Original Research Article

Effect of the Stimulating Paste Conservation Period and Economic Benefit of Ethrel-Palm oil Mixture in Rubber Tree Plantations in South-Eastern Côte d'Ivoire

Abstract

To maintain and increase sustainable productivity and natural rubber yield, the hormonal stimulation management of rubber yield should be controlled from operation centers. A study was conducted to determine the type of prepared stimulant paste and the maximum efficiency period of this paste in southeastern Côte d'Ivoire, during two years from 2012 to 2014. The study was conducted on the clone GT 1, according to single tree plot design of 15 treatments of 30 replicate trees per treatment, corresponding to different conservation periods of the prepared stimulant paste. Two stimulant products namely Ethrel mixed with palm oil and Almephon (ready-to-use), have been stored at ambient temperature and left for cool. The prepared stimulant paste was kept for 0, 3, 7, 14, 21, 28, 42 and 56 days. The results indicated that rubber yield, radial vegetative growth, physiological profile and health of rubber trees haven't been affected by both conservation of the stimulant paste and different conservation periods. However the prepared stimulant paste could be kept up to 56 days, at ambient temperature as well as in cold, without losing their efficiency. These important results help predict the management of hormonal stimulation of rubber yield, without electrical power, from centers which will ensure distribution to different plantations. The stimulant paste Ethrel-palm oil substantially cheaper, with an important local and profitable product.

Keywords: Hevea brasiliensis, Hormonal stimulations, Conservation periods, Rubber yield, Côte d'Ivoire.

1. INTRODUCTION

The tapping of trees from a plantation certainly yields rubber. However, the latex production of mature rubber tree is limited as the latex cells already prepared latex when its reached to tappable girth. It can't be adjusted to the needs of users and especially the tapping cannot alone enable the recovery of the tree yield potential [1]. Today, systematically, hormonal stimulation of the yield is added to the tapping system [2-4]. This consists in preparing the stimulant paste or by diluting the stimulant product ready for use, or mixing Ethrel (containing the Ethephon which is the active matter (a. m.) to palm oil in order to obtain concentrations in a. m. Ethephon, of 2.5 or 5 % [5-8] and then to apply to the tree in order to improve its productivity rubber [5,9].

Unfortunately in Africa and particularly in Côte d'Ivoire, where rubber cultivation is dominated by smallholders, including villagers [10]. And also, the controlling of the use of the practice of stimulation of the production is very little controlled [11] or even in disorder. Indeed, in hope of substantially increasing rubber yield, the tapper and/or farmer practices, repeatedly and excessively hormonal stimulation, concern the frequency of application as well as the concentration in the active matter (Ethephon) during the preparation of the stimulant paste. These poor practices can affect in the long-term the latex flow (tapping panel dryness) and leading to lowering the yield [3,11,12]. The distribution of stimulant pastes correctly prepared to rubber plantations of smallholders' sector, especially villagers, from management centers of plantation stimulation operations (preparation and distribution) is a plausible solution to the control of this operation. The option of controlling hormonal stimulation on rubber yield of smallholders, is centralizing the preparation of the stimulant paste and its distribution from center operation. Although this is a best initiative but keep on observation of the stimulant power of the paste prepared at the time of application. Since, the time between the preparations of the stimulant paste and its application can vary and be long according to distance from the center to the appropriate plantation and the necessary logistics. This period may reasonably exceed one week and raise thus the problem of the efficiency of this stimulant paste at the time of application. This, especially in common usage and, efficient and recommended is the preparation of the stimulant paste on the day of stimulation of the tree although. Also, some works have reported that once prepared, the stimulant could be kept for about one week at room temperature without loosing its efficiency [5].

The period between the preparations of the stimulant paste and its application may also vary due to the nature of the stimulant paste (ready to use or Ethrel and palm oil mixture) even though their effect on rubber productivity is similar [7,13,14]. Moreover, by the different nature of the two types of stimulant products, they will not necessarily have the same market value. Thus, in the interests of efficiency and lower cost, it would be appropriate to make the choice of the best of these two stimulants in order to recommend its use for rubber industries especially of smallholders.

In order to deal with the above facts of national rubber industries, a study on the effect of conservation periods and cost of the stimulant paste on rubber productivity of rubber trees was conducted during two years in southeastern of Côte d'Ivoire. This paper discuss only on the important results.

2. MATERIALS AND METHODS

2.1. Materials

Plant Material

The experiment was carried out on the experimental Research Station of *Centre National de Recherche Agronomique* (CNRA) of Bimbresso, located in Anguédédou, southeastern of Côte d'Ivoire. The plant material consisted of clone GT 1 of *Hevea brasiliensis,* belonging to the class

of moderate vegetative growth and also moderate metabolic activity [15,16]. Clone GT 1 (*Gondang Tappen* or GT) originates from Malaysia. Its yield per tree isn't very high but is largely compensated by the good homogeneity and secondary characteristics such as physiological parameters with the exception of moderate inorganic phosphorus content expressing an average activity metabolism. These physiological characteristics are favorable to the yield [17]. This clone was planted in 1997, on a monoclonal plot with an area of 6.25 hectares at a density of 555 trees/ha (6 m x 3 m). The trees were opened in 2003 and the experiment was began in 2012 in virgin bark of high panel (S/4 d4). Before the experiment, the trees were tapped in virgin bark in half spiral downward every four days, with one day rest in the week, 12 months on 12 (S/2 d4 6d/7 12 m/12) during nine years. There were stimulated with Ethrel-oil palm at the concentration of 2.5 % of Ethephon.

Stimulant products

The stimulant products used were marketed under the names of Ethrel and Almephon were manufactured respectively by ALM West Africa and CFPI. They were applied 48 hours before tapping. The concentration of active ingredient (Ethephon) for each of the stimulant products used in this experiment was 5 %. The Almephon, dose at 10 % Ethephon at delivery, is a ready to use product. As for the Almephon, Ethrel is dose at 480 g of active matter (Ethephon) per liter ($g.a.m.l^{-1}$). The density of product was 1.2 and it titles 400 g of active matter per kilogram ($g.a.m.kg^{-1}$).

2.2. Experimental design

The experimental design adopted was a " *single tree plot design* " where each treatment applied 30 replicate rubber tress as described bellow. The trees selected were distributed into 15 treatments corresponding to different modes of conditioning and conservation periods of the stimulant paste prepared and applied, 30 replicate trees per treatment. All the trees were tapped in quater spiral upward every four days, with one day rest in the week, 12 months on 12 (S/4U d4 6d/7 12 m/12). The stimulant products marketed (namely Ethrel and Almephon) dialuted with oil palm and clean water respectively. These stimulant pastes used to stimulate selected trees as described above. These pastes were conserved as showed in the Table 1, before being applied to the trees. The temperature for the cold condition kept was 4℃. Trees stimulated with the Ethrel whose stimulant paste was prepared and conserved at ambient temperature during 7 days, made up the control 1 (ETH-air 7). Similarly, trees stimulated with Almephon whose stimulant paste was prepared the day of the stimulation of the trees, made up the control 2 (ALM-air-Dd). The study was conduct during two years (2012-2014).

Table 1. Different treatments applied to trees of clone GT 1

ETHREL	ALMEPHON
ETH-air-Dd : stimulant paste prepared from Ethrel on the same day of use	**ALM-air-Dd :** stimulant paste prepared from Almephon on the same day of use (**Control 2**)
ETH-air-3 : stimulant paste prepared from Ethrel and conserved at room temperature during 3 days	**ALM-air-3 :** stimulant paste prepared from Almephon and conserved at room temperature during 3 days
ETH-air-7 : stimulant paste prepared from Ethrel and conserved at room temperature during 7 days (**Control 1**)	**ALM-air-7 :** stimulant paste prepared from Almephon and conserved at room temperature during 7 days
ETH-air-14 : stimulant paste prepared from Ethrel and conserved at room temperature during 14 days	**ALM-air-14 :** stimulant paste prepared from Almephon and conserved at room temperature during 14 days
ETH-air-21 : stimulant paste prepared from Ethrel and conserved at room temperature during 21 days	**ALM-air-21 :** stimulant paste prepared from Almephon and conserved at room temperature during 21 days
ETH-air-28 : stimulant paste prepared from Ethrel and conserved at room temperature during 28 days	**ALM-air-28 :** stimulant paste prepared from Almephon and conserved at room temperature during 28 days
ETH-air-42 : stimulant paste prepared from Ethrel and conserved at room temperature during 42 days	**ALM-air-42 :** stimulant paste prepared from Almephon and conserved at room temperature during 42 days
ETH-air-56 : stimulant paste prepared from Ethrel and conserved at room temperature during 56 days	**ALM-air-56 :** stimulant paste prepared from Almephon and conserved at room temperature during 56 days
ETH-cold-3 : stimulant paste prepared from Ethrel and conserved at cold condition during 3 days	**ALM-cold-3 :** stimulant paste prepared from Almephon and conserved at cold condition during 3 days
ETH-cold-7 : stimulant paste prepared based Ethrel and conserved at cold condition during 7 days	**ALM-cold-7 :** stimulant paste prepared from Almephon and conserved at cold condition during 7 days
ETH-cold-14 : stimulant paste prepared from Ethrel and conserved at cold condition during 14 days	**ALM-cold-14 :** stimulant paste prepared from Almephon and conserved at cold condition during 14 days
ETH-cold-21 : stimulant paste prepared from Ethrel and conserved at cold condition during 21 days	**ALM-cold-21 :** stimulant paste prepared from Almephon and conserved at cold condition during 21 days
ETH-cold-28 : stimulant paste prepared from Ethrel and conserved at cold condition during 28 days	**ALM-cold-28 :** stimulant paste prepared from Almephon and conserved at cold condition during 28 days
ETH-cold-42 : stimulant paste prepared from Ethrel and conserved at cold condition during 42 days	**ALM-cold-42 :** stimulant paste prepared from Almephon and conserved at cold condition during 42 days
ETH-cold-56 : stimulant paste prepared from Ethrel and conserved at cold condition during 56 days	**ALM-cold-56 :** stimulant paste prepared from Almephon and conserved at cold condition during 56 days

ETH: Ethrel, ALM: Almephon, 0…56: During of stimulant paste after its preparation (days),

Dd: 0 day.

2.3. Preparation and application of stimulant paste

The two types of stimulant paste were prepared by adding water to the Almephon (v/v) and oil palm to the Ethrel then the product was mixed until complete homogenization.

For the application of stimulant paste based on Almephon, measuring in the preparation of 10 % in this experiment, the same volume of water as that of Almephon so as to bring its concentration in Ethephon 5 %. Thus, for 1 g of stimulant paste 5 % Ethephon, 0.5 g of water was mixed with 0.5 g of Almephon.

In the case of Ethrel, the quantities of Ethrel and of oil palm to be mixed to obtain the required concentration are such that for 1 g of stimulant paste of concentration 5 % Ethephon, mixed 0.125 g of Ethrel to 0.875 g of palm oil. This mixture, the same as that of Almephon was applied per tree per stimulation. The stimulation was made in this study, on a band of 1 cm wide on the tapping cut and parallel to and above thereof in an amount of 0.7 g of stimulant paste per tree per application [6,7,18]. Exception of stimulant pastes used on the day of their preparations, other pastes were conserved at different times or in cold, or in the open air as shown in Table 1.

2.4. Measurements and data processing

Latex Yield

Each replicate tree latex yield collected in the field was measured by weighing the cumulative coagulated rubber in every four weeks. The transformation coefficient (CT), which is the rate of dry matter of a given sample of fresh rubber, was used to calculate the yield of dry rubber in grams per tree ($g.t^{-1}$).

Latex Micro Diagnosis

Some collected latex helped determine annually, by the method of Latex Micro Diagnosis (LMD), the value of dry rubber content (DRC (%)) and dose the sucrose (Suc), inorganic phosphorus (iP) and thiol groups (R-SH) contents of the latex, expressed in $mmol.l^{-1}$. In practice, the dry matter content of the latex was determined by weighing 1 ml of latex into a pillbox of 10 ml, before and after drying in oven at 80 °C during 24 h. But long before, the vacuum out of the pillbox was determined. Then the difference in weight expressed in percentage shows the rate of dry content. Inorganic phosphorus, sucrose and thiol groups were determined from the trichloroacetic acid serum (TCA-serum) obtained by mixing 1 ml of latex and 9 ml of 2.5 % TCA. Using a wand, the coagulated rubber was squeezed out and separated from 2.5 % TCA. Finally, the different dosages were applied:

- sucrose (Suc) was assayed by the method of [19] based on anthrone that in the presence of concentrated acid, hexoses dehydrate to give a furfural reacting with anthrone giving a blue-green coloring whose optical density (OD) is read at the wavelength of 627 nm ;

- inorganic phosphorus (Pi) was assayed by the method of [20] method using molybdate ammonium : phosphorus is complexed with an excess of molybdate ammonium. The complex formed is then reduced by ferrous sulfate, a blue color, whose absorbance value is read at the wavelength 680 nm ;
- thiol groups (R-SH) was performed by the method described by [21] using dinitro-2,2'-dithio-5-5'-dibenzoic acid (BTNB) thiol groups react with the acid to give the thio-nitro-benzoic acid (TNB) high-absorbency at 412 nm. TNB is revealed by Tris buffer (yellow coloration).

Thus, for each treatment, the characteristics of different clones were determined using the latex micro diagnosis.

Radial vegetative growth

The radial growth (Girth) was measured at 1.70 m above the ground on each tree treated at the beginning and the end of the experiment. The average annual girth increment (cm.year^{-1}) was determined by the following relation :

$$\boxed{Girth_{inc} = (Girth_{beginning} - Girth_{end}) / i}$$

With **$Girth_{inc}$:** Girth increment; **$Girth_{end}$:** tree girth at the end of the experiment ; **$Girth_{beginning}$:** initial girth of trees at the beginning of experiment ; **i :** number of years of experiment.

Visual estimation of the tapping panel dryness

The method of quick survey by visual assessment helped to account for the onset and progress of tapping panel dryness. For each tree tapped, it was assigned a number between 0 and 6.
The specific account of trees' condition was performed and the rate of length of dry cut (LDC (%)) for each treatment was calculated as follows:

$$\boxed{LDC\ (\%) = (0.1\ n_1 + 0.3\ n_2 + 0.5\ n_3 + 0.7\ n_4 + 0.9\ n_5 + n_6 + DT) \times N^{-1}}$$

N : Total number of trees ; **n_i :** Number of trees per tapping panel dryness class ; DT **:** Number of trees whose tapping has already been stopped due to full tapping panel dryness.

2.5. Determination of the best stimulant product

Evaluation of agro physiological parameters

The rubber yield in gram per tree (g.t^{-1}), the average annual increase in girth, as well as the physiological parameters of latex micro diagnosis of two types of stimulants were assessed. The above parameters were used to determine the best stimulant product.

Evaluation of Economical parameters

The cost of stimulation per hectare with each type of stimulant product were determined by the fallowing calculations. The number of trees per hectare is about 500.

$Cost_{Ha}Stim_{ET} = Cost_{ET} + Cost_{OP}$ (1)

$Cost_{Ha}Stim_{Alm} = Nb_{Tree/ha}$ x 0.7 g/tree x 5 USD x 500 g^{-1} (2)

With:

$Cost_{ET} = V_{ET}$ (L) x 13 USD.L^{-1}

V_{ET} (L) = $Nb_{Tree/ha}$ x 0.7 g/tree x 0.0625/1200 g x L

$Cost_{OP} = V_{HP}$ (L) x 1 USD. L^{-1}

V_{OP} (L) = $Nb_{Tree/ha}$ x 0.7 g/tree x 0.9375/900 g x L

2.6. Statistical analysis

Rubber yield data were performed using the software XL-STAT, Pro 6.1.9. The level of significance of differences between averages was estimated by the Duncan test at 5 % threshold level.

3. RESULTS AND DISCUSSION

3.1. Effects of the conservation period of the stimulant products on their efficiency

Agro physiological parameters

Agronomical, physiological and TPD parameters of rubber trees treated with Ethrel are presented in Table 2. The rubber yield of the control 1 (927.215 $g.t^{-1}$) was statistically identical to those of all other periods except of those periods ETH- cold-21 (685.625 $g.t^{-1}$) and ETH-cold-56 (1139.744 $g.t^{-1}$). Indeed, the control trees had a significantly (p>o.o5) higher rubber yield than that of ETH-cold-21 treatment and significantly (p>o.o5) lower than that of ETH-cold-56 treatment.

With regard to the physiological profile, rubber yield of rubber trees treated with stimulant pastes prepared with Almephon and also subject to different conservation periods are expressed in $g.t^{-1}$ and presented in Table 3. According to the results showed that rubber yields of control 2 (1153.947 $g.t^{-1}$) have been statistically similar to those of other treatments except the treatments of ALM-cold-3 (1382.911 $g.t^{-1}$) and ALM-cold-56 (984.615 $g.t^{-1}$). And also the yields of control 2 have been, significantly higher than those of ALM-cold-56 treatment and lower than those of ALM-cold-3 treatment.

In overall whatever the stimulant paste used, the rubber yield haven't been influenced either by the mode of conservation of the stimulant paste or either by the different conservation periods.

As for latex micro diagnosis (LMD) results which showed in Table 2 and indicated that the dry rubber content (DRC), sucrose and inorganic phosphorus contents are statistically equivalent for all the trees of the different treatments of stimulant paste based on Ethrel. The trees of the different treatments had on average very high DRC (54.98 %), average sucrose contents (7.23

mmol.l^{-1}) and inorganic phosphorus (16.63 mmol.l^{-1}). When considering the thiol contents, they varied from 0.10 to 1.45 mmol.l^{-1}. Thiol contents of control treatments statistically equivalent to those of other treatments except the treatments of ETH-air Dd (1.38 mmol.l^{-1}) and ETH-cold-56 (1.45 mmol.l^{-1}) which were the highest.

The results of physiological parameters of the rubber trees treated with the stimulant pastes prepared and based on Almephon are shown in Table 3. The values of dry rubber content (DRC), of sucrose and inorganic phosphorus contents are statistically equivalent for all the trees of the different treatments. The trees of the different treatments had on average very high rates of DRC (55.83 %), average sucrose contents (6.88 mmol.l^{-1}) and inorganic phosphorus (16.74 mmol.l^{-1}). With regard to the thiols contents, they have fluctuated between 0.11 and 1.64 mmol.l^{-1}. The thiol contents of the control have been statistically identical to those of the other treatments and significantly higher than those of treatments ALM-air-56 (0.12 mmol.l^{-1}), ALM-cold-42 (0.13 mmol.l^{-1}) and ALM-cold-56 (0.11 mmol.l^{-1}). The highest thiol contents were recorded by the treatments ALM-air-Dd (0.70 mmol.l^{-1}), ALM-air-3 (1.64 mmol.l^{-1}), ALM-air-14 (0. 66 mmol.l^{-1}) and ALM-cold-3 (1.02 mmol.l^{-1}).

The results of the latex micro diagnosis have supported characterizing the trees subjected to different conservation periods and conditioning methods. Thus, mean value of inorganic phosphorus contents of the trees of clone GT 1 were 16.63 mmol.l^{-1} and 16.74 mmol.l^{-1} corresponded to the trees treated with Ethrel and Almephon respectively. This is related to the metabolic functioning of this clone with moderate metabolism as shown by several authors [13,17,22-25].

The rates of dry rubber content (DRC) of trees of the different treatments were all equivalent and very high (Tables 2 and 3). They were 54.98 and 55.83 % on average, corresponding to the trees treated with Ethrel and Almephon respectively. The high rate of DRC is the expression of a very good regeneration of latex exported during tapping [26]. This is consecutive to stimulation as shown by many authors [22,25,24]. The high contents in thiol groups of trees subjected to different treatments were favorable to yield [17], as there are positive correlations between thiol contents and yield [22].

Furthermore, the evolution in thiol group contents of the different treatment groups was independent of the conservation periods of stimulant pastes. The significant variations of the different thiol groups contents might be due to their degradation, related to the conservation period of the serum-TCA intended to their dosage [1].

The sucrose content of the latex of all trees treated was an average, because the use of sucrose for the biosynthesis of rubber [27] is important [11]. Indeed, the ethylene stimulation highly activates the metabolism of the clones and the result is a high transformation of the sucrose present in latex vessels, into rubber [11,28,29]. Thus, a strong response to the stimulation of a clone makes predict that there is a good availability of sucrose within latex vessels, reflecting a good supply [23]. Overall, the characteristics of physiological parameters of

the latex of trees of the different treatments were all similar to each other. This is the good sign that the various periods and conservation methods of the stimulant pastes were statistically equivalent and do not affect negatively the physiological parameters of the latex of trees. These characteristics are favorable for rubber yield of clone GT 1 [17], and are corroborated by a very good average annual girth increment, absolutely above the national average of this clone (> 2 cm.year^{-1}) in Côte d'Ivoire.

The isodiametric growth of the trunk of trees treated with Ethrel, did not show any significant differences in terms of annual girth increases without considering the treatments. And also it is observed that girth increments varied between 3.38 and 5.67 cm.year^{-1}.

Similar results were found while considering isodiametric growth of trees treated with Almephon (Table 3).

Several studies revealed that the rubber trees tapping commencement creates competition for assimilates and energy between radial vegetative growth and latex yield, resulting in a decrease of one of the parameters [15,25,28,30,31]. The results of the present study, in relation to the isodiametric vegetative growth, were all equivalent and indicated average significant increase of 4.37 cm.year^{-1} and 4.43 cm.year^{-1} with the trees treated by Ethrel and Almephon respectively. These results are in accordance with those of other study [17]. Therefore the isodiametric vegetative growth are independent of periods and conservation methods of the stimulant pastes to which the trees have been submitted.

Sensitivity to tapping panel dryness (TPD)

Sensitivity to tapping panel dryness namely, the average values of the rates of dry tapping cut length (LDC (%)) and trees with TPD (%) of the trees of clone GT 1 treated with Ethrel are also presented in Table 2. The results showed that except for the trees of treatment ETH-air-21 which showed a significantly high rate of dry tapping cut length (7.0 %); and all the trees had rates of tapping panel dryness statistically identical to each other. These rates varied from 0 to 2 %. As for the treated trees, except for ETH-air-21 which recorded a significantly highest rate of TPD (5 %), all trees treated with stimulant pastes prepared with Ethrel were free from trees with TPD regardless of the conservation time and conditioning mode of the stimulant paste prepared.

With reference to sensitivity to tapping panel dryness, the average values of the rate of dry tapping cut length (LDC (%)) and trees with TPD (%) of the trees of clone GT 1 treated with Almephon are shown in Table 3. The results indicate that all treatments with Almephon are statistically identical relating to the rates of dry tapping cut length of the trees of clone GT 1. These rates also varied from 0 to 2 % compared with trees treated with Ethrel. Considering the trees with TPD, no any cases have been observed in trees treated with stimulant pastes prepared and based on Almephon without considering the conservation time and the conditioning method.

The rates of dry tapping cut length, all treatments included, except for ETH-air-21, which showed a high rate of dry-cut length (7.0 %) were low, confirming that clone GT 1 expresses a moderate sensitivity to this syndrome [17]. The conservation period of the stimulant paste corresponding to treatment ETH-air-21 is neither the shortest nor the longest. This sensitivity to tapping panel dryness expressed by the trees submitted to treatment ETH-air-21 would be independent of periods and conservation methods of stimulant pastes. It would be due to edaphic factors[11].

300 **Table 2. Agronomical, physiological and TPD parameters of rubber trees treated with ETHREL**

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Treatments	Yield ($g.t^{-1}$)	Physiological parameters				Isodiametric increase ($cm.year^{-1}$)	LDC (%)	Trees with TPD (%)
		DRC (%)	Suc ($mmol.l^{-1}$)	Pi ($mmol.l^{-1}$)	R-SH ($mmol.l^{-1}$)			
ETH-air-Dd	894.74 cd	54.96 a	8.29 a	13.49 a	1.38 a	4.17 a	1.50 b	0.00 b
ETH-air-3	997.50 bcd	57.72 a	6.15 a	14.60 a	0.62 bc	4.45 a	1.00 b	0.00 b
ETH-air-7 (Control 1)	**927.21 bcd**	**52.85 a**	**7.37 a**	**13.97 a**	**0.23 cd**	**5.67 a**	**1.00 b**	**0.00 b**
ETH-air-14	934.37 bcd	59.84 a	6.79 a	16.45 a	0.61 bc	5.10 a	2.00 b	0.00 b
ETH-air-21	901.87 cd	54.50 a	9.04 a	17.24 a	0.27 cd	4.74 a	7.00 a	5.00 a
ETH-air-28	1015.00 abc	56.08 a	7.36 a	14.91 a	0.77 bc	4.34 a	0.50 b	0.00 b
ETH-air-42	900.00 cd	57.23 a	9.18 a	16.01 a	0.09 d	3.76 a	1.00 b	0.00 b
ETH-air-56	1058.44 ab	50.05 a	5.84 a	18.78 a	0.20 cd	4.63 a	0.00 b	0.00 b
ETH-cold-3	886.25 cd	56.02 a	6.11 a	16.84 a	0.60 bc	3.48 a	0.00 b	0.00 b
ETH-cold-7	887.82 cd	54.36 a	5.49 a	18.25 a	0.15 cd	4.80 a	0.00 b	0.00 b
ETH-cold-14	865.62 d	53.01 a	7.62 a	18.12 a	0.10 d	3.86 a	2.00 b	0.00 b
ETH-cold-21	685.62 e	55.96 a	9.25 a	18.07 a	0.23 cd	4.11 a	0.50 b	0.00 b
ETH-cold-28	1006.25 abcd	57.46 a	6.84 a	15.88a	0.19 cd	5.28 a	0.00 b	0.00 b
ETH-cold-42	975.79 bcd	49.30 a	7.37 a	17.50 a	0.16 cd	3.61 a	1.00 b	0.00 b
ETH-cold-56	1139.74 a	54.98 a	5.81 a	19.42 a	1.45 a	3.58 a	0.00 b	0.00 b
Means	**938.41**	**54.98**	**7.23**	**16.63**	**0.47**	**4.37**	**1.16**	**0.33**

302 *In each column, the values followed by the same letter are not significantly different (test of DUNCAN at 5 %)*
303 *g/t: gram per tree, DRC: dry rubber content, Suc: sucrose, iP: inorganic phosphorus, R-SH: thiol components, LDC: dry tapping cut length*
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313 **Table 3. Agronomical, physiological and TPD parameters of rubber trees treated with ALMEPHON**
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Treatments	Production ($g.t^{-1}$)	Physiological parameters				Isodiametric increase ($cm.year^{-1}$)	LDC (%)	Trees with TPD (%)
		DRC (%)	Suc ($mmol.l^{-1}$)	Pi ($mmol.l^{-1}$)	R-SH ($mmol.l^{-1}$)			
ALM-air Dd (Control 2)	**1153.94 bcd**	**53.87 a**	**8.12 a**	**14.60 a**	**0.70 abc**	**3.84 a**	**1.00 a**	**0.0 a**
ALM–air-3	1307.69 ab	56.52 a	7.18 a	18.16 a	1.64 a	3.58 a	1.00 a	0.0 a
ALM–air-7	1161.33 bcd	54.66 a	6.24 a	15.48 a	0.48 bc	5.15 a	1.00 a	0.0 a
ALM–air-14	1231.64 abc	55.80 a	5.23 a	16.54 a	0.66 abc	3.79 a	1.50 a	0.0 a
ALM–air-21	1205.62 bc	56.21 a	5.94 a	15.00 a	0.41 bc	4.92 a	1.00 a	0.0 a
ALM–air-28	1089.87 cd	53.76 a	8.24 a	17.99 a	0.20 bcd	3.61 a	0.00 a	0.0 a
ALM–air-42	1245.62 abc	56.82 a	5.16 b	17.15 a	0.25 bcd	3.88 a	0.00 a	0.0 a
ALM–air-56	1112.50 cd	57.33 a	8.95 a	19.35 a	0.12 d	4.93 a	0.00 a	0.0 a
ALM–cold-3	1382.91 a	55.88 a	6.51 a	15.74 a	1.02 a	5.22 a	0.00 a	0.0 a
ALM–cold-7	1162.98 bcd	58.91 a	6.67 a	13.06 a	0.40 bc	4.27 a	0.50 a	0.0 a
ALM–cold-14	1068.59 cd	54.27 a	5.31 a	17.85a	0.41 bc	4.03 a	2.00 a	0.0 a
ALM–cold-21	1097.50 cd	57.05 a	6.63 a	17.24 a	0.39 bc	5.19 a	1.50 a	0.0 a
ALM–cold-28	1085.33 cd	54.03 a	8.17 a	18.47 a	0.26 bcd	4.91 a	2.00 a	0.0 a
ALM–cold-42	1137.01 bcd	56.01 a	7.16 a	17.77 a	0.13 d	4.74 a	0.00 a	0.0 a
ALM–cold-56	984.61 d	56.37 a	7.70 a	16.71 a	0.11 d	4.46 a	1.00 a	0.0 a
Means	**1161.81**	**55.83**	**6.88**	**16.74**	**0.48**	**4.43**	**0.83**	**0**

In each column, the values followed by the same letter are not significantly different (test of DUNCAN at 5 %)

g/t: gram per tree, DRC: dry rubber content, Suc: sucrose, iP: inorganic phosphorus, R-SH: thiol components, LDC: dry tapping cut length

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3.2. Comparative effects of cold and ambient air on the efficiency of Ethrel and of Almephon

Tables 4 and 5 show the values of the agro physiological parameters and rates of dry-cut length of trees stimulated with the Ethrel and Almephon respectively. The analysis of these two tables showed that the two conditionally methods are statistically equivalent relative to agro physiological and health parameters. In fact, regardless of the conservation periods and the type of stimulant products, no significant difference between treatments had been observed, considering the values of agro physiological and health parameters.

The action of stimulant treatment is limited in time [32]. Thus, once prepared, the Ethrel-based stimulant paste could be kept for one week at ambient temperature without the stimulant losing its effect [33], while Almephon-based stimulant paste can be maintained one day without the stimulant losing its effect [33]. In this study, results are not in accordance with the foregoing. Indeed, the results of the different treatments in relation to their rubber yields were not influenced either by stimulant paste conditioning, or by the different conservation periods to which these treatments were submitted. The conservation periods and the conditioning of the stimulant paste would therefore not, in this work, be limiting factors for the efficiency of the stimulant products. Regarding the independence of the efficiency of stimulant products vis-a-vis the conservation periods and the conditioning of stimulant pastes, in the present results are in accordance with those of the works of [34] which showed that the amplitude of the response to the stimulation may be limited by climatic factors causing a water deficit and a reduction in the internal turgor pressure. Thus, it is recommended to avoid stimulating the trees during periods of frequent rain and during periods of low luminosities [9]. Indeed, frequent rain water washes the stimulant paste applied to the tree sometime after the rain fell, while low light conditions are unfavorable to the synthesis of photosynthetic assimilates a fraction of which is diverted to a regeneration metabolism of the latex [9,11]. To this end, stimulation in expressed *in fine* by a significant latex export but also by a strong activation of cellular metabolism and an increased consumption of carbohydrate matters resulting from the photosynthetic activity [27,32].

Furthermore, most commercial products used in the stimulation of rubber in Côte d'Ivoire, including Ethrel and Almephon have the same active ingredient chloro-2-ethylphosphonic acid (Ethephon), in very acid solution (pH = 1). Stable with very acid pH, Ethephon decomposes and releases ethylene if the pH of the solution becomes greater than 4.5, which is occurs actually inside the bark tissues: this account for the fact that the stimulant treatments leads to a release of ethylene in the bark tissue [32]. The stimulant product would be active and efficient if its center of action becomes less acid. Thus, on the biochemical and metabolic level, there are also significant and intrinsic modifications of rubber trees. The pH of the latex shows a tendency to alkalinization which has a significant impact on certain enzyme activities, especially on the glycolysis, which is accelerated sharply [35,36]. Therefore, the efficiency of the stimulant product would not be primarily dependent of the conservation period or conditioning of the stimulant paste but the pH of its environment for action: this could explain the results of the

different treatments in terms of their rubber yield. The results of the present study do not fundamentally undermine those of previous studies as the present study carried out for a short periods. To some extent, these results confirm the first but indicate that the period is probably longer. The present results are comforting because they provide a clear response to the concerns of national rubber industries to provide the means to control hormonal stimulation of the latex yield of smallholders. Technically, this activity can be managed from a stimulant preparation center in any way whatsoever and distribute to all the plantations under such center.

Table 4. Agronomical, physiological and TPD parameters of rubber trees treated with ETHREL in function of the conditioning of stimulant pastes

Treatments	Yield ($g.t^{-1}$)	Physiological parameters				Isodiametric increase ($cm.year^{-1}$)	LDC (%)	Trees with TPD (%)
		DRC (%)	Suc ($mmol.l^{-1}$)	Pi ($mmol.l^{-1}$)	R-SH ($mmol.l^{-1}$)			
ETH-air	953.64 a	55.4 a	7.50 a	15.68 a	0.12 a	4.55 a	1.75 a	0.62 a
ETH-cold	906.81 a	54.5 a	6.93 a	17.72 a	0.09 a	4.02 a	0.50 b	0.00 a

In each column, the values followed by the same letter are not significantly different (test of DUNCAN at 5 %)
g/t: gram per tree, DRC: dry rubber content, Suc: sucrose, iP: inorganic phosphorus, R-SH: thiol components, LDC: dry tapping cut length

Table 5. Agronomical, physiological and TPD parameters of rubber trees treated with ALMEPHON in function of the conditioning of stimulant pastes

Treatments	yield ($g.t^{-1}$)	Physiological parameters				Isodiametric increase ($cm.year^{-1}$)	LDC (%)	Trees with TPD (%)
		DRC (%)	Suc ($mmol.l^{-1}$)	Pi ($mmol.l^{-1}$)	R-SH ($mmol.l^{-1}$)			
ALM-air	1188.53 a	55.62 a	6.88 a	16.78 a	0.13 a	4.21 a	0.69 a	0.00 a
ALM-cold	1131.28 a	56.07 a	6.89 a	16.69 a	0.10 a	4.43 a	1.00 a	0.00 a

In each column, the values followed by the same letter are not significantly different (test of DUNCAN at 5 %)
g/t: gram per tree, DRC: dry rubber content, Suc: sucrose, iP: inorganic phosphorus, R-SH: thiol components, LDC: dry tapping cut length

3.3. Cost of stimulation of a hectare plantation in downward tapping about stimulant paste based on Ethrel and Almephon and determination of the best stimulant

The report $CostHa_{StimAlph}/CostHa_{StimET}$ gives 5.843, which signifies that the stimulant product ready for use, especially the Almephon is at least 5 times more expensive than based on the Ethrel (Table 6).

In the present experimental setting, all the other parameters being equal, the ratio $CostHa_{StimAlm}/CostHa_{StimET}$ giving 5.843, enables to assert that hormonal stimulation with the stimulant product ready for use. However Almephon, at least 5 times more expensive than the Ethrel-based one is significantly less expensive than the latter. In addition, some works (personnal communication with Obouayeba) have shown that the bark stimulated with an Ethrel-based stimulant paste is softer than the stimulant product of ELS (ready-to-use). These two results in favor of the Ethrel-based stimulant paste, reinforces in us the idea that the best stimulant product is Ethrel.

Table 6. Cost of Stimulation of hectare plantation in downward tapping about stimulant paste based on EHTREL and ALMEPHON

Stimulant product	Cost/Ha (USD)
Ethrel	0.599
Almephon	3.500

4. CONCLUSION

The conservation of stimulant pastes prepared with stimulant products marketed under the names of Ethrel and Almephon enabled to note that the efficiency of the stimulant has been influenced neither by the conditioning of stimulant pastes previously prepared, nor by the different conservation periods to which these pastes were submitted. The response to stimulation could be due either to climate factors or the tendency to latex alkalinization which has a particularly significant impact on latex glycolysis. Conservation recommendations for one day or one week of the stimulant pastes prepared previously are now obsolete. The stimulant pastes prepared already can thus be conserved for more than one week, specifically up to 56 days, at room temperature as well as in cold, without the stimulant losing its efficiency.
The ratio $CostHa_{StimAlm}/CostHa_{StimET}$ worth 5.843, is an illustration of the fact that the stimulant product ready to use, Almephon, is more expensive than the Ethrel-based one. It is therefore the best stimulant for natural rubber yield.

REFERENCES

1. Jacob JL, Serres E, Prevot JC, Lacrotte R, Clement-Vidal A, Eschbach JM D'auzac J. Mise au point du diagnostic latex. *Agritrop*. 1988;(12):97-118.
2. Obouayeba S. Exploitation de l'hévéa en milieu non industriel. *Document de vulgarisation* validé, utilisé dans le cadre du projet "Hévéa 5 bis : Recherche-Formation pour le développement des petites et moyennes plantations d'hévéa en Côte d'Ivoire"; 1995.
3. Obouayeba S, Soumahin EF, Dobo M, Lacrotte R, Gabla O, Doumbia A. Agronomic performance of clone IRCA 111 of *Hevea brasiliensis* under different frequencies of tapping and stimulation in South-West Côte d'Ivoire. *Journal of Rubber Research*. 2009;12(2):93-102.
4. Soumahin E.F. Optimisation des systèmes d'exploitation en hévéaculture par la réduction des intensités de saignée. Thèse unique de doctorat, UFR Biosciences, Université Cocody, Côte d'Ivoire; 2010.
5. Anonymous. La stimulation de l'hévéa. *Fiche technique* n° E1. IRCA-CIRAD, Paris, France; 1988.
6. Obouayeba S. Estimation de la quantité de pâte stimulante appliquée sur hévéa en fonction de leur circonférence au sud-est de la Côte d'Ivoire. *Agronomie africaine*. 1992;5(1):26-32.
7. Obouayeba S, Boa D, Keli ZJ. Adéquation entre quantité de pâte stimulante et production de caoutchouc d'*Hevea brasiliensis* dans le sud-est de la Côte d'Ivoire. *Tropicultura*, 1996;14:54-58.
8. Traoré MS. Effets de différentes fréquences annuelles de stimulation éthylénique sur les paramètres agrophysiologiques des clones d'*Hevea brasiliensis* Müll. Arg. (*Euphorbiaceae*), PB 235, PB 260, GT 1 et PB 217 cultivés au sud-est de la Côte d'Ivoire. Thèse de doctorat d'Université, Université Félix Houphouët-Boigny, Abidjan, Côte d'Ivoire; 2014.
9. Compagnon P. Le caoutchouc naturel. Coste R. Ed. G. P. *Maisonneuve et Larose*, Paris; 1986.
10. Obouayeba S, Boko AMC, Soumahin EF, Elabo AAE, Dea GB, Kouamé C, Kéli ZJ. *Hevea brasiliensis*, composante de systèmes de production rurale stables : synthèse de quarante ans de travaux, *Rapport de synthèse-Programme Hévéa*, Centre National de Recherche Agronomique (CNRA); 2013.
11. Traoré MS, Dick EA, Elabo AEA, Soumahin EF, Assi EGM, Atsin OGJ, Allé YJ, N'guessan AEB, Kouamé C, Obouayeba S. Effets de différentes fréquences annuelles de stimulation éthylénique sur les paramètres agrophysiologiques et de sensibilité à l'encoche sèche *d'Hevea brasiliensis* au sud-est de la Côte d'Ivoire : cas des clones PB 235 et PB 260 de la classe d'activité métabolique rapide. *International Journal of Biological and Chemical Sciences*, 2014;8(3):956-974.
12. Okoma KM, Dian K, Obouayeba S, Elabo AAE, N'guetta ASP. Seasonal variation of tapping panel dryness expression in rubber tree *HEVEA BRASILIENSIS* Müll. Arg. In Côte d'Ivoire. *Agriculture and Biology Journal of North America*, 2011;2(3):559-569.

13. Gohet E, Prevot JC, Eschbach JM, Clement A, Jacob JL. Clone croissance et stimulation, facteurs de la production de latex. *Plantations, recherche, développement*, 1996;3(1):30-38.
14. Gohet E, Lacrote R, Obouayeba S, Commere J. Tapping systems recommended in West Africa. *Proceedings Rubber Research Institute Malaysia Rubber Growers' Conference,* 1991; 1998.
15. Obouayeba S, Boa D, Ake S, Lacrotte R. Influence of age and girth at opening on growth and productivity of Hevea. *Indiana Journal of Natural Rubber Research.* 2002;15(1):66-71.
16. Obouayeba S. Contribution à la détermination de la maturité physiologique de l'écorce pour la mise en saignée d'*Hevea brasiliensis* Muell. Arg. (*Euphorbiaceae*) : Normes d'ouverture. Thèse de doctorat d'Université, Université de Cocody, Abidjan, Côte d'Ivoire; 2005.
17. Anonymous. Recueil de fiches clones Hévéa. CIRAD-CP, 12 Square Pétrarque-75116 Paris, CSR, St Gely du Fesc. Juin 1993.
18. Eschbach JM, Tonnelier M. Influence de la méthode de la stimulation, de la concentration du stimulant et de la fréquence de son application sur la production du clone GT 1 en Côte d'Ivoire. In*:* C.R. Coll. Expl. Physiol. Amél. Hévéa, IRCA-CIRAD, ed*.,* Montpellier, France, 295 – 306. *Hevea brasiliensis*. *Physiologie Végétale,* 1984;22:295-304.
19. Ashwell G. Colorimetric analysis of sugar. Methods Enzymol., 1957;3:73-105.
20. Tausssky HH, Shorr E. A micro colorimetric method for the determination of inorganic phosphorus. *Journal of Biological Chemistry,* 1953;202:625-685.
21. Boyne AF, Ellman GI. A methodology for analysis of tissue sulphydryl components. *Analytical Biochemistry*., 1972;46:639-653.
22. Eschbach JM, Roussel D, Van De Sype H, Jacob JL, D'auzac J. Relationships between yield and clonal physiological characteristics of latex from *Hevea brasiliensis. Physiologie Végétale,* 1984;22:273-295.
23. Prévot JC, Jacob JL, Lacrotte R, Vidal A, Serres E, Eschbach JM, Gigault J. Physiological parameters of latex from *Hevea brasiliensis*. Their use in the study of the laticiferous system. Typologie of functioning production metabolism. Effects of stimulation. In*: International Meeting of Physiology Exploitation, International Rubber Research and Development Board,* Hainan, Chine; 1986.
24. Serre E, Clement A, Prevot JC, Jacob JL, Commere J, Lacrotte R, Eschbach JM. Clonal typology of laticiferous producing vessels in *Hevea brasiliensis*. In *CR*. *International Rubber Research and Development Board*;7:79-84.
25. Jacob, J.L., Prevot, J.C., Lacrotte, R. And Eschbach, J.M. 1995. Le diagnostic latex. *Plantations, recherche, développement,* 1988;2(2):34-37.
26. Milford GGJ, Paardekooper EC, Ho CY. Latex vessels plugging, its importance to yield and clonal behaviour. *Journal of Rubber Research Institute of Malaya,* 1969;21:274-282.

27. Lacrotte R. Etude des relations entre la teneur en sucre du latex et la production : approches des mécanismes du changement en saccharose des laticifères d'*Hevea brasiliensis* Muell. Arg. Thèse de Doctorat d'Université, Université des Sciences et techniques du Languedoc, Montpellier II. 2010;266.
28. Gohet E. La production de latex par Hevea brasiliensis. Relation avec la croissance. Influence de différents facteurs : Origine clonale, stimulation hormonale, réserves hydrocarbonées. Thèse Doct. 3e cycle. Université des Sciences & Technique de Languédoc, Ponpellier II, France. 1996;343.
29. Jacob JL, Prévôt JC, Lacote R, Gohet E, Clément A, Gallois R, Joet T, Pujade-Renaud V, D'auzac J. Les mécanismes biologiques de la production de caoutchouc par *Hevea brasiliensis*. Communication de l'IRRDB 1997 Workshop, Ho Chi Minh-Ville, Viêt Nam, du 14 au 15 octobre 1997. 1998; 13.
30. Paardekooper EC. Exploitation of rubber tree. In rubber eds. C. C. Webster et W. J. Baulkwill. 1989;398-414.
31. Obouayeba S, Boa D. Fréquence et repos annuel de saignée d'*Hevea brasiliensis*, clone PB 235, dans le sud-est de la Côte d'Ivoire. *Cahiers Agricultures*. 1993;2(6):387-393.
32. Tonnelier M, Primot L. La stimulation de la production en latex de l'hévéa. PG/MM, IRCA. 1979;8p.
33. Anonymous. ALM Afrique de l'ouest. Distributeur du produit stimulant commercialisé sous le nom d'Almephon. N° d'homologation : 02 05920 Pp. 18, rue du Docteur Blanchard, 01 BP 3823 Abidjan 01. 2010
34. Pakianathan SW. Some factors affecting yield response to stimulation with chloroethyl phosphonic acid. *Journal of Rubber Research of Malaysia*, 1977;25(1):30-60.
35. Tupy J. The regulation of invertase activity in the latex of *Hevea brasiliensis*. The effects of growth regulators bark wounding and latex tapping. *Journal of Experimental Botany,* 1973;24:516-524.
36. Tupy J, Primot L. Control of carbohydrate metabolism by ethylene in latex vessels of *Hevea brasiliensis* MÜLL. Arg. In relation to rubber production. *Biologia Plantarum,* 1976;18:373-384.