



Seed Germination and Growth Parameters of *Zea mays* L. as Influenced by Municipal Solid Waste Incineration Fly Ash (MSWIFA) and Sewage Sludge (SS) Amended Soil

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Abstract

Seed germination trials have relevance in measuring soil toxicity. The current study was therefore conducted to investigate the effect of municipal solid waste incineration fly ash (MSWIFA) and sewage sludge (SS) from the Phuket municipal waste incinerator and wastewater treatment plant as fertilizer substitutes on seed germination and growth parameters of sweet corn (*Zea mays* L.). A field experiment was conducted at the National Corn and Sorghum Research Station, Nakhon Ratchasima Province, using a randomized complete block design (RCBD). The treatments conditions were: 1) non-amended soil, 2) 378.125 kg MSWIFA ha⁻¹, 3) 543.75 kg SS ha⁻¹, 4) 756.25 kg MSWIFA ha⁻¹, 5) 1087.5 kg SS ha⁻¹, and 6) 378.125 kg MSWIFA ha⁻¹ + 543.75 kg SS ha⁻¹. The results indicated that application rates of MSWIFA and SS have profound effects on plant establishment and growth characteristics. The highest germination index (22.26), vigor index (146.4), leaf number (5.65), and fresh and dry weight (5.78 and 2.35 g plant⁻¹), respectively) were recorded from seeds treated with SS (1087.5 kg ha⁻¹), while the highest seed germination was found in 378.125 kg MSWIFA ha⁻¹ + 543.75 kg SS ha⁻¹. The result suggested the alternative use of MSWIFA and SS without addition of fertilizer on agricultural soils.

Keywords: Municipal solid waste; Incineration fly ash; Sewage sludge; Sweet corn; *Zea mays* L.

Introduction

Population increase and the rapid growth of tourism in Phuket Province, Southern Thailand

have greatly exacerbated the problem of disposal of environmentally harmful waste products. The fast-increasing volume of municipal waste

is already overwhelming the capacity of Phuket Municipality to incinerate these types of waste [1]. The disposal of municipal solid waste incineration fly ash (MSWIFA) and sewage sludge (SS) from the wastewater treatment plant of Phuket Municipality requires consideration of an integrated waste management strategy. Thus, reuse and recycling of these wastes might be an environmentally friendly and cost-effective approach, offering the benefit of recycling residuals rather than disposing of them. The use of some types of waste as a soil ameliorant for soil quality improvement is of special interest.

MSWIFA consists of hollow spheres, and contains high concentrations of aluminum oxides and hydroxides which might be used to adsorb pollutants such as heavy metals. Moreover, MSWIFA can also be used as a soil additive due to its capacity to supply sufficient amount of plant nutrients (Cu, Mg, S, K, P, Fe, Mn, Zn, etc.) as well as its physical structure that can improve chemical and physical properties of soils due to its high pH. When used as a soil amendment, MSWIFA has favorable effects on soil water holding capacity and aeration [2]. Sewage sludge has been regarded as a problematic solid waste, mainly due to the presence of toxic elements (Cd, Cr, Pb, etc.). Nevertheless, sewage sludge is a good source of essential plant nutrients (Ca, Cu, Mg, K, P, S, Fe, Zn, etc.) for plant growth and can improve soil properties. The characteristics of sewage sludge can vary depending on the individual source and treatment process. In Thailand, more than 47% by weight basis of sewage sludge comprises organic waste [3]. The combination of sewage sludge with MSWIFA may have synergistic effects in improving soil quality by counteracting problems such as waterlogging, contamination with pathogenic microorganisms, loss of soil structure, and a lowering of the availability of metals in amended soils [4].

Soil amendments can be used as fertilizer substitutes, and can improve soil fertility and

promote plant growth. The effectiveness of different amendments in increasing soil fertility is well covered in the literature; however, relatively few studies have been reported on the efficacy of the combination of MSWIFA and sewage sludge from Phuket Municipality as fertilizer substitutes.

Sweet corn (*Zea mays* L.) was chosen for the seed germination studies because it is one of Thailand's most important crops [3, 5]. Germination of seeds is one of the most critical phases of the plant life [6] and is highly vulnerable to environmental factors, particularly soil moisture, soil nutrients and soil temperature [7]. Therefore, this research aims to investigate the effect of MSWIFA and sewage sludge (SS) as fertilizer substitutes on seed germination and growth parameters of sweet corn (*Zea mays* L.) in its early growth stages. Seed germination can be used as a bioindicator for measuring soil toxicity [8]. Such an investigation may therefore help in evaluation of the potential use of MSWIFA and sewage sludge in agricultural fields as a quick and reliable test to assess final crop yield and quality.

Methodology

1) Description of the study area

The field experiment was conducted in October 2015 at National Corn and Sorghum Research Station (Suwan Farm), Kasetsart University, Nakhon Ratchasima Province, North-eastern of Thailand (Lat. 14° 38' 44''N; Long. 101° 18' 59''E). The location of the study area is shown in Figure 1. Average temperatures during the experiment ranged from 22 to 30 °C with the average monthly precipitation of 4.1 mm [9].

2) Preparation and analysis of soil, municipal solid waste incineration fly ash (MSWIFA), sewage sludge (SS), and seeds of sweet corn

Soil samples were collected for characterization at 0 to 15 cm depths prior to commence-

ment of the field trials. Samples were air-dried, ground and sieved through a stainless steel 2-mm sieve, then stored in low-density polyethylene (LDPE) plastic zip-lock bags at room temperature ready for determination of physicochemical characteristics using standard methodologies.

Sweet corn (*Zea mays* L.) single-cross hybrid "Insee 2" obtained from the National Corn and Sorghum Research Station (Suwan Farm) was used in this experiment for plant analysis. The seeds were sieved for size separation before mixing with metalaxyl to control fungal infections. Subsequently, the seeds were air-dried and the healthy ones selected for planting.

Samples of municipal solid waste incineration fly ash (MSWIFA) was collected from the mass-burn incinerator, while sewage sludge (SS)

was collected from the wastewater treatment plant of Phuket Municipality. The samples of MSWIFA and SS were air-dried, ground and sieved through a stainless steel 2-mm sieve and characterized before use as soil amendments. The physicochemical characteristics of the amendments and the soil were determined, including pH (1:1 and 1:2 for soil to water ratio, and amendments to water ratio, respectively) [10], electrical conductivity (EC), organic matter content (%OM) by the Walkley-Black procedure [11], cation exchange capacity (CEC), exchangeable potassium (exc.-K) using 1 N NH_4OAc extraction (pH 7), total nitrogen (total-N) by the Kjeldahl procedure, available phosphorus (avail.-P) by Bray II method, soil texture analysis using the pipette method and metal analysis according to the USEPA method 3052 protocol [12].



Figure 1 Experimental study site

3) Sweet corn field experiment

The sweet corn experiment was laid out in a randomized complete block design (RCBD) with four replicates for a short-term period of 1 month under normal field conditions and agricultural practices. The field experiment was designed to test whether the use of these waste materials as fertilizer substitutes can promote seed germination and growth of the sweet corn. For field preparation, soils were prepared by plowing, sowing and exposing to natural sunlight for 7-15 days prior to application of the amendment. The sub-plots were 6 m long and 3 m wide, with 9 rows with 30 cm intra-row and 75 cm inter-row spacing. Each sub-plot was spaced 1 m apart and 2 m spacing between blocks. Seeds were planted in rows with three seeds per hill; seedlings were thinned to a single plant per hill two weeks after emergence. Treatment details are shown in Table 1.

The study comprised eighteen treatments, with 6 main treatments (including non-amendment as control) and 2 sub-treatments of the applied fertilizer (including non-fertilizer) with 4 replicates per treatment. Therefore, there were 48 sub-plots in the experiment. The commonly used chemical fertilizer (15-15-15) for the initial soil dressing for sweet corn is used at a rate of 93.75 kg ha⁻¹ [13]. Therefore, the specified ratio of the amendment used in this study was based on the mentioned recommended fertilizer rate. The field was prepared and managed by the farmer according to standard corn growing practice. In each treatment, the amendments were

applied at once before planting as banding incorporation throughout the experiment. All MS-WIFA-SS-fertilizer mixtures were then weighed and mixed by hand until homogenous. All sub-plots were watered using sprinkler after seeding and planting, and irrigated once a week using sprinkler. Disease and insect prevalence was also checked regularly.

4) Germination study

Seed germination was recorded daily up to day 9, after the planting and watering. Five seedlings were randomly selected from each sub-plot and the averages of the following parameters were calculated; 1) seed emergence of 2 mm radicle at time of observation was determined by counting the number of seeds emerging. Then, the seed germination percentage [(the number of seeds emerged/the number of seeds sown) x 100] was determined for each of the treatments; 2) fourteen days after seedling emergence, plant height (cm) and plant weight (dry weight and fresh weight, g plant⁻¹) were measured; 3) coefficient of germination were calculated according to Taye et al. (2013) as co-efficient of germination = $(A_1 + A_2 + \dots + A_X) / (A_1 T_1 + A_2 T_2 + \dots + A_X T_X) \times 100$, where A1 is the number of seedlings counted on the first day and T1 is the number of days until the last collection and so on [14]; 4) Germination Index (GI) = \sum [number of seeds emerged on the day/day after planting]; and 5) Vigor Index (VI) was calculated by multiplying seed germination (%) and seedling length (cm) according to Cao et al. (2008) [15].

Table 1 Treatments used in this experiment

Main treatments	Sub-Treatments
M1: Non-amended soil (Ctrl)	F1: No fertilizer
M2: 378.125 kg MSWIFA ha ⁻¹	F2: 93.75 kg fertilizer ha ⁻¹
M3: 543.75 kg SS ha ⁻¹	F3: 187.5 kg fertilizer ha ⁻¹
M4: 756.25 kg MSWIFA ha ⁻¹	
M5: 1087.5 kg SS ha ⁻¹	
M6: 378.125 kg MSWIFA ha ⁻¹ + 543.75 kg SS ha ⁻¹	

5) Statistical analysis

Statistical analysis was based on two-way analysis of variance (ANOVA) for comparison of statistical significance among different treatments using Statistical Product and Service Solutions (SPSS) version 17.0 and Duncan's test at $*p < 0.05$ was used for mean separation.

Results and Discussion

1) Characteristic of soil, municipal solid waste incineration fly ash (MSWIFA), and sewage sludge (SS)

Characteristics of the soil, MSWIFA, and SS used in this study are shown in Table 2. The soil used for in this study had inherently low nutrient content compared to both MSWIFA and SS. The soil was neutral (pH 7.3), predominantly comprising a clay fraction with very little silt and some sand (Table 2). Based on the percentage composition of clay, slit and sand, soils can be classified as clay according to the Soil Survey and Classification Division, Land Development Department [16]. The studied soil had low nitrogen, phosphorus and potassium contents (Table 2). Cd and Pb concentrations were 24.79 and 34.13 mg kg^{-1} , respectively, which fall within the soil quality standard for habitats and agricultural purposes in Thailand (37 mg kg^{-1} and 400 mg kg^{-1} , respectively) [17].

For the amendments, the pH of MSWIFA and SS were 11.5 and 7.1, respectively. Concentrations of Cd and Pb were 77.48 and 794 mg

kg^{-1} and 34.49 and 52 mg kg^{-1} for MSWIFA and SS, respectively. In terms of essential plant nutrients, MSWIFA had the highest exc.-K of 45,063 mg kg^{-1} , while SS exhibited the highest amounts of total-N (3.24%) and avail.-P (1,375 mg kg^{-1}). These high nutrient levels make both materials suitable as fertilizer substitutes. However, the high concentrations of heavy metals, especially in MSWIFA, present the important concern limiting its use for agricultural purposes.

Numerous studies have been conducted on heavy metal accumulation in plants using MSWIFA and SS as soil amendments. It has been reported that heavy metal concentrations in a mixture of fly ash (FA) and SS were substantially below the toxicity limits and the results indicated that use of a mixture of FA and SS poses no risk to the food chain [18]. Phytotoxicity studies of the SS - FA mixture with *Zea mays* L. showed that bioavailability of Zn, Cu, and Co were all decreased by addition of FA [19]. In addition, a reduction of the most bioavailable form of Cd in treated soils (35.4-54.5%) and its transformation into an insoluble fraction was observed when boiler ash was used as a soil amendment; this study highlighted the beneficial effects of waste-products in promoting sugarcane growth and Cd stabilization in soil [20]. Therefore, in order to minimize the associated risk of elevated Cd and Pb concentrations in the MSWIFA used in this study, plant uptake of heavy metals were also investigated.

Table 2 Characterization of the soil, MSWIFA, and SS used in the experiment

Properties	Soil	MSWIFA	SS
pH	7.3	11.5	7.1
OM ^a (%)	1.93	3.40	40.56
EC (dS m ⁻¹)	0.49	77.60	9.38
CEC ^b (cmol _c kg ⁻¹)	18.20	12.00	74.20
Total N ^c (%)	0.11	0.02	3.24
Avail-P ^d (mg kg ⁻¹)	123	ND	1,375
Exc-K ^e (mg kg ⁻¹)	130.44	45,063	1,298
Cd conc. (mg kg ⁻¹)	24.79	77.48	34.49
Pb conc. (mg kg ⁻¹)	34.13	794	52

^aWalkley-Black Method; ^bfAm. acetate 1N pH 7.0 extraction; ^cKjeldahl Method; ^dBrayII; ^eND: not detected

2) Impact of MSWIFA and sewage sludge (SS) on seed germination and emergence

Successful germination and seedling establishment are essential to achieving a good crop of maize [21]. The percentage of seed germination in this study is shown in Table 3. Application of the different amendment rates had no effect on germination ($p > 0.05$). Germination on all treatments exceeded 80%, compared with 86.5% for the untreated control. The highest percentage seed germination (89.7%) was observed in F1M6 treatment (378.125 kg MSWIFA ha^{-1} + 543.75 kg SS ha^{-1}). The results in this study are in line with those reported by Katiyar et al. (2012) who reported the non-significant effect of fly ash on seed germination of mung bean (*Vigna radiate* L.) [22]. The results indicate that F1M4, F1M5, F1M6, and F2M6 treatments had higher levels of germination compared to the untreated control (F1M1), but the differences were not statistically significant at $p > 0.05$. The higher application rate in treatments F1M4 to F1M6 and F2M4 to F2M6 might have resulted in release of N for immediate plant uptake [23].

A similar pattern was observed for germination index (GI), with treatments F1M2-F1M6 and F2M2, F2M4-F2M6 exhibiting higher GI compared to treatments without addition of amendments (F1M1 and F2M1, Table 3). Vigor index (VI) is an important parameter for determining fast and uniform emergence and seedling establishment under a wide range of field conditions [24]. The F1M5 treatment revealed the highest GI value (22.26) and VI (146.4) compared to other treatments. The findings indicated that application of sewage sludge at a rate of 1,087.5 kg ha^{-1} (F1M5) resulted in the most vigorous seedling growth, and can be completely substituted without requiring additional fertilizer.

All treatments showed the same average emergence time of 9 days; thus, there was no observed impact of treatments on time to emer-

gence. The weather at the sampling site during the experiment was relatively cool (21.9 to 30.21 °C) compared to the other growing periods. The time to emergence might therefore be affected as germination is sensitive to soil temperature [25]. The coefficient of germination (%) is one of the indexes measuring seed germination speed and velocity [26] and the germination uniformity or variability in relation to mean germination time [27]. There was no significant difference in the coefficient of germination (%) among treated soils ($p > 0.05$) throughout the experiment (Table 3). The highest value of coefficient of germination (12.88) was observed under F1M6 with the incorporation of 378.125 kg MSWIFA/ha and 543.75 kg SS ha^{-1} . The results indicate that addition of MSWIFA and SS in the soil as fertilizer substitutes have no impact on germination (%) or on the coefficient of germination (%) since all the treatments showed values similar to the F1M1 control. However, treatment F1M6 showed slightly higher levels compared with the other treatments.

3) Impact of MSWIFA and SS on seed growth parameters.

3.1) Number of leaves and shoot height (cm)

After two weeks, five seedlings from each treatment were sampled and measured for leaf number and shoot height (cm). A significantly higher ($p < 0.05$) mean number of leaves per plant was observed in F1M5 (1,087.5 kg SS ha^{-1}) compared to the control, whilst the lowest average leaf number (4.60) was recorded from F1 M4 (756.25 kg MSWIFA ha^{-1}) (Table 3). This indicates that higher application rates of SS, containing significant amounts of total-N (3.24%) and avail.-P (1,375 mg kg^{-1}), can stimulate leaf production without the need for chemical fertilizer. In addition, application of F2M4 (756.25 kg MSWIFA ha^{-1} + fertilizer 93.75 kg ha^{-1}) significantly ($p < 0.05$) increased shoot height (6.75 cm) as compared to the F1M1 control

(5.48 cm). The results indicate that only F2M4 (93.75 kg fertilizer ha⁻¹ + 756.25 kg MSWIFA ha⁻¹) and F1M5 (1,087.5 kg SS ha⁻¹) were statistically different from other treatments in terms of these two seed growth parameters. Similar findings have been previously reported following application of biochar and SS amendments at 15 t ha⁻¹ for SS and biochar, and 7.5 t ha⁻¹ each for SS and biochar [28].

3.2) Shoot fresh weight and dry weight (g plant⁻¹)

Biomass production, expressed in shoots fresh weight and dry weight (g plant⁻¹) of sweet corn grown in different treated soils was mea-

sured on day 14 (T14) (Figure 2). No significant differences in biomass production were observed among treatments ($p>0.05$); however, F1M5 showed the highest shoot fresh weight and dry weight (g plant⁻¹). This result could be due to the higher levels of N and P in this treatment. The study's results are consistent with another study investigating application of municipal solid waste landfill from Khon Kaen Municipality, Thailand on corn yield (*Zea mays* L.) by Prabpai et al. (2009), who found that corn yield increased by 50, 72, 85 and 71% when 20, 40, 60 and 80% of municipal solid waste landfill were used [3]

Table 3 Seed germination (%), germination index (GI), vigor index (VI), coefficient of germination (%), shoot height (cm) and number of leaves of the sweet corn (*Zea mays* L.) as affected by MSWIFA and SS.

Treatments	Seed germination (%)	GI	VI	Coefficient of germination (%)	Shoot height (cm)	No. of leave
F1M1	86.5±12 ^a	19.42±5.5 ^a	104±22 ^a	12.29±0.6 ^a	5.48±0.7 ^{abc}	4.85±0.4 ^{ab}
F1M2	85.7±12 ^a	19.90±6.6 ^a	103±31 ^a	12.85±0.4 ^a	5.23±0.7 ^{ab}	4.65±0.4 ^a
F1M3	85.0±7 ^a	19.45±3.6 ^a	106±4.4 ^a	12.12±0.7 ^a	5.55±0.9 ^{abc}	4.85±0.6 ^{ab}
F1M4	87.4±9 ^a	21.48±6.4 ^a	105±36 ^a	11.98±0.8 ^a	4.85±0.8 ^a	4.60±0.7 ^a
F1M5	89.5±11 ^a	22.26±5.1 ^a	146±46 ^b	12.19±0.8 ^a	6.53±0.8 ^{cd}	5.65±0.1 ^c
F1M6	89.7±10 ^a	21.98±4.9 ^a	121±26 ^{ab}	12.88±0.5 ^a	5.53±0.3 ^{abc}	5.30±0.7 ^{abc}
F2M1	79.9±4 ^a	17.37±5.5 ^a	100±5.7 ^a	12.55±0.5 ^a	5.78±0.5 ^{abc}	5.20±0.3 ^{abc}
F2M2	83.8±7 ^a	21.59±6.6 ^a	130±16 ^{ab}	12.49±0.5 ^a	6.05±0.5 ^{bcd}	5.10±0.4 ^{abc}
F2M3	82.1±6 ^a	17.33±2.9 ^a	106±14 ^{ab}	12.47±0.7 ^a	6.18±0.6 ^{bcd}	5.50±0.1 ^{bc}
F2M4	84.4±11 ^a	19.60±2.7 ^a	133±26 ^{ab}	12.11±0.7 ^a	6.75±0.7 ^d	5.00±0.2 ^{abc}
F2M5	85.0±7 ^a	19.29±1.8 ^a	108±17 ^{ab}	12.10±1.2 ^a	5.59±0.6 ^{abc}	4.90±0.8 ^{abc}
F2M6	88.2±9 ^a	19.84±3.7 ^a	126±19 ^{ab}	11.87±1.0 ^a	6.25±0.8 ^{bcd}	5.25±0.2 ^{abc}

Means followed by different letters are significantly different at 5% level of probability

Where: F1M1 = no fertilizer and no amendment (Ctrl); F1M2 = no fertilizer + 378.125 kg MSWIFA ha⁻¹; F1M3 = no fertilizer + 543.75 kg SS ha⁻¹; F1M4 = no fertilizer + 756.25 kg MSWIFA ha⁻¹; F1M5 = no fertilizer + 1087.5 kg SS ha⁻¹; F1M6 = no fertilizer + 378.125 kg MSWIFA ha⁻¹ + 543.75 kg SS ha⁻¹; F2M1 = 93.75 kg fertilizer ha⁻¹ + no amendment; F2M2 = 93.75 kg fertilizer ha⁻¹ + 378.125 kg MSWIFA ha⁻¹; F2M3 = 93.75 kg fertilizer ha⁻¹ + 543.75 kg SS ha⁻¹; F2M4 = 93.75 kg fertilizer ha⁻¹ + 756.25 kg MSWIFA ha⁻¹; F2M5 = 93.75 kg fertilizer ha⁻¹ + 1087.5 kg SS ha⁻¹; F2M6 = 93.75 kg fertilizer ha⁻¹ + 378.125 kg MSWIFA ha⁻¹ + 543.75 kg SS ha⁻¹; F3M1 = 187.5 kg fertilizer ha⁻¹ + no amendment; F3M2 = 187.5 kg fertilizer ha⁻¹ + 378.125 kg MSWIFA ha⁻¹; F3M3 = 187.5 kg fertilizer ha⁻¹ + 543.75 kg SS ha⁻¹; F3M4 = 187.5 kg fertilizer ha⁻¹ + 756.25 kg MSWIFA ha⁻¹; F3M5 = 187.5 kg fertilizer ha⁻¹ + 1087.5 kg SS ha⁻¹; F3M6 = 187.5 kg fertilizer ha⁻¹ + 378.125 kg MSWIFA ha⁻¹ + 543.75 kg SS ha⁻¹

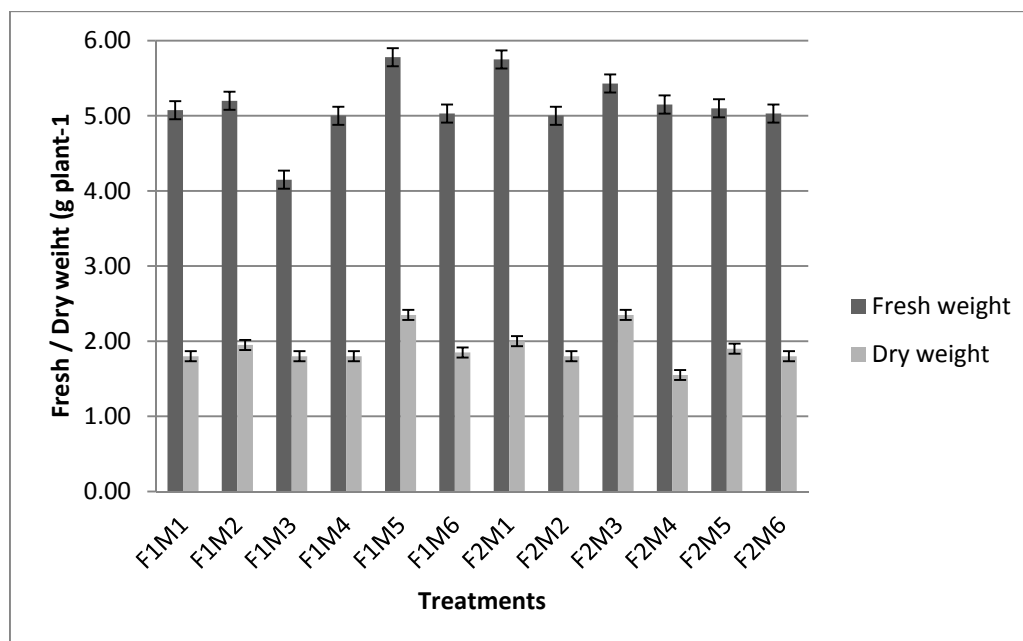


Figure 2 Shoot fresh weight and dry weight (g plant⁻¹) of the sweet corn grown in different treated soils at 14 days.

Conclusion

Different application rates of municipal solid waste incineration fly ash (MSWIFA) and sewage sludge (SS) had profound effects on plant establishment and growth characteristics of sweet corn relative to the conventional NPK fertilizer (15-15-15) application. The highest germination index (22.26), vigor index (146.4), average leaf numbers (5.65), and fresh and dry weight (5.78 and 2.35 g plant⁻¹, respectively) were recorded from maize seeds treated with SS (F1M5, 1087.5 kg ha⁻¹), while the highest percentage seed germination and coefficient of germination were found with the F1M6 treatment (378.125 kg MSWIFA ha⁻¹ + 543.75 kg SS ha⁻¹). Both MSWIFA and SS treatments were found to promote germination and growth parameters as compared to the control. These results support consideration of these materials as potential soil amendments. Nevertheless, the long term effect of repeated application of these materials and heavy metal accumulation in plants still requires further investigation.

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