

## RECYCLING OF OIL PALM EMPTY FRUIT BUNCH AS POTENTIAL CARRIER FOR BIOFERTILIZER FORMULATION

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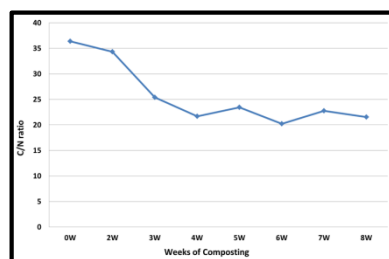
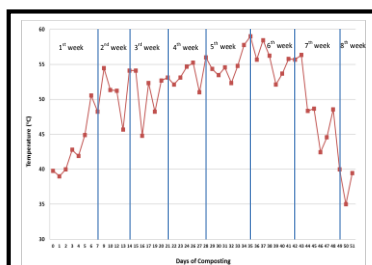
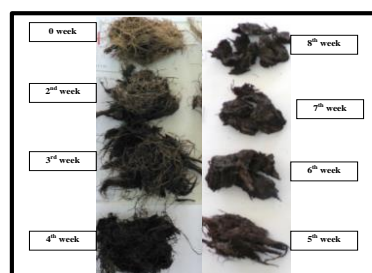
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### Graphical abstract



### Abstract

The palm oil industry generates large amount of biomass waste such as oil palm empty fruit bunch (EFB) and palm oil mill effluent (POME). This biomass would be typically recycled to produce EFB compost that is a valuable agricultural input. This study was conducted to evaluate the suitability of using EFB compost as carrier for nitrogen fixing bacteria (NFB) and phosphate solubilizing bacteria (PSB). Mixture (50-60% moisture) between EFB (shredded short fibres) and POME (anaerobic pond) were added with Effective Microorganisms (EM) to accelerate the composting process. The EFB compost reached thermophilic phase after 4-6 weeks with consistent temperature between 50-60°C. After 7 weeks of composting, EFB compost reached the mesophilic phase with continuous reduction of temperature to 35-40°C at week 8. The maturity of the compost is supported from the reduction of the C/N ratio from 36 (initial) to 20 (after 6 weeks). Mature EFB compost was sun-dried and ground into fine particle size (1 mm) prior to be used as carrier. Dried EFB compost carrier has the following characteristics (dry weight); average pH at 7.5, C/N ratio - 13.5, moisture - 17.4%, organic matter - 74.5%, total N - 3.06%, P - 0.37%, K - 4.74%, Ca - 3.32% and Mg - 0.79%. The inoculation of NFB and PSB into the EFB compost carrier from single cultures of *Serratia marcescens* and *Enterobacter cloacae* showed high viable cell count at  $4.05 \times 10^9$  CFU/g and  $2.75 \times 10^8$  CFU/g respectively at day three after inoculation. Meanwhile, the mixed culture of *Burkholderia cenocepacia* with *Serratia marcescens* showed  $2.45 \times 10^8$  CFU/g and  $4.31 \times 10^9$  CFU/g respectively. This clearly indicates the potential of using EFB as a useful alternative for bacterial immobilization prior to application in the oil palm industries.

Keywords: EFB, POME, compost, carrier, biofertilizer

### Abstrak

Industri minyak sawit menjana jumlah sisa biomass yang banyak seperti tandan kelapa sawit kosong (EFB) dan efluen kilang minyak sawit (POME). Biomass ini biasanya dapat dikitar semula untuk menghasilkan kompos EFB sebagai input pertanian yang bernilai. Kajian ini dijalankan untuk menilai kesesuaian penggunaan kompos EFB sebagai pembawa untuk bakteria pengikat nitrogen (NFB) dan bakteria pelarut fosfat (PSB). Campuran (kelembapan 50-60%) antara EFB (gentian yang dicincang pendek) dengan POME (dari kolam anaerobik) serta ditambah dengan mikroorganisma efektif (EM) bagi mempercepatkan pemprosesan kompos EFB. Kompos EFB mencapai fasa 'thermophilic' selepas 4-6 minggu pemprosesan dengan suhu yang konsisten di antara 50-60°C. Selepas 7 minggu, EFB kompos memasuki fasa akhir 'mesophilic' di mana suhu akan menurun kepada 35-40°C pada minggu ke-8 pemprosesan. Kematangan kompos EFB dapat ditunjukkan melalui penurunan nisbah C/N daripada 36 di peringkat

awal pemrosesan kepada 20 selepas 6 minggu pemrosesan. Kompos EFB matang tersebut dikeringkan dan dikisarkan serta diayak untuk mendapatkan kompos EFB halus (1 mm) sebelum digunakan sebagai pembawa bakteria. Kompos EFB halus yang digunakan sebagai pembawa bakteria tersebut mempunyai ciri-ciri seperti berikut (berat kering); pH purata pada 7.5, nisbah C/N - 13.5, kelembapan - 17.4%, bahan organik - 74.5%, jumlah N - 3.06%, P - 0.37%, K - 4.74%, Ca - 3.32% dan Mg - 0.79%. Inokulasi NFB dan PSB seperti *Serratia marcescens* dan *Enterobacter cloacae* ke dalam kompos EFB halus menunjukkan jumlah sel hidup yang tinggi pada  $4.05 \times 10^9$  CFU/g dan  $2.75 \times 10^8$  CFU/g masing-masing pada hari ke-3 selepas inokulasi. Manakala, inokulasi campuran daripada *Burkholderia cenocepacia* dengan *Serratia marcescens* menunjukkan jumlah sel hidup pada  $2.45 \times 10^8$  CFU/g dan  $4.31 \times 10^9$  CFU/g masing-masing. Keputusan ini jelas menunjukkan bahawa kompos EFB adalah berpotensi digunakan sebagai pembawa alternatif kepada bakteria sebelum diaplikasikan ke perladangan kelapa sawit.

*Kata kunci:* EFB, POME, kompos, pembawa, bakteria berfaedah

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## 1.0 INTRODUCTION

Palm oil is the major edible oil in world consumption and it had contributed about 33% of the world total vegetable oils production in year 2010/2011. Malaysia is the second largest palm oil producer in the world which had contributed about 39% of the total world palm oil production in 2010/2011. The total oil palm planted area in Malaysia was estimated about 4.9 million hectares (ha), where Peninsular Malaysia remain as the major planted area with 2.5 million ha, followed by Sabah (1.4 million ha) and Sarawak (0.9 million ha). The average oil palm yield in Malaysia was about 18.03 tones/ha and about 88.3 million tones of oil palm fresh fruit bunches was produced yearly [1].

The palm oil mill normally generated several wastes such as oil palm empty fruit bunches (EFB), palm oil mill effluent (POME), oil palm fruit mesocarp fibres and kernel shell [2]. These wastes are potentially to be recycled to produce synthetic bio-fuel for power generation [3], bio-ethanol [4] and bio-composite product from the cellulose and lignin [5]. Of these, EFB and POME contributed about 22% and 67% respectively of the total waste discharge from the palm oil mill [6]. Therefore, EFB and POME were among the palm oil mill wastes that had extensively been recycled to produce quality compost as organic fertilizer for nutrient input in agricultural activities [7, 8, 9].

Bioconversion of EFB and POME into compost is the current trend of many oil palm mills in Malaysia on waste management with the main objectives to reduce the oil palm mill waste discharge into the river and also returning back the nutrients to the plant nutrient cycle. Composting of organic waste is a better waste management approach where this system is cheaper, environmental friendly, sustainable, promote clean and readily marketable compost product to agriculture sector, able to minimize the greenhouse gases emission and enhances the recycling of material [10].

EFB normally has higher C/N ratio, lower moisture and nitrogen contents as compared to POME [11]. Therefore, mixing of EFB and POME for composting is an ideal approach which could provide better moisture content and adequate nutrients for microbial activities during the degradation process of the organic matter. Meanwhile, mixing of EFB and POME at a ratio of 1 to 3 for composting also directly assisted palm oil mill to achieve nearly zero waste discharge where the waste composition of EFB and POME are around 22% and 67%. Utilization of EFB and POME through composting is capable to manage the waste produced from the mill as well as generating new income source from the commercialization of the compost produced to the agricultural sector. Besides, recent study discovered the existence of NFB in the mature EFB compost and suggested the possibility of using EFB compost as the carrier for bacteria [12]. EFB powder was also studied as potential carrier for beneficial microbes and showed high viable cell count after 30 days of inoculation into the carrier [13]. Carrier can be derived from plant waste materials (compost, farmyard manure, soybean meal, press mud and agricultural waste), soils (peat, coal, clays and inorganic soil) and inert materials (vermiculite, perlite, ground rock phosphate, calcium sulphate and alginate beads) [14]. Unavailability of suitable carrier material can be the major constraint in biofertilizer production which can affect the shelf-life of the microbial activities [15].

Therefore, this study was carried out to evaluate the suitability of using EFB compost as environmental-friendly carrier for the immobilization of NFB and PSB, during the formulation of biofertilizer.

## 2.0 MATERIAL AND METHODS

### 2.1 EFB Composting

The EFB were shredded into short fibres about 7-8 inches in length and stacked to build a 40 tonnes composting windrow with 50 m long, 1.5 m height and 3 m width. About 10 tonnes of POME from anaerobic pond was pumped and drenched on the EFB composting windrow every 4 days until the cumulative volume of 120 tonnes was reached. The mixture was well-mixed by mechanize turner to ensure even distribution of the mixture components, provide aeration for microbial activities and maintaining the moisture content about 50-60%. Solution of Effective Microorganism (EM) was sprayed at 20 L onto the composting windrow in the initial stage to accelerate the composting process. The composting windrow was covered by the air permeable canvas during composting period to avoid process disturbances from the rain water and excessive heat build-up.

### 2.2 Monitoring of Composting Temperature

The temperature of the composting windrow was monitored throughout the 51 days composting process using the composting thermometer. The thermometer probe was inserted into the composting windrow at the depth around 1-1.5 foot and the reading was recorded after around 5 minutes or until the reading was consistent. The temperature of the composting windrow was averaged from 9 samplings points along the composting windrow.

### 2.3 Monitoring of Nutrient Content

The shredded EFB from the composting windrow was collected for nutrient analysis during the composting period. The samples were collected from three consistent sampling points at 10 m, 25 m and 40 m along the composting windrow. The samples were collected at 0, 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup>, 5<sup>th</sup>, 6<sup>th</sup>, 7<sup>th</sup> and 8<sup>th</sup> weeks during composting. The parameters of the nutrient analysis included total nitrogen (N), total carbon (C), carbon to nitrogen (C/N) ratio, phosphorous (P<sub>2</sub>O<sub>5</sub>), potassium (K<sub>2</sub>O) and magnesium (MgO).

### 2.4 Carrier Preparation

The mature EFB compost was sun-dried for 2 weeks and ground into smaller particle size by using hammer-mill. The grinded EFB compost was further screened through 1 mm sieve to get the fine powder form for biofertilizer inoculation [16]. The fine EFB compost was then oven-dried at 70°C for 48 hours and placed in airtight polyethylene bags for sterilization in autoclave at 121°C for 20 min in 3 consecutive days before used for bacteria inoculation [17].

### 2.5 Bacteria Inoculation Into Carrier

About 70 ml of the pure culture of *Serratia marcescens*, *Enterobacter cloacae* and mixed culture of *Burkholderia cenocepacia* with *Serratia marcescens* were aseptically injected by sterilized syringes into the polyethylene bags that contain 130 g of sterilized carrier (fine EFB compost) to obtain the moisture content about 35% [18]. The punctured area was wiped with 70% alcohol and the punctured hole was covered with cellophane tape. The bags were kneaded thoroughly to ensure well mixed of the bacteria culture with fine EFB compost [19].

### 2.6 Viable Cell Count of Inoculants

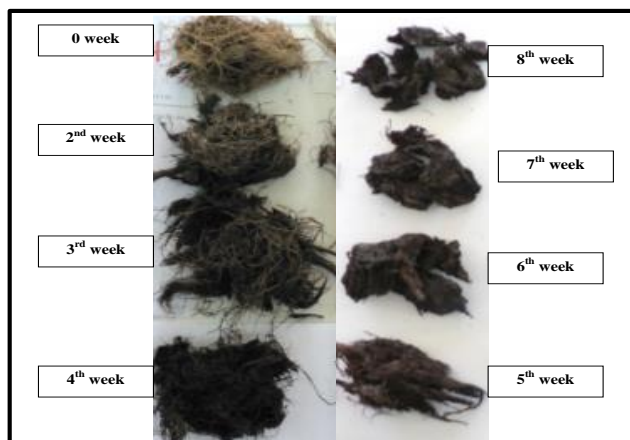
The cell concentration (CFU/g) of the inoculants in the carrier (fine EFB compost) was determined at 3<sup>rd</sup> day of inoculation. A total of 1 g of bacteria inoculated compost was sampled and placed into test tubes containing 9 ml of sterilized distilled water. It was shaken thoroughly to ensure complete separation of bacterial from the carrier. The solution was diluted in a 10-fold serial dilution and 0.1 ml of each solution was plated onto nutrient agar with three replications. All the plates were incubated at room temperature for 24 hours and the colony formed was counted to determine the viable bacteria cell count per gram of carrier (CFU/g) [19].

## 3.0 RESULTS AND DISCUSSION

### 3.1 Physical Characteristic

EFB fibres remained as coarse fibre until the 4<sup>th</sup> weeks of composting but the colour turned from light brown to black. After 6 weeks of composting, the EFB fibres became soft and short which can be tear manually, which indirectly indicate that the EFB compost was started to enter the mature phase. The EFB compost showed full maturity after 8 weeks of composting where the EFB fibres were in lumps of short fibres when wet, dark brown and can be easily shredded manually (Figure 1).

The EFB fibres normally contain about 52.5% of cellulose and 17.1% lignin, therefore, the composting of EFB fibres normally need longer period to reach the maturity stage as compared to other materials. The mature EFB compost normally can be produced after 8 weeks of composting when the final compost product exhibited in blackish colour and earthy smell [11]. The dark colour of the compost is caused by the conjugation of C=O group on quinones and ketones of the humic substances [20].



**Figure 1** The physical changes of shredded EFB during composting

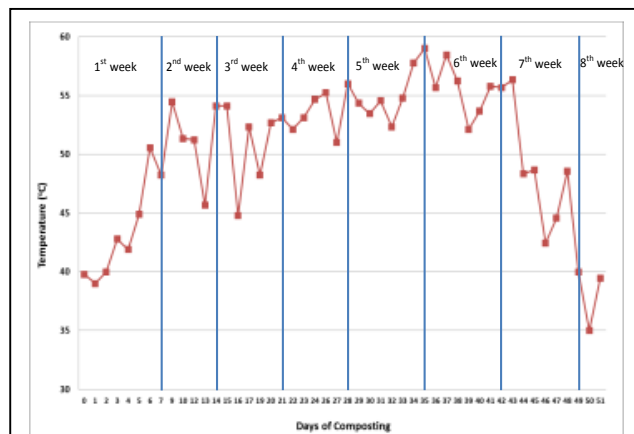
### 3.2 The Temperature Profile

The temperature during the initial stage of EFB composting was 40°C and drastically increased to 50°C during the 1<sup>st</sup> week of composting (Figure 2). Normally the lag phase of the temperature on EFB composting was very short and the drastic increase of the temperature of the composting windrow indicates active composting process through intensive microbial activity [21]. This is reflected from the high degradation rate of organic matter during the early stage of EFB composting [22]. The EFB composting was in early mesophilic stage during the 2-3 weeks after composting, where the compost temperature was fluctuated between 45-55°C.

The temperature was stable around 50-55°C from 4-5 weeks after composting and reached the highest temperature around 55-60°C after 6 weeks of composting. Normally, the thermophilic phase of composting can last for 2 weeks and the temperature was high and able to increase until 58°C [23]. The microbial colonization activity in EFB composting was active during 4<sup>th</sup> to 6<sup>th</sup> weeks with continuous high temperature which also indicated the composting occurring at the thermophilic stage. The temperature range of 52-60°C is the most favourable condition for microbial activity in composting and the pathogenic microorganism can be killed when the composting temperature rises above 55°C. However, the microbial activity will decline rapidly when the composting temperature surpassed the various optimum thermophiles temperature above 62°C [24].

The composting process enter the mesophilic phase after 7 weeks of composting where the temperature was starting to decline to around 45°C (Figure 2). The declined of temperature was due to the reduction of microbial activity which can also be associated with the depletion of degradable organic matter and the recolonization of the compost by mesophilic microorganism to degrade the remaining sugar, cellulose and hemicellulose [24]. The

temperature dropped to 35-40°C after 8 week of composting indicated the compost was entering into mature stage (Figure 2), where the bio-oxidation of the composting was considered completed [25].



**Figure 2** The temperature changes of EFB composting

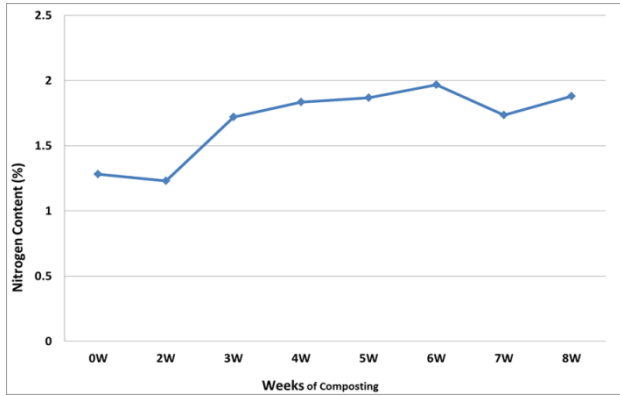
### 3.3 The Nutrient Changes During Composting

The N content of EFB fibre was around 1.2% during the initial stage of composting which increased to about 1.9% after 8 weeks of composting (Figure 3). Other researcher has also reported on the increase in N content during EFB composting (2.05%) after 12 weeks of composting [22]. The increase trend of N content in the compost showed the mineralization of organic compounds with formation of ammonium during the active phase of composting [24]. The increase of N content in the mature compost was also due to the concentration effect, where the strong degradation of organic compound caused reduction of compost mass weight [25].

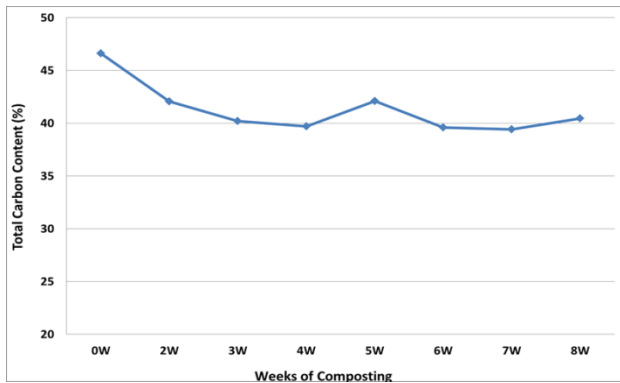
The initial high C content of 47% gradually decreased to 40% after 3<sup>rd</sup> weeks of composting which then fluctuates between 39-41% until the end of composting period (Figure 4). During the composting process, the organic compounds are degraded by microorganisms to carbon dioxide gas (CO<sub>2</sub>) and ammonia (NH<sub>3</sub>) with the consumption of O<sub>2</sub> under aerobic condition [24]. High initial C/N ratio of 36 was recorded during the early stage of composting process. After 4 weeks of composting, the C/N ratio was reduced to 22 and fluctuates between 20-22 until the end of the composting period (Figure 5). The compost is considered to reach maturity where the C/N ratio is less than 25 [26].

Potassium (K<sub>2</sub>O) concentration increased from <2% (at the initial stage) to around 3.5-4.0% after 5 weeks of composting (Figure 6). Similar trend was also observed for phosphorous (P<sub>2</sub>O<sub>5</sub>) with slight increased from 0.20% to 0.55% after 8 weeks of composting (Figure 7). The EFB fibres showed low magnesium (MgO) content of 0.35% in the initial stage. However, the MgO content was increased to around 0.6-0.7% after 4 weeks of composting (Figure 8). The nutrient

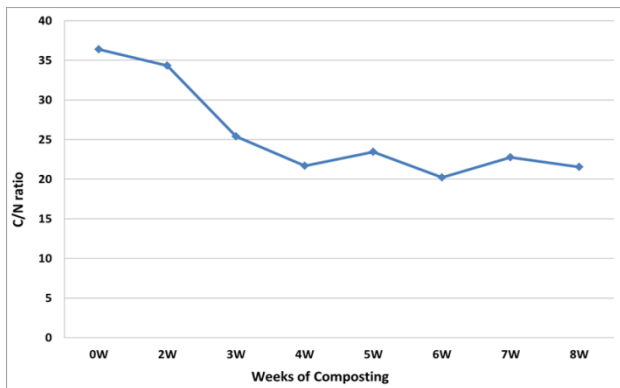
analysis of the composting indicated that the nutrient content was able to improve at minimum 4-5 weeks after composting, where the increase of these minerals was due to the dry weight losses of the organic compound weight [24]. These nutrient contents were comparable with the similar composting system which had recorded 0.69% of  $P_2O_5$ , 1.16% of  $K_2O$  and 0.28% of  $MgO$  [21].



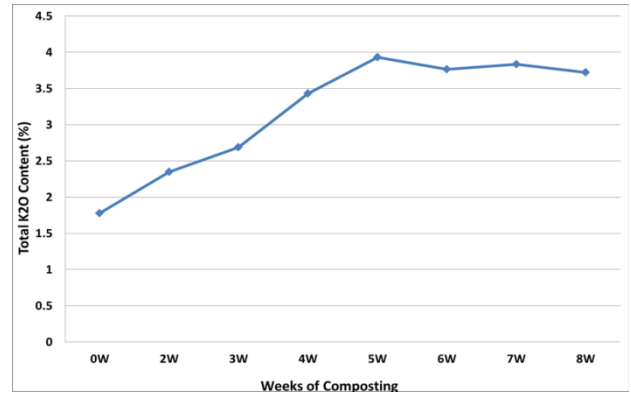
**Figure 3** Changes of nitrogen content of EFB compost during composting



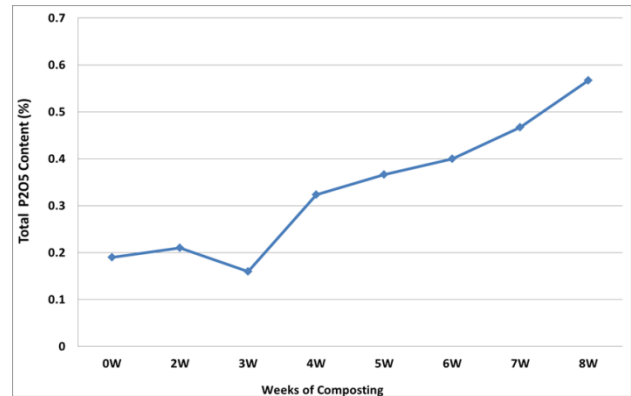
**Figure 4** Changes of carbon content of EFB compost during composting



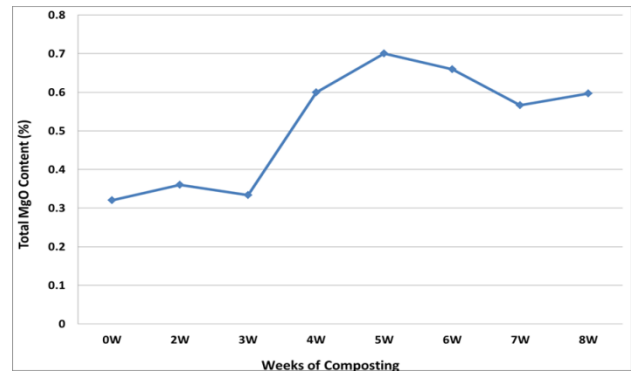
**Figure 5** Changes of C/N ratio of EFB compost during composting



**Figure 6** Changes of  $K_2O$  content of EFB compost during composting



**Figure 7** Changes of  $P_2O_5$  content of EFB compost during composting



**Figure 8** Changes of  $MgO$  content of EFB compost during composting

### 3.4 Nutrient Content of Carrier

Chemical analysis of the fine, grounded and sieved EFB compost (to remove the coarse undecomposed fibres and fine palm kernel shells) showed that the carrier is rich in mineral contents with values of 3.06% of nitrogen, 4.74% of potassium, 3.32% of calcium, 0.79% of magnesium and 0.37% of phosphorous. The pH is 7.5, moisture content is low at 17.4% and the C/N ratio was 13.5 which indicated the EFB compost (carrier) was mature.

### 3.5 Viable Cell Count of Inoculated Bacterial

The viable cell count of *Serratia marcescens* and *Enterobacter cloacae* pure culture in the fine EFB compost carrier at third day after inoculation was recorded at  $4.05 \times 10^9$  CFU/g and  $2.75 \times 10^8$  CFU/g respectively. Meanwhile, the mixed culture of *Burkholderia cenocepacia* with *Serratia marcescens* recorded  $2.45 \times 10^8$  CFU/g and  $4.31 \times 10^9$  CFU/g respectively. The high viable cell count of the bacterial inoculants in the fine EFB compost indicated the suitability of this material as carrier for bacterial inoculation.

## 4.0 CONCLUSION

This study demonstrated the feasibility of using fine EFB compost as carrier for bacterial inoculation during the formulation of biofertilizer. This finding is important as it provides some insight into the potential use of alternative material to be used as carrier for useful microbes required for biofertilizer formulation. Nevertheless, more studies need to be carried out before the commercial application.

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