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Research Article

Evaluation of Bokashi Compost Phytotoxicity on Seeds of Some Selected Arable Crops using Heavy Metals and Germination Bioassay Test

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ABSTRACT

Organic amendments such as bokashi can increase crop productivity but may also pollute soils if they have not been sufficiently decomposed. This study assessed the phytotoxicity of bokashi compost (BC) on cabbage, cucumber, okra and sesame seeds using heavy metals and bioassay tests. The experiment consisted of 5 treatments arranged in a Completely Randomised Design (CRD) with 4 replicates. Two sets of data were used. First data assessed the concentration of Copper, Cadmium, Chromium, Lead and Nickel in dried BC. The results revealed that cadmium, chromium and Lead were not detected. The values of copper (5.9 mg kg^{-1}) and Nickel (12.1 mg kg^{-1}) were within the acceptable limits. Analysis of variance was first used on the second data set (BC treatments). The results indicated a significant difference in the average number of sesame seeds germinated, with the highest value (6.75) obtained in 75% BC ($P < 0.05$). Also, okra gave significantly higher average radicle length in all bokashi treatments over the control. The global germination index (GI), which considers the average Gi of 50% and 75% bokashi extract, was used for the bioassay test. The highest GI (217.3%) was obtained on okra, which was > 136.9% (sesame) > 84.1% (cucumber) > 34.1% on cabbage, indicating that the BC was non-phytotoxic on okra, sesame and cucumber. This study demonstrated that the BC was safe with a phytostimulatory effect, thus recommended on wide range of crops. Future studies on BC should investigate phytotoxic substances other than heavy metals.

Keywords: Arable crop seeds; Bokashi compost; Germination bioassay; Heavy metals; Phytotoxicity

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INTRODUCTION

Organic farming is increasingly gaining relevance in recent years all over the world, especially in response to the rising cost of synthetic fertilizers, and the growing interest in agro-ecological practices (Urra *et al*., 2019). Crop producers are therefore switching from conventional to more profitable and eco-friendly organic farming (Reganold & Wachter, 2016). Organic fertilizers can supply crops with the needed nutrient but, they may contain heavy metals and other compounds that are toxic, capable of hindering crop growth and productivity (Gashua *et al.,* 2022; Kebrom *et al*., 2019a).

A compost may be regarded as stable when it resists further decomposition process, and can be considered mature when most of the phytotoxic compounds that are toxic to plants disappear (Bernal *et al*., 2009; Kebrom *et al*., 2019a; Komilis, 2015). The quality of compost is governed by its toxicity, which is usually assessed using seed germination index

(Kebrom *et al*., 2019b; Young *et al*., 2016b). Therefore, Composting is a cheap and safe means of recycling agricultural waste thereby minimizing the levels of phytotoxic compounds (Young *et al*., 2016a). However, the quality of compost needs to be evaluated and seed germination bioassay test is an important tool used to evaluate compost phytotoxicity. The intoxication of living plants by compounds found in the growing medium that are absorbed and deposited in plant tissue is known as phytotoxicity (Araújo & Monteiro, 2005). The most obvious impact of phytotoxic compounds on crop plants is to inhibit seed germination (Visioli *et al*., 2014). Phytotoxicity serves as the crucial parameter for evaluating the suitability of finished compost by means of germination bioassays using different crop seeds (Milon *et al*., 2022).

The concept of seed germination index was first proposed by Zucconi with colleagues (1981) using cress seeds to evaluate compost toxicity. Hence, a germination index (GI) is now frequently used to assess compost's phytotoxicity (Luo *et al*., 2018). The GI is widely accepted because it combines both the relative seed germination (RSG) and the relative root growth (RRG), the two of which are compost quality indicators (Luo *et al*., 2018).

Bokashi composting is a highly versatile procedure that is used to recycle wasted organic material, the finished product of which can be used to produce leachate or tea (Phooi *et al*., 2022). Bokashi composting uses certain group of microorganisms to ferment organic material anaerobically and give a useful product that can be used to enrich soil (Footer, 2013). The bokashi compost made from organic waste has the potentials to increase soil fertility, crop productivity and sustainability and is ecofriendly (Evcİm, 2022). Bokashi tea was reported to give tomato a good start and improved its growth and quality compared to untreated plants (Olle, 2020). The use of bokashi compost leachate on crop has also been reported (Buang, 2019; Lind, 2014). Aqueous extracting solutions have been used to test for seed germination and the extent of root length growth (Aylaj & Adani, 2023; Qasim *et al*., 2018).

Arable crops are those range of crops cultivated all year-round including grains (such as millet), pulses (such as cowpea), oilseed (such as sunflower), pasture crops (such as grasses), fibre crops (such as cotton), and tuber crops (such as yam) (Olohungbebe et al., 2023). Arable crops also refer to those crops that are grown on lands that are suitable for tilling, and may include grains such as rice, wheat and corn; or legumes such as beans and peas; or oilseed crops such as sesame and sunflowers; or vegetables such as tomatoes, onions and peppers.

The number of plant species that may be utilized for phytotoxicity test needs to be increased because different plant species may have significantly different susceptibility to pollutants, especially metals. Additionally, this might help build a database on plant seed phytotoxicity that would track the growing use of these kinds of bioassays. Among the dicot plants that were reported to be very sensitive to phytotoxic compounds are cucumber, radish and red clover, with millet and wheat among the monocot (Visioli *et al*., 2014). Also, while various types of compost and organic manure are being used by smallholder farmers, there still remains a significant gap in knowledge regarding the safety, efficiency, and crop compatibility of such organic amendments, especially the fermented organic extracts like bokashi compost tea. Also, the inappropriate use or application of premature compost can lead to phytotoxic effects such as reduced germination rates, stunted root growth, and delayed crop establishment especially at the early seedling stage, where sensitivity to toxic compounds is highest. Yet, farmers apply compost with the aim of increasing yield without scientific evaluation of its phytotoxic effects on both crop and soil. Additionally, studies on seed germination bioassays using compost are lacking in semi-arid region of Nigeria such as Gashua, where climatic and edaphic conditions may alter the amendment behavior. Without such knowledge, the full potentials of bokashi compost as a safe and cost-effective organic fertilizer may continue to remain untapped. Thus, this study seeks to fill this gap by conducting a laboratory-based seed germination bioassay to assess whether bokashi compost is phytotoxic on cabbage, cucumber, okra and sesame seeds. The objective of this research was therefore to determine the suitability of this bokashi compost on these selected arable crops and to assess the seed with highest global germination index.

MATERIALS AND METHODS

Site of the Experiment

This experiment was conducted in May 2025, in the instrumental laboratory of the Central Laboratory, Federal University Gashua, Nigeria.

Determination of Heavy Metals in the Bokashi Compost

To determine the heavy metals (HM) in the bokashi compost, already prepared bokashi compost (aged 3 weeks) was collected from the Agronomy Department, Federal University Gashua, Nigeria. The dried bokashi sample was taken to Bayero University Kano, Nigeria, for the determination of heavy metals content. The heavy metals were determined by the dry ashing method, as described by Jones (1984).

Seed Germination Bioassay Test

The bokashi compost used for the study was made of dung from cattle, sheep and goat, rice husk, wheat bran, and ash that were treated with yeast and lactic acid bacteria and the whole process lasted for 21 days when its colour and texture changed and there was no odour coming out from the heap. The use of yeast and lactic acid bacteria as effective microorganisms to facilitate bokashi composting with subsequent release of plant nutrients was reported (França *et al*., 2016).

For the seed bioassay test, 50 g of the bokashi compost were measured and dissolved in a 500 mL distill water (1:10 w/v) as described by Kebrom *et al*. (2019), and shaken for 6 hours on a mechanical shaker. This formed the bokashi tea which was sieved using muslin cloth and later centrifuged at 4000 rpm for 1 hour at 20 ^{0}C. Centrifugation is a quick and effective means of removing particles from the extract (Luo *et al*., 2018). The bokashi tea was later diluted with distill water to make 100%, 75%, 50%, 25% and 0% extract of the supernatant solution. The seeds of four different crops, Cabbage (*Brassica oleracea*) from FAO-2024 release, Cucumber (*Cucumis sativus*) obtained from Jubaili Company, Okra (*Abelmoschus esculentus*) NHAR47-4 variety from Nihort, Bagauda, Kano, and Sesame (*Sesamum indicum*) NCRIBEN-04E (Ex-Sudan) variety from National Cereal Research Institute (NCRI), Badeggi, Nigeria were selected for the test. For each crop and for each of the extracting solution, four petri dishes (serving as the replicates) sized 90 mm were used which were lined with Whatman filter paper No 1 or tissue paper as the case may be. To each dish, 3 mL of the appropriate treatment solution was added and the control dish also received 3 mL of distill water. Seven seeds of each crop were sown on each petri dish. The seeds were germinated under dark condition at room temperature (25 ^{0}C) for 72 hours.

Treatments and Experimental Design

This study consisted of five treatments. The treatment formulations were

T1 - 100% distill water = (control)

T2 – 25% bokashi = (75% distill water + 25% bokashi compost extract)

T3 – 50% bokashi = (50% distill water + 50% bokashi compost extract)

T4 – 75% bokashi = (25% distill water + 75% bokashi compost extract)

T5 - 100% bokashi = (100% bokashi compost extract)

These were arranged in a Completely Randomized Design (CRD) and replicated four times (Plate 1).

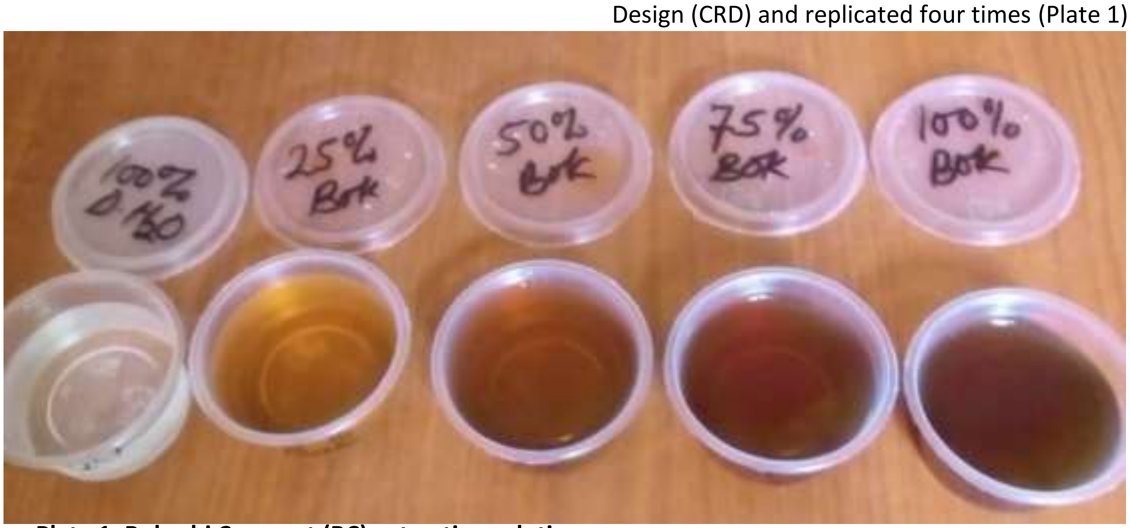

Plate 1. Bokashi Compost (BC) extracting solutions

Data Collection and Analysis

Data were collected on the average number of seeds germinated and average radicle length of the crops. The number of seeds germinated (G) was counted and the radicle length (L) was measured using centimeter rule. A seed is considered germinated when the radicle sprout is ≥1 mm long (Visioli *et al*., 2016). The germination index (Gi) was computed using the formula GI = (G/Go × L/Lo) × 100, where G and L were the total number of seeds germinated and the average radicle length from the bokashi extract, respectively, and Go and Lo are the respective total number of seeds germinated and average radicle length obtained from the control (distilled water). Thus, the global germination index (GI) used was the mean Gi of 50% and Gi of 75% of the bokashi extracting dilutions (Gashua *et al*., 2022; Luo *et al*., 2018).

$$\text{Gi } 50\% = [(\frac{w}{a}) \times (\frac{y}{b})] \times 100 \quad (1)$$

$$\text{Gi } 75\% = [(\frac{x}{a}) \times (\frac{z}{b})] \times 100 \quad (2)$$

$$GI = \frac{(Gi\ 50\% + Gi\ 75\%)}{2} \quad (3)$$

Where the ratios w/a and y/b were the relative seed germination (RSG) and relative radicle growth (RRG), respectively in 50% bokashi compost extract; and x/a and z/b denote the relative seed germination (RSG) and relative radicle growth (RRG), respectively in 75% bokashi compost extract.

RESULTS AND DISCUSSION

Table 1 revealed the heavy metals content in the bokashi compost used for this study. The results showed that most of the heavy metals (Cadmium, Chromium and Lead) were not detected from the sample following the laboratory analysis. However, the amounts of Copper and Nickel were 5.9 mg kg^{-1} and 12.1 mg kg^{-1}, respectively. These values were within the acceptable limit for compost according to Dutch legislation (Sloot *et al*., 2025). Similarly, the concentration of Cu (5.9 mg kg^{-1}) was below the range of Cu values (7.63 – 20.78 mg kg^{-1}) as reported by Gashua *et al*. (2022) using horse waste-based bokashi compost because this bokashi was well composted using yeast and lactic acid bacteria as effective microorganisms that facilitated the composting process (França *et al*., 2016). One of the best and most popular approaches to solid waste management is by composting (Bo, 2017; Mohee & Soobhany, 2014). A well-decomposed compost that is mature and stable may have less amounts or traces of heavy metals whose values may be safe for soil application. In a similar study on compost made by home composters, the findings revealed that all the compost samples were devoid of heavy metals (Bo, 2017). Therefore, the bokashi compost tested was devoid of the toxic heavy metals, thus, safe for agricultural application.

Table 1. Heavy metal contents in the dried bokashi compost

		Concentration (mg kg^{-1})	
S/No	**Heavy metals**	**Bokashi compost**	**Legal threshold amounts based on Dutch legislation for compost (Sloot et al., 2025)**
1	Cu	5.9	90
2	Cd	ND	1
3	Cr	ND	50
4	Pb	ND	100
5	Ni	12.1	20

Cu = Copper, Cd = Cadmium, Cr = Chromium, Pb = Lead, Ni = Nickel, ND =not detected

In this study, the bokashi compost extract was evaluated on seeds of four arable crops. In Table 2, results revealed the treatment effect on the average number of seeds germinated and average radicle length of cucumber, cabbage, okra and sesame after 72 hours. The treatments did not significantly affect the average number of seeds germinated on cucumber, cabbage and okra. However, the treatment effect was significant on the germination of sesame seeds. The average number of seeds germinated was lowest (4.25) in 25% bokashi extract and highest (6.75) in 75% bokashi extract although, this did not vary significantly with the values obtained in other treatments including the control.

Similarly, the treatment effect on the average radicle length for cucumber, cabbage and sesame was not significant. However, the effect of treatments on the average radicle length for okra was significant, with the longest radicle (36.92 cm) obtained on okra seeds treated with 100% bokashi extract. This explained little about the phytotoxicity of the compost on the test crops. Therefore, a seed germination bioassay was used by calculating the GI values to evaluate the phytotoxicity of this bokashi compost on these arable crops. This is because the germination index is considered the best tool to assess the compost phytotoxicity because the results are fast, simple and reliable (Lončarić *et al*., 2024).

Results presented in Table 3, Plates 2a and b, indicated the germination bioassay test for the bokashi compost using cucumber seeds. A total of 27 seeds germinated in the control out of 28 sown with the total of average radicle length of 238.2 cm. However, the total numbers of seeds germinated in 50% and 75% bokashi extract were 24 and 26, respectively. Also, the totals of the average radicle length growth in 50% and 75% were 215.0 and 217.0 cm. A GI of 84.1% was obtained indicating that the bokashi compost used was not phytotoxic on the crop. A GI value above 80% demonstrated the absence of phytotoxicity in the compost (Ravindran *et al*., 2019). In a study to evaluate compost maturity on some selected plant species, a similar GI of 84% on cucumber was reported (Lončarić *et al*., 2024).

Table 2. Treatment effect on the average number of seeds germinated and average radicle length of cucumber, cabbage, okra and sesame after 72 hours

	Average number of seed germinated after 72 hours				Average Radicle Length (cm) after 72 hours			
Treatments	**Cucumber**	**Cabbage**	**Okra**	**Sesame**	**Cucumber**	**Cabbage**	**Okra**	**Sesame**
Control	6.75^{a}	3.50^{a}	2.25^{a}	6.00^{a}	59.55^{a}	5.85^{a}	17.23^{b}	30.18^{a}
25% BC	6.50^{a}	3.00^{a}	3.00^{a}	4.25^{b}	54.60^{a}	5.75^{a}	23.43^{ab}	32.52^{a}
50% BC	6.00^{a}	2.00^{a}	3.00^{a}	6.50^{a}	53.75^{a}	2.53^{a}	32.73^{a}	34.27^{a}
75% BC	6.50^{a}	2.25^{a}	2.00^{a}	6.75^{a}	54.30^{a}	4.00^{a}	35.17^{a}	40.48^{a}
100% BC	6.50^{a}	1.50^{a}	2.25^{a}	6.25^{a}	60.30^{a}	3.50^{a}	36.92^{a}	26.38^{a}
SE±	0.310	0.588	0.725	0.433	3.93	1.458	4.79	5.05

Means followed by the same letter(s) in the same column are not significantly different using Duncan's Multiple Range Test (DMRT) at 5% level of probability. BC = bokashi compost, SE = standard error.

Table 3. Germination bioassay test for the bokashi compost extract using Cucumber seeds

		Number of seeds germinated in the treatments					Mean radicle length in the treatments					
Dish No	No of seeds	SGo	25%	50%	75%	100%	RLo	25%	50%	75%	100%	**GI (%)**
1	7	7	6	6	7	6	66.3	62.8	51.3	54.0	67.5	
2	7	7	6	6	7	6	55.0	57.0	50.8	42.1	48.0	
3	7	6	7	6	5	7	64.5	46.9	61.2	53.4	66.4	84.1
4	7	7	7	6	7	7	52.4	51.7	51.7	67.7	59.3	
Total	28	27 a	26	24 w	26 x	26	238.2 b	218.4	215.0 y	217.2 z	241.2	

SGo= Number of seeds germinated in the control, RLo = Average radicle length growth in the control, a = total number of seeds germinated in the control, b= total of the average radicle length growth in the control, w= total of the number of seeds germinated in 50% bokashi extract, x = total of the number of seeds germinated in 75% bokashi extract, y= total of the average radicle length growth in 50% bokashi extract, z = total of the average radicle length growth in 75% bokashi extract, GI = Global germination index

Plate 2a. Cucumber seeds germinated in 50% BC

Plate 2b. Cucumber seeds germinated in 75% BC

Table 4, Plates 3a and b, revealed the germination bioassay test results for the bokashi compost using cabbage seeds. Only 14 seeds germinated in the control out of 28 sown with the total average radicle length of 23.4 cm. However, the total numbers of seeds germinated in 50% and 75% bokashi extract were 8 and 9, respectively. Also, the totals of the average radicle length growth in 50% and 75% were 10.1 and 16.0 cm. The GI obtained was 34.1%. This low GI value below 50% indicated that the bokashi compost was toxic on the cabbage seeds used. The viability of the cabbage seed used cannot be ascertained because a GI of 90.1% was reported on cabbage (Gashua *et al*., 2022). The bokashi might have contained other toxic compounds such as organic acids, phenols ethylene and ammonia (Lončarić *et al*., 2024) that were not covered by this study and were phytotoxic to cabbage. The source of the cabbage seeds (FAO-2024) could also be responsible for the low GI value.

Table 5, Plates 4a and b, revealed the germination bioassay test for the bokashi compost using okra seeds. A total of 9 seeds germinated in the control out of 28 sown with the total of average radicle length of 68.9 cm. However, the total numbers of seeds germinated in 50% and 75% bokashi extract were 12 and 8, respectively. Also, the totals of the average radicle length growth in 50% and 75% were 130.9 and 140.7 cm, respectively. The GI obtained was 217.3%.

This was the highest GI value obtained compared to the GI of cucumber, cabbage and sesame seeds. A value of GI above 100% not only indicated the absence of phytotoxicity but also the phytostimulative effect of the bokashi compost used (Lončarić *et al*., 2024). That is, despite absence of phytotoxic effect on okra, the compost has stimulated early growth of the crop as evident on Table 2. The okra seeds might have adsorbed the extract and utilized the nutrients in the compost for different metabolic processes, including cell division, root elongation and differentiation (Cho *et al*., 2016).

Table 6, Plates 5a and b present results of the germination bioassay test for the bokashi compost using sesame seeds. A total of 24 seeds germinated in the control out of 28 sown with the total of average radicle length of 120.7 cm. However, the total numbers of seeds germinated in 50% and 75% bokashi extract were 26 and 27, respectively. Also, the totals of the average radicle length growth in 50% and 75% were 137.1 and 161.9, respectively. The GI obtained was 136.9. Although the GI value was below that of okra, but GI above 100% indicated non-phytotoxic effect and phytostimulatory effect of the bokashi compost.

Table 4. Germination bioassay test for the bokashi compost extract using Cabbage seeds

Dish No	No of seeds	Number of seeds germinated in the treatments					Mean radicle length in the treatments					GI (%)
		SGo	25%	50%	75%	100%	RLo	25%	50%	75%	100%	
1	7	3	4	2	2	2	6.0	4.5	5.6	2.0	4.0	
2	7	5	3	0	3	0	5.7	10.7	0.0	4.0	0.0	
3	7	4	2	4	1	2	10.0	1.5	1.0	4.0	3.5	34.1
4	7	2	3	2	3	2	1.7	6.3	3.5	6.0	6.5	
Total	28	14	12	8	9	6	23.4	23.0	10.1	16.0	14.0	
		a		W	x		b		y	Z		

SGo= Number of seeds germinated in the control, RLo = Average radicle length growth in the control, a = total number of seeds germinated in the control, b= total of the average radicle length growth in the control, w= total of the number of seeds germinated in 50% bokashi extract, x = total of the number of seeds germinated in 75% bokashi extract, y= total of the average radicle length growth in 50% bokashi extract, z = total of the average radicle length growth in 75% bokashi extract, GI = Global germination index.

Plat 3a. Cabbage seeds germinated in 50% BC

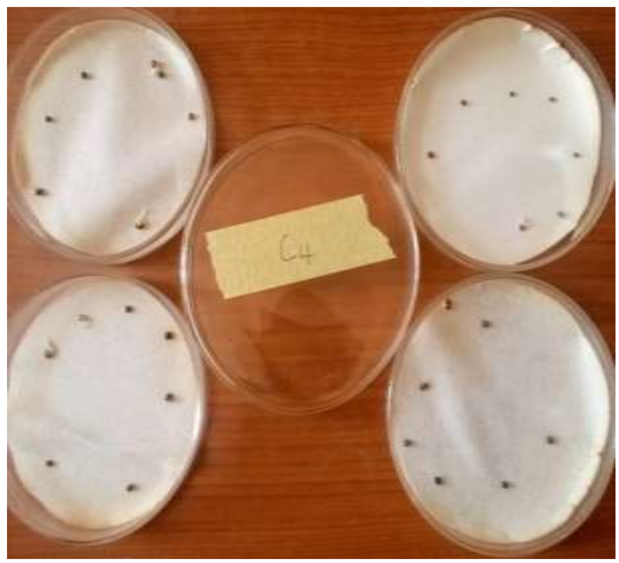

Plate 3b. Cabbage seeds germinated in 75% BC

Table 5. Germination bioassay test for the bokashi compost extract using okra seeds

Dish No	No of seeds	Number of seeds germinated in the treatments					Mean radicle length in the treatments					GI (%)
		SGo	25%	50%	75%	100%	RLo	25%	50%	75%	100%	
1	7	5	2	3	1	1	26.2	14.5	36.7	45.0	40.0	
2	7	0	3	2	2	1	0.0	26.7	22.5	36.5	38.0	
3	7	3	4	4	3	2	26.7	30.0	25.0	36.7	42.5	217.3
4	7	1	3	3	2	5	16.0	22.5	46.7	22.5	27.2	
Total	28	9	12	12	8	9	68.9	93.7	130.9	140.7	147.7	
		a		W	x		b		y	z		

SGo= Number of seeds germinated in the control, RLo = Average radicle length growth in the control, a = total number of seeds germinated in the control, b= total of the average radicle length growth in the control, w= total of the number of seeds germinated in 50% bokashi extract, x = total of the number of seeds germinated in 75% bokashi extract, y= total of the average radicle length growth in 50% bokashi extract, z = total of the average radicle length growth in 75% bokashi extract, GI = Global germination index.

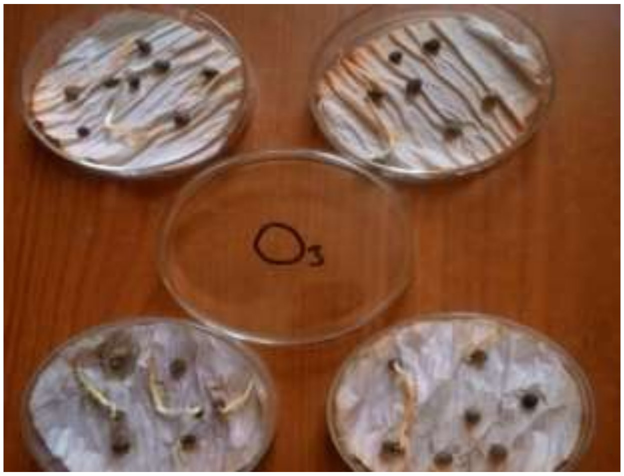

Plate 4a. Okra seeds germinated in 50% BC

Plate 4b. Okra seeds germinated in 75% BC

Table 6. Germination bioassay test for the bokashi compost extract using sesame seeds.

Dish No	No of seeds	Number of seeds germinated in the treatments					Mean radicle length in the treatments					GI (%)
		SGo	25%	50%	75%	100%	RLo	25%	50%	75%	100%	
1	7	7	4	6	7	7	19.0	31.5	28.0	44.4	22.1	
2	7	4	4	6	7	6	28.8	27.0	40.5	29.3	31.7	
3	7	6	5	7	7	5	50.8	46.6	40.7	34.0	21.0	136.9
4	7	7	4	7	6	7	22.1	25.0	27.9	54.2	30.7	
Total	28	24	17	26	27	25	120.7	130.1	137.1	161.9	105.5	
		A		w	x		b		y	z		

SGo= Number of seeds germinated in the control, RLo = Average radicle length growth in the control, a = total number of seeds germinated in the control, b= total of the average radicle length growth in the control, w= total of the number of seeds germinated in 50% bokashi extract, x = total of the number of seeds germinated in 75% bokashi extract, y= total of the average radicle length growth in 50% bokashi extract, z = total of the average radicle length growth in 75% bokashi extract, GI = Global germination index.

Plate 5a. Sesame germinated in 50% BC

Plate 5b. Sesame germinated in 75% BC

CONCLUSION

This study evaluated the phytotoxicity of bokashi compost on cucumber, cabbage, okra and sesame seeds using heavy metals and bioassay assessments. The results from the heavy metals analysis of the bokashi compost indicated that the concentrations of copper and nickel were within the international safety limits, with cadmium, chromium and lead not detected. The seed germination bioassay also indicated that the bokashi compost was not only non-phytotoxic but had phytostimulatory effect on crops such as okra and sesame. The highest germination index value was obtained from okra seeds. The findings from this study suggest that future studies should consider a broader range of crops to include cereals and legumes. Future studies should not only evaluate heavy metals in bokashi compost but, also assess other phytotoxic compounds such as organic acids, phenolic compounds, ethylene and ammonia.

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Conflict of Interest

The authors declare that there is no conflict of interest

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