Effective Microorganisms as a Potential Tool for the Remediation of ^{137}Cs-contaminated Soils

Aleksander N. Nikitin, Shuichi Okumoto, Galina Z. Gutzeva, Masaki Shintani, Galina A. Leferd, Ihar A. Cheshyk and Teruo Higa

***Abstract*— During the development of a method using effective microorganisms (EM) to reduce the soil-to-plant transfer of ^{137}Cs on land contaminated with radioactive Cs, an unexpected effect of EM on the reduction of the ^{137}Cs activity in soil samples was observed. Laboratory experiments were then conducted to evaluate the impact of EM and fermented organic fertilizer (EM Bokashi) on the ^{137}Cs activity in soil samples to investigate this observation. The experimental results indicate an increase in the ^{137}Cs decay rate of up to 4 times the theoretical decay rate corresponding to the half-life of ^{137}Cs, which is equal to 30.17 years. Our results suggest that EM accelerates the radioactive decay of ^{137}Cs in soil.**

***Index Terms*— cesium-137, radioactive isotopes, radioactive decay, effective microorganisms, soil**

I. Introduction

Nuclear accidents are highly undesirable but potential events in the process of operating nuclear power plants. Fukushima, Chernobyl, and other accidents have caused considerable areas to become contaminated with radioactive isotopes [1, 2]. One of the most critical and challenging tasks following such accidents is the decontamination of man-made radioisotopes in the soil. Current methods of decontamination are expensive and ineffective for large areas [3, 4]. Therefore, it is important to find innovative technologies to clean contaminated areas.

One option to economically use farmlands contaminated with ^{137}Cs is to substantially increase the application of potassium chemical fertilizers [5, 6]. However, this approach may lead to a decrease in the economic efficiency compared to non-contaminated areas as well as adverse ecological effects [7, 8]. Consequently, it is critical to develop new approaches to regulate the uptake of radioactive isotopes by plants.

Soil microorganisms can affect the bioavailability of man-made radioisotopes in the soil [9, 10]. Our research team (Institute of Radiobiology of the National Academy of Sciences of Belarus and EM Research Organization Inc., Japan) has investigated the possibility of using microbiological soil improver additives to decrease ^{90}Sr and ^{137}Cs transfer into crops [11-15]. Toward this goal, we tested mixtures of microorganisms known as effective microorganisms (EM). An EM is a people-friendly and environmentally safe microbial product that achieves synergistic effects by combining beneficial microorganisms that exist in nature, such as lactic acid bacteria, yeast, and phototrophic bacteria.

EM was initially developed in 1982 by Professor Teruo Higa to improve soils [16]. Following multiple studies, EM has now been in use for over 25 years in numerous fields, including sustainable agriculture, animal husbandry, and environmental conservation [17]. In addition, EM is distributed in more than 100 countries worldwide.

The results of our experiments confirm that EM and EM Bokashi reduce the transfer of ^{137}Cs and ^{90}Sr in corn, barley, lettuce, and vegetables [12, 13]. We suggested two mechanisms for this effect: a decrease in the bioavailable (soluble and exchange) forms of the radioisotopes in soil under the impact of EM and an increased migration of these radioisotopes downward in the soil profile. In our attempts to test these hypotheses, we observed an imbalance in the ^{137}Cs in the experimental system. After checking all the possible ways of losing radionuclides and all the sources of error, we suggest another hypothesis: the application of EM to soil increases the rate of ^{137}Cs radioactive decay. This study is devoted to confirming this hypothesis.

II. Materials and Methods

A. Soil samples

Sod-podzolic soil from the Chernobyl exclusion zone with a ^{137}Cs activity of approximately 10 KBq/kg was used in this experiment. The soil samples were dried and sieved through a 1-mm sieve. The entire volume of the soil was evenly mixed to

Manuscript received July 14, 2018. This work was supported in part by the EM Research Organization Inc. and in part through Belarussian State Program of Scientific Research "Nature Management and Ecology".

A. N. Nikitin is with the Institute of Radiobiology of the National Academy of Sciences of Belarus, Gomel 246007 Belarus (phone: 375-232-682035; fax: 375-232-683226; email: nikitinale@gmail.com).
S. Okumoto is with EM Research Organization Inc., Okinawa 901-2311 Japan (e-mail: sokumoto@emro.co.jp).
G. Z. Gutzeva is with the Institute of Radiobiology of the National Academy of Sciences of Belarus, Gomel 246007 Belarus (email: guzewa@mail.ru).
M. Shintani is with EM Research Organization Inc., Okinawa 901-2311 Japan (e-mail: m-shintani@emro.co.jp).
G. A. Leferd is with the Institute of Radiobiology of the National Academy of Sciences of Belarus, Gomel 246007 Belarus (email: leferd@mail.ru).
I. A. Cheshyk is with the Institute of Radiobiology of the National Academy of Sciences of Belarus, Gomel 246007 Belarus (email: cheshik@gsmu.by).
T. Higa is with International EM technology center, Meio University, Okinawa 905-8585 Japan.

provide a homogenous distribution of the ^{137}Cs activity and the physical and chemical properties of the soil. The soil was placed in a 100-ml container and mixed with different concentrations of EM or EM Bokashi.

B. Effective microorganisms (EM)

The EM (EM・1®) used in the experiments was supplied by EM Research Organization Inc., Japan. For application to soil, the EM was prepared in two forms: a liquid form (an EM solution) and a solid form (EM Bokashi). The EM solution was prepared by mixing EM with sugar cane molasses and water with a ratio of 1:1:20 (v/v). The mixed ingredients were transferred to a plastic container, which was tightly closed with a plastic lid and incubated for 20–25 days at 35±2°C to promote fermentation. The EM solution was considered ready for use when it produced a pleasant fermentation smell and the pH was below 3.5.

In Japanese, bokashi refers to fermented organic matter. EM Bokashi is an anaerobic fermentation product made from solid agricultural byproducts inoculated with EM. In EM Bokashi, organic matter serves as a growth medium for the microorganisms and provides a suitable microenvironment for EM in the soil. The EM Bokashi was prepared according to the method described in Ref. [16]. A mixture of 0.4 L of EM, 0.4 L of sugar cane molasses, and 4 L of chlorine-free tap water was added to 10 kg of wheat bran and mixed until homogeneous. The mixture was then placed in a plastic bag, which was hermetically sealed and kept under dark and warm conditions for 30 days. After the 30-day fermentation period, the EM Bokashi had a sweet-sour smell. The EM Bokashi was dried at room temperature prior to application.

C. Experimental layout

The different conditions of the experiment were as follows.

1. Absolute control (dry soil)
2. Positive control (wet soil)
3. Positive control 2 (wet soil + organic matter (OM))
4. EM-1 1% + molasses
5. EM-1 5% + molasses
6. EM-1 10% + molasses
7. EM Bokashi 1%
8. EM Bokashi 5%
9. EM Bokashi 10%
10. EM-1 10%

There were three types of control experiments: dry soil, wet soil with the addition of water, and wet soil with the addition of water, molasses, and wheat bran as organic matter (OM). The experimental treatments were 1%, 5%, and 10% EM with molasses and 1%, 5%, and 10% EM Bokashi. A treatment with EM but without molasses was also performed. Different exposure periods to the experimental treatments were set to 6 months, 12 months, and 18 months. Each treatment for each exposure period was repeated 15 times.

EM Bokashi was applied to the soil only at the beginning of the experiment. The EM, water, and molasses were added to the soil at the beginning of the experiment and every six weeks. Before adding the solutions, the containers were opened to allow for natural water evaporation. For the remainder of the time, the containers were sealed.

Soil samples were kept at room temperature (20–24°C) under natural light conditions.

D. Measurements

The ^{137}Cs activity was measured before and after each period of the experiment. The activity of the samples was determined in the containers where the samples were kept. The samples ware positioned precisely on the same axis as the detector to obtain a more stable result.

A GX 2018 gamma-spectrometry complex CANBERRA with a coaxial germanium detector with an extended energy range was used for the measurements. The γ-spectrometry measurement time was 600 s. The relative error in the measurements of the ^{137}Cs activity was less than 0.5%.

At the time of analysis, water was added to the samples to return the sample weight to its value at the time of the first measurement.

III. Results and Discussion

Table 1 shows the ^{137}Cs activity for each treatment at the beginning and end of the experiments during the 6-, 12-, and 18-month exposure periods. The ^{137}Cs activities at the beginning of the experiment were in the range of 692–929 Bq. Each sample in the experiment included precisely 100 g of evenly homogenized soil; however, we could not obtain a lower level of variability in these naturally contaminated samples.

After 6 months, the ^{137}Cs activity in the variants decreased to 686–924 Bq, and after 12 months, it decreased to 686–919 Bq.

TABLE I
THE ^{137}CS ACTIVITY IN THE SAMPLES AFTER DIFFERENT EXPOSURE PERIODS.

Treatments	6-month exposure		12-month exposure		18-month exposure	
	May 2016	November 2016	May 2016	May 2017	May 2016	November 2017
Absolute control (dry soil)	924 ± 30	924 ± 32	929 ± 57	919 ± 47	890 ± 24	883 ± 33
Positive control (wet soil)	692 ± 51	686 ± 51	694 ± 27	686 ± 23	743 ± 59	720 ± 63
Positive control 2 (wet soil + OM)	845 ± 31	824 ± 30	842 ± 18	811 ± 15	836 ± 25	812 ± 25
EM-1 1% + molasses	912 ± 20	893 ± 20	916 ± 22	861 ± 23	917 ± 25	873 ± 26
EM-1 5% + molasses	901 ± 15	879 ± 21	915 ± 25	886 ± 20	904 ± 23	880 ± 15
EM-1 10% + molasses	898 ± 40	860 ± 44	901 ± 48	869 ± 20	916 ± 32	874 ± 32
EM-bokashi 1%	929 ± 41	901 ± 39	913 ± 36	862 ± 29	908 ± 45	865 ± 47
EM-bokashi 5%	918 ± 29	877 ± 26	880 ± 34	829 ± 32	886 ± 25	845 ± 21
EM-bokashi 10%	854 ± 48	830 ± 36	901 ± 48	869 ± 37	852 ± 43	825 ± 35
EM-1 10%	904 ± 24	886 ± 23	912 ± 32	887 ± 37	917 ± 19	903 ± 22

This is equivalent to 0–4.4% and 1.0–5.9% decreases with respect to the initial activity, respectively.

In the 18-month experiment, the ^{137}Cs activity in the samples decreased to 720–903 Bq. Therefore, the measured ^{137}Cs activities in the samples decreased by 0.81–4.75% after 18 months.

The decrease in the ^{137}Cs activity after 6 months due to radioactive decay is 1.1% because the half-life of the ^{137}Cs radioisotope is 30.17 years. The reduction rates in the dry soil and wet soil control groups were nearly the same as the theoretical decay rate (Fig. 1). In all the other treatment groups, the rates were higher than the theoretical decay rate. In particular, a significant difference was detected in the wet soil with OM, 5% and 10% EM, and 1%, 5%, and 10% EM Bokashi treatments. The difference between these treatments and the theoretical decay rate was 1.3–3.3% (Fig. 1).

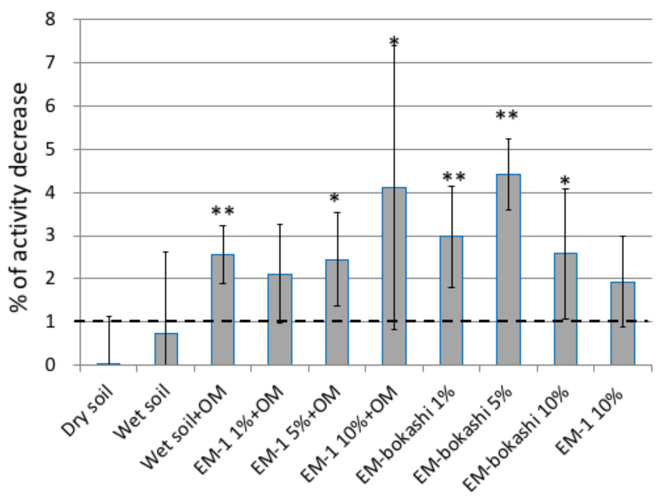

Fig. 1. The reduction rate of the ^{137}Cs activity in soil samples after the 6-month exposure treatments (mean ± confidence interval, p = 0.05). The dashed line indicates the calculated decrease in the activity due to radioactive decay with a half-life, $T_{1/2}$, of 30.17 years. * and ** indicate significant differences with respect to the calculated value at $p < 0.05$ and $p < 0.01$, respectively.

The theoretical decrease in the ^{137}Cs activity for 12 months is 2.3%. The actual reduction rates were higher than the theoretical decay rate for all treatments except the dry soil and wet soil treatments (Fig. 2). In addition, there were significant differences in the reduction rates of the wet soil with organic matter, 1% EM, and 1% and 5% EM Bokashi treatments compared to the theoretical decay rate. The difference between these treatments and theoretical decay rate was 1.3–3.7%.

The theoretical decrease in the ^{137}Cs activity for 18 months is 3.4%, which is shown by the dashed line in Fig. 3. The 1% EM and 1% and 5% EM Bokashi treatments had a significantly higher reduction rate than the theoretical decay rate (Fig. 3). The difference between these treatments and the theoretical decay rate was 1.2–1.4%.

Therefore, in the 6-, 12-, and 18-month exposure experiments, the 1% and 5% EM Bokashi treatments consistently had significantly higher reduction rates than the theoretical decay rate. While not consistent, the 1% EM treatment had a significantly higher reduction rate for the 12- and 18-month exposure experiments. Therefore, the application of EM and EM Bokashi to soil appears to have an impact on the radioactive decay rate.

The physical half-life of radionuclides is known to be very stable. Therefore, this observation is counter to the current understanding of radionuclides. However, similar results have been confirmed in another experiment and in field observations.

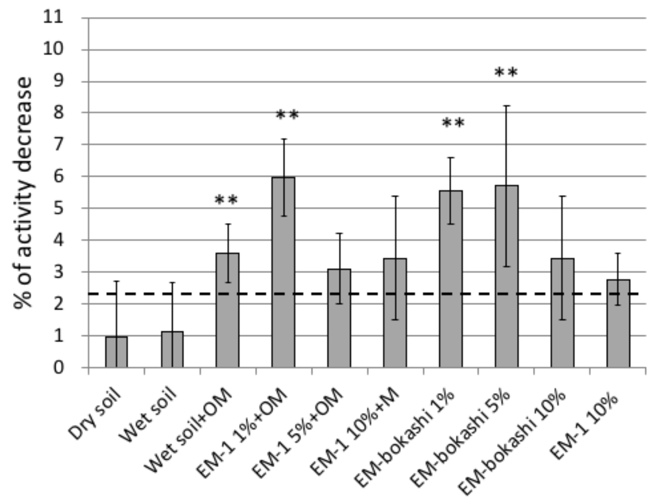

Fig. 2. The reduction rate of the ^{137}Cs activity in soil samples after the 12-month exposure treatments (mean ± confidence interval, p = 0.05). The notation is the same as in Fig. 1.

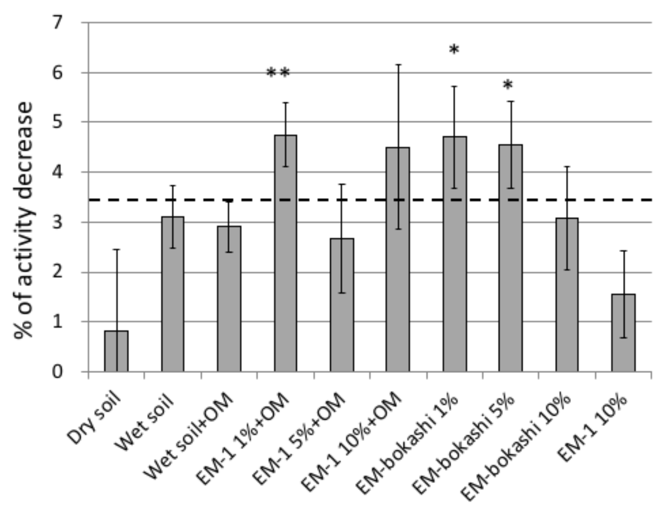

Fig. 3. The reduction rate of the ^{137}Cs activity in soil samples after the 18-month exposure treatments (mean ± confidence interval, p = 0.05). The notation is the same as in Fig. 1.

A similar laboratory experiment was conducted in Fukushima [18]. In the 690 days of the experiment, the reduction in the ^{137}Cs activity in soil treated with EM with molasses was confirmed to be greater than the theoretical decay value.

Monitoring of the $^{134+137}$Cs contamination of soils in paddy fields with long-term (more than 20 years) and short-term applications of EM in the Fukushima area shows significantly more rapidly decreasing radioisotope activities compared to the physical decay value [19]. Furthermore, the rate of decreasing radioactive cesium concentration in the soil on another farm was higher in a field treated with EM-fermented cow manure compost than in an adjacent area using a chemical fertilizer [20].

According to the conventional scientific paradigm, the rate of radioactive decay is not affected by operations such as heating, the addition of water, or the addition of organic matter. It should only decrease exponentially according to the law of radioactive decay. However, according to the hypothesis of bio-transmutation [21-23], some microorganisms may alter the rate of radioactive decay.

The obtained results suggest that EM accelerates radioactive cesium decay in soil. In view of the magnitude of these findings for mitigating radiation disasters, we need to obtain additional confirmations of this phenomenon under different conditions. If confirmed, it could be used for the remediation of areas contaminated with radioactive isotopes of cesium.

IV. Conclusions

The results of the experiments suggest that effective microorganisms (EM) accelerate the radioactive decay of ^{137}Cs in soil. Further studies are needed to understand the impact of EM on the ^{137}Cs activity.

References

[1] Y. A. Izrael, "Chernobyl radionuclide distribution and migration," *Health Phys.*, vol. 93, no. 5, pp. 410–417, Nov. 2007.

[2] T. J. Yasunari, A. Stohl, R. S. Hayano, J. F. Burkhart, S. Eckhardt, and T. Yasunari, "Cesium-137 deposition and contamination of Japanese soils due to the Fukushima nuclear accident.," *Proc. Natl. Acad. Sci. U. S. A.*, vol. 108, no. 49, pp. 19530–4, Dec. 2011.

[3] K. Wada, T. Yoshikawa, and M. Murata, "Decontamination Work in the Area Surrounding Fukushima Dai-ichi Nuclear Power Plant: Another Occupational Health Challenge of the Nuclear Disaster," *Arch. Environ. Occup. Health*, vol. 67, no. 3, pp. 128–132, Jul. 2012.

[4] S. Fesenko, P. Jacob, A. Ulanovsky, A. Chupov, I. Bogdevich, N. Sanzharova, V. Kashparov, A. Panov, and Y. Zhuchenka, "Justification of remediation strategies in the long term after the Chernobyl accident," *J. Environ. Radioact.*, vol. 119, pp. 39–47, May 2013.

[5] N. Yamaguchi, I. Taniyama, T. Kimura, K. Yoshioka, and M. Saito, "Contamination of agricultural products and soils with radiocesium derived from the accident at TEPCO Fukushima Daiichi Nuclear Power Station: monitoring, case studies and countermeasures," *Soil Sci. Plant Nutr.*, vol. 62, no. 3, pp. 303–314, 2016.

[6] N. Kato, N. Kihou, S. Fujimura, M. Ikeba, N. Miyazaki, Y. Saito, T. Eguchi, and S. Itoh, "Potassium fertilizer and other materials as countermeasures to reduce radiocesium levels in rice: Results of urgent experiments in 2011 responding to the Fukushima Daiichi Nuclear Power Plant accident," *Soil Sci. Plant Nutr.*, vol. 61, no. 2, pp. 179–190, 2015.

[7] S. Savci, "Investigation of Effect of Chemical Fertilizers on Environment," *APCBEE Procedia*, vol. 1, pp. 287–292, Jan. 2012.

[8] S. A. Khan, R. L. Mulvaney, and T. R. Ellsworth, "The potassium paradox: Implications for soil fertility, crop production and human health," *Renew. Agric. Food Syst.*, vol. 29, no. 01, pp. 3–27, Mar. 2014.

[9] S. Ehlken and G. Kirchner, "Environmental processes affecting plant root uptake of radioactive trace elements and variability of transfer factor data: a review," *J. Environ. Radioact.*, vol. 58, no. 2–3, pp. 97–112, Jan. 2002.

[10] J. R. Lloyd and J. C. Renshaw, "Microbial transformations of radionuclides: fundamental mechanisms and biogeochemical implications.," *Met. Ions Biol. Syst.*, vol. 44, pp. 205–40, 2005.

[11] G. Z. Gutzeva, G. A. Leferd, and S. O. Gaponenko, "Impact of mineral and bacterial preparations on lettuce productivity and accumulation of ^{137}Cs in plants," in *Proc. 5th Congr. of Belarussian Society of Soil and Agrochemistry Scientists*, Minsk, 2015, pp. 302—305.

[12] G. Z. Gutzeva, A. N. Nikitin, and N. V. Telitsina, "Accumulation of ^{137}Cs by biomass of barley in dependence from conditions of bacterization and mineral nutrition," in *Proc. Int. Sci. Conf. Radiobiology: «Mayak», Chernobyl, Fukushima*, Gomel, 2015, pp. 72—75.

[13] N. V. Shamal, E. A. Klementieva, R. A. Korol, R. K. Spirov, S. O. Gaponenko, A. N. Nikitin et al., "Application of EM-technologies for growing lettuce on the contaminated soils," in *Proc. Int. Conf. Current Status and Perspectives of Innovation Development of Vegetable Growing*, Samochvalovichi, 2015, pp. 45—47.

[14] A. N. Nikitin "Prospects for the use of microbiological preparations for regulating the accumulation of man-made radionuclides by plants," in *Proc. Int. Sci. Conf. Radiobiology: «Mayak», Chernobyl, Fukushima*, Gomel, 2015, pp. 157—161.

[15] N. Shamal, E. Klementjeva, R. Korol, S. Gaponenko, R. Spirov, A. Nikitin et al., "Application microbiological preparation EM-1 and mineral sorbent for growing lattice on the soils contaminated by radionuclides," in *Proc. 3d Int. Conf on Radiation and Applications in Various Fields of Research*, 2015, p. 574.

[16] T. Higa, "Effective Microorganisms: A Biotechnology for Mankind," in *Proc.* 1st *Int. Conf. on Kyusei Nature Farming*, USDA, Washington, DC, 1991, pp. 8-14.

[17] M. Olle, I. H. Williams, "Effective microorganisms and their influence on vegetable production – a review," *Journal of Horticultural Science & Biotechnology*, vol. 88, no. 4, pp. 380—386, 2013.

[18] S. Okumoto, M. Shintani, and T. Higa, "Possibilities of effective microorganisms (EM) technology for reducing radioactive cesium contamination in soil," in *Proc. Int. Sci. Conf. Chernobyl: 30 years later*, Gomel, 2016, pp. 157—159.

[19] S. Okumoto, M. Shintani, and T. Higa, "Analysis of radioactive cesium in paddy fields applied with effective microorganisms (EM 1®) in Fukushima," in *Proc. Int. Sci. Conf. Radiobiology: Challenges of the XXI Century*, Gomel, 2017, pp. 20—23.

[20] S. Okumoto, M. Shintani, and T. Higa, "Influence on the suppression of transfer of radioactive cesium from soil to grass using cow manure compost ant its effluent fermented by effective microorganisms™," in *Proc. Int. Sci. Conf. Radiobiology: «Mayak», Chernobyl, Fukushima*, Gomel, 2015, pp. 20—23.

[21] V. I. Vysotskii, and A. A. Kornilova, "Nuclear Fusion and Transmutation of Isotopes in Biological Systems," Mir, Moscow, 2003.

[22] J.-P. Biberian, "Biological Transmutations: Historical Perspective," *J. Condensed Matter Nucl. Sci.*, vol. 7, pp. 11-–25, 2012.

[23] H. Kozima, "Biotransmutation as a Cold Fusion Phenomenon," in *Proc. JCF16*, 2016, pp. 216–239.