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MITIGATION MEASURES TO REDUCE ROADKILL OF FIRE SALAMANDER *SALAMANDRA SALAMANDRA* (LINNAEUS, 1758) USING PASSIVE TRAPS IN PLITVICE LAKES NATIONAL PARK

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Amphibians are a significantly and globally threatened group of vertebrates. In the European context, this particularly refers to the fire salamander (*Salamandra salamandra* Linnaeus, 1758), assessed as a vulnerable species due to a decreasing population trend. Threats affecting the population include climate change, habitat fragmentation due to road and infrastructure construction, and the fungal disease *Bsal*. Roadkill of amphibians during migration cycles also affects their abundance. In Plitvice Lakes National Park we monitored the number of fire salamander individuals on the road during spring and autumn migrations in 2023, including animals captured in passive traps along eight sections, and those found road-killed. Seven amphibian and one reptile species were recorded during this research. Selected mitigation measures to prevent fire salamander roadkill with the installation of temporary fences with pitfalls and funnel traps, singled out certain sections as more active, providing insight into the sex ratio of individuals and recording the number of casualties. The fire salamander was the most represented amphibian with 170 individuals captured in passive traps, 36 counted alive on the road, and 50 road-killed. Using individual recognition software *Amphibian and Reptile Wildbook*, 93.81% of individuals were confirmed to be captured only once during the study period. Through ongoing activity, additional data will be gathered on the number of migration routes, including road-killed individuals opposite the temporary fences, identifying new critical points, and documenting roadkills.

Keywords: *Salamandra salamandra*, protected area, roads, migrations, funnel traps

Poljak, K., Vurnek, M., Špoljarić, I., Franjković, D., Čulinović, K. & Rendulić, Ž.: Mjere ublažavanja stradavanja pjegavog daždevnjaka *Salamandra salamandra* (Linnaeus, 1758) na prometnici pomoću pasivnih zamki u Nacionalnom parku Plitvička jezera. Nat. Croat., Vol. 34, No. 1, 85-100, 2025, Zagreb.

Vodozemci su značajno i globalno ugrožena skupina kralješnjaka. Posebno se to u europskom kontekstu odnosi na pjegavog daždevnjaka (*Salamandra salamandra* Linnaeus, 1758) ocijenjenom kao osjetljiva vrsta zbog opadanja trenda populacije. Prijetnje koje utječu na populaciju uključuju klimatske promjene, fragmentaciju staništa izgradnjom prometnica i infrastrukture i gljivičnu bolest *Bsal*. Stradavanje na prometnicama tijekom migracijskih ciklusa vodozemaca utječe na njihovu brojnost. U Nacionalnom parku Plitvička jezera tijekom 2023. godine u proljetnim i jesenskim migracijama pratili

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smo broj jedinki pjegavog daždevnjaka na prometnici, kako uhvaćenih jedinki u pasivnim zamkama na osam dionica, tako i stradalih. Ukupno je tijekom praćenja zabilježeno sedam vrsta vodozemaca i jedna vrsta gmaza. Odabrane mjere ublažavanja stradavanja pjegavog daždevnjaka postavljanjem privremenih ograda s lovnim posudama i tunelima izdvojile su pojedine dionice kao aktivnije, i pružile uvid u udio jedinki prema spolu, kao i u broj stradalih jedinki. Pjegavi daždevnjak bio je najzastupljeniji vodozemac sa 170 jedinki uhvaćenih u pasivnim zamkama, 36 jedinki zatečenih živih na prometnici i 50 stradalih. Upotrebom softvera za individualno prepoznavanje *Amphibian and Reptile Wildbook*, za 93,81% jedinki potvrđeno je da su uhvaćeni samo jednom tijekom razdoblja praćenja. Kroz aktivnost koja je u tijeku predviđamo prikupljanje dodatnih podataka o broju migracijskih puteva, uključujući pojedinačna stradavanja nasuprot privremenih ograda, identificiranje novih kritičnih točaka i bilježenje pojava stradalih na cesti.

Ključne riječi: *Salamandra salamandra*, zaštićeno područje, prometnice, migracije, lovni tuneli

INTRODUCTION

After the second mass extinction in the Late Devonian (~ 364 Mya) when the first amphibians evolved and until the fifth mass extinction at the Cretaceous-Tertiary boundary (~ 65 Mya), amphibians survived global extinctions with changes in climate, temperature, and atmosphere (Wake & Vredenburg, 2008). However, initial global assessments of amphibians demonstrated that amphibians are far more threatened than birds or mammals, with 32.5% of amphibian species assessed as globally threatened (Stuart *et al*., 2004). These numbers are changing even more dramatically, as new data on the status of amphibian species worldwide continues to express further deterioration with 40.7% of amphibians now globally threatened (Luedtke *et al*., 2023). With human activities being directly or indirectly associated with current amphibian extinction (Wake & Vredenburg, 2008), the most commonly documented threats are habitat loss and degradation, including agriculture (affecting 77% of species), timber and plant harvesting (53% species), and infrastructure development (40% species) (Luedtke *et al*., 2023). The complex life cycle of many amphibian species exposes them to both aquatic and terrestrial environmental changes (Blaustein & Bancroft, 2007; Bickford *et al*., 2018). Despite the non-uniform geographical pattern in status deterioration, and the differences in primary drivers such as over-exploitation, climate change effects or habitat loss responsible for this status, an emerging threat in Europe is the fungal disease caused by *Batrachochytrium salamandrivorans* (Luedtke *et al*., 2023). The fire salamander (*Salamandra salamandra*) has been assessed both globally and for the EU27 Member States as vulnerable (A3ce) due to a projected population decline estimated to be greater than 30% over the next 21–45 years. This assessment is based on the potential impact of *B. salamandrivorans* (*Bsal*) on the species, and on ongoing habitat loss and degradation occurring throughout its range (IUCN SSC, 2023). At the EU level, the fire salamander (*Salamandra salamandra*) is listed in Annex III of the Convention on the Conservation of European Wildlife and Natural Habitats (BERN CONVENTION, 1979). In Croatia, there are 13 strictly protected amphibian species (ZZOP, 2024), and within the genus *Salamandra*, only *S. atra* is strictly protected (OG 144/13, 73/16).

Road mortality is particularly high for reptiles and amphibians that cross roads (Holdergger & Di Giulio, 2010) for the sake of food, shelter or reproduction. They are regularly impacted by barriers creating habitat isolation (Iuell *et al*., 2003). A significant proportion of the road ecology literature is focused on evaluating the use and effectiveness of mitigation measures that aim to restore connectivity for wildlife or reduce rates of animal-vehicle collisions (Van Der Ree *et al*., 2011). Massive road mortality of amphibians is recorded during spring and/or autumn migrations. Many species

depend on exact timing for reproduction, and in spring, animals from a wider area migrate towards reproduction habitats. Significant mortality of the reproductive part of a population occurs when there is a relatively large road on their migration route, which can ultimately have a long-term negative impact on the entire population size (Janev Hutinec, 2008). For reducing traffic-related mortality and disturbance, a specific measure is fencing, while for reducing the barrier effect, especially for amphibians, a potential measure can be passages below the infrastructure as a series of underpasses in close proximity. Fences serve two purposes: preventing animals from accessing infrastructure (road) and guiding them to safe crossing points (Iuell *et al.*, 2022).

At Plitvice Lakes National Park, which has a long history of scientific research (over 120 years), scientists have conducted research into the herpetofauna to determine the number of amphibian and reptile species and their locations within the protected area. Recent studies included several new areas, especially high elevation areas (Koren *et al.*, 2023). According to the Management Plan of Plitvice Lakes National Park, there are 14 species of amphibians, six of which are enlisted as strictly protected in the park area (Kovačević, 2019). The Management Plan stipulates a range of activities for the conservation of natural values and prescribes the protection of migration paths (especially for amphibians) during the reproduction cycle (Kovačević, 2019).

The objective of this paper is to present one-year research data concerning dthe main migration routes of the fire salamander (*Salamandra salamandra*) on a section of road in Plitvice Lakes National Park subsequent to the installation of temporary fences with pitfalls and funnel traps. Road sections chosen for monitoring had been determined as critical points in a previous study in 2019/20 (Poljak, 2021), where high numbers of roadkills and live individuals of the fire salamander were recorded.

MATERIALS AND METHODS

Site description

Plitvice Lakes National Park is a part of the Dinaric karst area, situated in mountainous Croatia and bounded by the Lika region, Kordun plateau, and Ogulin-Plaščanska valley, between the massifs of Mala Kapela to the northwest and Lička Plješivica to the southeast, in an elevation range from 450 to 1280 meters (Fig. 1A). Due to its natural values, it was declared a national park in 1949 and inscribed on the UNESCO World Heritage List in 1979. With an area of almost 300 km^2 it is Croatia's largest national park. It is characterised by various habitat types, with forests occupying 81%, water surfaces about 1%, grasslands around 15%, and approximately 3% of the area is significantly altered by human activities (Kovačević, 2019). According to the Regulation of the Ecological Network, Plitvice Lakes National Park is included in the Natura 2000 ecological network under code HR5000020 (Martinić, 2019).

The study area lies along a stretch of a road between station 2 and station 3 that is frequented by panoramic vehicles and other park vehicles, spanning approximately 2 km between coordinates N 44.870479, E 15.601961 and N 44.874229, E 15.612161. This road is carved into the slopes of Mt Kosa and runs parallel with the habitat of the Upper Lakes aquatic ecosystems (Fig. 1B). The lake waters serve as breeding grounds for fire salamanders and their development until metamorphosis. Due to the specific terrain, the distance of study sites or sections from the breeding grounds varied. The average distance of the lakes' area (breeding grounds) and study sites was 128.8 m, with the

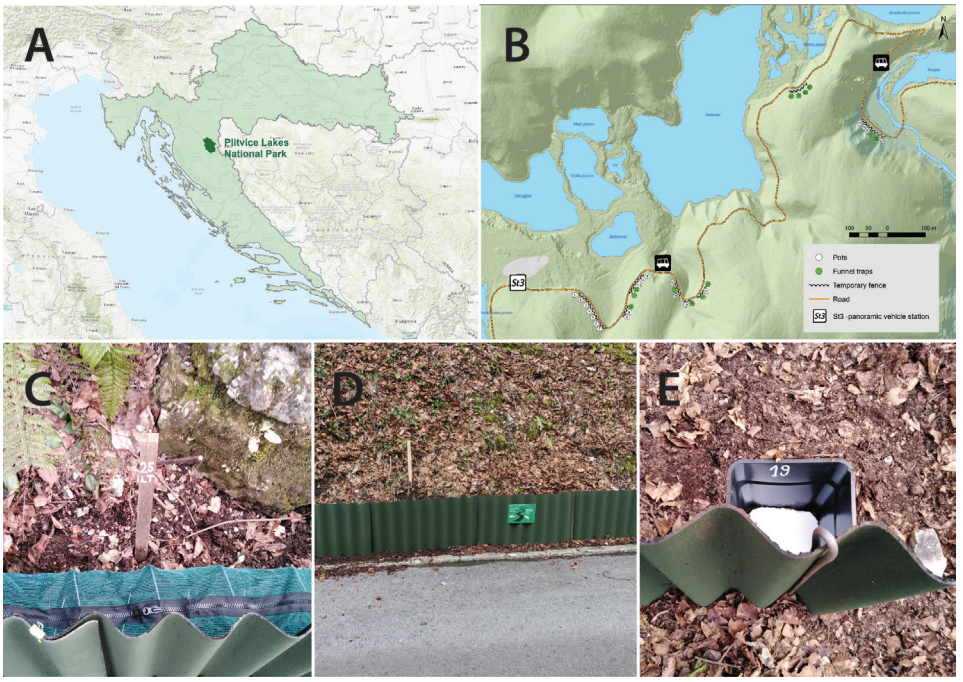

Fig. 1. Geographical position of Plitvice Lakes National Park on the map of Croatia (A); panoramic vehicle road with locations of sections, funnel traps and pitfalls (B); example of positioned funnel trap (C); temporary fence at a section (D); example of positioned pitfall (E).

minimum distance of 40 m (section 7), and maximum distance of 220 m (section 5). After metamorphosis, fire salamanders are considered fully terrestrial and mainly inhabit forested areas near aquatic habitats (Manenti *et al.*, 2017). On the eastern side of the road, there is a strict protection zone with the Natura 2000 forest habitat of Illyrian beech forests (*Aremonio-Fagion*) (Martinić, 2019), where fire salamanders find hibernation sites and daytime resting places. There is no traffic on the road during the night.

Methodology

The chosen approach to reduce fire salamanders roadkill along the road involved installing temporary fences only on one side of the road, equipped with pitfalls and funnel traps (Fig. 1B). Poljak (2021) identified eight critical points on this road, indicated in this study as sections 1–8 (Fig. 1B) varying in length from 20 to 120 meters. Each section was signed with an informative board (Fig. 1D). Along the sections, temporary solid fences 40 centimetres in height with pitfalls were installed (Figs. 1D & 1E). In a case in which sinking pitfalls was not possible, funnel traps were installed (Fig. 1C). The pitfalls and funnel traps were numbered sequentially from 1 to 33. Unlike the funnel traps, which were placed on both sides of the fence, pitfalls were positioned behind the fences with dividers and Styrofoam to prevent amphibians from drowning. This type of position provided researchers with a better estimation of the direction of the animals; those that were trapped on the other side of the fence were taken across the road in the direction of the breeding site (lake), and those that were

trapped in the funnel traps outside the fence were moved to the direction of the forest. After field surveys, passive traps (pitfalls and funnel traps) were disinfected.

Based on the data obtained from Poljak (2021), this field survey was carried out in the seasons of highest activity for fire salamander, during April and September 2023. Weather conditions for activities of *S. salamandra* were established in the previous study. Field surveys were conducted on days with air temperatures ranging from 8 to 14°C with relative humidity levels over 60%, and on days after heavy rain and during light precipitation (rain, drizzle, mist) with wind speeds ranging from 0–2 m/s. It took three days to install the fences and traps both in April and September. When favourable conditions for amphibians started (it was a day after the installation), we began the field survey. In April, the patrols were conducted in two periods, the first lasting 8 days and the second 4 days, the short period between them being marked by weather conditions unfavourable for amphibians. In September, the first survey period lasted for 3 days and the second one 10 days. On days with unfavourable weather conditions, the funnel traps were removed, the pitfalls were closed, and the fences at funnel trap positions were opened. Patrols were conducted at least twice in the morning (8–10 am, 11–12 am), and once at dusk (6–8 pm). The survey in 2023 was carried out over 25 days with 59 patrols, occupying a total of 84 working hours (Tab. 1).

Tab. 1. Monitoring periods in April and September 2023 in days and invested hours.

April			September		
Dates of field survey		Hours	Dates of field survey		Hours
13 – 20	24 – 27	47	15 – 17	19 – 28	37

Each individual encountered within the observed area was documented as a data point. Coordinate points in WGS 84 were collected using GPS. Different data were recorded for live and road-killed *S. salamandra* individuals. Collected live *S. salamandra* individuals were identified and their sex (male/female), adult or juvenile, location (pitfall or funnel trap and section number) were recorded. Each live individual was photographed on the dorsal and ventral sides. Road-killed individuals were identified, and age (if possible) and location were recorded. Live individuals were handled using gloves and relocated either towards or away from the lake, while dead individuals (road-killed) were removed from the road to avoid double counting.

The sex determination of *S. salamandra* was performed based on cloaca features. According to Manenti *et al.* (2017), males develop a swollen cloaca upon sexual maturity, and particularly during mating the cloaca tends to be well developed, large, and convex (bulging), covering the tail, with the cloacal opening (vent) appearing as a deep extended slit. In females, the cloacal lips are typically poorly developed, low, and flattened, with the cloacal opening shifted backward to cover the tail (Zakrzewski, 2007). Individuals shorter than the smallest recognizable male or female were considered juveniles without sex markings, while individuals identifiable by sex were considered adults. General differences between sexes were also taken into account, as females are usually larger than males, especially noticeable during the gestation period, while males have slender bodies, long tails, and long limbs (*Labus et al.*, 2013).

Amphibian and Reptile Wildbook software

The least intrusive method for individual identification of wild animals is not through physical tagging, but rather by utilising their natural individual 'markers' through photographic identification (capture-recapture, CR). Such unique colour patterns are present in many amphibian species (Schlüpmann & Kupfer, 2009). *S. salamandra* and related spotted species (*S. algira, S. corsica, and S. infraimmaculata*) are characterised by a black background covered with varying amounts of common yellow (but in certain taxa orange or red) spots or stripes (Speybroeck & Steenhoudt, 2017). To match the spot pattern of *S. salamandra* individuals, we used the software *Amphibian and Reptile Wildbook* developed by Wild Me (https://www.wildme.org), which is based on a computer algorithm that detects and reliably identifies each individual from photographs. The software allows the uploading of a large number of photographs without prior processing. So far, the software has enabled the recognition of individuals of the fire-bellied toad (*Bombina variegata*) and adult individuals and larvae of *S. salamandra* (Schulte *et al.*, 2022).

Photographs of live adults were imported into the software, regardless of location (passive traps or on the road). A photo database was established, and each fire salamander was assigned a unique ID. The photograph of each captured animal was compared with the photographs of each individual stored in the database. Using pattern recognition software, a comparison of dorsal patterns and consecutive individual identification was conducted. In cases where the software did not provide a matching photograph from the database, a manual comparison was undertaken to ensure accurate identification of the animal. Additional comparisons were made using photographs of the ventral side of individuals. If a suitable photograph was already present in the database, the capture was noted as a recapture. Conversely, if the animal's image was not present in the database, it was incorporated into the photo database and assigned a new individual identification.

RESULTS

A total of 450 herpetofauna individuals were found on the road and in the pitfall/funnel traps during April and September 2023. In addition to seven species of amphibians, one reptile species was also recorded, with one individual found road-killed. Among the Caudata, the most abundant species was the fire salamander (*S. salamandra*) with 256 individuals (56.89%), of which 170 were caught in the pitfall/funnel traps, 36 recorded live on the road, and 50 road-killed. Among the Anurans, the most numerous species was the common toad (*Bufo bufo*) with 18% (Tab. 2).

Field surveys were performed at different times of day. In April 2023, 23 patrols were conducted in the morning, while there were 8 at dusk. In September, there were 26 patrols in the morning and 2 at dusk. During field surveys in 2023 (April and September), the highest numbers of *S. salamandra* individuals were captured during the 1st morning patrol (129), followed by the 2nd morning patrol (15), and the 3rd was the dusk patrol (26). However, apart from the difference in the number of fire salamander individuals caught in passive traps during the spring (118) and autumn (52) migrations, there was also a difference in the number of individuals caught during times of day. In April, there were 80 individuals in the 1st morning patrol, 14 in the 2nd, and 24 at dusk in the 3rd patrol, whereas, in September, there were 49 individuals in the 1st patrol, 1 in the 2nd, and 2 in the 3rd (Fig. 2).

Tab. 2. Species and status of individuals recorded in the pitfalls/funnel traps and on the road.

No.	Species	Pitfall/Funnel trap		Road		Σ	%
		Live	Died	Live	Roadkill		
AMPHIBIANS							
1.	*Salamandra salamandra*	170	0	36	50	256	56.89
2.	*Bufo bufo*	79	0	0	2	81	18.00
3.	*Rana temporaria*	38	0	0	1	39	8.67
4.	*Lissotriton vulgaris*	36	1	0	2	39	8.67
5.	*Rana dalmatina*	21	0	0	3	24	5.33
6.	*Ichthyosaura alpestris*	9	0	0	0	9	2.00
7.	*Rana ridibunda*	1	0	0	0	1	0.22
REPTILES							
8.	*Natrix tesselata*	0	0	0	1	1	0.22
Total		**354**	**1**	**36**	**59**	**450**	**100%**

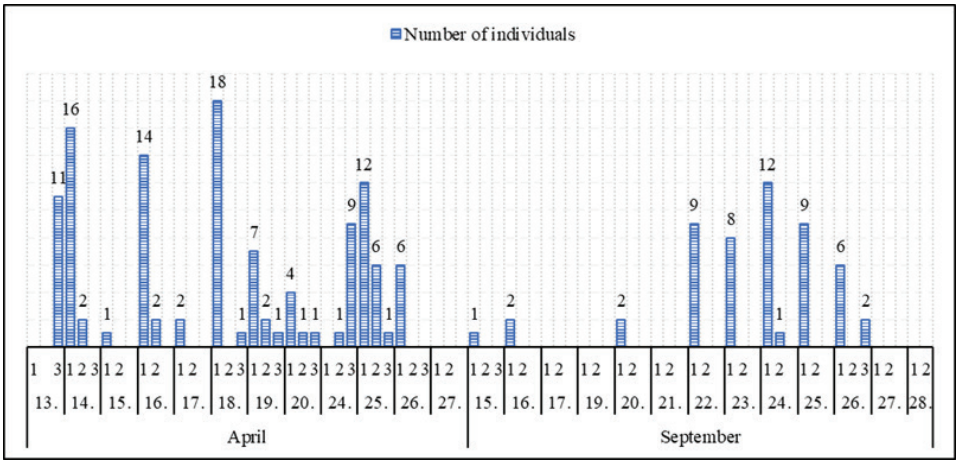

Fig. 2. The number of *S. salamandra* individuals caught in passive traps during monitoring period by dates and daily patrols.

Each section and passive trap exhibited variable activity, indicating places where an increased or decreased number of fire salamander individuals were captured (Fig. 3). Several sections were more active in April (1st, 3rd, 4th, and 7th) than in September (2nd and 6th), while section number 5 was completely inactive (Tab. 3). Additionally, the effectiveness of passive traps revealed differences in migration patterns during the migration seasons. According to the collected data, pitfall number 13 was very active for female fire salamanders in April (30 captured individuals) though only two individuals were captured in September (Fig. 4). For males, this pitfall was also active in April but with fewer individuals (4) and only one captured in September, as opposed to funnel trap number 12 that was more active for male fire salamanders in September with seven captured individuals (Fig. 5).

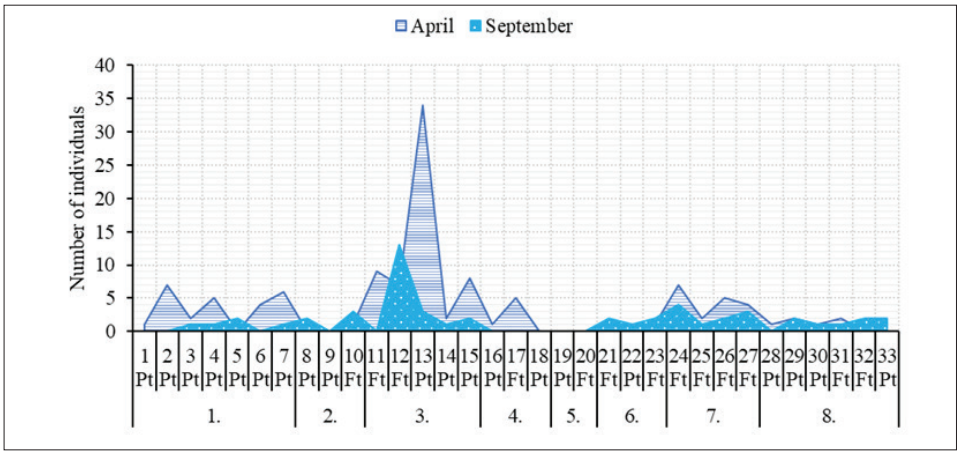

Fig. 3. Comparison of the number of captured *S. salamandra* individuals by section (1-8) and passive traps (1-33) in April and September 2023.

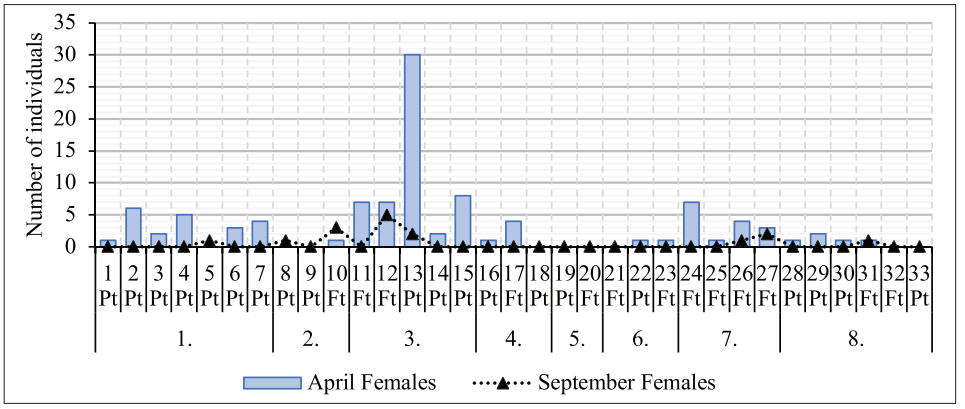

Fig. 4. Number of captured female *S. salamandra* individuals on eight sections and in passive traps in April and September 2023.

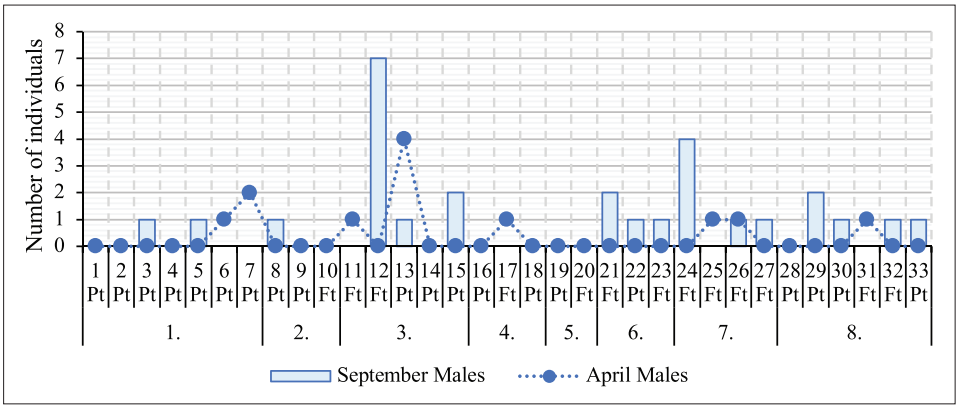

Fig. 5. Number of captured male *S. salamandra* individuals on eight sections and in passive traps in April and September 2023.

Tab. 3. Summary of captured fire salamanders on the road sections during the spring and autumn migrations in 2023.

Sections		1.	2.	3.	4.	5.	6.	7.	8.	∑
Month	April	25	1	60	6	0	2	18	6	118
	September	5	5	19	0	0	5	10	8	52
∑		**30**	**6**	**79**	**6**	**0**	**7**	**28**	**14**	**170**

Considering age, 159 adults and 11 (3 in April and 8 in September) juvenile *S. salamandra* were captured in passive traps. Among adults, 75% (119) were female and 25% (40) male. Differences in the sex ratio were observed during both the spring and autumn migrations. Although the proportion of females captured in April was higher than for males, the sex ratio was reversed during September. Of the 115 adults captured in April, the proportion of females was 88% (101) and males 11% (14), while during September, of the 44 adult individuals, the proportion of females was 36% (16) and males 64% (28) (Fig. 6).

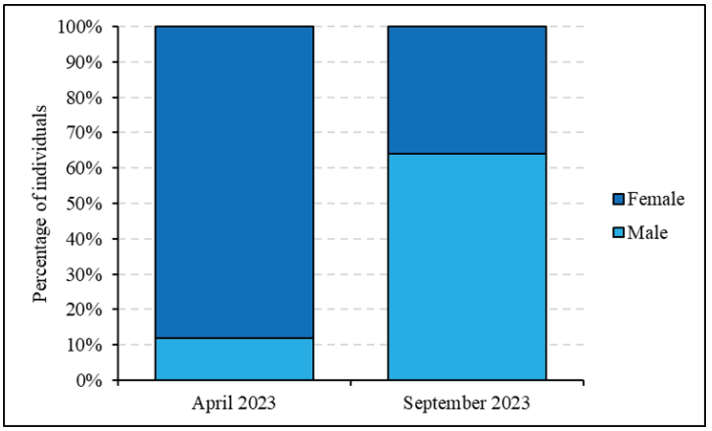

Fig. 6. The proportion of male and female *S. salamandra* individuals captured in passive traps in April and September 2023.

Observing migrations from sections 1 to 8, we monitored the number of live and road-killed individuals, with 86 individuals (both live and dead) recorded. Between sections, where there was no temporary fence and passive traps, we observed an increased number of individuals on the road, both live (26) and roadkills (43). On road areas situated on the opposite side of our study sections, we observed a decreased number of individuals, both live (10) and dead (7) (Tab. 4). In April, we observed increased activity of live individuals on the road (34), as opposed to September (2), while the number of roadkills was the same in both April and September (25 individuals each). No differences in the sex of roadkills was recorded, since individuals could not be properly sexed due to damage.

Tab. 4. Number of fire salamander individuals (live and roadkills) on different road sections and months in 2023.

ROAD	Between sections		Opposite sections	
	Live	Roadkills	Live	Roadkills
Number of individuals	26	43	10	7
Total	69		17	
MONTH	**April**		**September**	
	Live	Roadkills	Live	Roadkills
Number of individuals	34	25	2	25
Total	**59**		**27**	

After evaluating the data of all fire salamander individuals killed along the road, one part of the road showed a higher number of roadkills than all other sections, and therefore can be considered a potentially critical point (Fig. 7), highlighting a potential migration route after section 6 of the road. On this stretch of nearly 90 m of road, we counted 22 killed individuals (18 in April and 4 in September).

We imported 194 dorsal side photos of fire salamander individuals of the total number of 206 individuals (both captured in passive traps and live on a road) in the software *Amphibian and Reptile Wildbook* (Fig. 8). Each individual was assigned an identification number (ID). For better identification of each individual, we used an additional photo of the ventral side. During the study period, 182 individuals (93.81%) were captured once, 11 individuals (5.67%) twice, and only 1 individual (0.51%) three times.

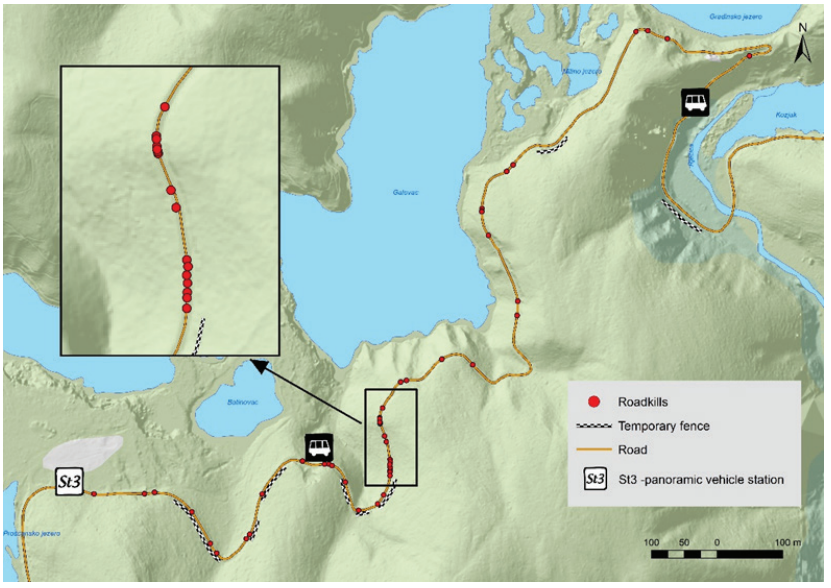

Fig. 7. Part of the road after section 6 with the most observed roadkills of fire salamander individuals.

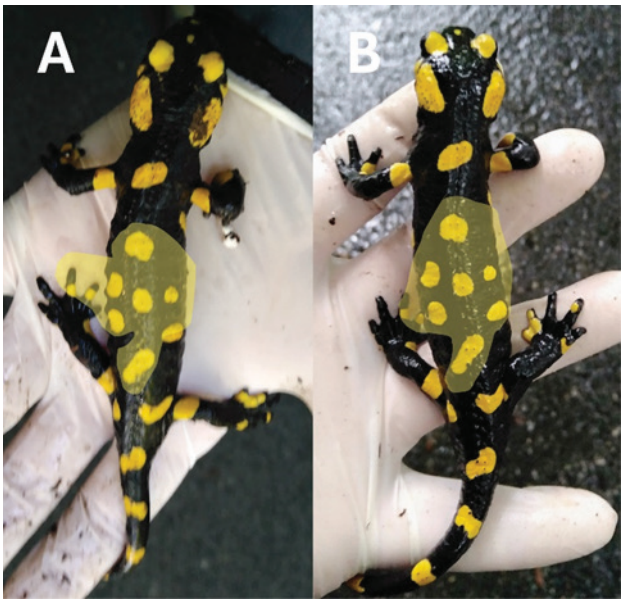

Fig. 8. Fire salamander individual captured for the first time on 20 September (A) and recaptured on 25 September (B). Recognised through photographic comparison of dorsal yellow spots in the *Amphibian and Reptile Wildbook* software.

DISCUSSION

The total number of amphibian individuals recorded in this monitoring confirms that the area of Plitvice Lakes National Park along the road and in the close vicinity of the lakes is a very active amphibian habitats. The increased number of *S. salamandra* individuals on the monitored road highlights the importance of this area for their reproduction and migration cycles. Salamanders are more mobile than usually thought, which could be due to the inappropriate definition of the spatial range of these amphibian populations or may reflect migration between populations (Schmidt *et al.*, 2007). The fire salamander is a common species in the park, recorded throughout its territory with the exception of the far southwest and northeast sections (Koren *et al.*, 2023).

Migration activities of the fire salamander based on the number of captured and live individuals in this study confirmed that the activity level is higher in spring (April) than in autumn (September), in line with their reproduction migrations from terrestrial (wintering sites) to freshwater ecosystems. Additional confirmation of this activity was the high proportion of females (88%) to males (11%) in April, since females return to water bodies only for parturition during which they deposit their offspring in water (Oswald, 2022). According to the collected data, it can be confirmed that the reproduction cycle of fire salamanders starts in April. It is generally accepted that in Europe, salamanders reach sexual maturity at 4 years old (Joly, 1968).

Patrols conducted at different times of day indicated that the activity level of fire salamanders is in the morning from 8–10 am. This was observed in both April and September. During their peak migrations, amphibians move during the day (Puky,

2005), though more diurnal species live at a greater risk of impact of traffic than nocturnal species (Hels & Buchwald, 2001). In the case of Plitvice Lakes National Park, the observed road is not used during the night, which reduces the potential of roadkills.

The most active sections with higher numbers of individuals enabled us to determine the potential migration routes, particularly sections 1 (30 individuals), 3 (79 individuals) and 7 (28 individuals). Section 3 with passive traps numbers 12 and 13 had the most captured individuals, indicating potential migration routes. However, the sections and passive traps active in 2023 should be confirmed in future field surveys. Pond-breeding amphibians are characterised by an annual life cycle with periodic migration among key habitats (breeding grounds, summer habitats, overwintering areas). Adult *S. salamandra* are exclusively terrestrial and inhabit forests (Joly, 1968). It is generally reported that only females migrate to aquatic habitats to lay larvae while males do not return to the water after metamorphosis (Manenti *et al.*, 2017). Furthermore, it is known that animals tend to stay in the vicinity of water bodies, and it is not uncommon to find them within a range of up to 500 meters (Ficetola *et al.*, 2012). Migration movements often have a limited range, but in adult *S. salamandra*, migration distances of up to 500 m from the breeding site are not uncommon (Manenti *et al.*, 2010).

While moving between habitats, amphibians in general and especially fire salamanders are at risk on roads. Mitigation measures to install temporary fences have proven to be efficient, with fewer casualties in sections with installed fences (7 individuals) in comparison with sections without fences (43 individuals). According to Schmidt & Zumbach (2008), temporary fences have been used for a long time, and the method involves setting up temporary fences with pitfalls, typically spaced 15–20 meters apart, which volunteers empty usually in the evening and/or morning, and carry captured amphibians safely to the other side of the road. Well-constructed fences can be highly effective. Todd *et al.* (2007) reported several studies in which temporary fences with pitfalls and funnel traps are found to be a superior way to maximise the number of captured individuals and species. Temporary fences are vertical barriers that intercept amphibian movement and lead them toward installed pitfalls and funnel traps, thus increasing the capture rate. The effectiveness of this method is significantly influenced by weather conditions, season, and amphibian activity (Waqas *et al.*, 2018). However, despite the installed temporary fences, we observed an increased number of roadkills on the part of the road after section 6, and despite the short stretch of approximately 90 m, this indicates a potentially critical part of the migration route of fire salamanders. Future monitoring should examine whether similar pattern will be observed in the next two years.

Finally, the software *Amphibian and Reptile Wildbook* proved useful in monitoring capture-recapture identification. This non-invasive method enabled us to determine the number of individuals captured only once, which was higher than those captured two or three times. In general, photographic identification has become a popular, non-invasive alternative for recognising individuals by the natural variations in their markings (Treilibs *et al.*, 2016). Among amphibians with highly variable colour patterns, the photographic recognition software can be applied to less variable samples of adult individuals and larvae of fire salamanders (Osvald, 2022; Faul *et al.*, 2022). The shapes, number, and size of spots in *S. salamandra* are highly diverse, and no two individuals have the same pattern (Zakrzewski, 2007).

CONCLUSION

The study conducted in Plitvice Lakes National Park sheds light on the significant impact of roadkill on the amphibian population, particularly on fire salamanders, during their migration cycles. Implementation of measures such as temporary fences with pitfalls and funnel traps aimed at mitigating roadkill incidents, proved to be have an effect on amphibian activity levels. Field surveys conducted during peak activity periods in April and September 2023 provided valuable insight into the migration patterns and sex ratio among captured individuals. The analysis of collected data revealed variations in activity levels across sections and highlighted seasonal differences in the number of captured individuals. Determining the spatial locations on roads where amphibians cross in larger numbers indicates the presence of the main migration routes. The study emphasises the importance of continuous monitoring and the application of adaptive conservation measures to protect the vulnerable amphibian population in the region. Through concerted efforts and adaptive management approaches it is important to mitigate the adverse effects of road infrastructure on wildlife and ensure the long-term conservation of the biodiversity of Plitvice Lakes National Park.

AUTHORS' CONTRIBUTION

Katica Poljak and Maja Vurnek wrote the first version of the paper, while other authors contributed to the final version of the manuscript. Katica Poljak, Maja Vurnek, Ivanka Špoljarić, Dragana Franjković and Krešimir Čulinović conducted fieldwork during the study period in 2023. Željko Rendulić developed the maps and contributed to the graphic design of the figures.

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SUMMARY

Mitigation measures to reduce roadkill of fire salamander Salamandra salamandra (Linnaeus, 1758) using passive traps in Plitvice Lakes National Park

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Amphibian species worldwide continue to experience declines, with 40.7% of amphibians now globally threatened. Human activities are directly or indirectly associated with the current amphibian extinction, while the most commonly documented threats are habitat loss and degradation (agriculture, timber and plant harvesting, and infrastructure development). The complex life cycle of many amphibian species exposes them to both aquatic and terrestrial environmental changes. An emerging threat in Europe for the fire salamander (*Salamandra salamandra*) is a fungal disease caused by *Batrachochytrium salamandrivorans (Bsal)*, and according to the last IUCN assessment *S. salamandra* is assessed both globally and for the EU27 Member States as vulnerable due to a projected population decline. At the EU level, fire salamander (*S. salamandra*) is listed in Annex III of the Bern Convention. The presented study was performed in Plitvice Lakes National Park, Croatia's largest protected area and a UNESCO World Heritage site since 1979. Covering almost 300 km^2 of diverse habitats like forests, water surfaces, and grasslands, it is the most famous tourism area in Croatia and the Lika region. The national park is also part of the Natura 2000 ecological network. Strictly protected zones, including the Natura 2000 forest habitat of Illyrian beech forests, provide vital hibernation and resting spots for fire salamanders, which prefer forested areas near water bodies. However, a section between the forested area and lake area about 2 km in length lies between stations 2 and 3 along the road used for tourism vehicles and park vehicles. Notably, there is no night-time traffic on this road, ensuring minimal habitat disruption. The effort to reduce roadkill of fire salamanders along the road involved installing temporary fences (at eight sections) with pitfalls and funnel traps (33 in total) at critical points identified through prior research. Field surveys conducted during peak

activity seasons (April and September) in 2023 focused on days with favourable weather conditions. Patrols were carried out periodically over several consecutive days, totalling 59 patrols (morning and at dusk) over 25 days. Encountered salamanders on road were documented with GPS coordinates. Live individuals were identified by sex, age, location, and photo (dorsal/ventral) while road-killed individuals were documented by age and photo (dorsal) before removal. Sex determination was based on cloacal features, with additional considerations for juvenile identification and general differences between sexes. The software *Amphibian and Reptile Wildbook* was used as a non-intrusive method for identifying *S. salamandra,* using computer algorithms to detect and identify individuals from photographs by recognising distinct and unique colour patterns. Recaptures were noted if a match was found, otherwise, a new individual identification was assigned. In April and September 2023, a total of 450 individuals of amphibians (7 species) and one reptile were recorded on the road and in pitfalls/funnel traps. Among amphibians, the fire salamander was the most numerous species, representing 56.89% of the total count with 256 individuals. Of these, 170 were captured in passive traps (159 adults and 11 juvenile), 36 were found alive and 50 were road-killed. The highest number of *S. salamandra* individuals were captured during the first patrol, followed by the second and third patrols. There was a notable difference in the number of individuals captured during spring and autumn migrations, with regard to passive traps and patrols, with significantly higher numbers captured in April than in September. The passive traps across different sections showed varying activity levels indicated by the number of *S. salamandra* captured. In April, sections 1, 3, 4, and 7 were more active, while sections 2 and 6 were more active in September. Section 5 showed no activity. Migration patterns differed between seasons, with more adults captured in April than in September. In pitfall trap 13, 34 adults were captured in April but only 3 in September. In funnel trap 12, 7 adults were captured in April and 13 in September, along with 1 juvenile. These findings suggest seasonal differences in migration behaviour that is crucial for understanding the movement and identifying priority areas for conservation efforts. Notable differences in sex ratios were observed between spring and autumn migrations. In April, females dominated, with 88%, reflecting their migration towards aquatic habitats for reproduction, while males constituted only 11%. Conversely, September showed a reversal in the sex ratio, with females comprising 36% of captures and males 64%. Juvenile captures were minimal, with three in April and eight in September. These findings highlight seasonal variations in sex ratios during *S. salamandra* migrations. The field survey revealed a total of 86 living and road-killed individuals on the road. Between sections without temporary fences and passive traps, there was a notable increase in both live (26) and road-killed (43) individuals. Conversely, decreased numbers were observed on the side opposite these sections (ten live, seven road-killed). Seasonal differences between live individual activities on the road were noted, while the number of roadkills was the same. A critical point emerged after section 6, with 22 roadkills noted in a 90-meter stretch, higher in April (18) than in September (4). During the monitoring period, 194 dorsal side photos of fire salamander individuals were imported into the *Amphibian and Reptile Wildbook* software, and the data revealed that 93.81% were captured once, 5.67% were captured twice, and only 0.51% were captured three times. The study emphasises the importance of continuous monitoring and the application of adaptive conservation measures to protect the vulnerable amphibian population in the region. Mitigating roadkills, protecting critical habitats, and monitoring population trends are essential steps towards ensuring the long-term survival of these vulnerable amphibians in the study area.