Weed Technology 2007 21:166–170

Reduced Translocation Is the Cause of Antagonism of Glyphosate by MSMA in Browntop Millet (*Brachiaria ramosa*) and Palmer Amaranth (*Amaranthus palmerii*)

Ian C. Burke, Clifford H. Koger, Krishna N. Reddy, and John W. Wilcut*

Studies were conducted in growth chambers to characterize absorption and translocation of ^{14}C-glyphosate applied alone or in mixture with MSMA in browntop millet and Palmer amaranth. MSMA antagonized activity of glyphosate in both weed species. Absorption of ^{14}C-glyphosate in Palmer amaranth was rapid and increased with time from 11.1% at 0.5 h after treatment to 68.1% at 168 HAT. Absorption of ^{14}C-glyphosate in browntop millet ranged from 1.6% at 0.5 HAT to 39.1% at 168 HAT. MSMA in mixture with glyphosate did not affect the absorption of glyphosate. In browntop millet, only 2.8% of the applied radioactivity translocated out of the treated leaf to the rest of the plant when glyphosate was applied in mixture with MSMA compared to 10.8% when glyphosate was applied alone at 72 HAT. Similarly, in Palmer amaranth, 3.2% of the applied radioactivity had translocated out of the treated leaf when glyphosate was applied in mixture with MSMA compared to 10.6% when glyphosate was applied alone. Reduced translocation appears to be the cause of the previously observed antagonism of glyphosate by MSMA.

Nomenclature: Glyphosate; MSMA; browntop millet, *Brachiaria ramosa* (L.) Stapf PANRA; Palmer amaranth, *Amaranthus palmerii* S. Wats. AMAPA.

Key words: Absorption, herbicide interaction, translocation.

MSMA is registered for control of sedge, grass, and broadleaf weeds in cotton (Askew et al. 2002; Culpepper et al. 2004; Porterfield et al. 2002). Before the advent of glyphosate-resistant cotton, MSMA was applied over the top often as a salvage treatment when early-season broadleaf weed control failed (Monks et al. 1999) and still is registered for postemergence (POST) and post-directed (PD) weed control in cotton. MSMA is applied PD extensively for control of late-season weeds in glyphosate-resistant and conventional cotton. Boll abortion following glyphosate applications during reproductive stage of cotton is a concern in glyphosate-resistant cotton (Pline-Srnic et al. 2004), and glyphosate is not registered for POST application on cotton past the four-leaf stage (Anonymous 2005).

Cotton growers often apply glyphosate and MSMA in mixture to maximize weed control (Stanley Culpepper, personal communication). In field and greenhouse studies evaluating compatibility of MSMA in tank mixtures with glyphosate or glufosinate for broadleaf and grass weed control, MSMA antagonized glyphosate efficacy on barnyardgrass [*Echinochloa crus-galli* (L.) Beauv.], browntop millet, hemp sesbania [*Sesbania exaltata* (Raf.) Rybd. Ex A. W. Hill], Palmer amaranth, and redroot pigweed (*Amaranthus retroflexus* L.) (Koger et al. 2007). Antagonism of glyphosate by MSMA was often overcome by applying glyphosate at a 2× rate (1.68 kg ae/ha), although MSMA is not registered for mixture with glyphosate for weed control in cotton (Koger et al. 2007).

The mode of action of MSMA is not well known. However, MSMA does cause cell disruption where it comes in contact with susceptible plant tissue (Duke 1992). Knowles and Benson (1983) demonstrated that methanearsonate can be photochemically reduced by photosystem I of photosynthesis to form sulfhydryl group reagents, including arsenomethane. Arsenomethane then reacts with sulfhydryl groups of enzymes involved in carbon fixation and its regulation, inhibiting carbon fixation. The reduction of carbon fixation in bright light causes rapid photo-oxidative damage due to uncontrolled dissipation of absorbed light energy, ultimately resulting in cell disruption. Herbicides that cause cell disruption often reduce the absorption and/or the translocation of other herbicides applied in mixture (Croon et al. 1989; Culpepper et al. 1999; Olson and Nalewaja 1982; Pereira and Crabtree 1986). The objective of this study was to characterize absorption and translocation of glyphosate applied alone and in mixture with MSMA to determine if reduced absorption or translocation is the cause of antagonism of glyphosate with MSMA in browntop millet and Palmer amaranth.

Materials and Methods

Plant Material. Seeds of browntop millet and Palmer amaranth were planted in a 1:1 mixture of potting media[1] and soil (Bosket sandy loam, fine-loamy, mixed thermic Molic Hapludalfs) in 7-cm × 5-cm plastic pots. After emergence, plants were thinned to one per pot. Plants were grown in a growth chamber set to 32/25 C (±3 C) day/night temperature with a 14-h photoperiod (400 µmol/m^2/s) and relative humidity of about 50%. Pots were subirrigated with water as needed. Experiments were conducted separately for

---

DOI: 10.1614/WT-06-064.1

* First and third authors, Postdoctoral Research Associate and Plant Physiologist, respectively, USDA-ARS, Southern Weed Science Research Unit, 141 Experiment Station Road, P.O. Box 350, Stoneville, MS 38776; second author, Research Agronomist, USDA-ARS, Crop Genetics and Production Research Unit, 141 Experiment Station Road, P.O. Box 345, Stoneville, MS 38776; and fourth author, Professor, Crop Science Department, North Carolina State University, Raleigh, NC 27695-7620. Current address of second author: Assistant Research Professor, Delta Research and Extension Center, Mississippi State University, Stoneville, MS 38776. Corresponding author's E-mail: icburke@wsu.edu

each species in a split-plot arrangement of treatments in a randomized complete block design with three replications. The main treatments were glyphosate application with and without MSMA and subtreatments were harvest timings. The experiment for each species was repeated in time.

Uptake and Translocation. Technical-grade glyphosate (^{14}C-methyl labeled with 2.0 kBq µmole^{-1} specific activity and 99.5% radiochemical purity in an aqueous stock solution of 7.4 MBq/ml as *N*-[phosponomethyl] glycine) was used in the study. Two ^{14}C-glyphosate solutions were prepared by diluting ^{14}C-glyphosate in one solution containing a commercial formulation of glyphosate[2] to give a final concentration of 0.84 kg ae/ha and another solution containing commercial formulations of glyphosate (final concentration of 0.84 kg/ha) and MSMA (2.24 kg ai/ha) plus non-ionic surfactant[3] (0.25% v/v), all prepared in double-distilled water. Five 1-µl droplets of the treatment solutions, containing approximately 4.3 kBq of radioactivity, were placed on the adaxial surface of the second youngest leaf of browntop millet and Palmer amaranth.

Plants were harvested at 0.5, 1, 4, 24, 72, and 168 h after ^{14}C-glyphosate treatment (HAT). Treated leaves were excised and washed by shaking for 10 s in 15 ml methanol:water (1:1, v/v) and 0.25% (v/v) nonionic surfactant[4] to remove nonabsorbed glyphosate. Two 1-ml leaf wash aliquots were mixed with 15 ml scintillation fluid,[5] and radioactivity was quantified via liquid scintillation spectrometry (LSS).[6] Plants were divided into the treated leaf, roots, and foliage above and below the treated leaf. Plant parts, including washed roots, were wrapped in tissue paper,[7] dried for 72 h at 40 C, weighed, and combusted with a biological sample oxidizer.[8] The evolved $^{14}CO_2$ was trapped in 10 ml Carbosorb and 10 ml Permafluor E$^+$.[9] Radioactivity in the oxidized samples was quantified by LSS.

Data were subjected to ANOVA with the use of the repeated-measures statement in PROC GLM (SAS 1998). Log transformation slightly improved homogeneity of variance based on visual inspection of plotted residuals; therefore, data were transformed prior to ANOVA. The six harvest timings were considered within-subject effects, the herbicide treatments were considered between-subjects effects. The five plant portions of quantified radioactivity were combined as the total absorbed into the plant (the sum of the radioactivity from the treated-leaf, above-treated-leaf, below-treated-leaf, and root portions), the total radioactivity translocated out of the treated leaf (the sum of the radioactivity from the above-treated-leaf, below-treated-leaf, and root portions), the above-treated-leaf portion, and the below-treated-leaf portions (the sum of the radioactivity from the below-treated-leaf and root portions). Where within-subject main effects or interactions were significant, the Gompertz equation was used to explain the relationship of measured responses over time. The Gompertz equation used was

$$y = a \times e^{-\exp[-(x-k)/b]} \qquad [1]$$

where *y* is the percent absorption or translocation expressed as the percent of the applied, *a* is the asymptote or the maximum absorption or translocation expressed as the percent applied, *k* is the point of inflection (in hours) and *b* is the slope of the curve at the point of greatest inflection.

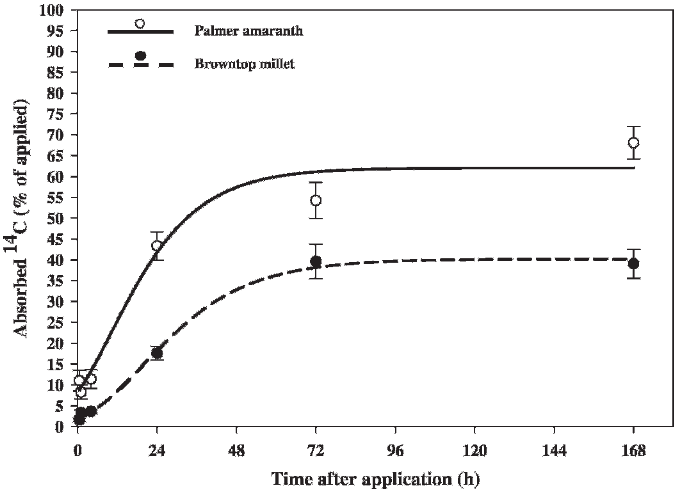

Figure 1. Foliar absorption of ^{14}C-glyphosate expressed as percent of applied in browntop millet and Palmer amaranth. Response modeled with the use of the Gompertz equation (Equation 1). See Table 1 for regression coefficients, standard errors, and R^2 values. Error bars represent the standard error of the mean.

Coefficients of determination (R^2) were calculated for all regressions. For the Gompertz equation fitted to the data, an approximate R^2 value was obtained by subtracting the ratio of residual sums of squares to corrected total sums of squares from 1 (Draper and Smith 1981). The R^2 and residual mean squares were used to determine goodness of fit to nonlinear models.

Results and Discussion

Absorption. Absorption and translocation data were averaged over experiments due to experimental effect for browntop millet or Palmer amaranth. Recovery of ^{14}C averaged 93% in both species. ANOVA indicated no difference in absorption when glyphosate was applied alone or in mixture with MSMA for either species, indicating that antagonism of glyphosate activity by MSMA was not caused by reduced absorption of glyphosate. As the treatment main effect was not significant, absorption was averaged over treatments and modeled over time with the use of the Gompertz equation (Equation 1) for each species (Figure 1). Absorption of ^{14}C-glyphosate by Palmer amaranth was rapid and increased with time from 11.1% at 0.5 HAT to 68.1% at 168 HAT (Figure 1). Absorption of ^{14}C-glyphosate by browntop millet was less than that of Palmer amaranth at each harvest interval when expressed as percent of applied. Absorption of ^{14}C-glyphosate by browntop millet ranged from 1.6% at 0.5 HAT to 39.1% at 168 HAT (Figure 1). When modeled, the trend in uptake over time was different for each species (Table 1), with time to the model inflection point of Palmer amaranth nearly half (10.4) of that of browntop millet (19.8).

The amount of ^{14}C-glyphosate absorption observed in this study is similar to that observed for other weed species. Li et al. (2005) observed 50.9 to 57.8% absorption of glyphosate at

Table 1. Regression parameters (and standard errors) for absorption and translocation of ^{14}C-glyphosate in browntop millet and Palmer amaranth.

Movement of ^{14}C-glyphosate	Plant species or plant portion	Herbicide treatment	Regression parameters (and standard errors)[a] a	b	k	R^2
Absorption into treated leaf	Browntop millet		40.2 (1.1)	17.7 (2.0)	19.8 (1.5)	0.99
	Palmer amaranth		62.1 (4.3)	14.7 (4.1)	10.4 (2.9)	0.97
Total translocation into plant	Browntop millet	Glyphosate	11.5 (0.1)	16.1 (0.7)	24.3 (0.4)	0.99
		Glyphosate + MSMA	2.1 (0.4)	16.6 (12.5)	18.9 (9.4)	0.86
	Palmer amaranth	Glyphosate	15.0 (0.6)	10.4 (1.6)	12.1 (1.6)	0.99
		Glyphosate + MSMA	3.1 (0.4)	5.8 (4.4)	16.6 (6.6)	0.94
Translocation within browntop millet	Above treated leaf	Glyphosate	0.7 (0.1)	35.9 (7.3)	38.8 (6.3)	0.99
		Glyphosate + MSMA	0.1 (0.0)	5.8 (4.2)	5.5 (2.4)	0.98
	Below treated leaf	Glyphosate	10.9 (0.1)	15.5 (1.0)	24.1 (0.5)	0.99
		Glyphosate + MSMA	2.0 (0.4)	18.0 (13.2)	19.7 (10.3)	0.82
Translocation within Palmer amaranth	Above treated leaf	Glyphosate	8.0 (0.1)	13.2 (1.2)	19.4 (0.8)	0.99
		Glyphosate + MSMA	2.5 (0.2)	8.4 (6.1)	16.4 (5.8)	0.98
	Below treated leaf	Glyphosate	7.5 (0.9)	5.8 (5.4)	6.7 (4.3)	0.93
		Glyphosate + MSMA	0.7 (0.2)	1.5 (5.3)	5.1 (5.9)	0.85

[a] The Gompertz equation used was $y = a \times e^{-\exp[-(x - k)/b]}$, where y is the percent absorption or translocation expressed as the percent of the applied, a is the asymptote or the maximum absorption or translocation expressed as the percent applied, k is the point of inflection (in hours), and b is the slope of the curve at the point of greatest inflection. R^2 represents the coefficient of determination.

74 HAT, depending on glyphosate formulation, in common waterhemp (*Amaranthus rudis* Sauer). Absorption of glyphosate in this study is also similar to that reported by Koger and Reddy (2005), who observed 44.1% absorption of glyphosate at 192 HAT in pitted morningglory (*Ipomoea lacunosa* L.). The level of absorption in this study was consistent with others, and MSMA in mixture with glyphosate did not affect the absorption of glyphosate.

Translocation. Differences in translocation of ^{14}C-glyphosate were significant for glyphosate applied with and without MSMA in browntop millet and Palmer amaranth. Translocation patterns were examined in three different ways: total translocation out of the treated leaf (sum of the radioactivity in the above the treated-leaf portion, below the treated-leaf portion, and root portion); acropetal translocation to foliage the above the treated-leaf portion; and basipetal translocation to the foliage below the treated leaf and roots.

Total translocation out of the treated leaf was similar for both browntop millet and Palmer amaranth at 0.5 and 1 HAT (Figure 2). By 4 HAT, a larger amount of glyphosate had translocated out of the treated leaf when glyphosate was applied alone. In browntop millet at 72 HAT, only 2.8% of the applied radioactivity had translocated out of the treated leaf to the rest of the plant when glyphosate was applied with MSMA compared to 10.8% when glyphosate applied alone. Translocation of glyphosate peaked at 72 HAT with no further increase at 168 HAT for both species. A similar trend was observed for Palmer amaranth, where by 24 HAT 3.2% of the applied radioactivity had translocated out of the treated leaf when glyphosate was applied with MSMA. At the same harvest interval, 10.6% of the applied glyphosate had translocated out the treated leaf when glyphosate was applied alone.

Translocation above and below the treated leaf followed different trends for each plant species. In browntop millet, more ^{14}C-glyphosate translocated below the treated leaf than

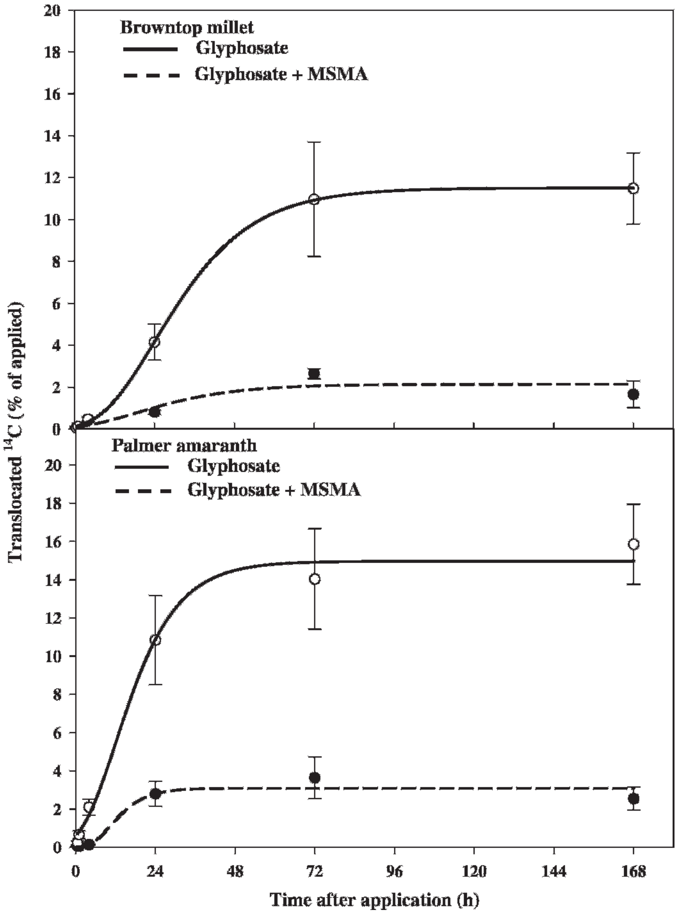

Figure 2. Translocation of ^{14}C-glyphosate (sum of the radioactivity in the above-treated-leaf, below-treated-leaf, and root portions) in browntop millet and Palmer amaranth. Responses were modeled with the use of the Gompertz equation (Equation 1). See Table 1 for regression coefficients, standard errors, and R^2 values. Error bars represent the standard error of the mean.

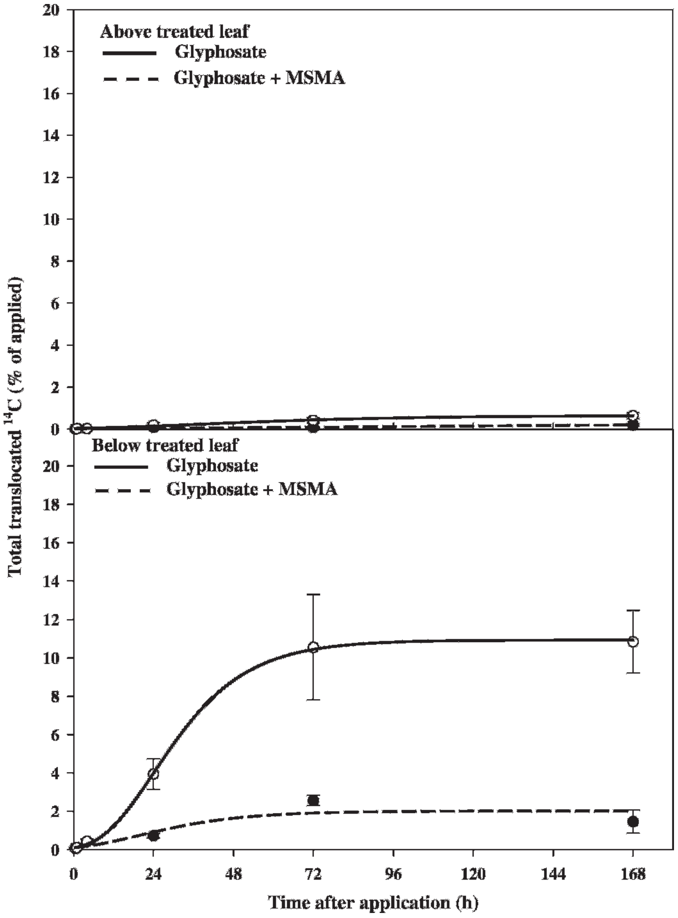

Figure 3. Translocation of ^{14}C-glyphosate above the treated leaf and below the treated leaf in browntop millet. Responses were modeled with the use of the Gompertz equation [1]. See Table 1 for regression coefficients, standard errors, and R^2 values. Error bars represent the standard error of the mean.

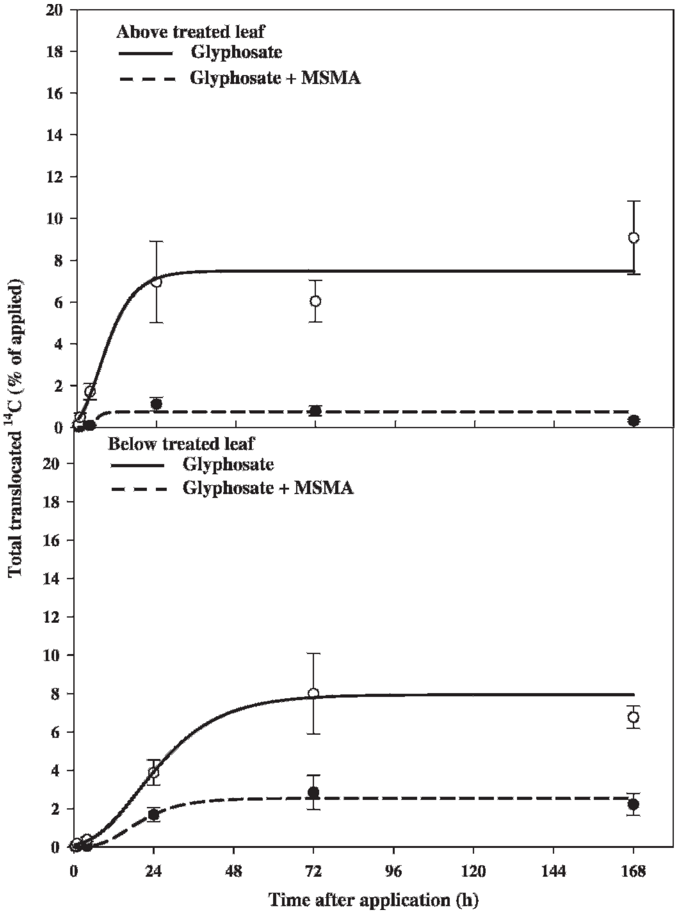

Figure 4. Translocation of ^{14}C-glyphosate above the treated leaf and below the treated leaf in Palmer amaranth. Responses were modeled with the use of the Gompertz equation (Equation 1). See Table 1 for regression coefficients, standard errors, and R^2 values. Error bars represent the standard error of the mean.

above the treated leaf (Figure 3). MSMA reduced the amount of ^{14}C-glyphosate translocated below the treated leaf, but not above the treated leaf. In browntop millet, less than 1% of applied ^{14}C glyphosate was translocated above the treated leaf, regardless of treatment (Figure 3, Table 1). Glyphosate typically translocates to actively growing meristematic regions, and in browntop millet, the meristematic region is the intercalary meristem located at the base of the whorl. When glyphosate was applied with MSMA, there was a reduction in the amount of radioactivity translocated to the intercalary meristematic region. Movement of glyphosate within a plant is related to plant growth stage, the amount of herbicide absorbed, and source–sink relationships (Dewey and Appleby 1983; Sandberg et al. 1980). Consequently, the plant portion below the treated leaf (including the roots) of browntop millet accumulated higher amounts of ^{14}C-glyphosate over time compared with the plant portion above the treated leaf, as the lower leaves, the intercalary meristem, and roots were a "sink" rather than a "source" for photosynthates. In contrast, partitioning of ^{14}C-glyphosate in Palmer amaranth was much more equitable, with similar amounts of radioactivity recovered from above the treated leaf and below the treated leaf portions, including the roots (Figure 4, Table 1). As with browntop millet, there was a considerable reduction in translocated radioactivity when MSMA was applied in mixture with glyphosate in Palmer amaranth.

MSMA causes cell disruption where it comes in contact with susceptible plant tissue (Duke 1992). Cell disruption may be the primary cause of the observed antagonism of glyphosate by MSMA in field and greenhouse trials (Koger et al. 2007). However, the magnitude of antagonism in field and greenhouse trials was not as great as what may be hypothesized based on this research. Control of browntop millet and Palmer amaranth was 74 and 77%, respectively, when glyphosate and MSMA were applied in mixture to greenhouse-grown plants at similar rates as used in this study. MSMA alone controlled browntop millet and Palmer amaranth 41 and 58%, respectively, whereas control of both weeds with glyphosate alone was 98%; suggesting that even with the extremely limited translocation observed in this study, glyphosate was still translocated in sufficient quantity to cause injury. It is likely that certain environmental conditions that favor rapid glyphosate translocation may offset antagonistic effects of MSMA. Alternatively, the amount of glyphosate translocated may be of greater biological significance when the entire plant is treated.

In conclusion, MSMA in mixture with glyphosate had no effect on absorption of glyphosate but did reduce translocation of glyphosate in browntop millet and Palmer amaranth. Reduced translocation appears to be the cause of the observed antagonism of glyphosate by MSMA.

Sources of Materials

[1] Jiffy mix, Jiffy Products of America Inc., Batavia, IL 60510.

[2] Roundup WeatherMax™, potassium salt of glyphosate, Monsanto Company, 800 North Lindbergh Boulevard, St. Louis, MO 63167.

[3] Induce® nonionic low foam wetter-spreader adjuvant contains 90% nonionic surfactant (alkylarypolyoxyalkane ether and isopropanol), free fatty acids, and 10% water, Helena Chemical Company, Suite 500, 6075 Poplar Avenue, Memphis, TN 38137.

[4] Induce® nonionic low foam wetter-spreader adjuvant contains 90% nonionic surfactant (alkylarypolyoxyalkane ether and isopropanol), free fatty acids, and 10% water, Helena Chemical Company, Suite 500, 6075 Poplar Avenue, Memphis, TN 38137.

[5] EcoLume, ICN, 3300 Hyland Avenue, Costa Mesa, CA 92626.

[6] Tri-carb 2500TR Liquid Scintillation Analyzer, Packard Instrument Company, 2200 Warrenville Road, Downers Grove, IL 60515.

[7] Kimwipes EX-L, Kimberly-Clark Corporation, 1400 Holcomb Bridge Road, Roswell, GA 30076.

[8] Packard Oxidizer 306, Packard Instruments Company, 2200 Warrenville Road, Downers Grove, IL 60515.

[9] CarboSorb E and Permafluor E$^+$, Packard Instruments Company, 2200 Warrenville Road, Downers Grove, IL 60515.

Acknowledgment

The authors thank Efren Ford for technical assistance.

Literature Cited

Anonymous. 2005. Roundup WeatherMax label. St. Louis, MO: Monsanto. 19 p.

Askew, S. D., W. A. Bailey, G. H. Scott, and J. W. Wilcut. 2002. Economic assessment of weed management for transgenic and nontransgenic cotton in tilled and nontilled systems. Weed Sci. 50:512–520.

Croon, K. A., M. L. Ketchersid, and M. G. Merkle. 1989. Effect of bentazon, imazaquin, and chlorimuron on the absorption and translocation of the methyl-ester of haloxyfop. Weed Sci. 37:645–650.

Culpepper, A. S., T. M. Webster, A. C. York, R. M. Barrentine, and B. G. Mullinix Jr. 2004. Glyphosate/MSMA mixtures in glyphosate-resistant cotton (*Gossypium hirsutum*). J. Cotton Sci. 8:124–129.

Culpepper, A. S., A. C. York, D. L. Jordan, F. T. Corbin, and Y. S. Sheldon. 1999. Basis for antagonism in mixtures of bromoxynil plus quizalofop-P applied to yellow foxtail (*Setaria glauca*). Weed Technol. 13:515–519.

Dewey, S. A. and A. P. Appleby. 1983. A comparison between glyphosate and assimilate translocation patterns in tall morningglory (*Ipomoea purpurea*). Weed Sci. 31:308–314.

Draper, N. R. and H. Smith. 1981. Applied Regression Analysis. New York: J. Wiley. Pp. 33–42.

Duke, S. O. 1992. Modes of action of herbicides used in cotton. Pages 403–437 *in* C. G. McWhorter and J. L. Abernathy, eds. Weeds of Cotton. Memphis, TN: The Cotton Foundation.

Knowles, F. C. and A. A. Benson. 1983. The biochemistry of arsenic. Trends Biochem. Sci. 8:178–180.

Koger, C. H., I. C. Burke, D. K. Miller, A. J. Kendig, K. N. Reddy, and J. W. Wilcut. 2007. MSMA antagonizes glyphosate and glufosinate efficacy on broadleaf and grass weeds. Weed Technol 21:159–165.

Koger, C. H. and K. N. Reddy. 2005. Glyphosate efficacy, absorption, and translocation in pitted morningglory (*Ipomoea lacunosa*). Weed Sci. 53:277–283.

Li, J. M., R. J. Smeda, B. A. Sellers, and W. G. Johnson. 2005. Influence of formulation and glyphosate salt on absorption and translocation in three annual weeds. Weed Sci. 53:153–159.

Monks, C. D., M. G. Patterson, J. W. Wilcut, and D. P. Delaney. 1999. Effect of pyrithiobac, MSMA, and DSMA on cotton (*Gossypium hirsutum* L.) growth and weed control. Weed Technol. 13:6–11.

Olson, W. and J. D. Nalewaja. 1982. Effect of MCPA on ^{14}C-diclofop uptake translocation. Weed Sci. 30:59–63.

Pereira, W. and G. Crabtree. 1986. Absorption, translocation, and toxicity of glyphosate and oxyfluorfen in yellow nutsedge (*Cyperus esculentus*). Weed Sci. 34:923–929.

Pline-Srnic, W. A., K. L. Edmisten, J. W. Wilcut, R. Wells, and J. L. Thomas. 2004. Effect of glyphosate on fruit retention, yield, and fiber quality of glyphosate resistant cotton. J. Cotton Sci. 8:24–32.

Porterfield, D., J. W. Wilcut, and S. D. Askew. 2002. Weed management with CGA-362622, fluometuron, and prometryn in cotton. Weed Sci. 50:642–647.

Sandberg, C. L., W. F. Meggitt, and D. Penner. 1980. Absorption, translocation and metabolism of glyphosate-C^{14} in several weed species. Weed Res. 20:195–200.

[SAS] Statistical Analysis Systems. 1998. SAS/STAT User's Guide. Release 7.00. Cary, NC: Statistical Analysis Systems Institute. 1028 p.

Received March 30, 2006, and approved May 24, 2006.