I. Introduction

This report describes the results of the analytical phase of the field dissipation study of Ridomil® 5G on bare ground conducted at a test site in Hollandale, Minnesota during 1987-1988. This study was conducted to satisfy the registration guideline requirements specified in subdivision N, 164-1 of FIFRA. The biology phase was conducted by Landis Associates, Inc., Valdosta, Georgia. Details of the biology phase of the study are provided by Landis Associates, Inc., Valdosta, Georgia.

Soil samples from the test site at depths of 0-6", 6-12", 12-18", 18-24", 24-36" and 36-48" were processed and analyzed by Tegeris Laboratories for metalaxyl (CGA-48988) and the major metabolite CGA-62826. The stuctures and nomenclature for metalaxyl and CGA-62826 are shown in Figure 1. The analyses were conducted using an analytical protocol developed by Tegeris Laboratories. Details of this protocol as well as a detailed description of the analytical method is provided in the Appendix of this report.

This report provides the results of the analyses of soil samples received thus far, namely for 364 days. Any additional analyses for later intervals, if necessary, will be provided as an addendum at a later date.

II. Materials and Procedures

A. Standards

CGA-48988, Lot # S85-0831, purity: 96.5%, received 5/28/87
CGA-62826, Lot # S85-0650, purity: 98.1%, received 5/28/87

B. Reagents

1. Alumina, Basic (Woelm) W200, activity Grade I (prepared by the addition of the 76 ml of water to 324g of Activity Grade Type I Alumina)
2. Diazomethane, ethyl ether solution, prepared according to Organic Syntheses, Coll. Vol. IV, 250 (1963).
3. Ethyl ether, anhydrous, reagent grade
4. Acetone, residue grade
5. Hexane, residue grade
6. Dichloromethane, residue grade
7. 7% (V/V) ethyl ether in hexane
8. 70% (V/V) ethyl ether in hexane
9. 1N Hydrochloric acid, reagent grade
10. 1N Sodium hydroxide, reagent grade
11. Methanol, residue grade
12. 50% (V/V) Methanol in water
13. Isooctane, residue grade

C. Apparatus

1. Centrifuge bottle, 250 ml, polyethylene
2. Flask, round bottom, 250 ml
3. Separatory funnel, 500 ml
4. Filter paper, Whatman 1
5. Centrifuge, with head to accomodate 250 ml centrifuge bottles
6. Rotary evaporator, Buchi, or equivalent
7. Mechanical shaker
8. Disposable chromatographic columns, 8 ml
9. Vacuum manifold to accept disposable chromatographic columns
10. N-evap, Organamation or equivalent
11. Centrifuge tubes, graduated, 15 ml

D. Analytical Method

1. Principle

Residues of CGA-48988 and CGA-62826 were extracted from soil with 50% aqueous methanol. An aliquot of the extract was diluted with water and basified with sodium hydroxide. Residues of the parent CGA-48988 were partitioned into dichloromethane. If necessary, the dichloromethane extract was cleaned up by column chromatography on a basic alumina column and an appropriate aliquot was injected into a gas chromatograph equipped with a nitrogen-phosphorus detector. Residues of the acid metabolite, CGA-62826, were extracted from the initial alkaline aqueous solution by acidification with hydrochloric acid and partitioning with dichloromethane. After evaporation of the solvent, the CGA-62826 was reacted with diazomethane to form the methyl derivative of CGA-62826. The derivative, present as the parent CGA-48988, was cleaned up by column chromatography (if necessary) and analyzed by gas chromatography under the same conditions as the original CGA-48988. A flow diagram of the procedure is depicted in Figure 2.

2. Procedure

a. Extraction
A 50 g soil was extracted using 200 ml of 50% aqueous methanol on a mechanical shaker for 90 minutes. After centrifugation for 20 minutes at 2500 ppm, the supernatant was filtered and basified to pH 10 with 12N NaOH.

b. Partition of CGA-48988
The aqueous solution was partitioned with three 50 ml portions of dichloromethane (DCM) which were filtered through a cotton plug and sodium sulfate into a 250 ml round bottom flask. The aqueous layer was saved for CGA-62826 analysis. The combined DCM extract was taken to

dryness on a rotary evaporator with a water bath at 35°-40°C. The residue was transferred with two 3 ml portions of hexane to either an alumina cleanup column (see below) or to a graduated centrifuge tube for GC analysis.

c. Cleanup of CGA-48988 (if necessary)
If gas chromatographic analysis indicated that additional cleanup were necessary, the extract from the partion step was subjected to column chromatography. A cleanup column was prepared adding 3.0 g of 16% basic Super I Alumina to a disposable 8 ml chromatographic column. The column was placed on a vacuum manifold and 10 ml of hexane was passed through the column. The two 3 ml portions of hexane extract from the partitioning step were transferred to the column. Five ml of 7% ether was then passed through column and discarded. The CGA-48988 was then eluted with 10 ml of 70% ether in hexane. The eluate was transferred to a graduated centrifuge tube for GC analysis.

d. Partitioning of CGA-62826
The aqueous extract of the DCM partitioning step was acidified to pH 3.0 with 1N HCl and extracted with three 50 ml portions of DCM. The combined DCM extracts were filtered through a cotton plug into a 250 ml round bottom flask and taken to dryness on a rotary evaporator at 35°-40°C.

e. Derivatization of CGA-62826
Five ml of methanol and approximately 5 ml of diazomethane ethyl ether solution was added to the residue and the solution allowed to stand for at least 20 minutes with occasional swirling. The diazomethane reagent was added to maintain a yellow color indicating excess diazomethane reagent. The solution was evaporated to dryness on a rotary evaporator at 35°-40°C. The residue was then dissolved in two 3 ml portions of hexane which were then subjected to column cleanup as described in 2.c. or transferred to a graduated centrifuge tube.

f. Gas chromatographic analysis
The extract contained in the graduated centrifuge tube from one of the above steps was evaporated to dryness on an N-evap under a stream of nitrogen. After the residue was reconstituted with isooctane to the appropriate volume, usually 1.5 ml, the solution was subjected to gas chromatography using one of the following systems:

(1) Instrument: Tracor 560 NPD
Packing: 3% OV/17 on Chromosorb W-HP 80/100
Length: 6 ft x 4 mm
Temp:
Column: 205°C
Injector: 250°C
Detector: 250°C

Gas Flow: He: 65 ml/min
Air: 125 ml/min
H_2 6.8 ml/min

Attn: 01
Min. Det. Sens: 0.5 ng

(2) Instrument: Shimadzu GC-9A NPD
Packing: 3% SP 2250 on Supelcoport 100/120
Length: 2.6m x 1.6 mm
Temp: Column: 210°C
Injector: 250°C
Detector: 250°C

Gas Flow: He: 37 ml/min
H_2: 2.7 ml/min
Air: 135 ml/min

Attn: 1
Min. Det. Sens. 0.5 ng

(3) Instrument: Hewlett Packard 5890A with NPD
Column: Methyl Silicone, 530 u ID
Length: 5 m x 530 u
Temp: Column: 165°C
Injector: 180°C
Detector: 220°C

Gas Flow: He: 20 ml/min
Air: 100 ml/min
H_2: 3.5 ml/min

Attn: 0
Min. Det. Sens: 0.5 ng

E. Calculations

Standards were injected into the gas chromatograph and peak heights obtained from a Hewlett Packard 3390A, Shimadzu C-R6A or Hewlett Packard 3393A integrator. From the amount the standard injected and the corresponding peak heights, a linear regression analysis was made to determine the amounts of CGA-48988 and CGA-62826 (as CGA-48988). The concentration in parts per million (ppm) of CGA-48988 and CGA-62826 were calculated as follows:

$$\text{ppm (dry)} = \frac{\text{amount residue found(ng)}}{\text{mg soil injected}} \times \frac{100}{100\text{-M}} \times \frac{100}{100\text{-R}}$$

where M is the moisture remaining after air drying and R is average recovery for the recovery samples. If R was greater than 100, R was taken as 100. That is, the procedural recovery was only used to correct the found values for recoveries less than 100%.

Since residues of CGA-62826 were determined as CGA-48988 in both samples and standards no stiochiometric correction feature was required to convert residues determined as CGA-62826 to equivalents of CGA-48988.

The concentration of CGA-48988 or CGA-62826 as received (or wet basis) was determined by the following equation:

$$\text{ppm (as received)} = \text{ppm(dry)} \times \frac{100\text{-M'}}{100}$$

where M' is the moisture as received.

Figure 2. Flow Diagram of the Analytical Procedure for the Determination of CGA-48988 and CGA-62826 in Soil