

AN ABSTRACT OF THE THESIS OF

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Title: DIFFERENTIAL SENSITIVITY OF TWO COMMON
GROUNDSEL BIOTYPES (Senecio vulgaris L.) TO
SEVERAL s-TRIAZINE HERBICIDES

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Dr. Arnold P. Appleby /

Studies were initiated to determine the response of two common groundsel biotypes (Senecio vulgaris L.) to several s-triazine herbicides. Herbicides tested were: 2-chloro-4,6-bis(ethylamino)-s-triazine (simazine), 2-chloro-4-(ethylamino)-6-(isopropylamino)-s-triazine (atrazine) 2-(sec-butylamino)-4-(ethylamino)-6-methoxy-s-triazine (GS-14254), 2,4-bis(isopropylamino)-6-methoxy-s-triazine (prometone), 2-(tert-butylamino)-4-(ethylamino)-6-methylthio-s-triazine (terbutryn), and 2,4-bis(isopropylamino)-6-methylthio-s-triazine (prometryne). One biotype was much more susceptible than the other. Sensitive plants were effectively controlled by 0.5 ppm of atrazine and simazine, 1 ppm of GS-14254 and prometone, and 4 ppm of prometryne. The resistant biotype failed to show any symptoms of photosynthesis inhibition at the highest

rates tested, i. e. 4 ppm of simazine and 30 ppm for atrazine, GS-14254, prometone, and prometryne. Both biotypes were resistant to terbutryn at 30 ppm.

When a triazine herbicide was applied, the susceptible plants became chlorotic and died; resistant plants never exhibited these symptoms. Photosynthesis was completely inhibited by simazine in susceptible (S) plants but resistant (R) plants were unaffected. Photosynthesis in the susceptible biotype resumed when the herbicide was removed after 24 hours.

Absorption and metabolism of simazine were explored as possible explanations for the herbicide tolerance exhibited by the R biotype. Both biotypes absorbed the herbicide equally well, and no differences in simazine metabolism were found which could explain the mechanism of resistance. Plants of both biotypes were subjected to ^{14}C -simazine for up to 96 hours. The greatest concentration of ^{14}C activity (80 to 90%) was located in the chloroform-soluble fraction of the foliage tissue of each biotype. The ^{14}C in this fraction of the plant extracts was determined by thin-layer chromatography to be similar to ^{14}C -simazine. Small amounts of ^{14}C activity (10-15%) were isolated in the water-soluble fraction of the plant extracts, but time-course studies revealed no differential increase in water-soluble simazine metabolites by either biotype. A similar metabolism study using corn was conducted, which substantiated the

findings of numerous workers. Several alternative explanations for the difference in triazine sensitivity between the two common groundsel biotypes are suggested.

Differential Sensitivity of Two Common Groundsel
Biotypes (Senecio vulgaris L.) to Several
s-Triazine Herbicides

by

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For her love, patience, and support during the years of study, I dedicate this manuscript to Lynn, my wife.

To our children, Steve and Susan, who endured partial loss of a father while this manuscript was prepared, I also dedicate this thesis.

TABLE OF CONTENTS

<u>Section</u>	<u>Page</u>
I THE RESISTANCE OF ORGANISMS TO PESTICIDES-- A REVIEW OF THE PROBLEM	1
II THE EFFECT OF CERTAIN <u>s</u> -TRIAZINE HERBICIDES ON TWO BIOTYPES OF COMMON GROUNDSEL	5
Introduction	5
Materials and Methods	6
Experiment 1: A Determination of Tolerance to Atrazine of Common Groundsel Previously Reported to be Triazine Resistant	6
Experiment 2: A Determination of Tolerance to Atrazine of Common Groundsel Found in the Willamette Valley	7
Experiments 3-8: The Dose Response of Two Common Groundsel Biotypes to Several <u>s</u> -Triazine Herbicides	7
Plant Material	8
Herbicides	10
Experiment 9: The Dose Response of Corn to Atrazine	11
Results	11
Experiment 1: A Determination of Tolerance to Atrazine of Common Groundsel Previously Reported to be Triazine Resistant	11
Experiment 2: A Determination of Tolerance to Atrazine of Common Groundsel in the Willamette Valley	11
Experiments 3-8: The Dose Response of Two Common Groundsel Biotypes to Several <u>s</u> -Triazine Herbicides	13
Experiment 9: The Dose Response of Corn to Atrazine	20
Discussion	20
III PHOTOSYNTHESIS, ABSORPTION, AND METABOLISM STUDIES OF TWO COMMON GROUNDSEL BIOTYPES TREATED WITH SIMAZINE	24
Introduction	24
Materials and Methods	29

<u>Section</u>		<u>Page</u>
III	Plant Material	29
	Experiment 10: The Photosynthetic Response of Two Common Groundsel Biotypes to Simazine	30
	Experiment 11: The Absorption of Simazine by Two Biotypes of Common Groundsel as a Function of Time	31
	Experiment 12: The Metabolism of Simazine by Two Biotypes of Common Groundsel as a Function of Time	34
	Extraction and Counting Procedure	35
	Experiment 13: The Metabolism of Simazine by Corn (<u>Zea mays</u> L.)	37
	Results	38
	Experiment 10: The Photosynthetic Response of Two Common Groundsel Biotypes to Simazine	38
	Experiment 11: The Absorption of Simazine by Two Biotypes of Common Groundsel as a Function of Time	41
	Experiment 12: The Metabolism of Simazine by Two Biotypes of Common Groundsel as a Function of Time	48
	Experiment 13: The Metabolism of Simazine by Corn	51
	Discussion	55
IV	SUMMARY AND CONCLUSIONS	57
	BIBLIOGRAPHY	60
	APPENDIX	65

LIST OF FIGURES

<u>Figure</u>		<u>Page</u>
1.	The Dose Response of Two Common Groundsel Biotypes to Simazine.	14
2.	The Dose Response of Two Common Groundsel Biotypes to Atrazine.	15
3.	The Dose Response of Two Common Groundsel Biotypes to Terbutryn.	16
4.	The Dose Response of Two Common Groundsel Biotypes to GS-14254.	17
5.	The Dose Response of Two Common Groundsel Biotypes to Prometryne.	18
6.	The Dose Response of Two Common Groundsel Biotypes to Prometone.	19
7.	The Dose Response of Corn to Atrazine.	21
8.	Metabolic Scheme for Atrazine Metabolism in Higher Plants.	26
9.	The Photosynthetic Response of Two Common Groundsel Biotypes to Simazine. Treatment Date 1.	34
10.	The Photosynthetic Response of Two Common Groundsel Biotypes to Simazine. Treatment Date 2.	40
11.	Total Simazine Absorbed by Two Common Groundsel Biotypes as a Function of Time.	42
12.	Simazine Absorbed per Gram Fresh Weight by Two Common Groundsel Biotypes as a Function of Time.	43
13.	Simazine Absorption per Gram Dry Weight of Two Common Groundsel Biotypes as a Function of Time.	44

<u>Figure</u>		<u>Page</u>
14.	Absorption of Simazine by Two Common Groundsel Biotypes as a Function of Transpiration.	45
15.	Radioautograph of Thin-layer Chromatogram.	52

LIST OF TABLES

<u>Table</u>	<u>Page</u>
1. Chemical Structure and Water Solubility of Six <u>s</u> -triazine Herbicides used in Experiments 3 through 8.	9
2. The Dose Response to Atrazine of Common Groundsel from a Willamette Valley Seed Source and a Seed Source Previously Reported to be Atrazine Resistant.	12
3. The Uptake of Simazine and Water by Two Biotypes of Common Groundsel as a Function of Time. Treatment Date 1.	46
4. The Uptake of Simazine and Water by Two Biotypes of Common Groundsel as a Function of Time. Treatment Date 2.	47
5. The Distribution of ^{14}C Activity from Applied ^{14}C -simazine in Extracts of Foliage and Roots of Two Common Groundsel Biotypes as a Function of Time. Study A.	50
6. The Distribution of ^{14}C Activity from Applied ^{14}C -simazine in Extracts of Foliage and Roots of Two Common Groundsel Biotypes as a Function of Time. Study B.	50
7. Rf Values for ^{14}C -simazine and ^{14}C Activity Extracted from Two Common Groundsel Biotypes Previously Treated with ^{14}C -simazine.	53
8. The Distribution of ^{14}C Activity in Foliage and Roots of Corn After Treatment with ^{14}C -simazine for 72 Hours.	53

DIFFERENTIAL SENSITIVITY OF TWO COMMON GROUNDSEL
BIOTYPES (Senecio vulgaris L.) TO SEVERAL
s-TRIAZINE HERBICIDES

I. THE RESISTANCE OF ORGANISMS TO PESTICIDES--
A REVIEW OF THE PROBLEM

Many entomologists consider resistance of insects to insecticides to be one of their greatest problems. Although this phenomenon was encountered before the modern era of pest control, it has only become a source of worry since the introduction of DDT and other synthetic organic pesticides. Within two years after DDT was available for testing, a strain of housefly resistant to it had been developed and by the late 1940's the cause of housefly resistance was known. The degradation of DDT to nontoxic DDE was more rapid in resistant insects than susceptible ones (Hoskins and Perry, 1950).

Weed scientists also are concerned about resistance but reports of plants becoming resistant to herbicides occur rarely in weed science literature. However, there are many reports of plants which are tolerant to a particular herbicide, since this is the basis for selective weed control. It seems of benefit, therefore, when considering the problem of herbicide resistance by plants to review some earlier entomological studies concerning insecticide resistance.

There are numerous examples of insect and mite species which can no longer be controlled by formerly useful chemicals. Brown

(1963 and 1964) lists 171 insect species which are resistant to at least one insecticide. There are several good reviews on the subject of insecticide resistance (Brown, 1948; Brown, 1963 and 1964; Crow, 1957; Hoskins and Gordon, 1956; and Terriere, 1966) as well as many technical articles dealing with specific examples of resistance.

Pesticide resistance appears to be under genetic control. There is little doubt that the ability to resist a poison is transmittable from generation to generation. Crow (1957) has suggested that the genetics of resistance be explained by either postadaptation or preadaptation. Postadaptation is a genetic change directly induced by the chemical, while preadaptation is the genetic difference in resistance already present within the population. In the latter case, the chemical or poison acts as a selection agent favoring the resistant genotype. Crow (1957) further indicates that little evidence is available to suggest postadaptation but explores in length the preadaptive mechanism for resistance in both insects and bacteria. He concludes by stating that the preadaptive hypothesis is well established and the sole effect of the insecticide is as a selective agent in the population (Crow, 1957).

Hoskins and Gordon (1956) suggest that mechanisms of insecticide resistance are primarily biochemical. However, they also indicate several other mechanisms by which an insect can resist a chemical. These include behavior patterns, cuticle thickness,

and cuticle permeability. Resistance arising from such defenses was classified as vigor tolerance.

Insecticides exert their lethal action by interfering with some function essential to life, and for resistance to occur the action of the insecticide must be countered (Hoskins and Gordon, 1956). This could happen in two ways: by addition of a protective mechanism which inhibits the lethal interaction of the insecticide with the vital function, or by replacing the essential life function with an "insensitive mechanism" not affected by the chemical. Terriere (1966) suggests three possible mechanisms of biochemical resistance. The insect could detoxify the chemical, thus preventing it from reaching the site of action. The sensitive site may be protected from the chemical, or the insect may have developed a new or alternate biochemical process not affected by the toxicant.

As indicated earlier, reports of plants which have become resistant to herbicides occur sparingly. Several reasons have been suggested for this apparent lack of herbicide resistance in plants. These are: a large source of new and old seed, seed dormancy (dispersal through time), a single reproductive cycle each year, vegetative propagation, escapes or large nontreated areas (tends to keep the gene pool infiltrated with susceptible genes), and the use of herbicide rotations or alternate methods of weed control. However, there is little reason to assume that mechanisms which allow

pesticide resistance to develop in other organisms do not act in a similar manner in plants. Recently, several reports have indicated that variation in sensitivity to herbicides does occur with certain plant species. These articles are further reviewed in the following sections.

II. THE EFFECT OF CERTAIN S-TRIAZINE HERBICIDES ON TWO BIOTYPES OF COMMON GROUNDSEL

Introduction

The continued use of a herbicide has been known to raise the level of tolerance within weed populations. This incidence of incomplete weed control is often observed as a shift to more tolerant plant species; for example, the increase of grass weeds and the decrease of many dicotyledonous weeds from repeated applications of 2, 4-D. Recently, additional information concerning herbicide variation within a plant species has been found. Ryan (1970) reported failure of atrazine and simazine to control common groundsel from a location where these herbicides had been in continued use. Plants of the same species which had not been exposed to continuous triazine pressure were effectively controlled. Plants from both locations were susceptible to other non-triazine herbicides. Data presented by Schooler, Bell, and Nalewaja (1972) indicate a genetic basis governing tolerance of foxtail barley (Hordeum jubatum L.) to siduron [1-(2-methylcyclohexyl)-3-phenylurea]. They concluded that under proper selection pressure, a siduron-tolerant foxtail barley could be developed. Similarly, Wiemer (1960) observed variability in tolerance to TCA (trichloroacetic acid) and dalapon (2, 2-dichloro propionic acid) by 20 strains of bermudagrass

[Cynodon dactylon (L.) Pers.].

These reports suggest that increased herbicide tolerance within a weed species may result from repeated applications and indicate a need for herbicide rotations when possible.

This study was initiated to observe the response to several triazine herbicides of common groundsel previously reported resistant by Ryan (1970). The response to several triazine herbicides of the same weed species from a location of non-triazine use also was studied.

Materials and Methods

Experiment 1: A Determination of Tolerance to Atrazine of Common Groundsel Previously Reported to be Triazine Resistant

Two hundred seeds were selected and pregerminated from the entire supply received from Ryan (1970). Seeds were germinated on moist filter paper in petri dishes and maintained at 25° C, 16 hr daylength, and light intensity of 8600 lux. After 10 days, seedlings were transplanted into 3 by 3 cm plastic pots containing moist 1:1 v/v sand and peat mix and allowed to grow for 20 days under greenhouse conditions. Seventy-five plants were then selected and subjected to several rates of atrazine. Atrazine treatments ranged from 0 to 8.96 kg/ha. The herbicide was applied as a uniform spray to both foliage

and to the surface of the sand-peat mix.

All treated plants were allowed to mature and seed was harvested from those plants surviving the highest atrazine rate for use in subsequent studies. The experiment was terminated 45 days after the herbicide application. Foliage dry weights were determined and differences were statistically compared.

Experiment 2: A Determination of Tolerance to
Atrazine of Common Groundsel Found in the
Willamette Valley

Seeds were collected at random from plants growing in several groundsel-infested fields in the Willamette Valley. The entire supply was bulked and 50 seeds were selected for germination and transplanting as described earlier (Experiment 1). Twenty-five uniform plants were then subjected to several rates of atrazine as described in Experiment 1. The experiment was terminated 45 days after herbicide application. Foliage dry weights were determined and differences were compared statistically.

Experiments 3-8: The Dose Response of
Two Common Groundsel Biotypes to
Several s-Triazine Herbicides

This study consisted of six similar experiments, each conducted using a different triazine herbicide. A list of herbicides used and the corresponding experiment number is presented in

Table 1. Each experiment was conducted twice with four replications each time. In experiments 3 and 4, resistant and susceptible common groundsel biotypes were subjected to differing rates of herbicide. For this reason, they were maintained in separate but adjacent experimental areas and conducted simultaneously under identical conditions.

Plant Material

Seeds of the susceptible and resistant sources were germinated under conditions previously described in Experiment 1. After 3 to 7 days, the seedlings were transferred to black polyethylene containers having 225 ml of Hoagland's solution. Experiments 3 through 6 were conducted using 0.5-strength Hoagland's solution the first time; thereafter 0.1-strength Hoagland's solution was used. Each seedling was placed between the halves of a split cork and fitted into a hole in the container lid with roots extending into the nutrient solution. Each pot containing three plants was maintained in a 21 °C water bath and was continually aerated. (The first series of experiments 3 through 6 were conducted without the water bath.) When 30 to 32 days old, plants were selected for uniformity and subjected to several rates of herbicide. The herbicide was introduced directly to the roots by removing the original solution and replacing it with that containing the appropriate herbicide concentration.

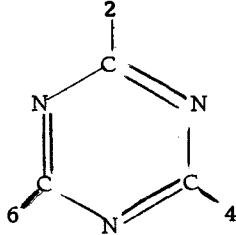
All experiments were terminated when complete necrosis was observed on plants of the susceptible biotype receiving the highest rate of herbicide. Plants treated with lower rates often were completely necrotic at the same time. For this reason, the lowest rate which resulted in 100% necrosis (L. D. 100) was visually determined. Dry weights of the entire plants were determined and statistically analyzed.

Herbicides

The chemical structure and water solubility of the six triazine herbicides used are presented below. Each technical herbicide was dissolved in methanol to a high concentration. The herbicidal rates were made by diluting an appropriate amount of the methanol-herbicide solution with 0.1-strength Hoagland's solution. The rates of herbicide used ranged from 0 to 30 ppm. (Conversions from ppm to molar concentration are given in Appendix Table 1 for each herbicide tested.)

A preliminary study was conducted (Appendix Table 2) to determine the phytotoxic concentration of methanol to both common groundsel biotypes. The methanol concentration was determined to be toxic only for experiment 3 and was removed from the treating solution under vacuum. In each experiment, a treatment having only nutrient solution plus methanol was included.

Table 1. Chemical structure and water solubility of the six s-triazine herbicides used in experiments 3 through 8.

Experiment Number	Common Name or Designation	Basic Ring	Groups on Ring			Water Solubility at 20° C (ppm)
			2	4	6	
3	simazine		Cl	NHC ₂ H ₅	NHC ₂ H ₅	5
4	atrazine		Cl	NHC ₂ H ₅	NH-i-C ₃ H ₇	33
5	terbutryn		SCH ₃	NHC ₂ H ₅	NH-t-C ₄ H ₉	58
6	GS-14254		OCH ₃	NHC ₂ H ₅	NH-s-C ₄ H ₉	620
7	prometryne		SCH ₃	NH-i-C ₃ H ₇	NH-i-C ₃ H ₇	48
8	prometone		OCH ₃	NH-i-C ₃ H ₇	NH-i-C ₃ H ₇	760

^a Adapted from the Herbicide Handbook of The Weed Science Society of America

Experiment 9: Dose Response of Corn
(*Zea mays* L.) to Atrazine

Corn seeds, variety KE 435 (Northrup King), were germinated in moist paper under conditions described in experiment 1. After 3 days, seedlings were transferred to black polyethylene containers having 225 ml of 0.25-strength Hoagland's solution. Each germinated seed was wrapped in cotton and placed into a hole in the container lid. Each pot contained three plants, was maintained in a 21 °C water bath, and was continually aerated. When 7 days old, plants were selected for uniformity and subjected to several rates of atrazine, ranging from 0 to 30 ppm. The herbicide was introduced directly to the roots as previously described. The experiment was terminated 10 days after the herbicide application. Dry weights of the entire plant were determined and statistically analyzed. Visual evaluations of control were also made. The experiment was conducted twice with three replications each time.

Results

Experiments 1 and 2

Foliage dry weights of plants from each common groundsel seed collection are presented in Table 2 and Appendix Tables 4 and 6.

Table 2. The dose response to atrazine of common groundsel from a Willamette Valley seed source and a seed source previously reported to be atrazine resistant. Dry weights determined 45 days after atrazine application.

atrazine rate	Average Foliage Dry Weight From:	
	Ryan ^a	Willamette Valley ^b
(kg/ha)	(mg/plant)	(mg/plant)
0	347	440
1.12	366	14
2.24	368	14
4.48	351	15
8.96	342	23

^aTriazines in continued use since 1958. (15 plants/treatment)

^bTriazines not in continued use (5 plants/treatment)

Plants from the Willamette Valley seed source were effectively controlled by atrazine at the lowest rate applied. The plants obtained from seed provided by Ryan were not affected even at the highest rate of atrazine. These data indicate that large differences in tolerance to atrazine by common groundsel exist and substantiate the terms "susceptible" and "resistant." In all future studies, the plants arising from the progeny of the Willamette Valley seed will be referred to as susceptible (S). Plants arising from progeny of the seed provided by Ryan will be termed resistant (R). The term "biotype" will be used during these studies to denote those members of the common groundsel population which exhibit a similar response to triazine herbicides.

Experiments 3-8: The Dose Response
to Two Common Groundsel Biotypes to
Several s-Triazine Herbicides

The dose response of both common groundsel biotypes to six s-triazine herbicides are presented in Figures 1 through 6 and Appendix Tables 8 through 42. The values depicted in all figures are averages of eight replications unless otherwise stated. Plants of both biotypes were pregerminated, grown in aqueous nutrient culture, and subjected to several rates of herbicide. Experiments conducted in this manner exclude seed depth in the soil, germination characteristics, differential growth rates, and some morphological differences as factors affecting herbicide tolerance.

Plants of the susceptible (S) biotype were effectively controlled by atrazine and simazine concentrations as low as 0.5 ppm (2.31×10^{-6} M and 2.48×10^{-6} M, respectively). However, simazine concentrations as high as 4 ppm (1.98×10^{-5} M) and atrazine concentrations up to 30 ppm (1.39×10^{-4} M) failed to adequately control the resistant biotype. Terbutryn had no phytotoxic effect upon the resistant biotype and prometryne significantly increased its growth at all levels tested. The susceptible biotype also was quite tolerant of terbutryn and required 4 ppm (1.66×10^{-5} M) of prometryne to produce the characteristic phytotoxic symptoms. One ppm (4.4×10^{-6} M) of either GS-14254 or prometone effectively inhibited

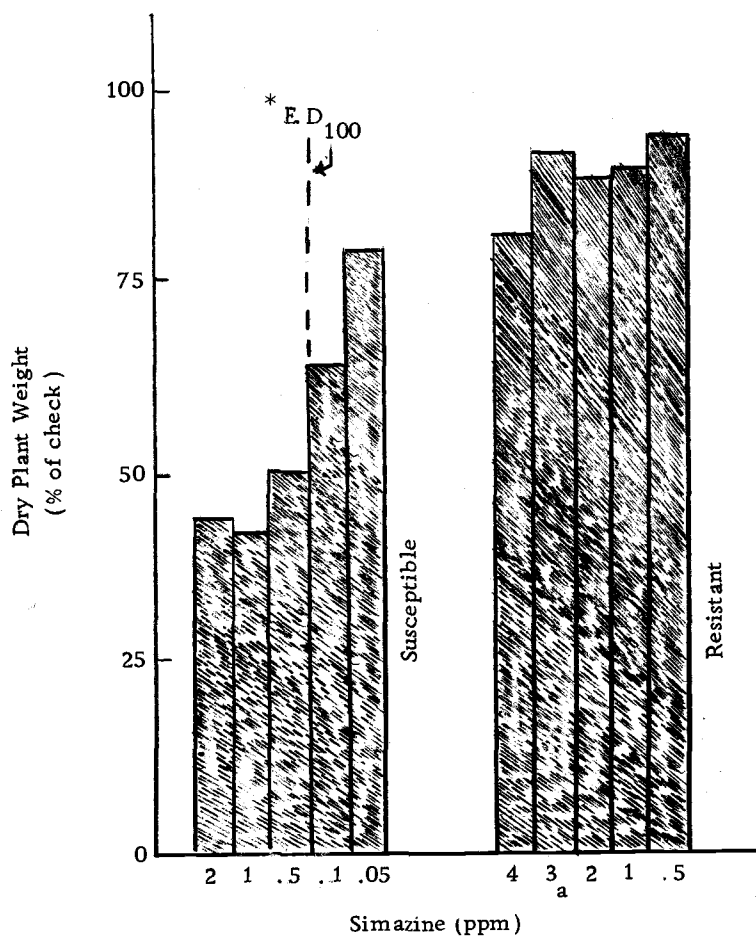


Figure 1. Experiment 3. The dose response of two common groundsel biotypes to simazine. Plants treated 30-32 days after germination.

*Effective dose 100%: values to left of line determined by visual evaluation equal to complete control.

^a Average of 4 replications.

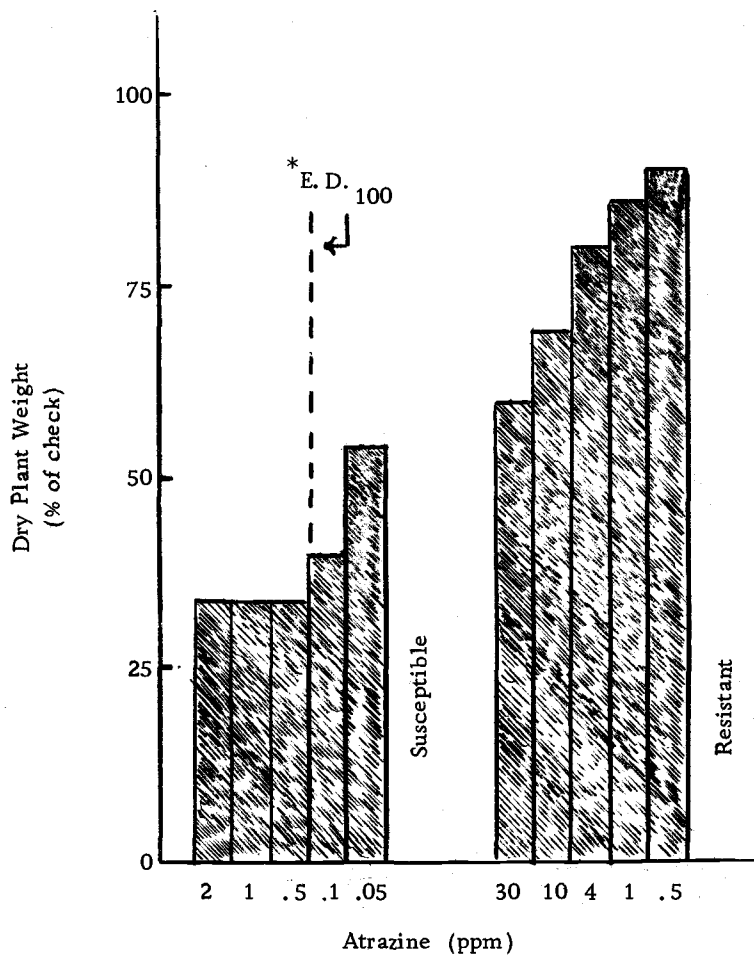


Figure 2. Experiment 4. The dose response of two common groundsel biotypes to atrazine. Plants treated 30-32 days after germination.

* Effective dose 100: values to left of line determined by visual evaluation equal to complete control.

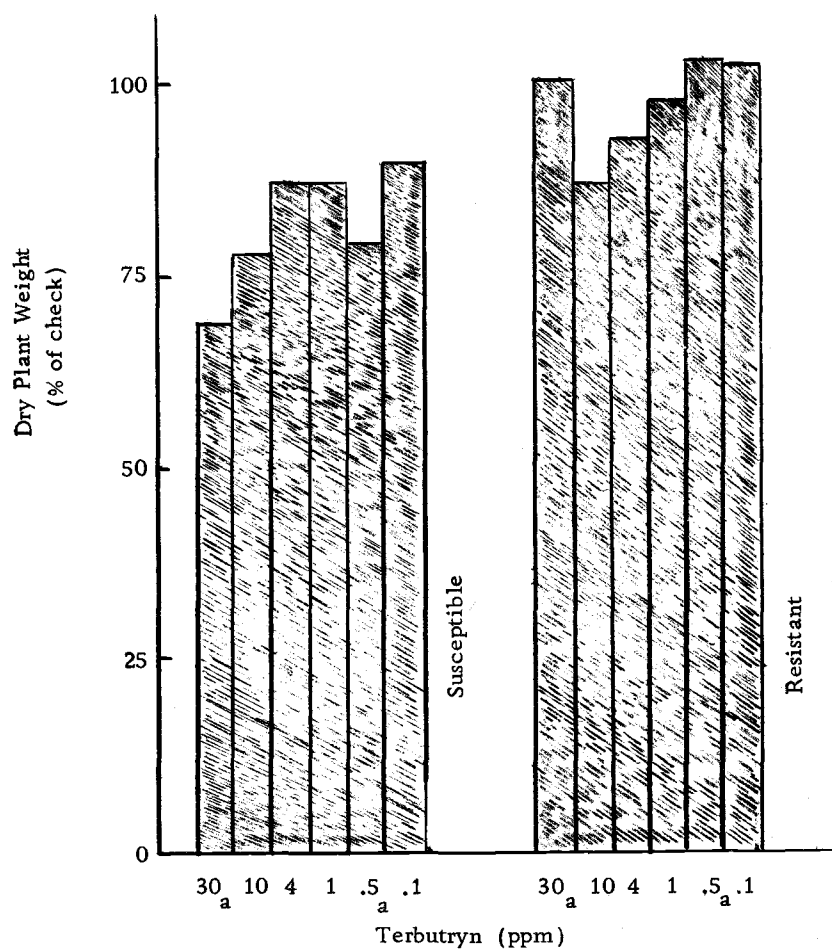


Figure 3. Experiment 5. The dose response of two common groundsel biotypes to terbutryn. Plants treated 30-32 days after germination.

^a Average of 4 replications.

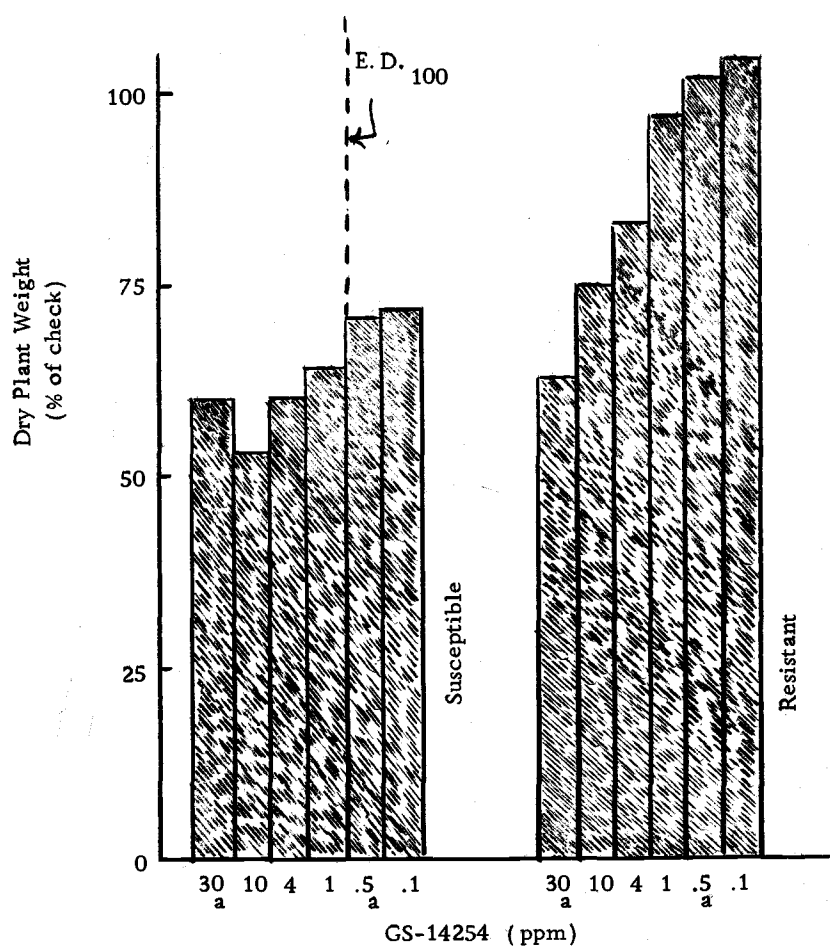


Figure 4. Experiment 6. The dose response of two common groundsel biotypes to GS-14254. Plants treated 30-32 days after germination.

*Effective dose 100%: values to the left of line determined by visual evaluation equal to complete control.

^a Average of 4 replications.

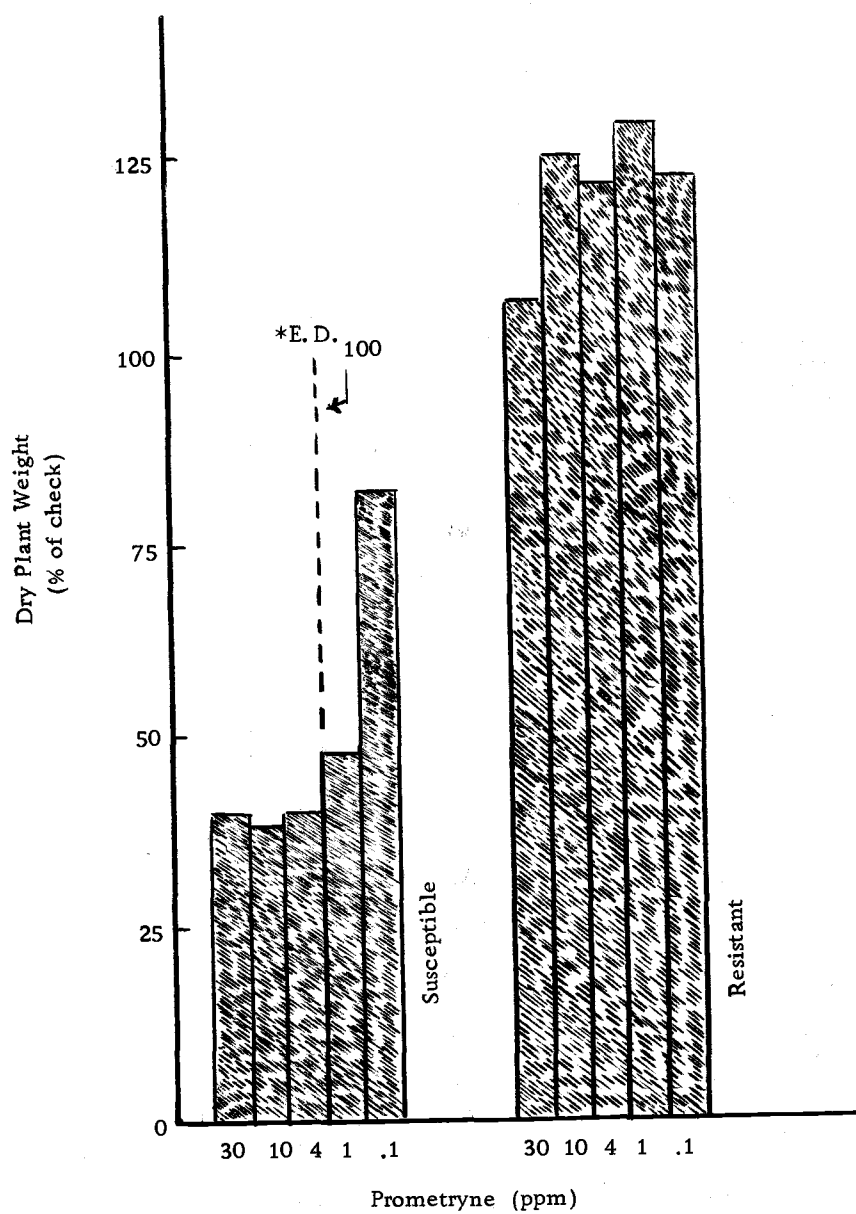


Figure 5. Experiment 7. The dose response of two common groundsel biotypes to prometryne. Plants treated 30-32 days after germination.

*Effective dose 100%: values to the left of line determined by visual evaluation equal to complete control.

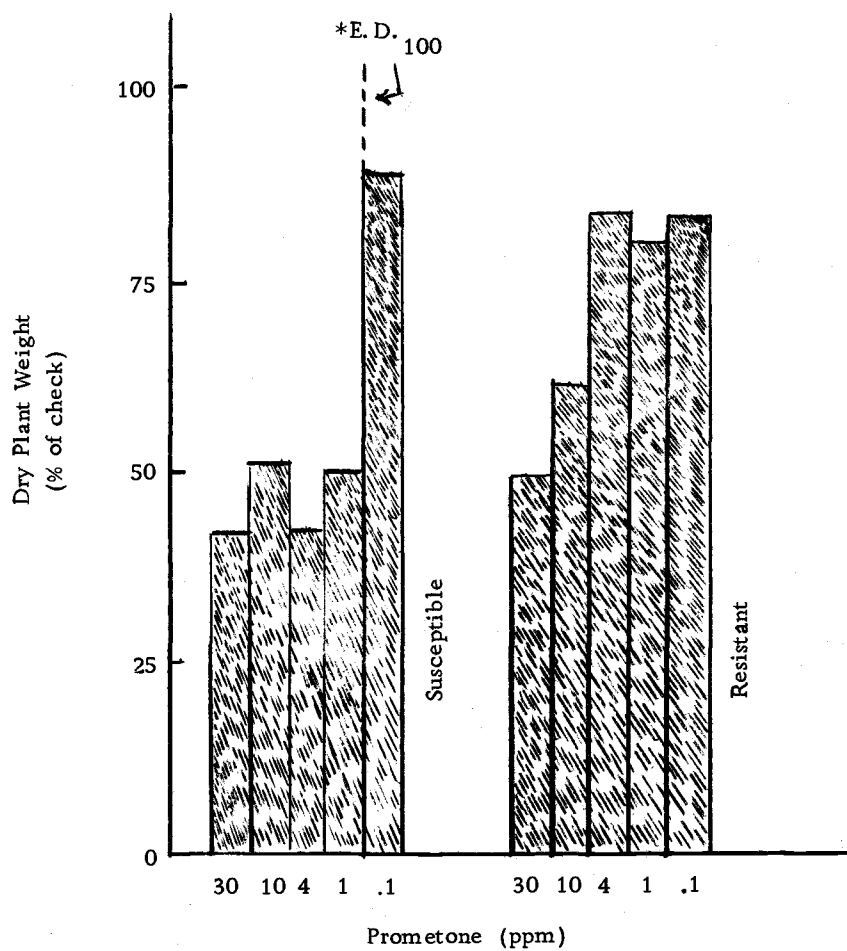


Figure 6. Experiment 8. The dose response of two common groundsel biotypes to prometon. Plants treated 30-32 days after germination.

*Effective dose 100%: values to the left of the line were determined by visual evaluation equal to complete control.

the susceptible biotype but 30 ppm (1.33×10^{-4} M) did not completely control the R plants. A degree of growth reduction of the resistant biotype was noted at the highest concentration of atrazine, GS-14254, and prometone. However, the characteristic symptoms of photosynthetic inhibition were not observed at any time in the resistant common groundsel.

Experiment 9: The Dose Response of Corn to Atrazine

The dose response of corn to several high rates of atrazine are presented in Figure 7 and Appendix Tables 44 through 49. Corn was subjected to 30, 15, and 5 ppm concentrations of atrazine. Growth inhibition was noted at each of these herbicide rates; however, symptoms of photosynthetic inhibition were not observed.

Discussion

Ryan (1970) showed a differential response between two biotypes of common groundsel to atrazine and simazine. One biotype was found to be much more susceptible than the other. The results of this study support Ryan's observations and, further, suggest that these differences in response are due to an inherent physiological difference between the two biotypes in the presence of a triazine herbicide. Prior to herbicide treatments, seeds were pregerminated and plants were grown in nutrient culture. This procedure

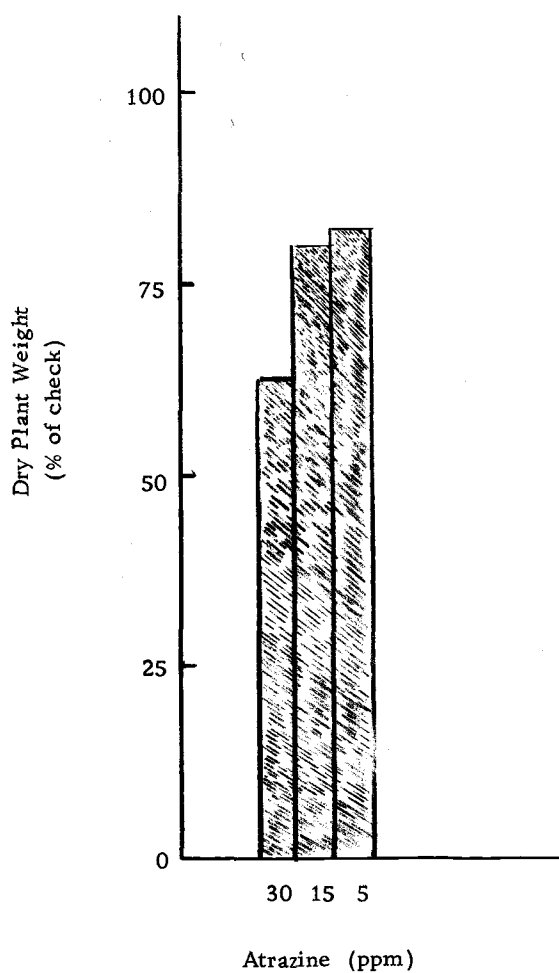


Figure 7. The dose response of corn to atrazine. Plants treated 7 days after germination.

eliminated soil factors, morphological differences, and growth rates as factors causing the differential herbicide response. Examination of both R and S biotypes also revealed similar morphologic structure. In all cases, when the R biotype was subjected to a triazine herbicide, the resistance was maintained. This resistance was found to be similar to corn which is noted for its high degree of atrazine tolerance. Growth inhibition of the resistant biotype was often noted at the highest concentration of several of the herbicides studied. However, the characteristic symptoms of photosynthetic inhibition were never observed with this biotype. Jordon, Marashige, Mann, and Day (1966), using nonphotosynthetic tobacco callus, presented evidence for an alternate mechanism of action of many photosynthesis-inhibiting herbicides. Perhaps such a mechanism could explain the phytotoxicity to the R biotype at the highest concentration of atrazine, GS-14254, and prometone. The S common groundsel biotype was significantly more susceptible to all triazines except terbutryn than its resistant counterpart. These and Ryan's data, in which effective control was observed when either biotype was subject to non-triazine herbicides, suggest the mechanism of resistance to be basic to the triazine structure.

The results of these studies demonstrate the necessity for having more than one herbicide for a particular crop use. They

also point out that if herbicides are to be rotated, the sequence should not contain members of the same herbicide classification.

III. PHOTOSYNTHESIS, ABSORPTION, AND METABOLISM STUDIES OF TWO COMMON GROUNDSEL BIOTYPES TREATED WITH SIMAZINE

Introduction

Montgomery and Freed (1964) stated that plant tolerance varies widely to triazine herbicides with different molecular structures. Often, closely-related plants exhibit marked differences in response to a given triazine. Likewise, certain plants especially noted for their tolerance to one triazine are frequently quite sensitive to other types. In recent years, considerable investigation has been initiated to study those factors responsible for plant tolerance to these herbicides. Several factors have been suggested (Montgomery and Freed, 1964). They include: restricted herbicide uptake by tolerant species but not sensitive ones, differential rates of metabolism or detoxification of the herbicide, and inhibition of biochemical systems in sensitive plants which are not affected in resistant plants. Also possible are restricted herbicide movement to the site of action and storage of the intact herbicide in areas, where it cannot disrupt plant function.

Restricted herbicide uptake by tolerant species as compared to susceptible species is often proposed as a mechanism for herbicide selectivity. This has not been fully substantiated with triazine herbicides. Davis, Gramlich, and Funderburk (1965), in studies with soybean, cotton, and corn, found atrazine susceptibility to coincide

with the amount of herbicide absorbed. However, Sheets (1961) and Negi, Funderburk, and Davis (1964) concluded that atrazine or simazine absorption could not be related with plant tolerance. A direct relationship between transpiration and absorption of the herbicide was observed in both studies. In most cases, the resistance of plants to triazine herbicides is inadequately explained by herbicide absorption alone (Montgomery and Freed (1964).

With triazines, different plants apparently possess varying capacities to metabolize or detoxify the herbicide to non-phytotoxic products. Numerous investigations have shown a relationship between the amount of triazine tolerance and rate of metabolism (Castelfranco, Foy, and Deutsch (1961); Davis, Funderburk, and Sansing (1959); Hamilton (1964); Hamilton and Moreland (1964); Montgomery and Freed (1964); Roth (1957); Roth and Knüsli (1961); Shimabukuro, Kadunce, and Frear (1966); Shimabukuro, Swanson, and Walsh (1970). These studies have prompted Shimabukuro et al. (1970) to propose the following metabolic scheme for atrazine metabolism in higher plants.

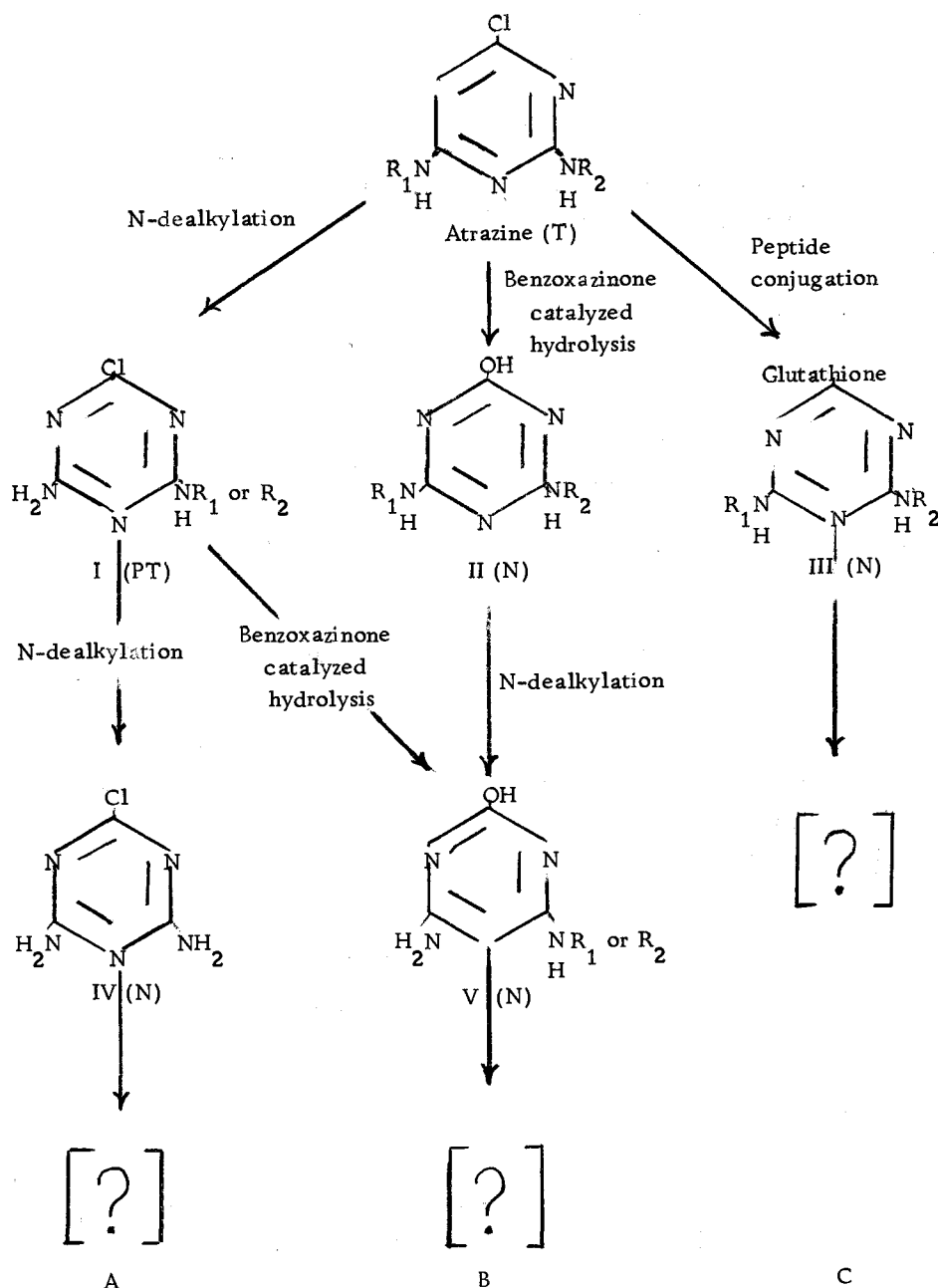


Figure 8. Metabolic scheme for atrazine metabolism in higher plants. I: 2-chloro-4-amino-6-(isopropylamino)-s-triazine or 2-chloro-4-(ethylamino)-6-amino-s-triazine; II: hydroxyatrazine; III: GS-atrazine; IV: 2-chloro-4,6-diamino-s-triazine (isolated from sorghum and identified by infrared and mass spectrometry, unpublished data); V: hydroxy compounds I and II (Shimabukuro *et al.*, 1970).

Evidence indicates that higher plants are capable of detoxifying atrazine by at least three metabolic pathways. Pathway A (N-dealkylation) is an enzymatic process in which hydrocarbon side chains are removed from the herbicide molecule. It is important in intermediately susceptible species such as pea or cotton. Pathway B involves both nonenzymatic benzoxazinone-catalyzed hydrolysis (described by Castelfranco *et al.*, 1961; Roth and Knüsli, 1961; and Hamilton and Moreland, 1962) and enzymatic (N-dealkylation) reactions to give several nonphytotoxic products (Shimabukuro, 1968). Pathway B seems to be limited to species such as wheat and corn which contain benzoxazinone and is primarily associated with root tissue (Shimabukuro *et al.*, 1970). Pathway C appears to be active in highly resistant species, corn and sorghum (Lamoureux, Shimabukuro, Swanson, and Frear, 1970; Shimabukuro *et al.*, 1970). Compound III (see Figure 8) is apparently formed in corn and sorghum by an enzymatically-catalyzed condensation of glutathione with atrazine. Pathway C is especially active in foliage tissue (Shimabukuro *et al.*, 1970).

Extensive metabolism of simazine has also been demonstrated by Montgomery, Freed, and Fang (1958) and Funderburk and Davis (1963). In these studies either ring- or side-chain-labeled ^{14}C -simazine was applied to several plant species. $^{14}\text{CO}_2$ evolution by plants treated with either type of labeled herbicide indicated that all

portions of the triazine ring are subject to complete oxidation. These studies implicate the metabolism of the triazine herbicide as an important role in plant tolerance.

Another possible explanation for the tolerance of certain plants is that the herbicide may not be active against some biochemical systems inhibited in sensitive species. In exhaustive studies, investigators have shown the triazines to be potent inhibitors of photosynthesis. Moreland, Gentner, Hilton, and Hill (1961) observed that simazine could greatly reduce the photochemical capacity of isolated barley chloroplasts. However, a supply of exogenous sucrose could partially overcome the lethal effects of the chemical. Ashton (1965) found that the phytotoxicity of monuron and atrazine increased with increasing light intensity and also increased at light wavelengths absorbed by chlorophyll a and b. Thus, herbicide toxicity has been associated with the light reaction of photosynthesis and not strictly plant starvation. Much of the evidence concerning the cause for toxicity by substituted phenyl urea herbicides such as monuron and diuron, may also explain the phytotoxic effects of triazines. Stanger and Appleby (1972) have proposed a mechanism for diuron-induced photooxidation of chlorophyll. They believe this occurs as a result of inhibition of NADPH which is necessary to maintain a carotenoid protective system within the intact chloroplast. Such a mechanism might also explain the lethal action of many triazine herbicides.

In previous experiments (see section II, experiments 3-8), when simazine was applied to either biotype of common groundsel, the susceptible plants quickly became chlorotic and died. The resistant biotype did not exhibit these characteristic symptoms of photosynthetic inhibition. The purpose of this study was to determine the photosynthetic response of both biotypes in the presence of simazine. The experiments described herein were also undertaken to study the uptake and distribution of simazine in both S and R biotypes and to determine the metabolic change of simazine in both plant tissues.

Materials and Methods

Plant Material

Seeds of an atrazine- and simazine-resistant biotype of common groundsel were obtained (Ryan, 1970) and plants from this source were subjected to several rates of atrazine. Seed from those plants surviving the highest rate of atrazine (8.96 Kg/ha) were collected and used in the following experiments. Atrazine- and simazine-susceptible plants were obtained by randomly gathering seed from common groundsel in the Willamette Valley and subjecting plants from a portion of this seed source to several rates of atrazine. These plants in all cases failed to survive the lowest application rate

(1.12 kg/ha). (See section II, experiments 1 and 2.)

The seed of both biotypes were germinated on moist filter paper at 25° C, 19,000 lux, and 16-hour day length. After 7 days, seedlings were transferred to black polyethylene containers with 0.1-strength Hoagland's solution. Plants grown for photosynthesis studies were placed in 225-ml pots, five plants per container.

Plants for absorption and metabolism experiments were grown in 1-liter freezer containers and roots were kept separate by glass dividers. Nine plants were grown in each container. At the time of transfer, seedlings were placed between the halves of a split cork and fitted into a hole in the container lid with roots extending into the nutrient solution. All plants were continually aerated and all experiments were conducted under the above described growth chamber conditions unless otherwise stated. At flowering (approximately 30 days after transfer), plants of each biotype were selected for uniformity and subjected to either photosynthesis, absorption, or metabolism study.

Experiment 10: The Photosynthetic Response of Two Common Groundsel Biotypes to Simazine

Plants of S and R biotypes were grown as described above, selected for uniformity, and subjected to either 0 or 0.5 ppm of simazine. Herbicide application was made directly to the roots by

supplying the appropriate amount of technical herbicide dissolved in 0.1-strength Hoagland's solution.

After several lengths of herbicide exposure, net CO₂ fixation was measured for 10 minutes with an infrared gas analyzer. Simazine exposure times ranged from 0 to 48 hours. The herbicide was removed from the S biotype after 24 hours; however, photosynthesis monitoring was continued.

During the time of herbicide exposure and photosynthesis measurement all plants were maintained under continuous illumination, 19,000 lux, and 25° C. Foliage dry weights were determined when the experiment was terminated and the data were expressed as mg CO₂ fixed/gram dry foliage weight/10 minutes. The experiment was conducted as a factorial arrangement of a completely randomized design. Each treatment was replicated three times and the entire experiment was repeated. Plants used during the initial experiment were 1 week older than those used later.

Experiment 11: The Absorption of Simazine by Two Biotypes of Common Groundsel as a Function of Time

At the beginning of flowering, plants of both biotypes were selected for uniformity and transferred to glass cups containing 75 ml of 5×10^{-4} M calcium nitrate. Each container was fitted with a plexiglass lid designed for holding plants. Lids were cut into half

circles and held together with a single hinge. The inside surface of each half circle was lined with foam rubber to avoid crushing plant stems. Light entry and possible herbicide adsorption to the plexiglass was prevented by wrapping each lid with aluminum foil. Each container, holding 4 plants, was placed in a light-proof box with only foliage exposed.

The plants were kept in calcium nitrate for 24 hours prior to being transferred into 75 ml of experimental solution. The experimental solution consisted of 5×10^{-4} M calcium nitrate, 130.8 μg simazine, and 19.2 μg uniformly ring-labeled ^{14}C -simazine (20.8 $\mu\text{c}/\text{mg}$). This gave a total concentration of 2.0 ppm of labeled and unlabeled herbicide in solution. Exposure times ranged from 1 to 24 hours.

Following exposure to the experimental solution, the plant roots were rinsed for one minute. The composition of the rinsing solution was identical to the experimental solution except it contained 2.0 ppm of unlabeled simazine and no radioactive simazine. The purpose of this rinse was to replace any ^{14}C -simazine adsorbed to the root surface with unlabeled herbicide. Corrections for evaporation, herbicide volatilization, and adsorption to the container walls were made following each exposure time using a control treatment, identical to the others except that it contained no plants. In addition, a preliminary study was conducted to determine the extent of simazine

adsorption to container walls as a function of time (see Appendix table 52). Less than 8.0% of the herbicide was adsorbed within 192 hours and maximum effects were observed within 15 minutes.

Immediately following each exposure period, the experimental solution was measured and returned to its original volume, thus correcting the herbicide concentration for plant uptake and giving a measurement of transpiration. Plants were sectioned into roots and foliage. Roots were blotted and fresh and dry weights of each plant part were determined.

The amount of simazine absorbed by the plants was found by measuring the 14 carbon activity in 1.0-ml aliquots from both experimental and rinse solutions using liquid scintillation counting. The difference between the sum of these two solutions and the total activity provided indicated the amount of simazine absorbed by the plants. All counting values were corrected for background and quenching. One liter of scintillation fluid contained 666 ml toluene, 333 ml Triton X-100, 5.5 grams of 2,5-diphenyloxazole (PPO) and 0.1 gram of 1,4 bis[2-(5-phenyloxazolyl)]benzene (POPOP). Each counting vial, in addition to the sample aliquot, contained 10 ml of scintillation fluid and 2 ml toluene.

The experiment was conducted as a factorial arrangement of a randomized block design containing two replications. The experiment also was repeated (the duplicate experiment was conducted

using 5 plants per treatment container).

Experiment 12: The Metabolism of Simazine by Two Biotypes of Common Groundsel as a Function of Time

This experiment was conducted as two similar studies. Unless otherwise stated, plant materials were grown and herbicide absorption was determined as earlier described. Plants of both biotypes used in study A (four plants per treating container) were exposed to an experimental solution containing 126.0 μg simazine and 24.0 μg uniformly ring-labeled ^{14}C -simazine (20.8 $\mu\text{c}/\text{mg}$) for 24 hours. The experimental solution used during study B contained 14.4 μg uniformly ring-labeled ^{14}C -simazine (20.8 $\mu\text{c}/\text{mg}$) and 135.6 μg simazine. In both cases total herbicide concentration was 2.0 ppm and total ^{14}C carbon activity was 0.5 μc and 0.3 μc respectively. Five plants per treating container were used in study B.

After the initial 24 hours of herbicide absorption, plants were removed from the experimental solution. Roots were rinsed in 2.0 ppm simazine for one minute (see experiment 11), and each plant was transferred into 2.0 ppm of nonlabeled herbicide solution for various lengths of time. The purpose of these incubation periods was to allow a greater amount of simazine metabolism. Incubation times ranged from 0 to 48 hours in study A and from 0 to 72 hours for study B. Every 24 hours, plants were transferred into fresh

calcium nitrate or a 2.0 ppm non-radioactive simazine solution during studies A and B, respectively. The maximum herbicide exposure time for each study was therefore 72 and 96 hours. Each exposure period was terminated by sectioning plants into root and foliage, determining fresh weights of each plant section and plunging each section into liquid nitrogen. Plant material was then stored at -30°C until extraction.

Extraction and Counting Procedure

At the end of each exposure period the roots and shoots were dipped into liquid nitrogen and then extracted by homogenizing the material with 20 ml of either 95% (study A) or 80% (study B) methanol for 10 minutes in an omnimixer. The homogenate was filtered and the residue was reextracted twice by rehomogenizing with 20 ml of the appropriate water-methanol solution. This mixture (for root tissue only) was boiled for several minutes. Residue in most cases was white to pale green in color. Methanol was evaporated under vacuum at 40°C from the combined filtrate, and the remaining aqueous portion was concentrated to near dryness in study A and to a small volume in study B. All evaporation and concentration operations were conducted under identical conditions.

The aqueous foliage extract was washed three times with equal volumes (5 ml) of chloroform. The aqueous root extract was washed

twice with chloroform followed by boiling for several minutes after each wash to remove any remaining chloroform. One-ml aliquots of the resulting chloroform and aqueous solutions were assayed for ^{14}C activity by liquid scintillation counting (see experiment 11).

Before adding scintillation fluid, chloroform was removed under vacuum from each vial. Portions of the root and foliage residue were analyzed for ^{14}C activity by gel-scintillation counting. Each sample vial contained 20 ml of scintillation gel plus the portion of finely ground residue. The scintillation gel per liter of toluene consisted of 40 grams Cab-O-Sil, 5.5 g PPO, and .1 g POPOP. All samples were corrected for quenching and background and counts are expressed as disintegrations per minute (DPM).

The chloroform partition was further concentrated to a 2-ml volume, and 20- μl aliquots from each sample were spotted on plates coated with silica gel (EM Laboratories, Inc.) 250 microns in thickness. Aliquots of extracts from both S and R biotypes were developed on the same plate for direct comparison. Thinlayer chromatograms were developed in benzene-acetic acid (50:4). Radioactivity on the chromatograms was detected with a radiochromatogram scanner. Rf values were calculated and compared. Thinlayer chromatograms with only ^{14}C -simazine were also prepared for comparison to treatments. Detection of ^{14}C -simazine and ^{14}C activity in the chloroform extract of one observation of study B was also made by radioautography

of thinlayer plates on Kodak No Screen X-Ray Film.

Both studies were conducted as factorial arrangements of a completely randomized design. Study A contained two observations per treatment while study B contained three.

Experiment 13: The Metabolism of
Simazine by Corn (*Zea mays* L.)

Corn seeds, variety KE 435 (Northrup King), were germinated at 25° C, 19,000 lux, and 16-hour day length. After 3 days seedlings were transferred to 100 ml of 0.25 Hoagland's solution in containers described in experiment 11. At 7 days of age, plants (4 plants per container) were transferred to 75 ml of 5×10^{-4} M calcium nitrate for 24 hours. All plants were then exposed to 75 ml of experimental solution for 72 hours. The experimental solution used was identical to that described in study B of experiment 12. Following exposure to the experimental solution, the plant roots were rinsed for 1 minute in 75 ml of 2.0 ppm unlabeled simazine. Corrections for evaporation, herbicide volatilization, and adsorption to the container walls were made as described in experiment 11. Immediately following the exposure period, the volume of experimental solution remaining was measured and returned to its original volume. One ml aliquots were then taken and counted by liquid scintillation (see experiment 11). Plants were sectioned into roots and shoots, and fresh weights

of each section were determined. Plant parts were plunged into liquid nitrogen, and stored at -30°C until extraction. The amount of simazine absorbed was determined as described in experiment 11. Extractions were accomplished in the same manner as study B of experiment 12. Residue, chloroform, and aqueous extracts were counted by gel or liquid scintillation techniques (see experiment 12).

Results

Experiment 10: The Photosynthetic Response of Two Common Groundsel Biotypes to Simazine

Data for this experiment are illustrated graphically in Figures 9 and 10 and presented in Appendix Tables 50 and 51. Net photosynthesis of the S biotype was completely inhibited following an application of simazine (0.5 ppm). However, when the R biotype was subjected to a similar rate of the herbicide, no effect was observed. Also noted was a significantly higher photosynthetic rate by the untreated resistant plants when compared to the untreated plants of the triazine-susceptible biotype. Because complete photosynthetic inhibition of the S biotype occurred within 24 hours after simazine application, the herbicide was removed from those plants at that time. A definite recovery of photosynthetic ability was observed upon removal of the herbicide.

These data suggest several possible explanations for the

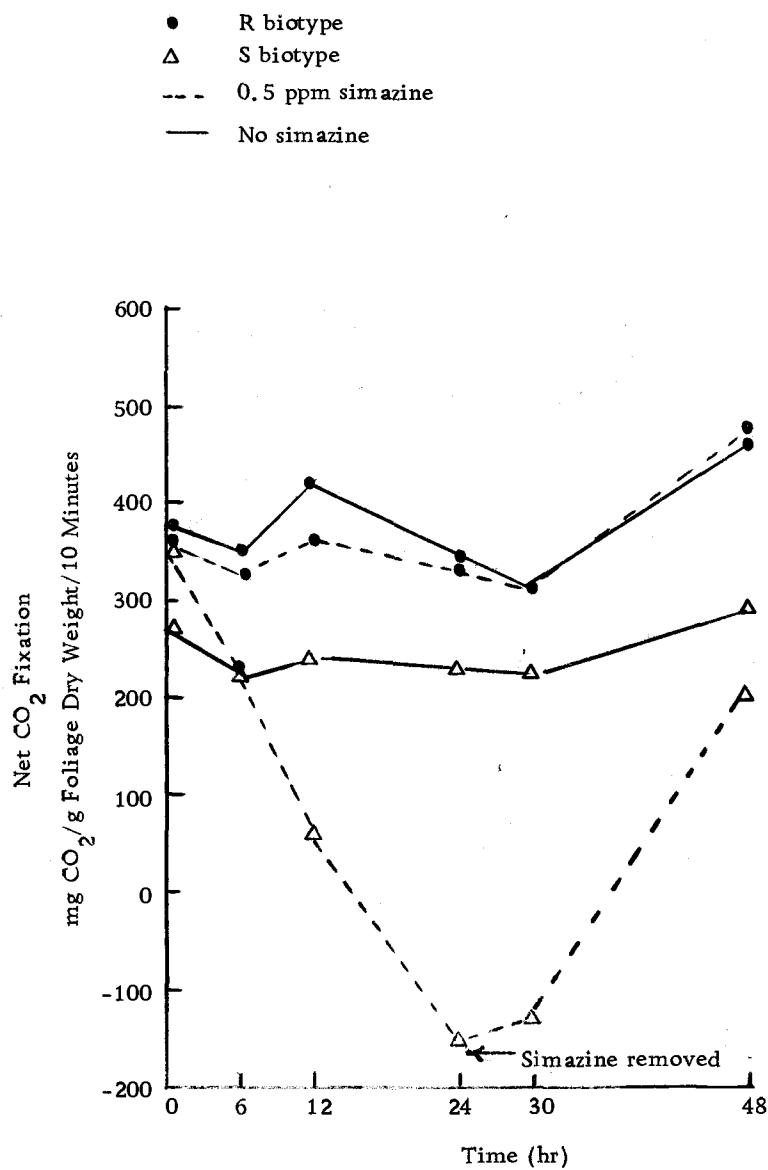


Figure 9. The photosynthetic response of two common groundsel biotypes to simazine. Treatment Date 1.

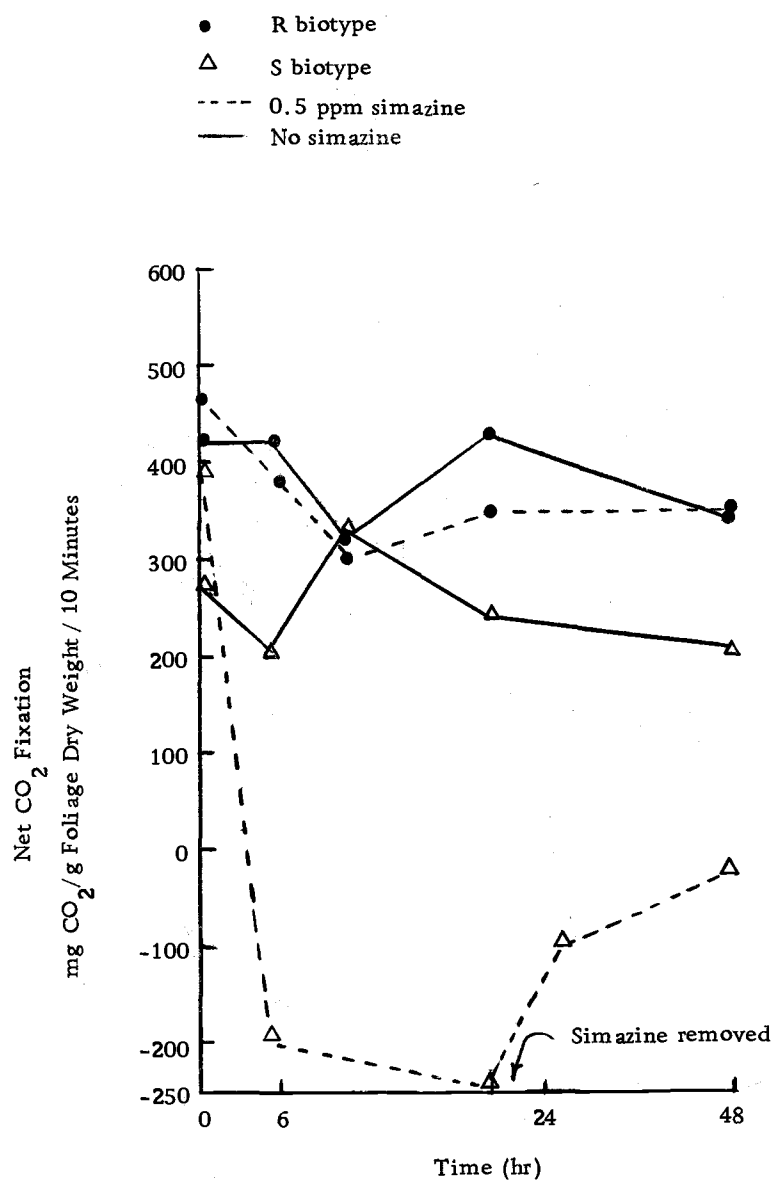


Figure 10. The photosynthetic response of two common groundsel biotypes to simazine. Treatment Date 2.

simazine resistance exhibited by the R biotype. The herbicide may be absorbed in phytotoxic quantities by the susceptible plants but not by the resistant plants. It may be absorbed in equal quantities by both biotypes and detoxified to nontoxic products in the R biotype but not the S biotype. The simazine may be stored or in some manner made unavailable to the site of herbicide action in resistant plants and not in susceptible plants. Finally, the herbicide may be absorbed equally and reach the site of action but chlorophyll may be rapidly replenished by resistant plants.

Experiment 11: The Absorption of Simazine by Two Biotypes of Common Groundsel as a Function of Time

Data for experiment 11 (two treatment dates) are summarized in Tables 3 and 4 and presented in Appendix Tables 53 through 66. Also, data for treatment date 2 are illustrated in Figures 11 through 14.

Total simazine uptake by the S biotype always exceeded the uptake by the R biotype (see Tables 3 and 4 and Figure 11). However, when simazine absorption per gram fresh or dry weight were compared for both biotypes, little difference existed (see Figures 12 and 13). These data support the findings of Abe (1971), who concluded that differential absorption does not play a role in the mechanism of resistance of common groundsel to simazine. It does not seem

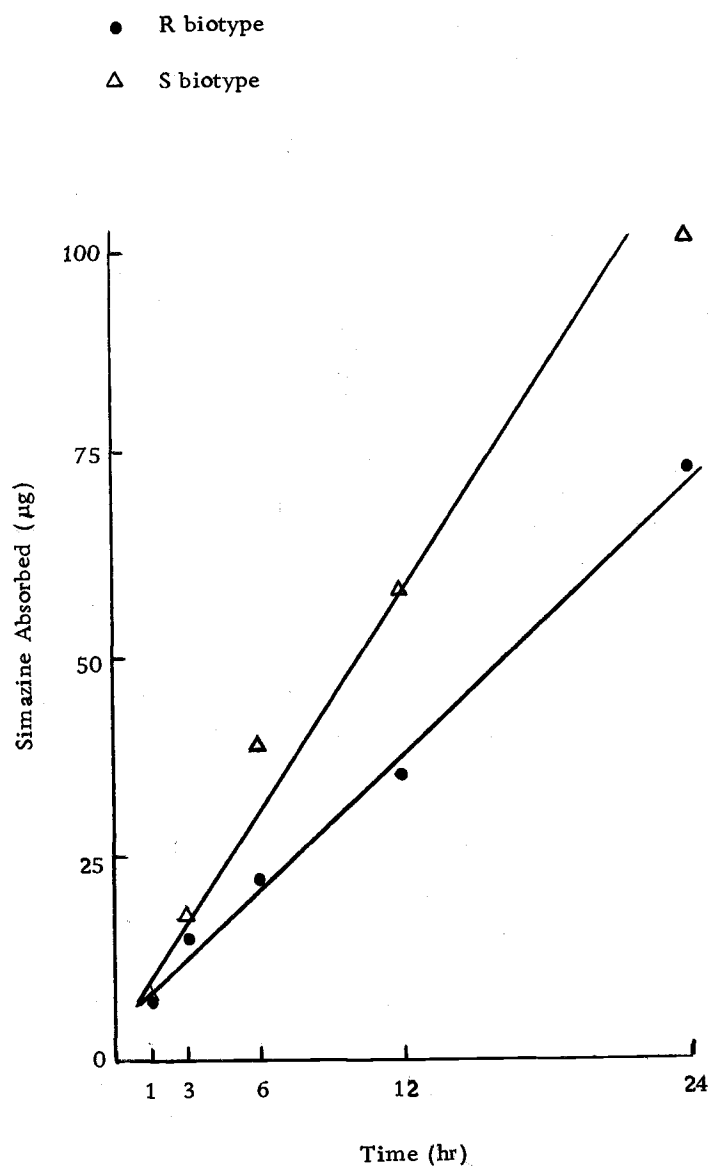


Figure 11. Total simazine absorbed by two common groundsel biotypes as a function of time. Treatment Date 2. Avg. of 10 plants.

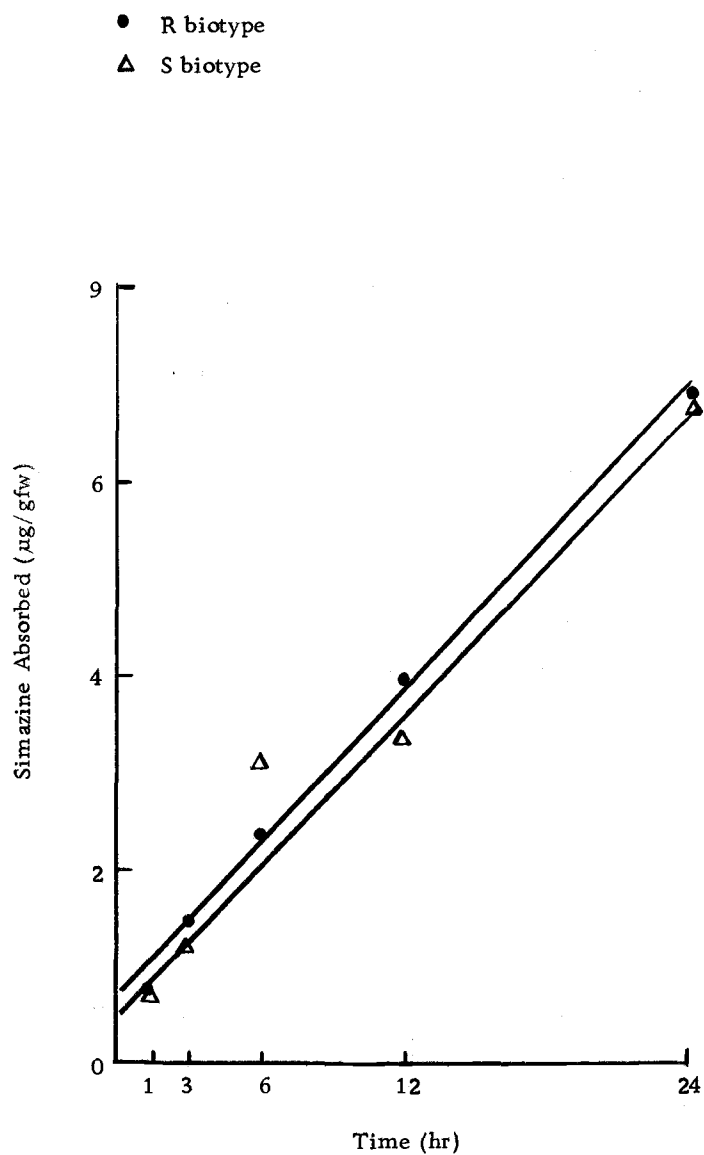


Figure 12. Simazine absorption per gram fresh weight by two common groundsel biotypes as a function of time. Treatment Date 2. Avg. of 10 plants.

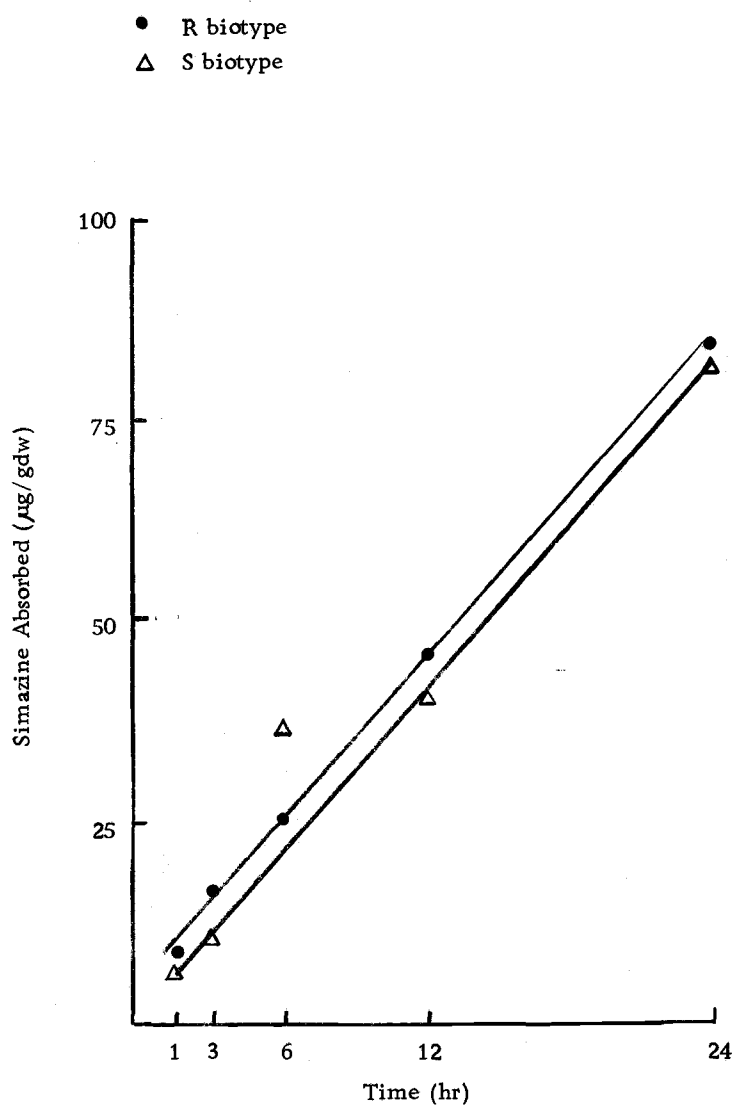


Figure 13. Simazine absorption per gram dry weight of two common groundsel biotypes as a function of time. Treatment Date 2. Avg. of 10 plants.

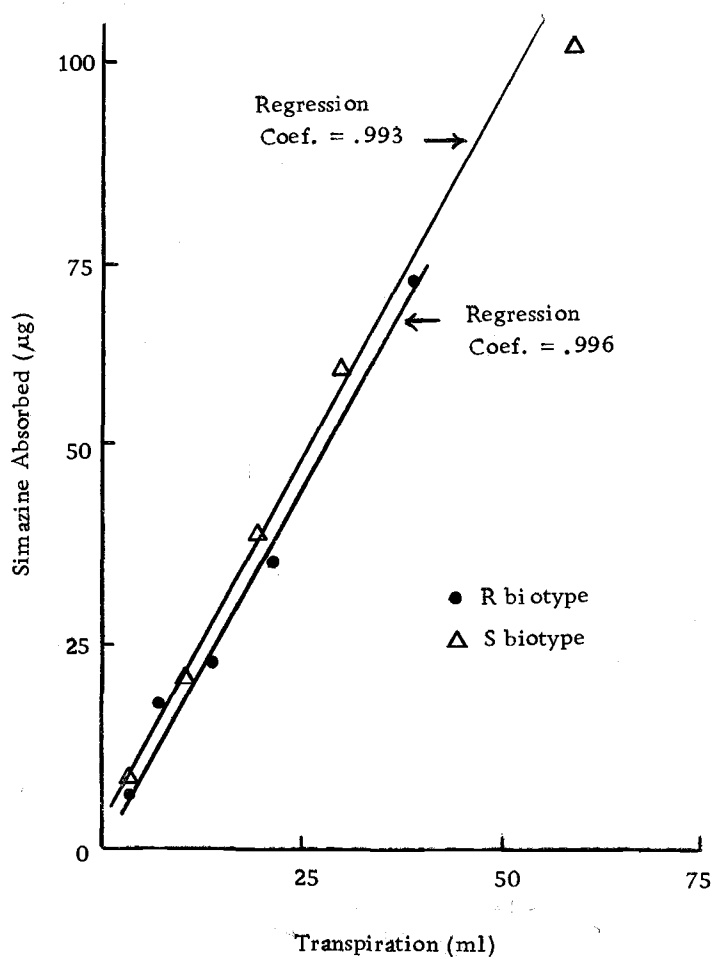


Figure 14. Absorption of simazine by two common groundsel biotypes as a function of transpiration. Treatment Date 2. Avg. of 10 plants.

Table 3. The uptake of simazine and water by two biotypes of common groundsel as a function of time. ^a Treatment date 1.

Exposure Time	Transpiration	Simazine Absorbed		
		Total	Per g Fresh Weight	Per g Dry Weight
	(ml)	(μg)	($\mu\text{g} / \text{g}$)	($\mu\text{g} / \text{g}$)
<u>Resistant</u>				
1	1.3	3.1	0.7	7.4
3	1.6	3.1	0.8	7.9
6	4.9	2.7	0.8	7.8
12	8.7	15.4	4.1	42.8
24	13.8	16.0	4.2	47.3
<u>Susceptible</u>				
1	1.0	3.4	0.4	4.6
3	3.4	6.8	0.9	9.9
6	4.6	14.2	2.8	23.8
12	12.0	19.4	3.0	34.4
24	14.4	26.7	6.3	48.3

^a Averages of 8 plants.

Table 4. The uptake of simazine and water by two biotypes of common groundsel as a function of time. ^a Treatment date 2.

Exposure Time	Transpiration	Simazine Absorbed		
		Total	Per g Fresh Weight	Per g Dry Weight
(hr)	(ml)	(μ g)	(μ g/g)	(μ g/g)
<u>Resistant</u>				
1	3.5	7.0	0.7	9.2
3	7.7	14.9	1.4	17.5
6	13.7	22.2	2.3	26.4
12	17.7	35.2	3.9	46.1
24	39.4	73.3	6.9	84.2
<u>Susceptible</u>				
1	3.6	8.4	0.6	6.9
3	10.8	17.8	1.2	14.2
6	17.0	39.2	3.1	36.8
12	30.2	58.6	3.3	40.6
24	59.0	102.8	6.8	81.4

^a Average of 10 plants

likely, therefore, that differences in simazine tolerance by the two common groundsel biotypes can be explained by differential uptake of the herbicide.

A clear and definite relationship between water uptake and simazine absorption is indicated in Tables 3 and 4 and Figure 14. These data were subjected to linear regression analysis and simazine absorption was found to be highly correlated to transpiration by both biotypes. These data support the observations of Sheets (1961) and Davis et al. (1965). This suggests that the herbicide enters the plant in a passive manner with established moisture gradients.

Experiment 12: The Metabolism of Simazine by Two Biotypes of Common Groundsel as a Function of Time

Differential plant tolerance resulting from herbicide detoxification is most often explained by differences in metabolic rate, differences in metabolic pathway, or both. Shimabukuro et al. (1970) has proposed a metabolic scheme for atrazine metabolism in higher plants (see Figure 8) and metabolism of triazine herbicides has been implicated as an important factor determining plant tolerance to these chemicals.

Aqueous plant extracts of two common groundsel biotypes previously subjected to ^{14}C -simazine were made and partitioned several times with chloroform. The resulting water and chloroform

fractions would then contain any hydrophilic and lipophilic metabolites which may be present. Intact simazine and any dealkylated metabolites would largely be located in the chloroform fraction, while any water-soluble metabolites, such as the 2-hydroxy metabolite or the glutathione complex would be found in the water portion. Data from these studies are summarized in Tables 5 and 6 and further presented in Appendix Tables 67 through 78.

The greatest proportion of ^{14}C was recovered from the chloroform fraction of the foliage extracts of both S and R biotypes. Furthermore, negligible differences in ^{14}C activity were observed between the two biotypes at any time period studied. Much less ^{14}C was observed in the water extracts of both roots and foliage. However, foliage also tended to accumulate the water-soluble metabolites. No difference in amount or rate of formation of water-soluble metabolites was apparent between the two common groundsel biotypes. These data suggest that if differential metabolism of simazine occurs in common groundsel, the difference between the S and R biotype must be located in the chloroform-soluble fraction of foliage tissue.

Aliquots of the chloroform extract from R and S common groundsel foliage were spotted on silica gel thinlayer plates. The thinlayer chromatograms were developed in benzene-acetic acid (50:4), as described by Shimabukuro et al. (1966) for detection of

Table 5. The distribution of ^{14}C activity from applied ^{14}C -simazine in the extracts of foliage and roots of two common groundsel biotypes as a function of time. ^a Study A.

Biotype	Treatment Time (hr)	^{14}C Recovered from Plant Extraction			
		% in Foliage		% in Roots	
		Chloroform	Water	Chloroform	Water
Susceptible	24	87	4	8	1.0
	48	86	9	4	1.5
	72	81	14	2	1.5
Resistant	24	87	4	2	1.0
	48	86	10	1	1.0
	72	92	3	2	1.5

^a Averages of 8 plants.

L. S. D. for extracts = 4.1%
.01

Table 6. The distribution of ^{14}C activity from applied ^{14}C -simazine in foliage and root extracts of two common groundsel biotypes as a function of time. ^a Study B.

Biotype	Treatment Time (hr)	^{14}C Recovered from Plant Extraction			
		% in Foliage		% in Roots	
		Chloroform	Water	Chloroform	Water
Susceptible	24	79	9	7	2
	48	81	14	2	2
	96	83	14	2	1
Resistant	24	72	12	12	2
	48	82	13	3	2
	96	83	13	2	1

^a Averages of 12 plants.

L. S. D. for treatment times = 4.8%
.05

L. S. D. for extracts = 3.8%
.01

atrazine and chloroform-soluble dealkylated metabolites of atrazine. The ^{14}C was detected using a radiochromatogram scanner and Rf values were determined. (See Table 7, Appendix Tables 79 through 84.) Radioautographs of thinlayer chromatograms also were made as shown in Figure 15.

A single peak was detected from chromatographs of either S or R biotypes. The Rf values for both biotypes were in most cases within .31-.36 (see Table 7 and Appendix Tables 79 through 84). When the Rf of samples were compared to intact ^{14}C -simazine and subjected to statistical analysis, no differences were found. However, a small but repeatable difference in Rf's between the S and R biotypes was observed. This difference is likely caused by differential amounts of plant material in each sample, thus hindering ^{14}C movement on the chromatograms in some cases. These data suggest that all ^{14}C -simazine introduced to the plants and recovered in chloroform foliage extract remained intact. Similar results were observed in the chloroform extracts of R and S root tissue.

Experiment 13: The Metabolism of Simazine by Corn

Data for experiment 13 are summarized in Table 8 below and presented in Appendix Tables 88 through 91. Aqueous extracts of corn plants previously subjected to ^{14}C -simazine for 72 hours were made and partitioned several times with chloroform. Intact

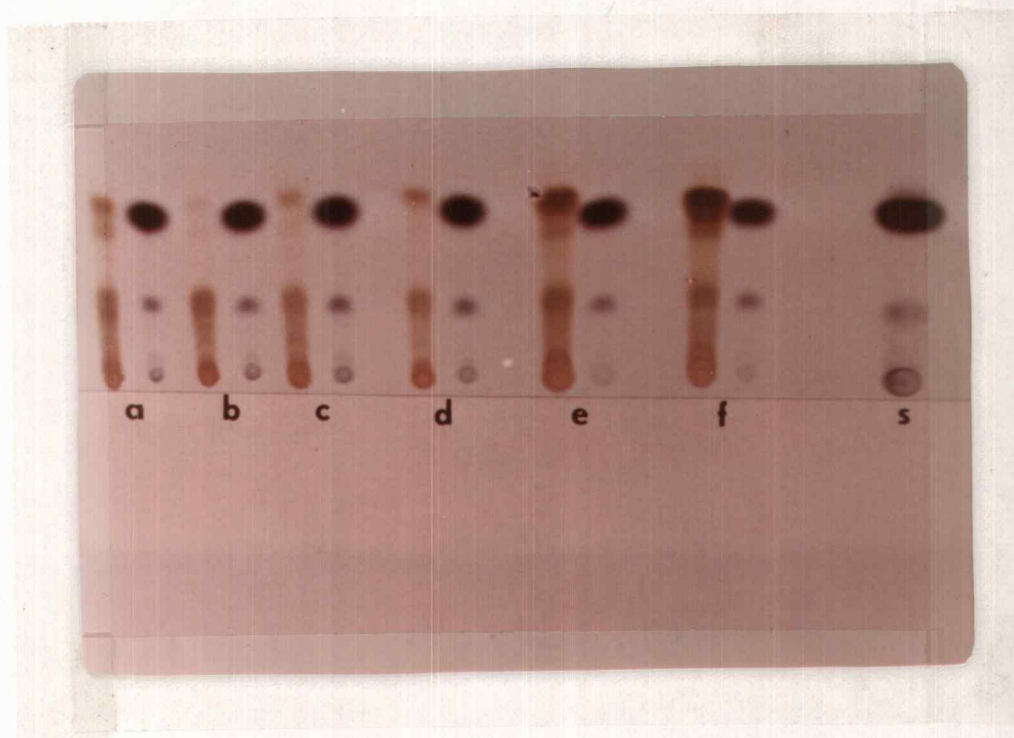


Figure 15. Radioautograph of thin-layer chromatogram. One observation of experiment 12, study B. Each letter a-f refers to a chromatogram (left) and radioautograph (right), respectively. a, c, and e are chloroform extracts of resistant biotype at 24, 48, and 96 hours of exposure to ^{14}C -simazine. b, d, and f are chloroform extracts of the susceptible biotype at 24, 48, and 96 hours of exposure to ^{14}C -simazine. s equals radioautograph of ^{14}C -simazine. Chromatograph developed in benzene:acetic acid (50:4).

Table 7. Rf values for ^{14}C -simazine and ^{14}C activity extracted from foliage of two common groundsel biotypes previously treated with ^{14}C -simazine. Studies A and B.

Study	Rf Value of ^{14}C from:		
	^{14}C -simazine	Resistant Biotype	Susceptible Biotype
A	.35	.28-.32	.29-.33
B	.35	.34-.39	.33-.34

Table 8. The distribution of ^{14}C activity in foliage and roots of corn after treatment with ^{14}C -simazine for 72 hours. A summary of data. ^a

Plant Part	% ^{14}C Activity Recovered in:		
	Chloroform	Water	Residue
Foliage	21	30	.2
Roots	17	31	.3
Total	38	61	.5

^a Average of 12 plants.

^{14}C -simazine and any dealkylated metabolites would be located in the chloroform fraction while hydroxylated metabolites or a glutathione complex (see Figure 8) would be found in the water portion. The greatest proportion of ^{14}C -activity in both roots and foliage of the treated plants was recovered as water-soluble metabolites. These data support the observations of numerous workers (Castelfranco et al., 1961; Davis et al., 1959; Hamilton, 1964; Montgomery and Freed, 1964; Roth, 1957; Roth and Knüsli, 1961; Shimabukuro et al., 1970) and indicate the basis for simazine tolerance by corn to be rapid conversion of the herbicide to nonphytotoxic water-soluble metabolites.

^{14}C -simazine was applied directly to the roots of the treated plants. However, ^{14}C activity was observed in root and foliage tissue in both chloroform and water partitions. These data suggest that a water-soluble metabolite of simazine is formed in the roots and translocated along with the herbicide to the leaves. These observations are also supported by the numerous workers cited earlier. This does not exclude the possibility, however, that some intact herbicide is metabolized to another water-soluble form in the foliage. Such a mechanism was found to exist in atrazine-resistant corn and sorghum (Lamoureux et al., 1970 and Shimabukuro, 1970). These data emphasize that the techniques used in earlier studies (see experiment 12) are sufficient for the isolation of water-soluble

metabolites of simazine and therefore support these observations.

Discussion

Two biotypes of common groundsel showed a differential photosynthetic response to simazine. Net CO₂ fixation, in the presence of the herbicide, was markedly inhibited in the S biotype but not in the R biotype. When simazine was removed from susceptible plants, photosynthesis resumed quickly. Furthermore, in the absence of the herbicide, resistant plants were able to maintain photosynthesis at a significantly more efficient level than susceptible plants.

Montgomery and Freed (1964) include differential herbicide uptake and metabolism as factors responsible for plant tolerance to triazine herbicides. Absorption studies revealed no differences in the ability of R and S biotypes to absorb simazine. These studies support the observations of Abe (1971) who concluded that differential absorption does not play a role in the resistance mechanism of common groundsel to simazine.

Metabolism studies, using techniques similar to Shimabukuro et al. (1966), Shimabukuro et al. (1970), and Thompson, Houghton, Slife, and Butler (1971) were conducted with R and S common groundsel biotypes. Greatest concentration of ¹⁴C activity was located in chloroform partitionings of foliage tissue from both biotypes. The ¹⁴C in this fraction of the extract was determined by thinlayer

chromatography to be similar to intact ^{14}C -simazine. While small amounts of ^{14}C were isolated in water partitions of the plant extracts, time-course studies revealed no differential increase in water-soluble simazine metabolites by either biotype. Furthermore, an experiment to study the metabolism of simazine by resistant corn plants revealed that the extraction techniques used in these studies were adequate for the isolation of water-soluble simazine metabolites. In light of these studies, it must be concluded that metabolism does not play an important part in the resistance of common groundsel to simazine.

IV. SUMMARY AND CONCLUSIONS

Two biotypes of common groundsel showed a differential response to chloro-, methoxy-, and methylmercapto-s-triazines. One biotype was found to be much more susceptible than the other. The results of this study support earlier observations by Ryan (1970) and Abe (1971) and suggest the mechanism for resistance to be basic to the triazine structure.

The use of herbicide rotations is often suggested as a means of preventing or postponing resistance in plants. Results of these studies demonstrate the need for having more than one herbicide for a particular crop use. They also point out that if herbicides are to be used in sequence, the rotation should not include members of the same herbicide classification. In addition to chemicals, other methods of weed control, such as plant rogueing and cultivation may be employed to help prevent resistance to herbicides.

When a triazine herbicide was applied, the susceptible plants quickly became chlorotic and died. The resistant biotype never exhibited these characteristic symptoms of photosynthetic inhibition. However, growth inhibition of the resistant biotype was often noted at high herbicide concentrations. A mechanism similar to that described by Jordon et al. (1966) might explain the phytotoxicity to the R biotype by high herbicide concentrations.

Photosynthesis was completely inhibited by simazine at 0.5 ppm in susceptible (S) plants but resistant (R) plants were unaffected. Furthermore, the susceptible biotype quickly recovered if the herbicide was removed. Muzik and Abe (1972) have proposed that the mechanism of resistance is due to inhibited root development of the S biotype but not the R biotype in the presence of simazine. While this may be an effect of the herbicide, it is difficult to relate the toxicity of this potent photosynthetic inhibitor with poor root development.

Two possible explanations for the simazine tolerance exhibited by the R biotype were explored. These were differential absorption and metabolism of the herbicide. Absorption studies revealed no differences in ability of R and S biotypes to absorb simazine. These findings support the observations of Abe (1971), who also worked with two common groundsel biotypes.

Metabolism studies involving both S and R biotypes indicated that simazine metabolism does not play an important role in the mechanism of resistance. However, the voluminous amounts of data implicating metabolism as a major factor in determination of plant tolerance to triazine herbicides must be considered. Perhaps the R biotype in the presence of simazine forms a weakly-bonded conjugate which is inactive in intact plants but restored to simazine during extraction. Hydrogen bonding of the hydrogen atoms of the ethylamino

groups on the simazine molecule with an unknown plant constituent might provide such a metabolite. H-bonding of the triazine molecule with a protein or enzyme involved in oxidation of water has been proposed by Good (1961). While this complex is proposed to explain the mechanism of phytotoxic action, it does suggest that such conjugants are possible in plant systems.

Montgomery and Freed (1964) also have suggested that sensitive biochemical systems may be affected in susceptible plants but not tolerant ones. Investigations have shown triazines to be potent inhibitors of photosynthesis. Plants of the resistant common groundsel biotype were observed to maintain photosynthesis at a significantly more efficient level than susceptible plants, even in the absence of simazine. This may suggest a rapid turnover rate of chlorophyll molecules in resistant plants. If so, rapid replenishment of chlorophyll at the site of toxic action might provide another explanation for the differential response of the two biotypes to simazine.

Foy (1961) has presented data indicating that storage of atrazine in lysigenous glands of tolerant varieties of cotton prevents the herbicide from reaching the site of action. Since only intact simazine was found in both common groundsel biotypes, a similar mechanism may also be suggested for tolerance of the R biotype.

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APPENDIX

Appendix Table 1. Conversions from Part Per Million (ppm) to Molar Concentration for Six s-Triazine Herbicides.

Dose (ppm)	Concentration (uM)					
	Simazine	Atrazine	Terbutryn	Prometryne	GS-14254	Prometone
30		138.89	124.48	124.28	133.33	131.31
15		72.82				
10		46.30	41.49	41.43	44.44	44.37
5		23.15				
4	19.80	18.52	16.60	16.57	17.78	17.75
3	14.85					
2	9.90	9.26	8.30	8.29	8.89	8.88
1	4.95	4.63	4.15	4.15	4.44	4.44
.5	2.48	2.31	2.07	2.07	2.22	2.22
.1	0.50	0.46	.41	.41	.44	.44
.05	0.25	0.23				

Appendix Table 2. The Dose Response of Two Common Groundsel Biotypes to Methanol.

Dose % (v/v)	% of Control							
	Susceptible Biotype				Resistant Biotype			
	I	II	III	Avg	I	II	III	Avg
1.43	64.1	103.2	106.8	91.3	63.9	59.2	71.2	64.8
.72	67.0	121.4	120.5	103.0	71.3	67.0	103.1	80.5
.14	78.8	124.7	121.6	108.4	75.9	67.7	103.1	81.6
.03	63.9	131.5	102.2	99.2	99.8	92.9	60.5	84.4
.015	76.6	91.8	112.7	93.7	98.0	114.7	79.3	97.3

Appendix Table 3. Analysis of Variance for Data in Appendix Table 2.

Source	df	SS	MS	F
Replications	2	3230.8	1615.4	4.03*
Treatments	(9)	(4414.2)	490.5	1.22
Strains	1	2281.1	2281.1	5.68*
Rates	4	1215.9	304.0	.76
Strain x Rate	4	971.2	242.8	.61
Error	18	7224.0	401.3	
Total	29	14869.0		

*Significant at 95% level

LSD_{.05} for Strains = 12.7%

CV = 22%

Appendix Table 4. Experiment 1. The Dose Response to Atrazine of a Common Groundsel Biotype Previously Reported to be Atrazine Resistant

Atrazine Rate Kg/ha	Foliage Dry Weight (mg)															
0	301	279	347	372	437	385	308	336	359	327	331	425	273	342	381	347
1.12	339	391	367	314	372	354	404	354	370	261	347	370	435	407	414	366
2.24	341	371	382	330	317	343	348	391	371	363	434	407	390	366	358	368
4.48	367	290	325	386	390	332	354	401	302	349	296	419	349	356	355	351
8.96	324	297	311	336	371	359	321	330	365	335	317	394	358	342	380	342

Appendix Table 5. Analysis of Variance for Data in Appendix Table 4.

Source of Variation	df	SS	MS	F
Treatments	4	8070.0	2017.5	1.388 n.s.
Error	70	101778.6	1453.9	
Total	74	109848.6		

LSD_{.05} = 27.8

LSD_{.01} = 36.9

CV = 2.77%

Appendix Table 6. Experiment 2. The Dry Weights of Common Groundsel from a Willamette Valley Seed Source 45 Days after an Application of Atrazine.

Atrazine Rate		Dry Weight (mg)					Avg
Kg/ha							
0	356	350	533	538	421		440
1.12	20	9	17	9	13		14
2.24	15	15	9	19	13		14
4.48	21	22	7	11	13		15
8.96	26	18	26	23	20		23

Appendix Table 7. Analysis of Variance for Data in Appendix Table 6.

Source of Variance	df	SS	MS	F
Treatment	4	716,999.8	179,249.9	105.0**
Error	20	34,137.2	1,706.9	
Total	24	751,137.0		

LSD_{.05} = 54.5

LSD_{.01} = 74.3

CV = 18.3%

Appendix Table 8. Experiment 3. The Dose Response to Simazine of Two Common Groundsel Biotypes. Treatment Date 1.

Biotype	Simazine (ppm)	Dry Weight (g)					% of Control				
		I	II	III	IV	Avg	I	II	III	IV	Avg
Susceptible	2	.421	.454	.334	.282	.375	43.5	54.5	44.8	34.8	44.4
	1	.370	.372	.329	.269	.335	38.2	44.6	42.9	33.2	39.7
	.5	.460	.291	.340	.282	.336	47.5	34.9	44.3	34.8	40.4
	.1	.494	.313	.526	.232	.391	51.0	37.6	68.6	48.0	51.3
	.05	.573	.333	.591	.483	.495	59.1	40.0	77.1	59.6	59.0
	methanol check	.969	.833	.767	.811	.845					
Resistant	4	.500	.449	.375	.294	.406	71.8	71.8	73.4	79.4	74.1
	2	.575	.490	.435	.326	.457	82.6	78.4	85.1	88.1	83.7
	1	.578	.500	.433	.434	.486	83.1	80.0	84.7	117.3	91.3
	.5	.584	.498	.459	.407	.487	83.9	79.7	89.8	110.0	90.9
	methanol check	.696	.625	.511	.370	.551					

Appendix Table 9. Analysis of Variance for Susceptible Biotype Data in Appendix Table 8.

Source of Variation	df	SS	MS	F
Replications	3	598.8	199.6	2.42
Treatments	4	1059.4	264.9	3.21 n.s.
Error	12	989.2	82.4	
Total	19	2647.5		

n.s. = nonsignificant at 5% level

CV = 19.3%

Appendix Table 10. Analysis of Variance for Resistant Biotype Data in Appendix Table 8.

Source of Variation	df	SS	MS	F
Replications	3	1089.8	363.2	7.11
Treatments	3	776.6	258.9	5.07 n.s.
Error	9	459.6	51.1	
Total	15	2326.0		

n.s. = nonsignificant at 95% level

CV = 8.4%

Appendix Table 11. Experiment 3. The Dose Response to Simazine by Two Common Groundsel Biotypes. Treatment Date 2.

Biotype	Simazine (ppm)	Dry Weight (g)					% of Control				
		I	II	III	IV	Avg	I	II	III	IV	Avg
Susceptible	2	.0970	.0711	.0744	.1678	.1026	45.7	39.4	22.2	65.2	43.1
	1	.0680	.0921	.1152	.1551	.1076	32.0	51.0	34.5	60.1	44.4
	.5	.0746	.1553	.1131	.2131	.1390	35.1	86.0	35.4	82.7	59.8
	.1	.1061	.1862	.1696	.2622	.1810	50.0	103.2	50.6	102.0	76.5
	methanol check	.2123	.1804	.3347	.2576	.2463					
	check	.2213	.3611	.5152	.6459	.4359					
Resistant	4	.0434	.0785	.0975	.1482	.0919	70.1	116.3	72.9	90.7	87.5
	3	.0529	.0664	.1342	.1369	.0976	85.1	98.4	100.2	83.9	91.9
	2	.0607	.0745	.0968	.1302	.0906	97.8	110.3	72.3	77.9	89.6
	1	.0371	.0521	.1404	.1669	.0991	59.7	77.1	104.9	02.3	86.0
	.5	.0755	.0797	.0802	.1398	.0938	121.6	118.2	59.9	85.5	96.3
	methanol check	.0621	.0672	.1338	.1633	.1067					
	check	.0923	.1225	.546	.2460	.1539					

Appendix Table 12. Analysis of Variance for Susceptible Biotype Data in Appendix Table 11.

Source of Variation	df	SS	MS	F
Replications	3	4595.7	1531.9	9.66
Treatments	4	7878.8	1969.7	12.42**
Error	12	1901.7	158.5	
Total	19	14376.3		

**Significant at 99% level

LSD_{.05} = 15.9%

LSD_{.01} = 23.9%

CV = 19%

Appendix Table 13. Analysis of Variance for Resistant Biotype Data in Appendix Table 11.

Source of Variation	df	SS	MS	F
Replications	3	1372.0	457.3	1.08
Treatment	4	2612.2	65.4	.154 n.s.
Error	12	5091.9	42.2	
Total	19	6725.6		

n. s. = nonsignificant

CV = 7.0%

Appendix Table 14. Experiment 4. The Dose Response to Atrazine by Two Common Groundsel Biotypes. Treatment Date 1.

Biotype	Atrazine (ppm)	Dry Weight (g)					% of Control				
		I	II	III	IV	Avg	I	II	III	IV	Avg
Susceptible	2	.319	.365	.242	.280	.320	30.3	36.1	33.7	50.7	37.7
	1	.419	.420	.248	.244	.333	39.8	4.15	34.5	40.6	39.1
	.5	.342	.420	.290	.239	.323	32.5	41.5	40.4	43.3	39.4
	.1	.391	.497	.522	.461	.468	41.7	47.8	61.6	57.8	52.2
	.05	.391	.497	.522	.461	.468	37.3	49.2	72.7	83.5	60.6
	methanol check	1.053	1.011	.718	.552	.834					
Resistant	30	.523	.304	.393	.266	.372	86.2	47.1	66.3	54.5	63.5
	10	.499	.374	.422	.438	.433	82.2	58.0	71.2	89.7	75.3
	4	.623	.564	.468	.422	.519	102.6	87.4	79.0	86.5	88.9
	1	.472	.513	.541	.507	.508	77.8	79.5	91.2	103.9	88.1
	.5	.687	.470	.578	.439	.544	113.2	72.9	97.5	90.0	93.4
	methanol check	.607	.645	.593	.488	.583					

Appendix Table 15. Analysis of Variance for Susceptible Biotype Data in Appendix Table 14.

Source of Variation	df	SS	MS	F
Replications	3	959.5	319.8	3.97
Treatments	4	1645.8	411.5	5.11 n.s.
Error	12	966.5	80.5	
Total	19	3571.9		

n.s. = nonsignificant at 5% level

CV = 19.6%

Appendix Table 16. Analysis of Variance for Resistant Biotype Data in Appendix Table 14.

Source of Variation	df	SS	MS	F
Replications	3	1435.7	478.6	3.63
Treatment	4	2399.7	599.9	4.56 n.s.
Error	12	1580.3	131.7	
Total	19	5415.8		

n.s. = nonsignificant at 5% level

CV = 14.0%

Appendix Table 17. Experiment 4. The Dose Response to Atrazine by Two Common Groundsel Biotypes. Treatment Date 2.

Biotype	Atrazine (ppm)	Dry Weight (g)					% of Control				
		I	II	III	IV	Avg	I	II	III	IV	Avg
Susceptible	2	.1071	.1211	.1298	.1592	.1293	22.2	27.3	40.5	29.1	29.9
	1	.0867	.1414	.1254	.1594	.1282	17.9	26.1	9.1	29.1	28.1
	.5	.1215	.1423	.0978	.1806	.1356	25.1	26.2	30.5	33.1	28.7
	.1	.1200	.1851	.1569	.2302	.1731	25.0	34.1	49.0	42.1	37.6
	.05	.1927	.2073	.2033	.2611	.2161	40.6	38.2	63.5	47.7	47.5
	methanol check	.4839	.5427	.320	.5468	.4737					
	check	.5361	.3796	.4383	.5416	.4739					
Resistant	30	.0863	.0835	.1208	.1244	.1038	57.1	49.8	58.3	57.0	55.6
	10	.0813	.0966	.1120	.1808	.1177	54.3	57.7	54.2	82.9	62.3
	4	.1084	.1320	.1434	.1650	.1372	72.4	78.7	69.4	75.6	71.7
	1	.0972	.1436	.1500	.2502	.1603	64.9	85.6	72.6	114.6	84.2
	.5	.1262	.1490	.1620	.2076	.1612	84.3	88.8	78.3	95.2	86.7
	methanol check	.1497	.1677	.2069	.2183	.1857					
	check	.1219	.1564	.2536	.2704	.2006					

Appendix Table 18. Analysis of Variance for Susceptible Biotype Data in Appendix Table 17.

Source of Variation	df	SS	MS	F
Replications	3	820.8	273.6	14.11
Treatments	4	1094.9	273.7	14.12**
Error	12	232.6	19.4	
Total	19	2148.3		

**Significant at 99% level

LSD_{.05} = 5.5%

LSD_{.01} = 8.34%

CV = 13%

Appendix Table 19. Analysis of Variance for Resistant Biotype Data in Appendix Table 17.

Source of Variation	df	SS	MS	F
Replications	3	1161.4	387.1	4.34
Treatment	4	2936.8	734.2	8.23**
Error	12	1071.0	89.3	
Total	19	5169.3		

**Significant at 99% level

LSD_{.05} = 11.9%

LSD_{.01} = 17.91%

CV = 13%

Appendix Table 20. Experiment 5. The Dose Response to Terbutryn by Two Common Groundsel Biotypes. Treatment Date 1.

Terbutryn (ppm)	Dry Weight (g)									
	Susceptible					Resistant				
	I	II	III	IV	Avg	I	II	III	IV	Avg
10	.374	.711	.636	.534	.589	.587	.608	.502	.397	.591
4	.410	.691	.516	.584	.550	.597	.606	.629	.491	.581
1	.370	.853	.599	.779	.650	.731	.557	.652	.513	.563
.5	.367	.653	.514	.581	.529	.634	.508	.714	.559	.604
.1	.577	.681	.488	.772	.630	.721	.567	.701	.642	.658
methanol check	.523	.862	.640	.820	.711	.656	.558	.568	.563	.586

Appendix Table 21. Experiment 5. The Dose Response to Terbutryn by Two Common Groundsel Biotypes. Treatment Date 1.

Terbutryn (ppm)	% of Control									
	Resistant					Susceptible				
	I	II	III	IV	Avg	I	II	III	IV	Avg
10	89.5	109.6	88.4	70.5	89.5	71.5	82.5	88.8	68.7	77.9
4	91.0	109.0	110.7	87.2	99.5	78.4	80.2	80.6	71.2	77.6
1	111.4	99.8	114.8	91.1	104.3	70.8	99.0	93.6	95.0	89.6
.5	96.7	91.0	125.7	99.3	103.2	70.2	75.8	80.3	70.9	74.3
.1	110.0	101.6	123.4	114.0	114.5	110.3	79.0	76.3	94.2	90.0

Appendix Table 22. Analysis of Variance for Data in Appendix Tables 20 and 21.

Source of Variation	df	SS	MS	F
Replications	3	759.1	265.0	2.23
Treatment	(9)	6296.2	699.6	5.89
Biotype	1	4129.0	4129.0	34.75**
Rate	4	401.9	100.5	3.71*
Error	27	3207.9	118.8	
Total	39	10299.2		

*Significant at 95% level

CV = 13.3%

**Significant at 99% level

LSD_{.01} for Biotypes = 9.5%

LSD_{.05} for Terbutryn rates = 11.2%

Appendix Table 23. Experiment 5. The Dose Response to Terbutryn by Two Common Groundsel Biotypes. Treatment Date 2.

Terbutryn (ppm)	Dry Weight (g)									
	Susceptible					Resistant				
	I	II	III	IV	Avg	I	II	III	IV	Avg
30	.205	.269	.397	.605	.369	.090	.143	.266	.257	.189
10	.194	.379	.465	.471	.377	.081	.104	.189	.244	.155
4	.223	.559	.388	.639	.452	.088	.099	.176	.282	.161
1	.247	.34	.440	.528	.390	.084	.134	.156	.309	.171
.1	.168	.422	.568	.407	.391	.076	.113	.217	.335	.185
methanol check	.285	.485	.527	.556	.463	.080	.135	.252	.309	.194
check	.335	.448	.488	.541	.453	.100	.113	.177	.268	.165

Appendix Table 24. Experiment 5. The Dose Response to Terbutryn of Two Common Groundsel Biotypes. Treatment Date 2.

Terbutryn (ppm)	% of Control									
	Resistant					Susceptible				
	I	II	III	IV	Avg	I	II	III	IV	Avg
30	112.5	105.9	105.7	83.1	101.8	72.0	55.5	75.5	73.3	69.1
10	100.7	77.2	75.3	78.8	84.0	68.2	78.3	88.2	84.8	79.9
4	110.0	73.6	69.8	91.1	86.1	81.6	115.4	73.7	115.0	96.4
1	105.2	99.6	62.0	99.8	91.6	86.8	70.8	83.6	95.0	84.1
.1	94.6	83.5	86.1	108.2	93.1	59.2	87.2	107.8	108.7	90.7

Appendix Table 25. Analysis of Variance for Data in Appendix Tables 23 and 24.

Source of Variation	df	SS	MS	F
Replications	3	774.6	258.2	.96
Treatment	(9)	3064.6	340.5	1.27
Biotype	1	535.1	535.1	2.00 n.s.
Rate	4	548.7	137.2	.51 n.s.
Biotype x Rate	4	1980.6	495.2	1.85
Error	27	7229.6	267.8	
Total	39	10394.0		

n.s. Nonsignificant at 95% level

CV = 18.6%

Appendix Table 26. Experiment 6. The Dose Response to GS-14254 of Two Biotypes of Common Groundsel. Treatment Date 1.

GS-14254 (ppm)	Dry Weight (g)									
	Susceptible					Resistant				
	I	II	III	IV	Avg	I	II	III	IV	Avg
10	.479	.208	.497	.310	.374	.494	.465	.618	.410	.497
4	.594	.465	.460	.257	.444	.545	.635	.724	.431	.584
1	.704	.400	.466	.275	.461	.700	.640	.768	.507	.654
.5	.605	.388	.575	.591	.540	.652	.522	.775	.661	.653
.1	.763	.434	.848	.633	.670	.692	.547	.652	.597	.622
methanol check	.780	.510	.946	.704	.735	.591	.681	.757	.549	.645

Appendix Table 27. Experiment 6. The Dose Response to GS-14254 by Two Biotypes of Common Groundsel. Treatment Date 1.

GS-14254 (ppm)	% of Control									
	Resistant					Susceptible				
	I	II	III	IV	Avg	I	II	III	IV	Avg
10	83.6	68.3	81.6	74.7	77.1	61.4	40.8	52.5	44.0	49.7
4	92.2	93.2	95.6	78.5	89.9	76.2	91.2	48.5	36.5	63.1
1	118.4	94.0	101.5	92.3	101.6	90.3	78.4	49.2	39.1	64.3
.5	110.3	76.7	102.5	120.4	102.5	83.6	76.1	60.8	83.9	76.0
.1	117.9	80.6	86.1	108.7	99.8	97.8	85.1	89.6	89.9	90.6

Appendix Table 28. Analysis of Variance for Data in Appendix Tables 26 and 27.

Source of Variation	df	SS	MS	F
Replication	3	1897.6	632.5	2.42
Treatment	(9)	11904.8	1322.8	5.05**
Biotype	1	6307.6	6307.6	24.09**
Rate	4	4677.2	1169.3	4.47**
Biotype x Rate	4	920.8	230.2	.88
Error	27	7069.3	261.8	
Total	39	18974.1		

**Significant at 99% level

CV = 19.9%

LSD_{.01} for biotypes = 14.2%

LSD_{.01} for GS-14254 rate = 20.4%

Appendix Table 29. Experiment 6. The Dose Response to GS-14254 by Two Biotypes of Common Groundsel. Treatment Date 2.

GS-14254 (ppm)	Dry Weight (g)									
	Susceptible					Resistant				
	I	II	III	IV	Avg	I	II	III	IV	Avg
30	.292	.257	.195	.181	.231	.214	.205	.099	.092	.153
10	.276	.257	.194	.161	.222	.233	.181	.109	.092	.154
4	.244	.281	.243	.148	.229	.215	.150	.148	.136	.162
1	.280	.260	.265	.182	.247	.277	.172	.161	.170	.195
.1	.260	.245	.332	.162	.250	.296	.211	.221	.197	.231
methanol check	.527	.388	.504	.227	.412	.307	.204	.198	.139	.212
check	.410	.597	.453	.355	.454	.347	.265	.197	.177	.246

Appendix Table 30. Experiment 6. The Dose Response to GS-14254 by Two Biotypes of Common Groundsel. Treatment Date 2.

GS-14254 (ppm)	% of Control									
	Resistant					Susceptible				
	I	II	III	IV	Avg	I	II	III	IV	Avg
30	69.8	66.0	50.3	66.2	63.1	55.4	66.3	38.7	79.7	60.0
10	75.9	88.6	55.2	66.2	71.5	52.4	66.2	38.6	71.0	57.1
4	70.0	73.4	74.8	97.8	79.0	46.3	72.3	84.3	65.4	58.1
1	90.4	84.2	81.6	122.4	94.7	53.2	67.1	52.7	80.2	63.3
.1	96.4	103.3	111.7	143.6	113.8	49.5	63.0	65.8	71.4	62.4

Appendix Table 31. Analysis of Variance for Data in Appendix Tables 29 and 30.

Source of Variation	df	SS	MS	F
Replication	3	3547.9	1191.6	10.91**
Treatment	(9)	12449.7	1383.3	12.68**
Biotype	1	5863.7	5863.7	53.70**
Rate	4	3905.0	976.2	8.94**
Biotype x Rate	4	2681.0	670.2	6.14**
Error	27	2947.7	109.2	
Total	39	18945.3		

**Significant at 99% level

LSD_{.01} for GS-14254 rate = 14.5%

LSD_{.01} for Biotypes = 9.2%

CV = 14.5%

Appendix Table 32. Experiment 7. The Dose Response to Prometryne by Two Common Groundsel Biotypes. Treatment Date 1.

Prometryne (ppm)	Dry Weight (g)									
	Resistant					Susceptible				
	I	II	III	IV	Avg	I	II	III	IV	Avg
30	.093	.066	.098	.053	.077	.106	.065	.087	.061	.079
10	.221	.155	.109	.102	.146	.118	.096	.071	.073	.089
4	.177	.131	.094	.100	.125	.157	.100	.087	.067	.102
1	.277	.192	.119	.101	.172	.171	.116	.107	.117	.127
.1	.132	.119	.132	.104	.121	.332	.227	.181	.205	.236
methanol check	.139	.096	.090	.110	.108	.451	.333	.244	.335	.340
check	.190	.188	.130	.153	.165	.293	.243	.345	.407	.322

Appendix Table 33. Experiment 7. The Dose Response to Prometryne by Two Common Groundsel Biotypes. Treatment Date 1.

Prometryne (ppm)	% of Control									
	Resistant					Susceptible				
	I	II	III	IV	Avg	I	II	III	IV	Avg
30	66.9	68.8	108.9	48.2	73.2	23.5	19.5	35.7	18.2	24.2
10	159.	161.5	121.1	92.7	133.6	26.2	28.8	29.1	21.8	26.5
4	127.3	136.5	104.4	90.9	114.8	34.8	30.	35.7	20.	30.1
1	199.3	200.	132.2	91.8	155.8	37.9	34.8	43.9	34.9	37.9
.1	95.	124.	146.7	94.6	115.1	73.6	68.2	74.2	61.2	69.3

Appendix Table 34. Analysis of Variance for Data in Appendix Tables 32 and 33.

Source of Variation	df	SS	MS	F
Replication	3	5752.0	1917.3	4.42
Treatment	(9)	85678.4	9519.8	21.94
Biotype	1	65431.9	65431.9	150.86**
Rate	4	11594.1	2898.5	6.68*
Biotype x Rate	4	8652.4	2163.1	19.95
Error	27	11710.9	433.7	
Total	39	103141.3		

*Significant at 95% level

CV = 26.7%

**Significant at 99% level

LSD_{.01} for Biotype = 18.2%

LSD_{.05} for Prometryne rate = 21.3%

Appendix Table 35. Experiment 7. The Dose Response to Prometryne by Two Common Groundsel Biotypes. Treatment Date 2.

Prometryne (ppm)	Dry Weight (g)									
	Resistant					Susceptible				
	I	II	III	IV	Avg	I	II	III	IV	Avg
30	.172	.232	.377	.150	.232	.218	.193	.182	.179	.193
10	.238	.207	.321	.095	.215	.227	.220	.139	.087	.168
4	.277	.248	.205	.132	.215	.215	.213	.100	.166	.173
1	.285	.075	.275	.142	.194	.239	.222	.155	.190	.201
.1	.327	.260	.222	.121	.232	.429	.358	.210	.309	.326
methanol check	.361	.128	.218	.095	.200	.337	.340	.384	.347	.352
check	.468	.169	.378	.211	.306	.489	.530	.516	.341	.469

Appendix Table 36. Experiment 7. The Dose Response to Prometryne by Two Common Groundsel Biotypes. Treatment Date 2.

Prometryne (ppm)	% of Control									
	Resistant					Susceptible				
	I	II	III	IV	Avg	I	II	III	IV	Avg
30	47.7	181.3	172.9	163.	141.2	64.7	56.8	47.4	51.6	55.1
10	65.9	161.7	147.3	103.3	119.6	67.4	64.7	36.2	25.1	48.4
4	76.7	193.8	94.0	143.5	127.0	63.8	62.7	26.0	47.8	50.1
1	79.0	58.6	126.2	154.4	104.6	70.9	65.3	40.4	54.8	57.9
.1	90.6	203.1	101.8	131.5	131.8	127.3	105.3	54.7	89.1	94.1

Appendix Table 37. Analysis of Variance for Data in Appendix Tables 35 and 36.

Source of Variation	df	SS	MS	F
Replication	3	8978.4	2992.8	2.24
Treatment	(9)	49166.2	5462.9	4.08
Biotype	1	40487.8	40487.8	30.27**
Rate	4	5308.5	1327.1	.99
Biotype x Rate	4	3369.9	842.5	.63
Error	27	36111.4	1337.5	
Total	39	94256.0		

**Significant at 99% level

LSD_{.01} for Biotype = 32.0%

CV = 39.3%

Appendix Table 38. Experiment 8. The Dose Response to Prometone by Two Biotypes of Common Groundsel. Treatment Date 1.

Prometone (ppm)	Dry Weights (g)									
	Resistant					Susceptible				
	I	II	III	IV	Avg	I	II	III	IV	Avg
30	.075	.054	.043	.036	.052	.119	.103	.052	.028	.075
10	.061	.099	.042	.067	.067	.066	.101	.046	.072	.071
4	.102	.112	.067	.072	.088	.082	.053	.065		.066
1	.116	.099	.069	.064	.087	.169	.121	.031	.051	.093
.1	.160	.095	.066	.118	.109	.198	.137	.223	.084	.160
methanol check	.195	.104	.065	.180	.136	.282	.162	.127	.093	.166
check	.191	.138	.071	.121	.130	.317	.223	.215	.129	.221

Appendix Table 39. Experiment 8. The Dose Response to Prometone by Two Biotypes of Common Groundsel. Treatment Date 1.

Prometone (ppm)	% of Control									
	Resistant					Susceptible				
	I	II	III	IV	Avg	I	II	III	IV	Avg
30	38.5	51.9	66.2	20.0	44.2	42.2	63.6	40.9	30.1	44.2
10	31.3	95.2	64.6	37.2	57.1	23.4	62.4	36.2	77.4	49.9
4	52.3	107.7	103.1	40.0	75.8	29.1	32.7	51.2	37.3	37.6
1	59.5	95.2	106.2	35.6	74.1	59.9	74.7	24.4	54.8	53.5
.1	82.1	91.4	101.5	65.6	85.2	70.2	84.6	75.6	90.3	80.2

Appendix Table 40. Analysis of Variance for Data in Appendix Tables 38 and 39.

Source of Variation	df	SS	MS	F
Replication	3	5519.4	1839.8	5.00**
Treatment	(9)	10583.6	1176.0	3.20**
Biotype	1	2017.8	2017.8	5.48*
Rate	4	6656.3	1664.1	4.52**
Biotype x Rate	4	1909.5	477.4	1.30
Error	27	9932.5	376.9	
Total	39	26035.5		

*Significant at 95% level

CV = 31.9%

**Significant at 99% level

LSD_{.05} for Biotypes = 12.6%

LSD_{.01} for Prometone rates = 26.6%

Appendix Table 41. Experiment 8. The Dose Response to Prometone by Two Common Groundsel Biotypes. Treatment Date 2.

Prometone (ppm)	Dry Weights (g)									
	Resistant					Susceptible				
	I	II	III	IV	Avg	I	II	III	IV	Avg
30	.139	.130	.090	.070	.107	.216	.146	.105	.073	.135
10	.158	.113	.007	.109	.114	.238	.207	.126	.115	.171
4	.337	.232	.078	.194	.210	.201	.201	.137	.076	.153
1	.259	.190	.087	.206	.185	.229	.159	.109	.118	.153
.1	.259	.143	.092	.186	.170	.348	.217	.156	.250	.242
methanol check	.284	.360	.089	.201	.233	.364	.331	.295	.315	.326
check	.338	.266	.195	.240	.260	.498	.433	.371	.445	.437

Appendix Table 42. Experiment 8. The Dose Response to Prometone by Two Common Groundsel Biotypes. Treatment Date 2.

Prometone (ppm)	% of Control									
	Resistant					Susceptible				
	I	II	III	IV	Avg	I	II	III	IV	Avg
30	48.9	36.1	101.1	34.8	55.2	59.3	44.1	35.6	23.2	40.6
10	55.6	31.4	86.5	54.2	56.9	65.4	62.5	42.7	36.5	51.8
4	118.7	64.6	87.6	96.5	91.8	55.2	60.7	46.4	24.1	46.6
1	91.2	52.8	97.8	102.5	86.1	62.9	48.0	37.0	37.5	46.4
.1	91.2	39.7	103.4	92.5	81.7	95.6	65.6	52.9	79.4	73.4

Appendix Table 43. Analysis of Variance for Data in Appendix Tables 41 and 42.

Source of Variation	df	SS	MS	F
Replication	3	3464.8	1154.9	3.12*
Treatment	(9)	12371.0	1374.6	3.71**
Biotype	1	5114.4	5114.4	13.81**
Rate	4	4506.4	1126.6	3.04*
Biotype x Rate	4	2750.2	687.6	1.86
Error	27	10000.0	370.4	
Total	39	25867.9		

*Significant at 95% level

**Significant at 99% level

LSD_{.01} for biotypes = 16.9%

LSD_{.05} for prometone rates = 19.7%

CV = 30.5%

Appendix Table 44. Experiment 9. The Dose Response of Corn to Atrazine. Treatment Date 1.

Atrazine (ppm)	Foliage Dry Weights (g)			Avg
	I	II	III	
30	0.79	0.81	0.84	.84
15	1.01	0.87	0.82	.90
5	1.11	0.98	0.99	1.02
methanol check	1.19	1.17	1.43	1.26
check	1.27	0.93	1.13	1.11

Appendix Table 45. Experiment 9. The Dose Response of Corn to Atrazine. Treatment Date 1.

Atrazine (ppm)	% of Check			Avg
	I	II	III	
30	66.4	69.2	58.7	64.8
15	84.9	74.4	57.3	72.1
5	93.3	83.8	67.4	81.5

Appendix Table 46. Analysis of Variance for Data in Appendix Tables 44 and 45.

Source of Variation	df	SS	MS	F
Replication	2	663.0	331.5	10.46
Rates	2	422.0	211.0	6.66 n.s.
Error	4	126.8	31.7	
Total	8	1211.8		

n.s. = not significant at 95% level

CV = 7.73%

Appendix Table 47. Experiment 9. The Dose Response of Corn to Atrazine. Treatment Date 2.

Atrazine (ppm)	Foliage Dry Weight (g)			
	I	II	III	Avg
30	0.69	0.65	0.78	.71
15	0.80	1.14	0.97	.97
5	1.39	0.89	0.68	.99
methanol check	1.57	0.87	1.19	1.21
check	1.25	0.96	1.18	1.13

Appendix Table 48. Experiment 9. The Dose Response of Corn to Atrazine. Treatment Date 2.

Atrazine (ppm)	% of Check			
	I	II	III	Avg
30	44.0	74.7	65.5	61.4
15	51.0	131.0	81.5	87.8
5	88.5	100.0	57.1	81.9

Appendix Table 49. Analysis of Variance for Data in Appendix Tables 47 and 48.

Source of Variation	df	SS	MS	F
Replication	2	2853.3	1426.7	3.02
Rates	2	1153.2	576.6	1.22 n.s.
Error	4	1889.5	472.4	
Total	8	5896.0		

n. s. = nonsignificant at 95% level

CV = 28.2%

Appendix Table 50. Experiment 10. The Photosynthetic Response of Two Common Groundsel Biotypes to Simazine. Treatment Date 1.
Simazine Removed from Susceptible Biotype after 24 Hours.

Simazine (ppm)	Exposure Time (hr)	Net CO ₂ Fixation (mg CO ₂ /gram dry foliage wt./10 min)								LSD . ₀₅	LSD . ₀₁
		Resistant				Susceptible					
		I	II	III	Avg	I	II	III	Avg		
0	0	358	371	391	373	242	287	267	266	38	64
	6	343	334	366	347	217	218	230	222	29	48
	12	394	454	414	420	240	231	260	243	54	90
	24	345	335	373	351	214	242	241	232	40	67
	30	299	297	333	310	198	236	237	224	48	80
	48	454	445	477	459	258	308	298	288	50	83
0.5	0	290	384	406	360	347	319	382	389	111	184
	6	259	346	375	327	217	221	247	228	100	166
	12	294	373	426	365	91	-24	117	62	160	266
	24	246	360	400	335	-125	-177	-139	-147	120	199
	30	235	315	376	309	-49	-79	-71	-66	116	192
	48	477	452	480	469	157	221	236	205	41	119

Appendix Table 51. Experiment 10. The Photosynthetic Response of Two Common Groundsel Biotypes to Simazine. Treatment Date 2.
Simazine Removed from Susceptible Biotype After 24 Hours.

Simazine (ppm)	Exposure Time (hr)	Net CO ₂ Fixation (mg CO ₂ /gram dry foliage wt./10 min)								LSD . ₀₅	LSD . ₀₁
		Resistant				Susceptible					
		I	II	III	Avg	I	II	III	Avg		
0	0	400	395	461	419	233	310	262	268	86	142
	6	577	292	396	421	156	186	271	204	250	415
	24	301	313	354	322	213	254	188	328	69	115
	30	529	362	396	429	196	287	234	239	159	264
	48	333	320	357	340	136	266	217	206	109	181
0.5	0	375	521	503	466	320	529	316	388	233	386
	6	314	382	448	381	-178	-225	-179	-194	115	191
	24	318	294	296	302	-251	-210	-272	-244	55	91
	30	340	346	372	352	-83	-43	-158	-95	97	161
	48	352	393	296	347	-8	-11	-67	-22	93	154

Appendix Table 52. The Sorption of Simazine by Plastic and Glass Containers as a Function of Time.

Exposure Time (hr)	Container Type			
	Plastic		Glass	
	μg Simazine Sorbed	% Simazine Sorbed	μg Simazine Sorbed	% Simazine Sorbed
.25	13.8	9.2	11.2	7.5
.50	14.0	9.4	10.0	6.7
1	14.1	9.4	8.1	5.4
3	11.6	7.8	7.9	5.3
6	13.7	9.2	8.4	5.6
18	12.4	8.3	7.9	5.2
24	12.1	8.1	7.1	4.7
48	10.4	7.0	6.3	4.2
96	13.6	9.1	5.2	3.5
192	11.4	7.6	6.8	4.5

Appendix Table 53. Experiment 11. Transpiration, Fresh and Dry Weights of Resistant and Susceptible Common Groundsel Plants Used During Simazine Absorption Study. Treatment Date 1.

Biotype	Exposure Time (hr)	Fresh Weight (g)			Dry Weight (g)			Transpiration (ml)		
		I	II	Avg	I	II	Avg	I	II	Avg
Resistant	1	5.391	4.242	4.817	.512	.390	.451	1.2	1.4	1.3
	3	4.286	3.771	4.029	.416	.380	.398	1.6	1.6	1.6
	6	3.533	3.377	3.455	.315	.327	.339	3.4	6.4	4.9
	12	3.653	3.855	3.754	.333	.384	.359	8.2	9.2	8.7
	24	4.200	3.414	3.807	.383	.309	.346	12.8	1.48	13.8
Susceptible	1	9.107	5.282	7.195	.847	.496	.672	1.2	0.8	1.0
	3	7.311	7.158	7.235	.697	.687	.692	3.8	3.0	3.4
	6	4.941	5.360	5.151	.697	.490	.594	4.4	4.8	4.6
	12	8.010	5.977	6.994	.665	.532	.599	10.4	13.6	12.0
	24	6.320	6.114	6.217	.538	.566	.522	13.2	15.6	14.4

Appendix Table 54. Experiment 11. Total Simazine Absorbed (μg) by Two Common Groundsel Biotypes as a Function of Time. Experiment Date 1.

Exposure Time (hr)	Resistant Total Simazine Absorbed (μg) ^a			Susceptible Total Simazine Absorbed (μg) ^a		
	I	II	Avg	I	II	Avg
1	1.635	4.530	3.083	5.505	1.395	3.450
3	.990	5.115	3.053	3.660	9.975	6.818
6	2.730		2.730	17.160	11.253	14.207
12	13.475	17.352	15.414	10.626	28.115	19.371
24	14.458	17.601	16.030	23.799	29.641	26.720

^a Values are averages of 4 plants/container.

Appendix Table 55. Experiment 11. Simazine Absorbed ($\mu\text{g/gfw}$) by Two Common Groundsel Biotypes as a Function of Time. Treatment Date 1.

Exposure Time (hr)	Resistant Simazine Absorbed ^a ($\mu\text{g/gfw}$)			Susceptible Simazine Absorbed ^a ($\mu\text{g/gfw}$)		
	I	II	Avg	I	II	Avg
1	.303	1.068	.656	.605	.264	.434
3	.231	1.356	.794	.501	1.394	.947
6	.773		.773	3.473	2.099	2.786
12	3.689	4.501	4.095	1.327	4.704	3.015
24	3.442	5.156	4.299	3.766	8.848	6.307

^aValues are averages of 4 plants/container.

Appendix Table 56. Experiment 11. Simazine Absorbed ($\mu\text{g/gdw}$) by Two Common Groundsel Biotypes as a Function of Time. Treatment Date 1.

Exposure Time (hr)	Resistant Simazine Absorbed ^a ($\mu\text{g/gdw}$)			Susceptible Simazine Absorbed ^a ($\mu\text{g/gdw}$)		
	I	II	Avg	I	II	Avg
1	3.191	11.615	7.403	6.502	2.813	4.657
3	2.380	13.461	7.920	5.251	14.520	9.885
6	7.778		7.778	24.620	22.965	23.793
12	40.465	45.188	42.826	15.979	52.848	34.413
24	37.749	56.961	47.355	44.200	52.370	48.285

^aValues are averages of 4 plants/container.

Appendix Table 57. Analysis of Variance Table for Total Simazine Absorption Data Presented in Appendix Table 54.

Source of Variation	df	SS	MS	F
Replication	1	56.68	56.68	3.08
Treatments	9	1281.80	142.42	7.74**
Strains	1	183.09	183.09	9.96**
Times	4	1005.84	251.46	13.67**
Strains x Times	4	92.87	23.18	1.26
Error	10	183.94	18.39	
Total	19	1522.78		

**Significant differences at 99% level CV = 38.67%

LSD_{.01} TMTS = 13.59

Appendix Table 58. Analysis of Variance Table for Simazine Absorption Data (ug/gfw) Presented in Appendix Table 55.

Source of Variation	df	SS	MS	F
Replication	1	7.31	7.31	8.12*
Treatments	9	78.87	7.76	9.73**
Strains	1	1.60	1.60	1.78
Times	4	63.53	15.88	17.64**
Strains x Times	4	7.67	1.92	2.13
Error	10	9.00	0.90	
Total	19	95.49		

*Significant differences at 95 % level

CV = 39.36%

LSD_{.01} = 9.51

**Significant differences at 99 % level

LSD_{.05} = 6.68

Appendix Table 59. Analysis of Variance Table for Simazine Absorption Data (ug/gdw) Presented in Appendix Table 56.

Source of Variation	df	SS	MS	F
Replication	1	426.93	426.93	4.04
Treatments	9	5930.85	658.98	6.24**
Strains	1	12.02	12.02	0.11
Times	4	5591.34	1397.80	13.23**
Strains x Times	4	327.49	81.87	0.77
Error	10	1056.75	105.68	
Total	19	6987.60		

**Significant differences at 99 % level

CV = 43.88%

LSD_{.01} = 28.67

Appendix Table 60. Experiment 11. Transpiration, Fresh and Dry Weights of Resistant and Susceptible Common Groundsel Plants Used During Simazine Absorption Study. Treatment Date 2.

Biotype	Exposure Time (hr)	Fresh Weight (g)			Dry Weight (g)			Transpiration (ml)		
		I	II	Avg	I	II	Avg	I	II	Avg
Resistant	1	7.0395	9.4494	8.2445	.6194	.7608	.6901	3.5	3.5	3.50
	3	8.2739	11.9525	11.1132	.6916	.9551	.8234	6.4	8.9	7.65
	6	11.6165	8.5600	10.0883	.9391	.7715	.8553	15.3	12.0	13.65
	12	9.7801	8.1014	8.9408	.8367	.6914	.7641	18.2	16.8	17.50
	24	9.9607	11.4389	10.6998	.8289	.9141	.8715	37.5	41.3	39.40
Susceptible	1	13.9650	15.4933	14.7292	1.1803	1.2503	1.2153	3.5	3.7	3.60
	3	15.4679	15.3177	15.3928	1.3128	1.1993	1.2561	10.4	11.1	10.75
	6	13.6912	11.9111	12.8012	1.1451	.9726	1.0589	10.0	15.1	17.05
	12	16.9637	18.9103	17.9370	1.4412	1.4440	1.4426	28.6	31.7	30.15
	24	16.7151	13.9305	15.3228	1.4624	1.1400	1.3012	57.4	60.7	50.05

Appendix Table 61. Experiment 11. Total Simazine Absorbed (μg) by Two Common Groundsel Biotypes as a Function of Time. Treatment Date 2.

Exposure Time (hr)	Resistant			Susceptible		
	Total Simazine Absorbed ^a (μg)			Total Simazine Absorbed ^a (μg)		
	I	II	Avg	i	ii	Avg
1		6.96	6.96	7.28	9.50	8.39
3	9.44	20.31	14.88	16.89	18.59	17.74
6	20.54	23.94	22.24	45.15	33.27	39.20
12	38.09	32.24	35.17	62.07	55.16	58.62
24	71.75	74.75	73.25	92.52	113.00	102.76

^aValues are averages of 5 plants/container.

Appendix Table 62. Experiment 11. Simazine Absorbed ($\mu\text{g/gfw}$) by Two Common Groundsel Biotypes as a Function to Time. Treatment Date 2.

Exposure Time (hr)	Resistant			Susceptible		
	Simazine Absorbed ^a ($\mu\text{g/gfw}$)			Simazine Absorbed ^a ($\mu\text{g/gfw}$)		
	I	II	Avg	I	II	Avg
1		0.74	0.74	0.52	0.61	0.57
3	1.14	1.70	1.42	1.09	1.21	1.15
6	1.77	2.80	2.29	3.30	2.79	3.05
12	3.90	3.98	.94	3.66	2.92	3.29
24	7.20	6.54	6.87	5.54	8.11	6.83

^aValues are averages of 5 plants/container.

Appendix Table 63. Experiment 11. Simazine Absorbed ($\mu\text{g/gdw}$) by Two Common Groundsel Biotypes as a Function of Time. Treatment Date 2.

Exposure Time (hr)	Resistant ^a Simazine Absorbed ($\mu\text{g/gdw}$)			Susceptible ^a Simazine Absorbed ($\mu\text{g/gdw}$)		
	I	II	Avg	I	II	Avg
1		9.15	9.15	6.17	7.60	6.89
3	13.65	21.27	17.46	12.87	15.50	14.19
6	21.87	31.00	26.43	39.43	34.21	36.82
12	45.52	46.63	46.08	43.09	38.20	40.64
24	86.56	81.77	84.17	63.27	99.12	81.40

^aValues are averages of 5 plants/container.

Appendix Table 64. Analysis of Variance Table for Total Simazine Absorption Data Presented in Appendix Table 61.

Source of Variation	df	SS	MS	F
Replication	1	14.50	14.50	.38
Treatments	9	17809.36	1978.80	52.07**
Strains	1	1101.87	1101.90	29.00**
Times	4	16090.38	4022.60	105.86
Strains x Times	4	617.11	154.30	4.06*
Error	10	380.04	38.00	
Total	19	18203.90		

*Significant differences at 95 % level

**Significant differences at 99% level

CV = 16.03%

LSD_{.05} = 13.73 µg

LSD_{.01} = 19.53 µg

Appendix Table 65. Analysis of Variance Table for Simazine Absorption Data (µg/gfw) Presented in Appendix Table 62.

Source of Variation	df	SS	MS	F
Replication	1	0.32	0.32	0.75
Treatments	9	96.16	10.68	24.96**
Strains	1	0.03	.03	.07
Times	4	94.06	23.76	55.52
Strains x Times	4	1.08	0.27	0.63
Error	10	4.28	.43	
Total	19	100.76		

**Significant Differences at 99 % level

CV = 21.7%

LSD_{.01} = 2.08 µg/gfw

Appendix Table 66. Analysis of Variance Table for Simazine Absorption Data ($\mu\text{g/gdw}$) Presented in Appendix Table 63.

Source of Variation	df	SS	MS	F
Replication	1	90.19	90.19	1.38
Treatments	9	13965.60	1551.73	23.81**
Strains	1	2.25	2.25	0.04
Times	4	1380.17	345.04	5.30*
Strains x Times	4	158.61	39.65	0.61
Error	10	651.46	65.16	
Total	19	14707.24		

*Significant differences at 95% level

**Significant differences at 99% level

CV = 22.2%

LSD_{.05} = 17.98 $\mu\text{g/gfw}$

LSD_{.01} = 25.58 $\mu\text{g/gfw}$

Appendix Table 67. Foliage Fresh Weights (g) of Two Common Groundsel Biotypes for Simazine Metabolism Study (Experiment 12; Study A).

Treatment Time (hr)	Susceptible			Resistant		
	I	II	Avg	I	II	Avg
24	4.65	6.50	5.57	4.90	5.64	5.27
48	6.72	7.74	7.23	4.52	4.52	4.52
72	10.58	6.63	8.60	4.05	3.83	3.94

Appendix Table 68. Root Fresh Weights (g) of Two Common Groundsel Biotypes for Simazine Metabolism Study. (Experiment 12; Study A)

Treatment Time (hr)	Susceptible			Resistant		
	I	II	Avg	I	II	Avg
24	2.97	3.00	2.98	1.98	2.48	2.23
48	3.62	3.74	3.68	1.81	1.65	1.73
72	3.93	2.87	3.40	2.08	1.69	1.88

Appendix Table 69. Total Fresh Weights (g) of Two Common Groundsel Biotypes for Simazine Metabolism Study. (Experiment 12; Study A)

Treatment Time (hr)	Susceptible			Resistant		
	I	II	Avg	I	II	Avg
24	7.62	9.50	8.56	6.88	8.12	7.50
48	10.34	11.48	10.91	6.33	6.17	6.25
72	14.56	9.50	12.03	6.13	5.11	5.62

Appendix Table 70. The Recovery of ^{14}C -Activity From Two Common Groundsel Biotypes After Treatment With ^{14}C -Simazine for 24 Hours. After 24 hours of initial ^{14}C -simazine uptake all plants were incubated in $5 \times 10^{-4}\text{M}$ calcium nitrate for 0, 24, 48 hours. (Experiment 12, Study A)

Biotype	Treatment Time (hr)	^{14}C Recovered								
		(dpm)			(dpm)			(dpm)		
		Initial Activity			Experimental Solution + Rinse			Plant		
		I	II	Avg	I	II	Avg	I	II	Avg
Susceptible	24	1216950	1223737	1220343	831210	466975	649092	297461	766436	531948
	48	1222185	1215510	1218847	692775	619350	656062	222081	529823	375952
	72	1167757	1220857	1194307	351975	740625	546300	405165	250889	328027
Resistant	24	1179225	1217880	1198552	761360	784565	772967	337159	358971	348065
	48	1228492	1151632	1190062	748725	700425	724565	356797	286988	321892
	72	1180552	1208017	1194284	550575	760575	655575	249009	288854	268931

Biotype	Treatment Time (hr)	^{14}C Recovered						
		% Recovered			% Absorbed by Plant/24 hr.			
		I	II	Avg	I	II	Avg	
Susceptible	24	93	101	97	32	62	47	
	48	75	95	85	43	49	46	
	72	65	81	73	70	39	54	
Resistant	24	93	94	93	35	36	35	
	48	90	86	88	39	39	39	
	72	68	87	78	53	37	45	

Appendix Table 71. The Distribution of ^{14}C -Activity in the Extracts of Foliage and Roots of Two Common Groundsel Biotypes as a Function of Time.
(Experiment 12; Study A).

Biotype	Treatment Time (hr)	% of ^{14}C -Recovered in Foliage						% of ^{14}C -Recovered in Roots					
		Chloroform			Water			Chloroform			Water		
		I	II	Avg	I	II	Avg	I	II	Avg	I	II	Avg
Susceptible	24	80	94	87	5	4	4	13	3	8	1	1	1
	48	83	89	86	10	8	9	5	3	4	2	1	1.5
	72	83	80	81	13	16	14	2	2	2	1	2	1.5
Resistant	24	87	87	87	5	4	4	1	3	2	1	1	1
	48	87	86	86	11	10	10	1	2	1.5	1	1	1
	72	93	92	92	4	3	3	3	1	2	2	1	1.5

Appendix Table 72. Analysis of Variance for Data in Appendix Table 71. % ¹⁴C-Recovered in Foliage Chloroform and Water Extracts (Study A).

Source of Variation	df	SS	MS	F
Biotype	1	.66	.66	.06 n. s.
Time	2	27.00	13.50	1.24 n. s.
Extracts	1	37446.00	37446.00	3456.55 **
Biotype x Time	2	1.33	.66	.06
Biotype x Extracts	1	73.50	73.50	6.78
Time x Extracts	2	39.00	19.50	1.80
Biotype x Time x Extracts	2	169.00	84.50	7.80
Error	12	130.00	10.83	
Total	23	37886.50		

**Significant at 99% level

LSD for Extracts = 4.1%

Appendix Table 73. Foliage Fresh Weights (g) of Two Common Groundsel Biotypes Used During Simazine Metabolism Study. (Experiment 12; Study B)

Treatment Time (hr)	Susceptible				Resistant			
	I	II	III	Avg	I	II	III	Avg
24	8.94	8.01	7.97	8.31	7.47	6.68	8.43	7.53
48	8.44	8.23	9.13	8.60	6.24	7.73	9.87	7.61
96	8.43	8.65	6.92	8.00	7.97	7.55	5.73	7.08

Appendix Table 74. Root Fresh Weights (g) of Two Common Groundsel Biotypes Used During Simazine Metabolism Study. (Experiment 12; Study B)

Treatment Time (hr)	Susceptible				Resistant			
	I	II	III	Avg	I	II	III	Avg
24	1.86	1.94	2.33	2.04	2.22	1.92	2.14	2.09
48	2.06	2.47	2.16	2.23	2.51	2.24	2.32	2.36
96	2.15	2.71	2.56	2.47	3.03	2.37	2.05	2.48

Appendix Table 75. Total Fresh Weights (g) of Two Common Groundsel Biotypes Used During Simazine Metabolism Study. (Experiment 12; Study B)

Treatment Time (hr)	Susceptible				Resistant			
	I	II	III	Avg	I	II	III	Avg
24	10.80	9.95	10.30	10.35	9.69	8.60	10.57	9.62
48	10.50	10.70	9.29	10.16	8.75	8.97	10.19	9.97
96	10.58	11.36	9.48	10.47	11.00	9.92	7.78	9.57

Appendix Table 76. The Recovery of ^{14}C -Activity From Two Common Groundsel Biotypes After Treatment with ^{14}C -Simazine for 24 Hours. After 24 hours of initial ^{14}C -simazine uptake, all plants were incubated in 2.0 ppm simazine for 0, 24, or 72 hours. (Experiment 12; Study B)

Biotype	Treatment Time (hr)	^{14}C -Recovered											
		Initial Activity (dpm)				Experimental + Rinse Solution (dpm)				Plants (dpm)			
		I	II	III	Avg	I	II	III	Avg	I	II	III	Avg
Susceptible	24	660097	693429	656082	669869	387750	3876916	343050	369239	267621	239607	245248	250758
	48	651140	624186	572950	616092	397575	393343	335250	375389	210504	241044	212621	221390
	96	532586	537247	494904	521579	292095	281325	260250	277890	171722	255135	109378	178745
Resistant	24	647146	617945	649800	638297	307800	414847	406275	376305	222357	177802	187954	196038
	48	648654	601628	530030	600104	395550	385350	349825	343575	193341	171081	231578	198670
	96	508439	481082	500124	496548	314100	245475	292500	284025	136090	152230	105440	131253

Biotype	Treatment Time (hr)	^{14}C -Recovered							
		% Recovered				% Absorbed by Plant/ 24 hr			
		I	II	III	Avg	I	II	III	Avg
Susceptible	24	99	89	90	93	41	45	48	45
	48	93	101	96	97	39	37	41	39
	96	87	100	75	87	64	48	47	53
Resistant	24	82	96	91	90	52	33	37	41
	48	95	93	88	92	39	36	54	43
	96	89	83	80	84	38	49	42	43

Appendix Table 77. The Distribution of ^{14}C -Activity in the Extracts of Foliage and Roots of Two Common Groundsel Biotypes as a Function of Time (Experiment 12; Study B).

Biotype	Treatment Time (hr)	% of ^{14}C -Recovered in Foliage								% of ^{14}C -Recovered in Roots							
		Chloroform				Water				Chloroform				Water			
		I	II	III	Avg	I	II	III	Avg	I	II	III	Avg	I	II	III	Avg
Susceptible	24	74	81	83	79	14	8	6	9	6	7	8	7	2	2	3	2
	48	82	79	81	81	12	16	13	14	3	2	2	2	2	2	3	2
	96	77	90	81	83	18	8	15	14	3	1	1	2	1	0	2	1
Resistant	24	77	70	70	72	10	13	15	13	9	13	12	12	1	1	4	2
	48	86	76	84	82	9	18	11	13	3	3	2	3	2	2	2	2
	96	82	86	81	83	13	11	15	13	2	1	2	2	1	1	1	1

Appendix Table 78. Analysis of Variance for Data in Appendix Table 77. % ¹⁴C Recovered in Foliage Chloroform and Water Extracts (Study B).

Source of Variation	df	SS	MS	F
Biotype	1	4.69	4.69	.28 n.s.
Time	2	156.22	78.11	4.71 *
Extracts	1	41141.36	41141.36	2480.88 **
Biotype x Time	2	6.22	3.11	.18
Biotype x Extracts	1	14.69	14.69	.88
Time x Extracts	2	38.88	19.44	1.17
Biotype x Time x Extracts	2	69.55	34.77	2.09
Error	24	398.00	16.58	
Total	35	41829.63		

*Significant at 95% level

**Significant at 99% level

LSD for Time = 4.8%

LSD for Extracts = 3.8%

Appendix Table 79. The Rf Value of ¹⁴C-Simazine.

I	I	II	II	III	III	Avg
.36	.34	.37	.33	.33	.37	.35

Appendix Table 80. The Rf Value of ¹⁴C Isolated from Foliage of Two Common Groundsel Biotypes (Study A).

Treatment Time (hr)	Resistant					Susceptible					*LSD .05
	I	I	II	II	Avg	I	I	II	II	Avg	
24	.30	.30	.33	.29	.31	.32	.30	.32	.28	.31	.03
48	.27	.32	.33	.35	.32	.29	.30	.33	.38	.33	.04
72	.25	.30	.30	.28	.28	.26	.27	.31	.32	.29	.04

*Comparison made to mean Rf value in Appendix Table 79.

Appendix Table 81. Analysis of Variance for Data in Appendix Table 80.

Source of Variation	df	SS	MS	F
Biotype	1	.00015	.00015	.204
Time	2	.0049	.0025	3.33
Error	20	.015	.0007	
Total	23	.020		

Appendix Table 82. Experiment 12. The Rf Values of ^{14}C Isolated from Foliage of Two Common Groundsel Biotypes (Study B).

Treatment Time (hr)	Resistant							Susceptible							*LSD .05
	I	I	II	II	III	IV	Avg	I	I	II	III	III	III	Avg	
24	.33	.33	.35	.36	.40	.40	.36	.31	.32	.33	.34	.38	.36	.34	.03
48	.37	.32	.27	.32	.5	.39	.4	.34	.30	.31	.32	.35	.23	.33	.04
96	.39	.45	.34	.44	.32	.37	.39	.34	.39	.32	.32	.27	.37	.34	.05

*Comparisons made to mean Rf value in Appendix Table 79.

Appendix Table 83. Experiment 12. The Rf Value of ^{14}C Isolated from Roots of Two Common Groundsel Biotypes after 24 Hours (Study B).

	Resistant							Susceptible							*LSD .05
	I	I	II	II	III	III	Avg	I	I	II	II	III	III	Avg	
	.33	.35	.33	.32	.27	.32	.32	.31	.34	.33	.32	.33	.28	.32	.03

*Comparison made with mean Rf value in Appendix Table 79.

Appendix Table 84. Analysis of Variance for Data in Appendix Table 82.

Source of Variation	df	SS	MS	F
Biotype	1	.0059	.0059	4.30
Time	2	.0068	.0034	2.50
Error	32	.0437	.0013	
Total	35	.0565		

CV = 10.3%

Appendix Table 85. Analysis of Variance for Data in Appendix Table 83.

Source of Variation	df	SS	MS	F
Biotype	1	.000008	.000008	.01 n.s.
Error	10	.00588	.00059	
Total	11	.00589		

CV = 7.5%

Appendix Table 86. The Recovery of ^{14}C -Activity from Foliage and Root Residue of Two Common Groundsel Biotypes as a Function of Time. After 24 Hours of Initial ^{14}C -Simazine Uptake all Plants were Incubated in $5 \times 10^{-4}\text{ M}$ Calcium Nitrate for 0, 24, or 48 Hours. (Study A).

Treatment Time (hr)	% ^{14}C Recovered in Foliage Residue						% ^{14}C Recovered in Root Residue					
	Resistant			Susceptible			Resistant			Susceptible		
	I	II	Avg	I	II	Avg	I	II	Avg	I	II	Avg
24	0.1	0.1	0.1	0.2	0.1	0.2	0.1	0.1	0.1	0.1	0.1	0.1
48	0.3	0.2	0.3	0.1	-	0.1	0.1	-	0.1	0.1	-	0.1
72	0.1	0.3	0.2	-	0.1	0.1	-	-	-	-	-	-

Severe quenching noted for all values

Appendix Table 87. The Recovery of ^{14}C -Activity from Foliage and Root Residue of Two Common Groundsel Biotypes as a Function of Time. After 24 Hours of Initial ^{14}C -Simazine Uptake All Plants Were Incubated in 2.0 ppm Simazine for 0, 24, or 72 Hours (Study B).

Treatment Time (hr)	% ^{14}C Recovered in Foliage Residue								% ^{14}C Recovered in Root Residue							
	Resistant				Susceptible				Resistant				Susceptible			
	I	II	III	Avg	I	II	III	Avg	I	II	III	Avg	I	II	III	Avg
24	3.7	3.0	0.6	2.4	2.6	2.9	0.4	2.0	0.3	0.1	0.3	0.2	0.1	0.1	0.1	0.1
48	1.1	0.7	0.7	0.8	0.7	0.6	1.2	0.8	0.2	0.3	0.3	0.3	0.3	0.3	0.1	0.2
96	1.6	0.3	0.3	1.1	2.6	1.1	1.4	1.7	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1

Appendix Table 88. Fresh Weight of Corn Plants Used During Experiment 13.
The Metabolism of Simazine by Corn (*Zea mays* L.)

Plant Part	I	II	III	Avg
Foliage	3.84	3.59	3.00	3.48
Roots	2.62	2.43	1.71	2.25
Total	6.46	6.02	4.71	5.73

Appendix Table 89. The Recovery of ¹⁴C-Activity from Corn After Treatment with ¹⁴C-Simazine for 72 Hours.

Initial (dpm)				¹⁴ C Recovered Experimental + Rinse Solution (dpm)				Plant (dpm)			
I	II	III	Avg	I	II	III	Avg	I	II	III	Avg
288798	254136	361679	334871	79578	87865	99424	88956	154231	193350	241095	196225
% Recovered				% Absorbed							
I	II	III	Avg	I	II	III	Avg				
80.6	79.4	94.1	84.7	72.5	75.2	72.5	73.4				

Appendix Table 9Q The Distribution of ¹⁴C-Activity in Foliage and Roots of Corn after Treatment with ¹⁴C-Simazine for 72 Hours.

Plant Part	^a % ¹⁴ C-Activity Recovered in:											
	Chloroform				Water				Residue			
	I	II	III	Avg	I	II	III	Avg	I	II	III	Avg
Foliage	17	24	24	21	25	33	32	30	0.2	0.3	0.2	0.2
Root	18	18	16	17	40	25	28	31	0.3	0.3	0.2	0.3
Total	35	42	40	38	65	58	60	61	0.5	0.6	0.4	0.5

^aValues calculated as percent of ¹⁴C-Activity recovered from plants.

Appendix Table 91. Analysis of Variance for Data in Appendix Table 90. % ¹⁴C Distribution in Chloroform and Water Extracts of Corn Foliage and Roots.

Source of Variation	df	SS	MS	F
Plant Part	1	363.00	363.00	14.81**
Extracts	1	8.33	8.33	.34
Error	9	220.67	24.52	
Total	11	59.20		

**Significant at 99% level

LSD_{.01} for parts = 8.1%

CV = 16.5%