

## Quality Assessment of Compost from Laboratory Reactor by Germination Index

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### ABSTRACT

Composting is a useful waste treatment for environmental problem, but composting experiment is challenging due to large-scale requirement. Thus, laboratory-scale reactor is of interest for composting experiment. The capability of laboratory-scale reactor to simulate large-scale composting was determined by assessing the quality of produced compost via germination index. The laboratory-scale reactors were set up from mini-reactor (internal volume 101 mL) with fixed temperature inside an incubator and two air flow rates (5.5 mL/min, 1.375 mL/min). Rabbit food was chosen as model food waste for reproducible results. During the course of the experiment, the progress of CO<sub>2</sub> evolution rate from laboratory-scale composting was similar to that of large-scale composting. After 10 days of composting, both air flow rates produced compost with organic matter degradation more than 70%. Throughout the course of the composting, sample was collected for germination index test. The germination index test showed that raw materials did undergo transformation to become less phytotoxicity. The germination index of final product was higher than 60%, indicating a matured and stabled compost. Thus, the composting in laboratory-scale system can simulate the progress in large-scale composting.

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## 1. Introduction

Composting is the biologically aerobic degradation and stabilization of organic matters, with the development of high temperature as a result of heat emitted from microbial activity. If the temperature of the composting pile is more than 50 °C for several days, final product of composting will be free of pathogens and active plant seeds, and can be applied to farmland [1]. Therefore, composting has been studied extensively over time for sustainable development.

The choice for composting experiments is either large- or laboratory-scale. Large-scale composting does not require complicated equipment to carry out, but it does require considerable raw materials piled up in an open space. As a result, its disadvantages are time and labor-consuming, controlling difficulty, high heterogeneity, fluctuating conditions (such as temperature, rainfall...). Laboratory-scale is developed to simulate the conditions of large-scale composting and would mitigate all the problems of large-scale composting, but it is often argued to not achieve product similar to large-scale composting because it does not completely generate the organic matter degradation of large-scale composting [2].

The objective of this study is to simulate the large-scale composting with laboratory-scale reactor, then evaluate the produced compost quality via phytotoxic activity using the germination index. Germination index (GI) is one of the bioassay methods to assess compost applicability to farmland [3]. The reason is compost might contain toxic substances to plant growth, such as volatile organic acids [4], ammonium ion [5]..., which are the intermediates of organic matter degradation during composting. The detection of these substances is either time-consuming (plant growth experiment) or costly. GI was introduced as quick, simple, inexpensive, and effective method for analysis of compost toxicity [6]. Specific type of seeds is submerged in either compost extract or water (control sample) until the seeds start germinating and growing after a certain time. The effect of toxic substances in compost on seed

germination is analyzed by difference in length of seed root and number of seed germinated between compost extract and water. In order to collect sample for GI, a laboratory-scale reactor system was designed to achieve consistent composting conditions. This system comprises multiple mini-reactors installed within an incubator, allowing precise control over the composting temperature. This experimental system is crucial to ensure uniform conditions are maintained throughout the composting course. The organic matter degradation of microbial activity was monitored via CO<sub>2</sub> evolution rate. Once the composting was completed, the produced compost was tested by germination index for maturity and stability. The result of this study would suggest a laboratory-scale reactor with better control and reproducible experimental results without the need for large space and materials.

The next sections are outlined as follows: Section 2 describes how to conduct the laboratory-scale composting and how to assess the produced compost by germination index; Section 3 explains the experimental results of the laboratory-scale composting and the compost assessment, and Section 4 concludes the important findings of this study.

## 2. Materials and Methods

### Composting material

The raw material for composting was prepared by mixing model food waste – rabbit food (Timothy, Easter Co. Ltd., Tatsuno, Japan) with bulking agent – sawdust, and seeding material (Alles G; Matsumoto Laboratory of Microorganism Co. Ltd., Matsumoto, Japan) at a ratio of 10:9:1 (dry weight basis). Rabbit food, having C/N ratio between 15 and 30, was chosen as model food waste [7]. The physicochemical properties of raw material and its components are presented in Table 1. The raw material was adjusted to 60% moisture content using water. The initial pH was adjusted to about 8.5 by adding Ca(OH)<sub>2</sub> to offset the low pH inhibition from microbial activity [8].

**Table 1.** Physicochemical properties of raw mixture and its components. A CHN corder (Micro corder JM10, J-Science Lab Co. Ltd., Kyoto, Japan) was employed for analysis of total carbon and nitrogen.

	Total C (%)	Total N (%)	C/N ratio	Moisture content (%)
Rabbit food	45.43	2.81	16.17	6.87
Sawdust	48.64	0.14	347.43	6.84
Seeding material	16.44	1.98	8.30	10.3
Raw material	48.8	1.68	28.93	60

### Composting operation

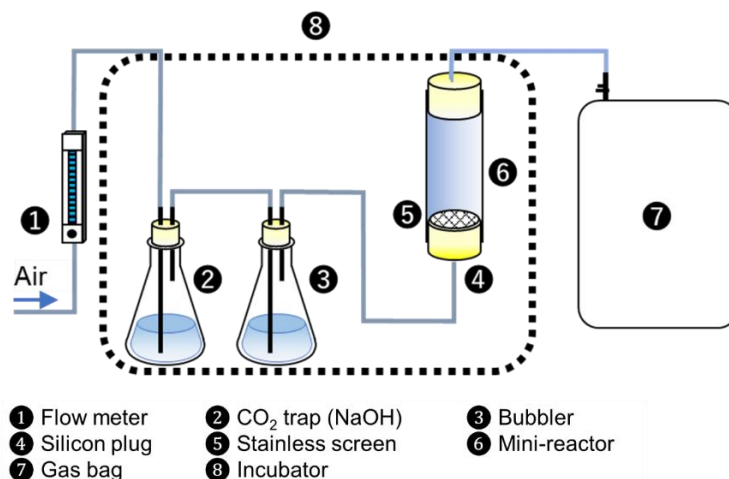
In Figure 1, the laboratory-scale reactor system was designed as follows: a cylindrical mini-reactor (Pyrex glass, inside diameter: 40 mm, height: 80 mm) was sealed with two silicone rubber stoppers at the top and bottom. Raw material (12 g, wet weight) was deposited in the mini-reactor, then the mini-reactor was fixed in an incubator (Model IS 800; Yamato Scientific Co., Ltd., Tokyo, Japan).

To achieve reproducible results, the laboratory-scale reactor system in this study was provided consistent conditions throughout the composting course as follows:

- Constant oxygen supply: two constant air flow rates 5.5 mL/min (Run HA) and 1.375 mL/min (Run LA) were introduced into the system. The inlet air was first introduced into one flask containing 0.1 M NaOH to remove CO<sub>2</sub>, then moisturized in the next flask containing water.
- Controlled temperature: The temperature of the mini-reactors was maintained in an incubator by setting up a temperature program: temperature initiated from 30 °C, and increased at a constant rate of 2.5 °C/h to 60 °C, then kept until the end of composting.
- Daily mixing: After 24 h, daily mixing of raw material for each mini-reactor was performed to accelerate composting, ensure oxygen distribution and homogeneity of raw material.
- Optimum moisture range: During mixing, by observing the changes of raw material, distilled water was added intermittently to ensure the moisture content of the compost was between 40 – 60% (optimum for composting) [9].

Gas bag (10 L, SMART bag PA AA-10; GL Sciences Inc., Tokyo, Japan) was connected to the mini-reactor to collect outlet gas. After 24 h, the gas collected inside the gas bag was analyzed for total accumulated volume and CO<sub>2</sub> concentration by a dry gas meter (DC-1C, Shinagawa Corporation, Tokyo, Japan), and a CO<sub>2</sub> sensor (Model RI-555, Riken Keiki, Tokyo, Japan), respectively. The organic matter degradation (the conversion of carbon) in this study was the degree of organic matter mineralized into CO<sub>2</sub> and calculated from the ratio between the accumulated CO<sub>2</sub> evolution (the sum of CO<sub>2</sub> evolution rate until a certain day) and the carbon content in rabbit food.

At the start of composting (day 0), eight reactors were prepared for replicating and analyzing the samples. On days 1, 3, 4, 7, and 10, one reactor was collected for physiochemical analysis (pH, moisture content), and germination index.



**Figure 1.** Setup of laboratory-scale reactor composting system

### Germination index

The progress of compost quality was analyzed via germination index of compost extracts [6]. In this study, germination index was determined by comparing the growth of komatsuna seeds (*Brassica campestris* L. var. *rapifera*froug, Takii Co. Ltd., Kyoto, Japan) in between compost extracts and water (control experiment). Compost samples from days 0, 1, 3, 4, 7 and 10 were homogenized with distilled water at a ratio of 1:9 (wet weight/weight) and then filtered through a 0.20 mm hydrophilic PTFE filter to obtain compost extracts. Ten milliliters of either compost extracts or distilled water were transferred into one disposable petri dish having 50 komatsuna seeds fixed on filter paper (Tanepita; Fujihira Co. Ltd., Tokyo, Japan). For each compost sample, GI was carried out in triplicates (three petri dishes). All dishes were lidded and transferred to an incubator without lighting for growing for 96 hours at 25 °C to avoid contamination. The germination index (GI) of the compost extracts was defined as follows:

$$GI (\%) = \left( \frac{G}{G_c} \right) \times \left( \frac{L}{L_c} \right) \times 100 \quad (1)$$

where G and G<sub>c</sub> are the number of germinated seeds growing in compost extract and distilled water, respectively; L and L<sub>c</sub> are the root length of each seed in the compost extract and distilled water, respectively.

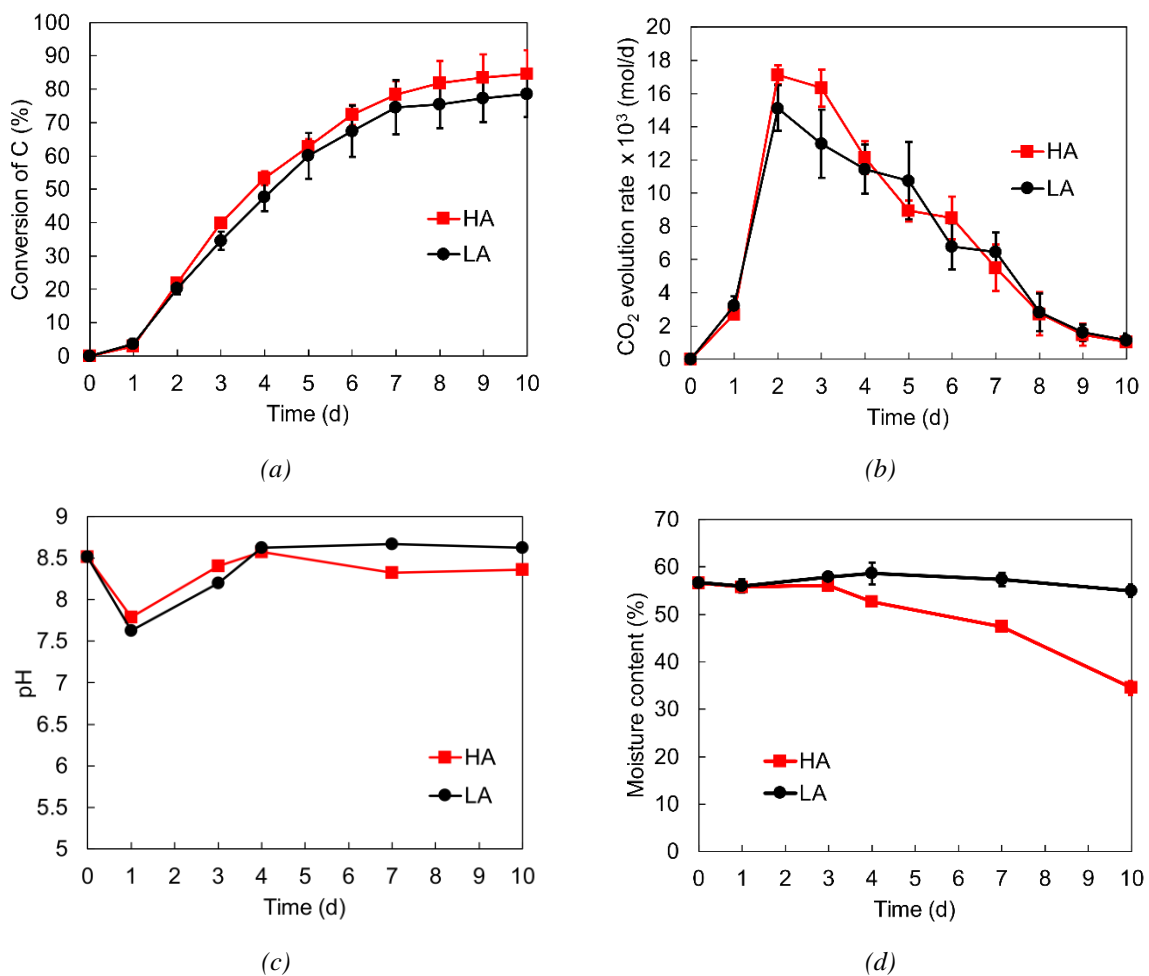
## 3. Results and Discussion

### 3.1. Organic matter degradation in laboratory-scale reactor

The time courses of the CO<sub>2</sub> evolution rate, conversion of C, pH, and moisture content of Runs HA and LA are shown in Figure 2. The shape of CO<sub>2</sub> evolution rate (Figure 2a) was similar for both runs as follows: During the first day of increasing from mesophilic to thermophilic temperature, the CO<sub>2</sub> evolution rates were slightly increased. From day 2 to day 7, large CO<sub>2</sub> emission was observed although the decreasing slope of CO<sub>2</sub> evolution rate of Run HA was steeper than that of Run LA. From day 8 to day 10, the CO<sub>2</sub> emission rates were negligible. The time course of CO<sub>2</sub> evolution rate in this study is

also similar to that of changes in CO<sub>2</sub> concentration in large-scale composting. However, it should be noted that the changes in CO<sub>2</sub> concentration in large-scale composting are not a quantitative parameter for monitoring the changes of organic matter degradation. The reason is the output flow rate in large-scale composting is usually not quantified due to composting being carried out in open space. The conversion of C (Figure 2b) in both runs was close to each other, reaching more than 70%, indicating the high efficacy of organic matter degradation. In large-scale composting, due to the difficulty of quantifying CO<sub>2</sub> emission, other parameters, such as volatile organic matter (VOM), or water-soluble carbon, are measured for changes in organic matter degradation [10]. Those parameters show declining trend as organic matters are degraded and released to environments. The conversion of C in this study shows increasing trend because it records the mineralizing of organic matters, which are released to surroundings in the form of CO<sub>2</sub>. Therefore, the results of this study regarding the time courses of organic matter degradation are consistent with previous studies for large-scale composting.

In both runs, the pH dropped in the first stage of composting and slowly rose to alkaline pH later (Figure 2c). The pH of the compost is the result of the organic matter degradation by microbial activity. As organic matters are metabolized, intermediate organic acids are produced and accumulated in the compost. Subsequently, those organic acids can be either further consumed by microorganisms or neutralized by alkaline compounds, such as ammonia, which is also the product of microbial activity. Additionally, as seen in Figure 2d, the optimum moisture content (40 – 65%) was successfully maintained when organic matter degradation happened vigorously (when CO<sub>2</sub> evolution rate was significantly high), thus the interference of fluctuating moisture was reduced with laboratory-scale reactors. Run HA on day 10 had moisture content of 35% because the added water was not enough to compensate for the loss of moisture in Run HA, which had higher air flow rate.

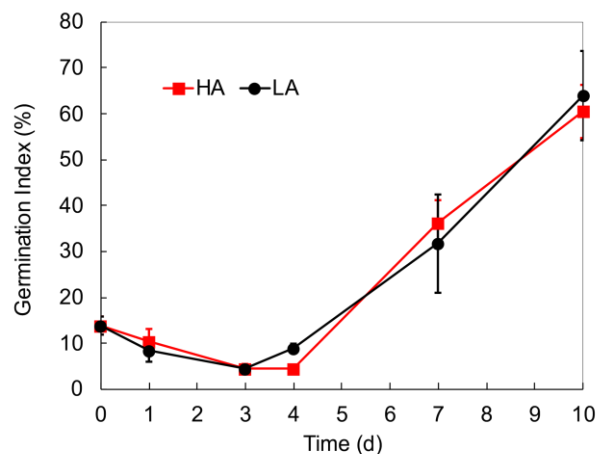


**Figure 2.** Time course of CO<sub>2</sub> evolution rate (a), Conversion of C (b), pH (c), and moisture content (d) of Run HA and LA

Generally, from the progress of CO<sub>2</sub> evolution rate, conversion of carbon, and pH, the laboratory-scale reactor was able to simulate the changes in organic matter in large-scale composting. Typically, during the first stage of composting, labile organic compounds, such as volatile fatty acids, are consumed by microorganisms, microorganisms start to grow, and small amounts of CO<sub>2</sub> are released. Once all the labile organic compounds are exhausted, more complex compounds such as carbohydrates, proteins, and fats, are consumed by the growing microorganisms. Due to microbial activities, heat is released into the composting pile, raising the heap temperature. Because of elevated temperature, reactions occur even more violently, and various compounds are mineralized into CO<sub>2</sub>, leading to significant emission of CO<sub>2</sub> (high CO<sub>2</sub> concentration in this stage). Additionally, the temperature usually reaches 50 to 60°C and lasts for weeks because the heat is retained inside the composting heap. This is why the thermophilic temperature in this study was maintained until the end of composting. After all organic matters are mineralized, only hardly degradable organic matters are left, such as cellulose, and lignin, which coincides with the small CO<sub>2</sub> emission.

### 3.2. Assessment of compost quality by germination index

The compost produced from laboratory-scale reactors is often questioned about its capability to apply to farmlands because of short composting time. For example, the composting time in this study was 10 days, compared to 30 days of large-scale composting. Thus, germination index was carried out to confirm that composting in laboratory-scale reactors would be able to reduce phytotoxicity of organic matter. As seen in Figure 3, the time course of germination index was in accordance with the time course of organic matter degradation, particularly CO<sub>2</sub> evolution rate and pH. At the early stage of composting (from day 1 to day 4), hydrolyzed and denatured compounds present in the compost. Low pH during this stage also indicates the presence of organic acids. At later stage, ammonium ions can also inhibit plant growth. From day 7 to day 10, all organic matters are mineralized and ammonium ions are evaporated in the form of ammonia, the GI increased, indicating that the compost is now suitable for plant growth. The course of GI is the inverse of CO<sub>2</sub> evolution rate as phytotoxins are mineralized. Compost from both runs achieved more than 60%, which is safe according to some regulations [11]. Interestingly, the GI are similar in both runs although their flow rates are different, which is consistent with the result of conversion of C. Thus, the use of laboratory-scale reactor is justified for the simulation of composting.



**Figure 3.** Germination index of Run HA and LA

## 4. Conclusions

Laboratory-scale reactor was able to simulate the course of organic matter degradation in composting. On the first day (starting stage), small CO<sub>2</sub> was released, and pH was slightly dropped. These results are explained by the microbial consumption of labile organic matters. After day 1 (middle stage), CO<sub>2</sub> emission peaked on day 2 and day 3, which corresponds to the vigorous organic matter degradation when microorganisms proliferate under high temperature. After day 2 and day 3 (final stage), CO<sub>2</sub> emission decreased and stabilized at low level due to the exhaustion of organic matter. After

day 1, pH increased probably due to the consumption of organic matter and/or the neutralization by alkaline compounds, such as ammonia, which are the intermediate of organic matter degradation. The changes in organic matter also reflected in germination index. During starting stage and middle stage, the GI of compost was below 20%, indicating the high number of organic compounds due to the ongoing organic matter degradation. In final stage, the GI of compost increased to about 60%, indicating the low number of organic compounds due to the ceasing of organic matter degradation. The compost produced in laboratory-scale reactor had low phytotoxic, which could be used for further analysis. Future studies should focus on how to standardize the GI test procedure for better application in regulation.

### Conflict of Interest

The authors declare no conflict of interest.

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