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Elevated radioactive contamination from the Fukushima nuclear power plant accident in aquatic biota from a river with a lake in its upper reaches

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1 **Elevated radioactive contamination from the Fukushima nuclear power plant accident in aquatic**
2 **biota from a river with a lake in its upper reaches**

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Abstract: Five years have passed since the Fukushima Daiichi Nuclear Power Plant (FDNPP) accident occurred. Forests, streams, and lakes remain radioactively contaminated, with slight sign of convergence. The radiocesium concentration of brown trout in Lake Chuzenji (160 km from the FDNPP) still exceeds the Japanese regulatory limit of 100 Bq/kg, likely due to elevated contamination in Lake Chuzenji. In this study, the concentration of ^{134}Cs and ^{137}Cs in algae, litter, sand substrate and aquatic insects in a river originating from Lake Chuzenji (Daiya site) and in a nearby river (Watarase site) from 2013 to 2015 were compared. At the Daiya site, ^{134}Cs and ^{137}Cs concentrations of algae and aquatic insects were high (e.g. ^{137}Cs in algae: 160 Bq/kg at the Watarase site and 320 Bq/kg at the Daiya site) and still increasing in some groups such as Perlodidae and Heptageniidae, though the average air dose rate (0.05 $\mu\text{Sv/h}$) was lower than that at the Watarase site (0.11 $\mu\text{Sv/h}$). We attributed this to high flow out of higher ^{134}Cs and ^{137}Cs concentration originating from Lake Chuzenji. Thus, lower reaches fed by contaminated headwaters will likely experience prolonged contamination.

26

Keywords: freshwater insects · lake and reservoir · litter · lower reaches · radioactive cesium concentration · sand · stream algae

29

30 **Introduction**

31

32 Streams that pass through radioactivity contaminated forests are important sources of drinking water
33 and components of urban environments. The first phase of investigation after the Fukushima Daiichi
34 Nuclear Power Plant (FDNPP) accident in 2011 revealed that large proportions of deposited ^{134}Cs and ^{137}Cs
35 were trapped in the forest canopy and litter layer on the forest floor (Hashimoto et al. 2012). ^{137}Cs is readily
36 adsorbed onto clay minerals and organic matter in soil (Kruyts and Delvaux 2002) and can be transported
37 by eroding soils and as dissolved organic matter via hydrologic channels into streams (Fukuyama et al.
38 2005; Kato et al. 2010; Wakiyama et al. 2010). Radioactive Cs in stream water exists in dissolved and
39 suspended forms (Ueda et al. 2013). Dissolved ^{134}Cs and ^{137}Cs in running water are relatively free from soil
40 adsorption and are readily taken up by microbes, algae and plants. This pathway of ^{134}Cs and ^{137}Cs
41 transport eventually leads to uptake by aquatic insects and freshwater fish at higher trophic levels.

42 The Chernobyl accident released more than five million terabecquerels of radionuclides, substantial
43 amounts of which spread to Finland, Sweden and Norway, 2000 km to the northwest. The broad effects of
44 the Chernobyl accident have been examined, and management measures have been designed for huge
45 geographic areas (Report of the Chernobyl Forum Expert Group, 2006; Yablokov et al. 2009). Changes in
46 the populations, biodiversity, mutation rate and fecundity of bird species after the Chernobyl accident have
47 also been studied (Møller and Mousseau 2011). In addition, Krivolutzkii & Pokarzhevskii (1992) described
48 the changes in animal populations in the soil after this accident. A few years after the accident, the densities

49 and biomasses of these populations were unrelated to the distance from the Chernobyl nuclear power
50 station. Jackson et al. (2005) also reported the abundances of terrestrial invertebrates in relation to
51 radioactivity levels, but failed to analyse the effects of radiation. Moreover, Møller & Mousseau (2009)
52 reported reductions in the abundances of insects in proportion to increases in radiation. Mietelski et al.
53 (2010) reported the accumulation of radionuclides in terrestrial invertebrates after the Chernobyl accident
54 and revealed that the contamination levels differed depending on the species.

55 Snowmelt water brings large deposits of ^{134}Cs and ^{137}Cs from dry riverbeds to streams, which alters the
56 levels of contamination of freshwater ecosystems. In the Dnieper River floodplain ecosystem, which drains
57 into the Black Sea, 85–97% of ^{137}Cs contamination was accumulated in aquatic plants, 1–8% in zoobenthos
58 and 1–8% in fish (Yablokov et al. 2009). Five to nine years after the accident, ^{134}Cs and ^{137}Cs
59 concentrations had fallen in lake water, but radioactivity had increased in sediments (Yablokov et al. 2009).
60 Furthermore, Galkovskaya & Molotkov (2001) described the lack of a relationship between planktonic
61 rotifer biodiversity in the Chernobyl region and radioactive contamination. Fish in lakes have also been
62 monitored for radioactivity and it was found that activity concentrations of ^{137}Cs in brown trout had
63 declined (Brittain et al. 1991; Kryshev et al. 1993; Kryshev 1995) but the level of contamination recently
64 has been constant (Brittain and Gjerset, 2010). ^{137}Cs contamination of water bodies was also shown to
65 have declined (Vakulovsky et al. 1994; Kryshev 1995). However, ^{137}Cs tends to be absorbed to the bottom
66 of the reservoir compared with ^{90}Sr (Vakulovsky et al. 1994) because ^{137}Cs is transported as colloids but
67 ^{90}Sr is present in the form of low molecular (Salbu et al. 1992). The broad effects of the Chernobyl accident

68 have been examined and management measures have been designed for large geographic areas. However,
69 the ecological consequences of radioactive contamination in aquatic ecosystems are poorly understood.

70 Fish in Japan were contaminated with radioactive substances after the Fukushima accident even where
71 dose rates in the atmosphere were relatively low (Wada et al. 2016). For example, in Lake Chuzenji (160
72 km from the FDNPP), ^{137}Cs concentrations in the stomach and muscle tissues of brown trout (*Salmo trutta*)
73 exceeded the Japanese regulatory limit of 100 Bq/kg in the winter of 2012 – 2013 (Yoshimura and Akama
74 2014; Yoshimura and Yokoduka 2014), and elevated ^{137}Cs concentrations persisted even in 2016 (Ministry
75 of Agriculture, Forestry and Fisheries (MAFF) 2015). Contamination of ^{137}Cs in muscle and egg tissue of
76 salmonid fish has also been investigated (Yamamoto et al. 2014 a, b). Rainbow trout (*Oncorhynchus*
77 *mykiss*) and kokanee (*Oncorhynchus nerka*) also inhabit Lake Chuzenji, but ^{137}Cs concentrations in the
78 muscle tissues of these species are nearly below the Japanese regulatory limit of 100 Bq/kg for radioactive
79 Cs (Yoshimura and Akama 2014; Yoshimura and Yokoduka 2014) and have been decreasing (MAFF 2015).

80 The Daiya River flows out of Lake Chuzenji and materials containing radioactive Cs are assumed to
81 flow downstream from the lake. To achieve a precise description of the mechanisms by which Cs deposited
82 in Lake Chuzenji is exported, we measured the concentrations of Cs in algae, litter, sand substrate and
83 aquatic insects inhabiting the Daiya River. Here, we report the results of investigations 45 months after the
84 FDNPP accident.

85

86 **Materials and methods**

87

88 **Study site**

89 The study was conducted in the Oku Nikko and Ashio regions, approximately 160 km southwest of the
90 FDNPP. According to an aircraft radioactivity survey (MEXT, 2013), the air dose rate in this region was
91 0.1–0.25 $\mu\text{Sv/h}$ and the deposition of $^{134+137}\text{Cs}$ ranged from 10,000 to 40,000 Bq/m^2 on May 31, 2012
92 (Table 1). The study catchment area is mostly forested and the dominant tree species are broadleaf and
93 deciduous. Other areas in the region are forested with Japanese cedar and cypress plantations used for
94 timber production. The field survey was conducted at one site along the Daiya River, part of the Kinu River
95 system in the Nikko region, and another site along the Watarase River in the Ashio region, which forms the
96 upper drainage component of the Tone River system, both in Honshu, Japan. The Daiya River originates
97 from Lake Chuzenji (Fig. 1).

98

99 **Sampling**

100 Air dose rates 1-m above ground were measured using a γ survey meter (NaI scintillation counter;
101 ALOKA TCS-172). Electrical conductivities (EC) of the streams were measured with a portable compact
102 twin conductivity meter (B-173; HORIBA); pH was measured with a portable compact twin pH meter
103 (B-212; HORIBA). Wetted river widths (RW) were measured with a measuring tape, and river velocities
104 were measured with a portable velocity meter (V-303, VC-301, KENEK). The data were sampled once in
105 each of February, July and November 2014 (the Watarase site) and once in each of July and November

106 2014 and February 2015 (the Daiya site).

107 Algae, litter and sand substrate were sampled from river riffles in February, July and October 2013 and
108 February, July and November 2014 at the Watarase site, and in July and November 2014 and February 2015
109 at the Daiya site. Periphytic algae attached to riverbed rocks were collected by brushing the substratum
110 with a toothbrush. These algae are primary producers at the base of river food webs. Prior to brushing, we
111 gently hand-rinsed the stone surface under running water to remove other organic matter from the epilithic
112 benthos. Litter comprising immersed leaves shed into the water by riparian trees was sampled after gentle
113 hand-rinsing. Litter leaves also form the base of river food webs. Sand substrate was sampled in each of the
114 riffles to a depth of 5-cm. When sand was not directly visible in the river substrate, stones were removed
115 and the sand underneath was sampled.

116 Aquatic insects from eight families (Heptageniidae, Perlidae, Perlodidae, Stenopsychidae,
117 Rhyacophilidae, Baetidae, Ephemerellidae and Tipulidae) were qualitatively sampled from riffles at the
118 Watarase site on six sampling days from February 2013 to November 2014 and at the Daiya site on six days
119 from July 2014 to February 2015. In addition to these eight families, aquatic insects from three families
120 (Ameletidae, Chloroperlidae and Ephemeridae) were qualitatively sampled at riffles. Six families
121 (Perlodidae, Ameletidae, Ephemerellidae, Tipulidae, Chloroperlidae and Ephemeridae) were qualitatively
122 sampled at pools at the Watarase site on February 12 of both 2013 and 2014. At each site, we placed a
123 D-frame net with a 1-mm mesh downstream from the sampling area on the substrate in water with a depth
124 of 20–25 cm. We then disturbed the substrate upstream of the net, allowing insects to drift into the D-frame

net. The sampled aquatic insects were identified to the family level in the field and then frozen.

Radiocesium analysis

Samples of algae were concentrated by evaporation and dried in an oven at 75°C. Samples of aquatic insects were freeze dried. Samples of litter and sand were dried in an oven at 75°C. After that, samples of sand were placed in a sieve (Iida, Japan, mesh size 2 mm) and sand that passed through the sieve was used. The respective samples were homogenised and packed into 100-ml polystyrene containers (U-8). We performed gamma-ray spectrometric measurements on each sample. The radioactive concentrations of ^{134}Cs (604 keV) and ^{137}Cs (662 keV) were measured with an HPGe coaxial detector system (GEM40P4-76, Seiko EG&G, Tokyo, Japan) at the Forestry and Forest Products Research Institute (FFPRI) for 3600 s or longer. Gamma-ray peaks of 604 and 662 keV were used to identify ^{134}Cs and ^{137}Cs , respectively. The measurement system was calibrated using a standard gamma-ray source (MX033U8PP, Japan Radioisotope Association, Tokyo, Japan), and a standard soil sample (IAEA-444) was used to check accuracy.

Homogenised samples of aquatic insects were also homogenised with Na_2SO_4 for radiocesium analysis. The processed aquatic insects were packed into 20-ml plastic tubes. We performed gamma-ray spectrometric measurements on each tube. Total concentrations of ^{134}Cs and ^{137}Cs were measured using a gamma particle counter with a NaI crystal detector (2480WIZARD², PerkinElmer, Downers Grove, IL, USA) at the FFPRI for 21,600 s. The measurement system was calibrated with a standard gamma-ray source (E-26-5/107F16-3, Canberra Industries, Oak Ridge, TN, USA), and a standard soil sample

(IAEA-444) was used to check accuracy. There was a strong correlation between the data obtained with the HPGe coaxial detector system and the data obtained with the NaI crystal detector. Therefore, the data obtained with the NaI crystal detector were used because they enabled us to count a smaller number of aquatic insects.

Because the sample size was small we conducted nonparametric tests for statistical analysis. The Kendall rank correlation test was used to clarify the relationship between the sampling day and ^{137}Cs concentration. Mann–Whitney *U*-tests were used to compare ^{137}Cs concentrations between riffles and pools and between the Daiya site and the Watarase site. To check the difference of decline between Daiya and Watarase, a test involving comparison of two regression slopes was conducted. Statistical analysis was performed using SYSTAT version 10 (SPSS Inc., 2000). Radiocesium concentrations that were below the detection level because of insufficient weight for radiocesium analysis were excluded from the statistical analysis.

Results

The radioactive dose rates in the air averaged 0.11 $\mu\text{Sv/h}$ (Watarase site) and 0.05 $\mu\text{Sv/h}$ (Daiya site). There were no differences in air temperature, water temperature, pH or dissolved oxygen between the Watarase site and the Daiya site. Conversely, EC, current velocity and dose rate did differ between these sites (Table 1).

The radioactive cesium concentrations of algae, litter and sand substrate in riffles varied in accordance with time since the accident at the Watarase site. The concentrations of ^{134}Cs and ^{137}Cs in algae and litter were not statistically correlated, but gradually decreased with time since the accident (Fig. 2a-d). The concentrations of ^{134}Cs and ^{137}Cs in sand substrate were significantly correlated and decreased with time since the accident (Fig. 2e,f). Because the half-life of ^{134}Cs is shorter than that of ^{137}Cs , the concentration of ^{134}Cs in some samples was below the detection limit. However, there was a close correlation between these radionuclides (algae: $\tau = 1.00$, $n = 5$, $P < 0.05$; litter: $\tau = 0.56$, $n = 12$, $P < 0.05$; sand substrate: $\tau = 0.82$, $n = 10$, $P < 0.001$; Kendall rank correlation test).

The concentrations of ^{134}Cs and ^{137}Cs in four families of aquatic insects in riffles tended to decrease with time since the accident, although a pattern was significant only in the Perlidae (Fig. 3a-h). The concentrations of ^{134}Cs and ^{137}Cs in two families of aquatic insects tended to increase with time since the accident (Fig. 3i-l). A close correlation was evident between these radionuclides (Heptageniidae: $\tau = 0.87$, $n = 6$, $P < 0.05$; Perlidae: $\tau = 0.91$, $n = 7$, $P < 0.005$; Perlodidae: $\tau = 1.0$, $n = 3$, ns; Stenopsychidae: $\tau = 0.80$, $n = 5$, $P < 0.05$; Ephemerellidae: $\tau = 1.00$, $n = 5$, $P < 0.05$; Tipulidae: $\tau = 0.4$, $n = 5$, ns; Kendall rank correlation test).

The radioactive ^{137}Cs levels in eight families of aquatic insects in riffles and pools at the Watarase site are indicated in Table 2. Radioactive ^{137}Cs levels differed greatly between insects sampled from riffles and those taken from pools. The radiocesium levels in four families (Perlodidae, Ephemerellidae, Tipulidae, Chloroperlidae) decreased between 2013 and 2014 but this change was significant only for pool samples

(riffles: $z = 0.58$, $n = 8$, ns; pools: $z = 2.02$, $n = 8$, $P < 0.05$; Mann-Whitney U-test). The radioactive ^{137}Cs levels of sand substrate in the riffles were 18 Bq/kg and those in the pools were 22 Bq/kg.

The radioactive cesium levels in algae, litter and sand substrate from the riffles at the Daiya site also varied in accordance with time since the accident. The concentrations of ^{134}Cs and ^{137}Cs in algae were not significantly correlated with time since the accident, but decreased overall (Fig. 4a,b). The concentrations of ^{134}Cs and ^{137}Cs in litter decreased significantly with time since the accident (Fig. 4c,d). The concentrations of ^{134}Cs and ^{137}Cs in sand substrate were not significantly correlated with time since the accident, but increased overall (Fig. 4e,f). A close correlation between these radionuclides was evident (algae: $\tau = 0.93$, $n = 9$, $P < 0.005$; litter: $\tau = 0.64$, $n = 14$, $P < 0.01$; sand substrate: $\tau = 0.64$, $n = 8$, $P < 0.05$; Kendall rank correlation test).

The concentrations of ^{134}Cs and ^{137}Cs in eight families of aquatic insects were not significantly correlated with time since the accident, but tended increase in seven families (Fig. 5a-n) while decreasing in one (Tipulidae: Fig. 5o,p). A close correlation between these radionuclides was evident (Heptageniidae: $\tau = 0.67$, $n = 4$, ns; Perlidae: $\tau = 1.00$, $n = 4$, $P < 0.05$; Perlodidae: $\tau = 1.0$, $n = 5$, $P < 0.05$; Stenopsychidae: $\tau = 1.00$, $n = 5$, $P < 0.05$; Rhyacophilidae: $\tau = 1.00$, $n = 5$, $P < 0.05$; Baetidae: $\tau = 1.00$, $n = 5$, $P < 0.05$; Ephemerellidae: $\tau = 0.80$, $n = 5$, $P < 0.05$; Tipulidae: $\tau = 0.33$, $n = 3$, ns; Kendall rank correlation test).

We calculated the degree of slope coefficient between time since the accident and the concentrations of ^{134}Cs and ^{137}Cs as a logarithmic function, and examined the difference in coefficient between the Watarase and Daiya sites by comparing the slopes of the resulting plots. These slopes differed significantly for

Perlidae taken from the Watarase and Daiya sites (^{134}Cs : $t = 3.45$, $n = 11$, $P < 0.02$; ^{137}Cs : $t = 4.28$, $n = 11$, $P < 0.005$, test of comparison of two regression slopes).

All of the value for radiocesium concentration on a dry weight basis at the Watarase and Daiya sites refer to a value for September 1, 2013 and were compared. There were no significant differences in ^{134}Cs and ^{137}Cs concentrations between the sites in algae and litter (Fig. 6). However, the concentrations of ^{134}Cs and ^{137}Cs in sand substrate were significantly higher at the Watarase site (Fig. 6).

The concentrations of ^{134}Cs and ^{137}Cs in eight families tended to be higher at the Daiya site than at the Watarase site, and they were significantly so for the Perlodidae and Heptageniidae (Fig. 7).

Discussion

The concentrations of ^{134}Cs and ^{137}Cs in algae and litter at the Daiya site are higher than those obtained from the upper reaches of Lake Chuzenji streams in 2012 (the six star symbols in Fig. 1, see Yoshimura and Akama 2014), although those of sand substrate are similar. The reason why the algae and litter are so contaminated at the Daiya site is expected to be associated with Lake Chuzenji. Three to four years had passed since the Fukushima accident, during which time flooding would have occurred on several occasions. Each flood would have resulted in the export of large amounts of radiocesium from the lake. When the flooding subsided, spot pools filled with radionuclides would remain and algae growing in these environments would be highly contaminated. Likewise, litter immersed in the pools would also be highly

contaminated. Thus, with no disturbances, radionuclide decay and the concentrations of ^{134}Cs and ^{137}Cs in algae and litter gradually decrease. By contrast, the sand substrate would have been contaminated with radiocesium when it was carried from Lake Chuzenji. Sand substrates are easily washed away even by small disturbances. So, little radiocesium contamination could accumulate at the Daiya site.

The concentrations of ^{134}Cs and ^{137}Cs in algae, litter and sand substrate at the Watarase site decreased in accordance with time since the accident. The same was true for aquatic insects at the Watarase site with the exception of a few groups. Thus, in general, radioactive Cs levels decreased in accordance with time since their deposition.

Although a decrease of radioactive concentrations in algae, litter and sand was observed at the Watarase site, the radioactive Cs level in sand substrate at the Daiya site increased gradually in accordance with time after deposition. The gradual increase in ^{134}Cs and ^{137}Cs concentrations in the sand substrate indicates that small amounts of sand with radioactive Cs have been carried downstream to the Daiya site from Lake Chuzenji. In this study, sand samples for measurement had been sieved (2mm), so they included all materials smaller than 2mm such as silt and clay. Radiocesium tends to be attached to the clay. They tend to be washed away by disturbance but not washed away under stable condition. The upper reaches of the Daiya site include Lake Chuzenji. Radioactive contamination has been observed in Lake Chuzenji fish (Yoshimura and Yokoduka 2014) and levels in excess of 100 Bq/kg are still found in brown trout inhabiting the deeper portions of the lake. Thus, we would infer that a large amount of radioactive cesium is deposited in the depths of Lake Chuzenji. In fact, ^{137}Cs level in a deep layer of Lake Chuzenji water (depth: 169 m)

and shallow layer (depth: 0.5 m) as well as Daiya River water are less than 1 Bq/L. However, ^{137}Cs in lake sand was found to be 450 Bq/kg (dry) in February 2013 (MOE 2013). In turn, the gradual increase of radioactive Cs at the sand substrate of the Daiya site suggests that radionuclides are slowly trickling down from the upper reaches, that is, Lake Chuzenji. The concentration of ^{137}Cs has been reported to be higher for clay than for silt (Bostick et al 2002, Tsukada et al 2008). Therefore, ^{137}Cs would trickle down to the lower reaches as accretion to clay. In Japan, there are many steep slopes, so a large amount of silt and clay with cesium would be brought to the lower reaches after heavy rainfall. Conversely, radioactive Cs levels in algae and litter at the Daiya site decreased in accordance with time since deposition.

The concentrations of ^{134}Cs and ^{137}Cs in aquatic insects at the Watarase site generally decreased with time since the accident, except for two groups: Ephemerellidae and Tipulidae. By contrast, radiocesium contamination increased in seven of eight aquatic insect groups at the Daiya site. The concentrations of ^{134}Cs and ^{137}Cs in the Perlidae, Perlodidae and Stenopsychidae at the Daiya site were higher than those in the upper reaches of Lake Chuzenji streams in 2012 (the six star symbols in Fig. 1, see Yoshimura and Akama 2014), although the concentrations in Stenopsychidae were similar. Primary consumers such as the Heptageniidae and Nemouridae eat algae and litter respectively (Merrit and Cummins 1996). Sand substrate and silt carried down from Lake Chuzenji would gradually accumulate in the river sometimes to the point of covering algae and litter. Primary consumers in the Daiya stream would eat silt covered algae and litter, and in turn be consumed by aquatic insects of the Perlidae and Perlodidae. Consequently, the concentrations of ^{134}Cs and ^{137}Cs in these families at the Daiya site would exceed those in the upper reaches.

By contrast, the Stenopsychidae construct filters between stones and eat materials that are trapped by these filter (Merrit and Cummins 1996). Thus, unlike the Perlidae and Perlodidae, the Stenopsychidae consume materials that flow down from the upper reaches, and their filtration technique would allow radionuclides associated with silt to be washed away by the current, because the mesh size of the net is large enough for silt and clay to pass through.

Because the air dose rate at the Watarase site exceeded that at the Daiya site, it is unsurprising that radiocesium levels were higher at the Watarase site. However the concentrations of ^{134}Cs and ^{137}Cs in aquatic insects were higher at the Daiya site, being several times higher than at the Watarase site in some families such as Perlodidae and Heptageniidae. This includes the effect of elevated contamination in Lake Chuzenji. There is a possibility that the difference of radioactive cesium concentration between Daiya and Watarase is depends on the natural level of potassium. However, the geological features of the surface soil are the same in Watarase, Daiya and the upper reaches of the lake and higher radioactive potassium concentration is related to higher radioactive cesium concentration, so this explanation is unlikely.

A pool is one of the easiest places for radionuclides to be deposited. In Norway, higher levels of radionuclides of several species were detected in pools and riffles due to fallout from Chernobyl although the examined species was different, and radioactive Cs concentrations of several species in pools and riffles decreased in a year (Solem and Gaare 1992). However, in our study of aquatic insects, the concentrations of ^{134}Cs and ^{137}Cs in pools decreased over a year. Radionuclide concentrations in riffles were lower than those in pools and no significant year-to-year differences in radionuclide levels were found in samples taken from

riffles. Because the ^{134}Cs and ^{137}Cs concentrations in aquatic insects in pools (lentic space) decreased, we might assume that a similar trend would occur for aquatic animals in the lake (lentic space). However, the radionuclide concentration in brown trout in Lake Chuzenji remained high. Pools generally experience limited but detectable water flow, which can contribute to the perpetual decrease of radioactive Cs in the pool and it would effect on the ^{134}Cs and ^{137}Cs concentrations of aquatic insects. Thus, we can infer that there is essentially no water movement in brown trout habitats in Lake Chuzenji. The difference in ^{137}Cs concentrations among species may arise from habitat differences even in a lake. Brown trout typically reside in the benthic substrate and middle strata of the lake interior, whereas rainbow trout reside in the middle to upper depths near the shoreline (Johnson 2016). Thus, the higher concentration of ^{137}Cs in brown trout indicates that radioactive substances tend to accumulate more in deeper areas where less water flow occurs. Most of the radionuclides in ponds are bound to the sediment and aquatic insects acquire them by ingesting the biotic components of the sediment (Voshell et al. 1985). Thus, the brown trout's preference for benthic habitat would lead to high levels of contamination. Ameletidae tend to move more easily between pools and riffles than the other sampled groups (Merritt and Cummins 1996), probably explaining the lack of a difference in Cs levels between the riffle and pool samples for this family.

The data presented in this manuscript are from approximately the first 3 years after contamination, a period during which activity concentrations were decreasing rather rapidly, although some ecosystem components higher up the food chain may still be showing increases. This is partly the reason for much of the variation in the data. Published data have shown variation, for example between individual fish or litter

samples (Yoshimura and Akama 2014; Yoshimura and Yokoduka 2014). Previous studies of Chernobyl fallout in aquatic systems have also shown that activity concentrations only began to stabilize after 10-15 years (Brittain and Gjerset, 2010). The variation in the data may also be partly due to seasonality. Higher water temperature brings higher contamination of ^{137}Cs in fish (Rowan and Rasmussen 1994). Litter is brought to the stream seasonally. Physiology of aquatic insects shows seasonality.

Brown trout in the Norwegian lake contaminated by radioactive fallout from the Chernobyl accident still showed substantial radioactive concentrations more than 20 years after the accident. Moreover, these levels were reported not to be declining any longer (Brittain and Gjerset 2010), probably because of continued input of radioactive substances from the catchment and the remobilization of lake sediment. ^{137}Cs activities in vegetation and water bodies in the United Kingdom have also been reported to have stabilized (Smith et al. 2000). The contamination of Lake Chuzenji seems to be prolonged. Rivers in contaminated areas with reservoirs in their upper reaches should be monitored to understand and help control the flow of radionuclides.

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Figure legends

Fig. 1. Study site of Lake Chuzenji and two rivers in the Oku Nikko and Ashio regions, Tochigi Prefecture, Japan. ●: sampling sites, different sampling site in Yoshimura & Akama (2014) ★: sampling sites in 2012-2013, Yoshimura & Akama (2014), ■: FDNPP, ▲: study area.

Fig. 2. Correlation between time since the accident and the radiocesium concentrations (^{134}Cs , ^{137}Cs) of algae, litter and sand substrate at Watarase site. Solid line: significant correlation, dotted line: tendency. a) ^{134}Cs of algae, $\tau = -0.56$, $n = 5$, ns; b) ^{137}Cs of algae, $\tau = -0.56$, $n = 5$, ns; c) ^{134}Cs of litter, $\tau = -0.43$, $n = 12$, ns; d) ^{137}Cs of litter, $\tau = -0.38$, $n = 12$, ns; e) ^{134}Cs of sand substrate, $\tau = -0.89$, $n = 10$, $P < 0.001$; f) ^{137}Cs of sand substrate, $\tau = -0.78$, $n = 10$, $P < 0.05$; Kendall rank correlation test.

Fig. 3. Correlation between time since the accident and the radiocesium concentrations (^{134}Cs , ^{137}Cs) of six families of aquatic insects at Watarase site. Solid line: significant correlation, dotted line: tendency. a) ^{134}Cs of Heptageniidae, $\tau = -0.47$, $n = 6$, ns; b) ^{137}Cs of Heptageniidae, $\tau = -0.33$, $n = 6$, ns; c) ^{134}Cs of Perlidae, $\tau = -0.90$, $n = 7$, $P < 0.005$; d) ^{137}Cs of Perlidae, $\tau = -0.93$, $n = 7$, $P < 0.01$; e) ^{134}Cs of Perlodidae, $\tau = -0.45$, $n = 3$, ns; f) ^{137}Cs of Perlodidae, $\tau = -0.45$, $n = 3$, ns; g) ^{134}Cs of Stenopsychidae, $\tau = -0.40$, $n = 5$, ns; h) ^{137}Cs of Stenopsychidae, $\tau = -0.20$, $n = 5$, ns; i) ^{134}Cs of Ephemerellidae, $\tau = 0.20$, $n = 5$, ns; j) ^{137}Cs of Ephemerellidae, $\tau = -0.20$, $n = 5$, ns; k) ^{134}Cs of Tipulidae, $\tau = 0.00$, $n = 5$, ns; l) ^{137}Cs of Tipulidae, $\tau = 0.60$, $n = 5$, ns; Kendall rank correlation test.

Fig. 4. Correlation between time since the accident and the radiocesium concentrations (^{134}Cs , ^{137}Cs) of algae, litter and sand substrate at Daiya site. Solid line: significant correlation, dotted line: tendency.

a) ^{134}Cs of algae, $\tau = -0.51$, $n = 8$, ns; b) ^{137}Cs of algae, $\tau = -0.42$, $n = 8$, ns; c) ^{134}Cs of litter, $\tau = -0.44$,
 $n = 14$, ns; d) ^{137}Cs of litter, $\tau = -0.54$, $n = 14$, $P < 0.01$; e) ^{134}Cs of sand substrate, $\tau = 0.51$, $n = 9$, ns;
 f) ^{137}Cs of sand substrate, $\tau = 0.42$, $n = 9$, ns; Kendall rank correlation test.

Fig. 5. Correlation between time since the accident and the radiocesium concentrations (^{134}Cs , ^{137}Cs) of

eight families of aquatic insects at Daiya site. Solid line: significant correlation, dotted line: tendency.

a) ^{134}Cs of Heptageniidae, $\tau = 0.55$, $n = 4$, ns; b) ^{137}Cs of Heptageniidae, $\tau = 0.91$, $n = 4$, ns; c) ^{134}Cs
 of Perlidae, $\tau = 0.18$, $n = 4$, ns; d) ^{137}Cs of Perlidae, $\tau = 0.18$, $n = 4$, ns; e) ^{134}Cs of Perlodidae, $\tau =$
 0.22 , $n = 5$, ns; f) ^{137}Cs of Perlodidae, $\tau = 0.22$, $n = 5$, ns; g) ^{134}Cs of Stenopsychidae, $\tau = 0.45$, $n = 5$,
 ns; h) ^{137}Cs of Stenopsychidae, $\tau = 0.45$, $n = 5$, ns; i) ^{134}Cs of Rhyacophilidae, $\tau = 0.55$, $n = 4$, ns; j)
 ^{137}Cs of Rhyacophilidae, $\tau = 0.55$, $n = 4$, ns; k) ^{134}Cs of Baetidae, $\tau = 0.59$, $n = 5$, ns; l) ^{137}Cs of
 Baetidae, $\tau = 0.59$, $n = 5$, ns; m) ^{134}Cs of Ephemerellidae, $\tau = 0.00$, $n = 5$, ns; n) ^{137}Cs of
 Ephemerellidae, $\tau = 0.22$, $n = 5$, ns; o) ^{134}Cs of Tipulidae, $\tau = -0.82$, $n = 3$, ns; p) ^{137}Cs of Tipulidae, τ
 $= 0.00$, $n = 3$, ns; Kendall rank correlation test.

Fig. 6. Comparison of the radiocesium concentrations (^{134}Cs , ^{137}Cs) of algae, litter and sand substrate

between the Watarase and Daiya sites. All radiocesium concentrations are compared to a reference

value from 1 September 2013. a) ^{134}Cs of algae, $z = 1.32$, $n = 13$, ns; b) ^{137}Cs of algae, $z = 1.32$, $n =$

13 , ns; c) ^{134}Cs of litter, $z = 0.14$, $n = 14$, ns; d) ^{137}Cs of litter, $z = 0.31$, $n = 26$, ns; e) ^{134}Cs of sand

substrate, $z = 2.09$, $n = 15$, $P < 0.05$; f) ^{137}Cs of sand substrate, $z = 2.87$, $n = 22$, $P < 0.005$;

Mann-Whitney U-test.

Fig. 7. Comparison of the radiocesium concentrations (^{134}Cs , ^{137}Cs) of eight families of aquatic insects between the Watarase and Daiya sites. All radiocesium concentrations are compared to a reference value from 1 September 2013. a) ^{134}Cs of Heptageniidae, $z = 2.56$, $n = 10$, $P < 0.05$; b) ^{137}Cs of Heptageniidae, $z = 2.56$, $n = 10$, $P < 0.05$; c) ^{134}Cs of Perlidae, $z = 0.00$, $n = 8$, ns; d) ^{137}Cs of Perlidae, $z = 0.00$, $n = 8$, ns; e) ^{134}Cs of Perlodidae, $z = 1.94$, $n = 8$, ns; f) ^{137}Cs of Perlodidae, $z = 2.23$, $n = 8$, $P < 0.05$; g) ^{134}Cs of Stenopsychidae, $z = 0.10$, $n = 10$, ns; h) ^{137}Cs of Stenopsychidae, $z = 10.10$, $n = 10$, ns; i) ^{134}Cs of Rhyacophilidae, $z = 0.78$, $n = 7$, ns; j) ^{137}Cs of Rhyacophilidae, $z = 0.78$, $n = 7$, ns; k) ^{134}Cs of Baetidae, $z = 1.17$, $n = 7$, ns; l) ^{137}Cs of Baetidae, $z = 1.17$, $n = 7$, ns; m) ^{134}Cs of Ephemerellidae, $z = 1.15$, $n = 10$, ns; n) ^{137}Cs of Ephemerellidae, $z = 1.15$, $n = 10$, ns; o) ^{134}Cs of Tipulidae, $z = 1.04$, $n = 8$, ns; p) ^{137}Cs of Tipulidae, $z = 1.04$, $n = 8$, ns; Mann-Whitney U-test.

Table 1. The amount of deposition on May 31, 2012, dose rate 1-m above the ground and the averaged value of dose rate 1-m above the ground and seven environmental factors on Daiya and Watarase sites.

	Watarase	Daiya	Mann-Whitney U-test (z)
Deposition ¹³⁷ Cs (KBq/m ²)	35	<10	-
Dose rate (μSv/h)	0.11	0.05	2.02*
Air temperature (°C)	14.9	14.2	0.22
Water temperature (°C)	10.1	10.8	0.22
pH	7.4	7.7	1.55
Electric Conductivity (μs/cm)	70	122	1.96*
Dissolved oxygen	9.8	10.4	1.09
Current velocity (m/s), riffle	0.53	0.46	2.32*
Altitude (m)	750	800	-

*: $P < 0.05$. The data in July 2014, November 2014, February 2015 were averaged at Daiya site, and the data in February 2014, July 2014, November 2014, were averaged at Watarase site.

Table 2. The concentration of ^{137}Cs in eight families of aquatic insects in riffle and pool at Watarase site on February 12 in 2013 and 2014. Dry weight of eight families for radio-caesium analysis and dominant species are also indicated.

		Radioactive ^{137}Cs (Bq/kg)		Dry weight for analysis (g)		Dominant species
		rifle	pool	rifle	pool	
Perlodidae	2013	89	200	0.43	0.16	<i>Sopkalia yamadae</i>
	2014	7	110	0.17	0.19	
Ephemerellidae	2013	14	180	1.37	0.23	<i>Drunella basalis</i> , <i>Cincticostella orientalis</i> , <i>Cincticostella elongatula</i>
	2014	33	49	0.61	0.34	
Tipulidae	2013	59	120	0.39	0.44	<i>Tipula</i> sp., <i>Hexatoma</i> sp.
	2014	110	16	0.22	0.13	
Ameletidae	2013	96	69	0.44	0.31	<i>Ameletus</i> sp.
	2014	24	63	0.54	0.20	
Heptageniidae	2013	63	-	0.77	-	<i>Epeorus latifolium</i>
	2014	41	-	0.45	-	
Perlidae	2013	51	-	0.47	-	<i>Acroneuria</i> sp., <i>Kamimuria</i> sp., <i>Calineuria</i> sp.
	2014	30	-	0.20	-	
Chloroperlidae	2013	-	290	-	0.14	<i>Sweltsa</i> sp.
	2014	-	190	-	0.11	
Ephemeridae	2013	-	46	-	0.25	<i>Ephemera japonica</i>
	2014	-	360	-	0.06	