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Effects of 1,1-Dimethylpiperidinium Chloride on the Pests and Allelochemicals of Cotton and Pecan

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The growth regulator, PIX (mepiquat chloride = 1,1-dimethyl-piperdinium chloride), when applied to cotton (Gossypium hirsutum L.) and pecan (Carya illinoensis Koch), caused internode shortening. PIX did not elicit an increase in resistance in cotton to the tobacco budworm [Heliothis virescens (Fab.)], or in pecan to pecan scab [Cladosporium caryigenum (Ell. et Lang) Gottwald]. Also, changes in content of four known allelochemicals (condensed tannins, gossypol, anthocyanins, flavonoids) for these pests were minimal. An unexpected finding was the increase in content of several nutritional factors that may be related to greater, rather than lesser, growth of tobbaco budworm larvae feeding on cotton tissues.

Plant growth regulators have an important role in the growth and developmental processes of plants. In cotton, (Gossypium hirsutum L.) termination of late season fruiting has been achieved with potassium 3,4-dichloroisothiazole-5-carboxylate, thus depriving the pink bollworm [Pectinophora gossypiella (Saunders)] of food and oviposition sites (1). Of perhaps greater importance would be the control of insects during the growing season. Plant growth regulators have been shown to increase the biosynthesis of certain secondary plant constituents that, in turn, decrease plant attack by insects. Gibberellic acid for example, elicits increased terpene biosynthesis in citrus (Citrus sp.), thus decreasing attack by fruit flies (Anasterpha sp.) (2,3).

Phytoalexin is a general term for compounds that are induced, whether by an infectious agent or by a chemical compound such as a bioregulator (4). More phytoalexins are synthesized when the plants are subjected to stress.

Plant growth regulators have been both isolated from and applied to cotton. Recently, 1,1-dimethyl-piperidinium chloride (mepiquat chloride = PIX) has been found to control undesirable vegetative growth, and to promote boll set (5,6). There have been recent reports about the effects of PIX on insect pests of

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cotton. Zummo et al. (7) reported less plant damage, decreased bollworm [Heliothis zea (Boddie)] growth, and 10-20% increased terpenoids, tannins, and astringency (biological tannin) in a Texas field plot test. Ganyard (8) in North Carolina, observed a 23% decrease in bollworm damage in PIX treated cotton.

There do not appear to be any reports about the application of PIX to pecans (Carya illinoensis Koch). Many other plant growth regulators have been applied to a number of fruit and nut trees and their effects recorded. Borazjani (9) has reviewed much of this literature. Increased fruit set and chlorophyll, and decreased internode lengths, have often been observed.

This study was initiated to determine whether PIX and other growth regulators enhanced a) the intrinsic resistance of cotton to insects by increasing allelochemics, and b) the resistance of pecan to pecan scab. These crops and their pests were selected for study because they were conveniently available, and are the subjects of comprehensive on-going breeding programs at this location.

Materials and Methods 1/

1982 PIX Cotton Test. Pix was applied to Stoneville 213 (ST 213) cotton on July 6 and July 21 at the rates of 0, 0.4, 1.2 and 3.6 liter/hect A.I. There were six replications. Plant tissues were harvested immediately before the first and second treatments, and also at 2 weeks following the second treatment. They were freeze-dehydrated and ground, then held at -20C in cap-closed vials until analyzed. Second instar tobacco budworm larvae housed individually in plastic cages in the laboratory were fed plant tissues (either leaves or buds) for three days. Rate of growth, in mg/day, was determined.

Analysis of Allelochemicals. Analyses for gossypol and related terpenoid aldehydes were performed on cyclohexane/ethyl acetate/acetic acid: 500/500/1 (CHEA) extracts of plant tissue by the phloroglucinol reaction (2% in abs. ETOH/Con HCl: 1/1, stand 1 hr) with subsequent spectrometic analysis at 550 nm. The concentration was determined by comparison with data obtained from authentic gossypol, and is expressed as gossypol equivalents. Condensed tannin analyses were performed on 70% aqueous methanol (MW) extracts of tissue. The anthocyanidin chromophore was developed by boiling 1 hr with n-butanol/HCl:95/5. The concentration was determined by comparison with the color obtained at 550 nm, from a purified cotton condensed tannin sample, the structure of which has recently been elucidated by Collum et al. (10). The anthocyanin content was determined by measuring the absorbancy at 540 nm of an extract of freeze dehydrated tissue, extracted with methanol/water/HCl: 79/19/3, using the molar extinction coefficient (E) of Cyanidin-3-β-glucoside (11). Flavonoids were determined after extraction of freeze-dehydrated tissue with 70% aqueous acetone. Diphenylboric acid-ethanolamine complex (Natural

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Product reagent A, Aldrich Chem. Co. 1%) in methanol was added, and the chromophore absorptivity at 440 nm was determined and compared to that obtained from a purified sample of isoquercitrin, the most prevalent flavonoid in cotton. AOAC methods were used for the following analyses: Solids (moisture); 14.083, crude fat; 14.019, crude fiber; 14.118, ash; 14.114, and nitrogen free extract (NFE) by difference from 100%.

1981 Pecan Test of Growth Regulators on Pecans. Fifteen mature Van Deman pecan trees located on the Black Belt Experiment Station in Brooksville, MS were used to evaluate the effects of four growth regulators on juglone synthesis and/or accumulation and growth habits. A randomized complete block experimental design with 3 replications was employed, using one-tree plots. Growth regulators for this study were Indole-3-acetic acid (Sigma Chemical Co., St. Louis, MO), Gibberellic acid (GA_3) (Sigma Chemical Co., St. Louis, MO), Para-coumaric acid (Sigma Chemical Co., St. Louis, MO), and PIX (1,1-dimethyl-piperidinium chloride, BASF Co., Limburgerhof, West Germany). A single spray application was made on April 24, 1981, using a hydraulic sprayer (Tricone^R nozzle 10-14 kg/cm^2). Each material was applied to the foliage at a concentration of 100 ppm to the point of "coverage without run-off" (approximately 56.8 L/tree). Sampling procedures for juglone analyses were carried out at three week intervals beginning in May and ending in late September. Two samples of leaves and nuts were taken from individual terminals in widely spaced random locations on each tree. Each sample of leaves and nuts consisted of no less than 10 g. Samples were transported in an ice chest to the laboratory and placed in the freezer.

1982 Test of PIX on Pecans. Twenty-one mature Van Deman pecan trees, located at the Black Belt Experiment Station in Brooksville, MS were used for this study. A randomized complete block design was employed, using one-tree plots and 3 replications. PIX was applied to the foliage by using a hydraulic sprayer (Tricone nozzle 10-14 kg/cm^2) at concentrations of 100 and 200 ppm (ai) to the point of "coverage without run-off" (56.8 L/tree). The first applications were made when first leaves were one-third grown and applications of some treatments were repeated at 3-week intervals. There were seven treatments, including 1, 2, 3 and 4 applications at 100 ppm, 1 and 2 applications at 200 ppm, and a check.

Analyses and Measurement of Pecan, 1981-2. Juglone analyses were accomplished using a technique developed by Hedin et al. (12). Extractions were made from 10 g of leaves or nuts by grinding in chloroform/methanol (2:1). The filtrate from 3 successive extractions of tissue was combined and concentrated to 25 mL by evaporation under vacuum at 50 C. A 1 mL aliquot was banded on a Silica Gel G TLC plate and chromatographed with methylene chloride/pentane (1:3). Juglone was observed as a yellow-orange band at R_f 0.40, scraped into a test tube, eluted from the silica gel with methylene chloride, filtered, and diluted to 10 mL for spectrometric analysis at 420 nm. For comparison, a standard curve was prepared with dilutions of 0.05-0.50 mg/10 mL of an authentic sample of juglone (Aldrich Chemical Co., Milwaukee, WI.). MS analysis showed that the fragmentation patterns of the commercial

sample and the sample from pecan leaves were identical. Analyses for condensed tannins were performed as described earlier. Samples were oven dried, ground, and analyzed for P, K, Ca, Mg, Mn, Fe, Zn, Cu, and B using spark emission spectroscopy procedures described by Jones and Warner (13). Total nitrogen was measured by the micro-Kjeldahl procedures of Bremner (14).

Ten terminals were taken from each tree at widely spaced random locations on June 18 and again on September 24, 1981, for the growth habit study. Terminals were wrapped in moist paper and transferred to the laboratory for measurements. Leaf area was measured by means of an Automatic Area Meter (Hayashi Denko Co., Tokyo, Japan). Terminal length, nut weight, nut number, terminal leaf area, specific leaf weight, and number of leaflets/leaf were also measured.

Results and Discussion.

1982 PIX Cotton Test.

In evidence that PIX had elicited its expected agronomic effects, the internode lenths of ST-213 cotton were decreased by approximately 25%; similar reductions were reported by Namken and Gausman (5). When cotton plant tissues were harvested and fed to tobacco budworm larvae in the laboratory for 3 days, the growth rate was significantly increased by 55% at the level of 3.6 l/ hect; .59 mg/mg larva/day vs control; .38 mg/mg larva/day (15).

The concentrations of allelochemicals in terminals and buds are listed in Tables I, II, III, and IV and show changes by the end of the experiment. The allelochemicals measured (tannins, gossypol, anthocyanin, and flavonoids) have been reported to contribute to resistance in cotton to the tobacco budworm (16). Gossypol levels in buds, but not in the leaves, increased significantly 4 weeks after treatment of the plants with the 3.6 l/hect. rate. The average concentration of gossypol, over time, was also significantly increased by this treatment. Leaf tannin and flavonoids were significantly decreased 4 weeks after applying PIX. Except for a small decrease in leaves and buds at the highest level of treatment, there was no significant effect of PIX on the anthocyanin level.

The increase in larval growth rate may be partly explained by increases in cotton terminal nutrients (Table V), perhaps coupled with the decrease of flavonoids in leaves. Increases were obtained for minerals (26%), protein (14%), and lipids (42%). Decreases were obtained for nitrogen free extract (9%), and more notably for crude fiber (16%).

In summary, increases were recorded in PIX treated cotton leaves for larval growth rate, protein, lipid, minerals, and gossypol in buds; there were decreases in leaf flavonoids and tannins, crude fiber, and internode distance (Table VI), but no effect on anthocyanins. The growing year, 1982, had more rainfall, than average, so that the cotton was not drouth stressed. The higher rate of larval growth may have been partially attributable to the higher nutrient concentrations and, perhaps, to the lower flavonoid level in PIX treated plants. The decreased larval growth rates observed in Texas by Zummo et al. (7) and in North Carolina by Ganyard (8) (1982) were obtained during adverse growing seasons when the cotton was stressed. Additionally, they grew larvae on growing plants and we used excised terminal leaves.

Table I. Effect of PIX on tannin levels of leaves and buds of ST 213 cotton over time, test commenced July 6

Treatment	Prior to treatment		2 weeks [1] later		4 weeks [2] later		$\bar{x}$ over time	
	leaf	buds	leaf	buds	leaf	buds	leaf	buds
ℓ/hect.	- % -							
0	8.21a [3]	8.62a	13.78a	9.64a	21.93a	10.67a	14.64a	9.64a
0.4	7.86a	8.06a	13.55a	9.73a	19.14ab	9.49a	13.52a	9.09a
1.2	10.59a	8.07a	14.79a	8.63a	16.34 bc	10.19a	13.91a	8.96a
3.6	8.84a	10.35a	14.40a	9.74a	13.68 c	11.33a	12.32a	14.47a

1/ Sampled immediately before second PIX application.

2/ Four weeks after first application, two weeks after second PIX application.

3/ Means within a column followed by the same letter are not significantly different at 0.05 level, as determined by Duncan's Multiple Range Test.

Table II. Effect of PIX on gossypol levels of terminal leaves and buds of ST 213 cotton over time; test commenced July 6

Treatment	Prior to treatment		2 weeks later [1]		4 weeks later [2]		$\bar{x}$ over time	
	leaf	buds	leaf	buds	leaf	buds	leaf	buds
ℓ/hect.	- % -							
0	0.43a [3]	0.35 b	0.29a	0.14a	0.24a	0.22a	0.32a	0.24a
0.4	0.40a	0.34ab	0.28a	0.16a	0.29a	0.25a	0.32a	0.25a
1.2	0.42a	0.28ab	0.32a	0.21a	0.24a	0.31a	0.33a	0.26a
3.6	0.48a	0.24a	0.36a	0.22a	0.29a	0.34 b	0.37 b	0.26a

1/ Sampled immediately before to second PIX application.

2/ Four weeks after first PIX application, two weeks after second PIX application.

3/ Means within a column followed by the same letter are not significantly different at the 0.05 level, as determined by Duncan's Multiple Range Test.

Table III. Effect of PIX on anthocyanin levels of terminal leaves and buds of ST 213 cotton over time; test commenced July 6

Treatment	Prior to treatment		2 weeks later[1]		4 weeks later[2]		$\bar{x}$ over time	
	leaf	buds	leaf	buds	leaf	buds	leaf	buds
ℓ/hect.	- - - - - - - - - - - - - - - - - - % - - - - - - - - - - - - - - - - - -							
0	1.15a[3]	0.10a	0.53a	0.07a	0.53a	0.59a	0.74a	0.25a
0.4	0.79a	0.09a	0.40a	0.11a	0.50a	0.56a	0.56a	0.25a
1.2	0.94a	0.09a	0.43a	0.08a	0.45a	0.52a	0.61a	0.23a
3.6	0.76a	0.10a	0.50a	0.09a	0.43a	0.44 b	0.56 b	0.21a

1/ Sampled immediately before to second PIX application.

2/ Four weeks after first PIX application, two weeks after second PIX application.

3/ Means within a column followed by the same letter are not significantly different at the 0.05 level, as determined by Duncan's Multiple Range Test.

Table IV. Effect of PIX on flavonoid levels of terminal leaves and buds of ST-213 cotton over time, test commenced July 6

Treatment	Prior to treatment		2 weeks later[1]		4 weeks later[2]		$\bar{x}$ over time	
	leaf	buds	leaf	buds	leaf	buds	leaf	buds
ℓ/hect.	- % -							
0	2.76a[3]	0.84a	2.93a	1.27a	3.09a	1.35a	2.92a	1.15a
0.4	2.71a	0.79a	2.75a	0.13a	2.57 b	1.12a	2.67 b	1.00a
1.2	2.74a	0.77a	2.96a	1.02a	2.51 b	1.46a	2.73 b	1.08a
3.6	2.67a	0.85a	2.67a	1.13a	2.13 c	1.39a	2.50 b	1.12a
LSD .05	ns	ns	ns	ns	0.37		0.22	ns

1/ Sampled immediately before to second PIX application.

2/ Four weeks after first PIX application, two weeks after second PIX application.

3/ Means within a column followed by the same letter are not significantly different at the 0.05 level, as determined by Duncan's Multiple Range Test.

Table V. Effects of PIX on ST-213 proximate composition of cotton terminals after 28 days

Treatment	Ash	Crude Protein	Crude Fat	Crude Fiber	NFE
1/hect.			%		
0	5.95	21.41	3.16	7.63	47.47
1.2	7.16	22.50	3.92	6.61	46.50
3.6	7.52	24.38	4.48	6.39	43.07

Table VI. Summary; Effects of PIX (3.6 1/hect.) on ST-213 cotton terminal content after 28 days

Increases		Decreases		No Change
Protein	14%	Flavonoids	31%	Gossypol
Lipid	42%	Tannins	38%	Anthocyanins
Minerals (ash)	26%	Internode Distance	25%	
Larval Growth Rate	55%	Crude Fiber	16%	

1981 Pecan test. The effects of the growth regulators on growth habits at 8 and 21 weeks after application are given in Tables VII and VIII. Only the specific leaf weight of differed significantly among the five treatments at 8 weeks. Leaves of trees treated with PIX had a higher specific weight (g/cm^2) than those treated with other chemicals on both observation dates. At the second sampling date, the number of leaflets/leaf for GA_3 and IAA treated leaves was significantly higher than for the other treatments (Table VII). When the data from the two observation dates were combined and analyzed collectively, all sprayed trees showed a significant decrease in terminal length, except those treated with GA. Also, PIX treated trees appeared to have a higher nut set, nut weight, and specific leaf weight for terminal leaves, but only the specific leaf weight of terminal leaves was significantly different at the $P=0.05$ level.

The increased specific leaf weight with PIX agrees with the findings of Gausman et al. (6) that this growth retardant increases leaf weight and reduces the leaf area due to changes in the mesophyll structure of leaves. Reduced height of cotton was also reported for PIX by (Gausman et al. (6). Since GA_3 and IAA promote new growth, the increase in number of leaflets/leaf seems consistent. The increased length of terminals observed here is consistent with the known elongation of internodes by GA_3 and their ability to cause stem elongation and even to reverse genetic dwarfism in some plants (17).

Juglone in the leaves of PIX and IAA treated trees was significantly higher than in the check leaves 3 weeks following application (Table IX). The juglone level was significantly lower in all treatments at 9 weeks. Although some differences were observed among treatments in the following weeks, none was significant. Juglone levels in nuts for each sampling date are given in Table X. No significant differences were observed among nut tissues until 12 weeks following application, when PIX and IAA-treated

Table VII. Pecan Growth Habit Measurements (6/18/81)

Treatment[1]	Terminal Length	Nut/ Treatment	Nut Wt.	Leaflet/ Leaf	Leaf Wt./ Terminal	Terminal Leaf Area	Terminal Leaf	Terminal Leaf Rachis Length
	cm		g		g	cm^2	mg/cm^2	cm
PIX	7.8 a[2]	2.6 a	1.7	9.8	25.5	298	17 a	34.5
IAA	6.8 a	2.7 a	1.6	9.9	24.6	295	15 b	34.2
GA_2	8.0 a	2.6 a	1.6	9.9	23.8	305	15 b	34.7
P-Coumaric acid	7.6 a	2.4 a	1.4	9.2	25.1	301	14 b	34.0
Check	7.6 a	2.6 a	1.6	9.7	27.1	330	14 b	35.4

1/ 100 PPM to dip off on 4/24 (56.8 L/tree).

2/ Values within a column followed by the same letter are not significantly different, as determined by Duncan's Multiple Range Test, p=0.05.

Table VIII. Pecan Growth Habit Measurements (9/24/81)

Treatment [1]	Terminal Length	Nut/ Treatment	Nut Wt.	Leaflet/ Leaf	Leaf Wt./ Terminal	Terminal Leaf Area	Terminal Leaf	Terminal Leaf Rachis Length
	cm		g		g	cm^2	mg/cm^2	cm
PIX	5.6 a [2]	2.3 a	9.8	9.5	25.2	297	16 a	35.3
IAA	7.6 a	1.9 a	8.1	10.1	25.9	308	13 b	34.5
GA_3	9.5 a	1.9 a	8.6	10.3	28.8	336	13 b	38.1
PCA	6.8 a	1.8 a	8.1	9.4	24.0	316	13 b	33.1
check	8.6 a	1.6 a	9.3	9.3	26.4	288	13 b	32.8

1/ 100 PPM to drip off on 4/24 (56.8 L/tree).

2/ Values within a column followed by the same letter are not significantly different, as determined by Duncan's Multiple Range Test, p=0.05.

Table IX. Pecan Leaf Juglone Concentration (mg/g fresh tissue)

Treatment[1]	1981 Sampling Dates						
	May 15	June 4	June 25	July 16	Aug 6	Aug 27	Sept 17
	mg/g fresh weight						
PIX	1.23 a[2]	0.93	0.52	0.37	0.56	0.33	0.25
IAA	1.21 ab	0.76	0.44	0.54	0.58	0.27	0.20
GA_3	0.89	0.85	0.49	0.40	0.60	0.41	0.23
PCA	0.81 bc	0.86	0.70 b	0.62	0.38	0.32	0.34
Check	0.65 c	0.82	1.06 a	0.71	0.55	0.30	0.16

1/ 100 PPM to drip-off on 4/24 (56.8 L/tree).

2/ Values within a column followed by the same letters are not significantly different, as determined by Duncan's Multiple Range Test, p=0.05.

Table X. Pecan Nut Juglone Concentration (mg/g fresh tissue)

Treatments[1]	1981 Sampling Dates					
	June 4	June 25	July 16	Aug 6	Aug 27	Sept 17
PIX	0.10 a[2]	0.16	0.45 ab	0.25 b	0.33 a	0.32 a
IAA	0.12 a	0.12	0.50 a	0.27 b	0.39 a	0.18 b
GA_3	0.13 a	0.12	0.30 bc	0.33 b	0.18 a	0.26 a
PCA	0.11 a	0.17	0.30 bc	0.52 a	0.08 a	0.25 a
Check	0.13 a	0.22	0.30 c	0.30 b	0.21 a	0.29 a

1/ 100 PPM to drip off on 4/24 (56.8 L/tree).

2/ Values within a column followed by the same letters are not sigificantly different, as determined by Duncan's Multiple Range Test, p=0.05.

trees showed significantly higher juglone levels than the controls(Table X). The p-coumaric acid treatment showed a significantly higher juglone content than the other plant growth regulators at 15 weeks following application. Also, on the last sampling date (21 weeks after application), IAA treated nuts showed lower juglone content than the others.

The lack of continued influence of bioregulators on juglone levels may result from dilution by increased plant biomass with time after treatment. IAA moves rapidly from the young green tissue to older tissue, and it is constantly being destroyed by indole-3-acetic acid oxidase (17). Gibberellin is also reported to lose its biological activity gradually after treatment (18); the coumarins are known to bind to sugar to form their glucosides (19). IAA, GA, and p-coumaric acid are reported to stimulate respiration (20), possibly by activating the oxidative enzyme system, thereby enhancing synthesis of phenolic compounds such as juglone. There is no report about the effect of PIX on phenolic pathways.

Delayed juglone accumulation in nut tissues (until 12 weeks after application) suggests that time may be related to translocation of excess material observed earlier in the leaves. Conversely, compounds may have accumulated that were subsequently metabolized amd permitted the increase in juglone.

1982 Pecan Test. Terminal shoot growth of all treated trees was significantly reduced by the first measurement on June 29 (Table XI). However, there were no significant differences from the unsprayed check for any of the other growth measurements that were made on this date.

Table XI. Effect of PIX on growth of mataure Van Deman trees

Treatment Number	Mepiquat Chloride Concentration	Number of Applications	Terminal Shoot Length
			-cm- [1]
1	200 ppm	1	6.93 b
2	200 ppm	2	7.15 b
3	100 ppm	1	6.60 b
4	100 ppm	2	7.13 b
5	100 ppm	3	6.86 b
6	100 ppm	4	6.70 b
7	Unsprayed check	0	9.50 a

Note: Measurements were made on 6/29/82; each value is the average of 3 replications.

1/ (p = 0.05) according to Duncan's Multiple Range Test.

Significant differences were recorded for all growth parameters measured in September (Tables XII, XIII). Terminal shoot growth was reduced from 30 to 50% among the various treatments (Table XII). This dramatic effect is significant, since the concentration of PIX used in this experiment was only a fraction of that commonly used on cotton. There were essentially no differences between treated and non-treated trees for number of nuts/terminal or in weight. Otherwise, there was a slight reduction in the number of leaflets/leaf (Table XII) and total leaf

Table XII. Effect of PIX on mature Van Deman pecan trees, as measured on Sept. 22, 1982

Treatment Number	PIX concentration	Number application	Terminal shoot length	Number of nuts/terminal	Nut weight	Number of leaflets/leaf
			(cm)		(g)	
1	200 ppm	1	5.60 b [1]	2.10ab	11.75ab	7.28 b
2	200 ppm	2	6.18 b	2.10ab	11.14ab	7.27 b
3	100 ppm	1	5.95 b	2.36a	12.54a	8.03 b
4	100 ppm	2	5.31 b	1.84 b	12.50a	6.96 b
5	100 ppm	3	4.58 b	2.00ab	10.75 b	7.32 b
6	100 ppm	4	6.30 b	1.93 b	12.32ab	7.64 b
7	Unsprayed check		9.15a	1.92 b	11.82ab	9.36a

Note: Each figure represents an average of 3 replications.

1/ Means within a column not followed by the same letter differ significantly ($p = 0.05$) according to Duncan's Multiple Range Test.

Table XIII. Effect of PIX on leaf characteristics of mature Van Deman pecan trees, as measured on Sept. 22, 1982

Treatment Number	PIX Concentration	Number Applications	Total leaf wt/ Terminal (cm)	Average Leaf Area	Specific Leaf Wt. (gm/cm^2/)	Average Rachis Length (cm)
1	200 ppm	1	10.28 b [1]	216 d	.013 abc	28.50 bc
2	200 ppm	2	15.43 b	264 b	.016 a	26.26 d
3	100 ppm	1	14.25 b	288 b	.014 abc	28.56 bc
4	100 ppm	2	12.03 b	239 c	.013 abc	29.02 bc
5	100 ppm	3	11.48 b	256 c	.012 c	28.06 c
6	100 ppm	4	14.75 b	278 b	.013 abc	29.78 b
7	Unsprayed check		22.05 a	328 a	.015 ab	32.00 a

Note: Each figure represents an average of 3 replications.

1/ Means within columns not followed by the same letter differ significantly (p = 0.05) according to Duncan's Multiple Range Test.

weight/terminal (Table XIII). Reductions in average leaf weight/unit area and average rachis length (Table XIII) were generally more pronounced.

These data suggest that PIX may be useful in reducing pecan tree size. This material may hold potential for those using precocius (early bearing) varieties in close spacing. These compounds delay crowding in these plantings, thus permitting longer periods of high yield before thinning to reduce the crowding effects.

Significant effects on seasonal juglone patterns in both leaves and nuts were noted for all treatments. However, the patterns among the treatments tended to be somewhat erratic and difficult to interpret. The patterns for the two highest rates, (four treatments at 100 ppm and two treatments at 200 ppm) and the check are given in Figure 1. The juglone level of leaves for the unsprayed check is consistent with that found for the Van Deman cultivar in previous studies. The juglone levels for nuts at the two highest rates and the check are given in Figure 2. The greatest effect on juglone levels in nuts may occur near the end of the season.

PIX is systemic and is not readily metabolized in cotton plants (17). Therefore, repeated applications during the growing season might be expected to increase the juglone level, but this trend was not consistent in this study. This may be due to a dilution of PIX by increased plant biomass, or photodegradation of PIX on leaf surfaces. The juglone content was studied because juglone has been shown to be a factor of resistance in pecan to pecan scab. In these PIX treatments, however, the incidence of pecan scab was not significantly reduced.

Tannin levels in leaves increased temporarily for all treatments through 3 and 6 weeks (April 28 and May 18 sampling dates respectively) after first applications (Table XIV). Therefore, following this, no significant differences among treatments were observed, with few exceptions, for the remainder of the season. PIX had no appreciable influence on the foliar levels of the nutrients (N,P,K,Ca,Mg,Mn,Fe,Zn,Cu,and B).

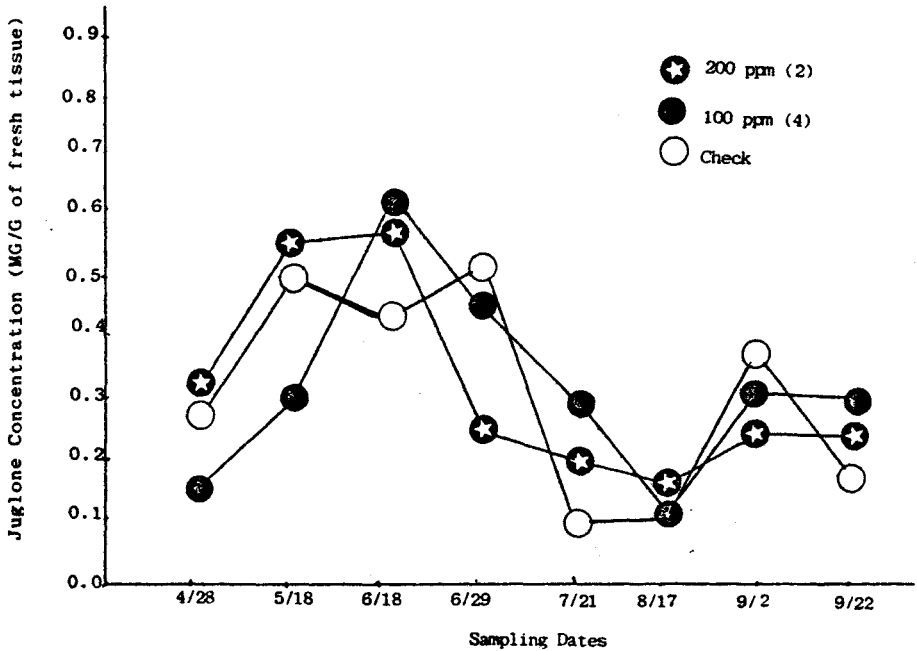

Figure 1. Seasonal Juglone concentrations in leaves of Van Deman pecan trees treated with PIX (200 ppm, 2 applications and 100 ppm, 4 applications) as compared with unsprayed check. Each datum point represents an average of 3 replications.

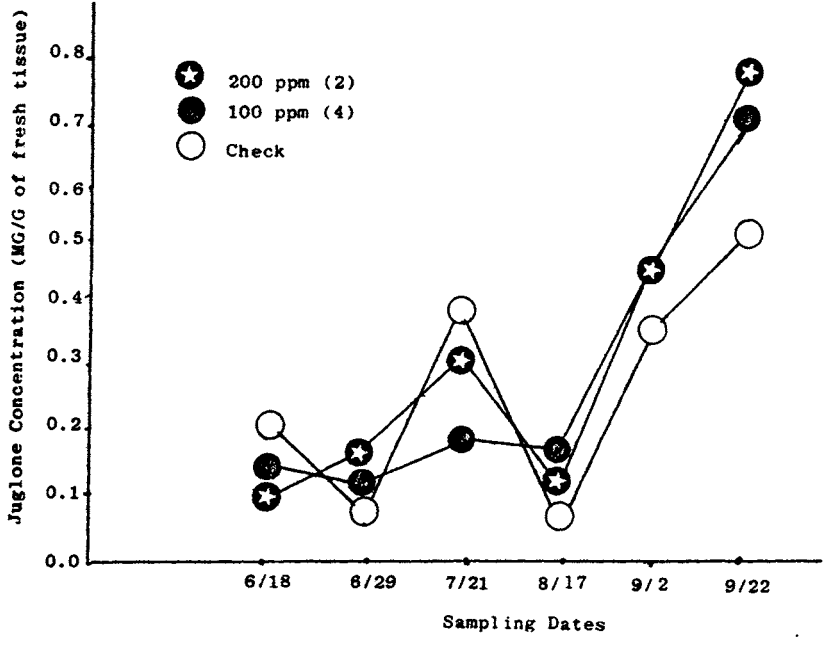

Figure 2. Seasonal juglone concentrations in nuts of Van Deman pecan trees treated with PIX (200 ppm, 2 applications and 100 ppm, 4 applications) as compared with unsprayed check. Each datum point represents an average of 3 replications.

Table XIV. Tannin concentration in leaves of Van Deman pecan trees treated with PIX

Treatment Number	PIX Conc.	# of appl.	Sampling Dates							
			Apr. 28	May 18	June 8	June 29	July 21	Aug. 12	Sept. 12	Sept. 22
			mg/g fresh wt.							
1	200 ppm	(1)	2.45a [1]	3.01 b	3.57ab	5.04a	4.97ab	5.46 cd	7.07ab	5.39 b
2	200 ppm	(2)	2.52a	2.94 c	3.57ab	4.41ab	3.65 c	5.81 cd	6.44ab	5.32 b
3	100 ppm	(1)	2.27a	2.76 e	3.57ab	4.48ab	3.85 bc	6.44a	6.09 b	7.21a
4	100 ppm	(2)	2.45a	3.01 b	3.36ab	4.87ab	4.20abc	6.02abc	6.58ab	6.02 b
5	100 ppm	(3)	2.73a	3.17a	4.04a	4.20ab	5.37a	6.3 ab	7.28a	5.25 b
6	100 ppm	(4)	2.34a	2.91a	3.20ab	3.99 b	3.64 c	5.25 d	6.58ab	5.25 b
7	Unsprayed check		1.78 b	2.56	3.01 b	4.06 b	4.55abc	5.95abc	6.37ab	6.16 b

Note: Each figure represents an average of 1982 data from 3 replications.

1/ Means within columns not followed by the same letter differ significantly at ($p = 0.05$) according to Duncan's Multiple Range Test.

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