Weed Technology 2013 27:317–322

Effect of Ambient Moisture on Aminocyclopyrachlor Efficacy

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Aminocyclopyrachlor (AMCP) is a newly developed synthetic auxin herbicide for broadleaf weed control in turfgrass systems. AMCP has been observed to undergo rapid photodecomposition in shallow water when exposed to sunlight. Most herbicide applications on golf courses occur during the morning when dew is still present on the turfgrass canopy. These conditions could result in efficacy loss if photolysis occurred while AMCP is suspended in dew droplets. Research was conducted to determine the effect of ambient moisture on AMCP efficacy. AMCP (79 and 105 g ae ha^{-1}), aminopyralid (280 g ae ha $^{-1}$), and two AMCP granular formulations (84 g ha^{-1}) were applied to dew-covered (WET) and dew-excluded (DRY) 'Tifway' bermudagrass plots. Herbicide treatments applied to WET plots had greater visually rated bermudagrass injury than respective treatments applied to DRY plots at 7 and 21 d after treatment (DAT), with the exception of aminopyralid at 21 DAT. Normalized difference vegetative index on turfgrass quality complemented visual ratings, indicating greater turfgrass quality reductions when applied to WET vs. DRY plots. These results indicate that AMCP applications made to dew-covered turfgrass can increase herbicidal efficacy, and no significant losses due to photodegradation were observed.

Nomenclature: Aminocyclopyrachlor, 6-amino-5-chloro-2-cyclopropyl-4-pyrimidine-carboxylic acid; aminopyralid, 4-amino-3,6-dichloro-2-pyridinecarboxylic acid; bermudagrass, *Cynodon dactylon* (L.) Pers. × *Cynodon transvaalensis* Burtt-Davy 'Tifway'.

Key words: Dew, herbicide loss, photolysis, turf injury.

Aminocyclopyrachlor (AMCP) es un herbicida del grupo de las auxinas sintéticas recientemente desarrollado para el control de malezas de hoja ancha en sistemas de céspedes. Se ha visto que AMCP sufre una rápida fotodescomposición en aguas superficiales cuando se expone a la luz solar. La mayoría de las aplicaciones de herbicidas en campos de golf se dan durante las mañanas cuando el rocío está todavía presente sobre el dosel del césped. Estas condiciones podrían resultar en una pérdida de eficacia si ocurre fotólisis mientras AMCP se encuentra suspendido en las gotas de rocío. Se realizó una investigación para determinar el efecto de la humedad ambiental sobre la eficacia de AMCP. Se aplicó AMCP (79 y 105 g ae ha^{-1}), aminopyralid (280 g ae ha^{-1}), y dos formulaciones granulares de AMCP (84 g ha^{-1}) a parcelas del césped *Cynodon dactylon* 'Tifway' cubiertas con rocío (WET) y sin rocío (DRY). Los herbicidas aplicados a parcelas WET tuvieron un mayor daño evaluado visualmente que los tratamientos respectivos aplicados a parcelas DRY a 7 y 21 días después del tratamiento (DAT), con la excepción de aminopyralid a 21 DAT. El índice de diferencia vegetativa normalizada de la calidad del césped complementó las evaluaciones visuales, indicando mayores reducciones en la calidad del césped cuando se aplicaron parcelas en WET vs. DRY. Estos resultados indican que las aplicaciones AMCP hechas en césped cubierto con rocío pueden incrementar la eficacia del herbicida y no se observaron pérdidas significativas debido a fotodegradación.

Synthetic auxin herbicides are commonly used in turfgrass settings to control broadleaf weeds (Senseman 2007). Aminocyclopyrachlor (AMCP) is a synthetic auxin herbicide belonging to the newly developed pyrimidine carboxylic acid herbicide family with a similar mode of action and chemical structure to phenoxy and pyridine compounds (Claus et al. 2008; Senseman 2007). These compounds mimic the natural plant hormone indole-3-yl-acetic acid but are not metabolized rapidly in susceptible dicotyledonous plants (Grossmann 2009). AMCP and other synthetic auxin compounds are translocated systemically through the phloem or xylem to meristematic plant regions where injury can be characterized by a loss in apical dominance, leaf cupping, epinastic curvature, and unregulated plant growth (Cobb and Reade 2010; Flessner et al. 2011a; Grossmann 2009). For herbicides to be efficacious, the compound must first contact the leaf surface, absorb through the plant cuticle, translocate to the intended target site, and affect a specific biochemical process (Ross and Lembi 1999). Although the aforementioned process seems easily achievable through typical herbicide application methods, numerous physiological, biochemical, and environmental barriers can inhibit synthetic auxin herbicide absorption into the targeted plant, causing reduced efficacy (Cobb and Reade 2010; Hess and Falk 1990; Monaco et al. 2002).

Terrestrial plant leaf surfaces are covered with a protective cuticular layer composed of hydrophobic/lipophilic trichomes, waxes, cutin, and pectin, which protects from desiccation and pathogen invasion; however, the cuticle poses a major physiological barrier to herbicide uptake (Cobb and Reade 2010; Currier and Dybing 1959; Hess and Falk 1990; Ross and Lembi 1999; Taiz and Zeiger 2006). Using scanning electron microscopic techniques, Hess and Falk (1990) noted that diverse epidermal morphology among various plant species greatly affected herbicide distribution on the leaf surface. The authors observed less MCPA distribution on bermudagrass compared with sugar beet (*Beta vulgaris* L), which was attributed to the thick epicuticular wax layer in bermudagrass. The physicochemical properties of AMCP denote high water solubility ($K_s = 4{,}200$ mg L^{-1}) and low

DOI: 10.1614/WT-D-12-00131.1

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Table 1. Environmental conditions at the Lake Wheeler Turf Field Laboratory, Raleigh, NC surrounding initiation of experimental runs.[a]

	October 1, 2009					October 1, 2011				
Time (EST)	Air temp.[b]	Dew point[c]	Relative humidity[d]	Soil temp.[e]	PAR[f]	Air temp.	Dew point	Relative humidity	Soil temp.	PAR
4:00 A.M.	12.5	8.1	74	15.9	0	12.4	6.6	67	20.3	0
5:00 A.M.	12.6	8.1	74	15.9	0	12.0	5.7	65	19.9	0
6:00 A.M.	12.3	7.9	74	15.8	0.866	11.3	4.1	61	19.6	3.389
7:00 A.M.	12.7	7.9	72	15.7	148.9	11.0	4.1	62	19.3	233.9
8:00 A.M.*	14.3	7.6	63	15.7	466.7	12.3	4.0	57	19.1	650.7
9:00 A.M.	16.4	8.5	59	15.7	745	13.5	3.6	51	19.3	1033
10:00 A.M.	17.9	9.8	59	15.7	1082	14.8	3.7	47	19.4	1359
11:00 A.M.	18.9	10.7	58	15.9	1071	15.6	4.0	46	20.2	1538
12:00 noon	20.1	10.1	52	16.2	1148	16.8	3.6	41	20.9	1758
1:00 P.M.	21.5	10.9	51	16.6	1136	17.0	2.7	38	21.6	1671
2:00 P.M.	21.8	11.4	51	16.8	1012	16.6	1.7	36	21.9	1438
3:00 P.M.	22.2	11.3	50	17.1	858	16.5	1.6	36	22.1	1097
4:00 P.M.	21.9	11.2	50	17.2	493.6	15.3	2.9	43	21.8	755.3

[a] Abbreviations: EST, eastern standard time; Temp., temperature; PAR, photosynthetically active radiation.

[b] Air temperature (C) from a 2-m height.

[c] Calculated dew point from a 2-m height.

[d] Percent relative humidity from a 2-m height.

[e] Soil temperature (C) from 0.1-m depth.

[f] Photosynthetically active radiation (µmol s^{-1} m^{-2}) from a 2-m height.

* Denotes time of herbicide applications.

octanol–water partition coefficient ($K_{ow} = -2.48$), indicating that the compound does not bind strongly to lipophilic cuticular constituents on the leaf surface (Claus et al. 2008). However, foliar herbicide absorption is also dependent on environmental conditions before, during, and after herbicide application. Periods of high relative humidity surrounding a herbicide application can increase efficacy due to prolonged spray droplet drying time on the leaf surface (Richardson 1977). Research from Pallas (1960) reported greater 2,4-D absorption and translocation in kidney bean (*Phlaseolus vulgaris* L.) under 70 to 74% humidity than between 34 to 48% humidity. Cuticular thickness may increase under low-humidity conditions or extended dry periods, reducing herbicide penetrability (Currier and Dybing 1959). Furthermore, low-humidity conditions can quicken spray droplet drying, creating unabsorbable synthetic auxin crystalline deposits on the leaf surface (Hess and Falk 1990). Because of the high K_s of synthetic auxin herbicides, dew formation after an application may resuspend crystalline deposits, making them available for plant uptake (Claus et al. 2008; Monaco et al. 2002; Senseman 2007).

AMCP has a broad-spectrum weed control at low application rates compared with phenoxy and pyridine compounds, making it an ideal alternative for broadleaf weed control in turfgrass systems (Claus et al. 2008; Curtis et al. 2009; Finkelstein et al. 2008; Flessner et al. 2011b; Gannon et al. 2009). AMCP received registration under the trade name Imprelis® in 2010 for use in commercial and residential fine turf (Anonymous 2010). Before registration, a field study conducted by DuPont Crop Protection indicated that AMCP underwent rapid field photolysis after a morning application to a dew-covered centipedegrass [*Eremochloa ophiuroides* (Munro.) Hack.] turfgrass stand (Jon S. Claus, personal communication). Literature has also reported AMCP to undergo rapid photodegradation in shallow water when exposed to sunlight (half-life = 1.2 d) (Claus et al. 2008). Potential AMCP photodegredation in dew-covered turfgrass is concerning as most herbicide applications on golf courses are made during the early morning to avoid patron exposure and utilize the dew pattern created by sprayer tires for guiding proper application coverage. Because of current application practices utilized by golf course managers, it is hypothesized potential photolysis could result in efficacy loss if AMCP applications are made to a dew-covered turfgrass stand. On the basis of this premise, research was conducted to determine the effect of ambient moisture on AMCP efficacy.

Materials and Methods

Field trials were initiated October 1, 2009 and October 1, 2011 at the North Carolina State University Turf Field Laboratory in Raleigh, NC to mature 'Tifway' bermudagrass (Table 1). Bermudagrass was selected as an indicator species because it displays an intermediate tolerance to AMCP (Flessner et al. 2011c). Experimental design was a 6 by 2 factorial treatment arrangement (six herbicide treatments by two ambient moisture levels) in a randomized complete block with four replications and two experimental runs.

Herbicide treatments included: foliar-applied AMCP (79 and 105 g ae ha^{-1} [Imprelis® herbicide, E. I. DuPont de Nemours, Wilmington, DE 19805]) and aminopyralid (280 g ha^{-1} [Milestone® herbicide, Dow AgroSciences, Indianapolis, IN 46268]); two granular AMCP formulations (coarse and fine prill size at 84.1 g ha^{-1} [aminocyclopyrachlor granular herbicide, E. I. DuPont de Nemours]); and a nontreated check. Although not labeled for use in commercial or residential turfgrass, aminopyralid was applied for comparative purposes because of its similar chemical structure, mode of action, and physicochemical properties to AMCP (Senseman 2007). Foliar herbicide applications were made with a

https://doi.org/10.1614/WT-D-12-00131.1 Published online by Cambridge University Press

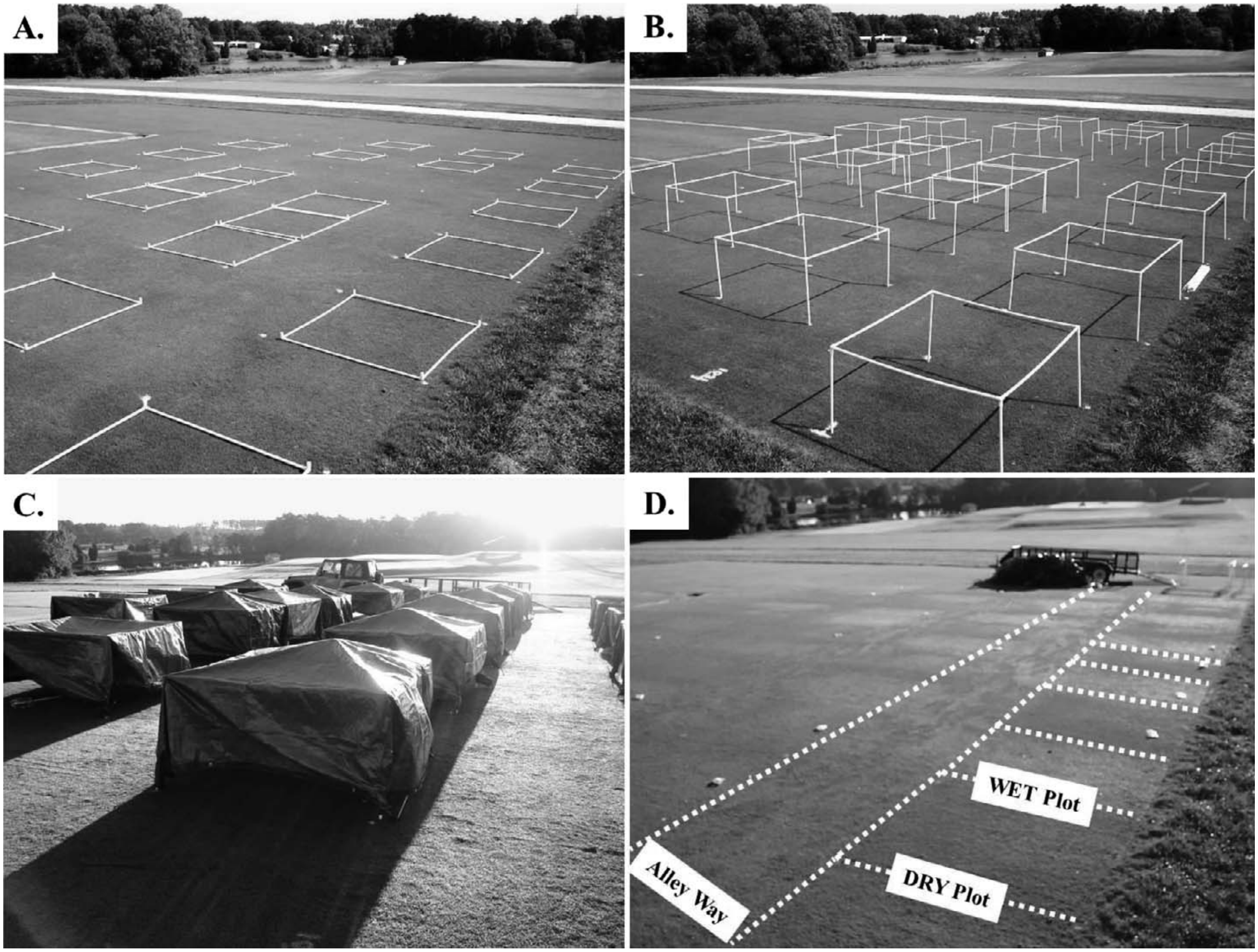

Figure 1. Schematic of tent structures used for dew exclusion: (A) and (B) Polyvinyl chloride frame assembly over DRY plots the evening before experiment initiation; (C) tarped tent structures the following morning; and (D) DRY (dew-excluded) vs. WET (dew-covered) plots before herbicide applications.

CO_2 pressurized spray boom (CO_2-pressurized sprayer, Spraying Systems Co., Wheaton, IL 60189) equipped with four TeeJet 8002 XR flat-fan nozzles (Teejet nozzles, TeeJet Technologies, Springfield, IL 62703) on 24-cm spacings calibrated to deliver 304 L ha^{-1} at 224 kPa. Granular herbicide applications were made using shaker jars (Ball® 16 oz canning jar, Jarden Home Brands, Daleville, IN 47334).

Ambient moisture levels included plots (2.25 m^2; 1.5 m by 1.5 m) covered overnight to prevent dew formation (DRY) and plots left uncovered so natural dew could form (WET) (Figure 1). Tent structures on the DRY plots were constructed using polyvinyl chloride piping (1.25-cm diameter) to create a 1.5 m by 1.5 m by 1 m frame covered with a 7.3-m^2 plastic tarp (Wel-Bilt® woven poly tarp, Northern Tools + Equipment, Raleigh, NC 27606). To allow air movement for guttation reduction, a 15-cm gap was left between the bottom of the tarp and turfgrass canopy. The tent structures were placed over DRY plots the evening before experiment initiation and removed the following morning before herbicide applications. A 1.5 m by 1.5 m buffer was included between replications to not disturb dew formation in WET plots while removing tent structures.

Data collection included visual bermudagrass injury rated weekly on a 0 to 100% scale (0% = no visible turfgrass injury; 100% = complete turfgrass death). In addition, normalized difference vegetation index (NDVI) was collected 21 d after treatment (DAT) using a Field Scout™ TCM 500 NDVI Turf Color Meter (Spectrum™ Technologies, Inc. Plainfield, IL 60585). Four reflectance readings were recorded within each plot and NDVI was calculated. NDVI values were converted to turfgrass quality ratings (1 to 9 scale: 1 = poorest quality; 7 = acceptable quality; and 9 = highest quality) using the equation: $[(\text{NDVI} \times 6.6) + 2.26]$ (Kieffer 2009). Research has indicated that NDVI can accurately and nonsubjectively evaluate turfgrass quality in relation to herbicide injury (Bell et al. 2002).

ANOVA was conducted in SAS (SAS statistical software, Version 9, SAS Institute Inc., Cary, NC 27513) using mixed-

Table 2. Interaction of herbicide treatment and ambient moisture level main effects on 'Tifway' bermudagrass visual injury and NDVI turfgrass quality 7 and 21 DAT, respectively[ab].

Herbicide treatment	Rate g as ha^{-1}	Ambient moisture	Visual injury[c] 7 DAT	Visual injury[c] 21 DAT	Turf quality[d] 21 DAT
			%	%	1–9
AMCP	79	DRY	17 CD[e]	27 D	7.1 AB
—	—	WET	21 B	32 BC	6.9 BC
AMCP	105	DRY	19 BC	33 B	6.8 C
—	—	WET	28 A	44 A	6.4 D
Aminopyralid	280	DRY	14 DE	28 D	6.2 E
—	—	WET	21 B	29 CD	5.5 G
AMCP fine	84	DRY	1 G	21 E	6.5 D
—	—	WET	7 F	26 D	6.0 EF
AMCP coarse	84	DRY	10 E	17 F	6.0 EF
—	—	WET	15 D	21 E	5.9 F
Nontreated	—	DRY	0 G	0 G	7.3 A
—	—	WET	0 G	0 G	7.2 A

[a] Abbreviations: NDVI, normalized difference vegetation index; DAT, days after treatment; AMCP, aminocyclopyrachlor; AMCP fine, aminocyclopyrachlor fine granular; AMCP coarse, aminocyclopyrachlor coarse granular; DRY, treatments applied in the absents of dew; WET, treatments applied in the presence of dew.

[b] Pooled analysis over two experimental runs.

[c] Evaluated visually on a 0 to 100% scale (0% = no injury; 100% = complete plant death).

[d] Evaluated using TCM 500 Turf Color Meter radiometer on a 1 to 9 scale (1 = poorest quality; 7 = acceptable quality; and 9 = highest quality).

[e] Means within columns with same letter (A–G) are not significantly different according to Fisher's Protected LSD ($P < 0.01$).

model methodology (SAS 2004). Herbicide treatment and ambient moisture levels were considered fixed variables in the model and evaluated to determine if there was an interactive effect. Experimental run was considered a random variable, allowing for the comparison of treatment means and interactions over multiple environments (Carmer et al. 1989). Experimental run, replication, and interactions between these effects were considered random effects in the model. Means were separated using Fisher's Protected LSD ($P \leq 0.05$).

Results and Discussion

ANOVA determined that a significant interaction ($P < 0.01$) was evident between the herbicide treatment and ambient moisture main effects on visual bermudagrass injury and NDVI turfgrass quality at 7 and 21 DAT; therefore, only the interactive effects will be reported (Table 2).

Herbicide Treatment and Ambient Moisture Level Interaction. At 7 DAT, visually rated bermudagrass injury was greater from all herbicide treatments applied to WET plots than DRY plots (Table 2). Foliar-applied AMCP at 79 and 105 g ha^{-1} to WET plots injured bermudagrass 21 and 28%, respectively. Conversely, injury from the aforementioned AMCP rates was reduced to 17 and 19%, respectively, when applied to DRY plots. The comparative aminopyralid treatment also demonstrated greater bermudagrass visual injury when applied to WET vs. DRY plots (21 vs. 14%, respectively), indicating that this exacerbated response to ambient moisture may also occur with other synthetic auxin herbicides with similar physicochemical properties (Claus et al. 2008; Senseman 2007). As observed with foliar-applied AMCP and aminopyralid, fine and coarse AMCP granules applied to WET plots injured bermudagrass more (7 and 15%, respectively) than DRY plots (1 and 7%, respectively). No visual bermudagrass injury was observed in WET or DRY nontreated plots.

At 21 DAT, greater bermudagrass injury was observed from all herbicide treatments applied to WET vs. DRY plots, with the exception of aminopyralid (Table 2). AMCP applied to WET plots at 79 and 105 g ha^{-1} injured bermudagrass 32 and 44%, respectively, whereas the same rates applied to DRY plots injured 27 and 33%, respectively. Regarding all herbicide treatments, the fine and coarse AMCP granular applied to DRY plots had the least amount of bermudagrass injury (21 and 17%, respectively). NDVI turfgrass quality showed a similar response to visual injury evaluations, as quality was reduced more from foliar-applied AMCP (105 g ha^{-1}), aminopyralid, and fine AMCP granular applied to WET plots compared with DRY plots (Table 2). Although not statistically different, a trend in reduced turfgrass quality in WET vs. DRY plots was noted from the low AMCP rate and coarse AMCP granular formulation. Excluding the low AMCP rate applied to DRY plots, all herbicide treatments applied to WET or DRY plots had reduced turfgrass quality (≤ 6.9) compared with the WET and DRY nontreated (7.3 and 7.2, respectively).

It should be noted that visually rated injury and NDVI data collection ceased 21 DAT (October 21, 2009 and 2011) as environmental conditions caused the bermudagrass stand to enter dormancy, thereby reducing the effectiveness of the aforementioned data collection techniques. Flessner et al. (2011c) indicated that bermudagrass injury from AMCP applications was most evident from 28 to 56 DAT when applied during the summer months. However, the 21-d evaluation period over the course of this study provided sufficient evidence to conclude that herbicidal efficacy increased when applied in the presence of ambient moisture.

Furthermore, these findings are congruent with past research indicating AMCP to cause reduced bermudagrass visual quality and percent green cover (Flessner et al. 2011c). Evaluations to the experimental plots resumed the following seasons to find that no treatments delayed bermudagrass spring green-up (data not shown).

Peason's Correlation Coefficients. Pearson's correlation coefficients were analyzed between bermudagrass visual injury and NDVI turfgrass quality taken 21 DAT. Visual injury and NDVI turfgrass quality demonstrated a strong negative relationship ($r = -0.88$; $P < 0.001$), indicating that an increase in visual injury corresponded to a decrease in NDVI turfgrass quality. This result is similar to past research indicating strong correlations between visual ratings and nonsubjective rating assessments, supporting the utility of visual evaluations to assess herbicide injury within the scientific community (Lewis et al. 2010; Yelverton et al. 2009).

Research Implications. Results from this study were contrary to the original hypothesis that AMCP applied to dew-covered turfgrass would experience efficacy loss due to photolysis. In fact the exact opposite was observed as herbicidal efficacy increased when applied to WET plots. Similarly, Kogan and Zu'ñiga (2001) reported that dew did not affect glyphosate efficacy when applied at low and medium application volumes (150 and 300 L ha^{-1}). The presence of dew on turfgrass leaves can result in cuticle hydration and increase herbicide coverage, leading to greater herbicide absorption (Caseley 1989; Johnstone 1973). Flessner et al. (2011c) reported that AMCP injured bermudagrass more in greenhouse studies than in field experiments, which could be attributed to greater AMCP absorption in greenhouse-conditioned bermudagrass due to reduced cuticle thickness. Dew may also reduce crystalline herbicide deposits from forming on the leaf surface, leading to greater foliar absorption (Hess and Falk 1990). Because of the high water solubility of AMCP and aminopyralid, it is speculated that coverage increased as the foliar-applied herbicides became suspended in dew droplets on the turfgrass leaves. Although aminopyralid is not labeled for use in turfgrass systems, these findings may be extrapolated to bermudagrass pastures, ranges, recreational areas, and roadside settings where the compound is currently registered (Anonymous 2008). Herbicide applications to WET plots likely allowed for a longer retention time on the leaf surface than applications made to DRY plots, which may have undergone formation of crystalline deposits as spray droplets evaporated. Granular herbicide formulations are intended for root absorption; however, the presence of dew at application can suspend the herbicide on the turfgrass leaves, leading to foliar uptake. Although AMCP photodegradation in shallow water bodies has been reported to have a half-life of 1.2 d, the dew was only present on the leaf surface in this study for approximately 4 h after application (Claus et al. 2008). On the basis of the reported observations, suspension time in dew had minimal effects on AMCP photodegradation and did not reduce herbicidal efficacy. These findings may warrant future research to quantify the increased AMCP activity on bermudagrass when applied in the presence of ambient moisture, perhaps through examining uptake and translocation using radiolabeled compounds.

As of August 2011, the Environmental Protection Agency ordered DuPont Crop Protection to remove AMCP from the turfgrass marketplace as reports indicated that Imprelis® applications were responsible for widespread injury to conifer tree species (USEPA 2011). Although AMCP is no longer used within turfgrass systems, future AMCP markets may include pasture, range, and roadside areas that could contain bermudagrass and other intermediately tolerant turfgrass species (Claus et al. 2008). Regardless of the setting, AMCP applicators need to be aware that herbicidal efficacy can increase when applied in the presence of ambient moisture on the leaf surface, potentially causing undesired bermudagrass injury.

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Received September 4, 2012, and approved November 8, 2012.