

THE INFLUENCE OF HERBICIDES ON SELECTED GROUND COVER PLANTS

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ABSTRACT

THE INFLUENCE OF HERBICIDES ON SELECTED GROUND COVER PLANTS

By Donald Bernard Carlson

Weed control in ground cover plantings has been a problem, both economically and aesthetically, to commercial nurserymen and homeowners alike. Though the cost factor is more important to the former, the labor involved for hand weeding practices, is just as tedious to the latter. With this in mind, the following investigations were conducted on the effectiveness of pre- and postemergence applications of herbicides on new and established ground cover plantings. At the same time, studies were made to gain a better understanding of triazine and phenylurea induced chlorosis. An elucidation of this mechanism would eliminate potential herbicide users fear of such injury.

Pre-emergence application of eight chemicals on new plantings, and postemergence application of nine chemicals on simulated established plantings were made, and subsequently rated for weed control, and the occurrance of injury on the ground cover species.

Simazine, at two pounds per acre, diuron at two pounds per acre, and simazine plus paraquat at two plus one-eighth pound per acre, respectively, gave the best results on new plantings. Postemergence applications of linuron at two and four pounds per acre gave good weed control, but ground cover burning was so severe that these treatments would be commercially unacceptable.

Greenhouse experiments were conducted to study the chlorosis problem. Treatments were made with herbicide solutions at concentrations ranging from 0.25 to 40 ppm.

There was no true correlation between the solubility of a herbicide and the type of chlorosis caused. In general, herbicides of higher solubility cause an interveinal chlorosis, whereas, those of lower solubility induce veinal chlorosis. This leads to the hypothesis that the availability of a herbicide, as influenced by its water solubility as well as the mode of action, will influence the type of chlorotic injury induced.

THE INFLUENCE OF HERBICIDES ON SELECTED GROUND COVER PLANTS

Ву

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A THESIS

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INTRODUCTION

"In the beginning God created the heavens and the earth. And God said, 'Let the earth put forth vegetation, plants yielding seed, and fruit trees bearing fruit in which is their seed, each according to its kind, upon the earth.' And it was so. The earth brought forth vegetation, plants yielding seed, each according to their own kinds." (1) And so it was that part of the vegetation was weeds, the curse of the agrarian societies.

Weeds have plagued man since earliest time, and their control has presented a challenge that often seems insurmountable. One area where the control of weeds has attracted the interest of many people is in ornamental plantings. Great progress has been made with the use of herbicides in this area since 1955. But many plantsmen are still reluctant to reap the harvest of advantages offered by this control method. One of the major reasons for this hesitation has been the fear of injury that can result from the misuse of herbicides. Increased research since 1959, and publications of the results, has brought about a phenominal growth in the number of users of chemical control measures, but still little has been done to investigate the basis of herbicide induced injury. Unusual injury symptoms such as veinal and interveinal chlorosis have been noted with the use of herbicides having almost identical modes of action. In addition to this, the area of weed control in new and established ground cover plantings still presents a problem.

This study was undertaken to evaluate herbicides for weed control in new and established ground cover plantings, to determine the tolerance of various ground cover species to herbicides, and as a preliminary study of herbicide induced chlorosis. The names, terms and abbreviations used throughout this thesis in reference to chemical materials are those adapted by the Weed Society of America, as reported in Weeds, Volume 10, Number 3, July, 1962.

REVIEW OF LITERATURE

Herbicide History

Weed control has progressed through the ages from hands and hoe to cultivator and plow. The present use of chemicals, however, marks the outstanding technological advancement in this field. One of the first records of the use of chemical materials to kill weeds was around the latter 1890's, when Bonnett in France, Schultz in Germany, and Bolley in the United States, all working independently, found that solutions of copper salts would selectively kill broadleaved weeds in cereals. In 1908 Bolley found that table salt, iron sulfate, copper and sodium arsenite would successfully control weeds in wheat. These discoveries were followed by Pokorny's chemical synthesis techniques for 2,4-dichlorophenoxyacetic acid, and its later application as a growth regulator and selective herbicide by workers such as Zimmerman and Hitchcock (30), Blackman (6), and Hamner and Tukey (15, 16).

Though herbicides are a relatively new tool in man's fight to control plant competition in his environment, if used properly they can often do the job better and more economically than other methods.

Weed Damage

Poor growth of ground cover plantings due to weeds is a factor worthy of note when considering control. In new plantings that become infested with weeds, the necessity for cultivation and hoeing reduces the nurseries efficiency since such practices increase cost. Furthermore, damage to the

Excerpt from Klingman (18) page 9.

²Portions taken from Klingman (18) pages 9 and 10.

roots of wanted plants is always a risk associated with cultivation. Of great importance is the fact that many ground cover plants at their start do not effectively compete against heavy weed growth. One of the major purposes of ground covers is to form a thick mat preventing weed growth. Thus, the necessity for getting vigorously growing plants as soon as possible is obvious. By eliminating weeds you eliminate competition for factors, such as: soil moisture and soil nutrients, so important for the growth of a plant.

In 1960 it was reported that good weed control had been obtained with 2-chloro-4, 6-bis(ethylamino)-s-triazine, (simazine), and 2-chloro-4-ethylamino-6-isopropylamino-s-triazine, (atrazine), used on ornamentals at rates up to ten pounds of active ingredient per acre (21). The results of these experiments, conducted over a three year period from 1957 to 1959, showed that injury to most nursery crops was negligible, but injury symptoms, including yellowing of the follage, occurred in newly planted and heavily watered Pachysandra and other ground covers. Light infestations of quackgrass, (Agropyron repens), and seedlings of annual weeds beyond the initial stage of growth occurred in the treated plots. It is interesting to note that when these plants were fertilized weekly throughout the summer, they regained their green color.

Birdsell, et. al. (5) did evaluation tests on five herbicides for weed control and toxicity to nursery plants. Included among the plants treated were two ground covers, Vinca minor (Myrtle) and Hedera helix baltica (Baltic Ivy). The chemicals used were 1-n-butyl-3-(3,4-dichlorophenyl)-1-methylurea, (neburon), at two pounds per acre; 2,4-dichlorophenoxyacetic amide, (2,4-D amide), at two pounds per acre; 2-chloro-4,6-bis(diethylamino)-s-triazine, (chlorazine), at eight pounds per acre; 2,3,6-trichlorobenzoic acid, (2,3,6-TBA), at two pounds per acre; and 2-(2,4,5-trichlorophenoxy)

ethyl-2,2-dichloropropionate, (erbon), applied at the rate of eighty pounds per acre. Though chlorotic injury due to 2,4-D amide appeared on some larger ornamentals, neither ground cover suffered from this type of symptom. Both, however, exhibited epinasty with the Myrtle recovering after approximately three months. In contrast, most of the Ivy plants were dead after eleven weeks. Chlorazine, on the other hand, caused moderate to severe chlorosis on Ivy, but the Myrtle was uninjured. The Ivy remained in poor condition even after eleven weeks. Neburon caused slight chlorosis on Ivy, but no other injury was particularly evident. 2,3,6-TBA showed unfavorable responses on both Myrtle and Ivy. The former showed epinasty, and either chlorosis or necrosis of the leaves and dieback of the tips, while the latter exhibited slight chlorosis and necrosis of the leaves. Erbon injury to both ground covers was very severe, eventually causing the death of all the Ivy plants.

Control of broadleaved weeds by four of the herbicides, (2,4-D amide, chlorazine, neburon and 2,3,6-TBA), was good to excellent after one months time, but grass weed control varied. 2,4-D amide, chlorazine and neburon gave fair grass weed control, but the use of 2,3,6-TBA resulted in only poor grass weed control. After two months only chlorazine continued to show excellent control of broadleaved weeds, and improved control of grasses. 2,4-D amide, neburon, and 2,3,6-TBA all showed poor control of grass weeds. Neburon's good control of broadleaved weeds remained unchanged, while all others decreased.

Rewarding results in field tests conducted by Ries, Grigsby and Davidson (22) during 1957 paved the way for further greenhouse tests.

Ground covers included among the plants that were treated were:

Hedera helix baltica, Pachysandra terminalis and Euonymus fortunei vegeta. The poor growth of weed seeds planted in the pots did not permit

their use as an index of weed control, but an evaluation of injury was possible. Euonymus exhibited severe chlorosis with the use of 2-chloro-4,6-bis(isopropylamino)-s-triazine, (propazine), at four pounds per acre, and slight chlorosis with a combination treatment of simazine and 2,2-dichloropropionic acid, (dalapon), at four and ten pounds per acre. Dalapon on Pachysandra at the rate of ten and twenty pounds per acre showed chlorotic injury, increasing respectively.

The use of various soil fumigants as a preplanting measure to control weeds in ground covers has been reported by different workers (21, 22). Efficient weed control was achieved in most cases. If planting was delayed, little or no injury resulted. But such measures usually require the use of a water seal following application of the various chemicals, which is not always feasible.

The foliage in Euonymus on median strip plantings has been reported to become 10% chlorotic when 3-amino-1,2,4-triazole, (amitrole), was used at two pounds per acre. At this rate only 30% weed control was obtained (2).

Other tests made to determine herbicide toxicities as well as seedling weed control ability of various chemicals have given fair to good results (9, 20).

Chlorotic injury symptoms have been found in plants other than ground covers (23). On grapes, Hemphill (16), reported that 3-(p-chlorophenyl)l, 1-dimethylurea, (monuron), caused chlorosis of young transplants, whereas, 3-(3,4-dichlorophenyl)-1,1-dimethylurea, (diuron), did not. Carlson (8) reported similar symptoms when monuron was used on light soils. Doll (12) on the other hand, found that diuron in addition to simazine caused mottled chlorosis of grape leaves, with the injury disappearing by the end of the season at lower rates. Plants

treated at higher rates continued to show symptoms throughout the season, with the higher rates producing consecutively more severe symptoms.

Apple trees, treated with sixteen pounds per acre of simazine, showed interveinal chlorosis and necrosis, which was followed by defoliation. Diuron injury, on the other hand, was characterized by faint veinal chlorosis on trees treated with eight pounds per acre, and extremely severe veinal chlorosis when used at sixteen pounds per acre. The chlorosis on the leaves of these trees rapidly advanced and changed to necrosis, ultimately causing complete defoliation of the tree (26).

In a study designed specifically to study the response of plants to monuron, Christoph and Fisk (10) found that soybeans developed chlorotic areas followed by red-brown spots, at rates as low as one pound per acre. An interesting observation was the fact that there was a marked reduction in the amount of mature xylem tissue, compression and collapse of the cambium, and disorganization of the phloem in the stems. Following cellular breakdown the xylem vessels in the leaves became plugged, thus, in the leaves this plugging was probably a secondary effect of the herbicide. Whether or not these conditions aggravated the chlorosis was not hypothesized.

Mode of Action of Chlorosis Inducing Herbicides

The actual cause of herbicide induced chlorosis is not known. The steps to a clear understanding could not even be started without having some knowledge of the mode of action of the herbicides causing such injury. The substituted urea (phenylurea) herbicides and the triazines have been referred to as causing chlorotic injury. The mechanism of

both groups has a common physiological action, that of upsetting photosynthesis (4). A detailed description of photosynthesis is unnecessary, but a review with emphasis on the phases affected by herbicides, and in particular those places where the effect could be causing chlorosis, may be helpful.

Chlorophyll, the wonder pigment in the plant which absorbs the light, is found in association with protein, forming chlorophyll-protein molecules that are arranged in layers. These layers of molecules form the functional units which trap the light. When photons of light strike this chlorophyll unit they throw electrons out of their track, leaving holes behind them. A certain number of these electrons will fall back into the holes, and as a result the chlorophyll emits light in the form of red fluorescence. There are some electrons that escape and are attracted by electron acceptors; these are finally returned to the chlorophyll where they came from by means of electron carriers such as cytochromes (19). Before they are returned, however, they transfer some of their energy for the creation of high energy phosphate bonds. As a result there is the formation of ATP with its high chemical bond energy. Here the electrons from the chlorophyll are returned to it by a seemingly closed circuit, or cyclic system, and ATP generation by this means is therefore known as cyclic phosphorylation. In a second case, the electrons do not flow back into the chlorophyll, but after being shot out are captured by triphosphopyridine nucleotide or TPN. (This process is referred to as non-cyclic phsophorylation). The TPN molecule with an extra electron now has a negative charge and as a result attracts a proton (H⁺) from water forming TPNH (reduction). It is TPNH and ATP that store the energy of light for future use in plant

processes. The holes left in the chlorophyll by the electrons captured by TPN are refilled with electrons from the hydroxyl ion (OH), left behind from the water after a proton was removed by TPN forming TPNH. In the process oxygen, along with water, is generated as a product. Evidence seems to indicate that the substitute urea and triazine herbicides interfere at this point where electrons are required to refill the holes in the chlorophyll molecule. As a result the holes left behind are not refilled since the supply of electrons is blocked by the herbicide. The higher the light intensity the more electrons that are shot out, and the more holes that are not refilled. Eventually the molecule has so many holes it is severely damaged. Van Overbeek (28) cites the following backing up this hypothesis. Remitted light in the form of red fluorescence is given off by all photosynthetic organisms after illumination. This reemission of light, as stated previously, is probably due to electrons refilling the holes in the chlorophyll. It has been shown that the substitute urea monuron, strongly inhibits this reemission of light.

A somewhat similar account of this phase of photosynthesis is given by Good (14). However, he does not commit himself as to whether it is the chlorophyll molecule that looses the electrons upon exposure to light, and then gains them back, except those lost to TPN. He states that the sun's energy is passed to a molecule of chlorophyll A, which is associated with two unidentified carrier substances designated as X and Y. "The reactivity of the light activated chlorophyll is such that Y is oxidized by loosing electrons or hydrogen atoms and X is reduced by gaining electrons or hydrogen atoms. Light energy is thus converted into potential chemical energy. It should be noted that either X or Y may, or may not, be a part of the chlorophyll molecule itself." The reduced carrier X can transfer its excess electrons or hydrogen atoms to a number of electron acceptors, the normal acceptor in

intact plant cells being TPN. The oxidized Y must be reduced again by some donar of electrons or hydrogen atoms, otherwise this whole process of the conversion of energy ceases. Under normal conditions oxidized Y obtains hydrogen from water, leaving oxygen gas as the product. However, in the presence of substituted ureas, such as monuron, this step is inhibited, and as a consequence there is an interference of the photosynthetic cycle. Both researchers seem to agree that the point of inhibition is at the stage where oxidation of water takes place. Very recent studies by Ashton, et. al. (3) on changes in the fine structure of chloroplasts in relation to herbicides affecting photosynthesis would tend to support Van Overbeek's assumption that it is the chlorophyll molecule itself that is damaged.

Though there is a drastic upset of the oxidation-reduction equilibrium, it must be pointed out that these herbicides would not just affect the chlorophyll molecule. They must have other consequences. Crafts (11) cites many examples such as: the loss of turgor, chlorosis, progressive dieback of leaves and retardation of mitosis in meristems in barley. In soybeans and tomatoes: chlorosis, collapse of young leaves, and disorganization of palisade tissue is common. Good also points out that there must be additional effects aside from inhibition of photosynthesis and ultimate starvation of the plant.

As stated previously, the triazine herbicides, an example of which is simazine, interfere with photosynthesis just as do the substituted ureas. Klingman (18) states that scientists do not fully understand just how simazine causes death of the plant, but it has been shown that simazine interferes with the cleaving of water into hydrogen and oxygen, an essential part of the Hill reaction. This is, of course, the same point where the substitute ureas have their effect, and therefore a complete reiteration of the inhibitory mechanism is unnecessary.

Naturally the ultimate cessation of photosynthesis due to this inhibition must eventually result in starvation of the plant. Though this is not a primary cause of death, it may have something to do with the chlorotic injury. If the chlorophyll molecule is injured, it will not be able to utilize the nutrients needed for food and chlorophyll synthesis. Certainly this would antagonize the chlorotic situation even more. Nutrient balances have been found to be upset by the use of herbicides. Analysis of peach leaves that were treated with simazine showed magnesium levels to be much higher than control plant leaves. Other element levels, such as boron, were also drastically upset. Additional mineral effects have been noted; for example, an interaction between phosphorus and diuron has been shown to occur (11). When phosphorus is applied to the soil of cotton and ryegrass cultures, it will counteract the effects of diuron in the one-quarter to two part per million range, and bring the green weight production of treated cultures up to that of controls.

Though the past information gives—some insight into the cause of chlorotic injury, it does not explain the reason for symptoms such as veinal and interveinal chlorosis caused by herbicides which presumably have the same killing mechanism. One explanation for this difference has been proposed by Good (14), who states that if the inhibitor (herbicide) is more soluble in some cellular substances than in water, it may accumulate in the first cells it comes to without spreading throughout the plant. Thus, a herbicide that is more soluble in fatty substances of the cell would accumulate immediately around the veins, causing a typical veinal chlorosis. A herbicide held less tightly in the cells would be washed out of these regions by the passage of the transpiration stream, and would accumulate in the interveinal area and along the edges. The injury symptom of this herbicide would appear as interveinal chlorosis.

Undoubtedly, no one factor is responsible for chlorosis, but rather several interacting factors, such as the mode of action and solubility of a herbicide. The problem will not be solved until these, and other questions related to them are accurately answered.

MATERIALS AND METHODS

Field Experiments - General

Two experimental plots, located at the Michigan State University

Horticulture Farm, were planted on a Hillsdale fine sandy loam during

October of 1963. Both plots were clean cultivated before planting with

rooted cuttings of the following ground covers: Euonymus fortunei vegeta

(wintercreeper), Pachysandra terminalis (Pachysandra or Japanese Spurge),

Vinca minor (Myrtle), and Hedra helix baltica (Baltic Ivy).

Pre-emergence Studies

The objective of the first experiment conducted in the field was to evaluate established and recently introduced pre-emergence herbicides in new ground cover plantings.

A completely randomized design was used having nine treatments, including the control. All treatments were replicated three times, each replication consisting of four plants of each of the above mentioned species. The treatments made shortly after planting were as follows: 2-chloro-4,6-bis (ethylamino)-s-triazine, (simazine), at 2 lb/A; 2,6-dichlorobenzonitrile, (dichlobenil), at 3 lb/A; dichlobenil at 6 lb/A; simazine plus 1,1-dimethyl-4,4-dipyridylium cation, (paraquat), at 2 plus 1/8 lb/A respectively; N,N-dimethyl-2, 2-diphenylacetamide, (diphenamid), at 8 lb/A; 3-(3,4-dichlorophenyl)-1,1-dimethylurea, (diuron), at 2 lb/A; 2,6-dinitro-N,N-di-n-propyl-a,a,a-trifluoro-ptoluidine, (trifluralin), at 6 lb/A; 2,4-bis(isopropylamino)-6-methylmercapto-s-triazine, (prometryne), at 2 lb/A, and a nontreated control.

Postemergence Studies

The second field experiment was conducted to study the use of herbicides for postemergence weed control in established ground cover plantings.

The experimental plot was allowed to become infested with weeds so that there would be a good source of seed the following spring. The intention here was to simulate an established ground cover planting, having actively growing weeds, and then to treat these plots in an attempt to remove the weeds without harming the ground covers. In addition to having much thinner ground cover growth, the weed populations under these conditions were heavier and more varied than in a normal established planting. However, it was felt advisable to study the herbicides' effects on simulated plantings before laying out plots in established plantings. A completely randomized design of the same type used on the fall treated plots was used, with the exception that ten treatments were applied, including the control. The treatments applied on June 26, 1964 were as follows: untreated control; hoe weeded check; N-cyclooctyl-N, N-dimethylurea plus N-phenyl-N-methyl-Nmethoxyurea, (H-150), at 2 lb/A; 3-(3,4-dichlorophenyl)-1-methoxy-1-methylurea, (linuron), at 2 lb/A; linuron at 4 lb/A; simazine at 2 and 4 lb/A; 80\% 3,4-dichlorobenzyl-N-methylcarbamate 20% 2,3-isomer, (UC 22463), at 2 lb/A; dichlobenil at 4 lb/A, and 2,2-dichloropropionic acid, (dalapon), at 5 lb/A.

Treatments for both the fall and spring tests were applied with a small plot sprayer, as designed by Ries and Terry (24).

Both plots were irrigated, and fertilized with 500 lb/A of 12-12-12 fertilizer. The pre-emergence plots were rated on May 21, June 17, and July 20, 1964. The postemergence studies were rated on July 15 and August 4, 1964. The ratings for weed control were on a 1 to 9 scale, where 1 was no control, 6 commercially acceptable control and 9 excellent control (little or no weeds). Injury ratings were made on the basis of a 1 to 5 scale, 1 being no visible injury, 3 severe chlorosis or burning, and 5 being death.

The data were statistically evaluated using analysis of variance. Where a significant F value occurred, Duncan's Multiple Range Test was used to compare mean difference.

MATERIALS AND METHODS

Greenhouse Experiments

Experiment I

The first of a series of experiments was started December 15, 1963, in the Horticulture Greenhouse at Michigan State University. These experiments were designed to determine at which levels herbicides could be used to induce chlorotic injury rather than severe burning, and to study possible causes of herbicide induced veinal and interveinal chlorosis.

Injured leaves were analyzed since it was felt that nutrient imbalances played a role in the development of chlorotic symptoms. All analyses were made at the plant analysis laboratory in the Department of Horticulture, Michigan State University, East Lansing. Samples were analyzed spectrographically for phosphorus, calcium, magnesium, sodium, manganese, iron, copper, boron, zinc, molybdenum and aluminum. Nitrogen determinations were made by the standard Kjeldahl method and potassium was determined by use of the flame photometer.

Rooted cuttings of Pachysandra treminalis (Japanese Spurge) and Hedra helix baltica (Baltic Ivy) were planted in a 1-1-1 mixture (soil, sand and peat) in four inch pots. Two cuttings of Japanese Spurge were planted in each pot. Those pots containing Ivy had only one cutting each of this species. The plants were then treated with four herbicides: simazine, prometryne, diuron, and simazine plus 1-n-butyl-3-(3,4-dichlorophenyl)-1-methylurea, (neburon). All four treatments were applied at three rates, four-tenths, four, and forty parts per million (ppm). 100mls of the solution were applied each time an application was made. Soybean seedlings were grown in each pot as indicator plants. Observations and notations were made on the type of injury that resulted.

Experiment II

A second experiment was started on March 21, 1964, to study the cause of herbicide chlorosis, and, if possible, gain some insight into the reasons for veinal and interveinal symptoms.

Rooted Pachysandra cuttings were potted in a fashion similar to those in the first experiment. Two cuttings were planted in each four inch pot, containing a 1-1-1 mixture of soil, sand and peat. The herbicides used were: simazine, prometryne, diuron and neburon. Neburon was substituted for the simazine-neburon combination used in the first greenhouse experiment. This was because no injury occurred to Pachysandra with the use of this combination.

A completely randomized design was used, with each of the four treatments being replicated six times. Each replication consisted of one pot containing two plants. 100 ml of solution were applied each time the treatments were made. All chemical solutions were mixed using commercial grade wettable powders.

Experiment III

A third greenhouse experiment, similar to the second test, but using soybeans, was started on April 15, 1964, in an endeavor to get more rapid results. Six seeds were sown in each four inch pot containing the same soil mixture used in the past. After a few weeks of growth, three plants were removed leaving the better plants for experimentation. The four treatments, used at the rates of .25, 1 and 4 ppm were as follows: simazine, prometryne, diuron and neburon.

A completely randomized design was used with each of the four treatments being replicated six times at each concentration. Once again, as in previous experiments 100 milliliters of solution were applied each time a treatment

was made.

In both the second and third greenhouse experiments injured leaves were removed from the plant and nutrient analysis was conducted.

RESULTS

Field Experiments

Pre-emergence Studies -- The results of the first rating showed that control with all eight herbicides was excellent (Table 1). The second rating made on June 17 showed that all treatments except trifluralin gave better control than the checks. However, only diuron at two pounds per acre, simazine, at two pounds per acre, and the simazine-paraquat combination, at two pounds plus one-eighth pound per acre respectively, gave control that was commercially acceptable. At the time of the final rating only simazine resulted in better control than the check plots, and this was not commercially acceptable.

All the Ivy plants were winterkilled, and as a result no injury ratings could be made relating to these plants. The myrtle in one replication of the diuron treatment, applied at two pounds per acre, was killed but no injury occurred to this species in any of the other replications of this treatment, so death was assumed to be due to some other factor.

Postemergence Studies -- The results from the second field experiment concerned with weed control in established ground cover plantings were not as rewarding as the first. Of the eight herbicides used, only one gave good postemergence control (Table 2). This was linuron at the rate of two and four pounds per acre. Unfortunately, the injury to the ground covers was severe, and would make the control commercially unacceptable. The hoe weeded check gave good weed control for only three weeks. Some interesting trends appeared that may be worthy of further experimentation; Simazine at two and four pounds per acre and dalapon at five pounds per acre gave noticeable control of grasses. There was some minor injury to the ground covers.

Table 1. - Effect of fall application of herbicides on pre-emergence weed control in ground cover plantings.

Treatments	Rate	Weed Control Ratings_*					
	1b/A	5/21/		6/17/		7/20/64	
simazine	2	9.0	С	8.0	С	4.7	b
simazine & paraquat	2 & 1/8	8.3	c	8.3	С	3.0 a	b
diuron	2	8.7	c	8.3	С	2.7 a	b
diphenamid	8	8.0	С	5.0	b	1.0 a	
prometryne	2	8.7	С	4.7	b	1.0 a	
dichlobenil	3	7.7	c	4.0	b	1.0 a	
dichlo benil	6	8.0	С	4.3	b	1.0 a	
trifluralin	6	3.7 k)	2.7 a	b	1.0 a	
none		1.0a		1.0a		1.0 a	

 $[\]stackrel{*}{-}$ All ratings are the average for three replications, rating scale

Values, in columns, followed by the same letter are not significantly different.

Values not followed by the same letter are significantly different at the 1% level.

^{1 =} no control; 6 = commercially acceptable control;

^{9 =} excellent control.

Table 2. - Effect of spring application of herbicides on postemergence weed control in ground cover plantings.

		Ratings						
Treatments	Rate lb/A	7/15/64		8/4/64				
		Weed control	Injury	Weed control	Injury			
none		1.0	1.0	1.0	1.0			
hoe check		9.0	1.0	5.3	1.0			
H-150	2	1.3	1.0	1.0	1.0			
linuron	2	7.0	4.0	2.0	4.0			
linuron	4	7.3	4.0	6.0	4.0			
simazine	2	2.3	2.7	2.0	2.3			
simazine	4	2.7	1.0	2.7	2.0			
UC22463	2	1.3	1.7	1.3	2.3			
dichlobenil	4	1.3	1.0	1.0	1.0			
dalapon	5	2.7	2.3	2.0	2.7			

All ratings are the average for three replications; rating scale for weed control: 1 = no control, 6 = commercially acceptable control, 9 = excellent control; injury rating scale: 1 = no injury, 3 = severe chlorosis and burning, 5 = death.

DISCUSSION

Field Experiments

Pre-emergence Studies -- Certain factors should be mentioned when considering the time period over which commercially acceptable control was achieved. Abnormally large sources of certain weed seed, such as grasses, could have come from adjacent treatments which gave poor control as early as May. In addition, certain ecological factors should be considered, that may have had an effect on the results. Both the simazine and the simazineparaquat combination had a replication in the western corner of the plot which was consistantly lower in its control rating. A probable cause of this was the existance of a low spot in that particular location into which much water from adjacent areas drained. This probably caused excessive leaching out of the herbicide. Stroube and Bondarenko (27) reported that five months after treatment with two pounds per acre of simazine, during which approximately ten inches of rain fell, the equivalent of 1/4 pound per acre remained in the top six inches of the soil. Burnside et. al. (7) also reported the leaching of simazine into the lower layers. At the same time weed seed was carried into this low region in wash water. Evidence of this can be found by examining the weed populations that were present. A recording of the weed populations found in each replication was made on July 7, 1964. In each case the replications for these two treatments in this particular location contained a larger number and variety of weeds than those in other sections of the plot.

An examination of the plots in September indicated that simazine was still showing discernible weed control, even though not commercially acceptable.

Postemergence Studies -- The methods by which the more popular ground covers spread could account for the difficulty in controlling weeds in established

plantings without injuring wanted plants. Pachysandra spreads by means of rhizomes which give rise to new shoots. Myrtle and Ivy, on the other hand, spread by roots forming at the nodes of trailing stems. In each case the major root mass is located at, or very near, the soil surface. This is the area where many herbicides are most active.

On the basis of the herbicides tested, postemergence control of extensive masses of weeds in established ground cover plantings using herbicides is not yet possible, and will be an important area for research in the present and future.

RESULTS

Greenhouse Experiments

Experiment I

Seven treatments were applied over a period of thirty-nine days before any symptoms appeared. (First and second treatment separated by thirteen days). Injury occurred first on the indicator plants at the highest (40 ppm) concentration, with death occurring rapidly. Two to seven days later, most of the Ivy plants at this concentration became necrotic in patches, localized mainly along the leaf margins. Some yellowing of the leaf margins and eventually the entire leaf occurred, but this rapidly changed to brown necrotic areas without any definite veinal or interveinal chlorosis developing. The injury started in the leaves nearest the base of the plant, with abscission occurring before the leaves in the tip region were completely dead. The same symptoms developed for all four herbicides, and death of all the Ivy plants at this highest concentration for all four treatments occurred approximately three and one-half months after the first application.

Injury symptoms did not show as rapidly with Pachysandra. They were first apparent about ninety days after the treatments were initiated in the case of prometryne and diuron, and about one-hundred days in the case of simazine. No injury was noted on the simazine-neburon combination. Once again, injury was confined to those plants receiving the highest concentration (40 ppm). The injury symptoms started as yellowing and then changed to small brownish necrotic patches which enlarged covering the entire leaf. Some mild veinal chlorosis was noted on the diuron treated plants. Death of all the plants receiving prometryne and diuron occurred approximately ninety days after the first treatment. In the case of simazine, only four out of eight plants were killed, and no death occurred among those plants treated with simazine plus neburon in combination at the highest concentration.

Since the injury found on Ivy was not usually of a distinct chlorotic nature, it was decided to discontinue the use of this plant and concentrate on Pachysandra which had produced the desired symptoms.

One other problem was the long period of time needed before the symptoms fully developed. Having noted that the soybean indicator plants achieved fully developed symptoms rapidly, it was decided that a third experiment would be conducted using soybeans as the plant material rather than ground covers.

Experiment II and III

Injury symptoms appeared on both Pachysandra and soybean plants that were treated with prometryne. The characteristic injury was a definite venial chlorosis. Pachysandra also showed a veinal chlorosis with diuron, yellowing being most prominant at the basal portion of the petiole. Some minor yellowing occurred on diuron treated soybeans, but could not be distinguished as veinal or interveinal. Simazine caused an interveinal chlorosis on soybeans, but no injury occurred on Pachysandra plants treated with this same chemical at a concentration of 40 ppm. Neither the soybeans nor the Pachysandra showed any injury from neburon treatments.

Analysis of the leaf material collected showed differences between controls and treated plants in the element levels of sodium, calcium, iron and zinc. Not enough leaf material could be collected to replicate the treatments, consequently these results cannot be considered as trends in the nutrient levels of various herbicide treated plants.

DISCUSSION

Greenhouse Experiments

Unfortunately, not enough leaf material could be collected to fully study nutrient imbalances in treated leaves as had originally been planned. Some points should be noted. As stated in the literature review, Good (14) feels that veinal and interveinal injury may be accounted for due to differences in the solubility of the herbicides in some cellular substances, in contrast to solubility in water. He states that the phenylureas appear to be less tightly held in the cells. Presumably, the transpiration stream in passing through the main vein region of the leaf washes out the herbicide, which then accumulates at the leaf margins and in the interveinal areas, causing an interveinal chlorosis. The phenylurea used by Good, leading to this hypothesis, was monuron, having a solubility in water of 230 ppm. One of the phenylureas used in this study was diuron, which, as stated in the foregoing results, caused a veinal chlorosis. This was also reported by other workers using diuron (26). It is interesting to note that the solubility in water of diuron is only 42 ppm. Relating the work done by Good with the results obtained from experiments conducted in this study, it appears that the water solubility of a herbicide within a certain group (phenylureas, triazines, acylanilides, etc.,) plays a role in the form of chlorotic injury it induces. It is logical to hypothesize that if two materials are both in the root zone, the one of higher solubility will be more readily absorbed by the plant, all other factors being equal. As a consequence a greater amount of the more soluble herbicide would be present in the plant and, provided it isn't held tightly by the cell, could be washed into the intercostal areas by the transpiration stream. Plausible as this seems with the phenylureas, it

raises question with the triazines. Here simazine having a solubility in water of only five ppm, causes interveinal chlorosis, whereas, prometryne with a solubility in water of 48 ppm causes veinal chlorosis. It should be noted, however, that these two compounds differ in that simazine possesses a chloro group on the benzene ring, whereas, prometryne has a thio group instead. This could make some difference in the ability of the plant to absorb the simazine ion. Once inside the plant, C ¹⁴ labled simazine has been shown to move readily in the transpiration stream (13). In a test simulating conditions in the apoplast, paper was used with water as the only solvent. Movement was not correlated either directly or indirectly with water solubility (13). Thus, it seems that once in the plant, regardless of the water solubility and provided the material is not held by the cell, movement in water won't be affected. How readily it may be absorbed by the plant may have an effect on the total amount moved, and as a result on the injury incurred.

SUMMARY

The use of herbicides in ground cover plantings, and triazine and phenylurea induced chlorosis were studied.

Field tests were conducted to evaluate fall applied pre-emergence herbicides in new ground cover plantings, and spring applied postemergence herbicides in established ground cover plantings. Simazine at two pounds per acre, diuron at two pounds per acre, and simazine plus paraquat at two plus one-eighth pound per acre respectively gave the best results when applied as a pre-emergence treatment.

None of the postemergence herbicides used gave acceptable weed control without causing severe injury to the ground cover plants.

Investigation of triazine and urea caused chlorosis under greenhouse conditions showed that there was no true correlation between solubility and chlorosis caused, but indicated that the availability of a herbicide to the plant, as influenced by it's water solubility, can have an effect on the expression of veinal versus interveinal chlorosis.

LITERATURE CITED

- 1. Anonymus. "The Holy Bible," Revised Standard Version, Book of Genesis I, Verses 1, 11, 12.
- Ashton, Floyd M., Ernest M. Gifford, Jr. and Thana Bisalputra.
 "Structural Changes in Phaseolus vulgaris induced by Atrazine,
 II Effect on fine Structure of Chloroplasts." Bot. Gazette. 127 (5)
 pp 336-343. 1963.
- 4. Audus, L.J. "The Physiology and Biochemistry of Herbicides." Academic Press. pp 387-399. 1964.
- 5. Birdsell, Duncan G., Donald P. Watson and Buford H. Grigsby.
 "Toxicity and Efficiency of Selected Herbicides on Representative Ornamental Plants." Weeds. 6:34-41. 1958.
- 6. Blackman, G.E. "Plant Growth Substances as Selective Weed Killers.

 A comparison of certain plant-growth substances and other selective herbicides." Nature (London) 155:497. 1945.
- 7. Burnside, O.C., E.L. Schmidt and R. Behrens. "Dissipation of Simazine from the Soil." Weeds. 9:477-484. 1961.
- 8. Carlson, R.F. "CMU as a Herbicide in Vineyards." Quart. Bull., Mich. Agr. Exp. Sta. 38:409-412. 1956.
- 9. Carlson, R.F. and F. B. Widmoyer. "A progress report of the Effect of N-1-Naphtyl phtalamic (Alanap) and 2,4-dichlorophenoxyethyl sulfate (Sesone) on 11 Species of Ground Cover Plants." Mich. Agr. Exp. Sta. Quart. Bull. 34, (4), 599-604. 1957.
- Christoph, Roy J. and Emma L. Fisk. "Response Plants to the Herbicide 3-(p-chlorophenyl)-1, 1-dimethylurea." Botanical Gaz. 116(1):1-14. 1954.

- 11. Crafts, A.S. "The Chemistry and Mode of Action of Herbicides." Interscience Publishers. 1961.
- 12. Doll, C.C. "Chemical Weed Control Studies in Young Vineyards."
 Proc. 14th N. Centr. Weed Cont. Conf. 48-49. 1957.
- 13. Foy, C.L. and P. Castelfranco. "Distribution and Metabolic Fate of C¹⁴ Labeled 2-chloro-4, 6-bis-(ethylamino)-s-triazine, (Simazine and Four Related Alkylamino triazines in Relation to Phytotoxicity." Proc. of the Plant Physiol. Meetings, Vol. 35, Supplement. p 28. Aug. 1960.
- 14. Good, Norman E. "Inhibitors of Photosynthesis as Herbicides." World Review of Pest Control, Part 1. 19-28. Spring 1962.
- 15. Hamner, C.L. and H. B. Tukey. "Herbicidal Action of 2,4-dichlorophenoxyacetic and 2,4,5-trichlorophenoxyacetic acid on Bindweed." Science. 100:154-155. Aug. 18, 1944.
- 16. Hamner, C.L. and H. B. Tukey. "Selective Herbicidal Action of Midsummer and Fall Applications of 2, 4-dichlorophenoxyacetic acid." Bot. Gaz. 106:232-245. Apr. 1944.
- 17. Hemphill, D.D. "Chemical Weed Control in Vineyards." Proc. 13th N. Centr. Weed. Cont. Conf. 64-65. 1956.
- 18. Klingman, Glenn C. and Lyman J. Noordhoff. "Weed Control as a Science." John Wiley & Sons, Inc. 1961.
- 19. Meyer, Bernard S., Donald B. Anderson and Richard H. Böhning. "Introduction to Plant Physiology." D. Van Nostrand Com., Inc. pp 292. 1960.
- 20. Pridham, A.M.S., Granular Herbicides for Weeds in Ground Cover Plantings. Proc. N.E.W.C.C. 12:108-110. 1958.
- 21. "Crop, Weed and Soil Aspects of Simazine, Atrazine and other Herbicides in Greenhouse and Field Tests." Proc. 14th N.E.W.C.C. pp 142-147. 1960.

- 22. "Granular Mylone A Preplanting Measure for Control of Perennial Weeds of Nursery and Ornamental Plantings." Proc. 13th N.E.W.C.C. pp 384-385. 1959.
- 23. Ries, S.K., B.H. Grigsby and H. Davidson. "Evaluation of Herbicides for several species of Ornamentals." Weeds. 7:409-417. 1959.
- 24. ____ and C. W. Terry. "The design and evaluation of a small-plot sprayer." Weeds 1:160-174. Jan. 1952.
- 25. Runge, George F. "Nursery Weed Control." Proc. 14th N.E.W.C.C. 165-166. 1960.
- 26. Saidak, W. J. and W.M. Rutherford. The Tolerance of Young Apple Trees to Amitrole, Diuron and Simazine. Canadian Jour. Sci. 43:113-118. April 1963.
- 27. Stroube, Edward W, and Donald D. Bondarenko. Persistance and distribution of Simazine applied in the field. Proc. 17th N.C.W.C.C. 40-41. 1960.
- 28. Van Overbeek, J. "Physiological Responses of Plants to Herbicides." Weeds. 10 (3):170-173. 1962.
- 29. Wallace, T. "The Diagnosis of Mineral Deficiencies in Plants by Visual Symptoms." His Majesty's Stationary Office. pp 31-32. 1944.
- 30. Zimmerman, P.W. and A.E. Hitchcock. Substituted Phenoxy and Benzoic Acid Growth Substances and the Relation of Structure to Physiological Activity. Contr. Boyee Thompson Inst. 13: 313-322. 1942.

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