Chemosphere 294 (2022) 133769

Contents lists available at ScienceDirect

Chemosphere

journal homepage: www.elsevier.com/locate/chemosphere

Characterizing the exposure of streams in southern Ontario to agricultural pesticides

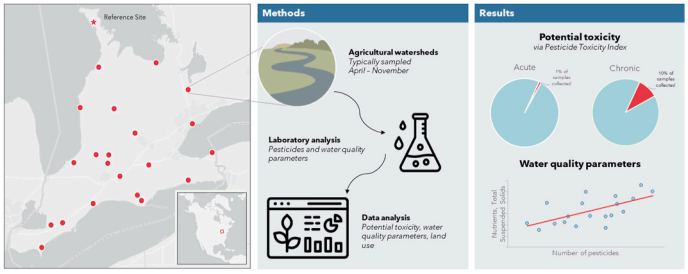
Melanie Raby [a,*], Linda Lissemore [b], Georgina Kaltenecker [a], Denise Beaton [c], Ryan S. Prosser [d]

[a] Ontario Ministry of the Environment, Conservation and Parks, Toronto, ON, Canada
[b] Laboratory Services Division, University of Guelph, Guelph, ON, Canada
[c] Ontario Ministry of Agriculture, Food, and Rural Affairs, Guelph, ON, Canada
[d] University of Guelph, School of Environmental Sciences, Guelph, ON, Canada

HIGHLIGHTS

- Pesticides can transport to streams where aquatic communities may be exposed.
- Water samples from ≥18 streams were analyzed for pesticides from 2012 to 2019.
- Frequently detected pesticides included neonicotinoid insecticides and herbicides.
- Pesticides detected were related to upstream land use and crops.
- Hazard to aquatic communities was found to generally be low, with some exceptions.

GRAPHICAL ABSTRACT



ARTICLE INFO

Handling Editor: Myrto Petreas

Keywords:
Agricultural pesticides
Invertebrate toxicity
Pesticide toxicity index
Watershed
Stressor

ABSTRACT

Aquatic communities can be exposed to pesticides transported from land. Characterizing this exposure is key to predicting potential toxic effects. In this study, samples of streamwater from 21 sites were used to characterize pesticide exposure to aquatic communities. Sites were in agricultural areas of southwestern Ontario, Canada and were sampled monthly from 2012 to 2019 from April to November. Samples were analyzed for a suite of hundreds of pesticides and pesticide degradation products and other water quality indicators (e.g., nutrients). Frequently detected pesticides included herbicides (2,4-D; bentazon; MCPP; metolachlor) and neonicotinoid insecticides (NNIs) (clothianidin; thiamethoxam) which were detected in >50% of samples collected between 2015 and 2019. Non-metric multidimensional scaling (NMDS) was used to explore connections between pesticide concentrations and upstream land use and crop type. Detectable concentrations of the NNI clothianidin and many herbicides were related to corn, soybean, and grain/cereal crops while concentrations of the NNI imidacloprid, insecticide flonicamid, and fungicide boscalid were related to greenhouse/nursery land use. Potential toxicity to aquatic communities was assessed by comparing pesticide concentrations to Pesticide Toxicity Index (PTI) values. Few samples exceeded levels where acute (1% of samples) or chronic toxicity (10.5%) would be expected. The diamide insecticide chlorantraniliprole was detected in several streamwater samples at levels that may cause toxicity to aquatic invertebrates, highlighting the need for continued toxicity research into this

* Corresponding author. Environmental Monitoring and Reporting Branch, Ontario Ministry of the Environment, Conservation and Parks, 125 Resources Rd, Etobicoke, ON, M9P 3V6, Canada.
E-mail address: melanie.raby@ontario.ca (M. Raby).

https://doi.org/10.1016/j.chemosphere.2022.133769
Received 2 November 2021; Received in revised form 24 January 2022; Accepted 25 January 2022
Available online 28 January 2022

pesticide class. The number of pesticides detected was positively correlated with nutrient and total suspended solids levels, underscoring the multiple stressors aquatic communities are exposed to in these habitats.

1. Introduction

Pesticide use plays an important role in maximizing productivity in many types of agriculture (Headley, 1968; Sexton et al., 2007). Pesticides used in agriculture can travel from the field of application to surface water through spray drift, run-off due to a precipitation or irrigation event, and/or leaching (Traub-Eberhard et al., 1994; Williams et al., 1995; Schulz et al., 2001). The propensity of a pesticide to move into surface water is a factor of physicochemical properties (e.g., solubility in water, ability to sorb to soil, half-life) (Nicholls, 1988; Brown and van Beinum, 2009), mode of application, and environmental conditions. When pesticides enter surface water, they can pose a risk to various components of aquatic ecosystems, e.g., primary producers, macroinvertebrates, fish (Gustavsson et al., 2017; Szöcs et al., 2017). An important component of assessing the risk of pesticides to aquatic ecosystems is being able to characterize the exposure of the ecosystem (Solomon et al., 2000). Characterizing exposure involves measuring the concentrations of pesticides in aquatic ecosystems (i.e., water, sediment, tissue) and understanding how the concentrations of pesticides in the various matrices in the aquatic ecosystems change over time (e.g., acute, chronic, episodic) (Solomon et al., 1996; Williams et al., 2014).

To this end, many jurisdictions have implemented monitoring programs to collect data on the exposure of aquatic ecosystems to pesticides, including Germany (Arle et al., 2016), Italy (Finizio et al., 2011), Sweden (Boye et al., 2019), and the United States (USGS, 2020). In the Canadian province of Ontario, the Ontario Ministry of Agriculture, Food, and Rural Affairs (OMAFRA) and the Ontario Ministry of the Environment, Conservation, and Parks (MECP) collaborate to administer a pesticide monitoring program in streams throughout the province. Most of these monitoring stations are located in the southern portion of the province where most agricultural land uses are found. The collection and measurement of pesticides in water samples is only the first step in pesticide monitoring. An equally important component of a monitoring program is the analysis of the data collected to determine whether the number and concentration of pesticides in water bodies are changing over time and whether the pesticides detected pose a risk to aquatic ecosystems (Schreiner et al., 2016; Di Guardo and Finizio, 2018; Spycher et al., 2018; Chow et al., 2020). The monitoring data can also provide insight on how land use in the upstream catchment of monitoring stations influences the type and concentration of pesticides detected in water samples. (Black et al., 2000; Glozier et al., 2012).

Consequently, the objective of this study was to use the data from the MECP/OMAFRA monitoring program collected in Ontario from 2012 to 2019 to describe the occurrence and concentration of pesticides in streams located in various watersheds across southern Ontario. This study also investigated the relationships between pesticide concentrations and other water quality indicators along with land use in the upstream catchment. In addition, the monitoring data was used to assess the potential hazard of pesticide exposure to aquatic communities using the Pesticide Toxicity Index (adapted from Nowell et al., 2014). The analysis conducted in this study is instrumental to assessing the exposure of Ontario's streams to pesticides and the associated risk to aquatic ecosystems.

2. Methods

2.1. Site selection and descriptions

Streamwater samples were collected from a total of 20 study streams and one reference stream in southern Ontario (Fig. 1). Monitoring sites were chosen to represent a wide geographic coverage and a broad mix of agricultural activities. Upstream catchments ranged in size from 15 km^2

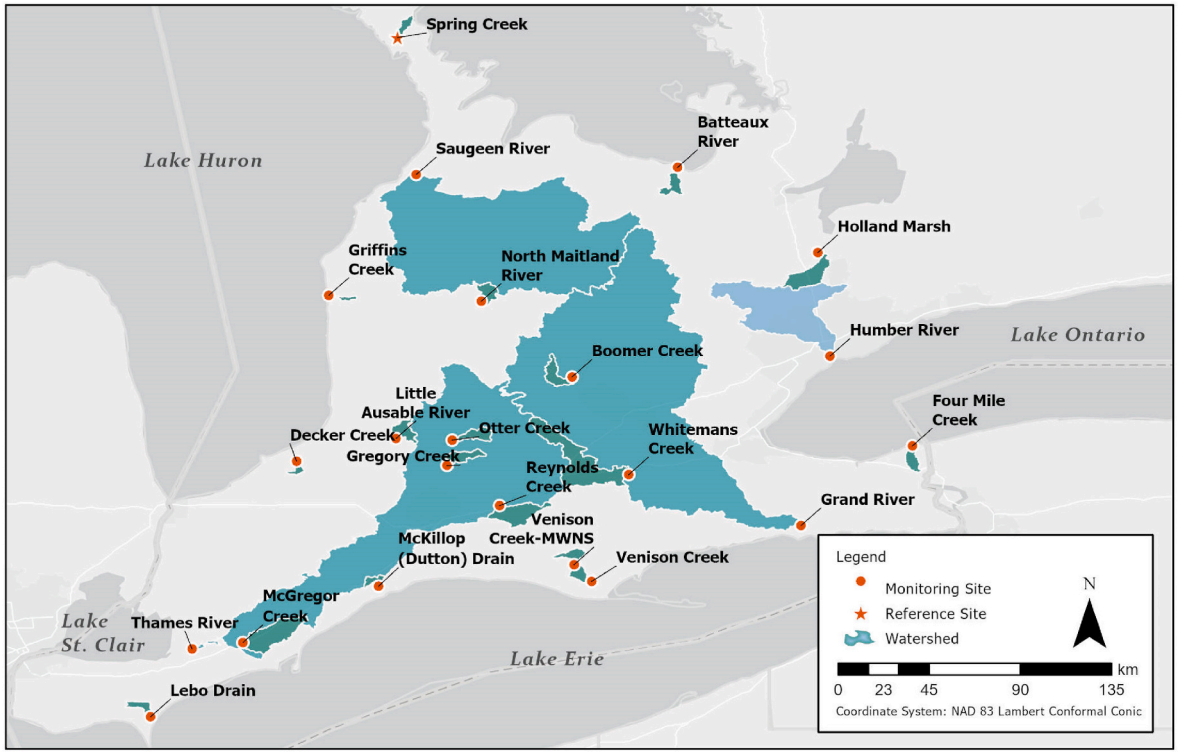


Fig. 1. Map of southern Ontario (Canada) showing the sites that are part of the MECP/OMAFRA pesticide monitoring program.

to 6312 km^2, with 60% of catchments <100 km^2, and 85% < 1000 km^2 (Supplemental Data Table S1). All monitoring sites had >60% agricultural land use in their upstream catchment, except for the reference site at Spring Creek and two sites with partial urban influence in the Humber River and Holland Marsh. The reference site Spring Creek was located outside a National Park and drained mainly forested land.

2.2. Stream water sampling

Stream water samples were collected by Conservation Authorities 1–8x/year during the ice-free months (March–November) from 2012 to 2019. Sampling was typically concentrated during the growing and pesticide application season, with most samples taken during the months of May–October. Samples were collected at approximately 0.3–0.4 m stream depth in multiple pre-cleaned 1 L amber glass bottles and shipped on ice to the University of Guelph Laboratory Services Division for analysis of dissolved pesticides. Samples were not filtered. Sites were often (two-thirds of the time) concurrently sampled for general water chemistry (in-stream measurements of pH, conductivity, dissolved oxygen and water temperature, and grab samples for laboratory analysis of total nitrogen, nitrate, nitrite, ammonium, phosphate, total suspended solids, and at some sites, metals) under the Provincial Water (Stream) Quality Monitoring Network (PWQMN) (https://www.ontario.ca/environment-and-energy/map-provincial-stream-water-quality-monitoring-network, see Supplemental Data Table S2 for sampling details). Samples were collected on an approximately monthly basis and were typically collected based on a schedule rather than weather or stream conditions, although some samples may have been collected during wet-weather flow events. In particular, samples from Gregory Creek, Otter Creek, and Reynolds Creek may have been collected during or after wet-weather flow events and these concentrations may be underestimated due to dilution or overestimated due to wet-weather flushing.

2.3. Pesticide analysis

Water samples were analyzed for pesticides and pesticide degradation products using both gas and liquid chromatography coupled with tandem mass spectrometry at the University of Guelph's Agriculture and Food Laboratory (AFL). The AFL is a Canadian Association of Laboratory Accreditation (CALA) and International Organization of Standardization and International Electrotechnical Commission (ISO/IEC 17025) accredited laboratory. In 2012, each sample was analyzed for 48 pesticides; by 2019, more than 500 pesticides were measured in each water sample using three different analytical methods. The following describes the current analytical methodologies as of 2019.

The first method screened water samples for 512 analytes (parent, metabolites and isomers) using liquid chromatography/electrospray ionization-tandem mass spectrometry (LC/ESI-MS/MS) and gas chromatography/electron impact ionization-tandem mass spectrometry (GC/EI-MS/MS). Pesticides were extracted from water using the technique known as the quick, easy, cheap, effective, rugged, and safe (QuEChERS) approach which applies dispersive solid phase extraction (d-SPE). A sub-sample of the water sample was extracted into acidified acetonitrile in the presence of anhydrous sodium acetate and magnesium sulfate. The supernatant was split, concentrated, and then diluted into either methanol/ammonium acetate (for LC analysis) or hexane (for GC analysis). The reconstituted extracts were analyzed in positive ion mode using both a SCIEX 5500 ESI-MS/MS coupled with an Agilent 1260 HPLC and an Agilent GC Quadrupole 7890A GC-MS/MS (electron impact ionization). The second method measured phenoxy acid herbicides and neonicotinoid insecticides (NNIs) in water. Prior to extraction, the water sample was acidified using sulfuric acid and separated from co-extractives using a reverse phase polymeric solid phase extraction (SPE) column. The pesticides were eluted from the column using methanol and then concentrated and analyzed in both positive and negative ionization modes using a SCIEX 5500 ESI-MS/MS. This method screened for 9 NNIs (positive ion mode) and 19 phenoxy acid herbicides (negative ion mode; except dithiopyr in positive ion mode). The third method measured glyphosate and its main degradation product aminomethylphosphonic acid (AMPA) in water samples. Prior to extraction, the water was acidified using phosphoric acid and separated from co-extractives using a PS/DVB SPE column, suited to the extraction of polar organic residues. The final eluant was analyzed in positive ionization mode using a SCIEX 4000 ESI-MS/MS coupled with an Agilent 1260 HPLC. For all three methods, several data points were used to confirm the presence of a pesticide in the sample: retention time, M + H target mass, two fragment qualifier ions, and the ratio of the two fragment ions.

For the identification and quantification of the compounds, all three methods utilized deuterium labelled internal standards, and either matrix-matched or solvent based five-point calibration curves. A total of ten deuterated standards were used including acetamiprid-d3, bentazon-d7, carbendazim-d4, carbofuran-d3, clothianidin-d3, imidacloprid-d4, thiabendazole-d6, thiamethoxam-d3, glyphosate-d2 and AMPA-d2. Internal standards were used to correct for matrix impacts on quantification and were added at the beginning of the method and carried through the entire extraction process. Method performance was monitored for analyte recovery using fortified quality control and reagent blank, matrix blank (environmental water) and duplicate samples in all analysis. Duplicate samples results were evaluated for consistency. The method detection limit (MDL) and method quantitation limit (MQL) for each pesticide are provided in Supplemental Data Table S3. The MDL for a number of pesticides has decreased from 2012 to 2019.

2.4. Data availability and analysis

Dissolved pesticide concentrations and general water chemistry (Provincial Water (Stream) Quality Monitoring Network) data and associated metadata, including MDLs and MQLs, are available online through Ontario Open Data Portal (https://data.ontario.ca/organization/environment-conservation-and-parks). All data analysis was completed in R v.3.6.3/RStudio v.1.1.463 and associated packages as described. Hypothesis tests were conducted with $\alpha = 0.05$.

The number of analytes increased over the study period from 48 in 2012 to 512 in 2019. Method detection and quantitation limits changed over the study period, with notable decreases in NNIs MDLs of an order of magnitude. Limits are provided in Supplemental Data Table S3. Pesticide results reported above the MDL, including those reported as less than the MQL were treated as detections. Detection frequencies of individual pesticides were calculated for the years 2012–2014 and 2015–2019 to account for a data artefact produced by the lowering of many MDLs and MQLs in 2014/2015. Descriptive statistics were calculated for frequently detected pesticides using left-censored data techniques to account for censored data (i.e., data less than MDL or MQL). Summary statistics (mean, median concentrations) were estimated using robust regression-on-order statistics (robust ROS) via the R 'NADA' package (v. 1.6–1) (Bolks et al., 2014; Lee, 2017), and confidence intervals around the mean were estimated using bootstrapping with 1000 iterations (Bolks et al., 2014).

Relationships between the number of pesticides detected and agricultural land use were assessed using the Kendall rank correlation test in the R package 'ggpubr' (Kassambara, 2020). Non-metric multidimensional scaling (NMDS) was used to explore the relationships between land use and mean pesticide concentrations. NMDS is a type of ordinal scaling that represents data along a set number of axes while preserving the rank-order relationships among them (Bocard et al., 2018). To gather land use data, upstream drainage basins were characterized using the Ontario Flow Assessment Tool (https://www.ontario.ca/page/watershed-flow-assessment-tool), and the resulting shapefiles were then used to calculate annual land use using Agriculture and Agri-Food Canada's (AAFC) Annual Crop Inventory (ACI) (AAFC, 2020). In 2009,

AAFC started producing digital maps of crop type using optical (Landsat-5, AWiFS, DMC) and radar (Radarsat-2) based satellite images (AAFC, 2020). Land use classifications changed from year-to-year and became more specific over time. To standardize land use across time and sites, individual land use types in acres were standardized to a percentage of total land use and were summarized into broader categories for the NMDS (Supplemental Data Table S4). Yearly data was then averaged to obtain mean land use over the 2015–2019 time period to match the time period with the most robust pesticide data. Pesticide data was first subset to include only the most frequently detected pesticides using a cut-off of >10% detection in 2015–2019 samples. One pesticide, imazethapyr, met this cut-off but was removed as it was only added as an analyte in 2018 and therefore had a fewer number of samples. Mean pesticide concentrations as calculated via ROS were used in the NMDS. Where mean concentrations could not be calculated due to <3 observations, a value of zero was assigned to obtain a complete data matrix for use in NMDS. Mean pesticide concentrations were then standardized to a value between 0 and 1 using the 'decostand' function and 'range' argument in the R package 'vegan' (Oksanen et al., 2019). NMDS was run on a matrix of mean pesticide concentrations for each site using the 'metaMDS' function in 'vegan' using the Gower distance metric, a maximum of 20 random starts, and 2 specific axes. Pesticides and land use vectors were fit on to the NMDS ordination with the 'envfit' function in the R package 'vegan' (999 permutations) (Oksanen et al., 2019).

Relationships between individual pesticide concentrations and other water quality indicators (dissolved nutrients, total suspended solids) were assessed using the Kendall rank correlation coefficient (tau) and associated p-value computed for singly or doubly censored data via the 'cenken' function in the R 'NADA' package (Lee, 2017). Correlations were restricted to paired pesticide and water chemistry data for samples collected from 2015 to 2019. A secondary analysis was conducted for only samples collected during the planting season (May and June) during these years to assess relationships when pesticide application is typically at the highest level.

The Pesticide Toxicity Index (PTI) is used as a screening-level tool to assess potential toxicity to fish, cladocerans, and benthic invertebrates as a result of exposure to a mixture of measured pesticides (Nowell et al., 2014). The PTI method scales observed chemical concentrations against a measure of toxicity or effect in a toxic unit (TU) approach. Toxic unit values for each pesticide are summed to give the PTI (Nowell et al., 2014). In this study we used the existing PTI database in Nowell et al. (2014) and amended it to include potential toxicity to primary producers as results showed herbicides were frequently detected in streams. We included a sensitive toxic concentration for a green algae (typically a 2 to 5-d EC50 for population abundance for *Raphidocelis subcapitata*, formerly known as *Pseudokirchneriella subcapitata*) and a macrophyte (typically a 7 to 14-d EC50 for population abundance for *Lemna minor* or *L. gibba*) for the 12 herbicides that were detected in >10% of samples (2, 4-D; 2,4-DP; acifluorfen; atrazine; bentazon; dicamba; imazethapyr; MCPA; MCPP; metolachlor; metribuzin; triclopyr). Toxic concentrations were sourced from the ECOTOX database, and priority was given to data derived using active ingredient over a pesticide formulation, and to data for standardized test durations (4 to 5-d for algae, 14-d for *Lemna* sp.) (Supplemental Data, Table S5). The sensitive PTI score was calculated for each sample according to equation (1), where E_i is the concentration of pesticide *i*, n is the number of detected pesticides in the sample, and STC_{it} is the sensitive toxic concentration for the pesticide *i*, for the taxa, *t*.

$$Sensitive - PTI_t = \sum_{i=1}^{n}\left(\frac{E_i}{STC_{i,t}}\right) \quad \text{Equation 1}$$

The sensitive PTI score was compared to two thresholds as used by Nowell et al. (2017): a threshold of 1 for acute toxicity and 0.1 for chronic toxicity, with toxicity expected where PTI scores exceeded the threshold. The PTI is based on acute toxicity data and not sub-lethal endpoints (Nowell et al., 2014); the use of a threshold of 0.1 for chronic toxicity is comparable to assuming an acute-to-chronic ratio of 10:1. Additionally, the pesticide resulting in the largest TU as part of the PTI score was identified and TU_{max}. Results reported as less than MDL or MQL were treated as zero when calculating the PTI and are therefore not included in the toxicity assessment. Where detected pesticides did not have an associated toxic concentration given in Nowell et al. (2014), that pesticide was not included in the PTI score. While this approach could underestimate the PTI score, few pesticides that were frequently detected and quantified had missing toxic concentrations for benthic invertebrates, cladocerans, or fish (notable pesticides with missing concentrations were MCPP, MCPA, 2,4-DP, triclopyr, flonicamid, and imazethapyr). Potential differences in PTI scores between the years 2012–2014 and 2015–2019 were tested via a Wilcoxon test ($\alpha = 0.05$).

3. Results and discussion

3.1. Pesticide occurrence and connections to land use

A total of 893 samples were collected across the 21 study sites from 2012 to 2019 (Supplemental Data Table S6), with 90 pesticides (parent or degradation products) detected in at least one sample (40 herbicides, 26 fungicides, and 24 insecticides). Most detected pesticides were registered for use in Canada, however infrequent detections of pesticides not registered for use in agriculture (i.e., dicloran, fenuron, propazine, propham, 2,4,5-T, 2,4,5-TP, DDT and metabolites, dinotefuran, and methamidophos) did occur in 2% of samples. Some were legacy pesticides which were once used but are no longer registered such as fenuron, propham, DDT, 2,4,5-T and methamidophos, in which case detection is likely the result of legacy contamination or transformation in the environment, while others may be due to unregistered use or in applications other than agriculture. For example, formulations containing dinotefuran are registered in Canada for veterinary and industrial pest control uses but not for use in agriculture (PMRA, 2020). Pesticides were rarely detected at the Spring Creek reference site, which is likely due to the upstream catchment of this site being dominated by forested land. Four samples were collected at Spring Creek from 2017 to 2018; pesticides were not detected above the MDL in two of the samples, while two samples contained detectable levels of bentazon, MCPA, thiamethoxam, and/or 2,4-D, but these pesticides were often below the MQL. One sample contained a detectable residue of 0.0017 μg/L of 2,4-D, which is 100x lower than the mean concentration at sites with intensive agriculture.

Detection frequencies were calculated for each pesticide over two time periods: 2012–2014, and 2015–2019. The most frequently detected pesticides are presented in Table 1. Of the 292 samples collected from 2012 to 2014, 215 (74%) contained at least one detected pesticide. The herbicide 2,4-D was the most frequently detected pesticide (42%), followed by the herbicides, metolachlor (38%), bentazon (34%), atrazine (34%), MCPP (23%), and MCPA (16%). Of the 601 samples collected from 2015 to 2019, all contained at least one detectable pesticide. The herbicide 2,4-D was again the most frequently detected pesticide (92% detection frequency), followed by bentazon (90%), clothianidin (88%), thiamethoxam (87%). MCPP (65%), metolachlor (54%). The observed increase in both the number and frequency of pesticides detected may be due to the lowering of detection limits and addition of new analytes in 2014–2015, rather than an actual increase in pesticides in streams from an increase in application. The increased detection of clothianidin and thiamethoxam is likely due to improved analytical methodology which lowered MDLs by an order of magnitude in 2015. Summarized results for each site are available in Supplemental Data Tables S7 to S24.

Most stream water samples contained pesticide mixtures of two or more detected pesticides. In samples taken from 2012 to 2014, 60% contained two or more detected pesticides, and 45% contained three or more. With the introduction of lowered detection limits in 2015, some pesticides were more frequently detected. In samples collected between 2015 and 2019, 99% contained two or more, and 52% contained eight or

Table 1
Summary of pesticide detections and concentrations across all sites for pesticides detected in >10% of all samples.

Pesticide	Acronym	2012–2014[a]		2015–2019[b]		All years: 2012–2019[c]		
		Detection limit(s) (μg/L)	Detection frequency (%)	Detection limit(s) (μg/L)	Detection frequency (%)	Estimated median concentration[d]	Estimated mean concentration with 95% CI (μg/L)[d]	Maximum concentration (μg/L) and site (year)
2,4-D	2,4-D	0.01	42	0.0006	92	0.014	0.111 (0.073–0.155)	13, McGregor Creek (2017)
Bentazon	BTZ	0.01	34	0.0008	90	0.008	0.044 (0.036–0.054)	2.2, Otter Creek (2012)
Clothianidin	CLO	0.084	9.8	0.002	88	0.015	0.036 (0.038–0.044)	0.97, McKillop Drain (2015)
Thiamethoxam	THX	0.09	9.8	0.0006	87	0.010	0.051 (0.043–0.061)	1.7, McKillop Drain (2015, 2016)
MCPP	MCPP	0.03; 0.01	23	0.0007	65	0.003	0.014 (0.010–0.018)	0.51, Holland Marsh (2019)
Metolachlor	MET	0.1	38	0.03	54	0.045	0.483 (0.314–0.706)	77, Reynolds Creek (2017)
Imidacloprid	IMI	0.126	6.2	0.0006	54	0.001	0.058 (0.030–0.100)	11, Lebo Drain (2012)
MCPA	MCPA	0.03; 0.01	16	0.0007	48	0.0005	0.033 (0.015–0.058)	8.7, Gregory Creek (2019)
Dicamba	DIC	0.02; 0.01	11	0.002	31	0.001	0.123 (0.064–0.204)	25, McGregor Creek (2012)
Atrazine	ATZ	0.1	34	0.07	27	0.022	0.249 (0.183–0.332)	18, McKillop Drain (2015) and Otter Creek (2019)
Imazethapyr	-	NA	NA	0.005	22	0.003	0.009 (0.006–0.012)	0.19 (Reynolds Creek, 2019)
2,4-DP	2,4-DP	0.02; 0.01	9.4	0.0007	20	0.0001	0.007 (0.003–0.012)	1.7, Lebo Drain (2015)
Triclopyr	TRI	0.03	4.5	0.001	19	0.0001	0.024 (0.006–0.057)	14, McGregor Creek (2016)
Boscalid	BSC	NA	NA	0.1	12	0.032	0.102 (0.081–0.141)	3.9, Lebo Drain (2015)
Flonicamid	FLO	NA	NA	0.1; 0.002	11	0.0001	0.053 (0.028–0.085)	6.6, Lebo Drain (2015)
Metribuzin	MET	0.1	4.5	0.05	11	0.004	0.079 (0.055–0.111)	7.2, Decker Creek (2018)
Acifluorfen	ACI	0.01	4.0	0.001	10	0.00004	0.0006 (0.0004–0.001)	0.067, Decker Creek (2019)

Note: Pesticides were considered detected if they were measured above the method detection limit.
[a] N = Only samples collected during years 2012–2014; 292 samples analyzed for all pesticides.
[b] N = Only samples collected during years 2015–2019; 601 samples analyzed for all pesticides.
[c] N = All samples collected during years 2012–2019; 893 samples analyzed for all pesticides.
[d] Concentrations estimated via robust regression-on-order statistics (ROS).

more. The sites with the highest number of detected pesticides were in Lebo Drain (median 17 detected pesticides), Holland Marsh (median 13), McGregor Creek and Thames River (median 11), and Reynolds Creek and Gregory Creek (median 10). The most pesticides detected in a single sample was 23 in a sample from Holland Marsh in summer 2018. The most common pesticide mixture detected were the NNIs clothianidin and thiamethoxam which were detected together in 81% of samples collected from 2015 to 2019. Clothianidin is known to be a breakdown product of thiamethoxam, and their co-occurrence has been previously demonstrated (Struger et al., 2017). Neonicotinoids are commonly used as seed treatments on corn and soybean (Douglas and Tooker, 2015). The most recent seed sale data available online through Ontario Open Data Portal shows half of all corn and a quarter of all soybean seed sold in Ontario during the years 2016–2017 was treated with clothianidin or thiamethoxam (Ontario MECP, 2018). Corn and soybean are the most popular crops in most of the sub-watersheds sampled (see breakdown of major crop type for each site in Supplemental Data Table S1). Other frequently detected mixtures included bentazon and the NNIs clothianidin and thiamethoxam (74–78% of samples) (Supplemental Data Table S25).

A significant moderate correlation was observed between the percentage of land use in the sub-watershed used for agriculture and the number of pesticides detected in a given sample for both the time periods 2012–2014 (tau = 0.57, $p < 0.001$) and 2015–2019 (tau = 0.37, $p = 0.034$) (Supplemental Data Figure S1). Non-metric multidimensional scaling (NMDS) was used to further explore the relationship between pesticides and watershed land use. Stress of the NMDS was 0.095. In NMDS plots in Fig. 2, the distance between points approximates the rank order of distance between the mean pesticide concentrations, so points closer together in space have higher similarities. NMDS revealed similarities between mean dissolved pesticide concentrations in streams and commonly used pesticides on corresponding crops in the watershed. The neonicotinoid insecticide (NNI) clothianidin, and herbicides (atrazine, metribuzin, metolachlor, MCPA, and bentazon) ordinated in similar space to corn, soybean, and grain and cereal crop land use (Fig. 2), meaning there was similarity between these datasets. The pesticides that ordinated with corn and soybean crops can be partially explained by patterns of pesticide use in Ontario. From 1973 to 2014, OMAFRA organized a farmer survey of agricultural pesticide use in Ontario every 5 years (Farm & Food Care Ontario, 2015). The results of the most recent survey conducted in 2013/2014 showed that the herbicides that were most readily used on corn and soybean in the province by mass were glyphosate (1,151,051 kg on corn, 1,544,954 kg on soybean), s-metolachlor (547,774 kg on corn, 187,581 kg on soybean), atrazine (293,208 kg on corn, not registered on soybean), and bentazon (341 kg on corn, 13,845 kg on soybean (Farm & Food Care Ontario, 2015). Glyphosate and its primary degradation product AMPA were detected in <10% of water samples from this study, even though a larger mass of glyphosate was applied to fields in Ontario than any other pesticide. Glyphosate constituted 54% of the mass of pesticide used across all types of crops in 2013/2014 according the survey (Farm & Food Care Ontario, 2015). The low frequency of detection of glyphosate in the water samples (4% of 292 samples from 2012 to 2014 and 9% of 601 samples from 2015 to 2019) collected for this study, despite its relatively high use, may be due to glyphosate's relatively high affinity to bind to soil (Mamy and Barriuso, 2005; Okada et al., 2016). A large proportion of glyphosate

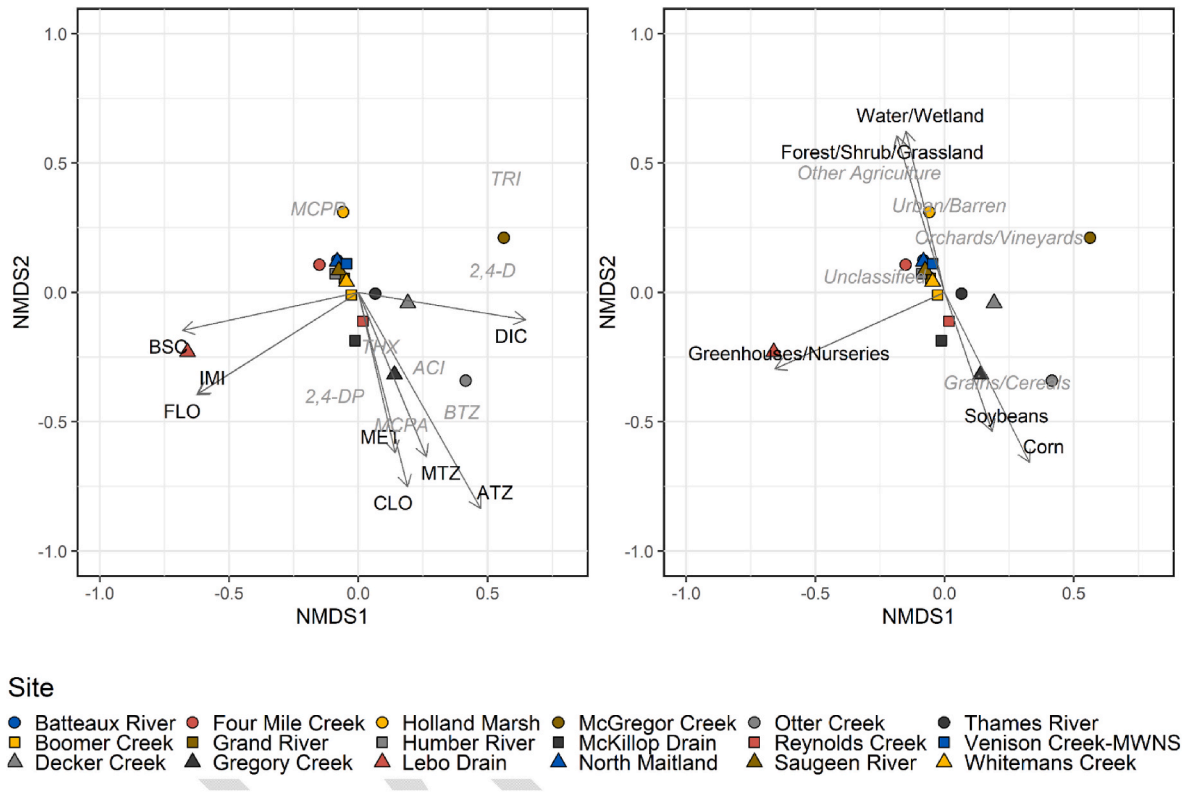


Fig. 2. Non-metric multidimensional scaling (NMDS) plot of mean dissolved pesticide concentrations and upstream land use. Fitted pesticide and land use factors in grey were not significant ($p > 0.05$) however are plotted for qualitative visualization; those in black were significant ($p < 0.05$). Acronyms are as follows and listed in Table 1: ACI = acifluorfen; ATZ = atrazine; BSC = boscalid; BTZ = bentazon; CLO = clothianidin; DIC = dicamba; FLO = flonicamid; IMI = imidacloprid; MET = metolachlor; MTZ = metribuzin; THX = thiamethoxam; TRI = triclopyr.

residues are retained in the field due to binding to soil particles and the glyphosate that does travel to surface water tends to bind to organic matter or sediment particles, and not be dissolved in the water column so dissolved concentrations would be expected to be low (Maqueda et al., 2017). Additionally, a relatively high method detection limit for glyphosate in this study of 20 μg/L from 2012 to 2016, and 1 μg/L from 2017 to 2019 may have masked lower concentrations. Environment and Climate Change Canada's Great Lakes Water Quality Surveillance program that monitors pesticide levels in Ontario tributaries detected glyphosate in 50% of samples from 2002 to 2016 with a method detection limit of 0.00017 μg/L (ECCC, 2020). NNIs were not reported in the farmer survey for any field crops, which is likely due to seed treatments not being considered in the survey (Farm & Food Care Ontario, 2015). As NNIs are commonly used as seed treatments for field crops (Douglas and Tooker, 2015) it is not unexpected for clothianidin and thiamethoxam in water samples to be associated with the cultivation of corn in the upstream catchment.

The pesticides boscalid, flonicamid, and imidacloprid ordinated in similar space as greenhouse/nursery land use (Fig. 2). Flonicamid was not reported in the Ontario pesticide survey conducted in 2013/2014, potentially because it was a relatively new insecticidal active ingredient at the time (Farm & Food Care Ontario, 2015). It was first registered for use in Canada in 2010 and currently there is only one product with flonicamid as the active ingredient registered in Canada (BELEAF 50SG, ISK Biosciences Corp.) (PMRA, 2020). BELEAF 50 SG is registered for use on flax, field vegetables, pome & stone fruit, outdoor ornamentals, and Christmas trees (PMRA, 2020), so it is not surprising that flonicamid in water samples was associated with greenhouses and nurseries in the upstream catchment (Fig. 2).

Non-agricultural land uses (forest/shrub/grassland, water/wetland, urban/barren) and other agricultural crops (e.g., canola, vegetables, berries, pulses, sod, tobacco, and others; see Supplemental Data Table S4) ordinated oppositely (and therefore were most dissimilar) to most pesticides and soybean, corn, grains/cereals and greenhouses/nursery agricultural land uses. A number of studies have observed that a greater proportion of natural land use (forest/shrubland/grassland; lower proportion of agricultural land use) in the upstream catchment can relate to a lower exposure of streams to pesticides (Schriever et al., 2007; Bunzel et al., 2014; Sanford and Prosser, 2020). The herbicides MCPP (also known as mecoprop), 2,4-D, and triclopyr had positive NMDS2 scores and ordinated along with non-agricultural land use. MCPP is a selective herbicide that is registered for use on turfgrass and field crops to control broadleaf weeds and is often formulated with other phenoxy herbicides (e.g., 2,4-D, MCPA, dicamba) (PMRA, 2020). MCPP was not listed in the survey on agricultural pesticide use in Ontario conducted in 2013/2014 which is expected as herbicide formulations containing MCPP are mainly used on turfgrass (e.g., golf courses) in Ontario (Farm & Food Care Ontario, 2015). While golf course land use was not specifically categorized in this study, given the relatively large size of some watersheds and the prevalence of golf courses in Ontario, it is likely there were golf courses in some upstream catchments that may have been captured in the 'urban/barren', 'other agriculture' or 'forest/shrub/grassland' land use categories. Triclopyr is a selective herbicide registered for use in forest and woodland management areas to control broadleaf weeds (PMRA, 2020). Like MCPP, triclopyr is used outside of agricultural applications on land uses that may have been captured in the 'urban/barren', or 'forest/shrub/grassland' land use categories. 2,4-D is a widely used herbicide for many applications that could span a number of land use categories. The ordination location of 2,4-D in 'neutral' space between land uses was therefore expected.

It is important to acknowledge that there are a number of potential explanatory variables that could explain the concentration of pesticides in water which have not been considered due to a lack of data. Other studies have shown data on the quantity and timing of pesticides used in

the upstream catchment correlates directly with the type and concentration of pesticides observed in streams (Kreuger, 1998; Hunt et al., 2006; Stackpoole et al., 2021). However, this data is not collected in Ontario. As mentioned, Ontario relies on a voluntary farmer survey to collect data on the type and magnitude of pesticides used in the province and this survey was not conducted as scheduled in 2018 due to low participation from a number of sectors of the agricultural industry (Farm & Food Care Ontario, 2015). This study also did not incorporate data on precipitation, soil type, extent of riparian buffer, and extent of tile drainage in agricultural fields in the upstream catchment of each sampling site. These factors play an important role in pesticide travelling from the field of application to surface water (Novak et al., 2001; Fortin et al., 2002; Montoya et al., 2006; Tran et al., 2010; Wettstein et al., 2016; Schmidt et al., 2019; Prosser et al., 2020). Some of these data types were not incorporated in this study due to availability and/or robustness of the data for the area covering our study sites. Collecting this type of data across a large province can be logistically challenging, however, this study underscores the importance of collecting this type of data to gain insight on pesticide exposure to streams.

3.2. Correlation between dissolved pesticide concentrations and additional water quality variables

Significant correlations were observed between pesticide

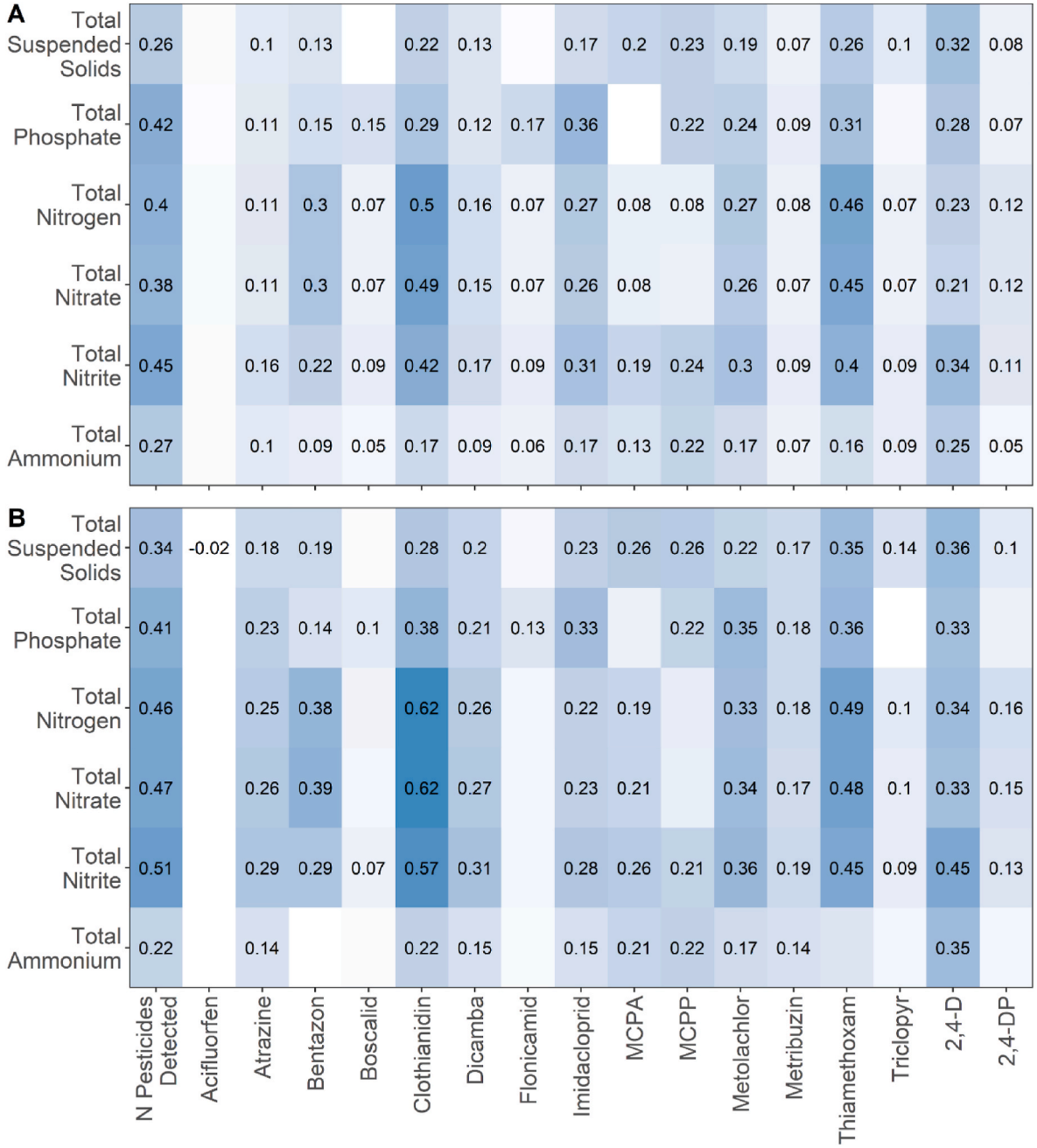


Fig. 3. Non-parametric correlation coefficient (Kendall's tau) for concentrations of pesticides and number of pesticides detected versus other measured water quality variables across all 18 study sites from 2015 to 2019. Panel A (top) shows correlations for samples collected year-round; panel B (bottom) shows correlations for only samples collected during the months of May and June. Colour indicates strength of correlation, with blue indicating a strong correlation and white indicating a weak correlation. Tau coefficients are only listed for significant ($p < 0.05$) relationships. N = 353–392 for all correlation tests. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

concentrations and other water quality variables for samples collected year-round, however most correlations were weak to moderate in strength ($p < 0.05$, Kendall's tau 0.05–0.5 (Fig. 3A, Supplemental Data Table S26). Most correlations improved in strength when only samples collected during crop planting season (May, June) were considered (Fig. 3B, Supplemental Data Table S26). Pesticides used during the early stages of crop growth such as the herbicides atrazine, MCPA, 2,4-D, neonicotinoid insecticides (NNIs) applied as seed treatments (clothianidin, thiamethoxam), and the total number of pesticides detected in a sample showed the most improvement in correlation strength when samples from May and June were isolated. The number of pesticides detected in a sample often showed a stronger correlation with nutrients and suspended solids than individual pesticide concentrations did, except for the NNIs clothianidin and thiamethoxam and the herbicide 2,4-D (Fig. 3). Concentrations of the NNIs clothianidin and thiamethoxam showed the strongest correlations (Kendall's tau 0.17–0.5, $p < 0.01$ for year-round samples) with concentrations of nutrients and suspended solids, indicating that NNIs, nutrients, and suspended solids could share a transport pathway from land to streams, as particular land uses/activities in the upstream catchment could promote the release of NNIs, nutrients, and suspended solids. A number of studies have observed that in catchments with a high density of agricultural land use, elevated concentration and number of pesticides detected in streams often corresponds with elevated nutrients (Mitchell et al., 2005; Falcone et al., 2018; Solis et al., 2019; Marrochi et al., 2020). While it is easy to focus on pesticide exposure, the correlation between NNIs, nutrients, and suspended solids highlights the importance of considering that aquatic ecosystems can be exposed to multiple factors that may affect their health due to activities upstream (Barmentlo et al., 2018; Cavallaro et al., 2019; Chara-Serna et al., 2019). For example, Barmentlo et al. (2018) observed that the effect of the NNI thiacloprid on freshwater invertebrates was reduced under conditions with elevated nutrients, which they hypothesized was due to increased primary production providing increased forage for invertebrates.

3.3. Screening-level hazard assessment to aquatic life

The Pesticide Toxicity Index (PTI) was used to assess the potential hazard of each sample to aquatic life at the point-of-collection, and the distribution of PTI scores was used to assess overall hazard to aquatic life in southern Ontario agricultural streams. Since data below the MDL and the MQLs was treated as zero in the PTI calculation, elevated MDL and MQLs for samples collected and analyzed prior to 2014/2015 likely artificially lowered PTI scores and PTI scores therefore represent minimum values. Comparisons of the distribution of PTI scores before and after 2015 showed that at many sites, scores were significantly different (Wilcoxon test, $p < 0.05$) for samples collected between 2012–2014 and 2015–2019. (Supplemental Data Figures S2–S6). Consequently, we focus here on PTI results from years 2015–2019 as they capture a more recent and comprehensive outlook of hazard to aquatic life.

Hazard to aquatic life was assessed based on the distribution of PTI scores for each of five taxa: benthic invertebrates, cladocerans, fish, algae, and macrophytes. Benthic invertebrates had the highest overall PTI scores across all sites (median 0.0024). Scores for algae and macrophytes were an order of magnitude lower, and scores for cladocerans and fish were very low ($<10^{-5}$) (Table 2). In general, PTI scores were under thresholds where acute or chronic toxicity would be expected. However, at some sites PTI scores for some samples did exceed chronic, and in very few cases, acute thresholds for toxicity, indicating hazard to aquatic communities may be present at select sites.

Very few samples exceeded the threshold for acute toxicity with a PTI score >1.0 (8/601, 1.3%). The majority (7/8) were also collected at Lebo Drain, where concentrations of chlorantraniliprole and imidacloprid were the drivers of potential toxicity. One sample from Reynolds Creek had highly elevated levels (77 μg/L) of the herbicide metolachlor, which was sufficiently high to potentially cause an acutely toxic response to macrophytes and algae; however, this was the highest concentration of any pesticide observed across all sites and years and was more than double the next highest concentration of metolachlor (29 μg/L).

Table 2
Summary of Pesticide Toxicity Index (PTI) scores for 601 samples collected from 18 sites from 2015 to 2019.

Taxa	PTI Score			Percent exceeding hazard threshold	
	Mean	Median	Range	Chronic (PTI >0.1)	Acute (PTI >1.0)
Algae	0.0203	0.0006	0–1.669	4.0	0.2
Benthic Invertebrates	0.0900	0.0024	0–13.38	7.8	1.1
Cladocerans	0.0125	0.00001	0–1.151	2.8	0.5
Fish	0.0009	0.00001	0–0.0869	0	0
Macrophytes	0.0193	0.0001	0–1.749	4.0	0.1

A total of 63/601 (10.5%) of samples exceeded the threshold for chronic toxicity with a PTI score >0.1 for at least one taxon. The majority (29/63) of these samples were concentrated at one site, Lebo Drain, where elevated concentrations of the insecticides chlorantraniliprole and imidacloprid were sufficiently high to cause toxicity to cladocerans and benthic invertebrates, respectively (Fig. 4, Supplemental Data Tables S27–S28 and Figure S12). The remaining samples that showed potential chronic toxicity were spread throughout 12 other sites, and in all cases comprised less than 20% of the overall samples from a given site. In addition, while at Lebo Drain the taxa expected to be most at risk were cladocerans and benthic invertebrates, at most other sites there was an equal or greater hazard to primary producers (algae, macrophytes) (Fig. 4).

The pesticide with the greatest contribution to the overall PTI score of an individual sample was identified and the contribution of that pesticide was termed the TU_{max}. The neonicotinoid insecticide imidacloprid was found to be the dominant driver behind toxicity to benthic invertebrates (Fig. 5). Imidacloprid and other NNIs have repeatedly been shown to be toxic to benthic invertebrates in laboratory and mesocosm-based studies at environmentally-relevant concentrations (e.g., Raby et al., 2018; Cavallaro et al., 2018) and have been implicated in the decline of the aquatic insect community in exposed water bodies (Morrissey et al., 2015; Hladik et al., 2018). The toxic concentration values for benthic invertebrates in the Pesticide Toxicity Index database for clothianidin and thiamethoxam of 59 μg/L and 967 μg/L, respectively (Nowell et al., 2014), do not consider more recent toxicological studies that would lower these toxic benchmark values. Draft water quality guidelines by the Canadian Council of Ministers of the Environment (CCME) and Health Canada's Pest Management Regulatory Agency make use of similar methodology as the PTI (namely the species sensitivity distribution (SSD) approach) and have derived values several orders of magnitude lower than those in the PTI. Other benchmark values such as the United States Environmental Protection Agency's Office of Pesticide Products Aquatic Life Benchmarks apply a 'level of concern' to an applicable fish or invertebrate acute toxicity value, some of which are lower than those in the PTI (US EPA, 2020). If such values were used in place of existing PTI values, calculated PTI scores would be considerably higher, and a larger proportion of samples collected in this study would have scored as potentially toxic. For example, applying the United States Environmental Protection Agency's Office of Pesticide Products Aquatic Life Benchmarks to this same dataset showed 22% of samples exceeded the chronic benchmark of 0.01 μg/L imidacloprid, and 25% of samples exceeded the chronic benchmark of 0.05 μg/L clothianidin (US EPA, 2020). Few samples exceeded US EPA Aquatic Life Benchmarks for other NNIs (e.g., thiamethoxam, acetamiprid, thiacloprid). The use of a desktop hazard assessment such as the PTI may not however accurately capture toxic effects, as demonstrated by Schepker et al. (2020) who found a negative associated between NNI

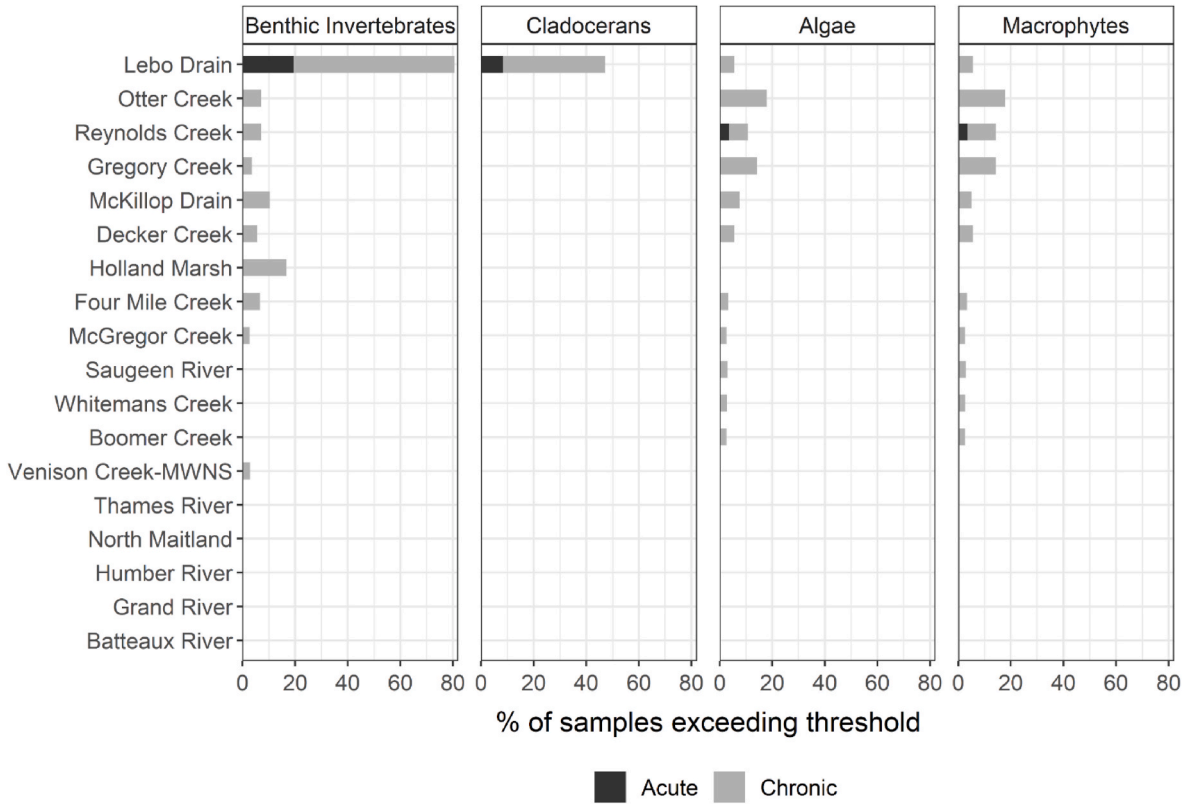


Fig. 4. Percentage of 601 stream water samples collected from 2015 to 2019 across 18 agricultural stream sites in southern Ontario that exceeded Pesticide Toxicity Index (PTI) thresholds of 0.1 for chronic toxicity and 1.0 for acute toxicity.

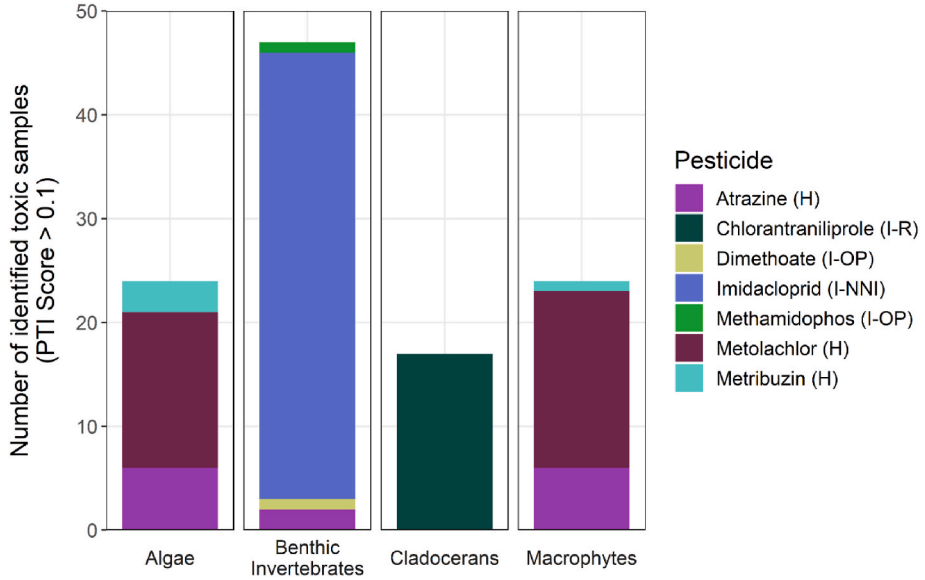


Fig. 5. Distribution of pesticides with the greatest contribution towards PTI scores (termed maximum toxic unit, or TU_{max}) in samples with Pesticide Toxicity Index score (PTI score) > 0.1. All fish-PTI scores were <0.1. Data restricted to samples collected between 2015 and 2019. Pesticide class is listed in the legend after the pesticide: H = herbicide; I = insecticide; R = ryanoid class; OP = organophosphate class; NNI = neonicotinoid class.

concentrations and aquatic invertebrate biomass in wetland study areas, despite NNI concentrations falling below US EPA Aquatic Life Benchmarks. Of the pesticides monitored in water, elevated concentrations of NNIs pose the greatest hazard to aquatic invertebrate communities in Ontario's streams exposed to pesticides. This conclusion echoes several recently published studies in stream (Liess et al., 2021; Stackpoole et al., 2021) and wetland (Schepker et al., 2020) habitats.

The ryanoid insecticide chlorantraniliprole was the major driver behind toxicity to cladocerans. Recent toxicological studies have shown toxicity to *Chironomus dilutus* (96-h LC50 = 4.0 μg/L, Maloney et al. (2020)) and *Neocloeon triangulifer* (96-h LC50 = 2.9 μg/L, Sanford et al. (2021) at levels that are within the range of those detected in the present study at Lebo Drain (median measured concentration of chlorantraniliprole 0.545 μg/L, maximum 8 μg/L). Given that the use of

chlorantraniliprole and other diamide/ryanoid insecticides may increase in future years as a response to restrictions on neonicotinoid insecticides, future toxicological studies and environmental monitoring should include this class of pesticides.

Pesticide concentrations and PTI scores showed a seasonal pattern with the highest concentrations and PTI scores occurring in the month of June (Supplemental Data, Figures S7-S11). This pattern coincides with the agricultural growing season in southern Ontario and the average timing of herbicide and insecticide application (e.g., herbicide application made prior to and after crop emergence, or NNI application via planting treated seed). This 'spring flush' phenomenon has been repeatedly observed in Ontario and North America with herbicides (Thurman et al., 1991; Nowell et al., 2018); and neonicotinoid insecticides (Struger et al., 2017). Very little sampling occurred in this study during the winter months (December, January, February, March) with only 23 winter samples collected in 12 sites. However, herbicide concentrations (MCPP, MCPA, 2,4-DP and triclopyr) in the samples that were collected showed levels similar to those in samples collected during the growing season suggesting some level of pesticide transport during the winter months. Too few samples were collected during winter months to draw conclusions on pesticide concentrations in the winter months and additional sampling is needed at all sites in the winter months to determine seasonal patterns in pesticide concentrations over the entire year.

Several pesticides that were frequently detected (in >10% of samples) did not have PTI toxic concentrations for benthic invertebrates, cladocerans, or fish. The absence of PTI toxic concentrations for pesticides that are frequently detected in samples would result in underestimation of the PTI score across sites, and thus, hazard would be underestimated. The pesticides for which this applied (MCPP, MCPA, 2,4-DP and triclopyr) were detected in most sites across the province meaning any underestimation of hazard was not concentrated to a specific site. It is also important to note that the four frequently detected pesticides missing PTI toxic concentrations were all herbicides. Consequently, their toxicity to vertebrates and invertebrates is relatively low compared to insecticides and fungicides (Fargašová, 1994; Belgers et al., 2007; Fairchild et al., 2009). This means that the underestimation of hazard to aquatic vertebrates and invertebrates using the PTI approach in this study is very low. However, this highlights a limitation in the PTI approach to assess the hazard of pesticide mixtures. The approach does not include a PTI score for aquatic primary producers (e.g., algae, macrophytes), which means the PTI score (without the amendments added in the present study) does not speak to the hazard of pesticide mixtures to primary producers. This is a concern as the majority of pesticides (12/17) detected in >10% of samples in this study were herbicides (Table 1). The addition of toxic concentrations for algae and macrophytes showed some evidence of chronic hazard (<20% of samples exceeded the chronic threshold, Fig. 4) to primary producers at most sites that would have otherwise been overlooked. The use of primary producer acute toxicity values, however, could overestimate toxicity as herbicides temporarily suppress the growth of non-target algae and macrophytes, but populations tend to recover once exposure is reduced (Fairchild, 2011). The use of single species toxicity tests generally do not reflect the complexity of natural systems and their capacity for recovery (Fairchild, 2011). The result of some chronic hazard to primary producers found in this study may therefore be considered very conservative. Other limitations of the PTI approach include assuming that pesticide toxicity is only additive (i.e., no synergism or antagonism is taking place) and the inherent uncertainty in using laboratory toxicity tests with standard test species to extrapolate to real-world potential toxicity (Nowell et al., 2014).

The results of this study were influenced by the sampling design. Samples were collected on an approximately monthly basis and for most sites, sample collection during wet-weather events were not targeted. This sampling design was chosen to balance resources with obtaining ambient pesticide concentrations. Work by Norman et al. (2020) examining the influence of sampling frequency on pesticide concentrations showed weekly discrete grab samples were likely to miss peak concentrations resulting in acute exposure being underestimated. Given the sampling design used in the present study, this was likely the case and acute (short-term) exposure was underestimated. In addition, the results of this study were influenced by improving analysis methods, so the number of analytes measured improved over time, and the concentrations at which those analytes could be measured decreased. More pesticides were therefore detected and at lower concentrations in the later years (2015–2019) than earlier (2012–2014). This underscores the importance of placing pesticide monitoring results in the context of analysis methods. These limitations are important to note against the backdrop of the usefulness of the PTI as an indicator of relative pesticide toxicity and water quality.

4. Conclusions

In Ontario's streams, herbicides and neonicotinoid insecticides are the most frequently detected pesticides in water samples. Dissolved pesticide concentrations measured in stream water were related to upstream land use, with the majority of pesticides detected commonly used in agriculture and some (MCPP, triclopyr, 2,4-DP) used in non-agricultural vegetation management. Based on the data collected as part of this monitoring program, the hazard to aquatic life based on the PTI approach is relatively low across southern Ontario. The exception was one monitoring site (the Lebo Drain) where concentrations of insecticides chlorantraniliprole and imidacloprid were sufficiently high to present a potential hazard to aquatic invertebrates. In addition, the number of pesticides detected in a streamwater sample was positively correlated with nutrients and suspended solids which highlights the importance of considering multiple factors/stressors to aquatic health.

Credit author statement

Melanie Raby: Conceptualization, Methodology, Formal analysis, Investigation, Data curation, Project administration, Visualization, Writing – original draft, Writing – review & editing. **Linda Lissemore**: Methodology, Formal analysis, Writing – review & editing. **Georgina Kaltenecker**: Conceptualization, Data curation, Project administration, Writing – review & editing. **Denise Beaton**: Conceptualization, Project administration, Writing – review & editing. **Ryan Prosser**: Conceptualization, Formal analysis, Investigation, Writing – original draft, Writing – review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

The authors are grateful to Ontario's Conservation Authorities for sample collection and the Laboratory Services Branch of the Ontario Ministry of the Environment, Conservation and Parks for sample analysis of nutrient and solids parameters. Grace Arabian made the map of monitoring sites and watersheds and the graphical abstract. We also thank Jennifer Winter and Mike Cowbrough and three anonymous reviewers for comments that improved this manuscript.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.chemosphere.2022.133769.

References

AAFC, 2020. Annual Crop Inventory. Agriculture and Agri-Food Canada. Canada, Ottawa, ON. https://open.canada.ca/data/en/dataset/ba2645d5-4458-414d-b196-6303ac06c1c9.

Arle, J., Mohaupt, V., Kirst, I., 2016. Monitoring of surface waters in Germany under the Water Framework Directive - a review of approaches, methods and results. Water 8, 217.

Barmentlo, S.H., Parmentier, E.M., De Snoo, G.R., Vijver, M.G., 2018. Thiacloprid-induced toxicity influenced by nutrients: evidence from in situ bioassays in experimental ditches. Environ. Toxicol. Chem. 37, 1907–1915.

Belgers, J.D.M., Van Lieverloo, R.J., Van Der Pas, L.J.T., Van Den Brink, P.J., 2007. Effects of the Herbicide 2,4-D on the Growth of Nine Aquatic Macrophytes, vol. 86, pp. 260–268.

Black, R., Haggland, A., Voss, F., 2000. Predicting the probability of detection organochlorine pesticides and polychlorinated biphenyls in stream systems on the basis of land use in the Pacific Northwest, USA. Environ. Toxicol. Chem. 19, 1044–1054.

Bocard, D., Gillet, F., Legendre, P., 2018. Numerical Ecology with R. Springer International Publishing.

Bolks, A., DeWire, A., Harcum, J.B., 2014. Baseline assessment of left-censored environmental data using R. Tech Notes 10. June 2014. Developed for U.S. Environmental Protection Agency by Tetra Tech, Inc., Fairfax, VA, p. 28. Available online at: https://www.epa.gov/sites/default/files/2016-05/documents/tech_notes_10_jun2014_r.pdf.

Boye, K., Lindström, B., Kreuger, J., 2019. Long-term data from the Swedish national environmental monitoring program of pesticides in surface waters. J. Environ. Qual. 48, 1109–1119.

Brown, C., van Beinum, W., 2009. Pesticide transport via sub-surface drains in Europe. Environ. Pollut. 157, 3314–3324.

Bunzel, K., Liess, M., Kattwinkel, M., 2014. Landscape parameters driving aquatic pesticide exposure and effects. Environ. Pollut. 186, 90–97.

Cavallaro, M.C., Liber, K., Headley, J.V., Peru, K.M., Morrissey, C.A., 2018. Community-level and phenological responses of emerging aquatic insects exposed to 3 neonicotinoid insecticides: an in situ wetland limnocorral approach. Environ. Toxicol. Chem. 37 (9), 2401–2412.

Cavallaro, M.C., Main, A.R., Liber, K., Phillips, I.D., Headley, J.V., Peru, K.M., Morrissey, C.A., 2019. Neonicotinoids and other agricultural stressors collectively modify aquatic insect communities. Chemosphere 226, 945–955.

Chara-Serna, A.M., Epele, L.B., Morrissey, C.A., Richardson, J.S., 2019. Nutrients and sediment modify the impacts of a neonicotinoid insecticide on freshwater community structure and ecosystem functioning. Sci. Total Environ. 692, 1291–1303.

Chow, R., Scheidegger, R., Doppler, T., Dietzel, A., Fenicia, F., Stamm, C., 2020. A review of long-term pesticide monitoring studies to assess surface water quality trends. Water Res. X 9, 100064.

Di Guardo, A., Finizio, A., 2018. A new methodology to identify surface water bodies at risk by using pesticide monitoring data: the glyphosate case study in Lombardy Region (Italy). Sci. Total Environ. 610–611, 421–429.

Douglas, M.R., Tooker, J.F., 2015. Large-scale deployment of seed treatments has driven rapid increase in use of neonicointoid insecticides and preemptive pest management in U.S. field crops. Environ. Sci. Technol. 49, 5088–5097.

ECCC, 2020. Great Lakes Water Quality Monitoring and Surveillance Data. Government of Canada, Ottawa, ON, Canada. https://open.canada.ca/data/dataset/cfdafa0c-a644-47cc-ad54-460304facf2e?=undefined&wbdisable=true.

Fairchild, J.F., Feltz, K.P., Allert, A.L., Sappington, L.C., Nelson, K.J., Valle, J.A., 2009. An Ecological Risk Assessment of the Exposure and Effects of 2,4-D Acid to Rainbow Trout (*Onchorhyncus mykiss*), vol. 56, pp. 754–760.

Fairchild, J.F., 2011. Structural and Functional Effects of Herbicides on Non-target Organisms in Aquatic Ecosystems with an Emphasis on Atrazine. Herbicides and the Environment, pp. 383–404.

Falcone, J.A., Murphy, J.C., Sprague, L.A., 2018. Regional patterns of anthropogenic influences on streams and rivers in the conterminous United States, from the early 1970s to 2012. J. Land Use Sci. 13, 585–614.

Fargašová, A., 1994. Comparative Study of Plant Growth Hormone (Herbicide) Toxicity in Various Biological Subjects, vol. 29, pp. 359–364.

Farm & Food Care Ontario, 2015. Survey of Pesticide Sse in Ontario, 2013/2014 - Estimates of Pesticides Used on Field Crops and Fruit and Vegetable Crops. Farm & Food Care Ontario, Guelph, ON, Canada.

Finizio, A., Azimonti, G., Villa, S., 2011. Occurrence of pesticides in surface water bodies: a critical analysis of the Italian national pesticide survey. J. Environ. Monit. 13, 49–57.

Fortin, J., Gagnon-Bertrand, E., Vézina, L., Rompré, M., 2002. Preferential Bromide and Pesticide Movement to Tile Drains under Different Cropping Practices, vol. 31, p. 1940.

Glozier, N., Struger, J., Cessna, A., Gledhill, M., Rondeau, M., Ernst, W., Sekela, M., Cagampan, S., Sverko, E., Murphy, C., Murray, J., Donald, D., 2012. Occurrence of glyphosate and acidic herbicides in select urban rivers and streams in Canada, 2007. Environ. Sci. Pollut. Control Ser. 19, 821–834.

Gustavsson, M., Kreuger, J., Bundschuh, M., Backhaus, T., 2017. Pesticide mixtures in the Swedish streams: environmental risks, contributions of individual compounds and consequences of single-substance oriented risk mitigation. Sci. Total Environ. 598, 973–983.

Headley, J., 1968. Estimating the productivity of agricultural pesticides. Am. J. Agric. Econ. 50, 13–23.

Hladik, M., Main, A., Goulson, D., 2018. Environmental risks and challenges associated with neonicotinoid insecticides. Environ. Sci. Technol. 52, 3329–3335.

Hunt, J.W., Anderson, B.S., Phillips, B.M., Tjeerdema, R.S., Richard, N., Connor, V., Worcester, K., Angelo, M., Bern, A., Fulfrost, B., Mulvaney, D., 2006. Spatial relationships between water quality and pesticide application rates in agricultural watersheds. Environ. Monit. Assess. 121, 245–262.

Kassambara, A., 2020. Ggpubr: 'ggplot 2' Based Publication Ready Plots, R Package version 0.2.5. https://CRAN.R-project.org/package=ggpubr.

Kreuger, J., 1998. Pesticides in stream water within an agricultural catchment in southern Sweden, 1990-1996. Sci. Total Environ. 216, 227–251.

Lee, L., 2017. NADA: nondetects and data analysis for environmental data. Package Ver. 1 (1).

Liess, M., Liebmann, L., Vormeier, P., Weisner, O., Altenburger, R., Borchardt, D., Reemtsma, T., 2021. Pesticides are the dominant stressors for vulnerable insects in lowland streams. Water Res. 201, 117262.

Maloney, E., Sykes, H., Morrissey, C., Peru, K., Headley, J., Liber, K., 2020. Comparing the acute toxicity of imidacloprid with alternative systemic insecticides in the aquatic insect *Chironomus dilutus*. Environ. Toxicol. Chem. 39, 587–594.

Mamy, L., Barriuso, E., 2005. Glyphosate adsorption in soils compared to herbicides replaced with the introduction of glyphosate resistant crops. Chemosphere 61, 844–855.

Maqueda, C., Undabeytia, T., Villaverde, J., Morillo, E., 2017. Behaviour of glyphosate in a resevoir and the surrounding agricultural soils. Sci. Total Environ. 593–594, 787–795.

Marrochi, M.N., Hunt, L., Solis, M., Scalise, A.M., Fanelli, S.L., Bonetto, C., Mugni, H., 2020. Land-use impacts on benthic macroinvertebrate assemblages in pampean streams (Argentina). J. Environ. Manag. 10, 111608, 1016/j.jenvman.2020.111608.

Mitchell, C., Brodie, J., White, I., 2005. Sediments, nutrients and pesticide residues in event flow conditions in streams of the Mackay Whitsunday Region, Australia. Mar. Pollut. Bull. 51, 23–36.

Montoya, J.C., Costa, J.L., Liedl, R., Bedmar, F., Daniel, P., 2006. Effects of Soil Type and Tillage Practice on Atrazine Transport through Intact Soil Cores, vol. 137, pp. 161–173.

Morrissey, C., Mineau, P., Devries, J., Sanchez-Bayo, F., Liess, M., Cavallaro, M., Liber, K., 2015. Neonicotinoid contamination of global surface waters and associated risk to aquatic invertebrates: a review. Environ. Int. 74, 291–303.

Nicholls, P., 1988. Factors influencing entry of pesticides into soil water. Pest Manag. Sci. 22, 123–137.

Norman, J.E., Mahler, B.J., Nowell, L.H., Van Metre, P.C., Sandstrom, M.W., Corbin, M. A., et al., 2020. Daily stream samples reveal highly complex pesticide occurrence and potential toxicity to aquatic life. Sci. Total Environ. 715, 136795.

Novak, S.M., Portal, J.M., Schiavon, M., 2001. Effects of soil type upon metolachlor losses in subsurface drainage. Chemosphere 42, 235–244.

Nowell, L.H., Norman, J.E., Moran, P.W., Martin, J.D., Stone, W.W., 2014. Pesticide toxicity index—a tool for assessing potential toxicity of pesticide mixtures to freshwater aquatic organisms. Sci. Total Environ. 476, 144–157.

Nowell, L.H., Moran, P.W., Schmidt, T.S., Norman, J.E., Nakagaki, N., Shoda, M.E., Hladik, M.L., 2018. Complex mixtures of dissolved pesticides show potential aquatic toxicity in a synoptic study of Midwestern US streams. Sci. Total Environ. 613, 1469–1488.

Okada, E., Costa, J., Bedmar, F., 2016. Adsorption and mobility of glyphosate in different soils under no-till and conventional tillage. Geoderma 263, 78–85.

Oksanen, J., Blanchet, F., Friendly, M., Kindt, R., Legendre, P., McGlinn, D., Minchin, P., O'Hara, R., Simpson, G., Solymos, P., Stevens, M., Szoecs, E., Wagner, H., 2019. Vegan : community ecology package, R package. version 2.5-6. https://CRAN.R-project.org/package=vegan.

Ontario Ministry of the Environment, Conservation and Parks, 2018. Corn and Soybean Neonicotinoid-Treated Seed Data [data File]. Retrieved from: https://data.ontario.ca/dataset/corn-and-soybean-neonicotinoid-treated-seed-data.

PMRA, 2020. Pesticide Product Label Search. Health Canada, Ottawa, ON, Canada. https://pr-rp.hc-sc.gc.ca/ls-re/index-eng.php.

Prosser, R.S., Hoekstra, P.F., Gene, S., Truman, C., White, M., Hanson, M.L., 2020. A review of the effectiveness of vegetated buffers to mitigate pesticide and nutrient transport into surface waters from agricultural areas. J. Environ. Manag. 261, 110210.

Raby, M., Zhao, X., Hao, C., Poirier, D.G., Sibley, P.K., 2018. Chronic toxicity of 6 neonicotinoid insecticides to Chironomus dilutus and Neocloeon triangulifer. Environ. Toxicol. Chem. 37 (10), 2727–2739.

Sanford, M., Prosser, R.S., 2020. High-frequency sampling of small streams in the agroecosystems of southwestern Ontario, Canada, to characterize pesticide exposure and associated risk to aquatic life. Environ. Toxicol. Chem. 39 (12), 2570–2587.

Sanford, M., Washuck, N., Carr, K., Prosser, R., 2021. Pulsed exposure of the macrophyte Lemna minor to herbicides and the mayfly *Neocloeon triangulifer* to diamide insecticides. Chemosphere 273, 128582.

Schepker, T.J., Webb, E.B., Tillitt, D., LaGrange, T., 2020. Neonicotinoid insecticide concentrations in agricultural wetlands and associations with aquatic invertebrate communities. Agric. Ecosyst. Environ. 287, 106678.

Schmidt, T.S., Van Metre, P.C., Carlisle, D.M., 2019. Linking the agricultural landscape of the midwest to stream health with structural equation modeling. Environ. Sci. Technol. 53, 452–462.

Schreiner, V., Szöcs, E., Bhowmik, A., Vijver, M., Schäfer, R., 2016. Pesticide mixtures in streams of several European countries and the USA. Sci. Total Environ. 573, 680–689.

Schriever, C.A., Ball, M.H., Holmes, C., Maund, S., Liess, M., 2007. Agricultural intensity and landscape structure: influences on the macroinvertebrate assemblages of small streams in northern Germany. Environ. Toxicol. Chem. 26, 346.

Schulz, R., Peall, S., Dabrowski, J., Reinecke, A., 2001. Spray deposition of two insecticides into surface waters in a South African orchard area. J. Environ. Qual. 30, 814–822.
Sexton, S., Lei, Z., Zilberman, D., 2007. The economics of pesticides and pest control. Int. Rev. Environ. Resour. Econ. 1.
Solis, M., Arias, M., Fanelli, S., Bonetto, C., Mugni, H., 2019. Agrochemicals' effects on functional feeding groups of macroinvertebrates in Pampas streams. Ecol. Indicat. 101, 373–379.
Solomon, K., Baker, D., Richards, R., Dixon, K., Klaine, S., La Point, T., Kendall, R., Weisskopf, C., Giddings, J., Giesy, J., Hall, L., Williams, W., 1996. Ecological risk assessment of atrazine in North American surface waters. Environ. Toxicol. Chem. 15, 31–76.
Solomon, K., Giesy, J., Jones, P., 2000. Probabilistic risk assessment of agrochemicals in the environment. Crop Protect. 19, 649–655.
Spycher, S., Mangold, S., Doppler, T., Junghans, M., Wittmer, J., Stamm, C., Singer, H., 2018. Pesticide risks in small streams - how to get as close as possible to the stress imposed on aquatic organisms. Environ. Sci. Technol. 52, 4526–4535.
Stackpoole, S.M., Shoda, M.E., Medalie, L., Stone, W.W., 2021. Pesticides in US rivers: regional differences in use, occurrence, and environmental toxicity, 2013 to 2017. Sci. Total Environ. 787, 147147.
Struger, J., Grabuski, J., Cagampan, S., Sverko, E., McGoldrick, D., Marvin, C.H., 2017. Factors influencing the occurrence and distribution of neonicotinoid insecticides in surface waters of southern Ontario, Canada. Chemosphere 169, 516–523.
Szöcs, E., Brinke, M., Karaoglan, B., Schäfer, R., 2017. Large scale risks from agricultural pesticides in small streams. Environ. Sci. Technol. 51, 7378–7385.
Thurman, E.M., Goolsby, D.A., Meyer, M.T., Kolpin, D.W., 1991. Herbicides in surface waters of the Midwestern United States: the effect of spring flush. Environ. Sci. Technol. 25, 1794–1796.
Tran, C.P., Bode, R.W., Smith, A.J., Kleppel, G.S., 2010. Land-use proximity as a basis for assessing stream water quality in. New York State (USA) 10, 727–733.
Traub-Eberhard, U., Kördel, W., Klein, W., 1994. Pesticide movement into subsurface drains on a loamy silt soil. Chemosphere 28, 273–284.
USEPA, 2020. Aquatic Life Benchmarks and Ecological Risk Assessments for Registered Pesticides. USEPA. https://www.epa.gov/pesticide-science-and-assessing-pesticide-risks/aquatic-life-benchmarks-and-ecological-risk.
USGS, 2020. Pesticides and Water Quality. United States Geological Survey, Washington, DC, USA. https://www.usgs.gov/mission-areas/water-resources/science/pesticides-and-water-quality?qt-science_center_objects=0#qt-science_center_objects.
Wettstein, F.E., Kasteel, R., Garcia Delgado, M.F., Hanke, I., Huntscha, S., Balmer, M.E., Poiger, T., Bucheli, T.D., 2016. Leaching of the neonicotinoids thiamethoxam and imidacloprid from sugar beet seed dressings to subsurface tile drains. J. Agric. Food Chem. 64, 6407–6415.
Williams, R., Brooke, D., Matthiessen, P., Mills, M., Turnbull, A., Harrison, R., 1995. Pesticide transport to surface waters within an agricultural catchment. Water Environ. 9, 72–81.
Williams, W., Giddings, J., Purdy, J., Solomon, K., Giesy, J., 2014. Exposure of aquatic organisms to the organophosphorus insecticide, chlorpyrifos resulting from use in the United States. In: Giesy, J., Solomon, K. (Eds.), Reviews of Environmental Contamination and Toxicology - Ecological Risk Assessment of Chlorpyrifos in Terrestrial and Aquatic Systems in the United States. Springer.