

## Effect of Deficiency-adjusted Macronutrients to Cure Brown Bast Syndrome in Rubber Tree (*Hevea brasiliensis*)

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### ABSTRACT

The brown bast (BB) syndrome causes a 15-20% loss of annual rubber (*Hevea brasiliensis*) production in Malaysia, and no sustainable remedy has been developed yet. Initial investigation showed a macronutrients deficiency in the bark of affected plants compared to non-affected plants. Therefore, this study was undertaken to know the effect of spraying the deficient macronutrients in curing BB syndrome and increasing latex productivity. The treatments were selected by adjusting deficient nutrients, especially in N, K, and S, compared to healthy plants. The treatments consisted of 13 combinations of nutrient concentrations and a control (no application on healthy rubber trees). One liter of individual treatment per tree was applied on the trunk of BB-affected trees once a week for one month. Data on latex production was collected one week after the last application and continued for up to three months. The results showed that the treatment T<sub>9</sub> (0.5% N,

0.8 mg/L K, and 0.2% S), in addition to other prescribed nutrients, made a 100% cure of BB syndrome after three weeks of application, and the nutrient contents of the bark of the affected plants appeared to be similar to the healthy plants. Applying these additional nutrients enhanced the latex production at the rate of 51.85 g/tapping/tree after two months of treatment application under the agro-ecosystem of FELCRA, Simpang Renggam, Johor, Malaysia. It is

### ARTICLE INFO

#### Article history:

Received: 05 May 2023

Accepted: 27 June 2023

Published: 02 November 2023

DOI: <https://doi.org/10.47836/pjtas.46.4.15>

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a new finding to 100% cure BB syndrome and to increase the latex productivity in sustaining the rubber industry in Malaysia.

*Keywords:* Brown bast syndrome, *Hevea brasiliensis*, latex productivity, macronutrient, tapping frequency d3, tapping panel dryness, tapping system S/2

## INTRODUCTION

Rubber (*Hevea brasiliensis*) is one of the significant economic plantations besides oil palm in Malaysia. The country plays an essential role in producing and exporting rubber worldwide. As per the first Economic Transformation Programs (ETPs) of Malaysia, the average national rubber productivity needs to be increased to 2,000 kg/ha by 2020 (Said & Ghani, 2012) since rubber is an important commodity under the National Key Economic Area (NKEA) of the country. However, since 2019, latex productivity has been reduced, with an average of 1,440 and 1,420 kg/ha in 2017 and 2018, respectively, due mainly to brown bast (BB) syndrome and other agronomic management problems. Rubber production has declined from 799.1 thousand tonnes in 2019 to 719 thousand tonnes in 2020. Meanwhile, rubber consumption in Malaysia increased from 1,075.7 thousand tonnes in 2019 to 1,170.1 thousand tonnes in 2020 (Malaysian Rubber Council [MRC], n.d.). Due to this decline in rubber production and increased demand, the country has relied on other countries to meet its rubber needs. According to MRC (n.d.), rubber imports in Malaysia increased from 1,677.4 thousand tonnes in 2019 to 1,702.1 thousand

tonnes in 2020. Therefore, an increase in rubber productivity in Malaysia is of prime importance.

Brown bast or tapping panel dryness (TPD) is a syndrome related to partial or entire dryness of a rubber tapping panel. The syndrome is regarded as a physiological disorder that causes severe damage to the rubber trees if not treated immediately. TPD is characterized by the drying up the latex-producing tissues beneath the tree's bark, resulting in reduced or halted latex flow. This condition is a major concern for rubber plantations, significantly impacting latex production and overall yield. This disorder causes an imbalance between the latex regeneration and tapping process. The bark becomes brown, thick, and finally cracks in severe cases. Brown bast has been reported to cause a loss of 15-20% in rubber production in Malaysia (Nandris et al., 2004). Qi et al. (2014) reported that around 14.75% of rubber trees are damaged by tapping panel dryness. Another report says that about 12–50% of productive trees are affected by TPD in almost every rubber-growing region (Venkatachalam et al., 2007). TPD can persist for extended periods and cause significant economic losses for rubber plantations.

The exact cause of BB or TPD is not yet fully understood, and it is believed to result from a combination of physiological, anatomical, and environmental factors. Some possible contributing factors include hormonal imbalances, nutrient deficiencies, excessive or improper tapping practices, genetic factors, and environmental stressors

such as drought, extreme temperatures, or waterlogging (Venkatachalam et al., 2007). The researchers have been trying to develop methods to treat BB syndrome for a long time. As part of the efforts, the syndrome portion was isolated from healthy high bark to avoid infection in healthy parts of rubber trees (Keralafarmer, 2019), but that was unsuccessful. The injection of chemical stimulants such as ethephon to increase latex productivity tended to cause a high incidence of this physiological syndrome (Nazri, 2020). The introduction of a newly formulated ethephon and water-based stimulant (RRIM HYDROBEST™) by the Malaysian Rubber Board (MRB) (2009) indicated that this stimulant could improve land productivity and lower the incidence of TPD (Budiasih et al., 2020; Nazri, 2020; Sainoi & Sdoodee, 2012).

However, treatment with the stimulants depends on the cell biochemistry of the plants (Nik Hashyati et al., 2022) and, for longer use, affects the latex physiology (Lacote et al., 2010). Nik Hashyati et al. (2022) found that the bark dryness of rubber trees could be cured by treating them with a specific fertilizer formulation of macro and micronutrients in liquid form and spraying on the tree. In light of the findings mentioned above, it was speculated that there might be differences in nutrient contents in the bark, leaf, and soil ecosystems of healthy and BB-syndromed rubber plants, which must be solved. Therefore, this study was undertaken firstly to know if there is any difference in macronutrients in the ecosystems of BB-affected and non-affected rubber trees;

secondly, if the difference exists whether adjustment of deficient nutrients can cure BB-syndrome, and thirdly, to know if the application of adjusted macronutrients can increase the yield of latex.

## **MATERIALS AND METHODS**

### **Sampling Location and Climatic Conditions**

The study was conducted at FELCRA, Simpang Renggam, Johor (1°44'55.1"N, 103°20'02.6"E) with the clone RRIM 2002 from November 2019 to February 2020. The area's three hundred and ninety trees, representing 56.52%, were infected by BB syndrome. Of these, 39 trees (10%) were randomly selected and used as study samples, and healthy plants were selected from the same area. The average temperature during the experiment was similar and varied from 33°C (November 2019) to 33.2°C (February 2020). The rainfall was high at the beginning and then reduced gradually. The flow was below: November 2019 = 67 mm, December 2019 = 84 mm, January 2020 = 47 mm, and February 2020 = 25 mm, respectively.

### **Pretreatment Nutrient Assessment and Treatment Determination**

First, some affected and healthy plants' soil, leaf, and bark nutrient contents were analyzed. Significant differences were identified in N, K, and S contents, especially in the barks between the two groups of plants. Therefore, in the new formulation, the nutrient deficiency in the affected plants was adjusted based on healthy plants

(Table 1). The N, K, and S nutrient rates used in the formulation were based on the difference observed between affected and unaffected rubber trees. For example, BB-affected plants' N contents differ by 0.2 to 0.8%. Putting the lowest and highest values in the Box-Behnken design, the predicted formulation rates were estimated for use as treatments (Kumari et al., 2021). The different nutrient concentrations used in the study are shown in Table 1. Treatment T<sub>1</sub> represents the healthy rubber tree as the control, i.e., without treatment application. Treatments T<sub>2</sub> to T<sub>14</sub> are estimated treatments, respectively. As per treatment specification, the nutrients of N, K, and S, which were obtained using ammonium nitrate, triple super phosphate, and ammonium sulfate that were purchased from RM Phosphate & Chemicals Pvt. Ltd. (Malaysia), were

mixed, stirred, and diluted in 1.0 L of distilled water.

The treatments were applied directly to the tree base to 6 feet upright of the trunk using a hand sprayer once a week for one month. Thirty-nine soil, bark, and leaf samples were collected from affected and non-affected rubber trees. The leaves and the barks were dried in the oven at 50°C for three days. The soil samples were collected at a depth of 30 cm from the area 2 m away from the base of affected and non-affected rubber trees using an auger. The soil samples were then separated into two different depths: 0-15 and 15-30 cm, respectively. The samples were air-dried, ground, and sieved through a 2 mm sieve before analysis for their macronutrients, such as calcium (Ca), nitrogen (N), phosphorus (P), potassium (K), magnesium (Mg), and

Table 1  
*Concentration of additional nutrients used in the treatment formulation*

Symbol	Rate		
	N (%)	K (mg/L)	S (%)
T <sub>1</sub> (control)	0	0	0
T <sub>2</sub>	0.2	0.2	0.5
T <sub>3</sub>	0.2	0.5	0.2
T <sub>4</sub>	0.2	0.5	0.8
T <sub>5</sub>	0.2	0.8	0.5
T <sub>6</sub>	0.5	0.2	0.2
T <sub>7</sub>	0.5	0.2	0.8
T <sub>8</sub>	0.5	0.5	0.5
T <sub>9</sub>	0.5	0.8	0.2
T <sub>10</sub>	0.5	0.8	0.8
T <sub>11</sub>	0.8	0.2	0.5
T <sub>12</sub>	0.8	0.5	0.2
T <sub>13</sub>	0.8	0.5	0.8
T <sub>14</sub>	0.8	0.8	0.5

sulfur (S). The samples were also analyzed using appropriate methodologies to know their pH and electrical conductivity (EC). Leaf and bark analyses for the elements N and S were done using LECO CNS TruMac Analyzer (Netherlands) (Kowalenko et al., 2001). The macronutrients P and K were determined by the dry ashing method (Varley, 1966) using an auto-analyzer (LACHAT Instrument, QuikChem FIA+ 8000 series, Malaysia), while Ca and Mg were analyzed using atomic absorption spectroscopy (PerkinElmer PinAAcle 900T, USA). Soil pH was determined using a calibrated pH meter (Forster, 1995; Paul et al., 2017). EC analysis was carried out with a ratio of 1:5 of soil to water (Foster, 2015) using a conductivity meter.

#### Data Collection and Statistical Analysis

Data on latex yield were collected one week after the last treatment and continued for 3 months with the tapping system S/2 (half-spiral cut downward) and tapping frequency d3 (once every 3 days) to prevent stress on the rubber trees. The cup lump was weighed and recorded before and after the tapping collection.

The percentage of brown bast cure was measured by using a measuring tape and calculated by the formula below:

$$\% \text{ Brown bast cure} = \frac{\text{Total length of tapping cut} - \text{Length of cured tapping cut}}{\text{Total length of tapping cut}} \times 100\%$$

(Rhoades, 1993)

The experiment was conducted in a randomized complete block design (RCBD) with 3 replications. In the first stage of statistical analysis, a comparison between healthy rubber trees and BB-affected rubber trees was done using a *t*-test. In the second stage of the study, the variance (ANOVA) analysis was done to identify the significant effectiveness of applying the new formulation using the concerned elements shown in the first stage. All the data were analyzed using the statistical software SAS (version 9.4). The significant differences were determined using Tukey's honest significant difference (HSD) test at a 5% level of probability ( $p \leq 0.05$ ).

## RESULTS AND DISCUSSION

### Soil pH, Soil EC, and Nutrient Contents in Soil, Leaf, and Barks

The results of nutrient analyses indicated no significant difference in soil nutrients between affected and unaffected rubber trees, as shown in Table 2. Soil pH ranged between 4.5 and 5.5 (Table 2), which is considered optimum acidic for rubber plantation (Daud, 2013; Priyadarshan, 2003), and the soil EC of both the affected and unaffected rubber trees was 1–2 dS/m (Table 2), which was in a non-saline state ("Wintering season," 2021).

Soil nutrient contents similarly influenced the nutrient contents of the leaf, as shown in Table 3. The insignificant difference in nutrient content in both soil and leaves was assumed to be the plant's adequate nutrient supply. However, the nutrient contents of the bark of BB-affected

rubber trees were significantly varied, especially in N and S contents, while the other nutrients, such as P, K, Ca, and Mg, were similar (Table 4). Bark and leaves usually correlate positively with their nutrient contents (Jones et al., 2019), but the authors found a difference between leaves and bark. Based on this difference, when a new formulation with additional fertilizers

was made, it caused positive responses to the healthiness of the bark of the BB-affected rubber trees. After treatment with a new formulation of fertilizers, the nutrient contents of the bark tissues of BB-affected trees were increased, and they became like non-affected healthy trees (Table 5). The bark of plants can absorb nutrients when the fertilizers are applied directly to the trees.

Table 2  
*Differences in pH, EC, and macronutrients in soils of healthy and BB-affected rubber trees before treatment application at two soil depths*

Rubber plant	Depth (cm)	pH	EC (dS/m)	N (%)	Available P (mg/kg)	K (mg/kg)	Ca (mg/kg)	Mg (mg/kg)	S (%)
Affected tree	0-15	5.232	2.090	0.073	12.677	42.030	210.200	22.633	0.065
Healthy tree		5.140	1.540	0.055	15.365	27.982	88.798	16.483	0.068
<i>p</i> -value		0.460 <sup>ns</sup>	0.187 <sup>ns</sup>	0.053 <sup>ns</sup>	0.607 <sup>ns</sup>	0.114 <sup>ns</sup>	0.063 <sup>ns</sup>	0.130 <sup>ns</sup>	0.203 <sup>ns</sup>
Affected tree	15-30	5.103	1.566	0.067	10.462	33.915	129.100	15.350	0.065
Healthy tree		4.947	1.687	0.070	10.208	30.032	68.513	13.033	0.066
<i>p</i> -value		0.381 <sup>ns</sup>	0.562 <sup>ns</sup>	0.827 <sup>ns</sup>	0.906 <sup>ns</sup>	0.563 <sup>ns</sup>	0.154 <sup>ns</sup>	0.423 <sup>ns</sup>	0.684 <sup>ns</sup>

Note. EC = Electrical conductivity; BB = Brown bast; ns = Not significant at  $p > 0.05$

### Effect of New Formulation on Brown Bast Cure

The new formulation of fertilizers, especially of N, K, and S, caused a positive response to cure BB syndrome in rubber trees, leading to increased latex production. Figure 1 shows that before treatment application, the latex flow in T<sub>1</sub> (healthy tree) was good (100% latex flow), but the BB-affected plants represented by T<sub>2</sub> to T<sub>14</sub> had no latex flow on the tapping cut. Figures 2, 3, 4, and

5 show the effect of treatments with new formulations on the percent cure of brown bast, respectively. For instance, Figure 2 indicates that, on average, 11.33% BB cure occurred after the first week of treatment application. Although the percent BB cure increased a bit, it was still significantly lower than the healthy plant (T<sub>1</sub>), and no significant differences were observed between treatments, T<sub>2</sub> to T<sub>14</sub>. However, significant increases (33%) were found in

the treated trees ( $T_2$  to  $T_{14}$ ) after two weeks of treatment application (Figure 3), and it caused a similar cure of healthy plants except for a few treatments ( $T_2$ ,  $T_5$ ,  $T_{12}$ , and  $T_{14}$ ). More prominent results (69% increase) were observed after three weeks of treatment application (Figure 4).

The results indicated no significant difference between healthy trees and the treated trees ( $T_2$  to  $T_{14}$ ) regarding the percent cure of BB syndrome. Noticeably,  $T_9$  produced a 100% brown bast cure after three weeks of treatment application, as

shown in Figures 4 and 5, and was not significantly different from  $T_1$ . The effects of other treatments produced differential BB cures, but there was no significant difference between all other treatments. This result was in line with a previous similar study using both macro and micronutrients, which was conducted by Nik Hashyati et al. (2022), who observed that brown bast syndrome could be cured by the application of macro- and micronutrients in liquid form and applied on the affected trees.

Table 3  
*Differences in macronutrients between leaf tissue of healthy and BB-affected trees before treatment application*

Rubber plant	N (%)	P (mg/kg)	K (mg/kg)	Ca (mg/kg)	Mg (mg/kg)	S (%)
Affected tree	2.779	302.7	3660.2	1,8621.7	812.7	0.244
Healthy tree	2.724	341.7	3,715.7	1,8282.2	985.8	0.24
<i>p</i> -value	0.765 <sup>ns</sup>	0.737 <sup>ns</sup>	0.686 <sup>ns</sup>	0.908 <sup>ns</sup>	0.449 <sup>ns</sup>	0.775 <sup>ns</sup>

Note. BB = Brown bast; ns = Not significant at  $p > 0.05$

Table 4  
*Differences in macronutrients between bark tissue of healthy and BB-affected trees before treatment application*

Rubber plant	N (%)	P (mg/kg)	K (mg/kg)	Ca (mg/kg)	Mg (mg/kg)	S (%)
Affected tree	0.463	894.8	4,063.7	9,741.3	1,546	0.092
Healthy tree	0.559	958.3	4,206.8	9,263.8	1,496.8	0.105
<i>p</i> -value	0.023*	0.617 <sup>ns</sup>	0.282 <sup>ns</sup>	0.789 <sup>ns</sup>	0.785 <sup>ns</sup>	0.007*

Note. BB = Brown bast; ns = Not significant at  $p > 0.05$ ; \* = Significant at  $p \leq 0.05$



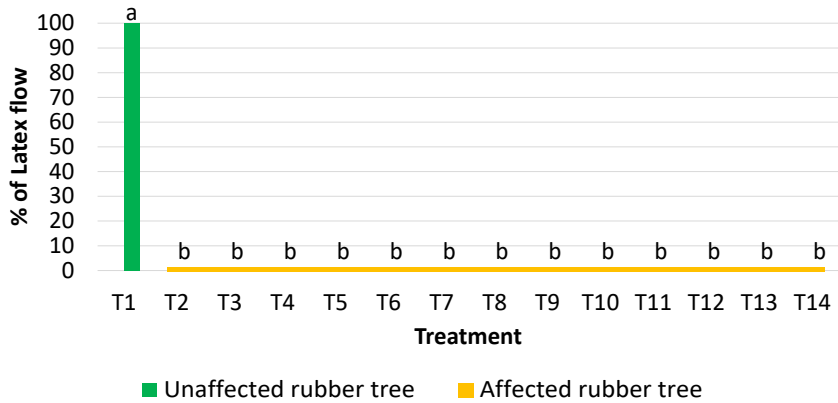


Figure 1. Initial percentage of latex yield of healthy trees and brown bast-affected trees

Note. Bars with the different letters are significantly different at  $p \leq 0.05$  according to Tukey's honest significant difference test [T<sub>1</sub> = Control; T<sub>2</sub> = 0.2% N, 0.2 mg/L K, and 0.5% S; T<sub>3</sub> = 0.2% N, 0.5 mg/L K, and 0.2% S; T<sub>4</sub> = 0.2% N, 0.5 mg/L K, and 0.8% S; T<sub>5</sub> = 0.2% N, 0.8 mg/L K, and 0.5% S; T<sub>6</sub> = 0.5% N, 0.2 mg/L K, and 0.2% S; T<sub>7</sub> = 0.5% N, 0.2 mg/L, and 0.8% S; T<sub>8</sub> = 0.5% N, 0.5 mg/L K, and 0.5% S; T<sub>9</sub> = 0.5% N, 0.8 mg/L, and 0.2% S; T<sub>10</sub> = 0.5% N, 0.8 mg/L K, and 0.8% S; T<sub>11</sub> = 0.8% N, 0.2 mg/L K, and 0.5% S; T<sub>12</sub> = 0.8% N, 0.5 mg/L K, and 0.2% S; T<sub>13</sub> = 0.8% N, 0.5 mg/L K, and 0.8% S; T<sub>14</sub> = 0.8% N, 0.8 mg/L K, and 0.5% S]

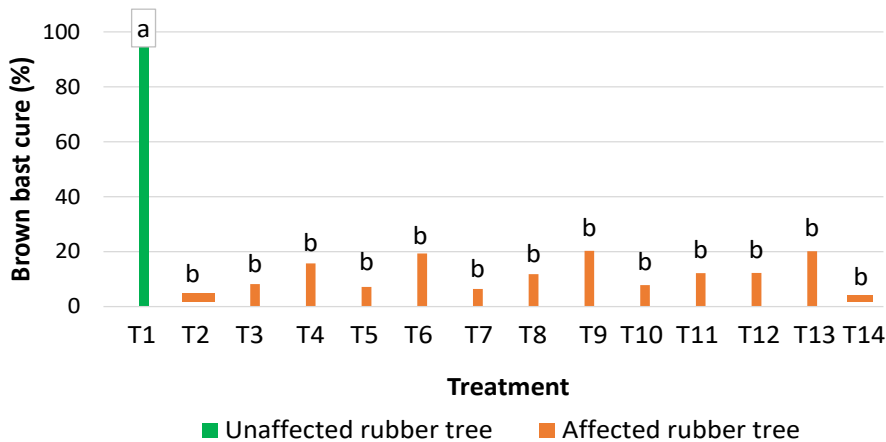


Figure 2. Percentage of brown bast cure after 1 week of treatment application

Note. Bars with the different letters are significantly different at  $p \leq 0.05$  according to Tukey's honest significant difference test [T<sub>1</sub> = Control; T<sub>2</sub> = 0.2% N, 0.2 mg/L K, and 0.5% S; T<sub>3</sub> = 0.2% N, 0.5 mg/L K, and 0.2% S; T<sub>4</sub> = 0.2% N, 0.5 mg/L K, and 0.8% S; T<sub>5</sub> = 0.2% N, 0.8 mg/L K, and 0.5% S; T<sub>6</sub> = 0.5% N, 0.2 mg/L K, and 0.2% S; T<sub>7</sub> = 0.5% N, 0.2 mg/L, and 0.8% S; T<sub>8</sub> = 0.5% N, 0.5 mg/L K, and 0.5% S; T<sub>9</sub> = 0.5% N, 0.8 mg/L, and 0.2% S; T<sub>10</sub> = 0.5% N, 0.8 mg/L K, and 0.8% S; T<sub>11</sub> = 0.8% N, 0.2 mg/L K, and 0.5% S; T<sub>12</sub> = 0.8% N, 0.5 mg/L K, and 0.2% S; T<sub>13</sub> = 0.8% N, 0.5 mg/L K, and 0.8% S; T<sub>14</sub> = 0.8% N, 0.8 mg/L K, and 0.5% S]



Curing of Brown Bast in Rubber Trees

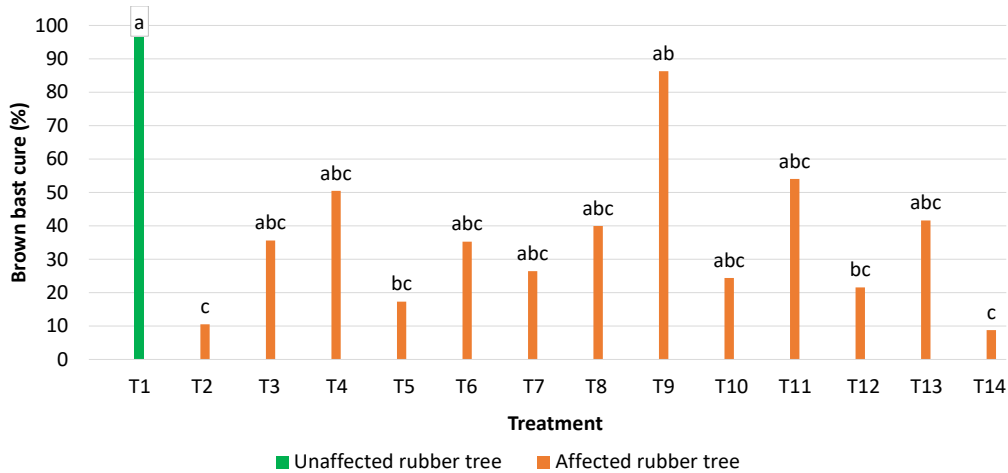


Figure 3. Percentage of brown bast cure after 2 weeks of treatment application

Note. Bars with the different letters are significantly different at  $p \leq 0.05$  according to Tukey's honest significant difference test [T<sub>1</sub> = Control; T<sub>2</sub> = 0.2% N, 0.2 mg/L K, and 0.5% S; T<sub>3</sub> = 0.2% N, 0.5 mg/L K, and 0.2% S; T<sub>4</sub> = 0.2% N, 0.5 mg/L K, and 0.8% S; T<sub>5</sub> = 0.2% N, 0.8 mg/L K, and 0.5% S; T<sub>6</sub> = 0.5% N, 0.2 mg/L K, and 0.2% S; T<sub>7</sub> = 0.5% N, 0.2 mg/L, and 0.8% S; T<sub>8</sub> = 0.5% N, 0.5 mg/L K, and 0.5% S; T<sub>9</sub> = 0.5% N, 0.8 mg/L, and 0.2% S; T<sub>10</sub> = 0.5% N, 0.8 mg/L K, and 0.8% S; T<sub>11</sub> = 0.8% N, 0.2 mg/L K, and 0.5% S; T<sub>12</sub> = 0.8% N, 0.5 mg/L K, and 0.2% S; T<sub>13</sub> = 0.8% N, 0.5 mg/L K, and 0.8% S; T<sub>14</sub> = 0.8% N, 0.8 mg/L K, and 0.5% S]

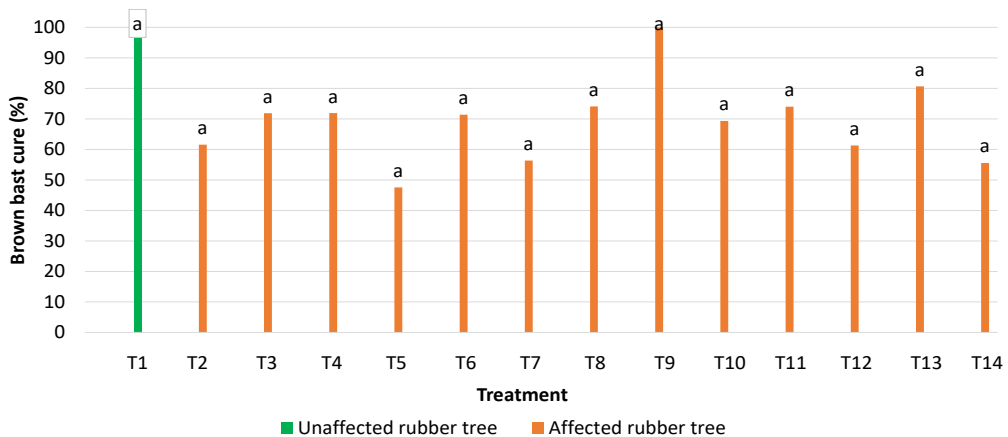


Figure 4. Percentage of brown bast cure after 3 weeks of treatment application

Note. Bars with the different letters are significantly different at  $p \leq 0.05$  according to Tukey's honest significant difference test [T<sub>1</sub> = Control; T<sub>2</sub> = 0.2% N, 0.2 mg/L K, and 0.5% S; T<sub>3</sub> = 0.2% N, 0.5 mg/L K, and 0.2% S; T<sub>4</sub> = 0.2% N, 0.5 mg/L K, and 0.8% S; T<sub>5</sub> = 0.2% N, 0.8 mg/L K, and 0.5% S; T<sub>6</sub> = 0.5% N, 0.2 mg/L K, and 0.2% S; T<sub>7</sub> = 0.5% N, 0.2 mg/L, and 0.8% S; T<sub>8</sub> = 0.5% N, 0.5 mg/L K, and 0.5% S; T<sub>9</sub> = 0.5% N, 0.8 mg/L, and 0.2% S; T<sub>10</sub> = 0.5% N, 0.8 mg/L K, and 0.8% S; T<sub>11</sub> = 0.8% N, 0.2 mg/L K, and 0.5% S; T<sub>12</sub> = 0.8% N, 0.5 mg/L K, and 0.2% S; T<sub>13</sub> = 0.8% N, 0.5 mg/L K, and 0.8% S; T<sub>14</sub> = 0.8% N, 0.8 mg/L K, and 0.5% S]

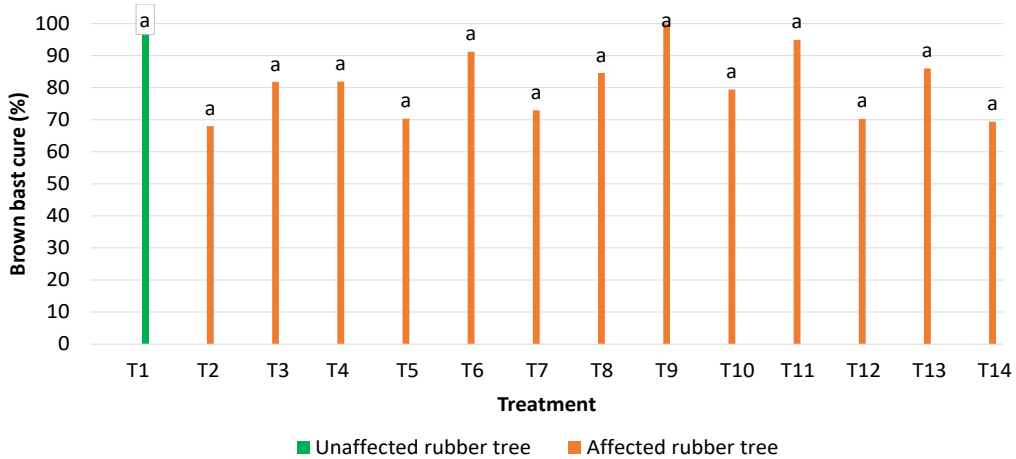


Figure 5. Percentage of brown bast cure after 4 weeks of treatment application

Note. Bars with the different letters are significantly different at  $p \leq 0.05$  according to Tukey's honest significant difference test [T<sub>1</sub> = Control; T<sub>2</sub> = 0.2% N, 0.2 mg/L K, and 0.5% S; T<sub>3</sub> = 0.2% N, 0.5 mg/L K, and 0.2% S; T<sub>4</sub> = 0.2% N, 0.5 mg/L K, and 0.8% S; T<sub>5</sub> = 0.2% N, 0.8 mg/L K, and 0.5% S; T<sub>6</sub> = 0.5% N, 0.2 mg/L K, and 0.2% S; T<sub>7</sub> = 0.5% N, 0.2 mg/L, and 0.8% S; T<sub>8</sub> = 0.5% N, 0.5 mg/L K, and 0.5% S; T<sub>9</sub> = 0.5% N, 0.8 mg/L, and 0.2% S; T<sub>10</sub> = 0.5% N, 0.8 mg/L K, and 0.8% S; T<sub>11</sub> = 0.8% N, 0.2 mg/L K, and 0.5% S; T<sub>12</sub> = 0.8% N, 0.5 mg/L K, and 0.2% S; T<sub>13</sub> = 0.8% N, 0.5 mg/L K, and 0.8% S; T<sub>14</sub> = 0.8% N, 0.8 mg/L K, and 0.5% S]

Table 5  
Effect of treatment application on the nutrient content of bark tissue

Factor	N	S	K	P	Mg	Ca
<i>ANOVA analysis</i>						
Treatment	0.847 <sup>ns</sup>	0.126 <sup>ns</sup>	0.777 <sup>ns</sup>	0.484 <sup>ns</sup>	0.090 <sup>ns</sup>	0.490 <sup>ns</sup>
Time	<0.0001*	0.0002*	0.280 <sup>ns</sup>	0.071 <sup>ns</sup>	0.676 <sup>ns</sup>	0.062 <sup>ns</sup>
Mean comparison of treatments						
<i>Time</i>						
Before	0.409 <sup>b</sup>	0.088 <sup>b</sup>	0.430 <sup>a</sup>	0.101 <sup>a</sup>	0.076 <sup>a</sup>	1.232 <sup>a</sup>
After	0.619 <sup>a</sup>	0.113 <sup>a</sup>	0.457 <sup>a</sup>	0.162 <sup>a</sup>	0.079 <sup>a</sup>	1.311 <sup>a</sup>
Least significant difference (LSD)						
<i>Treatment</i>						
T <sub>1</sub>	0.522 <sup>a</sup>	0.142 <sup>a</sup>	0.403 <sup>a</sup>	0.112 <sup>a</sup>	0.043 <sup>a</sup>	1.413 <sup>a</sup>
T <sub>2</sub>	0.595 <sup>a</sup>	0.097 <sup>a</sup>	0.507 <sup>a</sup>	0.118 <sup>a</sup>	0.085 <sup>a</sup>	1.316 <sup>a</sup>

Table 5 (continue)

Factor	N	S	K	P	Mg	Ca
T <sub>3</sub>	0.480 <sup>a</sup>	0.092 <sup>a</sup>	0.415 <sup>a</sup>	0.114 <sup>a</sup>	0.064 <sup>a</sup>	1.349 <sup>a</sup>
T <sub>4</sub>	0.534 <sup>a</sup>	0.095 <sup>a</sup>	0.501 <sup>a</sup>	0.117 <sup>a</sup>	0.077 <sup>a</sup>	1.231 <sup>a</sup>
T <sub>5</sub>	0.513 <sup>a</sup>	0.097 <sup>a</sup>	0.421 <sup>a</sup>	0.115 <sup>a</sup>	0.092 <sup>a</sup>	1.119 <sup>a</sup>
T <sub>6</sub>	0.460 <sup>a</sup>	0.092 <sup>a</sup>	0.423 <sup>a</sup>	0.117 <sup>a</sup>	0.086 <sup>a</sup>	1.274 <sup>a</sup>
T <sub>7</sub>	0.555 <sup>a</sup>	0.094 <sup>a</sup>	0.415 <sup>a</sup>	0.116 <sup>a</sup>	0.098 <sup>a</sup>	1.315 <sup>a</sup>
T <sub>8</sub>	0.533 <sup>a</sup>	0.116 <sup>a</sup>	0.512 <sup>a</sup>	0.342 <sup>a</sup>	0.077 <sup>a</sup>	1.165 <sup>a</sup>
T <sub>9</sub>	0.473 <sup>a</sup>	0.099 <sup>a</sup>	0.427 <sup>a</sup>	0.115 <sup>a</sup>	0.084 <sup>a</sup>	1.270 <sup>a</sup>
T <sub>10</sub>	0.515 <sup>a</sup>	0.092 <sup>a</sup>	0.444 <sup>a</sup>	0.120 <sup>a</sup>	0.056 <sup>a</sup>	1.227 <sup>a</sup>
T <sub>11</sub>	0.477 <sup>a</sup>	0.093 <sup>a</sup>	0.423 <sup>a</sup>	0.114 <sup>a</sup>	0.080 <sup>a</sup>	1.242 <sup>a</sup>
T <sub>12</sub>	0.481 <sup>a</sup>	0.109 <sup>a</sup>	0.472 <sup>a</sup>	0.113 <sup>a</sup>	0.082 <sup>a</sup>	1.268 <sup>a</sup>
T <sub>13</sub>	0.515 <sup>a</sup>	0.095 <sup>a</sup>	0.411 <sup>a</sup>	0.110 <sup>a</sup>	0.071 <sup>a</sup>	1.260 <sup>a</sup>
T <sub>14</sub>	0.546 <sup>a</sup>	0.093 <sup>a</sup>	0.445 <sup>a</sup>	0.120 <sup>a</sup>	0.092 <sup>a</sup>	1.352 <sup>a</sup>
LSD	0.241	0.055	0.229	0.305	0.057	0.384

Note. ns = Not significant at  $p>0.05$ ; \* = Significant at  $p\leq 0.05$ ; Means followed by the same letters in a column are not significantly different at  $p>0.05$  using Tukey's honest significant difference test [T<sub>1</sub> = Control; T<sub>2</sub> = 0.2% N, 0.2 mg/L K, and 0.5% S; T<sub>3</sub> = 0.2% N, 0.5 mg/L K, and 0.2% S; T<sub>4</sub> = 0.2% N, 0.5 mg/L K, and 0.8% S; T<sub>5</sub> = 0.2% N, 0.8 mg/L K, and 0.5% S; T<sub>6</sub> = 0.5% N, 0.2 mg/L K, and 0.2% S; T<sub>7</sub> = 0.5% N, 0.2 mg/L, and 0.8% S; T<sub>8</sub> = 0.5% N, 0.5 mg/L K, and 0.5% S; T<sub>9</sub> = 0.5% N, 0.8 mg/L, and 0.2% S; T<sub>10</sub> = 0.5% N, 0.8 mg/L K, and 0.8% S; T<sub>11</sub> = 0.8% N, 0.2 mg/L K, and 0.5% S; T<sub>12</sub> = 0.8% N, 0.5 mg/L K, and 0.2% S; T<sub>13</sub> = 0.8% N, 0.5 mg/L K, and 0.8% S; T<sub>14</sub> = 0.8% N, 0.8 mg/L K, and 0.5% S]

The results indicate that the applications of deficient macronutrients could cure the brown bast problem in rubber trees and restore latex generation similar to healthy rubber trees, as shown in Figure 6. Adequate fertilizer application does not only positively impact economic growth but could equally minimize nutrient loss. Moreover, adequate nutrients could maintain rubber trees' healthy growth and yield (Njukeng et al., 2013). Keralafarmer (2010) stated that nutrient deficiency is one of the factors that cause brown bast in rubber trees. In Malaysia, no more study has been done on curing brown bast syndrome with the addition of deficient

macronutrients. Therefore, this finding sheds new light on managing BB syndrome in rubber plantations.

It can be noted that there was a significant difference ( $p<0.047$ ) between S and Mg contents in the bark of *H. brasiliensis* after treatment application, with a negative correlation ( $r=-0.539$ ) between these elements (Table 6). The increased level of Mg in the rubber bark would decrease the level of S in the bark. Magnesium is generally essential for photosynthesis and latex production. However, the excessive use of fertilizer containing Mg would cause latex instability (Karunanayake &



Figure 6. Experimental rubber trees: (a) before treatment application (no latex flow) and (b) after treatment application (with latex low)

Priyanthi Perera, 2006), resulting in pre-coagulation and increased brown bast problems. Phosphorus is another essential macronutrient that is necessary for the growth and development of rubber trees. It plays a significant role in the plant's photosynthesis, respiration, and energy transfer. Adequate phosphorus supply increases latex production and improves the quality of latex.

Sulfur is generally essential in protein synthesis, enzyme activation, and

photosynthesis. According to Kashyap (2009), the sulfur requirement of plants is more significant than P. Meanwhile, Mg ranks as the least abundant macronutrient in plants compared to Ca, P, and S. Therefore, S was required in higher quantities than Mg.

Therefore, when the deficient macronutrients were supplied to the plant, especially applied directly to the trunk, the bark absorbed the nutrients, and the deficiency was recovered, which led the rubber trees to make regular latex flow.

Table 6  
Correlation between nutrient content and brown bast cure of *Hevea brasiliensis*

	N	P	K	Ca	Mg	S
N	1.000	0.165 0.573	0.438 0.117	0.120 0.684	0.105 0.720	0.083 0.777
P	0.165 0.573	1.000	0.525 0.054	-0.404 0.152	0.001 0.997	0.321 0.263
K	0.438 0.117	0.525 0.054	1.000	-0.330 0.249	0.198 0.497	0.061 0.835

Table 6 (continue)

	N	P	K	Ca	Mg	S
Ca	0.120	-0.404	-0.330	1.000	-0.318	0.299
	0.684	0.152	0.249		0.268	0.299
Mg	0.105	0.001	0.198	-0.318	1.000	-0.539
	0.720	0.997	0.497	0.268		0.047
S	0.083	0.321	0.061	0.299	-0.539	1.000
	0.777	0.263	0.835	0.299	0.047*	

Note. \* = Significant at  $p \leq 0.05$

### Effect of New Formulation on Moisture Content in Bark Tissue

The moisture content in the bark indicated a significant difference ( $p < 0.001$ ) between before and after treatment (Table 7). The moisture content before treatment application was higher (42.8%) than after treatment application (39.6%). Several factors can contribute to the development of this BB syndrome. Moisture content in the bark has been identified as a key factor of this syndrome (Cheng et al., 2019; Zhang et al., 2021).

High moisture levels in the bark create conditions that favor the growth of microorganisms, including fungi and bacteria, and can cause damage to the bark

and cambium tissue. It can also lead to brown bast syndrome in rubber trees (Cheng et al., 2019). On the other hand, maintaining appropriate moisture levels in the bark can help prevent the development of brown bast syndrome.

The low moisture content in the bark after treatment initiates a greater ability of barks to absorb water. That also helped the plants to absorb nutrients and improved latex production. It also increases the viscosity of latex. According to Lin and Lai (2009), the rheological behavior of plant hydrocolloids can be improved by using water, and the viscosity results from the polymer charge in water.

Table 7  
Effect of treatment application on the moisture contents of bark tissue

Factor	Moisture content
ANOVA analysis	
Treatment	0.115 <sup>ns</sup>
Time	<0.001*
Mean comparison of treatment	(%)

Table 7 (continue)

Factor	Moisture content
Time	
Before	42.786 <sup>a</sup>
Factor	Moisture content
After	39.593 <sup>b</sup>
Least significant difference (LSD)	1.881

Table 7 (continue)

Factor	Moisture content
<i>Treatment</i>	
T <sub>1</sub>	42.175 <sup>a</sup>
T <sub>2</sub>	43.820 <sup>a</sup>
T <sub>3</sub>	42.563 <sup>a</sup>
T <sub>4</sub>	40.973 <sup>a</sup>
T <sub>5</sub>	42.652 <sup>a</sup>
T <sub>6</sub>	37.562 <sup>a</sup>
T <sub>7</sub>	41.797 <sup>a</sup>
T <sub>8</sub>	43.222 <sup>a</sup>
T <sub>9</sub>	41.473 <sup>a</sup>
T <sub>10</sub>	37.792 <sup>a</sup>
T <sub>11</sub>	44.292 <sup>a</sup>
T <sub>12</sub>	41.067 <sup>a</sup>
T <sub>13</sub>	38.783 <sup>a</sup>
T <sub>14</sub>	38.477 <sup>a</sup>
LSD	8.715

Note. ns = Not significant at  $p > 0.05$ ; \* = Significant at  $p \leq 0.05$ ; Means followed by the same letters in a column are not significantly different at  $p > 0.05$  using Tukey's honest significant difference test [T<sub>1</sub> = Control; T<sub>2</sub> = 0.2% N, 0.2 mg/L K, and 0.5% S; T<sub>3</sub> = 0.2% N, 0.5 mg/L K, and 0.2% S; T<sub>4</sub> = 0.2% N, 0.5 mg/L K, and 0.8% S; T<sub>5</sub> = 0.2% N, 0.8 mg/L K, and 0.5% S; T<sub>6</sub> = 0.5% N, 0.2 mg/L K, and 0.2% S; T<sub>7</sub> = 0.5% N, 0.2 mg/L, and 0.8% S; T<sub>8</sub> = 0.5% N, 0.5 mg/L K, and 0.5% S; T<sub>9</sub> = 0.5% N, 0.8 mg/L, and 0.2% S; T<sub>10</sub> = 0.5% N, 0.8 mg/L K, and 0.8% S; T<sub>11</sub> = 0.8% N, 0.2 mg/L K, and 0.5% S; T<sub>12</sub> = 0.8% N, 0.5 mg/L K, and 0.2% S; T<sub>13</sub> = 0.8% N, 0.5 mg/L K, and 0.8% S; T<sub>14</sub> = 0.8% N, 0.8 mg/L K, and 0.5% S]

### Effect of New Formulation on Latex Production

The effect of treatment application indicated a significant ( $p < 0.05$ ) increment in latex yield (Table 8). For instance, in December 2019, the healthy plants (T<sub>1</sub>) had a higher latex yield (79.69 g/tapping/tree) than other treatments (T<sub>2</sub> to T<sub>14</sub>), except for T<sub>3</sub>

(62.09 g/tapping/tree). In January 2020, significant differences were noticed between T<sub>1</sub> with, T<sub>4</sub> and T<sub>12</sub>. However, there was no significant difference between T<sub>1</sub> with T<sub>3</sub>, T<sub>9</sub>, T<sub>2</sub>, T<sub>5</sub>, T<sub>7</sub>, T<sub>8</sub>, and T<sub>13</sub>. The treatment T<sub>3</sub> produced the highest yield (57.73 g/tapping/tree) after two months of treatment application, similar to T<sub>9</sub> (51.73 g/tapping/tree). Macronutrients are essential nutrients required in large quantities by rubber trees to grow and produce high-quality latex (Chowdhury et al., 2019; Zhou et al., 2018). For example, an adequate nitrogen supply increases the number of latex vessels and the latex yield.

However, excessive nitrogen supply can lead to the production of low-quality latex. Potassium is an essential macronutrient that regulates the plant's water balance and enhances its stress resistance. It also increases the number of latex vessels, the latex yield, and the quality of latex. Calcium is important for the structural development of rubber trees and is related to the thickness and elasticity of the latex vessels, increases the latex yield, and improves the quality of latex. Magnesium is necessary for the synthesis of chlorophyll, which is essential for photosynthesis. Adequate magnesium supply also increases the latex yield and improves the quality of latex. In summary, macronutrients are crucial in rubber trees' growth and development and significantly affect latex production and quality. A balanced supply of these nutrients is essential for high latex yield and quality.

Salisu and Daud (2016) reported that the optimum fertilizer level was achieved at 150% of the standard dose (780 kg/ha of a

Table 8  
*Latex yield (g/tapping/tree) of rubber trees after treatment application*

Factor	Latex yield			
	November 2019	December 2019	January 2020	February 2020
<i>ANOVA analysis</i>				
Treatment	<0.0001*	0.002*	0.046 <sup>ns</sup>	0.002*
(g)				
<i>Mean comparison of treatment</i>				
<i>Treatment</i>				
T <sub>1</sub>	73.86 <sup>a</sup>	79.69 <sup>a</sup>	74.53 <sup>a</sup>	62.20 <sup>a</sup>
T <sub>2</sub>	0 <sup>b</sup>	14.67 <sup>c</sup>	29.66 <sup>abc</sup>	24.39 <sup>bcd</sup>
T <sub>3</sub>	0 <sup>b</sup>	62.09 <sup>ab</sup>	57.73 <sup>ab</sup>	40.44 <sup>b</sup>
T <sub>4</sub>	0 <sup>b</sup>	9.42 <sup>c</sup>	10.40 <sup>c</sup>	11.39 <sup>d</sup>
T <sub>5</sub>	0 <sup>b</sup>	20.87 <sup>c</sup>	52.11 <sup>abc</sup>	37.81 <sup>bc</sup>
T <sub>6</sub>	0 <sup>b</sup>	20.41 <sup>c</sup>	20.14 <sup>bc</sup>	20.30 <sup>bcd</sup>
T <sub>7</sub>	0 <sup>b</sup>	32.33 <sup>bc</sup>	38.56 <sup>abc</sup>	30.57 <sup>bcd</sup>
T <sub>8</sub>	0 <sup>b</sup>	10.71 <sup>c</sup>	32.50 <sup>abc</sup>	26.94 <sup>bcd</sup>
T <sub>9</sub>	0 <sup>b</sup>	22.34 <sup>c</sup>	51.85 <sup>abc</sup>	26.57 <sup>bcd</sup>
T <sub>10</sub>	0 <sup>b</sup>	15.68 <sup>c</sup>	18.98 <sup>bc</sup>	14.93 <sup>cd</sup>
T <sub>11</sub>	0 <sup>b</sup>	40.41 <sup>bc</sup>	22.48 <sup>bc</sup>	20.77 <sup>bcd</sup>
T <sub>12</sub>	0 <sup>b</sup>	6.51 <sup>c</sup>	9.60 <sup>c</sup>	7.93 <sup>d</sup>
T <sub>13</sub>	0 <sup>b</sup>	36.36 <sup>bc</sup>	43.17 <sup>abc</sup>	30.44 <sup>bcd</sup>
T <sub>14</sub>	0 <sup>b</sup>	16.33 <sup>c</sup>	12.68 <sup>bc</sup>	12.19 <sup>d</sup>
Least significant difference	0.1055	56.259	70.499	38.734

*Note.* ns = Not significant at  $p > 0.05$ ; \* = Significant at  $p \leq 0.05$ ; Means followed by the same letters in a column are not significantly different at  $p > 0.05$  using Tukey's honest significant difference test [T<sub>1</sub> = Control; T<sub>2</sub> = 0.2% N, 0.2 mg/L K, and 0.5% S; T<sub>3</sub> = 0.2% N, 0.5 mg/L K, and 0.2% S; T<sub>4</sub> = 0.2% N, 0.5 mg/L K, and 0.8% S; T<sub>5</sub> = 0.2% N, 0.8 mg/L K, and 0.5% S; T<sub>6</sub> = 0.5% N, 0.2 mg/L K, and 0.2% S; T<sub>7</sub> = 0.5% N, 0.2 mg/L K, and 0.8% S; T<sub>8</sub> = 0.5% N, 0.5 mg/L K, and 0.5% S; T<sub>9</sub> = 0.5% N, 0.8 mg/L K, and 0.2% S; T<sub>10</sub> = 0.5% N, 0.8 mg/L K, and 0.8% S; T<sub>11</sub> = 0.8% N, 0.2 mg/L K, and 0.5% S; T<sub>12</sub> = 0.8% N, 0.5 mg/L K, and 0.2% S; T<sub>13</sub> = 0.8% N, 0.5 mg/L K, and 0.8% S; T<sub>14</sub> = 0.8% N, 0.8 mg/L K, and 0.5% S]

blend of 10–16–9-2). A study by Chowdhury et al. (2019) reported a significant increase in latex yield by 18.3% with NPK fertilizers. Mokhtatar et al. (2012) reported that the currently recommended fertilizer doses are insufficient, and for optimum growth, a precise fertilizer application should

be considered to optimize fertilizer use efficiency. However, in February 2020, the latex yield was lowered in the treated rubber trees (T<sub>2</sub> to T<sub>14</sub>) along with healthy plants compared to January yields, which might be due to annual wintering (February to May) and leaf senescence in Malaysia (Qi



et al., 2014). The wintering caused leaves to fall and disturbed photosynthetic activity, directly affecting latex production.

## CONCLUSION

Since significant differences in macronutrient contents, especially in the bark of brown bast-affected rubber trees, are found, the nutrient-adjusted formulation of fertilizers is the best option to cure the brown bast disease and to improve the latex yield. Treatment T<sub>9</sub>, comprised 0.5% N, 0.8 mg/L K, and 0.2% S is the best treatment (BB cure = 100%, yield = 51.85 g/tapping/tree) to cure the brown bast of rubber trees for the study area. The treatment T<sub>3</sub> (0.2% N, 0.5 mg/L K, and 0.2% S) can also be used, as it caused an 80% cure of BB syndrome after four weeks of treatment application and produced the highest latex yield (57.73 g/tapping/tree) after two months of treatment application. The curing of brown bast by applying this nutrient-adjusted formulation caused stability of latex flow, increment in nutrient content, and improved the latex yield. Therefore, it is recommended that first, the difference in macronutrients in the barks of BB-affected and healthy plants of any concerned area should be studied, and the deficiency, if any, should be adjusted in the liquid fertilizer formulation for treating the brown bast syndrome and to increase latex yield of rubber trees. It is the first report on the cure of brown bast syndrome in rubber trees by the trunk application of deficient-adjusted macronutrients.

## ACKNOWLEDGEMENTS

The authors would like to thank Universiti Putra Malaysia (UPM) for the financial support (grant number 9645300) under the Graduate Research Fellowship (GRF) program of UPM to complete this research successfully. We would also like to thank the staff of FELCRA, Simpang Renggam, Johor, Malaysia for assisting in data collection and field selection. We gratefully acknowledge all the lab assistants of UPM Serdang, Selangor, who graciously contributed to the success of this work.

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