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Population size of endemic *Rana tavasensis* in its terra typica, Turkey

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Abstract: We applied capture–mark–recapture (CMR) methods to estimate the population size of the Tavas frog *Rana tavasensis* in its terra typica. For this purpose, we used Pollock's robust design in program MARK in the 2011–2015 breeding seasons in its terra typica. Based on the selected model, equal catchability of each individual and absence of temporary migration were found to be the most likely biological hypotheses. Population sizes were estimated as 398, 348, 275, and 117 individuals during the four study years, respectively. Annual capture probabilities were estimated to average 0.07, and annual survival rates across years averaged 0.19. The year-specific estimations showed a remarkable decline in population size and survival rates. Anthropogenic factors, such as off-road activities, recreational activities, and animal grazing, might have played a role in this decline. This trend provides us with useful knowledge for conservation and management activities.

Key words: Tavas frog, population size, amphibian decline, survival rate, Çakıroluk, Tavas, Denizli

1. Introduction

In 1989, a decline of the global amphibian population was reported at the 1st World Congress of Herpetology. The general loss of amphibian biodiversity has been well documented recently; however, we have little information on the life cycles and population trends of certain populations or species (Barinaga, 1990; Blaustein and Wake, 1990; Stuart et al., 2004; Başkale and Kaya, 2012). Understanding population dynamics and estimating demographic parameters are key issues in amphibian conservation biology (Marsh and Trenham, 2001). Capture–mark–recapture (CMR) is an accepted method for estimating population size and other parameters based on ratios of marked to unmarked individuals. CMR data obtained at different points in time from a target population can provide fundamental insights into the ecology of the species and enable the estimation of demographic parameters such as immigration, emigration, capture probability, survival rate, and population size. Without such basic information, we are unlikely to understand and recover amphibian declines (Schmidt et al., 2002; Stuart et al., 2004).

The Tavas frog is on the IUCN Red List of Threatened Species and is categorized as endangered (EN) because of its restricted geographic distribution and declining population size since 2009 (IUCN, 2016). This species is also distributed in Girdev Lake and its close vicinity west of Elmali (Max Kasparek, pers. comm. November 2008; Franzen et al., 2008), as well as in streams near the town of Kızılcabölük (Yakup Kaska, pers. comm.). The population is estimated as having a maximum of 500 mature individuals (IUCN, 2015). No further information is available about the population size, ecological requirements, or habitat characteristics of this species.

In this respect, we aimed to estimate the population size and its related parameters, and determine the demographic structure of the population. This is an important step in raising public and scientific awareness of targeted conservation efforts.

2. Materials and methods

To estimate the population size of the Tavas frog, CMR studies were performed at the terra typica of the species, which, according to official reports, inhabits a single location. Çakıroluk (37°41′N, 29°02′E) is located on Akdağ Mountain (Tavas, Denizli) and the vertical distribution of the species is approximately 1670 m above sea level. The distribution area of the Tavas frog in Çakıroluk is about 0.6 ha, and is covered by grass and irregular sparse trees. In the spring time, grass length extends to 50 cm. The ground surface is usually wet due to melting snow water, spring waters, and the Çakıroluk fountain. Although this location is far from human settlements, due to these features Çakıroluk is generally used as a camping or picnic site and

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for sheep and goat grazing. The study area is surrounded by conifer forests consisting of *Pinus nigra* and *Juniperus excelsa*.

This study was conducted during the 2011–2015 breeding seasons, and a minimum of four campaigns were performed each year by two–three persons. During the field studies, the water temperature ranged from 8 to 16 °C and the air temperature from 4 to 22 °C. Tavas frogs were captured during the day by hand or using a dip net. They were kept in a plastic container until they were photographed. Afterwards, they were released into the same habitat. Photographs in the field were taken using Nikon D5000 digital cameras. Dates and image numbers were recorded as codes for all individuals. Dorsal maculation was suitable for recognizing each individual. All images were transferred to a computer and classified into different folders. All images of the individuals in the folders were matched visually, and the images of the same individuals from different folders were recorded using Microsoft Office Picture Manager.

For analyzing the CMR data, we used Pollock's (1982) robust design to estimate population size and parameters in the program MARK v. 4.3 (White and Burnham, 1999; Cooch and White, 2004). This method enables estimating capture probabilities (p), recapture probabilities (c), and population size (N) within primary sessions. Primary sessions are separated by longer time intervals (i.e. years). It also considers that the population is open and that immigration, emigration, birth, and death occurred between primary sessions. Thus, it also permits estimating annual survival (Φ) as well as temporary emigration (γ). Under Pollock's robust design, primary sessions contain secondary sessions that are separated by a short time interval, and it is assumed that the population is effectively closed (i.e. no births, deaths, immigration, or emigration). We constructed six models to test our hypothesis, which represents a biologically alternate hypothesis. We assumed that population size N(t) and survival rates are year-specific [$\Phi(t)$], and capture and recapture probabilities are equal [$p = c$] in all six models. Each constructed model yielded capture and recapture probabilities (constant [$p(\cdot\cdot) = c(\cdot\cdot)$] or time-specific [$p(t\cdot) = c(t\cdot)$], and temporary emigration (constant [$\gamma(\cdot\cdot)$], time specific [$\gamma(t\cdot)$] or absent [$\gamma(\cdot\cdot) = 0$]). Model selection was based on Akaike's information criterion and on Burnham and Anderson (2002). To improve model selection, we calculated mean Akaike weight (w) for each model across all years. We assessed the relative importance of each parameter by adding the mean Akaike weight across all models. For the CMR models, we assumed that 1) populations are closed within years and open from year to year, 2) marks are not lost over the sampling period owing to the photo-recognition method, 3) being caught, handled, and marked once or more has no effect on an individual's subsequent chance of capture such as trap happiness or trap shyness, and 4) there is equal catchability for each individual in every sample session and catchability does not vary among individuals. We also provided that marked and unmarked animals fully mix within the habitat between sampling sessions because of the long intervals between the sampling sessions. In addition, we used the program CAPTURE under Program MARK to test the population size of each year.

3. Results

The capture histories were generated from the field surveys. Overall, the CMR study captured a total of 251 individuals. Of these captures, 40 individuals were recaptured at least two or more times. We captured a total of 118 females and 93 males. Accordingly, the male:female ratio for the Çakıroluk population was 1.27. Only six adult individuals (2 males, 4 females) were captured during five occasions in the 2015 breeding season. Therefore, the 2015 CMR data were insufficient to estimate population size for that year. Successful population size estimations were, however, obtained for the 2011–2014 breeding seasons.

Model selection indicated that the model [$\Phi(t)\ p(\cdot\cdot) = c(\cdot\cdot)\ \gamma\ (\cdot) = 0\ N(t)$] was the most appropriate for population size estimation (Table 1). The selected model [$\Phi(t)\ p(\cdot\cdot) = c(\cdot\cdot)\ \gamma\ (\cdot) = 0\ N(t)$] suggested that individuals were not affected by the marking method, had a subsequent chance of capture, and exhibited equal catchability for each individual in every sampling session. Additionally, this model explained that temporary migration was absent, i.e. individuals did not skip a breeding season and normal life activities continued each year in the same habitat.

Accordingly, population sizes were estimated as 398, 348, 275, and 117 individuals for the four consecutive years (Table 2). These results show a systematic population decline from 2011 to 2014, including in the 2015 breeding season (Table 2). The field observations revealed that this

Table 1. Candidate model selection for population size estimation in Çakıroluk. Population size (Nt) is always year-specific, and the capture and recapture probabilities are always equal. K is the number of parameters and w is the Akaike weight of the models.

Model name	AICc	ΔAIC	K	W
$\Phi(t)\ p(\cdot\cdot) = c(\cdot\cdot)\ \gamma\ (\cdot) = 0\ N(t)$	–724.49	0.000	11	0.68814
$\Phi(t)\ p(\cdot\cdot) = c(\cdot\cdot)\ \gamma\ (\cdot)N(t)$	–722.32	2.171	12	0.23246
$\Phi(t)\ p(\cdot\cdot) = c(\cdot\cdot)\ \gamma\ (t)N(t)$	–720.14	4.356	13	0.07794
$\Phi(t)\ p(t\ \cdot) = c(t\ \cdot)\ \gamma\ (\cdot)=0\ N(t)$	–711.51	12.984	23	0.00104
$\Phi(t)\ p(t\ \cdot) = c(t\ \cdot)\ \gamma\ (\cdot)N(t)$	–709.15	15.347	24	0.00032
$\Phi(t)\ p(t\ \cdot) = c(t\ \cdot)\ \gamma\ (t)N(t)$	–706.77	17.728	25	0.00010

Table 2. Population size, annual capture probabilities, annual survival rates, and their 95% confidence interval (CI). Population size and population parameter estimations are generated from the most appropriate model [$\Phi(t)\ p(\cdot\cdot) = c(\cdot\cdot)\ \gamma\ (\cdot) = 0\ N(t)$].

Parameters	Years	Estimate	SE	95% CI
Population size	2011	398	112.78	245–708
	2012	348	77.18	238–552
	2013	275	163.41	109–844
	2014	117	60.35	55–326
Annual capture probabilities	2011	0.066	0.0198	0.037–0.117
	2012	0.086	0.0205	0.054–0.136
	2013	0.038	0.0235	0.011–0.122
	2014	0.068	0.0369	0.023–0.186
Annual survival rates (Φ)	2011–2012	0.36	0.123	0.162–0.615
	2012–2013	0.15	0.113	0.027–0.506
	2013–2014	0.07	0.086	0.007–0.463

habitat has been damaged by off-road activities since autumn 2012. Thus, many adults, juveniles, larvae, and eggs may have been destroyed by off-road vehicles. In addition, recreational activities (camping sites) and animal-grazing activities had been carried out in and around this habitat.

Annual capture probabilities were estimated to average 0.07 and differed considerably among primary seasons (Table 2). Accordingly, in most cases we recaptured less than one quarter of the breeding individuals in each year. Annual survival rates across years averaged 0.193 and also varied among primary seasons (Table 2).

4. Discussion

In Turkey, the first amphibian population size study was carried out on *R. holtzi* (Baran et al., 2001). That study reported 7–11 mature individuals per m^2 on the edge of Karagöl Lake and, based on those values, concluded that a population of approximately 30,000 frogs inhabited the lake. According to that study, the common carp (*Cyprinus carpio*) disrupted the biological balance of the lake and reduced the *R. holtzi* population size by 60%–70% compared to the previous year. Subsequent studies on the population size of *R. holtzi*, using comparative estimation methods, supported this decline by stating that conservation measures had to be taken quickly for the generation to continue (Kaya et al., 2005, 2010; Yıldız and Göçmen, 2012). These studies showed that the long-term monitoring studies yielded valuable information about population dynamics. In addition, population size estimations were performed for *Pelophylax ridibundus* (Kaya and Erişmiş, 2001), *Pelophylax bedriagae* (Ayaz et al., 2007; Başkale and Kaya, 2012), *Triturus ivanbureschi* (Mermer et al., 2008), *Ommatotriton ophryticus* (Mermer et al., 2008; Başkale et al., 2013), *Bombina bombina* (Çevik et al., 2008), and *Rana macrocnemis* (Çiçek et al., 2011). Although the Tavas frog is an endemic and endangered species, there has been no published report about its population size.

In addition, other brown frogs under the subgenus Rana, such as *Rana arvalis*, *Rana dalmatina*, and *Rana macrocnemis*, have a wide distribution area in Europe and the Caucasus (Gasc et al., 1997; Tarkhnishvili and Gokhelashvili, 1999). These species have declined in their distributional range and some populations became extinct during the last decades. Habitat alteration seems to be the main threat for *Rana dalmatina* (AmphibiaWeb, 2016). *Rana arvalis* is widely distributed throughout Europe and is not considered a concern, although its range is steadily decreasing. Sas et al. (2008) suggested that *Rana arvalis* has already vanished from several localities due to habitat destruction as a result of damming and dyking. Although *Rana macrocnemis* is a common and abundant amphibian species of the Caucasus, it has declined significantly as a result of raccoons and deforestation (Tarkhnishvili and Gokhelashvili, 1999).

When the capture probabilities were examined in detail, the capture probability values were low and changed from year to year. These differences were possibly caused by changes in population size or by animal behavior being affected by anthropogenic factors. Furthermore, we estimated a remarkable decrease in survival rates after off-road activities started. Similarly, population size decreased in consecutive years. The most dramatic drop occurred in the 2014 breeding season, when the value decreased by almost a quarter compared to previous years.

There seems to be a consensus that among the factors that negatively affect amphibian populations, human activities are responsible for most of the decline. Nonetheless, the paucity of natural field experiments has limited our ability to fully identify the mechanisms of decline (Fellers and Drost, 1993). If amphibian populations naturally decrease more often than they increase, as Alford and Richards (1999) suggest, then it may be impossible to detect a real decline. Although the dynamics of local populations alone can be poor indicators, numerous studies have identified factors potentially contributing to population decline and local extinctions (Alford and Richards, 1999; Gardner, 2001; Collins and Storfer, 2003). Habitat destruction is the major problem for amphibian populations in our locality. In this respect, we identify off-road activities, recreational activities, and animal grazing in field observations, stated in interviews with local people, as anthropogenic factors that reduced the population size of the Tavas frog. Road mortality is one factor that has received little attention for amphibians (Fahrig et al., 1995; Beebee, 2013). Road traffic can crush or maim the animals directly, or roads may lead to habitat fragmentation (Blaustein et al., 1990; Mader et al., 1990; Groot and Hazebrook, 1996; Reed et al., 1996).

In many cases, effective conservation of amphibian populations is limited by the lack of species-specific ecological knowledge and by the lack of information on population structure. In this study, we detected a remarkable decline in population size and decreasing survival rates. Accordingly, we propose to help the population recover by introducing captive breeding and/or fencing the core habitat, as has been performed in similar studies conducted elsewhere (Griffiths and Pavajeau, 2008; Bowkett, 2009). Reliable amphibian monitoring studies, such as the present study, can help initiate long-term monitoring actions and provide helpful knowledge for conservation and management actions.

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