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Phytotoxicity Evaluation of MSWC by Germination of Gram Seeds *(Cicer arietinum)* and Fenugreek Seeds (*Trigonella foenum-graecum)*

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Abstract **– The compost obtained from composting organic fraction of Municipal solid waste using different initial mix ratios (70%, 80% and 90%) was used to evaluate phytotoxicity of gram seeds (Cicer arietinum) and fenugreek seeds (Trigonella foenum-graecum) using a seed germination method. The results showed that composting generally reduced the phytotoxicity of the mixtures. A germination index was the highest in the mix ratio of 1:9 after 72hrs in the gram seeds (Cicer arietinum) obtained from the compost of municipal solid waste. The germination percentage, germination index and vigour index values were relatively higher in the compost. The vigour index was found to be maximal in the mix ratio of 1:9 after 72hrs in the fenugreek seeds (Trigonella foenum-graecum) from the municipal solid waste compost. The effects of compost water extracts on seed germination were measured to calculate the germination index (GI).**

Keywords **– Cicer Arietinum, Compost, Germination Index, Phytotoxicity, Trigonella Foenum-Graecum.**

I. INTRODUCTION

Phytotoxicity is one of the most important criteria for evaluating the suitability of compost for agricultural purposes and to avoid environmental risks before these composts can be recycled back to agricultural land [1][2][3]. These effects occur because immature compost induces high microbial activity (which reduce oxygen concentration in the soil), blocks the existing soil available nitrogen [4]. Immature compost also introduces phytotoxic compounds such as heavy metals [5]. Phenolic compounds [6], ethylene and ammonia [5], excess accumulation of salts [5], and organic acids [7] which could retard seed germination and plant growth. Acetic acid is probably the most damaging organic acid released from immature compost, but there are also other compounds that contribute to the phytotoxic effect [8]. In fact, application on soil of no stabilized organic materials could affect both crops and the environment because of the presence of phytotoxic compounds [9]. High concentrations of salt and the release of organic acids into the composts are also correlated to inhibition of germination and growth. Phytotoxicity is often best evaluated by conducting germination or growth tests [1][10], but the test plants have to be chosen with care [11]. Germination Index (GI) is the best way to test the phytotoxicity of compost to plant growth because the results of it are quite straightforward and reliable. Germination bioassays are widely used to test for salinity, soil pathogens, toxic substances and some other physical and chemical properties of compost [12][13], which could be the major potential reasons of phytotoxicity. Several investigators reported that phytotoxic compounds are gradually eliminated during the composting process, which could explain the GI increases with composting time. The Germination index (GI), which combines measures of relative seed germination (G%) and relative root elongation (L%), has been used to evaluate the toxicity of compost. [5][14]. It has been noted that a GI value of 80% indicated the disappearance of phytotoxins in composts [13]. [3] used this value not only as indication of the disappearance of phytotoxicity but also as an indication of the maturity of compost. The germination index is a maturity test based on seed germination and initial plant growth using a liquid extract from the compost [13]. It reflects the phytotoxicity of the compost extracts at different stages of composting. The compost is considered mature when the germination index is higher than 60 %, compared to the control with distilled water [15]. The most popular germination test used by researchers is from cress test [13]. In their opinion, the compost is non-toxic when the germination rate is more than 85% or the plant seedling weights are more than 90%. Beside, composting scientists found that GI at each composting time did not show any significant changes with the dilutions of the extract, or even when the extract was diluted up to 75% with distilled water. They conducted that ammonia and low molecular weight organic acids were two phytotoxic substances proposed and plant growth increased as these disappeared. [16] reported that when the GI is more than 80 % Compost is considered mature and practically free of phytotoxic substances. An increased GI is indicative of decreased phytotoxicity and thus of a more mature product [3][17][18].

II. METHODS AND MATERIAL

Study area - Jamshedpur is a city located in the popular state of Jharkhand in India, founded by the late Jamshedji Nusserwanji Tata. The city of Jamshedpur, known as the Steel City, is part of the East (Purba) Singhbhum District of the newly formed state of Jharkhand. Jamshedpur is the headquarters of the East Singhbhum District which was formed by isolating 9 blocks from greater Singhbhum on 16th January 1990. [19]

Jamshedpur was the first planned city in India, nurtured by Tata Steel. Jamshedpur is also the first ISO 9005 certified city in INDIA Jamshedpur has one of the highest per capita incomes in the country and is the only town whose municipality is governed by a corporate house. [19]
Phytotoxicity evaluation of MSWC- The phytotoxicity of compost extracts was evaluated by the seed germination

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technique [4][5][20]. Gram seeds (*Cicer arietinum*) and fenugreek seeds (*Trigonella foenum-graecum*) were surface sterilized by immersion in 75% alcohol for three minutes followed by transferring in 0.001 HgCl2 solution for two minutes with periodical agitation and finally thoroughly washed with sterilized distilled water to get rid of toxic chemicals[21]. 10 ml of water compost extract was applied to filter paper in a Petri dish and 5 seeds were then placed on the filter paper. All experiments were run in replicate.

$$\text{Seed germination } \% = \frac{\text{No. of seed grminated in compost extract}}{\text{No. of seed germinated in control}}$$

$$\textit{Root elongation } \% = \frac{\text{Mean root length in compost extract}}{\text{Mean root lenth in control}}$$

$$\text{Germination index} = \frac{\text{Seed germination } \% \times \text{root elongation } \%}{100}$$

III. Observation

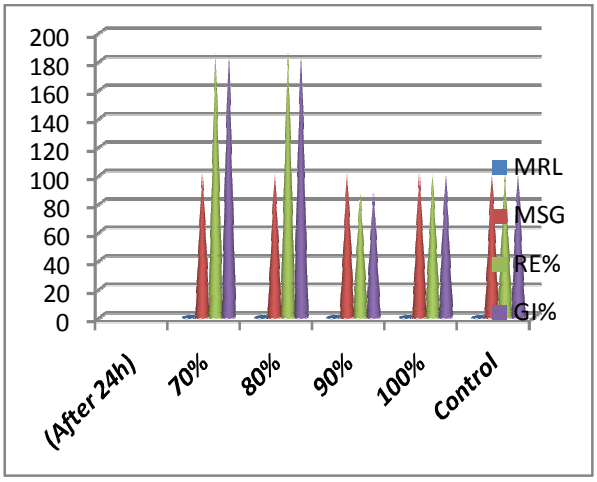
The phytotoxicity of compost extracts was evaluated by the seed germination technique. Water extract was obtained by shaking 10g of dried compost with 100 ml of distilled water for 15h at room temperature in the dark. The flask was then centrifuged for 20min and the supernatant was filtered through whatman No. 42 filter paper. The pH and electrical conductivity (EC) of these extract was examined.

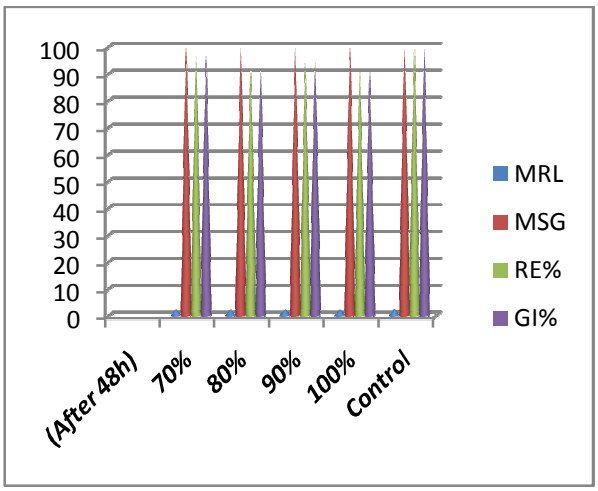
10ml of compost extract was applied to filter paper in a Petri dish and 5 seeds were then placed on the filter paper. All experiments were run in replicate. The Petri dishes were sealed with tape to minimize water loss while allowing air penetration and then were incubated in the dark for 72 hours at room temperature, the seed germination percentage and root length of the plants in the extracts were determined. The seed germination in distilled water was used as control. The percentage of seed germination, root elongation and germination index (GI) was calculated.

The data of seed germination after 72 hours for the 2 replicates containing 5 seeds in each compost mix are shown in the (Table 1 and 2). The GI values in compost mixtures of initial percentage of compost extract were 70%, 80% and 90%. The seed germination percentages were the highest in reactor 80% for the green gram and 70% for the fenugreek seeds. The relatively low germination value in 90% compost extract mixture for green gram and 80% for fenugreek seeds.

	MRL	*MSG*	*RE%*	*GI%*
(After 24h)				
70%	*0.13*	*100*	*185.71*	*185.71*
80%	*0.13*	*100*	*185.71*	*185.71*
90%	*0.08*	*100*	*87.5*	*87.5*
100%	*0.07*	*100*	*100*	*100*
Control	*0.07*	*100*	*100*	*100*
(After 48h)				
70%	*2.25*	*100*	*96.99*	*96.99*
80%	*2.14*	*100*	*92.25*	*92.25*
90%	*2.19*	*100*	*94.39*	*94.39*
100%	*2.11*	*100*	*90.95*	*90.95*
Control	*2.32*	*100*	*100*	*100*
(After 72h)				
70%	*3.73*	*100*	*98.07*	*98.07*
80%	*3.83*	*100*	*82.63*	*82.63*
90%	*3.71*	*100*	*100.39*	*100.39*
100%	*3.82*	*100*	*79.15*	*79.15*
control	*3.81*	*100*	*100*	*100*

MRL- mean root length,
MSG- mean seed germination
RE- root elongation
GI- germination index





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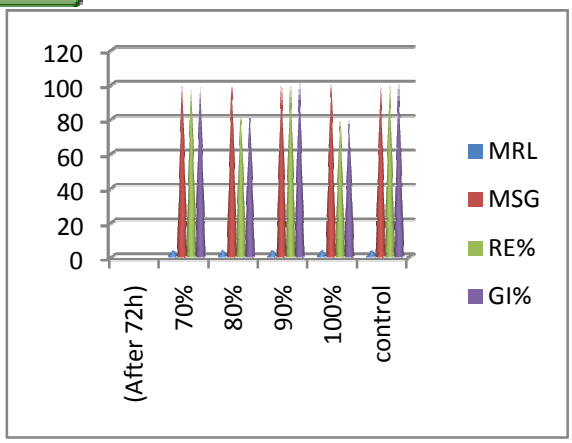
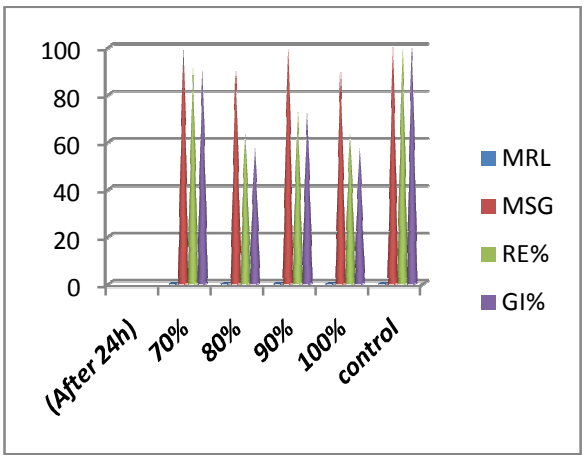


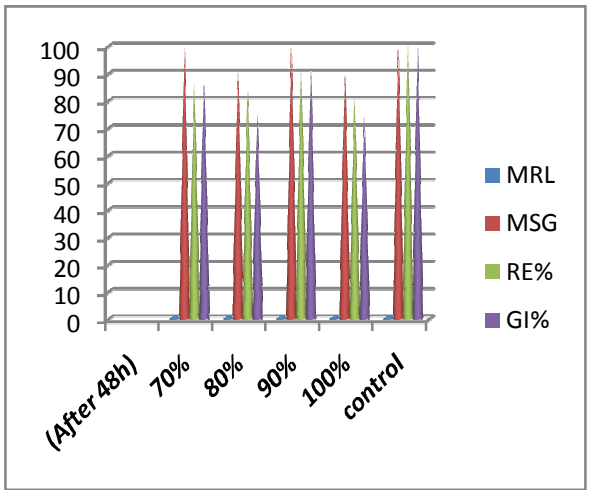
Fig 1. showing the growth of gram seeds at different time intervals

Table 2. Seed germination table for Fenugreek seeds (Trigonella foenum-graecum)

	MRL	***MSG***	***RE%***	***GI%***
(After 24h)				
70%	*0.1*	*100*	*90.91*	*90.91*
80%	*0.07*	*90*	*63.64*	*57.28*
90%	*0.08*	*100*	*72.73*	*72.73*
100%	*0.07*	*90*	*63.64*	*57.28*
control	*0.11*	*100*	*100*	*100*
(After 48h)				
70%	*1.28*	*100*	*86.49*	*86.49*
80%	*1.24*	*90*	*83.79*	*75.42*
90%	*1.35*	*100*	*91.22*	*91.22*
100%	*1.21*	*90*	*81.76*	*73.59*
control	*1.48*	*100*	*100*	*100*
(After 72h)				
70%	*2.54*	*100*	*98.07*	*98.07*
80%	*2.14*	*90*	*82.63*	*74.37*
90%	*2.60*	*100*	*100.39*	*100.39*
100%	*2.05*	*90*	*79.15*	*71.24*
control	*2.59*	*100*	*100*	*100*

MRL- mean root length,
MSG- mean seed germination
RE- root elongation
GI- germination index





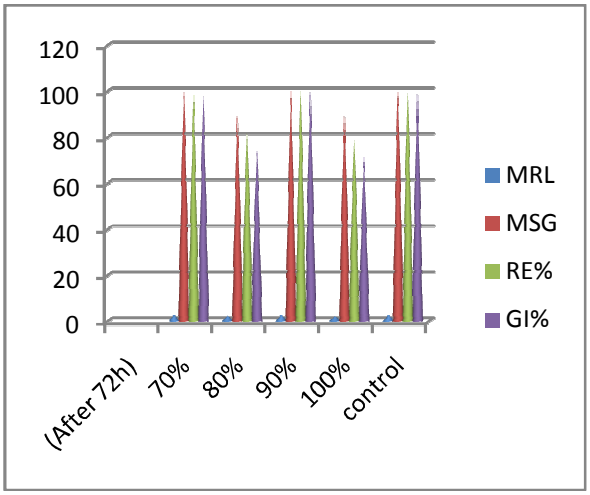


Fig 2. showing the growth of fenugreek seeds at different time intervals

The GI values of gram seeds in composting mixtures of initial mix ratios of 70%, 80,%,90% and 100% were 98.07, 82.63, 100.39 and 79.15 on day 3 respectively (Table 1), Similarly the GI values obtained from fenugreek seeds in composting mixtures of initial mix ratios of 70%, 80,%,90% and 100% were 98.07, 82.63, and 79.15 respectively (Table 2).

The responses of gram seeds *(Cicer arietinum)* and fenugreek seeds *(Trigonella foenumgraecum)* to the toxicity of the compost water extracts during the phytotoxic evaluation in term of the relative seed germination and relative root elongation percentages were illustrated in (Figure 1 and 2). The percentage of seed germination, root elongation and germination index (GI) was calculated. GI of gram seeds and fenugreek seeds was very low as compared to the control using distilled water.

The GIs in the composting mixtures of initial mix ratios 70%, 80%, 90% were over 80% during the composting. The GI value of gram was maximum (100.39) in reactor 90% and for fenugreek seed the GI value was maximum (98.07) in reactor 70%.

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IV. Results and Discussion

The percentage of seed germination, root elongation and germination index (GI) was calculated according to [4]. The responses of gram seeds *(Cicer arietinum)* and fenugreek seeds *(Trigonella foenumgraecum)* to the toxicity of the compost water extracts during the phytotoxic evaluation in term of the relative seed germination and relative root elongation percentages were illustrated in Figure 1 and 2. The pH and electrical conductivity (EC) of these extract were determined [13][20].

The phytotoxicity of compost extracts was evaluated by the seed germination technique [4][5][20]. Gram seeds *(Cicer arietinum)* and fenugreek seeds *(Trigonella foenum-graecum)* were surface sterilized by immersion in 75% alcohol for three minutes followed by transferring in 0.001 HgCl2 solution for two minutes with periodical agitation and finally thoroughly washed with sterilized distilled water to get rid of toxic chemicals [21].

The elimination of phytotoxicity has also been widely used as a measure of compost maturity [6]. GI of gram seeds and fenugreek seeds was very low as compared to the control using distilled water, probably due to the phytotoxic effects of ammonia and low molecular weight of organic acids [16].

The GI values of gram seeds in composting mixtures of initial mix ratios of 70%, 80,%,90% and 100% were 98.07, 82.63, 100.39 and 79.15 on day 3respectively (Table 1). Similarly the GI values obtained from fenugreek seeds in composting mixtures of initial mix ratios of 70%, 80,%,90% and 100% were 98.07, 82.63, and 79.15 respectively (Table 2). [4] reported that a GI value of more than 80% is an indication of phytotoxic-free and mature compost. Similar suggestions were also reported by [5]. The GIs in the composting mixtures of initial mix ratios 70%, 80%, 90% were over 80% during the composting. The GI value of gram was maximum (100.39) in reactor 90% and for fenugreek seed the GI value was maximum (98.07) in reactor 70%.

This result is an agreement with that obtained by [17], which reported that moisture was the most important factor inhibiting seed germination and root elongation of all species examined.

V. Conclusion

The physicochemical and phytotoxicity changes during the reactor composting of MSW, have been investigated at initial mix ratios of 70%, 80%, 90% and 100%. The final characteristics indicate the stability and usability of the compost in the entire ratio. The final GI in composting mixture of reactors the percentage 70, 80 and 90 was over 80%; however, the GI in the composting mixtures of mix ratio 100% for gram and fenugreek was not over 80% during the test.

The results of the current work indicate the necessity to stabilize the compost before its application to agricultural soil in order to avoid environmental problems and phytotoxicity.

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