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Effect of Sandwich Compost Leachate on *Allium tuberosum* Seed Germination

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ABSTRACT

Food waste is a serious global issue, and one way to reduce the impact of food waste is by composting. Sandwich compost is a type of fermented food waste compost created with microbial fermentation; meanwhile, the composting leachate provides nutrients for plants. Studies have shown that seed germination may be enhanced when treated with sandwich compost leachate. Furthermore, few studies have been on sandwich compost leachate used for seed priming. The objective of this study was to determine the effect of varying leachate concentrations of food waste sandwich compost and priming durations on the performance of Chinese chive (*Allium tuberosum*) seed germination. Chinese chive (*Allium tuberosum*) was chosen as the test crop. It is widely used as a flavouring herb with high economic potential; however, its seed germination time is long and requires pre-treatment such as crushing and seed priming to speed up the germination process. The study used four replications and a complete randomisation design (CRD). The seeds were exposed to different percentages of sandwich compost leachate (0.0%, 0.2%, 0.4%, 0.6%, 0.8%, and 1.0%) and priming duration (4, 8, and 12 hours). A significant interaction between the bio-nutri-priming concentration and priming duration was demonstrated by measuring the standard error of germination rate ($S_{\overline{V}}$) and corrected germination rate index ($S_{corrected}$). A longer bio-nutri-priming duration was key for a higher seed vigour index. The bio-nutri-priming concentration and priming duration, however, had no significant interaction.

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Longer bio-nutri-priming durations were recommended to obtain better germination performance of Chinese chive. The study showed that a twelve-hour bio-nutri-priming duration and a 0.6 % leachate concentration significantly enhanced the Chinese chive seed germination and helped break seed dormancy.

Keywords: Bio-nutri-priming, Bokashi, Chinese chive, kucai, seed germination, seedling vigour index

INTRODUCTION

Food waste is a serious problem around the globe. Hence, food waste utilisation is vital to reduce the environmental impact of food waste. Most people are not vegetarian; thus, composting methods that accept meat and dairy compost is crucial. The sandwich compost method utilises meat and dairy waste products without attracting pests at home. This form of food waste management could extend landfill life. Furthermore, leachate derived from food waste sandwich compost is considered an eco-friendly source by recycling nutrients for food production. The use of food waste in seed priming as the raw material meets the United Nations Sustainable Development Goals, including reducing poverty, hunger, and sustainable consumption.

Seed priming is a common solution to improve seed germination performance. Priming is an adjustment of water potential, which allows for seed imbibition but prevents germination. Biopriming, a mix of beneficial microbes and bioactive molecules, is associated with endophytic connections between flora and specific microbial. Biopriming is a sustainable method to support plant growth and development (Toribio et al., 2021). For instance, phytohormones production, abiotic and biotic stress resistance, and germination performance were enhanced by biopriming (Makhaye et al., 2021; Moeinzadeh et al., 2010; Paparella et al., 2015). Biopriming has significantly enhanced seed germination and plant growth performance of bread, wheat, and sunflower (Liela et al., 2010; Moeinzadeh et al., 2010).

Nutrient seed priming with molybdenum, zinc, boron, and phosphate was widely studied in Asian countries such as India, Nepal, Pakistan, and Bangladesh (Harris et al., 2001). Nutrient seed priming enhanced nutrient-use efficiency, photosynthetic rates, and translocation of reserves in an integrated manner (Davis & Quick, 1998). Surprisingly, the wheat yield increased up to 36% (Harris et al., 2001). Nutrient seed priming not only showed a positive effect in wheat seeds but also in corn seeds (Harris et al., 2001; Imran et al., 2013; Rasool et al., 2019), barley (Ajouri et al., 2004), and mung beans (Shah et al., 2012). Micronutrient seed priming also significantly enhanced the tolerance of corn to abiotic stress like salinity (Imran et al., 2018).

Therefore, the approach of bio-nutri-priming could shorten the priming duration with different concentrations of the leachate. Biopriming showed a positive effect on seed germination performance, particularly sandwich compost leachate (Bisen et al., 2015). Sandwich compost leachate is the by-product of fermented composting, resulting in nutritive liquid leachate enriched with fermentative microbial. Therefore, sandwich compost leachate has the potential to improve seed germination performance by biopriming. The biopriming duration and leachate concentration are crucial in seed biopriming with sandwich compost leachate. Biopriming with sandwich compost leachate increased plant nutrient uptake and enhanced the stem diameter of tomato transplants by up to 13% (Olle, 2020).

Chinese chives (*Allium tuberosum*) were used as a test crop in this study. They are a widely used allium with aromatic flavoured leaves (sulphur-containing compounds) (Wang et al., 2008). Chinese chives have many health benefits, such as anti-diabetic and hepatoprotective properties (Tang et al., 2017). In addition, they are produced vegetatively with nonedible storage rhizome normally (Kamenetsky & Rabinowitch, 2017). Therefore, propagating the seed can increase genetic diversity. Nonetheless, the seed germination period is long, generally between 7 to 14 days. Usually, the seeds are primed between 12 to 24 hours to enhance germination. However, despite undergoing the priming process using different solutions, a longer priming period was required to enhance the germination performance. For instance, Chinese chives primed with 100 mg L^{-1} of gibberellin for 12 to 24 hours showed a higher germination performance (Sun et al., 2010).

Thus, the objective of this study was to determine the effect of varying leachate concentrations of food waste sandwich compost and priming durations on the performance of Chinese chive (*Allium tuberosum*) seed germination.

MATERIALS AND METHODS

Treatments and Experiment Design

The experiment was carried out at Universiti Putra Malaysia (UPM) with coordinates 2°59’34.0”N, 101°42’52.3”E. The treatments consisted of six varying concentration percentages of sandwich compost leachate (0%, 0.2%, 0.4%, 0.6%, 0.8%, and 1.0%) (Table 1), and three priming durations (4, 8, and 12 hours). Seventy-two experimental units were arranged using a completely randomised design (CRD) with four replications. Each replication consisted of 30 seeds. At room temperature, the seeds were germinated in diameter (∅) a 9 cm petri dish with two layers of moist tissue paper. The experiment was conducted at 27 ± 1°C under a 16:8 h light/dark photoperiod. The tissue paper was kept moist by spraying tap water every four hours.

Germinated seeds were counted daily until day ten. Then, daily counts of seedlings during the germination test were performed, whereby the seeds were considered to have germinated when there was visible radicle protrusion of at least 0.2 cm. Germination traits were calculated using the germination metrics package run in the R-program statistical software, which includes germination percentage (GP), standard error of germination rate ($S_{\overline{V}}$), germination rate as the reciprocal of the median time (V_{50}), corrected germination rate index ($S_{corrected}$), germination index (GI), and peak value (PV) (Aravind et al., 2019).

Table 1
Physiochemical parameter of tap water and sandwich compost leachate

Physiochemical parameter	Tap water	Sandwich compost leachate
pH	6.98±0.02*	4.78±0.011
Electric conductivity (dS m^{-1})	0.134±0.00	0.3357±0.0003
Total dissolved salt (mg L^{-1})	85.76±0.00	22.19±0.2133
Osmotic potential (bar)	0.04824±0.00	0.0125±0.0001
Total N (%) (Distillation and titration)	$0.00056 \pm 1.63 \times 10^{-18}$	0.2135±0.0052
Phosphorous (ppm)	Not detected	5833±223
Potassium (ppm)	3.64±0.0415	3941±131
Calcium (ppm)	13.8±0.150	528±18.6
Sodium (ppm)	Not detected	332±151
Manganese (ppm)	Not detected	72.0±2.67
Iron (ppm)	0.306±0.015	160±42.5
Zinc (ppm)	Not detected	161±7.51

Note. *mean ± standard error

Seedling Vigour Index

ImageJ (Fuji, Japan) was used to analyse the root and shoot length at day ten. Seedling vigour is the total sum of seed properties that determine the seed or seed lot's level of activity and performance during seed germination and seedling emergence (International Seed Testing Association [ISTA], 1995). Low seed vigour means the seeds cannot perform all the physiological functions that allow them to germinate (ISTA, 1995). The seedling vigour index of the 10-day-old seedlings was calculated using the equation: root length + shoot length × germination percentage (%).

Sandwich Compost Leachate Preparation

The sandwich compost preparation method was modified according to Christel (2017) and Phooi et al. (2021) (Figure 1). Effective microorganisms (EM) were used to prepare the sandwich taster. EM contains a larger number of lactic acid bacteria and yeasts and a minor quantity of phototrophic bacteria, filamentous fungi, and actinomycetes in a pH 3.5 liquid culture (Higa, 2001; Higa & Parr, 1994). An initial mixture was made with one part EM and one part of molasses dissolved in 45 parts of water. Next, the sandwich taster was prepared with one part of the mixture mixed with two parts of rice bran. The taster was kept in an opaque garbage bin and covered with a black garbage bag for two weeks before sun-dried. The sandwich compost bucket was self-made using two garbage bins: the upper bin with 26 holes (∅2 mm in size) drilled at the bottom and the lower bin with a tap. The ratio of 3:2 of cropped 2 cm collected raw and cooked plant and animal-based food waste were layered in the bin. The waste mixture was alternately layered and compacted with 1 cm of sandwich taster and 5 cm of food waste. The leachate was harvested on day 14 of fermentation.

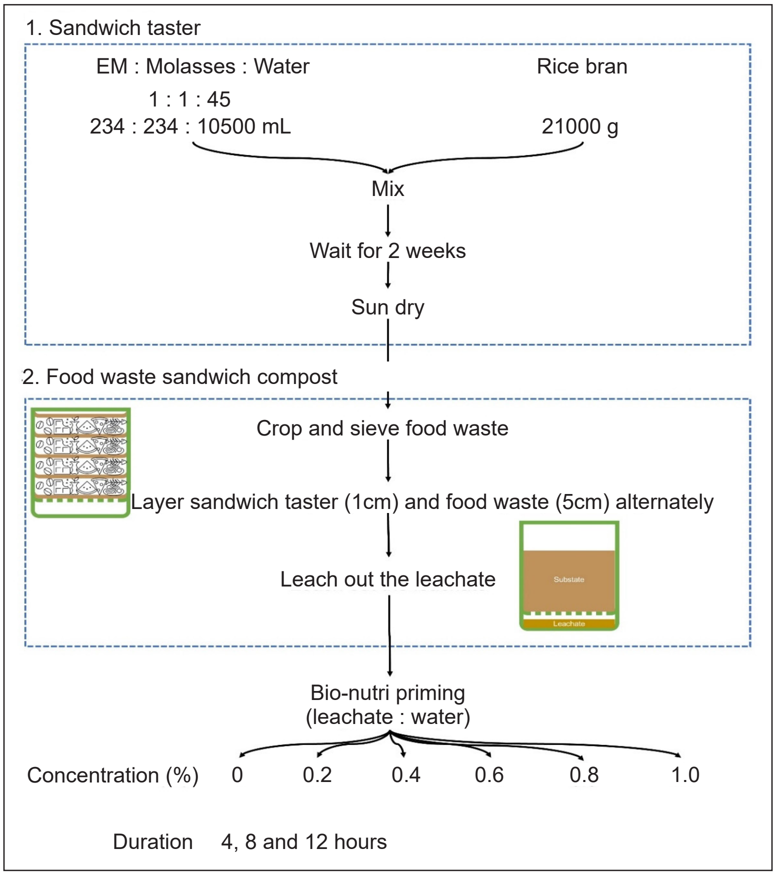

Figure 1. The procedure to prepare food waste sandwich compost leachate

Statistical Analysis

Data were subjected to a two-way analysis of variance (ANOVA) using R software (version 4.1.2). If the *F* values were significant at the $p<0.10$ level, treatment means were compared and separated using the Fisher's least significant difference (LSD).

RESULTS AND DISCUSSION

Seed priming was controlled by various factors such as priming agent concentration and duration (Waqas et al., 2019). Results indicated that the concentration of leachate and bio-nutri-priming duration showed a significant interaction on the standard error of germination rate ($S_{\overline{V}}$) and the corrected germination rate index ($S_{corrected}$) (Table 2).

A study has shown that micronutrient corn seed priming had significant interaction in the variables of germination percentage (GP), germination rate, the coefficient of the velocity of germination, days to germination, and mean germination time (Nciizah et al., 2020). However, in this study, there is no significant interaction between the different treatments on germination percentage (GP), germination rate as the reciprocal of the median time (V_{50}), germination index (GI), and peak value (PV) (Table 2). Therefore, to obtain a better seedling vigour index, a 0.6% leachate concentration with a 12-hour bio-nutri-priming duration is recommended (Table 2).

Some studies state that nutrient toxicity might occur in seed coats with longer priming durations and high leachate concentration levels and is deleterious to the seed germination performance. For instance, corn seed metabolism changes with toxicity, thus, decreasing the utilisation of seed food reserves (Nciizah et al., 2020). Germination percentage was significantly reduced at a high concentration of micronutrient priming (0.5%) for a long duration (24 hours) (Nciizah et al., 2020). A 0.08% ginger rhizome aqueous extractant significantly decreased the germination percentage of chive (*Allium schoenoprasum* L.) (Han et al., 2008). Also, a 100 mM Zn micronutrient priming significantly reduced the seed germination percentage (Ajouri et al., 2004).

Micronutrient priming such as zinc, boron, and manganese significantly shortened the mean germination time in corn (Rasool et al., 2019). In addition, priming improved seed germination compounds production (Varier et al., 2010). For instance,

Table 2
Germination performance based on bio-nutri-priming concentrations and duration

	GP (%)	$S_{\overline{V}}$ (day^{-1})	V_{50} (day^{-1})	$S_{corrected}$ (day^{-1})	GI	PV (% day^{-1})	Seedling vigor index
Concentrations (%)							
0.0	24.97b	0.055a	0.53a	0.44a	1.09b	8.63b	43.43b
0.2	28.03ab	0.066a	0.58a	0.45a	1.26ab	9.23ab	45.96b
0.4	28.75ab	0.055a	0.62a	0.49a	1.28ab	0.98ab	47.06b
0.6	34.86a	0.058a	0.57a	0.44a	1.50a	11.90a	70.34a
0.8	28.32ab	0.065a	0.57a	0.44a	1.16b	9.48ab	48.62ab
1.0	31.77ab	0.055a	0.60a	0.47a	1.39ab	11.12ab	58.07ab
Duration (hours)							
4	28.99a	0.059ab	0.54b	0.43b	1.23a	9.28b	47.63b
8	27.57a	0.054b	0.58ab	0.46ab	1.26a	9.72ab	44.86b
12	31.78a	0.068a	0.62a	0.47a	1.35a	11.67a	64.25a
Significant level							
Concentration (c)	ns	ns	ns	ns	ns	ns	ns
Duration (d)	ns	.	*	ns	ns	*	*
c×d	ns	**	ns	*	ns	ns	.
Mean	29.45	0.060	0.58	0.45	1.28	10.22	52.25
Coefficient of variation (CV)	31.29	31.81	18.72	14.62	28.23	33.57	50.87

Note. GP = Germination percentage; $S_{\overline{V}}$ = Standard error of germination rate; V_{50} = Germination rate as the reciprocal of the median time; $S_{corrected}$ = Corrected germination rate index; GI = Germination index; PV = Peak value. Means with the same letter were not significantly different between treatments (p>0.05) using LSD. *** p<0.00; ** p<0.001; * p<0.01;. p<0.10; ns no significant p<1.00

DNA, RNA, and protein may be triggered to produce during the biopriming of corn (Afzal et al., 2008; Nciizah et al., 2020). Seed priming also enhanced seed protease and α-amylase activity for carbohydrates metabolism and eventually improved assimilation and translocation (Jafar et al., 2012).

Different bio-nutri-priming durations showed a significant positive effect on the germination rate as the reciprocal of the median time (V_{50}), peak value (PV), and seedling vigour index (Table 2). Bio-nutri-priming durations controlled the V_{50}, PV, and seed vigour index (Table 2). The longer the seed bio-nutri-priming duration, the better V_{50}, PV, and seed vigour index (Table 2). A long priming duration was key for enhanced germination despite studies showing that a prolonged nutrient priming duration may result in toxicity during seed germination (Nciizah et al., 2020). Hence, this study has demonstrated that seed bio-nutri-priming for 12 hours significantly improved the Chinese chive seed germination performance. Furthermore, because of long bio-nutri-priming, toxicity was not observed during the Chinese chive seed germination as the germination parameters significantly improved during the 12-hour bio-nutri-priming.

The 12-hour bio-nutri-priming duration had enhanced the germination rate of V_{50}, PV, and seedling vigour index, likely due to the dormant seed needing time to undergo the priming mechanism from imbibition, lag/activation, and germination phase. The 4- and 8-hour bio-priming duration may cut off the phases and be directed to the germination phase (Pawar & Laware, 2018). In the second lag phase, low water intake resulted from slight biomass improvement (Pawar & Laware, 2018). For instance, cabbage showed a high germination rate under 200 mmol L^{-1} urea priming agent (Yan, 2015). Twelve (12) hours of Zn and Mn priming significantly improved the germination rate of marigold up to 93 % (Mirshekari et al., 2012).

The priming duration may vary between 8 hours to 14 days depending on different plant species, osmotic solution, osmotic potential, and temperature (Finch-Savage et al., 1991; Waqas et al., 2019). Nonetheless, an extended priming duration reduced soybean yield; hence, the suitable priming duration was 6 hours for germination performance and yield (Arif et al., 2008).

CONCLUSION

Twelve hours of bio-nutri-priming duration with 0.6 % leachate is recommended for improved germination parameters in Chinese chives. However, this study was only limited to 10 days of seed germination. Thus, future studies can extend to the several harvest cycles to explore the significance between priming duration and leachate concentrations and better understand Chinese chives' nutrient utilisation capacity and accumulation. Furthermore, biotic stress could be applied to the bio nutrient of the sandwich compost leachate primed plant to understand the post-priming memory to later growth.

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