FACTORS AFFECTING N-(PHOSPHONOMETHYL) GLYCINE (GLYPHOSATE) ACTIVITY IN TURFGRASS AND ALFALFA [MEDICAGO SATIVA (L.)] SEEDLING ENVIRONMENTS AND DEGRADATION IN THE SOIL

Dissertation for the Degree of Ph. D.

MICHIGAN STATE UNIVERSITY

LOREN JAMES MOSHIER

1977

This is to certify that the

thesis entitled

FACTORS AFFECTING N-(PHOSPHONOMETHYL)GLYCINE (GLYPHOSATE) ACTIVITY IN TURFGRASS AND ALFALFA [MEDICAGO SATIVA (L.)] SEEDLING ENVIRONMENTS AND DEGRADATION IN THE SOIL

presented by

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has been accepted towards fulfillment
of the requirements for

Ph.D. degree in Dept. of Crop and Soil Sciences

Major professor

Date June 3, 1977

O-7639

T-010

ABSTRACT

FACTORS AFFECTING N-(PHOSPHONOMETHYL)GLYCINE (GLYPHOSATE) ACTIVITY IN TURFGRASS AND ALFALFA [*MEDICAGO SATIVA* (L.)] SEEDLING ENVIRONMENTS AND DEGRADATION IN THE SOIL

By

Loren James Moshier

The effects of several concentrations of N-(phosphonomethyl)glycine (glyphosate) on germination and seedling growth of four Kentucky bluegrass (*Poa pratensis* L.), two creeping bentgrass (*Agrostis palustris* Huds.) and two red fescue (*Festuca rubra* L.) cultivars were evaluated in petri dishes under controlled environmental conditions. None of the glyphosate concentrations had a detrimental effect on germination of any of the turfgrasses examined. A high concentration of glyphosate (10^{-4} M) did reduce shoot growth in all but two cultivars. Siduron, a selective preemergence herbicide used in turfgrass establishment, had no effect on either germination or shoot lengths. Glyphosate or siduron applied to the soil and incorporated did not affect turfgrass emergence or growth. When glyphosate was sprayed over the soil with the seed lying on the soil surface, red fescue germination was reduced and shoot growth of all three turfgrass species was inhibited at the high application rate (17.9 kg/ha). Under field conditions, none of the glyphosate treatments reduced turfgrass emergence or growth.

The extent that glyphosate affected alfalfa (Medicago sativa L.) germination, emergence and seedling growth in several systems was also evaluated. Glyphosate and EPTC, a selective preemergence herbicide used in alfalfa establishment, caused no significant reduction in percent germination of either Saranac or Vernal cultivars grown in petri dishes. Glyphosate reduced shoot length of both cultivars. Vernal appeared to be more susceptible to glyphosate than Saranac. Soil applications of glyphosate also had no effect on percent emergence of either cultivar and only reduced plant height when applied to the soil at 17.9 kg/ha. Glyphosate applications to a Kentucky bluegrass sod in a simulated sod seeding alfalfa establishment study in the greenhouse reduced dry weight of both alfalfa cultivars. Application of glyphosate in four management systems showed that a 3-day planting delay of water application after spraying reduced growth inhibition of alfalfa seeded in treated sod.

Factors influencing degradation of ^{14}C-glyphosate to $^{14}CO_2$ was studied in three soils. Glyphosate degradation occurred primarily by microorganisms and at variable rates. Phosphate additions stimulated degradation to a limited extent in the Collamer silty clay loam but not in the Norfolk loamy sand. Additions of Fe^{+++} and Al^{+++} reduced degradation in the Spinks sandy loam. It is postulated that formation of colloidal Fe and Al precipitates in modified soils with concomitant adsorption of glyphosate was responsible for

decreased availability of glyphosate to microorganisms. Mn^{++} additions were found to increase degradation. Spinks soil and organic carbon amendments failed to increase glyphosate degradation in soils with low degradation rates.

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A DISSERTATION

Submitted to
Michigan State University
in partial fulfillment of the requirements
for the degree of

DOCTOR OF PHILOSOPHY

Department of Crop and Soil Sciences

1977

ACKNOWLEDGEMENTS

The author wishes to express his sincere appreciation to Dr. Donald Penner for his guidance and patience at all times during this study and in the preparation of this dissertation.

Gratitude is expressed to Dr. W. F. Meggitt for his cooperation and guidance and to Dr. B. G. Ellis, Dr. A. R. Putnam and Dr. J. M. Tiedje who also served on my guidance committee.

Appreciation is extended to Dr. A. D. Kern and Dr. D. B. Sharp, Monsanto Agricultural Products Co., for their contribution of ideas. The assistance of numerous work-study students in the greenhouse and laboratory aspects of this study, and the assistance of my wife, Deanna, in typing the manuscript is gratefully acknowledged.

Appreciation is also extended to Monsanto Agricultural Products Co. for their financial assistance and for supplying ^{14}C-glyphosate.

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INTRODUCTION

The discovery of the selective phytotoxicity of the phenoxy herbicides (59) ushered in an era of organic herbicide use as a means of controlling weeds. Biodegradability of organic compounds represented a major advantage over use of persistent inorganic compounds which had been used for selective weed control since 1900 (50). The list of chemicals for use in postemergence applications has grown steadily since and today includes both selective and nonselective herbicides (5). Nonselective postemergence herbicides are now used in conventional preplant, minimum tillage, and chemical fallow cropping systems and include both soil and foliar applied chemicals (5). Nonselective foliar applied herbicides used in these systems include dalapon (2,2-dichloropropionic acid), paraquat (1,1-dimethyl-4,4-bipyridinium ion) and glyphosate [N-(phosphonomethyl)glycine]. Phytotoxic residues from these applications must be removed from or inactivated within the seed environment to prevent inhibition of germination and seedling growth of succeeding crops. Dalapon residues remain mobile in the soil environment (90) and therefore planting of susceptible crops should be delayed for 4 weeks after dalapon applications (11, 61, 82). Paraquat is readily inactivated within the soil by adsorption to

soil colloids (2), but plant residues can remain in sufficient amounts to inhibit growth of turfgrass seedlings in contact with the treated foliage (49). Glyphosate is also rapidly inactivated within soils (6, 80) but residues appeared to be present in sufficient quantities in a treated sod environment to affect legume and grass seedling growth in contact with the treated foliage (14). A period of 3 weeks between spraying and seeding appeared necessary to insure complete dissipation of the residues. Studies (31, 71, 79, 90) with ^{14}C-glyphosate applied to various soils showed a wide variation in degradation rates. The possibility of residual characteristics in glyphosate treated sod and soils with low degradation rates is an environmental concern.

The objectives of this study were (1) to determine the magnitude of inhibition on turfgrass and alfalfa seedlings by glyphosate residues in several management systems and (2) to determine why ^{14}C-glyphosate degradation to $^{14}CO_2$ proceeded at a relatively slow rate in some soils and not in others.

CHAPTER 1

LITERATURE REVIEW

DISSIPATION OF GLYPHOSATE IN SOIL

Introduction

Numerous processes influence the activity and fate of herbicides introduced into the soil environment (66, 88). Transfer and degradation processes immediately act on the herbicide and are intimately associated with each other (88). Transfer processes include absorption and exudation by plants and animals, retention in vegetation and removal by harvesting, adsorption by soil particles, movement in the atmosphere as a gas, surface runoff and movement through the soil via capillary flow or in the leachate. Degradation processes include chemical decomposition, photodecomposition and biological decomposition.

Chemical and Photodecomposition

Photodecomposition of glyphosate has been reported to be negligible (71, 90). Other studies (71, 81, 90) have shown that chemical degradation is also a minor decomposition mechanism in soil. Evolution of ^{14}C as $^{14}CO_2$ accounted for less than 1 percent of total ^{14}C applied as glyphosate in a sterilized soil after 28 days (81).

Microbial Decomposition

Degradation by soil microorganisms is an important biological mechanism by which nonselective postemergence herbicides such as glyphosate are removed from the environment. Microbial degradation rates of herbicides are influenced by the amount of herbicide available for degradation. Adsorption of herbicides to soil clay and organic matter fractions may play a major role in determining the amount of herbicide degraded. Additions of montmorillonite clay and charcoal to a loamy sand decreased the degradation rate of atrazine [2-chloro-4(ethylamino)-6-isopropylamino-s-triazine] and chlorthiamid (2,6-dichlorothiobenzamide) (63). Microbial decomposition of diquat [6,7-dihydrodipyrido (1,2-a:2′,1′-c) pyrazinediium ion] was quantitatively reduced by addition of montmorillonite clay to nutrient solution cultures (86). In the same study, additions of kaolinite clay had no significant effect on the decomposition of diquat. Further studies revealed that ^{14}C-diquat retained on the internal surfaces of montmorillonite clay in aqueous soil suspensions was not degradable by microorganisms over a one year period (88). At the end of the experiment, the herbicide was extracted in its original form. However, these same investigators observed that diquat and paraquat (1,1′-dimethyl-4,4′-bipyridinium ion) which was ionically adsorbed to soil organic matter was still available for microbial degradation.

Soil pH can also affect availability of herbicides to microorganisms. Microbial degradation of metribuzin [4-amino-6-tert-butyl-3-(methylthio)-as-triazin-5(4H)one] in pH adjusted soils increased as soil pH increased (53). This phenomenon was attributed to increased mobility of the non-protonated form of the compound at higher pH levels.

Glyphosate appears not to be available to plants grown in treated soils (6, 80). Studies with ^{14}C-glyphosate (72, 80) have shown that ^{14}C absorbed by both corn (Zea mays L. and soybean (Glycine max (L.) merr.) plants grown up to 8 weeks in treated soils accounted for 1 percent or less of ^{14}C applied. A bioassay study (80) revealed that adsorption rather than microbial degradation was responsible for glyphosate inactivation since plants grown in autoclaved soils exhibited no herbicidal effects.

Glyphosate does appear to be readily available to soil microorganisms in some soils since degradation studies have revealed approximately fifty percent evolution of applied ^{14}C-glyphosate as $^{14}CO_2$ occurred after approximately 1 month incubation periods (31, 71, 81). However, other soils have shown extremely low degradation rates (31, 71) even though the number of microorganisms found in these soils were comparable to the number found in soils with high degradation rates (71). This is strong evidence that glyphosate availability to microorganisms is affected by soil characteristics rather than microbial populations.

Soil adsorbent characteristics and pH are soil factors that may affect availability of glyphosate to microorganisms. Adsorption of ^{14}C-glyphosate was found to be dependent on clay type and hydroxides present in adsorbent:water suspensions (81). A soil thin-layer chromatography study (81) revealed that glyphosate mobility was very limited but was affected by soil type and pH. Increased pH levels in soil resulted in increased mobility of glyphosate and was attributed to changes in ionization of the glyphosate molecule. A bioassay study (80) showed that at high rates of glyphosate application, phytotoxicity increased as pH increased. However, a correlation study (31) on 9 soils showed that correlation coefficients calculated between glyphosate adsorption and percent clay, percent carbon, or pH were not statistically significant.

Adsorption studies with glyphosate (31, 81) have shown that glyphosate is bound to soil constituents through the phosphonic acid moiety and competed with inorganic phosphate for available sorption sites. The correlation coefficients between glyphosate adsorption and unoccupied phosphate sorption sites was statistically significant (31). Mobility on soil thin layer chromatography plates increased when soils with added orthophosphate were used (81). It appears that orthophosphate additions would increase availability of glyphosate to microorganisms although degradation studies (79) showed increased availability in only 1 of 2 soils examined.

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Valency of cations present in the soil system may also affect glyphosate availability. Bioassay studies (81) with wheat revealed that glyphosate phytotoxicity was inversely related to valency of cations present on clay and organic matter surfaces. It was suggested that the strong adsorption of glyphosate to Al^{+++} and Fe^{+++}-saturated surfaces may be due to a mechanism similar to soil formation of colloidal precipitates of Al and Fe phosphates. These forms of P are known to be somewhat available to plants but are able to form unavailable crystalline phosphates (41).

Sequence of microbial degradation of compounds such as glyphosate in soil is also of interest. Various forms of aminoalkylphosphonic acids are known to exist in nature (39, 40, 47, 48, 76). Studies (3, 15, 32, 70, 92) on the growth of soil bacteria on alkyl and aminoalkylphosphonic acids revealed that these compounds could be used as sole P sources. Nine of ten bacterial strains were found to be able to utilize P for growth from at least one of eight different aminoalkylphosphonic acids (32). It was suggested that the ability to catabolize the C-P bond is widespread among bacteria. Studies with an isolated enzyme from cell-free extracts of a strain of Bacillus cereus indicated that breakdown of 2-aminoethylphosphonic acid involved a transamination reaction (57). Other investigators (15) have shown that 3-aminopropylphosphonic acid and 2-(3-alanylamino)ethylphosphonic acid can be degraded by the bacterium,

Pseudomonas aeruginosa, without modification of the amine groups on the two substrates.

Phosphonic acids have also been known for some time to exhibit toxicological properties in some microorganisms, higher animals, and plants (26). A number of aminoalkylphosphonic acids were found to be toxic to silkworms and chick embryos and to be able to repress tobacco (Nicotiana tobacum L.) mosaic virus and tobacco rootlets (73). Metabolism studies revealed that cleavage of the carbon-phosphorous bond did not occur in kidney, liver, and plant tissue from these organisms. Another study (28) indicates that a methylphosphonate compound had slight auxin activity in pea (Pisum sativum L.) epicotyl tests. Disodium methylphosphonate was found to markedly depress the dry weight of the tops of rape (Brassica Napus L.) seedlings. Aminomethylphosphonic acid (AMPA) was found to be an extremely active growth retardant in barley (Hordeum vulgare L.) (4). These observations indicate that cleavage of the carbon-phosphorous bond does not occur readily in these plant tissues.

Ethephon [(2-chloroethyl) phosphonic acid] activity appears directly related to the release of ethylene after treatment (20, 23, 67, 85, 91). Ethylene evolution with concomittant release of phosphate and chloride ions occurred when ethephon was placed in a reaction flask with sodium hydroxide or with etiolated pea epicotyls (85). Earlier studies (60) have shown that alcohols and phenols are phosphorylated with high yield by reaction with various

2-chloroalkylphosphonic acids at 20-50°C. The presence of three or four molar equivalents of cyclohexylamine or triethylamine is required and the products formed in addition to phosphorylated compounds are the corresponding alk-1-ene and chloride ions. These observations indicate that decomposition of ethephon in plant tissue may be nonenzymatic. Ethephon additions to soil have also resulted in ethylene release which has been useful in stimulating witchweed (Striga lutea Lour.) germination (24). The decomposition mechanism here could be either enzymatic or nonenzymatic in nature.

Investigations (71, 81) have shown that degradation of glyphosate in soils is predominantly due to microbial activity. Aminomethylphosphonic acid (AMPA) was found to be the only significant metabolite when glyphosate was degraded in soil shake flasks (71). This compound then underwent rapid degradation as evidenced by measurement of $^{14}CO_2$ evolution. This suggests that soil microorganisms eventually cleave the C-P bond but as the secondary step in the degradation sequence of glyphosate. Metabolism studies (72) in corn, cotton (Gossypium hirsutum L.), soybeans, and wheat (Triticum aestivum L.) also revealed that AMPA was a primary metabolite indicating that the C-P bond cleavage was also not a primary degradation step. Incorporation of labeled material into natural products suggested that eventually the C-P bond was cleaved in these plants.

Influence of Management Practices on Herbicide Dissipation

In recent years, there has been an increased emphasis on use of nonselective and selective postemergence herbicides for weed control in cropping systems. These herbicides include glyphosate, paraquat, dalapon, bentazon in addition to several new compounds currently being developed. Since most of these compounds are not intended to have herbicidal activity in the soil, rapid dissipation of these compounds from the soil is ecologically desirable.

Procedures which acknowledge herbicide transfer and decomposition processes occurring in soil (88) may be utilized to hasten dissipation of herbicide residues present in soils (12). These procedures include correct cropping systems, fallowing and tillage, plowing, irrigation, chemical additions, and microbial additions. The emphasis in this review will be placed on the latter three procedures.

Irrigation can be used to dissipate herbicide residues in the seed environment by increasing biological and non-biological decomposition of these residues. Early studies (10, 21, 37, 52) revealed that 2,4-D [(2,4-dichlorophenoxy) acetic acid] dissipated at a faster rate in soil when the moisture content was increased. A study (69) with radioactive ring-labeled atrazine showed that microbial populations and $^{14}CO_2$ evolution were greater in soils with higher moisture content. Non-biological dissipation of ^{14}C-trifluralin (a,a,a-trifluoro-2,6-dinitro-N,N-dipropyl-p-

toluidine) was also found to be greater in soils with higher moisture (62). However, bioassay studies (29, 56) revealed that phytotoxicity of simazine [2-chloro-4,6-bis (ethylamino)-s-triazine] increased as soil moisture level increased. This would indicate that irrigation should be used as a means of dissipation of herbicide residues prior to planting crops.

Leaching by irrigation has been shown to be a useful means of herbicide dissipation in certain soils (13, 58, 68, 84). In a semi-arid region, herbicides apparently were bound and then retained in an unaltered form for long periods until water was applied (58). Rewetting the soil by irrigation water appeared to be a practical means of releasing and removing herbicide residues from these soils. A study (14) conducted on sod seeding of alfalfa (Medicago sativa L.) showed that the residual effect of glyphosate was less severe if a 35 day interval was allowed to occur between spraying and seeding. The rainfall received during this period was felt to be a contributing factor in the dissipation of the residues in the sod environment.

Additions of chemicals to soil may also represent a practical means of herbicide dissipation. Chemicals may include adsorbents, surfactants, catalysts, microbial substrates, and substances to adjust pH, absorb heat, and conserve soil moisture (12). Numerous studies (22, 29, 34, 75, 77, 83) have shown that organic matter content is the dominant soil factor affecting phytotoxicity reduction to

herbicides. In modified soil studies, addition of organic matter reduced herbicidal activity of simazine (68), prometryne [2,4-bis(isopropylamino)-6-(methylthio)-s-triazine], fluometuron [1,1-dimethyl-3-(a,a,a-trifluoro-m-tolyl)] urea, and trifluralin (89). In both studies, modification of the clay content had less of an effect on biological activity than altering organic matter content. Reduced biological activity was attributed to an increase in cation exchange capacity in the modified soils. Other investigators (51, 68) have attributed the reduction in phytotoxicity to an increase in microbial activity in soils with higher organic matter contents.

A study (78) on pyrazon [5-amino-4-chloro-2-phenyl-3(2H)-pyridazinone] dissipation showed that phytotoxicity was directly related to organic matter content of the soil. The persistent level of phytotoxicity was attributed to slow release of pyrazon from the organic matter.

Activated charcoal has received considerable attention as a means of inactivation of herbicide residues in soil (25). The mechanism responsible for this detoxication is envisioned as simple adsorption of the herbicide so that it is unavailable to the crop. Investigators found that inactivation depended on the herbicide used, dosage, plant species, quantity of activated C applied (8) and environmental conditions (35). Exposure to alternate freezing and thawing reduced the capability of activated C to detoxify atrazine in a

Si sand potting medium (35). It therefore appears that long term effects of organic matter and activated C additions on herbicide activity should be considered and examined in greater detail.

Other chemical additives may be added to the soil to modify adsorption, mobility, leaching and non-enzymatic degradation of herbicide residues. Equilibrium adsorption studies (1) revealed that fluometuron adsorption decreased and prometryne increased when $CaCl_2$ was added to the soil solution. Surfactants were found to affect depth of penetration, activity and persistence of diuron [3-(3,4-dichlorophenyl)-1,1-dimethyl urea] in soil (7). Researchers (16) found that pyridine and hydroxylamine enhanced non-enzymatic detoxification of simazine. Increased rates of chemical degradation of amitrole (3,amino-s-triazole) were observed after addition of metallic salts (44). In other studies (46) a mixture of liquid NH_3 and metallic Na was useful in detoxifying herbicide residues.

Additions of substances to serve as microbial substrates may be useful in herbicide dissipation. Soils amended with glucose, a readily available substrate, were found to have greater degradation rates of chlorobromuron [3-(4-bromo-3-chlorophenyl)-1-methoxy-1-methyl urea] (74) and fluometuron (9). Studies revealed that additions such as manure (10) and leaf mold (52) favored dissipation of 2,4-D in soils. In another study (30), soil amendments with 1

percent alfalfa was found to increase dissipation of DDT (dichlorodiphenyl trichloroethane). Garbage compost added to soils reduced phytotoxicity of fluometuron and trifluralin (39). A study (64) on organic matter amendments of variable N content revealed that lower C:N ratios favored increased rates of urea herbicide dissipation.

The addition of lime or S to adjust soil pH may be yet another means of herbicide residue dissipation. An extensive study (18) on the influence of indogenous soil pH on herbicide phytotoxicity revealed that phytotoxicity was dependent on herbicide structure. Adjustment of soil pH influenced rate of detoxication of five out of eleven herbicides studied (17). Optimum levels for detoxication of these herbicides varied from 5.3 to 7.5. A study conducted on a Hillsdale sandy loam with pH previously adjusted revealed an increase in metribuzin phytotoxicity with increasing soil pH (54). Leaching and ultimately dissipation of metribuzin residues in the same soil (53) increased with increasing soil pH. Phytotoxicity was found to be less at pH 4.5 than at 6.5 after prometryne was applied to synthetic soil media (65).

Investigators have shown that increases in soil temperature accelerate biological (10, 52, 69, 74) and nonbiological (87) degradation of herbicides in soils. In incubated soils, atrazine degraded 2 to 3 times faster with each 10°C increase in temperature between 15° and 35° (69). Studies on comparison

of herbicide persistence within regions in the United States revealed less persistence to occur in southern states (33). Previous mention has been made regarding the beneficial effect of soil moisture on herbicide degradation in soil. It therefore appears that materials such as black plastic which absorb heat and hold moisture in soil may be a useful means of herbicide dissipation.

Inoculation of soils with specific organisms may be a useful method of herbicide dissipation. Several reviews have been written on the detoxication of herbicides by specific microorganisms (19, 36, 43). Soil enrichment techniques for the proliferation of specific organisms effective in degrading herbicides (42) and other organic compounds (27) have been used by microbiologists for some time. Investigators (45) were able to successfully enhance DDT degradation by inoculating soils with Aerobacter aerogenes.

Summary

Dissipation of glyphosate appears to be primarily by microbial degradation. Procedures which increase microbial activity or glyphosate availability would appear to be useful in dissipation of residues that may be present in soils or sod environments. Other procedures may involve transfer processes such as leaching by irrigation or procedures to increase chemical decomposition. However, any method used to increase residue detoxiation must be proven to be effective and economical. Additional research in this area is needed.

CHAPTER 2

EFFECTS OF GLYPHOSATE AND SIDURON ON TURFGRASS ESTABLISHMENT

Abstract

The effects of several concentrations of glyphosate [N-(phosphonomethyl)glycine] and siduron [1-(2-methylcyclohexyl)3-phenylurea] on germination and seedling growth of Kentucky bluegrass (Poa pratensis L. 'Nugget', 'Prato', 'Fylking', 'Park'), creeping bentgrass (Agrostis palustris Huds. 'Penncross', 'Seaside'), and red fescue (Festuca rubra L. 'Pennlawn', 'Wintergreen') were evaluated in petri dishes under controlled environmental conditions. A high concentration of glyphosate (10^{-4} M) reduced shoot growth in all but two cultivars. Siduron had no effect. Glyphosate or siduron applied to the soil and incorporated did not affect turfgrass emergence or growth. When the glyphosate was sprayed over the soil and on the seed lying on the soil surface, red fescue germination was reduced and shoot growth of Kentucky bluegrass, creeping bentgrass, and red fescue was inhibited at the high rate of glyphosate application (17.9 kg/ha). Under field conditions, none of the glyphosate treatments reduced turfgrass emergence or growth.

Introduction

Glyphosate, a nonselective postemergence herbicide introduced in 1971, has excellent herbicidal activity on a diverse group of perennial grasses (1, 3). Glyphosate also has been shown to be absorbed and translocated basipetally in numerous annual and perennial herbaceous plants (4). Applications of glyphosate at proposed rates did not injure crops planted immediately after herbicide treatment (7). Therefore, it appears that glyphosate would be an excellent chemical for renovating turfgrass communities.

Siduron is used to control germinating annual weed grasses in newly seeded Kentucky bluegrass, red fescue, and some varieties of creeping bentgrass (2).

The purpose of this study was to determine if treatments of glyphosate and siduron affected percent germination and growth of seedlings of Kentucky bluegrass, creeping bentgrass, red fescue and perennial ryegrass (Lolium perenne L. 'Pennfine') in the laboratory and the field.

Materials and Methods

The effects of glyphosate and siduron on percent germination and shoot lengths of three turfgrass species were studied by germinating the seeds in petri dishes in a controlled environment chamber. Twenty-five seeds each of 'Nugget' Kentucky bluegrass, 'Prato' Kentucky bluegrass, 'Fylking' Kentucky bluegrass, 'Park' Kentucky bluegrass,

'Penncross' bentgrass, 'Seaside' bentgrass, 'Pennlawn' red fescue, and 'Wintergreen' red fescue were placed on two layers of Whatman No. 2 filter papers in the petri dishes. Each turfgrass variety was treated with 10 ml per petri dish of 10^{-6} M, 10^{-5}M, and 10^{-4} M glyphosate and 10 ml of 10^{-6} M, 10^{-5} M, and 10^{-4} M siduron. Additions of 10 ml of distilled water to the seeds of each of the eight turfgrass varieties served as the control. The dishes were placed in a growth chamber with a photoperiod of 14 hr, at 25±1 C and a 10-hr dark period of 20±1 C, and moistened with distilled water when necessary. Germination counts and shoot length measurements were made 12 days later for the bentgrass and red fescue varieties and 19 days later for the Kentucky bluegrass varieties. The treatments were replicated four times in a completely randomized design.

A study was initiated in the growth chamber to determine if soil applications of glyphosate and siduron reduced percent germination, emergence and plant heights of the same turfgrass species. Styrofoam flats 14.5 cm by 10 cm were filled with a Conover loam soil containing 2.2% organic matter with a pH of 7.0 and randomly split into three equal sections in order to accommodate one species per section. Preemergence applications were simulated by applying glyphosate as a spray at 0, 1.1, 2.2, 4.5 and 17.9 kg/ha and siduron at 11.2 and 22.4 kg/ha at 935 L/ha volume and 2.11 kg/cm^2 pressure after fifty seeds each of 'Park' Kentucky bluegrass, 'Penncross' bentgrass, and 'Pennlawn' red fescue were placed

on the soil surface and pressed lightly into the top 0.1 cm layer of soil. Preplant incorporated applications were simulated by applying glyphosate at 1.1, 2,2, 4.5, and 17.9 kg/ha and incorporating in the upper 0.6 cm of soil followed by placing fifty seeds of each turfgrass species 0.3 cm deep in the treated soil. All flats were subirrigated daily during the first week and then surface irrigated the second week. After 14 days, germination and emergence counts and plant height measurements were made for each turfgrass species. The treatments were replicated three times in a completely randomized design.

Data presented in Tables 1 to 4 are the means of two experiments with two replications in each and in Tables 5 and 6 are the means of two experiments with three replications each.

A field study was initiated July 31, 1975, in Urbana, Illinois, on a Flanagan silt loam soil containing 6% organic matter with a pH of 6.6 to determine the effects of preplant incorporated and preemergence applications of glyphosate on emergence and subsequent growth of 'A-34' Kentucky bluegrass and 'Pennfine' perennial ryegrass.[1] Seeding rates were 1 and 3 kg/are for the 'A-34' and 'Pennfine', respectively. Glyphosate was applied at 0, 1.1, 2.2, 4.5, 9.0 and 17.9 kg/ha prior to seeding, and lightly raked into the top 2.5 cm of

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[1]This study was done by A. J. Turgeon, Asst. Prof. Dept. of Hort., Univ. of Illinois, Urbana.

soil, or immediately after seeding. Plots were 1.52 m^2 each in area. Irrigation was performed as needed to encourage germination. Treatments were replicated three times in a randomized complete block design. After 5 weeks, clipping yields were measured using a 2.5 cm cutting height. Afterward, the plots were moved twice weekly at 3.8 cm. Stand counts were made 8 weeks after planting from two 25 cm^2 plugs taken from each replication.

Results and Discussion

Treatments of glyphosate and siduron caused no significant reduction in percent germination of the eight turfgrass varieties tested (Tables 1 and 2). In the case of 'Wintergreen' red fescue and 'Park' Kentucky bluegrass an increase in percent germination was observed at the low rates of glyphosate compared to the control. Shoot lengths of all four Kentucky bluegrass varieties were severely reduced when seeds were treated with a 10^{-4} M glyphosate solution (Table 3). Reduction in shoot lengths were also observed for 'Penncross' creeping bentgrass and 'Pennlawn' red fescue (Table 4). Thus, under conditions allowing absorption of glyphosate, turfgrass growth could be inhibited by the 10^{-4} M glyphosate treatment although germination did not appear to be affected.

Preemergence application of glyphosate at 1.1, 2,2, 4.5, and 17.9 kg/ha to the soil did not significantly reduce percent germination of Kentucky bluegrass, creeping bentgrass or red fescue with the exception of the 17.9 kg/ha rate on

Table 1. The effect of glyphosate and siduron treatments on germination of four Kentucky bluegrass varieties grown in petri dishes.

Treatment	Percent germination[a]			
	'Nugget' (%)	'Prato' (%)	'Fylking' (%)	'Park' (%)
Control	66 a	64 a	90 a	57 a
10^{-6} M glyphosate	72 a	66 a	82 a	72 ab
10^{-5} M glyphosate	68 a	65 a	79 a	81 b
10^{-4} M glyphosate	52 a	56 a	73 a	66 ab
10^{-6} M siduron	80 a	75 a	83 a	78 ab
10^{-5} M siduron	78 a	68 a	76 a	77 ab
10^{-4} M siduron	64 a	61 a	73 a	65 ab

[a]Means within columns with common letters are not significantly different at the 5% level by Duncan's multiple range test. Percent values were transformed to the arcsine for statistical analysis.

Table 2. The effect of glyphosate and siduron treatments on percent germination of bentgrass and red fescue grown in petri dishes.

Treatment	Percent germination[a]			
	'Penncross' creeping bentgrass (%)	'Seaside' creeping bentgrass (%)	'Pennlawn' red fescue (%)	'Wintergreen' red fescue (%)
Control	90 a	56 a	82 a	40 a
10^{-6} M glyphosate	86 a	68 a	72 a	60 b
10^{-5} M glyphosate	82 a	57 a	77 a	52 ab
10^{-4} M glyphosate	86 a	66 a	68 a	50 ab
10^{-6} M siduron	96 a	62 a	74 a	51 ab
10^{-5} M siduron	89 a	67 a	87 a	54 ab
10^{-4} M siduron	75 a	65 a	71 a	49 ab

[a]Means within columns with common letters are not significantly different at the 5% level by Duncan's multiple range test. Percent values were transformed to the arcsine for statistical analysis.

Table 3. The effect of glyphosate and siduron treatments on the shoot length of four varieties of Kentucky bluegrass grown in petri dishes.

Treatment	Shoot lengths[a]			
	'Nugget' (mm)	'Prato' (mm)	'Fylking' (mm)	'Park' (mm)
Control	12.6 b	11.1 bc	14.7 c	18.9 b
10^{-6} M glyphosate	15.9 c	9.8 b	14.8 c	19.3 b
10^{-5} M glyphosate	14.4 bc	9.1 ab	10.5 ab	19.7 b
10^{-4} M glyphosate	8.6 a	7.4 a	7.9 a	15.3 a
10^{-6} M siduron	15.6 c	12.7 c	14.4 c	19.6 b
10^{-5} M siduron	15.4 bc	12.5 c	13.1 bc	22.3 b
10^{-4} M siduron	15.3 bc	10.7 bc	13.6 bc	22.1 b

[a]Means within columns with common letters are not significantly different at the 5% level by Duncan's multiple range test.

Table 4. The effect of glyphosate and siduron treatments on the shoot lengths of bentgrass and red fescue grown in petri dishes.

Treatment	Shoot lengths[a]			
	'Penncross' creeping bentgrass (mm)	'Seaside' creeping bentgrass (mm)	'Pennlawn' red fescue (mm)	'Wintergreen' red fescue (mm)
Control	10.9 b	7.3 a	30.0 c	17.5 ab
10^{-6} M glyphosate	9.6 b	9.7 a	26.7 bc	24.2 b
10^{-5} M glyphosate	10.2 b	10.1 a	25.3 bc	22.5 ab
10^{-4} M glyphosate	7.9 a	7.0 a	15.5 a	13.7 a
10^{-6} M siduron	10.8 b	9.1 a	19.5 ab	25.8 b
10^{-5} M siduron	10.7 b	9.9 a	28.4 c	24.2 b
10^{-4} M siduron	10.4 b	8.9 a	25.0 bc	25.0 b

[a]Means within columns with common letters are not significantly different at the 5% level by Duncan's multiple range test.

red fescue (Table 5). Following the 17.9 kg/ha preemergence application where the spray in direct contact with the seed lying on the soil surface, the plant height was reduced for all three turfgrass species (Table 6). However, when glyphosate was applied at this rate preplant and incorporated, no significant reduction in plant height of the three turfgrass species occurred. This is evidence that the glyphosate was rapidly inactivated by the soil as documented by previous reports (5, 6). Siduron did not appear to have a deleterious effect on germination or the shoot heights of the three turfgrass species grown in the controlled environment chamber (Table 6).

In a field study, no significant reductions in clipping yield or stand density of 'A-34' Kentucky bluegrass or 'Pennfine' perennial ryegrass resulted from preplant incorporated or preemergence applications of glyphosate (Tables 7 and 8). However, a trend toward higher density and yield was observed with increasing rate of glyphosate.

The results of this study indicate that a number of turfgrass species can be safely established following glyphosate application even under abnormally high herbicide application rates.

Table 5. The effect of glyphosate and siduron applications on percent germination of three turfgrass species grown in soil in the controlled environment chamber.

Treatment	Percent germination[a]		
	Kentucky bluegrass (%)	Creeping bentgrass (%)	Red fescue (%)
Control	63 ab	66 ab	70 b
Glyphosate, 1.1 kg/ha PRE[b]	60 ab	68 ab	69 b
Glyphosate, 2.2 kg/ha PRE	57 ab	68 ab	69 b
Glyphosate, 4.5 kg/ha PRE	57 ab	77 b	69 b
Glyphosate, 17.9 kg/ha PRE	50 a	54 a	51 a
Siduron, 11.2 kg/ha PRE	63 ab	70 ab	69 b
Siduron, 22.4 kg/ha PRE	67 b	72 ab	65 b
Glyphosate, 1.1 kg/ha PPI[c]	60 ab	64 ab	61 ab
Glyphosate, 2.2 kg/ha PPI	67 b	66 ab	66 b
Glyphosate, 4.5 kg/ha PPI	60 ab	68 ab	69 b
Glyphosate, 17.9 kg/ha PPI	58 ab	70 ab	67 b

[a]Means within columns with common letters are not significantly different at the 5% level by Duncan's multiple range test. Percent values were transformed to the arcsine for statistical analysis.

[b]PRE = preemergence herbicide application.

[c]PPI = preplant incorporated herbicide application.

Table 6. The effect of glyphosate and siduron applications on the plant heights of three turfgrass species grown in soil in the controlled environment chamber.

Treatment	Plant heights[a]		
	Kentucky bluegrass (cm)	Creeping bentgrass (cm)	Red fescue (cm)
Control	2.7 b	3.1 c	5.7 b
Glyphosate, 1.1 kg/ha PRE[b]	2.4 b	3.0 c	5.7 b
Glyphosate, 2.2 kg/ha PRE	2.8 b	3.1 c	5.6 b
Glyphosate, 4.5 kg/ha PRE	2.7 b	2.6 bc	5.5 b
Glyphosate, 17.9 kg/ha PRE	1.5 a	1.1 a	4.1 a
Siduron, 11.2 kg/ha PRE	2.6 b	2.8 bc	5.7 b
Siduron, 22.4 kg/ha PRE	2.4 b	2.6 bc	5.7 b
Glyphosate, 1.1 kg/ha PPI[c]	2.5 b	2.8 bc	5.2 b
Glyphosate, 2.2 kg/ha PPI	2.5 b	2.7 bc	5.8 b
Glyphosate, 4.5 kg/ha PPI	2.5 b	3.0 c	5.2 b
Glyphosate, 17.9 kg/ha PPI	3.2 c	2.5 b	5.3 b

[a]Means within columns with common letters are not significantly different at the 5% level by Duncan's multiple range test.

[b]PRE = preemergence herbicide application.

[c]PPI = preplant incorporated herbicide application.

Table 7. Effects of preplant incorporated and preemergence applications of glyphosate on stand density of two turfgrass species 8 weeks after planting.

Glyphosate (kg/ha)	Kentucky bluegrass		Perennial ryegrass	
	PPI[b]	PRE[c]	PPI[b]	PRE[c]
	(No. shoots/25 cm^2)[a]			
0	25.7 a	26.5 a	19.8 a	24.3 a
1.1	25.3 a	31.8 a	23.2 a	27.2 a
2.2	33.7 a	29.0 a	27.8 a	22.2 a
4.5	17.5 a	28.8 a	15.5 a	34.8 a
9.0	36.5 a	29.0 a	28.5 a	23.3 a
17.9	30.7 a	20.3 a	27.3 a	15.2 a

[a]Means within columns with common letters are not significantly different at the 5% level by Duncan's multiple range test.

[b]PPI = preplant incorporated herbicide application.

[c]PRE = preemergence herbicide application.

Table 8. Effects of preplant incorporated and preemergence applications of glyphosate on clipping yield of two turfgrass species 5 weeks after planting.

Glyphosate (kg/ha)	Kentucky bluegrass		Perennial ryegrass	
	PPI[b]	PRE[c]	PPI[b]	PRE[c]
	(g clipping dry wt/56 dm^2)[a]			
0	1.29 a	1.29 a	28.25 a	27.53 a
1.1	1.62 a	1.39 a	31.75 a	24.67 a
2.2	1.74 a	1.80 a	30.98 a	28.47 a
4.5	2.67 a	1.51 a	28.59 a	28.28 a
9.0	2.05 a	1.23 a	38.13 a	30.60 a
17.9	2.40 a	1.75 a	34.72 a	31.67 a

[a]Means within columns with common letters are not significantly different at the 5% level by Duncan's multiple range test.

[b]PPI = preplant incorporated herbicide application.

[c]PRE = preemergence herbicide application.

Literature Cited

1. Baird, D. D., R. P. Upchurch, W. B. Homesley, and J. F. Franz. 1971. Introduction of a new broad spectrum post-emergence herbicide class with utility for herbaceous perennial weed control. Proc. N. Cent. Weed Contr. Conf. 26:64-68.

2. Herbicide Handbook Weed Science Society of America. 1974. 3rd Ed. Weed Science Society of America, Champaign, Illinois. 430 pp.

3. Sprankle, P. and W. F. Meggitt. 1972. Effective control of quackgrass with fall and spring applications of glyphosate. Proc. N. Cent. Weed Control Conf. 27:54.

4. Sprankle, P., W. F. Meggitt, and D. Penner. 1975. Adsorption, action and translocation of glyphosate. Weed Sci. 22:235-240.

5. Sprankle, P., W. F. Meggitt, and D. Penner. 1975. Absorption mobility and microbial degradation of glyphosate in the soil. Weed Sci. 22:229-234.

6. Sprankle, P., W. F. Meggitt, and D. Penner. 1975. Rapid inactivation of glyphosate in the soil. Weed Sci. 22: 224-228.

7. Upchurch, R. P. and D. D. Baird. 1972. Herbicidal action of MON 9573 as influenced by light and soil. Proc. West. Weed Sci. Soc. 25:41-45.

CHAPTER 3

USE OF GLYPHOSATE IN SOD SEEDING ALFALFA ESTABLISHMENT

Abstract

The effect of glyphosate [N-(phosphonomethyl)glycine] on alfalfa (Medicago sativa L.) germination, emergence, and seedling growth was studied to evaluate the potential of glyphosate for weed control in alfalfa establishment in forage renovation. Glyphosate and EPTC (S-ethyl dipropylthiocarbamate), a selective preemergence herbicide used in alfalfa establishment, caused no significant reduction in percent germination of either Saranac or Vernal cultivars grown in petri dishes. Glyphosate significantly reduced shoot length of both cultivars. Soil applications of glyphosate also had no effect on percent germination of either cultivar and only reduced plant height when applied to the soil at 17.9 kg/ha. Applications of glyphosate at 2.2, 4.5, and 9.0 kg/ha to a Kentucky bluegrass (Poa pratensis L.) sod in a simulated sod seeding alfalfa establishment study in the greenhouse reduced dry weight of both alfalfa cultivars. Application of glyphosate in four management systems showed that a 3 day planting delay or water application after spraying reduced growth inhibition of alfalfa seeded in treated sod. These results

were similar to those obtained with paraquat (1,1'-dimethyl-4,4'-bipyridinium ion) in these management systems. Kentucky bluegrass clippings treated with paraquat 3 days or less before contact with alfalfa seedlings significantly reduced seedling dry weight. Clippings treated with glyphosate did not significantly reduce alfalfa seedling dry weights.

Introduction

Alfalfa establishment in a renovation program can improve productivity and nutritional quality of large acreages of forage within the North Central states. This acreage includes both old hay fields and pastures where perennial grasses such as quackgrass (Agropyron repens L.) and Kentucky bluegrass have become the dominant species. Farmers can renovate these forage stands by utilizing one of three methods of alfalfa establishment. One method is to till the land, then plant alfalfa and a cereal crop as a companion crop. A second method is to plant alfalfa alone in cultivated soil and use an appropriate preemergence herbicide to control annual weeds. A third method is no-tillage sod seeding. This method involves applications of a nonselective herbicide to suppress prior existing perennial species followed by seeding of alfalfa directly into sod.

Glyphosate has potential in forage renovation for removing existing vegetation because of three key characteristics: 1) it is a non-selective herbicide and therefore will control

a wide spectrum of perennial species, 2) it is readily translocated throughout these treated plants, and 3) it is rapidly inactivated within most soils.

Recent reports (3, 4) have shown that glyphosate sprayed at 4.5 kg/ha, did not inhibit germination of Kentucky bluegrass, red fescue (Festuca rubra L.), tall fescue (F. arundinacea Schreb.) and creeping bentgrass (Agrostis palustris Huds.) when applied to the soil before seeding or when applied directly over the seeds. Red clover (Trifolium pratense L.) germination was inhibited when glyphosate was applied at 2.0 kg/ha with seeds covered by soil at the time of treatment (5). Germination of annual ryegrass (Lolium multiflorum Lam.) seed, directly treated or covered by soil, was reduced when glyphosate was applied at 3.0 kg/ha and 4.0 kg/ha, respectively (5). In petri dish studies (2), glyphosate applied at 2.2 kg/ha and in direct contact with seeds, did not inhibit germination of prickly sida (Sida spinosa L.), velvet leaf (Abutilon theophrasti Medic.), barnyard grass [Echinochloa crusgalli (L.) Beauv.], and johnsongrass [Sorghum halepeuse (L.) Pers.]. Glyphosate, at 2.2 kg/ha, caused a 1.6 to 3-fold stimulation in red root pigweed (Amaranthus retroflexus L.) germination. Radicle growth inhibitions of the germination was observed with all five weed species. Other petri dish studies (4) showed that shoot lengths of Kentucky bluegrass, creeping bentgrass, and red fescue were reduced when seeds were treated with 10^{-4} M glyphosate solution.

The purpose of this study was to determine whether glyphosate could be safely used for establishing alfalfa by assaying glyphosate effects on percent germination and seedlings growth under several management systems. Furthermore, it was of interest to compare the effects of glyphosate with those of EPTC and paraquat.

Materials and Methods

The effect of glyphosate on percent germination and shoot lengths of two alfalfa cultivars was studied by germinating the seeds in petri dishes in a growth chamber within a day temperature of 24°C and night temperature of 14°C in a 16 hr photoperiod regime. Fifty seeds each of Saranac or Vernal alfalfa were placed on two layers on Whatman No. 2 filter papers in the petri dishes containing 10 ml of 0, 10^{-6} M, 10^{-5} M, and 10^{-4} M glyphosate or 10 ml of 10^{-6} M, 10^{-5} M, and 10^{-4} M EPTC. EPTC was included in this study because it is recommended for use in preemergence weed control in alfalfa (6). Germination counts and shoot length measurements were made after 7 days. The treatments were replicated three times in a completely randomized design.

A study was initiated in the growth chamber to determine if applications of glyphosate to the soil reduced percent emergence and plant heights of the two alfalfa cultivars. Styrofoam flats 14.5 cm by 10 cm were filled with a Spinks sandy loam containing 3.9% organic matter with a pH of 6.1 and

split into two equal sections to accommodate one cultivar per section. Preplant incorporated applications were simulated by applying glyphosate at 0, 2.2, 4.5, 9.0, and 17.9 kg/ha a.i. at 935 L/ha volume and 2.11 kg/cm^2 pressure and incorporating in the upper 0.6 cm of soil followed by placing 50 seeds 0.3 cm deep in the treated soil. Preemergence applications were simulated by applying glyphosate at 0, 2.2, 4.5, 9.0 and 17.9 kg/ha after 50 seeds of each cultivar were placed 0.3 cm deep in soil. All flats were subirrigated daily throughout the study. After 14 days, emergence counts and plant height measurements were made for each alfalfa cultivar. The treatments were replicated three times in a completely randomized design.

The effects of glyphosate applications on alfalfa in a simulated sod seeding system were studied in the greenhouse. Sixty seeds of Saranac and Vernal were planted in Kentucky bluegrass sod grown in flats 35 cm by 51.4 cm and split equally to accommodate one cultivar per section. Glyphosate then was applied at 0, 2.2, 4.5, and 9.0 kg/ha at 234 L/ha volume and 1.83 kg/cm^2 pressure. Alfalfa was also planted in nontreated soil which then served as a bare soil control. After 4 weeks, the alfalfa was harvested, surviving plants counted and dry weights measured. The treatments were replicated three times in a completely randomized design.

A second study was initiated to determine the interaction between glyphosate applications and four management systems on subsequent growth of alfalfa. Fifty seeds of

Vernal alfalfa were planted in a Kentucky bluegrass sod grown in flats 15 cm by 30 cm and glyphosate then applied at 0, 2.2, 4.5, and 9.0 kg/ha. Paraquat was also applied at 0.6 and 1.2 kg/ha. Applications were made at the time of seeding or prior to seeding according to the management system involved. The first two management systems consisted of single spray-seed operations. The second management system also involved application of 2.5 cm of water 1 day after herbicide treatment. The third and fourth management systems consisted of a 3-day delay in planting after herbicide application. No water was applied for a period of 7 days in the third system whereas in the fourth system 2.5 cm of water was applied 1 day after the herbicide was applied. Alfalfa was also planted in non-treated soil which then served as a bare soil control. Four weeks after planting the alfalfa was harvested and the dry weight measured. The treatments were replicated three times in a randomized complete block design.

Kentucky bluegrass clippings treated with paraquat or glyphosate were placed in contact with alfalfa seedlings to determine whether they affected subsequent growth. Alfalfa was planted in a potting soil mix, thinned to 100 plants per flat and grown for a period of 2 weeks in the greenhouse. Concurrently, Kentucky bluegrass sod in flats was allowed to grow to a height of 15 cm and then was sprayed with paraquat at 0 or 0.6 kg/ha or glyphosate at 2.2 kg/ha 0, 3, 7, and 10 days prior to clipping and placed in contact with the alfalfa

seedlings. The flats were subirrigated and also surface irrigated daily with a 10 second fine mist spray. The alfalfa plants were harvested 4 weeks later and the dry weight measured. The treatments were replicated three times in a completely randomized design. All data presented are the means of two experiments.

Results and Discussion

Glyphosate and EPTC treatments caused no significant reduction in percent germination of either cultivar examined in petri dish studies (Table 1). Glyphosate treatments at 10^{-6} M concentration did not have a detrimental effect on shoot length of Saranac cultivar but did reduce shoot length of the Vernal cultivar (Table 2). Increasing concentrations of glyphosate further inhibited shoot length of both cultivars. It therefore appears that under conditions allowing absorption of glyphosate, alfalfa seedling growth can be inhibited.

Soil applications of glyphosate had no detrimental effect on percent emergence of either cultivar even when glyphosate was applied eight times the normal use rate of 2.2 kg/ha (Table 3). Preplant incorporated applications of glyphosate did not significantly reduce plant height (Table 4) of either cultivar examined. Preemergence applications of glyphosate also did not significantly reduce plant heights of either cultivar when applied at 2.2, 4.5, and 9.0 kg/ha.

Table 1. The effect of glyphosate and EPTC treatments on germination of two alfalfa cultivars grown in petri dishes.

Treatment	Percent germination[a]	
	Saranac (%)	Vernal (%)
Control	86 a	96 a
10^{-6} M Glyphosate	86 a	92 a
10^{-5} M Glyphosate	90 a	93 a
10^{-4} M Glyphosate	85 a	93 a
10^{-6} M EPTC	85 a	94 a
10^{-5} M EPTC	84 a	93 a
10^{-4} M EPTC	86 a	93 a

[a]Means within columns with common letters are not significantly different at the 5% level by Duncan's multiple range test.

Table 2. The effect of glyphosate and EPTC treatments on the shoot length of two alfalfa cultivars grown in petri dishes.

Treatment	Shoot length[a]	
	Saranac (mm/plant)	Vernal (mm/plant)
Control	29.1 de	29.0 d
10^{-6} M Glyphosate	27.0 cd	25.8 c
10^{-5} M Glyphosate	17.4 b	17.4 b
10^{-4} M Glyphosate	12.9 a	12.5 a
10^{-6} M EPTC	25.9 c	25.9 c
10^{-5} M EPTC	27.7 cde	29.4 d
10^{-4} M EPTC	30.2 e	27.7 cd

[a]Means within columns with common letters are not significantly different at the 5% level by Duncan's multiple range test.

Table 3. The effect of glyphosate applications on percent emergence of two alfalfa cultivars grown in soil in the controlled environment chamber.

Rate (kg/ha)	Method	Percent emergence[a]	
		Saranac (%)	Vernal (%)
Control		41 a	53 ab
2.2	PPI[b]	44 a	49 ab
4.5		40 a	52 ab
9.0		38 a	47 a
17.9		38 a	44 a
2.2	PRE[c]	46 a	64 b
4.5		43 a	56 ab
9.0		43 a	46 a
17.9		45 a	49 ab

[a]Means within columns with common letters are not significantly different at the 5% level by Duncan's multiple range test.

[b]PPI = preplant incorporated herbicide application.

[c]PRE = preemergence herbicide application.

Table 4. The effect of glyphosate applications on plant heights of two alfalfa cultivars grown in soil in the controlled environment chamber.

Rate (kg/ha)	Method	Plant height[a]	
		Saranac (mm/plant)	Vernal (mm/plant)
Control		34 b	35 bc
2.2	PPI[b]	33 ab	35 bc
4.5		33 ab	33 ab
9.0		33 ab	35 bc
17.9		32 ab	33 ab
2.2	PRE[c]	36 bc	34 ab
4.5		34 b	35 bc
9.0		38 c	36 c
17.9		31 a	31 a

[a]Means within columns with common letters are not significantly different at the 5% level by Duncan's multiple range test.

[b]PPI = preplant incorporated herbicide application.

[c]PRE = preemergence herbicide application.

Only when glyphosate was applied at eight times the normal use rate, did a significant reduction in plant height occur. It therefore appears that glyphosate applications onto bare soil would result in injury to alfalfa seedlings only when applied at high rates and allowed to remain on the soil surface.

In the simulated sod-seeding study, percent survival of Saranac alfalfa grown in sod sprayed immediately after planting with glyphosate at 2.2 kg/ha was not significantly different from that grown in non-treated soil (Table 5). Percent survival of Vernal alfalfa was significantly reduced when grown in sod treated with glyphosate at 2.2 kg/ha. Increasing rates of glyphosate further reduced percent survival of both cultivars. Percent survival of alfalfa planted in non-treated sod was drastically reduced due to severe competition of Kentucky bluegrass. Glyphosate applied at the normal use rate did not affect dry weight yields of Saranac but did reduce yields of Vernal (Table 6). Increasing rates further inhibited growth of both cultivars. It therefore appears that glyphosate residues are present in treated bluegrass sod and are not as readily inactivated as are soil applications of glyphosate.

Interactions between paraquat applications and four management systems utilized in a second simulated sod-seeding study were observed (Table 7). Alfalfa growth in paraquat treated sod was reduced if a single spray-seed operation was

Table 5. The effect of glyphosate applications on percent survival of two alfalfa cultivars 4 weeks after planting in Kentucky bluegrass using a single spray-seed operation.

Planting medium	Glyphosate (kg/ha)	Percent survival[a]	
		Saranac (%)	Vernal (%)
Soil	0	77 e	87 f
Sod	0	15 a	12 a
	2.2	74 de	71 de
	4.5	67 cd	59 c
	9.0	44 b	45 b

[a]Means with common letters are not significantly different at the 5% level by Duncan's multiple range test.

Table 6. The effect of glyphosate applications on dry weight yields of two alfalfa cultivars 4 weeks after planting in Kentucky bluegrass sod using a single spray-seed operation.

Planting medium	Glyphosate (kg/ha)	Dry weight[a]	
		Saranac	Vernal
		(mg/900 cm^2)	
Soil	0	1697 e	2003 f
Sod	0	83 a	62 a
	2.2	1700 e	1627 e
	4.5	1615 e	1292 e
	9.0	835 c	590 b

[a]Means with common letters are not significantly different at the 5% level by Duncan's multiple range test.

Table 7. The effect of paraquat applications and four management systems on dry weight yields of 'Vernal' alfalfa 4 weeks after planting in Kentucky bluegrass sod.

Planting medium	Paraquat (kg/ha)	Dry weight[a]			
		Mgt a	Mgt b	Mgt c	Mgt d[b]
		(mg/225 cm^2)			
Soil	0	1081 cd	1117 d	1121 d	1077 cd
Sod	0	272 a	513 ab	400 a	338 a
	0.6	395 a	932 bcd	1189 d	1003 cd
	1.2	620 abc	945 bcd	1136 d	1056 cd

[a]Means with common letters are not significantly different at the 5% level by Duncan's multiple range test.

[b]Mgt a = planted, sprayed at time of planting.
Mgt b = planted, aprayed at time of planting, 2.5 cm water 1 day later.
Mgt c = sprayed, planted 3 days later, no water for 7 days.
Mgt d = sprayed, 2.5 cm water 1 day later, planted 3 days later.

used (Management System A). The dry weight yield of alfalfa grown in treated sod was not significantly different from the yield of alfalfa grown in non-treated soil if application of water simulating rainfall was made shortly after spraying (Management System B) or a 3-day lapse allowed to occur between spraying and seeding (Management Systems C and D). Alfalfa growth in non-treated sod was again severely reduced due to severe competition of Kentucky bluegrass.

Similar interactions were observed in the case of glyphosate applications (Table 8). Glyphosate applied at a normal use rate reduced alfalfa growth when applied at the time of planting (Management System A). Yields were not significantly reduced if application of water (Management System B) was made or a 3-day lapse allowed to occur between spraying and seeding (Management Systems C and D). Glyphosate applications at twice the normal use rate reduced growth when an application of water was made, but yields were not significantly reduced if a 3-day lapse was allowed to occur. Glyphosate applied at four times the normal use rate reduced growth when a 3-day lapse occurred (Management System C). At this rate, both an application of water and a 3-day lapse (Management System D) was necessary to reduce the inhibition in alfalfa growth. The results presented agree with those of Campbell (1) when he surface seeded alfalfa on glyphosate treated sod. However, in his studies an excess of 14 days was required before the danger of inhibiting alfalfa

Table 8. The effect of glyphosate application and four management systems on dry weight yields of 'Vernal' alfalfa 4 weeks after planting in Kentucky bluegrass sod.

Planting medium	Glyphosate (kg/ha)	Dry weight[a]			
		Mgt a	Mgt b	Mgt c	Mgt d[b]
		(mg/225 cm^2)			
Soil	0	1081 cde	1117 de	1121 de	1077 c-e
Sod	0	272 a	513 ab	400 ab	338 ab
	2.2	424 ab	644 a-d	1300 e	1157 de
	4.5	343 ab	586 abc	869 a-e	1209 e
	9.0	318 a	549 ab	564 abc	783 a-e

[a]Means with common letters are not significantly different at the 5% level by Duncan's multiple range test.

[b]Mgt a = planted, sprayed at time of planting.
Mgt b = planted, sprayed at time of planting, 2.5 cm water 1 day later.
Mgt c = sprayed, planted 3 days later, no water for 7 days.
Mgt d = sprayed, 2.5 cm water 1 day later, planted 3 days later.

establishment was no longer a problem. This would indicate that placing the seed in the soil increases the margin of safety.

Dry weight of alfalfa seedlings 4 weeks after initial contact with paraquat treated Kentucky bluegrass clippings were significantly reduced when paraquat was applied 3 days or less prior to contact (Table 9). Clippings treated with glyphosate and placed in contact with the alfalfa seedlings did not significantly reduce seedling dry weight even when clippings were sprayed immediately prior to contact. Studies by Klingman and Murray (3) showed that clippings treated with glyphosate did not inhibit Kentucky bluegrass, red fescue, and tall fescue germination. Clippings treated with paraquat were found to inhibit germination of the same three species.

Results presented here showed that paraquat or glyphosate residues remaining in the treated sod environment may inhibit alfalfa growth when applied at time of planting. Initial injury may result when emerging seedlings absorb residues that are present in the litter at the soil surface. Further growth inhibition may occur when treated foliage collapses onto emerged seedlings. However, results from the simulated sod seedling studies indicate that a 3-day lapse between spraying and seeding operations will allow successful use of glyphosate in alfalfa establishment.

Table 9. Dry weight of 'Vernal' alfalfa seedlings 4 weeks after initial contact with paraquat and glyphosate treated clippings.

Application	Time prior to contact (days)	Dry weight[a] (g/100 plants)
Control	-	1.83 b
Paraquat 0.6 kg/ha	0	1.35 a
	3	1.38 a
	7	1.48 ab
	10	1.56 ab
Glyphosate 2.2 kg/ha	0	1.47 ab
	3	1.72 ab
	7	1.49 ab
	10	1.50 ab

[a]Means followed by common letters are not significantly different at the 5% level by Duncan's multiple range test.

Literature Cited

1. Campbell, M. H. 1974. Effects of glyphosate on the germination and establishment of surface-sown pasture species. Aust. J. Exp. Anim. Husb. 14:557-560.

2. Egley, G. H. and R. D. Williams. 1977. Effects of glyphosate and paraquat upon weed seed germination. Weed Sci. Soc. Amer. Abstr. p. 81.

3. Klingman, D. L. and J. J. Murray. 1976. Germination of seeds of turfgrasses as affected by glyphosate and paraquat. Weed Sci. 24:191-193.

4. Moshier, L., A. J. Turgeon, and D. Penner. 1976. Effects of glyphosate and siduron on turfgrass establishment. Weed Sci. 24:445-448.

5. Segura, J., S. W. Bingham, and C. L. Foy. 1977. Susceptibility of some pasture species to glyphosate. Weed Sci. Soc. Amer. Abstr. p. 55.

6. Weed Science Society of America. 1974. Herbicide Handbook, 3rd ed., Champaign, Illinois. 430 pp.

CHAPTER 4

FACTORS INFLUENCING MICROBIAL DEGRADATION OF ^{14}C-GLYPHOSATE TO $^{14}CO_2$ IN SOIL

Abstract

Glyphosate degradation occurred primarily by microorganisms and varied among the three soils examined. Phosphate additions stimulated glyphosate degradation to a limited extent in the Collamer silt loam but not in the Norfolk loamy sand. Additions of Fe^{+++} and Al^{+++} ions reduced degradation in the Spinks sandy loam. It is postulated that formation of colloidal Fe and Al precipitates in modified soils with concomitant adsorption of glyphosation is responsible for decreased availability of glyphosate to microorganisms. Mn^{++} additions were found to increase degradation. Spinks soil and carbon substrate amendments failed to increase degradation in both soils with low degradation rates.

Introduction

Herbicides, applied to the soil, may be subject to microbial, chemical and photodecomposition. Photodecomposition of glyphosate has been reported to be negligible (7, 12). Chemical degradation has also been found to be of minor importance for glyphosate applied to soil (7, 10, 12). After

28 days, evolution of $^{14}CO_2$ accounted for 1 and 17 percent of total ^{14}C applied as ^{14}C-glyphosate in a sterilized and non-sterilized soil respectively (10) providing evidence that microbial degradation is the predominant mechanism for glyphosate decomposition in soils.

Past studies (1, 7, 10, 12) have shown a wide variation in the degradation rate of ^{14}C-glyphosate in the soils. This variation may be due either to the level of microbial activity present in these soils or to availability of the herbicide in the soil environment to microbial degradation. Factors influencing microbial activity such as pH, moisture, temperature, organic substrate levels and nutrient levels are known to affect herbicide degradation in soil (3). Attempts have also been made to increase herbicide degradation by soil inoculation techniques (2, 4). Few investigations have dealt with the effect of herbicide availability on microbial degradation. On a Hillsdale sandy loam soil with a previously established pH range, microbial degradation of metribuzin increased with increasing pH (5). This increase was attributed to decreased binding of the nonprotonated form of the compound at higher soil pH.

The objectives of this study were to determine the influence of various factors on degradation of glyphosate to CO_2 in three different soil types.

Materials and Methods

Soils studied included a Spinks sandy loam, a Collamer silt loam from New York State, and a Norfolk loamy sand from Virginia. Chemical characteristics of the soils studied as determined by the soil testing laboratory at Michigan State University, are presented in Table 1. Glyphosate degradation in various soils collected in summer of 1976 was investigated according to procedures adapted from Tiedje and Mason (11). A 25 gram portion of each soil was placed in 125 ml erlenmeyer flasks and received 5 ml of 6.3×10^{-4} M NH_4NO_3. A 1-ml portion of solution containing 0.005 uCi of ^{14}C-methyl-labeled glyphosate (spec. act.=1.0 mCi/mmole) was added to each flask. Three days prior to glyphosate application the 25 grams of the Spinks sandy loam soil also received 3 ml of propylene oxide which was allowed to volatilize. This treatment served as the sterilized soil control. A 2-ml beaker containing 1 M KOH was suspended in each flask to absorb the $^{14}CO_2$. The flasks were sealed and placed at 25 ± 2°C under room light and sampled 1, 2, 4, 8, 16, and 32 days later. At sampling time, the contents of the 2-ml beaker were placed in a vial containing 15 ml of a solution consisting of 1:1 (v/v) CAB-O-SIL[1] solution: 60 g naphthalene, 4 g PPO, 0.2 g POPOP, 100 ml methanol, 20 ml ethylene glycol made up to a liter with p-1,4-dioxane. The samples were radioassayed by liquid scintillation spectrometry.

[1]Product of Research Products Intl. Corp. Elk Grove Village, ILL 60007.

Table 1. Chemical characteristics of soils used in glyphosate degradation study.

Soil type	pH	Organic matter (%)	Phosphate (kg/ha)
Collamer silt loam	5.6	3.7	68
Norfolk sand	4.4	1.2	72
Spinks sandy loam	6.6	3.8	302

The effect of P addition on glyphosate degradation was examined by incorporating $CaHPO_4$ into the low P Collamer and Norfolk soils at concentrations of 0, 200, 400, and 800 ppm and 0, 200, and 400 ppm oven dry soil basis respectively. After thorough mixing, 25 g samples of each of the amended soils were incubated for 1, 2, 4, 8, 16, and 32 days for the degradation study.

Studies on cation influence on glyphosate degradation included additions of $FeCl_3 \cdot 6H_2O$, $FeNH_4(SO_4)_2 \cdot 12H_2O$, Fe_2O_3, $AlCl_3$, $CaCl_2$, $MnCl_2 \cdot 4H_2O$ and NaCl. Ferric chloride was incorporated into the Spinks sandy loam soil at a concentration of 0, 200, 400 Fe^{+++} ppm. Other ferric compounds and metal salts were incorporated to obtain a concentration of 200 ppm of the cation in soil for comparison. Incubation periods for the ferric chloride study were 1, 2, 4, 8, 16, and 32 days and for the Fe^{+++} compound and cation comparison studies were 1, 2, and 8 days.

The effect of soil pH on microbial degradation of glyphosate was examined by addition of 0, 0.1, 1.0, and 10.0 milliequivalents of ammonium carbonate to the low pH Norfolk soil. Glyphosate then was added to the soil and degradation monitored for incubation periods of 1, 2, 8 and 16 days.

To ascertain whether the low level of ^{14}C-glyphosate conversion to $^{14}CO_2$ in the Collamer and Norfolk soils was related to lack of microbial content, presence of microbial inhibitors, or insufficient substrate to support microbial population, a series of experiments with soil amendments was

initiated. Five grams of non-sterilized Spinks sandy loam soil, known to be active in degrading glyphosate, was added to 20 g of sterilized Spinks, non-sterilized Collamer and Norfolk soils. Five ml of a combined solution of 6.3×10^{-4} M NH_4NO_3 and 0.1% glucose was added to a Spinks sandy loam soil prior to addition of glyphosate. Five ml of 0.5% glucose, sarcosine, and glycine was added to a Norfolk soil as a means of comparing different microbial substrates. Alfalfa meal was also incorporated in soil to obtain a concentration equal to that of other organic substrates. The Norfolk soil was selected because of the low organic matter content in this soil (Table 1). Incubation periods were 1, 2, 8, and 32 days for the Spinks soil inoculum and glucose amendment studies and 1, 2, and 8 days for the substrate comparison study.

All data presented are the means of two experiments with three replications per experiment.

Results and Discussion

Glyphosate degradation occurred readily in the non-sterilized Spinks sandy loam soil and was severely reduced by soil sterilization (Figure 1). This evidence confirms that microbial degradation is the predominant mechanism in soil degradation of glyphosate. Evolution of $^{14}CO_2$ accounted for 3.0 and 9.5 percent of ^{14}C applied as glyphosate in the Norfolk and Collamer soils, respectively, whereas in the Spinks soil, $^{14}CO_2$ evolution accounted for 40 percent of applied ^{14}C after 32 days (Figure 2).

Figure 1. Patterns of ^{14}C-glyphosate degradation in Spinks soil.

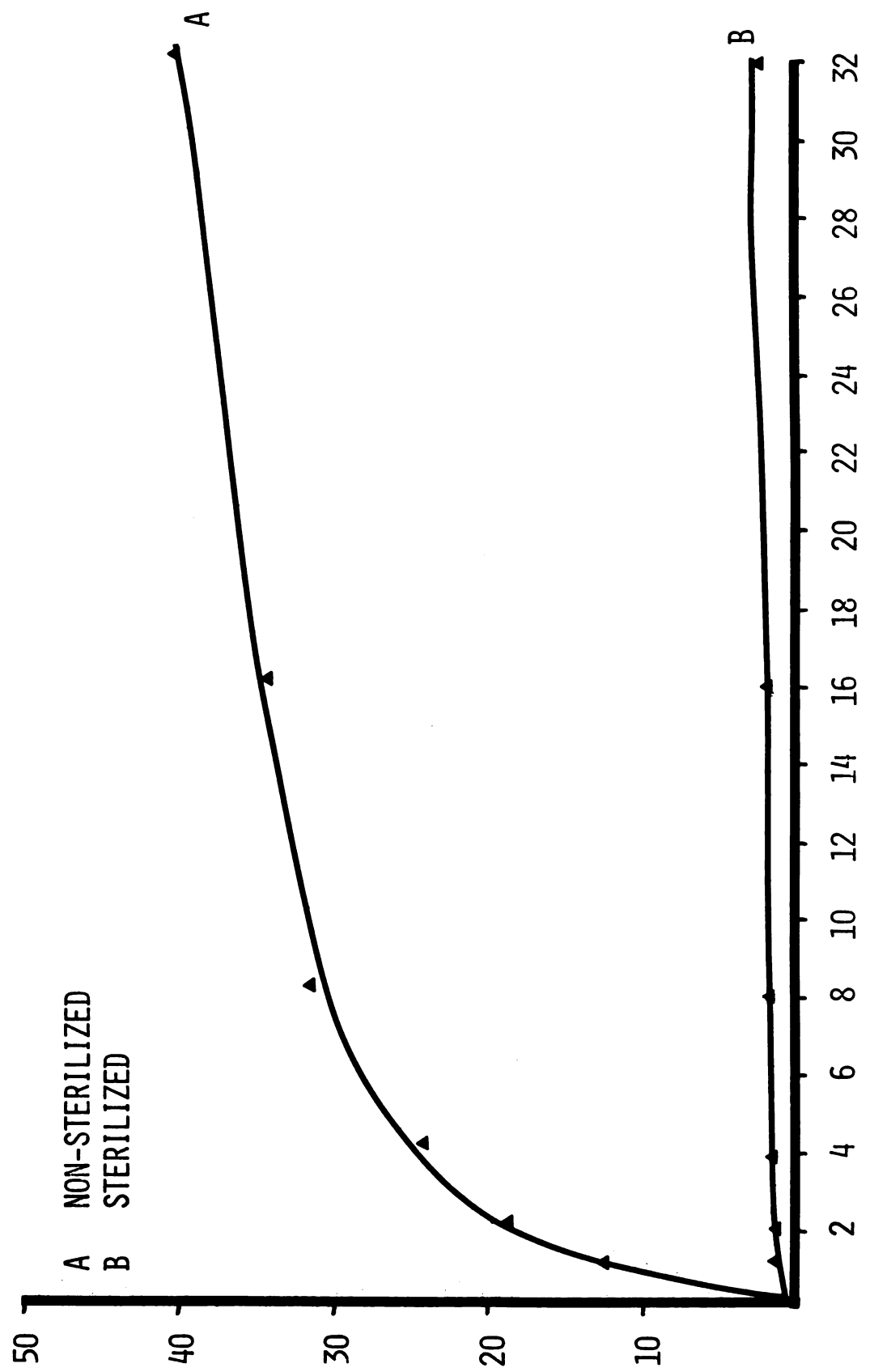
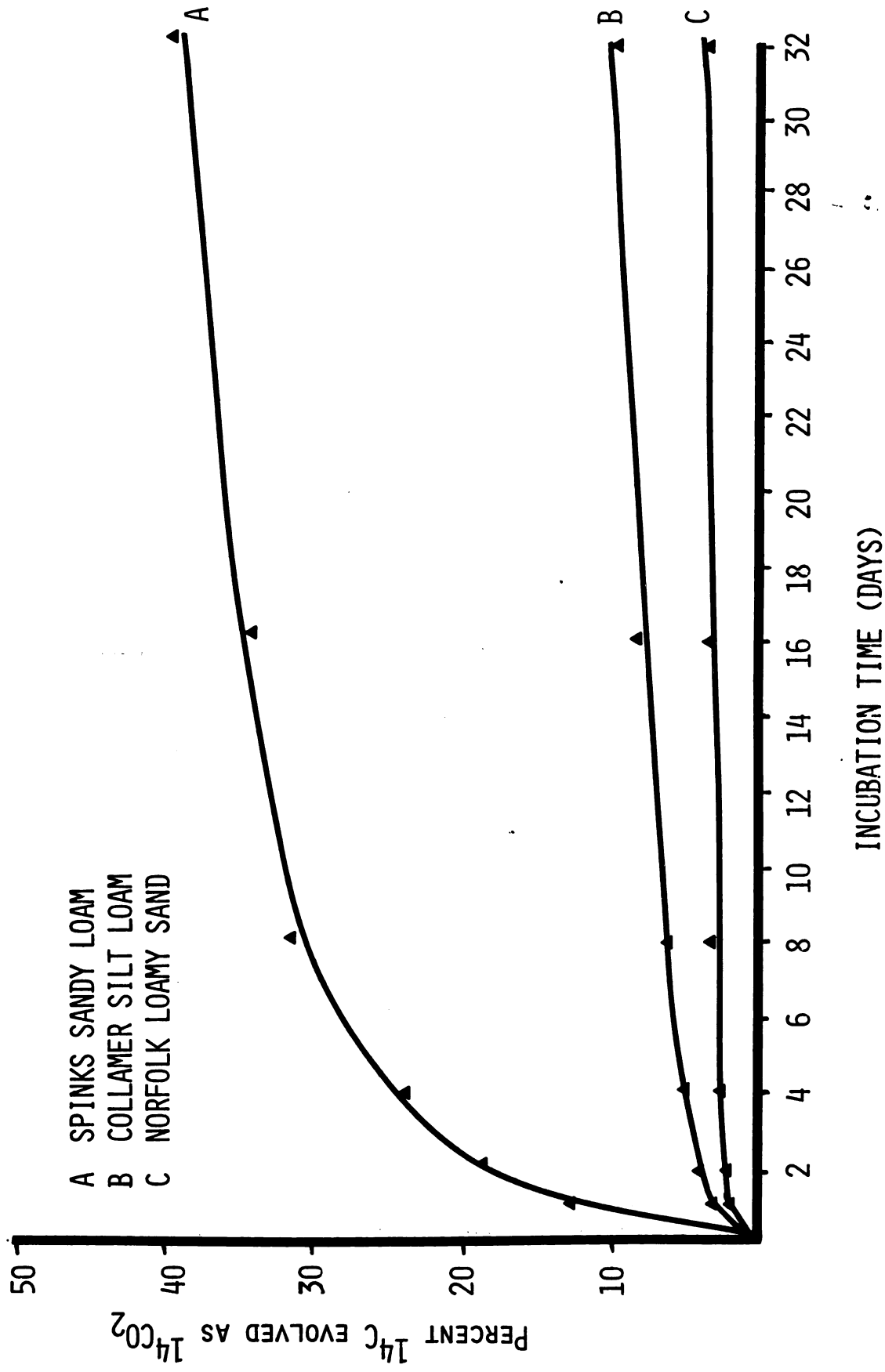

A NON-STERILIZED
B STERILIZED
A
B
50
40
30
20
10
2
4
6
8
10
12
14
16
18
20
22
24
26
28
30
32
PERCENT ^{14}C EVOLVED AS $^{14}CO_2$
INCUBATION TIME (DAYS)

Figure 2. Patterns of ^{14}C-glyphosate degradation in several soils.


A SPINKS SANDY LOAM
B COLLAMER SILT LOAM
C NORFOLK LOAMY SAND
PERCENT ^{14}C EVOLVED AS $^{14}CO_2$
INCUBATION TIME (DAYS)
A
B
C
10
20
30
40
50
2
4
6
8
10
12
14
16
18
20
22
24
26
28
30
32

Addition of phosphate to the Collamer soil at 200 and 400 ppm concentration levels significantly increased glyphosate degradation after 4 or more days (Table 2). After 32 days, increases were 30 and 47 percent for the 200 and 400 ppm concentration level respectively. Phosphate added at 800 ppm concentration level did not increase degradation further. Phosphate additions to the Norfolk soil essentially had no effect on glyphosate degradation (Table 3). It was expected that addition of phosphate would increase glyphosate availability to a limited extent by competing with glyphosate for sorption sites. Glyphosate is believed bound to soil constituents through the phosphonic acid moiety and competes with inorganic phosphate for available sorption sites (1, 10). The correlation coefficient between glyphosate adsorption and unoccupied phosphate sorption sites in nine soils was found to be statistically significant (1). Mobility of glyphosate on soil thin-layer chromatography plates increased when soils with added orthophosphate were used (10). However, in a previous study, effect of phosphate additions on glyphosate degradation was also observed to be dependent on soil type (9). Phosphate additions prior to soil collection significantly increased glyphosate degradation in a Conover sandy loam soil but had no effect on degradation in a Toledo clay loam soil. Since the addition of phosphate allowed only limited increase in microbial conversion of ^{14}C-glyphosate to $^{14}CO_2$ either the glyphosate bound through the phosphonic acid

Table 2. Effect of phosphate addition on degradation of ^{14}C-glyphosate in a Collamer silt loam.

Phosphate (ppm)[a]	Incubation time (days)					
	1	2	4	8	16	32
	(% ^{14}C evolved as $^{14}CO_2$)[b]					
0	2.3 a	2.9 bc	3.8 e	5.2 g	6.9 g	9.1 j
200	2.3 a	3.3 cd	4.7 f	6.8 h	9.2 j	11.8 l
400	2.6 ab	3.7 de	5.2 g	7.5 i	10.1 k	13.4 m
800	2.3 a	3.5 de	5.6 g	7.9 i	10.6 k	13.8 m

[a]Concentration calculated on oven dry soil basis.

[b]Means followed by common letters are not significantly different at the 5% level by Duncan's multiple range test.

Table 3. Effect of phosphate additions on degradation of ^{14}C-glyphosate in a Norfolk loamy sand soil.

Phosphate (ppm)[a]	Incubation time (days)					
	1	2	4	8	16	32
	(% ^{14}C evolved as $^{14}CO_2$)[b]					
0	1.1 a	1.4 b	1.8 c	2.3 d	2.8 e	3.5 f
200	1.1 a	1.4 b	1.8 c	2.3 d	2.8 e	3.6 f
400	1.1 a	1.4 b	1.8 c	2.3 d	2.8 e	3.8 g

[a]Concentration calculated on oven dry soil basis.

[b]Means followed by common letters are not significantly different at the 5% level by Duncan's multiple range test.

moiety is already quite available to the microbial population or there are other means of binding glyphosate in these soils which predominate and render the glyphosate unavailable for microbial degradation to $^{14}CO_2$, or there are inhibitors or insufficient substrate present in these soils.

Glyphosate degradation to CO_2 was drastically reduced after $FeCl_3 \cdot 6H_2O$ was added to the Spinks soil (Table 4). Addition of $FeNH_4(SO_4)_2 \cdot 12H_2O$ reduced glyphosate degradation to a similar extent as did $FeCl_3 \cdot 6H_2O$ additions whereas addition of Fe_2O_3 had no effect (Table 5). Solubilities and immediate reactions involving Fe^{+++} in the soil solution may explain the apparent inactivation of glyphosate.

Known reactions of Fe^{+++} in aqueous solutions (6) indicate that Fe^{+++} reacts in soil with OH^- ions to form colloidal ferric oxide precipitates. Sufficient hydroxyl ions would be present in the Spinks soil with a pH of 6.6 (Table 1) to allow rapid conversion of soluble Fe^{+++} to insoluble precipitates. These colloidal precipitates could then be involved in glyphosate inactivation due to their large surface areas with numerous exposed hydroxyl groups. Adsorption of glyphosate could occur in the modified soil due to replacement of the terminal phosphate group on the herbicide molecule with exposed hydroxyl groups on the colloidal surfaces. Glyphosate could also be tightly bound via a condensation reaction between the hydroxyl species present on the herbicide molecule and a hydroxyl group on the colloidal surface.

Table 4. Effect of ferric chloride additions on degradation of ^{14}C-glyphosate in a Spinks sandy loam soil.

Fe^{+++} added (ppm)[a]	Incubation time (days)					
	1	2	4	8	16	32
	(% ^{14}C evolved as $^{14}CO_2$)[b]					
0	8.5 f	12.6 h	17.9 j	22.7 l	30.0 m	34.4 n
200	3.5 c	5.3 d	8.1 f	10.9 g	16.7 i	21.7 k
400	1.3 a	2.3 b	3.0 bc	3.7 c	5.1 d	6.5 e

[a]Concentration calculated on weight Fe^{+++} per weight oven dry soil basis.

[b]Means followed by common letters are not significantly different at the 5% level by Duncan's multiple range test.

Table 5. Effect of different ferric compounds on degradation of ^{14}C-glyphosate in a Spinks sandy loam soil.

Compound[a]	Incubation time (days)			
	1	2	8	16
	(% ^{14}C evolved as $^{14}CO_2$)[b]			
Control	14.4 d	21.7 f	32.8 h	41.2 i
$FeCl_3 \cdot 6H_2O$	5.9 a	10.2 b	18.3 e	27.7 g
$FeNH_4(SO_4)_2 \cdot 12H_2O$	6.7 a	11.7 bc	20.6 f	29.4 g
Fe_2O_3	12.7 cd	21.4 f	33.0 h	41.2 i

[a]Added as 200 ppm (weight Fe^{+3} per weight oven dry soil).

[b]Means followed by common letters are not significantly different at the 5% level by Duncan's multiple range test.

Fe_2O_3 additions would not result in formation of colloidal precipitates of iron since the solubility constant for Fe_2O_3 is lower than that for the $Fe(OH)_3$ colloidal precipitate (8).

Addition of Al^{+++} also decreased glyphosate degradation in the Spinks soil (Table 6). Colloidal Al precipitates will form in aqueous $AlCl_3$ solutions as pH is adjusted from 3.5 to 7.0. Glyphosate adsorption in Al modified soils could then be similar to that which may occur in Fe modified soils. Ca^{++} and Na^{+} additions had essentially no effect on glyphosate degradation whereas addition of Mn^{++} increased glyphosate degradation. Mn^{++} may be required as a cofactor involved in microbial conversion of glyphosate to CO_2.

Addition of carbonate to increase pH did not significantly increase glyphosate degradation (Table 7). Hydroxyl ions present in pH adjusted soils could react with naturally occurring Fe and Al to form fresh precipitates which then could inactivate glyphosate by adsorption mechanisms previously discussed.

Mixing non-sterilized Spinks soil with sterilized Spinks soil resulted in observed rates of glyphosate degradation greater than the sums of glyphosate degradation rates occurring in each soil alone (estimated rate) calculated on a per gram basis (Table 8). This indicated that the microbial populations of the Spinks soil were capable of growing in new habitats and degrading available glyphosate residues in those habitats. Observed rates of degradation were also

Table 6. Effect of metal additions on degradation of ^{14}C-glyphosate in a Spinks sandy loam soil.

Ionic species[a]	Incubation time (days)			
	1	2	8	16
	(% ^{14}C evolved as $^{14}CO_2$)[b]			
Control	14.4 d	21.7 g	32.8 j	41.2 kl
Fe^{+++}	5.9 a	10.2 b	18.3 e	27.7 h
Al^{+++}	6.4 a	10.6 b	18.3 e	27.2 h
Ca^{++}	12.8 c	20.0 f	31.9 j	40.7 k
Mn^{++}	30.1 i	42.1 l	53.9 m	60.3 n
Na^{+}	13.1 cd	20.9 fg	32.4 j	41.4 kl

[a]Added as chloride salt to equal 200 ppm (weight ionic species per weight oven dry soil).

[b]Means followed by common letters are not significantly different at the 5% level by Duncan's multiple range test.

Table 7. Effect of pH as adjusted by ammonium carbonate additions on degradation of ^{14}C-glyphosate in a Norfolk loamy sand soil.

pH	Incubation time (days)		
	1	2	8
	(% ^{14}C evolved as $^{14}CO_2$)[b]		
4.4[a]	0.6 ab	1.1 cd	1.8 f
5.0	0.6 ab	1.0 cd	1.5 e
6.3	0.7 b	1.2 d	1.8 f
8.9	0.5 a	0.9 c	1.5 e

[a]Original pH.

[b]Means followed by common letters are not significantly different at the 5% level by Duncan's multiple range test.

Table 8. Degradation rates of ^{14}C-glyphosate in various soil mixtures with Spinks sandy loam soil.

Soil mixture			Degradation rate[a]	Incubation time (days)			
Amended soil		Spinks soil		1	2	8	32
	(gm)	(gm)		(% ^{14}C evolved as $^{14}CO_2$)			
Sterilized Spinks	20	5	Observed	9.0	12.7	12.7	12.9
			Estimated	3.2	4.3	7.3	10.7
			Difference	5.8	8.4	5.4	2.2
Collamer	20	5	Observed	5.9	7.4	10.9	15.3
			Estimated	4.2	6.3	11.0	15.2
			Difference	1.7	1.1	- 0.1	0.1
Norfolk	20	5	Observed	4.5	4.6	6.0	7.9
			Estimated	3.7	5.1	8.5	11.1
			Difference	0.8	- 0.5	- 1.5	- 3.2

[a]Estimated rates calculated using experimental data from incubation studies done on each soil component alone (Figures 1, 2), i.e. estimated rate = 20g/25g x amended soil degradation rate + 5g/25g x nonsterilized Spinks soil degradation rate.

higher than estimated degradation rates after incubation periods of 1 and 2 days in the Collamer-Spinks soil mixture but then were essentially equal to each other after 8 and 32 days. The Spinks soil may have inoculated the Collamer soil initially resulting in an increase of degradation of available glyphosate but this stimulatory effect was only temporary. Glyphosate residues after longer incubation periods appeared to become inactivated in the soil and had become unavailable to microorganisms. Observed degradation rates were lower in the Norfolk-Spinks soil mixture than estimated degradation rates for all incubation periods. This indicated that the Norfolk soil was able to adsorb glyphosate in a manner which made it unavailable to the active microbial populations in the Spinks soil inoculum. Perhaps the relatively high aluminum levels in the Norfolk played a role in the biological inactivation of glyphosate in these two soils.

Glucose additions to the Spinks soil failed to increase rates of glyphosate degradation (Table 9). Glucose, a readily available microbial substrate, apparently allowed rapid growth of microorganisms that were incapable of degrading glyphosate. Various organic amendments, in addition to glucose, did not increase glyphosate degradation in the Norfolk loamy sand soil (Table 10). This soil had a relatively low organic matter content thus, conceivably organic amendments would stimulate microbial activity. The presence of sarcosine and glycine but not alfalfa meal in the soil environment inhibited glyphosate degradation after an 8 day incubation period. This

Table 9. Effect of added glucose on degradation of ^{14}C-glyphosate in a Spinks sandy loam soil.

Soil medium	Incubation time (days)			
	1	2	8	32
	(% ^{14}C evolved as $^{14}CO_2$)[b]			
Spinks soil	12.5 b	18.0 c	31.5 e	39.0 f
Spinks soil + glucose[a]	9.6 a	16.3 c	26.8 d	32.3 e

[a]Concentration of glucose in soil equal to 200 ppmw (moist soil basis).

[b]Means followed by common letters are not significantly different at the 5% level by Duncan's multiple range test.

Table 10. Effect of organic amendments on degradation of ^{14}C-glyphosate in a Norfolk loamy sand soil.

Organic Amendment[a]	Incubation time (days)		
	1	2	8
	(% ^{14}C evolved as $^{14}CO_2$)[b]		
Control	0.6 ab	1.1 de	1.8 h
Glucose	0.6 ab	1.0 cd	1.9 h
Sarcosine	0.5 a	0.9 cd	1.4 g
Glycine	0.5 a	0.9 cd	1.3 fg
Alfalfa	0.7 b	1.2 ef	1.9 h

[a]Added as 200 ppmw oven dry soil basis.

[b]Means followed by common letters are not significantly different at the 5% level by Duncan's multiple range test.

was unexpected since both of these compounds are similar in structure to glyphosate.

In summary, microbial degradation rates of glyphosate varied with soil type. Addition of phosphate stimulated glyphosate degradation in only one of two soils examined. Fe^{+++} and Al^{+++} additions to the Spinks soil drastically decreased glyphosate degradation indicating that formation of colloidal Fe and Al precipitates may play a role in glyphosate binding. The presence of additional Ca^{++} and Na^{+} ions had essentially no effect whereas Mn^{++} stimulated degradation. Spinks soil and organic carbon amendments failed to increase glyphosate degradation in soils with low degradation rates.

Literature Cited

1. Hance, R. J. 1976. Adsorption of glyphosate by soils. Pestic. Sci. 7:363-366.

2. Kaufman, D. D. 1964. Microbial degradation of 2,2-dichloropropionic acid in five soils. Can. J. Microbiol. 10:843-852.

3. Kaufman, D. D. 1974. Degradation of pesticides by soil microorganisms. p. 133-202. In W. D. Guenzi (ed.) Pesticides in soil and water. Soil Sci. Soc. Amer., Madison, Wisconsin.

4. Kaufman, D. D., J. R. Plimmer, P. C. Kearney, J. Blake and F. S. Guardia. 1968. Chemical versus microbial decomposition of amitrole in soil. Weed Sci. 16:266-272.

5. Ladlie, J. S., W. F. Meggitt and D. Penner. 1976. Effect of soil pH on microbial degradation, adsorption, and mobility of metribuzin. Weed Sci. 24:477-481.

6. Lindsay, W. L. 1972. Inorganic phase equilibria of micronutrients in soils. p. 43-46. In J. J. Mortvelt (ed.) Micronutrients in agriculture. Soil Sci. Soc. Amer., Madison, Wisconsin.

7. Rueppel, M. L., J. T. Marvel and B. B. Brightwell. 1975. The metabolism and dissipation of N-phosphonomethylglycine in soil and water. Amer. Chem. Soc. Abstract No. 26.

8. Sillen, L. G. and A. E. Martell. 1964. 2nd ed. Stability constants of metal-ion complexes. Spec. Pub. No. 17, The Chemical Society, London.

9. Sprankle, P. L. 1974. Factors affecting the herbicidal activity of N-(phosphonomethyl)glycine (glyphosate) on quackgrass [Agropyron repens (L.) Beauv.] and in the soil. p. 87-89. Ph.D. dissertation, Michigan State University, East Lansing, Michigan.

10. Sprankle, P. L., W. F. Meggitt and D. Penner. 1976. Adsorption, mobility and microbial degradation of glyphosate in soil. Weed Sci. 23:229-234.

11. Tiedje, J. M. and B. B. Mason. 1974. Biodegradation of nitrilo-triacetate (NTA) in soils. Proc. Soil Sci. Soc. Amer. 38:278-283.

12. Weed Science Society of America. 1974. Herbicide Handbook, 3rd ed., p. 201-204. WSSA, Champaign, Illinois.

CHAPTER 5

SUMMARY AND CONCLUSIONS

Laboratory, greenhouse and field studies were initiated to determine the extent glyphosate residues affected turfgrass and alfalfa seedlings grown in several management systems and to determine the extent that various soil amendments affected glyphosate degradation.

Germination of four Kentucky bluegrass, two creeping bentgrass, and two red fescue cultivars was not affected when these turfgrasses were placed in contact with solution of glyphosate in petri dishes. A high concentration of glyphosate (10^{-4}M) did reduce shoot lengths of all three turfgrass species. Germination and shoot length of the three turfgrass cultivars were not affected when exposed to siduron. Soil applications of glyphosate had no effect on seedling growth of the turfgrasses unless glyphosate was applied at very high rates (17.9 kg/ha) and allowed to remain on the soil surface in contact with exposed seeds. Under field conditions, none of the glyphosate treatments reduced turfgrass emergence or growth.

Germination of two alfalfa cultivars, Vernal and Saranac, also was not affected when placed in direct contact with glyphosate solutions in petri dishes. Glyphosate did reduce shoot lengths of Vernal to a greater extent than

Saranac. Soil applications of glyphosate had no effect on percent emergence of either cultivar. Reduction in plant height occurred only when glyphosate was surface applied at 17.9 kg/ha. Glyphosate applied to a Kentucky bluegrass sod at 2.2, 4.5, and 9.0 kg/ha in a alfalfa establishment study utilizing a simulated sod seeding procedure reduced dry weight of both cultivars. Examination of various management systems revealed that a 3 day planting delay or water application after spraying reduced growth inhibition of alfalfa in treated sod.

Degradation studies showed that glyphosate is degraded primarily by microorganisms and at variable rates in different soils. Phosphate additions stimulated degradation to a limited extent in the Collamer silt loam but not in the Norfolk loamy sand. Additions of Fe^{+++} and Al^{+++} to the Spinks soil drastically inhibited degradation, indicating that formation of colloidal Fe and Al precipitates may play a role in glyphosate binding. The presence of additional Ca^{++} and Na^{+} ions had essentially no effect whereas Mn^{++} stimulated degradation. Adding Spinks soil and various carbon substrates as amendments failed to increase glyphosate degradation in soils with low degradation rates.

LIST OF REFERENCES

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1. Abernathy, J. R. and J. M. Davidson. 1971. Effect of calcium chloride on prometryne and fluometuron adsorption in soil. Weed Sci. 19:517-521.

2. Akhavein, A. A. and D. L. Linscott. 1968. The dipyridylium herbicides, paraquat and diquat. Residue Rev. 23: 98-145.

3. Alam, A. V. and S. H. Bishop. 1969. Growth of Esherichia coli on some organophosphonic acids. Can. J. Microbiol. 15:1043-1046.

4. Anderson, M. M. 1976. Growth regulation and metabolic effects of aminomethylphosphonic acid (AMPA) in barley. Plant Phys. Abstracts 57:61.

5. Ashton, F. M. and A. S. Crafts. 1973. Mode of action of herbicides. Wiley-Interscience, New York. 504 p.

6. Baird, D. D., R. P. Upchurch, W. B. Homesley and J. E. Franz. 1971. Introduction of a new broadspectrum postemergence herbicide class with utility for herbaceous perennial weed control. Proc. No. Centr. Weed Contr. Conf. 26:64-68.

7. Bayer, D. E. 1967. Effects of surfactants on leaching of substituted urea herbicides in soil. Weeds 15:249-252.

8. Bovey, R. W. and F. R. Miller. 1969. Effect of activated carbon on the phytotoxicity of herbicides in a tropical soil. Weed Sci. 17:189-192.

9. Bozarth, G. A. and H. H. Funderburk, Jr. 1971. Degradation of fluometuron in sandy loam soil. Weed Sci. 19: 691-695.

10. Brown, J. W. and J. W. Mitchell. 1948. Inactivation of 2,4-D in soil as affected by soil moisture, temperature, manure and autoclaving. Bot. Gaz. 109:314-323.

11. Buchholtz, K. P. 1962. How to get the best of quackgrass. Proc. No. Centr. Weed Contr. Conf. 19:12.

12. Burnside, O. C. 1974. Prevention and detoxification of pesticide residues in soils. p. 387-412. In W. D. Guenzi (ed.) Pesticides in soil and water. Soil Sci. Soc. Am., Madison, Wisconsin.

13. Burnside, O. C., C. R. Fenster and G. A. Wicks. 1971. Soil persistence of repeated annual applications of atrazine. Weed Sci. 19:290-293.

14. Campbell, M. H. 1974. Effects of glyphosate on the germination and establishment of surface-sown pasture species. Aust J. Exp. Agric. Anim. Husb: 14:557-560.

15. Cassaigne, A., A. Lacoste and E. Neuzil. 1976. Recherches sur le catabolisme des aeides phosphoniques: biodegradation de la liaison C-P par Pseudomonas aeruginosa Biochimic Microbienne Serie D:1637-1639.

16. Castelfranco, P. and M. S. Brown. 1962. Purification and properties of the simazine resistance factor of Zea mays. Weeds 10:131-136.

17. Corbin, F. T. and R. P. Upchurch. 1967. Influence of pH on detoxication of herbicides in soil. Weeds 15:370-377.

18. Corbin, F. T., R. P. Upchurch and F. L. Selman. 1971. Influence of pH on the phytotoxicity of herbicides in soil. Weed Sci. 19:233-239.

19. Cullimore, D. R. 1971. Interaction between herbicides and soil microorganisms. Residue Rev. 35:65-80.

20. Dennis, F. G., H. Wilczynski, M. de la Guardia and R. W. Robinson. 1970. Ethylene levels in tomato fruits following treatment with ethrel. Hort Science 5:168-170.

21. DeRose, H. R. 1947. Persistence of growth regulators in the soil. Soil Sci. Soc. Amer. Proc. 12:222-226.

22. Dubey, H. D. and J. F. Freeman. 1967. Influence of soil properties and microbial activity on the phytotoxicity of linuron and diphenamid. Soil Sci. 97:334-340.

23. Edgerton, L. J. and G. D. Blanpied. 1968. Regulation of growth and fruit maturation with 2-chloroethanephosphonic acid. Nature 219:1064-1065.

24. Egley, G. H. and J. E. Dale. 1970. Ethylene, 2-chloroethylphosphonic acid and witchweed germination. Weed Sci. 18:586-589.

25. Foy, C. L. and S. W. Bingham. 1969. Some research approaches toward minimizing herbicidal residues in the environment. Residue Rev. 29:105-135.

26. Freedman, L. B. and G. O. Doak. 1957. The preparation and properties of phosphonic acids. Chem. Rev. 57:502-523.

27. Gray, T. R. G. and S. T. Williams. 1971. Soil micro-organisms. p. 55-65. Hofner Publishing Company, New York.

28. Greenham, O. G. 1953. Auxin activity of ethyl hydrogen 2,4-dichlorophenoxymethylphosphonate in pea epicotyls. Australian J. Sci. 16:66-68.

29. Grover, R. 1966. Influence of organic matter, texture, and available water on the toxicity of simazine in soil. Weeds 14:148-151.

30. Guenzi, W. D. and W. E. Beard. 1968. Anaerobic conversion of DDT to DDD and aerobic stability of DDT in soil. Soil Sci. Soc. Amer. Proc. 32:522-524.

31. Hance, R.J. 1976. Adsorption of glyphosate by soils. Pestic. Sci. 7:363-366.

32. Harkness, D. R. 1966. Bacterial growth on aminoalkyl-phosphonic acids. J. Bacteriol. 92:623-627.

33. Harris, C. I., E. A. Woolson and B. E. Hummer. 1969. Dissipation of herbicides at three soil depths. Weed Sci. 17:27-31.

34. Harrison, G. W., J. B. Weber and J. V. Baird. 1976. Herbicide phytotoxicity as affected by selected properties of North Carolina soils. Weed Sci. 24:120-126.

35. Harvey, R. G. 1973. Influence of cropping and activated carbon on persistence of atrazine in sand. Weed Sci. 21: 204-206.

36. Helling, C. S., P. C. Kearney and M. Alexander. 1971. Behavior of pesticides in soils. Advan. Agron. 23:147-240.

37. Hernandez, T. P. and Warren, G. F. 1950. Some factors affecting the rate of inactivation and leaching of 2,4-D in different soils. Proc. Amer. Soc. Hort. Sci. 56: 287-293.

38. Hiltbold, A. E. and G. A. Buchanan. 1969. Use of garbage compost in reduction of toxic residues of herbicides in soil. Proc. Ass. S. Agr. Workers 66:102.

39. Horiguchi, J. S. and M. Kandatsu. 1959. Isolation of 2-aminoethanephosphonic acid from rumen protozoa. Nature 184:901-902.

40. Horiguchi, J. S. and M. Kandatsu. 1960. Ciliatine: A new aminophosphonic acid contained in rumen ciliate protozoa. Bull. Agri. Chem. Soc. Japan 24:565-570.

41. Juo, A. S. R. and B. G. Ellis. 1968. Chemical and physical properties of iron and sluminum phosphate and their relation to phosphorous availability. Soil Sci. Soc. Amer. Proc. 32:216-221.

42. Kaufman, D. D. 1966. An inexpensive, positive pressure, soil perfusion system. Weeds 14:90-91.

43. Kaufman, D. D. 1974. Degradation of pesticides by soil microorganisms. p. 133-202. In W. D. Guenzi (ed.) Pesticides in soil and water. Soil Sci. Soc. Amer. Madison, Wisconsin.

44. Kaufman, D. D., J. R. Plimmer, P. C. Kearney, J. Blake and F. S. Guardia. 1968. Chemical versus microbial decomposition of amitrole in soil. Weed Sci. 16:266-272.

45. Kearney, P. C., E. A. Woolson, J. R. Plimmer and A. R. Isensee. 1969. Decontamination of pesticides in soils. Residue Rev. 29:137-149.

46. Kennedy, M. V., B. J. Stojanovic and F. L. Shuman, Jr. 1972. Chemican and thermal aspects of pesticide disposal. J. Environ. Quality 1:63-65.

47. Kittredge, J. S. and R. R. Hughes. 1964. The occurrence of -amino-B-phosphonopropionic acid in the zoanthid, Zoanthus sociatus, and the ciliate, Tetrahymena pyriformis. Biochemistry 3:991-996.

48. Kittredge, J. S., E. Roberts and D. G. Simonsen. 1962. The occurrence of free 2-aminoethylphosphonic acid in the sea anemone, Anthopleura elegantissima. Biochemistry 1:624-628.

49. Klingman, D. L. and J. J. Murray. 1976. Germination of seeds of turfgrass as affected by glyphosate and paraquat. Weed Sci. 24:191-193.

50. Klingman, G. C. and F. M. Ashton. 1975. Weed science: principles and practices. Wiley-Interscience, New York. 431 p.

51. Koren, E., C. L. Foy, and F. M. Ashton. 1968. Phyto-toxicity and persistence of four thiocarbamates in five soil types. Weed Sci. 16:172-175.

52. Kries, O. H. 1947. Persistence of 2,4-dichlorophenoxyacetic acid in relation to content of water, organic matter and lime. Bot. Gaz. 108:510-525.

53. Ladlie, J. S., W. F. Meggitt and D. Penner. 1976. Effect of soil pH on microbial degradation, adsorption and mobility of metribuzin. Weed Sci. 24:477-481.

54. Ladlie, J. S., W. F. Meggitt and D. Penner. 1976. Effect of pH on metribuzin activity in the soil. Weed Sci. 24:505-507.

55. Ladlie, J. S., W. F. Meggitt and D. Penner. 1976. Role of pH on metribuzin dissipation in field soils. Weed Sci. 24:508-511.

56. Lambert, S. M., P. E. Porter and R. H. Schieferstein. 1965. Movement and sorption of chemicals applied to the soil. Weeds 13:185-190.

57. LaNauze, J. M., H. Rosenberg and D. C. Shaw. 1970. The enzymatic cleavage of the carbon-phosphorous bond: Purification and properties of phosphonatase. Biochim. Biophys. Acta 212:332-350.

58. Lange, A. H. 1967. Problems of sod contamination, accumulation of residues, and the effect on subsequent croppings. Proc. West Weed Contr. Conf. 27:30.

59. Marth, P. C. and J. W. Mitchell. 1944. 2,4-dichlorophenoxyacetic acid as a differential herbicide. Botan. Gaz. 106:224.

60. Maynard, J. A. and J. W. Swan. 1963. Organo-phosphorous compounds. I. 2-chloroalkylphosphonic acid as phosphorylating agents. Australian J. Chem. 16:596-608.

61. Meggitt, W. F. 1960. Herbicide combinations for quackgrass control. Proc. No. Centr. Weed Contr. Conf. 17:83.

62. Messersmith, C. G., O. C. Burnside and T. L. Lavy. 1971. Biological and non-biological dissipation of trifluralin from soil. Weed Sci. 19:285-290.

63. Mayer, J. R., R. J. Hance and C. E. McKone. 1972. The effect of adsorbents on the rate of degradation of herbicides incubated with soil. Soil Biol. Biochem. 4:307-311.

64. Murray, D. S., W. L. Rieck, and J. Q. Lynd. 1969. Microbial degradation of five substituted urea herbicides. Weed Sci. 17:52-55.

65. Obrien, S. R. and R. E. Green. 1969. Degradation of atrazine in four Hawaiian soils. Weed Sci. 17:509-514.

66. Ogle, R. E. and G. F. Warren. 1954. Fate and activity of herbicides in soils. Weeds 3:257-273.

67. Randall, D. I. 1968. 2-haloethanephosphonic acids as ethylene releasing agents for the induction of flowering in pineapples. Nature 218:974-975.

68. Rodgers, E. G. 1962. Leaching of four triazines in three soils as influenced by varying frequencies and rates of simulated rainfall. Proc. SWC 15:268.

69. Roeth, F. W., T. L. Lavy and O. C. Burnside. 1969. Atrazine degradation in two soil profiles. Weed Sci. 17:202-205.

70. Rosenberg, H. and J. M. LaNauze. 1967. The metabolism of phosphonates by microorganisms. The transport of aminoethylphosphonic acid in Bacillus cereus. Biochim. Biophys. Acta 141:79-90.

71. Rueppel, M. L., J. T. Marvel and B. B. Brightwell. 1975. The metabolism and dissipation of N-phosphonomethyl-glycine in soil and water. Amer. Chem. Soc. Abstract No. 24.

72. Rueppel, M. L., J. T. Marvel and L. A. Suba. 1975. The metabolism of N-phosphonomethylglycine in corn, cotton, soybeans and wheat. Amer. Chem. Soc. Abstract No. 26.

73. Ryzhkov, V. L., M. I. Kabachnik, L. M. Tarasevich, T. Y. Medved, N. A. Zeitlech, N. K. Marchenko, V. A. Yagzhanova, E. F. Ulanova, and N. V. Cheburetch. 1955. Toxicity of aminoalkylphosphonic acids in animal and plant tissue. Chem. Abstracts 49:3404.

74. Savage, K. E. 1973. Adsorption and degradation of chlorbromuron in soil. Weed Sci. 21:416-420.

75. Savage, K. E. and R. D. Wauchope. 1974. Fluometuron adsorption-desorption equilibria in soil. Weed Sci. 22:106-110.

76. Schimizu, H., Y. Kakimoto, T. Nakajuma, A. Kanazawa and I. Sano. 1965. Isolation and identification of 2-amino-ethylphosphonic acid from bovine brain. Nature 207:1197-1198.

77. Sheets, T. J. 1958. The comparative toxities of four phenyl urea herbicides in several soil types. Weeds 6: 413-425.

78. Smith, D. T. and W. F. Meggitt. 1970. Persistence and degradation of pyrazon in soil. Weed Sci. 18:260-264.

79. Sprankle, P. L. 1974. Factors affecting the herbicidal activity of N-(phosphonomethyl) glycine (glyphosate) on quackgrass [Agropyron repens (L.) Beauv.] and in the soil. Ph.D. thesis, Michigan State University, East Lansing, Michigan.

80. Sprankle, P. L., W. F. Meggitt and D. Penner. 1975. Rapid inactivation of glyphosate in the soil. Weed Sci. 23:224-228.

81. Sprankle, P. L., W. F. Meggitt and D. Penner. 1975. Adsorption, mobility and microbial degradation of glyphosate in the soil. Weed Sci. 23:229-234.

82. Thiegs, B. J. 1955. The stability of dalapon in soils. Down to Earth 11:2.

83. Upchurch, R. P. and D. D. Mason. 1962. The influence of soil organic matter on the phytotoxicity of herbicides. Weeds 10:9-14.

84. Upchurch, R. P. and W. C. Pierce. 1957. The leaching of monuron from lakeland sand soil. Part I. The effect of amount, intensity, and frequency of simulated rainfall. Weeds 6:321-330.

85. Warner, H. L. and A. C. Leopold. 1969. Ethylene evolution from 2-chloroethylphosphonic acid. Plant Physiol. 44:156-158.

86. Weber, J. B. and H. D. Coble. 1968. Microbial decomposition of diquat adsorbed on montmorillonite and kaolinite clays. J. Agr. Food Chem. 16:475-478.

87. Weber, J. B., P. W. Perry and K. Ibaraki. 1968. Effect of pH on the phytotoxicity of prometryne applied to synthetic soil media. Weed Sci. 16:134-136.

88. Weber, J. B. and S. B. Weed. 1974. Effects of soil on the biological activity of pesticides. p. 223-256. In W. D. Guenzi (ed.) Pesticides in Soil and Water. Soil Sci. Soc. Amer. Madison, Wisconsin.

89. Weber, S. B., S. B. Weed and T. W. Waldrep. 1974. Effect of soil constituents on herbicide activity in modified-soil field plots. Weed Sci. 22:454-459.

90. Weed Science Society of America. 1974. Herbicide handbook. 3rd ed. WSSA, Champaign, Illinois. 430 p.

91. Yang, S. F. 1969. Ethylene evolution from 2-chloroethylphosphonic acid. Plant Physiol. 44:1203-1204.

92. Zeleznick, L. D., T. C. Myers, E. B. Titchener. 1963. Growth of Escherichia coli on methyl- and ethylphosphonic acids. Biochim. Biophys. Acta. 78:546-547.