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Increasing Quality Transplants through the Use of Microbial and Soil Amendments against Damping-Off Disease in Onion

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Abstract

The study was undertaken to identify the best microbial and soil amendment in seedbeds that yielded quality transplants and to determine the effect of microbial and soil amendments in seedling production. The following treatments were evaluated: T1-Bacillus subtilis at 25 g; T2-Trichoderma spp. (Asperellum and longibrachiatum) at 25 g; T3-Vermicompost at 1.0 kg; T4-Composted goat manure at 1.0 kg; T5-Mancozeb M-45 at 10 g; and T6-Untreated (no application). The trial was laid in a Randomized Complete Block Design (RCBD) and replicated three times. Observations were done at 10 DAS for % seed germination, 16 DAS, and 25 DAS for % damping-off, % seedling survival at 30 DAS, seedling height, root length, and fresh weight of seedlings at pulling stage (35 DAS). The results of the study showed that the Red Pinoy onion variety has the lowest percentage of damping-off disease from the application of vermicompost at 16 DAS and 25 DAS. Highest percent seed germination was observed at 10 DAS, highest percent seedling survival at 30 DAS, tallest seedlings, and heaviest weight of seedlings were produced by the application of composted goat manure. For Yellow Granex onion variety, Trichoderma spp. obtained lower percentage of damping off at 25 DAS statistically similar to the effect of standard control Mancozeb M-45. Composted goat manure produced highest percent seed germination and vermicompost obtained higher percent seedling survival comparable to the standard control Mancozeb M-45. Results obtained showed a significant (P=0.05) effect on the treatments on the different characters measured. Based on result, application of all treatments have shown fungal activity that promoted better growth and quality of onion seedlings for transplant.

Key Words: vermicompost, onion variety, *Trichoderma spp.*, *Bacillus subtilis*, composted goat manure

Introduction

Success in the production of onion is possible provided that quality seedlings are used for transplants. It is very essential to select the seedlings prior to planting to achieve good bulb yield , (Department of Agriculture, Sri Lanka [DOASL], 2006). Seedling production is an important part in any vegetable production system. Its success depends much on the quality of transplants. Onion is among the crops usually raised in seedling form before being transplanted in the field (Mastouri et al., 2010). In some economic crops, production is done by using reliable producers; thus, the nursery business is increasing. In the nursery, disease management has become a concern during production of transplants (Dabire etal., 2016). They have to be protected from a variety of diseases like damping-off disease which causes serious problems in onion nurseries that it affects the germinating seeds and young seedlings. Some pathogenic fungi including *Pythium* spp., *Phytophthora* sp., *Rhizoctonia solani, Sclerotium rolfsii*, and *Fusarium oxysporum* were reported to be the causative agents of seedling damping-off disease (Adongo et.al.,2015). These soilborne fungi generally do not produce airborne spores but are easily transported from contaminated field soil. A study showed that the capability of two *Trichoderma spp.* to suppress *F. solani* is through formation of loops and coils and attachment of hyphal tips. They also had the ability to produce chitinase and volatile metabolites that controlled the growth of *F. solani* (Gunaratna et al.,2019). Results of the study conducted by Dabire et al. (2016) revealed that the use of an indigenous isolate of *T. harzianum* could be a novel biocontrol strategy against the damping-off of onion in an environmentally sustainable way.

Chemicals are highly effective in preventing and controlling the disease pathogens. However, the intensive use of pesticides has not only created problems of pesticide resistance and increased contamination of the environment but may also have adverse mammalian health effects (Gill & Garg, 2013). Biological control of plant diseases, particularly using microbial and soil amendments against the pathogen, has been expanded and may supplement or serve as an alternative to chemical pest control (Deshayes, 2017). Specifically, the objectives of the study were to: identify the best microbial and soil amendment in seedbeds that yield quality transplants and to determine the effect of the combination of microbial and soil amendments in seedling production.

Materials and Methods

Experimental Area and Design

The study was conducted in the research experimental area of Central Luzon State University, Science City of Muñoz, Nueva Ecija with a previous history of damping-off organisms. The area was thoroughly prepared before sowing. Each raised seedbed of about 30 cm high measured about 50 x 300 cm or 1.5m². The study was laid out following the Randomized Complete Block Design with three replications. Distance between block and treatment was 1 meter.

The following treatments were evaluated:

	Rate of application Amount[a] g/ml/seedbed (1.5m²)
T1-*Bacillus subtilis*	25 g
T2-*Trichoderma* spp. (*asperellum* and *longibrachiatum*)	25 g
T3-Vermicompost	1 kg
T4-Composted goat manure	1 kg
T5-Mancozeb M-45	10 g
T6-Untreated	none

[a] Volume of water used per seedbed was 3 liters

Sowing of Seeds and Treatment Application

Red Pinoy and Yellow Granex onion varieties were used. Seeds were sown in line on seedbeds at the rate of 10 g/1.5 m². After sowing, seedbeds for Treatments 1 and 2 were drenched with *Bacillus subtilis and Trichoderma* spp. at the rate of 25 g/3 L of water per seedbed, while in Treatments 3 and 4 were broadcasted with vermicompost and composted goat manure at the rate of 1 kg/seedbed/treatment.

Mancozeb M-45 was applied for Treatment 5 by drenching method at the rate of 10 g/3 L of water per seedbed. Plots were covered with dried rice straw to allow better germination and growth of onion seedlings. Treatment application was done 9 DAS, 16 DAS, and 23 DAS for *Bacillus subtilis* and *Trichoderma spp.,* while vermicompost and composted goat manure were incorporated before seed sowing in between rows at 16 DAS and 23 DAS.

Evaluation / Assessment

The effects of treatments were assessed based on the number of germinated seeds at 10 DAS, percent damping-off at 16 and 25 DAS, and percentage seedling survival at 30 DAS taken in 25 x 25 cm quadrant. At harvest (35 DAS), seedling height and root length were taken from 20 randomly selected plant samples. Fresh weight of seedlings at 35 DAS was taken from 25cm x 25cm quadrant at pulling stage (Patricio, 2017).

Statistical Analysis

Statistical Analysis System (SAS) software for statistical analysis of collected data was used for the analysis of variance (ANOVA) in Randomized Complete Block Design (RCBD) and LSD test at $P<0.05$ was used for mean comparison among treatments (Lawson, 2010).

Results and Discussion

Red Pinoy Variety

For the Red Pinoy variety, the highest percentage germination was recorded using composted goat manure. However, no significant differences were gathered using Mancozeb M-45, *Bacillus subtillis, Trichoderma* spp., and vermicompost.

On the occurrence of damping-off, the application of vermicompost obtained the least percentage of damping-off in two observations at 16 DAS and 25 DAS. Moreover, at 16 DAS, vermicompost had the lowest percentage incidence of damping-off comparable with *Bacillus subtillis*. At 25 DAS, the lowest percentage of damping-off using vermicompost was comparable with *Trichoderma* spp.

Meanwhile, composted goat manure resulted as the top most in terms of the percentage seedling survival 30 DAS (62.54 %) and the untreated had the least survived seedlings of 51.36%, respectively (Table 1). Results showed that application of composted goat manure had the highest percentage survival which means that it was an effective treatment as microbial and soil amendment that can be used in improving the quality of transplants to withstand the infection of damping-off disease in onion.

Table 1

Red Onion Variety Seed Germination, Damping-Off, and Seedling Survival as Affected by Different Treatments

Treatments	% Seed Germination 10 DAS	% Damping-off 16 DAS	% Damping-off 25 DAS	% Seedling Survival 30 DAS
T1-*Bacillus subtilis*	69.76[a]	7.48[bc]	17.96[a]	57.12[ab]
T2-*Trichoderma* spp. (*asperellum* and l*ongibrachiatum*)	67.45[ab]	11.12[ab]	14.16[ab]	56.98[ab]
T3-Vermicompost	67.63[ab]	5.21[c]	9.86[b]	61.00[a]
T4-Composted Goat manure	74.38[a]	12.75[a]	15.79[a]	62.54[a]
T5- Mancozeb M-45	72.65[a]	13.23[a]	14.57[a]	61.75[a]
T6-Untreated	59.55[b]	9.83[ab]	13.62[ab]	51.36[b]

Table 2 shows that composted goat manure produced the tallest seedling height but the increase did not differ significantly with vermicompost and Dithane 45 fungicide, but was better than untreated control plants. All treatment application had no influenced in root length as indicated by insignificant differences.

Regarding the weight of seedlings, composted goat manure provided the heaviest seedlings presumed to have more number of harvested seedlings Tilley (2018) mentioned that composted goat manure can add nutrients to the soil, promote healthier plant growth, and increase crop yields without the use of chemicals. He stated further that this manure contains adequate amounts of the nutrients that plants need for optimal growth.

The lightest among treated seedlings was obtained in *Bacillus subtilis* attributed by a decrease in survival rate. Mancozeb M-45 resulted on lighter weight of seedlings at par with *Trichoderma* spp. The untreated produced the lowest weight of seedlings among treatments.

Table 2

Red Onion Variety Seedling Height, Root Length, and Weight at 35 DAS as Affected by Different Treatments

Treatments	Seedling height (cm)	Root length (cm)	Fresh weight of seedlings in grams (25 x25 cm) per quadrant
T1-*Bacillus subtilis*	26.41[cd]	6.8	321.4[d]
T2-*Trichoderma* spp. (*asperellum* and *longibrachiatum*)	26.78[bc]	6.4	404.4[bc]
T3-Vermicompost	29.06[ab]	7.1	437.0[b]
	30.87	6.8	480.1[a]

Table 2 (cont'd)

Red Onion Variety Seedling Height, Root Length, and Weight at 35 DAS as Affected by Different Treatments

Treatments	Seedling height (cm)	Root length (cm)	Fresh weight of seedlings in grams (25 x25 cm) per quadrant
T4-Composted goat manure	30.87^{a}	6.8	480.1^{a}
T5- Mancozeb M-45	28.77^{ab}	6.6	383.9^{c}
T6-Untreated	23.99^{d}	6.8	257.0^{e}

Yellow Granex Variety

For the Yellow Granex variety, as presented on Table 3, the percentage seed germination in composted goat manure plants was comparable with vermicompost, *Trichoderma* spp., and Mancozeb M-45.

At 25 DAS, Mancozeb M-45 had the lowest damping-off but not significantly different from *Trichoderma* spp.

For percent seedling survival, Mancozeb M-45 had the highest percentage seedling survival with a mean of 49.25 percent, comparable to vermicompost and composted goat manure and *Trichoderma* spp. with percentages seedling survival of 49.22%, 48.15% ,and 48.04%, respectively. As stated by Dabire et al. (2016), the use of an indigenous isolate of *Trichoderma harzianum* could be a novel biocontrol strategy against the damping-off of onion seeds in an environmentally sustainable way. Similarly, Mastouri et al. (2010) mentioned that *Trichoderma* spp. are endophytic plant symbionts that are widely used as seedling treatments to control diseases and to enhance plant growth and yield.

Table 3

Yellow Granex Variety Percent Seed Germination, Damping-Off, and Seedling Survival as Affected by Different Treatments

Treatments	% Seed Germination 10 DAS	% Damping-off 16 DAS	% Damping-off 25 DAS	% Seedling Survival 30 DAS
T1-*Bacillus subtilis*				44.02^{bc}
T2-*Trichoderma* spp.	51.99^{b}	8.43	16.84^{ab}	
(*asperellum* and				48.04^{ab}
longibrachiatum)	56.03^{ab}	7.83	14.26^{c}	49.22^{a}
T3-Vermicompost	58.87^{a}	6.61	16.43^{b}	48.15^{ab}
T4-Composted Goat	58.97^{a}	8.61	18.37^{a}	49.25^{a}
manure	57.51^{ab}	8.76	14.11^{c}	42.56^{c}
T5- Mancozeb M-45	52.49^{b}	8.47	16.63^{ab}	
T6-Untreated				

Table 4 shows no significant differences on seedling height, and root length that were observed among treatments. The heaviest weight of seedlings was recorded in using vermicompost but found similar in the application of *Trichoderma* spp., composted goat manure, and Mancozeb M-45. A similar study was conducted by Abolmaaty and Fawas (2016) which showed that vermicompost treatment at the rate of 5 tons/fed combined with EM1 treatment was the best treatment for increasing the weight of onion bulb more than the control treatment.

Untreated onion plants had the lightest seedling weight comparable to *Bacillus subtilis*.

Table 4

Yellow Granex Variety Seedling Height and Root Length and Weight (35 DAS) as Affected by Different Treatments

Treatments	Seedling height (cm)	Root length (cm)	Fresh weight of seedlings in grams (25 x25 cm) per quadrant
T1-*Bacillus subtilis*	27.8	5.19	212.9[b]
T2-*Trichoderma* spp. (*asperellum* and *longibrachiatum*)	26.9	5.29	233.8[a]
T3-Vermicompost	28.1	5.34	238.1[a]
T4-Composted Goat manure	27.6	5.47	231.9[a]
T5- Mancozeb M-45	27.4	5.50	229.9[a]
T6-Untreated	26.0	5.23	211.7[b]

T. clypeatus was reported in Southern and Central Luzon by previous researchers (Arenas et al., 2015; Tadiosa et al., 2011; De Leon et al., 2015; De Castro & Dulay, 2015) who based their identification on the morphological features of their collection. *T. bulborhizus* was not mentioned in the said reports, however two unidentified species of *Termitomyces* collected in the provinces of Central Luzon namely, Pampanga, Tarlac, and Zambales were emphasized by De Leon et al. (2015) in addition to *T. clypeatus, T. striatus,* and *T. robustus*. In this paper, most of the samples examined which were obtained from different geographical areas in Central Luzon are *T. bulborhizus.*

Conclusion

The result of this study concluded that soil amendments and biocontrol agents applied to the two onion varieties have shown antifungal activity against the organisms tested. The degree of fungal activity with the corresponding rates of application was associated with the varieties of onion with regards to the different growth and seedling yield parameters. Generally, application of all treatments have shown fungal activity that promoted better quality of seedlings for transplant. Further evaluation using standardized application techniques will provide more comprehensive information about the effect of antifungal activities of the different treatments used in the study against common fungal pathogens that affect the growth and quality of onion seedlings in nurseries.

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