On the status of the fire-bellied toad, *Bombina bombina*, in Lobau (Vienna, Donau-Auen National Park)

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We investigated the population ecology of fire-bellied toads, *Bombina bombina*, at three study areas in Lobau, a part of the National Park Donau-Auen in Vienna, Austria. In 2008, we estimated population sizes using call surveys and recapture analysis of individually registered toads, with best estimates ranging from 115 to 650 individuals per study area. High proportions of subadult toads indicated recent successful reproduction at two of the three study sites. Yearlings gained on average 1.1 mm in snout-vent length and 0.3 g in body mass every ten days from April to August. Assessment of conservation status is hampered by the absence of data on migration and dispersal, which could allow delimitation of conservation units. We discuss implications of our results for surveillance and habitat management, stressing the importance of temporary water bodies for reproduction and recommending to focus surveys on metamorphic success.

Philippi D. & Gollmann G., 2014: Zum Erhaltungszustand der Rotbauchunke, *Bombina bombina*, in der Lobau (Wien, Nationalpark Donau-Auen). Populationsökologische Untersuchungen an der Rotbauchunke, *Bombina bombina*, wurden in drei Untersuchungsgebieten in der Lobau, dem in Wien gelegenen Teil des Nationalparks Donau-Auen, durchgeführt. Im Jahr 2008 wurden Populationsgrößen durch Ruferzählung und mittels Wiederfanganalyse individuell registrierter Unken geschätzt, wobei die besten Schätzwerte von 115 bis 650 Individuen pro Teilgebiet reichen. Ein hoher Anteil an subadulten Tieren in zwei der drei Untersuchungsgebiete zeigte erfolgreiche Fortpflanzung im Jahr davor an. Von April bis August nahmen die Jungtiere im Durchschnitt alle zehn Tage um 1.1 mm an Länge und 0.3 g an Masse zu. Die Abwesenheit von Daten zu Wanderungen und Ausbreitung erschwert die Einschätzung des Erhaltungszustands, da die Grundlage für eine Abgrenzung von Populationen fehlt. Wir erörtern Implikationen unserer Ergebnisse für Monitoring und Habitatmanagement, wobei wir die Bedeutung temporärer Gewässer für die Fortpflanzung betonen und empfehlen, besonders auf den Metamorphoseerfolg zu achten.

Keywords: Amphibia, Bombinatoridae, conservation, population ecology, recapture analysis, call surveys.

Introduction

The fire-bellied toad, *Bombina bombina* (Linnaeus, 1761) lives in lowlands of Eastern and Central Europe, reaching its western range border in Austria (Gollmann et al. 2012). It is included in Annex II of the Habitats Directive and classified as vulnerable (VU) in the Austrian Red List (Gollmann 2007a).

The main distribution areas of *B. bombina* in Austria are the floodplains along the Danube and March rivers, and Lake Neusiedler See with its surrounding wetlands (Cabela et al. 2001). Thus, most populations in Austria live in large protected areas, but only little quantitative information on them is available (Grabenhofer 2004).

Bombina bombina is widely distributed in the National Park Donau-Auen, but so far no population data were available that could support evaluation of its conservation status. Here, we present results of studies carried out in Lobau, the westernmost part of the National Park Donau-Auen, which allow quantitative estimates of the size of subpopulations and first inferences on population dynamics. We discuss implications of these findings for

the assessment of population status, as required by the Habitats Directive, and for habitat management in this urban wetland.

Methods

In 2008, field work was conducted in three study areas known as breeding sites from earlier surveys (Fig. 1). Study site 1 is an occasionally submerged reed zone adjacent to a permanent water body known as Seeschlacht. It contains several small puddles, which often are connected by narrow channels, depending on water level. Site 2, Königshaufen, consists of two large temporary ponds situated close to the oil harbour. These ponds are exposed to strong water level fluctuations, because they are situated on the riverside of the major longitudinal flood protection dam. Site 3, Göthenwasser, is a narrow, mostly shaded former side arm, which was periodically filled by emerging groundwater.

Fig. 1: Location of study sites: 1. Seeschlachtgraben, 2. Königshaufen, 3. Göthenwasser. The shading indicates the area of the Donau-Auen National Park in Vienna. Map source: Vienna GIS: www.wien.gv.at/viennagis/. – Abb. 1: Lage der Fundorte: 1. Seeschlachtgraben, 2. Königshaufen, 3. Göthenwasser. Die dunkle Schattierung zeigt die Fläche des Nationalparks Donau-Auen in Wien. Quelle: Stadt Wien - ViennaGIS: www.wien.gv.at/viennagis/.

Spawning sites were surveyed regularly from beginning of April to mid-October in intervals of approximately two weeks (Philippi 2013). During site visits, calling males were counted when calling activity seemed to be highest. Numbers were estimated in classes of five when more than ten males were calling simultaneously.

Fire-bellied toads were captured on sight with a dip net or by hand. The unique colour pattern on the ventral surface of each individual was photographed with a digital camera to allow for later identification (Nilsson 1954). Individuals already show distinct belly patterns shortly after metamorphosis. Snout-vent length was measured with plastic vernier callipers to the nearest 0.1 mm and animals were weighed on a portable digital gram scale (Voltcraft PS 250) with an accuracy of 0.05 g. Adults were sexed according to external secondary sexual characters: male *B. bombina* show nuptial pads on arms and fingers during the mating season and have internal vocal sacs.

Estimates of population size (N_E) were calculated for each month on basis of the highest number of calling males counted following the equation given by Fog et al. (2011):

$N_E = 8.526 \bullet 1.121^x$ for numbers of up to 25 calling males (x) or $N_E = 8.526 \bullet 1.121^x + 2 \bullet (x - 25)$ if the number of counted males (x) exceeded 25 individuals.

For estimation of population size from recapture data, a standard two occasion Peterson-Lincoln method modified by Chapman (in Amstrup et al. 2005) was used between two consecutive months. All capture occasions within one month were pooled and double captures of individuals excluded from the data set.

For discussion of reproductive success and movement patterns, we refer also to 9 days field work in 2007 (including observations on study sites 1 and 2; Gollmann 2007b) and to 38 days of surveys in 2009, mostly focused on the Danube crested newt (Schedl et al. 2009).

Results

A total of 505 individuals (metamorphs excluded) were registered at the three study sites over the activity season in 2008 (Tab. 1). A fraction of 30.3 % were classified as adults born in 2006 or earlier, whereas 69.7 % were classified as yearlings born in 2007 based on body size (see below). Numbers of metamorphs exceeded hundreds and thousands at two study sites, respectively. A representative number were captured and measured shortly after metamorphosis.

Numbers of calling males counted on each site visit with calling activity varied between 1 and 35 (Tab. 2). No correlation was found between the number of males registered calling

Tab. 1: Overall numbers of captured fire-bellied toads in 2008 for all study sites combined and separately. Metamorphs are not included in the number of individuals and recapture events. – Tab.1: Alle in der Saison 2008 registrierten und wiedergefangenen Unken für jede Untersuchungsfläche sowie die Gesamtzahlen. Metamorphlinge sind nicht in den Individuen- sowie Wiederfangzahlen inkludiert.

Study site	Individuals	Recaptures	Adults	Yearlings	Metamorphs
Seeschlacht	66	17	42	24	4
Königshaufen	392	153	96	296	127
Göthenwasser	47	4	15	32	47
total	505	174	153	352	178

Tab. 2: Maximum number of calling males (M_C) registered during site visits per month at the three study sites. Population size (N_E) was estimated using the formula provided by FOG et al. (2011). Population size estimates based on recapture data are also shown for comparison. – Tab. 2: Maximalanzahl gleichzeitig erfasster rufender Männchen innerhalb eines Monats (M_C). Anhand der Ruferzählungen wurde die jeweilige Populationsgröße (N_E) mittels der Formel von FOG et al. (2011) geschätzt. Zum Vergleich sind die Populationsschätzungen auf Basis der Wiederfangdaten angegeben.

Study site	April		May		June		July		Individuals captured	Population size after Chapman
	M_C	N_E	M_C	N_E	M_C	N_E	M_C	N_E		
Seeschlacht	5	30	15	95	10	53	0	NA	66	115
Königshaufen	35	969	18	133	11	60	23	236	392	650
Göthenwasser	10	53	25	296	7	38	2	21	47	NA

and males captured during the same site visit (r_S (30) = 0.06, p = 0.75; Fig. 2). Calling activity was highest from the second half of April until the beginning of June. With temporal variation in number of calling males, population size estimates fluctuate considerably over time (Tab. 2).

In May, 146 individuals were captured and released at the Königshaufen ponds. In June, 42 out of 189 captured individuals were recaptures. These numbers result in an estimated population size of 650 individuals in this time frame (lower 95 % CI: 508 individuals, upper 95 % CI: 792 individuals). At Seeschlachtgraben, 45 individuals were captured in April; among 24 individuals captured during May, nine were recaptures. Population size at this site was estimated at 115 individuals (lower 95 % CI: 68 individuals, upper 95 % CI: 162 individuals). Recapture rate at the Göthenwasser study site was too low for population size estimations.

Yearlings grew about 1.1 mm in average every ten days between April and end of August, and increased their body mass by 0.3 g on average every ten days during the same time. Thus, towards the end of the season, some yearlings reach adult body size (Fig. 3), although mean values still differ between adults and yearlings (PHILIPPI 2013). Migration of 24 year-

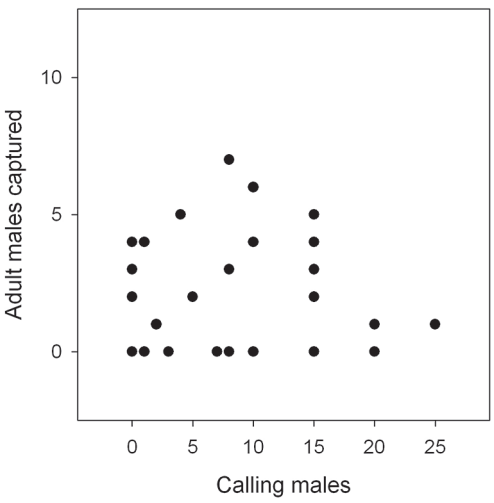

Fig. 2: Calling males counted and adult males captured during the same site visit. – Abb. 2: Vergleich der Anzahl an durch Rufzählung registrierten Männchen mit der Zahl der beim selben Besuch gefangenen adulten Männchen.

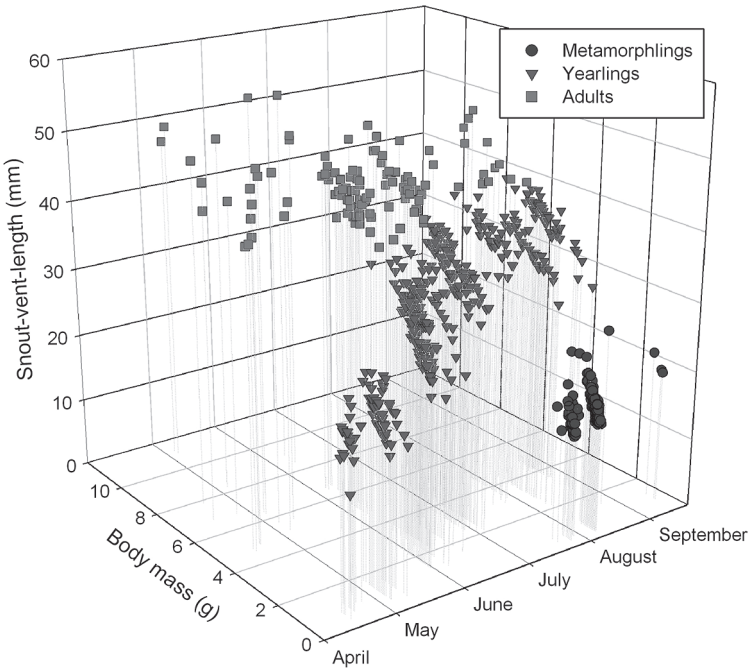

Fig. 3: Snout-vent length and body mass of every captured individual at the study site Königshaufen in 2008 (modified from Philippi 2013). – Abb. 3: Kopf-Rumpf Länge und Körpermasse von allen am Laichplatz Königshaufen in der Saison 2008 registrierten Individuen (verändert aus Philippi 2013).

lings between the two Königshaufen ponds was registered after a dry phase with very low water levels during July 2008. Seven individuals registered in 2008 at these ponds were found in a water body about 300 to 450 m away on the other side of the dam in 2009.

In 2007, a dry year, successful reproduction (metamorphosis) was observed at four sites in Lobau, including Königshaufen (Gollmann 2007b). In 2008, sucessful metamorphosis was observed at all three study sites; whereas we found only four metamorphs at Seeschlachtgraben, there were hundreds in Göthenwasser and thousands at Königshaufen.

In 2009, a wet year, adult and subadult *B. bombina* were observed in many temporary water bodies that had been dry in the years before. In June 2009, an exceptional flood introduced fish to many sites, including the Königshaufen ponds, thus strongly reducing metamorphic success of amphibian larvae. Successful metamorphosis of *B. bombina* was observed in an area just north of Seeschlachtgraben, which only became submerged with the flooding in June.

Discussion

Population structure and dynamics

Subadults grow rapidly in the season after their first hibernation. Yearlings and adults are clearly distinct in body size and mass in April and May, but these distributions merge in the summer (Fig. 3); at the same time, a new cohort of metamorphs may enter the population. Yearlings outnumbered the adults in two of our three study areas in 2008, indicating

high reproductive success in the previous year. In the same two areas, metamorphic success was also high in 2008. Recapture data between years are too scarce to allow estimation of survival rates and longevity in this population.

Our recapture analysis allowed estimation of population sizes at two study sites. Estimates based on a closed population model may overestimate population size if migrations occur during the study period. As we selected periods with high water levels and calling activities for our estimations, we presume this potential bias to be low. At Königshaufen, population size in 2008 was much higher than expected from preliminary observations in 2007, when the ponds had been dry in early spring (Gollmann 2007b). At Seeschlacht, estimated population size was also slightly higher than in 2007, but still substantially lower than indicated by observations from the late 1990s (Gollmann et al. 2013). Severe flooding in 2002 and the drought of 2003 probably reduced the numbers of many amphibian species in Lobau. Breeding success observed at several sites in 2007 and 2008, and the widespread occurrence of subadults in 2009, demonstrate the potential for recovery of *B. bombina* after this decline. In the Seeschlacht area, introduction of fish to the permanent water body during the flooding in 2002 may have been disadvantageous for the amphibian populations.

Almost no data are available on home range sizes of adults and dispersal distances of juveniles. Seasonal migrations following changes in the water table clearly occur, but their extent is unknown. The occurrence of subadults in water bodies that had been dry in the years before indicates dispersal of prereproductive individuals. Quantitative information on these movement patterns would be essential for determining conservation units, i.e. to decide whether *B. bombina* in Lobau should – for purposes of management and surveillance – better be regarded as a single population or as several independent units. To investigate genetic and demographic connectedness of subpopulations, two complementary approaches can be applied: recapture studies of individually registered toads spanning several years, and analysis of spatial genetic variation. Microsatellite markers allowing fine-scale resolution of population structure are now available for *B. bombina* (Hauswaldt et al. 2007, Dolgener et al. 2012, 2014).

Conservation status

Criteria for the assessment of population status in the frame of the Habitats Directive were proposed by Schedl (2005). The threshold value of population size for favourable status, 500 individuals, was surpassed at a single study site (Königshaufen) in 2008. As discussed above, due to the lack of data on migrations and dispersal it is unclear whether one or several management units should be discerned in Lobau. Even if we chose to distinguish several units, all numbers were probably in the favourable range in 2008 and 2009, as both the Seeschlacht and Göthenwasser sites are well connected with other habitats of *B. bombina*.

Regularity of reproduction is more difficult to evaluate, as data are too fragmentary. At two of three study sites, size distributions indicated reproductive success in two consecutive years (2007, 2008). Long-term observations are absent, however, and we probably studied some of the most productive sites. Reproductive success seems to be highest in temporary water bodies. More sampling effort across the spectrum of available habitats will be needed to support or correct this impression.

Implications for surveillance and habitat management

Numbers of callers and of captured males were not correlated in our investigation (Fig. 2). This lack of correlation may have several reasons: not all males present at a site are calling;

with more toads calling, it becomes increasingly difficult to count or estimate their number (Grabenhofer 2004); it is difficult to capture calling males, if they occupy poorly accessible positions, e.g. in dense reed stands or in deep water close to lying treetrunks. Hence, call surveys are a good method for mapping distribution of *B. bombina*, but should only be used with great caution for estimating population size. Nevertheless, population size estimates based on call surveys during days with high calling activity yielded similar values to those based on recapture analysis (Tab. 2).

Surveillance should extend through the whole activity season, as local abundance may vary greatly, depending on changes in the water table (Gollmann et al. 2013). We suggest that observations should focus on metamorphs, because knowledge of reproductive success is crucial for identifying the characteristics of source habitats in a metapopulation.

Since the river regulation in the 1870s, the wetlands of Lobau have been heavily modified, and their hydrological dynamics greatly reduced (Hohensinner et al. 2013). Due to sinking water tables and sedimentation, number and areas of water bodies have been declining (Reckendorfer et al. 2013). As a mitigation measure, a water enhancement scheme was initiated in the late 1990s, and implemented with some success (Weigelhofer et al. 2011). Whereas increased water levels are certainly beneficial for amphibians, reduced temporal variability and higher connectivity of water bodies may be disadvantageous. Further habitat management needs to consider the requirements of *B. bombina* and other protected amphibian species, especially the importance of temporary water bodies for their reproduction (Gollmann et al. 2013).

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