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

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

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11 **Abstract**

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

31 *Keywords:* refuge habitat, retention ponds, farmlands, semi-natural habitats, modified landscape, pollution

32 1. Introduction

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2
3 33 Behind over-exploitation, agriculture is the next-leading cause of current biodiversity decline and
4
5 34 affects all taxonomic groups (Butchart et al., 2010; Maxwell et al., 2016). The habitat loss induced by land
6
7 35 consolidation leads to a great number of local population extinctions through the modification of the use and
8
9 36 layout of land (Fischer and Lindenmayer, 2007). The creation of vast monocultural farmlands causes the
10
11 37 fragmentation of residual semi-natural elements and has negative impacts on species richness, population
12
13 38 abundances, growth rate and distribution, trophic chain length, breeding and dispersal success (Benton et al.,
14
15 39 2003; Fahrig, 2017). Roads are another cause of biodiversity loss (Forman and Alexander, 1998; Maxwell et al.,
16
17 40 2016). Their negative impacts are observed in the *road-effect zone* and are numerous (Richard et al., 2000; Van
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19 41 Der Ree et al., 2011). For example, roads decrease habitat sizes and quality, affect the life-history traits and
20
21 42 population characteristics of species and increase direct and indirect mortality rates (Redon (de) et al., 2015;
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23 43 Spellerberg, 1998). They also contribute to landscape fragmentation through the linearization of landscape
24
25 44 (Holderegger and Di Giulio, 2010). These negative effects have numerous adaptations and evolutionary
26
27 45 consequences such as changes in vocal pitch and activities in response to traffic noise in birds and amphibians
28
29 46 species (Lengagne, 2008; Slabbekoorn and Peet, 2003), higher pollutants tolerance (Brady, 2012), or change of
30
31 47 bird wing length in response to roadkill (Brown and Bomberger Brown, 2013; Kiang, 1982). These
32
33 48 consequences should be taken into account on impacts studies so that the adaptation would not be
34
35 49 underestimated and protection measures would be effective (Brady and Richardson, 2017). Together, intensive
36
37 50 agricultural activities and roads form highly modified landscapes in which few natural and semi-natural elements
38
39 51 remain, where biodiversity can be very low, and where impacts from both roads and agricultural activities are
40
41 52 present (Donald et al., 2001; Foley et al., 2005; Stoate et al., 2001).

42
43
44 53 Wetlands are particularly impacted by anthropogenic activities like roads and agriculture. At least 64%
45
46 54 of them have disappeared over the past century, and little data is available for the conservation status of what
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48 55 remains. There is a continuing decline of wetlands, accelerated by urban expansion, agricultural intensification,
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50 56 land consolidation and the construction of motorways (Davidson, 2014). Thus, they are internationally protected
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52 57 as they provide important ecosystemic services such as biomass and resource production, pollutant and climate
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54 58 regulation, flood abatement and erosion decrease (Bolund and Hunhammar, 1999; Russi et al., 2013; Zedler,
55
56 59 2003). Moreover, one third of non-marine vertebrate species inhabit wetlands, giving them a high ecological
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58 60 value (Dudgeon et al., 2006). In highly modified landscapes such as farmlands fragmented by roads, wetland
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61 species can survive habitat loss by colonizing the poor quality, isolated remains of semi-natural habitats
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2 62 (McKinney, 2006; Sinsch et al., 2012).
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4 63 To prevent some of the alteration of aquatic systems, wetlands and other habitats in modified
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6 64 landscapes, artificial ponds called stormwater ponds are built. Located next to roads, urban areas or industrial
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8 65 ones, they are designed to collect, stock and decontaminate runoff otherwise released into a nearby stream. These
9
10 66 structures protect outside water from chronic and exceptional contamination (Karouna-Renier and Sparling,
11
12 67 2001; Scher and Thiéry, 2005). They also aim to prevent and control flooding events, and to store chenal and
13
14 68 canal volume (EPA, 2009). They are required by European legislation for certain surfaces and associated water
15
16 69 volumes, and for new or existing infrastructures (Le Viol et al., 2009). There are several types of stormwater
17
18 70 ponds (such as micropool, stormwater wetlands...), two of which are retention ponds, which permanently
19
20 71 maintain a pool of polluted water throughout the year (permanent hydroperiod), and detention ponds, which hold
21
22 72 clean water for a short period of time before it enters the stream (temporary hydroperiod). While detention ponds
23
24 73 are usually a simple hole, retention ponds are designed with additional characteristics such as waterflow
25
26 74 regulation structures, hydrocarbon separators and waterproof covers (EPA, 2009). Retention ponds can be
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28 75 underground or open-air structures.
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32 76 Although roads contribute to wetland loss (Van der Ree et al., 2015), amphibian roadkill (Fahrig et al.,
33
34 77 1995 ; Elzanowski et al., 2009) and the invasion of non-indigenous species (Jodoin et al., 2008), they may also
35
36 78 provide alternative semi-natural habitats for wetlands species. Like natural wetlands, stormwater ponds can
37
38 79 provide ecosystem services and are inhabited by flora, birds, invertebrates, snakes and fishes (Ackley and
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40 80 Meylan, 2010; Bishop et al., 2000; Karouna-Renier and Sparling, 2001; Le Viol et al., 2009; Moore and Hunt,
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42 81 2012). The biodiversity of stormwater ponds can be equivalent to that of semi-natural wetlands (Hassall and
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44 82 Anderson, 2015) and these habitats can be inhabited by rare and protected species (Le Viol et al., 2012). A better
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46 83 understanding of their ecological function on a large scale and in highly modified landscapes is however needed
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48 84 (Brand and Snodgrass, 2010; Scheffers and Paszkowski, 2013). The small number of studies focusing on
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50 85 stormwater pond biodiversity are relatively recent (Bishop et al., 2000). This scarcity of literature could be
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52 86 explained by disinterest of pond managers for stormwater pond biodiversity (Hassall and Anderson, 2015), or by
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54 87 their desire to avoid colonisation by amphibians as stormwater pond water can be polluted (Massal et al., 2007;
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56 88 Snodgrass et al., 2008) and therefore these ponds could be ecological traps (Battin, 2004). Indeed, runoff
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58 89 collected by stormwater ponds can contain agents toxic for aquatic fauna. They contain heavy metals (Wik et al.,
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60 90 2008), polycyclic aromatic hydrocarbons PAHs (Neff et al., 2005) and chlorides (Gallagher et al., 2014). Water
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91 column pollutants can vary through time, an example being the high increase in chloride concentrations in late
1 92 winter due to the use of road salt as de-icing agent (Collins and Russell, 2009). Overall pollutant concentrations
2 93 can quickly increase after storm rainfall on the road surface. However, only a small proportion of these
3 94 pollutants reaches stormwater ponds as the majority evaporates, stays on road surface, or is degraded by sun
4 95 exposure (Pagotto, 1999). Pollutant concentration of the sediment is more stable over time, and high
5 96 concentrations are accumulated, especially if sediment is rarely removed. This can be a non-negligible threat for
6 97 species that hide or winter in sediment and also for their predators because of the bioaccumulation effect (Bishop
7 98 et al., 1995; Brand et al., 2010). Concentrations of pollutants can also vary according to the landscape. Maximum
8 99 threshold levels of nitrites and nitrates from agricultural chemicals can be recorded in highly modified
9 100 landscapes such as farmlands (Hayes et al., 2006). Thus, despite evidence of the habitat potentiality of
10 101 stormwater ponds provided by a number of studies (Brown et al., 2012; Scheffers and Paszkowski, 2013), the
11 102 stormwater ponds could still be ecological traps. Therefore some countries including France demand, at great
12 103 expense, the installation of enclosure measures such as fences, walls, cattle grids or handrails (Morand and
13 104 Carsignol, 2019)

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28
29 105 Amphibians are wetlands species which can be found in stormwater ponds (McCarthy and Lathrop,
30 106 2011; Sievers et al., 2019). One third of them is threatened with extinction all over the world, due to several
31 107 causes such as diseases and habitat loss (Arntzen et al., 2017; Becker et al., 2007; Dudgeon et al., 2006;
32 108 Eterovick et al., 2005; Mazerolle et al., 2005). This last threat is particularly serious as the habitat needs of pond-
33 109 breeding amphibians vary greatly according to the biological traits and biodemographic strategies of each
34 110 species (Van Buskirk, 2005). Amphibians are also vulnerable to the impact of road traffic due to their immobility
35 111 facing motor vehicles (Gibbs and Shriver, 2005; Mazerolle et al., 2005) and all their population-scale
36 112 movements (Joly, 2019). Indeed, the amphibian mass migration concerning most species can reach a distance of
37 113 15 km for anurans and occurs twice a year (Beebee, 1996; Sinsch, 1990). Therefore, it leads amphibians to cross
38 114 many roads. In highly modified landscapes where only few wetlands remain, it can be difficult for every
39 115 amphibian species to find suitable ponds (Hamer and McDonnell, 2008). By varying in location, shape, design,
40 116 function and management, stormwater ponds could serve as alternative habitat for some amphibian species.
41 117 However, there are very few studies focusing on amphibian community in stormwater ponds. Further research is
42 118 required to understand the factors, especially pollutants, driving these communities and to find ways of
43 119 improving the habitat quality of stormwater ponds for amphibians (Brand and Snodgrass, 2010; Scheffers and
44 120 Paszkowski, 2013; Scher and Thiéry, 2005). With sufficient concentration, pollutants can induce sterility and
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121 external and internal abnormalities in amphibians, stunt their growth and increase difficulties during
122 metamorphosis (Bryer et al., 2006; Egea-Serrano et al., 2012; Sievers et al., 2018; Wagner et al., 2014).
123 Concurrently to a bad stormwater pond design which can trap amphibians in dead-end, pollutants could make
124 stormwater ponds ecological traps and therefore toll the bells for these species in modified landscape where few
125 wetlands remain (Clevenot et al., 2018).

126 **2. Research objectives**

127 This study focuses on the agricultural plain of the Bas-Rhin (Bas-Rhin: 4 755km², 23 inhabitants per
128 km²), a highly modified landscape composed of vast monoculture areas and roads. We performed a one-breeding
129 season survey of the Anura and Caudata amphibian communities found in stormwater ponds located along public
130 roads with moderate traffic.

131 Our objectives were to: (1) Qualify and quantify the amphibian community biodiversity and species
132 richness of stormwater ponds, and to; (2) Identify and quantify factors driving the constitution parameters of
133 amphibian communities of stormwater ponds (species and density of population).

134 We expected that: Firstly, a significant proportion of stormwater ponds hosted amphibians and that
135 inter-species differences existed, following factors like ponds design (size, depth, slopes, etc.) and surrounding
136 land use. Secondly, a negative influence of pollutants, and particularly chlorides, nitrites and nitrates, and no
137 effect of enclosure measures on amphibian communities.

138 **3. Material and Methods**

139 3.1. Protocol

140 *3.1.1. Studied area*

141 The Department of the Bas-Rhin (Alsace, Grand-Est, France, **Fig. 1**) has a semi-continental climate
142 with an average temperature of 10.4 °C and an atmospheric temperature range of 30 °C. The annual precipitation
143 is about 700 mm per year and the average altitude is 150 m. The road density is 1.9 km/km², with more than
144 3 654 km of public roads with moderate traffic and 240 km of major roads (mainly private large road with high
145 traffic). In the western part of the Bas-Rhin, a range of low mountains forms the geomorphological unit of the
146 Vosges. Its flora is mainly dominated by Picea, fir, common beech and oak. In the East, the Rhine river is
147 bordered by many wetlands that are certified by the RAMSAR convention (n°FR7200025) and protected by the

148 Natura 2000 European network (n°FR4211811, n°FR4211810). These wetlands make up less than 1% of the
149 Bas-Rhin area, the successive building of dykes along the Rhine (1842-1876, 1928-1959) having substantially
150 diminished the area of wetlands in this region. The intensification of agricultural practices also decreased their
151 surfaces through land consolidation. This is illustrated by the courses of the Rhine tributaries from the Vosges,
152 which cross a large agricultural plain landscape that is currently dominated by wheat and maize crops. There is
153 no precise inventory of wetland loss in the Bas-Rhin. On a national scale, France has lost over 67% of total
154 wetland area since the beginning of the 20th century (Ximenès et al., 2007). The studied area hosts 18 amphibian
155 species.

3.1.2. Studied ponds

157 We monitored 82 of the 84 open-air stormwater ponds of the Bas-Rhin public roads with moderate
158 traffic (**Fig. 1**), and could not safely access the two remaining ponds. Traffic on those public roads is mainly
159 below 10 000 vehicles/day. Stormwater ponds were mainly located in the agricultural plain. Six were located in
160 the Vosges Mountains and only one was in the RAMSAR area near the Rhine (**Fig. 1**). Stormwater ponds
161 differed substantially in terms of age (from less than one year old to 24), volume (from to 50 to 7 000 m³) and
162 design (e.g., nature of enclosure measures, type of substrate, angle of banks, etc.). Most (73) were retention
163 ponds (permanent hydroperiod).

3.1.3. Sampling design

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

170 The sampling protocol was adapted from the “POP Amphibiens communauté” (Barrioz et al., 2016). Sampling
171 began at dusk. For each pond, sampling began by five minutes of listening to estimate the species richness and
172 number of adult males. Visual sampling was then performed with headlamps and flashlights. The sampling was
173 stopped after two patrols around each pond in order to have an equivalent sampling effort between ponds.
174 Indeed, most of them differed substantially in length but only slightly in width. The number of individuals,
175 amplexus, eggs and larvae was noted for each species. The number of floating carcasses and any visible

176 morphological abnormalities on adults was also recorded. To avoid any mistakes due to incorrect identification,
177 edible frogs (*Pelophylax kl. esculentus*), and pool frogs (*Pelophylax lessonae*) were gathered in a “green frogs”
178 group. We used the Miaud and Muratet, 2004 identification key. The experimental protocol was authorized by
179 the 5/6/2016 nominative authorisation of the French Environmental Code and followed EU Directive
180 2010/63/EU guidelines for animal experiments.

181 3.1.4. *Environmental factors*

182 Several factors were also checked and classified in factor groups (**Table 1**). Environmental factors (e.g.,
183 pH, water and air temperature, visibility, etc) were recorded immediately after samplings for each stormwater
184 pond at every field session. These factors were gathered in the “*immediate environmental factors*” group.
185 Simultaneously, biological factors were checked (e.g., presence of branches, aquatic plants, fishes, etc.) and
186 gathered in the “*biotic factors*” group. During the afternoons of the first field session, the stormwater pond
187 characteristics were noted (e.g., nature and state of enclosure measures, nature of substrate, slopes, volume...)
188 and gathered in the “*pond design*” group. The surface area of landscape elements (e.g., wetlands, crops, forests,
189 etc.) was calculated using GIS with a precision of 1:10 000 (data from CIGAL 2013) in a 500 m-wide buffer
190 (“*local-scale group*”) and 5 km-wide buffer (“*large-scale group*”) around each stormwater pond (Smith et green
191 2005). The pollutants of 34 stormwater ponds were quantified between November 2015 and January 2016 in
192 water samples (“*water fraction pollutants*” group) and sediment (“*sediment fraction pollutants*” group) collected
193 in the centre of ponds. The entire pollutant sampling protocol was performed by the road maintenance authority
194 as part of its pollutant survey, and was not part of the present study. The set up protocol to collect samples was
195 similar between ponds and pollutants. Samples were always collected after an intense rainy event, with a 10 L
196 flask, and close to the outlet without precise depth. Samples were individually homogenised without creating
197 vortex inside. The **SM Table in Supplementary Materials** summarizes the list of the measured pollutants, the
198 methods used, the ISO norms, French legal thresholds (Bas-Rhin, 2008; MEEM, 1998) authorized for
199 stormwater pond outlets, and sublethal thresholds, when available. PH was measured twice: once as part of the
200 *water fraction pollutants* group, and once for the *immediate environmental factors* group. Finally, the list and
201 number of species known to be present in the Bas-Rhin and in a 10 km radius around each stormwater pond was
202 evaluated using a local biodiversity atlas (Odonat, 2017).

203 3.2. Methods

204 3.2.1. *Objective (1): Qualifying and quantifying amphibian communities*

205 The global and specific presence (=occurrence), the global and specific abundance of adults and the
1 amphibian species richness were calculated for each stormwater pond. The global occurrence was set at 1 for a
2 206 stormwater pond when at least one amphibian adult, larvae, tadpole or egg was found during one of the three
3 207 field sessions, whatever the species. The specific occurrence was calculated in the same way, but for each
4 208 species. Specific abundance was defined as the maximal abundance of adult amphibians observed in a
5 209 stormwater pond among the three field sessions for each species. Global abundance was calculated as the sum of
6 210 specific abundances. Species richness was defined as the number of different amphibian species observed in one
7 211 stormwater pond during the three field sessions, whatever the developmental stage of individuals (adult, larvae,
8 212 tadpole or egg). However, as tadpole, eggs and larvae could be difficult to identify and find for some species, we
9 213 excluded them from species richness and abundance analysis. Shannon diversity (H) and equitability (E_H)
10 214 indices were also calculated (Beals et al., 2000). Descriptive statistics are mean \pm standard error of the mean.
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23 216 3.2.2. Objective (2): Identifying factors driving the amphibian community of stormwater ponds

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26 217 The global occurrence, global abundance and species richness values of each stormwater pond were
27 218 used as dependent variables (hereafter referred to as *Community Parameters*). The factors described in **Table 1**
28 219 were used to explain the three dependent variables. A mean of the three field sessions was calculated for the
29 220 “immediate environmental factors”. Measured pollutant concentrations were compared to French legal
30 221 thresholds and sublethal/lethal thresholds, when available in the literature.
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37 222 The collinear factors were identified and removed with a stepwise procedure based on the Variance Inflation
38 223 Factor (significance threshold = 3) and associated with ACP and Spearman tests corrected by Holm’s method
39 224 (Holm, 1979). Factors with insufficient class sample size (<10) were removed from analysis. When possible,
40 225 different classes were gathered to increase class sample size (**Table 1**). Boosted regression tree (BRT) models
41 226 were created (Elith et al., 2008) to explain variations in dependent variables (community parameters: global
42 227 occurrence, global abundance and species richness). These models do not provide p-values but indicate the
43 228 relative influence of each factor on explained variation. As suggested by Albeare (2009), the learning rate Lr was
44 229 set to obtain at least 1 000 regression trees with a tree complexity of two. To avoid overparameterization, each
45 230 model created was simplified by using a cross-validation method (*dismo* package; Elith & Leathwick, 2017)
46 231 based on deviance reduction. For each dependant variable, a preliminary model was firstly created for each
47 232 group of factors (e.g., design, pollutants, etc.). A general model was then created using solely the factors
48 233 remaining after simplification procedures of preliminary models. Only factors with at least 3% of relative
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234 influence and 5% of summed relative influences were considered significant and retained for analysis and the
1 discussion. For the three final models (one for each dependant variable), the pseudo-R², called D², was calculated
2 235
3
4 236 (Albeare, 2009). To analyse effects per species of these retained factors, a detrended correspondence analysis was
5
6 237 performed for each group of factor (*vegan* and *ggord* package) using specific abundances. Statistics were
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8 238 performed with R software (v3.3.0) and GIS analysis with QGIS (v2.18.3).
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10 239 4. Results

11 240 4.1. Objective (1): Qualifying and quantifying amphibian communities

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14 241 The presence of amphibians was observed in 69 (84%) of the 82 stormwater ponds. Fourteen species
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16 242 were found (summarized in **Figure 2**), representing 78% of the 18 species known in the study area if we
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18 243 consider that the “green frogs” group counted as two species (Odonat, 2017). The four species that were not
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20 244 found were the fire salamander (*Salamandra salamandra*), the moor frog (*Rana arvalis*), which has not however
21
22 245 been seen in the study area for nine years, the common midwife toad (*Alytes obstetricans*) and the yellow-bellied
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24 246 toad (*Bombina variegata*). The species richness of stormwater ponds was 2.44 ± 0.19 species (as a reminder,
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26 247 descriptive statistics are “mean \pm standard error of the mean”). We recorded a total of 2 046 adults, with an
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28 248 average of $19.51 (\pm 3.42)$ adults per stormwater pond. Abundances differed substantially among species (**Figure**
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30 249 **2**).
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35 250 Shannon indices and Equitability index were 0.57 ± 0.06 and 0.71 ± 0.03 , respectively.
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38 251 No morphological abnormalities were found on adults. Red fox carcasses were found floating in two stormwater
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40 252 ponds. The first pond had vertical concrete banks and the second had steep banks covered with geomembrane.
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42 253 No service bank or reachable exits were present on the latter, but claw marks were observed on the
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44 254 geomembrane. The month after the study period (August 2016), we observed the death of all the tadpoles in five
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46 255 detention ponds during drought events.
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49 256 4.2. Objective (2): Identifying factors driving the amphibian community of stormwater ponds

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52 257 The proportion of explained deviance D² was high for the three RBT models, one created for each
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54 258 community parameter (global occurrence, global abundance and species richness; **Table 2**). Chlorides were the
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56 259 only pollutant kept in the statistical BRT model. They had a relative influence of 2.69% on the global amphibian
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58 260 abundance alone, with a minor negative correlation. No stormwater pond pollutants were above French legal
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261 thresholds. For sediment pollutants, chromium and PCBs never exceeded thresholds in the ponds studied. For
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2 262 other sediment pollutants, between 23.08% and 92.31% of stormwater ponds were above known sub-lethal
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4 263 thresholds. For the water fraction, only Polycyclic Aromatic Hydrocarbons (PAHs) were above known sub-lethal
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6 264 thresholds (17.5% of stormwater ponds). No characteristic of enclosures measures was correlated with any of the
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8 265 studied community parameters. Fitted functions of each factor with more than 3% relative influence are
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10 266 presented in **Figure 3** (occurrence), **Figure 4** (abundance) and **Figure 5** (species richness).

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13 267 Detrended correspondance analysis (results of significant factors per species) show that the age of stormwater
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15 268 ponds was negatively correlated with abundances of the European green toad and green frogs (**Figure 6**). The
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17 269 presence of many helophytes was negatively correlated with the presence of vegetative fragements in water. The
18
19 270 alpine newt abundance was positively correlated with presence of helophytes whereas the agile frog and the
20
21 271 marsh frog were negatively correlated with it (**Figure 7**). The European green toad abundance was positively
22
23 272 correlated with pH and possibly with the water temperature too. Similar but less clear correlations were also
24
25 273 found for the march frog and for the smooth newt (**Figure 8**). At local scale (500 m ; **Figure 9**), green frogs and
26
27 274 the European green toad abundances were positively correlated with surface of annual crops (and also at large
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29 275 scale for green frogs ; **Figure 10**). The surface of deciduous forest was positively correlated with the smooth
30
31 276 newt abundance (**Figure 9**). At large scale (5000 m), the European green toad abundance was positively
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33 277 correlated with surface of road and green frogs abundance was also positively correlated with wetlands surface
34
35 278 (**Figure 10**).

36 37 38 279 **5. Discussion**

39 40 280 5.1. The amphibian communities of stormwater ponds

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42 281 As hypothesised, most of the stormwater ponds (84%) hosted amphibians. This suggests a high
43
44 282 attractiveness of stormwater ponds for amphibians, probably driven by necessity as they cannot find suitable
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46 283 ponds (semi-natural or other). The four species that were never observed on these sites (*Salamandra*
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48 284 *salamandra*, *Rana arvalis*, *Alytes obstetricans*, *Bombina variegata*) were not expected to be seen in stormwater
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50 285 ponds as they either avoid large ponds and modified landscapes, or are forest species (Räsänen et al., 2003; Vos
51
52 286 and Chardon, 1998). A previous study showed that more than a quarter of the total numbers of individuals of the
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54 287 European green toad (*Bufo viridis*) in the study area were located in stormwater ponds (Sané and Didier,
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56 288 2007). Our study shows a similar result, as it was the species with the second highest recorded mean abundances
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58 289 in stormwater ponds. It suggests that stormwater ponds play a crucial role in the conservation of this species,
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60 290 which is strongly protected in this study area. The marsh frog (*Pelophylax ridibundus*) was also frequently found
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291 in stormwater ponds. Those two species were both pioneer species (Pagano et al., 2001; Sinsch et al., 2007), as
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2 292 observed by Scher and Thiéry (2005) in Mediterranean motorway stormwater ponds. If the European green toad
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4 293 tolerates small puddles and gravel pits, the marsh frog needs large ponds, with sufficient depth and high
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6 294 exposure to sunlight (Kuzmin et al., 2009), such as retention stormwater ponds with concrete banks and riprap
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8 295 providing basking sites. The other species, especially newts and those needing more natural conditions, were less
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10 296 abundant. Indeed, even if some stormwater ponds looked like natural ponds with helophytes, clear water,
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12 297 protected species (e.g., odonata, amphibians) and aquatic ecosystems, most (56%) were vegetation-free with
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14 298 cloudy/turbid water. The number of amphibians found in studied ponds was probably underestimated as we did
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16 299 not adjust numbers according to the species detection probability (Schmidt, 2004). However, the global number
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18 300 of individuals per pond was quite high compared to other ponds in the studied area (2.95 time higher for green
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20 301 toad ; Vacher et al., 2015). Stormwater ponds therefore host high number of adults and species with great
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22 302 interspecies variations in population, as found by Scheffers & Paszkowski (2013) and Gallagher et al. (2014).
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24 303 Shannon and Equitability values were low, suggesting long-standing populations. This is corroborated by the
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26 304 presence of amphibians in the studied stormwater ponds, whatever the age of the latter. Although our single
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28 305 breeding season observation study leads us to consider stormwater ponds as a real habitat or to have high
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30 306 ecological potentiality in the Bas-Rhin, it is necessary to monitor populations over many years and to do so in
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32 307 other landscapes. Indeed, pollutant-induced genotoxicity could significantly reduce population viability
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34 308 throughout the years (Hamer et al., 2012). This would imply that a suitable stormwater pond can at terms
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36 309 increase the extinction probability although it could contain high amphibian abundances and species richness. An
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38 310 approach based on life-history traits is needed in order to conclude about the potential trap effect of stormwater
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40 311 ponds (Sinsch et al., 2007). This approach should compare at least reproductive success between stormwater
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42 312 ponds and controls. In the current study, we initially wanted to include semi-natural ponds as controls but they
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44 313 were located in too different landscapes associated with other communities, or their number were too low to be
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46 314 significant. Therefore, we can't exclude an ecological trap effect and caution is needed. However, the study
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48 315 highlights the relevance of studying the ecological potentiality of stormwater ponds as they can be the last
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50 316 wetlands remaining in highly modified landscapes.

5.2. Factors driving the amphibian community of stormwater ponds

56 318 Despite the known negative impacts of pollutants (Brand et al., 2010; Brand & Snodgrass, 2010; Collins
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58 319 & Russell, 2009; Gallagher et al., 2014; Karraker, Gibbs, & Vonesh, 2008), the concentrations observed in this
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60 320 study were probably too low to have a significant effect. Despite our initial hypothesis, no clear evidence was
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321 found of an overall effect of pollutants on amphibian communities at the observed concentrations in stormwater
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2 322 ponds in the Bas-Rhin. This result was supported by the absence of morphological abnormalities. However, the
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4 323 method used in the present study (examination of adult abnormalities) is not as precise as the examination of
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6 324 tadpoles (Wagner et al., 2014). Further experiments should therefore be performed to examine the effect of the
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8 325 observed concentrations of pollutants on amphibian development and reproduction. pH had the highest relative
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10 326 influence on the studied community parameters (global occurrence, global abundance and species richness), with
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12 327 a negative correlation for pH values of eight to ten (**Fig. 3-5**). As only one stormwater pond had an acid water
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14 328 (<7), only the basic effect of pH was revealed by models. The negative effect of basic pH (>7) on amphibian
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16 329 communities can be due to herbicides, whose negative effects are amplified in this condition. Moreover,
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18 330 pollutant analysis performed between November 2015 and January 2016 revealed a neutral pH in stormwater
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20 331 ponds. This indicates that pH became basic during spring, probably due to nitrites from agricultural activities. As
21
22 332 these results are based on winter concentrations, the effects of nitrites may be also underestimated. However,
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24 333 detrended correspondence analysis showed a positive relation between pH and abundance of the European green
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26 334 toad (**Fig. 8**), which is consistent as the species mainly use ponds in farmlands (Michel et al., 2017 ; also shown
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28 335 here **Fig. 9**)
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31 336 Inside the “*pond design*” group, pond volume was the main predictive factor of amphibian communities.
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33 337 Volumes of less than 1 000 m³ had a significant positive linear effect on community parameters. As the depth
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35 338 was not correlated to the volume and was not retained in final models, we suggest that only the surface of
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37 339 stormwater ponds and the perimeter size were of importance for amphibian communities (Guderyahn et al.,
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39 340 2016; Morand and Joly, 1995). There were too few stormwater ponds with volumes of over 1 000 m³ to consider
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41 341 any fitted function over this threshold to be significant. Moreover, the effect of volume seems only accurate for
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43 342 global parameters as no specific correlation was observed (**Fig. 6**). The age of ponds is an interesting factor, as it
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45 343 could reflect the long-term sustainability of amphibian populations. The age of ponds showed no correlation with
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47 344 the presence of most species. However, it did show a reverse relationship with Green frogs and European green
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49 345 toad population (**Fig. 6**). The main hypothesis is that those two species colonise the stormwater ponds as soon as
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51 346 they are created, inducing a highest population in very young stormwater ponds, maybe decreasing later with the
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53 347 colonization of ponds by helophytes (**Fig. 7**). In this case, a regular dredging should favour those species.
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56 348 Until now, stormwater ponds were intended to be unreachable for any species due to the pollutants contained in
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58 349 the water. However, enclosure measures were clearly demonstrated to be ineffective against amphibians. Even
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60 350 the most overprotected stormwater ponds equipped with concrete walls hosted amphibians. Indeed, there are
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351 many ways to enter stormwater ponds. Amphibians can follow street gutters and fall into the drain network.
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2 352 Depending on the design of the pond, they can also enter via the outlet. Another hypothesis is that birds could
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4 353 carry eggs of amphibian or fish on their legs, but no publication to date validates this popular thought. However,
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6 354 local fishermen indicated us regular observation of Grey heron (*Adrea cinerea*) drop off in ponds living
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8 355 amphibians and fishes kept in their beak. Eggs may also be involuntarily carried by humans during maintenance
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10 356 process or fishing activities (some stormwater ponds were even used as stock ponds of Northern pike). Poor
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12 357 vegetation maintenance also allows tall grass to grow against the fences and walls, helping amphibians to reach
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14 358 the top. Access can also be gained through large holes left in small fences after careless maintenance work.
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16 359 Finally, fence staples can eventually fall out, creating gaps through which amphibians can pass. If the current
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18 360 exclosure measures cannot keep amphibians out of stormwater ponds, the overprotection of stormwater ponds
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20 361 has to be questioned. The cost of creating completely unreachable stormwater ponds is high. It would require
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22 362 high concrete walls, large cattle grids under portal access, bird netting, fine grids over floor drains and outlets,
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24 363 etc., and would require permanent maintenance to avoid the obstruction of hydraulic pipes and the deterioration
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26 364 of netting. Although creating an underground stormwater pond would be far less expensive, the potential
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28 365 ecological value of stormwater ponds is not to be brushed aside in the current context of modified landscape
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30 366 where the number of semi-natural ponds is limited.
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33 367 Few biotic and immediate environmental factors (such as temperature or wind during the field sessions) were
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35 368 retained in the final models. Surprisingly, the presence of fish in the ponds had no effect on community
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37 369 parameters (a similar result to Le Viol et al., 2012), despite evidence that the predation by fish negatively affects
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39 370 the occurrence of amphibians (Brown et al., 2012; McCarthy and Lathrop, 2011). It can indicate that amphibian
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41 371 species had not enough evolutionary time to avoid stormwater ponds with higher densities of predators, or that
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43 372 amphibian had no other ponds to go to. Surprisingly, abundances of odonata larvae were positively associated
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45 373 with those of amphibians despite the fact that they are tadpole predators. However, odonata larvae presence
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47 374 could highlight an overall-high water quality leading to higher number of amphibians (Corbet, 1999 in Kalkman
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49 375 et al., 2008).
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52 376 Abundant aquatic vegetation is known to be positively correlated with amphibian occurrence and species
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54 377 richness (McCarthy and Lathrop, 2011; Shulse et al., 2010; Simon et al., 2009). This was partially confirmed in
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56 378 this study, as the presence of helophytes increased global levels of amphibian abundance. However, high
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58 379 quantities of helophytes slightly decreased the amphibian abundance levels. This may be explained by an
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60 380 observational bias, because of the hard detectability of amphibians among dense vegetation rather than because
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381 of a real effect of helophytes on the amphibians. A similar effect can probably be observed for algae and
1 382 vegetative fragments.

3 383 As commonly found in the literature, the main factors driving the parameters of amphibian communities were
4 384 the landscape element factors (Babbitt, Baber, & Brandt, 2006; Bix-Raybuck et al., 2010; Malmgren, 2002;
5 385 Pillsbury & Miller, 2008; Scheffers & Paszkowski, 2013; Scher & Thiéry, 2005; Sinsch et al., 2012). They alone
6 386 explained more than half of the observed variations. Some correlations (for example with wetlands, meadows,
7 387 surrounding species richness and deciduous forests) were self-explanatory given their accordance with the
8 388 biology of amphibians (Bix-Raybuck et al., 2010; Guderyahn et al., 2016; Le Viol et al., 2012; Simon et al.,
9 389 2009). One of the main discoveries was the unexpected negative correlation between species richness and
10 390 stormwater ponds located in intermediary zone between cultivated lands and deciduous forest (Grillet et al.,
11 391 2015). A shared hypothesis can be made for intermediate areas of annual crops as the same negative correlation
12 392 was found. In fact, the landscape dynamic of the studied area could be separated into two types of landscape
13 393 elements (as ponds were never located in cities): annual crops and semi-natural elements mainly composed of
14 394 forests and wetlands. When the surface of annual crops decreased, the landscape was mainly composed of more
15 395 natural elements and potentially suitable habitats for amphibians. Thus, the greater number of amphibians could
16 396 induce a greater number of amphibians in the stormwater ponds. A similar result was observed in newts by Joly
17 397 et al., 2001. Fewer semi-natural elements were present in the landscape when the surface area of annual crops
18 398 increased, so stormwater ponds were probably the only suitable habitats for amphibians and were used as
19 399 refuges (Le Viol et al., 2012, 2009). Another hypothesis could be simply the change in species composition as
20 400 the Smooth newt was mainly observed in deciduous forests landscape and as Green frogs and the European
21 401 green toad were mainly found in farmlands (**Fig. 9**). In this case, it would suggest that an “intermediate” pond
22 402 could be less efficient to hold large amphibian diversity than fully located in farmland or in deciduous forest
23 403 ponds. On the other hand, at a local scale, roads were found to have a strong negative effect on amphibians.
24 404 Although this effect could be due to road pollutants (light, noises, chemicals), we think the barrier effect of road
25 405 played the biggest role. Indeed, no amphibians were found when ponds were totally surrounded by roads, as
26 406 reported by Scheffers and Paszkowski (2013) and Parris (2006). However, at large scale, a positive correlation
27 407 was found between land covered by roads and abundances of European green toad (**Fig. 10**), possibly due to an
28 408 higher amount of stormwater ponds (roads or industrial ones).

58 409 5.3. Recommendations

410 As enclosure measures are inefficient in preventing stormwater ponds access to amphibians, the time
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2 411 and financial means dedicated to them should be reallocated to developing a better sediment cleaning process
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4 412 and creating substitute semi-natural ponds that are not connected to road runoff. Observed pollutant
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6 413 concentrations were observed to have no effect on the studied amphibian community parameters (global
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8 414 occurrence, global abundance and species richness) ; it is therefore futile to overprotect stormwater ponds which
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10 415 have real ecological value, but caution is still required. A sensible approach could be to consider all newly
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12 416 constructed stormwater ponds polluted, and remove the enclosure measures once the low pollutant
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14 417 concentrations have been demonstrated. An easier alternative for road managers would be to install efficient
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16 418 enclosure measures or other compensatory measures only if pollutant concentration reached sub-lethal known
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18 419 thresholds after a five-year survey or before this time if very high pollutants concentrations are measured.
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21 420 The design of stormwater ponds should be systematically validated by an herpetologist to avoid creating
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23 421 potentially mortal traps for animals. For example, the unclimbable banks of two stormwater ponds in this study
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25 422 were traps for mammals and possibly for amphibians too (Chang et al., 2011; Zhang et al., 2010). Exit ladders
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27 423 should be systematically installed. As detention pond water is not permanent, all the tadpoles of these sites can
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29 424 die during droughts (Brand and Snodgrass, 2010; Gallagher et al., 2014). Although some amphibian larvae can
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31 425 accelerate their growth and development in harsh ecosystems, they cannot survive these sudden events, which
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33 426 appear to be more frequent with global warming (Morand et al., 1997; Newman, 1992). We therefore
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35 427 recommend the creation of a permanent minimum water level that can easily be dredged – not only in detention
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37 428 ponds, but also in newly created semi-natural ponds. This extension of the hydroperiod would enhance species
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39 429 occurrences and abundances (Guderyahn et al., 2016).
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42 430 Other recommendations can be found in Clevenot et al. (2018) about other factors not developed in this
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44 431 study, such as dredging and vegetation maintenance.
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47 432 **6. Conclusions**

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50 433 The present study demonstrated a high potential ecological value of stormwater ponds for amphibian
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52 434 communities in the agricultural plain of the Bas-Rhin. Although no pollutant effect was observed on studied
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54 435 community parameters, we cannot unequivocally conclude that pollutants have no effect on the viability of the
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56 436 stormwater ponds amphibian populations and therefore that stormwater ponds are not ecological traps. However,
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58 437 we have demonstrated the ineffectiveness of enclosure measures. In these conditions, we recommend avoiding
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60 438 the installation of costly partitioning measures and reallocating efforts to ecological engineering to create better
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439 ecological stormwater ponds and more semi-natural ponds in the study area to limit a possible ecological trap
1
2 440 effect. Further analysis are needed in other landscapes and over many years to generalise our results.
3

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12
13

14 445 **Author Contributions**

16
17 446 All authors approve this version of the manuscript. They agree to be accountable for the aspects of the
18
19 447 work that they conducted, and will ensure that any questions related to the accuracy or integrity of any part of
20
21 448 their work are appropriately investigated and resolved. JJ conceived the idea and design methodology; JJ and JL
22
23 449 collected the data; JJ and JL carried out data analysis; JJ, YH, AM, LP and FB organised the writing of the
24
25 450 manuscript. All authors played an active part in the drafting of the manuscript.
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Figures

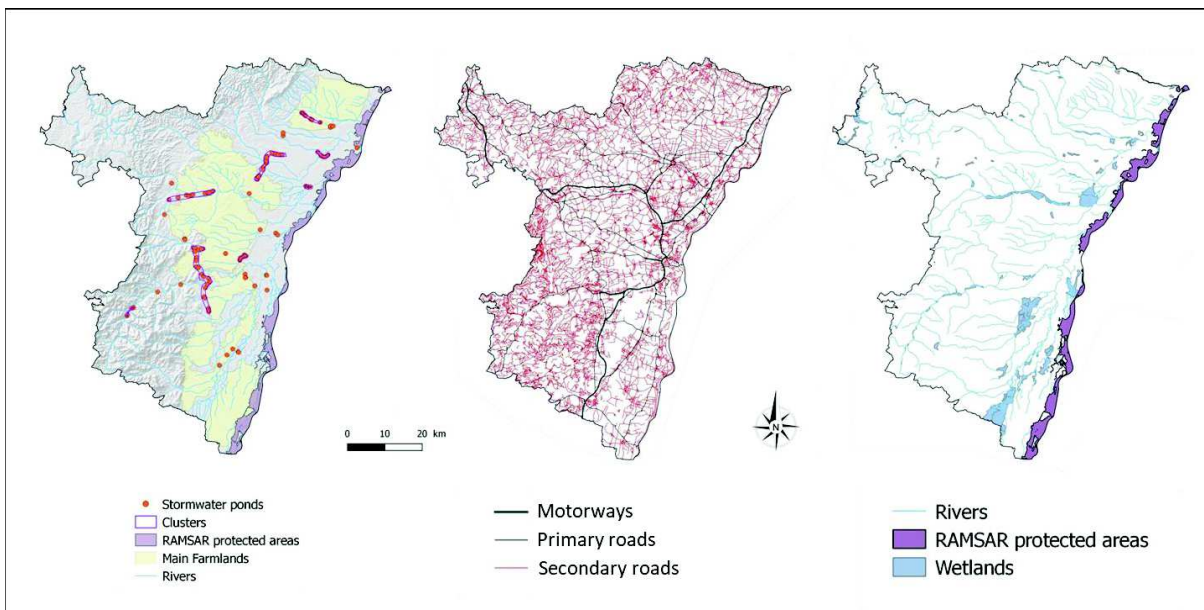


Figure 1: Bas-Rhin (Alsace, Eastern France). Map, left: location of studied stormwater ponds (orange). The stormwater ponds were grouped in clusters (purple), forming linear networks of ponds. Map, centre: motorways (wide black line), primary roads (thin black line) and secondary roads (red line) forming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

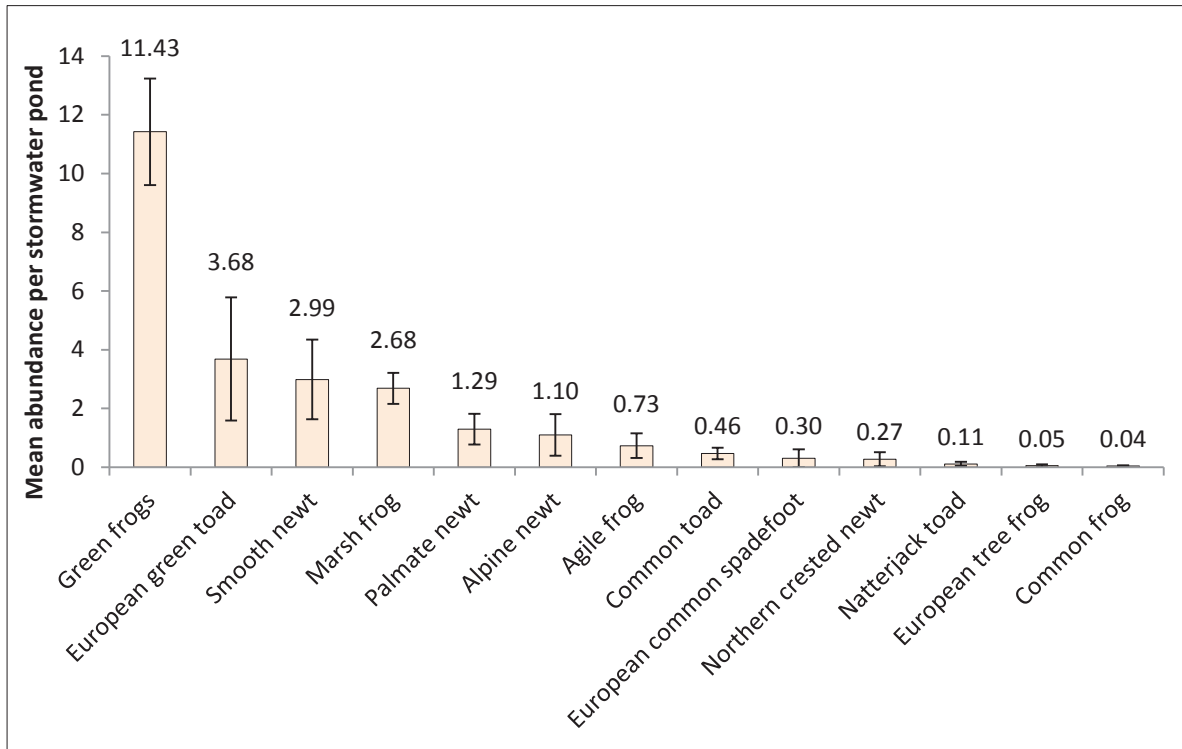


Figure 2: Specific mean abundances per stormwater pond. Error bars are standard error of the mean. Abundances are the maximal number of adult amphibians observed in a stormwater pond among the three field sessions for each species. Most observed species were Green frogs, European green toad, Smooth newt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

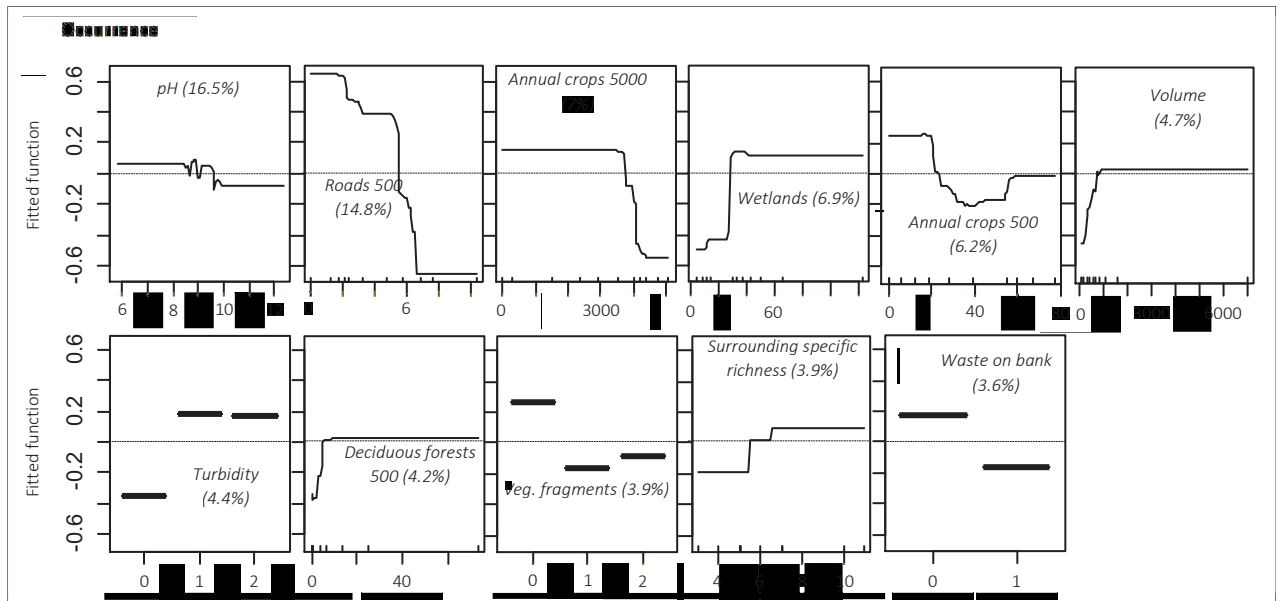


Figure 3: Fitted functions of factors with more than 3% relative influence on the deviance of the **global occurrence model**. Meaning of factors lettering is in **Tab.1**. Basic pH, roads at local scale (presumably a barrier effect), and annual crops were the main factors having negative impacts on amphibian presence in stormwater ponds. Wetlands and ponds volume were the main positive ones.

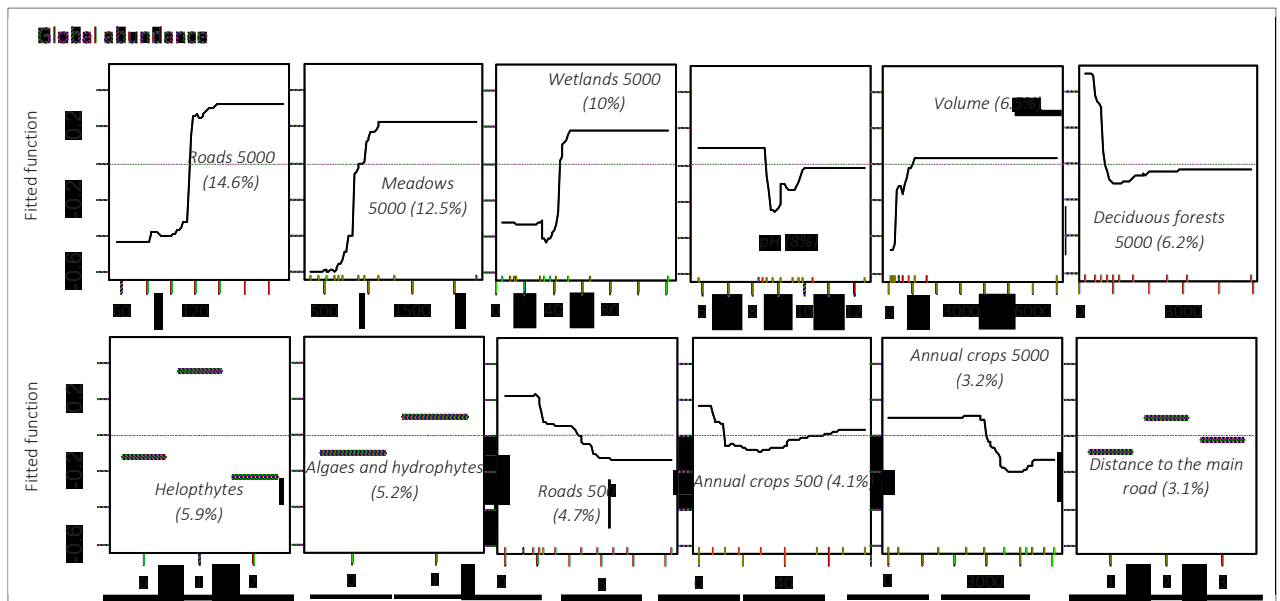


Figure 4: Fitted functions of factors with more than 3% relative influence on the deviance of the **global abundance model**. Meaning of factors lettering is in **Tab.1**. Basic pH and surface of deciduous forests at large scale were the main factors having negative impacts on amphibian abundances in stormwater ponds. Road at large scale, meadows, wetlands and ponds volume were the main positive ones.

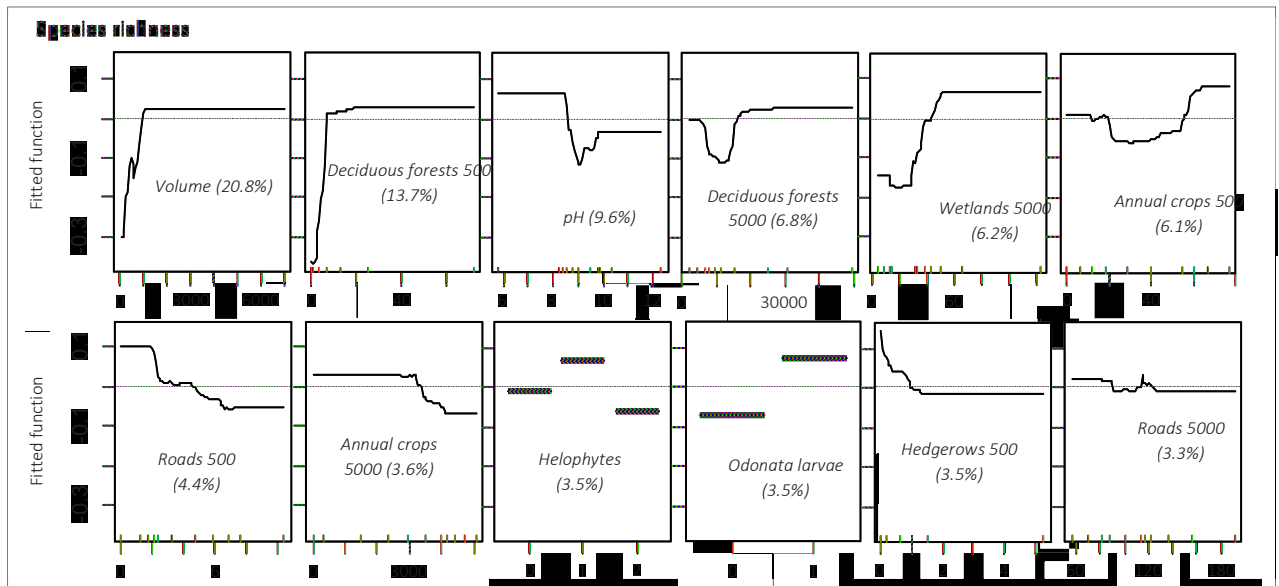


Figure 5: Fitted functions of factors with more than 3% relative influence on the deviance of the *species richness model*. Meaning of factors lettering is in **Tab.1**. Basic pH and roads at local scale were the main factors having negative impacts on amphibian abundances in stormwater ponds. Ponds volume, deciduous forests, wetlands and annual crops (possible a refuge effect) were the main positive ones.

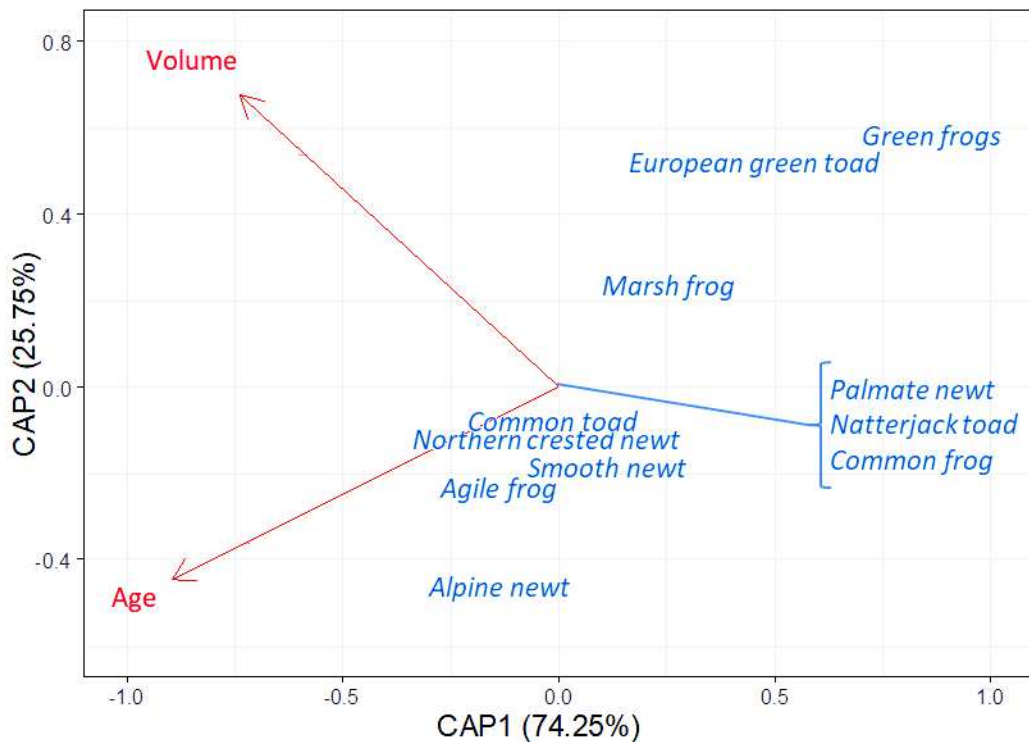


Figure 6: Correlation between species abundances (based on adults) and significant factors of Pond Design group. The abundance of a species whose name is located near the extremity of a factor is positively correlated with the factor. When the name is at the opposite, the correlation is negative. Greens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

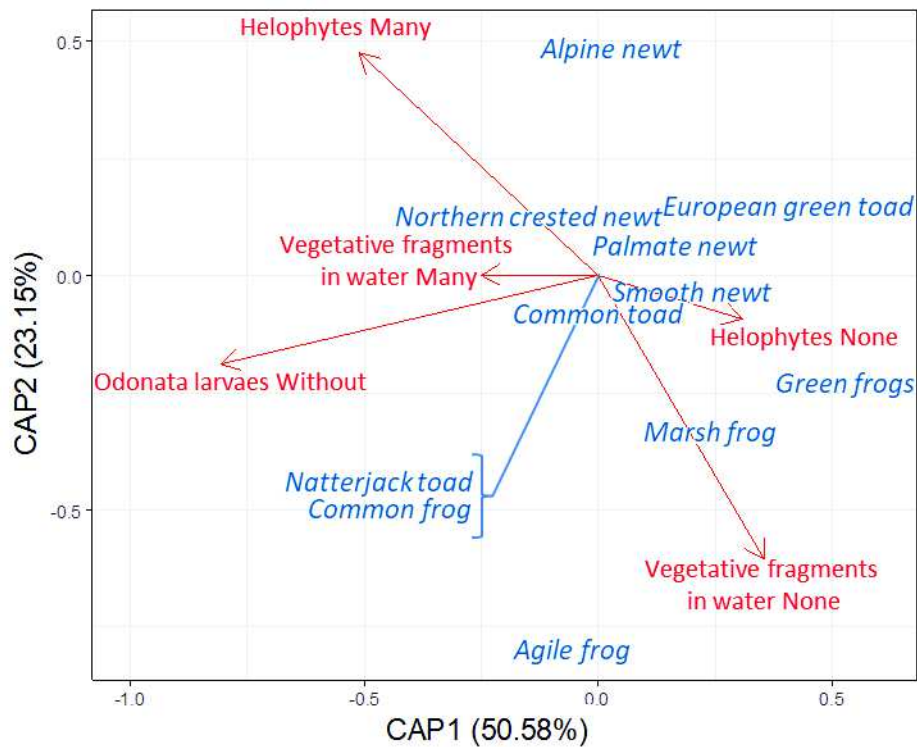


Figure 7: Correlation between species abundances (based on adults) and significant factors of Biotic factors group. The abundance of a species whose name is located near the extremity of a factor is positively correlated with the factor. When the name is at the opposite, the correlation is negative.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

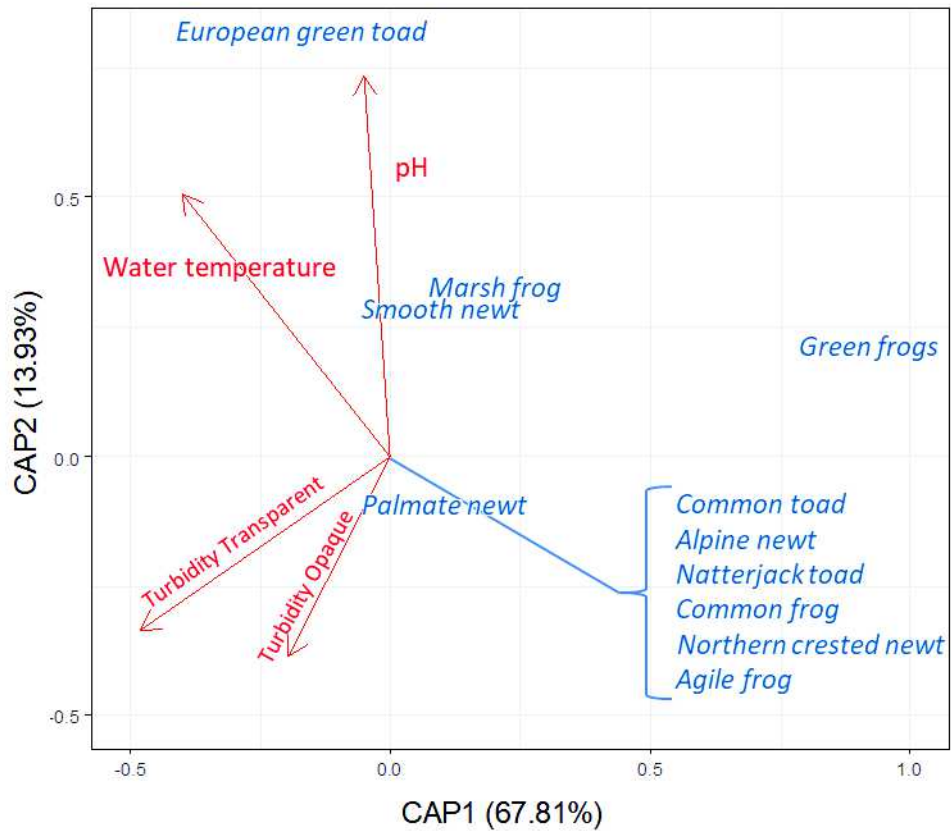


Figure 8: Correlation between species abundances (based on adults) and significant factors of immediate environmental factors group. The abundance of a species whose name is located near the extremity of a factor is positively correlated with the factor. When the name is at the opposite, the correlation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

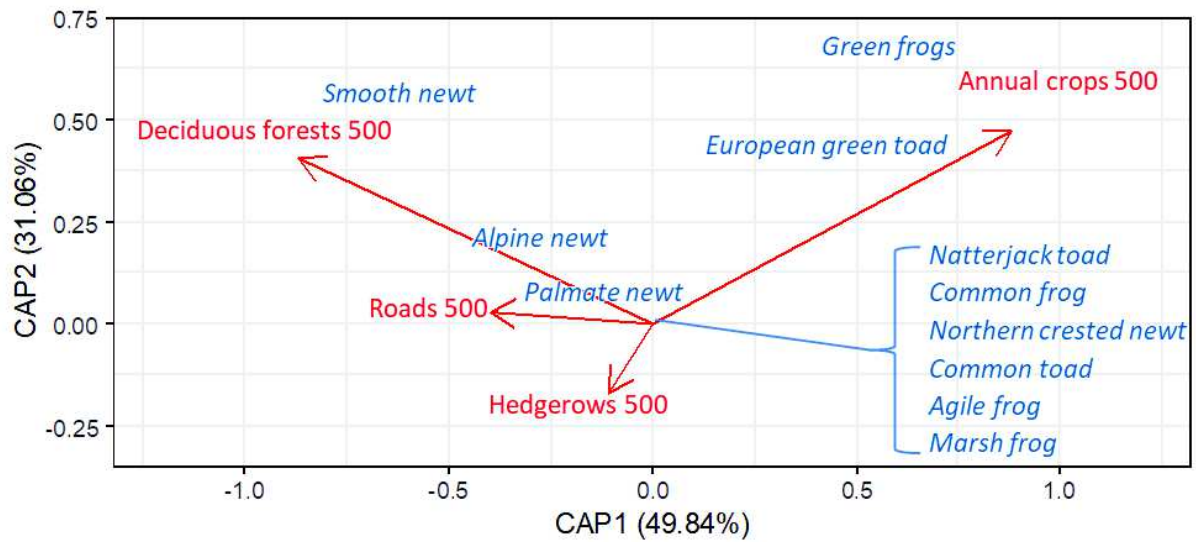


Figure 9: Correlation between species abundances (based on adults) and significant factors of Land use at Local scale group. The abundance of a species whose name is located near the extremity of a factor is positively correlated with the factor. When the name is at the opposite, the correlation is negative.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

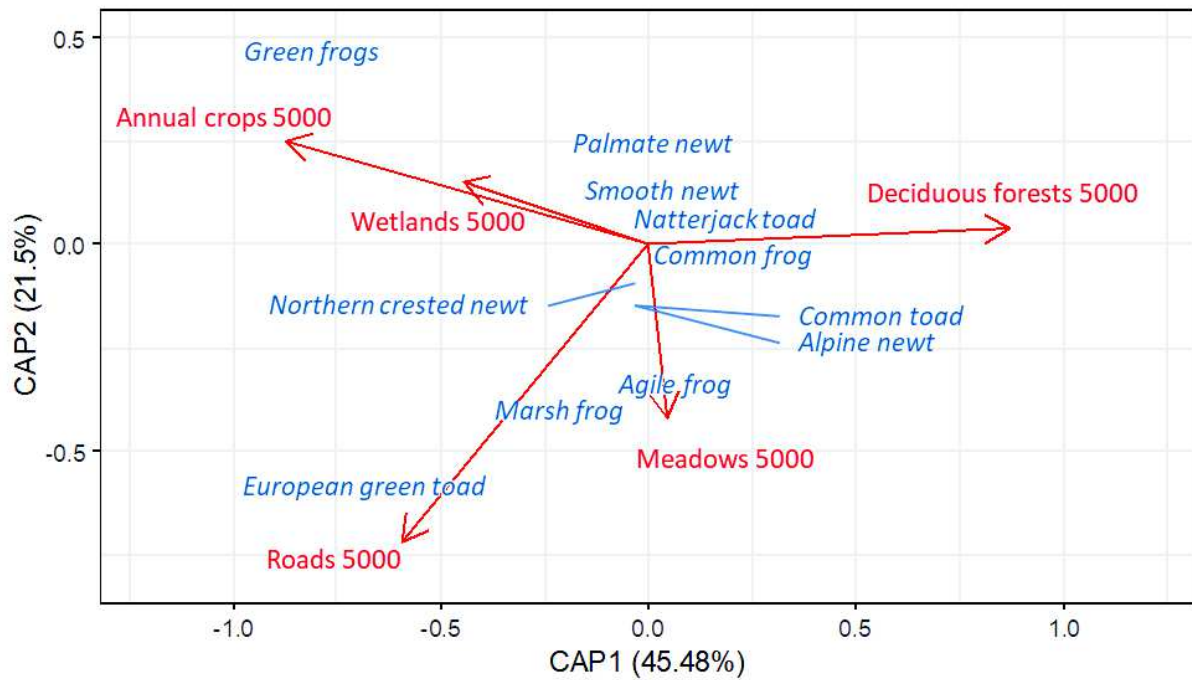


Figure 10: Correlation between species abundances (based on adults) and significant factors of Land use at Large scale group. The abundance of a species whose name is located near the extremity of a factor is positively correlated with the factor. When the name is at the opposite, the correlation is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

Tables

Table 1: Description of factors used to explain variations of dependent variables. (a): factors which initially included more classes but were pooled due to low sample size; bold: factors with sufficient sample size that were retained after correlation analyses.

Group	Factors	Min/class1	Max/class2	Median/class3	IQ range/class4	Var. Type	Measurement type
Pond design	Volume (m ³)	50	7 000	539	667	Quantitative	Data from road manager
	Depth (m)	0.3	2.5	1	0,25	Quantitative	Data from road manager
	Age (y)	0	35	8	15	Quantitative	Data from road manager
	Large fauna fence (12cm mesh)	No	Yes			Qualitative	Visual
	Small fauna fence (6.5mm mesh)	No	Yes, but without bent top	Yes, with bent top		Qualitative	Visual
	Small fauna fence condition (a)	Intact	1-2 holes >3cm	3+ holes >3cm		Qualitative	Visual
	Service gate condition (a)	Intact	With holes >3cm			Qualitative	Visual
	Cattle grid under service gate	No	Yes			Qualitative	Visual
	Concrete wall	No	Yes			Qualitative	Visual
	Pond watertightness/Permanent water	No	Yes			Qualitative	Visual
	Pond enclosure by vegetation/stagnant water	No	Yes			Qualitative	Visual
	Distance to the main road	< 3m	3 - 10m	> 10m		Qualitative	Laser telemeter
	Service bank	No	Yes			Qualitative	Visual
	Length of banks	< 3m	> 3m			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 as shelters)	None	Few	Many		Qualitative	Visual
	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 regrouped)	High			Qualitative	Visual
	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 (a)	No	Cloudy			Qualitative	Visual
Visibility (as visibility control)	<3m	3-10m	>10m		Qualitative	Visual	
Air temperature (°C)	9	24	16	8	Quantitative	thermometer	

	Water temperature (°C)	11	19.5	16.33	2	Quantitative	thermometer
	pH	5.8	12.35	9.05	1,3	Quantitative	pH-meter
Local scale (500m wide buffer) / Large scale (5000m wide buffer)	Hedgerows (ha)	0 / 14.83	5.28 / 96.72	0.15 / 50.19	0.84 / 18.65	Quantitative	SIG
	Roads (ha)	0 / 43.69	10.40 / 263.80	3.12 / 104.37	4.15 / 47.78	Quantitative	SIG
	Railways (ha)	0 / 0	3.63 / 177.49	0 / 22.31	0.54 / 23.48	Quantitative	SIG
	Meadows (ha)	0 / 329.77	47.16 / 2 538.59	11.04 / 819.07	17.14 / 500.97	Quantitative	SIG
	Vines (ha)	0 / 0	35.50 / 1 926.15	0 / 0	0 / 5.46	Quantitative	SIG
	Urban (ha)	0 / 264.60	65.90 / 3 761.17	9.32 / 929.51	18.69 / 659.77	Quantitative	SIG
	Plantations (ha)	0 / 0	6.00 / 274.36	0 / 7.87	0 / 31.01	Quantitative	SIG
	Wetlands (ha)	0 / 4.19	5.20 / 125.11	0 / 38.49	1.49 / 43.77	Quantitative	SIG
	Grasslands (ha)	0 / 0	5.05 / 211.21	0 / 12.22	0 / 23.21	Quantitative	SIG
	Moors (ha)	0 / 0.96	7.78 / 343.89	0 / 28.42	<0.01 / 59.01	Quantitative	SIG
	Ponds (ha)	0 / 0.55	7.71 / 132.56	0 / 4.79	0.10 / 7.06	Quantitative	SIG
	Annual crops (ha)	0 / 0	77.26 / 6 269.13	23.28 / 3 223.69	43.60 / 2 188.04	Quantitative	SIG
	Softwood forests (ha)	0 / 0	24.97 / 2 816.30	0 / 23.42	0 / 271.32	Quantitative	SIG
Mixed forests (ha)	0 / 0	29.77 / 1 759.29	0 / 64.57	0 / 213.64	Quantitative	SIG	
Deciduous forests (ha)	0 / 80.92	75.23 / 4 925.78	0.81 / 1 128.07	10.90 / 1 066.59	Quantitative	SIG	
Pollutants (Water / Sediment)	(µS/cm) Conductivity	64 / -	937 / -	162.6 / -	140.8 / -	Quantitative	See Tab SM
	(mg/l) Salinity	0 / -	420 / -	0 / -	127.92 / -	Quantitative	See Tab SM
	pH	5.8 / -	12.35 / -	9.05 / -	1.3 / -	Quantitative	See Tab SM
	(mg/l) Chloride	1.7 / -	107.6 / -	8.9 / -	15.6 / -	Quantitative	See Tab SM
	(mg/l) Suspended materials	0 / -	260 / -	11.45 / -	19.07 / -	Quantitative	See Tab SM
	(mg/l) Nitrates	0 / -	21.3 / -	0.77 / -	1.25 / -	Quantitative	See Tab SM
	(mg/l) Nitrites	0 / -	0.21 / -	0.03 / -	0.07 / -	Quantitative	See Tab SM
	(mg/l) PAH (mg/kgMS)	0 / 0.07 / -	0.68 / 15.51	0 / 0.53 / -	0 / 10.90	Quantitative	See Tab SM
	(mg/l) Phosphorus	0 / -	0.4 / -	0 / -	0.1 / -	Quantitative	See Tab SM
	(mg/l) Mercury (mg/kgMS)	0 / 0.01	0 / 0.34	0 / 0.13	0 / 0.06	Quantitative	See Tab SM
	(µg/l) Arsenic (mg/kgMS)	0 / <0.01	6.7 / 16.4	0.7 / 7.70	1.39 / 8.99	Quantitative	See Tab SM
	(µg/l) Cadmium (mg/kgMS)	0 / <0.01	0.02 / 95	<0.01 / 0.5	<0.01 / 0.9	Quantitative	See Tab SM
	(µg/l) Chromium (mg/kgMS)	0 / 0.1	18 / 95	1.67 / 41.6	1.32 / 69.6	Quantitative	See Tab SM
	(µg/l) Copper (mg/kgMS)	1.1 / <0.01	23.1 / 390	6.77 / 84.5	6.27 / 212.5	Quantitative	See Tab SM
	(µg/l) Nickel (mg/kgMS)	0 / 0.1	13.4 / 45.4	0 / 20.6	0 / 29	Quantitative	See Tab SM
	(µg/l) Lead (mg/kgMS)	0 / 0.12	16.9 / 118	0.74 / 40	1.7 / 69.70	Quantitative	See Tab SM
	(µg/l) Zinc (mg/kgMS)	0 / 0.2	99 / 3 085.2	25.5 / 685.4	25.25 / 1 214.2	Quantitative	See Tab SM
	Fluoranthene (µg/kgMS)	- / 0.05	- / 1748	- / 1.682	- / 414.42	Quantitative	See Tab SM
Benzo(b)fluoranthene (µg/kgMS)	- / 0.06	- / 913	- / 1.37	- / 235.01	Quantitative	See Tab SM	
Benzo(a)pyrene (µg/kgMS)	- / 0.05	- / 793	- / 23	- / 228.78	Quantitative	See Tab SM	
Sum 7 PCB (µg/kgMS)	- / 0.04	- / 29.5	- / 0.10	- / 0.01	Quantitative	See Tab SM	

Table 2: Relative influences of significant factors on deviance in each model. In line one, 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.

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)	i.e. 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 (y)	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