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CIBA-GEIGY CORPORATION
GREENSBORO, NC

UPTAKE AND CHARACTERIZATION OF ϕ -¹⁴C-CGA-48988
AND ITS SOIL METABOLITES IN FIELD ROTATION CORN

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A B S T R A C T

Corn was grown as a rotation crop to white potatoes in a field plot on the CIBA-GEIGY Research Farm at Livingston, New York. The plot was treated by spraying ϕ -¹⁴C-CGA-48988 over-the-top six times at a rate of 0.40 lb. a.i./A and at fourteen day intervals. Chemical names, codes and structures are in Figure 1. The first spraying was 47 weeks prior to planting the corn.

Radioactivity equivalent to ϕ -¹⁴C-CGA-48988 in the 0-3" and 3-6" soil layers decreased from 0.33 ppm to 0.20 ppm in twenty weeks. At the same time radioactivity in the 6-9" layer increased from 0.16 ppm to 0.24 ppm. This increase is not sufficient to account for the loss of radioactivity from the 0-3" and 3-6" layers by leaching. Therefore, the soil metabolites of ϕ -¹⁴C-CGA-48988 are probably being decomposed and the radioactivity lost as ¹⁴CO₂.

The balance data show that radioactivity in the organic fraction decreased from 45.3% at planting to below the level of quantitation at final harvest. During this period, nonextractable radioactivity increased from 41.3% to 86.1%. The radioactivity in the polar fraction remained relatively constant at or below 10%. These data suggest that nonpolar materials, possibly some parent ϕ -¹⁴C-CGA-48988, were adsorbed to soil particles.

The uptake of soil radioactivity by rotation corn was low. All samples at all harvests were less than 0.1 ppm equivalent to ϕ - ^{14}C -CGA-48988. This level is too low for further characterization. The mature grain contained 0.03 ppm, mature cobs contained 0.02 ppm and mature stalks contained 0.06 ppm. Therefore, the rotation corn does not take up inordinate levels of ϕ - ^{14}C -CGA-48988 soil metabolites.

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INTRODUCTION

N-(2,6-dimethylphenyl)-N-(methoxyacetal)-alanine methyl ester*, CGA-48988, is a fungicide proposed for the control of late blight in potatoes. The objectives of this study were to: 1) determine the uptake of the soil degradation products of [U-ring- ^{14}C] N-(2,6-dimethylphenyl)-N-(methoxyacetyl)-alanine methyl ester** in rotation corn, 2) determine the movement and degradation of ϕ - ^{14}C -CGA-48988 in field soil and 3) characterize the radioactive metabolites of ϕ - ^{14}C -CGA-48988 in soil and in corn grown as a rotation crop to potatoes (1).

EXPERIMENTAL

Preparation and Planting of the Field Plot: A 3' x 19' plot was prepared and planted with white potatoes at the CIBA-GEIGY Research Farm at Livingston, New York. The potatoes were sprayed over-the-top six times at fourteen day intervals with ϕ - ^{14}C -CGA-48988 at a rate of 0.40 lb. a.i./A. The first spray treatment was six weeks after plant emergence (7/1/77). The ϕ - ^{14}C -CGA-48988 was dissolved in an ethanol/water (1:1) solution and applied with a miniature boom sprayer. Forty-seven weeks after the first treatment, a 3 x 3' subplot was prepared for rotation corn by tilling to a depth of approximately 8". Seeds were planted at three inch intervals in a single row.

Radioactive Dose: A total of 1,755 mg of ϕ - ^{14}C -CGA-48988 was applied to the plot (sp. act. = 30 $\mu\text{Ci}/\text{mg}$ or 9.01 mCi/mM). No additional CGA-48988 or radioactive chemicals were applied for the duration of the studies.

Sampling: Planting and sampling dates are given in Table I. Monthly rainfall data are in Table II. Corn was sampled 5, 9, 14 and 21 weeks after planting. Soil was sampled at planting and at each corn sampling. Soil cores were divided into 0-3", 3-6", and 6-9" segments for analysis.

* Chemical names and structures are given in Figure 1.

**Hereafter referred to as ϕ - ^{14}C -CGA-48988.

Sample Preparation and Analysis: Plants were homogenized with dry ice in a Wiley Mill (2) and 150-200 mg samples were combusted in a Harvey Oxidizer (3). Biphasic extractions were in accordance with AG-214 to produce organic, polar and nonextractable fractions (4).

Soil samples of approximately two grams each were combusted in a Harvey Oxidizer (3). Extractions were in accordance with AG-254 (5).

Samples were analyzed upon arrival.

Radioactivity Measurements: Radioassays were done in a Beckman LS-255 or Searle Mark III liquid scintillation counter. Efficiencies were obtained by external standardization. Limits of detection and quantitation were determined in accordance with AG-276 (6).

RESULTS AND DISCUSSION

Soil: The levels of radioactivity equivalent to ϕ - ^{14}C -CGA-48988 in field soil (Table V) are shown in Table III. The level in the 0-3" and 3-6" layers showed a drop from approximately 0.33 ppm at the time of planting to 0.20 ppm twenty-one weeks later. The radioactivity in the 6-9" layer showed a slight decrease, from 0.18 ppm to 0.13 ppm during the course of the study. Based on prior data (7) and these soil data, the decrease in radioactivity in the top two layers was probably due to degradation of the ϕ - ^{14}C -CGA-48988 ring.

The balance data show a trend of decreasing radioactivity in the organic fraction with a concomitant increase of radioactivity in the nonextractable fraction. The radioactivity in the organic fraction decreased from 45.3% to below the level of quantitation during the twenty-one week study. The nonextractable radioactivity increased from 41.3% to 86.1% during this time. However, there was no accumulation of radioactivity in the polar fraction which was generally less than 10% of the total. These data indicate that organic materials, possibly some parent ϕ - ^{14}C -CGA-48988, were adsorbed to soil particles.

Corn: The levels of radioactivity in rotation corn equivalent to ϕ - ^{14}C -CGA-48988 are shown in Table IV. The radioactivity at all samplings and all plant parts is 0.06 ppm or less. Corn at fourteen weeks contained only 0.05 ppm equivalent to ϕ - ^{14}C -CGA-48988 and mature grain and stover contained 0.03 and 0.06 ppm respectively. This level is too low for further characterization. These data show that rotation corn takes up very little CGA-48988 or its metabolites from soil.

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2. 5.1.2.e Woo, AG-223, "Blending of Soils and Homogenization of Biological Materials for Radioassay and Extraction."
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TABLE III: DISTRIBUTION AND BALANCE OF RADIOACTIVITY EQUIVALENT TO ϕ - ^{14}C -CGA-48988 IN FIELD SOIL

Interval (weeks) ^a	0 (47)			5 (52)			9 (56)			14 (61)			21 (68)		
	0-3	3-6	6-9	0-3	3-6	6-9	0-3	3-6	6-9	0-3	3-6	6-9	0-3	3-6	6-9
Depth (inches)	0-3	3-6	6-9	0-3	3-6	6-9	0-3	3-6	6-9	0-3	3-6	6-9	0-3	3-6	6-9
Total ppm	0.29	0.36	0.19	0.31	0.22	0.14	0.19	0.18	0.17	0.25	0.23	0.22	0.19	0.21	0.13
Balance	(% of Total ^{14}C in 0-3" Layer)														
Organic	45.3			22.7			22.8			12.1			<*7.4	6.4	<*7.4
Polar	10.2			6.1			6.8			7.4			<*2.5	<*3.1	<*5.9
Nonext.	41.3			63.8			62.8			70.9			86.1	86.5	84.0
Total	96.8			92.6			92.4			90.4			86.1	92.9	97.3

^aNumbers in parentheses indicate the elapsed time (weeks) since the first treatment with ϕ - ^{14}C -CGA-48988 (7/1/77).
 <*indicates that the level of radioactivity was below the level of quantitation (6).

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Table IV: UPTAKE AND BALANCE OF RADIOACTIVITY EQUIVALENT
TO ϕ - 14 C-CGA-48988 IN ROTATION CORN

Interval (weeks)	Whole plants			Stalks	Cobs	Grain
	5	9	14			
Total ppm	0.05	0.06	0.05	0.06	0.02	0.03

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TABLE V: CHARACTERISTICS OF FIELD PLOT SOIL

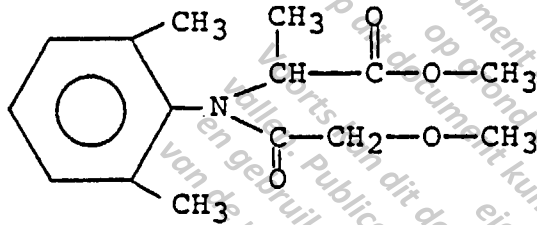
Location: CIBA-GEIGY New York Research Farm,
Livingston, New York

Texture	Silt	Loam
pH	5.5	
% Organic Matter	1.8	
% Sand	44.4	
% Silt	44.0	
% Clay	11.6	

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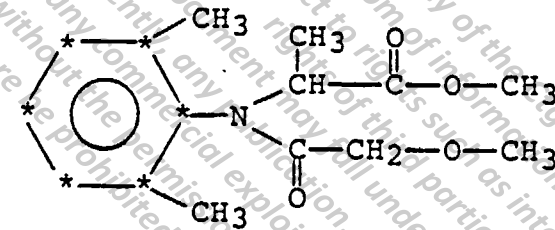
CGA-48988



N-(2,6-dimethylphenyl)-N-(methoxyacetyl)-alanine methyl ester

Radioactive Compound

ϕ - ^{14}C -CGA-48988

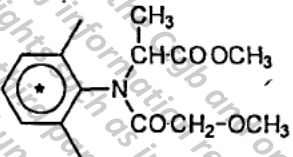


[U-ring- ^{14}C] N-(2,6-dimethylphenyl)-N-(methoxyacetyl)-alanine methyl ester

* = ^{14}C

Figure 1: CHEMICAL NAMES AND STRUCTURES

Annex II - 6.2. /07 : Uptake and characterization of Φ - ^{14}C -CGA 48988 and its soil metabolites in field rotation corn

General Information	
Title of the study:	Uptake and characterization of Φ - ^{14}C -CGA 48988 and its soil metabolites in field rotation corn
Report and/or project number:	ABR-79004
Author:	5.1.2.e/Woc
Ciba File Number (Desire):	48988/3566
Name and address of testing facility:	Ciba-Geigy Corp., Livingston, NY, USA (Biological phase) Ciba-Geigy Corp., Greensboro, N.C., USA (Analytical phase)
Study period:	5/78 - 10/78
Date of report:	January 2, 1979
Compliance with GLP:	Yes [] No, but complies with sound scientific principles [X]
Test guideline(s) used:	-
Deviations from the test guideline:	-
Test substance	
Test substance (code number):	CGA 48988
Batch:	-
^{14}C -labeled test substance :	Yes [X] No []
Specific activity of [U- ^{14}C -phenyl] label:	1.11 MBq/mg (= 30 $\mu\text{Ci}/\text{mg}$)
Radiochemical purity of test substance:	not available
Structural formula: (Position of label)	[U- ^{14}C -phenyl]-CGA 48988 $\ast = ^{14}\text{C}$ 
Formulation used for study:	no
Test system	
Target crop:	field grown potatoes
Formulation (spray application): Formulation N° (spray application): Solvent for application (if used):	ethanol/water (1:1) solution
Application: Field experiment:	Spray applications with a miniature boom sprayer: 6 over-the-top sprays (starting 6 weeks after plant emergence) at 14 days intervals at a rate of 0.40 lb./A (= 292.3 mg ^{14}C -CGA 48988 / 3' X 19' plot/ treatment (= 8.77 mCi) i.e. 1755 mg ^{14}C -CGA 48988/ 6 treatments (= 52.65 mCi for all 6 applications)
Rotational crop (planting / harvest):	corn planting: 47 weeks after the first treatment of target potatoes harvest: 21 weeks after planting the soybeans or 68 weeks after the first treatment of target potatoes

Soil:	Soil from Livingston, NY, USA
	Texture: Silt Loam
	pH: 5.5
	% Organic Matter: 1.8
	% Sand: 44.4
	% Silt: 44.0
	% Clay: 11.6

Summary of findings

Corn was grown as a rotation crop to white potatoes in a field plot on the CIBA-GEIGY Research Farm at Livingston, New York. The plot was treated by spraying Φ -¹⁴C-CGA 48988 over-the-top six times at a rate of 0.40 lb. a.i./A and at fourteen-day intervals. The first spraying was 47 weeks prior to planting the corn.

Radioactivity equivalent to Φ -¹⁴C-CGA 48988 in the 0 - 3" and 3 - 6" soil layers decreased from 0.33 ppm to 0.20 ppm in twenty weeks. At the same time radioactivity in the 6 - 9" layer increased from 0.16 ppm to 0.24 ppm. This increase is not sufficient to account for the loss of radioactivity from the 0 - 3" and 3 - 6" layers by leaching. Therefore, the soil metabolites of Φ -¹⁴C-CGA 48988 are probably being decomposed and the radioactivity lost as ¹⁴CO₂.

The balance data show that radioactivity in the organic fraction decreased from 45.3% at planting to below the level of quantitation at final harvest. During this period, nonextractable radioactivity increased from 41.3% to 86.1%. The radioactivity in the polar fraction remained relatively constant at or below 10%. These data suggest that nonpolar materials, possibly some parent Φ -¹⁴C-CGA 48988, were adsorbed to soil particles.

The uptake of soil radioactivity by rotation corn was low. All samples at all harvests were less than 0.1 ppm equivalent to Φ -¹⁴C-CGA 48988. This level is too low for further characterization. The mature grain contained 0.03 ppm, mature cobs contained 0.02 ppm and mature stalks contained 0.06 ppm. Therefore, the corn does not take up inordinate levels of Φ -¹⁴C-CGA 48988 soil metabolites.

Tab 1 Uptake, distribution and balance of radioactivity equivalent to Φ -¹⁴C-CGA 48988 in rotation corn and soil (at harvest)

Plant part Soil Layer	Total Residues [ppm]	Organic Phase	Water Phase	Non extractable	Total
Stalks	0.06				
Cobs	0.02				
Grain	0.03				
0 - 3"	0.19	<*7.4	<*2.5	86.1	86.1
3 - 6"	0.21	6.4	<*3.1	86.5	92.6
6 - 9"	0.13	<*7.4	<*5.9	84.0	97.3

a <* indicates that the level of radioactivity is detectable but below the level of quantitation

PP 2.52/ JK, 11.3.94