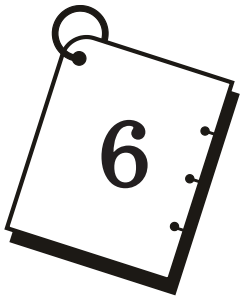

SPORE PRODUCTION OF ARBUSCULA MIKORIZA FUNGI GLOMUS SP. AS AFFECTED BY ENDOSIMBIOTIC BACTERIA AND HOST SPECIES

By

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ABSTRACT

Five bacteria isolats (B1, B2, B3, B4 dan B5) were isolated from Glomus sp spore and tested for their capability to increase Arbuscular Mycorrhizal fungi (AMF) spore production in the green house condition. There were two host plants Sorghum bicolor and Pururera javanica were used in this experiment. Growth medium used was pure zeolit. Based on the analysis of variance showed that the endosimbiotic bacteria significantly increase AMF spore production in P. javanica host plant only. The bacteria isolat B4 give significantly increase AMF spore production on P. javanica host plant with amount of spore number 1.115 spore/30 g media.

Key words: Spore production, Arbuscular mycorrhizal fungi, host plant, endosimbiotic bacteria

Introduction

Previous study demonstrated that Arbuscular mycorrhizal Fungi (AMF) could improved crop productivity in Agroforestry System in Gunung Walat Educational Forest (Budi, 2007). It is necessary to produce an inoculum of this fungus in large scale for its application in the field.

Arbuscular mycorrhizal fungi (AMF) cannot be grown in pure culture and consequently the techniques used for inoculum production are different from those generally employed for other biotechnologically intersesting fungi (Gianinazzi and gianinazzi-Pearson, 1990). Several methode have been developed to produce inoculum production such as nutrient film technique (Mosse, Thompson and Smith, 1980), axenic culture using disinfected spore on excised roots or whole plants (Williams, 1992) and

open pot culture (Gianinazzi, Gianinazzi-Pearson, and Trouvelot, 1990). The fisrt two methode for the moment is not practically viable for mass production.

Mycorrhizal development and sporulation are in general highly dependent on various biotict and abiotic factors, such as light intensity, temperature, fertilization, substrat and the host genome (Powell and Bagyaraj, 1984). In addition, bacteria are known to have beneficial (Bagyaraj, 1984), or detrimental effect on mycorrhizal development (Garbaye, Dupnnois and Wahl (1990).

The objective of this experiment was to investigate the effect of bacteria isolate and host plant on spore production and mycorrhizal development.

Materials and methods

Preparation of Host plant

Sorgum (*Sorghum bicolor*) and Kudzu (*Puraria javanica)* were used as a host plant of Arbuscular mycorrzizal fungi. The sorghum and Kudzu seed was soaked in water for about 4 h and then surface sterilized by shaking in 10 % NaCLO solution for 5 min then rinsed with sterile distillated water. The seed were shown in a plastic flat containing sterilized zeolite and grown under the greenhouse and allowed to germinate for 7 days.

Preparation of Arbuscular mycorrhizal spores

Spore of AM fungi Glomus sp were isolated from pot culture by wet sieving and decanting methods (Nicolson and Gardeman, 1963). The spores were than pick up under the compound microscope and stored in the refrigerator until used. The spores were originally isolated from mineral soil of Gunung Walat Educational Forest, Sukabumi by trap culture.

Preparation of Bacteria

The bacteria were isolated from spore of Glomus sp, according the methode of Budi (1999). Pure bacteria isolate then grown in Nutrient Broth medium for 24 h. The bacteria culture was centrifuge for 5 min, the supernatant through away and the pellet was collected for inoculation. Each plant was inoculated by 10 ml bacteria of 10^9 cfu/ml.

Inoculation and Maintenance

Two weeks old of *P. javanica* and *S. bicolor* were transplanted to plastic pot containing 250 ml zeolit and inoculated by Arbuscular mycorrhizal fungi and bacteria isolate. The plants were fertilized once per week by 5 ml red Hyponex® with concentration of 3 g/l.

Statistical analysis

Values obtained were checked by analysis of variance (ANOVA). Duncan Multiple Test was applied for multiple comparisons.

Results and Discussion

Effect on Spore Production

As shown in Figure 1, the treatments of bacteria isolate have different response to spore production. All of bacteria isolate tested have tendency to inhibit spore production when applied with *S. bicolor* as a host plant, in the contrary when applied with *P. javanica* as a host plant, there were three pattern response ; neutral for bacteria isolate B1, inhibition for bacteria isolate B2, B3 and B5 and stimulation for Bacteria isolate B4.

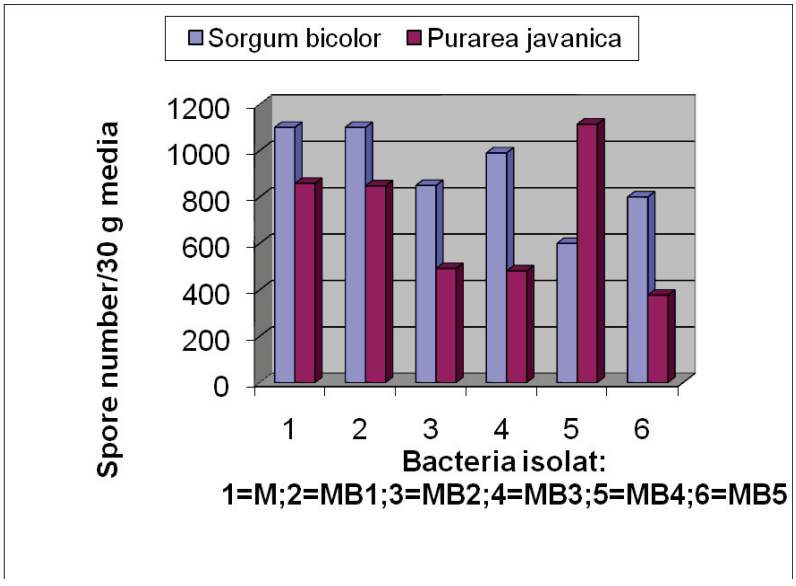

Figure 1 Spore number as affected by bacteria isolat in two different host species

Based on this results, seemly that the stimulation effect on spore production by bacteria highly dependent on host plant. This finding is an

accordance with research by von Alten, Lindermann and Schonbeck (1993) who demonstrate that the stimulation of spore production by *Bacillus mycoides*, increased by 144 % only when applied with barley as a host plant whereas when applied with Flax as a host plant the spore production increased by 209 %.

The affinity of host plants to AMF will determine the degree of sporulation which accurs (Hetrick, 1984). Schenck and Kinloch (1980) observed that the incidence of AMF species (determined as spore number in media) depend on plant species which was colonized. A woody site was newly planted with 6 agronomic crops and grown in monocultur for 7 yerars, spore number of Gigaspora spp. were more nummerous around soybean plants, whreas Glomus and Acaulospora spp. predominated around monocotil crops. It appears that the host plant can affect sporulation.

Effect on mycorrhizal development

Roots colonization by AMF are influenced by a wide range of environmental, host and fungal effect (Hetrick, 1984). Figure 2 shows that almost all of bacteria isolates have tendency to stimulate root colonization in both host plant species except bacteria isolate B5 inhibit root colonization of *S. bicolor.*

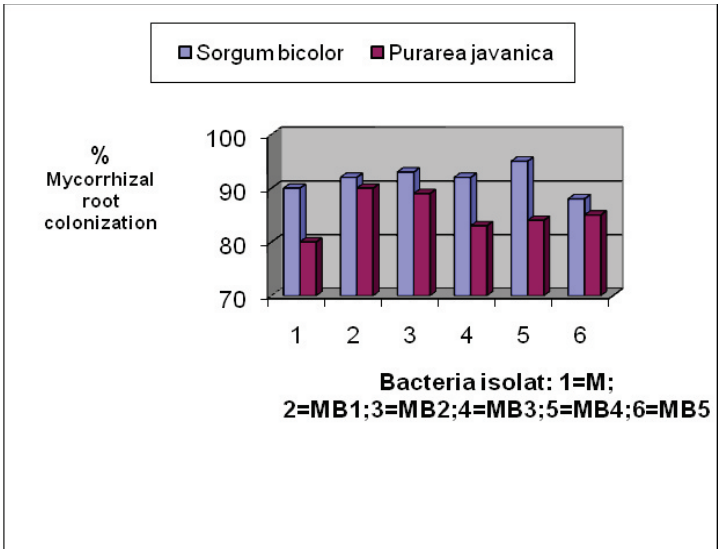

Figure 2 Roots colonization as affected by bacteria isolat in two different host species

Garbaye, Dupnnois and Wahl (1990) demonstrate that, only three bacterial isolates of 47 isolated from *Laccaria laccata* ectomycorrhizas or sporocarps significantly increased mycorrhizal development on Douglast fir seedlings. According to Puppi, Azcon and Hoflich (1994) bacteria isolates that could be beneficial to plants and their activities sinergistically interacting with Arbuscular mycorrhizal fungi can be used as inoculants.

As shown in Figure 2, the effect of bacteria isolates on mycorrhizal roots colonization were varie with host plant. This was in agrrement with the research of by von Alten, Lindermann and Schonbeck (1993) who demonstrated the different degree mycorrhizal root colonization when inoculated by *Bacillus mycoides.*

Although AMF have extremly wide host range (Gianinazzi, Gianinazzi-Pearson, and Trouvelot, 1986), the existence of host preference has been suggested by many researcher. This preferential association between certain plant and fungal species can be evaluated with respect to combinations which provide the greatest plant growth stimulation and the greatest root colonization or maximum sprorulation, but these three factors are not necessarily correlated (Mosse, 1975). Therefore as shown in Figure 1 and 2, there is no correlation between roots colonization and spore production.

Conclusions

Based on research results, it concluded that: (1) Bacteria isolate B4 give significantly increase AMF spore production on *P. javanica* host plant with amount of spore number 1.115 spore/30 g media. (2), All bacteria isolates tested stimulate roots colonization on *S. bicolor* and *P. javanica,* except bacteria isolate B5 inhibit roots colonization on **S. bicolor.**

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