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Exogenous Application of Calcium, α-Naphthaleneacetic Acid and 1-Methylcyclopropene Improved Fruit Growth and Oil Yield of Oil Palm (*Elaeis guineensis* Jacq.) Grown on Ultisol

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ABSTRACT

Soil acidity is one of the main factors limiting the growth and sustainable production of oil palm in Indonesia. The aim of this study was to evaluate the effect of various exogenous compounds on improving fruit growth and oil yield of palm oil grown on Ultisol. The experiment employed three treatments which were arranged in a Randomized Complete Block Design (RCBD) with three replications. The treatments were: 1-MCP (0, 100, 200, and 300 ppm), $CaCl_2$ (0 and 50 ppm) and NAA (0 and 200 ppm). Application of Ca^{2+}, NAA and 1-MCP either single or in combination improved fruit growth and oil yield by increasing almost all variables, except the number of bunches, bunch weight and fruit set. The 100 ppm 1-MCP + 50 ppm Ca^{2+} + 200 ppm NAA treatments showed the highest oil-to-dry mesocarp content. To get the highest oil to bunch (OB), the combination of 100 ppm 1-MCP + 200 ppm NAA; 50 ppm Ca^{2+} + 200 ppm NAA, and a single treatment of 50 ppm Ca^{2+} were very promising to be utilized.

INTRODUCTION

Oil palm (*Elaeis guineensis* Jacq.) is an important crop for many developing countries because this crop has become the main source of income for farmers as well as an alternative fuel source. The Indonesian palm oil industry continues to grow to become the largest producer of palm oil in the world, where in 2021 it is estimated that Indonesia's oil palm plantation area reached 16.76 million ha with the production of crude palm oil (CPO) of about 49.71 million tons (Directorate General of Plantation, 2021). The average CPO productivity in Indonesia on 2021 was 3.7 t/ha, much lower than its potential of 6 t/ha (BPS, 2022). Many oil palm plantations, however, are faced with problems of low soil fertility and Al toxicity, as most of the oil palm plantations are grown on acid soils with low pH and high Al. Aluminum toxicity is a major obstacle that inhibits growth and reduces yield of many crops grown on acidic soils. It has been reported that acidic soils occupy about 50% of agricultural land worldwide (Rahman & Upadhyaya, 2021), such as land in Indonesia which is dominated by Ultisols (56%). There are several constraints on the growth and production of fruit and oil content of oil palm under high aluminum concentrations. There are evidences that aluminum toxicity reduced root and shoot growth (Khoiriyah et al., 2016; Ratnasari et al., 2017; Utami et al., 2019), particularly in non-tolerant varieties (Putra et al., 2021). The recent study using oil palm seedlings grown in nutrient culture showed that high Al concentrations (400-1600 µM) induced an increase of MDA, and decreased the photosynthesis rate, resulting in impaired root and shoot growth (Hidayah et al., 2020). Several studies have proven that Al toxicity reduces the growth and production of oil palm in the field (Shamshuddin et al., 2014; Supena et al., 2014), the condition of which also occurred in our plantation. Based on the observation in that plantation which is dominated

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by Ultisol with high Al^{3+} content (Al^{3+} saturation > 70%) shows that the production of bunches is only about 70-75%, while the ratio of oil to bunch is only 65-70% of its potential. This is presumably due to the very low fruit set percentage (± 45%) which causes the high number of abnormal (rejected) bunches (> 50%) under the conditions of low pH and high Al. To improve fruit growth and oil yield of oil palm, therefore, a practical and inexpensive method is needed.

It is known that the percentage of oil content in fresh palm fruit bunches will determine the amount of CPO. Moreover, the number of bunches harvested, the weight of bunches, and the oil content of fruit will also determine the oil yield (Corley & Tinker, 2015). Mandal & Babu (2008) stated several factors that affect oil yield, including fruit maturity, mesocarp thickness, mesocarp weight, percentage of oil to mesocarp weight and moisture content. If the percentage of oil in the dry weight of the mesocarp is high and the weight of the mesocarp is high, it will invariably contain a high oil content. The process of fruit formation, mesocarp and oil is strongly influenced by metabolic processes that are closely related to the influence of enzymes and plant growth regulators. Therefore, some efforts are needed to improve cultivation techniques to increase CPO productivity. Several exogenous compounds have been reported to enhance flower and fruit development such as NAA (Sangwiroonthon et al., 2017; Romero et al., 2021), 1-MCP (Rath et al., 2017) in oil palm and apple (Tomala et al., 2020) and Ca^{2+} in mangosteen (Tanari et al., 2018) and tomato (Rachmah et al., 2017). Tanari et al., (2018) showed positive interaction between Ca^{2+} and NAA to increase the number of normal fruits of mangosteen. The aim of this study was to evaluate the effect of various exogenous compounds, namely NAA, 1-MCP and Ca^{2+} on improving fruit growth and oil yield of oil palm grown on acidic soil Ultisol.

MATERIALS AND METHODS

The research was carried out from 2018 to 2019 at PT Waru Kaltim Plantation, East Kalimantan, Indonesia. The averages of temperature, annual rainfall and relative humidity were 27.5°C, 1906 mm/year, and about 82-86%, respectively (BPS Penajam Paser Utara, 2022). The plants used in this experiment were 5-year-old Socfind variety, which were planted on Ultisol having pH of 4.27, exchangeable Al^{3+} 6.57 cmol/kg and Al^{3+} saturation 76.6%. The treatment was three exogenous materials, i.e. $CaCl_2$, 1-MCP (Methylcyclopropene), and NAA (Naphthalene Acetic Acid) at different concentrations which were arranged in a Randomized Complete Block Design (RCBD) with three replications. The treatments were four levels of 1-MCP (M0 = 0 ppm, M100 = 100 ppm, M200 = 200 ppm and M300 = 300 ppm), two levels of $CaCl_2$ (C0 = 0 ppm and C50 = 50 ppm) and two levels of NAA (N0 = 0 ppm and N200 = 200 ppm). Each treatment combination was repeated 3 times consisting of 5 bunches of each experimental unit (plot), so that there were 48 plots with a total of 240 bunches. Plot marking of sample bunches was carried out based on the uniform receptive female flowers, which was initiated by selecting receptive female flowers simultaneously on the same day to obtain homogenous samples. This method was carried out separately for each replication, where in each replicate 80 receptive flowers were selected. The selection of sample bunches for the second and third replications was carried out using the same method at 2-week intervals. Measurement of the number of bunches per tree was conducted every month, while observations of fruit morphology, fruit weight, average bunch weight and bunch analysis were carried out after harvest.

Application of $CaCl_2$ and NAA for each treatment concentration was performed 1 week after flower anthesis (flower cloves). The application of $CaCl_2$ and NAA was carried out twice, of which, the first was carried out on the day when the female flowers were receptive and the second was applied one week after the recepive period of female flowers. Exogenous compound 1-MCP was also applied twice, i.e., the first was when the fruit bunches had turned reddish (18 weeks after receptivity) and the second was 10 days after the first application. Each treatment of exogenous compound was applied to oil palm bunches using a hand sprayer with a volume of 50 ml per flower/bunch.

Fruit Morphology

The bunches were harvested at the appropriate period to obtain optimal oil content. The harvested bunches were weighed and recorded as bunch weight. Measurement of fruit length, width and weight were carried out after the bunches were harvested. Fruit length and width measurements were carried out using a caliper, the average values were obtained from three samples of oil palm bunches. Fruit weight was obtained from the weight of all pithy fruits divided by the total number of pithy fruits.

Bunch Analysis

The bunch analysis was performed according to Mandal & Babu (2008) with modifications. The harvested bunches were weighed and the spikelet was separated from the bunch stalks, then 30 spikelets were selected randomly. All the fruits were separated from the spikelet and measured individually. Fruits were grouped based on fruit criteria (pipe fruit, parthenocarpy fruit and infertile fruit). The percentage of fruit to bunch was calculated by the formula for the weight of the pithy fruit divided by the total weight of the fruit. The pithy fruits of 30 selected spikelets were composited and divided into 4 parts. A total of 30 pieces from 1 part of the composite were selected as samples for oil analysis. Fruit samples for oil analysis were weighed and recorded. The mesocarp was sliced or separated from the nut manually with a knife. The mesocarp slices were weighed and oven-dried at 105°C for 24 hours. Wet and dry mesocarps were weighed to measure mesocarp moisture content and 5 g of dry mesocarp was used for oil estimation by standard method. Soxhlet apparatus with an extraction capacity of 200 ml was used in the extraction process for 14 hours. The oil content in the mesocarp was measured. The oil content to bunch was calculated by the formula:

FB = (Fertile fruit weight)/(Total fruit weight) x 100%..1)

WMF = (Wet mesocarp weight)/(Fruit weight) x 100 % ...2)

OWM = (Oil content in mesocarp)/
(Wet mesocarp weight) x 100% ....3)

Oil to bunch = (FB xWMF)/100 x OWM/100 ....4)

Where: FB = Fruit to bunch (%), WMF = Wet mesocarp to fruit (%), OWM = Oil to wet mesocarp (%).

Data Analysis

Data were analyzed by ANOVA (Analysis of Variance) using Genstat v19.1.0.21390 statistical software. Significant mean differences were separated using Duncan's Multiple Range Test at $p < 0.05$. To determine the relationship between variables, the Pearson correlation test was used.

RESULTS AND DISCUSSION

The recapitulation of ANOVA (analysis of variance) on the treatment of various exogenous compounds on oil palm fruit is presented in Table 1. The F-test of the three exogenous compounds, both single and interaction, showed a significant effect on oil palm fruit, especially on fruit quality, as indicated by the variables WMF (wet mesocarp to fruit), DMF (dry mesocarp to fruit), OWM (oil to wet mesocarp), FB (fruit to bunch) and OB (oil to bunch). The effect on fruit size was observed in fruit weight (FWG) and fruit length (FL) which occurred mainly in the interaction of 3 exogenous compounds, which represents the effect on fruit growth.

Table 1. The recapitulation of ANOVA (Analysis of Variance) for the treatment of various exogenous compounds on oil palm fruit

Exogenous compound treatments	FL	FWD	FWG	WMF	DMF	NB	ABW	FS	ODM	OWM	FB	OB
MCP	ns	ns	ns	*	*	ns	*	ns	*	*	ns	ns
Ca	ns	ns	*	*	*	ns	ns	ns	ns	ns	*	*
NAA	ns	ns	*	*	*	ns	ns	ns	*	*	*	*
MCP * Ca	ns	ns	*	*	*	ns	ns	ns	ns	*	ns	*
MCP * NAA	ns	ns	ns	*	*	ns	ns	ns	ns	*	ns	ns
Ca * NAA	ns	ns	*	*	*	ns	ns	ns	ns	*	*	*
MCP * Ca * NAA	*	ns	*	*	*	ns	ns	ns	*	*	*	*

Remarks: * = significant at α=0.05; ns = not significant at α=0.05; FL: fruit length; FWD: fruit width; FWG: fruit weight; WMF: wet mesocarp to fruit; DMF: dry mesocarp to fruit; NB: number of bunches; ABW: average bunch weight; FS: fruit set; ODM: oil to dry mesocarp; OWM: oil to wet mesocarp; FB: fruit to bunch; OB: oil to bunch

There was a significant effect of the single factor MCP, Ca^{2+}, and NAA on several variables, except FL (fruit length), FWD (fruit width), NB (number of bunches), and FS (fruit set). In this study, FWD (fruit width), NB (number of bunches), and FS (fruit set) were not affected by any treatments. The average of bunch weight (ABW) was only influenced by a single MCP treatment, in which the application of 300 ppm MCP reduced ABW, while other concentrations did not have a significant effect on ABW (data not shown). The application of these three exogenous compounds was carried out at 2 weeks after anthesis for NAA and Ca^{2+}, and 18 weeks after anthesis for MCP, so it would not affect NB and FS. Application of exogenous compounds after anthesis has the opportunity to change ABW, as revealed by the negative effect of MCP at a high concentration of 300 ppm. However, NAA and Ca^{2+}, either single or in interaction, did not affect ABW.

Interaction effects between factors, both 2 factors and 3 factors, were also observed for several variables, except FWD, NB, ABW, and FS. The interaction of the 3 factors of MCP, Ca^{2+} and NAA gave the most significant effect on several variables that represent fruit growth and quality, including FL (fruit length), FWG (fruit weight), WMF (wet mesocarp to fruit), DMF (dry mesocarp to fruit), ODM (oil to dry mesocarp), OWM (oil to wet mesocarp), and the very important variables FB (fruit to bunch) and OB (oil to bunch). Therefore, the discussion will be focused on the results of the interaction of these 3 factors.

Fruit Growth and Ratio Mesocarp to Fruit

The interaction effect of exogenous compounds 1-MCP, Ca^{2+} and NAA on fruit length, width and weight and ratio of mesocarp to fruit is presented in Table 2. The results showed an interaction between 1-MCP, $CaCl_2$ and NAA in almost all variables, except fruit width. In control plants grown on Ultisols, the average fruit length, fruit width and fruit weight were 2.96 cm, 2.13 cm and 10.29 g, respectively. Meanwhile, the ratio of mesocarp to fruit based on wet weight and dry weight in control treatment was 84.48% and 56.09%, respectively (Table 2). Under control conditions, the values of these variables for 5-year-old oil palm were slightly lower than normal according to the plantation observed data. Exogenous application of Ca^{2+}, NAA and 1-MCP, however, gave a positive effect on all variables in Table 2, except for fruit width.

Fruit Length

Four treatment combinations of exogenous compounds which gave significant higher fruit lengths compared to controls (Table 2) were (a) 50 ppm Ca^{2+}, (b) 100 ppm 1-MCP, (c) 100 ppm 1-MCP + 50 ppm Ca^{2+}, and (d) 200 ppm 1-MCP + 50 ppm Ca^{2+} + 200 ppm NAA, where these 4 treatments were able to increase fruit length by 12.2, 23.4, 70.8, 70.8%, respectively. Exposing fruits to the single and lowest level of 100 ppm 1-MCP or 50 ppm Ca^{2+} resulted in a significant and efficient increase in fruit length.

Fruit Width

There was no significant effect of either single or combination of three exogenous compounds (1-MCP, Ca^{2+} and NAA) on fruit width. The width of the fruit was in the range of 2.04 - 2.57 cm (Table 2).

Fruit Weight

There was an increase in the weight of oil palm fruit in all treatments of exogenous compounds 1-MCP, Ca^{2+} and NAA, either single or in combination treatment (Table 2). The highest fruit weight (13.27 g) was achieved at 50 ppm Ca^{2+} + 200 ppm NAA, which was not significantly different with 50 ppm Ca^{2+}, 100 ppm 1-MCP + 50 ppm Ca^{2+}, 200 ppm 1-MCP + 50 ppm Ca^{2+} + 200 ppm NAA, and 300 ppm 1-MCP + 50 ppm Ca^{2+} + 200 ppm NAA. The increase in fruit weight under these four treatments was quite high reaching 28.9%, 21.1%, 20.9% and 21.3%. The 50 ppm Ca^{2+} as single and the lowest level of exogenous compound was significant and most efficient in increasing the fruit weight.

Ratio Mesocarp to Fruit (WMF and DMF)

Exposing bunches to exogenous compound 1-MCP, Ca^{2+} and NAA has brought about the increase of ratio mesocarp to fruit, the increase of which occurred at both variables i.e. wet weight ratio (WMF, wet mesocarp to fruit) and dry weight ratio (DMF, dry mesocarp to fruit) of oil palm (Table 2). Table 2 revealed that all treatments of exogenous compounds 1-MCP, Ca^{2+}, and NAA enabled oil palm to increase WMF as well as DMF, except the single treatment of 200 ppm 1-MCP. Similar to other variables, the 50 ppm Ca^{2+} as a single and the lowest level of exogenous compound was significant and most efficient on increasing the ratio of mesocarp to fruit.

The results showed a positive effect of Ca^{2+}, NAA and 1-MCP on fruit length, and ratio mesocarp to fruit which led to an increase in fruit weight. There are few references regarding the positive effect of Ca^{2+} and NAA on increasing the size and weight of the fruit of other plants. The addition of Ca^{2+} to the flowers and young fruit of oil palm was reported to support the formation of fruit cell walls (Marschner, 2012). According to Marschner (2012), Ca^{2+} in fruit and seeds is less than in leaves because Ca^{2+} translocation in plant tissues is affected by the transpiration process which leads to a greater increase of Ca^{2+} accumulation in leaves than in other parts of the plant. In addition, Ca^{2+} is immobile in the phloem so that the availability of Ca^{2+} in plant tissues is very limited, thus the availability of Ca^{2+} is highly dependent on direct delivery from the xylem (Marschner, 2012). It is known that the need for auxin in young fruits and seeds is very high, in which Ca^{2+} could act as a stimulator of cell division and elongation (Taiz et al., 2015). Therefore, the addition of Ca^{2+} and NAA to bunches 1 week after anthesis was expected to increase the adequacy of Ca^{2+} and NAA needed for the development of oil palm fruit. Adequacy of nutrients in plant growth will synergize with plant growth regulators so that plant metabolism will take place more optimally. The increase in cell division and elongation due to the addition of Ca^{2+} and NAA would probably increase in the length and size of the fruit, which might increase the size of the mesocarp. According to Marschner (2012), Ca^{2+} accumulates a lot in the cell wall (apoplast), when a new cell wall is formed due to the addition of NAA, Ca^{2+} will easily occupy the newly formed apoplast. This synergistic effect has been proven in mangosteen. Tanari et al. (2018) used 4.8 kg/tree of Ca^{2+} and 200 ppm NAA through spraying on the mangosteen fruit had increased the quality of fruit with reduced total yellow sap in fruits.

Table 2. Effect of exogenous compounds on fruit length, width, weight and ratio of mesocarp to fruit

Treatments	Fruit length (cm)	Fruit Width (cm)	Fruit weight (g)	Ratio of Mesocarp to fruit	
				Wet weight ratio, WMF (%)	Dry weight ratio, DMF (%)
M0C0N0	2.96^{c}	2.13	10.29^{e}	84.48^{g}	56.09^{d}
M0C0N200	3.14^{bc}	2.04	11.44^{cd}	89.91^{ab}	63.80^{bc}
M0C50N0	3.52^{ab}	2.39	12.46^{ab}	89.12^{bcde}	66.60^{abc}
M0C50N200	3.07^{bc}	2.15	13.27^{a}	89.55^{abcd}	67.63^{ab}
M100C0N0	3.46^{ab}	2.28	11.69^{bcd}	87.97^{de}	66.20^{abc}
M100C0N200	3.26^{bc}	2.04	11.55^{bcd}	89.54^{abcd}	67.96^{a}
M100C50N0	3.64^{a}	2.31	12.45^{ab}	88.2^{cde}	63.62^{bc}
M100C50N200	3.10^{bc}	2.15	11.57^{bcd}	90.97^{a}	64.60^{abc}
M200C0N0	3.21^{bc}	2.14	11.31^{cd}	86.84^{f}	58.09^{d}
M200C0N200	3.12^{bc}	2.04	11.10^{cd}	88.22^{cde}	65.63^{abc}
M200C50N0	3.23^{bc}	2.23	11.76^{bcd}	89.46^{abcd}	65.84^{abc}
M200C50N200	3.64^{a}	2.57	12.47^{ab}	90.38^{ab}	66.02^{abc}
M300C0N0	3.34^{bc}	2.28	11.95^{bcd}	87.48^{e}	64.06^{abc}
M300C0N200	2.96^{c}	2.21	12.16^{bc}	87.47^{e}	63.74^{bc}
M300C50N0	3.21^{bc}	2.05	11.02^{cd}	88.15^{cde}	62.32^{c}
M300C50N200	3.33^{bc}	2.27	12.48^{ab}	88.7^{bcde}	63.89^{bc}

Remarks: The numbers followed by the same letters in the same column show no significant difference based on DMRT at the level α=5%; 1-MCP (M0 = 0 ppm, M100 = 100 ppm, M200 = 200 ppm and M300 = 300 ppm), $CaCl_2$ (C0 = 0 ppm and C50 = 50 ppm) and NAA (N0 = 0 ppm and N200 = 200 ppm).

Those treatments elevated Ca-pectate content in the pericarp and reduced the percentage of fruit contaminated with yellow sap compared to the control. The addition of NAA has been thought to cause an increase in cell division so that new apoplasts were formed in the fruit, which allows Ca^{2+} to occupy the newly formed apoplast. So, the fruit becomes a strong sink because the newly formed apoplast requires Ca^{2+} to structure its cell walls. The presence of Ca^{2+} which functions structurally strengthens the cell wall of mangosteen. Whether the effect of Ca^{2+} and NAA on mangosteen also occurred in oil palm fruits, however, it remains to be proven. These results in mangosteen gave rise to the use of Ca^{2+} and NAA in order to improve fruit growth and oil yield of oil palm grown on acidic soil Ultisol, which has a low fruit set and fruit to bunch. The result obtained indicated that the application of exogenous 1-MCP, Ca^{2+} and NAA could improve fruit growth and oil content in fruit of oil palm.

In this study, the increase in fruit weight can be attributed to the increase in fruit length and ratio of mesocarp to fruit (WMF and DMF). The highest fruit weight was 13.27 g at 50 ppm Ca^{2+} + 200 ppm NAA, while the control was 10.29 g (Table 2). Thus, it could be predicted that the higher fruit weight was supported by longer fruit size, and higher mesocarp weight to fruit due to higher cell division and elongation processes which were influenced by the addition of these two compounds, i.e Ca^{2+} and NAA. Purnama (2016) pointed out that Ca^{2+} has an important role in increasing the cell wall strength of fruits such as mangosteen. Rachmah et al. (2017) mentioned that Ca^{2+} plays a role in maintaining fruit quality and fruit growth of tomatoes. The number of normal tomatoes was higher in the Ca^{2+} treatment. Besides Ca^{2+}, according to Taiz et al. (2015) young fruits and seeds contain high auxin which functions to accelerate cell division and elongation. One of the auxin groups that play a role in accelerating the process of division and elongation of plant cells is NAA (Taiz et al., 2015). Several studies have proven that exogenous application of NAA could increase the fruit size and quality, such as in blueberries (Milić et al., 2018) and pumpkin (Chen et al., 2022), thereby increasing yield per plant. Chen et al. (2022) stated that the increase in pumpkin fruit size could be attributed to the effects of NAA and EBR in increasing source, translocation, and sink strength, which could promote the synthesis and distribution of assimilate in pumpkin. This study strengthens the conclusion that the role of NAA in fruit improvement in several plants is quite clear. This overall effect of NAA may promote the synthesis and distribution of assimilate, thereby increasing fruit size, which may also occur in oil palm. An increase in fruit length and weight in oil palm due to the addition of exogenous NAA was reported by Sangwiroonthon et al. (2017), although the mechanism was not explained.

The results indicated that either a single application of 100 ppm 1-MCP or combination with Ca^{2+} and NAA enabled oil palm to increase fruit length, fruit weight and ratio mesocarp to fruit when fruits were subjected to this exogenous substance 18 to 20 weeks after anthesis (about 5-6 weeks to fruit ripening). The 1-MCP compound is known as an anti-ethylene agent which works by blocking ligand receptors, which is often used as a plant regulator in cut flowers, nursery plants and foliage, as well as in stored fruit and vegetables. Tomala et al. (2020) showed that 1-MCP application as preharvest treatment delayed harvesting and reduced the quality deterioration during the storage of apples. There is no sufficient explanation to reveal the effect of 1-MCP on the improvement of oil palm fruit and its mechanisms. There is no reference explaining why in a short period (5-6 weeks) from the time of its application until the fruit is harvested, 1-MCP can increase fruit length, fruit weight and mesocarp size. Perhaps the application of 1-MCP 5-6 weeks before harvesting will delay harvesting, which allows the fruit to still increase in size for fruit length and mesocarp, thereby increasing fruit weight.

Number of Bunches, Average Bunch Weight and Fruit Set

Exogenous application of Ca^{2+}, 1-MCP and NAA did not affect the number of bunches per tree, average bunch weight and fruit set (Table 3). Few reports indicate that the formation of bunches, which will determine the number of bunches, is strongly influenced by the availability of the hormone auxin. There was no report, however, related to the importance of the presence of Ca^{2+} and 1-MCP, it seems that the role of Ca^{2+} is more to the formation of fruit cell walls, and 1-MCP contributes more to inhibiting ethylene in ripe fruit.

Regarding the role of NAA, Romero et al. (2021) stated that NAA is very important for inducing bunch formation, fruit growth and oil accumulation. Romero et al. (2021) showed that oil palm bunches could be induced by application of NAA before anthesis and up to 14 days after anthesis. According to Sotelo-Silveria et al. (2014), the effect of auxin on fruit formation in several plants is closely related to auxin levels before and after anthesis. In tomatoes, Kim et al. (2020) and Shinozaki et al. (2020) revealed that auxin level at two-days before anthesis was low and began to increase after anthesis, reaching a maximum value at four-days after anthesis, then decreased rapidly. In oil palms, not all NAA application trials were successful in inducing fruit, as happened in several plantations in Africa (Thomas et al., 1973). Failure may occur even if parthenocarpy is achieved when the hormone is used very close to anthesis (Somyong et al., 2018). In this study, application of NAA at anthesis might not be effective in inducing bunch number.

In oil palm, bunch weight is determined by several components, namely number of spikelets, number of flowers per spikelet, fruit set, weight per fruitlet, and non-fruit bunch component weight. Based on these components, it is likely that the low fruit set in this study caused the number of bunches and the average bunch weight to be less responsive to various treatments of exogenous compounds. Furthermore, the formation of fruit set was quite low, which occurred in all treatments and controls (< 60%), which could be related to low pollination due to low pollen availability, which might be caused by high $Al^{3+.}$ saturation in soil (>70%). Woittiez et al. (2017), however, stated that bunch weight and oil content were less affected by stresses, such as abiotic stress, than the number of bunches, but had a large impact on yield. Hidayah (2020) reported that oil palm planted in soil with high Al^{3+} saturation (> 80%) showed a very high number of abnormal bunches (rejected bunches > 60%). The application of Ca^{2+}, citric acid, B and Cu through basal drench was very effective in increasing the number of normal bunches and the oil content. Therefore, the normal number of bunches and bunch weight will be very important to achieve high oil production.

The result of the Pearson correlation test (Table 4) indicated that average bunch weight (ABW) was strongly influenced by the percentage of fruit set. The results of the correlation test between the average bunch weight (ABW) with several variables showed that the highest correlation coefficient was with the fruit set (R^2 = 0.664**). The correlation between the ABW variable and other variables such as fruit weight ratio to bunch was lower (R^2 = 0.479**), and it was even lower with the number of bunches (R^2 = -0.366**), oil ratio to mesocarp dry weight (R^2 = 0.356**) and fruit length (R^2 = 0.299**) (Table 4).

Table 3. Effect of exogenous compounds on the number of bunches, average bunch weight and fruit set

Treatments	Number of bunches	Average bunch weight (kg)	Fruit set (%)
M0C0N0	9.0	6.55	42.0
M0C0N200	9.1	7.77	35.0
M0C50N0	8.9	7.90	45.0
M0C50N200	8.7	7.68	39.0
M100C0N0	8.2	7.56	40.0
M100C0N200	8.8	6.40	40.0
M100C50N0	8.5	7.58	39.0
M100C50N200	8.4	7.49	42.0
M200C0N0	8.3	8.06	50.0
M200C0N200	8.2	7.28	37.0
M200C50N0	8.6	7.10	31.0
M200C50N200	8.7	7.68	36.0
M300C0N0	9.6	6.63	32.0
M300C0N200	9.5	6.39	40.0
M300C50N0	8.2	6.49	32.0
M300C50N200	8.1	6.67	38.0

Remarks: 1-MCP (M0 = 0 ppm, M100 = 100 ppm, M200 = 200 ppm and M300 = 300 ppm), $CaCl_2$ (C0 = 0 ppm and C50 = 50 ppm) and NAA (N0 = 0 ppm and N200 = 200 ppm)

Fruit to Bunch and Oil Content

Exposing flowers to Ca^{2+} and NAA and fruits to exogenous compound 1-MCP increased oil content in fruit mesocarp (ODM, oil to dry mesocarp, and OWM, oil to wet mesocarp), fruit to bunch (FB), and oil to bunch (OB) (Table 5). This evidence was exhibited by several treatments, either alone or in combination of exogenous compounds as illustrated below.

Oil Ratio in Mesocarp (OWM and ODM)

The value of OWM was elevated by all treatments (Table 5), followed by an increase of ODM in all treatments, except for the treatment of a single 300 ppm 1-MCP and 300 ppm 1-MCP + 50 ppm Ca^{2+}. The best treatment for increasing ODM was 100 ppm 1-MCP + 50 ppm Ca^{2+} + 200 ppm NAA, which was not significantly different from a single treatment of 100 ppm 1-MCP; and 200 ppm 1-MCP + 50 ppm Ca^{2+} + 200 ppm NAA. The highest increase of which, however, was about 8.1% of the ODM, from 75.83% in control to 82.0%.

Fruit to Bunch (FB)

Four treatments of exogenous compounds enabled oil palm trees grown on Ultisol to improve the value of FB (Table 5), namely a single treatment of 50 ppm Ca^{2+}; 50 ppm Ca^{2+} + 200 NAA; 100 ppm 1-MCP + 200 ppm NAA, and a single treatment of 200 ppm 1-MCP.

Table 4. Summary of *Pearson correlation test*

Variable	Fruit set	Fruit to bunch	DMF	OWM
ABW	0.664**	0.479**	0.076tn	0.167tn
Oil to bunch	0.354*	0.637**	0.321*	0.416**

Remarks: ** = Correlation is significant at the 0.01 level (2-tailed); * = Correlation is significant at the 0.05 level (2-tailed); tn is not significant at the 0.05 level (2-tailed); ABW = Average bunch weight; DMF = dry mesocarp to fruit; OWM = oil to wet mesocarp.

Table 5. Effect of exogenous compounds on fruit to bunch and oil content

Treatments	Oil ratio in mesocarp		Fruit to bunch (FB) (%)	Oil to bunch (OB) (%)
	ODM (%)	OWM (%)		
M0C0N0	75.83^{f}	50.61^{f}	51.67^{bc}	21.65^{c}
M0C0N200	80.04^{bc}	56.8^{cd}	48.67^{bc}	24.74^{bc}
M0C50N0	79.05^{bcd}	57.37^{bcd}	65.67^{a}	33.96^{a}
M0C50N200	79.21^{bc}	56.99^{cd}	66.33^{a}	33.9^{a}
M100C0N0	80.54^{ab}	60.5^{a}	56.67^{b}	30.16^{ab}
M100C0N200	78.16^{cde}	59.32^{ab}	65.33^{a}	34.73^{a}
M100C50N0	79.38^{bc}	56.78^{cd}	49.00^{bc}	22.74^{ab}
M100C50N200	82.00^{a}	57.23^{cd}	55.33^{b}	28.51^{ab}
M200C0N0	78.78^{bcd}	55.91^{d}	65.67^{a}	30.10^{ab}
M200C0N200	78.80^{bcd}	57.87^{bcd}	52.67^{bc}	26.86^{ab}
M200C50N0	78.02^{cde}	58.33^{bc}	41.00^{c}	21.61^{c}
M200C50N200	80.69^{ab}	57.71^{bcd}	48.33^{bc}	24.99^{bc}
M300C0N0	77.25^{ef}	56.3^{cd}	51.67^{bc}	25.42^{bc}
M300C0N200	78.32^{cde}	57.11^{cd}	55.67^{b}	27.89^{bc}
M300C50N0	77.47^{ef}	53.91^{e}	45.00^{c}	21.59^{bc}
M300C50N200	79.3^{bc}	56.99^{cd}	59.67^{ab}	30.54^{ab}

Remarks: The numbers followed by the same letters in the same column show no significant difference based on DMRT at the level α= 5%; ODM = Oil to dry mesocarp; OWM = oil to wet mesocarp; 1-MCP (M0 = 0 ppm, M100 = 100 ppm, M200 = 200 ppm and M300 = 300 ppm), $CaCl_2$ (C0 = 0 ppm and C50 = 50 ppm) and NAA (N0 = 0 ppm and N200 = 200 ppm)

Oil to Bunch (OB)

Only 6 treatments had no effect on oil to bunch (OB) (Table 5), while the other 9 treatments had a positive effect in improving OB of oil palm trees grown on acidic soil. The best treatment was a single treatment of 50 ppm Ca^{2+} and 50 ppm Ca^{2+} + 200 ppm NAA, which resulted in the highest increase of ratio oil to bunch that reaching about 56.8%, a value that promises benefits. Similar to other variables, the 50 ppm Ca^{2+} as a single and the lowest level of exogenous compound was also significant and most efficient in increasing the ratio of fruit to bunch and oil content.

This study showed that the effect of exogenous compounds on the oil-to-bunch ratio (OB) had a high variation. This was presumably due to a high variation of FB (fruit to bunch) between treatments. Although the difference in ODM (oil to dry mesocarp) in the treatment of exogenous compounds (1-MCP, Ca^{2+} and NAA) with the control was quite significant, it could not directly determine the value of OB. It has been understood that the value of OB will be influenced by many factors. According to Mandal & Babu (2008), the value of OB will be determined by the weight of bunches, FB, ODM (oil to dry mesocarp) and OWM (oil to wet mesocarp) as it can be observed in Table 4 which explained the results of Person correlation test.

The correlation test between OB and other variables showed a positive and significant relationship. The correlation between OB and FB was the highest (R^2 = 0.637**), followed by other variables with smaller correlation values. The correlation between OB and OWM was R^2 = 0.416**, with fruit set was R^2 = 0.354*, and with DMF (dry mesocap to fruit) was R^2 = 0.321*. It could be observed that results in Table 5, that several treatments which had higher FB than control generally produced significantly higher OB than control. The results of this study were in agreement with Corley & Tinker (2015), that the value of OB will increase if the ratio of fruit weight to bunch (FB) and fruit set increases. These four treatments, namely 50 ppm Ca^{2+} as a single treatment; 50 ppm Ca^{2+} + 200 ppm NAA; 100 ppm 1-MCP + 200 ppm NAA, and a single treatment of 200 ppm 1-MCP had a higher FB value than control and exhibited also a higher OB than control. The best treatment (single 50 ppm Ca^{2+}, and 50 ppm Ca^{2+} + 200 ppm NAA) achieved an OB value of 33.9 % which was 56.9% higher than the control. In this experiment, however, there was no effect of application of these exogenous compounds on fruits set.

In this study, the positive effects of 1-MCP, Ca^{2+} and NAA in increasing oil yield, which were represented by ODM (oil to dry mesocarp) and OB (oil to bunch), were in accordance with several previous studies in oil palm. Rath et al. (2017) reported that 1-MCP was able to increase the oil yield when applied to the fruit before harvest. Romero et al. (2021) showed an increase of oil yield by adding NAA up to 1200 ppm. So far, there has been no report regarding the positive effect of Ca^{2+} on the increase of ODM or OB. In this study, however, the positive effect of Ca^{2+} either alone or in combination could be observed. The best treatment that provided the highest ODM was 100 ppm 1-MCP + 50 ppm Ca^{2+} + 200 NAA. The highest oil to bunch (OB) was obtained in the single treatment of 50 ppm Ca^{2+}, and 50 ppm Ca^{2+} + 200 ppm NAA. Regarding the effect of NAA on oil yield, the trials can be carried out with higher NAA concentrations up to 1200 ppm, as recommended by Romero et al. (2021). With a total dose of 1200 ppm NAA, applied 3-4 times, it was able to produce a better fruit set, similar average bunch weight, and oil to dry mesocarp than those obtained with assisted pollination. It was also reported that NAA increased oil to bunch in 36% compared to control (Romero et al., 2021). Romero et al. (2021) using NAA 1200 ppm, and in this experiment using 200 ppm NAA as a single treatment resulted in lower number of bunches (9.1), bunch weight (7.77 kg) and fruit set 35%. Moreover, a single treatment of 200 ppm NAA had no significant effect on oil to bunch compared to control (24.74 % versus 21.65%). By comparing the results of the two studies, it could be proposed to increase the concentration of NAA up to 1200 ppm in future studies. This research has shown that addition of NAA stimulated fruit growth resulting in an increase in fruit weight, fruit length, and fruit width, thereby increasing fruit to bunches. Basuchaudhuri (2016) suggested that NAA plays a role in stimulating cell division, cell elongation, photosynthesis and fruit formation as well as increasing the production of fruit plants. It is suspected that NAA can increase dissolved carbohydrates in fruit (Singh et al., 2018), leading to an increase in fruit volume and oil content in treated fruit. The oil content in the fruit begins to increase when the fruit is formed until it reaches the maximum level.

Regarding the application of 1-MCP, it could affect the ethylene receptors and block them, thus resulting in a possible delay in harvest, hence an increase in oil content might be obtained. On the other hand, some available literature showed that the use of ethylene in oil palm fruit could also increase the oil content. However, the effect of ethylene on increasing the oil content of palm fruit is still unclear. Chan et al. (1972) showed an increase in oil content in fruit due to the treatment of ethephon and other hormones. In that study, exogenous application on fruits with NAA, GA, or ethephon at anthesis (5-day intervals until abscission) inhibited fruit abscission, thus delaying harvest by up to 5 days without an increase in the number of detached fruits. Moreover, oil yield increased by more than 5%, with no change in harvesting costs or oil quality. It has been reported that ethylene has a role in triggering oil accumulation and fruit ripening in several plants, such as in oil palm (Abdul Wahab et al., 2020) and tomatoes (Brumos, 2021; Herrera-Ubaldo, 2022). The role of ethylene in the ripening process of oil palm fruit is related to the expression of the EgERD3 gene in mesocarp tissue, where the expression of this gene follows the ripening pattern of oil palm fruit. The EgERD3 gene plays a role in ethylene synthesis and signaling pathways in the mesocarp (Nurniwalis et al., 2018). It is known that 1-MCP is an anti-ethylene agent which works by blocking ligand receptors. Therefore, it is predicted that the application of the ethylene antagonist 1-MCP will have the opposite effect to ethylene on the change of oil content. On the contrary, however, Rath et al. (2017) found an increase of oil content when 100 ppm 1-MCP was applied before harvest of the fruits of oil palm. It seems that there is a high correlation between endogenous ethylene and oil content of oil palm fruit as posited by Tranbarger et al., (2011). Tranbarger et al. (2011) stated that the key phases of oil palm mesocarp development are characterized by morphology, cellular, biochemical and hormonal. In the maturation phase around 120 DAP (day after pollination) ethylene biosynthesis and oil content in the mesocarp increase simultaneously. The peak increase of endogenous ethylene is approximately at the beginning of ripening at 140 DAP and stops at around 150 DAP, while the oil content continues to increase until its peak at around 160 DAP (Tranbarger et al., 2011). Therefore, application of 1-MCP at 126 DAP and 136 DAP in this study might be effective in inhibiting ethylene biosynthesis, so that fruit ripening and aging could be prevented. This condition is thought to have contributed to a more optimal synthesis of oil in the fruit, thereby increasing fruit weight and oil content. In another study, the application of anti-ethylene 1-MCP on freshly harvested fruit showed no effect on the total oil content, even though there was a delay in the ripening of the fruit. In that study, Nualwijit et al. (2013) showed that the application of 1-MCP to freshly harvested fruit caused a delay in the ripening of oil palm fruit, which was characterized by a change in the color of the fruit skin from black to reddish orange. However, there was no change in the total oil content of the fruit, while the percentage of FFA (free fatty acids) in fruit treated with 1-MCP was lower than the control. In conclusion, the application of 1-MCP has the potential to maintain FFA levels in palm fruit after harvest, and allows longer transportation times from fields to producers. It can be concluded that the application time of 1-MCP as well as ethylene on fruit is very crucial and determines fruit ripening and oil content in fruits. Application of 1-MCP at anthesis until before abscission might stimulate an increase in oil content, whereas application of 1-MCP to freshly harvested fruit only affects the delay in fruit ripening without changes in oil content

CONCLUSION AND SUGGESTION

The exogenous application of Ca^{2+}, NAA and 1-MCP either single or in combination enabled oil palm tree grown on Ultisol to improve fruit growth and oil yield by increasing fruit length, fruit weight, ratio mesocarp to fruit, OWM (oil to wet mesocarp), ODM (oil to dry mesocarp), FB (fruit to bunch) and OB (oil to bunch). The best treatment for increasing ODM was 100 ppm 1-MCP + 50 ppm Ca^{2+} + 200 ppm NAA. Meanwhile, to get the highest oil to bunch (OB), the combination of 100 ppm 1-MCP + 200 ppm NAA, 50 ppm Ca^{2+} + 200 ppm NAA and a single treatment of 50 ppm Ca^{2+} were very promising to be utilized. However, from all of the above treatments, the 50 ppm Ca^{2+} as single and the lowest level of the exogenous compound was significantly and most efficient in improving fruit growth and oil yield of oil palm. Moreover, the addition of surfactants to the solution of exogenous compounds is seen to be able to increase the effectiveness in increasing oil yields.

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