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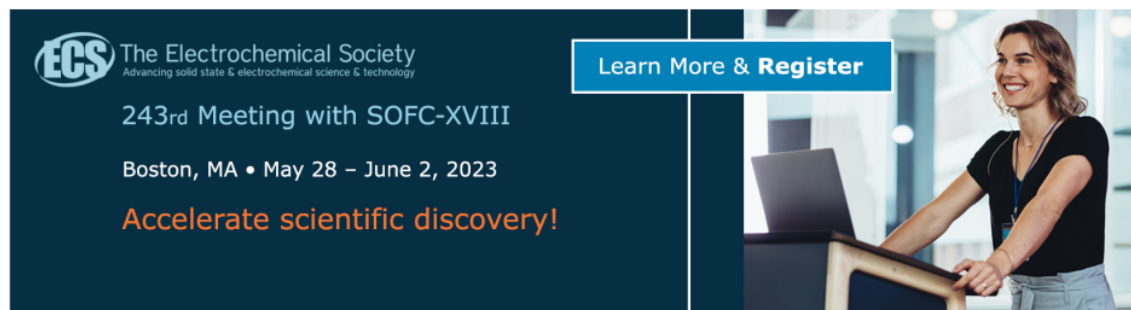
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Growth and production of red onion (*Allium ascalonicum* L.) with the application of monosodium glutamate and endophytic fungi

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Abstract. This study aimed to reveal the effect of giving monosodium glutamate (MSG) and endophytic fungi (*Trichoderma* sp., *Mycorrhiza*, and *Beauveria bassiana*) to red onion (*Allium ascalonicum* L.) plants which predicted to increase its growth and production while decrease diseases infection without harming the environment. The experiment was designed by Factorial-Randomized Block Design. There were two factors tested in this present study: (1) MSG treatment with three dosages: 0 g L⁻¹, 5 g L⁻¹, and 10 g L⁻¹; (2) endophytic fungi treatments with single or pure culture and mix-culture combinations: *Trichoderma* sp., *Beauveria bassiana*, *Mycorrhiza*, the combination of *Trichoderma* sp. and *Beauveria bassiana*, the combination of *Trichoderma* sp. and *Mycorrhiza*, the combination of *Mycorrhiza* and *Beauveria bassiana*, and the combination of *Trichoderma* sp., *Beauveria bassiana*, and *Mycorrhiza*. The parameters measured were plant growth, harvest, and disease infection intensity. The treatments did not give significant improvement to all parameters. This study concluded that *Mycorrhiza* treatment produced the highest rate value in red onion harvest.

Keywords: Endophytic fungi, monosodium glutamate, red onion

1. Introduction

Red onion cultivations have been carried out in all provinces and designated as one of seven strategic commodities by the Indonesian Government. The increase of the human population in the world is causing the increase of red onion's demand [1]. However, the red onion crop productivity is not easy to be increased due to several problems faced during the cultivation process.

Giving fertilizers additional nutrients is one of the crucial factors to improve crop quality and productivity. Chemical-based fertilizers have been used by farmers intensively which further leads to environmental problems. Therefore, developing organic fertilizers is highly required to replace the previous one. Liu et al. [2] revealed that there is a possibility to replace chemical-based fertilizers with monosodium glutamate (MSG) due to it contains nutrients that suitable for supporting plant growth. The high sodium content in MSG will increase the plant fertility level, accelerate plant growth, stimulate the appearance of flowers, and provide nutrition for plants [3]. Nutrients in MSG have been reported to be



similar to those contained in the chicken manure which is very beneficial for soil microbes [4]. Besides, to increase red onion productivity, the number of pathogens' infection during the cultivation is another problem that needs to be solved.

Fungicide and pesticides are often used excessively to kill pathogens which further the excess of these chemicals causes environmental problems in the long-term application. As the alternative strategy, endophytic fungi, living microorganisms in plant tissues have been reported to be potential biological control agents to kill pathogenic microorganisms while reducing fungicide application [5]. Biological agents that can be used in red onion farming are *Beauveria bassiana*, *Mycorrhiza*, and *Trichoderma* sp. The application of *Beauveria bassiana* on red onion can suppress the attack of red onion caterpillar (*Spodoptera exigua*); reduce pathogenic microorganisms attack; increase plant productivity by providing additional nutrition [6]. *Trichoderma* sp. is an endophytic and antagonist fungi for plant pathogenic microorganisms that can be also acted as plant growth-promoting fungi (PGPF) by colonizing inside the roots and stimulates plant growth [7]. Other fungi belong to the PGPF group is *Mycorrhiza*. *Mycorrhiza* and *Trichoderma* sp. can increase the surface area of the plant root transportation system. *Mycorrhiza*'s hyphae produce enzymes and organic acid to accelerate the formation of the P element, whereas *Trichoderma* sp. facilitates plant growth and increases biological activity which beneficial for soil microorganisms [8,9]. Therefore, *Mycorrhiza* and *Trichoderma* sp. can be considered as potential fungi for red onion cultivation [10,11]. This research is aimed to study the effect of MSG and endophytic fungi (*Trichoderma* sp., *Mycorrhiza*, *Beauveria bassiana*) application to red onion cultivation to improve its growth and production while also revealed its ability to reduce plant diseases infection.

2. Methods

2.1. Experimental design

This research was conducted at the Experimental Farm, Faculty of Agriculture, Hasanuddin University Makassar, South-Sulawesi Province, Indonesia. The cultivation of red onion started from seed plantation until harvesting took place during the long dry season in Makassar, Indonesia from August to November 2019. The temperatures were ranged from 24.5°C - 28.9°C, altitudes were around 1 - 22 m above sea level. The place was at 119°24'17"38 "E and 5°8'6"19" S. The research was carried out from July to November 2019. Red onion bulbs of Super Philip variety, MSG, and endophytic fungi cultures (*Trichoderma* sp., *Beauveria bassiana*, and *Mycorrhiza*) were obtained from the collection of the Faculty of Agriculture, Hasanuddin University Makassar and the office of Makassar Forestry and Research Institute. The experimental plot was arranged in a randomized Block Design consisted two factors: the dose of MSG and the type of endophytic fungi. The MSG level treatments were 0 g L⁻¹ / control (M0), 5 g L⁻¹ (M1), and 10 g L⁻¹ (M2). Eight groups of endophytic fungi treatments used were: negative control or without the augmentation of any endophytic fungi (C0), *Trichoderma* sp. (C1), *Beauveria bassiana* (C2), *Mycorrhiza* (C3), the mix of *Trichoderma* sp. and *Beauveria bassiana* (C4), the mix of *Trichoderma* sp. and *Mycorrhiza* (C5), the mix of *Mycorrhiza* and *Beauveria bassiana* (C6), the mix of *Trichoderma* sp., *Beauveria bassiana* and *Mycorrhiza* (C7).

The experiment of each treatment was repeated 3 times so that 72 experimental plots were prepared. The size area of each plot used was 1.4 m x 1.4 m, the distance between treatments was 20 x 15 cm, the distance between plots was formed as drainage canals with an additional 40 cm wide. The height of each plot was 20 cm. The basic fertilization using goat dung was held 2 weeks before planting. Goat dung as fertilizer was applied on the field/experimental plots with a dose of 10 tons ha⁻¹ for each treatment.

The endophytic fungi were sub-cultured into potato dextrose agar (PDA) and incubated for 7 days at room temperature (25°C). The fungi were transferred into rice media then incubated for 14 days at 25°C until ready to be used.

Trichoderma sp. was applied in 2 weeks before planting with a dose of 5 g per planting hole (with a total spore density of 5.8 x 10⁹). *Mycorrhiza* was applied together at the time of planting with a dose of

3 g placed under the planting hole [12]. *Beauveria bassiana* in the form of a growing solid was separated from the medium then weighed to 10 g L⁻¹ water (total spore density 8.6 x 10⁹). Afterwards, the fungi were filtered by sieve while adding water. The suspension as a result of the filtering fungi was then applied to the red onion from age 2 weeks until before the harvesting time (7 days interval with 6 times spraying). Fungi suspension was sprayed on a red onion plantation with a volume of 1 L per plot. The MSG treatment was given after the plant has grown healthy. The plant was treated according to the concentration of each treatment 2 times, by watering the area around the bulbs for 50 mL for 21 days after planting (DAP) and 75 mL at 28 DAP. The growth of red onions was maintained by re-planting, irrigation/watering, weed control, soil management, also pest and disease control according to the experimental application.

2.2. Parameters and data collection

The research data were collected based on the field observations in each trial plot, from the beginning of the planting to the time of harvesting. The parameters observed were plant height (cm), number of leaves per plant (leaves), wet weight of red onion waste product (g), dry weight of red onion waste product (g), dry weight of bulbs (g), the diameter of bulbs (cm), and disease infection intensity (%). The calculation of disease intensity can be obtained by the equation as follows [13]:

$$P = (A / N) \times 100\%$$

P = Damaged plant level (%)

A = Total of infected plants

N = Total of plants observed

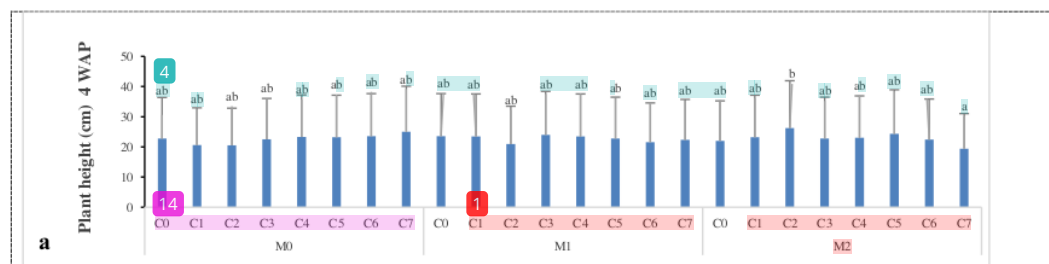
2.3. Data analysis

The data obtained were analyzed statistically based on two-way analysis of variance (ANOVA) using IBM SPSS Statistic 24 for Windows 8.1 and then followed by Duncan's multiple range test (DMRT) to determine the differences of the data at a significance level of 5%.

3. Results

3.1. Plant growth

Plant height and number of leaves are used to assess plant growth parameters. MSG and endophytic fungi into red onion plants in this present study did not have a significant ($p > 0.05$) effect on plant growth based on the plant height (Figure 1a-b) and the number of leaves (Figure 2a-b) both at 4 and 6 Week After Planting (WAP) observations. The highest average value of plant height at 4 WAP was 26.20 cm, the treatment with 10 g L⁻¹ MSG and *Beauveria bassiana* (Figure 1a), while at 6 WAP the highest average value was 26.66 cm treated with 5 g L⁻¹ MSG and *Trichoderma* sp (Figure 1b). In the number of leaves term, the highest average value was 17.25 leaves obtained at 4 WAP with the combination of 5 g L⁻¹ MSG and *Trichoderma* sp. and *Mycorrhiza*, (Figure 2a). At 6 WAP, the highest value was 24.48 leaves found in the mix of 5 g L⁻¹ MSG and *Trichoderma* sp., *Beauveria bassiana*, and *Mycorrhiza* treatment (Figure 2b).



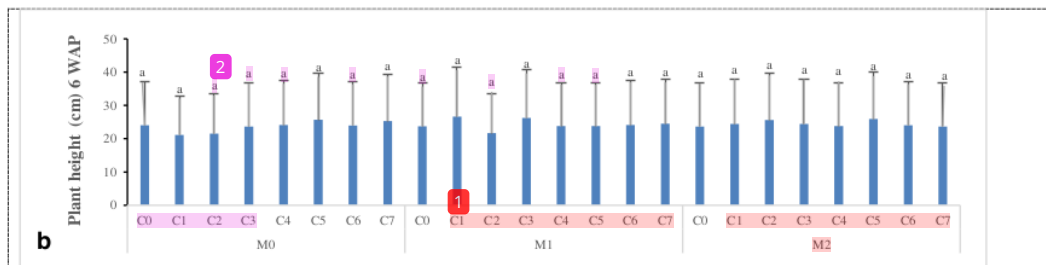


Figure 1. The average plant height at 4 (a) and 6 (b) WAP. The different letters in the same graph indicate significantly different based on Duncan's test $\alpha = 0.05$. Control (M0), 5 g L⁻¹ MSG (M1), 10 g L⁻¹ MSG (M2), control (C0), *Trichoderma* sp.(C1), *Beauveria bassiana* (C2), *Mycorrhiza* (C3), *Trichoderma* sp. and *Beauveria bassiana* (C4), *Trichoderma* sp. and *Mycorrhiza* (C5), *Mycorrhiza* and *Beauveria bassiana* (C6), *Trichoderma* sp., *Beauveria bassiana* and *Mycorrhiza* (C7).

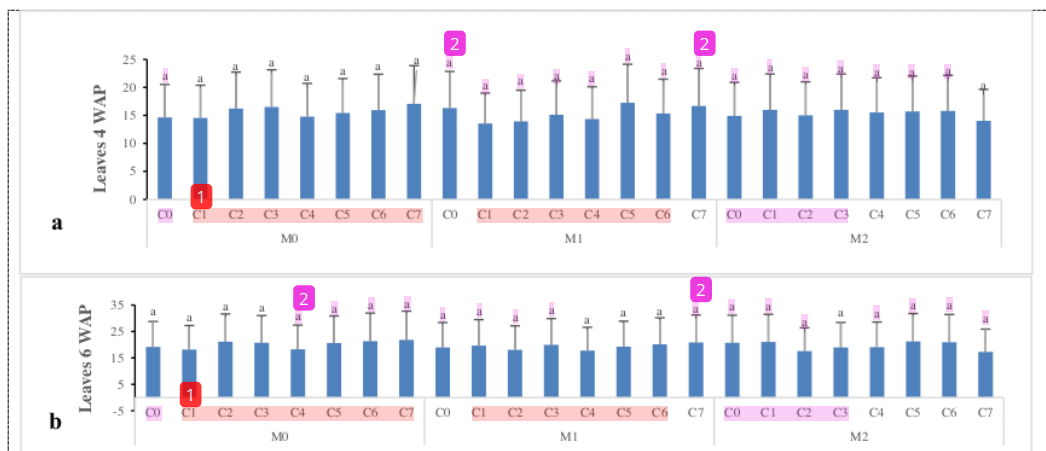


Figure 2. The average of plant leaves at 4 (a) and 6 (b) WAP. The different letters in the same graph indicate significantly different based on Duncan's test $\alpha = 0.05$. Control (M0), 5 g L⁻¹ MSG (M1), 10 g L⁻¹ MSG (M2), control (C0), *Trichoderma* sp. (C1), *Beauveria bassiana* (C2), *Mycorrhiza* (C3), *Trichoderma* sp. and *Beauveria bassiana* (C4), *Trichoderma* sp. and *Mycorrhiza* (C5), *Mycorrhiza* and *Beauveria bassiana* (C6), *Trichoderma* sp., *Beauveria bassiana* and *Mycorrhiza* (C7).

3.2. Red onion production quality

The production of red onions after giving the treatments with MSG and endophytic fungi was monitored based on the diameter of red onion bulbs, wet weight of red onion waste product, dry weight of the waste product, and dry weight of red onion bulbs. The wet weight of red onion waste measured bulb and leaves after harvest, whereas the dry weight of weight product measured bulb and leaves after 2 weeks. The data was collected after harvesting the red onion plantation. It can be seen that the treatment of fungi alone gave a significant effect ($p < 0.05$) on the four observed parameters, whereas conversely the treatment of MSG alone and the combination of fungi with MSG showed no significant effect ($p > 0.05$) (Figure 3). Control and *Mycorrhiza* and *Beauveria bassiana* treatment produced the highest average

value of 2.05 cm diameter of the bulb (Figure 3a). Mycorrhiza produced the highest average value of 35.62 g wet weight of the waste product (Figure 3b), *Trichoderma* sp. *Mycorrhiza* alone produced the highest average value of 27.41 g dry weight of the waste product (Figure 3c). The treatment with *Mycorrhiza* produced the highest value, 31.33 g dry weight of red onion bulbs (Figure 3d).

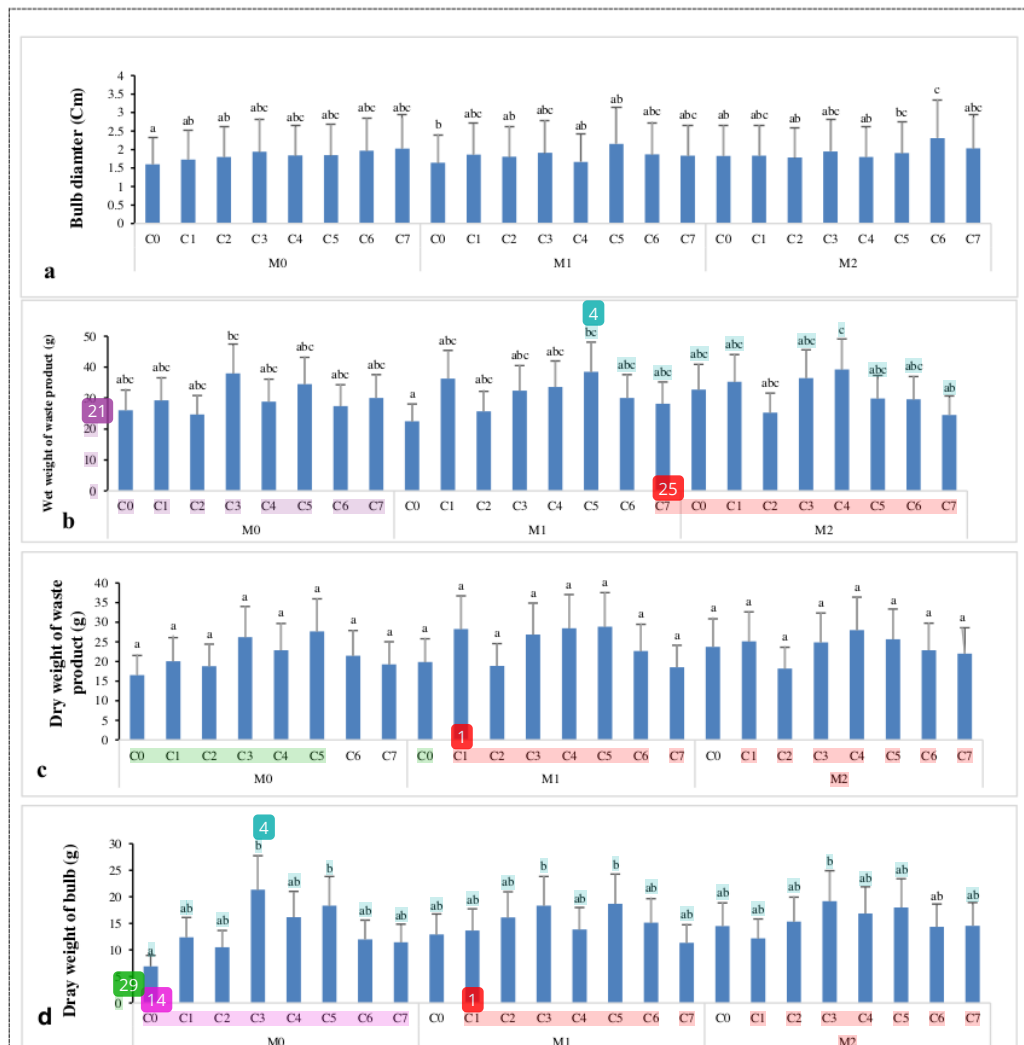


Figure 3. The average of red onion bulbs diameter (a), wet weight of the waste product (b), dry weight of the waste product (c), and dry weight of bulbs (d). The different letters in the same column indicate significantly different based on Duncan's test $\alpha = 0.05$. Control (M0), 5 g L⁻¹ MSG (M1), 10 g L⁻¹ MSG (M2), control (C0), *Trichoderma* sp. (C1), *Beauveria bassiana* (C2), *Mycorrhiza* (C3), *Trichoderma* sp. and *Beauveria bassiana* (C4), *Trichoderma* sp. and *Mycorrhiza* (C5), *Mycorrhiza* and *Beauveria bassiana* (C6), *Trichoderma* sp., *Beauveria bassiana* and *Mycorrhiza* (C7).

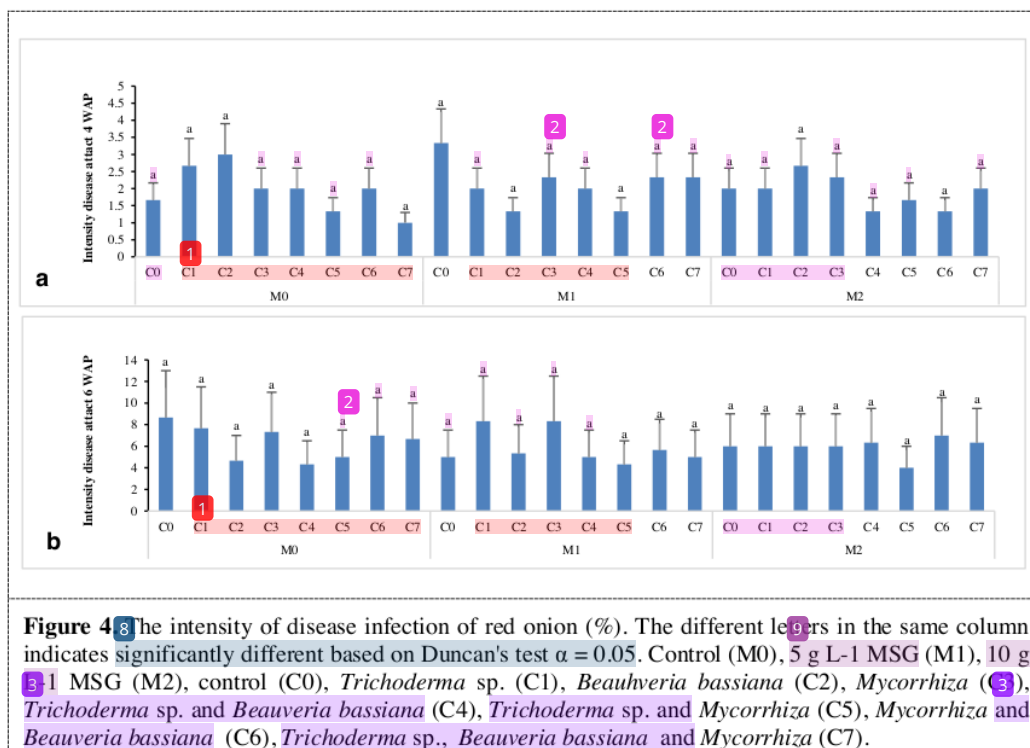


Figure 4 The intensity of disease infection of red onion (%). The different letters in the same column indicates significantly different based on Duncan's test $\alpha = 0.05$. Control (M0), 5 g L⁻¹ MSG (M1), 10 g L⁻¹ MSG (M2), control (C0), *Trichoderma* sp. (C1), *Beauveria bassiana* (C2), *Mycorrhiza* (C3), *Trichoderma* sp. and *Beauveria bassiana* (C4), *Trichoderma* sp. and *Mycorrhiza* (C5), *Mycorrhiza* and *Beauveria bassiana* (C6), *Trichoderma* sp., *Beauveria bassiana* and *Mycorrhiza* (C7).

3.3. The intensity of disease infection

The statistical analysis result showed that giving MSG alone, fungi alone, and a combination of fungi and MSG did not have a significant effect ($p > 0.05$) on the intensity of disease infection at both observation times at 4 and 6 WAP. However, the treatment of *Trichoderma* sp., *Beauveria bassiana*, and *Mycorrhiza* without MSG was showed to produce the lowest intensity of disease infection (1.00%) at 4 WAP and (4.00%) at 6 WAP compared to other doses (Figure 4).

4. Discussion

This present study tried to explore the effect of giving MSG, endophytic fungi, and a combination of MSG and endophytic fungi in red onion growth and production. Unfortunately, the result gave contrary findings. All trials that have been tested in this present study did not give significant results to all parameters monitored. However, giving *Mycorrhiza* showed a potential effect to increase the production of red onion compared with no-*Mycorrhiza* treatments. The combination of *Mycorrhiza* and *Beauveria bassiana* produced the highest diameter of bulbs while giving *Mycorrhiza* alone produced the highest wet weight of the waste product and dry weight of the bulb, and the combination of *Trichoderma* sp. and *Mycorrhiza* produced the highest dry weight of the waste product. This result was similar to a study conducted by Bolandnazar [14] who revealed that giving *Mycorrhiza* will help to produce large diameter sizes of bulbs of red onion. According to Farias et al. [15], the greater root development associated with increased *Mycorrhiza* colonization in plants could give more efficient absorption of nutrients and water. *Mycorrhiza* colonization enhances plant growth by increasing the hydraulic conductivity of red onion to improve water absorption [14]. Moreover, the development of plant roots can be improved by

Arbuscular mycorrhiza (AM) [16]. AM reduces the number of zinc and selenium in the soil and stimulates plant roots development [17].

The combination of *Trichoderma* sp., *Beauveria bassiana*, and *MychMycorrhiza*duced the lowest percentage of disease infection intensity. This result was in line with a previous study which used a combination of beneficial microbes such as *Pseudomonas fluorescens*, *Trichoderma harzianum*, and *Glomus intraradices* will be better at controlling the *Fusarium* wilt disease in tomato plants compared to every single microbial treatment [18,19]. However, the combination of endophytic fungi sometimes could not provide better or even give worse results compared with a single treatment [20]. The lack of interaction between endophytic fungi and plants might be the reason why the experiments were done in this present study could not be able to reduce the intensity of disease infection in the field. This poor interaction can be due to climatic conditions that greatly affect the development of plants. The best endophytic fungi isolates in triggering plant growth were not necessarily as good as in response to various environmental stresses [21].

The application of MSG to the red onion cultivation has no significant effect on all of the parameters tested. This might be presumably due to the environmental conditions of growing red onion plants which experienced drought stress. The ability of plant adaptation in drought conditions has a strong relationship with plant production. Drought stress conditions limit plants to grow well especially during the reproduction process which can be showed by limiting leaf areas such as losing more cell expansion and chlorophyll content in leaves color [14]. While in the combination of MSG and endophytic fungi, both factors did not have a mutually influential relationship to improve the growth and production of red onion. As the result described, it can be seen that each factor affecting red onion growth and production separately from each other. This is consistent with the previous study explained when the effect of different interactions was not real, it was concluded that among the treatment factors acting independently of one to another [22].

5. Conclusions

MSG treatments did not affect all parameters observed, but *Mychorrhiza* treatments give an improvement of the red onion production. Moreover, there is no interaction between the use of MSG and endophytic fungi in enhancing the red onion growth, production, and decreasing the infection of disease. Further research is needed to determine the effect of monosodium glutamate and other types of endophytic fungi on growth parameters both in the rain and dry season because different seasons and environmental conditions will have a different effect on plants. In addition, it would be better if, in future research, the nutrient or Physico-chemical content of red onion is considered as the additional parameters that should be measured.

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