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日本原子力研究所
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A Microbiological Study on Irradiated Sludge Composting

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Effect of fermentation temperature on microorganisms in sewage sludge compost and suppressive effect of the compost on Fusarium oxysporum were investigated. Dehydrated sewage sludge was irradiated at 10 kGy by cobalt 60 gamma ray source and fermented at various temperatures with six different seed-composts. It was found that microorganisms showed higher growth in irradiated sludge at the temperature around 30 to 40 °C. One of the seed-composts and compost produced from the seed-compost showed the remarkable effects of suppression on F. oxysporum. It can be also observed that the composts produced by lower temperature fermentation showed higher suppression.

Keywords : Radiation, Sewage Sludge, Microorganisms, Composting, Fusarium oxysporum, Suppressive Effect

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放射線照射汚泥コンポスト化の微生物学的研究

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(1993年2月3日受理)

下水汚泥コンポスト中の微生物に対する発酵温度の影響，並びに植物病原菌Fusarium oxysporumに対する抑制効果について検討した。脱水汚泥の照射は，コバルト60ガンマ線源により行い，6種の種コンポストを用いて発酵を行った。照射汚泥に植え付けた微生物は，30から40℃付近の発酵温度において高速で増殖した。用いた種菌の一つとその種菌を用いて得られたコンポストはF. oxysporumに対して顕著な抑制効果を示した。また，低温発酵により得られたコンポストの方が，より高い抑制効果を示した。

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1. Introduction

Management of sewage sludge is considered as one of the most serious problems in large cities. Land disposal of huge amounts of sludge causes environmental problems such as bad odor, generation of harmful insects, pathogenic contamination etc.

On the other hand, continuous extensive agricultural practice that depends heavily on application of chemical fertilizer has resulted in loss of organic matter, an increase in acidity, and accumulation of toxic elements in fertile soil, creating an environment favorable for development of certain soil-bone diseases. Generally, an application of composts to agricultural land is very useful for improvement of the quality of soils because the composts contain high level of organic matter and various kind of microorganisms helpful to support the plant growth.

There are various ways of sewage sludge management. Recently, composting has become widespread as a treatment process of sewage sludge from a viewpoint of recycling of organic materials and other trace elements useful for plant growth. The composting is an aerobic fermentation process of organic solid. The purpose of composting of sewage sludge is stabilization of easily decomposable substances in sludge which may cause bad odor by anaerobic fermentation in soil after application. Another purpose is sanitation by heat which is generated during fermentation process. The compost product, therefore, can be applied to land without adversely affecting the environment. The compost product is mainly used as a fertilizer or a soil conditioner.

Control of sludge composting process involves several factors, such as ventilation, temperature, moisture content and etc. Temperature, however, seems to be a critical element of control. Nakasaki [1] stated that a high temperature during composting of various materials is effective for the pasteurization of pathogenic microorganisms in the materials, for the promotion of water evaporation from the composting solid materials and for the acceleration of the rate of degradation of organic matter in the composting materials. Golueke [2] showed that the range of optimum temperature for the composting process as a whole is broad from 35 to 55 ˚C, because various microorganisms are involved in the decomposition of organic matter. Kawakami [3] reported that the optimum temperature for irradiated sewage sludge composting is around 50 ˚C.

Many microbiological study on composting also have been conducted. The microbial population in agricultural waste composting was reported by Fergus [4] and in municipal waste composting by Kain and Mullins [5] and Finstein [6]. Nakasaki et al. [1] has studied the change in microbial population during composting process of sewage sludge. Some composts are known to have sup-

pressive effect on occurrence of plant diseases as shown by Phae et al. [7].

In the process of conventional composting method of sewage sludge, high temperature is used to pasteurize pathogenic microorganisms in composting materials. According to a recommendation of the Association for Utilization of Sewage Sludge in Japan, it is necessary to keep composting temperature higher than 65 °C for more than 2 days during composting [8]. Such temperature is well above the thermal death points of most pathogens. In such case, even useful microorganisms are also eliminated.

It is possible to separate disinfection from composting by ionizing radiation because irradiation is an effective means of reducing the pathogens. Effect of radiation on disinfection of sewage sludge is reported by Ahlstrom [9]. Low temperature composting is possible in this case and the products may contain various kinds of microorganisms useful for plant growth. However, there has been no information on microbiological research on low temperature composting.

In this paper, the microbiological study on irradiated sewage sludge composting was investigated. The purposes of the study are to investigate the effect of temperature on microorganisms in composts produced from irradiated sewage sludge and also to evaluate the quality of the compost products by testing their suppressive effect on a plant pathogenic fungi, F. oxysporum.

2. Materials and methods

2.1 Apparatus

The fermentor used is a cylindrical glass container (70 mm in diameter, 55 mm in depth) with a perforated plate at the bottom to distribute the air from a compressor. Detail of the fermentor is shown in Fig. 1. It was covered with silicone cap directly connected with an exhaust gas carrying unit and a water trap unit. Three fermentors were contained in one set and each fermentor was separately removable. Those fermentors were embedded in a water bath to control the temperature during operation. Air from the compressor was split into five streams. The first was supplied for measuring carbon dioxide contained in air with a flow rate of 100 ml/min. The second was for dilution of the exhaust gas from the fermentors. The flow rate of dilution gas was set to be 300 ml/min. The third to fifth were supplied through each fermentor with 100 ml/min. The schematic diagram of the apparatus is shown in Fig. 2.

2.2 Disinfection of sludge

The sludge was disinfected by gamma-ray irradiation of 10 kGy. The dose rate was 5 kGy/h.

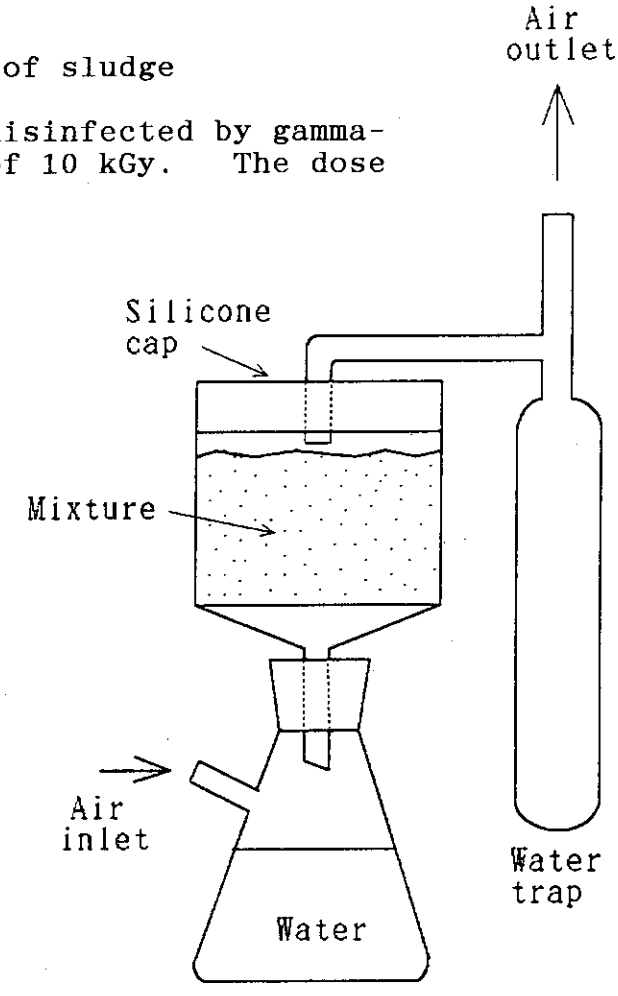

Fig. 1 Details of fermentor

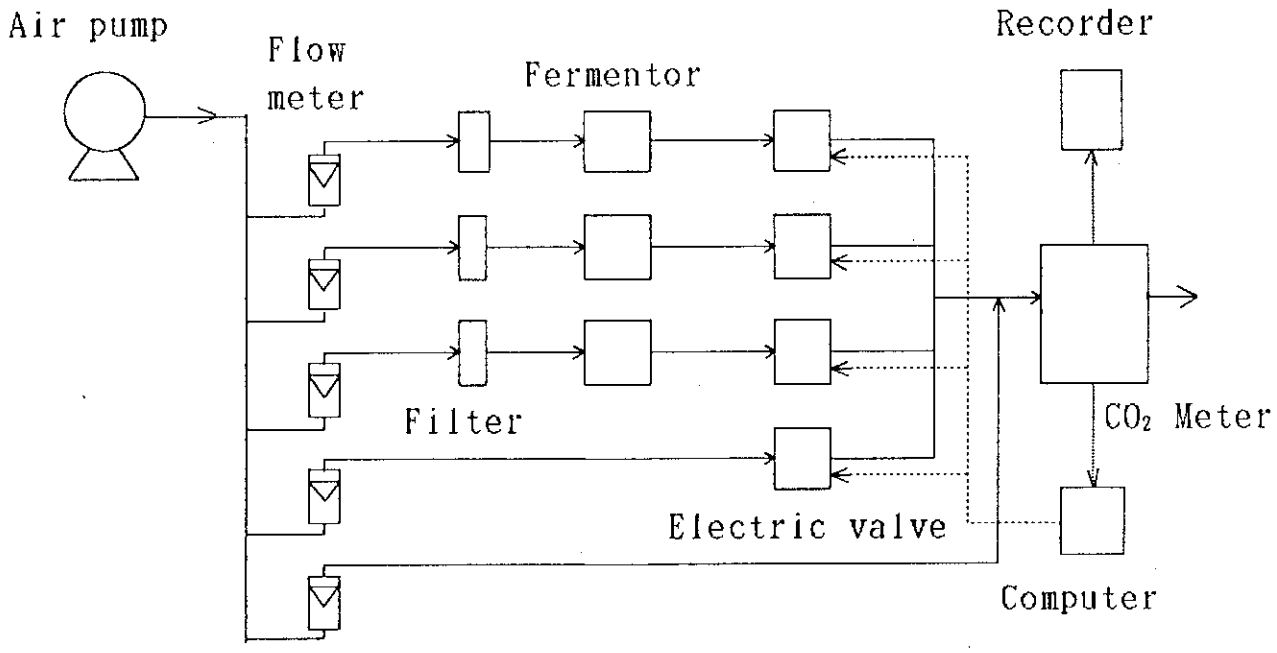

Fig.2 Schematic diagram of the experimental apparatus

2.3 Composting

Dehydrated sewage sludge from Akutsu Sewage Treatment Facility in Takasaki-city was used for the experiment. Moisture contents were ranged from 70.9 to 72.1 % and volatile solid contents were ranged from 56.7 to 57.4 %. The composting was performed at 30, 40, 50 and 65 °C. Six seed-composts were used and their properties were shown in Table 1. The composting materials for each run consisted of 30 g of irradiated sludge, 3 g of seed-compost, 15 g of sterilized perlite as a bulking agent, and 240 mg of sodium carbonate for adjusting pH to be around 7. After mixing, the composting materials were transferred into a sterilized fermentor, then covered with sterilized cap. After embedding the fermentor in the water bath, the operation was started by means of a micro-computer controlling. The operation was stopped when CO_2 evolution decreased and became constant.

Table 1. Name and properties of seed-composts

property	Seed-compost					
	A	B	T-A	O-1	O-2	R
pH	8.2	6.1	7.6	6.3	6.8	6.4
MC	19.1	62.5	60.5	13.5	15.0	43.8
VS	61.0	77.4	76.5	79.4	50.3	30.3

MC: Moisture content(%), VS: Volatile solid content(%)

CO_2 concentration from exhaust gas of each fermentor was continuously measured every 1 hour after it was diluted with 300 ml/min flow rate of air stream. The moisture contents of compost product and seed-compost were determined from the loss of the weight after drying at 105 °C until the weight becoming constant. The dried solids were heated at 600 °C for 4 h. The loss of weight was calculated as volatile solid content. The pH of compost product and seed-compost were measured by using Horiba compact pH meter C-1.

2.4 Isolation and enumeration of microorganisms

Numbers of total bacteria, actinomycetes, molds and yeasts in seed-composts and compost products were enumerated. The isolating media are shown in Table 2.

Three grams of sample was placed into 18 ml of sterilized water containing 0.01% Tween 20 in a sterilized polyethylene bag. The mixture was homogenized with a stomacher Lab-blender 80 for 2 min. The slurry was filtrated through sterilized four-layer gauze. After serial dilution of filtrate in sterilized water containing 0.01 % Tween 20, 0.2 ml of each appropriately diluted solution was plated in triplicate onto the three kinds of media as mentioned above. These plates had been heated overnight at 40 °C before use to minimized spreading of colonies because of excess water. The incubation condition are also shown in Table 2. The average number of microorganisms from three plates was calculated.

2.5 Suppressive effect test

1) Agar assays for antagonists

Two approaches were used. The first, 0.1 ml of spore suspension of F. oxysporum was spread onto Difco-Potato Dextrose Agar (PDA: potatoes, infusion form, 200 g; Bacto-dextrose, 20 g; Bacto agar, 15 g; distilled water, 1 liter [pH 5.5]) plate. The spore suspension was adjusted to be 10^6/ml in distilled water containing 0.01 % Tween 20 by counting chamber method. 0.5 g of sample was placed at the center of plate. The plate was kept

at 30 °C for 20 - 30 days. Inhibition zones around the sample were recorded. The second, the inoculated PDA was prepared as mentioned in the first approach. Two grams of a sample were suspended in sterilized 18 ml of distilled water containing 0.01 % Tween 20. The mixture was homogenized with a stomacher Lab-blender for 2 min. The slurry was filtrated through sterilized four-layer gauze. 0.2 ml of filtrate was dropped onto PDA plate by using drop plate method. The plate was kept at 30 °C for 7 - 15 days. Inhibition zones around the drop of filtrate were recorded. Two plates were prepared for each sample.

Table 2 Types of media and incubation condition for isolation and enumeration of microorganisms

Microorganisms	Isolating media	Formula & ingredients per liter		Incubation temp. and time
Bacteria	Nutrient agar	Beef extract	3 g	30 °C, 48 hr
		Peptone	5 g	
		Agar	15 g	
		Distilled water	1 l	
		Final pH	6.8	
Actinomycetes	Soluble starch agar	Soluble starch	10 g	30 °C, 4-5 days
		Ammonium sulfate	0.5 g	
		Di-Potassium hydrogen phosphate	0.5 g	
		Agar	20 g	
		Distilled water	1 l	
		Final pH	7.0	
Molds and Yeasts	Malt yeast extract agar	Malt extract	10 g	30 °C, 3 days
		Yeast extract	4 g	
		Glucose	4 g	
		Chloramphenicol	20 mg	
		Agar	20 g	
		Distilled water	1 l	
		Final pH	5.5	

2) Compost inoculation assays

Three grams of a sample was placed into a sterilized vial tube (30 ml) and inoculated with 0.3 ml of spore suspension of F. oxysporum. The spore suspension was adjusted to be 10^7/ml. The vial tube was stopped with a sterilized porous silicone cap to allow gas exchange, then kept at 30 °C. The F. oxysporum was enumerated by colony counts on malt-yeast extract agar after 7 days incubation.

To enumerate the F. oxysporum in the inoculated sample from a vial tube, three grams of sample was suspended into 27 ml of sterilized water containing 0.01% Tween 20 in sterilized polyethylene bag. The mixture was homogenized with a stomacher Lab-blender 80 for 2 min. After filtration through sterilized four-layer gauze, 0.2 ml of appropriate dilution was plated in duplicate onto the malt-yeast extract agar. All plates were incubated at 30 °C for 3 days. The number of F. oxysporum was obtained as the average of two samples in each vial tubes.

3. Results and discussion

3.1 Effect of temperature on microorganisms in composts

3.1.1 Microbial number in seed-composts

Number of microorganisms isolated from six different seed-composts are shown in Table 3. Seed A is the compost produced by the pilot plant in Akutsu Sewage Treatment Facility [10]. Other five seed-composts are commercial. The data show that number and types of microorganisms isolated from each seed-compost are different. The seed A contains the highest number of bacteria which are in the order of 10^8 cfu/g while seed O-1 contains the lowest number, about 10^6 cfu/g. For the counts of molds, it is found that seed T-A contains the highest counts. On the other hand, seed O-2 contains the lowest counts of molds. Seed R contains the highest number of actinomycetes while the lowest counts of those are obtained in seed O-1. In case of yeasts, they are undetectable level in seeds T-A and R. While the highest counts are obtained in seed O-1.

Table 3. Number of microorganisms in seed-compost

kind of micro-organisms	Seed name					
	A	B	T-A	O-1	O-2	R
TBC	5.7×10^8	8.0×10^7	6.7×10^7	7.0×10^6	1.7×10^7	4.0×10^7
TM	6.8×10^4	5.0×10^4	6.7×10^5	2.3×10^4	1.3×10^2	2.6×10^4
TY	8.3×10^5	4.4×10^5	<D	2.2×10^6	4.4×10^4	<D
TA	1.4×10^5	2.8×10^6	4.5×10^5	4.7×10^4	1.5×10^5	2.3×10^7

TBC: Total bacterial counts (cfu/g), TM: Total molds (cfu/g), TY: Total yeasts (cfu/g), TA: Total actinomycetes (cfu/g), <D: undetectable.

3.1.2 Effect of temperature on microorganisms in seed-composts

Number of microorganisms in six seed-composts at various incubation temperatures are shown in Table 4. The number and types of

To enumerate the F. oxysporum in the inoculated sample from a vial tube, three grams of sample was suspended into 27 ml of sterilized water containing 0.01% Tween 20 in sterilized polyethylene bag. The mixture was homogenized with a stomacher Lab-blender 80 for 2 min. After filtration through sterilized four-layer gauze, 0.2 ml of appropriate dilution was plated in duplicate onto the malt-yeast extract agar. All plates were incubated at 30 °C for 3 days. The number of F. oxysporum was obtained as the average of two samples in each vial tubes.

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TBC	5.7×10^8	8.0×10^7	6.7×10^7	7.0×10^6	1.7×10^7	4.0×10^7
TM	6.8×10^4	5.0×10^4	6.7×10^5	2.3×10^4	1.3×10^2	2.6×10^4
TY	8.3×10^5	4.4×10^5	<D	2.2×10^6	4.4×10^4	<D
TA	1.4×10^5	2.8×10^6	4.5×10^5	4.7×10^4	1.5×10^5	2.3×10^7

TBC: Total bacterial counts (cfu/g), TM: Total molds (cfu/g), TY: Total yeasts (cfu/g), TA: Total actinomycetes (cfu/g), <D: undetectable.

3.1.2 Effect of temperature on microorganisms in seed-composts

Number of microorganisms in six seed-composts at various incubation temperatures are shown in Table 4. The number and types of

those are observed to be maximum at the temperature around 30 - 40 °C. At the temperature above 50 °C, the number and types of microorganisms decreases, particularly, most of microorganisms are destroyed at temperature 65 °C,. It is noticeable that most of the seed-composts contains high number of thermophilic microorganisms which can survive even at 65 °C. But, microorganisms in seed-compost A are almost completely destroyed at temperature 65 °C. According to those results, it can be seen that the incubation temperatures directly affect on microorganisms in the seed-composts. The high growth of the microorganisms is obtained at low incubation temperature.

Table 4. Number of microorganisms detected from seed-compost at various temperature

Seed-compost	Number of microorganisms (cfu/g)	Temperature (°C) 30	40	50	65
A	TBC	5.7×10^{8}	3.3×10^{7}	1.2×10^{7}	$<1.7\times10^{3}$
	TM	6.8×10^{4}	$<1.7\times10^{2}$	$<1.7\times10^{2}$	<D
	TY	1.1×10^{5}	2.5×10^{4}	$<1.7\times10^{2}$	<D
	TA	1.1×10^{5}	1.1×10^{5}	3.1×10^{6}	<D
B	TBC	8.0×10^{7}	1.1×10^{8}	2.5×10^{7}	4.0×10^{6}
	TM	5.0×10^{4}	7.3×10^{5}	$<1.7\times10^{2}$	<D
	TY	4.4×10^{5}	5.5×10^{4}	2.2×10^{3}	<D
	TA	2.8×10^{6}	3.1×10^{7}	9.2×10^{6}	7.5×10^{5}
T-A	TBC	6.7×10^{7}	1.8×10^{7}	3.8×10^{6}	2.6×10^{6}
	TM	6.7×10^{5}	$<1.7\times10^{2}$	<D	<D
	TY	<D	<D	<D	<D
	TA	4.5×10^{5}	1.0×10^{6}	3.0×10^{5}	<D
O-1	TBC	7.8×10^{6}	5.1×10^{6}	6.7×10^{6}	1.6×10^{6}
	TM	2.3×10^{4}	2.7×10^{4}	$<1.7\times10^{2}$	<D
	TY	2.2×10^{6}	2.8×10^{6}	1.9×10^{4}	<D
	TA	4.7×10^{4}	2.0×10^{6}	1.3×10^{5}	$<1.7\times10^{3}$
O-2	TBC	1.7×10^{7}	3.5×10^{7}	1.8×10^{7}	7.8×10^{5}
	TM	1.3×10^{2}	6.7×10^{2}	<D	<D
	TY	4.4×10^{4}	1.8×10^{3}	<D	<D
	TA	1.5×10^{5}	<D	<D	<D
R	TBC	4.0×10^{7}	6.4×10^{7}	3.0×10^{7}	1.7×10^{6}
	TM	2.6×10^{4}	4.5×10^{3}	6.3×10^{3}	<D
	TY	<D	<D	<D	<D
	TA	2.3×10^{7}	2.4×10^{7}	1.6×10^{7}	$<1.7\times10^{2}$

TBC: Total bacterial counts, TM: Total molds, TY: Total yeasts, TA: Total actinomycetes, <D: Undetectable level.
$<1.7\times10^{2}$ means less than 1.7×10^{2}.

3.1.3 Effect of temperature on composting

CO_2 evolution rate was calculated as the weight of CO_2 evolved from 1 kg of volatile solid in sludge per hour. Carbon conversion (C-conversion) was calculated as the ratio of carbon evolved as CO_2 during the fermentation to total carbon in sludge. The examples of CO_2 evolution and C-conversion during composting at various temperatures are shown in Fig. 3 and 4, respectively.

Table 5 shows the experimental results of irradiated sewage sludge composting at four different temperatures by using different seed-composts. The variation of moisture content (MC), volatile solid (VS) and pH of the composts produced from each seed-compost at various temperatures are observed especially at high temperature. At 65 °C, pH of the composts became low. This probably due to low growth of microorganisms because pH increases by production of NH_3 during fermentation. MC of the composts is very low at high temperature because the water evaporation is large at high temperature. The CO_2 evolution rate and C-conversion are maximum around 40 - 50 °C in most cases. Although the seed B contains large number of thermophilic microorganisms, the CO_2 evolution rate and C-conversion at 65 °C are very low as same as other cases. Therefore, it can be concluded that the optimum temperature of composting of irradiated sludge is around 40 - 50 °C. This result is consistent with the report of Kawakami et al. [3].

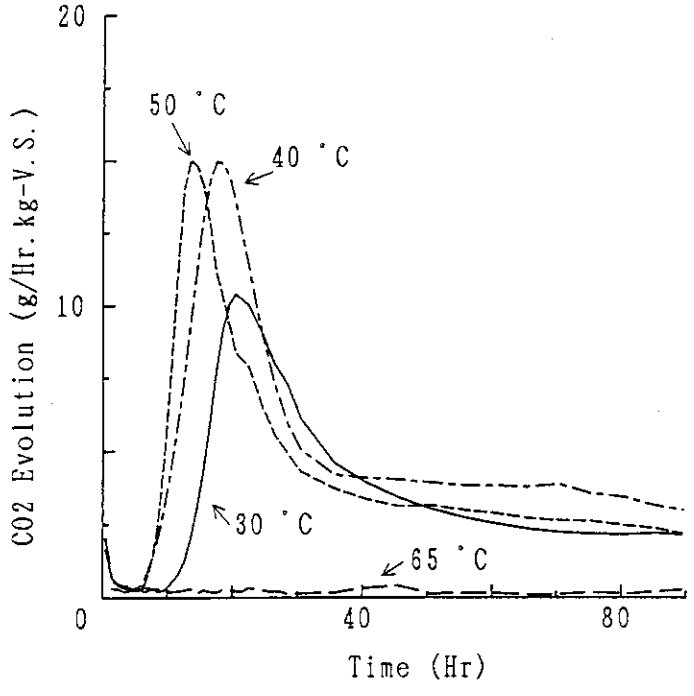

Fig. 3 An example of CO_2 evolution during composting

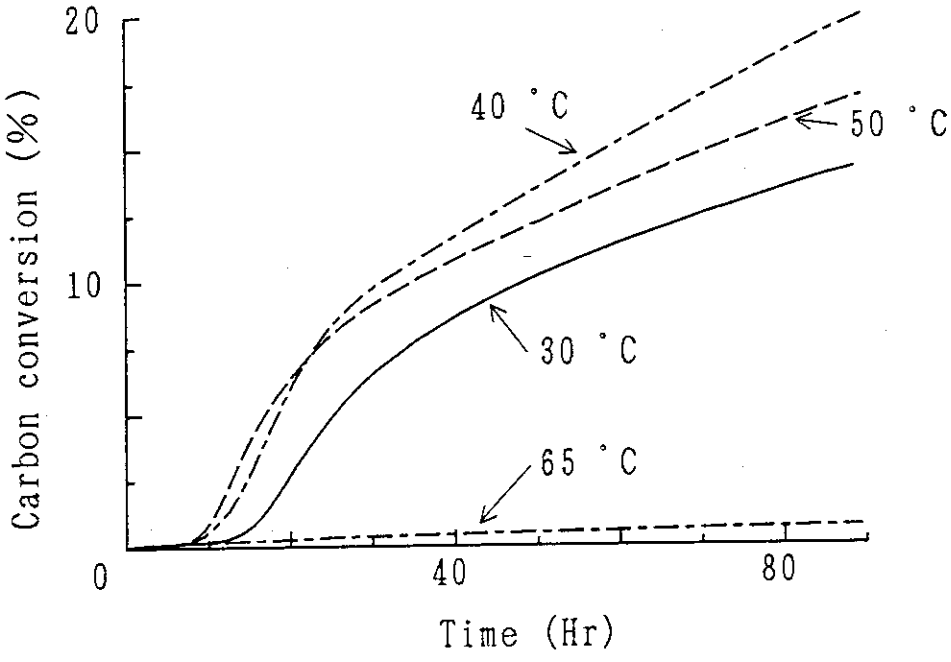

Fig. 4 An example of C-conversion during composting

Table 5. Composting at different temperatures by using different seed-composts

Seed-compost	T (°C)	Sludge property		Compost property			Max. CO_2 evol. rate (g/h/kg.V.S.)	C-conv. (%)
		MC(%)	VS(%)	MC(%)	VS(%)	pH		
A	30	71.4	57.4	48.4	20.8	8.5	7.6	12.8
	40	70.9	56.9	43.3	22.3	8.5	10.1	12.8
	50	71.4	57.4	48.4	20.8	8.5	7.6	12.8
	65	72.1	57.1	46.2	26.8	6.3	*	0.3
B	30	71.4	57.4	53.3	21.1	8.7	6.6	9.5
	40	70.9	56.9	47.8	21.9	8.5	6.4	11.2
	50	71.4	57.4	45.1	18.4	8.6	7.6	13.7
	65	72.1	57.1	40.4	24.6	7.7	3.0	7.3
T	30	71.4	57.4	42.3	22.4	8.2	6.2	9.9
	40	70.9	56.9	45.4	20.8	8.3	9.2	13.7
	50	71.4	57.4	47.8	18.6	8.4	11.2	16.3
	65	72.1	57.1	27.6	20.7	6.3	0.7	0.9
O-1	30	71.4	57.4	45.6	23.4	8.8	10.4	14.2
	40	70.9	56.9	46.5	22.3	7.7	15.0	16.8
	50	71.8	56.7	42.8	20.8	8.6	14.9	19.9
	65	70.9	56.7	34.2	26.8	6.3	*	0.7
O-2	30	71.4	57.4	44.4	22.0	8.6	6.4	9.7
	40	70.9	56.9	45.4	22.8	8.3	7.8	12.4
	50	71.8	56.7	37.6	17.9	8.3	16.9	16.0
	65	70.9	56.9	25.1	20.7	6.4	*	0.9
R	30	71.4	57.3	44.3	17.6	8.4	7.9	10.7
	40	70.9	56.9	45.7	18.5	8.4	11.7	12.6
	50	71.8	56.7	44.0	18.1	8.5	9.5	12.9
	65	70.9	56.9	29.8	21.7	6.7	1.2	2.1

T: Composting temperature, MC: Moisture content, VS: Volatile solid content, C-conv.: Carbon-conversion, Max. CO_2 evol.: Maximum CO_2 evolution, *: No fermentation occurred.
Composting time: 140-143 hr at 30 °C, 92-98 hr at 40 °C, 92-95 hr at 50 and 65 °C.

As shown in Table 6, number and types of microorganisms in composts produced at various temperatures from different seed-composts decrease with increase in temperature. In all cases, molds and yeasts are at undetectable level in composts produced at 65 °C. The same results are also observed on seed composts as shown in Table 4.

Table 6. Microorganisms in composts produced from various seed-composts

Seed-compost	Number of microorganisms (cfu/g)	Temperature (°C) 30	40	50	65
A	TBC	4.5×10^{9}	$3.\times10^{7}$	1.8×10^{7}	$5.0\ 10^{2}$
	TM	4.0×10^{6}	$<1.7\times10^{3}$	<D	<D
	TY	$<1.7\times10^{2}$	$<1.7\times10^{2}$	<D	<D
	TA	2.0×10^{6}	$<1.7\times10^{2}$	$<1.7\times10^{2}$	<D
B	TBC	2.1×10^{10}	1.0×10^{9}	1.2×10^{9}	1.2×10^{5}
	TM	1.0×10^{6}	$<1.7\times10^{3}$	<D	<D
	TY	$<1.7\times10^{2}$	$<1.7\times10^{2}$	<D	<D
	TA	1.0×10^{8}	7.2×10^{6}	1.5×10^{6}	<D
T-A	TBC	3.7×10^{9}	4.9×10^{7}	5.2×10^{6}	3.5×10^{4}
	TM	1.8×10^{6}	$<1.7\times10^{3}$	<D	<D
	TY	$<1.7\times10^{2}$	$<1.7\times10^{2}$	<D	<D
	TA	1.7×10^{8}	2.1×10^{6}	4.3×10^{6}	1.2×10^{3}
O-1	TBC	9.5×10^{8}	1.3×10^{9}	8.8×10^{6}	3.6×10^{4}
	TM	2.0×10^{5}	<D	<D	<D
	TY	$<1.7\times10^{2}$	3.3×10^{2}	$<1.7\times10^{2}$	<D
	TA	1.0×10^{6}	4.6×10^{4}	1.4×10^{6}	<D
O-2	TBC	2.2×10^{9}	7.3×10^{7}	2.0×10^{6}	9.7×10^{5}
	TM	1.6×10^{6}	5.0×10^{2}	$<1.7\times10^{2}$	<D
	TY	$<1.7\times10^{2}$	2.7×10^{2}	$<1.7\times10^{2}$	<D
	TA	3.5×10^{8}	7.5×10^{5}	<D	<D
R	TBC	1.5×10^{9}	3.2×10^{8}	4.1×10^{6}	1.1×10^{5}
	TM	1.5×10^{5}	<D	$<1.7\times10^{2}$	<D
	TY	$<1.7\times10^{2}$	3.3×10^{2}	$<1.7\times10^{2}$	<D
	TA	3.8×10^{8}	2.3×10^{6}	1.5×10^{5}	<D

TBC: Total bacterial counts, TM: Total molds, TY; Total yeasts, TA; Total actinomycetes, <D; Undetectable level.

The highest number and varieties of microorganisms in composts are observed at temperature around 30 - 40 °C but the optimum temperature of composting from the view-point of CO_2 evolution is around 40 - 50 °C. Although temperature at 30 °C is not optimum for composting, large number and varieties of microorganisms in the composts are observed. Therefore, high temperature composting is effective for quick stabilization of sludge by decomposing organic substances in sludge. On the other hand, low temperature composting is useful to produce compost with various kinds of microorganisms including those with suppressive effect on plant pathogenic microorganisms.

3.2 Preliminary study of suppressive effect of composts on F. oxysporum

3.2.1 Suppressive effect of seed-composts on F. oxysporum

1) Agar assays for antagonists

From six seed-composts as mentioned in Table 1, those were tested for antagonistic effect by first approach mentioned before. It was found that none of seed-composts showed growth inhibition of F. oxysporum. However, there are various kinds of mechanism of biological control as described by Millner et al. [11]. The method of first approach is only effective when the microorganisms can produce antibiotics or such kinds of suppressive substances.

2) Compost inoculation assays

As shown in case 1 of Table 7, five seed-composts out of six showed reduction of F. oxysporum. Seed O-1 and O-2 show the appreciable reduction of F. oxysporum while seed R does not show the reduction. It is noticeable that moisture contents (MC) of Seeds A, O-1 and O-2, which show reduction level of F. oxysporum, are very low. As known that spores of F. oxysporum can tolerate in dry condition, therefore, the appreciable declined level should not have occurred by only the effect of moisture content. So that in case of seeds O-1 and O-2, antagonist effect should be considered as a factor of the reduction level of F. oxysporum. Case 2 of Table 7 shows effect of seed-compost on F. oxysporum after adjusting MC to be 50 %. It is observed that seeds A and O-1 show the appreciable reduction of F. oxysporum. Seed O-2, on the other hand, does not show the reduction.

Table 7. Reduction of F. oxysporum by seed-composts

	Seed name	Seed property		Number of F. oxysporum (cfu/g)	
		MC(%)	pH	0 day	7 days
Case 1	A	19.1	8.2	1.7×10^{5}	3.1×10^{4}
	B	62.5	6.1	2.1×10^{5}	6.3×10^{4}
	T	60.5	7.6	1.9×10^{5}	7.1×10^{3}
	O-1	13.5	6.3	1.4×10^{5}	$<2.5\times10^{2}$
	O-2	15.0	6.8	1.7×10^{5}	$<2.5\times10^{2}$
	R	47.8	6.4	2.5×10^{5}	1.6×10^{5}
Case 2	A	50.0	8.2	1.4×10^{5}	7.5×10^{2}
	O-1	50.0	6.3	9.4×10^{4}	$<2.5\times10^{2}$
	O-2	50.0	6.8	1.6×10^{5}	5.0×10^{5}

MC: Moisture content.

3.2.2 Suppressive effect of composts on F. oxysporum

Table 8 shows reduction of F. oxysporum by composts produced at various temperature from different seed-composts. Five composts out of six produced at 30 °C show inhibition of F. oxysporum. The compost from seed O-2 shows the highest reduction with 2 log cycles. On the other hand, the compost from seed R does not show the suppressive effect.

Table 8. Reduction of F. oxysporum by composts

Seed name	Composting condition		Compost property			Number of F. oxysporum (cfu/g)	
	T(°C)	Time(h)	MC(%)	VS(%)	pH	0 day	7 days
A	30	140	48.4	20.8	8.5	1.9×10^{5}	5.1×10^{4}
	40	92	43.3	22.3	8.5	1.3×10^{5}	1.7×10^{5}
	50	95	48.4	20.8	8.5	1.9×10^{5}	3.0×10^{6}
	65	94	46.2	26.8	6.3	1.4×10^{5}	2.4×10^{5}
B	30	140	53.3	21.1	8.7	1.7×10^{5}	6.0×10^{4}
	40	92	47.8	21.9	8.5	1.5×10^{5}	1.5×10^{5}
	50	95	45.1	18.4	8.6	2.0×10^{5}	1.1×10^{5}
	65	94	40.4	24.6	7.7	1.7×10^{5}	1.7×10^{7}
T	30	140	42.3	22.4	8.2	2.1×10^{5}	6.3×10^{4}
	40	92	45.4	20.8	8.3	1.6×10^{5}	1.5×10^{5}
	50	95	47.8	18.6	8.4	1.6×10^{5}	5.6×10^{5}
	65	92	27.6	20.7	6.3	1.9×10^{5}	2.6×10^{7}
O-1	30	143	45.6	23.4	8.8	2.0×10^{5}	3.5×10^{4}
	40	98	46.5	22.3	7.7	1.5×10^{5}	2.7×10^{5}
	50	92	42.8	20.8	8.6	9.6×10^{5}	9.6×10^{5}
	65	95	34.2	26.8	6.3	1.5×10^{5}	5.3×10^{6}
O-2	30	143	44.4	22.0	8.6	2.3×10^{5}	2.3×10^{3}
	40	98	45.4	22.8	8.3	1.1×10^{5}	2.9×10^{4}
	50	92	37.6	17.9	8.3	1.0×10^{6}	4.7×10^{6}
	65	95	25.1	20.7	6.5	1.1×10^{5}	1.2×10^{7}
R	30	143	44.3	17.6	8.4	2.3×10^{5}	2.6×10^{5}
	40	98	45.7	18.5	8.4	1.5×10^{5}	1.7×10^{5}
	50	92	44.0	18.1	8.5	9.5×10^{5}	1.1×10^{6}
	65	95	29.8	21.7	6.7	1.2×10^{5}	2.5×10^{4}

T: Temperature, MC: Moisture content, VS: Volatile solid.

It can be also seen that the compost produced at 40 ˚C by using seed O-2 is suppressive to F. oxysporum. The reduction, however, is only 1 log cycle which is less than that observed from the results at 30 ˚C. According to the results, it can be concluded that the compost produced at 30 ˚C shows the higher suppressive effect on F. oxysporum than the others.

In case of the compost produced at 65 ˚C from seed R, it shows 1 log cycle reduction of F. oxysporum but no reduction is observed from other composts produced at 65 ˚C. In general, growth suppression on F. oxysporum by compost depends on microbes in the compost. As mentioned before, growth of microorganisms in compost from seed R was also large at low temperature as same as other seed-composts but no suppressive effect was observed at low temperature. Furthermore, seed R does not show suppressive effect on F. oxysporum as shown in Table 7. Therefore, the occurrence of reduction level in the compost produced from seed R can not be considered as effect of suppression by microorganisms.

The effect of suppression by composts on F. oxysporum was not clarified by this study using the compost inoculation assays. Therefore, further study should be required.

4. Conclusion

1) Microorganisms in seed composts showed higher growth at low temperature.

2) Optimum temperature for composting of irradiated sewage sludge was found to be around 40 - 50 ˚C as based on CO_2 evolution rate.

3) Composts produced at low temperature contained lager number and varieties of microorganisms than those produced at the higher temperature than 50 ˚C.

4) Suppression of F. oxysporum was observed in composts produced from seed-composts with suppressive effect and higher suppression was observed from composts produced at low temperature.

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