Evaluating Acute Toxicity of Methyl Parathion Application in Constructed Wetland Mesocosms

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ABSTRACT: Wetland ecosystems have reduced ambient levels of various organic and metallic compounds, although their effectiveness on agricultural pesticides is not well documented. Five stations within each of two 10 × 50 m constructed wetlands (two vegetated, two nonvegetated) were selected to measure the fate and effects of methyl parathion (MeP). Following a simulated storm event (0.64 cm of rainfall), aqueous, sediment, and plant samples were collected and analyzed spatially (5, 10, 20, and 40 m from the inlet) and temporally (after 3–10 days) for MeP concentrations and for the impact of those concentrations on the aquatic fauna. Aqueous toxicity to fish decreased spatially and temporally in the vegetated mesocosm. *Pimephales promelas* survival was significantly reduced, to 68%, at the 10-m station of the nonvegetated wetlands (3 h postapplication), with pesticide concentrations averaging 9.6 μg MeP/L. *Ceriodaphnia* in both the vegetated and nonvegetated wetlands was sensitive (i.e., a significant acute response to MeP occurred) to pesticide concentrations through 10 days postapplication. Mean MeP concentrations in water ranged from 0.5 to 15.4 μg/L and from 0.1 to 27.0 μg/L in the vegetated and nonvegetated wetlands, respectively. *Hyalella azteca* aqueous tests resulted in significant mortality in the 5-m vegetated segment 10 days after exposure to MeP (2.2 μg/L). Solid-phase (10-day) sediment toxicity tests showed no significant reduction in *Chironomus tentans* survival or growth, except for the sediments sampled 3 h postapplication in the nonvegetated wetland (65% survival). Thereafter, midge survival averaged >87% in sediments sampled from both wetlands. These data suggest that wetlands play a significant role in mitigating the effect of MeP exposure in sensitive aquatic biota.

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INTRODUCTION

The efficiency of aquatic plants in removing toxic chemicals from waters as well as their effectiveness in reducing suspended sediments and nutrients from the water column has been thoroughly documented in wetland research (Wolverton and Harrison, 1973; Nichols, 1983; Catallo, 1993). Over the last two decades various researchers have reported on the structure and function of wetlands in improving water quality and providing specific properties that mitigate the effects of metals, industrial/municipal wastewater, urban storm water, and petroleum products (Jackson, 1989; Liv-

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ingston, 1989; Skousen et al., 1994; Nix et al., 1994; Detenbeck et al., 1996; Hawkins et al., 1997; Gillespie et al., 2000; Mastin et al., 2001). Fewer studies, however, have examined the fate and effects of exposures to specific pesticide (i.e., organophosphate insecticides) in constructed wetlands (Rodgers and Dunn, 1992; Moore et al., 2002; Schulz et al., 2003a). The beneficial role of wetland ecosystems in improving water quality has been established through investigations examining their potential for trapping or transforming contaminants (Cooper et al., 1994). Investigations associated with agricultural runoff may provide landowners with guidance for how to reduce pesticide levels prior to their entering critical receiving streams.

Agricultural drainage ditches are integral components of the Mississippi Delta production landscape and have been proposed as comparable substitutes for edge-of-field constructed wetlands (Moore et al., 2001). Conventional wetland attributes (e.g., plant species, soil, and a hydrologic regime) can be found in agricultural drainage ditches. The length of time water is resident in a drainage ditch, aquatic plant communities, and associated sediments that often include organic matter and fine particles facilitate the transformation of pesticides known to pose risks to sensitive aquatic communities.

The recent recognition that drainage ditches have an inherent ability to transform pesticide-associated agricultural runoff supports their use as a tool to transfer and transform contaminants, decreasing their potential to affect downstream receiving systems because of non–point source runoff (Cooper et al., 2002; 2003). Agricultural ditches receive drainage water that may have resulted from both irrigation and storm-water runoff and therefore may contain elevated sediment, nutrient, and pesticide loads, often in pulsed doses (Morton et al., 2000; Reinert et al., 2002). These ditches not only become an efficient means to promote drainage and reduce flooding on production acreage but also provide a buffer between areas of intensive chemical use and aquatic ecosystems at potential risk. Concern over the presence of various pesticides in surface- and groundwater recently has resulted in consideration of higher-tier aquatic risk assessments (Solomon et al., 1996; Campbell et al., 1999; Morton et al., 2000; U.S. EPA, 2000; Giddings et al., 2001; Solomon et al., 2001). Research is now needed to quantify ditch attributes (i.e., runoff residence time, plant communities, water quality, and geomorphology) that support the use of agricultural drainage systems as best management practices for runoff-related issues.

This project evaluated aquatic and habitat attributes that support the use of drainage ditches as best management practices for runoff-related issues by measuring the partitioning, movement, and transformation of agrochemicals. It presents a specific approach for assessing the fate and effects of methyl parathion (*O,O*-dimethyl *O*-4-nitrophenyl phosphorothioate) via constructed wetland mesocosms (vegetated and nonvegetated) to reduce insecticide concentrations associated with agricultural runoff. In addition, it evaluated the various attributes common to agricultural drainage ditches, often associated with wetland ecosystems, for their potential to mitigate the effects of agricultural runoff.

MATERIALS AND METHODS

Study Site

Experiments were conducted in two outdoor wetland mesocosms at a University of Mississippi field station near Oxford, Mississippi. Water was provided by a spring-fed source and rainfall collection. The project objectives included using vegetated and nonvegetated wetlands to determine the fate and effects of exposure to varied concentrations of methyl parathion . Wetlands used in this study were constructed in 1988 (average dimensions: 50 × 10 × 0.32 m; average bed slope: 0.006%) and had been planted previously with wild rice (*Zizania aquatica*), cattail (*Typha latifolia*), and bulrush (*Scirpus cyperinus*); however, natural succession resulted in a vegetative community that included 86% rush (*Juncus effusus*) and 14% cut grass (*Leersia oryzoides*). The latter two plant species were identified and quantified (256 and 43 plants/m^2, respectively) for the vegetated wetland exposure. Elevated platforms were built at each station (5, 10, 20, and 40 m from inlet) to minimize disturbance during water, plant, and sediment collection.

Exposure Regime

Methyl parathion (MeP) is a highly toxic organophosphate insecticide that is primarily used on cotton, and most formulations are classified as restricted-use pesticides by the U.S. Environmental Protection Agency (EPA). Its use in the lower Mississippi Delta averages approximately 402 000 kg of active ingredient per year. The physical properties of methyl parathion include high water solubility (55–60 mg/L at 25°C; Kidd and James, 1991) and a low octanol/water coefficient (K_{ow} = 3.5–3.8; U.S. Public Health Service, 1995), which is considered relatively hydrophilic.

Two types of wetland mesocosms were used in this study: vegetated (VW) and nonvegetated (NVW) systems. Each wetland was divided lengthwise with metal flashing to provide replication within each wetland cell. Each side was treated with commercial grade MeP (43.8% active ingredient) at a rate of 0.43 kg active ingredient/hectare (ha). Wetland exposures utilized a stock solution of wetland water and 6.6 mg MeP/L, with approximately 6435 L of pesticide mixture introduced to each mesocosm at a rate of 9.5 L/s. Pesticide application simulated a storm event of 0.64 cm of rainfall on a 50-ha agricultural field; 5 kg of dry sediment was added to the pesticide mixture to simulate the typical suspended sediment load (400 mg/L total suspended solids) in the Mississippi Delta ecoregion (Smith et al., 2002).

Analytical Methods

Water-quality characteristics measured in both the VW and the NVW following MeP amendments included pH, dissolved oxygen, temperature, and conductivity (APHA, 1995). The temperature ranged from 23.7°C to 27.8°C in the VW and from 27.0°C to 28.6°C in the NVW. Conductivity was higher in the VW, with values ranging from 103 to 125 μs/cm, whereas values measured in the NVW ranged from 37 to 48 μs/cm. Values for pH were relatively neutral for both mesocosms, ranging from 6.5 to 7.1. Dissolved oxygen measurement indicated some differences between the two mesocosms, with measured values ranging from 1.1 to 4.2 mg/L in the VW, and from 5.8 to 7.2 mg/L in the NVW.

Plants, sediments, and surface-water samples were analyzed throughout the 10-day exposure period for MeP uptake at predetermined locations (stations) in the wetland mesocosms (Bennett et al., 2000; Moore et al., 2001; Cooper et al., 2002). Samples were collected after 0.5, 3, 24, and 96 h and 10 days for water and sediment and after 0.5, 3, and 24 h and 10 days for plants. All samples were collected and immediately placed on ice for shipment to the laboratory and subsequent analysis. Methyl parathion was analyzed with a Hewlett-Packard 6890 gas chromatograph (GC) equipped with a DB5 MS column according to methods described by Moore et al. (2001) and Bennett et al. (2000). A multilevel calibration procedure was used with standards and updated every ninth sample. Limits of detection (LOD) for MeP in water, sediments, and plants were 0.1 ng/L, 0.02 μg/kg, and 0.02 μg/kg, respectively. Mean extraction efficiencies were greater than 90%.

Acute Toxicity

Water and sediment were collected at 3, 24, and 96 h and 10 days postexposure for acute toxicity bioassays using *Ceriodaphnia dubia*, *Pimephales promelas*, *Hyalella azteca*, and *Chironomus tentans*. All samples were collected spatially to identify MeP movement through each wetland. Surface-water samples were collected 5, 10, 20, and 40 m from the inlet point. Sediment samples were collected only 5, 10, and 20 m from the inlet point on the basis of preliminary sediment analyses of MeP, which indicated that the pesticide had not bound to sediment particles beyond 20 m. Water and sediment samples for toxicity testing were transported on ice within 6 h of collection to Arkansas State University's Ecotoxicology Research Facility, where test organisms were immediately exposed to test media. Aqueous 48-h toxicity tests were conducted using *C. dubia*, *P. promelas*, and *H. azteca* following methods outlined by the U.S. EPA (1993). Comparison of organism survival with that in control exposures (moderately hard synthetic water: hardness = 80–100 mg $CaCO_3$/L) was the measured end point for all aqueous tests.

Sediment tests were conducted using 10-day solid-phase acute toxicity assays to determine the relative inhibition of survival and growth in exposed *C. tentans* (U.S. EPA, 1994). A static flow-through system was used to renew overlying water in the test chambers twice daily. In addition, each test replicate chamber (6 per site) was fed 1 ml of a Tetramin® solution (4 g/L) daily. Test controls for the sediment toxicity bioassays, used in the determination of statistical significance, included sediments collected from the same ecoregion that had been shown to support survival and growth in *C. tentans* (Moore et al., 1996).

Statistical Analysis

Results from the toxicity bioassays were statistically analyzed using Toxcalc®, version 5.0.25 ($\alpha = 0.05$). Sediment data were tested for normality using the Kolmogorov D test and for homogeneity of variance using Bartlett's test. The Wilcoxon rank-sum test with a Bonferroni T adjustment was used to determine if differences between stations were statistically significant. Aqueous data were tested for normality using Shapiro-Wilk's test and Steel's many–one rank test to determine significance of survival.

RESULTS AND DISCUSSION

In this study, constructed wetlands proved beneficial in mitigating the toxic effects of MeP in simulated storm events. Analytical data indicated that MeP partitioning predominantly occurred in plant tissues. Previous studies reported support for plant species having a significant role in limiting the bioavailability of organophosphate pesticides in field and laboratory exposures (Moore et al., 2002; Schulz et al., 2003b). In this study, the uptake of MeP from the water column by aquatic vegetation reduced the insecticide concentrations and the magnitude of acute toxicity in exposed laboratory organisms. The water solubility of this chemical and its subsequent partitioning into the soil allows for its uptake by aquatic plants, as was shown by the levels of MeP in plant material extracted at selected segment locations.

Our research supports the importance of assessing the temporal and spatial movement of MeP through the wetlands and the significance of these systems in efficiently transforming MeP from the water column to plant tissues or sediments. Using these mesocosms, we measured the fate of the insecticide in the constructed wetlands, concurrent with its effects on the aquatic and sediment test organisms. These data provide quantitative spatial and temporal end points that could be used to determine the specific distance and residence time needed in order to reduce water- and sediment-bound MeP below toxic levels.

Methyl Parathion Partitioning

For this study methyl parathion from sediment and plant tissues was analyzed prior to application in order to determine the ambient concentrations in the wetland mesocosms. Sediment and plant tissue concentrations of MeP in the VW ranged from 21 to 230 and from 67 to 212 μg/kg, respectively. MeP concentrations in the NVW sediment were somewhat lower, ranging from 31 to 115 μg/kg.

Measured aqueous concentrations of MeP in the VW and NVW mesocosms generally decreased as runoff progressed through the wetland (Table I). Sediment concentrations of MeP in the VW were as high as 688 μg/L (10 days; 2.5-m station). Likewise, in the NVW, sediment concentrations of MeP measured more than 10 000 μg/L (24 h; 2.5-m station). Neither of these concentrations, however, elicited a significant decrease in survival in exposed test organisms (*C. tentans*). Although Ali and Mulla (1976) reported a 24 h LC_{50} of 58 μg/L to C. tentans, no decreased survival was noted upon exposure to sediments from these sites.

Measured sediment and plant tissue concentrations of MeP in the VW and NVW generally decreased as runoff progressed through 20 m, with a latent increase of plant-bound pesticide at 40 m (Table I). Methyl parathion partitioning in sediments occurred as early as after 0.5 h (concentrations in the VW and NVW ranged from 0.0 to 230 and from 2.8 to 9270 μg/L, respectively), and the concentration decreased of MeP as it progressed through the wetlands. A peak of sediment-bound MeP in the NVW occurred at the 2.5-m station 24 h postapplication (average = 10 190 μg MeP/L). Sediment concentrations in the NVW may have been higher because of a lack of organic material (e.g., plant tissue) for partitioning to occur in. Within the VW, a greater proportion of measured MeP was bound to plant tissues (average = 2600 μg MeP/L) than on sediment particles (average = 78 μg MeP/L) postapplication. Although wetland vegetation uptake has been quantified, specific research into the type and number of plant species that most efficiently assimilate MeP has not been determined.

Howard (1989) reported a half-life of MeP of 8 days in surface water during the summer months. Using these results, following application the highest aqueous concentrations for the VW (15.4 μg MeP/L) and the NVW (27.0 μg MeP/L) would be reduced to nondetectable limits (0.1 ng MeP/L) after 136 and 144 days, respectively. Our analytical data indicated that MeP measured *in situ* from the VW was reduced to nondetectable limits within 96 h. However, analysis of water in the NVW indicated that it would take longer than 10 days for aqueous concentrations to reach nondetectable limits.

Toxicity Responses

In this study constructed wetlands reduced the toxicological effects of MeP on sensitive aquatic biota. Our research utilized the assessment of water and sediment quality along with toxicity methods in wetland mesocosms (1) to determine which temporal and spatial measurements would be needed to sufficiently (>80% survival) mitigate MeP exposure, and (2) to relate this information to similar vegetated agricultural ditch systems.

3 h Postapplication

Significant mortality was measured in *C. dubia* at the 5-, 10-, and 20-m stations in the VW and NVW, with average aqueous concentrations ranging from 3.5 to 9.6 μg MeP/L (Table II). The only toxic response by *P. promelas* was seen after exposure to water from the 10-m station in the NVW (67.5% survival), where MeP concentrations measured 0.5 μg MeP/L. LC_{50} values were not determined for *P. promelas* because there were not enough partial responses in any of the fish tests to calculate a value. Toxicity tests conducted for pesticide registration supported these findings and indicated that the average LC_{50} values calculated for fathead minnow and Daphnia magna are 8.5 $\pm$ 1.2 mg MeP/L and 12.3 μg MeP/L, respectively (U.S. EPA, 2000). Maximum MeP concentrations for *P. promelas* in those studies were 3 orders of magnitude greater than those found in our study (9.6 μg/L).

24 h Postapplication

Significant mortality was measured in *C. dubia* at the 5-, 10-, and 20-m stations in the VW and NVW, where MeP concentrations ranged from 6.6 to 15.4, and from 2.9 to 14.8 μg/L, respectively. While 48-h LC_{50} values could not be calculated in this study because there were no partial responses, calculated LOEC (lowest observable effect concentration) for both wetland mesocosms averaged 15.0 $\pm$ 0.4 μg MeP/L. Similar studies using *C. dubia* support the sensitivity of this invertebrate with 48-h LC_{50} values ranging from 2.6 to 5.5 μg MeP/L (Norberg-King et al., 1991). Reported EC_{50} values for *Daphnia magna* (average 12.3 $\pm$ 14.4 μg/L) and one LC_{50} value for *Gammarus fasciatus* (3.8 μg/L) indicate sensitivity similar to that in the toxicity tests conducted in this study, where MeP concentrations ranged from 0.3 to 27.6 μg/L (U.S. EPA, 2000).

96 h Postapplication

Significant mortality occurred in exposed *C. dubia* from all stations (5, 10, 20, and 40 m) in the NVW, with pesticide concentrations averaging 3.1 ($\pm$ 0.3), 13.6 ($\pm$ 15.9), 18.1 ($\pm$ 2.6), and 6.9 ($\pm$ 1.3) μg MeP/L, respectively. *Ceriodaphnia* mortality in the VW occurred only at the 5- and 10-m stations [average concentrations were 1.8 ($\pm$ 1.9) and 11.5 ($\pm$ 11.8) μg MeP/L, respectively].

10 Days Postapplication

In this study organisms were subjected to long-term exposures (e.g., >1 week) in order to determine the lethal and

TABLE I. Average methyl parathion concentrations (±1 SD) measured in water, sediment, and plants from vegetated and nonvegetated wetlands (sample size, *n*, is 2, unless otherwise noted)

	Vegetated			Nonvegetated	
Station[a]	H_2O (μg/L)	Sediment (μg/kg)	Plant (μg/kg)	H_2O (μg/L)	Sediment (μg/kg)
0 h postapplication					
2.5 m	—	82.7 (n =1)	—	—	—
5 m	—	—	211.6	—	30.8 (n = 1)
10 m	—	—	—	—	—
20 m	—	20.9 (n = 1)	66.9	—	89.2 (n = 1)
40 m	—	230.6 (n = 1)	148.5	—	115 (n = 1)
0.5 h postapplication					
2.5 m	5.14 (1.88)	229.8 (325)	8,155 (10,984)	7.89 (3.33)	9,270 (4,949)
5 m	11.47 (0.89)	15.2 (21.4)	2,533 (2,161)	27.01 (14.12)	1,444 (1,934)
10 m	6.67 (5.73)	0.8 (1.1)	1,275 (1,671)	4.36 (5.29)	581 (793)
20 m	11.16 (12.92)	0.0 (0.0)	2.1 (2.9)	3.64 (4.54)	20.7 (26.7)
40 m	ND	24.6 (30.7)	425.3 (128.4)	0.11 (0.03)	2.8 (4.0)
3 h postapplication					
2.5 m	7.22 (4.36)	53.9 (68.3)	21,523 (28,454)	17.68 (4.90)	585 (327)
5 m	6.74[b] (2.24)	108.4 (142.2)	4,650 (5,638)	8.82[b] (0.67)	131.0 (160.4)
10 m	4.09[b] (2.44)	77.2 (109.2)	4,519 (5,891)	9.62[b] (0.50)	0.5 (0.6)[b]
20 m	6.60[b] (8.95)	7.7 (6.4)	26.2 (18.1)	3.52[b] (0.92)	0.0 (0.0)
40 m	ND	12.8 (0.2)	249.7 (145.0)	0.24 (0.12)	4.2 (6.0)
6 h postapplication					
2.5 m	6.76 (4.11)	—	—	9.84 (0.61)	—
5 m	4.77 (1.58)	—	—	5.62 (3.22)	—
10 m	9.41 (11.31)	—	—	15.16 (7.89)	—
20 m	2.29 (2.27)	—	—	5.78 (2.76)	—
40 m	ND	—	—	0.47 (0.15)	—
24 h postapplication					
2.5 m	4.05 (0.33)	214.8 (293.6)	2,481 (2,993)	3.94 (0.39)	10,189 (9,432)
5 m	15.38[b] (17.21)	40.3 (57)	713.1 (16.9)	14.75[b] (9.61)	75.6 (3.2)
10 m	7.22[b] (9.75)	5.4 (7.7)	2,674 (3,688)	13.11[b] (7.03)	0.7 (0.9)
20 m	6.57[b] (8.12)	0.0 (0.0)	73.5 (60.6)	2.86[b] (2.01)	0.0 (0.0)
40 m	ND	12.3 (4.0)	120.6 (30.4)	5.13[b] (n = 1)	7.2 (10.2)
96 h postapplication					
2.5 m	7.42 (1.66)	—	—	5.82 (3.67)	—
5 m	1.76[b] (1.96)	—	—	3.09[b] (0.34)	—
10 m	11.51[b] (11.79)	—	—	13.60[b] (15.86)	—
20 m	0.53 (0.55)	—	—	18.07[b] (2.62)	—
40 m	ND	—	—	6.93[b] (1.29)	—
10 days postapplication					
2.5 m	—	687.8 (752.1)	133.5 (n = 1)	2.99 (0.59)	687 (752)
5 m	3.45[b] (0.02)	18.1 (8.4)	2,186 (n = 1)	2.18[b] (0.90)	18.1 (8.4)
10 m	0.48 (n = 1)	18.6 (14.0)	96.0 (n = 1)	2.79 (0.03)	18.6 (14.0)
20 m	ND	12.9 (1.0)	75.7 (30.9)	0.51 (0.01)	12.9 (1.0)
40 m	ND	23.3 (5.0)	180 (13.4)	0.44 (0.04)	23.3 (5.0)

[a] Stations represent distance from inlet point (0 m).
[b] Biological test organisms elicited significant toxicity at these pesticide concentrations.
— Not sampled for toxicity testing or chemical analysis.
ND: Not detected (LOD = 0.1 ng/L).

sublethal effects of MeP to exposed organisms. Significant mortality was measured in *C. dubia* at 5 m (40% survival) in the NVW (2.2 μg MeP/L) only. This may suggest that by 10 days MeP was no longer bioavailable in surface waters at concentrations that would elicit an acute response. LC_{50} and LC_{10} values were calculated separately for *C. dubia* and *H. azteca* using survival data for all stations and wetlands to determine the lethal concentration of MeP in these species. The calculated LC_{50} for *C. dubia* in this study was 1.8 μg MeP/L. LC_{10} values were calculated for a more conservative effort to protect a greater percentage of the test organisms. The calculated LC_{10} for *C. dubia* was 0.7 μg MeP/L.

TABLE II. Results of acute (aqueous) toxicity survival results from vegetated and nonvegetated wetland mesocosms; stations represent distance from inlet point (0 m)

	3-h Collection			
	C. dubia (% Survival)		*P. promelas* (% Survival)	
Station	Vegetated	Nonvegetated	Vegetated	Nonvegetated
5 m	0[a]	0[a]	97.5	90
10 m	0[a]	0[a]	92.5	67.5[a]
20 m	0[a]	0[a]	97.5	92.5
40 m	75	100	87.5	90

	24-h Collection			
	C. dubia (% Survival)		*P. promelas* (% Survival)	
Station	Vegetated	Nonvegetated	Vegetated	Nonvegetated
5 m	0[a]	0[a]	80	92.5
10 m	0[a]	0[a]	100	90
20 m	50[a]	0[a]	100	70
40 m	100	0[a]	100	100

	96-h Collection	
	C. dubia (% Survival)	
Station	Vegetated	Nonvegetated
5 m	0[a]	0[a]
10 m	0[a]	0[a]
20 m	100	0[a]
40 m	100	0[a]

	10-day Collection			
	C. dubia (% Survival)		*H. azteca* (% Survival)	
Station	Vegetated	Nonvegetated	Vegetated	Nonvegetated
5 m	70	40[a]	30[a]	62.5
10 m	100	70	92.5	67.5
20 m	90	100	92.5	100
40 m	100	100	97.5	100

[a] Significantly different from control results.

Vegetated wetlands would be protective of this species for 6 h and at a distance of 40 m from the inlet. The toxicity results and analytical data for the NVW indicated that the residence time of MeP would need to be greater than 10 days for the protection of *C. dubia.*

Hyalella were not significantly affected in the NVW the shortest distance (5 m) from the inlet for the 10-day collection. However, there was a measured impact on *H. azteca* (30% survival) in the VW at the 5-m station; associated MeP concentrations at the same station averaged 3.5 (± 0.02) μg/L. The LC_{50} of *H. azteca* in this study was 2.9 μg/L. Studies conducted by Anderson and Lydy (2002) support the sensitivity of *H. azteca* exposures to MeP; they reported 96-h LC_{50} values as low as 2.1 μg/L. Their study also indicated that MeP is more toxic to amphipods than are other organophosphate pesticides (i.e., chlorpyrifos and diazinon). As shown in our study, MeP was still detectable in water in both the VW and NVW, with concentrations of 3.5 and 2.8 μg/L, respectively. For the protection (90% of the test population) of *H. azteca* in this study, we used calculated LC_{10} values (1.0 μg MeP/L) to determine how wetlands could effectively mitigate MeP. Both wetland types, vegetated and nonvegetated, would provide protection to *H. azteca* with a residence time of 10 days and a distance of 10 and 20 m, respectively.

Schulz et al. (2003b) conducted similar studies using *H. azteca*. MeP detection in a nonvegetated wetland extended the entire length (40 m) of the mesocosm, with concentrations ranging from 0.3 to 550 μg/L. Aqueous MeP concentrations in vegetated wetlands indicated that the insecticide was only detected through 20 m, suggesting that the vegetation contributed to reducing its bioavailability and toxicity to exposed *H. azteca*. Toxicity in these wetlands was significantly reduced as time and distance from the inlet increased. The 48-h LC_{50} calculated for *H. azteca* was 9.0 μg MeP/L.

Other toxicity studies utilizing *C. tentans in situ* for aqueous exposures showed significant acute toxicity 3 h postapplication at 5,- 10-, and 20-m stations in unvegetated wetland exposures with MeP (Schulz et al., 2003a). Results from the same study indicated that significant toxicity to exposed midges was only measured through 10 m in vegetated wetlands.

Sediment Toxicity

Sediment-bound MeP reduced *C. tentans* survival (65%) at 10 m in the NVW 3 h postapplication. Methyl parathion concentrations for this station averaged 0.5 μg/kg. Average sediment concentrations of MeP for all other stations ranged from 7.7 to 131 μg/kg. No significant mortality or reduction in growth was measured in any of the other mesocosm stations through 24 h postapplication (Table III). Toxicity at the 10-m station may have been a result of MeP photolysis to other toxic byproducts (e.g., 4-nitrophenol) not analytically measured in this study. Toxicity assays with *C. tentans* in other studies have determined that midges are less sensitive to MeP than are other test organisms (Pape-Lindstrom and Lydy, 1997). Their research indicated that acute toxicity of *C. tentans* in single-chemical aqueous tests resulted in EC_{50} values of 32.3 μg MeP/L.

Best Management Practices

The data collected from this constructed wetland study could be used to redefine existing uses of agricultural drainage ditches, from a traditional conveyance structure during

TABLE III. ***Chironomus tentans*** **acute toxicity (sediment) survival and growth results from vegetated and nonvegetated wetland mesocosms; stations represent distance form inlet point (0 m)**

	3-h Collection			
	Survival (%)		Growth (mg)	
Station	Vegetated	Nonvegetated	Vegetated	Nonvegetated
5 m	73.8	93.8	2.3	2.7
10 m	83.8	65[a]	2.9	2.5
20 m	77.5	91.3	2.7	2.4
	24-h Collection			
	Survival (%)		Growth (mg)	
Station	Vegetated	Nonvegetated	Vegetated	Nonvegetated
5 m	87.5	86.3	2.6	2.0
10 m	95	92.5	2.6	2.2
20 m	86.3	92.5	2.5	2.3

[a] Significantly different from controls.

storm events or irrigation practices to a functional ecosystem capable of mitigating pesticides and lessening the impact on aquatic communities. Although the ultimate goal of utilizing wetlands is to achieve no observable effects in organisms once runoff enters a downstream receiving system, the objective of this research was to utilize toxicity assays to measure temporal and spatial reduction of pesticide concentrations associated with agricultural runoff.

The results of these toxicity bioassays indicate that as both distance from inlet and time following application increase, VW mesocosms become more effective in mitigating MeP. Vegetated wetlands provided more protection for aquatic organisms (sediment dwelling, epibenthic, and water column) because of the partitioning of MeP into sediment and plant material (Fig. 1). Although the protection (90%) of *C. dubia* and *H. azteca* differed depending on the type of wetland (VW and NVW) in which they were exposed, their overall responses indicate that effective mitigation of MeP would be achieved following a 10-day residence time. For the protection of *P. promelas*, a constructed wetland or agricultural drainage ditch would need to be at least 5 m long with a chemical residence time of at least 3 h to mitigate MeP beyond its toxic effects and, consequently, to protect sensitive vertebrates. Moore et al. (2001) concluded that the use of vegetated ditches was a comparable substitute for vegetated wetlands in mitigating agricultural-associated chemicals. In our study we found that constructed wetlands could sufficiently mitigate the toxic effects of MeP on all tested organisms at a distance of 40 m from the inlet. Using this model as an approach for agricultural drainage ditches, unvegetated ditches would need to be maximized for distance and residence time for the reduction of MeP, whereas shorter ditch lengths and residence times could be implemented if aquatic plant communities were present in the system during the growing season.

Drainage ditches often mimic wetland characteristics in mitigating contaminants, and these data can be utilized by agriculture to manage ditch systems in a way that more efficiently utilizes existing land use (e.g., ditch length and width) and reduces pesticide exposures in receiving streams. Furthermore, drainage ditches can be managed in a way that optimizes crop production by minimizing land loss (addition of riparian buffers for mitigation) from other BMPs that provide only an approximation for drainage ditch mitigation.

Although characteristics of vegetated agricultural ditches often go unnoticed as potential ecological benefits (i.e., ability to reduce contaminant concentrations below acutely toxic levels), their use in fate and effects studies provide more realistic pesticide exposure regimes than do single-species laboratory toxicity tests because the assimilative capacity of the drainage ditch is accounted for in the exposure response of test organisms. For this reason, several researchers are now examining the fate and effects of insecticides in mesocosms that resemble drainage ditches. Schulz et al. (2003) described similar fate and transport of MeP through a constructed wetland but assessed the impact on test organisms *in situ.* That study also provided support for the effectiveness of wetlands in mitigating specific organophosphate pesticides. Bouldin et al. (2004) summarized the fate and effects of the insecticide esfenvalerate, over a 56-day exposure using an agricultural drainage ditch. Their results indicated that in a vegetated ditch, esfenvalerate loses its toxic effects after a residence time of at least 3 h and at a minimum distance of 80 m from the point of entry (inlet).

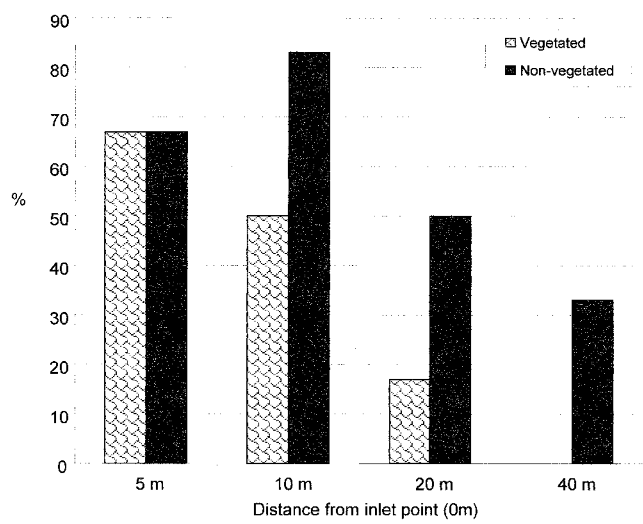

Fig. 1. Percentage of the toxicity tests (sediment and water) conducted during the study that elicited significant mortality in exposed organisms.

CONCLUSIONS

This study utilized standardized laboratory toxicity tests and pesticide analyses to quantify specific distances from the inlet and residence times needed to sufficiently mitigate (>90% survival) pesticide effects on biological communities (invertebrates and vertebrates). These data indicate that pesticide concentrations can be reduced to no-effect levels within relatively short distances and residence times, and if allowed to contact aquatic vegetation, pesticide concentrations are often reduced at shorter distances and times because of the uptake by plant tissues. Vegetated and unvegetated wetland mesocosms were used to reflect similar structural and functional aspects of agricultural drainage ditches and to assess acute toxicity and the fate of MeP for 10 days postapplication. Consequently, agricultural management techniques can be refined (e.g., allow aquatic plants to cultivate in drainage ditches, design drainage ditches to allow for specific distances from receiving streams, etc.) based on these fate-and-effects studies and applied to in-field drainage systems in order to reduce the potential impact of pesticides to downstream receiving systems. To date, there tends to be insufficient published data that specifically addresses the use of agricultural drainage ditches as mitigating structures to reduce agrochemicals in receiving streams. This study may provide the guidance needed to quantify the specific structural and temporal requirements of these drainage systems to protect in-stream biological communities.

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