non-governmental organizations and efforts like the American River Parkway Foundation, Save the American River Association, and Bushy Lake Restoration Project work to manage and implement solutions to these disturbances (SAFCA, 2015; Stevens, 2015).

**The Bushy Lake wildlife refuge**

*Bushy Lake as part of the American River riparian corridor*

The Bushy Lake wildlife refuge is a highly disturbed riparian habitat in an urban setting, lying in the Lower American River floodplain (Figure 1). The study area of the Bushy Lake site is located at 38°35'16.0"N 121°25'51.5"W, positioned in between the California Exposition and State Fair facility and the American River in Sacramento, CA (Figure 2). Major past disturbances to the Bushy Lake site include initial development for the construction of a golf course that resulted in soil compaction, fires in 2014 and 2016, and sheep grazing (Stevens, 2015; Stevens, 2017). As part of the American River Parkway, the Bushy Lake site experiences the same prevalent disturbances common to the parkway including the significant presence of invasive understory plant species, human foot traffic, and an increasing presence of homeless encampments (Stevens, 2017).

**SARA and the Bushy Lake Preservation Act, AB 889**

In 2009, the Save the American River Association advocated for California Assembly Bill No. 889 which passed the State legislature. AB 889 expanded the American River Parkway as a protected “open-space greenbelt” to encompass the Bushy Lake site and requires the
California Exposition and State Fair Board of Directors to maintain Bushy Lake “in a manner consistent with the definition of a state park” and “the features of a natural preserve” (California State Government, 2010). This piece of legislation effectively protected the Bushy Lake site as a wildlife refuge without formally recognizing it as a state park, by grouping it under the American River Parkway. Since being protected in the legislature, all developmental activity in the Bushy Lake site has been prohibited except for the Bushy Lake Restoration Project.

The Bushy Lake restoration project and Sac State

As of 2014, the Bushy Lake Restoration Project has been managed by Dr. Stevens of the CSUS Environmental Studies Department in collaboration with Sacramento County Parks and under the jurisdiction of the California Exposition and State Fair Board of Directors (Stevens, 2015; California State Government, 2010). From 2015-2019, Dr. Stevens’ Restoration Ecology class has been engaged in active restoration and wildlife monitoring at the Bushy Lake site during the Spring semester. Further, Bushy Lake has been a topic of research used by a number of Environmental Studies students over the past few years for their senior theses. Past student experiments at Bushy Lake and various senior theses over the years provide a foundation to inform the implementation of this one.

Understory vegetation

The restoration of understory vegetation is an important part of remediating a degraded site. The understory, composed of groundcover, grass, bush, and small tree species, provides important habitat for native animal species and is a rapidly productive part of the ecosystem (Alpert, Griggs, & Peterson, 1999). The existence of understory plants creates habitat for animal species by providing shelter and food (Golet et al., 2008). Through burning or natural decomposition, understory species provide nutrients that replenish and maintain a healthy soil
(Lacey, 1974). Following a disturbance to an ecosystem, growth in the understory is the first step of natural succession (Lacey, 1974). In riparian ecosystems, understory vegetation provides the crucial ecological role of bank stabilization by holding the soil together through root networks to reduce erosion (Golet et al., 2008). The productivity of understory species also provides carbon sequestration with a much faster growth period than that of canopy vegetation or trees (Alpert et al., 1999).

**Traditional Ecological Knowledge**

**What is Traditional Ecological Knowledge?**

Traditional Ecological Knowledge refers to the knowledge systems of native or indigenous people having to do with the environment and ecosystems (Zedler & Stevens, 2018). Traditional Ecological Knowledge is a system of understanding parallel to the scientific knowledge associated with Western academia and common peer-reviewed literature. These two knowledge systems can be complementary (Berkes, Colding, & Folke, 2000), and the utilization of both provides a more informed perspective than relying solely on one or the other.

**Prescribed burning**

For generations, the Sacramento valley has had a long history of being tended by native tribes through prescribed fires (Stevens, 2004b). The Nisenan, Miwok, and Maidu tribes among others all utilized prescribed burning of understory vegetation to stimulate renewal of the understory while simultaneously developing an open habitat with revitalized soils (Hankins, 2013). Fire was also used as a tool to clear out riparian and woodland areas to make them more walkable, inhabitable for animal and human use, and easier to hunt in (Berkes et al., 2000). Through intermittent burning of habitat patches, people native to the Sacramento valley successfully managed biodiverse and functional riparian ecosystems (Stevens, 2004b). The effect
of prescribed fire on rhizomatous understory grasses such as *Leymus triticioides* and *Carex barbarae* was the rapid production of tender, harvestable rhizome shoots (Stevens, 2004b).

**A Focus on Traditional Ecological Knowledge of understory vegetation**

In addition to the scientific literature, this experiment is guided on the Traditional Ecological Knowledge of tribes native to the Sacramento region including the Nisenan, Maidu, and Miwok. The understory vegetation species selected to test the hypothesis of companion planting with *Artemisia douglasiana* are *Leymus triticioides* and *Carex barbarae*. Both *Leymus triticioides* and *Carex barbarae* are rhizomatous grasses native to the Sacramento valley that have been historically tended and harvested by these tribal groups (Stevens, 2006; Zedler & Stevens, 2018). *Artemisia douglasiana* is a native rhizomatous herb that is also of cultural importance to the Nisenan, Maidu, and Miwok tribes (Stevens, 2004b).

*Artemisia douglasiana*

*Artemisia douglasiana* is a perennial herb native to the Sacramento Valley that is commonly known as California Mugwort (Calflora, 2019a). According to Calflora (2019a), it is an herbaceous dicot that occurs in California’s wetland, grassland, woodland, chaparral, and riparian ecosystems. Within these ecosystems, it is found in the understory of open to shady areas. The blooming period for *Artemisia douglasiana* is May through November (Shultz, 2012). *Artemisia douglasiana* visibly grows as a single herbaceous stalk up to a length of .5-2.5 meters above the ground. It also grows underground as part of a rhizome (Shultz, 2012).

*Artemisia douglasiana* has important ceremonial, medicinal, and practical uses in the cultures of the Nisenan, Maidu, and Miwok tribes. Mugwort is used as a medicine to treat both physical and spiritual ailments (Stevens, 2004b). Physical ailments that are in various ways treated with *Artemisia douglasiana* include menstrual and intestinal cramps, stomach aches,
sprains, bruises, rheumatism, swelling, inflammation, and athlete’s foot (Stevens, 2004b).

_Artemisia douglasiana_ is considered a sacred plant in native culture, used for spiritual purification in ceremonies including funerals, prayers, and burning “smudge” (Stevens, 2004b). Smudge burning is a cultural practice in which herbs such as Mugwort are burned to provide spiritual cleansing, as Mugwort is used to generate positive thoughts and dreams and protect against nightmares and haunting (Stevens, 2004b). _Artemisia douglasiana_ leaves emit a strong scent which also make them useful to native tribes as an insect repellant to protect food (Stevens, 2004b).

**Leymus triticoides and Carex barbarae**

*Leymus triticoides* is a perennial grass native to the Sacramento Valley commonly known as Creeping Wild Rye or Beardless Wild Rye (Calflora, 2019c). Another name used to refer to *Leymus triticoides* in the scientific literature is *Elymus triticoides*. The blooming period of Creeping Wild Rye is May through July (Calflora, 2019c; Calflora, 2019d). *Carex barbarae* is a perennial grass native to the Sacramento Valley commonly known as Santa Barbara Sedge or White Root (Calflora, 2019b; Stevens, 2006). The blooming period of *Carex barbarae* is June through August (Calflora, 2019b).

Both *Leymus triticoides* and *Carex barbarae* are herbaceous, rhizomatous grasses that are tended and harvested by tribes native to the Sacramento valley (Calflora, 2019b; Calflora, 2019d; Stevens, 2004a). The rhizomes of these grasses have historically been harvested by native people as a primarily important fibrous weaving material used to construct items including baskets, fishnets, and cradleboards (Stevens, 2006; Zedler & Stevens, 2018). These plants play an important role in native culture as the weaving of these items from their rhizomes is an important practical and spiritual tradition (Stevens, 2004a).
**Fire and Riparian Habitat**

Riparian ecosystems in the Sacramento Valley have evolved to be adapted to fire. Wildfires serve the ecological function of providing an intermediate level of disturbance which improves the ecological diversity of riparian areas (Roxburgh, Shea, & Wilson, 2004). As a fire burns through riparian ecosystems, it primarily destroys the understory vegetation. Understory vegetation is mostly herbaceous making it an available fuel for fires to burn through, whereas trees or canopy vegetation are woody and more resistant to fire (Hankins, 2013). The process of fire as a periodically recurring natural disturbance regime clears out the understory of riparian habitats (Dwire & Kauffman, 2003). This cyclical burning prevents understory overgrowth, burns fallen leaf litter or decomposing material, clears out low-lying branches of woody species, and ultimately triggers the process of natural succession in the understory (Griggs, 2009). The ground space made available after a fire event acts as an open niche for native vegetation to compete for. Further, the clearing of branches allows more percolation of sunlight through the canopy into the understory to stimulate increased vegetative growth. As a result of centuries of coevolution with fire, the native understory of riparian corridors in the Sacramento Valley consists of plant species that are adapted to fire (Dwire & Kauffman, 2003).

**Rhizomes**

Rhizomatous plants have an adapted stem or rhizome that lays horizontally under the ground surface rather than growing vertically out of the ground surface (Xu & Zhou, 2011). The horizontal rhizome stem spreads radially outwards from the original organism and sprouts clonal propagules (Lacey, 1974). Because rhizomes lie under the ground surface, when a fire burns through the understory vegetation, rhizomatous plants still have their rhizomes intact and...
are capable of sprouting back quickly (Lacey, 1974). *Artemisia douglasiana, Leymus triticoides,* and *Carex barbarae* are all rhizomatous plants and were chosen as the focus of this experiment.

Rhizomatous plants were chosen for the restoration of the Bushy Lake site for four reasons. First, the specific rhizomatous plants chosen are all cultural keystone species to the Nisenan, Miwok, and Maidu tribes. Second, rhizomatous understory species provide fire resiliency to riparian landscapes (Lacey, 1974). Third, rhizome networks persist in soils even following a fire and are capable of holding soil together for bank stabilization (Mallik & Rasid, 1993). Fourth, the nature of these rhizomatous plants to spread clonally from a single organism makes their use in restoration cheap, as planting them in lower densities has no negative effect on vegetative growth (Stevens, 2017).

**Goals**

The three goals of this project are: (1) To restore the degraded Bushy Lake site by establishing an understory composed of fire-resilient and drought tolerant native vegetation, (2) To establish a presence of ethnobotanically significant cultural keystone plant species, and (3) To conduct a manipulative experiment to evaluate whether the restoration strategy of companion planting with *Artemisia douglasiana* has an effect on the plant growth and survival of selected native understory vegetation species (*Leymus triticoides, Carex barbarae*).
Hypotheses

Hypothesis 1: The restoration strategy of companion planting with *Artemisia douglasiana* and response of *Leymus triticoides*

Null hypothesis of survivorship: H1S$_0$

The strategy of companion planting with *Artemisia douglasiana* will have no effect on the survivorship of *Leymus triticoides*.

Alternative hypothesis of survivorship: H1S$_A$

The strategy of companion planting with *Artemisia douglasiana* will have an effect on the survivorship of *Leymus triticoides*.

Null hypothesis of growth: H1G$_0$

The strategy of companion planting with *Artemisia douglasiana* will have no effect on the growth of *Leymus triticoides*.

Alternative hypothesis of growth: H1G$_A$

The strategy of companion planting with *Artemisia douglasiana* will have an effect on the growth of *Leymus triticoides*.

Hypothesis 2: The restoration strategy of companion planting with *Artemisia douglasiana* and response of *Carex barbarae*

Null hypothesis of survivorship: H2S$_0$

The strategy of companion planting with *Artemisia douglasiana* will have no effect on the survivorship of *Carex barbarae*.

Alternative hypothesis of survivorship: H2S$_A$

The strategy of companion planting with *Artemisia douglasiana* will have an effect on the survivorship of *Carex barbarae*. 
Null hypothesis of growth: H2G₀

The strategy of companion planting with *Artemisia douglasiana* will have no effect on the growth of *Carex barbarae*.

Alternative hypothesis of growth: H2Gₐ

The strategy of companion planting with *Artemisia douglasiana* will have an effect on the growth of *Carex barbarae*.

**Hypothesis 3:** The restoration strategy of companion planting with *Artemisia douglasiana* and response of nonplanted species

Null hypothesis for *Leymus triticoides*: H₃L₀

The strategy of companion planting *Leymus triticoides* with *Artemisia douglasiana* will have no effect on the growth of nonplanted species.

Alternative hypothesis for *Leymus triticoides*: H₃Lₐ

The strategy of companion planting *Leymus triticoides* with *Artemisia douglasiana* will have an effect on the growth of nonplanted species.

Null hypothesis for *Carex barbarae*: H₃C₀

The strategy of companion planting *Carex barbarae* with *Artemisia douglasiana* will have no effect on the growth of nonplanted species.

Alternative hypothesis for *Carex barbarae*: H₃Cₐ

The strategy of companion planting *Carex barbarae* with *Artemisia douglasiana* will have an effect on the growth of nonplanted species.
Methods

Site description

Climate and weather conditions

The Bushy Lake wildlife refuge study area (Figure 2) is located in Sacramento, CA and experiences a typical Mediterranean climate with mild, wet winters and hot, dry summers (Stevens, 2015). The Spring of 2017 during which this experiment was implemented was an exceptionally wet season, with multiple days setting record highs of precipitation that resulted in temporary flooding of the site (Figure 3).

Hydrology

Despite being along the American River, the water at the Bushy Lake site that constitutes the lake itself is actually pumped from groundwater reservoirs under management of the California Exposition and State Fair facility to remain consistent with requirements to maintain the Bushy Lake site under California AB 889 (Stevens, 2017; California State Government, 2010). The site and study area are not irrigated and do not receive active watering for restoration efforts. Precipitation and groundwater are the primary hydrologic sources to the site (Stevens, 2015). Located in close proximity to the site is Chicken Ranch Slough, a small concrete-lined waterway that transports stormwater through urbanized parts of Sacramento including the Arden-Arcade neighborhood to the American River (Stevens, 2017).

Biota

The Bushy Lake wildlife refuge provides habitat for many animal species. The Western Pond Turtle (*Actinemys marmorata*), a species of special concern in California, inhabits Bushy Lake along with an invasive turtle, the Red-Eared Slider (*Trachemys scripta elegans*). The site provides habitat for the Valley Elderberry Longhorn Beetle (*Desmocerus californicus*)
dimorphus), an endangered species, through the presence of Elderberry (*Sambucus mexicana*) trees (Stevens, 2017). Other animals observed to inhabit or visit the site include deer, coyotes, river otters, squirrels, hawks, and small birds.

The Bushy Lake study area includes both heavily wooded and open grassy areas. The understory of the Bushy Lake site has a large presence of invasive species. The most prevalent invasive species include Poison Hemlock (*Conium maculatum*), Himalayan Blackberry (*Rubus armeniacus*), Yellow Star Thistle (*Centaurea solstitialis*), and Black Mustard (*Brassica nigra*) (Stevens, 2017). The dominant native understory species include California Blackberry (*Rubus ursinus*), Coyote Brush (*Baccharis pilularis*), California Grape (*Vitis californica*), and the understory species this experiment is focused on (*Artemisia douglasiana*, *Leymus triticoides*, *Carex barbarae*). Native trees at Bushy Lake reflect the canopy of the American River Parkway, including various Willows, Oaks, Cottonwoods, Ashes, and Elderberry trees (Stevens, 2017).

**Soil**

The soils in the Bushy Lake wildlife refuge study area are dominantly composed of Rossmoor series fine sandy loam (UC Davis California Soil Resource Lab, 1989). According to the UC Davis California Soil Resource Lab (1989), the second most prevalent soil series on the site is Columbia, composing 6% of the floodplain soil. The Xerofluvents series is also present to a lesser extent, constituting the channel soil (UC Davis California Soil Resource Lab, 1989). The dominant condition of the site is well drained and would constitute ideal conditions for agriculture if irrigated (UC Davis California Soil Resource Lab, 1989). The formation of these soils on the Bushy Lake site is resultant from processes including the alluvial deposition of riverine sediment along the historic floodplain as well as compaction from the disturbances the site has experienced.
Disturbances

The Bushy Lake site was partially developed for the construction of a golf course before it was considered part of the American River Parkway, resulting in alterations to the natural soil formation, topography of the site, and shape of the lake (Stevens, 2017). In 2014 a fire burned through 160 acres along the Lower American River at the California Exposition and State Fairgrounds facility where the Bushy Lake site is located (Stevens, 2015). In 2016 another fire burned through the area of the Bushy Lake site, but the study area itself was protected from burning by the Fire Department who noticed flag-marked plots from former experiments part of the Bushy Lake Restoration Project (Stevens, 2017).

Experimental design

In order to test the hypotheses, two experiments were simultaneously conducted at the Bushy Lake wildlife refuge site: (1) an experiment to evaluate the effect of companion planting with *Artemisia douglasiana* on *Leymus triticoides*, and (2) an experiment to evaluate the effect of companion planting with *Artemisia douglasiana* on *Carex barbarae*. The experiments were implemented in situ at the Bushy Lake wildlife refuge study area (Figure 2). Within each experiment were two treatments, a control treatment represented by monotypic plots, and a companion treatment represented by interplanted plots. Interplanted plots were planted with the native grass respective to the experiment (either *Leymus triticoides* or *Carex barbarae*) and *Artemisia douglasiana* in an alternating pattern, switching every foot (Figure 4). Monotypic plots were planted with a composition of entirely the native grass of the experiment’s focus (Figure 4).

The plots were paired such that each pair consisted of: one monotypic plot to establish a control, and one interplanted plot to evaluate the effect of companion planting. The *Leymus triticoides* experiment consisted of 5 pairs of plots, for a total of 10 plots with 5 replicates of...
each treatment distributed throughout the study area (Figure 5). The *Carex barbarae* experiment consisted of 3 pairs of plots, for a total of 6 plots with 3 replicates of each treatment distributed throughout the study area (Figure 6). Plots were positioned throughout the study area based on where space was available, they were not positioned to specifically represent varying site factors such as shade or water. Any discernable factors varying from the standard visible condition of the site were noted. It should be noted that Plot 1 was positioned near a willow tree and that Plots 8 and 10 were positioned near a prior-existing patch of *Artemisia douglasiana*. (David, Kempton, & Nevison, 1996)

**Procedure**

**Plot Setup**

Experimental plots were implemented in the study area of the Bushy Lake wildlife refuge site in the early Spring of 2017. To set up the experimental plots prior to planting, surface vegetation was cleared exceeding each plot’s boundary by 1 foot in every direction on March 4th, 2017. Once the soil surface was cleared of existing vegetation, any existing root networks in the area of each plot were broken up thoroughly by shovels; roots that were found were removed from the plots. Students in the ENVS 151: Restoration Ecology class at CSUS for the Spring 2017 academic semester assisted in conducting the plot preparation.

Gridded quadrat frames were used to guide the position of planting on March 11th, 2017 (Figure 7). Plants were positioned in a square grid with 1 foot spacing in between each plant (Figure 4). Soil corers were used to dig holes 5.5 inches deep at each planting position. Each hole was inoculated with 1 teaspoon of mycorrhizal fungi (Green Gro Brand, Nature’s Pride with added biochar and mycorrhizae). Seedlings from Hedgerow Farms of *Leymus triticoides, Carex barbarae*, and *Artemisia douglasiana* were planted as according to the experimental design.
Figure 4) in their designated plots (Figure 5; Figure 6). Each planted seedling was marked with a flag to designate the planting position. Flag-marked sample plots allow for long-term monitoring after the implementation of a restoration project (Griggs, 2009).

**Survivorship Assessment**

Survivorship was measured on April 23rd, 2017. Each flag was monitored to assess whether the associated seedling had survived. Survival was determined by visible above-ground biomass where the seedlings were planted. Survivorship was measured as a tally count proportional to the total amount of planted seedlings.

**Biomass Assessment**

Biomass was harvested on April 28th, 2017 for the *Leymus triticoides* experiment and on May 22nd, 2018 for the *Carex barbarae* experiment. In order to minimize destruction of native vegetation through harvesting, 1/5th of each plot was randomly sampled to assess biomass. The gridded quadrat was laid over each plot and 1/5th of the grid area was randomly selected for harvest (Figure 7). Harvest consisted of removing all above-ground biomass present in the selected area. Vegetation sampled for harvest was categorized by species and stored in paper lunch bags specifically marked by species and plot for transportation (Figure 8).

Biomass samples were transported to the lab in Amador Hall 123, located on the CSUS campus. Each bag of vegetation was measured using an electronic scale in grams for a “green biomass” measurement, before being dried and remeasured for a “dry biomass” measurement. The weight of the paper lunch bag, both before and after drying, was independently measured and subtracted afterwards. The green biomass was measured on May 2nd, 2018 for the *Leymus triticoides* experiment and on May 23rd, 2018 for the *Carex barbarae* experiment. The vegetation samples were dried in the lab’s drying oven at a temperature of 104° Fahrenheit throughout May,
with measurement concluding on May 30th, 2018. Vegetation samples dried in the oven for varying amounts of time depending on the quantity of moisture present in the sample. Samples were left in the oven until they were completely dried, determined when additional drying did not result in any decreases to mass.

**Analytical notes**

Microsoft Excel was used to process data and produce Figures 9, 10, and 11. Analyses on Microsoft Excel consisted of paired t-tests to determine statistical. Standard error was calculated independently and error bars on the Microsoft Excel-generated Figures 9, 10, and 11 were adjusted to represent those values. Professor Rafael Diaz of the CSUS Mathematics and Statistics Department examined these analyses to confirm their validity. SPSS was also used to perform ANOVA and ANCOVA analyses through the assistance of Dr. Jamie Kneitel of the CSUS Biology Department.
Results

Survivorship of *Leymus triticoides* and *Carex barbarae*

Initial results for the first semester of the experiment consisted of survivorship counts from April 23rd, 2017. The mean survivorship of all treatments exceeded 99% of all planted seedlings (Figure 9). A paired t-test comparing the monotypic control treatment to the interplanted companion treatment found no significant difference between the means of the two treatments, in both the *Leymus triticoides* experiment and the *Carex barbarae* experiment. Further, the effect size between treatments in both experiments was not large enough to be meaningful, with differences of less than 1% (Figure 9).

Growth of *Leymus triticoides* and *Carex barbarae*

The biomass harvest from consisted of electronic scale measurements processed throughout May 2018. Dry mass measurements (in grams) were analyzed both as (a) mean masses and (b) converted into proportional biomass relative to the total biomass sampled from each plot. Statistical analyses comparing both the actual (mean) and relative (proportional) biomass of the selected native grass (either *Leymus triticoides* or *Carex barbarae*) did not discern a significant difference between the control treatment and the companion treatment in either of the experiments (Figure 10). ANCOVA and ANOVA tests factoring in the varying biomass of *Artemisia douglasiana* within each plot as a variable distinct from the treatment also did not detect a significant difference in the actual or relative biomass of the selected native grass between treatments.

Growth of nonplanted species

A paired t-test comparing the monotypic control treatment and the interplanted companion treatment found that monotypic plots had statistically significantly higher rates of
colonization by non-planted species than did interplanted plots in the *Carex barbarae* experiment (Figure 11), with a p-value of 0.04. The *Leymus triticoides* experiment had a similar trend but a statistically significant difference was not detected in the analysis, with a p-value of 0.052. In the case of both the *Leymus triticoides* and the *Carex barbarae* experiments, the effect size of the difference in colonization by non-planted species between the two treatments was 44%, with the control treatment having more than double the proportional amount of colonization as the companion treatment (Figure 11).

**Discussion**

**Survivorship**

Survivorship is a primary measure of evaluating the success of a horticultural restoration effort (Griggs, 2009). In the context of restoration of a degraded site through the reintroduction of native species, a high survivorship is indicative of success. While an almost even survivorship rate of 99% over the course of the initial semester does not make for data with any meaningful comparative difference (Figure 9), it is great news for satisfying this project’s first and second goals of increasing the presence of ecologically and culturally significant native plant species at the Bushy Lake wildlife refuge. The unusually high levels of precipitation throughout the Spring of 2017 were likely a contributing factor in the highly successful survivorship observed, providing substantial watering to a site that is not actively watered or irrigated at all (Figure 3). Without a statistically significant comparative result, the *Leymus triticoides* and *Carex barbarae* experiments both fail to provide sufficient evidence to reject the null hypotheses that there is no difference on the survivorship of the selected native species between the control and companion treatments (H1S₂ and H2S₂).
Growth

Planted native vegetation

The data did not reveal any significant differences in the plant growth of the selected native understory species as a result of the companion planting treatment (Figure 10). While not a significant difference in either experiment, the relative biomass of the selected native understory species was lower when interplanted with Artemisia douglasiana in both the Leymus triticoides and Carex barbara experiments (Figure 10). One explanation for this pattern could be that when within the same experimental plot, Artemisia douglasiana outcompetes the native species it is interplanted with. Without any significant differences observed, both the Leymus triticoides and Carex barbara experiments fail to provide any evidence sufficient to reject the null hypotheses that there is no difference in the growth of the selected native species between the control and companion treatments ($H_1G_0$ and $H_2G_0$).

Nonplanted vegetation

However, the data did reveal that companion planting with Artemisia douglasiana has the potential to exclude or prevent colonization by non-planted species (Figure 11). The Leymus triticoides experiment had a p-value of 0.052, just exceeding the maximum threshold established by an alpha value of 0.05 for significance, so it did not provide sufficient evidence to reject the null hypothesis of nonplanted species growth ($H_3L_0$). In the Carex barbara experiment, the difference between the companion and control treatments was not only statistically significant with a p-value of 0.04 given an alpha value of 0.05, but the difference had a substantial effect size (Figure 11). With a p-value below the threshold of statistical significance, the Carex barbara experiment provided sufficient evidence to reject the null hypothesis of nonplanted species growth ($H_3C_0$) and accept the alternative hypothesis ($H_3C_A$). In successfully providing
evidence to justify accepting the alternative hypothesis ($H_{3a}$) that companion planting with *Artemisia douglasiana* substantially reduces the amount of colonization by nonplanted species, this finding could potentially have significant implications for active restoration efforts. A main concern of restoration efforts is often dealing with the presence and pressure of nonplanted invasive species (Griggs, 2009). A strategy that has been shown to reduce the impact of nonplanted species, such as the strategy of companion planting with *Artemisia douglasiana*, could provide a solution for dealing with this known problem in active restoration efforts along the American River.

**Goal assessment**

Through the successful reintroduction of these plants, the two experiments achieved initial success in meeting the outlined goals of the project to increase (1) ecologically and (2) ethnobotanically significant native vegetation at the Bushy Lake site. Long-term monitoring is often lacking in the evaluation of restoration projects, without which the lasting impact of restoration cannot be accurately assessed (Palmer et al., 2005). This project’s goal to (3) conduct a manipulative experiment to test the effect of companion planting with *Artemisia douglasiana* was met through the execution of the *Leymus triticoides* and *Carex barbarae* experiments to provide answers to the stated hypotheses. The answers that are drawn from the data provided by these experiments are contingent upon their execution, the limitations of which are important to discuss.

**Confounding and limiting factors**

**Sample size**

The main limiting factor of this experiment was its small sample size. The objectives of the Bushy Lake restoration project, and in particular this experiment, include not just generating
scientific data but also performing active restoration to improve a degraded riparian habitat.

Experimental plots each had a minimum of over 25 seedlings each that were grouped together for the sake of establishing sizable patches of native vegetation in the understory and meeting the primary objective of the restoration effort (Figure 4). The grouping of these seedlings into large experimental plots severely limited the sample size relative to the quantity of seedlings. It would be an error of pseudo-replication to use the number of seedlings as sample size, given that the experimental unit is the plot. Arranging the experimental plots to have fewer seedlings could have allowed for a higher number of plots and subsequently a larger sample size provided the same resources. With sample sizes as small as were used in this experiment, it is difficult to illustrate statistical significance (Figure 5; Figure 6).

Measures of biomass

Above-ground biomass within the experimental plots may not be a very accurate measure of plant growth or success for the selected species *Leymus triticoides* and *Carex barbarae*, which are both perennial rhizomatous plants. As perennial species, these native grasses do not allocate as much of their resources to above-ground biomass as do annual species. Further, rhizomatous plants particularly invest their resources in maintaining their below-ground stem network for nutrient transfer between individual plant organisms and clonal or vegetative propagation of new individuals. In this process, the growth of these native grasses was not accounted for in two ways. First, below-ground biomass was systematically not considered in measures of biomass; it would be completely counter to the goals of restoration to destructively harvest or remove the ability of implemented native species to propagate on their own and recolonize the site. Second, the growth of new propagules sprouting outside of the experimental plots but stemming from plants within the plot are also not considered in measures of above-ground biomass, as the
rhizome connecting these plants is not present above the ground surface. For these reasons, it could be the case that our estimates of plant growth based on above-ground biomass are biased towards both annual species and non-rhizomatous species. Without the intention to destructively harvest the native plants that have been implemented for the purpose of restoration, or an alternative method of measuring the biomass of a stem network we cannot see underground, there is a limitation to how representative of growth our measures of biomass are.

Drying process

Another potentially confounding factor in the measurement of biomass is that vegetation samples were left to dry in the oven in varying durations. Different dried samples could have experienced varying levels of desiccation following treatment in the drying oven, which could have in turn biased the dry mass measurements.

Conclusion

This experiment was conducted with a very small sample size and yet still managed to produce statistically significant results in the case of exclusion of non-planted species. I primarily recommend further research on *Artemisia douglasiana* not as a companion plant embedded in the same experimental plots as other native grasses, but on the topic of using *Artemisia douglasiana* as a buffer or barrier plant between degraded and recently restored patches of habitat to prevent takeover by invasive species. With no evidence to suggest that *Artemisia douglasiana* has a positive effect on either the survivorship or growth of the species it is interplanted with (Figure 9; Figure 10), and a trend of reduced biomass of both the native grasses when interplanted with *Artemisia douglasiana* (Figure 10), the data collected indicates that there is no benefit to the establishment of the selected native species. It can be expected
I would also be interested to see further experiments to continue to evaluate the question of *Artemisia douglasiana* as a companion plant. One such future experiment could be to conduct this same experimental design in a controlled environment such as a greenhouse rather than at the Bushy Lake site. Another idea would be to just repeat the procedures of this experiment with a larger sample size to generate more data for a more representative assessment. It would be interesting to see an experiment focusing on the effect of companion planting with *Artemisia douglasiana* on other species than *Leymus triticoides* and *Carex barbarae*. I would specifically like to see if the selection of native annual grass species as opposed to perennial grass species reveals a more marked effect, as above-ground biomass is a more representative measure of growth for annual grasses than it is perennial grasses.

The use of *Leymus triticoides* and *Carex barbarae* was a strategic choice for the restoration effort at the Bushy Lake site because they both persist longer than annual grasses, and naturally spread outwards from where they are planted through their rhizomes. Further, establishing a rhizomatous network of native vegetation provides benefits to soil erosion prevention and landscape fire resiliency, as following an understory burn the rhizome below the ground should be able to sprout new propagules and quickly begin natural succession.

For future studies I recommend increasing the replicate sample size while possibly reducing the experimental plot size to allow restoration resources to be utilized in a manner that provides more representative and statistically relevant data. Through the reintroduction of these plants the quality of habitat at the Bushy Lake site has been improved. The restoration efforts at Bushy Lake remain ongoing to create a space for the benefit of wildlife, an assemblage of cultural keystone species, opportunity for future student experiments, and members of the community to enjoy.
Figures

Figure 1: Map of the Lower American River (Sacramento Area Flood Control Agency, 2015).

The Bushy Lake wildlife refuge site is indicated by the added arrow labeled “Bushy Lake”.

Figure 2: Map of the Bushy Lake site, the study area is circled in red.
Figure 3: Monthly precipitation charts for the City of Sacramento in the 2017 calendar year (National Oceanic and Atmospheric Administration, 2019).

Figure 4: Intraplot arrangement of the experimental treatments.
Figure 5: Interplot arrangement of the *Leymus triticoides* experiment.

Figure 6: Interplot arrangement of the *Carex barbarae* experiment.
Figure 7: Photograph of gridded quadrat frames used to set up experimental plots.
Figure 8: Photographs of biomass harvest on April 28th, 2018.
Figure 9: Mean survivorship of all planted seedlings in both the *Leymus triticoides* and *Carex barbarae* experiments. Data collected on April 23rd, 2017.
Figure 10: Mean above-ground biomass growth in grams, categorized by species in both the *Leymus triticoides* and *Carex barbarae* experiments. Data collected on May 22\textsuperscript{nd}, 2018.

Abbreviations: LeTr = *Leymus triticoides*, CaBa = *Carex barbarae*, ArDo = *Artemisia douglasiana*. 
Figure 11: Mean plot colonization by nonplanted species of the *Leymus triticoides* and *Carex barbarae* experiments. Data collected on May 22\textsuperscript{nd}, 2018.
References


