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I am submitting herewith a dissertation written by John London Oakes entitled "Whole Plant and Cellular Response of Wild Garlic (Allium vineale L.) to Thifensulfuron-Methyl." I have examined the final copy of this dissertation for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Doctor of Philosophy, with a major in Plant and Soil Science.

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WHOLE PLANT AND CELLULAR RESPONSE OF WILD GARLIC (*ALLIUM VINEALE* L.) TO THIFENSULFURON-METHYL

A Dissertation

Presented for the

Doctor of Philosophy

Degree

The University of Tennessee, Knoxville

John London Oakes

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DEDICATION

To my father, Gordon William Oakes, June 6, 1908 to November 4, 1991.

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ABSTRACT

Field, greenhouse and laboratory experiments were conducted to investigate whole plant and cellular response of wild garlic to thifensulfuron-methyl.

Thifensulfuron-methyl activity on wild garlic was evaluated as influenced by rate and time of application, antagonism and sequential application. Phytotoxicity to small grains was also assessed. Thifensulfuron-methyl at 9 g ai ha^{-1} was the lowest rate observed that suppressed aerial bulblet production greater than 90% when compared to the standard treatment of 2,4-D. Suppression was greatest at the 35 g ai ha^{-1} applied in late March to early April. The lowest rate for reduction of underground bulbs was 35 g ai ha^{-1} applied in late March.

Tank-mixing metribuzin at 210, 420 and 630 g ai ha^{-1} with thifensulfuron-methyl antagonized the latter's activity on wild garlic aerial bulblet suppression. Reduction in underground bulb and aerial bulblet weight was antagonized by tank-mixing metribuzin with thifensulfuron-methyl. The antagonism of thifensulfuron-methyl only occurred for rates of 4 to 9 g ai ha^{-1} and was totally overcome at 17.5 g ai ha^{-1}.

Sequential applications of thifensulfuron-methyl followed by metribuzin at 12 or 24 h or metribuzin followed by thifensulfuron-methyl at 12 or 24 h did not result in antagonized activity of thifensulfuron-methyl on wild garlic whole plant growth or aerial bulblet production. The lower rates of thifensulfuron-methyl followed at 12 h by metribuzin at 630 g ai ha^{-1} was antagonized as evidenced by increases in underground bulb weights.

Thifensulfuron-methyl decreased bulb quality, shoot weight, number of roots per bulb and root and shoot length even at 4 g ai ha^{-1} in the greenhouse. The addition of 420 g ai ha^{-1} to all rates of thifensulfuron-methyl reduced the latter's ability to inhibit shoot growth.

Thifensulfuron-methyl applied at rates up to 35 g ai ha^{-1} was not phytotoxic to wheat or barley.

Thifensulfuron-methyl reduced shoot and root growth of aerial bulblets in sterile culture. The amino acids valine, leucine and isoleucine were inconsistent in overcoming the effects of thifensulfuron-methyl on aerial bulblet growth. All concentrations of thifensulfuron-methyl except 0.001 mg L^{-1} inhibited growth of soft offset bulbs following 10 d of exposure. The addition of valine, leucine and isoleucine did not totally nullify the activity of thifensulfuron-methyl on soft offset bulb growth.

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CHAPTER I

INTRODUCTION

Wild garlic (Allium vineale L.) is a major weed problem in wheat for many areas of the United States. The origin of wild garlic is unknown but it likely originated in the Mediterranean region.

Zimdahl (1989) in the text; "Weeds and Words", discusses the origin of the name Allium vineale. Allium is Latin (L) meaning garlic; vinea is (L) for wine colored, red, or of vineyard; and vinealis is (L) for belonging to the vine. Linnaeus may have thought the pink or red basal portions of the plant or even the flowers resembled the color of wine. It is also possible he found wild garlic commonly growing in vineyards.

Wild garlic was brought to America in wheat (Triticum aestivium L.) grain during the 1600's by European settlers, and has since spread throughout much of the United States. Its geographic range includes the eastern United States and west into Oklahoma and Kansas. Wild garlic is often confused with wild onion (Allium canadense L.).

The growth and development of wheat parallels that of wild garlic, except the latter produces two plant types, scapigerous and nonscapigerous. Scapigerous plants produce aerial bulblets on scapes at approximately the same height as the head of wheat.

Aerial bulblets of wild garlic cannot be separated from wheat during harvest. Bulblets contain volatile chemicals (methyl disulfide, methyl trisulfide, n-propyl disulfide etc.) that give wheat an objectionable odor (Carson and Wong 1961; Freeman and Mossadeghi 1970; Davey et al. 1974; Shelby and Collin 1976). This results in reduced quality, interference with milling of wheat and off-taste in baked products if all bulblets are not removed.

Cultural practices and crop rotation have historically been used to control wild garlic (Sherwood 1944; Pieper and Rickey 1930; Watts 1895). These included rotation of crops and cultivation of fallow land. In the last four decades, herbicides have been used to control wild garlic. During this period 2,4-D (2,4-dichlorophenoxy acetic acid) was used with varying degrees of success (Sherwood 1944; Klingman 1950; Portz and Lobenstein 1955). Improved efficacy of herbicides for control of wild garlic is needed today.

Winter wheat is a major crop for Tennessee farmers. Winter wheat was grown on more than 160,000 ha in 1987 (Anonymous 1987). Though winter wheat is not usually subjected to heavy weed pressure in Tennessee, wild garlic does reduce the quality of harvested grain. Effective control of aerial bulblets from wild garlic would improve wheat grain quality and be financially advantageous to grain producers.

CHAPTER II

LITERATURE REVIEW

Geographic Distribution of Wild Garlic

Wild garlic (Allium vineale L.) is distributed throughout much of Europe and the United States. The origin of this plant is probably left to conjecture but it is assumed to have originated near the Mediterranean Sea (Peters et al. 1965). Through culture of small grains, it has spread throughout western and central Europe where it may occur as far north as Norway and east into the Soviet Union (Peters et al. 1965). Its introduction into the United States started during colonial settlement (Iltis 1949). It covers a geographic region in the United States from the eastern seaboard to the Mississippi River and extends into Kansas and Oklahoma (Peters et al. 1965). It is this area of the United States where wild garlic represents a serious pest in soft red winter wheat. Its effect on wheat grain quality as well as objectionable flavoring of milk is well known (Lee 1955; Peters et al. 1965; Peters 1975).

Morphology

Wild garlic may grow in association with similar bulb formers such as Allium cernuum L.(nodding wild onion), A. canadense L.(wild onion), A. mutabile Michx.(changeable, Michx. having confused two different species of wild onion) and Ornithogalum umbellatum (Star-of-Bethlehem) (Peters et al. 1965). A. canadense L.(wild onion) is often confused with A. vineale L. as evidenced by the attempts of numerous authors to clarify the difference between species (Richens 1947; Fernald 1950; Hakansson 1963; Peters et al. 1965); Peters 1976; Steyermark 1981. It also differs from cultivated garlic (Allium sativum L.) in bulb morphology (Mann 1952).

Peters and Stritzke (1976) noted that wild garlic looks much like cultivated onion and probably is one reason it is often confused with wild onion. Wild garlic and wild onion are often used interchangeably (Watts 1895; Pieper and Rickey 1930). Though they appear similar, they may be distinguished by leaf morphology. Wild garlic has a hollow leaf while wild onion has a flat, solid leaf (Peters and Stritzke 1976). There is also some confusion as to the correct scientific and common name for wild garlic (Iltis 1949; Richens 1947). Steyermark (1981) refers to A. vineale L. as field garlic, while A. oleraceum L. is called wild garlic. A. oleraceum has nearly flattened leaf blades toward the summit instead of hollow blades, yet the bulbs are simple.

This confusion is also noted by Richens (1947), who questioned the uniqueness of three varieties of A. vineale in Great Britain. Peters et al. (1965) commented in their review that all Allium species have a total of 16 chromosomes, but A. vineale has 32 chromosomes.

Wild garlic produces two plant types, scapigerous and nonscapigerous (Peters et al. 1965; Davis and Peters 1965; Peters 1975; Stritzke 1972). Scapigerous plants are often larger and produce aerial bulblets on scapes. (Peters 1965). Scapigerous plants may also produce flowers in some instances. Nonscapigerous plants are typically smaller, have fewer foliage leaves and do not produce a scape at the termination of growth (Peters et al. 1965).

Evidence indicates that a plant may produce up to 300 aerial bulblets per scape (Peters et al. 1965). No reason has been determined as to why some plants may be scapigerous or nonscapigerous. Richens (1947) thought development of scapigerous or nonscapigerous plants could be due to food reserves within the bulb.

Krochmal (1960) conducted germination studies on aerial bulblets. Aerial bulblets stored for 6 mon failed to germinate at temperatures of 0.5 and 4 C. Aerial bulblets stored at room temperature (22 C) for 6 mon had 24% germination.

Bulb distribution in the soil is quite variable and may range from near the surface to a depth of 1 M (Richens 1947).

Bulbs are most abundant between the 2- to 6-cm depth with diminishing frequency to about 15 cm (Richens 1947). Planting soft offset and central bulbs at depths of 10 cm or more reduced the number of scapes (Lazenby 1961). When bulbs were planted at 20 cm, scape production was reduced to 52%.

Freeman (1958) studied the seasonal emergence of bulb types from wild garlic. Total emergence percentages were: aerial bulblets at the 2.5-cm depth, 95%; soft shell bulbs at the 7.6-cm depth, 97%; terminal bulbs at the 7.6-cm depth, 99%; and hard shell bulbs at the 7.6-cm depth, 40%.

Stritzke and Peters (1970) found all hardshell bulbs were dormant at maturity of the parent plant. Only 20 to 34% of the hardshell bulbs sprouted after 1 yr. Dormancy period for the central and soft offset bulbs was 2 to 6 wk. The shorter dormancy period for central bulbs and soft offset bulbs was thought to be related to after-ripening of the embryonic shoot.

Anatomy of all bulb types of wild garlic has been studied by Stritzke and Peters (1972). Primordia development is the same in all bulb types. Embryonic plants in soft offset and central bulbs were the same, with numerous root primordia and four-leaf primordia. Embryonic plants of aerial bulblets and hardshell bulbs had three leaf primordia and one or two root primordia.

Aerial bulblets produced by scapigerous plants consist of a fleshy, cone-shaped scale containing a growing point at its base (Peters et al. 1965).

The scale is a bladeless storage leaf which is protected by a second scale, three cell layers in thickness. Iltis (1949) indicated that the second scale was bladeless in most cases, but could include a blade referred to as an "appendage". The scapigerous plant may also produce seed, but seed production is considered to be an insignificant means of reproduction.

Underground reproductive structures of scapigerous plants are hardshell and soft offset bulbs (Peters et al. 1965). The hardshell bulb is the second most numerous bulb type and contains a single storage leaf surrounded by a bladeless leaf forming a protective cover (Peters et al. 1965). Growth of this bulb type starts in mid-August or early September and usually ceases in October or November. The soft offset bulb is formed in the axil of the innermost leaf of scapigerous plants (Peters et al. 1965).

The nonscapigerous plant forms either underground hardshell bulbs or fleshy central bulbs. The hardshell bulb is the only bulb type produced by both scapigerous and nonscapigerous plants (Peters et al. 1965). The hardshell bulb is formed in underground axils of the outer leaves of both plant types. The fleshy central bulb is quite large and conspicuous at the end of the growing season. It develops around the main axis of the plant and may vary in size from that of an aerial bulblet to a soft offset bulb (Peters and Stritzke 1976).

Early Control Methods of Wild Garlic

Control prior to the 1950's was restricted to cultural practices. Cultural practices were often ineffective in controlling wild garlic, primarily due to a large food reserve in the bulbs (Peters et al. 1965).

Lazenby (1963) studied the effects of cutting on Allium vineale. Frequency of defoliation by cutting must be coordinated with seasonal timing for the greatest reduction in bulb weight and subsequent reduction in population.

Watts (1895), in early work at the University of Tennessee Experiment Station noted that preventing dissemention of bulbs and aerial bulblets was better than any known cure. In circumstances where wild garlic was already established the best control was deep plowing. Other possible measures included cultivation of certain row crops and mowing. All of these methods met with limited success. One cultural technique employed by Watts was shaving the soil. The method utilized a blade attached to a drawn implement which scrapes or shaves the soil surface. At the time of the experiment a high degree of success in eradication of wild garlic was noted. The practicality of this method was questioned and based on literature it was not widely adopted.

Wild (1928) investigated control methods for crow garlic (Allium vineale) and found that close grazing with cattle and sheep was the most effective control measure.

Pieper and Rickey (1930) advocated cultural practices for control of wild garlic over chemical treatments. They reported that in 1897 the U.S.D.A. experimented with fertilizers and chemicals such as carbolic acid, fuel oil, sodium arsenite and common salt. In using these compounds only the tops of plants were killed with little damage to the underground bulb. Chlorates were also used with greater success but were considered to be financially impractical for field applications. In research conducted by Pieper and Rickey (1930) chlorates were used to control wild garlic. Their results indicated reduction in wild garlic populations, but the treatment was considered to be financially impractical for an entire field.

During the 1940's and 1950's work continued with chlorates and 2,4-D (Sherwood 1944; Lee 1955; Klingman 1950). Sherwood (1944) recommended the use of sodium chlorate at a rate of 7 to 9 kg per 25 m^2. This proved to be expensive so an alternative recommendation was kerosene, waste oil or fuel oil at 1.9 L per 25 m^2. Lee (1955) recommended 2,4-D as a spring application over a period of 2 to 3 yr to control wild garlic. Raleigh (1951) utilized 2,4-D amine and ester formulations in the control of wild garlic. The ester and amine formulations at 1.1 to 2.2 kg ha^{-1} provided excellent control of wild garlic in pasture. Hard shell bulblets which germinated in the fall required the same treatment the following spring.

Portz and Lobenstein (1955) used the ester formulation of 2,4-D to control wild garlic in winter wheat. A 70% reduction in aerial bulblet production from wild garlic was obtained while wheat yields were 95% of the check. This rate reduced underground bulb production by 30%. Lobenstein and Portz (1956) evaluated 2,4-D on the control of wild garlic in grass sod. Reduction in underground bulb weight was dependent on the rate of 2,4-D. A reduction of 31%, 71%, and 85% of the untreated check was obtained at 0.8, 1.7 and 3.4 kg ai ha^{-1}, respectively. Allmond (1957) found 1.1 kg ha^{-1} 2,4-D applied in November to be effective in reducing wild garlic stand. Although the stand was reduced, hardshell bulblets remained a problem.

During the 1960's and 1970's much of the same control strategy continued. Davis et al. (1962) evaluated 2,3,6-TBA (2,3,6 trichlorobenzoic acid) for wild garlic control. The phytotoxicity of 2,3,6-TBA was increased with the addition of a wetting agent. Results revealed 2,3,6-TBA to be superior to 2,4-D in control of wild garlic. Davis et al. (1965) also found that 2,3,6-TBA at 2.2 kg ha^{-1} reduced hardshell bulb production more than 2,4-D. Binning and Meggitt (1968) noted that 2,4-D and dicamba (3,6-dichloro-o-anisic acid) at 0.56 and 0.28 kg ha^{-1}, respectively, resulted in good control of wild garlic with only a slight reduction in wheat yield. At the five-tiller stage, 2,4-D stunted hard red winter wheat (French et al. 1977).

Peters and McKelvey (1982) were successful in the eradication of wild garlic from test plots with 1.1 kg ha^{-1} of paraquat over a 4-yr period. When shallow tillage was included with paraquat (1,1'-dimethyl-4,4'bipyridinium ion) eradication occurred in less time.

Sulfonylurea Herbicides

History

The early sulfonylurea herbicides were derivatives of triazine herbicides and exhibited herbicidal symptoms comparable to the parent compound (Beyer et al. 1987). Herbicidal properties for the sulfonylurea herbicides were first reported in 1966 (Beyer et al. 1987). Development of sulfonylureas was slow until the 1970's when Levitt noted growth regulating activity of a sulfonylurea compound. By 1985 patents for sulfonylurea herbicides were held by 14 agricultural chemical companies (Beyer et al. 1987; Miyamoto and Kearney 1983).

Symptomology

The sulfonylurea herbicides inhibit plant growth in several ways. Symptoms include enhanced anthocyanin formation, loss of leaf nyctinasty, vein discoloration, terminal bud death and finally necrosis (Beyer et al. 1987). Although symptoms may develop over several days or weeks, sulfonylurea potency is unique. Activity has been noted at rates of 2 g ha^{-1} in field trials (Beyer et al. 1987).

Physical and Chemical Properties

Physical and chemical properties of sulfonylurea herbicides have been reviewed by several authors (Beyer et al. 1987; Levitt 1983). In general, sulfonylurea herbicides exhibit moderate melting points, low octanol-water partition coefficients at neutral pH, low vapor pressures and moderate water solubilities at neutral pH. They are weak acids and therefore solubility is affected by pH. For example, water solubility of chlorsulfuron at pH 5 and pH 7 is 60 and 7000 ppm, respectively (Beyer et al. 1987). Solution pH has an opposite effect on octanol-water partition coefficient. As pH increases more of the sulfonylurea exists in the anionic form (water soluble) and accordingly a decrease in partitioning into octanol.

Mode of Action

Sulfonylurea herbicides inhibit branch chain amino acid biosynthesis in plants and bacteria. The specific site for inhibition is acetolactate synthase (ALS) (Beyer et al. 1987; Ray 1985; Chaleff and Mauvis 1984; Ray 1984b; Erbes 1984; Dierk and Casida 1984; Anonymous 1986).

As early as 1980 the mode of action of the sulfonylurea, chlorsulfuron (2-chloro-N-[[(4-methoxy-6-methyl-1,3,5-triazin-2-yl)amino]carbonyl]benzenesulfonamide), was studied (Ray 1980, 1982). Ray (1980) observed 2.8 x 10^{-6} M chlorsulfuron reduced pea (Pisum sativum L.) shoot mitotic cell figures to 0.9 per 100 cells compared to 6.4 per 100 cells for the control.

Ray (1982) conducted studies to measure physiological and biochemical responses on corn (Zea mays L.) seedlings. The most noticeable response was a reduction in plant growth. Chlorsulfuron at 1 ppb totally inhibited shoot growth in corn. Chlorsulfuron also strongly inhibited cell division. DNA synthesis was strongly inhibited with little effect on RNA synthesis.

LaRossa and Schloss (1984) noted that many biochemical functions are the same in plants and bacteria. S.typhimurium growth was inhibited by sulfometuron-methyl (2-[[[[4,6-dimethyl 2-pyrimidinyl)amino]carbonyl]amino] sulfonyl]benzoic acid). Inhibition could be reversed by valine and isoleucine but not by other amino acids.

Ray (1984a) concluded that chlorsulfuron and sulfometuron-methyl inhibited valine and isoleucine synthesis by blocking ALS activity. Results indicated that for excised pea roots an ALS I_{50} value with chorsulfuron was 21 nM.

Chaleff and Mauvis (1984) utilized a chlorsulfuron resistant tobacco (Nicotiana tabacum L.) mutant to determine the mode of action of chlorsulfuron. Specific mutations from a herbicide resistant tobacco cell line exhibited resistance to chlorsulfuron, as indicated by ALS activity.

Growth of excised pea roots in culture was inhibited by chlorsulfuron at 28 nM (Ray 1985). Supplements of casein hydrolysate, valine, isoleucine and valine plus isoleucine would overcome this inhibition.

The addition of valine and isoleucine each at 100 uM gave complete protection against chlorsulfuron inhibition. These results agree with LaRossa and Schloss (1984) on reversal of growth inhibition with the addition of valine and isoleucine.

Tolerance to chlorsulfuron was studied by Hutchison et al. (1984). Flax (Linum usitatissimum L.) and black nightshade (Solanum nigrum L.) were evaluated for tolerance to ^{14}C-chlorsulfuron. Flax and black nightshade tolerated chlorsulfuron by metabolizing it to nontoxic metabolites.

Sensitivity of maize to chlorsulfuron was evaluated by Landi et al. (1989). Susceptible lines exhibited root length reduction. Field experiments measured the susceptibility of maize to chlorsulfuron by collecting information on shoot height, shoot dry weight, and percent shoot dry matter. Hybrids and parental lines had different responses to chlorsulfuron. There was a significant interaction of hybrid by line and herbicide rate for shoot height and dry weight. The general conclusion was that tolerance seemed to be under additive genetic control which resulted in tolerant hybrids from crosses among tolerant lines.

At the end of 6 wk chlorsulfuron and metsulfuron (2-[[[[(4-methoxy-6-methyl-1,3,5-triazin-2-yl)amino]carbonyl] amino]sulfonylbenzoic acid) reduced wild garlic shoot height, number of leaves and shoot dry weight (Leys and Slife 1988). Herbicide placement was not a factor in performance of chlorsulfuron or metsulfuron. Absorption and translocation of ^{14}C-chlorsulfuron or ^{14}C-metsulfuron was not different.

No difference was noted between herbicides in amount of radioactivity translocated from the treated leaf. Slow translocation through the symplast was evident for both herbicides. At 144 h following treatment, the treated leaf contained concentrations of 6000 and 3700 ng of chlorsulfuron and metsulfuron per mg fresh weight, respectively. At the same time, chlorsulfuron and metsulfuron were present in the roots at 100 and 80 ng mg^{-1} fresh weight, respectively. The slow rate of translocation and low dosages did not inhibit the control of wild garlic in this study.

Devine and Vanden-Born (1985) studied the translocation of ^{14}C-chlorsulfuron in Canada thistle (Cirsium arvense L.) After 144 h only 5% of the applied ^{14}C-chlorsulfuron was recovered from roots and developing buds.

In related work, Petersen and Swisher (1985) investigated the absorption, translocation and metabolism of ^{14}C (0.17 µCi) chlorsulfuron in Canada thistle. After 48 h 10% of foliar-applied ^{14}C-chlorsulfuron had translocated from the treated leaf and roots. When applied as a foliar treatment, ^{14}C-chlorsulfuron was not metabolized by Canada thistle after 48 h.

Devine et al. (1984), evaluated leaf-wash solutions and techniques for retention of ^{14}C-chlorsulfuron on Canada thistle. All wash solutions and techniques removed similar amounts of ^{14}C-chlorsulfuron from leaves.

Mersie and Foy (1985,1986) studied the influence of simulated rain on the phytotoxicity of chlorsulfuron to velvetleaf (Abutilon theophrasti Medic). Rates of chlorsulfuron penetration into leaves and absorption into roots from various pH solutions were examined. Chlorsulfuron phytotoxicity was enhanced by simulated rain at pH's of 2.5 and 3.4 compared to pH's of 4.4 and 5.6.

Bestman and Vanden-Born (1985) examined ^{14}C-chlorsulfuron movement in apical tissue of field pennycress (Thlaspi arvense L.). A 1 ug rate of ^{14}C chlorsulfuron was applied to the leaf and followed 3 or 24 h later by a 30 min pulse of $^{14}CO_2$. The pulse had no effect on ^{14}C-chlorsulfuron incorporated in the shoot 2 or 8 h after application.

Additional investigation of differential metabolism as a means of selectivity for chlorsulfuron was conducted by Sweetser et al. (1982). A number of sensitive species [sugar beets (Beta vulgaris L.), soybean (Glycine max L.), mustard (Brassica spp.) and cotton (Gossypium barbadense L.)] and tolerant species [wheat (Triticum aestivum L.), barley (Hordeum vulgare L.) and wild oats (Avena fatua L.)] were evaluated using ^{14}C-chlorsulfuron. During a 24-h study variability of penetration following treatment with ^{14}C-chlorsulfuron was noted. Mustard (Brassica kaber (D.C.) exhibited 98% penetration, while cotton exhibited only 56%. In both tolerant and sensitive species, the greatest translocation was in leaves and stems with very little into roots.

After 24 h mustard contained 18% of the original dosage of ^{14}C-chlorsulfuron in leaves and stems. Barley leaves and stems contained 1% of the original dosage. All tolerant species were able to metabolize chlorsulfuron to the O-glycoside metabolite of chlorsulfuron. Sensitive species metabolized little chlorsulfuron.

Ray (1984b) noted that 28 nM (10 ppb) chlorsulfuron inhibited excised pea roots. The enzyme ALS was sensitive to 21 nM (7.5 ppb). Erbes (1984) found that tolerant plants metabolized chlorsulfuron to hydroxylated and glucosylated derivatives. The hydroxylated-derivatives inhibited ALS from sulfonylurea sensitive and tolerant plants the same as the parent sulfonylurea. The glucose conjugate slightly inhibited ALS. Plant tolerance was due to metabolism of the parent compound to a glucose conjugate.

Chlorsulfuron inhibited Escherichia coli growth at 0.3 and 1.0 mM by suppressing RNA synthesis (Dierk and Casida 1984). Suspension culture of soybean cells had 50% inhibition of growth with 0.6 nM chlorsulfuron. Valine, leucine and 2-oxoisovalerate alleviated the inhibitory effects of chlorsulfuron.

Matsunaka et al. (1985), utilized tobacco cell culture to evaluate growth inhibition of chlorsulfuron at 1 ppb. Inhibitory effects were overcome with the addition of valine, leucine or isoleucine. Chlorsulfuron at 40 ppb totally inhibited the activity of ALS extracted from tobacco cell culture.

Recovery of chlorsulfuron induced inhibition was studied by Rost and Reynolds (1985). Excised pea root tips were grown in culture and treated with 28 nM chlorsulfuron. The percentage of mitotic figures did not recover following excision. Addition of valine and isoleucine reversed the effect of chlorsulfuron on cell mitotis. Similar research conducted by Rost (1984) noted the inhibitory effect of chlorsulfuron at 2.8 uM to 280 uM. All concentrations inhibited the progression of individual cells from G^2 (spindle fiber synthesis) to mitosis.

DeAgazio and Giardina (1984) evaluated the influence of chlorsulfuron on maize roots. They proposed the plasma membrane as the site of action for chlorsulfuron. Comparisons were made with the K^+ transport system from control and chlorsulfuron pretreated maize seedlings. Inhibition of K+ uptake was evident after 12 h pretreatment with 10 uM chlorsulfuron. The inhibition was evident only after maize root segments were washed for 2 h in a $CaSO_4$ solution.

There is little evidence to contradict that valine, leucine or isoleucine can bring about recovery from chlorsulfuron. Giardina et al. (1987) evaluated the role of valine and isoleucine as antidotes to chlorsulfuron. Additional studies were conducted with chlorsulfuron on root elongation and K^+ uptake. The addition of valine and isoleucine at 100 uM did not remove the inhibitory effect of chlorsulfuron at 28 uM.

The fresh weight of maize roots was essentially the same for roots with no amino acids or with valine plus isoleucine. Inhibition of ALS *in vitro* by chlorsulfuron, but not *in vivo*, was noted. Since the *in vivo* response was not noted, the authors suggested the possibility of more than one site of action for chlorsulfuron.

Klingman and Peeper (1989) used a bean root bioassay to evaluate the role of amino acids on nullifying the effects of CGA-131036 (N-(6-methoxy-4-methyl-1,3,5-triazin-2-yl amino-carbonyl)-2(2-chloroethoxy)-benzenesulfonamide). The addition of 100 uM valine and isoleucine partially nullified the growth inhibition caused by CGA-131036. CGA-131036 at 10 to 300 nM in the medium supplemented with valine and isoleucine produced bean root lengths greater than roots grown without supplemented amino acids.

Brown and Neighbors (1987) investigated soybean metabolism of chlorimuron-ethyl (2-([4-chloro-6-methoxy pyrimidine-2-yl) amino carbonyl] sulfonyl)benzoic acid, ethyl ester) and the physiological basis for selectivity. Herbicidal efficacy of chlorimuron-ethyl and its metabolites *in vitro* on soybean ALS was measured. Chlorimuron-ethyl at 6.2 ± 1.2 nM inhibited soybean ALS by 50%. Soybean ALS I_{50} values for the deesterified free acid analog, chlorimuron, and the chlorimuron-ethyl-GSH conjugate were >13,000 and >7,000 nM, respectively.

Weed Control Efficacy

Weed control in small grains with sulfonylureas is of primary interest (Dyer and Fay 1981; Ambach et al. 1984; Eberlein and Miller 1984; Sthalman 1983a,b; Hagood 1987; Morrow and Porter 1987; Webb and Johnson 1987; Dial et al. 1987). Use of chlorsulfuron has become widespread since its introduction in 1982 (Beyer et al. 1987; Oswald and Richardson 1985; Dyer and Fay 1981; Foy and Mersie 1984; Miller and Alley 1985; Nalewaja and Gillepsie 1984; Nalewaja and Wozinca 1985; O'Sullivan 1982; Schumacher et al. 1982; Retzinger and Kitchen 1984; Donald 1984; Thill et al. 1982).

Selly et al. (1985) evaluated chlorsulfuron and metsulfuron-methyl alone and in mixture for weed control in cereal grains. Control of broadleaf weeds was obtained when chlorsulfuron and metsulfuron-methyl were tank-mixed and applied in the autumn.

Leys and Slife (1986) compared fall and spring applications of chlorsulfuron for wild garlic control. Chlorsulfuron at 20 g ai ha^{-1} applied in the spring reduced aerial bulblet production by 99% while fall application only provided 59% reduction. Fall application of chlorsulfuron was better than the standard treatment of 2,4-D.

In a following study, Leys and Slife (1987) compared chlorsulfuron and metsulfuron for control of wild garlic. Chlorsulfuron at 20 g ha^{-1} applied in the spring was more effective than when applied in the fall.

Time of application was less critical for metsulfuron. Metsulfuron at 5 to 20 g ai ha^{-1} applied in the spring or fall reduced aerial bulblet production and plant density greater than 94%.

Ambach et al. (1984) evaluated thifensulfuron-methyl (formerly thiameturon, DPX-M6316) for control of a wide range of broadleaf weeds in small grains. Control of such species as wild buckwheat (Polygonum convolvulus L.), kochia (Kochia scoparia (L.) Schrod.) and Russian thistle (Salsola kali (L.) var. tenuifola Tausch.) has been achieved at 35 g ai ha^{-1}. Tolerance of small grains such as winter wheat, spring wheat and spring barley to thifensulfuron-methyl has been excellent. Persistence is not a problem since thifensulfuron-methyl has little soil activity (Brown et al. 1987).

Nalewaja and Gillespie (1984) evaluated thifensulfuron-methyl at rates of 3 to 28 g ha^{-1}. Weed control ranged from 30 to 98% for yellow foxtail (Setaria glauca (L.) Beauv.), wild mustard (Brassica kaber D.C.), kochia and redroot pigweed (Amaranthus retroflexus L.). Thifensulfuron-methyl applied in the spring controlled wild garlic with no carryover to double-cropped soybeans (Kaufman and Ritter 1987). Short soil residual activity as well as excellent weed control have also been noted by Ferguson et al. (1987).

Weise et al. (1986) noted activity from chlorsulfuron at 280 g ha^{-1} for 25 mon following treatment. The presence of chlorsulfuron was not injurious to sorghum (Sorghum bicolor L.) in a sorghum-wheat-sorghum cropping sequence.

Injury from chlorsulfuron has been noted on rotational crops (Peterson and Arnold 1985). Corn and sorghum were the most sensitive while flax was the least sensitive rotational crop after 1 y. Soybeans were injured and yields were reduced following wheat treated with 36 or 72 g ha^{-1} chlorsulfuron (Khodayari et al. 1985).

Brewster and Appleby (1983) did not notice any injury to wheat from chlorsulfuron at 140 g ha^{-1}. Residual activity of chlorsulfuron at 33 g ha^{-1} and greater was injurious to several rotational crops. Khodayari et al. (1985) noted little injury to wheat from chlorsulfuron applied at 9 or 18 g ha^{-1}, or in tank-mixtures with 2,4-D amine, bentazon, bromoxynil, metribuzin or diclofop.

Wild garlic aerial bulblet control has been excellent with thifensulfuron-methyl at rates from 7 to 21 g ha^{-1} with slight injury to oats (Avena sativa L.) in southern Illinois (Anderson and Kapusta 1987,1988; Kapusta and Anderson 1987). There was no reduction in oat yields from thifensulfuron-methyl (Durgan and Miller 1988).

Wild garlic control was excellent when thifensulfuron-methyl was applied to soft red winter wheat (Kapusta 1987). The best control of wild garlic aerial bulblets occurred with thifensulfuron-methyl applied on April 10 compared to March 24 or April 30. Double-cropped soybeans following wheat were unaffected by thifensulfuron-methyl.

Control of wild garlic offset bulbs was better for all rates of thifensulfuron applied in late March or early April compared to late April (Gast et al. 1990). The greatest reduction in offset bulb numbers occurred with thifensulfuron applied on March 21.

Wild garlic control was evaluated using chlorsulfuron, metsulfuron-methyl and 2,4-D low volatile ester (Kapusta and Strieker, 1983). All herbicides controlled aerial bulblets. Spring-applied chlorsulfuron injured developing double-cropped soybeans.

Aerial bulblets were reduced 90% by thifensulfuron-methyl at 9 to 70 g ai ha^{-1} applied on March 21, April 8, or April 28 (Gast and Slife 1987). Absorption studies revealed limited absorption of ^{14}C-thifensulfuron-methyl. After 12 h only 1% of the total thifensulfuron-methyl had been absorbed and only 12% was absorbed after 144 h.

The experimental sulfonylurea herbicide CGA-131036 controlled weeds and was selective in wheat and barley (Somody et al. 1987). Weeds less than 5-cm tall were most susceptible. Tank-mixtures of CGA-131036 with other herbicides resulted in antagonism on some weed species.

Chlorsulfuron and CGA-131036 at 9 and 10 g ai ha^{-1}, respectively, completely controlled flixweed (Descurainia pinnata Walt.) in winter wheat (Klingman and Peeper 1989). At similar rates, CGA-131036 was less effective than chlorsulfuron on henbit (Lamium amplexicaule L.) and Italian ryegrass (Lolium multiflorum Lam.).

Chlorsulfuron was evaluated for broadleaf weed control in newly seeded alfalfa (Medicago sativa L.) (Stannard and Fay 1987). The efficacy of chlorsulfuron in alfalfa was directly related to its soil persistence. Several alfalfa breeding lines were identified with tolerance to chlorsulfuron at 16 g ai ha^{-1}.

Anderson (1987) observed the response of sulfonylurea herbicides on broadleaf weeds in safflower (Carthamus tinctorius L.). Safflower tolerance to chlorsulfuron increased with plant size. Thifensulfuron-methyl applied at 5, 10 and 15 g ai ha^{-1} to safflower in the rosette growth stage did not reduce plant height or grain yield.

Hageman and Behrens (1984) investigated differences in response of eastern black nightshade (Solanum ptycanthum Dun.) and velvetleaf to foliar-applied chlorsulfuron. Eastern black nightshade metabolized 70% of absorbed ^{14}C-chlorsulfuron in 24 h and 81% within 72 h. Velvetleaf was less tolerant, with 7% of absorbed ^{14}C-chlorsulfuron being metabolized in 72 h.

Fielding and Stoller (1990), investigated the effects of additives on efficacy, uptake and translocation of thifensulfuron-methyl. Velvetleaf control and soybean injury increased with increasing rates of thifensulfuron (0, 2.2, 4.4 and 5.8 g ai ha^{-1}).

The addition of a nonionic surfactant (0.25% v/v) plus 28% UAN (urea:ammonium nitrate, 1:1) to thifensulfuron controlled velvetleaf better than nonionic surfactant alone.

Zhao et al. (1990) noted velvetleaf was more sensitive to thifensulfuron at 18 to 180 g ha^{-1} applied to the foliage than to the roots but corn was more susceptible to root than foliar application.

Tolerance of Small Grains

Winter and spring wheat have shown tolerance to postemergence chlorsulfuron (Palm et al. 1980; Cook and McKinley 1980; Morishita et al. 1980; Hageman and Behrens 1981; Martin et al. 1989). DPX-T6376 (metsulfuron-methyl) plus surfactant was evaluated when applied to wheat at the two- and five-tiller growth stages (Stahlman 1983a). Adding a surfactant increased injury and delayed heading of wheat at both growth stages. Wheat yields were also lower when metsulfuron-methyl plus surfactant was applied to 8-cm tillers. Preemergence or postemergence metsulfuron-methyl was applied to six wheat cultivars at three growth stages (two leaves, three tillers, and seven tillers) (Stahlman 1983a). A reduction in yield occurred regardless of application timing.

The mixture of 2,4-D with either thifensulfuron, DPX-L5300 (methyl 2-3[-(4-methoxy-6-methyl-1,3,5-triazin-2-yl]-N-methyl-amino]carbon-yl]amino]sulfonyl]benzoate) or DPX-R9674 (a mixture of 2:1 (w/w) thifensulfuron:DPX-L5300) controlled common chickweed (Stellaria media (L.) Vill.) in winter wheat better that 2,4-D alone (Harrison and Buerlein 1989).

Tolerance of small grains to postemergence chlorsulfuron was due to differential metabolism (Sweetser et al. 1982). Research to evaluate the basis for tolerance to soil residues of chlorsulfuron was conducted by Foley (1985). Wheat and barley were grown in nutrient solution containing ^{14}C-chlorsulfuron. Barley was less tolerant to chlorsulfuron than wheat. Barley root fresh weight was reduced compared to the control. The ^{14}C-chlorsulfuron in the roots of wheat was greater than in barley roots. Wheat and barley metabolized 89 and 77% of applied chlorsulfuron, respectively. The slower metabolism of chlorsulfuron by barley is not the basis for its increased sensitivity to root-applied chlorsulfuron. A possible explanation was a lower ALS I_{50} value in barley roots than previously reported.

Comparisons were made between chlorsulfuron and clopyralid on phloem mobility (Devine et al. 1987). Both herbicides eluted at equal rates when loaded tissue was transferred to a herbicide-free buffer. In whole plant experiments, pretreatment of an individual leaf with chlorsulfuron reduced the export of ^{14}C-sucrose applied to the same leaf 12 h later.

The reduction was not noted when plants were pretreated with a 10x dose of clopyralid.

Residual Activity and Soil Amendments

Chlorsulfuron has extended soil residual activity (Peterson and Arnold 1986; Walker and Brown 1983; Walker et al. 1989; Walker and Welch 1989). Residual activity and movement of chlorsulfuron in a sandy loam soil was investigated (Walker et al. 1983). Their model predicted the most rapid degradation of chlorsulfuron in warmer and wetter areas. A possible limitation of the model was that it did not take into account any soil factors such as pH. Results indicated chlorsulfuron degradation follows first order kinetics (Walker et al. 1983). By increasing soil temperature 10 C, the rate of loss was between 2 and 3 orders. Increasing the soil moisture content from 6 to 12% increased degradation two-fold.

Wondimagegneher and Foy (1985) noted chlorsulfuron became more toxic to corn as soil pH increased from 4.2 to 7.8. Adsorption of chlorsulfuron was stronger at low pH (4.2) than high pH (7.8). This suggested liming could increase chlorsulfuron phytotoxicity since it undergoes low adsorption and high desorption at neutral pH.

Chlorsulfuron adsorption was greatest on humic acids and iron oxides and negligible on montmorillonite clay (Borggard and Streiberg 1988). Chlorsulfuron adsorption decreased when pH increased from 4 to 8, with minimal adsorption at pH 8.

Fredickson and Shea (1986) evaluated the influence of pH on activity of chlorsulfuron in soil. Chlorsulfuron phytotoxicity to sorghum on a silty clay loam decreased as pH increased from 5.9 to 7.5. Uptake of ^{14}C-chlorsulfuron by wheat in a silty clay loam at pH 5.9 was 67 to 100% greater than at pH 7.5. The half-life of ^{14}C-chlorsulfuron in a silty clay loam was 1.9 wk at pH 5.6 and 10 wk at pH 7.5.

Activity of chlorsulfuron as influenced by straw ash and organic matter was investigated by Eleftherohorinos et al. (1985). Addition of 2% peat to a sandy loam soil with 4% organic matter increased chlorsulfuron absorption. Straw ash was considered to be a stronger absorbent with 2% ash being equivalent to 8% peat. There was no indication of decreased herbicide activity by either amendment.

The short residual for thifensulfuron-methyl is the result of rapid soil microbial degradation to herbicidally inactive products (Brown et al. 1987). Thifensulfuron-methyl degraded rapidly at a soil temperature of 30 C. The half-life in several fresh soils was 0.5 to 2.0 d. Soil pH, organic matter, sand, silt and clay content had no effect on degradation. Degradation of thifensulfuron-methyl is 10 to 50 times faster than chlorsulfuron.

Weise et al. (1988) studied the persistence of several sulfonylurea herbicides in a Pullman clay loam. Persistence was measured by sorghum injury over a range of pH (6.5 to 8.0) in a 3-yr wheat-sorghum-fallow rotation. As pH increased chlorsulfuron persistence increased.

When soil pH was 6.5, chlorsulfuron at 34 g ai ha^{-1} did not injure sorghum after 16 mon. When pH of the soil was increased to 7.5, chlorsulfuron at 34 g ai ha^{-1} injured sorghum planted at 25 mon.

Safeners with Sulfonylurea Herbicides

The use of safeners with chlorsulfuron has been evaluated by several researchers (Rubin and Casida 1985; Hatizos 1984; Parker et al. 1980; Sweetser 1985). Results indicate that dichlormid (2,2 dichlor-N,N-di-2-propenylacetamide) proved to be a more effective safener than flurazole (phenylmethyl 2-chloro-4-(trifluoromethyl)-5-thiazolecarboxylate) and CGA-92194 ([N-(1,3-dioxolan-2-yl-methoxy)imino-benzeneaceto-nitrile]) on several corn genotypes (Rubin and Casida (1985). Pretreatment with dichlormid at 1 kg ha^{-1} for 2 d prior to postemergence chlorsulfuron protected all genotypes from herbicide injury. Chlorsulfuron activity decreased root growth more than shoot growth. Safening by dichlormid was evident on root growth.

Safening of chlorsulfuron on cereal grains and the mode of action for antidotes were investigated (Sweetser 1985). Chlorsulfuron and sulfometuron-methyl were evaluated with the safeners dichlormid, cyometrinil((Z)-alpha [cyanomethoxy) imino] benzene aceto-nitrile) and K plus Na. Corn, a sensitive plant, had a significant increase in the rate of chlorsulfuron metabolism with all antidotes. The half-life for the herbicides was reduced from 3.6 h to 1 h in corn leaves treated with K plus Na.

Sensitive broadleaved species did not have a significant increase in metabolism of the sulfonylurea herbicides.

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CHAPTER III

EFFECT OF RATE, TIMING AND METRIBUZIN ON THIFENSULFURON-METHYL ACTIVITY ON WILD GARLIC (*Allium vineale* L.)

Introduction

Wild garlic (*Allium vineale* L.) represents a major weed problem in small grain production. Aerial bulblets cannot be separated from wheat during harvest. Volatile compounds (methyl disulfide, methyl trisulfide, n-propyl disulfide etc.) in aerial bulblets give wheat an objectionable odor (Carson and Wong 1961; Freeman and Mossadeghi 1970; Davey et al. 1974; Shelby and Collin 1976).

Wild garlic produces scapigerous and nonscapigerous plants (Peters et al. 1965; Davis and Peters 1965; Peters 1975; Peters and Stritzke 1976). Scapigerous plants produce aerial bulblets on scapes while nonscapigerous plants do not produce scapes (Peters et al. 1965).

Control of wild garlic has been by cultural and chemical methods (Pieper and Rickey 1930; Klingman 1950; Portz and Lobenstein 1955). The phenoxy herbicide, 2,4-D (2,4-dichlorophenoxy acetic acid), has been extensively used (French et al. 1977; Davis et al. 1965; Binning and Meggitt 1968). Common complaints with 2,4-D have been the lack of effective control of aerial bulblets, injury to small grains and effects on nontarget species from spray and vapor drift.

Wheat treated at the five-tiller stage with 2,4-D at 0.56 kg ai ha^{-1} produced twisted and deformed grain heads (French et al. 1977). Application of 2,3,6-TBA (2,3,6-trichlorobenzoic acid) failed to eradicate wild garlic (Davis et al. (1962).

The sulfonylurea herbicides chlorsulfuron (2-chloro-N-[[(4-methoxy-6-methyl-1,3,5-triazin-2 yl) amino]carbonyl] benzensulfonamide) and thifensulfuron-methyl (methyl (3-[[[(4methoxy-6-methyl-1,3,5-triazin-2yl)amino]carbonyl]amino] sulfonyl]-2-thiophenecarboxylic acid)) have controlled several weed species with little phytotoxicity to small grains (Ferguson et al. 1987; Gast and Slife 1987; Foley 1985; Selley et al. 1985; Nalewaja and Gillepsie 1984; Ambach et al. 1984; Kapusta and Anderson 1987; Anderson and Kapusta 1987; Durgan and Miller 1988).

Thifensulfuron-methyl controls wild garlic aerial bulblets when applied in April (Kapusta 1987). Control in natural stands depended on application timing (Gast 1988). Nearly complete control occurred during the latest application time (May 5), while earlier timings provided less control (Gast 1988).

The interaction of herbicides or other agrochemicals tank-mixed for crop production is of interest. The joint action of agrochemical mixtures on higher plants is classified as either synergism, antagonism or additive effect (Hatizos and Penner 1985).

Little evidence exists as to the effect of tank-mixing thifensulfuron-methyl and metribuzin. Antagonism between diclofop ((±)-2-[4-(2,4-dichlorophenoxy) phenoxy] propanoic) and thifensulfuron-methyl has been noted (Eberlein et al. 1988).

The objective of this study was to determine the most effective rate and time of application of thifensulfuron-methyl for control of wild garlic. An additional objective was to determine if antagonism existed between tank-mixed metribuzin and thifensulfuron-methyl on control of wild garlic.

Materials And Methods

Rate and Time of Application

Investigations into the activity of thifensulfuron-methyl on wild garlic (Allium vineale L.) were conducted from 1986 through 1988. Field experiments were established in areas naturally infested with wild garlic to evaluate the activity of thifensulfuron-methyl rate and time of application for suppression of aerial bulblet production. Experiments were conducted at the Knoxville Plant Science Farm near Knoxville TN, and Plateau Experiment Station near Crossville, TN during 1986. Treatments included thifensulfuron-methyl at 9, 17.5, and 35 g ai ha^{-1}; 17.5 g ai ha^{-1} thifensulfuron-methyl plus 420 g ai ha^{-1} metribuzin; and 1120 g ai ha^{-1} 2,4-D ester.

Nonionic surfactant at 0.25% v/v was included with each treatment. Treatments were applied postemergence with dates and wild garlic heights being: Knoxville: March 24, 20 to 25 cm; April 10, 25 to 30 cm and April 23, 35 to 38 cm and Crossville: April 1, 20 cm; April 12, 31 cm and April 25, 36 cm. Treatments were applied with a CO_2 backpack sprayer calibrated to deliver 98 L ha^{-1} at 158 kPa. Plot size was 2.4 by 7.6 m. Experimental design was a randomized complete block with a factorial arrangement of treatments. Treatments were replicated three times.

Visual rating of suppression in aerial bulblets was conducted on June 6 at Knoxville and June 11 at Crossville. Ratings based on visual observations were used to evaluate herbicide performance. Percentage values of 0 (no suppression) to 100 (complete suppression) were used in comparing herbicide efficacy to an untreated check (Frans et al. 1986). On July 12 and July 17 aerial bulblets were collected and weighed from a randomly selected 1-m^2 area of each plot at Crossville and Knoxville. Data were subjected to analysis of variance and means were separated by Duncan's Multiple Range.

An experiment at the Dairy Experiment Station near Lewisburg, TN was conducted during 1987. Treatments included thifensulfuron-methyl at 4, 9, and 17.5 g ai ha^{-1}. An additional treatment of 1120 g ai ha^{-1} of 2,4-D ester was included. Treatments included 0.25% v/v nonionic surfactant.

Treatments were applied postemergence with application dates and wild garlic heights being: March 27, 31 to 38 cm, April 9, 38 to 43 cm, April 22, 51 to 61 cm. Experimental design was a randomized complete block with a factorial arrangement of treatments. Treatments were replicated three times.

Visual rating of suppression in aerial bulblets was made on June 2. Data were subjected to analysis of variance and means were separated by Duncan's Multiple Range.

An experiment was conducted at Ames Plantation near Grand Junction, TN from 1986 to 1988. Treatments were repeated annually on the same plots. Treatments included thifensulfuron-methyl at 4, 9, 17.5 and 35 g ai ha^{-1} and thifensulfuron-methyl 17.5 g ai ha^{-1} plus metribuzin 420 g ai ha^{-1}. An additional treatment included 1120 g ai ha^{-1} of 2,4-D ester. A nonionic surfactant at 0.25% v/v was included with each treatment. Treatments were applied postemergence with dates of application and wild garlic heights being: March 26, 1986, 20 to 25 cm; April 4, 1986, 25 to 31 cm; April 14, 1986, 41 to 43 cm; March 20, 1987, 20 to 25 cm; April 7,1987, 31 to 36 cm, April 21, 1987, 41 to 46 cm; March 24,1988, 15 to 25 cm; April 6, 1988, 25 to 31 cm and April 21, 1988, 41 to 46 cm. Experimental design was a randomized complete block with a factorial arrangement of treatments. Treatments were replicated four times.

Visual ratings of percent suppression in aerial bulblets were conducted on May 23, 1986; June 3, 1987; and May 19, 1988.

On July 30, 1986 a 1-m^2 random area was selected from the rear one-half of each plot. Underground bulbs were exhumed and weights recorded. In 1987 aerial bulblets were collected from three random 0.5-m^2 areas per plot and weights were recorded. Data were subjected to analysis of variance and means separated by Duncan's Multiple Range.

Antagonism

An experiment was conducted at Ames Plantation near Grand Junction, TN in 1987 and 1988 to evaluate thifensulfuron-methyl at 4, 9, 17.5 g ai ha^{-1} applied as a tank-mixture with metribuzin at 210, 420 and 630 g ai ha^{-1}. Plots received identical treatments in 1987 and 1988. All treatments received 0.25% v/v nonionic surfactant. Experimental design was a randomized complete block with a factorial arrangement of treatments replicated four times. Plot size was 2.4 by 7.6 m. Treatments were applied postemergence with a CO_2 backpack sprayer calibrated to deliver 98 L ha^{-1} at 158 kPa.

Visual estimates measuring suppression of aerial bulblets were made on April 21 and May 15 in 1987; April 21 and May 19 in 1988. Ten plants were selected randomly from each plot on July 15, 1987 and July 12, 1988. Underground bulbs were exhumed and weighed. On July 25, 1987 and July 12, 1988 three 0.5-m^2 areas were selected randomly from each plot. Aerial bulblets were collected and weighed. Data analysis was based on a model described by Flint et al. (1988).

Data were utilized to produce regression statistics in the form of regression estimates for treatment with an associated probability value. For visual ratings on level of suppression for aerial bulblets, a negative estimate and probability being met indicates a poor response to treatment and antagonism. A positive estimate and appropriate probability being met indicates a synergistic response. In measuring bulb and aerial bulblet weight, a positive estimate with appropriate probability indicates antagonism, while a negative estimate indicates synergism.

Sequential Application

Experiments were conducted at the Middle Tennessee Experiment Station near Spring Hill, TN during 1987 and 1988 to evaluate sequential application of thifensulfuron-methyl followed by metribuzin and metribuzin followed by thifensulfuron-methyl. Treatments included thifensulfuron-methyl at 4, 9, 17.5 g ai ha^{-1} followed by metribuzin 12 or 24 h later at 210, 420 and 630 g ai ha^{-1}. A second experiment included the previous rates of metribuzin and thifensulfuron-methyl, with thifensulfuron-methyl applied 12 or 24 h following metribuzin. All treatments received 0.25% v/v nonionic surfactant. Treatments were applied postemergence on March 25, 1987. On April 5, 1988, plots received identical treatments as in 1987. Wild garlic heights at application were 15 to 25 cm in 1987 and 15 to 23 cm in 1988.

Experimental design for both experiments was a randomized complete block with factorial arrangement of treatments replicated four times. Plot size was 2.4 by 7.6 m. Treatments were applied with a CO_2 backpack sprayer calibrated to deliver 98 L ha^{-1} at 158 kPa.

Visual estimates of suppression of aerial bulblets were made on April 19 and May 14, 1987 and May 19, 1988. In 1987 ten plants were selected at random from each plot, exhumed and underground bulb weights recorded. Aerial bulblets were collected from three 0.5-m^2 areas selected at random within each plot on July 7, 1987. Data were analyzed as mentioned for the antagonism section and graphed using regression analysis.

Greenhouse

Four dormant central bulbs of wild garlic were planted in 13-cm diameter by 8-cm tall plastic pots filled with a 1:1 mixture of Pro Mix[1] media and washed sand. Natural lighting was utilized in the greenhouse and temperature ranged from 21 C during day to 10 C at night. Bulbs were allowed to produce two 15-to 18-cm leaves before treating. Experiments evaluated the efficacy of thifensulfuron-methyl and antagonism of metribuzin on thifensulfuron-methyl activity. An experiment on efficacy included thifensulfuron-methyl at 4, 9, 17.5 and 35 g ai ha^{-1} plus 0.25% v/v nonionic surfactant.

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[1]Premier Brands, Inc. Standford, CT. 06902.

Treatments were applied postemergence with a CO_2 backpack sprayer calibrated to deliver 98 L ha^{-1} at 158 kPa. Thifensulfuron-methyl activity was evaluated 30 d following application. Experimental design was a randomized complete block with treatments replicated four times. The experiment was repeated.

The antagonism experiment included thifensulfuron-methyl at 9, 17.5, 35 g ai ha^{-1} alone and in combination with metribuzin at 420 g ai ha^{-1} and metribuzin alone at 420 g ai ha^{-1}. All treatments included 0.25% v/v nonionic surfactant. Experimental procedures were identical to the efficacy experiment.

Plants from each experiment were collected 30 d following treatment. Data included bulb condition, bulb weight, shoot weight, number of roots, root length and shoot height. Data were subjected to analysis of variance and means separated by Duncan's Multiple Range.

Phytotoxicity to Small Grains

Thifensulfuron-methyl phytotoxicity to small grains was evaluated at the University of Tennessee Small Grains Farm near Knoxville, TN. Wheat cultivars 'Coker 747', 'Caldwell', 'Pioneer 2550', and 'Fillmore' and barley cultivars 'Barsoy' and 'Volbar' were seeded in the fall of 1986. Thifensulfuron-methyl at 17.5, 35 and 53 g ai ha^{-1} plus 0.25% v/v nonionic surfactant were applied postemergence at three stages of small grain development.

Application times occurred at Feekes (Large, 1954) wheat growth stage 4 on March 25, stage 6 on April 20 and stage 10 on April 27. Plots were 0.91 by 7.6 m. Treatments were applied using a CO_2 backpack sprayer calibrated to deliver 98 L ha^{-1} at 158 kPa. Experimental design was a randomized complete block factorial arrangement of treatments replicated four times.

Grain yields were determined by harvesting plots on July 2, 1987. Data were subjected to analysis of variance and means separated by Duncan's Multiple Range.

Results And Discussion

Rate and Time of Application

Thifensulfuron-methyl suppressed aerial bulblets in 3 yr of evaluation (Tables 1,2,3,4,5,6,7). The greatest suppression of aerial bulblets was with 35 g ai ha^{-1} of thifensulfuron-methyl. Aerial bulblet suppression ranged from 98% for 35 g ai ha^{-1} applied on April 10 at Knoxville to 20% for 4 g ai ha^{-1} applied April 14 at Grand Junction (Tables 1 and 3). Suppression of aerial bulblets was greater with thifensulfuron-methyl than with 2,4-D at Knoxville in 1986 (Table 1). Thifensulfuron-methyl at 17.5 g ai ha^{-1} tank-mixed with metribuzin at 420 g ai ha^{-1} applied April 23 resulted in a 75% suppression of aerial bulblets when compared to the control (Table 1). Suppression of aerial bulblets was less with 2,4-D than with thifensulfuron-methyl at all three dates.

Table 1. Effect of thifensulfuron-methyl rate and date of application on percent suppression of wild garlic plants at Knoxville in 1986[a].

Treatment[b]	Herbicide rate	Suppression on June 6		
		Application Date		
		24 Mar	10 Apr	23 Apr
	g ai ha^{-1}	%		
Thifensulfuron	9	96 ab	90 bc	93 a
Thifensulfuron	17.5	95 ab	96 ab	95 a
Thifensulfuron	35	98 a	98 a	94 a
Thifensulfuron + Metribuzin	17.5 420	88 b	87 c	75 b
2,4-D	1120	77 c	78 d	67 b

[a] Means in a column followed by the same letter do not differ at 0.05 level according to Duncan's Multiple Range mean separation.
[b] Thifensulfuron and thifensulfuron-methyl are the same.

Table 2. Effect of thifensulfuron-methyl rate and date of application on percent suppression of wild garlic plants at Crossville in 1986[a].

Treatment[b]	Herbicide rate	Suppression on June 11		
		Application Date		
		1 Apr	12 Apr	25 Apr
	g ai ha^{-1}	%		
Thifensulfuron	9	99 a	99 a	94 a
Thifensulfuron	17.5	99 a	99 a	99 a
Thifensulfuron	35	96 a	99 a	99 a
2,4-D	1120	80 b	77 b	73 b

[a] Means in a column followed by same letter do not differ at 0.05 level according to Duncan's Multiple Range mean separation.
[b] Thifensulfuron and thifensulfuron-methyl are the same.

Table 3. Effect of thifensulfuron-methyl rate and date of application on percent suppression of wild garlic plants at Grand Junction in 1986[a].

Treatment[b]	Herbicide rate	Suppression on May 23		
		Application Date		
		26 Mar	4 Apr	14 Apr
	g ai ha^{-1}	%		
Thifensulfuron	4	81 b	33 a	20 b
Thifensulfuron	9	96 a	51 a	29 ab
Thifensulfuron	17.5	98 a	44 a	54 a
Thifensulfuron	35	98 a	48 a	53 a
Thifensulfuron + Metribuzin	17.5 420	93 ab	26 b	51 a
2,4-D	1120	43 c	46 a	29 ab

[a] Means in a column followed by same letter do not differ at 0.05 level according to Duncan's Multiple Range mean separation.
[b]Thifensulfuron and thifensulfuron-methyl are the same.

Table 4. Effect of thifensulfuron-methyl rate and date of application on percent suppression of wild garlic plants at Grand Junction in 1987[a].

Treatment[b]	Herbicide rate	Suppression on June 3		
		Application Date		
		20 Mar	7 Apr	21 Apr
	g ai ha^{-1}	%		
Thifensulfuron	4	60 a	56 b	59 a
Thifensulfuron	9	64 a	83 ab	66 a
Thifensulfuron	17.5	75 a	91 a	66 a
Thifensulfuron	35	81 a	88 a	60 a
Thifensulfuron + Metribuzin	17.5 420	60 a	80 ab	59 a
2,4-D	1120	56 a	69 ab	53 a

[a] Means in a column followed by same letter do not differ at 0.05 level according to Duncan's Multiple Range mean separation.
[b]Thifensulfuron and thifensulfuron-methyl are the same.

Table 5. Effect of thifensulfuron-methyl rate and date of application on percent suppression of wild garlic plants at Grand Junction in 1988[a].

Treatment[b]	Herbicide rate	Suppression on May 19		
		Application Date		
		24 Mar	6 Apr	21 Apr
	g ai ha-1	%		
Thifensulfuron	4	63 a	50 ab	80 a
Thifensulfuron	9	49 a	74 ab	59 ab
Thifensulfuron	17.5	63 a	91 a	86 a
Thifensulfuron	35	75 a	94 a	81 a
Thifensulfuron + Metribuzin	17.5 420	68 a	49 ab	68 ab
2,4-D	1120	78 a	88 a	58 ab

[a] Means in a column followed by same letter do not differ at 0.05 level according to Duncan's Multiple range mean separation.
[b]Thifensulfuron and thifensulfuron-methyl are the same.

Table 6. Effect of thifensulfuron-methyl rate and date of application on percent suppression of wild garlic plants at Lewisburg in 1987[a].

Treatment[b]	Herbicide rate	Suppression on June 2		
		Application Date		
		26 Mar	9 Apr	22 Apr
	g ai ha-1	%		
Thifensulfuron	4	72 b	80 b	78 b
Thifensulfuron	9	87 a	87 ab	88 a
Thifensulfuron	17.5	88 a	93 a	90 a
2,4-D	1120	65 b	63 c	65 c

[a] Means followed by the same letter do not differ at 0.05 level according to Duncan's Multiple Range mean separation.
[b]Thifensulfuron and thifensulfuron-methyl are the same.

Table 7. Effect of thifensulfuron-methyl rate and date of application on wild garlic aerial bulblet production at Knoxville in 1986[a].

Treatment[c]	Herbicide rate	Aerial Bulblet Weights[b] Application Date		
		24 Mar	10 Apr	23 Apr
	g ai ha^{-1}		g m^{-2}	
Thifensulfuron	9	2 b	2 b	15 ab
Thifensulfuron	17.5	2 b	1 b	3 b
Thifensulfuron	35	2 b	0 b	6 b
Thifensulfuron + Metribuzin	17.5 420	7 b	4 b	5 b
2,4-D	1120	9 ab	6 b	16 a
Control	____	34 a	32 a	29 a

[a] Means in a column followed by the same letter do not differ at the 0.05 level according to Duncan's Multiple Range mean separation.
[b] Aerial bulblets were collected on July 17, 1986. Bulblet weight was recorded following air drying for 30 d.
[c]Thifensulfuron and thifensulfuron-methyl are the same.

The addition of 420 g ai ha^{-1} of metribuzin to 17.5 g ai ha^{-1} thifensulfuron-methyl antagonized the latter's activity on aerial bulblets at Knoxville in 1986.

Thifensulfuron-methyl rates did not differ in suppression of aerial bulblets at Crossville in 1986 (Table 2). Thifensulfuron-methyl at 9 to 35 g ai ha^{-1} suppressed aerial bulblets more than 2,4-D regardless of date applied. Suppression ranged from 94% for 9 g ai ha^{-1} applied April 25 to 99% for 17.5 g ai ha^{-1} on all application dates.

Time and rate of thifensulfuron-methyl application determined the level of aerial bulblet suppression at Grand Junction in 1986 (Table 3). All rates suppressed aerial bulblets more on March 26 compared to April 4 or 14. Gast and Slife (1985) observed that thifensulfuron-methyl applied on April 9 was more effective than on April 29, especially at higher rates. Thifensulfuron-methyl at 9, 17.5, and 35 g ai ha^{-1} suppressed aerial bulblets from 96 to 98% and was significantly better than at 4 g ai ha^{-1} on March 26. Aerial bulblet suppression was very poor (<54%) for all thifensulfuron-methyl rates applied April 4 and April 14. Metribuzin at 420 g ai ha^{-1} tank-mixed with 17.5 g ai ha^{-1} thifensulfuron-methyl antagonized the latter's activity on aerial bulblet suppression on April 4, but not on March 26 and April 14. Aerial bulblet suppression was poor for 2,4-D compared to thifensulfuron-methyl alone and in combination with metribuzin.

During 1987 and 1988 at Grand Junction application of 17.5 and 35 g ai ha^{-1} of thifensulfuron-methyl suppressed aerial bulblets more than at 4 and 9 g ai ha^{-1} (Tables 4 and 5). Aerial bulblet suppression was 91% with 17.5 g ai ha^{-1} thifensulfuron-methyl applied April 7 (Table 4). Fair control was noted for thifensulfuron-methyl rates of 4 and 9 g ai ha^{-1} applied on March 20, April 7 and April 21 (Table 4). Even though suppression of aerial bulblets was variable, there were differences among thifensulfuron-methyl treatments. Antagonism was not apparent for thifensulfuron methyl at 17.5 g ai ha^{-1} tank-mixed with 420 g ai ha^{-1} metribuzin in 1987. Results during 1988 were similar to those from 1987 (Table 5). Aerial bulblet suppression ranged from a high of 75% with 35 g ai ha^{-1} of thifensulfuron-methyl to a low of 49% with 9 g ai ha^{-1} when applied March 24. Suppression of 91 and 94% was obtained from 17.5 g ai ha^{-1} and 35 g ai ha^{-1} thifensulfuron-methyl applied on April 6. Thifensulfuron-methyl at 4 g ai ha^{-1} on April 21 was comparable to 35 g ai ha^{-1} in suppressing aerial bulblets. Tank-mixing thifensulfuron-methyl at 17.5 g ai ha^{-1} with 420 g ai ha^{-1} of metribuzin did not result in antagonism.

Thifensulfuron-methyl at 9 and 17.5 g ai ha^{-1} applied on three dates performed better than 2,4-D in suppression of aerial bulblets at Lewisburg (Table 6). Thifensulfuron-methyl at 17.5 g ai ha^{-1} suppressed aerial bulblets 93 and 90% on April 9 and 22, respectively.

Aerial bulblets (g m^{-2}) were reduced by all rates of thifensulfuron-methyl applied on March 24 or April 10 compared to the untreated control (Table 7). Thifensulfuron-methyl at 9 g ai ha^{-1} applied April 23 was not as effective in reducing aerial bulblets as on March 24 and April 10. Thifensulfuron-methyl at 17.5 g ai ha^{-1} tank-mixed with 420 g ai ha^{-1} metribuzin did not indicate antagonized activity on aerial bulblets.

Reduction of aerial bulblets was greatest for all application dates with 35 g ai ha^{-1} thifensulfuron-methyl at Grand Junction in 1988 (Table 8). Mean weights for aerial bulblets were 0 to 2 g per 1.5 m^2 on April 7 and April 21, respectively. Aerial bulblet weights were 22 and 27 g per 1.5 m^2 respectively for 4 and 9 g ai ha^{-1} of thifensulfuron-methyl applied on March 20 compared to 24 g per 1.5 m^2 in the untreated control. Leys and Slife (1987), evaluating chlorsulfuron and metsulfuron, found that 60 g ai ha^{-1} applied in late March eliminated aerial bulblet production. Chlorsulfuron at 20 g ai ha^{-1} in the spring (April 17) reduced aerial bulblets by 99% compared to 59% for fall application.

Antagonism of thifensulfuron-methyl tank-mixed with metribuzin was not evident. Reduction of aerial bulblets was not different for the tank-mixture compared to 35 g ai ha^{-1} thifensulfuron-methyl alone.

Table 8. Effect of thifensulfuron-methyl rate and date of application on reduction in wild garlic aerial bulblets at Grand Junction in 1987[a]

Treatment[c]	Herbicide rate	Aerial Bulblet Weights[b]		
		Application Date		
		20 Mar	7 Apr	21 Apr
	g ai ha^{-1}	g per 1.5 m^2		
Thifensulfuron	4	22 ab	11 b	10 b
Thifensulfuron	9	27 ab	5 b	2 b
Thifensulfuron	17.5	6 a	0 b	3 b
Thifensulfuron	35	1 b	0 b	2 b
Thifensulfuron + Metribuzin	17.5 420	7 ab	2 b	7 b
2,4-D	1120	12 ab	2 b	20 ab
Control	____	24 ab	26 a	30 a

[a]Means in a column followed by the same letter do not differ at 0.05 level according to Duncan's Multiple Range mean separation.
[b]Aerial bublets collected on June 24-25, 1987. Weights recorded after air drying 30 d.
[c]Thifensulfuron and thifensulfuron-methyl are the same.

Reduction of underground bulbs was greatest with 35 g ai ha^{-1} of thifensulfuron-methyl applied on March 26, but did not differ from 4 and 9 g ai ha^{-1} when applied on April 4 or 9 (Table 9). The lack of consistency in reduction of underground bulbs may have resulted from how the bulbs were collected. Separation of different bulb types was not conducted during collection. Bulb weights represent all bulb types from the exhumed area. Thifensulfuron-methyl had little effect on underground bulb production when applied on April 4 or 14.

Antagonism

During 1987 and 1988 tank-mixing metribuzin with thifensulfuron-methyl lead to antagonism of the latters activity on wild garlic aerial bulblet reduction (Tables 10 and 11). In 1987 activity of thifensulfuron methyl at 4 and 9 g ai ha^{-1} was antagonized by metribuzin at 210, 420, and 630 g ai ha^{-1} at 39 DAT (days after treatment) (Table 10).

As discussed in materials and methods, analysis of the data is based on a model by Flint et al. (1988). Rating data with a negative regression estimate for the herbicide combination and probability level of $P<0.05$ indicates antagonism on aerial bulblet suppression. If the sign is positive for the herbicide combination and probability level is met, the response is synergistic on aerial bulblet suppression.

Table 9. Effect of thifensulfuron-methyl rate and date of application on reduction of wild garlic underground bulb production at Grand Junction in 1986[a].

Treatment[c]	Herbicide rate	Underground Bulb Weight[b] Application Date		
		26 Mar	4 Apr	14 Apr
	g ai ha^{-1}		g m^2	
Thifensulfuron	4	50 ab	66 ab	77 a
Thifensulfuron	9	40 b	48 ab	83 a
Thifensulfuron	17.5	33 b	99 a	77 a
Thifensulfuron	35	30 b	83 ab	68 a
Control	____	81 a	59 ab	63 a

[a]Means in a column followed by same letter do not differ at 0.05 level according to Duncan's Multiple Range mean separation.
[b]Underground bulbs exhumed on July 30, 1986. Weights were recorded following washing and air drying.
[c]Thifensulfuron and thifensulfuron-methyl are the same.

Table 10. Effect of thifensulfuron-methyl and metribuzin alone and tank-mixed on aerial bulblet reduction at Grand Junction 1987[a].

Treatment[c]	Herbicide rate	Level of Reduction			
		39 DAT[b]		63 DAT[b]	
		Regression Estimate	P>|T|	Regression Estimate	P>|T|
	g ai ha^{-1}				
Intercept (control)	0	0.0000	1.0000	0.0000	1.0000
Thifensulfuron	4	0.5308	0.0001	0.2438	0.0137
Thifensulfuron	9	0.9336	0.0001	0.8117	0.0001
Thifensulfuron	17.5	0.7554	0.0001	0.8145	0.0001
Metribuzin	210	0.0125	0.8556	0.0625	0.5135
Metribuzin	420	0.0375	0.5853	0.0250	0.7936
Metribuzin	630	0.0125	0.8556	0.0501	0.6003
Thifensulfuron + Metribuzin	4 210	-0.4681	0.0001	-0.1504	0.2688
Thifensulfuron + Metribuzin	4 420	-0.5308	0.0001	-0.2563	0.0629
Thifensulfuron + Metribuzin	4 630	-0.5308	0.0001	-0.2438	0.0763
Thifensulfuron + Metribuzin	9 210	-0.9211	0.0001	-0.7072	0.0001
Thifensulfuron + Metribuzin	9 420	-0.9336	0.0001	-0.7990	0.0001
Thifensulfuron + Metribuzin	9 630	-0.8836	0.0001	-0.8118	0.0001
Thifensulfuron + Metribuzin	17.5 210	0.0556	0.5672	0.0210	0.8763
Thifensulfuron + Metribuzin	17.5 420	0.0583	0.5488	0.0830	0.5399
Thifensulfuron + Metribuzin	17.5 630	0.2060	0.0384	-0.0303	0.8224

[a]P<0.05 indicates significance and antagonism where thifensulfuron-methyl is tank-mixed with metribuzin.
[b]days after treatment
[c]Thifensulfuron and thifensulfuron-methyl are the same.

Table 11. Effect of thifensulfuron-methyl and metribuzin alone and tank-mixed on aerial bulblet reduction at Grand Junction in 1988[a].

Treatment[c]	Herbicide rate	Level of Reduction			
		28 DAT[b]		56 DAT[b]	
		Regression Estimate	P>|T|	Regression Estimate	P>|T|
	g ai ha^{-1}				
Intercept (control)	0	0.0000	1.0000	0.0000	1.0000
Thifensulfuron	4	0.1639	0.0422	0.4093	0.0030
Thifensulfuron	9	0.2976	0.0004	0.7883	0.0001
Thifensulfuron	17.5	0.2868	0.0007	1.1271	0.0001
Metribuzin	210	0.1007	0.2055	0.1135	0.3883
Metribuzin	420	0.0751	0.3429	0.0628	0.6320
Metribuzin	630	0.0500	0.5265	0.0628	0.6320
Thifensulfuron + Metribuzin	4 210	-0.2019	0.0752	-0.3686	0.0515
Thifensulfuron + Metribuzin	4 420	-0.1512	0.1793	-0.3845	0.0426
Thifensulfuron + Metribuzin	4 630	-0.1263	0.2604	-0.3970	0.0366
Thifensulfuron + Metribuzin	9 210	-0.2598	0.0236	-0.3842	0.0428
Thifensulfuron + Metribuzin	9 420	-0.1956	0.0845	-0.5940	0.0024
Thifensulfuron + Metribuzin	9 630	-0.2726	0.0179	-0.6324	0.0013
Thifensulfuron + Metribuzin	17.5 210	-0.0500	0.6539	-0.3780	0.0461
Thifensulfuron + Metribuzin	17.5 420	-0.5622	0.6146	-0.3728	0.0490
Thifensulfuron + Metribuzin	17.5 630	-0.0712	0.5237	-0.3334	0.0771

[a]P<0.05 indicates significance and antagonism where thifensulfuron-methyl is tank-mixed with metribuzin.
[b]days after treatment
[c]Thifensulfuron and thifensulfuron-methyl are the same.

A negative regression estimate and probability level of significance at $P<0.05$ indicates a reduction in aerial bulblet and underground bulb weight while a positive regression estimate indicates a synergistic response.

When metribuzin at 210, 420 and 630 g ai ha^{-1} were tank-mixed with 17.5 g ai ha^{-1} thifensulfuron-methyl antagonism did not occur. Thifensulfuron-methyl at 17.5 g ai ha^{-1} tank-mixed with metribuzin at 620 g ai ha^{-1} proved to be synergistic in aerial bulblet suppression. Thifensulfuron-methyl at 9 g ai ha^{-1} tank-mixed with metribuzin at 210, 420 and 630 g ai ha^{-1} antagonized aerial bulblet suppression. Thifensulfuron-methyl at 4, 9 and 17.5 g ai ha^{-1} suppressed aerial bulblets for 63 DAT.

During 1988 all tank-mixed treatments of thifensulfuron-methyl and metribuzin resulted in a negative regression estimate at 28 DAT (Table 11). Tank-mixed treatments containing 9.0 g ai ha^{-1} thifensulfuron-methyl and metribuzin at 210 and 630 g ai ha^{-1} were antagonistic at 28 DAT. At 56 DAT, antagonism occurred for all tank-mixed treatments of thifensulfuron-methyl and metribuzin except for thifensulfuron-methyl at 17.5 g ai ha^{-1} and metribuzin at 630 g ai ha^{-1}.

Reduction of underground bulbs and aerial bulblets was antagonized by tank-mixing metribuzin with thifensulfuron-methyl (Tables 12 and 13).

Table 12. Effect of thifensulfuron-methyl and metribuzin alone and tank-mixed on underground bulbs and aerial bulblets at Grand Junction in 1987[a].

Treatment[b]	Herbicide rate	Underground Bulbs			Aerial Bulblets		
		Regression Estimate	Expected Bulb Weight	P>|T|	Regression Estimate	Expected Bulblet Weight	P>|T|
	g ai ha^{-1}		g			g	
Intercept (control)	0	20.6200	20.6	0.0001	27.7700	27.8	0.0005
Thifensulfuron	4	-6.1200	14.5	0.1252	12.4125	40.1	0.2447
Thifensulfuron	9	-10.2375	10.4	0.0121	-3.3500	24.4	0.7519
Thifensulfuron	17.5	-11.2500	9.4	0.0062	-14.8950	12.9	0.1641
Metribuzin	210	-4.1225	16.5	0.2982	0.7575	28.5	0.9430
Metribuzin	420	-3.5600	17.0	0.3682	18.3075	46.1	0.0890
Metribuzin	630	-6.6400	13.9	0.0969	12.5050	40.3	0.2413
Thifensulfuron + Metribuzin	4 210	6.5350	27.1	0.2443	-12.2450	15.5	0.4153
Thifensulfuron + Metribuzin	4 420	10.5875	31.2	0.0623	2.9750	30.7	0.8426
Thifensulfuron + Metribuzin	4 630	15.1575	35.8	0.0089	-0.2150	27.5	0.9885
Thifensulfuron + Metribuzin	9 210	12.4800	33.1	0.0292	7.8800	19.9	0.5993
Thifensulfuron + Metribuzin	9 420	13.6925	34.3	0.0173	7.3225	20.4	0.6253
Thifensulfuron + Metribuzin	9 630	13.2500	33.9	0.0210	-9.9000	17.9	0.5096
Thifensulfuron + Metribuzin	17.5 210	5.8450	26.5	0.2970	9.1650	36.9	0.5414
Thifensulfuron + Metribuzin	17.5 420	2.4175	23.0	0.6646	-22.7875	4.9	0.1330
Thifensulfuron + Metribuzin	17.5 630	8.4675	29.1	0.1333	-19.9550	7.8	0.1870

[a]$P<0.05$ indicates significance and antagonism where thifensulfuron-methyl is tank-mixed with metribuzin.
[b]Thifensulfuron and thifensulfuron-methyl are the same

Table 13. Effect of thifensulfuron-methyl and metribuzin alone and tank-mixed on underground bulbs and aerial bulblets at Grand Junction in 1988[a].

Treatment[b]	Herbicide rate	Underground Bulbs			Aerial Bulblets		
		Regression Estimate	Expected Bulb Weight	P>|T|	Regression Estimate	Expected Bulblet Weight	P>|T|
	g ai ha^{-1}		g			g	
Intercept (control	0	27.2675	27.3	0.0001	24.8875	24.8	0.0001
Thifensulfuron	4	-15.0975	12.2	0.0001	8.1850	33.1	0.3246
Thifensulfuron	9	-17.9450	9.3	0.0001	-8.6750	16.2	0.2968
Thifensulfuron	17.5	-20.4550	6.8	0.0001	-16.1550	8.7	0.0555
Metribuzin	210	-10.8875	16.4	0.0028	-0.7150	24.2	0.9311
Metribuzin	420	-9.3650	17.9	0.0092	15.5200	40.4	0.0654
Metribuzin	630	-12.1600	15.1	0.0010	12.1075	37.0	0.1477
Thifensulfuron + Metribuzin	4 210	15.4505	42.7	0.0027	-8.6625	16.2	0.4600
Thifensulfuron + Metribuzin	4 420	19.2425	46.3	0.0003	5.5275	30.4	0.6367
Thifensulfuron + Metribuzin	4 630	21.2250	48.5	0.0001	-8.9175	15.9	0.4470
Thifensulfuron + Metribuzin	9 210	20.9625	48.2	0.0001	12.1425	37.0	0.3018
Thifensulfuron + Metribuzin	9 420	19.5825	46.8	0.0002	13.0600	37.9	0.2671
Thifensulfuron + Metribuzin	9 630	19.9550	47.2	0.0002	-6.9250	17.9	0.5543
Thifensulfuron + Metribuzin	17.5 210	13.0500	40.3	0.0102	3.3875	28.3	0.7721
Thifensulfuron + Metribuzin	17.5 420	12.6625	39.9	0.0125	-18.8975	6.0	0.1110
Thifensulfuron + Metribuzin	17.5 630	15.0875	42.3	0.0033	15.3625	9.5	0.1930

[a]P<0.05 indicates significance and antagonism where thifensulfuron-methyl is tank-mixed with metribuzin.
[b]Thifensulfuron and thifensulfuron-methyl are the same.

Expected bulb or bulblet weight was calculated by taking the regression estimate for the intercept and either adding or subtracting the estimate for treatment. A positive regression estimate and probability of $P<0.05$ indicates antagonism with increased bulb or bulblet weight for treatment. A negative regression estimate indicates synergism when decreased bulb or bulblet weight occurred. Treatments containing only thifensulfuron-methyl increased control of underground bulbs and aerial bulblets in 1987 and 1988. During 1987 expected underground bulb weight ranged from 14.5 to 9.4 g for 4 and 17.5 g ai ha^{-1} thifensulfuron-methyl, respectively (Table 12). A similiar response was apparent for aerial bulblets. Antagonism of thifensulfuron-methyl as evidenced by increased bulb weight was significant for treatments containing 4 g thifensulfuron-methyl plus 630 g ai ha^{-1} metribuzin and 9 g ai ha^{-1} thifensulfuron-methyl plus metribuzin at 210 and 630 g ai ha^{-1}.

No antagonism on aerial bulblets occurred from tank-mixing thifensulfuron-methyl with metribuzin (Table 12). It is apparent that thifensulfuron-methyl tank-mixed with metribuzin reduced aerial bulblet weight more than thifensulfuron-methyl alone. It is possible that the 17.5 g ai ha^{-1} rate of thifensulfuron-methyl tank-mixed with metribuzin produced a synergistic response on aerial bulblet reduction. Metribuzin has been evaluated solely for wild garlic aerial bulblet reduction (York and Slife 1977). Reduction in aerial bulblet production was 50% of control

when metribuzin was applied at 560 g ai ha^{-1} to test plots in 1976. In greenhouse experiments control of 7-wk old wild garlic plants was 50 and 83% for metribuzin applied at 560 and 840 g ai ha^{-1}, respectively.

Thifensulfuron-methyl at all rates reduced underground bulb production in 1988 (Table 13). Aerial bulblets were reduced to 8.7 g per 1.5 m^2 at the 17.5 g ai ha^{-1} rate. Lower rates of 4 and 9 g ai ha^{-1} thifensulfuron-methyl did not reduce bulblet weight in 1988. The apparent increase in bulblet weight both years for thifensulfuron-methyl at 4 g ai ha^{-1} compared to the control is difficult to explain. Controlling some of the other weed competition in these plots by thifensulfuron-methyl at the 4 g ai ha^{-1} is a possible explanation.

A significant antagonistic effect on underground bulb weights occurred at all rates of thifensulfuron-methyl tank-mixed with all rates of metribuzin in 1988. There was no antagonistic response for tank-mixed treatments of thifensulfuron-methyl and metribuzin on aerial bulblet production in 1988. Thifensulfuron-methyl at 17.5 g ai ha^{-1} appeared to overcome the antagonism of metribuzin at 420 and 630 g ai ha^{-1} by reducing aerial bulblets more than the other tank-mix combinations. This trend was also noted in 1987 (Table 12).

The reason thifensulfuron-methyl activity on wild garlic is antagonized by metribuzin is not known.

Metribuzin is a photosynthetic inhibitor and might cause phytotoxicity that could interfere with absorption and translocation of thifensulfuron-methyl. Metribuzin suppressed underground bulb development in 1987 and 1988, but not aerial bulblets (Tables 12 and 13). It is possible to conclude that metribuzin could be utilized to reduce wild garlic infestations where carryover to a succeeding crop would not be harmful. One reference (York and Slife 1977) noted that postemergence metribuzin at 560 g ai ha^{-1} controlled 50% of the wild garlic in a natural population.

Loss of herbicidal activity (antagonism) when two or more herbicides are mixed together has been reported. (Colby et al. 1965; Hall et al. 1982; Hatizos and Penner 1985; O'Donovan and Sullivan, 1982; O'Donovan et al. 1983; O'Sullivan and Vanden Born 1978; Prendeville et al. 1969; Todd and Stobbe 1980). Antagonism can be due to several factors. These could include alterations in metabolism of one herbicide caused by a second or changes in plant metabolism by one herbicide adversly affecting the phytotoxicity of another herbicide (O'Donovan et al. 1983; Todd and Stobbe 1980). When thiameturon (thifensulfuron) was tank-mixed with diclofop ((+-)-2-[4-(2,4-dichlorophenoxy) phenoxy] propanoic acid), antagonism of diclofop activity on wild oats was observed (Eberlein et al. 1988). Use of a formulation blank for thiameturon added to diclofop spray solution did not reduce wild oat control compared to diclofop alone.

It was concluded that thiameturon itself was responsible for reducing diclofop activity on wild oats. It is possible that the active ingredient in metribuzin reduces thifensulfuron-methyl activity in the underground bulb, but does not in aerial bulblets. Metribuzin causes leaf tissue to become necrotic which could reduce photosynthetate production and phloem loading to the sink ultimately resulting in decreased thifensulfuron-methyl activity on underground bulbs.

Sequential Application

Thifensulfuron-methyl followed by metribuzin at 12 h in 1987 did not antagonize thifensulfuron-methyl (Figure 1). Reduction of wild garlic growth was greater than in the control at 50 DAT for all rates of thifensulfuron-methyl plus metribuzin (Figure 2). There was no significant difference among treatments of thifensulfuron-methyl followed by metribuzin at 24 h in reduction of wild garlic growth at 25 DAT (Figure 3). Thifensulfuron-methyl at 9 and 17.5 g ai ha^{-1} followed at 24 h by metribuzin at 210, 420 and 630 g ai ha^{-1} reduced wild garlic growth (Figure 4).

No significant reduction in aerial bulblet weight with thifensulfuron-methyl followed by metribuzin at 12 h (Figure 5). Thifensulfuron-methyl at 4, 9 and 17.5 g ai ha^{-1} reduced aerial bulblet weight when followed at 24 h by metribuzin at 210, 420 and 630 g ai ha^{-1} (Figure 6).

Reduction of underground bulbs by thifensulfuron-methyl was not antagonized by metribuzin at 12 or 24 h (Figures 7 and 8).

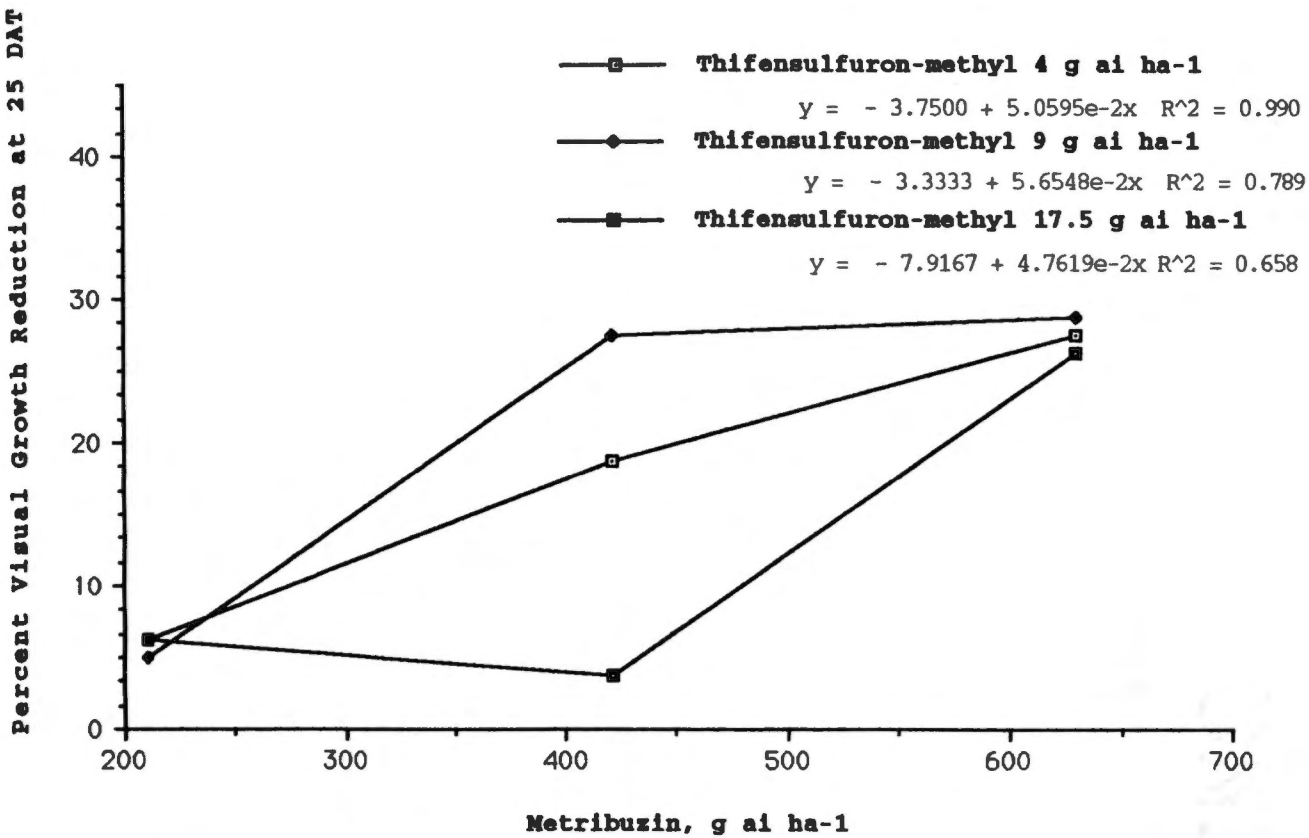

Figure 1. Percent reduction of wild garlic growth at 25 DAT from thifensulfuron-methyl followed at 12 h by metribuzin at Spring Hill in 1987.

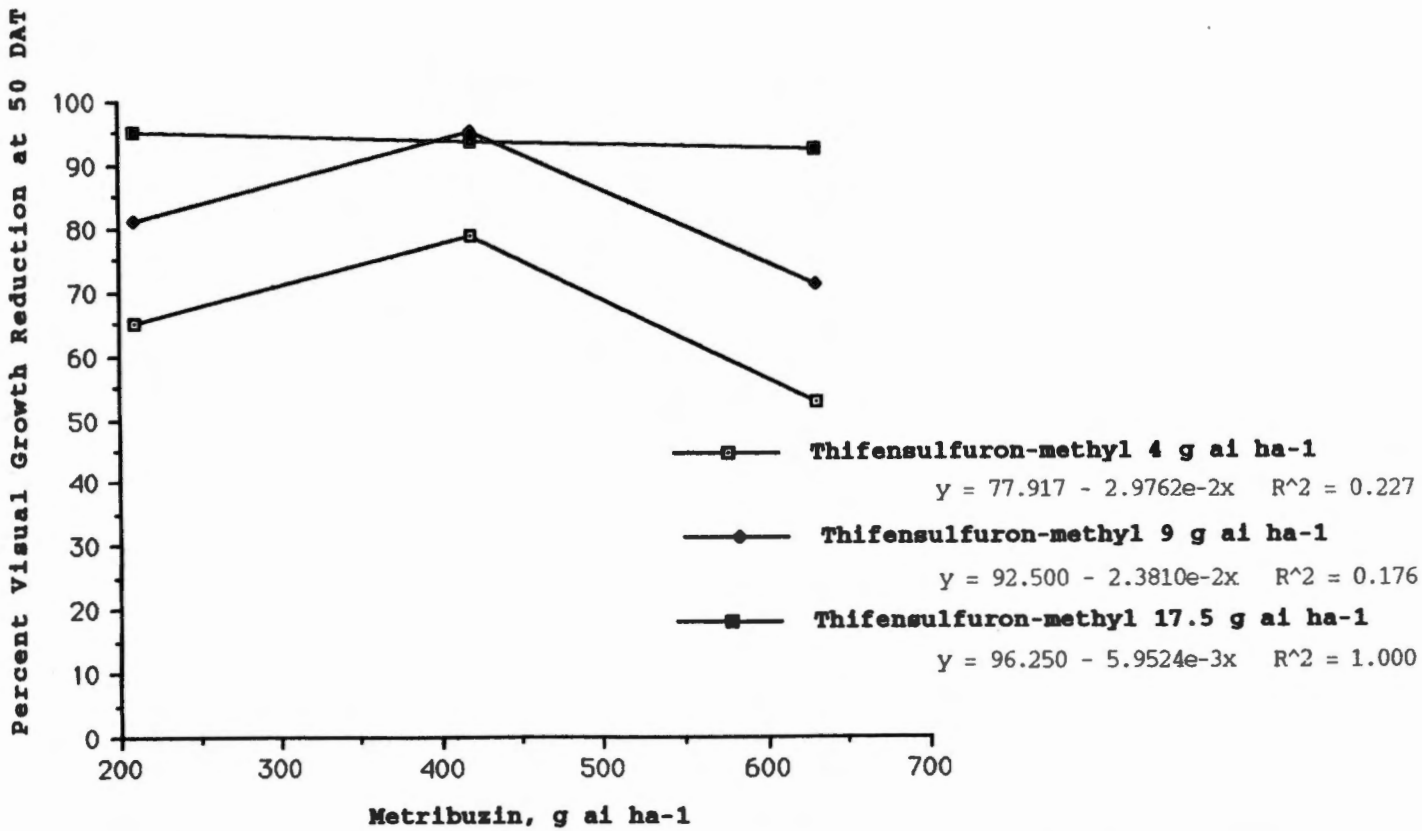

Figure 2. Percent reduction of wild garlic growth at 50 DAT from thifensulfuron-methyl followed at 12 h by metribuzin at Spring Hill in 1987.

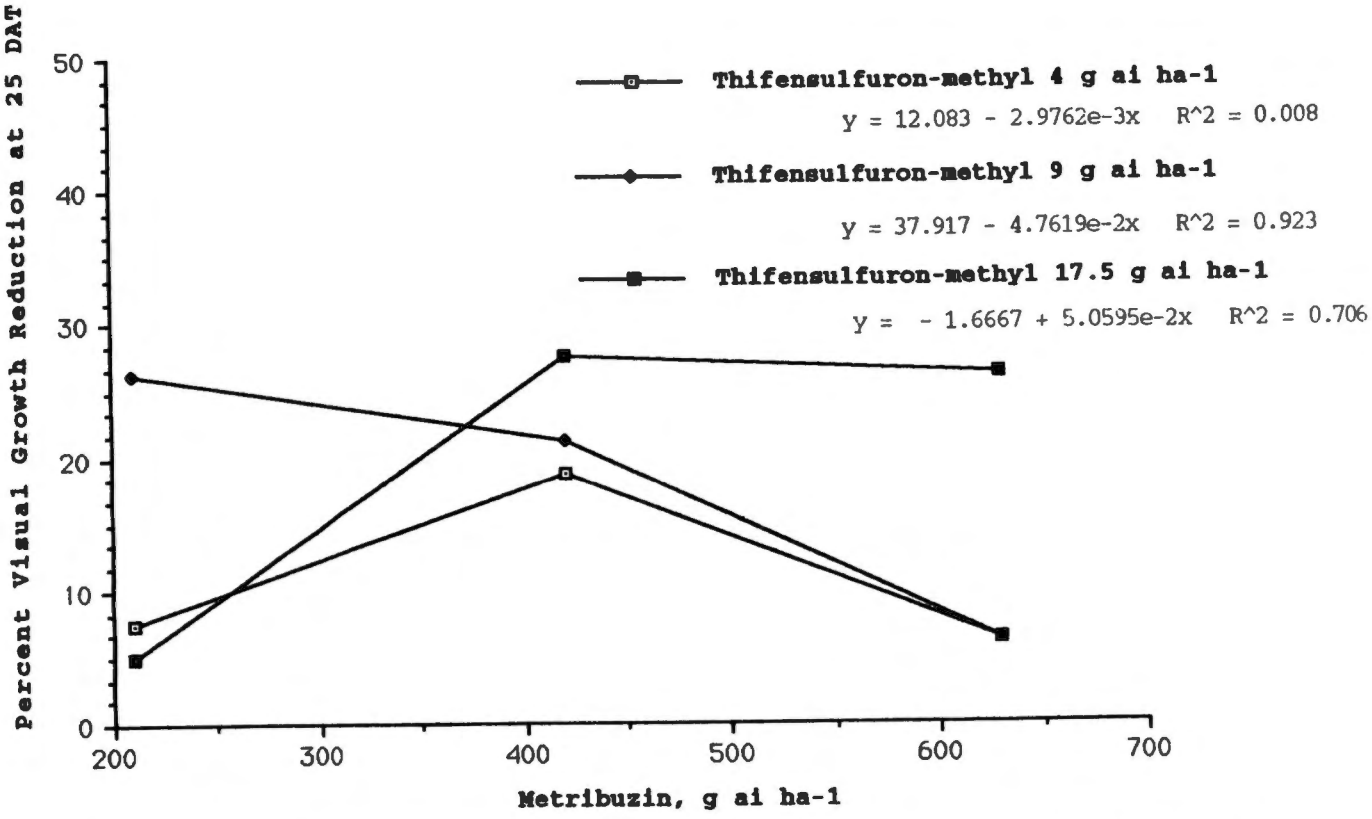

Figure 3. Percent reduction of wild garlic growth at 25 DAT from thifensulfuron-methyl followed at 24 h by metribuzin at Spring Hill in 1987.

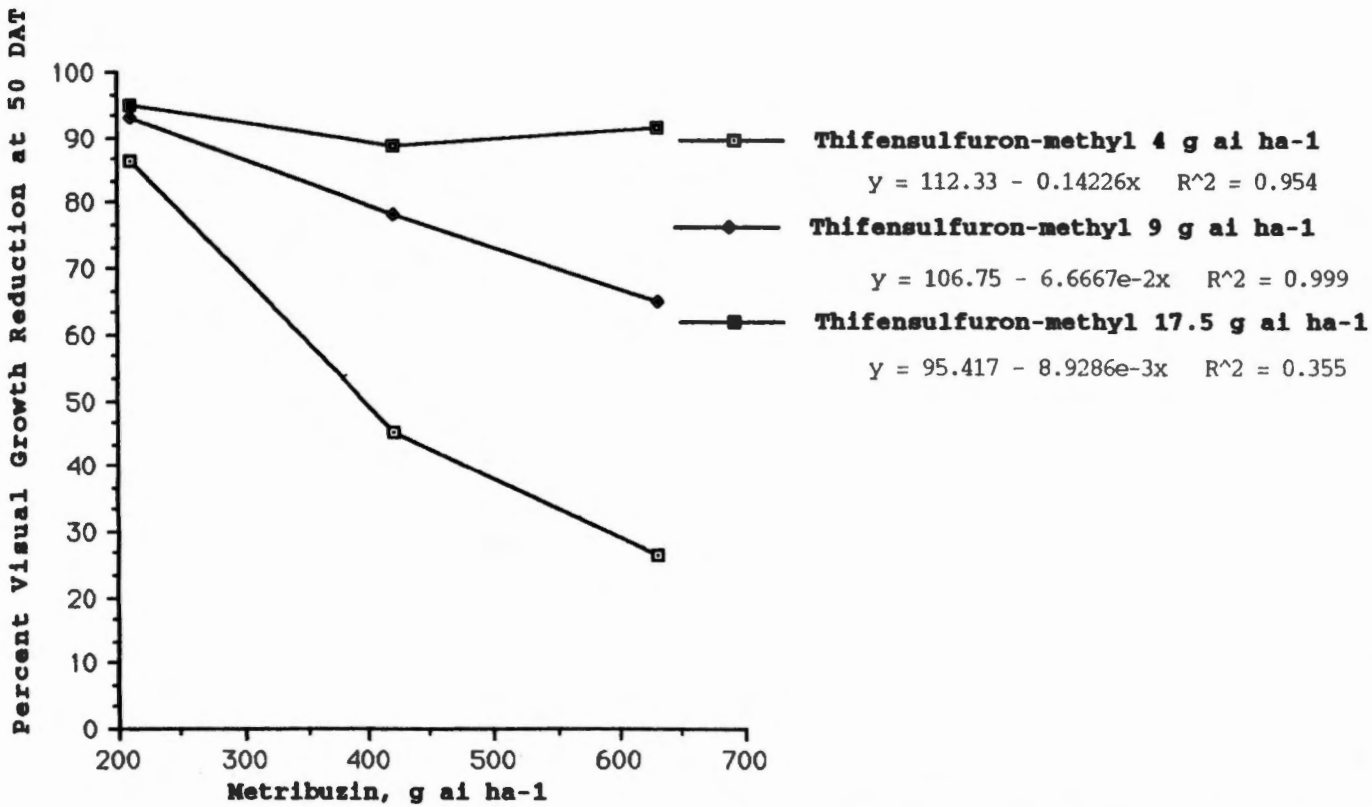

Figure 4. Percent reduction of wild garlic growth at 50 DAT from thifensulfuron-methyl followed at 24 h by metribuzin at Spring Hill in 1987.

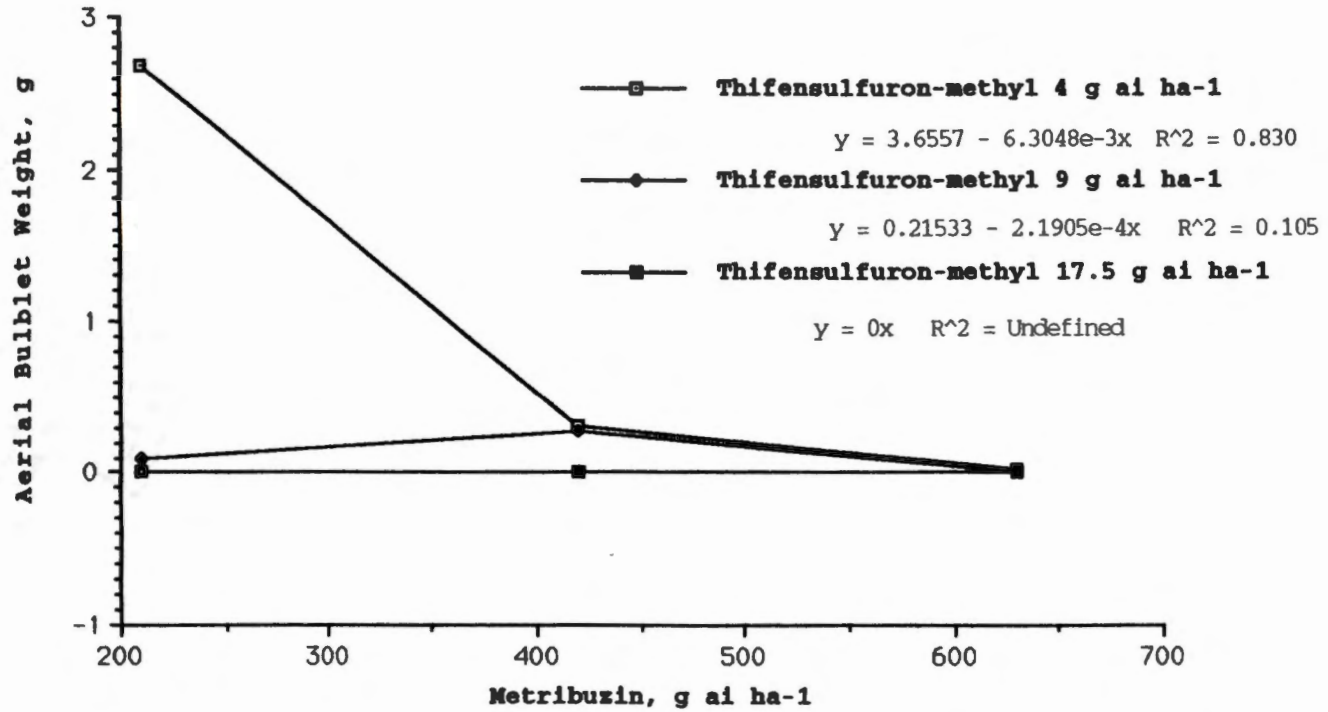

Figure 5. Reduction of wild garlic aerial bulblet weight from thifensulfuron-methyl followed at 12 h by metribuzin at Spring Hill in 1987.

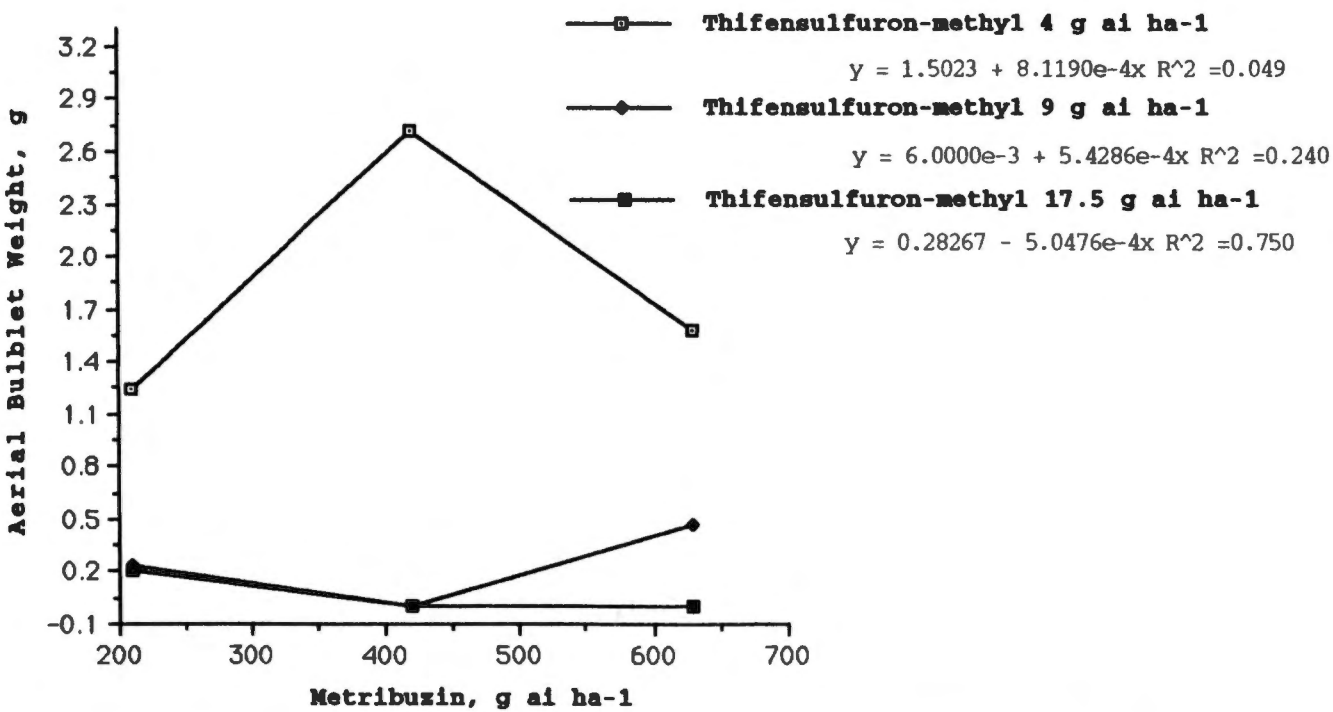

Figure 6. Reduction of wild garlic aerial bulblet weight from thifensulfuron-methyl followed at 24 h by metribuzin at Spring Hill in 1987.

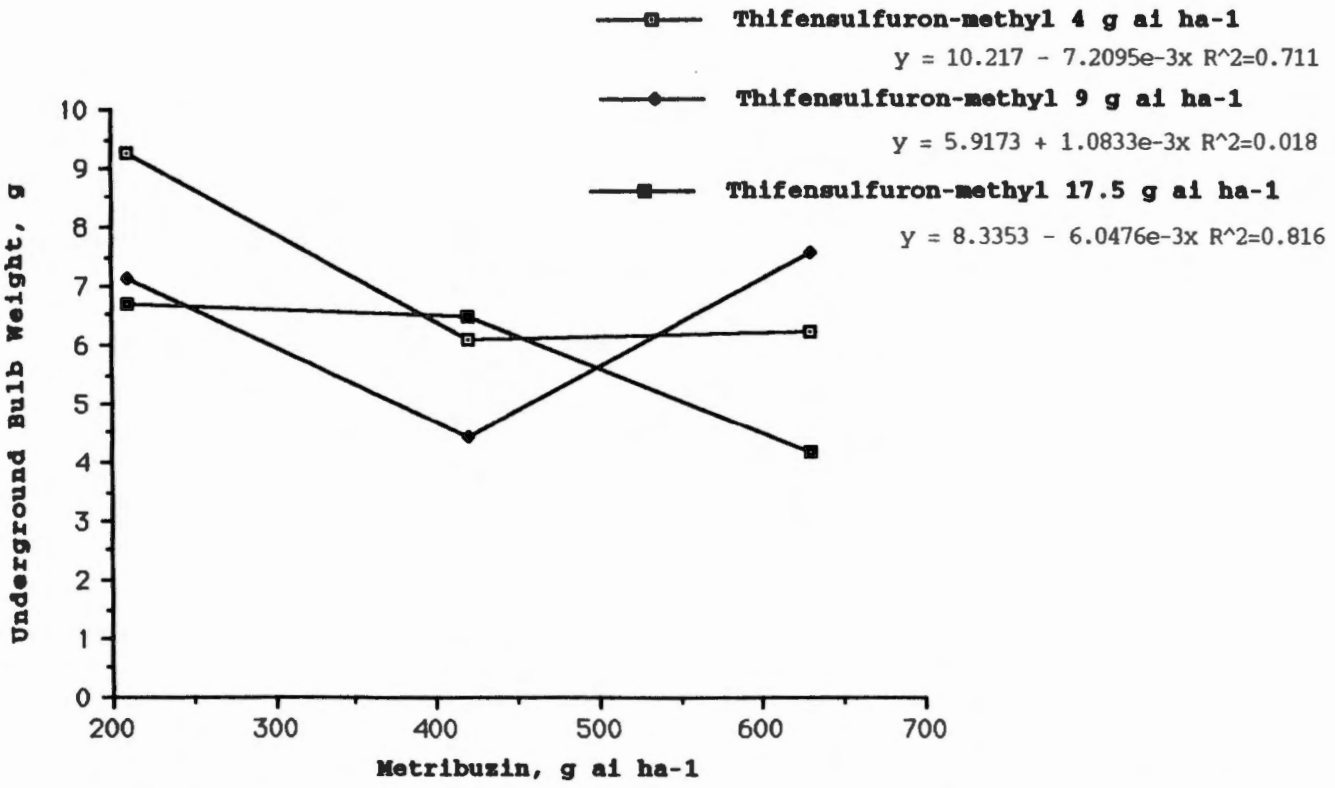

Figure 7. Reduction of wild garlic underground bulb weight from thifensulfuron-methyl followed at 12 h by metribuzin at Spring Hill in 1987.

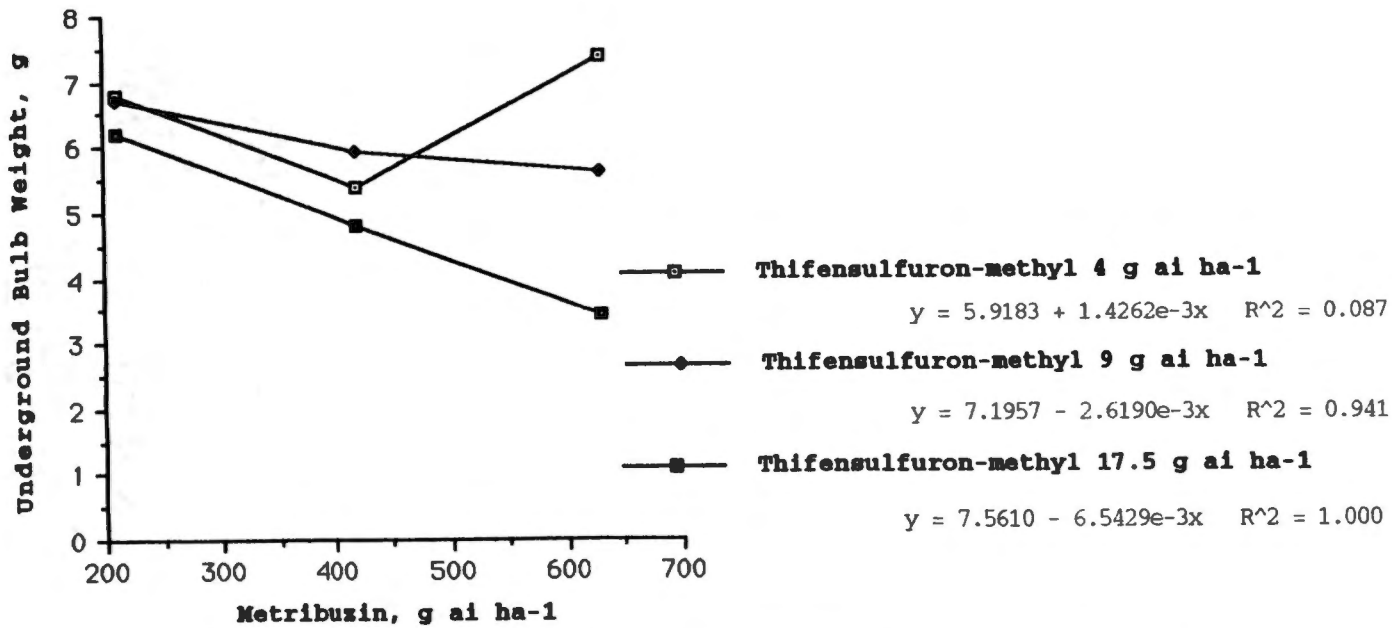

Figure 8. Reduction of wild garlic underground bulb weight from thifensulfuron-methyl followed at 24 h by metribuzin at Spring Hill in 1987.

The apparent antagonism at the lower rates of each herbicide is a result of the lack of activity on underground bulbs.

Control of wild garlic plants with thifensulfuron-methyl followed by metribuzin at 12 and 24 h was not affected (Figures 9 and 10). When thifensulfuron-methyl was applied 12 or 24 h after metribuzin, wild garlic control was not affected (Figures 11,12,13,14). All treatments resulted in whole plant reduction at 25 and 50 DAT. Similar results were obtained in 1988 (Figures 15 and 16).

Thifensulfuron-methyl activity on aerial bulblets was not affected when applied 12 or 24 h after metribuzin (Figures 17 and 18). All rates of thifensulfuron-methyl inhibited aerial bulblet production.

Thifensulfuron-methyl activity when applied 12 h after metribuzin was not antagonized (Figures 19 and 20). Increases in underground bulb weights occurred with the lower rates of thifensulfuron-methyl preceded by 630 g ai ha^{-1} metribuzin. Antagonism of thifensulfuron-methyl activity on underground bulbs occurred at 420 g ai ha^{-1} metribuzin followed by 9 g ai ha^{-1} thifensulfuron-methyl at 24 h. This treatment had 8.5 g per 1.5 m^2 of underground bulbs compared to 4.0 g per 1.5 m^2 in the control.

Greenhouse

Wild garlic bulb quality, shoot weight, roots per bulb and root and shoot length were reduced by thifensulfuron-methyl (Table 14).

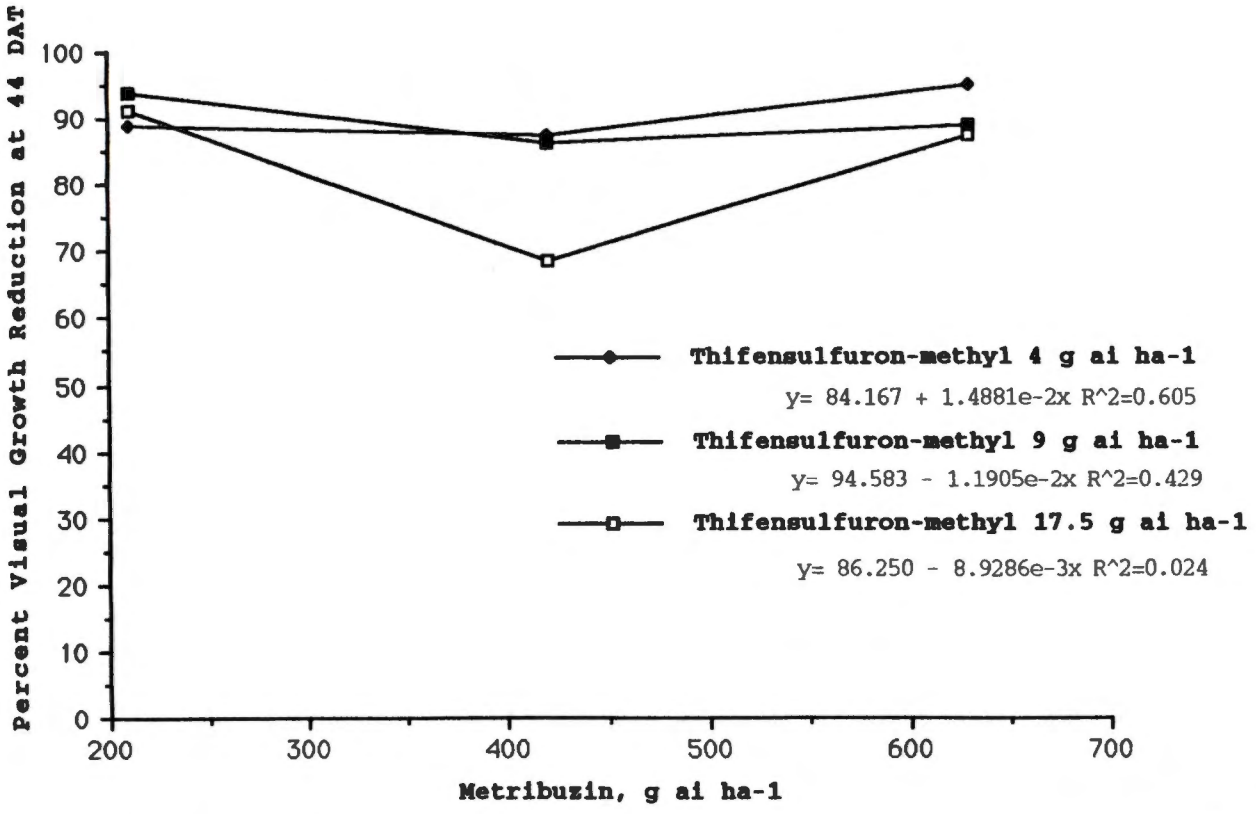

Figure 9. Percent reduction of wild garlic growth at 44 DAT from thifensulfuron-methyl followed at 12 h by metribuzin at Spring Hill in 1988.

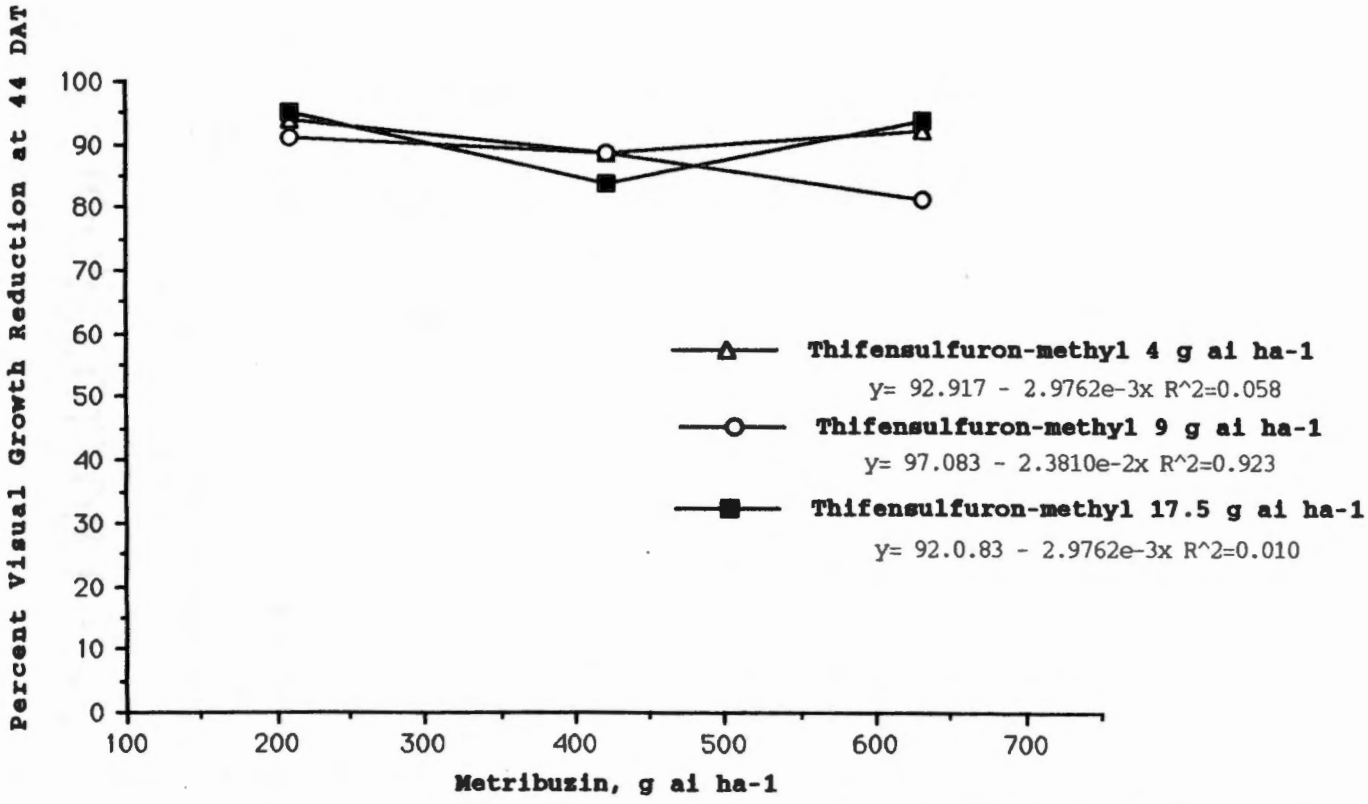

Figure 10. Percent reduction of wild garlic growth at 44 DAT from thifensulfuron-methyl followed at 24 h by metribuzin at Spring Hill in 1988.

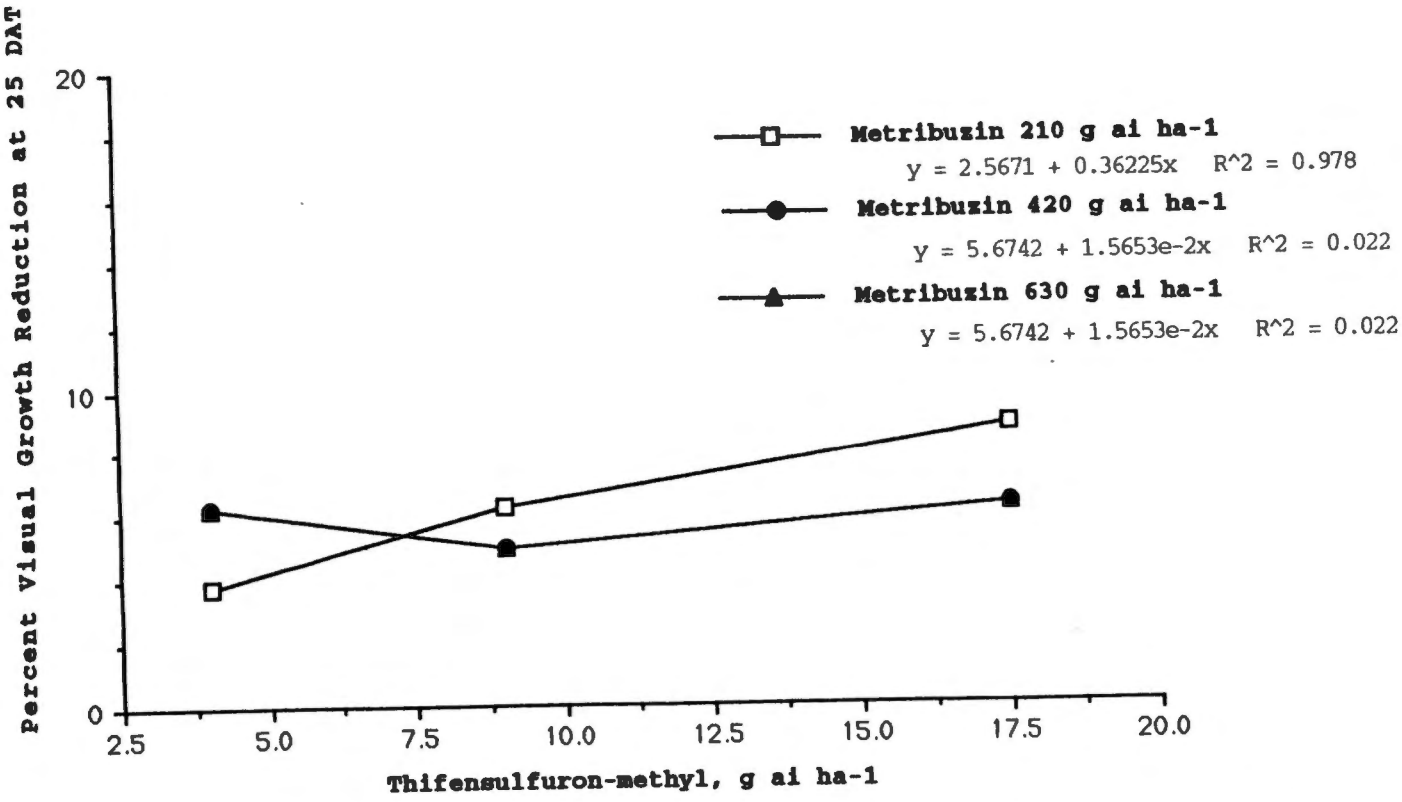

Figure 11. Percent reduction of wild garlic growth at 25 DAT from metribuzin followed at 12 h by thifensulfuron-methyl at Spring Hill in 1987.

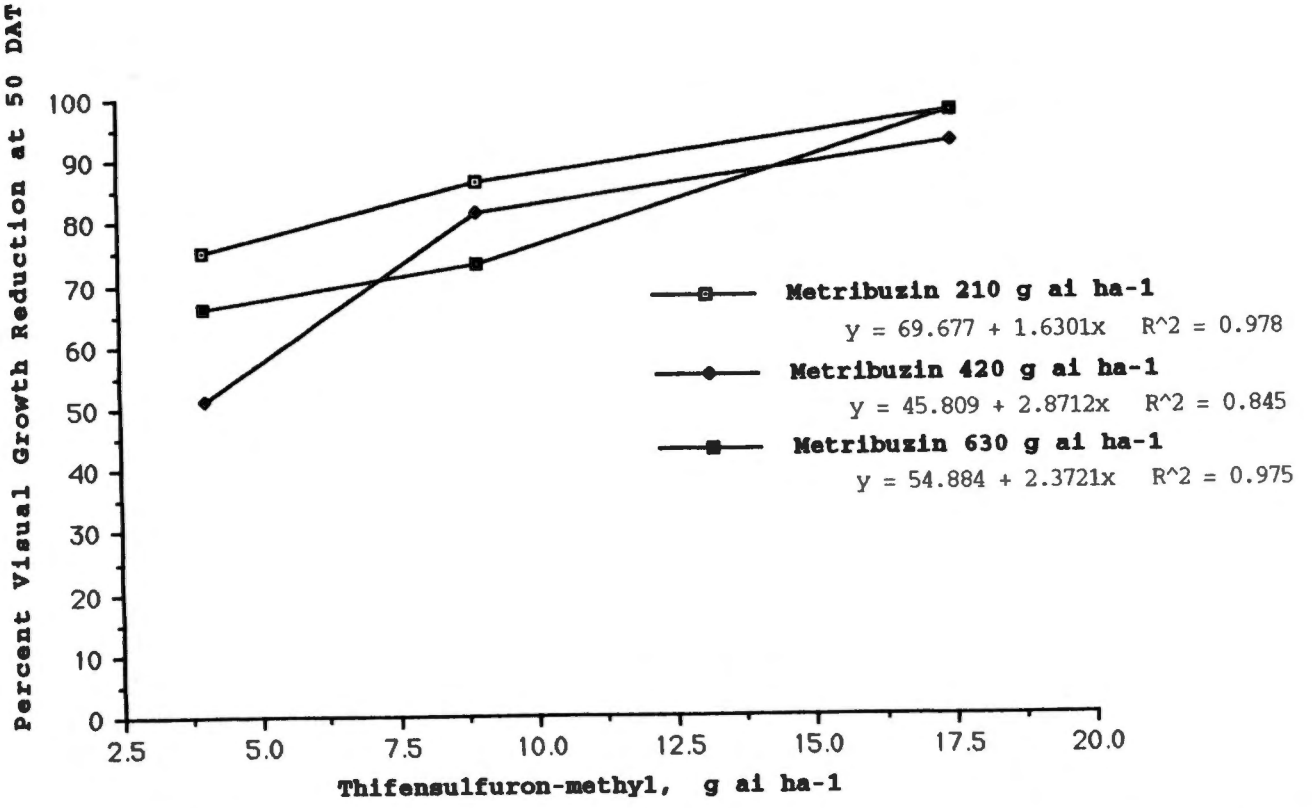

Figure 12. Percent reduction of wild garlic growth at 50 DAT from metribuzin followed at 12 h by thifensulfuron-methyl at Spring Hill in 1987.

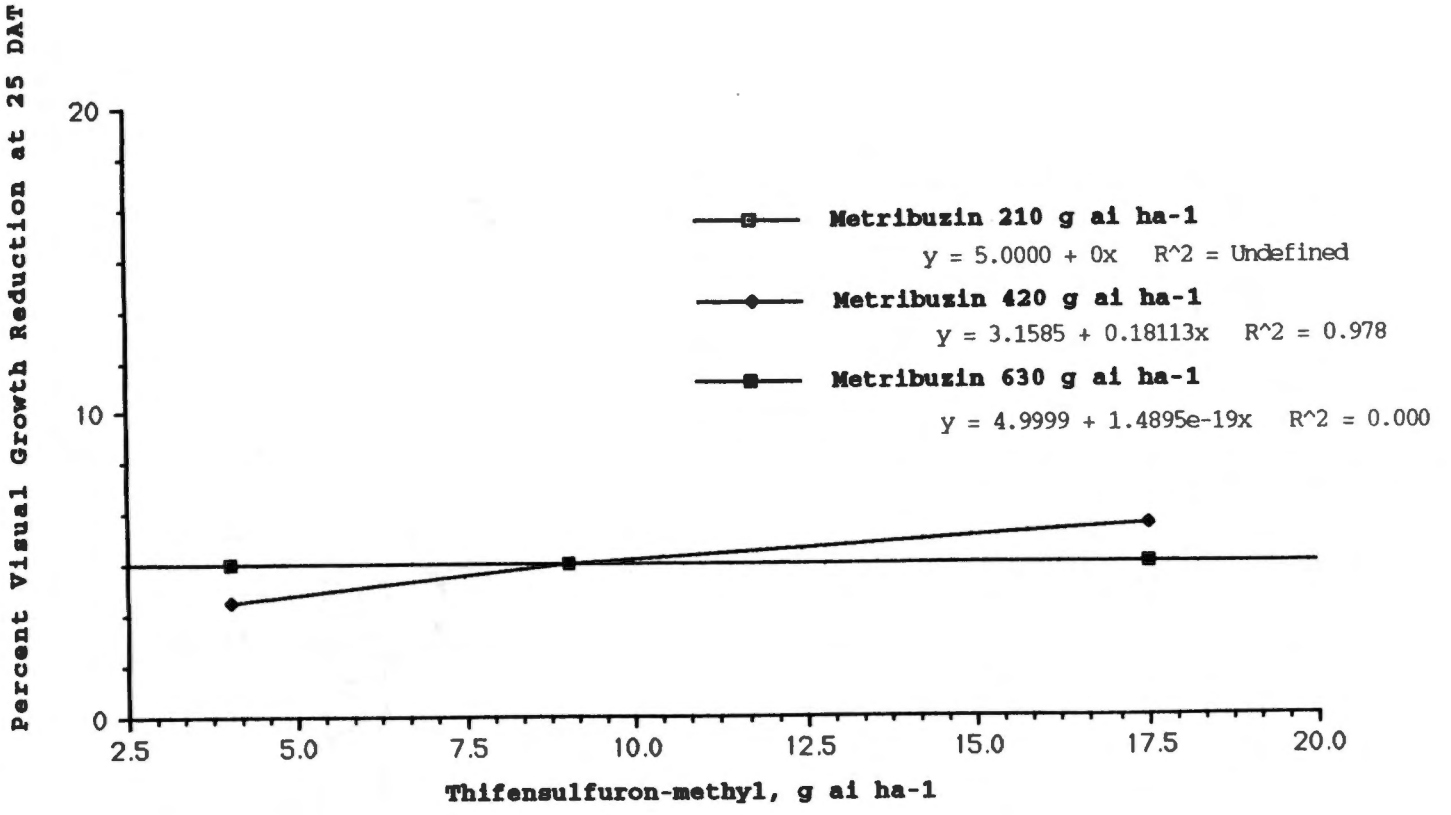

Figure 13. Percent reduction of wild garlic growth at 25 DAT from metribuzin followed at 24 h by thifensulfuron-methyl at Spring Hill in 1987.

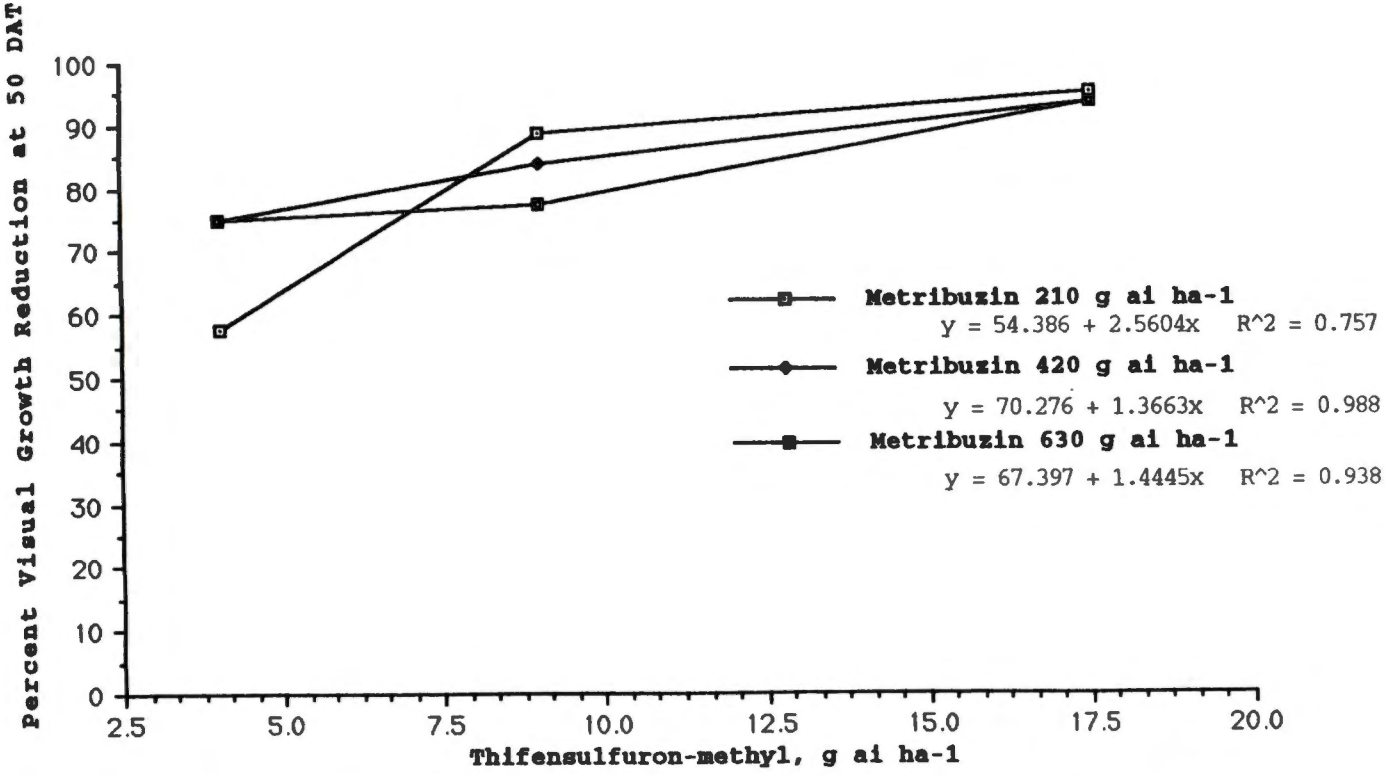

Figure 14. Percent reduction of wild garlic growth at 50 DAT from metribuzin followed at 24 h by thifensulfuron-methyl at Spring Hill in 1987.

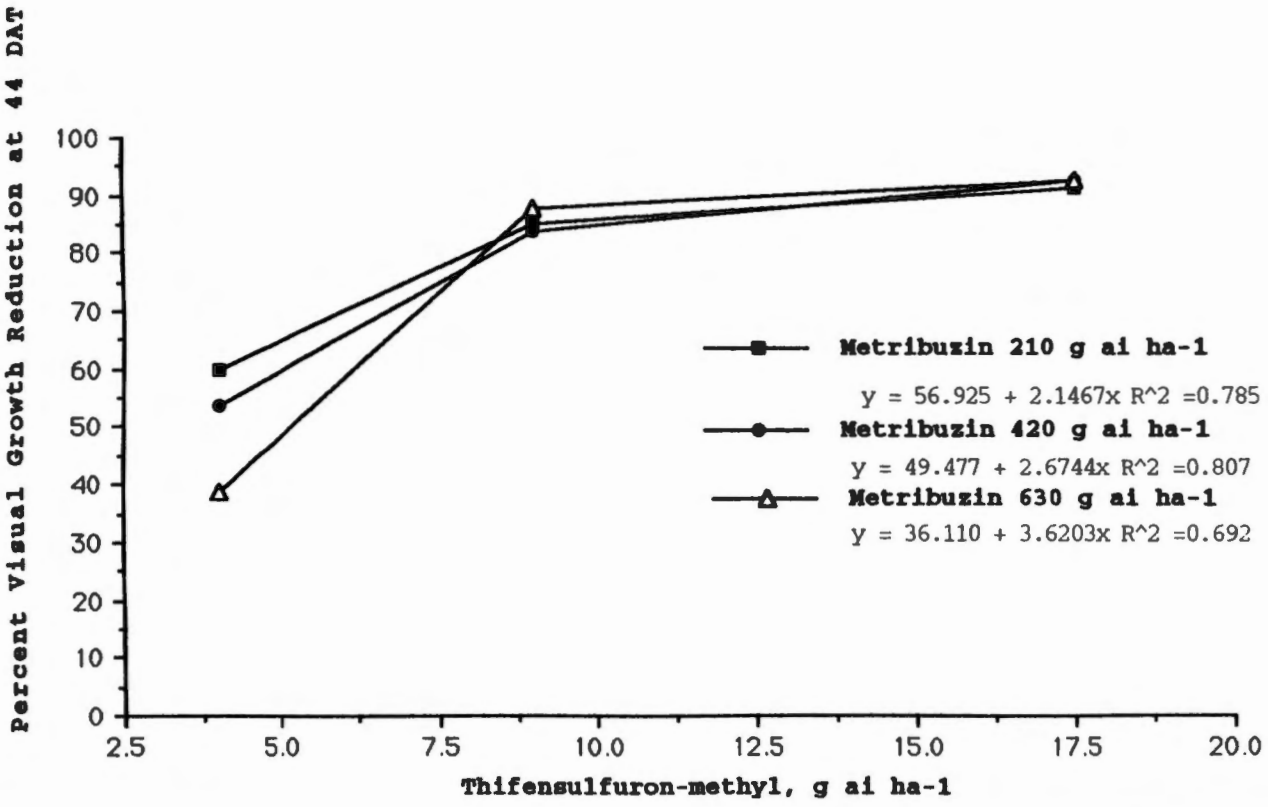

Figure 15. Percent reduction of wild garlic growth at 44 DAT from metribuzin followed at 12 h by thifensulfuron-methyl at Spring Hill in 1988.

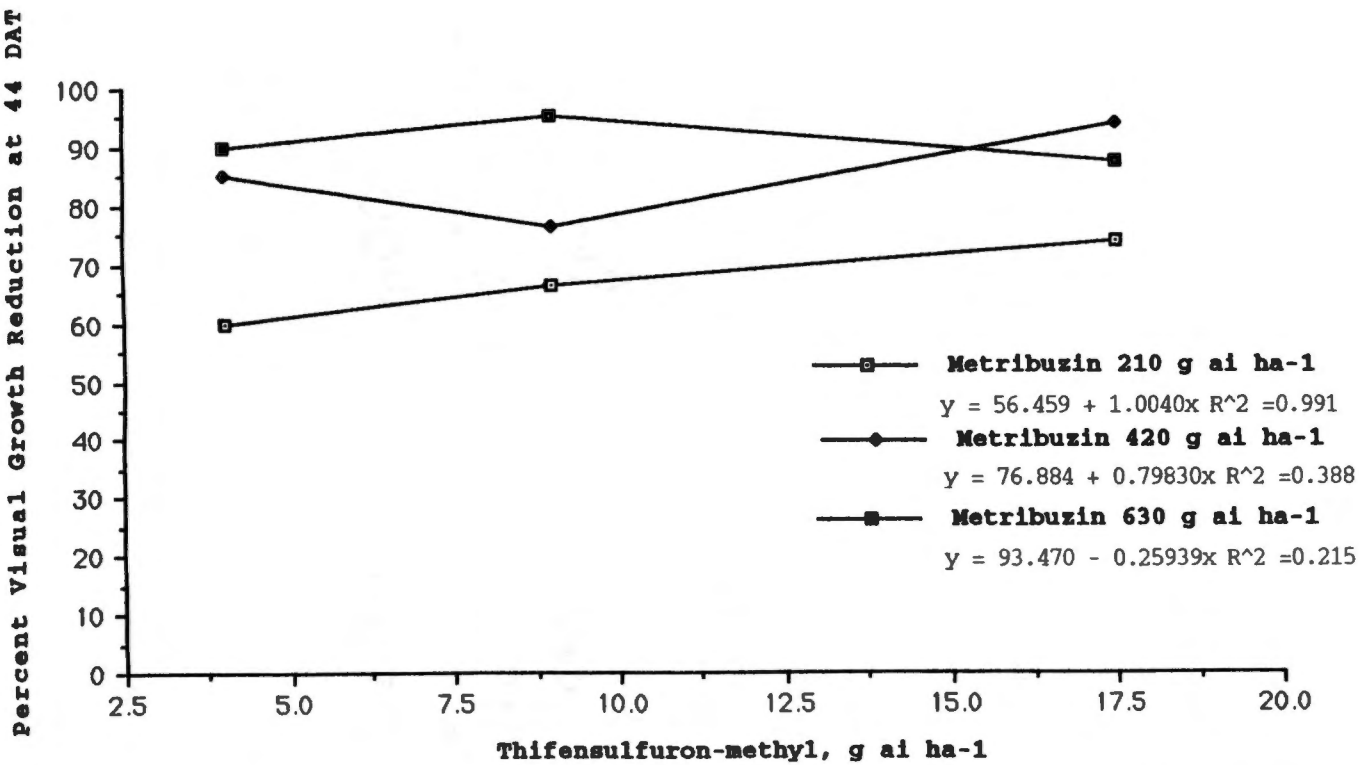

Figure 16. Percent reduction of wild garlic growth at 44 DAT from metribuzin followed at 24 h by thifensulfuron-methyl at Spring Hill in 1988.

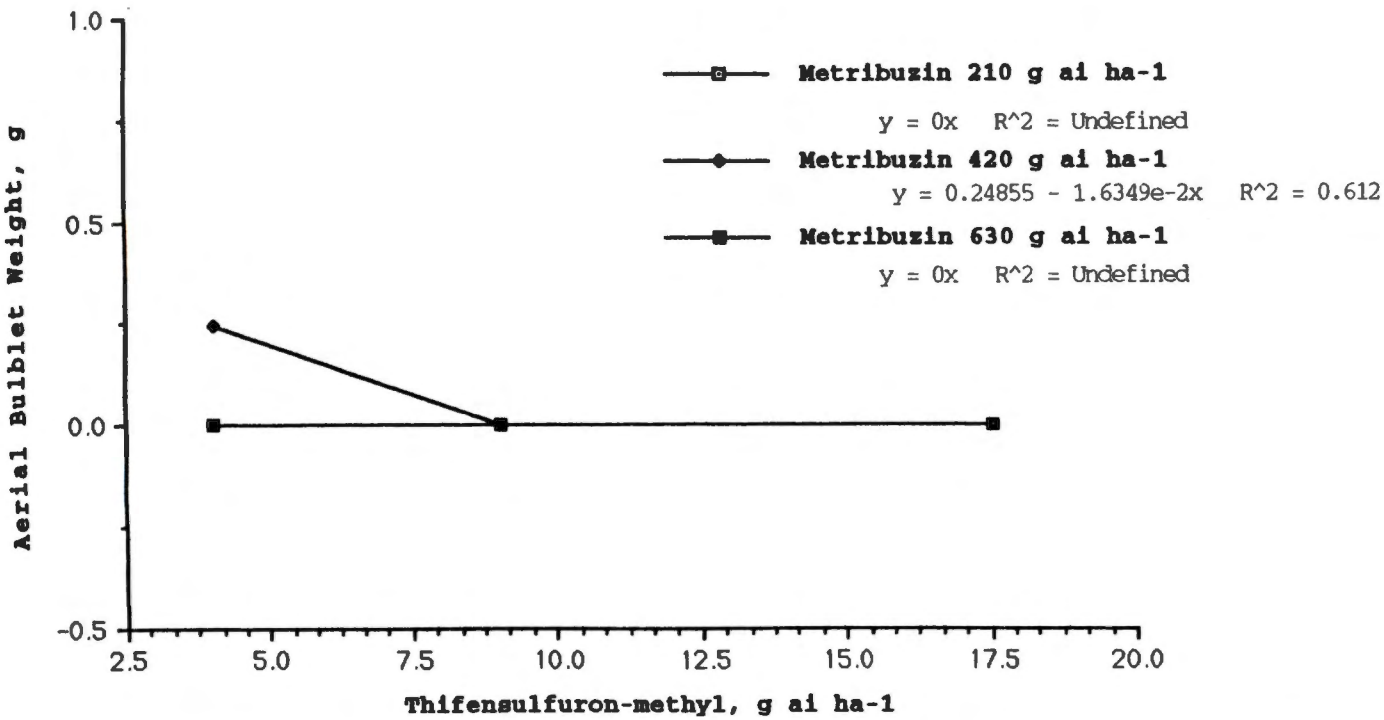

Figure 17. Reduction of wild garlic aerial bulblet weight from metribuzin followed at 12 h by thifensulfuron-methyl at Spring Hill in 1987.

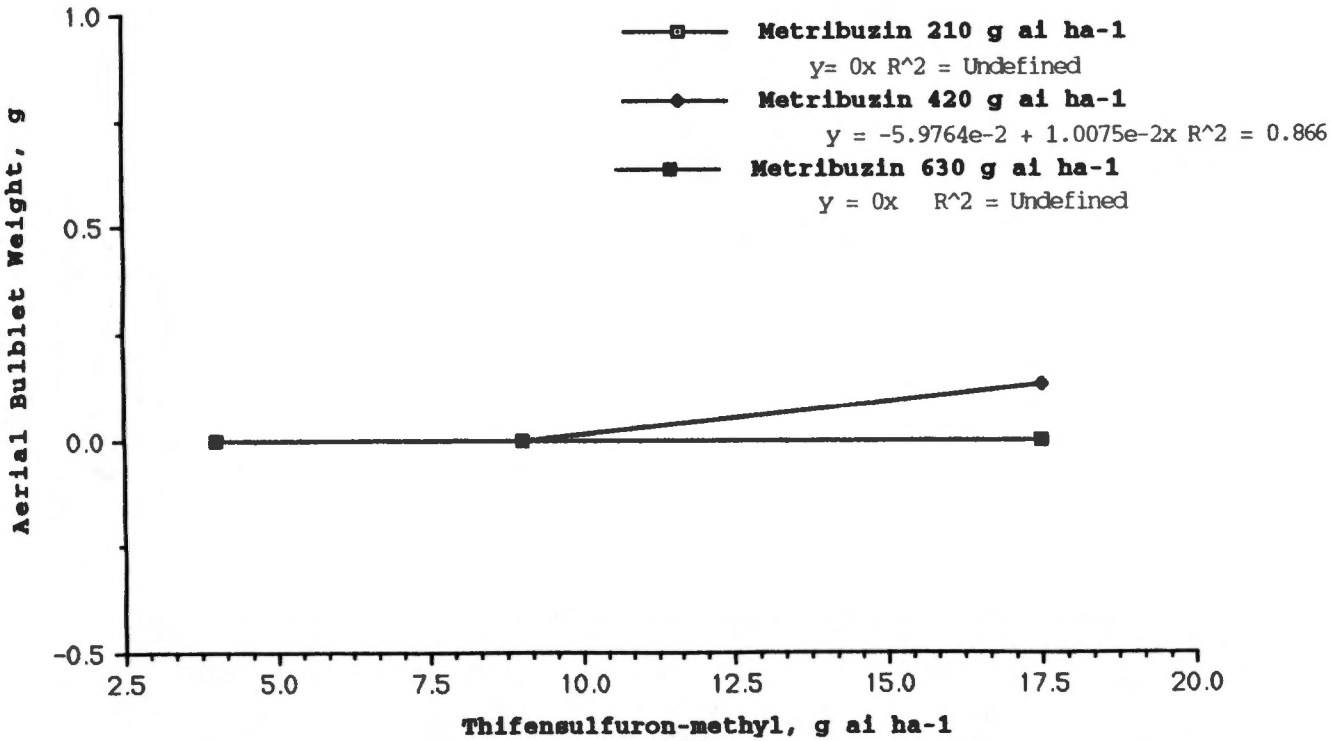

Figure 18. Reduction of wild garlic aerial bulblet weight from metribuzin followed at 24 h by thifensulfuron-methyl at Spring Hill in 1987.

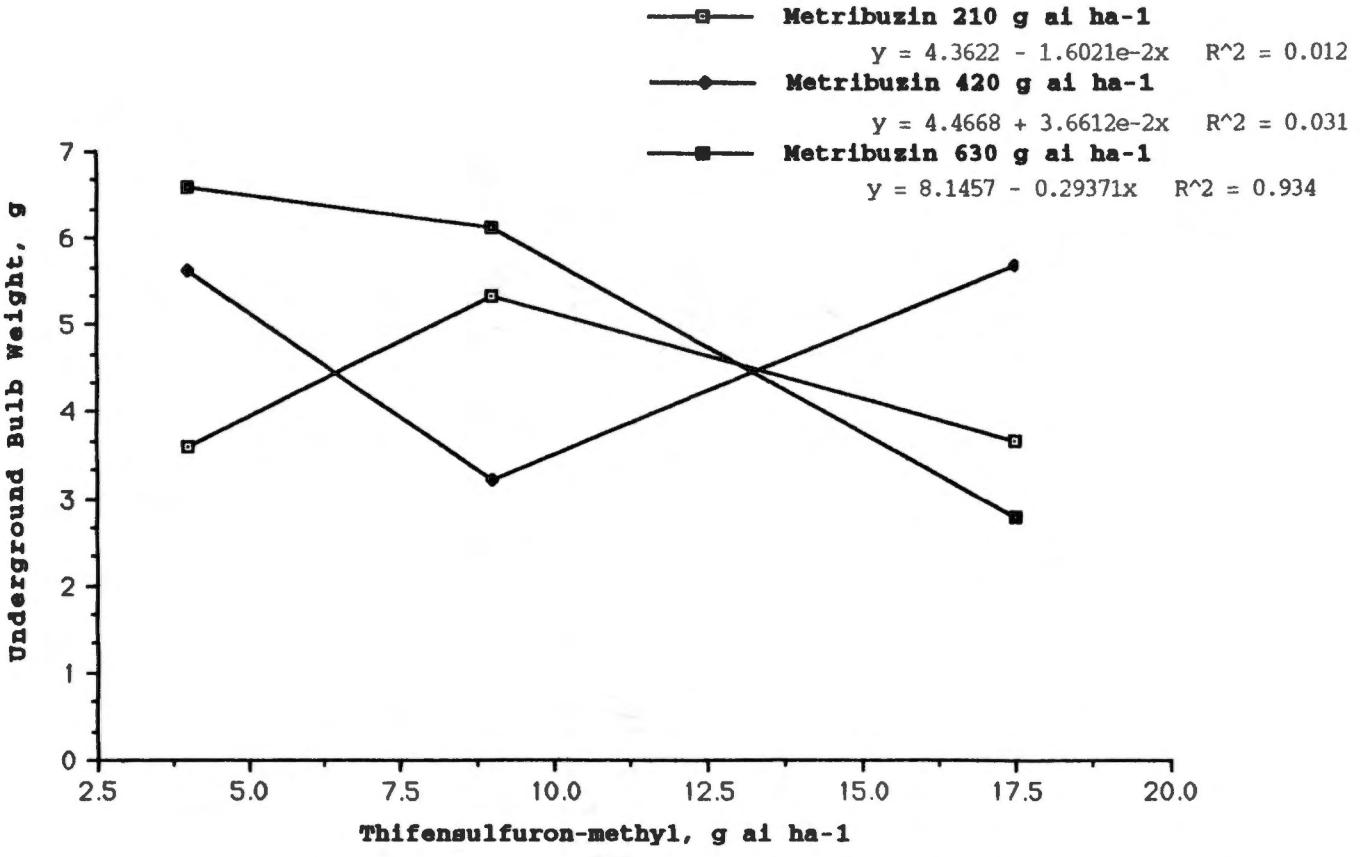

Figure 19. Reduction of wild garlic underground bulb weight from metribuzin followed at 12 h by thifensulfuron-methyl at Spring Hill in 1987.

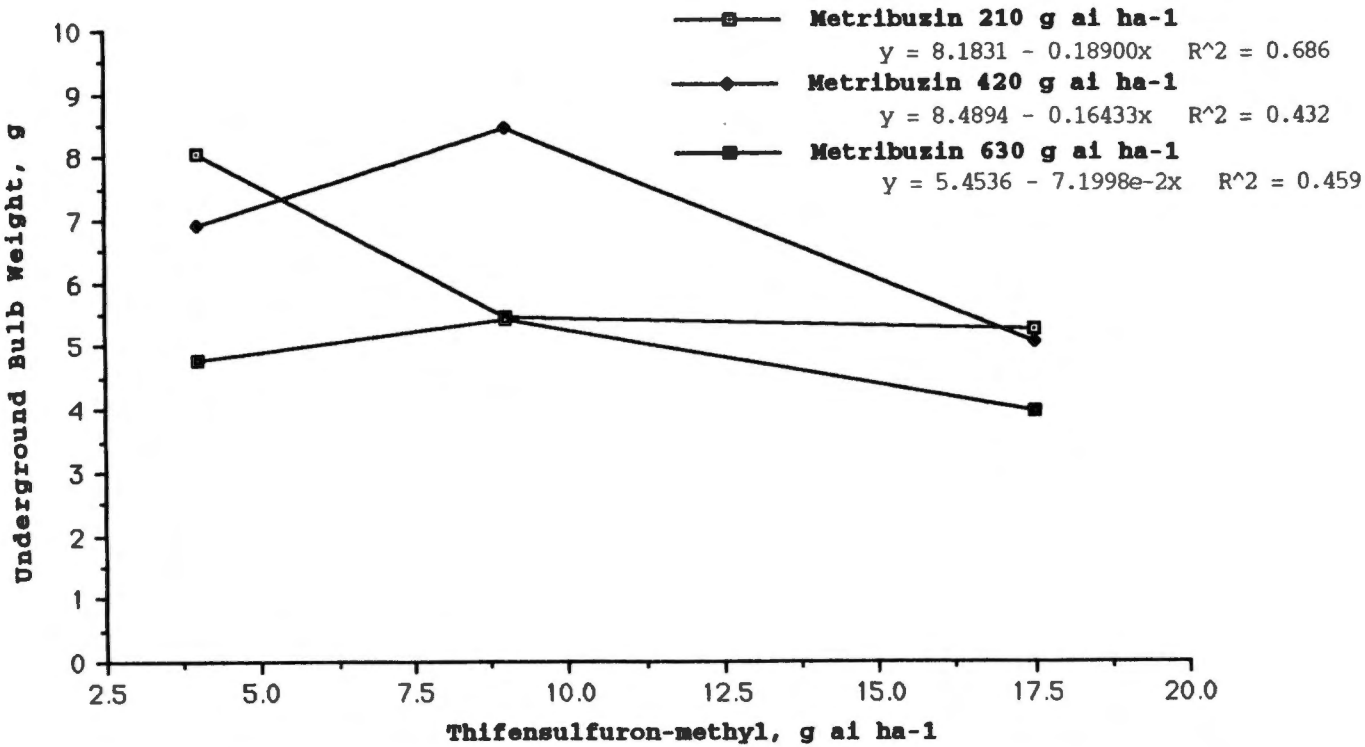

Figure 20. Reduction of wild garlic underground bulb weight from metribuzin followed at 24 h by thifensulfuron-methyl at Spring Hill in 1987.

Table 14. Effects of thifensulfuron-methyl on wild garlic shoots, roots and underground bulbs in the greenhouse[a].

Treatment[b]	Herbicide rate	Bulb[c] Quality	Bulb[d] Weight	Shoot[e] Weight	Roots[f] per Bulb	Root[g] Length	Shoot[h] Length-
	g ai ha^{-1}	0-100	g	g	No.	cm	
Thifensulfuron	4	89 a	1.8 a	0.4 b	13 b	15.3 b	8.4 b
Thifensulfuron	9	86 a	1.8 a	0.4 b	12 b	11.8 c	6.3 b
Thifensulfuron	17.5	83 a	1.5 a	0.3 b	13 b	10.1 c	5.7 b
Thifensulfuron	35	69 a	1.6 a	0.3 b	8 b	6.0 d	1.5 c
Control	_____	95 b	1.5 a	0.9 a	25 a	47.5 a	14.0 a

[a]Experiment was repeated and results were combined. Means in the same column followed by the same letter do not differ at the 0.05 level according to Duncan's Multiple Range mean separation.
[b]All treatments received 0.25% v/v nonionic surfactant. Thifensulfuron and thifensulfuron-methyl are the same.
[c]Bulb quality rated on scale of 0 (poor) to 100 (excellent).
[d]Bulb weight (g) is the mean weight per bulb.
[e]Shoot weight (g) is the mean weight per bulb.
[f]Root number is the mean number per bulb.
[g]Root length (cm) is the mean root length per bulb.
[h]Shoot length (cm) is the mean shoot length per bulb.

Statistically, all rates of thifensulfuron-methyl reduced growth of wild garlic bulbs compared to the control. Shoot weight decreased to 0.3 g compared to 0.9 g in the control. There appears to be a greater reduction of bulb quality and growth at 35 g ai ha^{-1} as compared to the lower rates.

Antagonism of thifensulfuron-methyl activity by metribuzin occurred in the greenhouse (Table 15). The addition of 420 g ai ha^{-1} of metribuzin to 9 through 35 g ai ha^{-1} thifensulfuron-methyl reduced the latters ability to inhibit shoot growth. Shoot weight did not differ between 35 g ai ha^{-1} of thifensulfuron-methyl alone and in tank-mixtures with 420 g ai ha^{-1} metribuzin. Thifensulfuron-methyl at 35 g ai ha^{-1} alone reduced wild garlic shoot length to 1.8 cm compared to 7.5 cm when tank-mixed with 420 g ai ha^{-1}, indicating strong antagonism. Metribuzin alone at 420 g ai ha^{-1} had little activity on wild garlic compared to thifensulfuron-methyl. However, some suppression was noted for most growth parameters when comparing metribuzin to the control.

The inhibition of shoot and root development from bulbs by thifensulfuron-methyl indicates its activity is at the meristematic areas. It is difficult to determine if the bulb becomes necrotic due to the suppression of these meristems. Thirty days following treatment in the greenhouse, bulb quality was still rated at 69 on a scale of 0 to 100 for thifensulfuron-methyl at 35 g ai ha^{-1} (Table 14).

Table 15. Effects of metribuzin and thifensulfuron-methyl alone and tank-mixed on growth of wild garlic in the greenhouse[a].

Treatment[b]	Herbicide rate	Bulb[c] Quality	Bulb[d] Weight	Shoot[e] Weight	Roots[f] per Bulb	Root[g] Length	Shoot[h] Length
	g ai ha^{-1}	0-100	g	g	No.	cm	
Thifensulfuron	9	88 a	1.1 ab	0.3 a	7 b	32.5 b	5.3 d
Thifensulfuron	17.5	98 a	0.8 a	0.2 abc	9 b	41.5 b	4.7 d
Thifensulfuron	35	83 a	0.7 b	0.1 c	4 b	26.2 b	1.8 e
Thifensulfuron + Metribuzin	9 420	98 a	0.7 b	0.2 ab	7 b	45.3 b	9.8 b
Thifensulfuron + Metribuzin	17.5 420	100 a	1.3 a	0.1 bc	8 b	40.3 b	9.3 b
Thifensulfuron + Metribuzin	35 420	83 a	0.9 b	0.2 abc	4 b	16.5 b	7.5 c
Metribuzin	420	96 a	0.7 b	0.2 abc	6 b	31.5 b	9.9 b
Control	____	100 a	1.4 a	0.3 a	16 a	79.0 a	14.8 a

[a]Experiment was repeated, with combined analysis. Means in the same column followed by the same letter do not differ at the 0.05 level according to Duncan's Multiple Range mean separation
[b]All treatments received 0.25% v/v nonionic surfactant. Thifensulfuron and thifensulfuron-methyl are the same.
[c]Bulb quality rated on scale of 0 (poor) to 100 (excellent).
[d]Bulb weight (g) is the mean weight per bulb.
[e]Shoot weight (g) is the mean weight per bulb.
[f]Root number is the mean number per bulb.
[g]Root length (cm) is the mean root length per bulb.
[h]Shoot length (cm) is the mean shoot length per bulb.

Antagonism of 35 g ai ha^{-1} thifensulfuron-methyl by metribuzin at 420 g ai ha^{-1} improved bulb quality to 83 out of 100 for the same time period (Table 15.)

Phytotoxicity to Small Grains

There was no phytotoxicity or yield reduction of small grain cultivars at any rate of thifensulfuron-methyl applied at any growth stage. (Tables 16 through 21). Yield differences between wheat and barley cultivars were due to winter kill of barley. 'Coker 747' wheat had the highest yield of all wheat cultivars (Table 16), while the highest yielding barley was 'Volbar' (Table 21). Previous research has demonstrated the tolerance of small grains to thifensulfuron-methyl and chlorsulfuron (Kapusta and Anderson 1987; Durgan and Miller 1988; Anderson and Kapusta 1988; Eberlein and Miller 1984; Ambach et al. 1984; Khodayari et al. 1985; Brewster and Appleby 1983). Chlorsulfuron was the first sulfonylurea herbicide to be used in small grains and has shown minimal phytotoxicity to wheat and barley at rates up to 70 g ai ha^{-1} (Miller and Nalewja 1979).

In conclusion, thifensulfuron-methyl activity on aerial bulblets is influenced by rate and time of application. Generally, 35 g ai ha^{-1} proved most effective in reducing aerial bulblets if applied in late March or early April. Rates of 4, 9 and 17.5 g ai ha^{-1} were not as effective as 35 g ai ha^{-1} regardless of date when applied.

Table 16. Influence of thifensulfuron-methyl applied at three stages of development on yield of 'Coker 747' wheat[a].

Treatment[c]	Herbicide rate	Feekes Growth Stage[b]	Yield
	g ai ha^{-1}		kg ha^{-1}
Thifensulfuron	17.5	4	4200 ab
"	"	6	4770 a
"	"	10	4130 ab
"	35	4	4500 ab
"	"	6	3900 b
"	"	10	4170 ab
"	53	4	3900 b
"	"	6	4230 ab
"	"	10	4480 ab
Control	———	4	4590 ab
"	"	6	4170 ab
"	"	10	4400 ab

[a]Means in a column followed by the same letter do not differ at 0.05 level according to the Duncan's Multiple Range mean separation.
[b]Large, E. C. 1954. Growth stages in cereals: Illustrations of the Feekes scale. Plant Path. 3:128-129.
[c]Thifensulfuron and thifensulfuron-methyl are the same.

Table 17. Influence of thifensulfuron-methyl applied at three stages of development on yield of 'Caldwell' wheat[a].

Treatment[c]	Herbicide rate	Feekes Growth Stage[b]	Yield
	g ai ha^{-1}		kg ha^{-1}
Thifensulfuron	17.5	4	3494 abc
"	"	6	3400 abc
"	"	10	3460 abc
"	35	4	3030 bc
"	"	6	3440 abc
"	"	10	3470 abc
"	53	4	3320 abc
"	"	6	2930 c
"	"	10	3760 ab
Control	———	4	3580 abc
"	"	6	3280 abc
"	"	10	3880 a

[a]Means in a column followed by the same letter do not differ at 0.05 level according to the Duncan's Multiple Range mean separation.
[b]Large, E. C. 1954. Growth stages in cereals: Illustrations of the Feekes scale. Plant Path. 3:128-129.
[c]Thifensulfuron and thifensulfuron-methyl are the same.

Table 18. Influence of thifensulfuron-methyl applied at three stages of development on yield of 'Pioneer 2550' wheat[a].

Treatment[c]	Herbicide rate	Feekes Growth Stage[b]	Yield
	g ai ha^{-1}		kg ha^{-1}
Thifensulfuron	17.5	4	4040 a
"	"	6	4180 a
"	"	10	3920 a
"	35	4	4460 a
"	"	6	4350 a
"	"	10	4270 a
"	53	4	4030 a
"	"	6	4230 a
"	"	10	3880 a
Control	____	4	4190 a
"	"	6	3940 a
"	"	10	4350 a

[a]Means in a column followed by the same letter do not differ at 0.05 level according to Duncan's Multiple Range mean separation.
[b]Large, E. C. 1954. Growth stages in cereals: Illustrations of the Feekes scale. Plant Path. 3:128-129.
[c]Thifensulfuron and thifensulfuron-methyl are the same.

Table 19. Influence of thifensulfuron-methyl applied at three stages of development on yield of 'Fillmore' wheat[a].

Treatment[c]	Herbicide rate	Feekes Growth Stage[b]	Yield
	g ai ha^{-1}		kg ha^{-1}
Thifensulfuron	17.5	4	3760 a
"	"	6	3360 a
"	"	10	3370 a
"	35	4	3190 a
"	"	6	3340 a
"	"	10	3480 a
"	53	4	3690 a
"	"	6	3280 a
"	"	10	3430 a
Control	____	4	3210 a
"	"	6	3330 a
"	"	10	3800 a

[a]Means in a column followed by the same letter do not differ at 0.05 level according to Duncan's Multiple Range mean separation.
[b]Large, E. C. 1954. Growth stages in cereals: Illustrations of the Feekes scale. Plant Path. 3:128-129.
[c]Thifensulfuron and thifensulfuron-methyl are the same.

Table 20. Influence of thifensulfuron-methyl applied at three stages of development on yield of 'Barsoy' barley[a].

Treatment[c]	Herbicide rate	Feekes Growth Stage[b]	Yield
	g ai ha^{-1}		kg ha^{-1}
Thifensulfuron	17.5	4	1080 ab
"	"	6	1070 ab
"	"	10	1460 a
"	35	4	1130 ab
"	"	6	1110 ab
"	"	10	1220 ab
"	53	4	1130 ab
"	"	6	870 ab
"	"	10	1410 ab
Control	———	4	1240 ab
"	"	6	1310 ab
"	"	10	770 b

[a]Means in a column followed by the same letter do not differ at 0.05 level according to Duncan's Multiple Range mean separation.
[b]Large, E. C. 1954. Growth stages in cereals: Illustrations of the Feekes scale. Plant Path. 3:128-129.
[c]Thifensulfuron and thifensulfuron-methyl are the same.

Table 21. Influence of thifensulfuron-methyl applied at three stages of development on yield of 'Volbar' barley[a].

Treatment[c]	Herbicide rate	Feekes Growth Stage[b]	Yield
	g ai ha^{-1}		kg ha^{-1}
Thifensulfuron	17.5	4	3310 a
"	"	6	2880 a
"	"	10	3490 a
"	35	4	3080 a
"	"	6	3540 a
"	"	10	3150 a
"	53	4	2850 a
"	"	6	3310 a
"	"	10	3380 a
Control	____	4	3150 a
"	"	6	3130 a
"	"	10	2630 a

[a]Means in a column followed by the same letter do not differ at 0.05 level according to Duncan's Multiple Range mean separation.
[b]Large, E. C. 1954. Growth stages in cereals: Illustrations of the Feekes scale. Plant Path. 3:128-129.
[c]Thifensulfuron and thifensulfuron-methyl are the same.

Antagonism of aerial bulblet suppression did not occur when metribuzin at 210, 420 or 630 g ai ha^{-1} was tank-mixed with thifensulfuron-methyl at 17.5 g ai ha^{-1}. Tank-mixing 4, 9 and 17.5 g ai ha^{-1} of thifensulfuron-methyl with 210, 420 and 630 g ai ha^{-1} of metribuzin resulted in antagonized activity on underground bulbs. Sequential application of thifensulfuron-methyl followed by metribuzin at 12 h was not antagonistic on aerial bulblets. The ability of thifensulfuron-methyl to reduce underground bulb weight was antagonized by metribuzin at 12 and 24 h. When thifensulfuron-methyl was applied 12 h after metribuzin, activity on aerial bulblets was not affected. Thifensulfuron-methyl at 53 g ai ha^{-1} may be applied to small grain cultivars with no apparent phytotoxicity or yield reduction.

The benefit of thifensulfuron-methyl in small grain production is apparent from this research. Historically the adverse effects of wild garlic bulblets on small grain quality necessitated numerous control measures which afforded only partial suppression of aerial bulblets in small grains. The use of 2,4-D improved the suppression of aerial bulblets, but had little effect on the development of underground bulbs. Also, injury to small grains could occur from 2,4-D, while thifensulfuron-methyl may be applied safely to small grains. Research suggests the most effective rate in suppression of aerial bulblets is 35 g ai ha^{-1} applied in late March to early April.

Long term suppression and control of wild garlic underground soft shell bulbs by thifensulfuron-methyl at 35 g ai ha^{-1} does occur, but evidence indicates that bulbs may visually appear viable up to 30 d following treatment. Whether all meristematic areas in the bulb are killed is not known. Also, the activity of thifensulfuron-methyl on all underground bulb types has not been evaluated. The often dormant hardshell bulb may represent a potential for continued infestation in small grain production for many years.

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CHAPTER IV

RESPONSE OF WILD GARLIC (*ALLIUM vineale* L.) AERIAL BULBLETS AND SOFT OFFSET BULBS TO THIFENSULFURON-METHYL IN STERILE CULTURE

Introduction

Wild garlic represents a serious weed pest in small grains. Aerial bulblets reduce small grain quality by releasing volatile compounds in processing (Carson and Wong 1961; Freeman and Mossadeghi 1970; Davey et al. 1974; Shelby and Collin 1976).

Sulfonylurea herbicides represent new herbicide technology with low rates (g ha^{-1}) and specificity of action by inhibiting ALS (acetolactate synthase)(Beyer et al. 1987; Ray 1985; Chaleff and Mauvais 1984; Ray 1984; Erbes 1984; Dieck et al. 1984).

ALS catalyzes the first step of the biosynthetic pathway for branched chain amino acids and was found to be highly sensitive in susceptible species to the first commercial sulfonylurea herbicide, chlorsulfuron (Ray 1982). Growth reduction was closely associated with inhibition of plant cell division.

Ray (1984) reported that chlorsulfuron at 2.8 nM significantly inhibited root growth of peas (*Pisum sativum* L.) in culture. The amino acids valine, leucine, isoleucine and alanine at 100 nM alleviated chlorsulfuron induced growth inhibition.

Giardina et al.(1984) found the addition of the amino acids valine and isoleucine at 100 nM each did not remove the inhibitory effect of chlorsulfuron at 28 nM on maize (Zea mays L.) roots in culture. These results lead to the conclusion that the mechanism of action for chlorsulfuron, and the sulfonylurea herbicides in general, may encompass more than one site.

Evaluation of thifensulfuron-methyl activity on wild garlic (Allium vineale L.) has been investigated in field and laboratory studies (Kaufman and Ritter, 1987; Anderson and Kapusta, 1987 and 1988; Gast, 1988). Translocation of ^{14}C thifensulfuron-methyl in wild garlic was slow but the meristematic region of the parent bulb had the highest concentration (Gast 1988).

An alternative to greenhouse studies in evaluating herbicide activity may be the use of treated growth medium in sterile culture. Evaluation of phytotoxicity to a given compound may use various parts of the plant ranging from aseptically grown seedlings to suspension cultures (Martensson and Nilsson 1989; Mersie and Foy 1987; Hess et al. 1990; Zilkah and Gressel 1977).

The objectives of this research were to: (1) evaluate the activity of thifensulfuron-methyl on shoot and root development of aerial bulblets and soft offset bulbs grown in sterile culture;

(2) evaluate the ability of the amino acids valine, leucine and isoleucine to overcome the activity of thifensulfuron-methyl on shoot and root growth of aerial bulblets and soft offset bulbs growing in culture and (3) view microscopically, histological and anatomical effects of thifensulfuron-methyl on the meristem of soft offset bulbs.

Materials And Methods

Evaluation of Aerial Bulblet Sensitivity to Thifensulfuron-Methyl

Laboratory studies were conducted using aerial bulblets of Allium vineale L. To facilitate sterilization of aerial bulblets, the outer scale leaf was removed from individual bulblets. Sterilization was conducted using commercial bleach (5.25% sodium hypochlorite) diluted 1:1 with distilled water. To enhance surface activity, 3 to 5 drops of Triton X-100[1] was added to the solution. Bulblets were placed in a sterile basket strainer inside a Laminare Flowhood[2] and suspended in the stirring bleach solution for 10 min. Following sterilization bulblets were rinsed in three individual changes of sterile distilled water and placed on a sterile surface to drain.

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[1]Mallinckrodt, Inc. Paris, Ky.40361.
[2]Laminare Corporation,960 East Hazelwood Ave., Rahway, N.J.07065

SH media was prepared in accordance with Schenk and Hildebrandt (1972). Media pH (5.6) was adjusted with 0.1 N NaOH and autoclaved for 20 min. Stock solutions of technical grade (95%) thifensulfuron-methyl were used to obtain treatments of 1, 0.1, 0.01 and 0.001 mg L^{-1}. Thifensulfuron-methyl treatments were sterile filtered by a syringe equipped with a 0.2 micron Acrodisc[3] filter into cooling media and gently stirred. Media was poured into sterile disposable 100 by 15-mm petri dishes and allowed to solidify. All procedures including sterilization of bulblets and pouring media were conducted in a flowhood. Treatments consisted of two bulblets per plate with 10 replications. Plates were incubated in the dark at 24 C, for 10 d. Evaluation of thifensulfuron-methyl activity included shoot and root length measurement of aerial bulblets.

A similar experiment to the previous study was conducted except the plates were placed in a lighted incubator, 16 h (290 uE m^{-2} s^{-1} photosynthetic photon flux density) at 24 C, followed by 8 h of darkness at 18 C.

Experimental design for both experiments was a randomized complete block with 10 replications. A minimum of two trials were conducted for each experiment. Statistical analysis was performed by analysis of variance on combined data from experimental trials.

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[3]Gelman Sciences, 600 S. Wagner Rd., Ann Arbor, MI. 48106.

Inhibition of Thifensulfuron-Methyl Activity on Aerial Bulblets by Amino Acids

Experiments were conducted to evaluate the influence of analytical grade valine, leucine and isoleucine (L isomers) at 100 uM on thifensulfuron-methyl activity. Thifensulfuron-methyl technical grade (95%) was used to obtain 1, 0.1, 0.01 and 0.001 mg L^{-1} with the amino acids individually and in all possible combinations. Procedures of bulblet and media preparation were described earlier. Experiments were conducted in a lighted incubator as described previously. Shoot and root length from aerial bulblets were measured after 10 d. Experimental design was a randomized complete block with 10 replications. A minimum of two trials were conducted. Statistical analysis was by analysis of variance on combined data from experimental trials.

Response of Soft Offset Bulbs to Thifensulfuron-Methyl

Soft offset bulbs were collected during dormancy and stored in cool, (5 C) dry conditions until used. Bulbs were placed in seedling flats filled with potting media[4] and allowed to grow. Actively growing bulbs were selected at random and washed thoroughly in tap water prior to cutting into bulb stem, leaf and roots.

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[4]Premier Brands, Inc. Standford, CT. 06902.

Caution was exercised in cutting of bulb stem and roots, if too much tissue was removed the meristem enclosed within the rudimentary leaves would be damaged by sterilization. Bulbs were placed in a sterile basket strainer in a flowhood and suspended in a stirring 1:1 bleach (5.25% sodium hypochlorite) and distilled water solution for 10 min. Following sterilization bulbs were rinsed in three changes of sterile distilled water and placed on a sterile surface to drain.

SH media containing thifensulfuron-methyl was prepared as described previously. Thifensulfuron-methyl technical grade (95%) was used for treatments of 1, 0.1, 0.05, 0.25, 0.01 and 0.001 mg L^{-1}. Media was poured into sterile Pyrex culture tubes (25 mm O.D. by 200 mm) and allowed to solidify. One bulb was transferred under sterile conditions to each tube. Root and shoot length were measured at 2-, 5- and 10-d intervals.

The meristem was then removed from the bulb and placed in a vial containing 50% FAA[5] (formalin-acetic acid-alcohol) fixing solution for 48 h. Vials were placed under vacuum to assure removal of air and infiltration. Meristems were dehydrated in a graded tertiary butanol-ethanol series (Johansen 1940; Sass 1958).

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[5]50 ml of 95% ethanol; 40 ml of distilled water; 5 ml of 37% formalin; 5 ml of glacial acetic acid.

Tissue was oriented for longitudinal sectioning and embedded in Paraplast[6] (melting point 56 to 57 C). Serial 10 um sections were mounted using distilled water on glass slides dried and cleared with Histo-Clear[7]. Sections were stained with 0.1% toluidine blue in phosphate buffer (pH 7.0). Photomicrographs of longitudinal sections were taken with a light microscope. The experimental design was a randomized complete block with 10 replications per treatment. The experiment was conducted a minimum of three times. Data were analyzed by analysis of variance.

Inhibition of Thifensulfuron-Methyl Activity on Soft Offset Bulbs as Influenced by Valine, Leucine and Isoleucine

Soft offset bulbs were prepared and utilized as in the previous experiment. Treatments consisted of analytical grade valine, leucine and isoleucine (L isomers) at 100 uM plus thifensulfuron-methyl technical grade (95%) at 1, 0.1, 0.05, 0.25, 0.01 and 0.001 mg L^{-1}. Treatments were added to media in the method previously described. Media was poured into sterile Pyrex culture tubes (25 mm O.D. by 200 mm) and allowed to solidify. One bulb was transferred under sterile conditions to each tube. Root and shoot length were measured at 2, 5 and 10-d intervals.

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[6]Monofet Scientific, div. of Sherwood Medical, St. Louis, Mo. 63103.
[7]National Diagonistics, 1013-1017 Kennedy Blvd. Manville, N. J. 08835.

The meristem was then removed from each bulb and prepared for histological examination as previously described.

The experimental design was a randomized complete block with 10 replications per treatment. The experiment was conducted a minimum of three times. Data were analyzed by analysis of variance.

Results And Discussion

Evaluation of Aerial Bulblet Sensitivity to Thifensulfuron-Methyl

Thifensulfuron-methyl inhibited shoot growth of aerial bulblets more in the dark than in light (Figure 21). All rates of thifensulfuron-methyl reduced shoot growth in the dark. Shoot growth ranged from 53 mm for the 0.001 mg L^{-1} to 17 mm for the 1 mg L^{-1} rate. Reduction of shoot growth occurred with all rates of thifensulfuron-methyl in the light, however reduction of shoot growth from thifensulfuron-methyl was not as great in the light as in the dark, due possibly to photosynthetic activity of aerial bulblets in the light. Shoot growth for the light ranged from 78 mm for 0.001 mg L^{-1} to 63 mm for 1 mg L^{-1}.

Light or dark environments had little affect on thifensulfuron-methyl activity on root growth of aerial bulblets (Figure 22).

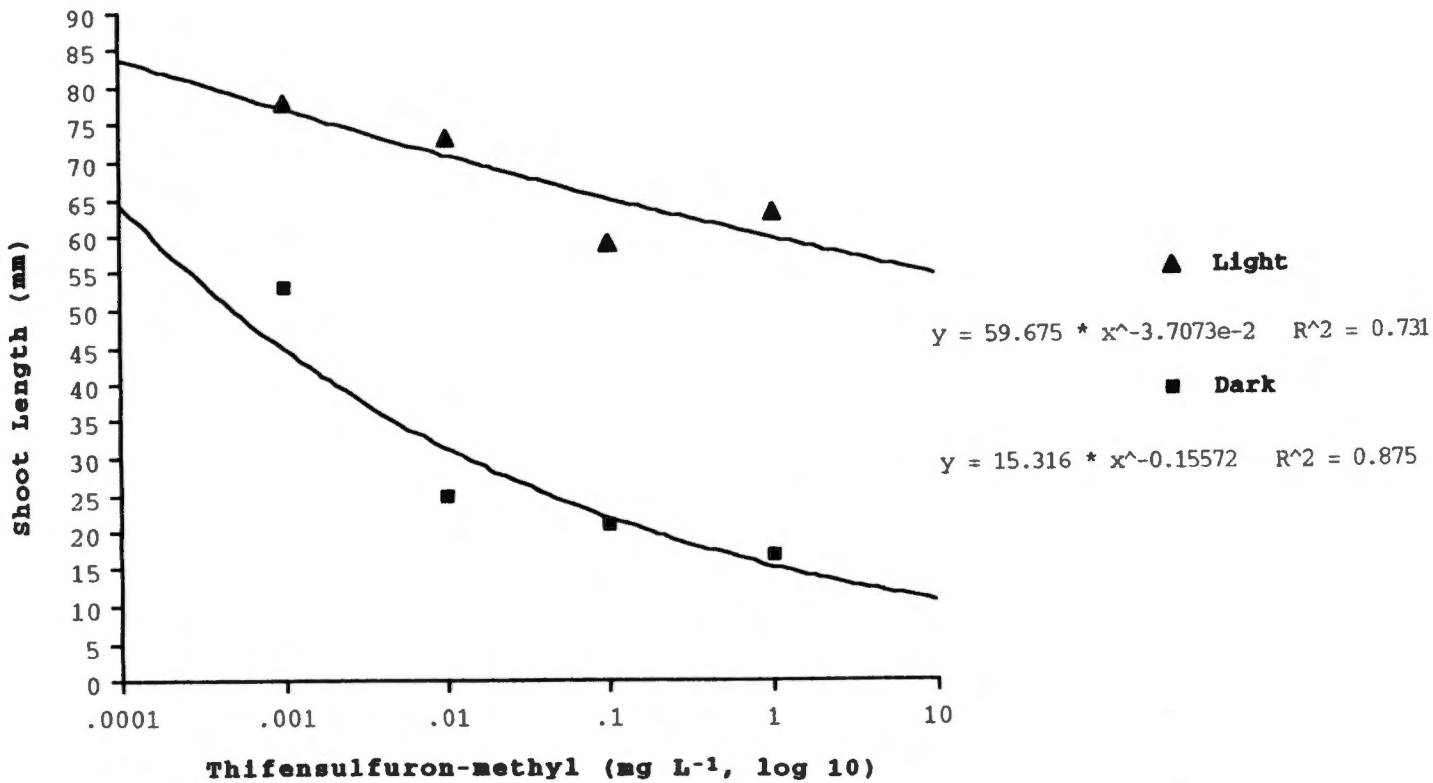

Figure 21. Effect of thifensulfuron-methyl on shoot growth of aerial bulblets grown in the light and dark for 10 d. Values are combined treatment means from conducting a minimum of two trials for the experiment. Untreated check shoot length in the light and dark was 90 and 71 mm, respectively.

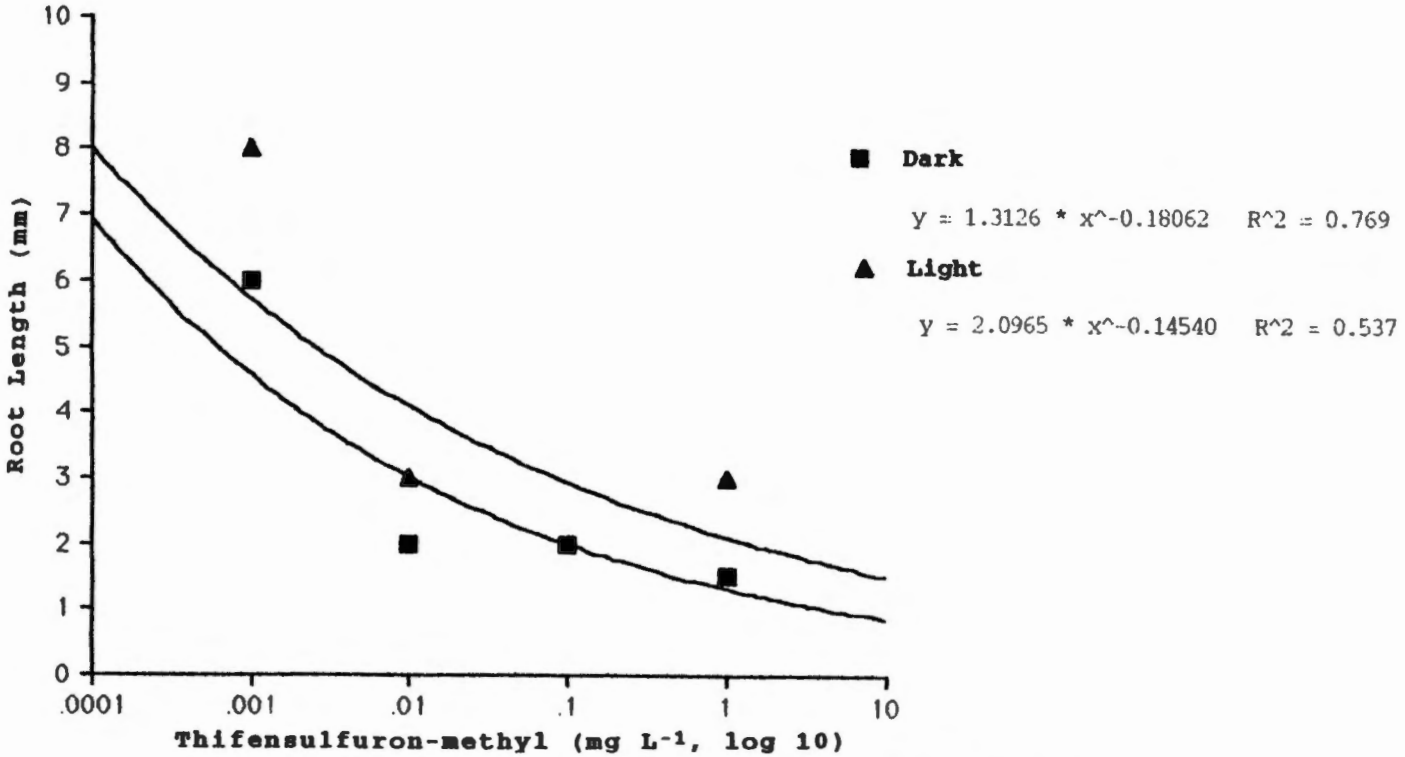

Figure 22. Effect of thifensulfuron-methyl on root growth of aerial bulblets grown in the light and dark for 10 d. Values are combined treatment means from conducting a minimum of two trials for the experiment. Untreated check root length in the light and dark was 27 and 28 mm, respectively.

The effect of thifensulfuron-methyl on root growth from aerial bulblets did not differ between light and dark environments as would be expected since they are not photosynthetically active. For both environments the trend was toward reduced root growth with increasing concentration of thifensulfuron-methyl. Reduction of root growth in sterile culture has been noted for chlorsulfuron (Klingman and Pepper 1989). A bean root bioassay using 'Kentucky Wonder' beans (Phaseolus vulgaris L.) evaluated root growth as affected by chlorsulfuron and CGA-131036. Both herbicides inhibited root growth at 10 to 300 nM.

Inhibition of Thifensulfuron-Methyl Activity on Aerial Bulblets by Amino Acids

Evaluation of the amino acids valine, leucine and isoleucine either individually or in combination at 100 uM concentration produced variable results in alleviating thifensulfuron-methyl activity on aerial bulblet growth (Figure 23). Thifensulfuron-methyl inhibition of aerial bulblet shoot elongation at 0.01 mg L^{-1} was not alleviated by amino acids (Figure 23). Shoot growth inhibition was negligible at 0.001 mg L^{-1} thifensulfuron-methyl and therefore any influence of the amino acids could not be determined (Figure 24). Reduction of root growth was pronounced at all rates of thifensulfuron-methyl. Even at 0.001 mg L^{-1} inhibition was alleviated by the addition of valine, leucine and isoleucine (Figure 25).

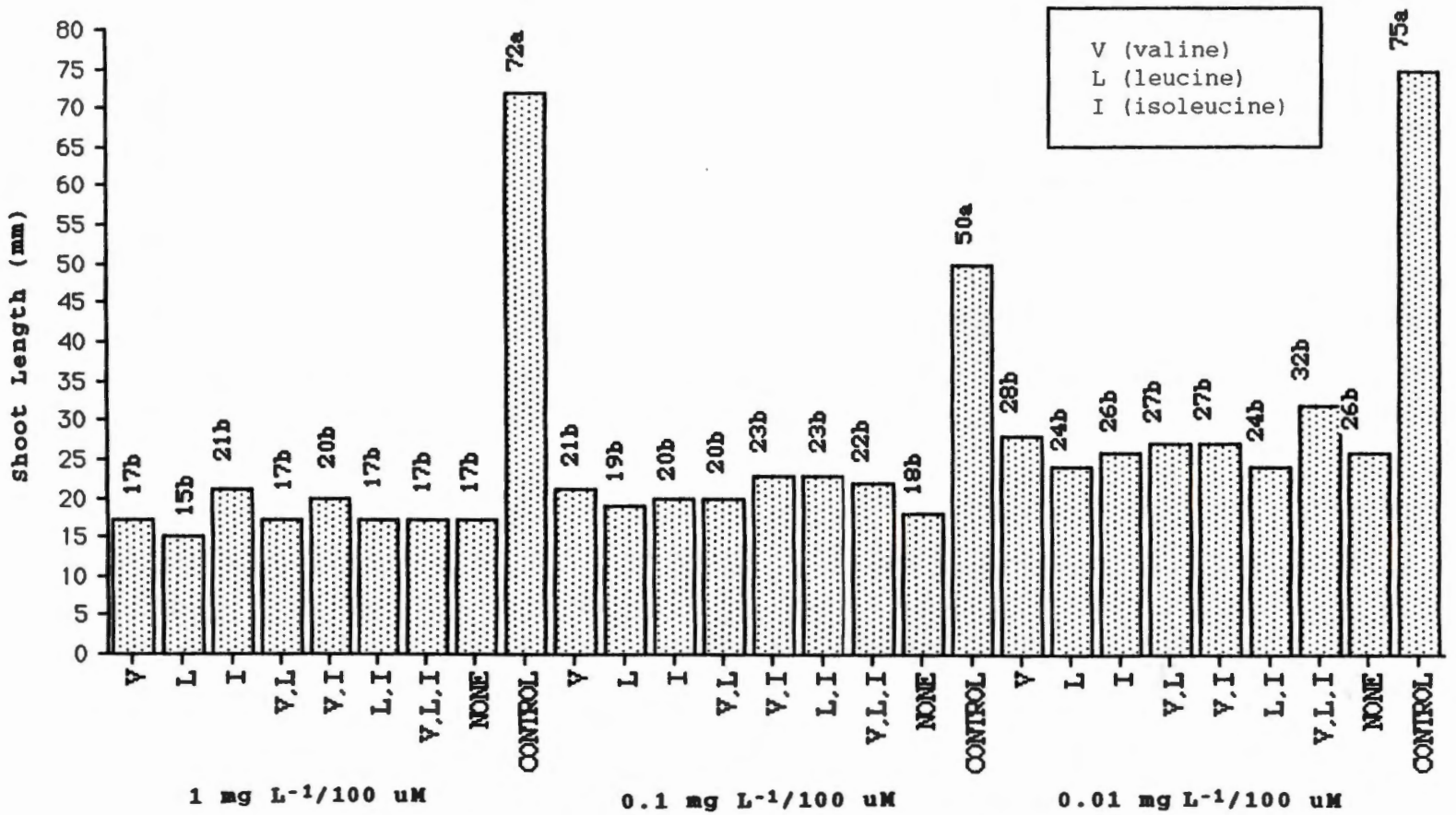

Figure 23. Activity of thifensulfuron-methyl at 1, 0.1 and 0.01 mg L^{-1} on aerial bulblet shoot growth as affected by the amino acids valine, leucine and isoleucine at 100 uM. Values are combined treatment means from conducting a minimum of two trials for the experiment. Means followed by the same letter do not differ at the 0.05 level according to Duncan's Multiple Range Test.

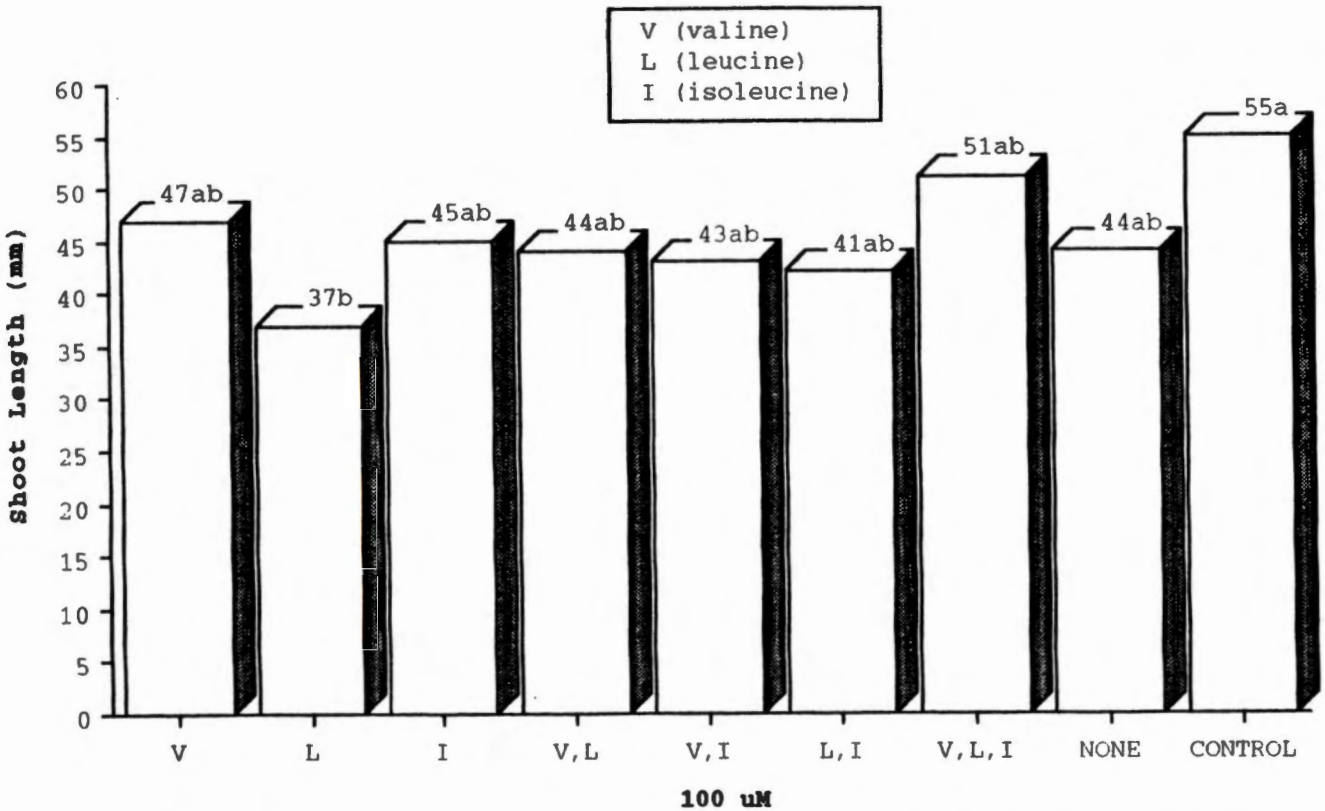

Figure 24. Activity of thifensulfuron-methyl at 0.001 mg L^{-1} on aerial bulblet shoot growth as affected by the amino acids valine, leucine and isoleucine. Values are combined treatment means from conducting a minimum of two trials for the experiment. Means followed by the same letter do not differ at the 0.05 level according to Duncan's Multiple Range Test.

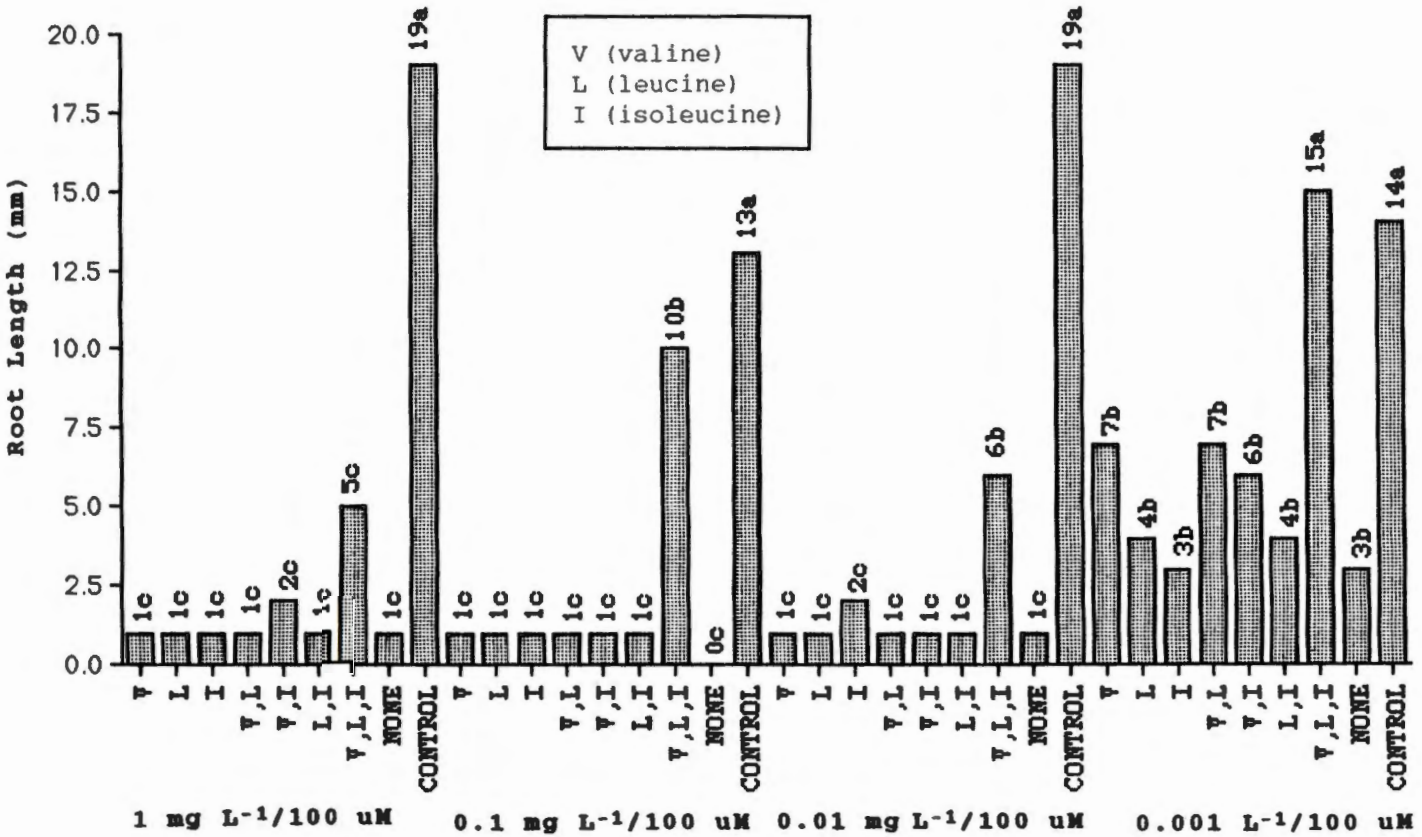

Figure 25. Activity of thifensulfuron-methyl at 1, 0.1, 0.01 and 0.001 mg L^{-1} on aerial bulblet root growth as affected by the amino acids valine, leucine and isoleucine at 100 uM. Values are combined treatment means from conducting a minimum of two trials for the experiment. Means followed by the same letter do not differ at the 0.05 level according to Duncan's Multiple Range Test.

Previous research combining 100 uM valine and leucine with chlorsulfuron at 28 nM was ineffective in reversing chlorsulfuron activity on peas and maize roots (Giardina et al. 1987).

Response of Soft Offset Bulbs to Thifensulfuron-Methyl

Thifensulfuron-methyl inhibited soft offset bulb shoot elongation over 10 d of evaluation (Figure 26). Shoot growth following 2 and 5 d indicated inhibition at all concentrations of thifensulfuron-methyl (Figure 26). After 10 d of exposure, soft offset bulb shoot growth was inhibited by all concentrations of thifensulfuron-methyl except 0.001 mg L^{-1} (Figure 26). It is possible that endogenous levels of amino acids in soft offset bulbs are sufficient to reverse the effects of thifensulfuron-methyl at 0.001 mg L^{-1}.

Root growth from soft offset bulbs was not present following 2 d incubation (data not shown). All concentrations of thifensulfuron-methyl suppressed root growth at 5 d with complete suppression at 10 d except at 0.001 mg L^{-1}. (Figure 27). These data indicate that suppression of root growth from soft offset bulbs was comparable to root growth of aerial bulblets. It appears that the root meristem is an active site for thifensulfuron-methyl in both soft offset bulbs and aerial bulblets of wild garlic. This observation was noted for soft offset bulbs treated with thifensulfuron-methyl in greenhouse experiments.

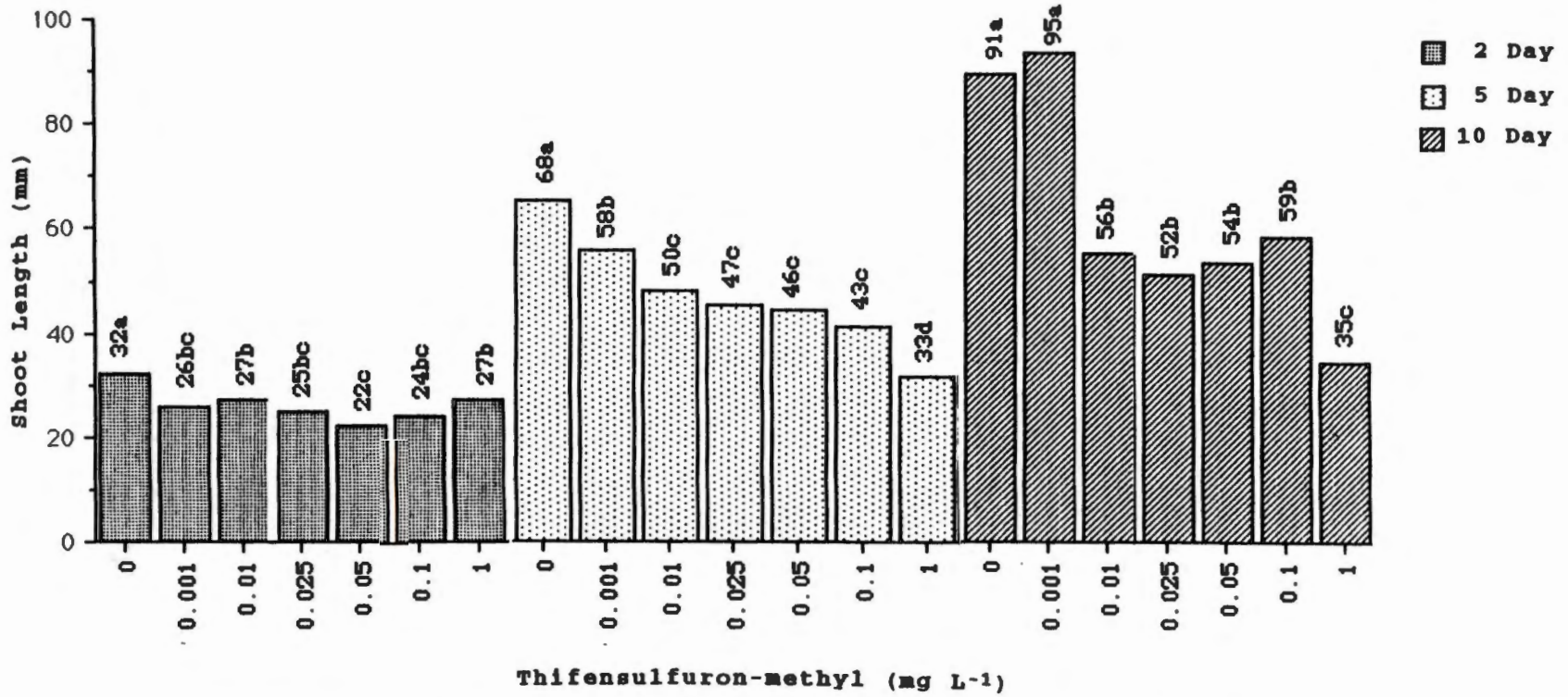

Figure 26. Effect of thifensulfuron-methyl after 2, 5 and 10 d on shoot growth of soft offset bulbs. Values are combined treatment means from conducting a minimum of two trials for the experiment. Means followed by the same letter within the same elongation period do not differ at 0.05 level according to Duncan's Multiple Range Test.

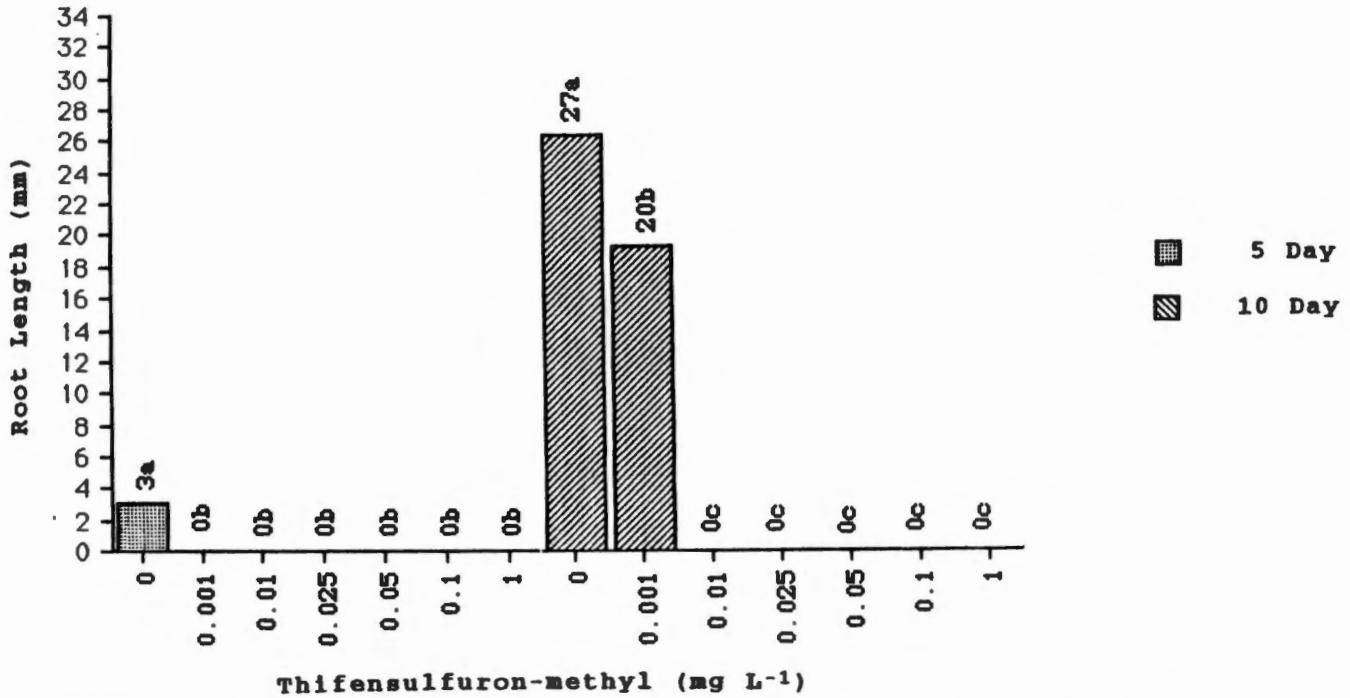

Figure 27. Effect of thifensulfuron-methyl after 5 and 10 d on root growth of soft offset bulbs. Values are combined treatment means from conducting a minimum of two trials for the experiment. Means followed by the same letter within the same elongation period do not differ at 0.05 level according to Duncan's Multiple Range Test.

Evidence for meristematic activity of thifensulfuron-methyl is shown in the photomicrographs of treated soft offset bulbs (Figures 28 through 34). Normal meristem development was observed after 10 d for soft offset bulbs treated with thifensulfuron-methyl at rates of 1 to 25 ppb (Figures 29 through 31). Thifensulfuron-methyl at 0.5 to 1 ppm had cells lacking nuclei and the leaf tissue was necrotic (Figures 32 through 34). Thifensulfuron-methyl reduced drastically the shoot growth of soft offset bulbs (Figure 26). Necrotic leaf and shoot tissue as well as viable leaf bud and meristem were present in a wild garlic bulb treated with 1 ppm thifensulfuron-methyl for 10 d (Figure 34). In greenhouse studies soft offset bulbs treated with 35 g ai ha^{-1} thifensulfuron-methyl were not totally necrotic after 30 d. Had the experiments been of longer duration, complete death of all meristematic areas in the bulb may have occurred. This should be a consideration in any future research with thifensulfuron-methyl on soft offset bulbs and other bulb types of wild garlic.

Inhibition of Thifensulfuron-Methyl Activity on Soft Offset Bulbs as Influenced by Valine, Leucine and Isoleucine

Activity of 0.001 and 0.01 mg L^{-1} thifensulfuron-methyl on shoot growth of soft offset bulbs after 2 d was partially nullified by the amino acids valine, leucine and isoleucine (Figure 35).

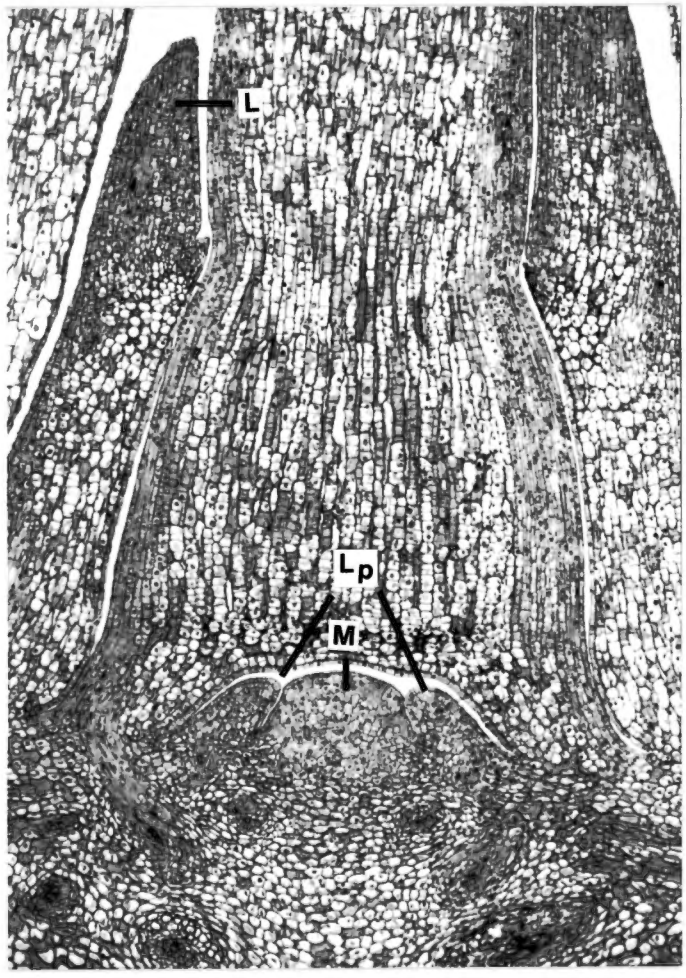

Figure 28. Photomicrograph of an untreated wild garlic bulb meristem following 10 d in sterile culture. A meristematic dome (M) is indicated as are leaf primordia (Lp) at the periphery of the meristem. An elongating leaf (L) is also shown. Magnification is 40X.

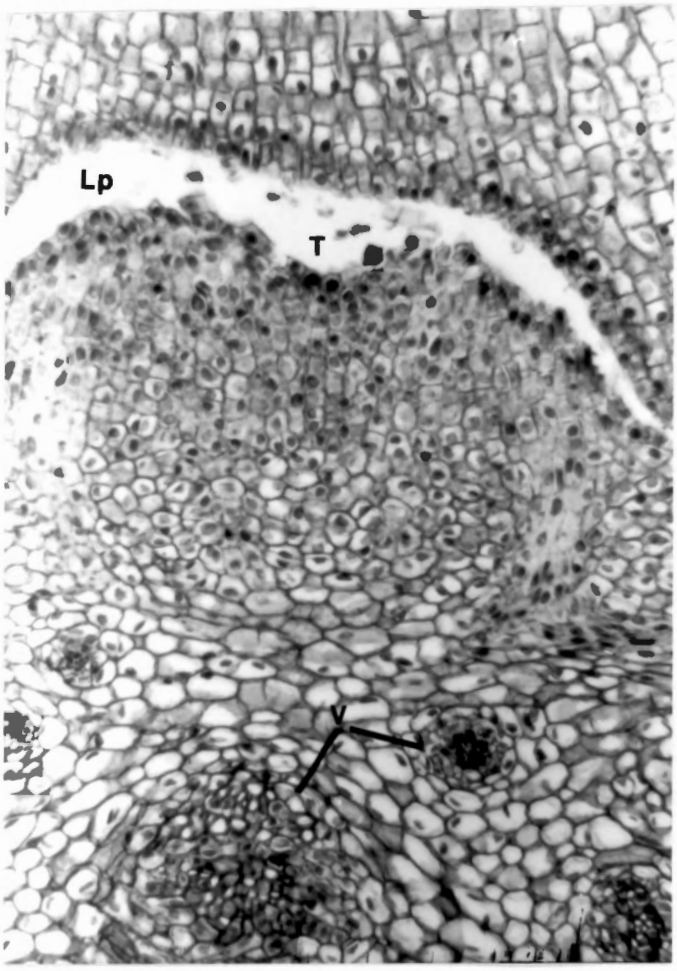

Figure 29. Photomicrograph of wild garlic bulb meristem following 10 d in sterile culture treated with 1 ppb thifensulfuron-methyl. A tunica layer (T) and leaf primordia (Lp) are shown. Vascular bundles (V) are basal to the meristem. Magnification is 63X.

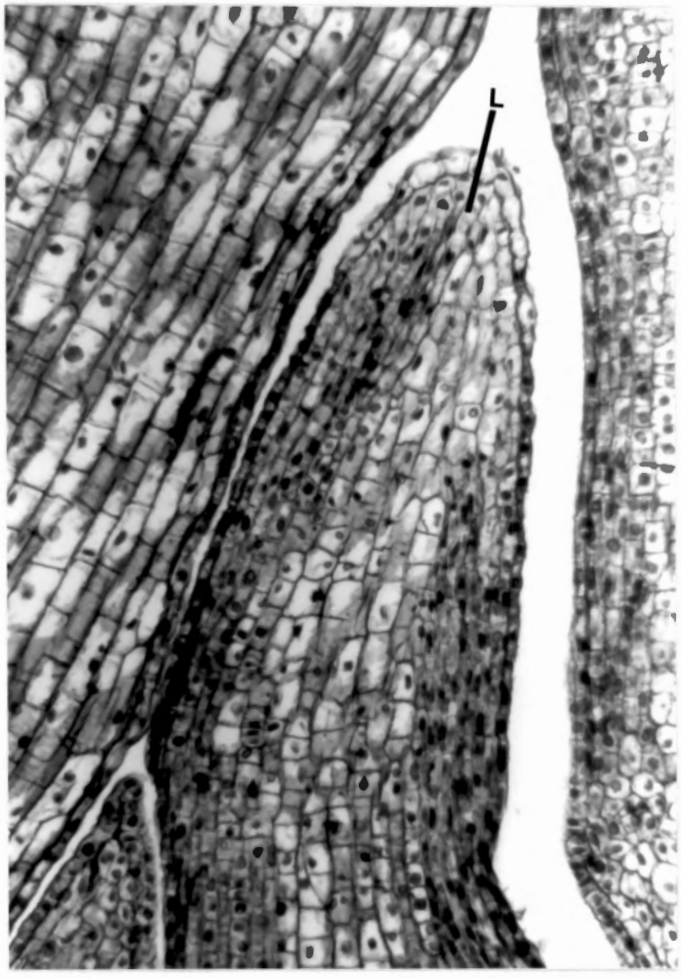

Figure 30. Photomicrograph of wild garlic bulb meristem following 10 d in sterile culture treated with 10 ppb thifensulfuron-methyl. A growing leaf (L) is shown with most cells containing a nucleus. Magnification is 63X.

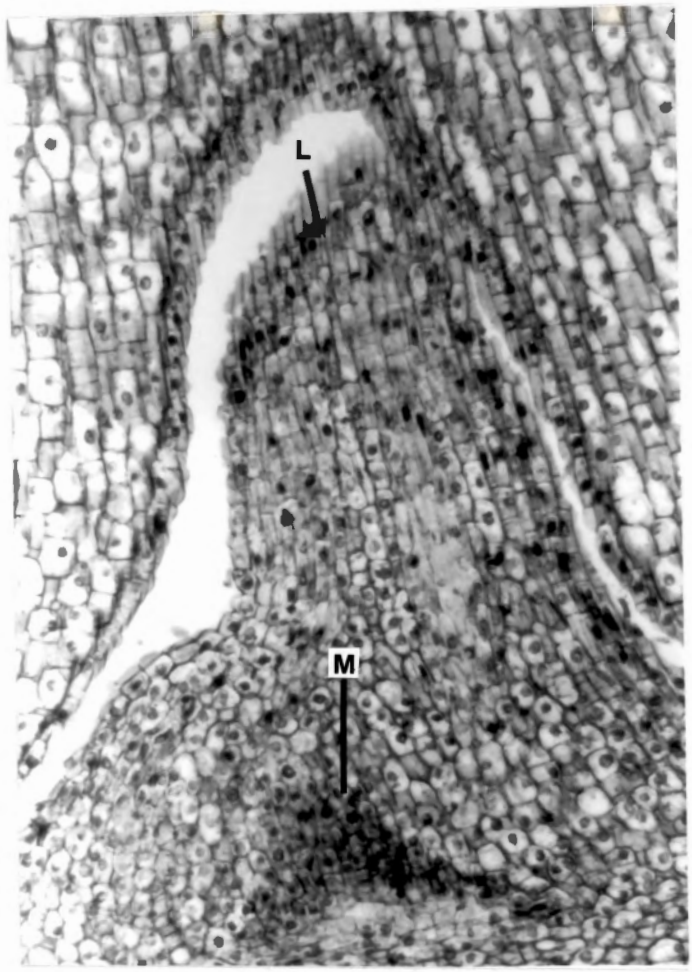

Figure 31. Photomicrograph of wild garlic bulb meristem following 10 d in sterile culture treated with 25 ppb thifensulfuron-methyl. Area of meristematic activity (M) is noted by heavy staining at base of leaf (L). Magnification is 63X.



Figure 32. Photomicrograph of wild garlic bulb meristem following 10 d in sterile culture treated with 50 ppb thifensulfuron-methyl. Leaf shoot with cells devoid of a nucleus (Nn). Magnification is 63X.

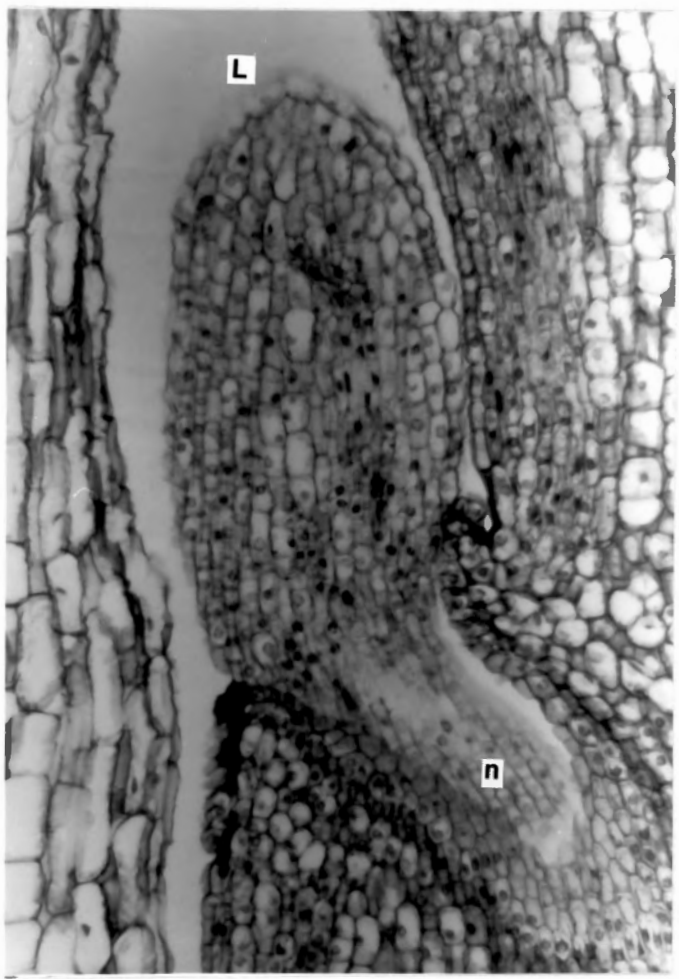

Figure 33. Photomicrograph of wild garlic bulb meristem following 10 d in sterile culture treated with 100 ppb thifensulfuron-methyl. An elongating leaf (L) with necrotic area (n) at base is shown. Magnification is 63X.

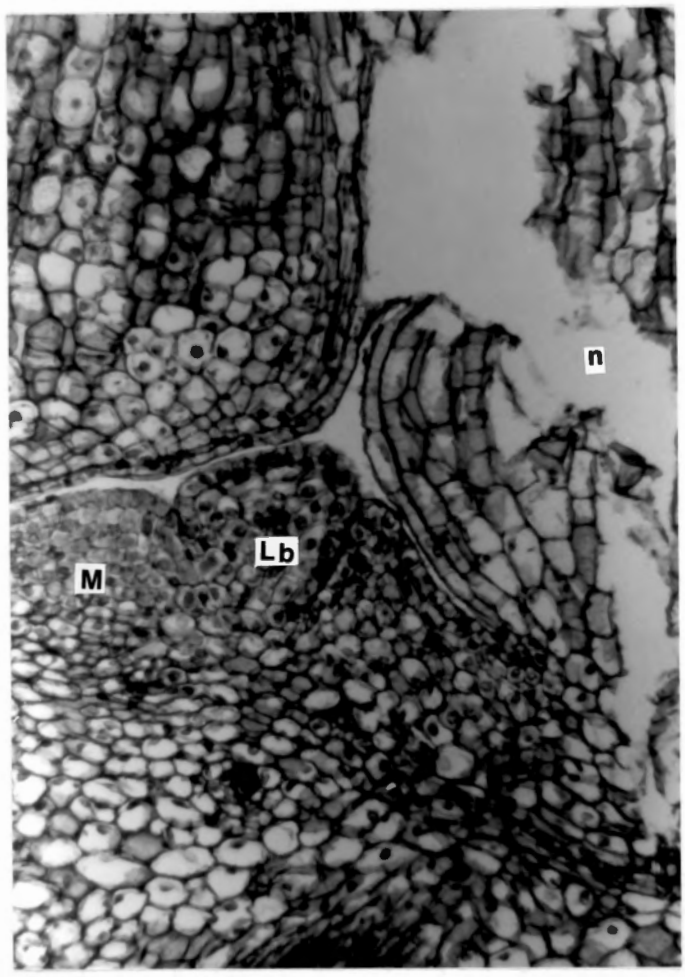

Figure 34. Photomicrograph of wild garlic bulb meristem following 10 d in sterile culture treated with 1 ppm thifensulfuron-methyl. Necrotic area (n) of meristem, the meristem (M) and leaf bud (Lb), not actively growing but still viable with nucleated cells, are shown. Magnification is 63X.

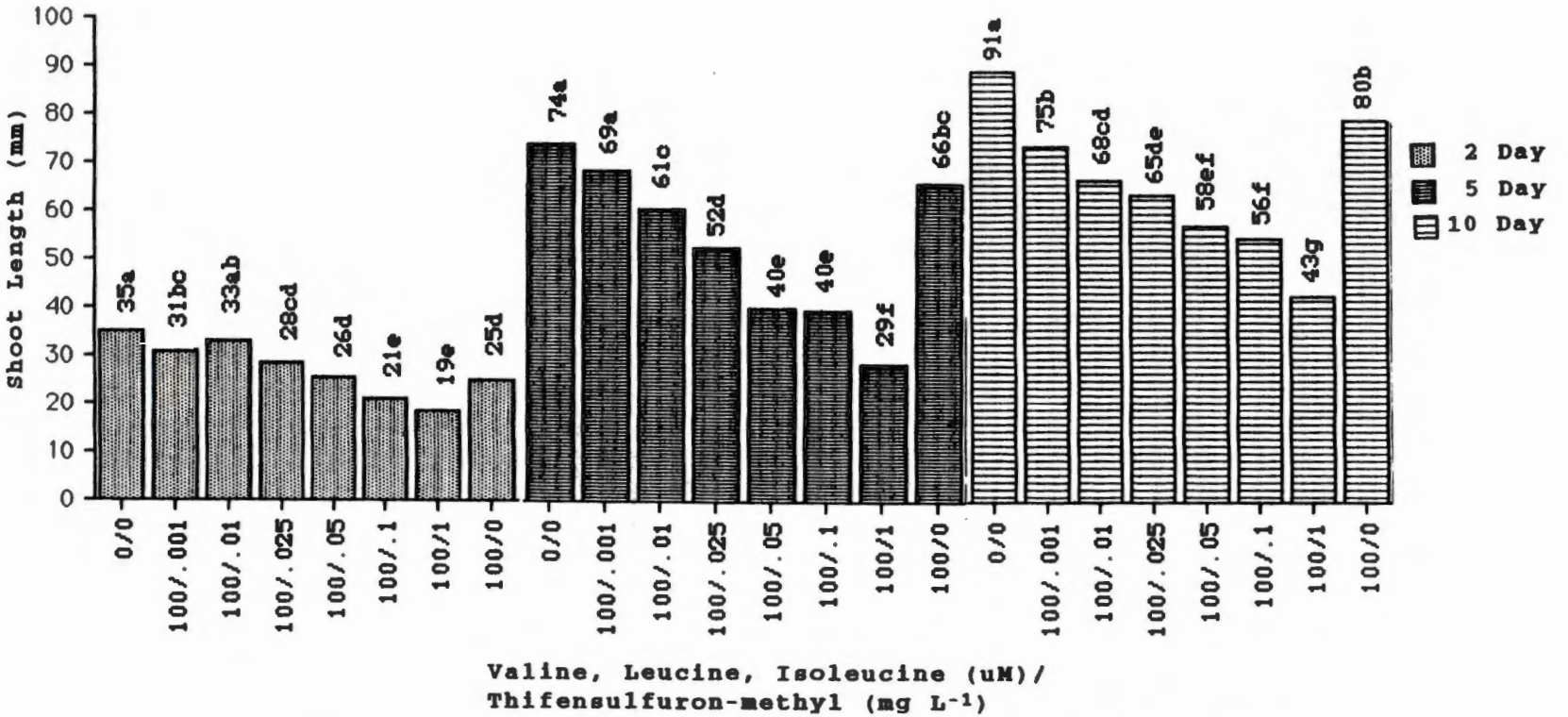

Figure 35. Effect of valine, leucine and isoleucine on the activity of thifensulfuron-methyl after 2, 5 and 10 d on shoot growth of soft offset bulbs. Values are combined treatment means from conducting a minimum of two trials for the experiment. Means followed by the same letter within the same elongation period do not differ at the 0.05 level according to Duncan's Multiple Range Test.

Following 5 d of exposure to thifensulfuron-methyl, soft offset bulbs had a greater response to the influence of amino acids. The influence of increasing rates of thifensulfuron-methyl on shoot growth was not as pronounced at 5 d as at 2 d. Valine, leucine and isoleucine at 100 uM partially reversed thifensulfuron-methyl activity from 0.001 to 0.1 mg L^{-1}. The reversal was greatest for the lowest rates of thifensulfuron-methyl. With 10 d exposure, reduction of shoot growth in soft offset bulbs was still evident. Valine, leucine and isoleucine at 100 uM could not totally nullify thifensulfuron-methyl activity.

There appeared to be a rapid increase in shoot growth through 5 d compared to the 2 d exposure. Increased rate of growth following 10 d exposure to all rates of thifensulfuron-methyl plus 100 uM valine, leucine and isoleucine was not as great as the 5 d evaluation. A trend appears for a decreasing effect on shoot growth of thifensulfuron-methyl at 0.001 to 0.1 mg L^{-1} through 10 d. This trend might be more pronounced if the duration of exposure was increased. Bulbs receiving 100 uM amino acids alone grew slower than the control, possibly indicating some adverse effect on the meristem. Single treatments of 15 amino acids at 1 or 2 mM inhibited fresh weight development of barley embryos (Miflin 1969).

There was no root growth for soft offset bulbs with any treatment including the control at the 2 d evaluation (data not shown).

Following 5 d of exposure only the control had root growth (Figure 36). Valine, leucine and isoleucine did not suppress the activity of any rate of thifensulfuron-methyl. After 10 d the amino acids alleviated activity of thifensulfuron-methyl at concentrations of 0.001 to 0.025 mg L^{-1}. Suppression of root growth by thifensulfuron-methyl at 0.5 to 1 mg L^{-1} was apparent. The possible inhibitory effect on shoot growth by the amino acids alone was not observed for root growth. Root growth length was in the same range as that of the untreated control.

Chlorsulfuron inhibition of root and shoot development has been noted by several researchers (Ray 1982,1984; Landi et al.1989; Rost and Reynolds 1985; Giardina et al. 1987; DeAgazio and Giardina. 1984). Chlorsulfuron at 1 ppb inhibited root growth in corn seedlings (Ray 1982). Since plant growth consists of cell expansion and cell division, Ray's research determined that chlorsulfuron had little effect on cell expansion but inhibited cell division. This was evident in photomicrographs of soft offset bulbs where cells were absent of nuclei (Figure 32). Alfalfa (Medicago sativa L. var. 'Vertus') growing in plant medium with 0.28 pM chlorsulfuron had deformed root hairs (Martensson and Nilsson 1989). Root hair deformation led to inhibition of nodule formation.

Valine and isoleucine at 100 uM failed to reduce the activity of 0.28 uM chlorsulfuron on maize shoots and roots (Giardina et al. 1987).

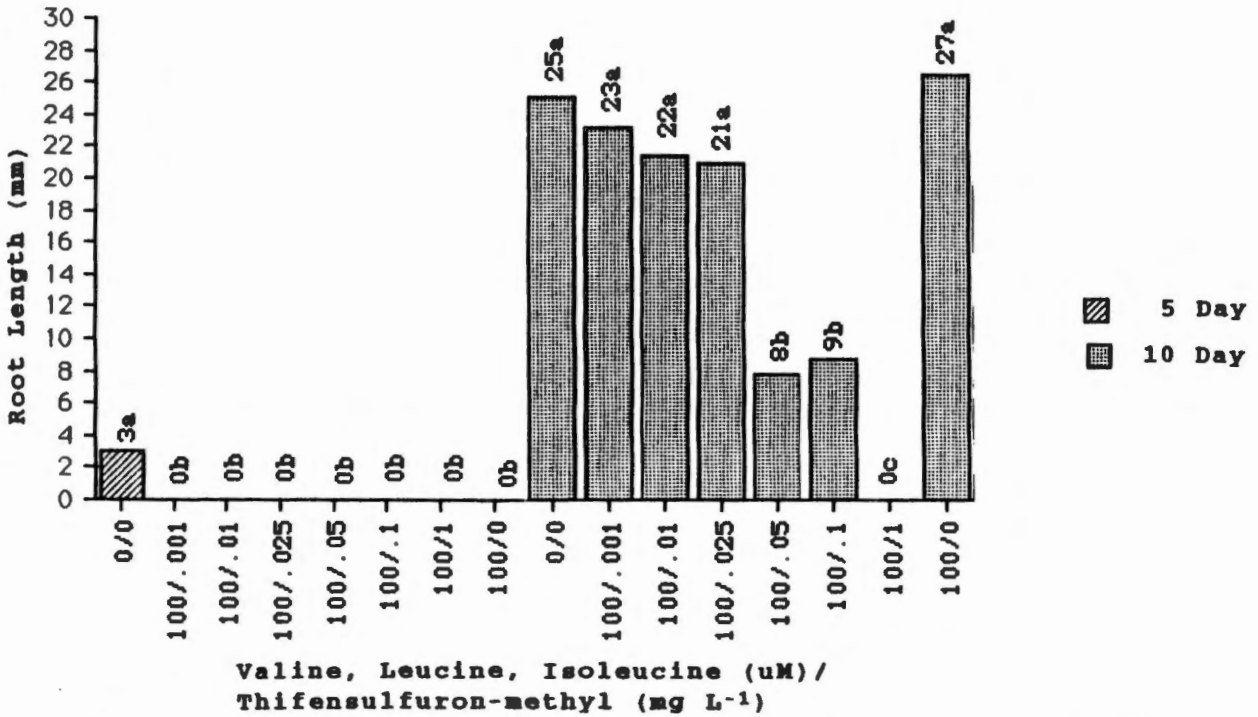

Figure 36. Effect of valine, leucine and isoleucine on the activity of thifensulfuron-methyl after 5 and 10 d on root growth of soft offset bulbs. Values are combined treatment means from conducting a minimum of two trials for the experiment. Means followed by the same letter within the same elongation period do not differ at the 0.05 level according to Duncan's Multiple Range Test.

The inhibition of corn root elongation by 28 nM chlorsulfuron was only partially alleviated by 1 mM valine, leucine or isoleucine (Rubin and Casida 1985). Exogenous levels of valine, leucine and isoleucine have been examined with the sulfonylurea herbicide chlorsulfuron and the imidazolinone herbicides (Rubin and Casida 1985; Anderson and Hibberd 1985). Growth inhibition of maize cell suspension culture by the imidazolinone herbicide AC 243 997 at 10 nM to 1mM was alleviated by the addition of 1 mM valine, leucine and isoleucine (Anderson and Hibberd 1985).

Photomicrographs of soft offset bulbs treated with thifensulfuron-methyl and 100 uM valine, leucine and isoleucine for 10 d indicated that amino acids partially nullified the activity of thifensulfuron-methyl (Figures 37 through 43). Normal cellular development was noted where 100 uM valine, leucine and isoleucine were added to thifensulfuron-methyl rates of 0.001 to 0.50 mg L^{-1}. Bulbs treated with 0.1 to 1 mg L^{-1} thifensulfuron-methyl plus 100 uM of the amino acids had cells without a nuclei as well as necrotic leaf tissue.

The growth of shoots and roots of soft offset bulbs supplemented with valine, leucine and isoleucine could not totally overcome the effects of thifensulfuron-methyl. The protective effects of the amino acids cannot be explained on the grounds of growth stimulation since the controls grown with the amino acids did not differ from those without amino acids.

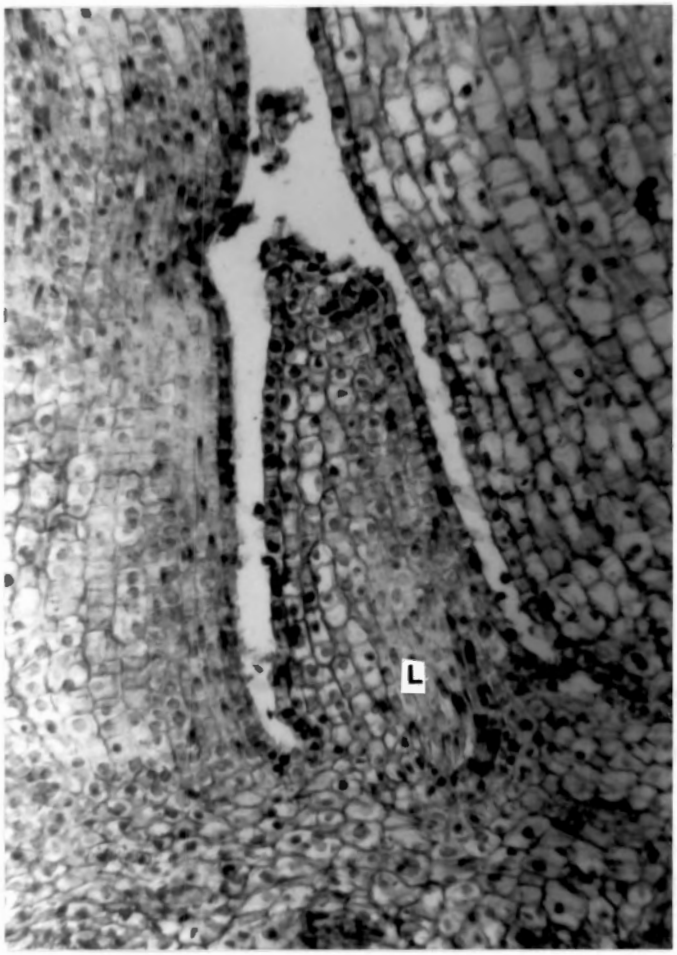

Figure 37. Photomicrograph of a wild garlic bulb meristem after 10 d in sterile culture treated with 100 uM valine, leucine and isoleucine. Normal leaf (L) development is indicated. Individual cells contain nuclei as indicated by the level of staining. Magnification is 63X.

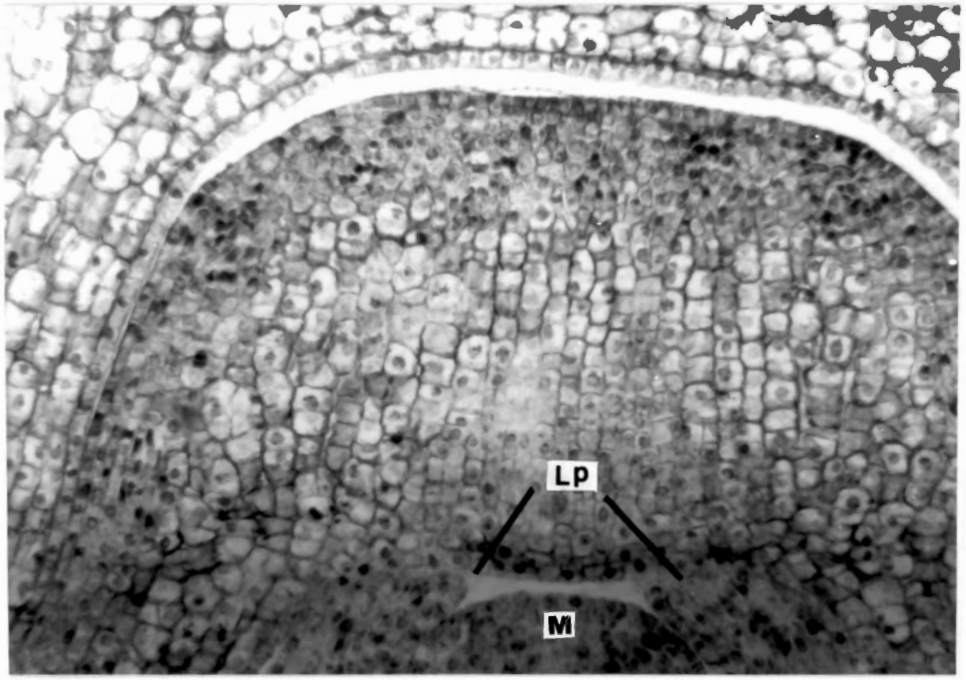

Figure 38. Photomicrograph of a wild garlic bulb meristem after 10 d in sterile culture treated with 100 uM valine, leucine and isoleucine plus 1 ppb thifensulfuron-methyl. Meristem (M) with leaf primordia (Lp) shows normal development of cells. Magnification is 63X.

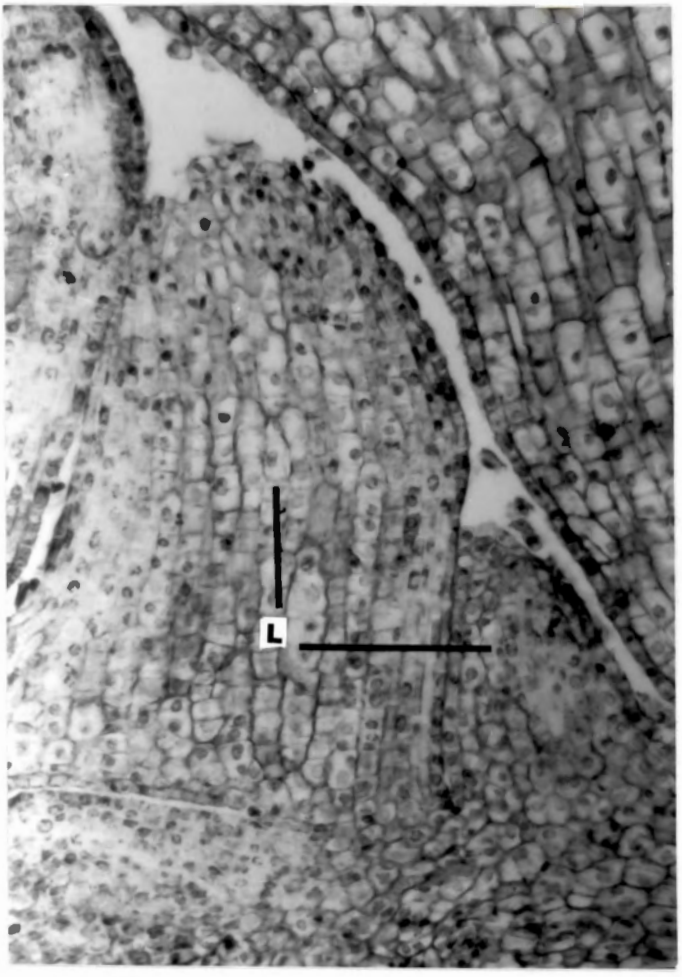

Figure 39. Photomicrograph of a wild garlic bulb meristem after 10 d in sterile culture treated with 100 uM valine, leucine and isoleucine plus 10 ppb thifensulfuron-methyl. Leaf development (L) is normal and not affected by thifensulfuron-methyl. Magnification is 63X.

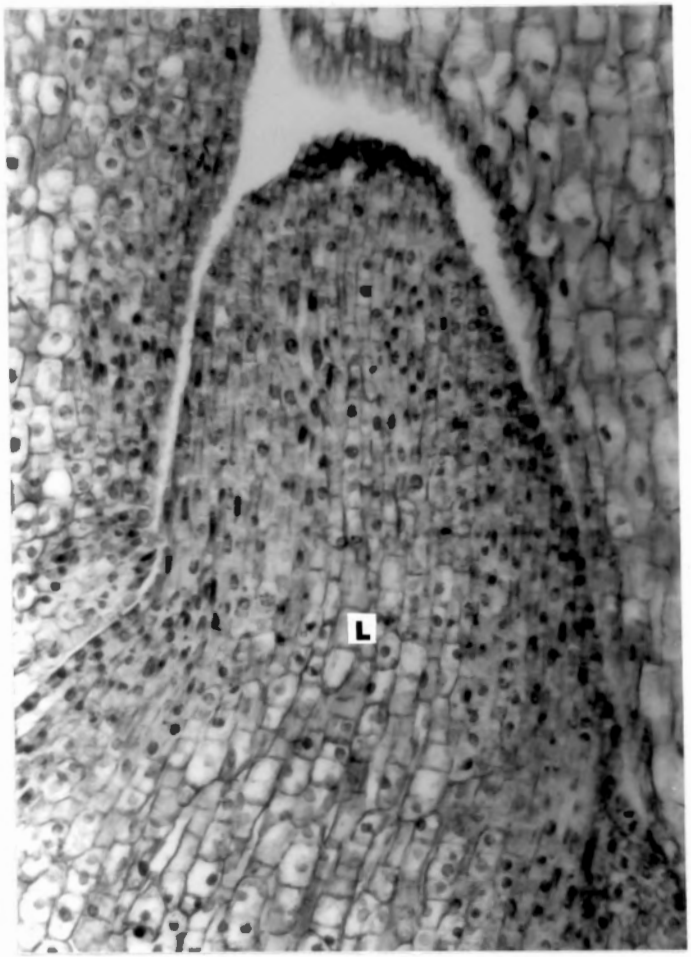

Figure 40. Photomicrograph of a wild garlic bulb meristem after 10 d in sterile culture treated with 100 uM valine, leucine and isoleucine plus 25 ppb thifensulfuron-methyl. Leaf development (L) is normal. Magnification is 63X.

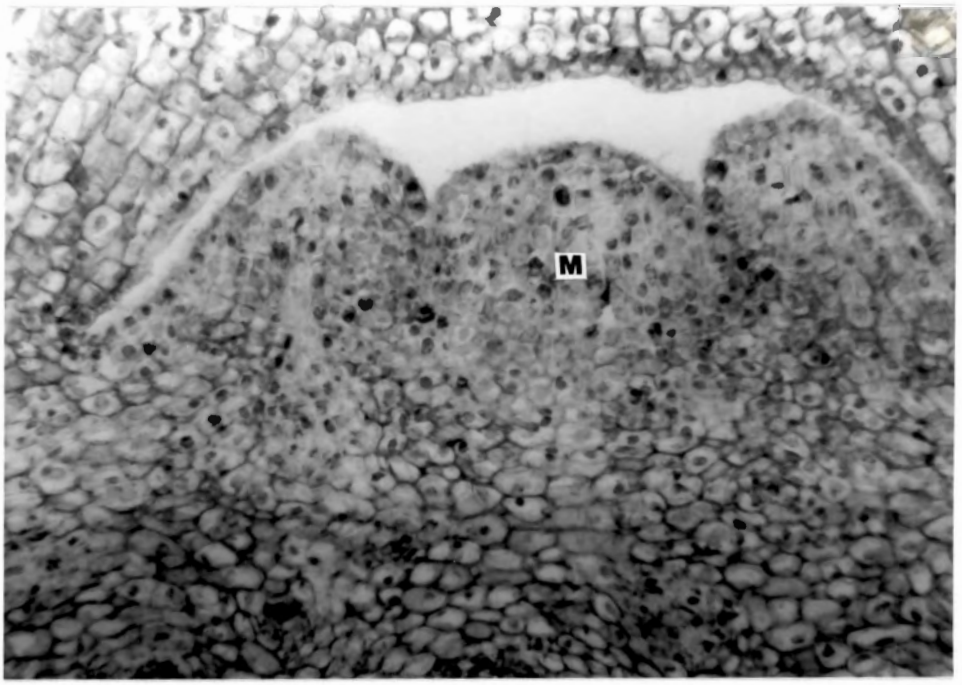

Figure 41. Photomicrograph of a wild garlic bulb meristem after 10 d in sterile culture treated with 100 uM valine, leucine and isoleucine plus 50 ppb thifensulfuron-methyl. Meristem (M)shown with a leaf primordia. Magnification is 63X .

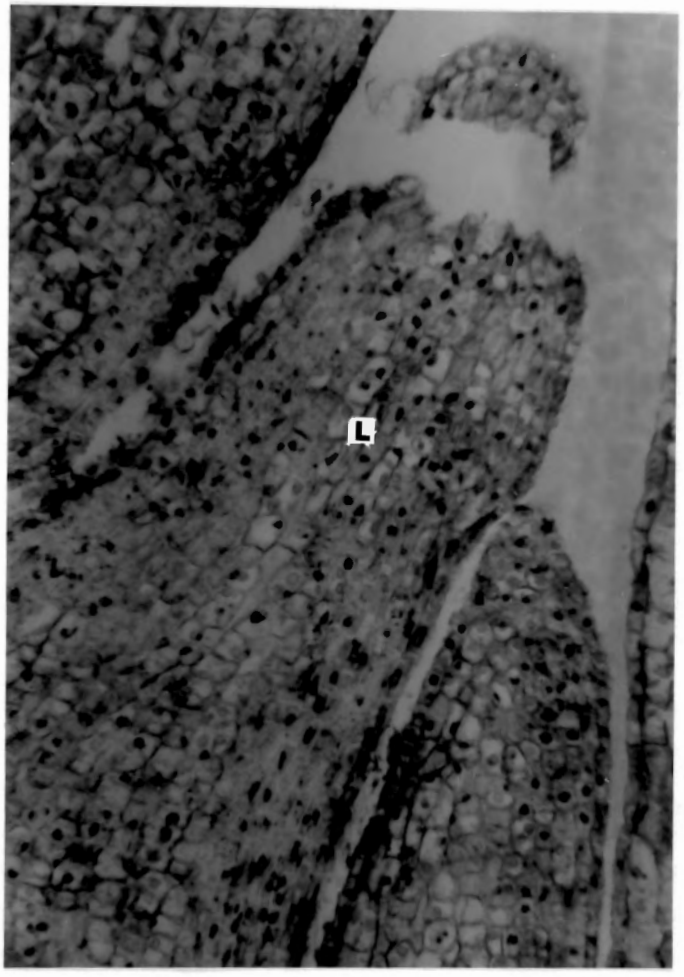

Figure 42. Photomicrograph of a wild garlic bulb meristem after 10 d in sterile culture treated with 100 uM valine, leucine and isoleucine plus 100 ppb thifensulfuron-methyl. Necrosis of leaf (L) and cells lacking nuclei are shown. Magnification is 63X.

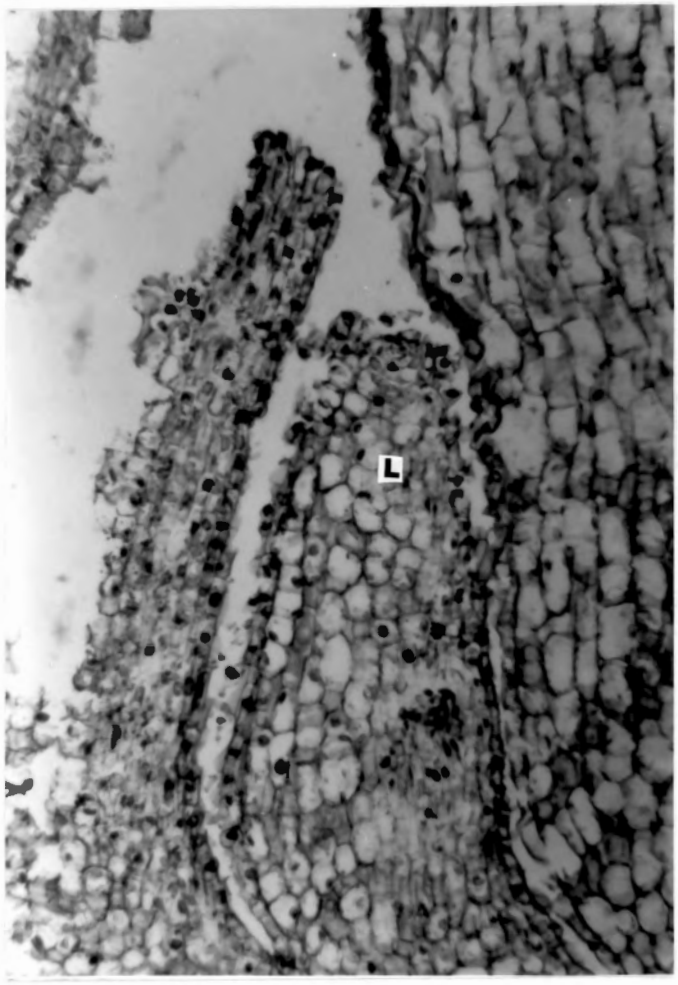

Figure 43. Photomicrograph of a wild garlic bulb meristem after 10 d in sterile culture treated with 100 uM valine, leucine and isoleucine plus 1 ppm thifensulfuron-methyl. Necrotic tissue is evident in the developing leaf (L). Magnification is 63X.

It is possible that more than one mechanism of action may exist for the sulfonylurea herbicides as proposed for chlorsulfuron (Giardina et al. 1987).

A decrease in export of ^{14}C activity by chlorsulfuron treated leaves of field pennycress (Thlaspi arvense L.) indicates that chlorsulfuron has an effect on assimilate translocation (Bestman et al. 1990). Further work showed that intact seedlings treated with chlorsulfuron plus L-valine, L-leucine and L-isoleucine had little or no effect in decreasing assimilate transport.

The evidence is not conclusive as to the role of supplemental branched chain amino acids in alleviating the activity of sulfonylurea herbicides. Pool size of the branched chain amino acids as well as tissue type may influence the response to sulfonylurea herbicides. Data are not available describing levels of branch chain amino acids in wild garlic bulb types or whole plants. Since soft offset bulbs were actively growing prior to preparation and subsequent exposure to thifensulfuron-methyl plus branched chain amino acids, the pool size of these amino acids may have decreased. The addition of these amino acids coupled with the lower concentrations of thifensulfuron-methyl may have permitted growth to continue through export of assimilates from the bulb. Where concentrations of thifensulfuron-methyl were highest the assumption is that assimilates were not actively transported due to inadequate levels of branched chain amino acids both endogenously and exogenously.

In the literature there appear to be differences as to minimal dosage of sulfonylurea herbicides required to produce inhibition of growth in susceptible species. Further research is needed to determine the endogenous levels of the amino acid pool of wild garlic and how this may influence herbicide response. Experimentation into dose by time by amino acid level could give further insight into the translocation properties of thifensulfuron-methyl. Of interest would be pretreatment of bulbs with amino acids for varying times and then exposing the bulbs to thifensulfuron-methyl.

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APPENDIXES

APPENDIX A

TABULAR DATA FOR SEQUENTIAL APPLICATIONS OF THIFENSULFURON-METHYL FOLLOWED BY METRIBUZIN AND METRIBUZIN FOLLOWED BY THIFENSULFURON-METHYL IN 12 AND 24 H ON WILD GARLIC AT SPRING HILL 1987 AND 1988

Table 22. Reduction in wild garlic whole plant growth from thifensulfuron-methyl followed in 12 h by metribuzin at Spring Hill in 1987[a].

Treatment[c]	Herbicide rate	12 h Application Timing			
		25 DAT[b]		50 DAT	
		Level of Reduction			
		Regression Estimate	P>|T|	Regression Estimate	P>|T|
	g ai ha^{-1}				
Intercept (control)	0	0.0000	1.0000	0.0000	1.0000
Thifensulfuron + Metribuzin	4 210	0.0625	0.7519	0.7152	0.0002
Thifensulfuron + Metribuzin	4 420	0.1956	0.3268	0.9447	0.0001
Thifensulfuron + Metribuzin	4 630	0.3166	0.1176	0.5895	0.0014
Thifensulfuron + Metribuzin	9 210	0.0500	0.8004	1.0115	0.0001
Thifensulfuron + Metribuzin	9 420	0.3508	0.0845	1.2532	0.0001
Thifensulfuron + Metribuzin	9 630	0.3633	0.0746	0.8437	0.0001
Thifensulfuron + Metribuzin	17.5 210	0.0625	0.7519	1.2532	0.0001
Thifensulfuron + Metribuzin	17.5 420	0.0375	0.8496	1.2198	0.0001
Thifensulfuron + Metribuzin	17.5 630	0.3174	0.1168	1.1865	0.0001

[a]$P<0.05$ indicates significance and antagonism of thifensulfuron methyl by metribuzin.
[b]days after treatment.
[c]Thifensulfuron and thifensulfuron-methyl are the same.

Table 23. Reduction in wild garlic whole plant growth from thifensulfuron-methyl followed in 24 h by metribuzin at Spring Hill in 1987[a].

Treatment[c]	Herbicide rate	24 h Application Timing			
		25 DAT[b]		50 DAT	
		Level of Reduction			
		Regression Estimate	P>|T|	Regression Estimate	P>|T|
	g ai ha^{-1}				
Intercept (control)	0	0.0000	1.0000	0.0000	1.0000
Thifensulfuron + Metribuzin	4 210	0.0751	0.6723	1.1316	0.0001
Thifensulfuron + Metribuzin	4 420	0.1935	0.2805	0.4675	0.0137
Thifensulfuron + Metribuzin	4 630	0.0625	0.7247	0.2678	0.1427
Thifensulfuron + Metribuzin	9 210	0.3383	0.0649	1.2119	0.0001
Thifensulfuron + Metribuzin	9 420	0.2313	0.1991	0.9332	0.0001
Thifensulfuron + Metribuzin	9 630	0.0625	0.7247	0.8332	0.0001
Thifensulfuron + Metribuzin	17.5 210	0.0500	0.7781	1.2992	0.0001
Thifensulfuron + Metribuzin	17.5 420	0.3299	0.0713	1.0938	0.0001
Thifensulfuron + Metribuzin	17.5 630	0.2944	0.1055	1.1717	0.0001

[a]P<0.05 indicates significance and antagonism of thifensulfuron-methyl by metribuzin.
[b]days after treatment.
[c]Thifensulfuron and thifensulfuron-methyl are the same.

Table 24. Reduction in wild garlic aerial bulblet weight from thifensulfuron-methyl followed in 12 and 24 h by metribuzin at Spring Hill in 1987[a].

Treatment[b]	Herbicide rate	Application Timing					
		12 h			24 h		
		Regression Estimate	Expected Bulblet Weight	P>|T|	Regression Estimate	Expected Bulblet Weight	P>|T|
	g ai ha^{-1}		g			g	
Intercept (control)	0	2.0475	2.0	0.0564	5.4075	5.5	0.0001
Thifensulfuron + Metribuzin	4 210	0.6300	2.7	0.6679	-4.1700	1.2	0.0002
Thifensulfuron + Metribuzin	4 420	-1.7325	0.3	0.2433	-2.6925	2.7	0.0095
Thifensulfuron + Metribuzin	17.5 630	-2.0175	0.0	0.1761	-3.8300	1.6	0.0005
Thifensulfuron + Metribuzin	9 210	-1.9550	0.1	0.1895	-5.1700	0.2	0.0001
Thifensulfuron + Metribuzin	9 420	-1.7700	0.3	0.2335	-5.4075	0.0	0.0001
Thifensulfuron + Metribuzin	9 630	-2.0475	0.0	0.1700	-4.9425	0.5	0.0001
Thifensulfuron + Metribuzin	17.5 210	-2.0475	0.0	0.1700	-5.1950	0.2	0.0001
Thifensulfuron + Metribuzin	17.5 420	-2.4705	0.0	0.1700	-5.4075	0.0	0.0001
Thifensulfuron + Metribuzin	17.5 630	-2.4705	0.0	0.1700	-5.4075	0.0	0.0001

[a]$P<0.05$ indicates significance and antagonism of thifensulfuron-methyl by metribuzin.
[b]Thifensulfuron and thifensulfuron-methyl are the same.

Table 25. Reduction in wild garlic underground bulb weight from thifensulfuron-methyl followed in 12 and 24 h by metribuzin at Spring Hill in 1987[a].

Treatment[b]	Herbicide rate	Application Timing					
		12 h			24 h		
		Regression Estimate	Expected Bulb Weight	P>|T|	Regression Estimate	Expected Bulb Weight	P>|T|
	g ai ha^{-1}		g			g	
Intercept (control)	0	5.1225	5.1	0.0000	5.2100	5.2	0.0001
Thifensulfuron + Metribuzin	4 210	4.1375	9.3	0.0027	1.5675	6.8	0.2093
Thifensulfuron + Metribuzin	4 420	0.9525	6.1	0.4545	0.1875	5.4	0.8789
Thifensulfuron + Metribuzin	4 630	1.1100	6.2	0.3843	2.1675	7.4	0.0866
Thifensulfuron + Metribuzin	9 210	1.9875	7.1	0.1249	1.5150	6.7	0.2245
Thifensulfuron + Metribuzin	9 420	-0.6800	4.4	0.5924	0.7275	5.9	0.5555
Thifensulfuron + Metribuzin	9 630	2.4450	7.6	0.0619	0.4150	5.6	0.7361
Thifensulfuron + Metribuzin	17.5 210	1.5950	6.7	0.2146	0.9900	6.2	0.4237
Thifensulfuron + Metribuzin	17.5 420	1.3700	6.5	0.2847	-0.4225	4.8	0.7315
Thifensulfuron + Metribuzin	17.5 630	-0.9450	4.2	0.4580	-0.7575	3.5	0.1608

[a]P<0.05 indicates significance and antagonism of thifensulfuron-methyl by metribuzin.
[b]Thifensulfuron and thifensulfuron-methyl are the same.

Table 26. Reduction in wild garlic whole plant growth from thifensulfuron-methyl followed in 12 and 24 h by metribuzin at Spring Hill in 1988[a].

Treatment[c]	Herbicide rate	12 h Application Timing		24 h Application Timing	
		44 DAT[b]		44 DAT	
		Level of Reduction			
		Regression Estimate	P>|T|	Regression Estimate	P>|T|
	g ai ha^{-1}				
Intercept (control)	0	0.0000	1.0000	0.0000	1.0000
Thifensulfuron + Metribuzin	4 210	1.1050	0.0001	1.2198	0.0001
Thifensulfuron + Metribuzin	4 420	1.0852	0.0001	1.1185	0.0001
Thifensulfuron + Metribuzin	4 630	1.2532	0.0001	1.1865	0.0001
Thifensulfuron + Metribuzin	9 210	1.2198	0.0001	1.1605	0.0001
Thifensulfuron + Metribuzin	9 420	1.0457	0.0001	1.1124	0.0001
Thifensulfuron + Metribuzin	9 630	1.1333	0.0001	0.9973	0.0001
Thifensulfuron + Metribuzin	17.5 210	1.1605	0.0001	1.2532	0.0001
Thifensulfuron + Metribuzin	17.5 420	0.8709	0.0001	1.0353	0.0001
Thifensulfuron + Metribuzin	17.5 630	1.1004	0.0001	1.2198	0.0001

[a]P<0.05 indicates significance and antagonism of thifensulfuron-methyl by metribuzin.
[b]days after treatment.
[c]Thifensulfuron and thifensulfuron-methyl are the same.

Table 27. Reduction in wild garlic growth from metribuzin followed in 12 h by thifensulfuron-methyl at Spring Hill in 1987[a].

Treatment[c]	Herbicide rate	12 h Application Timing			
		25 DAT[b]		50 DAT	
		Level of Reduction			
		Regression Estimate	P>|T|	Regression Estimate	P>|T|
	g ai ha^{-1}				
Intercept (control)	0	0.0000	1.0000	0.0000	1.0000
Metribuzin + Thifensulfuron	210 4	0.0375	0.0402	0.9183	0.0009
Metribuzin + Thifensulfuron	210 9	0.0625	0.0013	1.1649	0.0001
Metribuzin + Thifensulfuron	210 17.5	0.0876	0.0001	1.4120	0.0001
Metribuzin + Thifensulfuron	420 4	0.0625	0.0013	0.5986	0.0214
Metribuzin + Thifensulfuron	420 9	0.0500	0.0078	0.9928	0.0004
Metribuzin + Thifensulfuron	420 17.5	0.0750	0.0002	1.1939	0.0001
Metribuzin + Thifensulfuron	630 4	0.0625	0.0013	0.8044	0.0028
Metribuzin + Thifensulfuron	630 9	0.0500	0.0078	0.9676	0.0005
Metribuzin + Thifensulfuron	630 17.5	0.0625	0.0013	1.4120	0.0001

[a]P<0.05 indicates significance and antagonism of thifensulfuron-methyl by metribuzin.
[b]days after treatment.
[c]Thifensulfuron and thifensulfuron-methyl are the same.

Table 28. Reduction in wild garlic growth from metribuzin followed in 24 h by thifensulfuron-methyl at Spring Hill in 1987[a].

Treatment[c]	Herbicide rate	24h Application Timing			
		25 DAT[b]		50 DAT	
		Level of Reduction			
		Regression Estimate	P>|T|	Regression Estimate	P>|T|
	g ai ha^{-1}				
Intercept (control)	0	0.0000	1.0000	0.0000	1.0000
Metribuzin + Thifensulfuron	210 4	0.0500	0.0001	0.6844	0.0004
Metribuzin + Thifensulfuron	210 9	0.0500	0.0001	1.1050	0.0001
Metribuzin + Thifensulfuron	210 17.5	0.0500	0.0001	1.2532	0.0001
Metribuzin + Thifensulfuron	420 4	0.0375	0.0001	0.8829	0.0001
Metribuzin + Thifensulfuron	420 9	0.0500	0.0001	1.0172	0.0001
Metribuzin + Thifensulfuron	420 17.5	0.0625	0.0001	1.2658	0.0001
Metribuzin + Thifensulfuron	630 4	0.0500	0.0001	0.8497	0.0001
Metribuzin + Thifensulfuron	630 9	0.0500	0.0001	0.9146	0.0001
Metribuzin + Thifensulfuron	630 17.5	0.0500	0.0001	1.2733	0.0001

[a]P<0.05 indicates significance and antagonism of thifensulfuron-methyl by metribuzin.
[b]days after treatment.
[c]Thifensulfuron and thifensulfuron-methyl are the same.

Table 29. Reduction in wild garlic growth from metribuzin followed in 12 and 24 h by thifensulfuron-methyl at Spring Hill in 1988[a].

Treatment[c]	Herbicide rate	12 h Application		24 h Application	
		44 DAT[b]		44 DAT	
		Level of Reduction			
		Regression Estimate	P>|T|	Regression Estimate	P>|T|
	g ai ha^{-1}				
Intercept (control)	0	0.0000	1.0000	0.0000	0.0001
Metribuzin + Thifensulfuron	210 4	0.7065	0.0012	0.6878	0.0001
Metribuzin + Thifensulfuron	210 9	1.0333	0.0001	1.2198	0.0001
Metribuzin + Thifensulfuron	210 17.5	1.1531	0.0001	1.0790	0.0001
Metribuzin + Thifensulfuron	420 4	0.6346	0.0030	0.7468	0.0001
Metribuzin + Thifensulfuron	420 9	1.0708	0.0001	1.0309	0.0001
Metribuzin + Thifensulfuron	420 17.5	1.1939	0.0001	1.1271	0.0001
Metribuzin + Thifensulfuron	630 4	0.4149	0.0424	0.8586	0.0001
Metribuzin + Thifensulfuron	630 9	1.0902	0.0001	0.9102	0.0001
Metribuzin + Thifensulfuron	630 17.5	1.1939	0.0001	1.2532	0.0001

[a]P<0.05 indicates significance and antagonism of thifensulfuron methyl by metribuzin.
[b]days after treatment.
[c]Thifensulfuron and thifensulfuron-methyl are the same.

Table 30. Reduction in wild garlic aerial bulblet weight from metribuzin followed in 12 and 24 h by thifensulfuron-methyl at Spring Hill in 1987[a].

Treatment[b]	Herbicide rate	Application Timing					
		12 h			24 h		
		Regression Estimate	Expected Bulblet Weight	P>|T|	Regression Estimate	Expected Bulblet Weight	P>|T|
	g ai ha^{-1}		g			g	
Intercept (control	0	5.2675	5.3	0.0001	4.6250	4.6	0.0001
Metribuzin + Thifensulfuron	210 4	-5.2675	0.0	0.0008	-4.6250	0.0	0.0009
Metribuzin + Thifensulfuron	210 9	-5.2675	0.0	0.0008	-4.6250	0.0	0.0009
Metribuzin + Thifensulfuron	210 17.5	-5.2675	0.0	0.0008	-4.6250	0.0	0.0009
Metribuzin + Thifensulfuron	420 4	-5.0200	0.3	0.0012	-4.6250	0.0	0.0009
Metribuzin + Thifensulfuron	420 9	-5.2675	0.0	0.0008	-4.6250	0.0	0.0009
Metribuzin + Thifensulfuron	420 17.5	-5.2675	0.0	0.0008	-4.4975	0.1	0.0011
Metribuzin + Thifensulfuron	630 4	-5.2675	0.0	0.0008	-4.6250	0.0	0.0009
Metribuzin + Thifensulfuron	630 9	-5.2675	0.0	0.0008	-4.6250	0.0	0.0009
Metribuzin + Thifensulfuron	630 17.5	-5.2675	0.0	0.0008	-4.6250	0.0	0.0009

[a]P<0.05 indicates significance and antagonism of thifensulfuron-methyl by metribuzin.
[b]Thifensulfuron and thifensulfuron-methyl are the same.

Table 31. Reduction in wild garlic underground bulb weight from metribuzin followed in 12 and 24 h by thifensulfuron-methyl at Spring Hill in 1987[a].

Treatment[b]	Herbicide rate	Application Timing					
		12 h			24 h		
		Regression Estimate	Expected Bulb Wt.	P>|T|	Regression Estimate	Expected Bulb Wt.	P>|T|
	g ai ha^{-1}		g			g	
Intercept (control	0	4.8075	4.8	0.0001	4.9175	4.9	0.0003
Metribuzin + Thifensulfuron	210 4	-1.2100	3.6	0.4131	3.1375	8.1	0.0696
Metribuzin + Thifensulfuron	210 9	0.5225	5.3	0.7224	0.5675	5.5	0.7351
Metribuzin + Thifensulfuron	210 17.5	-1.1375	3.7	0.4413	0.3275	5.2	0.8451
Metribuzin + Thifensulfuron	420 4	0.8050	5.6	0.5848	1.9900	6.9	0.2411
Metribuzin + Thifensulfuron	420 9	-1.5975	3.2	0.2821	3.5600	8.5	0.0412
Metribuzin + Thifensulfuron	420 17.5	0.8875	5.7	0.5471	0.1525	5.1	0.9275
Metribuzin + Thifensulfuron	630 4	1.7800	6.6	0.2319	-0.1350	4.8	0.9358
Metribuzin + Thifensulfuron	630 9	1.3050	6.1	0.3779	0.4975	4.1	0.7667
Metribuzin + Thifensulfuron	630 17.5	-2.0275	2.8	0.1750	-0.9500	4.0	0.5719

[a]$P<0.05$ indicates significance and antagonism of thifensulfuron methyl by metribuzin.
[b]Thifensulfuron and thifensulfuron-methyl are the same.

APPENDIX B

PHOTOGRAPHS OF THE EFFECTS OF THIFENSULFURON-METHYL ALONE AND THIFENSULFURON-METHYL PLUS VALINE, LEUCINE AND ISOLEUCINE AFTER 2, 5 AND 10 D IN STERILE CULTURE ON THE GROWTH OF WILD GARLIC SOFT OFFSET BULBS

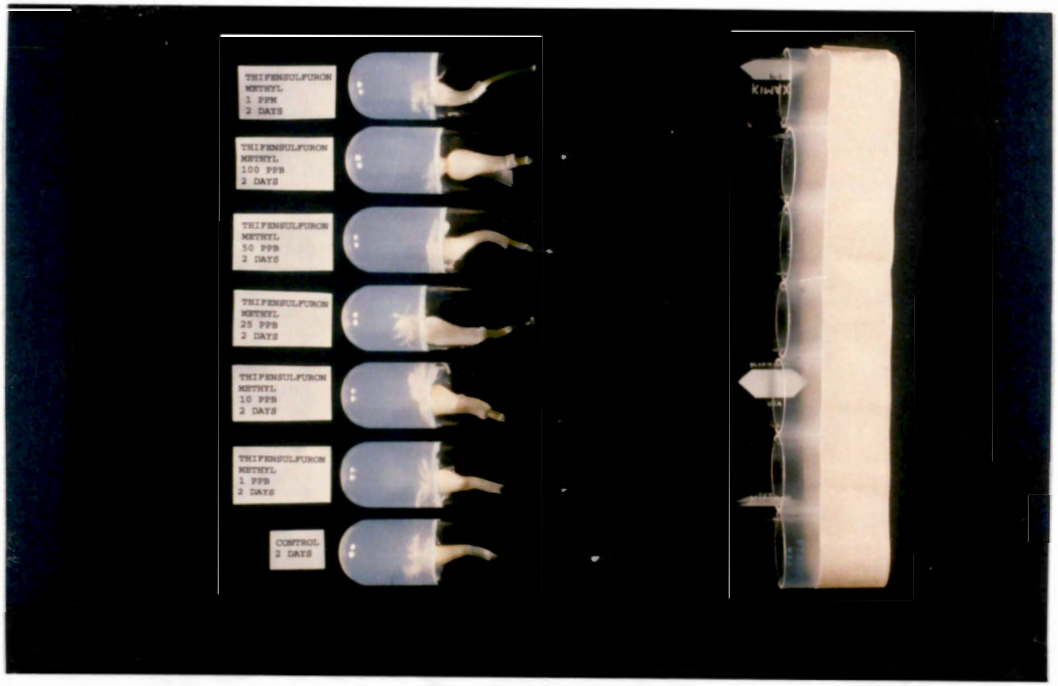

Figure 44. The effect of thifensulfuron-methyl after 2 d on the growth of soft offset bulbs.



Figure 45. The effect of thifensulfuron-methyl after 5 d on the growth of soft offset bulbs.

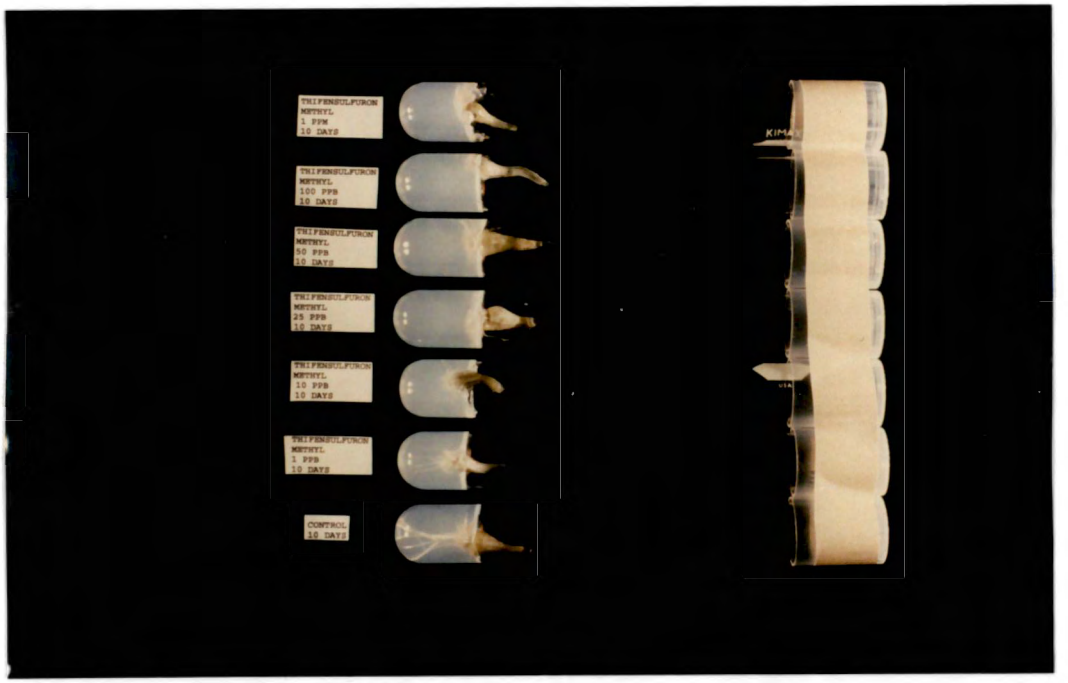

Figure 46. The effect of thifensulfuron-methyl after 10 d on the growth of soft offset bulbs.

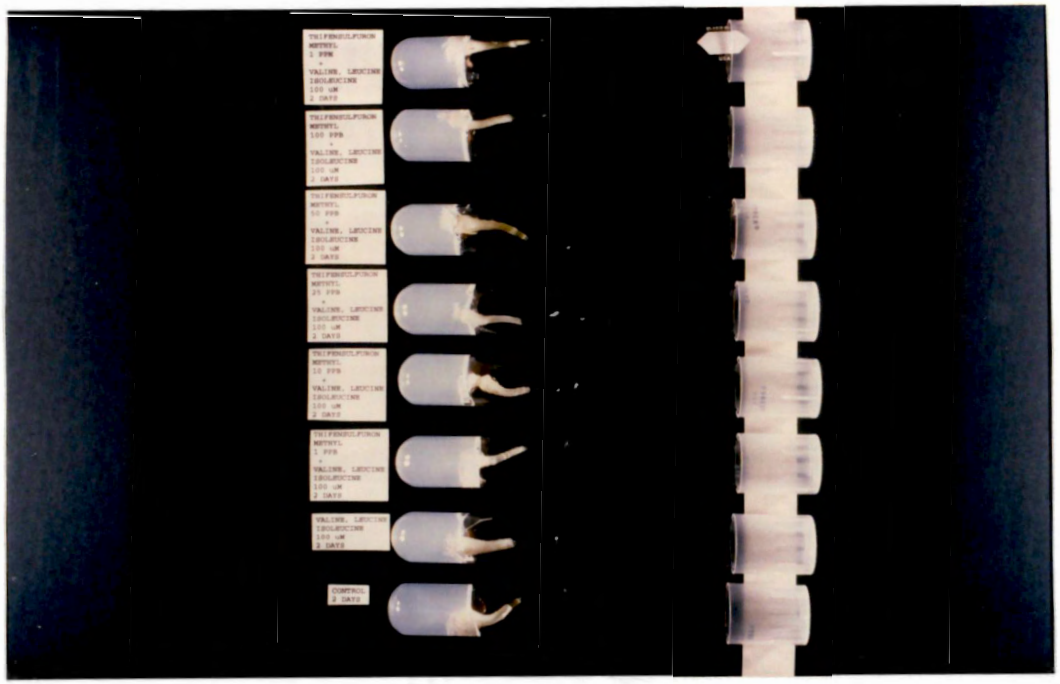

Figure 47. The effect of valine, leucine and isoleucine on the activity of thifensulfuron-methyl after 2 d on growth of soft offset bulbs.



Figure 48. The effect of valine, leucine and isoleucine on the activity of thifensulfuron-methyl after 5 d on growth of soft offset bulbs.

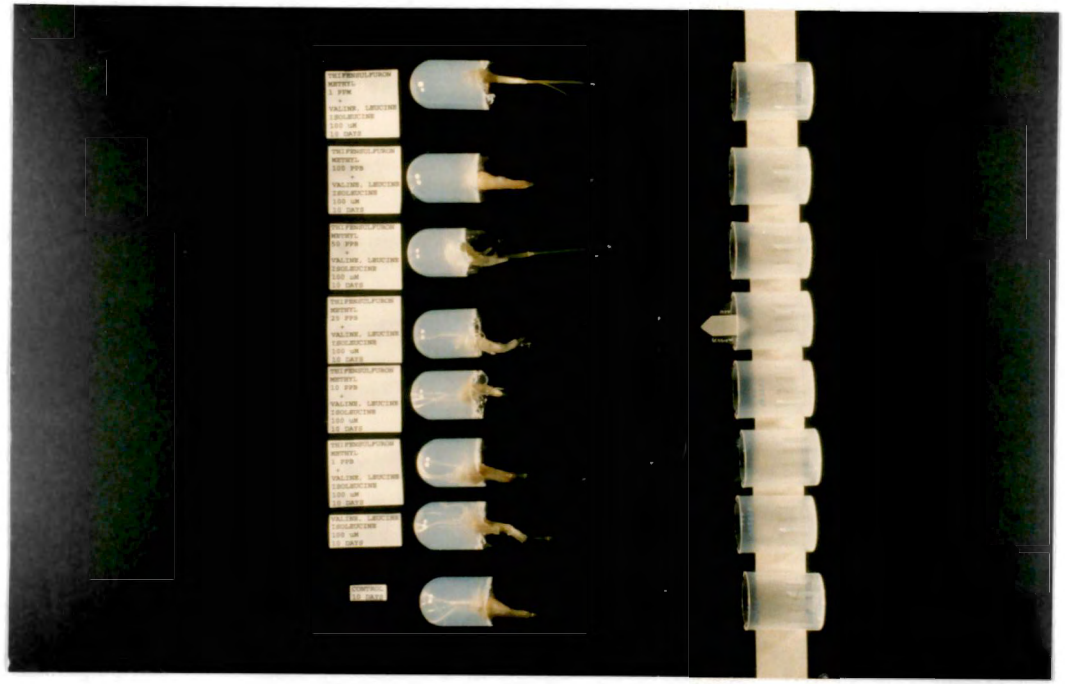

Figure 49. The effect of valine, leucine and isoleucine on the activity of thifensulfuron-methyl after 10 d on growth of soft offset bulbs.

APPENDIX C

LIST OF ABBREVIATIONS

ai..... active ingredient

^{0}C..... degrees Celsius

cm..... centimeter

d..... day or days

DAT..... days after treatment

g..... gram

ha^{-1}..... per hectare

h..... hour

kg..... kilogram

kPa..... kilopascals

L^{-1}..... per liter

m..... meter

m^{2}..... square meter

ml..... milliliter

mon..... month

ppb..... parts per billion

ppm..... parts per million

v/v..... volume for volume

wk..... week

y..... year

VITA

John London Oakes was born in Logan County, Kentucky on November 3, 1950. He attended elementary and high school at Auburn, Kentucky and graduated from Auburn High School in May of 1969. He entered Western Kentucky University in the fall of 1969 and graduated in August 1973 with a Bachelor of Science in Agriculture. He reentered Western Kentucky University in August of 1975 and in December of 1977 received a Master of Science in Agriculture. In August of 1980 he received a Master of Science from the Horticultural Food Science Department at the University of Arkansas. In the fall of 1980 he was employed as an Instructor of Agriculture in the Agriculture Department of Northeast Louisiana University, Monroe, Louisiana.

In January of 1985 he entered the Plant and Soil Science Department at the University of Tennessee to pursue a Doctor of Philosophy degree. In 1986 and 1987 he served as Graduate Student Representative to the faculty of the Plant and Soil Science Department. During the pursuit of this degree his parents developed long term health problems, resulting in home care and nursing home care for both and guardianship for a retarded brother. This resulted in his placement in a family home setting regulated through a state agency for disadvantaged individuals.