



Occurrence of pesticides and their transformation products in headwater streams: Contamination status and effect of ponds on contaminant concentrations

François Le Cor^{a,b,*}, Sylvain Slaby^a, Vincent Dufour^a, Alain Iuretig^a, Cyril Feidt^a, Xavier Dauchy^b, Damien Banas^a

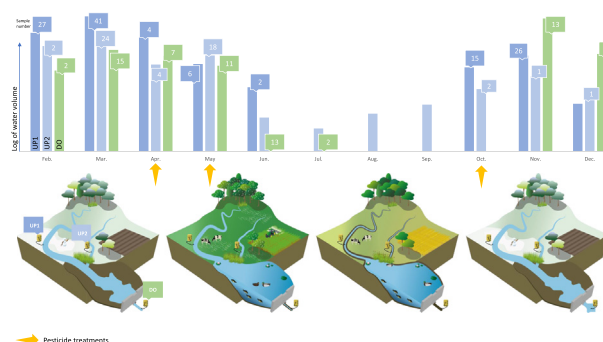
^a Université de Lorraine, INRAE, URAFFA, F-54000 Nancy, France

^b ANSES, Nancy Laboratory for Hydrology, Water Chemistry Department, 40 rue Lionnois, F-54000 Nancy, France

HIGHLIGHTS

- Streams are highly contaminated with pesticides in agricultural watersheds.
- Pesticide transformation products account for half of the contamination
- Ponds buffer pesticide contamination by reducing simultaneously detected compounds.
- Environmental and ecotoxicological thresholds are never exceeded downstream from ponds.

GRAPHICAL ABSTRACT



ARTICLE INFO

Article history:

Received 9 March 2021

Received in revised form 7 May 2021

Accepted 8 May 2021

Available online 13 May 2021

Editor: Jay Gan

Keywords:

Pesticides
Water quality
Agricultural watershed
Pond mitigation
Transformation products
Small waterbodies

ABSTRACT

In France, more than 90% of monitored watercourses are contaminated with pesticides. This high contamination level increases at the head of agricultural watersheds, where dilution capacities are low and transport from treated lands is direct. Ponds, numerous around headwater streams, could provide additional protection against pesticide pollution. Because of their long hydraulic residence time and large water volumes, they mitigate pesticide concentrations between upstream and downstream rivers. However, pesticide transformation products may also be responsible for the degradation of environments, owing to their presence at high concentrations and their persistence, but related data are scarce, particularly because of their high level of molecular diversity. We first reported on the state of water contamination in agricultural headwater streams, based on high frequency water sampling. Analysis of 67 molecules (HPLC-ESI-MS/MS) showed pesticides and pesticide transformation product mixtures of up to 29 different compounds in one sample. Regardless of the sampling location, transformation products represented at least 50% of the detected compounds. Then, we demonstrated the capacity of a pond to reduce contaminant concentrations in downstream rivers for 90% of the detected compounds. Upstream from this pond, environmental quality or ecotoxicological standards were exceeded during sampling, with pesticide and transformation product sum concentrations of up to 27 µg/L. Downstream from the study pond, few exceedances were observed, with a maximum total concentration of 2.2 µg/L, reflecting significant water quality improvement.

© 2021 Elsevier B.V. All rights reserved.

* Corresponding author at: Université de Lorraine, UR AFPA – ENSAIA, 2 avenue de la Forêt de Haye, F-54505 Vandœuvre-lès-Nancy cedex, France.
E-mail address: francois.le-cor@univ-lorraine.fr (F. Le Cor).

1. Introduction

Diffuse and acute pollution of water by pesticides brings together global health (Carvalho, 2017; Kim et al., 2017), ecological (Kaur and Garg, 2014; Osborne, 2012; van Lexmond et al., 2015), and economic concerns (Burgess and Pimentel, 2014; Pimentel, 2005). With the expansion of their use over the last 60 years, environmental issues have emerged on a large scale (Carson, 1962; Özkara et al., 2016). In Europe, the total quantity of pesticides used annually increased from 440,000 tons to more than 475,000 tons between 2000 and 2017 according to the Food and Agriculture Organization of the United Nations (FAO, 2020), questioning the overall state of ecosystems (Sharma et al., 2019). According to the same source, France (16% of all agricultural land in the European Union in 2016) has been ranked 6th in the world, over the last 10 years, among countries having the highest pesticide consumption, with more than 69,600 tons of active substances used for agriculture.

It has been estimated that depending on substances, agricultural equipment, and weather conditions before, during, and after treatment, various amounts of pesticides are likely to be transported to adjacent ecosystems (Dhananjayan et al., 2020; Mantzos et al., 2016a; Rousis et al., 2017; Sharma et al., 2019). Pesticides will spread through water resources in runoff induced by rainfall, resulting in rapid and widespread contamination events (Lefrancq et al., 2017). Headwater streams are particularly subject to this contamination, owing to their low dilution capacities and their proximity to agricultural land (Lorenz et al., 2017; Szöcs et al., 2017). Because of their position at the top of hydrographic networks, they are key to preserving the quality of downstream ecosystems (Dodds and Oakes, 2008; Rasmussen et al., 2013) as well as their rich biodiversity (Meyer et al., 2007). In this regard, several in situ structures can be implemented to limit their contamination by agricultural activities, such as constructed wetlands (Gaullier et al., 2019; Tournebise et al., 2012, 2013; Vallée et al., 2015; Vymazal and Březinová, 2015), vegetated ditches (Stehle et al., 2011), or riparian buffer strips (Bunzel et al., 2014; Chow et al., 2020).

Small waterbodies (i.e., 0.1 up to 100 ha) also seem to be relevant in the preservation of headwater streams. Ponds appear to have significant, but underestimated, pesticide mitigation capacities (Gaillard et al., 2015, 2016; Gregoire et al., 2009). Although small in size, these waterbodies, when added up together, cover an area three times larger than that covered by large natural and artificial lakes in France. On a European scale they cover nearly 270,000 km² (Bartout and Touchart, 2018). Usually, a high density of small waterbodies occurs at the head of agricultural watersheds (Drożdżyński, 2008; Lazartigues et al., 2012); they are, therefore, from the beginning of hydrographic networks, on the path of pesticide contamination. Moreover, they may also be particularly subject to transformation product (TP) transfers (Ulrich et al., 2018), making them even more relevant in the understanding of both levels of contamination (i.e., with parent compounds and TP). Field data regarding contamination of ponds with pesticides are scarce and, even though some do exist (Gaillard et al., 2016; Lazartigues et al., 2013; Ulrich et al., 2018), they rarely take into account TP. The collection of environmental data on pesticides and TPs seems, therefore, necessary to predict ecotoxicological effects that may occur.

Currently, waterbody status is mainly assessed against environmental quality standards (EQS) set by the Water Framework Directive (WFD, 2000/60/EC) for certain priority substances (e.g., EQS for metazachlor = 0.02 µg/L, EQS for MCPA = 0.5 µg/L). Ecotoxicological risks related to the occurrence and concentration of pesticides can also be assessed by comparing concentrations in waterbodies to toxicity thresholds (EC, LC50, or PNEC) established for various contaminants and model species (e.g., algae, invertebrates, fish). However, the diversity of active substances (466 potentially used in the European Union; Nagy et al., 2020) leads to gaps in the toxicity of certain substances and does not consider the effect of mixtures found in ecosystems. Consideration of this combination effect is necessary to assess the state of

natural environments in which multiple pesticides and TP are potentially present (Özkara et al., 2016). The current approach, based on the toxicity of individual compounds and not combinations of compounds, leads to an underestimation of the risk (Pereira et al., 2017).

Although ecotoxicological data on active substances are largely incomplete, 60% of the pesticides monitored in this work had at least one guidance value or toxicity threshold. As for TP, data are extremely scarce (i.e. 1%; Chemical Substances Portal, 2009). While issues relating to the latter have received increasing attention since the late 1990s (Belfroid et al., 1998; Melin et al., 2020), field data are still scarce (Fonseca et al., 2019; Rousis et al., 2017; Reemtsma et al., 2013). Given that TP, resulting from the transformation of parent compounds, are often more stable and therefore potentially more present in the environment (Phillips et al., 1999; Schuhmann et al., 2019), it seems relevant to accurately determine the extent of their environmental contamination (Gruchlik et al., 2018) especially in headwater streams. However, there is no official list of relevant TP to be monitored in the environment (Hernández et al., 2008; Melin et al., 2020). In addition, the scientific community knows little about the extent of their toxic potential, especially with long-term exposure. Therefore, in order to assess the extent of ecosystem contamination, it is necessary to gain such knowledge by analyzing as many substances (pesticides and TP) as possible (Fonseca et al., 2019) and over several growing seasons (Chow et al., 2020).

In this context, the present work had two main objectives. The first was to provide innovative data by assessing the numbers and concentrations of pesticides and several of their TP in small streams located at the head of a hydrographic web. The second was, on the one hand, to verify the reduction in pesticide concentrations downstream from a pond reported by Gaillard et al. (2016). On the other hand, we wanted to determine whether this reduction was counterbalanced by the production of TP and their release, downstream. Our work is a case study focusing on the high-resolution dynamics of a single pond over the long term, as an essential preliminary step before studying a wider range of cases at a larger scale.

2. Materials and methods

2.1. Study area, headwater stream, and pond characteristics

The study was carried out in North-Eastern France between February 8th and December 3rd, 2019 (Fig. 1). During this period, the average monthly rainfall was 70 mm with a maximum of 116.4 mm in October. Over the whole period, a total of 766 mm of rainfall was recorded. Twenty-three rainfall events (24-hours periods) greater than 8 mm were observed and the average temperature was approximately 12.3 °C ± 1.8 °C (Infoclimat, 2020).

The study streams were small first-order streams according to the Strahler classification (Strahler, 1952) receiving water from agricultural catchments. The study area was characterized by hydromorphic clay soils (Keuper marl). Streams were impeded by a dam pond built in the 15th century (Fig. 1). The study pond had a water surface area of 4.5 ha and supported extensive fish production (without any additional nutrient inputs). The average pond depth was 0.9 m, its maximum depth (near the dike) was 2.2 m, and its water residence time when the pond is at its fullest was 96 days (Gaillard et al., 2015). Regarding physicochemical characteristics, the pond water had a neutral to slightly alkaline pH. The study began with the pond dike gate closure and the pond being emptied, a few months before pesticides application in the agricultural catchment. The pond was then filled up with water from two first-order streams (i.e., tributaries) over the first two months of the study period (i.e., February and March). Next, it was stocked with selected fish species (i.e., *Cyprinus carpio*, *Tinca tinca*, *Rutilus rutilus*, and *Scardinius erythrophthalmus*) in late March. Finally, it was entirely drained in November in order to collect fish with fishnets. The whole water volume was discharged to a downstream stream.

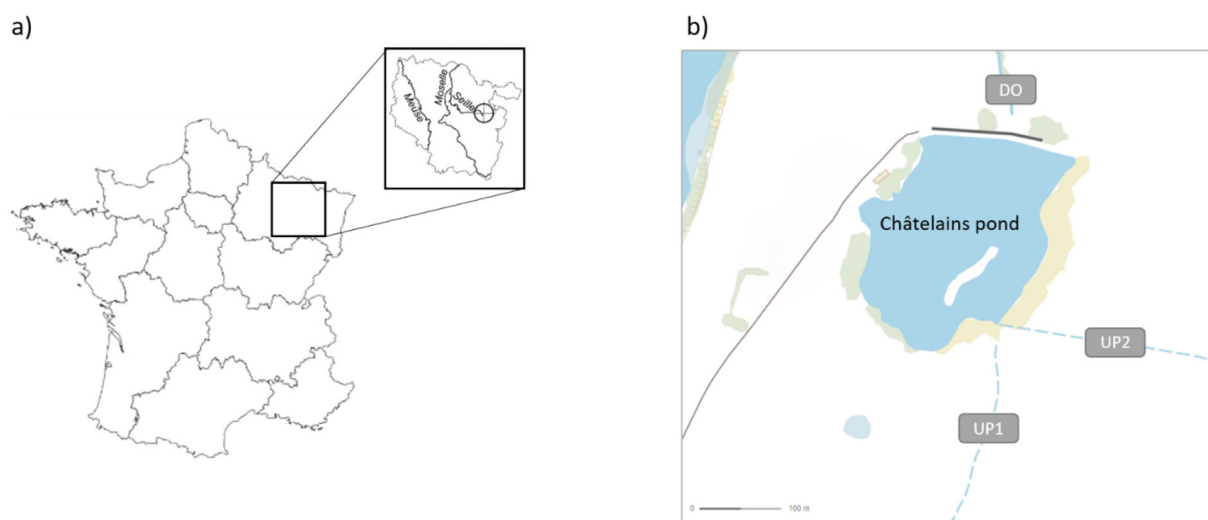


Fig. 1. a) The study pond is located in the Lorraine Region, upstream from the Seille River. b) Two sampling stations were located upstream from the pond (i.e., UP1 and UP2) and a single station was located downstream from the pond (i.e., DO).

The pond was supplied with water for the most part from two first-order streams (i.e., UP1 and UP2 representing 80% and 20% of its major water inputs, respectively). Pond water was discharged via a single outlet stream (i.e., DO) located 15 m downstream and mainly supplied with water from the pond. Even though dam ponds are no longer recommended by water agencies and the WFD because of the ecological discontinuity they cause, such a configuration is highly frequent among fish farming ponds that still represent historical production tools in Europe.

The pond watershed had a surface area of 86.2 ha. Since 2016, a large part (33.8 ha) had been cultivated according to a conventional 3-year rotation of rapeseed, wheat, barley, and potentially peas, whereas 42.5 ha had been used as permanent pastures (data retrieved by means of a farmer survey). Based on the information provided by the farmer, all data regarding treatment periods and crops grown on treated plots are given in Table 1.

2.2. Sampling strategy

The aforementioned three streams that were monitored in the study (i.e., UP1, UP2, and DO) were each equipped with a sampling station to carry out continuous flow-based water sampling. Mean sampled water volumes were $277 \pm 51 \text{ m}^3$ at UP1, $104 \pm 12 \text{ m}^3$ at UP2, and

$356 \pm 58 \text{ m}^3$ at DO. Each sampling station was composed of an automatic sampler (Hach Sigma SD-900, United States) controlled by an IFQ type logger (Flow-Tronic S.A., Belgium). Each automatic sampler allowed the storage of 24 polyethylene bottles of 1 L samples. Samples were retrieved once a week. The IFQ logger triggered water sampling by the automatic sampler following measurements of two sensors. The first was the Beluga flow sensor (Flow-Tronic S.A., Belgium). It emits in the water column a continuous 1 MHz frequency acoustic signal, and the receiver samples multiple frequency signals to calculate velocity with high accuracy ($\pm 0.01 \text{ m/s}$) and resolution (0.001 m/s). This probe allows the calculation of negative flow velocities (-2 to 6 m/s). Given the moderate slope (2.2%) that characterized the water inlet tributaries, this specification allowed us to take into account pond backflow events and obtain accurate water flow measurements. For level calculation, a second sensor was used simultaneously (ULS-02 ultrasonic level sensor, Flow-Tronic S.A., Belgium). It allows continuous level calculation by processing continuous echoed ultrasonic waves that are emitted toward the water flow, with high precision ($\pm 0.3 \text{ mm}$) and resolution (1 mm). Water flows were measured (based on measurements from the two probes) every minute at all three stations. The sampling frequency was thus automatically adjusted in order for the number of samples to be proportional to the volume of water flowing. Thus, samples were mainly collected during flood events and varied with seasonal precipitation.

During the study period, UP1, UP2, and DO sampling stations collected a total of 121, 52, and 64 water samples, respectively. Between July 3rd and October 15th, no samples were collected as streams were dry. During this period, DO station only sampled water coming from the main drain channel through dike leaks.

Samples were kept in the dark at -18°C and analyzed within 15 days to avoid degradation and sorption to sampling bottles. The stability of analyzed compounds in polyethylene bottles was tested prior to sampling and was found to be good for at least the first 15 days at 20°C and 17 days at 10°C (Table A.4 in the Supplementary appendix). Nevertheless, a few compounds showed low stability (i.e., less than 15 days at 10°C) in polyethylene bottles and were, therefore, removed from analysis (e.g., chlorpyrifos, CPO, chlorthiamide, fenthion). High temperatures in the summer season (37.6°C at their highest) were not an issue for sample conservation because no sampling was done at that time owing to lack of precipitation.

2.3. Pesticide and transformation product quantification in water

The list of pesticides and TP investigated in this study was established upon analysis of the treatments applied to the watershed

Table 1

Treatments applied to the watershed between 2017 and 2019 and crops grown on treated plots.

Year and crops	Month	Pesticide	Amounts (g)
2017 Barley and peas	April	Napropamide	8100
	August	Glyphosate	10,800
	September	Metazachlor	8100
	October	Metazachlor	3360
	October	Prosulfocarb	36,000
	October	Clopyralid	600
	October	Prosulfocarb	28,800
	October	Esfenvalerate	NA
2018 Rapeseed and wheat	April	Pyroxulam	225
		Florasulam	75
	April	Mesosulfuron-methyl	180
		Iodosulfuron-methyl-sodium	36
	April	Lambda-cyhalothrin	45
	April	Metsulfuron-methyl	36
2019 Sunflower and wheat	April	Pendimethalin	11,250
	April	Dimethenamid-P	9547
	May	Nicosulfuron	450
	October	Flufenacet	2880
	October	Flufenacet	2880

Table 2

Detection (FD) and quantification (FQ) frequencies of pesticides and TP and associated detection (LOD) and quantification (LOQ) limits in the two upstream rivers, at UP1 (N = 121), UP2 (N = 52), and DO (N = 64) stations.

	LOD (µg/L)	LOQ (µg/L)	FD UP1 (%)	FD UP2 (%)	FD DO (%)	FQ UP1 (%)	FQ UP2 (%)	FQ DO (%)
Herbicides and TP								
Acetochlor	0.01	0.02	0	0	0	0	0	0
Acetochlor-OXA	0.005	0.01	0	0	0	0	0	0
Alachlor	0.01	0.02	0	0	0	0	0	0
Alachlor-OXA	0.005	0.01	0	0	0	0	0	0
Alachlor-Acetochlor-ESA	0.005	0.01	0	0	0	0	0	0
Atrazine	0.0025	0.005	0	0	0	0	0	0
Atrazine-2-hydroxy	0.005	0.01	87.6	100	100	60.3	94.2	85.9
Desethylatrazine (DEA)	0.005	0.01	0	0	0	0	0	0
DEA-OH	0.005	0.01	0	0	0	0	0	0
Deisopropylatrazine (DIA)	0.005	0.01	0	0	0	0	0	0
DIA-OH	0.01	0.02	0.8	0	0	0.8	0	0
Desethyldeisopropylatrazine (DEDIA)	0.01	0.02	0	0	0	0	0	0
Bentazone	0.0025	0.005	99.2	0	3.13	85.1	0	0
Folpet	NA	NA	NA	NA	NA	NA	NA	NA
Benzamide	0.025	0.05	0.8	0	0	0	0	0
Chloridazon	0.0025	0.005	0	0	0	0	0	0
DPC (desphenyl chloridazon)	0.1	0.2	10	5.8	0	0	1.9	0
Me DPC	0.005	0.01	9.99	0	0	1.7	0	0
Chlorotoluron	0.0025	0.005	51.2	0	39.06	41.3	0	9.4
Dimethachlor	0.0025	0.005	16.5	0	0	13.2	0	0
Dimethachlor-ESA	0.0025	0.005	100	96.2	100	100	82.7	100
Dimethachlor-OXA	0.025	0.05	69.4	61.5	0	53.7	42.3	0
Dimethenamid	0.005	0.01	62	13.5	51.6	47.1	5.8	23.4
Dimethenamid-ESA	0.0025	0.005	99.2	0	85.9	99.2	0	70.3
Dimethenamid-OXA	0.01	0.02	97.5	9.6	57.8	66.1	0	57.8
Flufenacet	0.0025	0.005	92.6	69.2	37.5	91.7	67.3	31.3
Flufenacet-ESA	0.0025	0.005	100	100	100	100	100	100
Flufenacet-OXA	0.005	0.01	100	80.8	93.75	100	78.8	89.1
Glyphosate	0.033	0.1	69.4	13.5	9.4	28.9	5.8	1.6
AMPA	0.0083	0.025	96.7	40.4	21.9	81.8	5.8	0
Isoproturon	0.0025	0.005	23.1	0	0	18.2	0	0
Desmethyl-Isoproturon	0.0025	0.005	29.8	1.9	12.5	0	0	0
Didesmethyl-Isoproturon	0.0025	0.005	0	0	0	0	0	0
MCPA	0.005	0.01	4.1	21.2	4.7	2.5	17.3	3.1
Metazachlor	0.0025	0.005	99.2	0	21.9	98.3	0	14.1
Metazachlor-ESA	0.01	0.02	100	100	100	100	100	100
Metazachlor-OXA	0.01	0.02	100	100	100	100	100	100
Metsulfuron-Methyl	0.0025	0.005	3.3	25	0	1.7	25	0
Saccharin	0.01	0.02	0.8	17.3	3.1	0	11.5	3.1
Metolachlor	0.0025	0.005	100	15.4	62.5	100	11.5	39.1
Metolachlor-ESA	0.0025	0.005	100	75	100	100	69.2	100
Metolachlor-OXA	0.01	0.02	97.5	17.3	98.4	97.5	0	82.8
CGA-357704	0.01	0.02	71.1	0	21.9	47.1	0	1.6
CGA-368208	0.005	0.01	65.3	0	0	28.9	0	0
CGA-37735	0.0025	0.005	0	0	0	0	0	0
CGA-50267	0.0025	0.005	0	0	0	0	0	0
CGA-50720	0.01	0.02	5.8	0	0	0	0	0
S-Metolachlor-NOA	0.025	0.05	81	0	45.3	52.1	0	3.1
Terbumeton	0.0025	0.005	0	0	0	0	0	0
Terbumeton-Desethyl	0.0025	0.005	0	0	0	0	0	0
Terbuthylazine	0.0025	0.005	17.4	0	12.5	13.2	0	1.6
Terbuth-Desethyl-OH	0.0025	0.005	0	0	0	0	0	0
Terbuthylazine-Desethyl	0.0025	0.005	1.7	0	17.2	0	0	1.6
Terbuthylazine-Hydroxy	0.0025	0.005	0	0	0	0	0	0
Fungicides and TP								
Boscalid	0.0025	0.005	100	100	65.6	99.2	100	9.4
Tebuconazole	0.0025	0.005	100	61.5	96.9	100	25	78.1
Acetamiprid	0.005	0.01	0	0	0	0	0	0
IM-1-4	0.005	0.01	0	0	0	0	0	0
Carbofuran	0.0025	0.005	0	0	0	0	0	0
Clothianidin	0.005	0.01	0	0	0	0	0	0
TZMU (Thiazolyl methylurea)	0.025	0.05	0	0	0	0	0	0
Dimethoate	0.0025	0.005	0	0	0	0	0	0
Imidacloprid	0.005	0.01	1.7	9.6	0	0	5.8	0
Imidacloprid-Urea	0.0025	0.005	0	0	0	0	0	0
Imida-Olefin	0.05	0.1	0	0	0	0	0	0
6-Chloronicotinic-Acid	0.01	0.02	0	0	0	0	0	0
Omethoate	0.0025	0.005	0	0	0	0	0	0
Thiamethoxam	0.005	0.01	0	0	0	0	0	0

since 2000 and historic contaminants now banned from use. It included 67 substances in total, with 18 herbicides and 35 of their TP, 2 fungicides, and 8 insecticides and 4 of their TP (Table 2). Each water sample was analyzed by high-performance liquid chromatography-electrospray ionization-tandem mass spectrometry (HPLC-ESI-MS/MS) using HPLC-LC20AD (Shimadzu, Marne-la-Vallée, France) coupled with QTRAP® 5500 system (Sciex, Villebon-sur-Yvette, France). Limits of detection (LOD) and quantification (LOQ) are given in Table 2. Details regarding the method of analysis and hardware setup are given in Tables A.1 to A.3 in the Supplementary appendix.

Briefly, a 1 mL aliquot was taken from each initial sample and supplemented with 10 µL of an 8 ng/mL internal standard solution (HPC standards GmbH, Neochema, Bodenheim, Germany; Fluka, Toronto Research Chemicals, North York, Canada; Cambridge Isotope Laboratories, Tewksbury, United States) (purity >96%; the full list of internal standards is provided in Tables A.1 and A.2 in the Supplementary appendix). Then, samples were centrifuged at 20,800g for 15 min at 18 °C (Eppendorf 5810R, Montesson, France), allowing suspended particles to separate. The supernatant was transferred into 2 mL glass vials and 100 µL of preparation was injected and analyzed. In between each sample, the automatic sampler was rinsed with a mixed solution of ultrapure water, methanol, acetonitrile, and isopropanol (in equal proportions, 25% of final volume), and then with ultrapure water. Ultrapure water, formic acid, and acetonitrile were of ULC-MS quality (Biosolve, Dieuze, France). One sample per injection series was spiked with analytical standards to control accuracy (recovery range, 75%–125%). Blank samples were added at the beginning and end of each series, and quantification was performed with internal calibration. Samples with high concentrations (above the highest calibration point, $\pm 10\%$) were diluted to meet the calibration range. Injection results were then interpreted using MultiQuant software (version 3.0.1, Sciex, Villebon-sur-Yvette, France).

Concerning glyphosate and AMPA (aminomethanesulfonic acid), the analysis was based on online solid-phase extraction coupled with HPLC-MS/MS. As they are too polar to be analyzed with classical systems, a derivatization step with the FMOC (fluorenylmethoxycarbonyl) protection group was necessary before injection. Briefly, 1540 µL of water sample was spiked with 7.5 µL of a 40 µg/L solution of ¹³C¹⁵N AMPA-FMOC, 100 µL of 250 mM disodium EDTA, 200 µL of 50 mM sodium tetraborate buffer, and 160 µL of FMOC mix (0.5 g/L in acetonitrile). Derivatization was performed with a thermomixer at 50 °C and for 20 min. The reaction was then stopped with 20 µL of H₃PO₄ (2% in ultrapure water) and the mixture was transferred into a 1.5 mL polypropylene injection vial for analysis (details on the analytical methods are available in Tables S.4 and S.5 in the Supplementary appendix). One sample per injection series was spiked with analytical standards to control recovery accuracy. Blank samples were added at the beginning and end of each series, and quantification was performed with internal calibration for AMPA, while glyphosate was quantified with external calibration based on comparison with ultrapure water, spiked at 10, 20, 50, 100, 150, 200, 300, and 400 ng/L, and extracted as field samples. Samples with high concentrations (above the highest calibration point, $\pm 10\%$) were diluted to meet the calibration range. Injection results were then interpreted using TraceFinder (4.1) and Xcalibur (2.2 SP1.48) software.

All solvents and reagents used were of LC-MS purity grade.

2.4. Data analysis

Pesticide and TP concentrations allowed the calculation of mean concentrations \pm standard error of the mean (SEM) upstream and downstream from the pond as well as maximum concentrations of all analyzed compounds. Only substances having a frequency of quantification (FQ) of at least 80% were averaged, in order to avoid estimating concentrations for pesticides and TP poorly represented in the dataset.

Concentrations below the LOD were set to 0, those between the LOD and the LOQ were set to LOQ/2.

Based on the LOD and LOQ, we calculated the frequency of detection (FD) and quantification for each compound at upstream and downstream locations. The numbers of samples with values above the LOD or LOQ were divided by the total number of samples ($N = 121$ at UP1, 52 at UP2, and 64 at DO) and set in percentage form. Maximum and mean reduction rates (%) were calculated based on the decrease in pesticide and TP concentrations between upstream (UP1 and UP2) and downstream (DO) locations. For compounds with a FQ > 80%, a reduction rate of the mean concentration was calculated (i.e., downstream mean concentration/upstream mean concentration). In the same way, a reduction rate of the maximum concentration was obtained for the highest quantified concentrations. When both tributaries showed different mean and/or maximum concentrations, data from UP1 were used to calculate the reduction rates in order to take into account the major water source, which represented at least 80% of water inputs every time.

For each substance (and for the sum of all quantified substances) and at each sampling location (UP1, UP2, and DO), based on pesticide and TP concentrations, the events exceeding environmental or ecotoxicological standards were accounted for. Environmental and ecotoxicological standards were retrieved from either the INERIS database on chemical substances (Portail Substances Chimiques, 2009) or the IUPAC (International Union of Pure and Applied Chemistry) database on pesticide properties (Lewis et al., 2016).

Comparisons of contaminant concentrations between upstream and downstream stations were assessed using Welch's two-sample *t*-tests. Graphical representations and statistical analyses were done with R software (version 4.0.2, © The R Foundation).

3. Results

3.1. Occurrence of pesticides and TP in headwater streams

Over the 10-month study period (i.e., an entire fish breeding cycle from pond filling to pond emptying), 173 samples were retrieved from the upstream stations. Out of the 67 investigated substances, 38 were at least detected in one sample (Table 2). Three TP (i.e., flufenacet-ESA, metazachlor-ESA, and OXA) were present in all samples. At UP1, the number of pesticides detected in one sample ranged from 5 to 12. The number of detected TP was even higher and ranged from 10 to 17 compounds. At UP2, this number ranged from 1 to 9 for pesticides and from 4 to 12 for TPs in any sample (Fig. 2). Upstream from the pond, TPs represented on average $65.8\% \pm 0.6\%$ of the detected substances. Whatever the water sample (UP and DO stations alike), a mixture of at least 5 compounds was detected, reaching 29 different contaminants in one sample at its highest (at UP1 station, Fig. 2). The average numbers of compounds (i.e., pesticides and TP) detected per sample in UP1 and UP2 stations were 22.5 ± 0.51 and 11.3 ± 0.78 , respectively. Among the 38 detected substances, 31 showed concentrations higher than the LOQ at UP1 and 20 at UP2. In both cases, TP accounted for more than 50% of the substances.

At both upstream locations, several groups showing a high FQ were formed by pesticides and their respective TP. Indeed, the most quantified herbicides were flufenacet, metazachlor, and metolachlor (with a FQ as high as 100% for metolachlor at UP1) and TP of metazachlor were present in all samples at the upstream stations. In addition, the most quantified herbicide TP upstream were AMPA, atrazine-2-hydroxy, dimethachlor-ESA, dimethenamid-ESA, flufenacet-ESA and OXA, and metolachlor-ESA and OXA. Regarding fungicides, boscalid was highly quantified at both UP1 and UP2 stations, and tebuconazole mainly at UP1. Insecticides were rarely quantified or detected (Table 2).

Downstream from the pond, 64 water samples were collected and 27 different compounds were at least detected in one sample (Table 2), 59% of them being TP. The average number of compounds

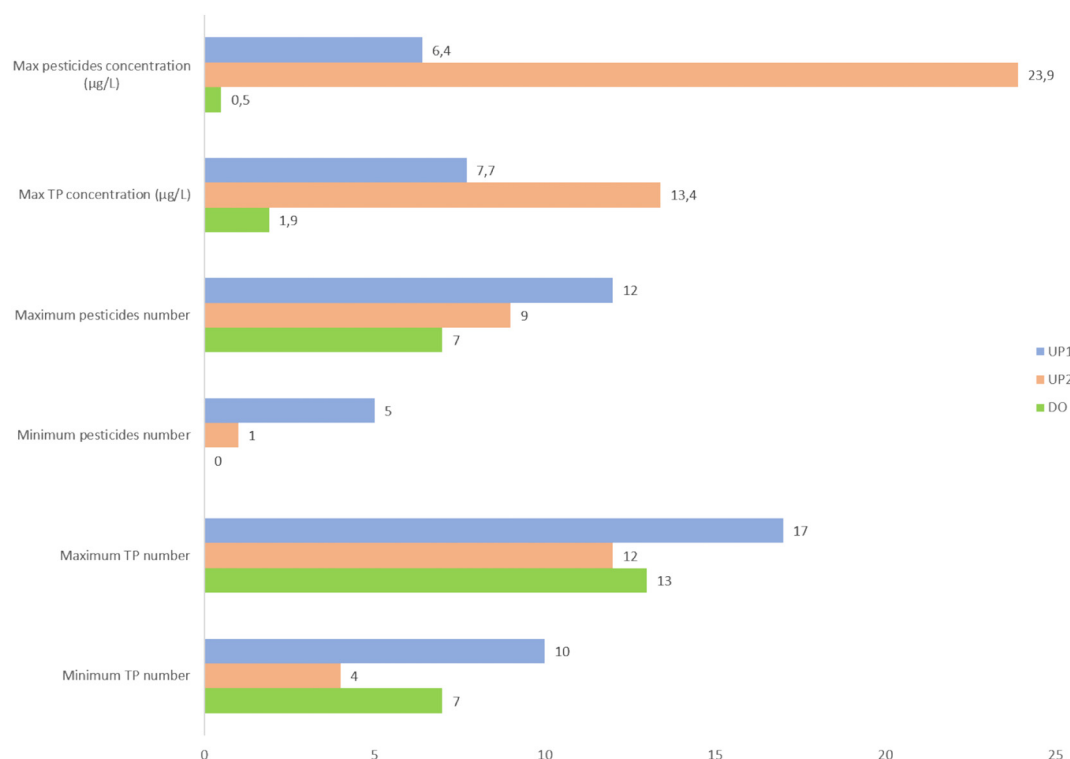


Fig. 2. Summary of maximum and minimum characteristics of upstream (UP1 and UP2) and downstream (DO) water samples ($N = 121, 52$, and 64 , respectively). Maximum pesticide and TP concentrations (sum of all pesticides or TP in one sample) as well as maximum and minimum numbers of pesticides and TP (for molecules with concentrations $> \text{LOD}$) are shown.

detected at DO station was 14.6 ± 0.8 . The number of pesticides and TP in one sample ranged from 0 to 7 and from 7 to 13, respectively (Fig. 2).

Out of the 27 detected compounds, 24 showed concentrations above the LOQ. Dimethenamid, flufenacet, and metolachlor were the most quantified herbicides with a FQ between 23.4% and 39.1%. The fungicide tebuconazole was quantified in 78.1% of the samples (Table 3). More than half (54%) of these 24 substances were TP.

At DO station, just like at UP1/UP2 stations, groups of compounds showing a high FQ were formed by pesticides and their respective TP (Table 2). Flufenacet, metazachlor, and metolachlor were quantified in many samples, as were their TP. For instance, metolachlor was quantified in 39.1% of the samples, and metolachlor-ESA and OXA in 100% and 82.8%, respectively.

3.2. Pesticides and TP concentrations in agricultural headwater streams and pond-induced reduction rates

According to the analytical method used (FQ $> 80\%$), 16 substances allowed calculation of an average concentration $\pm \text{SEM}$ upstream from the pond (Table 3). Considering all 38 compounds per sample (those with a concentration $> \text{LOD}$), the mean concentration recorded over the study period was $3.5 \pm 0.18 \mu\text{g/L}$ at UP1 and 5.3 ± 0.9 at UP2 (all pesticide and TP kinetics during the study period are shown in the Supplementary appendix). The maximum concentrations of pesticides were $6.4 \mu\text{g/L}$ and $23.9 \mu\text{g/L}$, and those of TP were $7.7 \mu\text{g/L}$ and $13.4 \mu\text{g/L}$ at UP1 and UP2, respectively (Fig. 2).

During the study period, flufenacet had a mean concentration of $0.529 \pm 0.09 \mu\text{g/L}$ at UP1 (Table 3) and was quantified at high concentration in November and December at UP1 and UP2 (5.4 and $3.1 \mu\text{g/L}$, respectively). In contrast, its TP (i.e., flufenacet-ESA and OXA) were found to be at respective concentrations as high as 6.2 and $5.6 \mu\text{g/L}$ at UP2 throughout the whole study period (Fig. 3). Metazachlor was also highly quantified throughout the same period, with a mean concentration of $0.0473 \pm 0.007 \mu\text{g/L}$ at UP1 (Table 3). Both its monitored TP showed high concentrations during sampling (Fig. 3). Higher mean and

maximum concentrations were observed at UP2, with a mean concentration of $0.871 \pm 0.09 \mu\text{g/L}$ for metazachlor-ESA (maximum of $2.8 \mu\text{g/L}$) and $0.816 \pm 0.1 \mu\text{g/L}$ for metazachlor-OXA (maximum of $4.8 \mu\text{g/L}$). Metolachlor had a mean concentration of $0.0752 \pm 0.008 \mu\text{g/L}$ at UP1 (Table 3) and was mostly present at the beginning of the study period (Fig. 3). Throughout the course of the study, its TP metolachlor-ESA and OXA were observed to have mean concentrations of 0.156 ± 0.008 and $0.169 \pm 0.01 \mu\text{g/L}$ (Table 3), respectively, with the highest concentrations in November and December (Fig. 3).

At DO location, the mean total concentration (i.e., all substances per sample considered) was $1.08 \pm 0.07 \mu\text{g/L}$ during the study. The maximum concentration of pesticides was $0.5 \mu\text{g/L}$ and that of their TP was $1.9 \mu\text{g/L}$ (Fig. 2).

Flufenacet-ESA and OXA were present throughout the whole study period (Fig. 3) with mean concentrations of $0.244 \pm 0.02 \mu\text{g/L}$ and $0.0719 \pm 0.006 \mu\text{g/L}$, respectively. Metazachlor-ESA and OXA were always present at higher concentrations at DO than their parent compound (Fig. 3) and had mean concentrations of $0.179 \pm 0.008 \mu\text{g/L}$ and $0.263 \pm 0.01 \mu\text{g/L}$, respectively (Table 3) during the same period. Both metolachlor-ESA and OXA were present in 100% and 82.8% of the water samples (Table 2), with mean concentrations of $0.0394 \pm 0.002 \mu\text{g/L}$ and $0.0422 \pm 0.003 \mu\text{g/L}$, respectively, throughout the study (Table 3 & Fig. 3).

Reduction rates were calculated for both mean and maximum concentrations (Table 3). When UP1 and UP2 had different values, UP1 was selected for the calculation of the reduction rate, in order to consider the main water input to the pond. For all substances having a calculated mean concentration (FQ $> 80\%$) except atrazine-2-hydroxy, reduction rates were positive, meaning that mean concentrations at DO were lower than those at UP1 and/or UP2. Regarding atrazine-2-hydroxy, its mean concentration at DO was 16% higher than that at UP2, which was not significantly different. Regarding dimethachlor-ESA, flufenacet-ESA, and metazachlor-ESA and OXA, their mean concentrations at DO were significantly lower than those at UP2. The reduction rates of the mean concentrations ranged from 34% for flufenacet-ESA

Table 3Pesticide and TP mean and maximum concentrations ($\mu\text{g/L}$) at UP1, UP2, and DO stations and associated reduction rates.

	Mean ^a UP1 \pm SEM	Mean UP2 \pm SEM	Max UP1 ^a	Max UP2	Mean DO \pm SEM	Max DO	Reduction rates ^b (mean/max)
Herbicides and TP							
Atrazine-2-hydroxy	–	0.021 \pm 0.002	0.05	0.05	0.0243 \pm 0.002	0.04	–16/20
DIA-OH	–	–	0.09	0	0	0	100/100
Bentazone	0.0086 \pm 0.0006	–	0.05	0	–	0.003	NA/94
Folpet	NA	NA	NA	NA	NA	NA	NA/NA
Benzamide	–	–	0.003	0	0	0	100/100
Chloridazon	–	–	0	0	–	0	NA/NA
DPC	–	–	0	0.2	0	0	100/100
Me-DPC	–	–	0.02	0	0	0	100/100
Chlorotoluron	–	–	0.06	0	–	0.009	NA/85
Dimethachlor	–	–	0.01	0	0	0	100/100
Dimethachlor-ESA	0.0522 \pm 0.003***	0.0707 \pm 0.008***	0.2	0.2	0.0196 \pm 0.0008	0.04	63/80
Dimethachlor-OXA	–	–	0.1	0.2	0	0	100/100
Dimethenamid	–	–	0.7	0.03	–	0.2	NA/71
Dimethenamid-ESA	0.114 \pm 0.01	–	1	0	–	0.05	NA/95
Dimethenamid-OXA	–	–	1.5	0.01	–	0.1	NA/93
Flufenacet	0.529 \pm 0.09	–	5.4	3.1	–	0.5	NA/91
Flufenacet-ESA	0.371 \pm 0.03**	0.992 \pm 0.2***	1.5	6.2	0.244 \pm 0.02	0.7	34/53
Flufenacet-OXA	0.169 \pm 0.02***	–	1.2	5.6	0.0719 \pm 0.006	0.2	58/83
Glyphosate	–	–	1.7	0.2	–	0.1	NA/94
AMPA	0.0739 \pm 0.007	–	0.6	0.05	–	0.01	NA/98
Isoproturon	–	–	0.03	0	0	0	100/100
Desmethyl-isoproturon	–	–	0.003	0.003	–	0.003	NA
MCPA	–	–	1.4	23.7	–	0.02	NA/99
Metazachlor	0.0473 \pm 0.007	–	0.6	0	–	0.02	NA/97
Metazachlor-ESA	0.459 \pm 0.02***	0.871 \pm 0.09***	1.5	2.8	0.179 \pm 0.008	0.4	61/73
Metazachlor-OXA	0.728 \pm 0.04***	0.816 \pm 0.1***	2.6	4.8	0.263 \pm 0.01	0.6	64/77
Metsulfuron-Methyl	–	–	0.02	0.5	0	0	100/100
Saccharin	–	–	0.01	0.06	–	0.06	NA/–500
Metolachlor	0.0752 \pm 0.008	–	0.6	0.02	–	0.03	NA/95
Metolachlor-ESA	0.156 \pm 0.008***	–	0.6	0.02	0.0394 \pm 0.002	0.08	75/87
Metolachlor-OXA	0.169 \pm 0.01***	–	1.1	0.001	0.0422 \pm 0.003	0.009	75/99
CGA-357704	–	–	0.1	0	–	0.02	NA/80
CGA-368208	–	–	0.02	0	0	0	100/100
CGA-50720	–	–	0.01	0	0	0	100/100
S-Metolachlor-NOA	–	–	0.1	0	–	0.06	NA/40
Terbutylazine	–	–	0.01	0	–	0.006	NA/40
Terbutylazine-desethyl	–	–	0.003	0	–	0.01	NA/–233
Fungicides and TP							
Boscalid	0.0128 \pm 0.0007	0.0436 \pm 0.004	0.05	0.1	–	0.006	NA/88
Tebuconazole	0.052 \pm 0.006	–	0.5	0.01	–	0.02	NA/96
Insecticides and TP							
Imidacloprid	–	–	0.005	0.02	0	0	100/100

^a For the calculation of mean and maximum concentrations at UP1, UP2, and DO, a non-detected molecule was set to 0 and an unquantified molecule (above the LOD) was set to half its LOQ.^b Reduction rates were calculated regarding mean/max pesticide and TP concentrations (%). UP1 concentrations were used when different from UP2 concentrations, in order to take into account the major water input, * Molecules having mean concentrations significantly different between UP1 and DO and/or UP2 and DO (*p-value <0.05, **p-value <0.01, ***p-value <0.001).

up to 75% for metolachlor-ESA and OXA. The low FQs of DIA-OH and several other compounds including benzamide and dimethachlor did not allow their mean concentrations to be expressed at UP1/UP2, but since they were never quantified at DO, positive reduction rates were expected.

Based on the maximum concentrations (i.e., peak concentrations), reduction rates were mainly positive, meaning that maximum concentrations at DO were lower than those at UP1/UP2. Considering highly quantified compounds, maximum reduction rates ranged from 20% for atrazine-2-hydroxy up to 100% for DIA-OH, benzamide, and eight other compounds (Table 3).

3.3. Suppression of threshold exceedances events downstream from the pond

In upstream locations and during late 2019, 37 flufenacet exceedance events were detected regarding regulatory acceptable concentration (RAC) 15 exceedance events of EC₅₀ for the bioindicator *Raphidocelis subcapitata* (13 at UP1 and 2 at UP2) were detected regarding flufenacet concentrations. In addition, 90 exceedances of the EQS value for metazachlor, set by the WFD (Order of July 27, 2015), were detected at UP1 between February and May, and again in November (Table 4 and Fig. 3).

Mid-May, 8 exceedances of the regulatory acceptable concentration (RAC) values were highlighted for metsulfuron-methyl at UP2 (Table 4). Also, the EQS threshold was exceeded for MCPA mainly at UP1 in May. At DO location, 5 exceedances of RAC values were observed for flufenacet (Table 4).

4. Discussion

4.1. Occurrence of pesticides and TP in headwater streams

At the very beginning of the headwater streams, mixtures of pesticides and TP were present in surface water (22.5 and 11.3 substances on average at UP1 and UP2, respectively). At DO station, the number of detected substances was found to be 14.6 on average, therefore smaller than at UP1 but greater than at UP2, the reason for the latter observation being a greater number of TP (Fig. 2) coming from UP1. Moreover, TP represented more than half of the substances quantified upstream and downstream from the pond. This result seems to be in agreement with studies carried out in lentic systems, whose characteristics are similar to those of ponds, and which reported the occurrence of metazachlor and flufenacet TP in 39% to 94% of their water samples

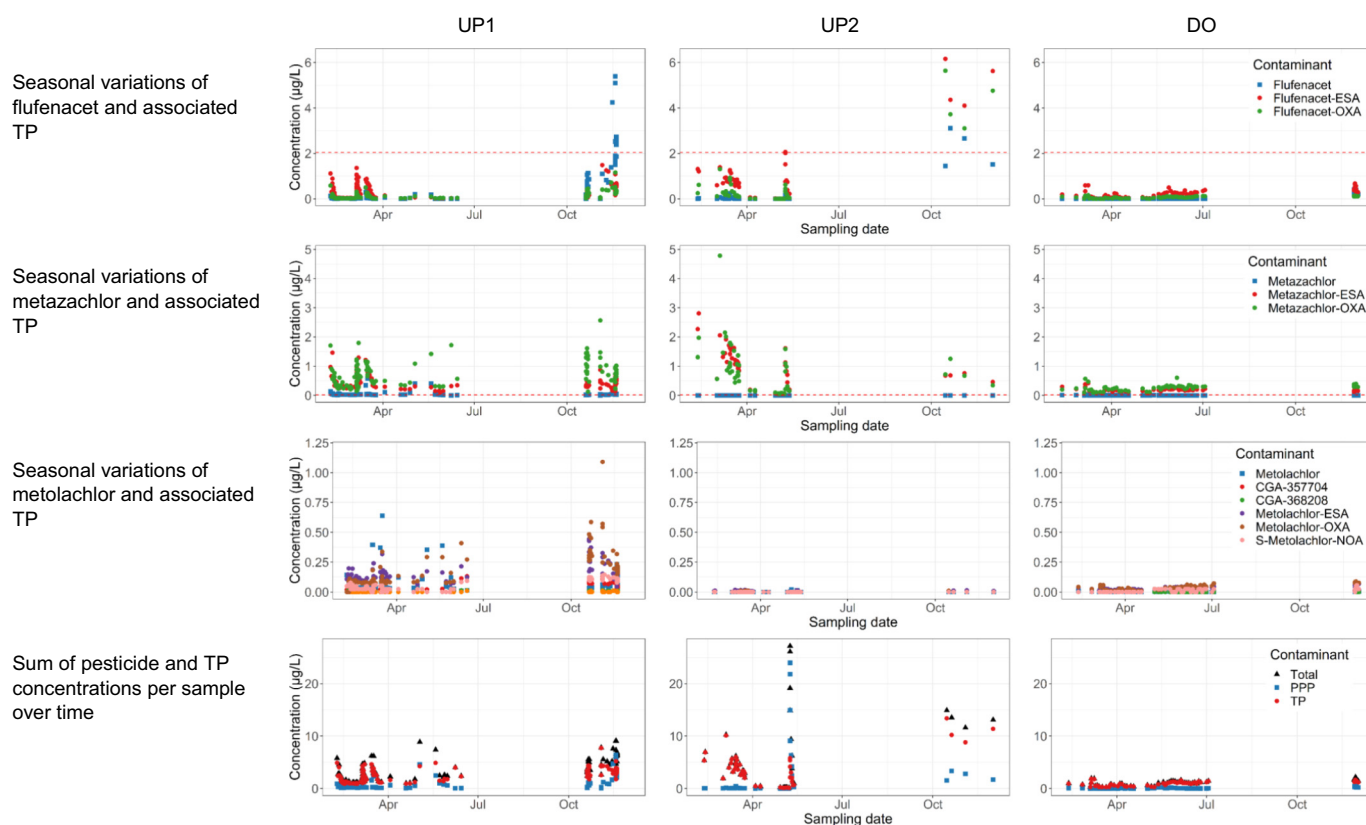


Fig. 3. Seasonal variations in pesticide and associated transformation product (TP) concentrations at UP1, UP2, and DO stations over the study period (February to December 2019). Flufenacet, metazachlor, metolachlor, and sum of pesticide and TP concentrations per sample are represented as a function of time. Dotted red lines stand for algae (*R. subcapitata*) acute 72-hour EC₅₀ (2.04 µg/L) for flufenacet, environmental quality standard (0.02 µg/L) for metazachlor. Concentrations below the LOD were set to 0 and those between the LOD and LOQ were set to LOQ/2. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

(Ulrich et al., 2018). In the same way, Fonseca et al. (2019) noted the occurrence of TP in (Spanish) streams, which accounted for 22% of the detected pesticide-related substances. These results call attention to the occurrence of TP in the environment. They show the importance of considering compound mixtures when studying ecotoxicological effects. In a study carried out across several European countries (France, Germany, and the Netherlands) and the United States, Schreiner et al. (2016) revealed, based on the analysis of routine monitoring data (292 compounds for France), the presence of 2 to 5 substances per sample, the majority of which being herbicides. Our results suggest higher levels of contamination of headwater streams with pesticide mixtures (as high as 29 different compounds per sample), especially in areas of intensive agricultural activities. Furthermore, other studies showed an occurrence of TP in surface waters greater than that of the present study. Indeed, Moschet et al. (2014) reported water samples containing between 15 and 25 different TP (as high as 17 in our samples), in rivers with larger agricultural/urbanized catchments (between 38 km² and

105 km²) than that studied here, potentially representing an even greater source of contamination.

Our analysis suggests a positive effect of the pond on the number of detected substances: by decreasing the number of pesticides and TP, ponds may induce a reduction in ecotoxicity risks (Cedergreen, 2014; Relyea, 2009; Rizzati et al., 2016).

The same trend was observed in Ohio (Midwestern United States), where TP are present at the beginning of channelized agricultural watersheds and mixtures (pesticides and TP) are generally more frequent than single substances. Concentrations depend on treatments types and climatic conditions, and can vary annually and seasonally (Smiley Jr et al., 2014). Therefore, our approach, based on frequent water sampling, appears to be relevant in highlighting the occurrence of TP in headwater streams. The same results were obtained in southwestern Germany, where more than 80% of the water samples coming from studied agricultural headwater streams contained more than 5 substances (pesticides and TP), and half of them showed high concentrations of TP, such as metazachlor-OXA (0.89 µg/L) (Weber et al., 2018). Our results showed higher concentrations for some compounds, with metazachlor-OXA and metolachlor-OXA concentrations exceeding those of Weber et al. (2018) by a factor of 5.3 and 2.75, respectively.

Concerning herbicide parent compounds, flufenacet, metazachlor, and isoproturon were present in our samples, which is consistent with the type of crops grown in the watershed (Table 1) and the importance of herbicides in the crop protection market (Bonanno et al., 2017). Flufenacet was particularly often quantified upstream from the pond, having been applied in the watershed in 2019. Although this herbicide has a relatively low organic carbon sorption coefficient (K_{oc}) of 401 (Lewis et al., 2016), which characterizes it as being moderately mobile, it may be highly mobile in agricultural catchment areas (Willkommen et al., 2019). The latter authors showed significant and continuous

Table 4

Number of events exceeding environmental/ecotoxicological standards at UP1, UP2, and DO stations.

	Flufenacet	MCPA	Metazachlor	Metsulfuron-methyl
Sampling station				
UP1	33	13	1	90
UP2	4	2	8	0
DO	5	0	0	0
Standard limit (µg/L)	0.357 ^a	2.04 ^b	0.5 ^c	0.019 ^d

^a Regulatory acceptable concentration (RAC).

^b Algae (*Raphidocelis subcapitata*) acute 72-hour EC₅₀ (µg/L).

^c Environmental quality standard (WFD, 2000/60/CE).

leaching of flufenacet soon after application under both dry and wet conditions. Our result tends to confirm this mobility, as flufenacet was applied at the beginning of October, and quantified as early as mid-October (Fig. 3). Metazachlor, which was used in 2017, was also often quantified at UP1 (FQ = 99.2%). It has a relatively low 50% dissipation time (DT_{50}) in soil (6.8 days under field conditions) characterizing a non-persistent compound but it is stable in the water phase (DT_{50} of 216 days) (Lewis et al., 2016). However, field studies have shown that metazachlor could be detectable in soil up to 170 days after application (LOD of 0.57 ng/g in soil), depending on soil characteristics (Mantzos et al., 2016b). According to Mantzos et al. (2016b), metazachlor is a potential risk for adjacent surface water within the first month after application. However, our results suggest a more lasting presence, even two years after its application on treated lands. Rising aquifers could explain its occurrence in the study streams, but its low leaching potential ($GUS = 1.75$ according to Lewis et al., 2016) makes it unlikely to reach groundwater bodies during treatment and then be transferred to surface water, like polar molecules, as TP could do (Hintze et al., 2020). Thus, it can be assumed that quantification of metazachlor over the whole study period resulted from accidental spill or undeclared treatments, but no evidence could confirm it.

Moreover, isoproturon, which had no longer been approved as an agricultural treatment since 2016 (EU 2016/872), was still quantified in water samples but at a level far below the EQS limit value of 1 µg/L. It was typically used in cereal crops to control annual grasses and weeds, and considered as a priority hazardous substance by the WFD because of its high environmental accumulation (Muhammad et al., 2019). This molecule is still frequently detected in surface water samples (Casado et al., 2019; Durak et al., 2020), notably because of its historic use and variable DT_{50} in soil ranging from 3 to 200 days (Alletto et al., 2006), thus potentially explaining its presence in our samples even though it was not actually used during the sampling period, nor had it been since 2016.

The two fungicides used in the study catchment (i.e., boscalid and tebuconazole) were detected in all UP1 samples and in 100% and 61.5% of UP2 samples, respectively. Boscalid is one of the most widely used succinate dehydrogenase inhibitor (SDHI) fungicides in the world. A recent study (Bénit et al., 2019) has established that human, honeybee, earthworm, and fungal succinate dehydrogenases (SDHs) are all affected by SDHIs. Furthermore, these authors have shown that cell death due to SDHI exposure was significantly accelerated in fibroblasts derived from patients with neurodegenerative diseases. For these reasons, even though this effect has not been highlighted in vivo yet, SDHIs have come under the close scrutiny of phytopharmacovigilance committees. Contamination of surface water by tebuconazole and boscalid has recently been established in an agricultural catchment in Germany (Tauchnitz et al., 2020). Our results confirm Tauchnitz et al.'s findings but with maximum concentrations that were higher by a factor of 10 (0.01 vs. 0.1 µg/L at UP2) and 50 (0.014 vs. 0.5 µg/L at the highest at UP1) for boscalid and tebuconazole, respectively.

Regarding groups of substances formed by pesticides and their respective TP, an overall decrease in FQ was observed for pesticides between the upstream and downstream stations (FQ of 91.7% and 67.3% at UP1 and UP2, respectively, vs. 31.3% at DO for flufenacet). TP tended to remain highly present in the samples (FQ of 100% at UP1, UP2, and DO for both TP of metazachlor). Greater persistence of TP (compared to pesticides) has been established for metazachlor and flufenacet in waterbodies (Ulrich et al., 2018), which thus can explain their detection at high concentration in the water sampled in the present study, even downstream from the pond.

4.2. Pesticides and TP concentrations in agricultural headwater streams and pond-induced reduction rates

A significant decrease in mean concentrations (p -value < 0.001) was observed between UP1/UP2 and DO stations (3.5 ± 0.2 µg/L and 5.3 ± 0.9 at UP1 and UP2, respectively, vs. 1.09 ± 0.07 µg/L at DO). Such a

decrease was observed for 7 different substances. A clear buffering of contaminant peaks was also observed, with a maximum of 27.2 µg/L at UP2 compared to 2.16 µg/L at DO. Reduction in pesticide concentrations between upstream and downstream rivers has already been demonstrated for ponds, with reduction rates of up to 99% (Gaillard et al., 2016, 2015), as well as for constructed wetlands, which showed variable reduction rates (Budd et al., 2009; Gaullier et al., 2019; Tournebise et al., 2017; Vymazal and Březinová, 2015). Our results indicate a similar trend not only for pesticides, with a 100% reduction rate for several substances such as dimethachlor, isoproturon, and metsulfuron-methyl, but also for TP such as benzamide, dimethachlor-OXA, and DIA-OH. Such retention effect was never highlighted in such an extent, at sampling as well as molecular diversity levels. Considering pesticides and TP, the mean reduction rates of their mean concentrations were 100% and $72.4\% \pm 8.9\%$, respectively. Better reduction rates were observed for parent compounds, reflecting potentially greater stability of TP, which may be less affected by abiotic and biotic processes occurring in the pond (Picó and Barceló, 2015). Atrazine-2-hydroxy had the lowest reduction rate of maximum concentration as compared to the other TP, which may be explained by its great persistence (with a DT_{50} potentially as high as 164 days; Lewis et al., 2016) or low volatility (with a Henry's law constant at 25 °C of $6.36e-8$ Pa·m³/mol). Atrazine and its TP have been reported to be highly persistent in water and soil (Jablonowski et al., 2011). These authors showed that, in Germany, atrazine was the most abundant pesticide in groundwater samples even after having been banned from use for 15 years. Schreiner et al. (2016) have also reported high atrazine and atrazine TP contamination in France, Germany, the Netherlands, and the USA, and identified them as being among the most detected pesticides and TP. The persistence of atrazine-2-hydroxy, combined with the continuous release of atrazine TPs from surrounding agricultural lands being then transported by the main tributaries of the pond (FQs of 60.3% and 94.2% at UP1 and UP2, respectively) and subsurface flows, may thus explain their occurrence at DO station.

It was surprising to observe, on the one hand, the presence of TPs upstream from the pond and, on the other hand, their diversity ($10 \leq TPs \leq 17$ at UP1) (Fig. 2). Indeed, the pond can be seen as a degrading environment, thus leading to increased TP presence downstream from the pond. Their presence upstream reflected high contamination at the beginning of the study headwater streams. Such contamination is related to the soil transformation that takes place between the time of treatment application (Raina et al., 2011), the first rain events post application, and rain events heavy enough to lead to the transportation of these substances (Ulrich et al., 2018).

Nevertheless, by showing the occurrence of TP at a stable high concentration (6.2 µg/L for flufenacet-ESA at UP2) over a long period of time, we highlighted that they should be more considered in monitoring programs. Maximum concentrations of metazachlor and flufenacet TP were particularly high in our samples compared to those of Ulrich et al. Flufenacet-ESA and OXA were quantified at a maximum concentration of 6.2 µg/L (vs. 1.9 in Ulrich et al.) and 5.6 µg/L (vs. less than 0.75 µg/L in Ulrich et al.), respectively, and metazachlor-OXA was quantified at 4.8 µg/L at its highest compared to less than 4 µg/L in Ulrich et al. TP were found to be at higher concentration than their parent substances, which could potentially be correlated to the time of treatment of arable lands, as well as to molecule characteristics. A high FQ of flufenacet in UP1 and UP2 samples may have been caused by direct transfer from arable lands, after pre-emergent treatment in October. Having both pesticides and TP at UP1 and UP2 stations is explained by winter treatments (of 2019 and potentially of previous years) and specific flufenacet characteristics. Longer DT_{50} have been estimated under laboratory conditions, correlated with soil clay content (Gupta and Gajbhiye, 2002), and even higher DT_{50} have been determined in field studies, ranging from 117 up to 145 days (Marín-Benito et al., 2020). Flufenacet will be slowly degraded in treated soils, leading to the formation of TP. However, the parent molecule will also be present when

heavy rainfall occurs, explaining the simultaneous presence of pesticides and TP in water (Willkommen et al., 2019).

Moreover, it raises the question of monitoring pesticide occurrences and concentrations without taking into account related TP, which may be, on average, more concentrated ($0.459 \pm 0.02 \mu\text{g/L}$ and $0.728 \pm 0.04 \mu\text{g/L}$ for metazachlor-ESA and OXA, respectively, vs. $0.0473 \pm 0.007 \mu\text{g/L}$ for metazachlor at UP1) and persistent (Fig. 3). Studies that question the relevance of TP in control and monitoring programs agree and highlight the need to include these substances on official lists. However, given the range of potential substances to be monitored, prioritizing pertinent TP is necessary (Melin et al., 2020). Ecotoxicological effects should be considered, which is the approach adopted by the French Agency for Food, Environmental and Occupational Health, and Safety (ANSES). Furthermore, the selection of the TP to be analyzed should be based on extensive environmental monitoring data (regarding the number of substances searched for and study times), as has been done in the present study, to determine TP actually occurring in anthropized natural environments (Fig. 2).

Contrary to Gaillard et al. (2016), no isoproturon increase was observed in DO samples. This may be due to a decrease in overall pollution, since the substance was banned from agricultural use, or to a difference in sampling strategy. The strategy used in the present study is potentially more precise because based on stream flows. Thus, we were less likely to miss isoproturon peaks at upstream stations during heavy precipitation events, which represent crucial leaching events (Beulke et al., 2002; Filipović et al., 2016).

Counter-intuitively, the pond did not release TP having formed because of hydraulic conditions through biotic (Vidali, 2001; Zablotowicz et al., 2004) and abiotic (Sarmah and Sabadie, 2002) transformation processes. Indeed, they were already present upstream from the pond (Fig. 3), which, however, may have increased their dissipation via these biotic and abiotic processes, allowing concentration reductions downstream. The year 2019 was marked by sparse rainfall, which resulted in incomplete filling of the pond and thus water not reaching the spillway. As a result, hydraulic residence time, normally close to 3 months, was increased to 7.7 months; it is thus likely that TP reaching and forming in the pond were degraded over a longer period of time, explaining such effective reduction between UP1/UP2 and DO stations. The importance of hydraulic residence time has indeed already been highlighted for parent molecules (Stehle et al., 2011).

It is also relevant to consider the storage/release capacities of the pond between the sediment compartment and the water column (Vallée et al., 2014). Indeed, according to their characteristics, pesticides and TP are likely to have a high affinity for particulate matter, thus being added to the sediment compartment. It is generally accepted that the sorption coefficient (K_{oc}) and the octanol/water partition coefficient (LogP) of pesticides are good indicators of their fate in waterbodies, and of their binding to suspended matter (Boithias et al., 2014; Vryzas, 2018). The importance of the sediment compartment in the retention of contaminants, whether pesticides or TP, should therefore also be considered and explored in order to explain the observed upstream/downstream difference.

Finally, it is also important to take into account the dilution effect induced by the pond. Studies focusing on large rivers have shown that pesticide loads are strongly correlated with hydrological parameters, showing a decrease in concentrations in downstream locations where water volumes are large (Climent et al., 2019). For compounds occasionally detected or quantified at UP1 and UP2 but not at DO, such as DIA-OH, benzamide, DPC, Me-DPC, dimethachlor, metsulfuron-methyl, and imidacloprid, and thus having a maximum reduction rate of 100%, dilution may explain the upstream/downstream reduction. All these substances had FQ lower than 20%, and for some of them less than 1% (DIA-OH and benzamide) at UP1 or UP2 location, and were never quantified at DO. They were likely to have remained undetected in the pond and at DO location (regardless of low LOD) because of large water volumes and possibly intra pond transformation processes.

4.3. Suppression of threshold exceedances events downstream from the pond

A total of 159 exceedances of environmental/ecotoxicological standards were observed over the whole sampling period at UP1 and UP2 stations, as against 5 at DO station (Table 4).

MCPA and metsulfuron-methyl are compounds used in cereal crops, and their occurrence at UP2 could be explained by cultivation upstream from the sampling station (see Section 2.1). The presence of tebuconazole could be linked to the type of crops grown in the watershed in previous years (see Section 2.1). It has been reported to have moderate to high persistence depending on soil properties, explaining possible leaching events even though not applied during the study period (Lewis et al., 2016).

In Europe, studies have shown that pesticide concentration thresholds are exceeded in water streams during site-sampling events (Casado et al., 2019; Schreiner et al., 2016; Szöcs et al., 2017; Weber et al., 2018). These events are strongly correlated with pesticide treatments (Zivan et al., 2016) but highly variable because they depend on environmental conditions and substance intrinsic properties such as solubility or soil sorption (Kellogg et al., 2002). Flufenacet was used as winter treatment in late 2019 when precipitation levels were high, thus leaving little time for compound transformation to occur and inducing direct runoff from arable lands. Low stable concentrations were observed at the beginning of the study period (Fig. 3), suggesting that most of the applied doses had already been washed off during rainfall events of late 2018. Flufenacet is well documented, and it is admitted that first runoff events will induce major transport after application (Milan et al., 2013), and then potential toxic effects on aquatic organisms like the bioindicator *R. subcapitata*. Our high frequency sampling indicated peaks of flufenacet (33 events at UP1 and 4 at UP2) from October until the end of December, suggesting significant chronic toxicity risks for aquatic organisms. Regarding metazachlor, concentrations were less variable along the study period (reaching $0.6 \mu\text{g/L}$ at UP1), but higher concentrations were observed at the beginning, in accordance with rapeseed treatment (Fig. 3). Major runoff is expected after rainfall events in treatment periods (Mantzios et al., 2016a). Both of these compounds are characterized by low to moderate K_{oc} and are thus likely to be transported by surface and subsurface runoff during precipitation events.

Regarding total pesticide and TP concentrations and the 44 exceedance events induced (Table 4 & Fig. 3), it is important to note that they take into account all 67 investigated substances (pesticides and TP). If we consider only parent molecules, then only 7 samples showed concentrations above the established threshold (5 events at UP2 and 2 at UP1). In both cases, exceeding events were only detected at UP locations, bringing to the fore the positive effect of the pond on pesticide and TP mixture contamination in agricultural headwater streams. TP were highly present in our water samples; their inclusion in regulations, such as EQS, but also in ecotoxicological risk assessments in the future therefore seems relevant. Nevertheless, many ecotoxicological standards and environmental limits are missing from the literature and pesticide databases. For instance, INERIS provided environmental and toxicity thresholds for 68% of the 28 pesticides monitored in the present study, but only 3% of the 39 TP. Therefore, there is an urgent need to obtain toxicity data on these compounds whose occurrence has been evidenced in the natural environment, such as in headwater streams.

5. Conclusion

Headwater streams are of prime importance when considering the chemical status of surface water. Our study emphasizes their contamination by pesticides and TP, when located in an agricultural watershed. We detected a minimum of 5-compounds mixtures in each sample and highlighted that regardless of the sampling location, TP represented

more than half of the detected and quantified substances. Considering that their concentrations were, in some samples, higher than those of their parent molecules, it seems essential to collect data on them in anthropized environments. Our results showed large pesticide and TP mixtures in water, and suggested several relevant compounds, such as flufenacet TP, to be included in monitoring lists in the future.

The buffer effect of the pond was brought to the fore by the decrease in the diversity of substances occurring in downstream water. Indeed, by reducing mixture diversity it may be able to protect downstream ecosystems from toxicity and the occurrence of synergic and/or additive effects. Finally, exceedances of guidance and threshold values were observed during sampling for several pesticide concentrations upstream from the pond. No exceedances were detected downstream from the pond, but these results should be interpreted with caution as many guidance values were missing concerning TP.

The question raised, then, relates to underlying processes inducing such an upstream/downstream difference. A dilution effect should not be ruled out, and reconciling concentration data with waterflows should make it possible to overcome the biases induced by this approach. Moreover, intra-pond sampling of the sedimentary matrix should make it possible in future work to highlight its role in pesticide and TP dynamics, the storage of a substantial part of the contaminants being a relevant hypothesis.

The demonstration of the prolonged presence and high concentration of TP highlights the importance of their inclusion in ecotoxicological studies, especially when several substances are combined, which is likely to occur repeatedly and on a scale as small as the watershed. Regarding the understanding of the fate of pesticides in the environment, as well as their dynamics, TP analysis is required to avoid missing this insidious contamination.

Funding

This research was carried out within the REPONSE project, funded by the Rhin-Meuse Water Agency and the French Agency for Food, Environmental and Occupational Health & Safety (ANSES).

CRediT authorship contribution statement

François Le Cor: Conceptualization, Methodology, Investigation, Visualization, Formal analysis, Resources, Writing – original draft. **Sylvain Slaby:** Investigation, Visualization, Writing – original draft. **Vincent Dufour:** Investigation, Writing – original draft. **Alain Iuretig:** Investigation, Methodology. **Cyril Feidt:** Conceptualization, Methodology, Supervision. **Xavier Dauchy:** Conceptualization, Validation, Resources, Writing – review & editing, Supervision, Project administration. **Damien Banas:** Conceptualization, Validation, Resources, Writing – original draft, Supervision, Project administration, Funding acquisition.

Declaration of competing interest

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

Acknowledgements

The authors are sincerely grateful to A. and H. Lhuillier for their significant help during sampling, and for providing access to the study pond. They also thank Mr. Nicolai Miguel of the Rhine-Meuse Water Agency for his help and advice in the selection of relevant pesticides and transformation products to be monitored. Finally, they would like to thank L. Pasquini, C. Hollard, and M-C. Clavos for their work during sample analysis. Finally, the authors sincerely thank ABC translation for the proofreading work done.

Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.scitotenv.2021.147715>.

References

- Alletto, L., Coquet, Y., Benoit, P., Bergeaud, V., 2006. Effects of temperature and water content on degradation of isoproturon in three soil profiles. *Chemosphere* 64, 1053–1061. <https://doi.org/10.1016/j.chemosphere.2005.12.004>.
- Bartout, P., Touchart, L., 2018. The limnic footprint index. *Proceedings Air and Water Components of the Environment*, pp. 137–145.
- Belfroid, A.C., Van Drunen, M., Beek, M.A., Schrap, S.M., Van Gestel, C.A.M., Van Hattum, B., 1998. Relative risks of transformation products of pesticides for aquatic ecosystems. *Sci. Total Environ.* 222 (3), 167–183. [https://doi.org/10.1016/S0048-9697\(98\)00298-8](https://doi.org/10.1016/S0048-9697(98)00298-8).
- Bénit, P., Kahn, A., Chretien, D., Bortoli, S., Huc, L., Schiff, M., Favier, J., Gressens, P., Rak, M., Rustin, P., 2019. Evolutionarily conserved susceptibility of the mitochondrial respiratory chain to SDHI pesticides and its consequence on the impact of SDHIs on human cultured cells. *PLoS One*, 1–20. <https://doi.org/10.1371/journal.pone.0224132>.
- Beulke, S., Brown, C.D., Fryer, C.J., Walker, A., 2002. Lysimeter study to investigate the effect of rainfall patterns on leaching of isoproturon. *Pest Manag. Sci.* 58, 45–53. <https://doi.org/10.1002/ps.419>.
- Boithias, L., Sauvage, S., Srinivasan, R., Leccia, O., Sánchez-Pérez, J.M., 2014. Application date as a controlling factor of pesticide transfers to surface water during runoff events. *Catena* 119, 97–103. <https://doi.org/10.1016/j.catena.2014.03.013>.
- Bonanno, A., Matera, V.C., Venus, T., Wesseler, J., 2017. The plant protection products (PPP) sector in the European Union: a special view on herbicides. *Eur. J. Dev. Res.* 29, 575–595. <https://doi.org/10.1057/s41287-017-0088-1>.
- Budd, R., O'Geen, A., Goh, K.S., Bondarenko, S., Gan, J., 2009. Efficacy of constructed wetlands in pesticide removal from Tailwaters in the Central Valley, California. *Environ. Sci. Technol.* 43, 2925–2930. <https://doi.org/10.1021/es802958q>.
- Bunzel, K., Liess, M., Kattwinkel, M., 2014. Landscape parameters driving aquatic pesticide exposure and effects. *Environ. Pollut.* 186, 90–97. <https://doi.org/10.1016/j.envpol.2013.11.021>.
- Burgess, M., Pimentel, D., 2014. Integrated pest management: pesticide problems, vol.3. *Integr. Pest Manag. Pestic. Probl.* Vol.3, 1–474. <https://doi.org/10.1007/978-94-007-7796-5>.
- Carson, R., 1962. Silent Spring.
- Carvalho, F.P., 2017. Pesticides, environment, and food safety. *Food Energy Secur.* 6, 48–60. <https://doi.org/10.1002/fes3.108>.
- Casado, J., Brigden, K., Santillo, D., Johnston, P., 2019. Screening of pesticides and veterinary drugs in small streams in the European Union by liquid chromatography high resolution mass spectrometry. *Sci. Total Environ.* 670, 1204–1225. <https://doi.org/10.1016/j.scitotenv.2019.03.207>.
- Cedergreen, N., 2014. Quantifying synergy: a systematic review of mixture toxicity studies within environmental toxicology. *PLoS One* 9. <https://doi.org/10.1371/journal.pone.0096580>.
- Chow, R., Scheidegger, R., Doppler, T., Dietzel, A., Fenicia, F., Stamm, C., 2020. A review of long-term pesticide monitoring studies to assess surface water quality trends. *Water Res.* X 9, 100064. doi:<https://doi.org/10.1016/j.wroa.2020.100064>.
- Climent, M.J., Herrero-Hernández, E., Sánchez-Martín, M.J., Rodríguez-Cruz, M.S., Pedreros, P., Urrutia, R., 2019. Residues of pesticides and some metabolites in dissolved and particulate phase in surface stream water of Cachapoal River basin, central Chile. *Environ. Pollut.* 251, 90–101. <https://doi.org/10.1016/j.envpol.2019.04.117>.
- Dhananjayan, V., Jayakumar, S., Ravichandran, B., 2020. Conventional methods of pesticide application in agricultural field and fate of the pesticides in the environment and human health, in: K. R. R., Thomas S., Volova T., K.J. (Ed.), *Controlled Release of Pesticides for Sustainable Agriculture*. Springer, Cham, pp. 1–39. doi:https://doi.org/10.1007/978-3-030-23396-9_1.
- Dodds, W.K., Oakes, R.M., 2008. Headwater influences on downstream water quality. *Environ. Manag.* 41, 367–377. <https://doi.org/10.1007/s00267-007-9033-y>.
- Drożdżyński, D., 2008. Studies on residues of pesticides used in rape plants protection in surface waters of intensively exploited arable lands in Wielkopolska province of Poland. *Ann. Agric. Environ. Med.* 231–235.
- Durak, J., Rokoszak, T., Skiba, A., Styszko, K., 2020. Environmental risk assessment of priority biocidal substances on Polish surface water sample. *Environ. Sci. Pollut. Res.* <https://doi.org/10.1007/s11356-020-11581-7>.
- FAO, 2020. FAOSTAT [WWW Document]. Agri-environmental Indic./Pestic. URL <http://www.fao.org/faostat/en/#data/EP> (accessed 11.10.20).
- Filipović, V., Coquet, Y., Pot, V., Houot, S., Benoit, P., 2016. Modeling water and isoproturon dynamics in a heterogeneous soil profile under different urban waste compost applications. *Geoderma* 268, 29–40. <https://doi.org/10.1016/j.geoderma.2016.01.009>.
- Fonseca, E., Renau-Pruñonosa, A., Ibáñez, M., Gracia-Lor, E., Estrela, T., Jiménez, S., Pérez-Martín, M.A., González, F., Hernández, F., Morell, I., 2019. Investigation of pesticides and their transformation products in the Júcar River Hydrographical Basin (Spain) by wide-scope high-resolution mass spectrometry screening. *Environ. Res.* 177, 108570. <https://doi.org/10.1016/j.envres.2019.108570>.
- Gaillard, J., Thomas, M., Lazartigues, A., Bonnefille, B., Pallez, C., Dauchy, X., Feidt, C., Banas, D., 2015. Potential of barrage fish ponds for the mitigation of pesticide pollution in streams. *Environ. Sci. Pollut. Res.* 23, 23–35. <https://doi.org/10.1007/s11356-015-5378-6>.
- Gaillard, J., Thomas, M., Iuretig, A., Pallez, C., Feidt, C., Dauchy, X., Banas, D., 2016. Barrage fishponds: reduction of pesticide concentration peaks and associated risk of adverse

- ecological effects in headwater streams. *J. Environ. Manag.* 169, 261–271. <https://doi.org/10.1016/j.jenvman.2015.12.035>.
- Gaullier, C., Baran, N., Dousset, S., Devau, N., Billet, D., Kitzinger, G., Coisy, E., 2019. Wetland hydrodynamics and mitigation of pesticides and their metabolites at pilot-scale. *Ecol. Eng.* 136, 185–192. <https://doi.org/10.1016/j.ecoleng.2019.06.019>.
- Gregoire, C., Elsaesser, D., Huguenot, D., Lange, J., Lebeau, T., Merli, A., Mose, R., Passepote, E., Payraudeau, S., Schütz, T., Schulz, R., Tapia-Padilla, G., Tournébeize, J., Trevisan, M., Wanko, A., 2009. Mitigation of agricultural nonpoint-source pesticide pollution in artificial wetland ecosystems. *Environ. Chem. Lett.* 7, 205–231. <https://doi.org/10.1007/s10311-008-0167-9>.
- Gruchlik, Y., Linge, K., Joll, C., 2018. Removal of organic micropollutants in waste stabilisation ponds: a review. *J. Environ. Manag.* 206, 202–214. <https://doi.org/10.1016/j.jenvman.2017.10.020>.
- Gupta, S., Gajbhiye, V.T., 2002. Effect of concentration, moisture and soil type on the dissipation of flufenacet from soil. *Chemosphere* 47, 901–906. [https://doi.org/10.1016/S0045-6535\(02\)00017-6](https://doi.org/10.1016/S0045-6535(02)00017-6).
- Hernández, F., Sancho, J.V., Ibáñez, M., Grimalt, S., 2008. Investigation of pesticide metabolites in food and water by LC-TOF-MS. *TrAC Trends Anal. Chem.* 27, 862–872. <https://doi.org/10.1016/j.trac.2008.08.011>.
- Hintze, S., Glauser, G., Hunkeler, D., 2020. Influence of surface water – groundwater interactions on the spatial distribution of pesticide metabolites in groundwater. *Sci. Total Environ.* 733, 139109. <https://doi.org/10.1016/j.scitotenv.2020.139109>.
- Infoclimat [WWW Document], 2020. URL <https://www.infoclimat.fr/climatologie/annee/2019/buhl-lorraine/valeurs/000C0.html> (accessed 11.11.20).
- Jablonowski, N.D., Schäffer, A., Buraue, P., 2011. Still present after all these years: persistence plus potential toxicity raise questions about the use of atrazine. *Environ. Sci. Pollut. Res.* 18, 328–331. <https://doi.org/10.1007/s11356-010-0431-y>.
- Kaur, H., Garg, H., 2014. Pesticides: environmental impacts and management strategies. *Pestic. - Toxic Asp.* <https://doi.org/10.5772/57399>.
- Kellogg, R.L., Nehring, R.F., Grube, A., Goss, D.W., Plotkin, S., 2002. Environmental indicators of pesticide leaching and runoff from farm fields. *Agric. Product.*, 213–256. https://doi.org/10.1007/978-1-4615-0851-9_9.
- Kim, K.H., Kabir, E., Jahan, S.A., 2017. Exposure to pesticides and the associated human health effects. *Sci. Total Environ.* 575, 525–535. <https://doi.org/10.1016/j.scitotenv.2016.09.009>.
- Lazartigues, A., Banas, D., Feidt, C., Brun-Bellut, J., Thomas, M., 2012. Pesticide pressure and fish farming in barrage pond in Northeastern France part I: site characterization and water quality. *Environ. Sci. Pollut. Res.* 19, 2802–2812. <https://doi.org/10.1007/s11356-012-0784-5>.
- Lazartigues, A., Thomas, M., Cren-Olivé, C., Brun-Bellut, J., Le Roux, Y., Banas, D., Feidt, C., 2013. Pesticide pressure and fish farming in barrage pond in Northeastern France. Part II: residues of 13 pesticides in water, sediments, edible fish and their relationships. *Environ. Sci. Pollut. Res.* 20, 117–125. <https://doi.org/10.1007/s11356-012-1167-7>.
- Lefrancq, M., Jadas-Hécart, A., La Jeunesse, I., Landry, D., Payraudeau, S., 2017. High frequency monitoring of pesticides in runoff water to improve understanding of their transport and environmental impacts. *Sci. Total Environ.* 587–588, 75–86. <https://doi.org/10.1016/j.scitotenv.2017.02.022>.
- Lewis, K.A., Tzilivakis, J., Warner, D.J., Green, A., 2016. An international database for pesticide risk assessments and management. *Hum. Ecol. Risk Assess.* 22, 1050–1064. <https://doi.org/10.1080/10807039.2015.1133242>.
- van Lexmond, M.B., Bonmatin, J.M., Goulson, D., Noone, D.A., 2015. Worldwide integrated assessment on systemic pesticides global collapse of the entomofauna: exploring the role of systemic insecticides. *Environ. Sci. Pollut. Res.* 22, 1–4. <https://doi.org/10.1007/s11356-014-3220-1>.
- Lorenz, S., Rasmussen, J.J., Süß, A., Kalettka, T., Golla, B., Horney, P., Stähler, M., Hommel, B., Schäfer, R.B., 2017. Specifics and challenges of assessing exposure and effects of pesticides in small water bodies. *Hydrobiologia* 793, 213–224. <https://doi.org/10.1007/s10750-016-2973-6>.
- Mantzou, N., Hela, D., Karakitsou, A., Antonopoulou, M., Konstantinou, I., 2016a. Dissipation and runoff transport of metazachlor herbicide in rapeseed cultivated and uncultivated plots in field conditions. *Environ. Sci. Pollut. Res.* 23, 20517–20527. <https://doi.org/10.1007/s11356-016-7233-9>.
- Mantzou, N., Hela, D., Karakitsou, A., Antonopoulou, M., Konstantinou, I., 2016b. Dissipation and runoff transport of metazachlor herbicide in rapeseed cultivated and uncultivated plots in field conditions. *Environ. Sci. Pollut. Res.* 23, 20517–20527. <https://doi.org/10.1007/s11356-016-7233-9>.
- Marín-Benito, J.M., Carpio, M.J., Mamy, L., Andrades, M.S., Sánchez-Martín, M.J., Rodríguez-Cruz, M.S., 2020. Field measurement and modelling of chlorotoluron and flufenacet persistence in unamended and amended soils. *Sci. Total Environ.* 725, 138374. <https://doi.org/10.1016/j.scitotenv.2020.138374>.
- Melin, J., Guillon, A., Enault, J., Esperanza, M., Dauchy, X., Bouchonnet, S., 2020. How to select relevant metabolites based on available data for parent molecules: case of neonicotinoids, carbamates, phenylpyrazoles and organophosphorus compounds in French water resources. *Environ. Pollut.* 265, 114992. <https://doi.org/10.1016/j.envpol.2020.114992>.
- Meyer, J.L., Strayer, D.L., Wallace, J.B., Eggert, S.L., Helfman, G.S., Leonard, N.E., Judy, L., Strayer, D.L., Wallace, J.B., Eggert, S.L., Helfman, G.S., Leonard, N.E., 2007. The contribution of headwater streams to biodiversity in river networks 43. doi:<https://doi.org/10.1111/j.1752-1688.2007.00008.x>.
- Milan, M., Ferrero, A., Letey, M., De Palo, F., Vidotto, F., 2013. Effect of buffer strips and soil texture on runoff losses of flufenacet and isoxaflutole from maize fields. *J. Environ. Sci. Health, Part B* 48, 1021–1033. <https://doi.org/10.1080/03601234.2013.824239>.
- Moschet, C., Wittmer, I., Simovic, J., Junghans, M., Piazzoli, A., Singer, H., Stamm, C., Leu, C., Hollender, J., 2014. How a complete pesticide screening changes the assessment of surface water quality. *Environ. Sci. Technol.* 48 (10), 5423–5432. <https://doi.org/10.1021/es500371t>.
- Muhammad, M., Rasul Jan, M., Shah, J., Ara, B., 2019. Determination of Isoproturon in environmental samples using the QuEChERS extraction-spectrofluorimetric method. *Environ. Toxicol. Chem.* 38, 2614–2620. <https://doi.org/10.1002/etc.4589>.
- Nagy, K., Duca, R.C., Lovas, S., Creta, M., Scheepers, P.T., Godderis, L., Ádám, B., 2020. Systematic review of comparative studies assessing the toxicity of pesticide active ingredients and their product formulations. *Environ. Res.* 181, 108926. <https://doi.org/10.1016/j.envres.2019.108926>.
- Osborne, J.L., 2012. *Bumblebees and pesticides*. *Nature* 491, 3–5.
- Özkara, A., Akyil, D., Konuk, M., 2016. Pesticides, Environmental Pollution, and Health. *Environ. Heal. Risk - Hazard. Factors to Living Species* 3–28. <https://doi.org/10.5772/63094>.
- Pereira, A.S., Daam, M.A., Cerejeira, M.J., 2017. Evaluation of FOCUS surface water pesticide concentration predictions and risk assessment of field-measured pesticide mixtures—a crop-based approach under Mediterranean conditions. *Environ. Sci. Pollut. Res.* 24 (21), 17394–17406. <https://doi.org/10.1007/s11356-017-9393-7>.
- Phillips, P.J., Wall, G.R., Thurman, E.M., Eckhardt, D.A., Vanhoesen, J., 1999. Metolachlor and its metabolites in tile drain and stream runoff in the canajoharie creek watershed. *Environ. Sci. Technol.* 33, 3531–3537. <https://doi.org/10.1021/es9811997>.
- Picó, Y., Barceló, D., 2015. Transformation products of emerging contaminants in the environment and high-resolution mass spectrometry: a new horizon. *Anal. Bioanal. Chem.* 407, 6257–6273. <https://doi.org/10.1007/s00216-015-8739-6>.
- Pimentel, D., 2005. Environmental and economic costs of the application of pesticides primarily in the United States. *Environ. Dev. Sustain.* 7, 229–252. <https://doi.org/10.1007/s10668-005-7314-2>.
- Portail Substances Chimiques [WWW Document], 2009. URL www.ineris.fr/substances.
- Raina, R., Etter, M.L., Buehler, K., Starks, K., Yowin, Y., 2011. Phenoxyacid herbicides in stormwater retention ponds: urban inputs. *Am. J. Anal. Chem.* 02, 962–970. <https://doi.org/10.4236/ajac.2011.28112>.
- Rasmussen, J.J., McKnight, U.S., Loinaz, M.C., Thomsen, N.I., Olsson, M.E., Bjerg, P.L., Binning, P.J., Kronvang, B., 2013. A catchment scale evaluation of multiple stressor effects in headwater streams. *Sci. Total Environ.* 442, 420–431. <https://doi.org/10.1016/j.scitotenv.2012.10.076>.
- Reemtsma, T., Alder, L., Banasiak, U., 2013. Emerging pesticide metabolites in groundwater and surface water as determined by the application of a multimethod for 150 pesticide metabolites. *Water Res.* 47 (15), 5535–5545. <https://doi.org/10.1016/j.watres.2013.06.031>.
- Relyea, R.A., 2009. A cocktail of contaminants: how mixtures of pesticides at low concentrations affect aquatic communities. *Oecologia* 159, 363–376. <https://doi.org/10.1007/s00442-008-1213-9>.
- Rizzati, V., Briand, O., Guillon, H., Gamet-Payastre, L., 2016. Effects of pesticide mixtures in human and animal models: an update of the recent literature. *Chem. Biol. Interact.* 254, 231–246. <https://doi.org/10.1016/j.cbi.2016.06.003>.
- Rousis, N.I., Bade, R., Bijlsma, L., Zuccato, E., Sancho, J.V., 2017. Monitoring a large number of pesticides and transformation products in water samples from Spain and Italy. *Environ. Res.* 156, 31–38. <https://doi.org/10.1016/j.envres.2017.03.013>.
- Sarmah, A.K., Sabadie, J., 2002. Hydrolysis of sulfonylurea herbicides in soils and aqueous solutions: a review. *J. Agric. Food Chem.* 50, 6253–6265.
- Schreiner, V.C., Szöcs, E., Bhowmik, A.K., Vijver, M.G., Schäfer, R.B., 2016. Pesticide mixtures in streams of several European countries and the USA. *Sci. Total Environ.* 573, 680–689. <https://doi.org/10.1016/j.scitotenv.2016.08.163>.
- Schuhmann, A., Klammler, G., Weiss, S., Gans, O., Fank, J., Haberhauer, G., Gerzabek, M.H., 2019. Degradation and leaching of bentazone, terbutylazine and S-metolachlor and some of their metabolites: a long-term lysimeter experiment. *Plant Soil Environ.* 65, 273–281. <https://doi.org/10.17221/803/2018-PSE>.
- Sharma, A., Kumar, V., Shahzad, B., Tanveer, M., Sidhu, G.P.S., Handa, N., Kohli, S.K., Yadav, P., Bali, A.S., Parihar, R.D., Dar, O.I., Singh, K., Jasrotia, S., Bakshi, P., Ramakrishnan, M., Kumar, S., Bhardwaj, R., Thukral, A.K., 2019. Worldwide pesticide usage and its impacts on ecosystem. *SN Appl. Sci.* 1, 1446. <https://doi.org/10.1007/s42452-019-1485-1>.
- Smiley Jr., P.C., King, K.W., Fausey, N.R., 2014. Agriculture, ecosystems and environment annual and seasonal differences in pesticide mixtures within channelized agricultural headwater streams in central Ohio. *Agriculture, Ecosyst. Environ.* 193, 83–95. <https://doi.org/10.1016/j.agee.2014.04.022>.
- Stehle, S., Elsaesser, D., Gregoire, C., Imfeld, G., Niehaus, E., Passepote, E., Payraudeau, S., Schäfer, R.B., Tournébeize, J., Schulz, R., 2011. Pesticide risk mitigation by vegetated treatment systems: a meta-analysis. *J. Environ. Qual.* 40, 1068–1080. <https://doi.org/10.2134/jeq2010.0510>.
- Strahler, A., 1952. Hypsometric (area-altitude) analysis of erosional topography. *Bull. Geol. Soc. Am.* 63, 1117–1142. [https://doi.org/10.1130/0016-7606\(1952\)63](https://doi.org/10.1130/0016-7606(1952)63).
- Szöcs, E., Brinke, M., Karaoglan, B., Schäfer, R.B., 2017. Large scale risks from agricultural pesticides in small streams. *Environ. Sci. Technol.* 51, 7378–7385. <https://doi.org/10.1021/acs.est.7b00933>.
- Tauchnitz, N., Kurzius, F., Rupp, H., Schmidt, G., Hauser, B., Schrödter, M., Meissner, R., 2020. Assessment of pesticide inputs into surface waters by agricultural and urban sources – a case study in the Querne/Weida catchment, central Germany. *Environ. Pollut.* 267, 115186. <https://doi.org/10.1016/j.envpol.2020.115186>.
- Tournébeize, J., Gramaglia, C., Birmant, F., Bouarfa, S., Chaumont, C., Vincent, B., 2012. Co-design of constructed wetlands to mitigate pesticide pollution in a drained catch-basin: a solution to improve groundwater quality. *Irrig. Drain.* 61, 75–86.
- Tournébeize, J., Passepote, E., Chaumont, C., Fesneau, C., Guenne, A., Vincent, B., 2013. Pesticide de-contamination of surface waters as a wetland ecosystem service in agricultural landscapes. *Ecol. Eng.* 56, 51–59. <https://doi.org/10.1016/j.ecoleng.2012.06.001>.

- Tournebize, J., Chaumont, C., Mander, Ü., 2017. Implications for constructed wetlands to mitigate nitrate and pesticide pollution in agricultural drained watersheds. *Ecol. Eng.* 103, 415–425. <https://doi.org/10.1016/j.ecoleng.2016.02.014>.
- Ulrich, U., Hörmann, G., Unger, M., Pfannerstill, M., Steinmann, F., Fohrer, N., 2018. Lentic small water bodies: variability of pesticide transport and transformation patterns. *Sci. Total Environ.* 618, 26–38. <https://doi.org/10.1016/j.scitotenv.2017.11.032>.
- Vallée, R., Dousset, S., Billet, D., Benoit, M., 2014. Sorption of selected pesticides on soils, sediment and straw from a constructed agricultural drainage ditch or pond. *Environ. Sci. Pollut. Res.* 21, 4895–4905. <https://doi.org/10.1007/s11356-013-1840-5>.
- Vallée, R., Dousset, S., Schott, F.X., Pallez, C., Ortar, A., Cherrier, R., Munoz, J.F., Benoît, M., 2015. Do constructed wetlands in grass strips reduce water contamination from drained fields? *Environ. Pollut.* 207, 365–373. <https://doi.org/10.1016/j.envpol.2015.09.027>.
- Vidali, M., 2001. *Bioremediation. An overview **. *Pure Appl. Chem.* 73, 1163–1172.
- Vryzas, Z., 2018. Pesticide fate in soil-sediment-water environment in relation to contamination preventing actions. *Curr. Opin. Environ. Sci. Heal.* 4, 5–9. <https://doi.org/10.1016/j.coesh.2018.03.001>.
- Vymazal, J., Březinová, T., 2015. The use of constructed wetlands for removal of pesticides from agricultural runoff and drainage: a review. *Environ. Int.* 75, 11–20. <https://doi.org/10.1016/j.envint.2014.10.026>.
- Weber, G., Christmann, N., Thiery, A., Martens, D., Kubiniok, J., 2018. Pesticides in agricultural headwater streams in southwestern Germany and effects on macroinvertebrate populations. *Sci. Total Environ.* 619–620, 638–648. <https://doi.org/10.1016/j.scitotenv.2017.11.155>.
- Willkommen, S., Pfannerstill, M., Ulrich, U., Guse, B., Fohrer, N., 2019. How weather conditions and physico-chemical properties control the leaching of flufenacet, diflufenican, and pendimethalin in a tile-drained landscape. *Agric. Ecosyst. Environ.* 278, 107–116.
- Zablotowicz, R.M., Locke, M.A., Lerch, R.N., Knight, S.S., 2004. Dynamics of Herbicide Concentrations in Mississippi Delta Oxbow Lakes and the Role of Planktonic Microorganisms in Herbicide Metabolism 134–149. doi:<https://doi.org/10.1021/bk-2004-0877.ch010>.
- Zivan, O., Segal-Rosenheimer, M., Dubowski, Y., 2016. Airborne organophosphate pesticides drift in Mediterranean climate: the importance of secondary drift. *Atmos. Environ.* 127, 155–162. <https://doi.org/10.1016/j.atmosenv.2015.12.003>.