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Enhancing Maize (Zea mays L.) Growth Through Inoculation with Arbuscular Mycorrhizal Fungi (AMF)

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ABSTRACT

This study aims to analyze the effects of arbuscular mycorrhizal fungi (AMF) inoculation on the growth and yield of maize (*Zea mays* L.) as a solution to fertilizer shortages in Indonesia. The continuous use of inorganic fertilizers can degrade soil quality, making environmentally friendly alternatives, such as biofertilizers, necessary. The study observed the growth optimization of several corn varieties inoculated with AMF. The eight-month study was conducted in the Biology Laboratory at Cenderawasih University's Faculty of Mathematics and Natural Sciences using a pot culture method with three varieties of maize: regular maize (RM), sweet corn (SC), and butter sweet corn (BSC). Parameters observed included plant height, leaf number, and relative growth rate based on dry weight. The results showed that AMF inoculation significantly increased corn plant height by 21.56%, leaf number by 18.14%, and the relative growth rate by 1,425% at six weeks after planting. Butter sweet corn and sweet corn was given 15 g of AMF inoculation per polybag.

Key words: AMF; biofertilizer; growth; Zea mays.

INTRODUCTION

Fertilizers are one of the essential inputs in agricultural cultivation (Maheta *et al.*, 2023). The Ministry of Agriculture has made efforts to improve performance achievements, one of which is by enhancing the availability and facilities for fertilizer production. Fertilizers in agriculture are categorized into two types: subsidized fertilizers, which are part of a government program, and non-subsidized fertilizers, including inorganic, organic, biofertilizers, and soil amendments. Fertilizers play a significant role in increasing the cropping index, ultimately contributing to higher agricultural productivity. However, fertilizers also

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represent a considerable production cost burden for farmers. Therefore, the government has promoted programs that encourage farmers to independently produce organic fertilizers, biofertilizers, soil amendments, or natural pesticides, and utilize them as alternative solutions to the rising cost of commercial fertilizers (Ministry of Agriculture, 2023).

The use of biofertilizers can serve as an alternative to improve plant quality compared to organic fertilizers. One of the most popular biofertilizers today is mycorrhizal biofertilizer (Hazrah *et al.*, 2023). Arbuscular Mycorrhizal Fungi (AMF) are biofertilizers expected to enhance the content of nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), and magnesium (Mg) in plants. AMF colonization in roots promotes increased root branching and the elongation of secondary roots, thereby improving nutrient uptake in maize plants. The limited availability of

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Figure 1. Structure of AMF in the roots of *Z. mays*. V: vesicle, IH: intraradical hyphae.

agricultural land has led to increased reliance on biofertilizers, including AMF, highlighting the need for support in large-scale AMF production (Aryanto *et al.*, 2018).

Maize remains the second most strategic crop after rice, due to its high economic value. Besides its role as food and feed, maize is increasingly being utilized as a source of bioenergy (fuel) and raw material for various industries, with demand rising annually (Silitonga et al., 2023). AMF, as a biofertilizer, offers a promising approach to reduce dependence on inorganic fertilizers. Further research and development are needed to optimize the use of AMF. Therefore, this study focuses on optimizing the growth of Zea mays through AMF inoculation. This research holds strategic value not only as a solution to the rising cost of fertilizers but also in building a sustainable agricultural system. As a biofertilizer, AMF offers multiple benefits, including increased productivity of maize-a high-value commodity-and the preservation of soil health.

MATERIALS AND METHODS

Time and Research Location

This study was conducted over a period of 8 months at the Biology Laboratory, Faculty of

Mathematics and Natural Sciences, Cenderawasih University, using an experimental method involving the inoculation of arbuscular mycorrhizal fungi into soil via a pot culture technique.

Tools and Materials

The tools used in this study included a microscope, cover glass, object glass, sterilization drums for soil, analytical balance, polybags, watering cans, and woven boards for plant drying. The materials used consisted of *Glomus etunicatum*, AMF obtained from SEAMEO BIOTROP Bogor, sterilized soil, corn seeds, zeolite, trypan blue root stain solution, distilled water, FAA solution, 10% KOH, 2% HCl, and lactoglycerol.

Experimental Procedure

AMF was inoculated into *Z. mays* plants using pots. The corn seeds used included three varieties: regular corn, sweet corn, and buttery sweet corn. Observations were conducted until the vegetative growth phase of *Z. mays*.

This research used a Completely Randomized Design (CRD) with a factorial pattern involving two factors: *first factor*: The amount of AMF propagule inoculation: 0, 10, 15, and 20 grams of zeolite medium containing AMF obtained from Bogor; *second factor*: Three corn varieties – regular maize (RM), sweet corn (SC), and buttery sweet corn (SCM).

The root staining technique was employed to observe AMF colonization on the plant roots. Root colonization was indicated by the presence of hyphae, vesicles, arbuscules, or any combination of these structures (Pulungan, 2013). The hyphae formed bulbous suspensors or rounded hyphal bases and had auxiliary cells, which are referred to as external vesicles, appearing bright yellow in color (Nusantara *et al.*, 2012).

Data Analysis

The parameters used in this study were plant height and number of leaves, which were analyzed using Analysis of Variance (Anova). If significant differences were observed between treatments, the analysis was continued with Duncan's Multiple Range Test (DMRT) using SPSS version 22.0. Furthermore, the relative growth rate (RGR) was calculated following the equation (Suharno *et al.*, 2017):

$$RGR = \frac{lnW2 - lnW1}{T2 - T1}$$

where:

RGR = Relative Growth Rate

 W_2 = Dry weight at measurement 2

 W_1 = Dry weight at measurement 1 T_2 = Time of measurement 2

 T_1 = Time of measurement 1.

MATERIALS AND METHODS

The results of this study indicate that Arbuscular Mycorrhizal Fungi (AMF) are compatible and capable of forming symbiotic associations with maize (*Z. mays*). This is evidenced by the structural formation within the root tissues of the plant (Figure 1), consisting of intraradical hyphae and vesicles.

Mycorrhizal association in maize can be identified through the formation of structures such as vesicles, intraradical hyphae, arbuscules, and intraradical spores. Root colonization reflects the level of mycorrhizal colonization in plant roots. The life cycle of AMF begins with the development of spores. Once AMF penetrates the host root, it forms various structures including extraradical intraradical hyphae, hyphae, arbuscules, and vesicles. Arbuscules are the sites of carbon, energy, and nutrient exchange between the host plant and AMF. Arbuscules have a shorter lifespan compared to hyphae and vesicles; they exist only during the initial phase of colonization, whereas hyphae and vesicles can persist for several months (Alayya & Prasetya, 2022; Sobat, 2022).

According to research by Alayya & Prasetya (2022), among sweet potatoes, maize, cassava, and rice, maize had the highest AMF colonization. AMF colonization can be influenced by the root system of the host plant. Maize, having a fibrous root system, supports the growth and development of AMF effectively. Maize roots

contain high levels of carbohydrates, which facilitate the growth of AMF. Root exudates that induce sugars and increase amino acid content can trigger the spores to produce flavonoid acids, which play a role in stimulating hyphal growth in AMF.

AMF colonization in plant roots influences nutrient absorption, plant tolerance to pests and diseases, drought resistance, exposure to heavy metals, and overall growth and adaptability to abiotic environmental conditions. The presence of AMF can enhance various plant growth parameters, including more efficient phosphorus and nitrogen uptake (Suharno *et al.*, 2023).

Effect of AMF on the plant height of Z. mays

emphasize the role of Arbuscular То Mycorrhizal Fungi (AMF) on the growth of several maize varieties, observations were made on key growth parameters, including plant height and leaf number of Z. mays (Figure 2). The results showed that AMF inoculation significantly enhanced plant growth across all maize varieties. Sweet corn plants inoculated with 15 g of AMF exhibited the highest weekly increase in plant height compared to other mThe most effective inoculation treatments were 10 g and 15 g of AMF, which increased plant height by up to 21.56%. Table 1 shows a significant difference in sweet corn treated with 15 g of AMF, which was 9.47 cm taller than the other treatments. During the third week after planting, although no statistically significant difference was observed, sweet corn was consistently taller, with a difference of 7.1 cm compared to the other varieties.

As shown in Figure 2, by the 8th week after planting, sweet butter corn treated with 15 g of AMF exhibited the highest increase in plant height, reaching 12.01 cm more than other treatments. Control plants (non-mycorrhizal) had the lowest plant height growth compared to those treated with AMF.

Musfal (2010) reported that root infection in maize increased with the application of AMF up to a dose of 15 g. This finding aligns with studies by Silitongan & Nasution (2020) and Farida & Chozin (2015), who observed that maize plants inoculated with AMF showed better plant height responses compared to the control group.

Nutrients play a crucial role in plant growth. One such nutrient is phosphorus. FMA decomposes the unavailable phosphorus element into a form that is available to plants. This increases plant height and allows the growth process to run optimally (Silitongan & Nasution, 2020). Extraradical hyphae increase the availability of nitrogen (N) and phosphorus (P) in plants and play an important role in nutrient absorption from the soil. Ginting et al. (2018) reported that combining FMA with 60% organic fertilizer increases plant height. This is due to the presence



Figure 2. Growth in plant height of maize (*Z. mays*) inoculated with AMF from 4 to 8 weeks after planting (WAP).

| | Table 1. | Growth res | sponse in p | plant height of | f Z. mays | inoculated [•] | with AMF | at 3 WA | Р |
|--|----------|------------|-------------|-----------------|-----------|-------------------------|----------|---------|---|
|--|----------|------------|-------------|-----------------|-----------|-------------------------|----------|---------|---|

| Treatmont | | Auorago | | | |
|-------------------------|---------|---------|----------|----------|-----------|
| Heatment | 0 | 10 | 15 | 20 | – Average |
| RM : regular corn | 5.65 c | 5.79 с | 6.36 bc | 7.14 abc | 6.23 x |
| SC : sweet corn | 7.69 bc | 6.63 bc | 9.47 a | 8.2 ab | 7.10 x |
| BSC : butter sweet corn | 5.86 c | 5.94 bc | 7.52 abc | 5.32 с | 6.14 x |
| Average | 6.40 k | 6.12 k | 7.781 | 6.87 kl | |

Notes: RM = Regular Maize, SC = Sweet Corn, BSC = Butter Sweet Corn. Means followed by the same letters in columns, rows, and treatment combinations indicate no significant difference based on Duncan's Multiple Range Test (DMRT) at a 95% confidence level.

| Table 2. Resp | onse of Z. mays | plant height | growth inoculated | with AMF a | at 8 WAP |
|---------------|-----------------|--------------|-------------------|------------|----------|
| | | | • / | | |

| | 00 | | | | |
|-------------------------|--------|---------|--------|--------|---------|
| Treatmonte | | Auorago | | | |
| Treatments | 0 | 10 | 15 | 20 | Average |
| RM : regular corn | 7.24bc | 8.42bc | 7.29bc | 8.17bc | 7.78z |
| SC : sweet corn | 7.55bc | 11.43ab | 9,30ab | 9.94ab | 9.55x |
| BSC : butter sweet corn | 6.62c | 8.76bc | 12.01a | 9,3ab | 9.17xy |
| Average | 7.13 k | 9.531 | 9.531 | 9.13 1 | |

Notes: RM = Regular Maize, SC = Sweet Corn, BSC = Butter Sweet Corn. Means followed by the same letters in columns, rows, and treatment combinations indicate no significant difference based on Duncan's Multiple Range Test (DMRT) at a 95% confidence level.

of nutrients, especially phosphorus, which the FMA makes available to corn plants.

Soil-dwelling microorganisms play an important role in decomposing, mineralizing, and recycling organic matter. These microorganisms stimulate the production of phytohormones, such as gibberellin and auxin, in plant roots. FMA can stimulate the formation of auxin hormones, which promote cell elongation and division. This triggers an increase in plant height and root development (Budi & Hardhani, 2020; Handayani *et al.*, 2023; Silitongan & Nasution, 2020).

Furthermore, according to Faizi & Purnamasari (2019), the height of sweet corn



Figure 3. Leaf number growth of maize plants (*Zea mays*) inoculated with AMF up to 8 weeks after planting (WAP).

Table 3. Response of leaf number growth in Z. mays inoculated with AMF at 3 WAP.

| Treatmont | | A 11072 CO | | | |
|-------------------------|---------|------------|---------|---------|---------|
| Heatment | 0 | 10 | 15 | 20 | Average |
| RM : regular corn | 3.44 ab | 3.77 b | 3.45 ab | 3.78 b | 3.61 x |
| SC : sweet corn | 2.89 a | 3.33 ab | 3.88 b | 3.44 ab | 3.38 x |
| BSC : butter sweet corn | 2.77 a | 2.94 a | 3.50 ab | 3.11 a | 3.08 x |
| Average | 3.03 k | 3.34 kl | 3.611 | 3.44 kl | |

Notes: RM = Regular Maize, SC = Sweet Corn, BSC = Butter Sweet Corn. Means followed by the same letters in columns, rows, and treatment combinations indicate no significant difference based on Duncan's Multiple Range Test (DMRT) at a 95% confidence level.

Table 4. Response of leaf number growth in Z. mays inoculated with AMF at 8 WAP.

| Å | | <u>v</u> | | | |
|-------------------------|---------|-------------|---------|----------|-----------|
| Treatmente | | 1 110112 00 | | | |
| Treatments | 0 | 10 | 15 | 20 | — Average |
| RM : regular corn | 3.66 ab | 5.26 abc | 3.20 a | 4,32 abc | 4.11 x |
| SC : sweet corn | 3.12 a | 6.06 bc | 6.52 bc | 5,60 abc | 5.32 x |
| BSC : butter sweet corn | 3,14 a | 6.26 bc | 7.34 c | 6,94 bc | 5.92 x |
| Average | 4.96 k | 5.861 | 5.681 | 5.621 | |

Notes: RM = Regular Maize, SC = Sweet Corn, BSC = Butter Sweet Corn. Means followed by the same letters in columns, rows, and treatment combinations indicate no significant difference based on Duncan's Multiple Range Test (DMRT) at a 95% confidence level.

plants treated with AMF showed a significant difference compared to the control. Suharno et al. (2024) also reported that AMF application to *Andrographis paniculata* increased plant height. This condition highlights the critical role of AMF in enhancing plant growth.

AMF can have a positive impact on plant growth by forming symbiotic relationships with host plant roots to facilitate nutrient uptake, particularly phosphorus (Suharno et al., 2024). The more AMF propagules are applied, the more nutrients can be absorbed and efficiently utilized by the plant (Faizi & Purnamasari, 2019). However, excessively high inoculation rates of AMF under certain conditions may reduce the optimization of plant growth (Suharno *et al.*, 2021).

Budi & Hardhani (2020) stated that the height growth of African mahogany seedlings is influenced not only by mutualistic symbiosis between AMF and the plant, but also by nutrient content derived from rice husk charcoal in organic potting media, which supports better seedling development. In this study, the three maize varieties were grown in pure soil pot cultures without the addition of organic materials or other nutrient sources. Saputra et al. (2019) explained that nutrient availability in tropical soils is generally insufficient to meet plant growth and productivity needs, thus requiring additional fertilization as a nutrient source.

Maize plants at 8 weeks of age are in the vegetative growth stage, where the most critical nutrients are nitrogen (N), phosphorus (P), and potassium (K), which stimulate plant height and stem diameter. During this phase, a large amount of nitrogen is required. Nutrient uptake by plants is influenced by the availability of nutrients in the soil. If the nutrient availability is equal across treatments, then the plant response will generally be similar (Amrizal *et al.*, 2021).

Response of AMF colonization on the leaf number of *Z. mays*

Observations revealed that the leaf number of butter sweet corn peaked in the seventh week, followed by a decline in the eighth week (Figure 3; Table 3; Table 4). This may have been influenced by environmental factors or nutrient deficiency. According to Budi & Hardhani (2020), macronutrients play an essential role in plant growth. Phosphorus deficiency can cause the lower leaves to yellow, dry out, or even turn greenish-brown to black. Meanwhile, nitrogen deficiency may stunt plant growth, resulting in dwarf plants with pale-colored leaves.

Inoculation with AMF significantly increased leaf number growth overall but not across maize varieties during the early growth stages (up to the third and sixth weeks). However, the interaction between AMF dosage and maize variety showed statistically significant differences (Table 3; Table 4). In the eighth week, butter sweet corn produced the highest number of leaves compared to other maize varieties.

Application of AMF at doses of 10, 15, and 20 grams per polybag significantly increased the number of leaves by up to 18.14%, with the highest increase observed at the 10 g treatment. This result suggests that although the increase was relatively small, it was statistically significant compared to the non-mycorrhizal control (Table 4). While the main effect of maize variety on leaf number was not statistically significant, butter sweet corn consistently showed higher leaf growth than the other varieties.

These findings contrast with those of Sufaati & Aryuni (2009), who reported that AMF and litter application at certain doses did not significantly improve the height or leaf number in shallot plants. Similarly, Faizi & Purnamasari (2019) also found no significant increase in leaf number. This condition is likely due to the stronger influence of genetic factors; leaf number is closely linked to plant genotype.

Sweet corn and butter sweet corn plants without AMF application (0 g) produced fewer leaves compared to those treated with AMF. Leaves are crucial for plants as the primary sites of photosynthesis, generating carbohydrates essential for plant growth and development. An increased number of leaves is indirectly associated with greater light capture for photosynthesis. The differences in leaf number across treatments may also be attributed to the variation in AMF colonization in the growth medium. Additionally, leaf area contributes to enhanced photosynthetic capacity; the broader the leaf surface, the greater the photosynthate production, which positively affects the plant growth rate (Sufaati & Aryuni, 2009; Fitria *et al.*, 2022; Suharno *et al.*, 2024).

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Relative growth rate (RGR) of maize (Z. mays)

The results of the study indicated that butter sweet corn exhibited better growth performance compared to other maize types. The relative growth rate (RGR) for this variety reached 0.43,

| Table 5. Relative growth rate o | f Z. <i>mays</i> plants inocu | lated with arbuscular m | vcorrhizal fungi (AMF) |
|---------------------------------|-------------------------------|-------------------------|------------------------|
| () | | | |

| Treatment | | Inoculum (g) | | | | |
|-------------------------|--------|--------------|----------|----------|---------|--|
| ireatment | 0 | 10 | 15 | 20 | - | |
| RM : regular corn | 0.02 a | 0.20 ab | 0.06 a | 0.27 abc | 0.13 k | |
| SC : sweet corn | 0.01 a | 0.01 a | 0.26 abc | 0.90 c | 0.29 kl | |
| BSC : butter sweet corn | 0.10 a | 0.43 bc | 0.52 bc | 0.68 bc | 0.431 | |
| Average | 0.04 x | 0.21 xy | 0.28 xy | 0.61 y | | |

Notes: The same letters in the mean values of columns, rows, and treatment combinations indicate no significant difference according to Duncan's Multiple Range Test (DMRT) at a 95% confidence level.

the highest among all observed types. Treatment with 20 g of AMF inoculum showed the highest RGR of 0.61, surpassing the growth rates of the other varieties (Table 5).

The findings further demonstrated that both butter sweet corn and sweet corn had higher RGR values. These two maize types are hybrid cultivars, which tend to exhibit enhanced growth compared to conventional maize.

AMF inoculation also had a significant impact on RGR. The highest relative growth rate was recorded in the treatment with 20 g of AMF per polybag, reaching 1.425%, followed by the 15 g treatment, which achieved 600%. This underscores the critical role of arbuscular mycorrhizal fungi (AMF) in improving plant growth performance.

Relative growth rate is closely associated with the overall increase in plant biomass. As shown in Table 5, the RGR of all three maize varieties treated with AMF was higher than that of untreated plants. This result is attributed to the symbiotic relationship where AMF utilizes carbon compounds from photosynthetic fixation and enhances the uptake of essential nutrients, namely 80% of nitrogen (N) and 100% of phosphorus (P). AMF is known to form symbiotic relationships with approximately 78% of terrestrial plants (Amrizal *et al.*, 2021).

AMF has a positive influence on both dry and fresh plant biomass. A higher dry weight is indicative of better plant growth. Dry weight reflects the accumulation of organic compounds synthesized by plant from the inorganic substances, including nutrients, water, and carbohydrates (Budi & Hardhani, 2020). Suharno et al. (2021) reported that AMF significantly increased the growth of *Pogostemon cablin* in sandy soil. Therefore, the application of AMF can enhance overall biomass production, resulting in optimal plant growth and improved yield quality.

CONCLUSION

The results of this study show that arbuscular mcorrhizal fungi (AMF) inoculation significantly enhances the growth performance of maize (*Zea* *mays*), increasing plant height by 21.56%, leaf count by 18.14%, and showing a remarkable improvement in relative growth rate (RGR) of up to 1,425% at 6 weeks after planting. Among the tested varieties, butter sweet corn and sweet corn responded more positively to AMF inoculation compared to regular maize. The best growth response was observed in butter sweet corn treated with 15 g of AMF inoculum per polybag.

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