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# The effect of organic fertilizer types and Endophyte bacteria application on the productivity of hot pepper (*Capsicum ascalonicum*)

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**Abstract.** This study was aimed to examine the interaction between the types of manure which used as basic fertilizer and the intervals of giving endophyte bacteria consortium to support hot pepper plant growth. The study was designed as completely randomized design in factorial experiment. The first factor is types of manure (P), while the second factor is the interval of endophyte bacteria consortium (N) application. Parameters observed were the growth of hot pepper namely plant height and number of leaves, and harvest quantity namely fruit number dan fruit resulted. As the result, there is interaction between type of manure treatment with the intervals of giving the endophyte bacteria. The application of endophyte bacteria able to increase the efficiency of manure given. The most recommended combination to produce best quality of harvested hot pepper is cow manure and intervals of 5 days of endophyte bacteria. Using this combination, the productivity of hot pepper could be reached to 19.5 tons per ha.

## 1. Introduction

In Indonesia, the total of hot pepper consumption per capita was reached at 3.05 kg in 2019 [1]. Java itself contribute 51.22% of total hot pepper production [2]. The increase price of hot pepper has been significantly affecting the inflation rate. Price fluctuation is almost found in every year; thus, the stability of hot pepper productivity is needed. To overcome that problem, government try to increase planting area and to spread culture technology that support sustainability and productivity.

Agricultural techniques using local components in the aim to increase production must be supported and applied at the farm level [3]. One of the local components is to utilize endophyte bacterial groups. In the soil, it is common to find certain bacterial populations, which promote rhizo-bacterial growth, promote plant growth and or reduce the incidence of soil diseases [3].

Endophyte bacteria promote plant growth in direct and indirect ways. The direct effect is to increase plant growth through various mechanisms. The mechanisms are including fixation of free nitrogen transferred into plants, production of siderophores that make iron (Fe) available to plant roots, and dissolving minerals such as phosphorus and synthesis of phytohormones [3]. It is known that endophyte bacteria are also capable of synthesizing auxins and cytokinin [4]. The effect of endophyte bacteria in indirect way is by increasing plant growth by suppressing the phytopathogens through antagonistic mechanisms. This includes the ability to synthesize anti-fungal metabolites such as antibiotics, such as hydrogen cyanide, endophyte bacteria as well as pathogenic competitors to obtain nutrients or special places in plant roots.

This group of bacteria have the potential to be developed into biotechnology products as biological fertilizers. One of product is a liquid organic fertilizer product, namely POC Garuda.

## 2. Materials and methods

### *2.1. Materials*
The research was conducted in Bedali District, Malang Regency, at ± 600 m above sea level. The research was started in August to December 2019. The materials were Jawara hot pepper seeds, organic fertilizers from fermented manure (cow, goat, chicken and mix), endophyte bacteria consortium in the form of liquid organic fertilizer (POC Garuda), pesticides and Santamikro as inorganic fertilizer. The tools were black silver mulch, bamboo, hoe, measurement tools and stationery.

### *2.2. Experimental design*
The study was arranged in factorial completely randomized design. The first factor is types of manure (M) while the second factor is the interval application of the endophyte bacteria consortium (I). Each treatment was described as follows: $M_1$: cow manure fertilizer, $M_2$: goat manure fertilizer, $M_3$: chicken manure fertilizer, $M_4$: mixed manure fertilizer. Meanwhile the interval of endophyte application is $I_1$: 5 days interval, $I_2$: 10 days, $I_3$: 15 days. Endophyte bacteria applied until the end of vegetative phase. Each treatment combination will be repeated four times.

### *2.3. Cultivation of hot pepper*
- Soil is processed using a hand tractor then forming beds
- Seeding on hot pepper seeds, transplanting is done at 3 weeks after seedlings
- Planting of hot pepper was carried out with a spacing of 30 cm x 30 cm. With a bed area of 9 $m^2$, in each bed there were 60 populations of plants. Experiment unit was determined among the population randomly.
- The organic fertilizer was used as base fertilizer at 300 grams per hole. The treatment is applied according to the fertilization recommendations. Fertilization treatment was repeated at 150 gr per plant at two weeks after planting.
- Endophyte bacteria sprayed with a dose of 200 cc per bed. With concentration of 10 $cc.lt^{-1}$[5]. It consists of *Bacillus sp*. ($3.2 \times 10^4$), *Pseudomonas sp*. ($1.6 \times 10^6$) and *Azotobacter* ($1.3 \times 10^6$)
- Maintenance is carried out by; replanting dead or damaged plants on first week after transplanting; applying fertilizers according to recommendations; preventing pests and diseases by spraying pesticides, weeding of weeds and watering.
- Harvesting is done when the plants at 60-67 days after planting, which is the first harvest period of 5 to 6 harvest periods during the generative phase.
- Observations were done periodically in accordance to the parameters to be observed. Variable observed were growth and crop yields. The growth components observed included: height of plants aged 1, 2, 3 and 4 weeks after planting and total of leaves. Leaves is counted at the leaves which have been opened perfectly, and done by using a hand counter. No pruning treatment was done in observation period. The yield obtained from total fruit and total weight per plant as experiment units.

### *2.4. Statistics analysis*
Data analyzed in two-way ANOVA, followed with the Duncan multiple range-test.

## 3. Results and discussion

### *3.1. Effect on plant growth*

The results showed that the highest plant is in treatment of cow manure at 15 day's intervals (Figure 1). The chicken manure at 10 day's intervals ($M_3I_2$ Treatment) has relatively the same effect, meanwhile, goat manure requires more frequent application of endophyte bacteria to be effective to support hot pepper growth. The process of breaking down the organic matter into nutrients which needed by the plants, was more efficient in cow manure in this study.

The transformation of manure into plant-available N is modulated by soil abiotic and biotic conditions. Solid manure must be decomposed first, before organic N are mineralized to release $NH^4$. The decomposition was beginning with physical fragmentation of larger particles to increase microbial colonization. Detritivorous microbe including endophyte bacteria are responsible for shredding and fragmenting the organic materials in solid manure. *Azotobacter*, *Bacillus* and *Pseudomonas* were mostly found in the liquid organic fertilizer applied. As bio-fertilizer its considered to be key factors to develop an integrated nutrient management system [3].

The C-rich compound such as in chicken manure stimulate microbial growth. But also cause gradual loss of C through microbial respiration. Solid manure contain a large amount of C could immobilize the N which available for plant [6]. The differences on water retention, nutrient supply and minor elements in each type of manure made different condition to plant development [7]

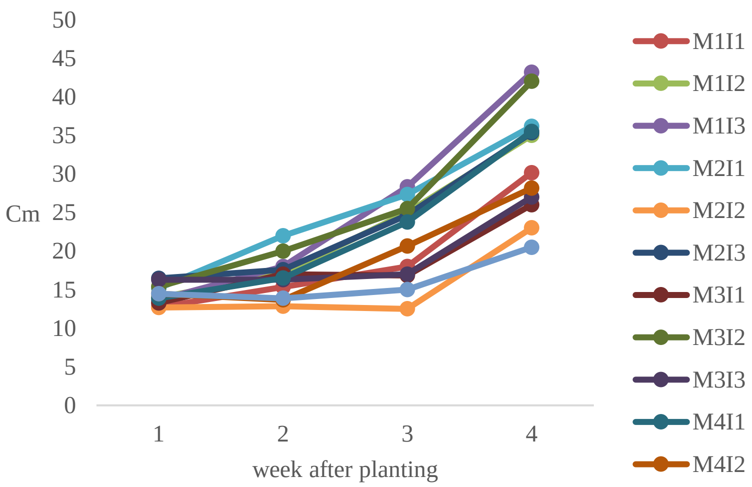


**Figure 1**. Comparison of plant height (cm) in time series per treatment

There are differences also found in total of leaves. The best treatment is obtained in cow manure (Figure 2). The total of leaves affects to production if the proportion of leaves serving as the source is greater than the leaves that function as sinks (users). However, from the point of view of production parameters, it shows that, without pruning, treatment of cow manure produces more leaves that function as sinks. Re-engineer the rhizosphere through inoculation with microbes must consider the connection between type of manure and its native microbiome [8]. Chicken manure need more bacteria activity to recycle lignocellulose-rich materials [9]. So, the more frequent induction of bacteria is needed. *Azotobacter sp*. need carbon and phosphate in order to N fixation [10]. Negative respond could be found if there were hyper-auxiny in the plant. The more microbe application could be induced. Moreover, over synthesis of auxin could inhibit plant growth [11]

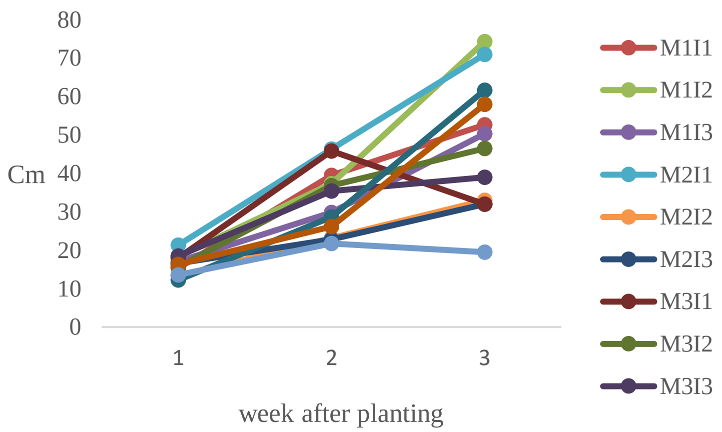


**Figure 2**. Comparison of total leaves per treatment

### 3.2. *Effect on plant productivity*

Treatment $M_1I_1$, $M_3I_2$ and $M_3I_3$ resulted total fruit range from 14 to 15 per plant. According to the boxplot, M1I1 has the narrowest (Figure 3). Based on the results of the ANOVA (Table 1), there was interaction between types of manure and the intervals ($P < 0.05$). Thus, the effectiveness of manure on the harvest quantity of hot pepper is influenced by the interval of given endophyte bacteria.

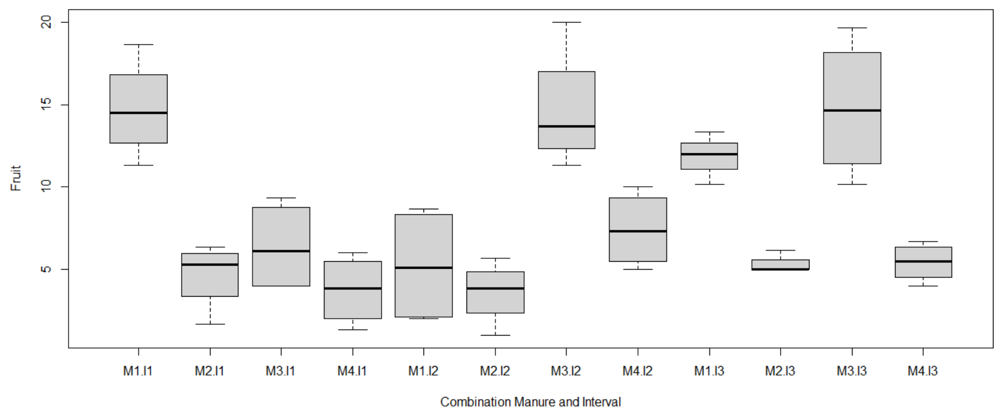


**Figure 3.** Boxplot of total fruit on first day harvested per treatment

Cow, goat, chicken and mixed manure produced different effects on the quantity of fruit. The results of DMRT showed that the highest number of fruits was produced by hot pepper which using cow and chicken manure as fertilizer (Table 2). The test resulted further confirm that the best treatment combination was obtained from either chicken manure with interval of 10 and 15 days ($M_3I_3$ and $M_3I_2$) and cow manure with interval 5 and 15 day. The average hot pepper fruit per plant was ranged from 14.67 to 14.79.

**Table 1.** Results of ANOVA in total fruits

| Source of Diversity | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Types of Manure | 487.379 | 3 | 162.460 | $23.489^{**}$ | 0.000 |
| Interval | 35.332 | 2 | 17.666 | 2.554 | 0.092 |
| Manure* Interval | 375.364 | 6 | 62.561 | $9.045^{**}$ | 0.000 |
| Error | 248.996 | 36 | 6917 | | |
| Total | 4,333.558 | 48 | | | |

Note: Asterix ($^*$) indicate highly significant differences ($p < 0.01$)

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**Table 2.** Results of total fruit

| Treatment Combination | Average of Total Fruit per Plant ± SD |
|---|---|
| $M_3I_3$ | 14.79 ± 4.211[a] |
| $M_1I_1$ | 14.75 ± 3.035[a] |
| $M_3I_2$ | 14.67 ± 3.732[a] |
| $M_1I_3$ | 11.88 ± 1.301[a] |
| $M_4I_2$ | 7.42 ± 2.315[a] |
| $M_3I_1$ | 6.38 ± 2.783[b] |
| $M_4I_3$ | 5.42 ± 1.667[b] |
| $M_2I_3$ | 5.29 ± 0.583[b] |
| $M_1I_2$ | 5.21 ± 3.619[b] |
| $M_2I_1$ | 4.64 ± 2.056[b] |
| $M_4I_1$ | 3.75 ± 2.132[b] |
| $M_2I_2$ | 3.58 ± 1.932[b] |

Note: Data shown is the mean ± SD, n= 4. Values in each row are denoted by letters indicate significant differences using two-way ANOVA ($p < 0.05$) with post hoc Duncan multiple-range test.

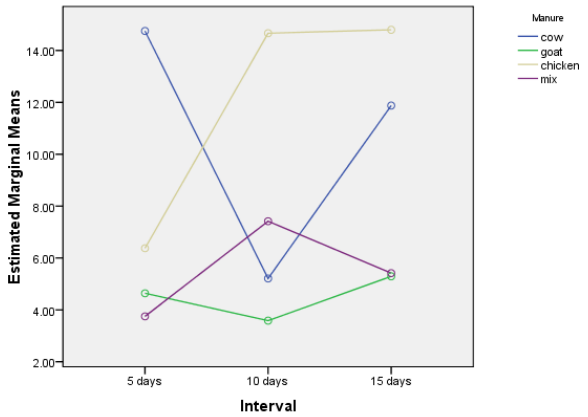


**Figure 4**. Interaction form of the treatment combination on total fruit

Cow manure is relatively less responsive to the intervals of endophyte bacteria application, where frequent or infrequent application tends to be the same. Chicken manure is the most responsive to endophyte bacteria, while rare application gives best product quantity (Figure 4). Bacteria activity affect the reduction of total losses of N and C in the composting process [12]. Different types of manure lead to different nitrification which play a key role in N cycling. Its transform ammonium into more mobile nitrate [13]. It seems that nitrification in chicken manure needs more bacteria activity than that in cow manure.

For the parameter of fruit weight per plant, data were obtained from the first harvest. The harvested fruit is the perfectly ripe fruit. If properly maintained, hot pepper has the potential to produce fruit up to the 6$^{th}$ harvest. The boxplot (Figure 5) showed that $M_1I_1$ has the best result, although it has wide variance.

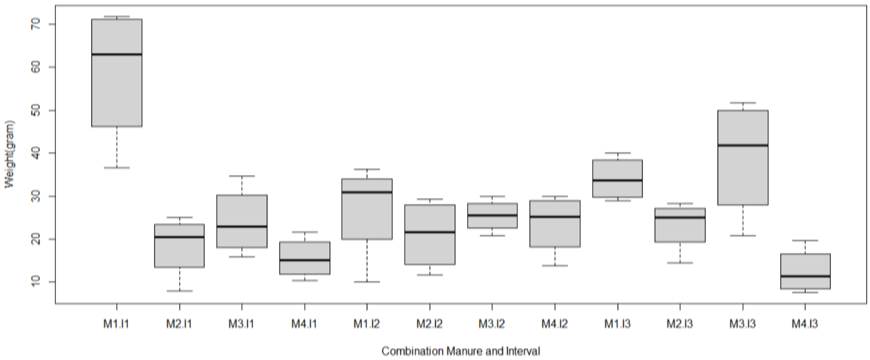


**Figure 5.** Boxplot of fruit weight per plant

Based on ANOVA (Table 3), there is a significant interaction between the type of manure and the intervals of giving endophyte bacteria. The result is relatively the same as the number of hot peppers, because the weight parameter is automatically determined by the number of fruits.

**Table 3.** Results of ANOVA in Fruit Weight

| Source of Diversity | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Type of Manure | 3652.227 | 3 | 1217.409 | $15.024^{**}$ | 0.000 |
| Interval | 196.369 | 2 | 98.185 | 1.2120 | 0.310 |
| Type of Manure * Interval | 2855.477 | 6 | 475.913 | $5.8730^{**}$ | 0.000 |
| Error | 2917.054 | 36 | 81.029 | | |
| Total | 4,4347.862 | 48 | | | |

Note: Asterix ($^*$) indicate highly significant differences ($p < 0.01$)

The results of further tests showed that the best treatment for hot pepper fruit parameters was obtained in a combination of cow manure with the most frequent interval (once every 5 days). This treatment has the most effect on minimizes fruit fly attack. The potential yield per bed can reach 17.58 kg per bed or equivalent to 19.5 tons per hectare. As known, manure has been applied as substitute for mineral fertilizer. Its main function is maintaining food security toward restore soil structure and fertility [14]. The use of manure as base fertilizer could support organic farming. It will minimize external non-renewable inputs [15], not only in-organic fertilizer but also the use pesticide.

**Table 4.** Result of weight per plant

| Treatment combination | Average weight ± SD (gr) |
|---|---|
| $M_1I_1$ | $58.60 \pm 16.33^{a}$ |
| $M_3I_3$ | $39.02 \pm 14.04^{b}$ |
| $M_1I_3$ | $34.06 \pm 5.191^{bc}$ |
| $M_1I_2$ | $26.98 \pm 11.63^{bcd}$ |
| $M_3I_2$ | $25.46 \pm 3.862^{bcd}$ |
| $M_3I_1$ | $24.13 \pm 8.105^{cd}$ |
| $M_4I_2$ | $23.59 \pm 7.193^{cd}$ |
| $M_2I_3$ | $23.24 \pm 6.095^{cd}$ |
| $M_2I_2$ | $21.08 \pm 8.315^{cd}$ |
| $M_2I_1$ | $18.50 \pm 7.418^{d}$ |
| $M_4I_1$ | $15.59 \pm 4.884^{d}$ |
| $M_4I_3$ | $12.53 \pm 5.369^{d}$ |

Note: Data shown is the mean ± SD, n= 4. Values in each row are denoted by letters indicate significant differences using two-way ANOVA ($p < 0.05$) with post hoc Duncan multiple-range test.

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The form of interaction between types of manure and intervals of giving endophyte bacteria is shown in Figure 6. It appears, that there is a large difference between the results of cow manure compare with chicken manure. This is due to the fact that hot pepper in the chicken manure treatment is much reduced due to fruit fly attack. So that the most suitable recommendation to produce the best quality harvest of hot pepper is to use cow manure at intervals of 5 days of endophyte bacteria. Inoculation bacterial as root symbiont can contribute to increase agronomic efficiency by reducing cost production [3].

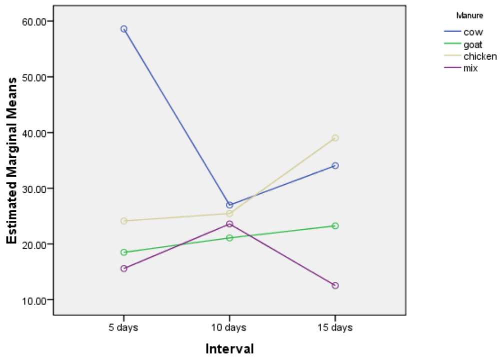


**Figure 6.** Interaction form of treatment combination on fruit weight per plant

## 4. Conclusions

According to the current study, t is concluded that there is an interaction between types of manure treatment with intervals of giving endophyte bacteria. The application of endophyte bacteria able to increase the efficiency of the manure given. Cow manure need more endophyte bacteria activity in order to have highest hot pepper productivity. The potential productivity of hot peppers can reach 19.5 tons per ha. In addition, treatment interactions can also reduce the potential loss in fruit ripening caused by fruit fly.

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