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# Effective composting of empty fruit bunches using potential *Trichoderma* strains

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#### ABSTRACT

Oil palm fibres are easy to degrade, eco-friendly in nature and once composted, they can be categorized under nutrient-enriched biocompost. Biocompost is not only a good biofertilizer but also a good biocontrol agent against soil-borne pathogens. In this research, experimental works on the composting of empty fruit bunches (EFB) from the oil palm industry were conducted using two potential *Trichoderma* strains. Analysis of pH initially found the soils to be slightly acidic. However, after composting, the soils were found to be alkaline. *Trichoderma* propagules increased by 72% in the soils compared to other fungi. Soil electrical conductivity was found to be  $50.40 \ \mu$ S/cm for compost A,  $42.10 \ \mu$ S/cm for compost B at 2.79, and then the control at 1.55. The highest percentages of nitrogen (N), phosphorus (P), and potassium (K) were found in compost A (0.91:2.13:6.68), which was followed by compost B (0.46:0.83:5.85) and then the control (0.32:0.26:5.76). Thus, the biocomposting of oil palm fibres shows great potential for enhancing soil micronutrient, plant growth performance, and crop yield production. (© 2016 The Author(s). Published by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

#### 1. Introduction

The oil palm industry has been growing exponentially over the past years. This has led to increasing amounts of waste products from the oil palm industry, especially empty fruit bunches (EFB). EFB is produced in large quantities in localized areas. Recycling EFB by conversion into usable by products has been found to be the most suitable way to reduce this raw waste material. Currently, there are a few solutions available including the use of EFB as fuel to generate steam at mills [1] and the composting of EFB [2,3] for use as mulch at plantations [4]. Most of these solutions are not very effective and are costly.

Using traditional composting methods, EFB may take months or years to reach maturation. The high C:N ratio and the presence of polymers such as cellulose and lignin in EFB act as natural barriers against natural biodegradation [5]. The addition of specific microorganisms to convert of EFB into compost may help reduce composting time. The compost produced from this process usually acts as a good nutrient-enriched biofertilizer, which at the same time helps to reduce the use of chemical fertilizers. The utilization of organic wastes as biofertilizer has ability to increase crop productivity, improve soil health and reduce waste disposal problems [5].

According to Goyal et al. [6], fungi are actively involved in the decomposition of cellulose, hemicelluloses, and lignin present in organic matter. The composting process can be accelerated to as take as little as one month by the inoculation of cellulolytic fungi such as *Aspergillus* and *Trichoderma* [7–9] due to their ability to produce enzymes that can degrade cellulose, hemicelluloses and lignin [10,11]. The *Trichoderma* species has proven to be able to effectively degrade organic matter. The *Trichoderma* species has also been used as a biological control agent as it competes for space and nutrients, and produces toxins against phytopathogenic fungi. The *Trichoderma* species thus enhances plant growth performance [12,13].

In 2005, Heraux et al. [14] reported that the mixing of *T. virens* with composted chicken manure and rye cover crops showed significant control of weeds. Later, Gaind and Nain [5] found that *T. reesei* is able to improve soil biochemical properties. Studies in 2010 by Pandya and Saraf [13] reported that the genus *Trichoderma* acts as an important biocontrol agent against several phytopathogenic fungi. Dayana Amira et al. [9] reported that the addition of

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*T. virens* to empty fruit bunches and palm oil mill effluents decreased the composting period due to the higher levels of enzymatic activity.

In this study, EFB was composted using two potential *Trichoderma* strains and the resulting composts were added to soil in field experiments. The objective of this study was to examine the physio-chemical changes such as temperature, pH, electrical conductivity (EC), C:N ratio and percentages of N, P, and K during the composting process and in field experiments.

#### 2. Materials and methodology

#### 2.1. Selection of trichoderma isolates based on enzymatic activity

The procedure for isolating, screening and measuring enzymatic activity of *Trichoderma* isolates has been previously described by Shafawati et al. [10]. Based on the enzymatic activity results, the two best potential strains were selected and used in the composting process.

#### 2.2. Preparation of compost and the composting process

EFB was used as the composting substrate. The EFB was collected from Sahabat palm oil industry, Kinabatangan, Lahad Datu, Sabah Malayisa. The dry EFB was soaked in water overnight, and then placed inside a plastic container with holes for at least 3 h in order to remove excess water while still ensuring that the EFB was moist enough for the composting process. About 350 g of EFB was packed into clear white plastic bags and 5% sugar was added. The EFB was then autoclaved at 121 °C/1.05 kg/m<sup>2</sup> for 15 min. After autoclaving all plastic bags were kept under laminar flow to reduce relatively low temperatures. After that, two types of composts (Compost A and Compost B) were made by using two effective Trichoderma strains. Strain SICCI was used for compost A and strain 11B was used for compost B. The strains were mixed with the EFB in the plastic bags, which were the tightly closed with rubber bands and inoculated for 30 days. Thus, this process occurred under anaerobic conditions.

Chilli plants were grown with three types of soil treatments, compost A, compost B and a control without compost. Each of the treatment plots were planted with five chilli seedlings with five replications. All chilli seedlings were originally from the same seed, planted with same volume of soil, in the same polybag and provided with the same volume of water. No additional fertilizers were added to the treatment plots, except for the compost. Three repetitions of the whole experimental work were conducted.

#### 2.3. Trichoderma colony forming unit (CFU) estimation

Soil samples were taken before and after applying compost to the treatment plots (polybag), and then mixed homogenously. Ten grams of soil was weighed out and 100 mL sterilized distilled water (121 °C/1.05 kg/m<sup>2</sup> for 15 min) was added before shaking with the orbital shaker for 30 min at 210 rpm. After that, 1 mL of the solution was added to 9 mL water for the first  $(10^{-1})$  dilution. The serial dilutions of 10<sup>-3</sup> and 10<sup>-5</sup> were used for Colony Forming Unit (CFU) measurements. Approximately 1 mL of soil solution was pipetted out and seeded into each Petri dish followed by pouring 9 mL of sterilized Trichoderma Selective Medium (TSM) [0.20 g of MgSO<sub>4</sub>·7H<sub>2</sub>O, 0.90 g of K<sub>2</sub>HPO<sub>4</sub>, 0.15 g of KCl, 1.0 g of NH<sub>4</sub>NO<sub>3</sub>, 3.0 g of glucose, 0.15 g of Rose Bengal, 20.0 g of agar (Difco, USA) and 1000 mL of distilled water] as reported by Elad et al. [15]. Each Petri dish was swirled manually before being allowed to solidify and then incubated at  $28 \pm 2$  °C for 7 days. Fungal colonies could be seen as small whitish growth spots. Each single colony appearance

was scored as a Colony Forming Unit (CFU), checked and reisolated onto Potato Dextrose Agar (PDA) (Oxoid, UK).

2.4. Measurement of the biological changes in temperature, pH and electrical conductivity

The temperature was measured every week throughout the composting period based on the procedures in Erwan et al. [16]. The temperature was measured at three different points, namely, the top, middle and lower parts of the compost bags using a thermometer. The thermometer was inserted for five minutes inside the compost to take the readings.

The pH of the compost was measured weekly during the composting period. About 10 g of compost was placed in a 500 mL beaker and 50 mL of sterilized distilled water was added. This was then placed in a shaker for 30 min [17], and then the pH was measured.

Electrical conductivity (EC) was also measured every week. Ten grams of each soil sample was added to 100 mL of distilled water (1:10 v/v), placed in a shaker for 30 min, and then kept for 24 h. Salinity was determined using an Electrical Conductivity Meter and expressed as dS/m [16].

#### 2.5. Measurement and chemical analysis of the C:N ratio and NPK

From each of the soil samples, 15 g was taken and put inside a furnace for 5 h. The temperature for the first hour was 300 °C and for the remaining 4 h was 500 °C. The samples were then left overnight before the ashes were weighed. The percentage of carbon content was calculated using the following formula [16]:

$$\%$$
ofCarbon =  $\frac{(Sample of dry weight) - (Weight of ash)}{Sample dryweight}$ X100

Nitrogen content was determined using the digestion method. Each of the soil samples were dried for 72 h at 60 °C and ground to pass through a 20 mm mesh sieve. About 0.25 g of soil sample was then put into a digestion flask with 5 mL of concentrated sulphuric acid ( $H_2SO_4$ ) and digested at 200 °C for 30 min. Then the temperature was increased to 360 °C for 1 h, after which 10 mL of hydrogen peroxide ( $H_2O_2$ ) was added until the reaction was completed. The final solution was made up with 100 mL distilled water. Nitrogen was determined using the Autoanalyser (System 4, Chemlab). The percentage of nitrogen content was calculated using the following formula [16]:

% Nitrogen = 
$$\frac{(\text{ppm x 400 x dilution})}{10^6}$$
X 100

Approximately 0.50 g of finely ground samples were added to 3 mL of 37% HCl and 1 mL of 68% HNO<sub>3</sub> in 50 mL Kjedahl flask and heated at 110 °C in a digestion block until 1 mL of the sample solution remained. If the sample colour had not changed to white, 3 mL HCl and 1 mL HNO<sub>3</sub> were added again. After the sample cooled down, 10 mL of 1.2% HNO<sub>3</sub> was added (v/v) and digestion was allowed to continue for 30 min at 80 °C. Distilled water was added to cool the sample solution and to ensure the sample solution maintained a volume of 20 mL while heated at 80 °C for 30 min. After cooling, distilled water was added to make up a final volume of 20 mL. The solution was then used to determine the values of P and K micronutrients by using an Atomic Absorption Spectrophotometer [16].

The C:N ratio was determined using the following formula [16]:

Table 1   Percentage of Trichoderma and Non-Trichoderma found in soil samples during 15 weeks of field experiment.									
Week	Control	Compost A							
	T CELL Non T CELL	T CEU Non T CEU							

WCCK	Control		compose A		Compost b		Total
	T. CFU	Non T. CFU	T. CFU	Non T. CFU	T. CFU	Non T. CFU	
0	1	0	-	-	-	-	
2	7	2	8	1	1	1	
4	2	1	15	0	14	0	
6	6	3	7	3	4	3	
8	7	2	5	6	7	4	
10	2	0	2	3	8	3	
12	1	1	2	3	2	2	
14	0	1	5	2	4	1	
15	1	1	7	1	3	1	
Total	27	11	51	19	43	15	166
Trichoderma sp. CFU			121	Non Trichoderma sp. CFU			45
Percentage of Trichoderma found in soil samples (%	6)		72.9	Percentage of non-Trichoderma found in soil samples (%)			27.1

#### 2.6. Statistical analysis

All the experiments were conducted using a completely randomized design (CRD) with three replications. Results were subjected to analysis of variance (ANOVA) and tested for significance using Least Significance Difference (LSD) in PC-SAS version 9.0.

#### 3. Results

#### 3.1. Trichoderma colony forming unit (CFU) estimation

Table 1 shows the estimated *Trichoderma* CFU for the 15 weeks of the field experiment using chilli plants. In the first week, very few *Trichoderma* colonies were found. During the second week, the number of *Trichoderma* colonies increased and in week 4 the highest number of *Trichoderma* colonies were found in soil samples with a total of 29 colonies for both compost A and compost B. The soils treated with compost were found to have fewer non-*Trichoderma* colonies as compared to the control soil. However, over the next few weeks the *Trichoderma* and non-*Trichoderma* colonies gradually decreased in number until the end of the field experiment. The total *Trichoderma* CFU recorded during field experiment amounted to about 72.9%.

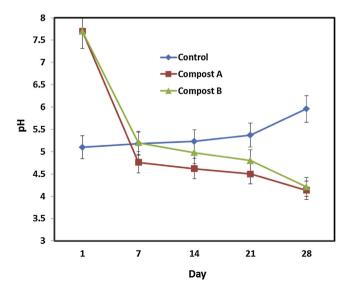


Fig. 1. pH analysis during four weeks composting process.

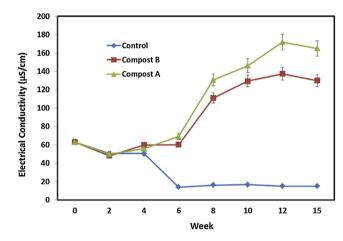


Fig. 2. Measurements of electrical conductivity during field experiment.

#### 3.2. Analysis of temperature

The compost temperature measured for four weeks during the incubation process was found to stay at  $24^{\circ}$ C. No significant changes in temperature were recorded. The compost temperature was maintained at  $24^{\circ}$ C till the end of the composting process.

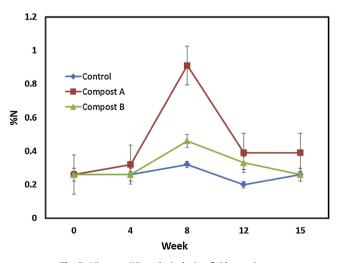


Fig. 3. Nitrogen (N) analysis during field experiment.

Total

Compost B

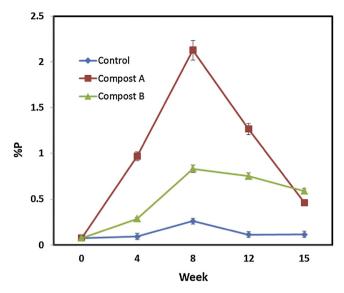


Fig. 4. Phosphorus (P) analysis during field experiment.

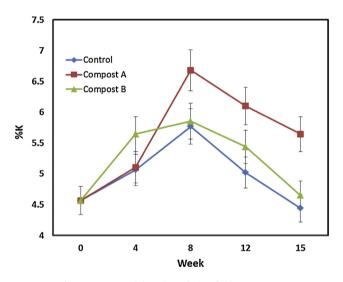


Fig. 5. Potassium (K) analysis during field experiment.

#### Table 2

Changes in C:N ratio of EFB composts during field experiment.

Treatment	Week of field experiment						
	Initial	Week 4	Week 8	Week 12	Week 15		
Compost A	10.42	10.89	6.48	4.01	3.33		
Compost B	10.42	9.47	6.03	3.87	2.79		
Control	10.42	7.88	5.00	3.04	1.55		

#### Table 3

Comparison of composting periods of different composts.

#### 3.3. Changes in pH

The pH values of the composts for all treatments are presented in Fig. 1. All treatments were found to be alkaline (pH 7.7) at the beginning of the compost process. Over the four weeks of composting, the composts became more acidic. At the end of the composting process, the pH value for compost A reached 4.1 and compost B reached 4.2.

#### 3.4. Electrical conductivity (EC)

The EC value significantly differed for each composting treatment. Initially, the EC value was  $62.96 \,\mu$ S/cm. The highest EC value was recorded by compost A which was 171.97  $\mu$ S/cm during week 12 of the field experiment while compost B and the control showed lower EC values on the same week, respectively 137.46  $\mu$ S/cm and 15  $\mu$ S/cm (Fig. 2).

#### 3.5. Chemical analysis of NPK and C:N ratio

#### 3.5.1. Nitrogen analysis

Fig. 3 shows the variation in nitrogen levels between the three types of field experiments conducted over 15 weeks. Compost A obtained the highest percentage of nitrogen (N) with a value of 0.91 in week 8 of the field experiment when the N value of compost B was 0.46 and the control was 0.32.

#### 3.5.2. Phosphorus analysis

The variation in phosphorus levels over the duration of the field experiment between the three types of field experiments are presented in Fig. 4. A comparison of the phosphorus (P) values at week 8 found that compost A reached the highest P value of 2.13 while compost B and the control had lower P values at 0.83 and 0.26, respectively.

#### 3.5.3. Potassium analysis

A comparison of the potassium (K) levels in the experimental soils found that K levels were highest in week 8 of the field experiment with compost A having the highest K level with a value of 6.68, compared to compost B at 5.85 and the control at 5.76 (Fig. 5).

#### 3.5.4. Carbon and nitrogen (C/N) ratio

The changes in the C:N ratio for all the compost treatments during the field experiment are shown in Table 2. The highest C:N ratio was obtained by compost A at 3.33, followed by compost B at 2.79 and the control at 1.55 (Table 2).

#### 4. Discussions

The decomposition of the EFB into mature compost after being inoculated with the *Trichoderma* species took four weeks in this study (Table 3). This can be compared to a study done by Dayana Amira et al. [9] which found that the addition of fungi to compost

Materials/microorganism	Time to complete composting process	Reference
Selected substrates + POME + EFB + Wheat flour + P. chrysosporium + T. harzianum + A. niger	2 months	[20]
EFB + T. virens	28 days/4 weeks	[9]
Different combination and ratio of Oil Palm Frond compost + chicken manure + rice bran	21 days	[16]
Rice straw + EFB + T.harzianum	12 weeks	[21]
Vegetable and animal waste + peat soil + <i>T. asperellum</i>	17 days	[22]
Olive pomace + P. chrysosporium + T. harzianum	50 days	[19]
EFB + Trichoderma sp.	28 days	This study

reduces the decomposition time of three lignocellulosic components, cellulose, hemicelluloses, and lignin. By day 22 these lignocellulosic components were significantly decomposed. When compared to the control, the addition of fungi to the compost reduced maturation time by 60.9%. A similar study done by Singh and Sharma [18] and Haddadin et al. [19] reported rapid degradation of compost when using fungi as compared to compost without bioinoculants which had longer degradation times.

Gaind and Nain [5] reported that a single inoculation of *T. reesei* showed positive effects on many types of soil parameters. Hydrophobin, which is produced by the *Trichoderma* species, is involved in mycelium attachment to the surface, alteration of biotic or abiotic surface properties and lowering of water tension. Due to these properties, fungi may play a strong functional role in soil aggregation and soil structure change, which may affect microbial activity [23].

A further benefit of the addition of fungi to compost is the production of digestive enzymes, which can be seen from the higher levels of xylanase and cellulase activity. This is in agreement with Gaind and Nain [5], who reported that an increase in enzymatic activity was found with a trend towards effective degradation and maturity of compost. They also found that *T. reesei* showed higher levels of carboxymethyl cellulose, cellobiase and xylanase activity as compared to *Aspergillus awamori* and *Phanerochaete chrysosporium*.

Organic substances with high lignin and cellulose content have low microbial degradability [24]. Therefore, the addition of accelerating fungi such as the *Trichoderma* species promotes the degradation process as well as improves physical properties such as water holding capacity, aeration and texture.

#### 4.1. Trichoderma colony forming unit (CFU) estimation

The CFU estimation technique was used to calculate the number of *Trichoderma* found in the soil. After mixing the soil and compost, soil samples were taken to measure the *Trichoderma* CFU. Based on the results, there were very few *Trichoderma* colonies in the initial week. The number of *Trichoderma* CFU increased in the second week and highest amount of CFU was found in week 4. In the following weeks the *Trichoderma* CFU dropped, although they could still be found until week 15. During the field experiment, the number of *Trichoderma* CFU was always higher compared to the non-*Trichoderma* CFU. At the end of the experiment, a total of 121 *Trichoderma* colonies. These results show that mixing the soils with compost increased the number of *Trichoderma* colonies.

The *Trichoderma* require a few weeks to adapt after the addition of the enriched compost to the soil. As can be seen from the CFU estimation results, the highest numbers of *Trichoderma* colonies were found in week 4. The effectiveness of using compost for disease control particularly against fungal pathogens has been studied extensively by several authors [21,25–27]. Studies done by Siddiqui et al. [21] found that the use of *T. harzianum* in compost made from rice straw (RST) and empty fruit bunches (EFB) was able to control the disease severity caused by *Choanephora cucurbitarum* wet rot in okra production by 85.04%. Although the number of *Trichoderma* colonies gradually decreased towards the end of experiment, *Trichoderma* colonies were still found in higher numbers than non-*Trichoderma* colonies.

Sariah et al. [28] stated in their report that the distribution of *Trichoderma* species decreased with the depth of soil. It was found in this study that when the soil is mulched with empty fruit bunches, there is a significant increase in the isolation frequency and the depth profile for *Trichoderma*.

#### 4.2. Analysis of temperature

There were no significant variations in temperature during the four weeks of composting, perhaps because the compost was kept in small packages made of plastic. In a similar statement by Al-Khawaldeh [29], it was stated that where the compost piles are too small, a rapid loss of heat can be expected. However, the experimental observation of the compost found that the colour of the compost become greener at the end of the composting process, which was different from the initial colour which was brownish. This was due to the massive growth of the *Trichoderma* strains on the surface of the compost which created the green impression.

#### 4.3. Changes in pH

The pH value indicates the acidity or alkalinity of a substance. Soil pH is defined as the negative logarithm of the hydrogen ion concentration. Soil pH will turn acidic when the amount of hydrogen ions in the soil increases. In this experiment, the pH of the soil was found to be around 5.2–5.5.

The final compost was found to be acidic compared to the intial pH recorded. The effect of the inoculums (the *Trichoderma* strains) on the rate of organic matter degradation leads to large amounts of ammonium that could decrease the compost pH as a consequence of nitrification [5]. In a similar work reported by Gaind and Nain [5], pH dropped significantly when the compost was mixed with *T. reesei.* 

The low pH found in this study was caused by the presence of *Trichoderma*. Kirk et al. [30], Reid [31] and Haddadin et al. [19] have reported that the optimal pH for cellulose and lignin degradation by fungi and the activities of their enzymes is 4. Similar results have been reported by Zeilinger et al. [32], where the optimal pH was found to be 4 for the degradation of cellulose and enzyme activity of *T. harzianum*. In this study, the pH at the end of the composting process was found to be 4.0. This is in agreement with study done by Sariah et al. [28], where the distribution of the *Trichoderma* species was favoured by acidic soil.

#### 4.4. Electrical conductivity (EC)

Soil electrical conductivity is an important indicator of soil properties that affect crop productivity and plant nutrient availability, including soil texture, cation exchange capacity (CEC), organic matter level, salinity, subsoil characteristics and activity of soil microorganisms which influence key soil processes [33]. Measuring EC levels can give an indication of the presence of ions or salt compounds such as nitrates, potassium, sodium, chloride, sulphate and ammonia. Determining the EC of soil can help in estimating the amount of nitrogen (N) available for plants.

During the field experiment, the soil treated with compost A was found to have the highest EC value in week 12 at 171.97  $\mu$ S/cm. This value decreased to 165.00  $\mu$ S/cm at the end of the field experiment. The EC values of the soil treated with compost B and the control soil were lower than compost A (Fig. 2). These results show better EC values compared to the studies done by Haddadin et al. [19] who reported that the use of *T. harzianum* and *P. chrysosporium* resulted in EC values of only 11.30  $\mu$ S/cm, while their control was 10.60  $\mu$ S/cm, which are relatively low EC values. Higher values of EC (>5.00 dS/cm) might cause dehydration and death of plants [34].

#### 4.5. C:N ratio

Compost A had the highest C:N ratio at the end of the field experiment with a value of 3.33, followed by compost B (2.79) and

#### Table 4

Comparison of C:N ratio of different composts during composting process.

Materials/microorganism	C/N ratio	Reference
EFB + fresh POME	15	[40]
Different combination and ratio of Oil Palm Frond compost + chicken manure + rice bran	21.34	[16]
Selected substrates + POME + EFB + Wheat flour + P. chrysosporium + T. harzianum + A. niger	17	[20]
Olive pomace + P. chrysosporium + T. harzianum	59.30	[19]
EFB + Trichoderma sp.	3.33	This study

#### Table 5

Comparison of macronutrient content of different composts.

Materials/microorganism	N (%)	P (%)	K (%)	Reference
Wheat Straw + T. harzianum + A. Niger + Azotobacter chrococcum	0.98	0.19	0.55	[18]
Olive pomace + T. harzianum + Phanerochaetechrysosporium	1.60	1.10	19.80	[19]
Empty fruit bunches (EFB) and palm oil mill effluent + T. harzianum	1.30	0.50	0.65	[42]
Rice straw + okara + vinasse + buffalo manure + rock phosphate	1.94	1.95	0.95	[43]
EFB + POME + chicken dung + T. virens	1.31	0.54	0.65	[9]
Different combination and ratio of Oil Palm Frond compost + chicken manure + rice bran	2.33	2.02	1.80	[16]
EFB + Trichoderma sp. (Compost A)	0.91	2.13	6.68	This study

the control (1.55). Nagasaki et al. [35] have proposed that the desirable C:N ratio for composting should be within the range of 16 to 21. However, the C:N ratio in this study was found to be lower than other studies (Tables 2 and 4). Marstop [36] identified soluble carbohydrates and amino acids as the major sources of carbon (C) for the microorganisms involved in the decomposition of plant residues. Bernal et al. [37] in their study stated that a high C:N ratio is frequently found with organic waste decomposition, so it is widely accepted that a high substrate C:N ratio implies a low mineralization rate due to N deficiency.

According to Iglesias-Jimenez and Perez-Garcia [38], C:N ratios lower than 12 for municipal waste compost indicate a good degree of maturity. In well humid soils the C:N ratio can be close to 10 and the addition of materials with a C:N ratio below 15 to the soil may not alter the microbiological equilibrium of the soil [39]. This type of compost is more suitable for use in the agricultural industry.

#### 4.6. Macronutrients analysis

Compost A was found to reach the highest values for nitrogen (N), phosphorus (P) and potassium (K) with the values of 0.91: 2.13: 6.68 in week 8 as compared to compost B and the control (Figs. 3–5). The macronutrient amounts were found to be highest in week 8, after which they decreased until the field experiment ended in week 15. Erwan et al. [16] found that the percentage of C and N affect the macronutrient levels in compost. Chabbey [41] has proposed that the high values for these macronutrients might be due to the reduction of organic content during the composting process (decomposition).

The high nitrogen content detected in compost A and B as compared to control is due to the increased decomposition of organic matter by the fungal *Trichoderma* strains [44]. This is in agreement with Gaind and Nain [5] who in their study on the effect of mixed fungal inoculants found nitrogen content increased significantly compared to single inoculation. In addition, Young et al. [45] have explained high nitrogen content as being governed by the initial nitrogen content of the raw materials. The increasing concentration of nitrate-nitrogen (NO<sub>3</sub><sup>-</sup>N) at the end of the composting process increased the total nitrogen content. However, ammonium nitrate (NH<sub>4</sub><sup>-</sup>N) was high in the early stages of composting but decreased gradually as the compost matured.

The phosphorus content was high in both compost A and compost B treated soils as they reached full maturation. This is because phosphorus is not lost during the composting process to volatilization or lixiviation. The concentration of phosphorus increases as composting proceeds [45]. Gaind and Nain [5] have suggested that the addition of fungi may increase phosphatase activity. Similarly, the potassium values of both composts reached high levels compared to the initial values. This is due to the release of  $PO_4^-$  ions from humic colloids into the system [44]. The increased percentage of phosphorus and potassium in the treated soils is due to the presence of the fungal Trichoderma strains, which play an important role in increasing P and K content during the composting process [9]. Table 5 shows the macronutrient content of compost from several different studies using various raw materials and microorganisms. The percentage of nitrogen varied but was always higher in the other studies. The percentage of phosphorus and potassium observed in this study was low compared to most of the other studies. These variations in NPK content may be due to several factors such as the composition and type of raw materials, and the microorganisms used in the compost.

#### 5. Conclusion

The outcome of this study is the benefit of being able to convert waste materials into a valuable by product within a short period of time. The EFB composting process required only 30 days. The compost obtained provides a high source of micronutrients (N, P, and K) and improves soil acidity and electrical conductivity. The composting materials are easy to degrade and eco-friendly in nature and the resultant compost can be categorized under nutrient-enriched biocompost. This process of recycling transforms the disposable waste of the palm oil industry into a valuable product for agriculture on an economically viable basis.

#### **Conflicts of interest**

The authors declare no conflict of interest the above title manuscript.

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#### References

- A.N. Ma, S.N. Chean, M.C. Chow, et al., Current status of palm oil processing waste management, in: Yeoh (Ed.), Waste Management in Malaysia, Current Status and Prospects for Biodegradation, 1993, pp. 111–136.
- [2] S.N. Shafawati, S. Siddiquee, Composting of oil palm fibres using *Trichoderma* as the biological control agent: a review, Int. Biodeterior. Biodegrad. 85 (2013) 243–253.
- [3] M.A. Damanhuri, Hands on experience in the production of empty fruit bunches (EFB) compost, Paper Presented at the CETDEM Malaysian Organic Farming Seminar, Petaling Jaya, Selangor, Malaysia, 1998.
- [4] N. Mohammad, M.Z. Alam, N.A. Kabbashi, A. Ahsan, Effective composting of oil palm industrial waste by filamentous fungi: a review, Resour. Conserv. Recy. 58 (2012) 69–78.
- [5] S. Gaind, L. Nain, Chemical and biological properties of wheat soil in response to paddy straw incorporation and its biodegradation by fungal inoculants, Biodegradation 18 (2007) 495–503.
- [6] S. Goyal, D.K. Dhull, K.K. Kapoor, Chemical and biological changes during composting of different organic wastes and assessment of compost maturity, Bioresour. Technol. 96 (2005) 1584–1591.
- [7] D.R. Biswas, G. Narayanasamy, Mobilization of phosphorus from rock phosphate through composting using crop residue-fertilizer, News B (2002) 53–56.
- [8] K. Beata, Lignocellulose Degradation and Humus Modification by the Fungus Paecilomyces Inflatus, University of Helsinki, 2007.
- [9] R. Dayana Amira, A.R. Roshanida, M.I. Rosli, M.F. Siti Fatimah Zahrah, J. Mohd Anuar, C.M. Nazrul Adha, Bioconversion of empty fruit bunch (EFB) and palm oil mill effluent (POME) into compost using *Trichoderma virens*, Afr. J. Biotechnol. 10 (81) (2012) 18775–18780.
- [10] S.N. Shafawati, S. Siddiquee, C.M.V.L. Wong, S.V. Kumar, Lignocellulolytic activities among *Trichoderma* isolates from lahad datu, sabah and deception island, antarctic, J. Microb. Biochem. Technol. 6 (5) (2014) 295–302.
- [11] S. Gaind, A.K. Pandey, J. Lata, Microbial biomass: p-nutrition and enzymatic activities of wheat soil in response to phosphorus enriched organic and inorganic manures, J. Environ. Sci. Health Part B 41 (2006) 177–187.
- [12] N.A.I. Mohamad Zainuddin, A. Faridah, Disease suppression in *Ganoderma* treated with *Trichoderma harzianum*, Plant Protect. Sci. 44 (2008) 101–107.
- [13] U. Pandya, M. Saraf, Application of fungi as a biocontrol agent and their biofertilizer potential in agriculture, J. Adv. Develop. Res. 1 (1) (2010) 90–99.
- [14] F.M.G. Heraux, S.G. Hallett, S.C. Weller, Combining Trichoderma virensinoculated compost and a rye cover crop for weed control in transplanted vegetables, Biol. Control 34 (2005) 21–26.
- [15] Y. Elad, I. Chet, Y. Henis, A selective medium for improving quantitative isolation of Trichoderma spp. from soil, Phytoparasitica 9 (1) (1981) 59–67.
- [16] M.R. Erwan Ismail, H. Mohd Saud, S.H. Habib, S. Siddiquee, H. Kausar, Physical, chemical and biological changes during the composting of oil palm frond, Afr. J. Microbiol. Res. 6 (19) (2012) 4084–4089.
- [17] Y.K. Jeong, J.S. Kim, A new method for conversation of nitrogen in aerobic composting process, Bioresour. Technol. 79 (2001) 129–133.
- [18] A. Singh, S. Sharma, Composting of crop residue through treatment with microorganisms and subsequent vermin composting, Bioresour. Technol. 85 (2002) 10–11.
- [19] M.S.Y. Haddadin, J. Haddadin, O.I. Arabiyat, B. Hattar, Biological conversion of olive pmace compost by using *Trichoderma harzianum* and *Phanerochaete chrysosporium*, Bioresour, Technol, 100 (2009) 4773–4782.
- [20] N.A. Kabbashi, M.Z. Alam, M. Ainuddin, Bio-composting process development by SSF for utilization agro-industrial wastes, 3rd Kuala Lumpur International Conference on Biomedical Engineering 2006 (Biomed 2006), IFMBE Proceedings (2006).
- [21] Y. Siddiqui, S. Meon, M.R. Ismail, A. Ali, *Trichoderma*-fortified compost extracts for the control of choanephora wet rot in okra production, Crop Protect. 27 (2008) 385–390.
- [22] L. Cotxarrera, M.I. Trillas-Gay, C. Steinberg, C. Alabouvette, Use of sewage sludge compost and *Trichoderma asperellum* isolates to suppress *Fusarium* wilt of tomato, Soil Biol. Biochem. 34 (2002) 467–476.

- [23] M. Vos, A.B. Wolf, S.J. Jennings, G.A. Kowalchuk, Micro-scale determinants of bacterial diversity in soil, FEMS Microbiol. Rev. 37 (6) (2013) 936–954.
- [24] S. Barrington, D. Choiniere, M. Trigui, W. Knight, Effect of carbon source on compost nitrogen and carbon losses, Bioresour. Technol. 83 (2002) 189–194.
- [25] S. Siddiquee, U.K. Yusuf, K. Hossain, S. Jahan, In vitro studies on the potential Trichoderma harzianum for antagonistic properties against Ganoderma boninense, J. Food Agric. Environ. 7 (3&4) (2009) 970–976.
- [26] H.C. Weltzein, Biocontrol of foliar fungal disease with compost extracts, in: J.H. Andrews, S.S. Hirano (Eds.), Microbial Ecology of Leaves, Springer-Verlag, New York, 1991, pp. 430–450.
- [27] H.A.J. Hoitink, A.G. Stone, D.Y. Han, Suppression of plant diseases by composts, HortScience 32 (1997) 184–187.
- [28] M. Sariah, C.W. Choo, H. Zakaria, M.S. Norihan, Quantification and characterization of *Trichoderma* spp. from different ecosystems, Mycopathologia 159 (2005) 113–117.
- [29] G. Al-Khawaldeh, Bioconversion of Olive Pomace into a Broiler Feed High in Protein and Metabolizable Energy. Master of Science Thesis, University of Jordan, Amman, Jordan, 1997.
- [30] T.K. Kirk, E. Schultz, W.J. Connors, L.F. Lorenz, J.G. Zeikus, Influence of culture parameters on lignin metabolism by Phanerochaete chrysosporium, Arch. Microbiol. 41 (1978) 465–505.
- [31] I.D. Reid, Biodegradation of lignin, Can. J. Bot. 73 (1995) 1011-1018.
- [32] S. Zeilinger, D. Kristufek, I. Arisan-Atac, R. Hodits, P.C. Kubicek, Condition of foratio: purification and characterization of an-galactasidase of *Trichoderma reesei* RUT C-30, Appl. Environ. Microbiol. 59 (1993) 1347–1353.
- [33] R. Grisso, M. Alley, D. Holshouser, W. Thomason, Precision Farming Tools: Soil Electrical Conductivity, Virginia Cooperative Extension, 2009.
- [34] M.L. Gullino, A. Garibaldi, Influence of soilless cultivation on soilborne diseases, ISHS Acta Hort: Intl. Symp. New Cult. Syst. Greenhouse (1994) 361.
- [35] T. Nagasaki, C.J. Chapin, G.G. Gundersen, Distribution of detyrosinated microtubules in motile NRK fibroblasts is rapidly altered upon cell-cell contact: implications for contact inhibition of locomotion, Cell Motil. Cytoskeleton. 23 (1) (1992) 45–60.
- [36] H. Marstop, Influence of soluble carbohydrates free amino acids, and protein content on the decomposition of *Lolium multiflorum* shoots, Biol. Fert. Soils 21 (1996) 257–263.
- [37] M.P. Bernal, M.A. Sanchez-Monedero, C. Paredes, A. Roig, Carbon mineralization from organic wastes at different composting stages during their incubation with soil, Agr. Ecosyst. Environ. 69 (1997) 175–189.
- [38] E. Iglesias-Jimenez, V. Perez-Garcia, Determination of maturity indices for city refuse composts, Agr. Ecosyst. Environ. 38 (1992) 331–343.
- [39] F.E. Allison, Soil Organic Matter and Its Role in Crop Production, Elsevier, New York, 1973.
- [40] F. Schuchardt, D. Darnoko, P. Guritno, Composting of oil palm empty fruit bunch (EFB) with simultaneous evaporation of oil mill waste water (POME), International Oil Palm Conference, July 8–12, 2002, Nusa Dua, Bali, Indonesia, 2016.
- [41] L. Chabbey, Heavy metals, maturity and cleanness of the compost produced on the experimental site of Chatillon, Preoceedings of the ReC'93 International Recycling Congress, Palexpo, Geneva, Switzerland, 1993, pp. 62–68.
- [42] R.D. Amira, A.R. Roshanida, M.I. Rosli, M.F. Siti Fatimah Zahrah, J. Mohd Anuar, C.M. Nazrul Adha, Bioconversion of empty fruit bunches (EFB) and palm oil mill effluent (POME) into compost using Trichoderma virens, Afr. J. Biotechnol. 10 (81) (2011) 18775–18780.
- [43] F.M. Rashad, W.D. Saleh, M.A. Moselhy, Bioconversion of rice straw and certain agro-industrial wastes to amendments for organic farming systems: 1. Composting quality, stability and maturity indices, Bioresour. Technol. 101 (2010) 5952–5960.
- [44] M. Pramanik, I. Chakraborty, S. Mondol, S.S. Islam, Structural analysis of a water-soluble glucan (Fr.I) of an edible mushroom, *Pleurotus* sajor-caju, Carbohydrates Resour. 342 (2007) 2670–2675.
- [45] K.K. Wong, J.N. Saddler, Trichoderma xylanases, their properties and application, Crit. Rev. Biotechnol. 12 (5–6) (1992) 413–435.