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Wetland buffers are no substitute for landscape-scale conservation

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Abstract. Wetlands in farmland are at risk of contamination by fertilizers and pesticides. One recommendation for reducing wetland contamination is to maintain a buffer of contiguous uncropped land around the wetland (a wetland buffer). Many agricultural water protection policies around the world recommend 5 to 50-m wide uncropped buffers around water bodies, but it is unclear how large wetland buffers must be to effectively protect against these chemicals. In addition, it is unclear whether wetland buffers have similar—or stronger—effects on fertilizer and pesticide contamination than reducing the amount of cropped land within the larger landscape context around wetlands. Our study, conducted across 37 wetlands in eastern Ontario, Canada, addressed the following questions: (1) Does increasing buffer width, or increasing the amount of contiguous uncropped land within recommended buffer width guidelines, reduce nutrient and pesticide levels in agricultural wetlands? (2) Does increasing uncropped land cover in the broader landscape reduce nutrient and pesticide levels in agricultural wetlands? and (3) What is the relative importance of buffer size and landscape-scale uncropped cover for reducing nutrient and pesticide levels in agricultural wetlands? A rigorous site selection process was employed to minimize the correlation between buffer size and landscapescale uncropped cover, minimize spatial gradients in these predictor variables, and minimize variation in potentially confounding variables. We obtained nutrient and pesticide data by collecting water samples from each wetland under similar weather conditions in June-July 2015. Nitrate concentrations were measured using ion chromatography, and atrazine and neonicotinoid (pesticide) concentrations using a combination of high-performance liquid chromatography and mass spectrometry. We found that nitrate, atrazine, and neonicotinoid concentrations in study wetlands were unaffected by wetland buffer size. However, concentrations of each chemical decreased with uncropped land cover in the surrounding 150 to 300-m radius landscapes. To effectively protect water in agricultural wetlands from contamination by nitrate-based fertilizers and atrazine or neonicotinoid pesticides, we recommend either increasing the policy-recommended width of wetland buffers to at least 150 m, or abandoning the buffer paradigm in favor of landscape-scale conservation.

Key words: agricultural wetland; atrazine; buffer zone; land use; neonicotinoids; nitrogen; pesticide; riparian buffer; scale of effect; spatial extent; vegetated filter strip; water quality.

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Introduction

Increasing agricultural land use and farming intensity can increase nutrient loads and

pesticide contamination in nearby water bodies, with negative consequences for ecosystem health (Carpenter et al. 1998, EC-WSTD 2011, Schäfer et al. 2011). Nutrients such as nitrogen and

phosphorus occur naturally in aquatic systems, but artificially high amounts can alter community composition via eutrophication (Carpenter et al. 1998) and can have direct lethal or sublethal effects on aquatic species (Hecnar 1995, Camargo et al. 2005). Agricultural pesticides such as atrazine and neonicotinoids can also kill or harm aquatic organisms, and reduce biodiversity (Graymore et al. 2001, EC-WSTD 2011, Schäfer et al. 2011, Beketov et al. 2013, Morrissey et al. 2015).

The most direct solution is to limit the spread and intensification of agriculture, and conserve uncropped land in landscapes surrounding water bodies of interest, such as wetlands. Crosbie and Chow-Fraser (1999) found that nutrient levels in marshes increased with the percentage of farmland in the watershed and decreased with the amount of woodland in the watershed. Similarly, Houlahan and Findlay (2004) showed that nitrogen and phosphorus concentrations in wetlands declined as woodland cover in the surrounding landscape increased. Smaller-scale studies suggest that pesticide levels in water bodies also increase with the amount of nearby agriculture (Rasmussen et al. 2011, McMurry et al. 2016).

However, conserving uncropped land at a landscape scale around wetlands in agricultural regions can be challenging. For instance, the appropriate landscape scale is unclear: Different studies have found surrounding land use from a 100-m radius up to the entire watershed to significantly affect water quality in lentic wetlands (Crosbie and Chow-Fraser 1999, Houlahan and Findlay 2004, Declerck et al. 2006). Furthermore, if the appropriate landscape scale exceeds the size of one person's property, landscape-scale conservation would require coordinating the efforts of multiple landowners.

Another way to reduce agricultural wetland contamination is to implement buffers, that is, strips of uncropped land adjacent to wetlands (Muscutt et al. 1993, Castelle et al. 1994, Coukell et al. 2004). Buffers have been recommended around water bodies by local, regional, and national governments in many parts of the world. In Canada, buffers 5–120 m wide are frequently recommended for aquatic habitat protection, with most recommendations falling between 5 and 50 m (Huel 2000, Coukell et al. 2004, PEI Legislative Counsel Office 2012,

Niagara Planning and Development Services Department 2014); in the USA, buffer width recommendations range from 4.6 to 106.7 m (McElfish et al. 2008). Similar guidelines exist in other parts of the world, such as Europe, South America, and Australia (State of Western Australia 2005, European Court of Auditors 2014, Machado and Anderson 2016). Buffers along rivers and streams have been shown to intercept runoff, reduce nutrient loads, and reduce pesticide contamination (Castelle et al. 1994, Vought et al. 1995, Wenger 1999).

However, it is unclear what size of buffer would effectively protect water quality in lentic agricultural wetlands. This is partly because most field studies of buffer effectiveness have focused on riparian buffers along streams and rivers rather than on buffers around lentic wetlands in regions with flat topography, as in many agricultural regions. Furthermore, studies focusing on riparian buffers along streams and rivers have come to widely different conclusions, with different studies recommending buffer widths anywhere between 5 and 90 m (Castelle et al. 1994, Vought et al. 1995, Wenger 1999, Rasmussen et al. 2011). In addition, buffer size is difficult to measure. Official buffer guidelines usually focus on buffer width, measured in an agricultural context as the minimum distance from a water body to cropped land (OFEC et al. 2004, McElfish et al. 2008, European Court of Auditors 2014), but real buffers vary in width. The most ecologically relevant way to measure buffer size has not been empirically determined and is rarely discussed in buffer guideline documents.

The effectiveness of wetland buffers relative to landscape-scale conservation of uncropped land around wetlands is also unclear. Most multi-scale studies of land use effects on water quality focus exclusively on spatial scales larger than recommended wetland buffer widths, partly because the resolution of available land cover maps is often too low to examine scales smaller than 100 m (Knutson et al. 1999, Sliva and Williams 2001). However, given data of sufficient resolution, it should be possible to address the importance of wetland buffers relative to land-scape-scale uncropped land around wetlands.

We examined the question of whether the use of wetland buffers as defined in existing guidelines, or landscape-scale conservation of uncropped land is a more effective tool for protecting lentic agricultural wetlands from contamination by fertilizers and pesticides. With the help of high-resolution land cover maps, we used a focal patch study design to examine the effects of buffer size and landscape-scale uncropped cover on nitrate, atrazine, and neonicotinoid levels in wetlands. Specifically, we addressed three questions:

- Does increasing buffer width, or increasing the amount of contiguous uncropped land within recommended buffer width guidelines, reduce nitrate and pesticide levels in agricultural wetlands? If so, how large should wetland buffers be?
- 2. Does increasing uncropped land cover in the broader landscape reduce nitrate and pesticide levels in agricultural wetlands?
- 3. What is the relative importance of buffer size and landscape-scale uncropped cover for reducing nitrate and pesticide levels in agricultural wetlands?

METHODS

Study region

All research was conducted in the agriculture-dominated region of eastern Ontario, Canada, in the Mixedwood Plains ecozone (Fig. 1). Topography of this region is very flat, and soils are primarily brunisols and gleysols on a base of sedimentary rock. Approximately 50% of this region is farmed, 20% is woodland, 10% is wetland, 10% is a combination of woodland and wetland, and 4% is developed; the remainder consists of a variety of land uses such as quarries and abandoned farmland. Most woodland in this region is unmanaged. Nitrogen is applied to crop fields at rates of ~15–314 kg/ha, and total pesticides are applied at rates of ~1 kg/ha (Huffman et al. 2008, McGee et al. 2010).

Site selection

Overall, our goals in wetland selection were to minimize the correlation between the two predictor variables (landscape-scale uncropped cover and buffer size), avoid spatial gradients in predictor variable values, and minimize variation in potentially confounding variables. We achieved this in several steps. First, we used existing shapefiles and aerial photographs in ArcGIS (ESRI 2015) to locate wetlands in our study region. We then minimized variation in potentially confounding variables by limiting wetlands considered to those with clearly defined edges (that is, ponds) of similar sizes (<0.5 ha), that were at least 150 m from major watercourses and at least 40 m from roads, and that occurred in landscapes with low amounts of developed land and pasture (<10% cover of each) and low road density ($<2.5 \text{ km road/km}^2$, with all roads ≤ 2 lanes wide). This step resulted in ~300 potential study wetlands. Next, we estimated predictor variable values for each of these wetlands by measuring landscape-scale woodland around each wetland (our best measure of uncropped land during site selection) using existing shapefiles in ArcGIS, and measuring wooded buffer size (our best measure of uncropped buffers during site selection) from aerial photographs in ArcGIS. Note that buffers in our study region frequently varied in width around the wetland perimeter, likely because they existed for historical reasons rather than representing a conscious decision by landowners to implement buffers based on official guidelines. Once we had predictor variable estimates, we identified wetlands with the two rare predictor variable combinations, (1) small buffers and high landscape-scale uncropped cover and (2) large buffers and low landscape-scale uncropped cover, and that were at least 2 km apart, to avoid spatial autocorrelation as much as possible while providing adequate sample size. We contacted landowners who had wetlands in these categories on their properties, and received permission to study 9-10 wetlands for each of the rare predictor variable combinations. We then approached other landowners to acquire similar numbers of study wetlands—also at least 2 km apart—with the two more common predictor variable combinations, (3) small buffers and low landscape-scale uncropped cover, and (4) large buffers and high landscape-scale uncropped cover. To avoid spatial gradients in predictor variables, we selected these wetlands such that they were interspersed among the type (1) and (2) wetlands already selected (Fig. 1). We note that when we re-calculated buffer sizes later in this work (see Uncropped land, buffer size, and landscape scale),

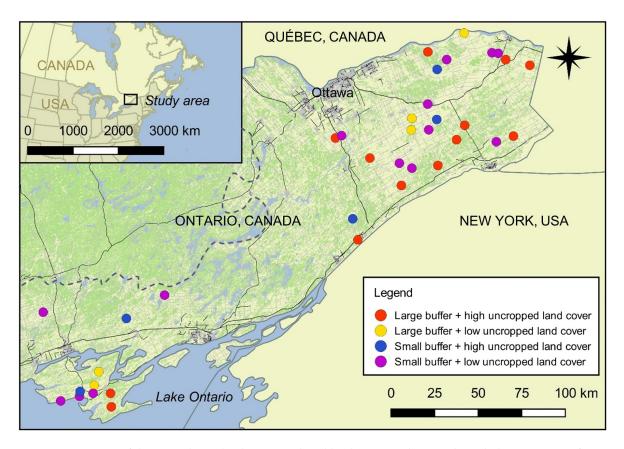


Fig. 1. Locations of the 37 study wetlands in agricultural landscapes in the Mixedwood Plains ecozone of eastern Ontario, Canada. The dashed line shows the boundary between the Mixedwood Plains ecozone (south and east) and the Boreal Shield (northwest). Wetlands were selected to minimize correlations between our two predictor variables—buffer size and landscape-scale uncropped cover—and to minimize spatial gradients in these variables by interspersing sites with dissimilar predictor variable values. Although buffer size and landscape-scale uncropped cover were continuous variables in analyses, for the purposes of this map, buffer widths are large if larger than the median buffer width (8 m) and small otherwise, and percent uncropped land cover is high if higher than the median percentage of land within 200 m of the wetland that was uncropped (44%), and low otherwise.

our distribution of study sites across predictor variable combinations (1–4) became less uniform, but a minimum of five study wetlands still fell into each of the four categories (Fig. 1).

Through this process, we ultimately selected 37 ponds for study (Fig. 1). All studied ponds fit the ecological definition of wetlands: Areas covered with water and characterized by vegetation adapted to water-saturated soils. Three of these study wetlands were natural; the remainder were originally dug or blasted but had not been used for agriculture (e.g., for irrigation or watering livestock) or quarrying for at least 5 or 20 yr,

respectively. None of the study wetlands was adjacent to pasture.

Data sources and software.—Most of the wetland, landscape, and buffer information used during site selection was collected from government maps and aerial photographs. Potential study wetlands were located using the Wetland Unit dataset (OMNR 2011), the Ontario Hydro Network Waterbody dataset (OMNR 2010), aerial photographs from the Digital Raster Acquisition Project Eastern Ontario (OMNR 2009), and satellite imagery from Google Maps (Google 2015). Buffer size was estimated from the above-mentioned

aerial photographs and satellite imagery. Woodland cover within a 1-km radius of potential study wetlands (our estimate of uncropped land cover during site selection) was based on the Agricultural Resource Inventory (OMAFRA 2010) and the Southern Ontario Land Resource Information System (OMNR 2002). Other information about surrounding landscapes was gathered from these sources, as well as from the Ontario Hydro Network Small Scale Watercourse dataset (OMNR 2012) and the National Road Network Ontario dataset (OMNR 2014). Information about wetland history and current usage was gathered by speaking with landowners and through field observations. All calculations of wetland size, landscape-scale variables, and buffer size were conducted in ArcMap 10.3.1 (ESRI 2015).

Nutrient and pesticide levels

We measured concentrations of nitrate, the herbicide atrazine, and four neonitcotinoid insecticides (acetamiprid, clothianidin, imidacloprid, and thiomethoxam) in all study wetlands during the 2015 growing season. Nitrate is one of the most common contaminants from agricultural fertilizers (Ongley 1996), and atrazine and neonicotinoids are common agricultural pesticides of concern (Graymore et al. 2001, Main et al. 2014, Schaafsma et al. 2015). In sufficient concentrations, all of these chemicals can be detrimental to aquatic organism health or survival, and can change community composition (Carpenter et al. 1998, Beketov et al. 2013, CCME 2014, Morrissey et al. 2015).

We collected two water samples from each wetland in June 2015—one to be assessed for nitrate and the other to be assessed for atrazine and neonicotinoids—as well as one sample in July 2015 to be assessed for nitrate. We expected agricultural chemical contamination to be greatest in June (Lapp et al. 1998, Byer et al. 2011, Main et al. 2014) and therefore had initially planned to sample water in June only. However, we took an additional water sample to test for nitrate in July because we had detected nitrate in very few wetlands in June and we wanted to be sure that we were not missing critical information. In each sampling period, we sampled southerly sites first, because we expected that fertilizers and pesticides would be applied earlier at lower latitudes. We took all water samples a minimum of 36 h after rain events of more than 2 mm, to avoid potentially confounding effects of major rains. On each sampling occasion, we collected the water sample(s) from one location in the wetland, away from dense vegetation or algae and ~2 m from shore, by wafting sampling bottles gently up and down through the middle 80% of the water column. Water samples to be analyzed for nitrate were collected in polyethylene terephthalate bottles, and those to be analyzed for pesticides in amber glass bottles. All water samples were refrigerated at 4–5°C until analysis and were analyzed by laboratories accredited by the Canadian Association for Laboratory Accreditation (Ottawa, Ontario, Canada).

One June water sample and the July water sample from each wetland were analyzed for nitrate within 5 d of sampling using ion chromatography with chemical suppression of eluent conductivity. This work was conducted by Laboratory Services at the City of Ottawa using an ion chromatograph (ICS-1000; Dionex, Sunnydale, California, USA) interfaced with Chromeleon analytical software (version 6.8; Dionex 2006; details in Haas et al. 2015, Sawatzky 2016). The lower detection limit for nitrate was 0.04 mg/L, which is <1% of the minimum concentration at which nitrate is thought to be detrimental to the long-term health of freshwater organisms (CCME 2014).

The second June water sample from each wetland was analyzed for atrazine and neonicotinoids (acetamiprid, clothianidin, imidacloprid, and thiomethoxam) in September-October 2015 using solid phase extraction followed by highperformance liquid chromatography-mass spectrometry (HPLC-MS; details in Prosser et al. 2016, Sawatzky 2016, Robinson et al. 2017). This work was conducted by Laboratory Services at Environment Canada's National Wildlife Research Centre. An Oasis HLB cartridge (225 mg, 60-µg particle size, Waters, Massachusetts, USA) was used for solid phase extraction, and a high-performance liquid chromatograph (1200 Series; Agilent Technologies, Santa Clara, California, USA) coupled with a mass spectrometer (API 5000 Triple Quadropole Mass Spectrometer and Turbo V; SCIEX, Framingham, Massachusetts, USA) and Analyst software (version 1.5; SCIEX 2010) for HPLC-MS. The lower detection limits were 0.00040 μg/L (atrazine), 0.00010 μg/L (acetamiprid),

 $0.00025~\mu g/L$ (clothianidin), $0.00025~\mu g/L$ (imidacloprid), and $0.00020~\mu g/L$ (thiamethoxam). These detection limits represent concentrations <1% of minimum concentrations thought to be detrimental to freshwater organisms in the long term (CCME 2014).

Nitrate concentrations were converted to binary values (0 if never detected or 1 if detected) due to low detection rates (see *Results*). Atrazine concentrations were used without modification. We summed acetamiprid, clothianidin, imidacloprid, and thiamethoxam concentrations to create a single variable, neonicotinoid concentration, because detection rates for several individual neonicotinoids were low (see *Results*), and because all four neonicotinoid insecticides are believed to have similar toxicity to aquatic organisms (Morrissey et al. 2015). If a pesticide was not detected in a wetland, its concentration was recorded as 0 µg/L.

Uncropped land, buffer size, and landscape scale

The two main predictor variables were (1) buffer size (Questions 1 and 3) and (2) uncropped cover in the landscape within an appropriate distance of each wetland (Questions 2 and 3). During site selection, we used woodland to estimate buffer size and uncropped land cover within 1 km of wetlands, because information about woodland was readily available and relatively precise, and woodland and agricultural cover are strongly negatively correlated in our study region ($r \approx -0.8$). Although this approximation was necessary during site selection, we manually obtained more detailed, up-to-date measures of predictor variables immediately after our water sampling period, in August 2015.

To update our predictor variable estimates, we took copies of the aerial photographs used during site selection to the field, and manually examined the area within 700 m of study wetlands, the maximum practical scale for this onthe-ground mapping. Due to the very flat topography of the study region, mapping cover types at a watershed scale was neither feasible nor likely to be a critical component in explaining wetland water quality. We identified land cover as row crop (e.g., corn or soybean), hay, pasture, orchard/vineyard, woodland, scrub, meadow, residential, road, or quarry, and recorded these cover types on the aerial photographs. We

digitized the resulting maps in ArcGIS to gain accurate estimates of land cover amounts around each wetland, and then estimated percent cover of uncropped land, buffer size, and landscape scale as follows.

Uncropped land.—We defined uncropped land in the same way both in wetland buffers and at the landscape scale. We focused on uncropped land because buffer guidelines in agricultural areas usually focus on the proximity of crops to water bodies (C. Flemming, personal communication), and as such, they effectively consider all uncropped land to be equivalent. We considered that hay and row crops are frequently rotated in our study region between and even within years, so we defined uncropped land as land used neither for row crops nor for hay during the study period. Most uncropped land was woodland, scrub, or meadow, with small amounts of residential land, pasture, orchards, roads, or quarries.

Buffer size.—We measured buffer size in three ways: wetland buffer width, area-based correspondence with buffer guidelines, and perimeter-based correspondence with buffer guidelines (Fig. 2). Buffer width was measured as the minimum width of adjacent uncropped land, in meters, as per Ontario guidelines (C. Flemming, personal communication). Area-based correspondence with buffer guidelines was measured as the percentage of the area circumscribed by a recommended buffer width that contained uncropped land contiguous with the wetland edge. Perimeter-based correspondence with buffer guidelines was measured as the percentage of a wetland's perimeter that was contiguous with uncropped land at least as wide as a recommended buffer width. For the area-based and perimeter-based measurements of correspondence with buffer guidelines, we examined seven buffer widths recommended in Ontario: 5, 9, 13, 16, 30, 50, and 120 m (Coukell et al. 2004, OFEC et al. 2004, Niagara Planning and Development Services Department 2014). Although we focused on Ontario guidelines here, our findings are relevant to other regions because most buffer policies around the world have minimum width recommendations of 5-50 m, with occasional recommendations of ~100 m (State of Western Australia 2005, McElfish et al. 2008, European Court of Auditors 2014, Macfarlane et al. 2014).

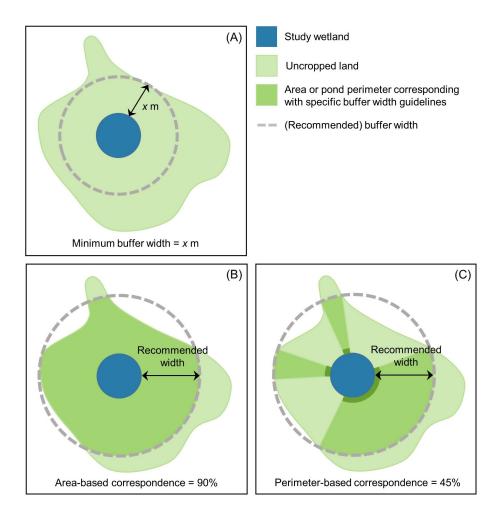


Fig. 2. The three ways that buffer size was measured. (A) Buffer width was measured as the minimum distance from the wetland edge to crop fields. (B) Area-based correspondence with buffer guidelines was measured as the percentage of the area within a recommended buffer width that contained uncropped land contiguous with the wetland edge. (C) Perimeter-based correspondence was measured as the percentage of the wetland perimeter that had a buffer at least as wide as a recommended buffer width.

We note that by measuring buffer sizes in the field in August, we obtained a generous, rather than conservative, estimate of buffer size. Wetlands in our area are largest in spring after snowmelt, and subsequently shrink. Thus, if a farmer planted crops to the edge of a wetland (zero buffer) in spring, by August, when we measured buffer size, the wetland would have appeared to have a buffer several meters in width. Buffer guidelines are unclear about the time of year at which buffers should be measured, but we estimate that our measured buffers of 2 m or less in August represent ponds that earlier in the year would have had zero buffer.

Landscape scale.—We measured percent uncropped land cover within 50, 100, 150, 200, 250, 300, 400, 500, 600, and 700-m radii around wetlands, and determined the most appropriate landscape scale using multi-scale analyses (see *Analyses*). We used 50 m as the smallest radius to ensure that our landscapes were larger than most recommended buffer widths for aquatic habitat protection (typically ≤50 m, although they can reach >100 m), and 700 m as the largest due to logistical constraints. We examined radii in increments of 50 or 100 m so that we would have high precision when finding the appropriate landscape scale, while also examining landscape

scales with different enough uncropped cover values that differences in models run at various landscape scales (see *Analyses*) would be meaningful.

Analyses

Our main study questions dealt with the effects of buffer size and landscape-scale uncropped cover on wetland nutrient and pesticide levels. Before we could address these questions, we conducted a preliminary analysis to find the appropriate landscape scale(s), that is, the scale(s) at which uncropped cover most strongly affected our water quality variables. We then evaluated our main study questions by examining the relationships between nitrate/atrazine/neonicotinoid concentrations and (1) buffer size (Questions 1 and 3) and (2) landscape-scale uncropped cover (Questions 2 and 3). Post hoc, we checked for effects of geographical location by comparing response variable values found in the southwestern vs. northeastern portions of our study area (Fig. 1) and checking residuals of all models for spatial autocorrelation. All statistical tests were performed in R (version 3.4.3; R Core Team 2017). R² values, regression coefficients, and P values were calculated using base R packages. Pseudo-R² values were calculated using the package, pscl (Jackman 2017). Moran's I spatial autocorrelation tests were performed using the package, ape (Paradis and Schliep 2018). We omitted one of the 37 study wetlands from analyses of nitrate because we were only able to visit it once, rather than twice. Data used in these analyses are available through Mendeley Data (Sawatzky and Fahrig 2019).

Landscape scale.—We found the most appropriate landscape scale for each water quality variable by examining the relationships between the water quality variables and percent cover of uncropped land within 50, 100, 150, 200, 250, 300, 400, 500, 600, or 700 m from the wetland edge. For nitrate, we conducted a logistic regression of nitrate detection (0 = not detected, 1 = detected) on percent uncropped land cover, and calculated Nagelkerke's pseudo- R^2 , at each spatial scale. For atrazine and neonicotinoids, we conducted log-linear regressions of pesticide concentrations on percent uncropped land cover, and calculated R^2 , at each spatial scale. The appropriate landscape scale for each water

quality variable was indicated by the analysis producing the highest pseudo- R^2 or R^2 value for the relationship between that variable and uncropped land cover. Based on these analyses, in addressing our main objectives we identified 150 m as the landscape radius for nitrate, 300 m as the landscape radius for atrazine, and 200 m as the landscape radius for neonicotinoids (see *Results*).

Evaluation of study questions.—We evaluated and compared the effects of buffer size and landscape-scale uncropped cover on nitrate detection and atrazine and neonicotinoid concentrations using multiple regression. The full models for each water quality variable were structured as follows: Water quality variable = $b_0 + b_1 \times$ (buffer size) + $b_2 \times$ (uncropped cover in landscape at its appropriate scale). We measured buffer size in 15 ways: buffer width, area-based correspondence with seven different buffer guidelines, and perimeter-based correspondence with seven different buffer guidelines. Therefore, for each of the three water quality response variables we examined 15 full models, each with one of the 15 buffer variables and uncropped cover in the landscape (at its appropriate scale). For each of the three water quality response variables, we also evaluated all 16 of the sub-models containing only one predictor each, that is, one of the 15 ways of measuring buffer size or the landscape predictor variable (uncropped cover at its appropriate scale), and evaluated the intercept-only null model. The landscape variable was always percent uncropped cover within the area from the edge of the wetland to the appropriate landscape scale of 150, 300, or 200 m, for nitrate, atrazine, and neonicotinoids, respectively. We used logistic regression models for nitrate (0 = not)detected, 1 = detected), and log-linear regression models for atrazine and neonicotinoid concentrations. We standardized all predictor variables to a mean of 0 and standard deviation of 1 for these analyses. For each water quality variable, we evaluated buffer size effects (Question 1) by looking at the 15 sub-models with only a buffer size predictor variable—that is, water quality variable = $b_0 + b_1 \times$ (buffer size)—and we evaluated landscape-scale uncropped cover effects (Question 2) by looking at the one sub-model with only uncropped cover in the landscape at its appropriate spatial extent—that is, water quality variable = $b_0 + b_2 \times$ (uncropped cover in landscape at its appropriate scale). To evaluate significance, we used $\alpha = 0.05$. We evaluated relative effects of buffer size and uncropped cover in the landscape (Question 3) by examining the 15 full models that contained both a buffer size variable and uncropped cover in the landscape—that is, water quality variable = $b_0 + b_1 \times$ (buffer size) + $b_2 \times$ (uncropped cover in landscape at its appropriate scale). Here we examined not only which predictor variables had significant effects, but also the relative effect sizes (partial standardized regression coefficients) of the predictor variables.

Effects of geographical location.—After addressing the main study questions, we conducted further tests to confirm that geographical position did not unduly influence our results. First, we used Moran's I tests to check for spatial autocorrelation of model residuals. In addition, because our study sites occurred in two distinct parts of the Mixedwood Plains ecozone (northeast and southwest, henceforth sub-regions; Fig. 1), and nitrate detection rates differed between these two sub-regions (Appendix S1: Fig. S1), we re-ran all models used to evaluate study questions (Evaluation of study questions), with study sub-region incorporated as an additional predictor variable, to see if the previously observed effects of predictor variables would still be present after accounting for sub-region.

RESULTS

Nutrient and pesticide levels

Nitrate was detected in 16 of 36 wetlands, with a maximum concentration of 14.28 mg/L. Atrazine was detected in all 37 wetlands, with concentrations of $0.0051-1.6~\mu g/L$. Acetamiprid, clothianidin, imidacloprid, and thiomethoxam were detected in 2, 25, 10, and 18 wetlands, respectively; at least one of these neonicotinoids was detected in 28 of 37 wetlands. The maximum individual neonicotinoid concentration was $1.3~\mu g/L$ (thiamethoxam), and the maximum total neonicotinoid concentration was $1.3231~\mu g/L$.

Landscape scale and predictor variables

Landscape scales for the effect of uncropped cover were 150 m for nitrate detection, 300 m for atrazine concentration, and 200 m for

neonicotinoid concentration (Fig. 3). Uncropped land cover at these spatial scales around study wetlands ranged from 11% to 99% (mean = 49%). Measured buffer widths were 0.85–135 m (mean = 23 m). Five wetlands had buffers <2 m wide. As we measured buffer width in August after the summer dry-down, these wetlands likely had zero buffer in spring when the crops were planted.

Area and perimeter-based buffer sizes within a given width guideline were highly correlated with each other (Spearman's $\rho = 0.901-0.984$, depending on the width guideline) and ranged from 30% to 100% for 5-m guidelines, and 0% to 100% for 120-m guidelines. As expected (see *Site* selection), correlations between buffer size and landscape-scale uncropped cover were relatively low, with most Spearman's ρ < 0.6, although these correlations did increase in strength when buffer size was measured within larger width guidelines (Table 1). Neither buffer size nor landscape-scale uncropped cover was correlated strongly with latitude, longitude, wetland area, broader-scale water cover, road density, or developed land cover (Table 2).

Evaluation of study questions

Buffer effects on water quality (Question 1).— There was little evidence for an effect of buffer size on nitrate detection, atrazine concentration, or neonicotinoid concentration, regardless of whether buffer size was measured as minimum buffer width or as correspondence with buffer guidelines. Exceptions occurred only when 120-m buffer guidelines were considered: In relation to this width guideline, the likelihood of nitrate detection decreased with area-based buffer size (P = 0.043), and neonicotinoid concentration significantly decreased with both area-based and perimeter-based buffer size (P = 0.007 and P = 0.002, respectively).

Landscape effects on water quality (Question 2).— Nitrate detection probabilities, atrazine concentrations, and neonicotinoid concentrations in wetlands decreased significantly with the proportion of uncropped land cover within the surrounding landscape (nitrate—landscape radius = 150 m, P = 0.019; atrazine—landscape radius = 300 m, P = 0.023; neonicotinoids—landscape radius = 200 m, P = 0.001; Fig. 4).

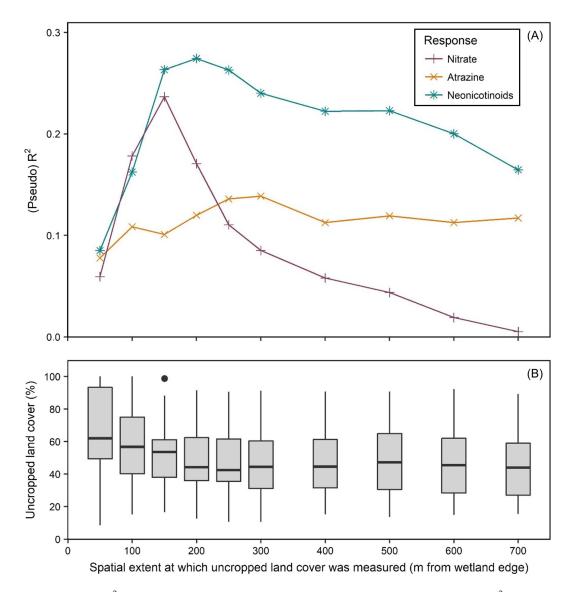


Fig. 3. (A) Pseudo- R^2 values for logistic regressions of nitrate detection in 36 wetlands, and R^2 values for loglinear regressions of neonicotinoid and atrazine concentrations in 37 wetlands (36 of which were the same wetlands as for nitrate detection), on percent cover of uncropped land in the surrounding landscape. Uncropped land cover was measured between the wetland edge and spatial extents of 50–700 m. Pseudo- R^2 values were calculated using the Nagelkerke method. Spatial extents with the strongest relationship (largest pseudo- R^2 or largest R^2) between water quality variables and uncropped land cover were 150 m for nitrate detection, 200 m for neonicotinoid concentration, and 300 m for atrazine concentration. These spatial extents were used as the landscape scales in subsequent analyses. (B) Distribution of percent cover of uncropped land at spatial extents of 50– 700 m around 37 wetlands—the independent variable in (A).

Relative effects of landscape-scale uncropped land and wetland buffer size on water quality (Question 3).—Landscape-scale uncropped cover had more consistent and stronger effects than did buffer size on nutrient and pesticide levels in study wetlands. In additive models containing both landscape-scale uncropped cover and a buffer size predictor variable, landscape-scale uncropped

Table 1. Spearman's rank correlation coefficients between buffer size predictor variables and landscape-scale uncropped cover predictor variables for 37 study wetlands.

Variable	Uncropped land cover within 150 m	Uncropped land cover within 200 m	Uncropped land cover within 300 m	
Buffer width	0.67	0.50	0.21	
Area-based correspondence with 5-m guideline	0.50	0.38	0.12	
Perimeter-based correspondence with 5-m guideline	0.50	0.38	0.13	
Area-based correspondence with 120-m guideline	0.96	0.83	0.56	
Perimeter-based correspondence with 120-m guideline	0.93	0.87	0.66	

Notes: Buffers were considered to include uncropped land contiguous with wetlands. Buffer size was measured either as minimum buffer width or as the percent correspondence between the actual buffer and recommended buffer size guidelines, ranging from 5 to 120 m. For brevity, only correlations for buffer width and 5 and 120 m guidelines are presented here. Landscape-scale uncropped land was the percentage of land within a given distance from the wetland edge that was uncropped. Only 150, 200, and 300-m landscape scales are included here as these were the radii with the strongest effects on nitrate, neonicotinoid, and atrazine concentrations, respectively.

cover nearly always had a significant effect on nutrient and pesticide concentrations (13/15 models for nitrate detection, 11/15 models for atrazine concentrations, and 13/15 models for neonicotinoid concentrations; Appendix S2: Tables S1-S3). Exceptions occurred when buffer size was measured at larger spatial extents, that is, ≥50 m, and was thus more correlated with the landscape-scale measure of uncropped land (Appendix S2: Tables S1–S3). In contrast, buffer size never had a significant effect on nutrient or pesticide concentrations in any of the additive models. In addition, landscape-scale uncropped cover always had a larger standardized partial regression coefficient in additive models than did buffer size (Fig. 5).

Effects of geographical location

Evidence for effects of geographical location on nitrate, atrazine, and neonicotinoids differed depending on the water quality variable. Residuals for ~10% of nitrate detection models were significantly spatially autocorrelated (Appendix S1: Table S1), and nitrate was detected more often in southwestern study sites than in northeastern ones (Appendix S1: Fig. S1). When sub-region was added to nitrate detection models, approximately half of the previously observed significant effects retained their statistical significance (Appendix S2: Table S1). In contrast, we did not observe any spatial autocorrelation of model residuals for atrazine or neonicotinoid concentrations (Appendix S1: Tables S2 and S3), and neither atrazine nor neonicotinoid concentration differed significantly between study sub-regions (Appendix S1: Fig. S1). When sub-region was added to atrazine and neonicotinoid concentration models, all previously observed significant effects remained significant (Appendix S2: Tables S2 and S3). Interestingly, even after accounting for sub-region, landscape-scale uncropped cover always had a larger standardized partial regression coefficient in additive models than did buffer size, regardless of the water quality response variable.

DISCUSSION

Nitrate and pesticide levels in study wetlands ranged from undetectable to harmful. Canadian Environmental Quality Guidelines recommend that for long-term protection of freshwater organisms, nitrate concentrations should be <13 mg/L, atrazine concentrations <1.8 μ g/L, and imidacloprid (neonicotinoid) concentrations <0.23 μ g/L (CCME 2014). These limits were exceeded in four of our 37 study wetlands—we observed one nitrate concentration of 14.28 mg/L, and neonicotinoid concentrations of 0.29, 0.40, and 1.32 μ g/L. We also detected 12.4 mg/L nitrate and 1.6 μ g/L atrazine in two wetlands, which approach the harmful thresholds.

Our results indicate that landscape-scale uncropped cover at relatively small spatial extents (150–300 m landscape radius) can reduce nutrient and pesticide levels in wetlands more effectively than compliance with buffer guidelines as they are currently written. This result is

Table 2. Spearman's rank correlation coefficients between predictor variables and potentially confounding variables for 37 study wetlands.

Variable	Latitude	Longitude	Wetland area	Water cover within 1 km	Road density within 1 km	Developed land cover within 1 km
Buffer width	0.15	0.28	-0.21	0.17	-0.16	-0.04
Area-based correspondence with 5-m guideline	-0.01	0.18	-0.16	0.04	-0.09	-0.03
Perimeter-based correspondence with 5-m guideline	< 0.005	0.19	-0.15	0.02	-0.12	-0.05
Area-based correspondence with 120-m guideline	0.21	0.34	-0.24	0.29	0.07	0.05
Perimeter-based correspondence with 120-m guideline	0.15	0.33	-0.15	0.37	0.06	0.07
Uncropped land cover within 150 m	0.14	0.28	-0.17	0.34	0.11	0.04
Uncropped land cover within 200 m	0.15	0.25	-0.06	0.42	0.20	0.06
Uncropped land cover within 300 m	0.15	0.17	0.05	0.53	0.26	-0.03

Notes: Predictor variables included buffer size and landscape-scale uncropped cover. Buffers were considered to include uncropped land contiguous with wetlands. Buffer size was measured either as minimum buffer width or as the percent correspondence between the actual buffer and recommended buffer size guidelines ranging from 5 to 120 m. For brevity, only correlations for buffer width and 5 and 120-m guidelines are presented here. Landscape-scale uncropped land was the percentage of land within a given distance from the wetland edge that was uncropped. Only 150, 200, and 300-m landscape scales are included here as these were the radii with the strongest effects on nitrate, neonicotinoid, and atrazine concentrations, respectively.

highly robust. Although we examined models using 15 different buffer size measurements for each wetland, which should have increased the probability of finding spuriously significant results, we found few significant effects of buffer size. The only exceptions were when buffer size was measured in relation to a 120-m buffer guideline—that is, as the percent cover of contiguous uncropped land within 120 m of the wetland or as the proportion of the wetland perimeter with contiguous uncropped land at least 120 m wide—and no landscape-scale variable was included in the model. However, the 120-m buffer guideline is very close to the empirically determined landscape scales of 150-300 m, and buffer sizes measured within the 120-m buffer guideline were correlated with landscapescale uncropped cover (Table 1), suggesting that the apparent effects of buffer size within the 120m buffer guideline could be due to landscapescale uncropped land. In further support of this inference, buffer size never had a significant effect on nitrate detection or pesticide concentrations when landscape-scale uncropped cover was also included as a predictor variable in the model. In contrast to the lack of buffer effects, we found consistently strong effects of uncropped land cover in the landscape on nitrate detection and atrazine and neonicotinoid concentrations. Uncropped land cover at a landscape scale always significantly affected nitrate or pesticide concentrations in univariate models. The effect of uncropped land on atrazine and neonicotinoid concentrations remained statistically significant after accounting for sub-region (Appendix S2: Tables S2 and S3), although its effect was less significant on nitrate after accounting for sub-region (P = 0.065; Appendix S2: Table S1), likely because nitrate detection was somewhat spatially autocorrelated (Appendix S1: Table S1) and because models for nitrate detection had a simplified response variable (nitrate concentration converted to nitrate detection, with 0 = not detectedand 1 = detected) and were thus less likely to produce significant results in the first place than models with continuous response variables. In additive regression models, landscape-scale predictor variables always explained more variation in nitrate and pesticide levels than did buffer size (Fig. 5). This was true even in the few (8/45)models where the landscape-scale variable did not have a statistically significant effect; the lack of effect in these cases was probably because buffer size was measured within guidelines beginning to approach the landscape extent such that the two predictor variables were correlated. This

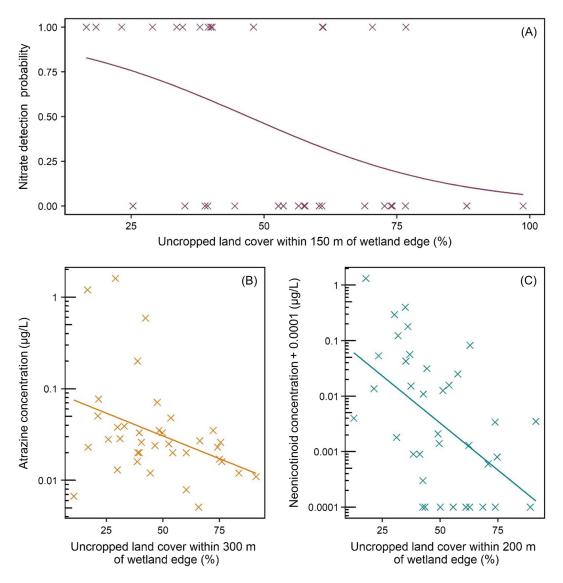


Fig. 4. The effect of uncropped land cover on (A) nitrate detection, (B) atrazine concentration, and (C) neonicotinoid concentration in wetlands (n = 36 for nitrate detection; n = 37 for atrazine and neonicotinoid concentrations). All relationships were statistically significant (P < 0.05).

was also true after accounting for study subregion. In other words, uncropped land does not need to be contiguous with wetlands to reduce contamination by nitrate, atrazine, or neonicotinoids, and landscape-scale uncropped cover affects levels of these chemicals more strongly than does buffer size.

Nitrate detection rates, atrazine concentrations, and neonicotinoid concentrations were significantly lower in wetlands that had more uncropped land within 150, 300, or 200 m of the

wetland edge (Fig. 4), respectively. We acknowledge that the data support the choice of landscape spatial scale more clearly for nitrate detection and neonicotinoid concentration than for atrazine concentration (Fig. 3), but overall our data indicate scales of effect of 150–300 m. These empirically determined landscape scales are smaller than those reported for rivers and streams, which often respond to landscape composition at the scale of a catchment or watershed (Sliva and Williams 2001, King et al. 2005). They

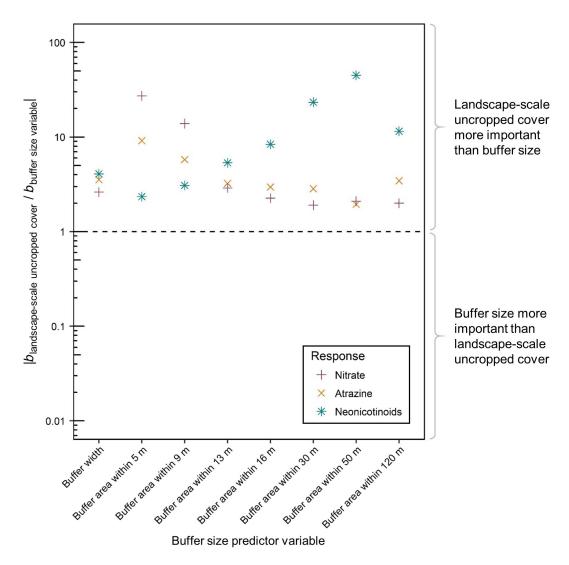


Fig. 5. Absolute values of the ratios between partial standardized regression coefficients for landscape-scale uncropped cover and buffer size, when water quality response variables in wetlands were regressed on both predictor variables in additive models. A |ratio| > 1 (above dashed line in figure) indicates an additive model where landscape-scale uncropped cover had a larger standardized partial regression coefficient than did buffer size. Landscape-scale uncropped cover was measured as percent cover within 150, 300, and 200 m for nitrate, atrazine, and neonicotinoid response variables, respectively. Buffer size was measured as buffer width or as areabased buffer size using one of seven buffer width guidelines (see Fig. 2); partial regression coefficients for perimeter-based buffer size (not shown) were very similar to values for area-based buffer size.

are also smaller than those reported for large wetlands: For example, Houlahan and Findlay (2004) empirically determined that woodland cover within 2000–2500 m of focal wetlands most strongly explained variation in water quality. However, Declerck et al. (2006), examining water quality in small farmland ponds, found suspended solids to most strongly decrease with woodland cover and increase with crop cover within 100 m of the pond, despite examining spatial scales up to 3200 m in radius. Although we measured different water quality variables, we found similarly small landscape scales. Another possible reason for our small landscape scales is

that the scale of effect of the uncropped land in the landscape on nitrate and pesticide levels is larger than the maximum scale we studied, that is, 700 m. However, this is unlikely because the effects of uncropped land cover on nitrate detection and pesticide concentrations leveled off or weakened beyond 150–300 m (Fig. 3).

Due to the high correlation between uncropped land and natural land (mainly woodland) in our study region, we cannot say whether the observed effects of uncropped land at a landscape scale on nitrate and pesticide levels are caused by reduced cropland leading to decreased chemical application, or by increased permanent vegetation leading to increased interception of contaminants. Previous studies have found some evidence for both reduced agriculture and increased forest cover reducing water contamination (Crosbie and Chow-Fraser 1999, Declerck et al. 2006, Lee et al. 2009); however, the relative importance of the two variables is unclear in the literature. Disentangling the effects of woodland and cropland on wetland contaminant concentrations would require an additional study in which study landscapes are selected to minimize their correlation. However, whatever the underlying mechanism, our results suggest that reducing cropped land cover in favor of uncropped land can reduce nitrate and pesticide levels in agricultural wetlands.

There are two main, non-exclusive potential explanations for why we did not observe wetland buffer effects on nitrate, atrazine, or neonicotinoids. First, wetland buffers may not be effective against these contaminants in regions with flat topography. Our study sites were all in relatively flat areas and may have been fed less by surface runoff than by groundwater or subsurface runoff. A meta-analysis of nitrogen removal in buffer zones found that buffer size had no effect on nitrogen transport by subsurface runoff (Mayer et al. 2007). Presumably buffers would have even less of an effect on contaminant transport in groundwater. Second, wetland buffers may intercept highly soluble compounds less effectively than sediment-bound compounds. The focal compounds in our study-nitrate, atrazine, and neonicotinoids—are all highly soluble. Although several studies have shown buffers to successfully intercept soluble compounds such as nitrate, it is generally acknowledged that buffers are more effective at intercepting particulate or sediment-bound pollutants than those in solution (Schmitt et al. 1999, Helmers et al. 2008).

Based on these arguments, we speculate that our observed lack of buffer effect may apply to many other agricultural systems. Many agricultural regions are flat (e.g., North America's Prairie Pothole Region, England's Fens, and China's Huang-Huai-Hai Plain), and several components of fertilizers and pesticides are highly soluble. If buffers are generally ineffective in this combination of conditions, then conservation planners in many agricultural regions should change their focus from implementing buffers around wetlands to limiting agricultural expansion and/or limiting the use of fertilizers and pesticides on agricultural lands.

On the other hand, it could be argued that the lack of buffer effect we observed was because even the smallest buffers reduced contaminants in our wetlands. In other words, what matters for contaminant interception is the presence of a buffer, not the size of the buffer. Although five of our wetlands had essentially zero buffer in the early spring, with crops planted very close to the edge of the wetland, these buffers became wider throughout the summer as wetlands dried down. Thus, all wetlands may have been buffered from contaminants to some extent. However, if these small buffers had been highly effective in reducing contaminants, we should not have observed the strong effects of uncropped cover in the landscape that we did observe. Therefore, we infer that the effectiveness of small buffers is very limited, and that the lack of buffer effect that we observed is real.

In short, our results indicate that wetland buffers as currently defined in agricultural guidelines do not effectively reduce overall nitrate and pesticide levels in small water bodies in flat agricultural areas during the growing season. Buffers may still help to reduce acute contaminant pulses associated with major rain events, or protect against contaminants other than those we measured, but we cannot draw such conclusions from our study.

It is important to note that our study was limited to the role of uncropped land—whether in buffers or at a landscape scale—in reducing nitrate and pesticide levels in wetlands. Thus, evaluating the role of specific vegetation types

such as shrubs or grasses, or examining ways to protect wetlands from other sources of contamination such as roads or urbanization, were outside the scope of our study. During our rigorous site selection process, we aimed to minimize the variation of potentially confounding variables. Some of these variables, especially roads and urban development, may themselves affect the water quality of wetlands. Different kinds of noncrop vegetation may also have different effects on nitrate and pesticide concentrations in wetlands. Further study will be necessary to determine how contaminants from other land uses can be best prevented from entering wetlands, and whether the benefits of uncropped land can be enhanced by using certain vegetation types.

With these caveats in mind, we recommend that to reduce nitrate and pesticide contamination of agricultural wetlands targeted for conservation, landscape-scale conservation should be pursued rather than the current focus on buffer guidelines, particularly in regions with flat topography. Uncropped land should be maintained within a 150 to 300-m radius of focal wetlands. This could be done through mandating 150 to 300-m wide wetland buffers such that 100% of the land within that radius would be conserved, or by instituting landscape-scale protection measures that require landowners to conserve a certain proportion of uncropped land within 150-300 m of wetlands on their properties. For example, our results suggest that about 40% of the landscape in uncropped land would be sufficient to reduce nitrate and pesticide concentrations in wetlands (Fig. 4).

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