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Factors driving the distribution of an amphibian community in stormwater ponds: a study case in the agricultural plain of Bas-Rhin, France

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Abstract

During road construction, stormwater ponds are created to address sanitation, water treatment and the containment of any accidental pollution issues. These environments are not intended to be habitats, so exclosure measures (e.g. fences, barriers) are implemented to prevent animals to gain access to them. However, the modification of the natural landscape for human needs resulted in the disappearance of most wetlands. Our hypothesis was that depending on the water pollutant concentrations, the stormwater water ponds could serve as refuge habitat for wetland species like amphibians. Thus, we evaluated the suitability of stormwater ponds as a habitat for amphibians by studying 82 such structures in the agricultural plain of Bas-Rhin. The proportion of stormwater ponds hosting amphibians and specific species abundances and richness were quantified as community parameters. They were explained using factors such as pond design (e.g. size, depth, slopes), road-induced pollutants, land use and exclosure measures. Significance of these factors was assessed by boosted regression tree models. Species-dependent effects were studied using detrended correspondence analysis. Amphibians were found in 84% of stormwater ponds, with an average of 19.51 adults and 2.44 species per pond. We found 83% of species previously detected in Bas-Rhin, including rare and protected ones. Neither exclosure measures nor pollutant concentrations were correlated with community parameters. The best explanatory factors were land use and pond design. For ponds with pollutant concentrations similar to those quantified in this study, we recommend reallocating the efforts made for exclosure to improve pond design and to the creation of semi-natural ponds as additional compensatory measures. Design of stormwater ponds should be systematically validated by a herpetologist to avoid mortal traps. Ponds should be large and have a permanent minimum water level even in droughts.

Keywords Refuge habitat · Retention ponds · Farmlands · Semi-natural habitats · Modified landscape · Pollution

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Introduction

Behind overexploitation, agriculture is the next-leading cause of current biodiversity decline and affects all taxonomic groups (Butchart et al. 2010; Maxwell et al. 2016). The habitat loss induced by land consolidation leads to a great number of local population extinctions through the modification of the use and layout of land (Fischer and Lindenmayer 2007). The creation of vast monocultural farmlands causes the fragmentation of residual seminatural elements and has negative impacts on species richness, population abundances, growth rate and distribution, trophic chain length, breeding and dispersal success (Benton et al. 2003; Fahrig 2017). Roads are another cause of biodiversity loss (Forman and Alexander 1998; Maxwell et al. 2016). Their negative impacts are observed

in the road-effect zone and are numerous (Richard et al. 2000; Van Der Ree et al. 2011). For example, roads decrease habitat sizes and quality, affect the life history traits and population characteristics of species and increase direct and indirect mortality rates (Redon (de et al. 2015; Spellerberg 1998). They also contribute to landscape fragmentation through the linearisation of landscape (Holderegger and Di Giulio 2010). These negative effects have numerous adaptations and evolutionary consequences such as changes in vocal pitch and activities in response to traffic noise in birds and amphibians species (Lengagne 2008; Slabbekoorn and Peet 2003), higher pollutant tolerance (Brady 2012) or change of bird wing length in response to roadkill (Brown and Bomberger Brown 2013; Kiang 1982). These consequences should be taken into account on impact studies so that the adaptation would not be underestimated and protection measures would be effective (Brady and Richardson 2017). Together, intensive agricultural activities and roads form highly modified landscapes in which few natural and semi-natural elements remain, where biodiversity can be very low and impacts from both roads and agricultural activities are present (Donald et al. 2001; Foley et al. 2005; Stoate et al. 2001).

Wetlands are particularly impacted by anthropogenic activities like roads and agriculture. At least 64% of them have disappeared over the past century, and little data is available for the conservation status of what remains. There is a continuing decline of wetlands, accelerated by urban expansion, agricultural intensification, land consolidation and the construction of motorways (Davidson 2014). Thus, they are internationally protected as they provide important ecosystemic services such as biomass and resource production, pollutant and climate regulation, flood abatement and erosion decrease (Bolund and Hunhammar 1999; Russi et al. 2013; Zedler 2003). Moreover, one third of non-marine vertebrate species inhabit wetlands, giving them a high ecological value (Dudgeon et al. 2006). In highly modified landscapes such as farmlands fragmented by roads, wetland species can survive habitat loss by colonising the poor-quality, isolated remains of semi-natural habitats (McKinney 2006; Sinsch et al. 2012).

To prevent some of the alteration of aquatic systems, wetlands and other habitats in modified landscapes, artificial ponds called stormwater ponds are built. Located next to roads, urban areas or industrial ones, they are designed to collect, stock and decontaminate runoff otherwise released into a nearby stream. These structures protect outside water from chronic and exceptional contamination (Karouna-Renier and Sparling 2001; Scher and Thièry 2005). They also aim to prevent and control flooding events and to store channel and canal volume (EPA 2009). They are required by European legislation

for certain surfaces and associated water volumes and for new or existing infrastructures (Le Viol et al. 2009). There are several types of stormwater ponds (such as micropool and stormwater wetlands), two of which are retention ponds, which permanently maintain a pool of polluted water throughout the year (permanent hydroperiod), and detention ponds, which hold clean water for a short period of time before it enters the stream (temporary hydroperiod). While detention ponds are usually a simple hole, retention ponds are designed with additional characteristics such as waterflow regulation structures, hydrocarbon separators and waterproof covers (EPA 2009). Retention ponds can be underground or open-air structures.

Although roads contribute to wetland loss (Van der Ree et al. 2015), amphibian roadkill (Fahrig et al. 1995; Elzanowski et al. 2009) and the invasion of nonindigenous species (Jodoin et al. 2008), they may also provide alternative semi-natural habitats for wetland species. Like natural wetlands, stormwater ponds can provide ecosystem services and are inhabited by flora, birds, invertebrates, snakes and fishes (Ackley and Meylan 2010; Bishop et al. 2000; Karouna-Renier and Sparling 2001; Le Viol et al. 2009; Moore and Hunt 2012). The biodiversity of stormwater ponds can be equivalent to that of semi-natural wetlands (Hassall and Anderson 2015), and these habitats can be inhabited by rare and protected species (Le Viol et al. 2012). A better understanding of their ecological function on a large scale and in highly modified landscapes is however needed (Brand and Snodgrass 2010; Scheffers and Paszkowski 2013). The small number of studies focusing on stormwater pond biodiversity is relatively recent (Bishop et al. 2000). This scarcity of literature could be explained by disinterest of pond managers for stormwater pond biodiversity (Hassall and Anderson 2015) or by their desire to avoid colonisation by amphibians as stormwater pond water can be polluted (Massal et al. 2007; Snodgrass et al. 2008), and therefore, these ponds could be ecological traps (Battin 2004). Indeed, runoff collected by stormwater ponds can contain agents toxic for aquatic fauna. They contain heavy metals (Wik et al. 2008), polycyclic aromatic hydrocarbons (PAHs) (Neff et al. 2005) and chlorides (Gallagher et al. 2014). Water column pollutants can vary through time, an example being the high increase in chloride concentrations in late winter due to the use of road salt as deicing agent (Collins and Russell 2009). Overall pollutant concentrations can quickly increase after storm rainfall on the road surface. However, only a small proportion of these pollutants reach stormwater ponds as the majority evaporates, stays on road surface or is degraded by sun exposure (Pagotto 1999). Pollutant concentration of the sediment is more stable over time, and high

concentrations are accumulated, especially if sediment is rarely removed. This can be a non-negligible threat for species that hide or winter in sediment and also for their predators because of the bioaccumulation effect (Bishop et al. 1995; Brand et al. 2010). Concentrations of pollutants can also vary according to the landscape. Maximum threshold levels of nitrites and nitrates from agricultural chemicals can be recorded in highly modified landscapes such as farmlands (Hayes et al. 2006). Thus, despite evidence of the habitat potentiality of stormwater ponds provided by a number of studies (Brown et al. 2012; Scheffers and Paszkowski 2013), the stormwater ponds could still be ecological traps. Therefore, some countries including France demand, at great expense, the installation of exclosure measures such as fences, walls, cattle grids or handrails (Morand and Carsignol 2019).

Amphibians are wetland species which can be found in stormwater ponds (McCarthy and Lathrop 2011; Sievers et al. 2019). One third of them is threatened with extinction all over the world, due to several causes such as diseases and habitat loss (Arntzen et al. 2017; Becker et al. 2007; Dudgeon et al. 2006; Eterovick et al. 2005; Mazerolle et al. 2005). This last threat is particularly serious as the habitat needs of pondbreeding amphibians vary greatly according to the biological traits and biodemographic strategies of each species (Van Buskirk 2005). Amphibians are also vulnerable to the impact of road traffic due to their immobility facing motor vehicles (Gibbs and Shriver 2005; Mazerolle et al. 2005) and all their population-scale movements (Joly 2019). Indeed, the amphibian mass migration concerning most species can reach a distance of 15 km for anurans and occurs twice a year (Beebee 1996; Sinsch 1990). Therefore, it leads amphibians to cross many roads. In highly modified landscapes where only few wetlands remain, it can be difficult for every amphibian species to find suitable ponds (Hamer and McDonnell 2008). By varying in location, shape, design, function and management, stormwater ponds could serve as alternative habitat for some amphibian species. However, there are very few studies focusing on amphibian community in stormwater ponds. Further research is required to understand the factors, especially pollutants, driving these communities and to find ways of improving the habitat quality of stormwater ponds for amphibians (Brand and Snodgrass 2010; Scheffers and Paszkowski 2013; Scher and Thièry 2005). With sufficient concentration, pollutants can induce sterility and external and internal abnormalities in amphibians, stunt their growth and increase difficulties during metamorphosis (Bryer et al. 2006; Egea-Serrano et al. 2012; Sievers et al. 2018; Wagner et al. 2014). Concurrent to a bad stormwater pond design which can trap amphibians in dead end, pollutants could make stormwater pond ecological traps and therefore toll the bells for these species in modified landscape where few wetlands remain (Clevenot et al. 2018).

Research objectives

This study focuses on the agricultural plain of Bas-Rhin (Bas-Rhin 4755 km², 23 inhabitants per km²), a highly modified landscape composed of vast monoculture areas and roads. We performed a one-breeding season survey of the Anura and Caudata amphibian communities found in stormwater ponds located along public roads with moderate traffic.

Our objectives were (1) to qualify and quantify the amphibian community biodiversity and species richness of stormwater ponds and (2) to identify and quantify factors driving the constitution parameters of amphibian communities of stormwater ponds (species and density of population).

We expected that firstly, a significant proportion of stormwater ponds hosted amphibians and that interspecies differences existed, following factors like pond design (size, depth, slopes, etc.) and surrounding land use, and secondly, a negative influence of pollutants, and particularly chlorides, nitrites and nitrates, and no effect of exclosure measures on amphibian communities.

Material and methods

Protocol

Studied area

The Department of Bas-Rhin (Alsace, Grand Est, France; Fig. 1) has a semi-continental climate with an average temperature of 10.4 °C and an atmospheric temperature range of 30 °C. The annual precipitation is about 700 mm per year, and the average altitude is 150 m. The road density is 1.9 km/km², with more than 3654 km of public roads with moderate traffic and 240 km of major roads (mainly private large road with high traffic). In the western part of Bas-Rhin, a range of low mountains forms the geomorphological unit of the Vosges. Its flora is mainly dominated by Picea, fir, common beech and oak. In the East, the Rhine River is bordered by many wetlands that are certified by the Ramsar convention (no. FR7200025) and protected by the Natura 2000 European network (no. FR4211811, no. FR4211810). These wetlands make up less than 1% of the Bas-Rhin area, the successive building of dykes along the Rhine (1842–1876, 1928–1959) having substantially diminished the area of wetlands in this region. The intensification of agricultural practices also decreased their surfaces through land consolidation. This is illustrated by the courses of the Rhine tributaries from the Vosges, which cross a large agricultural plain landscape that is currently dominated by wheat and maize crops. There is no precise inventory of wetland loss in Bas-Rhin. On a national scale. France has lost over 67% of total wetland area since the



Fig. 1 Bas-Rhin (Alsace, Eastern France). Map, left: location of studied stormwater ponds (orange). The stormwater ponds were grouped in clusters (purple), forming linear networks of ponds. Map, centre: motorways (wide black line), primary roads (thin black line) and

beginning of the twentieth century (Ximenès et al. 2007). The studied area hosts 18 amphibian species.

Studied ponds

We monitored 82 of the 84 open-air stormwater ponds of the Bas-Rhin public roads with moderate traffic (Fig. 1) and could not safely access the two remaining ponds. Traffic on those public roads is mainly below 10,000 vehicles/day. Stormwater ponds were mainly located in the agricultural plain. Six were located in the Vosges Mountains, and only one was in the Ramsar area near the Rhine (Fig. 1). Stormwater ponds differed substantially in terms of age (from less than 1 year old to 24 years old), volume (from 50 to 7000 m³) and design (e.g. nature of exclosure measures, type of substrate, angle of banks). Most (73) were retention ponds (permanent hydroperiod).

Sampling design

The presence of amphibians was checked in all the studied stormwater ponds. Three one-night field sessions were carried out for each pond between March and July 2016 to observe early- and late-breeding season amphibian species. Field groups included up to ten geographically close ponds. Each pond in a given field group was inspected during the same night. The order in which field groups were checked was

secondary roads (red line) forming fragmented landscapes. Map, right: location of wetlands (blue) and Ramsar-certified wetlands (purple). Most are located close to the Rhine River in the eastern part of the territory

randomly chosen during the first field session and replicated in the following sessions.

The sampling protocol was adapted from the "POP Amphibiens communauté" (Barrioz and Miaud 2016). Sampling began at dusk. For each pond, sampling began by 5 min of listening to estimate the species richness and the number of adult males. Visual sampling was then performed with headlamps and flashlights. The sampling was stopped after two patrols around each pond in order to have an equivalent sampling effort between ponds. Indeed, most of them differed substantially in length but only slightly in width. The number of individuals, amplexus, eggs and larvae was noted for each species. The number of floating carcasses and any visible morphological abnormalities on adults were also recorded. To avoid any mistakes due to incorrect identification, edible frogs (Pelophylax kl. esculentus) and pool frogs (Pelophylax lessonae) were gathered in a "green frogs" group. We used the identification key of Miaud and Muratet (2004). The experimental protocol was authorised by the 5/6/2016 nominative authorisation of the French Environmental Code and followed EU Directive 2010/63/EU guidelines for animal experiments.

Environmental factors

Several factors were also checked and classified into factor groups (Table 1). Environmental factors (e.g. pH, water and air temperature, visibility) were recorded immediately after

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Group	Factors	Min/class 1	Max/class 2	Median/class 3	IQ range/ class 4	Var. type	Measurement type
Pond design	Volume (m ³)	50	7000	539	667	Quantitative	Data from road
	Depth (m)	0.3	2.5	1	0.25	Quantitative	manager Data from road
	Age (years)	0	35	8	15	Quantitative	manager Data from road
	Large fauna fence (12-cm mesh)	No	Yes			Qualitative	manager Visual
	Small fauna fence (6.5-mm mesh)	No	Yes, but without hent ton	Yes, with bent	top	Qualitative	Visual
	Small fauna fence condition ^a	Intact	1-2 holes > 3 cm	3+ holes >3 cm		Qualitative	Visual
	Service gate condition ^a	Intact	With holes $> 3 \text{ cm}$			Qualitative	Visual
	Cattle grid under service gate	No	Yes			Qualitative	Visual
	Concrete wall	No	Yes			Qualitative	Visual
	Pond water tightness/permanent water	No	Yes			Qualitative	Visual
	Pond enclosure by vegetation/stagnant water	No	Yes			Qualitative	Visual
	Distance to the main road	<3 m	3-10 m	> 10 m		Qualitative	Laser telemeter
	Service bank	No	Yes			Qualitative	Visual
	Length of banks	<3 m	>3 m			Qualitative	Laser telemeter
	Bank soil	Natural	Artificial			Qualitative	Visual
	Bank steepness ^a	Steep (U)	Gentle (V)			Qualitative	Visual
	Hydrocarbon separator (upstream)	No	Yes			Qualitative	Visual
	Hydrocarbon separator (downstream)	No	Yes			Qualitative	Visual
	Service ladder	No	Yes			Qualitative	Visual
	Sunlight	Low	High			Qualitative	Visual
Biotic factors	Vegetative fragments in water (as food)	None	Few	Many		Qualitative	Visual
	Branches (for fastening eggs)	None	Few	Many		Qualitative	Visual
	Helophytes (for fastening eggs and as shelters)	None	Few	Many		Qualitative	Visual
	Helophyte types	None	Phragmites	Molinia	Both	Qualitative	Visual
	Algae and hydrophytes (for fastening eggs and	None	Few	Many		Qualitative	Visual
	as shelters)	:	;			;	
	Odonata larvae (as predators)	No	Yes			Qualitative	Visual
	Leaves (for food and sunlight reduction)	None	Few	Many		Qualitative	Visual
	Fishes (as predators)	No	Yes			Qualitative	Visual
	Snakes (as predators)	No	Yes			Qualitative	Visual
	Surrounding species richness	3	11	5	2	Quantitative	Odonat (2017)
Immediate environmental factors	Waste on bank ^a	No	Yes			Qualitative	Visual
	Waste on water ^a	No	Yes			Qualitative	Visual
	Luminosity (as visibility control) ^a	Low (has been	High			Qualitative	Visual
		regrouped)				:	
	Rain (as visibility control) ^a	No	Yes			Qualitative	Visual
	Wind (as visibility control) ^a	Low	High			Qualitative	Visual
	Turbidity (as visibility control)	Transparent	Cloudy	Opaque		Qualitative	Visual
	HAP on surface"	No	Yes	:		Qualitative	Visual
	Visibility (as visibility control)	<3 m	3-10 m	>10 m		Qualitative	Visual
	Air temperature (°C)	9	24	16	8	Quantitative	Thermometer

Table 1 (continued)							
Group	Factors	Min/class 1	Max/class 2	Median/class 3	IQ range/ class 4	Var. type	Measurement type
	Water temperature (°C) pH	11 5.8	19.5 12.35	16.33 9.05	2 1.3	Quantitative Quantitative	Thermometer pH meter
Local scale (500-m-wide buffer)/large scale	Hedgerows (ha)	0/14.83	5.28/96.72	0.15/50.19	0.84/18.65	Quantitative	GIS
(5000-m-wide buffer)	Roads (ha)	0/43.69	10.40/263.80	3.12/104.37	4.15/47.78	Quantitative	GIS
	Railways (ha)	0/0	3.63/177.49	0/22.31	0.54/23.48	Quantitative	GIS
	Meadows (ha)	0/329.77	47.16/2538.59	11.04/819.07	17.14/500.97	Quantitative	GIS
	Vines (ha)	0/0	35.50/1926.15	0/0	0/5.46	Quantitative	GIS
	Urban (ha)	0/264.60	65.90/3761.17	9.32/929.51	18.69/659.77	Quantitative	GIS
	Plantations (ha)	0/0	6.00/274.36	0/7.87	0/31.01	Quantitative	GIS
	Wetlands (ha)	0/4.19	5.20/125.11	0/38.49	1.49/43.77	Quantitative	GIS
	Grasslands (ha)	0/0	5.05/211.21	0/12.22	0/23.21	Quantitative	GIS
	Moors (ha)	0/0.96	7.78/343.89	0/28.42	< 0.01/59.01	Quantitative	GIS
	Ponds (ha)	0/0.55	7.71/132.56	0/4.79	0.10/7.06	Quantitative	GIS
	Annual crops (ha)	0/0	77.26/6269.13	23.28/3223.69	43.60/2188.04	Quantitative	GIS
	Softwood forests (ha)	0/0	24.97/2816.30	0/23.42	0/271.32	Quantitative	GIS
	Mixed forests (ha)	0/0	29.77/1759.29	0/64.57	0/213.64	Quantitative	GIS
	Deciduous forests (ha)	0/80.92	75.23/4925.78	0.81/1128.07	10.90/1066.59	Quantitative	GIS
Pollutants (water/sediment)	(μS/cm) conductivity	64/-	937/-	162.6/-	140.8/-	Quantitative	See SM Table
	(mg/l) salinity	-/0	420/-	-/0	127.92/-	Quantitative	See SM Table
	hd	5.8/-	12.35/-	9.05/-	1.3/-	Quantitative	See SM Table
	(mg/l) <i>chloride</i>	1.7/-	107.6/-	8.9/-	15.6/-	Quantitative	See SM Table
	(mg/l) suspended materials	-/0	260/-	11.45/-	19.07/-	Quantitative	See SM Table
	(mg/l) nitrates	-/0	21.3/-	0.77/-	1.25/-	Quantitative	See SM Table
	(mg/l) <i>nitrites</i>	-/0	0.21/-	0.03/-	0.07/-	Quantitative	See SM Table
	(mg/l) PAH (mg/kg SM)	-/0.0/0	0.68/15.51	0/0.53/-	0/10.90	Quantitative	See SM Table
	(mg/l) phosphorus	-/0	0.4/-	-/0	0.1/-	Quantitative	See SM Table
	(mg/l) mercury (mg/kg SM)	0/0.01	0/0.34	0/0.13	0/0.06	Quantitative	See SM Table
	(µg/l) arsenic (mg/kg SM)	0 < 0.01	6.7/16.4	0.7/7.70	1.39/8.99	Quantitative	See SM Table
	(µg/l) cadmium (mg/kg SM)	0 < 0.01	0.02/95	< 0.01/0.5	< 0.01/0.9	Quantitative	See SM Table
	(µg/l) chromium (mg/kg MS)	0/0.1	18/95	1.67/41.6	1.32/69.6	Quantitative	See SM Table
	(µg/l) copper (mg/kg SM)	1.1 < 0.01	23.1/390	6.77/84.5	6.27/212.5	Quantitative	See Tab SM Table
	(µg/l) nickel (mg/kg SM)	0/0.1	13.4/45.4	0/20.6	0/29	Quantitative	See SM Table
	(µg/l) lead (mg/kg SM)	0/0.12	16.9/118	0.74/40	1.7/69.70	Quantitative	See SM Table
	(µg/l) zinc (mg/kg SM)	0/0.2	99/3085.2	25.5/685.4	25.25/1214.2	Quantitative	See SM Table
	Fluoranthene (µg/kg SM)	-/0.05	-/1748	-/1.682	-/414.42	Quantitative	See SM Table
	Benzo(b)fluoranthene (µg/kg SM)	-/0.06	-/913	-/1.37	-/235.01	Quantitative	See SM Table
	Benzo(a)pyrene (μg/kg SM)	-/0.05	-/793	-/23	-/228.78	Quantitative	See SM Table
	Sum 7 PCB (µg/kg SM)	-/0.04	-/29.5	-/0.10	-/0.01	Quantitative	See SM Table
Factors with sufficient sample size that were ret	tained after correlation analyses are styled i	n italice					

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^a Factors which initially included more classes but were pooled due to low sample size

GIS Geographic information system, SM Supplementary Materials

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samplings for each stormwater pond at every field session. These factors were gathered in the "immediate environmental factors" group. Simultaneously, biological factors were checked (e.g. presence of branches, aquatic plants, fishes) and gathered in the "biotic factors" group. During the afternoons of the first field session, the stormwater pond characteristics were noted (e.g. nature and state of exclosure measures, nature of substrate, slopes, volume) and gathered in the "pond design" group. The surface area of landscape elements (e.g. wetlands, crops, forests) was calculated using GIS with a precision of 1:10,000 (data from CIGAL 2013) in a 500-mwide buffer ("local-scale group") and 5-km-wide buffer ("large-scale group") around each stormwater pond (Smith and Green 2005). The pollutants of 34 stormwater ponds were quantified between November 2015 and January 2016 in water samples ("water fraction pollutants" group) and sediment ("sediment fraction pollutants" group) collected in the centre of ponds. The entire pollutant sampling protocol was performed by the road maintenance authority as part of its pollutant survey, and was not part of the present study. The set-up protocol to collect samples was similar between ponds and pollutants. Samples were always collected after an intense rainy event, with a 10-L flask, and close to the outlet without precise depth. Samples were individually homogenised without creating vortex inside. The SM Table in Supplementary Materials summarises the list of the measured pollutants, the methods used, the ISO norms, French legal thresholds (Bas-Rhin 2008; MEEM 1998) authorised for stormwater pond outlets and sublethal thresholds, when available. PH was measured twice: once as part of the water fraction pollutants group, and once for the immediate environmental factors group. Finally, the list and number of species known to be present in Bas-Rhin and in a 10-km radius around each stormwater pond were evaluated using a local biodiversity atlas (Odonat 2017).

Methods

Objective 1: qualifying and quantifying amphibian communities

The global and specific presence (*occurrence*), the global and specific abundance of adults and the amphibian species richness were calculated for each stormwater pond. The global occurrence was set at 1 for a stormwater pond when at least one amphibian adult, larvae, tadpole or egg was found during one of the three field sessions, whatever the species. The specific occurrence was calculated in the same way, but for each species. Specific abundance was defined as the maximal abundance of adult amphibians observed in a stormwater pond among the three field sessions for each species. Global abundance was calculated as the sum of specific abundances. Species richness was defined as the number of different amphibian species observed in one stormwater pond during the three field sessions, whatever the developmental stage of individuals (adult, larvae, tadpole or egg). However, as tadpole, eggs and larvae could be difficult to identify and find for some species, we excluded them from species richness and abundance analysis. Shannon diversity (*H*) and equitability (*E_H*) indices were also calculated (Beals et al. 2000). Descriptive statistics are mean \pm standard error of the mean.

Objective 2: identifying factors driving the amphibian community of stormwater ponds

The global occurrence, global abundance and species richness values of each stormwater pond were used as dependent variables (hereafter referred to as *community parameters*). The factors described in Table 1 were used to explain the three dependent variables. A mean of the three field sessions was calculated for the immediate environmental factors. Measured pollutant concentrations were compared to French legal thresholds and sublethal/lethal thresholds, when available in the literature.

The collinear factors were identified and removed with a stepwise procedure based on the variance inflation factor (significance threshold = 3) and associated with ACP and Spearman tests corrected by Holm's method (Holm 1979). Factors with insufficient class sample size (<10) were removed from analysis. When possible, different classes were gathered to increase class sample size (Table 1). Boosted regression tree (BRT) models were created (Elith et al. 2008) to explain variations in dependent variables (community parameters: global occurrence, global abundance and species richness). These models do not provide p values but indicate the relative influence of each factor on explained variation. As suggested by Albeare (2009), the learning rate (Lr) was set to obtain at least 1000 regression trees with a tree complexity of two. To avoid overparameterisation, each model created was simplified by using a cross-validation method (dismo package; Elith and Leathwick 2017) based on deviance reduction. For each dependant variable, a preliminary model was firstly created for each group of factors (e.g. design, pollutants). A general model was then created using solely the factors remaining after simplification procedures of preliminary models. Only factors with at least 3% of relative influence and 5% of summed relative influences were considered significant and retained for analysis and discussion. For the three final models (one for each dependant variable), the pseudo- R^2 , called D^2 , was calculated (Albeare 2009). To analyse effects per species of these retained factors, a detrended correspondence analysis was performed for each group of factor (vegan and ggord packages) using specific abundances. Statistics were performed with R software (v3.3.0) and GIS analysis with QGIS (v2.18.3).

Results

Objective 1: qualifying and quantifying amphibian communities

The presence of amphibians was observed in 69 (84%) of the 82 stormwater ponds. Fourteen species were found (summarised in Fig. 2), representing 78% of the 18 species known in the study area if we consider that the green frogs group counted as two species (Odonat 2017). The four species that were not found were the fire salamander (*Salamandra salamandra*); the moor frog (*Rana arvalis*), which has not, however, been seen in the study area for 9 years; the common midwife toad (*Alytes obstetricans*); and the yellow-bellied toad (*Bombina variegata*). The species richness of stormwater ponds was 2.44 ± 0.19 species (as a reminder, descriptive statistics are "mean \pm standard error of the mean"). We recorded a total of 2046 adults, with an average of 19.51 (\pm 3.42) adults per stormwater pond. Abundances differed substantially among species (Fig. 2).

Shannon diversity and equitability indices were 0.57 ± 0.06 and 0.71 ± 0.03 , respectively.

No morphological abnormalities were found on adults. Red fox carcasses were found floating in two stormwater ponds. The first pond had vertical concrete banks, and the second had steep banks covered with geomembrane. No service bank or reachable exits were present on the latter, but claw marks were observed on the geomembrane. The month after the study period (August 2016), we observed the death of all the tadpoles in five detention ponds during drought events.

Objective 2: identifying factors driving the amphibian community of stormwater ponds

The proportion of explained deviance (D^2) was high for the three BRT models, one created for each community parameter (global occurrence, global abundance and species richness; Table 2). Chlorides were the only pollutant kept in the statistical BRT model. They had a relative influence of 2.69% on the global amphibian abundance alone, with a minor negative correlation. No stormwater pond pollutants were above French legal thresholds. For sediment pollutants, chromium and PCBs never exceeded thresholds in the ponds studied. For other sediment pollutants, between 23.08 and 92.31% of stormwater ponds were above known sublethal thresholds. For the water fraction, only PAHs were above known sublethal thresholds (17.5% of stormwater ponds). No characteristic of exclosure measures was correlated with any of the studied community parameters. Fitted functions of each factor with more than 3% relative influence are presented in Fig. 3 (occurrence), Fig. 4 (abundance) and Fig. 5 (species richness).

Detrended correspondence analysis (results of significant factors per species) shows that the age of stormwater ponds

was negatively correlated with abundances of the European green toad and green frogs (Fig. 6). The presence of many helophytes was negatively correlated with the presence of vegetative fragments in water. The alpine newt abundance was positively correlated with the presence of helophytes whereas the agile frog and the marsh frog were negatively correlated with it (Fig. 7). The European green toad abundance was positively correlated with pH and possibly with the water temperature too. Similar but less clear correlations were also found for the marsh frog and for the smooth newt (Fig. 8). At local scale (500 m; Fig. 9), the green frog and European green toad abundances were positively correlated with the surface of annual crops (and also at large scale for green frogs; Fig. 10). The surface of deciduous forest was positively correlated with the smooth newt abundance (Fig. 9). At large scale (5000 m), the European green toad abundance was positively correlated with the surface of road and the green frog abundance was also positively correlated with wetland surface (Fig. 10).

Discussion

The amphibian communities of stormwater ponds

As hypothesised, most of the stormwater ponds (84%) hosted amphibians. This suggests high attractiveness of stormwater ponds for amphibians, probably driven by necessity as they cannot find suitable ponds (semi-natural or other). The four species that were never observed on these sites (Salamandra salamandra, Rana arvalis, Alytes obstetricans, Bombina variegata) were not expected to be seen in stormwater ponds as they either avoid large ponds and modified landscapes or are forest species (Räsänen et al. 2003; Vos and Chardon 1998). A previous study showed that more than a quarter of the total numbers of individuals of the European green toad (Bufotes viridis) in the study area were located in stormwater ponds (Sané and Didier 2007). Our study shows a similar result, as it was the species with the second highest recorded mean abundances in stormwater ponds. It suggests that stormwater ponds play a crucial role in the conservation of this species, which is strongly protected in this study area. The marsh frog (Pelophylax ridibundus) was also frequently found in stormwater ponds. Those two species were both pioneer species (Pagano et al. 2001; Sinsch et al. 2007), as observed by Scher and Thièry (2005) in Mediterranean motorway stormwater ponds. If the European green toad tolerates small puddles and gravel pits, the marsh frog needs large ponds, with sufficient depth and high exposure to sunlight (Kuzmin et al. 2009), such as retention stormwater ponds with concrete banks and riprap providing basking sites. The other species, especially newts and those needing more natural conditions, were less abundant. Indeed, even if some stormwater ponds

Fig. 2 Specific mean abundances per stormwater pond. Error bars are standard error of the mean. Abundances are the maximal number of adult amphibians observed in a stormwater pond among the three field sessions for each species. Most observed species were green frogs, European green toad, smooth newt and marsh frog



looked like natural ponds with helophytes, clear waterprotected species (e.g. Odonata, amphibians) and aquatic ecosystems, most (56%) were vegetation free with cloudy/turbid water. The number of amphibians found in studied ponds was probably underestimated as we did not adjust numbers according to the species detection probability (Schmidt 2004). However, the global number of individuals per pond was quite high compared to that in other ponds in the studied area (2.95

Table 2 Relative influences of significant factors on deviance in each model

Factor	Relative influence (%)				
	Global occurrence $(D^2 = 0.64)$	Global abundance $(D^2 = 0.76)$	Species richness $(D^2 = 0.54)$	Sum of relative influence (%)	
pH	16.55	8.05	9.56	34.15	
Volume (m ³)	4.44	6.53	20.83	31.81	
Roads (500 m ²)	14.81	4.66	4.37	23.84	
Wetlands (5000 m ²)	6.94	10.02	6.24	23.20	
Deciduous forests (500 m ²) (c.t. mixed and softwood forest)	4.17	2.64	13.73	20.54	
Roads (5000 m ²)	2.18	14.57	3.30	20.04	
Annual crops (500 m ²) (meadows, urban)	6.26	4.06	6.05	16.37	
Deciduous forests (5000 m ²) (i.e. mixed and softwood forest)	1.87	6.22	6.83	14.92	
Annual crops (5000 m ²) (i.e. vines, copses)	6.96	3.21	3.57	13.73	
Meadows (5000 m ²)	1.08	12.46	0	13.54	
Helophytes (to fix eggs to branches and as shelters)	1.87	5.93	3.54	11.34	
Algae and hydrophytes (to fix eggs to branches and as shelters)	2.85	5.17	0	8.02	
Water temperature (°C) (i.e. air temperature)	2.87	2.12	2.71	7.70	
Odonata larvae (as predators)	2.65	1.39	3.54	7.58	
Hedgerows (500 m ²)	2.10	1.76	3.52	7.38	
Age (years)	1.45	2.52	2.99	6.95	
Surrounding species richness	3.85	0.62	1.77	6.24	
Vegetative fragments in water (as food)	3.87	0	2.00	5.87	
Hedgerows (500 m ²)	2.10	0	3.52	5.62	
Surrounding species richness	3.85	0	1.77	5.62	

In line 1, the pH explained 16.55% of variations of the global occurrence model, which itself explained 64% of the variation of the global occurrence. Landscape element factors explained 56% of the summed relative influences



Fig. 3 Fitted functions of factors with more than 3% relative influence on the deviance of the global occurrence model. The meaning of factors lettering is in Table 1. Basic pH, roads at local scale (presumably a barrier

effect) and annual crops were the main factors having negative impacts on the presence of amphibian in stormwater ponds. Wetlands and pond volume were the main positive ones

times higher for green toad; Vacher et al. 2015). Stormwater ponds therefore host a high number of adults and species with great interspecies variations in population, as found by Scheffers and Paszkowski (2013) and Gallagher et al. (2014). Shannon and equitability values were low, suggesting long-standing populations. This is corroborated by the presence of amphibians in the studied stormwater ponds, whatever the age of the latter. Although our single breeding season observation study leads us to consider stormwater ponds as a real habitat or to have high ecological potentiality in Bas-Rhin, it is necessary to monitor populations over many years and to do so in other landscapes. Indeed, pollutant-induced genotoxicity could significantly reduce population viability throughout the years (Hamer et al. 2012). This would imply that a suitable stormwater pond can at terms increase the extinction probability although it could contain high amphibian abundances and species richness. An approach based on life history traits is needed in order to conclude about the potential trap effect of stormwater ponds (Sinsch et al. 2007). This approach should compare at least reproductive success between stormwater ponds and controls. In the current study, we initially wanted to include semi-natural ponds as controls but they were located in too different landscapes associated with other communities, or their number was too low to be significant. Therefore, we cannot exclude an ecological trap effect and caution is needed. However, the study highlights



Fig. 4 Fitted functions of factors with more than 3% relative influence on the deviance of the global abundance model. The meaning of factors lettering is in Table 1. Basic pH and surface of deciduous forests at large

scale were the main factors having negative impacts on amphibian abundances in stormwater ponds. Road at large scale, meadows, wetlands and pond volume were the main positive ones



Fig. 5 Fitted functions of factors with more than 3% relative influence on the deviance of the species richness model. The meaning of factors lettering is in Table 1. Basic pH and roads at local scale were the main

factors having negative impacts on amphibian abundances in stormwater ponds. Pond volume, deciduous forests, wetlands and annual crops (possible a refuge effect) were the main positive ones

the relevance of studying the ecological potentiality of stormwater ponds as they can be the last wetlands remaining in highly modified landscapes.

Factors driving the amphibian community of stormwater ponds

Despite the known negative impacts of pollutants (Brand et al. 2010; Brand and Snodgrass 2010; Collins and Russell 2009; Gallagher et al. 2014; Karraker et al. 2008), the concentrations observed in this study were probably too low to have a significant effect. Despite our initial hypothesis, no clear evidence of an overall effect of pollutants was found on amphibian communities at the observed concentrations in stormwater

ponds in Bas-Rhin. This result was supported by the absence of morphological abnormalities. However, the method used in the present study (examination of adult abnormalities) is not as precise as the examination of tadpoles (Wagner et al. 2014). Further experiments should therefore be performed to examine the effect of the observed concentrations of pollutants on amphibian development and reproduction. pH had the highest relative influence on the studied community parameters (global occurrence, global abundance and species richness), with a negative correlation for pH values of 8 to 10 (Figs. 3, 4 and 5). As only one stormwater pond had acidic water (< 7), only the basic effect of pH was revealed by models. The negative effect of basic pH (>7) on amphibian communities can be due to herbicides, whose negative effects are amplified in this

Fig. 6 Correlation between species abundances (based on adults) and significant factors of the pond design group. The abundance of a species whose name is located near the extremity of a factor is positively correlated with the factor. When the name is at the opposite, the correlation is negative. Greens frogs and the European green toad were more abundant in recent stormwater ponds, and no clear species correlation was found with the pond volume



Fig. 7 Correlation between species abundances (based on adults) and significant factors of the biotic factors group. The abundance of a species whose name is located near the extremity of a factor is positively correlated with the factor. When the name is at the opposite, the correlation is negative. Agile frog was more abundant when no vegetative fragments were present in ponds. Conversely, alpine newt seemed to prefer ponds with many helophytes



condition. Moreover, pollutant analysis performed between November 2015 and January 2016 revealed a neutral pH level in stormwater ponds. This indicates that pH became basic during spring, probably due to nitrites from agricultural activities. As these results are based on winter concentrations, the effects of nitrites may be also underestimated. However, detrended correspondence analysis showed a positive relation between pH and abundance of the European green toad (Fig. 8), which is consistent as the species mainly use ponds in farmlands (Michel et al. 2017; also shown here in Fig. 9).

Inside the pond design group, pond volume was the main predictive factor of amphibian communities. Volumes of less than 1000 m^3 had a significant positive linear effect on community parameters. As the depth was

Fig. 8 Correlation between species abundances (based on adults) and significant factors of the immediate environmental factors group. The abundance of a species whose name is located near the extremity of a factor is positively correlated with the factor. When the name is at the opposite, the correlation is negative. The European green toad was more abundant in ponds with basic pH (note that the pH range of ponds was 5.8–12.35 with a mean value of 9.05)





Fig. 9 Correlation between species abundances (based on adults) and significant factors of the land use at local scale group. The abundance of a species whose name is located near the extremity of a factor is positively correlated with the factor. When the name is at the opposite,

the correlation is negative. Greens frogs and the European green toad were more abundant in stormwater ponds located in farmland whereas the smooth newt was mainly present in stormwater ponds located in deciduous forests

not correlated to the volume and was not retained in final models, we suggest that only the surface of stormwater ponds and the perimeter size were of importance for amphibian communities (Guderyahn et al. 2016; Morand and Joly 1995). There were too few stormwater ponds with volumes of over 1000 m³ to consider any fitted function over this threshold to be significant. Moreover, the effect of volume seems only accurate for global parameters as no specific correlation was observed (Fig. 6). The age of ponds is an interesting factor, as it could reflect the longterm sustainability of amphibian populations. The age of ponds showed no correlation with the presence of most species. However, it did show a reverse relationship with the green frog and European green toad population (Fig. 6). The main hypothesis is that those two species colonise the stormwater ponds as soon as they are created, inducing a highest population in very young stormwater

ponds, maybe decreasing later with the colonisation of ponds by helophytes (Fig. 7). In this case, a regular dredging should favour those species.

Until now, stormwater ponds were intended to be unreachable for any species due to the pollutants contained in the water. However, exclosure measures were clearly demonstrated to be ineffective against amphibians. Even the most overprotected stormwater ponds equipped with concrete walls hosted amphibians. Indeed, there are many ways to enter stormwater ponds. Amphibians can follow street gutters and fall into the drain network. Depending on the design of the pond, they can also enter via the outlet. Another hypothesis is that birds could carry eggs of amphibian or fish on their legs, but no publication to date validates this popular thought. However, local fishermen indicated us regular observation of grey heron (*Ardea cinerea*) drop off in ponds living amphibians and



Fig. 10 Correlation between species abundances (based on adults) and significant factors of land use at large scale group. The abundance of a species whose name is located near the extremity of a factor is positively correlated with the factor. When the name is at the opposite, the

correlation is negative. Greens frogs and the European green toad were more abundant in stormwater ponds located in farmland whereas the smooth newt was mainly present in stormwater ponds located in deciduous forests

fishes kept in their beak. Eggs may also be involuntarily carried by humans during maintenance process or fishing activities (some stormwater ponds were even used as stock ponds of northern pike). Poor vegetation maintenance also allows tall grass to grow against the fences and walls, helping amphibians to reach the top. Access can also be gained through large holes left in small fences after careless maintenance work. Finally, fence staples can eventually fall out, creating gaps through which amphibians can pass. If the current exclosure measures cannot keep amphibians out of stormwater ponds, the overprotection of stormwater ponds has to be questioned. The cost of creating completely unreachable stormwater ponds is high. It would require high concrete walls, large cattle grids under portal access, bird netting, fine grids over floor drains and outlets, etc., and would require permanent maintenance to avoid the obstruction of hydraulic pipes and the deterioration of netting. Although creating an underground stormwater pond would be far less expensive, the potential ecological value of stormwater ponds is not to be brushed aside in the current context of modified landscape where the number of semi-natural ponds is limited.

Few biotic and immediate environmental factors (such as temperature or wind during the field sessions) were retained in the final models. Surprisingly, the presence of fish in the ponds had no effect on community parameters (a similar result to Le Viol et al. 2012), despite evidence that the predation by fish negatively affects the occurrence of amphibians (Brown et al. 2012; McCarthy and Lathrop 2011). It can indicate that amphibian species had not enough evolutionary time to avoid stormwater ponds with higher densities of predators, or that amphibian had no other ponds to go to. Surprisingly, abundances of Odonata larvae were positively associated with those of amphibians despite the fact that they are tadpole predators. However, the presence of Odonata larvae could highlight an overall high water quality leading to a higher number of amphibians (Corbet 1999 in Kalkman et al. 2008).

Abundant aquatic vegetation is known to be positively correlated with amphibian occurrence and species richness (McCarthy and Lathrop 2011; Shulse et al. 2010; Simon et al. 2009). This was partially confirmed in this study, as the presence of helophytes increased global levels of amphibian abundance. However, high quantities of helophytes slightly decreased the amphibian abundance levels. This may be explained by an observational bias, because of the hard detectability of amphibians among dense vegetation rather than because of a real effect of helophytes on the amphibians. A similar effect can probably be observed for algae and vegetative fragments.

As commonly found in the literature, the main factors driving the parameters of amphibian communities were the landscape element factors (Babbitt et al. 2006; Birx-Raybuck et al. 2010; Malmgren 2002; Pillsbury and Miller 2008; Scheffers and Paszkowski 2013: Scher and Thièry 2005: Sinsch et al. 2012). They alone explained more than half of the observed variations. Some correlations (for example with wetlands, meadows, surrounding species richness and deciduous forests) were self-explanatory, given their accordance with the biology of amphibians (Birx-Raybuck et al. 2010; Guderyahn et al. 2016; Le Viol et al. 2012; Simon et al. 2009). One of the main discoveries was the unexpected negative correlation between species richness and stormwater ponds located in intermediary zone between cultivated lands and deciduous forest (Grillet et al. 2015). A shared hypothesis can be made for intermediate areas of annual crops as the same negative correlation was found. In fact, the landscape dynamic of the studied area could be separated into two types of landscape elements (as ponds were never located in cities): annual crops and semi-natural elements mainly composed of forests and wetlands. When the surface of annual crops decreased, the landscape was mainly composed of more natural elements and potentially suitable habitats for amphibians. Thus, the greater number of amphibians could induce a greater number of amphibians in the stormwater ponds. A similar result was observed in newts by Joly et al. (2001). Fewer semi-natural elements were present in the landscape when the surface area of annual crops increased, so stormwater ponds were probably the only suitable habitats for amphibians and were used as refuges (Le Viol et al. 2012; Le Viol et al. 2009). Another hypothesis could be simply the change in species composition as the smooth newt was mainly observed in deciduous forest landscape and as green frogs and the European green toad were mainly found in farmlands (Fig. 9). In this case, it would suggest that an intermediate pond could be less efficient to hold large amphibian diversity than fully located in farmland or in deciduous forest ponds. On the other hand, at a local scale, roads were found to have a strong negative effect on amphibians. Although this effect could be due to road pollutants (light, noises, chemicals), we think the barrier effect of road played the biggest role. Indeed, no amphibians were found when ponds were totally surrounded by roads, as reported by Scheffers and Paszkowski (2013) and Parris (2006). However, at large scale, a positive correlation was found between land covered by roads and abundances of European green toad (Fig. 10), possibly due to a higher amount of stormwater ponds (roads or industrial ones).

Recommendations

As exclosure measures are inefficient in preventing stormwater ponds access to amphibians, the time and financial means dedicated to them should be reallocated to developing a better sediment cleaning process and creating substitute seminatural ponds that are not connected to road runoff. Observed pollutant concentrations were observed to have no effect on the studied amphibian community parameters (global occurrence, global abundance and species richness); it is therefore futile to overprotect stormwater ponds which have real ecological value, but caution is still required. A sensible approach could be to consider all newly constructed stormwater ponds polluted and to remove the exclosure measures once the low pollutant concentrations have been demonstrated. An easier alternative for road managers would be to install efficient exclosure measures or other compensatory measures only if pollutant concentration reached sublethal known thresholds after a 5-year survey or before this time if very high pollutant concentrations are measured.

The design of stormwater ponds should be systematically validated by a herpetologist to avoid creating potentially mortal traps for animals. For example, the unclimbable banks of two stormwater ponds in this study were traps for mammals and possibly for amphibians too (Chang et al. 2011; Zhang et al. 2010). Exit ladders should be systematically installed. As detention pond water is not permanent, all the tadpoles of these sites can die during droughts (Brand and Snodgrass 2010; Gallagher et al. 2014). Although some amphibian larvae can accelerate their growth and development in harsh ecosystems, they cannot survive these sudden events, which appear to be more frequent with global warming (Morand et al. 1997; Newman 1992). We therefore recommend the creation of a permanent minimum water level that can easily be dredged-not only in detention ponds but also in newly created semi-natural ponds. This extension of the hydroperiod would enhance species occurrences and abundances (Guderyahn et al. 2016).

Other recommendations can be found in the study of Clevenot et al. (2018) about other factors not developed in this study, such as dredging and vegetation maintenance.

Conclusions

The present study demonstrated a high potential ecological value of stormwater ponds for amphibian communities in the agricultural plain of Bas-Rhin. Although no pollutant effect was observed on studied community parameters, we cannot unequivocally conclude that pollutants have no effect on the viability of the stormwater pond amphibian populations, and therefore that stormwater ponds are not ecological traps. However, we have demonstrated the ineffectiveness of exclosure measures. In these conditions, we recommend avoiding the installation of costly partitioning measures and reallocating efforts to ecological engineering to create better ecological stormwater ponds and more semi-natural ponds in the study area to limit a possible ecological trap effect. Further analysis is needed in other landscapes and over many years to generalise our results.

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