Differential Tolerance of Cotton Cultivars to Prometryn

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Abstract

The tolerance of cotton cultivars to the herbicide prometryn was determined in greenhouse tests. Prometryn was applied preemergence from 0.3 to 12.0 lbs. a.i./A and postemergence from 1.2 to 12.0 lbs. a.i./A. Upland cultivars were very susceptible to injury from prometryn applied preemergence; whereas Pima S-6, Pima S-7, and Acala 1517-75 were tolerant. Postemergence treatments of prometryn applied two weeks after planting were less injurious to Upland cultivars than preemergence treatments, however, differential tolerance between Pima and Upland cultivars was evident at the high rates of application.

Introduction

Prometryn, the active ingredient in Caparol herbicide, is registered for use in cotton, celery and pigeon pea (1), although it may cause injury in cotton. The potential for injury is apparent from the precautions listed on the label that warn against use on glandless cotton, contact with the foliage or application to sand or loamy sand soils (1). Prometryn injury may occur when soil conditions favor leaching or volatilization, such as on the coarse-textured, low organic matter soils found in the arid, hyperthermic cotton growing regions of the southwest. The vapor pressure increases nearly 200-fold at temperatures between 10 and 50°C, that are readily obtained in the desert (2). Leaf injury may occur from the adsorption of prometryn vapors. Intervenal chlorosis results from vapor injury whereas venal chlorosis occurs when prometryn leaches into the root zone and is absorbed and transported to the leaves (3).

Recently, several cotton cultivars were found to differ in sensitivity to prometryn, but were equally susceptible to fluometuron and metolachlor applied at similar rates (4). Pima S-7, Pima S-6 (*G. barbadense*), and Acala 1517-75 (*G. hirsutum*) cotton cultivars had slight to moderate injury at high rates of prometryn (5.2 and 13.4 kg ha^{-1}) whereas fifteen upland cultivars, such as DP 5415 (*G. hirsutum*) died when exposed to rates between 0.6 and 1.3 kg ha^{-1} (5). The differential tolerance, observed between Pima S-7 and DP 5415, was not due to differences in the relative rates of herbicide absorption, translocation, metabolism, or the efficacy in inhibition of photosynthetic electron transport (5).

Tolerance to prometryn has been partially attributed to the presence of lysigenous glands, which may sequester prometryn and prevent it from reaching the active site (6-9). Autoradiographs of cotton treated with ^{14}C-prometryn showed prometryn accumulation in lysigenous glands (6-8). In addition, the prometryn tolerance of glanded and glandless isogenic lines of cotton was found to be directly associated with lysigenous gland density (9). The glanded cultivars of Empire 61 (WR), Westburn M, Delcot 277, TH149 and Stoneville 213 cotton were more tolerant to prometryn than their glandless counterparts (9). Plants with higher gland densities had higher photosynthetic rates, and reduction in photosynthesis was less and of shorter duration in glanded isolines than in their glandless counterparts (9). Whether the glands of tolerant cultivars were able to sequester more prometryn than susceptible cultivars has not been determined, although gland density or differential sequestration of prometryn into glands may be a possible mechanism for the tolerance. These processes have not been adequately described for prometryn. Although the tolerance of cotton to prometryn is enhanced by the presence and density of lysigenous glands, there may be other inherent differences among cultivars to account for the tolerance. The tolerance of cotton to atrazine and propazine, herbicides not registered for cotton, differed markedly between cultivars (10). Acala 4-42 glanded and glandless cultivars were equally tolerant to prometryn suggesting that the role of glands may be cultivar specific (11). Differential translocation, but not absorption and metabolism, accounted for the tolerance observed between cotton and soybean (7). The purpose of this report is to describe the susceptibility of cotton cultivars to prometryn and to further characterize the fate of prometryn in these cultivars.

Materials and Methods

Plant material. Plants for herbicide bioassays and thylakoid isolation were grown in 4" pots in the green house at a day/night regime 35°C /25°C and a photoperiod of 14 hrs. The light intensity exceeded 800 $\mu E\ m^{2}\ s^{1}$ daily. Plants were fertilized weekly with a 0.4% solution of Peat-lite Special 20-10-20 (Grace-Sierra Horticultural Product Company, Milpitas, CA) after seedlings had emerged. Seeds were planted atop of soil mixture consisting of equal parts by volume of peat, vermiculite, sand, and pima clay loam soil. The seeds were covered with 140 g of a pima clay loam/sand mixture (1:1), an amount to provide a soil depth of 12 mm. To evaluate the effects of prometryn on seedling growth, prometryn was added in methanol to the soil/sand mixture to achieve the rates of 0.3 to 12.0 lbs. a. i./A. (see Figure 1). After the methanol had evaporated, the prometryn-treated soil was thoroughly mixed and applied over the seed. Pots were irrigated from above to activate the herbicide. The effects of prometryn were determined at 5 WAT by measuring reduction in growth. Postemergence applications of prometryn were made to 2-week-old seedlings and evaluated 3 WAT. Acala 1517-75 seed was obtained from Dr. Roy Cantrell, New Mexico State University.

Gland number. To determine if gland numbers differed between cultivars, glands on cotyledon discs and hypocotyl sections were counted. Seedlings were grown until the cotyledons were fully expanded (11 days). Gland number was determined from 8 mm diameter cotyledon discs and 1 cm long hypocotyl segments cut at 1 cm below the cotyledons using a dissecting microscope.

^{14}C-prometryn in Glands. To determine whether the lysigenous glands actually sequestered more ^{14}C-prometryn than non glanded regions of the hypocotyl, sections of the hypocotyl with and without glands were excised and the amount of ^{14}C-prometryn was determined. In order to determine whether ^{14}C-prometryn partitioned into the glands, the glands of seedlings were excised and counted for radioactivity. Seedlings (DP 5415, Pima S-7, Acala 2711 glandless) were grown in vermiculite and transferred to hydroponic culture in 50% Hoaglands solution. After 24 hrs, 0.1 μCi of ^{14}C-prometryn (specific activity 30.7μCi/mg) was added to the incubation solution. After 48 hrs, hypocotyls were removed, tissues of the epidermis and cortex containing glands were excised. Tissue samples of equivalent size without glands were also excised. The tissue samples were homogenized in methanol and clarified by centrifugation. The supernatants were withdrawn and ^{14}C-prometryn was determined by scintillaton spectrometry.

To delineate whether there was differential uptake and translocation of prometryn into roots, stems, leaves and lysigenious glands, autoradiographs were prepared of the seedlings that had been incubated for 48 hours with 0.1 μCi ^{14}C-prometryn in hydroponic culture. The roots of the seedlings were washed with a methanol:water (1:1) solution, and further washed with deionized water to remove surface radioactivity. The seedlings were dried for two weeks between layers of absorbent paper and placed in a cassette with 18 X 13 cm X-ray film (Eastman Kodak, Rochestrer, NY). The film was exposed for two weeks at -80°C, developed and evaluated visually.

Results and Discussion

Pima S-7, Pima S-6, and Acala 1517-75 cotton cultivars had high levels of tolerance to prometryn compared to fifteen other upland cultivars commonly grown in the southwest (Table 1 and Figure 1a-d). Tolerance was determined by growth at 12.0 lbs. a. i./A. All of the cultivars germinated and grew until the cotyledons began to expand. At the cotyledon stage, the susceptible seedlings developed necrotic lesions that eventually spread throughout the cotyledons and stems. Some stature reduction and discoloration in the form of slight chlorosis to Pima S-7, Pima S-6 and Acala 1517-75 was noted at the high rates of prometryn (4.8 and 12.0 lbs. a. i./A). However, even at the highest herbicide rates, new leaves were formed and developed normally. The susceptible cultivars sustained severe injury, did not produce new growth and died at rates between 0.6 to 1.2 lbs. a. i./A. There was no indication of intermediate levels of tolerance for the cultivars. Postemergence applications of prometryn did not cause injury at the low rates indicating that as seedlings aged they rapidly became more tolerant of prometryn.

There was no relationship between cultivars with regard to the number of glands present and the level of tolerance (Table 1). The tolerant cultivars had gland numbers equivalent to, or less than, many of the susceptible cultivars. For example, Pima S-6 had the lowest number of glands but was tolerant and DP 5690 had the highest number but was susceptible. Although gland number may have contributed to tolerance at lower rates of prometryn, it is uncertain whether gland number could impact tolerance to prometryn at 12.0 lbs. a. i./A. The glanded and glandless isolines of Empire 61 (WR), Westburn M, Delcot 277, TH 149, and Stoneville 213 were only evaluated at 3.4 kg ha^{-1} (3.0 lbs. a. i./A) of prometryn (9). The cultivars, noted in the studies on cotton response to atrazine and propazine, were tested at only 0.56 kg ha^{-1} (10). However, other mechanisms of tolerance are probably present considering that there was no difference between glanded and glandless cultivars of Acala 4-42, both of which had good tolerance to prometryn (11).

Autoradiographs of seedlings showed accumulation of ^{14}C-prometryn in the lysigenous glands. Hence, the glands appeared to be sequestering the prometryn. In addition, the glanded sections of DP 5415, Pima S-7 and Acala 1517-75 hypocotyls contained two to three times more ^{14}C-prometryn than non glanded areas of the hypocotyl (Table 2) further indicating that prometryn was partitioning into the glands. Autoradiographs of Pima S-7 and DP 5415 cultivars showed that the nonglanded sections did not produce an image indicating that these regions contained little prometryn (Figure 2). An autoradiograph of a non glanded cultivar, Acala g2711, showed radiolabel evenly distributed in the hypocotyl (Figure 2). These results demonstrated that glands sequestered prometryn, as others have shown (6-8), however, the amount of prometryn sequestered in the glands was no greater in susceptible DP 5415 than in tolerant Pima S-7.

These results suggest that the mechanism of cotton tolerance to prometryn was not due to differences in sequestration in glands. Previously, it was shown that differences in tolerance were not due to the relative rates of herbicide absorption, translocation, metabolism, the efficacy in inhibition of photosynthetic electron transport (5). Hence the mechanism for tolerance remains elusive. Alternative mechanisms of tolerance are possible. For example, the tolerant cultivars may contain higher levels of the enzymes and substrates that prevent oxidative stress. Acala 1517-88 and 1517-SR2 cultivars were more salt tolerant than DP 50 and STV 825 and had higher constitutive levels of catalase and α-tocopherol (12). Peroxidase and glutathione reductase activites also increased following salt treatment in the salt tolerant, but not in the salt sensitive, cultivars (12). The oxidized to reduced ascorbate ratio was lower and the reduced to oxidized glutathione ratio was higher in the salt tolerant cultivars (12). Thus, cotton cultivars may contain different levels of protective mechanisms against oxidative stresses. If these mechanisms were present in the prometryn tolerant cultivars, less oxidative stress would be realized allowing for a greater potential for repair and recovery from the prometryn treatments. The basis for the differential tolerance is under investigation.

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Table 1 Gland Number and Prometryn Tolerance of Eighteen Cotton Cultivars*

Cultivars	Gland Number		Prometryn Tolerance
	Cotyledon	Stem	
Pima S-7	52.6 ± 3.6	76.5 ± 6.8	T
Pima S-6	44.4 ± 2.0	53.4 ± 2.1	T
Acala 1517-75	55.2 ± 2.8	61.6 ± 2.7	T
DP20	56.5 ± 3.5	59.1 ± 2.9	t
DP50	59.8 ± 3.9	71.2 ± 5.7	t
DP77	51.7 ± 3.5	60.3 ± 4.4	t
DP90	47.6 ± 3.2	50.0 ± 2.7	t
DP5415	47.4 + 4.6	69.8 ± 3.8	t
DP5461	56.5 ± 5.4	72.5 ± 4.9	t
DP5690	74.0 ± 4.1	64.0 ± 2.9	t
DP5816	61.4 ± 4.7	61.8 ± 4.5	t
STV311	51.8 + 5.6	49.6 ± 3.3	t
STV324	68.6 ± 2.2	63.7 ± 4.1	t
STV453	55.3 ± 3.7	58.8 ± 2.6	t
STV506	53.0 ± 2.7	68.8 ± 7.2	t
STV825	48.5 ± 1.9	68.1 ± 4.5	t
STV887	53.7 ± 3.5	69.6 ± 3.6	t
STV907	48.5 ± 2.6	73.7 ± 3.4	t

*Data are the means ± SE from two experiments (n = 20). T = tolerant to pereemergence applications of prometryn, t = susceptible to preemergence applications of prometryn.

Table 2. ^{14}C-prometryn partitioning into lysigenous glands of DP 5415 Pima S-7, and Acala 1517-75, seedlings after two days of incubation.

Cultivar	^{14}C-prometryn	
	Glanded	Non Glanded
DP 5415	141 ± 19	70 ± 10
Pima S-7	176 ± 36	76 ± 15
Acala 1517-75	143 ± 25	83 ± 15

^{14}C-prometryn uptake and partitioning in glands wasdetermined by excising glands and homogenizing them in methanol. Gland-free areas were also excised and counted. ^{14}C-prometryn in aliquots of the methanol extracts were determined by liquid scintillation counting. Data are the means ± SE of two experiments (n=12).

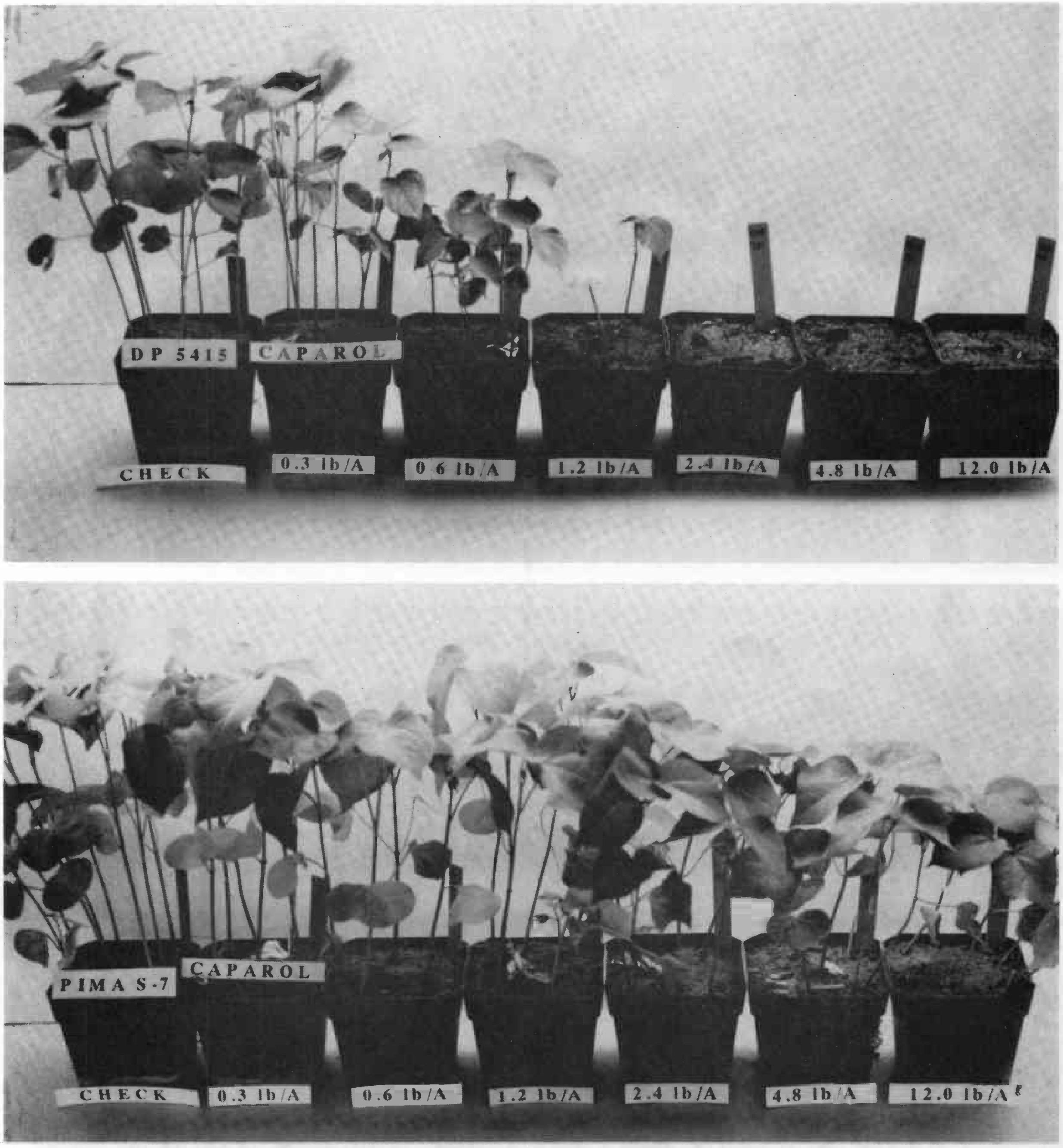

Figure 1a,b. Effect of prometryn (Caparol) on the growth of DP 5415 (a) and Pima S-7 (b) cotton. Prometryn was applied preemergence from 0.3 to 12 lbs. a. i./A. Germination was equivalent across all rates of prometryn. These are a representative photographs taken 5 WAT.

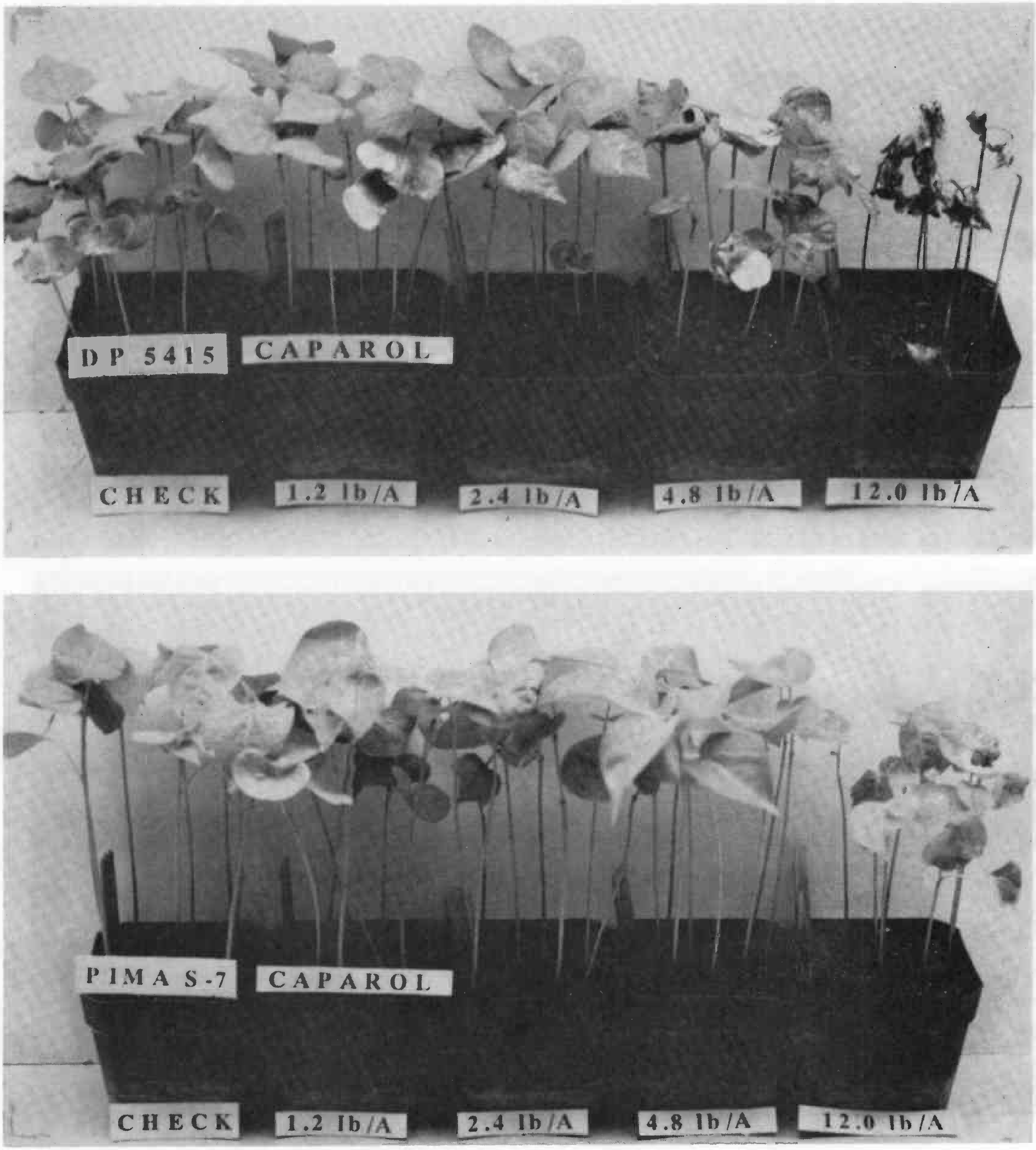

Figure 1c,d. Effect of prometryn (Caparol) on the growth of DP 5415 and Pima S-7 cotton. Prometryn was applied postemegence from 1.2 to 12 lbs. a.i./A. These are a representative photographs taken 3 weeks after treatment.

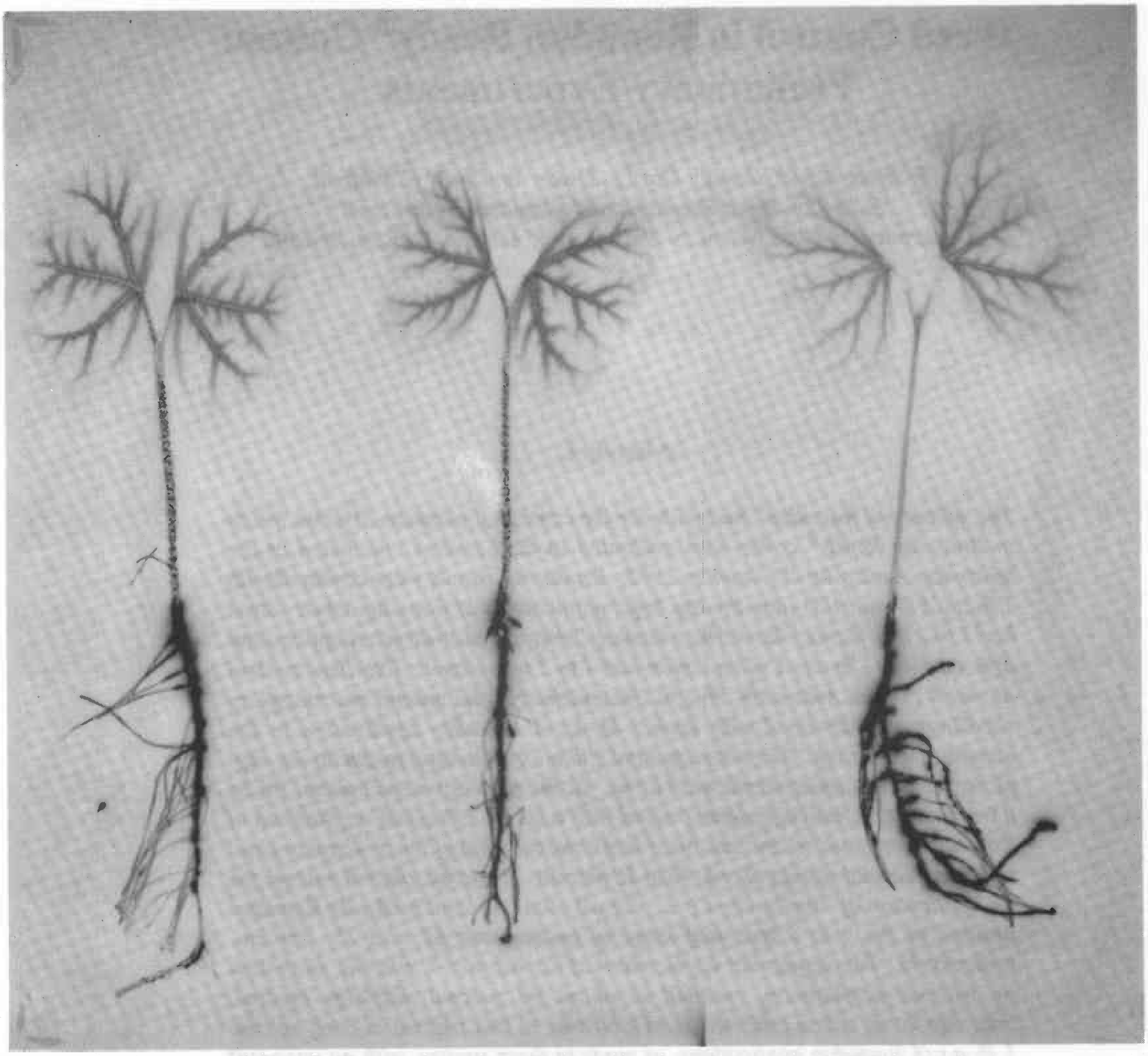

Figure 2. Autoradiographs of Pima S-7 (left), DP 5415 (center), and Acala g2711 (glandless) seedlings that had been incubated with ^{14}C-prometryn for 48 hours in hydroponic culture. Glanded cultivars show sequestration of ^{14}C-prometryn into the lysigenous glands of hypocotyls and movement of ^{14}C-prometryn into the cotyledons along the veins. The glandless cultivar shows uniform distribution of ^{14}C-prometryn in the hypocotyl and movement into the cotyledons along the veins.