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Wildlife road mortality in a National Park in the Romanian Carpathians (Jiu Gorge National Park)

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Abstract. Between April and October 2018, we studied the wildlife road mortality on two roads from the western region of the Southern Carpathians, Romania. One of these roads follows the course of the Jiu River and is situated in the Jiu Gorge National Park. The other road ascends to the Vulcan Pass. On both roads, wildlife road mortality was studied with monthly frequency on four road sectors, each with a length of 500 meters. We identified 2766 road-killed animals, which belonged to 72 taxa, on the surveyed roads. On the road from the Jiu River Gorge, the number of corpses was double compared to the road from the Vulcan Pass, but the number of taxa was slightly higher. The wildlife road mortality on the two studied roads reflects the composition and peculiarities of the fauna from the neighboring regions, but also the human impact from the past. Thus, the sectors with the lowest road mortality were those affected by other types of human impact besides the road (constructed areas, touristic resources, open areas, etc.). Moreover, the climatic peculiarities of the studied year had a noticeable imprint on the wildlife road mortality. Thus, the cold spring of 2018 made field activity impossible in March (when it was still very cold and snowy). Also, there was a severe cooling in June, as the study day was very cold and windy, which triggered an unusual reduction in road mortality intensity. Local differences in road mortality can be noticed in the case of both studied roads, although the differences between them were not significant. Thus, in the higher areas from Vulcan Pass, we found corpses of animals related to a colder mountain climate. At the same time, in the Jiu River Gorge, animals related directly or indirectly to water were killed (as the road parallels the Jiu River). In the southern areas of the Gorge, we found road-killed animals related to a warmer climate. The differences in wildlife road mortality between the two studied roads could be considered consequences of different road traffic and speeds. Still, they were also caused by differences in habitats, altitude, or fauna composition between the two areas. Because of the high number of road-killed protected animals on roads situated in a national park and in its vicinity, the road network from the protected region should be carefully monitored in the future as the protected area administration should search for appropriate mitigation measures and avoid any construction or modernization of roads.

Keywords: protected area, road-killed animals, temperature, humidity, periods, habitats, conservation.

Introduction

Natural protected areas conserve, or at least in theory they should conserve, the biodiversity of certain regions, their habitats, and protected, endemic, or umbrella species (e.g., Geldmann et al. 2013, Le Saout et al. 2013, Mi et al. 2023). With all that, natural protected areas and their biodiversity are subject to numerous threats (e.g., Schulze et al. 2018, Wade et al. 2020, Dioses-Salinas et al. 2022, Meng et al. 2023). Thus, sometimes protected areas do not succeed in adequately fulfilling their missions, as there are situations when they fail to protect some endemic species (Craigie et al. 2010, Oliveira et al. 2017, Acreman et al. 2020). At the same time, some animal groups, like insects, are insufficiently represented in protected areas (see Chowdhury et al. 2023). Among others, one of the threats to the biodiversity in natural protected areas is represented by the road network, as the impact is primarily noticeable through wildlife road mortality, an effect mentioned in numerous recent studies (Hartmann et al. 2011, Martínez-Freiría & Brito 2012, Healey et al. 2020, Covaciu-Marcov et al. 2022, Bakaloudis et al. 2023, Mulualem et al. 2023). Thus, on roads situated in protected areas, endemic or protected species were killed by road traffic (e.g., Cicort-Lucaciu et al. 2016, Healey et al. 2020, Covaciu-Marcov et al. 2022). In this context, actions aimed at reducing or even eliminating the victims of road traffic must be undertaken in protected areas, besides detailed studies that should verify their effectiveness (Martínez-Freiría & Brito 2012).

A natural protected area in Romania with clues about wildlife road mortality is Jiu Gorge Natural Park – JGNP (Covaciu-Marcov et al. 2009a). Nevertheless, since those few records of some road-killed amphibians and reptiles (Covaciu-Marcov et al. 2009a) in JGNP, there is no other recent information, nor specific studies, on wildlife road mortality, despite the importance of this protected area biodiversity (e.g. Covaciu-Marcov et al. 2009a, Tomescu et al. 2011, Telcean et al. 2017, Cicort-Lucaciu et al. 2020, Sucea et al. 2022). Moreover, the road network's impact on biodiversity is not the only environmental problem in JGNP, as this protected area is under considerable threat, represented by vast hydro-technical works aiming to realize micro-hydropower plants, which will determine a drastic reduction of Jiu River flow (e.g. Carpa et al. 2017, Marinescu & Mititelu-Ionuș 2019, Dejeu et al. 2022). Thus, we aimed to analyze the magnitude of a type of human impact on the biodiversity in JGNP, namely the impact of wildlife road mortality. Such studies are urgently needed in the context of the worldwide devastating impact of road mortality on wildlife (e.g., Ashley & Robinson 1996, Baxter-Gilbert et al. 2015, Covaciu-Marcov et al. 2022, Moore et al. 2023). Even more, in Romania, there is information on the wildlife road mortality from a natural park situated in the southwestern

area of the country, a study which indicated a high road mortality intensity and an increased number of road-killed protected and endemic species (Covaciu-Marcov et al. 2022).

Considering the above mentioned, we hypothesized that also in JGNP, the wildlife road mortality will be intense, and it will affect numerous threatened, protected, or endemic animals, just like in other cases (e.g., Glista & DeVault 2008, Wang et al. 2013, Ciolan et al. 2017, Healey et al. 2020, Covaciu-Marcov et al. 2022). Moreover, because in the immediate vicinity of the park in the previous years, a local road that leads to a tourist resort in Vulcan Pass was modernized (Baciu 2013, Guță 2017, 2022), we hypothesized that between that road and the road in JGNP, there would be differences in wildlife road mortality. Thus, the objectives of our study were the following: **1.** to determine the animals killed on several road sectors from JGNP and its neighboring areas, **2.** to establish the wildlife road mortality differences depending on the period, **3.** to establish the particularities of wildlife road mortality depending on the aspect of the roads neighboring habitats, altitude, and meteorological conditions.

Materials and methods

The region where the wildlife road mortality was studied is in a mountainous area, localized in the Parâng and Vulcan Mountains, massifs in the western part of the Southern Carpathians (Mânduț 2006). The Jiu River is the central element of the region; it is one of the largest rivers in the country, as it is a Danube affluent that originated from the Southern Carpathians (Ujvári 1972). JGNP is situated along the Jiu River and its neighboring massifs. Parallel to the Jiu River is a railway (line 202) and a national road (DN 66).

The field study was realized in 2018, between April and October. Although, in other cases in Romania, road mortality studies begin in March (Ciolan et al. 2017, Covaciu-Marcov et al. 2017) or were even realized over the entire length of a year (Covaciu-Marcov et al. 2022, Pop et al. 2023), in this case, we could begin our field research only in April. This fact resulted from a cold and prolonged winter, combined with the high altitude at which some of the studied road sectors are situated. Thus, the road to Vulcan Pass reaches an altitude of more than 1600 meters (even more than 1200 meters in the case of one of the studied sectors). Thus, snow patches were still present in this area, even in April, in the vicinity and upstream of the last analyzed road sector. Because of those mentioned above, the study involved seven months of field activity from April to October. We made one day of field study each month, as the road mortality research had a monthly frequency, like in other cases (e.g., Ciolan et al. 2017, Covaciu-Marcov et al. 2017, 2022, Pop et al. 2023).

During the study period, we analyzed the wildlife road mortality on two different types of roads in the Jiu River Gorge region. On one side, we studied a national road (DN 66) that passes through the Jiu River Gorge, parallel to the Jiu River. It is an important road with heavy traffic. Even if it is situated in a mountainous region, it has numerous curves, and thus, the speed is reduced to approximately 60 km/hour. The road has been recently rehabilitated and modernized (Guță 2023); therefore, the asphalt is in perfect condition. The road has one lane in each direction. It is parallel to the Jiu River valley; thus, it only has a reduced slope. The road is surrounded almost entirely by forests, mainly represented by beech forests, as JGNP is a densely forested area, covered mainly by beech forests (Theme no.11.RA/2004).

The second studied road leads to Vulcan Pass, starting from Vulcan town. It is situated approximately 10 km west of the previous road and is parallel to it. It is a sloped road that reaches a mountain peak; thus, it has an extremely steep slope in its superior segment near the peak of the pass. In the past, it was one of the main communication routes that crossed the Carpathians, but in the late 1800s, it lost that role in favor of the road situated in the Jiu Gorge, road which is parallel with the Jiu River (Baron & Dobre-Baron 2013). Until recently, it was a dirt road in a very precarious condition. Thus, it was challenging to drive on this road because of that and its steep slopes. In 2014, the road to Vulcan Pass was asphalted until the peak of the pass, which corresponds with the limit between Hunedoara and Gorj counties (Baciu 2013, Guță 2017, 2022). Nevertheless, the road sector in Gorj County remained a dirt road in poor condition. The road sector from Hunedoara County is in good condition due to its recent rehabilitation. Still, because of the steep slopes, it has a lot of curves, and thus, the speed is low (only approximately 30-40 de km/hour). Most of the road to the Vulcan Pass has two lanes (one in each direction), but in its superior part, near the peak of the pass, the road narrows to only one lane. In the upper part of the Vulcan Pass, near the upper limit of the forest, there is a small tourist resort with several villas and a gondola lift (Guță 2017).

In the case of both roads on which we studied the wildlife road mortality, we chose four road sectors, each with a length of 500 m, as in other cases (Covaciu-Marcov et al. 2022). On the road from the Jiu Gorge, the sectors were disposed from downstream to upstream (S I in the vicinity of the southern limit of JGNP, S II downstream of Lainici, S III upstream Pietrele Albe, S IV in Strâmbuța area). In the case of the road from Vulcan Pass, the sectors were also disposed from downstream to upstream (S I in the starting area of the climb, S II on the slope, S III in the touristic area, and S IV at the upper limit of the forest). In Jiu Gorge, the road surrounding habitats were similar, represented by the forest (usually beech forest and alder near the water) and areas with rocky slopes. In the case of Vulcan Pass, the first two road sectors were surrounded by forested areas (vast natural beech forest in S II and recovery forest, even with pine, in S I), relatively open areas with tourist constructions in S III, at the upper forest limit, at its contact with the mountain meadow in the case of S IV.

Each road sector was traveled on foot, as in other studies (e.g., Smith & Dodd Jr. 2003, Covaciu-Marcov et al. 2022, Pop et al. 2023). Each sector was walked each time by three persons who observed all animal corpses that were present on the two roads. Road-killed animals were determined directly on the roads, according to their morphological characters, until the lowest taxonomic level possible, a level comparable with the one reached in other studies (e.g., Popovici & Ile 2018, Covaciu-Marcov et al. 2022, Pop et al. 2023). Thus, vertebrates could be determined at a lower taxonomic level compared with invertebrates (even at a species level), a fact valid also in other cases (e.g., Seibert & Conover 1991, Covaciu-Marcov et al. 2022, Pop et al. 2023). The time required for the study differed between periods but also depending on the traffic intensity, as the sectors from the gorge required more time. Each sector was walked two times. We initially investigated one lane with a length of 500 meters and subsequently examined the other lane at the same distance as in other cases (Covaciu-Marcov et al. 2022).

Besides observing and determining road-killed animals, we also counted the number of cars that passed on the studied road sectors during our field activity. This number eventually was reported at one hour as the average number of cars/hours. The average number of cars/hours was calculated the same way in other similar studies (e.g., Covaciu-Marcov et al. 2022, Popovici et al. 2018, Pop et al. 2023). Also, with a thermometer and a hygrometer, we measured the temperature and humidity of each road sector and period. The results have been processed separately in the case of each studied road, each road sector, and each period. We have calculated the percentage abundance of road-killed animals in the case of each road, each road sector, and each period, and the frequency of occurrence according to sectors and periods. The diversity was calculated using the Shannon index and the similarity with the Jaccard index. The significance of the differences between periods and road sectors was calculated using the Kruskal-Wallis test. In the case of significant differences, we applied the Mann-Whitney test as a post-hoc test. The taxa composition and abundance between the two roads were compared with the t-test. The

correlation between different parameters (number of cars/hour, temperature, diversity, number of taxa, number of victims, etc.) was calculated using the Pearson correlation index. The statistics were made with the help of the Past program (Hammer et al. 2001).

Results

In total, on all sectors of the two roads from the Jiu River Gorge area, and in all study periods, a number of 2766 animals were killed by cars. The road-killed animals belonged to 72 taxa. Among the road-killed taxa, 21 belonged to Vertebrates, and 51 taxa belonged to the invertebrates. The diversity of road traffic victims registered a value of H=3.20. Between the road-killed animals, the highest percentage abundance was registered by Oligochaeta Lumbricidae (17.53%), followed by Diplopoda (10.45%) and Diptera Brachicera (7,92%). Nevertheless, the percentage abundance of Coleoptera from all families registered as road casualties surpasses that of Diplopoda. Among Vertebrates, only one taxon (*Bufo bufo*) registered a percentage abundance higher than 1% (2.06%), as the other taxa's percentage abundance was below 1%. Overall, the lowest percentage abundance of road-killed taxa was 0.04%, registered by six road-killed taxa (Table 1). The highest frequency of occurrence was registered by Oligochaeta Lumbricidae, with a value of 75%. The earthworms were followed with a frequency of occurrence of 71.43% by the Lepidoptera larvae and Diptera Brahicera, and after that, in the third place, by Diplopoda, with a frequency of occurrence of 67.86%.

The differences in wildlife road mortality between the two studied roads were noticeable. Thus, on the road from Jiu River Gorge, more animals (1844 compared with 922) and more taxa (64 compared with 50) were killed compared with the road to Vulcan Pass. Among the 72 road-killed taxa, 41 were killed on both roads, 23 were killed on the road from Jiu River Gorge, and only 8 were killed exclusively on the road from Vulcan Pass. On the road from Jiu River Gorge, the highest percentage abundance was registered by Lumbricidae, followed by Diplopoda and Hymenoptera Vespidae. On the road from Vulcan Pass, Gastropoda without shell, Oligochaeta Lumbricida, and Lepidoptera larvae registered the highest percentage abundance. Thus, between the two roads, there are differences not only regarding the number of victims and taxa but also in the case of their percentage abundance.

Table 1. Percentage abundance (P%) and frequency of occurrence (f%) of the road-killed taxa in the two roads (Jiu Gorge and Vulcan Pass) and for the total.

	Jiu Gorge		Vulcan Pass		Total	
	P%	f%	P%	f%	P%	f%
Oligochaeta Lumbricidae	19.69	67.86	13.23	82.14	17.53	75.00
Gastropoda (with shell)	0.38	25.00	-	-	0.25	12.50
Gastropoda (without shell)	0.33	21.43	15.40	60.71	5.35	41.07
Arachnida Araneida	0.60	28.57	3.04	42.86	1.41	35.71
Arachnida Euscorpius	0.11	7.14	-	-	0.07	3.57
Arachnida Opilionida	0.05	3.57	0.54	7.14	0.22	5.36
Oniscidea	0.16	10.71	0.33	7.14	0.22	8.93
Diplopoda	12.31	78.57	6.72	57.14	10.45	67.86
Chilopoda	1.46	35.71	0.54	17.86	1.16	26.79
Odonata	0.16	10.71	-	-	0.11	5.36
Ephemeroptera	0.27	10.71	-	-	0.18	5.36
Blatodea	0.05	3.57	0.54	3.57	0.22	3.57
Plecoptera	0.11	3.57	0.65	10.71	0.29	7.14
Trichoptera	2.87	46.43	-	-	1.92	23.21
Neuroptera	0.05	3.57	-	-	0.04	1.79
Mantodea	0.05	3.57	-	-	0.04	1.79
Orthoptera	4.07	60.71	6.83	50.00	4.99	55.36
Dermaptera	0.11	7.14	0.54	3.57	0.25	5.36
Coleoptera Dytiscidae	0.05	3.57	-	-	0.04	1.79
Coleoptera Carabidae	2.49	71.43	4.34	57.14	3.11	64.29
Coleoptera Scarabeidae	0.76	21.43	1.63	25.00	1.05	23.21
Coleoptera Geotrupes	0.43	14.29	8.35	50.00	3.07	32.14
Coleptera Melolontha	0.43	14.29	-	-	0.29	7.14
Coleoptera Coccinellidae	0.38	21.43	0.11	3.57	0.29	12.50
Coleoptera Crizomelidae	0.27	14.29	0.33	10.71	0.29	12.50
Coleoptera Tenebrionidae	0.33	10.71	-	-	0.22	5.36

Table 1 – continued next page

Table 1 - continuation

	Jiu Gorge		Vulcan Pass		Total	
	P%	f%	P%	f%	P%	f%
Coleoptera Cantharidae	0.49	17.86	1.41	10.71	0.80	14.29
Coleoptera Stafilinidae	0.27	14.29	0.22	3.57	0.25	8.93
Coleoptera Elateridae	0.38	14.29	0.76	14.29	0.51	14.29
Coleoptera Curculionidea	0.11	7.14	0.43	10.71	0.22	8.93
Coleoptera Cerambicidae	0.16	10.71	0.22	3.57	0.18	7.14
Coleoptera Lampyridae	-	-	0.54	10.71	0.18	5.36
Coleoptera Cetonia	1.25	42.86	0.22	3.57	0.90	23.21
Coleoptera Lucanidae	0.22	14.29	0.22	7.14	0.22	10.71
Coleoptera Silfidae	0.11	7.14	-	-	0.07	3.57
Coleoptera undetermined	0.43	17.86	1.30	25.00	0.72	21.43
Panorpata	0.16	10.71	0.22	7.14	0.18	8.93
Lepidoptera adults	3.09	75.00	1.41	28.57	2.53	51.79
Lepidoptera larvae	2.49	64.29	7.70	78.57	4.23	71.43
Homoptera Cicadina	0.11	7.14	1.63	14.29	0.61	10.71
Heteroptera undetermined	2.11	57.14	2.60	42.86	2.28	50.00
Heteroptera Pyrrhocoris	5.91	57.14	0.11	3.57	3.98	30.36
Hymenoptera Bombus	2.33	53.57	0.11	3.57	1.59	28.57
Hymenoptera Apidae	2.60	57.14	0.11	3.57	1.77	30.36
Hymenoptera Vespidae	11.12	64.29	-	-	7.41	32.14
Hymenoptera Formicidae	0.92	35.71	5.97	21.43	2.60	28.57
Hymenoptera undetermined	0.70	32.14	0.65	10.71	0.69	21.43
Diptera Brachicera	10.47	92.86	2.82	50.00	7.92	71.43
Diptera Brachicera Tabanidae	0.87	17.86	-	-	0.58	8.93
Diptera Nematocera Typulidae	0.54	32.14	0.43	14.29	0.51	23.21
Diptera Nematocera	0.43	14.29	-	-	0.29	7.14
Amphibia *Salamandra salamandra*	0.43	14.29	1.74	28.57	0.87	21.43
Amphibia *Lissotriton vulgaris*	-	-	0.22	7.14	0.07	3.57
Amphibia *Triturus cristatus*	-	-	0.22	7.14	0.07	3.57
Amphibia *Bombina variegata*	-	-	0.22	7.14	0.07	3.57
Amphibia *Bufo bufo*	1.63	57.14	2.93	50.00	2.06	53.57
Amphibia *Rana dalmatina*	0.38	25.00	1.19	25.00	0.65	25.00
Amphibia *Rana temporaria*	-	-	0.43	7.14	0.14	3.57
Reptilia *Lacerta viridis*	0.11	3.57	-	-	0.07	1.79
Reptilia *Podarcis muralis*	0.81	28.57	-	-	0.54	14.29
Reptilia *Zootoca vivipara*	-	-	0.33	7.14	0.11	3.57
Reptilia *Anguis colchica*	-	-	0.22	7.14	0.07	3.57
Reptilia *Natrix natrix*	0.11	7.14	0.11	3.57	0.11	5.36
Reptilia *Natrix tessellata*	0.33	10.71	-	-	0.22	5.36
Reptilia *Coronella austriaca*	0.05	3.57	-	-	0.04	1.79
Reptilia *Zamenis longissimus*	0.16	7.14	-	-	0.11	3.57
Reptilia *Vipera berus*	-	-	0.11	3.57	0.04	1.79
Aves undetermined	0.27	14.29	0.11	3.57	0.22	8.93
Mammalia undetermined	0.05	3.57	-	-	0.04	1.79
Mammalia Talpa	0.11	7.14	-	-	0.07	3.57
Mammalia Rodentia	0.11	3.57	-	-	0.07	1.79
Mammalia Chiroptera	0.16	10.71	-	-	0.11	5.36

The road mortality differences between the study periods were obvious in the case of both roads. Thus, on the road from Jiu River Gorge, the highest number of victims (538) was recorded in September, and the lowest number of victims

(115) was recorded in June (Table 2). The highest number of taxa (39) was road-killed in August, and the lowest number (29) was road-killed in October. A small number of taxa (31) were killed in April and June. The road traffic victim's percentage abundance was very different between the seven study months (Table 2). Thus, Lumbricidae varied in percentage abundance from 1.92% in May to 32.86% in August, but large fluctuations were also evident in the case of other taxa (Table 2). Ten taxa were road-killed in each study month, thus having a frequency of occurrence of 100%, and a number of 14 taxa were killed only in a single month, having a frequency of occurrence of only 14.28%. The highest victim diversity was registered in July (H=3.01%), and the lowest was registered in September (H=2.27). On the road to Vulcan Pass, the highest number of victims (320) and the highest number of taxa (31) were road-killed in May (Table 3). The lowest number of victims (54) was registered in July, and the smallest number of taxa (16) was registered in October. The highest diversity (H=2.69) was registered in April, and the smallest (H=2.29) in October. The differences between the road-killed taxa percentage abundance between periods were also noticeable on this road. Thus, for example, Gastropoda without shell had a percentage abundance of 27.27% in October and 2% in August and was completely absent in April. At the same time, Orthoptera had a percentage abundance of 19.54% in September, only 0,31% in May, and was completely absent in April. Only four taxa were road-killed in each period, having a frequency of occurrence of 100%, and 16 taxa were road-killed in only one month, having a frequency of occurrence of 14.28% (Table 3).

Table 2. Seasonal variation of the percentage abundance and frequency of occurrence (f%) of road-killed taxa in the Jiu Gorge

	Jiu Gorge							
	IV	V	VI	VII	VIII	IX	X	f%
Oligochaeta Lumbricidae	1.65	1.92	3.47	6.50	32.86	30.29	11.11	100
Gastropoda (with shell)	0.82	-	-	0.59	0.40	0.18	0.79	71.42
Gastropoda (without shell)	-	0.64	0.86	-	-	0.55	0.79	57.14
Arachnida Araneida	-	1.28	0.86	-	0.60	-	3.96	57.14
Arachnida Euscorpius	-	-	-	-	0.40	-	-	14.28
Arachnida Opilionida	-	-	0.86	-	-	-	-	14.28
Oniscidea	0.41	-	-	-	-	0.37	-	28.57
Diplopoda	35.26	3.84	1.73	3.55	8.61	11.33	19.04	100
Chilopoda	0.82	1.28	-	4.14	1.80	1.30	-	71.42
Odonata	-	-	1.73	0.59	-	-	-	28.57
Ephemeroptera	-	0.64	2.60	-	0.20	-	-	42.85
Blatodea	-	0.64	-	-	-	-	-	14.28
Plecoptera	-	1.28	-	-	-	-	-	14.28
Trichoptera	-	20.51	1.73	5.32	0.80	-	4.76	71.42
Neuroptera	-	-	0.86	-	-	-	-	14.28
Mantodea	-	-	-	-	-	-	0.79	14.28
Orthoptera	-	-	6.08	8.28	5.41	3.71	5.55	71.42
Dermaptera	-	-	-	-	-	-	1.58	14.28
Coleoptera Dytiscidae	-	-	-	-	-	0.18	-	14.28
Coleoptera Carabidae	5.39	6.41	1.73	5.32	0.80	1.30	0.79	100
Coleoptera Scarabeidae	3.31	0.64	0.86	1.18	-	0.37	-	71.42
Coleoptera Geotrupes	-	2.56	-	0.59	0.40	0.18	-	57.14
Coleptera Melolontha	0.82	3.20	-	0.59	-	-	-	42.85
Coleoptera Coccinelidae	0.41	-	0.86	0.59	0.20	-	2.38	71.42
Coleoptera Crizomelidae	-	-	0.86	1.77	0.20	-	-	42.85
Coleoptera Tenebrionidae	0.41	-	-	-	1.00	-	-	28.57
Coleoptera Cantaridae	0.41	3.84	1.73	-	-	-	-	42.85
Coleoptera Stafilinidae	-	0.64	-	1.18	0.20	0.18	-	57.14
Coleoptera Elateridae	1.24	1.28	-	-	0.40	-	-	42.85
Coleoptera Curculionidea	-	-	-	-	0.40	-	-	14.28
Coleoptera Cerambicidae	0.41	-	0.86	-	0.20	-	-	42.85
Coleoptera Cetonia	-	1.92	3.47	3.55	1.60	0.18	0.79	85.71
Coleoptera Lucanidae	-	-	-	0.59	0.60	-	-	28.57

Table 2 – continued next page

Table 2 - continuation

	Jiu Gorge							
	IV	V	VI	VII	VIII	IX	X	f%
Coleoptera Silfidae	-	-	-	0.59	0.20	-	-	28.57
Coleoptera undetermined	0.41	0.64	1.73	-	0.60	-	0.79	71.42
Panorpata	-	-	-	-	0.40	0.18	-	28.57
Lepidoptera adults	0.82	2.56	8.69	4.73	2.80	2.60	3.96	100
Lepidoptera larvae	1.65	0.64	1.73	1.77	1.80	2.78	9.52	100
Homoptera Cicadina	-	1.28	-	-	-	-	-	14.28
Heteroptera undetermined	1.24	0.64	9.56	-	1.00	2.97	2.38	85.71
Heteroptera Pyrrhocoris	15.76	19.87	1.73	3.55	2.60	3.34	0.79	100
Hymenoptera Bombus	0.41	3.84	8.69	9.46	-	1.85	-	71.42
Hymenoptera Apidae	7.46	0.64	13.91	1.77	1.20	0.55	0.79	100
Hymenoptera Vespidae	0.41	1.92	2.60	8.28	14.22	20.63	1.58	100
Hymenoptera Formicidae	-	3.20	-	2.95	0.40	0.37	2.38	71.42
Hymenoptera undetermined	-	1.28	1.73	0.59	0.80	0.55	0.79	85.71
Diptera Brachicera	9.12	4.48	14.78	14.20	10.62	11.15	7.93	100
Diptera Brachicera Tabanidae	-	-	0.86	0.59	2.80	-	-	42.85
Diptera Nematocera Typulidae	0.41	2.56	0.86	0.59	0.40	0.18	-	85.71
Diptera Nematocera	-	-	-	-	0.80	-	3.17	28.57
Amphibia *Salamandra salamandra*	2.48	0.64	-	-	-	-	0.79	42.85
Amphibia *Bufo bufo*	2.07	1.28	0.86	3.55	0.80	0.55	7.14	100
Amphibia *Rana dalmatina*	0.41	-	-	1.77	-	-	2.38	42.85
Reptilia *Lacerta viridis*	-	-	-	-	0.40	-	-	14.28
Reptilia *Podarcis muralis*	2.48	0.64	-	-	0.60	0.55	1.58	71.42
Reptilia *Natrix natrix*	0.41	-	-	-	-	0.18	-	28.57
Reptilia *Natrix tessellata*	1.24	-	-	0.59	-	0.37	-	42.85
Reptilia *Coronella austriaca*	-	-	-	-	-	0.18	-	14.28
Reptilia *Zamenis longissimus*	0.82	-	-	-	-	-	0.79	28.57
Aves undetermined	0.82	1.28	-	-	-	0.18	-	42.85
Mammalia undetermined	-	-	0.86	-	-	-	-	14.28
Mammalia Talpa	-	-	-	0.59	-	-	0.79	28.57
Mammalia Rodentia	-	-	-	-	-	0.37	-	14.28
Mammalia Chiroptera	-	-	-	-	0.40	0.18	-	28.57

Table 3. Seasonal variation of the percentage abundance and frequency of occurrence (f%) of road-killed taxa in the Vulcan Pass

	Vulcan Pass							
	IV	V	VI	VII	VIII	IX	X	f%
Oligocheta Lumbricidae	15.20	13.12	30.55	16.66	8.00	8.62	9.09	100
Gastropoda (without shell)	-	23.75	9.72	5.55	2.00	18.96	27.27	85.71
Arachnida Araneida	8.00	2.50	-	1.85	2.00	1.72	5.19	85.71
Arachnida Opilionida	-	-	-	-	1.00	2.29	-	28.57
Oniscidea	1.60	-	-	1.85	-	-	-	28.57
Diplopoda	5.60	1.56	2.77	3.70	14.00	13.21	11.68	100
Chilopoda	-	0.31	-	1.85	1.00	1.14	-	57.14
Blatodea	4.00	-	-	-	-	-	-	14.28
Plecoptera	2.40	0.62	-	-	-	0.57	-	42.85
Orthoptera	-	0.31	1.38	1.85	22.00	19.54	5.19	85.71
Dermaptera	-	-	-	-	-	2.87	-	14.28
Coleoptera Carabidae	9.60	4.37	4.16	-	8.00	1.72	-	71.42

Table 3 – continued next page

Table 3 - continuation

	Vulcan Pass							
	IV	V	VI	VII	VIII	IX	X	f%
Coleoptera Scarabeidae	5.60	0.62	4.16	-	-	1.72	-	57.14
Coleoptera Geotrupes	2.40	18.12	8.33	5.55	3.00	2.29	-	85.71
Coleoptera Coccinelidae	0.80	-	-	-	-	-	-	14.28
Coleoptera Crizomelidae	0.80	0.31	-	-	1.00	-	-	42.85
Coleoptera Cantaridae	1.60	3.43	-	-	-	-	-	28.57
Coleoptera Stafilinidae	1.60	-	-	-	-	-	-	14.28
Coleoptera Elateridae	3.20	0.62	1.38	-	-	-	-	42.85
Coleoptera Curculionidea	1.60	0.62	-	-	-	-	-	28.57
Coleoptera Cerambicidae	-	-	--	3.70	-	-	-	14.28
Coleoptera Lampyridae	-	1.56	-	-	-	-	-	14.28
Coleoptera Cetonia	-	0.62	-	-	-	-	-	14.28
Coleoptera Lucanidae	-	-	1.38	1.85	-	-	-	28.57
Coleoptera undetermined	0.80	2.50	-	-	1.00	0.57	1.29	71.42
Panorpata	-	0.62	-	-	-	-	-	14.28
Lepidoptera adults	-	1.25	8.33	3.70	1.00	-	-	57.14
Lepidoptera larvae	1.60	2.81	8.33	5.55	13.00	15.51	14.28	100
Homoptera Cicadina	0.80	4.37	-	-	-	-	-	28.57
Heteroptera undetermined	0.80	2.18	-	1.85	9.00	2.87	1.29	85.71
Heteroptera Pyrrhocoris	-	-	-	-	-	0.57	-	14.28
Hymenoptera Bombus	-	0.31	-	-	-	-	-	14.28
Hymenoptera Apidae	-	0.31	-	-	-	-	-	14.28
Hymenoptera Formicidae	21.60	3.43	-	20.37	2.00	2.29	-	71.42
Hymenoptera undetermined	-	1.87	-	-	-	-	-	14.28
Diptera Brachicera	4.00	4.06	1.38	3.70	1.00	1.72	1.29	100
Diptera Nematocera Typulidae	1.60	0.62	-	-	-	-	-	28.57
Amphibia *Salamandra salamandra*	2.40	1.87	-	-	-	0.57	7.79	57.14
Amphibia *Lissotriton vulgaris*	-	-	-	-	1.00	-	1.29	28.57
Amphibia *Triturus cristatus*	-	-	-	-	-	-	2.59	14.28
Amphibia *Bombina variegata*	-	-	1.38	-	-	-	1.29	28.57
Amphibia *Bufo bufo*	-	0.62	9.72	16.66	7.00	0.57	1.29	85.71
Amphibia *Rana dalmatina*	0.80	0.62	-	1.85	1.00	-	7.79	71.42
Amphibia *Rana temporaria*	-	-	4.16	1.85	-	-	-	28.57
Reptilia *Zootoca vivipara*	1.60	-	-	-	1.00	-	-	28.57
Reptilia *Anguis colchica*	-	-	1.38	-	1.00	-	-	28.57
Reptilia *Natrix natrix*	-	-	-	-	-	-	1.29	14.28
Reptilia *Vipera berus*	-	-	1.38	-	-	-	-	14.28
Aves undetermined	-	-	-	-	-	0.57	-	14.28

The differences in road mortality between sectors were also evident in the case of both roads (Table 4). Thus, on the road from Jiu River Gorge, on Sector IV were killed both the highest number of victims (703) and the highest number of taxa (49). Unlike these, in Sector II there were killed both the smallest number of victims (182) and the smallest number of taxa (37). Nevertheless, the victim diversity was exactly the opposite, having a value of H=3.21 on S II and a value of only H=2.22 on S IV. In Vulcan Pass, the highest number of victims (285) was recorded on S IV and the highest number of taxa (36) was recorded on S I. Unlike these, both the lowest number of victims (153) and the lowest number of taxa (22) was recorded on S III. The highest diversity was registered on S II (H=3.08) and the lowest on S III (H=2.20) (Table 4).

The differences between the percentage abundance of the road-killed taxa on the four road sectors in Jiu River Gorge were great (Table 5). Thus, the percentage abundance of Lumbricida varied between 0.54% on S II and 45.51% on S IV. Unlike these, Hymenoptera Vespida registered a percentage abundance of 31.36% on S I and only 0.42% on S IV. The differences between the road-killed taxa percentage abundance between the road sectors were also evident in the case of the road from Vulcan Pass. Thus, Gastropoda without shell varied in percentage abundance between 3.75% on S II and 30.71% on S III, and Coleoptera Geotrupes ranged between 0.65% on S III and 20% on S IV. On the road from Jiu

River Gorge, a number of 22 taxa had a frequency of occurrence of 100%, as they were identified on all road sectors. On the road from Vulcan Pass, a number of 12 taxa registered a frequency of occurrence of 100% (Table 5).

Table 4. Variation of temperature, humidity, cars/hour, number of individuals, number of taxa, and Shannon diversity on the two studied roads.

			Temperature (°C)	Humidity (%)	Cars/hour	No. of individuals (N)	No. of taxa (S)	Shannon diversity (H)
Total			18.23	53.05	114.90	2766	72	3.20
Jiu Gorge	Total		20.19	49.96	224.07	1844	64	2.97
	Sectors	S1	23.00	40.57	255.08	558	39	2.61
		S2	19.62	49.71	196.66	182	37	3.21
		S3	19.32	48.28	216.41	401	44	2.97
		S4	18.81	49.28	228.14	703	49	2.22
	Months	April	23.00	25.75	177.01	241	31	2.39
		May	17.12	58.75	208.56	156	34	2.87
		June	15.75	37.75	193.16	115	31	2.92
		July	24.75	49.00	219.38	169	32	3.01
		August	21.65	52.25	222.53	499	39	2.51
		September	21.32	46.00	290.95	538	33	2.27
		October	17.75	59.25	256.90	126	29	2.85
Vulcan Pass	Total		15.27	56.14	5.73	922	50	3.01
	Sectors	S1	17.01	55.42	4.89	271	36	2.83
		S2	15.77	56.71	7.85	213	30	3.08
		S3	15.11	55.28	5.64	153	22	2.20
		S4	13.20	57.14	4.51	285	26	2.41
	Months	April	13.75	44.50	5.00	125	25	2.69
		May	14.50	59.25	5.00	320	31	2.63
		June	11.75	46.00	4.75	72	17	2.34
		July	17.70	63.00	7.40	54	18	2.48
		August	23.05	45.50	5.19	100	21	2.48
		September	16.17	62.00	8.24	174	21	2.37
		October	10.00	72.75	4.52	77	16	2.29

The number of cars/hour was much higher in the Jiu River Gorge than in the Vulcan Pass (224.04 compared with 5.73). Even if there were differences regarding the average number of cars/hour, those differences were reduced (Table 4). In the case of the road from Jiu Gorge, the temperature was higher, and the humidity was lower than in the case of the road from Vulcan Pass. In the case of both roads, the temperature dropped from S I to S IV, and the humidity rose in the same direction (Table 4). The differences between temperatures and humidity were noticeable between periods on the road from Jiu River Gorge and the road from Vulcan Pass (Table 4). Generally, the temperature rose in the summer and subsequently dropped. Nevertheless, June represented an anomaly, as it was the coldest study month in Jiu River Gorge (Table 4). In the case of both roads, the humidity registered the highest value in October (Table 4).

There was a moderate positive correlation between the air temperature and the number of cars/hour ($r=0.65$, $p=0.0003$). Also positive was the correlation between the number of cars/hour and the number of taxa they killed ($r=0.45$, $p=0.023$). A negative correlation was registered between air temperature and humidity ($r=-0.53$, $p=0.005$) and between the number of cars/hour and humidity ($r=-0.41$, $p=0.37$). A positive correlation was registered between the number of road-killed taxa and the number of road-killed individuals ($r=0.83$, $p<0.0001$), between diversity and the number of individuals ($r=0.43$, $p=0.028$), and between diversity and the number of taxa ($r=0.054$, $p=0.005$).

According to the t-test, the differences in road mortality between the road from Jiu River Gorge and the road from Vulcan Pass were not significant ($t=1.1591$, $p=0.249$). According to the Kruskal-Wallis Test, the differences between the wildlife road mortality among the road sectors in Jiu River Gorge were not significant ($p=0.23$). Also not significant were the road mortality differences between the road sectors from Vulcan Pass ($p=0.066$) and the differences between periods on Jiu River Gorge ($p=0.379$). In this case, the only significant differences were between periods in Vulcan Pass ($p=0.029$). In Vulcan Pass, analyzing the wildlife road mortality differences between periods two by two with the Mann-Whitney Test, the mortality in May differed significantly from June ($p=0.005$), July ($p=0.006$), and October ($p=0.003$).

Table 5. Variation of the percentage abundance and frequency of occurrence (f%) of road-killed taxa in the Jiu Gorge and Vulcan Pass, according to the road sectors (S1 – S4) (N – number of individuals, S – number of taxa, H – taxa diversity).

	Jiu Gorge					Vulcan Pass				
	S1	S2	S3	S4	f%	S1	S2	S3	S4	f%
Oligochaeta Lumbricidae	2.50	0.54	6.98	45.51	100	8.11	9.85	22.87	15.43	100
Gastropoda (with shell)	-	1.64	0.24	0.42	75	-	-	-	-	-
Gastropoda (without shell)	0.53	1.09	-	0.14	75	11.80	3.75	30.71	19.29	100
Arachnida Araneida	0.35	-	0.99	0.71	75	6.27	2.81	-	1.75	75
Arachnida Euscorpius	0.17	-	0.24	-	50	-	-	-	-	-
Arachnida Opilionida	-	-	-	0.14	25	-	-	-	1.75	25
Oniscidea	0.35	-	0.24	-	50	1.10	-	-	-	25
Diplopoda	4.12	7.14	13.71	19.34	100	14.39	6.57	1.30	2.45	100
Chilopoda	0.53	1.64	3.49	0.99	100	0.36	1.40	-	0.35	75
Odonata	-	0.54	-	0.28	50	-	-	-	-	-
Ephemeroptera	-	0.54	0.24	0.42	75	-	-	-	-	-
Blatodea	-	-	-	0.14	25	1.84	-	-	-	25
Plecoptera	0.35	-	-	-	25	-	2.34	-	0.35	50
Trichoptera	4.48	1.64	2.24	2.27	100	-	-	-	-	-
Neuroptera	-	-	0.24	-	25	-	-	-	-	-
Mantodea	0.17	-	-	-	25	-	-	-	-	-
Orthoptera	5.19	11.53	3.99	1.28	100	2.21	8.45	9.15	8.77	100
Dermaptera	0.17	-	-	0.14	50	1.84	-	-	-	25
Coleoptera Dytiscidae	0.17	-	-	-	25	-	-	-	-	-
Coleoptera Carabidae	2.50	6.04	1.99	1.84	100	5.16	5.16	5.22	2.45	100
Coleoptera Scarabeidae	0.89	3.29	0.24	0.28	100	1.47	1.40	3.26	1.05	100
Coleoptera Geotrupes	-	-	0.49	0.85	50	2.58	5.63	0.65	20.00	100
Coleptera Melolontha	0.35	0.54	0.24	0.56	100	-	-	-	-	-
Coleoptera Coccinelidae	0.71	0.54	0.24	0.14	100	0.36	-	-	-	25
Coleoptera Crizomelidae	-	1.09	-	0.42	50	0.36	0.93	-	-	50
Coleoptera Tenebrionidae	0.71	-	-	0.28	50	-	-	-	-	-
Coleoptera Cantaridae	-	3.29	0.24	0.28	75	0.73	4.69	0.65	-	75
Coleoptera Stafilinidae	0.17	-	-	0.56	50	0.73	-	-	-	25
Coleoptera Elateridae	0.35	-	0.49	0.42	75	1.10	1.40	0.65	-	75
Coleoptera Curculionidea	0.17	-	-	0.14	50	1.10	-	0.65	-	50
Coleoptera Cerambicidae	-	-	0.49	0.14	50	0.73	-	-	-	25
Coleoptera Lampyridae	-	-	-	-	-	-	1.40	0.65	0.35	75
Coleoptera Cetonia	0.89	2.19	1.99	0.85	100	-	0.93	-	-	25
Coleoptera Lucanidae	0.17	0.54	0.24	0.14	100	-	0.46	0.65	-	50
Coleoptera Silfidae	-	-	0.49	-	25	-	-	-	-	-
Coleoptera undetermined	-	1.09	0.49	0.56	75	1.47	3.28	-	0.35	75
Panorpata	-	-	0.24	0.28	50	0.36	0.46	-	-	50
Lepidoptera adults	3.40	3.29	4.23	2.13	100	0.36	3.75	1.96	0.35	100
Lepidoptera larvae	2.15	9.34	2.99	0.71	100	1.47	11.26	8.49	10.52	100
Homoptera Cicadina	-	-	0.24	0.14	50	3.32	2.34	0.65	-	75
Heteroptera undetermined	0.71	2.74	5.23	1.28	100	1.84	6.10	1.96	1.05	100
Heteroptera Pyrrhocoris	6.09	2.19	14.46	1.84	100	0.36	-	-	-	25
Hymenoptera Bombus	1.25	4.39	4.23	1.56	100	-	-	-	0.35	25
Hymenoptera Apidae	5.37	4.94	1.49	0.42	100	0.36	-	-	-	25
Hymenoptera Vespidae	31.36	2.74	5.48	0.42	100	-	-	-	-	-
Hymenoptera Formicidae	1.61	1.64	0.24	0.56	100	19.92	-	0.65	-	50
Hymenoptera undetermined	1.07	-	0.99	0.42	75	0.36	1.40	1.30	-	75
Diptera Brachicera	15.77	7.69	12.96	5.54	100	2.58	3.75	6.53	0.35	100

Table 5 – continued next page

Table 5 - continuation

	Jiu Gorge					Vulcan Pass				
	S1	S2	S3	S4	f%	S1	S2	S3	S4	f%
Diptera Brachicera Tabanidae	2.50	0.54	0.24	-	75	-	-	-	-	-
Diptera Nematocera Typulidae	0.17	1.64	0.74	0.42	100	0.73	0.93	-	-	50
Diptera Nematocera	-	-	0.99	0.56	50	-	-	-	-	-
Amphibia *Salamandra salamandra*	-	3.29	0.24	0.14	75	-	2.34	0.65	3.50	75
Amphibia *Lissotriton vulgaris*	-	-	-	-	-	0.73	-	-	-	25
Amphibia *Triturus cristatus*	-	-	-	-	-	0.36	0.46	-	-	50
Amphibia *Bombina variegata*	-	-	-	-	-	-	-	-	0.70	25
Amphibia *Bufo bufo*	-	3.84	2.49	1.84	75	2.21	2.34	0.65	5.26	100
Amphibia *Rana dalmatina*	0.35	0.54	0.49	0.28	100	0.73	3.75	0.65	-	75
Amphibia *Rana temporaria*	-	-	-	-	-	-	-	-	1.40	25
Reptilia *Lacerta viridis*	0.35	-	-	-	25	-	-	-	-	-
Reptilia *Podarcis muralis*	0.89	-	0.49	1.13	75	-	-	-	-	-
Reptilia *Zootoca vivipara*	-	-	-	-	-	-	-	-	1.05	25
Reptilia *Anguis colchica*	-	-	-	-	-	-	0.46	-	0.35	50
Reptilia *Natrix natrix*	-	1.09	-	-	25	0.36	-	-	-	25
Reptilia *Natrix tessellata*	-	3.29	-	-	25	-	-	-	-	-
Reptilia *Coronella austriaca*	-	-	-	0.14	25	-	-	-	-	-
Reptilia *Zamenis longissimus*	-	0.54	0.49	-	50	-	-	-	-	-
Reptilia *Vipera berus*	-	-	-	-	-	-	-	-	0.35	25
Aves undetermined	0.53	0.54	0.24	-	75	-	-	-	0.35	25
Mammalia undetermined	-	0.54	-	-	25	-	-	-	-	-
Mammalia Talpa	-	-	-	0.28	25	-	-	-	-	-
Mammalia Rodentia	-	-	-	0.28	25	-	-	-	-	-
N	558	182	401	703		271	213	153	285	
S	39	37	44	49		36	30	22	26	
H	2.61	3.21	2.97	2.22		2.83	3.08	2.20	2.41	

According to the Jaccard similarity test, the similarity between the wildlife road mortality on the two studied roads registered a value of 0.569. In the case of sectors on the road from Jiu River Gorge, the highest similarity was recorded between S III and S IV (0.66), and the lowest similarity was registered between S I and S II. In the case of the road from Vulcan Pass, the most similar wildlife road mortality was between S I and S II (0.571), and the lowest similarity was between S I and S IV (0.319). On the road from Jiu River Gorge, the highest similarity was between July and August (0.543), and the lowest was between June and September (0.391). In Vulcan Pass, the highest similarity was between July and August (0.56), and the lowest was between April and June (0.235).

Discussion

Although the length of the studied sectors from the two studied roads was the same (2 km on each) the wildlife road mortality on these differed. Thus, on the road from the Jiu River Gorge, the number of road-killed animals was exactly double compared to the road from the Vulcan Pass, even if the number of road-killed taxa was only slightly higher. The most straightforward explanation for this difference is the traffic intensity, which was highly different between the two roads. Thus, the number of passing cars was much higher on the road from the Jiu Gorge, and the road traffic increase knowingly led to a fauna-mortality rise (e.g., Kobylarz 2001, Jones et al. 2014, Blais et al. 2023, Takahashi et al. 2023). It is known that on recently modernized roads, because of the increase in the number and speed of cars, the fauna road mortality has become more intense (Jones et al. 2014), and on the Jiu Gorge, not only the intensity of the traffic is higher, but also the speed of the cars. However, although simple, this explanation does not stand up entirely, as on some roads with much more reduced traffic than in the JGNP, the number of victims was higher relative to the same length of the road (Covaciu-Marcov et al. 2022), but in other cases, the number of corpses was lower (Ciolan et al. 2017). Thus, even the biodiversity of the surrounding areas could be responsible for these differences, as roads that cross high biodiversity zones cause an increased number of victims (Covaciu-Marcov et al. 2022). This fact means that despite the geographical proximity of the two roads, the ecological conditions at these sites differ to a certain extent, and this fact, combined with the traffic intensity, has determined the differences in fauna road mortality. At the same time, there is evidence of the fact that on roads with heavy traffic, road mortality decreases over time due to the decrease of animal populations from the

surrounding areas caused by the road (e.g., Fahring et al. 1995, Teixeira et al. 2017), and the road from the Jiu Gorge is a national and European road with heavy traffic. The fact that numerous animals continue to die on this road confirms the presence of a rich fauna in the JGNP (e.g., Covaciu-Marcov et al. 2009a, Tomescu et al. 2015, Sucea 2019, Cicort-Lucaciu et al. 2020).

The close number of taxa killed on the two roads indicates, however, the fact that in the nearby regions, the taxa diversity is alike, even if the number of individuals differs locally. Nevertheless, there were also killed taxa on the Vulcan Pass, which were not present on the road from the Jiu Gorge. These were six vertebrate taxa, mainly species linked to the mountain and cold climates, like *Zootoca vivipara* and *Vipera berus* (Fuhn & Vancea 1961), taxa that are missing from the lower area in the Jiu Gorge (Covaciu-Marcov et al. 2009a). These taxa are extremely rare in the JGNP, and each is present only in the highest areas of the park (Covaciu-Marcov et al. 2009a), and as such, they are absent on the road`s neighboring regions from the gorge. *Zootoca vivipara* is a rarity in the studies on road mortality in Romania; previously, it was mentioned only once, in a plain area from the western part of the country (Covaciu-Marcov et al. 2017), where it is considered a glacial relict (Covaciu-Marcov et al. 2009b). At higher altitudes, in the Vulcan Pass, the species ended up exposed to road mortality as the road, which was recently paved (Baciu 2013, Guţă 2017, 2022), was crossing its habitats. This fact shows again the negative effect of road construction and modernization on the fauna in an area that was not previously exposed to anthropogenic impact (Ciolan et al. 2017). At the same time, on the road from the Vulcan Pass, two newt species (*Lissotriton vulgaris* and *Triturus cristatus*) were killed, species that were absent on the road from the Jiu Gorge, which is a region where newts are generally rare due to the rarity of aquatic habitats (Covaciu-Marcov et al. 2009a). The presence of newts as victims on the road from the Vulcan Pass is probably due to the existence of some stationary aquatic habitats near the lower sectors of the road. Thus, these data indicate that newts are more common in areas neighboring JGNP, just like in other cases when road mortality studies were useful for documenting the presence of these animals (Covaciu-Marcov et al. 2022), moreover, as they are conservatively important (O.U.G. 57/2007).

The influence of an important watercourse (the Jiu River) near the road in the Jiu Gorge is evident. This is shown by the presence of victims belonging to some taxa with aquatic larvae, like Ephemeroptera or Trichoptera, or even a snake linked to water like *Natrix tessellata*. Ephemeroptera were killed in high numbers, even on a railroad along a watercourse (Pop et al. 2020), emphasizing these insects' vulnerability to road and rail traffic. In its turn, *N. tessellata* was frequently killed by cars on other roads that border rivers or other important aquatic basins (e.g., Ioannidis & Mebert 2011, Kambourova-Ivanova et al. 2012, Ciolan et al. 2017, Covaciu-Marcov et al. 2022). At the same time, on the road from the Jiu Gorge, there are also animals linked to a warmer climate and absent from the Vulcan Pass, like *Euscorpius carpathicus* or some lizard species. Of these, *E. carpathicus* is considered a Mediterranean and thermophilic element in the country, where it is endemic and present only in the Southern Carpathians (Bunescu 1959, Gherghel et al. 2016). It was also previously mentioned as a victim of road traffic in another zone of its distribution range (Covaciu-Marcov et al. 2022), indicating the species' vulnerability when exposed to this anthropogenic threat. Also, it needs conservative strategies in protected areas, even if the species is not protected by law (O.U.G. 57/2007). Thus, the impact of road traffic on this species is impossible to evaluate now, especially when detailed data referring to the species distribution are known from only one region (Covaciu-Marcov & Ferenţi 2019). Consequently, the protected areas where this species is present, including JGNP, must undertake actions to inventory the species distribution.

The influence of the habitats neighboring the road on road mortality has been a known fact for a long time, being reported in numerous studies (e.g., Smith & Dodd Jr. 2003, Covaciu-Marcov et al. 2017, 2022, Anđelković & Bogdanovic 2022, Gutema et al. 2023, Zevgolis et al. 2023). In the case of the road from the Jiu Gorge, the habitats surrounding the road are similar to each other (forested areas, rocky slopes); therefore, the fact that the differences between the road mortality on these were not significant was expected. However, even if statistically not significant, differences in road mortality between the road sectors from the JGNP can be easily observed. Thus, the lowest intensity of road mortality was observed on SII, an anthropogenically affected area near a religious and touristic objective (the Lainici Monastery). Low road mortality can also be because, in this area, the road is bordered by some secondary open areas, while high road mortality characterizes forested areas (e.g., Popovici et al. 2018, Magiolli et al. 2019). At the same time, at the moment of the study in the area, there was a temporary bridge with a single lane, limiting traffic and speed. All these elements have probably contributed to a decrease in road mortality on SII. At the same time, there are differences in taxa composition between the sectors because the number of southern elements decreases upstream in the gorge (Covaciu-Marcov et al. 2009a). Thus, in the Jiu Gorge, *Salamandra salamandra* and *Bufo bufo* were absent only on SI, as well as Ephemeroptera, but *Lacerta viridis* was present only on SI. The differences between the sectors result from increased humidity upstream in the gorge. Thus, Lumbricidae has the highest abundance in the gorge's most humid, high, and rocky zones on SIV. Differences between the sectors are also observable in the Vulcan Pass, where SIII, a sector that crosses an open area with touristic villas, presents the lowest road mortality. SIV, situated at the highest altitude, had victims of some mountain species, like *Zootoca vivipara* and *Vipera berus*.

Concerning seasons, the low number of victims in June is a consequence of the fact that the study overlapped with some unusually cold days, with low temperatures and very strong winds. This affected the fauna directly (animals that prefer warm weather were not active) and indirectly (the corpses of butterflies or flies were blown from the road by the wind). This fact modified the classic periodicity of road mortality peaks, in contrast to other cases, when the road mortality peak was registered in the spring (e.g., Ciolan et al. 2017, Covaciu-Marcov et al. 2017, Zevgolis et al. 2023). In our case, the cold weather in the study days of June has determined the atypical decrease in road mortality intensity. However, this decrease does not necessarily have to be considered a characteristic of the entire month but of the study day and the period around

this day. Anyway, this fact shows the effect of temporary climate oscillations on road mortality, a fact registered in other cases, too (e.g., Covaciu-Marcov et al. 2017, Anđelković & Bogdanovic 2022, Arca-Rubio et al. 2023). Therefore, if climate warming can influence road mortality (Covaciu-Marcov et al. 2022), a short cooling period can result in a secondary decrease, which masks the actual intensity of the phenomenon. Otherwise, the taxa killed by cars show foreseeable seasonal oscillations, the period of the year influencing the corpse composition depending on the group (e.g., Covaciu-Marcov et al. 2017, 2022, Anđelković & Bogdanovic 2022, Arca-Rubio et al. 2023). Thus, *Salamandra salamandra* was killed by cars in spring and autumn, missing during the summer, as in other cases (e.g., Covaciu-Marcov et al. 2022). The differences between the two roads are evident in the case of this species, too; thus, in the Vulcan Pass, *S. salamandra* appears in the autumn, a month earlier than in the gorge, due to climatic differences between these. On both roads, Orthopterans were absent in the spring, just like in other cases (Ciolan et al. 2017). The significant differences between the periods in the Vulcan Pass are a consequence of the greater habitat heterogeneity near the road and the higher altitudes, where seasonal oscillations are easier to notice.

There were more victims of some flightless taxa than some other flying taxa. Even from Lepidoptera in the Vulcan Pass were identified more larvae than adults, even if, in other cases, numerous adults were killed (McKenna et al. 2001, Rao & Girish 2007, Skórka et al. 2013). This fact can be explained by the low mobility of Lepidoptera larvae, which, in other cases, were more numerous than adults (Pop et al. 2023). At the same time, their corpses, once killed, remain stuck on the road, while adult corpses of butterflies are easily and rapidly removed from the road in several ways, like passing cars or scavengers (McKenna et al. 2001, Skórka 2016). Birds were rarely killed on this road, compared to reptiles, for example, even if, in other cases, they were killed in high numbers (e.g., Sacramento et al. 2022, Medrano-Vizcaíno et al. 2023, Pop et al. 2023). Among flying animals, on the road from the gorge, we identified the corpses of two bats, a fact rarely mentioned in Romania (Covaciu-Marcov et al. 2022, Pop et al. 2023) but frequently reported in other regions (e.g. Lesiński 2007, Damásio et al. 2021, de Figueiredo Ramalho 2021). Most road traffic victims were terrestrial animals, but occasionally, we identified a Coleoptera Dytistcida, an animal connected to the water (e.g., Radu & Radu 1967). This was determined on SI in the Jiu Gorge, and it probably arrived accidentally on the road from the water nearby or any other aquatic habitat in the JGNP. In the same way, aquatic or semiaquatic animals were killed occasionally in other cases, too (e.g., Popovici & Ile 2018, Balčiuskas et al. 2022).

The great similarity and the absence of differences between the sectors and periods are a consequence of the geographic and biological uniformity of the region. At least in the Jiu Gorge, the altitudinal difference between the sectors is low, and the aspect of the neighboring habitats is similar; thus, the mortality will also be similar. Differences were registered in the southern part of the gorge, where the submediterranean southern influences are felt just like in other cases (Covaciu-Marcov et al. 2009a). Mountain elements appear at the upper part of the gorge and the Vulcan Pass, like in other cases (Covaciu-Marcov et al. 2009a, Telcean et al. 2017, Sucea et al. 2023). The road mortality of the fauna reveals the composition, stratification, and distribution of the fauna in the studied area and, to some extent, the anthropogenic effect, which reduces road mortality in sectors where it is more pronounced, impacting the fauna in several other ways.

Even if the results of the study on the fauna road mortality in the Jiu Gorge mostly confirm the known information on faunistic studies on the zoogeographic particularities in the region (Covaciu-Marcov et al. 2009a), at the same time, these indicate the extremely high impact of roads on region`s fauna. The negative effects of this impact are even more pronounced as the road from the Jiu Gorge is situated in a national park, and the road from the Vulcan Pass is located mostly in its immediate vicinity. Moreover, on these roads, some strictly protected animals were killed, like *Triturus cristatus* or *Bombina variegata* (O.U.G. 57 / 2007), but also some endemic ones, like *Euscorpius carpathicus* (Bunescu 1959, Gherghel et al. 2016). If, in the case of a national and European road from the gorge, too few mitigation measures can be taken, the road from the Vulcan Pass, which is a local road with touristic importance, can easily be targeted by mitigation measures. First, the asphalting of this road should not be continued in the Gorj County sector. Even if the sector in the vicinity of the JGNP is short, the impact of the modernized road will undoubtedly be huge. A completely asphalted road will certainly have heavier traffic, and more traffic will make more victims (e.g., Kobylarz 2001, Blais et al. 2013, Jones et al. 2014, Takahashi et al. 2023), a fact observed even in the gorge. However, considering that the road will be realized anyway, this should have barriers and under-crossings, which proved to be efficient in reducing road mortality (e.g., Dodd Jr et al. 2004, Rytwinski et al. 2016), even if some recent studies indicate the accumulation of pollutants in these coming from the road traffic (White et al. 2023). Another way of reducing road mortality would be speed limitations, although due to the curves and the slope, it is hard to believe that speed could be even more reduced. In the gorge, road-mortality hotspots don`t seem to exist; thus, applying particular mitigation measures would be difficult. Also, the traffic speed is relatively reduced in the gorge because 50 km/h speed limit indicators are installed on most of it. Nevertheless, if on the existing roads, too little can be done because of economic reasons, the park administration should ensure that new roads will not be asphalted in the park or its vicinity, the harmful effect of road asphalting in forested areas being well-known (e.g. Ciolan et al. 2017, Popovici et al. 2018, Magiolli et al. 2019), and also, because JGNP is a heavily forested area (Theme no.11.RA/2004). If some major interests will make the realization or modernization of new roads necessary in JGNP, detailed studies must be realized previously.

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