

## Effect of Soil pH on Availability of Glyphosate in Soil to Germinating Ryegrass Seedlings<sup>1</sup>

Michael K. Kawate and Arnold P. Appleby<sup>2</sup>

### ABSTRACT

Soil activity of glyphosate (*N*-(phosphonomethyl)glycine) in greenhouse studies was inconsistent and infrequent. Glyphosate applied to two sandy soils at high rates (6.7 kg ae ha<sup>-1</sup>) reduced fresh weight of Italian ryegrass (*Lolium multiflorum* Lam.), measured 4 to 6 weeks after treatment, whereas glyphosate applied to finer-textured or high-organic soils did not injure plants. On the sandy soils, as soil pH increased, glyphosate injury to Italian ryegrass increased. Freundlich adsorption isotherms confirmed that as pH increased, glyphosate adsorption decreased. The incidence of damping-off (*Pythium* sp.) symptoms was higher on plants grown in glyphosate-treated soil than on plants grown in nontreated soil. High rates of glyphosate applied to coarse-textured soils, especially at high pH levels, can damage grass seedlings that emerge within a few days after herbicide application. Increased disease incidence may intensify this damage.

*Additional index words:* interaction, adsorption, isotherm, *Pythium*, *Lolium multiflorum*.

### INTRODUCTION

Glyphosate is an effective foliar-applied herbicide. Low soil activity and low volatility allow glyphosate to be used to control weeds preplant or preemergence to many crops. Glyphosate is generally rapidly inactivated in soil (Baird et al., 1971; Egley and Williams, 1978; Hensley et al., 1978; Klingman and Murray, 1976; Sprankle and Penner, 1975a, 1975b; Torstensson and Aamisepp, 1977). Usually when soil activity occurred, an exceptionally high rate was used (Sprankle and Penner, 1975b) or seeds of bioassay species were exposed to glyphosate spray (Moshier et al., 1976; Salazar and Appleby, 1982b; Segura et al., 1978). However, soil activity of glyphosate at

normal use rates has been reported. Brewster and Appleby (1972) reported preemergence glyphosate activity on wheat (*Triticum aestivum* L.) at 1.7 kg ae ha<sup>-1</sup> and suggested that glyphosate was not immediately inactivated, particularly when soil was moist. Dry weight of bentgrass (*Agrostis tenuis* Sibth. 'Highland') foliage was reduced when glyphosate was applied preemergence at 3.4 kg ae ha<sup>-1</sup> (Salazar and Appleby, 1982a). Nodule production was inhibited and root weight was reduced in subterranean clover (*Trifolium subterraneum* L.) planted 120 days after glyphosate was applied to a sandy loam at 2 mg g<sup>-1</sup> soil (Dao and Lavy, 1978). Glyphosate at 2.2 or 4.4 kg ha<sup>-1</sup> applied preemergence to a sandy loam soil reduced stand counts and fresh weights of tomato (*Lycopersicon esculentum* Mill.), redroot pigweed (*Amaranthus retroflexus* L.), and large crabgrass (*Digitaria sanguinalis* (L.) Scop.) (Boldt and Putnam, 1978). Marigold (*Tagetes* sp.) was injured when glyphosate at 2.2 kg ha<sup>-1</sup> was applied to a medium of peat, perlite, and vermiculite (Ahrens and Walton, 1986). Glyphosate at 4 kg ha<sup>-1</sup> on a peat soil reduced germination and dry weight of 'Tama' Italian ryegrass (*Lolium multiflorum* Lam.) (Rahman and Nishimoto, 1985). Soil pH may influence glyphosate availability similar to the way it affects phosphorus availability. Glyphosate may be bound to soil by the phosphonic acid moiety (Hance, 1976; Normura and Hilton, 1977, Sprankle and Penner, 1975a). As soil pH increased, glyphosate adsorption decreased (McConnell and Hossner, 1985). Sprankle and Penner (1975a) showed a similar response to soil pH, but concluded that the soil phosphorus level was more important.

Oregon seed growers have reported injury to grasses in fields treated with glyphosate shortly before grass emergence. The objective of these studies was to study further the effect of soil pH on glyphosate availability in several soils.

### MATERIALS AND METHODS

The five Oregon soils used were: (a) Woodburn silty clay loam (fine-silty, mixed mesic Aquultic Argixeroll); (b) Chehalis sandy loam (fine-silty, mixed mesic Cumulic Ultic Haploxeroll); (c and d) Semiahmoo muck (Euic

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<sup>2</sup>Former graduate research assistant and professor of crop science, Crop Sci. Dep., Oregon State Univ., Corvallis, OR 97331. Present address of first author: Univ. of Hawaii at Manoa, Department of Agricultural Biochemistry, 1800 East-West Road, Heneke 329, Honolulu, HI 96822

Table 1. Selected chemical and physical properties of five Oregon soils.

Soil <sup>a</sup>	pH	P	K	Ca	Mg	Na	CEC <sup>b</sup>	Organic matter	Particle Size		
									Sand	Silt	Clay
		------(cmol kg <sup>-1</sup> )-----					(cmol(p+) kg <sup>-1</sup> )	------(g g <sup>-1</sup> )-----			
Woodburn sicl	4.6	0.64	0.7	3.2	0.6	0.11	13.2	0.02	0.16	0.51	0.33
Chehalis sl	6.0	0.02	0.3	6.4	2.2	0.26	19.5	0.02	0.57	0.24	0.19
Semiahmoo muck no. 1	5.2	0.49	1.5	16.2	2.4	0.12	59.1	0.24			
Semiahmoo muck no. 2	4.9	0.61	0.7	20.6	2.3	0.12	68.5	0.45			
Crooked sl	8.2	0.07	1.6	4.8	2.0	0.62	13.7	0.01	0.70	0.13	0.17

<sup>a</sup>sicl = silty clay loam; sl = sandy loam.

<sup>b</sup>CEC = cation exchange capacity.

mesic, Typic Medisaprist), from two locations; and (e) Crooked sandy loam (loamy, mixed mesic, shallow Zeric Durothid)(Table 1).

**Plant response.** In the first experiment, three soils, and in the second experiment, five soils were treated with calcium hydroxide or sulfuric acid to create a range of pH from 4.9 to 8.8. Soils were placed in separate plastic bags and saturated with water. After 6 weeks, the soils were placed in 7.5- by 7.5-cm pots and 10 Italian ryegrass seeds were planted per pot. After subirrigating, glyphosate was sprayed on the soil in 280 l of water ha<sup>-1</sup> at 210 kPa using an 80015-E nozzle on a track-mounted sprayer.

Pots were kept in the greenhouse with 21 C/16 C day/night temperatures. No supplemental light was used and water was added by subirrigation. The experiments were randomized complete block designs with five replications. Plants were observed in both experiments for symptoms of chemical injury and other secondary effects. Fresh weights of Italian ryegrass foliage were recorded 4 (first experiment) or 6 (second experiment) weeks after treatment.

**Soil adsorption.** The two sandy loam soils, adjusted for pH from the second experiment, were used for adsorption studies. Methyl <sup>14</sup>C-labelled glyphosate acid with specific activity of 426 kBq mg<sup>-1</sup> was formulated as the isopropylamine salt to obtain an activity of 0.925 kBq/ml. Air-dried soil, 2.5 g, was weighed into 10-ml beakers. Eight concentrations of nonradioactive glyphosate, ranging from 2.5 x 10<sup>-6</sup> to 1.25 x 10<sup>-2</sup> g ml<sup>-1</sup> were formulated as the isopropylamine salt (without surfactant). One ml of each solution was placed in 10-ml test tubes, and 1 ul of radiolabelled glyphosate was added to each test tube to obtain the same level of radioactivity

in each sample. Additional water was added to each test tube to obtain at least 0.5 ml of soil solution after centrifugation (Chehalis, 0.2 ml; Crooked, 0.1 ml). The radioactive solution in each test tube was poured into the beaker containing air-dried soil and the treated soil was equilibrated for 24 h in 100% relative humidity chambers. Kinetic studies showed that glyphosate adsorption was complete 1 h after treatment.

The centrifugation method of Dao and Lavy (1978) was modified and used to extract soil solution. The modified apparatus consisted of a 10-ml plastic syringe instead of a small test tube. Glass wool was placed at the bottom of the syringe to cover the opening. A 13-mm section of 4.8-mm (inner diameter) polyethylene tubing was forced into the tip of the syringe. The syringe was then placed in a 30-ml glass centrifuge tube (25-mm outer diameter by 100-mm length). Treated soil was placed in the syringe. The apparatus was centrifuged at 2000 g for 20 min. A 0.5-ml sample of soil solution was placed into a scintillation vial containing 15 ml of scintillation solution and assayed for radioactivity using a liquid scintillation counter. The scintillation solution consisted of 0.1 g POPOP (1,4-bis(5-phenyloxazol-2-yl)-benzene), 5.0 g PPO (2,5-phenyloxazole), 400 ml 2-methoxyethanol, and 600 ml toluene. Preliminary studies determined that the apparatus (without soil) did not adsorb glyphosate. Treatments had two replications and the experiment was repeated.

The data were fitted to the Freundlich equation,  $\log x/m = \log K + (1/n)\log C_{eq}$ , where  $C_{eq}$  = equilibrium concentration of glyphosate (mg/ml) and  $1/n$  = linear constant. Regression analysis was performed and log K values were determined at  $\log C_{eq} = 0$  ug/ml.

**RESULTS AND DISCUSSION**

**Plant response.** Grass seedlings emerged 5 days after planting in both experiments. Differences were greater and more significant in the second experiment than in the first, but the general conclusions were similar. Glyphosate activity in soil was inconsistent and infrequent. Glyphosate did not injure Italian ryegrass in the silty clay or high-organic soils at any pH (data not

shown). However, Italian ryegrass seedlings were injured in the sandy loams, and injury was greatest at the higher pH levels (Table 2). Glyphosate injured Italian ryegrass only at high rates, such as those that would be used in spot applications.

**Soil adsorption.** Laboratory adsorption studies supported the plant response studies. As soil pH increased, Freundlich K values decreased significantly in both sandy loam soils (Table 3). This generally agrees with the results of McConnell and Hossner (1985).

**Table 2. Fresh weight of Italian ryegrass foliage grown in glyphosate-treated Chehalis or Crooked soils at several pH levels.<sup>a</sup>**

Soil	pH	Glyphosate rate (kg ae ha <sup>-1</sup> )					
		0.0		3.4		6.7	
		(g pot <sup>-1</sup> )	(% of ck)	(g pot <sup>-1</sup> )	(% of ck)	(g pot <sup>-1</sup> )	(% of ck)
Chehalis	5.7	2.0	100	1.9	93	1.6	78
	7.4	3.1	100	3.0	96	3.1	100
	8.0	3.8	100	3.6	94	3.1	83
	8.4 <sup>b</sup>	4.6	100	4.4	96	2.8	60
	Mean <sup>b</sup>	3.4	100	3.2	95	2.7	78
Crooked	5.0	2.6	100	2.6	99	2.2	83
	6.3	2.5	100	2.2	89	2.1	85
	7.3 <sup>b</sup>	2.7	100	2.3	86	0.9	32
	7.9 <sup>b</sup>	2.1	100	1.3	64	0.5	22
	Mean <sup>b</sup>	2.5	100	2.1	86	1.4	57

<sup>a</sup>Means of five replications.

<sup>b</sup>Significant (P = 0.05) glyphosate effect as measured by regression analysis.

**Table 3. Freundlich isotherm constants for glyphosate in two Oregon soils at several pH levels.**

Soil	pH	Freundlich K	Log K ± 95% C.I.	1/n	R <sup>2</sup>
Chehalis	5.7	112	2.05 ± 0.09	0.52	0.95
	7.4	30	1.48 ± 0.06	0.68	0.98
	8.0	32	1.51 ± 0.07	0.66	0.97
	8.4	16	1.19 ± 0.05	0.72	0.99
Crooked	5.0	23	1.37 ± 0.07	0.64	0.97
	6.3	18	1.25 ± 0.05	0.67	0.94
	7.3	9	0.94 ± 0.05	0.70	0.99
	7.9	6	0.81 ± 0.13	0.67	0.94

**Pythium interaction.** Injury symptoms on Italian ryegrass grown in the two sandy loam soils were not the same. Italian ryegrass plants in the Crooked soil treated with glyphosate were severely stunted. Leaves were streaked and the base of the stem swelled. These symptoms were typical of a chemical response. In contrast, lower stem tissue of injured plants in glyphosate-treated Chehalis soil was necrotic, and plants apparently died from severe water stress. These symptoms were typical of disease injury. The Plant Disease Clinic at Oregon State University identified a *Pythium* sp. as the causal organism. Incidence of damping-off symptoms were recorded at harvest.

Disease symptoms occurred more frequently on plants grown in glyphosate-treated soil (Table 4). In other studies (Holliday and Keen, 1982; Keen et al., 1982), a sublethal concentration of glyphosate caused a soybean variety (*Glycine max* (L.) Merr. 'Harosoy 63'), resistant to a particular race of *Phytophthora megasperma* f. sp. *glycinea*, to become susceptible to that race. Glyphosate interfered with glyceollin (a phytoalexin) production, which was partially responsible for disease resistance. Johal and Rahe (1984) reported that glyphosate at 10 mg plant<sup>-1</sup> killed bean (*Phaseolus vulgaris* L.) seedlings in nonsterilized soil, but not in sterilized soil. Nontreated seedlings in nonsterilized soil were healthy. Plants evidently died from combined injury from glyphosate and root pathogenic fungi (*Pythium* and *Fusarium*).

Our results suggest that Italian ryegrass grown in Chehalis soil was injured because of an interaction between glyphosate and *Pythium*. Glyphosate may have predisposed the plants to *Pythium* injury by suppressing their defense mechanism(s) (e.g., phytoalexin production), but this was not studied.

**Table 4. Incidence of damping-off symptoms on Italian ryegrass plants grown in glyphosate-treated Chehalis soil at several pH levels.**

Soil pH	Glyphosate rate (kg ae ha <sup>-1</sup> )		
	0.0	3.4	6.7
	------(infected plants pot <sup>-1</sup> )-----		
5.7 <sup>b</sup>	0.2	0.0	2.0
7.4	0.2	0.0	0.4
8.0	0.4	0.8	1.0
8.4	0.0	0.2	0.6
Mean <sup>b</sup>	0.2	0.3	1.3

<sup>a</sup>Means of five replications.

<sup>b</sup>Significant (P = 0.05) glyphosate effect as measured by regression analysis.

## CONCLUSIONS

Glyphosate occasionally can injure plants from soil uptake, especially at high rates in sandy, high-pH soils. Increased incidence of damping-off symptoms on Italian ryegrass plants growing in glyphosate-treated soil suggests that sublethal concentrations of glyphosate in soil could predispose plants to infection by pathogens. This may be important to seed growers who use glyphosate to control weeds after grass seed crops are planted but before they emerge. Application of fungicide to the seed may help reduce injury, but this was not studied in this paper. We suggest that glyphosate rates be kept as low as possible and application be made no less than 1 week before grass emergence is anticipated.

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## Improvement of Regar Meadow Bromegrass Seed Yield by Residue Removal and Nitrogen Fertilization<sup>1</sup>

L.E. Wiesner, L.E. Welty and S.F. Upton<sup>2</sup>

### ABSTRACT

Seed yields of 'Regar' meadow bromegrass (*Bromus biebersteinii* Roem and Schult) are high the first year of production and decline rapidly in subsequent years. This study was conducted to evaluate the effects of timing of nitrogen (N) fertilization and residue removal on Regar seed yield. Experiments

were conducted at Bozeman and Kalispell, Montana. Treatments consisted of four N application dates during the seeding year, four N applications to the established stands, and four dates of residue removal from established stands. Seeding year fall N fertilization increased first year seed yield at Kalispell, but not at Bozeman. The most economical stand life of Regar at each location was two years. Fall N application and fall residue removal increased first and second year seed yield at Kalispell and second year seed yield at Bozeman. Third and fourth year seed yield at Kalispell and third year yield at Bozeman was improved with N applied after harvest. However, these yields were low compared to first and second year seed yield.

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<sup>2</sup>Professor of Agronomy, Associate Professor of Agronomy, and Graduate Research Assistant, respectively, Plant and Soil Sci. Dep., Montana State Univ., Bozeman, MT 59717.

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