Integrated Management Strategies for Perennial Pepperweed

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Perennial pepperweed (Lepidium latifolium) has become one of California’s worst non-crop weed problems (see figure). It invades a wide range of environments including coastal areas, beaches, tidal shores, inland marshes, riparian areas, wetlands, grasslands, and roadways. Following disturbance, perennial pepperweed rapidly forms monoculture stands that displace desirable vegetation and prevent new vegetation establishment.

Currently, herbicides are the most common method for managing perennial pepperweed. Herbicides provide effective control, but they require repeat applications for several years, since perennial pepperweed often re-sprouts from its extensive root system and new seedlings emerge in bare-ground areas. Slow re-establishment of desirable vegetation also promotes perennial pepperweed re-invasion. In an effort to improve long-term control of perennial pepperweed, an experiment was started at two sites in Lassen County California in Fall 2002. The experiment examined management strategies that control perennial pepperweed and re-establish desirable vegetation. Study sites were heavily infested with perennial pepperweed with less than 5% competing vegetation cover.

Treatments evaluated the usefulness of site preparation methods such as winter burning, spring mowing, winter grazing, or fall disking for removing accumulated thatch to facilitate herbicide application and re-seeding. Treatments also examined the influence that site preparation methods had on chlorsulfuron (Telar), 2,4-D ester, or glyphosate (Roundup) efficacy applied at the flower-bud stage. Finally, the study determined if integrating herbicides and site preparation methods influenced native grass reseeding success and long-term perennial pepperweed control.
Figure 1: The Effect of Control Methods on Perennial Pepperweed Cover
June 2006 (4 years after treatment initiation)

Figure 1: All herbicides reduced perennial pepperweed cover compared to untreated plots one and two years after treatment, but some herbicide + site preparation combinations provided better control than others. Averaged across sites, chlorosulfuron, 2,4-D, or glyphosate applied alone, and chlorosulfuron, 2,4-D, or glyphosate in combination with burning, mowing, or grazing provided the best perennial pepperweed control. Fall disking before herbicide application often decreased perennial pepperweed control compared to using herbicides alone.

Figure 2: The Influence of Site Preparation Treatments and Herbicides on Perennial Grass Establishment in June 2006 (15 months after 2nd seeding)

Figure 2. Winter burning in combination with yearly 2,4-D or glyphosate applications, spring mowing in combination with yearly 2,4-D or glyphosate applications, and disking in combination with yearly 2,4-D or glyphosate applications provided the best site preparation for reseeding and resulted in the highest native grass establishment. Applying 2,4-D in early summer the year of re-seeding also improved grass establishment by suppressing perennial pepperweed re-sprouts and annual broadleaf weeds during seeding grass growth. Chlorosulfuron caused chlorosis and stunting to western wheatgrass, basin wildrye, and beardless wildrye seedlings when applied before seeding, but in other trials, chlorosulfuron was safe on these native grasses when applied after the 4 leaf stage. No herbicide offered 100% control after two years of treatment suggesting multiple follow-up herbicide applications are needed for long-term weed suppression and vegetation restoration.
Ryegrass (*Lolium* spp.) is a winter annual, common throughout California (Fig. 3). In 1998, two orchard sites were identified as possibly having glyphosate resistant ryegrass populations. These populations were confirmed as being resistant to glyphosate (Simarmata et al., 2003).

At least one orchard in the San Joaquin Valley was also reported to contain glyphosate resistant ryegrass (R. Vargas, personal communication). The species of ryegrass, although reported to be rigid ryegrass (*Lolium rigidum*), appears to be *L. multiflorum*, or possibly a hybrid between *L. rigidum* and *L. multiflorum*.

Although it has only been confirmed in two or three orchards, many other non-confirmed reports have indicated that glyphosate resistant ryegrass may be more common in California than was originally reported. If resistance to glyphosate is confirmed, then alternative weed management programs are needed.

The objective of this study was to screen ryegrass populations collected throughout California for resistance to glyphosate and to determine the level of resistance among populations.

**MATERIALS and METHODS**

**1. Seed collection**

Seed from mature annual ryegrass was collected in May, July and August, 2004, from roadside and agricultural fields and orchards throughout the Sacramento and San Joaquin Valleys.

Counties sampled and the number of sites per county were: Butte 4, Colusa 9, Fresno 1, Glenn 8, Madera 2, Merced 5, Monterey 1, Sacramento 1, San Joaquin 6, Solano 5, Stanislaus 3, Sutter 2, Tehama 2, Yolo 11.

A description of each collection site was noted, including collection date, and the GPS coordinates. At each site, seed from at least 15 plants were collected and combined into a single sample.

Attempts were made to collect at least 1,000 mature seed for each sample site. A total of 60 sites were sampled in 2004. Seed was cleaned and dry seed stored at 70°F until use.

![Figure 3. Resistant ryegrass in almonds.](image)

**2. Preliminary sensitivity experiment**

Seed from a known susceptible ryegrass population were planted into pots and grown in the greenhouse. Pots were placed in a temperature controlled greenhouse, 75°F day and 55°F night, with supplemental lighting (12 hr day length). Plants were thinned to 2 to 3 plants per pot at one week after emergence and allowed to grow until 4-6 inches in height. Once plants reached the desired height, they were treated with 8 glyphosate rates: 0.0 0.031, 0.062, 0.125, 0.25, 0.5, 1.0 and 2.0 lbs acid equivalent per acre (ae)/ac.

Four replicate pots per treatment were used. Glyphosate applications were made using a track sprayer, with final spray volume of 16 gal/ac. Distilled water was used for all applications, in order to avoid confounding effects of hard water.

Twenty one to 25 days after treatment, plants were harvested, and fresh weight determined. Percent control was calculated as the fresh weight of the treated plant.
divided by the fresh weight of the untreated plant. The rate that resulted in a 50% reduction in fresh weight (I₅₀) was calculated.

3. Screening experiment

Seed from each individual ryegrass population was planted into 12-inch x 18-inch flats to provide at least 20 plants in each flat. Application of glyphosate was made to each flat at 0.062 lbs ae/ac, the I₅₀ rate determined in the preliminary sensitivity experiment.

Plants were grown in the same greenhouse, under similar conditions used in the preliminary sensitivity experiment. Ryegrass was cut at ground level and shoot fresh weight was measured for each individual flat at 21 days after treatment. Samples were dried (120°F), then weighed. The distribution of responses within and between populations was compared.

4. Rate response experiment

In addition to comparing all populations at a low application rate, we also wanted to determine the rate of glyphosate necessary to kill all ryegrass from a population.

Following cutting for biomass measurement, ryegrass was allowed to regrow. Once plants reached 6 inches in height, they were once again treated with glyphosate, but at double the previous rate – 0.125 lbs ae/ac. At 21 days after treatment, plants were again cut at ground level for biomass measurement as described above.

This was repeated with glyphosate rate doubling [0.25, 0.75 (triple rate), 1.5, 3.1, 6.1 and 12.2 lbs ae/ac] at each successive treatment until all plants in a flat were killed.

RESULTS and DISCUSSION

Seeds were collected by driving until a mature ryegrass was seen, stopping and collecting a sample. Thus, many samples were collected on the roadside, but in all cases adjacent to agricultural fields. Ryegrass was common along roadsides and fields in the Sacramento Valley. While driving in the San Joaquin Valley, ryegrass was less common on roadsides and fields, particularly further south in the Valley.

Treating the 60 ryegrass populations with glyphosate at 0.062 lbs ae/ac resulted in minimal growth reduction in all populations (data not shown). The susceptible population was a population collected about 20 years ago from a location with no known glyphosate use. Thus, all the ryegrass populations appeared resistant relative to the susceptible ryegrass.

Table 1. Glyphosate rate required to kill ryegrass samples and the number of samples killed at that rate.

<table>
<thead>
<tr>
<th>Glyphosate rate</th>
<th>Number of samples</th>
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<tbody>
<tr>
<td>Lbs ae/ac</td>
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<tr>
<td>0.75</td>
<td>5</td>
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<td>1.5</td>
<td>3</td>
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<tr>
<td>3.1</td>
<td>29</td>
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<tr>
<td>6.1</td>
<td>12</td>
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<td>12.2</td>
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Of the 60 populations evaluated, only five populations were killed by the recommended glyphosate rate of 0.75 lb ae/ac (Table 1). At 1.5 lbs ae/ac less than 15% of the samples, 8 total, were killed. The observation that many populations are resistant to glyphosate is not surprising, since seed samples collected from areas where it is likely they would have been treated with glyphosate, in many cases repeatedly, and thus only resistant plants would remain. Eleven populations have not been completely killed by over 12 lbs of glyphosate per acre, and a few populations show less than 50% control from the 12.2 lb ae/ac rate, indicating a very high level of resistance.

The ryegrass populations in the northern counties, near the locations of the original observations of resistance, required a high rate of glyphosate for control. It is likely that most ryegrass populations in California contain some glyphosate resistant individuals and that once susceptible individuals are removed by treatment with glyphosate, resistant individuals breed with other resistant individuals, creating a highly resistant population.

The abundance of glyphosate resistant ryegrass along roadsides has likely allowed the rapid spread throughout the state, as mud picked up in tires could carry the seed. The high level of glyphosate resistance on roadside ryegrass may be a reflection of the repeated use of glyphosate by road crews, particularly in programs where residual herbicides are not used.

The data on glyphosate resistant ryegrass distribution and rate sensitivity will serve as a baseline, allowing other populations to be tested and compared in the future. Now that glyphosate resistant ryegrass has been confirmed in many areas of California, alternative management options will need to be implemented.
AVOIDING WEED SHIFTS AND RESISTANCE IN ROUNDUP READY ALFALFA

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Weeds are a challenge to profitable alfalfa production. The Roundup Ready (RR) alfalfa production system has potential to simplify weed management, and improve control of both annual and perennial weeds. Roundup has proven to be a reliable herbicide treatment in other transgenic crops and has improved weed management in the short term. However, weed species shifts and the selection for Roundup-resistant weeds have resulted from the increased use of this technology.

Alfalfa is especially vulnerable for several reasons: tillage is not practical, alfalfa is produced in large fields with a great diversity of weeds, and there is significant potential for long-term repeated use of a single herbicide because it is a perennial crop. Alfalfa growers can learn from the experience gained with other Roundup Ready crops to minimize weed shifts and the evolution of resistant weeds. Weed control systems that integrate tillage between plantings, crop rotations, rotations with herbicides of different modes of action (preferably soil residual herbicides), and tank mixtures are important preventative measures.

The long-term effectiveness and sustainability of the Roundup Ready system in alfalfa depends upon how well growers adopt the concept of herbicide rotation into their production systems. Use a preemptive approach—one should not wait until weed shifts and resistance occur before utilizing herbicide rotation strategies.

DEVELOPMENT OF RR ALFALFA

Alfalfa, is an important crop throughout the Western US. Because nearly all weeds reduce the palatability and nutritional value of alfalfa, livestock industries—especially the dairy and horse industry—expect nearly weed-free hay. This can be difficult to achieve using conventional alfalfa herbicides. Typically, no single herbicide controls all weeds present in a field and some weeds, especially perennial weeds, are not adequately controlled with any of the current conventional herbicides. In addition to being difficult to achieve, complete weed control in alfalfa is costly. Growers continually seek ways to enhance the level of weed control while minimizing costs.

Glyphosate (Roundup)-resistant or Roundup Ready alfalfa (RR alfalfa), developed in late 1997 and made commercially available in fall of 2005, makes broadcast applications of Roundup to alfalfa possible (fig. 4).

The biotechnology-derived plants have an altered enzyme that allows them to tolerate a Roundup application while susceptible weeds are killed. This technology allows growers to deal with some of the most difficult-to-control weed species.

Figure 4. Roundup ready alfalfa plots showing the safety on transformed varieties and the sensitivity of old alfalfa varieties.

Beginning in 2001, trials conducted by UC Farm Advisors throughout California demonstrated the effectiveness of this technology. Roundup was especially effective for seeded alfalfa, as less alfalfa injury resulted and superior weed control was realized compared to standard herbicides. One great advantage of this technology is that it suppresses perennial weeds such as dandelion, nutsedge, Bermudagrass, and quackgrass that have not been adequately controlled with conventional practices. Initial experience in commercial fields has confirmed these research findings and has further demonstrated the benefits of this technology—increased ease of use and superior weed control.

WEED SHIFTS & RESISTANCE WITH RR ALFALFA

The greatest concern with this new weed-management system is the potential for weed shifts and weed resistance. A weed shift occurs when weeds easily controlled by Roundup (such as chickweed) decline and difficult-to-control weeds (such as burning nettle) increase. Weed resistance is different from a weed shift. An example of weed resistance is ryegrass which is normally controlled by Roundup. If ryegrass develops resistance to Roundup then it can no longer be controlled by this herbicide. It is important to understand the distinction between weed shift and weed resistance.
because the significance, as well as the management approaches for dealing with each, is different.

The possibility of weed shifts and weed resistance is particularly a concern with RR alfalfa, the first perennial crop with this transgenic trait, due to a long alfalfa stand life and the potential for repeated use of a single herbicide over several years. For some growers alfalfa is the primary crop, and they want to minimize the amount of time the field is planted to low-profit rotational crops. Therefore, there may be a continuous alfalfa crop in a field for 14 or more years with only one year of a small grain rotation crop between alfalfa plantings. Stand life is shorter in warmer production areas like the Central Valley of California (3-4 year stand life is typical), and there are more rotation crops. However, cotton and corn are commonly rotated with alfalfa, and if transgenic RR varieties are produced, this again could result in a prolonged time period where a single herbicide is used repeatedly.

There are aspects of the alfalfa production system that both favor and discourage the development of weed shifts and resistant weeds. On one hand, rotation opportunities in a perennial crop like alfalfa are less compared with annual cropping systems. Resistance and weed shifts are believed to evolve more rapidly in crops like alfalfa that are solid seeded, relatively low value, and grown on large acreages. Mechanical cultivation and hand weeding are not practical in a solid-seeded low-value crop like alfalfa. On the other hand, alfalfa is an aggressive competitor with most weeds, and many weed species do not tolerate frequent cutting.

Weed shifts or resistant weeds are unavoidable and will eventually occur with any herbicide after repeated use. Fortunately, resistance to Roundup is not as common as resistance to many other herbicides. Two weed species have documented resistance to Roundup in California—rigid ryegrass and marestail. Roundup-resistant ryegrass was first found in California orchards where there was a long history of continual Roundup use. Roundup resistant marestail originated in the southern San Joaquin Valley of California in orchards, vineyards, and ditch banks where tillage was no longer used and Roundup was used continuously for several years, oftentimes with multiple applications per season.

Roundup, is the most effective broad-spectrum post-emergence herbicide available, and it would be a shame to lose its effectiveness as a result of mismanagement. Weed shifts and/or weed resistance have occurred with the other transgenic RR crops released before RR alfalfa. This has typically occurred after approximately 6 years of continual use. Alfalfa growers can learn lessons gained from experience with these other crops as a preemptive measure to avoid, or at least minimize problems with weed shifts and weed resistance. The obvious questions are: a) What management practices can be utilized to avoid weed shifts and weed resistance, and b) How do we effectively utilize this new technology?”

RECOMMENDED WEED MANAGEMENT PROGRAM FOR RR ALFALFA

Roundup Ready alfalfa is still a relatively new technology, so we have limited field experience to date. The following are some suggestions to consider based upon proven resistance management strategies, our understanding of alfalfa production practices, and our initial experience with RR alfalfa. Ultimately, growers and pest control advisors hold the key to avoiding weed shifts and resistance by reducing selection pressure.

Controlling weeds when alfalfa is in the seedling stage is the most challenging, as alfalfa is most vulnerable to weed competition during this stage. Additionally, complete weed control in seedling alfalfa is often difficult to achieve and frequently requires tank mixes of different herbicides to control the broad spectrum of weeds found in an individual field. Crop injury and yield loss are usually far greater in seedling than in established alfalfa. Numerous field trials throughout the country have proven the effectiveness of Roundup in herbicide-resistant transgenic alfalfa.

Therefore, it is only logical to use Roundup for weed control in transgenic seedling alfalfa for the cost savings, improved weed control, reduced crop injury, and to eliminate the alfalfa seedlings that do not carry the Roundup resistance gene. Ordinarily, the 1.0 pound active ingredient rate (22 oz of Roundup UltraMax) is sufficient. However, a higher rate may be needed if the field contains some of the tolerant weeds such as burning nettle or little mallow. A tank mix may be advised if especially tolerant weeds are present. For example, a tank mix with Raptor or Pursuit may be advised if burning nettle is present, or a tank mix with Prism may be necessary if the field contains ryegrass.

Alfalfa stand-life varies considerably throughout the West depending on the production area, grower practice, and the existence of profitable rotation crops. However, a stand-life of 3 to 4 years is common in the Central Valley of California. For the principles outlined above, it is unwise to rely solely on Roundup applications for weed control throughout the life of a transgenic alfalfa field. This practice would encourage weed shifts and resistance, and in most cases weed control would diminish over time. Once weeds become resistant, the usefulness of an herbicide is
greatly diminished, and once a resistant weed population has gained a foothold, it is practically impossible to eliminate. Most alfalfa producers treat alfalfa during the dormant season for winter annual weeds that infest the first cutting. It is strongly recommended that growers not rely solely on Roundup for their winter weed control program for the duration of the stand. They should rotate to another herbicide or tank mix at least once in the middle of the life of a stand, and perhaps twice, if the stand-life is over 5 years.

Fortunately, there are several herbicides to choose from that have a different target site of action than Roundup. The soil-residual herbicides applied during the dormant season to established alfalfa [such as hexazinone (Velpar), diuron (Karmex) and metribuzin (Sencor)] would be appropriate herbicides for a rotation or tank-mix partner. The rotation herbicide or tank-mix partner of choice depends on the weeds present in the field and their relative susceptibility to the herbicides. Paraquat (Gramoxone) is another candidate for rotation or tank mixing; however, Gramoxone, like Roundup, is applied late in the dormant season. By rotating Gramoxone with Roundup, growers could potentially be selecting for early-emerging weeds that may be too large to control at the typical timing for Roundup or Gramoxone. Or, they could be selecting for late emerging weeds that germinate after the application.

Weed control in the last year of an alfalfa stand is often challenging because the stand is typically less dense and competitive, and there are also fewer herbicide options to choose from due to the plant-back restrictions for many of the soil-residual herbicides. Therefore, Roundup is a good choice for controlling weeds in the final year of an herbicide-resistant alfalfa field. This underscores the importance of having rotated herbicides before the final year so that Roundup will remain effective and control most weeds in a last-year alfalfa stand.

Summer annual grasses, such as yellow and green foxtail, barnyardgrass, cupgrass and jungle rice, and sometimes pigweed, can be problematic in established alfalfa. These weeds emerge over an extended time period whenever soil temperatures and moisture are adequate, typically from late winter or early spring—as early as February in the Central Valley—throughout the summer. They may emerge between alfalfa cuttings, so several applications may be necessary for a foliar herbicide like Roundup to provide season-long control. Multiple applications of a single herbicide during a season is cited as promoting weed resistance. Therefore, growers should not rely solely on Roundup for summer grass control for multiple seasons. It still remains to be seen how many applications of Roundup will be required for season-long summer grass control. In long growing-season areas, as many as two to three applications per season may be needed. Rather than making multiple applications of Roundup, a better approach may be to apply the pre-emergence herbicide trifluralin (Treflan), and follow-up with Roundup as needed for escapes. This approach helps avoid weed shifts and resistance, and may be more economical, as well, compared with multiple applications of Roundup.

Herbicide rotation and use of tank mixtures should be done for both dormant applications to control winter annual weeds and for spring/summer applications intended to control summer annual weeds. For example, rotating to Velpar for winter annual weed control for a year does nothing to prevent weed species shifts or the development of resistance in the summer annual weed spectrum. Herbicides for summer annual weed control should be rotated as well.

There is no definitive rule on how often herbicides should be rotated. The suggestion to rotate or tank mix at least once in the middle years of the life of a stand (or more often for long-lived alfalfa stands) is only a suggestion. The key point, which cannot be overemphasized, is the importance of diligent monitoring for weed escapes. Producers should stay alert to the development of weed species shifts and resistant weeds. If the relative frequency of occurrence of a weed species increases dramatically, chances are that it is tolerant to Roundup and immediately rotating herbicides or tank mixing is advised. If a few weeds survive among a weed species that is normally controlled easily with Roundup, it could be an indication of possible weed resistance, assuming misapplication and other factors can be eliminated as possible causes. In these situations, it is imperative to prevent reproduction of a potential resistant biotype. Treat weed escapes with an alternative herbicide or other effective control measure.

CONCLUSIONS

The Roundup Ready production system has potential to simplify weed management, while also improving the spectrum of weed control. However, growers should learn from the experience gained in other crops and stay alert to the development of weed shifts and resistant weeds. The key is for growers to reduce selection pressure—not to rely on repeated applications of Roundup year-after-year, application-after-application. Rotate crops, rotate herbicides and utilize tank mixes as needed, depending on the weed species and weed escapes present. A grower should not wait for there to be a problem before he employs these practices; a preemptive approach is strongly encouraged.