Assessing chemical management options for the control of stinknet (*Oncosiphon piluliferum)*

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Summary:

- In Riverside county, controlling the spread of Stinknet (*Oncosiphon piluliferum)* is a pressing management objective.
- Pre-emergent herbicides were the most effective at reducing stinknet cover down to or close to zero percent cover, one year after treatment.
- Post-emergent herbicide strategy was effective at reducing stinknet cover during the drier year in 2018, but not as effective during the wetter 2019 year.
- Selecting the appropriate herbicide strategy should be weighed amongst the multiple management objectives and constraints for a site.

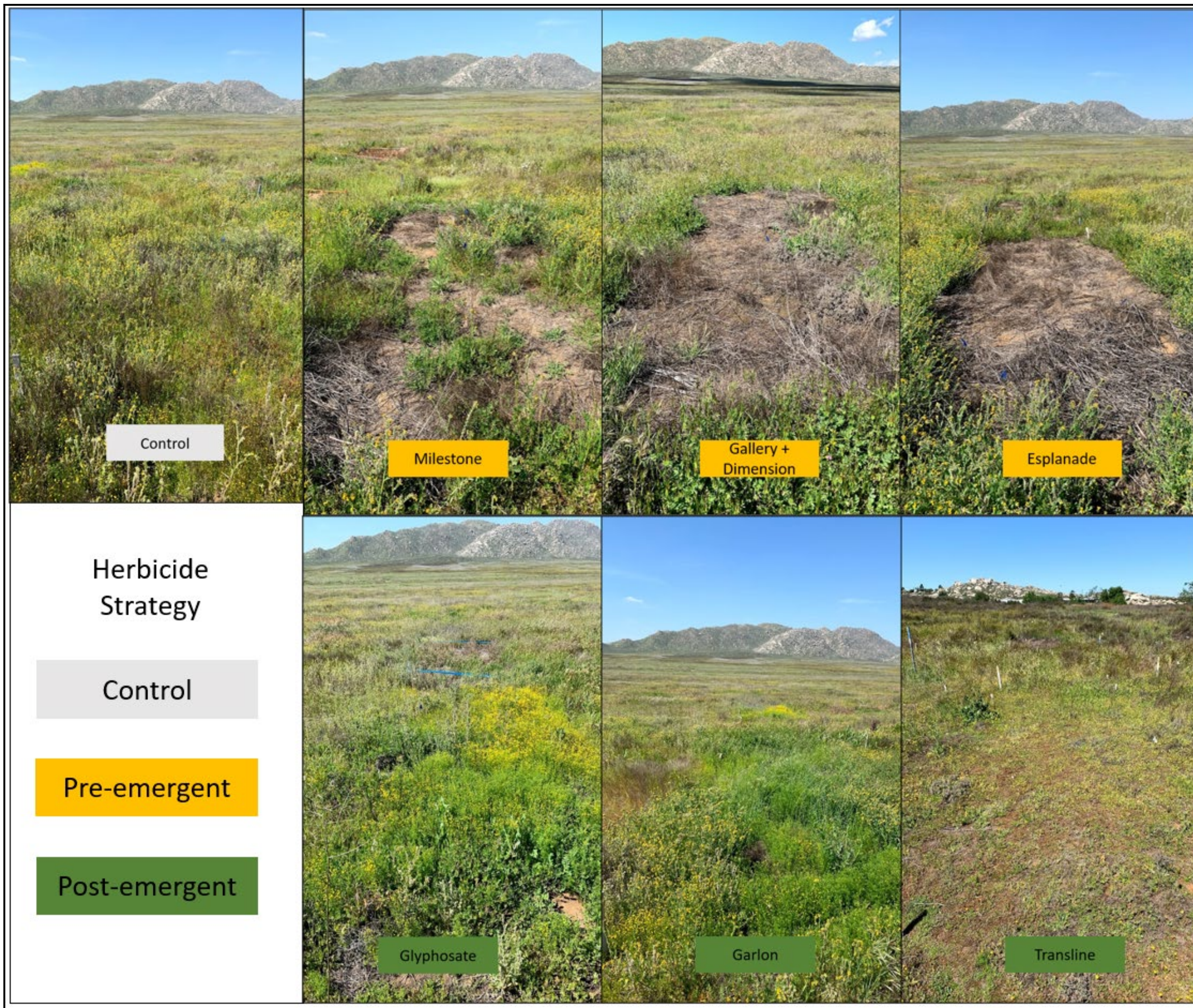

Figure 1. Experimental plots pictured one year after treatment, color coded by herbicide strategy. Note that photos were selected to depict average responses and come from different sites and years.

Project Overview. Stinknet (*Oncosiphon piluliferum*) is a winter annual forb native to South Africa that has recently become an invader of concern in North America as its distribution continues to expand rapidly. Stinknet produces many small seeds per individual and creates dense stands, altering community composition. In Riverside County, CA, Stinknet is threatening critical habitat for the endangered Stephens Kangaroo Rat (SKR, *Dipodomys stephens*), which has led land managers to utilize various methods of control for the invader, with reports of mixed results. This may be due to differences in the timing of application, or in differences of the phenological stage of the invader during application, making it difficult to make recommendations based on current reported management efforts. To help identify effective herbicide treatments, we initiated field trials aimed at:

(1) evaluating the efficacy of reducing initial establishment with pre-emergent herbicides applied in the fall vs reducing seed production with post-emergent herbicides applied in the spring,

(2) identifying within a given strategy: post- or pre-emergent, which herbicide treatment is most effective in reducing stinknet cover, and

(3) assessing herbicide impacts on non-target species and overall community composition.

This report covers the results of herbicide applications one year after treatment across our experimental sites and within each site. We will report in the future on the results of Stinknet cover two years after treatment as well as provide a comparison between a single and two years of application.

Study Design. We conducted field herbicide trials at three grassland sites within Riverside County: Lake Mathews Preserve, Lake Perris State Recreation Area and Motte Rimrock Reserve in 2018 and 2019. Using a randomized block with split-plot design, we set up three blocks at each site, with 16- 2m x 5m herbicide treatment plots arranged per block (Fig. 1). One of 6 treatments were randomly assigned with a paired control for each set of pre-emergent (aminopyralid, indaziflam, isoxaben+dithiopyr), and post-emergent (glyphosate, clopyralid, triclopyr) plots. We had three pre-emergent and three post-emergent plots per block, each with a paired control (n=8), three blocks per site, for a total of 72 plots per herbicide trial year (8 herbicide x 3 blocks x 3 sites), replicated over two years for a total of 144 plots (Fig. 1).

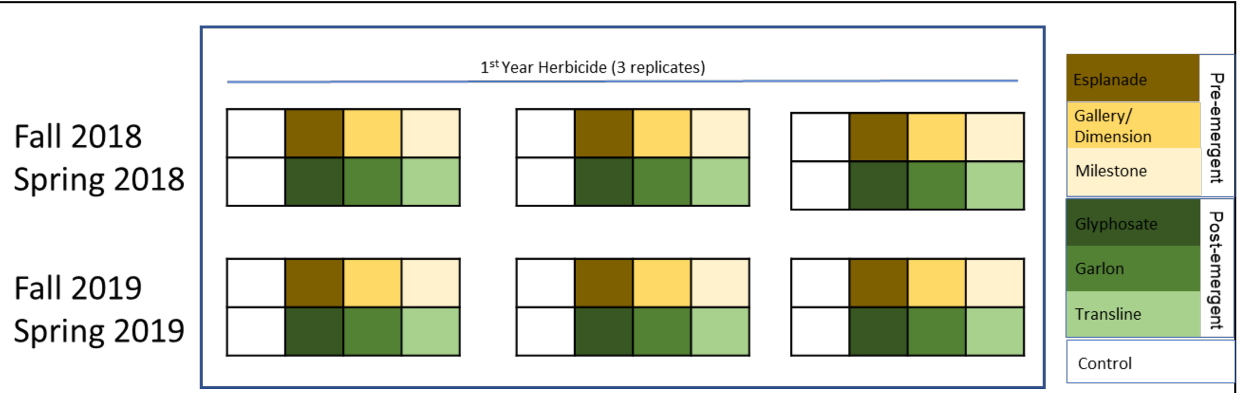

Figure 2. Study design layout for herbicide application, replicated at three sites: Lake Matthews, Lake Perris Recreation Area, Motte Rimrock Reserve.

Herbicides were applied using an 80-inch spray boom for one continuous spray (see Table 1 for concentrations used). Each plot was sprayed in the fall (November) if it was assigned a pre-emergent herbicide or in the spring (March/April) if it was assigned a post-emergent herbicide. Timing of spring application was dependent on the phenology of stinknet, as we sprayed when stinknet was starting to bud, and 5% of the population was flowering. The exact dates of application were (April 9, 2018-post; Nov 8-9, 2018-pre; Mar 19, 2019-post; Nov 15, 2019-pre).

Table 1. List of herbicides and concentrations used in experimental trials. No adjuvants were used with the herbicides.

Herbicide Strategy	Herbicide Name	Active Ingredient	Concentration	Herbicide applied per plot
Pre-Emergent	Milestone®	Triisopropanol-ammonium salt/ aminopyralid	7 oz/ac (2 lb/g a.e. formulation)*	0.52 ml/plot
Pre-Emergent	Esplanade®	indaziflam	7 oz/ac (1.67 lb/g a.i. formulation)**	0.52 ml/plot
Pre-Emergent	Gallery® + Dimension®	Isoxaben + dithiopyr	16-31 oz/ac (4.16 lb/g a.i. formulation)/ 2 qt/ac (1 lb/g a.i. formulation)	2.3 ml Garlon® + 2.4 ml Dimension®/plot
Post-Emergent	Roundup®	glyphosate	4 qt/ac (4 lb/g a.e. formulation)	9.5 ml/plot
Post-Emergent	Transline®	clopyralid	⅔ pt/ac (3 lb/g a.e. formulation)	0.80ml/plot
Post-Emergent	Garlon®	triclopyr amine or ester (ester preferred, but depends on wetland status)	8 qt/ac (3 lb/g a.e. formulation) or 1-4 qt/ac (4 lb/g a.e. formulation)	19ml/plot

* "a.e." = acid equivalent; ** "a.i." = active ingredient

Plant Sampling. To assess how the herbicides impacted the plant community, we made visual estimates of aboveground plant cover for each species within a 1 m² subplot, as well as total litter cover, and total bare ground cover. We took measurements in the spring one year after treatment (YAT). We classified litter as any dead vegetative growth from the previous year and classified bare ground as the total amount of the plot covered by barren soil. Species that were present as single individuals were recorded as 0.25 % cover. Because of the overlapping canopy cover of plant species, total cover in a subplot could be over 100 %. Species that were not present in the meter squared plot but were present within the larger 2 x 5 m experimental plot were recorded for richness measures.

Figure 3. Picture of 1m^2 plant sampling area within experimental plot.

Analyses. To determine the general effects of herbicides on stinknet control one year after treatment, we first analyzed stinknet cover as a function of *herbicide strategy/treatment (control, pre, post)* and *year treated* across sites. We then analyzed these factors per site to account for site level differences. To assess herbicide impacts on plant community composition, we relativized our species composition data into 4 cover category types: litter, bare, native, and non-native. We ran a multivariate analysis (PERMANOVA) that allowed us to evaluate simultaneously the impact of herbicides on the different plant cover classes. This would allow us to evaluate whether non-native or native species were differentially responding to the herbicide treatments.

Results Across Sites.

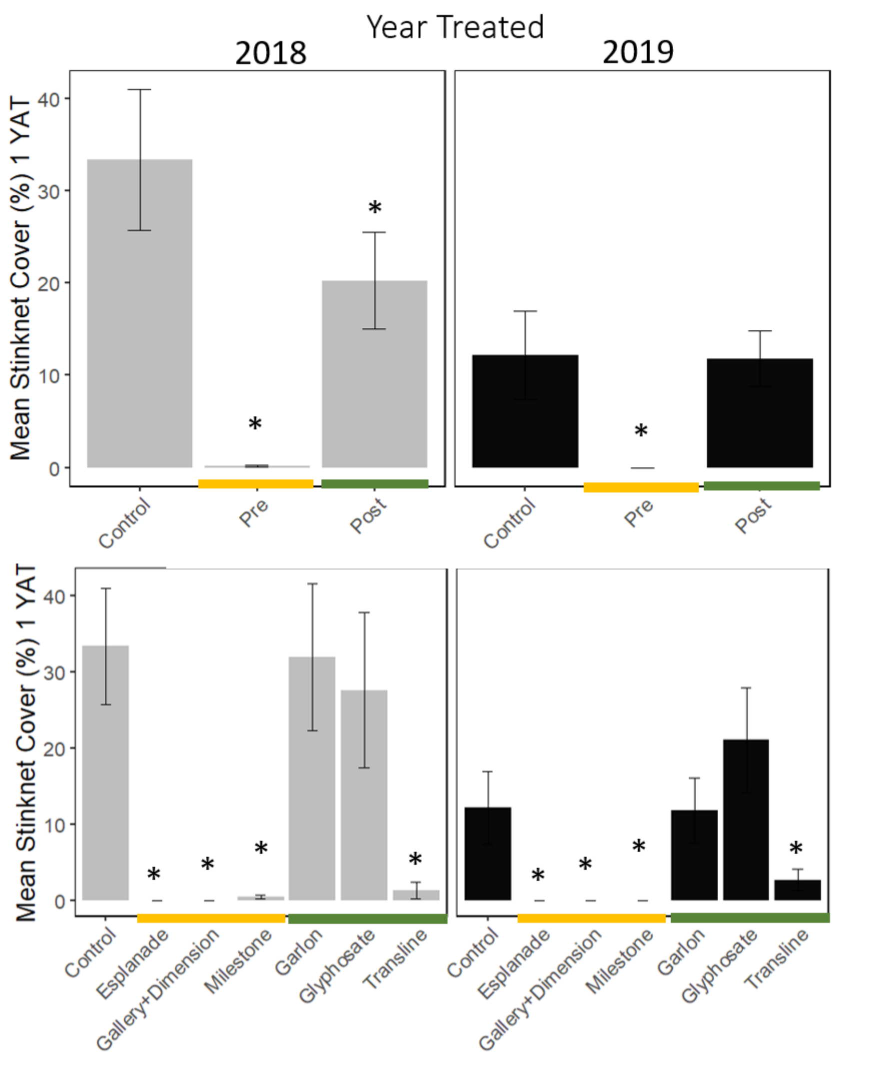

Figure 4. Mean stinknet percent cover one year after treatment plotted by herbicide treatment. Pre-emergents are designated by the orange bar, while post-emergents are designated by the green bar. Shading indicates year treated, gray = 2018, black = 2019. Error bars are 1 standard error. (*) indicates statistical difference from control p<0.001.

Evaluating the efficacy of herbicide strategies: We found that the overall herbicide strategy (pre- vs post-emergent) impacted stinknet cover. We also found that plant cover varied by year, as 2019 had significantly lower stinknet cover overall compared to 2018. Pre-emergent herbicides consistently reduced the amount of stinknet cover in treatment plots during both trial years, while post-emergent herbicides performed well in 2018, but their effect on controlling stinknet was reduced significantly in 2019 (Fig. 4).

Identifying effective herbicide treatments: The pre-emergent treatments (Esplanade®, Gallery®+Dimension®, Milestone®) were not significantly different from each other, as they were all effective at reducing stinknet cover one year after treatment during both trial years. As for the post-emergent herbicides (Garlon®, Glyphosate, Transline®), we found Transline® to be the only post-emergent treatment to significantly reduce stinknet cover one year after treatment. However, Transline® was less effective at controlling stinknet cover in the 2019 treated plots compared to the 2018 treated plots. The variability in treatment effectiveness may be due to the difference in precipitation received during each trial year. In 2019, Riverside received 14.5 inches of total annual rainfall (Jan-Dec), compared to 2018 which received 7.2 inches of total annual rainfall (*Current Results*, 2020). The additional soil moisture in 2019 may have accelerated the breakdown of the herbicide, thus reducing the effectiveness to control stinknet.

Assessing plant community responses: Plant community composition one year after treatment was significantly impacted by herbicide strategy and year treated (Fig. 5). Community composition for the post-emergent herbicide strategy did not differ from control plots, as these treatments were primarily dominated by non-native and native cover (i.e., green, and gray symbols overlapping non-native and native arrows in Fig 5). However, community composition in pre-emergent treated plots did differ from post-emergent and control communities, in that they contained more bare and/or litter cover, depending on the trial year (Fig. 5).

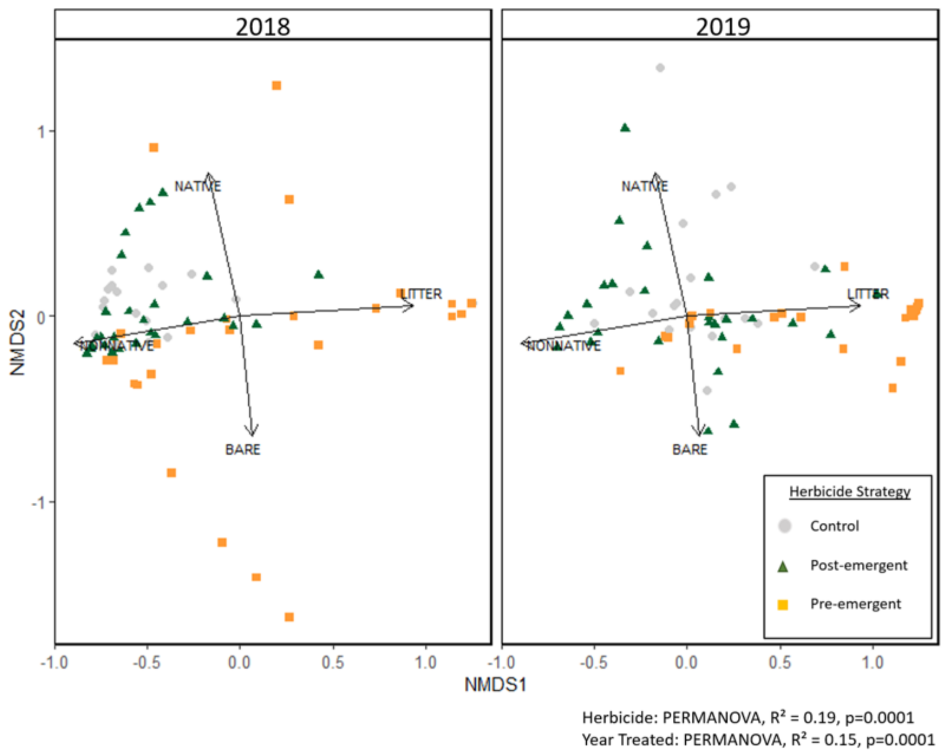

Figure 5. Changes in community composition within NMDS ordination space by herbicide strategy and year treated. Each point represents a plot that was treated, and the arrows represent the average magnitude and direction of community composition shift.

As for overall species richness, pre-emergent herbicide strategy resulted in significantly lower species richness compared to control and post-emergent treated plots (Fig. 6). Although Milestone®, a pre-emergent herbicide, had higher species richness than the other two pre-emergent treatments, it is important to note that these plots had higher non-native species present. Similarly, post-emergent treated plots did contain comparatively higher species richness, however most of the species were non-native.

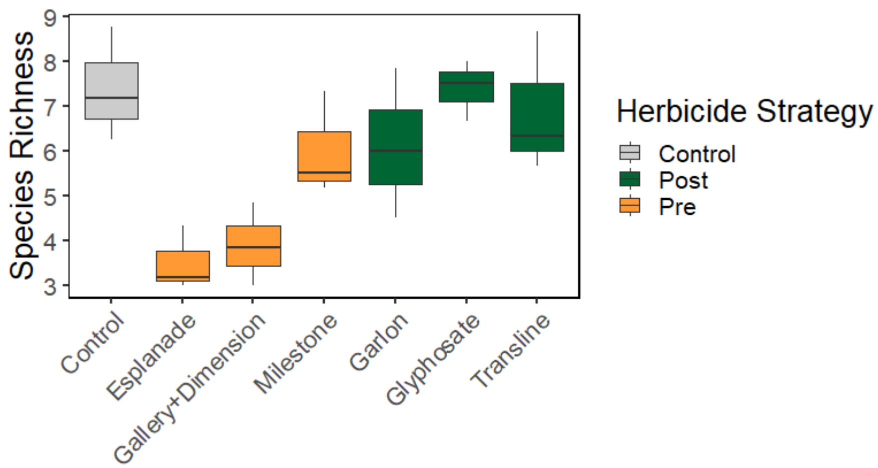

Figure 6. Boxplots showing the mean number of species richness on the y-axis, plotted by herbicide treatment on the x-axis. Treatments are coded by herbicide strategy.

Post-Emergent Herbicide Germination Trial seed collection: To assess the viability of stinknet seeds in plots sprayed with post-emergent herbicides, 5-10 stinknet seed heads were collected in late Spring of each herbicide trial year from the post-emergent treated plots and their paired control plots. Thus far, we have run one germination trial, which consists of 20 randomly selected seeds from the collected seed heads that are then placed into petri dishes and grown in growth chambers for four weeks. We are planning to replicate the trials to assess the viability of stinknet seeds from plots treated with post-emergent herbicides.

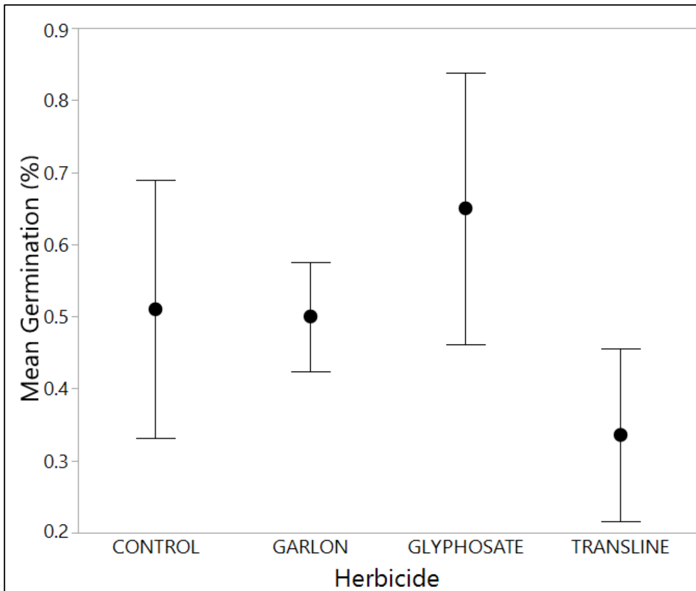

Figure 7. Preliminary results for post-herbicide germination trials showing the average percentage of seeds that germinated by herbicide treatment (20 seeds per plate, N=24 plates). Error bars are 1 standard error.

Based on our initial trial of comparing the viability of post-emergent treated stinknet seeds, we found that Transline® had the lowest germination percentage (33.5%), compared to the rest of the treatments which had over 50% germination (Fig. 7). This preliminary data may provide additional support for our findings, that Transline® is the most effective post-emergent herbicide treatment in controlling stinknet cover. Further, the germination data may explain why certain Glyphosate treated plots resulted in higher stinknet cover compared to control plots in our field trials as well.

Implications for management.

Pre-emergent herbicides: Our results suggest that pre-emergent herbicides are an effective way to reduce the establishment of stinknet one year after treatment. Even though all three pre-emergent herbicides were effective at reducing stinknet cover essentially to zero percent, their effects on community composition did vary slightly. Generally, the pre-emergent strategy resulted in increased bare or litter cover. Milestone®, which has pre- and post-emergent applications, had the highest vegetation cover of the pre-emergent treatments; however, most of the vegetation was non-native species. These results suggest that when using this strategy, practitioners should take into consideration other factors (e.g., timing of application, weather conditions, local vegetation) that might impact the overall recovery of the plant community in the treated area. For instance, it is important to consider the invasion history of the site to anticipate what species are stored in the local seedbank and will likely re-establish post treatment. In many of Riverside County's forbland communities, the native seedbank is limited or depleted (Allen et al 2005, Cox and Allen 2008). For this reason, if the management goal were to reduce stinknet cover and increase native plant cover, a pre-emergent herbicide would likely need to be paired with a follow up treatment (e.g., seeding) to promote native plant establishment.

Post-emergent herbicides: Our results suggest that post-emergent herbicides can be an effective strategy to control stinknet cover; however, the effectiveness can be more variable than pre-emergent herbicides. Transline® was the only post-emergent treatment to consistently reduce stinknet cover one year after treatment during both trials. This may be due to the small residual pre-emergent effects that Transline® can have after being sprayed as a post-emergent herbicide. Although Garlon® and Glyphosate have been shown to be effective at reducing stinknet cover during short term (0-3 months, unpublished data from C. McDonald), our germination and herbicide trial results suggest that these herbicides may not be the most effective treatments to control stinknet cover. Therefore, we recommend Transline® as the post-emergent treatment of choice. Equally important to consider when deciding which herbicide to use, is the consideration of application timing and weather conditions. Post-emergent herbicides are found to be most effective when applied on actively growing individuals, before the budding or flowering stage. Further, soil moisture is important to ensure the efficacy of post-emergent herbicides. For instance, soil that contains excess moisture can promote movement or degradation of the herbicides in the soil, resulting in reduced ability of the herbicide to control stinknet.

Future Work: This study analyzed the effects on stinknet control, one year after treatment of one seasonal application of herbicide. In the future, we plan to continue our investigation of identifying effective management techniques for stinknet control by also analyzing the effects of multiple years of herbicide application. We will also continue conducting germination trials for each herbicide trial year to assess the impacts of herbicide on stinknet seed viability.

As with any herbicide, it is important to always read the label and follow directions to avoid contamination or other environmental or health risks. For more information, please see the links below which contains additional product information.

Esplanade®	https://www.environmentalscience.bayer.us/-/media/prfunitedstates/documents/resource-library/product-labels/esplanade-f.ashx
Gallery®	https://www.corteva.us/products-and-solutions/turf-and-ornamental/gallery.html

Dimension® https://www.corteva.us/products-and-solutions/turf-and-ornamental/dimension.html
Transline® https://www.corteva.us/products-and-solutions/land-management/transline.html
Glyphosate https://natseed.com/pdf/Roundup%20Pro%20Label.pdf
Garlon® https://www.corteva.us/products-and-solutions/land-management/garlon-4-ultra.html

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1. E. B. Allen, R. D. Cox, T. Tennant, S. N. Kee, D. H. Deutschman, Landscape restoration in southern California forblands: Response of abandoned farmland to invasive annual grass control. *Isr. J. Plant Sci.* **53**, 237–245 (2005).

2. R. D. Cox, E. B. Allen, Composition of soil seed banks in southern California coastal sage scrub and adjacent exotic grassland. *Plant Ecol.* (2008).

3. Riverside - Recent Annual Temperatures and Rainfall.” *Current Results*, www.currentresults.com/Yearly-Weather/USA/CA/Riverside/recent-annual-riverside-temperature-precipitation.php.

Appendix. Results by Site.
Lake Matthews

The average stinknet cover across all the control plots at Lake Matthews was 1.66% in 2018, and 0.166% in 2019. This site had the least stinknet cover overall but was still consistent with the trend that 2019 had less stinknet cover compared to 2018.

Evaluating the efficacy of herbicide strategies: It is important to highlight that at this site, the max average stinknet cover was 3% one year after treatment. Post-and pre-emergent herbicide strategies were not significantly different from each other in 2018 or 2019. Although the difference between strategies or treatments was not significant, plots sprayed with pre-emergent herbicides consistently had lower stinknet cover compared to post-emergent and control plots (Fig. 8).

Identifying effective herbicide treatments: Similar to the overall trend across sites, the pre-emergent herbicides (Esplanade®, Gallery®+Dimension®, Milestone®) did not differ from each other as they were all able to control stinknet cover less than the control plots, down close to 0%. As for the post-emergent herbicides, plots treated with Glyphosate contained higher stinknet cover than the control plots in both 2018 and 2019. While Garlon® and Transline® treated plots reduced stinknet cover lower than the control plots in both 2018 and 2019.

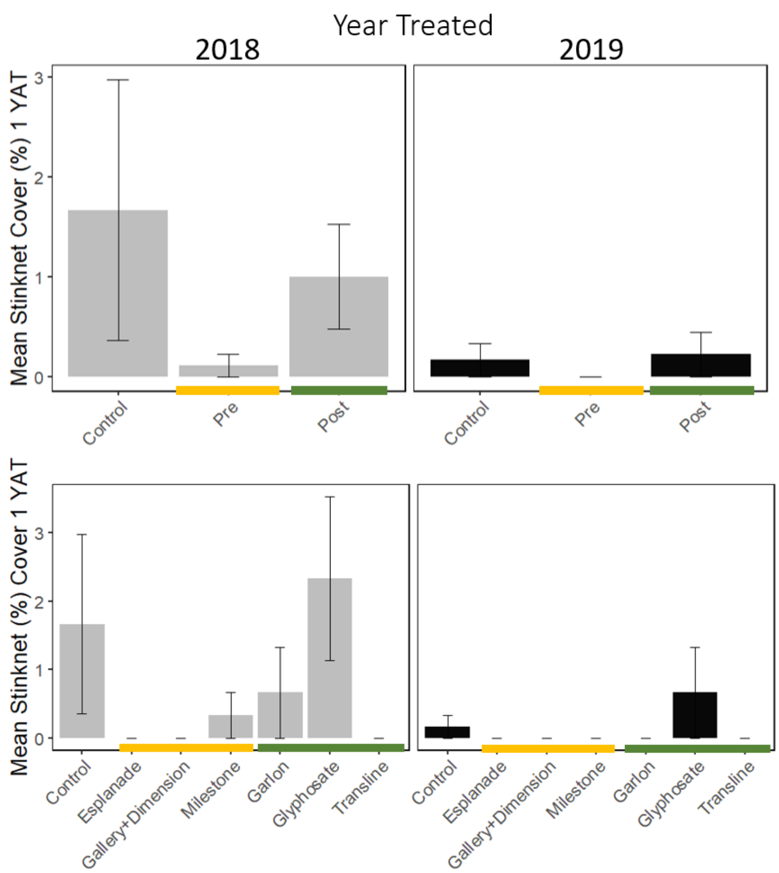

Figure 8. Mean stinknet percent cover at Lake Matthews one year after treatment, plotted by herbicide treatment. Pre-emergents are designated by the orange bar, while post-emergents are designated by the green bar. Shading indicates year treated. Error bars are 1 standard error.

Lake Perris

The average stinknet cover across all the control plots at Lake Perris was 30.83% in 2018, and 7.166% in 2019. This site had the second highest stinknet cover across the sites.

Evaluating the efficacy of herbicide strategies: At this site, the pre-emergent herbicide strategy consistently reduced stinknet cover to <1% in 2018 and 2019, while the efficacy of the post-emergent herbicide strategy varied by year. The post-emergent herbicides were able to reduce stinknet cover in 2018; however, in 2019, the strategy as a whole had higher stinknet cover compared to the control plots one year after treatment. This could be due to either a) high germination of an existing stinknet seed bank, b) the reduced efficacy of the herbicide as it could breakdown as a result of the higher soil moisture, or c) a combination of these factors.

Identifying effective herbicide treatments: At this site, Transline® was the only post-emergent to significantly reduce stinknet cover compared to control plots one year after treatment. Glyphosate and Garlon® treated plots had similar or slightly higher stinknet percent cover compared to the control plots (Fig. 9). As for the pre-emergent treatments, Esplanade®, Milestone® and Gallery®+Dimension® were all effective during both trial years, resulting in <1% stinknet cover.

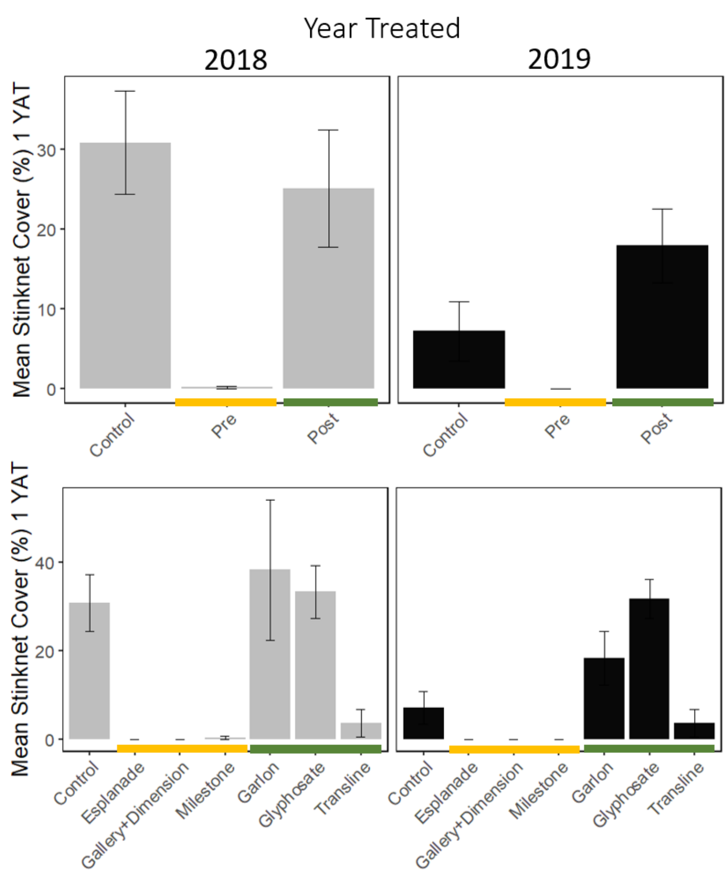

Figure 9. Mean stinknet percent cover at Lake Perris one year after treatment, plotted by herbicide treatment. Pre-emergents are designated by the orange bar, while post-emergents are designated by the green bar. Shading indicates year treated. Error bars are 1 standard error.

Motte Rimrock Reserve

The average stinknet cover across all the control plots at the Motte Rimrock Reserve was 67.5% in 2018, and 29.166% in 2019. This site had the highest stinknet cover of all of the sites.

Evaluating the efficacy of herbicide strategies: At this site, both pre-and post-emergent strategies resulted in reduced stinknet cover compared to the control. However, plots that were treated with pre-emergent herbicides consistently resulted in less stinknet cover compared to post-emergent treated plots in 2018 (pre: 0.22%, post: 34.66%) and 2019 (pre: 0.66%, post: 17.3%).

Identifying effective herbicide treatments: At Motte Rimrock Reserve, we observed similar trends to Lake Matthews, where Garlon® and Transline® reduced stinknet cover lower than the control, while glyphosate treated plots contained similar or higher cover of stinknet compared to the control (Fig. 10).

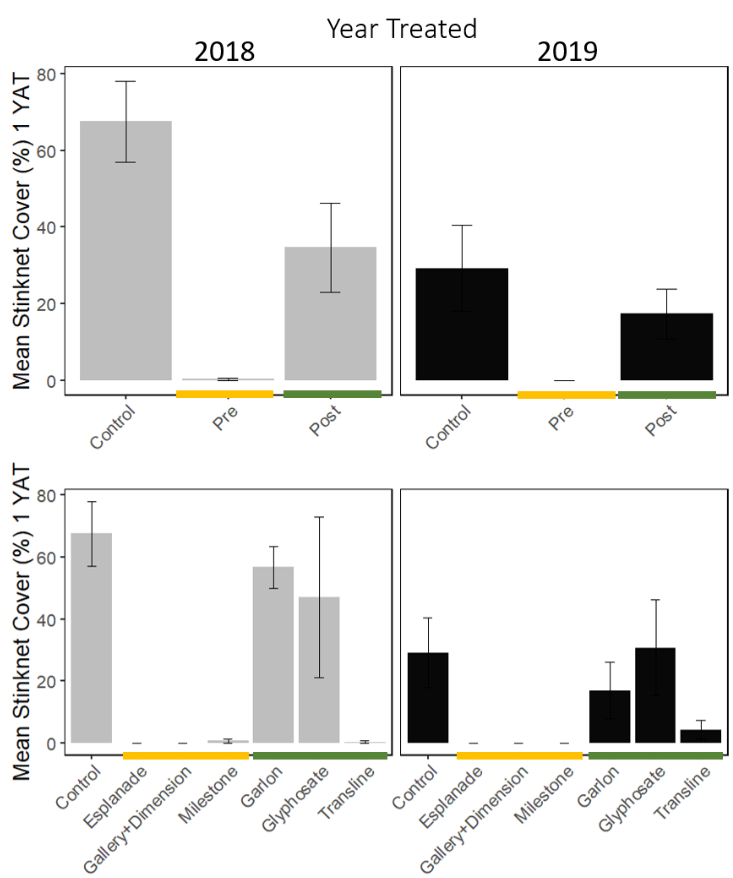

Figure 10. Mean stinknet percent cover at Motte Rimrock Reserve, one year after treatment, plotted by herbicide treatment. Pre-emergents are designated by the orange bar, while post-emergents are designated by the green bar. Shading indicates year treated. Error bars are 1 standard error.