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Ecological Half-Life of ^{137}Cs in Fungi

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Abstract: The ecological half-life of ^{137}Cs was calculated individually for four symbiotrophic fungi species (*Boletus edulis*, *Imleria badia*, *Suillus luteus*, *Paxillus involutus*) at 10 sampling sites in the Chornobyl exclusion zone and in the Kyiv region. It was found that the maximum rate of excretion of ^{137}Cs from the fungi organisms is characteristic for the territory with the maximum levels of soil contamination, i.e., for a zone near Chornobyl Nuclear Power Plant. In areas with low ^{137}Cs content, a slowing down of the excretion rate predominates. These results reveal different fungal response to the distinct concentration levels of ^{137}Cs in forest ecosystems. This observation further suggests that radiocaesium can be selectively accumulated by fungi and used in their life processes. Presence of this ^{137}Cs retention mechanism in fungi leads to a longer contamination of woody plants-symbionts.

Keywords: Caesium-137; ^{137}Cs; ecological half-life; fungi; Chornobyl accident

1. Introduction

^{137}Cs is a long-lived, biologically significant radionuclide [1,2]. It is considered that ^{137}Cs is a major artificial radionuclide present in today's environment. Radiocaesium is a by-product of nuclear fission of uranium, and it is released in the environment during accidents at the nuclear power plants, nuclear weapons testing, etc. Ecological and effective half-lives are used to describe the processes of removing radionuclides from the environment. Effective half-life (T_{eff}) of ^{137}Cs is the time required to reduce the activity of radiocaesium by 50% per unit mass, without considering differences between internal and external contamination or the specifics of elimination mechanisms [3,4]. T_{eff} combines the physical decay and ecological losses, and it is affected by physical (e.g., radioactive decay, precipitation, washout), chemical (e.g., changes in oxidation state, adsorption) and biological (e.g., changes in the food chain) factors. Ecological half-life (T_{eco}) expresses the ecological losses only, and it is an integral value that combines all processes that cause a radioactivity decrease in the environment in addition to physical decay [5,6]. Since T_{eco} does not take physical decay into account, it is identical for different isotopes of the same element [7]. The mathematical correlation between T_{eff} and T_{eco} is described by the formula: $\frac{1}{T_{eff}} = \frac{1}{T_{1/2}} + \frac{1}{T_{eco}}$ where $T_{1/2}$ represents physical half-life, which is equal to 30.05 years for ^{137}Cs [8]. Determination of T_{eff} and T_{eco} were carried out for various objects and territories contaminated with radiocaesium as a result of global fallout, after nuclear weapons tests, accidents at nuclear power plants, and discharges and emissions from nuclear facilities [9–16]. T_{eff} values (as well as the T_{eco}) found in the scientific literature can differ from each other even when calculated for the same environmental objects. For example, the Savannah River Site (SRS) South Carolina (USA) case. In this area, ^{137}Cs was released into the environment from nuclear production reactors during the years 1954–1974, and global fallout. Reported T_{eff} values obtained from the long-term, a 30-year environmental monitoring (1974–2005), show 14.9 years for soil and 11.6 years for vegetation [17]. The follow up study, conducted

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from 2006 to 2011, revealed that incorporation of new data to the previous 30-year study led to an increase of ^{137}Cs T_{eff} values, namely for soil to 17.0 years and for vegetation to 13.4 years [4]. Based on these results, it was concluded that natural ecological processes have the potential to remove ^{137}Cs from ecosystems faster than a physical half-life. It must be noted that about 90% of the SRS is covered by forests, which limits the anthropogenic impact on the study area.

Broad ranges of ^{137}Cs T_{eco} values were obtained for mosses and liches collected in Ordu province (Turkey) in 1997 [18] and 2007 [19]. Ordu province is among the areas with the highest ground contamination by ^{137}Cs in the Eastern Black Sea region after the Chornobyl accident. T_{eco} for the mosses varies between 1.8 and 10.4 years (with the mean of 4.4 years). For lichens, even a larger range of T_{eco} values were obtained, 2.1 to 13.7 years with a longer average ecological half-life (5.6 years).

A similar average T_{eco} value (3 to 4 years) in lichens was obtained in the study performed in an Alpine region (Bad Gastein, Austria) [20]. T_{eco} was determined by comparing ^{137}Cs activity concentrations measured between 2001 and 2003 with those measured between 1993 and 1996.

T_{eff} and T_{eco} values are also used to determine the levels of ^{137}Cs contamination in non-biotic objects as river water [21], bottom marine sediments [16] and bottom sediments in reservoirs [14].

In contrast to the number of publications reporting accumulation of ^{137}Cs in fungi, e.g., [22–29], there are not many publications in the scientific literature focused on ^{137}Cs T_{eco} or T_{eff}. In the comprehensive work summarizing the results of long-term measurements of the specific activity of ^{137}Cs in various objects of forest ecosystems in European countries, T_{eco} values were determined [30]. Data for three fungal species were calculated, namely: (a) *Imleria badia* in T_{eco} = 5.5 ± 2.1 years samples collected in Germany (Oberschwaben); (b) *Cantharellus cibarius* T_{eco} = 1.9 years, and (c) *Boletus edulis* T_{eco} = 2.7 ± 0.5 years, both collected in Ukraine (Zhytomyr region). Based on ^{137}Cs activity, mass concentration analyses performed in various forest wild fungi in the Czech Republic within 1986 and 2011, the overall T_{eff} (5.6 ± 0.6 years) and T_{eco} (6.9 ± 0.7 years) values were calculated [5]. Environmental monitoring of the ^{137}Cs and ^{134}Cs content in different environmental objects was performed in Northern Finland, 2.5 years after the Fukushima accident [31]. ^{137}Cs T_{eff} determined for fungi was 3.5 ± 1.3 years. Despite the short collection period and a very low level of ^{137}Cs fallout in Finland after Fukushima accident, these values were in accordance with previous data obtained in studies performed after the Chornobyl accident. In the comprehensive study focused on radiocaesium in *Tricholoma* spp., relatively long T_{eco} (16–17 years) for *Tricholoma equestre* in Poland (Augustów Primeval Forest) was calculated [11].

In our previous study, we demonstrated that the processes of ^{137}Cs accumulation by fungi after the Chornobyl accident took place in two stages. The first stage is characterized by an annual (linear) increase of the ^{137}Cs content in the fungi body. In the second stage, a (exponential) decrease of ^{137}Cs concentrations was observed [12]. We also observed that these two stages had different time durations for fungi belonging to the different ecological groups. For example, in ecological groups of saprotrophs and xylotrophs (*Lycoperdon perlatum*, *Fistulina hepatica*, *Laetiporus sulphureus*) the first stage lasted from 1986 to 1989–1991 depending on the sampling site, i.e., approx. 5 years [32]. On the other hand, for the fungi belonging to the ecological group of symbiotrophs, the first stage lasted until the mid-1990s, i.e., approx. 10–12 years after the Chornobyl accident [12,32]. Since that time, when the maximum concentration values of ^{137}Cs were determined, i.e., end of the first stage, a decrease of the ^{137}Cs content in all ecological groups has been observed. However, fungal species belonging to saprotrophs and xylotrophs accumulate ^{137}Cs in much lower amount compared to symbiotrophic fungi.

In this work, we focus on the determination of ^{137}C ecological half-life for four symbiotrophic fungi species (*Boletus edulis*, *Imleria badia*, *Suillus luteus*, *Paxillus involutus*). In contrast to other published works, we carried out T_{eco} calculations for each fungi species separately on each exactly determined, relatively small sampling site. All sampling sites belong to the same ecotope, nemoral Scots pine forest, which excludes the anthropogenic impact on the study area. The sampling sites differ only in the levels of soil contamination with radiocaesium as a result of the accident at the Chornobyl Nuclear Power Plant in 1986.

2. Materials and Methods

2.1. Sampling Sites

Fungi samples employed in this study were collected from Chornobyl Nuclear Power Plant (ChNPP) Exclusion Zone and the south of Kyiv region. The area of each fungi sampling site was approx. 100 m^2. In order to minimalize the differences in ecosystems, all the sampling sites were chosen in the forests where Scots pine (*Pinus sylvestris* L.) aged between 40 and 50 years prevails among woody vegetation. This type of the forest is denominated by European Environment Agency (EEA) as Nemoral Scots Pine Forest [33]. The soil type at the sampling sites was the sod-podzol and these areas belong to A1 ecotope type [34]. Exact location (latitude and longitude in decimal degrees format) of the fungi sampling sites within the Chornobyl exclusion zone (i.e., 30-kilometre zone) and the Kyiv region are summarized in the Table 1 and are shown on the map (Figure 1). South part of Kyiv region was selected because this area was more contaminated by radiocaesium than others, except for the north (ChNPP direction) region [35]. Sampling sites are listed according to the distance from ChNPP. For better overview, the table also includes the equivalent dose rate (H_T) as measured in year 2006.

Table 1. Fungi sampling sites within the Chornobyl exclusion zone (No. 1–7) and Kyiv region (No. 8–10).

Sampling Site		GPS Coordinates	Distance from ChNPP	H_T (µSv/h)
1	Yaniv	30.06408 E, 51.39017 N	2.2 km	10.00
2	Prypiat	30.02982 E, 51.41249 N	5.2 km	2.50
3	Novoshepelychi	30.01030 E, 51.41910 N	6.7 km	2.00
4	Leliv	30.15856 E, 51.32505 N	8.5 km	1.20
5	Paryshiv	30.32473 E, 51.30069 N	18.8 km	0.30
6	Opachychi	30.30404 E, 51.197143 N	25.0 km	0.25
7	Dytyatky	30.12449 E, 51.13088 N	29.9 km	0.25
8	Stare	30.99528 E, 50.06785 N	159.8 km	0.18
9	Staiky	30.98756 E, 50.05959 N	160.6 km	0.15
10	Rzhyshchiv	31.08317 E, 50.01686 N	167.5 km	0.15

2.2. Preparation of the Fungal Fruit Bodies Samples

In this study we used widely spread, edible wild fungal species, namely: *Boletus edulis* according to Bulliard (Bull.) (ENG: cep or porcini mushroom, UKR: Білий гриб), *Imleria badia* according to Fries (Fr.) Vizzini (2014) (ENG: bay bolete, UKR: Польський гриб), *Suillus luteus* according to Linnaeus (L.) Roussel (1796) (ENG: slippery jack or sticky bun, UKR: Маслюк звичайний), *Paxillus involutus* according to Fries (Fr.) Batsch (1838) (ENG: brown roll-rim, UKR: Свинуха тонка). All examined species are obligate symbiotrophic fungi, except for *P. involutus*, which is a facultative symbiotroph. The fungi were collected during the period of mass emergence of the fruiting bodies; this period occurs between the end of September and the end of October. For the calculations of T_{eco} we used average values from several samplings for each sampling site and for each fungi species for individual years. To avoid possible variables of seasonal changes, we excluded from the analysis the years where no fungi were found. Fungi fruit bodies of average weight and without visible severe external damage were collected and cleaned from surface contaminations. Fungi fruit bodies of each species were packed into a separate polyethylene bag, labelled and delivered to a laboratory. The samples were homogenized using a blender, placed into a graduated vessel for measurements, and stored in a freezer at −18 °C until the measurements were performed. The samples were taken out of the freezer and allowed to thaw at room temperature for 24 h before the measurements. Each fungus sample consisted of 3–31 fruit bodies [13].

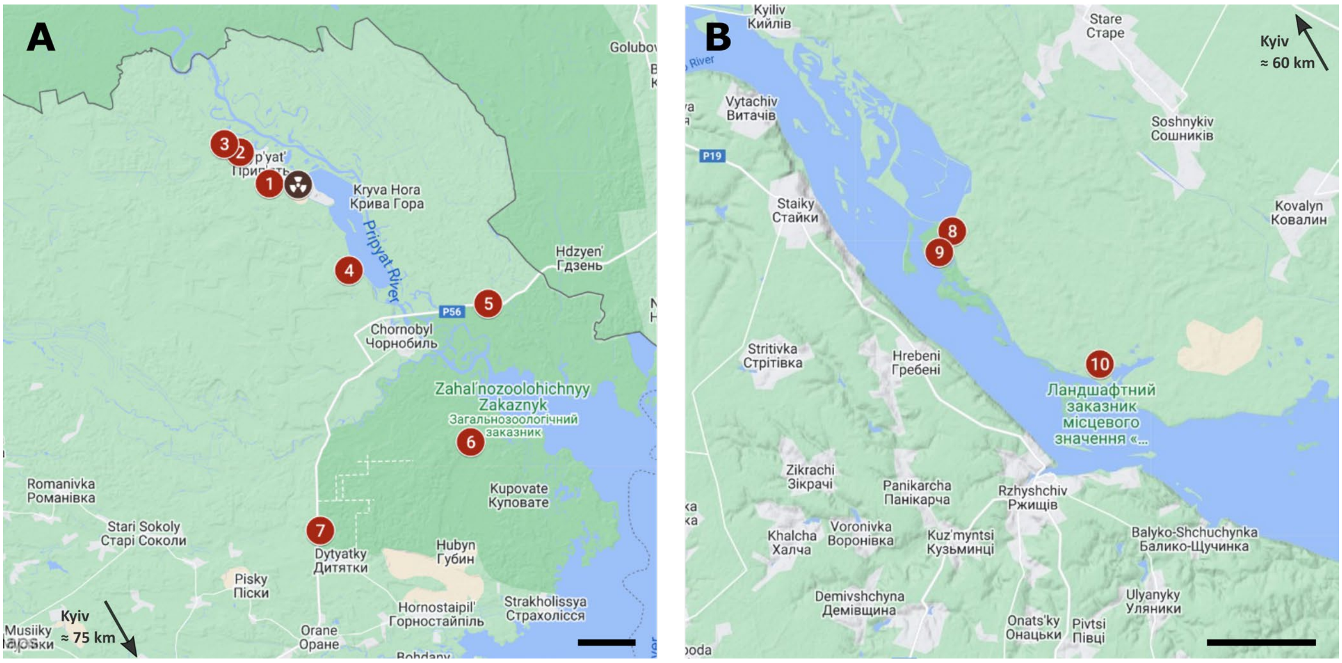

Figure 1. Fungi sampling sites within the Chornobyl exclusion zone (**A**) and Kyiv region (**B**). Scale bars represent 5 km (maps were created by Google My Maps (Google LLC)).

2.3. Radiometry

^{137}Cs specific activity measurements were performed employing a CANBERRA gamma-spectrometric set-up based on coaxial highly pure HPGe semiconductor detector, model GC6020 (Mirion Technologies, Atlanta, GA, USA). The detection unit was covered with 100-mm lead protection, allowing the effective measurement of samples with a relatively low radionuclide specific activity. Measurement time was from 600 to 14,400 s depending on the specific activity of the radionuclide in samples from the Chornobyl exclusion zone. In the samples taken outside the Chornobyl exclusion zone, the specific activity of ^{137}Cs samples was lower, so the sample measurement time was extended up to 86,400 s (24 h). The measurement errors of this series of samples did not exceed 10% and, as a rule, were within the limits of 3–5% of the radionuclide activity [13]. The ^{137}Cs specific activity (Bq/kg) in fungi was calculated on a raw, fresh weight basis.

2.4. Calculation of the Ecological Half-Life of ^{137}Cs

The rate of decay of radionuclides in environmental objects is characterized by the time during which the activity of that radionuclide decreases by a factor of 2. Radionuclide activity at time t is given by the formula: A(t) = A(0) $2^{-\mu t}$, where A(0) is the specific activity of the radionuclide at time zero. μ is a coefficient describing the rate of decay of activity with time; the greater the coefficient the faster radionuclide decays. When time $t = 1/\mu$, $2^{-\mu t}$ becomes 1/2 and A(t) = A(0)/2. If $1/\mu$ is denoted by T_{eff}, then $A(t) = A(0)\ 2^{-t/Teff}$. The exponent on a logarithmic scale is described by the straight line formula: L(t) = ln(A(0))−ln $2(t/T_{eff}) = b–at$, where b represents ln(A(0)) and a represents (ln 2)/T_{eff} (tangent of an angle of inclination of the straight line). From this it follows, T_{eff} = (ln 2)/a. Thus, we came up with the formula: $1/T_{eff} = 1/T_{1/2} + 1/T_{eco}$. The ecological half-life T_{eco} was calculated based on this ratio. $1/T_{eco}$ can be denoted by β—the rate of specific activity decreases in the ^{137}Cs value of the studied object. T_{eff}, T_{eco}, $T_{1/2}$ have the dimension of time (years), the dimension of β is 1/year.

In this work, we analysed data of our own long-term investigations of ^{137}Cs content in fungi on the territory of the Chornobyl exclusion zone and Kyiv region. The ecological half-life was calculated starting from the year when the maximum values of specific activity ^{137}Cs levels in the fruiting bodies of fungi in each sampling site were recorded. In order to enable a comparison of our data with those previously published, we report ^{137}Cs T_{eff} values as well.

Statistical analysis and graphical visualization were performed using GraphPad Prism 9.5.0 software (GraphPad Software, San Diego, CA, USA). Statistics were performed using one-way ANOVA and Tukey's multiple comparisons tests. A p-value ≤ 0.05 was considered as statistically significant.

3. Results

Based on the experimental values of the specific ^{137}Cs activity for each sampling site and fungi species, the rate of β concentration decrease of radiocaesium was calculated. In order to estimate the level of contamination for each sampling site, the maximum specific activity of ^{137}Cs in the fruiting bodies of fungi species (A_{max}) for the entire observation period was chosen. The β parameters were obtained as a function of lg(A_{max}). From the calculated β values, straight lines were fitted by the least squares method for each fungi species individually, across all sampling sites (Figure 2).

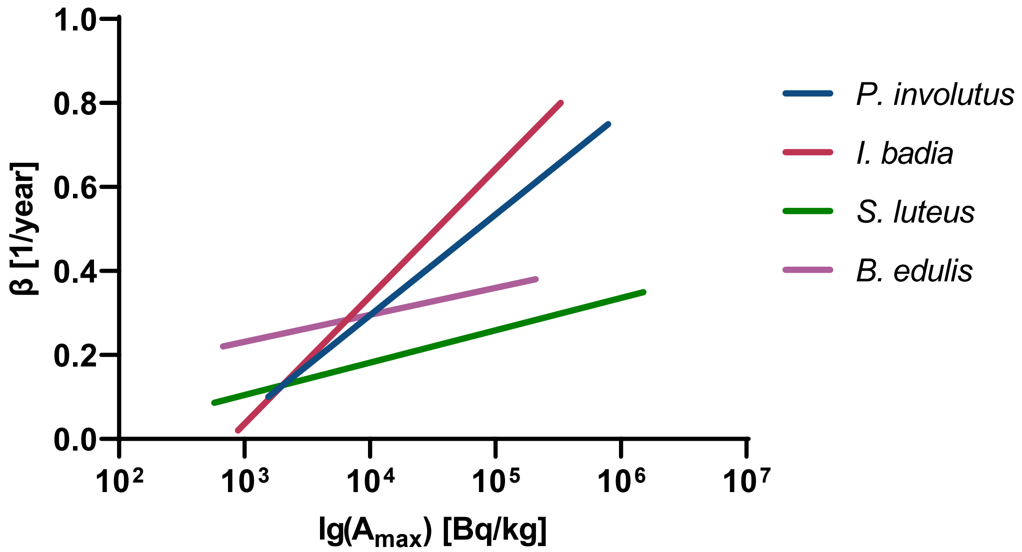

Figure 2. Dependence of the rate of ^{137}Cs concentrations decrease (β), on the logarithm of the ^{137}Cs specific activity (A_{max}) in fungi.

T_{eco} values for studied fungi species at individual sampling sites were calculated as inverse values of β. T_{eco} as well as T_{eff} values are summarised in the Table 2.

In general, the lowest values of T_{eco} for all fungi species were obtained in the samples collected on the sampling sites located near ChNPP (Yaniv, Prypiat, Novoshepelychi), i.e., on the sites with maximum ^{137}Cs contamination. This observation shows that the duration of the ecological half-life ^{137}Cs is not identical for fungi of the same species in different locations and it is inversely proportional to the levels of ^{137}Cs contamination in forest ecosystems. Low T_{eco} values (i.e., maximal radiocaesium concentration reduction rate) are characteristic for the territory of 7-km zone around ChNPP. On the other hand, highest T_{eco} values (up to 8+ years) were determined in distant locations as e.g., Stare or Rzhyshchiv. Differences of T_{eco} values for the same fungi species collected on different sampling sites represent a multiple up to 9 times, e.g., for *I. badia*. There is no statistically significant difference between the T_{eco} (or T_{eff}) values for individual fungi species depending on the sampling zones (Chornobyl vs. Kyiv regions), due to the high standard deviation and low number of the sample cohort (3–5 vs. 2). However, the same trend is observed for all studied species.

Table 2. T_{eco} and T_{eff} (values in parentheses) in years of ^{137}Cs in symbiotrophic fungi species.

Sampling Site		Fungi Species			
		S. luteus	*B. edulis*	*I. badia*	*P. involutus*
1	Yaniv	2.46 (2.27)			
2	Prypiat		3.54 (3.17)		
3	Novoshepelychi	3.14 (2.84)	1.73 (1.64)	0.93 (0.90)	1.16 (1.12)
4	Leliv			5.89 (4.92)	
5	Paryshiv	7.76 (6.17)	3.66 (3.26)	3.53 (3.16)	6.68 (5.47)
6	Opachychi		4.48 (3.90)		
7	Dytyatky	5.43 (4.60)	5.01 (4.29)	3.05 (2.77)	2.83 (2.59)
8	Stare			8.85 (6.84)	4.61 (4.00)
9	Staiky	5.94 (4.96)	7.56 (6.04)	3.36 (3.02)	3.35 (3.01)
10	Rzhyshchiv	8.21 (6.45)	2.79 (2.55)		

In order to analyse the data in more detail, we focused on the T_{eco} and T_{eff} values of all symbiotrophic fungi according to the sampling sites. For that, we divided the sampling sites into three groups according to the distance from ChNPP and H_T, i.e., group 1: ChNPP (No. 1–3, $H_T \geq 2.0$ µSv/h), group 2: Chornobyl exclusion zone (No. 4–7, H_T = 2.0 to 0.2 µSv/h), group 3: Kyiv region (No. 8–10, $H_T \leq 0.2$ µSv/h), Figure 3.

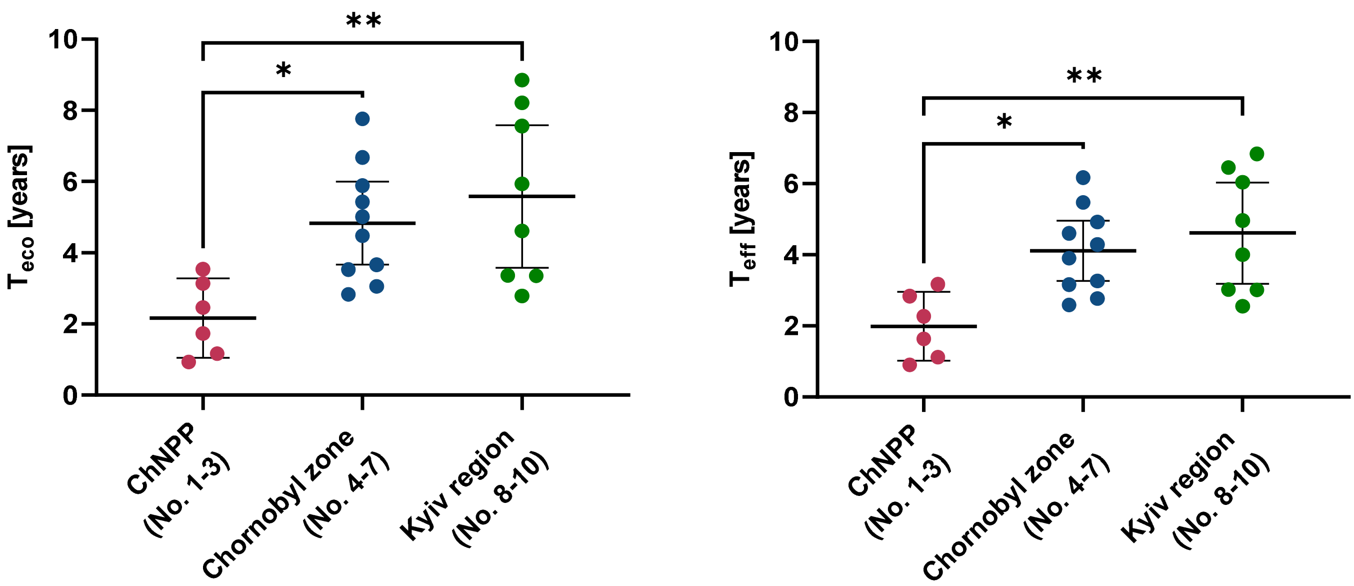

Figure 3. T_{eco} (**left**) and T_{eff} (**right**) values of symbiotrophic fungi according to the sampling site groups [red dots - group 1: ChNPP (No. 1–3, $H_T \geq 2.0$ µSv/h), blue dots–group 2: Chornobyl exclusion zone (No. 4–7, H_T = 2.0 to 0.2 µSv/h), green dots–group 3: Kyiv region (No. 8–10, $H_T \leq 0.2$ µSv/h)]. Data represent the mean and 95% confidence interval (CI) of the mean. Variables of significance (* $p \leq 0.05$, ** $p \leq 0.01$) were calculated by ordinary one-way ANOVA.

This data analysis shows that ^{137}Cs T_{eco} and T_{eff} values are significantly shorter in symbiotrophic fungi collected on the sampling sites at close vicinity to the ChNPP (No. 1–3) if compared to the fungi from more distant sampling sites (No. 4–7 or No. 8–10).

4. Discussion

Obtained values of the ^{137}Cs ecological half-life differ for each studied fungi species and for each sampling site. However, we observed a regular pattern when greater loss rate (i.e., lowest T_{eco} values) of ^{137}Cs by fungi was determined at locations with high levels of soil contamination. There is statistically significant difference in T_{eco} and T_{eff} values depending on the sampling site groups, even if both groups (Figure 3, No. 1–3 vs. No. 4–7) are located within 30-km Chornobyl zone. The different durations of the ecological half-life indicate the existence of differences in the mechanism of accumulation vs. removal of ^{137}Cs in fungi depending on pollution levels.

Pröhl and co-workers raised a hypothesis that the reason for the different T_{eco} values for the same studied objects is due to the differences in the type of soil and its heterogeneity [30]. Having this in mind, we selected sampling sites with the same sod-podzolic soil type. In order to minimalize the differences in ecosystems and possible anthropogenic impact on the study area all sampling sites were in nemoral Scots pine forests. Furthermore, collection of fungi was carried out practically during the same year period which excluded an influence of seasonal changes of radiocaesium concentration. Overall, in the present study, the main difference between sampling sites are different levels of soil ^{137}Cs contamination after the Chornobyl accident. Soil contamination is the main factor influencing the process of ^{137}Cs accumulation by fungi [13,36,37]. The maximum contribution to total soil contamination is made by fungi mycelium, mycelium contains from 10 to 63% of the total ^{137}Cs stock in the forest ecosystem [38,39].

Differences in the ecological half-life of ^{137}Cs on different sampling sites indicate distinct biota response to the different levels of ^{137}Cs contamination. Fungi from highly polluted areas remove more intensively "excessive" amounts of ^{137}Cs from the organism. At the same time, we see a slowing down of the ^{137}Cs removal rate from fungi collected in areas with low ^{137}Cs content. This is evident from Figure 3, as there is no statistically significant difference between T_{eco} (T_{eff}) values for greater Chornobyl zone (No. 4–7) vs. south part of Kyiv region (No. 8–10). Therefore, we hypothesize that the rate of ^{137}Cs concentrations decrease in fungi is greater in the areas with higher contamination of forest ecosystems.

There is a suggestion that radiation exposure at very low doses is beneficial, i.e., it has hormetic effects, for animals and plants [40–44]. According to our results of the ecological half-life, we suppose that the effect of incorporated ^{137}Cs in low concentrations is like the effect of X-rays, γ-rays or low doses of ^{40}K and it has a beneficial effect on fungi. Probably, the reaction of fungi to this positive impact is the activation of mechanisms that slow down the elimination of radiocaesium from their organism as much as possible. We hypothesize that fungi selectively accumulate ^{137}Cs in small amounts and use it in their life processes.

5. Conclusions

We determined T_{eco} values for four individual fungi species separately on an exactly defined sampling site as a result of our long-term radioecological monitoring. We showed that the ^{137}Cs T_{eco} in fungi of one species differs from the same species in the territories with different levels of soil contamination caused by the Chornobyl accident. The maximum removal rate of radiocaesium from all studied fungi species was observed at the sampling sites in close vicinity to ChNPP. Based on our experimental data, we hypothesize that there are mechanisms allowing fungi to accumulate ^{137}Cs selectively and use it in the processes of their vital activity. From this assumption, we conclude that the existence of a ^{137}Cs retention mechanism by fungi leads to a longer contamination of woody plants-symbionts, as the circulation of radiocaesium occurs along the soil-fungi-woody plants-fungi-soil chain. It would be desirable in the future to carry out calculations of the ecological half-life for other objects of forest ecosystems in areas contaminated with ^{137}Cs, e.g., lichens, fishes, bottom sediments.

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