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MOBILITY AND DISSIPATION STUDIES OF METRIBUZIN, ATRAZINE AND THEIR METABOLITES IN PLAINFIELD SAND USING FIELD LYSIMETERS

BRUCE T. BOWMAN
London Research Centre, Agriculture Canada, 1400 Western Road,
London, Ontario, Canada N6G 2V4
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Abstract—Mobility and persistence of metribuzin (Lexone DF) and its metabolites (deaminated, DA; diketo, DK; deaminated-diketo, DADK) were studied using 75-cm x 15-cm field lysimeters packed with Plainfield sand, and subjected to two moisture regimes (rainfall, supplementary watering). Atrazine was applied to all lysimeters as an internal reference. Each lysimeter set consisted of 24 lysimeters, divided into two moisture treatments of six pairs each. Effluent was monitored for metribuzin, DA, DK, DADK, atrazine and desethylatrazine. Selected core pairs were sectioned (7 x 10 cm) and analyzed to determine mobility and persistence for each chemical at weeks 1, 2, 4, 8, 12, and 21. No chemicals moved more than 30 cm, nor were they detected in the effluent of cores receiving rainfall. In cores receiving supplementary watering, substantial amounts of metribuzin moved more than 30 cm by week 2 and eluted on 21 occasions throughout the study. The DADK and DK were much more mobile than metribuzin in the lysimeters, eluting more frequently than metribuzin. Metribuzin and metabolites were considerably more mobile than either atrazine or desethylatrazine in Plainfield sand. Metribuzin disappearance closely followed first-order kinetics with $t_{1/2}$ values of 3.08 and 2.04 weeks, respectively, for the rainfall and supplementary watering treatments. The primary degradation pathway for metribuzin was through the DK intermediate rather than the DA intermediate.

Keywords — Leaching Soil core Metabolite Degradation

INTRODUCTION

Metribuzin is an asymmetrical triazine herbicide that has been shown to provide effective pre- or postemergence control of broad-leaved and grassy weeks in a variety of vegetable crops, as well as in soybeans and sugar beets [1,2]. Metribuzin has a water solubility of 1.2 g/L (20ºC) [2] and is not strongly adsorbed by most soils. K_{OC} values (soil-water partitioning coefficient [K_d] normalized to organic carbon fraction) calculated from the literature ranged from 38 [3] for surface horizons to 275 [4] for subsoil horizons, based on K_d units of $\mu g^{(1-N)}\ g^{-1} ml^N$, where N was the Freundlich exponent. Harper [4] found that clay content and soil pH had more influence over metribuzin adsorption than soil organic matter content, which may explain the rather high K_{OC} values for his subsoils. Harper also found that metribuzin adsorption was easily reversible on mineral soils, with only 5% of the original amount remaining after three desorption cycles. In contrast, Sharom and Stephenson [3] and Peter and Weber [5] reported that soil organic matter did influence metribuzin adsorption.

Sharom and Stephenson also found that metribuzin mobility was inversely related to organic matter content and that six desorption cycles were required to remove most of the metribuzin from their soils, while 10 desorption cycles still left substantial amounts on muck soils. Both Nicholls et al. [6] and Peter and Weber [5] reported that metribuzin was considerably more mobile in light textured soils than was atrazine.

Metribuzin degradation in soil appears strongly influenced by microbial activity [3] and by those soil factors that influence microbial activity, such as temperature, moisture and nutrient levels. Reported 50% disappearance times for metribuzin were in the 2.5- to 9-week range [7,8]. The apparent reaction order reported for metribuzin degradation in soil has varied widely (0.17-3.93) [7,9]. Allen and Walker [10] obtained a better linear relationship when the logarithm of metribuzin concentration was plotted against cumulative respiration than when it was plotted against time. They concluded that metribuzin degradation rates were con-trolled by a combination of microbial activity, availability of herbicide in the soil solution and some component of the particle size distribution (sand, silt, or clay). Moorman and

Harper [9] concluded that both metribuzin concentration and microbial biomass contributed to the second-order degradation reaction observed in their studies.

There are three major breakdown metabolites of metribuzin: deaminated metribuzin (DA), diketometribuzin (DK), and deaminated-diketo metribuzin (DADK). Possible breakdown pathways are illustrated in Figure 1. There is no consensus in the literature as to whether the degradation route from metribuzin to DADK is a two-step process through either DA or DK, as shown by Thornton and Stanley [11], or whether a direct conversion is possible. Webster and Reimer [12] suggested a direct conversion link from metribuzin to DADK, although they did not explain how this two-stage process could directly occur, nor did they show any pathways existing from DA or DK to DADK. Pape and Zabik [13] have shown the DA metabolite to be the major photodegradation product in water, and it is not clear whether DA can be generated by alternate processes in soil. Sharom and Stephenson [3] reported that after a six-month incubation of [^{14}C]metribuzin in a silt loam soil, the distribution of radioactivity was 10% metribuzin, 20% DK, 20% DA and 50% DADK, with apparently no other unidentified components. Moorman and Harper [9] also used ^{14}C-radiolabeled metribuzin and reported that after 91d, up to 14% of the soil-applied ^{14}C was unidentified chloroform-soluble metabolites. Webster and Reimer [12] found considerably less DA than either DK or DADK in degradation studies using fine sandy loam. There appears to be no information in the literature regarding the mobilities of metribuzin metabolites in soil.

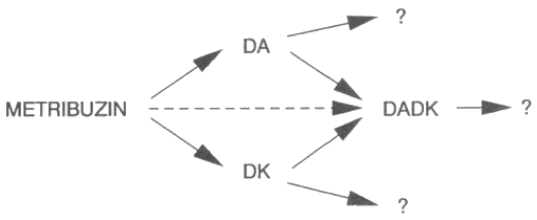

Fig. 1. Metabolic pathways for metribuzin in soil; DA = deaminated, DK = diketo, DADK = deaminated-diketo metribuzin.

The objective of this study was to investigate the mobility and persistence of metribuzin and its three major metabolites in Plainfield sand using field lysimeters, and to compare their behavior with that of atrazine, whose mobility and persistence characteristics have already been well defined[14].

MATERIALS AND METHODS

Compounds and application rates

Ciba-Geigy Canada Ltd. supplied analytical-grade standards (99.7% purity) of atrazine and desethylatrazine. Du Pont Canada supplied analytical standards of metribuzin and three of its metabolites—DA, DK, and DADK. Commercially formulated products were applied to the field lysimeters as follows: atrazine— Aatrex 480L; metribuzin — Lexone DF (granular). Atrazine was appliedat the maximum recommended rate of 2.25 kg active ingredient (a.i.)/ha (3,980 µg/core) [1], and thesame rate was used for metribuzin. Both atrazine and metribuzin formulations were uniformly applied by pipet (10-ml aqueous suspensions) to the surface of each soil core, at least 1 cm away from the cylinder walls.

Experimental setup

Details of the experimental setup, the leaching protocol and analytical procedures have been previously described [14]. Briefly, about 17 kg (dry wt.) of Plainfield sand (87.5% sand, 6.5% silt, 6.0% clay, 1.5% organic matter) was loaded into each lysimeter, producing a 70-cm x 15-cm soil core packed to within 5 cm of the top of the stainless steel cylinder. Lysimeters were buried in an outdoor sandbox (embanked on three sides with soil for temperature stability) to within 5 cm of the top of the unit. Approximately 10 to 12 L of water applied to each lysimeter over the two-week periodpreceding pesticide application on May 9, 1989 aided uniform packing of the cores. Effluent was removed from the collection beaker below the soil core via a stainless steel tube on the day following each rainfall or watering. One-half of the lysimeters received only rainfall, whereas remaining lysimeters received rainfall plus supplementary watering,simulating a 50-mm rainfall on days 2 and 9. After week 2, 25-mm water applications simulating an irrigation protocol were made: (a) 2 d following <10 mm rainfall, (b) 3 d following 10 to 25 mm rainfall and (c) 4 d following >25 mm rainfall, or the last watering. Effluent from each lysimeter was centrifuged at 40,000 g for 10 min and analyzed for the respective chemical by HPLC or by GLC. One pair of cores from each moisture regime was retrieved from the field at 1, 2, 4, 8, 12, and 21 weeks, and frozen for subsequent sectioning (7 x 10 cm) and residue analysis.

Soil core extraction and analysis

Glass sample bottles (450 ml, screw-capped) containing 100 g of treated soil and 80 ml of extraction solvent (90:10 HPLC-grade methanol-water) were heated in a circulating water bath to 45ºC for 10 min, sonicated for 10 min, then tumbled for 30 min at 40ºC in a temperature-controlled cabinet. Samples were left to settle for 30 min, then the supernatants were decanted into vacuum Buchner funnels and filtered. Remaining soil slurries in the sample bottles were extracted a second time and added (total transfer) to the contents of the first extraction in the Buchner funnels. The soil cake in each Buchner funnel was finally washed three times with 20-ml aliquot of extraction solvent. Filtrates were quantitatively transferred to 250 ml volumetric flasks, made to volume with methanol and centrifuged at 40,000 g for 20 min to remove sediment traces before HPLC or GLC analysis. Extraction efficiencies for metribuzin and its metabolites exceeded 95% for freshly spiked soil samples.

Atrazine, desethylatrazine, and metribuzin analyses were done by GLC, using the following operating parameters: column, J&W DB-17, 0.53 µm x 15 m (megabore, 1.0-µm film); carrier gas, ultra-pure helium (15-20 ml/min); detector, nitrogen-phosphorus, 260ºC; H_2 flowrate, 1 to 3 ml/min; injector temperature, 240ºC; injection volume, 2.0µl (autoinjected); temperature programs, 165ºC initial, 10ºC/min to 250ºC. Atrazine eluted near 195ºC; metribuzin eluted near 220ºC. Minimum detectable concentrations for atrazine, desethylatrazine, and metribuzin were, respectively: 2 to 5 µg/L, 5 to 10 µg/L, and 5 to 10 µg/L.

The three metribuzin metabolites—DA, DK, and DADK—were analyzed by HPLC. The operating parameters were: column, Waters Resolve radial compression cartridge, 5 mm *x* 10 cm, reversed-phase C_{18} 10-pm particle size, or Waters Resolve column, 15.0 cm x 3.9 mm, reversed-phase C_{18} 3 µm; pump speed, 0.8 ml/min; injection volume, 40µl; detector wavelength, 240 nm for DA and either 208 or 260 nm for DK and DADK; mobile phase, 40/60 acetonitrile/water for DA, 30/70 acetonitrile/water for DA and DADK. The minimum detectable concentration was 15 to 30 µg/L. All mobile-phase solvents for HPLC analyses were degassed by vacuum filtering through 0.7 µm glass microfiber filters. Peak areas of HPLC and GLC responses for the analytes were determined by a computer-based integrator using external standard solutions of the same solvent composition.

RESULTS AND DISCUSSION

Effluent data

No traces of metribuzin, atrazine, or their metabolites were detected in the effluent from lysimeters receiving only rainfall throughout the study, despite substantial rainfall during the first four weeks (Table 1). Small amounts of metribuzin eluted from 7 of the 12 lysimeters receiving supplementary watering by May 15, compared with one atrazine elution. Initial DK and DADK elutions occurred during week 10 (mid-July) and continued for several weeks. The total number of recorded elutions were as follows: metribuzin, 21; DA, 0; DK, 25; DADK, 29; atrazine, 8; desethylatrazine, 0. Total amounts eluted for each chemical were generally less than *20 µg* (0.005 fraction of applied) for a given lysimeter.

Mobility and persistence data

No chemicals were detected below 30 cm in lysimeters receiving rainfall throughout the study (Fig. 2B; maximum penetration bar graphs indicate only presence or absence of a chemical).Under supplementary watering, traces of atrazine and metribuzin quickly reached 60 cm by week 1 (Fig. 2A), whereas more substantial amounts of metribuzin moved at least 30 cm (Fig. 3). By week 2, metribuzin concentration increased in segment 4 (40 cm maximum). Maximum penetration for atrazine and metribuzin receded to 40 and 50 cm, respectively, from weeks 2 to 8 (Fig. 2A), by which time only 10 to 15% of applied metribuzin remained (Fig. 4). Traces of the metabolite DA reached 30 cm after one week and 40 cm after four weeks, but by week 12 had disappeared from the soil cores (Fig. 2A). Traces of DK and DADK reached 30 cm and 20 cm after two weeks, respectively, and both reached 50 cm by week 4. Traces of both DK and DADK remained

Table 1. Cumulative precipitation amounts received by lysimeters under both natural rainfall and supplementary watering treatments, May 10 to October *3,* 1989

	Cumulative precipitation (mm)	
Week	Rainfall	Watering
1	6.0	107.7
2	23.0	124.7
4	95.0	222.1
8	133.5	362.3
12	149.5	505.4
21	226.5	811.2

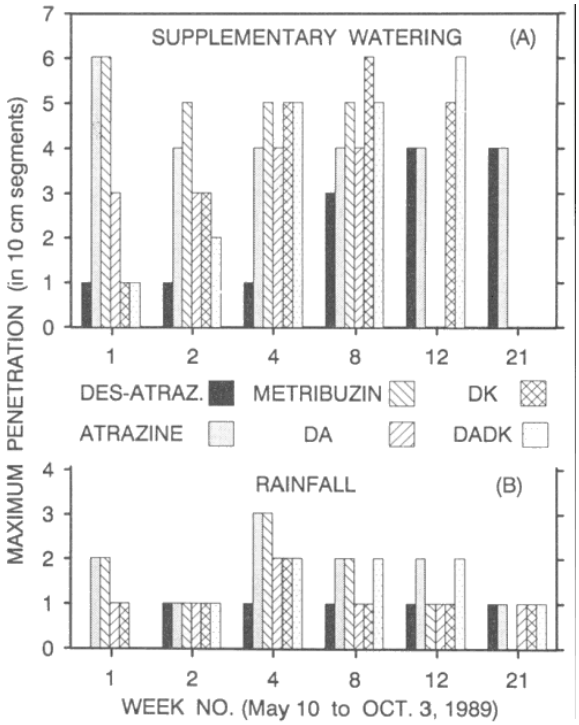

Fig. 2. Maximum penetration of atrazine, desethyl-atrazine (Des-Atraz.), metribuzin, DA, DK, and DADK in Plainfield sand lysimeters: **(A)** supplementary watering, **(B)** rainfall.

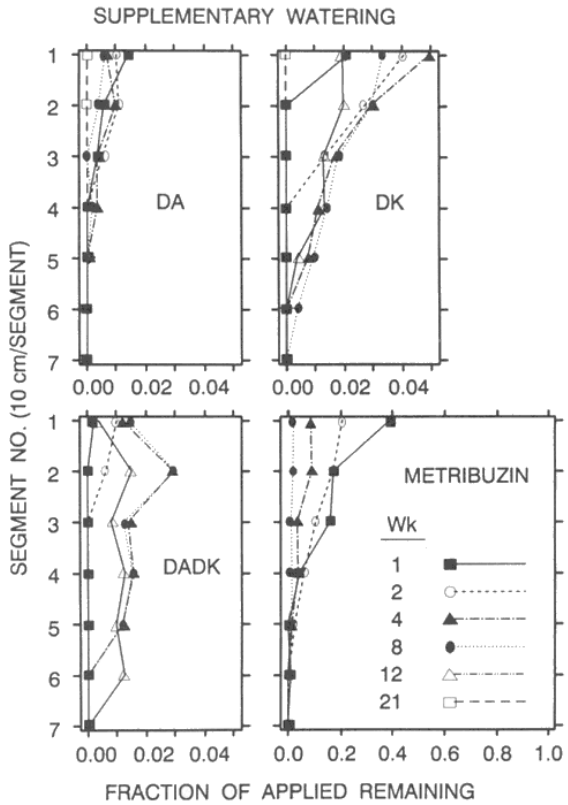

Fig. 3. Mobility profiles for metribuzin, DA, DK and DADK in Plainfield sand lysimeters under supplementary watering.

by week 21 in lysimeters receiving rainfall, but not in those receiving supplementary watering (Fig. 2A). DADK appeared somewhat more mobile than DK, judging from its greater levels in lower core segments by week 2 (Fig. 3) and by its greater elution frequency. It must be borne in mind that in situ conversion of metribuzin occurred as it leached downward, and this may have influenced "apparent" relative mobilities of the metabolites.

Metribuzin degradation data (Fig. 4) agreed reasonably well with those of Webster and Reimer [12]. Considerably less DA than either DK or DADK was detected in the lysimeters, and there was a distinct time lag in the appearance of the metabolites, especially for DADK. Metribuzin degradation in this study followed first-order kinetics quite closely, as reported by Webster and Reimer [12], but differed from some other studies [7,9]. Rate constants were 0.225 $week^{-1}$ (R^2 = 0.995) for cores receiving rainfall (12 weeks data) and 0.338 $week^{-1}$ (R^2 = 0.998) for those receiving supplementary watering (8 weeks data). Half-life ($t_{1/2}$) values for metribuzin were 3.08 and 2.04 weeks, under rainfall and supplementary watering, respectively. Shorter metribuzin persistence in cores receiving supplementary watering, relative to those receiving only rainfall, may reflect greater microbial activity at higher moisture levels, or perhaps some low-level redistribution to lower portions of the core or some washout from the cores. It is difficult to assess DA mobility accurately from these profiles (Fig. 3), because it occurred in amounts barely above detection limits. However, it is evident from both the effluent data and the mobility profiles that both DK and DADK were very mobile in this soil when subjected to supplementary watering.

Despite careful attempts at maintaining a mass balance for metribuzin and its metabolites in this study, approximately 50% of total metribuzin residues (including DA, DK, and DADK) had disappeared by four weeks (Fig. 4). Loss of recovery with time can be ascribed to several factors, including: (1) loss of extraction efficiency due to residue binding, primarily by soil organic matter; (2) production of unidentified metabolites; (3) degradation losses; or (4) volatilization losses. As noted above, metribuzin extraction efficiencies for freshly spiked soil samples exceeded

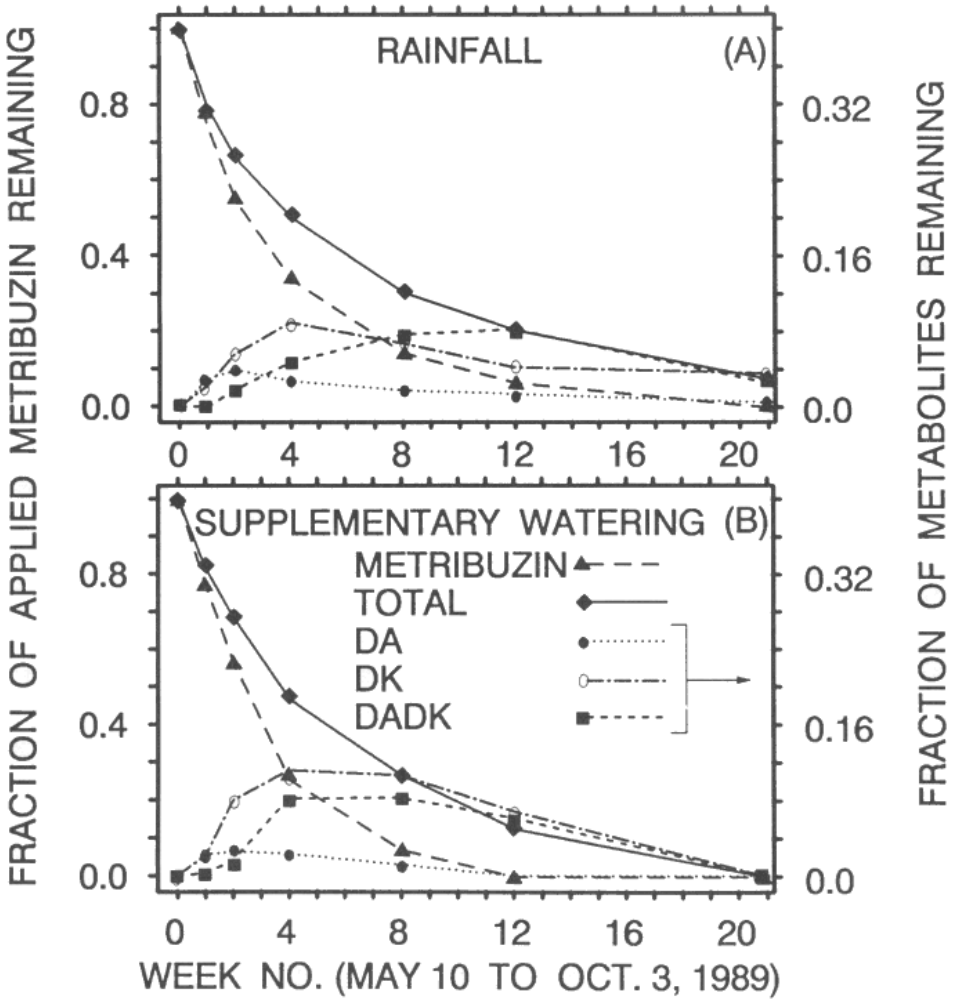

Fig. 4. Persistence of metribuzin, DA, DK and DADK in Plainfield sand lysimeters: **(A)** rainfall, **(B)** supplementary watering.

95%. As the organic matter content of this soil was only 1.5%, residue binding was not expected to be of major consequence with residue aging. Moorman and Harper [9] found that other unidentified breakdown products accounted for up to 14% of applied amounts in their study. Jensen et al. [15] found that shallow incorporation of metribuzin on both exposed and covered plots increased its half-life by twofold and threefold, respectively, relative to surface applications. They suggested that both photodecomposition and volatility decreased metribuzin persistence. Metribuzin was surface-applied in this study, and its persistence may have been reduced by those factors.

Both atrazine and desethylatrazine mobility (Fig. 5) and persistence behavior (Fig. 6) in this study were consistent with previously reported behavior [14]. Half-life values for atrazine were 4.3 and 4.9 weeks for cores receiving rainfall and supplementary watering treatments, respectively. Metribuzin (Fig. 3) was much more mobile than either atrazine or desethylatrazine (Fig. 6), but its shorter persistence, relative to that of atrazine, would tend to diminish its leaching hazard. These mobility data are consistent with the soil–water partitioning coefficients (K_d) for atrazine and metribuzin, which are 0.843 and 0.243 $\mu g^{(1-N)} g^{-1} ml^N$ respectively, where N is the Freundlich exponent. The K_d values for DA, DK, and DADK were estimated to be less than 0.1 $\mu g^{(1-N)} g^{-1} ml^N$, based on their mobilities relative to metribuzin.

The K_d values in the range below approximately 0.1 are not particularly useful in distinguishing mobility differences among compounds, due to their minimal sorption.

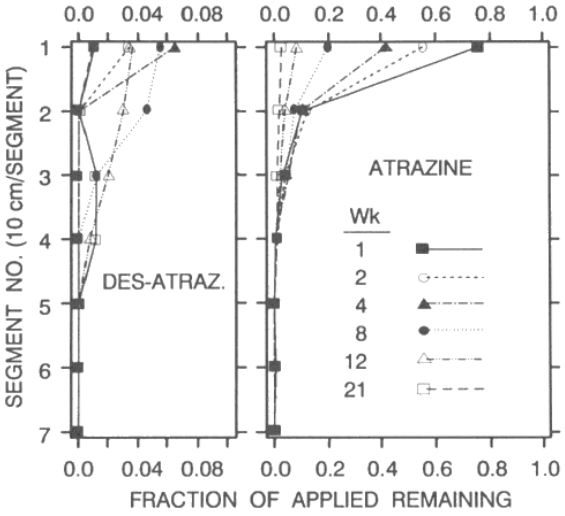

Fig. 5. Mobility profiles for atrazine and desethyl-atrazine (Des-Atraz.) in Plainfield sand lysimeters treated with metribuzin under supplementary watering.

CONCLUSIONS

Several significant findings from this study can be summarized as follows:

1. Under existing rainfall patterns of 1989, neither metribuzin, atrazine nor their metabolites leached through 70 cm Plainfield sand cores during the 21-week study, and in fact only limited chemical movement occurred. Under those conditions, a conventional field-scale study could not have assessed the leaching potential of those compounds.
2. The mobility ranking under supplementary watering for metribuzin, atrazine and their metabolites in Plainfield sand was:
   DADK $\geq$ DK > DA = metribuzin > desethylatrazine > atrazine.

   Using atrazine as an internal reference to earlier mobility studies [14,16], metribuzin and its metabolites were considerably more mobile than alachlor, metolachlor, terbuthylazine, isazofos, and atrazine, but considerably less mobile than aldicarb and its metabolites. Because DA was produced in rather small amounts, its leaching behavior was difficult to characterize.
3. Metribuzin disappearance in Plainfield sand followed first-order kinetics quite closely, unlike several other reported studies. Whereas DA levels in the cores peaked quickly by week 2, DK levels peaked at considerably higher levels by week 4. Observed DADK levels peaked after those of DK and DA.
4. Despite careful mass balance calculations, about 50% of total metribuzin residues (including metabolites) could not be accounted for after about four weeks in the lysimeters. The surface application of metribuzin may have favored increased photodecomposition or volatility losses, relative to an incorporated application.

The field lysimeter technique is an efficient way to study pesticide metabolism and mobility under natural climatic conditions. Amounts of supplementary water applied to the lysimeters are carefully controlled and repeatable from study to study(or year to year), thereby permitting consistent comparisons. It accentuates small mobility differences among chemicals, which would otherwise be observed only during wet weather and/or under an ongoing irrigation program. This technique would lend itself very well to the controlled use of radio-labeled pesticides as a means of more precisely tracking metabolic pathways and maintaining accurate mass balances in situations such as the current study, in which there were unexplained losses of applied pesticide.

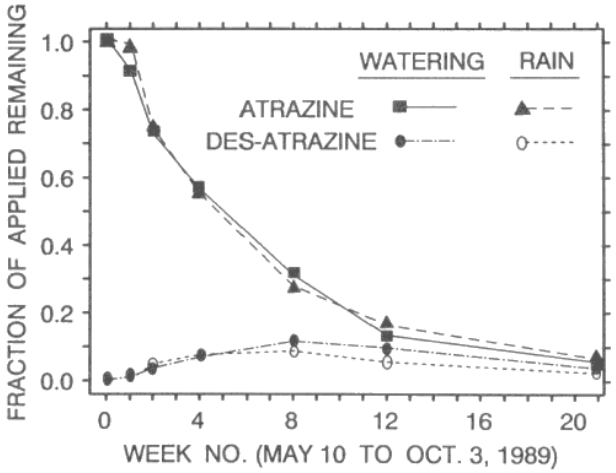

Fig. 6. Mobility profiles for atrazine and desethylatrazine (Des-Atraz.) in Plainfield sand lysimeters treated with metribuzin under both supplementary watering and rainfall.

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