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Effects of Urease Inhibitor and Nitrification Inhibitor on the Nitrogen Losses, Physiological Activity, and Oil Palm Yield on Red-Yellow Podzolic

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ABSTRACT
Nitrogen (N) fertilizer efficiency can be increased by adding N-(n-butyl) thiophosphoric triamide (NBPT) and N-(n-propyl) thiophosphoric triamide (NPPT) as urease inhibitor in urea fertilizer and adding 3,4-dimethyl pyrazole phosphate (DMPP) as nitrification inhibitor in ammonium sulfate (ZA) fertilizer. The research objective was to examine the effectiveness of urease inhibitor (NBPT and NPPT) and nitrification inhibitor (DMPP) on the physiological activity and oil palm yield on red-yellow podzolic. Field experiment was conducted using a Randomized Complete Block Design (RCBD) of single factor and three replications as block. The single factor was nine fertilization treatments of urea with or without urease inhibitor, and of ZA with or without nitrification inhibitor. The use of urease inhibitors decreased the amount of volatilization and N loss fertilizer. The higher dose of broadcasting N fertilizer (on the same type of fertilizer) resulted in the higher amount of N loss and volatilization. All fertilization treatments did not have any significant effect to the width and the length of opening stomata, nitrate reductase activity (NRA), N concentration in oil palm leaf, Oil Extraction Rate (OER), and Free Fatty Acid (FFA). Meanwhile, the oil palms at yellow red podzolic applied with 195 kg ha^{-1} of urea combined with 0.12% of NBPT-NPPT resulted in higher fresh fruit bunch productivity.

Keywords: Fertilizer, nitrification inhibitor, oil palm, urease inhibitor

INTRODUCTION

Oil palm (*Elaeis guineensis* Jacq) is one of plantation commodities which can produce a higher quantitiy of vegetable oil than the other commodities, so these commodities can be planted in a wide scale of cultivation. The maximum productivity of oil palm can be obtained through comprehending the characteristics and critical factor of its cultivation. Jannah *et al*. (2012) said that recently, the productivity of oil palm in Indonesia was 16 tons ha^{-1} of fresh fruit bunch (FFB), whereas the potential productivity of oil palm may attain up to 30 tons FFB ha^{-1}. The implementation of an appropriate cultivation technology may improve its potential productivity.

Fertilization is the determining factor in oil palm's productivity about 50 % of the production costs in the cultivation of oil palm are used for fertilization. Oil palm hybrids developed today are generally responsive to fertilization (Hakim, 2007 and Nurjaya, 2009). The appropriate fertilizations are able to supply the nutritious needs of the plants optimally so that the productivity will increase approaching its potential. One of the necessary fertilizations for oil palm maximum productivity is an appropriate nitrogen fertilization.

Nitrogen is one of the primary-macro nutrients for oil palm commodity. However, the nitrogen added in the fertilizer into the soil is easy to lose by the process of leaching and volatilization, and becomes unavailable to plant because of bunding by soil material. The loss of nitrogen in the soil can be caused by denitrification, volatilization, leaching, and absorbtion by the plant (Sanchez, 1979). The research done by Handayani (2015) showed that the amount of broadcasted N fertilizer in oil palm plantation in West Sumatra was about 163,772,466 kg for the area of 381,754 ha, meanwhile the amount of N fertilizer absorbed by the plants was only

73,697,609.70 kg so that the amount of N fertilizer wasted was 90,074,856.30 kg. The data indicated that the absorption efficiency value of N fertilization was only about 45 %.

The loss of NH_3 can be prevented by the inhibition of urease activity. Urease activity can inhibit the release of NH_4^+ from urea so that the N uptake in the form of NH_4^+ can be absorbed more significantly by plants. Besides, the alteration of NH_4^+ to NH_3 gets smaller due to slow release of NH_4^+ in urea. This situation can potentially improve the efficiency of N uptake by oil palm when the plants are fertilized with urea.

In addition, N loss from added fertilizer can also be prevented through the inhibitation of nitrification process. The use of nitrification inhibitor can decrease the change of ammonium (NH_4^+) to nitrate (NO_3^-) and decrease the production of N_2O gas through the process of nitrification and denitrification. Nainggolan *et al*. (2009) mentioned that broadcasting nitrogen fertilizer in the soil can increase the reaction of nitrification in the soil with the release of hydrogen ions so that the decrease in pH promotes the change in ammonium to nitrate. The activity of nitrosomonas and nitrobacter (autotrof obligate bactery) can increase the amount of nitrate in the soil.

This activity is formed through the process of nitrification (the changes of amonium into nitrate). The amount of nitrate in the soil is related to the concentration of ammonium in the soil. The amount of nitrate will increase if the amonium concentration increases in the soil. The increase of nitrate in the soil is the effect of the increasing of amonium. The process of nitrate formation is called nitrification and this process is mediated by nitrosomonas and nitrobacter.

Nitrosomonas can convert the ammonium to nitrit, meanwhile nitrobacter can convert nitrit to nitrate. N-(n-butyl) thiophosphoric triamide (NBPT) and N-(n-propyl) thiophosphoric triamide (NPPT) are known as the inhibitors of urease in urea fertilizer and 3,4-dimethyl pyrazole phosphate (DMPP) is widely used as the nitrification inhibitor in ZA fertilizer. Both of them are able to inhibit the loss of nitrogen in the soil. The research objective was to examine the effectiveness of urease inhibitors (NBPT and NPPT) and nitrification inhibitor (DMPP) on the physiological activity and oil palm yield in red-yellow podzolic.

MATERIALS AND METHODS

Study area: This research was conducted in Central Seruyan oil palm-plantation (OPP), Central Borneo from November 2016 to Mei 2017. The average age of the plants when the research was conducted was about 10 years old.

Experimental Design. The research was a single factor of field trial method designed in Randomized Complete Block Design (RCBD). This single factor was the package of urea and ZA fertilizer with or without inhibitor consisting of nine packages of fertilizer which are also called as the treatment factor. The treatments tested were nitrogen fertilizer package (N), consisting of nine packages of fertilizer i.e., without N (N0), Urea 195 kg ha^{-1} (N1), Urea 195 kg ha^{-1}+0.12% NBPT-NPPT (N2), Urea 156 kg ha^{-1} (N3), Urea 156 kg ha^{-1} 0.12% NBPT-NPPT (N4), ZA 427 kg ha^{-1} (N5), ZA 342 kg ha^{-1} + 0.8% DMPP (N6), ZA 342 kg ha^{-1} (N7), and ZA 342 kg ha^{-1} + 0.8 % DMPP (N8). The experiment plot consisted of 3 blocks as replications. The total number of plants in every treatment and in every plot was 5 oil palm trees. Moreover, the total number of oil palm trees used was about 135 tress. The fertilizer application method was a ring placement with the same basic fertilization in each treatment, i.e. TSP (46%P_2O_5) 130 kg ha^{-1}, KCL (60 % K_2O) 293 kg ha^{-1}, and dolomite 130 kg ha^{-1}.

Observation parameters. The observations consisted of: (1) Climate condition (rainfall , temperature, and humidity). The rainfall was measured by automated rainfall gauge placed near by the research block for six months, so that temperature and humidity was measured by thermohigrometer (Haar-synth, German) for six months; (2) Physiological component (the length and width of stomata opening and nitrate reductase activity (NRA)). This analysis was measured in the third and sixth month after the application of N fertilizer; (3) The analysis of plants tissue (N concentration in the plant's leave). This analysis was carried out in the BPTP Laboratory, Yogyakarta in the third and sixth month after the application of N fertilizer. Leaf samples were taken by taking leaves on the 17th leaf midrib; (4) Analysis of loss of NH_3 was done using foam method (Cantarella *et al*., 2003). This analysis was carried out at the second and fourth week after application of N fertilizer. The procedure of foam method, i.e. soil → insert soil ring → fertilize → NH_3 volatilization → tube inserted into the soil with an acidified foam as scrubber → NH_3 volatilization → desorption of absorbed NH_4^+ by water → NH_4^+ analysis as measure for volatilized NH_3 from soil; (5) The oil palm yield and the components of yield, i.e. Fresh fruit bunches (FFB), oil extraction rate (OER) and free fatty acid

Table 1. The treatment of fertilizer for oil palm trees

Treatment	Treatment Codes	Amount of N Fertilizer (kg ha^{-1})
Control	N0	0
Urea (46% N)	N1	195
Urea+0.12% NBPT&NPPT	N2	195
Urea (46% N)	N3	156
Urea+0.12% NBPT&NPPT	N4	156
Ammonium Sulfate (21% N)	N5	427
Ammonium Sulfate +0.8%DMPP	N6	427
Ammonium Sulfate (21% N)	N7	342
Ammonium Sulfate +0.8%DMPP	N8	342

Remarks: A table should stand by itself, any necessary information should be given here, such as NBPT, NPPT, and DMPP

Table 2. Rainfall (mm), temperature (ºc), and humidity (%) in the research area (December 2016 – Mei 2017)

Month	Rainfall (mm)	Temperature (ºC)	Humidity (%)
First month	572	29.5	72.2
Second month	160	28.7	76.1
Third month	647	28.4	71.0
Fourth month	336.5	30.6	72.9
Fifth month	664.5	29.3	67.5
Six month	168.5	29.0	67.5

(FFA) was measured on the fifth and sixth month after broadcasting the N fertilizer.

Statistical analysis. The data were analyzed using Analysis of Variance (ANOVA) with $\alpha = 5$ %. If the result of the analysis showed that F sum > F table, it means that there was a significant difference in every treatment, and it would be continued on its analysis using Duncan Multiple Range Test (DMRT) at test level of 5%.

RESULTS AND DISCUSSION

The environment condition at the research area

The environment condition at the research area was one of the influential factors of the growth and the yield of oil palm trees. Kiswanto *et al*. (2008) argued that oil palm is one of tropical plants that can grow well in Indonesia. The period for good solar radiation for oil palm plants was about 5–7 hours day^{-1}, and the optimal temperature was at 24ºC–28º C. The ideal altitude for the plants was about 1–500 meters above the sea level. Moreover, the maximum oil palm humidity for pollination process was about 80–90% and the wind velocity was 5–6 km $hour^{-1}$. According to Pahan (2010), oil palm can grow well in the area with annual rainfall of ≥ 2,000 mm $year^{-1}$ in dry season period (<100 mm $month^{-1}$) and less than 3 months. The appropriate temperature for oil palm plant was about 29º C–33º C in the daylight period and 22º C–24º C in night period.

The results of the research shown in Table 2 suggests that the rainfall and the temperature in the research area were ideal for the growth of oil palm plants. The average humidity in the research area was about 67 % to 76 %. It indicates that the humidity in the research area was under the maximum humidity for oil palm plants that was 80 % (Pahan, 2006).

The loss of N fertilizer

The N nutrient is one of the key factors in agricultural production and in the system area since the need and acumulation of nutrient is higher in the plants. However, N can lose from agroecosystem in many ways due to the result of natural process and agricultural process. The result of the research (Table 3) showed that the use of urease inhibitors (0.12% NBPT & NPPT) in urea fertilizer (N2 and N4) could decrease the amount of volatilization and the loss of N fertilizer rather than that of urea without inhibitor (N1 and N3). The amount of N loss fertilizer and volatilization found in the use of urea without inhibitor (N1) was higher than the urea fertilizer + urease inhibitors (N2) applied in the same dose of fertilizer. It was also found that in the treatment of urea without

Table 3. The amount of N volatilization and the loss of N fertilizer in the oil palm plantation during 1 month after sowing N fertilizer.

Treatment	TThe Amount of N Volatilization (ppm)	The Amount of N Loss Fertilizer (kg ha^{-1})
N0	289.33 d	0.00 d
N1	11,697.00 a	104.92 a
N2	7756.00 b	69.57 b
N3	10,346.00 ab	74.24 b
N4	4,307.33 c	30.91 c
N5	280.00 d	2.51 d
N6	182.00 d	1.63 d
N7	186.67 d	1.34 d
N8	147.00 d	1.06 d
Mean	3,910.15	31.80
CV (%)	22.98	20.85

Remarks: The means in one column followed by the same letter were not significantly different according to DMRT (α 5%).

inhibitor (N3), N loss fertilizer and volatilization in the urea fertilizer (N3) were higher than the urea fertilizer + urease inhibitors (N4) applied in the same dose of fertilizer. This research applied the higher dose of N1 and N2 fertilizer from N3 and N4 yet the higher of the fertilizer dose in the same type of fertilizer (N1 and N3) and (N2 and N4) led to higher of the amount of volatilization and N loss fertilizer.

There was no difference on the amount of volatilization and N loss fertilizer in the N5 and N6, and so was in n N7 and N8. Therefore, broadcasting ZA fertilizer with or without DMPP did not show the significant difference. This occured due to the function of DMPP on ZA fertilizer as the inhibitor of nitrification process (DMPP) inhibiting the process of the change of NH_4^+ to NO_3^-) so it decreased the loss of N in the form of NO_3^- through the leaching process. However, in this research the measurement of N loss fertilizer through the leaching process was not conducted as this research only focused on finding the amount of N loss in oil palm plantation through volatilitation process. This result was in line with the report by Menendez *et al*. (2012) stating that nitrification inhibitor 3,4-dimethyl pyrazole phosphate (DMPP) was able to decrease the conversion of ammonium (NH_4^+) to nitrate (NO_3^-) and N production in the form of gas through denitrification. Besides, Roberts *et al*. (2016) also detailed that nitrification inhibitor is the chemical material which can slow down the conversion of ammonium to nitrate by influencing the activity of Nitrosomonas bacteria and giving the capacity to decrease the leaching nitrate and denitrification loss. Based on the result of the research, the treatment of N0 (control) indicated that there was N fertilizer that lost through volatilization. The volatilization of NH_3 noticed comes from the decomposition of soil organic matter.

Physiological component

Based on the result of analysis, the treatment of N fertilizer with or without the use of urease inhibitor and nitrification in the different dose of fertilizer did not affect the width and length of the stomata opening in three to six months after broadcasting N fertilizer. It can be assumed that the N fertilizer is not the main factor that directly influences the width and the length of the opening of stomata.

The opening and closing of stomata can be influenced by some factors such as the sunlight, the concentration of CO_2, temperature, humidity and the growth hormon. The sunlight can open the stomata in daytime, on which the plants absorb the light with the use of its pigmen called chlorophyl. Meanwhile in night time, the increasing of CO_2 concentration and the drop of moisture can cause the closing of stomata (Kearn and Assmann, 1993).

The increasing of CO_2 diffusion from the athmosphere to the leaf tissue happened because the stomata opens widely, so that it will support CO_2 supply from the environment and it will increase the photosynthesis process. The increasing of photosynthesis process on oil palm plants will increase teh assimilation needed for the growth of the plants.

Crop productivity depends on adequate nitrogen supply. Nitrate is one form of nitrogen in the soil which can be absorbed by plant root. The process of nitrate reduction in plant cells was catalyzed by nitrate reductase (Kasim *et al*, 2013). Nitrate reductase is the main enzyme contained in the plant tissue that is active in photosynthesis process. Nitrate reductase plays very important role in the first synthesis process of amino acid. Nitrate reductase biosynthesis depends on the nitrogen nutrient and its activity can be induced with the availability of nitrate on the leaf (Lawlor, 2002). When NO_3^- was dominantly used as nitrogen source of plants, nitrate reductase was the most important enzyme responsible for nitrate assimilation in plants. The Nitrate Reductase Activity (NRA) was than considered to be limiting factor for growth, development, and protein synthesis (Solomonson and Barber, 1990) and was reported to be highly correlated with plant growth (Shen *et al*., 1993). The most absorbed nitrate are allocated to the leaf, so that it can increase NRA on the leaf (Iqbal, 2008). Based on the result of analysis, in the treatment of N fertilizer the existence of urease inhibitor and nitrification did not significantly affect the NRA in 3 and 6 months after broadcasting the N fertilizer. But from the all treatments, the average of NRA was higher in 6 months after broadcasting N fertilizer compared to that in three months.

The analysis of plant tissues

In the analysis, the means of N concentration in palm oil leaf in 3 and 6 months after broadcasting N fertilizer are 2.23 and 2.21, respectively. The availability of urease inhibitor and nitrification did not affect the N on the oil palm leaf in 3 and 6 months after broadcasting N fertilizer. The treatment of different doses of N fertilizer can cause the various N in the leaf , but it did not show a significant difference.

Most plants contain 1.50 to 6.00% nitrogen in dry weight matter with a sufficiency value of 2.50 to 3.50% in leaf tissue. A lower range of 1.80 to 2.20% was found in most fruit plants and a higher range of 4.80 to 5.50% was found in the legume type (Jones, 1998). According to Corley and Tinker (2003), the average N concentrations (% of dry matter) in the leaflets of 7, 14, and 20 years old palm near Benin City in Nigeria, with no fertilizer were 1.64%, 1.90%, and 1.94%, respectively.

The yield of oil palm and the components of yield

Oil palm productivity was influenced by some factors, such as the environmental, the genetic, and the cultivation technique factors. The environmental factors (enforce) that influence oil palm productivity consisted of abiotic factors (rainfall, soil and topography) and biotic factors (weeds, pest, the number of plant population ha^{-1}). Genetic factor (innate) consisted of the variety and the age of oil plam. Cultivation technique factor (induce) consisted of fertilizer, the management of water and soil, the restraint of weeds, pest, plant desease, and other cultivation activities. Those factors had a correlation and affected each other (Pahan, 2010).

The variety of oil palm in the research area was Marihat with the average potential productivity was about 24 ton ha^{-1} $year^{-1}$ and the highest potential productivity about 30 ton ha^{-1} $year^{-1}$ (Risza, 2010). The average age of the plant at the time of the research was 10 years. According to Corley and Tinker (2003), the bunch productivity of oil palm increases quickly and can reach its maximum at the age of 8–12 years, but it will decrease gradually as the plants grow old until its economical age of 25 years old.

Table 4 provides the weight of FFB per bunch (kg), FFB per tree (kg), and FFB per hectare (tons ha^{-1} $year^{-1}$). The treatment on N fertilizer with or without the use of urease inhibitor and nitrification affected those parameters. Meanwhile, the highest weight of FFB in the N2 treatment reached 35.99 $tons^{-1}$ ha^{-1} $year^{-1}$. In the treatment of N2, N3, and N6, the productivity of oil palm plants was above the potential average productivity of Marihat variety. The data of the weight of FFB per tree and FFB per hectare shows that the higher dose on broadcasting the urea and ZA without inhibitor resulted in lower weight of FFB per tree and FFB per hectare. Despite the difference in results, the disparity between them was not significantly different. Meanwhile, the treatment of urea+urease inhibitor and ZA+nitrification inhibitor showed contradictory.

The treatment of N fertilizer with or without the use of urease inhibitor and nitrification in the different dose of fertilizer did not affect the Oil Extraction Rate (OER) and free fatty acid (FFA) in 6 months after broadcasting N fertilizer. Many factors can affect the OER or the percentage of the oil coming from mesokarp (CPO) and FFA which are fruit maturity level, harvesting technique, and the treatment after the harvest. The level of FFA was determined by the harvest of the bunch to the factory process. Harvesting in the right time was one of the efforts to decrease FFA and also to increase the OER. FFA can be minimized through immediate boiling of FFB after harvest. It is because the increase of FFA is caused by the enzyme activity which hydrolyzes the oil.

Table 4. Fresh fruit bunches (FFB) per bunch, per tree and per hectare.

Treatment	The Weight of FFB Per Bunch (kg)	The Weight of FFB Per Tree (kg)	The Weight of FFB Per Hectare (ton ha^{-1} $year^{-1}$)
N0	15.90 a	17.67 b	19.22 b
N1	19.00 a	21.57 ab	23.46 ab
N2	17.88 a	33.08 a	35.99 a
N3	19.59 a	25.58 ab	27.83 ab
N4	21.36 a	20.98 ab	22.83 ab
N5	14.91 a	17.35 b	18.88 b
N6	24.93 a	24.93 ab	27.13 ab
N7	18.70 a	18.70 b	20.35 b
N8	16.63 a	16.63 b	18.10 b
Mean	18.77	21.83	23.75
CV (%)	26.23	19.83	19.83

Remarks: The means in one column followed by the same letter were not significantly different according to DMRT (α 5%).

Thus, to stop the activity, the boiling must be done. According to Pahan (2010) the standard point of FFA of oil palm was <3.50.

In Indonesia, oil palm plantation development has been done in marginal soils such as peat and red-yellow podsolic. Hardjowigeno (2007) stated that red-yellow podsolic land is a large enough land in Indonesia. The total area of red-yellow podsolic land in Indonesia is estimated at 48 million ha. Podsolic land is commonly found in Sumatra, Kalimantan, Sulawesi, and Irian Jaya. The problems arising in the red-yellow podsolic are acid soil reactions, the high content of Al and the low of nutrients. In that case, a good management such as fertilization and liming is needed, so that the soil will be productive and not degraded (Hardjowigeno, 2007). To increase the productivity of podsolic, it can be done through broadcasting the lime, fertilization, the addition of organic matter, adaptive soil planting, the minimum application of cultivation (or intercropping), terracing, drainage and tillage techniques (Hakim *et al*., 1986). Fertilization in oil palm plantation is a very important factor to improve productivity and quality of oil palm yield. Sutarta *et al*. (2003) mentioned that fertilization is one of the plant maintenance activities to provide sufficient nutrients to promote vegetative plant growth, the maximum and economic production of FFB, and the resistance of pests and diseases. A good fertilization can increase the production to achieve the standard productivity based on the class of land suitability.

The oil palm variety in the research area was Marihat variety, which is one of the superior varieties of such plants resulting from a cross between Dura and Pisifera elders. Dura variety is the female parent, while Pisifera is the male parent. The results of these crosses have show better quality and quantity than other varieties (Fauzi *et al*., 2002).

Fertilization in oil palm plants is a very important factor to increase productivity and the quality of production produced. One of the beneficial effects of fertilization is increasing soil fertility which causes the level of productivity of oil palm plants to be relatively stable and increases plant resistance to disease attacks and the influence of unfavorable climate. In addition, fertilization is useful to ensure the availability of nutrients in the soil so that the plant's nutrient requirements can be fulfilled which will ultimately provide maximum results.

Oil palm plants need large amount of N fertilizer. However, nitrogen in the soil is easily loss to the environment. The loss of nitrogen in the soil occurs in several ways, including through volatilization and denitrification.

The efforts to improve N fertilization efficiency can be done in several ways, including improving fertilization application techniques, improving the physical and chemical properties of fertilizers, changing the shape and size of fertilizers, and by adjusting nutrient content according to plant needs so that plants can be optimally utilized.

The result of the research showed that the use of urease inhibitors in urea fertilizer could decrease the amount of volatilization and the loss of N fertilizer rather than that of urea without inhibitor. The amount of N loss fertilizer and volatilization found in the use of urea without inhibitor was higher than the urea fertilizer+urease inhibitors applied in the same dose of fertilizer. In addition, the higher the fertilizer dose in the same type of fertilizer, the higher the amount of volatilization and N loss fertilizer would be.

Urease inhibitors were able to reduce N loss rates by about 21 %–43 % compared to fertilizers with no urease inhibitors (Bastos *et al*., 2015). In the research conducted by Suter *et al*. (2012), urease inhibitors inhibited urea hydrolysis between 2 to 14 days compared to urea which was not given urease inhibitor. The use of urease inhibitors could also reduce NH_3 volatilization by about 68% compared to urea without inhibitor.

The high effectiveness of NBPT and NPPT in inhibiting urease activity could reduce the rate of loss of N fertilizer in oil palm plantation. Economically, the benefit was the potential reduction in production costs due to the reduced dose of N fertilization. The potential for N contamination from oil palm plantation was the highest when compared to other commodities due to the high amount of N fertilizer for plants and the most extensive planting area. This condition occured if the N absorption efficiency of the given fertilizer was low. The use of NBPT and NPPT in urea and DMPP in ammonium sulfate had the potential to reduce production costs associated with the use of N fertilizer, ensuring the sustainability of oil palm plantation business, and creating environmental sustainability due to a decrease in the rate of N volatilization.

CONCLUSIONS

The use of urea without combined with NBPT-NPPT led to higher N volatilization and loss compared with the use of urea combined with NBPT-NPPT, at the same urea dose. All fertilization treatment did not result in any significant effect to the width and the length of opening stomata, NRA, N concentration in oil palm leaf, OER, and FFA. The oil palms at yellow red podzolic applied with urea 195 kg ha^{-1} combined with 0.12% of NBPT-NPPT resulted in higher fresh fruit bunch productivity. The higher dose of broadcasting the urea and ZA without inhibitor caused the lower the result of the weight of FFB per tree and FFB per hectare. So that the management of nitrogen fertilization in oil palm plants must consider the right dose for plant needs.

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