MECHANISMS OF SELECTIVITY OF PENDIMETHALIN (PROWL®) AND TRIFLURALIN (TREFLAN®) IN COTTON (*GOSSYPIUM HIRSUTUM*) AND WEEDS

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Abstract

The uptake, translocation, and metabolism of pendimethalin (PROWL®) and trifluralin(TREFLAN®) in cotton, (*Gossypium hirsutum*) redroot pigweed (*Amaranthus retroflexus*) and seedling johnsongrass (*Sorghum halepense*) were determined. The better cotton selectivity of pendimethalin versus trifluralin appears to be due to differences in metabolism and sequestration of pendimethalin in the lysigenous glands. Cotton roots metabolized pendimethalin but did not metabolize trifluralin. Redroot pigweed and johnsongrass were also able to metabolize pendimethalin, but only pigweed metabolized trifluralin. However these two weeds species absorbed 1.3 and 2 times more pendimethalin than trifluralin, respectively. This increased uptake compensated for increased metabolism such that the internal concentration of pendimethalin was much higher the concentration of trifluralin. This higher uptake of pendimethalin occurred in spite of 1.6 to 2 times higher adsorption of pendimethalin than trifluralin to the soil.

Introduction

Pendimethalin and trifluralin are two of the most widely used dinitroaniline herbicides for control of annual grass and small-seeded broadleaf weeds in cotton (*Gossypium hirsutum* L.) and soybean (*Glycine max* (L.) Merr), among other crops (Weed Sci. Soc. Am., 1995). These two herbicides control most weeds at equivalent rates when applied PPI. However there are differences in their selectivity on crops, particularly on cotton lateral root development (Jordan et al. 1978; Mitchell and Bourland, 1986; Bailey and Bourland, 1986). The reasons for this difference in selectivity of pendimethalin versus trifluralin in cotton is not clear.

The objectives of this research were to determine the potential mechanisms of selectivity of pendimethalin and trifluralin by comparing the soil bioavailability, uptake, and plant metabolism of pendimethalin and trifluralin in cotton, johnsongrass (*Sorghum halapense* (L.) Pers.) (SORHA) and redroot pigweed (*Amaranthus retroflexus* L.) (AMARE). The results show that johnsongrass and pigweed seedlings absorb more pendimethalin from the soil than trifluralin, even though pendimethalin is bound more tightly to the soil. In cotton, both pendimethalin and trifluralin are sequestered in the lysigenous glands, but only pendimethalin is metabolized to a non-toxic metabolite.

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Materials and Methods

Greenhouse Studies

Pendimethalin and trifluralin were applied at 1.12 kg ha^{-1} to Sassafras sandy loam soil (fine-loamy, siliceous, mesic Typic Hapludults) in greenhouse flats with a belt sprayer. The treated soil was placed in plastic bags and shaken to incorporate, and poured into individual cells (5 by 4 by 4 cm deep) of greenhouse flats. Flats with untreated soils were also included. Three cotton seeds (cultivars Delta and Pine Land (DPL) 20, 50, 90 and Paymaster HS-26, and DPL 90gl, a glandless isoline of DPL 90) were planted into the cells, one cell for each cultivar. Additionally, seeds of johnsongrass, redroot pigweed, prickly sida (*Sida spinosa* L.) (SIDSP), barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.) (ECHCG), common lambsquarters (*Chenopodium album* L.) (CHEAL), and entireleaf morningglory (*Ipomoea hederacea* var. *integruiscula* Gray) (IPOHG) were planted into treated soil in individual cells. The flats were placed in a greenhouse set at 14/10-hour light/dark cycle and 30 C day and 18 C night temperature. Flats were watered daily. The experiment was a randomized complete block design with four replications and was run twice. Species/cultivars were randomized within each flat. Control flats with no herbicide were included. Percent visual injury with 0 = no injury and 100 = complete kill were recorded for the weeds 19 days after treatment (DAT). For cotton, soil was washed from the roots 23 DAT and root dry weight determined after drying overnight in an oven.

Soil Adsorption Studies

Adsorption of pendimethalin and trifluralin in soils was studied using the batch-equilibrium technique with 10 mL of herbicide solution (in 0.01 M $CaCl_2$) and 1 g of soil. ^{14}C-labeled pendimethalin and trifluralin were used at a concentration of 0.5 *u*g g^{-1} soil. This is equivalent to a field use rate of 1000 g ha^{-1}. Soils that were tested included Sassafras sandy loam, Tippecanoe silt loam (fine-loamy, mixed, mesic Typic Argiudolls), Plano loam (fine-silty, mixed, mesic Typic Argiudolls), and Beardon clay loam (fine-silty, frigid, Aeric Calciaquolls). Each soil by herbicide combination had 3 reps and the experiment was run twice. The distribution coefficient (Kd), the ratio of the herbicide concentration in soil to the concentration in solution, was calculated for each herbicide on each soil by:

$$Kd = (ug \text{ herbicide/g soil})/(ug \text{ herbicide/ml solution})$$

The amount in the soil was assumed to be the difference between the radioactivity in the solution with no soil and the radioactivity in the solutions with soil.

Plant Bioavailability Studies

Bioavailability of pendimethalin and trifluralin to cotton, redroot pigweed, and johnsongrass was determined in Sassafras sandy loam and Plano loam soils. A preliminary study with nonradioactive compounds showed the weed species could grow without visible injury symptoms in 0.125 ppm herbicide (data not shown). Separate 25-g soil samples were treated with this rate of ^{14}C-labelled pendimethalin and ^{14}C-labelled trifluralin at 0.125 ppm. The herbicides were mixed with the soil and the soil placed in 2 by 2-cm by 5 cm deep pots. Cotton (3 seeds), redroot pigweed (approximately 20 seeds), and johnsongrass (approximately 15 seeds) were placed on the soil surface and gently pressed into the soil. A small amount of vermiculite was spread on the soil surface to help preserve moisture. Each cell had a paper wick that allowed for constant wetting of the soil from the bottom. The plants were placed in a growth chamber set on a 16/8-hour light/dark cycle with day temperature of 27 C and night temperature of 20 C for 10 days. The soil was then washed from the roots of all plants and the plants divided into roots and shoots. The tissues were dried in an oven, weighed, and ^{14}C present determined by combustion followed by liquid scintillation counting (LSC). The experiment had three replications and was conducted three times.

Metabolism Studies

The metabolism of pendimethalin and trifluralin in cotton, redroot pigweed, and johnsongrass was studied using plants germinated and grown in paper towels treated with ^{14}C-labeled herbicides. At the end of the exposure period the plants were divided into roots and shoots and the tissues were extracted by homogenizing 2 to 3 min. in a mixture of methanol, acetone, and water (1:1:1 by vol) at a ratio of 10 ml solvent per g of tissue fresh weight. The homogenate was centrifuged 20 min. at 600 x *g*, and the pellet rehomogenized with more extraction solvent and centrifuged two more times. Radioactivity in the pellet was quantified by combusting the pellet samples and LSC analysis to determine bound radioactivity. This method extracted greater than 90% of the radioactivity from the plants. The supernatants were combined and solvent removed to near dryness. The residue was resuspended in 4 to 7 mL of the extraction solvent and centrifuged. The supernatants were filtered through 0.45-*u*m membrane filters and aliquots subjected to reverse-phase HPLC for separation of parent compounds and metabolites. The distribution of radioactivity in each peak was calculated as the percent of the total recovered radioactivity for each injection. Cochromatography of authentic pendimethalin and trifluralin reference standards was used to identify the radioactive peaks corresponding to the parent compounds.

Results and Discussion

Greenhouse Studies

There were no differences between the herbicides in percent control of any of the weed species (Table 1). Both compounds controlled seedling johnsongrass and barnyardgrass at least 90%, and were slightly weaker on redroot pigweed and common lambsquarters. Neither compound controlled prickly sida or entireleaf morningglory. These data show pendimethalin and trifluralin provide similar weed control at equal rates when both are used preplant incorporated.

Analysis of variance of cotton root dry weight showed no significant cultivar by herbicide interaction, and the cultivar main effect was not significant. This shows that there was no difference between cultivars in their response to each herbicide. Averaged over cultivars, the herbicide factor was significant. Trifluralin reduced cotton root dry weight by 50% compared to untreated soil, while there was no effect from pendimethalin (Table 2). Mitchell and Bourland (1986) found pendimethalin and trifluralin reduced cotton lateral roots by 17 and 35%, respectively. They also found trifluralin reduced the number of lateral roots, while pendimethalin had no effect. Anderson et al. (1967) reported trifluralin inhibited cotton lateral root production in the treated soil zone and also caused plant stunting.

Soil Adsorption Studies

Analysis of variance showed no interactions between run, herbicide, and soil, so data for the two runs were analyzed combined. There was no significant soil by herbicide interaction for adsorption. However, the soil and herbicide main effects were significant. Averaged over soils, pendimethalin adsorption was 67% higher than trifluralin adsorption (Table 3). Adsorption of both herbicides was high, with Kds of at least 25, and generally increased as soil clay and organic matter content increased. This has been observed by other researchers with these compounds (Cooper et al., 1994; Hollist and Foy, 1971; Pedersen et al., 1995; Peter and Weber, 1985; Wheeler et al., 1979). Pedersen et al. (1995) also observed higher adsorption of pendimethalin than trifluralin.

Plant Bioavailability Studies

There was no difference between cotton cultivars in their response to each herbicide (data not shown). Therefore, data for each experiment were averaged over cultivar for analysis. There were no significant ($P = 0.05$) interactions between herbicide, soil, or species, so data were combined over soils. The main effects of herbicide and species were highly significant ($P < 0.01$). Averaged over species, more pendimethalin was found in the plants than trifluralin (Figure 1). However, the differences were minor for cotton. Redroot pigweed and seedling johnsongrass absorbed 1.3 and 2.0 times more pendimethalin than trifluralin, respectively. This occurred in spite of the higher pendimethalin adsorption to soils compared to trifluralin (Table 3). Thus, although there was less pendimethalin in the unbound, plant-available pool than trifluralin, the more efficient absorption of pendimethalin resulted in higher concentrations in the plant. Averaged over herbicide, redroot pigweed and seedling johnsongrass absorbed 2.0

and 1.5 times more herbicide than cotton, respectively (Figure 1). These differences were mostly accounted for by pendimethalin, especially for johnsongrass.

Most cotton plants contain numerous lysigenous glands in their seeds, stems, and leaves. These glands play a major role in herbicide selectivity by sequestering herbicides so that they are not available for phytotoxicity. For example, norflurazon (Strang and Rogers, 1975), diuron (Strang and Rogers, 1971), and prometryn (Foster et al., 1994) are sequestered in lysigenous glands of cotton. Glandless cotton lines are not tolerant to these herbicides because the herbicides are not sequestered and are available to inhibit plant processes. Whole plant autoradiography of cotton seedlings germinated in the presence of the radiolabelled pendimethalin or trifluralin clearly demonstrated the sequestration of these highly lipophilic compounds in the lysigenous glands of cotton. It appeared, based on the intensity of the exposure that sequestration of pendimethalin in these lysigenous glands was much more pronounce compared to trifluralin exposed seedlings (data not shown).

Metabolism

Pendimethalin was metabolized to at least two compounds that are more polar than the parent in roots and shoots of cotton (Table 4). Only parent was recovered from trifluralin-treated cotton roots and shoots. This high (approximately 30-40%) metabolism of pendimethalin explains the higher tolerance of cotton to pendimethalin than trifluralin. Results were similar for johnsongrass, in which 20% of pendimethalin was metabolized in 6 d while there was no metabolism of trifluralin (Table 4). Redroot pigweed metabolized both herbicides to a similar extent (Table 4).

Conclusions

The bioavailability studies (Figure 1) showed equal uptake of pendimethalin and trifluralin in cotton. Thus, the higher tolerance of cotton to pendimethalin than trifluralin may be because cotton can detoxify pendimethalin, so that there is less pendimethalin present in the plant to cause inhibition. While seedling johnsongrass could metabolize pendimethalin, the higher uptake of pendimethalin from soil (Figure 1) may compensate for the loss due to metabolism, so that control is equal to trifluralin. Redroot pigweed can metabolize both compounds, and, although there was a slightly higher concentration of pendimethalin than trifluralin in redroot pigweed, the differences were minor. Thus, approximately equal amounts of compound should be present in redroot pigweed for control.

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Table 1. Percent control of weeds from pendimethalin and trifluralin applied preplant incorporated at 1.12 kg ha^{-1} 19 DAT.

Weed Species	Weed Control (%) Herbicide	
	Pendimethalin	Trifluralin
SORHA	90	95
AMARE	73	84
SIDSP	45	21
ECHCG	96	92
CHEAL	91	75
IPOHG	32	19

LSD (0.05) species means = 16

LSD (0.05) herbicide means within species = NS

Table 2. Effect of pendimethalin and trifluralin applied preplant incorporated at 1.12 kg ha^{-1} on cotton root dry weight.

	Dry Wt (g)
Treatment	Root
Control	0.12
Pendimethalin	0.10
Trifluralin	0.06
LSD (0.05)	0.03

Table 3. Adsorption of pendimethalin and trifluralin in soil.

	Adsorption (Kd)	
Soil	Pendimethalin	Trifluralin
Sassafras	51.4	32.6
Tippecanoe	44.8	26.9
Plano	70.2	35.8
Beardon	98.3	63.0
Herbicide Avg.	66.2	39.6

LSD (0.05) to compare herbicide averages = 6.5

Table 4. Rate of metabolism of pendimethalin and trifluralin in cotton roots and shoots.

	Percent of radioactivity recovered as parent compound[a]		
Herbicide	Cotton	SORHA	AMARE
Pendimethalin	64	78	20
Trifluralin	99	100	20

[a]Cotton was germinated and grown in the presence of 2.5 ppm of the herbicides for 5 days. SORHA and AMRE were germinated and grown in the presence of 0.5 ppm of the herbicides for 6 days.

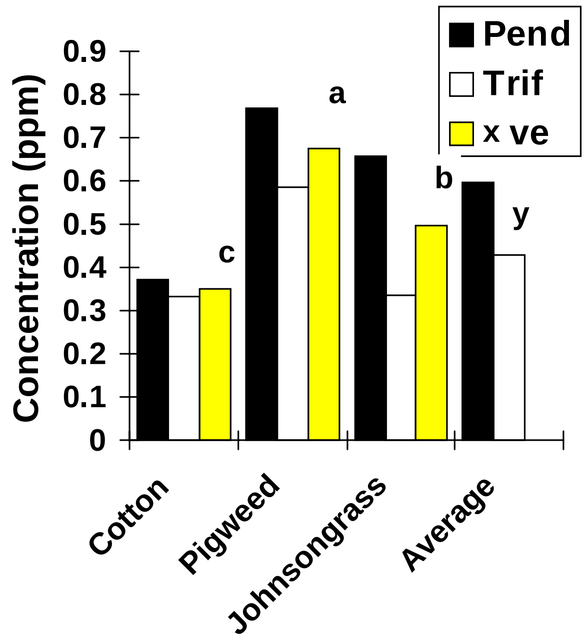

Figure 1. Pendimethalin and trifluralin concentration in plants after exposure to ^{14}C- labeled herbicide for 10 days. Herbicide*Species = NS; LSD (0.05) (species averages) = 0.13; LSD (0.05) (herbicide averages) = 0.12