Compatibility and effectivity of various mycorrhizal sources with cassava varieties in Gunungkidul

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Abstract. This study aims at understanding the interaction between various mycorrhizal sources and three local cassava varieties of Gunungkidul, specifically on their compatibility, population, mycorrhizal diversity, and their effect on cassava growth. The experiments were conducted based on Completely Randomised Design (CRD) with experimental factors using three treatments in triplicates. The experimental factors included three cassava varieties and various sources of mycorrhiza. The results demonstrated that among the interactions of various mycorrhizas with three local cassava varieties of Gunungkidul, the highest infection was seen from Mentega cultivar inoculated with indigenous mycorrhiza of the Alfisol soil of Gunungkidul (95%). Mycorrhizal diversity using indigenous mycorrhiza of Alfisol soil was higher than mycorrhiza of pandan rhizosphere and commercial inoculant, and the dominant mycorrhiza type was of spores belonging to genus Glomus. On the other hand, the indigenous mycorrhiza of Alfisol soil inoculant on various cassava varieties demonstrated similar effect and yielded the same number of spores, root length, and plant dry weight; despite the fact that on Kirik cultivar growth, the significant difference was observed on the plant height, leaf number and fresh net weight.

1. Introduction
Cassava is a local staple food in Gunungkidul area. However, cassava yield remains low compared to other plantation areas that have the potential to reach 300-400 kw/Ha. According to the data on Plant Food Sources collected from Gunungkidul area, the total cassava yield only reaches 844,773.26 tons with a productivity rate of only 155.05 kw/Ha despite the widespread cassava plantation areas (54,485 hectares) [1]. One of the leading factors of this low productivity is the naturally dry lands in Gunungkidul that only rely to rainfall as their water source. In Playen, Wonoasari, Karangmojo, Ponjong tengah and Semanu, the most ubiquitous soil type is the red-yellow Alfisol soil formed by decayed limestone with low concentration of organic matter, dry texture, and soil acidity ranging from slightly acidic to slightly basic [2].

Mycorrhiza can improve soil condition and increase plant growth due to the mutualistic symbiosis between fungi and the plant’s root system. According to Hajoeningtijas and Purnawanto [3], the use of mycorrhiza as biofertilizer induces a positive response on the growth and yield of cassava crops. Mycorrhiza can be utilized as a means to improve yield in marginal lands [4]. Arbuscular mycorrhiza is a type of endomycorrhiza that functions in the soil as an obligate symbiont by infecting plant roots. This improves the plant’s ability to absorb nutrients, enhances the soil conditions, increases the plant’s resilience against drought and diseases, maintains the opening of stomata and transpiration, improves roots system and supports plant growth. The infection process starts with the sprouting of
the spore in the soil. The hypha will penetrate the roots and develop in the cortex to form arbuscules, vesicles, as well as internal and external hyphae [5]. Mycorrhiza infects the roots of Dicotyledoneae plants (83%), Leguminosae and Monocotyledoneae plants (79%), and Gramineae [6]. However, mycorrhiza growth depends on its inoculant source and other environmental factors such as temperature, water content, soil pH, the availability of organic matter, light, nutrients, as well as the concentration of heavy metal and fungicides in the soil [7, 8]. A study conducted by Sarjiyah et al. [9] demonstrates the presence of more than 30 local cassava varieties in Gunungkidul that have the potential for future development including Mentega, Kirik and Ketan varieties. Therefore, future studies are needed to determine the compatibility of several mycorrhiza sources with the local cassava varieties of Gunungkidul as well as their adaptability and effectiveness during the propagation phase. We hypothesize that mycorrhiza isolates from the indigenous Alfisol soil of Gunungkidul would be compatible with the three local cassava varieties and display a robust propagation phase. In order to maximize the use of mycorrhiza as biofertilizer, we need to assess the type of mycorrhiza that can adapt and associate with a particular plant [6]. Some common Mycorrhiza genus include Glomus, Gigaspora, Acaulospora and Scutellospora [10]. Each Mycorrhiza yields a different response in improving plant yield [11]. Therefore, it is necessary to find a mycorrhiza isolate that is compatible with the plant [12].

Cassava demonstrates high dependability on mycorrhiza. The cassava cultivar influences the degree of this dependability. Aryaji [13] shows a strong association between cassava cultivar and the mycorrhiza type. Eight cassava varieties display a high degree (61.3-80%) of root colonization by mycorrhiza: Glomus sp., Scutellospora sp., Paraglomus sp., Gigaspora sp., Diversispora sp., and Acaulospora sp. Application of mycorrhiza biofertilizer affects cassava growth positively. Mycorrhizal inoculation is proven to increase cassava yield by increasing water and nutrient absorption through the external hyphae and improving plant metabolism [14]. Several farming groups in the Karangsari village, Kembaran subdistrict, Banyumas regency have applied mycorrhizal biofertilizer on the cassava plants [15]. Sieverding and Toro [16] shows that cassava yield declines without mycorrhiza inoculation. Cassava plan grown in mycorrhiza-sterilized soil lacks of phosphor and displays stunted growth. Hajoentingtijas and Purnawanto [3] shows that mycorrhizal biofertilizer increases the growth and yield of the cassava plants.

This study aimed to identify several mycorrhiza inoculants in the alfisol soil of Gunungkidul and assess the compatibility of these inoculants with the three local cassava varieties, as well as their effectiveness on cassava yield. The long-term goal of this study is to identify mycorrhiza inoculants that can improve cassava yield.

2. Materials and Methods

2.1. Research Period and Location
This study was conducted in March - August of 2018 at the Agrobiotechnology Laboratory and the Green House of the Faculty of Agriculture, Universitas Muhammadiyah Yogyakarta.

2.2. Materials of Research
Mycorrhiza collected from : Indigenous Alfisol rhizosphere of cassava from Gunungkidul regency, the rhizosphere of pandan plant from Bugel beach and commercial Bogor mycorrhizal inoculant. Propagation of a cultivar of mycorrhizal sources on the host corn crops using the trapping method. The cassava varieties indigenous from Gunungkidul: Mentega, Kirik and Ketan cultivars.

2.3. Research Methodology
The experiment was conducted in a greenhouse of the Agriculture Faculty UMY Yogyakarta Indonesia during 12 weeks on the Alfisol soil from Gunungkidul. The experimental design was a factorial (3x3) in Completely Randomised Design (CRD) principle using three treatments in triplicates. The first factor is the mycorrhizal source, delineated as A: indigenous Alfisol soil collected from Gunungkidul; B = rhizosphere of pandan plant collected from Bugel beach; C = commercial mycorrhiza inoculant. The second factor is the cassava cultivar, delineated as P= Mentega; Q= Kirik;
R= Keta. In total, 9 treatment combinations were conducted in triplicates to yield 27 units. Each unit consists of 3 samples, yielding a total of 81 plants.

Each pot received 4 kg of dry soil and contain 1 plant of Cassava and inoculated with 40 g crude inoculum of indigenous mycorrhiza Gunungkidul according to the assigned treatment. Plants were carefully watered as needed to maintain soil moisture near field capacity. After 12 weeks of growth, plants were cut at ground level separated from roots. Several experimental parameters were observed, including the percentage of mycorrhizal root colonization, the number of spores, root length, plant height, dry net weight, fresh net weight, and the number of leaves. Counting of spora number of mycorrhiza 100 g dry soil was using wet sieving and decanting technique [17]. Mycorrhizal root colonization was determined according to Kormanik and McGraw [18].

The analysis of variance on number of spora, mycorrhizal root colonization, root length, plant height, dry net weight, fresh net weight and number of leaves were done using procedure of SAS. Significant differences among of treatments were tested using Duncan's Multiple Range Test.

3. Results and Discussions

3.1. Compatibility of several mycorrhizal sources with an array of cassava varieties

The degree of compatibility between mycorrhiza and their host plants varies depending on the mycorrhiza species, the host plant species, and their environmental conditions. This compatibility is measured by the degree of mycorrhizal colonization on the plant roots and the portion of mycorrhiza-infected plant roots [19, 20]. Several factors may influence this compatibility, including mycorrhiza species, type of host plant, microbial interactions, host plant’s root architecture, competition among several mycorrhizal, and soil conditions [21].

This study shows that the same mycorrhizal source can yield various degrees of mycorrhizal infection on different cassava varieties. A mycorrhizal source that yields a higher degree of infection on a particular cassava cultivar is therefore more compatible with this plant than another mycorrhizal source that yields a lower degree of infection. The degrees of mycorrhizal infection on several cassava varieties are shown on Table 1.

| Mycorrhizal sources                  | Cultivars | Average of sources |
|-------------------------------------|-----------|--------------------|
|                                     | Mentega   | Kirik              | Ketan               |
| Indigenous alfisol, Gunungkidul     | 95.00a    | 55.00c             | 53.33c              |
| Pandan rhizosphere, Bugel Beach     | 75.00b    | 50.00c             | 50.00               |
| Commercial mycorrhizal inoculant    | 53.33c    | 60.00cb            | 45.00               |
| **Average of cultivars**            | 71.43     | 55.71              | 50.00 (+)           |

Different letters denote statistically significant results according to analysis of variance and Duncan’s multiple range test, with $\alpha=5\%$. (+) denotes no significant association.

Table 1 demonstrates a significant association between mycorrhizal source and cassava cultivar. We observed that the mycorrhizae collected from the indigenous Alfisol soil of Gunungkidul had the highest compatibility with the Mentega cassava cultivar as shown by the highest degree of mycorrhizal infection (95%). This finding is strongly supported by Aryaji [13], who observes a strong association between cassava cultivar and mycorrhizal source as exemplified by the high degree of root colonization (61.3-80.3%) on eight cassava varieties. This proves that a degree of compatibility between cassava cultivar and the mycorrhizal source is necessary to form root colonization through the hyphae. Hyphae penetrance leads to an increase in respiration, cytoplasmic size, and enzymatic activity of the host plant cells [22].

On the other hand, both mycorrhizas collected from the pandan rhizosphere in Bugel Beach and the commercial inoculants ones demonstrated low root colonization (<60%) on the Kirik and Ketan cassava varieties. The pandan rhizosphere mycorrhizae demonstrated a moderately high degree of
colonization on the *Mentega* cassava cultivar (75%). Different plant varieties will yield different interactions with a mycorrhizal source, which in turn influences the degree of mycorrhizal infection and colonization. This is highly dependent on the plant’s response to mycorrhizal infection and the dependency between the host plant and the mycorrhiza. These two factors are associated with the type of the host plant’s root architecture and its physiological conditions [19].

In conclusion, the compatibility of a mycorrhizal source with a host plant is highly variable and dependent on the mycorrhizal species and the plant cultivar, as shown by the degree of mycorrhizal infection. In addition, mycorrhizal infection is highly influenced by the host plant’s particular features and environmental factors such as temperature, humidity, soil pH, and light intensity [23].

3.2. Effectivity of several mycorrhizal sources on an array of cassava varieties

In order to determine the effectivity of a mycorrhizal source on plant growth, we evaluated the types of organelles that were formed on the host plant’s cortex. Root colonization is an easily observable feature to evaluate the effect of mycorrhizal inoculation on plant growth. A successful mycorrhizal infection leads to the formation of one of the following structures: vesicles, arbuscules, and external hyphae [24].

The seeds of the *Mentega*, *Kirik*, and *Ketan* cassava varieties were inoculated with mycorrhiza collected from the indigenous alfisol soil of Gunungkidul, the pandan rhizosphere of Bugel Beach, and a commercial mycorrhiza inoculant. Mycorrhizal colonization on the plant roots reached completion (100%) after 4 weeks; as shown by the formation of vesicles, internal hyphae, and arbuscules (Table 2). Arbuscules are hyphae formed by repeated dicothomic branching that resemble the symbiont’s branch structure [22].

| Mycorrhizal sources                  | MVA Characteristics (weeks) | Degree of infection (%) |
|--------------------------------------|-----------------------------|-------------------------|
|                                      | Internal hyphae | Arbuscules | Vesicles | External hyphae |                             |
|                                      | 4   | 12             | 4   | 12         | 4   | 12             |                             |
| Indigenous alfisol, Gunungkidul      | 2+   | 5+             | 2+   | 3+         | -   | 4+             | 4+                          | 100                          |
| Pandan rhizosphere, Bugel Beach      | 2+   | 4+             | 2+   | 3+         | -   | 2+             | 4+                          | 100                          |
| Commercial mycorrhizal inoculant     | 2+   | 4+             | 2+   | 3+         | -   | 2+             | 4+                          | 100                          |

The mycorrhizae collected from the indigenous alfisol soil in Gunungkidul demonstrated the highest activity compared to the other two mycorrhizal sources. This is demonstrated by the highest degree of infection (100%) and the higher formation of internal hyphae, external hyphae, and arbuscules on the colonized root.

Resti et al. [23] shows that indigenous mycorrhizae plays a pivotal role in nutrient acquisition to support plant growth. In the more advanced stage of plant development, the mycorrhizae continues to grow and forms more vesicles and external hyphae. The increasing number of vesicles is resulted from the terminal growth of the internal hyphae. They form fat-containing round structures and support nutrient storage for survival as well as spore formation in certain conditions [25]. At the tip of the external hyphae, mycorrhiza can form both free spores and sporocarps. The increased number and length of the external hyphae support the plant’s root architecture and plant growth.

Analysis of variance on the number of mycorrhizal spores showed no significant association between mycorrhizal source and cassava plant cultivar (Table 3). There was no significant difference across all treatments. The highest number of mycorrhizal spores was found in the rhizosphere of the *Kirik* cassava plants inoculated with the pandan rhizosphere from Bugel Beach (98 spores/100g). The lowest number of spores was found in the rhizosphere of the *Ketan* cassava plants inoculated with the indigenous alfisol soil from Gunungkidul (39 spores/100g). The average number of spores yielded by
the sum of the mycorrhiza treatments and the cassava varieties was 54.8 spores/100g and 53.63 spores/100g respectively. This is still lower than the acceptable standard of >60 spores/100 g [26].

### Table 3. Average number of spores in the cassava growing media (spores/100g).

| Mycorrhizal sources                  | Cultivars | Average of sources |
|-------------------------------------|-----------|--------------------|
|                                     | Mentega   | Kirik              | Ketan |
| Indigenous alfisol, Gunungkidul     | 39.0      | 56.0               | 51.3  |
| Pandan rhizosphere, Bugel Beach     | 45.5      | 98.0               | 49.0  |
| Commercial mycorrhizal inoculant    | 52.3      | 50.6               | 46.0  |
| **Average of cultivars**            | 45.1p     | 65.7p              | 50.1p |

Different letters denote statistically significant results according to analysis of variance and Duncan’s multiple range test, with α=5%. (-) denotes no significant association.

Spore production increases as the host plant matures. The number of spores does not directly demonstrate the number of formed root colonies. Spore formation is a dynamic process, as spore formation and sprouting can occur concurrently. The number of spores is an indicator of mycorrhizal growth and development in the soil that depend on the host plant’s metabolism [19].

We isolated spores from vesicles of arbuscular mycorrhiza collected from the Tanjungsari Village, Ponjong Subdistrict, Gunungkidul Regency. We identified these spores as members of the Glomus, Gigaspora, and Acaulospora genera. Their features and characteristics are shown in Table 4.

### Table 4. Types of mycorrhiza isolated from cassava growing media after 8 weeks

| Mycorrhizal sources                  | Cultivars |平均 | Ketan |
|-------------------------------------|-----------|-----|-------|
|                                     | Mentega   |     |       |
| Indigenous alfisol, Gunungkidul     | Glomus sp.| Glomus sp.| Glomus sp. |
| Pandan rhizosphere, Bugel Beach     | Acaulospora sp.| Gigaspora sp.| Glomus sp. |
| Commercial mycorrhizal inoculant    | Glomus sp.| Glomus sp.| Glomus sp. |
|                                     | Acaulospora sp.| Glomus sp.| Glomus sp. |

The spores collected from the above mycorrhizal sources were produced by *Glosmus* sp., Gigaspora sp., and *Acaulospora* sp. This is in agreement with the study conducted by Widiatma *et al.* [27] which identifies three mycorrhiza genera in the cassava rhizosphere, namely the *Glomus* sp., Gigaspora sp., *Acaulospora* sp. Aryaji [13] identifies 6 mycorrhiza genera in the rhizosphere of 8 cassava varieties, namely *Glomus* sp., *Gigaspora* sp., *Acaulospora* sp. Scutellospora sp., Paraglomus sp. and Diversispora sp. The *Acaulospora* and *Glomus* mycorrhizae have previously been isolated from the red onion rhizosphere. Likewise, the *Acaulospora* and *Glomus* mycorrhiza were isolated from the taro rhizosphere. However, the *Scutellospora* mycorrhizae were not isolated. A possible explanation is that the compatibility of a mycorrhiza species on the host plant is highly variable and dependent on the mycorrhizal species, host plant species, and other environmental factors [20].

The degree of compatibility between the mycorrhizas and their host plant is highly dependent on the degree of mycorrhizal infection on the plant roots [23]. Although mycorrhizas might not have an affinity for a particular symbiont, their ability to infect and colonize plant roots may differ from one another. This is thought to be the product of mycorrhizal adaptability to the soil conditions, the abundance of propagules, the physiological properties of the propagules, and the development of the fungi in the soil after root infection [20].

### 3.3. Growth Evaluation of An Array of Cassava Varieties

Analysis of variance on several plant growth parameters showed no significant association between...
mycorrhizal source and cassava plant cultivar (Table 5). There was no significant difference in root length (average: 18.13 cm) and dry net weight (average: 21.71 g). However, we observed significant differences in plant height, fresh net weight, and number of leaves. The inoculation of the Ketan cultivar with mycorrhiza from the indigenous Alfisol soil from Gunungkidul yielded the tallest plant (52.18 cm), followed by the Ketan cultivar (59.98 cm). Both the highest fresh net weight (110.8 g) and the highest number of leaves (89.11 units) were found on the Kirik cultivar.

### Table 5. Analysis of variance on cassava growth parameters

| Treatments                        | Root length (cm) | Plant Height (cm) | Dry net weight (g) | Fresh net weight (g) | Number of leaves (unit) |
|-----------------------------------|------------------|-------------------|-------------------|----------------------|------------------------|
| Mycorrhizal sources:              |                  |                   |                   |                      |                        |
| Indigenous alfisol, Gunungkidul   | 18.55a           | 52.18a            | 93.11a            | 22.88a               | 75.22ab                |
| Pandan rhizosphere, Bugel Beach   | 16.20a           | 48.98a            | 88.56a            | 19.78a               | 83.88a                 |
| Commercial mycorrhizal inoculant  | 19.47a           | 48.31a            | 87.31a            | 22.15a               | 67.55b                 |
| Cultivars:                        |                  |                   |                   |                      |                        |
| Mentega                           | 20.43p           | 44.22q            | 70.91q            | 17.69p               | 66.22q                 |
| Kirik                             | 18.52p           | 45.78q            | 110.80p           | 25.61p               | 89.11p                 |
| Ketan                             | 15.66p           | 59.98p            | 87.27p            | 22.19p               | 71.33q                 |
| Association                       | (-)              | (-)               | (-)               | (-)                  | (-)                    |

Different letters denote statistically significant results according to analysis of variance and Duncan’s multiple range test, with α=5%. (-) shows no significant association.

This finding supports the study by Brundrett et al. [10] which states that mycorrhizal inoculation stimulates root branching and increases proliferation. Despite the relatively equal root lengths across all treatments, root proliferation was highest on the cassava varieties inoculated with the mycorrhiza from the indigenous Alfisol soil from Gunungkidul and the pandan rhizosphere from Bugel Beach compared to the commercial inoculant ones. Improved nutrient acquisition leads to higher plant height (52.18 cm) and number of leaves (83.88 units). This supports the notion that mycorrhiza-inoculated plants demonstrate improved growth because of increased nutrient acquisition despite the lack of accessibility to the plant roots [28].

Kirik cassava cultivar is indigenous to Gunungkidul. Therefore, it is not surprising that it demonstrated a high adaptability to its environment, as shown by the high number of spores (98 spores/100g). Consequently, this cassava cultivar also demonstrated the highest growth as shown by the high fresh net weight (110.80 g) and number of leaves (89.11 unit) compared to the other varieties. The fresh net weight is a good indicator of plant metabolism, which is influenced by water content, nutrient acquisition, and metabolic outcomes. The leaves play an important role in processing nutrients, transpiration, and plant respiration. Therefore, a high number of leaves also leads to improved plant growth. Mycorrhizal inoculation from a cultivar of sources induced a positive response on the growth of several cassava varieties. The use of mycorrhiza as biofertilizer was proven to show good compatibility and effectivity on cassava growth.

### 4. Conclusions

The compatibility of various mycorrhizal sources and three local cassava varieties of Gunungkidul were tested. The most compatible pair, as shown by the highest degree of infection (95%), was the Mentega cassava cultivar inoculated with mycorrhizae from the indigenous Alfisol soil of Gunungkidul. Among the diverse mycorrhizal sources, the most effective source was the indigenous
Alfisol soil; as shown by the higher formation of internal hyphae, external hyphae, vesicles, and arbuscules by the mycorrhiza. The mycorrhizal population reached the value of 98 spores/100g and the dominant spores belonged to the Glomus genus. The indigenous mycorrhiza of Alfisol soil inoculant on various cassava varieties demonstrated similar effect as it resulted in the same number of spores, root length, and plant dry weight; despite the fact that on Kirik cultivar growth, the significant difference was observed on the plant height, leaf number and fresh net weight.

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