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Title: A KINETIC ANALYSIS OF HERBICIDE DEGRADATION IN SOIL

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Chehalis silt loam was treated with nine different herbicides. Each was applied at eight ppm and [3(3,4-dichlorophenyl)-1,1-dimethyl urea] diuron was also applied at two ppm. The degradation of diuron was followed at 6.5, 13.2 and 31.2 degrees centigrade. The degradation of all other herbicides was followed at 13.2 and 31.2 degrees centigrade. The soil moisture was maintained at approximately 50 percent of field capacity. Monthly samples were analyzed for remaining herbicide using the techniques of analytical chemistry. The rate constant for herbicide degradation was obtained by plotting the log of the concentration remaining in the soil vs. time in months. Evaluation of the rate constant at different temperatures permitted calculation of the heat or energy of activation from the Arrehenius equation.

It was determined that the rate of degradation of nine herbicides in three classes did follow a first order rate law, and that the rate could be related to molecular structure. The first order rate began only after an initial lag period of undetermined length in the case of the phenyl urea herbicides diuron; [ 3-(p-chlorophenyl)-1, 1-dimethyl urea], monuron; [ N,N-dimethyl-N'-phenyl urea], fenuron; N'-4(4-chlorophenoxy)-phenyl-N,N-dimethyl urea, chloroxuron. This was indicative of a microbial breakdown. The triazines: (2-methylthio-4-ethylamino-6-isopropylamino-s-triazine), ametryne; (2-chloro-4-ethylamino-6-isopropylamino-s-triazine) atrazine; and (2-chloro-4,6-bis-ethylamino-s-triazine) simazine, and the uracils: (5-bromo-3-sec-butyl-6-methyluracil) bromacil, and (3-tertbutyl-5-chloro-6-methyluracil) terbacil, had no detectable lag period, from which non-enzymatic degradation may be suspected.

The energies of activation for the phenyl urea molecules fell in the narrow range of 4-5 kilocalories per mole, which strongly suggested a common mode of degradation. Analysis of bond energies and published data supported the contention that this was an N-demethylation. The rate of N-demethylation was apparently attenuated by the substituents on the phenyl group. The energy of activation for the triazines and uracils did not fall in a narrow range which suggested breakage of a different bond in each case. Thus, it was concluded that a non-enzymatic conversion of the triazines to

the hydroxy derivative was occurring at the point of variance, i.e., the two position. As evidenced by the data, the uracils also appeared to be attacked by chemical hydrolysis at the point of variance, i.e., the halogen substituent.

Suggestions are made regarding the use of the parameters of rate and activation energy for predictions of persistence, hypotheses concerning the initial point of attack and the mechanism of degradation.

# A Kinetic Analysis of Herbicide Degradation in Soil

by

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# A KINETIC ANALYSIS OF HERBICIDE DEGRADATION IN SOIL

> The search for truth is in one way hard and in another easy. For it is evident that no one can master it fully nor miss it wholly. But each adds a little to our knowledge of Nature and from all the facts assembled there arises a certain grandeur.
>
> Aristotle

## I. INTRODUCTION

Application of a herbicide to the soil introduces the herbicide into a heterogeneous medium where a vast complexity of possible interactions exists. There is an abundance of scientific literature which attempts to explain these interactions and thereby explain the soil activity and persistence of herbicides. Webster defines persistence as "continuing steadily or firmly in some state, purpose, course of action or the like; to last or endure." The degree of persistence is, _in toto_, a result of the interactions between a herbicide and its environment. Herbicide residues which do persist in soil pose several potential environmental problems. They may cause injury to succeeding crops grown in rotation with a treated crop or accumulate at a rate faster than the rate of dissipation and cause more extensive damage. The accumulation of unlawful and often unknown residues in successive crops or in water sources is one of the

most publicized problems. Herbicides are applied to selectively eliminate weeds on agricultural land or to eliminate all vegetation on industrial sites. Their persistence, while not wholly undesirable, presents problems with important agricultural and public health implications. Within the last fifteen years the public has become increasingly aware of the dangers inherent in the presence of pesticide residues in the environment. This awareness and the concomitant mandate for solutions to this problem has created an increasing demand for methods and information that will facilitate accurate predictions of the residual life of herbicides in soil.

As Alexander (4) has pointed out, the issue regarding the use of pesticides has two cardinal assumptions. The first is that people must eat. The second, that they should not be poisoned now or in subsequent years because they cannot avoid eating. Therefore, the problem of herbicide persistence in soil is not of purely academic interest.

There are three fundamental characteristics of herbicides which relate to their effectiveness and persistence in soils (18). Each of these is capable of being measured quantitatively but is always influenced by the others. The first characteristic is the fixation of a herbicide in the soil as it relates to its availability to plants. The second is the uptake by and movement in plants. The final trait is that of lability with respect to decomposition and

detoxification, and it is this that will be examined herein.

All chemical reactions pass through an activated complex and a finite amount of energy is involved (19). The energy requirements are determined by the reactants and the medium. These experiments were designed to test the hypothesis that herbicide breakdown in the soil follows a first order rate law. It was theorized that the rate of breakdown and associated activation energy of different groups of compounds would be different and that the difference could be related to molecular structure. Thus, similar molecules would follow similar paths, and variability within and between classes of compounds would represent the influence of molecular structure. The experimental procedure measured the rate of breakdown and from this the activation energy was calculated. The relation of these values to molecular structure, and chemical and physical properties of the herbicides will be discussed.

# II. LITERATURE REVIEW

Upchurch (78) states that herbicide placement in the soil, whether by intent or as a result of processes in nature, is the dominant factor controlling soil behavior. However, placement, including method of application, might be more properly considered as an imposed factor which regulates the interaction of five primary factors on the chemical and physical properties of a herbicide and thus its persistence in any given soil situation. These factors are: (1) microbial breakdown; (2) adsorption; (3) leaching; (4) volatilization; and (5) chemical decomposition. Plant uptake and metabolism could also be included, but this is the goal of application and will not be considered in this review. Freed (31) has focused attention on a parallelepiped whose upper surface is a few centimeters above the vegetation and whose lower surface is a few centimeters below the soil-air phase boundary. Several variables will be operative in this defined system and thereby affect the soil life of herbicides. Among these will be: temperature or heat input, air flow, light intensity and quality, water flux, transevaporation rates, and chemical reaction potential. Herbicides which cannot survive the rigors of a given environment for the length of time required for uptake of toxic amounts by plants are of no value. However, great resistance to decomposition may be as serious a disadvantage. It is the

combination of the above-mentioned factors with the nature and properties of the particular molecule that determine the term of soil residence. Effectiveness, while correlated with soil life, is also related to the concentration in the region of soil where the effects are desired. This review will now deal with the current knowledge of how each of the five primary factors affects persistence.

## Microbial Decomposition

The existence of microbial decomposition is well documented for several classes of herbicides (7, 9). Compounds as diverse as aryl carbamates, phenoxyacetic acids, phenyl ureas and s-triazines have all been shown to undergo microbial degradation (33, 44, 49, 71). Sheets et al. (71) have reported very slow dissipation for most inorganic herbicides and for organics such as the benzoic acid derivatives. It cannot be assumed that microbial action will decompose all organic molecules. Radio-carbon datings have shown the organic compounds of certain peats to have an age of ± 34,000 years (52) and others 1580 to 2860 years (62). However, microbial action is often the primary mode of herbicide decomposition in soil. With some exceptions, microbes are of importance because they regulate the length of phytotoxicity in soil without affecting initial activity (78).

Two herbicide-microorganism interactions are possible.

These are: (1) potential inhibition of microorganisms and, (2) microbial alteration of the chemical as mentioned above. Alexander (2) has pointed out several factors of importance in the ability of microorganisms to degrade herbicides. These can be grouped as factors most favorable for microbial function. The optimum temperature range is $80^{o}$ to $90^{o}$ F, soil moisture is optimal at 50 to 100 percent of field capacity and aerobic conditions are best. There is also a very specific pH range for most microbial processes. Secondly, adsorption of herbicide, microorganism or enzyme may increase persistence by limiting interactions. At normal field application rates the effects of herbicides on microorganisms are negligible (12, 15, 82). An additional factor of importance is the amount of normal or more accessible substrate, such as organic matter, that is present (11). Biodegradability is probably a characteristic of definite ecosystems and the susceptibility to degradation will vary with the environment to which a herbicide is exposed.

Audus (7) has defined three phases in the time course of changes in the soil concentration of 2,4-dichlorophenoxyacetic acid (2,4-D) during microbial breakdown. He found an initial drop in concentration and assumed it was due to adsorption. There were ten to fourteen days with no change, followed by one to three days for rapid and complete disappearance. Microbial action was verified by using metabolic inhibitors in another experiment. The presence and length

of these phases of microbial decomposition have not been established for more persistent herbicides.

Alexander and his co-workers (3, 4, 5, 55) have delineated specific molecular characteristics that influence stability. Working with halogenated phenoxy acid derivatives they discovered the rate of microbial degradation was governed by the position of the halogen on the aromatic nucleus and by the linkage and type of aliphatic side chain. Halogenation of phenoxyaliphatic acids or of dichlorophenols in the meta position increased persistence. Attachment of the aliphatic side chain through the alpha carbon had a similar effect. This is an interesting approach to persistence but has not been followed for other classes of compounds. Upchurch (78) points out that such data can be understood only when the herbicidal properties of a given compound are known and compared with the inherent phytotoxicity of a reference compound in the class. Alexander (4) mentions evidence which indicates that persistence is not associated with the effect of structure on microbial decomposition per se, but rather with unknown interactions with non-biological soil constituents.

It has been established that many herbicides are decomposed in soil by microorganisms, and many species have been isolated and identified. The time for dissipation is known for some herbicides and in general for classes (4, 8, 28, 36, 43, 49). However, the real basis for the slow degradation of herbicides has not been

established. The microorganisms may be an extremely minor component of the soil population incapable of preferentially using the herbicide as an energy source within the imposed environment. The herbicide molecule could be competing with other soil components as a substrate or the requisite reaction might be suppressed in some way. A final hypothesis is that the toxic reaction of herbicides on microorganisms does exist and it prevents the build-up of organisms required for decomposition (78).

A detailed review of all of the pathways of microbial degradation that have been identified is beyond the scope of this review. A few references bear directly on the results and they will be discussed.

Kaufman and Kearney (50) and Duke (27) have reported specific soil microorganisms capable of degrading simazine and atrazine. This and subsequent work showed dealkylation with no ring cleavage.

McCormick and Hiltbold (57) found the inactivation of diuron and simazine was directly related to the metabolism of soil organic carbon. Repeated additions of energy material were closely associated with the rate of inactivation indicating an involvement of microorganisms. They found the rate of decomposition of atrazine doubled and diuron tripled for each ten degree rise in temperature from ten to thirty degrees centigrade. This paralleled the response of soil organic matter and was cited as a measure of microbial efficiency.

On the other hand, Ercegovich's (29) proposal of alteration of the triazines in the soil, by a number of chemical reactions, should not be neglected. Such reactions could occur in a microenvironment or after adsorption.

Specific information on the soil breakdown of the urea herbicides is also lacking. Geissbühler et al. (36) showed that soil microbes inactivate a specific N-methylated urea by a demethylation reaction leading to the appropriate aniline derivative. Dalton et al. (21) demonstrated a similar pattern for diuron with the removal of each methyl group followed by hydrolysis of the urea to 3,4 dichloro aniline. Sheets and Smith (73) have identified the monomethyl, demethylated and aniline derivatives of diuron in treated soil.

## Adsorption

The process of adsorption can be chemical (bonding forces) or physical (electrostatic forces) and is regulated by the characteristics of the soil solution and colloidal fraction. Its implications for pesticides have been extensively reviewed by Bailey and White (10). Hartley (42) indicates its importance when he mentions that many herbicides are more persistent in surface layers than would be expected from rainfall and solubility alone. He concludes that some factor other than low water solubility is holding back the herbicide and that the solubility of many herbicides in the soil is apparently

much greater than in the soil's water content alone. Adsorption determines how much herbicide will remain in solution and "active" and concurrently how much herbicide will be mobile and leached or subject to microbial decomposition or plant uptake. Adsorption regulates an equilibrium concentration that generally occurs within a few hours of application (34).

It has been reported that adsorption is greater in fine than coarse textured soils (10, 42, 65) and that bioactivity follows a reverse relationship (10). This is an expression of differences in adsorptive capacity and specific surface. It is a well established fact that clays and organic matter are the reservoirs for small metallic cation exchange in the soil. However, this fact may have no relevance to the adsorption of large organic molecules (39). Frissel (34) has examined the principles underlying adsorption of neutral and charged molecules on three clay minerals. He concluded that there was no especially strong attraction between herbicides and clays. At concentrations of practical interest, a soil composed entirely of clay would need 0.001 percent or less of its surface area to adsorb all of the added herbicide. Most workers agree the clay adsorption does occur but they have found a greater correlation with organic matter content (35, 39, 45, 77, 79, 83). Under similar climatic conditions it has been shown that soil organic matter increases with clay content. The correlation of these two factors

tends to explain the correlation of each with adsorption (66, 70, 79). In spite of these and others, it has been difficult to extend the results to soils in situ. One must not assume that an apparent correlation necessarily implies a causal relationship. Yuen and Hilton (83) have been able to prescribe herbicide dosage rates based on previously determined adsorption curves for individual soils. The fact that this could only be done on an individual soil basis reinforces Frissel's contention (34) that the forces involved in adsorption result from specific interacting forces of, a priori, unknown magnitude, and cannot be qualitatively predicted. Thus, it may not be possible to extrapolate general predictions based on a herbicide's physical and chemical properties without considering the specific soil parameters. Lambert et al. (51) have shown a high degree of predictability given the organic matter content and properties of the chemical. Upchurch has reported similar findings. Over many soil types a 14-fold range of toxicity for cotton and a 26-fold range for ryegrass were found for monuron (80). These findings were confirmed (79) and a 43-fold increase in herbicide concentration required to reach the $ED_{50}$ value of the same crops was detected as organic matter was increased from 0.5 to 17 percent. Based on several studies the assumption has been made that the organic matter adsorbs herbicide and thereby makes it biologically unavailable to the plant. This assumption is probably correct but the

question has been raised (78) whether such inactivation can be accounted for solely by adsorption or if it is in part associated with the greater microbial activity of soils with higher amounts of organic matter.

Adsorption, as a soil phenomenon is, without question, a fundamental determinant of herbicide persistence. It is the foremost determinant of concentration in the soil solution and affects each of the other four primary factors in soil persistance.

## Leaching

The amount of herbicide carried into the soil by water depends on the properties of the chemical, the amount of water available, and the speed of water movement through the chemical zone (33). Leaching results in the placement of herbicide at some point at which its specific characteristics or consequences may be modified (77). Downward flow is the predominant aspect of leaching but lateral diffusive movement and upward movement also occur, and with rill or furrow irrigation can concentrate herbicides at or near the soil surface (40) in bands which may or may not enhance the desired effect. The leaching process has been divided into two components (80). The first is the entrance of the herbicide into solution, as controlled by the specific solubility of the herbicide. This step implies that a leachable compound must also be soluble and not

totally adsorbed. Solubility may be related to leachability within a class of compounds because there is frequently a relation between adsorption and solubility among nonionic compounds (78). The second component of leachability is soil adsorption and plant uptake. This factor will be affected by soil texture, soil permeability, and the amount and frequency of rainfall (41, 80).

Downward movement of water in the form of a discontinuous film is produced by the combined effects of capillary and gravitational forces (33). The herbicide is alternately adsorbed and desorbed by soil surfaces. If solubility is low and adsorption is strong, only large amounts of water will affect downward flow and the herbicide will be concentrated near the surface. If solubility is great and adsorption is weak, small amounts of water will move the herbicide rapidly out of the root zone. The herbicide is in a dynamic equilibrium which is different at each finite soil level. At no time is all of the herbicide adsorbed on the surface of soil particles (33). Some prediction of the leaching of a specific herbicide can be made by considering the texture, percolation rate, and adsorptive characteristics of the soil, and the physical and chemical properties of the herbicide. The velocity of leaching has been related directly to the amount of percolating water (80) and the energy of adsorption which was shown to be related in a general way to the latent heat of solubility (32). The absolute amount leached will be determined by

the initial application rate and the sorption-solubility equilibrium (79).

## Volatilization

All herbicides have a finite vapor pressure. For some this is a negligible quantity, but loss by volatility can be significant for many surface applied herbicides (e.g. thiolcarbamates). Rapid microbial or chemical decomposition, adsorption, leaching, or placement in the soil will decrease or eliminate the occurrence of these losses. The tendency to volatilize from a soil surface has two opposed consequences (78). The vapor may be toxic to desirable plants present in the treated field or in adjacent untreated areas and cause unwanted injury and loss of herbicide (25). However, volatilization can also be beneficial to weed control as shown by Slater (75) for the effect of [isopropyl-(N-3-chlorophenyl carbamate)] CIPC vapors on dodder (Cuscuta spp.) seedlings. Volatility can also be a serious loss for most of the thiolcarbamate herbicides and result in a reduction in the extent of phytotoxic action (37). The tendency of any chemical to volatilize is a function of its vapor pressure, which is influenced by temperature.

Large losses of (ethyl-N,N-di-n-propyl thiolcarbamate) EPTC by volatilization have been demonstrated by Fang et al. (31). Their results explained the greater loss of EPTC from wet as opposed to

dry soil surfaces in terms of greater adsorption by the dry soil. If a herbicide is applied on dry soil it can immediately penetrate by diffusive and non-diffusive mechanisms. On the other hand, on wet soil the herbicide penetrates only with difficulty. Also, water appears to displace the chemical from the adsorptive sites. Once in the soil the EPTC molecules are held by adsorption and further volatility losses are reduced.

Deming (26) has shown that under certain circumstances the relation of volatilization and temperature was reversed to give a decreasing vapor loss with increasing temperature. This relation was influenced by soil water content; with increasing amounts of water losses due to volatilization increased with increasing temperature. Deming suggests that this results from direct competition for adsorption sites between herbicide and water molecules. At low soil moisture he detected less loss at higher temperatures and attributed the effect to more herbicide adsorption owing to less competition for sites.

An additional phenomenon that occurs when volatile herbicides are applied on warm, wet soil surfaces is steam or co-distillation. Hartley (42) has pointed out that this is actually a misnomer. The process actually occurring is one of herbicide concentration at the soil surface by water moving upward as a liquid. This movement concentrates the herbicide molecules at the soil surface until they

attain a vapor pressure at which they evaporate as fast as they arrive. Upchurch states that vaporization is best viewed as an interface phenomenon (78). The herbicide molecule is in a dynamic equilibrium between sub-surface areas and surface boundaries. The tendency to volatilize will depend on the concentration and strength of binding at interfaces.

## Chemical Decomposition

The final consideration among the primary factors affecting persistence is chemical or non enzymatic decomposition. Microbial action is a chemolytic process, however, there are numerous chemical reactions possible in soil that are not biologically mediated. Hartley (42) discusses the fact that adsorption can retard or more frequently accelerate chemical breakdown by a process of surface catalysis. Some workers have reported herbicides to be less persistent in soils with a high adsorptive capacity: simazine (13) and phenyl ureas (45, 79). This effect has been attributed to increased microbial attack and should be considered. However, increased adsorption and the concomitant lowering of effective pH might effect more favorable conditions for non-biological decomposition (34).

The conversion of simazine to hydroxysimazine 2-hydroxy-4,6,-bis (ethylamino)-s-triazine has been reported in soil (41). Similar findings have been reported for atrazine (6, 74). These

data place the question of relevance on the findings of Kaufman and Kearney (50) concerning the ability of microorganisms to metabolize the N-alkyl side chains of these molecules. One might also question whether the omnivorous appetite attributed to the ubiquitous soil microorganisms is always responsible for the degradation of chemicals. However, the possibility of combinations of modes of degradation should be recognized.

Photodecomposition is a form of chemical degradation. It is generally agreed that several herbicides will undergo photodecomposition which is an effect of radiation on internal chemical bonds. The absorption of electromagnetic radiation of specific wavelength can influence the excitation state of electrons and lead to bond rupture. Heterocyclic compounds and compounds containing nitrogen appear to be particularly susceptible (33). Studies have been made on the ultra-violet decomposition of herbicides: simazine (1), phenyl ureas (17, 48), diquat and paraquat (16), 2,4-D (60), amiben (67). These studies have been heavily dependent on laboratory work and have not measured decomposition from a natural soil surface. If such soil measurements were attempted the results were determined by bioassay which indicates what happened but not why. These facts support Hartley's (42) contention that direct photodecomposition may not occur. He maintains that it is very difficult to distinguish between photochemical losses and

losses due to volatilization owing to increased temperature at the soil surface, and that no one has successfully done so.

## Other Kinetic Studies

Hill et al. (44) were the first to examine the disappearance of urea herbicides from the soil by application of a first order rate law. They found that the values of k (rate constant) were essentially constant over a five fold range of $x_o/x$ ($x_o$ = amount present at $t_o$, x = amount present at time t). Recognizing that variable soil moisture and temperature conditions could accelerate or retard the rate of removal, they believed the first order rate law to be applicable under the usual field conditions. They also postulated that phenyl urea breakdown in the soil was mediated by microorganisms. They found a constant rate of breakdown for monuron as opposed to the lag phase breakdown proposed for 2,4-D (7). Burschel and Freed (14) examined herbicide decomposition from a more refined physical chemical point of view. They reasoned that herbicide losses in soil should follow a first order rate law. Therefore, because the soil, the moisture, and the microorganisms of the soil are already in abundance, or capable of becoming non-limiting, the rate limiting component should be concentration of the herbicide. Using a bioassay they demonstrated a first order rate of disappearance for (3-amino-1,2,4-triazole) amino triazole, CIPC and IPC

(isopropyl-N-phenylcarbamate). Using the Arrehenius equation and the rate constants at two temperatures they calculated the enthalpy of activation for each of these compounds. The values obtained were:

| | |
|---|---|
| amino triazole | 5374 Cals/mole |
| IPC | 7768 Cals/mole |
| CIPC | 21247 Cals/mole |

The authors proposed that the parameter of energy of activation could be used in estimating the residual life of a herbicide in soil where decomposition was the major factor in loss of activity. There was no explanation of the large difference between IPC and CIPC.

The hypothesis concerning a first order rate of disappearance has been reiterated in the literature (14, 33, 69). Riepma (64) investigated the soil breakdown of amitrole and calculated rate constants and the energy of activation. He found a first order rate constant and an energy of activation (5078 cals/mole) that agreed very well with the results of Burschel and Freed. However, Riepma reported a lag period in the breakdown that increased with increasing amitrole concentration in the soil. This suggests (38) that at higher rates the herbicide can initiate or prolong an adaptive phase in the metabolic processes of the soil microorganisms.

Grover (38) working with (4-amino-3, 5, 6-trichloropicolinic acid), picloram, also found a pronounced lag phase which increased with increasing concentration of chemical. He observed first order kinetics via a bioassay analysis for remaining picloram. The first order breakdown was not observed until after the initial lag period. Therefore, he proposed that assigning half-life periods to picloram degradation in soil is of doubtful value.

With the exception of the paper by Grover (38) and the earlier work by Hill et al. (44) there are no reports of kinetic studies of the degradation of the more persistent herbicides.

## Field Studies

The comparative and residual phytotoxicities of some of the herbicides included in this study have been compared in several field studies. Results representative of these data are included herein.

Sheets (65) and Sheets and Crafts (69) compared the phytotoxicities of phenylurea herbicides in several soil types, ranging from a clay to a sandy loam. They found the initial toxic level varied with soil type, with the lighter soils requiring less chemical to produce injury or death of plants. The rate of detoxification varied in the order fenuron, monuron, and diuron, with the latter being the most persistent. However, monuron had the highest initial

phytotoxicity with fenuron second. The variation in effective dosage suggested the adsorptive interaction with clay minerals was the greatest for Diuron. They proposed that the effect of the substituents on the phenyl moiety was evident in the pattern of persistence. In a later article Sheets (68) said that massive accumulation of diuron residues is not a problem in most soils but that injury was often found 6 to 12 months after application of one to four pounds per acre. Similar results were reported by Weldon and Timmons (81) who found that two pounds per acre disappeared in one growing season from a sandy clay loam soil under frequent irrigation but persisted two seasons under less frequent irrigation. Under deep furrow irrigation a two pound application remained phytotoxic to oats planted in the upper two inches of a loamy sand for at least 15 months.

Birk (11) reported 90 percent disappearance of monuron in one year even though the herbicide was concentrated in the upper two inches of soil for most of the growing season. Similar results have been reported by others (23, 30, 63). Loustalot et al. (53) reported that monuron toxicity persisted longer at room temperature than at 10 or 45 degrees centigrade. They also reported longer phytotoxicity at higher rates and under conditions unfavorable for microbial action. Erickson (30) reported 5 to 160 pounds per acre of monuron were toxic to peas and wheat for up to three years.

Although fewer data are available there is some work on the triazines. Holly and Roberts (46) reported that 7 to 27 weeks were required to dissipate 80 percent of an application of two pounds of simazine per acre. Dissipation was more rapid under high rainfall conditions. Sheets and Shaw (72) compared initial and residual phytotoxicities of 14 s-triazines in four soils. The initial effective levels of the methoxy and methylmercapto derivatives varied more among soils than did those of the corresponding chloro derivatives. In most cases methoxy-s-triazines were more toxic to the fifth oat crop than corresponding chloro-s-triazines, whereas the latter exhibited the greatest initial phytotoxicity. They suggested that the variation in results with the methoxy and methyl mercapto derivatives indicated that the availability of each was more affected by soil adsorption than was the chloro derivative. The study was done in the greenhouse with triazines whose solubility varied greatly. Consequently leaching and inactivation by soil adsorption as downward movement occurs may result in different persistence relations under field conditions.

# III. CHARACTERIZATION OF HERBICIDES AND SOIL

To facilitate the use of common names in the discussion, complete chemical descriptions are listed in Table II. Table I is a complete characterization, including microbial analysis, of the soil used in the experiment.

Table I

Soil Characterization[1]

| Parameter | Units | Value |
|---|---|---|
| pH | | 6.40 |
| phosphorus | ppm-P | 10.80 |
| potassium | me/100 g-K | 0.32 |
| calcium | me/100 g-Ca | 8.80 |
| magnesium | me/100 g-Mg | 6.10 |
| sodium | me/100 g -Na | 0.20 |
| total nitrogen | % | 0.054 |
| organic matter | % | 0.40 |
| cation exchange capacity | me/100 g | 21.79 |
| sand | % 50-2000 microns | 55.62 |
| silt | % 2-50 microns | 27.53 |
| clay | % < 2 microns | 16.85 |
| fungi | viable organisms per gram | 4500 |
| actinomycetes and bacteria | viable organisms per gram | 760,000 |

[1] Compiled by Soils Department, Oregon State University, Corvallis, Oregon.

Table II.

Physical and chemical properties of herbicides.

| Common name | Trade name and Manufacturer | Chemical name | Mole wt. | SP. GR. | Vapor pres. mm. Hg at °C | Melting point °C | Solubility at °C ppmw[1] | | State |
|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | Water | Organic solvents | |
| Phenyl Urea Herbicides | | | | | | | | | |
| chloroxuron | tenoran CIBA Corp. | N'-4(4-chloro phenoxy)-phenyl-N, N-dimethyl urea | 290.8 | | | 151-2 | 3.7/20 | mod. in acetone | odorless, white crystalline |
| diuron | karmex E. I. duPont de Nemours and Co. | 3(3, 4-dichloro phenyl) 1, 1 dimethyl urea | 233.1 | | $2x10^{-7}$/30 | 158-9 | 42/25 | 53000 acetone 1200 benzene | odorless, white crystalline |
| fenuron | dybar E. I. duPont de Nemours and Co. | N, N-dimethyl N' phenyl urea | 164.2 | 1.08 20/20 | $1.6x10^{-4}$/60 | 133-4 | 3850/25 | sparing in hydrocarbons | odorless, white crystalline |
| monuron | telvar E. I. duPont de Nemours and Co. | 3-(p-chloro phenyl)-1, 1-dimethylurea | 198.6 | 1.27 20/20 | $5x10^{-7}$/25 | 174-5 | 230/25 | 52000 acetone 2900 benzene | odorless, white crystalline |
| Triazine Herbicides | | | | | | | | | |
| ametryne | ametryne 80w Geigy Agricultural Chemicals | 2 methylthio-4 ethylamino-6 isopropylamino-s-triazine | 227.3 | | $8.4x10^{-7}$/20 | 84-5 | 185/20 | very sol. | white crystalline |

Continued on next page

Table II.
Continued

| Common name | Trade name and Manufacturer | Chemical name | Mole wt. | SP. GR. | Vapor pres. mm. Hg at °C | Melting point °C | Solubility at °C ppmw[1] | | State |
|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | Water | Organic solvents | |
| atrazine | atrazine 80-w Geigy Agricultural Chemicals | 2-chloro-4-ethylamino-6-isopropylamino-s-triazine | 215.7 | | $3.0 \times 10^{-7}$/20 | 173-5 | 70/27 | 52000 chloroform 18000 methanol | white crystalline |
| simazine | simazine 80-w Geigy Agricultural Chemicals | 2-chloro-4,6-bis ethylamino-s-triazine | 201.7 | | $6.1 \times 10^{-9}$/20 | 225-7 | 5/20 | 900 chloroform 400 methanol | white crystalline |
| Uracil Herbicides | | | | | | | | | |
| bromacil | hyvar-x E.I. duPont de Nemours and Co. | 5-bromo-3-sec-butyl-6-methyluracil | 261.1 | 1.55 20/20 | | 158-59 | 815/95 | 167000 acetone 88000 3% aq NaOH | odorless white crystalline |
| terbacil | sinbar E.I. duPont de Nemours and Co. | 3-tert-butyl-5-chloro-6-methyluracil | 216.7 | 1.34 20/20 | | 175-7 | 710/25 | sparingly | odorless white crystalline |

[1] ppmw = parts per million by weight.

# IV. EXPERIMENTAL METHODS AND PROCEDURE

## Sample Preparation and Sampling

Chehalis loam soil was air dried to 1.5 percent moisture and screened through a 20 mesh screen. A kilogram of soil was divided into six equal portions for treatment in Buchner funnels. Each of the herbicides (see Section III, page 24-25) was prepared at 1333 ppm w/v in acetone. Appropriate dilutions were made for two of the diuron treatments. Table III shows the grade of herbicide used.

Table III.

Grade of herbicide used for soil treatment.[1]

| Herbicide | Grade |
|---|---|
| ametryne | Technical |
| atrazine | Technical |
| simazine | Technical |
| chloroxuron | Technical |
| diuron | Recrystallized-technical |
| fenuron | Technical |
| monuron | Recrystallized-pure |
| bromacil | Recrystallized-technical |
| terbacil | Technical |

[1] Bromacil and terbacil obtained from E. I. duPont de Nemours & Co. All other compounds obtained from the Department of Agricultural Chemistry, Oregon State University.

The acetone solution was added, dropwise, to the soil surface in two five-ml aliquots. After the addition of each five-ml portion the soil was stirred and vacuum applied to remove the acetone. When the soil surface was again dry the six portions were combined in a rotary mixing shell and continuously mixed for one hour. This treatment was designed to yield a final concentration of eight ppm w/w in the soil. The first kilogram of soil was treated with diuron at eight ppm and when complete a recovery study was done before any additional soil was treated. The analytical method will be described; however, it was determined that 90 to 92 percent of the diuron theoretically applied could be consistently recovered. Therefore, the application and mixing techniques were considered to be adequate.

Three kilograms were treated for each rate and temperature combination and combined in gallon glass jars with lids. Untreated samples were prepared for each temperature. Although it was considered unlikely that any significant degradation would occur in dry soil, each sample was put in the freezer until all were prepared. After all herbicide-rate-temperature combinations were treated (75 kilograms of soil) all samples were moistened within two days.

The approximate field capacity was determined in a two cm glass column with a moistened glass wool plug by adding water to the top of a column of dry soil until water dripped out the bottom. This was determined to be 44 percent on a v/w basis. The prepared soil

samples were moistened with 500 mls of water per three kilograms (approximately 38 percent of field capacity). The entire sample was done at one time in a rotary conical blender wherein the soil in a revolving container was moistened by an internal nozzle. Afterwards each sample was stored at one of three selected temperatures which were: 6.5, 13.2, and 31.2°C. The lower temperatures were selected because they were readily available in existing storage facilities. The 31.2° temperature was designed to be about twice the 13.2 and it was obtained by using a refrigerator box heated by a thermostatically controlled 60 watt light bulb.

The following herbicide-rate-temperature combinations were employed:

Table IV.

Herbicides, rates and temperatures employed.

| | | Storage Temperature °C | | |
|---|---|---|---|---|
| Herbicide | Desired rate(s) | 6.5 | 13.2 | 31.2 |
| chloroxuron | 8 | | X | X |
| diuron | 2 | X | X | X |
| | 8 | X | X | X |
| fenuron | 8 | | X | X |
| monuron | 8 | | X | X |
| ametryne | 8 | | X | X |
| atrazine | 8 | | X | X |
| simazine | 8 | | X | X |
| bromacil | 8 | | X | X |
| terbacil | 8 | | X | X |
| Untreated | 0 | X | X | X |

When the samples were placed in storage, a zero time sample was removed and frozen. The sampling period approximated 30 days with samples taken on the first day of each month. Three hundred grams (wet weight) were removed, at each sampling, by scooping small portions from several sections of the jar as it was rotated. The sample jars were shaken after sample removal to assure uniform re-mixing of soil adhering to the sides. It was hypothesized that the biological activity of the soil would remain reasonably constant between 25 and 75 percent of field capacity. Therefore, rather than adopt a complicated re-wetting procedure, a simple system was employed. Water was added from a plastic squeeze bottle as needed after sampling. At the low temperature water was added once, while at 13 degrees it was added three times. At the highest temperature marked drying was noted on the edges of jars during the first month. This created problems in sampling. Water was added and the samples were thoroughly mixed. To prevent drying the light bulb was partially shielded with aluminum foil and to maintain a high humidity a gallon jar filled with water was placed on a tripod over the bulb. These procedures decreased drying in the samples adjacent to the bulb and maintained a higher humidity for all samples. Thus, after the first month water was added to these samples a total of three additional times. Because of the difficulty of removing a uniform sample from the gallon jar, it

is recommended, for future experiments, that samples of the appropriate size for analysis be stored in Erlenmeyer flasks and that one flask constitute a sample.

The water content of the soil was not checked during the course of the experiment. When the experiment was terminated the percent of field capacity at each temperature was:

| | | |
|---|---|---|
| 6.5°C | - | 44.5% |
| 13.2°C | - | 50.3% |
| 31.2°C | - | 47.0% |

Initially all samples were frozen after they were taken. As analytical methods were developed this was unnecessary and many samples were allowed to air dry immediately. The initial freezing limited further degradation prior to analysis.

## Analytical Methods

### Phenyl Urea Herbicides

Diuron, fenuron, and monuron. The determination of the aniline fragment formed during quantitative hydrolysis as originally developed by the duPont Co. (21, 54) for monuron and other ureas, was applicable for assaying the phenyl ureas. Basic hydrolysis cleaves the carbonyl nitrogen bond freeing the appropriate aniline

derivative. The aniline is then distilled into an acid solution and detected colorimetrically after diatoziation and coupling.

Reagents required:

a.) Sodium hydroxide - 20% aqueous solution

b.) 6 N hydrochloric acid

c.) 1 N hydrochloric acid

d.) Sodium nitrite - 2% - aqueous solution - prepare fresh daily

e.) Sulfamic acid - 10% - aqueous solution - prepare fresh daily

f.) N-(1-napthylethylenediamine dihydrochloride) - 2% - aqueous solution - prepare fresh daily

g.) Glacial acetic acid

Prior to analysis standard curves were prepared from analytical standards of aniline (fenuron), p-chloroaniline (monuron) and 3,4-dichloroaniline (diuron). One thousand microgram/ml solutions of each aniline were prepared by dissolving 0.5 grams in 500 ml of 1.0 N HCl. By dilution, solutions containing 10 micrograms/ml were obtained. Aliquots of these solutions containing 5, 10, 20, 30, 40, and 50 micrograms were quantitatively transferred to 50 ml volumetric flasks. Ten ml of 1.0 N HCl and 5 ml of glacial acetic acid were added. One ml of 2% sodium nitrite was added and the flasks left at room temperature (with occasional shaking) for ten

minutes. After the diazotization period one ml of sulfamic acid was added, the flasks were stoppered, and mixed gently over a ten minute period to assure destruction of excess nitrite. Two ml of N-(1-napthylethylenediamine dihydrochloride), hereinafter referred to as the coupler, were added and the following times were allowed for the development of the magenta color:

| | |
|---|---|
| aniline | 90 minutes |
| p-chloroaniline | 30 minutes |
| 3,4-dichloroaniline | 15 minutes |

These time periods were followed but it was found that minor deviations were of little consequence. After full color development the samples were read on a Klett-Summerson photoelectric colorimeter with a one cm cell, filter number 56 (wave length range 540-590 millimicrons) and a blank similar in every respect but without aniline. The standard curves prepared for these herbicides are shown in Figure 1.

Samples of soil taken monthly were air dried and stirred so no large lumps were present. Depending on the level of activity expected 50 or 100 grams of soil were placed in a 1000 ml flat bottom boiling flask. About 350 ml of 20 percent sodium hydroxide and a few boiling chips were added. Experience showed that foaming was not a problem with this soil so no anti-foam agent was used. Six

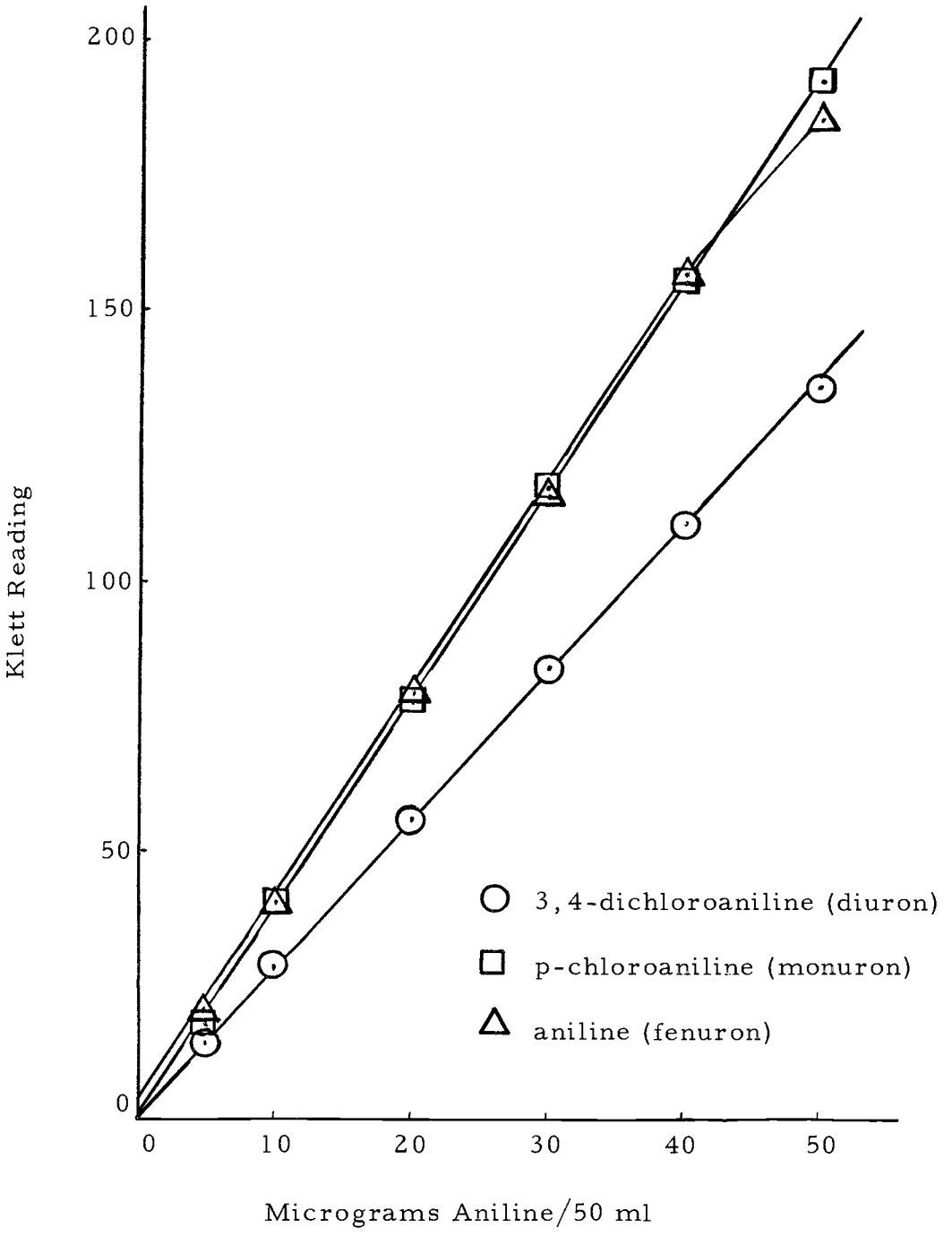


Figure 1. Standard calibration curve for aniline, p-chloroaniline and 3,4-dichloroaniline.

samples were prepared at one time and refluxed on a large hot plate for four hours from the time boiling began. After this period 60 ml was distilled into 10 ml of 6 N hydrochloric acid solution. This solution was quantitatively transferred to a 100 ml volumetric flask and aliquots of it were tested for their aniline content, as outlined under preparation of the standard curves. Experience showed that there was no gain in efficiency or accuracy of results if a chromatographic clean up step using Whatman cellulose columns was included after coupling.

The amount of phenyl urea in the original soil samples was calculated with the following equation:

$$\frac{\mu g}{\text{soil}}\text{gm} = \frac{\text{Klett reading}}{\text{slope of standard curve}} \times \text{Molecular weight factor} \times K\,(-\text{background}) \times 90\%\ \text{recovery}$$

$$K = \text{Dilution factor} = \frac{100\ \text{ml}}{\text{ml of distillate analyzed}}$$

$$\frac{\text{M. W.}}{\text{factor}} = \frac{\text{molecular weight of herbicide}}{\text{molecular weight of aniline derivative}}$$

Table V shows the molecular weight factors and the slope of the standard curve used for each of the three herbicides.

Table V.

Molecular weight factor and slope values for diuron, fenuron and monuron.

| Herbicide | Molecular Weight factor | Slope of Standard curve |
|---|---|---|
| diuron | 1.44 | 2.83 |
| fenuron | 1.558 | 3.87 |
| monuron | 1.76 | 3.99 |

Chloroxuron. The general procedure was the same as that for the previously described compounds, i.e. hydrolysis to the aniline and detection by diazotization and coupling to form a colored compound. However, repeated recovery studies with the parent compound and the 4-amino,4'-chlorodiphenylether, using distillation of the aniline after five hour hydrolysis, gave only 60-70 percent recovery when the hydrolyzed samples were compared with a non-hydrolyzed series of standards. Therefore, it was concluded that this aniline was less volatile than those from diuron, fenuron or monuron. To circumvent this, the basic solution was allowed to cool after hydrolysis and then positioned on an A-frame apparatus for overnight liquid-liquid distillation and extraction by n-hexane. The aniline was extracted with 3 N hydrochloric acid and the color test completed as before. Again, after trials with minor variation, recovery was poor. However, when the samples prepared in this

way were compared with a series of standards which had also been hydrolyzed, 90 percent recovery was consistently obtained. The same percentage could be recovered from spiked soil samples and this method was subsequently adopted. Although this method does give 90 percent recovery it does not present a wholly accurate picture because losses of the aniline occur in some unknown way. If such losses did not occur, the hydrolyzed and unhydrolyzed standard curves would be in very close agreement. The losses may be due to volatilization but the exact cause was not investigated.

With the exception of the substitution of hexane extraction for distillation and the subsequent acid extraction of the hexane, the procedure was the same as those outlined in the previous section. The color test was modified by using 10 instead of 5 ml of glacial acetic acid. This was found to give more stability to the final purple color. Color development time was two hours and fifteen minutes. If the color development period was shorter or longer than this a straight line relationship was still obtained but this time gave a line which passed through the origin and most closely adhered to Beer's law.

The molecular weight factor for chloroxuron is 1.323 and the slope of the standard curve shown in Figure 2 is 1.73.

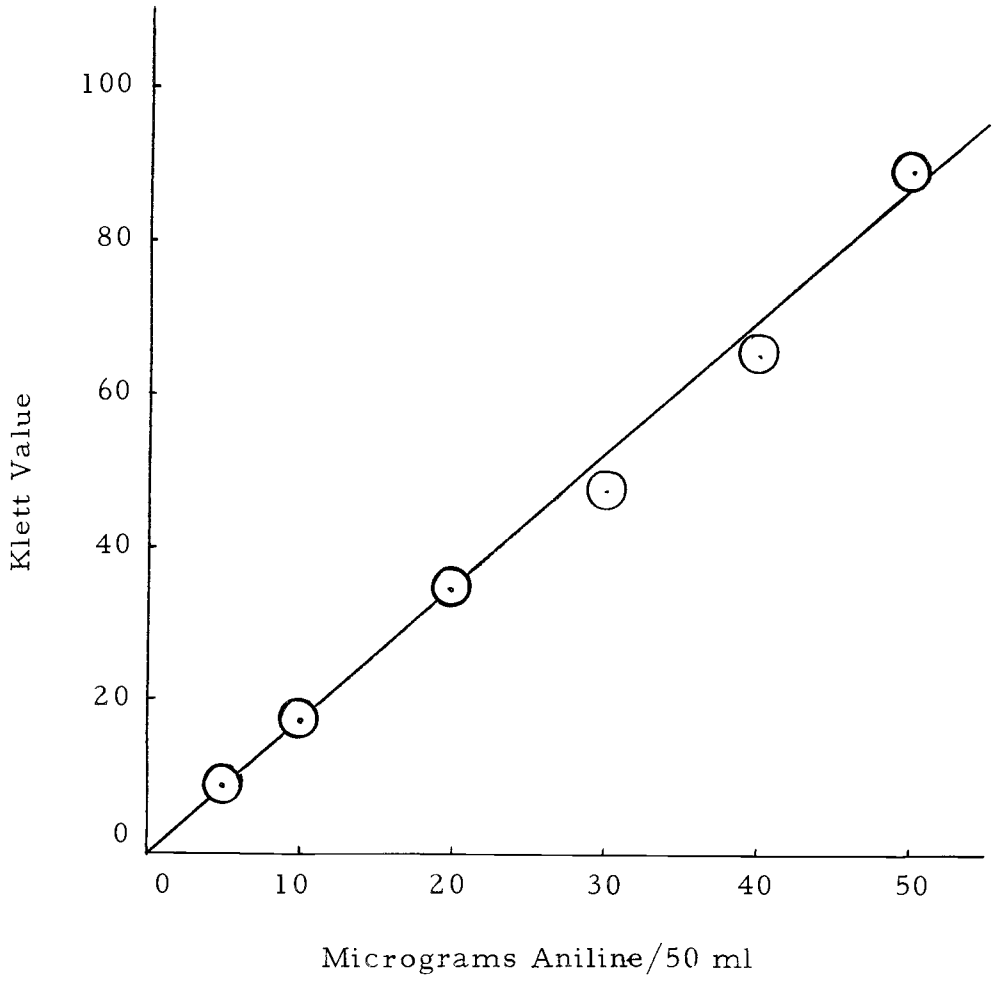


Figure 2. Standard calibration curve for 4-amino-4'-chlorodiphenyl ether (chloroxuron).

## Triazine Herbicides

Ametryne. Gas-chromatography offers a specific method for determining individual members of the triazine herbicide group and has been employed for each of the triazines in this experiment. The procedure for ametryne was based on the work of Mattson et al. (56).

The volatility of ametryne was of concern so the soil was analyzed without drying. The degree of saturation of the soil varied and was determined with a duplicate sample. The reason for this variation was that samples had been taken for four months before a method of extraction and detection was developed. In the interim samples were frozen and some freeze drying occurred.

Reagents required:

a.) Concentrated ammonium hydroxide

b.) Distilled ethyl ether

c.) Anhydrous sodium sulfate

d.) Distilled benzene

e.) Distilled hexane

f.) Woelm Activity Grade I basic alumina

Twenty-five grams of moist soil were placed in a 250 ml glass centrifuge bottle. To this was added one ml of concentrated ammonium hydroxide and 50 ml of distilled ethyl ether. The bottle was

capped and placed on an oscillating shaker for five minutes. This procedure was repeated two additional times (the $NH_4OH$ may be omitted the third time). After each period of shaking the ether was decanted through sodium sulfate into a 250 ml beaker. At this point the combined ether extracts may be passed through a column of sodium sulfate to remove any remaining water. Using a stream of nitrogen gas and warming on a steam bath the ether extract was evaporated to 10 ml. Then 3-4 ml of benzene were added with rinsing of the sides and evaporation continued, without heat, to about one ml. The sample in benzene was transferred quantitatively to a column containing 12 grams of 15% basic alumina. Hexane rinses were used to facilitate the transfer and wash the sample into the column. The column was washed with 75 ml of n-hexane which did not elute triazines. Ametryne was eluted with 150 ml of a mixture of 1:1 benzene and hexane (v/v). Using a stream of nitrogen gas or a rotary evaporator the eluate was evaporated to a volume suitable for chromatography.

A Dohrmann microcoulometric gas chromatograph model C-200 equipped with a T-200-S titration cell sensitive to sulfur was used. A five foot by one-quarter inch pyrex column packed with ten percent Dow 11 on 60/80 gas chrom Q proved to be satisfactory. The flow rate of nitrogen gas was 200 ml/minute at a setting of eight. A resistance of 200 ohms and a temperature of 165°C were

used. Injection volumes of two to ten microliters were routinely employed and at no time was there any sulfur interference in the soil blanks. It was usually necessary to condition the column with the standard until constant recovery (expressed as peak height) was obtained. Peak height was used as the basis for calculation. Retention time was three and one half minutes. Ten micrograms/ml was the concentration of the standard and two successive two microliter injections were made before every sample.

This extraction and detection procedure gave consistent 90 percent recovery in the range of one to ten ppm of ametryne in soil.

Atrazine and simazine. The Dohrmann microcoulometric gas chromatograph model C-100 with a titration cell sensitive to halides was used for each of these herbicides. The extraction and clean-up steps were almost identical and were those used by the Department of Agricultural Chemistry at Oregon State University[1]. McGlamary et al. (58) studied methods for the extraction and determination of atrazine from soil and found that the extraction procedure to be described was very satisfactory.

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[1] Personal communication from Marvin L. Montgomery and Eugene R. Johnson, Department of Agricultural Chemistry, Oregon State University, Corvallis, Oregon.

Reagents required:

a.) Distilled methanol - (atrazine)

b.) Distilled chloroform - (simazine)

c.) Distilled benzene

d.) Distilled hexane

e.) Carbon tetrachloride - reagent grade

f.) Distilled ether

g.) Woelm Activity Grade I - 15% basic alumina

Fifty or 100 gram samples of air dried, pulverized soil were weighed into asbestos extraction thimbles. These were placed in a soxhlet apparatus and extracted for 24 hours with distilled methanol for atrazine and chloroform for simazine. The difference in initial extracting solvent was the only difference in procedure. After extraction, the samples were cooled, placed on a rotary evaporator and evaporated almost to dryness. The flask was then rinsed with 25-50 ml of benzene and extracted water appeared as milkly droplets. The cycle of benzene rinse and evaporation was repeated until the solution was clear. Water was less of a problem with the chloroform extraction because it is not soluble in chloroform. When the water had evaporated the benzene was evaporated to about 10 ml and quantitatively transferred to a 50 ml beaker with benzene rinses. Using a steam bath and a stream of nitrogen gas this solution was evaporated to a volume of approximately 1 ml. This was quantitatively

transferred to a column prepared with 12 grams of 15 percent basic alumina. The sample was washed into the column with a small amount of n-hexane. The column was then washed successively with 50 ml of reagent grade carbon tetrachloride and 50 ml of distilled n-hexane. These were discarded. The triazine was eluted with 30 ml of a 60: 40 mixture of benzene and ether (v/v). This was evaporated to a volume appropriate for chromatography.

A Dohrmann microcoulometric gas chromatograph model C-100 equipped with a model T-200-S halide sensitive titration cell was used for detection. A three and one-half foot pyrex column (4 mm inside diameter) was packed with five percent SE-30 on 60/80 mesh gas chrom Q. The gas flow rate was 116 ml of nitrogen per minute at a setting of 40 on a Brooks-sho-rate R-2-15-AAA flow meter with a tank pressure of 40 psig. A resistance of 32 ohms and a temperature of 150$^{o}$C gave a retention time of 160 seconds for atrazine and 165 for simazine, for injection volumes of 20 to 70 microliters. A concentration of 100 micrograms per ml was used in the standard and 20 microliters were injected. The determination of triazine present was made by comparing integrator strokes of frequently injected standards with those obtained from unknown samples. No chlorine interference was detected. The procedure gave 95 percent recovery of both triazines.

Uracil Herbicides

The methods of extraction, clean-up, and detection of the uracil herbicides were adapted from three sources (47, 61) and [2]. The method outlined in reference (61) used programmed temperature gas chromatography, which was unavailable. Joliffe et al. (47) used partitioning into ethyl acetate, a step which was dropped.

Reagents required:

a.) Sodium hydroxide - 1.5% aqueous solution

b.) Sulfuric acid - 10.0 N

c.) Distilled chloroform

d.) Distilled n-hexane

e.) Distilled acetone

Fifty grams of air dried, pulverized soil were placed in a 250 ml centrifuge bottle and 90 ml of 1.5 percent sodium hydroxide was added. This mixture was shaken for 15 minutes on an oscillating table shaker. After shaking the mixture was centrifuged for ten minutes at 2000 rpm. The cloudy supernatant was decanted through a glass wool plug into a 250 ml separatory funnel. The soil was re-suspended, with the aid of an omni mixer, in 50 ml of 1.5 percent

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[2] Personal communication: Marvin L. Montgomery, Department of Agricultural Chemistry, Oregon State University, Corvallis, Oregon.

sodium hydroxide. Samples were shaken for five minutes, centrifuged, and the cloudy supernatant decanted through the glass wool plug into the separatory funnel. The combined extracts were acidified with 12 ml of 10 N sulfuric acid. It is important that the extract be acidic at this point. The acidified samples were then extracted successively with 90 and 50 ml of distilled chloroform. With careful shaking emulsification can be prevented on the first extraction whereas the second will emulsify. The emulsion was broken by centrifugation and the chloroform extracts combined in a separatory funnel. The chloroform was extracted twice with 75 ml of 1.5 percent sodium hydroxide and the base was extracted once with n-hexane which was discarded. Twelve ml of 10 N sulfuric acid were used to acidify the basic solution and the uracils were extracted with two 75 aliquots of chloroform and collected in a 250 ml round bottom flask. The chloroform was then evaporated to dryness on a rotary evaporator. The flask was rinsed with 25 ml of acetone and again taken to dryness. This step was repeated a minimum of two additional times, to assure removal of the chloroform. After the final acetone rinse the yellow solution was quantitatively transferred to the appropriate volume for chromatographic detection. All traces of chloroform must be removed because it will interfere with proper detection. This procedure gave better than 85 percent recovery with no soil interference for either herbicide.

Up to this point the extraction procedures for bromacil and terbacil were identical. Their final detection was not. Terbacil was detected with a Dohrmann microcoulometric gas chromatograph model C-100 equipped with a model T-200-S halide sensitive titration cell. The column, temperature, and gas flow rate were all identical to those used for atrazine. With an injection of 20 microliters of a 100 ppm standard a resistance of 16 ohms proved to be satisfactory. The retention time was 175 seconds for injection volumes of 20 to 80 microliters. The determination of terbacil present was made by comparing integrator strokes of frequently injected standards with those obtained from the unknown samples.

Initially the Dohrmann microcoulometer was to be used for the detection of bromacil. However, it was found that the determinations were non-stoichometric. The apparent low conversion was due to the fact that elemental bromine was formed in the oxidation rather then hydrogen bromide. Subsequent solvation in the electrolyte yields one mole of hypobromous acid (non-titratable). Therefore, the Wilkens Aerograph Hy-Fi, oven model 550 with Electrometer 500-D, electron capture gas chromatograph was used for bromacil detection. A 12 inch column with a 4 mm inside diameter was packed with a one inch plug of carbowax on the cell end followed by 10 inches of 5 percent SE-30 on 60/80 mesh gas chrom Q. The flow rate of nitrogen gas was approximately 50 ml per minute. The

oven temperature was 175° C and the range setting was 1.0 with an attenuation of 4. The total standing current was 2500 units and no more than ten percent of it was used. Retention times varied somewhat with different analyses due to the vagaries of the instrument but averaged 0.15 minutes. Two microliters of a 0.5 ppm standard solution in acetone were used. Bromacil was determined by comparing relative peak heights from injections of 2 to 8 ml of the unknown samples with the peak height of frequently injected standards.

## Derivation of Rate Laws and the Arrehenius Equation[3]

In chemical kinetics the term rate of reaction refers to the time rate of change, dc/dt, of the concentration of one of the reactants or products. The reaction rate constant k is a proportionality factor which relates the rate of reaction to the concentration of the reactants (22). The factor k may be determined graphically or mathematically. To treat reaction rates in a quantitative manner it is necessary to consider reactions from the standpoint of order (76). If no concentration terms appear in the kinetic equation a reaction is said to be of zero order. The rate of such a reaction is determined by a limiting factor other than concentration. An illustration of a zero order reaction is one where the rate is determined

[3] Much of the material in this section was obtained from references (19, 22).

by the quanta of light received by a reaction mixture per unit time or a reaction whose rate is determined by exposure to a specific catalyst. The equations for reaction speed are:

$$dx/dt = k \qquad (48\text{-}1)$$

$$x = kt + I \qquad (48\text{-}2)$$

$x$ = amount of product formed in a unit of time in a unit volume.

$k$ = velocity or rate constant

$I$ = constant of integration

A first order reaction is one whose rate is proportional to the first power of a reacting substance and independent of the concentration of all other substances that may be present, including the product (19). This is expressed by the simple mathematical relation

$$\text{Rate} = -\frac{da}{dt} = ka \qquad (48\text{-}3)$$

$da$ is the number of moles of reactant undergoing reaction/unit volume in a small amount of time $dt$. $k$ is the specific rate constant which considers the nature of the reacting substance, the temperature, and any other rate limiting or accelerating factors.

Integration of equation 48-3 on rearranging gives 49-1.

$$-\frac{da}{a} = kdt \qquad (49\text{-}1)$$

which gives,

$$\log a = -\frac{kt}{2.303} + I \qquad (49\text{-}2)$$

If equation 49-2 is integrated between the limits time = $\underline{0}$ and time = $\underline{t}$ with the appropriate concentration values of $\underline{a}^{o}$ and $\underline{a}$ equation 49-3 results

$$\log \frac{a^{o}}{a} = \frac{kt}{2.303} \qquad (49\text{-}3)$$

$a^{o}$ = initial concentration

$a$ = concentration at time $\underline{t}$

A first order reaction will obey these equations and this can be checked by substituting the value of $\underline{k}$ for a number of values of $\underline{t}$ and $\underline{a}$, nearly constant values should result. A plot of the logarithm of $\underline{C}$ against $\underline{t}$ will give a straight line if the reaction is first order. A third check is to use the half-life method. The half-life of a reaction is the time that must elapse before half the initial concentration has been dissipated. When this has occurred $\underline{a}$ is equal to $a^{o}/2$ . If this substitution is made in equation 49-3 equation 50-1 results.

$$\log \frac{a^o}{a^o/2} = \frac{kt_{1/2}}{2.303} \qquad (50\text{-}1)$$

This reduces to

$$\log 2 = \frac{kt_{1/2}}{2.303} \qquad (50\text{-}2)$$

or

$$t_{1/2} = \frac{2.303 \log 2}{k} = \frac{0.693}{k} \qquad (50\text{-}3)$$

Therefore, the half life is independent of initial concentration and if similar values are obtained for a number of different initial concentrations, the reaction is of first order.

In two concentration terms appear in the reaction equation and each is raised to the first power the reaction is second order. However, if only one concentration term appears and it is raised to the second power, the reaction is also second order. This situation gives rise to two second order equations. If the two reacting molecules are the same the equation is:

$$-\frac{da}{dt} = ka^2 \qquad (50\text{-}4)$$

When the molecules are different the equation is:

$$-\frac{da}{dt} = -\frac{db}{dt} = k\ (a \times b) \qquad (51\text{-}1)$$

If a = b then equation 51-1 becomes identical with 50-4. For purposes of simplification only this situation will be considered.

Integration of equation 50-4 gives two equations which correspond to those for first order reactions.

$$\frac{1}{a} = kt + I \qquad (51\text{-}2)$$

$$\frac{a^o - a}{a^o a} = kt \qquad (51\text{-}3)$$

A second order reaction must obey these equations and adherence to them can be checked by substituting values of k for sets of t and a data or by plotting the reciprocal of the concentration against t. A straight line confirms a second order reaction. A third check is provided by the half life method. Substitution of $a^o/2$ for a in equation 51-3 shows that the half life period is a function of the initial concentration.

$$\frac{a^o - a^o/2}{a^o \times a^o/2} = kt_{1/2} \qquad (51\text{-}4)$$

$$t_{1/2} = \frac{1}{a^o k} \qquad (51\text{-}5)$$

Therefore if similar half life periods are determined for two different initial concentrations and these periods are inversely proportional to the initial concentrations, the reaction is second order. The initial assumption of similarity of reacting molecules or initial concentrations is to be noted. Half lives for different reactants would be different.

The rate determining steps of most reactions are usually of first or second order. There are also third order reactions where k has the dimensions mole $^{-2}$ liter$^{2}$ sec$^{-1}$ as shown in equations 52-1 and 52-2.

$$-\frac{da}{dt} = k[A]^3 \qquad (52\text{-}1)$$

$$k_3 = \frac{1}{2t}\left(\frac{1}{[A]_t^2} - \frac{1}{[A]_o^2}\right) \qquad (52\text{-}2)$$

The $t_{i/2}$ for these reactions is dependent on a factor of $(\frac{1}{\text{initial concentration}})^2$. From the kinetic viewpoint most reactions are not simple but rather complex with a reaction mechanism consisting of several successive steps, each of which will normally be first or second order. Complications arise when reactions proceed to an equilibrium state appreciably short of completion and the reverse reaction becomes important. Fractional orders are also known but the derivation of appropriate equations for such complex

situations is attended with several problems.

The order of a chemical reaction should be regarded as a mathematical convenience, not as a fundamental property (76). The order can be changed by altering reaction conditions.

Evaluation of the rate constant $\underline{k}$ at different temperatures permits the calculation of the heat or energy of activation ($\Delta$ Ha*). This value is interpreted to be a measure of the amount of energy molecules must have in order to react. It is expressed in calories or kilocalories per mole. The dependence of many rate constants on temperature can be expressed in terms of the Arrehenius equation:

$$k = Ae^{-E/RT} \qquad (53\text{-}1)$$

A = Arrehenius parameter

H or E = Arrehenius activation energy

R = gas constant 1.987 Cal mole$^{-1}$ degree$^{-1}$

T = absolute temperature

e = base of natural logarithms

The relation of $\underline{k}$ and energy of activation can alternately be expressed as:

$$\frac{d(\ln k)}{dt} = \frac{\Delta Ha^*}{RT^2} \qquad (53\text{-}2)$$

$$\ln k = \frac{-\Delta Ha^*}{RT} + I \qquad (54\text{-}1)$$

$$\log \frac{k_2}{k_1} = \frac{\Delta Ha^*}{2.303\ R} \quad \frac{\Delta T}{T_1 \times T_2} \qquad (54\text{-}2)$$

The activation energy of a known or an unknown reaction can be calculated from the Arrehenius equation if the rate constant, $\underline{k}$, has been determined at two different temperatures and is a parameter of interest in the discussion to follow.

# V. RESULTS AND DISCUSSION

The purposes of this study were to determine whether the degradation of several herbicides followed a first order rate law in the soil, and if the rate and associated activation energy could be related to molecular structure. These two goals have been achieved and the data have provided evidence for some additional hypotheses as well as several new problems. The experiments were not designed to ascertain the pathways of degradation or to identify metabolites. The data to be discussed are not offered as precise parameters but are close approximations of the actual values. This thesis represents a first step in the examination of degradation from a kinetic point of view. More precise determinations of rates and energies of activation should now be possible.

## Phenyl Urea Herbicides

One definition of a first order reaction is that a plot of the log of concentration vs. time gives a straight line. Figure 3 shows the relative rates of degradation of four phenyl urea herbicides applied at eight ppm and stored at 31.2 degrees centigrade. Although the data vary due to inherent analytical variation and inaccuracies in sampling, Figure 3 indicates a first order rate of reaction. A second point of interest in Figure 3 is the origin of the curves. With

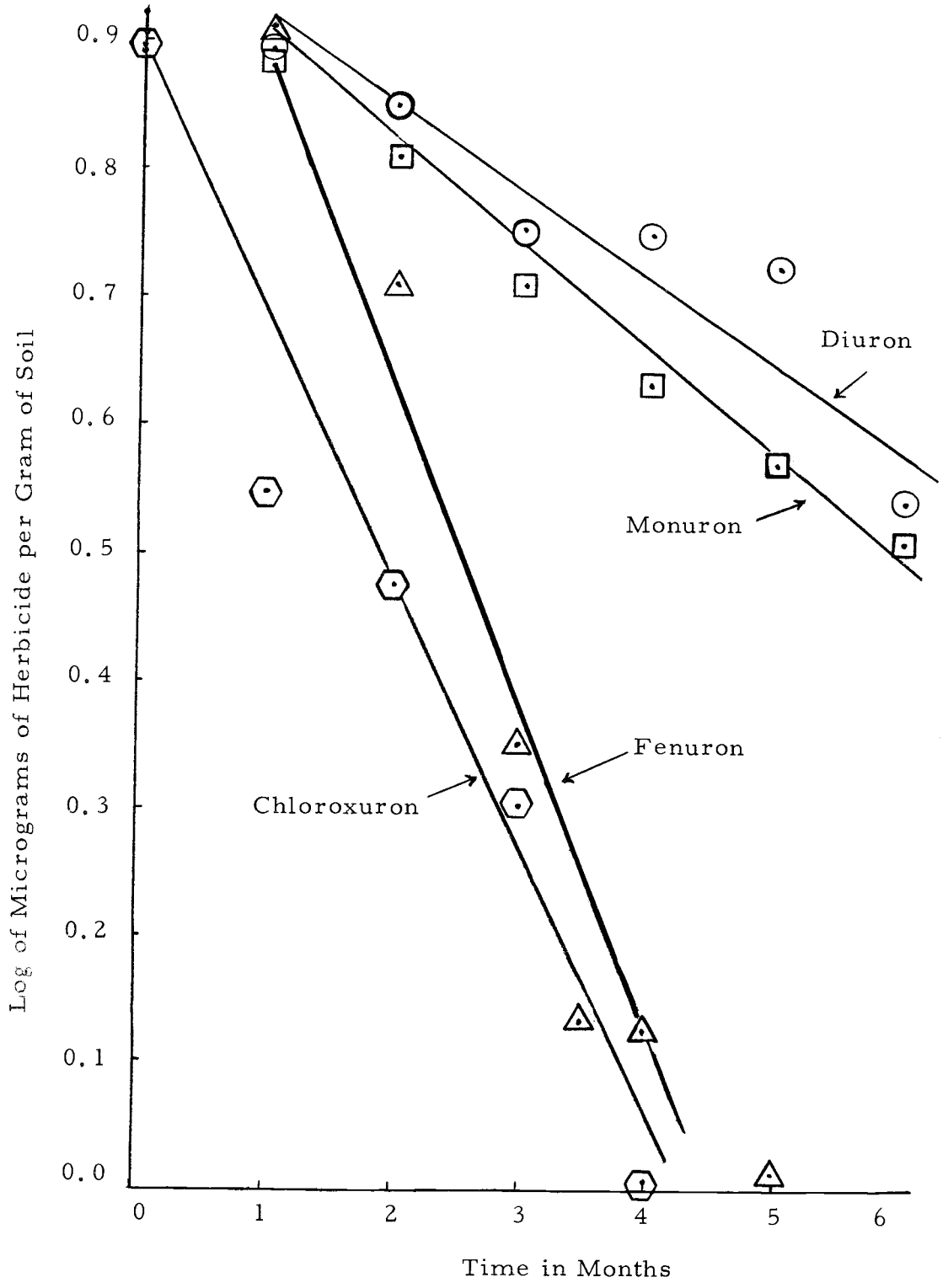


Figure 3. The rate of degradation of phenyl urea herbicides applied to soil at eight ppm and stored at 31.2°C.

the exception of chloroxuron, all of the curves originate at one month rather than time zero. This is indicative of a lag period in the microbiological degradation of these molecules. It is postulated that the herbicides may initiate or prolong an adaptive phase in the microorganisms giving rise to the lag period. The exact length of the lag period was not defined for any of the herbicides. The data indicated it was about the same but this may not be true. Chloroxuron did not show a lag period. The data do not support the conclusion that there was not a lag period. If the experiment was repeated with a shorter sampling interval, it is possible that a lag period would be found for chloroxuron. This would also permit more precise definition of the length of this period for the other molecules.

Similar observations were made on the rate of degradation at 13.2 degrees centigrade, as shown in Figure 4. Because of the proximity of the curves they are separated, but do present the same evidence, i.e., a differential rate and a lag period.

The indication of a lag period creates a question concerning the validity of employing a half life value as an estimate of precise herbicide persistence. Grover (38) has suggested that the half life value would be useful only after the first order reaction had begun and any estimate prior to this would be somewhat distorted. For field application this error may not be of serious consequence. Figure 5 shows the rate of degradation of diuron when applied to the

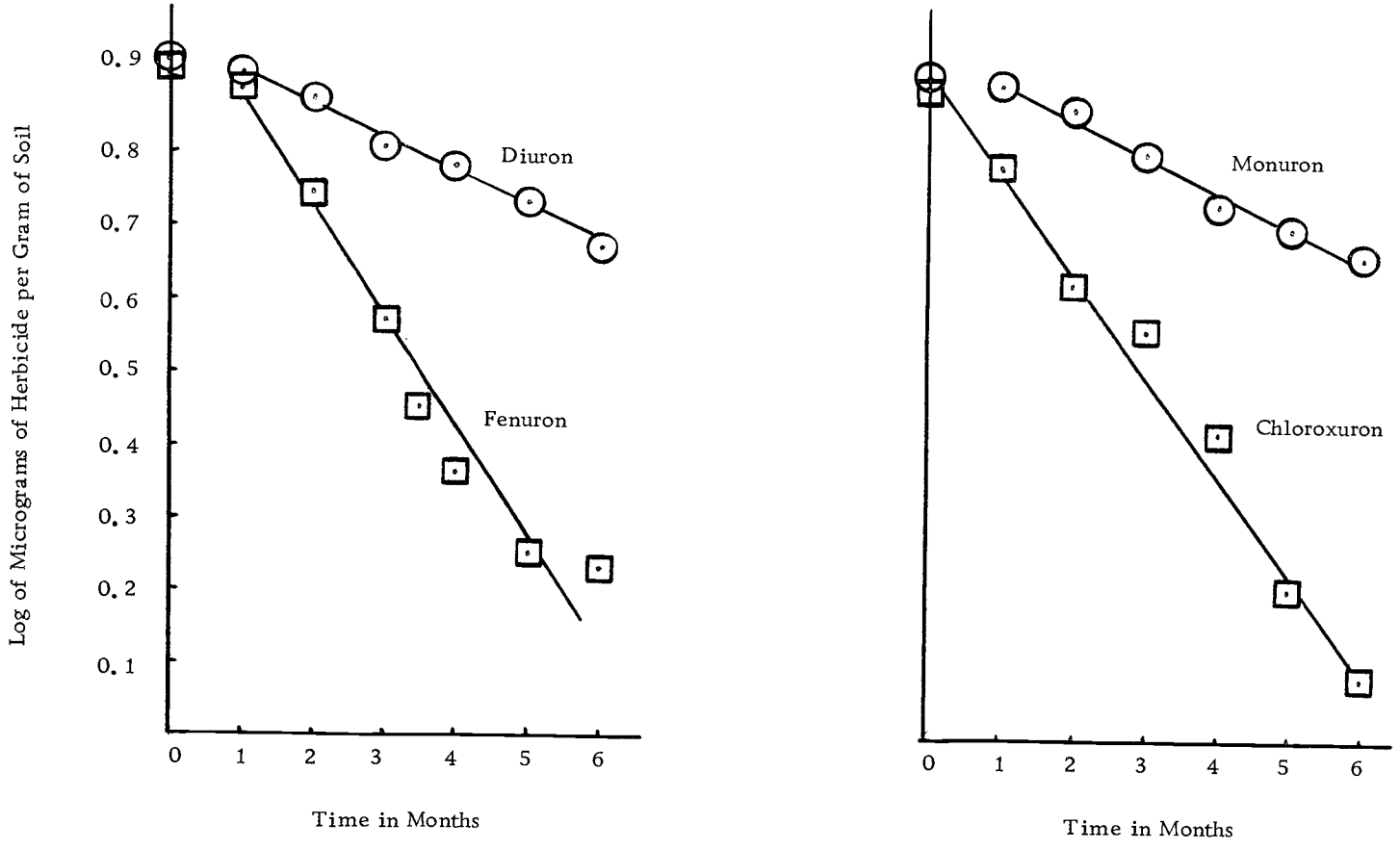


Figure 4. The rate of degradation of phenyl urea herbicides applied to soil at eight ppm and stored at 13.2°C.

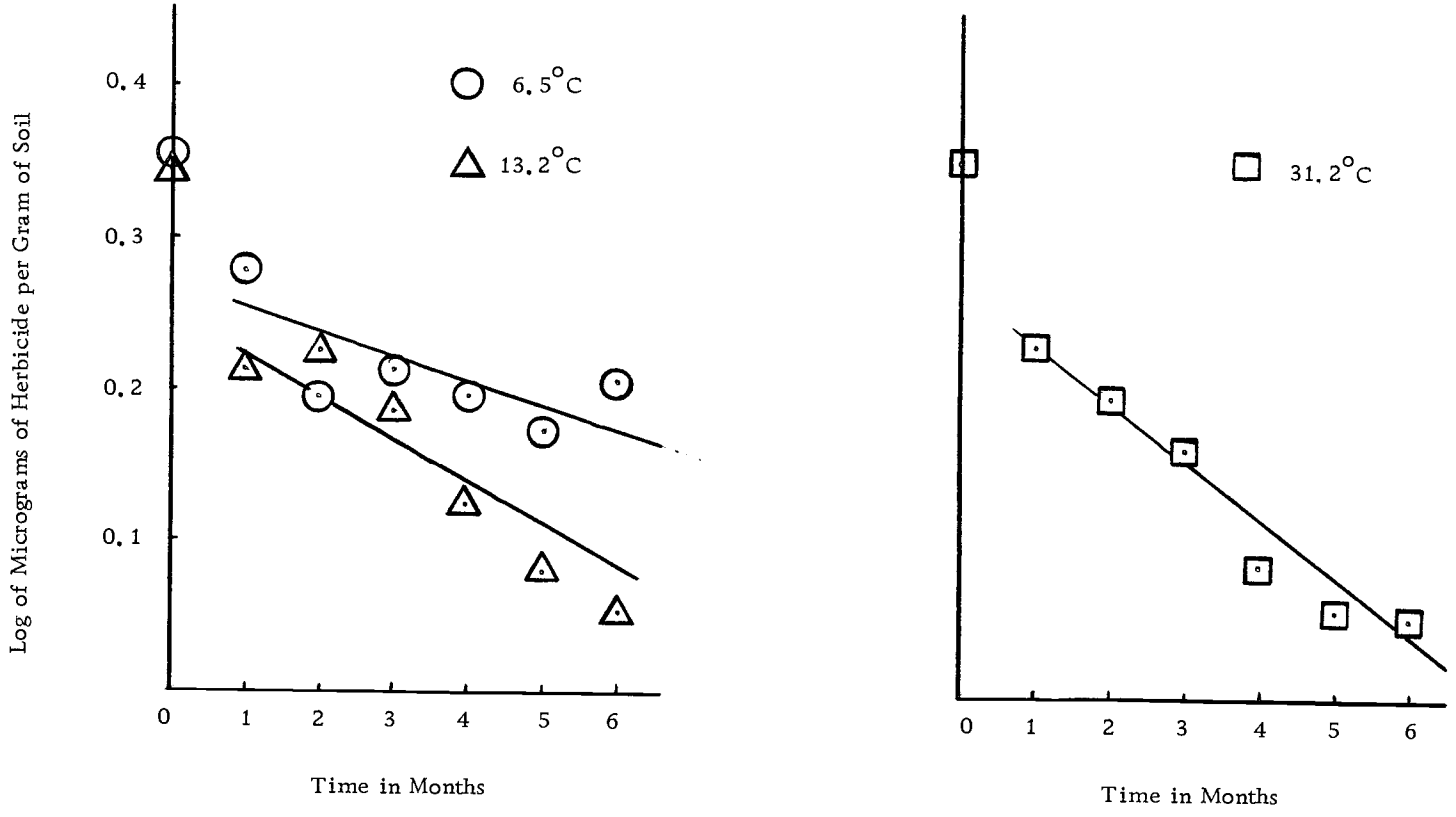


Figure 5. The rate of degradation of diuron applied to soil at two ppm and stored at 6.5, 13.2 and 31.2°C.

soil at two ppm and stored at 6.5, 13.2 and 31.2 degrees centigrade. These data show an increasing rate with increasing temperature but they do not show a lag period. It is possible that a lag period was not detected with a one month sampling interval. However, it is also possible that there was an effect of concentration on the duration of the lag period.

Table VI shows the results of graphic determination of the rates of degradation for the phenyl urea herbicides. These rates were found to be consistent with temperature. The rate could not be related to water solubility as shown in Table VI. Geissbühler et al. (35) found the adsorptive effect of clay to be more important for urea herbicides of low solubility, such as chloroxuron, than for more soluble ones. However, the data in Table VI do not support the hypothesis of a specific interaction between sorption, water solubility and the rate of degradation. The rate of degradation is attenuated by sorption and is dependent on the rate of microbial or enzymatic attack which can be hindered by molecular structure and conformation. Therefore, it is proposed that the rate of degradation is probably attenuated by the chlorine atom(s) of diuron and monuron and the 4'-chloro-biphenyl ether linkage of chloroxuron as opposed to the N-phenyl group of fenuron. (See Appendix Table I for molecular structures.) Although the reaction cannot be defined it can best be envisioned as an affect of the chlorine atom(s) on the formation of

the enzyme-substrate complex.

Table VI

The rate of degradation of four phenyl urea herbicides in soil.

| Herbicide and initial concentration in soil | Rate of degradation at storage temperature (°C) | | | Water Solubility ppm |
|---|---|---|---|---|
| | 6.5 | 13.2 | 31.2 | |
| diuron | | | | 42.0 |
| 2 ppm | 0.0935 | 0.1217 | 0.1891 | |
| 8 ppm | 0.1413 | 0.1870 | 0.2391 | |
| monuron | | | | 230.0 |
| 8 ppm | | 0.2087 | 0.3543 | |
| chloroxuron | | | | 3.7 |
| 8 ppm | | 0.5543 | 0.8391 | |
| fenuron | | | | 3850.0 |
| 8 ppm | | 0.6739 | 1.0891 | |

Table VII is another presentation of the data concerning the rate of degradation of the phenyl ureas. The percentage of herbicide remaining in the soil at each monthly interval shows the variation in the rate of breakdown with temperature. The data for diuron show that within reasonable limits the half life at two rates at a given temperature was nearly the same. This gives further support to the hypothesis of a first order rate of breakdown.

Table VII

The percentage of phenyl urea herbicides remaining in the soil at different times and storage temperatures.

| Herbicide | Storage temperature °C | Initial conc. ppm | Percent of original concentration remaining, months after treatment | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | | 1 | 2 | 3 | 4 | 5 | 6 |
| diuron | 6.5 | 2 | 84.4 | 69.8 | 72.4 | 69.8 | 66.2 | 71.6 |
| | 13.2 | 2 | 73.5 | 75.3 | 68.6 | 59.6 | 53.8 | 50.7 |
| | 31.2 | 2 | 75.6 | 70.2 | 64.9 | 54.2 | 50.7 | 50.2 |
| diuron | 6.5 | 8 | 100.5 | 93.8 | 88.2 | 81.3 | 82.5 | 72.5 |
| | 13.2 | 8 | 102.0 | 94.6 | 81.1 | 76.5 | 67.7 | 59.7 |
| | 31.2 | 8 | 101.2 | 88.1 | 70.3 | 69.7 | 66.1 | 43.2 |
| monuron | 13.2 | 8 | 97.4 | 91.1 | 79.3 | 67.6 | 62.7 | 57.5 |
| | 31.2 | 8 | 99.4 | 82.2 | 64.9 | 54.4 | 47.3 | 41.1 |
| chloroxuron | 13.2 | 8 | 78.1 | 54.1 | 46.6 | 33.8 | 20.6 | 15.7 |
| | 31.2 | 8 | 45.4 | 38.5 | 25.8 | 13.8 | 7.8 | 4.9 |
| fenuron | 13.2 | 8 | 91.2 | 65.8 | 43.9 | 27.2 | 21.1 | 20.0 |
| | 31.2 | 8 | 92.0 | 62.2 | 28.1 | 16.2 | 12.5 | 3.4 |

Dalton (20) and Sheets and Smith (73) have identified the pathway of phenyl urea degradation in plants and soil. The first step proposed by these workers was N-demethylation to the mono-methyl phenyl urea. This hypothesis is supported by the activation energies obtained from the Arrhenius equation (Chapter IV, page 53). Table VIII shows the similarity of activation energies ($\Delta$ Ha*) obtained for each of the phenyl urea herbicides. Thus, it is hypothesized that these data support the existing evidence for a common mechanism of breakdown, specifically, N-demethylation. It is recognized, that sorption, and hindrance from the phenyl end of the molecule will

influence the rate at which degradation proceeds but this will not influence the finite amount of energy required to cleave identical N-methyl bonds.

Table VIII

The heat of activation for the degradation of four phenyl urea herbicides in soil.

| Herbicide | Approximate initial concentration in soil (ppm) | Storage temperatures compared ($^{o}$C) | Average energy of activation ($\Delta$ Ha*) Cals/mole |
|---|---|---|---|
| diuron | 2 | 6.5 : 13.2 | |
| diuron | 2 | 13.2 : 31.2 | 5031 |
| diuron | 2 | 6.5 : 31.2 | |
| diuron | 8 | 6.5 : 13.2 | |
| diuron | 8 | 13.2 : 31.2 | 5243 |
| diuron | 8 | 6.5 : 31.2 | |
| monuron | 8 | 13.2 : 31.2 | 5093 |
| chloroxuron | 8 | 13.2 : 31.2 | 4136 |
| fenuron | 8 | 13.2 : 31.2 | 4614 |

Although bond energies indicate the theoretical amount of energy resident in a given bond they do not necessarily relate to the likelihood of attack by a catalytic organism or to the energy of activation. The point of attack may also be directed by sorption, hindrance of formation of the enzyme-substrate complex and the availability of a given site. However in this case the N-methyl bond

is perhaps one of the weakest bonds in the phenyl urea molecule (59) and is thus the most likely point of attack. Dapo and Mann (24) provide additional evidence for N-demethylation as the first step. They have shown that the first step in the electrochemical oxidation of tertiary amides is always cleavage of the N-methyl bond.

A justifiable criticism of these data is that the analytical method would not distinguish between the remaining parent molecule and its metabolites. The phenyl urea molecules do break down slowly so that the percentage of metabolites present at any one time would be small compared to the parent molecule. This assumes that the metabolites break down as rapidly as the parent.

To partially determine the extent of this problem several soil samples were analyzed for free aniline prior to basic hydrolysis. The soil was stirred for one hour with 1.0 N hydrochloric acid and then filtered. This solution was made basic to litmus and extracted with two 50 ml aliquots of benzene, followed by two 50 ml aliquots of 1.0 N hydrochloric acid. No aniline was detected after diazotization and coupling of aliquots of these samples. Therefore, it was concluded that when free aniline was formed it was probably broken down very rapidly.

## Triazine Herbicides

Three triazine herbicides were included in this experiment, they were: atrazine, ametryne and simazine. Atrazine has a chlorine atom in the two position while ametryne has a thiomethyl group. Simazine differs from atrazine in that it is 4-6-bis-ethyl-amino whereas atrazine is 4-ethylamino-6-isopropylamino. Because of these variations in molecular structure the data obtained have permitted the development of hypotheses concerning the initial point of attack and the relation between bond energy and the energy of activation.

Table IX shows the percentage of each of the herbicides remaining in the soil at each sampling period. Inspection of Table IX reveals the dependence of rate on temperature and an apparent difference in half lives. Figures 6, 7 and 8 graphically show that the rate of decomposition varied with temperature and that no lag period was apparent. This is not to imply that a lag period does not exist in the degradation of these molecules, only that these data do not show one. However, other data (6, 41) concerning the degradation of triazines have not indicated a lag period in the first step of degradation. Table X shows the rate of degradation for the triazine herbicides. These values were obtained from plots of the log of concentration vs. time. These data show that the rate of degradation of

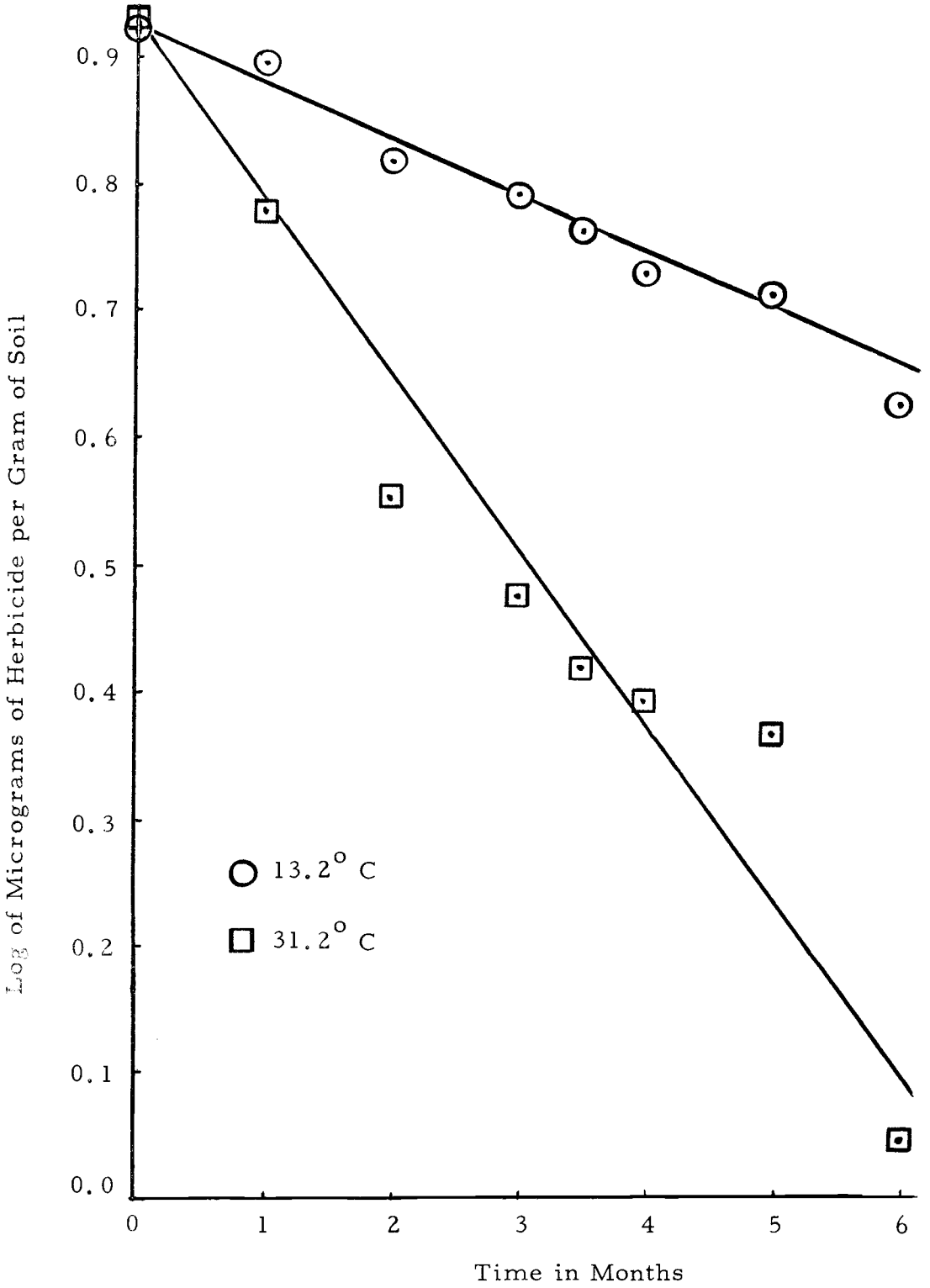


Figure 6. The rate of degradation of atrazine applied to the soil at eight ppm and stored at 13.2 and 31.2°C.

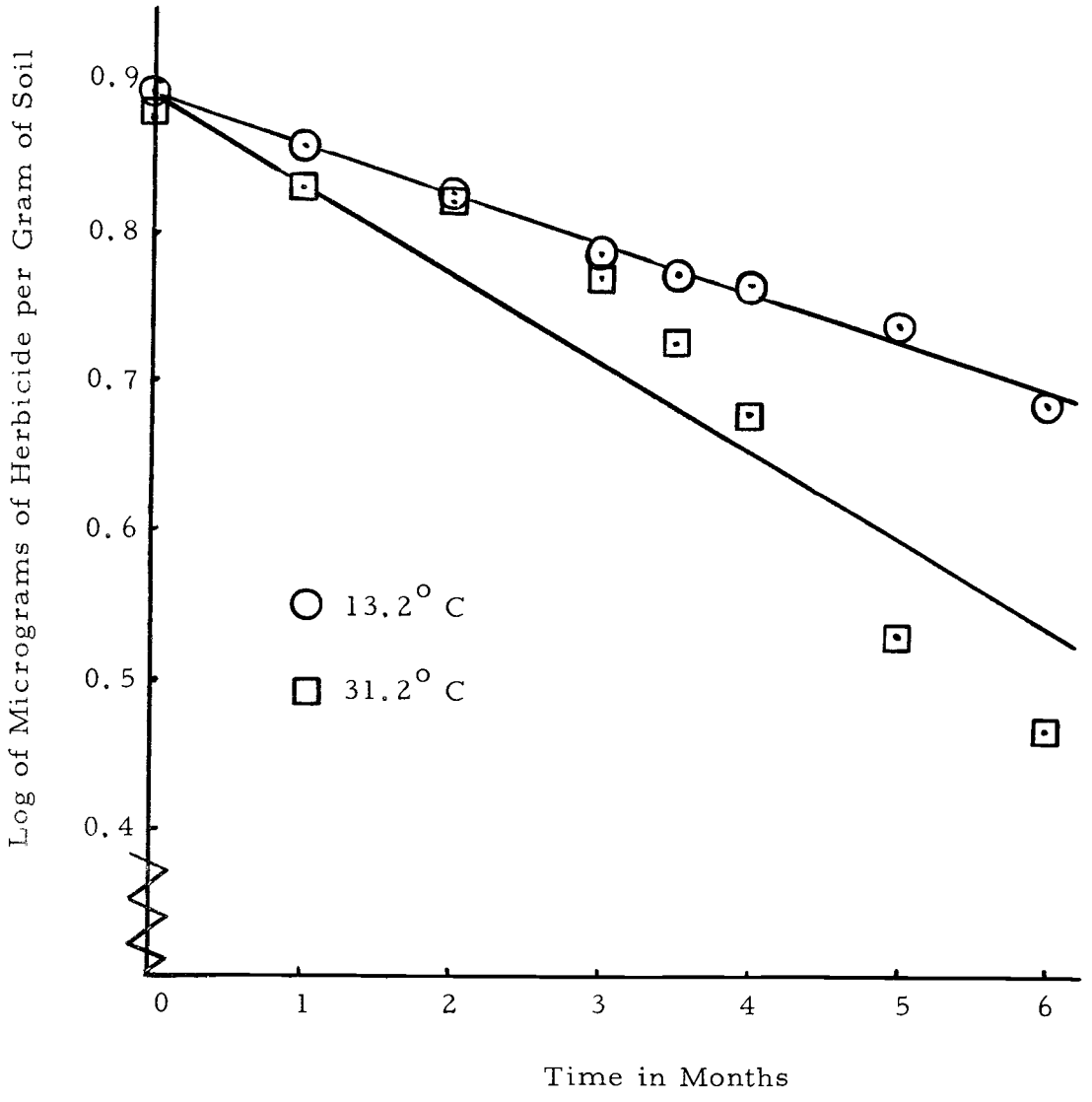


Figure 7. The rate of degradation of ametryne applied to soil at eight ppm and stored at 13.2 and 31.2° C.

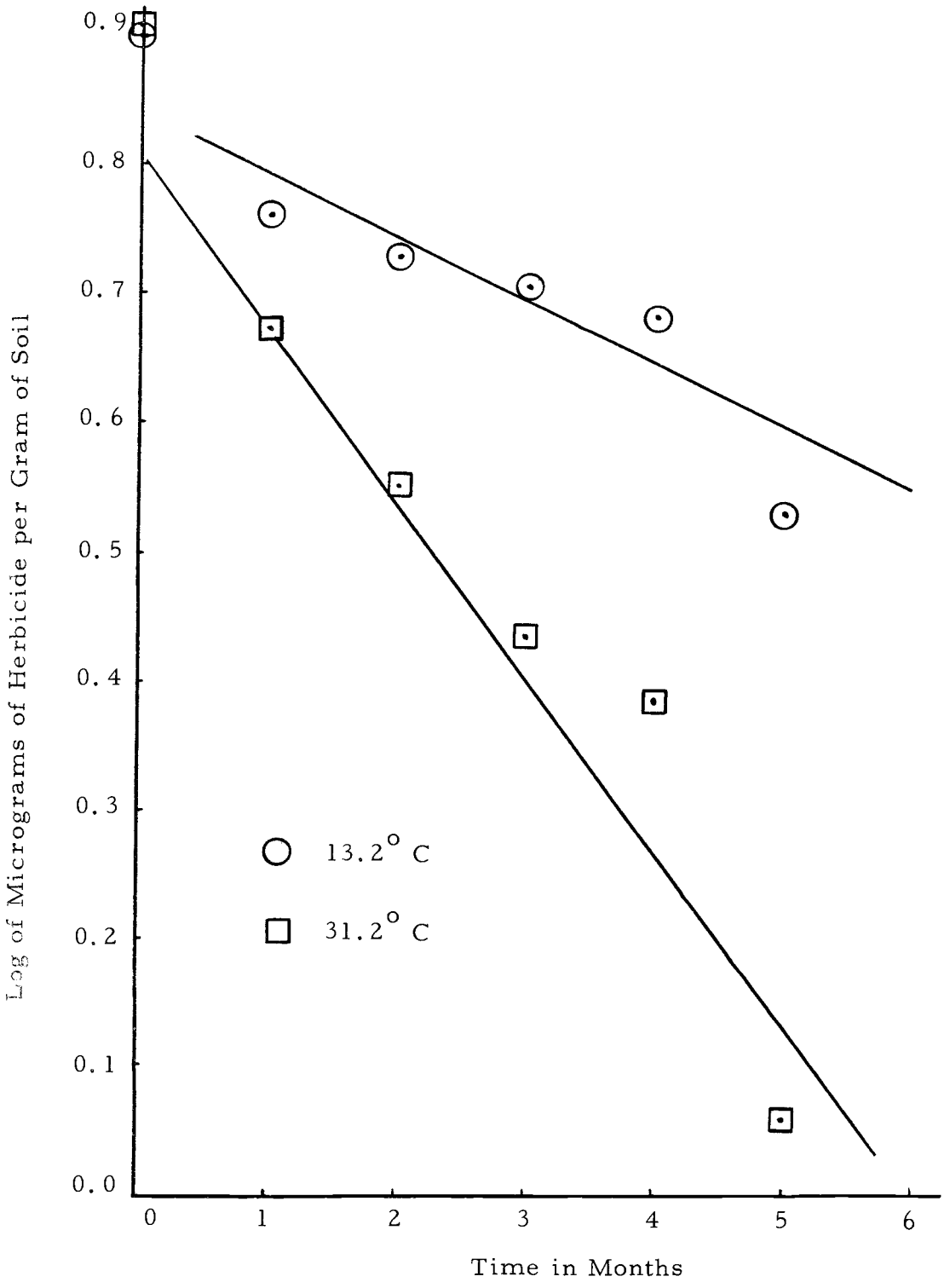


Figure 8. The rate of degradation of simazine applied to soil at eight ppm and stored at 13.2 and 31.2°C.

atrazine and simazine was quite similar and that they were more susceptible to the effects of temperature than was ametryne.

Table IX

The percentage of triazine herbicides remaining in the soil at different times and storage temperatures.

| Herbicide and initial concentration | Storage temperature °C | Percent of original concentration remaining, months after treatment | | | | | |
|---|---|---|---|---|---|---|---|
| | | 1 | 2 | 3 | 4 | 5 | 6 |
| atrazine 8 ppm | 13.2 | 94.9 | 79.4 | 74.9 | 65.0 | 62.7 | 51.2 |
| | 31.2 | 70.4 | 42.1 | 35.2 | 29.2 | 27.4 | 12.9 |
| ametryne 8 ppm | 13.2 | 96.7 | 89.7 | 81.6 | 78.1 | 73.6 | 64.9 |
| | 31.2 | 87.1 | 85.3 | 75.6 | 61.3 | 43.5 | 37.5 |
| simazine 8 ppm | 13.2 | 71.0 | 66.2 | 62.9 | 59.6 | 42.0 | |
| | 31.2 | 58.3 | 44.2 | 34.0 | 30.4 | 14.1 | |

Table X

The rate of degradation of three triazine herbicides in soil.

| Herbicide | Rate of degradation at storage temperature (°C) | |
|---|---|---|
| | 13.2 | 31.2 |
| atrazine | 0.1935 | 0.5978 |
| ametryne | 0.1370 | 0.2587 |
| simazine | 0.2130 | 0.5543 |

The activation energies for degradation of the triazine molecules are shown in Table XI. The activation energy values show that removal of a common group such as an N-alkyl is not the limiting step in degradation of these variously substituted triazines. Thus, it is postulated that attack probably occurs at the two position. This view is supported by the similarity of results with the two chloro substituted triazines. In view of the absence of a detectable lag period, it is also postulated that this is a chemical hydrolysis. Ercegovich (29) was among the first to suggest that the triazines are altered in the soil by chemical as well as microbial action. The non-biological conversion of simazine (41) and atrazine (6, 74) have been reported to occur in the soil. Others (27, 50) have reported specific microorganisms capable of degrading atrazine and simazine by N-dealkylation. However, these data indicate that a non-enzymatic conversion at the two position is probably the first step, while enzymatic N-dealkylation may occur subsequently.

The relative magnitude of activation energies also corresponds to the order of bond energies at the two position. The carbon-chlorine bond energy is 78.5 and the carbon-sulfur bond energy is 69.0 kilo-calories per mole (59). It is suggested that the activation energy values for other triazines will also correlate with the bond energy at the two position.

Table XI

Activation energies for degradation of three triazine herbicides in soil.

| Herbicide | Storage temperatures compared (°C) | Heat or energy of activation (Cals/mole) |
|---|---|---|
| atrazine | 13.2 : 31.2 | 10845 |
| ametryne | 13.2 : 31.2 | 6111 |
| simazine | 13.2 : 31.2 | 9195 |

## Uracil Herbicides

As shown by Figure 9 the degradation of terbacil and bromacil also followed a first order rate law. Neither compound appeared to have a lag phase and chemical (non-enzymatic) degradation, analogous to that of the triazines, is suspected as the first step in their breakdown. However, these data do not define the chemical mechanism of the process.

The rates of degradation showed the expected variation with temperature. Table XII shows the rates and the calculated activation energies. The bonds whose energies are shown are those suspected as the most likely point of attack. There are a finite number of reactions that can occur in the degradation of a given molecule. It is reasonable to assume that the reaction which cleaves the bond with the lowest energy, is the most likely to occur. It is not

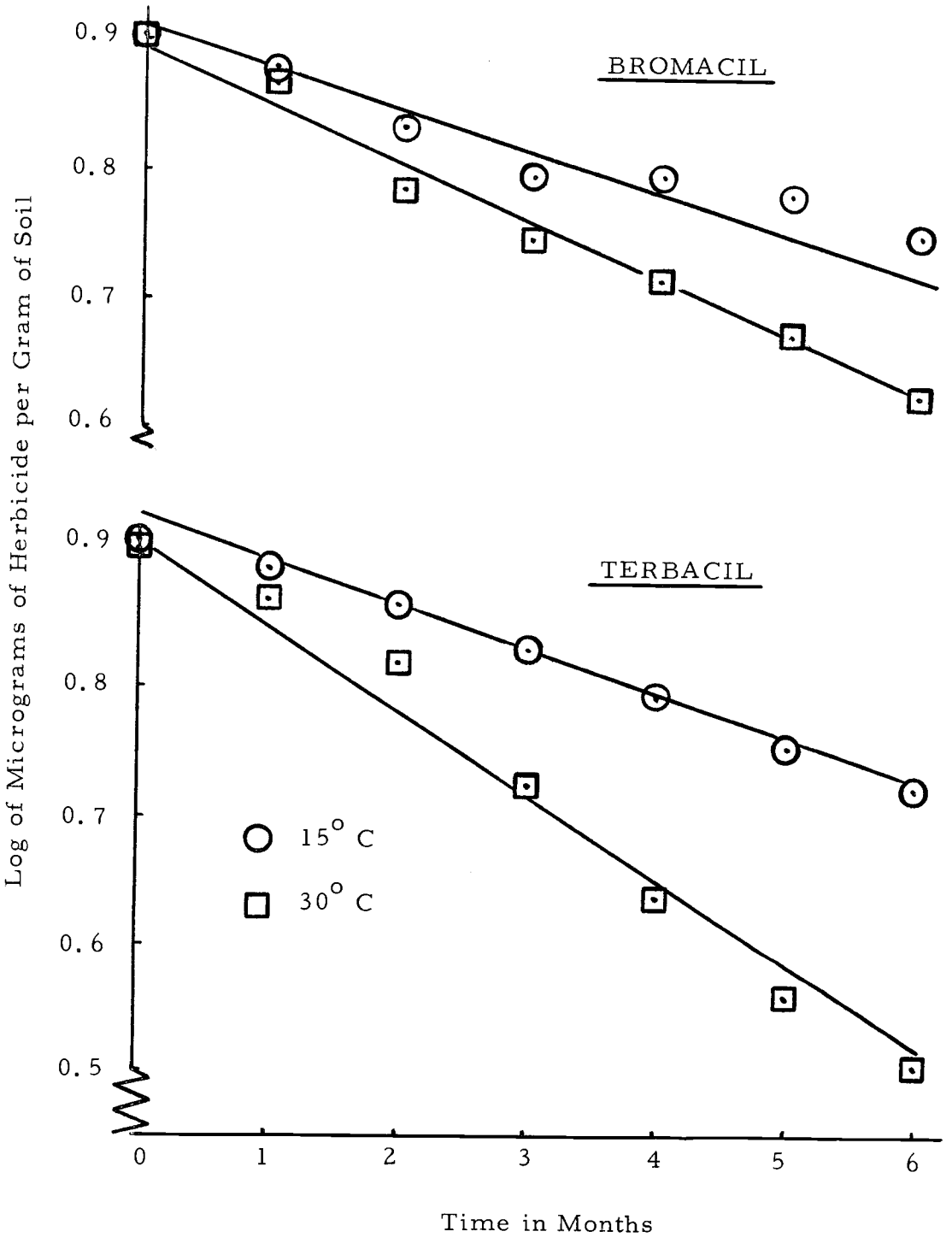


Figure 9. The rate of degradation of uracil herbicides applied to soil at eight ppm and stored at 13.2 and 31.2°C.

reasonable to assume a relation between bond energy and the energy of activation. The carbon-carbon bonds (see Appendix Table I for structural formulas) have a bond energy of 83.1 kilocalories per mole (59). These bonds are also more sterically hindered to chemical attack in both of the uracil molecules. Therefore, the most likely point of attack is the least hindered bond with the lowest energy. It is hypothesized that this initial attack is a hydrolysis leading to the hydroxyuracil, analogous to the hydroxytriazine. The activation energies fall in the same order as the bond energies, which lends support to the proposed site of attack. Because of the differences in the terbacil molecule, the activation energy for cleavage of the carbon-chlorine bond was not identical with that found for atrazine and simazine.

Table XII

Parameters of interest for the degradation of bromacil and terbacil in soil.

| Herbicide and Storage temperature | Rate of degradation | Activation energy (Cals/mole) | Bond | Bond energy (Kcals/mole) (59) |
|---|---|---|---|---|
| Bromacil | | | | |
| 13.2° C | 0.1413 | 3020 | C-Br | 65.9 |
| 31.2° C | 0.1935 | | | |
| Terbacil | | | | |
| 13.2° C | 0.3709 | 6113 | C-Cl | 78.5 |
| 31.2° C | 0.5869 | | | |

# VI. CONCLUSIONS

All studies of herbicide degradation in soil lead, sooner or later, to two parameters: chemical and environment. This study has been no exception and the results presented illustrate the fundamental and inseparable interaction of these two parameters. This study took place in a situation as close to reality as was possible under laboratory conditions. Emphasis was directed toward the usefulness of the techniques and conclusions with respect to herbicide persistence in soil. As pointed out in the Introduction, there is a vast complexity of possible interactions in the herbicide-soil milieu. However, from a practical point of view, most interactions can be ignored in an experiment of this type. This permits the use of very simple hypotheses on which to sketch the broad outlines of some of the parameters of degradation.

This study has established that nine herbicides in three different chemical families do follow a first order rate law in their breakdown. In the case of phenyl ureas, the first order rate began only after an initial lag period of undetermined length. This was indicative of a microbial breakdown which agrees with other published data. The triazines and uracils had no detectable lag period from which it may be suspected that they degraded non-enzymatically.

The energies of activation for the phenyl urea molecules fell

in the narrow range of 4-5 kilocalories per mole, which strongly suggested a common mode of degradation. Analysis of bond energies and published data supported the contention that this was an N-demethylation. The rate of N-demethylation was apparently attenuated by the substituents on the phenyl group. The energy of activation for the triazines and uracils did not fall in a narrow range which suggested breakage of a different bond in each case. Thus, it was concluded that a non-enzymatic conversion of the triazines to the hydroxy derivative was occurring at the point of variance, i.e., the two position. As evidenced by the data, the uracils also appeared to be attacked by chemical hydrolysis at the point of variance, i.e., the halogen substituent.

The parameters of rate and activation energy can be used as predictive tools for estimating the persistence of other compounds within a given class of herbicides. Both parameters are essential for this purpose. The rate of degradation is not specific for a given compound because it is attenuated by the characteristics of the soil environment. A soil with more organic matter than Chehalis loam will induce a slower rate of degradation. However, the relation of a given series of compounds will remain independent of soil characteristics. The activation energy required to initiate degradation of a herbicide will be quite constant and independent of the specific environment and dependent only on the nature of the bond involved.

Field studies[4] have shown that atrazine is phytotoxic longer than ametryne under most conditions in the United States. The data presented herein suggest that the converse is true. Therefore, it is porposed that there is probably not a 1:1 ratio between phytotoxicity and the amount of chemical detected by chemical analysis. If these experiments were repeated using bioassay techniques rather than the techniques of analytical chemistry different results would probably be obtained. Because of the stronger sorption of the ametryne molecule it may persist longer due to its unavailability as opposed to the less tightly bound atrazine. The preceding is not intended to imply that an answer obtained by analytical means is any more correct than one from a bioassay. The possibility of a difference should be recognized and results interpreted accordingly.

Although these data may be used as predictive parameters, this use is limited to a given class of herbicides with no obvious correlation between classes. This approach also provides another way to formulate hypotheses concerning the initial point of attack and the mechanism of degradation. The soil and its adsorptive capacity will influence the rate of degradation but the energy required and the point of attack will be constant. Therefore, it should be recognized that this method alone is not sufficient to precisely define the point of

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[4] Furtick, W. R. Personal communication. Corvallis, Oregon State University. 1967.

attack or mechanism of degradation and should be coupled with other complementary studies.

For the interest of those who may follow this line of research, the following suggestions are offered. The modification in the method of sampling proposed in Chapter IV should definitely be incorporated in future experiments. In addition, it is suggested that shorter sampling intervals be used in the first 1 or 2 months to define the length of biological lag period, if present. Finally, the combination of bioassay and chemical analysis would provide a very interesting approach to the problem.

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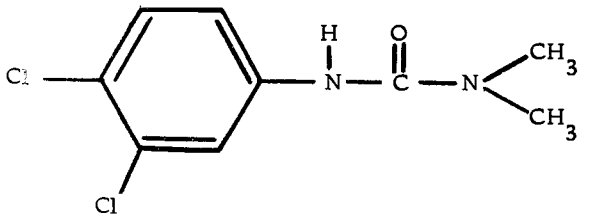
# APPENDIX

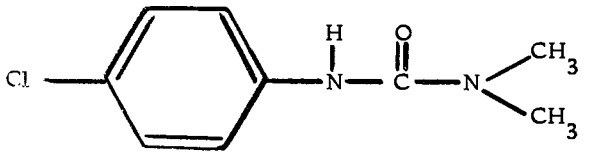
Appendix Table I. Structural formulas of experimental herbicides.

I. Phenyl Urea Herbicides

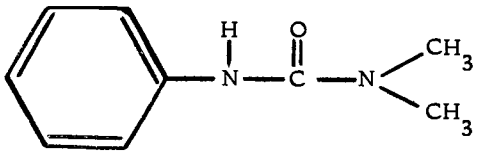
A. Diuron



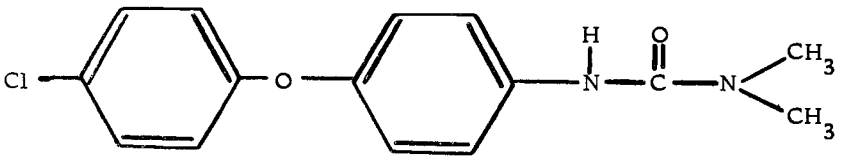
B. Monuron



C. Fenuron

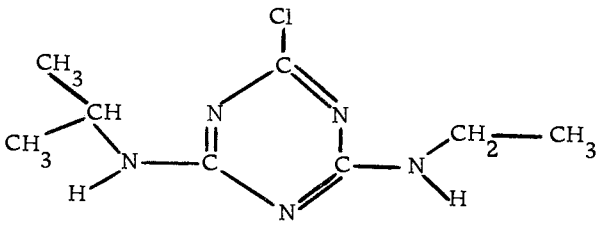


D. Chloroxuron

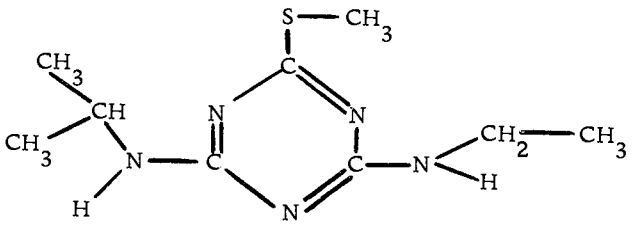


II. Triazine Herbicides

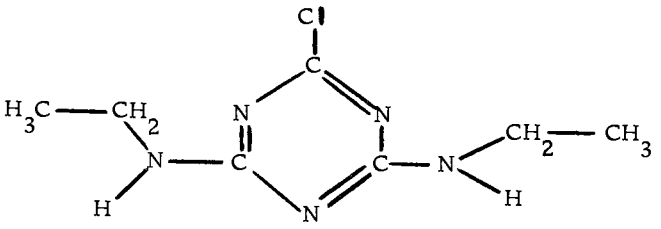
A. Atrazine



B. Ametryne

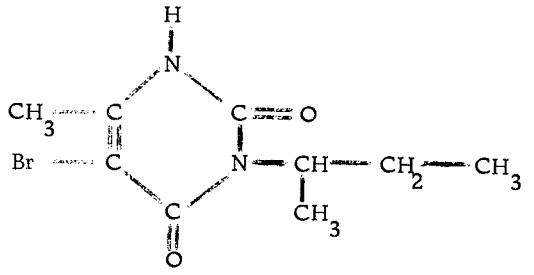


C. Simazine

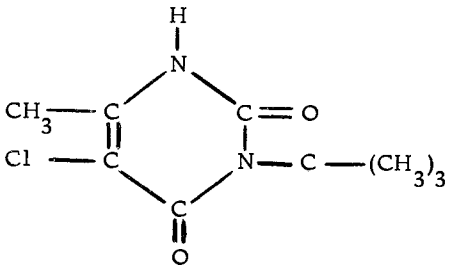


III. Uracil Herbicides

A. Bromacil



B. Terbacil



Appendix Table II.

The amount of herbicide remaining, months after storage (in ppm).

| Herbicide | Storage temperature °C | Initial concentration time zero | Months from time zero (ppm) | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | 1 | 2 | 3 | 3-1/2 | 4 | 5 | 6 |
| diuron | 6.5 | 2.25 | 1.90 | 1.57 | 1.63 | | 1.57 | 1.49 | 1.61 |
| diuron | 13.2 | 2.23 | 1.64 | 1.68 | 1.53 | | 1.33 | 1.20 | 1.13 |
| diuron | 31.2 | 2.25 | 1.70 | 1.58 | 1.46 | | 1.22 | 1.14 | 1.13 |
| diuron | 6.5 | 7.70 | 7.97 | 7.22 | 6.79 | | 6.26 | 6.35 | 5.58 |
| diuron | 13.2 | 7.95 | 8.11 | 7.52 | 6.45 | | 6.08 | 5.38 | 4.75 |
| diuron | 31.2 | 8.08 | 8.18 | 7.12 | 5.68 | | 5.63 | 5.34 | 3.49 |
| monuron | 13.2 | 8.12 | 7.91 | 7.40 | 6.44 | | 5.49 | 5.09 | 4.67 |
| monuron | 31.2 | 7.90 | 7.85 | 6.49 | 5.13 | | 4.30 | 3.74 | 3.25 |
| fenuron | 13.2 | 8.49 | 7.74 | 5.59 | 3.73 | 2.83 | 2.31 | 1.79 | 1.70 |
| fenuron | 31.2 | 8.25 | 7.59 | 5.13 | 2.32 | 1.36 | 1.34 | 1.03 | 0.28 |
| chloroxuron | 13.2 | 7.85 | 6.13 | 4.25 | 3.66 | | 2.65 | 1.62 | 1.23 |
| chloroxuron | 31.2 | 7.80 | 3.54 | 3.00 | 2.01 | | 1.08 | 0.61 | 0.38 |
| atrazine | 13.2 | 8.26 | 7.84 | 6.56 | 6.19 | 5.81 | 5.37 | 5.18 | 4.23 |
| atrazine | 31.2 | 8.50 | 5.98 | 3.58 | 2.99 | 2.63 | 2.48 | 2.33 | 1.10 |
| ametryne | 13.2 | 7.50 | 7.25 | 6.73 | 6.12 | 5.95 | 5.86 | 5.52 | 4.87 |
| ametryne | 31.2 | 7.82 | 6.81 | 6.67 | 5.91 | 5.33 | 4.79 | 3.40 | 2.93 |
| simazine | 13.2 | 8.11 | 5.76 | 5.37 | 5.10 | | 4.84 | 3.41 | |
| simazine | 31.2 | 8.25 | 4.70 | 3.56 | 2.74 | | 2.45 | 1.14 | |
| bromacil | 13.2 | 8.02 | 7.50 | 6.84 | 6.28 | | 6.28 | 6.06 | 5.04 |
| bromacil | 31.2 | 8.00 | 7.44 | 6.10 | 5.62 | | 5.20 | 4.72 | 4.25 |
| terbacil | 13.2 | 8.07 | 7.81 | 7.28 | 6.82 | | 6.20 | 5.66 | 5.26 |
| terbacil | 31.2 | 8.18 | 7.38 | 6.59 | 5.29 | | 4.34 | 3.62 | 3.18 |