

Tolerance of Three Clovers (*Trifolium* spp.) to Common Herbicides

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Clover inclusion may increase the sustainability of certain low-maintenance turfgrasses. However, selective weed control within mixed turfgrass–clover swards proves problematic because of clover susceptibility to herbicides. Research was conducted to identify common turf herbicides that are tolerated by three *Trifolium* species, including white clover, ball clover, and small hop clover, within low-maintenance turfgrass. Leaf and flower density, as well as plant height, were measured 4 wk after treatment as indicators of clover response to 14 herbicides. The three *Trifolium* spp. were moderately tolerant of bentazon (< 35% decrease in leaf density, height, or flowering). Simazine was well tolerated by white clover (< 5% decrease in all response variables), yet moderate injury to ball clover and small hop clover was observed (> 32% decrease in leaf density and > 27% decrease in flower density). Pronamide was well tolerated by white and ball clovers, with no effect on measured response variables; however, pronamide decreased small hop clover height and flower density (38 and 42%, respectively). Imazethapyr and imazamox were moderately well tolerated by white clover and small hop clover (< 39% decrease by all response variables), yet ball clover may be more susceptible to these herbicides than was anticipated based on previously reported tolerance. The herbicides 2,4-DB, halosulfuron, and metribuzin were well tolerated by white clover, with no effect on measured response variables; however, results suggest ball and small hop clovers were less tolerant. Clopyralid, 2,4-D, glyphosate, imazaquin, metsulfuron-methyl, and nicosulfuron resulted in varying degrees of injury across clover species and response variables, but, in general, these herbicides may not be viable options when attempting to maintain any of the three clover species tested. Further research is needed to quantify long-term effects of herbicide application on sward composition and clover succession.

Nomenclature: 2,4-D; 2,4-DB; bentazon; clopyralid; glyphosate; halosulfuron; imazaquin; imazethapyr; imazamox; metribuzin; nicosulfuron; pronamide; simazine; metsulfuron-methyl; ball clover, *Trifolium nigrescens* Viv.; small hop clover, *Trifolium dubium* Sibth. TRFDU; white clover, *Trifolium repens* L. TRFRE.

Key words: Clover, legume inclusion, mixed-sward, turfgrass.

La inclusión de *Trifolium* podría incrementar la sostenibilidad de varios céspedes de bajo mantenimiento. Sin embargo, el control selectivo de malezas en mezclas de céspedes con *Trifolium* es problemático debido a la susceptibilidad de *Trifolium* a muchos herbicidas. Se realizó una investigación para identificar herbicidas comunes para céspedes que son tolerados por tres especies de *Trifolium*, incluyendo *Trifolium repens*, *Trifolium nigrescens*, y *Trifolium dubium*, en céspedes de bajo mantenimiento. La densidad de hojas y flores, al igual que la altura de planta, fueron medidas 4 semanas después del tratamiento, como indicadores de la respuesta de *Trifolium* a 14 herbicidas. Los tres *Trifolium* spp. fueron moderadamente tolerantes a bentazon (< 35% de disminución en densidad de hojas, altura, o floración). *T. repens* también toleró simazine (< 5% disminución en todas las variables de respuesta), aunque se observó un daño moderado en *T. nigrescens* y *T. dubium* (> 32% disminución en densidad de hojas y > 27% disminución en densidad de flores). Pronamide fue tolerado por *T. repens* y *T. nigrescens*, sin ningún efecto en las variables de respuesta medidas. Sin embargo, pronamide disminuyó la altura y densidad de flores de *T. dubium* (38 y 42%, respectivamente). Imazethapyr e imazamox fueron moderadamente tolerados por *T. repens* y *T. dubium* (< 39% disminución de todas las variables de respuesta), aunque *T. nigrescens* podría ser más susceptible a estos herbicidas que lo que se anticipó con base en reportes previos de tolerancia. Los herbicidas 2,4-DB, halosulfuron, y metribuzin fueron bien tolerados por *T. repens*, sin efectos en las variables de respuesta medidas. Sin embargo, los resultados sugieren que *T. nigrescens* y *T. dubium* fueron menos tolerantes. Clopyralid, 2,4-D, glyphosate, imazaquin, metsulfuron-methyl, y nicosulfuron resultaron en varios grados de daño entre las especies de *Trifolium* y las variables de respuesta, pero en general, estos herbicidas no

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serían opciones viables al tratar de mantener alguna de las especies de *Trifolium* evaluadas. Investigaciones adicionales son necesarias para cuantificar los efectos a largo plazo de la aplicación de herbicidas en la composición del césped y la sucesión de especies de *Trifolium*.

Clover (*Trifolium* spp.) inclusion is a proposed means of increasing the sustainability of certain low-maintenance turfgrasses (Dudeck and Peacock 1983; McCurdy et al. 2013, 2014; Munshaw et al. 2015; Sincik and Acikgoz 2007). Clovers provide ecosystem services, such as nitrogen fixation (Ledgard and Steele 1992; McCurdy et al. 2014; McNeill and Wood 1990; Whitehead 1995) and insect habitat (Abraham et al. 2010; Rogers and Potter 2004). White clover improves sward color by contributing N to associated grasses (Sincik and Acikgoz 2007) and is suitable for maintaining roadside slopes (Roberts and Bradshaw 1985). Yet, herbicide applications are often required to maximize yields of mixed grass–legume swards (DiTomaso 2000; Seefeldt et al. 2005) and are especially important during seedling establishment (Carlisle et al. 1980; Evers et al. 1993; Young et al. 1992).

Few research efforts have evaluated herbicide tolerance across a diverse range of clover species present within maintained turfgrass. Furthermore, knowledge of weed control within mixed turfgrass–clover scenarios has largely been compared with that of forages, in which relevant clover herbicide-tolerance studies are narrowly focused on white clover, subterranean clover (*Trifolium subterranean* L.), and red clover (*Trifolium pratense* L.). Small hop clover and ball clover are commonly found in transportation right-of-ways, waste areas, and low-maintenance commercial turf in the southeastern United States. Previous research by McCurdy et al. (2013) reported differential herbicide tolerance among four clover species, including red clover, small hop clover, crimson clover (*Trifolium incarnatum* L.), and ball clover. Those results suggested 2,4-DB, imazethapyr, and bentazon were potentially useful herbicides when clover is a desirable companion crop.

Given labeling adaptation that meets the needs of mixed turfgrasses, selective herbicides may improve establishment, maintenance, and persistence where mixed swards of these clovers and companion grasses are desired. Simultaneously, knowledge of herbicidal efficacy may lead to improved weed control where clover species are undesirable.

Our objective was to further identify common turfgrass- and legume-labeled herbicides that are tolerated by three clover species within low-maintenance turfgrass scenarios. Those species included small hop clover and ball clover, both annuals, as well as white clover, a perennial capable of spreading via seed and stolons. Furthermore, research was conducted to determine the effects of herbicides on clover phenology, such as height and flower and leaf density, which may have bearing on seed dispersal and clover persistence within turfgrass.

Materials and Methods

Study Sites. Research was conducted in Auburn, AL, on naturalized stands of three clover species, including white clover, ball clover, and small hop clover. Each location was a mixed sward of respective clover species and dormant bermudagrass [*Cynodon dactylon* (L.) Pers.] or bahiagrass (*Paspalum notatum* Fluegg) or both. Site selection was based on clover uniformity and homogenous sward composition of respective clovers. In total, 12 separate sites were chosen during a 2-yr period (four for each clover species). Each site contained only one dominant clover species. Annual clovers had emerged approximately 2 mo earlier and were beyond seedling stage but had not begun to flower. White clover locations were perennial stands that were in various phases of flowering. Locations were proximal to the Auburn University campus and were mixed-use areas, including roadside rights-of-way and temporary recreational vehicle parking areas. Soil at all locations was a Marvyn loamy sand (fine-loamy, kaolinitic, thermic Typic Kanhapludults). Soil pH ranged from 6.0 to 6.3 (1 : 1 soil : H₂O). Soil organic matter within the top 10 cm was < 1.5% at all sites. Locations were mown regularly at approximately a 13-cm height during the fall season preceding the study implementation. Sites remained unmown during the winter and while studies were being conducted in the spring. Supplementary fertilizer was withheld to ensure adequate clover coverage.

Table 1. Treatments were applied March 1, 2011, and March 2, 2012, via a CO₂-pressurized backpack sprayer within a water carrier volume of 280 L ha⁻¹, and all treatments included a 0.25% v/v nonionic surfactant.^a

Common name	Formulation, manufacturer	Experiment rate
		100 m ⁻²
Bentazon	Basagran, Arysta LifeScience, Cary NC	11.2 g ai
Metribuzin	Sencor, Bayer CropScience, Research Triangle Park, NC	4.2 g ai
Simazine	Princep, Syngenta Crop Protection, Greensboro, NC	11.2 g ai
2,4-D	Amine 400, PBI/Gordon, Kansas City, MO	15.6 g ae
2,4-DB	Butyrac 200, Albaugh, Ankeny, IA	15.4 g ae
Clopyralid	Lontrel Turf and Ornamental, Dow AgroSciences, Indianapolis, IN	4.2 g ai
Glyphosate	Roundup PRO, Monsanto Company, St Louis, MO	11.2 g ai
Imazethapyr	Pursuit, BASF Corporation, Research Triangle Park, NC	0.70 g ai
Imazamox	Raptor, BASF Corporation, Research Triangle Park, NC	0.52 g ai
Imazaquin	Image, BASF Corporation, Research Triangle Park, NC	5.6 g ai
Halosulfuron	SedgeHammer Gowan Company, Yuma, AZ	0.53 g ai
Metsulfuron-methyl	MSM Turf, FarmSaver, LLC, Raleigh, NC	0.21 g ai
Nicosulfuron	Accent, E. I. du Pont de Nemours and Company, Wilmington, DE	0.26 g ai
Pronamide	Kerb, Dow AgroSciences, Indianapolis, IN	2.8 g ai

^a Activator 90, Loveland Products, Inc., Greeley, CO.

Experimental Design and Treatments. Studies were randomized complete blocks (four replications) by design and were repeated at two sites per year. Experimental units were 1.0 m² in size. Herbicide treatments (Table 1) were applied March 1, 2011, and March 2, 2012, via a CO₂-pressurized backpack sprayer within a water carrier volume of 280 L ha⁻¹ using a handheld, four-nozzle boom (TeeJet TP8002VS nozzles on 23 cm spacing; Spraying Systems Company, Wheaton, IL). Treatments included a nontreated control and glyphosate as a treated control. Herbicide treatments included a 0.25% v/v nonionic surfactant (Activator 90, Loveland Products, Inc., Greeley, CO). Studies received natural rainfall within 48 h of treatment application (2.1 and 1.9 cm in 2011 and 2012, respectively).

Experimental units were randomly subsampled within three polyvinyl hoops (324 cm²) for flower and trifoliate leaf densities and clover-stand height, 4 wk after treatment (WAT), which roughly corresponded to flower senescence for small hop clover and ball clover. Height was determined from the soil surface to the topmost trifoliate leaf to discount differences in inflorescence stalk length.

Data Analysis. Data were normalized to the nontreated control, with respect to study location within year and were subject to ANOVA within the SAS software procedure GLIMMIX using generalized linear mixed-models methodology (version 9.4,

SAS Institute, Cary, NC). Treatment was considered a fixed effect in the model. Year, location (nested within year), and iterations containing these effects were considered random in the model (Carmer et al. 1989). Basic model assumptions were confirmed. Means were separated based on adjusted 95% confidence intervals, which allowed for multiple comparisons by protecting against familywise error rate (Littell et al. 2006).

Results and Discussion

For all clover species studied, ANOVA indicated that location, year, and treatment interactions were not significant ($P > 0.05$) for response variables. Therefore, experiments were pooled across locations and years with respect to species. Results are discussed for each clover species. Injury symptoms varied greatly because of herbicide mode of action and response timing; thus, it was difficult to visually distinguish between clover control and transient symptoms. Leaf density was reported instead of visually determined injury.

White Clover. Of the three clovers assessed within this study, white clover was generally least injured by all herbicides (Tables 2, 3, and 4). The ability to detect differences that were due to treatment was diminished by a high degree of variability. This is evidenced by rather large standard deviations and 95% confidence intervals.

Table 2. White clover response 4 wk after treatment to a mixed turf–clover sward, with treatments applied March 1, 2011, and March 2, 2012.

Common name	Trifoliolate leaves	± 95% CI ^a	Height	± 95% CI	Flowers	± 95% CI
Bentazon	14 abc	15	14 a	15	23 abc	35
Metribuzin	−22 cdef	15	2 ab	15	−30 bcd	35
Simazine	−5 abcd	15	9 a	15	23 abc	35
2,4-D	−33 defg	15	−39 cd	15	−63 cd	35
2,4-DB	16 ab	15	8 a	15	21 abc	35
Clopyralid	−81 h	15	−60 d	15	−50 cd	35
Glyphosate	−57 fgh	16	−48 cd	15	−71 d	35
Imazethapyr	10 abc	16	−12 abc	15	−38 bcd	35
Imazamox	7 abc	15	8 a	15	76 a	35
Imazaquin	−46 efgh	15	−46 cd	15	−50 cd	35
Halosulfuron	22 a	15	3 a	16	65 a	35
Metsulfuron-methyl	−64 gh	15	−53 d	15	−72 d	35
Nicosulfuron	−18 bcde	16	−35 bcd	15	−55 cd	35
Pronamide	31 a	15	23 a	15	50 ab	36
Nontreated	0 abcd	16	0 ab	16	0 bcd	35

^a Means were separated based on adjusted 95% confidence intervals (95% CI). Overlapping 95% CIs indicate a lack of significant difference.

^b Mean ± SD nontreated trifoliolate leaf density = 3,064 trifoliolate leaves m^{−2} (± SD 1,342); nontreated height = 12 cm (± SD 10); nontreated flower density = 142 inflorescences m^{−2} (± SD 187).

White clover leaf density mean ± SD measured 3,064 trifoliolate leaves m^{−2} (± SD. 1,342) across 2011 and 2012 nontreated plots. Imazaquin, glyphosate, metsulfuron-methyl, and clopyralid

reduced white clover leaf densities between 46 and 81% relative to the nontreated control (Table 2). Pronamide, halosulfuron, 2,4-DB, bentazon, imazethapyr, imazamox, simazine, nicosulfuron,

Table 3. Ball clover response 4 wk after treatment to a mixed turf–clover sward, with treatments applied March 1, 2011, and March 2, 2012.

Common name	Trifoliolate leaves	± 95% CI ^a	Height	± 95% CI	Flowers	± 95% CI
Bentazon	−34 b	11	6 a	10	−7 ab	12
Metribuzin	−83 de	10	−63 c	9	−87 cd	11
Simazine	−34 b	10	−5 a	9	−28 b	11
2,4-D	−67 cd	10	−66 c	9	−94 d	11
2,4-DB	−34 b	10	−8 a	9	−29 b	12
Clopyralid	−81 de	11	−72 c	9	−99 d	12
Glyphosate	−98 e	10	−95 d	9	−97 d	11
Imazethapyr	−38 b	10	−55 bc	9	−90 cd	12
Imazamox	−19 ab	11	−40 b	9	−65 c	12
Imazaquin	−72 de	11	−75 cd	10	−98 d	12
Halosulfuron	−64 cd	10	−74 cd	9	−100 d	11
Metsulfuron-methyl	−70 d	10	−77 cd	9	−100 d	11
Nicosulfuron	−44 bc	10	−66 c	9	−92 cd	12
Pronamide	−6 a	10	−9 a	9	−16 ab	12
Nontreated	0 a	10	0 a	9	0 a	11

^a Means were separated based on adjusted 95% confidence intervals (95% CI). Overlapping 95% CIs indicate a lack of significant difference.

^b Mean ± SD nontreated trifoliolate leaf density = 6,514 trifoliolate leaves m^{−2} (± SD 2,265); nontreated height = 18 cm (± SD 12); nontreated flower density = 1,599 inflorescences m^{−2} (± SD 809).

Table 4. Small hop clover response 4 wk after treatment to a mixed turf–clover sward, with treatments applied March 1, 2011, and March 2, 2012.

Common name	Trifoliolate leaves	± 95% CI ^a	Height	± 95% CI	Flowers	± 95% CI
	% change relative to the nontreated ^b					
Bentazon	–11 ab	14	–11 ab	9	–11 a	11
Metribuzin	–61 def	14	–49 ef	9	–67 cde	11
Simazine	–33 bcd	14	–28 bcd	9	–57 cd	11
2,4-D	–89 fg	14	–81 ghi	9	–94 ef	11
2,4-DB	–49 cde	14	–47 def	9	–58 cd	11
Clopyralid	–81 efg	14	–82 hi	9	–95 f	11
Glyphosate	–97 g	14	–94 i	9	–100 f	11
Imazethapyr	12 a	14	–22 bc	9	–10 a	11
Imazamox	–10 ab	14	–29 bcde	9	–17 ab	11
Imazaquin	–58 def	14	–67 fgh	9	–91 ef	11
Halosulfuron	–80 efg	14	–77 ghi	9	–96 f	11
Metsulfuron-methyl	–72 efg	14	–79 ghi	9	–91 ef	11
Nicosulfuron	–61 def	14	–61 fg	9	–78 def	11
Pronamide	–22 abc	14	–38 cde	9	–42 bc	11
Nontreated	0 ab	14	0 a	9	0 a	11

^a Means were separated based on adjusted 95% confidence intervals (95% CI). Overlapping 95% CIs indicate a lack of significant difference.

^b Mean ± SD nontreated trifoliolate leaf density = 5,857 trifoliolate leaves m⁻² (± SD 3,352); nontreated height differed by location and year, possibly due to fall mowing frequency and timing, but average = 15 cm (± SD 7); nontreated flower density = 2,177 inflorescences m⁻² (± SD 819).

metribuzin, and 2,4-D did not reduce white clover leaf densities relative to the nontreated control (≤ 33% numerical reductions and up to 31% increase).

White clover height differed by location and year but averaged 12 cm (± SD 10) across 2011 and 2012 nontreated plots. Imazaquin, glyphosate, metsulfuron-methyl, clopyralid, and 2,4-D reduced white clover height between 39 and 60% relative to the nontreated control (Table 2). However, with the exception of 2,4-D, herbicides that did not reduce leaf density did not reduce white clover height either. Those herbicides were pronamide, halosulfuron, 2,4-DB, bentazon, imazethapyr, imazamox, simazine, nicosulfuron, and metribuzin (≤ 35% numerical reductions and up to 23% increase).

White clover flower density was highly variable, averaging 142 inflorescences m⁻² (± SD 187) across nontreated plots. With the exception of increases in flower density due to treatment with halosulfuron (65%) and imazamox (76%), herbicidal effects on white clover flower density were similar to that in the nontreated control (Table 2). Metribuzin, 2,4-D, clopyralid, glyphosate, imazethapyr, imazaquin, metsulfuron, and nicosulfuron all caused a 30% or greater reduction in flower

density; although, these were not statistically significant compared with nontreated plots. Additional research is necessary to determine whether these herbicides caused an unacceptable reduction in flower count. Flower density increases with imazamox and halosulfuron applications may be for several reasons, including herbicide growth regulation or decreased weed competition (although none was recorded).

Results suggest pronamide, halosulfuron, 2,4-DB, bentazon, imazethapyr, imazamox, simazine, nicosulfuron, and metribuzin are at least moderately well tolerated by white clover. Of the three clover species studied, white clover was unique because it is capable of vegetative establishment. For this reason, persistence seems less likely to be affected by reductions in flower production relative to annual clovers, such as ball and small hop clover.

Ball Clover. Ball clover leaf density measured 6,514 trifoliolate leaves m⁻² (± SD 2,265) across 2011 and 2012 nontreated plots. Glyphosate, metribuzin, clopyralid, and imazaquin reduced leaf density > 72% (Table 3). Metsulfuron-methyl, 2,4-D, and halosulfuron, reduced ball clover leaf density between 64 and 70%. Only pronamide and imazamox did not reduce ball clover leaf densities

relative to the nontreated control (6 and 19% reductions, respectively).

Ball clover height differed by location and year but averaged 18 cm (\pm SD 12) across 2011 and 2012 nontreated plots. Glyphosate, imazaquin, halosulfuron, and metsulfuron-methyl reduced ball clover height by $\geq 74\%$ (Table 3). Metribuzin, 2,4-D, clopyralid, and nicosulfuron reduced ball clover height approximately 65%. Imazethapyr and imazamox reduced ball clover height 55 and 40%, respectively, whereas 2,4-DB, bentazon, simazine, and pronamide did not cause a statistically significant reduction in ball clover height relative to the nontreated control ($\leq 9\%$ reductions and up to 6% increase).

Ball clover flower density averaged 1,599 inflorescences m^{-2} (\pm SD 809) across 2011 and 2012 nontreated plots. Ball clover flower density was reduced $> 65\%$ by metribuzin, imazethapyr, nicosulfuron, 2,4-D, glyphosate, imazaquin, clopyralid, halosulfuron, metsulfuron-methyl, and imazamox (Table 3). Simazine and 2,4-DB reduced ball clover flower density approximately 28%, whereas pronamide and bentazon did not reduce flower density relative to the nontreated (7% reduction).

Small Hop Clover. Small hop clover leaf density measured 5,857 trifoliolate leaves m^{-2} (\pm SD 3,352) across 2011 and 2012 nontreated plots. Glyphosate, 2,4-D, clopyralid, halosulfuron, and metsulfuron-methyl reduced small hop clover leaf density $> 72\%$ (Table 4). A range of intermediate responses were observed, with metribuzin, nicosulfuron, imazaquin, and 2,4-DB reducing leaf density of small hop clover between 49 and 61% relative to the nontreated control. Bentazon, imazamox, and pronamide caused a 10 to 22% reduction in leaf density, although that was not statistically significant, whereas imazethapyr caused a 12% increase.

The height of small hop clover differed by location and year but averaged 15 cm (\pm SD 7) across 2011 and 2012 nontreated plots. Glyphosate, clopyralid, 2,4-D, metsulfuron-methyl, and halosulfuron reduced the height of small hop clover $> 77\%$. Imazaquin nicosulfuron, metribuzin, 2,4-DB, and pronamide reduced the height of small hop clover between 38 and 67%. Imazamox, simazine, and imazethapyr reduced the height of small hop clover between 22 and 29%. Only bentazon did not reduce the height of small hop clover relative to the nontreated control.

The flower density of small hop clover averaged 2,177 inflorescences m^{-2} (\pm SD 819) across 2011 and 2012 nontreated plots. Glyphosate, halosulfuron, clopyralid, 2,4-D, metsulfuron-methyl, imazaquin, and nicosulfuron reduced the flower density of small hop clover $> 78\%$ (Table 4). Metribuzin, 2,4-DB, simazine, and pronamide reduced the flower density of small hop clover between 42 and 67%. Imazamox, bentazon, and imazethapyr did not reduce the flower density of small hop clover relative to the nontreated control (between 10 and 17% reduction).

Implications for Mixed Turf–Clover Swards. On a practical level, results indicated that bentazon was at least moderately well tolerated across the range of clover species tested ($< 35\%$ decrease by all response variables; Tables 2, 3, and 4). Previous results confirmed similar tolerance and, in some cases, increased clover height because of bentazon application (Ceballos et al. 2004; McCurdy et al. 2013). Bentazon can be applied to clover grown for seed (Anonymous 2015) and for broadleaf and sedge weed control within turfgrass (Anonymous 2014c).

Simazine was well tolerated by white clover ($\leq 5\%$ decrease by all response variables; Table 2), yet moderate injury to ball clover and small hop clover was observed (Tables 3 and 4). Simazine reduced ball clover and small hop clover leaf density approximately 33% and reduced flower density 28 and 57%, respective to species. Simazine is labeled for PRE and POST control of annual grasses and broadleaves within turf, including recommendations for control of newly emerged small hop clover (Anonymous 2014a). Results presented here do not fully support claims of small hop clover control; however, in this study, small hop clover had emerged more than 2 mo before treatment.

Pronamide was well tolerated by white and ball clovers, with no effect on response variables. However, pronamide decreased height and flower density of small hop clover, which may limit its use within stands where small hop clover is desired. Pronamide reduced the flower density of small hop clover by 42%, suggesting a possible selective option for suppressing small hop clover present as a weed within clover seed production. Pronamide can be applied to nonresidential turf for PRE and POST control of common grass and broadleaf weeds, including annual bluegrass (*Poa annua* L.) (Anon-

ymous 2009b; Toler et al. 2007). Pronamide is also intended for fall or winter applications to clover and several other leguminous crops (Anonymous, 2009a). However, reductions in white clover root nodulation have been reported when applied to 3-d-old seedlings (Clark and Mahanty 1991). Root nodulation was not measured within the presented study but is an avenue for future research.

Imazethapyr and imazamox were moderately well tolerated by white clover and small hop clover (< 39% decrease by all response variables), yet ball clover proved more susceptible to these herbicides than was anticipated based on previous results. McCurdy et al. (2013) reported that imazethapyr was tolerated by four clover species, with only a 12% reduction in ball clover height. Enloe et al. (2014) previously demonstrated white clover tolerance to imazethapyr within mixed grass–clover pastures. Similarly, Lins et al. (2005) demonstrated red clover tolerance to imazamox. Imazethapyr can be applied in clover and several other leguminous crops (Anonymous 2011). In newly established clover, it is applied after the second trifoliate leaf has emerged. In forage production, it is applied soon after mowing or harvest when clover foliage is absent. Imazamox can be applied on clover grown for seed in Washington and Oregon; however, its use requires the addition of bentazon to minimize clover injury (Anonymous 2014b). Likewise, Lins et al. (2005) reported increased red clover tolerance with the addition of bentazon. Imazamox can be used for foxtail (*Setaria* spp.) and crabgrass (*Digitaria* spp.) control, as well as for control of several problematic broadleaf weeds, including prostrate spurge [*Chamaesyce humistrata* (Engelm ex. Gray) Small] and chickweed (*Stellaria* spp.).

The herbicide 2,4-DB was well tolerated by white clover and moderately well tolerated by ball clover, but small hop clover was more susceptible. It has been suggested that 2,4-DB is a potential herbicide for maintaining clover within mixed grass–clover swards (Hawton et al. 1990; Smith and Powell 1979) although differential tolerance to 2,4-DB among clover species has previously been reported and may suggest utility for clover-in-clover weed control (McCurdy et al. 2013).

Flowering response to herbicides has not previously, to our knowledge, been reported for ball clover or small hop clover. Flower reductions likely indicate a disruption in seed production and

dispersal, ultimately leading to reduced clover persistence in a mixed stand. Thus, flower reductions are assumed to prevent reseeding of annuals, whereas vegetatively established white clover may recover during the growing season. On the other hand, halosulfuron and imazamox increased flower production in white clover, although it is unknown whether they had an effect on seed production or seed viability. Future research should evaluate the long-term effects of herbicides on stand persistence, rather than on short-term injury symptoms. Likewise, seed viability following herbicide applications and resulting stand loss over multiple years of application should be investigated.

An acceptable level of clover injury or stand density disruption has not previously been determined for low-maintenance, biodiverse turf. This is a shortcoming of this and prior research that may be addressed in future studies. Quantifying the effects of clover reduction on nitrogen fixation and composite grass–clover sward density may be two ways to determine acceptable clover injury.

There are numerous variables influencing clover tolerance to herbicides, including effects of mowing or herbivory, fertility, grass competition, and application timing, all of which should be investigated. The results presented here could also be discussed in relation to mixed grass–clover forage systems, although problematic weeds and management are often different. Importantly, weed control with suggested herbicides should be investigated so managers and producers can adapt these technologies widely.

Literature Cited

- Abraham CM, Held DW, Wheeler C (2010) Seasonal and diurnal activity of *Larra bicolor* (Hymenoptera: Crabronidae) and potential ornamental plants as nectar sources. *Appl Turf Sci*. DOI: 10.1094/ATS-2010-0312-01-RS
- Anonymous (2009a) Kerb® 50 WP herbicide product label. Indianapolis, IN: Dow AgroSciences. <http://www.cdms.net/LDat/ld5TU000.pdf>. Accessed August 13, 2015
- Anonymous (2009b) Kerb® 50 WP specialty herbicide product label. Indianapolis, IN: Dow AgroSciences. <http://www.cdms.net/ldat/ld5t2006.pdf>. Accessed August 13, 2015
- Anonymous (2011) Pursuit® herbicide product label. Research Triangle Park, NC: BASF Corporation. <http://www.cdms.net/LDat/ld01S011.pdf>. Accessed August 13, 2015
- Anonymous (2014a) Princep® Liquid product label. Greensboro, NC: Syngenta Crop Protection. <http://www.cdms.net/LDat/ld786000.pdf>. Accessed August 13, 2015

- Anonymous (2014b) Raptor® herbicide product label. Research Triangle Park, NC: BASF Corporation. <http://www.cdms.net/LDat/ld20T013.pdf>. Accessed August 13, 2015
- Anonymous (2014c) Basagran® T&O herbicide product label. Research Triangle Park, NC: BASF Corporation. <http://www.cdms.net/ldat/ldBUS000.pdf>. Accessed August 13, 2015
- Anonymous (2015) Basagran® herbicide product label., Cary, NC: Arysta LifeScience North America. <http://www.cdms.net/LDat/ld89N004.pdf>. Accessed August 13, 2015
- Carlisle RJ, Watson VH, Cole AW (1980) Canopy and chemistry of pasture weeds. *Weed Sci* 28:139–142
- Carmer SG, Nyquist WE, Walker WM (1989) Least significant differences for combined analysis of experiments with two- or three-factor treatment designs. *Agron J* 81:665–672
- Ceballos R, Palma G, Brevis H, Ortega F, Quiroz A (2004) Effect of five postemergence herbicides on red clover shoot and root growth in greenhouse studies. *Phytoprotection* 85:153–160
- Clark SA, Mahanty HK (1991) Influence of herbicides on growth and nodulation of white clover, *Trifolium repens*. *Soil Biol Biochem* 23:725–730
- DiTomaso JM (2000) Invasive weeds in rangelands: species, impacts, and management. *Weed Sci* 48:255–265
- Dudeck AE, Peacock CH (1983) Rate, method, and time of overseeding white clover on bermudagrass during the winter. *Proc Fla Hortic Soc* 96:159–161
- Enloe SF, Johnson J, Renz M, Dorough H, Tucker K (2014) Hairy buttercup control and white clover tolerance to pasture herbicides. *Forage Grazinglands* 12(1)
- Evers GW, Grichar WJ, Pohler CL, Schubert AM (1993) Tolerance of three annual forage legumes to selected postemergence herbicides. *Weed Technol* 7:735–739
- Hawton D, Johnson IDG, Loch DS, Harvey GL, Marley JMT, Hazard WHL, Bibo J, Walker SR (1990) A guide to the susceptibility of some tropical crop and pasture weeds and the tolerance of some crop legumes to several herbicides. *Trop Pest Manag* 36:147–150
- Ledgard SF, Steele KW (1992) Biological nitrogen fixation in mixed legume/grass pastures. *Plant Soil* 141:137–153
- Lins RD, Colquhoun JB, Cole CM, Mallory-Smith CA (2005) Postemergence small broomrape (*Orobancha minor*) control in red clover. *Weed Technol* 19:411–415
- Littell RC, Milliken GA, Stroup WW, Wolfinger RD, Schabenberger O (2006) SAS® for Mixed Models. 2nd edn. Cary, NC: SAS Institute Inc. Pp 15–56
- McCurdy JD, McElroy JS, Flessner ML (2013) Differential response of four *Trifolium* species to common broadleaf herbicides: implications for mixed grass-legume swards. *Weed Technol* 27:123–128
- McCurdy JD, McElroy JS, Guertal EA, Wood CW (2014) White clover inclusion within a bermudagrass lawn: effects of supplemental nitrogen on botanical composition and nitrogen cycling. *Crop Sci* 54:1796–1803
- McNeill AM, Wood M (1990) ¹⁵N estimates of nitrogen fixation by white clover (*Trifolium repens* L.) growing in a mixture with ryegrass (*Lolium perenne* L.). *Plant Soil* 128:265–273
- Munshaw G., Sparks B, Williams D, Barrett M, Beasley J, Woosley P (2015) Pre-plant cultivation techniques and planting date effects on white clover establishment into an existing cool-season turfgrass sward. *Hortscience* 50:615–620
- Roberts RD, Bradshaw AD (1985) The development of a hydraulic seeding technique for unstable sand slopes, II: field evaluation. *J Appl Ecol* 22:979–994
- Rogers ME, Potter DA (2004) Potential for sugar sprays and flowering plants to increase parasitism of white grubs by tiphiid wasps (Hymenoptera: Tiphidae). *Environ Entomol* 33:520–527
- Seefeldt SS, Stephens JMC, Verkaarik ML, Rahman A (2005) Quantifying the impact of a weed in a perennial ryegrass-white clover pasture. *Weed Sci* 53:113–120
- Sincik M, Acikgoz E (2007) Effects of white clover inclusion on turf characteristics, nitrogen fixation, and nitrogen transfer from white clover to grass species in turf mixtures. *Commun Soil Sci Plant Anal* 38:1861–1877
- Smith AE, Powell JD (1979) Herbicides for weed control during establishment of arrowleaf clover. Athens, Georgia: University of Georgia Research Report. 324 p
- Toler JE, Willis TG, Estes AG, McCarty LB (2007) Postemergent annual bluegrass control in dormant nonoverseeded bermudagrass turf. *Hortscience* 42:670–672
- Whitehead DC (1995). *Grassland Nitrogen*. Wallingford, U.K.: CAB International. Pp 397–117
- Young RR, Morthorpe KJ, Croft PH, Nico H (1992) Differential tolerance of annual medics, Nungarin subterranean clover and hedge mustard to broadleaf herbicides. *Aust J Exp Agric* 32:49–57

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