AN ABSTRACT OF THE THESIS OF

JONATHON DENNIS SCHREIBER for the Doctor of Philosophy
(Name of student) (Degree)

in SOILS presented on August 2, 1971
(Major) (Date)

Title: MOISTURE INFLUENCES ON BROMACIL DISTRIBUTION IN THE SOIL AND RESULTANT UPTAKE AND PHYTOTOXICITY

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Abstract approved: ______________________
Dr. V. V. Volk

The objectives of this research were to determine the distribution and phytotoxicity of 5-bromo-3-sec-butyl-6-methyluracil (bromacil) in the soil aqueous phase as influenced by soil texture and soil moisture. Three Oregon soils and a quartz sand were selected for study providing a range in soil texture and water holding properties.

A volumetric pressure membrane apparatus (VPMA) was used to extract soil water over a 15 bar moisture tension range from 375 g of soil treated with 7,100 μg of ^{14}C bromacil. Extracts were collected at pressure increments and analyzed for bromacil content. For all three soils and the quartz sand the concentration of bromacil in the extracts increased as the soil moisture tension increased from a 0.10 to 0.33 bar tension. From the 0.33 bar tension to a 3

bar tension the bromacil was relatively constant at 11, 15, and 26 μg/ml of extract for the Coker clay, Woodburn silt loam, and Ephrata loam soils. At higher tensions the bromacil concentration in the soil water extracts decreased due to a possible increase in bromacil adsorption at the drier soil conditions.

Differences in bromacil concentration of the extracts among the soils appears to be a function of the adsorptive capacity of each soil for bromacil. While bromacil phytotoxic differences can be expected among soils of different texture, the relative concentration of bromacil in the soil water over a moisture tension range from 0.33 to 3 bars would indicate that in this moisture range for each soil, bromacil phytotoxicity should be largely controlled by the soil water influence on the plant system. Total quantities of bromacil removed from the soils over the 15 bar tension range were 82, 45 (est.), 26, and 21% of that initially added to the quartz sand, Ephrata loam, Woodburn silt loam, and Coker clay respectively. The greater majority of bromacil extracted was removed at tensions less than 0.60 bars, indicating that movement of bromacil could occur at high soil moisture levels.

Wheat plant bioassay studies with the Ephrata loam, Coker clay, and Woodburn silt loam at four soil moisture levels indicated that both soil texture and soil moisture had a pronounced effect on the uptake and phytotoxicity of ^{14}C bromacil. The maximum concentration of bromacil in the plant tissue grown in the Ephrata loam, Woodburn silt loam, and Coker clay soils was 523, 137, and 115 μg bromacil per gram plant tissue respectively at the 4.0 ppm bromacil application rate. The variations in the uptake of bromacil by the plants from the three soils reflect the differences in the concentration of bromacil in the soil solution of the three soils as observed from the VPMA studies.

As soil moisture decreased from 42 to 12% for the Ephrata loam soil, the accumulation of bromacil increased at the 4.0 ppm bromacil rate form 36 to 49 μg. Over the same moisture range the plant growth increased from 25 to 49% of the check plant weight. Less dramatic, but similar observations were also made for the plants grown in the Coker clay and Woodburn silt loam soils.

The initially faster rates of transpiration and photosynthesis at higher soil moisture values are hypothesized explanations for the increased toxicity of bromacil at higher soil moisture values. At lower soil moisture values, the bromacil was not as toxic to plant growth and the plants continued to grow and assimilate bromacil, which by the end of the two-week growth period had resulted in higher concentrations of bromacil in the plant tissue.

The effects of soil moisture tension on the plant system in relation to the uptake of ^{14}C bromacil was studied by placing wheat plants (three weeks old) grown in Woodburn silt loam into osmotic solutions that regulated the soil moisture tension across a cellulose membrane at 0.35 and 2.5 bars. Measurements of bromacil uptake and transpiration were made as a function of time.

At both soil moisture tensions and bromacil application rates (2.0 and 4.5 μg/ml in 5 l chambers) the uptake of bromacil into the root and foliar portions increased in a linear fashion with the increase in time over a ten-day experimental period.

Soil moisture tension greatly influenced the uptake of bromacil into both the foliar and root portions of wheat. At the low and high bromacil applications 70 and 42% more bromacil respectively was taken up at the 0.35 bar tension compared to the 2.5 bar tension. At the same bromacil application rates, 25 and 69% more bromacil was taken up by the roots at the low moisture value.

Uptake of bromacil by the plants at both soil moisture tensions and bromacil application rates increased in direct proportion to the increase in water transpired by the plants. Calculation of bromacil concentration in the transpirational stream at both soil moisture tensions showed a constant bromacil concentration after two days at the 2.5 bar tension, whereas the bromacil concentration in the transpirational stream increased at the 0.35 bar tension over a ten-day period. As the transpiration rates of the plants were essentially the same after 40 hours, root permeability was cited as a major factor in limiting the uptake of bromacil into the plant system.

At the low bromacil application rate, a greater proportion of bromacil was translocated from the root to the foliar portions at the 0.35 bar tension compared to the 2.5 bar tension.

The increased toxicity of bromacil at the higher soil moisture values can be explained in part by the more rapid uptake and translocation of bromacil from the root to the foliar portions of the plant.

Moisture Influences on Bromacil Distribution in the Soil and Resultant Uptake and Phytotoxicity

by

Jonathon Dennis Schreiber

A THESIS

submitted to

Oregon State University

in partial fulfillment of
the requirements for the
degree of

Doctor of Philosophy

June 1972

APPROVED:

Redacted for Privacy

Associate Professor of Soils
in charge of major

Redacted for Privacy

Head of Department of Soils

Redacted for Privacy

Dean of Graduate School

Date thesis is presented August 2, 1971

Typed by Opal Grossnicklaus for Jonathon Dennis Schreiber

Dedicated to

Sharon

FOR: her patience with me and an understanding of the many long hours spent away from home during the graduate program

her long hours spent after the completion of her work day on the analysis of data, typing, and editing of the thesis

finding me and making my life more meaningful

ACKNOWLEDGEMENTS

A very special and warm thank you to Dr. V. V. Volk, my major professor during these past five years, for his careful and critical guidance of my graduate program, the concern expressed to me pertaining to my development as an individual as well as a soil scientist, and finally for his many hours devoted to the editing of this thesis and for the freedom to pursue a graduate program of my own design.

To Dr. L. Boersma for the many helpful suggestions and use of soil physics' equipment during the osmotic chamber studies. Also to Dr. C. H. Ullery for his editing of this thesis

To the remaining members of my committee: Drs. A. P. Appleby, J. W. Nibler, and R. M. Highsmith, Jr. for their helpful suggestions on the graduate program and editing of this thesis

To Drs. M. D. Dawson and D. O. Chilcote for use of growth chamber space during certain phases of the experimental work

To Drs. H. D. Skipper and F. O. Colbert for sharing with me their philosophies of research, critical and helpful suggestions, and their problems

To my fellow graduate students Wanee Akratanakul, Mohammad Fazilat, and Ali Sepaskhah for their aid so freely given during the osmotic chamber studies

To Dr. J. A. Gardiner of E. I. Du Pont de Nemours and Company for providing ^{14}C labeled bromacil

To E. C. Y. Kuo, J. D. Gaynor, and G. B. Tucker, members of the "soil chemistry group," for their interest expressed in my research

To the United States Department of Health, Education, and Welfare for financial support through the Pesticide Toxicology Training Grant

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INTRODUCTION

Soil applied herbicides must undergo various equilibrations and cross many barriers before their toxicity is ultimately expressed at a target site within the plant system. The entire quantity of a given amount of herbicide applied to a soil system is not adsorbed, but rather a certain quantity of the herbicide will remain in the soil solution. The amount of herbicide which will remain in the soil solution will depend upon the chemical and physical properties of both the soil and the herbicide. Depending upon the adsorption capacity of a soil for a specific herbicide, fluctuations in soil moisture can influence the concentration of herbicide in the soil solution. If the adsorption capacity of the soil for the herbicide is low, the concentration of herbicide in the soil solution may increase as soil moisture decreases, whereas if the adsorption capacity of the soil is high, any additional increase in herbicide concentration at lower soil moisture values may be adsorbed by the soil system.

An equilibration between the herbicide in the soil solution and that adsorbed or concentrated near root surfaces can also be expected. Soil moisture may influence this equilibrium by controlling not only the concentration of herbicide in the soil solution, but also by controlling the rate of water movement through the soil. These two factors in turn influence the mass flow of herbicide in the soil water as well as molecular diffusion, and hence limit the concentration of herbicide at the root surface.

If a soil applied herbicide must reach a reaction site inside the foliar portions of a plant, soil moisture effects on the plant system alone must also be considered. Soil moisture may influence plant physiological functions such as root permeability, transpiration, and photosynthesis. As soil moisture decreases, root permeability is also thought to decrease, which could limit the quantity of herbicide that enters the plant system. Decreased rates of transpiration at lower soil moisture values could influence not only the uptake of herbicide as a function of time, but also the subsequent translocation of herbicide from the root to the reaction site in the foliar portions. Thus, when considering the influence of soil moisture on the effectiveness of a soil applied herbicide, soil moisture effects on the plant system as well as the soil system must be considered.

DISTRIBUTION OF BROMACIL IN THE SOIL AQUEOUS PHASE

Literature Review

Introduction

The adsorption of herbicides takes place on the chemically active fractions of the soil such as clay and organic matter; thus, soil texture might be expected to influence the distribution of bromacil in the soil aqueous phase. At a given soil moisture tension, soils of different texture contain different amounts of water that might also influence the equilibrium of bromacil in the soil solution with that adsorbed on the clay and organic matter soil fractions.

Soil moisture

The adsorption of herbicides onto different soils is often characterized by a K value, which represents the concentration of herbicides in the adsorbed phase divided by the concentration of herbicide in the non-adsorbed phase (Green and Obien, 1969). A soil with a large K value (K=5) will not undergo changes in herbicide concentration as the soil moisture fluctuates. Variations of soil moisture in a soil with a small K value (K=0.1) may induce changes in the concentration of herbicide. Utilizing a porous ceramic cup filled with 250 g of either the A, B, or C horizon of a Kapaa soil, the distribution of atrazine in the soil solution was determined at soil moisture levels of 45, 50, 55, and 60% (Green and Obien, 1969). Changes in the soil moisture content of the A, B, and C horizons (horizon K values of 2.72, 1.06, and 0.35 respectively) caused only the C horizon to show a significant increase in atrazine concentration (15 μg/ml to 18 μg/ml) as the soil moisture decreased from 60 to 45% by weight. Over the same moisture range the atrazine concentration increased by only 1 μg/ml for the B horizon while essentially no increase in atrazine concentration was observed for the A horizon (Green and Obien, 1969). Data generated by a predictive equation based upon the appropriate K value and effective soil water content agreed with the experimental values of atrazine concentration in the soil water as a function of soil moisture for all three soil horizons of the Kapaa soil. The effective soil water content used in the predictive equation was defined as the soil water present in a soil at tensions less than 15 bars.

Standard soil slurry techniques involving vigorous shaking produced K values that were 20% higher than the K values determined by the use of soil aggregates that were not dispersed (Green and Obien, 1969). Thus, adsoprtion of herbicides can be over-estimated by soil slurry techniques. Adsorption of linuron and atrazine on a Begbroke soil was studied by Grover and Hance (1970) at soil to water ratios of 1:10, 4:1, and 1:1. As the soil to water ratio decreased from 4:1 to 1:10 there was a five fold increase in the adsorption of linuron and a three fold increase in the adsorption of atrazine. The dispersion of soil aggregates was greater at the soil to water ratio of 1:10 than at the 4:1 ratio. The increased surface area and exposure of additional adsorption sites upon soil dispersion were cited for the increased adsorption at the lower soil to water ratios.

A pressure membrane extractor was used by Scott and Lutz (1971) to study the release of several herbicides from kaolinite as a function of water content over a 15 bar moisture tension range. Six herbicides were studied and grouped into three classes based on the herbicide concentration in the extracts as a function of pressure. The three classes were 1) herbicide concentration of solution extracted higher than initial concentration, indicating negative adsorption (2, 4-D and fluometuron), 2) herbicide concentration of initial extracts slightly less than that added, with a further decrease as pressure increased (atrazine, chlorpropham, and diuron), and 3) herbicide concentration in the extracts increased as pressure increased (simazine) but at all times less than the initial concentration. The amount of chemical removed from the kaolinite clay by the pressure membrane extractor expressed as a percentage of the initial amount added ranged from 89.3% for 2, 4-D to 28.5% for diuron over the 15 bar pressure range. At a 1.0 ppm application rate, the percentage of 2, 4-D and diuron removed at the first pressure increment (0-0.3 bars) was 54.5 and 24.4% respectively. The remaining portions of each herbicide were removed in additional pressure increments from 0.3 to 15 bars.

Analysis of the soil water from a pressure membrane apparatus indicated that minor deviations in the concentration of some ions in the initial fractions of soil water collected could result from membrane filtration and the presence of insoluble carbonates (Reitemeier and Richards, 1944). These effects were minimized when the extracts were collected and analyzed in smaller fractions rather than as large bulk samples. Thus, caution should be exercised when interpreting herbicide concentration data of the first few fractions of soil water extracts from a volumetric membrane apparatus.

Microbial decomposition

Exposure of bromacil to high temperatures and sunlight indicate that soil loss of bromacil due to volatilization and photodecomposition are minimal (Bingeman et al. (1962). Bromacil degradation studies by Gardiner et al. (1967) under field conditions established a half-life of five to six months for bromacil. Approximately 24% of the initial bromacil applied to a Butlertown silt loam remained after a one-year exposure period in the field. Of the

^{14}C labeled compounds (bromacil, 5-bromo-3-sec-butyl-6-hydroxylmethyluracil, 5-bromo-3-(2-hydroxy-1-methylpropyl)-6-methyluracil, and an unidentified compound) which were extracted from the soil, 90% was the intact bromacil molecule.

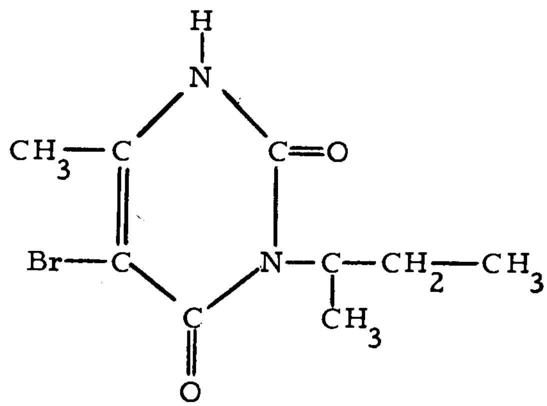
Soil diphtheroids and Pseudomonas Sp. of bacteria are both known to degrade bromacil (Reid, 1963). One mode of bromacil degradation hypothesized was hydroxylation of the side chain alkyl groups, followed by ring cleavage, and then metabolism to CO_2, ammonia, and hydrobromic acid (Gardiner et al., 1967).

Objectives of this study were to determine the distribution and removal of bromacil in the soil water as a function of soil texture and soil water. In order to fully discern the role of soil moisture on bromacil phytotoxicity it was necessary to know the distribution and concentration of bromacil in the soil solution as a function of soil moisture.

Materials and Methods

Chemical and physical properties of bromacil

The structural formula and various physical and chemical characteristics of bromacil are indicated below.



Formula	$C_9H_{13}BrN_2O_2$
Molecular weight	261.1
Solubility	0.815 g/l H_2O
Melting point	158-159° C
Mode of action	Inhibitor of photosynthesis

The 2-^{14}C-labeled bromacil was supplied in crystalline form by E. I. Du Pont de Nemours and Company.

Selection of soils

Three Oregon soils and a quartz sand were selected to give a range of physical and chemical properties (Table 1). The soil textures ranged from a clay (47.8% clay) to a loam (11.8% clay). For the same three soils organic matter varied from 4.7 to 0.6%. Another important consideration was the relative amounts of water present in the three soils at selected moisture tensions. At saturation the percent moisture in the Woodburn silt loam, Ephrata loam,

Table 1. Chemical and physical properties of several Oregon soils

Soil series	O. M.	Soil separates: sand	silt	clay	Moisture at field capacity	CEC	pH
	%	%	%	%	%	meq/100 g	
Woodburn sil	3.1	11.0	72.0	17.0	28.5	18.9	6.6
Ephrata 1	0.6	41.6	46.6	11.8	18.2	10.3	7.8
Coker c	4.7	n.d.	n.d.	63.7	47.8	45.2	6.9
quartz	n.d.	n.d.	n.d.	n.d.	2.3	n.d.	n.d.

n.d. not determined

Coker clay, and quartz sand was 48, 42, 76, and 33% respectively. The Coker clay soil contained predominantly montmorillonite clay whereas the Woodburn silt loam contained vermiculite, mica, and some beidellite and kaolinite. Clay minerals present in the Ephrata loam were montmorillonite, vermiculite, mica, and possibly kaolinite.

Distribution of bromacil in the soil aqueous phase

A volumetric pressure membrane apparatus (VPMA) consisting of a base containing a cellulose membrane and an enclosing cylinder was used to extract different soils treated with ^{14}C labeled bromacil. A total of 375 g (oven dry weight) of each soil was weighed into approximately six equal portions. The first portion of soil was added to the membrane surface of the VPMA and uniformly distributed with a small brush. An increment of aqueous ^{14}C labeled bromacil solution was added uniformly across the soil surface to bring the soil moisture level to saturation. Each increment of soil and ^{14}C labeled bromacil solution was added in a similar manner to bring the total 375 g to saturation. A total of 7,100 μg bromacil was added to each 375 g of soil.

The VPMA was sealed and the soil-bromacil system allowed to equilibrate for 24 hours. Following equilibration, pressure, using nitrogen gas, was applied to the VPMA in selected increments over a pressure range of 0.10 bars (1.5 PSI) to 15 bars (225.0 PSI). The effluent was collected volumetrically into vials from ports at the bottom of the VPMA. Care was taken to avoid evaporation of sample from the glass vials. The volume of effluent and collection time were dependent upon both the soil in the VPMA and the pressure increment at which the effluent was being collected. Equilibrium at each pressure increment was considered complete when the flow rate from the VPMA was less than 0.10 ml per hour. Each soil, except the quartz sand, was run in duplicate over the 15 bar tension range.

From each vial of effluent collected over the 15 bar tension range duplicate 1 ml samples were pipetted into a scintillation vial along with 10 ml of a triton X-100 fluor and counted for ^{14}C activity by liquid scintillation methods. Bromacil concentration was calculated and expressed μg/ml of effluent. Since the total volume of effluent was known the absolute quantity of bromacil removed in each fraction and for each pressure increment could also be calculated.

Results and Discussion

Moisture tension curves

Soil moisture content decreased with an increase in soil moisture tension for a clay, silt loam, loam, and a quartz sand soil (Figure 1). As the percentage of clay increased for the four soils the quantity of water retained at a given moisture tension value also increased. At the end of the 15 bar equilibration the soil moisture

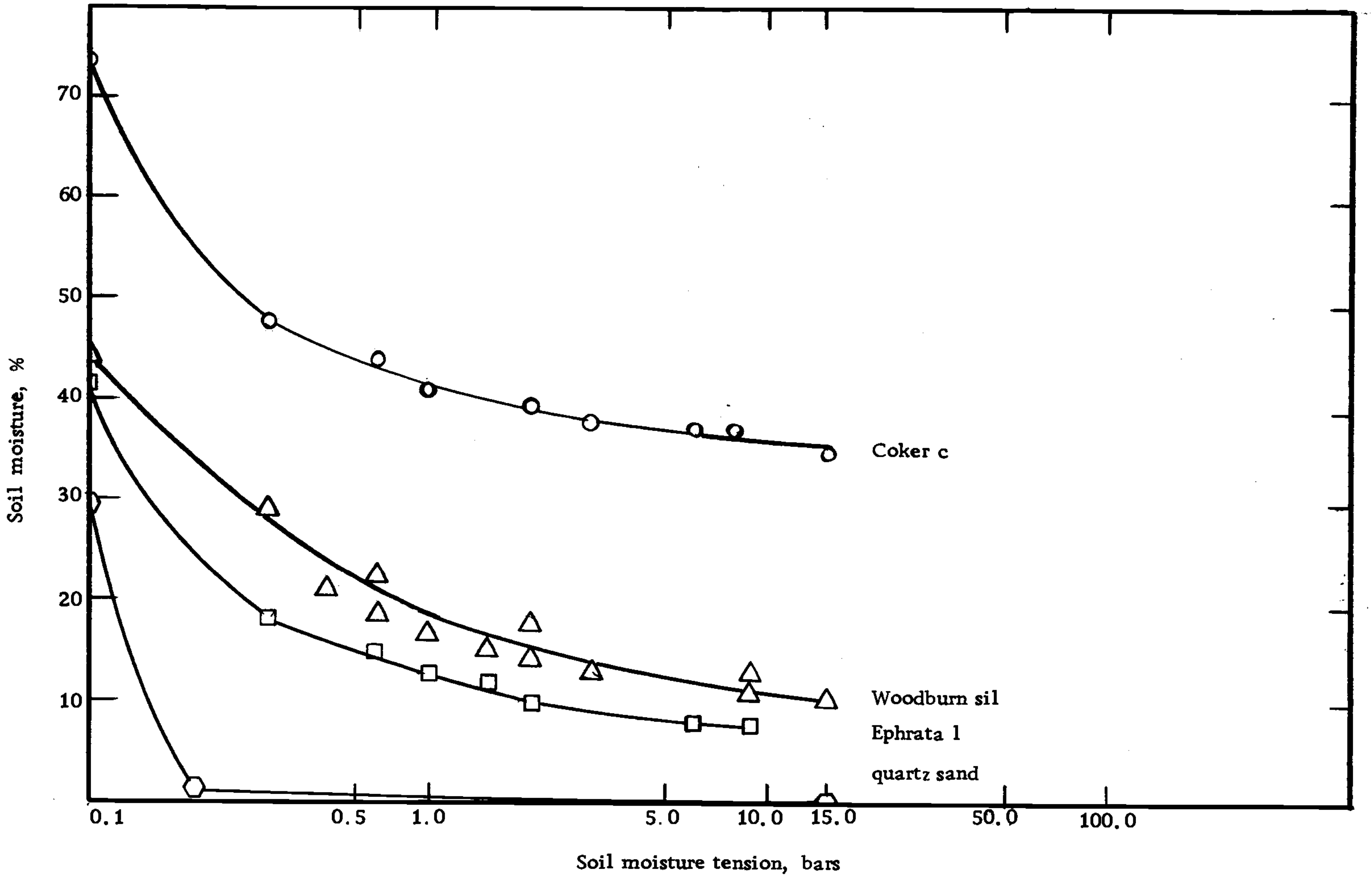


Figure 1. Soil moisture tension curves for three Oregon soils and a quartz sand

content for the Coker clay, Woodburn silt loam, Ephrata loam, and quartz sand was 35.5, 10.2, 6.5 and 0% respectively.

Bromacil distribution in soil water extracts

The differences in bromacil concentration of the extracts among the three soils and the quartz sand reflected the adsorptive capacity for bromacil by the three soils (Figure 2). Previous adsorption isotherm studies indicated that the three soils and the quartz sand placed in order of increasing bromacil adsorption were quartz sand < Ephrata loam < Woodburn silt loam < Coker clay, the reverse order in terms of bromacil concentration in the soil water extracts (Figure 2). In view of the large differences in percentages of clay between the Woodburn silt loam (17.0%) and Coker clay (63.7%) soils, the small differences in the concentration of bromacil in the soil solution for these two soils could be due to the adsorption of bromacil by the organic matter fraction of these soils.

Explanation of the differences in bromacil concentration of the soil solutions due to pH effects on adsorption would be doubtful as, in general, the pH of the Coker clay, Ephrata loam, and Woodburn silt loam were about the same (Table 1). The Ephrata loam and the Woodburn slit loam contained approximately the same percentage of clay, 11.8% for Ephrata loam and 17.0% for Woodburn silt loam, and also the same types of clay minerals such as montmorillonite, vermiculite, mica, and kaolinite. However, the Woodburn silt loam contained five times the amount of organic matter of the Ephrata loam soil.

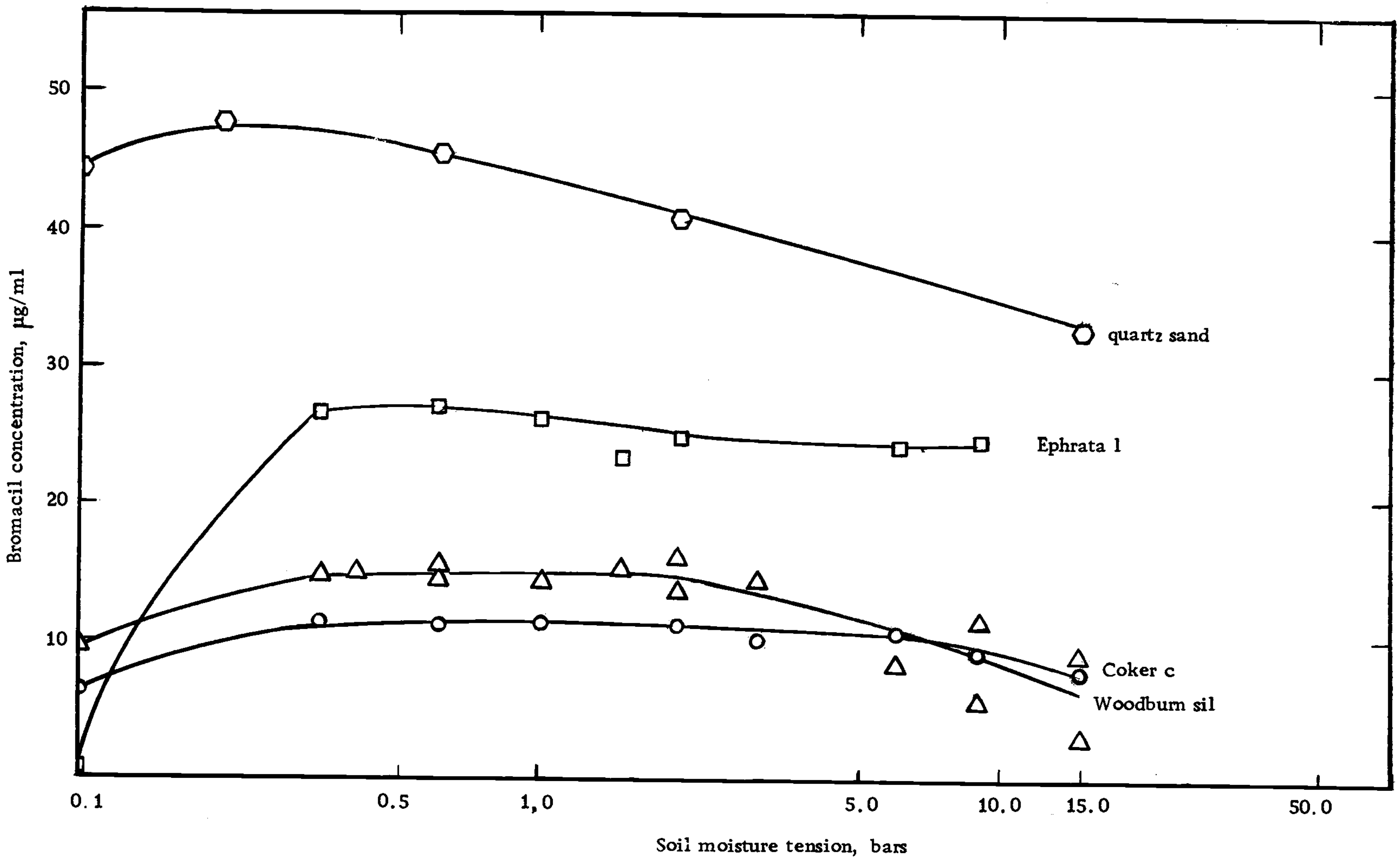


Figure 2. Bromacil concentration in soil water extracts at selected soil moisture tension values

In view of the bromacil concentration differences between the Ephrata loam and Woodburn silt loam soil water extracts, organic matter might be suspected as the predominate adsorption site for bromacil. As the Coker clay soil contained primarily montmorillonite clay as compared to a mixture of clay minerals for the remaining soils, higher adsorption could be expected for the Coker clay if cation exchange properties are important for the adsorption of bromacil. Results seem to indicate however, that organic matter plays a more important role in bromacil adsorption than does clay.

The bromacil concentration in the extracts collected from the VPMA increased for all three soils and the quartz sand from the 0.10 bar tension to the 0.33 bar tension or field capacity (Figure 2). From field capacity to the 3 bar tension the bromacil concentration was relatively constant at 11, 15, and 26 μg/ml of extract from the Coker clay, Woodburn silt loam, and Ephrata loam soils. Beyond the 3 bar tension value the bromacil concentration in the extract from the Coker clay remained constant until the 6 bar tension value where the bromacil concentration decreased from 10.5 μg/ml to 8.0 μg/ml at the end of the 15 bar tension equilibration. The concentration of bromacil in the extract from the Woodburn silt loam decreased from 14 μg/ml at the 3 bar tension to 6 μg/ml at the 15 bar tension value. At the tension range from 3 to 9 bars, the highest pressure increment achieved for the Ephrata loam soil, the bromacil concentration remained constant at 15 μg/ml. The extract concentration of bromacil from the quartz sand decreased continually from 47.7 μg/ml at the 0.20 bar tension to 33 μg/ml at the 15 bar tension.

The initial increase in bromacil concentration could be due to insufficient time allowed for the bromacil to completely diffuse throughout the soil. An initial soil increment of approximately 60 g was placed on the membrane surface of the VPMA prior to the first addition of bromacil solution. The initial soil increment was approximately 0.9 cm thick and contained approximately 2% moisture (1.2 g H_2O/60 g soil) as the soil was added air dry. It is also possible that due to a chromatographic effect, as a result of soil water and bromacil movement, the initial increments of soil water from the first increment of soil would contain a lower concentration of bromacil than the subsequent increments of soil water.

The constant concentration of bromacil in the soil water from 0.33 to 3.0 bars for each of the three soils might be an indication that the equilibrium between bromacil in the soil water and that adsorbed might be the same for the soil pores between 4.40 μ and 0.48 μ which correspond to the size of pores drained at the moisture tensions of 0.33 to 3.0 bars.

The decrease in the concentration of bromacil for the Coker clay, Woodburn silt loam, and quartz sand extracts might reflect a more adsorptive surface associated with soil pores smaller than 0.48 μ. Pore walls often contain clay and organic matter complexes which could account for increased adsorption of bromacil. Also, at the low soil moisture values, preferential adsorption for bromacil over water could result in a higher adsorption of bromacil. As the smaller pores drain at the higher tensions the possibility would exist for a portion of the extract to flow through larger pores already void of soil water. Competition between water molecules and chlorpropham was cited as one possible reason for a decrease in the chlorpropham concentration of extracts from kaolinite clay at a 15 bar tension (Scott and Lutz, 1971). Increased adsorption of simazine on the more available hydrophilic sites at low soil moisture values was hypothesized as an explanation for an increase in ED_{50} values as soil moisture decreased (Grover, 1965). The decreased toxicity of simazine at lower soil moisture values was explained by Scott and Lutz (1971) as due to the unavailability of soil water, and hence simazine, at soil water tensions greater than 15 bars.

The decrease in bromacil concentration of the soil water just discussed occurred in the last 20 ml of collected extract from the three soils and the quartz sand (Figure 3). This would tend to support the hypothesis that preferential adsorption of bromacil is responsible for the increased adsorption of bromacil at the higher soil moisture tensions.

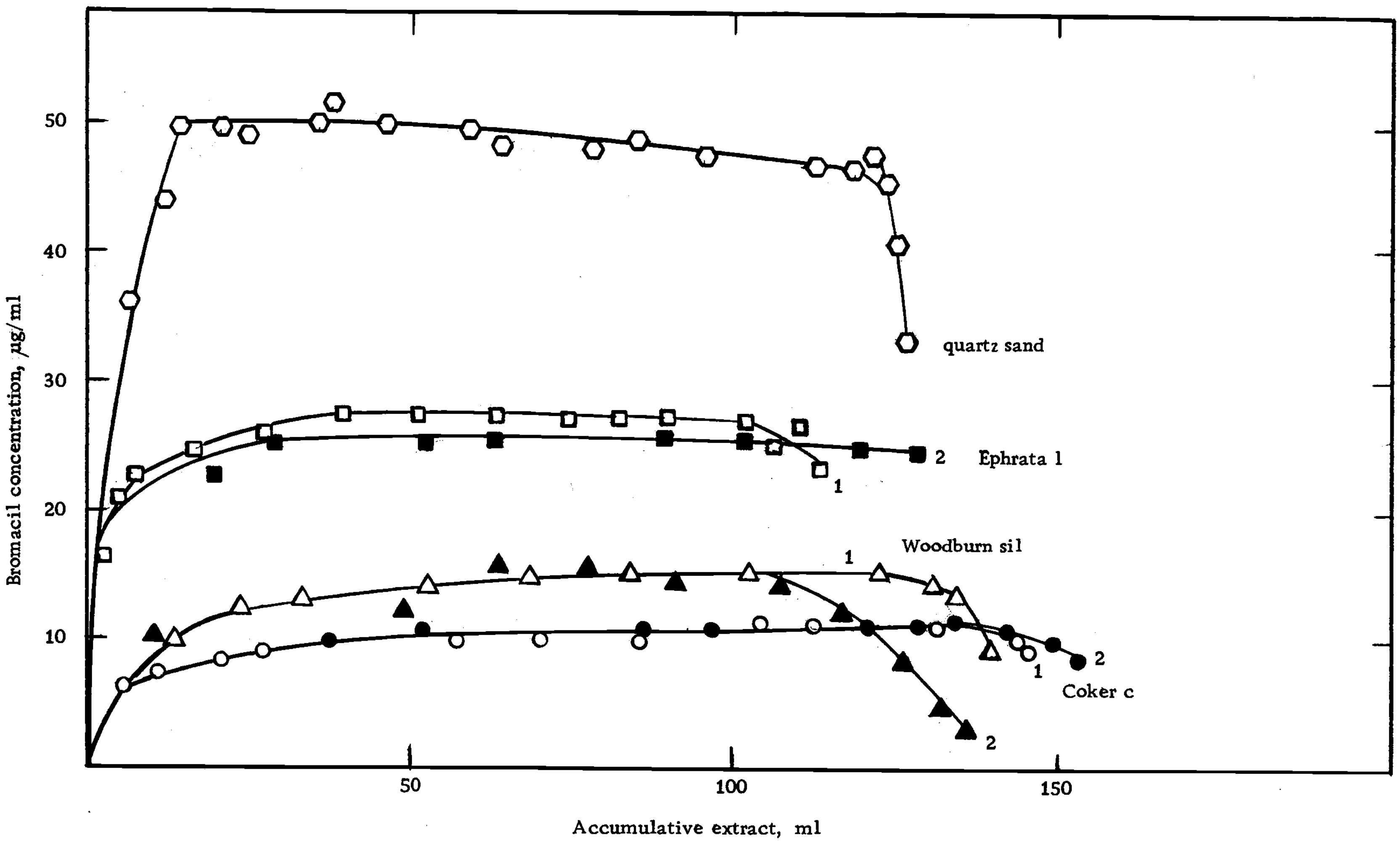


Figure 3. Bromacil concentration in the soil water extracts from three Oregon soils and a quartz sand

From a standpoint of bromacil concentration, bromacil should be equally available to plants from soils at field capacity as at a 4.0 bar soil moisture tension. Over this moisture tension range (0.33 to 4.0 bars) it would appear that toxicity of bromacil might be largely controlled by the influence of soil moisture on the plant system. In view of the bromacil concentration differences in the collected effluent for the soils at moisture tension values between 0.2 and 5.0 bars, bromacil phytotoxicity should increase for the soils studied in the order Coker clay < Woodburn silt loam < Ephrata loam < quartz sand.

Data collected from microbial degradation studies indicate that the maximum degradation of bromacil over a three-week period amounted to only 0.28% of the amount initially added to a Woodburn silt loam (Figure 5, appendix). Therefore, the decrease in bromacil concentration of the extracts at the higher soil moisture tensions cannot be attributed to microbial degradation.

Bromacil removal

Total quantities of bromacil released from the soils over the 15 bar tension range were 82, 45 (estimated),[1] 26, and 21% of that

[1] Presumably because of an equal pore size distribution very little water was extracted from the Ephrata loam at 9 bars. A 15 bar equilibration was not achieved for the Ephrata loam soil.

initially added to the quartz sand, Ephrata loam, Woodburn silt loam, and Coker clay respectively (Figure 4). The majority of bromacil was removed between the moisture tension values of 0.10 and 0.60 bars, indicating that movement of bromacil with the soil solution could be a problem at high soil moisture content.

The percentage of bromacil removed from the Ephrata loam, Coker clay, and Woodburn silt loam increased in direct proportion to the increase in percent soil water extracted (Figure 5). Even though the bromacil concentration in the extracts was lower for the Coker clay compared to the Woodburn silt loam, for a given percentage of water removed a larger percentage of bromacil was removed from the Coker clay relative to that removed from the Woodburn silt loam. Presumably this would be due to the larger absolute quantity of water removed from the Coker clay.

Adsorption of bromacil

Calculation of bromacil adsorption by the Woodburn silt loam, Coker clay, Ephrata loam, and quartz sand based upon Green's (Green and Obien, 1969) concept of 15 bar water indicate that the quantity of bromacil adsorbed by the soils is essentially constant throughout a wide moisture tension range (Table 2). Average adsorption values for the Coker clay, Woodburn silt loam, Ephrata loam, and quartz sand were 15.1, 14.0, 10.0, and 2.9 μg/g. As

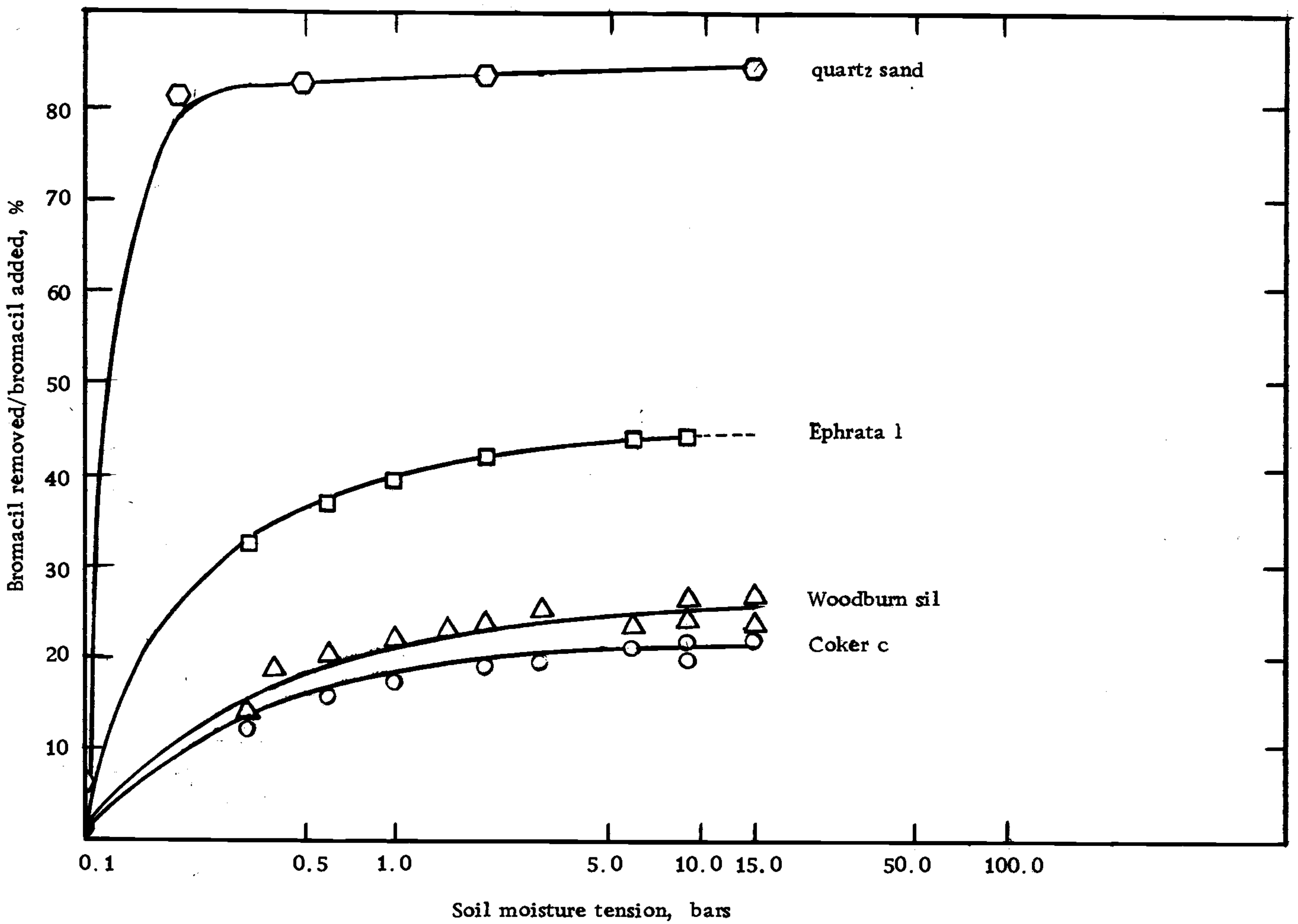


Figure 4. Bromacil removed from three Oregon soils and a quartz sand over a 15 bar tension range

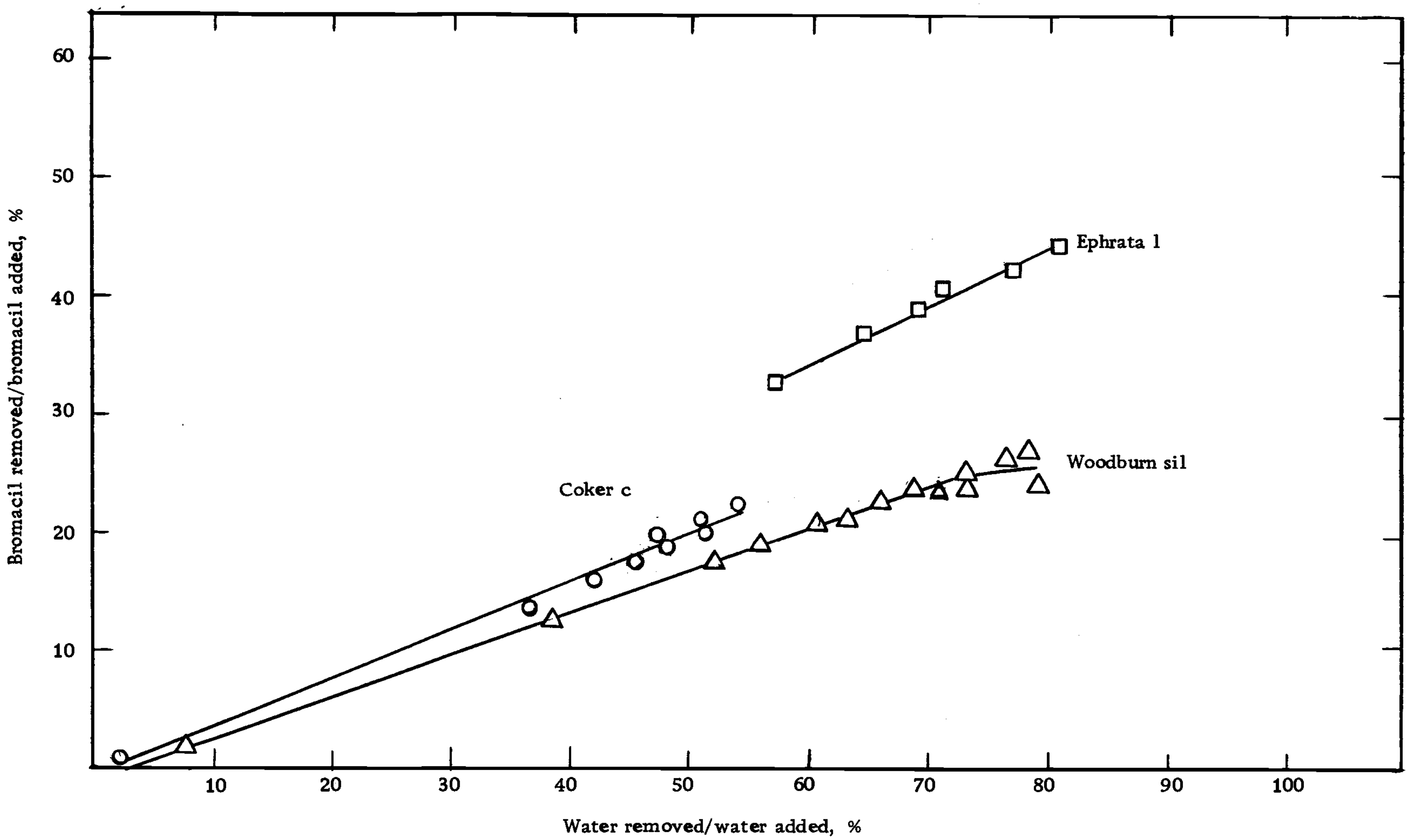


Figure 5. Bromacil removed as related to soil water removed from three Oregon soils

Table 2. Bromacil adsorption and K values for a quartz sand and several Oregon soils

Soil		Moisture tension bars								
		0.1	0.2	0.3	0.6	1.0	2.0	3.0	9.0	15.0
Woodburn silt loam										
	K value	1.6	--	1.1	0.9	1.0	0.9	1.0	1.1	--
	Adsorption, µg/g	15.3	--	14.4	13.8	13.8	13.8	13.8	13.8	--
Coker clay	K value	--	--	1.6	1.3	1.3	1.3	1.5	1.6	1.7
	Adsorption, µg/g	--	--	15.2	14.9	14.9	14.9	14.9	14.9	14.7
Ephrata loam	K value	--	--	0.4	0.4	0.4	0.4	--	0.4	--
	Adsorption, µg/g	--	--	10.0	9.7	9.8	10.1	--	10.1	--
quartz sand	K value	0.1	0.1	--	0.1	--	0.1	--	--	--
	Adsorption, µg/g	5.5	2.6	--	2.8	--	2.9	--	--	--

a relatively constant bromacil concentration was observed in the soil effluent, the value of K (μg bromacil per g soil divided by μg bromacil per ml soil solution) would also be constant for the soil water in all pore volumes (Table 2).

The average K values based upon the 15 bar water value were 1.5, 1.0, 0.4, and 0.1 for the Coker clay, Woodburn silt loam, Ephrata loam, and the quartz sand respectively (Table 2). Average K values based upon the adsorption isotherms show values of 1.0, 0.6, 0.3, and 0.1 for the Coker clay, Woodburn silt loam, Ephrata loam, and the quartz sand respectively. Except for the Coker clay and Woodburn silt loam, the K values calculated from the VPMA data and isotherm data agree relatively well. Calculations according to Green's (Green and Obien, 1969) predictive equation show that for herbicide-soil systems with K values less than one an increase in herbicide concentration can be expected as soil moisture decreases. The experimental data for the Woodburn silt loam, Coker clay, and quartz sand agree with Green's concept in that a decrease in the concentration of bromacil in the soil solution was observed as soil moisture decreased (Figures 2 and 3). The Ephrata loam did not demonstrate a decrease at the higher soil moisture tensions, but a 15 bar equilibration was not achieved for this soil.

Conclusions

Distribution of bromacil in the soil aqueous phase was studied as a function of soil texture and soil moisture. Soil texture influenced not only the concentration of bromacil in the soil water, but also the removal of bromacil from the soil system over the 15 bar moisture range. While Coker clay contained more than three times the amount of clay present in the Woodburn silt loam soil, the quantity of bromacil removed and the concentration of bromacil in the soil solution were very similar for the two soils. Soil organic matter may thus play a more important role than clay in determining the concentration of bromacil in the soil solution.

As soil texture and soil organic matter influence the distribution of bromacil in the soil solution, differences in bromacil phytotoxicity due to soil texture can be expected. Whereas the concentration of bromacil in the soil solution is relatively constant over a wide range of soil moisture values, soil moisture, at least from a bromacil concentration standpoint, should have little effect on bromacil phytotoxicity.

Of the total bromacil removed from each soil a high percentage was extracted at moisture tensions less than 0.50 bars. For the Coker clay, Woodburn silt loam, Ephrata loam, and quartz sand, 81, 73, 82 and 98% of the bromacil removed was extracted at tensions less than 0.60 bars. Significant removal of bromacil from the soil into runoff water should be a serious problem only when the soil moisture is above or slightly below field capacity.

Calculation of K values from the VPMA data showed values of 1.5, 1.0, 0.4, and 0.1 for the Coker clay, Woodburn silt loam, Ephrata loam, and quartz sand respectively. Values of K calculated from adsorption isotherms agreed well with the K values from the VPMA except for the Coker clay and Woodburn silt loam soils for which the K values were slightly lower. Low K values for the three soils and quartz sand predicted that as soil moisture decreased the concentration of bromacil in the soil solution should increase. An increase in bromacil concentration in the soil solution at high moisture tensions was observed for the Coker clay, Woodburn silt loam and quartz sand, but not for the Ephrata loam.

BROMACIL PHYTOTOXICITY AS INFLUENCED BY SOIL TEXTURE AND SOIL MOISTURE

Literature Review

Introduction

Several chemical and physical factors of a soil system influence the fate of soil applied herbicides. These factors include soil water, temperature, texture, pH, microbial activity, and organic matter. Equilibrium between the herbicide adsorbed on soil or root surfaces and that in the soil solution is greatly dependent upon two of the above factors, soil water and soil texture.

Root uptake of herbicide and water from light textured soils decreases the volume of water in the voids and generally increases the herbicide concentration (Hartley, 1964). This increase can be attributed to a lack of soil adsorptive surfaces. A similar phenomenon also occurs in heavy textured soils; however, because of the large number of colloidal particles in the system which contribute to adsorption, the concentration of herbicide generally does not increase.

Soil moisture

Soil moisture had a very significant effect on the toxicity of simazine to oats grown in a Regina clay soil (Grover, 1966). As soil moisture decreased from 60 to 30% three times as much simazine was needed to reduce the growth of oats by 50%. Grover hypothesized that as soil moisture decreased additional simazine was adsorbed on more hydrophilic sites.

Cotton grown in a Cecil sandy loam with no diuron added produced 50% more dry matter at high soil moisture levels than at low soil moisture levels (Upchurch, 1957). When diuron was mixed with the soil, the yield of green matter and dry matter, as well as plant height was the same at all moisture levels indicating that soil moisture had no absolute effect on diuron phytotoxicity. As soil moisture decreased the differential in plant weight between the herbicide treated plants and the check plants at each soil moisture level also decreased, indicating that soil moisture had a large relative effect on diuron phytotoxicity. Lambert (1966), utilizing data from the cotton-diuron experiment by Upchurch, demonstrated that as the concentration of diuron in the water phase increased, the green shoot weights expressed as a percentage of the unstressed cotton plant weight decreased. From these data it was concluded that the toxicity of diuron to cotton was constant at all soil moisture levels. The growth response of cotton to diuron was a function of soil moisture only in that the concentration of diuron in the water phase was a function of Kp (g compound/g organic matter divided by the concentration in the water phase), the sorption coefficient.

Igran toxicity to winter wheat grown on Chehalis loam was studied by Figuerola (1969) as a function of herbicide rate and soil moisture content. Igran applied at 2 lb/A caused serious injury at the highest soil moisture level (75-100% field capacity) but almost no injury at the lowest soil moisture value (25-50% field capacity). At the 8 lb/A application no difference in toxicity was observed at three moisture levels. Soil moisture content was thought to have a very important effect on the toxicity of Igran under conditions of high light intensity and temperature.

Toxicity of picloram to sunflower plants increased as soil moisture of a Regina clay soil decreased from 65 to 35%, slightly below field capacity (Grover, 1970). Increased toxicity was attributed to an increase in picloram concentration in the soil solution as soil moisture decreased. The soil pH was 7.7 under which conditions the adsorption of picloram was thought to be negligible.

Soil components

Soil pH did not significantly affect the phytotoxicity of the uracil herbicides although greater toxicity was observed with a sandy loam than with a clay loam soil (Pancholy and Lynd, 1969). Fresh weights of plants were a more sensitive indicator of bromacil phytotoxicity as the dry weights of the normal living plants were similar to the dry weights from the dead bromacil treated plants. Additions of organic matter to a Eufaula sand soil significantly reduced the toxicity of bromacil, which was highly correlated with an increase in the nitrogenous components of the organic and inorganic amendments.

Adsorption and movement of two uracils, triazines, phenylureas, and carbamate herbicides on a sandy loam and clay soil were studied by Hance (1967). Compared to the other herbicides, the two uracils showed low and intermediate adsorptions. Adsorption of both uracil herbicides was highest on the sandy loam soil which contained six times the organic matter of the clay soil. Uracil was not adsorbed on slurries of montmorillonite saturated with Na, Ni, Cu, Fe (III), Li, Mg, or Ca cations (Lailach, Thompson, and Brindley, 1968a, b).

Bromacil adsorption on nine Oregon soils was highly correlated with the percentage of organic matter (correlation coefficient with organic matter and Freundlich K value of 0.913) (Gaynor, 1971). On soils low in organic matter, bromacil adsorption appeared to be better correlated with the increase in exchangeable aluminum. Adsorption of bromacil by the nine soils increased as measured by the Freundlich K value with the bromacil concentration of the equilibrating solution.

The objective of this study was to determine the influence of soil moisture and soil texture on bromacil phytotoxicity as related to the uptake of bromacil by wheat grown at selected soil moisture values. Effects of bromacil on plant utilization of water as a function of time was also investigated.

Materials and Methods

Soil moisture content

Ephrata loam, Woodburn silt loam, and Coker clay were adjusted to four moisture levels (Table 1). The soil moisture percentage for each level was determined from soil moisture tension curves of each soil obtained by the use of a volumetric pressure membrane apparatus.

Wheat bioassay procedure

Each soil sample (150 g oven dry weight) was weighed into plastic bags which were then sealed. Eight soil samples were prepared for the study at each soil moisture level: No plants, check plants, and plants grown at two herbicide concentrations. Each treatment was conducted in duplicate. The wheat bioassays on the Coker clay and Woodburn silt loam soils were conducted simultaneously whereas the wheat bioassay on the Ephrata loam was completed at a later time.

Stock solutions of ^{14}C bromacil were prepared such that 5 ml of solution would contain either 75 μg (0.5 ppm based on oven dry soil) or 600 μg (4.0 ppm) bromacil. Five ml of the desired stock

Table 1. Soil moisture content for three Oregon soils at selected bar tension values

Soil series and texture	Moisture tension bars	Moisture percent
Coker clay	0.10	74.2
	0.20	55.0
	0.30	48.0
	2.50	38.0
Woodburn silt loam	0.10	44.0
	0.30	29.5
	0.40	20.6
	2.50	14.5
Ephrata loam	0.10	41.6
	0.25	20.5
	0.40	15.1
	1.00	11.5

solution was mixed with sufficient distilled water to bring the total volume of aqueous herbicide solution to that required for the desired soil moisture level.

Wheat seeds (Triticum aestivum L., var. Gaines) were pre-germinated for a three-day period. On the day of planting each 150 g portion of soil was placed in a beaker and the prepared aqueous herbicide solutions added. The soil was thoroughly mixed to ensure uniform distribution of water and herbicide in the soil. The water and herbicide was mixed directly with the soil in the plastic bags at the 0.1 bar moisture level. After mixing, the soil was transferred to 2.75 inch square plastic pots, lined with plastic. A straw perforated with holes was inserted in the center of each pot to three-quarters of the soil depth.

Fifteen uniform pre-germinated wheat plants were placed on the soil surface and covered with a thin layer of soil. Coarse expanded vermiculite was applied to the soil surface to reduce evaporation losses. The initial weight of each pot was determined to the nearest 0.01 gram.

After planting, the pots were placed in a growth chamber controlled for a 16-hour day at 23.8° C, a 15.6° C night, and a light intensity of 16,100 lumens/m^2. After three days the plants were thinned to ten wheat plants per pot. On each day until termination of the experiment the pots were weighed twice daily and sufficient water added via the straws to bring the pots back to their initial weights. After each weighing the pots were positioned in the growth chamber in a completely randomized fashion.

Plant harvest and tissue analysis

After thirteen days, the aerial portions of the plants were harvested, weighed fresh, and after oven drying at 40° C for one week. The plant growth at each level of herbicide application was expressed as a percent of the check plants at each soil moisture level. ED_{50} values, the amount of chemical required to give a 50% reduction in plant growth, were calculated at each moisture level and bromacil application rate.

The dry tissue was ground in a Wiley mill and 50 mg samples were weighed into liquid scintillation vials. Two-tenths ml perchloric acid followed by 0.5 ml of 30% hydrogen peroxide was added to the contents of each vial. Each vial was capped and placed in a water bath at 75° C for a six-hour digestion period. Upon complete digestion the vials were placed in an ice bath for one hour prior to the addition of the scintillation fluor. After cooling, 6 ml of cellusolve (ethylene glycol monoethyl ether) and 10 ml toluene phosphor solution containing 6 g PPO/l of toluene were added to each vial. In each digestion several plant samples which had not been treated with bromacil were spiked with ^{14}C bromacil to calculate a recovery percentage. The ^{14}C activity was counted for 20 minutes to a standard deviation of 2.5%. Bromacil concentration per gram of plant tissue was calculated. Water use by the plant was calculated by subtracting the values of the twice daily pot weighings from their respective initial weights. Evaporation as determined by water loss from pots which contained only soil was also subtracted from each pot weight.

Adsorption isotherm

Twenty-five ml of ^{14}C bromacil aqueous solution (1, 10, 50, or 100 ppm) was added to a 30 ml glass centrifuge tube which contained 5 g of air dry soil. The soil-bromacil system was placed in a water bath which was controlled at 25° C and shaken for 12 hours. After equilibration the soil-bromacil mixture was centrifuged at 9,000 revolutions per minute for ten minutes. Duplicate 1 ml samples of the supernatant were removed and counted for ^{14}C activity. Bromacil adsorbed to the soil was calculated and expressed as micrograms bromacil per gram soil.

Results and Discussion

Physical appearance of plants

Bromacil phytotoxicity symptoms were first observed on the sixth or seventh day of plant growth at the highest moisture level and highest bromacil application rate for each soil. Early bromacil toxicity symptoms were primarily a slightly lighter shade of green in the plant tissue. After the first week of plant growth the bromacil toxicity symptoms, lodging of the blades and a change in tissue coloration from a light green to a light brown, became more severe. Also as time continued the height of the check plants surpassed the height of the bromacil treated plants. At the lowest soil moisture level for each soil, the bromacil treated plants differed from the check plants only in being a slightly lighter shade of green and showing a slight reduction in plant height. The toxicity symptoms at the different bromacil application rates were detected earlier and were easier to distinguish as soil moisture increased.

Plant growth as related to soil moisture

Plant growth increased in direct proportion to the increase in soil moisture for the Ephrata loam, Woodburn silt loam, and Coker clay soils when no herbicide was present (Figure 1). The only exception was at the highest soil moisture level for the Woodburn silt

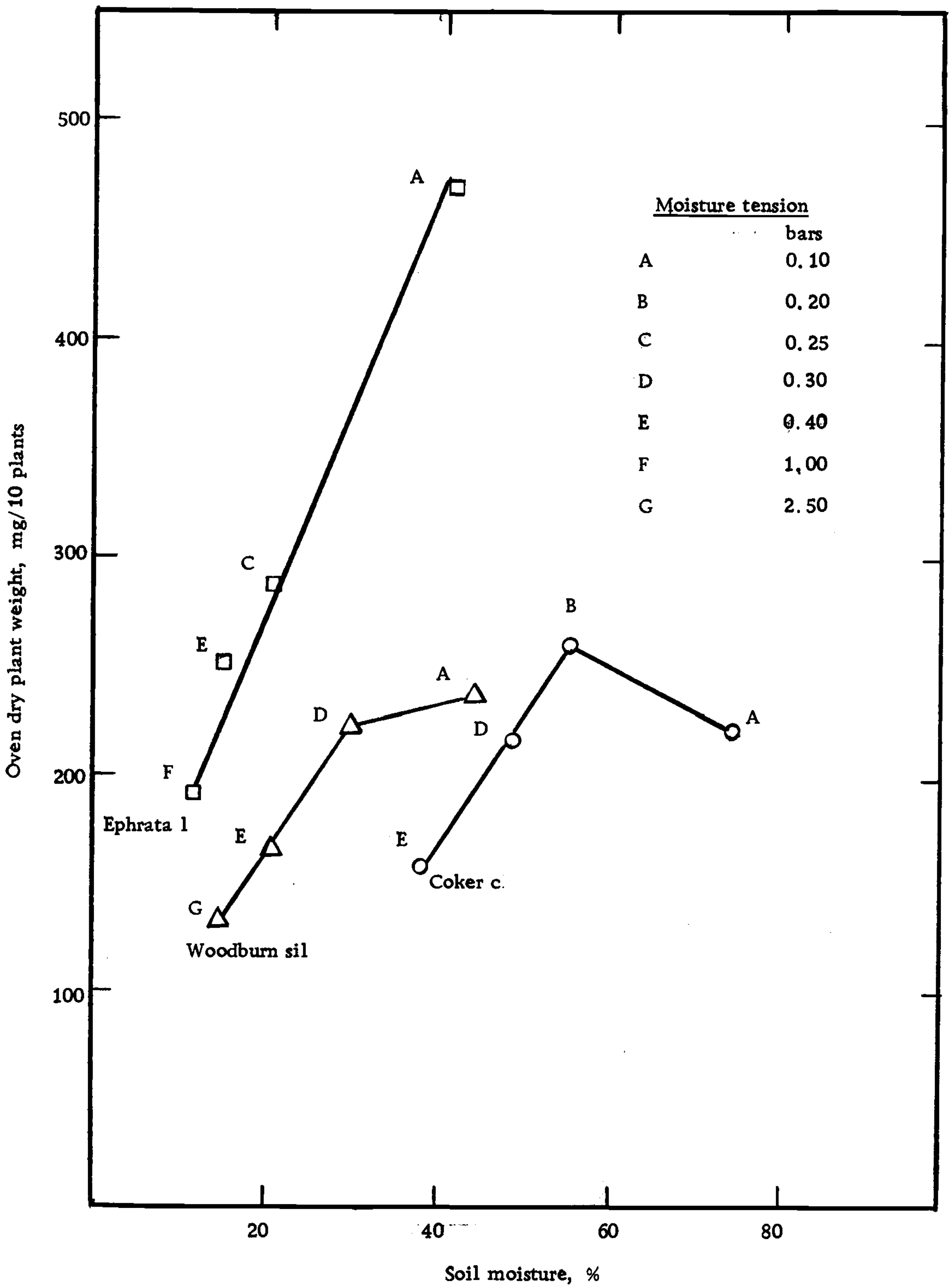


Figure 1. Wheat growth at several soil moisture contents

loam and Coker clay soils where plant growth was reduced. At extremely high soil moisture values plant physiological functions such as root respiration would become limited, and a resultant decrease in plant growth could be expected.

Bromacil severely reduced the quantity of water transpired by the plants grown in Ephrata loam at the 0.10 bar moisture level after 100 hours (Figure 2). The plants treated with 4.0 ppm bromacil transpired slightly larger amounts of water than the plants treated with 0.5 ppm bromacil. At the 0.25, 0.40, and 1.0 bar tensions, the herbicide treated plants transpired the same amount of water at both bromacil application rates, but less than the check plants after the first 100-hour growth period (Figure 3 and Table 2). Stomatal closure from high levels of CO_2 produced as a result of a decrease in photosynthesis is the suspected reason for the decrease in transpiration.

Similar observations in regard to quantities of water transpired can be made for the Woodburn silt loam (Table 2). The water usage data for the Woodburn silt loam are presented from a separate experiment in which non-labeled bromacil was utilized. The low quantities of water transpired by the plants grown in the Coker clay reflect the high humidity conditions of the growth chamber during the experimental period.

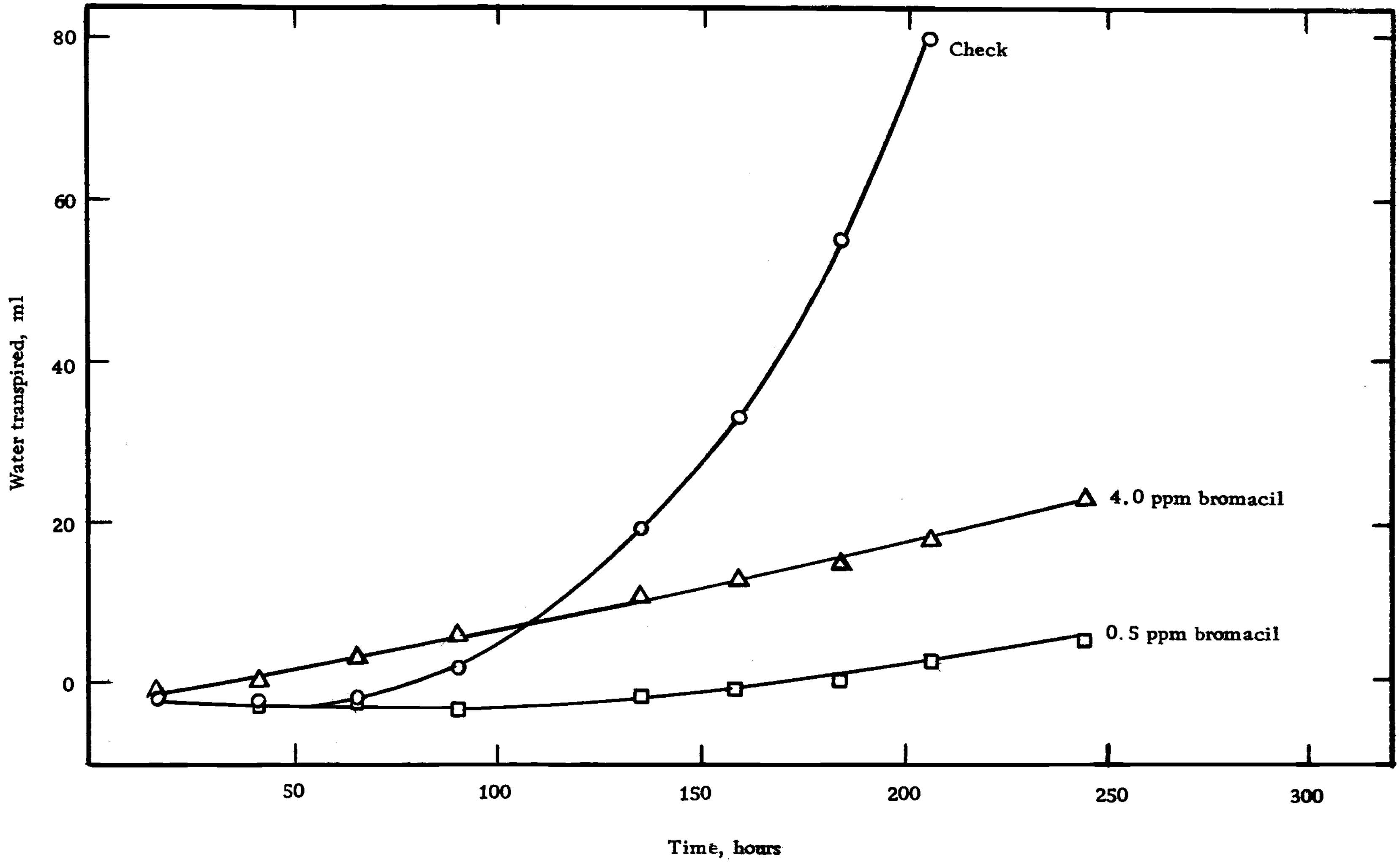


Figure 2. Water transpired by wheat plants grown in Ephrata 1 at 0.10 bar moisture

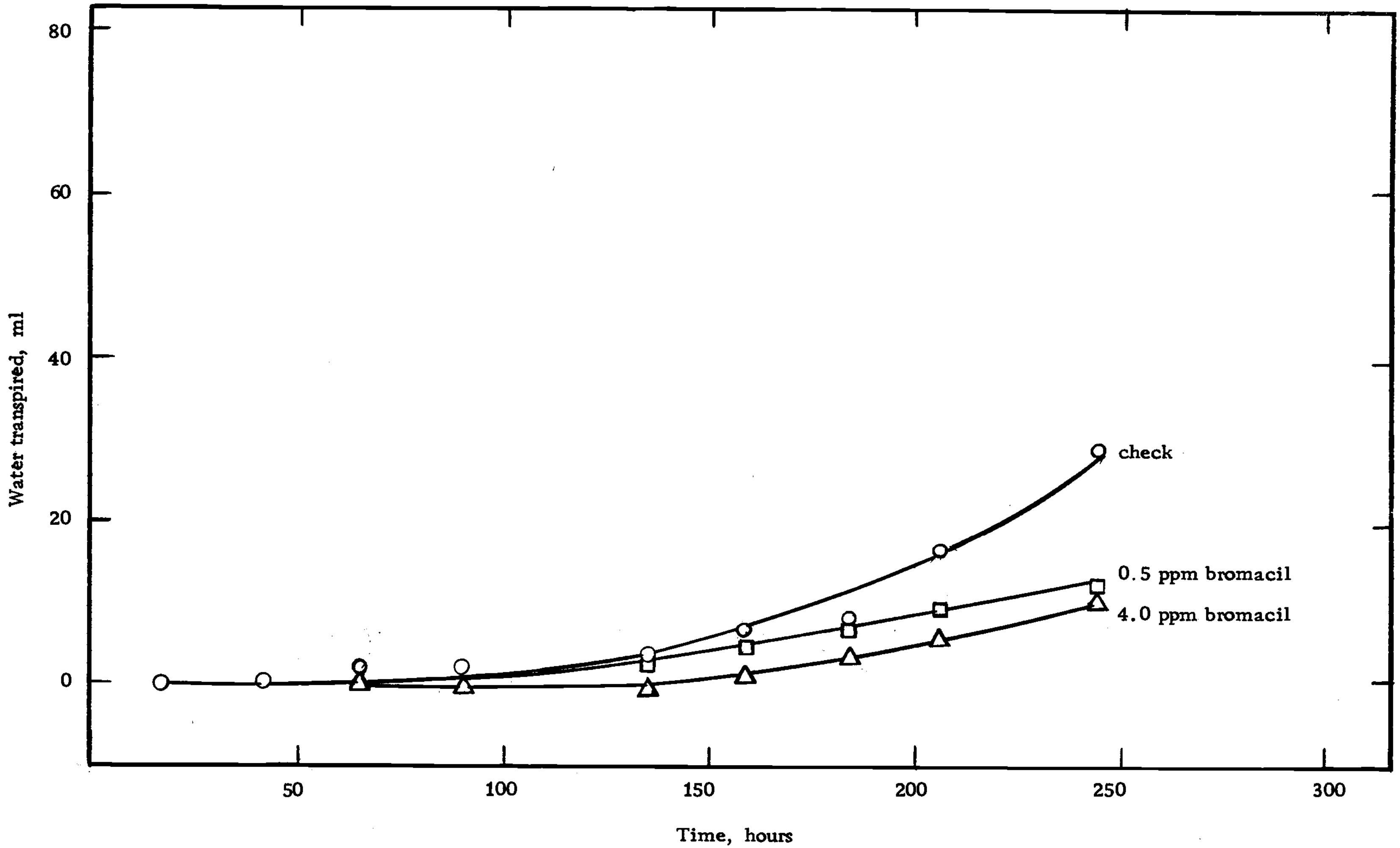


Figure 3. Water transpired by wheat plants grown in Ephrata 1 at 1.0 bar moisture.

Table 2. Water transpired by wheat grown in three Oregon soils

Soil	Moisture tension (bars)	Time (hours) 100			150			200		
		Bromacil application rate (ppm)								
		0	0.5	4.0	0	0.5	4.0	0	0.5	4.0
		Water transpired (ml)								
Ephrata loam	0.10	2.0	-3.0	6.5	27.5	-1.5	11.5	74.0	2.0	17.5
	0.25	5.6	1.0	3.0	17.5	4.0	6.5	42.5	7.0	8.6
	0.40	3.0	2.5	1.0	10.5	6.0	4.0	26.0	10.3	8.4
	1.00	2.0	1.6	-0.4	5.7	4.4	0.7	14.5	8.5	5.0
Woodburn silt loam	0.10	7.0	14.0	21.0	18.0	17.0	30.0	38.0	22.0	35.0
	0.40	9.0	5.0	6.0	18.0	8.0	9.0	32.0	10.0	13.0
	2.50	4.0	-5.0	3.0	3.0	-5.0	2.0	6.0	-4.0	1.0
Coker clay	0.10	3.8	2.6	0.40	8.2	6.6	2.3	10.8	7.8	3.6
	0.20	1.4	1.6	0.30	4.6	5.4	1.6	9.0	12.0	3.2
	0.30	1.3	1.4	2.30	3.2	3.8	4.8	6.2	6.5	7.1
	2.50	0.2	1.4	2.30	2.4	3.1	3.7	5.6	5.6	6.0

Bromacil uptake

At both the 0.5 and 4.0 ppm bromacil application rates, the concentration of bromacil in the plant tissue was highest for plants grown in Ephrata loam and lowest for plants grown in the Coker clay soils (Figure 4). The bromacil concentration in the plants grown on Woodburn silt loam was slightly higher than for the plants grown in the Coker clay soil. In general, as the percentages of soil clay and organic matter increased for the three soils in the order Ephrata loam < Woodburn silt loam < Coker clay, the uptake and concentration of bromacil in the wheat tissue decreased (Table 3). The difference in bromacil concentration in the plant tissue can be nicely explained by the differential in bromacil adsorption by the soils.

Table 3. Some physical properties of three Oregon soils

Soil series	Organic matter	Clay
	%	
Coker clay	4.70	63.7
Woodburn silt loam	3.10	17.0
Ephrata loam	0.56	11.8

Bromacil adsorption on the three soils increased in the order Ephrata loam < Woodburn silt loam < Coker clay when equilibrated in 1, 10, 50, and 100 ppm bromacil solutions (Figure 5). The adsorption isotherms indicate that at a given bromacil concentration

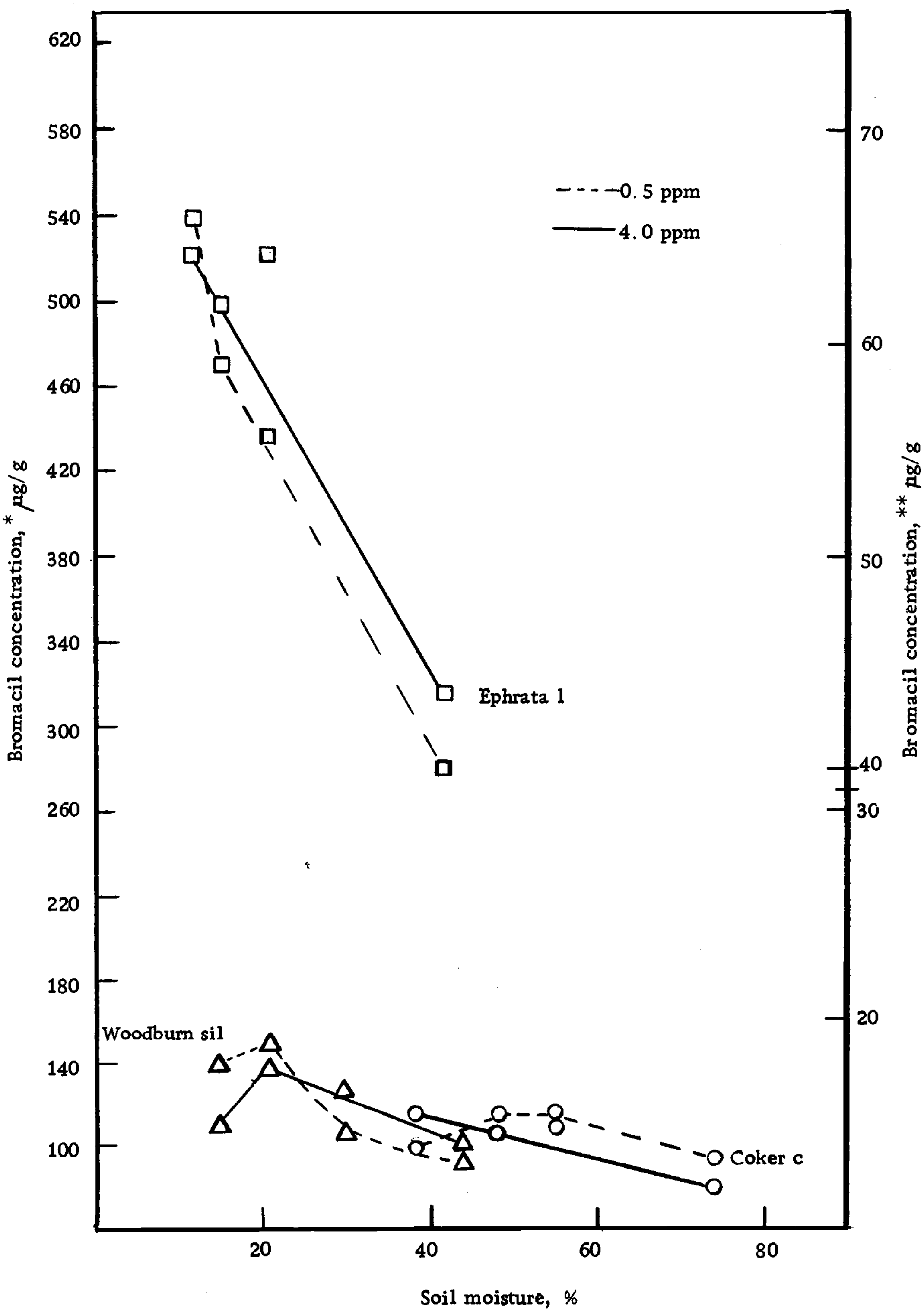


Figure 4. Bromacil concentration in wheat tissue

*Application rate of 4.0 ppm

**Application rate of 0.5 ppm

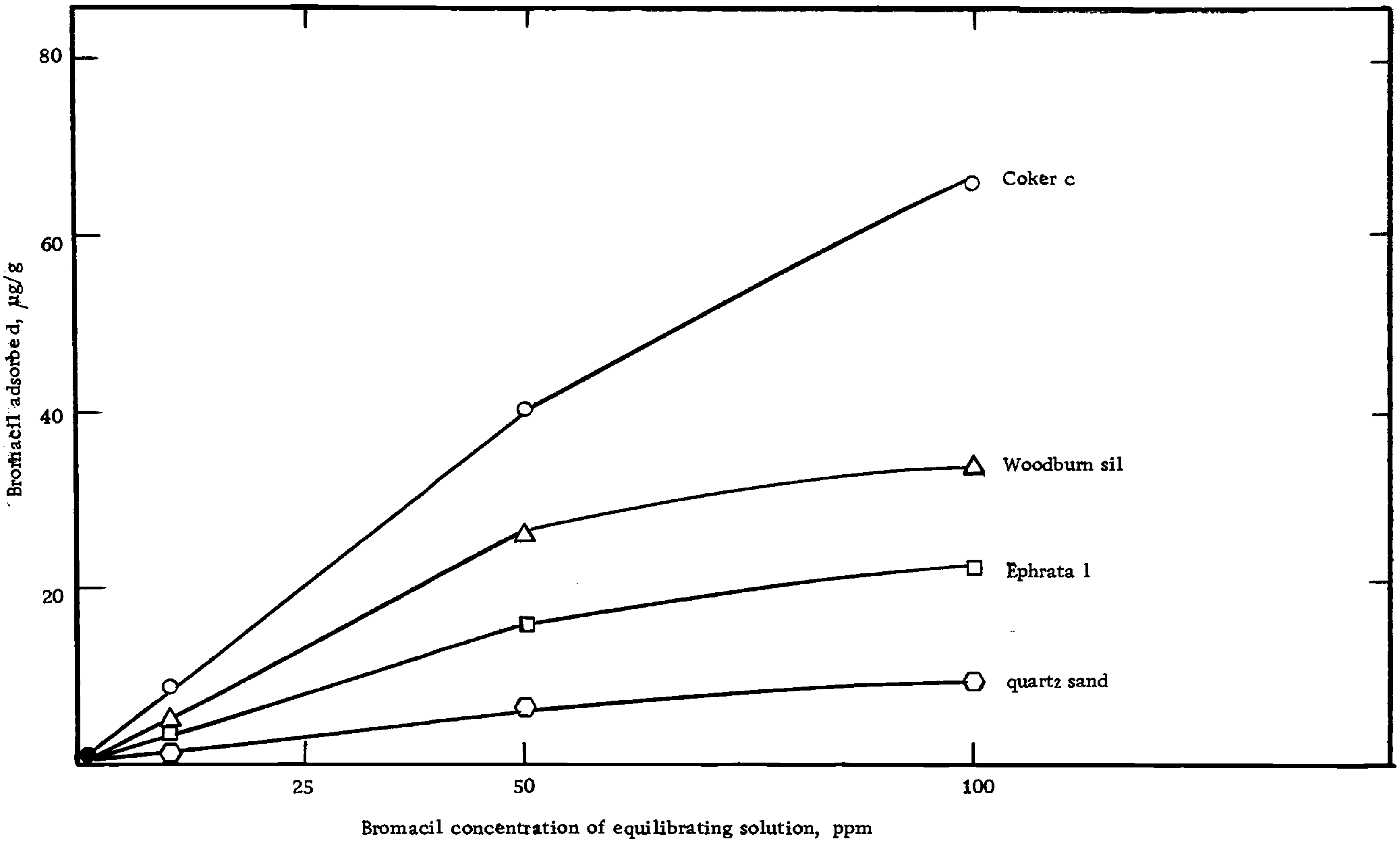


Figure 5. Bromacil adsorption on Coker c, Woodburn sil, Ephrata l, and a quartz sand

more bromacil would be in the soil solution in the order: Ephrata loam > Woodburn silt loam > Coker clay. A higher concentration of bromacil in the soil solution related directly to bromacil uptake by the plants. Uptake of ^{14}C labeled atrazine by corn increased as soil organic matter decreased, indicating that the amount of atrazine in the soil solution was quite influential in determining the amount of herbicide taken up by a plant (Lavy, 1968). For three soils of organic matter content which decreased from 4.4 to 1.7%, the ^{14}C activity of atrazine in corn leaf tissue increased from 2,800 to 11,000 counts per minute.

Differences in the relative humidity for the plants grown in the Ephrata loam soil compared to the plants grown in the Woodburn silt loam and Coker clay soils could also account for differences of bromacil concentration in the plant tissue. Plants were grown in the Coker clay and Woodburn silt loam soils during the same time interval and in the same growth chamber which resulted in high humidity conditions. Because of the fewer number of pots, the relative humidity was lower for the plants grown in the Ephrata loam at a different time period. Plants grown under high humidity conditions usually transpire at slower rates than plants grown at a low relative humidity. If a herbicide enters the plant root and is translocated with the transpirational stream, a faster accumulation of herbicide in the plant would take place under conditions of low relative humidity.

At both bromacil application rates, the concentration of bromacil increased in the plant tissue as soil moisture decreased (Figure 4). For the plants grown in the Ephrata loam, at the 0.5 ppm bromacil application rate, the concentration of bromacil in the plant tissue increased from 40 μg/g plant tissue at a soil moisture content of 42% to 66 μg at a soil moisture content of 11%. At the 4.0 ppm application rate bromacil concentration increased from 316 μg/g plant tissue to 522 μg/g at the same soil moisture values. Bromacil concentration in the wheat tissue also increased for the plants grown in the Coker clay and Woodburn silt loam, although increases in bromacil concentration were not as pronounced as for the Ephrata loam.

The increased bromacil concentration in the plant tissue with a decrease in soil moisture content can be explained on the basis that at the higher soil moisture values bromacil reduced or stopped plant growth earlier in the two week growth period than at the low soil moisture values. As plant growth decreased or stopped earlier at the high soil moisture levels the uptake of bromacil was also stopped or severely reduced. However, at the lower soil moisture values, the plants continued to grow and assimilate bromacil which by the end of the two week growth period had resulted in higher concentrations of bromacil in the plant tissue.

The quicker inhibition of plant growth at the higher soil moisture values can be explained by 1) the initial faster uptake of greater quantities of bromacil and 2) the faster expression of bromacil phytotoxicity. The growth curves of the check plants (no herbicide applied) indicate that the dry matter production increased in a direct proportion to the increase in soil moisture. As dry matter production increases in direct proportion to the rate of photosynthesis, the initial higher rates of photosynthesis at the higher soil moisture values could have allowed for a more rapid expression of bromacil phytotoxicity. Also, as transpiration rates usually increase at higher soil moisture values, the increased transpiration rates at the higher soil moisture values could have been responsible for the initial entry of larger quantities of bromacil into the plant system. As the translocation of a herbicide from the roots to the foliar portion of a plant may be inhibited by a decrease in soil moisture, it is also possible that herbicide movement from the xylem conduits to the chloroplasts would be reduced. If such a reduction in herbicide movement were to occur the toxicity of bromacil would be reduced at the lower soil moisture values as lesser quantities of the herbicide would reach the chloroplasts, the site of action for the bromacil molecule.

At both bromacil application rates the concentration of bromacil in the plant tissue decreased for the plants grown in the Woodburn silt loam at the lowest soil moisture value compared to the remaining soil moisture levels (Figure 4). A decrease in bromacil concentration for the plants grown in the Coker clay was also observed at the lowest soil moisture value and 0.5 ppm bromacil application rate. The decrease in bromacil concentration of the plant tissue at the lowest soil moisture values could be related to root permeability. A decrease in root permeability would make it more difficult for the bromacil molecule to enter the root, and hence limit the total uptake of bromacil into the plant system.

Except for the lowest soil moisture values there was a slight trend toward an increase in accumulation of bromacil in plant tissue as soil moisture decreased for the plants grown in the Woodburn silt loam and Coker clay soils (Table 4). The accumulation of bromacil in plant tissue as soil moisture decreased was much more pronounced for the plants grown in the Ephrata loam soil. At the 4.0 ppm bromacil application rate the accumulative uptake of bromacil for plants grown in the Ephrata loam increased from 36 μg to 49 μg bromacil as soil moisture decreased from 42 to 12 %.

Even though the bromacil concentration increased in the plant tissue as soil moisture decreased, the toxicity of bromacil to plant growth decreased (Figure 6). Bromacil was slightly more phytotoxic to wheat plants at the 4.0 ppm application rate than at the 0.5 ppm application rate. An equal reduction of plant growth at both bromacil application levels was observed for the plants grown in the Ephrata loam due to the larger quantity of bromacil present in

Table 4. Weight of bromacil treated plants grown in several Oregon soils and the resultant accumulation of bromacil

Soil	Moisture tension bars	Bromacil application rate ppm	Oven dry plant weight g/10 plants	Bromacil accum. µg
Ephrata loam	0.10	0.5	.1097	4.40
	0.10	4.0	.1156	36.48
	0.25	0.5	.1006	5.61
	0.25	4.0	.0950	49.72
	0.40	0.5	.0988	5.89
	0.40	4.0	.1009	50.21
	1.00	0.5	.1048	6.71
	1.00	4.0	.0932	48.64
Woodburn silt loam	0.10	0.5	.1184	1.57
	0.10	4.0	.0981	9.95
	0.30	0.5	.1084	1.57
	0.30	4.0	.1011	12.78
	0.40	0.5	.0937	1.79
	0.40	4.0	.0939	12.87
	2.50	0.5	.0983	1.75
	2.50	4.0	.0844	9.27
Coker clay	0.10	0.5	.1178	1.58
	0.10	4.0	.0992	7.74
	0.20	0.5	.1208	1.55
	0.20	4.0	.1018	11.12
	0.30	0.5	.1095	1.68
	0.30	4.0	.1066	11.17
	2.50	0.5	.0972	1.34
	2.50	4.0	.0922	10.58

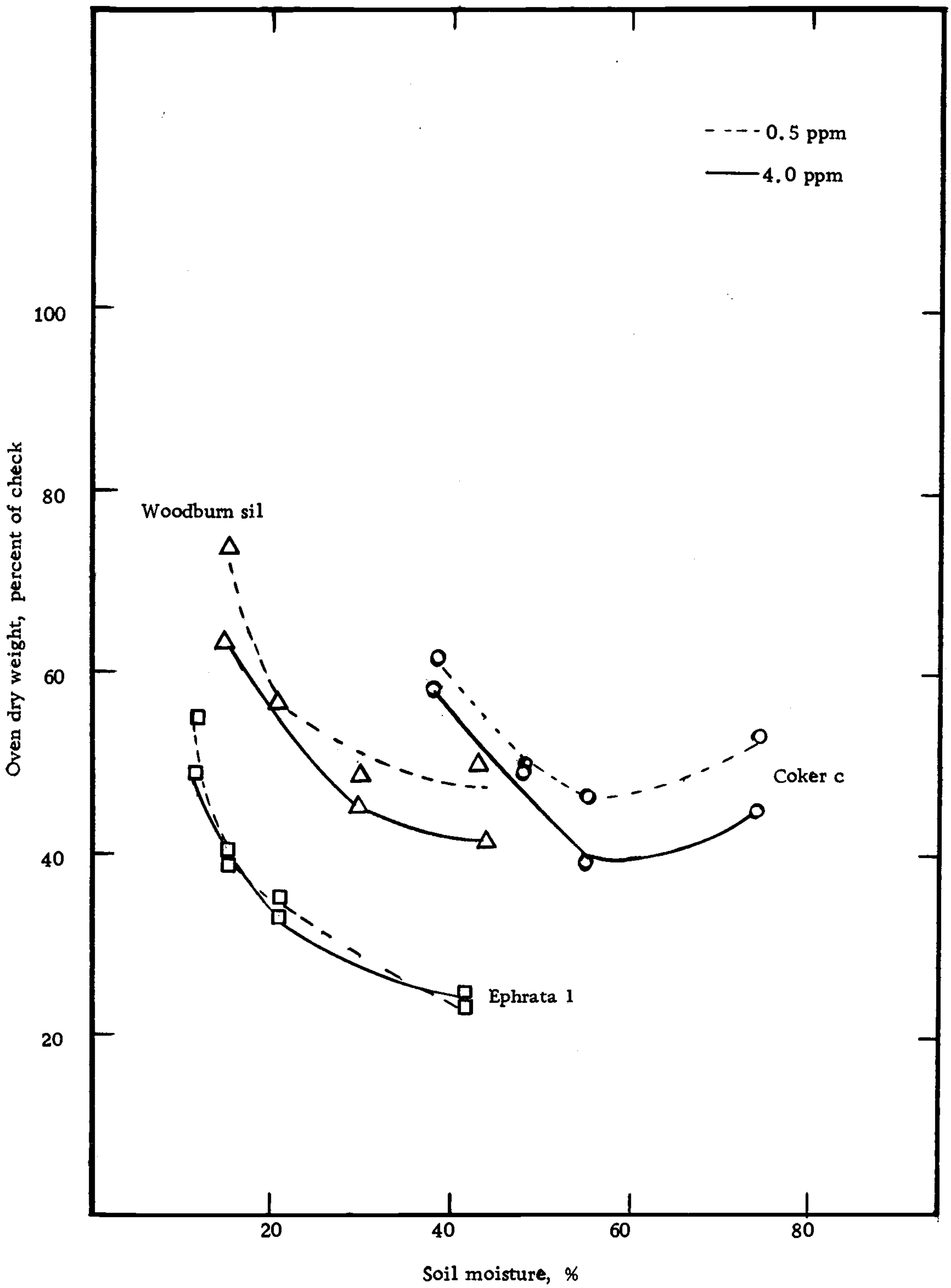


Figure 6. Wheat growth as affected by bromacil and soil moisture

the plant tissue. As the soil moisture of the Ephrata loam decreased from 42 to 12% the plant growth increased from 25 to 49% of the check plants at their respective soil moisture levels. Since the weight of the check plants grown in Coker clay at the 74% soil moisture level was less than the weight of the check plants at the 55% moisture value, bromacil toxicity to plant growth appeared to be less on a relative basis at the 74% moisture level as compared to the lower soil moisture value. However, on an absolute basis, plant growth was reduced as much if not more at the 74% soil moisture level as compared to the 55% soil moisture value (Table 4).

ED_{50} values (the amount of bromacil required for a 50% reduction in plant growth) were calculated for each soil at both bromacil application rates and for all soil moisture levels (Table 5). The ED_{50} values for each soil at both bromacil application rates increased in direct proportion to the decrease in soil moisture, except at the highest soil moisture values. In studies of soil moisture effects on simazine toxicity to oats, ED_{50} values were also observed to increase as soil moisture decreased (Grover, 1966).

Conclusions

Bromacil uptake by wheat was studied as a function of soil texture and soil moisture. As percentages of soil clay and organic matter decreased, the uptake and concentration of bromacil in wheat

Table 5. Bromacil ED_{50} values for wheat grown in three Oregon soils

Soil	Moisture tension bars	Bromacil application rate ppm 0.5 ppm	4.0 ppm
		ED_{50} value	
Ephrata loam	0.10	0.23	2.00
	0.25	0.35	2.65
	0.40	0.39	3.20
	1.00	0.55	3.90
Woodburn silt loam	0.10	0.50	3.33
	0.30	0.49	3.62
	0.40	0.57	4.54
	2.50	0.74	5.06
Coker clay	0.10	0.53	3.59
	0.20	0.46	3.13
	0.30	0.51	3.94
	2.50	0.62	4.68

tissue increased. The maximum concentration of bromacil in the plant tissue grown in the Ephrata loam, Woodburn silt loam, and Coker clay soils was 523, 137, and 115 μg/g plant tissue respectively at the 4.0 ppm bromacil application rate.

Soil moisture variables within each soil type influenced not only the uptake of bromacil, but also the phytotoxicity of bromacil inside the plant system. As soil moisture decreased from 42 to 12% for the Ephrata loam soil the accumulative uptake of bromacil at the 4.0 ppm application increased from 36 to 49 μg bromacil, yet the plant growth increased from 25 to 49% of the check plant weight in the same soil moisture range. In general similar observations, but less dramatic, were also made for the plants grown in the Coker clay and Woodburn silt loam soils. The initial faster rates of transpiration and photosynthesis at higher soil moisture values for each soil are hypothesized explanations for the increased toxicity of bromacil at the higher soil moisture values. A faster and more complete control of weeds should be obtained if bromacil is applied to a soil at or near field capacity than at drier soil conditions.
At each moisture level for the three soils, bromacil reduced the quantity of water transpired to less than that of the check plants.

Previous studies have shown that bromacil is present at the same concentration in the soil water over a wide range of soil moisture values. However, the present investigation has shown that bromacil phytotoxicity does vary with the soil moisture content. A study to further illucidate the fate of bromacil in the plant system as a function of time and soil moisture was completed to help explain bromacil phytotoxicity differences due to soil moisture.

BROMACIL UPTAKE BY WHEAT UNDER SOIL MOISTURE STRESS CONDITIONS

Literature Review

Introduction

Bromacil, (5-bromo-3-sec-butyl-6-methyluracil) is generally used on non-cropland areas for control of grasses and broadleaf weeds (Herbicide Handbook of the Weed Society of America, 1967). Its effectiveness may be influenced by soil water not only in the soil system, but also by the effects of soil water on the plant system. Herbicide transport through the soil to root surfaces occurs by two processes: mass flow and molecular diffusion. Soil water affects these processes by influencing both the rate of water movement and herbicide concentration in the soil. Plant physiological functions such as transpiration, photosynthesis, and root permeability are also controlled by soil moisture, which in turn may influence bromacil phytotoxicity.

Mechanism of action

The substituted uracil herbicides are potent and specific inhibitors of the Hill reaction. Addition of 0.5 ppm bromacil to spinach chloroplasts resulted in a 50% reduction of oxygen evolution whereas a 1.6 ppm concentration completely inhibited oxygen evolution (Hoffmann, McGahen, and Sweetser, 1964). Bromacil added at 4.0 ppm to illuminated Euglena cells stopped the production of oxygen within a ten-minute period. Concentrations of bromacil as high as 50 ppm had no effect on the respiration of the Euglena cells. Additional studies with bromacil and isocil demonstrated that a 1.4×10^{-6} M concentration of bromacil was required for a 50% inhibition of the Hill reaction in chloroplasts isolated from turnip greens. Inhibitory action was attributed to possible hydrogen bonding between the imino hydrogen of the nitrogen atom at the number one ring position, and the carbonyl oxygen of the carbon atom at the number two ring position with unidentified active sites in the chloroplasts (Hilton _et al._, 1964). Bromacil had no significant effect on non-photosynthetic tobacco callus tissue at concentrations below 10^{-3} M. At higher concentrations, growth was reduced by 16% of the control weight tissue (Jordon _et al._, 1966).

Recent studies have been completed concerning the degradation products of bromacil which occur in plant systems. The uptake and subsequent metabolism of ^{14}C bromacil was studied by placing two-year-old orange trees in nutrient solutions containing 10.3 ppm ^{14}C bromacil (Gardiner _et al._, 1967). After 27 days the roots, lower stems, upper stems, and leaves contained bromacil and its metabolites in concentrations of 8.5 ppm, 1.0 ppm, 1.1 ppm, and 1.2

ppm respectively. The compounds identified from tissue extracts were bromacil, 5-bromo-3-sec-butyl-6-hydroxy-methyluracil and an unknown metabolite in the ratio of 10:4:1. Bromacil was not metabolized to $^{14}CO_2$ as evidenced by lack of $^{14}CO_2$ evolution. Less than 5% of the total activity applied to the nutrient solutions was found in the plants.

Herbicide effect on photosynthesis and transpiration

If a herbicide is to be translocated via the transpirational stream, many barriers must be crossed as the herbicide enters the plant through the root system. The ion or molecule in question must diffuse across the apoplast of the root hair and come in contact with the outer symplast surface. Molecules accumulate at the outer symplast surface and are adsorbed to concentrations higher than the bulk solution surrounding the root. Movement then proceeds through the epidermis, across the cortex, through the endodermis, and into the stele. Migration continues through the stele to the apoplast or xylem conduits where movement may proceed rapidly to the foliar regions and possible re-entry into the symplast (Crafts, 1964).

Utilizing nutrient solutions, labeled herbicides, and autoradiographic techniques, Crafts and Yamaguchi (1960) studied the rate of movement of several herbicides from the root to the foliar portions of several plants. Atrazine and monuron, both inhibitors of the Hill reaction, were found to move rapidly from the roots to the foliar portions of several plants. Rapid translocation from the roots to the foliar portions would be an indication of movement via the transpirational stream through the xylem elements. Although movement could occur through the phloem tissues from the root, such movement would occur at a much slower rate. In a study on snap beans, Smith and Buchholtz (1964) found that isocil at 300 ppm reduced the bean transpiration rate by 45% within two and one-half hours.

Accumulative uptake of simazine from nutrient solution increased in both the foliar and root tissues of oat and cotton seedlings as a function of time (Sheets, 1961). Larger amounts of simazine were usually found in the foliar tissue relative to that in the root tissue. Conditions that increased the rate of transpiration also increased the concentration of simazine in the plant tissue; however, the simazine uptake per ml of water transpired rarely equaled the simazine concentration in the nutrient solution.

The uptake and translocation of ^{32}P phorate to cotton leaves has been studied in nutrient and sand cultures under conditions of different transpirational demand (Hacskaylo et al., 1961). The cotton transpired more rapidly at 35.0° C and 35% relative humidity than at 23.8° C and 95% relative humidity. In sand culture the phorate accumulation in the leaves increased linearly with time during the seven-day experiment. The movement of phorate from roots to leaves and the total phorate uptake was positively correlated with transpiration rates.

Most of the research on the uptake and translocation of herbicides has been accomplished with either nutrient solutions or foliar sprays. A noted exception was work completed by Sedgley and Boersma (1969) on the uptake of diuron at different soil moisture levels. Osmotic solutions (osmotic pressure controlled by Carbowax 6,000) containing diuron were used to control moisture stress on wheat plants grown in soil encased by a cellulose membrane. A linear relationship between the decrease in photosynthesis and water uptake was noted at each moisture tension. The relative rate of photosynthesis decreased more rapidly for plants grown at a 0.30 bar tension than for plants at a 2.5 bar tension. Root permeability was cited for the smaller decrease in the rate of photosynthesis per unit volume of water transpired at the 2.5 bar tension as compared to the 0.30 bar tension. Transpiration rates decreased during the first few days and then remained constant throughout the experiment.

In general, decreased transpiration rates from herbicide applications are a result of three different mechanisms: 1) inhibition of water uptake by roots, 2) increased cell permeability leading to dehydration of protoplasm in the guard cells, or 3) inhibition of photosynthesis resulting in CO_2 levels to which guard cells are sensitive. Decreased rates of plant transpiration, photosynthesis, and/or CO_2 fixation have been studied with atrazine (Wills, Davis, and Funderburk, 1963), simazine (Ashton, Zweig, and Mason, 1960), phenylmercuric acetate (Shimshi, 1963), and isocil (Hoffmann, McGahen, and Sweetser, 1964).

Soil moisture stresses often cause stomatal closure that reduces both photosynthesis and transpiration. Increased water stress decreased the rates of photosynthesis and transpiration for tomatoes and loblolly pine as demonstrated by Brix (1962). Photosynthesis and transpiration rates of Monterey pine were shown to decrease consistently with increasing soil moisture stress (Babolola, 1967).

The objective of this investigation was to relate the transpiration rate of wheat as influenced by soil moisture stress to the quantitative uptake of bromacil. Bromacil distribution in the root and foliar portions as a function of time was also investigated, as well as the influence of bromacil on the transpiration rate of the plant.

Materials and Methods

Preparation of wheat plants for osmotic solution

Wheat seeds (*Triticum aestivum* L., var. Gaines) were pre-germinated in glass trays for a three-day period. Ten of the seedlings were planted in each of 70 plastic soil cells, 30 cm × 8 cm ×

0.8 cm, which contained approximately 250 g of air dry Woodburn silt loam (Babolola, 1967). The wheat plants were grown for three weeks in a growth chamber controlled for a 16-hour day at 23.8° C, a 15.6° C night, and a light intensity of 16,100 lumes/m^2. On days one through seven 3 ml of water was added twice daily to the surface of each cell. The plants were thinned after five days to five plants per cell. On days eight through 19 the soil cells were subirrigated. The subirrigation water was replaced with one-half strength Hoagland solution on days 11, 16, and 19. Extra nitrogen (50 ppm ammonium nitrate) was added with the third Hoagland solution treatment.

Wheat growth in osmotic solutions

Twelve hours before the twentieth day the cells were removed from the growth chamber and excess water allowed to drain. On the twentieth day the plastic sides of the soil cells were removed and replaced with cellulose membranes. The soil cells were then equilibrated for 36 hours in osmotic solutions of 0.35 and 2.5 bar tensions which corresponded to 25.0 and 13.2% soil moisture for Woodburn silt loam. Carbowax 6,000, a polyethylene glycol with a molecular weight of 6,000, was utilized to control the osmotic pressure at the 0.35 bar tension (48.0 g/l) and at the 2.5 bar tension (150.0 g/l).

Following equilibration in the osmotic solutions, the soil cells were transferred to 20 individual osmotic chambers in an environmental growth room. Three soil cells (15 plants) were placed in each osmotic chamber. The soil moisture stress was controlled at either a 0.35 bar or a 2.5 bar tension. In experiment one the osmotic chambers contained no herbicide. In experiment two and three each chamber contained ^{14}C labeled bromacil at a concentration of 2.0 μg/ml, total of 10,880 μg or 4.5 μg/ml, total of 24,480 μg.

The wheat plants were grown under continuous light, a temperature of 24° C, relative humidity of 45%, wind speed over the plants at approximately 6 m/sec, and a light flux of 11,800 lumens/m^2 at a height of 20 cm above the soil surface.

Wheat plant harvest

Two osmotic chambers or 30 plants at each of the two moisture tensions were harvested on the second, fourth, sixth, eighth, and tenth day after placement in the herbicide and moisture tension controlled solutions. The plants were weighed fresh and after drying in an oven at 40° C for one week. The soil was removed from the plant roots in three soil cells selected at each osmotic tension and harvest date. Fresh and dry root weights were determined.

In each of the three experiments six soil cells were harvested prior to transfer to the herbicide solutions to obtain an initial plant weight. Incremental growth rates were computed with the increase or decrease in foliar growth measured at the end of each two-day harvest period, based upon the initial weight of the plants removed from the growth chamber.

Transpiration measurements

Transpiration was measured on each osmotic chamber with a Mariotte bottle constructed from a 100 ml burette. A constant level of water was maintained in the chambers throughout the experiment. Water transpired by the plants was replaced in the chambers from the burettes. The water level in each burette was measured every two hours, water loss calculated, and transpiration expressed as ml of H_2O per hour per 15 plants. Each chamber was completely sealed with modeling clay to eliminate evaporation.

Plant tissue analysis

Plant tissue samples of both the roots and foliar portions were digested and counted by liquid scintillation for ^{14}C activity (Mahin and Lofberg, 1970). Fifty mg of plant tissue was placed in a liquid scintillation vial to which was added 0.2 ml perchloric acid followed by 0.4 ml of 30% hydrogen peroxide. Contents of the vials were swirled after each addition of reagent. Each vial was capped and placed in a water bath at 75° C for a six-hour digestion period. Upon complete digestion the vials were placed in an ice bath for one hour prior to the addition of the scintillation fluor. After cooling, 6 ml of cellusolve (ethylene glycol monoethyl ether) and 10 ml of toluene phosphor solution containing 6 g PPO/l of toluene were added to each vial. In each digestion several plant samples which had not been treated with bromacil were spiked with ^{14}C bromacil to calculate a recovery percentage. The ^{14}C activity was counted for 20 minutes to a standard deviation of 2.5%. The bromacil concentration per gram of plant tissue was calculated.

Results and Discussion

Plant growth and uptake of bromacil

Bromacil accumulation in the root and foliar portions of wheat increased as a linear function over the ten-day growth period (Figure 1). The bromacil uptake by the plants grown at the 0.35 bar tension and 4.5 ppm bromacil concentration was erratic. The leveling off in accumulation of bromacil between the eight and ten-day period could have been a result of the larger amount of bromacil accumulated in the plant tissue resulting in a quicker termination of plant growth.

The accumulation of bromacil in the root tissue also increased with time. The bromacil increase however was more curvalinear in roots than in shoots with respect to time (Figure 2). Since the

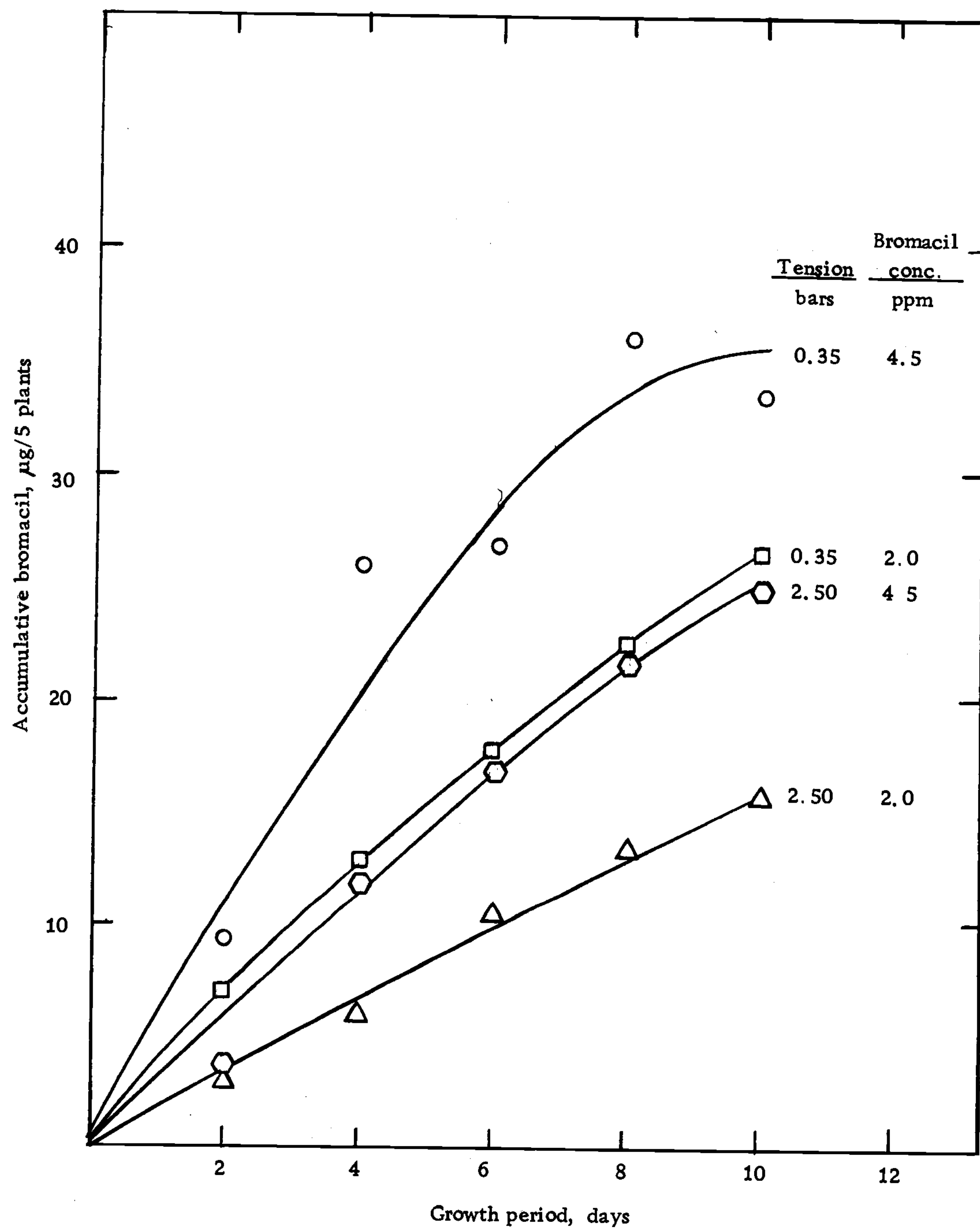


Figure 1. Bromacil accumulation in root and foliar tissue of wheat

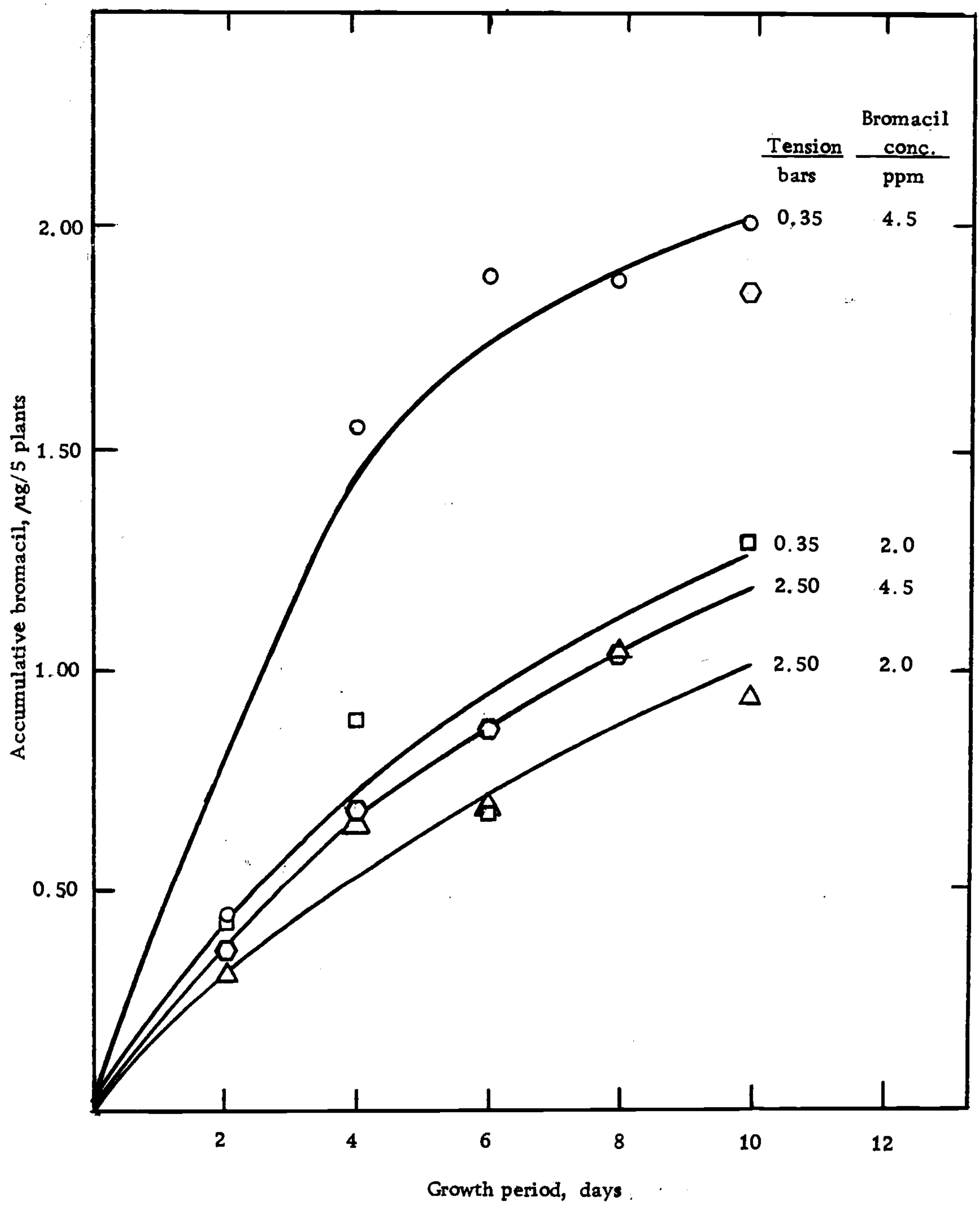


Figure 2. Bromacil present in the root tissue of wheat

bromacil accumulated in the roots was only 5 to 10% of that accumulated by the total plant, the bromacil accumulated by the total plant (Figure 1) essentially represents foliar accumulations. The total amount of bromacil accumulated in the 15 plants in each osmotic chamber was but a small percentage (0.5% maximum) of that initially added to the osmotic chamber.

The bromacil concentration in both the root and foliar tissues increased linearly with the age of the plant and exposure to the chemical in the osmotic solution (Figures 3 and 4). After eight days the bromacil concentration in the foliar tissue increased less rapidly. The wheat plants grown in the high bromacil concentration at the 0.35 bar tension were essentially dead, while the other plants, although showing injury symptoms, were still growing.

At both soil moisture tension values, 0.35 bar and 2.5 bar, nearly twice as much bromacil was present in the total plant and in the root tissue at the high application as compared to the low application rate (Tables 1 and 2). While the ratio of bromacil concentrations (high rate/low rate) in the osmotic chambers was 2.25, the same ratio ranged only from 1.1 to 2.1 based on bromacil concentration and accumulation in the plant tissues. Therefore, neither the concentration nor the accumulation of bromacil in the plant tissue was in direct proportion to the concentration of bromacil in the osmotic chambers.

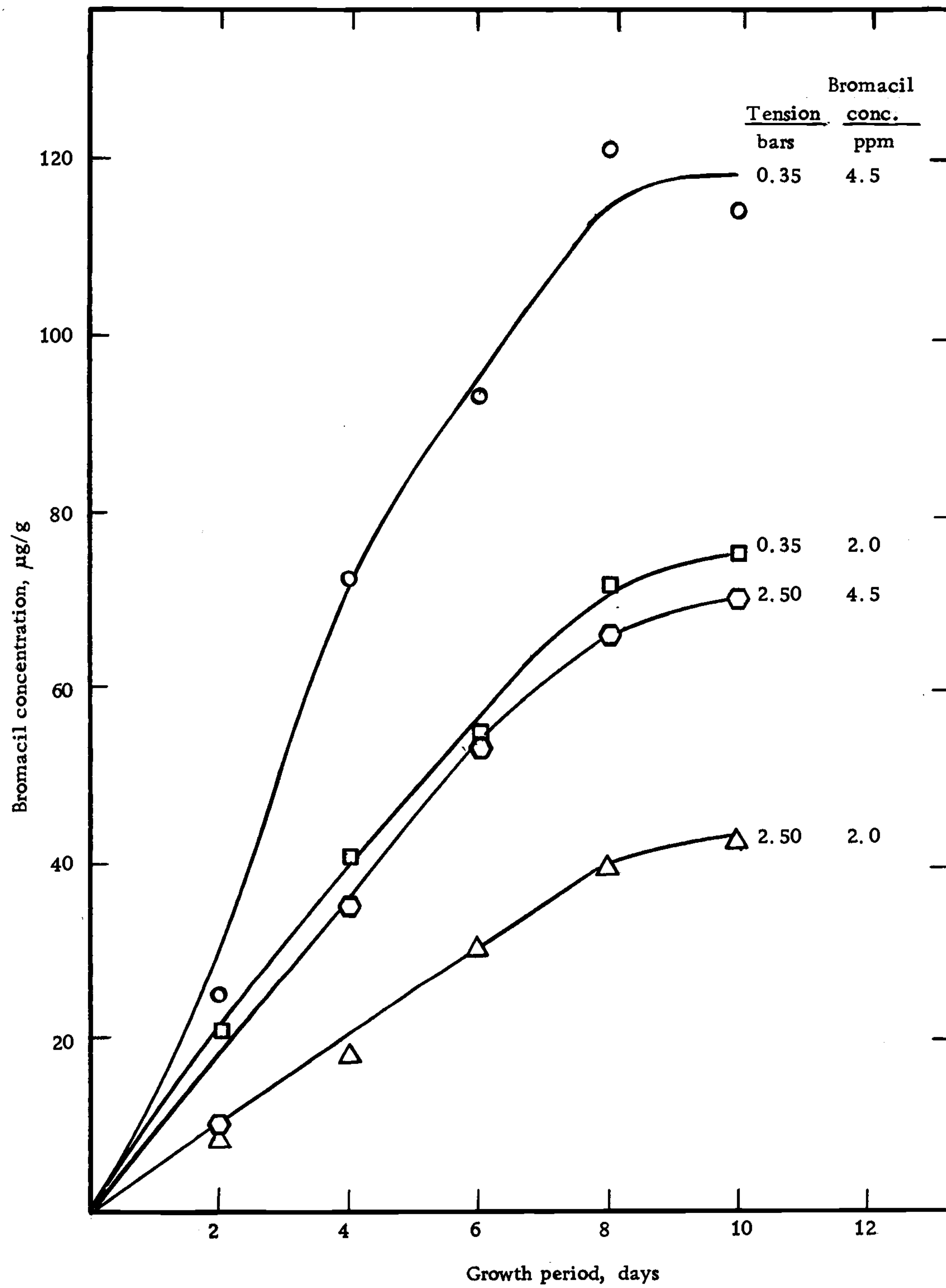


Figure 3. Bromacil concentration in the foliar tissue of wheat

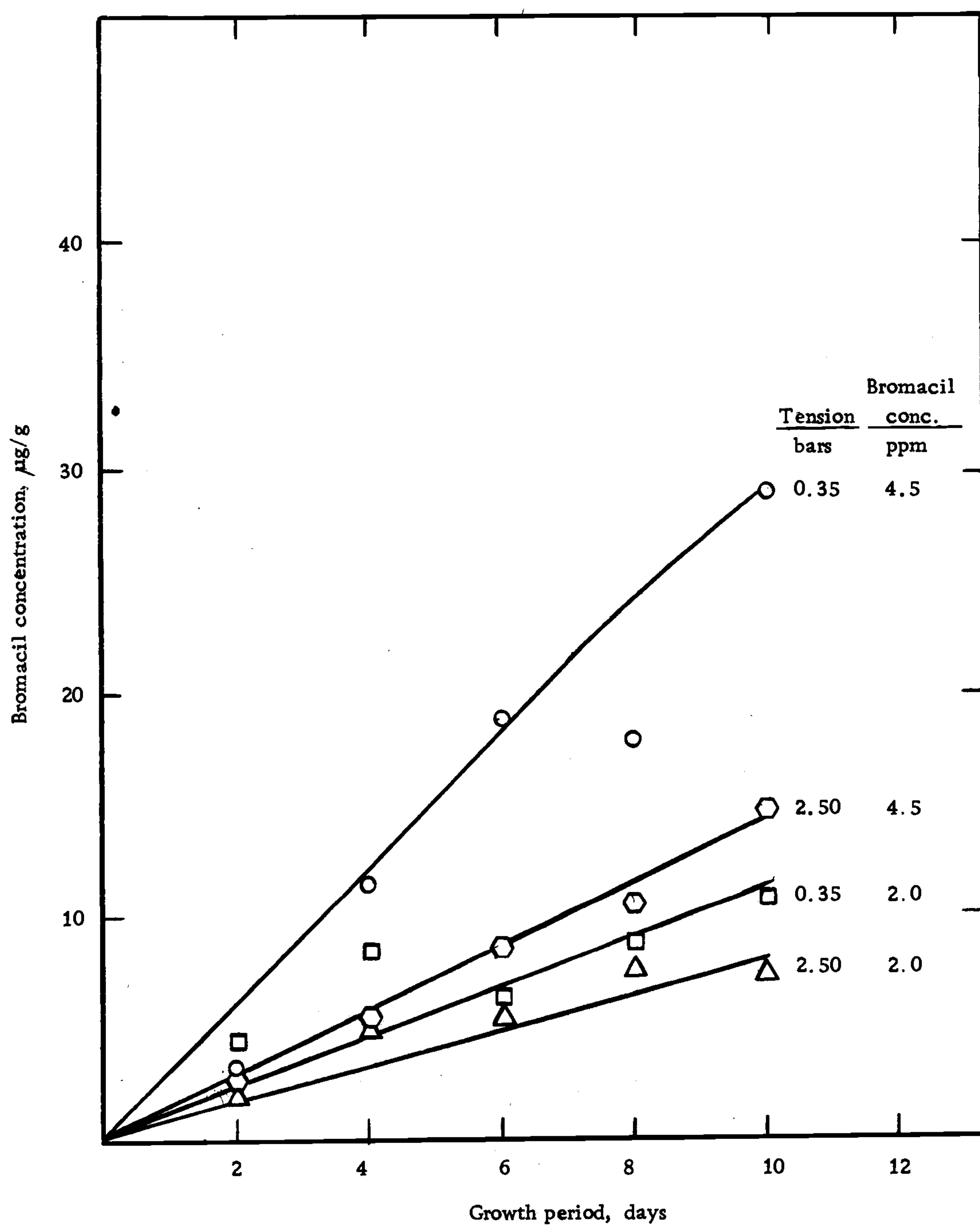


Figure 4. Bromacil concentration on the root tissue of wheat

Table 1. Bromacil distribution in wheat tissue as a function of soil moisture tension and bromacil application rate

Growth period	Moisture tension	Application rate	Bromacil conc. in root and foliar tissue	Bromacil accum. in root and foliar tissue	Bromacil in foliar and total plant tissue at two application rates		Bromacil in total plant tissue at two moisture levels
days	bars		μ/g foliar / μg root	μg foliar / μg root	conc. (foliar)	accum. (total plant)	μg .35 bar / μg 2.5 bar
					μg high rate / μg low rate		
2	0.35	2.0	4.9	15.6	1.2	1.4	2.1
		4.5	8.1	20.0			2.6
2	2.50	2.0	3.9	9.5	1.2	1.1	
		4.5	3.7	8.9			
4	0.35	2.0	4.8	13.6	1.8	2.0	2.2
		4.5	6.3	15.8			2.1
4	2.50	2.0	3.5	7.7	1.3	2.1	
		4.5	6.5	17.0			
6	0.35	2.0	8.8	25.7	1.7	1.5	1.7
		4.5	5.0	13.3			1.6
6	2.50	2.0	5.6	14.5	1.8	1.6	
		4.5	6.3	19.0			
8	0.35	2.0	8.2	20.6	1.7	1.6	1.7
		4.5	6.8	18.3			1.6
8	2.50	2.0	5.2	12.0	1.7	1.6	
		4.5	6.3	20.2			
10	0.35	2.0	7.0	19.7	1.5	1.3	1.7
		4.5	4.0	15.8			1.3
10	2.50	2.0	5.9	15.9	1.7	1.3	
		4.5	4.8	12.7			

Table 2. Ratios of bromacil in the root tissue as a function of soil moisture tension and bromacil application rate

Growth period	Bromacil in root tissue at two moisture tensions				Soil moisture tension	Ratio of bromacil in root tissue at two application rates	
	2.0 ppm		4.5 ppm				
	(conc.)	(accum.)	(conc.)	(accum.)		conc.	accum.
Days	µg .35 bar / µg 2.5 bar		ug .35 bar / ug 2.5 bar		Bars	µg high rate / µg low rate	
2	1.2	1.2	2.0	1.4	0.35	.7	1.1
					2.50	1.2	1.2
4	2.1	2.3	1.6	1.3	0.35	1.4	1.7
					2.50	1.0	1.0
6	2.2	2.2	1.1	1.0	0.35	3.0	2.8
					2.50	1.6	1.3
8	1.7	1.8	1.1	1.0	0.35	2.0	1.8
					2.50	1.4	1.0
10	1.9	1.1	1.5	1.4	0.35	2.7	1.6
					2.50	2.0	2.0

Bromacil uptake into the wheat plants based on a two-day time interval decreased as a function of time at the 0.35 bar tension and low application rate (Table 3). At the 2.5 bar tension the bromacil taken up into the plants at any two-day interval was about the same throughout the experiment. Approximately 13 μg of bromacil were taken up by the plants at the 0.35 bar tension by the end of the second two-day interval, while the plants grown at the 2.5 bar tension did not accumulate this quantity of bromacil until after eight days.

At the high bromacil application rate, more bromacil was accumulated in the wheat tissue after four days at the 0.35 bar moisture level than with any other treatment after ten days (Table 3). The rapid bromacil uptake essentially terminated growth of the plants at the 0.35 bar tension after eight days. As at the low application rate the plants at the 2.5 bar tension assimilated about the same amount of bromacil at each two-day interval.

The fresh weight of the herbicide treated plants, at both bromacil application levels, was substantially lower than the check plants at each corresponding soil moisture tension value after the initial two-day growth period. Fresh weight growth increments (initial plant weight - plant weight at harvest time) at the low application of bromacil indicate a possible stimulation effect by the herbicide during the intitial two-day growth period (Figure 5). The increased growth rates could also represent a water-saving effect brought about

Table 3. Uptake of bromacil by wheat at each two-day interval

Time interval days	Accumulative time days	Moisture tension bars	Application rate µg/ml	Chemical in plant per two day period µg	Water transpired per two day period ml
2	2	0.35	2.0	6.99	69.58
			4.5	9.47	65.35
2	2	2.50	2.0	3.27	56.64
			4.5	3.65	37.72
2	4	0.35	2.0	6.01	48.08
			4.5	16.62	71.28
2	4	2.50	2.0	2.64	49.65
			4.5	8.56	62.87
2	6	0.35	2.0	4.90	45.63
			4.5	0.94	50.29
2	6	2.50	2.0	4.63	50.28
			4.5	4.97	51.60
2	8	0.35	2.0	4.75	34.51
			4.5	9.20	45.21
2	8	2.50	2.0	2.94	46.12
			4.5	4.82	44.72
2	10	0.35	2.0	4.10	22.84
			4.5	--	36.96
2	10	2.50	2.0	2.36	31.98
			4.5	3.26	48.70

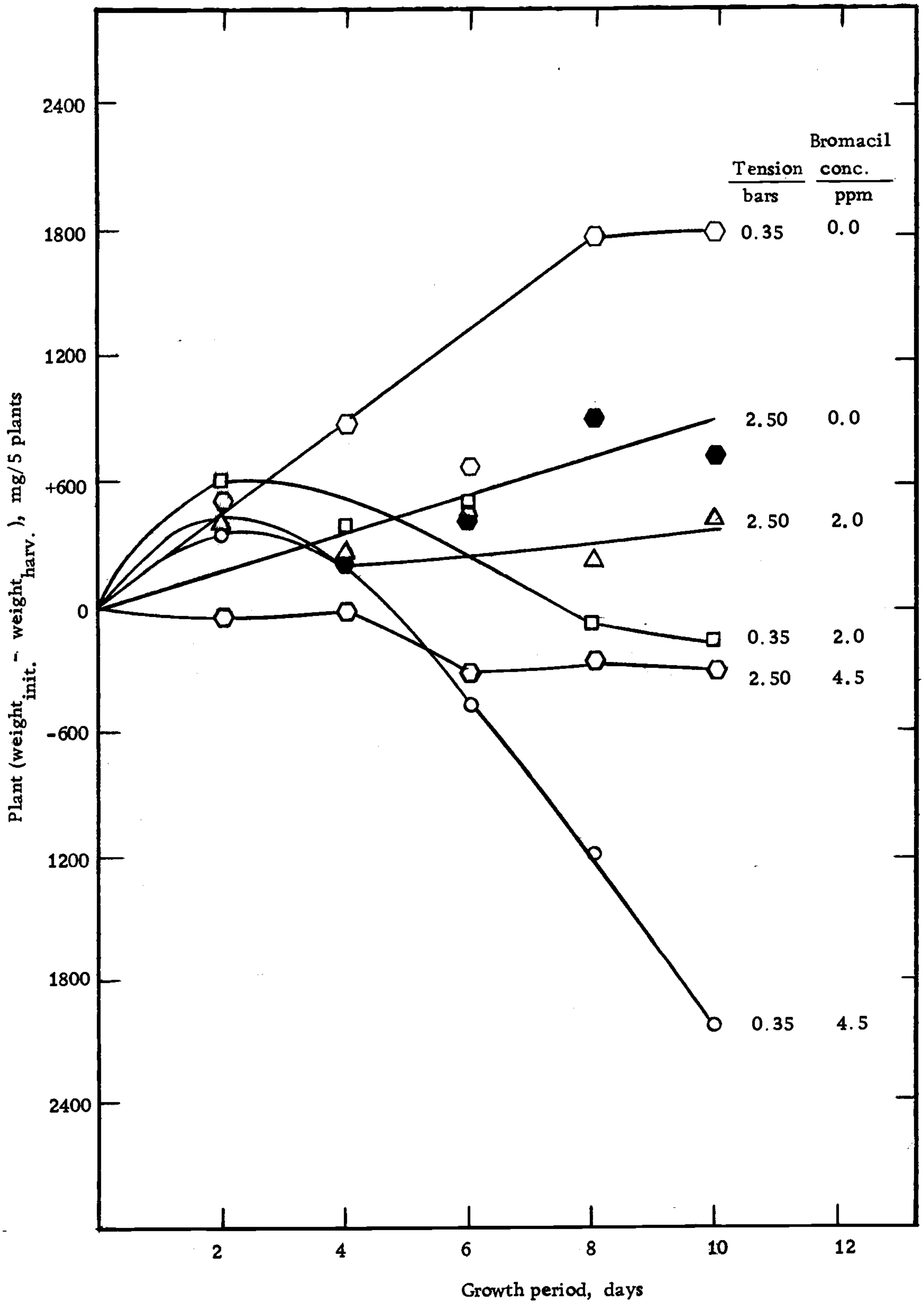


Figure 5. Wheat growth (fresh weight) in Woodburn silt loam treated with 2.0 and 4.5 ppm bromacil

by the reduced transpiration rates observed at both soil moisture tension values. Atrazine applied to soybean and cotton plants initially raised the plant moisture content above that of the non-treated plants (Wills et al., 1963). From the second day until termination of the experiment, the plants grown at the 0.35 bar tension and the 2.0 ppm bromacil application decreased in fresh weight. The plants grown at the 2.5 bar tension decreased slightly in fresh weight from the second to the fourth day, after which there was a very slight increase in the fresh weight of the plants.

At the high bromacil application rate, similar growth patterns were observed as for the low application rate except that no stimulation effect was observed during the first two-day period (Figure 5). At the 0.35 bar tension the loss in plant fresh weight was much more rapid at the high application rate than at the low application rate. The plants grown at the 2.5 bar tension decreased in weight during the first six days after which the growth rate remained constant.

The greater bromacil toxicity to wheat at the 0.35 bar tension at both bromacil application levels can be directly related to the higher rates of transpiration and photosynthesis that not only increased the total uptake of bromacil (Table 3), but also resulted in a quicker expression of bromacil toxicity. Increased rates of photosynthesis and transpiration at lower soil moisture tensions have been demonstrated by Brix (1962) and Babolola (1967).

In general, root weight decreased as a function of time, especially at the high rate of bromacil application (Table 8, appendix). The trend toward a decrease in root weight would reflect a decreased supply of photosynthates available for root growth.

Plant condition at harvest

After ten days the plants exposed to the high bromacil application rate and 0.35 bar tension were extremely flaccid, desiccated, yellow to brown in coloration, and had very narrow leaf blades (Figure 6a). The plants at the 2.5 bar tension were slightly flaccid, light green in color, but no desiccation or narrow leaf blades were observed (Figure 6b). At the low rate of bromacil application the plants at the 0.35 bar tension were somewhat flaccid, yellow to light green in color, slightly desiccated, and had but a few blades that were narrow in width (Figure 6c). Plants grown at the 2.5 bar tension differed only from the check plants in that there was a slight discoloration toward a lighter shade of green (Figure 6d).

Soil moisture

Soil moisture tension had a significant effect on the uptake and subsequent translocation of bromacil in wheat (Figures 1, 2, 3, and 4). Regardless of the stage of growth and the application rate, more bromacil was taken up by the plants at the 0.35 bar tension (Tables

1 and 3).

Soil moisture tension also controlled the accumulation of bromacil in root tissues (Figures 2 and 4). More bromacil was consistently present in the roots at the lower 0.35 bar moisture tension value (Table 2). An increase in soil moisture tension could adversely affect root permeability by inducement of changes in membrane structure, increased suberization of the epidermis, or an increase in cytoplasmic viscosity. A decrease in root permeability could make it more difficult for the bromacil molecule to enter the root and hence limit the total uptake of bromacil into the plant system.

At the low bromacil application rate and 0.35 bar tension, the concentration of bromacil in the transpirational stream increased as a function of time, whereas the concentration of bromacil in the transpirational stream at the 2.5 bar tension remained relatively constant from the second to the tenth day (Figure 7). At no time during the experiment (at both application rates) did the concentration of bromacil in the transpirational stream approach that of the osmotic solutions. These data may be an indication that decreased root permeability at the high soil moisture tension value is limiting the entry of bromacil into the root. During the first two-day growth interval the greater transpiration rate and total quantity of water transpired at the 0.35 bar tension could account for the larger quantity of bromacil in the plant (Table 3). However, after two days the

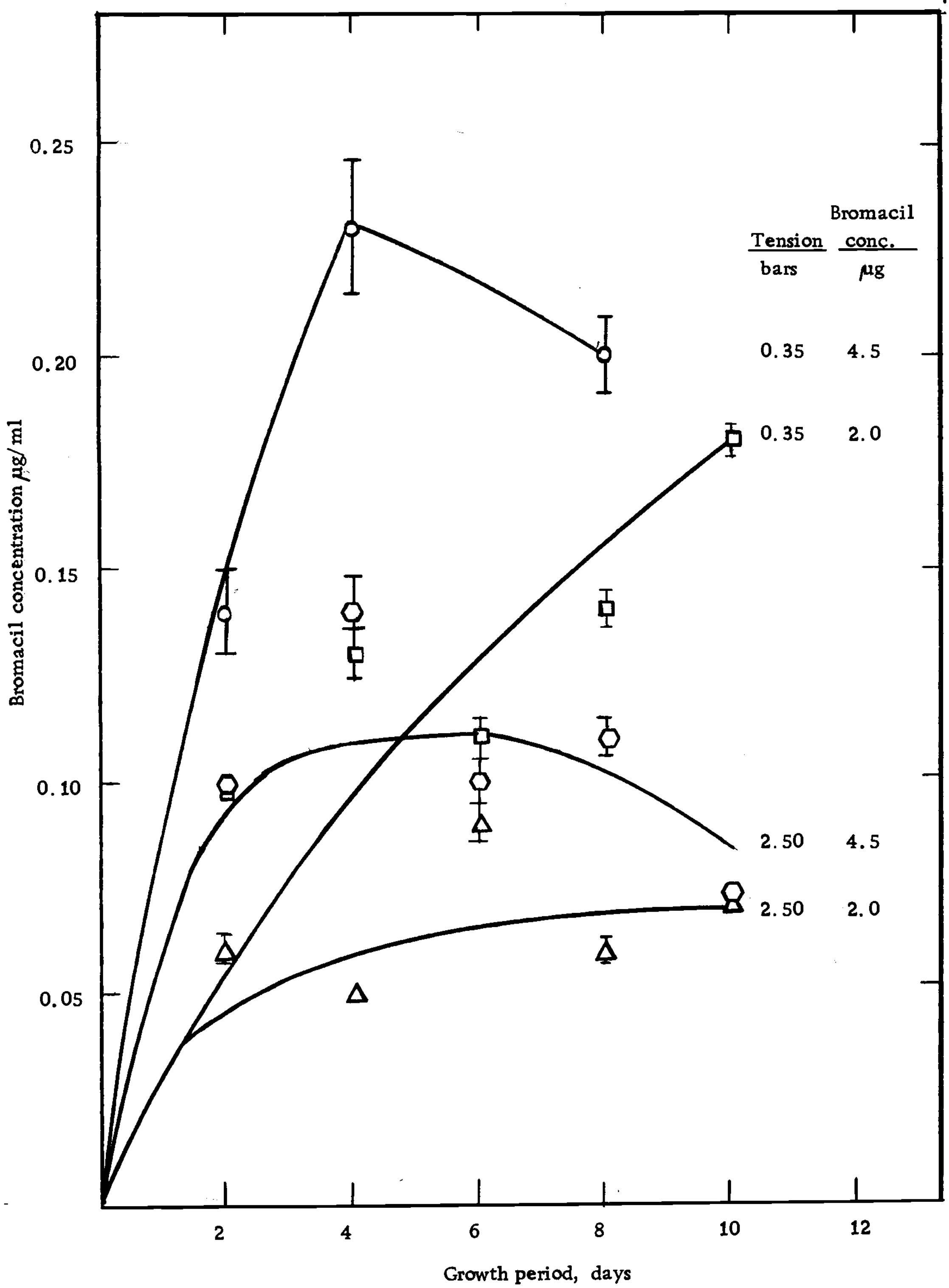


Figure 7. Concentration of bromacil in the transpirational stream

*Vertical line at each data point indicates the relative amount of bromacil taken up during the two-day period transpiration rate and total quantity of water transpired was greater at the 2.5 bar tension than at the 0.35 bar tension, yet, a larger amount of bromacil at each additional two-day interval was taken up by the plants at the lower 0.35 bar tension than at the 2.5 bar tension value. Similar observations, in regard to a root permeability effect, can also be made for the 0.35 bar and 2.5 bar tensions at the high bromacil application level.

The bromacil concentration in the transpiration stream was higher for the 4.5 ppm bromacil application rate at a given moisture stress. Also noted at the high bromacil application rate is the decrease in concentration of bromacil in the transpirational stream after four days at the 0.35 bar tension, and after eight days at the 2.5 bar tension. This decrease in bromacil concentration at the two tensions could be related to the greater physiological damage of the plants at the higher application rate. Decreased rates of transpiration at higher levels of soil moisture stress might also be expected to decrease the uptake of bromacil, assuming that bromacil enters the plant and is transported to the foliar portions via the transpirational stream.

Under normal field conditions the soil water between 0.33 (field capacity) and 15.0 bar tensions (permanent wilting point) is considered available for plant growth. Thus, the moisture tensions used in this experiment were approximately at field capacity and below field capacity. Soil moisture levels higher than the 15 bar permanent wilting point may also limit plant growth.

Bromacil in foliar and root tissues

Bromacil did not accumulate in significant quantities in the root, but rather was translocated quickly via the transpirational stream to the foliar portion of the plant (Table 1). Observed ratios of bromacil in the foliar portion to that present in the root were similar at both the high and low bromacil application rates at a given soil moisture level.

Soil moisture tension influenced the ratio of chemical in the foliar portions to that in the roots. At the low bromacil application rate the ratios of bromacil in the foliar portions to that in the roots were consistently larger at the 0.35 bar tension compared with the ratio values at the 2.5 bar tension. The lower ratios of bromacil in the foliar portions to that in the roots at the higher tension value would indicate that a smaller proportional amount of bromacil was translocated from the roots to the foliar portions of the plant. At the high bromacil application rate the ratio of bromacil in the foliar portions to that in the roots was more erratic and no definite trend could be observed as to the effect of soil moisture tension on the translocation of bromacil.

Transpiration rate and bromacil uptake

Transpiration rates over the ten-day growth period were higher for the check plants at the 0.35 bar tension than at the 2.5 bar tension (Figure 8). Average transpiration rates for the check plants over the ten-day period at the 0.35 bar and 2.5 bar tensions were 3.79 ml/hr/15 plants and 2.28 ml/hr/15 plants respectively.

Bromacil had a marked effect on plant transpiration rates at both soil moisture tensions and bromacil application rates (Figures 9 and 10). Initial plant transpiration rates for the first two to five hours at both the high and low levels of bromacil application were nearly the same as the initial transpiration rate of the check plants at the 0.35 and 2.5 bar moisture tension values. The greatest decrease in transpiration rate at both levels of bromacil application and soil moisture values occurred during the first 40 hours after transpiration rates were initially measured. During this time period, transpiration rates were decreased by 66 and 39% at the 0.35 and 2.5 tensions respectively.

Stomatal closure as a result of an increase in CO_2 concentration due to stoppage of the Hill reaction has been thought to be the primary reason for decreased transpiration rates of plants treated with a Hill reaction herbicide (Smith and Buchholtz, 1962, 1964). Since bromacil does inhibit the Hill reaction, it is postulated that

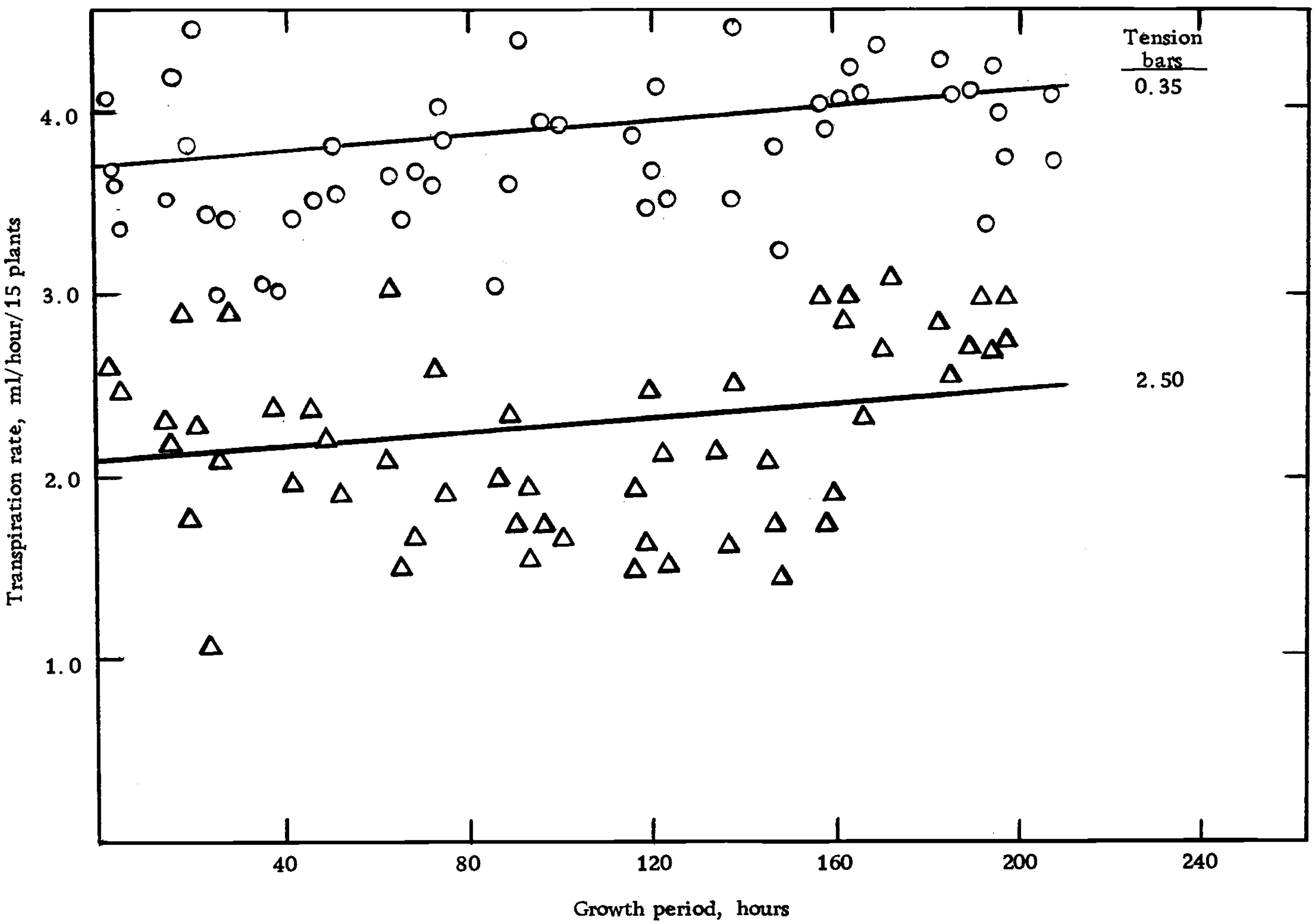


Figure 8. Transpiration rate of wheat at two moisture tensions

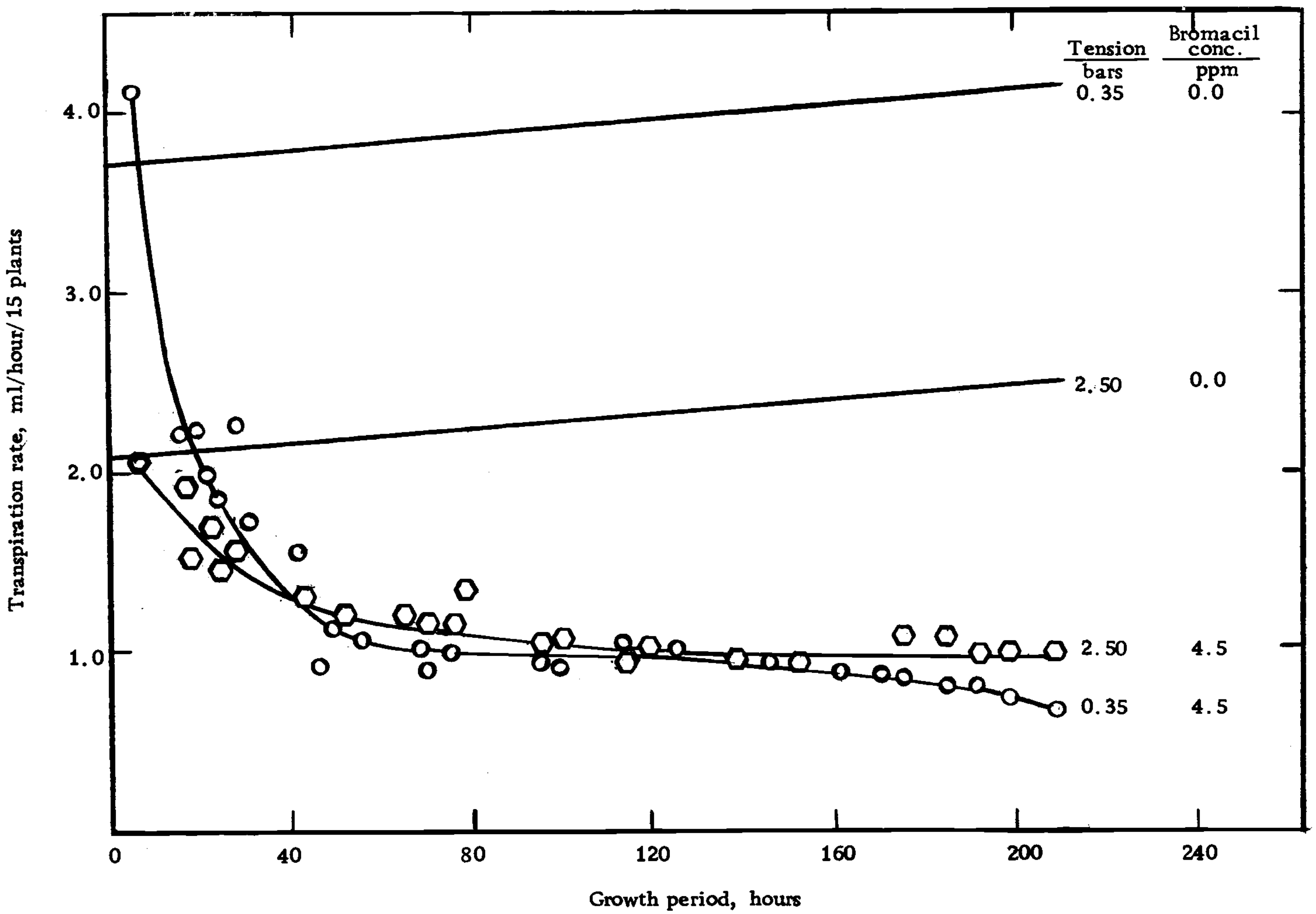


Figure 9. Transpiration rate of wheat at the high bromacil application rate

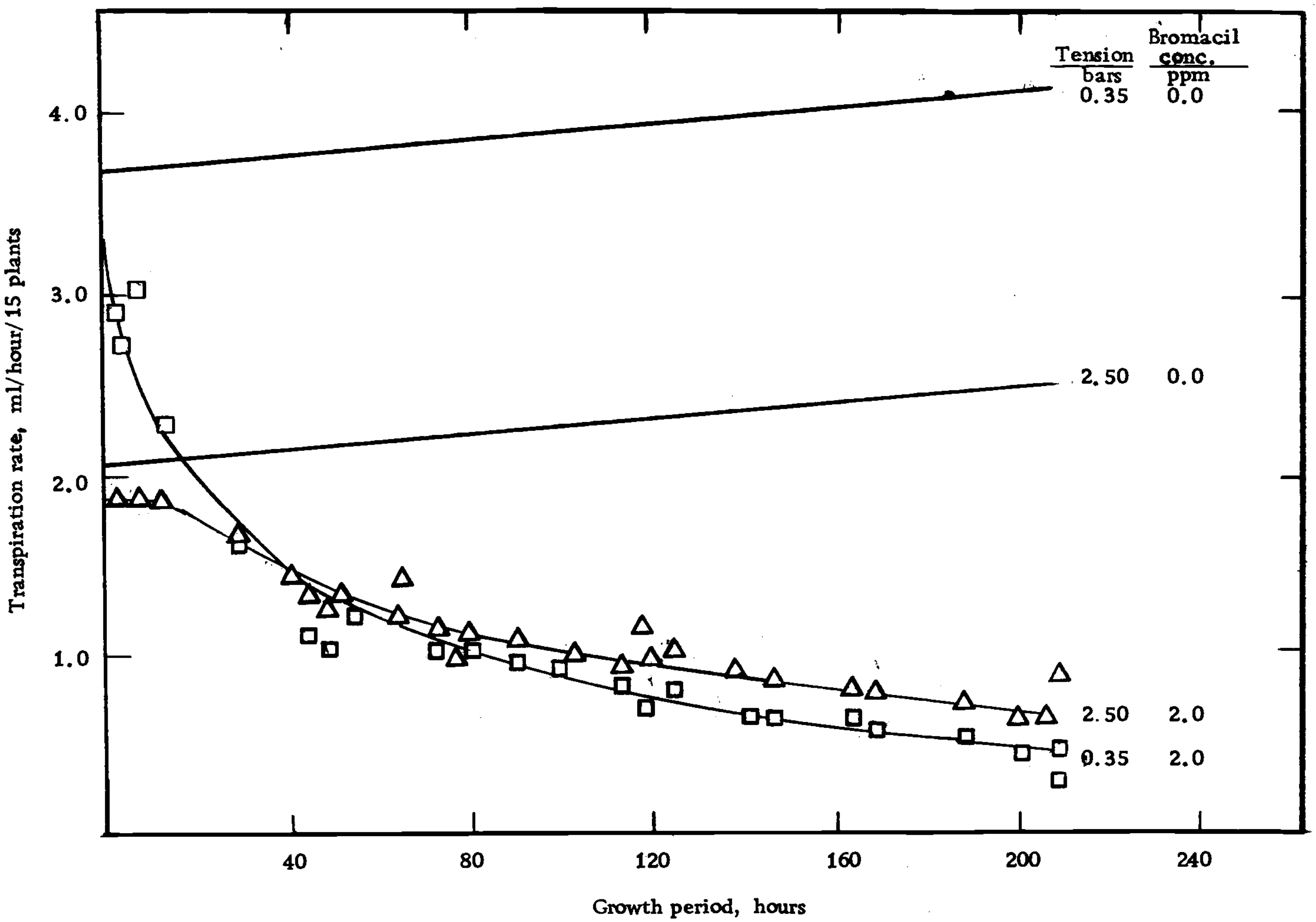


Figure 10. Transpiration rat e of wheat at the low bromacil application rate

the decreased rates of transpiration observed in this experiment were due to stomatal closure resulting from a build up of CO_2 around the guard cells of the stomata. At both the high and low bromacil application levels the transpiration rates at 0.35 bar and 2.5 bar tensions were essentially the same at the end of 50 hours. Although bromacil indirectly reduced transpiration rates, the accumulative uptake of bromacil into the plants increased in direct proportion to the water transpired at the two soil moisture tension values (Figure 11).

In diuron uptake studies with wheat, the decrease in photosynthesis was linear with the accumulative uptake of water at various soil moisture levels (Sedgley and Boersma, 1969), A smaller decrease in the rate of photosynthesis per unit volume of water transpired at the 2.5 bar tension compared to the 0.30 bar tension was considered to result from a decrease in root permeability at the higher soil moisture tensions. Assuming that bromacil moves in wheat similarly to diuron, the data presented by Sedgley and Boersma can be explained by the fact that a larger amount of chemical per unit volume of water transpired is present in the plant tissue at the 0.35 bar tension compared to the 2.5 bar tension (Figure 11), which in turn may be related to root permeability effects on the concentration of bromacil in the transpirational stream (Figure 7). Also, as reported previously, soil moisture may influence the translocation

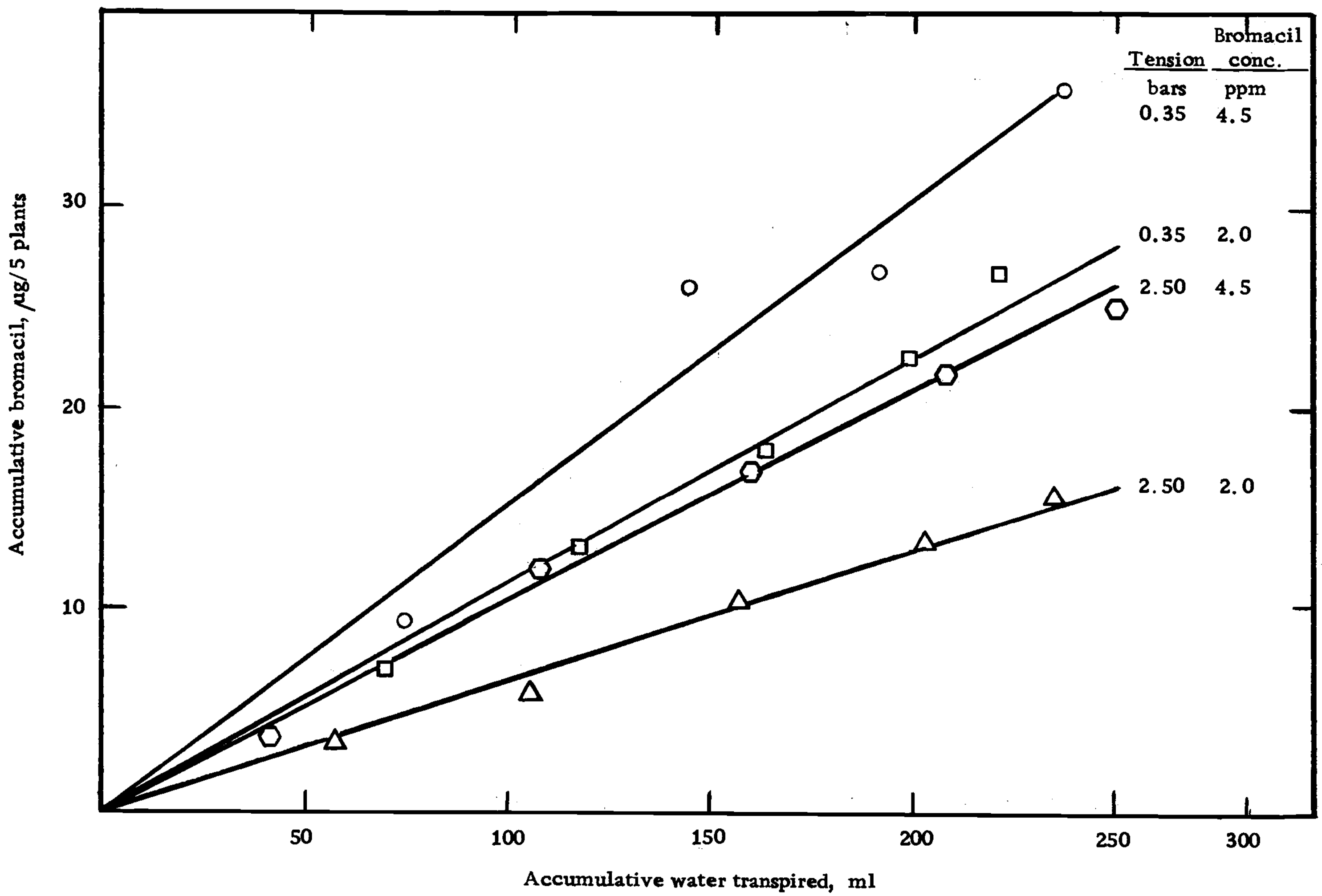


Figure 11. Bromacil accumulation in wheat as a function of transpiration

of chemical from the root to the foliar portions of wheat as well as influence root permeability (Table 1).

Conclusions

Uptake of bromacil by wheat was studied as a function of time, bromacil concentration, and soil moisture stress. Large quantitative differences in the uptake of bromacil at the two soil moisture tensions indicate that soil moisture plays an important role in the uptake of bromacil into the total plant system. At the low and high bromacil applications 70 and 42% more bromacil respectively was taken up at the 0.35 bar tension value as compared to the 2.5 bar tension value.

Soil moisture influences not only the distribution of a herbicide in the soil aqueous phase, but also can regulate important plant physiological functions such as root permeability and transpiration rate which in turn may influence the uptake of a herbicide. Larger values for the ratio of bromacil in the foliar to root portions at the 0.35 bar tension compared to the 2.5 bar tension indicate that translocation of bromacil from roots to the foliar portions may also be regulated by soil moisture stress. At the low and high bromacil applications 25 and 69% more bromacil was taken up by the roots at the low moisture tension than at the high moisture tension. The linear relationship between the accumulative uptake of bromacil and quantity of water transpired would indicate that bromacil uptake is directly proportional to the rate of transpiration.

SUMMARY AND CONCLUSIONS

Soil texture and organic matter had a pronounced effect on both the distribution of bromacil in the soil solution and removal of bromacil from soil over a 0.10 to 15.0 bar moisture tension range. Of the initial amount of bromacil added to each of four soils, 82, 45, 26, and 21% was released by application of pressure to a quartz sand, Ephrata loam, Woodburn silt loam, and Coker clay soil respectively over the 15 bar tension range. The major portion of the bromacil removed from each soil was recovered at moisture tensions less than 0.60 bars. Thus, movement of bromacil with the soil water could be a problem at high soil moisture values.

Over a moisture tension range of 0.30 to 3.0 bars, the concentration of bromacil in the soil solution of each soil was relatively constant. The average bromacil concentration in the soil solution over this tension range was 11, 15, and 26 μg bromacil per ml of effluent for the Coker clay, Woodburn silt loam, and Ephrata loam soils. As the Coker clay contained more than three times the amount of clay present in the Woodburn silt loam, and because of the small difference in the concentration of bromacil in the soil water extracts for the two soils, it appears that organic matter may be the predominant adsorption site for bromacil.

As the distribution of bromacil in the soil solution is affected by soil texture and organic matter, differences in bromacil uptake and phytotoxicity to plants grown in different soils can also be expected. Soil moisture, at least from a bromacil concentration standpoint, should have little effect on bromacil phytotoxicity for the Ephrata loam, Coker clay, and Woodburn silt loam soils over a moisture tension range of 0.30 to 3.0 bars.

Plant bioassay experiments demonstrated that at a 4.0 ppm bromacil application rate, the maximum concentration of bromacil in the plant tissue of plants grown in the Coker clay, Woodburn silt loam, and Ephrata loam soils was 115, 137, and 523 μg of bromacil respectively per gram plant tissue. The differences in bromacil concentration present in the plant tissue for the plants grown in the three soils reflect the bromacil concentration in the soil solution for the Coker clay (11 μg/ml), Woodburn silt loam (15 μg/ml), and the Ephrata loam (26 μg/ml) over the 0.30 to 3.0 bar moisture tension range. At the 4.0 ppm application rate an extrapolation of the data shows that at a common soil moisture of 40%, the herbicide treated plants were 25, 42, and 55% of the weight of the check plants grown in the Ephrata loam, Woodburn silt loam, and Coker clay soils respectively.

Soil moisture variables within each soil type influenced not only the uptake of bromacil, but also the phytotoxicity of bromacil inside the plant system. As soil moisture decreased for the three soils the uptake and concentration of bromacil in the plant tissue increased. A faster termination of plant growth at the higher soil moisture values could have decreased the uptake of bromacil, whereas at the lower soil moisture values plant growth continued as did the assimilation of bromacil. Initial faster rates of transpiration and photosynthesis of the plants at the higher soil moisture values are possible explanations for the increased toxicity of bromacil as soil moisture increased. Also, at the lower soil moisture values, movement of bromacil from the xylem to chloroplasts could have been restricted resulting in a decrease in phytotoxicity.

Soil moisture tension on the plant system greatly influenced the uptake of bromacil into both the foliar and root portions of wheat. At the high and low bromacil application rates 70 and 42% more bromacil respectively was taken up by the plants grown at a 0.35 bar tension as compared to plants grown at a 2.50 bar tension. At the same application rates, the amount of bromacil in the roots was 25 and 69% more at the low moisture tension value than at the high value. While the uptake of bromacil at both moisture tensions increased in direct proportion to the quantity of water transpired, calculation of bromacil concentration in the transpirational stream as a function of time showed a constant bromacil concentration in the transpiration stream at a 2.50 bar tension, whereas there was an increase in bromacil concentration at the 0.35 bar tension. Because the transpiration rates of the bromacil treated plants were essentially the same after 40 hours at both soil moisture tensions, root permeability was cited as a major factor in limiting the uptake of bromacil into the plant system. Also, at the low bromacil application rate, a greater proportion of bromacil was translocated to the foliar portions from the roots at the 0.35 bar tension as compared to the 2.50 bar tension. Thus, the increased toxicity of bromacil at higher soil moisture values can be explained in part by the more rapid uptake of chemical and faster translocation from the root to the foliar tissues. It is suspected that the higher rate of photosynthesis at higher soil moisture tensions may also play a role in the more rapid expression of bromacil phytotoxicity.

In general, these results indicate that for the maximum effectiveness of a soil applied herbicide, specifically bromacil, the soil at application time should be moist rather than dry, and the weeds to be controlled should be under environmental conditions that would allow for a rapid rate of transpiration.

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APPENDIX

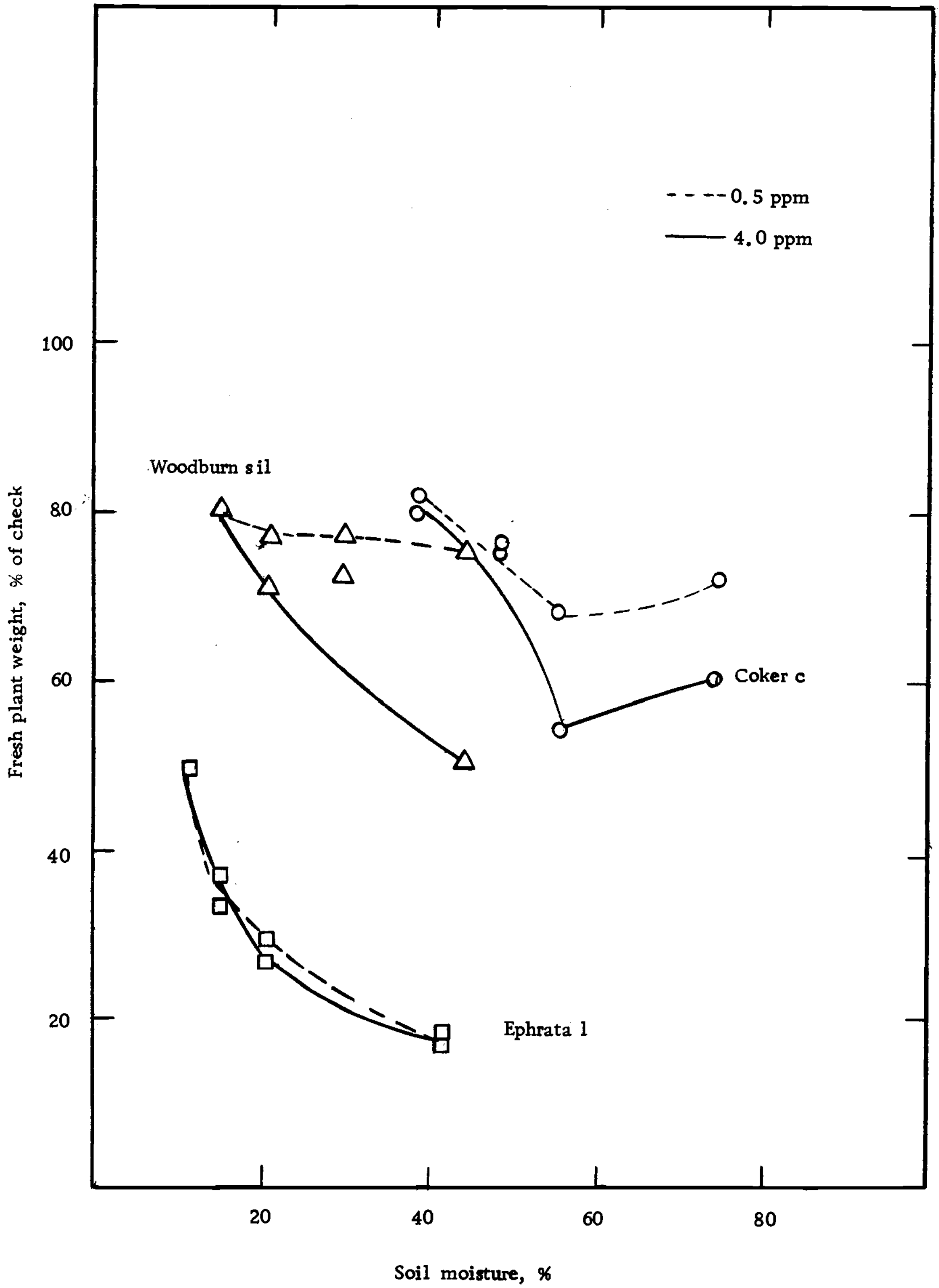

Figure 1. Wheat growth as affected by bromacil and soil moisture

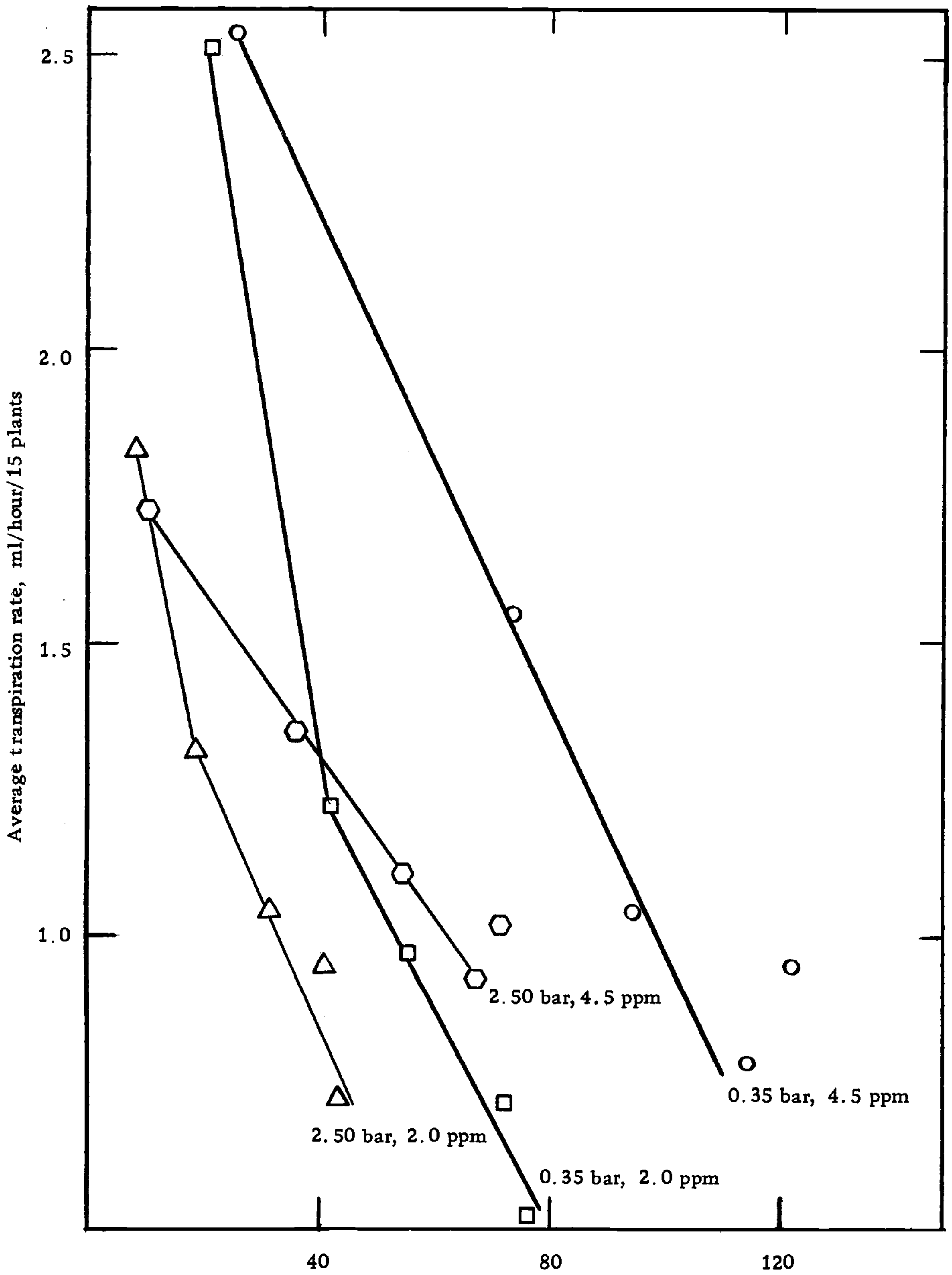

Figure 2. Bromacil in wheat foliar tissue as a function of transpiration

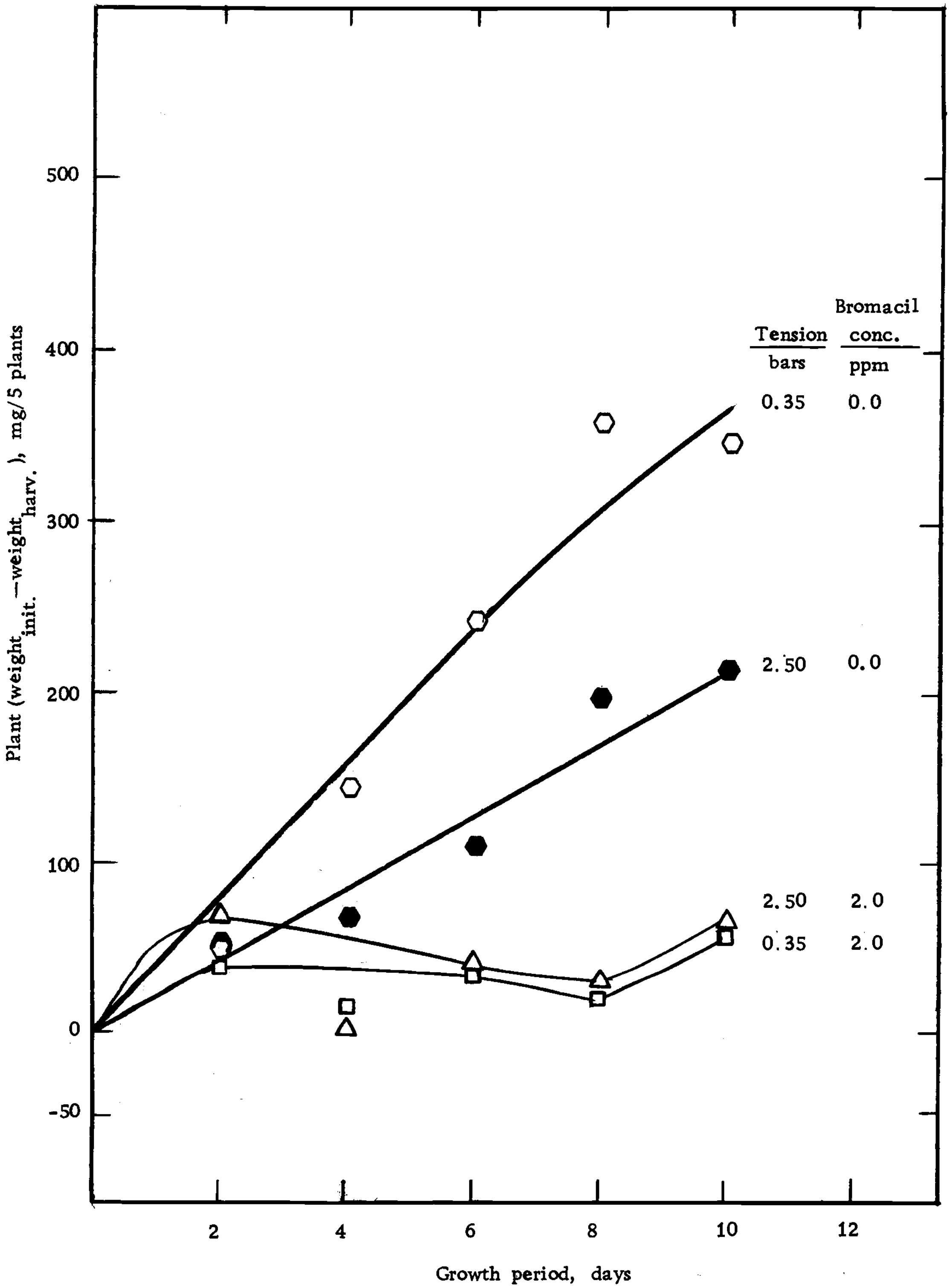

Figure 3. Wheat growth (oven dry weight) in Woodburn sil treated with 2.0 ppm bromacil

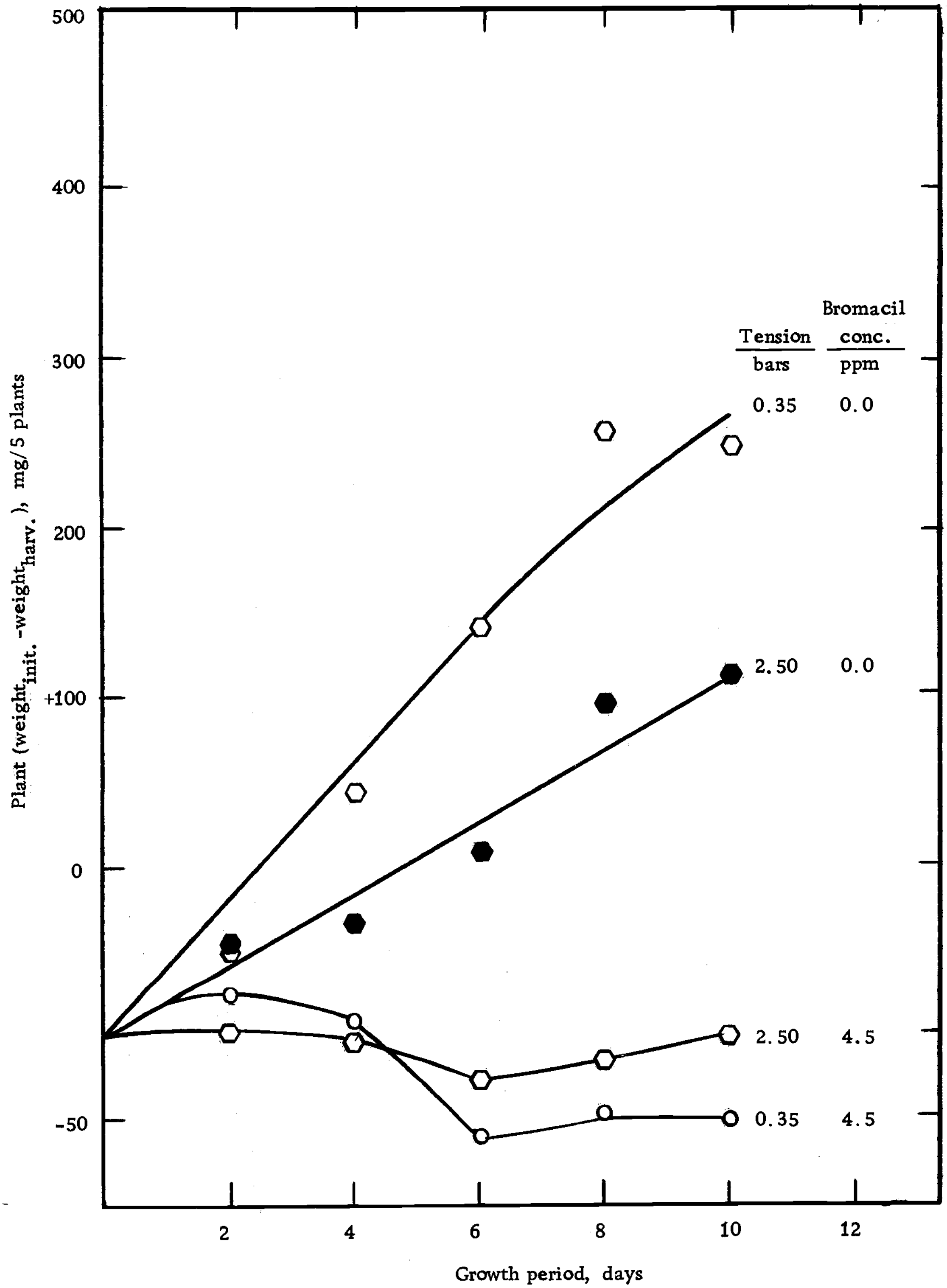

Figure 4. Wheat growth (oven dry weight) in Woodburn silt loam treated with 4.5 ppm bromacil

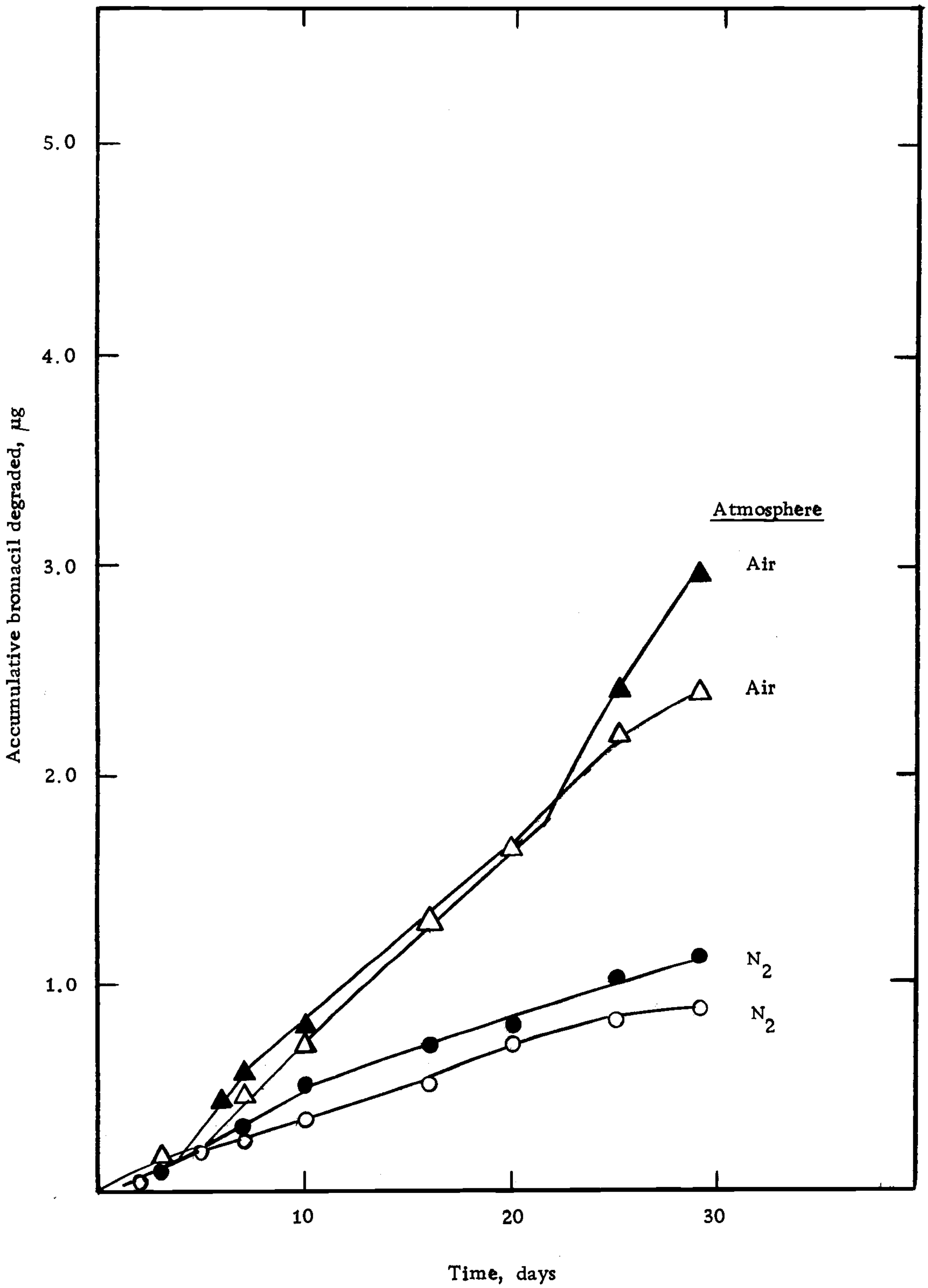

Figure 5. Microbial degradation of bromacil under two atmosphere systems*

*A total of 950 μg bromacil added to each degradation system

Table 1. Volumetric pressure membrane apparatus data for Ephrata loam

Moisture tension bars	*Bromacil conc. µg/ml		Extract collected ml		Accum. extract ml		Bromacil collected µg		Accum. bromacil µg	
	Experiment number									
	1	2	1	2	1	2	1	2	1	2
0.10	0.1	--	0.9	--	0.9	--	0.1	--	0.1	--
0.33	26.9	--	88.5	--	89.4	--	2329.8	--	2329.9	--
0.60	27.3	--	12.0	--	101.4	--	307.0	--	2636.9	--
1.00	26.6	--	7.4	--	108.8	--	168.6	--	2805.5	--
1.50	23.3	--	3.9	--	112.7	--	90.2	--	2895.7	--
2.00	--	25.0	--	120.8	--	120.8	--	3002.6	--	3002.6
6.00	--	24.7	--	6.2	--	127.0	--	148.6	--	3151.2
9.00	--	25.0	--	1.4	--	128.4	--	35.1	--	3186.3

*Concentration of bromacil in the last increment of the indicated tension

Table 2. Volumetric pressure membrane apparatus data for Coker clay

Moisture tension	*Bromacil conc.		Extract collected		Accum. extract		Bromacil collected		Accum. bromacil	
bars	µg/ml		ml		ml		µg		µg	
	Experiment number									
	1	2	1	2	1	2	1	2	1	2
0.10	6.3	--	6.3	--	6.3	--	39.2	--	39.2	--
0.33	11.5	--	97.4	--	103.7	--	929.2	--	968.4	--
0.60	11.1	--	14.5	--	118.2	--	164.3	--	1132.7	--
1.00	11.3	--	10.8	--	129.0	--	120.2	--	1252.9	--
2.00	11.3	10.6	6.9	133.9	135.9	133.9	78.1	1409.4	1331.0	1409.4
3.00	10.2	--	5.0	--	140.9	--	52.2	--	1383.2	--
6.00	--	11.0	--	12.1	--	146.0	--	126.5	--	1535.9
9.00	9.0	9.8	4.2	2.7	145.1	148.7	40.9	26.5	1424.1	1562.4
15.00	--	8.0	--	4.1	--	152.8	--	34.5	--	1596.9

*Concentration of bromacil in the last increment of the indicated tension

Table 3. Volumetric pressure membrane apparatus data for Woodburn silt loam

Moisture tension bars	*Bromacil conc. µg/ml		Extract collected ml		Accum. extract ml		Bromacil collected µg		Accum. bromacil µg	
	Experiment number									
	1	2	1	2	1	2	1	2	1	2
0.10	9.5	--	13.2	--	13.2	--	129.7	--	129.7	--
0.33	14.8	--	55.5	--	68.7	--	748.3	--	878.0	--
0.40	15.2	--	31.2	--	99.9	---	473.0	--	1351.0	--
0.60	15.5	14.6	8.1	92.8	108.0	92.8	121.3	1242.5	1472.3	1242.5
1.00	14.3	--	9.1	--	117.1	--	131.0	--	1603.3	--
1.50	15.4	--	5.4	--	122.5	--	78.9	--	1682.2	--
2.00	16.3	13.7	2.8	19.1	125.3	111.9	43.0	273.1	1725.2	1515.6
3.00	14.2	--	5.1	--	130.4	--	70.6	--	1795.8	--
6.00	--	8.3	--	13.9	--	125.8	--	148.8	--	1664.4
9.00	11.8	5.7	6.3	4.0	136.7	129.8	82.0	25.4	1877.8	1689.8
15.00	9.0	2.9	2.7	10.6	139.4	140.4	24.2	33.7	1902.0	1723.5

*Concentration of bromacil in the last increment of the indicated tension

Table 4. Volumetric pressure membrane apparatus data for quartz sand

Moisture tension bars	*Bromacil conc. µg/ml	Extract collected ml	Accum. extract ml	Bromacil collected µg	Accum. bromacil µg
0.10	44.1	11.5	11.5	456.4	456.4
0.20	48.0	109.7	121.2	5353.7	5810.1
0.60	45.7	1.8	123.0	80.9	5891.0
2.00	41.1	1.6	124.6	66.8	5957.8
15.00	33.4	1.7	126.3	54.9	6012.7

*Concentration of bromacil in the last increment of the indicated tension

Table 5. Wheat growth on several Oregon soils treated with bromacil (plant bioassay study)

Moisture tension	Tissue	Soil								
bars	fresh or oven dry	Coker clay			Woodburn silt loam			Ephrata loam		
		Bromacil application rate ppm								
		0.0	0.5	4.0	0.0	0.5	4.0	0.0	0.5	4.0
		Plant weight g/10 plants								
0.10	Fresh	1.5705	1.1348	0.9488	1.4170	1.060	0.7179	2.7235	0.4658	0.4924
	Oven dry	0.2208	0.1178	0.0992	0.2359	0.1184	0.0981	0.4696	0.1097	0.1156
0.20	Fresh	1.5028	1.0284	0.8129	--	--	--	--	--	--
	Oven dry	0.2599	0.1208	0.1018	--	--	--	--	--	--
0.25	Fresh	--	--	--	--	--	--	1.5960	0.4696	0.4352
	Oven dry	--	--	--	--	--	--	0.2866	0.1066	0.0950
0.30	Fresh	1.1757	0.9041	0.9007	1.1145	0.8599	0.8076	--	--	--
	Oven dry	0.2164	0.1095	0.1066	0.2234	0.1084	0.1011	--	--	--
0.40	Fresh	--	--	--	1.0296	0.7917	0.7302	1.3635	0.4568	0.5096
	Oven dry	--	--	--	0.1656	0.0937	0.0939	0.2517	0.0988	0.1009
1.00	Fresh	--	--	--	--	--	--	1.0228	0.5053	0.5183
	Oven dry	--	--	--	--	--	--	0.1910	0.1048	0.0932
2.50	Fresh	0.9685	0.7939	0.7741	0.9299	0.7475	0.7438	--	--	--
	Oven dry	0.1576	0.0972	0.0922	0.1334	0.0983	0.0844	--	--	--

Table 6. Bromacil present in wheat tissue of plants grown in several Oregon soils (plant bioassay study)

Moisture tension bars	Bromacil application rate ppm	Coker clay conc. µg/g	Coker clay accum. µg/10 plants	Woodburn silt loam conc. µg/g	Woodburn silt loam accum. µg/10 plants	Ephrata loam conc. µg/g	Ephrata loam accum. µg/10 plants
0.10	0.5	13.41	1.58	13.30	1.57	40.15	4.40
	4.0	78.04	7.74	101.48	9.95	315.54	36.48
0.20	0.5	15.35	1.85	--	--	--	--
	4.0	109.20	11.12	--	--	--	--
0.25	0.5	--	--	--	--	55.74	5.61
	4.0	--	--	--	--	523.34	49.72
0.30	0.5	15.36	1.68	14.47	1.57	--	--
	4.0	104.75	11.17	126.38	12.78	--	--
0.40	0.5	--	--	19.12	1.79	59.62	5.89
	4.0	--	--	137.09	12.87	497.66	50.21
1.00	0.5	--	--	--	--	66.02	6.91
	4.0	--	--	--	--	521.84	48.64
2.50	0.5	13.86	1.34	17.85	1.75	--	--
	4.0	114.75	10.58	109.85	9.27	--	--

Table 7. Fresh and oven dry weight of the foliar portion of wheat plants grown in Woodburn silt loam at two osmotic tension values (osmotic chamber studies)

Growth period days	Moisture tension bars	Bromacil application rate ppm 0		2.0 Plant weight g/5 plants		4.5	
		Fresh	Oven dry	Fresh	Oven dry	Fresh	Oven dry
0	0.35	2.2137	0.3066	2.0297	0.2773	2.5262	0.3275
	2.50	2.2137	0.3066	2.0297	0.2773	2.5262	0.3275
2	0.35	2.7058	0.3554	2.6209	0.3169	2.8740	0.3529
	2.50	2.6577	0.3608	2.5401	0.3494	2.4707	0.3301
4	0.35	3.0885	0.4501	2.4145	0.2927	2.7305	0.3371
	2.50	2.4761	0.3745	2.0961	0.2831	2.5047	0.3245
6	0.35	2.8648	0.5487	2.5161	0.3117	2.0574	0.2662
	2.50	2.6214	0.4167	2.4738	0.3183	2.2042	0.3015
8	0.35	3.9859	0.6641	1.9411	0.2992	1.3182	0.2825
	2.50	3.1294	0.5049	2.2724	0.3075	2.2735	0.3126
10	0.35	4.0082	0.6548	1.8727	0.3355	0.4891	0.2767
	2.50	2.9482	0.5229	2.4738	0.3447	2.2217	0.3287

Table 8. Oven dry weight of wheat roots from plants treated with bromacil at two application rates (osmotic chamber studies)

Growth period days	Moisture tension bars	Root weight (oven dry) g/5 plants Bromacil application rate ppm	
		2.0	4.5
2	0.35	0.0975	0.1431
	2.50	0.1424	0.1382
4	0.35	0.1059	0.1332
	2.50	0.1268	0.1247
6	0.35	0.1070	0.1002
	2.50	0.1238	0.1012
8	0.35	0.1184	0.1048
	2.50	0.1345	0.0982
10	0.35	0.1190	0.6960
	2.50	0.1294	0.1242

Table 9. Bromacil accumulation and concentration in wheat foliar and root tissue (osmotic chamber studies)

Growth Period days	Moisture tension bars	Bromacil conc. µg/g, Bromacil application rate ppm: 2.0		4.5		Bromacil accum. µg, Bromacil application rate ppm: 2.0		4.5	
		Root	Foliar	Root	Foliar	Root	Foliar	Root	Foliar
2	0.35	4.26	20.74	3.16	25.56	0.42	6.57	0.45	9.02
	2.50	2.15	8.48	2.65	9.93	0.31	2.96	0.37	3.28
4	0.35	8.36	41.38	11.61	72.80	0.89	12.11	1.55	24.54
	2.50	5.35	18.46	5.43	35.53	0.68	5.23	0.68	11.53
6	0.35	6.27	55.27	18.88	94.43	0.67	17.23	1.89	25.14
	2.50	5.48	30.98	8.57	54.09	0.68	9.86	0.87	16.31
8	0.35	8.83	72.19	17.92	121.61	1.05	21.60	1.88	34.35
	2.50	7.75	40.44	10.64	67.06	1.04	12.44	1.04	20.96
10	0.35	10.83	75.90	28.92	114.55	1.29	25.46	2.01	31.70
	2.50	7.27	43.23	14.89	71.21	0.94	14.90	1.85	23.41