

# EFFECT OF BIOFERTILISER CONTAINING DIFFERENT PERCENTAGE RATES OF CHEMICAL FERTILISER ON OIL PALM SEEDLINGS

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## ABSTRACT

*Extensive use of inorganic substances contained in chemical fertilisers leads to environmental pollution and makes the plant more susceptible to diseases. The utilisation of beneficial microbes in biofertilisers has become a key solution in reducing the usage amount of chemical fertilisers in agriculture. This study was conducted on oil palm seedlings (OPS) in a nursery and treated with biofertiliser with zero chemicals (BF0), biofertiliser containing two percentage levels of chemical fertiliser (CF) (BF + 40% CF and BF + 50% CF) and two controls consisting of 100% chemical (CF100) and standard organic (SO) fertiliser. The vegetative measurements and foliar nutrients analysis for the OPS were recorded three and five months after fertilisation (MAF), when the OPS were six and eight months old, respectively. The application of biofertilisers containing low CF rates resulted in higher vegetative measurements compared to application of 100% chemical. Nutrients analysis showed that application of biofertiliser assisted in the increase of essential nutrients uptake in the OPS. Thus, we concluded that combination of biofertiliser with small rates of chemical able to increase vegetative measurements and nutrients uptake. Biofertiliser application is also capable in reducing the sole use of 100% chemical fertilisers, which is not environmental-friendly and expensive.*

**Keywords:** biofertiliser, chemical fertiliser, vegetative measurements, nutrients uptake.

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## INTRODUCTION

For optimum plant growth, nutrients must be supplied sufficiently and are in balanced quantities (Chen, 2006). Intensive farming practices, that demand high yield and quality, require the extensive use of chemical fertilisers. However, the most important constraint that limit the plant growth is soil fertility (Mohammadi and Sohrabi, 2012). Due to the extensive use of chemical fertilisers, which are not environmental-friendly, has resulted in many environmental problems such as soil acidification, greenhouse gases emission and destruction of friendly microorganisms which reduce the soil

fertility due to the excessive nutrients, heavy metals and high concentration of radionuclides (Savci, 2012; Zhao *et al.*, 2016). Apart from that, sole application of chemical fertilisers has also displayed the disadvantages such as leaching and polluting water basins making the plants more susceptible to the attack of diseases and causing irreparable damage to the overall biological system (Ibiene *et al.*, 2012).

Researchers started to introduce beneficial microbes in inorganic fertilisers as the microbes are effective in promoting plant growth by secreting out plant growth promoters such as auxins (Zahir *et al.*, 2010). Biofertilisers which contain bacteria inoculants help to reduce environmental problems by maintaining many kinds of important nutrients in the soil via nitrogen fixation, phosphate and potassium solubilisation, release of plant growth

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promoter and biodegradation of organic matters in soil (Bhardwaj *et al.*, 2014).

Plant requires optimum supply of nutrients to enhance its growth and yield. The deficiency of important nutrients can adversely affect plant growth and performance (Miransari, 2013). Unlike chemical fertilisers which are not environmental-friendly, biofertilisers which contain different types of microorganisms, have become an essential component to sustain the oil palm growth by enhancing the nutrients uptakes (Mishra *et al.*, 2013). Inoculants containing beneficial microbes applied in fertilisers are responsible in supplying adequate nutrients to the plant and increase plant growth (Egamberdiyeva, 2007).

Nowadays, studies to reduce the usage of inorganic fertilisers have received great attention by researchers. Successful nutrient management in biofertilisers can provide sustainable yields and become an alternative to replace the usage of 100% chemical fertilisers. Essential nutrients such as nitrogen, phosphorus and potassium must be present in biofertilisers for uptake by roots (Munda *et al.*, 2015). Organism that are commonly used as biofertilisers component are nitrogen fixers (N-fixer), potassium solubiliser (K-solubiliser) and phosphorus solubiliser (P-solubiliser), or with the combination of fungi. Most of the microbes included in biofertiliser have closed relationship with plant roots (Mohammadi and Sohrabi, 2012). Apart from that, Gupta (2004) stated that microorganism contained in biofertilisers mainly bacteria and fungi possess the ability to make insoluble P available to the plants and to bring insoluble phosphate into soluble forms.

The sole application of chemical fertilisers has led to a decline in soil fertility (Savci, 2012). Thus, researchers agreed that biofertiliser application with the integration of small amounts of chemical fertilisers can maintain and increase the soil fertility while enhancing the plant development (Mekki, 2016). Mondal *et al.* (2015) studied the impact of reduced dose of chemical fertiliser (CF) with its combination with biofertiliser of mustard (*Brassica campestris* cv. B<sub>9</sub>). The fertiliser was formulated using a full recommended dose of CF (N:P:K-100:50:50) combined with biofertiliser. Apart from that, a study on application of organic fertiliser in combination with CF (N:P:K-15:15:15) also indicated significant effect on the growth of kenaf (*Hibiscus cannabinus* L.) (Hadi *et al.*, 2013). Therefore, this study was conducted to determine the effects of biofertilisers as well as to compare the effects of biofertiliser containing different rates of CF with the application of 100% chemical (CF100) and standard organic (SO) fertilisers on vegetative measurements and nutrients uptake of oil palm seedlings (OPS) after three and five months of fertiliser application.

## MATERIALS AND METHODS

### Preparation of Inoculum and Biofertiliser Formulation

Seventeen different kinds of microorganisms from several classes of bacteria and fungi were utilised in the production of biofertiliser in this research. The microbes were chosen in this formulation based on their capability to promote plant growth. The consortium of microbes consisted of five microbes from prokaryotes of group *Proteobacteria*; two bacteria were from the genus *Bacillus* and the other three bacteria were from the genera of *Providencia*, *Phyllobacterium* and *Sphingobacterium*. Fungi consisted of three species of *Trichoderma* and one species each of *Antrodia*, *Pichia*, *Pycnoporus* and *Phanerochaete*. Generally, these microbes were protected under Malaysia Patent No. PI 2017703375.

In order to prepare the microbial consortium formulation, for fungi, malt extract broth was prepared by dissolving 19 g of malt extract broth powder in 400 ml of distilled water. For bacteria, a volume of 400 ml nutrient broth was prepared by dissolving 3.2 g of nutrient broth powder in 400 ml distilled water. The fermentation media were sterilised by autoclaving at 121°C for 20 min before inoculating the individual microbes. After inoculation, the broths containing inoculum were incubated in an INNOVA44 incubator shaker at a speed of 170 rpm at 30°C for a period of three days to produce fermented products. The different individual fermented microbes were pooled together on the basis of synergistic behaviour.

### Nursery Trial

The experiment was conducted at a commercial nursery in Johor, Malaysia. The soil type used in polybag was clay loam. Three months old oil palm seedlings were fertilised with biofertiliser with zero chemical (BF0), biofertiliser containing two different percentage rates of CF (60% BF + 40% CF and 50% BF + 50% CF) and two controls consisting of 100% CF (CF100) with ratio of 12:12:17:2 (N: P: K: Mg) and SO medium in the form of unsieved premium compost. The five treatments were carried out in replicates of eight for every harvest. Therefore, a total of 40 experimental units were tested in this research for one harvest. The oil palm seedlings were provided by the Malaysian Palm Oil Board, Kluang, Johor.

### Vegetative Measurements

Vegetative measurements for each replicate of OPS were recorded after three and five months of fertiliser application which means that the age of the OPS were six and eight months old, respectively. The

measurements taken were plant height, meristem diameter, and number of fronds.

**Foliar Analysis**

At the age of six and eight months, leaflets of OPS were dried and then ground. Fifty milligrams of dried leaf powder were weighed into each crucible. The samples were then subjected to total nitrogen (N) analysis with Primacs SNC analyser (Skalar, UK). For analyses of other nutrients such as boron (B), magnesium (Mg), phosphorus (P), potassium (K), calcium (Ca), manganese (Mn), iron (Fe), copper (Cu), zinc (Zn), and molybdenum (Mo), the samples were first digested using a Microwave Oven (Memmert, Germany) according to the US EPA Test Method 3050B (Kimbrough and Wakakuwa, 1989). The digested samples were filtered using filter paper and diluted with MilliQ water prior to analysis using ICP Mass Spectrometer ELAN DRC (Perkin Elmer SCIEX, USA).

**Statistical Analysis**

Results of the nutrient analyses were analysed statistically using IBM SPSS Statistics software. All the data were reported as means ± SE (standard error). The mean for each treatment for the respective analysis was separated statistically using Duncan’s test as we wanted to test the differences among mean values of every treatment at p< 0.05 level of significance.

**RESULTS AND DISCUSSION**

Biofertilisers amended with inorganic fertiliser are used to increase nutrient availability to plant and can also affect the soil microbial biodiversity (Javoreková *et al.*, 2015). Generally, the microbial consortium includes plant growth promoting rhizobacteria, nitrogen fixing bacteria, plant disease suppressive bacteria and fungi, soil toxicant degrading microbes, actinomycetes which are

important for soil health, plant growth and yield as well as to increase soil microbial diversity (Haque *et al.*, 2011; Egamberdiyeva, 2007). Microbes from the genus *Bacillus* are the most commonly used as they always dominate in the rhizosphere (Morgan *et al.*, 2005). While microbes belonging to the genus *Trichoderma* have also been described to be beneficial for host plants as they are involved in a complex mechanism to also promote plant growth, development and protection (Nihorimbere *et al.*, 2011).

The vegetative measurements of OPS at six and eight months old are shown in *Table 1*. For both harvests, the plant height of OPS treated with BF + 50% CF were the highest at 37.75 cm and 64.75 cm, respectively. This shows that application of biofertiliser integrated with CF can increase height of OPS. Mekki (2016) also found that application of biofertiliser in combination with CF enhanced the growth and yield of plant. Similar observation was made on the number of fronds; the application of BF + 50% CF has led to the highest number of leaves for six (5.38) and eight months old (10.50) of OPS (*Table 1*). Meanwhile, the meristem diameters of OPS at the age of six months were comparable between all treatments ranging between 10.55-13.13 cm. However, at the age of eight months, the oil palm seedlings treated with BF + 50% CF were the highest at 34.00 cm and was significantly higher than CF100 (11.66 cm) and SO (2.33 cm). The effects of biofertilisers on growth of the seedlings were due to the plant growth-promoting microbes contained in the biofertiliser which are responsible for enhancing plant growth and protect them from diseases (Dimkpa *et al.*, 2009). For example, P-solubilising bacteria improved nutrients availability by increasing nitrogenase activity which resulted in better growth of OPS (Dadresan *et al.*, 2015).

Apart from that, significant improvements on the meristem diameter was aided by the microbes attaching to the roots and colonising the root surfaces efficiently and stimulated the development and health of the plants (Hayat *et al.*, 2010; Vacheron *et al.*, 2013; Van Der Heijden *et al.*, 2008). In addition,

**TABLE 1. VEGETATIVE MEASUREMENT FOR OIL PALM SEEDLINGS TREATED WITH BIOFERTILISER CONTAINING DIFFERENT PERCENTAGE RATES OF CHEMICAL FERTILISER, 100% CHEMICAL AND STANDARD ORGANIC FERTILISER**

Treatment	Six MOOS			Eight MOOS		
	Ph (cm)	Md (cm)	NOF	Ph (cm)	Md (cm)	NOF
BF0	35.75±0.98 <sup>b</sup>	12.75±0.59 <sup>a</sup>	5.25±0.25 <sup>a</sup>	61.63±3.55 <sup>b</sup>	31.25±1.13 <sup>c</sup>	10.50±0.50 <sup>b</sup>
BF + 40% CF	12.38±0.65 <sup>a</sup>	13±0.80 <sup>a</sup>	5.00±0.33 <sup>ab</sup>	56.50±2.34 <sup>ab</sup>	30.88±1.48 <sup>c</sup>	10.38±0.46 <sup>b</sup>
BF + 50% CF	37.75±2.21 <sup>b</sup>	13.125±0.58 <sup>a</sup>	5.38±0.26 <sup>b</sup>	64.75±2.35 <sup>b</sup>	34.00±2.97 <sup>c</sup>	10.50±0.73 <sup>b</sup>
CF100	13.19±0.38 <sup>a</sup>	10.55±1.45 <sup>a</sup>	4.63±0.26 <sup>ab</sup>	59.13±1.65 <sup>b</sup>	11.66±4.40 <sup>b</sup>	8.13±0.30 <sup>a</sup>
SO	13.25±0.88 <sup>a</sup>	12.38±0.75 <sup>a</sup>	4.25±0.16 <sup>a</sup>	49.38±3.82 <sup>a</sup>	2.33±0.26 <sup>a</sup>	8.13±0.40 <sup>a</sup>

Note: Data reported as mean and standard error for oil palm seedlings (OPS) treated with biofertiliser (BF) containing different percentage rates of chemical fertiliser (CF), 100% chemical (CF100) and standard organic (SO) fertiliser. <sup>abcd</sup> In the same row indicated the significant different [analysis of variance (ANOVA) p<0.05]. MOOS - month oil palm seedling, Ph - plant height, Md - meristem diameter, NOF - number of frond. BFO - biofertiliser with zero chemicals.

the increase in the vegetative growth of the OPS was due to the uptake of N from the soil as this nutrient is responsible for chlorophyll content and it is also vital for synthesis of food material in plants (Fraile *et al.*, 2015). It was revealed that the integration of microbial inoculants contained in biofertilisers was significant for plant height, diameter and yield production (El-Naggar, 2010). Biofertilisers are considered as an alternative to reduce the impact of chemical fertilisers in agriculture as it contain different kind of microbes which have the ability to convert nutrients from unavailable to available forms and produced plant-growth stimulating factors (Chen, 2006).

Chemical or microbial-based fertiliser has its advantages and disadvantages in terms of nutrient supply, soil quality and crop growth. Developing a biofertiliser by integration with a CF was believed to promote the nutrient uptake as it was aided by the beneficial microbes contained in the biofertiliser (Azab and El-Dewiny, 2018). Plants need adequate macronutrients and micronutrients to ensure a healthy plant growth as they are among the most important factors affecting the plant productivity. N, P, and K are macronutrients that play a vital role in ensuring the growth of the plant (Figures 1 and 2). Their availability in biofertilisers help the microbes to work efficiently (Miransari, 2013). The deficiency of some macronutrients will adversely affect the plant health and growth (Hirel *et al.*, 2011). For OPS at six months old, N was recorded highest in the seedlings treated with biofertiliser containing 50% CF (2.55%) as compared to the application of CF100

and SO (1.55% and 1.23%, respectively). After five months of application, when the OPS were eight months old, N was increased and maintained as the highest (3.19%) in OPS treated with biofertiliser containing 50% of CF compared to sole application of CF (3.07%) and SO (2.59%).

P is among the most important nutrient for root, seed and fruit development in plants. Sufficient uptake of P is vital for the development of plants (Silva and Uchida, 2000). P deficiency may lead to poor growth of the plant (Sharma *et al.*, 2013). The application of BF + 50% CF had resulted in the highest P uptake in the soil (1287.52 mg litre<sup>-1</sup>) as compared to the application of CF100 (347.88 mg litre<sup>-1</sup>) and SO (362.53 mg litre<sup>-1</sup>) for six months old OPS. However, for eight months old OPS, treatment with BF + 40% CF (2623.88 mg litre<sup>-1</sup>) indicated the highest P content compared to CF100 (624.38 mg litre<sup>-1</sup>) and SO (711.80 mg litre<sup>-1</sup>). The higher foliar P content in the OPS was due to the application of biofertilisers that contain phosphate solubilising bacteria, which have the capability to solubilise inorganic phosphate compounds and liberate the organic phosphate (Oteino *et al.*, 2015).

Macronutrient K is considered as one of the important nutrients to improve the growth and yield of plants. Apart from that, K is also an essential nutrient as it is involved in metabolic processes such as photosynthesis and transport of sugar. It is also responsible for the survival of plants under biotic and abiotic stresses (Wang *et al.*, 2013). The deficiency of K may lead to the scorching effects of leaf, slow and poor development of root (Prajapati and Modi,

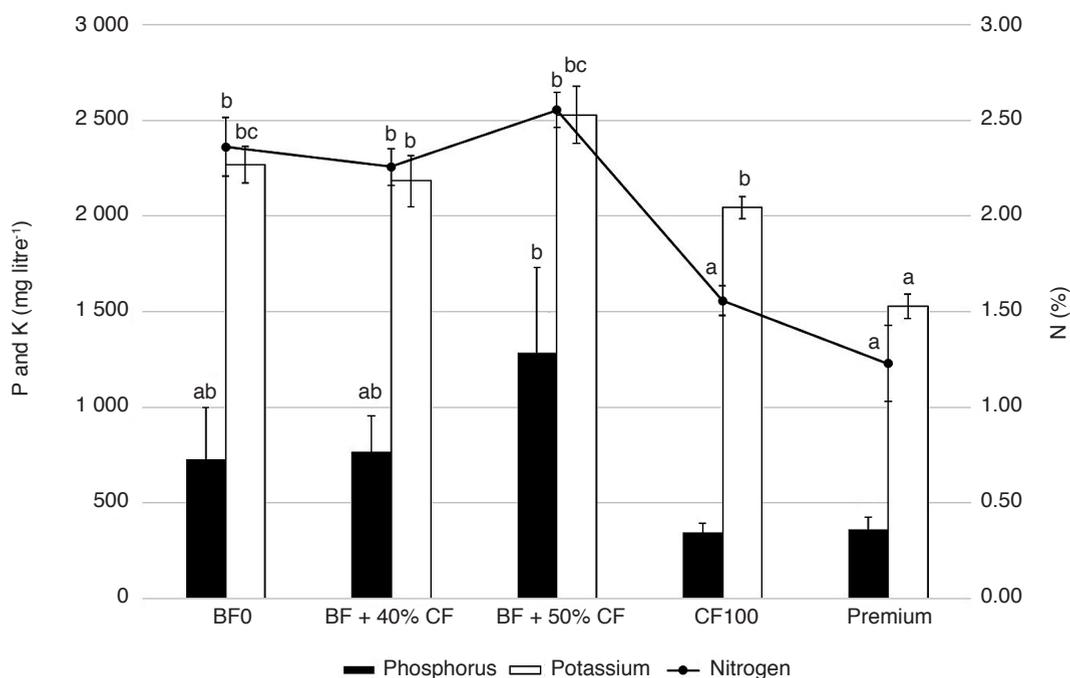


Figure 1. Nitrogen (N), phosphorus (P) and potassium (K) content in oil palm seedlings (OPS) of six months old treated with biofertiliser (BF) containing different percentage rates of chemical fertiliser (CF), 100% chemical (CF100) and standard organic (SO) fertiliser.

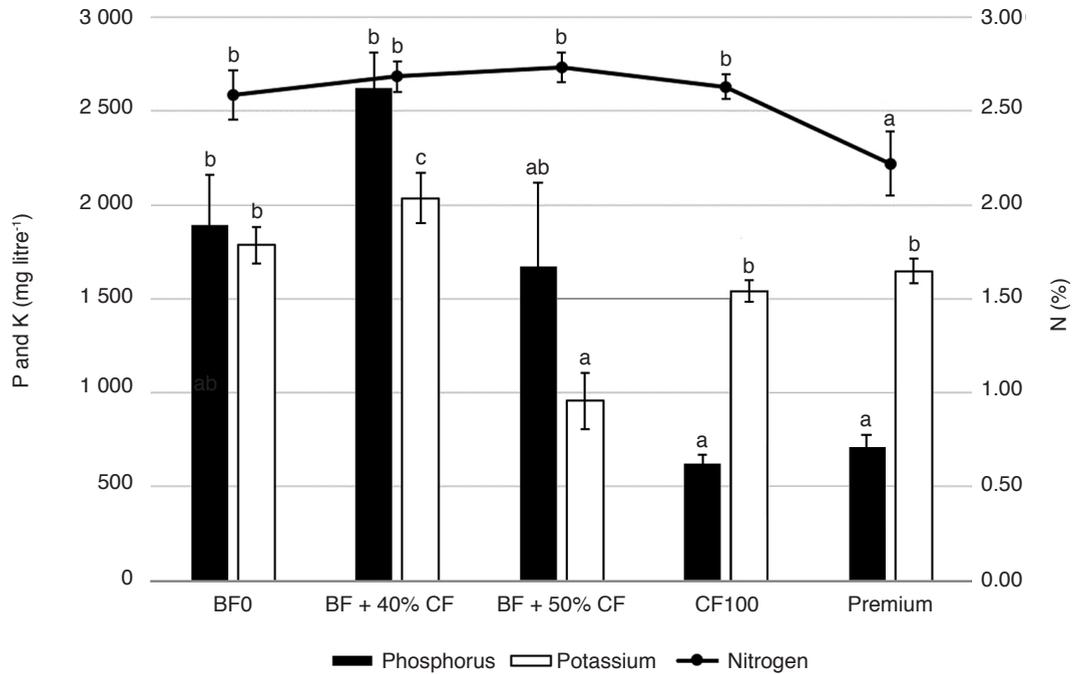


Figure 2. Nitrogen (N), phosphorus (P) and potassium (K) content in oil palm seedlings (OPS) of eight months old treated with biofertiliser (BF) containing different percentage rates of chemical fertiliser (CF), 100% chemical (CF100) and standard organic (SO) fertiliser.

2012). Similar to P, K was recorded highest in OPS of six months old which were treated with BF + 50% CF (2528.93 mg litre<sup>-1</sup>) compared to CF100 (2043.95 mg litre<sup>-1</sup>) and SO (1528.66 mg litre<sup>-1</sup>). However, at eight months old, K was significantly high in OPS treated with BF + 40% CF (2037.96 mg litre<sup>-1</sup>). Meanwhile, the OPS treated with CF100 and SO have low K levels which were 1543.54 mg litre<sup>-1</sup> and 957.833 mg litre<sup>-1</sup>, respectively.

Plants also need a right combination of micronutrients to live and grow (Tables 2 and 3). The application of biofertilisers has also increased the level of micronutrients B, Mg, Ca, Mn, Fe, Cu, Zn and Mo. An adequate amount of the micronutrients help to elevate biochemical and metabolic characteristics in plants (Tripathi *et al.*, 2015). Besides that, micronutrients also resulted in the increase of the vegetative measurements of the plants. The deficiency of micronutrients will give similar symptoms to the plants such as chlorosis and yellowing of the leaves (Bradley and Hosier, 1999). B is among the important trace element as it helps to develop the plasma membrane and is involved in several metabolic pathways. The deficiency of B in plants would impose unhealthy effects such as yellowing of the leaf tips, chlorosis and necrotic spot and stunting the growth of the plant (Cristobal *et al.*, 2008). In this research, the application of BF +50% CF has resulted in the highest boron content in the OPS of six months old which was 5.18 mg litre<sup>-1</sup> as compared to CF100, 3.54 mg litre<sup>-1</sup> and SO, 3.50 mg litre<sup>-1</sup>. However, after five months of fertiliser

application, B was highest in OPS treated with BF0 which was 6.14 mg litre<sup>-1</sup> compared to CF100 and SO which were 3.66 mg litre<sup>-1</sup> and 5.17 mg litre<sup>-1</sup> respectively. This result showed a promising effect in using biofertilisers rather than chemical which is harmful for the environment.

Adequate amount of Mg acts as a regulator of physiological processes in plants which influence their susceptibility to plant diseases. Deficiency of Mg could lead to the degradation of chlorophyll in leaves (Huber and Jones, 2013). Mg content in OPS after three months of fertiliser application was comparable and there was no significant difference between all treatments; whilst SO indicated slightly high Mg content which was 2368.92 compared to the three biofertilisers (2190.35-2323.57 mg litre<sup>-1</sup>) and CF (2243.83 mg litre<sup>-1</sup>). However, after five months, Mg was recorded the highest in OPS treated with BF + 50% CF which was 4081.89 mg litre<sup>-1</sup> compared to CF100 (1981.21 mg litre<sup>-1</sup>) and SO (3071.92 mg litre<sup>-1</sup>).

Higher Ca was recorded in six months old OPS, treated with BF + 50% CF (7630.68 mg litre<sup>-1</sup>) compared to the seedlings treated with CF100 and SO (3988.50 mg litre<sup>-1</sup> and 4699.45 mg litre<sup>-1</sup> respectively). The application of biofertiliser promoted the uptake of Ca from soil, which resulted in the increase of foliar Ca in OPS at eight months old. The highest recorded Ca was in OPS treated with BF + 40% CF, which was 3664.83 mg litre<sup>-1</sup>, compared to seedlings treated with CF100 (2489.97 mg litre<sup>-1</sup>) and SO (3081.95 mg litre<sup>-1</sup>). The increase

TABLE 2. MICRONUTRIENT CONTENT ANALYSIS IN OIL PALM SEEDLINGS TREATED WITH BIOFERTILISER CONTAINING DIFFERENT PERCENTAGE RATES OF CHEMICAL FERTILISER, 100% CHEMICAL AND STANDARD ORGANIC FERTILISER AT SIX MONTHS OLD

Treatment	Micronutrients (mg litre <sup>-1</sup> )								
	B	Mg	Ca	Mn	Fe	Cu	Zn	Mo	
BF0	4.41±0.23 <sup>b</sup>	2 323.57±138.15 <sup>a</sup>	6 158.58±411.55 <sup>a</sup>	77.46±28.57 <sup>a</sup>	137.79±7.79 <sup>a</sup>	5.19±0.50 <sup>c</sup>	133.50±9.32 <sup>b</sup>	0.43±0.05 <sup>ab</sup>	
BF + 40% CF	5.02±0.22 <sup>bc</sup>	2 190.35±76.04 <sup>d</sup>	5 363.69±274.76 <sup>a</sup>	355.26±172.47 <sup>b</sup>	321.04±22.90 <sup>b</sup>	3.36±0.13 <sup>ab</sup>	159.04±14.70 <sup>b</sup>	0.47±0.08 <sup>ab</sup>	
BF + 50% CF	5.18±0.24 <sup>c</sup>	2 260.63±58.41 <sup>a</sup>	7 630.68±3 722.46 <sup>a</sup>	119.87±17.79 <sup>ab</sup>	316.63±29.64 <sup>b</sup>	4.53±0.64 <sup>bc</sup>	142.37±6.85 <sup>b</sup>	0.90±0.46 <sup>b</sup>	
CF100	3.54±0.16 <sup>a</sup>	2 243.83±85.52 <sup>a</sup>	3 988.52±188.34 <sup>a</sup>	110.68±40.26 <sup>ab</sup>	177.74±12.74 <sup>a</sup>	4.14±0.29 <sup>bc</sup>	31.90±6.70 <sup>a</sup>	0.00±0.00 <sup>a</sup>	
SO	3.50±0.30 <sup>a</sup>	2 368.92±89.78 <sup>a</sup>	4 699.45±290.56 <sup>a</sup>	85.35±31.37 <sup>a</sup>	176.97±12.76 <sup>a</sup>	2.24±0.20 <sup>b</sup>	24.12±7.43 <sup>a</sup>	0.00±0.00 <sup>a</sup>	

Note: Data reported as mean and standard error for oil palm seedlings (OPS) of six months old treated with biofertiliser (BF) containing different percentage rates of chemical fertiliser (CF), 100% chemical (CF100) and standard organic (SO) fertiliser. <sup>abcd</sup>In the same row indicated the significant different [analysis of variance (ANOVA)  $p < 0.05$ ].

TABLE 3. NUTRIENT CONTENT ANALYSIS IN OIL PALM SEEDLINGS TREATED WITH BIOFERTILISER CONTAINING DIFFERENT PERCENTAGE RATES OF CHEMICAL FERTILISER, 100% CHEMICAL AND STANDARD ORGANIC FERTILISER AT EIGHT MONTHS OLD

Treatment	Micronutrients (mg litre <sup>-1</sup> )								
	B	Mg	Ca	Mn	Fe	Cu	Zn	Mo	
BF	6.14±0.40 <sup>b</sup>	3 405.22±147.07 <sup>b</sup>	2 726.30±132.69 <sup>a</sup>	45.60±8.26 <sup>a</sup>	274.55±29.90 <sup>a</sup>	548.96±54.40 <sup>b</sup>	175.03±8.28 <sup>b</sup>	74.22±33.09 <sup>b</sup>	
BF + 40% CF	5.38±0.44 <sup>b</sup>	3 972.86±567.60 <sup>b</sup>	3 664.83±517.01 <sup>b</sup>	50.68±4.35 <sup>a</sup>	236.38±21.66 <sup>a</sup>	532.45±48.50 <sup>b</sup>	185.19±23.22 <sup>a</sup>	34.57±7.24 <sup>a</sup>	
BF + 50% CF	5.63±0.68 <sup>b</sup>	4 081.89±642.93 <sup>b</sup>	3 359.25±370.82 <sup>ab</sup>	290.40±94.86 <sup>b</sup>	266.56±66.89 <sup>a</sup>	496.89±63.68 <sup>a</sup>	190.93±28.88 <sup>a</sup>	46.59±8.77 <sup>a</sup>	
CF	3.66±0.09 <sup>a</sup>	1 981.21±114.53 <sup>a</sup>	2 489.97±142.56 <sup>a</sup>	36.70±3.38 <sup>a</sup>	267.32±27.93 <sup>a</sup>	409.11±29.73 <sup>a</sup>	213.57±5.83 <sup>a</sup>	62.33±37.27 <sup>a</sup>	
SO	5.17±0.21 <sup>b</sup>	3 071.92±152.06 <sup>ab</sup>	3 081.95±90.36 <sup>ab</sup>	33.85±3.80 <sup>a</sup>	238.05±10.82 <sup>a</sup>	477.78±31.93 <sup>a</sup>	197.11±12.83 <sup>a</sup>	67.53±6.56 <sup>a</sup>	

Note: Data reported as mean and standard error for oil palm seedlings (OPS) of eight months old treated with biofertiliser (BF) containing different percentage rates of chemical fertiliser (CF), 100% chemical (CF100) and standard organic (SO) fertiliser. <sup>abcd</sup>In the same row indicated the significant different [analysis of variance (ANOVA)  $p < 0.05$ ].

of Ca in OPS treated with microbe-based fertiliser explained the increase in vegetative growth of OPS as Ca is important to promote plant development (Hepler, 2005). Stunted plant growth, necrotic leaf margins and eventual death of terminal buds and root tips are due to the deficiency of Ca in plants (Stevens *et al.*, 2002).

Similarly with Mn, biofertiliser application also resulted in its highest content in six months old OPS treated with BF + 40% CF (355.26 mg litre<sup>-1</sup>) compared to CF100 (110.68 mg litre<sup>-1</sup>) and SO (85.35 mg litre<sup>-1</sup>). A significant difference of Mn was seen in eight months old OPS fertilised with BF + 50% CF (290.40 mg litre<sup>-1</sup>). However, seedlings treated with CF100 and SO indicated the lowest Mn contents which were 36.70 mg litre<sup>-1</sup> and 33.85 mg litre<sup>-1</sup> respectively. Deficiency or excess of Mn lead to the yellowing of the leaves, premature fall of the leaf, delayed maturity, and formation of white and grey spots on leaves (Tripathi *et al.*, 2015).

Fe is also among the important elements which are required by plants for living. The increased amount of Fe could affect the uptake of other nutrients such as Ca, Mg, P, and K. Therefore, Fe is needed in adequate amounts to avoid excess amounts which in turn creates another micronutrient deficiency (Cui *et al.*, 2015). The application of BF + 40% CF promoted the uptake of the nutrient from the soil which has resulted in the highest foliar Fe in six months old OPS which was 321.04 mg litre<sup>-1</sup>, compared to application with CF100 (177.74 mg litre<sup>-1</sup>) and SO (176.97 mg litre<sup>-1</sup>). At eight months old, all treatments gave comparable results; with BF0 being slightly higher (274.55 mg litre<sup>-1</sup>) compared to other treatments (236.38 mg litre<sup>-1</sup> - 267.32 mg litre<sup>-1</sup>).

Cu is essential for promoting metabolism of plants and helps in increasing crop yield. However, excessive application might induce toxicity to the plant (Zhu *et al.*, 2012). Inadequate amounts of Cu in plants adversely affect plant growth and plant photosynthesis process (Peng *et al.*, 2013). After three months of fertilisation, Cu recorded highest in OPS treated with BF0 (5.19 mg litre<sup>-1</sup>) compared to application of CF100 (4.14 mg litre<sup>-1</sup>) and SO (2.24 mg litre<sup>-1</sup>). However, after five months of fertiliser application, biofertiliser managed to increase the uptake of Cu from the soil. Cu was seen comparable between all treatments which ranged between 409.11- 548.96 mg litre<sup>-1</sup>.

Johnson (2010) stated that the microbes in biofertiliser are able to increase the Zn uptake from the soil. Deficiency of Zn may lead to stunted development of plants and chlorosis effect on leaves (Sharma *et al.*, 2015). For OPS of six months old, application of BF + 40% CF resulted in the highest Zn content (159.04 mg litre<sup>-1</sup>) compared to application of CF100 (31.90 mg litre<sup>-1</sup>) and SO (24.12 mg litre<sup>-1</sup>). The amount of Zn increased after five months

of fertilisation; which was observed comparable between all treatments ranging between 175.03-213.57 mg litre<sup>-1</sup>.

Mo was highly significant in six months old OPS treated with all kinds of biofertiliser. Seedlings treated with BF + 50% CF yielded the highest Mo content (0.90 mg litre<sup>-1</sup>) whilst for CF100 and SO application, zero Mo was detected. For application of fertiliser after five months, BF0 which is chemical-free indicated the highest Mo which was 74.22 mg litre<sup>-1</sup> compared to sole application of chemical, CF100 (62.33 mg litre<sup>-1</sup>) and SO (67.53 mg litre<sup>-1</sup>). It was noted that using biofertiliser can help to improve plants uptake of Mo from the soil. Mo is known to be one of the important micronutrients for plant growth and development. Its deficiency leads to the chlorosis effect of the leaves (Fageria *et al.*, 2015).

The growth-promoting microbes contained in biofertiliser are able to regulate many enzymatic processes which consequently increase the vegetative growth of the plant. Furthermore, the utilisation of microbes in fertilisation can act as a biological control for plant pathogens, N fixation, and solubilisation of beneficial nutrients (Ge *et al.*, 2016). Researchers found out that the application of biofertiliser with integration of low rates of chemical was responsible in enhancing the nutrients uptake and vegetative measurements of plants. Apart from that, Rakha and El-Said (2013) stated that the mixture of microbial consortium with inorganic fertiliser can cause the plants to receive the best nutritional values resulting in the higher content of N, P, K and other nutrients content. In addition, its application can also reduce the sole use of chemical fertilisers which are not environmental-friendly and expensive.

## CONCLUSION

Compared to the sole application of 100% chemical, the application of biofertiliser containing low chemical rates significantly improved the vegetative measurements and the nutrients absorption of oil palm seedlings. The usage of microbial inoculants contained in biofertiliser helped to regulate the availability of nutrients in the soil which resulted in healthy oil palm. Adding to this, the beneficial microbes contained in the biofertiliser improved the plant quality, increased the soluble P content, helped in nutrients intake by the plants, and enhanced the populations of phosphate-solubilising and proteolytic bacteria. Based from our results, the application of biofertiliser with the combination of CF gave a higher quality effect on the OPS. Therefore, we conclude that the integration of environmental-friendly biofertiliser with low rates of CF provides a promising alternative to replace the usage of 100%

CF, which is expensive and less friendly to the environment.

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