



Research Article

A Fast and Reliable Screening Method of Organic Materials for Crop Cultivation

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Keywords: germination index; organic waste; raw material screening; inhibitory effect

Abstract

Strategic utilization of organic material for agriculture is important for improve efficiency in organic agriculture. Waste from food processing is a good candidate for compost material, and the first critical step is to choose proper raw material. In this study, we tried to establish a reliable screening method by comparing growth of different plant parts in different crops in combination with serial dilution of raw material extracts. The raw materials compared here are tangerine pomace, herbal medicinal plant waste, bread, spent coffee ground (SCG). We also compared effect of organic fertilizer made of SCG, natural pesticides made of pyrethrum extract and neem oil. The results obtained from different kind of crops and plant parts give insight into growth enhancing or inhibitory effect of tested material which will be useful screening raw material to make organic fertilizer and crop protective materials.

Introduction

Recycling organic waste into organic fertilizers has been widely practiced worldwide which is a sustainable agricultural practice protecting the environment and saving cost at the same time. For proper composting processes to be achieved, a thorough designing of raw material composition and method according to the physico-chemical properties of the raw materials (Stabnikova *et al.*, 2005; Moldes *et al.*, 2006; Diacono *et al.*, 2019). There can be raw materials that can reduce crop growth or productivity, however, most components in the raw material are changed by

microorganisms during the composting processes (Chefetz *et al.*, 1996). Still, it is safe to be aware of the properties of the original materials and the possibility of inhibitory component in the material. The knowledge of that can help designing composting processes and assessment of the compost product altogether.

As well as organic fertilizer, crop protective materials made of natural resources are gaining attention for sustainable agricultural practices. However, there the natural sources can

contain unknown chemicals and the effects of crops yet to be analyzed.

Germination index (GI), is a widely used parameter for assessing maturity of fertilizer or toxicity of materials. Most of safeness of organic fertilizers are judged by GI, and there are some report successfully assessing the toxicity of organic fertilizers for crops (Paik *et al.*, 1998; Mitelut and Popa 2011; Selim *et al.*, 2012; Yoon *et al.*, 2014). However, using only germination rate has limitation recognizing inhibitory properties of materials (Cesaro *et al.*, 2014). Some researchers take root growth in their account and put forward more comprehensive screening results (Selim *et al.*, 2012; Yoon *et al.*, 2014). However, considering different responses among plant tissues and different crops, the bioassay method needs to be more improved.

In this study, we tried to overcome such limitation represented by GI, by using three distinctively different crop in their morphology and taxonomy, and measured the length of radicle and cotyledon (or hypocotyl) separately. This method will provide more comprehensive and detailed information of the material for wide range of material that will be applied to crop as well as raw material for organic fertilizers.

Materials and Methods

Food Process Waste and Other Organic Materials

Tangerine pomace was obtained from a juice factory located in Jeju island, South Korea. Breads were mixture of various types of bread selling in retail stores. Herbal medicinal plant wastes were obtained from a local oriental medical clinic. After decocting together, each different source materials were separated for the experiment. Spent coffee ground were from remaining of capsules for home espresso machine, and espresso and decaffeine were tested separately. In addition to raw materials for organic fertilizers, plant protective material also tested, pyrethrum extract (Youngbiwon, South Korea)

and neem oil (Ozoneem, India), and 1000x and 500x water dilution was used as 1x according to the manufacturer's recommendation (Fig. 1).

The raw materials were immersed in water and incubated on a shaking incubator for a couple of hours at room temperature, and then centrifuged for 5 minutes at 4000 rpm, room temperature, and the supernatant were used as extract. The extracts were applied in three serial dilutions to the test plate containing 0.8% agar (Table 1).

Germination and Growth Test

To facilitate measuring radicle and hypocotyl, square dishes of 12cm x 12cm x 1.5cm were used. Twenty milliliter of 0.8% agar containing each extract at three different concentrations were poured in the square dish and three seeds of three different crops were placed on the plate, and then the plates were placed vertically. The three crops use for the test were rice (*Oryza sativa* L., cv. Koshihikari), young radish (*Raphanus sativa* L., cv. Jindong), lettuce (*Lactuca sativa* L., cv. Jeokchima). Seeds were soak in the water about 5-10 minutes before placing them on the test plates.

In five days after sowing, length of radicle and hypocotyl (for young radish) or cotyledon (rice and lettuce) were measured. The relative elongation rate (%) were calculated based on that of control as 100% (Byeon *et al.*, 2020).

$$\text{Relative elongation rate} = \frac{\text{length in treated plate}}{\text{length in control plate}} \times 100$$

Replication and Statistics

Each treatment was performed on three replications, and each replication consisted with three seeds of three different crops. Lengths of radicle and cotyledon or hypocotyl measured on fifth day of the test were averaged and the standard deviation were calculated for comparison.

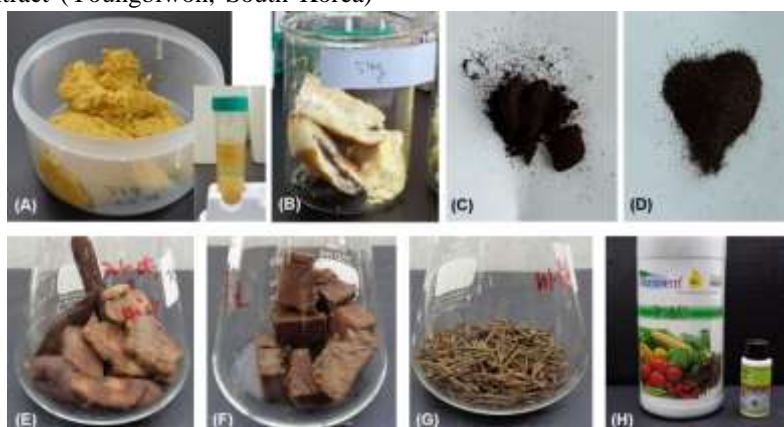


Fig. 1: Raw materials used in this study. Upper panel: (from left to right) tangerine pomace and the centrifuged extract, mixture of various types of bread, SCG (spent coffee ground), SCG compost. Lower panel: (from left to right) three waste of herbal medicine – peony root, *Pueraria* root, *Ephedra* and neem oil and pyrethrum extract.

Table 1: Chemical characteristics of raw material extracts and concentration of extracts and treatment

Raw material	Moisture content (%)	pH	EC (uS/cm)	Extract concentration	Final concentration
Tangerine pomace	86.9	4.1	3040	10% extract	0.5 / 1 / 1.5%
Bread	31.7	6.9	5620	25% extract	1.25 / 2.5 / 3.75%
Peony root	79.0	7.2	1635	30% extract	1.2 / 2.4 / 3.6%
<i>Pueraria</i> root	68.4	6.8	3750	30% extract	1.2 / 2.4 / 3.6%
<i>Ephedra</i>	31.2	7.1	2340	20% extract	1.2 / 2.4 / 3.6%
SCG-espresso	35.0	6.7	2480	10% extract	1, 2, 3%
SCG-decaffeine	40.0	6.8	2180	10% extract	1, 2, 3%
SCG fertilizer	18.0	6.4	9100	10% extract	1, 2, 3%
Pyrethrum extract *	NA	NA	NA	1/1000 dilution	0.5x / 1x / 2x
Neem oil*	NA	NA	NA	1/500 dilution	0.5x / 1x / 2x

* Pyrethrum extract and neem oil were recommended to use 1/1000 and 1/500 water diluted liquid and used those as 1x, respectively.
NA: Not analyzed

Results and Discussion

Effect of raw material extract on seedling growth

In this study, we compared growth rate of both radicle and cotyledon or hypocotyl on test plates containing extracts from various materials for organic fertilizers and insecticides. When the growth rate compared on the basis of that of the control plate, different growth rates were observed among seedlings grown in different extracts, as well as different kinds of crops (Fig. 2).

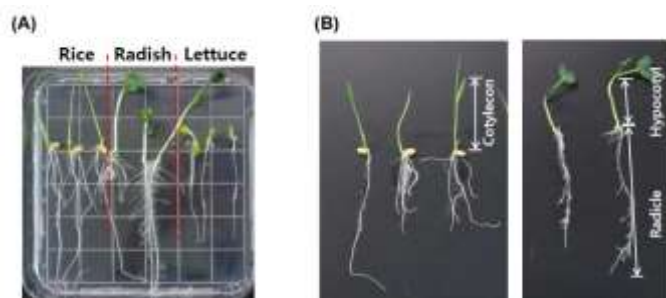


Fig. 2: Growth of seedlings and parameters. (A) Seedlings grown on the test plate (B) Measuring seedling growth: length of cotyledon, hypocotyl, and radicle

The growth rate of rice was affected little by different kinds of material, and radicle growth were slightly reduced in some medicinal plant extracts according to the concentration. Young radish also showed radicle growth inhibition by some medicinal plant extracts. On the contrary, growth of lettuce was not affected by medicinal plants extracts, however, that in tangerine pomace and bread extract were greatly inhibited. SCG extract enhanced growth of all three crops with increasing concentration, and this trend was stronger in organic fertilizer made of SCG.

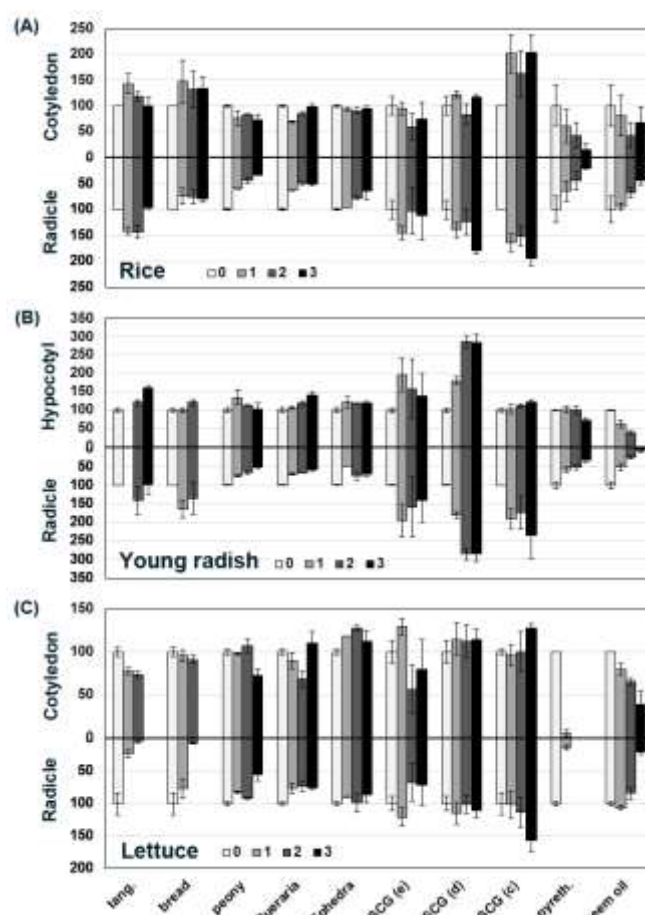


Fig. 3: Relative growth rate of seedlings. The relative growth rates of (A) rice, (B) young radish, and (C) lettuce were compared on the basis of non-treated control as 100%. The raw materials extract compared in the experiment are, tangerine pomace (tang.), bread, peony root, *Pueraria* root, *Ephedra* stem, SCG-espresso, SCG-decaffeine, SCG compost, pyrethrum extract (pyreth), neem oil. Numbers (0, 1, 2, 3) represent the concentration of extract added to the test plate and coincidence with a final concentration in Table 1.

Different Responses of Different Crops

The same substance can induce different response in different crops. Piccolo *et al.*, (1993) reported different germination ratio and seedling growth between lettuce and tomato induced by two humic substances. In their study, different responses were attributed to different cellular responses, such as cell elongation. It cannot be just one universal mechanism behind the different responses; however, this proves convenient way of screening unknown effects of the substance of inquiry. As the example, natural organic insecticide, pyrethrum extract and neem oil, resulted in different responses to different crops (Fig. 3). The growth inhibition effect of pyrethrum extract was severer in lettuce, however, that of neem oil was least severe on the same crop. On the contrary, neem oil most severely inhibited growth of young radish seedling, however, pyrethrum extract does not seem to have dose dependent growth inhibition effect on young radish seedlings. These results show that this type of screening can help early-stage crop growth by screening out inappropriate kind of substances for pest control. As these two materials are originated from plants, it is possible that seedling growth was inhibited by allelopathic effects. It is noteworthy that pyrethrum extract affected growth of lettuce seedlings (Fig. 3), and there are previous reports about allelopathic effects of plant raw material (Ali *et al.*, 2019) including chrysanthemum extract (Kil and Lee 1987; Beninger and Hall 2005; Salim and Abdalbaki 2017). The results of our study and previous report show that this screening method also can be applied to screen candidate materials for weed control.

Root Growth Inhibition of Plant Based Extracts

Most of the seedling showed similar morphology even in the growth was slightly reduced by treatment (Fig. 4). However, seedlings grown on test plates containing tangerine pomace and bread extracts were severely deformed (Fig. 5). The change of morphology was more obvious on radicle and hypocotyl than cotyledon. Root-hair was not observed in those defective radicles. The similar pattern of growth inhibition by allelopathic chemical were reported in previous study (Chon *et al.*, 2002). In their study, severe inhibition of root elongation in early seedling of alfalfa was accompanied by change of the root morphology. Based on these results, we conclude that the severe growth inhibition in fresh plant tissue based raw material can be at least partly attributed to allelopathic effect.

In this study, we were able to gain an overview of effect of raw material for organic fertilizers by comparing three different crops and using three serial dilutions of the extracts. Due to the change that occurs through composting process, these results may not be telling the effect of organic fertilizer from the raw material. Still, it offers valuable information for a possibility or existence of growth inhibitor that the users should be aware of. In that regard, the method we tried here

might be useful and applied to screening wide range of materials for fertilizer and plant protective material.

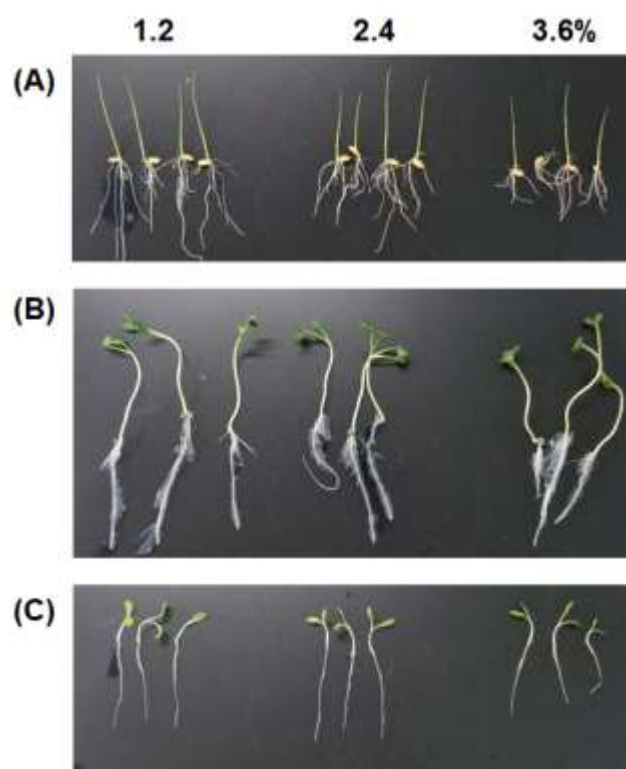


Fig. 4: Comparison of seedlings grown on test plates containing different concentrations of peony root extract. From top to bottom, rice, young radish, and lettuce.

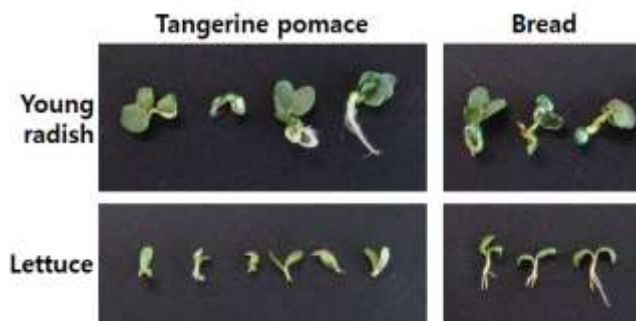


Fig. 5: The abnormal morphology of young radish and lettuce seedlings grown in test plates containing tangerine pomace or bread extract.

Author's Contribution

All authors contributed equally in all steps of the research and the finalization of this manuscript.

Conflict of Interest

The authors declare that there is no conflict of interest with present publication.

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