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Role of Fungi in the Determination of the Radiological Status of Terrestrial Ecosystems

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ABSTRACT

In an attempt to assess the importance of the role of fungi as bioindicators in terrestrial ecosystems, we compiled the results reported by more than 100 authors worldwide. These mostly were centred on the ^{137}Cs and ^{40}K contents. The genera *Paxillus*, *Xerocomus*, *Cantharellus*, and *Hebeloma* were among those with the highest radiocaesium contents. Studies in countries not highly contaminated in the Chernobyl accident, which included other anthropogenic and natural radionuclides in the assays find the order of relative accumulation to be: $^{40}K \geq {}^{137}Cs > {}^{228,230,232}Th \approx {}^{234,238}U \approx {}^{226}Ra \approx {}^{90}Sr >> {}^{239+240}Pu > {}^{241}Am$. Laboratory studies showed that the relative position of anthropogenic radionuclides (mainly radiocaesium and radiostrontium) depends on the moment at which fallout occurred. In particular, if the radionuclide deposition occurred long before the formation of the fruiting bodies, the accumulation of radiocaesium was higher than that of radiostrontium. Other factors, such as the concentration of stable elements and the bioavailability of the different radionuclides in the soil, also affect the radioactive content of the fruiting bodies and hence the dose due to their consumption. While the dose from ingestion of mushrooms in areas clearly affected by the Chernobyl Nuclear Power Plant accident is currently estimated at 1.8 mSv/year due to ^{137}Cs, the maximum dose in unaffected areas estimated on the basis of the present findings is $4.60 \cdot 10^{-3}$ mSv/year for the main natural and anthropogenic radionuclides present in the environment.

Keywords: anthropogenic radionuclides, dose, mushroom, natural radionuclides, radiocaesium

CONTENTS

INTRODUCTION

As a consequence of the atmospheric nuclear weapon tests carried out from the end of World War II until around the mid-1960s, and of accidents involving nuclear materials such as Thule, Palomares, Chernobyl, and the re-entry of SNAP-9 (UNSCEAR 1982), large quantities of anthropogenic radionuclides have been released into the environment. They can be transferred along the trophic chain and finally incorporated by humans, with the resulting implied health risk. Therefore, their content and transfer in different products – basically milk, meat, fish, vegetables, and their derivatives – have frequently been analyzed (Ng 1982). However, little attention was paid in these studies to the consumption of mushrooms. We consider that there may have been two reasons. On the one hand, mushrooms are strongly seasonal, appearing mainly in autumn and to a lesser degree in spring. This, together with their short life span led to the inference that they did not constitute a part of fresh market produce during a significant part of the year. And on the other hand, since mushrooms are consumed mainly by only a small number of people, usually of rural origin, it was supposed that the dose due to mushroom ingestion might be less than that of other produce.

However, after the Chernobyl Nuclear Power Plant accident in 1986, there was an increase in the number of studies of the radioactive contamination in mushrooms. Their focus was mainly on ^{137}Cs, and to a lesser extent on ^{90}Sr. These radionuclides are usually considered to be indicators of radioactive contamination risk. Both are products of nuclear

Received: 27 February, 2007. Accepted: 11 April, 2007.

Invited Review

fission, with significant production rates and relatively long half-lives, and they are chemical analogues of biologically important cations – potassium and calcium, respectively. From these studies, two surprising results arose. The radionuclide content, especially ^{137}Cs, of mushrooms was higher than that of other food items collected in the same area (Horyna 1991; Ruig *et al*. 1992; Skuterud *et al*. 1997), and generally higher than other forest products such as bilberries (Mietelski *et al*. 1996; IAEA 2006). In a review of the radioactive contamination in the Arctic (Alaska, Artic Finland, Artic Norway, Artic Sweden, Greenland, Iceland, Northern Canada, Northern Russia), Strand *et al*. (2002) found that the order of the level of ^{137}Cs contamination in products harvested in natural and semi-natural ecosystems was: Reindeer (~800 Bq/kg), mushrooms (~300 Bq/kg), freshwater fish (~50 Bq/kg) > lamb meat, goat cheese (~100-200 Bq/kg) > potatoes, vegetables (~3-9 Bq/kg) >> marine products (~0.1 Bq/kg).

Although the dietary ingestion of mushrooms was estimated at 7.8%, they presented the second highest ^{137}Cs content, only surpassed by reindeer meat.

In addition, the ecological half-life of radionuclides in mushrooms is longer than in other products (Shutov *et al*. 1996; Skuterud *et al*. 1997). This may explain the annual increase in some compartments of the trophic chain in highly contaminated areas (Shutov *et al*. 1996; Skuterud *et al*. 1997). Their consumption may even be a significant percentage of people's annual dose (Mietelski *et al*. 1994; Shutov *et al*. 1996; Skuterud *et al*. 1997).

Another pathway of incorporation to man is through the consumption of mushrooms by animals. Several studies have found a relationship between the consumption of mushrooms by ruminants, and the presence of radionuclides in their meat or milk. There have also been observed seasonal variations in the body content of radiocaesium in these animals due to the seasonal availability of mushrooms and lichens (Hove *et al*. 1990; Strandberg *et al*. 1994a; Mehli *et al*. 1998; Ávila *et al*. 1999; Zibold *et al*. 2001; IAEA 2006).

ACTIVITY LEVELS IN MUSHROOMS

Radiocaesium

In the literature, the radionuclides most extensively studied are two radiocaesium isotopes – ^{134}Cs and ^{137}Cs. As can be seen in **Fig. 1**, more than 90% of the studies that we consulted included radiocaesium assays. These studies were carried out mainly in northern, central, and eastern European countries, i.e., those that were most affected by the Chernobyl accident. We found only a small number of studies for the Americas (only Canada and Mexico) and Asia (only Japan and Taiwan). No references were found for Africa or Oceania. **Table 1** lists the ranges of ^{137}Cs content in mushrooms collected in different countries around the world, with the species that presented high contents and the corresponding references. The ^{137}Cs content is usually expressed on the basis of dry weight of mushroom, and only to a lesser extent on the fresh weight. In the latter case, we normalized the activity levels by assuming that the dry weight was about 10% of fresh weight. The most significant fact was the high variability of the accumulation of radiocaesium in mushrooms. The ^{137}Cs content in mushrooms ranged over about eight orders of magnitude, from 0.4 Bq/kg d.w. to 50 MBq/kg d.w. in some areas highly contaminated by the Chernobyl accident. Some studies have found the radiocaesium content to vary over about two or three orders of magnitude within mushrooms collected in the same area, and within mushrooms of the same species collected in different locations (Gentili *et al*. 1991; Mietelski *et al*. 1994). In an analysis of the influence of the intraspecific variation of ^{137}Cs activity concentration in sporocarps of *Suillus variegatus* in seven Swedish locations, Dahlberg *et al*. (1997) found that 40% of the variance of the ^{137}Cs content was explained by site variations, but that the other 60% could not be explained by genetic differences.

The distribution of radiocaesium content in mushrooms of a given population has been found to follow a non-symmetric, log-normal type of distribution (Mietelski *et al*. 1994; Zibold *et al*. 2001; Baeza *et al*. 2004a). It is therefore advisable not to use mean values and standard deviations to evaluate the degree of radiocaesium contamination of mushrooms in an ecosystem. In our opinion, it is better to describe it using the median value and the range observed.

Dependence on the nutritional mechanism of the mushroom has also been reported. Mycorrhizae, forming symbioses with tree roots, present higher ^{137}Cs contents than saprophytes or parasites (Guillite *et al*. 1994; Kammerer *et al*. 1994; Yoshida *et al*. 1994b; Baeza *et al*. 2004a; IAEA 2006). This factor was considered to be more important than the environmental conditions or the properties of the soil in which the mushrooms were growing (Heinrich 1992). Thus the relative order of radiocaesium contamination can be summarized as: Mycorrhiza > saprophyte ≈ parasite.

Some of the species that were found to have high radiocaesium contents are listed in **Table 1**. Most are mycorrhizae such as *Hydnum repandum*, *Paxillus involutus*, *Rozites caperata*, *Xerocomus badius*, and some species of the genera *Hebeloma*, *Cortinarius*, and *Lactarius*. Some authors suggest that mycorrhizae act as a filter through which their host plants take up nutrients. In this process, the host plant might discriminate against caesium in favour of potassium (Guillite *et al*. 1994; Kammerer *et al*. 1994). This hypothesis was confirmed to some extent in laboratory experiments, with the observation of a decrease in the uptake of ^{134}Cs by germinated seeds of *Picea abies* colonized by *Hebeloma crustuliniforme* mycorrhiza (Riesen *et al*. 1996) and an accumulation of ^{134}Cs in fungal hyphae (Brunner *et al*. 1996),

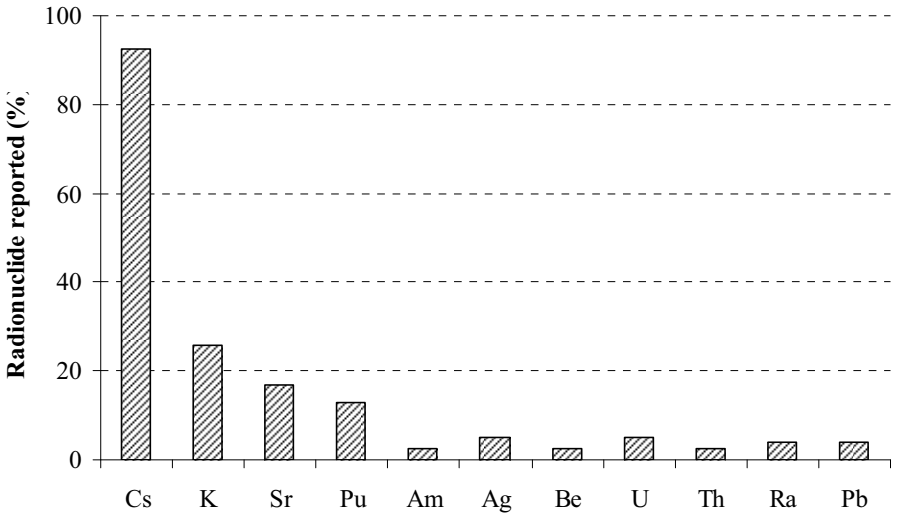

Fig. 1 Percentage of the studies consulted in the literature as listed in the References section which report activity levels for different radioelements.

Table 1 Range of ^{137}Cs content in mushrooms, expressed in Bq/kg d.w., in different countries worldwide, together with the species that showed major accumulations of radiocaesium and the corresponding literature references.

CONTINENT/ Country	Range (Bq/kg d.w.)	Species	References
EUROPE			
Austria	148 – 37370	*Xerocomus badius*, *Hydnum repandum*, *Rozites caperata*	Teherani 1987; Teherani 1988; Ismail *et al*. 1995
Belgium	160 – 102000	*Cortinarius brunneus*, *Cortinarius armillatus*, *Laccaria amethysana*, *Paxillus involutus*	Andolina *et al*. 1990; Fraiture *et al*. 1991; Guillite *et al*. 1991; Lambinon *et al*. 1998
Bulgaria	41 – 990	*Craterellus cornucopioïde*	Calmet *et al*. 1998
Czech Republic	50 – 150700	*Laccaria amethysana*, *Cortinarius armillatus*, *Xerocomus badius*	Řanda *et al*. 1990; Cibulka *et al*. 1996; Calmet *et al*. 1998; Dvořák *et al*. 2006
Denmark	212 – 13343	*Rozites caperata*, *Cortinarius alboviolaceus*	Strandberg *et al*. 1994a; Strandberg 1994b, 2004
Finland	58.6 – 121400	*Hydnum* sp., *Rozites caperata*	Ikáheimonen *et al*. 2003
France	2.5 – 5595	*Cantharellus* sp., *Rozites caperata*, *Xerocomus badius*, *Hydnum repandum*	Calmet *et al*. 1998; Kirchner *et al*. 1998
Germany	1000 – 11290	*Hebeloma* sp., *Hydnum repandum*, *Paxillus involutus*, *Rozites caperata*	Rückert *et al*. 1990; Römmelt *et al*. 1990; Heinrich 1993; Kammerer *et al*. 1994; Zibold *et al*. 2001
Great Britain	0.4 – 30500	*Hydnum repandum*, *Boletus badius*, *Cantarellus cibarius*	Barnett *et al*. 1999, 2001; Toal *et al*. 2002
Hungary	3 – 714	*Tricholoma terreum*, *Suillus granulatus*	Vaszari *et al*. 1992
Italy	95 – 135575	*Cantharellus lutescens*, *Clytocibe infandibuliformis*	Battiston *et al*. 1989; Giovanni *et al*. 1990; Borio *et al*. 1991; Gentili *et al*. 1991; Ingrao *et al*. 1992
Lithuania	42 – 3400	*Tricholoma equestre*, *Cantharellus cibarius*	Calmet *et al*. 1998
Norway	500 – 445000	*Lactarius* sp., *Amanita fulva*, *Amanita vaginata*, *Cortinarius armillatus*, *Rozites caperata*	Bakken *et al*. 1990; Hove *et al*. 1990; Amundsen *et al*. 1996
Poland	40 – 156700	*Xerocomus badius*, *Sarcodon imbricatum*	Bem *et al*. 1990; Mietelski *et al*. 1993; Grabowski *et al*. 1994; Mietelski *et al*. 1994; Zagrodski *et al*. 1994; Mietelski *et al*. 1996; Pietrzak-Fils *et al*. 1996; Calmet *et al*. 1998; Mietelski *et al*. 2002; Malinowska *et al*. 2006
Romania	85 – 360	*Hydnum repandum*	Calmet *et al*. 1998
Slovakia	323 – 966	*Suillus luteus*, *Russula aeruginea*	Čipáková 2004; Dvořák *et al*. 2006
Spain	0.5 – 647	*Hebeloma cylindrosporum*, *Lactarius deliciosus*	Arrondo 1988; Paniagua 1991; Baeza *et al*. 2004a
Switzerland	3 – 2000	*Boletus edulis*, *Rozites caperata*, *Xerocamus badius*	Froidevaux *et al*. 2006
Sweden	20 – 950000	*Dermocybe cinnamomea*, *Hygrophorus* sp., *Rozites caperata*, *Russula decolorans*, *Suillus variegates*, *Cortinarius collinitus*	Mascanzoni 1990; Guillitte *et al*. 1994; Smith *et al*. 1994; Dahlberg *et al*. 1997; Nikolova *et al*. 1997; McGee *et al*. 2000; Andersson *et al*. 2001
Ukraine/Russia	210 – 50700000	*Paxillus involutus*, *Xerocomus badius*, *Lactarius torminosus*	Hoshi *et al*. 1994; Smith *et al*. 1994; Tsvetnona *et al*. 1994; Lux *et al*. 1995; Mietelski *et al*. 2002; Travnikova *et al*. 2002; Vinichuk *et al*. 2003, 2004
Yugoslavia	360 – 117000	*Cortinarius armillatus*, *Laccaria amethysana*	Byrne 1988
AMERICA			
Canada	2 – 560	*Leucopaxillus giganteus*, *Laccaria laccata*	Mihok *et al*. 1989; Smith *et al*. 1994
Mexico	2 – 1357	*Clavariadelphus truncatus*, *Gomphus floccosus*, *Cortinarius caerukescens*	Gaso *et al*. 1998; Gaso *et al*. 2000
ASIA			
Japan	1.01 – 16300	*Hebeloma* sp., *Tricholoma flavovirens*	Muramatsu *et al*. 1991; Tsukada *et al*. 1998; Yoshida *et al*. 1994a, 1994b; Ban-Nai *et al*. 1997; Yoshida *et al*. 1998; Kuwahara *et al*. 2005
Taiwan	1.1 – 7.3	*Ganoderma tsuga*, *Lentinula edodes*	Wang *et al*. 1998

although no fruiting body was obtained in these experiments. One species analyzed in most of the studies was *Xerocomus badius*. According to Kottke *et al*. (1998), it contains a large amount of macro- and micro-elements (P, K, Ca, Mg, Fe, Zn, Mn, and Al) which they take to be connected to activity in the hyphal sheath and to the frequent occurrence of vascular bodies where deposition can occur. It was also found that some pigments present in the cap – derivatives of pulvinic acid, badione A1 and norbadione A2 – bind potassium and caesium (Aumann *et al*. 1989). These compounds are present in the brown pileus of fresh *Xerocomus badius*, and presented an enrichment factor of 3.4 regarding the activity of the fruiting body, quantified by γ spectrometry.

Prior to the Chernobyl accident, there were very few studies on radiocaesium accumulation in mushrooms. A wide range of variation was, however, observed from 370 to 18130 Bq/kg d.w. in mushrooms collected in different countries in Europe, with *Cortinarius armillatus*, *Rozites caperata*, and *Paxillus involutus* being the species that presented the highest ^{137}Cs content (Haselwandter, 1978; Bem *et al*. 1990). Bem *et al*. (1990) found a sharp increase of the ^{137}Cs content in mushrooms collected in Poland due to that accident – from a maximum activity level of 2704 Bq/kg d.w. with *Paxillus involutus* having the highest content before, to 8855 Bq/kg d.w. in *Xerocomus badius*, *Tricholoma flavovirens*, and *Paxillus involutus* after. Following Chernobyl, there were two sources of radiocaesium in soils: ^{137}Cs from global fallout, and ^{134}Cs and ^{137}Cs from Chernobyl. But their bioavailability in soils was considered to be different – ^{137}Cs pre-Chernobyl was considered to be more fixed to soil particles than "fresh" ^{134}Cs, due to the time that had passed since the global fallout (Muramatsu *et al*. 1991). Therefore the comparison of the ratio ^{134}Cs/^{137}Cs in mushrooms with that from Chernobyl fallout was used to determine the contribution of the latter to the ^{137}Cs content. In this way, Mietelski *et al*. (1994) found that in Poland, *Xerocomus badius* was able to accumulate about 3 kBq/kg d.w. of ^{137}Cs pre-Chernobyl.

Other anthropogenic radionuclides

The second most studied anthropogenic radionuclide, ^{90}Sr, presented a lower content in mushrooms, and less variabi-

lity. The variation was about three orders of magnitude considering studies worldwide (Mascanzoni *et al*. 1990; Römmelt *et al*. 1990; Paniagua 1991; Kozhevnikova *et al*. 1993; Mietelski *et al*. 1993; Baeza *et al*. 2004b, 2006b). The highest ^{90}Sr content found was 311 Bq/kg d.w. for *Paxillus involutus* in the 30 km zone around Chernobyl (Lux *et al*. 1995). For Poland, Germany, Sweden, Finland, and Spain, the highest activity level was about 4-5.6 Bq/kg d.w. for *Boletus edulis* and *Amanita porphyria*. The ratio ^{90}Sr/^{137}Cs for these same species and in different ecosystems was much less than unity, within the range 0.001-0.70 (Mascanzoni *et al*. 1990; Baeza *et al*. 2004b), indicating that mushrooms are able to accumulate ^{137}Cs better than ^{90}Sr. The strontium content of mushrooms was less than that found in other plants in the same ecosystems (Römmelt *et al*. 1990; Yoshida *et al*. 1998).

The isotope ^{110m}Ag has only occasionally been detected in Italy, Hungary, and Yugoslavia. Due to its short half-life ($T_{½}$ = 234 d) it was only detected after some recent deposition of radionuclides, such as after the Chernobyl accident. The highest activity level was 1872 Bq/kg d.w., detected in *Lycoperdon perlatum* collected in Italy. The ratio ^{110m}Ag/^{137}Cs was in the range 0.013-2.85 (Byrne 1988; Battiston *et al*. 1989; Gentili *et al*. 1991; Vaszari *et al*. 1992). From the above results, Byrne *et al*. (1979) concluded that the genus *Lycoperdon* can accumulate silver better than caesium (Byrne *et al*. 1979).

The content of $^{238,239+240}$Pu was lower than that of other anthropogenic radionuclides considered. The values of the $^{239+240}$Pu content ranged over about five orders of magnitude, from 1.2 mBq/kg d.w. to 82.8 Bq/kg d.w. (Mietelski *et al*. 1993; Mietelski *et al*. 1994; Lux *et al*. 1995; Yamamoto *et al*. 1995; Mietelski *et al*. 2002; Outola *et al*. 2003; Baeza *et al*. 2004b, 2006b). But the range was 0.39-82.8 Bq/kg d.w. considering only the Ukrainian mushrooms. *Cantharellus cibarius* presented the highest $^{239+240}$Pu activity, followed by *Xerocomus badius*, *Paxillus involutus*, and *Suillus luteus* (Lux *et al*. 1995; Mietelski *et al*. 2002). In other countries (Finland, Poland, and Spain), the range was lower (1.2-161 mBq/kg), and the species that had high contents were *Hebeloma cylindrosporum*, *Amanita muscaria*, and *Xerocomus badius*. The ratio ^{238}Pu/$^{239+240}$Pu reflected the origin of the contamination – for Ukraine samples it was about 0.5, confirming their Chernobyl origin, and lower for samples from other countries less affected by the Chernobyl fallout. The ^{241}Am content of mushrooms has only been analyzed in Finland and Spain (Outola *et al*. 2003; Baeza *et al*. 2006b). The range was 5.4-40 mBq/kg d.w., with *Clitocybe* sp., *Hebeloma cylindrosporum*, and *Lycoperdon perlatum* presenting the greatest accumulations. In general, the ratio ^{241}Am/$^{239+240}$Pu was less than unity.

In a few studies carried out after the Chernobyl accident, other anthropogenic radionuclides were detected in mushrooms (Battiston *et al*. 1989; Romeo *et al*. 1994; Lux *et al*. 1995). ^{125}Sb was only detected in *Russula vesca* and *Amanita muscaria*, 10,310 and 6755 Bq/kg d.w., respectively, collected within the 30 km zone around the Chernobyl plant (Lux *et al*. 1995). In the same study, the ^{60}Co, ^{106}Ru, ^{144}Ce, and ^{155}Eu contents of mushrooms were below the detection limit. ^{103}Ru was detected in Austria, Italy, and Ukraine in the range 10-1480 Bq/kg d.w., with *Macrolepiota procera*, *Cantharellus* sp., and *Clitocybe infundibuliformis* presenting the highest activity levels (Teherani 1987; Battiston *et al*. 1989; Romeo *et al*. 1994). For ^{131}I, we found only one reference – *Amanita rubescens* with 1.1 – 44.4 Bq/kg (Romeo *et al*. 1994).

Natural origin radionuclides

Of the radionuclides of natural origin, ^{40}K is the most extensively studied in the literature (see **Fig. 1**) (Muramatsu *et al*. 1991; Vaszari *et al*. 1992; Mietelski *et al*. 1994; Wang *et al*. 1998; Baeza *et al*. 2004a). The range of variation of ^{40}K content in mushrooms was not wide: 10^2-10^3 Bq/kg d.w., independently of the species considered or the location in which the mushrooms were collected. The frequency distribution for ^{40}K is symmetric and almost Gaussian, differing significantly from that of radiocaesium (Mietelski *et al*. 1994; Baeza *et al*. 2004a). It was also observed that ^{40}K content did not depend on the nutritional mechanism of the mushroom – mycorrhizae and saprophytes had the same mean values and standard deviations (Yoshida *et al*. 1994b; Baeza *et al*. 2004a).

The uranium content in mushrooms was in the range $5.88 \cdot 10^{-3}$ – 11.3 Bq/kg d.w. (Yamamoto *et al*. 1995; Mietelski *et al*. 2002; Baeza *et al*. 2004b; Jia *et al*. 2004). The highest activity was found in Kosovo, former Yugoslavia (Jia *et al*. 2004), but unfortunately the species was not mentioned. The maximum content of uranium in mushrooms collected in Spain was 7.0 Bq/kg d.w. for *Amanita ponderosa*, followed by *Russula cessans* and *Hebeloma cylindrosporum* (Mietelski *et al*. 2002; Baeza *et al*. 2004b). In Poland, the maximum activity was 240 mBq/kg d.w. in *Macrolepiota procera*, followed by *Xerocomus badius* (Mietelski *et al*. 2002). The ^{234}U/^{238}U ratio was around unity so that the two isotopes were in equilibrium in these mushrooms, and the ^{235}U/^{238}U ratio confirmed the natural origin of the uranium. In Kosovo, ^{236}U was also detected in one mushroom sample, with the ^{236}U/^{238}U ratio confirming the presence of depleted uranium in that species (Jia *et al*. 2004).

Thorium isotopes in mushrooms have only been analyzed in Poland and Spain (Mietelski *et al*. 2002; Baeza *et al*. 2004b). The ^{228}Th, ^{230}Th, and ^{232}Th contents varied within the ranges 1.8-13 Bq/kg d.w., $2.5 \cdot 10^{-3}$-6.5 Bq/kg d.w., and $2.5 \cdot 10^{-3}$-10.7 Bq/kg d.w., respectively. The activities were in the order ^{228}Th > ^{232}Th > ^{230}Th. The higher levels of ^{228}Th relative to ^{232}Th were probably due to the uptake of ^{228}Ra which later decayed to ^{228}Ac and then to ^{228}Th (Baeza *et al*. 2004b). A clear difference was observed between mushrooms from Poland and Spain. The ^{230}Th and ^{232}Th contents in Polish mushrooms were of the order of mBq/kg d.w., and ^{228}Th was not measured. Spanish mushrooms presented higher activity levels, up to the order of Bq/kg d.w., with the highest contents being in *Amanita ponderosa*, *Russula cessans*, *Tricholoma terreum*, and *Hebeloma cylindrosporum* for ^{228}Th and ^{232}Th, and *Amanita ponderosa*, *Russula cessans*, and *Macrolepiota procera* for ^{230}Th.

The radium content in mushrooms was 0.089-87 Bq/kg d.w. (Paniagua 1991; Yamamoto *et al*. 1995; Kirchner *et al*. 1998; Baeza *et al*. 2004a). *Terfezia arenaria*, *Lycoperdon perlatum*, and *Russula cessans* presented the highest radium activities. The frequency distribution was observed to be symmetrical and of a Gaussian type. There was no observed dependence of the radium content on the nutritional mechanism of the mushrooms (Baeza *et al*. 2004a).

The ^{210}Pb content in mushrooms was in the range 3.64-58.9 Bq/kg d.w. (Kirchner *et al*. 1998; Malinowska *et al*. 2006). *Boletus edulis*, *Xerocomus badius*, and *Clitocybe nebularis* presented the highest activities. Kirchner *et al*. (1998) found that the ^{210}Pb content in mushrooms was greater than that of ^{226}Ra in the same sample. The ^{210}Pb mainly originated from direct uptake from the soil. Deposition of atmospheric ^{210}Pb onto the fruiting bodies and uptake of ^{222}Rn in solution in the soil pore water which subsequently decayed into ^{210}Pb were only of minor importance. Skwarzec *et al*. (2006) analyzed the ^{210}Po content of mushrooms collected in Poland. The observed range was 2.11-76.47 Bq/kg d.w., with *Boletus edulis*, *Agaricus silvicola*, and *Macrolepiota procera* having the highest activities. But no analysis of ^{210}Pb was made in those samples.

The only detection of ^{7}Be was in two studies in Spain (Paniagua 1991; Baeza *et al*. 2004a). Its origin is cosmogenic and its deposition in soil is related to rainfall (Baeza *et al*. 1996). Its presence in mushrooms, in spite of its relatively short half-life ($T_{½}$ = 53.4 d), is because mushrooms appear mostly in autumn, the season when rainfall is also more frequent and abundant in Spain (Baeza *et al*. 2004a).

The ^{7}Be content was in the range 1.5-70 Bq/kg d.w., and the frequency distribution was of log-normal type. There was observed a dependence on the nutritional mechanism, with the content of saprophytes being higher than that of mycorrhizae.

Comparison of natural and anthropogenic radionuclides

In the previous sections we have analyzed the range of variation of natural and anthropogenic radionuclides worldwide. But it is also very useful to compare the activity levels of these radionuclides within an ecosystem. **Table 2** presents a summary of the ranges of the activities detected. Since the content of natural radionuclides other than ^{40}K has not been extensively studied, we only considered Spanish and Polish samples in which the uranium and thorium contents were also analyzed. The order of accumulation of radionuclides, considering mean and median values, was the following:

a. Spanish mushrooms, as an example of a region mainly affected by global fallout:
$^{40}K > {}^{137}Cs > {}^{228,230,232}Th \approx {}^{234,238}U \approx {}^{226}Ra \approx {}^{90}Sr >> {}^{239+240}Pu > {}^{241}Am$
b. Polish mushrooms, as an example of a region highly contaminated by the Chernobyl accident:
$^{137}Cs > {}^{40}K > {}^{90}Sr >> {}^{230,232}Th \approx {}^{234,238}U \approx {}^{239+240}Pu$

Table 2 Orders of magnitude of the content of anthropogenic and natural radionuclides in mushrooms collected in different countries.

Anthropogenic origin		Natural origin	
Radionuclide	**Range (Bq/kg d.w.)**	**Radionuclide**	**Range (Bq/kg d.w.)**
^{137}Cs	$10^{-1} – 10^{7}$	^{40}K	$10^{2} – 10^{3}$
^{90}Sr	$10^{-1} – 10^{2}$	^{234,235,236,238}U	$10^{-3} – 10^{0}$
^{110m}Ag	$10^{0} – 10^{3}$	^{228,230,232}Th	$10^{-3} – 10^{1}$
$^{238, 239+240}$Pu	$10^{-3} – 10^{1}$	^{226}Ra	$10^{-1} – 10^{1}$
^{241}Am	$10^{-3} – 10^{-2}$	^{210}Pb	$10^{0} – 10^{1}$
^{125}Sb	$10^{3} – 10^{4}$	^{210}Po	$10^{0} – 10^{1}$
^{103}Ru	$10^{1} – 10^{2}$	^{7}Be	$10^{0} – 10^{1}$
^{131}I	$10^{0} – 10^{1}$		

FACTORS AFFECTING THE RADIONUCLIDE CONTENT

The radionuclide content in mushrooms can be influenced by several factors including (i) the date at which radioactive fallout occurred, (ii) its quantity and chemical form, (iii) the location of the layer of soil from which the mycelium takes up nutrients, (iv) the presence of stable elements, and (v) the stage of development of the mushrooms.

Influence of the source term

The predominant source term of radionuclides in a given ecosystem was the most significant factor influencing their content in mushrooms. For the anthropogenic radionuclides, especially radiocaesium, a clear difference is observed between the content in countries strongly affected by Chernobyl fallout (Ukraine, Russia, Sweden, Finland, Poland, etc.) and others in which the predominant source was global fallout (Canada, Spain, Mexico). In the former, the order of magnitude of the radiocaesium content was up to 10^{7} Bq/kg d.w., and in the latter it was up to 10^{2} Bq/kg d.w. This factor also affected the kind of anthropogenic radionuclides detected. In the case of Chernobyl fallout, short half-life radionuclides such as ^{110m}Ag, ^{125}Sb, ^{103}Ru, ^{131}I, were detected, while for the case of global fallout only radionuclides with longer half-lives, such as ^{137}Cs, ^{90}Sr, $^{239+240}$Pu, and ^{241}Am, were detectable.

The time elapsed between the incorporation of anthropogenic radionuclides to the ecosystem and the appearance

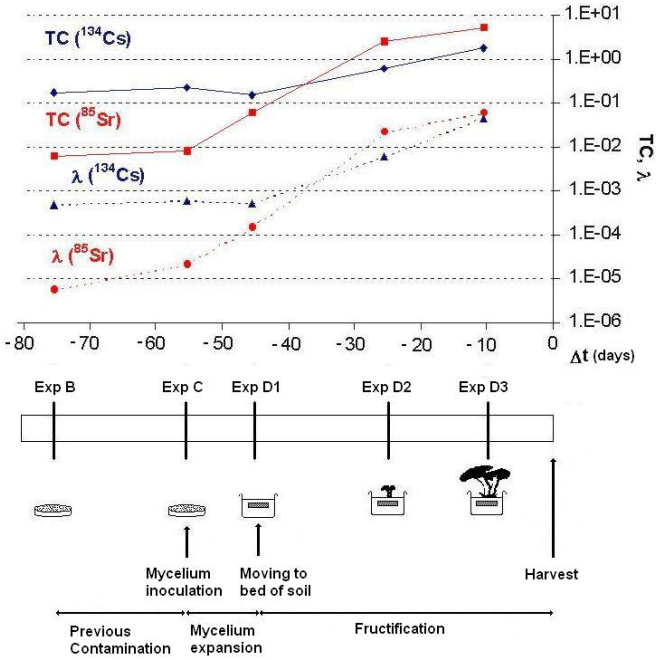

Fig. 2 Time-line of the experiments on *Pleurotus eryngii* culture grown under controlled laboratory conditions, and dependence of the transfer of ^{134}Cs and ^{85}Sr on the time lapse between the incorporation of the radionuclides and harvesting the fruiting bodies. Harvest was taken as time zero. In Exp. B, ^{134}Cs and ^{85}Sr were incorporated into the substrate 20 days before the inoculation with mycelium. In Exp. C, they were incorporated at the time of mycelium inoculation, 55 days before harvest. In Exps. D1, D2, and D3, they were incorporated on the surface soil at the different stages of growth of the fungus shown. Modifed from **Baeza *et al.*** (2002).

of the fruiting bodies also influenced the radionuclide content. This was analyzed in laboratory experiments under controlled conditions for the uptake of ^{134}Cs and ^{85}Sr by fruiting bodies of *Pleurotus eryngii* (Baeza *et al*. 2000, 2002). **Fig. 2** shows the time-line of the inoculation with the ^{134}Cs and ^{85}Sr contaminants at different stages of growth of the fungus. The results of the transfer to the fruiting bodies were evaluated by the transfer coefficients, TC in the figure, and the coefficients λ as calculated from a compartmental model. Both TC and λ were represented as a function of the elapsed time between contamination and harvest. It was found that the longer this elapsed time, the less ^{134}Cs and ^{85}Sr the fruiting bodies accumulated. It was also observed that if the radionuclides were incorporated a few days before harvest the accumulation of ^{85}Sr was greater than ^{134}Cs, probably due to a change in nutritional requirements. This was not observed in field conditions from global fallout or the Chernobyl accident, because the latter occurred in April 1986, several months before autumn. In these studies, together with this elapsed time, different uptake routes were considered – contamination of the mycelium, contamination of the surface soil, and direct deposition onto the fruiting bodies. The most effective incorporation route was the direct deposition onto the fruiting bodies, followed by soil and mycelium contamination for ^{134}Cs and these two in reverse order for ^{85}Sr.

Stage of maturity of fruiting bodies

There are few references in the literature discussing the influence of the stage of maturity on the radionuclide content. Most studies do not report this factor, so that it was supposed that the mushrooms collected were mature. A study under laboratory conditions (Baeza *et al*. 2006c) compared the uptake of ^{134}Cs and ^{85}Sr in young, mature, and old fruiting bodies of *Pleurotus eryngii*. The accumulation of both radionuclides was greatest in mature mushrooms. The

young and old mushrooms presented the same activity levels for ^{85}Sr, but the old mushrooms presented a greater accumulation of ^{134}Cs than the young ones.

Location of the mycelium

The location of the mycelium is a factor that significantly affects the radionuclide content in mushrooms. The radionuclide content of the soil layer from which the mycelium takes up nutrients seemed to be one of the most important factors controlling the content in the fruiting bodies. Wang *et al*. (1998) and Ban-Nai *et al*. (1997) observed that artificially cultivated mushrooms had a lower radiocaesium content than wild mushrooms due to the lower content in the medium used for growth – usually sawdust or rice bran. Yoshida *et al*. (1994a) also found differences between mushrooms with the mycelium growing in different habitats – wood, litter layer, surface soil layer (0-5 cm), and deeper layers of soil (>5 cm).

There are different methods of locating the mycelium in the soil. The first is the mechanical separation of the soil into different fractions such as rhizosphere, the root-soil interphase of fine roots and mycorrhizae, and the bulk soil (Nikolova *et al*. 1997, 2000; Vinichuk *et al*. 2003), but no fraction of pure mycelium has as yet been obtained. The species were identified by DNA determination. A second method is to assume that the ^{134}Cs/^{137}Cs ratio in the fruiting bodies of the fungi and in the layer of soil in which the mycelium is located is the same (Guillite *et al*. 1990; Rühm *et al*. 1997), because the mycelium takes nutrients from this layer of soil. Some remarks are in order about the conceptual aspects of this method. It is useful in a case in which the ecosystem received a recent deposition of ^{134,137}Cs significantly greater than that of the ^{137}Cs already present in the soil, such as occurred in certain regions following the Chernobyl NPP accident. This method could be questioned because it is based on the assumption that ^{134}Cs and ^{137}Cs have the same bioavailability in the soil (Muramatsu *et al*. 1991). And a third method is to estimate the fungal biomass by evaluating some organic compound present in the mycelium. Frequently ergosterol is used. It is the main sterol present in most fungi being a principal component of their cell walls, while it is absent from practically all other life forms, in particular from the roots of plants (Weete 1974). The disadvantage of this method is that it does not allow the species of fungi to be identified, so that it can only give an average of the distribution of mycelium at a certain depth of soil.

Olsen *et al*. (1990) estimated the fungal biomass in a mountain area of Norway to be in the range 12-138 mg fungal d.w./ g soil d.w., which was able to retain about 32% of the radiocaesium present in the soil. Nikolova *et al*. (2000) found the highest concentrations of ^{137}Cs, as well as the highest organic content, to be in the soil-root interface fraction of small roots and mycelium. Vinichuk *et al*. (2003) estimated the fungal biomass in soil to be about 13.8 mg/g soil, and to retain 0.1-50% (mean 15%) of the total soil inventory of ^{137}Cs. They also found a higher concentration of ^{137}Cs in the fruiting bodies than in the mycelium, with ratios in the range 0.1-66 (mean 9.9).

Bioavialability of radionuclides in soil

The bioavailability of a given radionuclide is defined as the fraction which is capable of being transferred. The main problem which arises with this concept is that there is no unique operative definition of the bioavailable fraction. In practice, each researcher uses an *ad hoc* definition that may vary according to the extraction scheme. The most common extractants are ammonium acetate and $MgCl_2$. **Fig. 3** shows, by way of example, the speciation of several natural and anthropogenic (mainly due to global fallout) radionuclides in a pinewood forest ecosystem. From this figure it is possible to deduce that, with respect to the anthropogenic radionuclides, ^{90}Sr was commonly observed to be the most

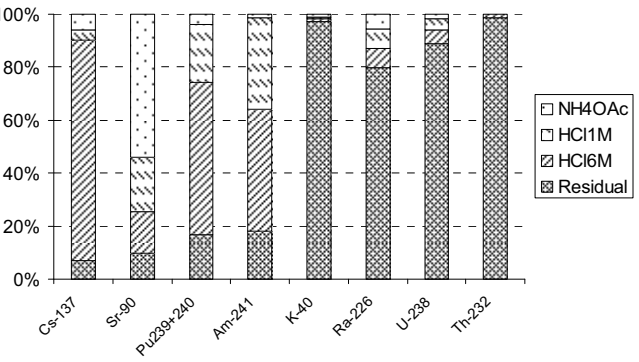

Fig. 3 Association of anthropogenic (^{137}Cs, ^{90}Sr, $^{239+240}$Pu, and ^{241}Am) and natural (^{40}K, ^{226}Ra, ^{238}U, and ^{232}Th) radionuclides with the exchangeable (extracted with NH_4OAc), dilute acid soluble (extracted with HCl 1M), strong acid soluble (extracted with HCl 6M), and residual fractions obtained during the sequential extraction procedure for the surface layer of soil from a Spanish pinewood ecosystem. The source of anthropogenic radionuclides was mainly global fallout. Modified from **Baeza *et al.*** (2005b, 2006a, 2006b).

mobile (Knatko *et al*. 1996; Krouglov *et al*. 1998; Baeza *et al*. 2005). The mobility of ^{137}Cs was lower and depended on the presence of clay in the soil (Ohnuki 1994). Humic substances in the soil, however, could reduce clay's absorption of radiocaesium (Dumat *et al*. 1999). $^{239+240}$Pu and ^{241}Am were more mobile than radiocaesium in a pinewood ecosystem (Baeza *et al*. 2006b). This was probably due to the strong association of these radionuclides to the organic matter in the soil (Fujikawa *et al*. 1999). Natural radionuclides were generally strongly bound to soil particles, because they formed an integral part of them (Baeza *et al*. 2005b, 2006a). ^{226}Ra was the most mobile natural radionuclide, followed by the uranium isotopes. ^{40}K and thorium isotopes were the least mobile radionuclides – more than 90% of their activeties were strongly attached to soil particles or associated with the residual fraction.

Some workers (Heinrih 1992; Gerzabek *et al*. 1998) have reported that a combination of these last two properties – bioavailability of the radionuclides and location of the mycelium – has the consequence that the type of ecosystem in which the fungi are growing affects their radiocaesium activity levels. The order of the incorporation is: Coniferous forest > deciduous leaf forest > prairies.

Relationship with stable elements

The content of different stable cations in mushrooms did not vary over a wide range, indicating that some of them were essential to the fungi's development (Ismail *et al*. 1995). Occasionally, a positive correlation was found between one stable cation and ^{137}Cs (Ismail *et al*. 1995), but this depended on the species of mushrooms considered, and was not general.

The content of alkali and alkaline earth elements in mushrooms was different from that found in plants of the same ecosystem (Yoshida *et al*. 1998). The values were in the following order:

a) Mushrooms: K > Mg > Na > Ca > Rb > Ba > Cs ≈ Sr
b) Plants: Ca > K > Mg > Na > Sr > Ba > Rb > Cs

Compared to plants, the elemental composition of mushrooms was characterized by high Rb and Cs contents and low Ca and Sr contents. Various studies found ^{137}Cs and stable Cs to be correlated (Řanda *et al*. 1990; Ingrao *et al*. 1992; Yoshida *et al*. 1998; Tsukada *et al*. 1998). Yoshida *et al*. (1998) found that the ratio ^{137}Cs/Cs in mushrooms was higher than that in the soil. Subsequent research (Dragović *et al*. 2003; Yoshida *et al*. 2004) found that the ^{137}Cs/Cs ratio was fairly constant for each sampling site, and of the

same order of magnitude as that in the organic layer of soil, suggesting that ^{137}Cs from Chernobyl was well mixed with stable caesium within the biologically connected compartments in the forest ecosystem. There was no correlation between the K and Cs content in mushrooms: the K content seemed to be species independent, while the Cs content varied widely. This suggested that Cs and K play different roles in mushrooms. The behaviour of Rb was intermediate: Rb was correlated with Cs, but the Rb/Cs ratio was not constant, but decreased with increasing Cs content. This suggested that Rb was partially incorporated by the same mechanism as Cs.

Producing the fruiting bodies of mushrooms under controlled laboratory conditions is very difficult. It has only been achieved with a few species, mainly saprophytes such as *Pleurotus eryngii*. Instead, some studies have analyzed the uptake of radionuclides and the influence of different stable elements using hydroponic cultures of the mycelium of various fungi species. Studies with the mycelium of the *Pleurotus* genus found a decrease in the stable Cs and ^{137}Cs content when the Cs content was increased in the culture medium (Kuwahra *et al*. 1998; Terada *et al*. 1998; Baeza *et al*. 2005).

DOSE DUE TO MUSHROOM CONSUMPTION

The consumption of mushrooms was found to make a significant contribution to radiocaesium incorporation in human body (Shutov *et al*. 1996; Skuterud *et al*. 1997). In some countries such as the Czech Republic it was assessed to be the greatest source of radiocaesium uptake in the diet (Horyna 1991). The dose from internal exposure was 0.2-5.35 mSv/y (Shutov *et al*. 1996; Beresford *et al*. 2001; Jacob *et al*. 2001; Dragović *et al*. 2003).

One of the main problems connected with the consumption of mushrooms is that, in zones affected by Chernobyl, the dose due to the consumption of wild and farm produce has fallen drastically since the accident. Since, however, the ^{137}Cs activity levels in mushrooms have remained practically constant (except of course for physical disintegration), their consumption today has a greater relative importance with respect to other routes of ingestion. In a study of the dose levels in different areas of subarctic Russia, Kaduka *et al*. (2005) concluded that, while in 1987 the consumption of mushrooms only contributed 3.6% of the dietary dose, in the period 1991-2004, this had risen to 52%.

In order to reduce the dose from mushroom consumption in countries with a significant Chernobyl contribution, a recommendation was made to prohibit their consumption in zones where the deposition of ^{137}Cs was either greater than 555 kBq/m^2 or greater than 185 kBq/m^2 and the internal dose was above 1 mSv. Indeed, this restriction of mushroom consumption reduced the dose levels by a factor of 3, and was the best action in terms of the cost-benefit ratio (Jacob *et al*. 2001; Shaw *et al*. 2001). However, in countries not highly contaminated by Chernobyl fallout, the annual effective dose due to mushroom ingestion was lower than those mentioned above. It was about 0.077 μSv in Japan (Ban-Nai *et al*. 1997), 0.22-1.2 μSv in Mexico (Gasó *et al*. 2000), 5.42 μSv in Great Britain (Barnett *et al*. 1999), and 4.6 μSv in Spain (Baeza *et al*. 2004a). It was found that in Mexico about 32-37% of the radiocaesium incorporated by diet is due to mushroom consumption (Gasó *et al*. 2000).

The radionuclide content in mushrooms is easily digested by humans. The nutritional quality of cultivated mushrooms contaminated with ^{134}Cs, ^{85}Sr, and ^{239}Pu was tested by the van Soest method (van Soest 1967). It was found that 97-99% of the radioactive contamination could be digested by man (Baeza *et al*. 2004c). Vinichuk *et al*. (2004) reported a significant finding regarding the digestibility of mushrooms – extraction with water at 20°C released 68% (mean value) of ^{137}Cs from ectomycorrhizae, and up to 93% if water at 80°C was used.

Mushrooms are not frequently consumed raw. Mushroom preparation and preservation was found to reduce the radiocæsium content significantly: cleaning and washing 20%, boiling and salting 40-50%, frying or pickling 70% (Kenigsberg *et al*. 1996; Beresford *et al*. 2001; Kalač 2001). Drying was the only mode of preparation that increased the radiocaesium content, due to the loss of water. The form of preparation thus affects radiocaesium intake, and should be taken into account to correctly estimate the internal dose.

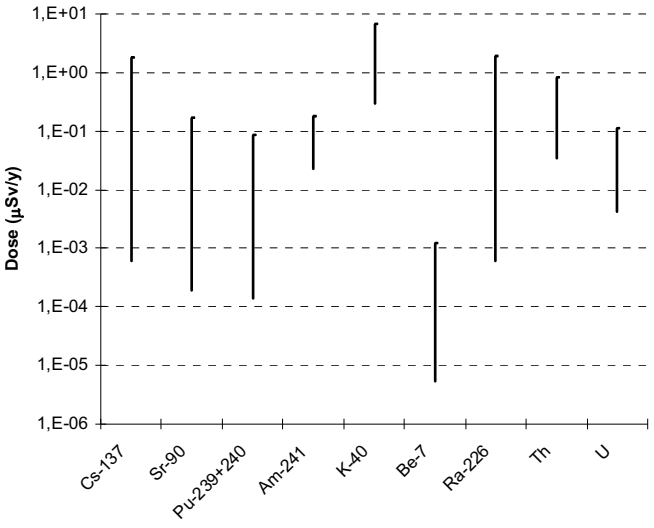

Fig. 4 Range of the dose from mushroom consumption in Spain due to anthropogenic and natural radionuclides.

Other radionuclides were also found to have a relatively significant contribution to the effective dose. In the case of ^{210}Pb, this was estimated at 8.3 μSv/year, similar to that of radiocaesium in the same ecosystem (Kirchner *et al*. 1998). The contribution of natural radionuclides to the dose due to consumption of fungi was especially significant in ecosystems that were not highly contaminated, such as the case of Spain. **Fig. 4** shows the range of the contribution from natural and anthropogenic radionuclides to the dose due to ingestion of mushrooms in this kind of ecosystems. The radiocaesium contribution was greater than that of the other anthropogenic radionuclides considered – ^{90}Sr, $^{239+240}$Pu, and ^{241}Am. Nevertheless, in some cases of low radiocaesium content, the contribution of natural radionuclides – ^{40}K, ^{226}Ra, and ^{228,230,232}Th – may be similar to or even greater than that of ^{137}Cs.

CONCLUSIONS

Mushrooms can bioaccumulate radionuclides in forest ecosystems. Radiocaesium has been the most widely and intensively studied radionuclide, showing a great range of variation of about eight orders of magnitude. The affinity of some species of fungi for radiocaesium was so high than they accumulated it even 40 years after the deposition, as in the case of global fallout. The accumulation or transfer of other radionuclides has been far less studied, especially the cases of alpha emitters. Generally, their activity levels were lower than that of radiocaesium, except in the case of ^{40}K in countries that are not highly contaminated. The activity levels of ^{40}K and radiocaesium in mushrooms were uncorrelated, and their frequency distributions were significantly different – log-normal type for radiocaesium and Gaussian for ^{40}K. These findings were taken to imply that the content of K in the mushrooms was somehow self-controlled. Short-lived radionuclides such as ^{125}Sb or ^{103}Ru were only detected shortly after the Chernobyl accident. Of the anthropogenic radionuclides that we found reported in the literature, $^{239+240}$Pu and ^{241}Am presented the lowest activity levels. With respect to natural radionuclides, their degree of variation was less than that of radiocaesium. In particular, the relatively high activity levels of ^{40}K and thorium and uranium isotopes were surprising given their low bioavailability in

soils.

The radioactive content of mushrooms can be influenced by several factors affecting the growth of fungi. The source term had a great influence. In sites greatly affected by the Chernobyl accident, there was a significant rise in the radiocaesium content, and some short-lived radionuclides were detected. It was observed that when fungi incorporated the radionuclides in the initial stages of their growth, the activity detected in the fruiting bodies was lower than when the incorporation was during the final stages. Mature fruiting bodies had higher contents than young or old ones. The location of the layer of soil from which the mycelium takes it nutrients and the degree of bioavailability of radionuclides in that layer were key factors in determining the soil-to-fungus transfer of radionuclides and their final accumulation in the fruiting bodies. The content of stable elements in that soil layer also affected the radiocaesium content of mushrooms which was observed to correlate with stable Rb and Cs in the sense of decreasing ^{137}Cs content with increasing content of those elements.

Currently, the consumption of mushrooms is the main contribution to the internal dose in highly contaminated areas, mainly because of their high radiocaesium content. In some subarctic areas, there has been observed a steady increase of the importance of the consumption of mushrooms to the dietary intake of radiocaesium, due mainly to a decrease of its content in agricultural produce. In other areas with a lower radionuclide contamination, the contribution of other radionuclides may be similar to or even higher than that of radiocaesium. The radionuclide content in mushrooms was estimated to be readily digestible by man, so that almost all the radionuclides present in mushrooms contributed to the internal dose. Mushrooms' radiocaesium content, and consequently the dose, can be reduced by some methods of preparation such as boiling or frying.

ACKNOWLEDGEMENTS

The present work was partially financed from Spain's Nuclear Safety Board (Consejo de Seguridad Nuclear) under the projects entitled "Study of the transfer of radioactivity to fungi. Interactions and consequences. 1st and 2nd phase", and from ENRESA under the project "Study of the transfer of radioactivity to fungi. Interactions and consequences. 2nd phase". The research carried out in the development of these projects gave the scientific knowledge necessary for the present work.

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