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BIOMAGNIFICATION OF ATRAZINE IN LAKE COLUMN SIMULATORS

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ON

GREAT LAKES

POLLUTION FROM LAND USE ACTIVITIES

BIOMAGNIFICATION OF ATRAZINE IN LAKE COLUMN SIMULATORS

by

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3.0 DISCLAIMER

The study discussed in this document was carried out as part of the efforts of the Pollution from Land Use Activities Reference Group, an organization of the International Joint Commission, established under the Canada-U.S. Water Quality Agreement in 1972. Funding was provided through Fisheries and Environment Canada. Findings and conclusions are those of the authors and do not necessarily represent the views of the Reference Group or its recommendations to the Commission.

4.0 ACKNOWLEDGEMENTS

The technical assistance of Mr. R. Bishop is gratefully acknowledged.

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8.0 SUMMARY

Additions of atrazine at two treatment levels were made to four lake column simulators in each of two experiments. The lowest treatment in both experiments was sufficient to yield concentrations in the range (0-30 ppb) frequently measured in agricultural watersheds in late spring and early summer while the other was an order of magnitude higher. The amount of atrazine added at each treatment level was similar between experiments but was applied over different lengths of time, 14 days in experiment I and 5 days in experiment II.

Atrazine was detected in most components of a simple food chain (algae → Daphnia → guppies) but concentrations, particularly in fish, were not much higher than in the water. Over 90% of atrazine added could be accounted for by adding quantities found in open water of the upper and lower layers. The bulk of atrazine added remained in solution in the upper layer.

Bench-scale experiments showed that Daphnia magna and guppies exposed to atrazine in water only, had residues in proportion to water concentrations. No trend in accumulation with time was noted in exposures up to a week's duration. De-ethylated atrazine was not detected in the columns experiments but was detected in guppies in bench-scale experiment in proportion to atrazine water concentrations. These smaller experiments indicated that a fraction of the atrazine residues found in biota in the columns experiments were due to uptake or

adsorption directly from water.

Neither impairment of photosynthesis by algae nor toxic effects to zooplankton and fish were apparent in columns experiments. Atrazine does not appear to be a threat to the Great Lakes ecosystem in terms of either toxicity or bioaccumulation.

9.0 INTRODUCTION

Sales of herbicides in Canada have risen markedly in the last two decades from approximately 5 million dollars in the 1950's to well over 20 million dollars by the early 1970's (Thomson, 1973). Herbicides accounted for over half of the total pesticide used in Ontario in 1973 with atrazine accounting for 42% of the herbicide use (Roller, 1975). Information on the fate of atrazine in the aquatic ecosystem and potential for biomagnification has been non-existent until recent years. Generally, herbicides are more water soluble than insecticides thereby enhancing leaching and erosional losses from agricultural watersheds. However, herbicides exhibit lower mammalian toxicity, are less persistent than chlorinated hydrocarbon insecticides and have a reduced potential for accumulation in the ecosystem (Edwards, 1975). These properties infer that atrazine should pose a minimal threat to the environment except when careless events occur, such as spills or rinsing of sprayers in streams. On the other hand, the immense quantities of this substance in use and the minimal information available on persistence and accumulation in the aquatic ecosystem suggested a need for further research.

The purpose of our study was to determine the extent of biomagnification of atrazine to various trophic levels under different levels and frequencies of application in model ecosystem experiments.

10.0 MATERIALS AND METHODS

10.1 LAKE COLUMN SIMULATORS - CHARACTERISTICS AND CONTROL

The LCS are eight stainless-steel columns, 4.5 m high, 1.0 m in diameter, with a volume of 3336 l when filled to 4.25 m. The LCS are located indoors in the wet lab area of the Great Lakes Biolimnology Laboratory at the Canada Centre for Inland Waters, Burlington. The sole source of irradiance for each column was two banks of 36 in fluorescent lamps, each containing four Warm-White and four Gro-lux lamps (Sylvania, Canada). The lamps were on a 15 h light 9 h dark photoperiod with dawn and dusk simulation provided by automatic switching of pairs of lamps every 15 min at the start and end of each light period. Quantum flux just above the water surface was approximately 175-225 μ Einsteins m^{-2} sec^{-1}.

A coil carrying a coolant mixture of ethylene glycol and water encircles the lower 2.0 m of the column (lower layer - LL) and is connected to a coolant reservoir equipped with a pump and refrigerating system. Temperature stratification can be established in the middle of the column with this system. Advective gains of heat to the (LL) through the steel walls are minimized by an insulative neoprene jacket. Temperatures in the LL averaged 10-12°C in all four columns during both experiments. Temperature in the upper layer of the columns (UL) is uncontrolled and in balance with the ambient air temperature due to heat gain through the uninsulated steel walls of this portion of the column.

Temperatures in the UL averaged 20-22°C and 25-26°C in experiments I and II respectively. Higher UL temperatures in experiment II were a result of the higher building temperatures at this time of the year (July) compared to the first experiment (Feb.).

The columns were isothermal from the surface to 1.5 m in experiment I but down to only 1.0 m in experiment II. Temperature discontinuity started between 1.5 m and 2.0 m in experiment I and between 1.0 and 1.5 m in experiment II. As a result, a thinner isothermal layer and thicker zone of temperature change were present in experiment II. This has important implications regarding distribution of atrazine in the columns.

Growth of attached algae on container walls is a common problem in model ecosystems. Biomass of attached algae tended to be restricted to the upper 0.5 m due to shading by algae in the open water and was controlled by frequently scrubbing the upper walls with abrasive pads.

10.2 NUTRIENT ADDITIONS

An amount of phosphorus required to raise the concentration in the entire column to 5 µg liter^{-1} was added prior to the experiment. Phosphorus loading was continued after this initial spike at ca. 1.0 g P$\cdot$m$^{-2}\cdot$ yr^{-1} or 2.82 mg$\cdot$column$^{-1}\cdot$ day^{-1}. The amounts of other macronutrients added were relative to the phosphorus loading but in a proportion to each other similar to Chu No. 10 (Chu, 1942) culture medium (Table 1).

Table 1. Quantities of macro- and micronutrients added to all columns

Element	Loading
	Macronutrients ($mg \cdot day^{-1}$)
P	2.82
N	10.75
Ca	15.35
Mg	3.84
Na	28.47
C	3.57
Si	9.03
Fe	0.44
	Micronutrients ($\mu g \cdot day^{-1}$)
Co	20.87
Cu	84.4
B	425.49
Mn	102.7
Mo	99.4
Zn	412.49

10.3 ATRAZINE TREATMENTS

Two experiments were conducted using the commercial preparation AAtrex (Ciba-Geigy Ltd.) which contained 90% atrazine (2-chloro-4-ethylamino-6-isopropylamino-s-triazine) according to the manufacturer's specifications. In both experiments, two columns received treatments differing by an order of magnitude and two columns served as controls for toxic effects and received no atrazine.

10.3.1 Experiment I

Each treatment received seven doses of atrazine applied on alternate days starting approximately four weeks after algae were added. The lower treatment (LT) to column 1 was 3.49 mg per dose and the higher treatment (HT) to column 4 was 34.9 mg of AAtrex per dose. Columns 2 and 3 were controls.

These applications were capable of raising the concentration of atrazine in the UL of the two treatments by 1 and 10 $\mu g \cdot liter^{-1} \cdot day^{-1}$ if all of the atrazine added remained in this layer (rationale for application rates chosen, see p. vii).

10.3.2 Experiment II

A rapid rise to high atrazine concentrations, similar to the trend in atrazine concentrations found in agricultural watersheds in southern Ontario during late June and early July (Frank et al., 1978), was simulated in this experiment. The LT (column 3) and the HT (column 1) received three doses each of 9.9 and 99.0 mg AAtrex respectively within a five-day period. Columns 2 and 4 were controls. These applications were capable of raising the atrazine concentration in the UL to 17 and 170 $\mu g \cdot liter^{-1}$ if all of the atrazine remained in this layer.

10.4 FOOD CHAIN

10.4.1 Algae

Inocula were taken from a dense, mixed-species, batch culture. In experiment I 20 g dry weight of this culture was added to each column and atrazine doses commenced four weeks later. In experiment II we encountered difficulty establishing a dense algal community. A second inoculum of 27 g dry weight, from the same type of culture as that used in experiment I, was made to each column six weeks after the initial inoculation. Atrazine doses commenced ten days after the second inoculum. All data presented here are for the time period after the second algal inoculum.

10.4.2 Zooplankton

Approximately 2 and 8 thousand Daphnia magna were added to each column in experiments I and II respectively. Zooplankton were added concurrently with algae in experiment I. In experiment II no further additions of zooplankton were made with the second algal inoculum. Animals were originally added concurrently with the first algal inoculum in experiment II.

10.4.3 Fish

Ten female guppies (Lebistes reticulatus) were added to each column in experiment I, one week after the additions of algae and zooplankton. In experiment II, four female and three male guppies were added to each column prior to the second algal inoculum.

10.5 ALGAL BIOMASS PARAMETERS

Composite water samples from the UL of each column were collected with a tube sampler. Composite samples from the LL were taken by pooling aliquots taken from different pressurized sampling ports. Analyses for chlorophyll *a* and particulate organic carbon were performed by the Water Quality Laboratories at CCIW, Burlington. Analytical procedures are outlined in the Analytical Methods Manual (1975).

10.6 PRIMARY PRODUCTION

Primary production was measured in experiment II only, using ^{14}C-tracer methods (Vollenweider, 1974). Bottles were suspended *in situ* at ca. 10 cm for 3-4 h. Total inorganic carbon was measured directly by the Water Quality Laboratory at CCIW.

10.7 SAMPLING FOR ATRAZINE RESIDUES

10.7.1 Water

Composite water samples were collected from both layers, in treated columns only after four atrazine doses, and from all columns two days after the last atrazine dose in experiment I. Samples from control columns were analyzed as a check for cross contamination between columns. Water samples were refrigerated in clean glass bottles until analyses.

Sample collection, preparation and storage were the same in experiment II. Filtered (GF/C, Whatman Co.) and unfiltered water samples were analyzed in experiment II.

Samples were collected from treated columns one week after, and from all columns two weeks after, atrazine additions had started in experiment II.

10.7.2 Suspended Particulates

Suspended particulate matter (excluding zooplankton) in both the UL and LL was collected by continuous-flow centrifugation of 20-30 l of water.

Samples were collected at two days and two weeks after atrazine treatments were terminated in experiments I and II respectively. Samples were oven-dried at 60°C for 24 h in experiment I and freeze-dried in experiment II.

10.7.3 Sedimented Particulates

Glass jars (8 cm deep, 9 cm diam) were suspended at 3 m for a period starting three weeks prior to and extending throughout the two-week period of atrazine additions in experiment I. Traps were retrieved, excess water decanted and the contents oven-dried at 60°C for 24 h.

In experiment II, the traps were retrieved every 3-4 days during a two-week period following the first atrazine addition. The contents from separate retrievals were pooled and freeze-dried.

10.7.4 Zooplankton

In experiment I, net collections of animals were made two days following the last atrazine addition and oven-dried at 60°C for 24 h.

Net collections were made only in the LT in experiment II, two weeks after atrazine additions were begun.

Fish

All fish, including those born during the experiment, were captured with a net and combined as one sample per column. Fish were oven-dried in experiment I and freeze-dried in experiment II.

10.8 ATRAZINE UPTAKE FROM WATER

Accumulation of atrazine from exposure to the substance in water only was investigated in two bench-scale experiments. Several hundred D. magna were placed in 3 L beakers of dechlorinated tap water without food, at atrazine concentrations of 17, 170 and 1700 ppb for periods of 1, 4 and 7 days at each concentration. At the end of each exposure time all animals (live and dead) were collected, rinsed and freeze-dried.

A similar experiment was conducted with 11 guppies exposed without food at each concentration and exposure period. All fish from each exposure time and concentration were combined as one freeze-dried sample.

10.9 ANALYTICAL PROCEDURE

All atrazine analyses were conducted by the Provincial Pesticide Residue Laboratory, Guelph, Ont. Details of the analytical procedure are given in Sirons et al., 1973. Only total atrazine was measured in experiment I but de-ethylated atrazine was also measured in experiment II.

11.0 RESULTS

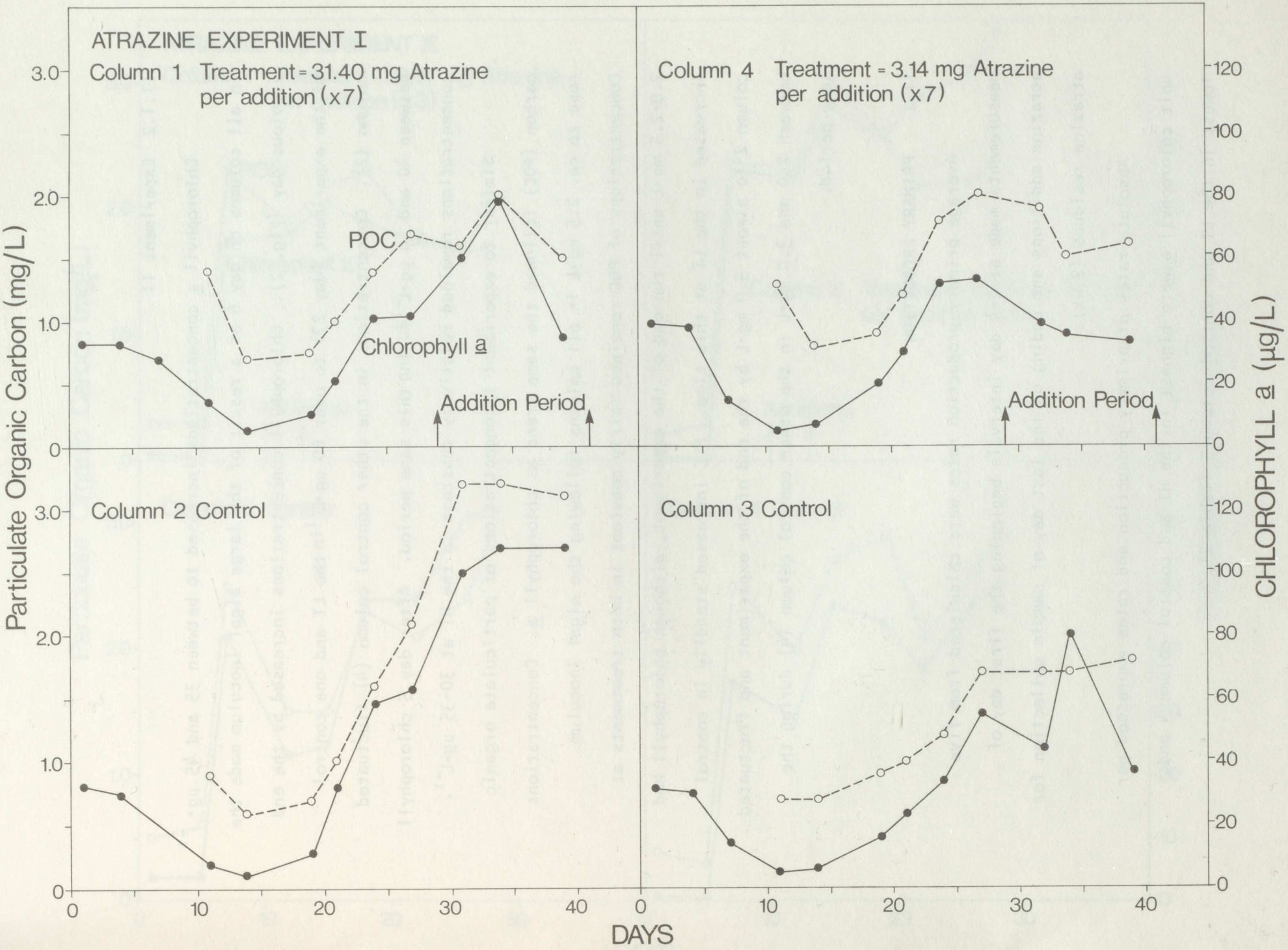
11.1 ALGAL BIOMASS PARAMETERS

11.1.1 Experiment 1

Chlorophyll a concentrations declined following inoculation from about 30 to 6 $\mu g \cdot L^{-1}$ by day 15. A subsequent increase occurred in all columns although the maximum concentrations varied.

Chlorophyll a concentrations in column 2 (control) peaked at over 100 $\mu g \cdot L^{-1}$ and remained at this level through the atrazine additions. In contrast, the HT and column 3 (control) attained maximum concentrations of 80 $\mu g \cdot L^{-1}$ during atrazine additions but concentration in both declined to ca. 35 $\mu g \cdot L^{-1}$. The LT reached its maximum chlorophyll concentration prior to atrazine additions and declined slowly throughout the treatment period.

Concentrations of particulate organic carbon (POC) followed a trend similar to that for chlorophyll a. In comparison, fewer fluctuations were noted in POC concentrations during the peak periods and treatments. The declines in chlorophyll a noted in one control and both treatments during atrazine additions were not as evident with POC. Maximum concentrations were between 1.5 and 2.0 mg•L in both treatments and one control (column 3) but reached higher levels (3.0 $mg \cdot L^{-1}$) in the other control (column 2), similar to chlorophyll a.


ATRAZINE EXPERIMENT I
Column 1 Treatment = 31.40 mg Atrazine per addition (x7)
Column 4 Treatment = 3.14 mg Atrazine per addition (x7)
Column 2 Control
Column 3 Control
POC
Chlorophyll a
Addition Period
Addition Period
Particulate Organic Carbon (mg/L)
CHLOROPHYLL a (µg/L)
DAYS
0
1.0
2.0
3.0
0
20
40
60
80
100
120
0
10
20
30
40

11.1.2 Experiment II

Chlorophyll a concentrations increased to between 35 and 45 $\mu g \cdot ^{-1}$ in all columns on day 6 as a result of the large algal inoculum made the previous day (Fig. 2). Chlorophyll concentrations increased by the end of the experiment (day 27) to over 60 $\mu g \cdot L^{-1}$ in the LT and one control column (2). Concentrations in the other control column (4) fluctuated between 40 and 50 $\mu g \cdot L^{-1}$ during this same period. After day 5 chlorophyll concentrations remained relatively constant in the HT at 30-35 $\mu g \cdot L^{-1}$.

Similar to experiment I, concentrations of particulate organic carbon (POC) followed the same trend as chlorophyll a. Concentrations rose to ca. 2.5 mg•L in all columns following the algal inoculum. Concentrations of POC remained fairly constant in both treatments at 2.0-2.5 mg•L until the end of the experiment, although chlorophyll had increased in the LT at this time. POC increased steadily in control column 2 to almost 3.0 mg•L by the end of the experiment and fluctuated between 2.0 and 3.0 mg•L in the other control column (4) during the same period.

11.2 PRIMARY PRODUCTION

Average primary production rates were calculated from five measurements made at 2-4 day intervals beginning the first day of atrazine additions and ending on the last day of sample collection for atrazine residues (2 wk).

Absolute rates of primary production and rates expressed per unit chlorophyll a were highest in the HT and control column 4 and lowest in the LT and control column 3 (Table 2).

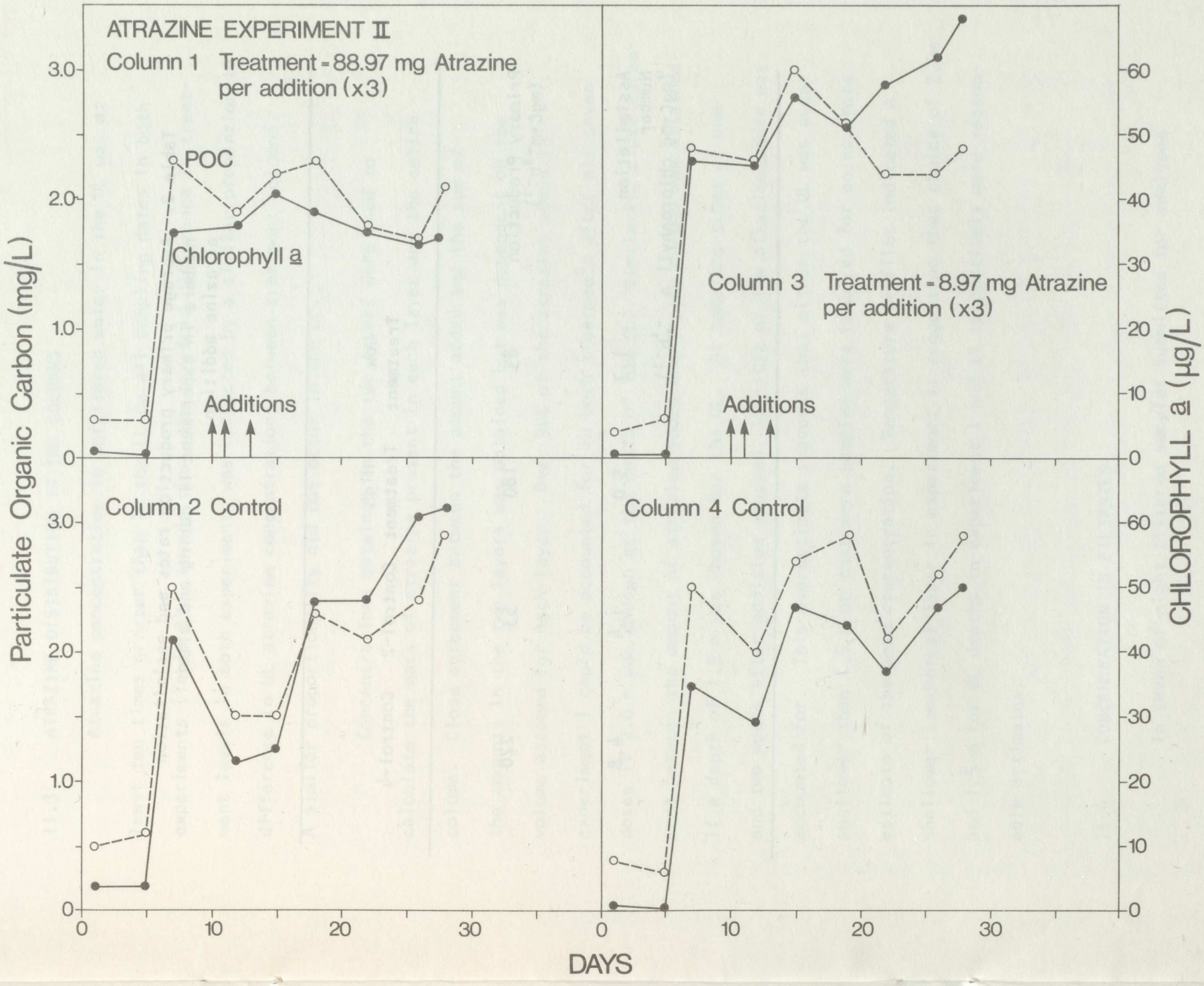

ATRAZINE EXPERIMENT II
Column 1 Treatment = 88.97 mg Atrazine per addition (x3)
POC
Chlorophyll a
Additions
Column 3 Treatment = 8.97 mg Atrazine per addition (x3)
Additions
Column 2 Control
Column 4 Control
Particulate Organic Carbon (mg/L)
CHLOROPHYLL a (µg/L)
DAYS
0
1.0
2.0
3.0
10
20
30
40
50
60

Table 2. Average primary production rates and assimilation numbers in experiment II during and after atrazine addition

	Low Treatment	High Treatment	Control-2	Control-4
Primary Production ($mgC \cdot m^{-3} h^{-1}$)	83	180	53	220
Assimilation Number ($mgC \cdot mg\ chlorophyll\ \underline{a}^{-1} \cdot h^{-1}$)	1.7	5.0	1.4	4.8

11.3 ATRAZINE DISTRIBUTION IN THE COLUMNS

Atrazine concentration in unfiltered water in the UL was at least ten times greater than in the LL on all sampling dates in both experiments (Table 3). In addition, the ten-fold difference in treatment levels in both experiments was reflected by a similar proportionate difference in UL atrazine concentrations between treatment columns. A similar proportionality did not occur in the LL.

Concentrations obtained in the two layers were used to calculate the mass of atrazine present in each layer and the entire column. Close agreement between the amount added and the sum of the amounts in the two layers was obtained but was dependent on the volume assumed for each layer. Over 90% of the atrazine added in experiment I could be accounted for in both treatments after all seven doses if 2.0 m was chosen as the depth of the UL. Similarly, close agreement between the amount of atrazine added and in the column was attained if a depth of 1.5 m was chosen for the UL. At sampling times of one and two weeks after addition stopped, 101-107% of the atrazine added was accounted for. This overestimate suggests that either the UL was even shallower than 1.5 m or that more samples were required for an accurate estimate of the mean concentration. Temperature profiles indicated a shallower isothermal layer in experiment II suggesting that choice of 2.0m and 1.5 m for UL depths in experiment I and II respectively were reasonable estimates.

11.4 CONCENTRATION IN FILTRATES

In seven of eight filtered samples submitted for analyses

Table 3. Concentrations and amounts of atrazine in the columns compared to the amount added.

Treatment	Amount Added (mg)	Concentration[a] (ppb)		Atrazine in Column[b]							
				UL - 1.5 m				UL - 2.0 m			
		UL	LL	UL	LL	Total	%	UL	LL	Total	%
Experiment I[c]											
LT-4	12.6	6.8	trace	8.0	-	8.0	63.5	10.7	-	10.7	84.9
LT-7	22.0	12.1	0.6	14.3	1.3	15.6	70.9	19.0	1.1	20.1	91.4
HT-4	125.6	74.5	4.1	87.8	8.9	96.6	76.9	117.0	7.2	124.2	98.9
HT-7	219.8	125.0	14.1	147.3	30.4	177.7	80.9	196.3	24.9	221.2	101
Experiment II[d]											
LT-1	26.7	22.0	0.7	25.9	1.5	27.4	101	34.5	1.2	35.8	133
LT-2	26.7	20.5	1.9	24.2	4.1	28.3	105	32.2	3.4	35.5	132
HT-1	266.9	235.7	3.1	277.7	6.7	284.4	107	370.1	5.5	375.5	141
HT-2	266.9	221.4	7.6	260.8	16.4	277.2	104	347.6	13.4	361.0	135

a - Unfiltered samples

b - UL - upper layer 1570 L, LL-lower layer 1766 L when upper layer 2.0 m deep; upper layer 1178 L, lower layer 2158 L when upper layer 1.5 m deep

c - Samples taken after fourth and seventh atrazine additions

d - Samples taken one and two weeks after start of atrazine additions.

at least 85% of the total atrazine in the water was "soluble" as defined by the filtration method used (Table 4). Data on filtered samples was available for experiment II only.

11.5 CONCENTRATIONS AND BIOMAGNIFICATION FACTORS

Atrazine concentrations in the UL of both low treatments were comparable to concentrations (0-30 $\mu g \cdot L^{-1}$) measured during late June and early July in agricultural watersheds in southern Ontario (Frank *et al.*, 1978).

Concentrations of atrazine in experiment I were detectable in water samples only in the LT (Table 5). Concentration in unfiltered samples was 12.1 and 0.6 ppb in the UL and LL respectively. In the HT, residues were detected in all components. Water concentrations were 125 and 14.1 ppb in the UL and LL respectively while suspended particulates from the UL had a concentration of 2.5 ppm. Residues were not detected in *Daphnia magna* and were low in the guppies at 0.2 ppm. Highest residues were found in sedimented particulate matter (21.3 ppm). Biomagnification factors were calculated as concentration in component ÷ concentration in water of UL and were only 20x for UL suspended particulates, 170X for sedimented particulates and 2x for guppies.

In experiment II, atrazine concentrations in water at the LT were 20.5 and 1.9 ppb in the UL and LL respectively. In contrast to experiment I, residues were significant in the particulate matter and biota at the lower treatment level. The highest atrazine concentrations in the LT were in zooplankton (9.3 ppm) and UL suspended particulate (8.1 ppm) and lowest

Table 4. Atrazine concentrations in filtered[a] and unfiltered water samples

Time after 1st addition	Layer	Atrazine Concentrations in Water (ppb)			
		Low Treatment		High Treatment	
		Unfiltered	Filtered	Unfiltered	Filtered
7 days	upper	22.0	19.5	235.7	207.7
	lower	0.7	0.6	3.1	3.1
13 days	upper	20.5	18.5	221.4	210.7
	lower	1.9	1.1	7.6	6.7

[a]GF/C filters

Table 5. Concentrations[a] and biomagnification factors(B_f)[b] for atrazine in water, particulates and biota.

Component	Experiment I				Experiment II			
	Low Treatment		High Treatment		Low Treatment		High Treatment	
	Concentration	B_f	Concentration	B_f	Concentration	B_f	Concentration	B_f
Water (unfiltered)								
upper layer	12.1	-[d]	125.0	-	20.5	-	221.4	-
lower layer	0.6	-	14.1	-	1.9	-	7.6	-
Particulates								
upper layer	ND[c]	-	2.5	20x	8.1	395x	5.1	23x
lower layer	ND	-	$\leq$ 0.1	< 1x	1.4	68x	4.3	19x
sedimented	ND	-	21.3	170x	6.1	298x	69.8	315x
Daphnia magna	ND	-	ND	-	9.3	454x	(no animals)	
Guppies	ND	-	0.2	2x	0.4	20x	2.1	9x

[a] Water concentrations in ppb; particulates and biota ppm

[b] Biomagnification factors are relative to water concentration in upper layer

[c] Not detectable

[d] Not applicable

in LL particulates (1.4 ppm) and guppies (0.4 ppm). Biomagnification factors ranged from only 20x for guppies to ca. 400x for UL particulates and zooplankton.

Atrazine concentrations in unfiltered water from the UL (221.4 ppb) in the HT showed an increase in proportion to the treatment. Concentrations in biota and particulates did not show a similar proportional increase with the exception of sedimented particulate matter (69.8 ppm). Upper layer particulates had lower concentrations (5.1 ppm) than in the LT. Residues in guppies were ten times those found in guppies at the HT in experiment I but were lower than residues in other components. Biomagnification was only 10-20x for suspended particulates and fish at the HT in experiment II but about 300x for sedimented particulates. De-ethylated atrazine was undetectable in water, particulates and biota in experiment II.

Atrazine in samples from control columns was always low or undetectable indicating no significant contamination of these columns.

11.6 ATRAZINE UPTAKE FROM WATER

Atrazine was not accumulated by zooplankton exposed to 17 ppb in the water in exposures lasting up to seven days (Table 6). Residues in zooplankton were similar at 5.2 and 5.6 ppm after one and four day exposures to concentrations of 170 ppb of atrazine in the water and

Table 6. Atrazine concentrations in zooplankton and fish exposed to atrazine in water only

Days of Exposure	Atrazine Residues (ppm)					
	Daphnia			Guppies		
	Exposure Concentrations (ppb)					
	17	170	1700	17	170	1700
1	0.06	5.6	54.4	ND	1.8	9.5
4	ND[a]	5.2	26.6	0.12	1.8	16.8
7	ND	3.7	19.1	0.08	2.4	20.3

[a]Not detectable

less after seven days at 3.7 ppm. At the highest exposure concentration of 1700 ppb of atrazine in the water, zooplankton had residues of 54.4, 26.6 and 19.1 ppm after exposures of 1, 4 and 7 days.

Accumulation of atrazine from the water by guppies was low at 17 ppb and showed no trend with time of exposure. At 170 ppb of atrazine in the water residues in guppies were ca. 2.0 ppm for all exposure times. Residues increased from 9.5 ppm after one day of exposure to 20.3 ppm after seven days of exposure at a concentration of 1700 ppb in the water.

Residues of de-ethylated atrazine in guppies showed no trend with time of exposure in this experiment but were proportional to the exposure concentration. Residues were ca. 0.28 ppm at an exposure concentration of 170 ppb and varied between 2.0 and 3.0 ppm at 1700 ppb of atrazine in the water (Table 7).

12.0 DISCUSSION

Atrazine residues were detectable in the suspended particulates and biota in most of the treatments. However, concentrations of atrazine along the food chain, particulates → zooplankton → fish were low. Residues in guppies were typically less than in other components of the simulated ecosystem. Klaassen and Kadoum (1979) also found that in farm ponds receiving applications of atrazine, residues were detectable in all physical and biological components soon after application but biomagnification to higher trophic levels was absent.

Table 7. Concentrations of de-ethylated atrazine in guppies exposed to atrazine in water only.

Days of Exposure	De-ethylated Atrazine Residues in Guppies (ppm)		
	Exposure Concentrations (ppb)		
	17	170	1700
1	ND [a]	0.27	2.91
4	ND	0.29	3.31
7	ND	0.28	2.32

[a] Not detectable

Similarly, Yu et al. (1975) found that cyanizine, another triazine herbicide, and its degradation products remained in the water with minimal bioaccumulation. Macek et al. (1976) also concluded that fish concentrate very little atrazine, particularly in comparison to other contaminants such as DDT, mercury and PCBs which may be magnified thousands of times. Our own work (unpublished results) on transfer and accumulation of PCBs in these same lake column simulators has yielded biomagnification factors thousands of times greater than those found in the atrazine experiments.

Similar to our findings with atrazine, a proportionality between application and water concentration has also been demonstrated with simazine, another triazine herbicide (Mauck et al., 1976). The large pool of soluble atrazine in the UL of all treatments suggested that processes such as sedimentation, volatilization and adsorption to column walls had a minimal effect on distribution of atrazine in the columns.

The bench-scale experiment on atrazine uptake from water indicated that atrazine residues found in biota in the columns may have been partially the result of sorption equilibria with "soluble" atrazine. Biomagnification factors for fish of 10x were similar in both types of experiments. In the bench-scale experiment, residues were proportional to exposure concentration suggesting this was the most influential factor. Streit (1978) found that the invertebrates he studied accumulated atrazine quickly through sorption processes proportionate to the exposure concentration. Biomagnification factors in his study ranged from 2-50x at water concentrations of 10-100 ppb.

Generally, we observed no obvious toxic effects to biota in our experiments. Hollister and Walsh (1973) demonstrated significant inhibition of photosynthesis of several algae at atrazine concentrations similar to those in our highest treatment. In our experiments, neither change in algal standing crops nor reduced assimilation numbers in experiment II bore any relationship to treatments. Short-term toxic effects of atrazine to invertebrates has been demonstrated by other researchers but concentrations required tend to be much higher than in our highest treatment. Macek et al. (1976) estimated a 48-hour LC50 of 6.9 $mg \cdot L^{-1}$ for D. magna however, they found impairment of reproduction in chronic exposures of 30-90 days at much lower concentrations. These workers suggested a "maximum acceptable toxicant concentration" for this species of 0.140-0.250 $mg \cdot L^{-1}$. Schoba and Lampbert (1977) also found impaired growth and reproduction in D. pulex at sublethal concentrations > 1.0 $mg \cdot L^{-1}$. General observations and net hauls made at the end of each experiment did not indicate any relationship between treatments and zooplankton abundance. The population of D. magna in the HT of experiment II had peaked prior to atrazine treatment and was on the decline once treatments began. This explains their complete absence when samples were collected. Factors other than atrazine concentration such as abundance and quality of food may have been responsible for the decline.

Although atrazine use is heavy in agricultural land in Ontario the threat to the environment through short-term toxicity to aquatic organisms or accumulation of harmful residues in biota should be minimal with the exception of results of careless incidents, prevention of which is by obvious means.

13.0 REFERENCES

Analytical Methods Manual. 1975. Water Quality Branch, Inland Waters Directorate, Ottawa, Canada.

Chu, S.P. 1942. The influence of the mineral composition of the medium on the growth of planktonic algae. I. Methods and culture media. J. Ecol. 30: 284-285.

Edwards, C.A. 1975. Persistent pesticides in the environment. - CRC Press. 2nd ed.: 1-70.

Frank, R., H.E. Braun, G. Sirons, M.Holdrinet, B.D. Ripley, D. Onn and R. Coote.1978. Stream flow quality - pesticides in eleven agricultural watersheds in Southern Ontario, Canada, 1974-1977. Prepared for PLUARG, Task C (Canada) Activity 1. (project 80645, Education, Research and Special Services Division, Ontario Ministry of Agriculture and Food).

Hollister, T.A., and G.E. Walsh. 1973. Differential responses of marine phytoplankton to herbicides: Oxygen Evolution. Bull. Environ. Contam. Toxicol. 6(5): 291-295.

Klaassen, H.E. and A.M. Kadoum. 1979. Distribution and retention of atrazine and carbofuran in farm pond ecosystems. Arch. Environ. Contam. Toxicol. 8:345-353.

Macek, K.S., K.S. Buxton, S. Sauter, S. Gnilka and J.W. Dean. 1976. Chronic toxicity of atrazine to selected aquatic invertebrates and fishes. U.S. Environmental Protection Agency Office of Research and Development, Environmental Protection Agency Duluth, Minnesota. EPA-60013-76-047.

Mauck, W.L., F.L. Mayer and D.D. Holz. 1976. Simazine residue dynamics in small ponds. Bull. Environ. Contam. Toxicol. 16(1): 1-8.

Muir, D.C.G., J.Y. Yoo and B.E. Baker. 1978. Residues of atrazine and N-de-ethylated atrazine in water from five agricultural watersheds in Quebec. Arch. Environ. Contam. Toxicol. 7: 221-235.

Roller, N.F. 1975. Survey of pesticide use in Ontario, 1973. Economic Information. Ont. Ministry Agric. and Food, Toronto, Ontario.

Schober, U. and W. Lampert. 1977. Effects of sublethal concentrations of the herbicide Atrazin ® on growth and reproduction of Daphnia pulex. Bull. Environ. Contam. Toxicol. 17(3): 269-277.

Sirons, G.J., R. Frank and T. Sawyer. 1973. Residues of atrazine, cyanizine, and their phytotoxic metabolites in a clay loam soil. J. Agr. Food Chem. 21(6): 1016-1020.

Streit, B. 1978. Aufnahme, Anreicherung und Freisetzung organischer Pestizide bei benthischen. SuB wasserinvertebraten 1. Reversible Anreicherung von Atrazin aus der wasserigen Phase. - Arch. Hydrobiol. 1 Suppl. 55: 1-23

Thomson, J.N. 1973. Environmental Contaminants Inventory Study No. 1 Statistics on the use of pest control products in Canada. Environment Canada, Report Series No. 35.

Vollenweider, R.A. 1974. A manual on methods for measuring primary production in aquatic environments. IBP Handbook No. 12. Blackwell Scientific Publications, Oxford and Edinburgh.

Yu, C.C., G.M. Booth and J.R. Larsen. 1975. Fate of triazine herbicide cyanizine in a model ecosystem. J. Agric. Food Chem. 23(15): 1014-1015.