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Assessing population size and survival rate of *Pelophylax bedriagae caralitanus*, in a well-protected Nature Park in Türkiye

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Abstract: Estimating population trends provides valuable information for conservation biologists. Although there are many methods for estimating demographic rates, capture-mark-recapture (CMR) methods are known to be the most realistic method that can provide detailed data on individuals and populations, including the achievement of conservation goals. This study focused on determining the population trend of *Pelophylax bedriagae caralitanus,* Beyşehir frog using the CMR method in a protected area during the 2011–2019 breeding seasons. Our CMR data led to the selection of model-considering constant survival rates, capture/recapture probabilities, and year-specific immigration/emigration patterns [$\Phi(\cdot\cdot)\ y'(t)\ y''(t)\ p(\cdot\cdot) = c(\cdot\cdot)\ N(t)$]-as the most fitting biological hypothesis among 22 constructed models. According to the best-fitted model, 6% of all individuals in Gölcük population can be captured during each sampling occasion. The annual survival rates show low variation between years, and the mean survival rate was estimated as 0.85, that means 85% of the individuals of *Pelophylax bedriagae caralitanus* in the Gölcük population were able to live on to subsequent breeding seasons. The average population size of Gölcük population for nine consecutive years was estimated as 5094 (range 4834–5382) individuals that shows minor and acceptable levels of population size fluctuations, and slightly increasing over the years. These findings can guide future research, aiding in assessing population size changes in both protected and nonprotected areas while understanding population decline trends.

Key word: Amphibia, population trend, long-term monitoring, nature park, conservation

1. Introduction

Amphibians, are a unique group of vertebrates containing over 8689 known species, of which 7653 are frogs and toads, 815 are newts and salamanders, and 221 are caecilians (AmphibiaWeb, 2023)[1], and Amphibian population decline and loss are becoming global concerns on a growing scale (Barinaga, 1990; Wyman, 1990; Blaustein et al., 1994; Alford and Richards, 1999; Houlahan et al., 2000; Gardner, 2001; Stuart et al., 2004). Amphibians play a vital role in their ecosystems by regulating insect populations and maintaining a delicate balance within the environment. Even though amphibians have thrived for over 300 million years, an alarming number of these creatures have faced extinction in just the past two decades. 7486 amphibian species listed on IUCN Red list (2024)[2], 37 of them were extinct (EX), two species were extinct in the wild (EW), nearly 798 species are believed to have gone extinct (CR) and at least 40.7% amphibian species' population trend are declining. This indicates a probable ongoing rise in the count of endangered and extinct species (Stuart et al. 2004). Hence, it is imperative to swiftly delve into understanding the reasons behind amphibian declines and initiate conservation efforts without delay. In addition to estimating the demographic characteristics such as survival rate, capture probability and population size, Capture-Mark-Recapture (CMR) data can provide important information on population dynamics and species conservation.

Arıkan (1988) described a new taxon, *Rana ridibunda caralitana*, identifying significant differences in morphometric characteristics, colour, and pattern in the Beyşehir population. Since the day this taxon was described, significant changes have been made in its taxonomic status, and there are still questions. The studies on the morphology, karyologic, genetics, and bioacoustics of the Anatolian Lake District populations showed significant

[1]AmphibiaWeb. 2023 University of California, Berkeley, CA, USA, [online]. Website <https://amphibiaweb.org> [accessed 6 Nov 2023].

[2]IUCN (2024). The IUCN Red List of Threatened Species [online]. Website <https://www.iucnredlist.org> ISSN 2307-8235. [accessed 02 2024].

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differences between caralitana and ridibundus (Alpagut and Falakalı 1995; Jdeidi 2000; Jdeidi et al. 2001; Plötner et al. 2001). As a result of these studies, caralitana was raised to the species level. However, Bülbül et al. (2011) and Sinsch et al. (2023) examined the taxonomic relationships of the aforementioned taxon using phylogenetic and bioacoustics techniques, and they suggested that *Pelophylax caralitanus* be classified as a subspecies of *Pelophylax bedriagae*. Considering these recent studies, we accepted the Gölcük population as *Pelophylax bedriagae caralitanus* in this study. *P. b. caralitanus*, distributed only in the Lakes District region of Türkiye. This subspecies generally inhabited permanent wetlands (Ayaz et al., 2007; Başkale and Çapar 2016; Arısoy and Başkale, 2019). In addition, *P. caralitanus* has been categorized as Vulnerable (VU) because it confronts continuous risks arising from climate change, habitat loss and overexploitation while *P. bedriagae* has been categorized as Least Concern (LC) because it confronts continuous risks arising from climate change, habitat loss and overexploitation with a wider distribution area (IUCN 2024).

The first CMR study on Amphibians in Türkiye was conducted on *Rana holtzi* by Baran et al. (2001). In subsequent years, the follow-up studies were carried out using comparative estimation methods for the same population and decrease in population size was supported by emphasizing the need for swift conservation measures to ensure the continuation of the generation (Kaya et al., 2005, 2010; Yıldız and Göçmen, 2012). In addition, from the early 2000s to the present, population size estimation studies have been expanded to include different anuran and urodelan species forming important foundations for the conservation of species with priority given to endemic and endangered species (i.e. Kaya and Erişmiş, 2001; Ayaz et al., 2007; Çevik et al., 2008; Mermer et al., 2008; Çiçek et al., 2011; Başkale and Kaya, 2012; Başkale et al., 2013; Başkale and Çapar, 2016).

In this paper, we use the CMR method to estimate the annual population size, survival rates, and capture probabilities in Gölcük populations of *P. b. caralitanus*. Thus, our objectives were (1) to provide the best fitting option of population models for the population structure of *P. b. caralitanus*, (2) to obtain a robust estimate of annual survival probabilities and population size from capture, mark, and recapture data, (3) to evaluate population size fluctuations among years in a protected site.

2. Material and methods

2.1. Study sites: Gölcük Lake, a crater lake, is located in Gölcük Nature Park, Isparta, Türkiye (37°72'N, 30°49E)

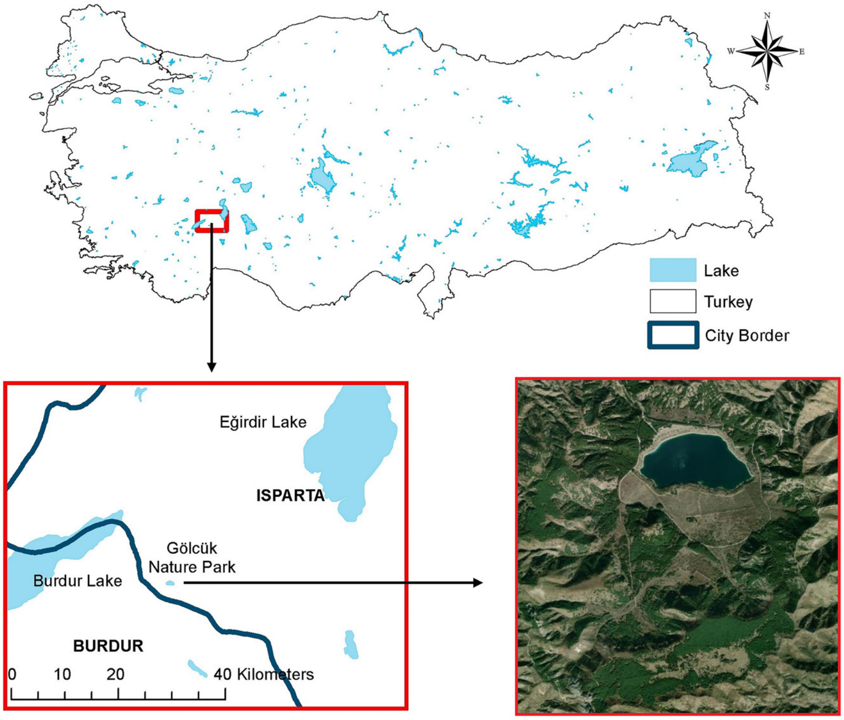

Figure 1. Location of Gölcük Nature Park Isparta, Türkiye and satellite view of Gölcük Lake, where Capture-Mark-Recapture studies were carried out.

and vertical distribution is 1378 m above the sea level (Figure 1). Gölcük Lake is not close to residential areas and is mostly used as an area where people perform their leisure activities during the daytime due to the lack of overnight accommodation facilities. The lake is a natural habitat for amphibians, and its main water sources are Karanlık Creek, Kayırlı Creek, and Koca Creek, along with underground water sources and rainfall. The surface of this lake has approximately 83 ha, 32 m depth, and a scarce vegetation cover. Gölcük Nature Park consists entirely of reforestation, predominantly featuring Black Pine (*Pinus nigra*), Scots Pine (*Pinus sylvestris*), False Acacia (*Robinia pseudoacacia*), and Lebanese Cedar (*Cedrus libani*). The surroundings of Gölcük Lake are regularly afforested, and with decomposed Salix sp. and some sections of these trees present on the lake's surface (Figure 2).

The average annual temperature of Gölcük Nature Park is 12.2 °C and the total annual precipitation is 564.0 mm (MGM, 2019). Tree frogs (*Hyla orientalis*) and green toads (*Bufotes viridis*) coexist in this lake alongside the Levantine frog. Moreover, Gölcük Nature Park hosts 9 mammal species, 67 bird species, and 99 insect species (Oğurlu et al., 2005).

Figure 2. The view of the habitat surrounding Gölcük Lake.

2.2. Field studies and recognizing the individuals: This study was conducted for eight consecutive years during the 2012–2019 breeding seasons. CMR data from 2011, was collected by Başkale et al. (2017), was also included in the population size estimations. A minimum of two, a maximum of four trapping occasions (Table 1) were conducted each year by 2–3 experts. Individuals of *P. b. caralitanus* were captured after sunset (start from 09:00 pm to 01:00 am) using torch light using a dip net. We used the photographic marking technique for recognizing individuals of *P. b. caralitanus* from their dorsal patterns (Figure 3). This technique is the cheapest and least harmful marking method for numerous amphibian species. We determined the sex of the individuals by observing their secondary sexual organs: males have tubercles on the first digit of their forelimb and a paired vocal sac on their head. Details regarding the marking of individuals and individual recognition processes are given in Başkale and Kaya (2012) and Başkale and Çapar (2016).

2.3. Statistical analyses: Using program MARK v. 4.3, we estimated Gölcük Lake's population parameters under Pollock's (1982) robust design (PRD) analysis (White and Burnham, 1999; [3]Cooch and White, 2014). PRD is one of the best estimation methods that gives intra- and interannual changes in parameters of population demography studies in long-term studies. It considers sampling sessions covering long time intervals (such as years) as an open population and referees as primary session. In this study, we configured the time interval as years for each primary session, allowing us to estimate

[3]Cooch E, White G (2014). Program MARK "A Gentle Introduction." 13th Ed. Available from: http://www.phidot.org/software/mark/docs/book .

Figure 3. The dorsal pattern of *Pelophylax bedriagae caralitanus* that captured in different trapping occasions. Arrows of different colours indicate matches of dorsal patterns of the same individual.

Table 1. The raw CMR data of the studied years at Gölcük Lake.

Years	2011*	2012	2013	2014	2015	2016	2017	2018	2019
Number of trapping occasions	4	3	3	2	3	2	2	3	3
Total number of captured individuals	1254	995	883	688	839	612	653	1025	935
Number of newly captured individuals	1142	935	831	665	792	594	631	961	880
Number of recaptured individuals	112	60	52	23	47	20	22	64	55

* CMR data was collected by Başkale et al. (2017) and reanalysed with cumulative data in this study using Pollock's (1982) robust design analysis.

annual population size, annual survival rates (Φ) as well as temporary immigration (γ') and emigration (γ"). Under the PRD, primary sessions encompass secondary sessions that are separated by a short time interval. In secondary sessions, it is assumed that the population is effectively closed, meaning there are no births, deaths, immigration, or emigration occurring within it. We designed the time intervals for secondary sessions between 7 days and 15 days within each year.

Both primary and secondary sessions data allow the construction of models which represent alternate biological hypothesis and the selection of the most appropriate model. In this context, we constructed 22 models to test our hypothesis. We made an assumption that the annual population size is time-specific [N(t)] in all models. We also fixed that capture (p) and recapture (c) probabilities are equal (p = c) due to the photo-recognition method. During the analysis, we took into account the equal capture and recapture probability parameters (p = c) are constant across years [p(··) = c(··)] or time-specific [p(*t*·) = c(t·)] during the construction of the models. The constructed models were also created with the combination and variation of the following parameters.

Temporary immigration (*γ'*): It refers to new individuals joining the population due to birth, internal migration, etc. This parameter was modelled with 3 variations; temporary immigration was constant [*γ'*(··)], time specific [*γ'*(*t*)] or absent [*γ'*(··) = 0].

Temporary emigration (*γ"*): It refers to individuals who leave the population due to reasons such as death or external migration. This parameter was modelled with 3 variations; temporary emigration was constant [*γ"*(··)], time specific [*γ"*(*t*·)] or absent [*γ"*(··) = 0].

Annual Survival rates (*Φ*): It refers to the survival rates of individuals in the population between primary seasons. This parameter was modelled with two variations; annual survival rates was constant [*Φ*(··)] or year-specific *Φ*(*t*).

We employed the Akaike's Information Criterion, this method is adjusted for small sample sizes to identify the optimal model (Burnham and Anderson 2002). To furnish more insights into the process of model selection, we computed the average Akaike weights (w) for every model over all years and by adding together the mean Akaike weights for all models, we were able to determine the relative significance of each parameter.

3. Results

The number of trapping occasions during years and the count of individuals captured and recaptured in the field surveys are given Table 1. Throughout our CMR study, we documented a sum of 7883 individuals spanning from 2011 to 2019, comprising 3797 females and 3386 males. The captured female: male ratio was calculated as 1.12 : 1 for Gölcük Lake.

The model selection suggested that the most accurate explanation for our data came from models that assumed a consistent survival rate, capture/recapture probability, and time-specific temporary immigration and emigration (Table 2). This model indicates that individuals of *P. b. caralitanus* have high survival rate and capture/recapture probability among primary sessions (years), and they also show low variation between years (Table 3). The mean survival rate was estimated as 0.85 (range = 0.64–0.98). This means that 85% of individuals can survive to the next breeding seasons. Similarly, the mean of annual capture probability was estimated as 0.062 (range = 0.059–0.066). This means that 6% of the total population can be captured during each sampling occasions.

Table 2. The constructed models selection for *Pelophylax bedriagae caralitanus* population in Gölcük Lake. K is the number of parameter; *w* is Akaike weight of the models; "*(t)*" = time specific; "(··)" = Constant; "(··) = 0" is absent. The retained model is in **bold**.

Model Number	Model name	K	AICc	ΔAIC	*w*
1	***Φ(··) y'(t) y"(t) p(··) = c(··) N(t)***	**79**	**–64,058.241**	**0.000**	**0.910**
2	*Φ(··) y'(t) y"(t) p(t) = c(t·) N(t)*	75	–64,053.558	4.683	0.874
3	*Φ(··) y'(t) y"(··) p(··) = c(··) N(t)*	81	–64,046.611	11.630	0.271
4	*Φ(··) y'(··) y"(t) p(··) = c(··) N(t)*	68	–64,010.565	47.676	0.112
5	*Φ(t) y'(t) y"(t) p(··) = c(··) N(t)*	117	–63,975.463	82.749	0.024
6	*Φ(t) y'(t) y"(t) p(t) = c(t) N(t)*	123	–63,970.286	87.995	0.000
7	*Φ(t) y'(··) y"(··) = 0 p(··) = c(··) N(t)*	109	–63,968.331	89.910	0.000
8	*Φ(t) y'(··) y"(··) p(··) = c(··) N(t)*	110	–63,954.223	104.018	0.000
9	*Φ(··) y'(··) y"(··) p(t) = c(t) N(t)*	90	–63,941.575	116.666	0.000
10	*Φ(t) y'(··) = 0 y"(··) p(··) = c(··) N(t)*	76	–63,929.576	128.666	0.000
11	*Φ(t) y'(··) = 0 y"(··) = 0 p(··) = c(··) N(t)*	76	–63,929.094	129.148	0.000
12	*Φ(··) y'(··) y"(··) p(··) = c(··) N(t)*	84	–63,921.687	136.554	0.000
13	*Φ(··) y'(··) = 0 y"(··) = 0 p(··) = c(··) N(t)*	58	–63,812.164	246.077	0.000
14	*Φ(t) y'(··) y"(t) p(··) = c(··) N(t)*	36	–63,770.379	287.863	0.000
15	*Φ(t) y'(t) y"(··) p(t) = c(t) N(t)*	37	–63,769.034	289.208	0.000
16	*Φ(··) y'(t) y"(··) p(t) = c(t) N(t)*	43	–63,761.678	296.563	0.000
17	*Φ(··) y'(··) y"(t) p(t) = c(t) N(t)*	43	–63,760.321	297.920	0.000
18	*Φ(t) y'(··) y"(··) p(t) = c(t) N(t)*	30	–63,759.850	298.391	0.000
19	*Φ(··) y'(··) = 0 y"(··) p(··) = c(··) N(t)*	44	–63,758.592	299.650	0.000
20	*Φ(t) y'(t) y"(··) p(··) = c(··) N(t)*	37	–63,752.904	305.338	0.000
21	*Φ(t) y'(··) y"(t) p(t) = c(t) N(t)*	49	–63,752.390	305.852	0.000
22	*Φ(··) y'(··) y"(··) = 0 p(··) = c(··) N(t)*	42	–63,752.282	306.902	0.000

Table 3. Population size and population parameter estimations according to the most appropriate model [$\Phi(\cdot\cdot)$ $y'(t)$ $y''(t)$ $p(\cdot\cdot) = c(\cdot\cdot)$ $N(t)$]. SE: Standard Error; CI: Confidence Interval.

Parameters	Years	Estimate	SE	95% CI
Population size	2011	5046	428.32	4294–5979
	2012	5382	638.11	4298–6818
	2013	4887	623.27	3839–6303
	2014	5128	995.99	3563–7540
	2015	4892	659.51	3791–6401
	2016	5172	1143.92	3421–8010
	2017	4834	969.07	3334–7170
	2018	5335	609.43	4295–6701
	2019	5171	642.47	4095–6634
Annual capture probabilities	2011	0.062	0.0055	0.052–0.074
	2012	0.060	0.0070	0.048–0.075
	2013	0.059	0.0076	0.046–0.076
	2014	0.060	0.0115	0.041–0.087
	2015	0.062	0.0072	0.049–0.077
	2016	0.061	0.0124	0.040–0.090
	2017	0.062	0.0126	0.042–0.092
	2018	0.064	0.0076	0.050–0.080
	2019	0.066	0.0069	0.053–0.086
Annual survival rates (Φ)	2011–2012	0.98	0.0060	0.53–1.00
	2012–2013	0.72	0.1970	0.28–0.94
	2013–2014	0.94	0.1200	0.31–0.99
	2014–2015	0.98	0.2620	0.16–1.00
	2015–2016	0.64	0.2460	0.18–0.94
	2016–2017	0.95	0.5010	0.16–1.00
	2017–2018	0.80	0.0005	0.57–1.00
	2018–2019	0.82	0.0060	0.56–1.00

The average population size for nine consecutive years was estimated as 5094 (range 4834–5382) individuals. The estimated fluctuations in the population size of *P. b. caralitanus* were distributed at an acceptable interval (Figure 4), as the fluctuations were located within the upper and lower limits in Gölcük population.

4. Discussion

Recently, there has been ample evidence of the decline in amphibian biodiversity worldwide (Arntzen et al., 2017; Scheele et al., 2019; Green et al., 2020; Brannelly et al., 2021; Womack et al., 2022). A key component of amphibian conservation biology is comprehending population dynamics and calculating demographic parameters (Marsh and Trenham, 2001). With this respect the CMR studies are a widely used method to determine population structures and trends not only in amphibians but also in fish (Bradshaw et. al., 2007; Kanno et. al., 2020), reptilians (Dyugmedzhiev et al., 2020; Rosa et al., 2022), birds (Lindberg, 2012; Lieury et al., 2017), and mammals (Hammond and Anthony, 2006; Kristensen et al., 2019).

Based on single-year data, Başkale et al. (2017) reported that the sex ratios (female: male) of *P. b. caralitanus* were calculated as 1.38 in the Gölcük population, 1.32 in the Beyşehir Lake Kuşluca location, and 1.13 in the Derebucak population in 2011. In our study, the captured female: male ratio was calculated as 1.12:1 for Gölcük population covering nine years of study (2011–2019). These differences in the sex ratio of the Gölcük population prove that long-term studies provide more stable information

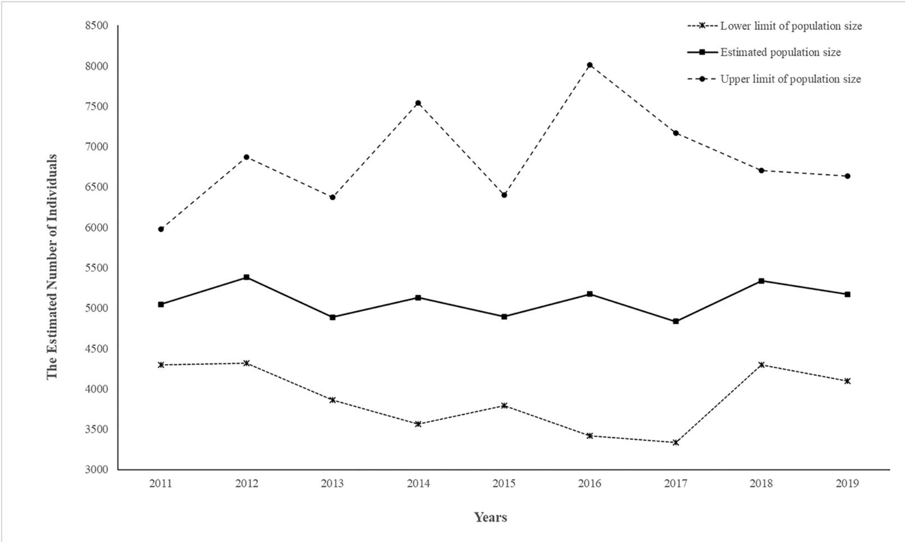

Figure 4. Annual fluctuation in population size for Gölcük population of *Pelophylax bedriagae caralitanus*.

about population dynamics. Moreover, the female:male ratios were reported that 1.27:1 for Çakıroluk population of *Rana tavasensis* (Başkale and Çapar 2016), 1.39:1 for Sülüklü Lake population of *P. bedriagae* (Ismail and Çiçek, 2017).

According to our CMR data, we constructed 22 models and analysed them to select the best fitted biological hypothesis using Pollock's (1982) robust design analysis. Model selections [*Φ(··) y'(t) y''(t) p(··) = c(··) N(t)*] for population estimate showed year-specific variation in temporary immigration and emigration. That implies the following: 1) individuals of *P. b. caralitanus* population can die between years, 2) individuals in the population may skip their reproductive periods, 3) new individuals may join the population every year due to reproduction, 4) individuals in other populations may migrate inward, 5) external migration may occur from the target population to another population. All of these phenomena are part of the ordinary life cycle in wild animals that can occur during the lifetime of individuals in natural populations (Donnelly and Guyer, 1994; Duellman and Trueb, 1994).

The best-fitted model presents the constant capture/recapture probability, meaning individuals were unaffected by the marking method, had consistent chances of subsequent capture, and displayed equal catchability in every sampling session. However, we observed very low variations in capture/recapture probabilities between years. The fluctuations on an annual basis could be responsible for alterations in the population size rather than being attributed to heterogeneity effects or animal behaviour influenced by the marking technique.

Similarly, the best-fitted model also showed that the Gölcük population of the *P. b. caralitanus* had a constant survival rate between years. Baskale et al. (2017) reported that survival rates of *P. b. caralitanus* were 0.66 for Gölcük population in 2011, 0.52 for Beyşehir-Kuşluca population in 2012 and 0.66 for Derebucak population in 2012. These survival rates are derived from the closed population model and represent the probabilities of individuals surviving within a given year (secondary session). The determined survival rates in our study refer to the rates at which individuals survive from one year to the next (primary session). The calculated high survival rates of secondary sessions showed that an average of 85% of the individuals in the population were able to live on to subsequent breeding seasons. High survival rates were reported in different populations of the species using the skeletal chronology method (Erişmiş, 2018; Arısoy and Başkale, 2019). In both studies, survival rates assumed constant for the sample of individuals across all age classes. Although the estimation method is different, the derived survival rates obtained are quite similar to this study. In addition, high annual survival rate is an indicator that factors such as habitat destruction, alien species, off-road activities, recreational activities that negatively affect amphibian populations.

Estimating population size for amphibians is becoming increasingly important to assessing current and future population structures of species. Single-year monitoring studies provide short-term data describing current status of populations. However, long-term amphibian monitoring programs are essential to comprehending complex ecosystem and population dynamics (Lindenmayer et al., 2012), and support planning for adaptive management strategies for the protection of species and/or ecosystems (Havstad and Herrick, 2003; Lindenmayer and Likens, 2009; Eyre et al., 2011). Additionally, long-term monitoring programmes may also offer valuable background information for evaluating the efficacy of management activities (Magurran et al., 2010; Dodds et al., 2012). They can establish connections to other ecological studies, generate new hypotheses (Lindenmayer and Likens, 2010; Dodds et al., 2012), and/or act as a biological indicator serving as an early warning system for the disruptions in ecological balance (Magurran et al., 2010). In this context, we gathered important information regarding population size and annual fluctuations within the scope of the *P. b. caralitanus* monitoring project, which we conducted uninterruptedly for nine years in the Gölcük Lake. It seems that in the Gölcük population of *P. b. caralitanus*, there were minor and acceptable interval fluctuations in population size within the upper and lower limits observed across different years.

Since the 1990s, the reasons for the extinction or decline of amphibians have been extensively researched and these factors are listed according to their importance (i.e. Barinaga, 1990; Alford and Richards, 1999; Gardner, 2001; Grant et al., 2016; Campbell et al., 2020). Recently, Luedtke et al. (2023) evaluated the reasons for these decreases at the global level according to the increase in the IUCN red list status of the species. This study in question delved deeper into a subset of species that had seen an elevation in their Red List categories over time. It categorized the primary drivers of deteriorating threat statuses into four main groups: diseases, impacts of climate change, habitat loss/degradation, and overexploitation. On the other hand, Chen et al. (2017) found that predicted reductions in amphibian richness, endemism, phylogenetic diversity, phylogenetic endemism, and suitable habitat were lower in protected areas than in nonprotected areas. Similarly, Kaensa et al. (2014) claimed that most anuran amphibian species in the protected area had larger population sizes than those in the nonprotected area. We found that the estimated population size of *P. b. caralitanus* is stable to slightly increasing over the years in Gölcük population. Gölcük Lake is protected as a nature park and encompasses an undisturbed amphibian habitat. It is one of the best-protected areas due to the absence of the many threatened factors for the amphibian population such as minimum human activity, absence of alien species, no over-collection, and no decrease in water level due to drought.

5. Conclusion

In conclusion, long-term studies give us important information about the population structures of threatened species. Especially if there is a decrease in population size, it guides in identifying the reasons for this decrease by considering ecological balances of the ecosystem and in creating appropriate conservation strategies. The previous studies highlighted the value of protected areas as a contributing factor to maintaining effective amphibian population size (Blaustein and Kiesecker, 2002; Von May et al., 2008; Ochoa-Ochoa et al., 2009; Kaensa et al., 2014). This study gives information on the population structure for a well-protected population of *P. b. caralitanus*. The obtained data on population structure from this study showed that the high survival rate and stable to slightly increasing population size of *P. b. caralitanus* in consecutive years in a well-protected nature reserve area. In this context, the study's results offer insights into the annual population size fluctuations of the *P. b. caralitanus* within a well-protected area. It is also informative and a guide for future studies in evaluating population size changes of a target species in protected and nonprotected areas.

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