Accelerated Soil Dissipation of Tebuconazole following Multiple Applications to Peanut

Thomas L. Potter,* Timothy C. Strickland, Hyun Joo, and Albert K. Culbreath

ABSTRACT

Repeated application may increase rates of pesticide dissipation in soil and reduce persistence. The potential for this to occur was investigated for the fungicide, tebuconazole (α-[2-(4-chlorophenyl)ethyl]-α-(1,1-dimethylethyl)-1H-1,2,4-triazole-1-ethanol), when used for peanut (Arachis hypogaea L.) production. Soil samples were collected from peanut plots after each of four tebuconazole applications at 2-wk intervals. Soil moisture was adjusted to field capacity as necessary and samples were incubated in the laboratory for 63 d at 30°C. Untreated plot samples spiked with the compound served as controls. Results indicated accelerated dissipation in field-treated samples with the time to fifty percent dissipation (DT50) decreasing from 43 to 5 d after three tebuconazole applications. Corresponding increases in rates of accumulation and decay of degradates were also indicated. Best-fit equations (r2 = 0.84–0.98) to dissipation kinetic data combined with estimates of canopy interception rates were used to predict tebuconazole and degradates concentrations in soil after each successive application. Predicted concentrations compared with values measured in surface soil samples were from twofold less to twofold greater. Use of kinetic data will likely enhance assessments of treatment efficacy and human and ecological risks from normal agronomic use of tebuconazole on peanut. However, the study indicated that varying soil conditions (in particular, soil temperature and water content) may have an equal or greater impact on field dissipation rate than development of accelerated dissipation. Results emphasize that extension of laboratory-derived kinetic data to field settings should be done with caution.

About 75% of the annual peanut crop in the United States is produced within the Atlantic Coastal Plain region, extending from Virginia to Alabama (USDA National Agricultural Statistics Service, 2004) and centered in south-central Georgia. Peanut is well adapted to the region’s soils and humid subtropical climate. These conditions also promote high disease pressures. Foliar and soilborne fungal pathogens are particularly troublesome. Estimated losses associated with yield and quality reductions and the cost of control in Georgia exceed $50 million annually (Guillebeau, 2003).

Typically growers make six to eight fungicide applications during the growing season beginning about four weeks after planting with sprays repeated at two-week intervals until harvest. Surveys indicate that >90% of the Georgia crop is treated with at least one fungicide and that the two most widely used active ingredients are chlorothalonil (2,4,5,6-tetrachloro-1,3-benzenedicarboxonitrile) and tebuconazole (USDA National Agricultural Statistics Service, 2000). Estimated percent of acreage treated was 85% for chlorothalonil and 47% for tebuconazole.

Chlorothalonil effectively controls foliar but has little impact on soilborne diseases whereas tebuconazole provides control of both (Brenneman et al., 1991; Brenneman and Culbreath, 1994; Branch and Brenneman, 1996). The difference in behavior of the two compounds may be linked to their difference in soil persistence. In prior work we found that the aerobic soil half-life (t1/2) of chlorothalonil in Tifton loamy sand (fine-loamy, kaolinitic, thermic Plinthic Kandiudult), a soil commonly used for peanut production in the Georgia Coastal Plain region, was very short, ranging from <1 to 3.5 d (Potter et al., 2001). The t1/2 also decreased about fourfold after three successive chlorothalonil applications two weeks apart indicating that accelerated dissipation conditions developed rapidly. The t1/2 of tebuconazole in the same soil was much longer, 49 d (Strickland et al., 2004). The potential for development of accelerated dissipation conditions was not evaluated.

Among the triazoles, the fungicide group to which tebuconazole belongs, soil persistence is a general feature. The aerobic soil t1/2 measured in some laboratory studies exceeded several years (Bromilow et al., 1999a) and, as occurs for many pesticides, a trend toward reduced persistence in field versus laboratory studies was reported (Bromilow et al., 1999b). There also appears to be potential for accelerated triazole biodegradation. Thom et al. (1997) observed that difenoconazole time to 50% dissipation (DT50) under aerobic conditions in a silt loam soil decreased by about twofold when dissipation in nonpretreated and pretreated soils was compared. Accelerated degradation has been reported to reduce the efficacy of many soil-applied pesticides (Suett et al., 1996). Our literature review did not identify any studies with tebuconazole in this regard nor field dissipation studies in peanut or other crops.

In this report we describe a combined field–laboratory study focused on determining tebuconazole dissipation kinetics in soil following multiple applications to peanut. The amount of tebuconazole reaching the soil surface when broadcast sprays were made to the plant canopy was measured, and dissipation of the parent

Abbreviations: DAP, days after planting; DT10, DT50, and DT90, time to 10, 50, and 90% dissipation, respectively; Koc, soil organic carbon–water partition coefficient; LAI, leaf area index; t1/2, half-life.
compound and accumulation and decay of principal degradates were monitored during laboratory incubations of soil collected from treated plots. Incubations of untreated soil spiked with tebuconazole in the laboratory served as a control. The primary objective of the study was to evaluate the soil persistence of tebuconazole in an environment where it is used heavily as this may have implications for optimizing applications for disease management. In addition there are water quality considerations. Weather patterns and soil conditions in the region indicate that surface waters are vulnerable to impacts from pesticide runoff (Goss et al., 1998). A key factor in assessing runoff risks of any pesticide is concentration in surface soil (Leonard, 1990); thus, there is a need to comprehensively assess post-application dissipation rates to estimate the amount of tebuconazole that remains available for runoff.

**MATERIALS AND METHODS**

**Field Conditions and Crop Management**

Peanuts (var. AT 201) were grown at a University of Georgia research farm near Tifton, Georgia (31°28’ N, 83°35’ W) in twin rows (0.5 m on center) on 7.6-m plots separated by 2.4-m alleys. Planting and digging dates were 27 May and 10 Oct. 2002, respectively. Plots used for the study were part of a comparative investigation of the curative activity of three fungicides, tebuconazole, pyraclostrobin, and chlorothalonil, applied from two to seven times during the growing season. Formulations were Folicur 3.6F (Bayer CropScience LP, Research Triangle Park, NC), Headline 2.09 EC (BASF Corp., Research Triangle Park, NC), and Bravo WeatherStik 720F (Syngenta Crop Protection, Greensboro, NC), respectively. There were four replicates per treatment group and four untreated controls in a randomized complete block design. The current study focused on plots that received four Folicur applications. Sprays were made using a tractor mounted CO₂-propellant sprayer with three D3-23 hollow cone nozzles per row at a tebuconazole target rate of 0.22 kg ha⁻¹. Total tebuconazole applied was the maximum label rate for a peanut crop (Bayer CropScience, 2002). Crops in the three previous years at the study site were: cotton in 1999, soybeans in 2000, and cotton in 2001 and they did not receive tebuconazole. Daily irrigation and rainfall amounts and dates of tebuconazole application, soil sample collection, and digging, and the average daily soil temperature measured at 5 cm at a weather station about 1 km south of the study site are shown in Fig. 1. Rainfall totaled 236 mm and irrigation 177 mm during the growing season. A preemergence herbicide application of ethalfluralin and metolachlor at 1 and 1.5 kg ha⁻¹, respectively, was made 4 d before planting. Soil at the test site is Tifton fine-loamy sand (1-2% slope). Composite surface (0-2 cm) soil samples collected at approximately 2-wk intervals (n = 7) had the following characteristics: median pH = 6.1, total organic carbon = 3.9 ± 0.3 g kg⁻¹, and organic total nitrogen = 0.24 ± 0.06 g kg⁻¹. Nitrogen and carbon analyses were performed on sieved (10 mesh) pulverized samples by dry combustion using a Carlo-Erba Model WA 1500 Series II analyzer (CE Elantech, Lakewood, NJ). Textural analysis was performed on two of the surface soil composites. Samples were 830 to 900 g kg⁻¹ sand, 30 to 80 g kg⁻¹ clay, and 50 to 150 g kg⁻¹ silt.

**Application Rate Measurement and Soil Sample Collection**

Before each tebuconazole application, two 7-cm-diameter cellulose filter papers were deployed on each of the four treated and control plots as spray targets. On each plot, one target was attached to plant tops using paper clips so that the filter paper was horizontal and the other was pinned to the soil surface at the mid-point between rows. One hour after application, spray targets were collected. A composite soil sample was then obtained by combining four subsamples collected at the midpoint between the two twin rows on each treated plot using a 13- by 10-cm stainless steel trowel with an effective sampling depth of 0 to 2 cm. Areas disturbed by prior sampling were avoided. Samples on 59 and 71 days after planting (DAP) were collected beneath the plant canopy, which closed between 54 to 58 DAP. Soil composites were obtained in the same way from control plots on 28 and 71 DAP.

**Soil Incubations**

Composites were prepared for incubations by removing visible plant debris and stones with tweezers followed by sieving using a 10-mesh stainless steel sieve. The sieved field-moist soil (50 g) was then weighed into 240-mL French-square glass bottles (n = 7). Soil moisture content was adjusted to 12% gravimetric as necessary by addition of distilled-deionized water. The water content was at the midpoint of the range of values for field capacity for Tifton surface soil (D.D. Bosch, personal communication, 2005). All bottles were closed with Teflon-lined screw caps after methanol (50 mL) was added to three of the bottles. Those containing methanol were placed in a chest freezer held at −20°C. The other bottles were closed and placed in a dark laboratory incubator maintained at 30 ± 1°C. The incubation temperature was based on available soil temperature data. The long-term average (June to August) measured at 5 cm at two sites in the county where the study was conducted was 29 ± 3°C (University of Georgia, 2004). Three bottles were removed on Days 1, 4, 7, 14, 21, 28, 42, and 63 and 50 mL of methanol was added to each. After recapping, bottles were transferred to the −20°C chest freezer. Termination of the incubations at 63 d was based on the draft USEPA–OECD harmonized guideline for aerobic soil metabolism studies (USEPA, 1998). All bottles, glassware, and steel implements were washed with soap and water and rinsed with distilled-deionized water and acetone and dried in a laboratory oven at 125°C overnight before use. Sixty-mesh quartz sand (200 mg) containing 192 ± 12 µg g⁻¹ tebuco-
Tebuconazole was added to bottles containing soil from control plots followed by capping and vigorous shaking. With the exception of the sand addition, control samples were handled as the field treated samples. The spiking sand was prepared by combining methanol containing tebuconazole with sand in a beaker and allowing the methanol to evaporate overnight. After drying the sand was stirred with a stainless steel spatula and three 200-mg subsamples were extracted with methanol and extracts analyzed to determine the tebuconazole concentration.

**Tebuconazole and Degradates Extraction and Analysis**

Procedures described by Strickland et al. (2004) were followed. Briefly, about 1 wk after termination of the last incubation, bottles containing soil and methanol (in sets of 27) were brought to room temperature and sequentially extracted (3 by 50-mL) with methanol. Spray targets were extracted by shaking for 1 h with 25 mL methanol. After filtration and concentration to 10 mL, extracts were analyzed by high performance liquid chromatography–tandem mass spectrometry using a Thermoquest LCQ DECA system (Thermoquest-Finnigan, San Jose, CA). An atmospheric pressure chemical ionization ion source was used. Tebuconazole concentrations in soil sample extracts and standards were divided by 1.2 to account for a matrix enhancement that was observed. In each analysis, the four degradates (Fig. 2) proposed by Strickland et al. (2004) were monitored. Their concentration was estimated using the assumption of equivalent response of their protonated molecular ions, \((M + H)^+\) to tebuconazole’s \((m/z = 308)\). Subsequently we were able to obtain reference standards of Degradates 2, 3, and 4. Their analysis confirmed proposed structural assignments for Degradates 2 and 4 and their presence in soil extracts, and provided relative response factors (RRFs) to tebuconazole. The RRFs were used to adjust initial concentration estimates of these compounds. Reference standard analyses also demonstrated that Degradate 3 (the hydroxy acid form) was separated chromatographically from the other degradates and tebuconazole using our analytical conditions. Re-examination of all chromatograms did not reveal a peak corresponding to Degradate 3; thus, we concluded that it was not detected. The estimated detection limit based on the lowest calibration standard analyzed was 0.002 \(\mu g \cdot g^{-1}\).

**Chemicals and Supplies**

Tebuconazole was purchased from Chem-Service (Chester, PA) and 2-chlorolepidine from Sigma-Aldrich (Milwaukee, WI). Reference standards of Degradates 2, 3, and 4 were donated by Bayer CropScience (Kansas City, MO). Optima-grade solvents, filters, and other supplies were purchased from Fisher Scientific (Hampton, NH).

**Quality Control**

All laboratory incubation samples were analyzed in triplicate. Among the 55 sample sets for which complete data were obtained, the relative standard deviation averaged 7.6 ± 6.3%.
Table 1. Mean ± standard deviation of tebuconazole application and soil deposition rates for the four broadcast sprays on peanut (based on filter paper spray targets).

<table>
<thead>
<tr>
<th>DAP†</th>
<th>Applied</th>
<th>Soil deposition</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>kg ha⁻¹</td>
<td></td>
</tr>
<tr>
<td>28</td>
<td>0.22 ± 0.05</td>
<td>0.19 ± 0.01</td>
</tr>
<tr>
<td>43</td>
<td>0.17 ± 0.08</td>
<td>0.23 ± 0.17</td>
</tr>
<tr>
<td>59</td>
<td>0.19 ± 0.04</td>
<td>no sample</td>
</tr>
<tr>
<td>71</td>
<td>0.22 ± 0.02</td>
<td>0.01 ± 0.01</td>
</tr>
</tbody>
</table>

† Days after planting.

Results from time zero spiked control samples provided data to evaluate storage stability and matrix-spike recovery. The average percent recovery was 87 ± 6.6% with the relative standard deviation = 4.5% (n = 6) demonstrating that tebuconazole was stable during storage, solvent extractions were quantitative, and measurement precision was relatively high.

Data Analysis

Tebuconazole dissipation was fit to logarithmically transformed forms of the first-order rate expression (linear) and the spatially distributed first-order model (nonlinear) proposed by Gustafson and Holden (1990), by linear regression using the spreadsheet Excel (Microsoft, 2003). Discussion of the use and applicability of these models is available in recent publications (Potter et al., 2002; Wolt et al., 2001)

RESULTS AND DISCUSSION

Fungicide Application and Foliar Interception

Mean application rates to the plant canopy, 0.17 to 0.22 kg ha⁻¹ (Table 1), were close to the target rate of 0.22 kg ha⁻¹. Deposition to soil, measured by positioning spray targets between rows, was also close to the target rate during the first two sprays on 28 and 43 DAP (Table 1). The crop canopy had not closed between rows, so spray targets staked to the soil surface were sprayed directly. Soil means, although higher, were not significantly different from measured canopy application rates. Spray targets were not deployed on the soil during the third application (59 DAP). During the fourth application (71 DAP), the percent deposition on soil compared with the canopy was 5.5 ± 2.7% of applied (Table 1). This was in close agreement with two recently published studies describing spray penetration of peanut canopies. Wauchope et al. (2004) reported that 4.4 ± 4.0% of foliar chlorothalonil applications at 64 and 80 DAP was detected in spray traps positioned at the soil surface beneath peanut plants. Zhu et al. (2004) using fluorescein dye reported that the average spray deposit at 75 DAP was 3 to 4% of the amount deposited on plant canopy surfaces.

The dye measurements also indicated that some adjustment for plant growth stage with regard to soil deposition rate may be appropriate. Bottom-of-canopy dye deposition was 8 to 12% at 41 DAP, 3 to 4% at 75 DAP, and 4 to 7% at 104 DAP. A trend to higher direct soil deposition at 104 when compared with 71 DAP was linked to a 20% decrease in leaf area index (LAI) (Zhu et al., 2004). Peanut LAI reduction of this magnitude or greater at this stage is common (Truman and Williams, 2001) and since one to two fungicide sprays are commonly made during this time period, increased soil deposition is likely. This type of behavior was inferred in a prior study with chlorothalonil (Potter et al., 2001). Three- to fivefold higher concentrations were detected in surface soil (0–2 cm) samples collected after a spray on 114 DAP compared with sprays on 59, 71, 87, and 100 DAP on peanut.

Tebuconazole Dissipation in Laboratory Incubations

The times to 10% (DT₉₀), 50% (DT₅₀), and 90% (DT₉₀) dissipation computed using the linear and nonlinear kinetic models were remarkably uniform for the spiked controls, and uniformly high r² values (>0.919) showed that both kinetic models provided a good fit to the data (Table 2). The extent of tebuconazole dissipation when incubations were terminated at 63 d was 82 to 86%. This was close to the guideline level, 87.5% (2.5 t₁₀), for study duration used for registrant submissions to the USEPA (Wolt et al., 2001). Tebuconazole dissipation along with degradate accumulation and decay are summarized in Fig. 2 for the 28 DAP and Fig. 3 for the 71 DAP soil samples. Data for incubations for corresponding treated plot soil samples are included for comparison.

For the spiked control samples the t₁₀ determined in both incubations, 25 d, was about half that found in a prior investigation using soil samples collected from the same field (Strickland et al., 2004). The later study was

Table 2. Computed tebuconazole time to 10% (DT₉₀), 50% (DT₅₀), and 90% (DT₉₀) dissipation during laboratory incubation of soil samples collected from treated and control plots using two kinetic models.

<table>
<thead>
<tr>
<th>Sample source (date of collection)</th>
<th>Cᵣ</th>
<th>Kinetic model</th>
<th>DT</th>
<th>r²</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>nmol g⁻¹</td>
<td></td>
<td>(d)</td>
<td></td>
</tr>
<tr>
<td>Treated plots (28 DAP†)</td>
<td>2.7 ± 0.07</td>
<td>nonlinear</td>
<td>6 41 140</td>
<td>0.952</td>
</tr>
<tr>
<td></td>
<td>3.1 ± 0.09</td>
<td>linear</td>
<td>6 41 140</td>
<td>0.952</td>
</tr>
<tr>
<td></td>
<td>2.4 ± 0.32</td>
<td>linear</td>
<td>4 29 95</td>
<td>0.918</td>
</tr>
<tr>
<td>Treated plots (71 DAP)</td>
<td>1.9 ± 0.14</td>
<td>nonlinear</td>
<td>2 13 45</td>
<td>0.872</td>
</tr>
<tr>
<td></td>
<td>1.9 ± 0.03</td>
<td>nonlinear</td>
<td>1 12 73</td>
<td>0.896</td>
</tr>
<tr>
<td>Treated plots (59 DAP)</td>
<td>1.9 ± 0.10</td>
<td>nonlinear</td>
<td>3 18 59</td>
<td>0.919</td>
</tr>
<tr>
<td>Spiked control (28 DAP)</td>
<td>1.9 ± 0.03</td>
<td>nonlinear</td>
<td>4 25 83</td>
<td>0.919</td>
</tr>
<tr>
<td>Spiked control (71 DAP)</td>
<td>1.9 ± 0.10</td>
<td>nonlinear</td>
<td>4 25 83</td>
<td>0.975</td>
</tr>
</tbody>
</table>

† Days after planting.
conducted at 24 ± 1°C. The incubation temperature was 30 ± 1°C in the current work. To more appropriately compare results between studies, the $t_{1/2}$ measured at 24°C was adjusted to 30°C using the Arrhenius equation (Boesten et al., 2004).

Use of the equation requires compound specific activation energy ($E_a$). No data were available for tebuconazole. An $E_a$ estimate of 52 ± 42 kJ mol$^{-1}$ was derived by taking the mean and standard deviation of $E_a$ computed from published studies describing the degradation of other triazole fungicides as a function of temperature (Beigel et al., 1999; Bromilow et al., 1999a; Singh and Duryea, 2000). Using the estimated $E_a$, $t_{1/2}$ adjusted to 30°C was 32 ± 11 d for the study conducted at 24°C. Based on this, $t_{1/2}$ values obtained in our current and prior work were in general agreement. Some contributing factors to the variation observed included sample collection several months apart (beginning of May in one case and end of June and end of July in the others) and use of higher initial tebuconazole concentration, about twofold in the 24°C study. However, it appears the influence of these factors was relatively small compared with the uncertainty associated with $E_a$ estimation and $t_{1/2}$ temperature adjustment. Activation energy was a highly sensitive parameter in a pesticide leaching model that took into account impacts of temperature on pesticide degradation rate (Wu and Nofziger, 1999).

In contrast to the spiked control plots, tebuconazole DT$_{10}$, DT$_{50}$ and DT$_{90}$ determined by incubation of soil samples collected from treated plots differed by up to 12-fold with a trend toward decreasing values with increasing DAP for sample collection and the nonlinear model provided an improved fit ($r^2$) to data for some samples (Table 2). Overall, percent tebuconazole dissipation on the incubation termination date (63 d) was 67, 82, 92, and 90% for soil samples collected after the first (28 DAP), second (43 DAP), third (59 DAP), and fourth (71 DAP) tebuconazole applications, respectively (Table 3). During incubation of the third (59 DAP) and fourth (71 DAP) samples, dissipation was more rapid at the initial stage than could be described accurately using the linear model. This type of behavior is often observed in laboratory and field pesticide dissipation studies and lead to development of a variety of nonlinear models (Wolt et al., 2001). Models of this type typically provide improved data fits due to increases in the number of variable parameters. Lower DT$_{10}$ and DT$_{50}$ and higher DT$_{90}$ are also often observed when compared with the linear first-order kinetic model. These trends were reflected in our data (Table 2).

Using nonlinear model results, DT$_{10}$ decreased from an initial value of 6 d for the first treated soil sample (28 DAP) to 0.5 d for the sample collected after the third application (59 DAP). The DT$_{10}$ for the fourth and last sample collected (71 DAP) was consistently low, 1 d, while the second sample (43 DAP) sample gave a value intermediate between the first (28 DAP) and third (59 DAP) samples. The same trends were observed for DT$_{50}$ and DT$_{90}$ with the highest value associated with the first (28 DAP) sample and the lowest for the third (59 DAP) sample and with the fourth (71 DAP) sample also returning a relatively low value. While trends were similar, the magnitude of the differences between the highest and lowest DT varied with DT$_{10}$ > DT$_{50}$ > DT$_{90}$. This is a common characteristic of dissipation studies that do not exhibit relatively long initial induction periods.

A likely explanation of observed DT decreases associated with increases in the DAP for spray application and soil sample collection was adaptation of the community of soil organisms contributing to tebuconazole degradation and development of accelerated degradation conditions. This is consistent with a report for another triazole fungicide, difenoconazole (Thom et al., 1997). The DT$_{50}$ decreased about two times following a single pretreatment about 1 mo before sample collection. The DT$_{50}$ in the sample collected after our second (43 DAP) tebuconazole application was about 1.5 times lower than for the sample collected after the first (28 DAP) application (Table 2). The decrease in DT$_{50}$ after the third application (59 DAP) was nearly eightfold. At this point
degradation conditions appeared to stabilize. The DT$_{50}$ for the sample collected after our fourth (71 DAP) was about two times higher than the sample after the third (59 DAP) application. No explanation is available for the apparent DT$_{50}$ increase with the exception of spatial variability in soil conditions. Walker et al. (2001) indicate that the relative standard deviation for pesticide DT$_{50}$ in soil samples collected from a single field can be expected to be 40% or greater. The magnitude of difference was approximately equal to the variation with our last two soil samples (59 and 71 DAP).

The conclusion that an adapted community of tebuconazole-degrading organisms developed was supported by comparison of treated and spiked control plot soil sample incubation results. The DT$_{10}$ and DT$_{50}$ values were 4 and 25 d, respectively, for both the first (28 DAP) and second (71 DAP) spiked controls. The consistency in results was an indication of the stability of degradation conditions until the soil microbial community was stimulated by tebuconazole treatment. The DT$_{50}$ for the fourth (71 DAP) treated soil sample was about two times lower than the value obtained for the soil sample collected on DAP 71 from control plots and spiked before incubation. The DT$_{50}$ for the first (28 DAP) sample collected from treated plots was about 1.6 times greater than the corresponding spiked control plot soil sample. The observed shift in dissipation kinetics is indicated in Fig. 2 and 3.

It was anticipated that values for first (28 DAP) treated sample and both (28 and 71 DAP) spiked control plot samples would be approximately equal since there was no tebuconazole prior treatment. A possible explanation of why faster dissipation was observed in the spiked samples was that the initial tebuconazole concentration was about two times lower when compared with the treated sample (Table 2). However, this argument is not supported by the observation described above that the spiked soil samples’ $t_{1/2}$ values were within the range of values computed for a prior study on differences in temperature were taken into account. Initial tebuconazole concentration for the published study was about two times greater than in spiked control plot samples in this study.

Alternatively, the apparent difference in percent dissipation times between the field-treated and laboratory-spiked samples may have been due to the difference in fungicide application technique. The spikes were prepared by mixing very fine sand coated with the fungicide with the soil. This likely resulted in more effective mixing with the soil matrix and greater bioavailability than in the field-treated scenario where soil was collected after spraying with the formulated commercial product. The fungicide after field treatment was adsorbed and or crystallized on the soil particles varying in size and shape. When these particles were mixed with bulk soil by sieving before incubation, mixing was likely less effective than in the laboratory spiking. Further work is needed to clarify what may be an important consideration when field and laboratory dissipation results are compared. Possible impact of formulation of field-applied pesticides may also merit investigation although studies that compared soil degradation rates of formulated and unformulated tri-ticonazole and trifluralin did not show appreciable differences (Jolley and Johnstone, 1994; Beigel et al., 1999).

### Degradate Behavior in Laboratory Incubations

Degradates 1, 2, and 4 were detected (Fig. 1) with structures of Degradates 2 and 4 confirmed by analysis of reference standards. Failure to detect Degradate 3, the γ-hydroxy acid likely formed by oxidative cleavage of the chlorophenyl group, may be explained by its rapid conversion to the corresponding γ-lactone, Degradate 2. In all incubations, Degradate 2 represented 92 to 97% of total degradates when incubations were terminated (63 d, Table 3). Although concentrations were generally low (<1%) relative to the mass of the parent dissipated, a trend to accumulation of the other degradates (1 and 4) was also indicated. However, differences in the behavior of these degradates were difficult to assess due to the uncertainty associated with the large number of concentration measurements that were near the limit of detection. Thus, the discussion below is limited to the sum of all degradates of which typically >92% was represented by Degradate 2.

In all incubations, with the exception of the fourth (71 DAP) field-treated soil sample, total degrade concentration increased steadily to its maximum when incubations were terminated. This is shown for the first (28 DAP) spiked and field-treated soil samples collected in Fig. 3. The second (43 DAP) and third (59 DAP) treated soil samples exhibited more rapid accumulation and higher total degradates levels. A fourfold increase in the relative molar percent of total degradates to the initial tebuconazole concentrations was observed when end-of-incubation results were compared between the later samples and the one collected after the first spray (Table 3). In the case of fourth (71 DAP) field-treated sample very rapid accumulation of total degradates followed by decay at the end of the incubation (Fig. 4) explains the decrease in degrade concentration observed.

In sum, degrade behavior provided another indication of adaptation and development of accelerated degradation conditions and suggests microbial community evolution during the growing season. The rate of degrade formation in field-treated samples increased with each successive tebuconazole application. Degradate accumulation kinetics was essentially the same when the two spiked control plot samples were compared.

Finally, total degrade concentrations when incubations were terminated were 16 to 66% of initial tebuconazole concentrations (molar) (Table 3). Though this indicates some potential for accumulation, the decrease in the relative amount of Degradate 2 at the end of the incubation of the 71 DAP treated soil samples suggests that accumulation is probably transitory.

### Comparison of Laboratory and Field Measurements

Tebuconazole concentration in soil samples collected after the four applications varied about 1.6-fold, 1.9 to 3.1 nmol g$^{-1}$, with the highest value observed after the
Table 4. Tebuconazole and degradate concentrations in soil expressed as a cumulative percent of tebuconazole applications made on 28, 43, 59, and 71 days after planting (DAP).

<table>
<thead>
<tr>
<th>DAP</th>
<th>Degradate 1</th>
<th>Degradate 2</th>
<th>Degradate 4</th>
<th>Tebuconazole</th>
</tr>
</thead>
<tbody>
<tr>
<td>28</td>
<td>~0.01</td>
<td>~0.01</td>
<td>~0.01</td>
<td>99 ± 23</td>
</tr>
<tr>
<td>43</td>
<td>0.2 ± 0.1</td>
<td>0.2 ± 0.1</td>
<td>0.5 ± 0.1</td>
<td>67 ± 16</td>
</tr>
<tr>
<td>59</td>
<td>0.7 ± 0.2</td>
<td>0.7 ± 0.2</td>
<td>1.2 ± 0.7</td>
<td>33 ± 7</td>
</tr>
<tr>
<td>71</td>
<td>0.5 ± 0.2</td>
<td>1.2 ± 0.2</td>
<td>1.1 ± 0.2</td>
<td>19 ± 3</td>
</tr>
</tbody>
</table>

were used for calculations were 29 ± 1°C (28–43 DAP), 30 ± 1°C (43–59 DAP), and 30 ± 1°C (59–71 DAP).

For the 43 DAP sample, the soil surface between rows was bare, so it was sprayed directly. Combining soil inputs associated with this spray and the predicted residues remaining from the first spray yielded a predicted tebuconazole concentration about 65% greater than measured. The 22 mm of rain received the day after the first (28 DAP) application may help to explain the difference between measured and predicted concentrations. Because the soil was bare between rows and plants, most of the rain fell directly on the soil surface. Infiltration for such storm events on Tifton soil is typically 80% of total rainfall (C.C. Truman, personal communication, 2004). Using this as an infiltration estimate and assuming equilibrium partitioning of tebuconazole between soil organic carbon and water, up to 25% of the tebuconazole reaching the soil surface during the first application (28 DAP) may have leached below the 2-cm sampling depth. In calculations, 0.4% was used as the average soil organic content and tebuconazole $K_{oc}$ (soil organic carbon–water partition coefficient) = 467 mL g$^{-1}$ (Xu et al., 1999).

Other dissipation processes that may account for the remainder of the difference were volatilization and soil photochemical transformation. Given tebuconazole’s relatively low vapor pressure (approximately $3.1 \times 10^{-3}$...
mPa; Food and Agriculture Organization of the United Nations, 1997), volatilization potential appears low, but this may be offset by extreme environmental conditions. Soil surface temperature at the study site may reach 40 to 45°C at mid-day on bare soil surfaces (T.L. Potter, unpublished data, 1999).

The canopy between rows closed between second (43 DAP) and third (59 DAP) applications; thus, the plants intercepted most of the third (59 DAP) spray. Canopy interception was estimated to be 95% for computation of the predicted concentration. The predicted tebuconazole in soil was within 5% of the measured in spite of the fact that there was a relatively large amount of rainfall and irrigation before the application (Fig. 4). The agreement between measured and predicted tebuconazole concentrations was presumably due to several factors. This included reduced leaching due to increases in plant evapotranspiration (as indicated by increased LAI) and foliar wash-off of residues remaining in the canopy from the second (43 DAP) application and their deposition on the soil.

Following the fourth (71 DAP) application, the measured soil concentration was two times greater than the predicted value. During this spray, canopy interception was again assumed high (95%) and residue in soil from prior sprays represented the bulk of the tebuconazole detected. The relatively low amount that was predicted was due to high soil dissipation rates inferred from laboratory incubation of the soil sample collected after the third (59 DAP) application. The most likely explanation of the difference in predicted in measured values was that dry soil conditions in the field inhibited degradation. There was only one 10-mm irrigation and no rainfall between the third (59 DAP) and fourth (71 DAP) tebuconazole applications. The gravimetric soil water content of bulk samples collected after these applications was 6.6 and 11.1%, respectively.

Back calculation using the first-order rate model showed that if $t_{1/2} = 37$ d, the predicted concentration would equal the fourth (71 DAP) application measured value. In turn, application of the soil water–$t_{1/2}$ adjustment function (Walker, 1974) indicated that if the gravimetric water content were 5.4%, the $t_{1/2}$ for the sample collected after third (59 DAP) application (Table 1) would be equal to the back-calculated value (i.e., 37 d). Since no data were available for tebuconazole $\beta$ in this equation, it was set equal to 0.8 (Boesten et al., 2004). The computed water content falls within the range of measured values for the bulk soil samples collected after the third (59 DAP) and fourth (71 DAP) tebuconazole applications.

Measured combined degrade concentration were assessed by comparison with predicted concentrations derived from fits of the laboratory incubation data to a two-parameter power function, $C = at^b$, where $C =$ concentration and $t =$ time, and $a$ and $b$ are constants. Relatively high $r^2$ (0.84–0.96) demonstrated that the equation fit the data well. The most notable result was for the sample collected after the fourth (71 DAP) application. The predicted total degrade concentration was greater than three times the measured. Dry soil conditions contributing to reduced rates of tebuconazole dissipation and degradates formation provided the most logical explanation of the poor agreement in the two values.

A final consideration regarding the degradates is that although leaching was not assessed there appears to be a potential for this to occur. This is suggested by estimates of water solubility and $K_{oc}$ for Degrade 2. The QSAR parameter estimation model, SPARC, provided high water solubility, 45 000 mg L$^{-1}$ (University of Georgia, 2004). In turn a very low $K_{oc} = 14$ mL g$^{-1}$ was calculated using the relationship, $K_{oc} = 3000 \times S^{-1/2}$ (Hornsby et al., 1996). This would make the compound susceptible to leaching.

Overall, comparison of field and laboratory results showed that although accelerated dissipation was indicated in laboratory studies, impacts may be offset by field conditions and point to a need to systematically assess impacts of varying soil temperature and water content, particularly in surface soil (0–2 cm), on pesticide dissipation rates. Results also have implications for the design, conduct, and interpretation of pesticide terrestrial field dissipation studies. Such studies are a key requirement for registration of pesticides in the United States and Europe (Barefoot and Clay, 2003). Current thinking in the United States is that study results should be used to validate the conceptual model of pesticide environmental fate developed from laboratory studies (Hendley, 2003). The relatively poor agreement between measured and predicted values in our study indicates that extension of laboratory derived kinetic data to field settings should be done with caution.

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