Original Research

Influence of Water Chemistry and Habitat Parameters on the Abundance of Pond-Breeding Amphibians

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Abstract

The aim of our study was to analyze the species composition of batrachofauna from urban water bodies and their relationship with habitat conditions. The analysis included 13 water bodies located within the administrative limits of Słupsk city. Characteristics of the water bodies included their morphometric features and physical-chemical parameters of water. Species composition and number of adult amphibians were determined during their breeding season (March-May). A total of 4 amphibian species (*Bufo bufo*, *Lissotriton vulgaris*, *Rana temporaria*, and *Rana esculenta* complex) were identified. The co-existence of all these species was documented in only one water body, and no amphibians were revealed in another five ponds. Redundancy analysis (RDA) showed positive correlations between the composition of batrachofauna and the morphometric parameters of water bodies, hardness, and water temperature. These correlations were the strongest in the case of *Rana esculenta* complex and *Rana temporaria*. Furthermore, the composition of batrachofauna correlated inversely with certain chemical parameters of the water, such as the concentration of nitrogen and total phosphorus, and N:P ratio. Moreover, we observed the inverse correlation between the prevalence of *Rana temporaria*, *Rana esculenta*, and *Lissotriton vulgaris* and the concentration of chlorophyll *a*. In our study, the physical parameters strongly differentiating the composition and abundance of amphibians, whereas chemical parameters of water had only little influence on it.

Keywords: urban ecosystems, small water reservoirs, amphibians, habitat factors

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Introduction

Urban ecosystems can be viewed as highly fragmented, heterogenous landscapes dominated by buildings, roads, and pavement, and often lacking in substantial vegetation cover [1]. The presence of lakes or ponds in this area significantly affects the quality of life in urban space by increasing amenities, providing recreational and educational activities, and even helping to modify the urban microclimate [2, 3]. Moreover, human interaction with blue space (e.g. canals, rivers, lakes, and ponds) is very important for its social and health-related benefits [4]. Urban lakes and ponds usually are small and shallow, with an average depth of 6 m or less, and are very sensitive to water pollution and eutrophication processes [5]. Thus, urban lake and pond managers are faced with a variety of ecological problems such as: high trophy (eutrophy or hypertrophy), very low water visibility, algal blooms containing toxic cyanobacteria, heavy metals and toxic organic compounds pollution, low sanitary conditions, odors associated with the rotting processes, and, consequently, potential risk for human and animal health [5-8].

Each modification of an urban ecosystem influences the number and species diversity of wild animals living in a given environment. Although some so-called urban factors can be more favorable than natural conditions, the others hinder the survival of many species or even eliminate them from urban and suburban areas [9, 10]. Batrachofauna seem particularly susceptible to anthropogenic influences; this results from their biphasic, aquatic, and terrestrial life cycle, and specific physiology, among others [11, 12]. Therefore, the existence of amphibians in a given environment can be impossible or impaired due to the presence of certain unfavorable conditions or factors.

The most common threats include fragmentation of habitats, enhanced urbanization, formation of migration barriers (roads) [13], use of fertilizers and pesticides [14-16], and global increase in environmental pollution, which can manifest itself by increased concentration of heavy metals, biogenic substances, and particles suspended in water [11]. Furthermore, unprotected permeable skin and the lack of scales or hair in the case of adult individuals or shells in embryos make amphibians quite susceptible to climate changes and UV-B radiation [17]. Consequently, the problem of global decline in the amphibian population, highlighted in 1989 for the first time, is still actual nowadays [17-19]. According to data presented in the IUCN, Red List amphibians have the highest proportion of species on the verge of extinction among the word's vertebrates, currently estimated as one in three species [11].

The aim of this study was to verify if the prevalence and number of amphibians is associated with the quality of water bodies in Słupsk city.

Material and Methods

The urban area of Słupsk (54º28'N; 17º01'E) (Pomeranian voivodeship) encompasses about 43 km^2 and more than 96,300 inhabitants (according to 2010 data). About 50% of the urban area is built-up, and the remaining 50% is covered by woodlands, including communal forests (12%), arable lands, allotment gardens and orchards, surface waters (8%), and urban green lands (3%).

This study included 13 ponds located in the southern (pond Nos. 1-7) and northern part of the city (pond Nos. 8-13). The location of analyzed water bodies is presented in Fig. 1. Although all the analyzed water bodies are of anthropogenic origin, they were modified to various degrees. Pond Nos. 1-5 are filled with water from clay excavation holes. Water bodies Nos. 6, 7, and 8 were formed during the regulation of the bed of the Słupia river. Finally, the origin of pond Nos. 9-13 dates back to the end of the 19th century, when a weir was formed on the flood crossing the area. Most of the analyzed water bodies is subjected to periodic cleaning, and their banks are successively hardened (pilled or concreted). Only two studied objects (pond Nos. 1 and 5) can be considered as relatively natural as they were not subject to the abovementioned procedures for many years. The analyzed ponds are characterized by small water table areas, below 2 ha, and average depth up to 1.5 m.

The prevalence of breeding amphibians in the area of Słupsk was recorded between the end of March and the end of May 2010. The water bodies were monitored regularly every 5 days, and the number of individuals belonging to each species was recorded in the middle of the day. Additionally, the night controls were conducted in May in order to determine the prevalence of night species. We surveyed the ponds 16 times each (12 daily and 4 night surveys), yielding a total number of 208 survey occasions. All sites were surveyed using two methods: visual encounter surveys and auditory surveys. A typical survey period involved systematically walking two observers through a shoreline of the pond, identifying adults found opportunistically and identifying amphibian calls heard during the survey. The number of amphibians breeding in a given water body was defined as the highest value of multiple measurements. The number of *Rana temporaria* Linnaeus, 1758 was determined in a 5-10-meter distance from the shore due to great skittishness of breeding individuals. The number of *Bufo bufo* (Linnaeus, 1758) was determined directly at the shore as most of the representatives of this species are not frightened by the presence of humans. The number of adult *Lissotriton vulgaris* (Linnaeus, 1758) was determined by means of direct observation from the shore. The total number of *Rana esculenta* complex (Linnaeus, 1758) was analyzed without classification into separate species.

Material for physicochemical tests was collected in April 2010. One measurement site was defined in each of the analyzed ponds in a 3-5-meter distance from the shore. The samples were collected under the water surface. The field measurements included pH (C-315 pH-meter, Elmetron) and temperature of the water (CC-401, Elmetron), plus the level of dissolved oxygen (HI 9146 Microprocessor Dissolved Oxygen Meter, Hanna). Other laboratory parameters, i.e. the concentrations of chlorophyll *a*, total nitrogen and phosphorus concentrations, were determined in a laboratory setting with the aid of standard analytical procedures. Concentration of chlorophyll *a* (Chl *a*) was determined spectrophotometricially according to the standard method in cold water with 90% acetone [20]. Concentrations of total phosphorus (TP) and total nitrogen (TN) were determined after their oxidation to nitrates and phosphates by autoclaving and digestion in perichloric acid according to respective colorimetric methods described in Hermanowicz et al. [21], using spectrophotometer SP-830 plus Metertech.

Based on the standardized physico-chemical and morphometric data, the investigated ponds were classified by use of hierarchical cluster analysis, Ward's Euclidean distance method within Statistica® v.10. The effect of environmental variables, i.e. select physicochemical characteristics of the water and morphometric parameters of water bodies, on the qualitative and quantitative characteristics of amphibians was verified by means of redundancy analysis (RDA) [22]. We selected RDA due to linear distribution of

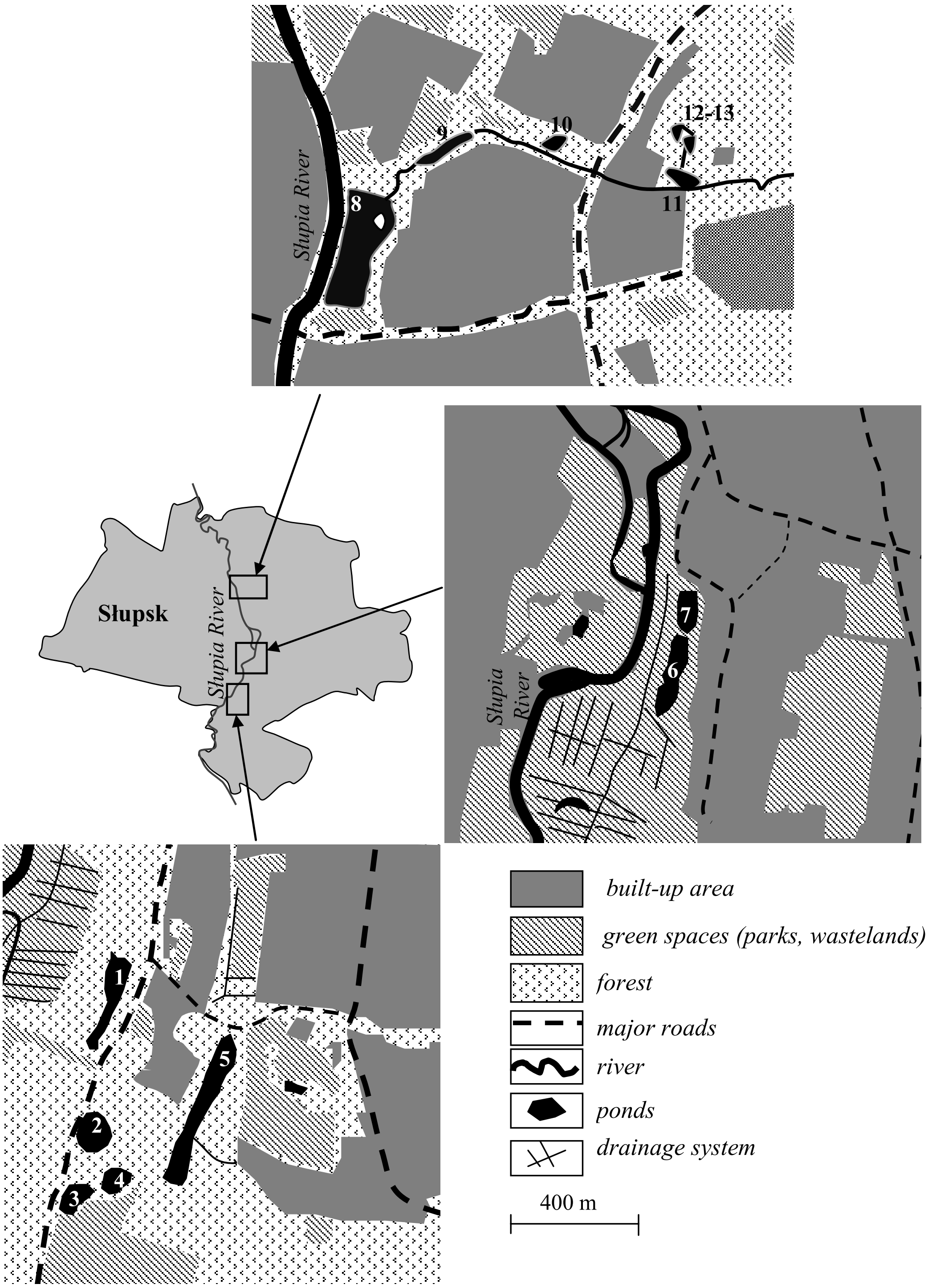

Fig. 1. Distribution of examined ponds within urban Słupsk.

Table 1. Distribution and abundance of amphibian species in the analyzed 13 ponds in Słupsk.

No.	Species and number of individuals			
	Rana temporaria Linnaeus, 1758	*Rana esculenta* complex (Linnaeus, 1758)	*Bufo bufo* (Linnaeus, 1758)	*Lissotriton vulgaris* (Linnaeus, 1758)
1	200	15	20	10
2	0	0	0	0
3	0	0	20	0
4	0	0	100	0
5	150	10	80	0
6	0	10	15	0
7	0	15	50	0
8	0	0	0	0
9	0	0	0	0
10	8	0	10	0
11	0	0	0	0
12	0	0	0	0
13	20	0	0	0

studied variables (DCA ordination; gradient length 1.939) and the fact that our material included water bodies without any amphibian species (empty samples). Furthermore, the Monte Carlo permutation test (499 permutations) was conducted to determine the significance of the model (p=0.012, F=6.65). CANOCO® 4.5 software was used for statistical calculations and graphical presentation of results.

Results and Discussion

Only four amphibian species, all of them commonly observed in Poland, were documented in the analyzed water bodies in Słupsk [23]. *Rana temporaria* and *Bufo bufo* were the most prevalent species, followed by *Rana esculenta* complex and *Lissotriton vulgaris* (Table 1). Adult amphibians were documented in 8 out of 13 analyzed ponds. However, we identified five ponds (Nos. 2, 8, 9, 11, and 12) that lacked any amphibian representatives (Table 1). A markedly higher number of amphibian species (n=11) was documented in Olsztyn by Nowakowski et al. [9]. Such a huge difference probably resulted from specific conditions of water bodies in Olsztyn (greater variability in size and type of aquatic environments) and the size of the city itself.

We identified two main groups of ponds on the basis of their similarity (Ward's method, Euclidean distance) (Fig. 2a). The first group included water bodies Nos. 1, 5, 7 and 8. These ponds were characterized by the largest area, between 0.78 ha (pond No. 1) and 1.65 ha (pond No. 5), as compared to the average area of all analyzed objects equal 0.54 ha (Table 2), and the highest values of shoreline length and shoreline development ratio, ranging between 470 m (pond No. 7) and 885 m (pond No. 5), and between 1.44 (pond No. 7), and 1.94 (pond No. 5), respectively. The second group included water bodies whose morphometric parameters did not exceed the average values for the whole sample (Table 2). It should be noted, however, that pond Nos. 10-13 differed markedly from the rest of this group. These were the smallest water bodies, with regular circular shape, area between less than 0.05 ha and 0.13 ha, and shoreline development ratio close to 1.

In turn, the cluster analysis of the physicochemical properties of the water (Fig. 2b) suggested that the analyzed ponds should be classified into four groups. The first group included water bodies Nos. 1 and 11-13 (Fig. 1). Their most distinctive feature was low concentration of chlorophyll *a*, ranging from less than 5 $mg \cdot m^{-3}$ (pond Nos. 11-13) to 15 $mg \cdot m^{-3}$ (pond No. 1), as compared to the average value for the whole sample equalling 48 $mg \cdot m^{-3}$ (Table 2). Moreover, they were characterized by relatively low concentrations of total phosphorus (between 0.046 mg $P \cdot dm^{-3}$ and 0.059 mg $P \cdot dm^{-3}$) and low levels of dissolved oxygen, especially in the case of pond Nos. 12 and 13, whose oxygen saturation approximated 6 $mg \cdot dm^{-3}$. In contrast, the second group included three ponds (Nos. 2, 3 and 4) which were characterized by the highest concentration of chlorophyll *a* (approximately 115 $mg \cdot m^{-3}$), and concentrations of nitrogen and total phosphorus amounting to 0.78-0.87 mg $N \cdot dm^{-3}$ and 0.051-0.06 mg $P \cdot dm^{-3}$, respec-

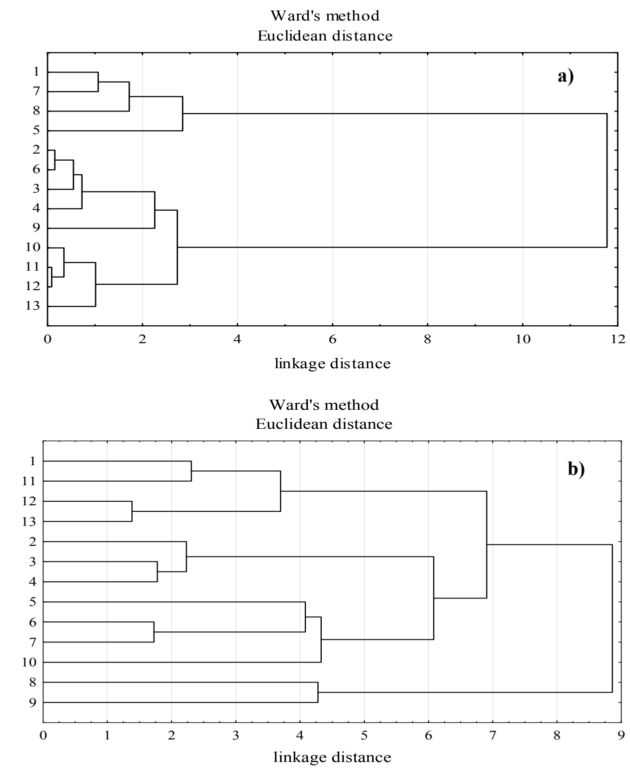

Fig. 2. Cluster diagram with urban ponds of Słupsk classification; a) – based on morphometric parameters, b) – based on physico-chemical parameters.

Table 2. Descriptive statistics of morphometric and physico-chemical variables of measured 13 ponds including mean, standard deviation (S.D.), minimum (Min.) and maximum (Max); (pond number in parentheses).

	Mean	S.D	Min.	Max
Morphometric data				
Area, [ha]	0.54	0.53	0.04(11-12)	1.65(5)
Shoreline lenght, [m]	321	224	70(11-12)	885(5)
Shoreline development	1.28	0.32	1.0(11-12)	1.94(5)
Physico-chemical data				
Temperature, [ºC]	9.6	1.4	8.1(8)	11.0(1)
pH	7.4	0.2	7.2(12-13)	7.9(8)
Dissolved oxygen, [mg $O_2 \cdot dm^{-3}$]	10.03	2.75	5.30(12)	16.63(8)
Hardness, [mval$\cdot dm^{-3}$]	9.06	1.87	6.67(9)	14.47(5)
Total nitrogen, [mg N$\cdot dm^{-3}$]	1.01	0.19	0.71(3)	1.42(8)
Total phosphorus, [mg P$\cdot dm^{-3}$]	0.067	0.016	0.046(1)	0.095(8)
N:P	15.7	3.9	10.7(7)	24.9(9)
Chlorophyll *a*, [mg$\cdot m^{-3}$]	48.1	45.1	1.4(11-12)	115.7(4)

tively (i.e. below the average levels of these biogenic substances in all analyzed water bodies). The third group (pond Nos. 5, 6 ,7, 10) comprised water bodies whose physico-chemical properties most closely resembled the average values of these parameters in the whole sample (Table 2). The latter, most distant group included two water bodies, Nos. 8 and 9 (Fig. 2b). These ponds, located in a densely built-up area, were connected with a regulated canal. They were characterized by one of the highest concentrations of total nitrogen and phosphorus (1.3-1.4 mg N$\cdot dm^{-3}$ and 0.09 mg P$\cdot dm^{-3}$, respectively – pond No. 8), the greatest oxygen saturation, up to 16.6 mg $O_2 \cdot dm^{-3}$ in water body No. 8, and the highest pH. Moreover, the water from pond Nos. 8 and 9 had the lowest temperature and hardness during the field measurements.

Using multivariate exploratory technique, we revealed that the first ordination axis, explaining 70% of the variance, was correlated positively with the morphometric parameters of water bodies (area, shoreline length, and shoreline development ratio), hardness, and temperature of water (Fig. 3.). The strongest correlations between these variables were documented in the case of *Rana esculenta* complex and *Rana temporaria*. Weak association with the first ordination axis was in turn revealed in the case of *Lissotriton vulgaris* and *Bufo bufo*. Moreover, we observed an inverse correlation between the first ordination axis and the chemical parameters of the water, such as concentrations of nitrogen and total phosphorus, and N:P ratio. Furthermore, we documented the inverse correlation between the prevalence of *Rana temporaria*, *Rana esculenta*, and *Lissotriton vulgaris,* and the concentration of chlorophyll *a*. Oxygen saturation and pH of the water had the weakest influence on the ordination diagram.

Similar effects of the size of water body, the length of its shoreline, and its variability were previously reported by Nowakowski et al. [24], who examined the prevalence of amphibians in the area of Olsztyn. Also, Kutka and Bachman [25] revealed statistically significant relationship between the area of ponds and the diversity of their batrachofauna.

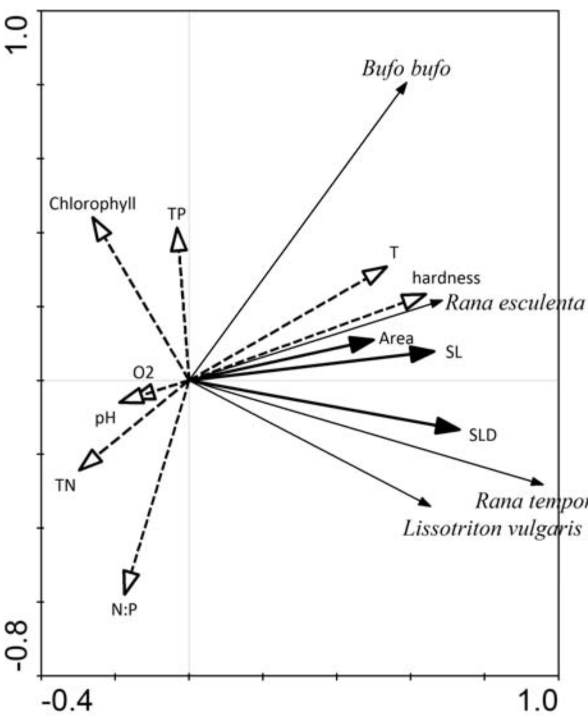

Fig. 3. Effect of environmental variables of the water bodies on the qualitative and quantitative characteristics of amphibians – RDA diagram.

Temperature of the water is an important environmental determinant of the prevalence of amphibians (Fig. 3). Too low a temperature or its sudden decrease are reflected by reduced activity or even death of breeding individuals, and prolong the duration of tadpole stage [11, 26]. Based on our analysis (RDA) we can suppose that the species most sensitive to temperature is Rana esculenta. Furthermore, an inverse relationship was observed between the concentration of biogenic substances and the prevalence of amphibians. No amphibian species were detected in ponds showing the highest concentration of biogenic substances (pond Nos. 8 and 9) located in a highly urbanized zone of the city. The amount of published research confirming the direct relationship between the concentrations of nitrogen and phosphorus and the prevalence and reproduction of amphibians is sparse [11]. According to Massal et al. [27], nitrogen pollution does not exert a significant effect on the development of amphibian embryos and larvae. However, many authors claimed the unfavorable effects of agriculture, fertilizers [14-16], and superficial runoff from agricultural lands [e.g. 28].

Performed redundancy analysis suggests that in the case of measured ponds, water pH exerted virtually no effect on the prevalence of amphibians and their number (Fig. 3). However, many authors emphasized the negative effect of environmental acidification on the existence and growth of amphibians [11, 28, 29]. Perhaps the lack of evident effect of pH on the batrachofauna of ponds analyzed in our study resulted from similar, neutral pH of the water in all 13 water bodies, approximating 7.4±0.2 (Table 2). According to literature, this is lower pH of the water (below 5), which is a strong limiting factor for amphibian survival [28, 30].

Although according to many authors the association between the prevalence of amphibians and water chemistry is quite weak [30-32], it is generally accepted that a high degree of water pollution, e.g. with heavy metals, is associated with decreased reproductive potential of amphibians and exerts a negative effect on their number and diversity of species [31, 32]. Consequently, the availability of bodies with optimal chemical composition of the water and plant structure is critical for the survival of amphibians in an urban environment [33].

Certain amphibian species live in the close neighborhoods of Słupsk but do not colonize the urban area. The list of these species includes *Bombina bombina*, *Pelobates fuscus*, *Epidalea calamita*, and *Rana arvalis* [34]. Also, the occurrence of Hyla arborea was observed in more distant areas [35]. However, these species are less prevalent and frequently show specific habitat requirements. Many authors have observed that a higher urbanization rate is reflected by reduced numbers of amphibian species [9, 11, 18, 29]. Various species respond differently to enhanced urbanization. Amphibian species with relatively low requirements seem better predisposed to survival in an urban and suburban setting. Therefore, the conservation strategies of amphibians living in urban and suburban areas should be oriented at the prevention of further loss and degradation of both terrestrial and aquatic habitats.

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