A MICROECOSYSTEM FOR FERTILIZER AND PESTICIDE FATE RESEARCH

B. E. BRANHAM, D. J. WEHNER, W. A. TORELLO, AND A. J. TURGEON

Abstract

A microecosystem was designed to study the behavior of pesticides, fertilizers, or related compounds applied to plant stands. The system consists of three parts: a brass base that holds the plant growth media, a glass atmospheric chamber that rests on the base, and a set of analytical traps. The brass base is fitted with a porous ceramic plate so that tension can be applied to the water in the growing media. Air enters the bottom of the glass atmospheric chamber and exits through the top into appropriate trapping systems to recover volatilized pesticides, ammonia, or metabolized {superscript}14CO{subscript}2 from labeled compounds. A port at the base of the chamber allows collection of leachate. The microecosystem was evaluated by applying N sources or a pesticide to intact turfgrass profiles and monitoring the fate of the applied compound. Leaching and volatilization losses of N ranged from 0 to 17% and 0.1 to 17% of the applied N, respectively, depending on N source, soil conditions, and whether tension was applied to the base of the system. Three weeks after the application of radiolabeled diazinon [O,O-diethyl-O-(2-isopropyl-4-methyl-6-primidinyl) phosphorothioate] to a turf, 47% of the label remained in the form of the parent compound, 22% had been metabolized and lost as {superscript}14CO{subscript}2, 1% had leached through the profile, 2% had been lost through volatilization, and 28% remained in the soil as a metabolite or in unextractable compounds. The microecosystem has proven to be an invaluable tool for turfgrass research and should be useful for fertilizer and pesticide fate studies with other crops.

Additional index words: Model ecosystem, Ammonia volatilization, Diazinon.

MICROECOSYSTEMS are useful tools for studying the fate of pesticides, fertilizer elements, or related compounds applied to plant stands. The advantages of using these systems over field studies are that they are closed, allowing for total accountability of a compound and its metabolites and they facilitate studies with radioactive or potentially toxic materials.

The first microecosystem designed to follow the movement of pesticides and metabolites through a food chain was developed by Metcalf et al. (10), and consisted of an aquatic-terrestrial system. Cole et al. (4)
developed a terrestrial microecosystem composed of a 19 L widemouthed bell jar containing 400 g of vermiculite, 50 corn (Zea mays L.) seed and a five-level food chain. A pesticide was applied and the airstream was sampled periodically for volatilized pesticide. At the end of the experiment, the top level of the food chain was analyzed for the concentration of the pesticide in various organs of the body. Nash et al. (12) and Gillett and Gillett (7) have developed microecosystems, however, neither system attempts to simulate field drainage.

The microecosystem presented herein was designed to allow simulation of field drainage conditions by including a porous ceramic plate under the growing media (1). The microecosystem was tested by applying fertilizers and pesticides to intact turf profiles and monitoring the fate of the applied compounds.

**Materials and Methods**

The microecosystem consists of three components: base, atmospheric chamber, and analytical trapping system. Eight systems are housed in a 7.3 X 3.7 m controlled environment chamber. Temperature inside the microecosystems is monitored with thermocouples.

**Microecosystem Base**

The microecosystem base that holds soil or other growing media (Fig. 1) was constructed from 32 mm thick brass plate with dimensions of 208 X 319 X 79 mm (L X W X D). Strips of brass 3.2 mm thick and 9.5 mm in width were soldered along the inside edge of the media base to provide a ledge upon which the porous ceramic plate rests. The ledge also forms a 3.2 mm deep free space on the bottom of the base. The ledge was covered with a 1.6 mm thick foam tape to provide an even seal for the porous ceramic plate. The porous ceramic plate (Soil Moisture Equip. Co.3) has dimensions of 306 X 295 X 13 mm and a bubbling pressure of 50 kPa. The plate was placed on the ledge and centered to provide a 3.2 mm channel along each of the four sides. An epoxy mix (Soil Moisture Equip. Co.) was layered into the channel to seal the plate into the base. A serrated hose fitting was soldered onto a corner of the base at the level of the free space to permit drainage. The hose fitting was connected to the analytical trapping system by 9.6 mm ID teflon tubing.

Along the top outside edge of the base an aluminum “Z” track was attached. The “Z” track formed a seat upon which the atmospheric chamber rested.

**Atmospheric Chamber**

The atmospheric chamber (Fig. 2) 311 X 319 X 356 mm (L X W X H) was constructed from 6.4 mm plate glass. The glass was cut and drilled by the Arrow Glass Co. of Urbana, IL and assembled in the laboratory using a silicone rubber sealant. Evenly spaced 6.4 mm air intake holes were drilled near the bottom of each glass plate. A total of 18 intake holes were drilled per chamber. The glass section which enclosed the top of the chamber had four 19 mm diam holes drilled on the corners of a centered 76 mm square. In the center of the square was a 11 mm hole. A glass manifold (114 X 114 X 38 mm) (L X W X H) was sealed to cover the five holes of the top glass section. A 11 mm diam hole was drilled in the center of the manifold which lined up directly over the hole of the same size on the glass chamber. These two holes formed the entrance for a 32 mm pipe to deliver water to the nozzle assembly. The nozzle assembly consisted of a full-cone spray nozzle (cat. no. 1/4 TGO.3, Spraying Systems Co., North Ave. at Schmale Rd., Wheaton, IL 60187) connected to a diaphragm body (cat. no. 8360, Spraying Systems Co.). The nozzle assembly was attached to the 32 mm pipe with the use of a reducing coupler. Another hole 12.7 mm in diameter was offset by 25 mm from the center of the manifold top. A 9.5 mm serrated brass hole fitting was cemented to the glass with litharge [260 g PbO ground in a mortar with 100 mL of diluted glycerine (2/1, glycerine/H2O)]. The brass hose fitting served as the air outtake port. The hose fitting was connected to the analytical trapping system by 9.5 mm ID teflon tubing.

**Analytical Trapping System**

The analytical trapping arrangement can be varied to meet the requirements of the particular compound being studied. Two different types of trapping systems were used to collect either pesticide-related volatiles or ammonia.

The trapping system for pesticides consisted of a solid adsorbent (chromosorb 101, Alltech and Assoc.) and was adapted from the method of Pellizzari et al. (13). An open-ended 7 mm diam glass tube was filled with 5 g of adsorbent and placed at the exit of the atmospheric chamber to trap...
any volatilized pesticide. A second set of traps was used to
scrub the airstream for any volatilized \(^{14}\)CO\(_2\) resulting from
the microbial degradation of the pesticide. Two flasks were
filled with 375 mL of 1 \(M\) NaOH and the airstream was bubbled
through the two traps.

In order to trap volatilized ammonia, the above trapping
scheme was modified by removing the solid adsorbent trap
and by using a 4.5 \(M\) boracic acid solution to trap ammonia
in the two bubbling tubes.

Following either scheme, the airstream then passed through
a desiccant and into an air flow meter (cat. no. VPAB-65-BV, Dwyer Instruments, Inc.) to monitor and regulate air flow.
The air flow meter was connected to a manifold, common
to all microecosystems, that was in turn connected to a
vacuum pump.

**Trapping Efficiency Tests**

Tests were conducted to evaluate the efficiency of trapping
\(^{14}\)CO\(_2\), diazinon, and NH\(_3\). A weighed amount of Ba\(^{14}\)CO\(_2\),
specific activity 1.16 \(\times\) \(10^2\) DPM/mg, was placed in a 10
mL beaker on a ringstand inside the atmospheric chamber.
A buret slowly dripped 5.9 \(M\) perchloric acid into the beaker
releasing \(^{14}\)CO\(_2\) over a period of 2 h. Four bubblers connected
to series were filled with 375 mL of 1 \(M\) NaOH and the airstream
(air flow 3.3 \(\times\) \(10^{-3}\) m\(^3\) s\(^{-1}\)) from the chamber was
swept through the solutions for 24 h. The solutions were
collected and the radioactivity assayed by adding 3.5 mL of
the NaOH solution to 15 mL of Aquasol (New England Nu-
clear). The samples were replicated three times and counted
using a Packard Tri-Carb liquid scintillation counter. The
solid adsorbent trap was evaluated by allowing a known
amount of diazinon to volatilize in the atmospheric chamber,
passing the airstream (air flow 6.6 \(\times\) \(10^{-3}\) m\(^3\) s\(^{-1}\)) through
the adsorbent, and then extracting the diazinon from the
adsorbent and assaying the amount of material trapped.

The trapping efficiency for NH\(_3\) was conducted as reported
by Torello et al. (16) by dripping 1.0 \(M\) KOH into a 50 mL
beaker containing a known amount of \((NH_4)_2SO_4\) within
each atmospheric chamber. Ammonia liberated and trapped
within the boric acid traps was then quantified by titrating
with 0.0231 \(M\) H\(_2\)SO\(_4\).

**Diazinon and Fertilizer Applications to Turf**

The microecosystem was evaluated by applying two dif-
ferent N sources and the pesticide diazinon to turf and mon-
toring the fate of the applied compounds. In all of these
evaluations, the intact turf profiles consisted of Kentucky
bluegrass (Poa pratensis L.) growing on a Flanagan silt loam
(fine, montmorillonitic, mesic Aquic Argiudoll). No volatil-
ized ammonia was detected from nonfertilized controls run
at the beginning of the research project. All samples were
taken directly from the field and placed in the microecos-
systems, allowed to equilibrate for 1 to 3 days, and then fer-
tilized or treated with diazinon. A brief outline of the pro-
cedures for these experiments is given below. Detailed
accounts of the procedures are given by Torello (14) and
Branham (2).

Several different fertilizer treatments were applied to turf
profiles and leaching and volatilization of N monitored. Am-
nnonium nitrate (33-0-0) at the rate of 293 kg N ha\(^{-1}\) and
urea (46-0-0) at the rates of 49 and 293 kg N ha\(^{-1}\) were
applied to turf subjected to a soil moisture tension of -40
kPa and irrigated with 5 mm water every 3 days. The first
irrigation came 3 days after the application of the fertilizer
treatments. The temperature inside the microecosystems was
21°C with a 12 h daylength and a light irradiance of 0.204
W m\(^{-2}\). Air flowed through the atmospheric chamber at the
rate of 6.6 \(\times\) \(10^{-3}\) m\(^3\) s\(^{-1}\) resulting in 6.7 volume changes
per hour.

The microecosystems were also used with the vacuum sys-
tem turned off so that no tension was applied to the soil (i.e.,
controlling grown plants). In this case, urea was applied at
rates of 49 and 245 kg N ha\(^{-1}\) to turf growing in a thatch
layer without the accompanying soil. The thatch layer was
25 mm thick and had been stripped from the soil surface
in the field and placed in the media base. The plant-thatch
layer was irrigated with 600 mL distilled water and then the urea
was applied. There were no subsequent irrigations.

Volatilized NH\(_3\) was collected for a 10 to 21 day period
in boric acid traps and assayed as described by Torello et al.
(16). The leachate was analyzed for total N by the Kjeldahl
procedure. From one to three replications of each treatment
were run.

Granular radiolabeled diazinon was applied to turf at the
rate of 4.9 kg ha\(^{-1}\) of active ingredient with enough label to
provide 1.48 \(\times\) \(10^{4}\) Bq of activity per microecosystem. The
temperature inside the microecosystems was held at 21°C
for 12 h with a light irradiance of 0.204 W m\(^{-2}\) and a night
temperature of 16°C. Irrigation (5 mm per application) was
applied every 4 days starting immediately after pesticide ap-
lication and tension was applied to the base of the micro-
ecosystem (-40 kPa). Air flowed through the systems at a rate of 6.6 \(\times\) \(10^{-3}\) m\(^3\) s\(^{-1}\). Volatilization, leaching, metab-
olism, and incorporation of the label into nonextractable soil
compounds was monitored. Branham (2) presents a detailed
account of the procedures for extraction of the parent com-
 pound from the soil as well as the separation of the metab-
olites by liquid chromatography. The volatile compounds
and \(^{14}\)CO\(_2\) released through pesticide metabolism were trap-
ped as outlined in the trapping efficiency section. This ex-
periment lasted 3 weeks.

**Results**

An important aspect of the microecosystems is the
analytical trapping arrangement. These traps can be
changed to accommodate the particular compound
under investigation. Table 1 contains a summary of
the trapping efficiencies for the various compounds
that were released in the microecosystems. As ex-
pected, the released compounds were trapped with
varying efficiencies. There were advantages to using
the solid adsorbent for volatile organic compounds as
it required less maintenance than wet chemical traps.
The chromosorb 101 does not have much retentive
capacity for water, unlike other adsorbents, e.g.,
molecular sieves, which proved to be inadequate as trap-
ing materials. Also, the solid adsorbent allowed the
air flow rate to be increased over the flow rate used
with the wet chemical traps. A higher flow rate min-
imizes water condensation on the sides of the atmos-
pheric chamber.

The losses of fertilizer N from the microecosystems
through volatilization and leaching are presented in
Table 2. These values, which are presented to
demonstrate the use of the microecosystem, were recorded
at various times during a research project on N volat-
ilization (14) and as such cannot be directly com-
pared with one another. However, the trends pre-
sent here agree with published results of N volatilization experiments in regard to source and rate effects (8, 16, 17) on NH$_3$ losses from turf. Essentially, N was lost through NH$_3$ volatilization from turf fertilized with ammonium nitrate and volatilization losses from urea-treated turf increased with N application rate. The leaching loss for ammonium nitrate was higher than for the same application rate of N from urea. The overall low levels of N leached are a reflection of the nutrient holding capacity of the Flanagan silt loam and the fact that the turf samples for our study were selected from an area that had not been fertilized for 8 years. The N volatilization from the turfs that were not subjected to suction were higher than where tension was applied. This was probably due to the fact that more N remained in the turf profile in the absence of leaching, the high level of urease found in turfgrass thatch (15), and the slow drying that occurred over the course of the experiment since there were no irrigations after fertilizer application.

The results of the experiment with diazinon (Table 3) indicated that after 3 weeks, 47% of the applied label remained in the form of the parent compound, 22% had been metabolized and lost as $^{14}$CO$_2$, 1% had leached through the soil profile, 2% had been lost through volatilization and 28% remained in the soil as a diazinon metabolite or in unextractable compounds. The half-life of diazinon in soil has been reported by a number of authors (3, 5, 6, 9, 11) to be between 7 and 56 days depending on conditions of the experiment. Our results using the microecosystem fall within the range of previously published results.

**Discussion**

Because the atmospheric chambers are constructed from glass, which is opaque to ultraviolet light, no measurement of photodecomposition is possible using the microecosystems. However, most other avenues of pesticide fate can be studied. Glass was used for the atmospheric chambers so that volatile compounds would not be absorbed by the walls. No attempt was made to control the relative humidity of the air entering the atmospheric chamber.

One of the unique features of this microecosystem is the presence of the porous ceramic plate in the bottom of the media base. The plate allows a suction to be applied to the water in the soil or other growing media. Because the soil base is only 50 mm deep, caution must be used in interpreting leaching data. For herbicide research the depth is adequate because very few weed seeds germinate below 50 mm in the soil. Herbicide that is leached out of the 50 mm zone would no longer be effective. Also, cool-season turfgrasses which have a fairly shallow root system under mowed conditions lend themselves to this depth. No attempts were made to determine the amount of nutrient or radioactive label that may be adsorbed to the ceramic plate. The use of a balance sheet approach in studying applied compounds will allow the researcher to determine if this is important.

In summary, the microecosystems have proven to be useful for carrying out studies on the fate of pesticides and fertilizers applied to turfgrass stands. They should be adaptable to other plant species by changing the system dimensions and trapping arrangements to suit the goals of the researchers. Experimental conditions, such as soil moisture, can be varied so that simple comparisons between soil type, plant species, etc. can be easily accomplished.

**Acknowledgments**

The authors wish to thank Drs. L. A. Spomer, R. F. Nystrom, and Mr. Howard Friese for their help with this project.

**References**


**Table 3. Fate of radiolabeled diazinon applied to turf growing on a Flanagan silt loam.**

<table>
<thead>
<tr>
<th>Percent of applied label</th>
<th>Week</th>
<th>Lost as $^{14}$CO$_2$</th>
<th>Leached</th>
<th>Volatilized</th>
<th>As unextractable compounds</th>
<th>Metabolite</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
<td>0.3†</td>
<td>0.6</td>
<td>1.8</td>
<td>16.2</td>
<td>6.1</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>4.4</td>
<td>0.9</td>
<td>2.0</td>
<td>18.3</td>
<td>7.5</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>22.2</td>
<td>1.2</td>
<td>2.0</td>
<td>23.4</td>
<td>4.0</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
| † 8-isopropyl-4-methyl-6-hydroxyprazimidine. | Values represent mean of two replications.