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Alternative Herbicides for Controlling Herbicide-Resistant Annual Bluegrass (*Poa annua* L.) in Turf

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Abstract: *Poa annua* is a cosmopolitan, cool-season grass species regarded as one of the most significant weeds of turfgrass. It is mainly controlled by herbicides; however, repeated use of herbicides in golf turf has resulted in the evolution of multiple-herbicide resistant *P. annua*. Four field experiments were performed in autumn and spring in golf turf to identify effective herbicide options to control multiple herbicide-resistant *P. annua*. In herbicide resistance screening, the trial site population (SA1) was found to be susceptible to amicarbazone and terbuthylazine, but resistant to simazine and metribuzin at the field rate of each herbicide. Consistent with the results of the pot study, the PSII-inhibiting herbicides amicarbazone and terbuthylazine provided the best control (80–100%) of *P. annua* in both autumn and spring trials with minimal damage to the turf. In contrast, the other two PSII-inhibiting herbicides, metribuzin and simazine, were relatively ineffective in controlling *P. annua* in the field. Indaziflam also performed well in both autumn trials and reduced *P. annua* occurrence by >75%. Pyroxasulfone and s-metolachlor only provided moderate weed control in both the autumn and spring trials, reducing *P. annua* occurrence by 50%. Among the nine different herbicides, amicarbazone and terbuthylazine were found to be most effective for spring and autumn application in turf. As resistance to some PSII-inhibiting herbicides has already evolved in this field population, the use of amicarbazone and terbuthylazine needs to be integrated with other herbicide modes of action and non-chemical tactics to delay the onset of resistance to them.

Keywords: *Poa annua* L.; annual bluegrass; amicarbazone; terbuthylazine; indaziflam; weed management

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1. Introduction

Annual bluegrass (*Poa annua* L.) is one of the most problematic weeds in sports turf, particularly in temperate climates [1,2]. It reduces the turf quality for sport and creates an uneven surface that affects ball roll [3,4]. This weed also competes for water and nutrients with the desired turfgrass species and reduces turf growth. It often produces panicles below the turf cutting height, which reduces the effectiveness of mowing for weed control [3]. *P. annua* is a genetically diverse weed species that typically germinates in autumn, grows in winter, and produces seed in spring; however, some germination can occur in spring as well [5,6]. During summer *P. annua* senesces, resulting in dead, bare, and unsightly patches that reduce the aesthetic value of turf [7]. It is considered the most problematic weed of golf courses in Australia and other countries, such as the USA. One feature that makes it difficult to control *P. annua* is its large seed bank of up to 200,000 seed m^{-2} [6] and potential for year-round germination [8].

Several alternative management strategies including manual, cultural, biological and chemical control are available for controlling *P. annua* [9,10]. However, chemical control tends to be most commonly used, because of the ease of application and reliability of weed control. Both PRE and POST herbicides are used to control *P. annua* in turf [11]. However, repeated use of herbicides has resulted in the evolution of herbicide resistant *P. annua* populations [12–14]. The loss of herbicides to resistance requires new weed management practices including additional herbicides to control *P. annua*.

Greenkeepers of the golf course used for this study had reported difficulty in controlling *P. annua* with several herbicides. Seeds were collected from the golf course trial site in October 2017 to screen for herbicide resistance. The population was confirmed resistant to four different herbicide modes of action HRAC (Herbicide Resistance Action Committee) group, 1, 2, 5 and 31 in initial research, with >9-fold resistance to HRAC group 1, 2 and 31 and >2-fold resistance to HRAC group 5. Therefore, alternate herbicides with different mode of action could be an option for controlling this population. Hence, four field trials were conducted at a golf course in spring and autumn seasons between 2018 and 2020. The treatments were selected based on some commonly used herbicides currently registered for controlling *P. annua* in golf turf in Australia and additional herbicides that are not currently registered for turf, but may be suitable for controlling *P. annua*. The objective of the study was to identify suitable herbicide options to control multiple herbicide-resistant *P. annua* in a bermuda grass (*Cynodon dactylon*) turf during autumn and spring.

2. Materials and Methods

2.1. Experimental Site and Field Trial Design

Field trials were conducted on a Bermuda grass (*Cynodon dactylon*) turf at a golf course (34.896°S 138.51°E) in spring and autumn between 2018 and 2020. The chosen site was a practice green, which received the same weed control measures as the remainder of the golf greens. The turf was well managed with regular mowing (twice a week) at a height of 9 mm and watered every third night depending on the weather conditions. The soil type of the trial site is sandy in texture with 1.48% organic matter (Table 1). Liquid fertilizer MP Brilliance (20-0-0 + 6Fe, 1Mg) was applied at 37 L ha^{-1} every 3-4 weeks to maintain the fertility of the golf course. Meteorological and soil properties of the experimental sites are presented in Tables 1 and 2.

Table 1. Calendar dates and environmental conditions during herbicide application in field trials in 2018−2020.

Application Timing	Trial	Date	Air Temperature at Spraying (°C)	Average Maximum Temperature during Trial (°C)	Average Minimum Temperature during Trial (°C)	Total Rainfall during Trial (mm)
Spring	Trial 1	24 October 2018	17.7	23.1	13.3	35.5
	Trial 2	30 September 2019	21.7	23.4	11.8	15.6
Autumn	Trial 1	8 March 2019	23.7	25.6	15.5	9.0
	Trial 2	24 March 2020	20.5	24.9	15.0	4.2

Table 2. Soil analysis data of the trial site.

Soil pH	Organic Matter (%)	Salinity EC 1:5 (dS m^{-1})	Texture	Physical Properties (%)		
				Sand	Silt	Clay
7.6	1.48	0.073	Sand	92	6.6	1.7

Nine different herbicide treatments were applied in the spring trial established in October 2018 and repeated in September 2019 (Table 3). Similarly, a field trial with nine herbicide treatments was undertaken in the autumn at the same location in March 2019 and repeated in March 2020 (Table 3). Dates of herbicide application and weather conditions are presented in Table 1. The timing of herbicide application in these trials is consistent with the weed management programs used by local greenkeepers. A range of both PRE and POST herbicides were used in both spring and autumn trials as emergence of *P. annua* occurs over a long period. Therefore, some herbicides e.g., s-metolachlor, indaziflam, which are mainly recommended for PRE application, were applied in both autumn and spring trials to test for both PRE and POST herbicide activity. Additionally, herbicides not currently registered for *P. annua* control were tested in these field trials. The trial was established in a randomised complete block design with four replications. The plot size was 5 m × 2 m with a 0.25 m gap between plots. A non-treated check treatment was included in each trial. Herbicides were applied using a CO_2-pressurized boom sprayer with medium droplet size flat-fan nozzles (TT01 and LD110-015) delivering 100 L ha^{-1} volume at a pressure of 200 kPa. The trials were assessed 28 DAT (days after treatment) and again 42 DAT.

Table 3. List of herbicides used in spring and autumn treatment.

Herbicide	Trade Name	HRAC Group [15]	Registered for Turf in Australia	Recommended Application Time	Used in Which Trial	Rate (g a.i. ha^{-1}) Used in Experiment	Company
Pendimethalin	Rifle® 440	3	Yes	PRE	Autumn	1496	Nufarm
Propyzamide (Pronamide)	Kerb® 500 SC	3	Yes	PRE, EPOST	Spring Autumn	600	Corteva Agrisci.
Metribuzin	Mentor® WG	5	No	PRE, EPOST	Spring Autumn	210	Adama
Simazine	Gesatop® 600 SC	5	Yes	PRE, POST	Spring Autumn	1200	Syngenta
Terbuthylazine	Terbyne® Xtreme® 875 WG	5	No	PRE, POST	Spring Autumn	875	Sipcam Pacific
Amicarbazone	Amitron® 700 WG	5	Yes	PRE, POST	Spring Autumn	700	Arysta LifeScience Australia Pty Ltd.
Pyroxasulfone	Sakura® 850 WG	15	No	PRE	Spring Autumn	100.3	Bayer
S-Metolachlor	Pennmag®	15	Yes	PRE	Spring Autumn	1920	Syngenta
Indaziflam	Specticle®	29	Yes	PRE, EPOST	Spring Autumn	50	Bayer
Endothall	Poachek®	31	Yes	EPOST, POST	Spring	262.5	Campbell Chemicals

2.2. Pot Trial Methodology

As most of the PRE herbicides applied in the trial were ineffective, a follow-up pot study was undertaken with the PRE herbicides (Table 4) to determine the resistance status of the *P. annua* population from the trial site (hereafter referred to as SA1). The *P. annua* population (SA1) used in this study was collected from the trial site during October 2018. One susceptible population collected from a non-golf course area was used as the susceptible control. Seed bulking was performed in 2019 and the pot trial conducted during July 2019 and repeated in June 2020 at the University of Adelaide following the method used by Barua, et al. [16]. Approximately 100 seeds were measured by volume (0.2 mL), placed onto the surface of standard potting mix [17] and herbicide applied directly onto the seed. Herbicides (Table 4) were applied using a laboratory moving boom sprayer equipped with a twin nozzle (Tee-jet 110° flat fan Spraying Systems, Wheaton, IL, USA) delivering an output of 118 L ha^{-1} at a pressure of 250 kPa and speed of 1 m s^{-1}. Immediately after herbicide application, the seeds were covered with 5 mm of potting mix. The pots were placed outside and watered as required. The experiment was assessed for seedling emergence 28 DAT (days after herbicide treatment). Plants that emerged and grew to the two-leaf stage were considered resistant to the herbicide treatment. The experiment was repeated.

In order to further investigate resistance in *P. annua* to PSII inhibiting herbicides, an additional pot trial was undertaken. In this dose-response pot trial herbicide rates used were the following: amicarbazone (0, 26.3, 52.5, 105, 210, 420, 840 and 1680 g a.i ha^{-1}), terbuthylazine (0, 32.8, 65.6, 131.3, 262.5, 525, 1050 and 2100 g a.i ha^{-1}), simazine (0, 26.3, 52.5, 105, 210, 420, 840 and 1680 g a.i ha^{-1}) and metribuzin (0, 13.1, 26.3, 52.5, 105, 210 and 420 g a.i ha^{-1}). The methodology used was the same as Barua et al. [16]. The susceptible population mentioned above was used as the susceptible control. Seeds were sown in trays

(330 by 200 by 50 mm) located outdoors. One to two leaf seedlings were transplanted into punnet pots (95 by 85 by 95 mm) containing the already described standard potting mix, with 5 plants per pot and replicated three times. At the 2-3 leaf stage, plants were treated with the herbicides using the laboratory moving boom sprayer mentioned above. Plants actively growing with new leaves after 28 days were classified as survivors, while plants with severe stunting or dead were considered susceptible [19]. The experiment was repeated.

Table 4. Rate of herbicides used and survival (%) of the field population SA1 to herbicides at PRE application in a pot study.

Chemical Name	HRAC Group [15]	Rate (g a.i. ha^{-1}) Used in Study	% Survival	Resistance Status *
Pendimethalin	3	1496	10	S
Propyzamide (Pronamide)	3	600	15	S
Metribuzin	5	210	30	R
Simazine	5	1200	34	R
Amicabazone	5	700	0	S
Terbuthylazine	5	875	19	S
Pyroxasulfone	15	100.3	0	S
S-Metolachlor	15	1920	14	S
Indaziflam	29	50	0	S

* R—Resistant: More than 20% survival compared to the untreated control is considered as resistant [18], S—Susceptible.

2.3. Data Collection and Analysis

Prior to herbicide treatment, a low density of *P. annua* was present on the site in the autumn trials, but a much larger number of *P. annua* plants were uniformly distributed at the trial site in spring trials. The occurrence of *P. annua* in the trials after treatment was determined with the use of a 1 by 1 m grid divided into four hundred 50 by 50 mm squares. The grid was placed in the centre of each plot at three random locations and the number of squares containing *P. annua* plants counted to determine the % occurrence. The treatments were visually assessed for turf phytotoxicity at 7 DAT, 28 DAT and 42 DAT using a scale of 0−100 where 0 = no visual injury and 100 = no green tissue [20].

The data of percent occurrence of *P. annua* were subjected to two way analysis of variance (ANOVA) with GenStat version 19 (VSN International Ltd. Hemel Hempstead, UK) with herbicide and experiment run treated as variables. As there was a significant variation between experimental runs ($p < 0.05$), the data of each run is presented separately. The data were square-root transformed before statistical analysis to normalize the distribution of the residuals. Where treatment differences were significant, the means of the transformed data were compared using Fisher's Protected LSD at $p = 0.05$. As there were no significant differences in turf quality between the two runs, the data were pooled for statistical analysis.

Dose response trials were set up as a completely randomized design and repeated. Data was subjected to three way ANOVA for each herbicide with population, rate and run as variables. For every herbicide there was a significant effect of rate ($p < 0.0001$) and population ($p < 0.0001$), but not for run. Therefore, data for each herbicide were pooled across experiments. Survival at each rate was converted to mortality and the data analyzed using PriProbit (1.63) [21] with the LD_{50} with 95% confidence intervals (CI) determined and resistance ratios calculated as LD_{50} Resistant/LD_{50} Susceptible. Population responses to the herbicides were considered different if confidence intervals of the LD_{50} did not overlap.

3. Results

3.1. Spring Trial Assessment

The occurrence of *P. annua* in the non-treated control for the two spring field trials varied between 86 and 100% of grids (Table 5), which indicates a relatively uniform spatial distribution at the site. In both spring trials, amicarbazone (2% occurrence) and terbuthylazine (15–20% occurrence) provided the greatest control. The remaining treatments were less effective, particularly endothall, which was ineffective in both spring trials due to herbicide resistance confirmed previously at this site Barua, et al. [16]. These less effective treatments showed inconsistency in weed control over the two years.

Table 5. Effect of spring herbicide treatments on % occurrence of *P. annua* in golf turf in 2018 and 2019 after 28 DAT.

Treatment	*P. annua* (% Occurrence) [a]	
	2018	**2019**
Propyzamide (Pronamide)	62.4 d	64.3 ef
Metribuzin	60.4 d	49.8 d
Simazine	66.3 d	57.7 e
Terbuthylazine	14.6 b	19.6 b
Amicarbazone	1.9 a	1.6 a
Pyroxasulfone	50.4 c	39.4 c
S-Metolachlor	55.5 cd	41.1 c
Indaziflam	59.6 d	39.9 c
Endothall	81.4 e	67.2 f
Non-treated	99.5 f	86.3 g

[a] values with different letters within each column are significantly different.

In 2018, amicarbazone provided the highest level of control by reducing *P. annua* occurrence to 2% followed by terbuthylazine that reduced *P. annua* occurrence to 14% (Table 5). However, the other two PSII-inhibiting herbicides (simazine and metribuzin) provided moderate control and reduced the occurrence of *P. annua* between 60 to 66% (Table 5). Other mode of action herbicides, such as indaziflam and propyzamide, also reduced occurrence to 60 to 62% (Table 5).

Amicarbazone and terbuthylazine were also the most effective weed control treatments in the 2019 spring trial, followed by pyroxasulfone, indaziflam and s-metolachlor. Amicarbazone reduced the occurrence of *P. annua* to 2% followed by terbuthylazine to 20% (Table 5). Simazine and metribuzin provided moderate control of *P. annua* similar to the results obtained in 2018. The next most effective treatments were pyroxasulfone and indaziflam that reduced *P. annua* occurrence to 39% followed by s-metolachlor to 41% (Table 5).

3.2. Autumn Trial Assessment

The occurrence of *P. annua* was high (>96%) in untreated plots for both 2019 and 2020 autumn trials (Table 6). Amicarbazone was once again the most effective herbicide in autumn 2019, followed by terbuthylazine, indaziflam, pyroxasulfone, and s-metolachlor. In contrast, pendimethalin, propyzamide and simazine were less effective treatments in both autumn trials. The occurrence of *P. annua* was reduced to 0% by amicarbazone, 6% by terbuthylazine, 22% by indaziflam, 43% by pyroxasulfone, and 45% by s-metolachlor (Table 6).

Table 6. Effect of autumn herbicide treatments on % occurrence of *P. annua* in golf turf in 2019 and 2020 after 28 DAT.

Treatment	*P. annua* (% Occurrence) [a]	
	2019	**2020**
Pendimethalin	83.5 f	80.5 g
Propyzamide (Pronamide)	84.9 f	56.1 e
Metribuzin	55.8 e	59.2 ef
Simazine	89.8 fg	68.4 f
Terbuthylazine	6.2 b	8.0 b
Amicarbazone	0.0 a	1.4 a
Pyroxasulfone	43.2 d	40.6 d
S-Metolachlor	44.9 de	55.0 e
Indaziflam	22.6 c	26.3 c
Non-treated	99.9 g	96.4 h

[a] values with different letters within each column are significantly different.

In 2020, amicarbazone was again the best treatment and reduced *P. annua* occurrence to below 2% (Table 6). Terbuthylazine was the second best treatment and reduced *P. annua* occurrence to 8%, followed by indaziflam to 26%, pyroxasulfone to 40%, and s-metolachlor to 55% (Table 6). Consistent with the spring trials, simazine and metribuzin were less effective than amicarbazone or terbuthylazine.

A difference in the performance of two herbicides (propyzamide and simazine) was observed in the two autumn trials (Table 6). As the soil type at the study site is sandy in texture (Table 2), higher rainfall in 2019 (Table 1) may have moved these herbicides through the soil profile reducing their performance in that year.

In all four trials, amicarbazone was the most effective treatment; however, it produced mild turf grass injury (data were not shown). This persisted for four weeks after herbicide application, followed by complete recovery of the turf at six weeks. No other treatments were phytotoxic to the turf. Perry, et al. [22] reported that amicarbazone provided superior control of *P. annua* through foliar and root uptake at 371 g ha^{-1} as compared to atrazine at 2025 g ha^{-1}. Another study suggested that amicarbazone is more active than other PSII inhibitors such as atrazine Dayan, et al. [23]. Amicarbazone has been recently registered for turf in Australia at 210 g ha^{-1} [24], a much lower rate than 700 g ha^{-1} used in our field trials that were initiated prior to the registration of amicarbazone. It is possible the lower rate on the label will provide less effective control of *P. annua* than observed in this trial and in other studies [22,23]. The efficacy of amicarbazone at lower rates on *P. annua* needs to be validated. Of all the herbicide treatments investigated, terbuthylazine consistently resulted in good turf quality followed by indaziflam, propyzamide and amicarbazone at six weeks after herbicide application. In both the autumn and the spring trials there was rapid recovery of turf grass and no difference in turfgrass density (data not shown here) at 6 weeks after herbicide application. However, terbuthylazine, which was the second most effective herbicide in these trials, is not registered for use in turf in Australia. Indaziflam was the next most effective herbicide for PRE application in autumn and is registered for use in Australia [25]. Two other herbicides, pyroxasulfone and s-metolachlor, provided similar levels of control (40−55%) in all the trials. S-metolachlor is registered in turf for *P. annua* control during the autumn season. Pyroxasulfone should be considered as an option for use in turf and would increase the range of herbicide options for *P. annua*. The use of herbicide rotations can help to slow the evolution of resistance in weeds.

3.3. Pot Trial Screening

To investigate why some PRE herbicides were not effective in the autumn field trial, a pot study was conducted with the trial site population (SA1) using these herbicides. Moderate levels of resistance (30 to 34% survival) were detected to metribuzin and simazine (Table 3), which could explain the poor efficacy of these two herbicides in the field. However, SA1 was susceptible to the other herbicides used in the field trials (Table 3). Therefore, other factors, such as weed growth stage, weed competition and environmental conditions appear to be responsible for the poor control of *P. annua* by these herbicides.

3.4. Dose-Response Trial

To further investigate the extent of resistance to the PSII inhibiting herbicides used in the trial, dose response experiments were conducted on SA1. Compared to the S population, SA1 had 2.6-fold resistance to simazine and 2.2-fold to terbuthylazine. The LD_{50} of SA1 population to metribuzin was 1.5-fold higher than the S population and for amicarbazone was 1.2-fold higher (Table 7). Low-level resistance does not always result in a failure of the herbicide in the field. Terbuthylazine controlled the S population well below the field rate, so this herbicide was effective despite the low-level resistance present. In contrast, metribuzin at the field rate only controlled the S population, so the increase in tolerance of SA1 is the likely cause of poor performance in the field. The dose response study confirmed the SA1 population exhibited greater resistance to simazine and lower resistance to terbuthylazine, but little resistance to amicarbazone.

Table 7. The dose required for 50% mortality (LD_{50}) of susceptible (S) and trial site population (SA1) to various PSII inhibiting herbicides with 95% confidence intervals (CI) in parentheses. R/S is the ratio of LD_{50} of resistant and susceptible populations.

Herbicides		**Population**	
		S	**SA1**
		LD_{50} g a.i. ha^{-1}	
Simazine		249.1 (193.9, 319.6)	638.4 (500.6, 818.6)
	R/S	-	2.6
Amicarbazone		284.0 (227.8, 353.9)	354.5 (284.7, 442.2)
	R/S	-	1.2
Metribuzin		72.8 (57.6, 91.9)	109.0 (86.3, 137.1)
	R/S	-	1.5
Terbuthylazine		258.2 (194.3, 342.9)	575.2 (440.3, 751.8)
	R/S	-	2.2

4. Discussion

Resistance to 10 herbicide modes of action has been reported in *P. annua* worldwide [12], which restricts the number of herbicide options available for its control in turf. Most of the herbicides used in the autumn trial exhibited poor control of *P. annua* in the field (Table 6), despite resistance not being confirmed in pot studies (Table 3). This indicates that factors other than herbicide resistance were responsible for the poor performance, such as high seed bank, herbicide application timing relative to weed emergence, soil type, thatch layer, and environmental conditions [10,26].

The timing of a herbicide application is important to maximise PRE herbicide effectiveness [11]. In the current study, herbicide timings were similar to those used by local greenkeepers. In our field study, the performance of indaziflam was greater in the autumn trial than the spring trial. Brosnan, et al. [27] reported that soil application of indaziflam was more effective than foliar application to control *D. ischaemum* and *P. annua*. They concluded that indaziflam needs to be absorbed by the roots to maximise POST control. Therefore, time of application is important for the success of indaziflam. It is likely that when this herbicide was applied in autumn some *P. annua* had already germinated, thus reducing the efficacy of the indaziflam. In previous research, indaziflam was shown to be more effective when used PRE and EPOST (early POST) [28], which could explain why it was less effective in our spring trials where weeds were established. Greenkeepers often reapply the same herbicide during the season to maximise activity on subsequent germinations, which was not done in these field trials. Multiple applications may be more effective than a single application. Thus, a follow-up application timing study is needed to determine the difference between a single and multiple applications of this herbicide.

Pronamide has been shown to provide good control *P. annua* with PRE and POST treatments [29]. However, in our study performance of pronamide only provided moderate efficacy (Tables 5 and 6). In a previous study, poor efficacy of pronamide used POST on *P. annua* in Georgia, USA was associated with reduced absorption and translocation [30]. Others have also claimed that unsuitable application timing (when plants are too large) can lead to the poor efficacy of pronamide [31].

Factors which influence the efficacy of PRE herbicides include solubility in soil solution, binding to organic matter and the half-life in soil [32]. Uptake of herbicides by the root occurs more readily when the herbicide is in soil solution. Out of the herbicides tested in the autumn trials, amicarbazone had the highest solubility in water [33] and was found to have the greatest efficacy. In contrast, terbuthylazine and indaziflam have lower water solubility that could account for their lower activity.

Soil organic matter content can also play an important role in PRE herbicide activity [34]. In our study, some the PRE herbicides were unsuccessful for controlling *P. annua* in the autumn trial, even though no herbicide resistance was detected. The soil at the trial site was sandy with 1.48% soil organic matter content (Table 2). Soil low in organic matter (low cation exchange capacity) has a lower tendency to bind herbicides and allows greater herbicide availability for uptake by weeds. However, heavy rainfall or frequent irrigation can move the herbicides through the soil profile before the compound has a chance to bind to the soil colloids and organic matter [32]. Since most *P. annua* seeds germinate in the top layer of soil [10,35], leaching of herbicides to lower layers of soil could result in reduced weed control.

Another factor that may have reduced the performance of PRE herbicides is the thatch layer, which is primarily organic matter (stems, stolons, roots), in turf that develops between the turf and soil surface [36]. This limits the movement of air, water and nutrition into the soil [37]. The thatch layer can also bind PRE herbicides, thereby reducing their effectiveness [38,39].

In conclusion, amicarbazone was the most effective herbicide for the reduction of P. annua occurrence (98–100%) followed by terbuthlazine (>80%), indazaflam (>63%) and pyroxasulfone (>57%) in both autumn and spring trials. Indaziflam performance was greater in the autumn trial than the spring trial indicating that indazaflam could be a good option for autumn application. The availability of multiple herbicides with different modes of action allows rotating herbicides in an herbicide management program, which is important to reduce the risk of resistance. Pyroxasulfone, although not currently registered in turf, could be a viable option for *P. annua* control. Amicarbazone and terbuthylazine controlled the trial site population (SA1) in pots indicating it was susceptible to both herbicides (Table 3). In contrast, simazine and metribuzin were less effective (Table 3). Terbuthylazine could be a potential candidate for *P. annua* control in turf. Given the extensive presence of herbicide resistance, it would be invaluable for the greenkeepers to test their populations for resistance status, so they can make informed choices of herbicides for *P. annua* control. The currently effective herbicides need to be used with care otherwise, they could also lose their effectiveness due to resistance.

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References

1. Heide, O. Flowering responses of contrasting ecotypes of *Poa annua* and their putative ancestors *Poa infirma* and *Poa supina*. *Ann. Bot.* **2001**, *87*, 795–804. [CrossRef]
2. Wódkiewicz, M.; Ziemiański, M.; Kwiecień, K.; Chwedorzewska, K.J.; Galera, H. Spatial structure of the soil seed bank of *Poa annua* l.—Alien species in the antarctica. *Biodivers Conserv.* **2014**, *23*, 1339–1346. [CrossRef]
3. McElroy, J.S.; Walker, R.H.; Wehtje, G.R.; Van Santen, E. Annual bluegrass (*Poa annua*) populations exhibit variation in germination response to temperature, photoperiod, and fenarimol. *Weed Sci.* **2004**, *52*, 47–52. [CrossRef]

4. Rana, S.S.; Askew, S.D. Measuring canopy anomaly influence on golf putt kinematics: Errors associated with simulated putt devices. *Crop. Sci.* **2018**, *58*, 900–910. [CrossRef]
5. Neylan, J.A.P. *Tu05001: A Lifecycle Approach to the Control of Poa Annua in Bentgrass Putting Greens, Final Project Report for Horticulture Australia Ltd (Hal)*; Hort Journal Australia: Queensland, Australia, 2009; p. 49.
6. Lush, W.M. Adaptation and differentiation of golf course populations of annual bluegrass (*Poa annua*). *Weed Sci.* **1989**, *37*, 54–59. [CrossRef]
7. Huff, D.R.; Casler, M.; Duncan, R. *Annual bluegrass (Poa annua l.). Turfgrass Biology, Genetics, and Breeding*; John Wiley & Sons: Hoboken, NJ, USA, 2003; pp. 39–51.
8. Galera, H.; Rudak, A.; Czyż, E.A.; Chwedorzewska, K.J.; Znój, A.; Wódkiewicz, M. The role of the soil seed store in the survival of an invasive population of *Poa annua* at point thomas oasis, king george island, maritime antarctica. *Glob. Ecol. Conserv.* **2019**, *19*, e00679. [CrossRef]
9. Vargas, J.; Turgeon, A.J. *Poa annua: Physiology, Culture, and Control of Annual Bluegrass*; John Wiley & Sons: Hoboken, NJ, USA, 2003.
10. Brosnan, J.T.; Breeden, G.K.; Zobel, J.M.; Patton, A.J.; Law, Q.D. Nonchemical annual bluegrass (*Poa annua*) management in zoysiagrass via fraise mowing. *Weed Technol.* **2020**, *34*, 482–488. [CrossRef]
11. Neylan, J.; Nickson, D. Poa Annua Control in Warm-Season Grass Sportsfield 2016. Available online: https://www.turfmate.com.au/uploads/STA%20Poa%20annua%20control%20in%20warm%20season%20grasses%20Final%20Report%20May%202016.pdf. (accessed on 15 June 2021).
12. Heap, I. International Survey of Herbicide Resistant Weeds. Available online: http://www.weedscience.org (accessed on 24 April 2020).
13. Cross, R.B.; Bridges, W.C.; McCarty, L.B.; McElroy, J.S. Evaluating annual bluegrass herbicide resistance evolution in golf course fairways. *Weed Technol.* **2015**, *29*, 488–500. [CrossRef]
14. Singh, V.; Dos Reis, F.C.; Reynolds, C.; Elmore, M.; Bagavathiannan, M. Cross and multiple herbicide resistance in annual bluegrass (*Poa annua*) populations from eastern texas golf courses. *Pest. Manag. Sci.* **2021**, *77*, 1903–1914. [CrossRef]
15. HRAC. Global Herbicide Classification Lookup. Available online: https://hracglobal.com/tools/classification-lookup/ (accessed on 15 June 2021).
16. Barua, R.; Boutsalis, P.; Malone, J.; Gill, G.; Preston, C. Incidence of multiple herbicide resistance in annual bluegrass (*Poa annua*) across southeastern australia. *Weed Sci.* **2020**, *68*, 340–347. [CrossRef]
17. Boutsalis, P.; Gill, G.S.; Preston, C. Incidence of herbicide resistance in rigid ryegrass (*Lolium rigidum*) across southeastern australia. *Weed Technol.* **2012**, *26*, 391–398. [CrossRef]
18. Panozzo, S.; Scarabel, L.; Collavo, A.; Sattin, M. Protocols for robust herbicide resistance testing in different weed species. *JoVE (J. Visual Exp.)* **2015**, *101*, e52923. [CrossRef]
19. Powles, S.B.; Lorraine-Colwill, D.F.; Dellow, J.J.; Preston, C. Evolved resistance to glyphosate in rigid ryegrass (*Lolium rigidum*) in australia. *Weed Sci.* **1998**, *46*, 604–607. [CrossRef]
20. Anderson, E. Herbicide Phytotoxicity Response and Eradication Studies in Miscanthus x Giganteus. Master's Thesis, University of Illinois at Urbana-Champaign, Urbana-Champaign, IL, USA, 2011. Available online: http://hdl.handle.net/2142/18498 (accessed on 10 June 2021).
21. Sakuma, M. Probit analysis of preference data. *Appl. Entomol. Zool.* **1998**, *33*, 339–347. [CrossRef]
22. Perry, D.; McElroy, J.; Walker, R. Effects of soil vs. Foliar application of amicarbazone on annual bluegrass (*Poa annua*). *Weed Technol* **2011**, *25*, 604–608. [CrossRef]
23. Dayan, F.E.; Trindade, M.L.; Velini, E.D. Amicarbazone, a new photosystem ii inhibitor. *Weed Sci.* **2009**, *57*, 579–583. [CrossRef]
24. Xtron* Herbicide Label. 2019. Available online: https://nuturf.com.au/product/xtron-700wg-herbicide/ (accessed on 15 June 2021).
25. Specticle®(200 g/L Indaziflam). Available online: www.environmentalscience.bayer.com.au (accessed on 11 June 2021).
26. Brosnan, J.T.; Elmore, M.T.; Bagavathiannan, M.V. Herbicide-resistant weeds in turfgrass: Current status and emerging threats. *Weed Technol.* **2020**, *34*, 424–430. [CrossRef]
27. Brosnan, J.T.; Breeden, G.K.; McCullough, P.E.; Henry, G.M. Pre and post control of annual bluegrass (*Poa annua*) with indaziflam. *Weed Technol.* **2012**, *26*, 48–53. [CrossRef]
28. Marble, S.C.; Chandler, A.; Archer, M. Impact of application rate, timing, and indaziflam formulation on early postemergence control of oxalis stricta. *Weed Technol.* **2016**, *30*, 701–707. [CrossRef]
29. Toler, J.E.; Willis, T.G.; Estes, A.G.; McCarty, L.B. Postemergent annual bluegrass control in dormant nonoverseeded bermudagrass turf. *Hort. Sci.* **2007**, *42*, 670–672. [CrossRef]
30. McCullough, P.E.; Yu, J.; Czarnota, M.A. First report of pronamide-resistant annual bluegrass (*Poa annua*). *Weed Sci.* **2017**, *65*, 9–18. [CrossRef]
31. CIPM. Is Herbicide Resistance to Blame? Available online: https://pesticidestewardship.org/resistance/herbicide-resistance/is-herbicide-resistance-to-blame-herbicide/ (accessed on 15 June 2021).
32. Hartzler, B. Absorption of Soil-Applied Herbicides. 2002. Available online: http://www.weeds.iastate.edu/mgmt/2002/soilabsorption.htm (accessed on 12 May 2010).
33. Hertfordshire, U. *Ppdb: Pesticide Properties Database*; University of Hertfordshire: Hatfield, UK, 2017. Available online: http://sitem.herts.ac.uk/aeru/ppdb/ (accessed on 10 June 2021).

34. GRDC. Pre-Emergent Herbicide Fact Sheet. Available online: https://grdc.com.au/data/assets/pdf_file/ (accessed on 5 June 2021).
35. Branham, B.E.; Hardebeck, G.A.; Meyer, J.W.; Reicher, Z.J. Turfgrass renovation using dazomet to control the *Poa annua* L. Soil seed bank. *Hort. Sci.* **2004**, *39*, 1763–1767. [CrossRef]
36. Martin, D.L.; Baird, J.; Sutherland, A. Thatch Management in Lawns. 2002. Available online: https://extension.okstate.edu/fact-sheets/thatch-management-in-lawns.html (accessed on 15 June 2021).
37. Clifton, N.; Dicklow, M.B.; Ebdon, S.; Jung, G.; Lanier, J.; Owen, M.; Prostak, R.; Spargo, J.; Vittum, P. Best Management Practices for Lawn and Landscape Turf. 2013. Available online: https://www.northeastipm.org/neipm/assets/File/Schools/Outdoors/BMPS_lawn_landscape_2013.pdf (accessed on 15 June 2021).
38. Schleicher, L.C.; Shea, P.J.; Stougaard, R.N.; Tupy, D.R. Efficacy and dissipation of dithiopyr and pendimethalin in perennial ryegrass (*Lolium perenne*) turf. *Weed Sci.* **1995**, *43*, 140–148. [CrossRef]
39. Dreistadt, S.H. Pests of Landscape Trees and Shrubs: An. Integrated Pest Management Guide. *UCANR Publ.* **2016**, *3359*. Available online: http://ipm.ucanr.edu/IPMPROJECT/ADS/manual_landscape.html (accessed on 15 June 2021).