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Efficiency of Banzai Biostimulant on the cherries production in healthy and cocoa trees affected by swollen shoot virus: A case study at Petit-Bondoukou Côte d'Ivoire

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ABSTRACT

The present study is to evaluate the efficiency of the Biostimulant "Banzai" on the production of cherries from healthy and diseased trees in a context of pressure from Swollen shoot cocoa disease in Petit-Bondoukou in the department of Soubré. The specific aim is to compare the effect of the single and double doses of Banzai on the production of cherries from healthy and diseased trees. The experimental design consists of a single block of four plots marked T0 (Control), T1 (a standard rate Banzai application without fertilizer), T2 (a double rate Banzai application without fertilizer) and T3 (a standard rate Banzai application with fertilizer). Each plot contains ten healthy trees and ten trees with swollen shoot disease. This device was repeated four times at the study site. Banzai was applied for four months and observations were made for six consecutive months on the number of cherries produced. The results of the Kruskal-Wallis test showed that the treated plots produced significantly more cherries than the control plots in both healthy and diseased trees at any dose. In addition, in healthy trees, the Kruskal-Wallis test showed that the single dose of Banzai without fertilizer (T1) significantly produced more cherries than other treatments. While in diseased trees, the Kruskal-Wallis test showed that the double dose of Banzai without fertilizer (T2) significantly produced more cherries than other treatments.

Keywords : *Cocoa trees, Biostimulant, Banzaï, CSSV, Côte d'Ivoire.*

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1. INTRODUCTION

Côte d'Ivoire is a tropical country, located in West Africa, whose economy is based on agriculture. Cocoa occupies a prominent place in the socio-economic fabric of Côte d'Ivoire, which produces about 1,650,000 tons annually, or 32% of the world's supply, making it the world's leading producer [1]. Cocoa cultivation generates 35% of the country's total export earnings and accounts for 15% of GDP (Gross Domestic Product) and 20% of tax revenues [2]. However, cocoa cultivation is affected by several constraints, including the existence of pests and diseases such as the Swollen Shoot, which has a negative influence on cocoa bean yields and quality. Indeed, Swollen Shoot is a viral disease of cocoa caused by a virus (CSSV: Cocoa Swollen Shoot Virus) of the genus *Badnavirus* of the *Caulimoviridea* family. The first form of the virus was detected in Ghana in 1936 on 15-year-old cocoa trees. Swollen Shoot was subsequently reported in other cocoa producing countries such as Côte d'Ivoire in 1943, Nigeria in 1944 and Togo in 1955. The natural transmission of the swollen shoot virus from a diseased plant to a healthy plant is through the mealybugs [3] of the *Pseudococcidae* family and in a semi-persistent manner. Once in the plant, CSSV causes physiological and morphological disturbances. The virus causes swelling followed by abortion of the taproot, abortion of the apexes of the twigs and inhibition of terminal meristem functioning followed by a slowing of tree growth [4]. The reproductive organs (pollen grains and eggs) become abnormal and sterile. Sick pods or cherries are abnormal and the affected tree has embossed leaves [4]. The virus causes more or less intense discoloration of the blade depending on its severity. The virus causes low flowering, low fruit set, very pronounced physiological drying and a rapid decrease in the number of cherries or pods. All these disruptions result in a rapid drop in production, followed by the death of attacked trees after at least three years of infection [5]. Post-harvest losses caused by this disease range from 30 to 40% [6; 7]. There is currently no treatment for this disease and the only methods adopted are the uprooting of diseased cocoa trees and replanting with resistant or tolerant varieties. The control method, which recommends that grubbing-up, is difficult for producers to accept because the areas destroyed are too large and have a negative impact on the socio-economic situation of producers. Faced with this situation, alternative solutions must be found to the uprooting of sick cocoa trees. Indeed, in recent years, a category of products and substances has developed within the agricultural input market "Biostimulants". Biostimulants provide solutions in terms of fertilization and often crop protection. They contain substances or microorganisms that stimulate natural processes to increase absorption, nutrient efficiency and tolerance to abiotic and biotic stresses. Biostimulants also increase crop quality and protect the health of the soil, environment and people [8]. This is why this study, whose main aim is to improve cocoa tree productivity in the context of Swollen Shoot disease with Banzai biostimulant, is justified. More specifically, the effect of Banzai Biostimulant on cherry production in both diseased and apparently healthy trees should be evaluated. It is also to assess the cumulative effect of Banzai biostimulant and fertilizer on the production of cherries.

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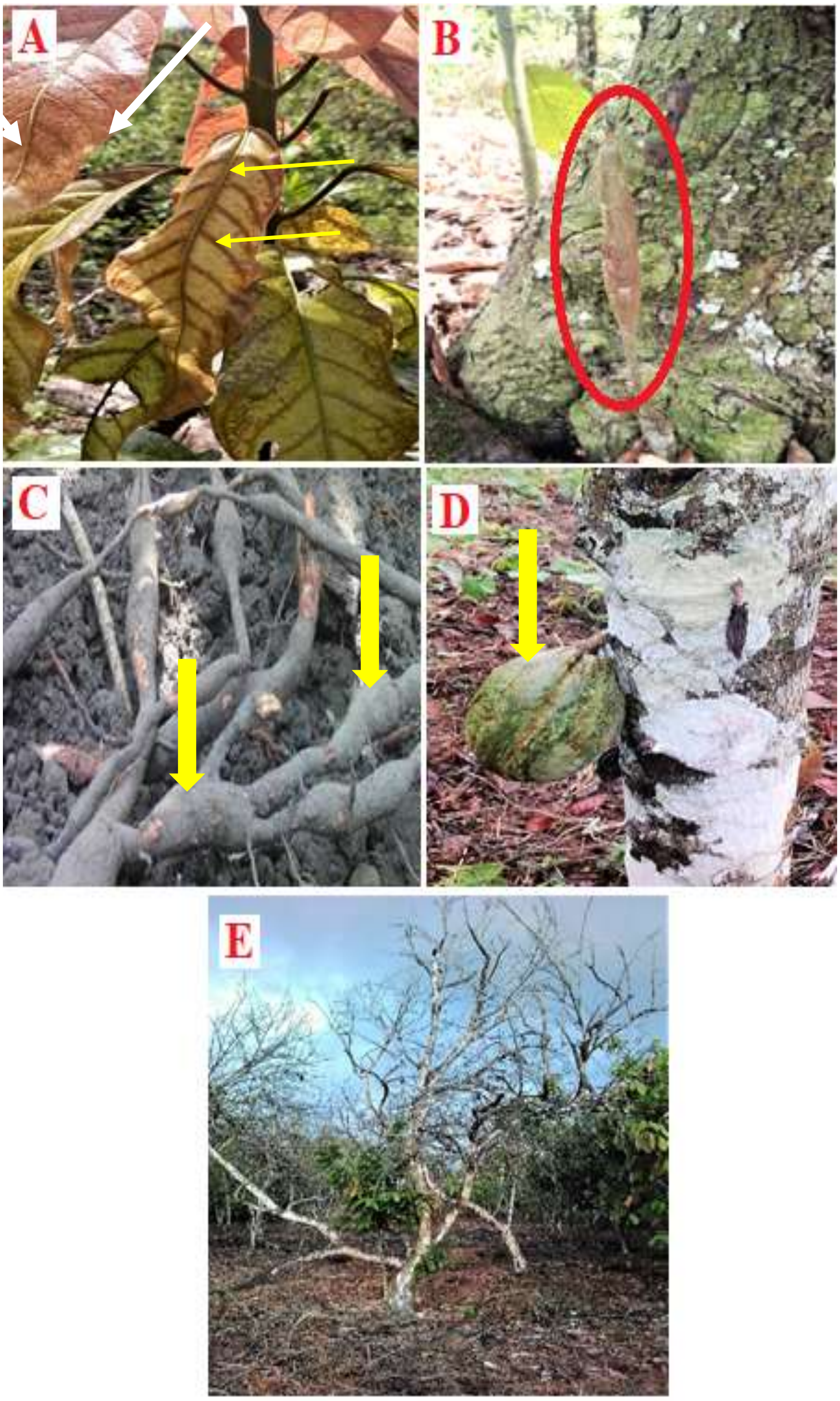

Figure 1: Symptoms of Swollen Shoot disease on different parts of the cocoa tree. (A) redness along the veins on young leaves, (B) apical swelling on orthotropic shoots, (C) swelling on roots, (D) stunting of the pod, thus reducing the size and quality of the beans, (E) damage of Swollen Shoot Virus in peasant plantations (Picture taken by Oro in 2017).

2. MATERIAL AND METHOD

2.1. Study area

This study was carried out in Petit-Bondoukou (Figure 2) located in Méagui (department of Soubré) in southwestern Côte d'Ivoire, the main cocoa production area with about 300 000 tons, or nearly 20% of national production during the 2013-2014 season [9]. Petit-Bondoukou is a village located 51 kilometers from Soubré. The study site is characterized by a humid tropical climate with an average rainfall of 1485 mm per year, and an average temperature of 25.8°C per year. Vegetation and soil are characterized by dense, moist forests and deep, permeable and well-drained soil that allows for human activity related to agriculture [10]. The soil

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of Soubré is suitable for all types of food and industrial crops. This vegetation is now giving way to shreds of forests and huge plantations of traditional or industrial perennial crops [9].

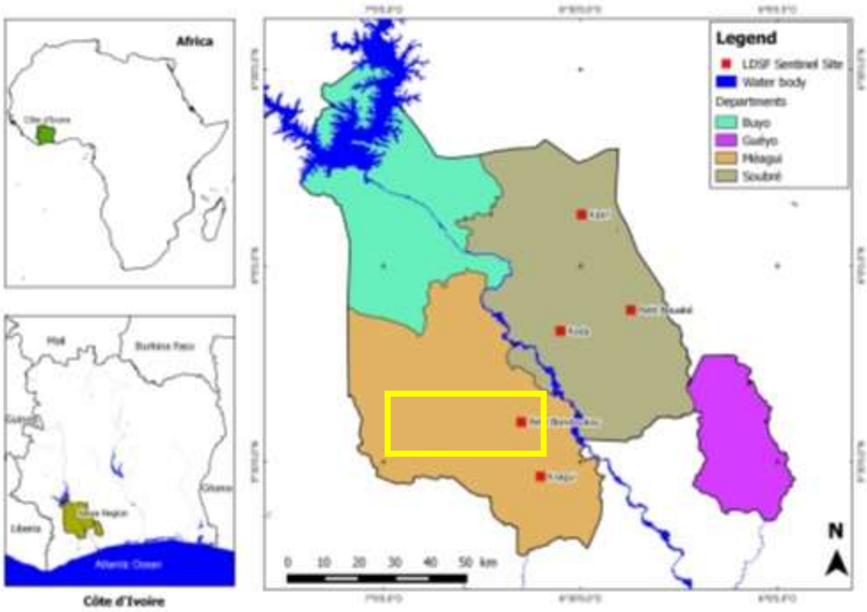

Figure 2: Map of the region of Nawa highlighting Petit-Bondoukou site [11].

2.2. Experimental Device

The experimental set-up is a simple block consisting of four (04) sub-plots marked T0, T1, T2 and T3. Each experimental plot is composed of twenty (20) cocoa trees, ten (10) of which are apparently healthy and 10 diseased. T0 represents the control without Banzai application and without fertilizer. T1 is the plot that represents a standard rate application of Banzai (800 ml/ha) and without fertilizer. T2 is a plot representing a double-dose Banzai application without fertilizer and T3 represents a normal-dose Banzai application with fertilizer. This device is repeated 4 times on Petit-Bondoukou site.

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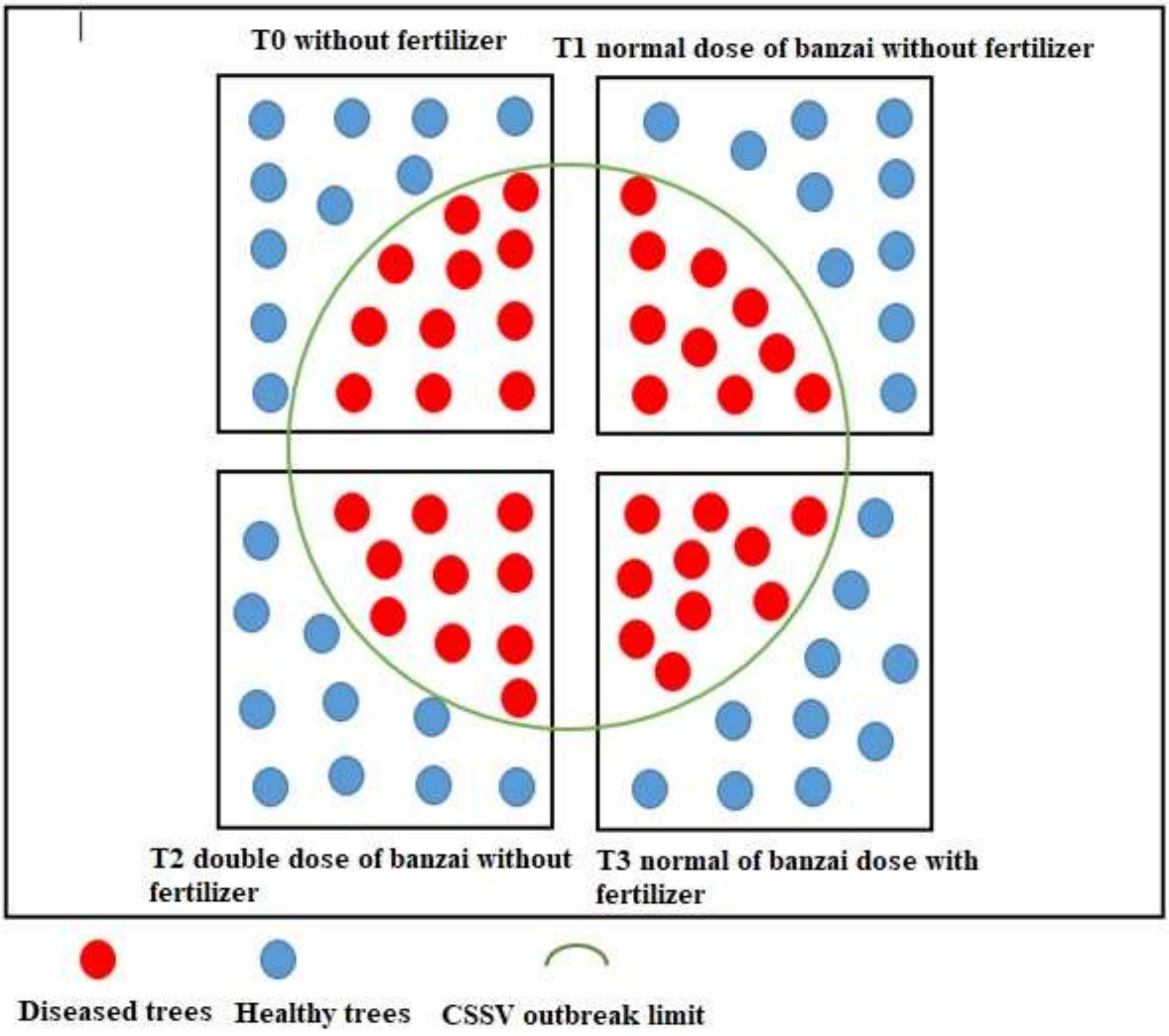

Figure 3: Experimental device with the different elementary plots around the CSSV infection site

2.3. Conduct of the test

The implementation of the test began with a prospective survey that consisted first of researching and selecting the experimental sites, then identifying the appropriate plots for the study. The selected sites were characterized by cocoa plots with young outbreaks containing trees affected by swollen shoot disease, but still in production and apparently healthy trees. Each elementary plot has an area of about 300 m^2 and contains 20 trees, including 10 diseased trees and 10 apparently healthy trees. These plots were delimited in the direction of progression of the infection site. The plant material consisted of all the test cocoa trees that were selected from the already delimited elementary plots. The technical equipment used to set up the test consisted of red paint cans and labels to mark the test trees, multi-colored ribbons to mark the boundaries of each block and each elementary plot, an atomizer for spreading the Banzai biostimulant, a decameter to measure the block boundaries and plot boundaries, and a camera for taking pictures. The treatment products consisted of SUPERCAO fertilizer formulated 0-23-19 + 10CaO + 5MgO + 6.5S +0.7 Zn which was applied to some elementary plots, then Banzai biostimulant with an indicator dose of 800 ml/ha which was applied to all elementary plots except the control plots.

Indeed, SUPERCAO fertilizer was applied twice, i.e. once when the device was installed in August 2017 and the other time in September 2017 (only on the elementary plot T3) at 200g per tree. Fertilizer application was carried out within a 50 cm radius around each test tree and was covered with soil and dead leaves. On the other hand, Banzai biostimulant was applied at the 800 ml/ha rate, considering that one hectare of cocoa trees contains 1333 cocoa trees. It is from this initial dose that the dose for 20 test cocoa trees (12 ml for 3 liters of water) and the double dose at 24 ml for 3 liters of water were deducted. Banzai biostimulant was applied to both sides of the cocoa tree from the foliage through the trunk to the neck of the tree. Thus, the normal Banzai rate was applied to the test trees in plots T1 and T3, while the double rate was applied to the test trees in plot T2.

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2.4. Observations

Observations in general were made each month for six (06) months (August to January) after the Banzai product was applied. These observations were made on the number of cherries for both the lot of diseased trees and the lot of apparently healthy trees.

2.5. Data collection

The total number of cherries was counted on the trunk of each test tree in a height range between 0 and 2.35 meters from the ground. Cherries are immature pods of less than 6 cm in length. The number of cherries was counted cumulatively between two observation periods.

2.6. Data Analysis

The production data were first subjected to a descriptive analysis in Excel 2013 and then to a comparative analysis using IBM SPSS Statistics version 20 software. The descriptive analysis of the data relating to cherries consisted first of all in aggregating the number of cherries produced per treatment and for a batch of test trees (Healthy/Diseased) during the six months of observation. Then histograms by treatment were made to compare the evolution of cherries production according to the observation period and for each batch of test trees. Finally, a comparison of the histograms was carried out to evaluate the production of cherries between the treatments of the lot of healthy trees and the lot of diseased trees. The comparative analysis made it possible to statistically evaluate the impact of Banzai biostimulant on the production of cherries. The analyses consisted first of comparing the minimums, maximums, sums, average and standard deviations between the different treatments as a whole, and then according to the health status of the tree. Then boxplots were made to visualize the distribution of the cherries produced by treatment. The hypotheses based on the boxplots were confirmed or disproved by the Kruskal-Wallis statistical test. When a difference was found from this test, a classification of treatments according to their efficacy was performed to evaluate the different doses of Banzai biostimulant.

3. RESULTS

3.1. Effect of Banzai Biostimulant on the production of cherries of apparently healthy trees

Figure 4 shows the production of cherries during the six months of observation. In the first month of observation (August to September), the production of cherries is similar (on average 261 cherries produced) for all treatments. From October to December, the production of T1, T2 and T3 treatments is more important than the production of the T0 control. The production of cherries in January is very low for all treatments. Overall, the T1 treatment produced the most cherries (721 cherries) than the other treatments with an average difference of 60 cherries. The T0 control was less productive with 287 cherries (**Figure 13**). **Table 1** presents the minimum, maximum, sum, average and standard deviation of the cherries produced per test tree according to the treatment. The small standard deviations obtained reflect the standardization of the production of apparently healthy trees. The graphical representation of these data (**Figure 5**) by boxplots showed two groups based on the medians: the trend of the control T0 with a median that is below 10 cherries and the trend of treatments (T1, T2, T3) with a median close to 20 produced cherries. The Kruskal-Wallis statistical test presented in Table 2 showed a significant difference ($p=0.001<0.05$) between the mean production of cherries per treatment. This test grouped the treatments into three classes: A, B and C top-down (**Table 3**). The T1 treatment (single dose without fertilizer) was more effective in the production of cherries than the other T2 (double dose without fertilizer) and T3 (single dose with fertilizer) treatments.

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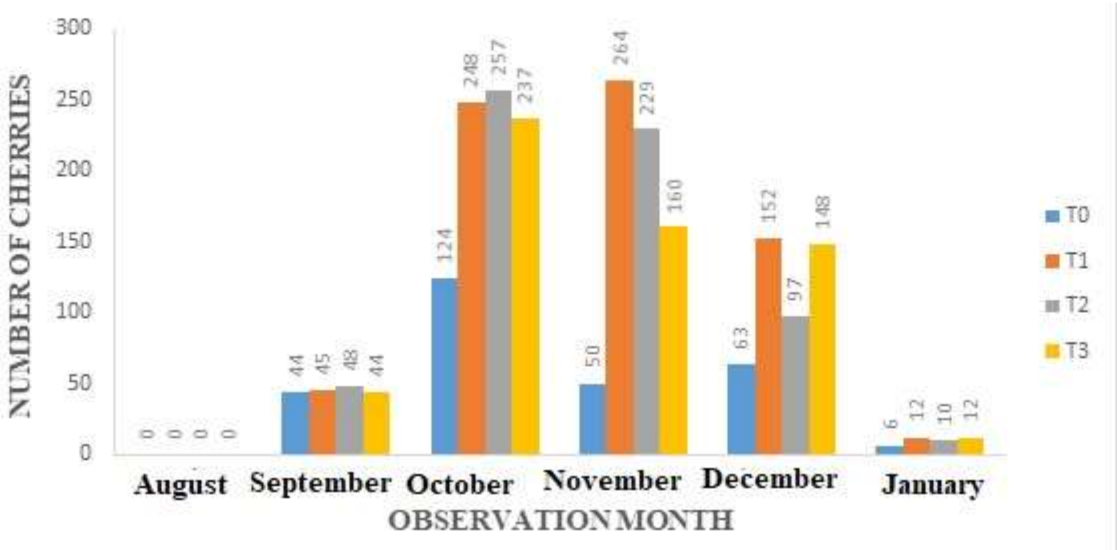

Figure 4: Evolution of cherries production on apparently healthy trees

Table 1 : Average production of cherries per tree according to treatment

	Accumulation of cherries from August to January				
Treatments	**Minimum**	**Maximum**	**Number**	**Average**	**Standard deviation**
T0	0	36	287	7.18	7.61
T1	0	64	721	18.03	16.03
T2	0	51	641	16.03	13.63
T3	0	58	601	15.03	14.56

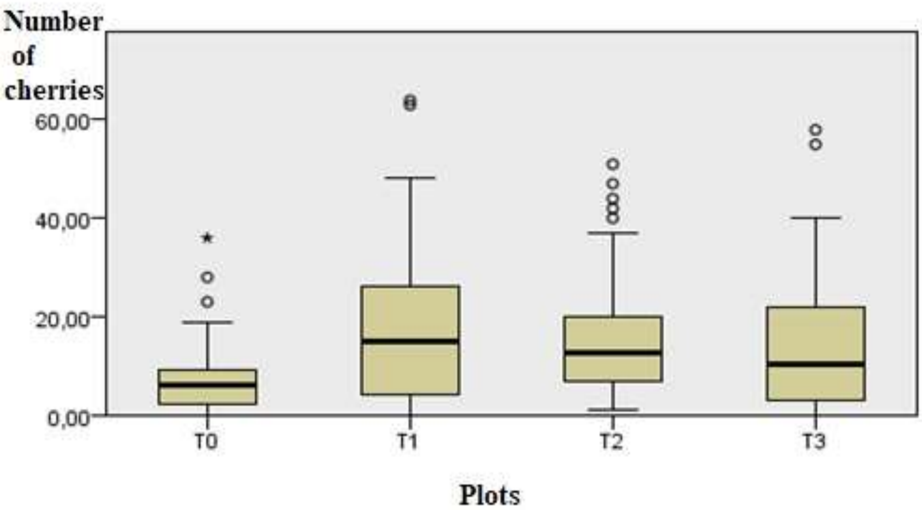

Figure 5: Boxplots representing the production of cherries by treatment

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Table 2 : Results of the Kruskal-Wallis test for the comparison of average production of cherries per treatment

Récapitulatif du test d'hypothèse

	Hypothèse nulle	Test	Sig.	Décision
1	La distribution de Cumul chérelles m1-m6 est identique sur les catégories de parcelles.	Test de Kruskal-Wallis à échantillons indépendants	,001	Rejeter l' hypothèse nulle.

Les significations asymptotiques sont affichées. Le niveau de signification est ,05.

Table 3 : Result of the classification of treatments according to the averages of the cherries produced

Treatments	Average	Classification
T1	18.03	A
T2	16.03	B
T3	15.03	B
T0	7.18	C

3.2. Effect of Banzai Biostimulant on the production of cherries from diseased trees

During the six months of observation, trees from the T1, T2 and T3 treatments produced more cherries than the control plot with peaks of 118 cherries respectively; 153 cherries and 90 cherries compared to 38 cherries for the control in October (Figure 6). At the end of the experiment, the T2 treatment represented by the double dose without fertilizer was more productive with 302 cherries than the other treatments with an average difference of 76 cherries. The T0 control produced fewer cherries than other treatments. Overall, the Banzai product had a positive impact on the production of cherries from diseased trees and exceptionally, the double dose of Banzai was more remarkable in the production of cherries. **Table 4** presents the minimum, maximum, sum, mean and standard deviation of the cherries produced per tree according to the treatment. The standard deviations are higher than the corresponding averages. This means that the production of cherries in the context of the presence of swollen shoot disease is not standardized from one tree to another. In terms of statistical analysis, the one-way ANOVA test revealed that there is no significant difference between the average production of cherries from treatment including the control (**Table 5**). Although there was no significant difference between the treatments, the T2 (double dose without fertilizer) and T1 (single dose without fertilizer) treatments were the most numerically productive compared to the T3 and T0 control treatments.

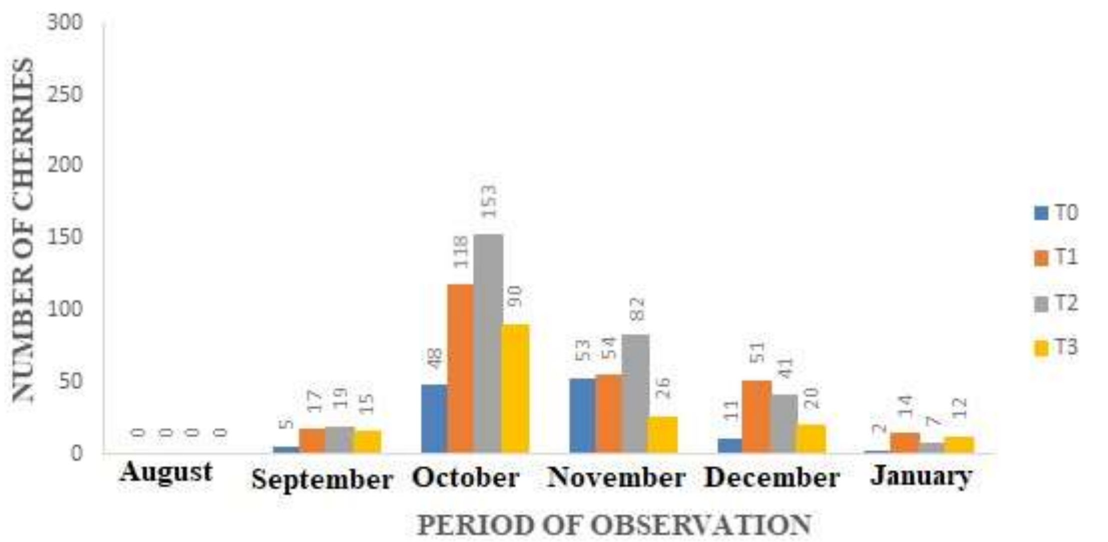

Figure 6: Evolution of cherries production by treatment and month of observation in diseased trees

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Table 4 : Average production of cherries by tree according to treatment on diseased trees

	Accumulation of cherries produced				
Treatments	**Minimum**	**Maximum**	**Number**	**Average**	**Standard deviation**
T0	0	29	119	2,98	5,20
T1	0	68	254	6,35	11,28
T2	0	49	302	7,55	9,42
T3	0	27	151	4,08	6,74

Tableau 5 : Result of the one-way analysis of variance for the comparison of the number of cherries produced per treatment

	One-way ANOVA				
	Number of squares	**ddl**	**Average of squares**	**F**	**Standard deviation**
Inter-groups	299.638	2	149.819	2.036	**0.134**
Intra-groups	11553.963	157	73.592		
Total	11853.600	159			

3.3. Comparison of Banzai's efficiency on the production of cherries between healthy and diseased trees

Overall, apparently healthy trees have the highest cherries production (2250) compared to diseased trees (826). This production of apparently healthy trees represents 73% of the cherries production in the whole system compared to 27% for diseased trees (Figure 7).

The Banzai effect is more remarkable in terms of cherries production on apparently healthy trees than on diseased trees.

Table 6 presents the minimum, maximum, sum, mean and standard deviation of the cherries produced per tree according to treatment and health status. The graphical representation of these data (**Figure 8**) by boxplots showed several groups that emerged on the basis of the medians. The Kruskal-Wallis statistical test showed a highly significant difference ($p=0.0001<0.05$) between the average production of the treatments (**Table 7**). The test groups these averages into 4 classes (A, B, BC and C) (**Table 8**). Healthy test trees that received the single dose of Banzai without fertilizer (T1) produced more cherries compared to all trees in other treatments, considering both healthy and diseased trees. Therefore, all apparently healthy test trees treated with Banzai were more productive than all diseased test trees treated.

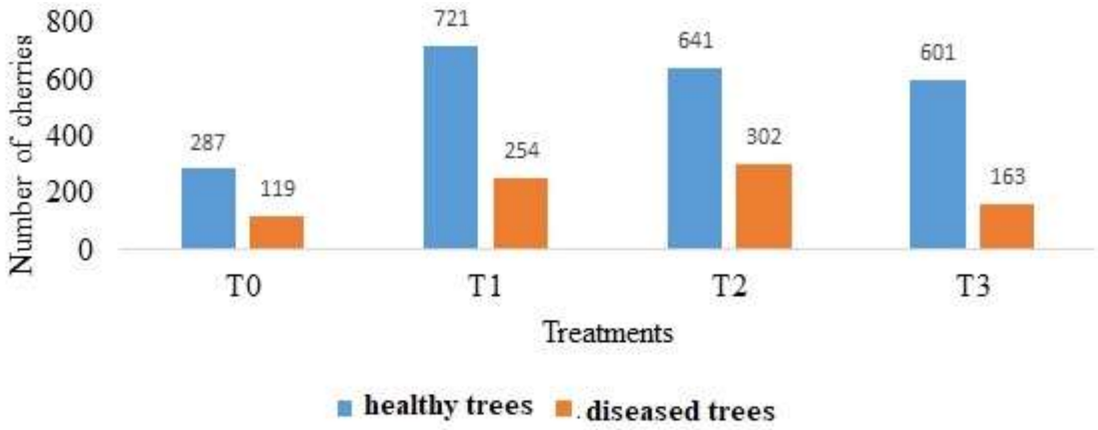

Figure 7: Comparison histograms of total cherries production of healthy and diseased trees

Table 6: Total and average production per treatment according to the status of the tree

	Accumulation of cherries produced

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Treatments	Minimum	Maximum	Number	Average	Standard deviation
T0_diseased	0	29	119	2.98	5.20
T1_diseased	0	68	254	6.35	11.28
T2_diseased	0	49	302	7.55	9.42
T3_diseased	0	27	151	4.08	6.74
T0_healthy	0	36	287	7.18	7.61
T1_healthy	0	64	721	18.03	16.03
T2_healthy	1	51	641	16.03	13.63
T3_healthy	0	58	601	15.03	14.56

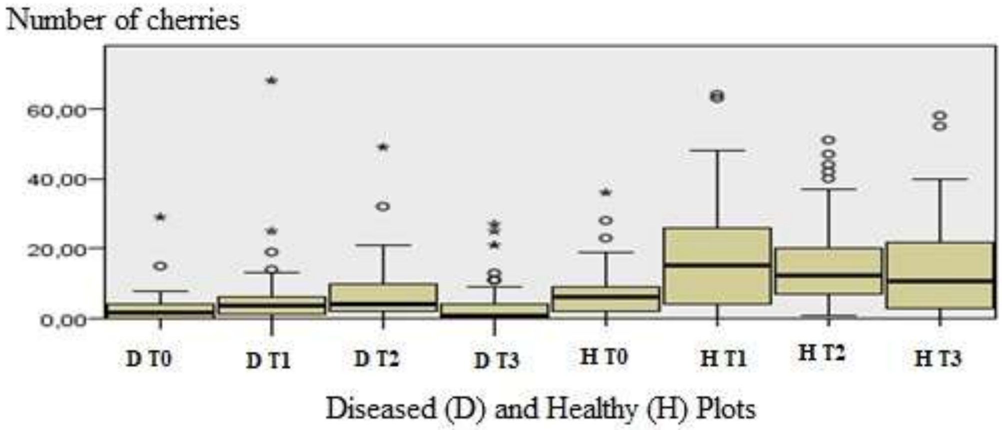

Figure 8: Boxplots representing the production of cherries by treatment according to health status.

Table 7 : Results of the Kruskal-Wallis test for the comparison of Average production of cherries per treatment according to health status.

Récapitulatif du test d'hypothèse

	Hypothèse nulle	Test	Sig.	Décision
1	La distribution de Cumul chérelle m1-m6 est identique sur les catégories de parcelles_Statut.	Test de Kruskal-Wallis à échantillons indépendants	,000	Rejeter l'hypothèse nulle.

Les significations asymptotiques sont affichées. Le niveau de significa est ,05.

Table 8 : Classification of treatments according to cherries production by health status

Treatments	Average	Classification
HealthyT1	18.03	A
HealthyT2	16.03	B
HealthyT3	15.03	B
DiseasedT2	7.55	BC
HealthyT0	7.18	BC
DiseasedT1	6.35	BC
DiseasedT3	4.08	C
DiseasedT0	2.98	C

4. DISCUSSION

The results of the Kruskal-Wallis statistical test show that the Banzai biostimulant treated plots produced more cherries than the control plots in both healthy and diseased trees. These results are consistent with those obtained by Stefano Pedrazzi and al. [12]. Indeed, these researchers obtained by combining the biostimulant based on *Pseudomonas fluorescens*, the biostimulant based on

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Trichoderma harzianum and the biostimulant based on *Glomus intraradices* a significant increase in tomato yield of more than 40% and a reduction in variability in weight per fruit.

In healthy trees, the results of the Kruskal-Wallis statistical test showed that the plots treated with the single dose without fertilizer (T1) were significantly more productive in terms of cherries than the plots of other treatments. This shows that Banzai biostimulant has provided the essential nutrients that enable trees to increase their production. This result is in harmony with the claims listed on the Banzai biostimulant data sheet.

In diseased trees, the results of the Kruskal-Wallis statistical test showed that the production of cherries from plots that were treated with the double dose of Banzai and without fertilizer (T2) is significantly similar to those that were treated with the single dose of Banzai with fertilizer (T3). On the other hand, the productions resulting from the T2 and T3 treatments are significantly different from those of the T1 treatment (standard Banzai dose without fertilizer) and those of the T0 control, both of which were less productive. Indeed, when a tree is stressed by biotic or abiotic diseases, it needs more nutritional resources to be able to function better and in a certain quantity. Biostimulant not only allows the stressed tree to compensate for the lack of elements essential to its survival, but also improves the tree's productivity. This result is in line with the statement made by EBIC in 2014 through its definition of biostimulant.

Indeed, the difference in cherries production between apparently healthy trees (73%) and trees with swollen shoot disease (27%) testifies to the fact that swollen shoot disease is irreversible. An affected tree is doomed to die regardless of the amendments made to it because once its vascular system is destroyed, the tree is unable to be physiologically reactivated [4]. On the other hand, an apparently healthy tree is at the beginning of infection with still a high production potential. Moreover, its physiological system is still functional compared to a diseased tree.

The statistical results of Kruskal-Wallis also showed that the fertilizer used during the experiment had little effect on the production of cherries in both healthy and diseased trees. The little effect produced by the fertilizer is related to the fact that the effect of the fertilizer is felt later after application and not immediately. Indeed, fertilizer must take some time to be assimilated by cocoa trees [13]. This author argues that the effect of fertilizers on productivity depends largely on growing conditions, rainfall and sunshine. In addition, the period over which a specific fertilizer formula can be used is limited. As for the Coffee-Cocoa Council [14], the most suitable periods for fertilizer application are March-April for the 1st application and July-August for the 2nd application.

5. CONCLUSION, RECOMMENDATIONS AND PERSPECTIVES

At the end of this study, it appears that the application of Banzai biostimulant at standard dose without fertilizer (T1) had a significant impact on the production of cherries than other treatments in apparently healthy cocoa trees. In contrast, in diseased trees, the double dose of Banzai without fertilizer or the single dose of Banzai with fertilizer provided a production equivalent to the production of apparently healthy trees that had not received any Banzai applications. These results can be used to improve methods to control swollen shoot disease. Indeed, these results could compensate for the destruction of apparently healthy trees in an outbreak as recommended and allow the grower to optimize his production despite the presence of the disease in his plot. Based on these results, it would be recommended to use the simple dose of Banzai without fertilizer for apparently healthy cocoa trees around the infection sites in order to optimize production. However, the double dose of Banzai can be used as an alternative to early uprooting of cocoa trees affected by Swollen Shoot disease.

In perspective, it would be interesting to study the effect of Banzai in the case of control of cherry rot, pod rot and mirid attacks to assess the product's performance against these pests.

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REFERENCES

1- Journée Nationale du Cacao et de Chocolat (JNCC), 2016. Evolution de la filière café-cacao de 2012-2016. 44p.

2- Dufumier M, 2016. L'adaptation de la cacao-culture ivoirienne au dérèglement climatique : L'agroécologie pourrait-elle être une solution ? Rapport de mission, Côte d'Ivoire, 16 p

3- Coterell, 1943, Report of the Central Cocoa Research Station, Tafo, Gold Coast, 1938-1942 1943 pp.46-55

4- Oro F. Z., 2011. Analyse des dynamiques spatiales et épidémiologie moléculaire de la maladie du swollen shoot du cacaoyer au togo : étude de la diffusion à partir des systèmes d'information géographiques, Montpellier supagro. Thèse de doctorat école doctorale sibaghe, 262p.

DOI: 10.31695/IJASRE.2020.33683

5- Mossu G., 1990. Le cacaoyer. Le technicien d'agriculture tropicale. N° 14, pp.: 9-109.

6- Dzahini-Obiatey H, Domfeh O, Amoah FM (2010). Review : Over seventy years of a viral disease of cocoa in Ghana: From researchers' perspective. Afr. J. Agric. Res. 5 (7): 476-485.

7- Kebe, Ismaël. (2005). Cacaoyère ivoirienne en danger, le swollen shoot progresse. Cnra, *lepoint2005*

8- EBIC, 2014 Ebic (European biostimulants industry council. 2014 : Promoting the biostimulant industry and the rule of plant biostimulants in making agriculture more sustainable. (http://www.biostimulants.eu/).

9- Monographie du département de Soubré sud-ouest de la Côte d'Ivoire. Disponible sur : https://news.abidjan.net/h/527425.html consulté le 03/04/2018.

10- N'go, P.K., Azzaoui, F.Z., Ahami, A.O.T., Aboussaleh, Y., Lachheb, A., Hamrani, A., 2012. Déterminants socioéconomiques, environnementaux et nutritionnels de l'échec scolaire : cas des enfants résidant en zone cacaoyère de Soubré (Côte d'Ivoire). Antropo, 28, 63-70.

11- Diby L., Kouassi G., N'guessan M., Yao E., Oro F., Aynekulu E., Kassin E., Kouamé C., Coe R., Shepherd K., (2014). Cocoa Land Health Surveillance: An evidence-based approach to sustainable management of Cocoa landscapes in the Nawa region, South-West Côte d'Ivoire. Working Paper 193, 34 p.

12- Stefano PEDRAZZI, Youness RECHKA, Pegah PELLETERET, Romain CHABLAIS, Julien CROVADORE et François LEFORT : 2016. Evaluation de biostimulants commerciaux en culture de tomates en sol. Revue suisse Viticulture, Arboriculture, Horticulture | V 358 ol. 48 (6): 358–364.

13- ARAG, 2017. *Theobroma cacao* L. groupe génétique "Guiana" Développement de la filière locale du Cacao guyanais Programme d'actions de l'association ARAG. 28p

14- Conseil café-cacao. 2015. manuel technique de cacaoculture durable. 13p

DOI: 10.31695/IJASRE.2020.33683