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Original Research Article

Effect of Bio Priming Seeds with Microbial Based Bio Fertilizers on Growth of Maize Seedlings

Moses Mutetwa[1], Karen Nyaera[2], Thomas Masaka[3], Tuarira A. Mtaita[4]

[1]Postgraduate Africa University Alumni, [2]Undergraduate Student, [3]Assistant Lab Technician Africa University, [4]Senior Lecturer and Head Department of Agriculture and Natural Sciences,
Africa University, Department of Agriculture & Natural Sciences, College of Health, Agriculture and Natural Sciences, Box 1320, Mutare - Zimbabwe

Corresponding Author: Moses Mutetwa; mosleymute@gmail.com

ABSTRACT

The world over agriculture is being threatened by adverse effects of climate change. For that reason, Sub-Saharan Africa needs to intensify agricultural production to feed her growing population adequately in a sustainable manner by the exploitation of beneficial microbes as a bio fertilizer for food security, food safety and sustainable crop production. The study aimed to assess the effects of bio priming with *Trichoderma* and *Bacillus* based-bio fertilizers on maize plant characteristics. A pot experiment was conducted for each bio fertilizer. The soaking treatments for the bio fertilizers were applied to the seeds as follows: Trt_1 *= 0 hours,* Trt_2 *= 8 hours,* Trt_3 *= 16 hours,* Trt_4 *= 24 hours, and* Trt_5 *= Control.* The experiment was laid out in a randomized complete block design with three replicates. The effect of soaking was significant ($P<0.05$) for the parameters (plant height, root length, fresh weight, dry weight) investigated. Number of leaves was only significant ($P<0.05$) for treatments wit *Bacillus*. Best results were obtained when soaking for 16hrs for both *Trichoderma* and *Bacillus* treatments. In view of these findings, the study conclude that when targeting for maximum plant height, fresh weight and dry weight in maize, soaking the seed for 16hrs can be adopted and further, soaking for 24 hrs do promote root length which can help assist in accessing water and nutrients from deeper horizons which can be beneficial in marginal production areas. Significant N and P uptake by the maize was as a result of incorporating the bio fertilizers.

Key Words: *Bio fertilizer, bio priming, Trichoderma, Bacillus, seed, maize, growth*

INTRODUCTION

The world population is estimated to reach about 9 billion by 2050. Africa, especially sub-Saharan Africa, has been predicted to contribute to bulk of the increase (Godfray et al, 2010; UNDESA, 2005). This global population increase coupled with climate change poses a challenge to worldwide agriculture production and has triggered competition in all forms of resources required for human survival such as land, water, energy and food. Perhaps, the most essential is the food resources that have become insufficient, and consequently, its increased production cannot be compromised (Asenso-Okyere & Jemaneh, 2012). Sub-Saharan Africa needs to intensify agricultural production to feed her growing population adequately in a sustainable manner and to find solutions to

combat abiotic and biotic stresses which are being threatened by adverse effects of climate change. For this reasons, the agriculture sector must increase productivity by an estimated 60% to meet an increasing global demand in food (Alex & Bru, 2012).

Dependence on inorganic chemical-based fertilizers pause an obstinate threat to human health and environment. Consequently, adoption of new technology such as an integrated nutrient management that harnesses the benefits of the interaction of plant and microorganisms in the rhizosphere is vital (Denning et al, 2009; Muzari et al, 2012). The exploitation of beneficial microbes as a bio fertilizer in agriculture sector plays a potential role in food safety and sustainable crop production. The need to focus on bio fertilizers' potential as an efficient integrated nutrient management in increasing smallholder farmer productivity and profitability cannot be over emphasized. Increasing bio fertilizer awareness and use is important for maintaining and improving ecological stability and alleviating poverty, especially among the rural dwellers.

In light of this background, this study was importantly undertaken to standardize the optimum duration for seed bio priming using *Trichoderma* and *Bacillus* as an efficient strategies by stakeholders to improve adoption of this technology among smallholder farmers to contribute to food security and sustainable crop production.

RESEARCH METHODOLOGY

Site Description

The experiment was conducted at Africa University Farm located at 18°53'70.3" South and 32°36'27.9" East and at an altitude of 1131m. The mean annual precipitation is approximately 800-1000 mm with most of rain falling between December and February. The average summer temperature is 27ºC and winter temperature is about 7ºC. The medium used in the pot experiment was a sandy loam soil. The soil at AU farm is a red sandy clay loam, Fersiallitic 5E soil under Zimbabwe soil classification system (Nyamapfene, 1991).

Experimental Design, Treatments And Establishment

A pot experimental study was conducted to determine the effect of a bio fertilizer on growth and development of maize seedlings. The experimental treatments were laid out in a Randomized Complete Block Design (RCBD) with three replications. Maize grains were surface sterilized by immersing in 70% ethanol for 2 min and then in 0.2% sodium hypochlorite (NaoCl) for 3 min. Seeds were washed several times with sterile distilled water. The microbial bio fertilizers selected for this study contained *Trichoderma harziunum* at a concentration of not less than $1.0x10^8$ cfu/g and the other *Bacillus oryzicola* YC7007 at a concentration of not less than $1.0x10^{10}$ cfu/g.

The soaking treatments for the bio fertilizers were applied to the seeds as follows: Trt_1 *= 0 hours,* Trt_2 *= 8 hours,* Trt_3 *= 16 hours,* Trt_4 *= 24 hours, and* Trt_5 *= Control (without bio fertilizer)*

Ten seeds were placed into a plastic pot, each filled with about 5 kg of sandy loam based garden soil and replicated three times in a randomized block design. The seeds were sown at 2 to 3 cm depth in each pot and when seedlings growth was 10 old days, the seedling density was reduced to 6 seedlings per pot. The pots were watered every 72 hours with equal amount of water. When the seedlings had reached 45 days old, 4 plants were carefully uprooted from the soil of each treatment.

Data collection;

Samples were collected for the following analyses: Plant height, number of leaves, fresh weight and dry matter of shoots and roots, and root length.

Statistical Analyses of Experimental Data

Data collected was statistically analyzed using the GenStat Analysis of Variance (ANOVA) software. Differences

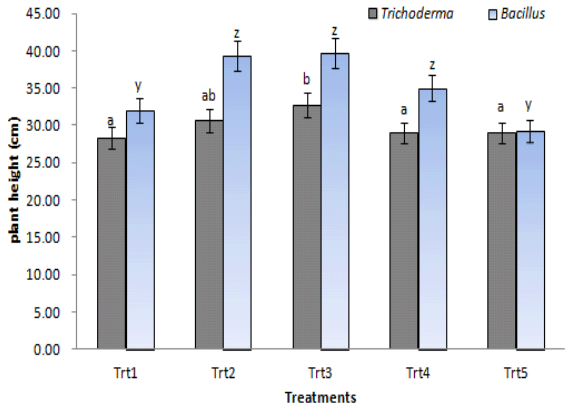
between means were determined using the Least Significant Difference (LSD) test at P=0.05 level.

RESULTS

Effect of Soaking Seed With Bio-Fertilizer

Plant Height

Data regarding the effect of soaking seeds with a *Trichoderma* based-bio fertilizer on plant height is shown in Figure 1.



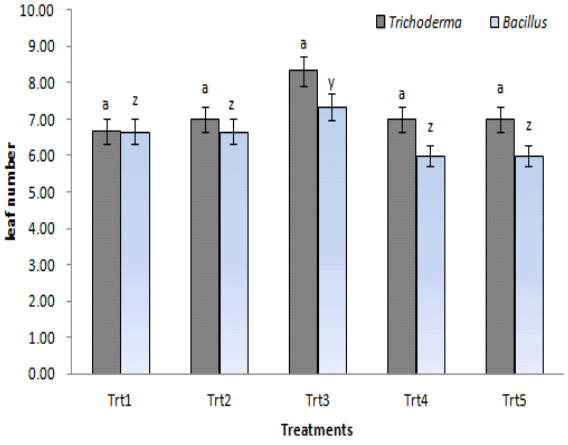
Soaking treatments: Trt1 = 0 hours Trt2 = 8 hours Trt3 = 16 hours Trt4 = 24 hours Trt5 = Control
**Figures not sharing a common letter in a column differ significantly at 0.05 probability.*
Figure 1 Effect of Soaking Seeds with bio fertilizer on plant height

Soaking for 0hrs and 8hrs (Trt_1 and Trt_2 respectively) was not significant differently from the Control (Trt_5) treatment. However, the tallest plants were observed from Trt_3 (32.67cm) and these were not statistically different from Trt_2 with a plant height of 30.67cm. The mean plant height recorded was 29.93cm.

Effect on plant height from soaking seeds with *Bacillus* based-bio fertilizers shows that 0hrs (Trt_1) of soaking and Control (Trt_5) treatments produced the lowest plant height and were not statistically (P>0.05) different (Figure 1). Trt_2, Trt_3 and Trt_4 produced the tallest plants which were not significantly (P>0.05) different from each other. The mean plant height was 35.07cm.

Leaf Number

Effect of soaking with *Bacillus* based-bio fertilizer on number of leaves was significant (P<0.05) (Figure 2).

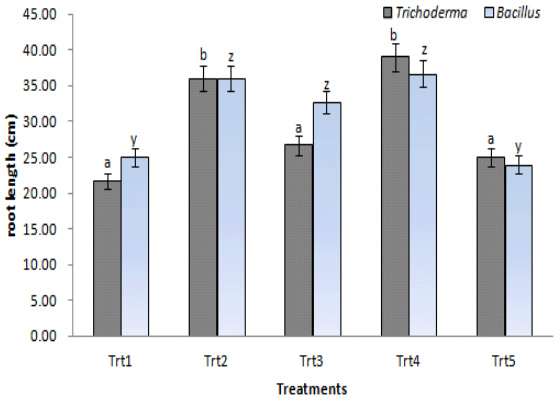


Soaking treatments: Trt1 = 0 hours Trt2 = 8 hours Trt3 = 16 hours Trt4 = 24 hours Trt5 = Control
**Figures not sharing a common letter in a column differ significantly at 0.05 probability.*
Figure 2 Effect of Soaking Seeds with bio fertilizer on number of leaves

Trt_3 produced the highest number of leaves while the rest of the other treatments (Trt_1, Trt_2 and Trt_4) were not statistically different from the Control treatment. The mean number of leaves recorded was 6.53.

Results from effect of *Trichoderma* based-bio fertilizer on number of leaves were not statistically (P>0.05) different between all the treatments.

Root Length



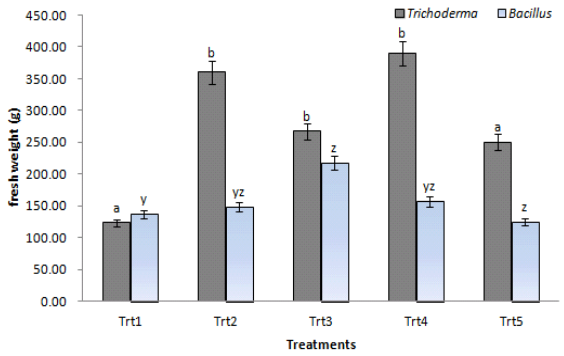
Soaking treatments: Trt1 = 0 hours Trt2 = 8 hours Trt3 = 16 hours Trt4 = 24 hours Trt5 = Control
**Figures not sharing a common letter in a column differ significantly at 0.05 probability.*
Figure 3 Effect of Soaking Seeds with bio fertilizer on root length

Figure 3 shows that Trt_1, Trt_3 and Trt_5 with root lengths of 21.7cm, 26.7cm and 25.0cm respectively were not statistically (P>0.05) different from each other for *Trichoderma* based-bio fertilizer. The longest root length was observed from Trt_2 and Trt_4, 36.0 and 39.0cm respectively. The mean root length recorded was 29.7cm.

Results for the *Bacillus* based-bio fertilizer on root length was significant (P<0.05) while results for Trt_1 and Trt_5 were not significantly (P>0.05) different from each other. However, Trt_2, Trt_3 and Trt_4 produced the largest root lengths; 36.0cm, 32.7cm and 36.7cm respectively, but were not statistically significant (P>0.05) different from each other. The mean root length recorded was 30.9cm.

Fresh Weight

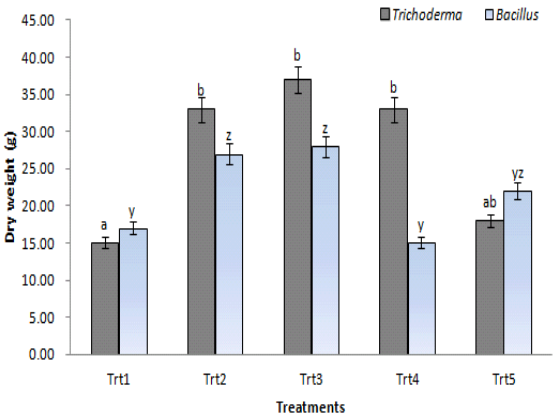
Results regarding fresh weight of the plant from different treatments are shown on Figure 4. The fresh weight from *Trichoderma* based-bio fertilizer for Trt_1 and Trt_5 recorded was the lowest and these were not significantly (P>0.05) different from each other. Fresh weight for Trt_2, Trt_3 and Trt_4 recorded the highest and were not significantly (P>0.05) different from each other. The mean fresh weight recorded was 169.0g. The fresh weight from *Bacillus* based-bio fertilizer shown in Figure 4 was significant (P<0.05) to Trt_1 and Trt_5 recorded the lowest fresh weight and are not significantly (P>0.5) different from each other.



Soaking treatments: Trt1 = 0 hours Trt2 = 8 hours Trt3 = 16 hours Trt4 = 24 hours Trt5 = Control
**Figures not sharing a common letter in a column differ significantly at 0.05 probability.*
Figure 4 Effect of Soaking Seeds with bio fertilizer on fresh weight

Dry Weight

Data regarding effect of *Trichoderma* on dry weight of the plant was significantly (P<0.05) different from each other (Figure 5).

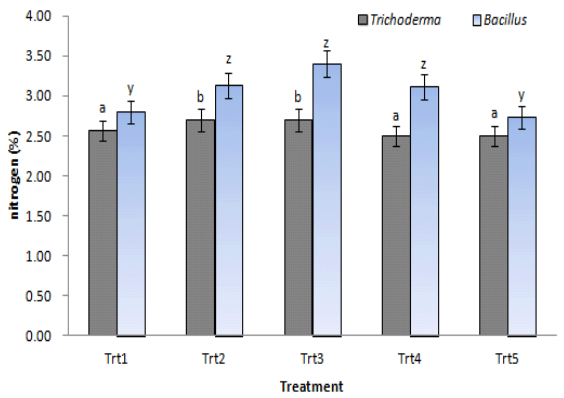


Soaking treatments: Trt1 = 0 hours Trt2 = 8 hours Trt3 = 16 hours Trt4 = 24 hours Trt5 = Control
**Figures not sharing a common letter in a column differ significantly at 0.05 probability.*
Figure 5 Effect of Soaking Seeds with bio fertilizer on dry weight

Trt_1 recorded the lowest (15.0g) dry weight and was not significantly (P>0.05) different from each other. Trt_5 recorded a minimum dry weight of 18.0g. However, the highest dry weight was recorded from Trt_2, Trt_3 and Trt_4 (33.0g, 37.0g and 33.0g respectively) and these were not statistically different from each other. The mean dry weight recorded was 27.0g. Effect on dry weight from soaking seeds with a *Bacillus* bio fertilizer was significantly (P<0.05) different. The least dry weight was recorded Trt4 (15.0g). From Trt_1 to Trt_2 to Trt_3 the dry weight was numerically increasing (17.0g, 27.0g and 28.0g respectively) but not statistically (P>0.05) different. The mean dry weight recorded was 22.0g.

Nitrogen (N)

Effect of bio priming with bio-fertilizers on uptake of nitrogen was significant (P<0.05) as shown in Figure 6.



Soaking treatments: Trt1 = 0 hours Trt2 = 8 hours Trt3 = 16 hours Trt4 = 24 hours Trt5 = Control
**Figures not sharing a common letter in a column differ significantly at 0.05 probability.*
Figure 6 Effect of Soaking Seeds with bio fertilizer on nitrogen content

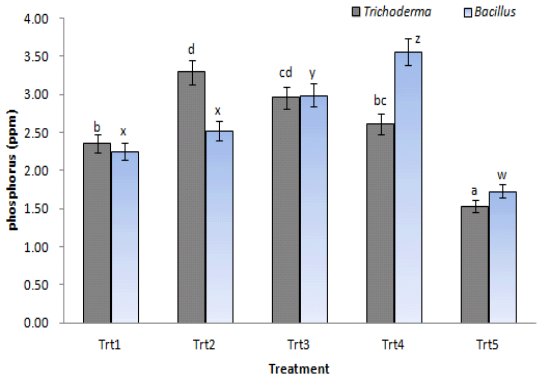
For bio priming with *Trichoderma* bio fertilizer, treatment Trt_2 and Trt_3 were not significantly (P>0.05) different from each other and the two recorded the highest (2.70%) N content percentage. The N percentage content for the control treatment Trt_5 (2.50%) was however not significantly different from percentages recorded from Trt_1 and trt_4 (2.57% and 2.50% respectively). The mean N percentage content recorded was 2.59%.

Bio priming with *Bacillus* recorded significant (P<0.05) differences among the treatments. Treatment Trt_1 which recorded 2.80% N content was however, not statistically (P>0.05) different from that of the control (2.73%). The highest N percentage content was recorded from Trt_3 but was not significantly different from Trt_2 and Trt_4 which recorded 3.13% and 3.12% N content respectively. The mean percentage N content was 3.04%.

Phosphorus (P)

Data regarding tissue phosphorus content as affected by bio priming with *Trichoderma* based bio fertilizer was significant (P<0.05) among treatments (Figure 7). All treatments recoded significantly (P<0.05) higher P content from the control. Treatment Trt_2 recorded the highest tissue P content (3.29 ppm) followed by Trt_3 (2.96 ppm). Trt_1 and Trt_4 were not significantly (P>0.05) different from each other for tissue P and recorded 2.36 ppm and 2.61 ppm respectively. The mean P content recorded was 2.55 ppm.

Bio priming with *Bacillus* was also significant (P<0.05) among the treatments with all the treatment recording means higher than the control. The trend exhibited was that the more the time for soaking the more the tissue P was recorded. The highest tissue P content was recorded for Trt_4 (3.57 ppm). The mean P content recorded was 2.62 ppm.



Soaking treatments: Trt1 = 0 hours Trt2 = 8 hours Trt3 = 16 hours Trt4 = 24 hours Trt5 = Control
**Figures not sharing a common letter in a column differ significantly at 0.05 probability.*
Figure 7 Effect of Soaking Seeds with bio fertilizer on phosphorus content

DISCUSSION

Trichoderma and *Bacillus* based bio fertilizer were evaluated for their effect on growth and development of maize seedlings in early development stage. All seed treatments by bio priming significantly (P<0.05) affected the co-efficient of variation. On average however, seeds from bio priming performed better in plant height, root length, fresh weight and dry weight as compared to the control treatment.

Fundamentally, the height of a plant is a character controlled genetically. However, the bio priming treatments had a significant influence on plant growth characters of the maize seedlings, resulting in improved growth. Numerous findings indicate that the height of a plant can be increased through the application of synthetic plant growth regulators. A significant difference in plant height

between the treatments by bio priming seeds for different time intervals and bio fertilizers used could be due to increased photosynthetic activity, development in the mobilization of photosynthates and change in the membrane permeability (Shukla et al., 1997).

Comparable type of findings were recorded by several researchers when they tried PGPR on different crops (Glick, 1995; Welbaum et al., 2004; Pirlak et al., 2007; Dursun et al., 2008; Jelin et al., 2013). In a study by Jelin et al. (2013) application of *Pseudomonas* strain in maize crop enhanced 17% plant height. These findings are at par with the findings from this current study. Likewise, Pirlak et al. (2007) indicated that the inoculation of *Pseudomonas* and *Bacillus* strains to the apple trees had significantly enhanced the length and diameter of apple stern compared to control. Additionally, Dursun et al. (2008) showed that plant growth promoting rhizobacteria have great prospective to promoting the growth parameters of rocket (*Eruca sativa*) plants. The improved plant growth can be attributed to additional phosphorus (P) as a result of P-solubilizing bacteria. *Bacillus* can mineralize organic P through enzymatic hydrolysis by secreting phosphatase enzymes. It also secrete organic acids such as oxalic, citric, gluconic and lactic acids to solubilize soil inorganic P (Adeleke, Nwangburuka, & Oboirien, 2017; Mohammadi, 2012; Rai, 2006) hence improved plant uptake of P recorded in this study.

The extrametrical fungal hyphae for the *Trichoderma* can extend several centimeters into the soil and absorb large amounts of nutrients for the host root which might have further contributed to higher photosynthesis and bio- chemical activities (Khan et al., 2000). These activities could have been the reasons of the improved results of growth parameter measured. The significantly improved fresh weight by bio priming with both *Trichoderma* and *Bacillus* might be due to enhance growth on plant height and root length. Fresh weight increase simultaneously increases the dry weight as well. Usually, micronutrients (Zn, Fe, Mn, Cu and Mo) form insoluble complexes in the soil, which are not readily accessible by cropswhile *T. harzianum* solubilizes minerals such as metallic Zn and MnO_2 by chelating and reducing mechanisms (Altomare et al., 1999) and these adds minerals along with improved N uptake by the plants as recorded in the study leading to improved plant growth. These results are in accordance with Gasoni et al., (2008) who reported an improvement in the seedling number and fresh weight of table beet by sowing beet seeds coated with a *T. harzianum* isolate. Prashikhan *et al*., (2018) in a study on impact of bio fertilizers on seedling vigor in cashew (*Anacardium occidentale* L.) also observed similar results. Dabiré et al,. (2016) observed accelerated seedling emergence speeds and significantly increased seedling emergence rates, seedling length and weight and root length compared to untreated substrate and uncoated seeds with bio fertilizer. Ozbay and Newman (2004) also reported bio fertilizers to have significantly increased the height, shoot and root dry weight in tomato seedlings transplanted into pots in the green house because of the efficiency if biofertilizers

CONCLUSION

The results of the study provided evidence of improved effect of bio priming treatments on maize seed using the different bio fertilizers. The bio fertilizers, *Trichoderma* and *Bacillus* had a significant influence on plant height, root length, fresh weight, and dry weight. Number of leaves was improved by bio priming treatment with *Bacillus* only. Therefore, when targeting for maximum plant height, fresh weight and dry weight in maize the seed can be soaked for 16hrs. Further soaking for 24 hrs do promote root length which can help assist in accessing water and nutrients from deeper horizons in maize production under marginal production areas.

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