



Impact of Intensive Agricultural Production on the Ecotoxicologic Quality of Associated Medium-Order Streams: Cereal and Oilseed *versus* Horticultural Production

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Abstract

Streams associated with agroecosystems receive inputs of chemicals used within a basin that negatively impact its environmental quality. In this work, we aimed at comparing, through a battery of ecotoxicological tests, the relative impact of the cereal and/or oilseed and vegetable and/or flower agricultural-production models on the ecotoxicologic quality of both the water column and the bottom sediments of medium-order streams. The study, performed over 4 years, involved two major agroproductive areas of Argentina, one predominating in cereal and/or oilseed crops (Area 1), the other in vegetable and/or flower agriculture (Area 2). Both productive systems impacted the associated surface water bodies negatively, with the intensive production of vegetables and flowers producing greater ecotoxicologic effects on diagnostic organisms. The intensive-agriculture systems associated with Area 2 caused greater negative impacts on the water column than those of Area 1, with this pattern occurring in reverse for the bottom sediments. Furthermore, the samples from the sites associated with horticulture were more frequently toxic than those from Area 1. Of the organisms used to assess sample toxicity—*Lactuca sativa*, *Daphnia magna*, and *Hyallela curvispina*—*L. sativa* was the most sensitive to the type of contaminants associated with the form of agricultural land use; whereas no differences in sensitivity were observed between the two crustaceans. We found that the sublethal effects were significantly more sensitive than the lethal. The findings from this work would strongly advocate more sustainable agricultural-management plans that employed phytosanitary products whose action were more environmentally sustainable.

Keywords Bioassays · Intensive agriculture · Water quality · Ecotoxicity

Introduction

Agriculture is considered the main cause of the degradation of surface and groundwater resources (FAO 2004). An estimated three million metric tons of pesticide active

ingredients are applied to cropland annually worldwide (Popp et al. 2013). The streams associated with agroecosystems receive inputs of agrochemicals by drift and runoff, becoming sinks of the chemicals used within the basin that lead to adverse influences on environmental quality (Scanlon et al. 2007; Stehle and Schulz 2015). An assessment of the full impact of agricultural pesticides on associated surface water bodies is essential for designing more sustainable and productive management strategies. The analysis, however, is highly complex because of the diffuse nature of the diverse sources of emission and the large geographical area that this activity occupies.

In countries with strong agroproductive profiles, the agricultural area is dominated by the cultivation of oilseeds and cereals whose production is mainly destined for export, while horticulture occupies the rest of the areas in the periurban belts around the main cities, with the output being mainly used to cover domestic demand (SCI 2012). In Argentina, for

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example, the area used for cereal and oilseed cultivation (soybeans, corn, wheat, sunflower, barley, sorghum, and cotton) reaches 37 million hectares, with soybeans covering almost half of that area (MAGyP 2020). The tillage systems used in those regions are quite limited, and the variety of plant-protection products is less than in more complex agroecosystems. Conversely, in horticulture, a broad range of plant species is commonly grown with different tillage systems, requiring a wide variety of agricultural supplies for soil fertilization and pest control (Colamarino et al. 2006). This substantial difference prompted us to speculate that these two contrasting models of agricultural cultivation could impact the associated natural ecosystems quite differently.

Among the tools provided by the different disciplines of environmental sciences to assess the impacts of agricultural activities on surface water bodies, ecotoxicological bioassays provide a simple and practical way of directly estimating the potential effects that agroinputs can have on the associated biota independently of the compounds present through taking into account the different types of possible interaction between those compounds (additivity, synergism, antagonism) (Newman 2015). Usually, strategies to evaluate the ecotoxicologic impacts of the use of pesticides consist in studying the toxicity of individual (or less commonly the mixed) plant-protection products on a model species in the laboratory, having detected several adverse effects (Sobrero et al. 2007; Ronco et al. 2008; Barreto et al. 2020; Venturino et al. 2007; Anderson et al. 2015). While this type of study is very useful in ascertaining the potential effects of pesticides, due to in laboratory pesticides toxicity is usually higher (Yorlano et al. 2021) an extrapolation of such results to real field circumstances has always been a major challenge. A step forward in achieving a greater realism in such assessments consists of working with batteries of toxicity tests that incorporate several organisms of different species along with evaluating the toxicity of natural water and sediment samples that had been under previous agricultural influence, since this approach facilitates an independent means of estimating the impact of the compounds used in the productive system. Studies of this type have already been carried out to evaluate the impacts of contrasting anthropic uses of the soil in urban-versus-agricultural environments on the associated surface water bodies (Ding et al. 2010; Rimoldi et al. 2018; Stone et al. 2014); but, to the best of our knowledge, no studies involving a comparison of the ecotoxicological impacts of different agricultural-production systems have as yet been reported.

In view of these antecedents, the aim of the present study was to determine the relative impact of the agricultural-production models used in the cereal and/or oilseed crops and in horticulture on the ecotoxicologic quality of medium-order streams through a battery of bioassays and evaluating both the water column and the bottom sediments.

Materials and Methods

Description of the Study Areas and the Sampling Sites

The study was carried out in two major agroproductive areas of Argentina (Fig. 1). Area 1 corresponded to a sector located north of the Buenos-Aires province. This zone is one of the main oilseed-production regions of the country (mainly soybean) and has been extensively cultivated for several decades. The landscape is dominated by rural areas with few urban agglomerates of low-to-medium population density and with close links to agricultural activity (Piquer-Rodríguez et al. 2018).

Area 2 is located in the periurban sector of the La-Plata city (southeast of Buenos-Aires city) and is one of the most productive horticultural belts of the country with a wide variety of species being grown in about 7500 ha of fields and greenhouses (SCI 2012). The predominant production in the area is family-style with several management systems based on different agricultural supplies (Blandi et al. 2016).

A reference area was also selected (Area 3), located in Punta Indio (about 80 km southeast of the city of La Plata) within a sector declared by United Nations Educational, Scientific and Cultural Organization as a World Biosphere Reserve, where anthropic activity is scarce. The areas have a large number of medium-order streams typically through plains, with meandering watercourses of low flow rate, running toward the Paraná River (Area 1) or into the Río-de-la-Plata estuary (Area 2, Area 3).

Small water bodies are freshwater ecosystems of great relevance, as they contain greater biodiversity compared to larger systems; but they receive a higher input of pesticides because of their close proximity to adjacent agricultural fields and their low volume, or discharge, of water. Because, at the same time, small streams feed larger watersheds downstream, an understanding of the pattern of exposure in small water bodies will facilitate an awareness of the need to implement measures aimed at reducing the movement of pesticides from agricultural areas.

In each productive area (Area 1 and Area 2), nine independent sampling sites were selected, located in sectors where each has an exclusive agricultural influence (Fig. 1); whereas in the reference area contained 3 sampling sites were chosen.

Sampling and Characterization of the Water Bodies

Four sampling campaigns were carried out in each area during the spring-summer period of the years 2012 to 2015. In all instances, the samples were taken at least 72 h after a rainfall to study the different systems under similar conditions. In each sampling campaign, the areas surrounding the sampling sites were visually inspected in order to detect

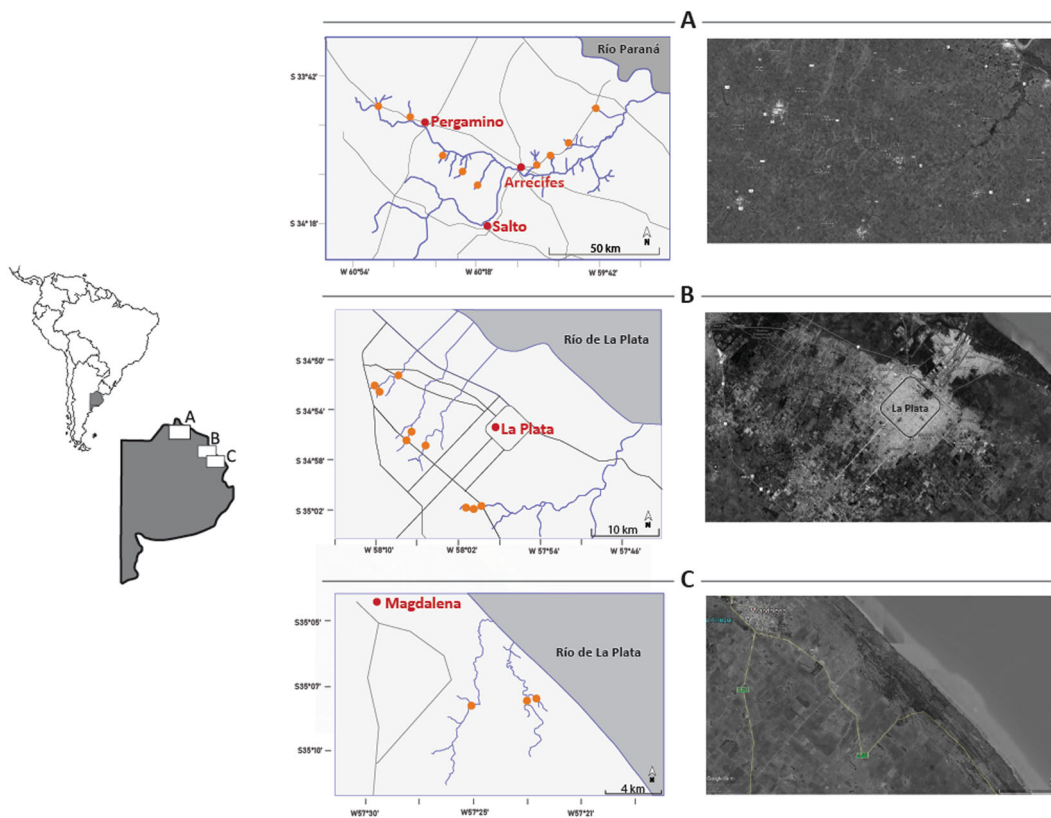


Fig. 1 Location of the sampling sites and satellite images corresponding to the extensive-production zone in the northeast of Buenos Aires (A), the intensive-production zone in the horticultural belt of the periurban area of La Plata (diamond-shaped area indicated in B), and reference area (C). The inset to the left of the panels illustrates the orientation of the sampling areas (white boxes) within the Province of

Buenos Aires and, in turn, the location of that province within Argentina and South America in general. The satellite images are registered exactly in correspondence with the respective maps of the sampling areas in the three panels. The sampling sites in the maps (red dots), accordingly are visible in the corresponding locations within the satellite images

potential changes in land use in the areas of greatest influence. In addition, the pH, temperature, dissolved-oxygen concentration, conductivity, total dissolved solids, and salinity of the water were determined with a Horiba U52 Multiparameter Meter™ in situ, and samples of the sediment and surface water were collected.

The samples obtained from the water were collected in plastic jars of 10 L for ecotoxicological testing then transported to the lab, where they were kept at 4 °C for no longer than 2 weeks before toxicity testing. In addition, the total hardness, alkalinity, and organic matter in the water were measured in the laboratory following standardized methodologies (APHA 1998, methods 2340, 2320).

Sediment samples were removed with an Eckman grab either from the riverbank or else by wading into the shallow water. Fine-grained sediments were preferentially collected at each site because hydrophobic pesticides have a higher affinity for such sediments (Di Toro 1991). Composite samples of the sediments were placed in 4-L plastic bags and kept cold during transport to the laboratory, where the sediments were homogenized by hand-mixing and the large debris, such as gravel and native organisms, were removed.

The sediment samples were stored at 4 °C for no longer than 2 weeks before toxicity testing. The dry weight was measured by heating at 105 °C to a constant value and the organic-matter content determined by calcination at 550 °C by weight loss on ignition (APHA 1998). Grain-size analysis was performed by passage through a set of standardized sieves larger than 63 µm to separate the sand particles, followed by the technique of settling velocity (Day 1965). The grain size of the fraction smaller than 63 µm was determined by a standard pipette technique (Folk 1954).

Ecotoxicological Bioassays

To evaluate biologic effects of the water and sediments a standardized battery of toxicity tests was selected to assess the relative toxicity of the water and sediment samples for the aquatic environment.

Toxicity testing of the water column

With this environmental matrix, the following bioassays were used to evaluate potential toxicology:

Acute-toxicity testing with the cladoceran *Daphnia magna* A static test under controlled conditions involving a 48-h exposure was used to evaluate organism mortality (USEPA 2002). Each bioassay was performed with 40 individuals per treatment (4 replicates of 10 neonates of age less than 24 h each). Species of the genus *Daphnia* (and in particular the species *D. magna*) are the most widely used as diagnostic organisms for toxicity tests in the world (USEPA 2021). The acute sensitivity of this species to various pollutants of anthropic origin such as insecticides, and the existence of standardized protocols that enable test reproducibility, give this bioassay particular value since the results obtained can be compared with those reported in other regions of the world. Although the species *D. magna* is not present in the water bodies studied here, other species of this same genus are abundant (Adamowicz et al. 2004).

Acute-toxicity testing with the amphipod *Hyaella curvispina* Water-toxicity tests were carried out following the USEPA standardized protocol involving a 96-h exposure (USEPA 2000). Three replicate test chambers (300-mL beakers containing 200 ml of test water and 10 individuals each) were used for each test, and the number of dead amphipods was registered at the end of the exposure. *Hyaella curvispina* is a native freshwater amphipod that lives in association with aquatic macrophytes. The feeding habits of this species play a fundamental role in food webs by facilitating the flow of energy in aquatic ecosystems through the direct conversion of detritus into small particles of organic matter and as a food source for other animals (Casset et al. 2001; Saigo et al. 2009). Consequently, the effects detected in this species can accordingly provide relevant information about the impact on local ecosystems associated with a given study area.

Acute-toxicity testing with *Lactuca sativa* seeds A standardized static test of exposure for 120 h was performed under controlled laboratory conditions before evaluating the percentage of seed germination (lethal effect) and root and hypocotyl length (sublethal effects), according to the United States Environmental Protection Agency [USEPA] (USEPA 1996). For each sample, 4 replicates of 20 seeds each were used. This toxicity bioassay has been recommended and applied by different environmental-protection agencies for the ecotoxicological evaluation of environmental samples and pure compounds, in addition to the evaluation of the phytotoxic effect of pesticides on nontarget species necessary for the registration of such agents (Boutin et al. 1993).

Sediment-toxicity test

The toxicity of the sediments was evaluated with *H. curvispina* as the diagnostic species. Whole-sediment tests were conducted for 10 days (USEPA 2000) according to the

standardized protocol with modifications (Peluso et al. 2011). The procedure stated in brief: After a 24-h stabilization period, five biological replicates were used for each sediment sample; in each replicate were placed 100 mL of sediment and 175 mL of overlying water along with ten individuals. The endpoints measured were survival (lethal effect) and growth (body length in millimeters—sublethal effect). The performance criterion for the reference sediment was an 80% survival. Each replicate having an amphipod survival higher than 50% was used for growth analysis (i.e., the sublethal test; Peluso et al. 2013).

General considerations

The organisms of the different species used in the battery of bioassay were obtained from cultures kept in the laboratory under controlled conditions. Lettuce seeds were obtained from a selected batch. In all the bioassays, the laboratory conditions (e.g., photoperiod, temperature) were monitored and kept constant. Moreover, according to the corresponding protocols, the requirements of quality control and quality assurance (control charts) along with the criteria for the acceptability of the experimental results—i.e., an effect <10% (*D. magna* and *H. curvispina*) along with a coefficient of variation <30% (*L. sativa* test) in the negative controls—were rigorously followed.

Statistical Analysis

The percentages of the biologic effects assessed (% mortality, % root and hypocotyl elongation, % growth inhibition) were adjusted with respect to the control values by the Abbott correction (Abbott 1925). With an aim at stabilizing the variances, before an analysis of variance (ANOVA), the arc sine of the angular transformation of the percentages, was applied. Subsequently, in each instance, we established the bioassays with significant endpoints with respect to negative controls (Zar 1998). In order to describe the proportion of positive tests (significant bioassay endpoints relative to the control) in each subgroup in terms of the explanatory variables we used a generalized linear model involving a binomial response and a logit-link function that was generated after considering the binary-response variable (Y)—i.e., 1, significant; 0, not significant—according to the categorical variables: Study Area (Reference/Extensive/Intensive), Environmental Matrix (Water/Sediment), Bioassay (*Daphnia*/*Lactuca*/*Hyaella*), Endpoint (Lethal/Sublethal) and Campaign. The model can be summarized as follows (equation 1):

$$Y \sim \text{Binomial}(N, p)$$

$$\text{logit}(p) = \log\left(\frac{p}{1-p}\right) = \mathbf{X}\boldsymbol{\beta}$$

$\mathbf{X}$: model matrix with categorical variables $\boldsymbol{\beta}$: parameter vector

(1)

Table 1 General characterization of the water bodies associated with each area of study

Environmental matrix	Parameter ^a	Reference area	Cereal and oilseed production	Horticultural production
Water	pH	8.62 ± 0.22	7.62 ± 0.21	7.44 ± 0.26
	T (°C)	19.30 ± 0.47	18.54 ± 2.02	20.90 ± 2.04
	DO (mg/L)	5.74 ± 1.77	6.28 ± 2.13	4.96 ± 1.31
	EC (mS/cm)	0.50 ± 0.14	0.71 ± 0.46	0.99 ± 0.34
	TDS (mg/L)	134 ± 4	377 ± 148	465 ± 157
	Hardness (mg/L)	67 ± 1	151 ± 2	119 ± 15
	Alkalinity (mg/L)	393 ± 22	381 ± 73	717 ± 257
	OM (mg/L)	0.81 ± 0.35	0.49 ± 0.10	0.62 ± 0.36
Bottom sediments	LOI (%)	4.87 ± 0.67	6.71 ± 1.93	4.65 ± 1.03
	Fines (%)	88 ± 3	95 ± 5	55 ± 10

The values correspond to the mean ± the standard deviation of the measurements made in each stream during sampling campaigns

^aT temperature, DO dissolved-oxygen concentration, EC electrical conductivity, TDS total dissolved solids, OM organic matter, LOI weight loss on ignition, Fines <63 µm

For the categorical variables (F and *p* value), marginal-hypothesis tests (Wald) were carried out and then a selection of variables made to generate the final model according to the minimization criteria of the Akaike information criterion (AIC). The analyses were carried out with the function `glm{stats}` and `stepAIC{MASS}` (Venables and Ripley 2002) in the R-software environment version 4.02 (R Core Team 2020).

To depict the results graphically, a multiple correspondence analysis was performed. The multivariate observations of the categorical variables were projected in a biplot built from the axes with greater inertia to visualize the most significant associations between the different kinds of categorical variables (Greenacre and Hastie 1987).

Results

General Characterization of the Areas of Study

Although none of the areas studied suffered substantial changes in land use; in the areas surrounding the sampling points during the sampling campaigns, areas 1 (cereal and/or oilseed production) and 3 (reference sites) remained much more stable than Area 2, where during this period the structure of the horticultural production was increasingly transformed by the use of greenhouse technology (Ferraris and Ferrero 2018). The production modality under cover has led to the confinement of the cultivated area and the continuous succession of crops throughout the year. In addition, liquid effluents from greenhouses reach surface water bodies on a more punctual basis through small canals. With respect to the water bodies monitored, no significant temporal or spatial variations were observed in the general characteristics of the water and the sediment measured

between those two forms of land use. Similarly, the surface water bodies of the different study areas evidenced similar characteristics (Table 1) with the exception of the total dissolved solids and water hardness, which parameters were somewhat lower in the streams of the reference sites, and the alkalinity, with values on the average double those of the streams associated with the extensive agricultural production (Table 1).

Ecotoxicologic Impact

The results of the bioassays involved two categories: (a) the quantitative assessment of the intensity of the biologic effect as indicated by the response of each one of the diagnostic species as well as the ecotoxicologic quality of the site; and (b) the frequency of ecotoxicological endpoints that proved to be significant with respect to the bioassay controls. Since different species manifested differential biologic responses to different compounds—and even more so when comparing phylogenetically more distantly related species—along with the number of events in which the biota are affected, the frequency of these endpoints can be reliably interpreted as an indicator of the diversity of stressors present at a given study site, but only if that site does not contain pollutants at high enough concentrations to become generally toxic to all organisms used in the battery.

Magnitude of effects

Figure 2 illustrates the differential effects (expressed as percentages of the biologic effect evaluated in relation to the negative control of the bioassay) produced in the water and sediment samples from the surface water bodies associated with each one of the productive systems studied and with the reference sites. As to the lethal effects within the water

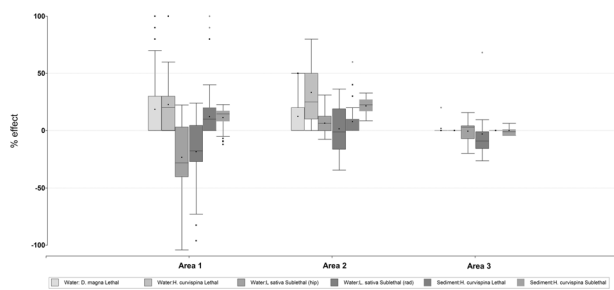


Fig. 2 Percentage of effects with respect to the negative controls for each endpoint. In the box plots, the upper and lower borders represent the quartiles of the data, the solid line the median, and the upper and lower outliers (the whiskers) the respective maximum and minimum values

column, no significant differences among the samples from the different areas studied were observed for any of the species (*D. magna*: $F = 1.414$, $p = 0.249$; *H. curvispina*: $F = 2.149$, $p = 0.148$; *L. sativa*: $F = 2.808$, $p = 0.066$). With respect to the sublethal effects on *L. sativa*, however, significant differences were observed between the samples coming from Area 1 and Area 2. Nevertheless, in neither of the agroecosystems studied was the root elongation statistically different from that produced in the samples from reference site, but significant differences were observed between Area 1 and Area 2 ($F = 5.374$, $p = 0.007$). Only the horticultural production (Area 2) induced significant effects on hypocotyl elongation with respect to the reference area ($F = 20.744$, $p < 0.0001$; Fig. 2). Note that since the sublethal effects measured in *L. sativa* were defined as elongation inhibition, the negative values in the figure refer to a greater elongation than recorded in the bioassay control.

The sediment samples did not evidence significant differences among the areas studied when the lethal effects were assessed on *H. curvispina* ($F = 1.038$, $p = 0.311$). Nevertheless, significant differences were observed when sublethal effects were considered. In this instance, the sediments from the streams associated with Area 1 (cereal and oilseed production) were more toxic than those of the horticultural land use, and both of these more toxic than those of the reference site ($F = 28.663$, $p < 0.0001$).

Thus, in general terms, the intensive productive horticultural systems produced greater effects than the cereal and oilseed ones on the water column, whereas this relationship was the reverse for the bottom sediments.

Frequency of effects

In general, if the totality of water and sediment samples evaluated in each study area and all the endpoints analyzed for each bioassay are taken into account (Table 2), the percentages of endpoints that were significant with respect to the bioassay controls were higher in the samples

corresponding to the sites associated with agricultural production (Area 1 and Area 2) than in those of the reference sites, where in the latter sublethal effects were recorded with only *L. sativa*—and there in fewer than 30% of the bioassays. In the sediments whose toxicity was evaluated with *H. curvispina* as the diagnostic organism, the sublethal effects clearly pointed to a greater response sensitivity than the lethal ones; whereas, for the water, regardless of the bioassay considered, no definite pattern of greater sensitivity between the lethal and sublethal effects was observed.

As to the categorical variables analyzed, no significant differences among the sampling campaigns ($F = 0.03$; $p = 0.86$) or between water and sediment ($F = 1.38$, $p = 0.24$), were observed. Nevertheless, in the final model, significant differences were obtained with respect to the area of study ($F = 9.55$; $p < 0.0001$), the diagnostic species ($F = 3.17$; $p < 0.0431$), and the endpoint analyzed ($F = 28.7$; $p < 0.0001$); thus, manifesting significant differences in the frequency of endpoints differing significantly from the respective controls (Fig. 3). Accordingly, the samples from Area 2 associated with horticultural cultivation were found to be more frequently toxic than those from the area of cereal and oilseed production, with both those frequencies being higher than that of the reference site (Fig. 4).

Regarding the different species included in the bioassay, *L. sativa* proved to be more sensitive to the type of contaminants associated with the intensive horticultural land use, whereas no such differences in sensitivity were observed with the two crustaceans. Moreover, with respect to the type of endpoint evaluated, we observed that the sublethal effects were significantly more sensitive—and thus more informative—than the lethal ones (Table 2).

Discussion

The advance of mankind in the occupation of territories for human development and use, especially in sectors with high productive potential, has led to the displacement and disappearance of natural ecosystems from those areas (Piquer-Rodríguez et al. 2018). Thus, when evaluating the impact of an anthropic activity on the environment, the absence of regions for use as reference that are without such an influence but otherwise manifest similar characteristics to the study sites is one of the most difficult obstacles to be surmounted by researchers. In the work reported here, we selected a reference area within a sector of land declared and maintained as a natural reserve where the anthropic influence is significantly lower than the areas under study with the existing few activities having a different productive profile than the latter (i.e., animal grazing in low densities).

The measurements in situ of certain general parameters of water quality enable an assessment of the general state of

Table 2 Percentage of endpoints that indicated significant differences from the controls with respect to each study area, environmental matrix, bioassay, and type of effect (*L* Lethal, *SL* Sublethal)

Study area	Enviromental matrix	Bioassay	Effect	<i>n</i> ^a	% Positive
Area 1	Water	<i>D. magna</i>	L	27	22
		<i>H. curvispina</i>	L	27	33
		<i>L. sativa</i>	SL	54	24
	Sediment	<i>H. curvispina</i>	L	35	23
		<i>H. curvispina</i>	SL	31	68
		<i>D. magna</i>	L	54	39
Area 2	Water	<i>H. curvispina</i>	L	30	30
		<i>L. sativa</i>	SL	96	57
		<i>H. curvispina</i>	L	62	24
	Sediment	<i>H. curvispina</i>	SL	44	61
		<i>D. magna</i>	L	11	0
		<i>H. curvispina</i>	L	6	0
Area 3	Water	<i>L. sativa</i>	SL	22	27
		<i>H. curvispina</i>	L	4	0
		<i>H. curvispina</i>	SL	4	0
	Sediment	<i>H. curvispina</i>	L	4	0
		<i>H. curvispina</i>	SL	4	0
		<i>H. curvispina</i>	SL	4	0

Area 1 correspond to cereal and oilseed production; Area 2 correspond to horticultural production, while Area 3 was the reference area of minimum anthropic impact

^aNumber of endpoints for each bioassay for the totality of the sites. Note that when significant lethal effects were detected, the sublethal effects were not considered

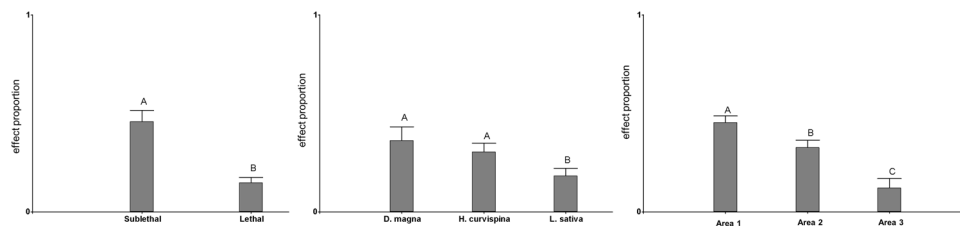


Fig. 3 Visualization of the relationships among the categorical variables studied. In each panel, the effect proportion (adjusted means) is plotted on the *ordinate* for the lethal and sublethal effects (Left panel), the assay species (Central Panel), and the three study areas (Right Panel) as indicated on the *abscissa*. Only the variables that manifested

significant differences are plotted (Fisher's least significant difference; $\alpha = 0.05$) by the *p* value—correction procedure (Benjamini and Hochberg 1995). Different letters above the boxes indicate significant differences ($p < 0.05$)

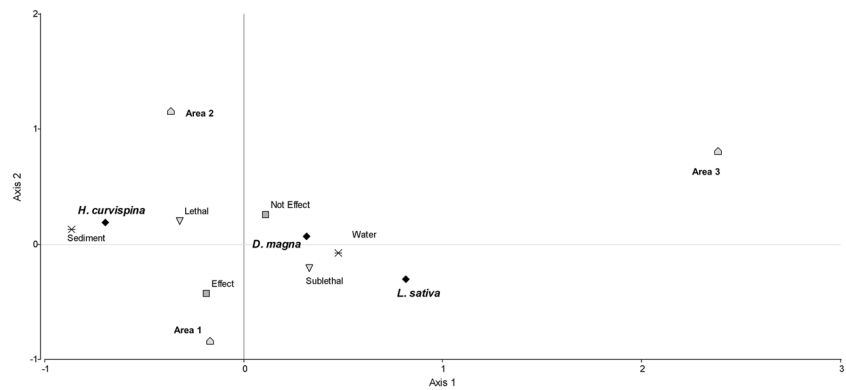
a water body under study. The electrical conductivity and total dissolved solids are among the parameters that become the most modified by the input of pollutants from urbanization-related processes. Kaushal et al. (2005), for example, studying surface water bodies associated with urban and/or industrial land use, found that rivers under those anthropic influences had salinity values (directly related to electrical conductivity) several times higher than those found in water bodies under the influence of agriculture or forestry. The general parameters measured in this study (Table 1) exhibited values comparable to those recorded by other authors in streams belonging to areas associated with similar land uses (Ronco et al. 2008; Solis et al. 2017; 2019; Mac Loughlin et al. 2020), and those values were far below the corresponding data cited for highly polluted water bodies that run through urban areas (Rimoldi et al. 2018; Islam et al. 2015). Therefore, on the

basis of the parameters evaluated and the results of the visual survey of the area, the streams studied in this work did not suggest an urban-industrial influence.

The overall impacts of agriculture on the environment have been well documented, with pesticide inputs being one of the main contributors to the deterioration of natural ecosystems (Davari et al. 2010; Ponge et al. 2013; Annett et al. 2014).

The production of cereals and oilseed involves the use of a substantial amount of pesticides. In the example of the chemical fallow used in direct sowing, all those forms of cultivation involve similar profiles and amounts of phytosanitary products; but, during the postseeding period, soybeans generally require a higher number and greater quantity of pesticides. Indeed, nearly 20 different types of herbicides, 15 different formulations of fungicides, and a similar number of insecticides are usually used in soybean

Fig. 4 Biplot of the multiple correspondence analysis built from the axes with greater inertia



production in Argentina (Pérez et al. 2017). In contrast, the great variety of species cultivated in horticulture is associated with the use of a wide diversity and quantity of pesticides (DP 2015). Especially, the use of fungicides and insecticides has increased in the last 15 years as a result of the growth in the production of vegetables in greenhouses (Baldini and Mendizabal 2019). For example, for merely tomato cultivation, more than 70 different fungicide, insecticide, and herbicide compounds are currently on the market (DP 2015).

The chemical-control practices associated with each productive area are reflected in the profile of pesticides found in the associated water bodies (Supplementary Material). According to the literature, in surface water bodies associated with cereal and oilseed production, herbicides and fungicides are the main pesticides found in the water column, whereas insecticides and glyphosate predominate in the sediments, which difference may be due to the physical-chemical characteristics of the latter compounds, with their greater affinity for particulate material causing them to end up in those bottom sediments (Mac Loughlin et al. 2020). In general, data on the environmental concentrations of pesticides in areas associated with intensive horticulture are scarce. In addition, the sediments of streams associated with intensive horticulture contain certain herbicides—such as glyphosate, trifluralin, atrazine, and acetochlor—at higher concentrations than those reported for sites associated with the predominant production systems in Area 1; whereas the insecticides apparently exhibit a similar profile and are at comparable concentrations in both productive systems, except with respect to certain extreme values registered in the intensive horticultural-production sectors (Mac Loughlin et al. 2017). Among the paucity of information found related to the concentration of pesticides in the water columns of streams under the influence of horticulture, the high concentrations of glyphosate and aminomethylphosphonic acid (the main degradation metabolite of glyphosate)—which exceed by up to two orders of magnitude those observed in water associated with cereal and oilseed production—are

especially noteworthy (Bonansea et al. 2017; Mac Loughlin et al. 2020).

Ecotoxicologically, the surface water bodies associated with the reference area (Area 3) are clearly of a better quality than those located in agricultural areas (Areas 1 and 2) (Table 2, Fig. 2), thus confirming that the former area was appropriate for use as a reference environment and that both contrasting models of agricultural production impacted negatively on the biota. Even though Area 3 was associated with cattle grazing, agriculture seems to have a stronger negative impact than that type of land use, although in this instance we need to clarify that the animal load in the reference area was notably lower than that existing in an area under intensive cattle raising. Solis et al. (2017)—studying the abundance and composition of the macroinvertebrate assemblages in different streams associated with livestock, agriculture, and the same biosphere reserve used by us as a reference site—found that the composition of the invertebrate assemblages of the streams through cultivated areas differed significantly from the others, with the abundance and richness of invertebrates being higher in streams of the reserve than in either of the other two.

Furthermore, the water samples from streams associated with horticulture induced a higher percentage of toxic effects than those of the sites under cereal and oilseed production (Table 2), which difference could be related to the presence of higher concentrations of pesticides found in the former sector. This conclusion is further strengthened if we consider that for *L. sativa* the water samples from the horticultural-production zone were more toxic than those from Area 1 (Table 2, Fig. 2), since this species exhibits a greater intrinsic sensitivity to herbicides such as the glyphosate that had been found in higher concentrations in this area than do either *D. magna* or *H. curvispina* (Miller et al. 1985). In general, the effects detected on *L. sativa* corresponded to sublethal alterations associated with the increase in radicle and hypocotyl growth. This type of effect, known as hormesis—i.e., a biphasic dose-response profile involving a transient stimulation followed by an inhibition—has been widely registered in ecotoxicological studies as a

response of organisms to exposure to low doses of contaminants (Calabrese and Baldwin 2002; Iavicoli et al. 2018). Note that the growth stimulation should not be interpreted as a favorable effect, but as a compensatory response to exposure to a pollutant. In the example of vascular plants, several studies have reported a growth stimulation associated with exposure to low doses of herbicides (Belz et al. 2011; Cedergreen 2008), which findings lead us to surmise that the response observed in this study could be associated with that type of pesticide. Although *L. sativa* is not a representative species of aquatic ecosystems, the information generated from this toxicity test provides data about the possible effect of pollutants on plant communities near the margins of contaminated water bodies, with *L. sativa* being also an informative species to consider due to the horticultural relevance (Sobrero and Ronco 2004; Priac et al. 2017). Thus, the sublethal effects associated with radicle and hypocotyl elongation are highly sensitive indicators for the evaluation of biologic effects on plants, providing information complementary to that provided by studying the effect on germination and indirectly related to the ability of future seedlings in the riparian vegetation associated with these water bodies to become established. *D. magna* and *H. curvispina* showed similar lethal responses to exposure to water samples. This similarity in the response between both species in environmental samples could be attributed to similar metabolic pathways, since the close phylogenetic relationship between both species (Maltby et al. 2005). As our study, Cooman et al. (2005) used *D. magna* and *D. pulex* to evaluate water quality in a watershed in Chile, detecting toxicity associated with agricultural land use mainly when rainfall is high. Also, these authors observed toxic effects on diagnostic organisms even when the pesticide concentrations detected were below known toxicity levels, attributing to additive and synergistic effects of the presence of mixture of pesticides as a possible cause of the observed toxicity, and reevaluating the use of the use of these tools to study surface water quality associated with agricultural land uses.

Conversely, with the sediments we did not observe large differences in the percentages of significant effects induced by the samples from the two areas (Table 2). The sublethal effects on *H. curvispina*, however, were of greater magnitude with the samples from the extensive-production area (Fig. 2), which finding could be related to the high sensitivity of this species to insecticides (especially pyrethroids) that are found at high frequencies and concentrations in those sectors (Hunt et al. 2016; Pérez et al. 2021). The amphipod *H. curvispina* has been used as a sentinel species owing to its wide distribution in the pampas region and its high sensitivity to pesticides (Mugni et al. 2012; Mac Loughlin et al. 2017). According to Solis et al. (2017) and Arias et al. (2020) this species is present in all the

ecosystems studied, but its abundance is much higher in the streams located in the reference area (2408 ind/m²), than in the streams affected by agricultural activity (21 ind/m²); thus suggesting—as found in this work—that agrochemicals are harmful to biota.

Conclusions

To the best of our knowledge the work reported here marks the first investigation comparing the impacts of two contrasting models of agricultural production on the ecotoxicologic quality of the surface water bodies associated with those respective practices, where the conclusions from this study ought to constitute a fundamental feedback for decision-making regarding the environmental management of those territories.

According to our results, both productive systems impacted the associated surface water bodies negatively, with horticulture being the one that seemed to produce a greater frequency of ecotoxicologic effects on the species studied in both the water and the sediment. The sediments associated with cereal and oilseed production, however, evidenced a greater magnitude of sublethal effects.

Taking into account that the sensitivity of the bioassay battery selected in this work as ecotoxicologic-impact-assessment strategy allows detect differences between the impacts of each activity, it appears to be a useful tool. Thus, the use of this battery of ecotoxicological bioassays appears to be an effective approach to be adopted by the organizations in charge of water-resource management.

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Compliance with Ethical Standards

Conflict of Interest The authors declare no competing interests.

Ethical Approval No approval of research ethics committees was required to accomplish the goals of this study because experimental work was conducted with an unregulated invertebrate species.

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References

- Abbott WS (1925) A method of computing the effectiveness of an insecticide. *J Econ Entomol* 18:265–267
- Adamowicz SJ, Hebert PDN, Marinone MC (2004) Species diversity and endemism in the *Daphnia* of Argentina: a genetic investigation. *Zool J Linn Soc* 140(2):171–205. <https://doi.org/10.1111/j.1096-3642.2003.00089.x>
- Anderson BS, Phillips BM, Voorhees JP, Petersen MA, Jennings LL, Fojut TL, Vasquez M, Siegler C, Tjeerdema RS (2015) Relative toxicity of bifenthrin to *Hyalella azteca* in 10 day versus 28 day exposures. *Integr Environ Assess Manag* 11:319–328
- Annett R, Habibi HR, Hontela A (2014) Impact of glyphosate and glyphosate-based herbicides on the freshwater environment. *J Appl Toxicol* 34:458–479. <https://doi.org/10.1002/jat.2997>
- APHA, American Public Health Association, AWWA, WEF (1998) Standard Methods for Examination of Water and Wastewater. American Public Health Association, Washington, DC
- Arias M, Scalise A, Solis M, Paracampo A, Indaco M, Fanelli S, Mugni H, Bonetto C (2020) Horticulture affects macro-invertebrate assemblages in adjacent streams (Buenos Aires, Argentina). *Knowl Manag Aquat Ecosyst* 421:5. <https://doi.org/10.1051/kmae/2019048>
- Baldini C, Mendizábal A (2019) Entre los commodities, el agronegocio y una población que demanda avanzar hacia la agroecología: pensar las políticas públicas agroecológicas en Argentina a partir de la reflexión sobre experiencias en Francia. *Revista Cardinalis*, Año 7, N° 13, 2do. Semestre 2019. pp. 82–116. <https://revistas.unc.edu.ar/index.php/cardi/issue/current>
- Barreto E, Costa S, Demetrio P, Lascano C, Venturino A, Natale GS (2020) Sensitivity of *Boana pulchella* (Anura: Hylidae) Tadpoles to Environmentally Relevant Concentrations of Chlorpyrifos: Effects at the Individual and Biochemical Levels. *Environ Toxicol Chem* 39:834–841. <https://doi.org/10.1002/etc.4664>
- Belz RG, Cedergreen N, Duke SO (2011) Herbicide hormesis - can it be useful in crop production? *Weed Res* 51:321–332. <https://doi.org/10.1111/j.1365-3180.2011.00862.x>
- Benjamini Y, Hochberg Y (1995) Controlling the false discovery rate: a practical and powerful approach to multiple testing. *J R Stat Soc Ser B* 57:289–300
- Blandi ML, Sioux Cavalcante M, Gargoloff NA, Sarandón SJ (2016) Prácticas, conocimientos y percepciones que dificultan la conservación de la agrobiodiversidad. El caso del Cinturón Hortícola Platense, Argentina. *Cuad de Desarro Rural* 13 (78):97–122
- Bonansea RI, Filippi I, Wunderlin DA, Marino DJG, Amé MV (2017) The fate of glyphosate and AMPA in a freshwater endorheic basin: an ecotoxicological risk assessment. *Toxics* 6:1–13. <https://doi.org/10.3390/toxics6010003>
- Boutin C, Freemark K, Keddy C (1993) Proposed guidelines for registration of chemical pesticides: Nontarget plant testing and evaluation, Technical Report Series NO. 145. Canadian Wildlife Service (Headquarters), Environment Canada, Ottawa
- Calabrese EJ, Baldwin L (2002) Applications of hormesis in toxicology, risk assessment and chemotherapeutics. *Trends Pharmacol Sci* 23:331–337. [https://doi.org/10.1016/S0165-6147\(02\)00234-5](https://doi.org/10.1016/S0165-6147(02)00234-5)
- Casset MA, Momo FR, Giorgi ADN (2001) Dinámica poblacional de dos especies de anfípodos y su relación con la vegetación acuática en un microambiente de la cuenca del río Luján (Argentina). *Ecolía Austral* 11:79–85
- Cedergreen N (2008) Herbicides can stimulate plant growth. *Weed Res* 48:429–438. <https://doi.org/10.1111/j.1365-3180.2008.00646.x>
- Colamarino I, Curcio N, Ocampo F, Torrandel C (2006) La producción hortícola en la Argentina. Dirección Nacional de Alimentos y Bebidas. Ministerio de Agricultura, Ganadería y Pesca. Presidencia de la Nación. Buenos Aires, Argentina. http://www.alimentosargentinos.gob.ar/contenido/revista/ediciones/33/articulos/mesa_todos.htm
- Cooman K, Debels P, Gajardo M, Urrutia R, Barra R (2005) Use of *Daphnia* spp. for the ecotoxicological assessment of water quality in an agricultural watershed in South-Central Chile. *Arch Environ Contam Toxicol* 200:191–200. <https://doi.org/10.1007/s00244-004-0218-6>
- Davari M, Tewari JC, Kaushish S (2010) Impact of agricultural practice on ecosystem services. *Int J Agron Plant Prod* 1:11–23
- Day P (1965) Particle fractionation and particle size analysis methods of soil analysis. In: Black C (Ed.) *Methods of Soil Analysis*, Part I. American Society of Agronomy, Wisconsin, p 545–566
- Ding YU, Harwood AMD, Foslund HEM, Lydy MJ (2010) Distribution and toxicity of sediment-associated pesticides in urban and agricultural waterways from Illinois, USA. *Environ Toxicol Chem* 29:149–157. <https://doi.org/10.1002/etc.13>
- DP (2015) Defensor del Pueblo de la Provincia de Buenos Aires: Relevamiento de la utilización de agroquímicos en la Provincia de Buenos Aires. Argentina: Mapa de situación e incidencia sobre la salud, Buenos Aires, <http://sedici.unlp.edu.ar/handle/10915/54451>
- Ferraris G, Ferrero GE (2018) Análisis de la estructura agraria en los sistemas hortícolas del AMBA- SUR (Área Metropolitana de Buenos Aires-Sur). *Rev de La Facultad de Agronomía-UNLP* 117:231–244
- Folk RL (1954) The distinction between grain size and mineral composition in sedimentary rock nomenclature. *J Geol* 62:344–359
- Food and Agriculture Organization (FAO) (2004) Socio-economic Analysis and Policy Implications of the Roles of Agriculture in Developing Countries. Summary Report, Roles of Agriculture Project, FAO, Rome, Italy
- Greenacre MJ, Hastie T (1987) The geometric interpretation of correspondence analysis. *J Am Stat Assoc* 82:437–447
- Hunt L, Bonetto C, Resh VH, Forsin D, Fanelli S, Marrochi N, Lydy MJ (2016) Insecticide concentrations in stream sediments of soy production regions of South America. *Sci Total Environ* 547:114–124. <https://doi.org/10.1016/j.scitotenv.2015.12.140>
- Iavicoli I, Leso V, Fontana L, Calabrese EJ (2018) Nanoparticle exposure and hormetic dose-responses: An update. *Int J Mol Sci* 19:3. <https://doi.org/10.3390/ijms19030805>
- Islam MS, Ahmed MK, Raknuzzaman M, Habibullah-AI-Mamun M, Islam MK (2015) Heavy metal pollution in surface water and sediment: a preliminary assessment of an urban river in a developing country. *Ecol Indic* 48:282–291
- Kaushal SS, Groffman PM, Likens GE, Belt KT, Stack WP, Kelly VR, Band LE, Fisher GT (2005) Increased salinization of fresh water in the northeastern United States. *Proc Natl Acad Sci USA* 102:13517–13520
- Mac Loughlin TM, Peluso L, Marino DLG (2017) Pesticide impact study in the peri-urban horticultural area of Gran La Plata, Argentina. *Sci Total Environ* 598:572–580. <https://doi.org/10.1016/j.scitotenv.2017.04.116>
- Mac Loughlin TM, Peluso ML, Aparicio VC, Marino DJG (2020) Contribution of soluble and particulate-matter fractions to the total glyphosate and AMPA load in water bodies associated with horticulture. *Sci Total Environ* 703: <https://doi.org/10.1016/j.scitotenv.2019.134717>
- MAGyP, Ministerio de Agricultura Ganadería y Pesca, Argentina (2020) <https://www.magyp.gob.ar/sitio/areas/estimaciones/monitor/>

- Maltby L, Blake N, Brock TC, van den Brink PJ (2005) Insecticide species sensitivity distributions: importance of test species selection and relevance to aquatic ecosystems. *Environ Toxicol Chem* 24(2):379–88. <https://doi.org/10.1897/04-025r.1>
- Miller W, Peterson SA, Greene JC, Callahan CA (1985) Comparative Toxicology of Laboratory Organisms for Assessing Hazardous Waste Sites Response of Bioassays to Chemical Subgroups The EC ~ 0 response values (LC₅₀ for earthworms). *J Environ Qual* 14:569–574
- Mugni H, Demetrio P, Paracampo A, Pardi M, Bulus G, Bonetto C (2012) Toxicity persistence in runoff water and soil in experimental soybean plots following chlorpyrifos application. *Bull Environ Contam Toxicol* 89:208–212
- Newman MC (2015) Fundamentals of ecotoxicology. The Science of Pollution, FOURTH EDITION. CRC Press Taylor & Francis Group 6000 Broken Sound, Parkway NW, Suite 300 Boca Raton, FL 33487-2742, p 708
- Peluso L, Bulus Rossini G, Salibián A, Ronco AE (2013) Physico-chemical and ecotoxicological based assessment of bottom sediments from the Luján River basin, Buenos Aires, Argentina. *Environ Monit Assess* 185:5993–6002
- Peluso L, Giusto A, Bulus Rossini GD, Ferrari L, Salibián A, Ronco AE (2011) *Hyalella curvispina* (Amphipoda) as a test organism in laboratory toxicity testing of environmental samples. *Fresenius Environ Bull* 20:372–37
- Pérez DJ, Iturburu FG, Calderon G, Oyesqui LAE, De Gerónimo E, Aparicio VC (2021) Ecological risk assessment of current-use pesticides and biocides in soils, sediments and surface water of a mixed land-use basin of the Pampas region, Argentina. *Chemosphere* 263. <https://doi.org/10.1016/j.chemosphere.2020.128061>
- Pérez DJ, Okada E, De Gerónimo E, Menone ML, Aparicio VC, Costa JL (2017) Spatial and temporal trends and flow dynamics of glyphosate and other pesticides within an agricultural watershed in Argentina. *Environ Toxicol Chem* 36. <https://doi.org/10.1002/etc.3897>
- Piquer-Rodríguez M, Butsic V, Gärtner P, Macchi L, Baumann M, Gavier Pizarro G, Volante JN, Gasparri NI, Kuemmerle T (2018) Drivers of agricultural land-use change in the Argentine Pampas and Chaco regions. *Appl Geogr* 91:111–122. <https://doi.org/10.1016/j.apgeog.2018.01.004>
- Ponge J, Pérès G, Guernion M, Ruiz-Camacho N, Cortet J, Pernin C, Cluzeau D (2013) Soil Biology & Biochemistry The impact of agricultural practices on soil biota: a regional study. *Soil Biol Biochem* 67:271–284. <https://doi.org/10.1016/j.soilbio.2013.08.026>
- Popp J, Peto K, Nagy J (2013) Pesticide productivity and food security. A review. *Agron Sustain Dev* 33:243–255. <https://doi.org/10.1007/s13593-012-0105-x>
- Priac A, Badot P, Crini G (2017) Comptes Rendus Biologies Treated wastewater phytotoxicity assessment using *Lactuca sativa*: Focus on germination and root elongation test parameters. *Comptes Rendus Biol* 340:188–194. <https://doi.org/10.1016/j.crv.2017.01.002>
- R Core Team (2020) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. <https://www.R-project.org/>
- Rimoldi F, Peluso L, Bulus Rossini G, Ronco AE, Demetrio PM (2018) Multidisciplinary approach to a study of water and bottom sediment quality of streams associated with mixed land uses: Case study Del Gato Stream, La Plata (Argentina). *Ecol Indic* 89:188–198. <https://doi.org/10.1016/j.ecolind.2018.01.063>
- Ronco AE, Carriquiriborde P, Natale GS, Martín ML, Mugni H, Bonetto C (2008) Integrated approach for the assessment of biotech soybean pesticides impact on low order stream ecosystems of the pampasic region. *Ecosystem Ecology Research Trends*. <http://www.scopus.com/inward/record.url?eid=2-s2.0-9953028745&partnerID=tZOtx3y1>
- Saigo M, Marchese M, Montalto L (2009) Hábitos Alimentarios de *Hyalella curvispina* Shoemaker, 1942 (Amphipoda: Gammaridea) en Ambientes Leníticos de la Llanura Aluvial del Río Paraná Medio. *Nat Neotropicalis* 1:43–59. <https://doi.org/10.14409/natura.v1i40.3876>
- Sansinena JA, Peluso L, Salgado Costa C, Demetrio PM, Mac Loughlin TM, Marino DJG, Alcalde L, Natale GS (2018) Evaluation of the toxicity of the sediments from an agroecosystem to two native species, *Hyalella curvispina* (CRUSTACEA: AMPHIPODA) and *Boana pulchella* (AMPHIBIA: ANURA), as potential environmental indicators. *Ecol Indic* 93:100–110. <https://doi.org/10.1016/j.ecolind.2018.04.061>
- Scanlon BR, Jolly I, Sophocleous M, Zhang L (2007) Global impacts of conversions from natural to agricultural ecosystems on water resources: Quantity versus quality. *Water Resour Res* 43. <https://doi.org/10.1029/2006WR005486>
- Secretaría de Comercio Interior (SCI) (2012) La producción de hortícolas en Argentina. Gerencia de calidad Tecnología
- Sobrero MC, Rimoldi F, Ronco AE (2007) Effects of the Glyphosate Active Ingredient and a Formulation on *Lemna gibba* L. at Different Exposure Levels and Assessment End-Points. *Bull Environ Contam Toxicol* 79:537–543. <https://doi.org/10.1007/s00128-007-9277-5>
- Sobrero MC, Ronco A (2004) Ensayo de toxicidad aguda con semillas de lechuga. In: Castillo G, Díaz MC, Pica Y, Ronco A, Sobrero C, Bulus G, Feota G, Forget G, Sánchez-Bain A (Eds.) Ensayos toxicológicos y Métodos de evaluación de calidad de aguas. Estandarización, intercalibración, resultados y aplicaciones. Centro Internacional de Investigaciones para el Desarrollo e Instituto Mexicano de Tecnología del Agua, México, p 71–79
- Solis M, Mugni H, Fanelli S, Bonetto C (2017) Effect of agrochemicals on macroinvertebrate assemblages in Pampasic streams, Buenos Aires, Argentina. *Environ Earth Sci* 76. <https://doi.org/10.1007/s12665-017-6476-1>
- Solis M, Paracampo A, Bonetto C, Mugni H (2019) Acute Toxicity of Chlorpyrifos to *Hyalella curvispina*: Comparison of Species Sensitivity and Assessment of Environmental Risk *Env Process* <https://doi.org/10.1007/s40710-019-00352-3>
- Stehle S, Schulz R (2015) Agricultural insecticides threaten surface waters at the global scale. *Proceedings of the National Academy of Sciences*, 112, 5750–5755. <https://doi.org/10.1073/pnas.1500232112>
- Stone WW, Gilliom RJ, Ryberg KR (2014) Pesticides in U.S. streams and rivers: Occurrence and trends during 1992–2011. *Environ Sci Tech* 48:11025–11030. <https://doi.org/10.1021/es5025367>
- Di Toro DM, Zarb SC, Hansen DJ, Swartz RC, Cowan CE, Pavlou SP, Allen HE, Thomas NA, Paquin PR (1991) Technical basis for establishing sediment quality criteria for nonionic organic chemicals using equilibrium partitioning. *Environ Toxicol Chem* 10:1541–1583
- USEPA United States Environmental Protection Agency (1996) Ecological Effects Test Guidelines (OPPTS 850.4200): Seed Germination/Root Elongation Toxicity Test
- USEPA United States Environmental Protection Agency (2000) Methods for measuring the toxicity and bioaccumulation of sediment-associated contaminants with freshwater invertebrates, 2nd edn. Office of Science and Technology. Office of Water. Report: EPA 600/R-99/064
- USEPA United States Environmental Protection Agency (2002) Methods for measuring the acute toxicity of effluents and receiving waters to freshwater and marine organisms, 5th edn. USEPA, Washington (EPA-821-R-02-012). Test Method 2000.0
- USEPA United States Environmental Protection Agency (2021) ECOTOX User Guide: ECOTOXicology Knowledgebase System. Version 5.3. Available: <http://www.epa.gov/ecotox/>

- Venables WN, Ripley BD (2002) *Modern Applied Statistics with S*. Springer, New York, 4th edn
- Venturino A, Montagna CM, de D'Angelo AMP (2007) Risk assessment of Magnacide H herbicide at Río Colorado irrigation channels (Argentina). Tier 3: studies on native species. *Environ Toxicol Chem* 26:177–182. <https://doi.org/10.1897/06-085r.1>
- Yorlano MF, Demetrio PM, Rimoldi F (2021) Riparian strips as attenuation zones for the toxicity of pesticides in agricultural surface runoff: Relative influence of herbaceous vegetation and terrain slope on toxicity attenuation of 2,4-D. *Sci. Tot. Environ.* <https://doi.org/10.1016/j.scitotenv.2021.150655>
- Zar JH (1998) *Biostatistical analysis*. Prentice Hall, New Jersey, 4° edición, 785 pp