Effects of simulated solar arrays on seed dynamics in California desert plant species

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Abstract Seed dormancy is an important bet-hedging strategy for annual species in variable environments, allowing persistence through unfavorable periods. Desert annual species are known for complex seed dynamics, with germination and dormancy influenced by environmental factors including soil moisture, temperature, and light availability. These same factors also affect soil microbes, and can stimulate activity of fungal pathogens affecting seed survival in the soil seed bank. Naturally occurring interactions between desert annual seeds and soil microbes may be affected by a renewable energy boom in California's desert, where large facilities alter natural shade and moisture patterns. If belowground effects differ among species, it could influence seedling emergence aboveground, and ultimately community composition at energy sites. To test whether these shifts in abiotic regimes affect seed dormancy, seed survival, and soil microbial activity, we used experimental panels to alter natural shade and moisture patterns. We asked how treatments affect seed germination rates and seed bank survival for a native annual species pair, one common and one rare. We also tested treatment effect on a widespread, noxious invader of the desert southwest. We found that seed germination was not affected by microhabitats created by experimental panels, or by fungicide treatment intended to reduce soil pathogen activity. We then divided apparently dormant seed between experiments testing for survival and pathogen infection, subjecting seed to a tetrazolium assay or plating them on medium supporting fungal growth. Microhabitat and fungicide affected seed survival only in the rare endemic. Microhabitat only affected fungal growth on plated invader seed, with higher growth on seed from the shade microhabitat; fungicide treatment did not affect fungal growth for any species. Regardless of microhabitat and fungicide treatment, we observed striking differences in germination across species, with a much higher proportion of seeds germinating for the invader and rare species compared to the common species.

KEYWORDS: *Eriophyllum wallacei, Eriophyllum mohavense, Brassica tournefortii*, desert annuals, Mojave Desert, solar development, seed dormancy, seed bank persistence

Introduction

The dwindling supply of fossil fuels and rising concern about the climate implications of their use are driving demand for alternative energy sources. As a result, solar energy facilities are proliferating in the desert southwest. California aims to produce 50% of the state's energy from renewable sources by 2030, converting 488,000 acres of desert landscape to renewable energy sites. The Desert Renewable Energy Conservation Plan (DRECP) is intended to facilitate this land conversion while remaining sensitive to potential impacts on threatened species - yet little work has been done to understand the ecological effects of large-scale renewable energy facilities. While effects from development induced habitat disturbance are anticipated (e.g., negative effects of road construction), but other effects such as shading and water runoff from solar panels may also facilitate invasion (Gelbard and Belnap 2003). The Mojave is home to many species that may respond differently to disturbances imposed by solar panels. Comparing responses between rare and common natives, as well as exotic invasive species may suggest whether ecological effects are likely to vary across species.

In undisturbed desert landscapes, topography drives variation in abiotic factors (e.g. temperature, moisture) that may in turn affect underground biotic diversity. Therefore, local microenvironments are characterized by shifts in both biotic and abiotic factors (Randall et al. 2010). Solar panels alter light availability, soil moisture, transpiration rates, and soil temperature (Armstrong et al. 2016, Hernandez et al. 2014, Lovich et al. 2011), with potentially strong consequences for soil communities and annual seed banks. In particular, panel effects on soil moisture should influence germination; conditions under panels are drier, and rainfall runoff increases moisture near the edges of panels. There is a known correlation between seed germination and moisture availability, with increasing moisture driving higher germination rates

(Pake and Venable 1996, Freas and Kemp 1983). Changes in soil moisture can also alter soil pH and nutrient availability (Pakeman et al. 2011), leading to shifts in soil microbial communities. In particular, an increase in soil moisture can promote higher abundance of fungal pathogens, influencing the development, distribution, and longevity of fungal spores (Mordecai 2012, Schafer and Kotanen 2003). Fungal pathogens can reduce fecundity in specific hosts, leading to shifts in seed bank composition that ultimately affect aboveground community composition (Schafer and Kotanen 2003).

In addition, poorly managed energy development may promote habitat fragmentation and spread of invasive plants (Randall et al. 2010). Invaders arriving from arid regions may already possess dormancy strategies suited to unpredictable desert rainfall (Dahlin et al. 2012); such invaders can compete strongly for soil moisture and nutrients, threatening native and endangered species in resource-limited ecosystems (Barrows et al. 2009, Berry et al. 2014). In the absence of such pre-adaptation, solar arrays could facilitate invasion by creating more mesic conditions favoring weedy species (e.g., via panel washing and road runoff). *Brassica tournefortii* is often found along roadsides, is highly fecund, and germinates under a wide range of conditions (Berry et al. 2014) – suggesting this desert invader will thrive near energy facilities. Once established, invaders like *B. tournefortii* may more easily colonize surrounding wildlands, altering community composition and leading to loss of plant and animal diversity (Berry et al. 2014, Dahlin et al. 2012).

Here we use experimental panels to create microhabitats similar to those present at photovoltaic arrays, testing for effects on seed dynamics of two natives and one invader: *Eriophyllum wallacei, Eriophyllum mohavense*, and *B. tournefortii*. We installed artificial seed banks in the open and in microhabitats created by experimental panels to test for effects on seed

germination and survival rates. We predict germination will be higher in microhabitats where soil moisture is higher, and that seed survival in mesic microhabitats will be lower. We test whether pathogens influence seed survival by treating half of our artificial seed banks with a fungicidal soil drench protecting against oomycetes and fungal pathogens. We hypothesize that novel microclimates created by experimental panels will influence fungal infection rates (e.g., higher infection rates in microhabitats with more moisture, or where moisture loss to evaporation is slower) Differences in germination, seed survival, or infection rate with fungicide treatment would suggest an effect of fungal pathogens on seed dynamics in this system.

Methods

Study system and sites

Our study sites are located within 40 km of Barstow, California, in the eastern Mojave Desert. Considered one of the driest areas in the United States, this region receives less than 30 centimeters of rainfall annually (Randall et al 2010). Daytime temperatures are warm, and high winds are frequent. All study sites are situated in creosote- bush scrub dominated by widely spaced shrubs including *Larrea tridentata, Yucca schidigera,* and associates (Randall et al. 2010). Each of our sites supports a large population of a single focal species, with permanent plots established during earlier demographic work. All sites varied in soil substrate type. *E.wallacei* site was the most heterogenous and had variation between coarse rocks and sizes of gravel. *B.tournefortii* site is comprised of a mixture of sand and finer gravel. *E.mohavense* site has the most refined soil of the three, in fine sandy soils.

Focal species

E.wallacei is a small annual herb that grows no taller than 15 cm. A member of the Asteraceae family, individual plants are generally tufted with small hairs and produce one flower head per stem with 5-10 yellow ray florets per head (Baldwin et al. 2012). *E.mohavense* is a rare, California endemic (California Rare Plant Rank 1B.2), with a narrow distribution centered near Barstow. It grows less than 3 cm tall, with white wooly hairs on stems and leaves (Baldwin et al. 2012). Cylindrical flower heads are 1-3 mm wide, containing 3-4 bright yellow disc flowers (Baldwin et al. 2012). Achenes (hereafter called seeds) of *E.wallacei* and *E.mohavense* are morphologically similar - both are less than 3 mm long and 1 mm wide (Baldwin et al. 2012). By studying the *Eriophyllum* species pair, we can assess whether there are differences in treatment response between a rare species and its common congener.

Native to Africa and the Middle East, *B.tournefortii* (Brassicaceae) is a common invader of disturbed sites and wildlands in the desert southwest (Minnich and Sanders 2000). Growing to a height of 100 cm, individuals may produce up to 16,000 seeds (Brooks et al. 2006). Seed germinates well under a wide range of temperatures and light levels, and it can tolerate moderate salt concentrations. Rapid, early growth also allows this species to establish before native desert annuals, pre-empting resources. All of these traits likely contribute to *B.tournefortii's* success as an invader (Brooks et al. 2006, Bangle et al. 2008).

Experimental panels

Experimental panels simulate commercial solar panels, at a reduced scale: 2' x 2' panels consist of a rebar frame holding a wooden shield at a 30° angle. Clear corrugated plastic sheeting (4 mm Coroplast, CorrugatedPlastics.net) was fixed to wooden shields to improve water runoff.

Panels are oriented to face south, and the shade and runoff microhabitats are defined by their position relative to panels (Figure 1). Previous analyses revealed a ~11°C difference in average soil temperature between shade and control microhabitats near solar noon, with large reductions in photosynthetically active radiation (~70%) under panels (*K. Tanner, manuscript in prep*). At each study site twenty experimental panels were randomly divided between the microhabitat-only and the microhabitat-fungicide experiments. Control locations were established 1 meter south of experimental panels in both treatment groups.

Artificial seed banks

In summer 2016 we sewed seeds into artificial seed banks constructed from polyorganza fabric, with individual cells for each seed (Figure 2). Due to the limited production of *Eriophyllum* seed in 2016, we used the 2016 seed crop in seed banks for the microhabitat-only experiment, and 2015 seed in seed banks for the microhabitat-fungicide experiment. Seed availability for each species determined the numbers of seeds included per seed bank (Table 1). The 2016 *B.tournefortii* seed crop provided enough seed to construct artificial seed banks for both experiments. In October 2016 we deployed seed banks at all three field sites. At each site, 60 seed banks were divided among the control, runoff, and shade locations at 10 plots in the microhabitat-only experiment. Sixty seed banks were divided among control, runoff, and shade locations at 10 plots in the microhabitat-fungicide experiment; half of the packets in each location were treated with fungicide (see *Fungicidal treatment* below). Seed packets were buried in each microhabitat with their long edge parallel to the prevailing physical gradient: we oriented packets east- west in the shade, to align with movement of the sun; and north- south in the runoff, to align with the moisture gradient created by runoff at the south edge of panels. We covered seed banks with a thin layer of soil and a hardware cloth cover held down by nails to

keep seed banks in place. For both experiments, seed banks were collected at the end of the growing season (March 2017), stored in paper envelopes, and brought to UCSC labs for processing.

Fungicidal treatment

To test for soil pathogen effects on germination and seed survival in the microhabitat-fungicide experiment, we treated half of the seed banks in each microhabitat with a soil drench protecting against soil fungi and oomycetes. The drench was prepared using Subdue MAXX (Syngenta) and Cleary's 3336 (NuFarm) (Table 2, Figure 3). Prior to applying the soil drench, seed banks were isolated with a physical barrier to prevent lateral spread of liquid to untreated packets nearby (Figure 3). The same volume of water was applied to untreated packets as a control.

Visual inspection for germination

We compared germination rate across microhabitat and fungicide treatment for each species. We carefully removed seeds from seed bank packets using forceps, and observed them individually under a Nikon SMZ800 microscope. Recovered seed was scored as "germinated" or "dormant." Seeds were scored as germinated if (1) only an empty seed coat remained; (2) the seed coat was ruptured; or (3) if we observed remnant a radicle or shoot (Figure 4). Intact seeds were assumed to be dormant, and were set aside for tetrazolium and plating assays.

Tetrazolium assays for seed survival in microhabitat-fungicide experiment

To test for seed survival we conducted a tetrazolium (TZ) stain assay. Respiring tissues in the seed convert colourless tetrazolium chloride to a water- insoluble red carmine formazans,

indicating active respiratory processes (Porter et al. 1947). We first conducted a pilot to test the accuracy of the TZ assay, boiling 10 seeds of each species in deionized water for 5 minutes before staining (Porter et al. 1947). None of the embryos from boiled seeds stained red, indicating the TZ assay is a reliable method to test for seed survival in our species.

Half of the apparently dormant seed for each species was allocated to TZ assays, and we first allowed seeds to imbibe deionoized water for 24 hours. We prepared a 1% solution of tetrazolium chloride using solid 2,3,5-triphenyltetrazolium chloride and deionized water. After soaking, seeds were cut longitudinally using a precision knife (Xacto #11 blade) to expose the embryo and pericarp, and cut seeds were placed in the prepared solution. *B. tournefortii* and *E. wallacei* seeds were soaked in TZ solution for 24 hours at 17°C. In preliminary tests, *E. mohavense* did not stain as readily as the other species. For such cases, specialists at Ransom Seed Labs in Carpteneria, CA suggested soaking seeds at higher temperature for longer periods to facilitate chemical reactions. We therefore soaked *E.mohavense* seeds in TZ solution for 6 hours at 35°C. Following TZ soak, seeds were observed under a Nikon SMZ800 microscope within 1 hour for signs of red stain. The intensity of staining varied across and within species, so we classified seed exhibiting any red stain as viable. Seeds lacking red stain were classified as dead (Figure 5).

Plating assays for fungal infection rate of seeds

We tested for differences in fungal infection of seeds in the microhabitat-fungicide experiment by plating half of remaining dormant seeds on medium promoting fungal growth (N (*E.wallacei*)= 121 seeds, N (*E.mohavense*)= 104 seeds, and N (*B.tournefortii*)= 39 seeds). To ensure that growth would reflect actual seed infection (i.e., agents penetrating the seed coat), we

surface sterilized each seed before plating. Seeds were rinsed for 30 seconds each in 70% ethanol solution, 10% bleach solution, and deionized water. Seeds were plated on Malt Extract Agar (MEA) media with 0.01% Chloramphenicol on 60 x 15 mm sterile culture dishes, with one seed per plate. We included Chloramphenicol, an antibiotic, in order to isolate growth to fungal taxa present. Seeds were checked for growth at 24 hour intervals. We recorded presence or absence of fungal growth.

Data Analysis

We analyzed data for each species assessing effects of microhabitat and fungicide treatment. We used the 'aov' function in the base package of R (version 1.1.442, R Core Team 2016) to build models for seed germination. In our models fixed effects were microhabitat and fungicide treatment and the interaction between the two. We used the 'Anova' function in the car package (Fox and Weisberg 2011) to extract model p-values.

We used the 'glm' function in base R to build generalized linear models (GLMs) using binomial error distributions for seed survival and fungal infection. Across all models we used the 'summary' function in base R to output results on model-fitting and to compare models. In cases where we observed significant predictor effects, we applied post-hoc Tukey's tests to try and identify which microhabitats differed.

Results

Germination in the microhabitat-only experiment

Microhabitat had a marginally significantly effect on *B.tournefortii* germination (Table 3; Figure 6); germination was lowest in the runoff microhabitat, and highest in the shade microhabitat. Microhabitat had no effect on germination of either native species. Differences in germination of the 2016 seed crop were striking across species; *E.wallacei* germination was

relatively low (~20%), while *E.mohavense* and *B.tournefortii* were much higher (~80% and ~85%, respectively; Figure 6).

Germination in the microhabitat-fungicide experiment

Germination rate did not differ across microhabitat or fungicide treatment for either the 2016 *B.tournefortii* seed crop or the 2015 *Eriophyllum* seed crops (Table 3, Figure 7). Differences in germination rates across species remained strong, with *E.wallacei* germination ~21%, *E. mohavense* germinatinon ~53%, and *B.tournefortii* germination ~92% (Figure 7).

Seed survival in the microhabitat-fungicide experiment

Microhabitat had a significant effect on *E.mohavense* seed survival; in the fungicide control group, seeds survived at the highest rate in the runoff and at the lowest rate in the shade. (Table 4). Fungicide was not significant as a main effect, but there was a marginally significant interaction between microhabitat and fungicide; survival was lower for fungicide-treated seeds in the control and shade microhabitats, and higher in the runoff microhabitat. We did not find an effect of microhabitat or fungicide treatment on survival of *E.wallacei* or *B.tournefortii* seed (Table 4). Similar to patterns for seed germination, survival was highest for *B.tournefortii*, lowest for *E.wallacei*, and intermediate for the endemic *E.mohavense* (Figure 8).

Testing for pathogenic agent/fungal growth in microhabitat-fungicide experiment

The interaction between microhabitat and fungicide treatment had a significant effect on fungal growth for plated *B.tournefortii* seed (Table 4). Within the control microhabitat, fungal growth was higher on seed from the fungicide treatment; within the shade and runoff microhabitats, fungal growth was lower on seed from the fungicide treatment. There was a marginally significant effect of microhabitat on fungal growth for plated *E.mohavense* seed (Table 4). The shade microhabitat had the largest proportion of fungal infection. There was no

effect of microhabitat, fungicide, or their interaction for fungal growth on plated *E.wallacei* seed. The endemic *E.mohavense* had the highest proportion of seeds with fungal growth (Figure 9) while its common congener, *E.wallacei*, had the lowest proportion (Figure 9).

Discussion

Seed Germination

We expected higher germination rates in the panel runoff microhabitat, where soil moisture should be higher compared to the shade and control microhabitats. We might also have expected lower germination in shade compared to the control, due to blocking of rainfall by panels. However, microhabitat rarely exerted an influence on germination. *B.tournefortii* had a near 100% germination rate regardless of microhabitat or fungicide treatment (Figure 6, Figure 7). This is perhaps unsurprising for an invader known for its ability to germinate under a wide range of temperature and light conditions (Brooks et al. 2006, Bangle et al. 2008). We observed a marginally significant effect of microhabitat on *B.tournefortii* seed germination in the microhabitat-only experiment (Figure 6, Table 3); surprisingly, the runoff microhabitat had the lowest germination rate. Although we expected lower germination in the shade due to panel blocking effects, we did not see any difference in germination between the shade and control microhabitat. The experimental panels are small and it is possible that rain could have been blown underneath- rainfall could have contributed to the observed outcomes. Despite using the same *B.tournefortii* seed crop for artificial seed banks in both the microhabitat-only and microhabitat fungicide experiments, we observed no effect of microhabitat in the latter (Table 3).

Germination of *E.mohavense* seed did not differ across control, shade, and runoff locations in either the microhabitat-only experiment or the microhabitat-fungicide experiment. However, we saw a relatively large difference in germination rate across the seed crops used in

these experiments - the 2016 *E.mohavense* seed crop had an average germination rate of 80%, while the 2015 seed crop had an average germination rate of 50% (Figure 6, Figure 7). This result suggests a reduction in viability with seed age- other authors have found a loss of viability with seed age for species using dormancy to spread risk of reproductive failure over multiple growing seasons (Phillipi 1991). However, because the 2015 seed crop was stored under ambient laboratory conditions until artificial seed banks were fabricated in 2016, it is unclear whether a similar loss of viability would have been observed for seed aging in the field.

E.wallacei germination rates were also unaffected by microhabitat and fungicide treatment. For this taxon, seed age appeared to have less effect on germination rate. The difference between seed crops was reduced, and the older seed actually germinated at a higher rate; the 2016 seed crop had an average germination rate of 15%, while the 2015 rate was 20% (Figure 6, Figure 7). This reversal suggests an alternative explanation for different germination rates across seed crop from different years. We know growing conditions can vary across years (e.g., due to differences in rainfall), and if the growing conditions experienced in a particular season affect seed quality, this could in turn affect germination patterns when comparing seed produced in different years.

We observed large differences in germination rate between the rare and common *Eriophyllum* species, which was not entirely unexpected. Other studies on desert species show that dormancy is largely unpredictable and is affected by life-history traits and environmental cues (Pakeman et al. 2011, Phillip 1991). However, *E.mohavense* had a higher germination rate than its common relative, which we did find surprising. This endemic species has a narrow distribution, and appears to be restricted to “soil islands” of relatively ancient origin exposed through weathering (ERT, 1988). In contrast, *E.wallacei* has a broad distribution in the Mojave

Desert, which suggests a more “generalist” life history strategy able to survive a broader range of environmental conditions. We might therefore expect E.mohavense to exhibit more refined germination cues than its common relative. However, our ability to compare these rates directly is somewhat limited. The sites occupied by these species are separated by ~60 kilometers of distance, and about 200 m in elevation – so weather conditions were not identical across sites. Furthermore, the substrate at these sites is quite different. The soil at the *E.mohavense* site is quite fine, and appears to hold moisture well. The substrate at the *E.wallacei* site is a mix of relatively coarse gravel and soil, likely with much lower water-holding capacity. Prior analyses of soil moisture content following rain do suggest better water-holding capacity at the *E.mohavense* site (*K. Tanner, manuscript in prep*). These differences in soil substrate and retention of soil moisture could influence germination rates, which are correlated with moisture availability (Ayerst 1969). Nonetheless, while favorable edaphic conditions have been linked to seedling success, environmental cues for germination can be highly variable (Pakeman et al. 2011, Pakeman and Small 2011).

Effects of fungicide on seed survival

Changes in edaphic factors can influence important soil pathogens, with increased soil moisture leading to increased pathogen activity (Ayerst 1969, Mordecai 2012). We predicted that survival of dormant seed could be mediated by soil pathogens. If so, we should observe higher seed survival in packets treated with fungicide. However, if mutualist fungal agents are more important than pathogenic agents in our system, we might expect to see lower survival for seeds treated with fungicide. Furthermore, the relative influence of pathogenic versus mutualistic agents on seed survival might shift across microhabitats. For *E.mohavense* alone, we observed a significant effect of microhabitat on survival, and a marginally significant interaction between

fungicide treatment and microhabitat; otherwise, fungicide treatment had no effect on seed survival (Table 4). For *E.mohavense,* survival of untreated seeds was higher in the control and runoff microhabitats, and survival of treated seeds was higher in the shade microhabitat (Figure 8). These results suggest a switch in the direction of fungicide's effect across microhabitats, which might be related to relative importance of pathogenic or mutualistic soil microbes across these locations. Our use of a broad spectrum fungicide treatment did not target soil pathogens specifically, and may have also killed beneficial fungal partners influencing seed survival; to tease out the relative influence of each class of soil microbes across microhabitat would require more a discriminating fungicidal treatment. While we did not observe clear patterns within a microhabitat we found significant differences in seed survival between microhabitats. For *E.mohavense,* the lowest average seed survival was in the control microhabitat and highest average seed survival in the shade microhabitat (Table 4). The effects of abiotic conditions and pathogen mediation are unclear raising the questions: are pathogens important in arid environments? While fungal activity may be mediated by soil moisture content, studies show that fungal community development in deserts can be influenced by other factors i.e. space and time (Zak et al. 1995).

Presence of pathogenic agents

We tested for fungal infection of seeds to assess efficacy of fungicide treatment, and to aid in interpretation of germination and survival patterns. We found no influence of microhabitat or fungicide treatment on fungal growth from plated *Eriophyllum* seed (Table 4). We can imagine at least three possible explanations for this pattern. First, fungal pathogens may infect seeds in this system, but our fungicidal treatment was ineffective, leading to the finding of no difference between treatments. We used a one-time, broad spectrum soil drench treatment

applied in fall, before the onset of the rainy season; our fungicide treatment may have degraded over time, and applying it more frequently or during the rainy season may have yielded the desired effect. Different fungicidal agents may also have yielded different outcomes. Lastly, it is also possible that soil pathogens are not important determinants of seed dynamics in this system. However, for the invader *B.tournefortii*, we observed a significant effect of the interaction between fungicide treatment and microhabitat: we saw higher fungal growth on untreated seeds in the runoff and shade microhabitats (Table 4, Figure 9). Although this finding is intriguing, the lack of difference in *B.tournefortii* seed survival and germination rates in the fungicide experiment suggests that these differences in fungal growth patterns are not driven by taxa influencing seed dynamics. One suggestion would be that native species, evolving with other plant residents, have had the opportunity to adapt to local abiotic and microbial conditions, leaving stronger effects of soil pathogens on invaders (Kawecki and Ebert 2004, Levine et al. 2004).

While we did not find clear correlation of fungal infection rates between treated and untreated seeds for *Eriophyllum* species, we found general differences in fungal infection rates across sites. The highest fungal growth was observed in seeds from the *E.mohavense* site; the least fungal growth on seeds from the *E.wallacei* site, and intermediate growth on seeds from the *B.tournefortii* site. These findings may be related to differences in substrate at each site and could be an important factor influencing microbial communities (Schafer and Kotanen 2003). The *E.mohavense* site is characterized by fine soil, with greater moisture holding capacity; the substrate *at E.wallacei* site is a heterogeneous mix of soil and medium fine to coarse gravel; and the *B.tournefortii* site is a mix of sandy soil and finer gravel. The soil at *E.wallacei* likely drains quickly, and may discourage flourishing soil microbial communities; in contrast, the more slowly

draining soil at the *E.mohavense* site may promote belowground fungal activity. Variation in substrate is known to influence spatial variation in microbial communities and contribute to microbial heterogeneity in the soil (Schafer and Kotanen 2003).

Implication for solar facilities

Solar facilities make important contributions to alternative energy capacity, and promote sustainable development – yet these benefits may not be without ecological cost. Large scale development may affect habitat quality and plant performance in a variety of ways, including disturbance from construction sites, maintenance roadways, and shading from panel arrays. With these studies, we hoped to reveal the influence of experimental panels on seed dynamics for two closely related natives and an exotic invader. However, we found little evidence for strong direct effects of microhabitat on seed dynamics, or for indirect effects of microhabitat mediated by seed pathogen activity in the soil. We did find differences in germination and seed survival among species, likely driven by differences in life history strategy. In this study, our invader exhibited very high rates of germination and seed survival regardless of conditions – as might be expected for an opportunistic species. In contrast, germination and survival rates for our native species were generally lower. Our Mojave Desert natives may therefore rely more heavily on temporal bet-hedging through seed dormancy. Whether this is a better strategy than opportunism depends on the frequency and duration of unfavorable periods; with very high germination rates, the invader seed bank could be drained relatively quickly if seeds germinate at high rates in years where reproductive success is low. On the other hand, to the extent that shading and water runoff from solar infrastructure mitigate drought conditions, opportunistic invaders could be favored even in years when drought conditions prevail. Clearly, a robust understanding of solar panel effects on plant performance will require longer-term study, capturing response across multiple

growing seasons in the highly variable desert environment. Our findings so far suggest that panels have no strong effect on seed dynamics for species considered here. However, it is important to note that this study did not examine aboveground life stages for any of our species, and because solar development impacts will play out across the life cycle, we cannot rule out strong effects of microhabitats at later life stages. It is also possible that future climate change will drive environmental conditions that render species more susceptible to microclimate effects, even though we do not now observe them. For these reasons, and due to the limited period of our study, the results reported here should be interpreted with caution.

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Appendix

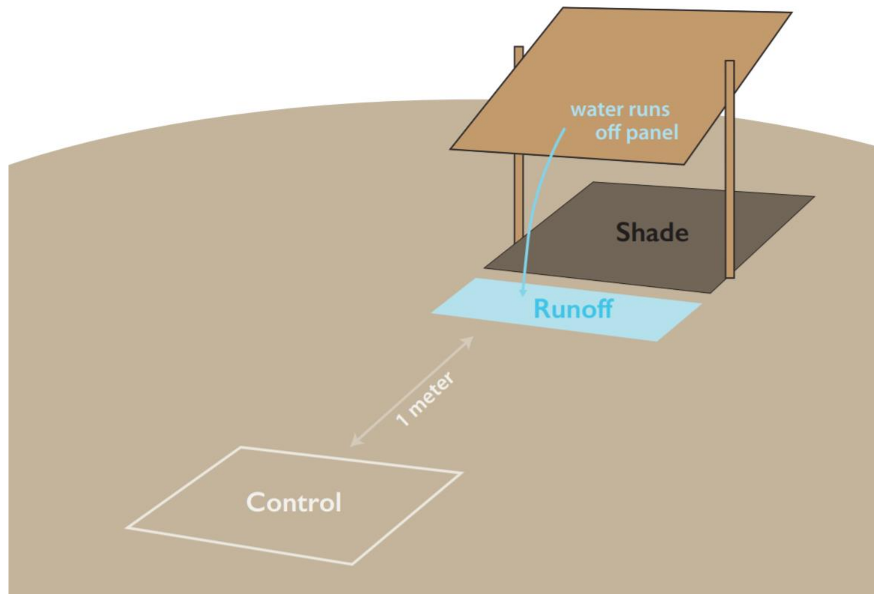

Figure 1: Diagram of an experimental solar panel and the microhabitats it creates: the shade microhabitat is directly underneath the panel, the runoff microhabitat is underneath the southern edge of the panel, and the control microhabitat is 1 meter south of the experimental panel. We used 20 experimental panels at each site: 10 panels for the microhabitat-only experiment and 10 panels for the microhabitat-fungicide experiment.

Figure 2: Stack of *B.tournefortii* artificial seed banks awaiting deployment and burial at plot. Packets were constructed from polyorganza bridal veil, with polyester thread dividing packets into cells holding one seed each.

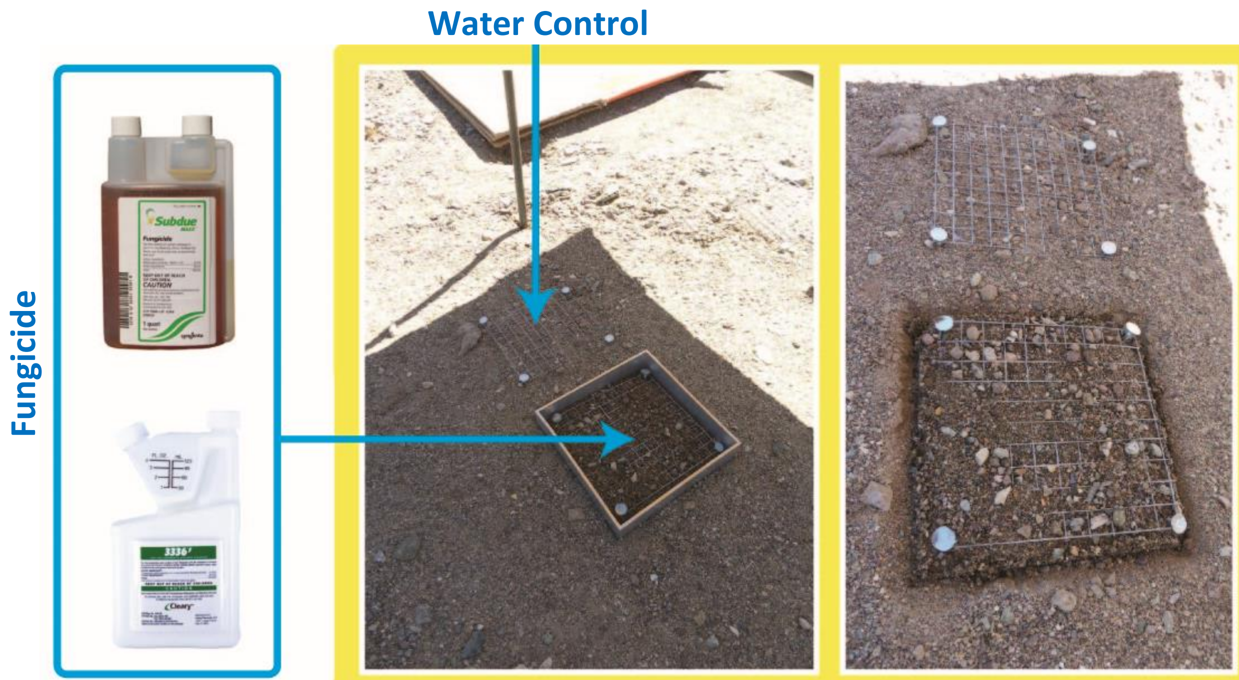

Figure 3: Visual representation of treatment in microhabitat-fungicide experiments at each plot (10 plots per site) and 3 microhabitats per plot (control, runoff, shade). Packets were installed in September 2016 and covered with minimal layer of soil and a square 1/2 " hardware cloth secured by 5" nails and given a one-time soil drench treatment. Fungicide plots were treated with 6oz of Subdue MAXX (Syngenta) and Cleary 3336^{F} (NuFarml) solution to offer broad protection against fungal and oomycete agents (Table 2). At each fungicide plot, a 6x6" open ended aluminum barrier box was centered on packets to be treated and hammered into the soil to isolate treated are from surrounding soil. To further isolate treated area, a 6" steel plate set on its edge was hammered in the ground between aluminum box and untreated packets. For untreated location applied same volume of water as a control (Table 2).

Figure 4: Representative images of dormant seed (left column) and germinated seed (remaining columns) for A) *E.wallacei,* B) *E.mohavense,* and C) *B.tournefortii.* Germinated seed was sorted into three possible categories; empty hull, hull with remnant embryo tissue visible, or stalk / radicle present. Photographs were taken from a Nikon SMZ800 microscope.

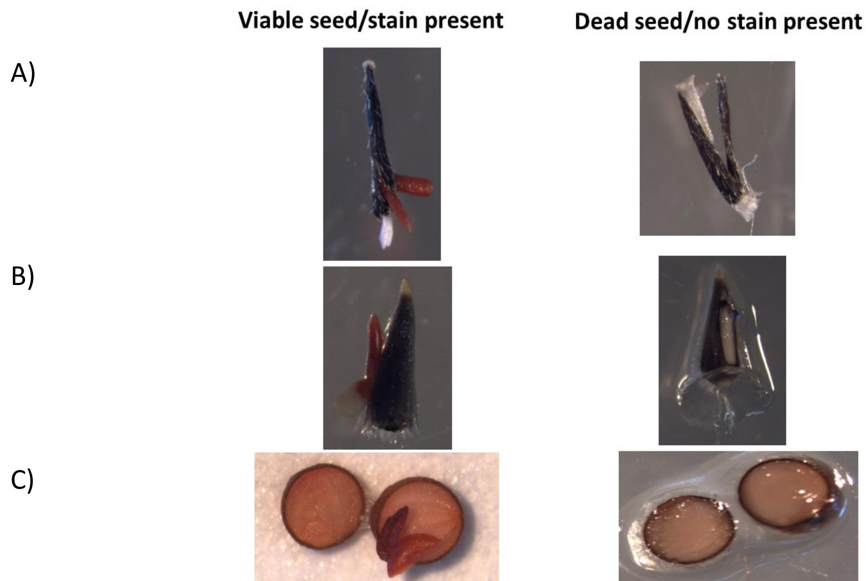

Figure 5: Representative images of living seed (left column) and dead seed (right column) from TZ staining assays are shown for A) *E.wallacei,* B) *E.mohavense,* and C) *B.tournefortii.* Photographs were taken with Infinity Analyze software from a Nikon SMZ800 microscope.

Table 1: Seed allocation to artificial seed banks for each experiment

Experiment	Species	Seed crop year	Total number of seeds	Number of seeds per artificial seed bank	*Number of artificial seed banks
Microhabitat-only	*E.wallacei*	2016	120	4	60
	E.mohavense	2016	540	18	60
	B.tournefortii	2016	540	18	60
Microhabitat-fungicide	*E.wallacei*	2015	840	14	60
	E.mohavense	2015	1080	18	60
	B.tournefortii	2016	1080	18	60

Table 2: Soil drench and water control treatments used in the microhabitat-fungicide experiment

Treatment	Volume (oz) per seed bank	Solution
Water control group	6	Water
Fungicide treatment group	6	Soil drench solution containing 4 drops Subdue MAXX and 2.5mL Cleary 3336F per gallon of water

Table 3: Results from Analysis of Variance for seed germination in each experiment

Species	Experiment	Factor	Df	F-value	p-value
E.wallacei	Microhabitat-only	Microhabitat	2, 27	0.481	0.623
	Microhabitat-fungicide	Fungicide	1, 54	0.002	0.960
		Microhabitat	2, 54	1.185	0.313
		MicrohabitatxFungicide	2, 54	1.357	0.266
E.mohavense	Microhabitat-only	Microhabitat	2, 26	0.301	0.742
	Microhabitat-fungicide	Fungicide	1, 51	0.064	0.800
		Microhabitat	2, 51	0.061	0.940
		MicrohabitatxFungicide	2, 51	0.032	0.967
B.tournefortii	Microhabitat-only	Microhabitat	2, 27	2.935	*0.070*
	Microhabitat-fungicide	Fungicide	1, 54	0.783	0.380
		Microhabitat	2, 54	1.142	0.326
		MicrohabitatxFungicide	2, 54	0.559	0.574

Italics indicate marginal significance p< 0.100.

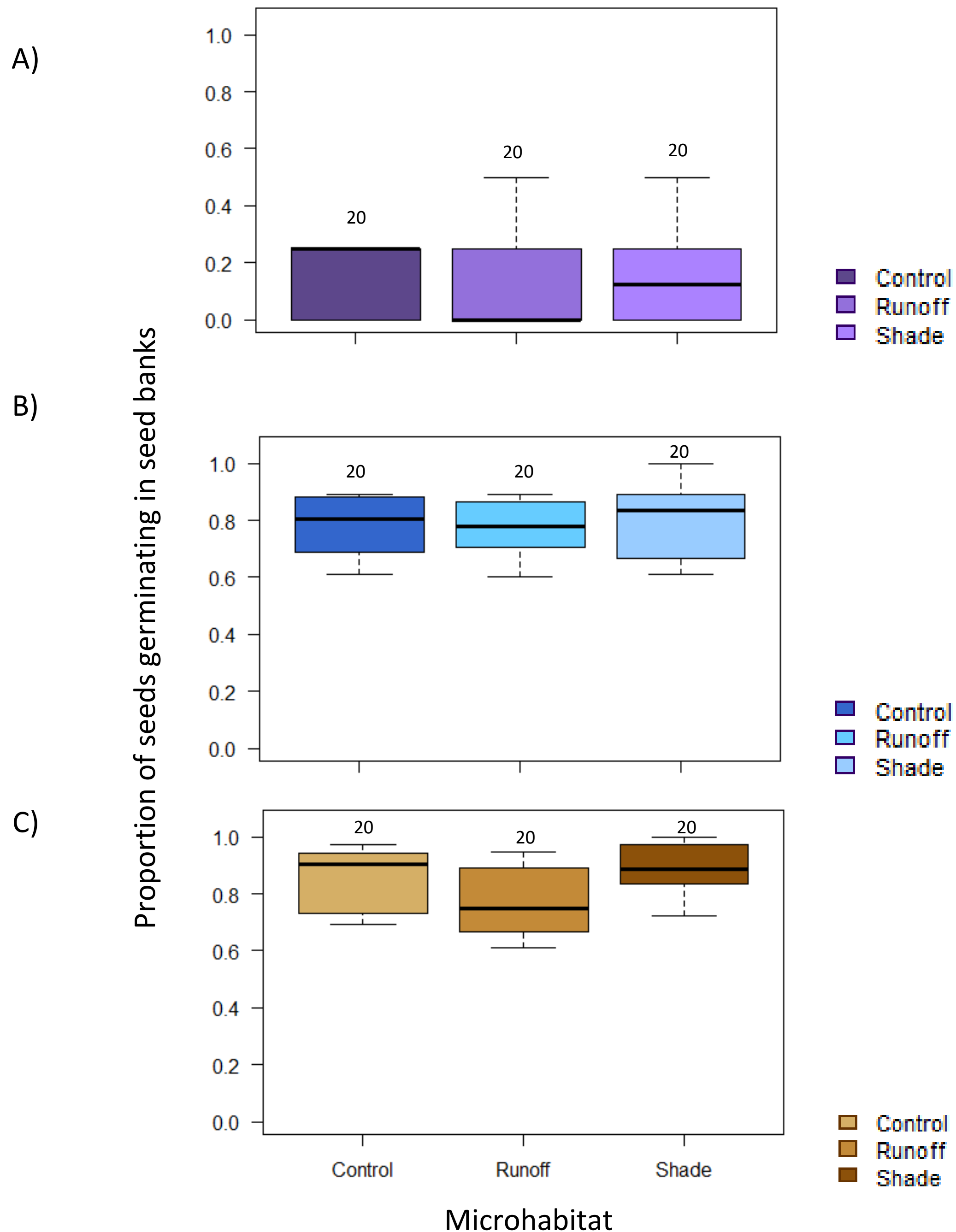

Figure 6: Germination rates from artificial seed banks in the microhabitat-only experiment for A) *E. wallacei,* B) *E.mohavense,* and C) *B.tournefortii.* Sample size, number of artificial seed banks used to estimate proportion values, is shown above each bar. Differences between sample sizes shown and the number of seeds originally allocated to seed bank packets (Table 1) result from seed loss through fabric tears in the field or during transport. Boxplots show upper and lower quantile, representing observations outside 9 – 91 percentile. Dark line represents the median. Whiskers represent standard deviation above and below mean. Data falling outside 9 – 91 percentile range are outliers.

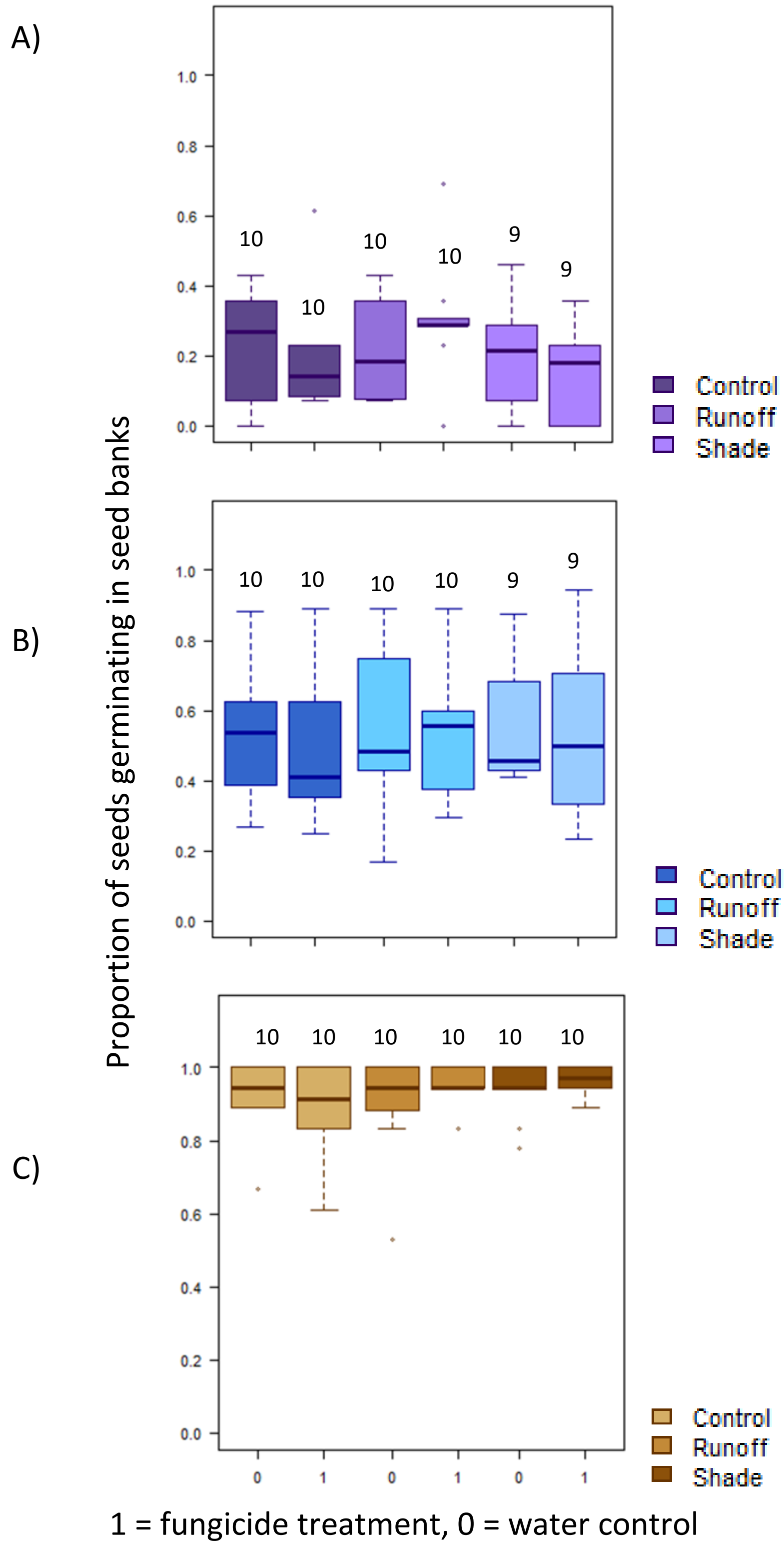

Figure 7: Germination rates from artificial seed banks in the microhabitat-fungicide experiment for A) *E. wallacei,* B) *E.mohavense,* and C) *B.tournefortii.* Seed banks were treated with either a one-time soil drench fungicide solution or a water control after burial. Sample size, number of artificial seed banks used to estimate proportion values, is shown above each bar. Differences between sample sizes shown and the

number of seeds originally allocated to seed bank packets (Table 1) result from seed loss through fabric tears in the field or during transport. Boxplots show upper and lower quantile, representing observations outside 9 – 91 percentile. Dark line represents the median. Whiskers represent standard deviation above and below mean. Data falling outside 9 – 91 percentile range are outliers.

Table 4: Results from Generalized Linear Model for seed survival and fungal growth in microhabitat-fungicide experiment

Species	Experiment	Factor	dF	X^2	p-value
E.wallacei	Seed Survival	Fungicide	1, 244	0.055	0.814
		Microhabitat	1, 244	1.432	0.231
		FungicidexMicrohabitat	1, 244	0.016	0.899
	Fungal Growth	Fungicide	1, 120	0.570	0.450
		Microhabitat	2, 120	1.580	0.453
		FungicidexMicrohabitat	2, 120	0.453	0.797
E.mohavense	Seed Survival	Fungicide	1, 188	3.111	0.777
		Microhabitat	1, 188	12.069	**0.000513**
		FungicidexMicrohabitat	1, 188	2.886	*0.089*
	Fungal Growth	Fungicide	1, 98	0.330	0.565
		Microhabitat	2, 98	4.684	*0.096*
		FungicidexMicrohabitat	2, 98	3.520	0.172
B.tournefortii	Seed Survival	Fungicide	1, 34	1	1
		Microhabitat	1, 34	1	1
		FungicidexMicrohabitat	1, 34	1	1
	Fungal Growth	Fungicide	1, 33	1.276	0.258
		Microhabitat	2, 33	0.134	0.935
		FungicidexMicrohabitat	2, 33	9.759	**0.007**

Seed survival results are derived from tetrazolium staining assays; fungal growth results come from plated seed assays. **Bold** values indicate significance, $p<0.050$. *Italics* indicate marginal significance, $p <0.100$

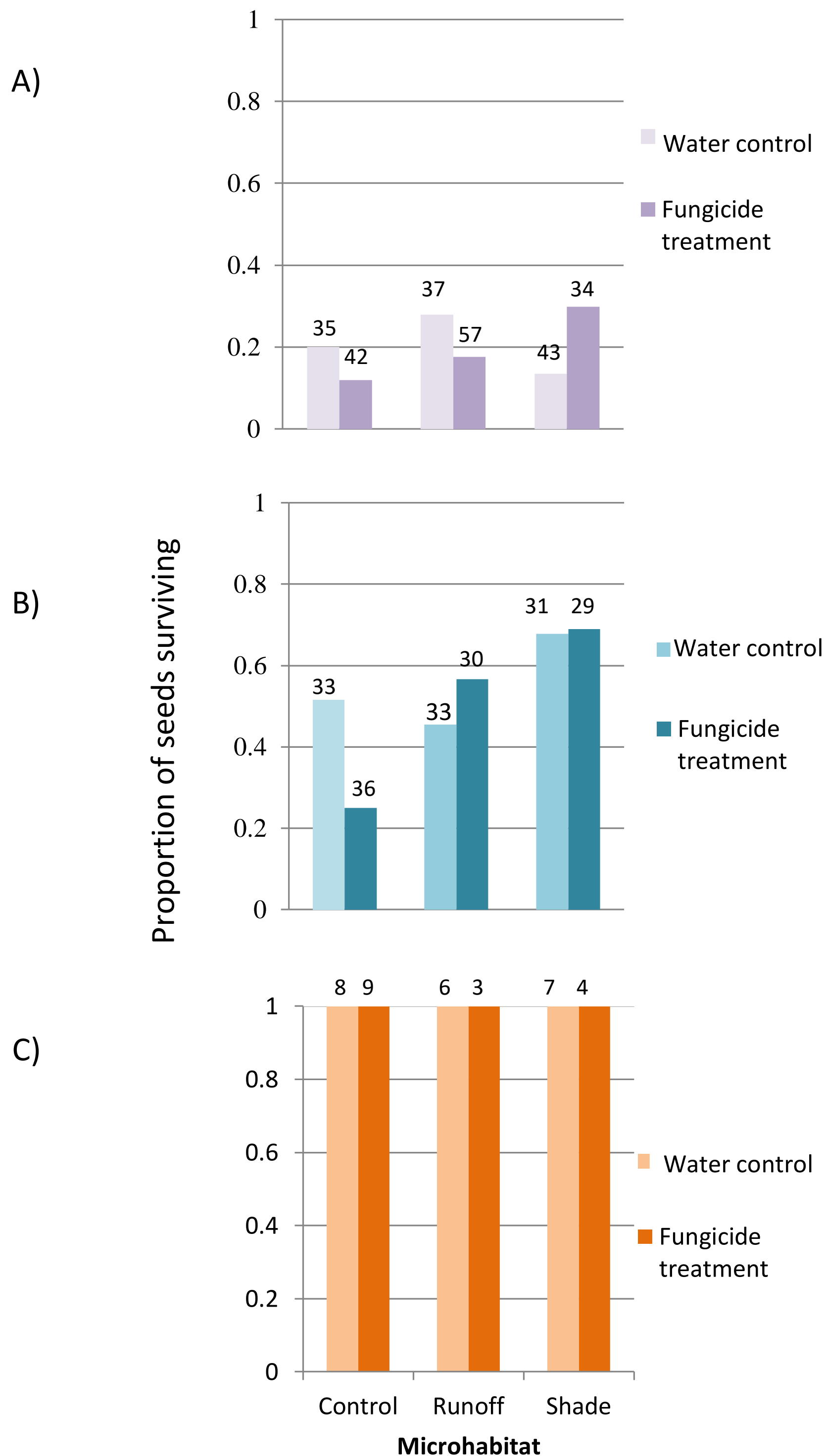

Figure 8: Proportion of dormant seeds surviving in artificial seed banks treated with fungicide or water control treatments in microhabitat-fungicide experiment for A) *E. wallacei,* B) *E.mohavense* , and C) *B.tourneforti.* Proportions shown represent the number of seeds staining red in TZ assays, for seed taken from seed banks in each microhabitat. Sample sizes above each bar indicate the number of dormant seeds allocated to this experiment. Sample sizes for *B.tournefortii* are low because nearly all seeds germinated in seed banks while they were deployed in the field (Figure 6), leaving few dormant seeds for experiments conducted in the lab.

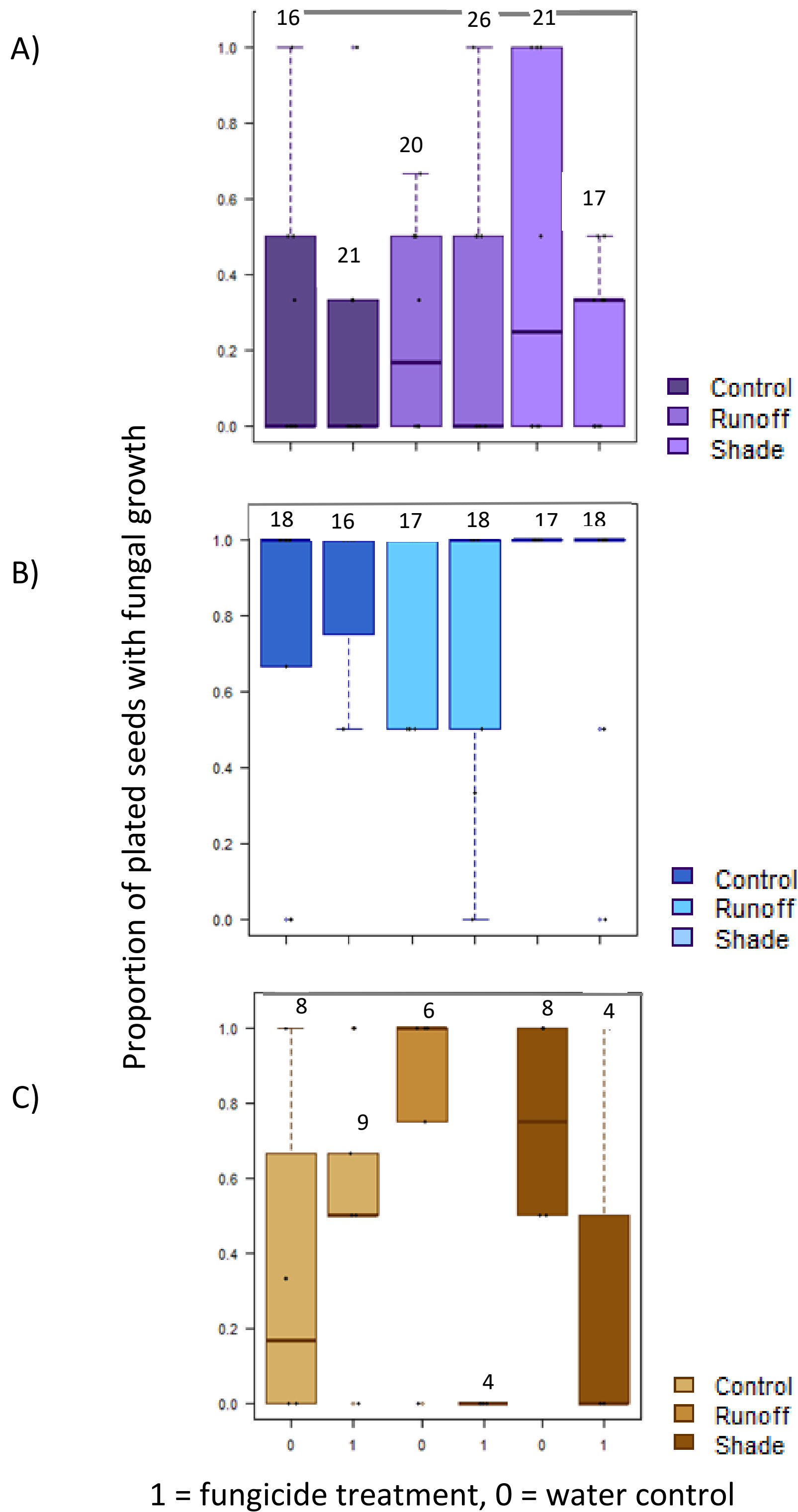

Figure 9: Proportion of plated dormant seeds showing fungal growth in the microhabitat-fungicide experiment for A) *E.wallacei,* B) *E.mohavense, and* C) *B.tournefortii.* Sample sizes above each bar indicate the number of dormant seeds allocated to this experiment. Sample sizes for *B.tournefortii* are low because nearly all seeds germinated in seed banks while they were deployed in the field (Figure 6),

leaving few dormant seeds for experiments conducted in the lab. Boxplots show upper and lower quantile, representing observations outside 9 – 91 percentile. Dark line represents the median. Whiskers represent standard deviation above and below mean. Data falling outside 9 – 91 percentile range are outliers.

References

Armstrong, A., Ole, N. J., & Whitaker, J. (2016). Solar park microclimate and vegetation management effects on grassland carbon cycling. *Environmental Research Letters*, *11*(7), 074016.

Ayerst, G. (1969). The effects of moisture and temperature on growth and spore germination in some fungi. *Journal of Stored Products Research*, *5*(2), 127-141.

Baldwin, Bruce G., and Douglas H. Goldman. *The Jepson Manual: Vascular Plants of California*. University of California Press, 2012.

Bangle, D. N., Walker, L. R., & Powell, E. A. (2008). Seed germination of the invasive plant Brassica tournefortii (Sahara mustard) in the Mojave Desert. *Western North American Naturalist*, *68*(3), 334-342.

Barrows, C. W., Allen, E. B., Brooks, M. L., & Allen, M. F. (2009). Effects of an invasive plant on a desert sand dune landscape. *Biological Invasions*, *11*(3), 673-686.

Berry, K. H., Gowan, T. A., Miller, D. M., & Brooks, M. L. (2014). Models of Invasion and Establishment for African Mustard (Brassica tournefortii). *Invasive Plant Science and Management*, *7*(4), 599-616.

Brooks, M. L., Draper, J. V., & Trader, M. R. (2006). Controlling Sahara mustard: evaluation of herbicide and mechanical treatments (California). *Restor Ecol*, *24*, 277-278.

Brown, J. S., & Venable, D. L. (1986). Evolutionary ecology of seed-bank annuals in temporally varying environments. *The American Naturalist*, *127*(1), 31-47.

Dahlin, K., König, E., Laubmeier, A., Wehn, A., & Rıos-Soto, K. (2012). Competition Model between the invasive Sahara Mustard and Native Plants in the Sonoran Desert.

Fox, John, and Sanford Weisberg. *An R Companion to Applied Regression*. 2nd ed., Sage, 2011.

Freas, K. E., & Kemp, P. R. (1983). Some relationships between environmental reliability and seed dormancy in desert annual plants. *The Journal of Ecology*, 211-217.

Gelbard, J. L., and J. Belnap. 2003. Roads as conduits for exotic plant invasions in a semiarid landscape. *Conservation Biology,* 17:420–432.

Hernandez, R. R., Easter, S. B., Murphy-Mariscal, M. L., Maestre, F. T., Tavassoli, M., Allen, E. B., ... & Allen, M. F. (2014). Environmental impacts of utility-scale solar energy. *Renewable and*

Sustainable Energy Reviews, *29*, 766-779.

ERT. 1988. E503 Luz SEGS VII Sensitive Plant Salvage Soil Survey Technical Report. Prepared for Luz Engineering Corporation, Los Angeles, California by James Nyenhuis, ERT, Fort Collins, Colorado.

Jeffrey E. Lovich, Joshua R. Ennen; Wildlife Conservation and Solar Energy Development in the Desert Southwest, United States, *BioScience*, Volume 61, Issue 12, 1 December 2011, Pages 982–992, https://doi.org/10.1525/bio.2011.61.12.8.

Juhren, M., Went, F. W., & Phillips, E. (1956). Ecology of desert plants. IV. Combined field and laboratory work on germination of annuals in the Joshua Tree National Monument, California. *Ecology*, *37*(2), 318-330.

Kawecki TJ, Ebert D. Conceptual issues in local adaptation. *Ecology Letters.* 2004;7:1225–1241.

Levine JM, Adler PB, Yelenik SG. A meta-analysis of biotic resistance to exotic plant invasions. *Ecology Letters*. 2004;7:975–989.

Minnich, R. A. and A. C. Sanders. 2000. *Brassica tournefortii*. In Bossard, C. C., J. M. Randall, and M. C.Hoshovsky, editors. editors. Invasive plants of California's wildlands. University of California Press. Berkeley

Mordecai, E. A. (2012). Soil moisture and fungi affect seed survival in California grassland annual plants. Plos One, 7(6), e39083.

Pake, C. E., & Venable, D. L. (1996). Seed banks in desert annuals: implications for persistence and coexistence in variable environments. *Ecology*, 77(5), 1427-1435.

Philippi, T. (1993). Bet-hedging germination of desert annuals: beyond the first year. *The American Naturalist*, *142*(3), 474-487.

Pimentel, D., Rodrigues, G., Wang, T., Abrams, R., Goldberg, K., Staecker, H., . . . Boerke, S. (1994). Renewable Energy: Economic and Environmental Issues. *BioScience, 44*(8), 536-547. doi:10.2307/1312281

Porter, R., Durrell, M. and Romm, H. (1947). The use of 2,3,5-triphenyl-tetrazoliumchloride as a measure of seed germinability. *Plant Physiol* 22(2): 149.

Randall, J. M., Parker, S. S., Moore, J., Cohen, B., Crane, L., Christian, B., …& Morrison, S. (2010).

Mojave Desert Ecoregional Assessment. *The Nature Conservancy*.

Schafer, M., & Kotanen, P. M. (2003). The influence of soil moisture on losses of buried seeds to fungi. *Acta Oecologica*, *24*(5-6), 255-263.

Venable, D. L. (2007). Bet hedging in a guild of desert annuals. *Ecology*, *88*(5), 1086-1090.

Zak, J. C., Sinsabaugh, R., & MacKay, W. P. (1995). Windows of opportunity in desert ecosystems: their implications to fungal community development. *Canadian Journal of Botany*, *73*(S1), 1407-1414