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DATA EVALUATION RECORD

STUDY 4

PC No. 600074

Triazole

(degradate of Tebuconazole (PC No. 128997))

§162-1

CAS No. 107534-96-3

DP Barcode D271910

FORMULATION-00-ACTIVE INGREDIENT

STUDY ID 45284038

Scholz, K. 1990. Metabolism of [3,5-<sup>14</sup>C]1,2,4-triazole in soil under aerobic conditions. Performing Laboratory ID PF 3364. Bayer Report No. 110153. Unpublished study performed by Bayer AG, D-5090 Leverkusen - Bayerwerk, Germany; and submitted by Bayer Corporation, Kansas City, MO.

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CONCLUSIONS

Metabolism - Aerobic Soil

1. This study is scientifically valid and provides supplemental information on the aerobic soil metabolism of 1,2,3-triazole.
2. In series of seven experiments (Experiments I, II, and IV conducted on Burscheid I silt loam; Experiment III on Burscheid II silt loam; and Experiments V-VII Leifers silt loam) it was found that triazole degradation in soil was microbial in nature, based on high rates of mineralization (<sup>14</sup>CO<sub>2</sub> = 56-70% of the applied at 360 days) in low-dosed (0.9 µg/100 g

soil) nonsterile silt loam soils and the lack of mineralization in similarly-treated sterile silt loam soil. Rates of mineralization were slow ( $^{14}\text{CO}_2 \leq 4.5\%$  of the applied at 360 days) in high-dosed (ca. 5 mg/100 g soil) nonsterile silt loam soils. Three 1,2,4-triazole degradates: hydroxytriazole (3-hydroxy-1,2,4-triazole), triazolyl alanine (1,2,4-triazole-1-alanine); and triazolyl acetic acid; were identified. All test data were not sufficient for calculation 1,2,3-triazole soil metabolism half-lives.

3. These studies did not follow Subdivision N Guidelines requirements for aerobic soil metabolism for the following reasons:
  - A. zero-time soil samples were not collected;
  - B. only German and Italian soils were tested;
  - C. the soils were not classified according to the USDA Soil Textural Classification System; and
  - C. the soil moisture was maintained at 40% maximum water capacity;
  
4. EFED notes that although this aerobic soil metabolism study does not fulfill Subdivision N Guidelines, there is sufficient information available from the other aerobic soil metabolism studies submitted by the registrant that support the results of this study. The registrant conducted four other aerobic soil metabolism studies with variety of experimental settings/conditions. All studies indicate that microbial degradation of 1,2,4-triazole appears to be rather slow process at higher application rates and faster at lower application rates. The half-lives ranged from 22 days to 375 days (Table 1). The aerobic soil metabolism degradation products were hydroxytriazole (3-hydroxy-1,2,4-triazole), triazolyl alanine (1,2,4-triazole-1-alanine), and triazolyl acetic acid (max. 18%; MRID 45297203 (Experiment II)),  $\text{CO}_2$ , and bound residues.

Table 1. Summary information of 1,2,4-Triazole aerobic soil metabolism study.

Treatment Level	Soil type	Half-life ( $T_{1/2}$ ) in days	Reference
ca. 0.06 ppm	German sandy loam	26.5	MRID 45284032
	German loamy sand	46.7	
	German silt loam	22.2	
50 ppm	Soil 2.2	343	MRID 45297203
	Soil 2.3	375	
1 ppm	Silty loam soil	155	MRID 45284027

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One study showed that 1,2,4-triazole residue may accumulate in crops after triazole application on the cropped soil. Total residues in the wheat plants were the maximum of 61.1-61.3% by 60-90 days after soil treatment with triazole (Experiment III, MRID 45297203). Another laboratory experiment showed that 1,2,4-triazole was almost completely metabolized in 14 days via a bacteria culture, *Nocardia coralline* (MRID 45297203), and triazolyl alanine was the principal metabolite isolated from the cultures.

## ABSTRACT

In series of seven experiments (Experiments I, II, and IV conducted on Burscheid I silt loam; Experiment III on Burscheid II silt loam; and Experiments V-VII Leifers silt loam) it was found that triazole degradation in soil was microbial in nature, based on high rates of mineralization ( $^{14}\text{CO}_2 = 56-70\%$  of the applied at 360 days) in low-dosed ( $0.9 \mu\text{g}/100 \text{ g soil}$ ) nonsterile silt loam soils and the lack of mineralization in similarly-treated sterile silt loam soil. Rates of mineralization were slow ( $^{14}\text{CO}_2 \leq 4.5\%$  of the applied at 360 days) in high-dosed (ca.  $5 \text{ mg}/100 \text{ g soil}$ ) nonsterile silt loam soils. Three 1,2,4-triazole degradates: hydroxytriazole (3-hydroxy-1,2,4-triazole), triazolyl alanine (1,2,4-triazole-1-alanine), and triazolyl acetic acid, were identified. All test data were not sufficient for calculation 1,2,3-triazole soil metabolism half-lives. The detail observations in each of the tests were as follows:

- A. **Burscheid I silt loam soil, low dose.** In **nonsterile** Burscheid I silt loam soil treated at  $0.9 \mu\text{g}/100 \text{ g soil}$ , triazole was 21.4% of the applied at 14 days, 15.1% at 28 days, 7.9% at 90 days, and 5.2% at 180 days posttreatment. Hydroxytriazole was a maximum of 3.1% of the applied at 61 days posttreatment. Triazolyl alanine was  $\leq 0.6\%$  of the applied during the study period. Unidentified [ $^{14}\text{C}$ ]residues were a maximum of 2.5% of the applied at 360 days posttreatment. The soil metabolism half-life could not be calculated because further extraction of triazole residues was performed on one (clean-up II) or two samples (clean-up III) only. Unextracted [ $^{14}\text{C}$ ]residues were a maximum 75.6% of the applied at 28 days, and were 43.0% at 360 days posttreatment.  $^{14}\text{CO}_2$  totaled 55.6% of the applied at 360 days, and organic volatiles were  $<0.1\%$  of the applied during the study.
- B. In **sterile** Burscheid I silt loam soil treated at  $1.0 \mu\text{g}/100 \text{ g soil}$ , triazole was 22.9% of the applied at 28 days, 14.1% at 180 days, and 13.3% at 360 days posttreatment. Unidentified [ $^{14}\text{C}$ ]residues were a maximum of 2.9% of the applied at 180 days posttreatment. Unextracted [ $^{14}\text{C}$ ]residues were 71.8-82.7% of the applied throughout the study, with a maximum at 360 days.  $^{14}\text{CO}_2$  and organic volatiles each totaled  $<0.1\%$  of the applied throughout the study period.
- C. **Burscheid I silt loam soil, high dose.** In **nonsterile** Burscheid I silt loam soil treated at  $4874.8 \mu\text{g}/100 \text{ g soil}$ , triazole was 54.0% of the applied at 14 days, 40.6% at 60 days, and 32.6% at 180 days posttreatment. Hydroxytriazole and unidentified [ $^{14}\text{C}$ ]residues were  $<0.1$  and  $\leq 0.6\%$  of the applied, respectively, during the study period. Unextracted [ $^{14}\text{C}$ ]residues were 42.7-65.0% of the applied throughout the study, with the maximum at 180 days posttreatment.  $^{14}\text{CO}_2$

was 2.5% of the applied at 360 days, and organic volatiles were <0.1% of the applied throughout the study period.

- D. **Leifers silt loam soil, low dose:** In **nonsterile** Leifers silt loam soil treated at 0.9 µg/100 g soil, triazole was 4.0% of the applied at 12 days, varied at 0.2-0.8% at 28-90 days, and was <0.1% at 180 days. Hydroxytriazole and triazolyl alanine were maximums of 30.8 and 0.7% of the applied, respectively, at 12 days posttreatment. Unidentified [<sup>14</sup>C]residues were a maximum of 2.4% of the applied at 28 days posttreatment. Unextracted [<sup>14</sup>C]residues were a maximum of 53.5% of the applied at 28 days, and were 33.7% at 360 days posttreatment. <sup>14</sup>CO<sub>2</sub> totaled 70.3% of the applied at 360 days, and organic volatiles were <0.1% of the applied throughout the study.
- E. **Leifers silt loam soil, high dose:** In **nonsterile** Leifers silt loam soil treated at 4874.8 µg/100 g soil, triazole was 59.3% of the applied at 14 days, 43.3% at 60 days posttreatment, and 33.8% at 180 days. Hydroxytriazole was 0.9% of the applied at 60 days posttreatment. Unidentified [<sup>14</sup>C]residues were ≤0.6% of the applied at 14-180 days posttreatment. Unextracted [<sup>14</sup>C]residues were 35.2-62.4% of the applied at 14 through 180 days posttreatment. <sup>14</sup>CO<sub>2</sub> totaled 4.5% of the applied at 360 days, and organic volatiles were <0.1% of the applied during the study period.

## MATERIALS AND METHODS

Sieved (2 mm) Burscheid I silt loam, Burscheid II silt loam, and Leifers silt loam soils were used in the study; the soil characteristics are as follows (Appendix 2, p. 28):

Soil type	% Sand	% Silt	% Clay	Organic carbon (%)	pH (KCl)	Maximum water-holding capacity (g water/100 g dry soil)
Burscheid I silt loam	5.0	80.3	14.7	2.0	6.0	62.1
Burscheid II silt loam	2.6	84.4	13.0	1.8	5.4	59.0
Leifers silt loam	33.6	61.1	5.3	5.1	6.8	76.2

To study the degradation of triazole under aerobic conditions, seven test variants were used. Soil samples were treated with [3,5-<sup>14</sup>C]1,2,4-triazole (radiochemical purity >99%, specific activity 1.758 MBq/mg; p. 6), dissolved in methanol:water, at a nominal rate of 0.9 µg. The test conditions employed for each variant were as follows (pp. 7-9, Appendices 4-5, pp. 30-31):

Test variant	Soil type	µg Triazole in 100 g soil	Application of triazole	Solvent	Incubation days
1	Burscheid I silt loam	0.9	Subsamples	Methanol	14, 28, 61, 90, 180, 360
2	Burscheid I silt loam	4874.8	Subsamples	Water	14, 21, 28, 60, 180, 360
3	Burscheid II silt loam	0.9	Direct	Water	14, 28, 60, 90, 108
4	Burscheid I (sterile) silt loam	1.0	Methanolic stock solution; in droplets	Methanol	28, 90, 180, 360
5	Leifers silt loam	0.9	Subsamples	Methanol	12, 28, 61, 90, 180, 360
6	Leifers silt loam	10.1	Direct	Water	12
7	Leifers silt loam	4874.8	Subsamples	Water	14, 21, 28, 60, 180, 360

Soil subsample applications (test variants 1, 2, 5, and 7) were made by surface-treating the subsamples (30 g) with the test substance, mixing thoroughly, then adding the treated subsamples to conical bottles containing the total soil samples (p. 8). The bottles were manually rotated 100 times each around the longitudinal and lateral axes; this procedure was repeated 10 times. The thoroughly mixed samples (100 g) were weighed into 250-L conical flasks. Total soil sample applications (test variants 3 and 6) were made by directly treating the soil samples (100 g dry weight) with the test substance, mixing thoroughly, and weighing the samples into 250-L conical flasks. Methanolic stock solution applications to sterile soil samples (variant 4) were made in droplets onto the soil and the sample was thoroughly mixed (p. 9). All test soils were adjusted to 40% of the maximum water capacity. The flasks were equipped with an inlet tube for blowing out air, and were stoppered with a glass tube (14.5 cm long) containing an oil-covered layer of quartz wool topped with alternating layers of quartz wool moistened with 2% paraffin oil in hexane and soda lime (Appendix 1, p. 27). Prior to moistening the soil or removing the soil samples, humidified, ambient air was forced (1 mL/sec then 9 mL/sec, for 5 minutes each) through the incubation vessel, then sequentially through the layers of quartz wool to collect organic volatiles and soda lime to collect CO<sub>2</sub>. The treated samples were incubated in darkness in a climatic cabinet maintained at 22 ± 1°C and 60-80% humidity for up to 360 days posttreatment. Soil moisture was maintained by weighing the flasks monthly and adding distilled water as needed (approximately 0.5 mL/100 g soil/month; p. 9). Additional flasks of untreated soil were prepared as described and served as controls. Single flasks of soil were collected for analysis at 14, 21 and/or 28, 60, 61, 90, 180 and 360 days posttreatment (Appendix 4, p. 30). Volatile traps were collected for analysis at each sampling interval. For test variant 3, only the traps were collected analysis; the soil samples were further incubated using new traps (p. 7). For test variant 4, sterile soil was treated to differentiate biotic and abiotic degradation.

The soil samples were extracted with methanol:water (4:1, v:v) by shaking for 1 hour, then were centrifuged for 10 minutes (Extraction I; p. 15). The supernatant decanted over a folded filter, and the extraction was repeated twice with methanol. For each soil, the extracts were combined, concentrated by rotary evaporator ( $\leq 25^{\circ}\text{C}$ ), and centrifuged. Aliquots of the extracts were analyzed for specific [ $^{14}\text{C}$ ]compounds by one-dimensional TLC on silica gel plates developed with the following solvent systems (pp. 12-13):

Solvent System	
System A	Chloroform:methanol:aqueous ammonia solution (65:28:8; v:v:v)
System B	n-Butanol:propionic acid:water (45:22:33; v:v:v)
System C	Chloroform:methanol:water (69:27:4; v:v:v)
System D	Chloroform:methanol:water:aqueous ammonia solution (66:30:4:4; v:v:v:v)
System E	Chloroform:methanol:water:acetic acid (86:26:4:2; v:v:v:v)
System F	Acetonitrile:water:aqueous ammonia solution (80:18:2; v:v:v)

Additional aliquots of the extracts were analyzed using reverse-phase TLC on RP-18 plates developed in methanol:water (1:4, v:v). The extracts were cochromatographed with unlabeled reference standards of triazole, hydroxytriazole (3-hydroxy-1,2,4-triazole), 1,2,4-triazole-3-alanine, and i-butyl triazolyl alanine, and radiolabeled reference standards of triazolyl alanine (1,2,4-triazole-1-alanine) and triazolyl acetic acid (Appendix 8, p. 34). Radioactive areas on the plates were compared with the zones of the reference standards. The following methods were used to visualize the reference standards: (i) spraying (sodium nitroprusside-potassiumhexacyanoferrate (III) solution and ninhydrin), (ii) linear analysis, or (iii) autoradiography (pp. 13-14). Radioactive areas were quantified by linear analysis and/or by scraping the radioactive zones off the silica gel plates and counting by LSC. The extracted soils were air-dried and analyzed for total radioactivity using LSC following combustion (pp. 10, 12). In attempts to release additional extractable [ $^{14}\text{C}$ ]residues from the test soils, selected postextracted soils from each test variant were subjected to one or two aggressive, alkaline extractions. Selected subsamples (50 g) of extracted soil were re-extracted with methanol:25% aqueous ammonia solution (7:3; v:v) by shaking for 2 hours at  $21^{\circ}\text{C}$ , then centrifuged (Extraction II; p. 15). The supernatant was analyzed for total radioactivity using LSC and for [ $^{14}\text{C}$ ]compounds using one-dimensional TLC as previously described. Selected subsamples (50 g) of extracted soil from extractions I and/or II were re-extracted with methanol:25% aqueous ammonia solution (7:3; v:v) by refluxing for 4 hours at  $60^{\circ}\text{C}$ , and centrifuged (Extraction III). The supernatant and combusted sediment were analyzed for total radioactivity by LSC and LSC following combustion, respectively, and for [ $^{14}\text{C}$ ]compounds using one-dimensional TLC as previously described. Recovery rates of triazole, triazolyl acetic acid, and triazolyl alanine in the Burscheid I and Leifers silt loam soils following Extractions I-III were 78.1-96.7% of the applied (Appendix 19, p. 43).

The identity of hydroxytriazole was confirmed by HPLC analysis of the 28-day methanol:water extracts from test variant V and by GC/MS analysis of the 12-day methanol:water extract from

test variant 6 (pp. 17-18). The identity of triazolyl alanine was confirmed by one-dimensional TLC analysis of the 12-day methanol:water extracts from test variants 5 and 6, before and after derivatization (test variant 5 only) with HCl-isobutanol solution (p. 18). The identities of triazole and derivatives of hydroxytriazole and triazolyl acetic acid (N-methyl-bis-trimethylsilyltrifluoroacetamide) and triazolyl alanine (HCl-isobutanol solution) were confirmed by GC/MS (p. 20).

The oil-coated quartz wool plug was extracted with ethyl acetate and aliquots of the extracts were analyzed for total radioactivity by LSC (p. 10). Aliquots of the soda lime trapping solutions were analyzed for total radioactivity by LSC. The presence of  $^{14}\text{CO}_2$  was confirmed by dissolving the soda lime in 18% HCl; the evolved  $^{14}\text{CO}_2$  was fed into a  $\beta$ -phenylethylamine/butyl PBD cocktail and analyzed by LSC (Appendix 6, p. 32). The presence of  $^{14}\text{CO}_2$  was also confirmed by [ $^{14}\text{C}$ ]benzoic acid, by means of the Grignard reaction (p. 11, Appendix 7, p. 33).

Microbial viability of the soils were determined at the initiation and during the study period (p. 6). Microbial viability of Burscheid I and Leifers silt loam soils at time 0 were 826 and 1386 mg C/kg dry soil, respectively, and at 360 days posttreatment were 352 and 861 mg C/kg dry soil, respectively (Appendix 10, p. 36; Appendix 13, p. 39).

## RESULTS/DISCUSSION

The aerobic metabolism of ([3,5- $^{14}\text{C}$ ]1,2,4-triazole; radiochemical purity >99%) was studied in two silt loam soils (Burscheid I and Leifers) that were treated at a nominal rate of 0.9  $\mu\text{g}/100\text{ g}$  soil and incubated in the dark at  $22 \pm 1^\circ\text{C}$  for up to 360 days. Seven test variants were employed. One sample was analyzed at each sampling interval; the first sampling interval for all test variants was 12, 14 or 28 days posttreatment. The proposed degradation pathway of triazole in soil under aerobic conditions is shown in Appendix 33 (p. 59).

Mineralization of 1,2,4-triazole at low concentrations (0.9  $\mu\text{g}$  of triazole/100g soil) reached 25% in Burscheid I soil and 51% in Leifers soil within 90 days. The main degradate of 1,2,4-triazole is hydroxytriazole (3-hydroxy-1,2,4-triazole;  $\leq 30.8\%$ ), minor degradates are triazolyl alanine (1,2,4-triazole-1-alanine) and triazolyl acetic acid. No mineralization occurred in sterile Burscheid I soil. Rates of mineralization were low ( $\leq 4.5\%$  of the applied) in high-dosed nonsterile silt loam soils.

**Test variants 1, 4, and 5** were performed to determine the nature of triazole degradation. Based on higher mineralization rates in nonsterile silt loam soils (test variants 1 and 5) compared to sterile silt loam soil (test variant 4), the degradation of triazole was biological in nature.

**Test variant 1:** In Burscheid I silt loam soil treated at 0.9  $\mu\text{g}/100\text{ g}$  soil, triazole was 21.4% of the applied at 14 days, 15.1% at 28 days, 7.9% at 90 days, and 5.2% at 180 days posttreatment (Appendix 10, p. 36). Hydroxytriazole was a maximum of 3.1% of the applied at 61 days posttreatment. Triazolyl alanine was  $\leq 0.6\%$  of the applied during the study period. Unidentified [ $^{14}\text{C}$ ]residues were a maximum of 2.5% of the applied at 360 days posttreatment. Unextracted [ $^{14}\text{C}$ ]residues were a maximum 75.6% of the applied at 28 days, and were 43.0% at 360 days

posttreatment.  $^{14}\text{CO}_2$  totaled 55.6% of the applied at 360 days, and organic volatiles were <0.1% of the applied during the study. Following one or two alkaline extractions of the 28- and 180-day postextracted soil, triazole comprised 24.1-44.1% of the applied (reviewer-calculated; summed data), and hydroxytriazole, triazolyl alanine, and unidentified [ $^{14}\text{C}$ ]residues were each  $\leq 1.9\%$  of the applied.

**Test variant 5:** In Leifers silt loam soil treated at 0.9  $\mu\text{g}/100$  g soil, triazole was 4.0% of the applied at 12 days, varied at 0.2-0.8% at 28-90 days, and was <0.1% at 180 days (Appendix 13, p. 39). Hydroxytriazole was 30.8% of the applied at 12 days, 11.0% at 28 days, 3.8% at 61 days, and 0.6% at 180 days posttreatment. Triazolyl alanine was 0.7% of the applied at 12 days and was not detected at other sampling intervals. Unidentified [ $^{14}\text{C}$ ]residues were a maximum of 2.4% of the applied at 28 days posttreatment. Unextracted [ $^{14}\text{C}$ ]residues were a maximum of 53.5% of the applied at 28 days, and were 33.7% at 360 days posttreatment.  $^{14}\text{CO}_2$  totaled 70.3% of the applied at 360 days posttreatment, and organic volatiles were <0.1% of the applied throughout the study. Following one or two alkaline extractions of the 12-, 28-, 61-, and 180-day postextracted soil, triazole comprised 1.4-10.3% of the applied, hydroxytriazole was 1.9-11.1%, triazolyl alanine and triazolyl acetic acid were each  $\leq 3.6\%$ , and unidentified [ $^{14}\text{C}$ ]residues were  $\leq 5.0\%$  of the applied.

**Test variant 4:** In sterile Burscheid I silt loam soil treated at 1.0  $\mu\text{g}/100$  g soil, triazole was 22.9% of the applied at 28 days, 14.1% at 180 days, and 13.3% at 360 days posttreatment (Appendix 10, p. 36). Unidentified [ $^{14}\text{C}$ ]residues were a maximum of 2.9% of the applied at 180 days posttreatment. Unextracted [ $^{14}\text{C}$ ]residues were 71.8-82.7% of the applied throughout the study, with the maximum at 360 days.  $^{14}\text{CO}_2$  and organic volatiles each totaled <0.1% of the applied throughout the study period. Following two alkaline extractions of the 180-day postextracted soil, triazole comprised 47.6% of the applied (reviewer-calculated) and unidentified [ $^{14}\text{C}$ ]residues were 0.4% of the applied.

**Test variant 6** was conducted at 10x the nominal application rate in order to obtain a sufficient amount of hydroxytriazole for structure elucidation. In Leifers silt loam soil treated at 10.1  $\mu\text{g}/100$  g soil, triazole was 12.7% of the applied, hydroxytriazole was 21.5%, and triazolyl alanine was 1.1% at the single sampling interval of 12 days posttreatment. Unidentified [ $^{14}\text{C}$ ]residues were 4.3%, unextracted [ $^{14}\text{C}$ ]residues comprised 61.0%, and  $^{14}\text{CO}_2$  was 5.4% of the applied (Appendix 13, p. 39). Following one alkaline extraction of the 12-day postextracted soil, triazole was 7.8%, hydroxytriazole and triazolyl alanine were each  $\leq 2.7\%$ , and unidentified [ $^{14}\text{C}$ ]residues were 0.4% of the applied.

**Test variants 2 and 7** were conducted at an exaggerated application rate to determine whether the low degradation of triazole correlated with the high amount of triazole and/or the soil.

**Test variant 2:** In Burscheid I silt loam soil treated at 4874.8  $\mu\text{g}/100$  g soil, triazole was 54.0% of the applied at 14 days, 40.6% at 60 days, and 32.6% at 180 days posttreatment (Appendix 12, p. 38). Hydroxytriazole and unidentified [ $^{14}\text{C}$ ]residues were <0.1 and  $\leq 0.6\%$  of the applied, respectively, during the study period. Unextracted [ $^{14}\text{C}$ ]residues were 42.7-65.0% of the applied throughout the study, with the maximum at 180 days posttreatment.  $^{14}\text{CO}_2$  was 2.5% of the applied at 360 days, and organic volatiles were <0.1% of the applied throughout the study period.



Following two alkaline extractions of the 180-day postextracted soil, triazole comprised 31.2% of the applied, and hydroxytriazole, triazolyl alanine, and triazolyl acetic acid were each  $\leq 0.3\%$  of the applied.

**Test variant 7:** In Leifers silt loam soil treated at 4874.8  $\mu\text{g}/100\text{ g}$  soil, triazole was 59.3% of the applied at 14 days, 43.3% at 60 days posttreatment, and 33.8% at 180 days (Appendix 15, p. 41). Hydroxytriazole was a maximum of 0.9% of the applied at 60 days posttreatment. Unidentified [ $^{14}\text{C}$ ]residues were 0.1% of the applied at 60 and 180 days. Unextracted [ $^{14}\text{C}$ ]residues were 35.2-62.4% of the applied at 14-180 days, with a maximum at 180 days.  $^{14}\text{CO}_2$  totaled 4.5% of the applied at 360 days posttreatment, and organic volatiles were  $<0.1\%$  of the applied during the study period. Following two alkaline extractions of the 180-day postextracted soil, triazole comprised 18.6%, hydroxytriazole, triazolyl alanine, and triazolyl acetic acid were each  $\leq 1.3\%$ , and unidentified [ $^{14}\text{C}$ ]residues were 3.3% of the applied.

**Test variant 3** was conducted to study mineralization of triazole using Burscheid II silt loam soil treated at 0.9  $\mu\text{g}/100\text{ g}$  soil; only volatile traps (for  $^{14}\text{CO}_2$  determination) were sampled. Throughout the study, the majority of the [ $^{14}\text{C}$ ]residues were  $^{14}\text{CO}_2$  (Appendix 16, p. 42).

#### DEFICIENCIES/DEVIATIONS

1. No 0-day soil samples were collected in any of the test systems. Additionally, only selected postextracted soil samples were subjected to further extraction to release additional extractible [ $\text{C}^{14}$ ] residues from the test soil. The half-life calculations are not possible without zero-time data and lack of subsequent extraction data for all sampling intervals.
2. The test soils were from Germany and Italy. These soils were not classified according to the USDA Soil Classification System, and were not compared to soils in the United States.
3. The soil moisture was maintained at 40% maximum water capacity (p. 9). Subdivision N Guidelines specify that soil moisture should be maintained at 75% of 0.33 bar.
4. Single soil samples were analyzed for each test soil at each sampling interval. Subdivision N Guidelines recommend that at each sampling interval, at least two samples be collected.
5. A fortification experiment was conducted in which soil was treated with [3,5- $^{14}\text{C}$ ]1,2,4-triazole-1- alanine at 0.5  $\mu\text{g}/100\text{ g}$  soil (p. 23). Two incubation batches were prepared, and only the volatile traps were collected for analysis (p. 7). At 108 days posttreatment,  $^{14}\text{CO}_2$  comprised 7.8% of the radioactivity, representing 75% of the recovered (reviewer-calculated) at that sampling interval. The results demonstrate rapid mineralization of the test substance (Appendix 16, p. 42).

6. The study author concluded that the aerobic degradation of triazole at 0.9  $\mu\text{g}/100\text{ g}$  soil occurs via hydroxytriazole to  $\text{CO}_2$  (p. 24). The author noted that the mineralization in the Burscheid I and Leifers soils increased from 25 and 51% of the applied at 90 days to 50 and 70% at 360 days posttreatment.
7. The mineralization of 1,2,4-triazole was studied using Variant 3, in which only the volatile traps were sampled and analyzed. The Grignard reaction was used to identify  $^{14}\text{CO}_2$  (p. 10). Based on the data presented in Appendix 16 (p. 42), most of the radioactivity in the 31-day samples was identified as  $^{14}\text{CO}_2$ . Based on these findings, the reviewer concluded that the majority of the [ $^{14}\text{C}$ ]residues in the trapping solutions was  $^{14}\text{CO}_2$ .
8. The maximum proposed or registered field application rate of triazole was not reported in the study. It was stated that the commercial use rate of triadimefon on pome fruit (50 g triadimefon/ha is equivalent to 11.7 g triazole/ $\text{dm}^2$ ) results in a maximum amount of 9  $\mu\text{g}$  triazole/kg soil at a 10-cm depth and a soil density of 1.3  $\text{g}/\text{cm}^3$  (p. 7).
9. Seven solvent systems were used in the thin layer chromatography for analysis of [ $^{14}\text{C}$ ] compounds (pp. 12-13). The solvent system(s) used for analysis of samples from each test variant was not specified.
10. The limits of detection and quantitation for the LSC, TLC, HPLC, and GC/MS methods were not reported.
11. Two radioactive samples ( $a_1$  and  $a_2$ ) of triazole were available. The specific activity of the two samples was reported as 1.758 and 30.780 Mbq/mg, respectively (p. 6). The reviewer reported the specific activity of the test substance as  $\geq 1.758$  Mbq/mg.
12. Representative TLC radiochromatograms and HPLC chromatograms presented in Appendices 18-29, pp. 44-55 indicated good separation of compounds.
13. The solubility of triazole in water was reported to be  $>2000$  ppm (MRID 45284023 p. 6)
14. Good Laboratory Practice and Quality Assurance Statements were not provided with the study.

ATTACHMENT 1  
Data Critical to the Study Interpretation

THE FOLLOWING ATTACHMENT IS NOT AVAILABLE ELECTRONICALLY  
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Page \_\_\_\_\_ is not included in this copy.

Pages 12 through 29 are not included in this copy.

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The material not included contains the following type of information:

- Identity of product inert ingredients.
- Identity of product impurities.
- Description of the product manufacturing process.
- Description of quality control procedures.
  
- Identity of the source of product ingredients.
- Sales or other commercial/financial information.
- A draft product label.
- The product confidential statement of formula.
- Information about a pending registration action.
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