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# Tolerance of Banana and Plantain Cultivars (*Musa* spp.) against the Epidemiology of BBTV, Grown in Open Fields, Maniema, DR Congo

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# ABSTRACT

The objective of the study was to evaluate the expression of banana and plantain (*Musa* spp.) cultivars grown in the open field in the face of the epidemiology of BBTV, Maniema, DR Congo. Specifically, to determine which banana and plantain (Musa spp.) cultivars are resistant, tolerant and sensitive to the epidemiology of BBTV. To achieve the objective, methods relating to the latter were used. A single factorial design of elongated plots with 11 repetitions and 11 cultivars of banana and plantains was set up. The spacings chosen were 3 m x 3 m, i.e. a density of 121 feet in a field of 40 m x 40 m. Thus, the results obtained showed that from the TAS ELISA in the presence of the vector (Pentalonia nigronervosa) the plants of banana and plantain cultivars with or without symptoms of BBTD are sometimes carriers of the banana bunchy top virus (i.e. 24.28%). There is a strong correlation between plant-vectordisease-production, that is to say; plants carrying aphid colonies are likely to be more infected by BBTV and cause significant yield losses. The correlation between production, incidence and severity makes it possible to uncover the resistance and tolerance of cultivars to BBTV. The cultivars Kamboloso, Lokoka, Yangambi Km 5 and Tala lola having achieved excellent yields are said to be resistant to bunchy top disease, their severity in the TAS ELISA oscillated between 0-10% of the general scale for evaluating viral diseases. Furthermore, the cultivars Libanga likale, Leese, Andula, Lokusu, Losakala and Nguku are tolerant to BBTD given that they obtained good yields and their ELISA severity level evolved between 11-40% and finally, the Kamaramasenge cultivar is sensitive to BBTD because it achieved an intermediate yield and a severity level varying between 41-75% of the general scale assessment of viral diseases. This study clearly confirms that there is a positive effect of aphid bite inoculum on virus transmission in banana and plantain cultivars grown in the open field. We recommend that genetic improvement studies be carried out by other researchers to explore the resistance and tolerance genes found in the cultivars Kamboloso, Lokoka, Yangambi Km 5, Tala lola, Libanga likale, Leese, Andula, Lokusu, Losakala and Nguku for better cultural control of BBTV and an improvement in the productivity of bananas and plantains in the sub-region of Africa to the east of the RD Congo in particular and throughout the world in general.

Keywords: tolerance, banana and plantain cultivars, epidemiology of BBTV

# **INTRODUCTION**

Cultivated bananas (dessert and plantains) constitute a food source for millions of people around the world with an annual production of 50 million tonnes of dessert bananas and 45 million tonnes of plantains (CIRAD, 2019). At the regional level, 57% of production comes from Asia, 28% from Latin America and 15% from Africa. As for the plantain type, Africa

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holds 60.9% of its global production, or more than 39 million tonnes (FAOSTAT, 2018). Sweet bananas are the most important fruits in tropical regions and the most consumed around the world both inside and outside the tropics. In terms of global fruit trade, bananas occupy first place in terms of turnover (Frisson & Sharrock, 1999). As for plantain, it constitutes one of the staple foods of many populations of Central Africa, in general, and of the Democratic Republic of Congo in particular. Plantain is generally consumed after cooking and plays an important role in the social structure of many rural communities because, in most cases, it becomes the sole source of income (Tollens, 2004).

In DR Congo, plantain and banana are produced under different cultivation systems, namely house crops, field crops, association or monoculture. Among these systems, the most sustainable is that of hut crops because the plants grown there produce over several cycles and are less affected by insects and diseases than those grown in fields (Dowiya *et al.*, 2009; Mobambo *et al.*, 1994). The Congo basin constitutes one of the important poles of plantain and the second center of diversity in the world after Asia (Mobambo *et al.*, 2010). In DR Congo, bananas and plantains are grown at different altitudes and are associated with several plants, especially on small farms. They are cultivated mainly for self-consumption, thus playing an important role in the food security of the population in the different agro-ecological zones (Dhed'a *et al.*, 2019). Despite the undeniable importance of bananas and plantains, in DR Congo several biotic and abiotic factors limit their production significantly. These include traditional farming practices, soil exhaustion, lodging due to increasingly violent winds at the start of torrential rains, attacks by diseases and pests (Dhed'a *et al.*, 2019).

The crop suffers from many diseases caused by fungi, bacteria as well as viruses. In recent decades, Banana Bunchy Top Disease (BBTD) has been one of the most important diseases of bananas and plantains in several regions of Africa, Asia and the South Pacific (Mukwa *et al.*, 2014). This disease is widely present in all banana-producing regions of the world, with the exception of Latin America (Wickramaachchi *et al.*, 2016). In many banana growing areas, banana production and area are reduced by 90-95% when the disease is present (Caruana, 2003). This disease is expanding worldwide (Kagy *et al.*, 2001). In DR Congo, Banana Bunchy Top Disease (BBTD) was introduced in 1958 in Yangambi by importing infected planting materials offered as a gift to the King (Swennen *et al.*, 2001). For the moment, BBTD is already reported mainly in the Central Congo, North and South Kivu Provinces as well as in the Tshopo Province (Dowiya *et al.*, 2009, Ngama *et al.*, 2015).

Like the province of Bandundu, Equateur, greater Kasai as well as Katanga particularly in its northern part, the province of Maniema is also affected by BBTD (Shadari *et al.*, 2023). This disease also called "banana bushy top" is due to the Marmor abaca HOLMES virus called Banana Bunchy Top Virus (BBTV). It is a single-stranded DNA-containing virus of the nanoviridae family belonging to the Babuvirus genus; the disease mainly affects growth and renders affected subjects unproductive in the vegetative phase (Nzawele, 2012, Ngama, 2010). BBTV is an obligate parasite which carries out its multiplication cycle in the host plant which is the banana tree (Caruana, 2003). It spreads from plant to plant by aphids and from place to place within the plantation by people transporting planting material from infected plants (Kallow *et al.*, 2021).

According to Niyongere *et al.* (2012), the virus is transmitted locally in a persistent and circulating manner by the vector agent *Pentalonia nigronervosa* Coquerel. Symptoms usually appear on the second emerging leaf after inoculation in accordance with the observations of Caruana (2003), then Busogoro *et al.* (2009). This rapid replication of the virus allows symptoms to appear immediately over time. Symptomatic levels will therefore evolve quickly when environmental conditions are favorable. The chronological stages involved in the initiation of the different defense reactions developed by plants following an attack by a pathogen have been the subject of more specific studies (Yoshikawa *et al.*, 1993). For several

years now, the mechanisms associated with the plant's response following microbial attack have also been the subject of in-depth studies with the aim of better understanding their biological functions in a spatio-temporal context (Benhamou, 1993, 1996). In a study on induced resistance: a new strategy for plant defense against pathogens Benhamou and Picard, (2000) had identified the process associated with the induction of resistance in plants, the effect of biological, microbiological elicitors and chemical effects on the cellular response of plants to a pathogenic attack.

This study indicated that pathogens produce toxic substances (toxins, enzymes, antibiotics, etc.) and, in return, the plant is capable of responding to aggression by synthesizing a variety of molecules with complementary biological functions. Contribute to the protection of tissues and organs. Thus, these authors had recommended, although there are only few examples of practical application of induced resistance as a method of combating plant diseases, the results obtained from some experiments carried out in full field and greenhouse are encouraging and that this approach has the potential to become an effective and sustainable strategy for combating a range of pathogens.

Research by Ondh *et al.* (2021) on the variability of sensitivity to Banana Bunchy Top Virus (BBTV) of *Musa* spp. species, cultivated in Greater Libreville, reports that the diversity of local cultivars of banana plants can be an asset in the search for varieties resistant to BBTV. The results of this study showed that not all cultivars have the same sensitivity. Dessert cultivars are more susceptible than plantain cultivars. Still Ondh *et al.* (2020) in another study on "the prevalence of Banana Bunchy Top Disease (BBTD) in the Ntoum zone in Gabon" recommended that researchers evaluate the varietal resistance of *Musa* spp. And to show the influence of biotic and abiotic factors on the spread of this disease.

In 1990, a screening trial for BBTV resistance was initiated in RWANDA in an area of high infection. The planting material (30 different clones) was vitro plants from INIBAP/IRAZ. The suckers of locally recognized high-yielding varieties (10 varieties) were planted at the same time as the vitro plants to serve as a source of infection, that is to say that a local variety returned each time after 2 clones of the 'INIBAP. Each variety was planted in 2 rows. A line comprised 8 plants at a spacing of 3 m x 3 m. Observations focused on the date of appearance of BBTV symptoms, the percentage of plants attacked, the cycle and the yield of unattacked plants. In the 1st cycle, 40% of varieties did not show symptoms of the virus. For the two cycles combined, only 25% of varieties or cultivars, including Gros Michel, were not attacked by BBTV (CIRAD-FLHOR, 1996). Furthermore, the study by Bola (2013) on the sanitation test of banana plants (Libanga likale, Litete) infected with BBTV by in vitro culture had shown that after in vitro culture (proliferation of meristematic buds and regeneration) of virus-infected banana plants, on the samples analyzed, 75% of Litete cultivar and 66.7% of Libanga likale were found to be free from the virus. On the other hand, 25% of the Litete cultivar and 33.3% of the Libanga likale cultivar showed the persistence of the virus by always being positive in the TAS ELISA.

Indeed, the study by Shadari (2022) on the spatial distribution of Banana Bunchy Top Disease (BBTD) in the different banana production areas of the Maniema Province. The study showed that, regarding the growing conditions; bananas are grown 100% in association with rice, cassava, corn or other crops; 95.24% of farmers use waste from neighbors' fields; 90.48% of farmers use waste from plantations located far from their areas and 52.38% of farmers use waste from the same plantation. The incidence of BBTD varied relatively between 78.52% and 88.15% throughout the study area; discoloration of leaves with normal size was the most frequent and highest symptom on all banana trees, i.e. 31.9%. The *Pentalonia* vector. *Nigronervosa* is widely distributed in Maniema with an average rate of 94.85%. A large colony with winged insects is the most remarkable form, on average with 40.31%. Multivariate factorial analyzes made it possible to classify in each territory of the Maniema Province, the

different groups of banana cultivars sensitive and less sensitive to BBTV. The groups made up of less sensitive cultivars were characterized by the absence of symptoms (zero incidence and severity) but, generally with the presence and importance of the vector (*P. nigronervosa*) of level 2 and 3 of the aphid rating scale.

However, despite all the progress made since then, the problem of viral diseases still contains many unknowns, mainly with regard to the means of control. This investigation aimed to determine the tolerance of banana and plantain cultivars to the epidemiology of the bushy top disease of banana trees, cultivated in the open field. The study allows us to know the level of sensitivity and tolerance of each cultivar studied in relation to the condition caused by BBTV.

#### MATERIAL AND METHODS

#### Framework of the Study

The study was carried out in the Democratic Republic of Congo, Maniema Province more precisely in the territory of Kailo, community-chiefdom of Bangengele, at PK17 Lokando axis, Kimbembe Village at an altitude of 500 mr. The geographical location of the experimental site was that of the Maniema Province. Located in the Center - East of DR Congo, the Province extends between 0° and 5° South latitude and 24° 55' and 28° 8' longitude, it is limited to the East by the North-Eastern provinces. Kivu and South Kivu, to the North by the Province of Tshopo, to the West by the Province of Sankuru and to the South by the Provinces of Lomami and Tanganyika. Its total area is 132,520 km2 or 5.6% of the total area of the country (Shadari *et al.*, 2023, Makondambuta, 1997).

Figure 1 below shows the map of the experimental site.



Figure 1: Descriptive map of the experimental site in Maniema Province, DR Congo

# Climate

The climate that prevailed throughout the test period was that of the province of Maniema. A hot and humid tropical climate of the AW type according to the Köppen and Martonne classification. The latter evolves towards the equatorial type (AW1) in the North and towards the Sudanese type (AW4) in the South. In these two trends, the duration of the dry

season is 2 to 3 months and 3 to 4 months respectively. It includes two major seasons; the dry season and the rainy season. The average annual temperature is 27°C and annual precipitation amounts to 1650 mm of water per year (Makondambuta, 1997).

### Soil

The soil of Maniema varies from sandy type (Kibombo) to compact clay type (Pangi and Kasongo). Depending on the parent rock, we distinguish three types of soil: sedimentary with soft, coarse and red sandstone, clay and shales; metamorphic with silicified limestone, quartozitz, gneiss and metamorphic rocks; eruptive with eruptive granitic rocks (PAPRBIO, 1999). It should be noted the presence of ferralsol of the Yangambi type which is identified in the North of Lubutu (Makondambuta, 1997; CTCPM, 2002).

# Vegetation

The experiment was carried out in a secondary forest, the previous crops identified were essentially made up of species such as: Pandamus musanga, Elaïs guineinsis. We also noted the presence of trees of the cycadaceae family and other species of branched and non-branched plants.

# Material

# **Plant** material

The material used as part of this experiment consists of banana and plantain cultivars collected in the 7 territories of the Maniema Province considered less sensitive to banana bushy top disease in accordance with the results of Shadari *et al.*, (2023); they are: Tala lola (Mbudi: AAB), Leese (Mukokoonde: AAB), Nguku (Kangindi: AAB), Kamboloso (Kakumbi: AAB), Litete (Bondjilo: AAB), Lokusu (Mbole: AAB), Losakala (Mudioko: AAB), Libanga likale (Kileka: AAB), Andula (Akondo: AAB) for plantains, Kamaramasenge (Kamera: AAA) and Yangambi Km 5 (IBOTA: AAA) for bananas (Dowiya *et al.*, 2009, Adheka *et al.*, 2018).

# Technical equipment

We used as technical equipment among others the following instruments: machete (it was used to clear and cut grass during field experimental activities); hoe (this instrument we used to do superficial plowing, stump removal, digging, and weeding); magnifying glass (this device helped us visualize aphids on leaf sheaths and banana leaves); tape measurer (allowed us to take different measurements of the field and banana plants when collecting data in the field); twine (allowed us to trace stakes and planting lines); scale (also used to weigh the weight of harvest products) and GPS (helped us to take the geographical coordinates of the experimental site in order to develop the map of said site).

### Methods

### **Experimental design**

A single factorial design of elongated plots with 11 repetitions was used. The spacings chosen were 3 m x 3 m, or a density of 121 feet. The field had a quadratic shape of 40 m x 40 m and plots 3 m wide by 33 m long. The total area of the field was 1600 m2. Indeed, the cultivar Kamaramasenge (Kamera AAB) was used because of its high sensitivity as a reference individual. On the other hand, the inoculation technique by aphid sting was applied. Hence, the disease was transmitted from one plant to another by the bites of aphids (Pentalonia nigronervosa) taken from diseased plants showing symptoms of level 4 to 5 of the BBTD rating scale (Shadari *et al.*, 2023).

# Collection of BBTV epidemiological data by TAS ELISA in banana and plantain cultivars

Enzyme-linked immunosorbent assay (ELISA) is a powerful and widely used molecular technique to detect and quantify the interaction between antigens and antibodies. It plays a crucial role in clinical diagnosis, research and various fields of biotechnology. The Enzyme Linked Immuno Sorbent Assay (ELISA) procedure involves several key steps to detect and quantify the antigen of interest in a sample. The following describes the general procedure for a direct ELISA test:

**1. Addition of sample:** The first step is to add the sample, which contains the antigen, into the wells of a microtiter plate. The plate is covered with a solid surface that allows the antigen to adsorb or bind to it. Antigen adsorption: Antigens present in the sample adsorb to the surface of the well through nonspecific interactions. The plate is then incubated to ensure sufficient binding between the antigens and the plate surface. Washing: After the incubation period, the plate is washed to remove any unbound or non-specifically bound antigens. This washing step reduces background noise and increases the specificity of the assay.

**2. Addition of enzyme-linked antibodies:** Next, an enzyme-linked antibody specific to the target antigen is added to the wells. This antibody is typically conjugated to an enzyme, such as peroxidase or alkaline phosphatase. The antibody binds specifically to antigens immobilized on the plate, forming an antigen-antibody complex. Washing: Similar to the previous step, the plate is washed again to remove any unbound or non-specifically bound antibodies. This washing step is crucial to remove any excess or unwanted antibodies that could interfere with the accuracy of the test.

**3.** Addition of chromogenic substrate: A chromogenic substrate specific for the enzyme linked to the antibody is added to the wells. This substrate undergoes a reaction with the enzyme, resulting in a color change. The reaction typically involves the conversion of a colorless substrate into a colored product.

**4. Visualization and interpretation of results:** The last step is to visualize the color change or the formation of a colored product. This can be done by visually observing the wells or by using a spectrophotometer to measure the absorbance at a specific wavelength. Color intensity or absorbance is proportional to the amount of antigen present in the sample. The results are then interpreted based on a standard curve or known concentrations of the antigen to determine the concentration of the antigen in the sample. Indeed, our TAS ELISA analyzes were carried out at the IITA-KALAMBO laboratory in BUKAVU, DR Congo.

# Presence of tolerant banana and plantain cultivars at BBTD

During their cycles, knowledge on tolerance was gained by comparing production with the different disease parameters below.

- 1. Impact % =  $\frac{\text{Number of infected clumps}}{\pi} \times 100$ 
  - Total number of tufts
- 2. Severity: The rating scale (Cialca, 2008) which was used to assess the severity of banana viral disease Bunchy top of banana tree is as follows:
  - Level 0: without symptoms,
  - Level 1: streaks on the leaves,
  - Level 2: dark streaks up to the pseudo-trunk,
  - Level 3: discoloration of leaves with normal size,
  - Level 4: reduced size of discolored leaves,
  - Level 5: bushy appearance at the top "Bunchy top".
- 3. Presence of the Aphid: The black banana aphid *P. nigreonervosa* is located between the old leaf sheath and the young leaf sheath. It can even be located on leaves, cigars and fruits. To observe the presence of aphids, the banana trees in each clump were inspected using a magnifying glass. The observation consisted of pulling out the false trunk of the

banana tree to visualize this aphid. The presence of the aphid Pentalonia nigronervosa was assessed from the ratio between the total number of tufts carrying aphids over all the tufts observed and expressed as a percentage.

- 4. Importance of the Aphid: As the different colonies of *P. nigronervosa* are generally located between the sheath of the most recent leaf and the old sheath, we observed the different colonies of this aphid on the different organs of the banana tree. The different forms of aphid (larvae, nymphs, alates or adults) forming the different colonies were observed on the cigar, the unfolded leaves and the leaf sheaths of the banana tree.
- 5. Vector importance on selected clumps was rated using the BBTD scale ranging from 0 to 5 with higher scores indicating increased *P. nigronervosa* aphid populations. The rating scale for *P. nigronervosa* aphids is as follows (Cialca, 2008; Ngama *et al.*, 2014):
  - Level 0: without vector on the tuft,
  - Level 1: a simple colony without winged insects,
  - Level 2: several simple colonies,
  - Level 3: a large colony with winged insects,
  - Level 4: several large colonies,
  - Level 5: widespread colonies on the leaves and pseudostem.

Indeed, to correlate the level of production and degree of sensitivity of different cultivars of bananas and plantains to banana bushy top disease and to give the conclusion the tolerance of the cultivars, we used the general scale for evaluating the viral diseases. However, Table 1 below presents the above-mentioned scale.

| Level | Symptom      | Impact (%) | Yield        | Category       |
|-------|--------------|------------|--------------|----------------|
| 1     | Absence      | 0          | Excellent    | Resistant      |
| 2     | Doubtful     | 1 to 10    |              |                |
| 3     | Weak         | 11 to 25   | Good         | Tolerant       |
| 4     | Moderate     | 26 to 40   |              |                |
| 5     | Intermediate | 41 to 60   | Intermediate | Sensitive      |
| 6     | General      | 61 to 75   |              |                |
| 7     | Intense      | 76 to 90   | Poor         | Very sensitive |
| 8     | Severe       | 91 to 99   |              |                |
| 9     | Death        | 100        | Very poor    | Very serious   |

Table 1: General scale for assessing viral diseases (CIAT, 1987)

# Statistical analysis

The data obtained were analyzed by multivariate methods. The factorial method allowed us to make the spatial distribution of the cultivars, while the cluster method allowed us to classify the cultivars into groups according to their susceptibility to banana bushy top disease using the STATGRAPHICS\_Centurion\_XVI version 16.20 software. 04.

# RESULTS

# Epidemiological Parameters and Virus Detection in Banana and Plantain Cultivars by TAS ELISA

The results which relate to the epidemiological parameters, notably the incidence and severity of BBTD, the presence and importance of the vector (*Pentalonia nigronervosa*) as well as the enzyme-linked immunosorbent assay (TAS ELISA) are presented in Tables 2 and 3 and as in Figure 2 below.

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| FACTORS |   |  |  |
|---------|---|--|--|
| FI (%)  | FII (%)   |  |  |
| 2,44    | 1,27  |  |  |
| 48,83   | 25,56   |  |  |
| 48,30*  | 74,399*   |  |  |
| 0,81    | -0,526406   |  |  |
| 0,82    | -0,503195   |  |  |
| 0,60147 | 0,708531  |  |  |
| 0,8     | 0,476495  |  |  |
| 0,27    | 0,138084  |  |  |
|         | FI (%)           2,44           48,83           48,30*           0,81           0,82           0,60147           0,8           0,27 |  |  |

# Table 2: Contribution of factors to the observed variance and their correlation to variables (incidence, severity of BBTD, presence, vector importance and TAS ELISA

Legend: FI: Factor 1; FII: Factor 2; \*: Accumulation of two factors (cultivars and fields)

In view of the results recorded in Table 2, it emerges that the banana and plantain cultivars were extracted with a variance of 74, 39% where positively the relative variance was 48, 83% to factor 1 and of 25, 56% to factor 2 on the dispersion of cultivars according to their degree of incidence, severity, presence, importance of the vector (*P. nigronervosa*) and confirmation of the virus by the TAS ELISA. Indeed, graph 2 below illustrates the spatial distribution of banana and plantain cultivars according to their sensitivity.



Figure 2: Spatial distribution of cultivar plants according to sensitivity to BBTD Legend: Dotted yellow: cultivar Kamaramasenge; Dotted in purple color: Kamboloso cultivar; Dotted black: cultivar Kamaramasenge; Dotted in dark red color: cultivar Leese; Dotted green: Andula cultivar; Dotted blue-purple color: Lokoka cultivar; Dotted in yellow-orange color: Lokusu cultivar; Dotted blue-green color: Losakala cultivar; Dotted in pink color: cultivar Yangambi Km 5; Dotted in light green color: Nguku cultivar; Dotted gray color: cultivar Tala lola

It appears from Figure 2 that the relative variance subdivided by two axes of factor I with (48, 83%) and factor II with (25, 56%) spatially distributed the banana and plantain cultivars according to their sensitivity in 4 groups. As a result, cultivars dispersed in negative zones are less and less susceptible to BBTD and those dispersed in positive zones are increasingly susceptible to the disease. However, Table 3 below presents the different groups of cultivars tested according to their degree of sensitivity to banana bushy top disease.

|           |        |           | Qualitative variables |                  |                              |                                |              |  |
|-----------|--------|-----------|-----------------------|------------------|------------------------------|--------------------------------|--------------|--|
| Groups    | Number | Frequency | Incidence<br>of BBTD  | Severity of BBTD | Presence<br>of the<br>vector | Importance<br>of the<br>vector | TAS<br>ELISA |  |
| Group I   | 30     | 24,79     | 0                     | 0                | 0                            | 0                              | 0            |  |
| Group II  | 48     | 39,67     | 0                     | 0                | 1                            | 1                              | 0            |  |
| Group III | 28     | 23,14     | 1                     | 3                | 1                            | 2                              | 0            |  |
| Group IV  | 15     | 12,4      | 0                     | 0                | 1                            | 2                              | 1            |  |

# Table 3: Groups of banana and plantain cultivars and their level of sensitivity to BBTD

Legend: BBTD: Banana Bunchy Top Disease; TAS ELISA: Triple Antibody Sandwish Enzyme Linked Immuno Sorbant Assay

Indeed, with regard to the results recorded in Table 3, it emerges that 30 plants of banana and plantain cultivars out of 121 plants, or 24.79%, are classified in group I and are characterized by plants which have not presented symptoms of BBTD (zero incidence and severity), absence of aphid colonies (Pentalonia nigronervosa) and revealed negative by TAS ELISA. On the other hand, 48 plants of banana and plantain cultivars out of 121, or 39.67%, are classified in group II and are characterized by plants not showing symptoms of the disease but with the presence and importance of the vector (P. nigronervosa) of the disease. Level 1 (a single colony without winged insects) of the aphid rating scale and TAS ELISA negative. While, 28 plants of banana and plantain cultivars out of 121 plants, or 23.14%, are classified in group III and characterized by plants of cultivars presenting disease symptoms with severity of level 3 (discoloration of leaves with pruning normal) of the BBTD rating scale and with presence, importance of aphid (P. nigronervosa) of level 2 (several simple colonies) and revealed negative in the TAS ELISA. Finally, it is observed that 15 plants of banana and plantain cultivars out of 121 plants, or 12.4%, are placed in group IV and are characterized by the plants having not presented the symptoms of BBTD, but with the presence and importance of aphid (P. nigronervosa) of level 2 (several single colonies) and revealed positive by TAS ELISA.

# Tolerance of Banana and Plantain Cultivars in Relation to Their Production Level and the Incidence of BBTD

The results consistent with the tolerance, resistance or sensitivity of banana and plantain cultivars tested in the open field, as well as the different variables are presented in Table 4 below.

| banana and plantain cultivars in relation to production |            |           |          |        |               |              |           |  |
|---|------------|-----------|----------|--------|---------------|--------------|-----------|--|
| Cultivars   | Vernacular | Genotypes | Presence | Impact | Severity with | Yield        | Category  |  |
|   | names      |           | of the   | (%)    | TAS ELISA     |              |           |  |
|   |            |           | vector   |        | (%)           |              |           |  |
| Libanga likale  | Kileka     | AAB       | 45,5     | 18,2   | 27,3          | Good         | Tolerant  |  |
| Kamboloso   | Kakumbi    | AAB       | 54,5     | 18,2   | 9,1           | Excellent    | Resistant |  |
| Kamaramasenge   | Kamera     | AAB       | 81,8     | 72,7   | 72,7          | Intermediate | Sensitive |  |
| Leese   | Mukokoonde | AAB       | 90,9     | 36,4   | 36,4          | Good         | Tolerant  |  |
| Andula  | Akondo     | AAB       | 90,9     | 27,3   | 36,4          | Good         | Tolerant  |  |
| Lokoka  | Bonjilo    | AAB       | 100      | 45,5   | 9,1           | Excellent    | Resistant |  |
| Lokusu  | Mbole      | AAB       | 90,9     | 18,2   | 18,2          | Good         | Tolerant  |  |

 Table 4: Expression of the disease and significance of tolerance and/or resistance of banana and plantain cultivars in relation to production

| Losakala      | Mudioko         | AAB | 81,8  | 9,1   | 27,3  | Good      | Tolerant  |
|---------------|-----------------|-----|-------|-------|-------|-----------|-----------|
| Yangambi Km 5 | Ibota           | AAA | 90,9  | 0     | 0     | Excellent | Resistant |
| Nguku         | Kangindi        | AAB | 72,7  | 45,5  | 27,3  | Good      | Tolerant  |
| Tala lola     | Mbudi           | AAB | 36,4  | 9,1   | 9,1   | Excellent | Resistant |
|               | General average |     | 76,03 | 27,29 | 24,81 |           |           |

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Legend: %: percentage; AAA: triploid genome of Musa acuminata; AAB: triploid genome of Musa acuminata and Balbisiana

The results shown in Table 4 show that the presence of aphids (Pentalonia nigronervosa) on all cultivars was 76.03%; while, the incidence of the disease was 27.29% on all cultivars and the average severity tested by the TAS ELISA in all cultivars was also 24.80% of plants carrying the disease. Banana bunchy top virus. Indeed, the cultivars Kamboloso, Lokoka, Yangambi Km 5 and Tala lola achieved yields described as "excellent" and are said to be resistant to banana bunchy top virus because their TAS ELISA severities varied between 0-10% of the scale. General assessment of viral diseases. However, the cultivars Libanga likale, Leese, Andula, Lokusu, Losakala and Nguku obtained yields qualified as "good" and are said to be tolerant to BBTV, given their severities varied between 11-40% of the general evaluation scale. Viral diseases. On the other hand, the Kamaramasenge cultivar achieved an intermediate yield and is said to be a cultivar sensitive to BBTV given its severity which oscillated between 41-75% of the general assessment scale for viral diseases.

### DISCUSSION

The analysis of epidemiological parameters, notably the incidence of BBTD, the severity, the presence and importance of the vector (Pentalonia nigronervosa) as well as the enzymelinked immunosorbent assay (ELISA) were important in allowing us to adopt the varietal control method; by choosing banana and plantain cultivars less susceptible to banana bunchy top viral disease. We found cultivars tolerant, resistant and sensitive to BBTD, among bananas and plantains tested and evaluated in the field. Indeed, the cultivars Kamboloso, Lokoka, Yangambi Km 5 and Tala lola are "resistant" to the banana bunchy top viral disease in accordance with the general scale for evaluating viral diseases because they have achieved "excellent" yields with an incidence rate varying between 0 to 10%, on the other hand, the cultivars Libanga likale, Leese, Andula, Lokusu, Losakala and Nguku are tolerant to BBTD given that they obtained "good" yields with an incidence rate varying between 11 to 40% and finally, the Kamaramasenge cultivar is sensitive to BBTD because it achieved an "intermediate" yield with a rate of incidence varying between 41 to 75% of the general assessment scale for viral diseases. Indeed, the high sensitivity observed in the Kamaramasenge (AAA) cultivar is not surprising and is justified by the results of Ondh et al. (2021) who observed that in general, the incidence of BBTV is higher in dessert bananas than in plantains. Indeed, we found plants with a total absence of the vector (Pentalonia nigronervosa) among the cultivars. This is justified by the removal of dead leaves from the pseudo-trunk. These results could also be justified in the sense that other cultivars have their leaf sheaths tightly rolled over the others, leaving no free access for the vector to settle there. We have seen on the one hand that there have been banana cultivars without symptoms of BBTD with the presence of the vector but tested negative for bunchy top virus. On the other hand, we found cultivar plants with apparent symptoms and presence of the vector, however, tested negative for the virus by ELISA. Furthermore, we also encountered in certain cultivar plants the absence of symptoms with the presence of the vector and carriers of the virus in the TAS ELISA; this phenomenon called "holy carrier". Indeed, these different results consolidate those of subsequent studies on bananas and plantains. Plants may be completely resistant to some

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pathotypes of the pathogen and susceptible to others. The reaction which does not allow the attack is qualified as incompatible; it is specific to both partners and is described as vertical or even mono genic, reports Van Der Plank (1968). It is total resistance because plants that have this type of resistance do not show any symptoms of the disease. The genotypic diversity of these different cultivars could explain the difference in varietal sensitivity from one cultivar to another. Indeed, all plantain cultivars have the same genotype (AAB). Dessert bananas, on the other hand, have three (5) different genotypes (AAA), (AAB), (ABB), (AB) and (AA) (Nzawele, 2012). This study clearly shows that there is indeed an "inoculum effect by aphid bite" on the banana and plantain cultivars studied given the presence of the observed symptoms and viruses in the plant tissues tested by the TAS ELISA. However, the observed phenomenon of the presence of aphids, absence of symptoms and vice versa is added to the series of results of the authors below: Qazi (2016) reports that vectors can quietly inoculate viruses which will replicate for a short period, at the point of aphid inoculation. Symptoms usually appear on the second emerging leaf after inoculation in accordance with the observations of Caruana (2003), and of Busogoro et al. (2009). This rapid replication of the virus allows symptoms to appear immediately over time. Symptomatic levels will therefore evolve quickly when environmental conditions are favorable. The importance of level 1 aphids can lead to the appearance of all the other visual symptoms of BBTD. Pentalonia nigronervosa is the only insect vector species known to date (Thomas et al., 2003). It needs to feed on an infected plant to acquire viral particles (Robson et al., 2007; Mukwa et al., 2014). An aphid feeding time of at least a few hours on an infected plant is necessary to effectively acquire the virus. This time is followed by a latency period during which the virus circulates through the body of the aphid to the salivary glands (Adegbola et al., 2013). The aphid then becomes infectious and capable of transmitting the virus for the rest of its life (Caruana, 2003). Thus, whatever the number of aphids or the number of aphid colonies present on the plant, it is enough that there is at least one aphid vector of BBTV for the plant to show the symptoms of the disease. Visual symptom level 5 is always associated with the highest levels of aphid importance. Indeed, at optimal temperatures of 28°C, P. nigronervosa reproduces at a rate of 25 to 30 generations per year, according to the work of Anhalt and Almeida (2008). Aphids generally occur in colonies located between the sheath of the newest leaf and the central vein. The alatae or winged forms appear after 7 to 10 generations of wingless (wingless). Generally speaking, wings appear when the numbers inside the colonies increase significantly, or when the nutritional quality of the plant deteriorates (aging or yellowing). This situation justifies the fact that the importance of level 3 aphids only appears when the plant presents the visual symptoms of levels 4 and 5, in accordance with the results obtained by Nelson (2004).

### CONCLUSION

This study aimed to evaluate the expression of tolerance of banana and plantain cultivars in the face of the epidemiology of the bushy top disease of banana trees, cultivated in the open field. Specifically, know the level of sensitivity and tolerance of each cultivar studied in relation to the condition caused by BBTV. To achieve the objectives, methods relating to the latter were used. A single factorial design of elongated plots with 11 repetitions was set up. The spacings chosen were 3 m x 3 m, i.e. a density of 121 feet in a field of 40 m x 40 m. The results obtained showed that: the TAS ELISA makes it possible to evaluate the epidemiological expression of BBTV in bananas and plantains.

It demonstrated that in the presence of the vector (Pentalonia nigronervosa) with or without symptoms of BBTD (24.28%) plants of cultivars studied were carriers of the banana bunchy top virus. There is a strong correlation between plant-vector-disease-production, that is to say; plants carrying aphid colonies are likely to be more infected by BBTV and cause significant yield losses. The level of production of banana and plantain trees infected by BBTV,

the incidence rate and severity of the disease makes it possible to confirm the tolerance, resistance and sensitivity of banana and plantain cultivars to the banana bunchy top virus. Help of the general assessment scale for viral diseases.

Indeed, the production correlation with the severity of each cultivar studied made it possible to discover the resistance of certain cultivars considered tolerant to BBTV. However, the cultivars Kamboloso, Lokoka, Yangambi Km 5 and Tala lola having achieved "excellent" yields are said to be resistant to bunchy top disease, their severity in the TAS ELISA oscillated between 0-10% of the general scale. Assessment of viral diseases. Furthermore, the cultivars Libanga likale, Leese, Andula, Lokusu, Losakala and Nguku are tolerant to BBTD given that they obtained "good" yields and their ELISA severity level evolved between 11-40% and finally, the Kamaramasenge cultivar is sensitive to BBTD because it achieved an intermediate yield and a severity level varying between 41-75% of the general scale assessment of viral diseases. This study clearly confirms that there is a positive effect of aphid bite inoculum on virus transmission in banana and plantain cultivars grown in the open field.

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