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The Role of Peg Based Stimulants Application on the Production and Physiology Character of Clone Pb 260

Murni Sari Rahayu[a*], Nurhayati[b], Radite Tistama[c], Yenni Asbur[d]

[a,b,d]*Department of Agrotechnology, Faculty of Agriculture, University Islam Sumatera Utara, Medan, Indonesia*

[c]*Rubber Research Institute, Sungai Putih, Deli Serdang, North Sumatra, Indonesia*

[a]*Email: murni.rahayu@fp.uisu.ac.id*

[b]*Email: nurhayatijb@yahoo.co.id*

[c]*Email: raditetistama@yahoo.co.id*

[d]*Email: yenni.asbur@fp.uisu.ac.id*

Abstract

The research's objective is to study the effect of PEG bases alternative stimulant to increase production and physiological character of Clone PB 260. This research was conducted at Sungai Putih Farm, Rubber Research Center, Deli Serdang Regency, Province of North Sumatera. The altitude is 25 m above sea level with Ultisol soil type. This research was to study the difference of production parameters g/p/s, inorganic phosphate, sucrose content, and Thiol. The results showed that the stimulants was significantly affect and increased production, but show no significant effect to increase the concentration of sucrose and Thiol. PEG application can also encourage increased production and inorganic phosphate although has no significant effect but increasing the levels of sucrose and Thiol. Combination of stimulants and PEG is increase on the production and inorganic phosphates parameters.

Keywords: Clone PB 260; Stimulant; PEG.

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* Corresponding author.

1. Introduction

Indonesia is a country with the largest rubber plantation in the world, namely 3.4 million hectares but its production is still low compared to the competitors, only around 1050 kg/hectare [1]. Official Data from The Ministry of Trade show that Indonesia's foreign exchange derived from natural rubber in 2014 is amounted to 4.7 billion US dollars [2]. Currently, the Indonesian government has set a target to develop natural rubber production that is 3.4 million tons/year by 2025 [3]. Increased production can be achieved through superior clones that are by optimizing the taping system. One effort to optimize the taping system is stimulants application to increase the productivity of rubber tree [4]. Stimulant has been widely known in rubber tapping to increase latex production by prolonging the latex flow. According Krisnakumar and his colleagues (2011), stimulant can increase latex production by prolonging the flow due to blockage on latex vessels [5]. Presently, stimulant that is widely used both in government and private rubber estate is etephon (2-chlorocthyphosponic acid). Such stimulant is able to prevent blockage which in turn increased latex flow [6.5]. However, continuous use of this stimulant has negative effect on trees that is more susceptible to dry tapping grooves (KAS) [4]. Therefore it is necessary to find and develop alternative stimulants that can increase production as well as safer to the physiological conditions of the rubber tree. PEG (Polyethylene glycol) is a potential substance to be used as stimulant. Such compound is able to decrease osmotic potential through the activity of ethylene oxide subunits that increase water molecules with hydrogen bonds [7]. Rahayu and his colleagues (2016) reported that 3% PEG application as stimulant is able to increase the latex production [8]. This research's objective is to study the effect of PEG based stimulant to increase the production and physiological characteristics of Rubber's Clone PB 260.

2. Research methods

2.1. Time and Place

This research was conducted at Sungai Putih Farm, Rubber Research Center, Deli Serdang Regency, Province of North Sumatera. The altitude is 25 m above sea level with Ultisol soil type. Experiment of stimulant and PEG application on 11 years old of clone PB 260 was conducted at field and physiology laboratory, White River Research Institute.

2.2. Materials and Tools

Material: the materials used are grouped into 2 ie material for field activity and for laboratory observation. Materials for the field activities are 11 years old of Clone PB 260, PEG, palmitic acid, NAA, Kinetin, and Etephon. Materials for physiological analysis in laboratory are concentrated sulfuric acid (H_2SO_4), Aquadest, Trish reaction tube, Anthron, Ethanol, dithiobis nitro benzoic acid (DTNB). Tools: Tapping knives, container bowls, gutters, buckets, brushes, stationery, red and yellow paint, digital scales and other support tools. Physiological tools are glass beaker, test tube, oven, Becleman DO 650 spectrometer, mortar, strier, and analytical scales. This research was arranged on Factorial Randomized Block Design with 2 treatment factors and 3 replications. Factor treatment ie Etephon stimulant concentration (S) has 4 treatment levels: S_0 = No

Stimulant, S_1 = N_2O_1 formulation, S_2 = Etephon 1.5% + N_2O_1 formulation, S_3 = 2.5% Etephon + N_2O_1 formulation and PEG concentration (P) has 2 levels: P_0 = without PEG, P_1 = PEG 3%.

3. Results

3.1. Latex Production (g/p/s)

The results showed that Stimulant (S) application has very significant effect on latex production. PEG (P) application has very significant effect on latex production. The combination of Stimulant and PEG is significantly affecting latex production (Table 1).

Table 1: Mean of Latex Production (g/p/s) on Stimulant and PEG Treatment

Treatment	Latex Production (g/p/s)
Stimulant	
S_0 (control)	29.51 cC
S_1 (Formulation N_2O_1)	40.36 aA
S_2 (etephon 1,5%+ Formulation N_2O_1)	30.69 cC
S_3 (etephon 2,5%+ Formulation N_2O_1)	32.81 bB
PEG application	
P_0 (control)	26.68 bB
P_1 (PEG 3%)	40.01 aA
Interaction	
S_0P_0	23.62 gG
S_0P_1	35.39 cC
S_1P_0	27.52 fF
S_1P_1	53.20 aA
S_2P_0	29.10 eE
S_2P_1	32.29 dD
S_3P_0	26.47 fF
S_3P_1	39.14 bB

Note : The numbers followed by the same letter on the same row or column are not significantly different at the 5% and 1% Duncan test. Formula N_20_1 (NAA 100 ppm + Kinetin 50 ppm + Palmitic Acid 2%)

Table 1 shows the highest latex production was found in S_1 treatment (formulation N_2O_1) namely 40.36 g/p/s, followed by S_3 (Etephon 2,5% + formulation N_2O_1), S_2 (Etephon 1,5% + formulation N_2O_1) and S_0 (Control). PEG application (P1) has higher latex production and effect on increasing latex production than no PEG (P_0) PEG application had very significant effect on increasing latex production, that is an increase by 49.96%. Combination of stimulant and PEG had very significant effect to support latex production. The highest latex production was found in S_1P_1 (formulations N_2O_1 and PEG), while the lowest was found in S_0P_0 (control). Increased of latex production with N_2O_1 and PEG formulation was 125.23%.

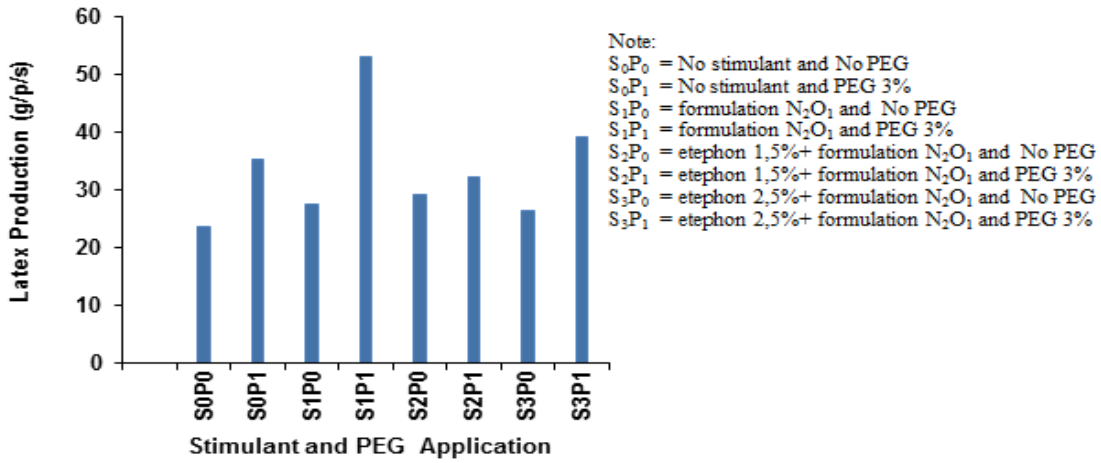

Figure 1: Histogram of Production Mean with Combination of Stimulant and PEG

3. 2. Inorganic Phosphate (IPs)

The research foud that stimulant treatment (S) has no significant effect on Inorganic Phosphate (IPs). PEG Treatment (P) has significant effect on Inorganic Phosphate. Combination of stimulant and PEG had very significant effect on Inorganic Phosphate (Table 2).

Table 2: Mean of Inorganic Phosphate (IPs) on Stimulant and PEG Treatment

Treatment	**Inorganic Phosphate (IPs)**
Stimulant	
S_0 (control)	3.38
S_1 (Formulation N_2O_1)	3.36
S_2 (etephon 1,5%+ Formulation N_2O_1)	3.26
S_3 (etephon 2,5%+ Formulation N_2O_1)	3.40
PEG application	
P_0 (control)	3.19 b
P_1 (PEG 3%)	3.51 a
Interaction	
S_0P_0	3.53 bB
S_0P_1	3.23 dD
S_1P_0	2.94 eE
S_1P_1	3.79 aA
S_2P_0	3.22 dD
S_2P_1	3.29 cC
S_3P_0	3.06 eE
S_3P_1	3.74 aA

Note: The numbers followed by the same letter on the same row or column are not significantly different at the 5% and 1% Duncan test. Formula N_20_1 (NAA 100 ppm + Kinetin 50 ppm + Palmitic Acid 2%)

Table 2 shows that PEG treatment had significant effect on the levels of Inorganic Phosphate. The highest inorganic phosphate was found in PEG (P1), namely 3.51 mM, while the lowest in P0 (control), namely 3.19 mM. Increased levels of Inorganic Phosphate with PEG application is 10.03%.

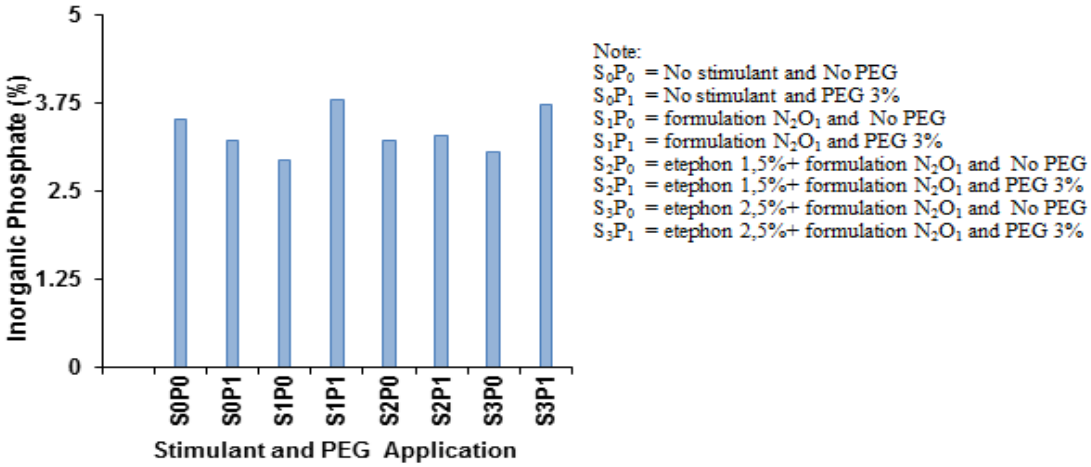

Figure 2: Histogram of Inorganic Phosphate Mean on Combination of Stimulant and PEG

3. 3. Sucrose

The result of statistic analysis showed that stimulant treatment (S) and PEG (P) did not have significant effect on sucrose content. Combination of stimulant and PEG application had no significant effect on the sucrose content (Table 3).

Table 3: Mean of Sucrose (mM) on Stimulant and PEG Treatment

Treatment	Sucrose (mM)
Stimulant	
S_0 (control)	3.29
S_1 (Formulation N_2O_1)	4.23
S_2 (etephon 1,5%+ Formulation N_2O_1)	3.94
S_3 (etephon 2,5%+ Formulation N_2O_1)	3.65
PEG application	
P_0 (control)	3.57
P_1 (PEG 3%)	3.99
Interaction	
S_0P_0	2.79
S_0P_1	3.79
S_1P_0	3.76
S_1P_1	4.70
S_2P_0	3.99
S_2P_1	3.90
S_3P_0	3.74
S_3P_1	3.55

Note: The numbers followed by the same letter on the same row or column are not significantly different at the 5% and 1% Duncan test. Formula N_20_1 (NAA 100 ppm + Kinetin 50 ppm + Palmitic Acid 2%)

Table 3 shows that stimulant and PEG has no significant effect on the sucrose content. There is an increase in the sucrose content with PEG treatment. Combination of stimulant and PEG had no significant effect on sucrose level, but there was a tendency of increase of sucrose level.

3.4. Thiol

The results of statistical analysis showed that Stimulant treatment (S) and PEG (P) did not significantly affect the Thiol level. The combination of Stimulant and PEG application also had no significant effect on Thiol level (Table 4). Table 4 showed that stimulant and PEG treatment had no significant effect on Thiol level. There is an increase of Thiol level with PEG treatment. The combination of stimulants and PEG had no significant effect on Thiol level; however there was an increase in Thiol leves.

Table 4: Mean of Thiol (mM) on Stimulant and PEG Treatment

Treatment	Thiol (mM)
Stimulant	
S_0 (control)	0.82
S_1 (Formulation N_2O_1)	0.67
S_2 (etephon 1,5%+ Formulation N_2O_1)	0.61
S_3 (etephon 2,5%+ Formulation N_2O_1)	0.66
PEG application	
P_0 (control)	0.70
P_1 (PEG 3%)	0.69
Interaction	
S_0P_0	0.82
S_0P_1	0.82
S_1P_0	0.75
S_1P_1	0.59
S_2P_0	0.56
S_2P_1	0.67
S_3P_0	0.66
S_3P_1	0.66

Note : The numbers followed by the same letter on the same row or column are not significantly different at the 5% and 1% Duncan test. Formula N_20_1 (NAA 100 ppm + Kinetin 50 ppm + Palmitic Acid 2%)

4. Discussion

The most important response of rubber tree to the treatment is the production. Interaction of stimulant S1 (Formulation N_2O) and PEG is significantly increase the production.The increase is due to the presence of

palmitic acid contained in the N_2O_1 formulation. Palmitic acid (fatty acid) will be converted to Acetyl Coenzyme A. Such acetyl CoA will form Tricarboxilie Acid (TCA) and produce energy. Energy generated from fatty acids breakdown is greater than carbohydrates breakdown. The energy will be partially utilized for vegetative growth (stem cell enlargement) with the addition of bark thickness and the number of latex vessels. Some of the Acetyl CoA will produce terpenoid compounds such as politerpen (latex). Therefore, application of palmitic acid stimulants basis is able to increase the latex production [8,9]. Besides increasing osmotic pressure, PEG application also induces the activity of PEP carboxylase enzyme [10]. Increased photosynthesis activity causes the level of sucrose also increased. Sucrose is a precursor of latex, therefore PEG-based stimulants will lead to increased of latex production [9]. Inorganic phosphate is an indicator of metabolic activity that is describing plant ability to convert raw material (sucrose) into rubber particles [11]. Inorganic Phosphate is a reflection of active metabolism because phosphate serves as a phosphorylated compound and an energy-forming compound. Inorganic phosphate levels show the intensity of metabolic activity in latex vessels [6]. The results showed that the interaction of stimulant and PEG had significant effect on the content of Inorganic Phosphate (IPs). Table 2 shows the highest inorganic phosphate content found at S_1P_1 (formulation of N_2O_1 and PEG), namely 3.79 mM. This is because formulation N_2O_1 contain palmitic acid that able to produce ATP (energy) derived from Tricarboxylie Acid (Respiration). In this case, ATP is a reflection of active metabolism that play a role in latex formation. PEG application increase Inorganic Phosphate, means that there will be an increase in photosynthetic activity due to increase in carboxylated PEP [10]. As the result, carbohydrate is also increases. Carbohydrates have important roles for plant life to grow and perform other physiological activities normally. In other words, carbohydrates are the main source of energy to life [12]. Yulinda (2010) confirm that triterpenoid secondary metabolite content in invitro culture of *Centella asiatica* increases with the addition of 1 of 2% PEG [13]. The higher levels of Inorganic Phosphate mean the more active metabolism of plants. The levels of Inorganic Phosphate in both treatments is not exceeded the threshold, that is 25 mM. If it level exceeds the threshold, indicates the plant response to stress or disease [11]. Sucrose is raw material of cis-polyisoprene synthesis which required by latex cells for regeneration [14]. Biosynthesis/latex regeneration takes place in latificiferous cells, use sucrose derived from leaves as raw material and as the center of photosynthesis [15]. There is no significant effect of treatment given, probably because the research was conducted at autumn season when leaves as place for photosynthesis begin to fall. Photosynthesis yield will be transferred to other organs in the form of sucrose. Tolbert (1979), Yusoff and Chow (2003) states that source-sink mechanism is an assimilate partition generally derived from the canopy to the bark that produces latex when tapped [16,17]. In this research, the content of latex sucrose is very low due to leaf decay so that although the level of sucrose starts to increase every month but there is a tendency of more assimilate use for leaf formation. However, Table 1 show that there is an increase in production due to existing sucrose molecules going through a series of enzymatic reactions will form acetic acid molecules or acetyl CoA derived from palmitic acid from the stimulant. Acetyl CoA is the main precursor of cis-polyisoprene formation. Thiol levels is important indication related to latex physiological susceptibility especially in the incidence of dry tapping grooves (KAS) [11]. Thiol function is to activate enzymes that play in environmental stress conditions. According to Thiol levels, the use of bark recovery based stimulants and PEG did not have negative affect on the observed parameters. Thiol is an antioxidant-related parameter that reflects the ability of plants (active oxygen species) to prevent cell damage by free radicals [18]. Thiol availability in latex is important since it functions as an activator of various enzymes

and associated with lutoid membrane stability to prolong the length of latex flow [6]. In turn, if latex regeneration works well it will support high production at the time of tapping. This research show that although stimulant, PEG and the combination did not have significant effect but overall still showed a safe level for the plants. This is as indicated by field observations that dry tapping grooves (KAS) have not encountered by the Rubber Tree.

5. Conclusions

1. Applied stimulants has significant effect and encourage increased production, although has no significant effect but increase in the level of sucrose and Thiol.
2. PEG application can also encourage increased production and inorganic phosphate although has no significant effect but increase the levels of sucrose and Thiol levels.
3. Combination of stimulant and PEG application found increase production and inorganic phosphate.

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