AN ABSTRACT OF THE THESIS OF

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Title: Yellow Nutsedge (Cyperus esculentus L.) Control with Herbicides: The Role of Tuberization.

Abstract approved: _____
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Research efforts involving the control of yellow nutsedge were directed toward inhibiting sprouting of parent tubers by soil-applied herbicides and inhibiting new tuber formation by postemergence herbicides. Trials were carried out under greenhouse, growth chamber, laboratory, outdoor pot, and field conditions to characterize stages of tuberization and to investigate the influence of herbicides. The effects of herbicides on tuberization and phytotoxicity at several growth stages, as well as on sprouting, growth characteristics, and survival of new tubers were determined.

Tuberization was a continuous process, but was modulated by plant age and environmental conditions. Glyphosate [N-(phosphonomethyl)glycine] toxicity to nutsedge was dependent on growth stage, with plants being more susceptible at an early growth stage than at late stages. The growth stage that included the time of first tuber initiation was the best for applying glyphosate and oxyfluorfen [2-chloro-1-(3-ethoxy-4-nitrophenoxy)-4-(trifluromethyl)benzene). At that time plants were less tolerant to herbicides and tuber production was synergistically reduced by

combinations of the two herbicides. Oxyfluorfen did not affect tuber initiation or tuber development, but glyphosate was very effective in killing rhizomes and blocking new tuber formation when applied at the early growth stage. Glyphosate reduced the enlargement of the immature tubers present at spray time, but allowed for their formation and maturation.

Plant age and length of period after spraying influenced glyphosate and oxyfluorfen absorption and translocation. Between 1 and 8 days after application of glyphosate to 30-day-old plants, absorption increased from 18 to 35% of the amount applied and translocation increased from 7 to 27%. Addition of unlabeled oxyfluorfen as a tank mixture with glyphosate increased absorption of ^{14}C-glyphosate to 27% after 1 day and 46% after 8 days and increased translocation into other plant parts to 15% and 42% for the 1- and 8-day periods. For oxyfluorfen applied alone, total absorption and translocation was about 17% and 2%, respectively. Concentration of glyphosate in the whole plant decreased from 111 dpm/mg to 42 dpm/mg when applied to 30-day- and 60-day-old plants, respectively. Also, translocation of glyphosate into the tubers was influenced by maturity of the individual tubers. Concentration in the immature tubers was about three times as much as in the mature ones.

Nutsedge regrowth ability was reduced with increasing plant age. Regrowth ability was also significantly reduced by glyphosate and oxyfluorfen applied to 40-day- and 70-day-old plants. Both herbicides reduced chlorophyll content at 6 days after spraying,

however this effect was temporary for oxyfluorfen and only glyphosate reduced nutsedge chlorophyll when measured at 15 days.

Proper timing of control measures for tuberous weeds such as nutsedge is extremely important because the tuberization process must be stopped before new tubers start to form and develop. Therefore, timing of postemergence herbicide applications relative to tuberization is crucial for overall control of yellow nutsedge.

When soil applied herbicides were compared in the field, consecutive applications of dichlobenil (2,6-dichlorobenzonitrile) and metolachlor [2-chloro-N-(2-ethyl-6-methylphenyl)-N-(2-methoxy-1-methylethyl)acetamide] for two years provided the best control of nutsedge. Dichlobenil and metolachlor reduced tuber populations at the end of 2 years by as much as 88% and 90%, respectively. A measure of long-term control was obtained by their residual effect on nutsedge tuber population.

YELLOW NUTSEDGE (*Cyperus esculentus* L.) CONTROL WITH HERBICIDES: THE ROLE OF TUBERIZATION

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Typed by Welington Pereira

This thesis is dedicated to

my sons Junior and Alex, to

my wife Nina, and to

my parents Joaquim and Clarinda

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Chapter 1

INTRODUCTION

Yellow nutsedge is one of the most troublesome perennial weeds of the U.S. (77, 93, 166, 186), and world (70) thriving over a range of soil and climatic conditions. Although eradication of nutsedge has been attemped since 1925 (119), changes in agriculture such as the increased use of herbicides for control of annual weeds, less handhoeing, reduced tillage, etc, have lead to dramatic increases in infestations in many areas of the U.S. (1, 45, 77, 93, 133, 166, 180, 189).

Yellow nutsedge proliferates by a complex system of underground rhizomes, basal bulbs and tubers (77, 103, 134). Since tubers are the most important propagule of this weed and an important factor in its dissemination in cultivated areas, efforts to develop rational control methods should be based on how to inhibit sprouting of tubers and/or how to inhibit new tuber formation.

Historically, herbicides from several chemical families have provided poor and only temporary control of this weed. Reasons include failures to translocate to the site of action, to inhibit sprouting of parent tubers, to prevent escapes, and to inhibit new tuber formation. A number of both soil- and foliage-applied herbicides have been evaluated for the control of yellow nutsedge

with the emphasis on determining the effect on parent tubers or on nutsedge foliage. A few studies have investigated the influence of herbicides relative to nutsedge growth stages, new tuber development, tuber sprouting, and growth characteristics. Inconsistent results have been reported caused by interactions resulting from differences in the physiological status of nutsedge plants and environment conditions. These studies have not provided adequate or precise enough information to understand herbicide effects on new tuber formation and the later ability of these nutsedge tubers to sprout.

The following studies were conducted to investigate the influence of herbicides on tuberization and control at several growth stages of yellow nutsedge, as well as on sprouting, growth characteristics, and survival of tubers. Trials were carried out under growth chamber, laboratory, greenhouse, outdoor pot, and field conditions. In chapter 2 a review of the biology, life cycle, tuber characteristics, and the action of different herbicide families on purple and yellow nutsedge will be discussed. Chapter 3 reports nutsedge response to glyphosate as affected by growth stages. The tuberization process was evaluated by growth analysis and related to glyphosate phytotoxicity. The interactions of glyphosate and oxyfluorfen as they affect tuberization and control, relative to plant growth stages of yellow nutsedge, are discussed in chapter 4. Phytotoxicity, absorption, and translocation of glyphosate and oxyfluorfen are discussed in chapter 5, elucidating the reasons for greater effectiveness of the herbicides applied at an early nutsedge

growth stage. Nutsedge response to soil-applied herbicides in greenhouse and under field conditons were evaluated, and single and repeated yearly applications are compared in chapter 6.

Chapter 2

REVIEW OF THE LITERATURE

1. Biology, life cycle, and tuber characteristics of yellow nutsedge

Yellow nutsedge is a perennial that proliferates mainly by a system of underground rhizomes, basal bulbs and tubers (77, 103, 134). Tubers are important in that they guarantee the vegetative propagation of this weed, especially in agricultural areas where tubers can be easily disseminated. The tubers lie dormant in the soil during fall and winter where they are naturally stratified by cold and wet conditions of winter, breaking tuber dormancy and sprouting by spring (13, 136, 162).

Tubers can be killed relatively easily by extreme cold or heat as well as by desiccation (33, 119, 138). A combination of low temperature and low humidity is more effective in reducing tuber viablility than either factor alone (157). Temperatures of -7 ^{o}C killed 50% of the tubers in the laboratory, while some tubers survived -20 ^{o}C in the field (135, 136). Low temperature apparently killed tubers near the soil surface during the winter (136), but the cold did not greatly affect those tubers deep in the soil because the surface insulated them from exposure to the severe cold (157). Variants have been found to differ in their responses to both temperature and desiccation and some survived dessiccation (35, 98, 158). Over 80% of the tubers in the soil will decay in less than 3 years (136, 158, 162) although longevity is dependent on tuber depth in the soil. Half-lives of about 4

months are expected for tubers 10 cm below the surface (136). Factors which will affect sprouting and emergence of viable tubers include temperature, moisture, light, depth of tuber in the soil, type of cultivation, crop, tuber dormancy, etc. Germination inhibitors are present in dormant tubers (76, 149, 163), but natural water movement through the soil during the dormant period aids in leaching inhibitors out of the tubers. Ethylene, chlorohydrin, thiourea, ethyl ether, ethephon, potassium thiocyanide, hydrogen peroxide, oxygen, gibberellic acid, and benzyladenine have been reported to break dormancy when applied at an adequate concentration (13, 156, 163). Such physical actions as stratification (2 to 5 ^{o}C) (13, 162), scarification (156), dessiccation (158), and leaching with water (162) are also effective in breaking tuber dormancy. Diurnal alternating temperatures, which occur in the soil in the spring, promote sprouting. Promoting tuber sprouting in the field as a prelude to control has been proposed but getting the germination promoting substances into the tubers in the soil would be difficult. In the spring when the soil warms up to about 12 ^{o}C, some tubers are stimulated to sprout (136). Sprouts readily emerge from tubers 30 cm deep and some through 46 cm of soil, but emergence is delayed as the tuber depth in the soil increases (148, 162). 61). Tillage operations have stimulated tubers to sprout in the field (33, 35).

When a tuber sprouts, one or more slender rhizomes elongate vertically from the buds near the terminal end of the tuber (136). The rhizome is a continuation of the growth of the rhizome that

formed a tuber, with the new rhizome expressing a negatively geotropic (upward) response on sprouting. Because the end of the scale-leaf at the tip of the rhizome is sharp and strong, they can penetrate through resistive substrates. The presence of numerous buds in a tuber allows for it to sprout several times or to produce several sprouts at one time (15, 134, 159). The first sprouting usually consumes most of the food reserves (60%), leaving the subsequent sprouts with reduced vigor (134). Apical dominance in tuber buds is expressed in as much as the attached developing ramet inhibits sprouting in the other buds. Dominance is released, allowing additional buds to sprout, when the sprouts are detached from the tuber or suppressed. The evolving ramet remains attached to the tuber by the vertical rhizome if it is not physically detached (140).

As the rhizome reaches the soil surface, the rhizome tip encounters sunlight and diurnal temperature fluctuations, which are the principal factors in stimulating a basal bulb to form under the soil (140, 141). A temperature alternation of 10 ^{o}C stimulated basal bulb formation. Phytochrome did not appear to be the photoreceptor for the stimulus, as both red and far-red light responses were identical to that of white light (140). In a uniform seedbed, all basal bulbs are at a comparable distance from the soil surface, regardless of the depth of the parent tuber. As a basal bulb forms, the nodes in the top of the vertical rhizome become compacted leaving very short internodes. Leaf length and function are dependent on internode length: short scalelike leaves develop at nodes attached to long internodes and long

photosynthetically active leaves develop at short internodes in the basal bulb (140). Leaves grow out of the bulb from a plicate triangular fascicle beginning with the outermost leaf and the fascicle terminates under proper conditions in a seed-bearing rachis.

The basal bulbs are considered the principal sites of vegetative activity and propagation (13, 48, 70, 77). Several weeks after sprouts emergence rhizomes develop from the basal bulb. These early season rhizomes elongate nearly horizontally from the bulb until the tips turn upward, differentiating into secondary basal bulbs similar in structure to the primary basal bulb. These secondary bulbs produce shoots, then rhizomes, as described for primary bulbs. As the ramet continues to grow bulbs of a higher order form on rhizomes from the secondary basal bulbs, giving rise to a complex system of rhizomes and basal bulbs that eventually lead to the production of tubers (140).

Secondary rhizomes arise from basal bulbs or from tubers during sprouting (77, 134, 183). A new rhizome grows as an indeterminate stem with nodes and internodes. The growing point of an indeterminate rhizome is enclosed by the terminal scale-leaves and can transform into either a basal bulb or a tuber (183). A rhizome growing from a sprouting tuber, however, is determinate and can transform only into a basal bulb. The rhizomes that develop from sprouting tubers are structurally similar to those that develop from basal bulbs (183).

Wilson (184) defined tuberization as the sum total of all

processes that lead to the development of the mature tuber. The individual processes involved are tuber initiation and tuber development. The locus of the primary events in tuber initiation depends on the origin of the tuber which may be either a specialized tuber-bearing organ such as a stolon of a potato or a rhizome of nutsedge, or an existing organ of the plant (e.g. on roots of carrots and beets). Under appropriate enviromental conditions and physiological status of the plant tubers are differentiated from meristems at the rhizome tip. The internodes at the tip become increasingly shorter, resulting in a group of appressed leaves that form a cone (15). Tuber formation begins in the meristematic region behind the leaf primordia (77, 183). The leaf primordia remain dormant as the parenchemma cells, both inside and outside the endodermis, enlarge and accumulate starch. As the tuber enlarges the rhizome vascular system develops into the tuber matrix (183). A newly differentiated tuber is white and fresh appearing, changing to tan and brown as it matures. Mature tubers clearly display their nodes, internodes, roots, scale leaves and tend to be spherical. Several morphological types however, based primarily on tuber color and shape, have been described (150, 156). Distorted shapes commonly result from physical constraints during development. Small thick-veined leaves are appressed against the tuber surface (15).

Photoperiod is considered the major known factor controlling the differentiation of rhizomes (48, 77), but temperature fluctuations, chemicals, and nutrition also affect differentiation (14, 48). Long days promote differentiation into basal bulbs and

as day length shortens late in the season rhizomes differentiate into tubers (48, 77), although under certain experimental conditions tuber initiation also occurred in 30-day-old nutsedge plants (116). Tuber initiation seems to be related to the age of plant and temperature (113, 116). At the end of the growing season more than one-third of the total plant dry weight is commonly in tubers (79).

Mature tubers vary greatly in size within a local population as well as among various populations (98, 159, 162). For example tubers ranged from 3 to 11 mm in diam and with an average weight of 150 mg in Illinois, from 2 to 7 mm and 70 mg in Minnesota; and 12 mm and 710 mg in Maryland. In most undisturbed soils, more than 75% of the tubers are produced in the surface 15 cm layer, with some tubers being produced as deep 46 cm in dense populations of yellow nutsedge. Tuber density in the soil is affected somewhat by the density of plants of yellow nutsedge. In a dense, pure stand, yields of 5 to 16 thousands tubers/m2 commonly weighing 9 to 15 tons/acre have been recorded (138, 162). In cultivated fields in competition with a crop, tuber densities on the order of 1,100/m2 have been observed (138). A single plant growing alone produced 9,000 tubers in a season (162).

Besides the quantity of tubers produced their quality is important with respect to vegetative propagation of this weed. Carbohydrates are the most abundant component in tubers, constituting from 45 to 75% of tuber dry weight. While starch is the major carbohydrate present, also present are small quantities

of fructose, glucose, sucrose, melobiose, and other polysaccharides (99). Lipids constitute from 5 to 14% of tuber dry weight (99, 138). The storage lipids, triglycerides, are present in the largest quantity (more than 80%) while polar lipids make up only about 1%. Oleic and linoleic are the principal fatty acids in tubers; other fatty acids present are palmitic, linoleic, and stearic. Protein content varies from 5 to 10% of tuber weight (99). The constituent amino acids of these proteins have not been characterized (140). Tubers contain a variety of phenolic compounds. Ferulic and p-coumaric acids are the most abundant, but vanilic, p-hydroxybenzoic, syringic, protocatechuic, and caffeic acids as well as eugenol and other phenolic compounds are also present (76, 149). Since these compounds are known inibitors of plant growth they may be important factors in inhibition of tuber sprouting. Tuber dormancy is considered one of the major obstacles in controlling this weed, because the presence of a population of dormant tubers makes many control methods ineffective. If all the tubers in the soil could be stimulated to germinate at one time, and if subsequent plants could be killed, yellow nutsedge could be eliminated (140). Tubers exibit maximum dormancy at the end of the growing season and least dormancy in the spring and early summer (13, 150), some dormancy may be retained for the entire year.

Populations of yellow nutsedge are composed of different varieties, biotypes, and tuber sizes, thus contributing to the variations discussed.

2. Action of Different Herbicide Families on Purple and Yellow Nutsedges

Historically, herbicides from several chemical families have provided poor and only temporary control of purple (Cyperus rotundus L.) and yellow (Cyperus esculentus L.) nutsedges. Reasons include failures to translocate to the site of action, to inhibit sprouting of parent tubers, to prevent escapes, and to inhibit new tuber formation. Since the advent of herbicides most soil- or foliage-applied herbicides have been evaluated for the control of these weeds, with the principal emphasis on determining the effect on parent tubers or on nutsedge foliage. Inconsistent research results were reported because of interactions resulting from differences in stage of nutsedge tuberization and environmental interactions. Therefore, it would seem appropriate that research efforts for the control of nutsedges should be directed in the area of inhibiting sprouting of tubers and inhibiting new tuber formation. A few studies have investigated the influence of herbicides relative to nutsedge growth stages, new tuber development, and the ability of these nutsedge tubers to sprout at a later date.

Growth regulators- Within the phenoxy group of herbicides, 2,4-D [(2,4-dichorophenoxy) acetic acid] appears to be the most active on nutsedge (97). Historically, 2,4 D has provided erratic control (31, 30, 60, 96, 107, 167) and repeated applications are required for both purple and yellow nutsedge (8, 63, 128, 131). Excellent purple nutsedge control resulted from 9 applications of

2,4-D initiated 1 or 2 weeks after shoot emergence (63). In yellow nutsedge, poor foliar wetting and retention of spray mixture and a low rate of absorption (less than 19%) and translocation (less than 12%) (16), may explain the slow control of foliage and tubers with 2,4-D. Applications of 2,4-D killed mostly nutsedge shoots (18, 19, 58), although inhibition of lateral rhizomes and tubers of yellow nutsedge following 2,4-D applications have been noted (21, 16). Perhaps this species may be biochemically susceptible since appreciable degradation of the 2,4-D was not observed (16). Loustalot et al. (97) found that purple nutsedge plants 3 to 4 weeks old began to produce new underground tubers which sprouted after the top was killed with 2,4-D.

Photosynthesis inhibitors- Inconsistent control of yellow nutsedge with atrazine [6-chloro-N-ethyl-N'-1-methylethyl-1,3,5-triazine-2,4-diamine] (66, 112, 125, 173, 185) may be due to differences in nutsedge varieties (87), biotypes (190), and size of tubers (168). Consistent control has been achieved with split applications of soil incorporated and directed postemergence treatments of atrazine along with combinations of alachlor [2-chloro-N-(2,6-diethylphenyl)-N-(methoxymethyl)acetamide, EPTC (S-ethyl dipropyl carbamothioate), or butylate [S-ethyl bis(2-methylpropyl)carbamothioate] (112). When postemergence sprays of atrazine are applied, thorough wetting of the foliage must be achieved since there is little basipetal translocation and the action is mainly "contact" rather than "systemic" (170).

Effective control of purple and yellow nutsedge has been

reported with bromacil [5-bromo-6-methyl-3-(1-methylpropyl)-2,4(1H,3H)pyrymidinedione] and terbacil [5-chloro-3-(1,1-dimethylethyl)-6-methyl-2,4(1H,3H)pyrymidinedione], especially when these herbicides were applied in the early stage of plant growth (86, 120, 123, 171, 178,, 189). Part of the success with the uracils may be because they translocate readily in nutsedge plants (121).

Linuron [N'-(3,4-dichlorophenyl)-N-methoxy-N-methylurea has been found to reduce nutsedge growth (20, 21, 100, 127), but not enough to provide commercial control.

Bentazon [3-(1-methylethyl)-(1H)-2,1,3-benzothiadiazin-4-(3H)-one 2.2-dioxide] has selectively controlled yellow nutsedge in many crops (80, 91, 137, 164, 174, 180, 186), particularly when applied in split applications 5 to 10 days apart (51, 137, 142, 174, 186) to actively growing nutsedge plants at the 4- to 6-leaf stage (51, 137, 174, 186). For this herbicide to be effective, complete coverage of nutsedge foliage is essential and temperatures must exceed 24 ^{o}C (113). In contrast, one study showed that young plants were controlled at lower temperatures, under low light intensities, and high soil moisture conditions (143). Evidence suggests that parent tubers were controlled with bentazon (137), although repeated annual treatments were needed (180).

Pigment synthesis inhibitors- Excellent control of purple nutsedge has resulted from two applications of amitrole (1H-1,2,4-traizol-3-amine), the first being applied 4 weeks after shoot emergence.

Conversely, nutsedge appeared resistent to amitrole when it was applied later than 6 weeks after emergence (62, 63). Amitrole readily penetrates and translocates in the nutsedge plant including movement into the attached tubers (1, 41, 59, 69). Results include prevention of new tubers and reduced germination of the seeds and sprouting of tubers produced from treated plants (41, 69, 147).

Fluridone [1-methyl-3-phenyl-5[3-(trifluoromethyl)phenyl]-4(1H)-pyridinone] provided good yellow nutsedge control in cotton as measured by shoot and tuber production (12). Control of yellow nutsedge with low rates of fluridone was better when incorporated in the soil than when it was applied preemergence to the soil surface (11, 12, 88, 108). Foliage regrowth from tubers exposed to fluridone for 2 weeks displayed a typical bleached appearance for a few weeks and new tuber production was prevented (12). Also, phytotoxicity to nutsedge has been increased synergistically by the combination of fluridone and bentazon applied to 30-day-old plants. Sterrett (132) related that a similar response was observed on Canada thistle [Cirsum arvense (L) Scop.], and suggested that fluridone apparently was active only on the young developing foliage of Canada thistle, whereas bentazon was mainly effective on the mature foliage. This foliage selectivity could explain, at least in part, the observed interaction between the two herbicides.

Preemergence applications of norflurazon [4-chloro-5-(methylamino)-2-(3-(trifluoromethyl)phenyl)-3-(2H)-pyridazinone]

have not provided the same level of yellow nutsedge control in the field as was observed in greenhouses (12, 113). The relatively low water solubility of norflurazon may prevent its movement to the deeper sprouting tubers under field conditions.

Cell membrane destroyers- Control of purple nutsedge with paraquat [1,1'-dimethyl-4,4'-bipyridinium ion] has been inconsistent (53, 131). Repeated applications of paraquat or dinoseb [2(1-methylpropyl)-4,6-dinitrophenol] reduce, but do not eliminate nutsedge stands (52, 118, 176). Paraquat will quickly dessicate the foliage (53, 100) and stop new tuber production (100), but effects are temporary and new sprouts emerge from parent tubers (53, 154) or basal bulbs (151). By killing an emerged shoot of purple nutsedge with paraquat, dormant buds on the basal bulb or tuber may be stimulated to produce new shoots as a result of released apical dominance (53, 153) resulting in possible increases in nutsedge populations where paraquat is used to maintain a zero-tilage system (153).

Activity of diphenyl ether herbicides is expressed as a foliage burn 4 to 6 hours after a postemergence application in the light. These chemicals are referred to as contact herbicides and apparently are not translocated following root or foliar application on young plants (2, 170). They induce lipid peroxidation which leads to increased membrane permeability and membrane destruction (170). Night applications of nitrofen [2,4-dichloro-1-(4-nitrophenoxy)benzene] with or without an herbicidal oil (11.7% aromatics) suppressed purple nutsedge foliage by 60%,

but was not considered selective in carrots. Activity of these chemicals was greater, probably as a result of increased penetration, during the warm wet season than when it was cold and dry (177). Oxyfluorfen [2-chloro-1-(3-ethoxy-4-nitrophenoxy)-4-(trifluoromethyl)benzene] quickly desiccated yellow nutsedge foliage and reduced tuber production by as much as 50% (114). Similar to paraquat, oxyfluorfen allowed resprouting from parent tubers. Combinations of glyphosate [N-(phosphonomethyl)glycine] and oxyfluorfen provided more rapid top kill and synergistically reduced tuber production (114, 116). Nutsedge tolerance to these herbicides increased with plant age, possibly a result of less wetting and leaf penetration in older plants (116). Also, fluorodifen [P-nitrophenyl (α,α,α-trifluoro-2-nitro-p-tolyl)ether] provided effective control of purple nutsedge when it was applied before the 4-leaf stage (46).

Shoot inhibitors of emerging seedlings- All herbicides in this chemical group are soil-applied and have high vapor pressures. They inhibit growth in meristematic regions of leaves of susceptible species resulting in disruption of cell division or induction of abnormal cell elongation and expansion (2). These herbicides can readily enter roots, but must translocate to the shoot growing point to be active since they have no direct effect on roots (170). EPTC and other thiocarbamate herbicides have been used extensively to suppress purple and yellow nutsedge during the early growth stages of many crops (13, 54, 64, 66, 81, 82, 88, 112, 122, 172, 173, 180, 185). Nutsedge tubers sprouted readily in

soil treated with EPTC, but shoot growth was temporarily suppressed. These herbicides did not kill tubers or eradicate nutsedge (71, 73, 109, 1250), thereby requiring repeated applications to maintain a satisfactory level of control (71, 109).

EPTC has been used for decades to suppress nutsedge in potato fields, but has not significantly affected the nutsedge population in areas where this crop is grown (147). This would indicate that EPTC probably suppressed nutsedge growth during periods when herbicidal levels were present in the soil, but failed to affect subsequent mother plants, new tubers, and future populations.

EPTC persisted longer and was more effective when applied and incorporated into dry soil than when incorporated into moist or wet soil. Under tropical conditions during the dry season, satisfactory control of purple nutsedge was achieved at half the rate normally used during the wet season (179). Mechanical incorporation of EPTC resulted in more effective control of nutsedge than incorporation by leaching with water (22, 61, 64, 109, 146).

Although some suppression of both purple and yellow nutsedge has been reported with butylate (3, 10, 42) and vernolate [S-propyl dipropylthiocarbamate] (65, 67), EPTC was more active and presents more potential for crop injury (13, 66, 173).

Root and shoot inhibitors of seedlings- The relative activity of chloroacetamide herbicides such as alachlor and metolachlor [2-chloro-N-(2-ethyl-6-methylphenyl)-N-(2-methoxy-1-methylethyl)

acetamide] on roots versus shoots varies with species (170). Generally, these herbicides have proven to be relatively effective in controlling nutsedge at low rates of application (37). The chloroacetamides are not strongly influenced by soil factors and apparently do not kill tubers, but act by delaying sprouting of tubers and killing young nutsedge shoots (5, 6, 13, 38, 72, 86, 110, 120, 139, 172). These two herbicides have been used to selectively control nutsedge in several crops (5, 6, 37, 85, 113, 133, 172, 173, 180), but metolachlor often provides better control with more residual activity than alachlor (37, 68, 108, 169). Most roots arising from the nutsedge basal bulb provide a more direct route for metolachlor movement into the shoots, and therefore increasing nutsedge susceptibility as compared to alachlor. Substantial acropetal translocation of root absorbed metolachlor may explain the greater level of control of yellow nutsedge with metolachlor than with alachlor (108). Persistence of alachlor and metolachlor in the soil affects the period of nutsedge control (110).

The level of nutsedge control generally declines after 2 to 4 months following a single application of alachlor or metolachlor, although repeated applications have extended control for an additional 6 weeks (133). These herbicides are more effective in controlling nutsedge when incorporated into the soil than when applied to the soil surface (38, 28), although seasonal control of yellow nutsedge has been obtained with metolachlor regardless of the method or depth of incorporation (68, 108, 113). Improved control was achieved by placing these herbicides in the soil above

the nutsedge tubers (5, 38, 113, 172). In one study, metolachlor applied in the spring to the soil surface in a pear orchard provided greater than 96% control for the season (115). By the end of the growing season, tuber production was reduced by 82% (113). Plots treated only once in the previous year were evaluated at the end of the second growing season when tuber production was still reduced by 75%. This would indicate that there is a residual effect of the herbicide on yellow nutsedge tuber population and more than seasonal control can be obtained (115).

Dichlobenil [2,6-dichlorobenzonitrile] kills germinating seeds and inhibits growth of young seedlings and sprouts of perennial weeds emerging through the soil. The mode of action is a mitotic poison that inhibits seed germination and cell division in meristematic regions (2). Both field and greenhouse studies with dichlobenil have indicated that this herbicide can provide good control of purple and yellow nutsedge (56. 113, 120, 126, 171). Winter surface applications of 6 kg/ha of dichlobenil in a pear orchard has afforded seasonal yellow nutsedge control of 96% or greater, and by the end of the season reduced tuber populations by 62% (113). Surface applications of dichlobenil in winter or incorporated applications in spring provided better control of nutsedge tuberization than surface applications in spring. This enhanced herbicidal activity was probably related to less evaporative loss of dichlobenil when it was incorporated, either mechanically or by rainfall. Plots treated only once in the previous year were evaluated at the end of the second growing

season when infestations remained at 60% of the control. As with metolachlor, this would indicate a residual effect of the herbicide on the yellow nutsedge tuber population (115).

Herbicides that interfere with plant amino-acid metabolism- Of the many postemergence herbicides tested for nutsedge control, glyphosate has provided the greatest potential for suppressing resprouting of parent tubers (4, 17, 40, 87, 88, 89, 100, 102, 117). This readily translocated herbicide also has shown considerable promise for inhibiting tuber formation when applied at the tuber initiation stage (116). Rates higher than 1 kg ai/ha have been needed to satisfactorily control yellow nutsedge (17, 116, 143, 180) although single or split applications at lower rates have controlled small plants (137, 113).

Hunt and Linscott (74) reported significant interactions between season, glyphosate rates, and time of application on viable tuber production. As the early part of the season progressed, less glyphosate was needed to obtain comparable reductions in number of tubers. A 2 kg/ha rate significantly reduced production of viable tubers from plants emerging in the 5 week period after May 30, whereas plants emerging before May 30 produced viable tubers at the same application rate (74).

This herbicide has been effective for inhibiting tuberization when applied no later than the time of first tuber initiation, but it was not very effective in the control of those rhizomes in which the tuberization process was visibly underway. Glyphosate killed young rhizomes without tubers, thereby blocking tuber

formation (116). Applications following tuber initiation appeared to reduce tuber enlargement slightly, but allowed for their formation and maturation (114, 116, 180). Regrowth of nutsedge under field conditions has been reported to be due either to the presence of dormant tubers which sprouted after treatment or to some escapes of basal bulbs (40, 113). Repeated applications of glyphosate were necessary to substantially reduce the number of both yellow and purple nutsedge plants (106, 113, 115, 161, 196).

When production of new yellow nutsedge shoots was induced by soil treatment with naptalam (N-1-naphthylphthalamic acid), a later foliar spray of glyphosate resulted in significantly greater control than when either herbicide was used alone (4). This suggested that a possible approach for improved control would be to inhibit apical meristems of rhizomes from being transformed into tuber meristems and thereby encourage the transformation of rhizomes into shoots with a subsequent treatment of a translocated foliage-active herbicide (4). This technique would not be effective unless a reasonable level of control of shoots is achieved and tuber formation is blocked (114, 116). Under some circunstances, nutsedge plants treated with naptalam have produced more shoots and tubers (55).

When tillage followed application of glyphosate, nutsedge control was increased and tuber populations reduced (23, 39, 49). Excellent control of purple nutsedge foliage has been achieved when treated in each of two or three consecutive seasons (spring to fall) (27, 191).

Glyphosate has been applied to nutsedge in combination with a

range of other herbicides and certain non-herbicidal additives (9, 114, 116, 144). Most herbicides often produce antagonistic effects with glyphosate, especially those which inhibit photosynthesis (9, 144). In contrast, 2,4-D amine, amitrole (144), and oxyfluorfen (116) provided additive or synergistic effects. Combinations of glyphosate with oxyfluorfen produced more rapid top kill and improved control of new tubers when applied at the tuber initiation stage (30 days after emergence). However, later applications at the mature tuber stage (60 days after emergence) with these herbicide combinations produced antagonistic effects (114, 116). This difference due to timing was attributed to greater absorption and translocation of each herbicide when applied at the tuber initiation stage of yellow nutsedge.

Many researchers have reported that glyphosate activity is affected by environmental factors such as temperature, relative humidity, light intensity, photoperiod, and soil moisture (9, 23, 24, 101, 165, 181). Purple nutsedge was somewhat more susceptible to glyphosate during the rainy season than during the dry season (23). Also, glyphosate effectiveness on nutsedge was reduced under high moisture stress (24, 102) and low humidity (24). Application of water to the roots one week before and one week after treatment with glyphosate enhanced the activity (102). High light intensities (143, 170) as well as short photoperiods (155) also increased glyphosate activity. Reports of temperature interactions have been inconsistent (116, 143, 152).
Extensive research has also indicated that glyphosate accumulates

in the meristematic regions of foliage, roots, rhizomes and tubers in a typical source to sink pattern. Nutsedge tubers are apparently a major sink for glyphosate accumulation following foliar applications, although considerable amounts of the herbicide have also been detected in other developing tissues such as flowers, newly formed leaves, basal bulbs, roots, and rhizomes (24, 25, 36, 47, 50, 57, 78, 90, 116, 124, 129, 130, 182, 188, 192). Zandstra and Nishimoto (192) reported that tubers from young purple nutsedge plants contained greater concentration of labeled glyphosate than leaves. With increasing plant age, specific activity decreased in both tubers and leaves. Total translocation to tubers increased with a concurrent slight decrease to leaves when the plant began to form new rhizomes and tubers. Leaves accumulated less glyphosate than tubers at all growth stages when tubers were present. In purple nutsedge, glyphosate appeared to move through mature tubers to the newly forming tubers at rhizome tips, probably because as the plant matures new root growth decreases and the assimilate flow shifts to the new rhizomes and tubers. There was no evidence of metabolism of glyphosate in purple nutsedge leaves or tubers 16 days after application. Differences were probably due to the physiological status of the plants in that older plants had more dormant tubers, strong apical dominance, and less active metabolism (160, 192). The authors concluded that the best time to apply glyphosate for purple nutsedge control was when a maximum number of newly produced rhizomes and tubers were connected to healthy foliage. In the same study, it was determined that the lack of translocation of

herbicide within purple nutsedge rhizome tuber chains was a major reason for poor control (192). Also, total below-ground translocation of labeled glyphosate was about 3% (24) which may explain a major reason for poor control. A minimum interval of 3 days between glyphosate application and tillage was considered necessary for adequate translocation to the tubers (23, 40, 192). Translocation of a phytotoxic dose of glyphosate was achieved within 72 hours at 1 kg/ha application rate, whereas 36 hours was sufficient at 2 kg/ha indicating that responses in tubers were rate dependent (40).

Moisture relations within the nutsedge plant also appeared to have a major impact on translocation of glyphosate. A three-fold increase in translocation into the underground parts of purple nutsedge occured at 90% as compared to 50% relative humidity, and twice as much translocation occured at -2 bars as compared to -11 bars of soil water potential (24).

Miscellaneous herbicides- Repeated applications with organic arsenical herbicides over a 2-year period were reported to reduce the number of nutsedge tubers (52, 84, 92, 175). The authors suggested that tuber mortality was caused by depletion of food reserves rather than accumulation of a specific level of arsenic (44, 72), although organic arsenical herbicides translocate readily in purple and yellow nutsedge (43, 44, 72, 84). Small and actively growing purple nutsedge tubers accumulated more arsenic than large quiescent tubers (5, 44, 72, 83). Soil fumigants have eradicated patches of perennial weeds (2). Both a friable soil

for good penetration, and a tarp to maintain the concentration of fumigant at the soil surface are required. Correct applications of methyl bromide, a multi-purpose soil fumigant, have nearly eliminated nutsedge infestations (34, 92). Proper soil moisture levels to ensure maximum tuber viability and presprouting is also essential to obtain control. The dithiocarbamate herbicide, metham (sodium methyldithiocarbamate), acts as a soil fumigant, and is recommended to control nutsedges. Metham degrades in moist soil to methyl isothiocyanate as the principal toxic product which rapidly diffuses through the soil in vapor form. Metham persists only for 1 to 5 hr in moist soil and the methyl isothiocyanate disappears almost completely in 2 to 3 weeks (2).

Oil carriers or adjuvants for increasing nutsedge control with herbicides- Oil carriers and adjuvants have enhanced the activity of many herbicides. Enhancement is due partially to the greater penetration caused by lower surface tension and greater wetting ability of the oil or adjuvant. Penetration of 2,4-D and linuron in yellow nutsedge was increased when these herbicides were applied in an oil mixture (20, 21, 107). Also, the addition of oil enhanced atrazine (26, 105, 112) and bentazon (137, 186) activity when applied on nutsedge foliage.

Combinations of nitrofen with an herbicidal oil are more effective in growth of purple nutsedge foliage than either herbicide applied alone. The authors believed that these herbicides used in combination remained active on the foliage for longer periods of time, thereby increasing herbicidal retention

and penetration (177).

Phytotoxicity of paraquat to nutsedge was increased with the use of nitrogenous carriers (154). Control was greater with potassium nitrate than with urea. The inference from this research was that a greater amount of paraquat moved into the tubers of purple nutsedge along with the nitrogen compounds.

Under field conditions ammonium sulfate significantly enhanced glyphosate activity in purple nutsedge in the dry season (144, 145). The authors suggested that a natural rejuvenating effect of the first rains of the season, or an artificial "rejuvenation" following clipping, fertilizing, or using a contact herbicide might have resulted in increased susceptibility to glyphosate or the activating effect of ammonium sulfate may have been more direct.

A summary of effects of time of application of herbicides- The proper time to control tuberous weeds such as nutsedges is extremely important because the tuberization process must be stopped before tubers start to form and develop. Developed tubers are usually unaffected by herbicides (116). Newly developed leaves are physiologically and anatomically younger and would be expected to more readily absorb foliar applied herbicides (32, 187), while young plants have more active physiological sinks. Therefore, timing of postemergence herbicide applications relative to tuberization is crucial for overall control of yellow and purple nutsedge (113, 116, 144).

Susceptibility of nutsedge to 2,4-D has been shown to

decrease with increased plant age (60, 63), possibly because 2,4-D would not translocate to mature tissues of older plants. Control improved with multiple applications of 2,4-D when nutsedge plants were first treated at an early stage development (53, 63, 131) as compared to applications beginning only at a later stage (21). Timing of amitrole applications were critical because nutsedge was most susceptible when applied 4 weeks after initial emergence, whereas nutsedge appeared resistent six weeks after emergence (62). herbicide (62). Applications of paraquat and oxyfluorfen prior to tuber formation reduced tuber numbers dramatically when compared to later applications (100, 116). Some research has indicated that later timings of glyphosate such as advanced prebloom stage provided more effective control (4, 9 170), but most researchers report better control with earlier applications (94, 114, 116, 137, 144, 155). The optimal time for glyphosate application has generally been determined to be four weeks after emergence (74, 114, 116), but may be earlier for plants starting growth in mid-season. In growth chamber and outdoor pot studies tuber initiation occurred in 30-day-old nutsedge plants (113, 116), a time when maximum control can be achieved with postemergence herbicides.

In summary, basic efforts for nutsedge control should be directed at inhibiting sprouting of parent tubers and inhibiting new tuber formation. Among many herbicides representing the several chemical families, it is apparent that herbicides which act as root, shoot, and photosynthesis inhibitiors, and those that

interfere with plant amino-acid metabolism are the most effective for nutsedge control. Mature nutsedge tubers are generally unaffected by herbicides. Important points to consider if one is to maximize the performance of postemergence herbicides for control of nutsedge include (a)- foliage penetration, absorption, and translocation of herbicides will be enhanced if applied at the proper growth stage of yellow nutsedge; (b)- herbicidal activity can be increased via use of a synergistic mixture of herbicides; and (c)- an understanding of the importance of environmental conditions and the physiological status of purple and yellow nutsedge plants can be used to advantage. A means of absolute control of nutsedge using herbicides has not been found, although available information indicates that a rational level of control of this weed is possible.

Development of new and selective herbicides are still necessary for improved nutsedge control in the future. Preconditioning nutsedge with applications of growth regulators or other chemicals for increasing subsequent sensitivity to herbicides remains an interesting possibility (11, 75, 152).

Chapter 3

THE ROLE OF YELLOW NUTSEDGE (*Cyperus esculentus* L.) GROWTH STAGE ON TIMING, PHYTOTOXICITY AND CONTROL WITH GLYPHOSATE

WELINGTON PEREIRA and GARVIN CRABTREE

Abstract. Growth chamber, greenhouse, and outdoor pot experiments were conducted from 1982 through 1984 to characterize stages of tuberization of yellow nutsedge (*Cyperus esculentus* L. var *leptostachyus* Boeck. #[a] CYPES) relative to age of the plant, to determine the effect of glyphosate [*N*-(phosphonomethyl)glycine] relative to nutsedge growth stage, to ascertain if glyphosate inhibited tuber formation, and to examine the possibility of split applications for increasing glyphosate effectiveness in controlling nutsedge. The role of the tuberization process was related to glyphosate phytotoxicity. Under experimental conditions, the first tuber initiation occurred about 30 days after nutsedge emerged. New tuber production started to decline about 14 days after maximum leaf area was reached, suggesting that new tuber formation is linked to active growth of the plant foliage. Tuberization was a continuous process, but was modulated by plant age, as well as by environmental conditions. Glyphosate toxicity to nutsedge was dependent on growth stage, with plants being more susceptible at an early growth stage than at late stages. The best time to apply glyphosate was at initiation of the first tubers. At that time nutsedge plants were less tolerant to the herbicide and tuber production was significantly reduced.

Split applications 10 days apart increased response to glyphosate at an early nutsedge growth stage. Glyphosate was very effective in killing rhizomes and blocking new tuber formation, although this herbicide was not effective in controlling tuberization in those rhizomes in which the tuberization process was visibly underway. Therefore, stage of nutsedge tuberization is a crucial point for timing glyphosate applications.

Additional index words - CYPES, tuberization, tuber formation, dry matter accumulation, growth analysis, herbicide, control, timing applications.

[a]Letters following this symbol are a WSSA-approved computer code from Important Weeds of the World, 3rd. ed., 1983. Available from WSSA, 309 West Clark St. Champaign, IL 61820.

Introduction

Generally, the best time to apply herbicides to control perennial plants is shortly before bloom stage. In contrast, the proper time to apply herbicides for the control of a tuberous perennial might be earlier because the tuberization process must be stopped before tubers develop and mature, since mature tubers are usually unaffected by herbicides (18).

There has been some research to indicate that timings of glyphosate applications to nutsedge as late as advanced pre-bloom stage provide more effective control (1, 4) and some reports of better control with earlier applications (14, 19, 20, 21). It was not clear whether these differences were due to size of the plants or the effect of short days as the growing season progressed. Also, these reports did not include characterization of nutsegde tuberization at time of herbicide treatment. Hunt and Lindscott (11) reported significant interactions between season, glyphosate rates, and time of application on viable tuber production. As the season progressed, less glyphosate was needed to obtain a comparable reduction in number of tubers. A 2 kg/ha rate significantly reduced production of viable tubers from plants emerging in the 5-week period after May 30, whereas plants emerging before May 30 produced viable tubers at the same application rate.

Also, susceptibility of nutsedge to 2,4-D [(2,4-dichlorophenoxy)acetic acid] has decreased with increased plant age (9), possibly because 2,4-D would not translocate effectively

to the mature tissues in older plants. Early application of 2,4-D, amitrole (1-H-1,2,4-triazol-3-amine), paraquat (1,1'-dimethyl-4-4'-bipyridinium ion), fluorodifen [P-nitrophenyl(α,α,α,-trifluro-2-nitro-p-tolyl)ether, and oxyfluorfen [2-chloro-1-(3-ethoxy-4-nitrophenoxy)-4-(trifluromethyl)benzene] have provided better results in controlling nutsedge as compared to applications at later stages (7, 8, 15, 18).

The objectives of these studies were (a)- to characterize stages of tuberization of yellow nutsedge relative to age of the plant, (b)- to determine the effect of glyphosate relative to nutsedge growth stage, (c)- to ascertin if glyphosate inhibits tuber formation, and (d)- to examine the effect of split applications of glyphosate for the control of nutsedge.

Materials and Methods

Experiments were carried out under growth chamber, greenhouse and outdoor conditions from 1982 through 1984. Nutsedge tubers were collected from a field near Dayton, OR in March 1982, multiplied in the greenhouse and stratified at 4.5 ^{o}C for subsequent studies. Nutsedge plants were grown from single pregerminated tubers in 3-L pots containing a screened mix of equal volumes of soil, sand, and peat. The medium was previously sifted in order to facilitate washing all particles from plants at harvest time for the purpose of assessing herbicide effect on tuber production. Top water was applied as needed and included a weekly application of water soluble 20-20-20 fertilizer. Plants were selected for morphological uniformity, and treatments arranged in a complete randomized design in each experiment. A track-mounted sprayer in the greenhouse was calibrated for about 340 L/ha and used to apply the herbicide. A 0.5% concentration of non-ionic surfactant was included in the spray mixture.

At the end of each experiment soil was washed from the plants and they were separated into parts to determine (a)- number of primary and secondary shoots and leaves; immature (young, white), and mature (large and brown) tubers, (b)- fresh and/or dry weight of shoot and leaves, rhizomes and roots, and tubers, (c)- visual ratings of nutsedge foliage injury with 0 = no effect and 100 = complete kill.

Growth analysis - Three experiments were conducted to characterize the growth and tuberization pattern of yellow nutsedge under experimental conditions. Treatments of experiments 1 and 3 were replicated four times and are listed in Figure 3.2. In experiment 1, nutsedge plants were propagated from pregerminated tubers after treatment with 75 ppm of gibberellic acid (GA_3) for 48 hours, and grown in a growth chamber regulated for 15 hours of daylightlength for 60 days and later for 9 hours for 50 days. Plants were maintained under a photosynthetic photon flux density of about 400 $\mu Em^{-2}s^{-1}$, with 30 ^{o}C and 60% relative humidity day and 20 ^{o}C and 90% relative humidity night conditions. Observations were made with plants harvested and evaluated at 10 day intervals from 30 days after emergence to 110 days. Experiment 2 was carried out under outdoor conditions from June 27, 1983 through October 29, 1983 (Figure 3.1). Nutsedge plants were grown from six single sprout pregerminated tubers in 15-L pots following procedures described above. Plants were harvested and evaluated at 10-day intervals, starting 25 days after emergence and extending to 125-day-old plants. Treatments were replicated four times and are listed in Figure 3.8 in appendix A.

In experiment 3, plants were grown under outdoor conditions from June 10, 1984 through October 13, 1984 (Figure 3.1), following the general procedures described above. Evaluations were made with plants harvested weekly during a 16-week period.

Timing glyphosate applications relative to nutsedge growth stages- Three trials similar to the growth analysis experiments were

established to determine the effect of glyphosate as influenced by nutsedge growth stage, and to investigate the effect of split applications on glyphosate activity on nutsedge. Included were experiments 4 and 5 conducted in a growth chamber under conditions similar to experiment 1. Experiment 6 was carried out under outdoor conditions, similar to experiment 3. Treatments for experiments 4 and 5 are listed in Figure 3.5 and Table 3.2, respectively, and treatments for experiment 6 are in Table 3.3. The amount of phytotoxicity after glyphosate applications was evaluated in experiment 4 at 30 days after each treatment time and at 110 days after emergence. Treatment and evaluation times are listed in Table 3.1.

The effect of glyphosate on tuber formation - Individual yellow nutsedge plants were propagated from pregerminated tubers in 3-L sand culture pots and grown with half strength Hoagland (10) solution in a greenhouse with supplemental lighting in the morning and evening to maintain a 15 hrs photoperiod during 60 days. Temperatures were maintained at about 28 ^{o}C during day and about 18 ^{o}C at night, during the summer of 1984. Twelve plants selected for morphological uniformity, at 40 days after emergence were washed free of sand and on each plant 10 rhizomes were labeled as to status of tuber development: 5 rhizomes without a new tuber at the tip, and 5 rhizomes with an immature tuber at the tip. Plant were repotted and allowed to grow for 1 day at which time they were either left untreated or treated with 1.5 kg ai/ha of glyphosate. Forty-five days after treatment, plants were

harvested, rewashed free of sand and labeled rhizomes were reevaluated for glyphosate effect on tuber formation. The experiment was repeated.

Results and Discussion

Growth analysis-Individual processes of tuberization have been defined as tuber initiation and tuber development (22), but nutsedge growth stages have not been described in relation to tuberization. Since tuber initiation seemed to be related to the age of plant, days after emergence was used to represent different stages of plant growth and as a reference for treatment application. New tuber formation started about 30 days after nutsedge emerged, with a higher production rate occurring after 42 days and a peak at about 80 days after emergence (Figure 3.2). Plants grown at high temperature (experiment 1) appeared to have accelerated tuber development, and an increase in the number of tubers maturing at an early growth stage (Figure 3.2). Conversely, under field conditons, at Hood River, OR, it was observed that nutsedge plants started to grow in the spring and took about 2 months from emergence to initiate tuber production. Late in the summer, growth slowed and eventually complete kill of the top occurred with fall frost. In experiment 3, maximum leaf area was produced 70 days after plants emerged, followed by progressive senescence and loss of leaf area (Figure 3.3). New tuber production started to decline about 14 days after maximum leaf area (Figure 3.3), suggesting that new tuber formation is linked to active growth of the plant foliage. Tuberization was a continuous process, but was modulated by plant age (Figures 3.2) and environmental conditions such as temperature and photoperiod (2, 6). Under appropriate environmental conditions and

physiological status of the plant tubers are differentiated from meristems at the rhizome tip (3). Normally, rhizomes carrying an immature tuber appeared fresh and were apparently physiologically active, but those rhizomes with mature tubers were generally brown and senescent and appeared to be less active metabolically. Short, thin roots were present on well developed tubers, possibly contributing to their final development and maturation. The rate of new tuber formation was unaffected when photoperiod shifted from long days (after 60 days) to short days (Figure 3.2A), although short days have been reported to enhance differentiation of rhizomes into tubers (12).

About one third of the total dry matter was found in tubers at the end of the season (Figure 3.4). These results substantiate work reported by Jordan (13). Similarly, the total underground (roots, rhizomes, and tubers) dry matter was about two thirds of the total plant dry matter (Figure 3.4). Dry matter was evenly distributed between the top and underground portions of nutsedge plants up to about 70 days after emergence, then the later became significantly greater than the top (Figure 3.4). After only 50 days after emergence the dry weight of tubers contributed significantly to the total underground dry matter (Figures 3.4). Dry matter accumulation in roots and rhizomes was slightly increased at the end of the season, in experiment 3. Coincidentally, 95 day old plants grown in a growth chamber showed a similar response when they were moved to lower temperatures (mean reduction about 14 ^{o}C) of outdoor conditions (Figure 3.4A). This suggests that roots and rhizomes still can grow even under

relatively unfavorable conditions for top growth. The data of experiment 2 are included in appendix A. Results were generally similar to experiments 1 and 3.

Timing glyphosate applications relative to nutsedge growth stages- Glyphosate toxicity to foliage was first noticed and more evident on those new (secondary or higher order) shoots that were present at treatment time. This supports previous results indicating that glyphosate accumulates in the meristematic regions of plants (55). Significant reductions in shoot numbers were observed when glyphosate was applied 30 to 50 days after emergence (Figure 3.5A). New shoots were significantly suppressed or killed until 50 days after emergence, but primary shoots were totally killed only on 30 day old plants (Figure 3.5A). This indicates that there is a greater effect from this herbicide on actively growing plant parts at early growth stages. Similarly, dry weight of shoot and leaves was significantly reduced by glyphosate applied 50 days after emergence of the plant (Figure 3.6A). Plants older than 60 days appeared more tolerant to the same rate of herbicide application.

Glyphosate toxicity to nutsedge underground parts was evidently similarly related to growth stage (Figures 3.5B, 3.6B, and 3.7). Glyphosate applications provided significant reductions in tuberization at all growth stages as compared to untreated plants, although more mature plants at the time of herbicide treatment were less responsive (Figures 3.5B and 3.6B). Greater than 90% reduction in tuberization was obtained if glyphosate was applied no later than 50 days after plant emergence, and

conversely, less than 61% if applied later (Figure 3.5B). Similar to its effect on new shoot production, glyphosate inhibited new tuber formation in nutsedge plants until about 50 days after emergence. This would indicate that the first tuber initiation growth stage is the best time to apply glyphosate, because the plant is apparently at the most sensitive stage and has produced no mature tubers (Figure 3.5B). Ability of nutsedge plants to survive glyphosate applications was evident by the increased dry weight accumulation in roots and rhizomes, but only on those plants treated at late stages (Figure 3.7). Since plant age influenced the response to glyphosate and stages of tuberization was dependent on age of nutsedge plants (Figures 3.2 and 3.5B) in that older plants had a large proportion of mature rhizomes and tubers (Figure 3.4), it would seem that glyphosate activity was generally influenced by physiological status of the plant. These observations substantiate work reported by Zandstra and Nishimoto (23) in which age of purple nutsedge did not appear to be the cause of differences in levels of control with glyphosate, but these differences appeared to be related to the physiological status of the purple nutsedge plants. Those authors concluded that the best time to apply glyphosate for purple nutsedge control was when a maximum number of newly produced rhizomes and tubers were connected to healthy foliage.

The fact that yellow nutsedge at early growth stages was more susceptible to glyphosate than at late stages substantiates some previous observations (1, 4), but contradicts others (14, 19, 20,

21) that support the hypothesis that the best time, as a general rule, to apply a postemergence herbicide to control perennial plants is just before bloom stage. This study provides evidence that the best time for glyphosate application is at about the first tuber initiation stage.

The amount of phytotoxicity (expressed as number and weight of shoots and tubers) after glyphosate applications was significantly affected by plant age at treatment time but this response was the same regardless of time of evaluation (Table 3.1). This would suggest there was no further development of toxicity of glyphosate to the nutsedge shoots and underground (tubers, roots, and rhizomes) parts 30 or more days after treatment.

Nutsedge plants grown under outdoor conditons of lower temperatures (Figure 3.1) than under growth chamber conditions (experiment 1) generally showed less response to glyphosate (Figure 3.5 and Table 3.3) for the same application rate. Many researchers have reported that glyphosate activity was affected by environmental factors such as temperature, relative humidity, light intensity, photoperiod, and soil moisture (1, 5, 16, 17). The physiological status of the plant grown under various environmental conditions probably also contributed to the differences observed.

When compared to a single application of the same total amount, split applications at 10-day intervals increased glyphosate activity at an early nutsedge growth stage by reducing weight of shoots and leaves, and tuber numbers (Table 3.2 and

3.3). This increased activity was probably a result of greater penetration into nutsedge foliage from the duplicated foliar wetting and greater total retention of spray solution on the foliage. This enhanced effect was less evident on older plants (Table 3.3). Tuberization is a continuous process in nutsedge and the sustained translocation of small amounts of glyphosate to the tubers was believed to result in the significant increase in glyphosate activity.

Effect of glyphosate on tuber formation- The effect of glyphosate on nutsedge tuberization depended on the physiological status of the plant at the time of herbicide application. Although glyphosate was very effective in killing rhizomes without tubers, this herbicide was not effective in controlling tuberization in those rhizomes in which the tuberization process was visably underway. Rhizomes with immature tubers at treatment time were able to support continued tuber development and maturation even if the plants were showing some response to the herbicide. These results indicate there is a potential for glyphosate to inhibit yellow nutsedge new tuber formation. Probably most of the reduction in nutsedge tuber production by glyphosate treatments, as reported by different researchers, could be due to the effectiveness in killing rhizomes without tubers and blocking new tuber formation on rhizomes, rather than by the killing action in tubers present at treatment time.

Figure 3.1 Temperatures (^{o}C) at outdoor location of experiments at Corvallis, OR. Values represent means of 5-day intervals during experimental times in 1983 and 1984.

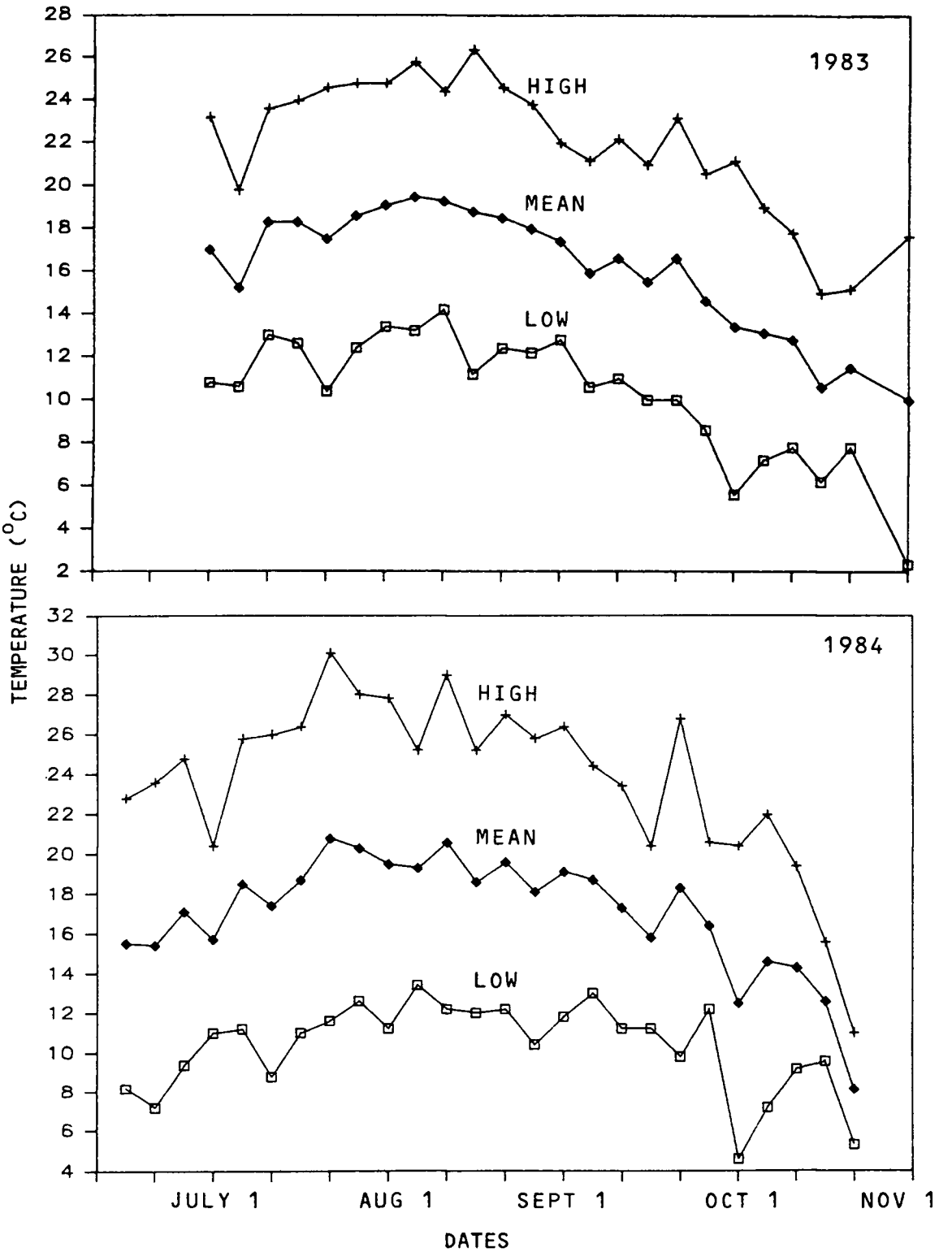
1983
HIGH
MEAN
LOW
1984
HIGH
MEAN
LOW
TEMPERATURE (°C)
JULY 1
AUG 1
SEPT 1
OCT 1
NOV 1
DATES

Figure 3.1

Figure 3.2 Number of immature and total (immature + mature) tubers of yellow nutsedge grown in a growth chamber (A, experiment 1) and under outdoor conditions (B, experiment 3). Error bars = standard deviation of the mean.

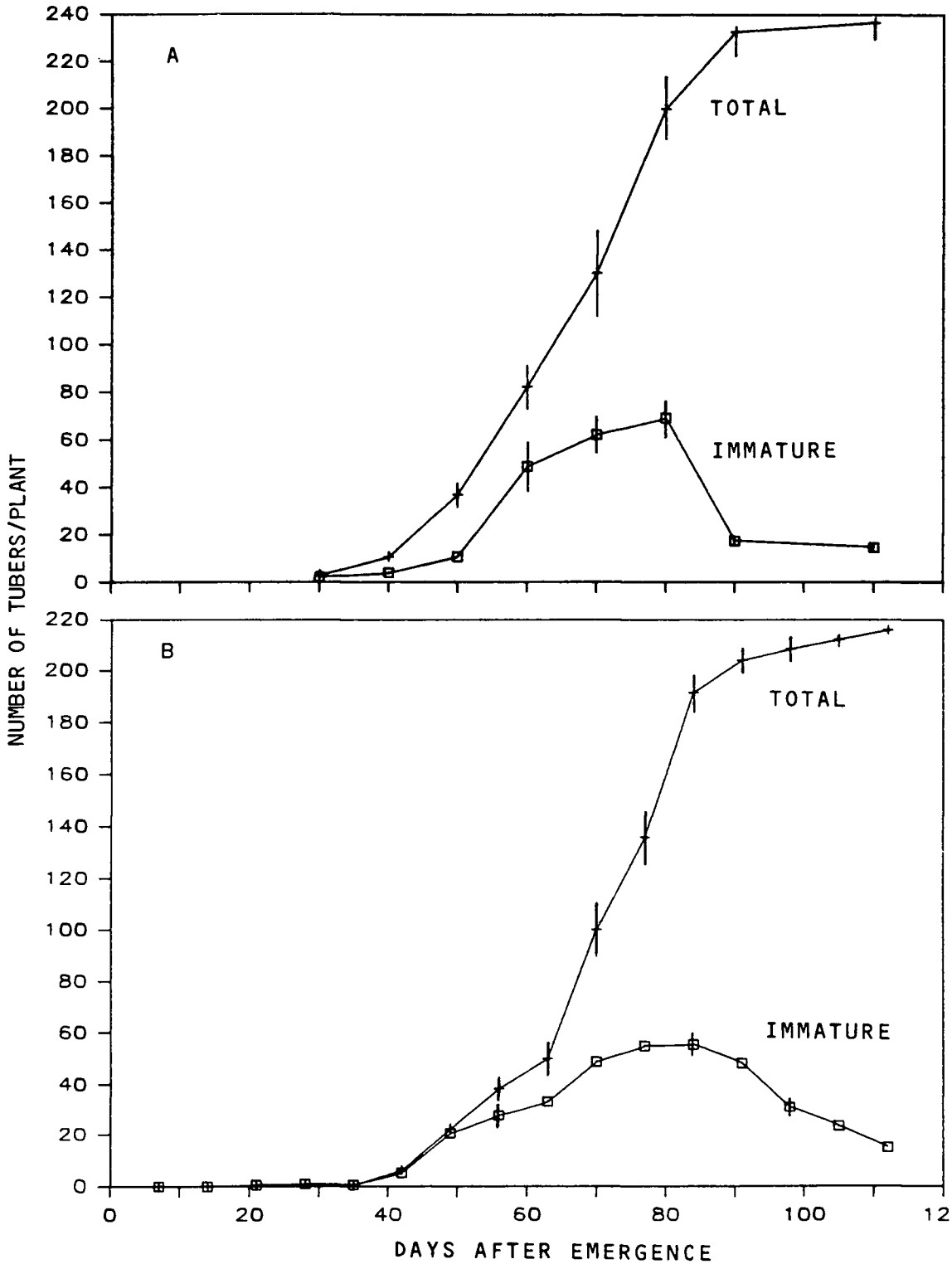
A
TOTAL
IMMATURE
B
TOTAL
IMMATURE
NUMBER OF TUBERS/PLANT
DAYS AFTER EMERGENCE
0
20
40
60
80
100
120
140
160
180
200
220
240

Figure 3.2

Figure 3.3 Leaf number and area (cm^2) of yellow nutsedge plants grown under outdoor conditions at Corvallis, OR in 1984 (Experiment 3). Error bars = standard deviation of the mean.

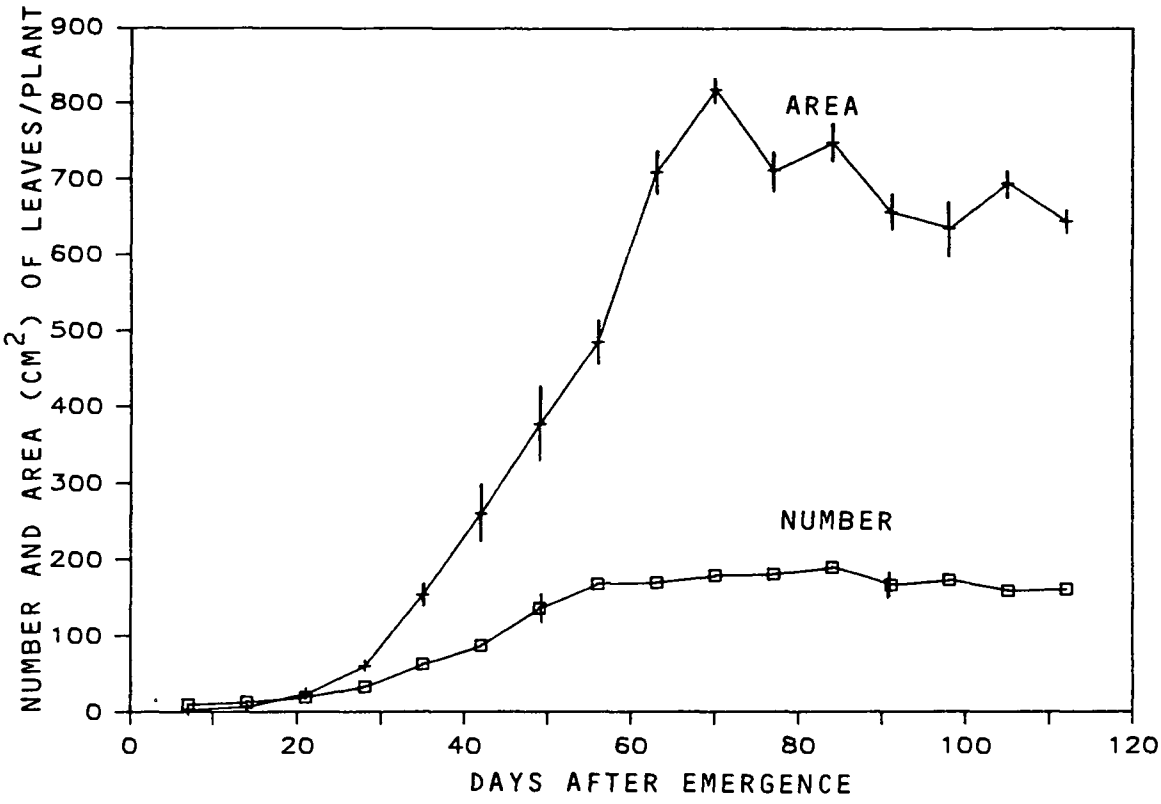
NUMBER AND AREA (CM^2) OF LEAVES/PLANT
900
800
700
600
500
400
300
200
100
0
AREA
NUMBER
0
20
40
60
80
100
120
DAYS AFTER EMERGENCE

Figure 3.3

Figure 3.4 Dry matter accumulation in yellow nutsedge plant parts grown in a growth chamber (A, experiment 1) and under outdoor (B, experiment 3) conditions. Error bars = standard deviation of the mean.

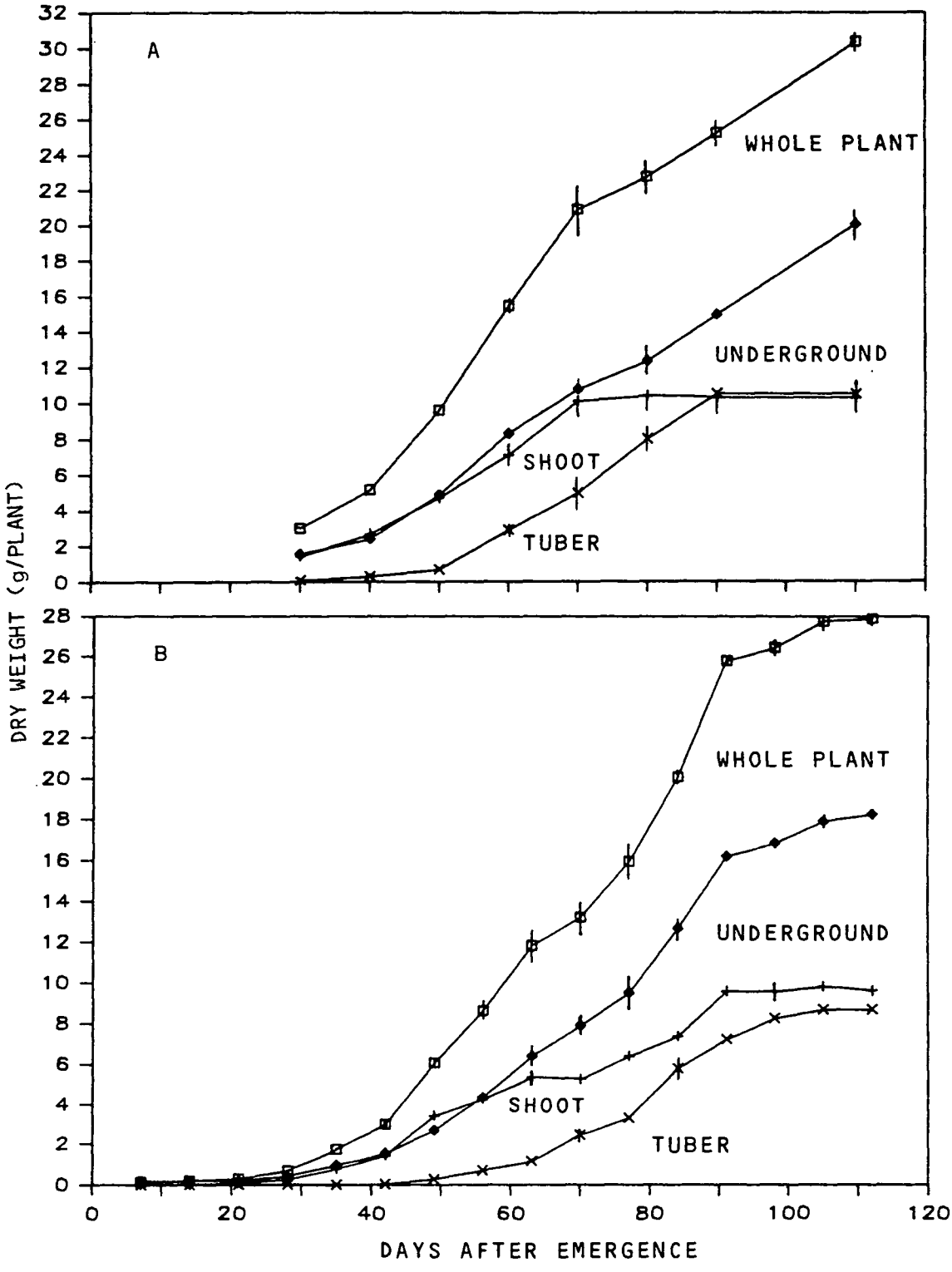
A
WHOLE PLANT
UNDERGROUND
SHOOT
TUBER
B
WHOLE PLANT
UNDERGROUND
SHOOT
TUBER
DRY WEIGHT (g/PLANT)
DAYS AFTER EMERGENCE
0
2
4
6
8
10
12
14
16
18
20
22
24
26
28
30
32
0
20
40
60
80
100
120

Figure 3.4

Figure 3.5 The effect of glyphosate (1 kg ai/ha) on number of new (secondary or higher order) shoots (A) and number of tubers (B) when treated at different nutsedge growth stages. Experiment 4 in a growth chamber.

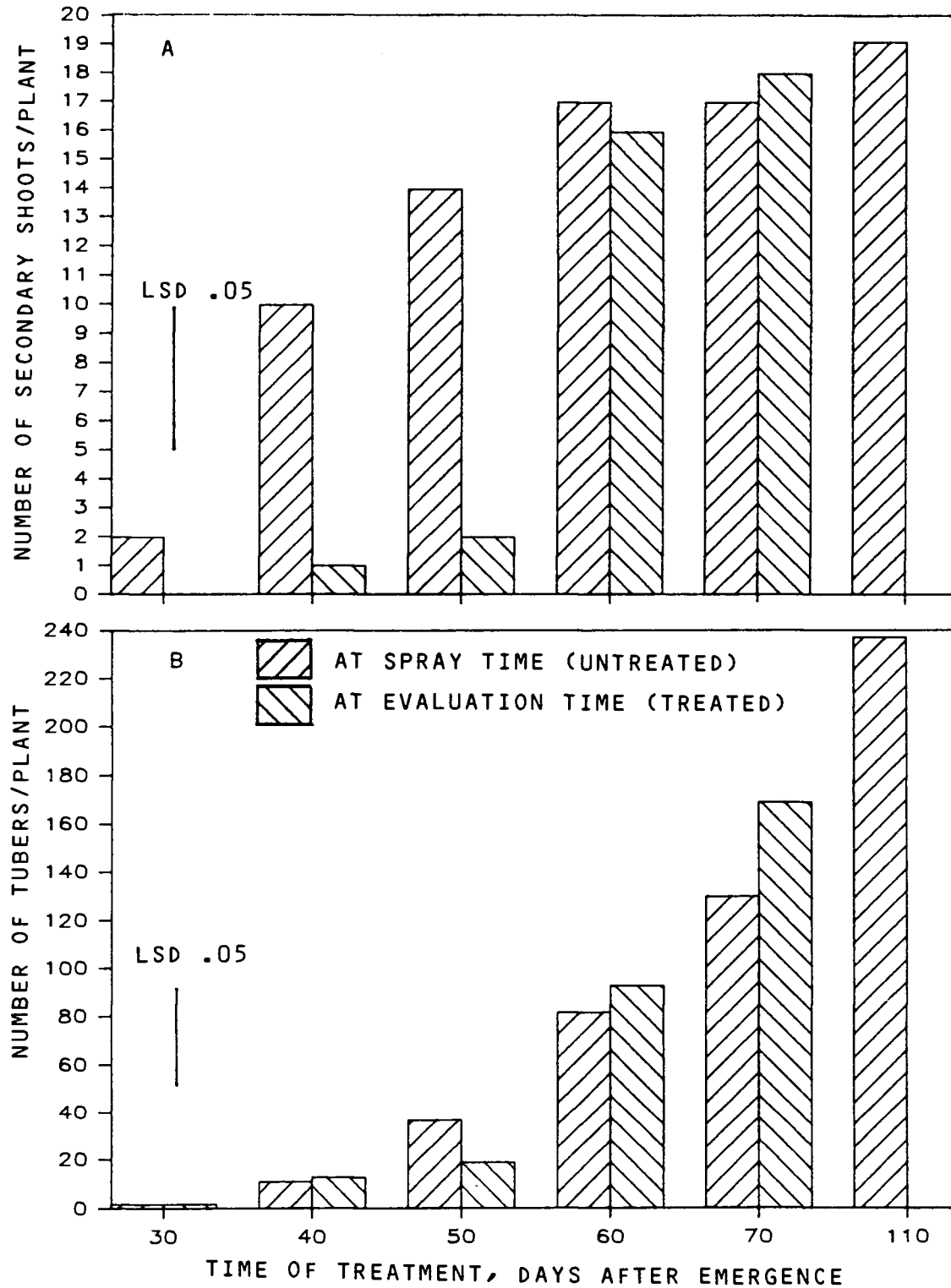
A
NUMBER OF SECONDARY SHOOTS/PLANT
LSD .05
B
AT SPRAY TIME (UNTREATED)
AT EVALUATION TIME (TREATED)
NUMBER OF TUBERS/PLANT
LSD .05
30
40
50
60
70
110
TIME OF TREATMENT, DAYS AFTER EMERGENCE

Figure 3.5

Figure 3.6 The effect of glyphosate (1 kg ai/ha) on dry weight of shoots (A) and fresh weight of tubers (B) relative to different nutsedge growth stages. Experiment 4 in a growth chamber.

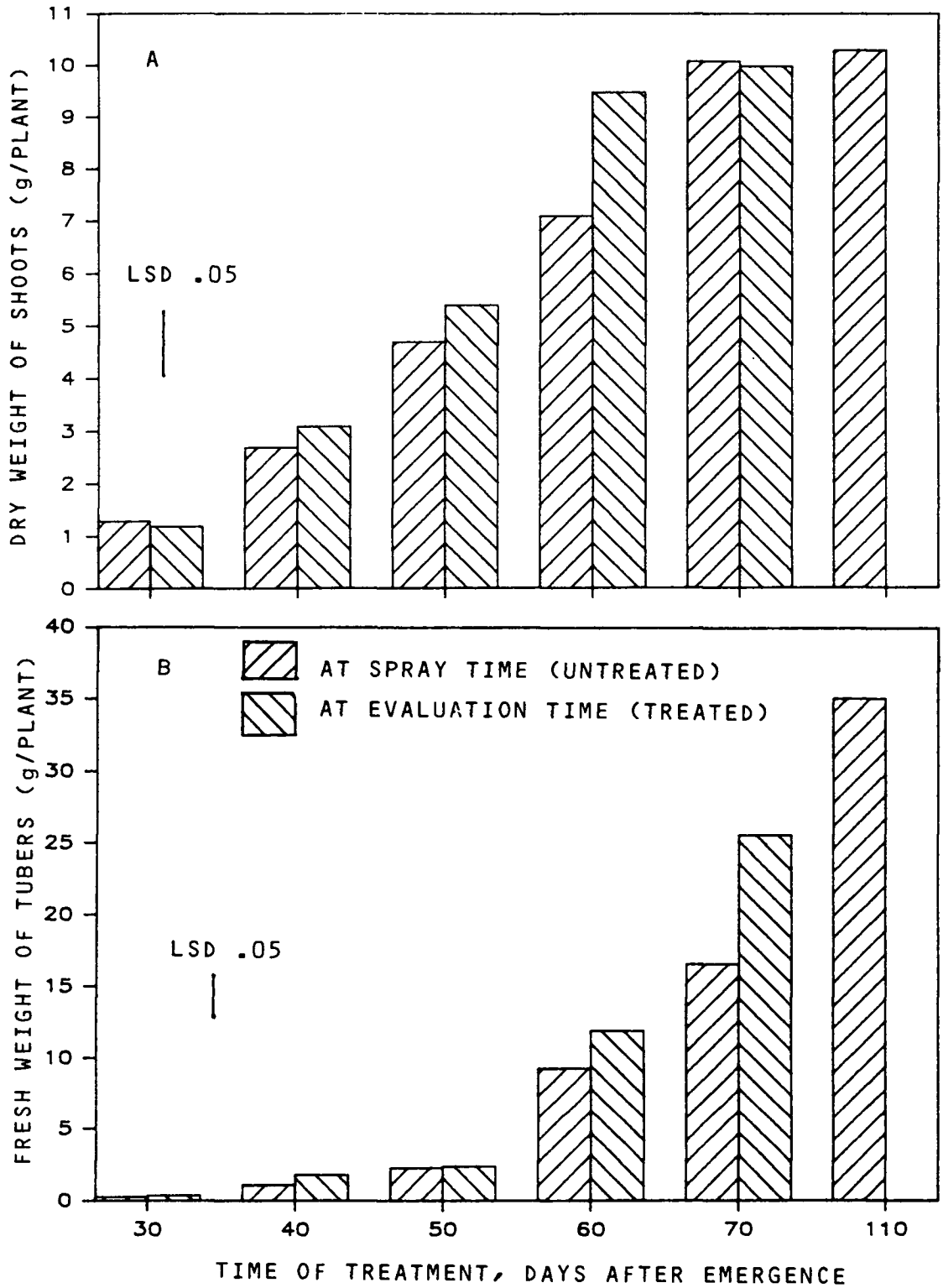
A
DRY WEIGHT OF SHOOTS (g/PLANT)
LSD .05
B
AT SPRAY TIME (UNTREATED)
AT EVALUATION TIME (TREATED)
FRESH WEIGHT OF TUBERS (g/PLANT)
LSD .05
TIME OF TREATMENT, DAYS AFTER EMERGENCE

Figure 3.6

Figure 3.7 The effect of glyphosate (1 kg ai/ha) on dry weight of roots and rhizomes relative to different nutsedge growth stages. Experiment 4 in a growth chamber.

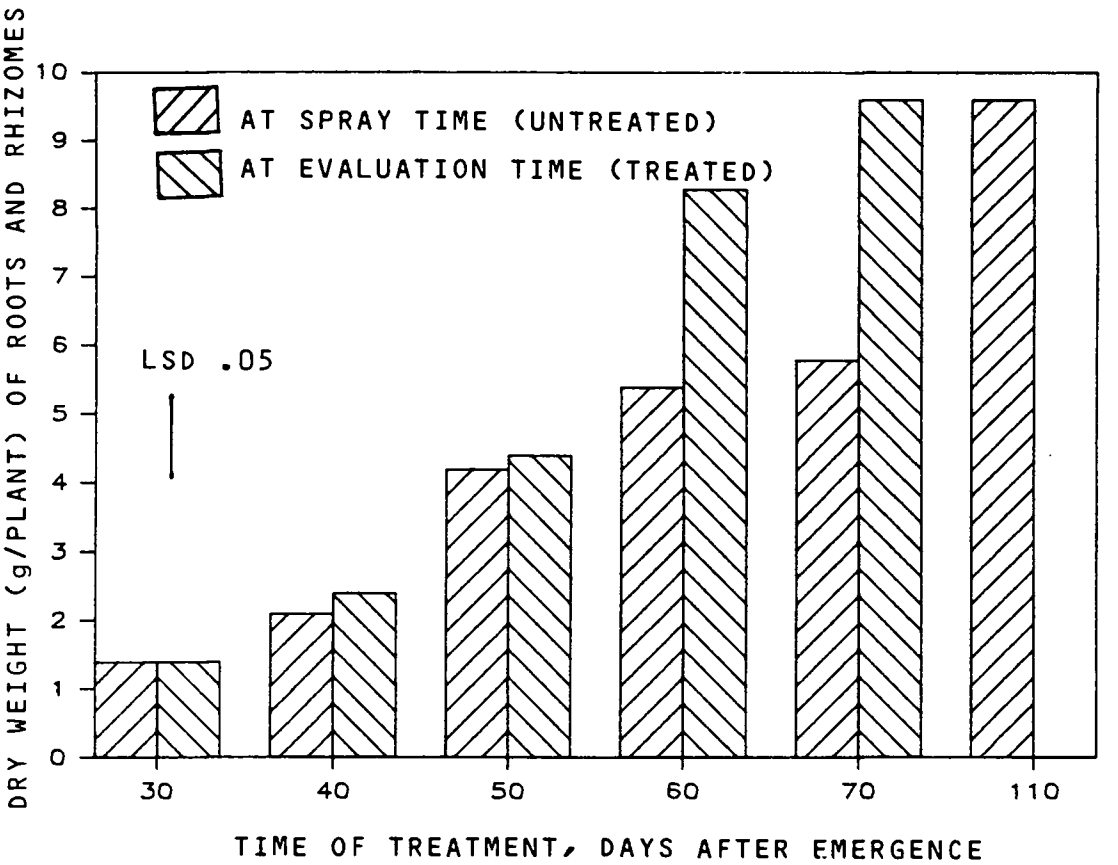
DRY WEIGHT (g/PLANT) OF ROOTS AND RHIZOMES
0
1
2
3
4
5
6
7
8
9
10
AT SPRAY TIME (UNTREATED)
AT EVALUATION TIME (TREATED)
LSD .05
30
40
50
60
70
110
TIME OF TREATMENT, DAYS AFTER EMERGENCE

Figure 3.7

Table 3.1 Early and late evaluations of yellow nutsedge response to glyphosate applied at different growth stages.[a]

Treatment Time(DAE)[b]	Evaluation Time (DAT)[b]	Tuber Number	Tuber Weight	Living Shoots	Shoot Dry Weight
Days	Days	#/plant	g/plant	#/plant	g/plant
30	30	4 d	0.4 d	3 b	2.1 d
	80	3 d	0.4 d	1 b	1.2 d
40	30	9 d	1.2 d	3 b	3.9 d
	70	13 d	1.8 d	3 b	3.1 cd
50	30	15 d	1.6 d	5 b	5.7 b
	60	19 d	2.4 d	4 b	5.4 bc
60	30	81 c	9.7 c	15 a	9.3 a
	50	93 c	11.9 c	18 a	9.5 a
70	30	169 b	25.6 b	21 a	10.3 a
Untreated	110	237 a	35.1 a	21 a	10.3 a

[a]Values within columns followed by the same letter are not significantly different at the 0.05 level according to Tukey's test. Data represent means of 4 replications from experiment 4 under growth chamber conditions.

[b]DAE and DAT = days after emergence and days after treatment, respectively.

Table 3.2 The effect of single and split application of glyphosate on 40-day-old yellow nutsedge plants.[a]

Rate	Tuber Number	Fresh Weight		
		Tubers	Roots & Rhizomes	Shoots & Leaves
kg ai/ha	#/plant	g/plant		
Untreated	474 a	18.8 a	47.1 a	25.2 a
0.75	189 b	13.8 b	16.7 b	11.0 b
0.37/0.37[b]	10 c	1.8 c	11.5 bc	4.7 c
1.50	2 c	0.3 c	5.6 c	2.2 c
0.75/0.75	1 c	0.1 c	5.0 c	2.5 c
2.25	2 c	0.2 c	4.7 c	2.0 c
1.12/1.12	2 c	0.3 c	6.7 c	2.2 c

[a]Values within columns followed by the same letter are not significantly different at the 0.05 level according to Tukey's test. Data represent average of 5 replications from experiment 5 in growth chamber.

[b]/ = split applications at 10 day intervals.

Table 3.3 Yellow nutsedge response to single and split applications of glyphosate as affected by plant growth stage.[a]

Treatment Time (DAE)[b]	Rate	Tuber Number	Tuber Weight	Dry Weight		
				Leaves	Roots & Rhizomes	Total
Days	kg/ha	#/plant	g/plant			
25	0.5	309 a	45.1 a	9.2 abc	9.6 abc	32.3 b
	1.0	291 ab	37.7 bc	7.7 c	6.0 c	25.1 bcd
	0.5/0.5[c]	131 cd	16.7 ef	7.4 c	5.6 c	18.1 d
35	0.5	305 ab	44.2 a	8.9 abc	9.2 abc	31.3 b
	1.0	296 ab	38.7 bc	8.0 bc	7.0 bc	26.7 bc
	0.5/0.5	264 ab	36.0 bcd	8.2 bc	7.9 bc	27.0 bc
45	1.0	294 ab	37.1 bcd	8.5 bc	7.8 bc	27.4 bc
	2.0	190 bc	23.5 def	8.1 bc	7.3 bc	22.5 bcd
	1.0/1.0	118 d	15.7 f	7.9 c	7.3 bc	19.9 cd
55	1.0	308 ab	41.0 bc	9.9 abc	10.5 abc	32.7 b
	2.0	209 bc	28.4 cdef	10.2 abc	9.3 abc	28.1 b
	1.0/1.0	202 bc	29.0 cdef	9.9 abc	10.9 ab	29.6 b
65	1.0	303 ab	39.6 bc	10.4 ab	11.0 ab	33.2 b
	2.0	221 bc	34.4 bcd	10.5 ab	11.9 ab	32.7 b
	1.0/1.0	217 bc	35.6 bcd	9.5 abc	9.7 abc	29.7 b
Untreated	(105)	388 a	57.6 a	11.1 a	13.7 a	42.0 a

[a]Values within columns followed by the same letter are not significantly different at the 0.05 level according to Tukey's test. Data are means of 4 replications from experiment 6 under outdoor conditions at Corvallis, OR, in 1984.

[b]DAE = days after emergence.

[c]/ = split applications at 10 day intervals.

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Chapter 4

INTERACTIONS OF GLYPHOSATE AND OXYFLUORFEN ON YELLOW NUTSEDGE (*Cyperus esculentus* L.) TUBERIZATION AS AFFECTED BY GROWTH STAGES

WELINGTON PEREIRA and GARVIN CRABTREE

Abstract. Research was conducted in a greenhouse, in outdoor pots, and under field conditions in 1983 and 1984 to evaluate interactions of glyphosate [*N*-(phosphomethyl)glycine] and oxyfluorfen [2-chloro-1-(3-ethoxy-4-nitrophenoxy-4-(trifluromethyl)benzene] on yellow nutsedge (*Cyperus esculentus* L. var *leptostachyus* Boeck. #[a] CYPES) tuberization and control. There were significant interactions between glyphosate, oxyfluorfen, and nutsedge growth stages. The response was synergistic between herbicides when they were applied in postemergence mixtures at an early nutsedge growth stage. Nutsedge tolerance to herbicides increased with plant age. Plants treated with oxyfluorfen or with low rates of glyphosate recovered from initial herbicide effects, suggesting that reapplications of herbicides may be required for satisfactory control. Generally, the higher rates of glyphosate that reduced sprouting of tubers, also reduced dry matter accumulation and new tuber formation in ramets originating from tubers of treated plants. There was no interaction between the herbicides on bulking (increase in size) of tubers. Oxyfluorfen did not affect tuber initiation or tuber development, but glyphosate killed young rhizomes and blocked tuber formation. Also, glyphosate significantly reduced the enlargement of immature

tubers present at application time, but allowed for their formation and maturation. Proper timing of control measures for tuberous weeds such as nutsedge is extremely important because the tuberization process must be stopped before new tubers start to form and develop. Therefore, timing of postemergence herbicide applications relative to tuberization is crucial for overall control of yellow nutsedge.

Additional index words - CYPES, herbicides, synergism, antagonism, long-term control, tuber sprouting, dry matter accumulation, tuber formation, and bulking.

[a]Letters following this symbol are a WSSA-approved computer code from Important Weeds of the World, 3rd ed., 1983. Available from WSSA, 309 West Clark St. Champaign, IL 61820.

Introduction

Of the many postemergence herbicides tested for yellow and purple nutsedge control, glyphosate has the greatest potential for suppressing resprouting of parent tubers (3, 7, 11, 14) and for inhibiting new tuber formation when applied at the tuber initiation stage of yellow nutsedge (17). Young and actively growing nutsedge plants (12, 16, 17, 19, 20, 22) were more susceptible to glyphosate than older ones (1, 3). Different amounts of glyphosate toxicity were observed in nutsedge plants grown under different environmental conditions (15, 17). Under field conditions nutsedge regrew from sprouting of dormant tubers after herbicide application or from basal bulbs which escaped the effect of the herbicide (7, 15) and reapplications of glyphosate were often necessary to substantially reduce plant populations (25).

The use of a mixture of herbicides will often maximize the performance of postemergence herbicides. Thus, glyphosate has been applied to nutsedge in combination with other herbicides and non-herbicidal additives. In mixtures with glyphosate most herbicides, especially those which inhibit photosynthesis, produced antagonistic effects (1, 20). In contrast, 2,4-D amine or amitrole provided additive effects (20). Ryan (18) reported that dormant plants of several coniferous species were severely injured by combinations of glyphosate and oxyfluorfen with rates that did not cause serious injury when either herbicide was applied alone. Pereira and Crabtree (16, 17) also preliminarily reported that a combination of glyphosate with oxyfluorfen killed nutsedge foliage

faster than when these herbicides were applied alone.

The objectives of this study were: (a)- to determine the combining effects of glyphosate and oxyfluorfen on yellow nutsedge tuberization, (b)- to study herbicide interactions as influenced by plant age, (c)- to evaluate the long-term herbicidal control of yellow nutsedge by reducing new tuber production and tuber survival, (d)- to determine if herbicides affect new tuber sprouting as well as growth of ramets from tubers of herbicide treated plants, and (e)- to ascertain if either glyphosate or oxyfluorfen affects bulking of tubers.

Materials and Methods

Greenhouse and outdoor studies- Yellow nutsedge tubers were collected from a field near Dayton, OR in spring of 1982, multiplied in the greenhouse and stratified at 4.5 ^{o}C for subsequent studies. Nutsedge plants were grown from single sprout pregerminated tubers in 3-L pots containing a screened mix of equal volumes of soil, sand, and peat. The medium was previously sifted in order to facilitate washing all particles from plants at the harvest time for the purpose of assessing herbicide effect on tuber production. Top water was applied as needed and included a weekly application of water soluble 20-20-20 fertilizer. Plants were selected for morphological uniformity and treatments arranged in a complete randomized design for experiments 1 and 2 under greenhouse and outdoor conditions, respectively. A track-mounted greenhouse sprayer used to apply the herbicides was calibrated for the application of 320 L/ha of water. A 0.5% concentration of non-ionic surfactant was included in the spray mixture.

In experiment 1, plants were grown from April through August in the greenhouse with supplemental lighting (photosynthetic photon flux density about 200 $\mu E.m^{-2}.s^{-1}$) in the morning and evening to provide a 15 hrs photoperiod during 60 days. Temperatures were maintained at about 24 ^{o}C during the day and 18 ^{o}C at night. Herbicides were applied 30 days after nutsedge emergence and visual ratings of foliage response to the herbicides were made 50 days after treatment. Living shoots were counted at 50 and 120 days after treatment to determine regrowth. Treatments

consisted of 4 rates of glyphosate (0, 0.4, 0.8, and 1.2 kg ai/ha) and 3 rates of oxyfluorfen (0, 0.4, and 1.2 kg ai/ha) in a factorial arrangement with 4 replications. At 150 days soil was washed from the plants and they were separated into parts to determine 1- number of primary and secondary shoots; immature (young, white) tubers, and mature (large, and brown) tubers, 2- fresh and/or dry weight of shoots; rhizomes and roots; and tubers, and 3- visual ratings of nutsedge foliage survival with 100 = no effect and 0 = complete kill.

Experiment 2 was conducted from July 5, 1983 through October 23, 1983 (Figure 4.1). Plants were grown under outdoor conditions until 110 days after emergence and herbicides were applied at 30 and 60 days after plant emergence. These application timings were used because tuber formation is influenced by age of nutsedge plants and glyphosate activity is a function of physiological status of the plant (17, 25). Four rates of glyphosate (0, 0.5, 1.0, and 1.5 kg ai/ha), 3 rates of oxyfluorfen (0, 0.5, and 1.5 kg ai/ha), and 2 growth stages were included in a factorial arrangement with 4 replications. At 110 days after emergence plants were harvested and evaluated following procedures described for experiment 1.

Field study - In a two year study (experiment 3), herbicides were applied on a natural stand of yellow nutsedge in a 3-year old pear orchard near Hood River, OR, with the objective of examining the long-term influence of herbicides on new tuber production and tuber survival. Plots 1.8 by 1.8 m were randomized in a block

design with four replications. Nutsedge plants started to emerge between the third or fourth week of April 1983 and 1984. Herbicides (Table 4.2) were applied in 325 L/ha water in split applications at 10 days intervals in late June of each year with a CO_2 plot sprayer. Each plot area was broadcast sprayed twice at each time of application. Plots treated in 1983 were kept untreated and evaluated at the end of the second season (1984). In the fall two soil samples were dug from each plot to assess herbicidal effect on tuber number and weight. Tubers were stratified for later tuber survival studies.

Nutsedge tuber sprouting and dry matter accumulation of ramets originating from tubers of treated plants - Tubers collected from treated plants in experiments 1, 2, and 3 were placed in Petri dishes lined with filter paper moistened with 0.03% captan [cis-N-((trichloromethyl)thio)-4-cyclohexene-1-2-dicarboximide] suspension, sealed in plastic bags and stratified at 4.5 ^{o}C for at least 4 weeks. Later in experiment 4 tuber sprouting and new plant growth characteristics were evaluated. A subsample of 100 tubers from each treatment, if available, was put in a germinator (8 hour 30 ^{o}C day, 16 hour 20 ^{o}C night). Sprouted tubers were counted every week for 4 weeks. Tubers that had not sprouted by 4 weeks were cut and visably classified either as dormant if they were firm and without apparent decay, or non-viable if were soft and decayed. From the first sprouted tubers a random sample of 7 ramets from each treatment was planted in Jiffy Mix Plus in a 3-L pot and placed in a greenhouse with conditions as described for

experiment 1. After 40 days shoots produced were counted and media was washed from the plants which were separated into top and underground parts, dried and weighed.

The effect of glyphosate and oxyfluorfen on bulking of yellow nutsdege tubers - Plants were propagated from pregerminated tubers in 3-L sand culture pots and grown with half strength Hoagland solution (9) in greenhouse conditions similar to experiment 1, but with a 28 ^{o}C day and 20 ^{o}C night. At 40 days after emergence plants were selected for morphological uniformity, washed free of sand and segregated into three rhizome tuber maturity classes: (a)- rhizomes without a visable tuber at the tip, (b)- rhizomes with immature (young, white) tubers, and (c)- rhizomes with mature (large, brown) tubers. Each rhizome class was labeled for maturity class and diameter of tubers at the tip measured. Plants were repotted and allowed to grow for 1 day and then treated with glyphosate or oxyfluorfen (1.5 kg ai/ha) alone or in combination, or left untreated. Thirteen rhizomes were labeled on each plant (5 for each of the 2 first maturity classes and 3 for the third class). This experiment 5 was conducted twice using a complete randomized design with 3 and 6 replications, respectively. Three weeks after applying the herbicides plants were harvested, rewashed free of sand, condition of the rhizomes reevaluated and tubers diameter measured. Plants were also divided into sections for dry matter determinations.

Results and Discussion

Greenhouse and outdoor studies - Tuberization of yellow nutsedge is a continous process but is modulated by plant age, as well as enviromental conditions such as temperature and photoperiod (8). The stage of tuberization of the 2 plant ages used in this study (30 and 60 days after emergence) represented distinct growth stages of nutsedge plants, characterized by the first tuber initiation stage and the mature tuber stage, respectively. In experiment 1, all rates of glyphosate, and the highest rate of oxyfluorfen applied on 30-day-old plants caused significant injury to nutsedge foliage, as compared to untreated plants when evaluated 50 days after herbicide application (Figure 4.2 A). At 120 days after treatment there was some plant recovery and no evident effect from oxyfluorfen or from the lower rate of glyphosate(Figure 4.2B). Conversely, combinations of oxyfluorfen rates at lower rates of glyphosate increased number (Figure 4.3) of nutsedge shoots as compared to untreated plants. This appears similar to the result found by Coupland and Caseley (6) who found that sub-lethal doses of glyphosate increased numbers of small shoots in Agropyron repens and suggested that sub-lethal doses of glyphosate can upset apical dominance. After breaking the apical dominance, bud growth and development was stimulated with continued cell division but reduced elongation (2). Oxyfluorfen quickly desiccated yellow nutsedge foliage but resprouting increased the number of shoots present 50 days after herbicide application (Figure 4.3). These results indicate that nutsedge

plants can recover and produce large amount of tubers when they are treated alone with either oxyfluorfen or a low dose of glyphosate. Herbicidal activity of oxyfluorfen is light dependent (13). The toxicity of oxyfluorfen to nutsedge in experiment 1 was relatively low as compared to experiment 2, possibly a result of low light conditions in the greenhouse (photosynthetic photon flux density about 200 $\mu E.m^{-2}.s^{-1}$). Under outdoor conditions, oxyfluorfen quickly desiccated yellow nutsedge foliage and reduced tuber production by as much as 50% when applied to nutsedge at the early growth stage (Figure 4.5).

Results from greenhouse and outdoor experiments (Figures 4.5, 4.6, and Table 4.1) show significant interactions between glyphosate, oxyfluorfen, and nutsedge growth stage. Glyphosate effectiveness on nutsedge was comparatively greater in the greenhouse than outdoors (Figure 4.4 and 4.5). Conversely, oxyfluorfen was more effective on nutsedge under outdoor conditions. This suggests that combinations of these herbicides would be advantageous and could improve results under variable weather conditions. The response of yellow nutsedge to combinations of glyphosate and oxyfuorfen was statistically analyzed according to Colby's (5) 'expected response' method and by using the isobole method of Tammes (21). The response was synergistic when herbicides were applied in postemergence mixtures at the early nutsedge growth stage but generally antagonistic at the late growth stage (Figure 4.6 and Table 4.1). Combinations of glyphosate and oxyfluorfen provided more rapid nutsedge foliage kill when applied at the early stage and

synergistically reduced tuber population (Figures 4.4 and 4.5). Plants were severely injured by the combinations of herbicides at rates that did not cause serious injury with either herbicide alone. Responsiveness to herbicide rates and combinations was greater between 0.4 and 1.0 kg ai/ha of glyphosate in both experiments. Greater foliar chlorosis of the nutsedge plants treated with glyphosate combined with oxyfluorfen than with single applications of the herbicides suggests that addition of oxyfluorfen increased the amount of glyphosate getting into the nutsedge plant parts, especially when the herbicides were applied at the early stage. Tolerance to the combinations of glyphosate and oxyfluorfen increased with plant age, possibly a result of less wetting and leaf penetration of the spray mixture in the older plants (Table 4.1). Generally, application of glyphosate in combination with other herbicides has produced antagonistic effects. Baird et al (1) reported that glyphosate activity was substantially reduced when it was applied in postemergence mixtures with linuron [N'-(3,4-dichlorophenyl)-N-methoxy-N-methylurea], diuron[N'-(3,4-dichlorophenyl)-N,N-dimethylurea], prometryn [N,N'-bis(1-methylethyl)-6-(methylthio)-1,3,5-triazine-2,4-diamine], terbacil [5-chloro-3-(1,1-dimethylethyl)-6-methyl-2,4,(1H,3H)pyrimidinedione], and alachlor [2-chloro-N'-2,6-diethylphenyl-N-(methoxymethyl)acetamide]. In contrast, this antagonism was not observed in another study with mixtures of glyphosate and diuron or simazine [6,-chloro-N-N'-diethyl-1,3,5-triazine,2,4-diamine] (4). Although it is not clear if these

differences can be attributed to plant size or age it is known (17, 25) that the status of tuberization in nutsedge plants is an important factor in the success of nutsedge control measures. For long-term effective control application of herbicides at the late growth stage must be avoided because yellow nutsedge plants are more tolerant to herbicides at that time and plants are in an advanced tuberization stage, bearing mature tubers which will be unaffected by the herbicides. Mature and/or dormant attached tubers have been less metabolically active (23) and generally unresponsive to herbicide applications (17, 25).

Field study - In this 2-year study all herbicide treatments significantly reduced the number and weight of nutsedge tubers formed (Table 4.2). When applied two consecutive years to the same plots glyphosate reduced tuber production to 15% and bentazon [3-(1-methylethyl)-(1H)-2,1,3,-benzothiodiazin-4(3H)one 2,2-dioxide] to 33% of the untreated plots. Comparatively, one application of glyphosate and bentazon reduced average tuber yields to 40% and 68%,respectively, of the untreated plots. These herbicides controlled nutsedge foliage for about 50 days, but it regrew from sprouting of dormant tubers or ramets which escaped the effect of the herbicide, and contributed to the amount of new tuber production (Table 4.2). This suggests that for effective long-term control reapplications of herbicides are required when there is recovery or regrowth. Nutsedge plants emerging in mid-season seemed to require less glyphosate to provide a comparable reduction in tuber numbers (10), however, as previously discussed

(16,17), application of glyphosate at the first tuber initiation stage was reported as the best time to suppress tuberization.

The residual effects in nutsedge tubers from single application of glyphosate or bentazon were apparent in the second growing season with tuber production reduced to 30% and 60%, respectively, of the untreated plots (Table 4.2). Glyphosate and oxyfluorfen applied together killed nutsedge foliage efficiently and regardless of plant regrowth, number and weight of tubers at the end of the season were significantly less than production in plots receiving a single herbicide. These results suggest that yearly reapplications of the herbicides are needed for a period of three or more years in order to achieve rational control of yellow nutsedge tuberization.

Nutsedge tuber sprouting and dry matter accumulation in ramets originating from tubers produced by treated plants - From outdoor and field experiments 2 and 3, respectively, glyphosate reduced sprouting of new tubers originating from nutsedge plants treated at the early stage (Tables 4.3 and 4.4). However, in the outdoor experiment this herbicide did not affect sprouting of new tubers originating from plants treated at the mature tuber stage. This suggests that glyphosate was accumulated in higher toxic doses in tubers attached to plants when treated at the early growth stage as compared to the late stage. At 40 days after emergence new plants had already produced immature (young, white) tubers, as reported previously (17), and glyphosate significantly reduced the formation of new tubers (Table 4.3). This indicates a residual

effect of this herbicide on new tuber production. Generally, the higher rates of glyphosate that reduced sprouting of tubers, also reduced dry matter accumulation of shoots, and roots and rhizomes. These observed effects of glyphosate on tuber sprouting and in dry matter accumulation in new nutsedge ramets may explain in part the residual effect of this herbicide in tubers from treated plants and the reduced tuber production in the second growing season (Table 4.2). Apparently, the herbicide translocated into the new tubers may have affected their sprouting and new plant growth characteristics.

The effect of glyphosate and oxyfluorfen on bulking of yellow nutsedge tubers - The data presented in Table 4.5 are averages of 2 experiments. These data show that there was no interaction between the two herbicides as they affected bulking of nutsedge tubers. Oxyfluorfen did not affect tuber initiation or tuber development, since tubers were formed on rhizomes without a visable tuber at the tip at herbicide application time. This suggests that oxyfluorfen affects tuber production indirectly by temporarily reducing photosynthetic capacity. Single applications of this herbicide reduced tuber production up to 50% when it was applied at the early growth stage (Figure 4.5). Conversely, glyphosate killed young rhizomes without visable tubers at treatment time, thereby blocking tuber formation (Table 4.5). This herbicide has been effective for inhibiting tuberization when applied no later than the time of first tuber initiation (17), but it was not very effective in controlling tuberization in those

rhizomes in which the process was visably underway (Table 4.5). Glyphosate significantly reduced enlargement of immature tubers present at application time, but allowed for their formation and maturation. The development of mature tubers (maturity class 3 in Table 4.5) expressed by their diameter was not significantly affected by the herbicide combinations. Glyphosate also reduced the remaining (from unlabeled rhizomes) number and fresh weight of tubers (Table 4.5), thus suggesting that this herbicide may have suppressed new tuber formation by rhizomes originating after the herbicide application.

Timing of control measures for tuberous weeds such as nutsedge is extremely important because the tuberization process must be stopped before tubers start to form and develop, since developed tubers are less metabolically active (23) and generally unaffected by herbicides (17, 25). Therefore, timing of postemergence herbicide applications relative to tuberization is crucial for overall control of yellow nutsedge.

Figure 4.1 Temperatures (^{o}C) at outdoor experiment location, Corvallis, OR. Values represent means of 5-day intervals.

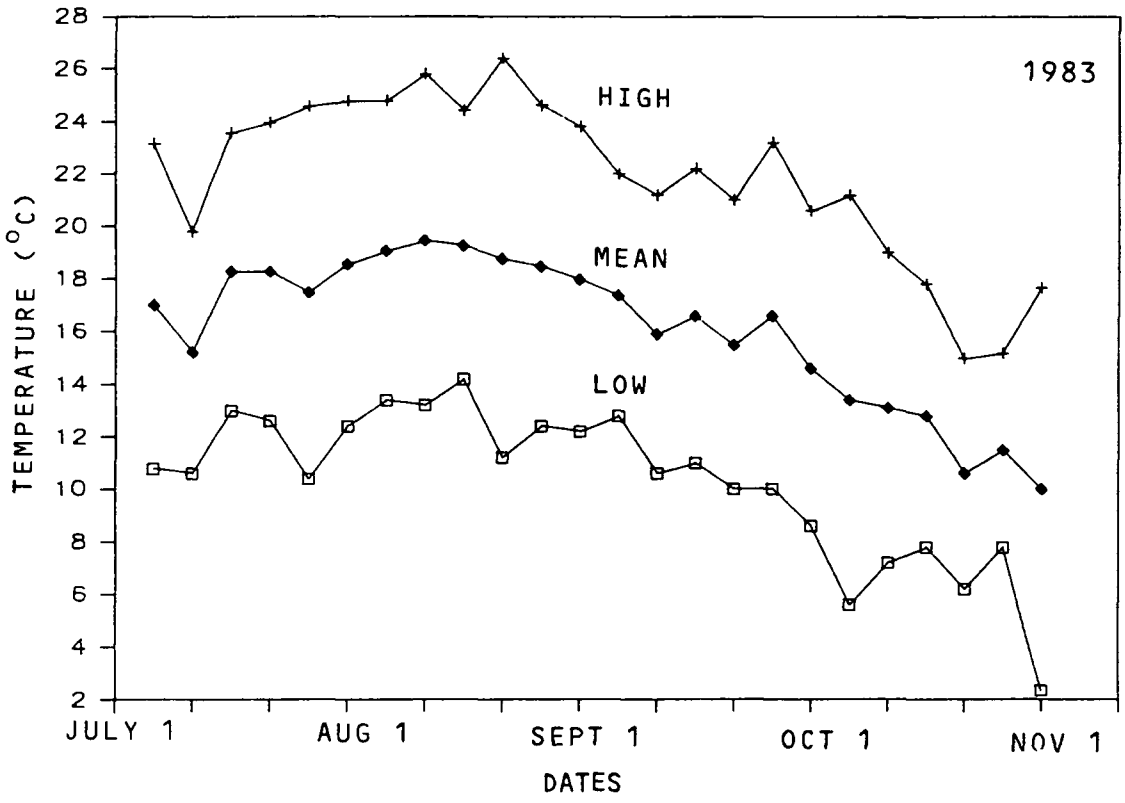
28
26
24
22
20
18
16
14
12
10
8
6
4
2
TEMPERATURE (°C)
1983
HIGH
MEAN
LOW
JULY 1
AUG 1
SEPT 1
OCT 1
NOV 1
DATES

Figure 4.1

Figure 4.2 Influence of glyphosate and oxyfluorfen on nutsedge grown in greenhouse experiment 1. Data represent percent of foliage survival at 50 days (A) and fresh weight of shoots at 120 days (B) after herbicide spray.

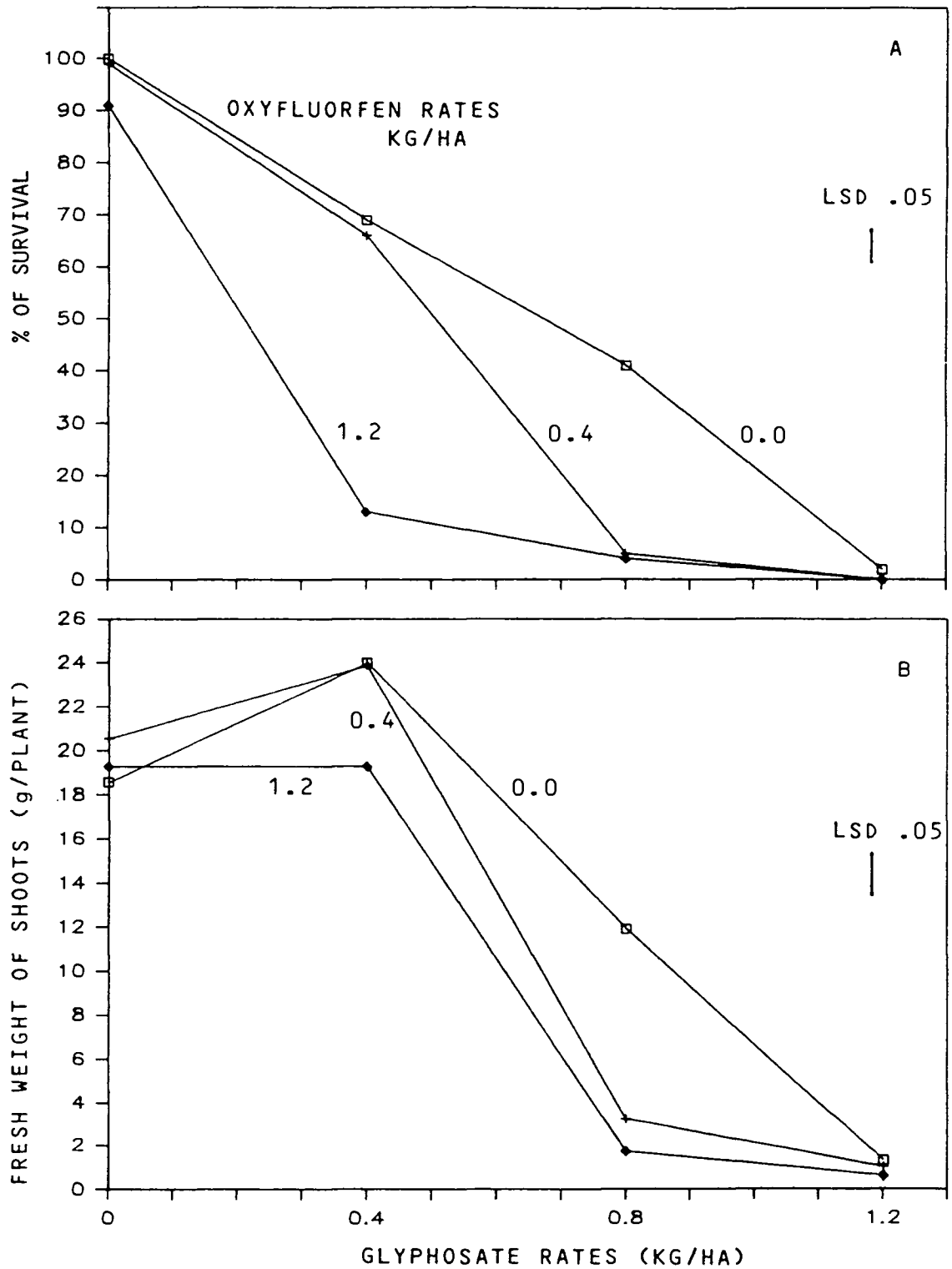
A
OXYFLUORFEN RATES
KG/HA
LSD .05
% OF SURVIVAL
100
90
80
70
60
50
40
30
20
10
0
1.2
0.4
0.0
B
0.4
1.2
0.0
LSD .05
FRESH WEIGHT OF SHOOTS (g/PLANT)
26
24
22
20
18
16
14
12
10
8
6
4
2
0
0
0.4
0.8
1.2
GLYPHOSATE RATES (KG/HA)

Figure 4.2

Figure 4.3 Effect of glyphosate and oxyfluorfen combinations on number of nutsedge shoots, at 50 days and 120 days after herbicide treatment (DAT), greenhouse experiment 1.

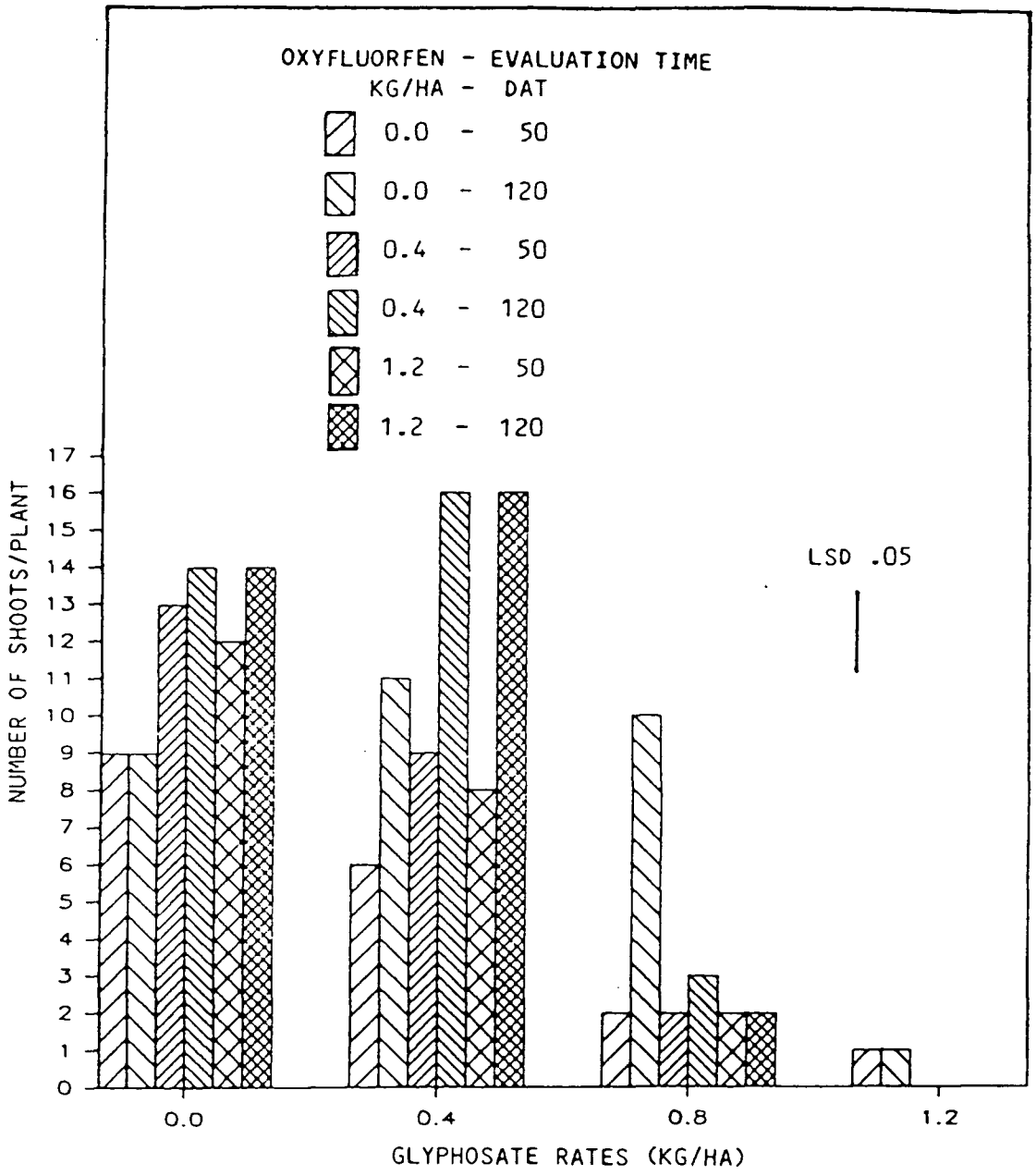
OXYFLUORFEN - EVALUATION TIME
KG/HA - DAT
0.0 - 50
0.0 - 120
0.4 - 50
0.4 - 120
1.2 - 50
1.2 - 120
LSD .05
NUMBER OF SHOOTS/PLANT
0
1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
0.0
0.4
0.8
1.2
GLYPHOSATE RATES (KG/HA)

Figure 4.3

Figure 4.4 Influence of glyphosate and oxyfluorfen on number (A) and fresh weight (B) of nutsedge tubers, greenhouse experiment 1.

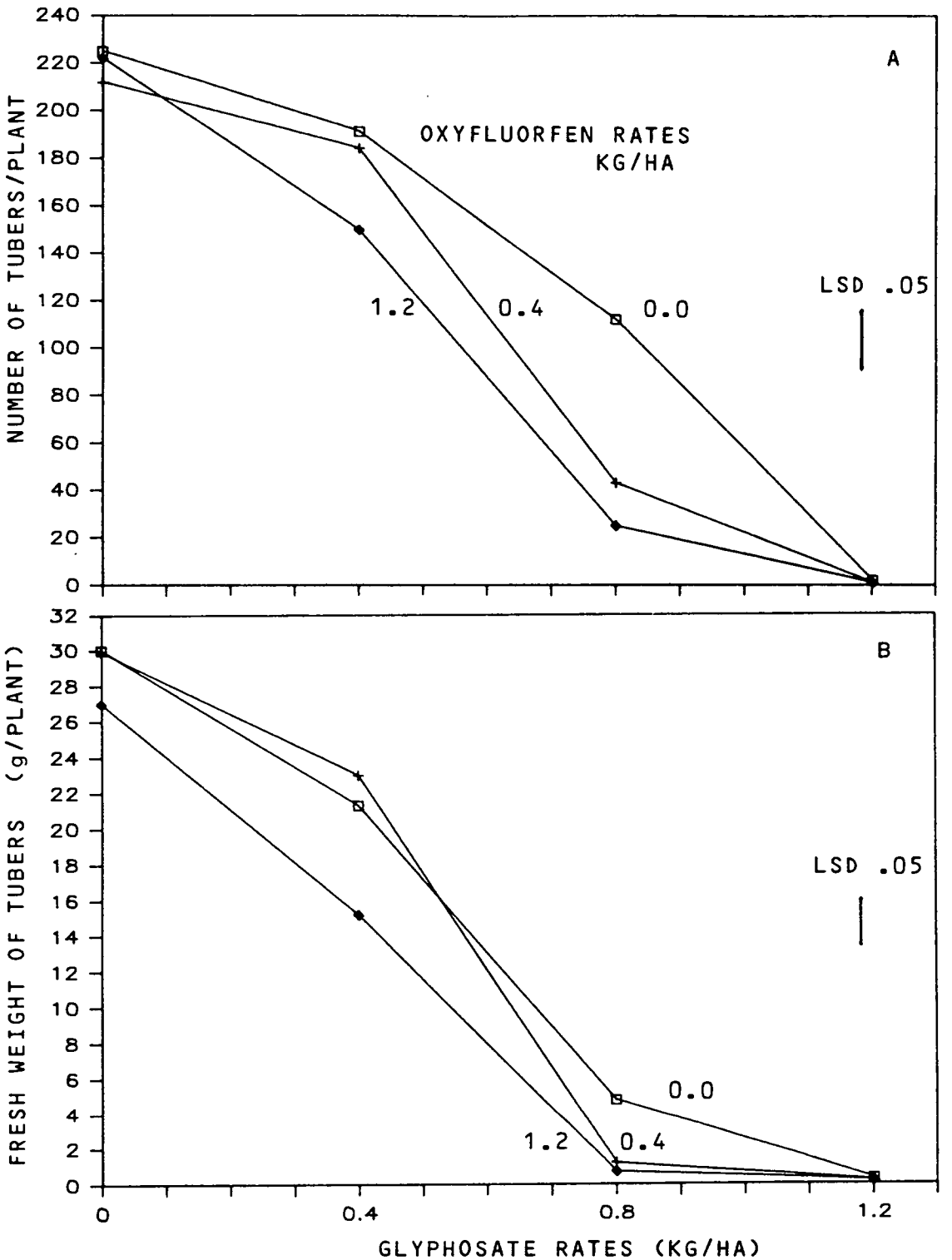
A
OXYFLUORFEN RATES
KG/HA
1.2
0.4
0.0
LSD .05
NUMBER OF TUBERS/PLANT
240
220
200
180
160
140
120
100
80
60
40
20
0
B
LSD .05
0.0
1.2
0.4
FRESH WEIGHT OF TUBERS (g/PLANT)
32
30
28
26
24
22
20
18
16
14
12
10
8
6
4
2
0
0
0.4
0.8
1.2
GLYPHOSATE RATES (KG/HA)

Figure 4.4

Figure 4.5 Interactions of glyphosate and oxyfluorfen on number of yellow nutsedge tubers as affected by 2 growth stages, herbicide application at 30 days and 60 days after emergence (DAE), outdoor experiment 2.

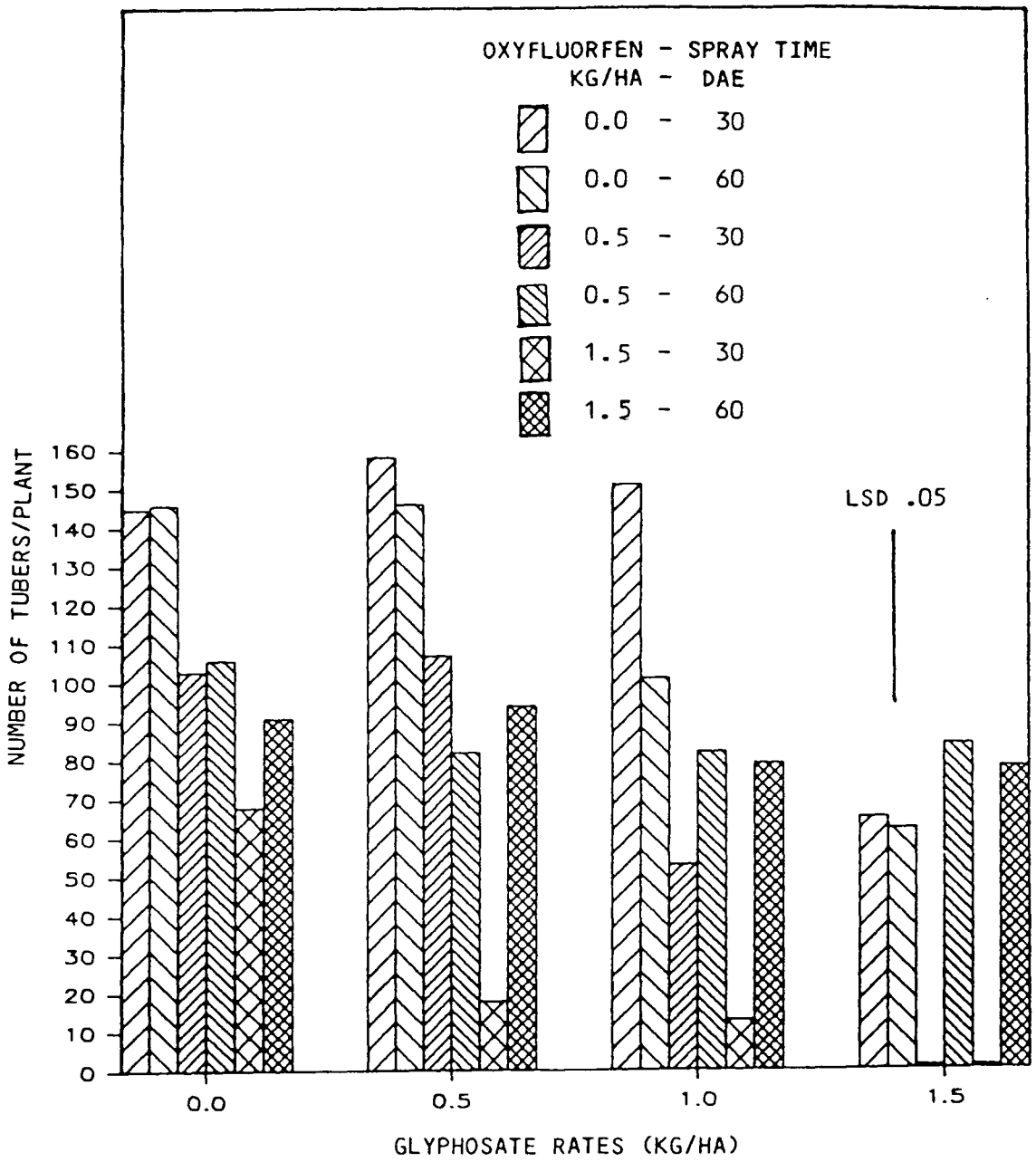
OXYFLUORFEN - SPRAY TIME
KG/HA - DAE
0.0 - 30
0.0 - 60
0.5 - 30
0.5 - 60
1.5 - 30
1.5 - 60
LSD .05
NUMBER OF TUBERS/PLANT
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
0.0
0.5
1.0
1.5
GLYPHOSATE RATES (KG/HA)

Figure 4.5

Figure 4.6 ID_{50} (tuber fresh weight inhibition) isobole of glyphosate and oxyfluorfen combinations sprayed on yellow nutsedge at 2 growth stages obtained by Tammes isobole method (21). Synergism for applications on 30-day- and antagonism on 60-day-old plants are indicated.

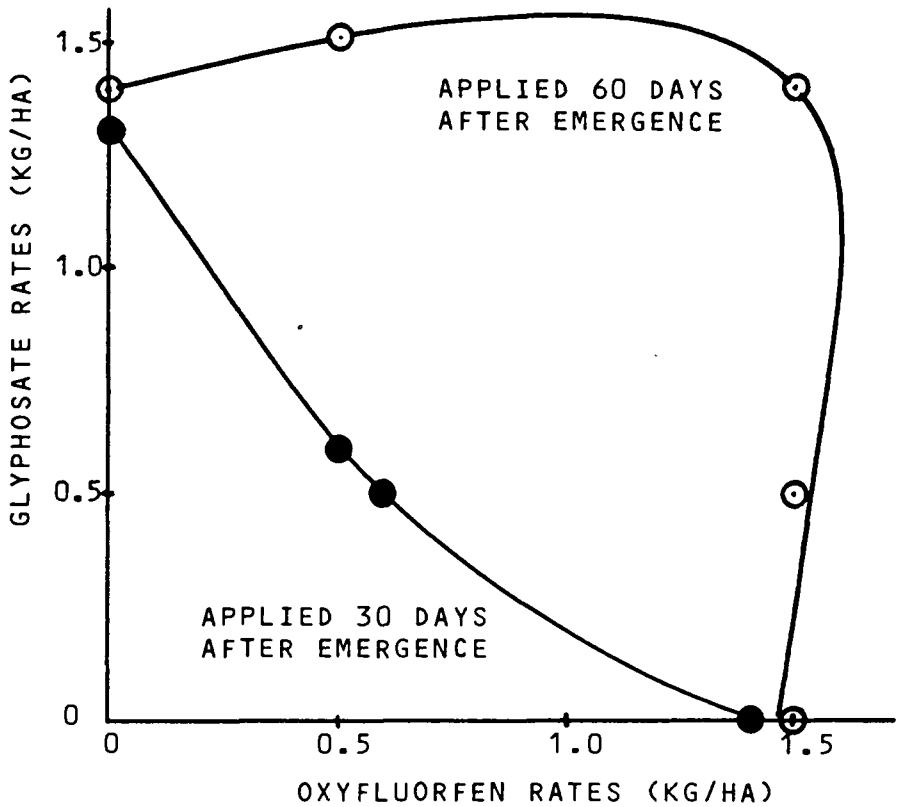
1.5
1.0
0.5
0
GLYPHOSATE RATES (KG/HA)
APPLIED 60 DAYS
AFTER EMERGENCE
APPLIED 30 DAYS
AFTER EMERGENCE
0
0.5
1.0
1.5
OXYFLUORFEN RATES (KG/HA)

FIGURE 4.6

Table 4.1 Interactive effects of glyphosate, oxyfluorfen and growth stage on tuberization of yellow nutsedge.[a]

Days after emergence	Glyphosate kg ai/ha	Oxyfluorfen (kg ai/ha) 0	0.5	1.5
Outdoor conditions				
		%-of-tuberization[b]		
30	0.0	100	69	48
	0.5	97	66 (67)[c]	14 (47)
	1.0	87	38 (60)	8 (42)
	1.5	39	1 (27)	1 (19)
60	0	100	66	71
	0.5	100	51 (66)	66 (71)
	1.0	60	60 (40)	55 (43)
	1.5	41	50 (27)	49 (29)
Greenhouse conditions				
Days after emergence	Glyphosate Rate kg ai/ha	Oxyfluorfen (kg ai/ha) 0	0.4	1.2
		%-of-tuberization		
30	0	100	94	99
	0.4	85	82 (80)	67 (84)
	0.8	50	19 (47)	11 (50)
	1.2	1	0 (1)	0 (1)

[a]Data are averages of 4 replications in each of the outdoor and greenhouse experiments.
[b]Relative to fresh weight as a % of untreated plants.
[c]Expected Colby value (5).

Table 4.2 The effect of herbicide applications over a two year period on tuber production by yellow nutsedge, field experiment 3.

Treatment	Time of application		Rate	1983 Tuber number	1983 Tuber weight	1984 Tuber number	1984 Tuber weight
			kg/ha	#/plot	g/plot	#/plot	g/plot
Check	--	--	--	550	88	587	65
Bentazon	1983	--	1/1[a]	437	79	334	43
Glyphosate	1983	--	1/1	227	31	193	26
	1983	--	2/2	206	30	151	19
Bentazon	1983	1984	1/1+1/1	384	61	193	24
Glyphosate	1983	1984	1/1+1/1	189	30	102	21
	1983	1984	2/2+2/2	163	22	72	12
Bentazon	--	1984	1/1	--	--	340	39
Glyphosate	--	1984	1/1	--	--	236	41
	--	1984	2/2	--	--	202	31
Oxyfluorfen	--	1984	1.5	--	--	416	54
Glyphosate + Oxyfluorfen	--	1984	2+1.5	--	--	186	28
Glyphosate + Oxyfluorfen	--	1984	1/1+1.5	--	--	158	26
LSD (5%)				58	8	46	8

[a]/ = split application at 10 day intervals.

Table 4.3 The effect of glyphosate and oxyfluorfen on sprouting of new nutsedge tubers and dry matter accumulation by 40-day-old ramets originating from tubers produced by 30-day-old treated plants. Outdoor experiment 2.[a]

Treatment	Rate	New tuber sprouting	Ramet dry weight		Immature tubers
			Shoots	Roots & rhizomes	
	kg/ha	-- % --	------- g -------		#/ramet
Untreated	-	100 a	10.1 a	3.3 ab	11 a
Oxyfluorfen	0.4	99 a	10.4 a	3.7 a	8 ab
	1.2	99 a	9.4 a	3.2 ab	8 ab
Glyphosate	0.4	97 a	6.6 ab	2.0 b	6 b
	0.8	89 b	3.1 b	0.7 c	2 c
Glyphosate + Oxyfluorfen	0.4 + 0.4	98 a	9.0 a	2.0 b	6 b
Glyphosate + Oxyfluorfen	0.4 + 1.2	98 a	8.9 a	2.1 b	6 b

[a]Values within columns followed by the same letter are not significantly different at the 0.05 level by using Tukey's test.

Tabel 4.4 The effect of bentazon and glyphosate on sprouting of new nutsedge tubers and dry matter accumulation by 40-day-old ramets originating from tubers produced by treated plants in 1983 under field conditions, experiment 4.[a]

Treatment	Rate	New tuber sprouting	Ramet dry weight	
			Shoots	Roots & rhizomes
	kg/ha	--- % ---	-------- g/pot --------	
Untreated	-	98 ab	10.0 a	2.6 a
Bentazon	1	98 ab	8.6 ab	2.2 ab
	1/1[b]	99 a	7.6 ab	2.1 ab
Glyphosate	1	96 abc	8.0 ab	2.0 ab
	1/1	93 bc	7.4 b	2.0 ab
	2/2	91 c	6.6 b	1.8 b

[a]Values within columns followed by the same letter are not significantly different at the 0.05 level accordinng to Tukey's test.

[b]/ = split application at 10 day intervals.

Table 4.5 The effect of glyphosate and oxyfluorfen on bulking of nutsedge tubers. Data are averages of repeated experiment 5.[a]

Tuber maturity class[b]	Herbicide rate kg ai/ha		Tuber diameter increment[c]	Average tuber diameter
	Glyphosate	Oxyfluorfen	mm	
1	0	0	5.8	5.8
	0	1.5	5.5	5.5
	Mean		5.6 a	5.6 a
	1.5	0	0	0
	1.5	1.5	0	0
	Mean		0 b	0 b
2	0	0	1.2	5.3
	0	1.5	1.3	5.0
	Mean		1.2 a	-
	1.5	0	0.4	4.8
	1.5	1.5	0.6	5.0
	Mean		0.5 b	-
3	0	0	0.2	6.1
	0	1.5	0.3	6.3
	1.5	0	0.2	5.8
	1.5	1.5	0.3	5.7
			Tuber number	Tuber fresh weight
			#/plant	-- g/plant --
Remaining	0	0	41	3.1
	0	1.5	33	2.8
	Mean		37 a	3.0 a
	1.5	0	8	0.5
	1.5	1.5	7	0.5
	Mean		7 b	0.5 b

[a]Values with columns followed by the same letter are not significantly different at the 0.05 level by using Tukey's test.

[b]/1 = rhizomes without a visable tuber at the tip, 2 = rhizomes with immature tubers, 3 = rhizomes with mature tubers, and remaining = unlabeled rhizomes with tubers.

[c]Increase in diameter from treatment time to evaluation time.

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Chapter 5

ABSORPTION, TRANSLOCATION AND TOXICITY OF GLYPHOSATE AND OXYFLUORFEN AS AFFECTED BY GROWTH STAGE OF YELLOW NUTSEDGE (Cyperus esculentus L.)

WELINGTON PEREIRA and GARVIN CRABTREE

Abstract. Nutsedge growth stage and interval between herbicide application and evaluation significantly influenced glyphosate [N-(phosphonomethyl)glycine] and oxyfluorfen [2-chloro-1-(3-ethoxy-4-nitrophenoxy)-4-(trifluromethyl)benzene] absorption and translocation in yellow nutsedge (Cyperus esculentus L. var leptostachyus Boeck. #[a] CYPES). Between 1 and 8 days after application of glyphosate to 30-day-old plants, absorption increased from 18 to 35% of the amount applied and translocation increased from 7 to 27%. Addition of unlabeled oxyfluorfen as a tank mixture with glyphosate increased absorption of ^{14}C-glyphosate to 27% after 1 day and 46% after 8 days and increased translocation into other plant parts to 15% and 42% for the 1- and 8-day periods. For oxyfluorfen alone, total absorption and translocation was about 17% and 2%, respectively. Translocation to tubers accounted for 22% of the 37% translocated to underground (root, rhizomes, and tubers) parts, indicating that tubers are a relatively strong sink for glyphosate accumulation. Translocation of glyphsosate into the tubers was influenced by maturity of the individual tubers. Glyphosate concentration in immature tubers was over three-times greater than in mature ones, indicating that

glyphosate effectiveness in nutsedge tubers was influenced by tuber maturity. Similarly, concentration in young roots and rhizomes was about two-fold that found in comparable mature tissues. Concentration of this herbicide in whole nutsedge plants, decreased from 111 dpm/mg to 42 dpm/mg when applied to 30-day-old and 60-day-old plants, respectively. Comparatively, oxyfluorfen values were as litle as 8 dpm/mg and 3 dpm/mg, respectively. Concentration of ^{14}C-glyphosate was increased in leaves and new tubers by using a combination of these 2 herbicides as compared to glyphosate used alone. Nutsedge regrowth ability was reduced with increasing plant age. Regrowth ability was also significantly reduced by glyphosate and oxyfluorfen applied to 40-day- and 70-day-old plants. Both herbicides reduced chlorophyll content of nutsedge leaves at 6 days after application, however this effect was temporary for oxyfluorfen and only glyphosate reduced nutsedge chlorophyll when measured at 15 days. Also, chlorotic regrowth in nutsedge leaves was observed only from glyphosate treatments.
Additional index words - CYPES, tuberization, tuber formation, herbicide combinations, regrowth abilility, chlorophyll.

[a]Letters following this symbol are a WSSA-approved computer code from Important Weeds of the World, 3 rd. ed., 1983. Available from WSSA, 309 West Clark St., Champaign, IL 61820.

Introduction

To be effective, most herbicides must be translocated within plants from their sites of entry to their sites of activity. Plant characteristics, herbicide chemical and physical properties, and enviromental conditions will determine the degree of absorption and translocation of herbicides (3).

The total amount of glyphosate absorbed and translocated in nutsedge plants varies considerably. Variation in the total amount of absorption can be influenced by many factors such as relative humidity (11, 14, 22), soil moisture (2, 6, 17), and temperature (11, 22) which may affect the rate and/or duration of absorption. Extensive research indicates that glyphosate accumulates in the meristematic regions of foliage , roots, rhizomes, and tubers in a typical source-to-sink pattern (6, 7, 17, 22, 24), although the total translocation to purple nutsedge underground parts may be as little as 3 % of the total applied (6). Zandstra and Nishimoto (24) reported that translocation of labeled glyphosate from a leaf of purple nutsedge to other plant parts increased from 5% of the amount applied at 1 day to 19% at 4 days after application. They observed that the lack of translocation of glyphosate within rhizome-tuber chains was a major reason for poor control. Pereira and Crabtree (16) reported that glyphosate toxicity to yellow nutsedge was related to stage of growth at time of application and that the best control was obtained at the time of first tuber initiation. At that time the nutsedge plants were less tolerant to the herbicides and subsequent tuber production was significantly

reduced.

In looking for increased activity, researchers have applied glyphosate on nutsedge in combination with a range of other herbicides and some non-herbicidal additives. Most herbicides produced antagonistic (4, 20) or additive effects (20). Pereira and Crabtree (16) have reported that a combination of glyphosate and oxyfluorfen provides more rapid yellow nutsedge foliage kill when applied at an early stage of plant development and synergistically reduced production of new tubers and other plant parts. The increased foliage chlorosis resulting from combining these herbicides suggested that the addition of oxyfluorfen increased the amount of glyphosate getting into young nutsedge plants.

Nutsedge plants treated either with oxyfluorfen or with low rates of glyphosate have recovered after showing herbicide injury symptoms (16) suggesting that reapplications of herbicides are required to obtain a satisfactory level of control. Reduction of chlorophyll to 75% of the control and carotenoids to 60% of the control in purple nutsedge leaves was observed 96 hours after application. Achlorophyllus regrowth was reported following sublethal doses of this herbicide (1). With excision eight hours after application, glyphosate reduced regrowth to 47% of that of untreated purple nutsedge (6). Oxyfluorfen exerted its predominant phytotoxic action by damaging membrane components, leading to reduced carotenoid content and subsequent destruction of chlorophylls. Chlorophyll content of plant tissue has been used as a measure of membrane degradation by this herbicide (13).

The overall objectives of these studies were to study the absorption, translocation, and toxicity of glyphosate and oxyfluorfen in yellow nutsedge. Specific objectives were (a)- to determine if combinations of glyphosate and oxyfluorfen increase absorption and translocation of these herbicides in nutsedge plants, (b)- to ascertain if nutsedge growth stage affects the interaction of the herbicides, (c)- to follow the time course distribution of herbicides in different plant parts (leaves, rhizomes and roots, and tubers), and (d)- to evaluate herbicide toxicity as measured by regrowth ability and chlorophyll content of nutsedge plants.

Material and Methods

Absorption and translocation study - Yellow nutsedge plants were propagated from pregerminated tubers with twin sprouts. These plants were grown in 3-L sand culture pots watered with half strength Hoagland (14) solution in a greenhouse. Temperatures were maintained at about 28 ^{o}C during day and 18 ^{o}C at night.

In experiment 1, plants were selected for morphological uniformity and treated with herbicides at 30 and 60 days after emergence. They were first treated with unlabeled glyphosate and oxyfluorfen applied singly or in combinations at 1 kg ai/ha in the evening and after 12 hours treated with labeled materials. Four herbicides treatments: A- unlabeled glyphosate and ^{14}C-glyphosate, B- unlabeled oxyfluorfen and ^{14}C-oxyfluorfen, C- unlabeled glyphosate + oxyfluorfen and ^{14}C-glyphosate, D- unlabeled glyphosate + oxyfluorfen and ^{14}C-oxyfluorfen were used in a complete randomized design with 3 replications. Plants were sampled 1, 4, and 8 days after treatment.

An aqueous solution of the radiolabeled herbicides was prepared (appendix B) so that the 25 µL used on each primary nutsedge twin shoot contained 0.2 µCi of ^{14}C. The labeled materials were applied to the adaxial surface of the fifth leaf from the apex of 30-day-old nutsedge shoots or to the third leaf of 60-day-old shoots. The labeled herbicides were spread between lanolin barriers spaced at 10 cm on the leaf surface. At each sampling time the treated leaf section was excised and immersed in 15 ml of 10% ethanol for glyphosate treatments or in acetonitrile

(water mixture 65:35 v/v) for oxyfluorfen treatments, followed with gentle shaking for 1 minute to wash unabsorbed labeled herbicide from the leaf surface. A 5 ml aliquot of the wash solution was assayed for ^{14}C. Remaining untreated leaves and inflorescences (60-day-old plants only) were cut and placed in individual paper bags and dried immediately. The underground parts were washed free of sand, frozen and separated into the following parts and further dried: (a)- mmature (young, white) tubers, (b)- mature (large, brown) tubers, (c)- parent tubers, (d)- roots and rhizomes. After drying, plant parts were weighed and ground. The radioactivity of plant material was determined by liquid scintillation counting (AQC microprocessor LS 7500) after combustion of solid samples in a Packard oxidizer. Resulting $^{14}CO_2$ was captured in a mixture of Oxisorb- CO_2 and Oxiprep-2. Samples of the wash solution were prepared for counting by mixing with Aquasol liquid scintillation cocktail. The resulting counts per minute (cpm) were corrected for efficiency and converted to desintegrations per minute (dpm). The microprocessor was programed for library program No. 4 with 10 minutes of preset time and/or 95.5% confidence limits for countings.

The radioactivity measured in the nutsedge plant material was assumed to be that of unaltered herbicides as studies on different species have not detected much degradation of ^{14}C glyphosate for periods up to 90 days after leaf application (7, 17).

Regrowth and chlorophyll content of glyphosate and oxyfluorfen

treated nutsedge plants - In experiment 2, plants were propagated in a greenhouse under conditions as described above. Forty days after emergence plants were selected for morphological uniformity and were either left untreated or treated with 1.5 kg ai/ha of glyphosate and oxyfluorfen applied alone or in combination. A complete randomized design with 6 replications was used. Six days after treatment plant tops were removed 1 cm above the soil surface and regrowth measured at 21 and 36 days after herbicide application. Plants were washed free of sand and development of underground parts evaluated.

Experiment 3 was carried out under outdoor conditions in the summer of 1984 at Corvallis, OR (Figure 5.1). Plants were grown from single sprout pregerminated tubers in 3-L pots containing a mix of equal volumes of soil, sand, and peat. Top water was applied as needed and included weekly applications of water soluble 20-20-20 fertilizer. Plants were selected for morphological uniformity and three rates (0, 0.75, and 1.5 kg ai/ha) of glyphosate and oxyfluorfen and 2 (40 and 70 days after emergence) growth stages at time of treatment were included in a factorial arrangement with 4 replications. Plants were cut (similarly to experiment 1) 6 and 15 days after herbicide treatments and regrowth was measured from recuttings at 15-day intervals.

Shoot tissue was collected from plants at 6 days after treatment in experiment 2 and from 6 and 15 days after herbicide application in experiment 3. This plant material was placed on ice

and transported to the laboratory for chlorophyll analysis. Twenty-five 6-mm diameter discs of tissue were cut in the fifth from the center fully developed leaves and chlorophyll content on a leaf area basis was determined. A modified method described by Sestak et al. (19) was used. Tissue was placed in 50 ml centrifuge tubes containing 20 ml of 80% (v/v) aqueous acetone and homogenized by a Polytron Pt10 Brickman Instruments homogenizer at 0 ^{o}C for one minute. The volume of homogenate was completed to 40 ml and centrifuged at 25,000 g for 10 minutes. From the supernatant absorbances at 663 and 645 nm were determined. Chlorophyll a + b were then estimated using the following equation:

$$\text{Chlorophyll a + b } (\mu g/cm^2) = 7.01(A_{663}) + 17.76(A_{645})$$

Results and Discussion

Absorption and translocation study - Plant age and interval between application and sampling significantly influenced the amount of glyphosate and oxyfluorfen absorbed and translocated in yellow nutsedge plants (Figures 5.2 and 5.3). The longer the time after application the greater the absorption observed for both single herbicide applications (Figure 5.2). Generally, it appeared that the amount of glyphosate translocation was dependent on the same factors affecting absorption (Figures 5.2 and 5.3) but total oxyfluorfen translocation was not affected by time following herbicide application. On the average, glyphosate and oxyfluorfen absorption was about 27% and 10% of the amount applied, respectively. Similarly, translocation of the herbicides was about 21% and 1% of the total applied, respectively. This confirms reports of limited movement of oxyfluorfen from foliar applications (9).

Between 1 and 8 days after application of glyphosate to 30-day-old plants, absorption increased from 18 to 35% of the amount applied and total translocation into plants parts increased from 7 to 27% of the amount applied. For 60 day old plants total translocation increased from 17% to 31%, respectively. Zandstra and Nishimoto (24) reported increased glyphosate translocation in purple nutsedge up to 4 days after application, ranging from 5% of the amount applied at 1 day to 19% at 4 days after application. For oxyfluorfen applied alone, total absorption and translocation was about 17% and 2%, respectively.

Glyphosate translocation to the tubers accounted for 22% out of the 37% total translocated to underground plant parts (Table 5.1). This indicates that tubers are a relative strong sink for glyphosate accumulation, although Chase and Appleby (6) reported that glyphosate translocation to underground purple nutsedge parts was only about 3%. These results suggest that glyphosate translocation seems to be greater in yellow nutsedge than reported for purple nutsedge (6, 24).

Since the total amount of herbicide translocated was greatest at 8 days after application, plants from this period were used for translocation comparisons. Dpm/mg was used as a measure of the relative concentration of ^{14}C in plant parts, while total activity (dpm) in the plant parts was used as an indicator of the general direction of translocation with increasing yellow nutsedge age.

Total activity of glyphosate translocated to various parts of nutsedge plants was slightly greater when the herbicide was applied on older plants, but the small amount of oxyfluorfen translocated was not affected by plant age. This slight increase (4.4%) of glyphosate translocation was generally a result of significantly increased (66%) activity found in the underground portions (roots, rhizomes, and tubers) of older plants (Table 5.1). Total activity of glyphosate was generally higher in the top than in underground parts at both plant ages. This result was contrary to those reported for purple nutsedge (24).

Concentration of glyphosate in whole nutsedge plants generally decreased from 111 dpm/mg to 42 dpm/mg when applied to 30-day- and 60-day-old plants, respectively. Comparatively,

oxyfluorfen values were 8 dpm/mg and 3 dpm/mg, respectively. The concentration of glyphosate in leaves, root and rhizomes, and new tubers was reduced to about one half the level found in the younger plants. These differences in concentration of ^{14}C of glyphosate support previous work (16) indicating that for the same dose of glyphosate applied per plant it would be more phytotoxic at the early nutsedge growth stage than at late stages. Translocation of glyphosate measured in yellow nutsedge tubers was influenced by maturity of the individual tubers. Immature tubers accumulated over three times as much radioactive glyphosate per dry weight basis as mature ones. Similarly, concentration in young roots and rhizomes was about two-fold that found in comparable mature tissues (Table 5.2). The relative amount of meristematic tissue in underground plant parts decreases as tubers develop and become mature, which could explain at least in part the decreasing concentration of glyphosate in older plants. These results are similar to those from Thullen and Keeley's (21) studies on the influence of yellow nutsedge tuber maturity in the accumulation of ^{14}C from urea and NAA (naphthaleneacetic acid). They concluded that sources of ^{14}C accumulated in tubers in decreasing amount as tubers became more mature and that accumulation in parent and mature tubers was insignificant.

Total activity in tubers increased with a concurrent slight decrease in activity in leaves as plant age increased. While specific activity in both tubers and leaves decreased with age, total activity in tubers increased with age. This would suggest

that as more weight is formed in roots, rhizomes and tubers a larger proportion of the glyphosate accumulates in the underground portions of the plant. Translocation to the tubers accounted for 22% of the 37% translocated to underground parts (roots, rhizomes, and tubers), indicating that tubers are relatively strong sinks for glyphosate accumulation. Studies on several species have indicated that glyphosate accumulates in the meristematic regions of foliage, roots, rhizomes, and tubers in a typical source-to-sink pattern (6, 7, 17, 22, 24).

The measured radioactivity of glyphosate was significantly increased by the addition of unlabeled oxyfluorfen in tank mixture applications of glyphosate on 30-day-old yellow nutsedge plants (Figure 5.2 and 5.3). Between 1 and 8 days after application of glyphosate absorption was increased from 27% to 46% of the amount applied and total translocation into other plant parts increased from 15% to 42% of the amount applied. Total translocation of glyphosate at 8 days increased from 27% to 42 of the amount applied when comparing single and combination applications. Conversely, the rate of glyphosate absorption and translocation was not increased up to 4 days after treatment on 60-day-old plants. From 4 to 8 days the rate was apparently increased, but addition of oxyfluorfen decreased glyphosate activity relative to its single application to the 60-day-old plants (Figures 5.2 and 5.3). The ability of old nutsedge plants to produce new growth after application of herbicides was insignificant as compared to young plants (Table 5.4). In the absence of new growth and leaf expansion on old plants, oxyfluorfen induced lipid peroxidation

probably lead to increased membrane permeability and leaking of glyphosate into intercellular spaces. The low tolerance and the greater ability of young nutsedge to grow (Table 5.4) with probably more leaf expansion might have contributed to the greater translocation of glyphosate. Crafts and Foy (8) and Wittwer and Teubner (23) reported that newly developed leaves are physiologically and anatomically younger and would be expected to more readily absorb foliar applied chemicals. The greater effectiveness of glyphosate when applied with oxyfluorfen on yellow nutsedge at the early growth stage is extremely important since the tuberization process must be stopped early in the development of the plant if control of the weed is to be successful. Greater concentration of glyphosate in young nutsedge plants (Table 5.2) treated with combinations of glyphosate and oxyfluorfen corroborate previous results showing that this combination of herbicides reduced synergistically production of new tubers and other plant parts (16). Concentration of ^{14}C-oxyfluorfen was not significantly increased by the combination of the two herbicides, suggesting that the greater reduction in tuber production might be a result of increased amounts of glyphosate reaching the sites of tuber formation. Although the concentration of ^{14}C-glyphosate when used in combination with oxyfluorfen was significantly increased in leaves and new tubers of 30-day-old yellow nutsedge, concentration in roots and rhizomes was unchanged. These increases in leaves and new tubers were not observed in applications of these two herbicides to 60-day-old

plants, however concentration in immature tubers was about 6 times as much as in mature ones.

Regrowth and chlorophyll content of glyphosate and oxyfluorfen treated nutsedge plant. - Both glyphosate and oxyfluorfen significantly reduced the ability of nutsedge plants to regrow when plants were cut off 1 cm above the soil surface 6 days after treatment (experiment 2). Most of the regrowth was from secondary shoots but dry matter accumulation in roots and rhizomes was similarly reduced. Number and fresh weight of new tubers were only significantly affected by the glyphosate treatment (Table 5.3). These results indicate that oxyfluorfen did not affect dry matter accumulation in tubers, although it reduced the amount of regrowth of shoots. Previous results (16) reporting on the effect of herbicides on bulking of tubers indicated that oxyfluorfen neither inhibited tuber formation nor affected enlargement of tubers.

In experiment 3, the interval between herbicide application and cutting did not significantly affect the influence of glyphosate and oxyfluorfen on nutsedge regrowth response. Regrowth ability was significantly reduced with increasing plant age. Regrowth abililty was also significantly reduced by glyphosate and oxyfluorfen applied to 40-day- and 70-day-old plants. Table 5.4 shows that combinations of these two herbicides was more phytotoxic to nutsedge than either herbicide applied alone. Also glyphosate reduced regrowth more than oxyfluorfen applications. These results suggest that combining the herbicides resulted in their increased translocation into the bellow ground portion of

the plants prior to the time of cutting. The relative amount of reduction of regrowth by herbicides was greater in older plants, however previous work (16) indicates that applications of herbicides to older plants is ineffective in providing long-term control since many mature tubers have been produced by 70 days after emergence and the reproductive cycle is not broken. Pereira and Crabtree (16) reported that the stage of tuberization at the time of herbicide treatment is extremely important because mature tubers are relatively inactive metabolically (21) and are generally unaffected by herbicides (16, 24). In experiments 2 and 3 glyphosate and oxyfluorfen significantly reduced chlorophyll content of nutsedge foliage when measured 6 days after treatment (Table 5.5). The reduction was greater with oxyfluorfen than with glyphosate. Although the chlorophyll content was reduced by both herbicides at 6 days after treatment, in oxyfluorfen treated plants the effect was not measured 15 days after application. This indicates that the action of this herbicide on nutsedge is temporary, allowing for healthy nutsedge regrowth. Reapplications of oxyfluorfen are required at relatively short intervals in order to achieve an effective sustained level of nutsedge control. At 6 days after application glyphosate significantly reduced chlorophyll content only at the higher rate (1.5 kg ai/ha), although the lower rate (0.75 kg ai/ha) was enough to exert phytotoxic action on nutsedge chlorophyll when measured 15 days after herbicide application. Chlorotic regrowth was qualitatively observed following sublethal dose applications of glyphosate. Kitchen et al. (12) reported that a reduction in chlorophyll

accumulation by glyphosate treated plants may be a result of chlorophyll synthesis inhibition or from increased chlorophyll degradation. Glyphosate has reduced chlorophyll content of purple nutsedge (1), quackgrass (5), corn, barley, and soybean hypocotyls (12).

Figure 5.1 Temperature (^{o}C) at outdoor experiment location, Corvallis, OR. Values represent means of 5-day intervals.

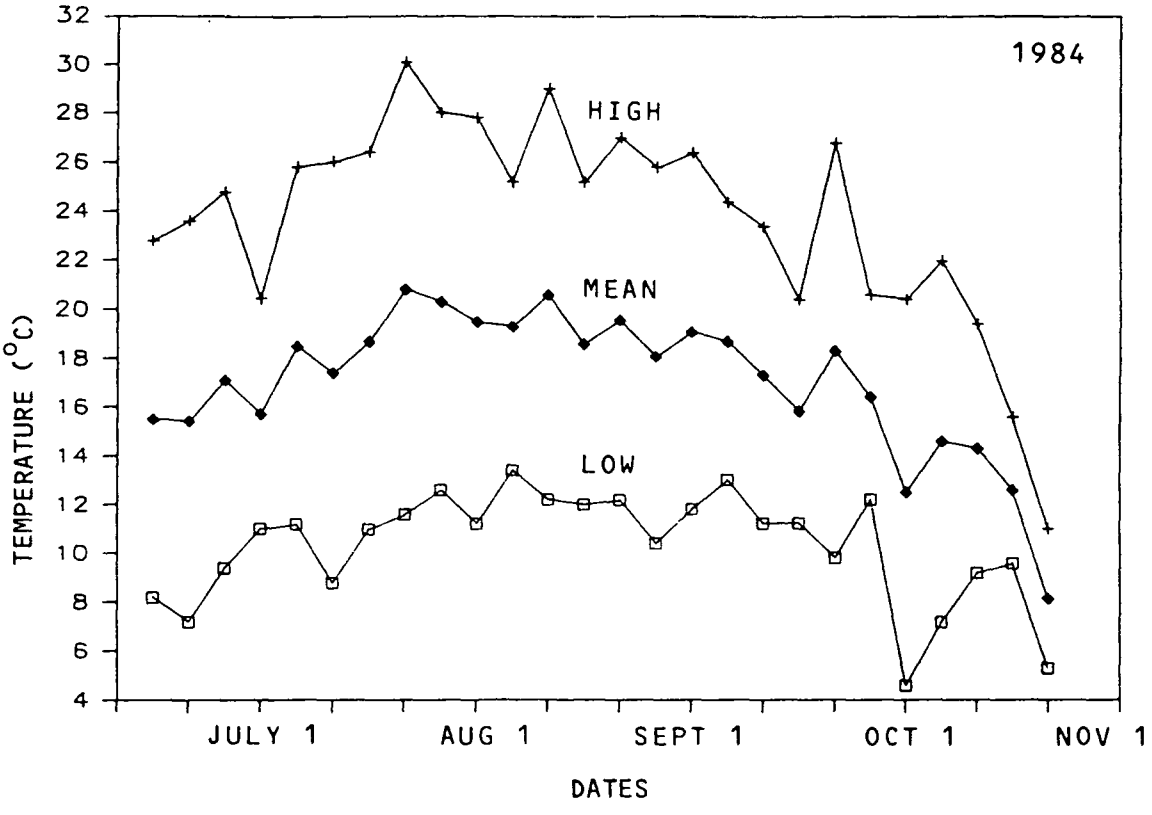

1984
HIGH
MEAN
LOW
TEMPERATURE (°C)
32
30
28
26
24
22
20
18
16
14
12
10
8
6
4
JULY 1
AUG 1
SEPT 1
OCT 1
NOV 1
DATES

Figure 5.1

Figure 5.2 Foliar absorption of ^{14}C-glyphosate and ^{14}C-oxyfluorfen combinations by applications on 30-day- and 60-day-old nutsedge plants, (A= unlabeled glyphosate with ^{14}C-glyphosate, B= unlabeled oxyfluorfen with ^{14}C-oxyfluorfen, C= unlabeled glyphosate and oxyfluorfen with ^{14}C-glyphosate, D= unlabeled glyphosate and oxyfluorfen with ^{14}C-oxyfluorfen).

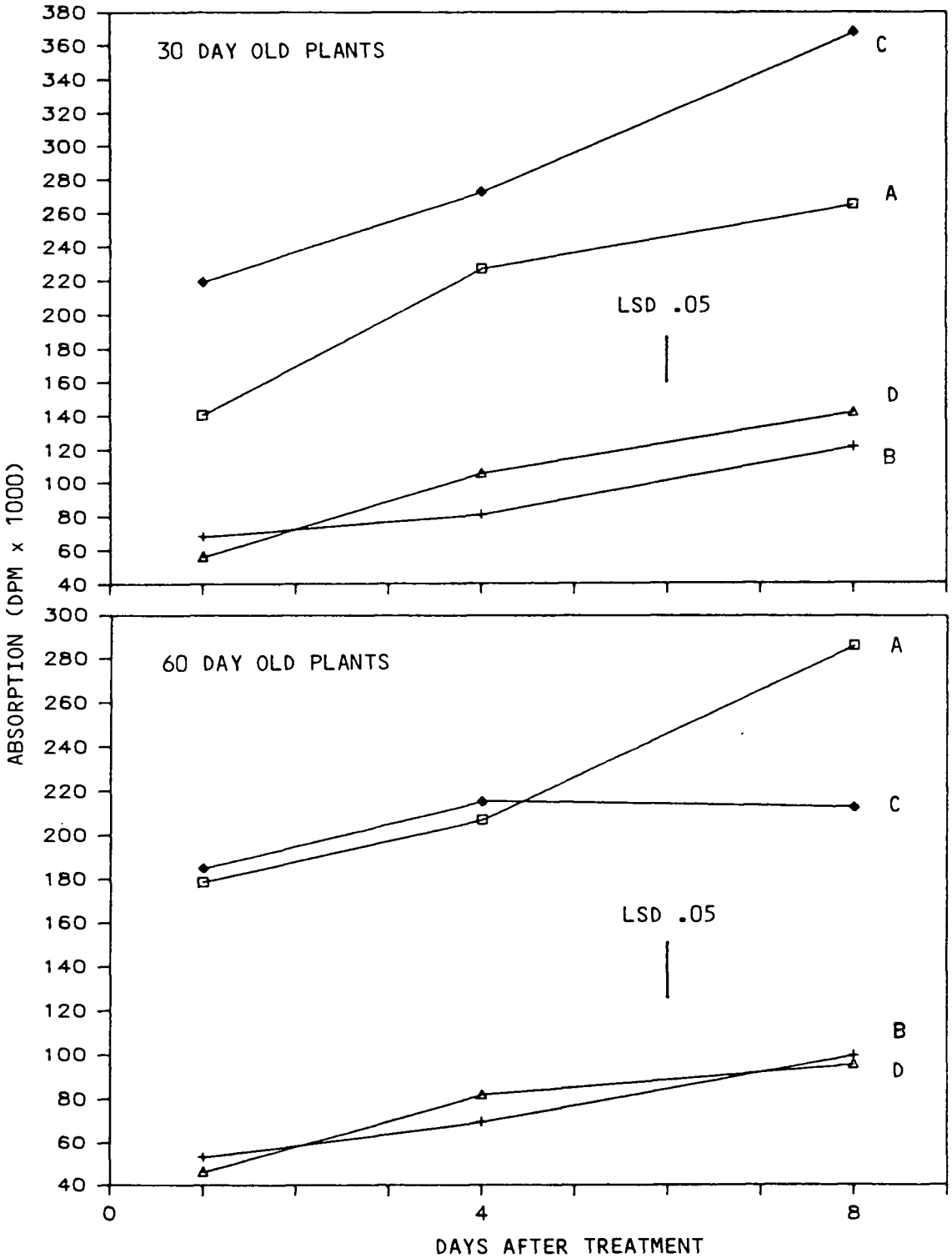
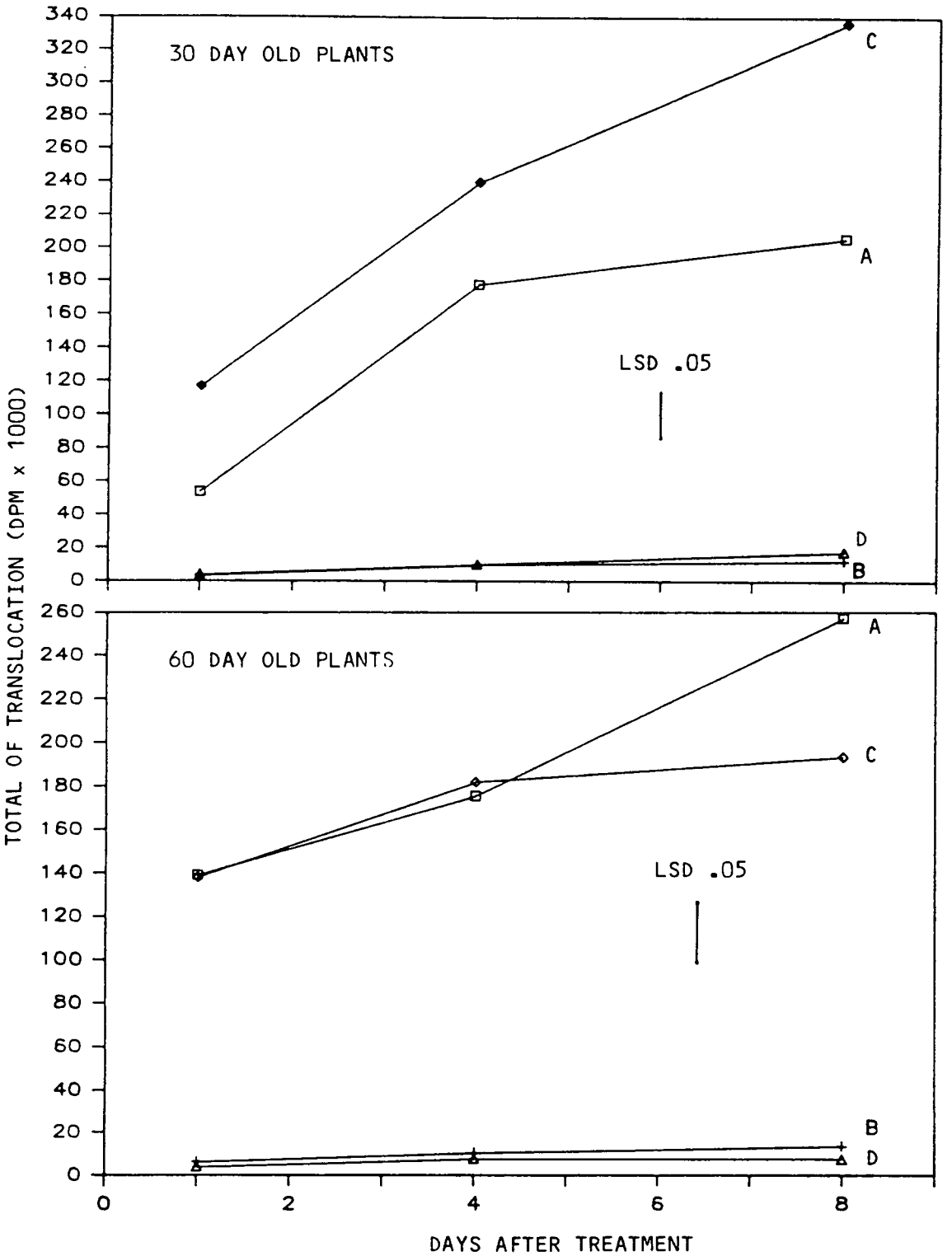

30 DAY OLD PLANTS
C
A
LSD .05
D
B
60 DAY OLD PLANTS
A
C
LSD .05
B
D
ABSORPTION (DPM x 1000)
380
360
340
320
300
280
260
240
220
200
180
160
140
120
100
80
60
40
300
280
260
240
220
200
180
160
140
120
100
80
60
40
0
4
8
DAYS AFTER TREATMENT

Figure 5.2

Figure 5.3 Translocation of ^{14}C-glyphosate and ^{14}C-oxyfluorfen combinations by applications on 30-day- and 60-day-old nutsedge plants, (A= unlabeled glyphosate with ^{14}C-glyphosate, B= unlabeled oxyfluorfen with ^{14}C-oxyfluorfen, C= unlabeled glyphosate and oxyfluorfen with ^{14}C-glyphosate, D= unlabeled glyphosate and oxyfluorfen with ^{14}C-oxyfluorfen).


30 DAY OLD PLANTS
60 DAY OLD PLANTS
LSD .05
LSD .05
A
B
C
D
A
B
C
D
TOTAL OF TRANSLOCATION (DPM x 1000)
DAYS AFTER TREATMENT
0
2
4
6
8
20
40
60
80
100
120
140
160
180
200
220
240
260
280
300
320
340

Figrue 5.3

Table 5.1 The effect of glyphosate and oxyfluorfen combinations and nutsedge growth stage on total translocation activity of ^{14}C-glyphosate and ^{14}C-oxyfluorfen recovered from various plant parts harvested 8 days after herbicide applications.

Days after emergence	Herbicide applied[a]	Whole plant	Inflorescence	Leaves	Roots & rhizomes	Tubers		
						Parent	Immature	Mature
		Total activity (dpm x 1000)						
30	A	206	-	140	52	5.3	5.8	3.0
	B	13	-	10	2	0.3	0.1	0.2
	C	336	-	260	59	8.5	6.1	5.4
	D	18	-	15	2	0.1	0.1	0.0
60	A	257	22	125	88	2.5	11.3	9.0
	B	13	1	8	4	0	0.1	0.1
	C	193	13	93	53	2.4	20.7	10.8
	D	7	1	4	2	0	0.1	0.2
LSD 0.05		35	-	40	28	5.4	2.6	2.6

[a] A= unlabeled glyphosate with ^{14}C-glyphosate
B= unlabeled oxyfluorfen with ^{14}C-oxyfluofen
C= unlabeled glyphosate and oxyfluorfen with ^{14}C-glyphosate
D= unlabeled glyphosate and oxyfluorfen with ^{14}C-oxyfluorfen

Table 5.2 The effect of glyphosate and oxyfluorfen combinations and nutsedge growth stage on concentration of ^{14}C-glyphosate and ^{14}C- oxyfluorfen recovered from various plant parts harvested 8 days after herbicide applications.

Days after emergence	Herbicide[a]	Whole plant	Leaves	Roots and rhizomes	Tubers			
					Parent	New[b]	Immature	Mature
		dpm/mg						
30	A	110.6	130.9	105.4	53.8	77.1	75.3	73.0
	B	7.7	11.3	4.7	4.2	2.3	6.2	0.9
	C	187.0	273.1	101.2	96.1	108.4	202.3	82.6
	D	11.2	18.5	4.9	0.9	1.1	1.7	0.5
60	A	41.6	75.0	54.0	46.9	35.7	62.6	22.4
	B	2.5	6.9	2.8	0.5	0.3	0.9	0.2
	C	30.1	42.0	26.8	35.4	36.1	100.2	17.0
	D	1.2	3.0	1.2	0.2	0.4	0.4	0.5
LSD 0.05		21.9	36.5	28.2	41.6	26.5	76.7	27.9

[a] A= unlabeled glyphosate with ^{14}C-glyphosate
B= unlabeled oxyfluorfen with ^{14}C-oxyfluorfen
C= unlabeled glyphosate and oxyfluorfen with ^{14}C-glyphosate
D= unlabeled glyphosate and oxyfluorfen with ^{14}C-oxyfluorfen

[b] New tubers = immature + mature.

Table 5.3 Influence of glyphosate and oxyfluorfen on regrowth ability of 40 day old yellow nutsedge after cutting 6 days after herbicide applications. Greenhouse experiment 2.[a]

Herbicide rate		Total shoot regrowth	Root & rhizome dry weight	Tuber fresh weight	Tuber number
Glyphosate	Oxyfluorfen				
kg/ha		g/plant			#/plant
0	0	9.0 a	2.3 a	2.7	25
0	1.5	3.5 b	1.6 b	2.8	21
Mean				2.7 a	23 a
1.5	0	0 c	1.0 c	1.3	11
1.5	1.5	0 c	0.8 c	1.4	12
Mean				1.3 b	11 b

[a]Values within columns following by the same letter are not significantly differents at the 0.05 level according to Tukey's test.

Tabel 5.4 Influence of glyphosate and oxyfluorfen on regrowth ability of yellow nutsedge at 40 days and 70 days after emergence. Outdoor experiment 3.[a]

Days after emergence	Glyphosate rate (kg ai/ha)	Oxyfluorfen rate (kg ai/ha)		
		0	0.75	1.5
		- Total fresh weight regrowth (g/plant) -		
40	0	20.1 A	15.4 B	13.2 BC
	0.75	11.3 C	4.6 D	2.9 DE
	1.50	0.2 E	0.0 E	0.0 E
70	0	5.6 a	4.8 a	3.5 ab
	0.75	0.4 b	0.5 b	0.1 b
	1.50	0.1 b	0.0 a	0.0 b

[a]Values within data groups representing each 'Days after emergence' followed by the same letter are not significantly different at the 0.05 level by using Tukey's test. Data from 6 and 15 days after herbicide applications were pooled for an appropriated statistical analysis.

Table 5.5 Influence of glyphosate and oxyfluorfen on chlorophyll content ($\mu g/cm^2$) in 40-day-old yellow nutsedge, measured at 6 and 15 days after herbicide applications.[a]

Days After Treatment	Glyphosate Rate kg ai/ha	Oxyfluorfen rate kg ai/ha 0.0	0.75	1.50	Mean
		Greenhouse conditions			
6	0	37.9	-	16.6	27.2 A
	1.5	26.5	-	11.5	19.0 B
	Mean	32.2 a	-	14.0 b	
		Outdoor conditions			
6	0	31.3	22.3	21.3	25.0 A
	0.75	31.0	22.8	19.3	24.3 A
	1.50	24.2	18.2	14.2	18.8 B
	Mean	28.8 a	21.1 b	18.2 b	
15	0	31.8	34.1	34.6	33.5 A
	0.75	21.3	20.6	20.4	20.5 B
	1.50	21.4	20.4	18.6	20.1 B
	Mean	24.8	25.0	24.5	

[a]Values within column or row followed by the same type of letter are not significantly different at the 0.05 level according to Tukey's test. Data are means of 6 and 4 replications from experiments 2 and 3 in greenhouse and under outdoor conditions, respectively.

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Chapter 6

EVALUATION OF DICHLOBENIL, METOLACHLOR, AND NORFLURAZON FOR CONTROLLING TUBERIZATION OF YELLOW NUTSEDGE (*Cyperus esculentus* L.)

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Abstract. Three soil-applied herbicides were evaluated in a pear orchard, in a greenhouse, and in outdoor pots to determine their effects on long-term control of yellow nutsedge (*Cyperus esculentus L.* var *leptostachyus* Boeck. #[a] CYPES) tuberization and tuber survival. Both dichlobenil (2,6-dichlorobenzonitrile) and metolachlor [2-chloro-*N*-(2-ethyl-6-methylphenyl)-*N*-2-methoxy-1-methylethyl)acetamide] consistently reduced shoot and tuber populations either in a greenhouse or under field conditions. In the field, consecutive applications for 2 years provided the best control of nutsedge and reduced tuber populations as much as 88% for dichlobenil and 90% for metolachlor. In the greenhouse, norflurazon [4-chloro-5-(methylalamino)-2-(3-(trifluoromethyl)phenyl)-3(2H)-pyridazinone] provided the best control (100%). There was a significant reduction in yellow nutsedge tuber populations from residual effects of the herbicides in the tubers produced from treated plots. Winter surface applications and spring surface applications of metolachlor reduced the percent of tuber sprouting with a concurrent increase in percent of decayed tubers.

Additional index words - CYPES, herbicides, soil-applied, long-

term control, tuber sprouting, dry matter accumulation, pear orchard.

[a]Letters following this symbol are a WSSA-approved computer code from Important Weeds of the World, 3rd. ed., 1983. Available from WSSA, 309 West Clark St. Champaign, IL 61820.

Introduction

Most soil-applied herbicides used to selectively control yellow nutsedge inhibit emergence of new shoots or rhizomes, but have little effect on parent tubers (13). The major fault with control programs using those herbicides is that they allow escapes and do not provide consistent control for a long period (4, 12). However, some herbicides have longer residual activity in soil. The chloroacetamide herbicides such as alachlor [2-chloro-N-(2,6-diethylphenyl)N-methoxymethyl)acetamide and metolachlor have proven to be relatively effective in controlling nutsedge at low rates of application (2, 5, 12). Also, both field and greenhouse studies with dichlobenil have indicated that this herbicide can provide good control of purple and yellow nutsedge (3, 8, 9, 11). Preemergence applications of norflurazon under greenhouse conditions have provided a high level of yellow nutsedge control but this success has not been carried to trials in the field (6, 11).

Pereira et al. (7) have reviewed the action of different herbicide families on purple and yellow nutsedges and suggested it would be appropriate that research efforts for the control of nutsedge be directed toward inhibiting sprouting of tubers and inhibiting new tuber formation.

The objectives of these studies were (a)- to compare the influence of dichlobenil, metolachlor, and norflurazon on yellow nutsedge tuberization, (b)- to evaluate long-term control of yellow nutsedge with herbicides in a pear orchard, (c)- to

determine the effect of rate and split applications of metolachlor on tuberization of yellow nutsedge, and (d)- to determine if herbicides affect new tuber sprouting as well as growth characteristics of ramets from tubers produced by treated plants.

Materials and Methods

Greenhouse and outdoor studies - Yellow nutsedge tubers were collected from a field near Dayton, OR in the spring of 1982, multiplied in the greenhouse and stratified at 4.5 ^{o}C for subsequent studies.

Tubers were submerged for 24 hours in a 75 ppm solution of gibberellic acid (GA_3) to break dormancy and then planted in a greenhouse sandy loam soil mix with 1% (experiment 1) and 4% (experiment 2 and 3) organic matter, in trays of 0.1 m^2 by 0.12 m. The medium was previously sifted in order to facilitate washing all particles from plants at harvest time, for the purpose of assessing herbicidal effect on tuber number and weight. Top water was applied as needed and included a weekly application of water soluble 20-20-20 fertilizer.

In experiments 1, 2, and 5 plants were grown in a greenhouse with supplemental lighting (photosynthetic photon flux density about 200 $\mu E.m^{-2}.s^{-1}$) in the morning and evening to provide a 15 hour photoperiod for 60 days. Temperatures were maintained at about 24 ^{o}C during the day and 18 ^{o}C at night. For experiment 1, eighteen tubers were planted per tray, kept in the greenhouse from February to early June 1983, and then moved outdoors. Treatments were placed in a complete ramdomized design with 4 replications. Dichlobenil, metolachlor, and norflurazon were applied preplant incorporated or preemergence (Table 6.1) with the objective of examining the long-term influence of herbicides on nutsedge plants and tuber production. The treatments from this trial were repeated

in a 2-year field study (experiment 4) in a pear orchard near Hood River, OR.

Experiments 2 and 3 were established to determine the effect of rates and split applications of metolachlor (Table 6.3 and Table 6.4) on yellow nutsedge tuberization. In experiment 2, 9 tubers per tray were planted and ramets grown in the greenhouse from mid-April to early July and then moved outdoors during the summer of 1983. Experiment 3 was conducted entirely outdoors during the summer of 1983 (Figure 6.1) with six tubers per 10-L pot planted in a greenhouse soil mix.

A track-mounted, greenhouse sprayer was calibrated for 340 L/ha and used to apply the herbicides. At the end of each experiment soil was washed from the plants and they were separated into parts to determine (a)- number of shoots, immature (young, white) tubers, and mature (large, brown) tubers; (b)- fresh and/or dry weight of shoots, rhizomes and roots, and tubers; and (c)- visual ratings of nutsedge foliage injury with 0 = no effect and 100 = complete kill.

Field study - In a 2-year study (experiment 4), herbicides were applied on a natural stand of yellow nutsedge in a 3-year old pear orchard near Hood River, OR with the primary objective of examining the long-term control of yellow nutsedge tuberization and tuber survival. The soil was a sandy loam with 1.9% organic matter. Plots 1.8 by 1.8 m were established in a randomized block design with 4 replications. Herbicides were applied in mid-January and March, representing winter and spring applications,

respectively (Table 6.2). Yellow nutsedge foliage injury was evaluated using a visual system (0 = no effect and 100 = complete kill), and in the fall two soil samples were dug from each plot to assess herbicidal effect on tuber number and weight. Tubers were stratified for later tuber survival studies.

Nutsedge tuber sprouting and dry matter accumulation of ramets originating from tubers produced by treated plants - Tubers collected from treated plants under field conditions (experiment 4) were placed in Petri dishes lined with filter paper moistened with a 0.03% captan [cis-N-((trichloromethyl)thio)-4-cyclohexene-1-2-dicarboximide] suspension, sealed in plastic bags and stratified at 4.5 ^{o}C for at least 4 weeks. Later, in experiment 5, tuber sprouting and new plant growth characteristics were evaluated. A subsample of 100 tubers from each treatment in 1983 was put in a germinator (8 hour 30 ^{o}C day, 16 hour 20 ^{o}C night) and sprouted tubers were counted every week for 4 weeks. The few tubers that did not sprout at 4 weeks were cut and visibly classified either as dormant if they were firm and without apparent decay, or non-viable if were soft and decayed. From those sprouted tubers a random sample of 7 ramets from each treatment was planted in Jiffy Mix Plus in a 3-L pot and placed in a greenhouse under conditions described above. After 40 days shoots were counted, and the medium was washed from the plants which were then separated into top and underground parts, dried and weighed.

Results and Discussion

Nutsedge tuber production was significantly reduced by dichlobenil, metolachlor, and norflurazon, compared to untreated plants when tested in greenhouse and field experiments (Tables 6.1 and 6.2). Both dichlobenil and metolachlor provided consistent results either in the greenhouse or under field conditions while norflurazon provided superior results in the greenhouse (Table 6.1) but inferior control in the field (Table 6.2).This difference in norflurazon activity under field and greenhouse conditions was also observed by Banks (1) who suggested that the relatively low water solubility (28 ppm) of norflurazon and the fact that it was applied preemergence may have prevented its movement to deeper sprouting yellow nutsedge tubers under field conditions. In experiment 1 (Table 6.1) tubers from norflurazon treatments sprouted and emerged at the same time as did untreated ones, but all shoots were dead by the end of the experiment. In the field (Table 6.2) nutsedge injury was determined to be about 25% in norflurazon plots at the end of the season. In experiment 1 dichlobenil and metolachlor applications inhibited shoot emergence for about 95 and 110 days, respectively. After 230 days the number of shoots in metolachlor treated trays was slightly greater than the number of shoots from untreated plants, but the height of plants was less than half and tuber production was reduced by as much as 63% of the untreated plants.

Incorporation of dichlobenil under field or greenhouse conditions significantly improved nutsedge control as measured by

shoot dry weight and tuber production (Table 6.1 and 6.2). Either winter surface or spring incorporated applications afforded better control of nutsedge tuberization than spring surface applications (Table 6.1). This enhanced herbicidal effectiveness was probably related to less evaporation loss of dichlobenil when it was incorporated, either mechanically or by winter rainfall. In the field (Table 6.2) a single winter surface application of dichlobenil or spring surface application of metolachlor reduced tuber populations about 62% and 68%, respectively. However, consecutive applications for 2 years provided the best control of yellow nutsedge and reduced tuber populations as much 88% and 90% as compared to the untreated plants, respectively. This indicates that reapplications of these herbicides are needed to improve control of yellow nutsedge tuberization.

Plots treated only in 1983 were kept without further treatment and evaluated at the end of the second growing season at which time it was observed that there was still significant reduction on the tuber population. This shows a residual effect of the herbicides in the yellow nutsedge tuber population and that a measure of control can be expected for more than one season.

Tuber production in 1984 was generally less than in 1983 and caused by an significant interaction between treatments and years. In 1984 there was some competition from other weeds in the experiment which could explain in part the differences in tuber production observed between years.

Rates of metolachlor of 1 kg ai/ha or greater significantly reduced yellow nutsedge tuber production in experiments 2 and 3

(Tables 6.3 and 6.4). In experiment 3, using a soil with 4% organic matter, there was no significant effect on nutsedge by increasing the rate above 1 kg ai/ha (Table 6.4) but the number of tubers was significantly reduced with higher rates applied to a sandy loam soil with 1% organic matter in experiment 2 (Table 6.3). Metolachlor rates of 1 kg ai/ha applied at one time allowed for shoot survival and relatively high tuber production (Table 6.3). In this experiment, splitting the application of metolachlor did not improve nutsedge control, although in experiment 3 (Table 6.4) there was a slight reduction in tuber production.

When tubers collected from plants in experiment 4 were evaluated, winter surface applications of dichobenil and spring surface applications of metolachlor significantly reduced the percent of tubers sprouting, with a concurrent increase in percent of decayed tubers. None of the herbicide treatments reduced shoot or root and rhizome dry matter accumulation by 40-day-old ramets growing from tubers collected from the plots of experiment 4 (Table 6.5).

Figure 6.1 Temperature (^{o}C) at outdoor experiment location, Corvallis, OR. Values represent means of 5-day intervals.

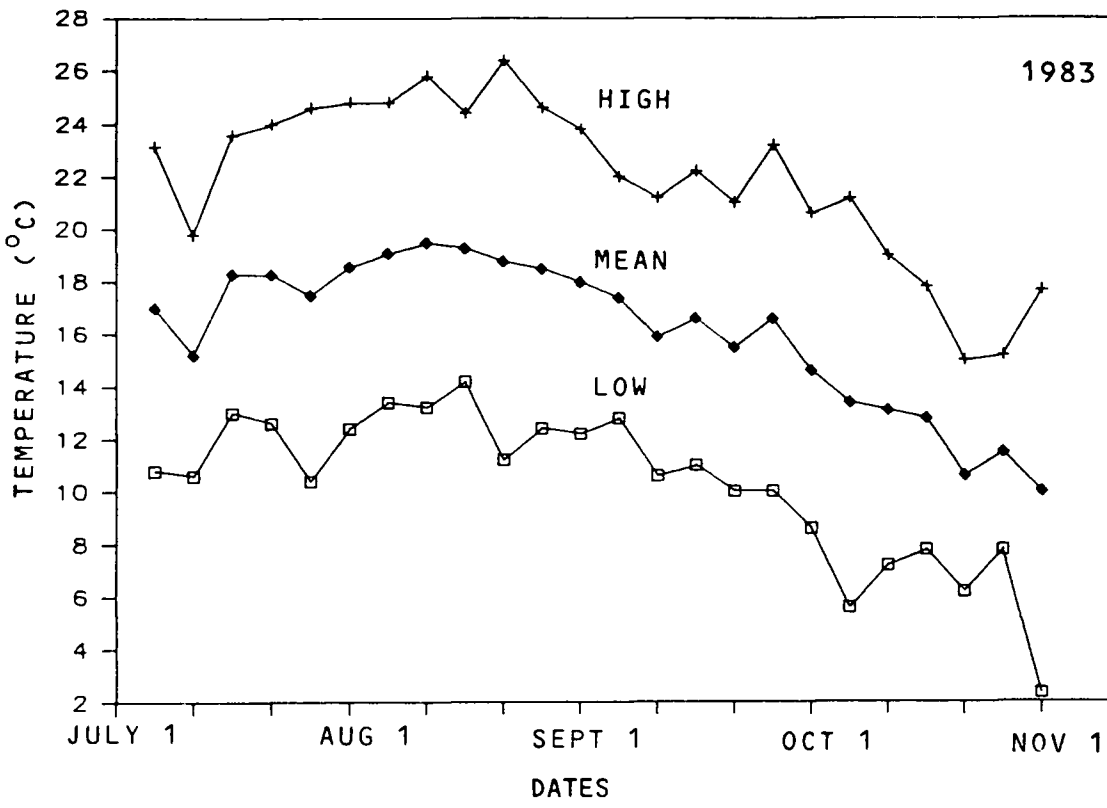
28
26
24
22
20
18
16
14
12
10
8
6
4
2
TEMPERATURE (°C)
1983
HIGH
MEAN
LOW
JULY 1
AUG 1
SEPT 1
OCT 1
NOV 1
DATES

Figure 6.1

Table 6.1 Long-term yellow nutsedge response to dichlobenil (6 kg ai/ha), metolachlor (4 kg ai/ha), and norflurazon (4 kg ai/ha), greenhouse experiment 1.[a]

Treatment	Time of application[b]	Live shoots at 140 days	Live shoots at 230 days	Shoot dry weight	Tuber fresh weight
		#/tray	#/tray	g/tray	g/tray
Dichlobenil	PRE	7 c	22 b	19.9 b	67.3 b
	PPI	0 d	8 c	5.9 c	21.4 cd
Metolachlor	PRE	22 ab	66 a	22.0 b	76.6 b
	PPI	16 bc	55 a	15.3 b	55.7 bc
Norflurazon	PRE	0 d	1 d	0.1 c	0.1 d
	PPI	0 d	0 d	0.0 c	0.0 d
Untreated	-	31 a	49 a	56.0 a	206.6 a

[a]Values within columns followed by the same letter are not significantly different at the 0.05 level according to Tukey's test.

[b]PRE and PPI = preemergence and preplant incorporated applications, respectively.

Table 6.2 Long-term response of yellow nutsedge to single or repeated applications of dichlobenil, metolachlor, and norflurazon. Field experiment 4.

Treatment	Time of application[a]	Rate kg ai/ha	1983 Foliage injury	1983 Tuber number	1983 Tuber weight	1984 Foliage injury	1984 Tuber number	1984 Tuber weight
			-- % --	#/plot	g/plot	-- % --	#/plot	g/plot
Untreated	-	-	0	537	75	0	478	52
Dichlobenil	PRE W 1983	6	90	162	29	43	198	22
	PRE S 1983	6	22	442	65	12	415	41
	PPI S 1983	6	73	277	56	18	285	32
Metolachlor	PRE S 1983	4	89	129	13	56	138	15
Norflurazon	PRE S 1983	4	21	448	47	23	477	32
Dichlobenil	PRE W 83&84	6+6	83	248	38	93	57	11
	PRE S 83&84	6+6	19	461	67	33	215	34
	PPI S 83&84	6+6	66	315	53	98	106	20
Metolachlor	PRE S 83&84	4+4	90	165	21	96	48	4
Norflurazon	PRE S 83&84	4+4	12	420	41	43	199	16
Dichlobenil	PRE S 1984	6	--	---	--	56	145	18
	PRE S 1984	6	--	---	--	18	278	34
	PPI S 1984	6	--	---	--	96	109	17
Metolachlor	PRE S 1984	4	--	---	--	58	161	11
Norflurazon	PRE S 1984	4	--	---	--	29	330	25
LSD (5%)			18	82	14	17	59	10

[a]PRE, PPI, W, and S = preemergence, preplant incorporated, winter and spring applications, respectively.

Tabel 6.3 The effect of rates and split application of metolachlor on yellow nutsedge 7 months after herbicide application. Greenhouse experiment 2.[a]

Rate	Number of living shoots	Dry weight		Tuber	
		Shoots	Roots & rhizomes	Fresh weight	Number
kg/ha	#/tray	g/tray			#/tray
1	27 ab	10.6 b	3.5 b	48.3 b	620 b
2	7 cd	1.8 cd	0.7 c	11.1 c	121 d
3	5 cd	1.2 cd	0.2 c	5.6 c	66 e
4	0 d	0.0 d	0.0 c	0.0 c	0 f
1/1[b]	25 ab	4.8 c	0.6 c	18.3 c	348 bc
2/2	1 d	0.4 d	0.1 c	2.4 c	20 ef
1/1/1	13 bc	4.0 cd	0.7 c	15.6 c	239 cd
2/2/2	1 d	0.1 d	0.0 c	0.2 c	2 ef
Untreated	34 a	23.6 a	23.0 a	111.7 a	1220 a

[a]Values within columns followed by the same letter are not significantly different at the 0.05 level by using Tukey's test.

[b]/ = split application at 25 day intervals.

Table 6.4 The effect of rates and split applications of metolachlor on yellow nutsedge at 110 days after herbicide application. Outdoor experiment 3.[a]

Rate	Number of living shoots	Dry weight of		Tuber	
		Shoots	Roots & rhizomes	Fresh weight	Number
kg/ha	#/tray	g/tray			#/tray
0.5	22 a	5.1 b	5.1 b	32.4 b	336 a
1.0	7 bc	1.5 c	1.7 b	14.0 c	125 b
2.0	7 bc	2.1 c	2.3 b	16.1 c	139 b
4.0	5 bc	1.4 c	2.5 b	9.7 cd	96 bc
0.5/0.5[b]	11 b	1.5 c	1.4 b	9.2 cd	95 bc
0.5/0.5/0.5	6 bc	1.7 c	2.1 b	11.2 cd	95 bc
1.0/1.0	6 bc	1.9 c	2.1 b	9.0 cd	100 bc
2.0/2.0	5 bc	1.5 c	1.8 b	7.5 cd	67 bc
4.0/4.0	2 c	1.2 c	1.8 b	7.0 d	57 c
Untreated	25 a	11.1 a	12.5 a	57.6 a	356 a

[a]Values within columns folowed by the same letter are not significantly different at the 0.05 level according to Tukey's test.

[b]/ = split application at 25 day intervals.

Table 6.5 The effect of dichlobenil (6 kg ai/ha), metolachlor (4 kg ai/ha), and norflurazon (4 kg ai/ha) on yellow nutsedge tuber sprouting and dry matter accumulation by 40 day-old ramets originating from tubers produced by plants treated in 1983. Experiment 5.[a]

Treatment	Time of application[b]	Tuber sprouting	Tuber decaying	Dry weight[c] Shoots	Dry weight[c] Roots & rhizomes	Number of new shoots
		%	%	g/pot	g/pot	#/pot
Dichlobenil	PRE W	87 c	9 a	8.8	2.2	16 b
	PRE S	97 ab	2 b	8.0	2.5	17 ab
	PPI S	95 abc	3 b	8.6	2.4	17 ab
Metolachlor	PRE S	88 c	7 ab	8.7	1.8	17 ab
Norflurazon	PRE S	98 a	2 b	8.5	2.1	21 a
Untreated	-	97 ab	2 b	8.8	2.2	17 ab

[a]Values within columns followed by the same letter are not significantly different at the 0.05 level by using Tukey's test.

[b]PRE, PPI, W, and S= preemergence, preplant incorporated, winter, and spring applications, respectively.

[c]Not significant at 5% of probability by F test.

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APPENDICES

Appendix A

Figure 3.8 Number of immature and total (immature + mature) tubers (A) and dry matter accumulation in yellow nutsedge plant parts (B) grown under outdoor conditions. Values are average of 4 replications from experiment 2. Error bars = standard deviation of the mean.

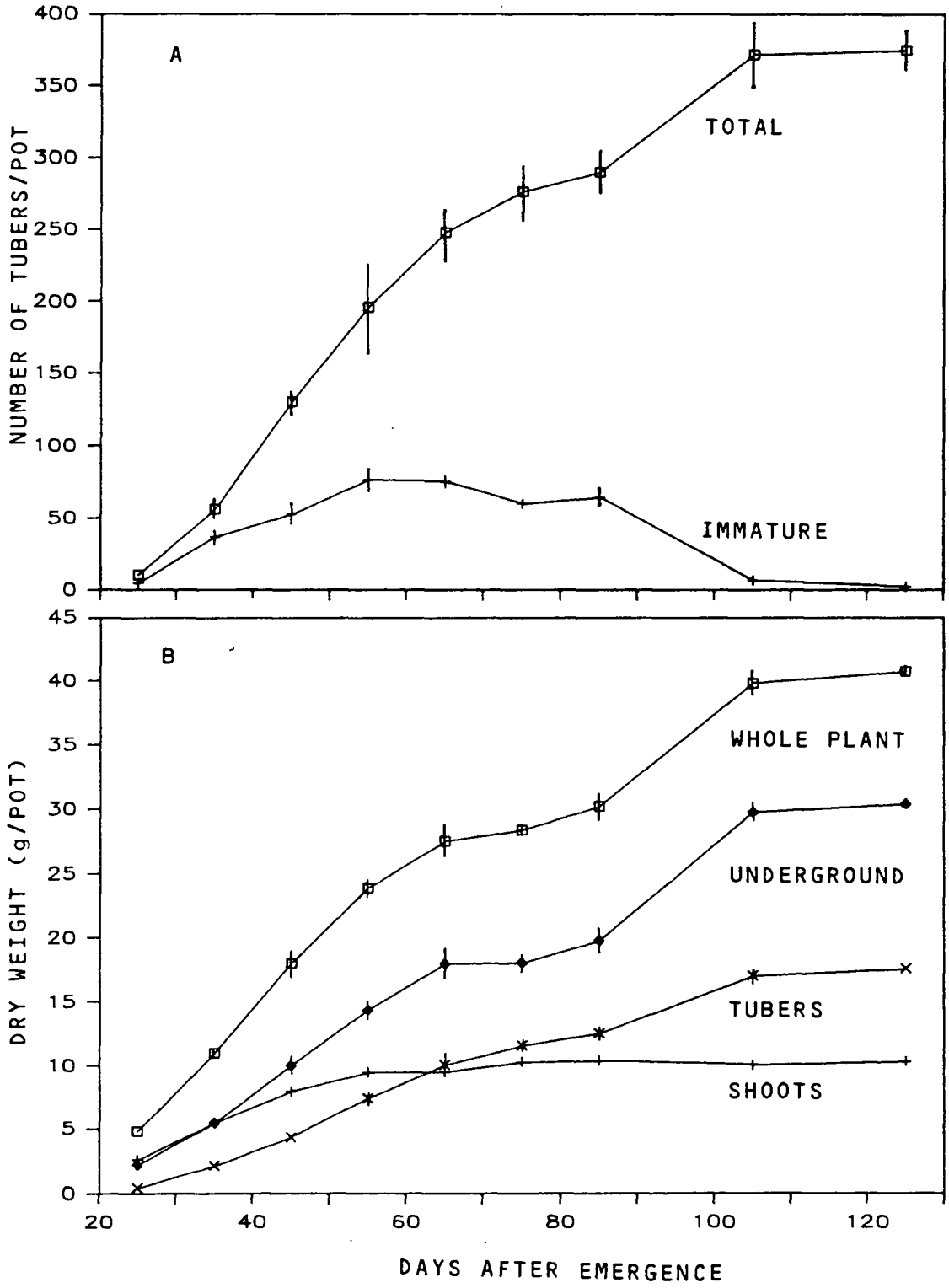
A
TOTAL
IMMATURE
NUMBER OF TUBERS/POT
400
350
300
250
200
150
100
50
0
B
WHOLE PLANT
UNDERGROUND
TUBERS
SHOOTS
DRY WEIGHT (g/POT)
45
40
35
30
25
20
15
10
5
0
20
40
60
80
100
120
DAYS AFTER EMERGENCE

Figure 3.8

Appendix B

Preparation of ^{14}C herbicide solutions:

^{14}C-glyphosate - An aqueous solution was prepared following the recommendations of Monsanto Agricultural Products Co. researchers:

To 5 ml of water add 40 mg of ^{14}C-glyphosate, 14 mg of isopropyl amine, and 20 mg of MON 0818. Dilute with water to make volume to 10 ml.

Formation of the isopropyl amine salt of glyphosate is an exothermic reaction and although for small quantities there should be no problems it was recommended that half of the water be cooled close to freezing while placed in the reaction vessel. The isopropyl amine is added while still maintaining a temperature close to freezing. The glyphosate is added in portions and the reactants allowed to come to room temperature, possibly with the need to heat the reactants. Surfactant is added to the completed isopropyl amine-glyphosate and finally the solution is brought up to volume by adding the rest of the water.

^{14}C-oxyfluorfen - was dissolved in absolute methanol. It was used in this form, without mixing with water or the addition of a surfactant.