



Small Rodents Are Appropriate Bioindicators for Total B-Activity Monitoring in Alpine Ecosystems

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Abstract: The priority of studying high mountain ecosystems is due to the increasing influence of a large number of natural and anthropogenic pollutants. As solar radiation levels increase, exposure to human-caused radionuclides in alpine regions decreases due to the decay of anthropogenic radioisotopes, as well as precipitation and demobilisation of heavier elements. The present article is based on biomonitoring studies of the total β -activity of small rodents conducted in the Rila Mountains. The purpose of the study is to present data on the biomonitoring potential of small rodents in relation to radioecological studies in these areas. The suitability of two rodent taxa is discussed in a comparative aspect: the omnivorous *Apodemus* sp. and the herbivorous representative *Chionomys nivalis*. The larger reduction was observed in *Apodemus* sp., followed by the snow vole, which shows high zoomonitor potential for alpine ecosystems. An analysis of the animal diet and determination of total β -activity in the dominant plant components of the food spectrum was also performed. This allows further detailed studies to track the biomagnification of radionuclides along food chains in these fragile ecosystems.

Key Words: biomonitoring, small mammals, total β -activity, alpine ecosystems

Introduction

Anthropogenic pollution is a significant issue, especially in diverse landscapes like mountain ecosystems. Recent studies clearly show that alpine environments are negatively influenced by environmental contamination, especially from long-distance transmission (BARREDO et al. 2020). The Alpine ecosystem, as the highest elevation belt of the mountains, is an important reservoir of fresh water and provides a habitat to unique species (BARREDO et al. 2020). It is projected that, soon, due to the effects of climate change, these habitats will decrease in size and become vulnerable to degradation due to various anthropogenic factors

(BARREDO et al. 2020). This will entail serious consequences for species diversity in the high mountains and ecosystem balance. At the same time, these regions have been selected for monitoring weather patterns, space radiation, and trans-border and global atmospheric contamination, with an active network of European high-mountain observatories monitoring all of these parameters (MASSON et al. 2016). This generates an increased interest in Alpine ecosystems due to the wealth of data, combined with the vulnerability of the high mountains to anthropogenic pressure (VERRALL & PICKERING 2020).

Complex radioecological and ecotoxicological biomonitoring has been conducted on Rila Moun-

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tain in Bulgaria during the period 1993-1996 within the framework of the French-Bulgarian OM2 project, principally identifying two appropriate monitor sites: Moussala peak (2925 m a. s. l.) as the highest monitoring point at Balkan Peninsula, as well as the coniferous forest belt (1400 – 1500 m a. s. l.) (IOVTCHEV et al.1995; IOVTCHEV et al.1996). Subsequent studies have been conducted two decades later in the same areas. On their basis, the most appropriate bioindicator and monitor small rodent species for different types of mountain ecosystems were identified. They are the snow vole (*Chionomys nivalis*) and wood mouse (*Apodemus sp.*) for the Alpine zone, *Apodemus sp.* and bank vole (*Myodes glareolus*) for the area of coniferous and mixed forests (IOVTCHEV et al.1996, SAMOURIS et al. 2007, OGUT 2015). Additionally, the snow vole (*Ch. nivalis*) has been well-characterised concerning its potential as a bioindicator species for Alpine ecosystems (IOVTCHEV et al.1996, METCHEVA et al. 2008). Radioecological monitoring nowadays shows a several-fold decrease in anthropogenic radioactive contamination after the initial peak following the Chernobyl accident in 1986 (BERESFORD 2016, BAVERSTOCK & WILLIAMS 2006). Whole-body total β -activity comparison results of the four rodent species were studied by OSTOICH et al. (2020). They demonstrated trends of an overall reduction by a factor of 10 between the two investigated time points.

There is a significant range of sources dealing with the use of small rodents as biomonitors of radioactive contamination (CHESSEY et al. 2001; CHESSEY et al. 2000; RODGERS et al. 2001; BERESFORD et al. 2008; OKANO et al. 2016). The studies in the Chernobyl Exclusion Zone in Ukraine have highlighted the suitability of the bank vole (*Myodes glareolus*) as a radioecological model organism due to its very high uptake of caesium radionuclides (^{134}Cs and ^{137}Cs) (CHESSEY et al. 2000). At the same time, the tundra vole (*Microtus oeconomus* Pallas, 1776) has been investigated in the past as a biomonitor for tundra ecosystems (TESTOV 1996; CHESSEY et al. 2001). It would be advantageous to compare biomonitoring data from Rila Mountain, Bulgaria, with results obtained from similar species elsewhere.

Not enough data exists about the place and role of small rodents as primary consumers and the degree of biomagnification from the plants as producers to the higher levels in the trophic chains.

The purpose of the current study is to present data on the biomonitoring potential of small rodent species from Rila Mountain (*Apodemus sp.* and *Ch. nivalis*) concerning radioecological studies. Data on food spectra and residual radioactivity in plant spe-

cies are presented. The analysis of the animal diet and the determination of the total β -activity in the dominant plant components of the food spectrum is a basis for further monitoring and calculating the radionuclides in food chains. These results could serve as a basis for future studies on the turnover of radionuclides in ecosystems. This study re-evaluates the radiological situation in the alpine zone of Rila Mountain concerning the diet of the small mammal species. Finally, conclusions are drawn regarding the comparative utility of different rodent species as suitable bioindicators for radioecological monitoring.

Materials and Methods

Small rodents, as well as the main plant species that composed animal's diet, were collected over 20 years from the same areas in the alpine zone of Rila Mountain, Bulgaria, at an altitude of 2925 m a.s.l. – the peak of Moussala. Wood mouse (*Apodemus sp.*) and snow vole (*Chionomys nivalis*) were used as model species for this investigation. The target internal organs like muscles, liver, and bones were collected separately. Whole-body total β -activity in Bq/kg was obtained for two rodent species from Moussala (*Ch. nivalis*, n=12, *Apodemus sp.*, n=12). The analysis of total β -activity was performed according to the procedures described in detail in OSTOICH et al. (2020).

Stomach content analysis was provided on the base of the food spectrum using snap traps captured animals (BOMFORD 1987). After a 24-hour preliminary lightening in glycerin, five glycerin-gelatin microscopic slides were prepared from each tested animal's stomach contents. Permanent histological slides from most plant species like *Achillea clusiana*, *Sesleria sp.*, and *Nardus stricta*, dominant in the studied alpine areas and expected to be prevalent in the animal's diet, were used as a key for food identification. Finally, an attempt was made to correlate the obtained results for the total β -activity of the rodent's body with the amount of β -activities of plants in the rodent's food spectrum.

Results

The data for total β -activity in the target internal organs and tissues in the investigated small rodent species from the alpine zone of Rila mountain are presented in Fig. 1. Comparisons were made using the same data and animal species as a study conducted 20 years ago, from 1993 to 1996 (OSTOICH et al. 2020).

The present results show higher β -activity in muscles. Comparable but slightly lower values were

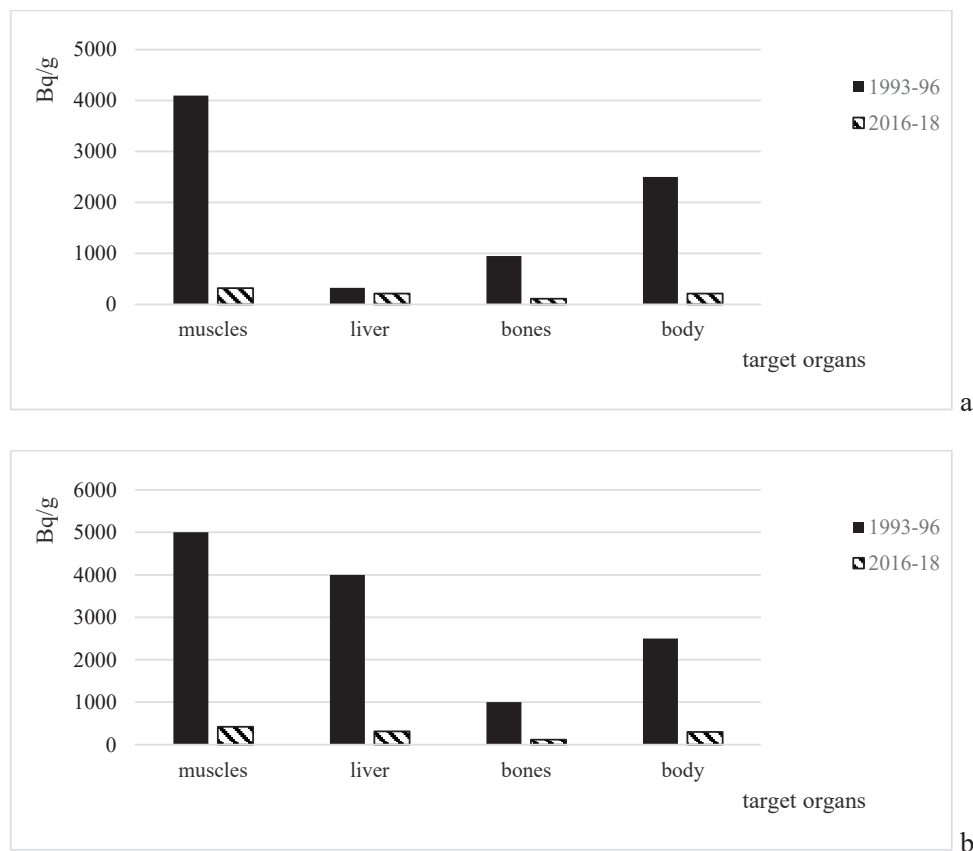


Fig. 1. Total β -activity in Bq/kg in the whole body and target organs and tissues of *Chionomys nivalis* (a) and *Apodemus sp.* (b) from the peak of Moussala in Rila Mountain in the beginning and the end of the twenty-year investigation period.

obtained for the liver, followed by bones. Species-specific differences in the accumulated amounts of total β -activity in the two small rodent species studied were found, but they nevertheless followed the trend:

muscles > liver > bones

It is well known that green vegetable parts accumulate radionuclides, particularly radiocesium, in higher quantities than seeds (METCHEVA et al. 2008). Therefore, in the studied area, the representatives of *Apodemus sp.*, primarily omnivores, accumulate lower doses of residual amounts of total β -activity than herbivorous voles. The conducted analysis of the food spectra of the two rodent species shows a significant dominance of the green vegetable parts in the diet of *Ch. nivalis*, with 88% of the diet consisting of greens and an additional 6% mosses. The representatives of *Apodemus sp.* are mainly omnivorous, with at least 9% animal matter in their diet, and strongly prefer seeds (~70% of the diet). The respective food spectra of the two investigated rodent species are presented in Fig. 2 and 3.

The food spectrum of *Ch. nivalis* correlates well with available food sources in the highest area of the peak Moussala, with a somewhat untypically high content of animal-based food in the diet (~6%

insects) compared to other vole species. This can nevertheless be explained by increased foraging for protein sources during the summer season.

Apodemus sp. displays a typical preference for a seed-based high-calorie diet, which is typical for the Muridae family representatives. Their adaptability and low levels of specialisation explain why *Apodemus sp.* can be found in the Alpine zone as well as in the coniferous/mixed forest in Rila Mountain.

In summary, the two rodents' analysed food spectra fit within their expected diet, with relatively little variation in the period 1993-2018 and slight differences with literature data that can be explained, in part, by the specific conditions of the investigated alpine habitat.

The current authors attempted to correlate the results for total β -activity of the two investigated rodent species to measured data from collected phytomonitor plant species in the periods 1993-1996 and 2017-2018 from the surroundings of peak Moussala. Five species of herbaceous plants, which form the overall part of the rodents' diet were selected and analysed for total beta activity: *Festuca spp.* (blue fescue), *Sesleria spp.* (blue moor grass), *Taraxacum officinale* (dandelion), *Nardus stricta* (matgrass),

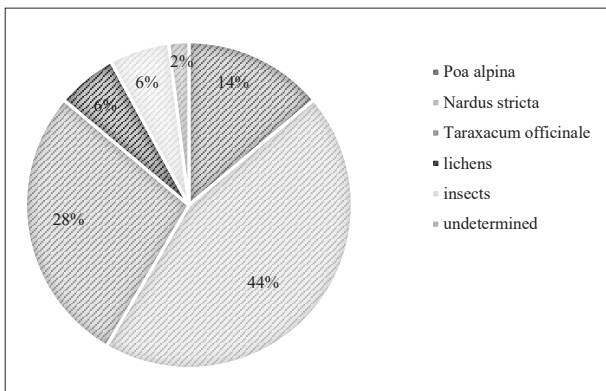


Fig. 2. Food spectrum of *Chionomys nivalis* from the investigated area of Rila Mountain.

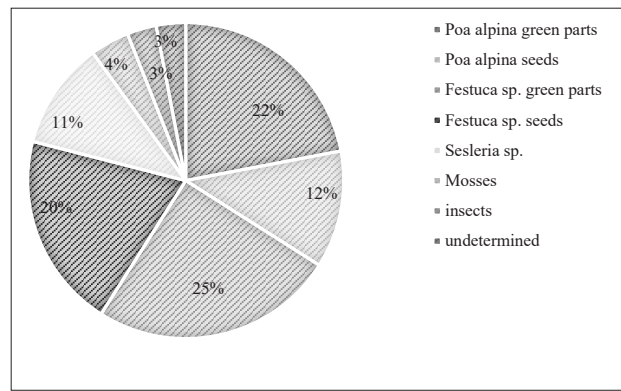


Fig. 3. Food spectrum of *Apodemus* sp. from the investigated area of Rila Mountain.

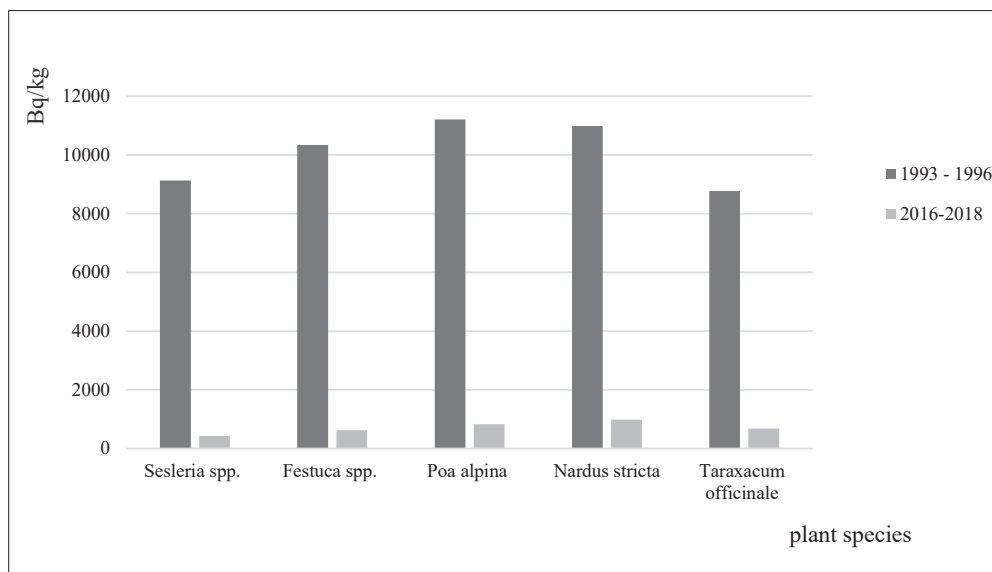


Fig. 4. Total β -activity in Bq/kg in plant species dominant in the animals' diet.

and *Poa alpina* (alpine meadow grass). The results are presented in Fig. 4.

The plant species display similar reduction factors in measured total β -activity: *Sesleria spp.* of 21.1, *Festuca spp.* – 16.7, *Poa alpina* – 13.6, *Taraxacum officinale* – 12.9 and *Nardus stricta* – a reduction factor of 11.1.

Discussion

The present results are in agreement with the current models of deposition, mobility, and decay of technogenic radioactive contamination developed for terrestrial ecosystems (BERESFORD et al., 2008). One of the most important anthropogenic radioisotopes, emitting beta-particles, is ^{137}Cs . In Bulgaria, as in all countries in South-Eastern Europe, the deposition of ^{137}Cs originates mainly from the fallout from the Chernobyl accident (BERESFORD et al. 2016). Recent

studies have indicated that the amount of ^{137}Cs in the surface soil layer of 0-5 cm for 2015 is comparable to that before the accident at the Chernobyl power plant. This decrease is mainly due to the natural decay of ^{137}Cs and, to a very small degree, to migration processes in the depth of the soil (YORDANOVA et al. 2016).

Due to toxic isotope biomagnification, including radionuclides, it is essential to track the flow of these elements through food chains. The health and sustainability of ecosystems where they circulate are closely dependent on the residual quantities of ^{137}Cs in the soil and in producers like plants.

The results highlight observations by other authors demonstrating the utility of the voles as biomonitors of residual radioactive contamination compared with omnivorous *Apodemus* species. Investigations conducted reached similar conclusions,

particularly noting the high bioaccumulation of caesium radionuclides (^{134}Cs and ^{137}Cs) in herbivorous species, which helps explain the comparatively lower reduction in measured total β -activity in *Ch. nivalis* (CHESSER et al. 2001; CHESSER et al. 2000; RODGERS et al. 2001; BERESFORD et al. 2008). The main residual radionuclides from Chernobyl at present are ^{137}Cs (half-life of about 30 years) and ^{90}Sr (half-life of about 29 years). Caesium tends to be more mobile in terrestrial ecosystems; it is expected that the main contribution to the total β -activity measured in *Ch. nivalis* and *Apodemus* sp. originates from incorporated ^{137}Cs . It is well established that caesium is preferentially accumulated in the green parts of plants and that voles have a high-volume, low-calorie diet with a strong preference for green plant matter. Together, these factors can explain why the snow vole displays a lower reduction factor of total β -activity among investigated rodent species, as well as the highest recorded values in the later investigated period. The decrease in total β -activity in small mammals depends on the radioactivity in plants comprising the main part of the animal's diet. The food spectra of the investigated rodents show slight variation over time and are consistent with data by other authors (BUTET & DELETTRE 2011). The available data on the total β -activity of plants includes 5 taxa from the investigated area. All these species form a main part of the diet of the investigated rodents and show comparable reduction factors between rodents from the alpine zone of Rila Mountain. This is somewhat unexpected, as the tendency of rodents to bioaccumulate caesium would lead to lower reduction factors in them. Nevertheless, this surprising observation leads us to believe that the rodent zoomonitors and the species they feed on respond in similar ways to the decrease in anthropogenic radioactive contamination.

Conclusion

It is assumed that the total β -activity in mammals should be less than 4800 Bq.kg⁻¹ (THORNE & VENNART 1976). If it is higher, measuring the concentrations of each separate radionuclide is necessary. The levels of total β -activity in the studied small rodents were relatively low compared to the established referent values (THORNE & VENNART 1976). The level obtained can be considered normal for the studied areas. It is possible to evaluate the presented values as a reference baseline and to use them for further monitoring investigations, as well as to trace the turnover of radionuclides through the trophic chain's levels in the ecosystems.

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