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Research Article

Empty Fruit Bunches as Potential Source for Biosilica Fertilizer for Oil Palm

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ABSTRACT
In Indonesia, the development of oil palm plantations has been going on a pervasive way; they covered about 14.03 million hectares in 2017. This massive coverage of land might then generate a tremendous amount of biomass per year, both in the form of both solid and liquid wastes. The processing of fresh fruit bunches (FFB) in palm oil mill (POM) produces wastes that primarily in the form of empty fruit bunches (EFB), which is amounting of up to 25% (w/w) of FFB. It has been being indicated that EFB contains a considerable amount of silica (Si) which attracts the Indonesian Research Institute for Biotechnology and Bioindustry (IRIBB) to investigate the potential use of EFB as a source of bio-available Si, in the form of H_4SiO_4 (mono silicic acid, BioSilAc). The experiment was carried out at Sungai Mirah Minting Estate, PT Bumitama Gunajaya Agro-Central Kalimantan. The EFB material was obtained from POM and chopped into 2.5-5.0 cm in size. A four-week bio-decomposition process was employed by using bio-decomposers containing *Trichoderma pseudokoningii*, *T. polysporum*, and *Phanerochaete chrysosporium*. Chemical analyses of composted EFB were conducted before and 28-days after decomposer application. The presence of Si in the compost was observed by scanning electron microscopy (SEM). The effect of Si-containing EFB compost on the immature and mature oil palm was evaluated. Seven treatments, i.e. combination of EFB compost and BioSilAc application with reduced-dosages of NPK fertilisers were arranged in a random block design with three replicates. The results show that large quantities of silica bodies attached to the surface of EFB fibres and amounting to 0.44% soluble Si. The FFB data indicated that the application of 75% NPK + 500 kg composted EFB + 2 L BioSilAc/ha/year on a five-year-old plant resulted in higher yield than that obtained from 100% standard dosage of NPK. The study also revealed that the application of EFB compost reduced 50% of BioSilAc dosage.

INTRODUCTION
Indonesia has been placed as the world's first producer of palm kernel and crude palm oils. In producing crude palm oil (CPO) and palm kernel oil (PKO), the oil palm industry is strongly dependent on the processing of the fresh fruit bunches (FFB) at palm oil mill (POM) and traded internationally. However, this process produces also a waste of solid organic waste [i.e. empty fruit bunch (EFB)], which reaches up to 25% of FFB. Therefore, if Indonesia produced 38 million tons of CPO in 2017, then it is equivalent to 190 million tons of FFB. Since FFB yielded in 20% CPO, it leads to the potential availability of EFB more than 47 million tons per year. In addition to the production of solid waste, the POM also produces nonsolid biomass called as palm oil mill effluent (POME) in huge quantity, material discharged from washing and sterilization of the palm fruits.

Oil palm EFB fiber has been identified as the single most important agriculture biomass in Indonesia was usually contains 30–35% lignocellulose, 1–3% residue oil, and roughly 60% of moisture (Gunawan *et al.* 2009). The lignocellulose or fiber consists of cellulose, hemicellulose, and lignin. Many types of research on EFB had been focused on energy, biochemicals, and wood-related products development purposes (Geng, 2013) as well as for compost (Goenadi, 2006). Therefore, this material could become an important portfolio to sustain the development and growth of energy demand, biochemicals, industrial materials, and sources of available nutrients for the plant. It has also been indicated that EFB contains a considerable amount of silica (Si) so then the Indonesian Research Institute for Biotechnology and Bioindustry (IRIBB) has investigated the potential use of EFB as a source of bio-available Si, in the form of H_4SiO_4 (mono silicic acid) (Santi *et al.* 2017). The IRIBB has also developed a bio-available Si product derived from quartz sand enriched with Si-solubilizing microbes (BioSilAc).

Silica (Si) is the second most abundant element in soils and can be found in noticeable concentrations in many terrestrial plants (Epstein, 1994; Keeping & Reynolds, 2009). Plant species vary in their ability to take up and accumulate Si as silicon dioxide (SiO_2) in their tissues; depends on this characteristic, plants are then classified as excluders, intermediate types, or accumulators (Mitani & Ma, 2005; Montpetit *et al.* 2012). Most dicots accumulate less than 0.1% Si on a dry weight basis, but many grasses species are able to accumulate as much as 10% (Montpetit *et al.* 2012; Ma *et al.* 2002; Vivancos *et al.* 2015). It has been widely reported that Si is able to suppress both physical stresses, such as drought, high temperature, UV, loading, and freezing, and chemical stress, including salinity, nutrient imbalance, and metal toxicity (Ma, 2004; Ma & Yamaji 2015). Silica has not been recognized as an essential element, although numerous studies have demonstrated that Si is beneficial for plant growth and development, especially under a wide range of abiotic stress conditions (Sanglard 2016; Shi *et al.* 2013; Yin *et al.* 2014). Si deposition occurs mainly as phytoliths ($SiO_2\_nH_2O$) (Ye *et al.* 2013). It acts as a physical barrier and thus improves plant resistance to pathogens and insects. Najihah *et al.* (2015) observed that the accumulation of Si in epidermal and endodermal cell walls protected oil palm roots from penetration of *Ganoderma boninense* fungus. The objective of our study reported here is to determine the potential use of composted EFB as a source for the availability of Si to oil palm.

MATERIAL AND METHODS

Composting Technology

The experiment was carried out at Sungai Mirah Minting Estate, PT Bumitama Gunajaya Agro-Central Kalimantan. EFB materials were obtained from POM and chopped into 2.5-5.0 cm length. A four-week bio-decomposition process was employed by using a bio-decomposer containing *Trichoderma pseudokoningii*, *T. polysporum*, and *Phanerochaete chrysosporium.* In brief, composting steps involved EFB collection, shredding, mixing with bio-decomposer, incubation, and harvesting. Dosages of bio-decomposer were 0.2% (w/w) with four weeks incubation period without turning following the process outlined by Goenadi (2006).

Table 1. Chemical characteristics of fresh and composted EFB

Parameters	Fresh EFB	EFB compost
pH	6.3	8.0
N (%)	0.7	1.9
P_2O_5 (%)	0.4	0.6
K_2O (%)	2.5	3.8
SiO_2 (%)	11.3	28.7
dissolved Si (%)	0.03	0.44
Ca (%)	0.4	1.3
Mg (%)	0.3	0.58
CEC (cmol+/Kg)	7.3	52.1
C-organic (%)	41.9	36.9
C/N	59.9	19.4

Chemical Analysis

Analysis of EFB

The number of chemical characteristics of EFB before and after bio-decomposition was determined at the laboratory of IRIBB. The EFB samples were air-dried and passed through 100 mesh sieves and analyzed for the following: pH, nitrogen (Kjeldahl), phosphorus (spectrophotometer), potassium (Atomic Absorption Spectrophotometer, AAS), total SiO_2 (gravimetry), soluble Si (spectrophotometer), calcium (AAS), magnesium (AAS), cation exchange capacity (CEC) by using SNI 13-3494-1994 standard method, and C-organic (spectrophotometer).

Soil and leaf analysis

Soil samples were air-dried and passed through 2 mm sieve and analyzed for the following: pH, soil texture, C-organic, nitrogen, phosphorus, potassium, calcium carbonate, CEC, Boron, and exchangeable Al and H. Whereas leaf nutrient analysis was N, P, K, Mg, and SiO_2.

BioSilAc preparation

A 150 g of 325-mesh Belitung quartz sand sample was boiled in 100 mL HCl at 5 N concentrations until almost all of the solution evaporated to dissolve any contaminant elements present. The treated samples were then washed out with tap water several times to eliminate the contaminants and the rest of the HCl solution. Wet samples were transferred on a

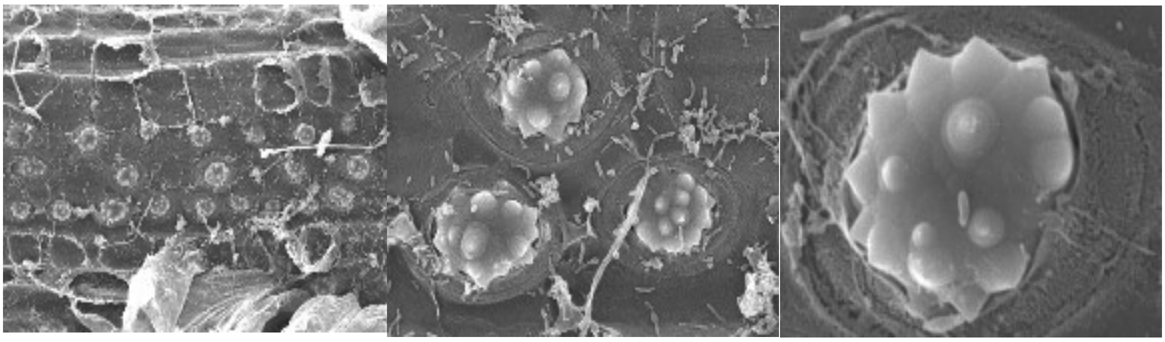

Figure 1. The silica body of fresh EFB found between fibre under microscopic magnification of 500x (left); 2,000x (middle); and 7,500x (right).

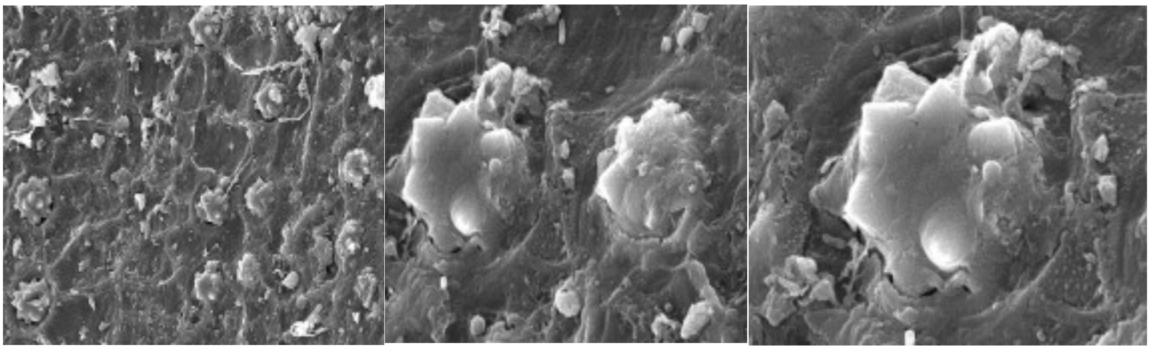

Figure 2. The silica body of EFB compost under microscopic magnification of 700x (left); 3,500x (middle); and 5,000x (right).

sheet of paper and dried out at 100°C in an oven until completely dry 60 g washed-sample was then mixed with 80 g NaOH (s) in a stainless pan and heated on the stove at 330°C while stirred manually until melted. The melted mixture was kept stirred until it dried out. After cooling at room temperature, 60 g pre-treated quartz sand was dissolved in 400 mL distilled water. The liquid obtained was the soluble silica (H_4SiO_4) (Santi *et al.* 2017), whereas the solid formula was prepared by the inoculation of acid-base-pretreated 325-mesh quartz sand with Si-solubilizing microbes i.e.: *Burkholderia cenocepacia*, *B. vietnamiensis*, *Aspergillus niger*, *Aeromonas punctata* (Santi & Goenadi, 2017). These two Si sources are called as BioSilAc. Silica concentration was determined by spectrophotometer.

Scanning Electron Microscopy Analysis

This analysis was performed to confirm the presence of Si in EFB tissues. All solid material of fresh and composted were examined with a Scanning Electron Microscope (SEM). A slice cut of EFB fiber was taken and prepared for SEM analyses. The electron beam is accelerated through a high voltage of 20 kV and passes through a system of apertures and electromagnetic lenses to produce a thin beam of electrons (Zhou *et al.* 2006). In the early stages, a material sample levelled with a special tool. After sputter coating the cast with 35 nm of gold-palladium (Au-Pd), electron micrographs were generated using a JeolJSM-5310LV SEM.

Field Experiment

A field experiment was conducted at Sungai Mirah Minting Estate, Central Kalimantan and arranged in seven treatments, i.e. combination of EFB compost and BioSilAc application with reduced dosages of NPK fertilizers, were arranged in a random block design with three replicates. The BioSilAc used was in liquid and solid forms. Soil chemical characteristics of immature-plant plot were pH 5.0; 83% sand; 14.5% clay; 2.5% silt; 0.12 (N); 1.98% (C-organic); 0.79 cmol+/kg (exchangeable Al); 0.32 cmol+/kg (exchangeable H); 3.67cmol+/kg CEC; 6.4 ppm (B); 0.006% (P_2O_5); 0.18% (K_2O); and 0.05% (CaO). The data from mature-plant plot were pH 4.8; 61.0% sand; 25.7% clay; 13.3% silt; 0.17 (N); 3.02% (C-organic); 1.4 cmol+/kg (exchangeable Al); 0.74 cmol+/kg (exchangeable H); 6.8 cmol+/kg CEC; 5.5 ppm (B); 0.102% (P_2O_5); 0.17% (K_2O); and 0.34% (CaO). Applied on a two-year (immature) and five-year (mature) old plants the treatments consist as follows: (i) 100% NPK standard dosage (T1); (ii) T1+ 225 kg BioSilAc /ha/year; (iii) 75% (T1) + 225 kg BioSilAc/ha/year; (iv) T1+ 4 L BioSilAc/ha/year; (v) 75% (T1) + 4 L BioSilAc/ha/year; (vi) T1 + 500 kg EFB compost + 2L BioSilAc/ha/year; (vii) 75% (T1) + 500 kg EFB compost + 2L BioSilAc/ha/year. The oil palm

Table 2. The leaf nutrient contents of immature oil palm one year after treatments.

Treatments	Leaf nutrient contents (%)				
	N	**P**	**K**	**Mg**	**SiO_2**
NPK 100% standarddosage (T1)	2.7	0.21	0.9	0.15	0.98
(T1) + 225 kg BioSilAc/ha/year	2.7	0.28	1.3	0.18	2.5
75% (T1) + 225 kg BioSilAc/ha/year	2.6	0.26	1.2	0.20	2.9
(T1) + 4 Liter BioSilAc/ha/year	2.7	0.24	1.0	0.18	1.04
75% (T1) + 4 Liter BioSilAc/ha/year	2.7	0.27	1.3	0.21	1.78
(T1) + 500 kg EFB compost + 2 L BioSilAc/ha/year	2.5	0.26	0.8	0.21	1.90
75% (T1) +500 kg EFB compost + 2 L BioSilAc/ha/year	2.7	0.27	1.0	0.20	1.10

Note: Classification of leaf nutrient contents of oil palm according to Fairhurst dan Hardter (2003): N (%) = <2.50 (deficient), 2.6-2.9 (optimum), >3.1 (high); P(%) = <0.15 (deficient), 0.16-0.19 (optimum), >0.25 (high); K (%) = <1.00 (deficient), 1.1-1.3 (optimum),> 1.8 (high); and Mg (%) = <0.20 (deficient), 0.30-0.45 (optimum), >0.70 (high).

plants were planted in 2013 (mature) and 2015 (immature). Each was applied on a plot consisting of 25 trees with nine of them in the middle as observed -trees. Selected parameters observed included leaf nutrient content (N, P, K, and Mg) of leaf no. 9, average weight and number of FFB. Data were analyzed by ANOVA and Duncan's Multiple Range Test (DMRT).

RESULTS AND DISCUSSION

Fresh and Composted EFB Chemical Characteristics

Chemical characterization of the fresh- and composted-EFB was conducted to determine the potential level of dissolved Si and other nutrients that can be utilized by plants. The characterization results are presented in Table 1. The results indicated that the levels of N, P, K, Ca, and Mg from the composted-EFB increased. Similarly, total SiO_2 and dissolved Si contents increased significantly, i.e. 2.5 and 14.6 times, respectively, in comparison to non-decomposed EFB. Furthermore, there was an increase in CEC and pH values, whereas the C/N ratio was decreased drastically from 59.9 to 19.4. Therefore, composting EFB with bio-decomposer for 28 days incubation has improved the quality of organic material, the nutrients content, and the availability of Si for plants.

The Present of Silica Body in EFB

In general, EFB has a thicker cell wall, thinner lumen, smaller diameter, and shorter fibers. EFB fibers have similar cell wall thickness and fiber length while having a thicker lumen and larger diameter when compared with hardwood species (Law *et al.* 2007; Jinn *et al.* 2015). The evidence of Si present in fresh and composted EFB was collected by using a scanning electron microscope (SEM). The SEM analysis showed that the amount of crystalline Si in the cross-sectional slices of EFB quite abundant in fresh (Figure 1) and composted EFB (Figure 2). Silica bodies were bind between the fibers. The silica bodies observed on fibre strands were a round-spiky shape. This evidence is in agreement with those reported by Harun *et al.* (2013) and Jinn *et al.* (2015). In composted EFB Si position nearly separated from fiber tissue because the tissue

Table 3. Growth of immature oil palm one year after treatments.

Treatments	Frond length (cm)	Number of leave (sheet)	Width of petiole (cm)	Dense of petiole (cm)
NPK 100% standarddosage (T1)	340.3^{ab}	218^{ab}	4.9^{ab}	3.2^{a}
(T1) + 225 kg BioSilAc/ha/year	328.0^{b}	211^{b}	4.9^{ab}	3.0^{ab}
75% (T1) + 225 kg BioSilAc/ha/year	372.8^{a}	208^{b}	5.2^{a}	3.3^{a}
(T1) + 4 Liter BioSilAc/ha/year	354.6^{ab}	230^{ab}	4.2^{c}	2.5^{b}
75% (T1) + 4 Liter BioSilAc/ha/year	320.9^{b}	251^{a}	4.3^{bc}	2.4^{b}
(T1) + 500 kg EFB compost + 2 L BioSilAc/ha/year	342.9^{ab}	239^{ab}	4.7^{abc}	2.9^{ab}
75% (T1) +500 kg EFB compost + 2 L BioSilAc/ha/year	383.2^{a}	223^{ab}	5.4^{a}	3.6^{a}
Coefficient variable (%)	6.6	7.7	8.0	11.4

Note: Values in the same column followed by the same superscript letter(s) are not significantly different according to Duncan's Multiple Range Test (P<0.05).

Table 4. The productivity of 2013 planting year oil palm, one year after treatments.

Treatments	Number of FFB	Average weight of FFB (kg)	FFB production (ton/ha/year)
NPK 100% standarddosage (T1)	1,823^{bc}	4.83^{a}	8.80^{ab}
(T1) + 225 kg BioSilAc/ha/year	1,750^{c}	4.70^{b}	8.23^{c}
75% (T1) + 225 kg BioSilAc/ha/year	1,800^{c}	4.70^{b}	8.47^{bc}
(T1) + 4 Liter BioSilAc/ha/year	1,889^{bc}	4.50^{c}	8.50^{a}
75% (T1) + 4 Liter BioSilAc/ha/year	1,973^{ab}	4.63^{b}	9.13^{a}
(T1) + 500 kg EFB compost + 2 L BioSilAc/ha/year	1,875^{bc}	4.70^{b}	8.80^{ab}
75% (T1) +500 kg EFB compost + 2 L BioSilAc/ ha/year	2,084^{a}	4.70^{b}	9.23^{a}
Coefficient variable (%)	4.6	1.4	2.9

Note: Values in the same column, followed by the same superscript letter(s) are not significantly different according to Duncan's Multiple Range Test ($P<0.05$).

conditions of EFB has become weak. The structure of composted EFB tissue was somewhat fragile that enables the release of silica from these tissues will become easier.

Field Experiment

The first application of EFB compost and BioSilAc was conducted in January 2017. The data from this experiment showed that the application of EFB compost and BioSilAc after one year on an immature oil palm could maintain P, K, Mg, and Si absorptions better than those of other treatments including normal dosage of NPK fertilizer. When applied in combination with 500 kg EFB compost + 2 L BioSilAc/ha/year, they reduced the rate of NPK up to 75% standard dosage. Leaf P, K, and Mg content of 500 kg EFB compost combined with 75% NPK dosages were considered to be optimum, i.e.28.6% (P); 11.1% (K); 33.3% (Mg); and 12.2% (SiO_2) compared to the standard NPK dosage treatment. Meanwhile, data analyses indicated that total P content was relatively high (0.27%), whereas N, K, and Mg content of leaf among treatment plots of BioSilAc was at an optimum level according to the classification made by Fairhurst & Hardter (2003) (Table 2). Furthermore, application EFB compost and BioSilAc increased SiO_2 content on the leaf of immature oil palm. Application of 500kg EFB compost + 2 L BioSilAc combined with 75% dosage of NPK resulted in significantly higher vegetative growth of frond length, width and dense of petiole than those of other treatments (Table 3).

Table 4 presents the effect of treatments on the yield of a five-year-old palm. It is evidenced that the highest yield average in terms of FFB was obtained from the application of a 75% dosage of NPK combined with 500 kg EFB compost + 2 L BioSilAc/ha/year. The application of these treatments resulted in higher yield (than that obtained from 100% standard dosage of NPK. This study revealed that the use of EFB compost reduced the need of Si up to 50% for both immature and mature oil palms which promote both better vegetative and productive performances of the palm.

Table 5. Soil characteristics determined at one year after treatments on mature plant plots.

Treatments	pH H_20	N (%)	P_2O_5 (%)	K_2O (%)	C_{org} (%)	Al (%)	exchangeable Al (cmol$^+$kg^{-1})
NPK 100% standarddosage (T1)	4.9^{a}	0.13^{d}	0.05^{b}	0.01^{c}	2.5^{b}	0.11^{ab}	1.50^{bc}
(T1) + 225 kg BioSilAc/ha/year	4.4^{de}	0.16^{bc}	0.01^{c}	0.03^{a}	2.6^{b}	0.06^{d}	1.81^{b}
75% (T1) + 225 kg BioSilAc/ha/year	4.7^{bc}	0.08^{e}	0.02^{c}	0.01^{c}	1.3^{c}	0.06^{d}	1.36^{c}
(T1) + 4 Liter BioSilAc/ha/year	4.4^{de}	0.18^{ab}	0.02^{c}	0.007^{cd}	2.9^{a}	0.10^{abc}	2.67^{a}
75% (T1) + 4 Liter BioSilAc/ha/year	4.5^{de}	0.19^{a}	0.08^{a}	0.01^{c}	3.1^{a}	0.12^{a}	1.74^{b}
(T1) + 500 kg EFB compost + 2 L BioSi-lAc/ha/year	4.7^{b}	0.08^{e}	0.04^{c}	0.004^{d}	1.2^{c}	0.10^{abc}	1.71^{b}
75% (T1) +500 kg EFB compost + 2 L BioSilAc/ha/year	4.6^{cd}	0.15^{cd}	0.06^{b}	0.02^{b}	2.9^{a}	0.09^{c}	1.21^{c}
Coefficient variable (%)	1.2	7.0	19.2	14.4	4.0	9.6	7.7

Notes: Figures in the same column followed by the same letter(s) are not significantly different according to Duncan's Multiple Range Test ($P<0.05$).

The effect of treatments on selected soil chemical properties after one year of application is shown in Table 5. In general, the selected chemical properties of the soil were not so much different among those treatments. No clear evidence noticed or shown relationships between Si application and P and Al contents. However, it is observed that in general application of Si tended to decrease the Al content in the soil. Britez *et al.* (2002) reported that silicon (Si) can make stable complexes with Al and reduce the harmful Al effects. Si can potentially increase the root elongation rate (RER) in Al-toxic solutions, with the magnitude of the effect increasing with the concentration of Si (Koppittke *et al.* 2017). Moreover, these researchers also confirmed that Si is not only deactivated Al in the rhizosphere but also in plant shoot tissue avoiding Al toxicity to the plant.

CONCLUSIONS

The application of Si (BioSilAc) improved the vegetative performance of immature and yield of mature oil palm (4.9%) and increased NPK fertilizer use efficiency (25%) one year after treatment. A 28-day bio-composted EFB could provide Si available to the plant and the addition of 500 kg EFB compost/ha/year combined with 75% NPK fertilizer reduced 50% the need for BioSilAc. Further research is needed to evaluate the long-term effect of Si application on the yield of oil palm.

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