EFFECTS OF ACIFLUORFEN ON TRANSLOCATION OF AUXINS AND ON THE ABSORPTION AND TRANSLOCATION OF FOUR GRAMINICIDES

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PART I

INTERACTION OF ACIFLUORFEN WITH ABSORPTION AND TRANSLOCATION OF FOUR GRAMINICIDES

Interaction of Acifluorfen with Absorption and Translocation of Four Graminicides

Abstract. The objective of this study was to determine the feasibility of using acifluorfen [5-(2-cholor-4-(trifluoromethyl)phenoxy)-2-nitrobenzoic acid] and four graminicides for the control of large crabgrass (Au[Digitaria sanguinalis] (L.) Scop # DIGSA) and Palmer amaranth (Amaranthus palmeri S. Wats # AMAPA). Preliminary studies showed that acifluorfen, when applied prior to the graminicide, caused deliterious effects on grass control by the different graminicides. Studies on the effects of acifluorfen on the absorption, translocation, and recovery of ^{14}C-labeled graminicides were then conducted. The four graminicides were haloxyfop [methyl 2-(4-((3-chloro-5-(trifluoromethyl)-2-pridinyl)oxy)phenoxy)propanoic acid], fluazifop [butyl (RS)-2(4-((5-(trifluoromethyl)-2-pyridinyl)oxy)phenoxy)propanoic acid], xylafop [2-(4-((6-chloro-2-quinoxalinyl)oxy)phenoxy)propionic acid, ethyl ester], and sethoxydim [2-(1-(ethoxyimino)butyl)-5-(2-(ethylthio)propyl)-3-hydroxy-2-cyclohexene-1-one].

Two-week-old grain sorghum (Sorghum bicolor (L.) Moench) plants were treated by placing two 1-µl drops of ^{14}C-labeled graminicide, with and without unlabeled acifluorfen, on the second oldest leaf 3 cm from the base of the leaf and either side of the midrib. The plants were harvested at 3 or 6 h, sectioned and analyzed for ^{14}C. In the field acifluorfen was applied postemergence alone to soybean (Glycine max (L.) Merr), in tank mixtures, or as sequential treatments with the four graminicides.

Acifluorfen decreased absorption of the ^{14}C-labeled graminicides into the plant leaf tissue and increased the ^{14}C recovered. Acifluorfen slightly enhanced percent acropetal and basipetal translocation. In the field, grass and broadleaf weed control and crop injury were evaluated. The four graminicides were more effective when acifluorfen was applied as a sequential treatment after, rather than prior to, graminicide treatment.

Additional index words: tank-mix, sequential treatment, haloxyfop, fluazifop, xylafop, sethoxydim, DIGSA, AMAPA.

INTRODUCTION

In 1984 there were approximately 3 million hectares of soybeans harvested in the United States. In Oklahoma 89 thousand hectares were harvested with a yield of about 1008 kg/ha. Although soybeans are not a major crop statewide in Oklahoma, they are an important crop in a few eastern Oklahoma counties. Broadleaf weeds and unwanted grasses reduce soybean yield and interfere with harvest.

Shurtleff and Coble (18) conducted field experiments in 1979, 1980, and 1981 to quantify soybean yield reductions from common cocklebur (Xanthium strumarium L. #[1] XANST), common ragweed (Ambrosia artemisiifolia L. # AMBEL), common lambsquarters (Chenopodium album L. #CHEAL), sicklepod (Cassia obtusifolia L. # CASOB), and redroot pigweed (Amaranthus retroflexus L. # AMARE) interference. They determined that at a density of 16 weeds per 10 m row all weeds reduced soybean yield, and that the leaf area of soybeans was higher at greater distances from the weed for all weed species. The range of soybean leaf area

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[1]Letters following this symbol are a WSSA-approved computer code from Composite list of Weeds, Weed Sci. 32. Suppl.2. Available from WSSA, 309 West Clark St., Champaign, IL 61820.

reductions occasioned by proximity to individual weed species corresponded fairly well with differences in soybean yield reduction. This and other data exhibit the need for effective broadleaf weed control in soybeans.

Acifluorfen was first reported in 1978 (9) to be effective on many broadleaf weeds in soybeans including current problem weeds such as common cocklebur, velvetleaf (Abutilon theophrasti Medik. # ABUTH), morningglory (Ipomoea sp.), and jimsonweed (Datura stramonium L. # DATST). Soybeans showed high tolerance to acifluorfen although there was, at times, some localized and temporary leaf damage. Hartnett (6) reported similar results with acifluorfen.

Ritter and Coble (16) found that treatment at high relative humidity (RH) resulted in a significant increase in acifluorfen phytotoxicity and a decrease in weed plant dry weight as compared to treatment at a lower RH. Temperature was not found to be as critical as RH.

In 1982 Lee and Oliver (10) investigated the effect of timing and application method on acifluorfen activity on broadleaf weeds. Their report indicated that with some weeds, lower rates (0.3 kg/ha) could be used on younger plants while older plants required higher rates (1.1 kg/ha). Control was reduced as the plants aged. With certain weed species, such as hemp sesbania (Sesbania exaltata (Raf.) Rydb. ex A.W. Hill # SEBEX), applications of acifluorfen during the dark were more effective than those applied at

sunup or midday. With some weed species, increased surfactant concentration enhanced weed control.

Ritter and Coble (17) obtained the best control (>90%) of common cocklebur 2 weeks after plant emergence. Murphy and Gossett (11) found that acifluorfen applied 7 days after cowpea (Vigna unguiculata (L.) Walp.) emergence gave better control of cowpeas than a single application at 21 days after emergence. The greatest soybean injury occurred when acifluorfen was applied at 7, 14, and 21 days after emergence, but seed yields were not reduced relative to the untreated weed-free control.

In recent years there have been efforts to find a way for farmers to have herbicides with broader spectrum weed control. Broad spectrum control is obtained by one or more applications of different herbicides. However, tank mixes can reduce the cost of application and, thus, they are often used. But some herbicide mixes may be physically incompatable in solution or physiologically incompatable. Acifluorfen has been shown to be antagonistic to weed control in tank-mixes with certain other herbicides (19). It is possible that such antagonism is due to an effect on absorption or translocation.

Studies have shown that mefluidide [N-(2,4-dimethyl-5-(((trifluoromethyl)sulfonyl)amino)phenyl)acetamide] and acifluorfen mixtures provide broad spectrum grass and broadleaf weed control without unacceptable soybean injury (4,5,8). The control of younger weeds is usually better

than control of more mature weeds. Sequential applications of mefluidide and acifluorfen often increased velvetleaf and common cocklebur control compared to either herbicide applied alone (4,8).

Retzinger et al. (14) investigated tank mixtures of acifluorfen, sethoxydim, and bentazon [3-(1-methylethyl)-(1H)-2,1,3-benzothiadiazin-4(3H)-one 2,2-dioxide]. The mixutures of bentazon and acifluorfen gave no control of johnsongrass but gave 100% control of sesbania and common cocklebur. A three-way mixture of the herbicides gave 80% johnsongrass (Sorghum halepense (L.) Pers. # SORHA) control with 90% or better control of pigweed (Amaranthus sp.), prickly sida (Sida spinosa L. # SIDSP), pitted morningglory (Ipomoea lacunosa L. # IPOLA), sesbania, and common cocklebur with no soybean injury. There was antagonism observed in a two-way mixture of sethoxydim and bentazon (12,14) but the addition of acifluorfen appeared to reduce the antagonism (14). Hartzler and Foy (7) reported that a 1.5 h or longer interval between separate applications of sethoxydim and bentazon seemed to eliminate the antagonism. Several studies have shown that mixing sethoxydim with other herbicides, such as bentazon and acifluorfen, significantly reduced johnsongrass control (1,15,19). Whitwell et al. (19) reported that acifluorfen was more antagonistic than bentazon and benazolin [4-chloro-2-oxo-3(2H)-benzothiazoleacetic acid] to the activity of postemergence grass herbicides and sethoxydim was more susceptable to

antagonism than CGA-82725 [2-propynyl-2(4-((3,5-dichloro-2-pyridinyl)oxy)phenoxy)propanoic acid] or RO-13-8895 [acetone-O-(D-2-(p-(α,α,α-trifluoro-p-tolyl)oxy)phenoxy) propionyl oxime]. The antagonism was attributed to the broadleaf herbicides, which caused soybean yield reductions of up to 67%.

The highest level of barnyardgrass (Echinochloa crusgalli (L.) Beauv. # ECHCG) control found by Chen and Penner (2) was obtained with a combination of sethyoxydim plus acifluorfen plus crop oil concentrate. This combination tended to increase foliar absorption of ^{14}C-acifluorfen by barnyardgrass but not by soybeans. But the combination of acifluorfen with diclofop-methyl [2-(4-(2,4-dichlorophenoxy)phenoxy)propanoic acid, methyl ester] appeared to reduce phytotoxicity to barnyardgrass and increase it to soybean.

Other herbicide mixtures not containing acifluorfen show varying results. Research indicated no antagonism between dalapon (2,2-dichloropropanoic acid) or TCA (trichloroacetic acid) and sethoxydim at 0.3 kg/ha applied to control volunteer barley (Hordeum vulgare L. # HORVX) and wheat (Triticum aestivum (L.) Neepawa) in flax (Linum usitatissimum (L.) Dufferin) although lower concentrations of MCPA [(4-chloro-2-methylphenoxy)acetic acid] and sethoxydim were antagonistic (3).

Sequential applications can sometimes overcome the antagonsitic effects seen by tank-mixed herbicides. Quershi

and Vanden Born (13) showed that a 4 day interval between MCPA and dicofop-methyl application prevented most of the antagonism that occurs with applications of a tank mix of the two herbicides.

Since acifluorfen has been shown to be antagonistic in tank-mixes with other herbicides (19) it is possible that this is an effect on absorption or translocation. The objective of this research was to determine whether acifluorfen mixed with four radioactive graminicides affected absorption and translocation of the graminicides. Sorghum, a susceptible grass, was used for these determinations. It was expected that this procedure would make it possible to determine to what extent the reported antagonism is related to the inhibition of herbicide translocation. Tank-mixtures and sequential applications of the herbicides also were used to show visual differences in percent weed control in soybeans under field conditions.

MATERIALS AND METHODS

A field experiment was conducted in the summer of 1985 at the Agronomy Research Station, Perkins, Oklahoma to evaluate acifluorfen interaction with haloxyfop, fluazifop, xylafop, and sethoxydim for weed control and crop response in soybeans that were seeded May 24,1985 on Teller loam (fine-loamy, mixed, thermic Udic Argiustoll; 0.3% organic matter; 5.9 pH) soil. The crop was watered by irrigation and rainfall.

Acifluorfen was applied postemergence alone with a crop oil concentrate (Agridex, Helena Chemical Company) added as an adjuvant, mixed with the four different graminicides, or as sequential treatments of each graminicide followed 24 h later by acifluorfen, and sequential treatments of acifluorfen followed 24 h later by each graminicide. The herbicides were applied at labeled rates with acifluorfen at 0.28 kg/ha, fluazifop, sethoxydim, and haloxyfop at 0.22 kg/ha, and xylafop at 0.14 kg/ha.

All treatments were applied with a tractor mounted compressed air sprayer at a delivery volume of 280 l/ha. The experiment was established with natural weed infestations. Stage of growth of the soybeans at treatment was V3: 12.5 to 17.5 cm. Weeds present at treatment were Palmer amaranth (1.25 to 3.75 cm tall, 2 to 4 leaf stage,

2.5 to 3.75 cm foliage diameter) and large crabgrass (1.25 to 5 cm tall, 3 to 5 leaf stage).

The experiment was conducted with a plot size of two 90 cm rows 7.62 m long. All tank-mix treatments were applied on June 18, 1985. Sequential treatments were applied on June 19, 1985. The experimental design was a randomized complete block with 4 replications.

Crop injury and weed control were visually rated on June 25, 1985 and July 10, 1985. Stand counts of soybeans were taken June 28, 1985. The results of this data are expressed as percent control of the weeds and percent injury of the crop.

Laboratory experiments were established to determine the interaction of acifluorfen with the absorption and translocation of four ^{14}C-labeled graminicides. The graminicides studied were haloxyfop (Dow, 10.6 Ci/M, phenoxy UL-^{14}C), fluazifop (ICI, 21.5 Ci/M), xylafop (DuPont, 8.1 Ci/M, phenol-^{14}C), and sethoxydim (BASF, 13.06 Ci/M, H-4-^{14}C). Paymaster grain sorghum was used as the indicator species in the laboratory experiments. In all of the experiments, the plants were established and maintained as follows: Five seeds per pot were planted in each 8.5x14 cm styrafoam pots containing 575 g of a Teller fine sandy loam soil (fine-loamy, mixed, thermic Udic Argiustoll; 0.8% organic matter; 5.9 pH). The pots were randomized and placed in a controlled temperature chamber with a 29° C day, 21° C night, 12 h photoperiod with a light intensity of 241

$\mu E \cdot m^{-2} \cdot s^{-1}$, and 80±5% RH. The soil was initially subirrigated to capacity and then was gravimetrically brought uniformly to near field capacity by adding water at approximately three day intervals. After emergence the seedlings were thinned to one plant per pot and treated at two weeks after planting.

Foliar absorption and translocation of the applied material was determined by placing two 1-µl drops of acetone each containing 0.05 µCi of ^{14}C-labeled technical graminicide, 0.4% Triton X100 (oxtoxynol), and with and without 2 µg unlabeled technical acifluorfen on each side of the leaf midrib 3 cm above the ligule on the second oldest leaf of the sorghum seedlings. The plants were allowed to absorb and translocate the graminicides for 3 or 6 h and then dissected into various plant parts. These included the roots, foliage below the ligule of the treated leaf, foliage above the ligule of the treated leaf, the lower 2 cm section of the treated leaf, a 2 cm treated area, and the upper portion of the treated leaf. The 2 cm treated area of each plant was washed for 30 s in 5 ml of 95% ethanol to remove any ^{14}C remaining unabsorbed on the surface of the leaf. The plant parts were then lyophilized to remove moisture, weighed, and homogenized in a total of 10 ml of 95% ethanol. An aliquot of each homogenate and wash was removed for ^{14}C analysis by liquid scintillation spectrophotometry. Each treatment was replicated 4 times and each experiment was

repeated once for a total of 8 replications per treatment. These were then pooled for analysis of variance.

Results are presented as: percent recovery, which is expressed as a percentage of the amount of ^{14}C applied; percent absorption, which is the percent of the recovered ^{14}C not removed by the wash; acropetal translocation which is expressed as a percentage of the total herbicide absorbed that moved up from the treated area; basipetal translocation which is expressed as a percentage of the total herbicide absorbed that moved down from the treated area; and the apoplastic/symplastic transport ratio, which is expressed as a ratio of acropetal to basipetal translocation.

DISCUSSION AND RESULTS

Experiments were conducted to determime the effects of tank-mixing and sequential treatments of acifluorfen with four different graminicides. The first experiments were conducted under field conditions where the herbicides were applied alone with a crop oil concentrate added as an adjuvant; applied as a mixture of graminicide and acifluorfen; applied sequentially with acifluorfen applied the first day followed by the graminicide the second day; and applied sequentially with the graminicide applied the first day followed by acifluorfen the second day. Stand counts of soybeans were recorded on June 28, 1985. The average stand count was 26 plants with no significant difference at the 0.05 level between any of the treatments (data not shown).

Most of the treatments showed good large crabgrass and Palmer amaranth control (Table 1). A sequential application of haloxyfop followed by acifluorfen gave excellent large crabgrass and Palmer amaranth control at both evaluation dates. This treatment also caused only a little soybean injury. Xylafop, fluazifop, and sethoxydim followed by acifluorfen were also fair to good treatments for large crabgrass control but Palmer amaranth control had decreased by the second evaluation date to a level that was lower than

the control exhibited by haloxyfop followed by acifluorfen. Sequential treatments of acifluorfen followed by xylafop, sethoxydim, or haloxyfop were less effective treatments for large crabgrass control at both evaluation dates than treatments where the graminicides were followed by acifluorfen treatment. Palmer amaranth control also decreased for sequential treatments of acifluorfen followed by xlyafop for both evaluation dates and haloxyfop for the second evaluation date as compared to these graminicides followed by acifluorfen treatments. Soybean injury among sequential treatments was significantly greater only for treatments of xylafop followed by acifluorfen.

In general the best sequential treatments were the graminicides followed by acifluorfen. These treatments were not greatly more effective than mixtures of acifluorfen and the graminicides applied simultaneously but there was a slight trend for better large crabgrass and Palmer amaranth control in the sequential treatments. However, this was only significant for treatments involving xylafop mixed with acifluorfen for large crabgrass control. There was an increase in soybean injury when fluazifop was tank mixed with acifluorfen as compared to fluazifop followed by acifluorfen sequential treatment.

Large crabgrass control was not significantly enhanced for either of the four graminicides by tank mixtures or sequential treatments of acifluorfen as compared to the graminicides applied alone. When the graminicides were

applied alone, Palmer amaranth control was very low. When acifluorfen was applied alone, Palmer amaranth control was fair, but not as good as when any of the four graminicides were applied with or as sequential treatments with acifluorfen as shown at the first evaluation date. However, when analyzed at the second evaluation date none of the graminicide-acifluorfen mixtures or sequential treatments were better than acifluorfen applied alone for the control of Palmer amaranth. When applied alone, acifluorfen was no more effective when applied with the surfactant Ag98 than when applied with a crop oil concentrate added as an adjuvant. But when acifluorfen was applied without an added surfactant, both Palmer amaranth control and soybean injury decreased significantly.

All of the graminicides except fluazifop were more effective when acifluorfen was applied as a sequential treatment after rather than prior to the graminicide application. This implies that acifluorfen may have been reducing the absorption or translocation of the graminicides. Therefore, experiments were also conducted in the laboratory to determine the rates of foliar absorption and translocation of the ^{14}C-labeled graminicides with and without acifluorfen at two treatment time periods (Tables 2 through 6).

After three hours, 7.8% of the haloxyfop was absorbed into the sorghum leaf tissue. With the addition of acifluorfen the absorption dropped to 3.9% (Table 2). The

amount of haloxyfop recovered from all plant parts during this treatment period was 83%. Percent recovery increased to 105% when acifluorfen was applied with haloxyfop. The increase in percent recovery with the addition of acifluorfen may be the result of less absorption and, thus, less metabolism by the enzyme systems of the cell cytoplasm in the interior of the cell.

After treatment for 6 h there were no significant differences in haloxyfop recovery and absorption between plants treated with haloxyfop and those treated with haloxyfop and acifluorfen. There were no significant differences in percent acropetal or basipetal translocation or the apoplastic/symplastic transport ratio between haloxyfop treatments and haloxyfop with acifluorfen treatments at either 3 or 6 h after treatment.

There were no significant effects of acifluorfen among any of the reported variables on translocation and absorption of fluazifop after the 3 and 6 h treatment periods (Table 3). However, there was a trend of decreasing absorption and increasing recovery after the 3 h treatment when acifluorfen was added to fluazifop. The treatment of fluazifop also showed no significant differences in efficacy for large crabgrass control when acifluorfen was applied as sequential treatments. Apparently the absorption of fluazifop is not affected by acifluorfen as greatly as is haloxyfop.

The effect of acifluorfen on the absorption and percent recovery of xylafop was similar to haloxyfop after the 3 h treatment. Absorption of xylafop decreased from 2.2 to 1.5% and percent recovery increased from 94 to 105% when acifluorfen was added (Table 4). There also was a significant increase in basipetal translocation from 11.0 to 14.6%. After 6 h the same trends in absorption and percent recovery were present but there were no significant differences.

The percent recovery of sethoxydim after 3 h was increased with the addition of acifluorfen as was the case for all the other graminicides except fluazifop (Table 5). Absorption decreased from 9.1% to 7.8% which was not a significant decrease, but recovery did increase from 82.4% to 97.0%. Both percent acropetal and basipetal translocation increased when aciflurfen was added to sethoxydim. Acropetal translocation increased from 7.1% to 11.2% and basipetal translocation increased from 18.2% to 23.1%. In all of the experiments with the other graminicides there was a tendency for acifluorfen to cause an increase of the acropetal and basipetal translocation. Although the differences for the other graminicides were not significant, there were no cases where acifluorfen appeared to inhibit either acropetal or basipetal translocation. There was no difference in the apoplastic/ symplastic transport ratio of sethoxydim as affected by acifluorfen and this was also true for the other graminicides.

There are significant interactions of acifluorfen levels, graminicides, and time periods on percent recovery, absorption, and translocation of the graminicides when analyzed together (Table 6). The addition of acifluorfen significantly decreased percent recovery and absorption and increased acropetal translocation when analyzed as an average of treatment time and graminicides. The four graminicides also showed significant interactions. Xylafop was recovered at a slightly greater extent than haloxyfop. All four graminicides were absorbed at different levels with sethoxydim absorbed at the highest level followed by fluazifop, haloxyfop, and xylafop. Sethoxydim showed higher levels of acropetal transloction than haloxyfop. Sethoxydim had the highest rate of basipetal translocation followed by fluazifop and haloxyfop while xylafop had the lowest rate. Xylafop had a significantly higher apoplastic/symplastic transport ratio than the other graminicides. Comparisons of the two treatment time periods showed percent recovery and basipetal translocation to decrease and absorption and the apoplastic/symplastic transport ratio to increase with time.

Acifluorfen inhibited the absorption of the four graminicides while percent recovery and acropetal translocation were increased. In veiw of these effects of acifluorfen on metabolism, absorption, and translocation it appears very probable that any deliterious effects on efficacy of the graminicides would have been caused by acifluorfen inhibition of absorption of the graminicides.

Table 1. The interaction effects of acifluorfen with four graminicides on percent control of large crabgrass, Palmer amaranth, and soybean injury.[a]

Herbicide	Large Crabgrass Rating 1[b]	Large Crabgrass Rating 2[c]	Palmer amaranth Rating 1[b]	Palmer amaranth Rating 2[c]	Soybean Injury 1[b]
	----------%				----------%
Alone					
Xylafop	91b-e	85a-d	3h	0h	13c-e
Fluazifop	85ef	93ab	1h	3h	3f
Sethoxydim	86d-f	94ab	0h	0h	3f
Haloxyfop	88c-e	91a-c	0h	5h	3f
Acifluorfen	5g	0f	75f	80b-d	20a-c
Ac.[d] + Ag98	5g	3f	87b-d	87a-d	14c-e
Mixed					
Xylafop + Ac.	90c-e	88a-c	86c-e	53f	26a
Fluazifop + Ac.	89c-e	84a-d	90b-d	70de	25ab
Sethoxydim + Ac.	85ef	80b-d	90b-d	65ef	25ab
Haloxyfop + Ac.	95a-c	95ab	95ab	76c-e	19a-c
Sequential					
Ac.; Xylafop	80f	71d	84e	38g	10ef
Ac.; Fluazifop	88c-e	75cd	93a-c	65ef	11de
Ac.; Sethoxydim	80f	48e	89b-d	73c-e	15c-e
Ac.; Haloxyfop	84ef	83b-d	91b-d	69de	9ef
Sequential					
Xylafop; Ac.	96ab	94ab	93a-c	64ef	20a-c
Fluazifop; Ac.	91b-e	83b-d	94ab	70de	15c-e
Sethoxydim; Ac.	89b-e	81bcd	94ab	75c-e	18b-d
Haloxyfop; Ac.	94a-d	100a	94ab	86bc	14c-e
Additional treatments					
Ac. w/o COC	3g	3f	55g	55f	1g
Weed free	100a	100a	100a	100a	0g
Weedy	0g	0f	0h	0h	0g
LSD (0.05)	7	16	7	13	7

[a](Values followed by the same letter are not significantly different at the 0.05 level.)
[b]Rating recorded on June 25,1985
[c]Rating recorded on July 10,1985
[d]Ac.=Acifluorfen

TABLE 2. The effects of acifluorfen[a] on absorption and translocation of haloxyfop[b] after 3 and 6 hour treatment periods.[c]

Herbicide	Recovery	Absorption	Translocation Acro-petal	Translocation Basi-petal	Apoplatic/ Symplastic Transport Ratio
	----------%		----------		
Three hour treatment					
Haloxyfop	83.3b	7.8a	3.8a	18.7a	0.27a
Haloxyfop + Acifluorfen	105.2a	3.9b	6.5a	21.0a	0.45a
LSD (0.05)	17.7	2.8	4.3	19.3	0.32
Six hour treatment					
Haloxyfop	79.0a	7.5a	4.2a	7.2a	0.61a
Haloxyfop + Acifluorfen	92.1a	4.4a	5.1a	7.5a	0.69a
LSD (0.05)	13.6	4.2	2.1	1.6	0.30

[a]Acifluorfen, 7.1 µg
[b]Haloxyfop, 57.9 µg (3 h); 8.10 µg (6 h)
[c]Values followed by the same letter are not significantly different at the 0.05 level.

TABLE 3. The effects of acifluorfen[a] on absorption and translocation of fluazifop[b] after 3 and 6 hour treatment periods.[c]

Herbicide	Recovery	Absorption	Translocation Acro-petal	Translocation Basi-petal	Apoplatic/ Symplastic Transport Ratio
	----------%		----------		
Three hour treatment					
Fluazifop	90.1a	7.4a	4.7a	19.7a	0.32a
Fluazifop + Acifluorfen	94.9a	5.6a	5.3a	22.0a	0.35a
LSD (0.05)	9.8	3.2	2.5	17.3	0.41
Six hour treatment					
Fluazifop	85.8a	9.1a	6.4a	11.2a	0.59a
Fluazifop + Acifluorfen	90.5a	8.4a	10.4a	11.1a	0.92a
LSD (0.05)	8.9	1.5	7.4	3.8	0.52

[a]Acifluorfen, 3.57 µg
[b]Fluazifop, 4.14 µg (3 h); 4.93 µg (6 h)
[c]Values followed by the same letter are not significantly different ath the 0.05 level.

Table 4. The effects of acifluorfen[a] on absorption and translocation of xylafop[b] after 3 and 6 hour treatment periods.[c]

Herbicide	Recovery	Absorption	Translocation Acro-petal	Translocation Basi-petal	Apoplatic/ Symplastic Transport Ratio
	----------%----------				
Three hour treatment					
Xylafop	94.1b	2.2a	6.8a	11.0b	0.55a
Xylafop + Acifluorfen	105.3a	1.5b	10.2a	14.6a	0.24a
LSD (0.05)	7.5	0.45	2.6	3.9	0.33
Six hour treatment					
Xylafop	85.3a	3.7a	3.9a	4.7a	0.79a
Xylafop + Acifluorfen	93.3a	2.9a	7.0a	6.4a	1.5a
LSD (0.05)	17.5	1.0	7.5	2.4	1.6

[a]Acifluorfen, 9.18 µg
[b]Xylafop, 8.22 µg (3 h); 11.21 µg (6 h)
[c]Values followed by the same letter are not significantly different at the 0.05 level.

TABLE 5. The effects of acifluorfen[a] on absorption and tanslocation of sethoxydim[b] after 3 and 6 hour treatment periods.[c]

Herbicide	Recovery	Absorption	Translocation Acro-petal	Translocation Basi-petal	Apoplatic/ Symplastic Transport Ratio
	----------%----------				
Three hour treatment					
Sethoxydim	82.4b	9.1a	7.1b	18.2b	0.39a
Sethoxydim + Acifluorfen	97.0a	7.8a	11.2a	23.1a	0.49a
LSD (0.05)	5.8	1.7	3.8	3.9	0.13
Six hour treatment					
Sethoxydim	92.6a	11.2a	8.7a	20.0a	0.47a
Sethoxydim + Acifluorfen	89.3a	8.7a	7.0a	20.5a	0.39a
LSD (0.05)	5.8	5.2	2.2	4.2	0.15

[a]Acifluorfen, 7.00 µg
[b]Sethoxydim, 5.58 µg (3 h); 8.87 µg (6 h)
[c]Values followed by the same letter are not significantly different at the 0.05 level.

TABLE 6. The interaction of acifluorfen, graminicides, and treatment time on absorption and translocation graminicides.[a]

Treatment	Recovery	Absorption	Translocation Acro-petal	Translocation Basi-petal	Apoplatic/ Symplastic Transport Ratio
	----------%----------				
Acifluorfen					
-Acifluorfen	86.4b	7.2a	5.7b	13.8a	0.50a
+Acifluorfen	95.9a	5.4b	7.8a	15.8a	0.62a
LSD (0.05)	3.3	0.93	1.8	2.5	0.19
Graminicide					
Haloxyfop	89.6b	5.9c	4.9b	13.6b	0.50ab
Fluazifop	90.3ab	7.6b	6.7ab	16.0b	0.54ab
Xylafop	94.5a	2.6d	7.0ab	9.1c	0.76a
Sethoxydim	90.3ab	9.2a	8.5a	20.4a	0.44b
LSD (0.05)	4.6	1.3	2.6	3.5	0.27
Time					
3 Hour	93.9a	5.7b	6.9a	18.5a	0.38b
6 Hour	88.5b	7.0a	6.6a	11.1b	0.74a
LSD (0.05)	3.3	0.93	1.8	2.5	0.19

[a]Values followed by the same letter are not significantly different at the 0.05 level.

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PART II

TRANSLOCATION OF AUXINS AS AFFECTED BY ACIFLUORFEN

Translocation of Auxins as Affected by Acifluorfen

Abstract. The effects of acifluorfen [5-(2-chloro-4-(trifluoromethyl)phenoxy)-2-nitrobenzoic acid] on auxin translocation were determined. The first objective was to determine the possible necessity of an auxin protein carrier as a mediator of acifluorfen effects on auxin translocation. The second objective was to determine the effects of nutrient source on the acifluorfen-mediated changes in auxin translocation patterns.

Nine-day-old bean (Phaseolus vulgaris (L.) cv Kentucky Wonder) seedlings were treated by injecting 1-µl amounts of 95% ethanol containing various concentrations of ^{14}C-labeled auxins and 25 µg of acifluorfen into the stem at the cotyledonary node. Plants were harvested 4 h after treatment, divided into various plant parts, and analyzed for ^{14}C.

Acifluorfen enhanced acropetal apoplastic translocation of indole-3-acetic acid (IAA) and inhibited basipetal symplastic translocation of the auxin. The effects appeared to be due to an effect of acifluorfen on vein unloading rather than an effect on vein loading of the auxin. The effects of acifluorfen on IAA translocation were equally as large in the presence of high concetrations of IAA as in the presence of small amounts of IAA, indicating that the acifluorfen effects were not mediated through carrier-mediated auxin transport systems. The effects of acifluorfen on IAA and 2,4-dichlorophenoxyacetic acid (2,4-D) translocation were much greater when plants were grown for 4 h prior to treatment in nutrient solutions containing ammonium rather than nitrate nitrogen. This effect may implicate changes in cytoplasmic pH as the mechanism being affected by acifluorfen.

Additional index words: nitrogen, nutrient source, protien carrier, IAA, 2,4-D.

INTRODUCTION

In 1969 Matsunaka (17) reported that diphenyl ether herbicides could be classified into two groups based on their light requirement for activation. He found that ortho-substituted diphenyl ethers are active only in the light but that meta-substituted diphenyl ethers are active in the light and dark. The acitvation by light is thought to be a photobiochemical process. Xanthophyll pigments from susceptible yellow mutants of rice plants appeared to make an important contribution to the photoactivation of ortho-substituted diphenyl ether herbicides acting as the acceptor of the light energy involved in the activation.

The exact mode of action of the ortho-substituted diphenyl ether herbicide acifluorfen is still unknown. The toxic effect in green plants is apparently initiated by absorption of light (6,20), although Lambert et al. (11) found that acifluorfen affected auxin translocation in the dark as effectively as in the light. It has been suggested that acifluorfen is activated in light by yellow pigments and is then involved in the initiation of a free radical chain reaction (19) especially involving cell membranes. This theorectically results in a loss of the membrane's selective permeability characteristics, thereby leading to cell death.

Kunert and Boger (9) reported in 1985 that diphenyl ether herbicides such as acifluorfen initiate lipid peroxidation in higher plants in the light. This peroxidation of polyunsaturated fatty acids is one of the most deteriorative reactions that can damage biomembranes of plant cells. They also found that the biotransformation of 1-galactano-1,4-lactone to vitamin C is a potent natural antidote against the peroxidation. Studying lipid peroxidation by oxyfluorfen [2-chloro-1(3-ethoxy-4-nitrophenoxy)-4-(trifluoromethyl)benzene], Kunert et al. (10) reported that under peroxidizing conditions, destruction of the alga Scenedesmus acutus cytochrome c is significantly higher than destruction of membrane-bound components, such as cytochrome f and chlorophyll.

Acifluorfen is the first compound reported (5) to induce tissue-specific isoflavonoid glucosides and key enzymes of their biosynthesis in any plant. Mature soybean (Glycine max) leaves normally contain kaempferol-3-glycoside but they accumulate no other flavonoids. Whole leaves sprayed with acifluorfen and maintained in the light developed smallnecrotic lesions and accumulated isoflavone aglycones, isoflavone glucosides, and pterocarpans.

In 1970 Moreland et al. (18) reported the effects of the three diphenyl ether herbicides nitrofen [2,4-dichloro-1-(4-nitrophenoxy)benzene), MC-1478 [2,4,6-trichlorophenyl-4'-nitrophenyl ether], and fluorodifen [2-nitro-1-(4-nitrophenoxy)-4-trifluoromethylbenzene] on phosphorylation

and electron transport in spinach (Spinacia oleracea L.). All three herbicides acted primarily as inhibitors of chloroplast noncyclic electron transport and the coupled photophosphorylation. A site of action close to light reaction II was suggested.

Acifluorfen has been shown to be inhibitory to those photosynthetic functions that require a functioning chloroplast envelope and stroma (26). This herbicide inhibits the unloading of electrons from cytochrome f in the chloroplast (20). Although diphenyl ether herbicides block electron transport (3), this apparently is not the mode of action of acifluorfen since light activation of the herbicide does not require photosynthetic electron transport (2,19,20). Kenyon et al. (8) in 1985 reported that although photosynthesis is affected early in the ontogeny of acifluorfen damage, it is probably due to chloroplast envelope breakage and not due to direct interaction with photosynthesis.

Hawton and Stobbe (7) proposed that a light-controlled biochemical process causes a diphenyl either such as nitrofen to polymerize which would enable it to combine with cell lipids that could affect certain membrane properties. Leong and Briggs (13) concluded that acifluorfen enhances the blue-light induced absorbance change in Triton X100-solubilized crude membrane preparations from etiolated oat coleoptiles. Enhancement of the spectral change is

correlated with a change in rate of dark reoxidation of a b-type cytochrome.

Although first positive curvature in oat (Avena sativa L.) coleoptiles is known to be mediated by lateral transport of auxins (2), it is unknown how the mechanism for this transport is induced. Acifluorfen sensitizes phototropism in dark-grown oat seedlings such that the first positive response occurs with blue light fluences as little as one-third of those required to elicit the same response in seedlings grown in the absence of the herbicide.

Shimabukuro et al. (24) found that the IAA-induced acidification in peeled coleoptiles of wheat (Tritcum aestivum (L.) cv Waldron) and oat was inhibited by the diphenyl ether herbicide diclofop-methyl [(2-(4-(2,4-dichlorophenoxy)phenoxy)propanoic acid, methyl ester] during a 3 to 4 h period. Shimabukuro et al. (25) in other research also reported that changes in net proton flux (external pH) corresponded to the depolarization of the cell membrane potential by 50 μM diclofop-methyl. This was followed by a repolarization response within 30 to 40 s after removal of diclofop-methyl and the addition of 10 μM IAA or 2,4-D.

Therefore, it is possible that part of the mode of action of acifluorfen is due to the inhibition of proton extrusion by blocking a b-type cytochrome in the electron transport chain and by acidification in the cell cytoplasm.

It is well known that auxin absorption is dependent on the maintenance of pH and electrical gradients in plant tissues (12,21,22). Schweizer et al. (23) found that nitrate and ammonium nutrient nitrogen sources have different effects on cell pH. Since diphenyl ethers are also known to collapse pH gradients (24), it appears that research should be done on the interaction of acifluorfen with the two nitrogen sources as they relate to the translocation of auxins.

Mangeot et al. (16) found that translocation of ^{14}C acifluorfen in ivyleaf morningglory (Ipomoea hederacea (L.) Jacq # IPOHE) was mainly acropetal and the herbicide accumulated in the meristematic tissue. Soybeans retained the radioactivity at the sites of initial contact of the treatment solution. In 1979 Mangeot and Rieck (15) reported that treated leaflets of soybeans contained 77 percent of the applied ^{14}C after 24 h with no further increases at 48 or 96 h samplings. Autoradiographs revealed translocation of the ^{14}C acifluorfen to the stem and plant shoot above the treated leaves.

Basler et al. (1) and Long and Basler (14) showed that stem injection used to apply ^{14}C-labled auxins to plants resulted in acropetal translocation with only low levels of basipetal movement. Acifluorfen mixed with 2,4,5-T-1-^{14}C showed increased 2,4,5-T-1-^{14}C translocation out of the treated area of stem injected bush bean seedlings, especially into the large primary leaves (11). This

translocation pattern is indictive of apoplastic translocation and suggests that acifluorfen inhibited vein loading or enhanced vein unloading of the auxin. Previous work (24) also showed that acifluorfen affected translocation of different auxins such as indole-3-acetic acid (IAA).

The research reported here was conducted to obtain additional information on the effects of acifluorfen on auxin translocation.

MATERIALS AND METHODS

Auxin translocation experiments were conducted using bush bean. The seeds were germinated in vermiculite moistened with one-half strength Hoaglands nutrient solution and grown for 4 days in complete darkness and 1 day under continuous light of 100 $\mu E \cdot m^{-2} \cdot s^{-1}$ at 32° C. At this time the bean seedlings were transferred to amber jars containing 400 ml of aerated, one-half strength Hoaglands nutrient solution and grown 4 days in the controlled temperature chamber at 29° C with 14 h days and light intensity of 223 $\mu E \cdot m^{-2} \cdot s^{-1}$ and 10 h nights at 27° C.

The 9 day old bean seedlings were then treated at 10:00 AM by injecting 1-µl of 95% ethanol containing 0.5 µg of 2,4-D-1-^{14}C (Amersham, 8.9 Ci/M), 0.25 µg of IAA-1-^{14}C (Amersham, 61 Ci/M), or 0.25 µg of IAA-1-^{14}C (Amersham, 40 Ci/M) and 20 µg unlabeled technical acifluorfen into the pith of the stem at the cotyledonary node. In some experiments the level of IAA injected was increased to 25 µg per plant by adding unlabeled IAA to the solution being injected. The syringe needle was inserted into the cotyledonary node and pushed down the hollow in the pith area about 1 cm where the chemicals were then deposited. Immediately following injection, plants were returned to the growth chamber.

After a treatment period of 4 h the plants were removed and dissected into various plant parts. These included the young shoot, primary leaves including petioles, epicotyl, 2 cm treated area including stem tissue 0.5 cm above the cotyledonary node down to 1.5 cm below the cotyledonary node, hypocotyl, and roots. A 5 ml aliquot of the nutrient solution was also taken for ^{14}C analysis.

The plant parts were lyophilized, weighed, and homogenized in 5 or 10 ml of 95% ethanol. An aliquot of the homogenate was removed and analyzed for ^{14}C by liquid scintillation spectrophotometry.

Experiments were also conducted to investigate the effect of nutrient solution nitrogen source on the acifluorfen-mediated changes in auxin translocation patterns. For these experiments the procedure was the same as stated for the previous experiment except for a 4 h pretreatment of one-fourth strength Hoaglands nutrient solutions containing 10 mM ammonium acetate, 10 mM ammonium bicarbonate, or 10 mM potassium nitrate as the nitrogen source. These were then compared to plants grown in one-fourth strength Hoagland's nutrient solution as the control. Additional carbon dioxide was mixed with air for aeration of the treatment using ammonium bicarbonate to bring the pH of the nutrient solution to 6.2 which is normal for the control nutrient solution.

The data collected was disintegrations per minute (dpm) of the ^{14}C in the homogenized plant part. This was then

converted to percent recovery, which is expressed as a percentage of the amount of ^{14}C applied; vein loading; and vein unloading.

When auxins are injected into the center of a bean stem they have to translocate radially to the vascular bundles before they can move out of the treated area. This translocation has to occur through the apoplast, between the cells in the cell wall area, or directly through the living cytoplasm or symplast of each cell. Apparently auxins do not translocate through the apoplast or cell wall area because metabolic inhibitors such as dicyclohexylcarbodi-imide (DCCD) have been shown to inhibit the movement of auxins out of the treated area (4).

Vein loading is represented by the following equation:

$$\text{Vein Loading} = \frac{\text{Total } ^{14}C \text{ recovered} - \text{Treated area } ^{14}C}{\text{Total } ^{14}C \text{ recovered}} \times 100$$

The percent translocation out of the treated area can be accredited to cell or vein loading because metabolic inhibitors such as DCCD were shown to greatly inhibit translocation of some compounds out of the treated area (4). Since metabolic inhibitors would not inhibit simple diffusion in the apoplast, this type of inhibition must be due to inhibition of cell or vein loading.

Vein unloading is represented by the following equation:

$$\text{Vein unloading} = \frac{\text{Primary leaves } {}^{14}C}{\text{Hypocotyl } {}^{14}C + \text{Roots } {}^{14}C + \text{Nut. Soln. } {}^{14}C}$$

This calculation is an expression of the apoplastic/ symplastic transport ratio and can be used as a measure of cell or vein unloading because metabolic inhibitors such as DCCD were also shown to enhance, rather than inhibit, apoplastic translocation of the auxin IAA to the large primary leaves and inhibit basipetal symplastic translocation. Since metabolic inhibitors cannot enhance simple diffusion in the apoplast, this type of enhancement must be due to enhanced unloading of cells.

DISCUSSION AND RESULTS

Previous research (11) showed that acifluorfen drastically affected the translocation of auxins in intact bean seedlings. The primary effect of acifluorfen was one in which vein unloading was increased. However, in some treatments vein loading was also increased. The first objective of this research was to determine the possible necessity of an auxin protein carrier as a mediator of acifluorfen effects on auxin translocation. If an auxin carrier is involved, the carrier should be saturated at high auxin concentrations and acifluorfen should have little effect on translocation.

The effects of low and high concentrations of unlabeled acifluorfen and indole-3-acetic acid on the translocation of radiolabeled IAA are shown in Table 1. At the low IAA concentration both vein loading and unloading were significantly affected with the addition of 25 µg of acifluorfen. Vein loading increased from 46% to 55% with acifluorfen. Vein unloading as indicated by the apoplastic/symplastic transport ratio, increased from 0.03 to 0.20. There was no difference in percent recovery between the two levels of acifluorfen although it appears as if the addition of acifluorfen caused less recovery of the labeled IAA.

There were significant interactions of IAA levels and acifluorfen effects on loading and unloading of IAA. Using the high concentration of IAA, the same trends with even greater rather than smaller differences were observed for loading and unloading of IAA as affected by acifluorfen. Vein loading exhibited a large increase from 46% to 79% with acifluorfen treatment. There also was a large increase in vein unloading from 0.10 to 1.0. This is equivalent to a 1070% change in vein unloading which is significantly greater than the 597% change that ocurred when acifluorfen was applied in the presence of low IAA levels. The implication is that acifluorfen affects simple diffusion of auxins rather than carrier-mediated auxin transport.

Shimabukuro et al. (25) reported antagonistic effects of auxins such as 2,4-dichlorophenoxyacetic acid (2,4-D) and indole-3-acetic acid (IAA), and diclofop-methyl which is a diphenyl ether herbicide, on growth, pH, and electropotential gradients across the plasma membrane. Diclofop-methyl appeared to act by reducing the electropotential gradient while the auxins tended to reestablish the gradient. This appeared to be the basis for the antagonistic herbicidal effects of the auxin 2,4-D and diclofop-methyl. Thus, one would expect antagonistic effects of high auxin concentrations and acifluorfen, which is also a diphenyl ether herbicide, on the translocation of auxins such as IAA. The expected results were not observed in as much as high auxin concentrations caused even greater

acifluorfen effects especially on vein unloading. This draws in to question the conclusion that auxins act antagonistically because they have antagonistic effects on pH and electropotential gradients. Electropotential and pH gradients are most likely to be the factors intimately involved in the control of loading and unloading of auxins.

If diclofop-methyl antagonized IAA activity by collapsing the electrogenic proton gradient across the plasmalemma, this may cause the perturbation of numerous cellular functions with serious physiological consequenses. But, Shimabukuro et al. (25) stated that the antagonistic interaction between IAA and diclofop-methyl was indirect because fusicoccin-induced coleoptile growth and cell wall acidification was also inhibited by diclofop-methyl.

The second objective was to determine the effects of nutrient nitrogen source on the acifluorfen-mediated changes in auxin translocation patterns. Previous work by Shimabukuro (24) on diphenyl ether herbicides implies that diphenyl ethers inhibit proton secretion and the buildup of a pH gradient. Other recent work has shown that nutrient solution nitrogen source also affects cellular pH gradients (23). Since auxin uptake into cells is well known to be influenced by pH gradients, the nitrogen source could be expected to interact with the diphenyl ether herbicide acifluorfen in its effect on auxin translocation.

The effects of nitrogen source and acifluorfen on the translocation of IAA and 2,4-D can be seen in Tables 2 and

3. When a one-fourth strength Hoagland's nutrient solution (4 mM nitrate) in addition to 10mM potassium nitrate was used, 25 µg of acifluorfen caused no significant effects on vein loading, vein unloading or percent recovery of 0.5 µg IAA (Table 2). When 10 mM ammonium acetate and one- fourth strength Hoagland's nutrient solution was used as the nitrogen source, there were no differences in vein loading and vein unloading from the values found when nitrate was used as the nitrogen source. However, when acifluorfen was applied, both vein loading and unloading were higher than when 10 mM potassium nitrate was used. Vein unloading increased from 0.05 to 0.27 and vein loading increased from 54% to 65%.

Three micrograms of the auxins IAA and 2,4-D were applied in the experiment described in Table 3. The high auxin levels increased the values for vein loading and unloading as was shown in Table 1 when high auxin levels were used. There was a significant (0.03 level) interaction of nutrient nitrogen source and acifluorfen effects on auxin vein unloading. Acifluorfen increased vein unloading of 2,4-D and IAA most when ammonium bicarbonate rather than potassium nitrate was used as the nitrogen source. This observation substantiates the conclusion drawn from the data of Table 2 that ammmonium rather than acetate was responsible for the increased acifluorfen effect. Acifluorfen, in the presence of ammonium bicarbonate, increased vein unloading of IAA 189% while vein unloading in

the presence of potassium nitrate was not significantly increased. The effect on vein unloading of 2,4-D was also significant as the increase was 177% after acifluorfen treatment in the presence of ammonium bicarbonate and there was no significant effect in the presence of potassium nitrate. Vein loading of the auxins was not affected by acifluorfen. Percent recovery was not greatly affected by acifluorfen but was slightly decreased for IAA and 2,4-D in the presence of ammonium bicarbonate.

Since high ammonium nutrition is thought to decrease the pH of the cytoplasm, it appears that acifluorfen interacts in pH effects to bring about the changes in auxin translocation patterns. Acifluorfen may possibly have more extensive effects when the cytoplasm pH is low and this may have been one reason why acifluorfen was more effective in the presence of high auxin concentrations. Fusicoccin, which like auxin causes proton excretion, also was shown to cause acidification of the cytoplasm (25). It appears possible that high concentrations of auxin may also cause cytoplasmic acidification and, thus, would act in the same way as high ammonium nutrition. The addition of acifluorfen tended to decrease the percent recovery of ^{14}C of the radioactive auxin.

In summary the greatest general effects of acifluorfen appear to cause vein unloading of auxins. The effects of acifluorfen on vein unloading were always greatest in treatments where it would be expected that the cytoplasmic

pH would be low, i.e., with ammonium nutrition or high auxin levels. This prominent interaction of acifluorfen with pH implies that acifluorfen effects on pH gradients may be responsible for the effects on auxin translocation.s

Table 1. The effects of 25 µg of acifluorfen and 25 µg of unlabeled IAA on the translocation of 0.25 µg of IAA-1-^{14}C during a 4 hour treatment period.[a]

Treatment	Percent recovery	Vein loading (%)	Vein unloading (apoplastic/ symplastic transport ratio)
0 µg unlabeled IAA			
0 µg acifluorfen	78b	46a	0.03a
25 µg acifluorfen	70a	55b	0.20b
25 µg unlabeled IAA			
0 µg acifluorfen	82b	46a	0.10ab
25 µg acifluorfen	83b	79c	1.5c
LSD (0.05)	7	4	0.12

[a]LSD (0.05) Significant differences between acifluorfen levels.

Table 2. The effects of nitrogen source and 25 µg of acifluorfen on the translocation of 0.5 µg of IAA-1-^{14}C during a 4 hour treatment period.[a]

Treatment	Percent recovery	Vein loading (%)	Vein unloading (apoplastic/ symplastic transport ratio)
10 mM potassium nitrate			
0 µg acifluorfen	73b	59ab	0.06ab
25 µg acifluorfen	66ab	54a	0.13b
10 mM ammonium acetate			
0 µg acifluorfen	71b	54a	0.05a
25 µg acifluorfen	63a	65b	0.27c
LSD (0.05)	8	8	0.08

[a]Values followed by the same letter are not significantly different at the 0.05 level.

Table 3. The effects of nitrogen source and 25 µg of acifluorfen on the translocation of 3.0 µg of IAA-1-^{14}C and 3.0 µg of 2,4-D-1-^{14}C during a 4 hour treatment period.[a]

Treatment	Percent recovery	Vein loading (%)	Vein unloading (apoplastic/ symplastic transport ratio)
IAA with 10 mM potassium nitrate			
0 µg acifluorfen	87bc	55b	0.47b-e
25 µg acifluorfen	82bc	55b	0.52d-f
IAA with 10 mM ammonium bicarbonate			
0 µg acifluorfen	77b	50ab	0.46b-d
25 µg acifluorfen	66a	46ab	0.87g
2,4-D with 10 mM potassium nitrate			
0 µg acifluorfen	90c	47ab	0.18a
25 µg acifluorfen	85bc	51ab	0.30ab
2,4-D with 10 mM ammonium bicarbonate			
0 µg acifluorfen	92c	48ab	0.31a-c
25 µg acifluorfen	78b	45a	0.55d-f
LSD (0.05)	10	9	0.21

[a]Values followed by the same letter are not significantly different at the 0.05 level.

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