

Bark Recovery of Rubber Clone PB 260 Through Application of NAA⁺, Kinetin and Oleo-chemicals

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Abstract. Clone PB 260 firstly planted in 1975 and popular due to its excellent vigor and productivity during maturity (1.5-2 tonnes/ha¹⁰), the dense foliage reduced extensive weeding and is resistance against major leaf diseases. The knowledge on the bark recovery rate is very important to maintain high yield of latex production on rubber plantation. The study which was conducted on the experimental farm of Rubber Research Institute, Sungei Putih, Deli Serdang, north Sumatra, Indonesia was aimed to obtain the optimum combination concentration of growth regulators (NAA⁺ and kinetin) and various oleo-chemicals to accelerate bark recovery and response of bark recovery rate for rubber clones PB 260. Factorial randomized block design, which consists of two factors, namely growth regulator concentrations (N₁=50 ppm NAA⁺ + 50 ppm kinetin; N₂=100 ppm NAA⁺ + 50 ppm kinetin; N₃=150 ppm NAA⁺ + 50 ppm kinetin) and oleo-chemicals (O₁=palmitic acid; O₂=oleic acid and O₃=stearic acid) were used in this study. The parameters measured were the number of latex vessels, skin thickness, sucrose levels, thiol levels, inorganic phosphate level, latex yield and total solid content (TSC). The results showed that growth regulator concentrations significantly affect the number of latex vessels but did not significantly affect the skin thickness, sucrose levels, thiol levels, inorganic phosphate levels, latex yield and total solid content (TSC). The most optimum growth regulator combination was N₂ at various parameters observed. Oleo-chemicals application showed significant effect on the number of latex vessels, skin thickness and latex yield, but showed no significant effect on levels of sucrose, thiol levels, inorganic phosphate level and total solid content (TSC). Thus, application of growth regulator and oleo-chemicals are beneficial to accelerate bark recovery rate in order to maintain latex production on rubber plantation.

Keywords: clone PB 260, oleo-chemicals, NAA, latex physiology

Introduction

Latex of rubber tree (*Hevea brasiliensis*) is one of the leading plantation commodities that have provided large foreign exchange for Indonesia. In 2014 alone, latex export generated approximately 4.3 billion US dollars (Tajulfitri, 2019). Indonesia with its 3.67 million ha in 2017 has the largest rubber plantation area in the world. However, in terms of production, Thailand leads at 3.23 million tons and Indonesia come in second place (Apriansyah and Sohieben, 2019; Tania *et al.*, 2017).

Rubber tree economic life is upto 25 years with production as much as 36 tonnes per hectare. Approximately 79% of production comes from small holder plantation, 10% of state owned enterprise plantations and 11% from private estates. Several factors

have been known to lower latex production such as white root fungus (WRF), tapping panel dryness (DTP) which reduced economics life of rubber tree in the field to averaged only 15 years. Maturity of ready-to-tap rubber tree reached at 6-7 years. Main characteristics of mature rubber tree observed as girth 45 cm at 100 cm height of the grafting link (elephant root). If the bark has reached the thickness of 6-7 mm, the tree is considered ready to be tapped (Rubber Research Center, 2014). However, if tapping conducted at rubber trees under 6 years old, the bark is still too thin (3-4 mm) and the tapping process will resulted in wound of the wood (Arja and Supijatno, 2018).

Tapping of natural rubber tree responsible for 90% of rubber productions in the world, which produced from secretory structures called laticifers. Numbers of laticifers determines the rubber yield, thus varied clones

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has been created (Silva *et al.*, 2021). Clone PB 260 is one of the clone created with high rubber yield and its characteristics makes it favourable in many plantations. Tapping, if conducted properly, would last for 20-30 years. Therefore, it is important to ensure excellent bark formation and good bark recovery if wound is happened (Yusriadi and Ikramullah, 2017).

Bark is very valuable for rubber plantation companies. Therefore, it must be managed properly. After tapping, the bark will recover after six years if all precautions are carried out properly. In practice, the recovered bark can be tapped again after eight to nine years. It is necessary to pay attention on bark recovery of the tapping panel as any mistakes in determining the tapping system and tapping depth will cause abnormal bark recovery. This will affect tree health and in turn, tree productivity. Thickness of recovered bark at minimum 7 mm determined that the recovered bark is suitable for re-tapping (Arrasyid *et al.*, 2018). Acceleration of bark recovery is extremely crucial in rubber plantation industry which includes standard tapping, fertilizing, maintaining regular tapping panel and other cultivation techniques. However, study on cambium activity after wounded is still very limited.

Recovery and bark recovery are crucial to ensure continuous flow of nutrients, water and sugar as raw materials in rubber formation. According to Belleville *et al.* (2020), rapid recovery of bark will resulted in re-tapping after bark thickness reach at least 7 mm within 7-8 years. Acceleration of bark recovery effort nevertheless critical for continuous production, along with fertilization, care of tapping area and other farming techniques. But efforts or research leading to or directly influence the activity of cambium is still limited.

Cambium growth as secondary meristem activity resulted in formation of bark tissue. In order to increase cambium activity, growth regulator and exogenous oleo-chemicals is needed. Moreover, open xylem and phloem of rubber (dicotyle) permitted the use of growth regulator to modulate cambium growth (Lopes *et al.*, 2020).

Previous research has found that the use of exogenous growth regulators on rubber may affect the activity of cambium. Chiatante *et al.* (2018) stated that the cambium activity depends on factors such as growth substance. Cambium cell division is influenced by auxin and cytokinins. Accordingly by (Sorice *et al.*, 2013; Du and Groover, 2010) stated that IAA regulator can increase the activity of the cambium. (Immanen *et al.*, 2016; Li

et al., 2013; Popko, 2010) concluded that regulator IAA can increase the activity of cambium, RNA and protein synthesis.

Good phloem growth is crucial to ensure latex vessel activity and increase the production. Growth regulator such as IAA reported to increase phloem growth rate. Mean while, Koryati and Elfiana (2019) reported that application of IAA, kinetin and paclobutrazol significantly affect girth growth of selected clone of rubber plants. Furthermore, oleo-chemicals are also known able to restore damaged tissue due to disease. Devappa *et al.* (2015) reported oleo-chemical application can increase tissue growth on the recovered bark of rubber tree with an average percentage is 40. Oleo-chemicals are industrial raw materials produced from vegetable oil, namely palm oil which contains glycerides of oleic acid, linolic acid (approximately 50%), glycerides of palmitic acid (25%), stearin acid (3-5%) and glycolglycerin acid (0.1%). This study aims to obtain the optimal combinations of NAA⁺, kinetin and oleo-chemicals to accelerate bark recovery and also to determine the response of accelerated bark recovery process on clone PB 260.

Materials and Methods

The research was conducted at the experimental farm of Rubber Research Institute of Sungei Putih, Deli Serdang Regency, north Sumatra province at 25 m above sea level with Ultisol soil type. Six years old clone PB 260, planted with spacing of 5 m x 2.5 m and tapping field which were at panel B0-1 (first bark care) were used in this research. NAA⁺, kinetin, palmitic acid, oleic acid and stearic acid used as bark wound chemical treatments. Physiological analysis uses trichloroacetic acid (TCA), H₂SO₄ 70%, dithio-bisnitro benzoic acid (DNIB) and selenomethionine.

Measuring tape, Tap SP, light microscope, tapping knives, containers, gutters, buckets, brushes, stationery, red green and yellow paint, analytical/digital balance, cameras and other supporting tools were used as well as analytical balance, beaker glass, test tubes, oven, spectrophotometer Beckleman DU 650, mortar and grinder for physiological analysis.

Research design. The design is factorial randomized block design, which consists of two factors, namely growth regulator concentrations (N) and oleo-chemicals type (O). Growth regulator concentrations (N) consists of 4 levels, namely: 0 ppm (N₀), NAA⁺50 ppm + 50

ppm kinetin (N_1), NAA+100 ppm + 50 ppm kinetin (N_2), NAA+ 150 ppm + 50 ppm kinetin (N_3). Oleo-chemicals (O) types consists of three levels namely no Oleochemicals (O_0), palmitic acid (O_1), oleic acid (O_2) and stearic acid (O_3).

NAA+ and oleo-chemicals are used in liquid form diluted growth regulators and oleo-chemicals were weighed according to the specified concentrations. NAA+ was dissolved in 60% ethanol, stirred until fully dissolved. The solution then mixed with selected oleo-chemicals, shaken using-magnetic stirrer for ± 30 min until homogeneous.

The mixtures were then applied using smearing technique two times a month at the tapped path of rubber stems. Parameters observed were increase of skin thickness (mm), number of latex vessels (once for three months), while sucrose, inorganic phosphate and thiol concentration were measured once a month as well as yield at tapping time.

Statistical analysis. Analysis of variance of experimental data were determined using the F test at 5% level of confidence which were then followed by Duncan's Multiple Range Test (DMRT) at the 5% level of confidence if the result showed significant difference.

Results and discussion

Number of latex vessels. The analysis showed that a treatment of NAA+ and kinetin at various concentrations and different oleo-chemicals application has various effects on number of latex vessels after three months of application. The average number of rubber latex vessels at 3 months after application is showed at Table 1.

N_1 and N_2 treatments resulted in highest latex vessels number per mm^2 and the results are not statistically different, indicating the presence of growth regulators at certain concentrations are crucial in latex vessels development, as shown in Table 1, while palmitic acid application affected latex vessels development even without growth regulator treatment better than oleic acid and stearic acid treatments as indicated in Table 1. It is interesting to highlight that N_0 and O_0 treatments showed the lowest latex vessel numbers, indicating NAA+and kinetin presence are crucial for latex vessel development. Observation of interaction between factors showed that N_1O_1 , N_2O_1 , N_3O_1 showed the highest latex vessels/ mm^2 and the results are not statistically different.

It is interesting to note that palmitic acid presence along with growth regulators (at all formulations concentration) resulted in highest latex vessel development/ mm^2 (Table 1).

Addition of bark thickness. The analysis showed that oleo-chemicals treatments showed significant effect on some parameter observes but the application of growth regulators and interaction of both factors showed insignificant effect on addition of bark thickness three months after application. The average of bark thickness addition of three months after application is presented in (Table 2).

Table 2 showed that growth regulators concentration showed insignificant effect on increase of bark thickness. Instead, oleo-chemicals types significantly affected the increase of bark thickness in which stearic acid application showed the highest bark thickness increment. At factors interaction, it is shown that stearic acid at N_3

Table 1. Latex vessels number by growth regulator application and oleo-chemicals treatment after three months application

Treatments	Latex vessel number/ mm^2
Growth regulator concentration N_0 (control)	16.33 ^c
N_1 (50 ppm NAA ⁺ +50 ppm kinetin)	20.92 ^a
N_2 (100 ppm NAA ⁺ +50 ppm kinetin)	21.50 ^a
N_3 (150 ppm NAA ⁺ +50 ppm kinetin)	19.58 ^b
Oleo-chemical types	
O_0 (control)	15.83 ^c
O_1 (palmitic acid)	23.08 ^a
O_2 (oleic acid)	19.67 ^b
O_3 (stearic acid)	19.75 ^b
Factors interaction	
N_0O_0	
N_1O_1	12.67 ^g
N_1O_2	24.33 ^a
N_1O_3	23.00 ^b
N_2O_1	24.67 ^a
N_2O_2	21.67 ^c
N_2O_3	19.67 ^d
N_3O_1	24.33 ^a
N_3O_2	14.67 ^f
N_3O_3	21.67 ^c

Note: The values followed by not the same letter in the same treatment group were significantly different at 5% level of DMRT. Values that are not lettered are not significantly different

Table 2. Bark thickness by growth regulators application and oleo-chemicals treatments after three months application

Treatments	Increase of bark thickness (mm)
Growth regulator concentration	
N ₀ (control)	1.09
N ₁ (50 ppm NAA ⁺ +50 ppm kinetin)	1.08
N ₂ (100 ppm NAA ⁺ +50 ppm kinetin)	1.09
N ₃ (150 ppm NAA ⁺ +50 ppm kinetin)	1.09
Oleo-chemicals type O ₀ (control)	
O ₁ (palmitic acid)	1.07 ^c
O ₂ (oleic acid)	1.12 ^b
O₃ (stearic acid)	1.16^a
Interactions	
N ₀ O ₀	1.00
N ₁ O ₁	1.04
N ₁ O ₂	1.09
N ₁ O ₃	1.18
N ₂ O ₁	1.10
N ₂ O ₂	1.11
N ₂ O ₃	1.15
N ₃ O ₁	1.00
N ₃ O ₂	1.22
N ₃ O ₃	1.14

Note: The values followed by not the same letter in the same treatment group were significantly different at 5% level of DMRT. Values that are not lettered and not significant.

concentration of growth regulators used, showed the highest bark thickness increased, followed by oleic acid treatments at N₁ and N₂ concentrations, even though the values are not statistically different.

Sucrose levels. The analysis showed that growth regulator concentrations and oleo-chemicals treatments and the interaction of both factors did not significantly affect levels of sucrose three months after application. The average of sucrose levels at three months after application is presented in Table 3.

Treatment of oleo-chemicals types has no significant effect on levels of sucrose (Table 3). There are no statistical differences observed on growth regulator concentration to sucrose level observed, while the application of oleic acid and palmitic acid markedly resulted with highest sucrose levels (1.96 mM and 1.95 mM, consecutively) to other oleo-chemicals treatment. The factors interaction, no trends observed on sucrose levels.

Table 3. The average of sucrose levels (mM) by growth regulator and oleo-chemicals treatments after three months of application

Treatments	Sucrose level (mM)
Growth regulator concentration	
N ₀ (control)	1.99
N ₁ (50 ppm NAA ⁺ +50 ppm kinetin)	1.91
N ₂ (100 ppm NAA ⁺ +50 ppm kinetin)	1.80
N ₃ (150 ppm NAA ⁺ +50 ppm kinetin)	1.88
Oleo-chemicals type	
O ₀ (control)	1.82
O₁ (palmitic acid)	1.95
O₂ (oleic acid)	1.96
O ₃ (stearic acid)	1.86
Factors interaction	
N ₀ O ₀	1.99
N ₁ O ₁	1.98
N ₁ O ₂	1.75
N₁O₃	2.01
N ₂ O ₁	1.82
N₂O₂	2.10
N ₂ O ₃	1.83
N ₃ O ₁	2.08
N₃O₂	1.78
N ₃ O ₃	1.73

Note: The values that are not lettered and not significant.

Thiol levels. Table 4 presented average thiol levels after three months of NAA⁺ and oleo-chemical applications. Results indicated that oleo-chemical types, instead of NAA⁺ concentrations, affecting thiol level concentrations on samples observed. O₁ in general showed highest thiol levels at N₂ and N₀ treatments, consecutively.

Table 4 shows that growth regulator concentrations have no significant effect on levels of thiol. The highest thiol level were found in NAA⁺ 50 ppm + 50 ppm (N₁) namely 0.73 mM, followed by NAA⁺ 0 ppm (N₀), 0.71 mM, NAA⁺ 100 ppm + 50 ppm (N₂), 0.69 mM and NAA⁺ 150 ppm + 50 ppm (N₃), 0.66 mM.

Inorganic phosphate (Pi). Inorganic phosphate is an indicator of metabolic activities which represents the plant's ability to change the raw materials (sucrose) to rubber particles. Statistical analysis showed that growth regulator concentrations insignificantly affecting inorganic phosphate level at the first to fourth month after application, as well as oleo-chemicals treatments. Moreover, analysis of factors combination effect showed insignificant results after three months application (Table 5).

Table 4. Thiol levels by growth regulator and oleo-chemicals treatments after three months application

Treatments	Thiol levels (mM)
Growth regulator concentration	
N ₀ (control)	0.71
N ₁ (50 ppm NAA ⁺ +50 ppm kinetin)	0.73
N ₂ (100 ppm NAA ⁺ +50 ppm kinetin)	0.69
N ₃ (150 ppm NAA ⁺ +50 ppm kinetin)	0.66
Oleo-chemicals type	
O ₀ (control)	0.71
O ₁ (palmitic acid)	0.72
O ₂ (oleic acid)	0.65
O ₃ (stearic acid)	0.70
Factors interaction	
N ₀ O ₀	0.68
N ₁ O ₁	0.73
N ₁ O ₂	0.72
N ₁ O ₃	0.76
N ₂ O ₁	0.77
N ₂ O ₂	0.61
N ₂ O ₃	0.65
N ₃ O ₁	0.65
N ₃ O ₂	0.57
N ₃ O ₃	0.69

Note: The values that are not lettered are not significantly different.

Latex production. The analysis showed that oleo-chemicals application significantly affecting latex production but NAA⁺ concentration and the interaction of both factors has no significant effect on the production of latex at three months after application. The average production of latex at three months after application is presented in Table 6.

Table 6 show that NAA⁺ concentrations have no significant effect on the latex production of the samples observed. The highest latex production obtained in (N₀) namely 40.19 g/tree/tapping, followed by (N₁), 39.26 g/tree/tapping; (N₃), 37.89 g/tree/tapping and (N₂), 37.77 g/tree/tapping consecutively, while oleo-chemicals treatments have no significant effect on the latex production (Table 6). The highest latex production was obtained in application of palmitic acid (O₁), namely 42.85 g/tree/tapping. This value is significantly different with stearic acid (O₃), namely 40.13 g/tree/tapping; oleic acid (O₂), 38.04 g/tree/tapping and no oleo-chemical (O₀), 34.09 g/tree/tapping.

Total solid content (TSC). Total solid content(TSC) is a reflection of the in site Latex biosynthetic ability.

Table 5. Inorganic phosphate content by growth regulator and oleo-chemicals treatments after three months application

Treatments	Inorganic phosphate (%)
Growth regulator concentration	
N ₀ (control)	3.42
N ₁ (50 ppm NAA ⁺ +50 ppm kinetin)	3.42
N ₂ (100 ppm NAA ⁺ +50 ppm kinetin)	3.58
N ₃ (150 ppm NAA ⁺ +50 ppm kinetin)	3.26
Oleo-chemicals type	
O ₀ (control)	3.36
O ₁ (palmitic acid)	3.51
O ₂ (oleic acid)	3.40
O ₃ (stearic acid)	3.41
Factors interaction	
N ₀ O ₀	3.47
N ₁ O ₁	3.32
N ₁ O ₂	3.58
N ₁ O ₃	3.51
N ₂ O ₁	3.77
N ₂ O ₂	3.35
N ₂ O ₃	3.62
N ₃ O ₁	3.58
N ₃ O ₂	3.14
N ₃ O ₃	3.19

Note: The values that are not lettered are not significantly different.

The analysis showed that NAA⁺ concentrations and types of oleo-chemicals and the interaction did not significantly effect on TSC three months after application. The average of TSC at three months after application is presented in Table 7.

Table 7 show that NAA⁺ concentrations have no significant effect on TSC. The highest TSC was obtained in N₀(39.67 g/p/s), followed by N₂(39.20 g/p/s), N₁(39.02 g/p/s) and N₃(38.85 g/p/s).

Treatments of oleo-chemicals types have no significant effect on TSC (Table 7). The highest TSC was obtained in application stearic acid (O₃) which was 9.43 g/p/s, followed by palmitic acid (39.24 g/p/s), no oleo-chemicals (O₀), 39.07 g/p/s and oleic acid (O₂), 38.99 g/p/s.

Results and Discussion

Effect of growth regulators concentration on bark recovery of rubber clones PB 260. The results showed that formulation of growth regulators concentrations has significant effect on the number of latex vessels,

Table 6. Latex production by NAA⁺ and oleo-chemicals treatment after three months application

Treatments	Production yield (g/p/s)
Growth regulators concentration	
N ₀ (control)	40.19
N ₁ (50 ppm NAA ⁺ +50 ppm kinetin)	39.26
N ₂ (100 ppm NAA ⁺ +50 ppm kinetin)	37.77
N ₃ (150 ppm NAA ⁺ +50 ppm kinetin)	37.89
Oleo-chemicals type	
O ₀ (control)	34.09 ^d
O ₁ (palmitic acid)	42.85 ^a
O ₂ (oleic acid)	38.04 ^c
O ₃ (stearic acid)	40.13 ^b
Factors interaction	
N ₀ O ₀	34.97
N ₁ O ₁	40.92
N ₁ O ₂	38.90
N ₁ O ₃	43.41
N ₂ O ₁	46.20
N ₂ O ₂	33.54
N ₂ O ₃	38.31
N ₃ O ₁	41.92
N ₃ O ₂	40.28
N ₃ O ₃	34.81

Note: The values followed by not the same letter in the same treatment group were significantly different at 5% level of DMRT. Values that are not lettered are not significantly different.

but showed no significant effect on the level of bark thickness, sucrose and thiol levels, inorganic phosphate levels, latex production and TSC.

In this study, the effects of formulation of three concentrations of growth regulators were tested at 0 ppm, 50 ppm NAA⁺+ 50 ppm kinetin, 100 ppm NAA⁺+ 50 ppm kinetin and 150 ppm NAA⁺+ 50 ppm kinetin on rubber bark recovery. Bark recovery shows linear response to the increase of growth regulators, particularly to NAA⁺ concentrations due to its function in cell division. NAA⁺ encourages cell division of one cell into two cells and so on. As the plant cell numbers increase, then tree will grow taller and have wider leaves (Schaller *et al.*, 2015).

According to Setiawati *et al.* (2018), plant requires addition of growth regulator to stimulate the growth, although it also produces the growth regulator endogenously. The concentration level of growth

Table 7. Total solid content by growth regulator and oleo-chemicals treatments after three months application

Treatments	Total solid content (g/p/s)
Growth regulators concentration	
N ₀ (control)	39.67
N ₁ (50 ppm NAA ⁺ +50 ppm kinetin)	39.02
N ₂ (100 ppm NAA ⁺ +50 ppm kinetin)	39.20
N ₃ (150 ppm NAA ⁺ +50 ppm kinetin)	38.85
Oleo-chemicals type	
O ₀ (control)	39.24
O ₁ (palmitic acid)	39.07
O ₂ (oleic acid)	38.99
O ₃ (stearic acid)	39.43
Factors interaction	
N ₀ O ₀	40.66
N ₁ O ₁	39.00
N ₁ O ₂	38.29
N ₁ O ₃	39.65
N ₂ O ₁	38.67
N ₂ O ₂	39.59
N ₂ O ₃	40.03
N ₃ O ₁	39.33
N ₃ O ₂	39.72
N ₃ O ₃	37.67

Note: The values that are not lettered are not significantly different.

regulators that is needed by each plant organ is different (Bienaime *et al.*, 2015). Increased concentrations of NAA⁺ and kinetin from 0 ppm to 150 ppm + 50 ppm were noticeably improved rubber bark recovery but this response is linear and the optimum concentration is not yet obtained. Buranov and Elmuradov (2010) reported of thickness increment of rubber bark during recovery using materials containing mercury naphthenate 0.1% and naphthyl acetic acid (NAA⁺) 0.05% to accelerate rubber bark recovery and production increased of 10% after two years of treatment.

NAA⁺ significantly affects the number of latex vessels but showed no significant effect on bark thickness, sucrose level, inorganic phosphate, thiol levels, latex production and total solid content (TSC). The best treatment was obtained at (N₂). It is proposed that the growth regulators which were applied directly onto the wound were absorbed by rubber's stem, which then directly affect the vascular tissue formation. In addition, cambium cell division is affected by auxin (in this case

NAA) and also by cytokinin (kinetin) that promoting and improving cell division in plant tissues and regulate plant growth and development (Ogunyale *et al.*, 2014).

The results showed an increase in bark thickness although no significant effect observed in the number of latex vessels. This is due to application of growth regulator spurred rapid cell division as mentioned by Koryati and Elfiana (2019), which reported that application of auxin and gibberelic acid increased the girth, bark thickness and latex vessel diameter. Furthermore, Ogunyale *et al.* (2014) confirmed that internal factors are important in influencing the activity of the cambium or bark recovery, among other photosynthesis, assimilate transport, genetic factors and growth regulators. External factors such as sufficient water availability do help rapid cells division or differentiation due to increasing cambium activity. However, sufficient water supply is not strong enough to maintain high cambium activity; instead, the effects are accelerated and prolonged (Ramage *et al.*, 2017). Cambium activity spurred by growth regulators application in the study is not significantly affecting the number of latex vessels. This might be due to limited time of the study (three months) so, that even though the number of latex vessels (laticifier) increases along with increasing of skin thickness, no significant results can be observed on such a short time period. The results showed that the NAA concentrations has significantly effect on the number of latex vessels but have no significantly effect on the level of bark thickness, sucrose, thiol, inorganic phosphate, latex production and TSC.

Effect of oleo-chemicals type on bark recovery of clones PB 260. Application of different oleo-chemicals has significant effect on the number of latex vessels, bark thickness and latex production. However, no significant effect observed on the level of sucrose, thiol, inorganic phosphate and TSC were observed. In this study, several types of oleo-chemicals were tested namely palmitic acid, oleic acid and stearic acid on rubber's bark recovery. Number of latex vessels showed the best response to application of stearic acid, followed by oleic, palmitic and no oleo-chemicals as control. The number of latex vessels shows linear response to application of oleo-chemicals, which are able to restore damaged tissue due to the presence of the disease. Using oleo-chemicals are able to promote the growth of recovered skin tissue on rubber's stem by 40% (Robiartini and Budiman, 2002).

Application of different oleo-chemicals types significantly affects bark thickness. The increase of recovered bark at each treatment was observed and calculated since the first application. Application with oleo-chemicals on rubber clones show acceleration of recovered bark thickening when compared with no oleo-chemicals. Such acceleration is influenced by material contained in oleo-chemicals and some studies confirm that oleo-chemical have good effect on the rate of bark recovery, increasing the number of latex vessels in Hevea rubber, such as cytokines, NAA or 2.4 D (Liu *et al.*, 2016).

Applications of some oleo-chemicals were significantly affecting latex production. The highest latex production was obtained in palmitic acid treatment after three months of application. Latex production signifies assimilates translocation and latex synthesis in latex vessels take place normally which in turn will increase the production of latex (Purwaningrum *et al.*, 2019). The benefits of oleo-chemicals to accelerated growth of recovered bark and have dry tapping girth reported by Robiartini and Budiman (2002). Such benefits are improvement in bark recovered after smearing once a month and once two months and once three months and also once four months. The higher the frequency of oleo-chemicals application, the thicker bark recovered generated after scraping, although the frequency of oleo-chemicals applications were varied and differ.

Stearic acid was significantly increasing the bark thickness, the number of latex vessels and latex production. Stearic acid may lead to increased plant growth especially the increase in bark thickness on the stem and also the latex vessels (laticifier) where the stimulation mechanism of stearic acid enzymes system thought be involved in photosynthetic reaction in the calvin cycle. The results of photosynthesis, namely carbohydrate and sucrose are used to enlarge the vegetative parts of the plant, especially stem cells and latex precursor. So, the latex production increased. Kankariya *et al.* (2019) confirm the use of 50 ppm of stearic acid can increase the growth of bean, marigolds, pumpkins and beets by 30-60%.

Interaction effect of growth regulators and oleo-chemicals on bark recovery of rubber clones PB 260.

Interaction of growth regulator concentration and various oleo-chemicals types has no significant effect on the recovery of rubber bark. This indicates that appropriate interaction between NAA⁺ concentration and oleo-chemicals type for rubber's bark recovery is not yet

found. NAA⁺ application affects the activity of cell division in the growing site, while oleo-chemicals act for leveling, solvent, penetrant and antioxidants. Such interaction has no significant effect allegedly due to the time of smearing is not the same and the activities of the two materials are in different places. NAA⁺ acts directly on the stem and leaves, while oleo-chemical is reported to be able to maintain the efficacy of fungicides from external influences, such as climate effects due to antioxidant properties (Tuntipaleepun and Prasertsit, 2011).

Conclusion

Application of growth regulators at tested concentrations significantly affect the number of latex vessels but has no significant effect on skin thickness, sucros and thiol levels, inorganic phosphate, latex production and TSC. oleo-chemical types significantly affect the number of latex vessels, skin thickness and latex production but have no significant effect on the levels of sucrose, Thiol, Inorganic phosphate and TSC. Interaction between NAA⁺ concentrations and oleo-chemicals types have no significant effect on the recovery of rubber's skin.

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