Doctoral Thesis

Radiocesium contamination in Japanese fir (*Abies firma* Sieb. et Zucc.) after the Fukushima Daiichi Nuclear Power Plant accident

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Contents

Abstract

On 11 March 2011, the magnitude 9.0 Great East Japan earthquake and resulting tsunami caused the extensive damage to the coastal areas in the Tohoku region of Japan. As a result of this disaster, the number of recorded deaths rose to 15,893 and those still missing total 2,556.

The breakdown of cooling systems for the reactors at Tokyo Electric Power Company Fukushima Daiichi Nuclear Power Plant (the FDNPP) led to a severe nuclear crisis. The loss of cooling functions caused hydrogen explosions, and a large amount of radioactive material from the damaged reactor buildings was released into the atmosphere. Radioactive material released from the FDNPP have polluted the surrounding terrestrial and aquatic environment.

Approximately 70% of Fukushima Prefecture is covered with forests. A large amount of radionuclides, especially radiocesium (^{137}Cs), was deposited onto the forested areas. To understand the effect on forest wild organisms and for the safety of workers and residents near forests, once radioactive materials are released into the environment, contamination levels of various organisms must be promptly assessed. However, radioactive contamination of forest ecosystems is quite complex, consisting of various ecological factors. Hence, the effects radioactive pollution in Japanese forests may differ from the effects in North European forests caused by the Chernobyl accident. Ecological factors affecting North European forest ecosystems are quite different from those in Japan. Therefore, an understanding of the behavior of radiocesium in Japanese forest ecosystems is required.

Forest trees have a huge biomass above the ground, and they have potential to provide storage of huge amounts of radioactive material and to be a source of radiation emission. After the Chernobyl accident, many studies on tree contamination were conducted and these reported the heterogeneity of distribution of radiocesium in a tree body. Moreover

there are large inter-individual variations in radiocesium contamination of forest trees, often affecting the results of statistical tests. To understand the true effects of ecological factors, we need to consider these large variations when investigating radioactivity contamination of forest trees; otherwise we cannot obtain an accurate assessment of the contaminated environment. Well-designed sampling methods help to avoid these large variations. In this dissertation, to establish an effective sampling method, I propose a new sampling method based on information obtained by my studies on endemic tree species in a Japanese forest ecosystem.

In this dissertation, I focused on the evergreen conifer, Japanese fir (*Abies firma* Sieb. et Zucc.). This species is commonly distributed in natural forests in coastal area of Fukushima Prefecture. *A. firma* has a specific branching pattern that allows us to identify the shoot age. Tree age is an important factor for the radiocesium distribution in tree bodies, and this information helps us to avoid a large variation caused by differences in age. Using this species, I provide information to establish an effective sampling method for the assessment of contaminated trees growing in Japanese forest ecosystems.

In Chapter 2, I show the distribution pattern of ^{137}Cs contamination in a young *A. firma* tree body. The results of this study indicate that visual classification of shoot age is a good working method for the assessment of ^{137}Cs contamination in *A. firma* tree organs. This study clarified the differences in ^{137}Cs contamination between different ages and between different tree organs such as needles, branches, wood and bark. Moreover, the vertical distribution of ^{137}Cs contamination in an individual tree body was also discussed. Both age and height information of trees allow us to distinguish external contamination from internal contamination. In conclusion, the heterogeneity of ^{137}Cs contamination in tree bodies was observed and this heterogeneity was brought about by (1) differences in organs such as needles, branches, wood and bark, (2) differences in age, and (3) position (height) of organs. Thus, standardization of sampling to avoid the effect of intra-individual variation is required for the assessment of radioactivity contamination in forest tree species.

The Chapter 3 presents additional information for highly accurate sampling methods. The branching pattern of *A. firma* is trifurcate. In this branching, we can see clear morphological differences between three shoots (one main shoot and two lateral shoots). I investigated the difference in ^{137}Cs concentration between three different shoots. The ^{137}Cs concentration in main shoots was significantly higher than those in lateral shoots. This indicates that there were differences in ^{137}Cs concentration between shoot type even for the same organs of the same age and at the same position (height). In conclusion, it is important to select samples while paying attention to shoot differentiation. Samples should be taken only from the main shoot or only from the lateral shoot (not a mix of shoots) for every sampled tree.

In Chapters 2 and 3, I discuss the intra-individual variation of ^{137}Cs concentration in *A. firma*. In these chapters, differences in ^{137}Cs concentration caused by different organs, ages and sampling positions when field sampling is conducted are considered important. Next, I tried to clarify the inter-individual variation of ^{137}Cs concentration in this species. A spatial autocorrelation is one of the factors affecting inter-individual variation in ecological data, causing a lack of independence of ecological data. Non-independence of ecological data leads to various statistical problems. Examination of the spatial structure of observations between ecological data is required to clarify the presence of spatial autocorrelation. However, the spatial structure of ^{137}Cs contamination in forest trees is still unclear. In Chapter 4, I investigated the spatial structure of ^{137}Cs concentration of *A. firma* in a natural secondary forest and examined whether or not spatial autocorrelation exists. A significant strong spatial autocorrelation was observed over short distance (<2.5 m), suggesting that spatial distribution needs to be considered in the evaluation of radioactive contamination in trees. Therefore, I strongly recommend taking position data of sampled trees when sampling is being conducted, because we can adjust the non-independence of data due to the spatial autocorrelation with the position data. Furthermore, I mentioned the number of samples required to evaluate radioactive contamination in trees based on the results of spatial analysis.

According to the sample-size analysis, a sample size of seven trees was required to determine the mean contamination level within an error of the mean of no more than 10%. This required sample size may be feasible for various fields.

In Chapter 5, I examined the low contamination levels in *A. firma* seeds. This result, together with the limited root uptake of ^{137}Cs due to a strong fixation of ^{137}Cs to clay minerals, suggests that ^{137}Cs contamination in future generations of this species growing in forests surrounding the FDNPP will be low. However, the risks of chronic radiation exposure for trees have been reported recently. There is still insufficient field data on the consequences of chronic radiation exposure for tree populations. Consequently, studies on the long-term biological consequences of radiation exposure after a nuclear crisis are needed in future.

Chapter 1

General Introduction

The Fukushima Daiichi Nuclear Power Plant accident

On March 11, 2011, a magnitude 9.0 earthquake, subsequently named ‘The 2011 Great East Japan Earthquake’, occurred in the Pacific Ocean near the coastal Tohoku district of Japan with an epicenter at 38°06.2′N, 142°51.6′E and a depth of 24 km (Japan Meteorological Agency). The great tsunami following the earthquake caused extensive damage to the coastal Tohoku district areas. The number of deaths and missing people as a result of the earthquake and tsunami has risen to 15,893 and 2,556, respectively (National Police Agency 2016). The earthquake and tsunami caused the Tokyo Electric Power Company Fukushima Daiichi Nuclear Power Plant (FDNPP) accident, which was the one of the most severe incidents that arose as a result of this event. At the time of the accident, Units 1, 2 and 3 of the FDNPP had been supplying electricity under ordinary commercial operations. After the earthquake occurred, the reactors of Units 1, 2 and 3 were shut down and cooling functions were operated immediately. However, the FDNPP was hit by the tsunami 50 minutes after the earthquake and the cooling functions of the Unit 1, 2 and 3 reactors were lost. The loss of the cooling functions caused water depletion of the pressure containers, and increased the temperature of the fuel rods. In addition, hydrogen gas was generated, and consequently the reactor buildings of Units 1, 2 and 3 were destroyed in a hydrogen explosion. This damage to the reactor buildings caused the release of a large amount of fission products into the environment. Furthermore, the venting systems of Units 1 and 3 released nuclear fission products into the atmosphere (Tokyo Electric Power Company).

Emission radionuclides from the Fukushima Daiichi Nuclear Power Plant

In general, several kinds of radionuclides are released from nuclear power plant accidents, which was also the case for the FDNPP accident. Many papers have reported the estimation of the total and different activity of the emitted radionuclides. According to Steinhauser et al. (2014), the total activity of radionuclides released was 520 PBq (1 PBq = 10^{15} Bq), with 340 and 800 PBq as lower and upper bounds, respectively. Radionuclides detected after the FDNPP accident were the noble gases krypton-85 and xenon-133, the volatile elements tellurium-129m/132, iodone-129/131/133 and cesium-134/136/137, and strontium-89/90 and plutonium-238/239+240/241 as elements with intermediate volatility (Chino et al. 2011; Stohl et al. 2012; Zheng et al. 2012; Ahlswede et al. 2013; Hou et al. 2013). It is noteworthy that two kinds of radionuclides were extensively released: iodine-131 and cesium-137 (^{137}Cs). Iodine-131 is known for the risk of health effects such as thyroid cancer. The long-lived environmental contaminant ^{137}Cs also represents a health risk following external and internal exposure. The estimated amount of iodine-131 and ^{137}Cs released from the shattered plants into the atmosphere were approximately 100–500 PBq and 6–20 PBq, respectively (UNSCEAR 2014). Morino et al. (2011) reported that approximately 87% of iodine-131 and 78% of ^{137}Cs was deposited into the ocean, and the rest was deposited over terrestrial Japan. According to the results of airborne surveys and soil sampling, the northwest of the FDNPP and the Naka-dori region of Fukushima Prefecture were most highly contaminated with radionuclides such as ^{137}Cs, cesium-134 (^{134}Cs), and iodine-131 (Yoshida and Takahashi 2012).

Cesium-137 in plants

Compared with iodine-131, ^{137}Cs is a more important element when a long-term environmental pollution of radioactivity is considered; ^{137}Cs has a longer half-life that is about 30.1 years (cf. iodine-131: 8 days). Cesium is an alkaline trace element and it is not necessary for plant growth (Kabata-Pendias 2011). However, cesium is transferred in ecosystems with potassium, which is an essential element for plants, because cesium acts like potassium due to a chemical analog (Davis 1963). This phenomenon can also explain the long duration of terrestrial environmental cesium pollution. For example, if long-lived organisms such as tree species keep cesium in their bodies for a long period, they can be a source of exposure. For the assessment of radiological dose effects on organisms including humans, understanding of the contamination level of forest ecosystems and their inhabiting tree species is important.

Species studied: Japanese fir (*Abies firma* Sieb. et Zucc.)

Japanese fir belongs to the family Pinaceae, and there are about 40 species in genus *Abies* in the Northern Hemisphere (Kenzo 2009). *A. firma* is an endemic species of Japan and a common coniferous species in forests from a cool-temperate zone to a warm-temperate zone in Japan. Tall trees of this species make up the canopy layer of the forests at about 20 m from the ground. The forests near the coastal area of Fukushima Prefecture have some abundant *A. firma* seedlings with fewer numbers of big trees. The currently observable small seedlings of *A. firma* are likely to become canopy trees in future by regeneration.

In the intermediate-temperate forests in the southern Tohoku district, *A. firma* trees are commonly found in forest ecotones from evergreen broadleaf forests to beech forests and from a mixed-forest with trees in *Quercus serrata* and hornbeam (trees in *Carpinus* species). They often form Momi-Tsuga forests with another coniferous tree species, *Tsuga sieboldii*. In general, *A. firma* appears in a late stage of forest succession (Kaji 1975).

A. firma has a high shade tolerance; seedlings and saplings are abundant under the shaded closed canopies. Trees in this species are mainly distributed in the sites with humid and nutrient rich soils such as valleys. Although we can find *A. firma* trees under the closed canopies, their growth is retarded. However, they grow well in canopy gaps and/or forest edge where the trees receive a larger amount of sunlight. Rapid growth of this species under such conditions results in a beautiful cone-shaped tree form.

Natural forests dominated by *A. firma* (rather than artificially planted forests) were converted into Sugi-Hinoki plantation forests by the development of forestry after World War II. Furthermore, many *A. firma* trees were killed by acid rain and/or air pollution (Suzuki 1992). This makes this species rare around urban areas. The hindrance of regeneration of this species caused by feeding damage of Japanese deer was also reported (Ogawa et al. 2006).

In this study, I focused on *A. firma* trees for the assessment of contamination levels by ^{137}Cs in naturally regenerating forest (hereafter referred to as 'natural forest'). The use of this species for the assessment of contamination levels by ^{137}Cs will bring two benefits. The first is the abundance and a wide distribution of this species in natural forests in Fukushima Prefecture (the ubiquity of this species). This species may be present in natural secondary forests located in coastal areas in Fukushima. The ubiquity of this species allows us to assess the contamination levels by ^{137}Cs in many natural forests (= a wide site application). The second benefit is related to its branching pattern. This species has a trifurcation branching (Fig. 2-1). The terminal (current-year) shoots of this species expand from winter buds which are made at the top of the previous-year shoots. Typically, the winter buds at the top of a previous year's shoot develop into one main shoot and two lateral shoots (trifurcation) in summer. We can distinguish the age of the shoots by tracing back the trifurcation branching modules.

The age of tree parts is important for the assessment of ^{137}Cs contamination levels because the contamination level of tree parts should be strongly affected by the age of the tree parts. For example, older parts that expanded before the FDNPP accident might have been more heavily contaminated than younger parts that expanded after the accident because of the direct external contamination. Furthermore, previous studies commonly show that the contamination level of needle leaves of some coniferous tree species changed with the age of the needles because of the internal translocation of ^{137}Cs (Fogh and Andersson 2001; Yoshihara et al. 2014). My preliminary survey shows the same phenomenon in *A. firma* (radioactivity in new shoots was higher than old shoots) (Fig. 1). Therefore, age information of trees is of paramount importance for the assessment of ^{137}Cs contamination levels and it can easily be determined from branching patterns for *A. firma*.

This beneficial aspect of *A. firma* is clear when we compare this species with other tree species. Deciduous tree species such as *Q. serrata* only have their current-year leaves. Another evergreen conifer species (e.g. *T. sieboldii* and *Cryptomeria japonica*) is found in Fukushima forests, but these species usually have a complex branching pattern rather than the simple trifurcation found in *A. firma*. The complex branching structure makes it impossible for us to trace back the shoot ages. One might expect that a shoot produces an annual ring and we can identify the shoot age by counting the number of annual rings. However, many tree species produce only faint annual rings when growing in shaded conditions (Kohyama 1980). Therefore, the determination of shoot age by counting annual rings is not feasible for the assessment of contaminated trees. In addition, clear morphological differentiation between different tree organs (e.g. needle-leaves vs branches) of *A. firma* is also useful to improve our understanding of ^{137}Cs accumulation in tree bodies. Therefore, these species characteristics are beneficial to enhance our understanding of the behavior of ^{137}Cs in tree.

Objectives of the study

After the Chernobyl accident in what is now Ukraine in 1986, forest contamination was widespread, and the behavior of ^{137}Cs in trees was investigated in various forests. These studies indicated that the environmental factors (e.g. climates, geography) have a great impact on the ^{137}Cs contamination of trees. The scientific knowledge obtained in previous studies is useful to help us understand the ^{137}Cs dynamics in Japanese forest ecosystems; however, it remains unclear whether the scientific knowledge can be applied to Japanese forests. Therefore, clarification of the ^{137}Cs dynamics in Japanese forest ecosystems is needed to determine whether the forests contaminated by the FDNPP accident were affected in the same way as forests contaminated by the Chernobyl accident.

As a first step toward this verification, effective methods should be established for an assessment of ^{137}Cs behavior in contaminated forests. Ecological data obtained using unpracticed methods are inaccurate and unsuited to assess the environmental pollution. In this study, therefore, I provide information to establish effective methods, especially sampling, for the understanding of ^{137}Cs behavior in contaminated forests in Japan. The aims of this dissertation were as follows:

1. To clarify the contamination levels of ^{137}Cs in *A. firma* growing in natural secondary forests in Fukushima.
2. To provide information about the sampling method of contaminated trees in Japanese forest ecosystems.
3. To predict the long-term effects of pollution on Japanese forest ecosystems.

Study sites

To achieve the above-mentioned aims, I conducted field surveys in natural secondary forests predominated by *A. firma* in Fukushima prefecture. Two study forests were selected: Soma (Chapters 2 to 4, described below) and Iitate (Chapter 5).

The study forest in Soma is in a deciduous broadleaf (*Quercus crispula* and *Cornus* *macrophylla*) and evergreen conifer (*Pinus densiflora* and *A. firma*) mixed forest (N37°45′41, E140°47′52) (Fig. 3-1). No bare land was located around the study forest and the soil type was brown forest soil. The forest was located approximately 45 km northwest of the FDNPP. The initial deposition of ^{137}Cs around the study site was 100–300 kBq m^{-2} according to the third airborne monitoring survey conducted from 31 May to 2 July in 2011 (MEXT 2011). The mean annual precipitation was 1361.6 mm between 1981 and 2010 at the nearest meteorological recoded station (approximately 12 km southwest of the study site). The Soma study forest is not within the evacuation zone defined after the FDNPP accident and thus people live in areas surrounding the study forest. Because the Soma study forest is located a few km from the nearest human habitation, few people visit the forest. Decontamination activities for radionuclides have not been conducted in this area. I chose this forest as a study site because it is a typical natural secondary forest predominated by *A. firma* in Fukushima prefecture and is located near human habitation but with limited human activities and disturbance.

The Iitate study forest area included a mixed deciduous and coniferous secondary forest. The study forest was located approximately 45 km northwest from the FDNPP (N37°44′05 and E140°49′35) (Fig. 5-1). The initial deposition of ^{137}Cs around the study site was 100–300 kBq m^{-2} according to the third airborne monitoring survey described above (MEXT 2011). The mean annual precipitation was 1361.6 mm between 1981 and 2010 at the nearest meteorological recorded station (approximately 10 km southwest of the study site). The Iitate study forest was in the planned evacuation zone, and the designation of the planned evacuation zone was removed in March 31, 2017. Therefore, people have not lived near the study forest in the last 6 years following the FDNPP accident. Decontamination activities for radionuclides in and around the Iitate study forest were moderately high. I chose this forest as a study site because it is a typical natural secondary forest predominated by *A. firma* in Fukushima prefecture located close to human habitation.

Outline of the dissertation

Chapter 2 describes the distribution of radioactive contamination of the whole aboveground tree components of *A. firma*. Using the ecological and morphological characteristics of this species, this chapter discusses the tendency of ^{137}Cs accumulation in a tree body and the differences in ^{137}Cs between different tree components.

Chapter 3 focused on the relationships between ^{137}Cs accumulation and the growth rate of shoots of *A. firma*. This chapter deals with the question of whether the growth rate affects the contamination level of ^{137}Cs in *A. firma* needles.

Chapter 4 addresses the question of the presence of spatial autocorrelation between radioactive contamination of forest trees growing in a natural secondary forest. This chapter provides information about the sampling strategy for the assessment of tree contamination in forest ecosystems.

Chapter 5 determines the contamination levels of diaspores of *A. firma* approximately 4.5 years after the FDNPP accident. Through the assessment of diaspores of this species, the aim of this chapter was to predict the contamination level in future generations growing in natural forests surrounding the FDNPP.

Chapter 6 summarizes the achievements of the study. I describe sampling methods for the assessment of ^{137}Cs contamination levels by using *A. firma* trees. I also discuss the application of the study for the assessment of ^{137}Cs contamination levels over forests in Fukushima Prefecture and the required future research for forests and people in Fukushima Prefecture.

Chapter 2

Accumulation of ^{137}Cs in different parts of Japanese fir trees after the Fukushima Daiichi Nuclear Power Plant accident

Abstract

The aim of this study was to understand the distribution of ^{137}Cs in Japanese fir (*Abies firma* Sieb. et Zucc.) trees growing in a natural secondary forest in Fukushima, Japan. The concentrations of ^{137}Cs in different parts of the trees (needles, branches, wood, and bark) were determined with special reference to the height and age of the tree parts. There were large differences in the ^{137}Cs concentrations among different tree parts, different heights, and tissues of different ages. In general, the current-year parts and older parts that existed at the time of the accident (> 6 years old) were more heavily contaminated than the other parts. On the basis of these results, I suggest that the organ type, age, and height should be considered when collecting samples to assess the ^{137}Cs levels in trees. This information will be useful for obtaining reliable measurements of the ^{137}Cs concentration in trees and its changes over time for the long-term monitoring of radioactive contamination in this area.

Key words

Abies firma **Sieb. et Zucc.; tree organs; age; height; stratified-clipping method; biomass profile**

Introduction

Many types of forests were contaminated with radionuclides released from the Fukushima Daiichi Nuclear Power Plant (FDNPP) accident in March 2011. After radionuclides are deposited in forests, they are circulated through forest material cycles and the food web, and very little is discharged from the forest ecosystem. Since these contaminants will persist in the environment for a long time, it is important to have reliable methods for the long-term monitoring of radionuclide contamination in forests.

One way to monitor the contamination levels of forest trees is to sample tree parts and measure the concentration of ^{137}Cs. Several studies have shown that the distribution of ^{137}Cs differs among various tree parts such as leaves, branches, and wood (Mamikhin et al. 1997; Strebl et al. 1999; Komatsu et al. 2016). In addition, the concentration of ^{137}Cs in the same tree part can vary among different heights and/or of different ages (Fogh and Andersson 2001; Yoshihara et al. 2014). Thus, the ^{137}Cs concentration determined for collected samples may differ depending on the organ sampled, its age, and its height, even within the same individual tree. This makes it difficult to monitor ^{137}Cs contamination reliably in forest ecosystems. Consequently, it is important to develop a standardized sampling method for the reliable measurement of ^{137}Cs concentrations. To develop such a method, we should determine which part of the tree is most suitable for measuring the ^{137}Cs concentration. Therefore, it is important to determine the distribution of ^{137}Cs within the tree body to design a standardized sampling method for use in continuous forest monitoring.

In this study, I focused on Japanese fir (*Abies firma* Sieb. et Zucc.), a dominant canopy tree species in natural Fukushima forests. The morphological characteristics of this tree species (simple trifurcation branching) allowed us to determine shoot age easily (Fig. 2-1). I investigated the distribution of ^{137}Cs in the whole *A. firma* tree body to clarify is accumulation characteristics in above-ground tree parts (needles, branches, wood, and bark), with special reference to the height and age of each organ. The results of this study provide essential information to develop a standardized sampling procedure for the reliable long-term monitoring of ^{137}Cs contamination in a Japanese forest ecosystem.

Materials and Methods

Study sites

The study was conducted in a natural secondary forest in Soma, Fukushima Prefecture approximately 45 km northwest of the FDNPP. The initial deposition of ^{137}Cs around the study site was 100－300 kBq m^{-2}, as determined in the third airborne monitoring survey conducted from 31 May to 2 July in 2011 (MEXT 2011). Deciduous broadleaf trees (*Quercus crispula*, *Quercus serrata*, and *Magnolia obovata*) and evergreen coniferous trees (*Pinus densiflora* and *A. firma*) are dominant in this natural secondary forest.

Sampling and processing

I selected two young *A. firma* trees; one was 265-cm high (S1) and the other was 263-cm high (S2). Each tree was cut at the base of the stem (5 cm above the ground). In this study, tree parts (needles, branches, wood, and bark) were grouped according to their height on the tree and their age. After cutting, the stem including branches was cut into 1-m sections, measured from the ground. The branches were sawn off and grouped into seven age groups (current-year, 1-year-, 2-year-, 3-year-, 4-year-, 5-year-, and > 6-year-old branches). The stem was cut into sections of different ages, as determined by morphological characteristics. In the laboratory, the dry mass of all samples (needles, branches, wood, and bark) was measured after oven-drying at 65°C for 72 h.

Sample treatment and ^{137}Cs analysis

Samples were oven-dried at 65°C for 72 h and then pulverized with a mill (Tube Mill control, IKA Japan K.K., Osaka, Japan). The milled samples stored in plastic containers (U-9 container, 50 ml). The ^{137}Cs concentration in samples was measured using a low background germanium detector (GEM30-70, Seiko EG & G, Tokyo, Japan). The detection efficiency of gamma-rays has been reported elsewhere (Shizuma et al. 2016). The measurement times were 10,000 s for needle, branch, and bark samples and 80,000 s for wood samples. The average uncertainty of measurement was 9%. The ^{137}Cs concentration was converted to that on the sampling day based on the physical decay rate.

Results and Discussion

Difference in biomass of tree parts (needles, branches, wood, and bark) with respect to height

The dry mass of each part (needles, branches, wood, and bark) of the sampled *A. firma* trees is shown in Figure 2-2. In this study, both sampled trees showed a similar trend of biomass allocation among the tree parts. The dry mass of the middle part (height, 100–200 cm) was greater than those of the lower and upper parts. The mass of needles and branches was also higher in the middle part than in the upper and lower parts. In contrast, the dry mass of wood and bark gradually decreased from the lower to the upper parts of the trees. The highest dry mass of wood and bark was in the bottom height class (0–100 cm in height). The biomass distribution patterns in the needles and branches may result from the death of older and lower branches due the darker conditions in the lower parts. The distribution pattern in the stem and bark may reflect the continuous accumulation of biomass in the stem.

Difference in concentrations of ^{137}Cs among tree parts (needles, branches, wood, and bark) with respect to height and age

The concentration of ^{137}Cs differed substantially among the needles, branches, and stems of both sample trees (Fig. 2-3), similar to the results reported by Mamikhin et al. (1997), Strebl et al. (1999), and Komatsu et al. (2016). The patterns of ^{137}Cs distribution among the tree parts were similar in the two sample trees (Fig. 2-3).

The highest ^{137}Cs concentrations in needles were almost the same among the three height classes. Among age groups younger than 6 years (i.e., the needles that grew after the accident), the ^{137}Cs concentration was highest in the current-year needles, regardless of the height class, suggesting that ^{137}Cs is translocated from old needles to the newest needles. As a result of this process, the current-year needles (the newest needles) may be the most heavily contaminated. Interestingly, the concentrations of ^{137}Cs were almost equal among 1- to 5-year-old needles. This trend was observed for both trees and across all height classes.

Comparing the average ^{137}Cs concentration in current-year needles among different height classes, the concentration was greater in the higher height class (200 – 300 cm) than in the lower height class (0 – 100 cm) (Fig. 2-3).

The ^{137}Cs concentration was higher in >6-year-old needles (i.e., those that grew before the accident) than in younger needles, apart from the current year needles. This result strongly suggested that needles more than 6 years old were polluted by the initial fall out of ^{137}Cs shortly after the accident, because they existed at that time. The pattern of ^{137}Cs distribution in branches was similar to that in needles (Fig. 2-3).

The highest ^{137}Cs concentration in bark was in the samples that were more than 6 years old, probably because of contamination by the initial fall out of ^{137}Cs shortly after the accident. The current-year bark also showed a high ^{137}Cs concentration, suggesting that ^{137}Cs is transported from old parts to new parts of the bark, like in needles.

Wood had the lowest concentrations of ^{137}Cs (Fig. 2-3, note different scales on horizontal axes). The youngest wood had the highest ^{137}Cs concentration in both sample trees. The ^{137}Cs concentration in older wood was 4 – 20 times that in the youngest wood.

Figure 2-4 shows the total ^{137}Cs per tree, calculated by multiplying the ^{137}Cs concentration in each part by the biomass of each part. In both trees, wood had the lowest total ^{137}Cs among the four tree parts. The total ^{137}Cs amount in the other tree parts (needles, bark, and branches) was 10 – 40 times higher than that in wood.

Comparing the different height classes, the total ^{137}Cs was lowest in the highest height class for both trees (200–265 cm) because this height class was smaller than the other two (see Fig. 2-2). However, the distribution of total ^{137}Cs between the middle and bottom height classes differed between the two sample trees. Tree S1 had higher total ^{137}Cs in the middle height class than in the bottom height class, regardless of the tree part. Tree S2 had higher total ^{137}Cs in the bottom height class than in the middle height class, regardless of the tree part. This reflected differences in dry mass distribution, because the total ^{137}Cs concentration did not differ significantly between the two sample trees (Figs. 2-2 and 2-3).

The concentration of ^{137}Cs differed markedly among the tree parts, and among tree parts of different ages and at different heights. Therefore, when collecting samples to measure ^{137}Cs concentrations, researchers should consider the height, age, and type of sample to ensure reliable estimates of the ^{137}Cs concentration. On the basis of the results, I suggest that researchers collect the newest needle from the same height on each tree to measure ^{137}Cs concentrations, especially for the long-term monitoring of ^{137}Cs contamination in forests.

Chapter 3

Difference in ^{137}Cs in needles between main and lateral shoots of Japanese fir growing in natural forests 5 years after the Fukushima accident

Abstract

Japanese fir (*Abies firma* Sieb. et Zucc.) usually shows a trifurcated branching form, differentiated into main and lateral shoots. I compared the radiocesium (^{137}Cs) concentration in needles between the terminal main shoot and two terminal lateral shoots of Japanese fir in order to examine whether the shoot property (main or lateral) affected ^{137}Cs concentration in needles. The ^{137}Cs in needles of main shoot was significantly higher than that of the two lateral shoots, suggesting that differences in shoot properties are a factor affecting the ^{137}Cs in needles. To examine the relationships between ^{137}Cs in needles and shoot morphological traits, the dry mass of needles and branches and the length of branches were measured. The main shoot had a statistically significant heavier needle and branch and longer branch than lateral shoots. However, the ^{137}Cs in needles was not significantly correlated with the morphological values of shoots. The results suggest that the difference in ^{137}Cs in needles between main and lateral shoots cannot be attributed to the difference in shoot morphology (dry weight and branch length). I recommend that when sampling needles for the evaluation of contamination of ^{137}Cs, the shoot properties should be considered to eliminate ^{137}Cs variation arising from these traits.

Key words

The Fukushima Daiichi Nuclear Power Plant; *Abies firma*; shoot position; shoot morphological differences; intra-individual variation

Introduction

The Fukushima nuclear crisis in March 2011 had a severe effect on the environment in terms of releasing a massive amount of radioactive material. Approximately 70% of the area contaminated by the Fukushima Daiichi Nuclear Power Plant (FDNPP) accident is covered by forests (Hashimoto et al. 2012).

Previous studies have reported that there is a heterogeneous distribution of radiocesium (^{137}Cs) within an individual tree depending on the tree parts such as leaves, branches, wood and roots (Mamikhin et al. 1997; Strebl et al. 1999; Komatsu et al. 2016). In addition, the ^{137}Cs concentrations differ within the same tree parts. Following the Chernobyl and Fukushima accidents, in evergreen coniferous species differences in ^{137}Cs concentration in needles were found between the upper and lower foliage and/or between new and old needles were observed (Fogh and Andersson 2001; Yoshihara et al. 2014). One explanation for these phenomena may be different of nutrient requirements of needles (Yoshihara et al. 2014). However, the detailed mechanisms for the translocation of ^{137}Cs in needles within a tree is not clear, and there is a possibility that other factors are affecting the process.

Generally, in Japanese fir (*Abies firma* Sieb. et Zucc.) terminal (current-year) shoots expand from winter buds formed on the top of the previous year's shoots every spring. In well-grown individuals, winter buds consist of one terminal bud and two or more terminal lateral buds. These become the terminal main shoot and terminal lateral shoots with growth, showing a trifurcate branching pattern. Usually these three terminal shoots elongate under the nearly same conditions (same age and same light environment). Therefore, a comparison between the three terminal shoots allows us to eliminate the effect of differences in shoot age and in light conditions such as between the upper and lower foliage of the tree.

In this study, I investigated the ^{137}Cs concentration in needles between the terminal main shoot and two terminal lateral shoots of *A. firma* after eliminating the variation caused by the differences in shoot age and light environment among shoots in an individual tree in order to examine whether the shoot property (main or lateral) affected the ^{137}Cs concentration in needles. First, I compared the ^{137}Cs in needles and the morphological values such as (1) dry weight of needles, (2) dry weight of branches and (3) length of branches between three terminal shoots. Second, I tested the relationships between ^{137}Cs in the needles of three terminal shoots and three morphological values to clarify the effect of morphological traits on the contamination levels of needles.

Materials and Methods

Study sites

The study site was located in a mixed deciduous broadleaf and evergreen coniferous forest in Soma, Fukushima Prefecture (37°45'41 N, 140°47'52 E) approximately 45 km northwest of the FDNPP (Fig. 3-1). The mean annual precipitation was 1361.1 mm from 1981 to 2010 and the mean annual temperature was 10.0°C from 1981 to 2010, recorded by the nearest meteorological observation point (approximately 12 km southwest of the study site) (Japan Meteorological Agency 2016). The initial deposition of ^{137}Cs around the study site was 100–300 kBq m^{-2} reported by the results of the third airborne monitoring survey conducted from 31 May to 2 July 2011 (MEXT 2011). The results from my preliminary field survey showed that the tree density was 2825 trees ha^{-1} (over 5 cm DBH) and the basal area was 45.5 m^2 ha^{-1} (plot size: 20 m × 20 m). The dominant tree species in this area were *Quercus crispula* (32.7%) and *A. firma* (12.8%).

Sampling strategy

A. firma is a common large evergreen conifer species in Fukushima forests. This species is shade-tolerant and saplings are abundant in shaded understory. In August 2016, shoot samples of *A. firma* were collected from 14 young trees of 100 to 200 cm in height growing on flat topography. Previous studies reported a large intra-individual variability of ^{137}Cs concentrations in an individual tree in some tree species. The contamination levels in needles of individual evergreen conifers showed a large variation between shoots depending on the shoot age (Sombré et al. 1994) and the height at which the shoot was located (Yoshihara et al. 2014). Therefore, in this chapter shoot samples were standardized to minimize the intra-individual variability of ^{137}Cs concentrations between shoots. I sampled three terminal (current-year) shoots on the lateral shoot of the previous-year terminal trunk from each tree (Fig. 3-2). The three sampled terminal shoots were divided into three groups (see Fig. 3-2: terminal main shoot, terminal lateral shoot 1 and 2). After grouping, the branch lengths of each shoot were measured.

^{137}Cs measurement

Shoot samples were oven-dried at 65°C for 72 h and separated into ‘needle samples’ and ‘branch samples’. The respective dry weight of each sample was measured. Needles were pulverized using a powdering machine (Tube Mill control, IKA Japan K.K., Osaka, Japan) and were packed into a plastic container (U-9 container, 50 ml). The concentration of ^{137}Cs in needles was determined with a low background germanium detector (GEM30-70, Seiko EG & G Co., Ltd., Tokyo, Japan). The measuring time was 10,000 s for each sample. The detection efficiency for gamma-rays is reported separately (Shizuma et al. 2016). The measurement uncertainty averaged less than 8% for all samples. Based on the rate of physical decay, the concentration of ^{137}Cs in needles was converted to that on the sampling date.

Statistical analysis

The ^{137}Cs concentrations in the needles of each shoot were transformed to a natural logarithm to fit the criteria of normal distribution (Kolmogorov-Smirnov Test, $p = 0.2$). The data for the dry weight of needles and branches and the length of branches obtained from all three shoots showed a normal distribution (Kolmogorov-Smirnov Test, $p = 0.2$).

I performed a one-way repeated measure analysis of variance (ANOVA) with Bonferroni correction in order to test for significant differences in ^{137}Cs in needles and the morphological values (dry weight of needles and branches, length of branches) between the terminal main shoot and the two terminal lateral shoots.

Previous studies reported a large inter-individual variability of ^{137}Cs in the same tree species (Ertel and Ziegler 1991). From my results of ^{137}Cs measurement of needles, the maximum value was about 16 times higher than the minimum value between different individuals. Commonly, a large variation among ecological data masks the effect being examined by statistical tests and decreases the power of statistical tests (Cohen 1988). Thus I tried to eliminate large inter-individual differences in ^{137}Cs concentration in needles among individuals as follows. First, the average ^{137}Cs concentration in the needles of each individual tree was quantified as,

$$the\ average\ {}^{137}Cs\ in\ needles\ of\ each\ tree = \frac{{}^{137}Cs\ in\ needles\ of\ main\ shoot + \left(\frac{{}^{137}Cs\ in\ needles\ of\ lateral\ shoot\ 1 + lateral\ shoot\ 2}{2}\right)}{2} \cdots (1)$$

Then the standardized ^{137}Cs concentration of each shoot was defined as,

$$\text{the standardized } {}^{137}Cs \text{ of each shoot} = \text{observed } {}^{137}Cs\ in\ needles\ of\ each\ shoot - (1) \cdots (2)$$

Using the values of Equation (2), I calculated Pearson's correlation coefficients between the standardized ^{137}Cs in the needles in each shoot and the morphological values (dry weight of needles and branches, length of branches). I also calculated Pearson's correlation coefficients between the morphological values. All statistical analyses were performed using SPSS version 21.0 (IBM SPSS Statistics 21.0 for Windows).

Results

The ^{137}Cs concentration in the needles of the terminal main shoot was significantly higher than that of the two terminal lateral shoots (one-way repeated measure ANOVA, $F = 16.486$, $n = 14$, df = 1.334, $p < 0.001$, Fig. 3-3a). However, there was no significant difference in the ^{137}Cs concentration in the needles between the two terminal lateral shoots (one-way repeated measure ANOVA using Bonferroni correction, $p = 1$, Fig. 3-3a). Comparisons of the dry weight of needles and branches, and the length of branches between the three shoot groups showed that the terminal main shoot had significantly higher values than those of the two terminal lateral shoots (one-way repeated measure ANOVA, needle-DW: $F = 41.558$, $n = 14$, df = 2, $p < 0.001$, branch-DW: $F = 171.403$, $n = 14$, df = 1.104, $p < 0.001$, branch length: $F = 71.703$, $n = 14$, df = 2, $p < 0.001$, Figs 3-3b, c, d). On the other hand, there was no significant difference in the morphological values between the two terminal lateral shoots (one-way repeated measure ANOVA using Bonferroni correction, needle-DW: $p = 1$, branch-DW: $p = 0.915$, branch length: $p = 1$, Figs 3-3b, c, d).

The relationships between the ^{137}Cs in the needles of each shoot and the morphological values of shoots were not significant (Pearson's correlation coefficient, Fig. 3-4). On the other hand, the relationships between the morphological values of shoots were all positively correlated with each other (Table 3-1).

Discussion

I found a significant difference in ^{137}Cs concentration in the needles between the terminal main shoot and the two terminal lateral shoots, although there were no differences in shoot age and light environment such as different shoot positions on a tree. The distribution of ^{137}Cs in an individual tree was heterogeneous even within the same tree parts. Several studies have reported that the radial distribution of ^{137}Cs in the stems of Scots pine contaminated by the Chernobyl fallout was different between sapwood and heartwood (Thiry et al. 2002; Soukhova et al. 2003). These studies indicated that the difference in the radial distribution of ^{137}Cs in the stem was due to the different morphological and functional compositions of sapwood and heartwood. A difference in ^{137}Cs concentration among foliar parts was observed also in some conifer species after the Chernobyl and Fukushima accidents (Raitio and Rantavaara 1994; Sombré et al. 1994; Fogh and Andersson 2001; Yoshihara et al. 2014). A high ^{137}Cs concentration in new needles compared with old needles was reported in these previous studies, and Yoshihara et al. (2014) concluded that the relocation of specific nutrients between foliage in a tree followed by a flush of new foliage may result in new foliage having a high ^{137}Cs concentration. This suggested that morphological and physiological differentiation among the same tree parts were important factors affecting the ^{137}Cs distribution in trees. In this study, the ^{137}Cs in the needles of the main shoot was significantly higher than that of the two lateral shoots, indicating that the differences in shoot properties (main shoots or lateral shoots) were a factor affecting the ^{137}Cs in needles (Fig. 3-3a). I also found significant differences in the morphological values between the main shoot and two lateral shoots and similar trends in the results of ^{137}Cs in needles was found (Figs 3-3b, c, d). However, the standardized ^{137}Cs in needles was not significantly correlated with both the dry weight of needles and branches and the length of branches (Fig. 3-4). These results may be explained by the two following reasons. One is that the morphological values of shoots (dry weight of needles and branches, branch length) may not be the predominant factors causing a difference in ^{137}Cs in needles between the main and lateral shoots. The other is that I could not totally exclude inter-individual variation in ^{137}Cs in needles between sampled trees using standardized ^{137}Cs in the needles of each shoot. Therefore, further investigation is necessary to examine the effect of shoot properties on ^{137}Cs distribution in needles after completely eliminating inter-individual variation. For example, multiple terminal shoots consisting of main and lateral shoots could be sampled from an individual tree under the terms of same shoot age and light environment, and the relationships between ^{137}Cs in needles and the morphological traits of each shoot can be examined without intra- and inter-individual variations.

I examined the effect of shoot properties on the ^{137}Cs concentration in needles between the main and lateral shoots of *A. firma* growing in mixed deciduous and coniferous forests in Fukushima. The main shoot had approximately 9% higher ^{137}Cs concentration than the lateral shoots (geometric mean-based comparison). This difference was statistically significant. Therefore, I should consider not only shoot age and shoot position but also the type of shoot (main or lateral) when sampling is conducted for the evaluation of ^{137}Cs contamination of trees having a similar branching like most Pinaceae tree species, in order to control intra-individual variation in a contaminated tree.

Chapter 4

Sampling design and required sample size for evaluating contamination levels of ^{137}Cs in Japanese fir needles in a mixed deciduous forest stand in Fukushima, Japan

Abstract

I estimated the sample size (the number of samples) required to evaluate the concentration of radiocesium (^{137}Cs) in Japanese fir (*Abies firma* Sieb. et Zucc.), 5 years after the outbreak of the Fukushima Daiichi Nuclear Power Plant accident. I investigated the spatial structure of the contamination levels in this species growing in a mixed deciduous broadleaf and evergreen coniferous forest stand. We sampled 40 saplings with a tree height of 150 cm to 250 cm in a Fukushima forest community. The results showed that: (1) there was no correlation between the ^{137}Cs concentration in needles and soil, and (2) the difference in the spatial distribution pattern of ^{137}Cs concentration between needles and soil suggest that the contribution of root uptake to ^{137}Cs in new needles of this species may be minor in the 5 years after the radionuclides were released into the atmosphere. The concentration of ^{137}Cs in needles showed a strong positive spatial autocorrelation in the distance class from 0–2.5 m, suggesting that the statistical analysis of data should consider spatial autocorrelation in the case of an assessment of the radioactive contamination of forest trees. According to my sample size analysis, a sample size of seven trees was required to determine the mean contamination level within an error in the means of no more than 10%. This required sample size may be feasible for most sites.

Key words

Fukushima nuclear accident; natural forests; *Abies firma*; inter-individual variation; spatial autocorrelation; Moran's *I*

Introduction

Once radioactive materials are artificially released into a natural environment, we need to promptly assess the contamination levels of various organisms including humans (cf. Science Council of Japan 2011). The contamination levels of a variety of species including wild plants (Papastefanou et al. 1989; Davids and Tyler 2003; Yoshihara et al. 2013) and animals (McGee et al. 2000; Moller et al. 2012; Hayama et al. 2013; Kubota et al. 2015; Matsui et al. 2015) were assessed after the Chernobyl and Fukushima nuclear reactor accidents.

Generically, the accumulation of pollutants into plant bodies is affected by physiological and ecological plant characteristics as well as environmental factors such as meteorological phenomena and soil types (Baker 1983). The use of plants as a biomonitor for environmental pollution needs to understand factors that influence the accumulation of pollutants into plant body and to standardize sampling methods, sample procedure and statistical analysis (Wagner 1993).

Spatial structures of distributions of organisms often affect the results of statistical test. In nature, organisms always have some functional spatial structures (Legendre and Legendre 1998). The first law of geography, which is 'everything is related to everything else, but near things are more related than distant things' proposed by Tobler (1970), can apply to the spatial structure in ecology. For example, plant populations often show aggregated spatial distribution caused by endogenous factors (e.g. dispersal, spatial competition) and/or exogenous factors (e.g. the heterogeneity of environmental conditions) (Fortin and Dale 2005). This spatial distribution pattern is known as spatial autocorrelation. According to Legendre (1993), spatial autocorrelation is loosely defined as the property of random variables taking values, as pairs of locations a certain distance apart, that are more similar (positive autocorrelation) or less similar (negative autocorrelation) than expected for randomly associated pairs of observations. When spatial autocorrelation exists among ecological data, there are some relationships among the data depending on spatial distance. Because of the relationships among the data, these data are not independent of each other (Legendre and Legendre 1998) and the lack of independence among the data affect the result of statistical analysis (e.g. underestimation of the variance of the mean) (Fortin and Dale 2005). However, if I obtain information about spatial autocorrelation, its information can assist in designing sampling methods (e.g. the distance between observations). In addition, the information can be used to adjust the underestimated variance (Cressie 1993), and this adjustment leads to more reliable estimation of the appropriate number of samples (Petersen and Calvin 1986). Therefore, understanding the spatial structure of observations plays an important role in good sample design and statistical analysis (Legendre et al. 2002). However, the spatial structure of radioactive contamination of forest tree species has been rarely studied.

When investigating environmental pollution such as aerial and soil pollution, tree leaves are often used as biomonitors (Sawidis et al. 1995; Moreno et al. 2003; Tomašević et al. 2004; Maher et al. 2008). In the case of radioactive contamination, many studies have been conducted examining the accumulation of radiocesium (^{137}Cs) in leaves, especially conifer needles. In this study, I investigated the contamination levels in needles of Japanese fir trees (*Abies firma* Sieb. et Zucc.) contaminated by ^{137}Cs released from the Fukushima Daiichi Nuclear Power Plant (FDNPP) to clarify the spatial structure of the radioactive pollution of this species. *A. firma* is a common large evergreen coniferous tree species of forests in Fukushima. It is a shade-tolerant species whose saplings are abundant in shaded understory. This chapter addresses following objectives; (1) to clarify the concentration of ^{137}Cs in current year needles of this species 5 years after the FDNPP accident; (2) to examine whether spatial autocorrelation exists among trees contaminated with ^{137}Cs; and (3) to estimate the number of samples that were necessary and sufficient (i.e. required sample size) to assess the contamination level of this tree species using the results of spatial analysis. The final goal of this study was to provide information about sampling methods for the assessment of contaminated forest trees.

Materials and Methods

Study sites

The study was conducted in a deciduous broadleaf (*Quercus crispula* and *Cornus macrophylla*) and evergreen coniferous (*Pinus densiflora* and *A. firma*) mixed forest located in Soma, Fukushima Prefecture (N37°45’41, E140°47’52) (Fig. 4-1). In a preliminary survey, two plots (20 m × 20 m) set in the forest showed that a tree density of 2825 trees (over 5 cm in DBH) ha^{-1} and basal area of 45.5 m^2 ha^{-1}. 24 tree species were recorded in the plots. The plots were dominated by *Q. crispula* and *A. firma* accounting for 32.7% and 12.8% of total tree number, respectively. The trees of *Q. crispula*, *Quercus serrata*, *P. densiflora* and *A. firma* made up canopy layer at about 20 m from the ground. No bare land was located in the forest. Soil types of the study site is brown forest soil. The forest was located approximately 45 km northwest of the FDNPP. According to the nearest meteorological observation point (approximately 12 km southwest of the study site), the average annual precipitation from 1981 to 2010 was 1361.1 mm and the average annual temperature from 1981 to 2010 was 10.0°C (Japan Meteorological Agency 2016). According to the results of the third airborne monitoring survey conducted from 31 May to 2 July in 2011, the initial deposition of ^{137}Cs around the study site was 100–300 kBq m^{-2} (MEXT 2011).

Samples and ^{137}Cs measurements

The sampling was carried out in March 2016. First, I selected 40 saplings varying in height from 150 cm to 250 cm. I recorded the coordinates of each tree by a level survey using a compass and a laser rangefinder (TruePulse 200, Centennial, Colorado, USA) and measured the tree height with a measuring tape.

Previous studies have commonly reported a large inter-individual variability of ^{137}Cs in the same tree species. For example, Ertel and Ziegler (1991) reported that the needles of *Picea abies* (L.) Karst most highly contaminated with ^{137}Cs had contamination levels two orders of magnitude higher than the least contaminated ones in a southeast Bavarian forest in 1985. A large intra-individual variability in an individual tree is one of the factors that a large inter-individual variability is generated. For example, in the case of evergreen coniferous species, the contamination levels of needle-leaves in an individual tree showed a large variation between branches depending on the age of branches (Sombré et al. 1994), the size of trees (Thiry et al. 2002) and the height at which the branch was located (Yoshihara et al. 2014). Generally, a large variation among environmental samples masks the effect being examined by statistical tests and lessens the power of statistical tests (Cohen 1988). Therefore, I used a sampling method designed to exclude the factors causing a large intra-individual variability (tree size, branch age and branch height) to decrease the variation between individuals. Branch growth in *A. firma* species occurs over spring and summer, sprouting from the ends of the previous year's growth. Buds are produced from summer to winter and remain dormant until branch growth starts again the following spring. This growth pattern allows us to identify branch age by measuring the number of nodal units. I sampled a lateral branch of the annual trunk that had grown in 2015 to standardize the sampling branch age (current year branches) and position on an individual tree. At the same time, I removed the litter layer and collected soils at a depth of 0–5 cm within a 15 cm radius from the base of trees using a soil core sampler (φ50×51 mm in height, 100 ml in volume). One soil sample was taken per tree. Sampled branches were oven-dried at 65°C for 72 h and needles were removed from the branches. The needles were pulverized using a powdering machine (Tube Mill control, IKA Japan K.K., Osaka, Japan) and packed into a plastic container (U-9 container, 50 ml). Soil samples were oven-dried at 80°C for 72 h and sieved using a 2-mm mesh sieve to remove plant roots and pebbles. After that, soil samples were packed into a plastic container (U-9 container, 50 ml).

The ^{137}Cs concentration in samples was determined using a low background germanium detector (GEM30-70, Seiko EG & G Co., Ltd., Tokyo, Japan). The detection efficiency of gamma-rays are reported separately (Shizuma et al. 2016). The measuring time was 10,000 s for each sample. Measurement uncertainty averaged under 10% for all samples. The ^{137}Cs concentration was converted to that on the sampling date based on the physical decay rate.

Statistical analysis

The concentration of ^{137}Cs in needles and soil were both transformed to a natural logarithm to meet the criteria of normal distribution (Kolmogorov-Smirnov Test, needles: $p = 0.072$, soil: $p = 0.2$). I calculated the sample size required from the sample variance as follows. I tested the spatial patterns of ^{137}Cs concentrations from sampled tree needles and soils for spatial autocorrelation using Moran's *I* statistics. The Moran's *I* statistic at distance class d (I (d)) is calculated from the following equation (1):

$$I(d) = \frac{\left(\frac{1}{W(d)}\right)\sum_{i=l}^{n}\sum_{j=l}^{n} w_{ij}(d)(x_i - \bar{x})(x_j - \bar{x})}{\frac{1}{n}\sqrt{\sum_{i=l}^{n}(x_i - \bar{x})^2}} \cdots (1),$$

where wij (d) is the distance class connectivity matrix that indicates whether a pair of sampling locations are in distance class d, xi and xj are the values of the ^{137}Cs concentrations (x) at sampling location i and j, and W (d) is the sum of wij (d) (Fortin and Dale 2005). Values for the Moran's *I* statistics run from −1 to +1 (Fortin and Dale 2005). Positive values indicate positive autocorrelation, and negative values show negative autocorrelation. If the value is close to 0, there is no significant autocorrelation (Fortin and Dale 2005). Fifty distance classes with 2.5-m distance intervals were used. The significance of the Moran's I statistics were obtained by a randomization test of 5000 permutations. This analysis was carried out using the software package R version 2.5.0 (R Development Core Team 2005).

I calculated the sample size required (n_{req}) using the equation by Petersen and Calvin (1986) as follows:

$$n_{req} = t_{0.95}^2 s^2 \Big/ D^2 \cdots (2),$$

where $t_{0.95}$ is the Student's t statistic with n-1 degrees of freedom at the 95% probability, s^2 is the sample variance of the ^{137}Cs concentration in needles, and D is the specified error limit. D denotes the allowable 95% confidence interval of the mean. In this study, I estimated the n_{req} to be the D smaller than or equal to 10% and 20% of the sample mean. When a positive spatial autocorrelation exists, the observed sample variance is known to be underestimated compared with the true variance (Fortin and Dale, 2005). The underestimation leads us to accept a smaller n_{req} than really exists. I corrected the underestimation of observed sample variance when a significant positive autocorrelation was found as follows. First, I calculated the correction factor (θ) following Cressie (1993). The correction factor is expressed by I (d) (Moran's I statistics) as:

$$\theta = \left[1 + 2\frac{I(d)}{1 - I(d)}\left(1 - \frac{1}{n}\right) - 2\frac{I(d)^2}{(1 - I(d))^2}\frac{1 - I(d)^{n-1}}{n}\right] \quad \ldots (3)$$

Then the equation4- (2) was modified as:

$$n_{req} = t_{0.95}^2 \theta s^2 \Big/ D^2 \quad \ldots (4)$$

Results

^{137}Cs concentration in needles and soil

The maximum (minimum) values of ^{137}Cs concentrations in the needles of lateral branches of annual trunk of Japanese fir and soil under the sampled trees were 7020 (156) Bq/kg-DW and 38100 (3940) Bq/kg-DW, respectively. The geometric mean ^{137}Cs concentration in the needles was 1530 Bq/kg-DW, whereas in the soil the mean was 10500 Bq/kg-DW. The standard deviation of ^{137}Cs concentration in needles and soil ranged from 738 to 3190 Bq/kg-DW and from 6340 to 17600 Bq/kg-DW, respectively. There was no significant correlation between the ^{137}Cs concentration in needles and soil (Pearson's correlation coefficient $r = 0.217$, $p = 0.179$).

The locations and contamination levels of needles taken from target trees and soil are shown in Figure 4-2. The Moran's *I* correlogram of the ^{137}Cs concentration in needles (Fig. 4-3a) showed strong significant positive autocorrelation at the distance class from 0 to 2.5 m (Moran's *I* autocorrelation coefficient, $I(d) = 0.375$, $p < 0.01$). In addition, 6 other significant autocorrelations were found. The Moran's *I* of soil showed no significant spatial autocorrelation at the 0–2.5 m distance class (Fig. 4-3b). The pattern of Moran's *I* spatial correlogram of the ^{137}Cs concentration of needles showed a quite different pattern from that of soils (Figs 4-3a and 4-3b).

The number of required samples

Because the ^{137}Cs concentration in needles showed a significant positive autocorrelation at the distance class of less than 2.5 m, I corrected the observed variance of the ^{137}Cs concentration in needles to calculate the required sample sizes. After correction, the required sample sizes were calculated as seven and two for allowable margins of error of 10% and 20% in the sample mean at the 95% probability level, respectively.

Discussion

The contamination levels of ^{137}Cs, 5 years after the FDNPP accident, in *A. firma* sapling current year needles that sprouted in 2015 were shown in Figure 4-2. Kato et al. (2015) reported that the concentrations of ^{137}Cs in throughfall and stemflow were low about 2 years after the FDNPP accident, especially in a mixed broad-leaved stand. I thus assumed that the possibility of external contamination and absorption from the surface of needles in this study was low. From the results that (1) there was no correlation between the ^{137}Cs concentration in sampled needles and soil, and (2) there was a difference in the spatial distribution pattern of the ^{137}Cs concentration between needles and soil, I deduce that the contribution of root uptake to the concentration of ^{137}Cs in new needles in this study may be small 5 years after the radionuclides were released into the atmosphere. Nishikiori et al. (2015) also indicated that there was a small contribution of root uptake to contamination in new leaves of *Cryptomeria japonica* that sprouted in 2012 in forest located approximately 160 km southwest of the FDNPP, and that ^{137}Cs in new leaves was translocated from other tree parts. I conclude that the ^{137}Cs concentration in new needles in the present study may be largely explained by the translocation of radiocesium within the tree body.

As far as we know, this is the first study to examine the geostatistical characteristics of ^{137}Cs in trees. My study showed a strong positive spatial autocorrelation of ^{137}Cs in trees at the distance class within 2.5 m. Although the underlying mechanism creating the spatial autocorrelation remains unclear, a spatial autocorrelation over a short distance might be applicable to other tree species growing in a similar environment. Spatial autocorrelation affects accurate sampling in that the presence of a significant positive autocorrelation violates the independence of observations, resulting in an underestimation of the variance of sampling means (Fortin and Dale 2005). According to Fortin and Dale (2005), it is important to understand the characteristics of spatial autocorrelation before the start of research. This understanding will make it possible to design sampling methods that avoid spatial autocorrelation, or to reduce the effect of spatial autocorrelation on following analyses. They suggest, therefore, that a pilot study for the design of sampling is required. However, the assessment of radioactive contamination of the environment is often urgent (Science Council of Japan 2011). Therefore, the best sampling methods would be designed to allow the analysis of a spatial pattern of radioactive contamination of forest trees at the same time that an assessment of contamination levels of forest trees is conducted. For example, the locations of trees are recorded to examine whether a significant positive spatial autocorrelation is present in a study plot. If a significant positive spatial autocorrelation is detected, an observed sample variance can be corrected using a correction factor calculated using Moran's *I* statistics. My results showed six additional significant autocorrelations in the ^{137}Cs concentration in needles in some distance classes. However, the processes underlying these autocorrelations are unclear. Further investigations are needed to clarify them.

The accumulation of ^{137}Cs into tree leaves is affected by various factors. Yoshihara et al. (2014) reported that the concentration of ^{137}Cs in leaves differed between leaves near the top of the tree and those in the lower parts of the tree. In addition, the concentrations of ^{137}Cs in leaves were affected by the leaf age (Sombré et al. 1994; Yoshida et al. 2004). If the sampling design ignores these factors, the variance of ^{137}Cs between trees may become unnecessarily large. A large variation between individuals may be reduced by a well-designed sampling method that excludes factors caused by a large intra-individual variability (Fisher 1966). It is therefore necessary to standardize the characteristics of sampled branches for the assessment of radioactive contamination of forest trees. This allows us to minimize variation generated by poor sampling methods, resulting in a small variance between trees. According to my sample-size analysis based on the observed variance between trees, the sample sizes required to fall within the 95% confidence limit of the population mean of ^{137}Cs concentrations, with allowable margins of error of 10% and 20% of the population mean, were seven and two, respectively. These sample sizes would be feasible for many case studies. Additional studies are needed to test if my results are applicable to other forest stands and other tree species.

I found the spatial structure of radioactive contamination of trees growing in a mixed deciduous broadleaf and evergreen coniferous forest. In the present study, however, the results were derived from only one forest stand. Several studies reported that the dynamics of ^{137}Cs in forest ecosystems depend on the type of forest stand (Melin et al. 1994; Goor and Thiry 2004; Komatsu et al. 2016). Thus, based on the present analysis, further investigations are needed to clarify the spatial structure of contamination of various forest stands, including stands in plantation and natural forests. Additionally, using these results, it is necessary to develop methods of scaling up from a stand-level evaluation to a forest-level evaluation in assessing the overall contaminated forest areas in future work.

Chapter 5

Radiocesium concentration in seeds of Japanese fir (*Abies firma* Sieb. et Zucc.) growing in Fukushima forests—implications for radiocesium contamination in offspring

Abstract

Seed soundness is of paramount importance for all plants. Seed soundness of forest trees might be negatively affected after the Fukushima Daiichi Nuclear Power Plant (FDNPP) accident in March 2011, when a massive amount of artificial radionuclides deposited on forest areas. However, information on seeds of forest tree species contaminated with radionuclides is limited because they are rarely encountered in forest ecosystems. Japanese fir (*Abies firma* Sieb. et Zucc.) is a predominant tree in natural secondary forests in the Abukuma Mountains in Fukushima, and this species shows masting; cone production occurs every 2 or 3 years, meaning that the contamination levels of *A. firma* seeds remains unknown. I investigated the contamination levels of ^{137}Cs in cone components (seeds, wings and cone scales) of *A. firma* approximately 4.5 years after the FDNPP accident. The ^{137}Cs concentration in seeds showed the lowest contamination level of the three cone components. In addition, I compared the ^{137}Cs concentration between cone components and current-year needles of this species; a low concentration of ^{137}Cs in seeds compared with needles was observed. My results suggested that the combination of low contamination levels of seeds of *A. firma* and small root uptake of ^{137}Cs from soil have led to low contamination levels of ^{137}Cs in recruits of this species.

Key words

^{137}Cs; translocation; evergreen conifer; cones; future generations

Introduction

The Fukushima Daiichi Nuclear Power Plant (FDNPP) accident in March 2011 had severe consequences for the terrestrial environment, releasing a large amount of radioactive materials into the atmosphere. Radiocesium, especially ^{137}Cs, is a particularly important radionuclide because of its long half-life (30.2 y), resulting in long-term contamination of ecosystems. Approximately 70% of the contamination area caused by the FNDPP accident is covered with forests (Hashimoto et al. 2012). After the deposition on forest areas, radiocesium is transferred into nutrient cycles in forest ecosystems over time, and accumulates in various forest components (IAEA 2006). In the initial phase of forest contamination after deposition, the adsorption of radiocesium on the canopy from the atmosphere and/or the absorption from the plant surface into tree bodies are important factors in determining tree contamination (Calmon et al. 2009). Following the initial phase, the contamination level of trees mainly depends on the following two processes; (1) a part of the radiocesium adsorbed on the plant surface is removed by rainfall and/or litterfall, and (2) root uptake and/or internal translocation of radiocesium affects tree contamination levels over a long period of time (Goor and Thiry 2004; Calmon et al. 2009). In tree bodies, radiocesium accumulates in different tree organs such as leaves, branches and wood, and the distribution of radiocesium among tree organs has been found to be heterogeneous (Strebl et al. 1999; Calmon et al. 2009; Kuroda et al. 2013; Yoshihara et al. 2013). For example, the concentration of radiocesium in leaves (needles), twigs and bark was higher than those in wood (Ertel and Ziegler 1991; Goor and Thiry 2004; Komatsu et al. 2016). Although many studies about forest trees contaminated by radiocesium were conducted after the Chernobyl and Fukushima accidents, few studies have investigated tree contamination related to a temporary event in the life history of trees such as reproduction, because of limited opportunities.

Abies firma Sieb. et Zucc. is a common large evergreen conifer species in the Abukuma Mountains in Fukushima, Japan. This species is shade-tolerant and saplings are abundant in shaded understory. This species regenerates mainly by sexual reproduction. Generally, *A. firma* shows masting; cone production occurs every 2 or 3 years (Yuruki and Aragami 1973; Aragami 1987; Mizunaga et al. 1998). Because of a supra-annual cycle of reproduction, there is limited opportunity to sample diaspore of this species. To adequately assess the contamination levels of offspring that create future generations, it is important to understand the contamination levels of diaspore. Fortunately, I encountered the masting of this species in Fukushima in 2015. In this study, therefore, I investigated for the first time the contamination levels of ^{137}Cs in cone components (seeds, wings and cone scales) of *A. firma* growing in a mixed deciduous and coniferous secondary forest in Fukushima. First, I examined the concentration of ^{137}Cs in three different cone components and the difference in contamination levels between these three parts. Next, I compared the ^{137}Cs concentration in cone components with the ^{137}Cs concentration in current-year needles of this species to evaluate the contamination levels of diaspore of *A. firma*. On the basis of these results, I discuss the effects of ^{137}Cs contamination on future generations of *A. firma* in Fukushima.

Materials and Methods

Study sites and sampling

The study site was a mixed deciduous and coniferous secondary forest in Iitate, Fukushima Prefecture, which was located approximately 45 km northwest from the Fukushima Daiichi Nuclear Power Plant (N37°44'05", E140°49'35"; Fig. 5-1). The mean annual precipitation from 1981 to 2010 was 1361.1 mm, and the mean annual temperature was 10.0°C recorded at the nearest meteorological observation point (Japan Meteorological Agency 2016).

After the FDNPP accident, the masting of *A. firma* in Fukushima was observed in 2013 and 2015 (my field observations). My field survey was conducted in November 2015. Cone samples were collected from the forest floor. Generally, *A. firma* produce winged seeds and the type of seed dispersal of this species is anemochory. Typically, winged seeds with cone scales are separated from the axis of the cone at cone maturity and depending on meteorological weather conditions (e.g., the wind speed), they fall onto the forest floor. Cones can often fall under the canopy without separating, but mature cones found on the ground are very fragile. Therefore, I identified maternal trees of cones on the ground by the distance from the trees and the condition of cones (non-separating). Nine cones were obtained from four maternal trees. Current-year needles in 2015 were also sampled from the maternal tree for comparison with the contamination level of the cones. I sampled 12 current-year shoots from each maternal tree. Because of the difficulty of accessing shoots of the upper foliar part of maternal trees, shoots were sampled from a height of approximately 5–7 m from the ground. The sample descriptions are shown in Table 5-1.

Sample procedures for ^{137}Cs measurements

A cone sample was separated into three groups (seeds, wings and cone scales) after being oven-dried at 65°C for 72 h (Fig. 5-2) and weighed to determine the mass of each part of the cone. Cone scales were pulverized with a powdering machine (Tube Mill control, IKA Japan K.K., Osaka, Japan). Seeds, wings and powdery cone scales were packed into a plastic container (U-9 container, 50 ml). Needle samples were removed from a branch after being oven-dried at 65°C for 72 h and pulverized with the powdering machine. This process was conducted for every shoot separately (n = 12 × 4 trees). Powdery needles were also packed into the plastic container.

The ^{137}Cs concentration in samples was measured using a low background germanium detector (GEM30-70, Seiko EG & G Co., Ltd., Tokyo, Japan). The detection efficiency of gamma-rays was reported separately (Shizuma et al. 2016). The measurement time was 20,000 s for each sample. Uncertainty of measurement averaged approximately 15% for cone samples and 10% for needle samples. The ^{137}Cs concentration was converted to that on the sampling day based on the physical decay rate.

Statistical treatment

I performed the Friedman test to compare the ^{137}Cs concentration, dry mass and the amount of ^{137}Cs among seeds, wings and cone scales. The p -values were corrected using the sequentially rejective Bonferroni technique for multiple comparisons. I also performed a Spearman rank correlation test to examine the relationships of ^{137}Cs between seeds, wings and cone scales. To compare the ^{137}Cs concentration in three different cone components with current-year needles (the average value (n = 12 shoots) per tree), a one sample t-test was performed. The mean value and 95%, 99% and 99.9% confidence interval (CI) of the mean ^{137}Cs concentration in current-year needles were calculated separately from every maternal tree. Based on the CI of the mean of ^{137}Cs concentration in needles of the maternal tree, I determined significant differences in ^{137}Cs concentration in seeds, wings and cone scales of a cone from that in the needles. For example, if the 95% CI of ^{137}Cs concentration in needles of the maternal tree did not include the ^{137}Cs concentration in seeds of a cone from the maternal tree, I determined that the seed had a significantly different concentration from the needle at the 5% significance level. These statistical analyses were carried out using IBM SPSS Statistics version 21.0 for Windows.

Results

The average values of ^{137}Cs were 175 Bq/kg-DW for seeds, 383 Bq/kg-DW for wings and 646 Bq/kg-DW for cone scales. The maximum–minimum values of the concentration of ^{137}Cs were 539–43 Bq/kg-DW for seeds, 1055–58 Bq/kg-DW for wings and 1875–204 Bq/kg-DW for cone scales. There were significant differences in ^{137}Cs concentration between seeds, wings and cone scales (Friedman test, $x^2 = 14.222$, $df = 2$, $p < 0.01$; Fig. 5-3a). The concentration of ^{137}Cs in seeds was significantly lower than that in wings and cone scales. Fig. 5-3b shows the dry mass of seeds, wings and cone scales. In addition, I estimated the amount of ^{137}Cs in each part of the cone based on the ^{137}Cs concentration and the total mass of each cone; the total amount of ^{137}Cs in seeds was significantly lower than that in cone scales, but not significantly lower than that in wings (Fig. 5-3c). The relationship of ^{137}Cs between seeds and wings was not significant (Spearman correlation coefficient = 0.633, $p = 0.067$, Fig. 5-4), but significant correlations were obtained between seeds and cone scales and between wings and cone scales (Spearman correlation coefficient = 0.85, $p < 0.01$ and Spearman correlation coefficient = 0.683, $p < 0.05$, respectively).

The comparison of ^{137}Cs concentration between seeds and needles showed that the ^{137}Cs concentration in seeds was relatively low when compared with that in needles; of nine comparisons, seven comparisons resulted in significantly lower concentrations in seeds, one comparison resulted in significantly higher concentrations in seeds and one comparison was neutral (Fig. 5-5). In contrast, the ^{137}Cs concentrations in wings and cone scales were relatively high when compared with those in needles (wings: five were significantly higher, three were significantly lower and one was not significantly different; and cone scales: seven were significantly higher, one was significantly lower and one was not significantly different).

Discussion

The transfer of ^{137}Cs into cones of *A. firma* was observed approximately 4.5 years after the FDNPP accident. The translocation of radiocesium into diaspore of trees was reported by several previous studies (Ertel and Ziegler 1991; Ipatyev et al. 1999; Fogh and Andersson 2001). However, there were few studies that referred to the contamination levels in different cone components. In this study, the differences in ^{137}Cs concentration between three different cone components were shown (Fig. 5-3a). The lowest ^{137}Cs concentration was observed in seeds, whereas cone scales showed the highest concentration of ^{137}Cs of the three cone components. One possibility of this result was the external contamination of cone scales. In the present study, cone samples were collected from the forest floor even if samples were on the litter layer. In forest ecosystems, soil often shows the highest contamination levels. Cone scales are the outermost parts of cones. Therefore, there was a possibility that external contamination was present on the cones. To examine whether cone components had external contamination, I performed autoradiography using imaging plates to clarify the radioactive distribution of cone samples. In autoradiography, generally, radioactivity caused by external contamination on plants shows different characteristics from internal contamination in the radioactive distribution images. From the autoradiography results, the radioactivity distributions caused by external contamination in cone components including cone scales were rarely found (Fig. 5-6). Therefore, I assumed that the possibility of external contamination was small. The mechanism generating the differences in ^{137}Cs concentration between the three cone components was unclear, therefore future work should address gaining a better understanding of the factors that influence the contamination levels in each cone component.

In Figure 5-5, two cone samples collected from the same maternal tree showed high ^{137}Cs concentrations compared with other samples. In this study, nine cone samples were collected from four maternal trees, and the distance between trees was always within 50 m, suggesting that there was a large variation in contamination level between cones of *A. firma* within a small spatial scale in forests. The underlying mechanisms generating the large variances remain unclear. After the Chernobyl accident in 1986, cones of Scots pine (*Pinus sylvestris* L.) were collected within the 30-km zone around Chernobyl in the years 1987–1991, and the contamination levels of cones showed an increasing tendency with increasing deposition of radioactive materials in soil (Ipatyev et al. 1999). In this study, however, I did not examine the ^{137}Cs contamination levels in soil. Thus, examination of the relationships between contamination levels of cones and soils in Fukushima forests should be conducted. Additionally, a large inter-individual variability of ^{137}Cs in the same tree species was reported by several previous studies (Ertel and Ziegler 1991; Yoshihara et al. 2014). According to Ertel and Ziegler (1991), the needles of *Picea abies* (L.) Karst most highly contaminated by ^{137}Cs had contamination levels two orders of magnitude higher than the least contaminated ones in a southeast Bavarian forest. Thus, further studies are required to investigate whether the contamination levels of maternal trees affect the contamination level of cones and therefore clarify the factors involved in determining the contamination levels of diaspore.

I compared the ^{137}Cs concentration between cone components and current-year needles of *A. firma*, with the result that concentrations of ^{137}Cs in seeds were low when compared with those in needles (Fig. 5-5). The distribution of ^{137}Cs in an individual tree is heterogeneous even when comparing the same tree parts. For example, the contamination levels in upper foliar parts of evergreen coniferous species have a tendency to be higher than those in lower foliar parts (Melin et al. 1994; Kaunisto et al. 2002; Yoshihara et al. 2014). Yoshihara et al. (2014) reported the vertical distribution of radiocesium in the canopy of several tree species, and the upper/lower ratios (ratios of radiocesium concentrations in the upper foliar parts to those in the lower foliar parts) of Japanese cedar were high (about 1.0–2.0). In fact, my preliminary survey showed that the ^{137}Cs concentration in needles of *A. firma* saplings taken from upper parts was higher than that taken from lower parts (Oba et al. unpublished data). The most popular explanation for this phenomenon is that the behaviour of cesium in tree bodies is similar to that of potassium; the high mobility of these elements is important for internal translocation to newly emerging parts of trees (Sombré et al. 1994; Kaunisto et al. 2002). Yoshida et al. (2011) also reported high concentrations of ^{137}Cs in young parts of Scots pine (*Pinus sylvestris* L.) 12 years after the Chernobyl accident, suggesting that ^{137}Cs has a tendency to translocate into growing parts. In the present study, needle samples were taken from the lowest foliar part of each sampled tree, and the ^{137}Cs concentration in sampled needles was still significantly higher than that in seeds (except for one individual tree, but the difference between ^{137}Cs concentration in needles and seeds was small; see Fig. 5-5). Therefore, the contamination level of ^{137}Cs in seeds appears to be lower than that in needles in a whole tree body.

Following the Chernobyl accident, 4–5 years after radioactive materials were released into the atmosphere, the major factors that determined the contamination level of trees were root-uptake and internal translocation of radiocesium (Goor and Thiry 2004; Calmon et al. 2009). Although the root uptake of ^{137}Cs has limitations because of fixation of ^{137}Cs to clay minerals over time (IAEA 2006), soil-to-plant transfer of ^{137}Cs is one factor that contributes to long-lasting forest tree contamination (Calmon et al. 2009). According to Goor and Thiry (2004), the root-uptake of ^{137}Cs into trees of a Scots pine stand in Gomel was observed 15 years after the Chernobyl accident, although the contribution of tree contamination was smaller than that of internal translocation. In contrast, Nishikiori et al. (2015) reported that the contribution of root uptake to contamination in new leaves of *Cryptomeria japonica* that sprouted in 2012 was already small in forest located approximately 160 km southwest of the FDNPP, and that ^{137}Cs in new leaves was translocated from other tree parts approximately 1 year after the accident. In the present study, from the results of low ^{137}Cs concentrations and small dry mass in seeds, the total ^{137}Cs in seeds per cone was very small.

Given the small contribution of root uptake and low contamination of seeds of *A. firma*, the contamination levels of this species in future generations might be very low. To understand the secondary contamination through reproduction events, long-term monitoring for assessment of forest tree contamination is required over generations.

Chapter 6

General Discussion

The dynamics of ^{137}Cs in a Fukushima forest ecosystem

The discharge of ^{137}Cs from forest ecosystems to the areas surrounding the forest has become an important issue following the ^{137}Cs contamination caused by the nuclear crisis. The discharge of ^{137}Cs may represent a new source of pollution of the environment and organisms. Tikhomirov and Shcheglov (1994) reported that the net ^{137}Cs transferred from a forest ecosystem contaminated by the Chernobyl accident was below 1%. Therefore, the amount of discharged ^{137}Cs could be low and thus the physical decay rate of ^{137}Cs is likely to be a major factor in defining the scope of the impact of radiation on the environment except human-induced transport of ^{137}Cs such as decontamination activities (IAEA 2006). In the case of the Fukushima accident, the changes in the amount of ^{137}Cs in forest areas were also small. Most of the ^{137}Cs was present in the surface layer of soil and the amount of ^{137}Cs in the mountain stream was small. This indicates that the export of ^{137}Cs out of a forest ecosystem can be assumed to be small (Forestry Agency 2017). Once ^{137}Cs has been deposited on forest areas from the atmosphere, it remains in forest ecosystems for a long time. Therefore, the understanding of a long-term dynamics of ^{137}Cs in forest ecosystems is of paramount importance for the management of forests after a nuclear crisis. The long-term dynamics of ^{137}Cs in a forest ecosystem may be strongly affected by several environmental factors such as climate conditions and vegetation, therefore these factors must be determined for both the Chernobyl forests and the Fukushima forests.

To consider the long-term dynamics ^{137}Cs in Fukushima forest ecosystems, we must pay special attention to deposition and interception, internal contamination and soil contamination, which play key roles in the long-term dynamics ^{137}Cs in a forest ecosystem. I next discuss these three features separately.

Deposition and interception

Tree canopies adsorb various pollutants from the atmosphere and act as an efficient filter. After the release of ^{137}Cs into the atmosphere, ^{137}Cs (either particulate ^{137}Cs (dry deposition) or contaminated water droplets (wet deposition)) is intercepted by forest canopies. In particular, evergreen conifer stands are known to be an effective filter for dry deposition (Bunzl et al. 1989). Bunzl et al. (1989) reported that evergreen conifer stands, which retain their foliage throughout a year are a particularly efficient filter of the interception mechanisms, unlike deciduous tree species.

After the deposition phase, ^{137}Cs deposited on plant surfaces was gradually removed by rainfall (such as throughfall and stemflow) and/or litterfall (biomass fall driven by tree phenology or extreme meteorological events such as typhoons) (Bunzl et al. 1989; IAEA 2006). The ^{137}Cs is transferred from forest canopies to the forest floor through these effects. In the case of the Fukushima accident, Kato et al. (2012) described the process of the ^{137}Cs transfer from tree canopies to forest floor as follows: (1) The amount of removal of ^{137}Cs from plant surfaces was less than 10% of the total deposition for about 2 weeks between March 11 and March 28 during the first rainfall event, (2) The leaching of ^{137}Cs from forest canopies was observed even 2 months after the accident, (3) The amount of ^{137}Cs at the forest floor showed logarithmic increases in various tree species stands, indicating that the ^{137}Cs in the canopy was removed and transferred to the forest floor over time, (4) About 5 months after the FDNPP accident, only approximately 60% of the total deposited ^{137}Cs was observed in forest canopies in cypress stands and 70% in cedar stands, (5) The half-life of ^{137}Cs absorbed in the canopies was estimated from the observation period (160 days after the beginning of radioactive fallout) as 620 days for the cypress and 890 days for the cedar stand, (6) These values were considerably longer than that of 100 days for the Norway spruce, and (7) This indicates that the removal of radionuclides through absorption by the cypress and cedar stands was substantially slower than that by the Norway spruce stand. In case of forests contaminated after the Chernobyl fallout, evergreen coniferous forest vegetation is likely to have intercepted 90% of the ^{137}Cs deposit, and 80% of the deposition was gradually transferred to the forest floor in 960 days (Gonze and Calmon 2017). This also supports the idea that the cypress and cedar stands have a slow removal rate.

Yoshihara et al. (2013) showed that the concentration of radiocesium (^{134}Cs + ^{137}Cs) in pre- and post-fallout sprouted leaves of evergreen species was higher than those in deciduous species, and suggested that this was because deciduous species did not possess foliar parts at the time of the accident. In contrast, Nishikiori et al. (2015) reported that there was no difference in ^{137}Cs concentration of leaves sprouted in 2012 between evergreen and deciduous species at their study sites, and they suggested that there was likely to be a more important factor determining tree contamination than the presence or absence of leaves at the time of radiocesium deposition.

Internal contamination

The transfer of ^{137}Cs into tree bodies could have begun after the deposition, and the accumulation of ^{137}Cs in wood was observed within a few months after the Fukushima accident (Gonze and Calmon 2017). Internal tree contamination may progress via translocation from (1) plant surfaces, (2) other parts of trees, and (3) root uptake from contaminated soil. The root uptake of ^{137}Cs is expected to be limited because of the strong fixation of ^{137}Cs to clay minerals over time (IAEA 2006), and therefore the internal translocation of ^{137}Cs (the movement of ^{137}Cs between tree parts) might be prolonged by tree contamination.

The internal translocation of ^{137}Cs has been observed and evaluated for various tree species. Wood is an important component of this translocation because it has a huge biomass and we use it for various purposes such as construction. In Japanese cedar (*Cryptomeria japonica*), red pine (*Pinus densiflora* Sieb. et Zucc.) and oak (*Quercus serrata* Thumb.), both sapwood and heartwood were contaminated by ^{137}Cs at the Ohtama site in Fukushima half a year after the Fukushima accident (Kuroda et al. 2013). However, the ^{137}Cs contamination in wood parts was lower than those of the bark, which suggests that the transfer of ^{137}Cs into the tree stem occurred at a very early phase of radioactive contamination (Kuroda et al. 2013).

Nishikiori et al. (2015) investigated the ^{137}Cs concentration of *C. japonica* leaves sprouted in 2010 by washing using water or chloroform to examine the presence of ^{137}Cs (inside the leaf or on leaf surfaces). They showed that 17–38% of ^{137}Cs was present inside the leaves and 62–83% was adhered to the leaf surfaces. Yoshihara et al. (2013) also reported radiocesium accumulation in the post-fallout-expanded leaf tissues, with a variable activity concentration that was irrespective of species. From the results, they suggested the influence of species-specific translocation of radiocesium in tree bodies or uptake from the soils. Nishikiori et al. (2015) provided information about the source of ^{137}Cs in *C. japonica* leaves sprouted in 2012. Their results indicated that ^{137}Cs in new leaves of *C. japonica* may be supplied by internal translocation from the other tree organs. They also suggested the contribution of root uptake must have been small. According to Goor and Thiry (2004) and Thiry et al. (2009), the translocation, especially from senescing foliage, contributed a major portion (60–70%) of the annual supply of ^{137}Cs in Scots pine contaminated by the Chernobyl accident. The translocation of ^{137}Cs toward the tip of *C. japonica* needles (old needles to young needles and subsequently to male flowers) was reported by Kanasashi et al. (2015). These results are strong evidence for the occurrence of acropetal translocation of ^{137}Cs from old needles to young needles, male flowers and pollen. Yoshihara et al. (2014) showed that newly expanded foliar parts contained higher ^{137}Cs concentrations than older parts for evergreen coniferous species and also indicated that ^{137}Cs translocation was correlated with that of nutritional flow.

Establishment of the assessment method of radioactive contamination for forest ecosystems using Japanese fir trees

Approximately 70% of Fukushima Prefecture is covered with forests. A large amount of radionuclides, especially radiocesium (^{137}Cs), was deposited onto the forested areas after the FDNPP accident. To understand the effect on forest organisms and for the safety of workers and residents near forests, contamination levels of various organisms must be promptly assessed after radioactive materials are released into the environment. However, radioactive contamination of forest ecosystems is complex, consisting of various ecological factors. Hence, the effects of radioactive pollution in Japanese forests may differ from the effects in North European forests caused by the Chernobyl accident. Ecological factors affecting North European forest ecosystems are substantially different from those in Japan. Therefore, an understanding of the behavior of radiocesium in Japanese forest ecosystems is required.

Forest trees have a huge aboveground biomass and they have the potential to provide storage of huge amounts of radioactive material and to be a source of radiation emissions. Many studies on tree contamination were conducted after the Chernobyl accident, and these reported the heterogeneity of distribution of radiocesium within a tree body (intra-individual variations). Moreover, there are large inter-individual variations in radiocesium contamination of forest trees, often affecting the results of statistical tests. We need to consider these large variations when investigating radioactive contamination of forest trees; otherwise we cannot understand the true effects of ecological factors or obtain an accurate assessment of the contaminated environment.

Spatial structures of field observations also frequently affect the results of statistical tests. In the field, organisms always have same functional spatial structures (Legendre and Legendre 1998).

However, the spatial structure of radioactive contamination of forest trees has been rarely studied. Well-designed sampling methods help to avoid the effects of large variations and special structures of observations on the results of statistical analysis. In this study, to establish an effective sampling method, I propose a new sampling method based on the results of my studies on an evergreen conifer species within a Japanese forest ecosystem.

In this study, I focused on the evergreen conifer, Japanese fir (*Abies firma* Sieb. et Zucc.). This species is commonly distributed in natural forests in the coastal area of Fukushima Prefecture. *A. firma* has a specific branching pattern that allows us to identify the shoot age. Tree age is an important factor for the radiocesium distribution in tree bodies, and this information helps us to avoid the large variation caused by differences in age. Based on this species, I have established an effective sampling method for the assessment of contaminated trees growing in Japanese forest ecosystems.

In Chapter 2, I show the distribution pattern of ^{137}Cs contamination in a young *A. firma* tree body. The results of this study indicate that visual classification of shoot age is an appropriate working method for the assessment of ^{137}Cs contamination in *A. firma* tree organs. This study clarified the differences in ^{137}Cs contamination between different ages and between different tree organs such as needles, branches, wood and bark. Moreover, the vertical distribution of ^{137}Cs contamination in an individual tree body was also discussed. Both tree age and tree height data allow us to distinguish external contamination from internal contamination. In conclusion, the heterogeneity of ^{137}Cs contamination in tree bodies was observed, which was brought about by (1) differences in organs such as needles, branches, wood and bark; (2) differences in age; and (3) position (height) of organs. Thus, standardization of sampling to avoid the effect of intra-individual variation is required for the assessment of radioactive contamination in forest tree species.

Chapter 3 presents additional information for highly accurate sampling methods. The branching pattern of *A. firma* is trifurcate. In this branching pattern, we can see clear morphological differences between three shoots (one main shoot and two lateral shoots). I investigated the difference in ^{137}Cs concentration between three different shoots. The ^{137}Cs concentration in main shoots was significantly higher than those in lateral shoots. This indicates that there were differences in ^{137}Cs concentration between shoot type even for the same organs of the same age and at the same position (height). In conclusion, it is important to select samples while paying attention to shoot differentiation. Samples should be taken only from the main shoot or only from the lateral shoot (not a mix of shoots) for every sampled tree.

In Chapters 2 and 3, I discussed the intra-individual variation of ^{137}Cs concentration in *A. firma*. The differences in ^{137}Cs concentration caused by different organs, ages and sampling positions when field sampling is conducted were considered important. Next, in Chapter 4, I attempted to clarify the inter-individual variation of ^{137}Cs concentration in this species. Spatial autocorrelation is one of the factors affecting inter-individual variation in ecological data, causing a lack of independence of ecological data. Non-independence of ecological data leads to various statistical problems. Examination of the spatial structure of observations between ecological data is required to clarify the presence of spatial autocorrelation. However, the spatial structure of ^{137}Cs contamination in forest trees remained unclear. In Chapter 4, I investigated the spatial structure of ^{137}Cs concentration of *A. firma* in a natural secondary forest and determined the presence or absence of spatial autocorrelation. A significant strong spatial autocorrelation was observed over short distances (< 2.5 m), suggesting that spatial distribution needs to be considered in the evaluation of radioactive contamination in trees. Therefore, I strongly recommend taking position data of sampled trees when sampling is being conducted as this allows adjustment of non-independent data due to the spatial autocorrelation with the position data. Furthermore, I described the number of samples required to evaluate radioactive contamination in trees based on the results of spatial analysis. According to the sample-size analysis, a sample size of seven trees was required to determine the mean contamination level within an error of the mean of no more than 10%. This required sample size may be feasible for various fields.

The ^{137}Cs contamination of future *A. firma* generations in Fukushima forests

In Chapter 5, I showed the low contamination levels in *A. firma* seeds. This result, together with the limited root uptake of ^{137}Cs due to the strong fixation of ^{137}Cs to clay minerals (IAEA 2006), suggests that ^{137}Cs contamination in future generations of this species growing in forests surrounding the FDNPP will be low. Although I predicted that their contamination levels in future would be low, it would be difficult to determine whether they would be safe. The risk of chronic exposure of irradiation for trees has recently been reported (Volkova et al. 2017). A dose rate of 100 μGy h^{-1} is considered to be safe for forest ecosystems by the United Nations Scientific Committee on the Effects of Atomic Radiation considers (UNSCEAR 2008). However, Volkova et al. (2017) showed biological effects in European Scots pine populations that were chronically exposed to significantly lower dose rates for 30 years following the Chernobyl accident. This study indicates that the chronic exposure by relatively low dose rates of radiation can be an ecological effective factor for trees, and we should take this into account for both forest management and tree species management (Volkova et al. 2017). Therefore, the effective methods for radiation protection should be developed based on the information on the effects on reference species, effects of different dose thresholds and effects on different types of reference ecosystems (Volkova et al. 2017). However, we still lack mechanistic understanding of the biological effects induced by the chronic exposure of low radiation dose rates and there are not currently sufficient field data about the real consequences of chronic radiation exposure for tree populations (Garnier-Laplace et al. 2013). Consequently, tree populations remain an important object for study of long-term biological consequences of radiation exposure after the nuclear crisis (Volkova et al. 2017). Needless to say, contamination level and species composition of Fukushima forests are quite different from forests around Chernobyl and those of Northern Europe. Therefore, we need to develop a long-term effective radiation protection approach for forest trees focused specifically on Japanese forest ecosystems.

Decontamination and monitoring of contamination level of forest ecosystems

According to the preliminary surveys conducted in August and September 2011 in Fukushima deciduous forests, the radiocesium concentration (total of ^{134}Cs and ^{137}Cs) in litter ranged from 24.1 to 319 kBq kg^{-1} and the proportion of radiocesium in the litter ranged from 22% to 66% of the total radiocesium in the forest ecosystems (Hashimoto et al. 2012). This observed high proportion of radiocesium in the litter was because the forest floor received a considerable level of radiocesium fallout without filtering by forest canopies as deciduous forests did not have leaves at the time of the FDNPP accident. Shortly after the incident, removing the contaminated litter could have been an efficient method of decontamination (Hashimoto et al. 2012). Although the dynamics of radiocesium is expected to differ depending on the amount of deposition, types of forest ecosystems, climatic and topographic conditions (Kliashtorin et al. 1994; Linkov and Schell 2012), the majority of ^{137}Cs would have already migrated to the mineral soil layer in Fukushima forests by 2012, suggesting that little ^{137}Cs remains within the litter layer in Fukushima forests. Therefore, removing the litter may now be ineffective for decontamination in the Fukushima forests (Hashimoto et al. 2012).

Although it may depend on the balance of costs and benefits, one option is to leave certain forest sites without decontamination and we should discuss this choice as one of the options (Hashimoto et al. 2012). After the Chernobyl accident, forest decontamination was considered as potentially labor-intensive and high-cost (IAEA 2011). If we were to conduct decontamination of forest ecosystems through removal of the contaminated litter, soil and/or trees, we need to first evaluate its negative effects on the forest ecosystem. Large-scale removal of trees and soils for decontamination may cause soil erosion and landslides, and may also lead to reduction of biodiversity and the degradation of soil nutrients and water retention functions. In conclusion, the potential reduction of radiation dose, feasibility, monetary cost, the potential negative ecological effects, and the social acceptability of both options (with or without decontamination) should be taken into account before a decision is made. However, in reality, the decision of whether to take the option of decontamination or no decontamination for a particular forest area will be governed by the ease of access and removal of contaminated materials (Guillitte and Willdrocht 1993; Guillitte et al. 1994; Fesenko et al. 2005; IAEA 2006; Hashimoto et al. 2012). Although we may decide not to decontaminate a forest site, this does not mean that we abandon efforts to manage the site. As a minimum requirement, we should monitor the contaminated forest ecosystem for a long period of time using the sampling methods proposed in this work.

What studies are needed in future

In this dissertation, I described the abovementioned sampling methods for the assessment of contamination levels for Japanese fir trees. However, the following items should be elaborated to further improve the assessments of ^{137}Cs contamination levels in natural forests in Fukushima Prefecture.

First, in Japanese fir trees, there is an uneven distribution of ^{137}Cs over tree body according to parts of the tree (needles, branches, wood and bark), age of the parts, and height of the parts. However, it remains unclear whether these conclusions are applicable to other tree species. Therefore, it may be necessary to assess the contamination levels by ^{137}Cs in other tree species than Japanese fir. Therefore, future work should consider the applicability of the results of the current research to other tree species. However, to achieve this, we would need to first invent a suitable method to identify ages of leaves for other tree species.

Second, I presented a method for the assessment of contamination levels by using Japanese fir trees in a small area (< 1 ha). However, Fukushima natural forests cover a far larger area than 1 ha. To assess the contamination level over the Fukushima natural forests, assessment is required of Fukushima forests other than the study sites. Based on the method provided for the assessment of contamination levels by using Japanese fir trees in a small area in the current work, we need to next make assessments for many forests as possible.

Komatsu et al. (2015) showed that contamination level by ^{137}Cs varied among five Fukushima forests depending on the form of initial ^{137}Cs deposition (dry or wet), weather conditions (rainfall) and forest structure (leaf biomass). Therefore, the best way to scale up a small area based assessment to the whole Fukushima natural forests assessment may be as follows. First, we classify Fukushima natural forests into groups using the criteria of the form of initial ^{137}Cs deposition (dry or wet), weather condition (rainfall) and forest structure (leaf biomass). We should then assess the contamination level by ^{137}Cs for every forest type using the method proposed in the current research. Finally, these assessment results should be combined for each forest type to scale up to cover all Fukushima forests. This design represents only a rough approach to upscaling the results of assessment of a small area to an assessment of all Fukushima natural forests. More studies are needed to construct a suitable upscaling method in future.

Finally, I only focused on small trees of a few meters in height. The question therefore arises whether the results are applicable to big, canopy Japanese fir trees. The parts, age, and height must be controlling factors for the distribution of ^{137}Cs for large, canopy-level Japanese fir trees. However, we need to check this using the methods described in Chapter 3.

In short, future research on contamination levels by ^{137}Cs in natural forests in Fukushima Prefecture should address the following three items:

(1) Clarification of the extent to which the results for Japanese fir trees are applicable for other tree species.

(2) Establishment of suitable methods to upscale the small-area-based assessment proposed by this study to an assessment of all Fukushima natural forests.

(3) To check whether the results are applicable for large canopy-forming Japanese fir trees.

References

Ahlswede J, Hebel S, Ross JO, Schoetter R, Kalinowski MB. 2013. Update and improvement of the global krypton-85 emission inventory. J Environ Radioact. 115:34–42.

Aragami K. 1987. Studies on the process of formation of Abies and Tsuga natural forest in the central mountain district of Kyushu. Bull. Kyushu Univ. For. 57:17–108. (in Japanese with English summary).

Baker DA. 1983. Uptake of cations and their transport within the plants. In: Robb, D.A. and Pierpoint, W.S. (eds.). Metals and Micronutrients: Uptake and Utilization by Plants. Academic Press, London. pp.3–19.

Bunzl K, Schimmack W, Kreutzer K, Schierl R. 1989. Interception and retention of Chernobyl-derived ^{134}Cs, ^{137}Cs and ^{106}Ru in a spruce stand. Sci Total Environ. 78:77–87.

Calmon P, Thiry Y, Zibold G, Rantavaara A, Fesenko S. 2009. Transfer parameter values in temperate forest ecosystems: a review. J Environ Radioact. 100:757–766.

Chino M, Nakayama H, Nagai H, Terada H, Katata G, Yamazawa H. 2011. Preliminary estimation of release amounts of ^{131}I and ^{137}Cs accidentally discharged from the Fukushima Daiichi nuclear power plant into the atmosphere. J Nucl Sci Technol. 48:1129–1134.

Cohen J. 1988. Statistical Power Analysis for the Behavioral Sciences, 2nd ed. Psychology Press, New York.

Cressie NAC. 1993. Statistics for Spatial Data Revised Edition. John Wiley & Sons, Inc., New York.

Davids C, Tyler AN. 2003. Detecting contamination-induced tree stress within the Chernobyl exclusion zone. Remote Sensing of Environment. 85:30–38.

Davis JJ. 1963. Cesium and its relationships to potassium in ecology. In: Schltz V, Klement Jr AW, editors. Radioecology. New York: Reinhold. pp.539–556.

Ertel J, Ziegler H. 1991. Cs-134/137 contamination and root uptake of different forest trees before and after the Chernobyl accident. Radiat Environ Biophysics. 30:147–157.

Fesenko SV, Voigt G, Spiridonov SI, Gontarenko IA. 2005. Decision making framework for application of forest countermeasures in the long term after the Chernobyl accident. J Environ Radioact. 82:143–166.

Fisher RA. 1966. The Design of Experiments, 8th ed. Oliver & Boyd, Edinburgh, London.

Fogh CL, Andersson KG. 2001. Dynamic behaviour of Cs-137 contamination in trees of the Briansk region, Russia. Sci Total Environ. 269:105–115.

Forestry Agency of Japan. 2017. [Internet]. The Results of Surveys of Distributions of Radioactive Elements in Forest Ecosystems. [In Japanese] [Accessed June 27 2017] Available from: http://www.rinya.maff.go.jp/j/kaihatu/jyosen/H28_jittaihaaku_kekka.html

Fortin MJ, Dale MRT. 2005. Spatial Analysis : a guide for ecologists. Cambridge University Press, Cambridge, N.Y.

Garnier-Laplace J, Geras'kin S, Della-Vedova C, Beaugelin-Seiller K, Hinton TG, Real A, Oudalova A. 2013. Are radiosensitivity data derived from natural field conditions consistent with data from controlled exposures? A case study of Chernobyl wildlife chronically exposed to low dose rates. J Environ Radioact. 121:12–21.

Gonze MA, Calmon P. 2017. Meta-analysis of radiocesium contamination data in Japanese forest trees over the period 2011–2013. Sci Total Environ. 601: 301–316.

Goor F, Thiry Y. 2004. Processes, dynamics and modelling of radiocaesium cycling in a chronosequence of Chernobyl-contaminated Scots pine (*Pinus sylvestris* L.) plantations. Sci Total Environ. 325:163–180.

Guillitte O, Tikhomirov FA, Shaw G, Vetrov V. 1994. Principles and practices of countermeasures to be carried out following radioactive contamination of forest areas. Sci Total Environ. 157:399–406.

Guillitte O, Willdrocht C. 1993. An assessment of experimental and potential countermeasures to reduce radionuclide transfers in forest ecosystems. Sci Total Environ. 137:273–288.

Hashimoto S, Ugawa S, Nank, K, Shichi K. 2012. The total amounts of radioactively contaminated materials in forests in Fukushima, Japan. Sci Rep. 2:416.

Hayama SI, Nakiri S, Nakanishi S, Ishii N, Uno T, Kato T, Konno F, Kawamoto Y, Tsuchida S, Ochiai K, Omi T. 2013. Concentration of radiocesium in the wild Japanese monkey (*Macaca fuscata*) over the first 15 months after the Fukushima Daiichi Nuclear Disaster. PloS One 8(7): e68530. doi: 10.1371/journal.pone.0068530.

Hou X, Povinec PP, Zhang L, Shi K, Biddulph D, Chang C-C, et al. 2013. Iodine-129 in seawater offshore Fukushima: distribution, inorganic speciation, sources, and budget. Environ Sci Technol. 47:3091–3098.

IAEA. 2006. Environmental consequences of the Chernobyl accident and their remediation: twenty years of experience report of the Chernobyl forum expert group 'environment'. Vienna. Radiol. Assess. Rep. Ser.

IAEA. 2011. [Internet]. Summary Report of the Preliminary Findings of the IAEA Mission on Remediation of Large Contaminated Areas Off-Site the Fukushima Dai-ichi NPP. [cited 2017 May 1]. Available from:https://www.iaea.org/sites/default/files/preliminaryfindings2011.pdf

Ipatyev V, Bulavik I, Baginsky V, Goncharenko G, Dvornik A. 1999. Forest and Chernobyl: forest ecosystems after the Chernobyl nuclear power plant accident: 1986-1994. J Environ Radioact. 42:9–38.

Japan Meteorological Agency. [Internet]. The 2011 Great East Japan Earthquake -Portal-. [cited 2017 May 1]. Available from:

http://www.jma.go.jp/jma/en/2011_Earthquake/2011_Earthquake.html

Japan Meteorological Agency [Internet]. Weather, Climate & Earthquake Information, 2016. [cited 2016 Jul 6]. Available from: http://www.data.jma.go.jp/obd/stats/etrn/view/nml_amd_ym.php?prec_no=36&block_no=1130&year=&month=&day=&view=.

Kabata-Pendias A. 2011. Trace elements in soils and plants. 4th edition. Boca Raton, USA: CRC Press, Taylor & Francis. pp.128–133.

Kaji M. 1975. Studies on the Ecological Status of *Abies firma* Forest in the Boso Peninsula. Bulletin of Tokyo university Forests. 68:1–23.

Kanasashi T, Sugiura Y, Takenaka C, Hijii N, Umemura M. 2015. Radiocesium distribution in sugi (Cryptomeria japonica) in Eastern Japan: translocation from needles to pollen. J Environ Radioact. 139:398–406.

Kato H, Onda Y, Hisadome K, Loffredo N, Kawamori A. 2015. Temporal changes in radiocesium deposition in various forest stands following the Fukushima Dai-ichi Nuclear Power Plant accident. J Environ Radioact. 166:449–457.

Kaunisto S, Aro L, Rantavaara A. 2002. Effect of fertilisation on the potassium and radiocaesium distribution in tree stands (*Pinus sylvestris* L.) and peat on a pine mire. Environ Pollut. 117:111–119.

Kenzo T. 2009. *Momi*. J. Jpn. Soc. Reveget. Tech. 34(4).659.

Kliashtorin AL, Tikhomirov FA, Shcheglov AI. 1994. Vertical radionuclide transfer by infiltration water in forest soils in the 30-km Chernobyl accident zone. Sci Total Environ. 157:285–288.

Kohyama T. 1980. Growth pattern ofAbies mariesii saplings under conditions of open-growth and suppression. Botanical Magazine Tokyo 9:13–24.

Komatsu M, Kaneko S, Ohashi S, Kuroda K, Sano T, Ikeda S, Saito S, Kiyono Y, Tonosaki M, Miura S, et al. 2016. Characteristics of initial deposition and behavior of radiocesium in forest ecosystems of different locations and species affected by the Fukushima Daiichi Nuclear Power Plant accident. J Environ Radioact. 161:2–10.

Kubota Y, Takahashi H, Watanabe Y, Fuma S, Kawaguchi I, Aoki M, Kubota M, Furuhata Y, Shigemura Y, Yamada F, Ishikawa T, Obara S, Yoshida S. 2015. Estimation of absorbed radiation dose rates in wild rodents inhabiting a site severely contaminated by the Fukushima Dai-ichi nuclear power plant accident. J Environ Radioact. 142:124–131.

Kuroda K, Kagawa A, Tonosaki M. 2013. Radiocesium concentrations in the bark, sapwood and heartwood of three tree species collected at Fukushima forests half a year after the Fukushima Dai-ichi nuclear accident. J Environ Radioact. 122:37–42.

Legendre P. 1993. Spatial autocorrelation: trouble or new paradigm? Ecology. 74:1659–1673.

Legendre P, Dale MRT, Fortin MJ, Gurevitch J, Hohn M, Myers D. 2002. The consequences of spatial structure for the design and analysis of ecological field surveys. Ecography. 25:601–615.

Legendre P, Legendre L. 1998. Numerical Ecology: Second English edition. Elsevier Science B. V., Amsterdam.

Linkov I, Schell WR. (Eds.). 2012. Contaminated forests: Recent developments in risk identification and future perspectives (Vol. 58). Springer Science & Business Media.

Maher BA, Moore C, Matzka J. 2008. Spatial variation in vehicle-derived metal pollution identified by magnetic and elemental analysis of roadside tree leaves. Atmospheric Environment. 42:364–373.

Mamikhin SV, Tikhomirov FA, Shcheglov AI. 1997. Dynamics of Cs-137 in the forests of the 30-km zone around the Chernobyl nuclear power plant. Sci Total Environ. 193:169–177.

Matsui S, Kasahara S, Morimoto G, Mikami OK, Watanabe M, Ueda K. 2015. Radioactive contamination of nest materials of the Eurasian Tree Sparrow Passer montanus due to the Fukushima nuclear accident: The significance in the first year. Environmental Pollution. 206:159–162.

McGee EJ, Synnott HJ, Johanson KJ, Fawaris BH, Nielsen SP, Horrill AD, Kennedy VH, Barbayiannis N, Veresoglou DS, Dawson DE, Colgan PA, McGarry AT. 2000. Chernobyl fallout in a Swedish spruce forest ecosystem. J Environ Radioact. 48:59–78.

Melin J, Wallberg L, Suomela J. 1994. Distribution and retention of cesium and strontium in Swedish boreal forest ecosystems. Sci Total Environ. 157:93–105.

Ministry of Education, Culture, Sports, Science and Technology Japan [MEXT]. 2011. [Internet]. Results of the third airborne monitoring survey by MEXT [cited 2016 Dec 1]. Available at: http://www.radioactivity.nsr.go.jp/en/contents/5000/4182/24/1304797_0708e.pdf.

Mizunaga H, Ishii H, Tsujino S, Haraguchi A, Kawabata Y. 1998. The annual variation of seed production and seedling density at *Abies*-*Tsuga* forest in Kirishima. Bull. Kagoshima Univ. For. 26:23–31. (in Japanese with English summary).

Moreno E, Sagnotti L, Dinarès-Turell J, Winkler A, Cascella A. 2003. Biomonitoring of traffic air pollution in Rome using magnetic properties of tree leaves. Atmospheric Environment. 37:2967–2977.

Moller AP, Hagiwara A, Matsui S, Kasahara S, Kawatsu K, Nishiumi I, Suzuki H, Ueda K, Mousseau TA. 2012. Abundance of birds in Fukushima as judged from Chernobyl. Environmental Pollution. 164:36–39.

Morino Y, Ohara T, Nishizawa M. 2011. Atmospheric behavior, deposition, and budget of radioactive materials from the Fukushima Daiichi nuclear power plant in March 2011. Geophysical research letters. 38:7.

National Police Agency. 2016. [Internet]. The damage and steps of the 2011 Great East Japan Earthquake. [cited 2017 May 1]. Available from:

https://www.npa.go.jp/archive/keibi/biki/higaijokyo.pdf

Nishikiori T, Watanabe M, Koshikawa MK, Takamatsu T, Ishii Y, Ito S, Takenaka A, Watanabe K, Hayashi S. 2015. Uptake and translocation of radiocesium in cedar leaves following the Fukushima nuclear accident. Sci Total Environ. 502:611–616.

Ogawa N, Yoshida S, Mizoue N, Murakami T. 2006. The relationship between growth of *Abies firma* saplings and light environment in planted Japanese cypress （*Chamaecyparis obtusa*） plantations at Kirishima mountains. Bulletin of Kyushu J. For. Res. 59:147–149.

Papastefanou C, Manolopoulou M, Sawidis T. 1989. Lichens and mosses – biological monitors of radioactive fallout from the Chernobyl reactor accident. J Environ Radioact. 9:199–207.

Petersen RG, Calvin LD. 1986. Sampling. Methods of Soil Analysis, Part 1, Physical and Mineralogical Methods 2nd ed. American Society of Agronomy-Soil Science Society of America, Wisconsin.

Raitio H, Rantavaara A. 1994. Airborne Radiocesium in Scots pine and Norway spruce needles. Sci Total Environ. 157:171–180.

Sawidis T, Marnasidis A, Zachariadis G, Stratis J. 1995. A study of air pollution with heavy metals in Thessaloniki city (Greece) using trees as biological indicators. Archives of Environmental Contamination and Toxicology. 28:118–124.

Science Council of Japan Great East Japan Earthquake Task Force, 2011. [Internet] Scientific Survey and Analysis of Movement of Radioactive Substances over a Wide Area. The 7th emergency recommendation regarding the response to the Great East Japan Earthquake. [cited 2016 June 28]. Available from: http://www.scj.go.jp/ja/info/jishin/pdf/t-110803e.pdf

Shizuma K, Oba Y, Takada M. 2016. A practical method for determining γ-ray full-energy peak efficiency considering coincidence-summing and self-absorption corrections for the measurement of environmental samples after the Fukushima reactor accident. Nucl Instrum Methods Phys Res B. 383:183–190.

Sombré L, Vanhouche M, Debrouwer S, Ronneau C, Lombotte JM, Myttenaere C. 1994. Long-term radiocesium behavior in spruce and oak forests. Sci Total Environ. 157:59–71.

Soukhova NV, Fesenko SV, Klein D, Spiridonov SI, Sanzharova NI, Badot PM. 2003. Cs-137 distribution among annual rings of different tree species contaminated after the Chernobyl accident. J Environ Radioact. 65:19–28.

Strebl F, Gerzabek MH, Bossew P, Kienzl K. 1999. Distribution of radiocaesium in an Austrian forest stand. Sci Total Environ. 226:75–83.

Steinhauser G, Brandl A, Johnson TE. 2014. Comparison of the Chernobyl and Fukushima nuclear accidents: a review of the environmental impacts. Sci Total Environ. 470:800-817.

Stohl A, Seibert P, Wotawa G. 2012. The total release of xenon-133 from the Fukushima Dai-ichi nuclear power plant accident. J Environ Radioact. 112:155–159.

Suzuki K. 1992. Fluctuation of Momi (*Abies firma*) dead standing trees and change of annual ring width at Mt. Ohyama and the around areas in Kanagawa Pref. Bulletin of Kanagawa Pref. Forest Experimental Station. 19:23–42.

Thiry Y, Colle C, Yoschenko V, Levchuk S, Van Hees M, Hurtevent P, Kashparov V. 2009. Impact of Scots pine (*Pinus sylvestris* L.) plantings on long term ^{137}Cs and ^{90}Sr recycling from a waste burial site in the Chernobyl Red Forest. J Environ Radioact. 100:1062–1068.

Thiry Y, Goor F, Riesen T. 2002. The true distribution and accumulation of radiocaesium in stem of Scots pine (Pinus sylvestris L.). J Environ Radioact. 58:243–259.

Tikhomirov FA, Shcheglov AI. 1994. Main investigation results on the forest radioecology in the Kyshtym and Chernobyl accident zones. Sci Total Environ. 157:45–57.

Tobler WR. 1970. A computer movie simulating urban growth in the Detroit region. Economic Geography. 46:234–240.

Tomašević M, Rajšić S, Đorđević D, Tasić M, Krstić J, Novaković V. 2004. Heavy metals accumulation in tree leaves from urban areas. Environmental Chemistry Letters. 2:151–154.

Tokyo Electric Power Company. [Internet]. [cited 2017 May 1]. Available from: http://www.tepco.co.jp/nu/fukushima-np/outline/index-j.html

UNSCEAR [United Nation Scientific Committee on the Effects of Atomic Radiation]. 2008. Report to the General Assembly with Scientific Annexes. Volume II, Scientific Annex E. Effect of ionizing radiation on non-human biota.

UNSCEAR [United Nations Scientific Committee on the Effects of Atomic Radiation]. 2014. [Internet]. Sources, effects and risks of ionizing radiation. UNSCEAR 2013 Report to the General Assembly with Scientific Annexes (Volume I, Scientific Annex A). [cited 2017 May 1]. Available from: http://www.unscear.org/docs/publications/2013/UNSCEAR_2013_Report_Vol.I.pdf

Volkova PY, Geras'kin SA, Kazakova EA. 2017. Radiation exposure in the remote period after the Chernobyl accident caused oxidative stress and genetic effects in Scots pine populations. Scientific Reports. 7.

Wagner G. 1993. Large-scale screening of heavy metal burdens in higher plants. In: Markert, B. (ed.). Plants as Biomonitors. Indicators for Heavy Metals in the Terrestrial Environment. VCH, Weinheim. pp.425–434.

Yoshida N, Takahashi Y. 2012. Land-surface contamination by radionuclides from the Fukushima Daiichi Nuclear Power Plant accident. Elements. 8:201–206.

Yoshida S, Muramatsu Y, Dvornik AM, Zhuchenko TA, Linkov I. 2004. Equilibrium of radiocesium with stable cesium within the biological cycle of contaminated forest ecosystems. J Environ Radioact. 75:301–313.

Yoshida S, Watanabe M, Suzuki A. 2011. Distribution of radiocesium and stable elements within a pine tree. Radiat Prot Dosim. 146:326–329.

Yoshihara T, Matsumura H, Hashida SN, Nagaoka T. 2013. Radiocesium contaminations of 20 wood species and the corresponding gamma-ray dose rates around the canopies at 5 months after the Fukushima nuclear power plant accident. J Environ Radioact. 115:60–68.

Yoshihara T, Matsumura H, Tsuzaki M, Wakamatsu T, Kobayashi T, Hashida SN, Nagaoka T, Goto F. 2014. Changes in radiocesium contamination from Fukushima in foliar parts of 10 common tree species in Japan between 2011 and 2013. J Environ Radioact. 138:220–226.

Yuruki T, Aragami K. 1973. Studies on natural regeneration of Momi (*Abies firma* S. et Z., Japanese fir) and Tsuga (*Tsuga Sieboldii* Carr., Japanese hemlock). Bull. Kyushu Univ. For. 47:77–124. (in Japanese with English summary).

Zheng J, Tagami K, Watanabe Y, Uchida S, Aono T, Ishii N, et al. 2012. Isotopic evidence of plutonium release into the environment from the Fukushima DNPP accident. Sci Rep. 2:304.

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Figures and Tables

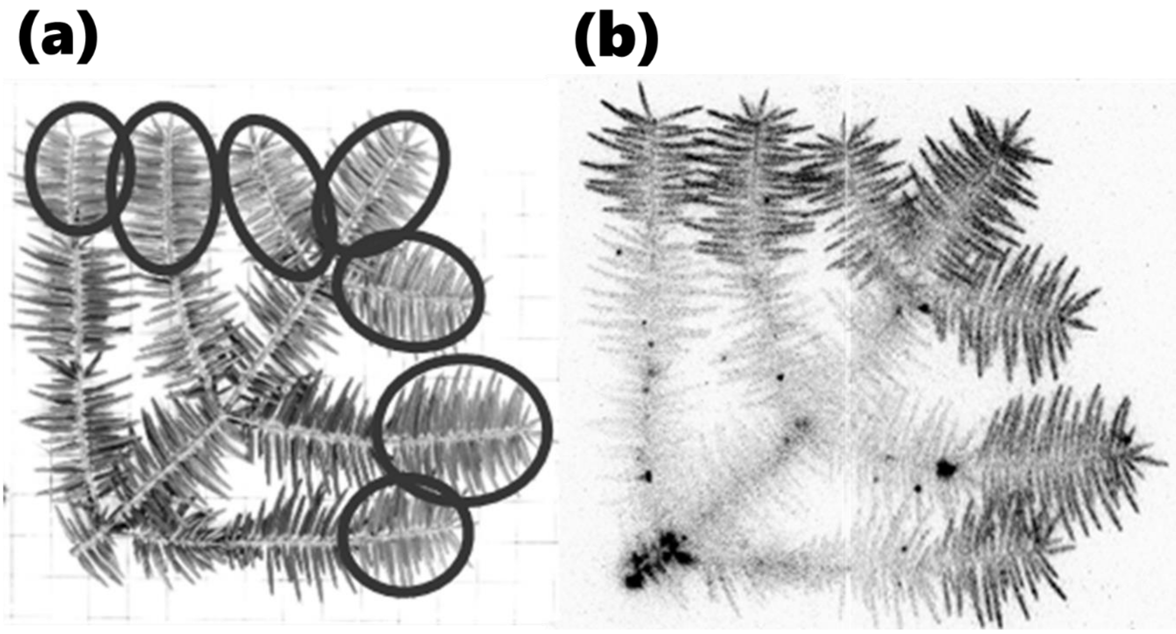


Figure 1. Radioactive distribution in shoots of *A. firma*. (a) Photograph of samples. Circles show the current-year shoots in 2013. (b) Radioactive distribution image obtained using imaging plates. In the image, radioactivity increases from white to black.

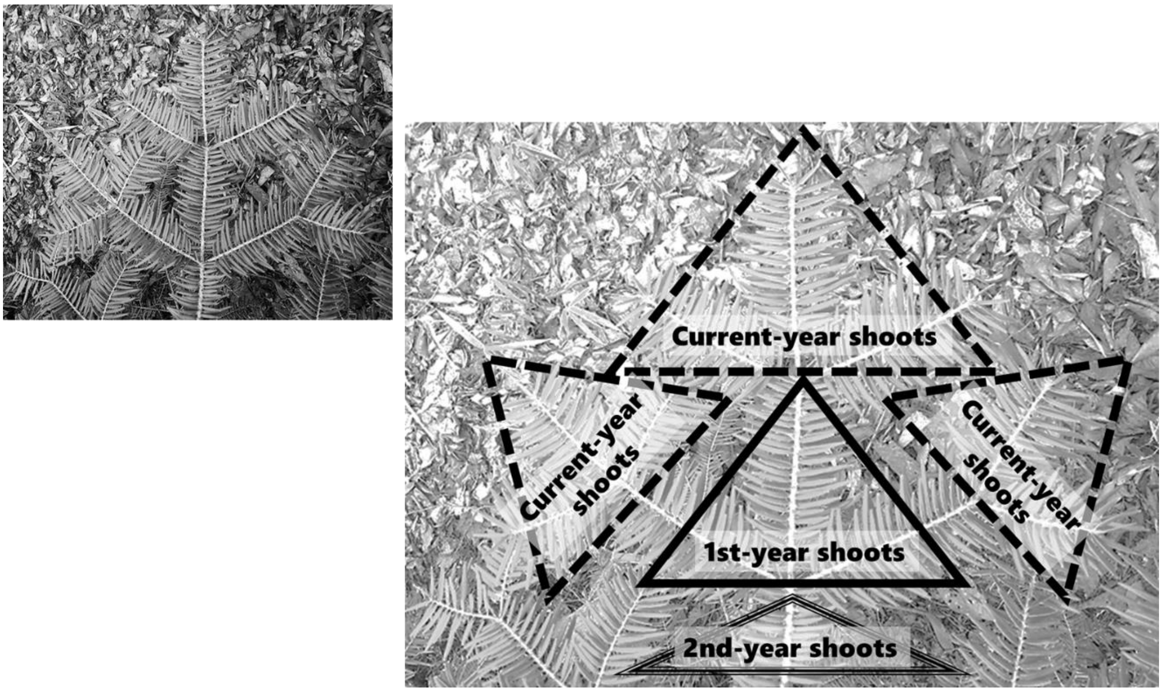
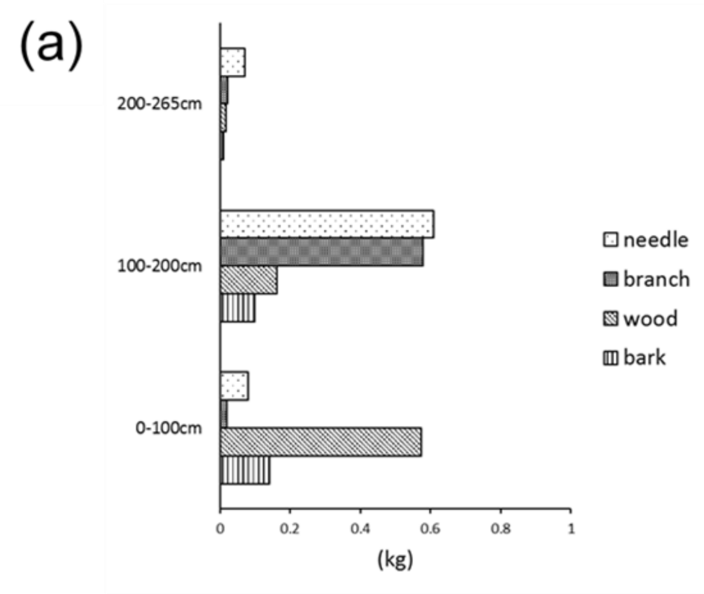


Figure 2-1. Typical branch of *A. firma*. Age of shoots can be recognized by the branch morphology of this species.



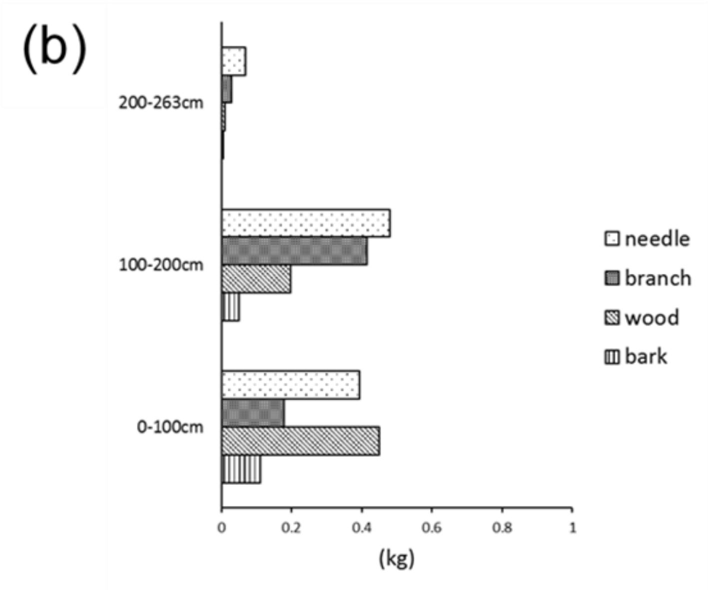


Figure 2-2. Dry mass of needles, branches, wood, and bark from each sampled *A. firma* tree. (a) Sample tree S1; (b) Sample tree S2.

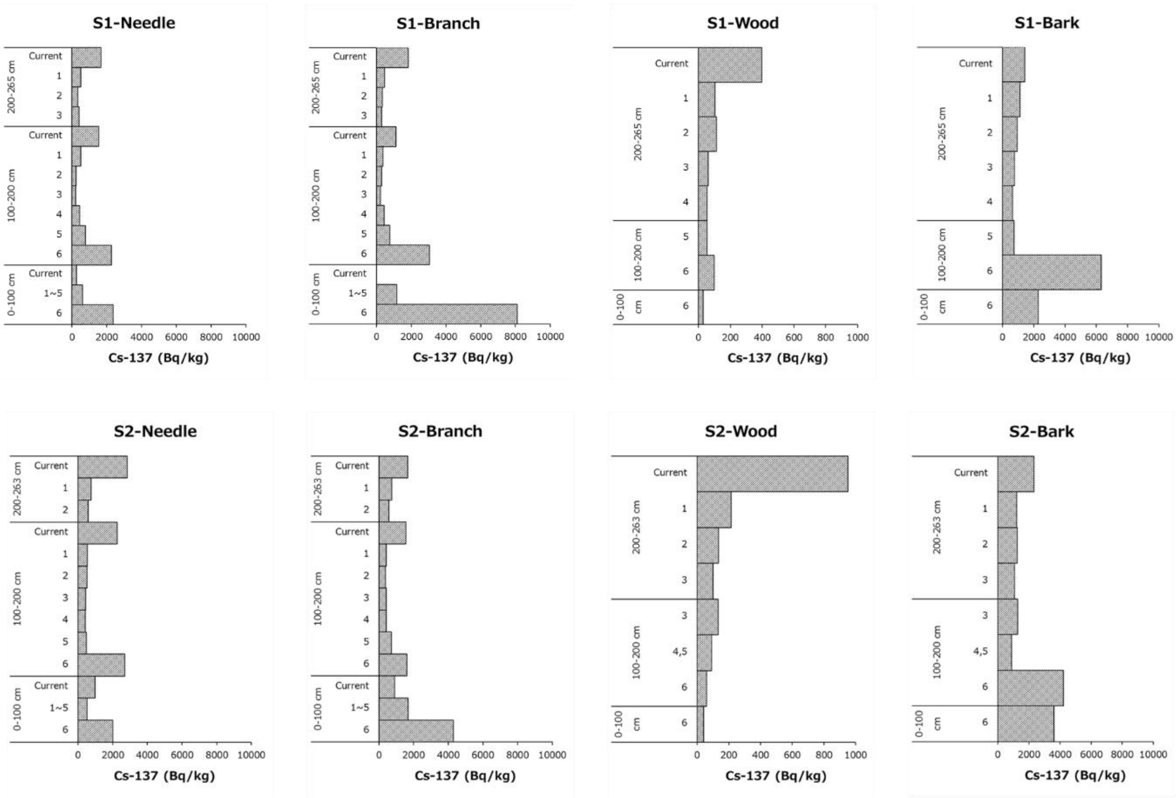


Figure 2-3. ^{137}Cs concentration in needles, branches, bark, and wood of sampled *A. firma* trees. Upper line, S1 tree; lower line, S2 tree.

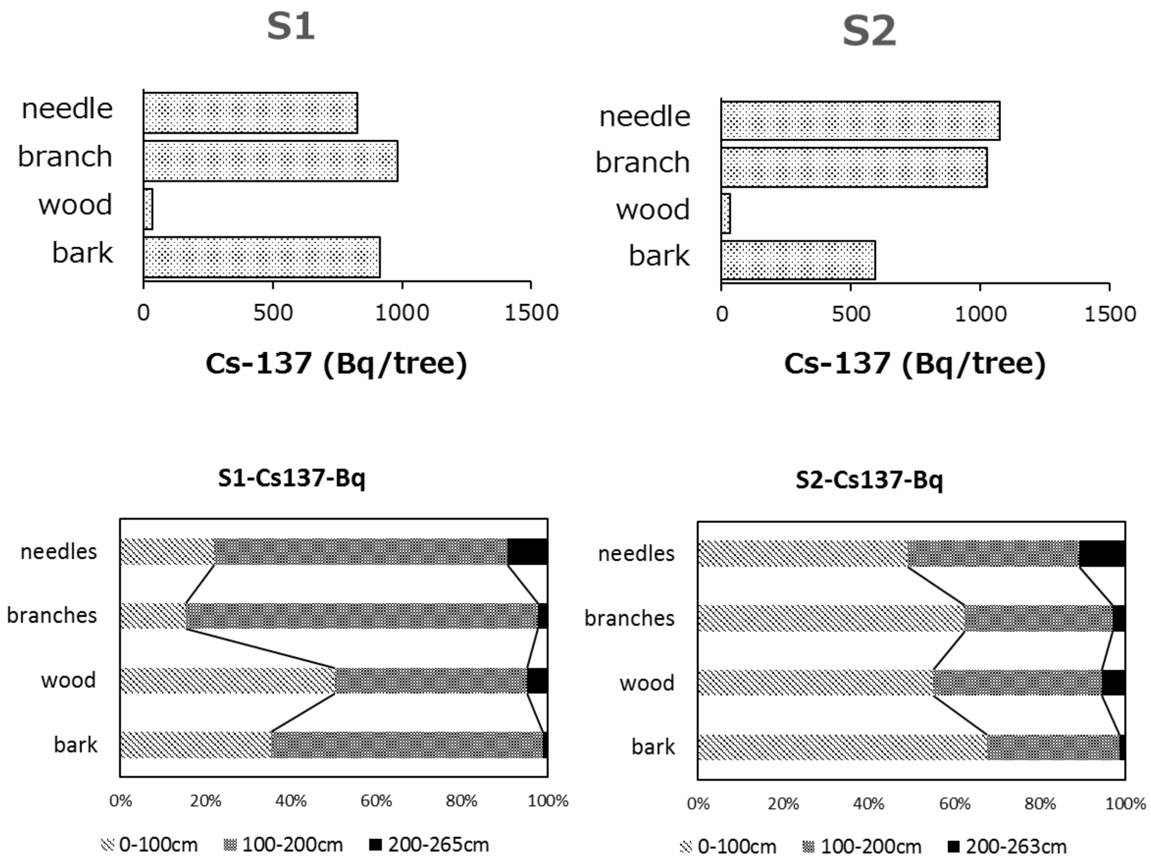


Figure 2-4. Total amount of ^{137}Cs in each part of S1 and S2 trees (upper panels). Total ^{137}Cs amount was determined by multiplying ^{137}Cs concentration in each part by biomass of that part. Lower panels: proportion of total ^{137}Cs in each tree part in three different height classes (0 – 100 cm; 100 – 200 cm; > 200 cm).

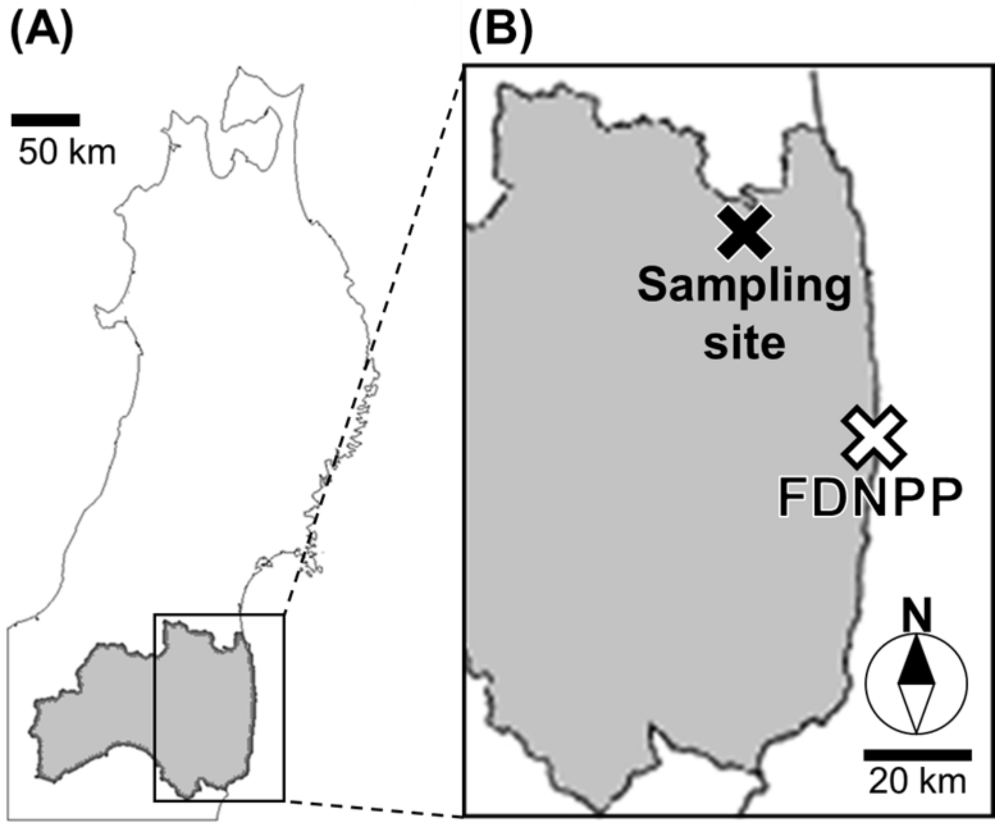


Figure 3-1. Map of the sampling location and the Fukushima Daiichi Nuclear Power Plant (FDNPP) in Fukushima, Japan. Fukushima Prefecture is shown in gray.

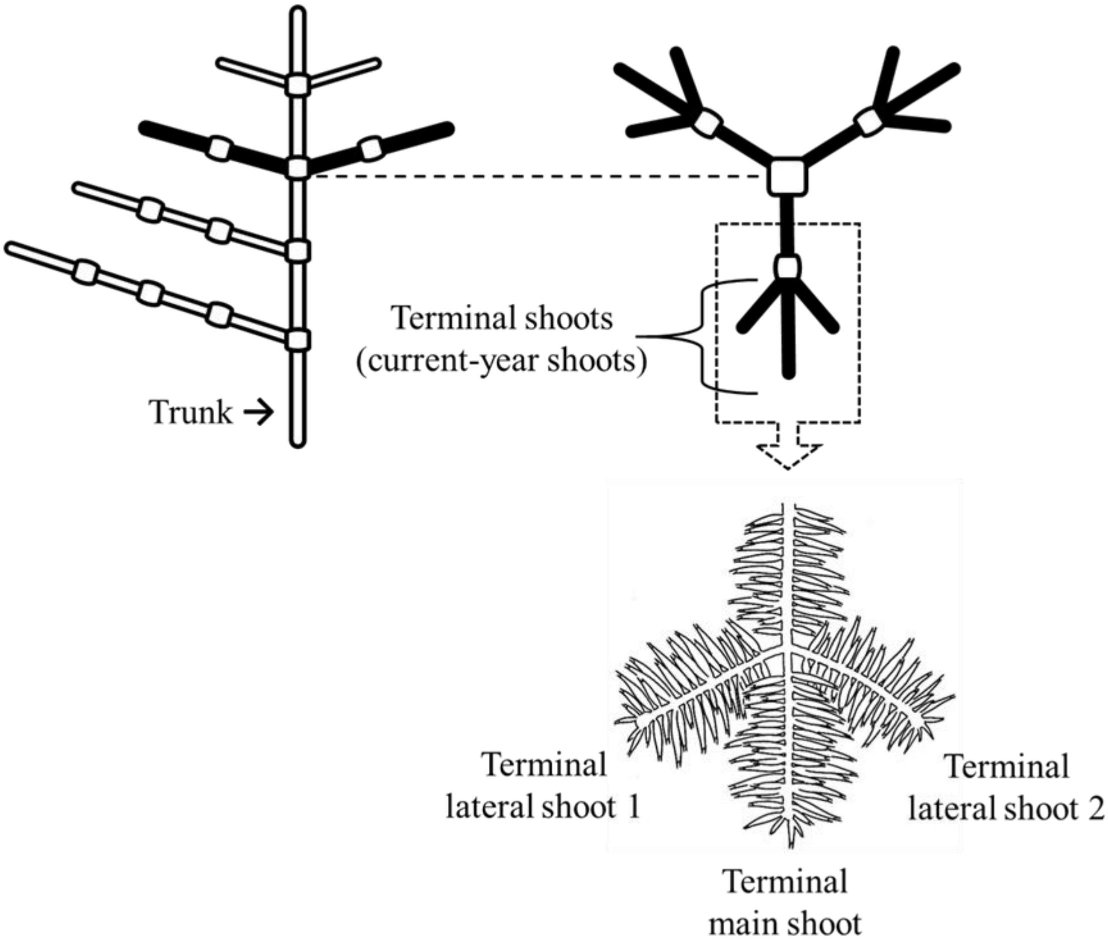


Figure 3-2. Morphological description of sampled young *A. firma* tree. Left figure shows side view of tree and right figures show views from directly above. Open squares show nodes (= scars of buds). Black bars represent the lateral shoots of the previous-year terminal trunk.

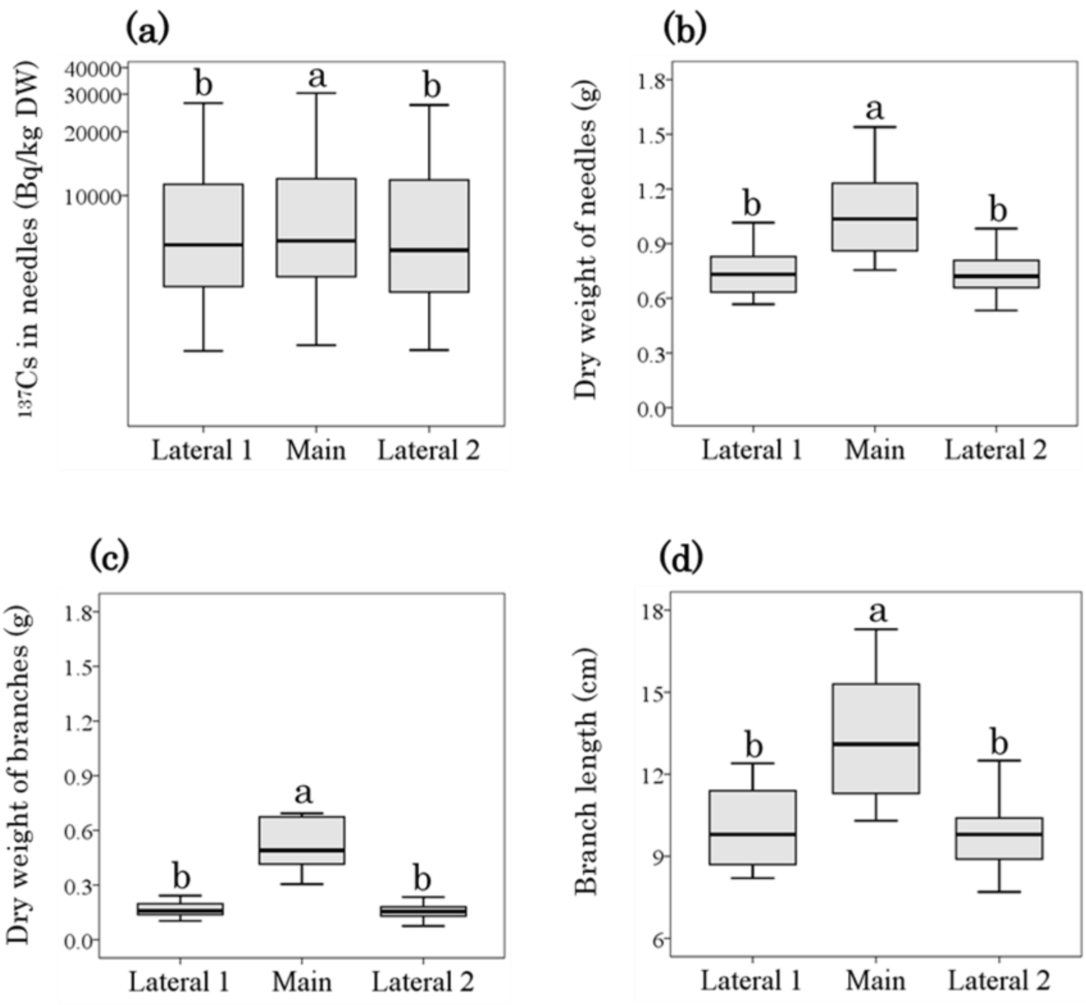


Figure 3-3. Box plots of (a) the ^{137}Cs concentration in needles, (b) the dry weight of needles, (c) the dry weight of branches, (d) the length of branches of the main and two lateral shoots. Different letters above the boxes show significant differences ((a); $p < 0.01$, (b), (c), (d); $p < 0.001$) between the values of each shoot group based on a one-way repeated measure ANOVA with Bonferroni correction. The line in the middle of the boxes shows the median value. The bottom of the box indicates the 25th percentile. The top of the box represents the 75th percentile. Approximately 95% of the data are expected to lie between the whiskers.

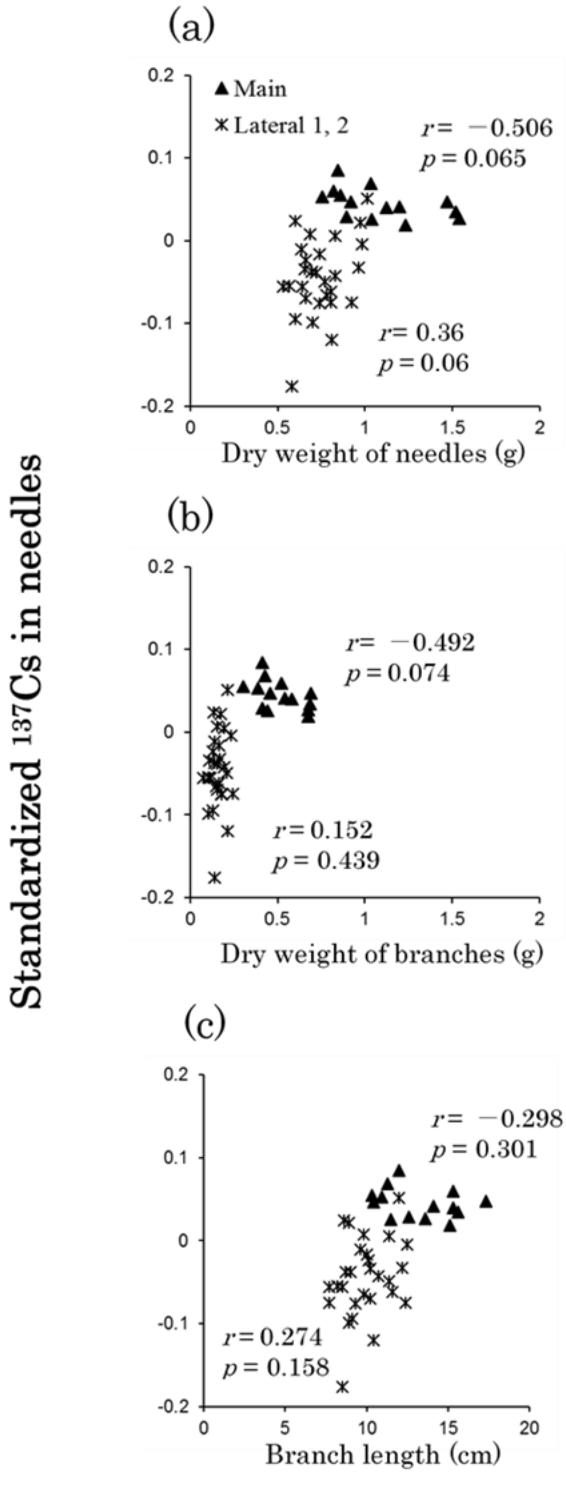


Figure 3-4. Relationships between the standardized ^{137}Cs in the needles of each shoot and (a) the dry weight of needles, (b) the dry weight of branches, and (c) the length of branches. The values (*r* and *p*) in each graph show the results of the Pearson's correlation coefficient.

Table 3-1. Results of the pairwise correlations between the morphological values of each shoot. The values show the r (Pearson's correlation coefficient).

	Main shoot	Lateral shoot 1	Lateral shoot 2
Needle DW × Branch DW	0.877**	0.829**	0.808**
Needle DW × Branch length	0.658*	0.67**	0.757**
Branch DW × Branch length	0.858**	0.780**	0.668**

(*: $p < 0.05$, **: $p < 0.01$)

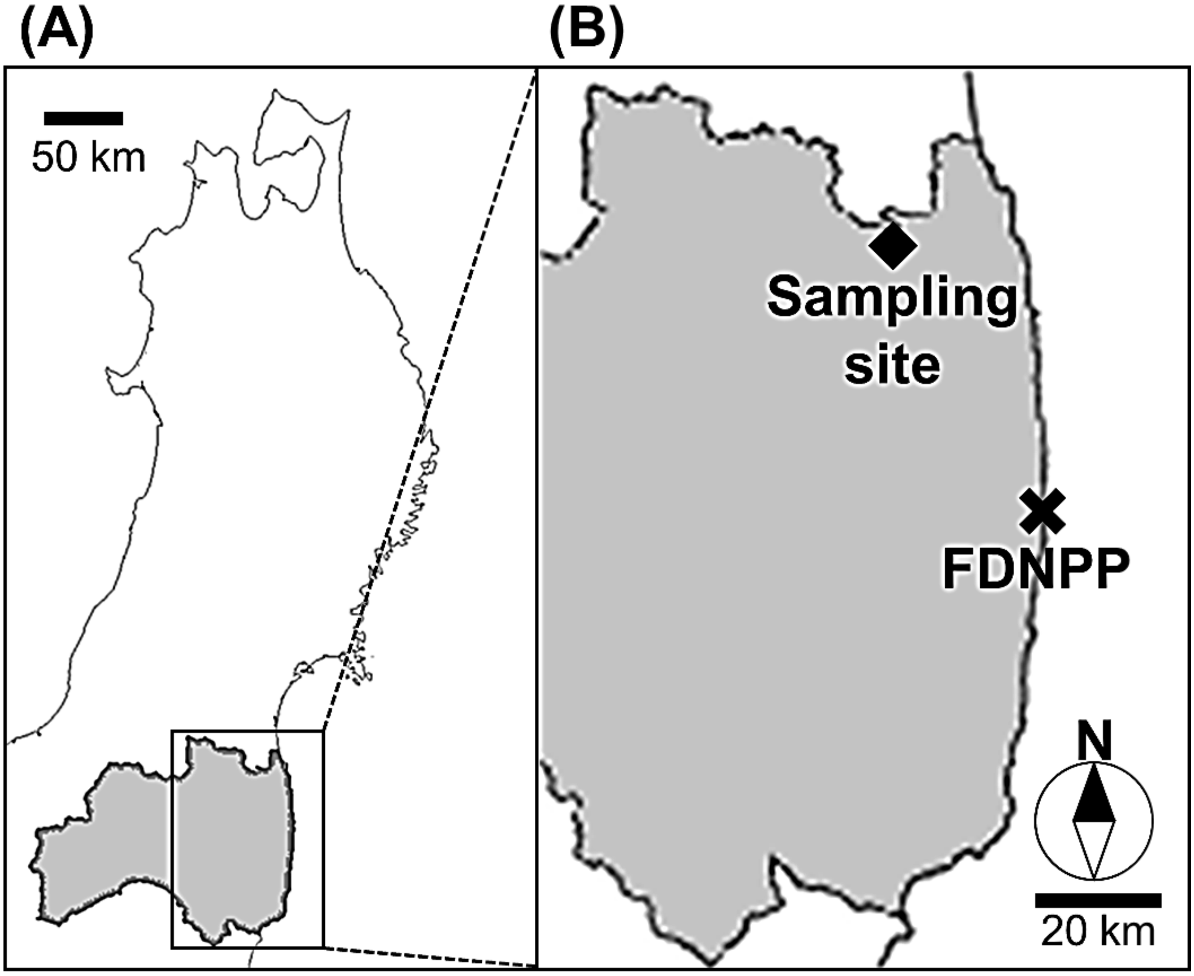


Figure 4-1. Maps showing the location of the sampling site and the Fukushima Daiichi Nuclear Power Plant (FDNPP), Fukushima, Japan. Gray parts represent Fukushima Prefecture.

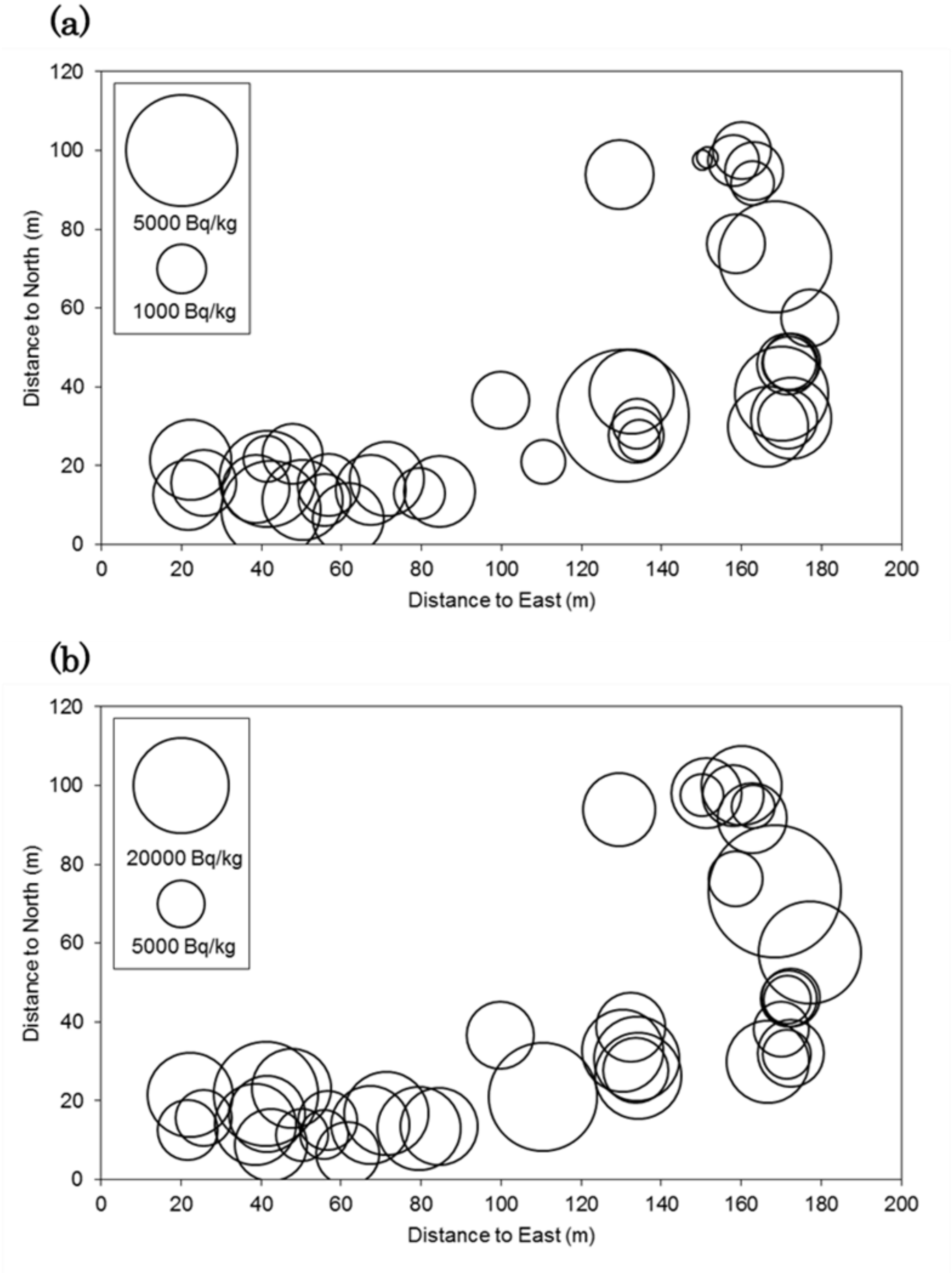


Figure 4-2. The locations of sampled trees of *A. firma* in the study area (n = 40). The size of circles shows (a) the concentration of ^{137}Cs in current year needles of lateral branches of annual trunk growing in 2015, (b) the concentration of ^{137}Cs in soil (0-5 cm) under the trees. Note that the size of circles showed different concentration of ^{137}Cs between (a) and (b).

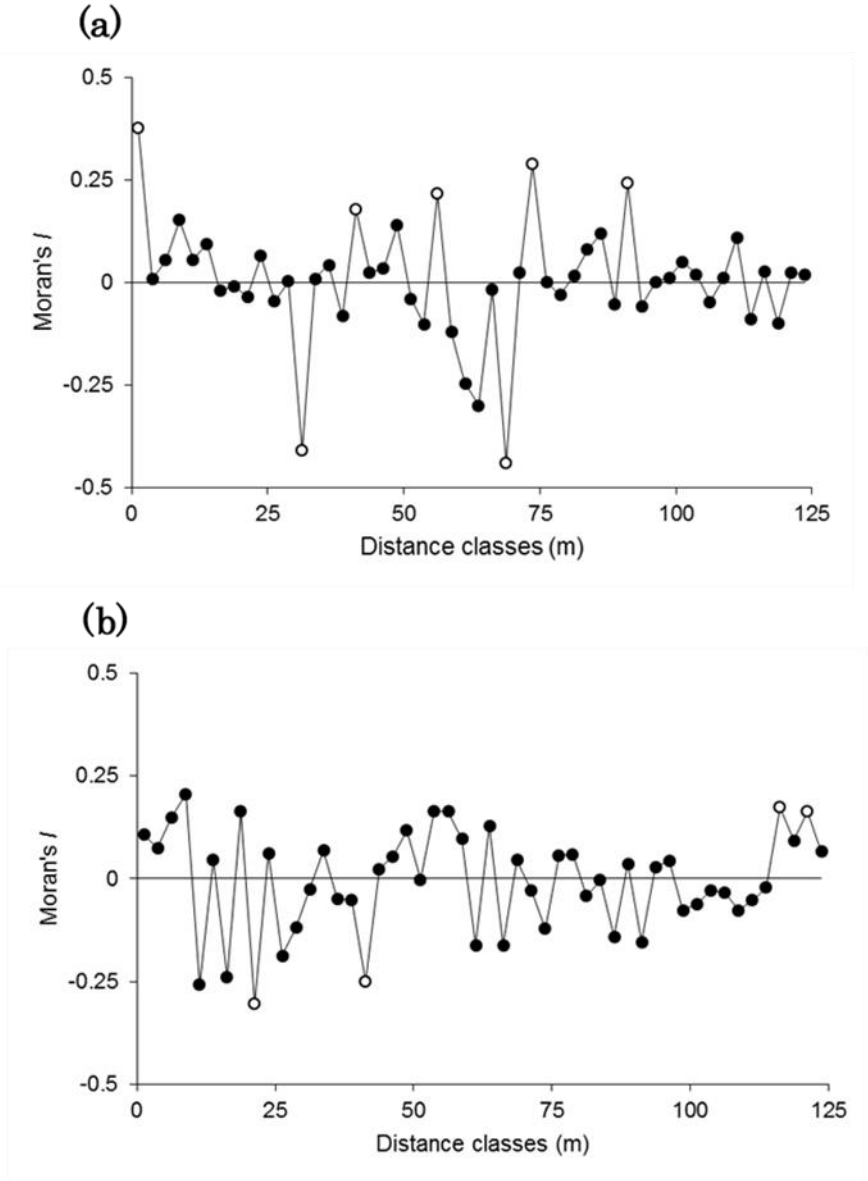


Figure 4-3. Spatial correlograms of the concentration of ^{137}Cs in (a) needles of lateral branches of annual trunks, and (b) soil under the sampling trees. Open circles show significant Moran's I statistics (p < 0.01) and closed circles show nonsignificant ones. Significance of Moran's I statistics was obtained by the randomization test of 5000 permutations. Distance class is at intervals of 2.5 m. Straight line shows Moran's $I = 0$.

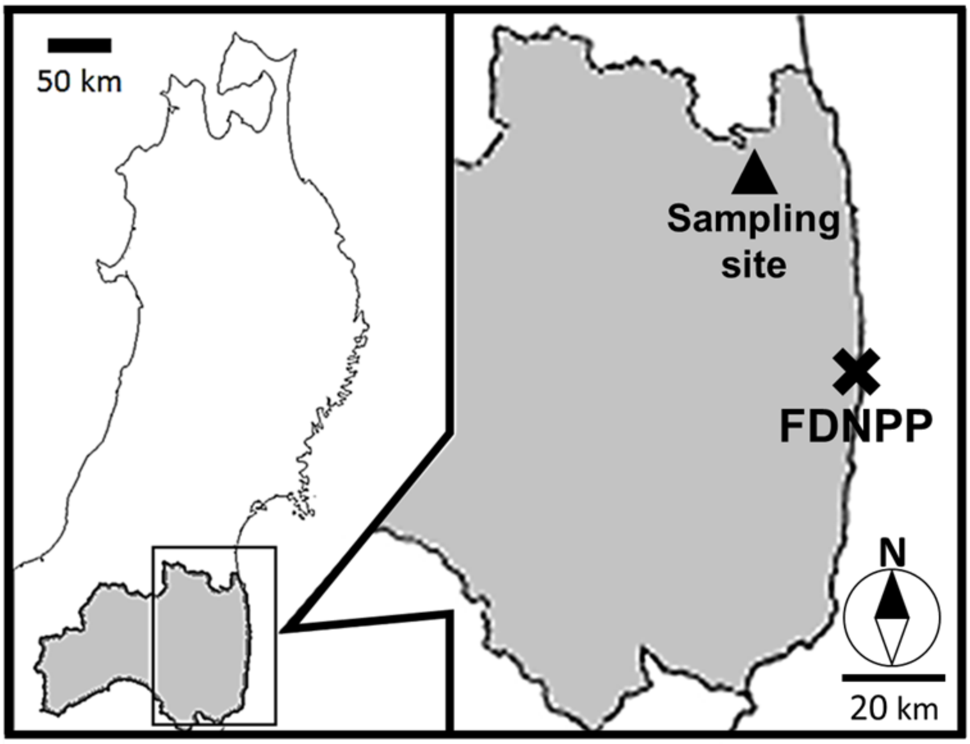


Figure 5-1. The locations of the sampling site and the Fukushima Daiichi Nuclear Power Plant (FDNPP), in Fukushima, Japan. Shaded areas show Fukushima Prefecture.

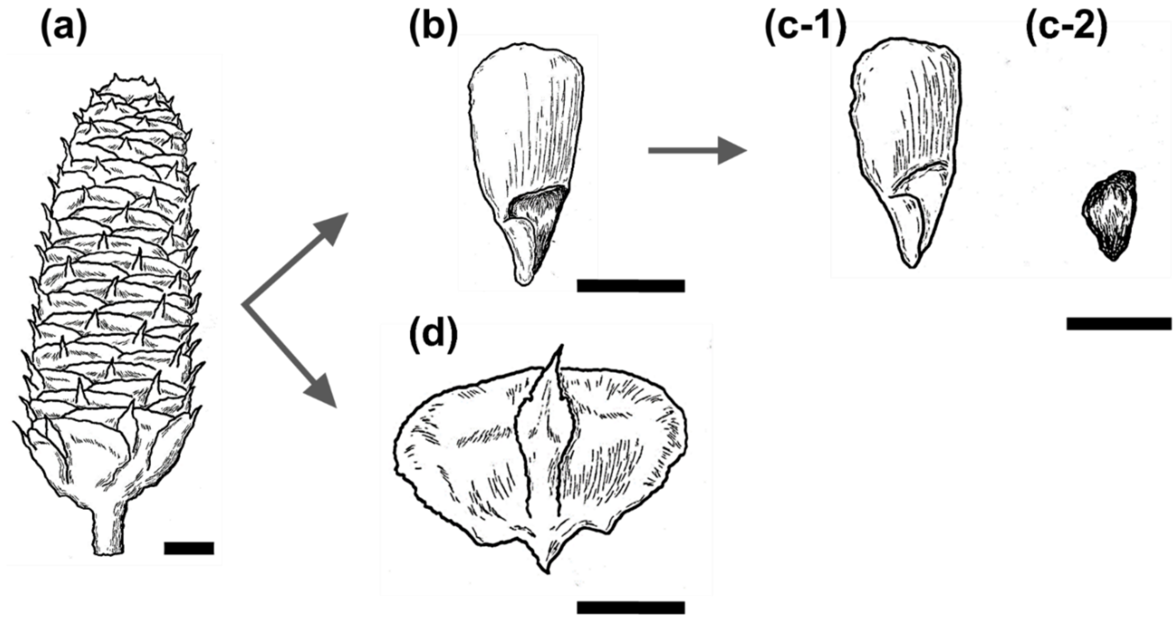


Figure 5-2. Description of the separation of cone samples. (a) cones, (b) winged seeds, (c-1) wings, (c-2) seeds, and (d) cone scales. The scale bar is 1 cm.

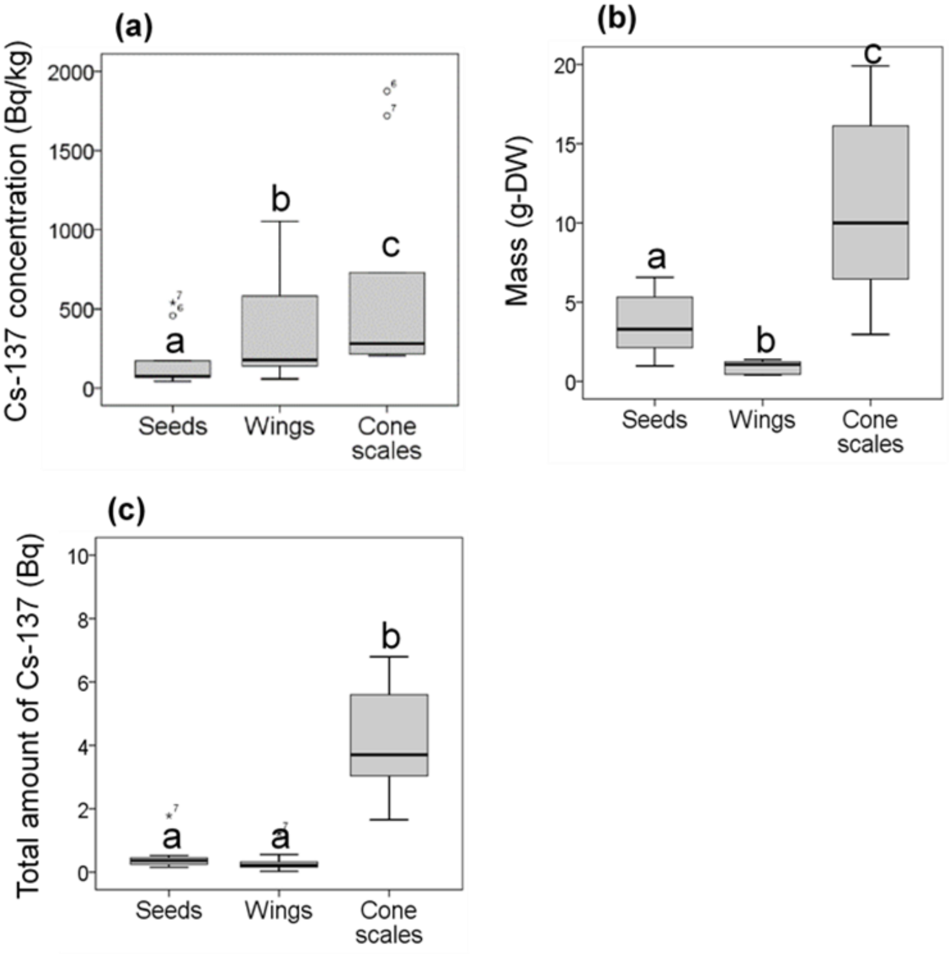


Figure 5-3. Box plots of (a) the ^{137}Cs concentration in seeds, wings and cone scales of nine cones, (b) the dry mass of seeds, wings and cone scales of nine cones, and (c) the total amount of ^{137}Cs in seeds, wings and cone scales of nine cones. Different letters above the boxes show significant differences ((a); $p < 0.05$, (b) $p < 0.01$, (c) $p < 0.01$) based on Friedman test followed by sequentially rejective Bonferroni. The line within the boxes represents the median value. The bottom of the box shows the 25th percentile. The top of the box indicates the 75th percentile. Approximately 95% of the data are expected to lie between the whiskers.

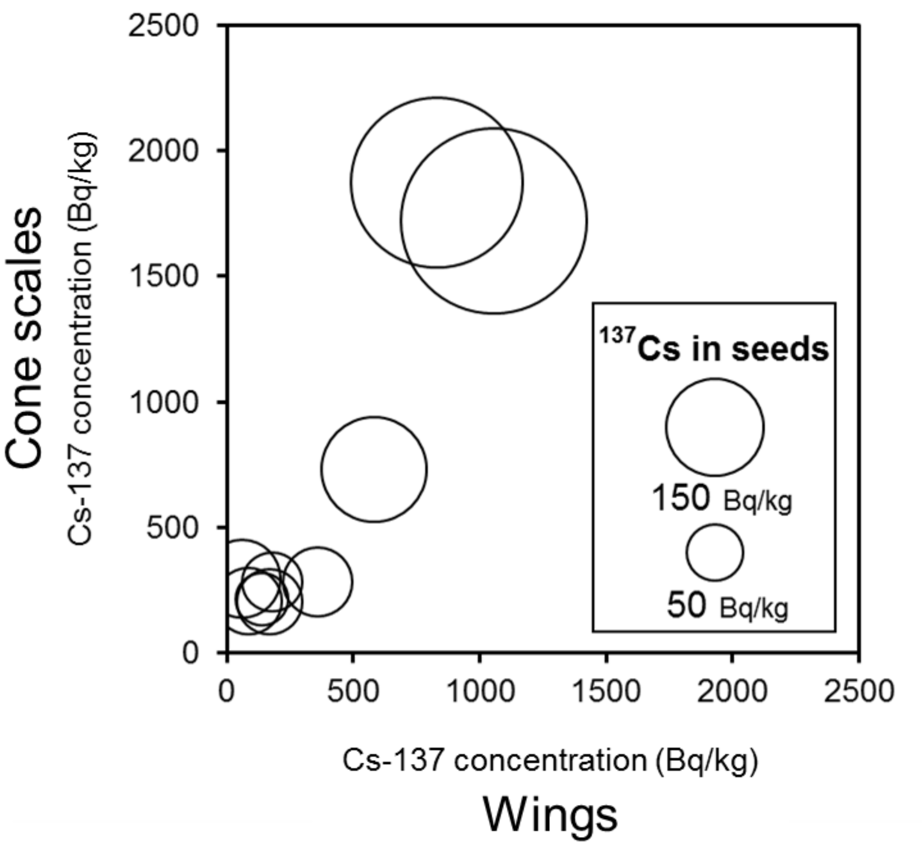


Figure 5-4. The relationships of ^{137}Cs concentrations in seeds, wings and cone scales of *A. firma*. Circle sizes show the ^{137}Cs concentrations in seeds.

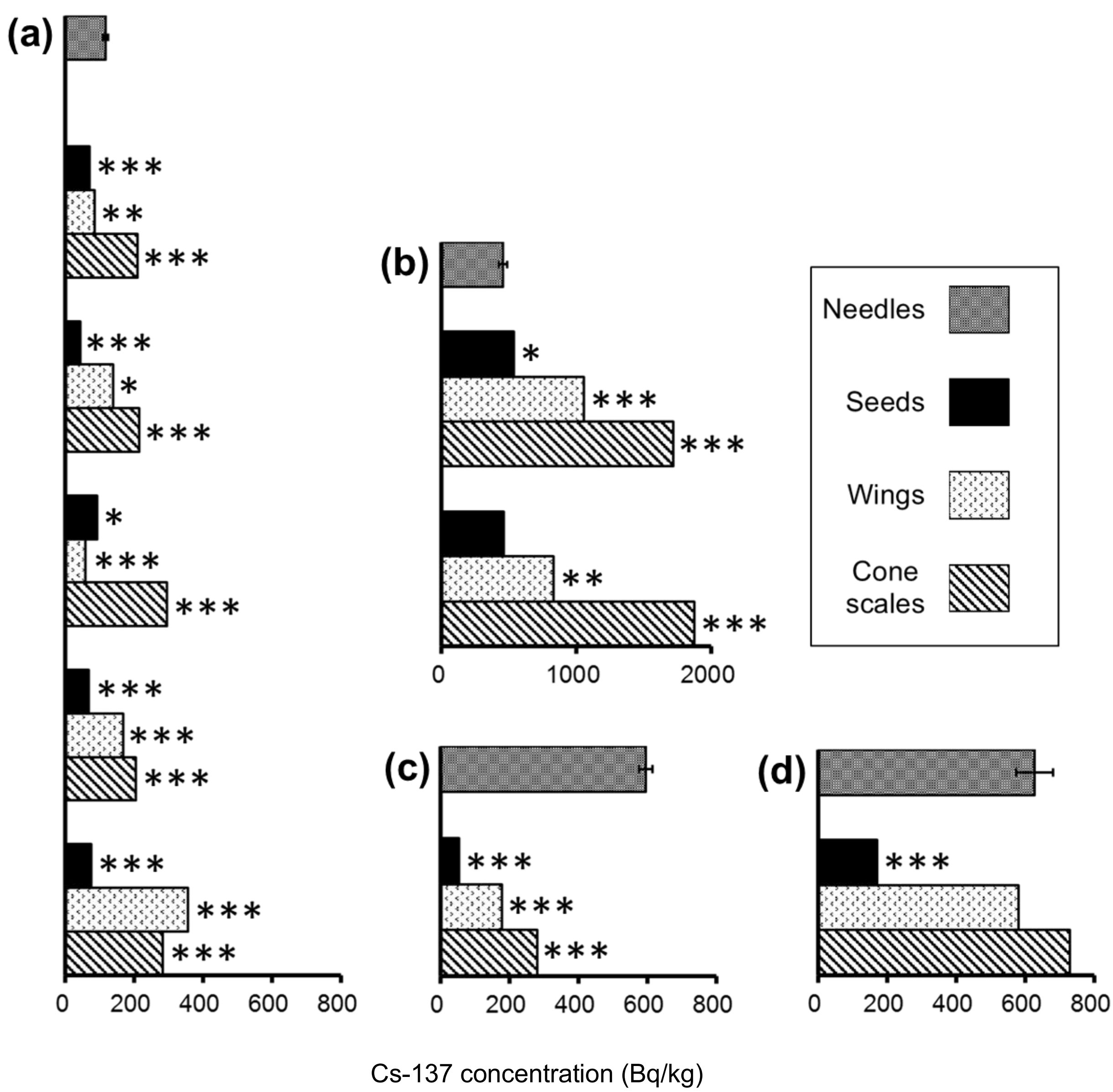


Figure 5-5. The ^{137}Cs concentration in needles, seeds, wings and cone scales of *A. firma*. The values of needles were the average values of 12 sample needles per tree. Asterisks show significant differences (***: $p < 0.001$, **: $p < 0.01$, *: $p < 0.05$) between needles and seeds or wings or cone scales based on the one sample t-test. (a) Tree No. 1 (5 cones), (b) Tree No.2 (2 cones), (c) Tree No.3 (1 cone), and (d) Tree No.4 (1 cone). Note that the scale of (b) is different from that of the other figures.

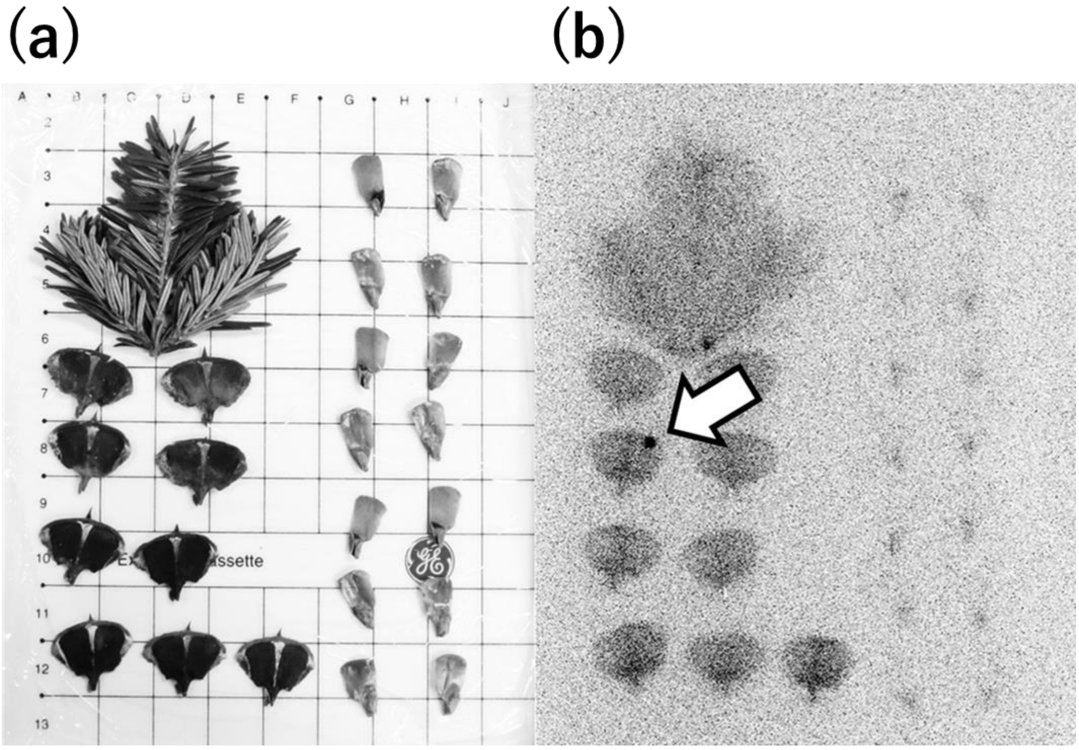


Figure 5-6. Radioactive distribution in shoots, winged seeds and cone scales. (a) Photograph of samples. (b) Radioactive distribution image obtained using imaging plates. Exposure time was 16 days. In the image, radioactivity increases from white to black. Radioactivity indicated by an arrow is probably caused by external contamination.

Table 5-1. Description of maternal trees (tree height, diameter at breast height: DBH), the number of cone samples and sampling height of needle samples.

Maternal trees			the number of sampled cones	sampling height of needles
Sample No.	Height (m)	DBH (cm)		
1	22.1	76.5	5	5
2	20.8	75.1	2	7
3	21.7	58.2	1	7.1
4	20.5	73.4	1	5