

Assessment of the Conservation Significance and Threats to the Amphibians and Reptiles in Three Wetlands in Southern Bulgaria, with Different Degree of Anthropogenic Influence

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Abstract. The species composition and conservation significance of the amphibians and reptiles from three wetlands in Southern Bulgaria, with different degree of anthropogenic influence - Protected Area (PA) "Zlato pole", "Tsalapitsa" Rice-Fields and "Plovdiv" Rice-Fields is presented in the current study. The registered contemporary threats for the herpetofauna in the studied wetlands are listed and discussed. The most dangerous for amphibian and reptile biodiversity and degradation of the habitats remain habitats destruction through drainage and land modification, pollution and direct killing of individuals.

Key words: threats, biodiversity, amphibians, reptiles, wetlands, Southern Bulgaria.

Introduction

Biodiversity provides several benefits to ecosystems including ecological efficiency, ecosystem stability and ecosystem productivity, among others (CARDINALE *et al.*, 2012). Amphibians and reptiles are an abundant and diverse component of many terrestrial and freshwater ecosystems, contributing to a diverse range of ecological functions (POUGH *et al.*, 2004; WELLS, 2007). They play vital role in the food chains, as they maintain the balance of the food web since they consume many insects and themselves are source of food for many avian and mammalian species. Moreover, they recycle nutrients between aquatic to terrestrial environments, and removal of these creatures from any ecosystem will lead to disturbances in predator-prey dynamics, invertebrate

populations, algae communities, leaf litter decompositions, and nutrient cycling (ALI *et al.*, 2018).

Wetlands serve as critical habitat for many species of amphibians and reptiles. They depend upon a variety of wetland types, which may include marshes, swamps, bogs and fens (and their associated terrestrial lands). Although some may be permanent and others ephemeral, amphibian populations can depend on multiple wetlands within a given area. To protect these species over the long term, the variety and density of suitable habitat sites within the landscape must be preserved, along with terrestrial corridors that connect the wetlands. For many reptiles, wetlands also serve as primary habitat, supplying them with an ample source of food and habitat for breeding and nursing.

Specially adapted reptiles that are able swimmers are likely to be found in wetlands (EPA, 2016).

In response to evidence of rapid population declines and species loss of amphibians and reptiles worldwide the purpose of the current study is to identify the real and potential threats to amphibians and reptiles in three wetlands in Southern Bulgaria, with different degree of anthropogenic influence - Protected Area "Zlato pole" (Dimitrovgrad District), "Tsalapitsa" Rice-Fields and "Plovdiv" Rice-Fields (Plovdiv District) and, to give some recommendations for conservation measures.

Materials and Methods

The assessment of the real and potential threats to amphibians and reptiles in three studied wetlands in Southern Bulgaria - Protected Area "Zlato pole" (Dimitrovgrad District), "Tsalapitsa" Rice-Fields and "Plovdiv" Rice-Fields (Plovdiv District) is based on systematic visits and research of different parts of the studied wetlands (Fig. 1). The field research is conducted within the territory of the studied wetlands once a month in the active for the amphibians and reptiles season - from March to November, during two-year period 2017-2018.

Amphibians and reptiles were determined visually using the field guide of Biserkov *et al.* (2007). For each species are given a valid Latin

and common name after Biserkov *et al.* (2007), Stojanov *et al.* (2011), Tzankov & Popgeorgiev (2015) and Tzankov (2016). Each observed amphibian and reptile was identified at the species level. In some cases observed amphibians and reptiles were captured by hand or using a net, loops, etc. for the more precise identification and released at the same place. Some specimens were identified by the sounds they make, their eggs or larvae and skin sheds.

The conservation status of the species in this study are presented according to the contemporary Bulgarian and European legislation (Table 1).

Results and Discussion

During the study period, three pre-designated wetlands - the "Zlato Pole" Protected Area was selected for a control due to its weak anthropogenic impact and status of protected area; rice fields near Plovdiv and rice fields near the village of Tsalapitsa, were selected as anthropogenically influenced zones. In all three areas the species composition and the conservation significance of amphibians and reptiles were studied in order to assess the real and potential conservation problems and threats for the herpetofauna.

The species composition of the recorded species of amphibians and reptiles in the three studied wetlands is presented in Table 2.

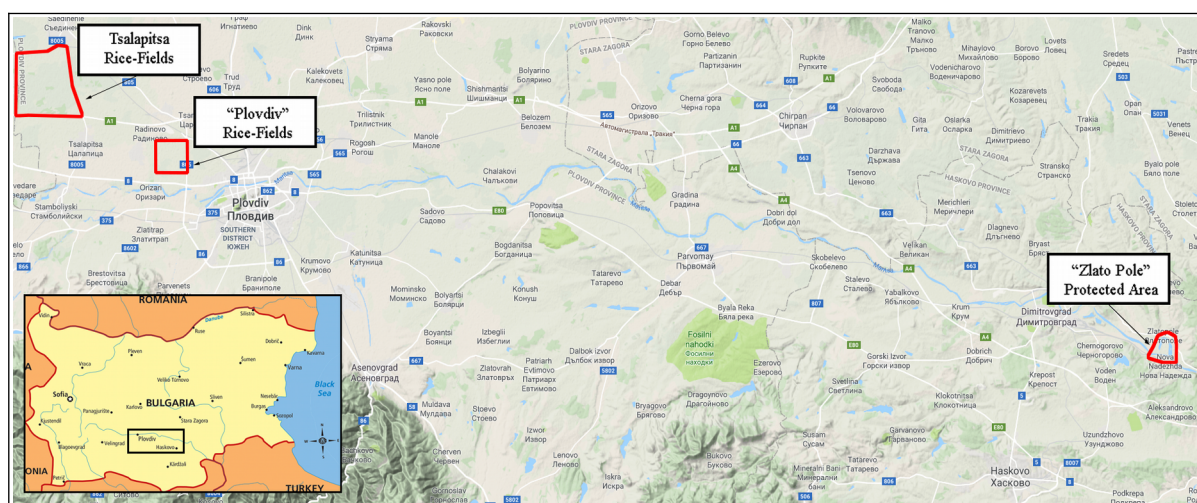


Fig. 1. Indicative map of the location of the three studied wetlands in Southern Bulgaria.

Table 1. Legal documents in the contemporary Bulgarian and European legislation, determining the conservation status of the registered species in the current study.

Abbreviation	Legal Document/Annexes	Source
BPA	Biodiversity Protection Act of Bulgaria	BPA (2002)
	Appendix II - species whose conservation requires the designation of special areas of conservation of their habitats. Appendix III - species protected in the whole of the country. Appendix IV - species under the mode of protection and regulated use of nature.	
DCE'92/43	Council Directive 92/43/EEC of 21 May 1992 on the conservation of natural habitats and of wild fauna and flora	EC (2006)
	Annex II - animal and plant species of community interest whose conservation requires the designation of special areas of conservation. Annex IV - animal and plant species of community interest in need of strict protection. Annex V - animal and plant species of community interest who is taking in the wild and exploitation may be subject to management measures.	
Bern	Convention on the Conservation of European Wildlife and Natural Habitats, Bern, 19.IX.1979	Bern Convention (1979)
	Appendix II - strictly protected fauna species (status in force since 1 March 2002). Appendix III - protected fauna species (status in force since 1 March 2002).	
IUCN	IUCN 2018. The IUCN Red List of Threatened Species. Version 2018-2.	IUCN (2018)
	NT - Near Threatened (a taxon is Near Threatened when it has been evaluated against the criteria but does not qualify for Critically Endangered, Endangered or Vulnerable now, but is close to qualifying for or is likely to qualify for a threatened category in the near future). LC - Least Concern (a taxon is Least Concern when it has been evaluated against the criteria and does not qualify for Critically Endangered, Endangered, Vulnerable or Near Threatened. Widespread and abundant taxa are included in this category).	
ERedList	European Red List of Amphibians European Red List of Reptiles	TEMPLE & COX (2009) COX & TEMPLE (2009)
	NT - Nearly Threatened - taxa that have been evaluated according to the IUCN criteria and although they do not currently meet any of the criteria for inclusion in the Critically Endangered, Threatened or Vulnerable categories, there is a lot likelihood in the near future to meet one of the criteria for these categories. LC - Least Concern - (a taxon is Least Concern when it has been evaluated against the criteria and does not qualify for Critically Endangered, Endangered, Vulnerable or Near Threatened. Widespread and abundant taxa are included in this category).	

No newts were recorded in any of the studied wetlands due to the fact that the method used does not allow it. So the analysis focuses only on the anurans and the reptiles.

Among the recorded amphibian species, the only species that has been registered in all three wetlands is the Marsh frog (*Pelophylax ridibundus*), which is the most commonly met in the three surveyed areas. Most amphibian species (6 species) were recorded in PA "Zlato Pole", the "Tsalapitsa" Rice-Fields

ranked second with 3 species and only two species were found in "Plovdiv" Rice-Fields.

From the reptiles, most species were recorded again in PA "Zlato Pole" - 6 species, in "Tsalapitsa" Rice-Fields - 5 species and in "Plovdiv" Rice Fields - 3 species.

From all three studied wetlands with the greatest species richness, with respect to the amphibians and the reptiles, stands out PA "Zlato pole", followed by "Tsalapitsa" Rice-Fields and the lowest species richness is registered in "Plovdiv" Rice-Fields.

Table 3 presents the conservation status of the amphibians and reptiles recorded in the studied wetlands according to the contemporary national and international conservation legislation. Eleven species are protected under the Biodiversity Protection Act (BPA, 2002): 2 species are included in Annex II; 10 are included in Annex III and one species is included in Annex IV (see Table 1 for more information).

One species (*Pelobates syriacus*) was included in the old edition of the Red Data Book of Bulgaria (BESHKOV, 1985) with the category "threatened". A series of studies carried out on this species have shown that *P. syriacus* has a much wider distribution than previously thought and its populations are stable. This led to the exclusion of this species from the new edition of the Red Data Book of Bulgaria in 2015 (GOLEMANSKI *et al.*, 2015). Thirteen species are included in the Convention for the Conservation of Wild European Flora and Fauna and Natural Habitats (Bern Convention, 1979); 10 are listed in Annex II as "strictly protected" and 3 are listed in Annex III. Eleven species are protected by Council Directive 92/43 (EC, 2006) on the conservation of natural habitats and of wild fauna and flora: 2 species is included in Annex II; 10 species are listed in Annex IV and one species included in Annex

V (see Table 1 for more information). All identified species are listed on the Red List of Europe (COX & TEMPLE, 2009; TEMPLE & COX, 2009), in the "LC" category - least concern (low risk) and one in category "NT" - near threatened. Thirteen species are included in the Red List of Endangered Species of the World Conservation Union (IUCN, 2018): eleven with "LC" category - least concern, one with "LR/LC" category (low risk) and one with "NT" category - near threatened. None of the recorded amphibians and reptiles is included in the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES, 1975).

The amphibians and reptiles found in the surveyed areas are characterized with high conservation significance at national and international levels. Although most of them are common species in the country, their conservation is necessary in order to preserve the native herpetofauna (as part of European and world fauna) and to preserve biodiversity in wetlands.

The main threats to amphibians and reptiles in the studied wetlands are invariably under the influence of the anthropogenic factor. Its impact will be addressed in several aspects: loss and habitat disturbance, direct anthropogenic pressure (animal killing), habitat pollution.

Table 2. Species composition of the amphibians and reptiles in PA "Zlato pole", "Tsalapitsa" Rice-Fields and "Plovdiv" Rice-Fields, recorded during the study period.

Species	PA "Zlato pole"	"Plovdiv" Rice-Fields	"Tsalapitsa" Rice-Fields
<i>Bombina bombina</i> (Linnaeus, 1761)	-	-	+
<i>Pelobates syriacus</i> Boettger, 1889	+	-	-
<i>Bufo bufo</i> (Linnaeus, 1758)	+	-	-
<i>Bufo viridis</i> complex	+	+	-
<i>Hyla arborea</i> complex	+	-	+
<i>Rana dalmatina</i> Fitzinger, 1838	+	-	-
<i>Pelophylax ridibundus</i> complex	+	+	+
<i>Emys orbicularis</i> (Linnaeus, 1758)	+	+	+
<i>Lacerta viridis</i> (Laurenti, 1768)	+	-	+
<i>Podarcis tauricus</i> (Pallas, 1814)	+	-	+
<i>Natrix natrix</i> (Linnaeus, 1758)	+	+	+
<i>Natrix tessellata</i> (Laurenti, 1768)	+	+	+
<i>Dolichophis caspius</i> (Gmelin, 1789)	+	-	-

Table 3. Conservation status of the amphibians and reptiles in PA “Zlato pole”, “Tsalapitsa” Rice-Fields and “Plovdiv” Rice-Fields, recorded during the study period. *Legend* (for more information, please see Table 1): *BPA* - Biodiversity Protection Act of Bulgaria; 92/43 - Council Directive 92/43/EEC of 21 May 1992 on the conservation of natural habitats and of wild fauna and flora (amended by Council Directive 2006/105/EC of 20 November 2006); *Bern* - Convention on the Conservation of European Wildlife and Natural Habitats, Bern, 19.IX.1979; *IUCN* - IUCN Red List of Threatened Species; *ERedList* - European Red List of amphibians and reptiles.

Species	BPA	92/43	Bern	IUCN	ERedList
<i>Bombina bombina</i> (Linnaeus, 1761)	II, III	II, IV	II	LC	LC
<i>Pelobates syriacus</i> Boettger, 1889	III	IV	II	LC	LC
<i>Bufo bufo</i> (Linnaeus, 1758)	III	-	III	LC	LC
<i>Bufotes viridis</i> complex	III	IV	II	LC	LC
<i>Hyla arborea</i> complex	III	IV	II	LC	LC
<i>Pelophylax ridibundus</i> complex	IV	V	III	LC	LC
<i>Rana dalmatina</i> Fitzinger, 1838	-	IV	II	LC	LC
<i>Emys orbicularis</i> (Linnaeus, 1758)	II, III	II, IV	II	NT	NT
<i>Lacerta viridis</i> (Laurenti, 1768)	III	IV	II	LC	LC
<i>Podarcis tauricus</i> (Pallas, 1814)	III	IV	II	LC	LC
<i>Natrix natrix</i> (Linnaeus, 1758)	-	-	III	LR/LC	LC
<i>Natrix tessellata</i> (Laurenti, 1768)	III	IV	II	LC	LC
<i>Dolichophis caspius</i> (Gmelin, 1789)	III	IV	II	LC	LC

Habitat loss, disturbance, fragmentation and isolation. In order to maintain healthy amphibian and reptile populations, wetland habitat must be protected. A watershed contains multiple habitats, all of which are affected by changes in hydrology, land use and water quality. Since no habitat is isolated from its surroundings, protection of amphibians and reptiles must take place at both the large-scale watershed level and at the smaller scale of individual wetlands (EPA, 2016). The most vulnerable to the loss of freshwater habitats are all amphibian species. They are directly dependent on the availability of suitable breeding ponds. For most of them, this is the determining factor for their distribution and development in wetlands. For *B. bufo* a potential threat may occur the drainage of water basins (especially in the rice-fields), since this species is attached to the pond, in which it has metamorphosed and when it becomes an adult it comes back to the same pond to lay its own eggs (BESHKOV *et al.*, 1986).

The Green toad (*B. viridis* complex), on the other hand, uses all sorts of breeding

ponds (KÜHNEL & KRONE, 2003). Since its breeding occurs mostly in small, temporary ponds, its eggs are the most vulnerable place in its life cycle. Often many of the eggs are destroyed due to the rapid drying or drainage of some of the water basins.

The Tree frog (*H. arborea* complex) is a species that is well adapted to the wetlands. It is not pretentious for the choice of breeding ponds and the eggs can withstand severe drought. It only suffers from pollution and the loss of suitable breeding grounds.

The rest of the registered amphibian species (*B. bombina*, *P. ridibundus* complex and *R. dalmatina*) directly depend on the availability of water for their existence. The Marsh frog is most adapted for living in natural as well as anthropogenically modified wetlands and is the most commonly met species in all three studied wetlands.

Direct anthropogenic pressure. During the study period we recorded few species killed on the adjacent roads around the wetlands. At PA “Zlato pole” - *P. syriacus* and *D.*

caspius; at “Tsalapitsa” Rice-Fields – *P. ridibundus* complex and “Plovdiv” Rice-Fields – *N. natrix*. During their spring and autumn migrations amphibians and reptiles are quite vulnerable to this type of threat, as stated in previous studies (KAMBOUROVA-IVANOVA *et al.*, 2012; MOLLOV *et al.*, 2013).

The Marsh frog (*P. ridibundus* complex) is subjected to collection by man for consumption, but most likely there is no direct danger of its disappearance. Even according to BESHKOV (1993), the demand for frogs' legs is mainly intended for the foreign market and in recent years there is no over-collecting of frogs in the study area and generally in the country.

The direct anthropogenic pressure also affects the aquatic reptiles. According to BESHKOV (1993) and according to our observations, occasionally *E. orbicularis* are caught by accident on the hooks of fishermen, and most of them are often killed afterward. The two species of aquatic snakes are also killed by fishermen because they rarely “attack” the caught fish.

Habitat pollution. Negative influence on all species directly related to water comes from the massive pollution of most wetlands. Amphibians are particularly sensitive to environmental pollutants due to their two-phase life cycle and their physiological requirements (BLAUSTEIN *et al.*, 1994). Most ponds in the rice-fields are often contaminated with a wide range of pollutants, including heavy metals, pesticides, unsolved substances, hydrocarbons and salts. Although this problem is not well researched in the studied wetlands, we assume that, except for the Marsh frog, which is proved to be species with a high ecological plasticity, which can survive even strong contamination (BESHKOV, 1993; LEONTYEVA & SEMENOV, 1999), more sensitive to this type of threat would be *B. bombina*, *R. dalmatina* and *H. arborea* complex, which could be confirmed by more detailed research on the subject.

Recommendations and conservation measures. On the basis of the results of the

present study, the following recommendations for the conservation of the diversity of amphibians and reptiles in the three studied wetlands can be proposed.

All conservation measures and efforts should be directed to preserving the habitats. It is a good idea to place information signs of conservation significance and ecology of this species in the proximity of these water basins in order to raise people's awareness of the protection of the amphibians in the wetlands.

Very negative impact is also the collection of sand and inert materials from the banks of the Maritsa River near PA “Zlato pole” (which is observed on many places along the river as well). This activity in some places may lead to destruction of the river banks, as some species are directly affected, such as *P. syriacus*, the two species of aquatic snakes and *E. orbicularis*.

A positive role for the distribution of many amphibian and reptile species has been the construction of a dense network of irrigation canals for the rice-fields. In order for this network to continue to function, it is necessary to maintain and repair it in many places.

In order to limit the harmful impact of road traffic, tunnels can be built under the road, in the places where amphibians and reptiles migrations are most often take place. If the already built infrastructure does not allow it, the only possible measure is to place signs in these places to raise the attention of the drivers.

Regular monitoring of actual and potential threats must be included in the Management Plan for PA “Zlato pole” and NATURA2000 zone “Tsalapitsa Rice-Fields”, in order to be achieved timely identification and appropriate response to eliminate the harmful effects. To deal with the problems, arising from the threats of anthropogenic origin, it is also recommended some assistance to municipalities in the neighboring settlements, in order that these problems are solved, through organizing awareness events, campaigns for better

nature and preserving biodiversity in the region.

Conclusions

From all three studied wetlands with the greatest species richness, with respect to the amphibians and the reptiles, stands out PA "Zlato pole", followed by "Tsalapitsa" Rice-Fields and the lowest species richness is registered in "Plovdiv" Rice-Fields. The studied wetlands are characterized by a number of threats with anthropogenic origin. The most dangerous for amphibian and reptile biodiversity and degradation of the habitats remain habitats destruction through drainage and land modification, pollution and direct killing of individuals.

Acknowledgements

This research was supported by Fund "Scientific research" at the Plovdiv University "Paisii Hilendarski"- Project FP17-BF-001.

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Received: 01.12.2018
Accepted: 20.12.2018