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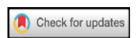

Impact of the Aerobic Mesophilic Microorganisms on Black Sigatoka of Bananas According to the Cropping Systems in the Region of Kisangani (*Case of the old secondary forest*)

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Abstract

The microorganisms play crucial roles in the cycle of matter and damage the organic substances, sources of electrons, source of carbon, and source of energy for their biosynthesis. In this work, we studied the impact of the aerobic mesophiles microorganisms on the black sigatokaof banana in the old secondary forest. The objective was to count and to identify the microbial diversity of the forest ecosystems, as well as to study their impact on the development of the black sigatoka of banana. The assessment of the microbial populations has been done in an experimental field according to a device in blocks of Fischer by the method of successive dilutions of coloration of Gram and by the discharge of ascospores. The results showed that the rain season was lower in total microbial biomass (700, 7 colonies) than subdry season (840,3 colonies). The *Bacillus* genera have been more represented more than the *Coccus*genera. The impact of black sigatoka of banana was raised at the cultivar LibangaLikale (40%) and low at Yangambi 5 Km (14%). The distribution of microorganisms in depth in the sub-dry season to the level of surface with vegetation was considerable either 3819, 3. 10^3 UFC(UNIT FORMAT COLONY) by gram of soil between 0 and 5 cm against 2754,5.10^3 UFC by gram of soil between 15 and 20 cm. This suggests that the raised number of microorganisms could have positive impact on soil fertility by decreasing the illnesses in this ecosystem.

Keywords: Aerobic mesophilic, Black Sigatoka, Cropping systems, Libanga Likale,Yangambi 5Km, Litete, Gros Michel, Mycosphaerellafijiensis Morelet, Bacillus.

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Contribution of this paper to the literature
The objective was to count and to identify the microbial diversity of the forest ecosystems, as well as to study their impact on the development of the black sigatoka of banana.

1. Introduction

The soil is not just the support in which plants take root and can provide the nutrients essential for their development, but also is a reservoir of microorganisms.

These soil microorganisms are important for plant productivity. They are the most abundant living organisms in the soil, and they play an active role in the nutrient and organic matter cycle, soil fertility, plant health and production. Over 90% of plants worldwide develop symbiotic associations with at least one type of mycorrhizae [1].

In the soil, there are beneficial bacteria that maintain very intimate relationships with the plant and strengthen its natural defenses against stress of biotic origin.

Bacteria are of considerable importance in biogeochemical cycles, such as those of carbon or nitrogen with a fundamental role in the fixation of atmospheric nitrogen, a function which has been much studied for several decades [2].

The soil is the outermost layer, marked by living things in the earth's crust. It is the site of an intense exchange of matter and energy between air, water and the global cycles of matter [3]."

It is considered to be one of the most biospheres and is therefore a major reservoir of microbial diversity. It is made up of insoluble mineral debris produced by the fragmentation and alteration of sedimentary rocks (limestone, etc.), organic matter (colloidal humus, etc.), living and dead organic matter, air and water, which allow the proliferation of telluric microorganisms [4].

The physical properties of the soil are determined by the sizes of the particles that compose it. We can classify them by decreasing diameter: sands, silts, clay. The organic part of the soil includes plant debris, animal remains and varying amounts of amorphous organic matter called humus. The gases contained in the soil are mainly oxygen, nitrogen, carbon dioxide (CO2) and mineral substances dissolved or dissociated by hydrolysis (mineral ion). Oxygen is important for the metabolism of plants and their growth. Its presence is also necessary for the development of bacteria, and other organisms.

In the soil, there are not only microorganisms that swarm. Animals, plants, and viruses are also found [5].

This population of the soil undergoes great fluctuations because of the physicochemical factors of the soil in particular the structure, the organic matters, the textures, the ionic concentration, the temperature, the humidity, the pH, the biotic factors: the types of relations (symbiosis parasitism, predation, and commensalism), the phenomenon of inhibition. To these is added the individual potentiality of each species (reproduction, adaptation, feeding).

Soil is considered today as a vital, non-renewable source, which should therefore be preserved, although the horticultural uses non-soil growing media. Most of the methods developed to ensure soil health apply to horticultural substrates and every effort should be made to limit the use of synthetic chemical inputs and depreciate natural resources such as silt [4].

Microorganisms play crucial roles in the cycle of matter and degrade organic matter, sources of electrons, source of carbon, and source of energy for their biosynthesis. By dying, microorganisms in turn help enrich the soil with different carbon compounds [6].

On the other hand, many of them promote the growth of plants, ensure the degradation of pollutants and provide compounds of interest, enzymes, antibiotics or other molecules.

Microorganisms are found in all types of environment found in nature: They colonize all ecosystems, such as soil, freshwater and marine water, air, but also more hostile environments such as the poles, deserts, geysers, the ocean floor, etc. Microorganisms found in extreme environments are called extremophiles [7].

Many microorganisms are associated with plants or animals with which they can maintain relationships of symbiosis, commensalism or parasitism. Some microorganisms can be pathogenic, that is, cause disease in plants or animals.

Man very early used the properties of microorganisms (bacteria) to feed, to heal. Today the fields of application are very varied. In the food industry, bacteria , such as *Lactobacillus*,*Lactococcus*or *Streptococcus*, yeasts and molds are involved in the production of fermented foods, such as cheese, yogurt, beer [7].

The ability of heterotrophic bacteria to degrade a wide variety of organic compounds is exploited for the treatment of polluted soils in bioremediation strategies or for the treatment of wastewater [8]. Bacteria are also used in septic tanks to purify them. In agriculture, some bacteria can be used in place of pesticides in biological control to fight plant parasites [9]. (ex: *Bacillus thuringiensis*), other bacteria will have a beneficial effect on the growth of plants [10].

Black Sigatoka or Black Streak Disease, caused by the fungus *Mycosphaerellafijiensis*Morelet, is considered to be the most devastating disease in the world banana crop [11]. This disease, which has a wide geographic distribution, causes early death of infected leaves and is responsible for more than 50% of the losses in banana production [11].

The banana tree is a giant and perennial herb 1.5 to 9 m tall. It consists of a corm (underground part) with roots and shoots, a pseudo trunk provided with leaves, and a bunch of fruits Jones [12]; Swennen and Vuylsteke [13]. Its reproduction is vegetative and ensured by suckers [14]. Bananas are monocots without an aerial vegetative stem [15] classified in the order of

Zingiberal, the *Musaceae*family and the genus of *Musa*.

The genus *Musa* is characterized by inflorescences with bracts inserted separately from the flowers, and includes 5 sections: *Australimusa*, *Callimusa*, *Rhodochlamys*, *Eumusa and Ingentimusa* Swennen and Vuylsteke [13]. The *Australimusa* include a species cultivated for its fibers, *M. textilis* and the Féhi of the Pacific islands bearing an erect inflorescence, fruit to be cooked and a colored sap. The *Callimusa* include a few ornamental species such as *M.coccinea*. The *Rhodochlamys* also include ornamental species such as *M. ornata*, *M. velutina* and *M. laterita*. Edible

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bananas belong to the *Eumusa*section and are divided into three categories according to consumption: cooking bananas, dessert bananas and wine bananas.

In the Democratic Republic of Congo (DRC), it is currently spread in all the provinces [16].

Bananas and plantains play an important economic, cultural and nutritional role worldwide, particularly in developing countries in the tropics [17]. They are among the main staple foods in producing countries and constitute a considerable source of employment and income through local and international markets [18].

In the Kisangani region, no studies have been done on the contribution of microorganisms to banana fungal diseases. That is why this study was carried out around the Masako Forest Reserve in order to determine the impact of aerobic mesophilic microorganisms associated with black Sigatoka of banana.

The causative agent of MRN for bananas is M. fijiensisMorelet. According to Cabi [19], this mushroom is classified as follows:
Phylum: Ascomycota.
Class: Ascomycetes.
Subclass: Dothideomycetidae.
Order: Mycosphaerellales.
Family: Mycosphaerellaceae.
Genus: Mycosphaerella.

2. Material and Methods

2.1. Study Area

The research was conducted in the Kisangani region, around Masako Forest Reserve (0 ° 36'N, 25 ° 13'E, Eastern Province) [20]. Covers an area of 2105 ha of which 1/3 is occupied by the northeast primary forest and at least 2/3 by the secondary forests in the northwest [21]. While the south part of the reserve is occupied by fallow and crops (see figure 1).

The Masako Forest Reserve is part of the relatively humid Guinean-Congolese rain forests. It includes six types of vegetation, including: ruderal vegetation, crop weed, post crop vegetation, *Musangacercropioid* vegetation, *Gilbertiondendrondewevrei* forest, and aquatic and semi aquatic vegetation. The soil of the experimental site is sandy-clay, lumpy, fine to medium, brittle, with many roots, dark brown in color [22]. All the eco-climatic data the position of the city of Kisangani are near the Equator, and give it an equatorial climate of the Af type in the Köppen classification [23].

2.2. Experimental Set Up

The experimental plots were established in fallow fields and secondary forest according to the Fisher block system [24].With 4 treatments and 5 repetitions per treatment. The blocks, also called repetitions, were made up of a group of plots almost forming a square. Five blocks or repeats each containing six plants of the same cultivar was placed in each plot (see figure 2). Four locally cultivated varieties (Gros Michel, Libanga Likale, Litete and Yangambi 5Km) were used and distributed randomly in the plots of each block.

2.3. Biological Material

The biological material used in this work consisted of soil samples taken under aseptic conditions, using a graduated auger, brand Eijkelkamppo Box 469872 G, in a field of old secondary forest. The type of soil examined in this work was of the silty-sandy type with a fine structure and a very strong anthropic activity.

2.4. Plant Material

The plant material consisted of 4 cultivars of bananas and plantains, including LibangaLikale (Plantain: Musa AAB), LibangaLikale (Plantain: Musa AAB), Gros-Michel (Banana: Musa AAA), and Yangambi Km5 (Banana: Musa AAA), the most cultivated by farmers in Kisangani region. The choice of these cultivars was motivated by their presence in the study environment and this is justified by several reasons mentioned by the inhabitants of the village (figure 3).

2.5. Sampling of Soil Microbial Populations

We collected the soil under aseptic conditions using an auger. We weighed 10 g of soil under the same conditions, and then fixed the sample in 90 ml of peptone water. A series of successive dilutions was made, but only one dilution for 2 Petri dishes due to 1 ml of inoculum per dish was seeded. The liquefied nutrient agar cooled to 45 ° C was poured into the seeded Petrie dish and after solidification, a second layer was poured. The incubation was carried out at 30 ° C for 72 hours. The number of CFU (Colony-forming Unit) was obtained using the following formula:

$$N = \frac{\sum Colonies}{Vml.(n1 + 0,1n2).D1}$$

Where: N: number of CFU / g (ml) of the initial product.
$\sum$ Colonies: sum of colonies of interpretable boxes.
Vml: volume of inoculum = 1 ml.
n1: number of boxes considered at D1 selected.
n2: number of boxes considered at D2 selected ; D1: Factor of the selected D1.

2.6. Identification of Aerobic Mesophilic Microorganisms

The bacteria were identified using at genus level using by the Gram staining method. The bacteria were classified into two groups: Gram positive colored purple and Gram negative colored pink. Gram staining was based on the principle that crystal violet stains of all bacteria purple or dark blue. Lugol played the role of an etcher. Alcohol acetone crossed the wall of some bacteria and discolored them. However, the wall of other bacteria

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remained impassable by alcohol and retained the purple or dark blue color which was well reinforced by lugol. Finally, saffron colored pink or red any bacteria which wall was discolored and did not act on bacteria colored purple or dark blue [25].

We took the inoculum and spread it on an object slide to get a smear smearusing a platinum loop. We then dried the smear at laboratory temperature and fixed the preparation by passing the bottom of the slide over the flame. Then we covered the entire smear successively with crystal violet, lugol, alcohol and saffron; while leaving a minute of reaction for each dye, except the alcohol which was left for 30 seconds of reaction. Before switching from one dye to another, the preparation was washed with tap water. Finally, we dried the preparation at laboratory temperature, applied a drop of immersion oil and observed under the microscope at 100x objective.

2.7. Mushroom Isolates

From the samples of collected leaves, the strains of Mycosphaerella spp. were isolated by the ascospore discharge technique on agar medium (H2O Agar), which were then subcultured on Potato Dextrose Agar medium (PDA) [24]. For discharge, the pieces of necrotic leaves were first cut, then soaked in sterile distilled water for 20 minutes to moisten them. The leaf pieces were then placed inside lids of Petri dishes which were placed on dishes containing 3% agar. The underside of the leaf was directed upwards, facing the culture medium. The dishes were incubated at 25 ° C overnight. The following day, the ascospores discharged onto the agar overnight were subcultured individually on the PDA medium (39 g / l).

Transplanting was done by observation with an inverted microscope (Motic AE31), carefully recovering, one by one, the discharged ascospores using a needle. The cultures were incubated at 25 ° C. and the phenotype of the strains observed after 45 days to evaluate the correspondence with that of M. fijiensis, M. musicola or M. eumusae. The strains obtained were subcultured in Petri dishes and in tubes in order to preserve the established collection.

2.8. Evolution of the Disease

Data were recorded on only one variable: the incidence of the disease.

The incidence of the disease (number of plants infected with *Mycosphaerella fijiensis* expressed as a percentage) was assessed every 2 weeks from 2 months after planting.

2.9. Data Analysis

Statistical analysis was performed using R.2.10.0 software.

3. Results and Discussion

The results of our investigations are presented in Tables 1 to 4 and illustrated by Figures 4 and 5. The total density of the microbial flora according to the depth during the dry season and that of rains is presented in table 2.

Table-1. Number of microbial colonies as a function of soil depth during two seasons.

Season	Depth (cm)	Number of colonies
Sub-dry	0-5	1238
	5-10	889
	10-15	628
	15-20	606
	Average	**840,3**
Rain	0-5	504
	5-10	588
	10-15	726
	15-20	788
	Average	**700,7**

By examining Table 1 relating to the density of bacteria during the dry season and that of rain, it appears that the number of bacteria varies according to the seasons as well as the depths. However, the rainy season is low in total microbial biomass with an average of 700.7 number of microbial colonies compared to the dry season which showed a high value of the number of colonies is 840.3. Furthermore, the number of microorganisms was higher at the surface, 1238 between 0 and 5 cm during the dry season and decreases with depth.

A contrary situation was observed during the rainy season when this number increases with depth since we observed 504 colonies on the surface and 788 in depth. This could be due to the unfavorable climatic conditions on the surface during the rainy season allowing the mesophilic bacteria to penetrate in depth and to grow between 20 and 45 ° C. Compared to previous work, Alexander [26] found 97,510 bacteria per gram of soil, the bacterial titer, while Meddah, et al. [27]. found up to 5,4,107. The distribution of bacteria in depth during the dry season and that of rain is illustrated in Figure 4.

Looking at Figure 4, it appears that a decrease in the number of microorganisms was observed with depth during the dry season while an increase in this number was observed during the rainy season. In addition, the distribution of microorganisms in depth in the dry season at the level of the planted surface increased from 3819.3.103 CFU to 3184.8.103 CFU per gram of soil. This suggests that the environmental conditions of the environment are recognized as playing a determining role in the dynamics of colonization by the microbial population.

In the middle of the forest, the densities are much higher and the highest concentrations are located at the level of the first 10 centimeters and decrease slightly at the last centimeter. This suggests that the high number of microorganisms could have an impact on soil fertility and thereby decrease the incidence of diseases in this ecosystem. Furthermore, abiotic factors can also affect the specific composition of microbial communities and their biochemical potential. As a result, we see that there is a drop in microorganisms and we have found that the number of colonies in certain boxes, for example in the last 20 centimeters deep, is higher than on the surface.

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The variations recorded are due to physicochemical factors as well as to crops which could exert certain influences on microbial populations in the soil. Also, abiotic factors can affect the specific composition of microbial communities and their biochemical potential.

The results of the identification of the bacteria by Gram staining during the dry season are presented in Table 2.

Table-2. Characterization of Bacteria after Gram staining during the dry season.

subsèche.

Strains	Form	Gram Staining	Genus	Density
S1	*Bacillus*	Gram+	*Bacillus*	++++
S2	*Coccus*	Gram+	*Staphylococcus*	++
S3	*Bacillus*	Gram+	*Bacillus*	++++
S4	*Coccus*	Gram+	*Staphylococcus*	++
S5	*Bacillus*	Gram+	*Bacillus*	++++
S6	*Bacillus*	Gram+	*Bacillus*	++++
S7	*Bacillus*	Gram+	*Bacillus*	++++
S8	*Bacillus*	Gram+	*Bacillus*	++
S9	*Bacillus*	Gram+	*Bacillus*	+

Legend: S: Strain
+: 25%
++ : 50%
+++ : 75%
++++ : 100%

In light of this table, it appears that during the dry season, the *bacillus* and shell forms were observed with a large dominance of the first form which also dominates in density. With regard to Gram staining, all the strains observed are Gram + belonging to the genus *Bacillus* and *Staphylococcus*. The method used being specific to genera, it did not allow us to determine the species.

The results of the identification of bacteria by Gram staining during the rainy season are presented in Table 3.

Table-3. Characterization of Bacteria after Gram staining in the rainy season.

Strains	Form	Gram Staining	Genus	Density
S1	Bacillus	Gram+	Bacillus	++++
S2	Bacillus	Gram+	Bacillus	++
S3	Bacillus	Gram+	Bacillus	+++
S4	Bacillus	Gram+	Bacillus	+
S5	Coccus	Gram+	Diplococcus	+++
S6	Coccus	Gram+	Streptococcus	++

Legend: S: Strain
+: 25%
++ : 50%
+++ : 75%
++++ : 100%

The observation in Table 3 shows that the *bacillus* form has been more observed (4 strains) than the shell form (2 strains) although all the bacteria are Gram +. As for the genus, the *bacillus* form has been represented by a single genus, *Bacillus*, while the shell form has been represented by the genera *Diplococcus* and *Streptococcus*.

Figure 5 illustrates the incidence of black Sigatoka in bananas and plantains in the old secondary forest. The overall rate for all cultivars was below 50%, and ranged from 14 to 40%. As in previous studies carried out in a hut garden [16]. we find that plantains were more susceptible to this disease and therefore had a high incidence, 40% for Libanga Likale and 32% for Litete than banana trees. In addition Gros Michel was more sensitive to this disease (34%) while Yangambi km 5 (14%) showed tolerance to black Sigatoka.

Table-4. Effect of depth on the density (CFU) of bacteria in the rainy season.

Depth(cm)	Sample	D1	D2	CFU (10^3)
0-5	B1	260	217	4113,6
	B2	271	157	
5-10	B1	219	133	2600
	B2	105	112	
10-15	B1	97	98	1650
	B2	89	79	
15-20	B1	72	68	1295,4
	B2	67	78	

Légend: B1 et B2 : Petridish 1 et 2, Cm : Centimeter, D : Dilution, CFU : ColonyFarmat Unit.

The findings showed that there is a high concentration of the CFU density of bacteria (4113.6) followed by the depth in CFU density of the bacteria of 2600 of 5-10, the depth on the density of UFC of 10-15 with 1650 UFC, and the depth of 15-20cm with 1295.4UFC of bacteria. Table 4 revealed that the density of bacteria decreased according to the depth in the rainy season. The density of bacteria was the highest at 0-5cm and lowest at 15-20cm.

Table 5 showed that the density of bacteria increased according to the depthin the dry season.

The bacteria were the highest at 15-20cm (with CFU of 2918.2.10^3) and lowest at 0-5 CFU of (995.5.10^3).As shown in Tables 4 and 5, the density of bacteria varied differently according to the depth between these two seasons.

This result may be explained due to during the dry season,the bacteria moved to moved deep to look for their nutrients.

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Table-5. Effect of depth on the density (CFU) of bacteria in the dry season.

Depth (cm)	Sample	D1	D2	CFU (10^3)
0-5	B1	45	47	995,5
	B2	69	58	
5-10	B1	72	69	1609,1
	B2	120	93	
10-15	B1	88	105	2145,5
	B2	99	180	
15-20	B1	111	210	2918,2
	B2	103	218	

Légend: B1 et B2 : Petridish 1 et 2, Cm : Centimeter, D : Dilution, CFU : ColonyFarmat Unit

Table-6. Incidence of black Sigatoka in old secondary forest.

Cultivars	LibangaLikale				Litete				Gros Michel				Yangambi Km 5			
Plants	N	PS	PI	%	N	PS	PI	%	N	PS	PI	%	N	PS	PI	%
Bloc 1	6	3	3	50	5	4	1	20	4	2	2	50	5	4	0	0
Bloc 2	5	3	2	40	4	3	1	25	5	3	2	40	5	5	0	0
Bloc 3	5	4	1	20	4	3	1	25	5	3	2	40	6	4	2	33
Bloc 4	4	2	2	50	5	3	2	40	5	4	1	20	5	4	1	20
Bloc 5	5	3	2	40	4	2	2	50	5	4	1	20	6	5	1	17
Avarage				40				32				34				14
Standard deviation				12.2				12.5				13.4				14

Legend: N: Number of Plant, P.S.: Plant Saint, P.I.: Plant infected.

Table 6 indicated that the highest incidence of black Sigatoka in old secondary forest was recorded in cultivar Libanga Likale (with average of 40%) and the lowest in Cultivar Yangambi 5Km (with average of 14%). We concluded that cultivar Yangambi 5Km was the most resistant to black Sigatoka.

4. Conclusion and Suggestions

The main objective of this work was to count and identify microbial diversity as well as to study the incidence of black Sigatoka in old secondary forest near the Masako forest reserve in Kisangani. The evaluation of microbial populations was carried out in an experimental field using a Fischer block device. Parameters such as density of microbial population, depth of soil, characterization of microorganisms and incidence of black Sigatoka were assessed during the dry and rainy seasons.

The results obtained during this work show that:

The rainy season is low in total microbial biomass (700.7 colonies) than the season dries up (840.3 colonies).

The bacteria rate increases with depth during the rainy season and decreases during the rainy season.

The distribution of bacteria in depth in the dry season at the level of the planted area is considerable, at 3,819.3. 103 CFU per gram of soil between 0 and 5 cm compared to 2754.5.103 CFU per gram of soil between 15 and 20 cm.

The number of CFUs during the rainy season varied from 2,990.9.103 to 3,581.8. 103 CFU per gram of soil between 0-5 cm and 15-20 cm respectively.

The incidence of black Sigatoka in bananas is high in the cultivar Libanga Likale (40%) and low in Yangambi Km 5 (14%). In addition Litete and Gros Michel were presented with intermediate values respectively 32 and 34%.

The number of bacteria is relatively high in this ecosystem and would reduce the incidence of black Sigatoka in bananas and plantains

Statistically, the one-factor analysis of variance has shown that there are significant differences between the parameters studied.

We suggest that:

In-depth studies are carried out to characterize microorganisms at the level of species existing in the soil in order to determine their relationships with soil fertility.

That other studies be carried out in more depth on the molecular characteristic of microbial communities in the Kisangani region in order to better understand them given their very small size.

References

[1] A. Faugier, "Bacterial diversity of soils: Access to populations with moniritary numbers “the rare biosphere," Unpublished Doctoral Thesis, ECL, 2010.

[2] P. M. Vitousek and R. W. Howarth, "Nitrogen limitation on land and in the sea: How can it occur?," *Biogeochemistry*, vol. 13, pp. 87-115, 1991. Available at: https://doi.org/10.1007/bf00002772.

[3] Swiss Society of Soil Science, "Definition of soil for public use," *Internal Document*, vol. 6, pp. 189-203, 1997.

[4] F. Aurelie, "Bacterial diversity of soils: Access to populations with a monetary workforce "the rare boisphere," Thesis Unpublished, Electronic, Electrotechnical and Qutomatic Doctoral School of Lyon, 2010.

[5] R. Mansisa, "Identifying the dominant soil bacterial taxa in libraries of 16S rRNA and 16S rRNA genes," *Applied and Environmental Microbiology*, vol. 72, pp. 1719-1728, 1982. Available at: https://doi.org/10.1128/aem.72.3.1719-1728.2006.

[6] R. Calbrix, "Impact of organic inputs and cultural practices on microbial biomass and the diversity of telluric bacteria," Unpublished Thesis, University of Rouen, 2005.

[7] S. Hanniffy, C. Pelaez, M. Martinez-Bartolome, T. Requena, and M. Martinez-Cuesta, "Key enzymes involved in methionine catabolism by cheese lactic acid bacteria," *International Journal of Food Microbiology*, vol. 135, pp. 223-230, 2009. Available at: https://doi.org/10.1016/j.ijfoodmicro.2009.08.009.

[8] K. Furukawa, "Super bugs' for bioremediation," *Trends Biotechnol*, vol. 21, pp. 187-190, 2003.

[9] M. Lucy, E. Reed, and B. R. Glick, "Applications of free living plant growth-promoting rhizobacteria," *Antonie van Leeuwenhoek*, vol. 86, pp. 1-25, 2004. Available at: https://doi.org/10.1023/b:anto.0000024903.10757.6e.

[10] B. Lugtenberg and F. Kamilova, "Plant-growth-promoting rhizobacteria," *Annual Review of Microbiology*, vol. 63, pp. 541-556, 2009.

[11] D. H. Marin, R. A. Romero, M. Guzmán, and T. B. Sutton, "Black Sigatoka: An increasing threat to banana cultivation," *Plant Disease*, vol. 87, pp. 208-222, 2003. Available at: https://doi.org/10.1094/pdis.2003.87.3.208.

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[12] D. R. Jones, "Diseases of banana, abacat and enset, londres," ed UK: CABI Publishing, 2000, pp. 4-11, 37-39 et 173-190.
[13] R. Swennen and D. Vuylsteke, "Bananier. In: Raemaekers, R. H. Agriculture in Tropical Africa," ed Brussels: DGCI, 2001, pp. 661-636.
[14] C. W. Wardlaw, "Banana diseases," ed London: Longmans Group, 1961, p. 648.
[15] A. Lassoudiere, "The banana tree and its culture," ed France: Edition Quae, 2007, p. 384.
[16] O. Onautshu, "Characterization of Mycosphaerella fijiensis populations and epidemiology of black Sigatoka of banana (Musa spp.) in the Kisangani region (DRC)," Unpublished Doctoral Thesis, UCL, 2013.
[17] B. K. Dadzie and J. E. Orchard, *Routine post-harvest screening of banana/plantain hybrids: Criteria and methods*. Montpellier, France: INIBAP, 1997.
[18] E. Frison and S. Sharrock, "The economic, social and nutritional importance of banana in the World. In C. Picq, E Fouré and E. Frison (Eds.), Bananas and Food security," in *Proceeding of an International Symposium held in Douala, Cameroon, 10 - 14 November 1998. INIBAP, Montpelleir, France*, 1999, pp. 21 – 35.
[19] Cabi, "Crop protection compendium. Retrieved from http://www.cabicompendium.org," 2007.
[20] A. Upoki, "Study of the stand of bulbuls (Pycnonotidae, Passériformes) in the Masako forest reserve at Kisangani (R.D.Congo)," Unpublished Doctoral Thesis, Fac. Sci., Kisangani, 2001.
[21] J. Lejoly, S. Lisowski, and M. Njele, "Catalog of vascular plants from the Kisangani and Tshopo sub-regions (Haut-Zaire)," 3rd ed Belgium: Labor. Botan. Syst. Phytos. ULB, 1988, p. 122
[22] K. Soki, "Biology and ecology of termites (Isoptera) from the rain forests of Northeast Zaire (Kisangani)," Unpublished Doctoral Thesis, ULB, 1994.
[23] M. Mate, "Growth, phytomass and mineralomass of hedgerows of improving legumes in alley crops in Kisangani," Unpublished Doctoral Thesis, 2001.
[24] J. Carlier, D. De Waele, and J. V. Escalant, "Overall assessment of the resistance of banana trees to Fusarium wilt, to leaf diseases caused by Mycosphaerella spp. and nematodes," INIBAP Technical Guides 7. Montpellier, France2003.
[25] W. Oleko, "Course," *Applied Microbiology*, vol. 1, p. 92, 2011.
[26] M. Alexander, *Introduction to soil microbiology*, 2nd ed. United States of America: John Wiley & Sons, Inc, 1977.
[27] N. Meddah, A. O. Touhami, and A. Douira, "Mycoflora associated with banana (Musa accuminata L.), Grande naine variety, grown under glass in the Gharb region (Morocco)," *Bulletin of the Scientific Institute, Rabat, Life Sciences Section*, vol. 32, pp. 1-11, 2010.

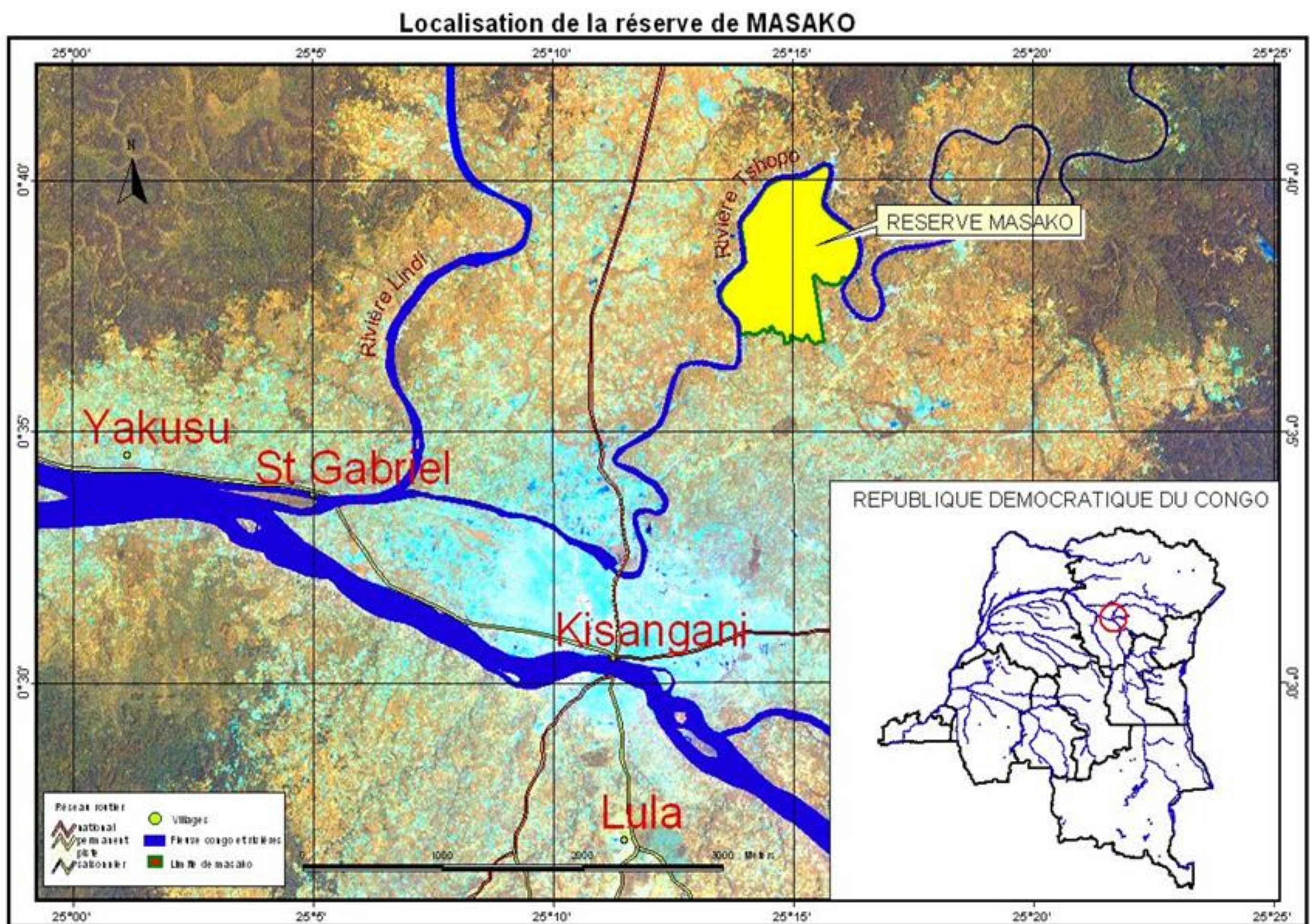

Figure-1. Location of the Masako Forest Reserve.
Source: Adaptation of 2010 satellite photo, of Lisingi and CFT.

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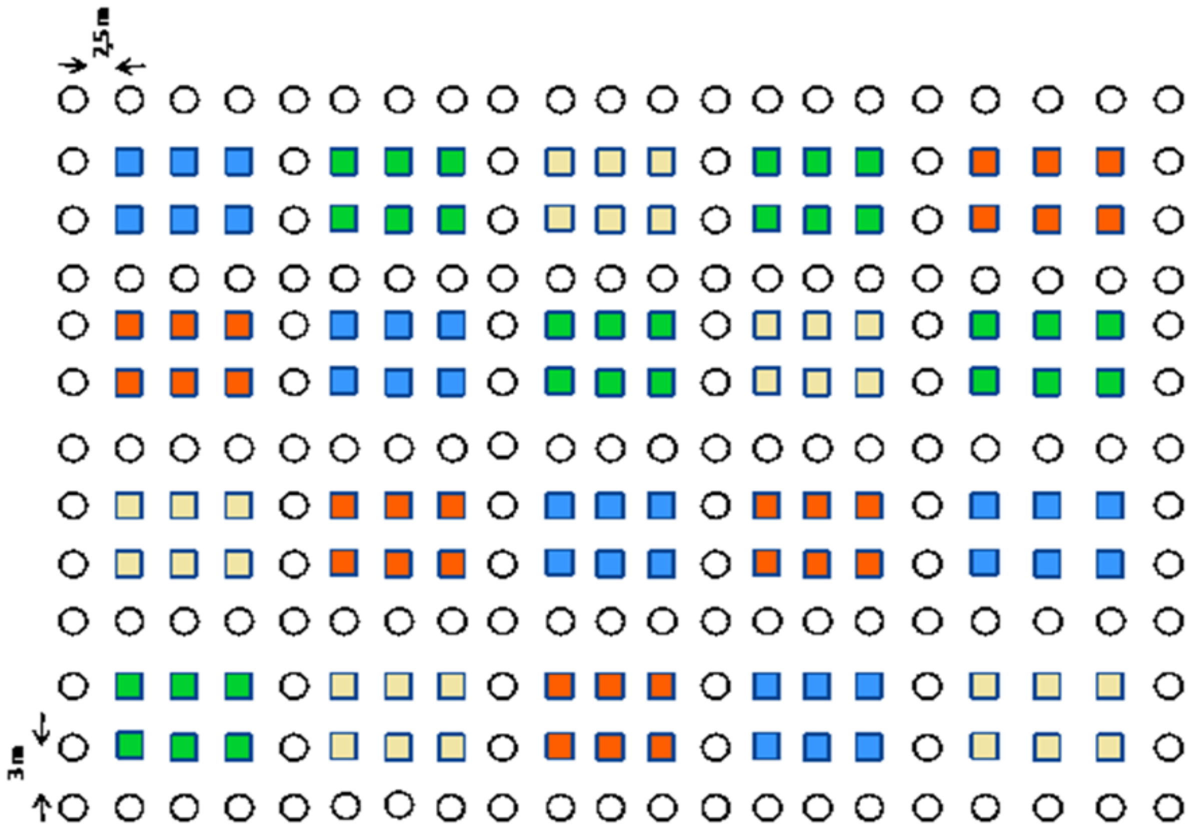

Figure-2. Fisher block device.

Legend:

: Gros Michel

: Libanga Likale

: Litete

: Yangambi Km 5

: Edge plants

Libanga Likale (Musa AAB)

Litete (Musa AAB)

Gros Michel (Musa AAA)

Yangambi Km 5 (Musa AAA)

Figure-3. Banana and plantain cultivars used

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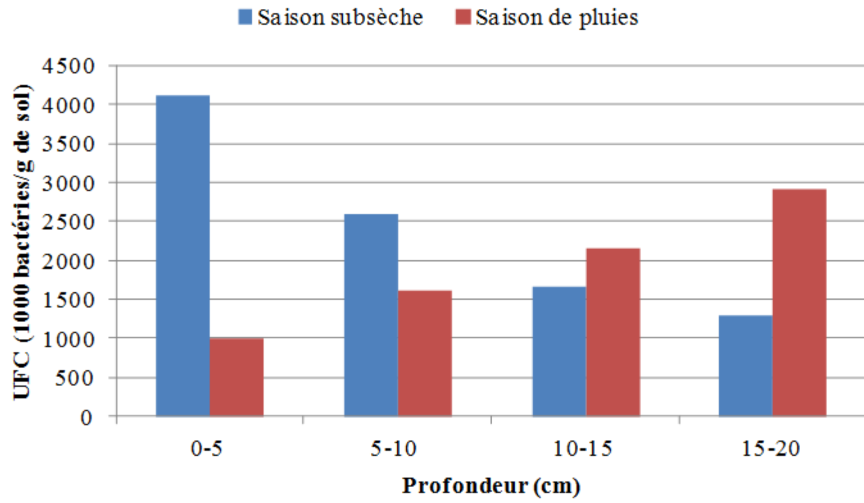

Figure-4. Distribution of bacteria by depth over two seasons.

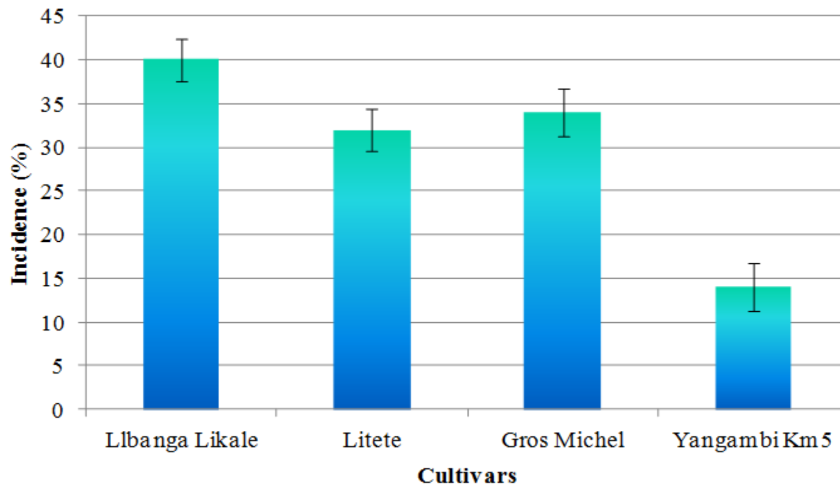

Figure-5. Incidence of black Sigatoka in bananas and plantains in old secondary forest in Masako in 2014.

Statistiques analysis result

- **ANOVA :CFU-Saison**

Df Sum Sq Mean Sq F value Pr(>F)
Saison 1 990722 990722 1.0264 **0.3282**
Residuals 14 13513548 965253
meansd n
Rainy 1917.075 755.6602 8
Dry 2414.750 1165.9694 8

- **ANOVA : CFU-Depth**

Df Sum Sq Mean Sq F value Pr(>F)
Depth 3 920840 306947 0.2712 **0.845**
Residuals 12 13583431 1131953
meansd n
0-5 cm 2554.55 1800.2359 4
10-15 cm 1897.75 286.0771 4
15-20 cm 2106.80 936.9240 4
5-10 cm 2104.55 572.0964 4

- **ANOVA : Incidence-Cultivar**

Df Sum Sq Mean Sq F value Pr(>F)
Cllivars 3 1880.0 626.67 3.6343 **0.03581** *
Residuals 16 2758.9 172.43

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
meansdn
Gros Michel 34 13.41641 5
LibangaLikale 40 12.24745 5
Litete 32 12.54990 5
Yangambi 5Km 14 14.22049 5

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