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Effect of Atrazine and its Carriers on the Grain Sorghum Plant

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EFFECT OF ATRAZINE AND ITS CARRIERS

ON THE GRAIN SORGHUM PLANT

BY

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This thesis is approved as a creditable and independent investigation by a candidate for the degree, Master of Science, and is acceptable as meeting the thesis requirements for this degree, but without implying that the conclusions reached by the candidate are necessarily the conclusions of the major department.

Thesis Adviser　　　　Date

Head, Plant Science Department　　　　Date

2661-11

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INTRODUCTION

The addition of crop oils to post-emergent applications of atrazine has enhanced the weed control properties of the atrazine. This treatment has been used very effectively for the control of annual weeds in corn and sorghum. Because the treatment was applied post-emergence, it could be employed after the farmer found he would have difficulty controlling annual weeds. The treatment was, then, an ideal salvage measure for the farmer who could not cultivate his field because of rainy weather. The atrazine plus oil treatment appeared to work better under rainy weather conditions than it did under dry weather conditions.

A rainy season in the southeastern part of South Dakota in 1967 caused many farmers to turn to atrazine plus oil to control the annual weeds they could not cultivate out of their corn and sorghum fields. Many farmers who used the atrazine plus oil treatment had satisfactory weed control and received no apparent injury to their crops. In other instances, sorghum fields sprayed with atrazine plus oil were severely injured. Most of this injury was confined to stunting and delay in maturity.

Because of such erratic results, a survey was conducted by Parker and Stritzke (40) to determine the extent of the injury and to study the type of weather conditions prior to and immediately following spraying. It appeared that the most severe injury was preceded by at least three days of cool weather when temperatures were less than seventy degrees Fahrenheit.

The injury to the sorghum raised questions about the effects of atrazine and its carriers on the sorghum plant. A study was initiated to measure the physiological responses of the sorghum plant and to note the gross physical responses of the plant in height, weight, and grain yield.

LITERATURE REVIEW

The triazine compounds have been primarily used as preemergent herbicides. They are readily absorbed by the root of plants and translocated to the leaves via the apoplast (13). Research workers recently have been experimenting with post-emergent applied triazines. In work done by Crafts (10) it was found that when simazine is applied to the leaf surface it penetrates the cuticle rapidly but moves in the leaf in only acropetal direction. Many research workers have found that the addition of a non-phytotoxic crop oil to the atrazine suspension has increased the weed control properties of the treatment (4, 8, 11, 14, 15, 26, 27, 31, 32, 39, 41, 47, 49, 50). Strand (39) found in his work that atrazine plus emulsifier was not as effective as atrazine plus oil with an emulsifier added, and that ten percent oil in the total spray solution was sufficient to give maximum weed control. In Smith's work (38) with C^{14} labeled atrazine, it was found that the addition of crop oil to the atrazine-water suspension increased the uptake of the atrazine by the leaf of foxtail plants.

The addition of oils or emulsifiers has an effect on the spray solution as well as on the leaf which is sprayed. Sargent and Blackman (33) found that the surface tension was most reduced by the addition of 0.1 percent emulsifier. Temple and Hilton (41) reported in their work done with thirty different emulsifiers and two triazines (ametryne and atrazine) that the triazines were most soluble in cationic emulsifiers, less soluble in anionic and even less soluble in nonionic types.

Surfactants have been reported to increase the herbicidal activity of many herbicides (3, 17, 18, 19, 20, 21). Ilnicki, et al., (25) found that the addition of surfactants increased the toxicity of atrazine to broad-leaf weeds and Biswas (5) reported that a surface-active agent increased foliar absorption of atrazine in detached whole peanut leaves.

Currier and Dybing (12) attributed the greater uptake of herbicides with surfactants to the following factors: 'a) improved coverage; b) removing air films between spray and leaf surface; c) reducing interfacial tensions between relatively polar and apolar submicroscopic regions of the cuticle; d) inducing stomatal entry; e) increasing the permeability of the plasma membrane, through stimulation or incipient toxicity; f) facilitating cell wall movement in the region of the wall-cytoplasm interface; g) acting as co-solvents; h) interacting directly with herbicide in some manner; i) acting as humectant secondarily.'

There are many environmental factors which tend to increase or decrease the uptake of various herbicides applied to plants grown under field conditions. Currier and Dybing (12) indicated that high relative humidity would prevent moisture stress in the plant, favor opening of the stomatal cavity, delay drying of the spray droplet and increase permeability of the cuticle.

Hammerton (22) indicated that high humidity and high temperatures in general increased susceptibility of plants to herbicides. Hammerton also found that low light intensity before and high intensity after spraying appear to increase susceptibility. He also discussed the relationship of the stage of growth and susceptibility.

Bukovac and Norris (6) found that by removing the wax from a plant leaf the penetration of the cuticle by water was aided. They also suggest that the chemical composition, in addition to the overall quantity of cuticular wax, may be of major importance. Holloway (24) found that the hydrophobic properties of waxes depend upon their chemical makeup and also upon the orientation of the molecules of their constituents. He also found that the wettability of wax constituents increases with the increase of polarity. Skoss (19) found that a thicker cuticle was produced at a median temperature and the greatest percentage of wax was produced at a high temperature. The plants undergoing water stress produced cuticles containing a greater portion of waxes than did plants with more favorable moisture conditions. Skoss also stated that permeability of a cuticle to water in an in vitro system was directly correlated with the amount of impregnating waxes. Dexter, Burnside and Lavy (14) found that the toxicity of atrazine plus surfactants increased as the relative humidity increased on emerged crabgrass (Digitaria sanguinalis).

Higher uptake and translocation of foliar atrazine on quackgrass (Agropyron repens) as the temperature increased from 68 to 80 degrees Fahrenheit was reported by Wax and Behrens (44). Stritzke and Parker (40) reported that the cases of injury to sorghum fields in South Dakota were associated with temperatures below seventy degrees Fahrenheit and with high relative humidity.

Plants capable of breaking down atrazine are tolerant to it. Negi et al. (29) reported that johnsongrass, grain sorghum and corn converted twice as much atrazine to hydroxyatrazine as did susceptible plants like oats, soybeans and beans. Wheeler and Hamilton (46) indicated that after prolonged treatment of root solutions of tolerant species, maize and sorghum had accumulated concentrations of unaltered atrazine in the leaves, comparable to those found in the sensitive species of wheat at the point of acute toxicity.

Shimabukuro (35) reported that the metabolism of atrazine by corn and sorghum was not the same. In corn, atrazine metabolized via both the 2-hydroxylation and N-dealkylation pathways, while the sorghum plants were only capable of the N-dealkylation pathway. Because the sorghum plant is only capable of the N-dealkylation pathway, the metabolism of atrazine is slower than in the corn plant. If atrazine is not broken down in the plant, its mode of action is to block the Hill reaction and thus prevent the CO_2 uptake of the susceptible plant. Some researchers have made use of the fact that the triazines block the Hill reaction by measuring the amount of $C^{14}O_2$ taken up by treated and untreated leaves (38, 9, 2).

In his work on uptake of atrazine and of atrazine plus oil combinations, Smith (38) used C^{14} atrazine to measure the uptake of atrazine by grasses. Couch and Davis (9) used $C^{14}O_2$ to measure the CO_2 uptake of corn, cotton and soybeans treated with atrazine, bromacil and diquat. Moss (28) used a method of measuring the CO_2 uptake of tolerant and susceptible plants and tried to correlate the rate of CO_2 uptake of these plants with their tolerance to the triazine herbicides. Olech (30) used a method of measuring the amount of CO_2 taken out of a standard gas as it was passed over the sample of treated intact tissue. An infra-red CO_2 gas analyzer measured the amount of CO_2 in the air sample. Burnside, Wicks and Fenster (7) measured the effects of atrazine plus surfactants on grain sorghum yields grown under field and growth chamber conditions.

MATERIALS AND METHODS

I. LABORATORY STUDIES

General Procedure

Background Information. Triazine herbicides inhibit the Hill reaction and thus limit assimilation of CO_2. A quantative measurement of this effect is obtained by measuring the net CO_2 uptake of the sorghum leaves treated with atrazine and its carriers.

The net CO_2 uptake was measured periodically after spraying of leaves which were unrolled and leaves which were not yet unrolled at the time of spraying. Plants grown under different light and temperature conditions were also evaluated to determine the effects that these environmental conditions had on the net CO_2 uptake rate following spraying.

Growing Sorghum Plants. The NK-133 hybrid grain sorghum seeds were placed in a germinator at 80 degrees Fahrenheit for two days. Uniform sprouts were then selected and planted in pots containing 500 grams of a sand-soil mix. The plants were then grown under constant temperature and light intensity with a sixteen-hour day. The plants were sprayed when the third leaf was fully unrolled and expanded.

Spraying. The plants were sprayed with atrazine and its carriers with a spray-chamber machine equipped with a tee-jet nozzle, applying twenty gallons of spray solution per acre. The soil in the pots was covered with perlite before spraying to prevent the spray solution from reaching the soil surface. After the spraying was completed, the perlite was poured off the soil to prevent the treatments from being leached to the rooting zone of the sorghum plants.

Sampling. One plant from each pot was harvested immediately after spraying. Leaves selected for analysis were excised from the plant near the leaf collar. The cut ends of the leaves were placed in plastic embedding capsules filled with distilled water. This maintained the leaves in a turgid condition during the period of analysis. The embedding capsule and the analyzed leaves are represented in (Figure 1).

Analysis and Equipment. The leaves were then placed in a machine equipped to measure the rate of net CO_2 uptake of the intact leaves. Figure 2 illustrates laboratory equipment used.

Glass tubes were suspended in a water bath equipped with a unit that cooled the water when illuminating lights were turned on. The bath was equipped with a plexiglass bottom and a bank of lights located below the bath. The leaves were placed in the tubes and were illuminated from both top and bottom by 4500 foot-candles of light. Nine tubes were suspended in the water bath; eight of these tubes were used for samples and one was used as a blank tube for standard reference. The incoming standard gas was divided in a copper coil manifold in the water bath, and was then delivered to the glass sample

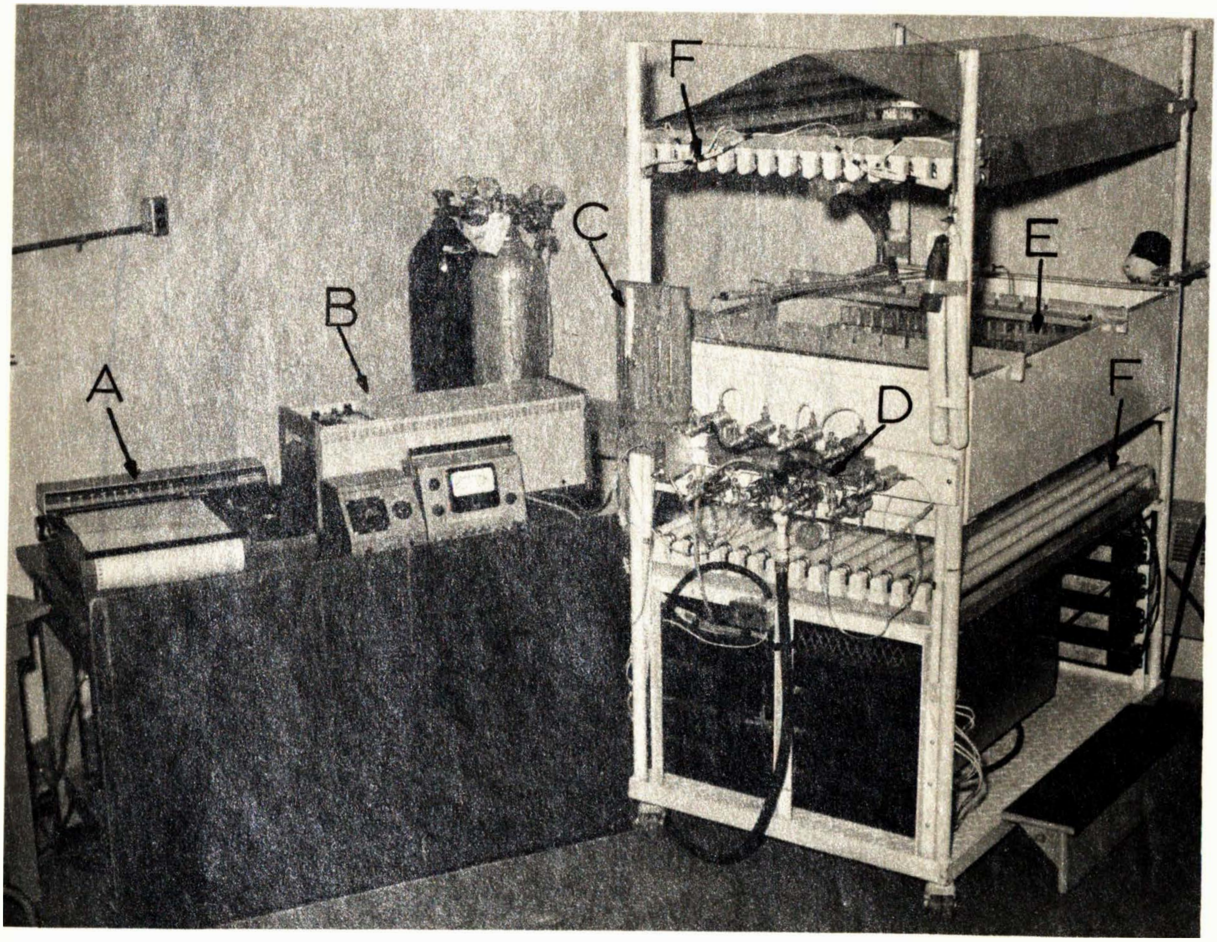

Figure 2. CO_2 measuring equipment as assembled in the laboratory.

A. Twelve-inch Beckman recorder.

B. Beckman 215 infra-red CO_2 analyzer.

C. Flow meter.

D. Electrical solenoids.

E. Water bath.

F. Light banks.

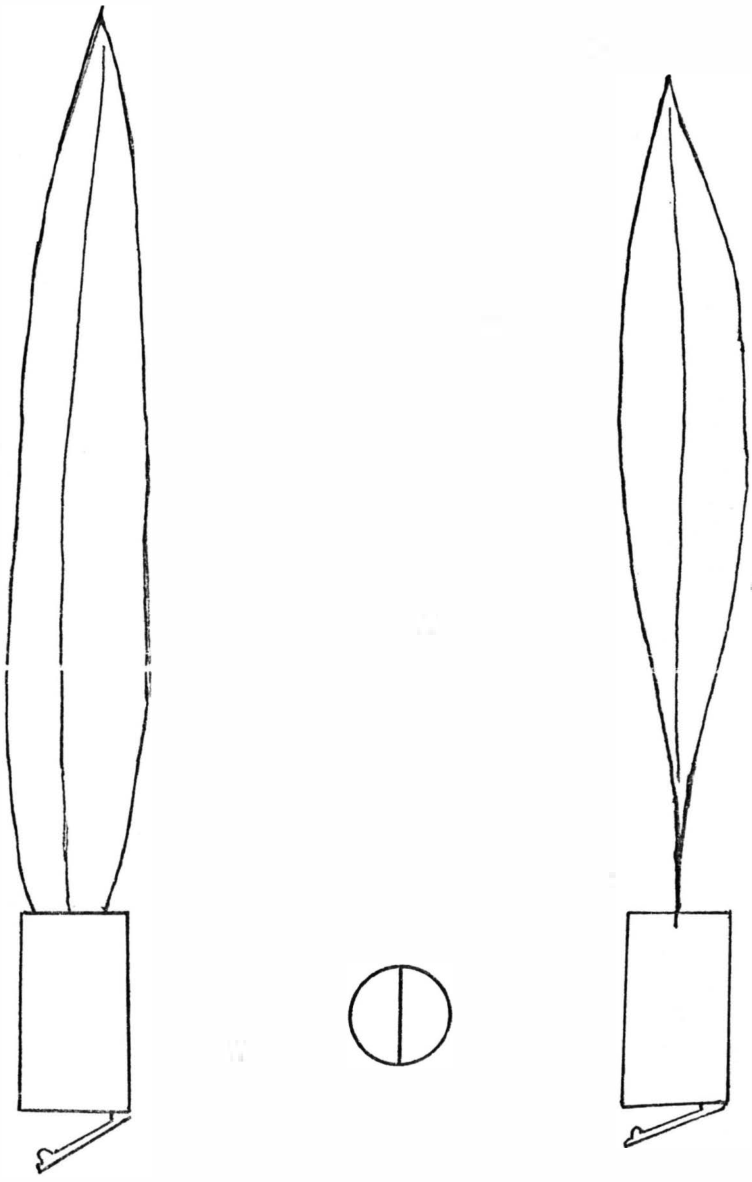

Figure 1. Excised leaves with cut ends placed in embedding capsules.

tubes by means of small plastic hoses. The air then passed through the glass sample tubes containing the intact leaf tissue. These sample tubes were connected to electric solenoids which either cycled the air to a Beckman 215 infra-Red CO_2 gas analyzer or to the atmosphere. The solenoids were controlled by an electric time clock which cycled the time of sampling. The analyzer registered the amount of CO_2 in the gas sample taken from each tube. The analyzer was coupled to an instrument which made a permanent record of the amounts of CO_2 in each sample analyzed. Calculations, expressed in μl CO_2 per gram dry weight per minute, were made from these data.

Effects of Atrazine on Net CO_2 Uptake

Initial Effect. This phase of the study was designed to determine the initial effect of atrazine and carriers on the sorghum plant. The plants were grown in a growth chamber with 1700 foot-candles of light, and a constant temperature of 76 degrees Fahrenheit and a sixteen-hour day length. The plants were sprayed when the third leaves were fully unrolled and the fourth leaf was still rolled. Immediately after spraying, the second and third leaves were excised for analysis of net CO_2 uptake. The foliage of the sorghum plants was sprayed with four treatments, as follows: (1) Control - no treatment; (2) Hundred second viscosity crop oil, one gallon per acre rate; (3) One pound per acre active ingredient atrazine plus

0.5 percent emulsifier in spray suspension; (4) One pound active ingredient atrazine per acre plus one gallon per acre crop oil in the spray suspension.

Due to the time required to sample and allow the tissue to come to equilibrium, the first valid determinations of net CO_2 uptake were made one and one-half hours following spraying. The leaves were periodically analyzed every 32 minutes from one and one-half to four and one-half hours after spraying. The data were converted to μ l CO_2 per gram of dry weight per minute. The μl CO_2 were then plotted against time to determine the initial effect of atrazine and its carriers on the net CO_2 uptake of the sorghum plant.

Persistence of Atrazine Effect. This phase of the study was designed to determine the initial and prolonged effects of atrazine plus oil on the net CO_2 uptake of the sorghum leaves. Leaves rolled and unrolled at the time of spraying were evaluated for the effect of atrazine plus oil on the net CO_2 uptake. The plants were grown in a growth chamber with 1700 foot-candles of light for sixteen hours per day at a constant temperature of 76 degrees Fahrenheit. The plants were sprayed when the third leaf was unrolled and the fourth leaf was exposed but not unrolled.

Immediately after spraying, the third leaves were excised from four plants and analyzed for the net CO_2 uptake. The third and fourth leaves were sampled and analyzed seven days following spraying.

Fourteen days following spraying, the third, fourth and fifth leaves were sampled and analyzed for net CO_2 uptake. The data were converted to μl CO_2 per gram dry weight per minute and was plotted against time.

<u>The Influence of Temperature and Light on Atrazine Effects.</u>

This phase of the study was designed to evaluate what effect various environmental factors had on the net CO_2 uptake rate of sorghum plants treated with atrazine plus oil. Two growth chambers with the same light intensity but with different temperatures were used. Different light intensity data were also desired, so one-half of the growth chamber was shaded with cheesecloth. The different temperature and light intensity factors evaluated were as follows: (1) A light intensity of 3800 foot-candles and a temperature of 80 degrees Fahrenheit; (2) A light intensity of 2100 foot-candles and a temperature of 80 degrees Fahrenheit; (3) A light intensity of 3800 foot-candles and a temperature of 68 degrees Fahrenheit; (4) A light intensity of 2100 foot-candles and a temperature of 68 degrees Fahrenheit.

All test chambers received sixteen hours of light per day and were maintained at a constant night and day temperature. The plants were sprayed when the third leaf of the sorghum plant was unrolled and the fourth leaf was still in the whorl. One pound of active atrazine plus one gallon crop oil per acre in twenty gallons per acre spray suspension was used, while the second group of plants were untreated.

Following the spraying, the third leaf was sampled and analyzed for CO_2 uptake immediately, and on the seventh, fourteenth and twenty-first day thereafter. These data were converted to μl CO_2 per gram dry weight per minute and were plotted against time.

II. FIELD STUDIES

General Field Procedure.

This portion of the study involved field plot evaluation of the effect of atrazine and its carriers on the sorghum plant. A series of three experiments were conducted, one in Brookings in 1968, one at the Southeast Research Farm in 1969, and one at Brookings in 1969. The experiments were set up in a randomized complete block design, and were replicated four times. The treatments were arranged in a factorial design using atrazine and oil as factors, as illustrated in tables one through eight. Rates are expressed in pounds per acre of active atrazine and gallons per acre of hundred second viscosity crop oil. Five-tenths of one percent emulsifier was used in combination with atrazine for one treatment. All treatments were applied using a tractor-type sprayer applying twenty gallons of spray suspension per acre when the third leaf was unrolled and the fourth leaf was still in the whorl. The experiments were maintained under weed-free conditions so that competition would not influence the maturity or yield of the grain sorghum.

Field Experiment A. This experiment was conducted at Brookings, South Dakota, during the 1968 growing season. NK-133 hybrid grain sorghum was planted on June 7, 1968. The plots were sprayed with the various treatments on July 25, 1968. The net CO_2 uptake rate was determined the day of spraying and three, six and twelve days after spraying. The sorghum plants were measured for height on July 16, 1968, and the percent head emergence in twenty feet of row was observed on August 2, 1968. Yield and moisture samples were recorded on October 8, 1968.

Field Experiment B. This experiment was conducted at Brookings, South Dakota, during the 1969 growing season. The NK-133 hybrid grain sorghum was planted on June 5, 1969. Resulting plant population was 31,500 plants per acre. The plots were sprayed with atrazine and its carriers on July 1, 1969, when the third leaf was unrolled and the fourth leaf was exposed but rolled up in the whorl. The weather before and after spraying was cool and cloudy. Net CO_2 uptake rate of the fourth leaf was measured the day of spraying and seven, fourteen and twenty-one days following spraying with atrazine and its carriers. The fifth leaf was also evaluated fourteen days after spraying. Height measurements were taken in each plot on July 15, July 22, and July 29, 1969.

Four plants from each plot were harvested for dry weight comparisons on July 10, July 20, and July 30, 1969. The number of heads emerged per twenty-five feet of row were counted on August 2, 1969. Grain and forage yields and percent moisture in the grain head samples were taken on October 9, 1969.

Field Experiment C. This experiment was conducted at the Southeast Research Farm near Beresford, South Dakota, during the 1969 growing season. The NK-133 hybrid grain sorghum was planted in thirty-inch rows on June 5, 1969. Resulting plant population was 105,000 plants per acre. The plots were sprayed on June 30, 1969, when the third leaf was unrolled and the fourth leaf was still rolled. The weather before and after treatment was cool and dry.

On July 14, 1969, the plant heights in each plot were measured. Number of heads emerged per twenty foot of row was counted on August 11, 1969. Grain and forage yields were taken on October 7, 1969, and percent moisture of the sorghum head was also determined on that date.

RESULTS

I. LABORATORY STUDIES

Effects of Atrazine on Net CO_2 Uptake.

Initial Effect. The second leaves were fully unrolled when sprayed and their response to the treatments was determined by measuring the CO_2 uptake (Figure 3). The atrazine plus oil had a more immediate effect in stopping or limiting the net CO_2 uptake of the second leaf than did the atrazine plus emulsifier treatment. By one and one-half hours after treatment the atrazine plus oil had reduced the net CO_2 uptake of the leaves to zero while the atrazine plus emulsifier had reduced the net CO_2 uptake by only about one-third that of the control leaves. By three and one-half hours after treatment, the atrazine plus emulsifier treatment had also reduced the net CO_2 uptake to zero.

The third leaf was fully unrolled when the plants were sprayed. In the evaluation of the third leaf, the atrazine plus oil treatment had a more immediate effect in stopping or limiting the net CO_2 uptake of the leaves than did the atrazine plus emulsifier treatment (Figure 4). By one and one-half hours after treatment, the atrazine plus oil treated leaves had a net CO_2 uptake of zero, while the atrazine plus emulsifier treated leaves were not as severely affected. By three and one-half hours after treatment, the atrazine plus oil treated leaves

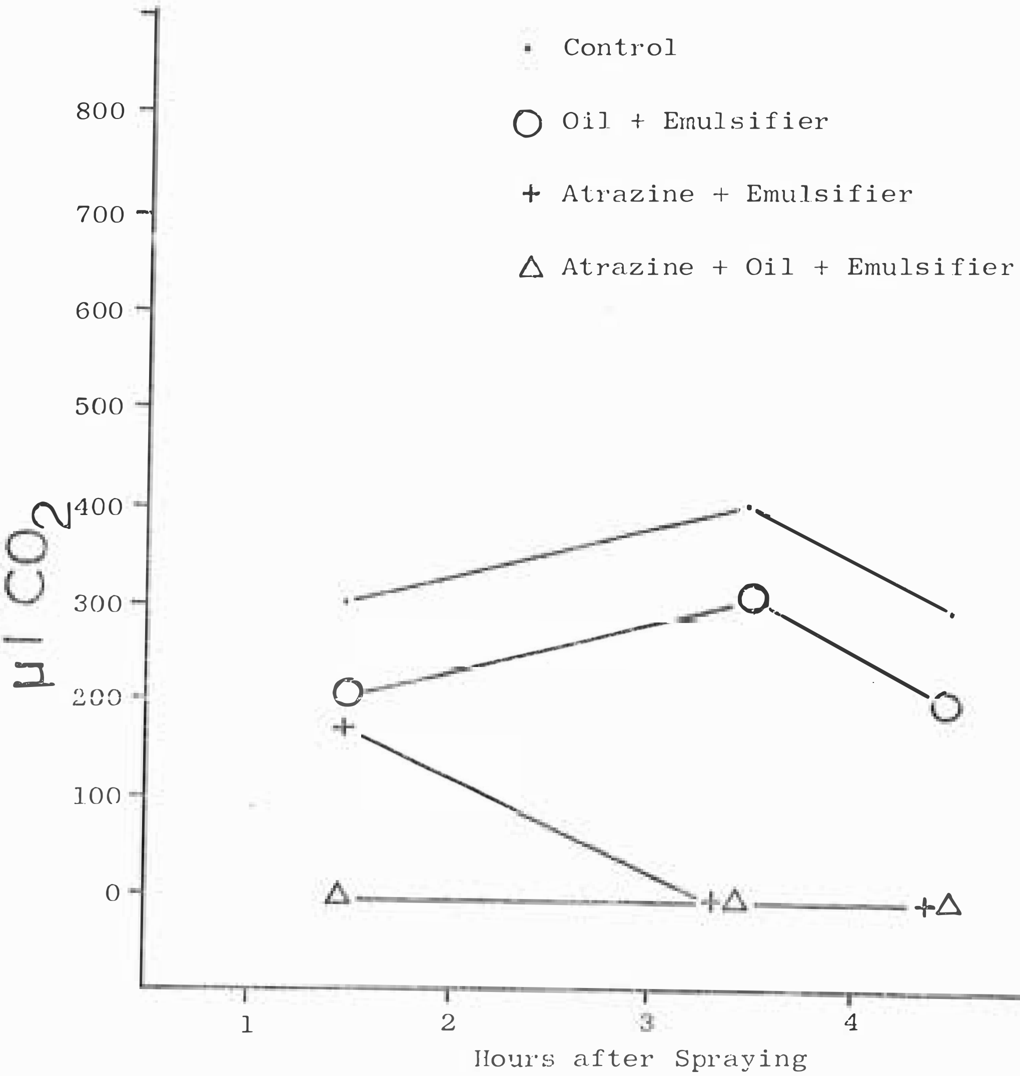

Figure 3. The initial effect of atrazine and its carriers on the net CO_2 uptake of the second leaf of the sorghum plant, as expressed in μl CO_2 per gram dry weight per minute.

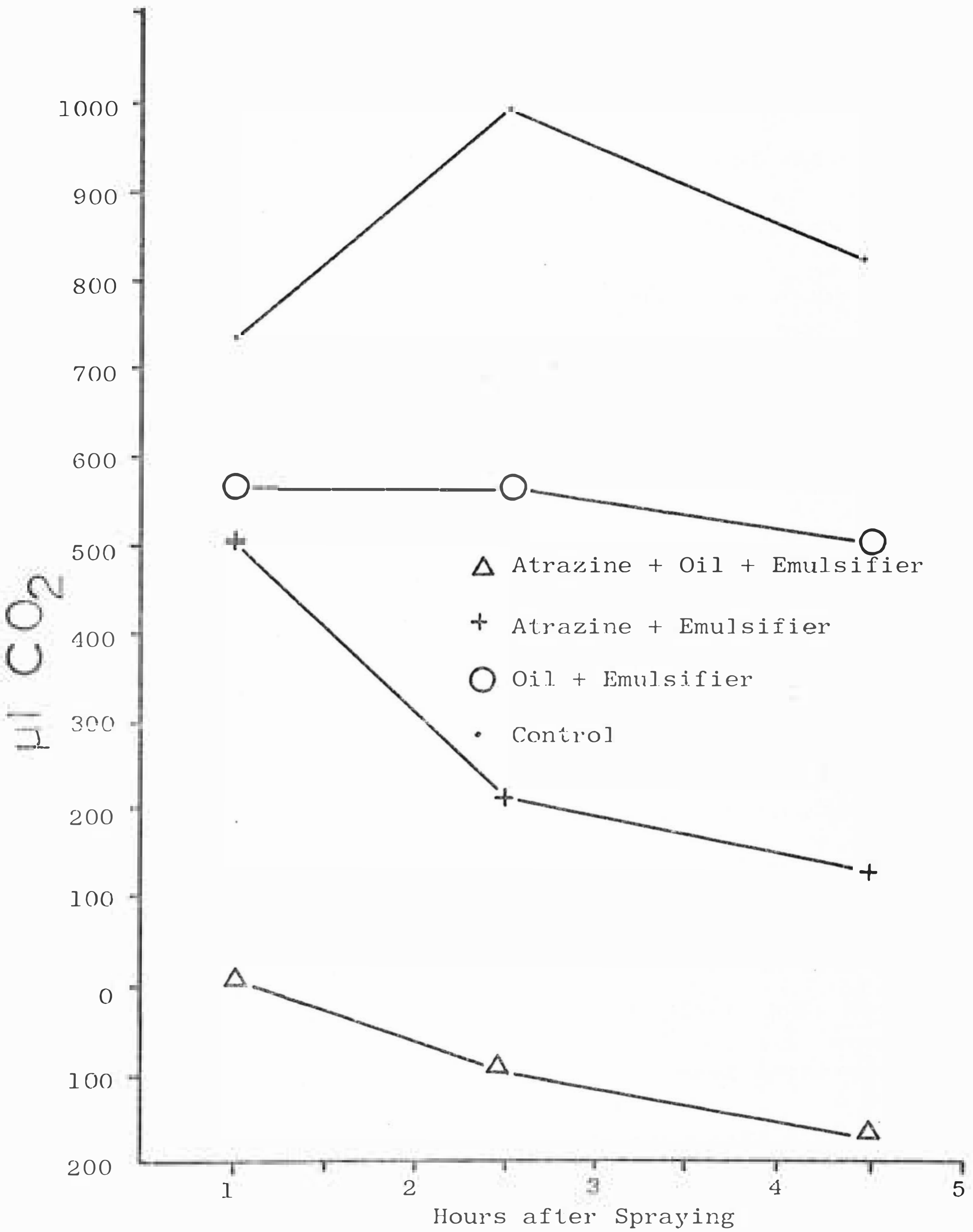

Figure 4. The initial effect of atrazine and carriers on the net CO_2 uptake of the third leaf of the sorghum plant, as expressed in $\mu l\, CO_2$ per gram dry weight per minute.

were liberating CO_2, indicating that respiration was taking place faster than was photosynthesis in these leaves.

Persistence of Atrazine Effect. The third leaf was unrolled when the plants were sprayed. The atrazine plus oil treatment reduced the level of net CO_2 uptake to zero the day of spraying (Figure 5). Seven days following spraying, the third leaf had recovered to a level of net CO_2 uptake slightly less than that of the control plants (Figure 5). The atrazine plus oil treated leaves continued to have a lower level of CO_2 uptake throughout the twenty-one days that the leaves were evaluated.

The fourth leaf was exposed but still rolled when the plants were sprayed and was not large enough to evaluate until seven days later. At this time, the net CO_2 uptake level for the atrazine plus oil treated leaves was below that of the control leaves, and remained at this suboptimum level throughout the experiment (Figure 6).

The fifth leaf was not exposed when the plants were sprayed and was not large enough to sample until fourteen days after spraying. At this time, the fifth leaf of the plants treated with atrazine plus oil had a net CO_2 uptake level approximately one-half that of the control plants (Figure 7). This indicates that even though the fifth leaf was not actually sprayed, it was affected by the atrazine which was sprayed on the exposed foliage of the plant.

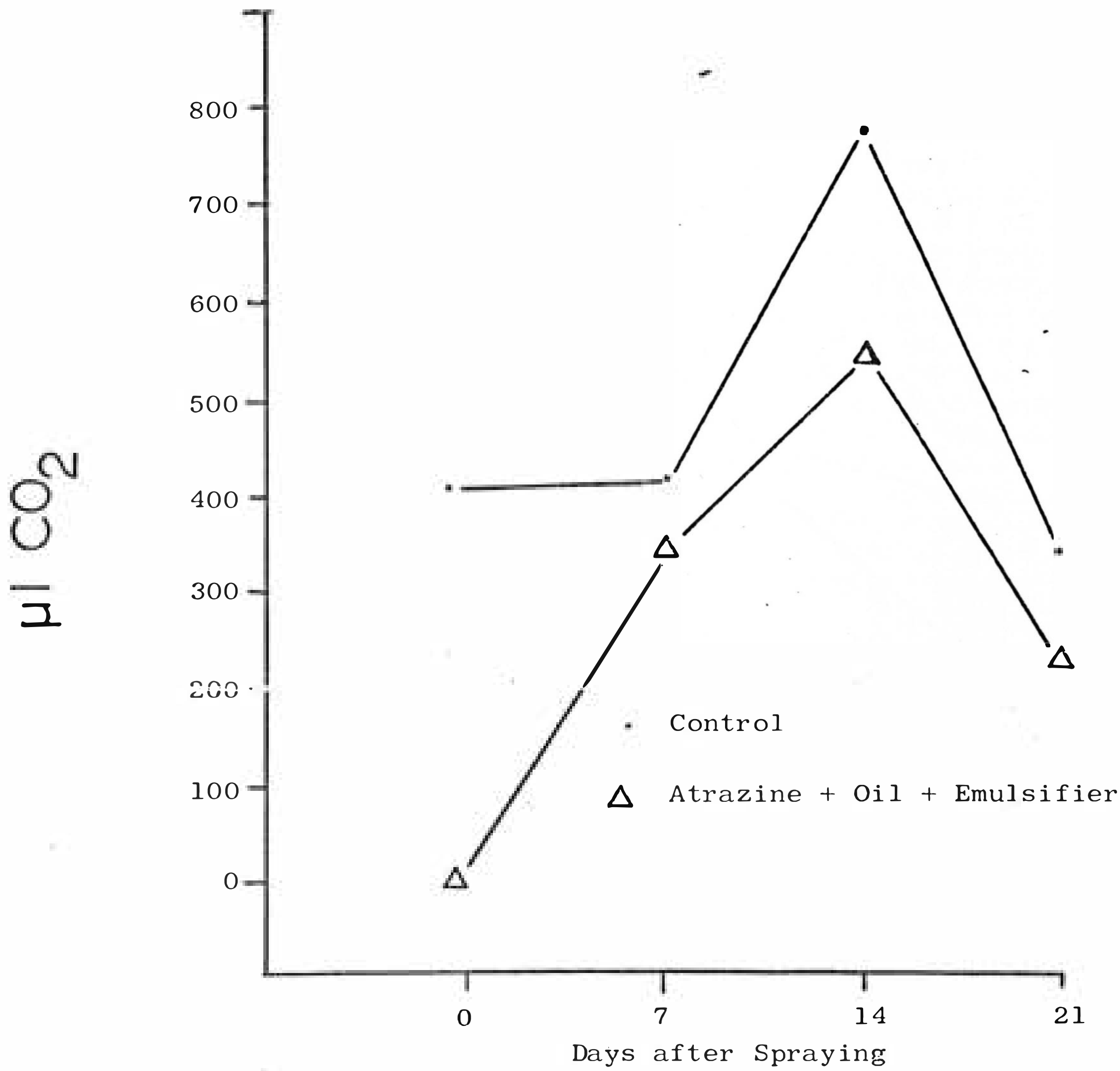

Figure 5. The net CO_2 uptake rate of the third leaf as affected by spraying with atrazine plus oil is expressed in µ l CO_2 per gram dry weight per minute.

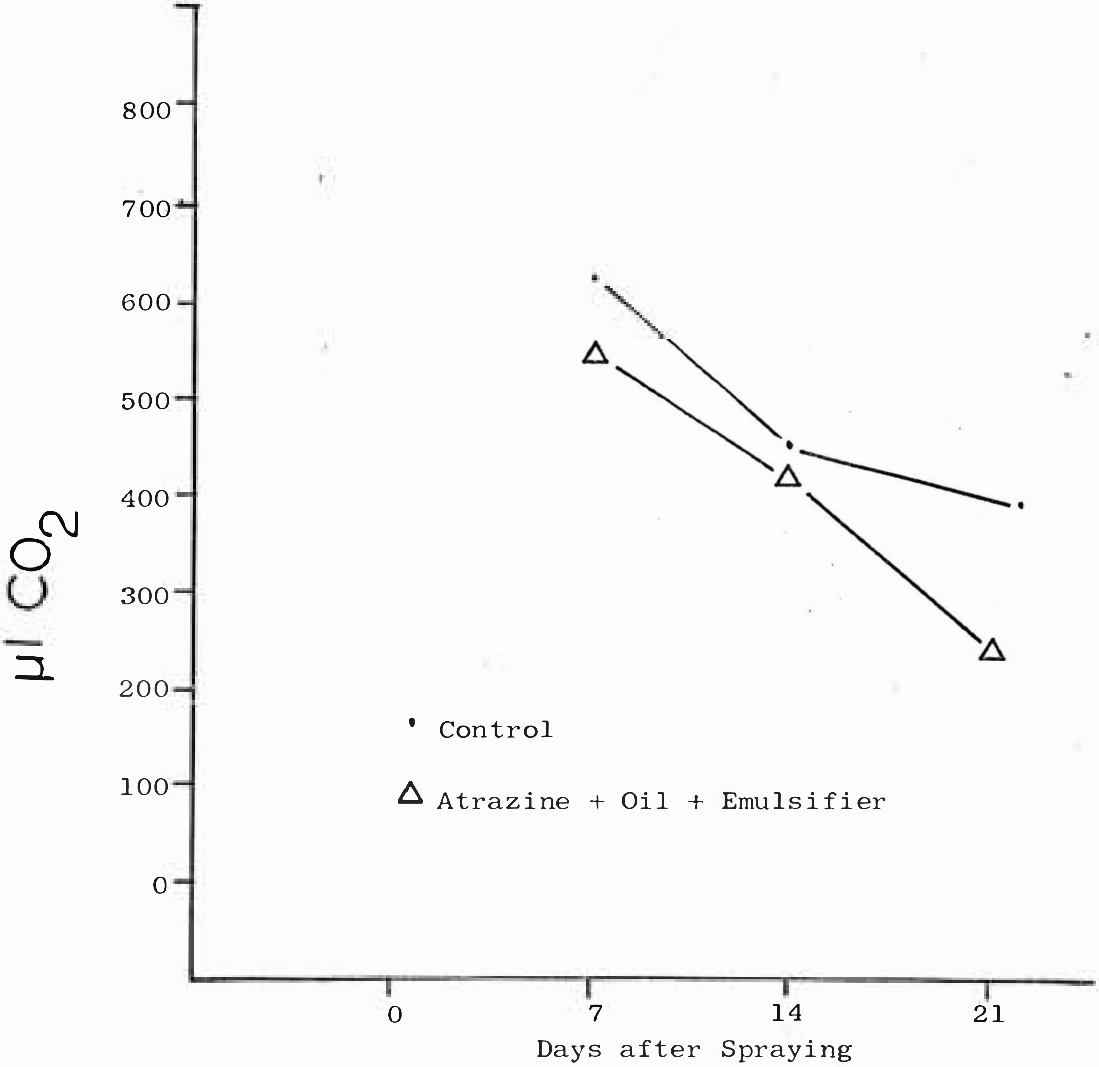

Figure 6. The net CO_2 uptake rate of the fourth leaf as affected by spraying with atrazine plus oil is expressed in μ l CO_2 per gram dry weight per minute.

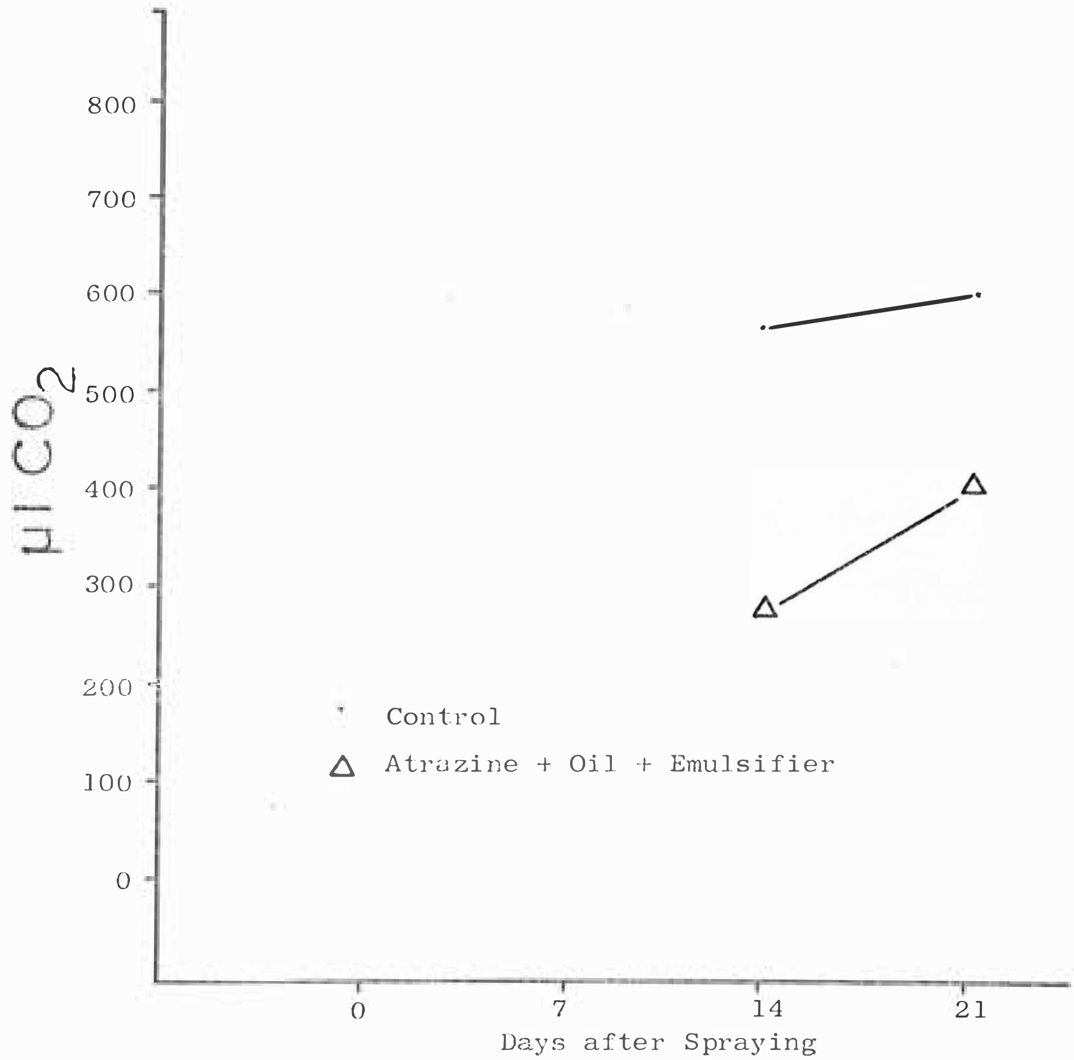

Figure 7. The recovery rate of net CO_2 uptake of the fifth leaf, after spraying with atrazine plus oil, as expressed in μ l CO_2 per gram dry weight per minute.

The Influence of Temperature and Light on Atrazine Effects.

80° Fahrenheit and 3,800 Foot-Candles Light Intensity. Under these growing conditions the atrazine plus oil treatment reduced the level of net CO_2 uptake of the treated leaves to less than one-fourth that of the untreated control plants (Figure 8). By seven days after spraying the treated leaves had recovered little in their rate of CO_2 uptake. At fourteen days after spraying the treated leaves had almost fully recovered in their level of CO_2 uptake.

68° Fahrenheit and 3,800 Foot-Candles Light Intensity. Initially the atrazine plus oil treatment reduced the level of CO_2 uptake of the treated leaves grown under these conditions so that the plants were liberating CO_2 (Figure 9). At seven days after spraying the level of net CO_2 uptake of the treated leaves was only about one-half that of the untreated control plants. The leaves treated with atrazine plus oil did not recover to a level of CO_2 uptake of more than one-half that of the untreated plants throughout the fourteen days the plants were analysed.

80° Fahrenheit and 2,100 Foot-Candles Light Intensity. Under these growing conditions the atrazine plus oil treatment reduced the level of net CO_2 uptake to a little more than one-fourth that of the untreated control plants (Figure 10). By seven days after treatment the treated leaves had recovered little in their level of net CO_2

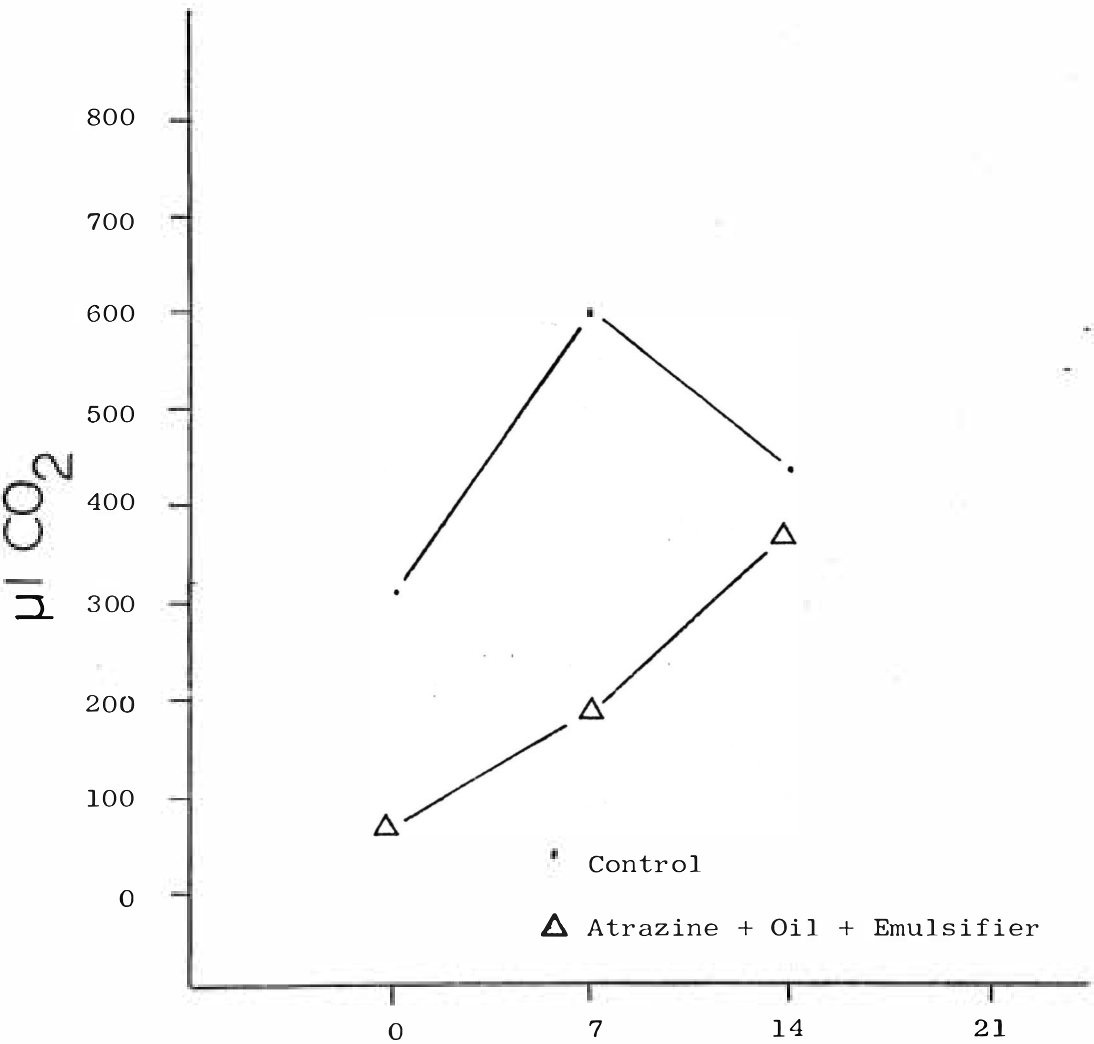

Figure 8. Net CO_2 uptake of the third leaf sprayed with atrazine plus oil under growing conditions of 80° Fahrenheit and 3,800 foot candles light intensity. The CO_2 uptake rates are expressed as μl CO_2 per gram dry weight per minute.

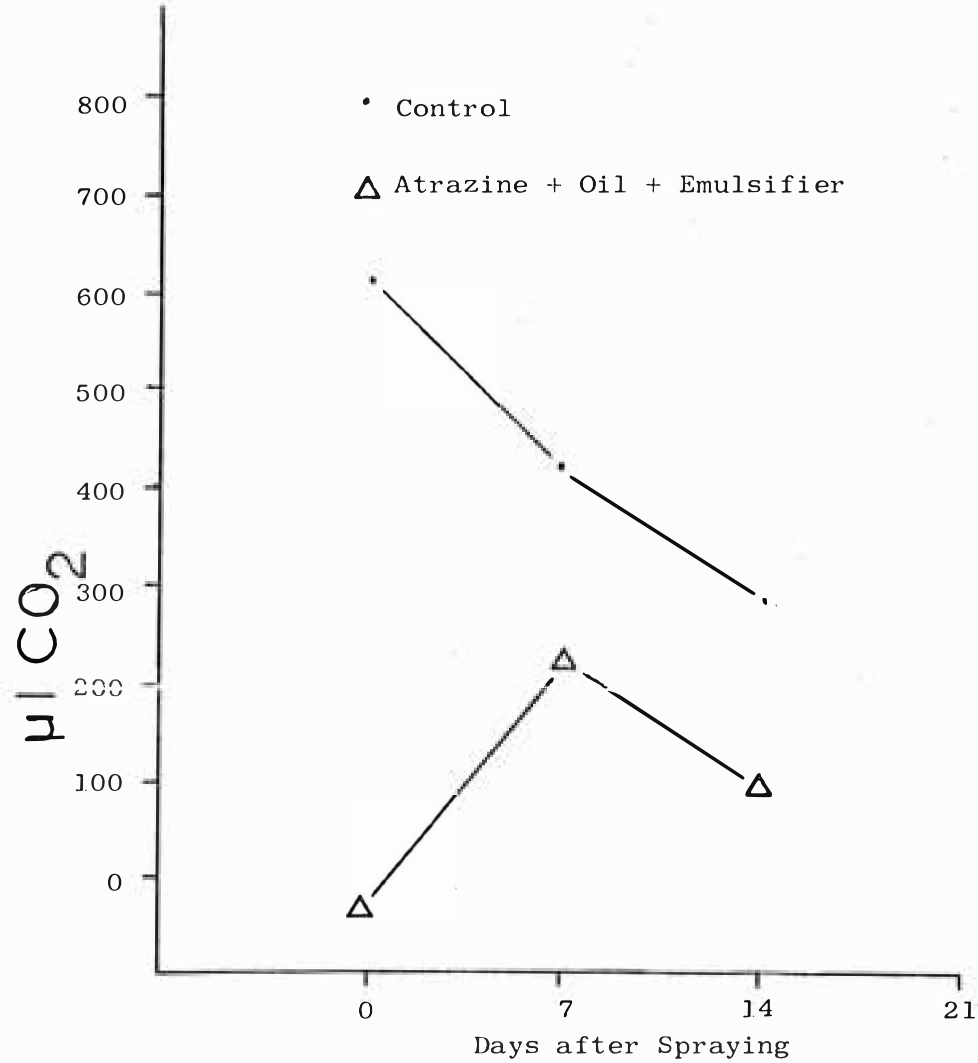

Figure 9. Net CO_2 uptake of the third leaf sprayed with atrazine plus oil under growing conditions of 68° Fahrenheit and 3,800 foot candles light intensity. The CO_2 uptake rates are expressed as μl CO_2 per gram dry weight per minute.

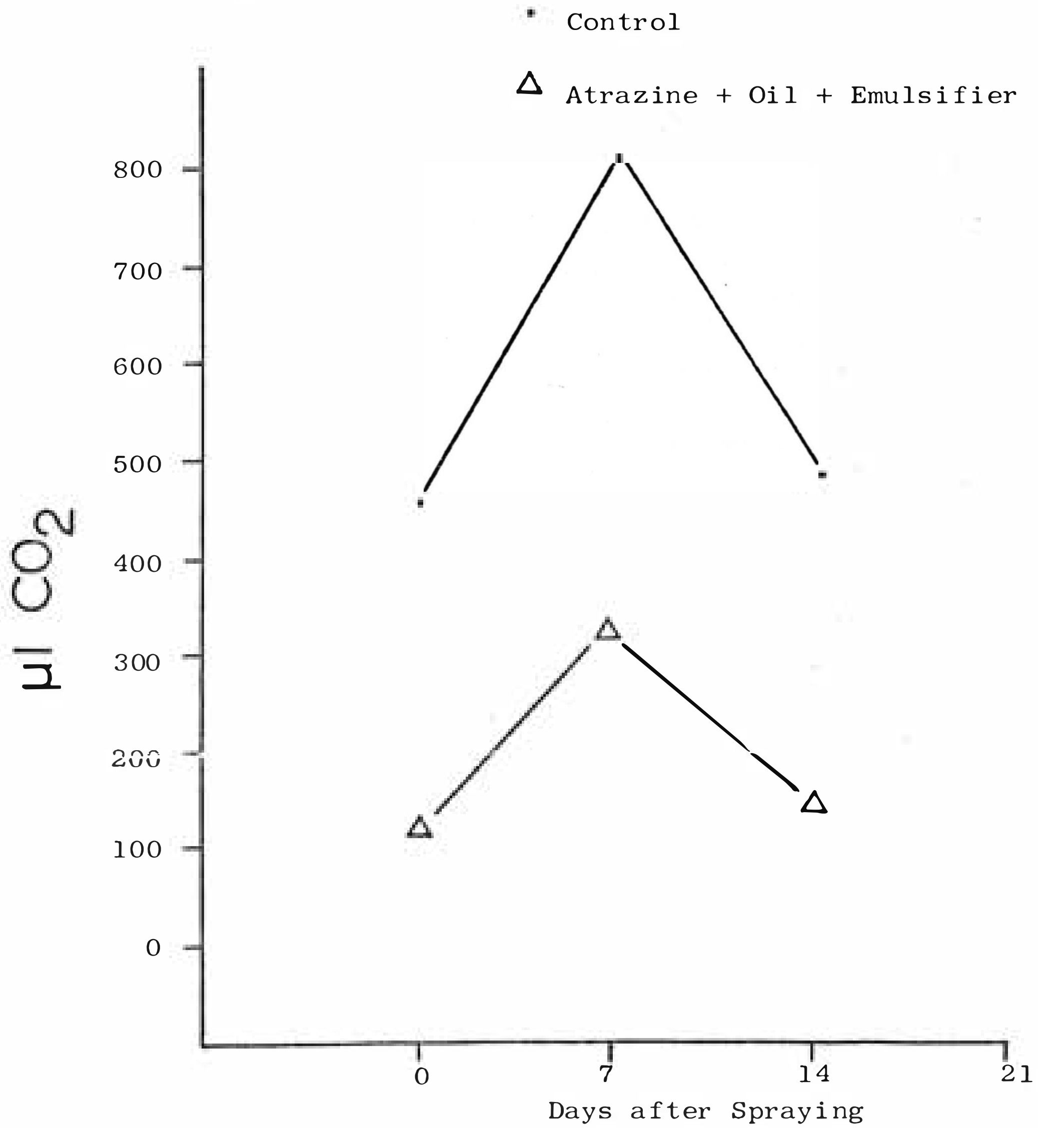

Figure 10. Net CO_2 uptake of the third leaf sprayed with atrazine plus oil under growing conditions of 80^o Fahrenheit and 2,100 foot candles light intensity. The CO_2 uptake rates are expressed as μ l CO_2 per gram dry weight per minute.

uptake. The atrazine plus oil treated leaves did not recover fully throughout the fourteen days they were analyzed.

<u>68° Fahrenheit and 2,100 Foot-Candles Light Intensity.</u> Initially the level of net CO_2 uptake of the atrazine plus oil treated leaves grown under these conditions was about one-fifth that of the untreated control plants (Figure 11). By seven days after spraying the treated leaves had recovered in their level of net CO_2 uptake to about one-half that of the untreated control leaves. At fourteen days after spraying the atrazine plus oil treated leaves had fully recovered in their level of net CO_2 uptake.

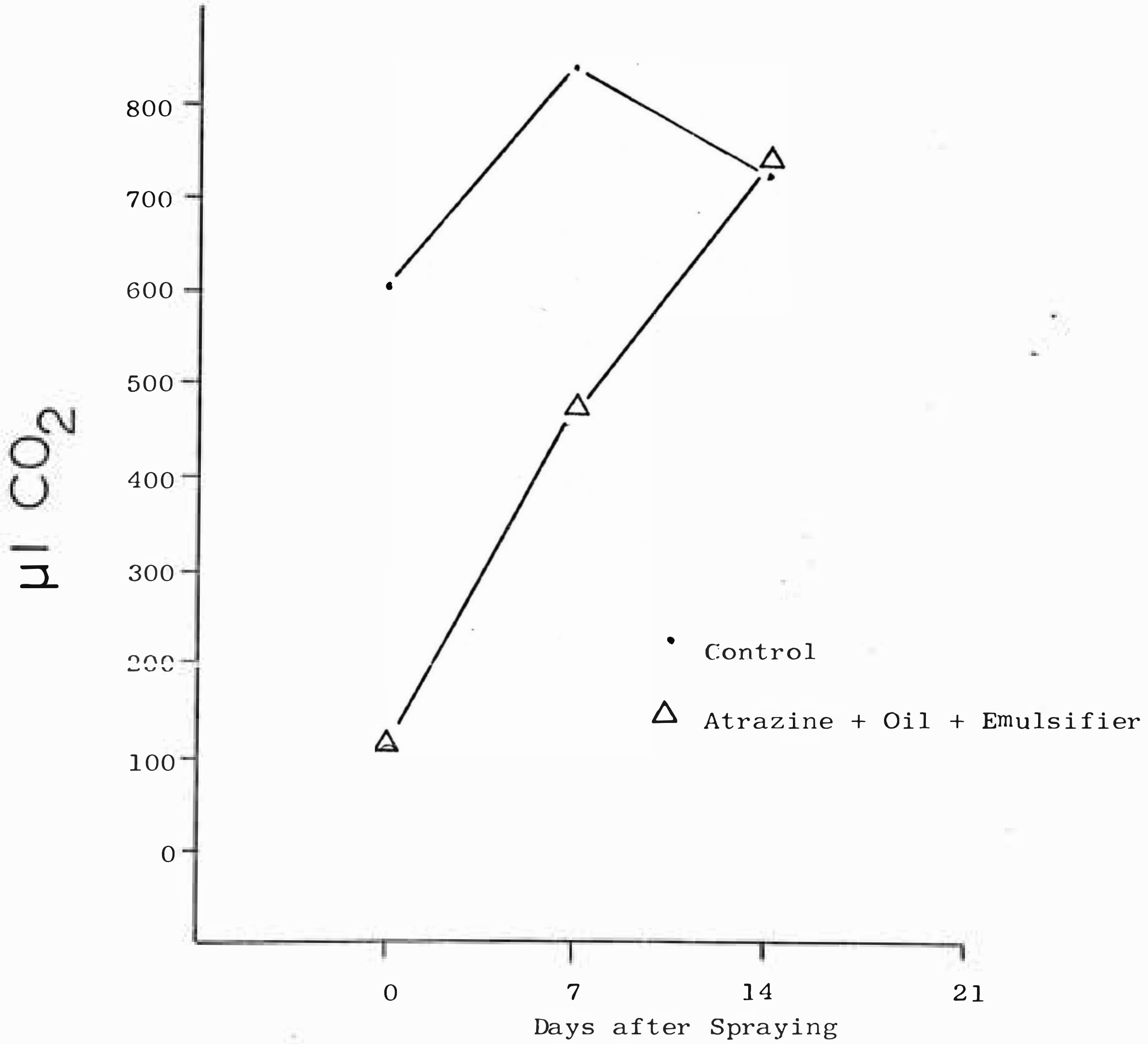

Figure 11. Net CO_2 uptake of the third leaf sprayed with atrazine plus oil under growing conditions of 68° Fahrenheit and 2,100 foot candles light intensity. The CO_2 uptake rates are expressed as μl CO_2 per gram dry weight per minute.

RESULTS

II. FIELD STUDIES

Experiment at Brookings, 1968.

Net CO_2 Uptake. Initially the atrazine plus oil treatment reduced the net CO_2 uptake level of the third leaf to twenty-five percent of that of the untreated control leaves (Figure 12). At both three and six days after spraying the atrazine plus oil treated leaves of the sorghum plant maintained a low level of net CO_2 uptake. By twelve days following spraying the atrazine plus oil treated leaves had recovered to one-third the rate of net CO_2 uptake of untreated control leaves. The atrazine plus emulsifier treatment initially reduced the net CO_2 uptake level severely, but by three days after treatment the leaves were recovering. At twelve days after spraying, the leaves treated with atrazine plus emulsifier had almost fully recovered in their level of net CO_2 uptake.

Height Measurements. No significant differences were noticeable among treatments at the ninty-five percent level of confidence twenty-one days after treatment (Table 1). Some trends were expressed in this table: The higher rates of atrazine plus oil reduced the height of sorghum plants grown under field conditions while the atrazine or the oil alone seemed to have very little effect on the sorghum plants' height.

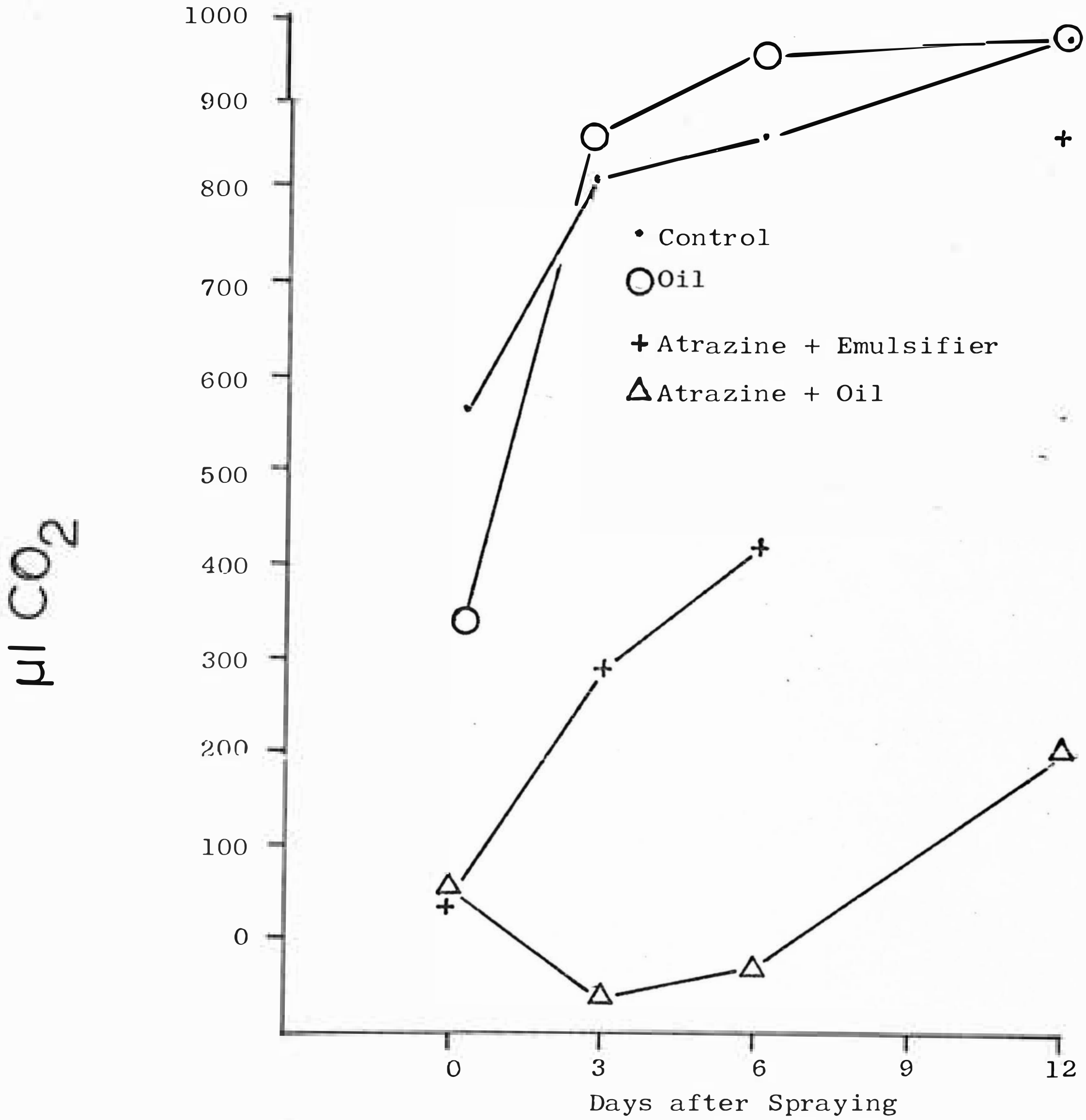

Figure 12. The effects of atrazine and its carriers on level of net CO_2 uptake of the third leaf of sorghum plants grown under field conditions at Brookings, South Dakota, during 1968. The data are determined in $\mu l\ CO_2$ per gram dry weight per minute.

Table 1. The effect of atrazine and its carriers on the growth of grain sorghum, at Brookings in 1968.

Treatment	Mean Height Cm. July 16	% Head Emergence August 2
No Oil		
Atrazine 0	58.8	34.0
Atrazine 1	58.0	32.0
Atrazine 2	56.5	26.0
One Gallon Oil		
Atrazine 0	58.3	39.0
Atrazine 1	55.5	17.5
Atrazine 2	55.5	15.5
Two Gallons Oil		
Atrazine 0	58.3	26.0
Atrazine 1	54.8	11.0
Atrazine 2	52.3	15.0
0.5% Emulsifier		
Atrazine 1	NA	32.0

NA data not available

* Mean is significantly different from the control mean according to Dunnett's test at the 95% level of confidence.

** Mean is significantly different from the control mean according to Dunnett's test at the 99% level of confidence.

Percent Head Emergence. The means shown in Table 1 indicated some trends with the higher rates of atrazine plus oil reducing the percent head emergence at the time the notes were taken. The rate of head emergence for the one pound of atrazine alone was the same as the rate of head emergence with application of one pound of atrazine plus an emulsifier.

Percent Moisture in the Sorghum Head. The percent of moisture of the sorghum heads with these treatments ranged from twenty-six to thirty-five percent at the time of harvest on October 8, 1968 (Table 2).

Grain Yields. No significant differences in grain yields were apparent among the treatments at the ninty-five percent level of confidence (Table 2). The means indicated a trend that the higher rates of atrazine plus oil reduced the yields, while the treatments of atrazine or oil alone did not.

Experiment B, Southeast Research Farm, 1969.

Height Measurements. The treatments of one pound of atrazine plus one gallon oil, and of two pounds atrazine plus one gallon oil, and two pounds atrazine plus two gallons oil significantly reduced the sorghum plants' height (Table 3). The means also show a trend that the higher rates of atrazine plus oil tend to reduce the height of

Table 2. The effects of atrazine and its carriers on the growth of grain sorghum at Brookings in 1968.

Treatments	Mean Yield Lbs./A.	Mean % Moisture of Head
No Oil		
Atrazine 0	4014	35.3
Atrazine 1	4002	28.3
Atrazine 2	4027	29.8
One Gallon Oil		
Atrazine 0	4081	28.3
Atrazine 1	3684	31.5
Atrazine 2	3659	29.0
0.5% Emulsifier		
Atrazine 1	3731	26.8

* Mean is significantly different from the control mean according to Dunnett's test at the 95% level of confidence.

** Mean is significantly different from the control mean according to Dunnett's test at the 99% level of confidence.

Table 3. The effects of atrazine and its carriers on grain sorghum at the Southeast Research Farm in 1969.

Treatment	Mean Height in Ft.	Mean No. Heads Emerged
No Oil		
Atrazine 0	1.4	42.8
Atrazine 1	1.4	36.0
Atrazine 2	1.3	31.0
One Gallon Oil		
Atrazine 0	1.5	31.3
Atrazine 1	1.3	55.8 **
Atrazine 2	1.0**	5.3 **
Two Gallons Oil		
Atrazine 0	1.4	39.8
Atrazine 1	1.1 *	3.3 **
Atrazine 2	1.1 *	1.5 **
0.5% Emulsifier		
Atrazine 1	1.6	38.5

* Mean is significantly different from the control mean according to Dunnett's test at the 95% level of confidence.

** Mean is significantly different from the control mean according to Dunnett's test at the 99% level of confidence.

the sorghum, while the atrazine or the oil alone affected sorghum height very little.

Number of Heads Emerged Per Twenty-Five Feet of Row. The number of heads emerged was significantly reduced by the following treatments: One pound atrazine plus one gallon oil; one pound atrazine plus two gallons of oil; two pounds atrazine plus one gallon oil; and, two pounds atrazine plus two gallons oil (Table 3). Analysis under a factorial arrangement indicated that atrazine treatments were significantly different at the ninty-five percent level of confidence and that oil treatments were not significantly different from the control rows.

Percent Moisture in Head of Sorghum Plant. The percent moisture in the head at harvest time was generally higher when atrazine plus oil treatments were applied to the plants than they were in the untreated control plants (Table 4).

Forage Yields. No significant differences among treatments were apparent in forage yields (Table 4). The average forage yields from the atrazine plus oil treated plots tended to be slightly higher than those that were untreated.

Grain Yields. There were no significant differences in grain yields. The average grain yields tended to be lower in atrazine plus oil treated plots than yields in the untreated plots (Table 4).

Table 4. The effects of atrazine and its carriers on grain sorghum at the Southeast Research Farm in 1969.

Treatments	Mean Forage Yield Lbs./A.	Mean Grain Yield Lbs./A.	% Moisture in Head at Harvest
No Oil			
Atrazine 0	11,770	6,560	29
Atrazine 1	11,990	6,440	29
Atrazine 2	11,820	6,350	29
One Gallon Oil			
Atrazine 0	11,410	6,600	30
Atrazine 1	11,850	6,350	32
Atrazine 2	11,920	6,250	32
Two Gallons Oil			
Atrazine 0	11,490	6,500	30
Atrazine 1	11,680	6,080	31
Atrazine 2	12,140	6,280	33
0.5% Emulsifier			
Atrazine 1	11,660	6,540	29

* Mean is significantly different from the control mean according to Dunnett's test at the 95% level of confidence.

** Mean is significantly different from the control mean according to Dunnett's test at the 99% level of confidence.

Experiment C, Brookings, 1969.

Net CO_2 Uptake. The third leaf was unrolled when the plants were sprayed. Initially the atrazine plus oil treatment reduced the net CO_2 uptake level of the treated leaves to below zero and the third leaf was liberating CO_2 (Figure 13). The atrazine plus emulsifier reduced the level of net CO_2 uptake to zero the day of spraying. Both atrazine treatments had recovered substantially by seven days following spraying and by twenty-one days after spraying the atrazine treated leaves appeared to be fully recovered in net CO_2 uptake rates.

The fifth leaf was not emerged when the plants were sprayed and the leaves were not large enough to evaluate until fourteen days following spraying. The net CO_2 uptake rates of the four treatments showed little difference at that time (Figure 14). The analysis was conducted until twenty-one days after spraying and at that time there appeared to be little difference in rate of net CO_2 uptake of the four applied treatments.

Height Measurements. The height of the sorghum plants in each plot was measured in centimeters on July 15, 1969, with the data expressed in Table 5. The results indicated that one or two pounds of atrazine, when used in combination with one or two gallons of oil, re- reduced the height of the sorghum plants significantly at the ninety-nine percent level of confidence.

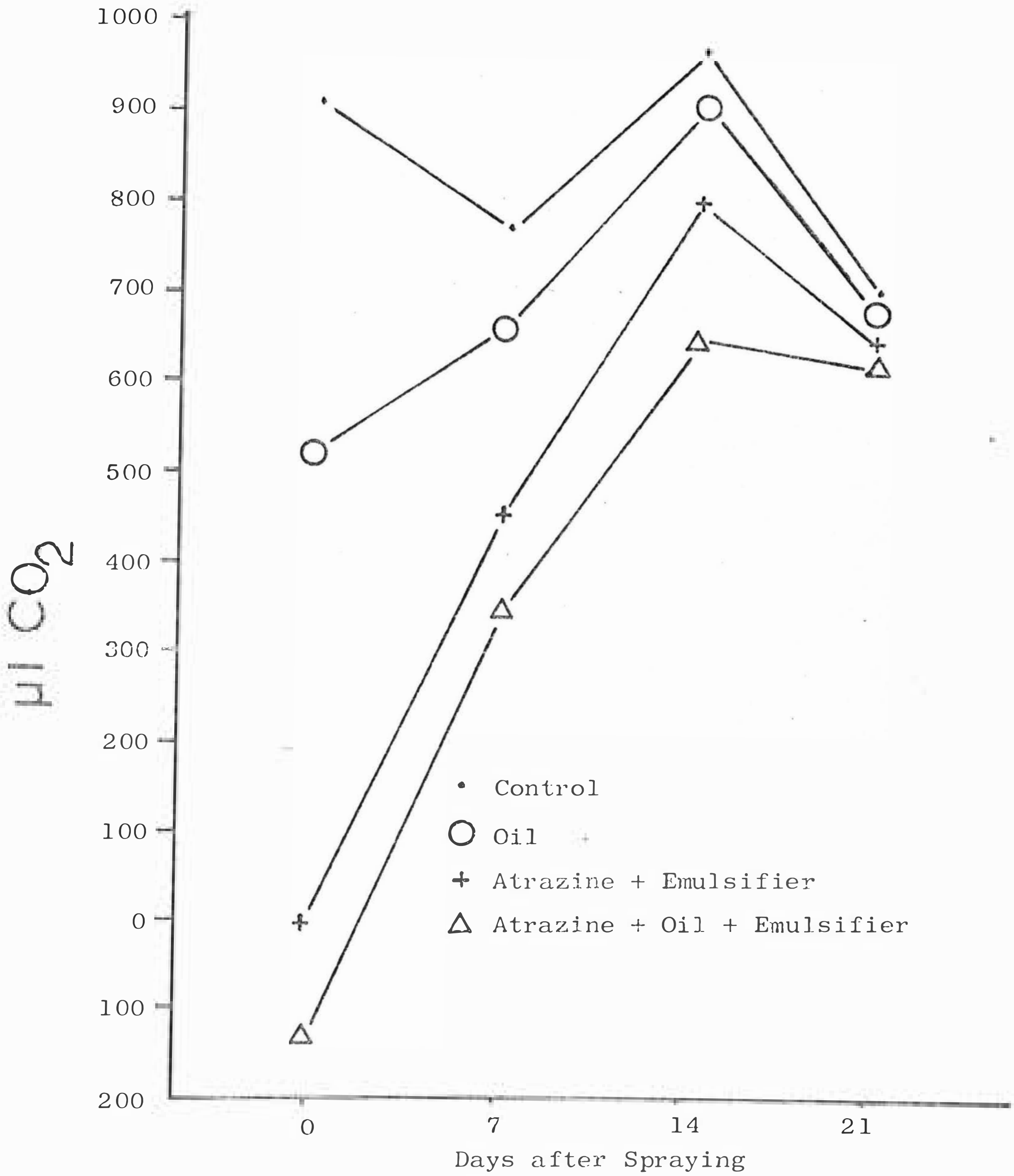

Figure 13. Net CO_2 uptake per gram dry weight per minute of the third leaf grown under field conditions at Brookings, South Dakota, 1969, as affected by atrazine and its carriers.

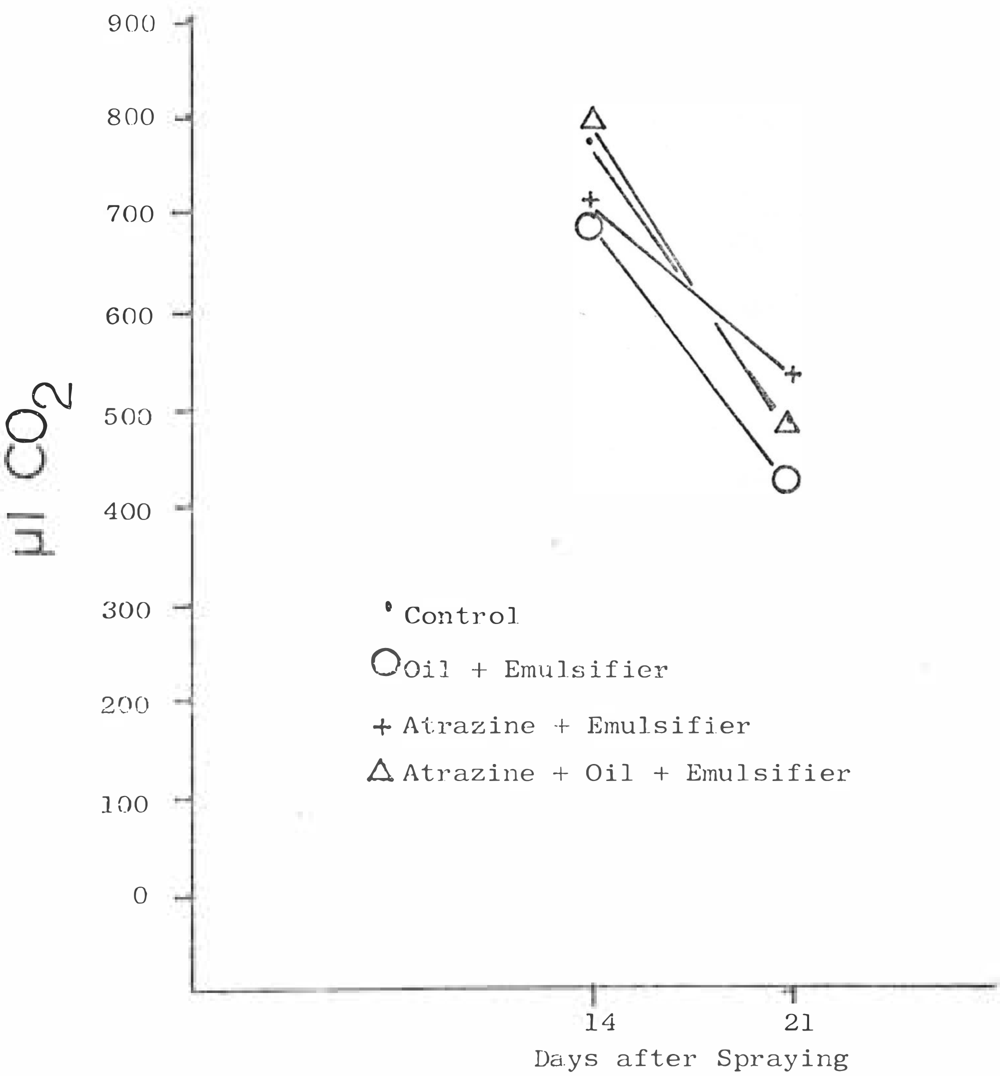

Figure 14. Net CO_2 uptake per gram dry weight per minute of the fifth leaf grown under field conditions at Brookings, South Dakota, 1969, as affected by atrazine and its carriers.

Table 5. The effects of atrazine and its carriers on the growth of grain sorghum at Brookings during the 1969 growing season.

	Height in Cm.		
Treatments	July 15 Mean	July 22 Mean	July 29 Mean
No Oil			
Atrazine 0	14.0	22.8	27.3
Atrazine 1	13.8	20.5	25.5
Atrazine 2	12.8	19.5	25.5
One Gallon Oil			
Atrazine 0	15.5	22.3	24.8
Atrazine 1	10.0 **	17.8**	23.3
Atrazine 2	8.8 **	16.0**	22.5*
Two Gallons Oil			
Atrazine 0	15.0	20.0	26.8
Atrazine 1	10.0**	15.8**	23.0*
Atrazine 2	8.5**	16.3**	22.8*
0.5% Emulsifier			
Atrazine 1	13.3	21.5	25.8

* Mean is significantly different from the control mean according to Dunnett's test at the 95% level of confidence.

** Mean is significantly different from the control mean according to Dunnett's test at the 99% level of confidence.

Plant height in each plot was again measured on July 22, 1969. It was noted that plants sprayed with atrazine plus oil treatments were significantly shorter than the control plants at the ninety-nine percent level of confidence (Table 5). The atrazine rates alone and the atrazine plus emulsifier treatments did not significantly influence the height. Analyzing the factorially arranged portion of the experiment revealed that the rates of atrazine caused a significant reduction in the sorghum plants' height at the ninety-nine percent level of confidence. The rates of oil significantly reduced height at the ninety-five percent level of confidence.

The height of sorghum plants was measured for the third time at twenty-nine days after treatment. At that time, the following treatments significantly reduced the height: two pounds of atrazine plus one gallon of oil, one pound of atrazine plus two gallons of oil, and two pounds of atrazine plus two gallons of oil (Table 5). Under factorial analysis, the atrazine rates significantly reduced the height of the sorghum plants, compared to the control plants' height. The height of the plants treated with oil was not significantly different from the height of the control plants at the ninety-five percent level of confidence.

Dry Weight Yield of Sorghum Plants.

July 10. Dry weight samples were taken ten days following spraying. At that time, all treatments except the one pound atrazine alone and the one gallon of oil alone were significantly lighter than were the control plants (Table 6). Under factorial analysis the atrazine and the oil treated plants were significantly lighter than the control plants at the ninety-five percent level of confidence.

July 20. All the treatments significantly reduced the dry weight of the sorghum plants except the one pound of atrazine alone, the one gallon of oil alone and the one pound of atrazine plus 0.5 percent emulsifier (Table 6). The higher rates of atrazine and atrazine in combination with oil reduced the dry weight of the sorghum plants significantly at the ninety-nine percent level of confidence.

July 30. The one pound and two pound rates of atrazine and atrazine in combination with oil reduced the sorghum plants' dry weight significantly (Table 6). The one and two gallons of oil and the one pound rate of atrazine in combination with 0.5 percent emulsifier did not significantly reduce the dry weight of the sorghum plant. When the data were analyzed under a factorial arrangement, it was found that the atrazine treatments were not significantly different from control plants. There was no interaction between the atrazine and the oil treatments.

Number of Heads Emerged per Twenty-Five Feet of Row. There was no significant difference among treatments (Table 7). The table of means indicated a trend showing the higher rates of atrazine plus oil reduced the number of heads emerged, while the atrazine alone or

Table 6. The effect of atrazine and its carriers on the growth of the grain sorghum plant at Brookings during the 1969 growing season.

Treatment	Dry Weight in Grams		
	July 10 Mean	July 20 Mean	July 30 Mean
No Oil			
Atrazine 0	2.9	39.0	75.3
Atrazine 1	2.6	38.2	66.3**
Atrazine 2	1.9*	41.8**	51.9**
One Gallon Oil			
Atrazine 0	2.2	37.4	73.2
Atrazine 1	1.2**	23.4**	50.4**
Atrazine 2	0.9**	26.1**	42.2**
Two Gallons Oil			
Atrazine 0	1.8**	30.1**	78.0
Atrazine 1	1.4**	24.5**	45.2**
Atrazine 2	0.9**	21.8**	43.2**
0.5% Emulsifier			
Atrazine 1	1.8**	35.1	81.5

* Mean is significantly different from the control mean according to Dunnett's test at the 95% level of confidence.

** Mean is significantly different from the control mean according to Dunnett's test at the 99% level of confidence.

Table 7. The effect of atrazine and its carriers on the maturity of grain sorghum grown at Brookings during the 1969 growing season.

Treatments	No. Heads Emerged Mean August 11	% Moisture of Head Mean October 7
No Oil		
Atrazine 0	15.5	43
Atrazine 1	26.0	41
Atrazine 2	25.0	41
One Gallon Oil		
Atrazine 0	21.3	39
Atrazine 1	5.5	43
Atrazine 2	6.0	45
Two Gallons Oil		
Atrazine 0	17.5	41
Atrazine 1	6.0	45
Atrazine 2	2.8	44
0.5% Emulsifier		
Atrazine 1	22.0	42

* Mean is significantly different from the control mean according to Dunnett's test at the 95% level of confidence.

** Mean is significantly different from the control mean according to Dunnett's test at the 99% level of confidence.

the atrazine plus 0.5 percent emulsifier did not. Under a factorial arrangement there was no significant difference among atrazine or oil treatments and there was no interaction.

Percent Moisture of the Sorghum Head. The percent moisture was determined at harvest time. The treatments containing atrazine plus oil generally had a higher percentage of moisture in the head than the untreated plants (Table 7).

Grain Yields. No significant differences appeared in grain yields among treatments. The average yields from plots treated with atrazine plus oil tended to be slightly lower than were the yields of the untreated plots (Table 8).

Forage Yields. The forage yields of treated plants were not significantly different from the untreated plants at the ninety-five percent level of confidence. The plots treated with higher rates of atrazine plus oil tended to produce a lower average forage yield (Table 8).

Table 8. The effect of atrazine and its carriers on the yield of grain sorghum grown at Brookings during the 1969 growing season.

Treatment	Forage Yield Lbs./A. Mean	Grain Yield Lbs./A. Mean
No Oil		
Atrazine 0	7,990	4,070
Atrazine 1	7,950	3,980
Atrazine 2	8,040	3,730
One Gallon Oil		
Atrazine 0	8,260	4,110
Atrazine 1	8,030	3,860
Atrazine 2	7,740	3,860
Two Gallons Oil		
Atrazine 0	8,270	4,380
Atrazine 1	7,750	3,580
Atrazine 2	7,670	3,540
0.5% Emulsifier		
Atrazine 1	7,860	3,820

* Mean is significantly different from the control mean according to Dunnett's test at the 95% level of confidence.

** Mean is significantly different from the control mean according to Dunnett's test at the 99% level of confidence.

DISCUSSION OF RESULTS

I. LABORATORY RESULTS

Initially, the one pound atrazine active ingredient plus one gallon of oil per acre reduced the level of net CO_2 uptake on leaves more than did the one pound atrazine active ingredient plus 0.5 percent emulsifier in spray suspension. This would corroborate the work performed by Smith (38). He found that more atrazine was taken up by the leaf when comparing C^{14} atrazine plus oil and C^{14} atrazine in water suspensions.

The leaves sprayed with these sub-lethal doses of one pound active ingredient of atrazine recovered slowly in their ability to assimilate CO_2. It was apparent that leaves treated with atrazine plus oil did not recover as quickly as those treated with atrazine plus an emulsifier. The third leaf of the sorghum plant had a higher level of net CO_2 uptake and was able to recover from atrazine treatments more quickly than was the second leaf, which had a lower net CO_2 uptake level. This is in agreement with the theory proposed by Moss (28) that plants with a higher photosynthetic rate recover from atrazine treatments faster than do plants with lower photosynthetic rates.

The fourth and fifth leaves, which were not emerged at the time of spraying, were evaluated for net CO_2 uptake. It was found that these leaves had a lower level of net CO_2 uptake than did the untreated control plants. This would indicate that the atrazine, when used in combination with oil in a water suspension, was capable of penetrating the leaf sheath and reaching those leaves not yet unrolled.

Effects of atrazine plus oil on plants grown under different light and temperature conditions were evaluated. The atrazine plus oil treatment reduced the CO_2 uptake level of the sorghum plant's third leaf grown under all temperature and light conditions. There appeared to be some interaction between light intensity and temperature. The high light intensity and low temperature resulted in severe initial injury. This same group of plants did not recover as rapidly as did plants treated under different growing conditions. The plants grown under low light intensity and low temperatures did not show severe injury initially and appeared fully recovered fourteen days following spraying, while the plants grown under different growing conditions did not. Results indicate that symptoms caused by atrazine are more apparent under 3,800 foot candles light intensity than under 2,100 foot candles of light intensity.

DISCUSSION OF RESULTS

II. FIELD RESULTS

The field results collected on net CO_2 uptake level of the leaves treated with atrazine and its carriers agreed with laboratory results. The atrazine plus oil suspension reduced the level of net CO_2 uptake more than did the atrazine plus emulsifier suspension. The plant leaves in the 1968 experiment were slower to recover to the normal net CO_2 uptake rate than were the leaves treated with atrazine plus oil in the 1969 experiment (Figures 10 and 11). The 1968 experiment was sprayed during a period when the daily mean temperature was well below average and remained below normal for the next two days (Appendix I). The 1969 experiment was sprayed when the mean daily temperature was near normal (Appendix I). These data indicate that plants sprayed under normal field light intensity with a low mean temperature exhibit a greater reduction in net CO_2 uptake than plants sprayed with atrazine plus oil under warmer temperatures.

The sorghum plants were generally stunted when treated with atrazine plus oil. All measurements taken on the height of the sorghum plant indicated that the higher rates of atrazine plus oil caused height reductions early in the growing season (Tables 1, 3, and 5). Dry weight determinations per plant were made in the 1969 Brookings experiment and these data also indicated some stunting (Table 6). During the growing season, the plants did partially recover from this stunting.

At the time the sorghum heads were emerging from the boot stage, there appeared to be a maturation delay in the plants treated with atrazine plus oil in all experiments conducted (Tables 3 and 7); however, by harvest time very little difference was indicated in the percent moisture of the sorghum head.

Atrazine plus oil treatments slightly lowered grain yields in all experiments (Tables 2, 4, and 8). These results may have been because of the early reduction in net CO_2 uptake and the uptake of the atrazine throughout the season by the roots of the sorghum plants that were grown in the atrazine-treated plots.

During the 1969 season, forage yields were taken at both locations. Plots treated with atrazine plus oil at the Southeast Research Farm had higher forage yields but lower grain yields than other plots in the experiment. The plant population in this experiment was 105,000 plants per acre. Forage yields at Brookings were reduced by the atrazine plus oil treatment and the plant population was 31,500 plants per acre. The delay in maturity caused by atrazine plus oil plus the high plant population at the Southeast Research Farm caused the plants to have more immature heads and a higher ratio of forage to the grain yield.

SUMMARY

The atrazine plus oil suspension severely limited the net CO_2 uptake of plant leaves grown both in the growth chamber and under field conditions. The growing conditions of the plants prior to and following spraying are very important. Plants grown when temperatures were less than seventy degrees Fahrenheit and with normal light intensity were severely affected by the atrazine plus oil treatment. The plants grown at temperatures greater than seventy degrees Fahrenheit and with normal light intensity were not as severely affected in their ability to recover to the normal CO_2 uptake rate. The effect of limited CO_2 uptake was expressed in the reduced height and dry weight of these plants treated with atrazine plus oil. The maturity was also delayed in the sorghum plants sprayed with atrazine plus oil. Grain yields of plants grown under weed-free conditions and sprayed with atrazine plus oil were reduced slightly when compared to weed-free control plants but these differences were not significant at the ninety-five percent level of confidence.

Results indicate that a farmer could spray sorghum plants and be assured of receiving minimum crop injury under growing conditions where the daily temperature prior to and following spraying was not below seventy degrees Fahrenheit. When the daily temperature was below seventy degrees Fahrenheit prior to and after spraying the farmer could expect more severe injuries to the sprayed sorghum plants.

A farmer, in making a decision whether to spray a sorghum field with atrazine plus oil, should consider the weather conditions prior to spraying and following spraying. The severity of the annual weed infestation should also be considered, as annual weeds can also reduce height, delay maturity, and reduce grain yields because of the competition, as determined in studies conducted under the direction of South Dakota State University.

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APPENDIX I

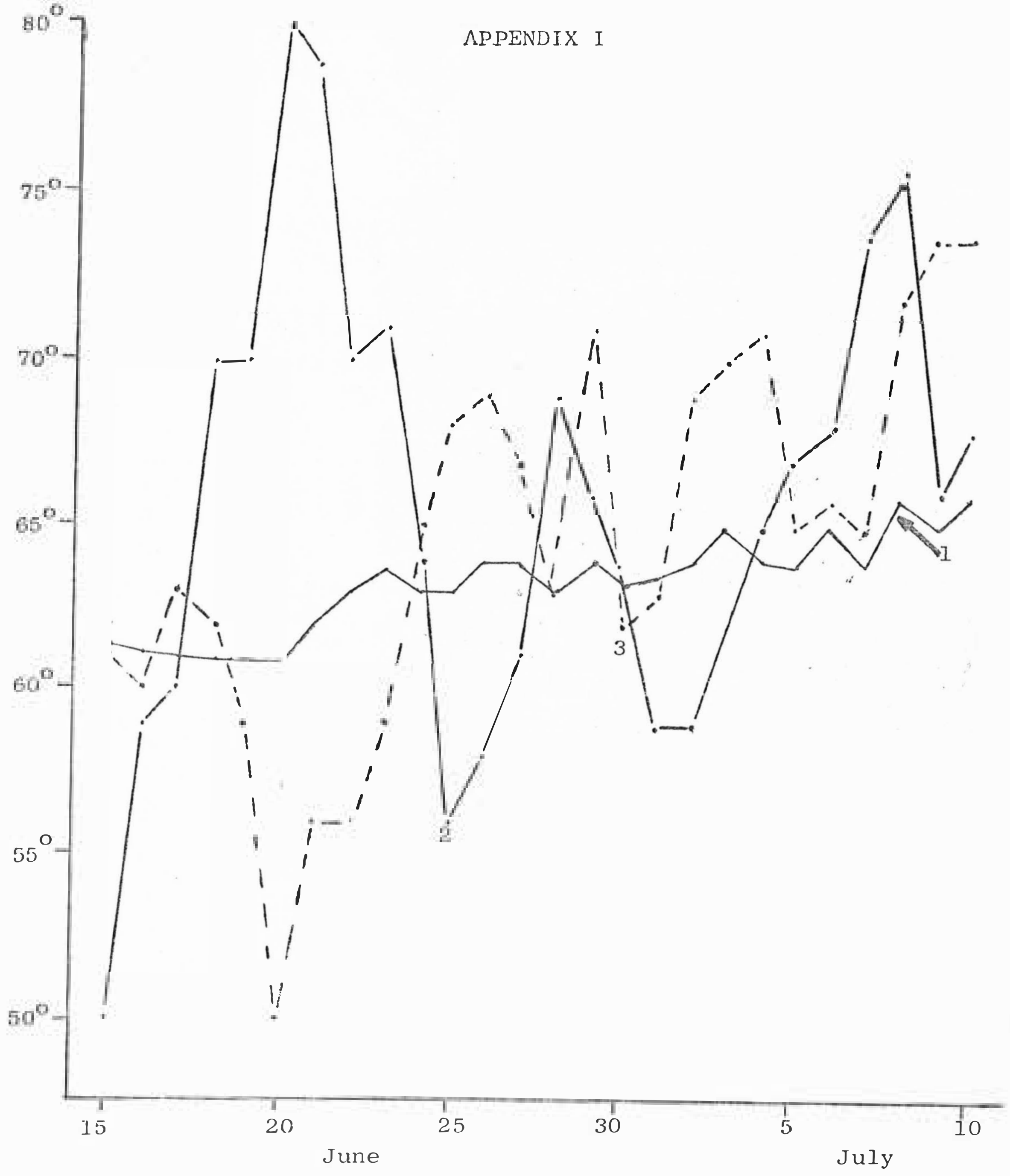

Figure I. The mean temperatures for Brookings location 1968, 1969, and thirty-year average.

1. Thirty-year average mean temperature.
2. June 25 date of spraying 1968.
3. June 30 date of spraying 1969.