***Original Research Article***

**Different morphotypes of *Fusarium oxysporum* isolated from *Xanthosoma sagittifolium* L. Schott roots: Action of ethanol leaf extracts of *Psidium guajava* on *in vitro* inhibition and on *X. sagittifolium* plants inoculated with *F. oxysporum***

Comment [A1]: Must changed to be more suitable and describe the content of the paper


**Abstract**

The objective of this study was to determine the different morphotypes of *F. oxysporum* present in the root of *X. sagittifolium* and evaluate the effect of alcoholic extracts of *Psidium guajava* on their *in vitro* inhibition. Strains of *Fusarium oxysporum* were collected in eight localities where *X. sagittifolium* is grown. *Fusarium* strains were isolated from roots of *X. sagittifolium* harvested in each locality on PDA media in the laboratory. The antifungal test using alcoholic extracts of *P. guajava* at 30 and 60% and the virulence test of each strain on young plants of *X. sagittifolium* aged three months were realizing. Eight strain of *Fusarium oxysporum* were successfully isolated. After maximum growth, five morphological types were observed (pionnotal, sclerotic, clowny, cottony and ras senescent). The cottony strain was abundant and present in all the locality. Histological analysis of the different strains obtained revealed the presence of septate or siphoned hyphae and three types of conidia (microconidia, macroconidia and sporangiospores or chlamidospores). The inhibition tests were very high with 60% of ethanol extract of *P. guajava*. 83.33% of inhibitory effect were observed after eight days of growth, in the strains collected in *X. sagittifolium* roots, in $L_3$ (Loum) and $L_4$ (Bangoua) localities. After infection of *X. sagittifolium* plants with each strain of *F. oxysporum* isolated, symptoms observed were yellowing and wilting of leaves. However, plants inoculated with the $L_3$ (Loum) strain showed both yellowing and wilting of leaves. These results obtained showed that *F. oxysporum* is not only saprophytic fungi, it's also able to induce yellowing and wilting of leaves in *X. sagittifolium*.


Comment [A2]: Explain the main objective, major methods and important results.


**Keys words**: *Fusarium oxysporum*, *Xanthosoma sagittifolium*, *P. guajava*, inhibition, antifungal test.


**INTRODUCTION**

Comment [A3]: rewriting with academic pattern (needs to be revised by a professional English speaker)

*Fusarium oxysporum* (Nectriaceae) is an ascomycete telluric, ubiquitous and plant parasitic fungi that collectively infect different hosts plant. The presence of this ascomycete in the soil leads to significant production losses in many crops such as *Musa acuminate* (Poon *et al*, 2020), *Gossypium hirsutum* (Asran et al., 2018), *Cucumis melo* (Dhaouadi *et al.*, 2019), *Lycopersicon esculentum* (Neela *et al*. (2014), *Solanum tuberosum* (Trabelsi et al. 2016; Upadhaya *et al.*, 2020). This soil-borne pathogen also has a great influence on the quality of plant seeds (Farshid *et al.* 2019). The visible symptoms due to the presence of *F. oxysporum* include yellowing of the leaves, wilting of tissue and vascular lesions thus causing death of the plant (Zhang *et al.*, 2008). Xin and Cun, (2020), showed that *F. oxysporum* are responsible of damping-off in *Pinus massoniana* plants.

This endophytic plant fungus can remain as dormant mycelium or chlamydospores without causing disease (Antônia and Menezes, 2006), virulent or non-virulent depending on the host species they infect (Van Der Does *et al*., 2008; Michielse and Rep, 2009). This may be why Pacumbaba *et al.* (1992), noted the presence of *Fusarium solani* and *Rhizoctonia solani* during root rot of *Xanthosoma sagittifolium* L. Schott caused by *Pythium myriotylum*. Hence, it would be good to know if, *Fusarium* species, actes as a saprophyte that benefits from the aggressiveness of *Pythium myriotylum* vis-à-vis the roots of *X. sagittifolium* plants, or remains in a state of latency due to the action of *P. myriotylum*. Could *Fusarium oxysporum* be an aggressive pathogen for *Xanthosoma sagittifolium* plants? Or does it remain only a profiteer during root rot during *X. sagittifolium – P. myriotylum*.

In plants species where *Fusarium* are absolute pathogens, some authors have shown that their action could be inhibited by medicinal plant extracts. Plant extracts are good efficient against the phytopathogenic fungi, due to the synergistic antimicrobial, antifungal, antibacterial activity of their various phytochemical constituents (Gahukar, 2012; Neela et *al*., 2014; Baka and Rashad, 2016; Abdulaziz *et al*., 2018). It is noted that aqueous extracts of *Curcuma longa* can be used against root rot caused by *Fusarium solani* in *Helianthus annuus* L. (Abdulaziz et *al*., 2018) and against *Fusarium oxysporum* (Chen et *al*., 2018). The antifungal activity of ethanol and acetone extract of leaves of *Piper betel*, *Lowsonia inermis*, *Psidium guajava*, *Carica papaya*, *Moringa oleifera*, *Mimosa pudica*, *Catharanthus roseus*, *Adhatoda vasica* and *Andrographis paniculata* have been effective against *Fusarium oxysporum* the causal agent of *Fusarium* wilt in tomato (*Lycopersicon esculentum* Mill.) Neela *et al.* (2014). Despite the type of solvent used, the antimicrobial activities of plants against a number of plant pathogens are linked to their richness in bioactive compounds. These compounds not only have an impact on the growth of the mycelium of the fungi, but

they also influence the production and sporulation of spores (Xoca-Orozco et *al*., 2018). Many studies show the remarkable presence of bioactive compound in medicinal plants (Dissanayake et al., 2014; Neela *et al.,* 2014; Abdulaziz et *al*., 2018 and Chen et *al*., 2018)

As a traditional medicinal plant, *Psidium guajava* L. (Myrtaceae), has received much attention for producing many complex compounds. The therapeutic properties of *P. Guajava* include insecticidal (Salah and Saadiya, 2017), antimicrobial (Kemegne et al, 2018, Mohammed *et al.,* 2017; Okechukwu *et al.,* 2012), antifungal (Vijayakumar *et al*., 2018), antiviral (Sobral – Souzaab *et al.*, 2019) and antioxidant properties (Manikandan1and Vijaya, 2015). Theses antioxidant and antifungal activities of leaf extract of *P. Guajava* against *Fusarium* sp, *Alterneria* sp. and *Colletotrichum* sp have been shown by Akanji *et al.*, 2009 and Mandal *et al*., 2010 studies. More, aqueous and ethanol leaf extract of *Psidium guajava* L. have shown their efficacy against *in vitro* inhibition of *Pythium myriotylum* Drechsl, main causative agent of root rot disease in *X. sagittifolium*.

So to understand if, *Fusarium oxysporum*, can be pathogenic or saprophytic organism to *X. sagittifolium* roots, this study aims to determine the different morphotypes present in the roots of *X. sagittifolium*, to evaluate the effect of ethanol extracts of *Psidium guajava* leaves on the *in vitro* inhibition of the fungal strains, this during the interaction *F. oxysporum* - *X. sagittifolium*.

## MATERIAL AND METHODS

Comment [A4]: rewriting with more scientific method and attraction

### Harvesting, isolation, cultivation and purification of *Fusarium oxysporum* strains of the *X. sagittifolium* roots

The harvest roots of *X. sagittifolium* used to isolate *F. oxysporum* strains was carried out between October and November (at the end of the rainy season and the beginning of the dry season), in *X. sagittifolium* plants with yellow leaves and infected roots (dark in color and having a mole end). This harvest of the X. sagittifolium plant roots was carried out in polyculture farms of *X. sagittifolium* and other food crops in eight localities (L). $L_1$ (Soa), $L_2$ (Banda), $L_3$ (Loum) $L_4$ (Bangoua), $L_5$ (Abong-Mbang), $L_6$ (Bansoa), $L_7$ (Santa) and $L_8$ (Ekona). In each harvest locality, three repetitions were carried out and the collected samples were mixed. Roots of *X. sagittifolium* collected were properly washed with running tap water, air-dried for a few minutes, cut into small pieces (1cm), soaked in sterilized water, with 1% sodium hypochlorite (NaClO) for 3 min and then rinsed with distilled water as described by Tongon and Soytong (2016). All of the small pieces of roots were transferred onto water agar (WA) medium in oder to first observe the appearance of colonies (Cao *et al.,* 2002) then sub-

cultured on PDA mixed with 200 mg/L of Streptomycin to prevent bacterial proliferation (Si Mohammed *et al*., 2016), until pure culture were obtained.

**Production, observation and identification of spores of *Fusarium oxysporum***

This production of *F. oxysporum* spores was carried out by thermal shock according to the protocol of (Widmer and Laurent, 2006). The mycelium of each fungal strain was collected according to the method of Dohou et al., (2004). After seven days of culture, the mycelia were homogenized in 50 ml of ice cold sterile distilled water for 10 to 15 min. This mixture was then placed in the dark for the release and germination of the spores. Morphological identification was done by observation of the fungal characteristic under binocular optical microscope (IVYMEN mark) at 400X. The isolates obtained were identified using the synoptic keys (Nelson *et al.*, 1983), dichotomous keys (Booth, 1971) and tabular keys (Burgess *et al*., 1988). These keys are based on the examination of the morphological characters of asexual structures.

**Evaluation of the effect of ethanol leaf extracts of *Psidium guajava* on the *in vitro* growth of *F. oxysporum***

The antifungal effect of the ethanol leaf extracts of *P. guajava* plants was evaluated using two concentrations (PDA + 30 and PDA + 60% of leaf extract). Inhibitory activity was evaluated with the poisoned food method (Das et *al*., 2010). Control plates consisted in PDA with ethanol. The culture of these *Fusarium* strains was carried out in the dark in a culture chamber at 25±1°C. Every 2 days, the mean radial diameter was measured. Growth inhibition relative to the control due to the efficacy of the ethanol leaf extract of *P. guajava* was calculated according to the following formula of Dissanayake (2014):

$$\text{Inhibition (\%)} = [(C - T)/C] \times 100$$

where, C and T represent the diameter of control and treated colony respectively. Here three replications were prepared for each treatment. Data on mycelial growth at 0,2,4,6 and 8 days after inoculation were recorded. To avoid bacterial contamination 0.5 g of antibacterial streptomycin was added to 1 L of PDA medium.

**Evaluation of the virulence of each strains of *F. oxysporum* on *X. sagittifolium***

*In vitro* of *X. sagittifolium* plants were obtained according to the protocol of Omokolo *et al.* (1995) and Djeuani *et al*. (2014). These vitroplants were transferred to pods containing a mixture of black earth, sand and wooden chips 2:1:1 (V/V) previously oven sterilized (mark

REPLEX) at 170°C for 24 hours for acclimation. After three months, for each strain of *F. oxysporum* isolated, 25 seedlings of *X. sagittifolium* were used. The inoculum was directly leached to the root zone through the soil. Each plant was inoculated with 5ml of the suspension containing $2x10^7$ cfu/ml concentration of each strain of *Fusarium oxysporum* according to the protocol of (Muhammed et *al.*, 2017). The plants were observed regularly for the appearance and development of disease symptoms. The severity of symptoms was scored on a scale ranging from 1 through 5: 1–No obvious symptoms, 2–Symptoms on 0-24% of leaves, 3–Symptoms on 25%- 50% of leaves, 4 –Symptoms on 51%-74% of leaves and 5–Symptoms on 75%-100% of leaves (Eni *et al*., 2008 modified) after 14 days.

**Evaluation of the effect of *P. guajava* ethanol leaf extracts on *X. sagittifolium* infected by *F. oxysporum***

*X. sagittifolium* plants used were divided into two groups: control plants, (treated only with the ethanol solution) and the plants previously inoculated with each strain of *F. oxysporum* isolated (treated with ethanol leaf extracts of *P. guajava*). In each treatment 25 *X. sagittifolium* plants aged three months were used. The *P. guajava* leaves were cut in small pieces; washed in distilled water; dried at 40°C for 48 hours in an oven (Barnstead/Lab-Line; USA; Model No. 121). The dried leaves were ground using a mortar and pestle into fine powder. 60g of *P. guajava* leaf powder were soaked in 100ml of 95% ethanol in a sterilized bottle and kept overnight at room temperature for 48h. The ethanol fraction was separated through sterilized Whatman filter paper (No.1). The supernatant collected constituted the ethanol extract of *P. guajava*. The extracts obtained were applied on aerial parts and the rhizosphere of each plant, using a hand sprayer until wetness of plants according to Yasser et *al*., (2017). Plants were treated twice time at day 7 and day 14 (for two weeks) and disease severity score evaluated at day 14 using method of Eni *et al*, (2008).

**Statistical analysis**

Results were expressed in the form of Means ± SD. All the statistical analysis was done using Microsoft excel. Duncan Multiple Range Test at 5 % significance was used for the comparative analyses of the results with the help of SPSS 20.0.

## RESULTS AND DISCUSSION

**Comment [A5]:** delete

## Results

### Morphological analysis of the different strains of *Fusarium oxysporum* isolated from *Xanthosoma sagittifolium*.

Comment [A6]: explain the relation between morphotypes of fungus and their pathogenicity of plants

Morphological analysis of the eight strains of *Fusarium oxysporum* collected in each locality shows that the growth is not the same for all strains after 8 days of *in vitro* culture (Fig.1). Strains collected in $L_1$ (Soa), $L_2$ (Banda), $L_3$ (Loum), $L_6$ (Bansoa) and $L_7$ (Santa), grew faster than those collected in $L_4$ (Bangoua), $L_5$ (Abong-Mbang) and $L_8$ (Ekona). the cottony or wooly form was present in all the harvesting locations (Table I). Similarly, a variation in the staining of the strains is visible and the most dominant is the white color (strains collected in $L_1$, $L_2$, $L_5$, and $L_8$) followed by the beige color compared to the pink and purplish pink (Table II). The pure culture showed that the cottony strain of hyphae is the most abundant in *F. oxysporum* in in all harvest areas.

### Structure of conidia of *F. oxysporum* strains

Microscopic observation of conidia of each strain of *Fusarium oxysporum* shows that zoospores differ from one strain to another (Fig. 2). Strains of $L_3$, $L_4$, $L_6$, and $L_7$ only have microconidia (nonseptate and monoseptate). Those of $L_1$, $L_2$, $L_5$, and $L_8$ showed two types of conidia, microconidia (nonseptate and monoseptate) and macroconidia (two-septate, three-septate and four-septate). The results also show that microconidia are most abundant in all cultivated *F. oxysporum* strains. Chlamydospores are also present in strains collected in $L_1$, $L_2$, and $L_5$.

### Effect of *P. guajava* ethanol leaf extracts on *in vitro* inhibition of radial growth of *F. oxysporum*

Comment [A7]: inhibition activity of ethanolic leaves extract of P. guajava on F. oxysporum growth in vitro

Inhibition tests of *F. oxysporum* with *Psidium guajava* ethanol leaf extract was significant at 30 and 60% compared to the control (Table III). This inhibition of growth is higher at 60%. The results show that radial growth of control is 9.0 cm, in strains obtained in $L_2$ (9.0 ± 0.00 cm), $L_3$ (9.0 ± 0.05 cm), $L_4$ (9.0 ± 0.02 cm), $L_5$ (9.0 ± 0.05 cm), $L_6$ (9.0 ± 0.04 cm), $L_7$ (9.0 ± 0.01 cm) and $L_8$ (9.0 ± 0.00 cm) after 8 days of growth (Table III). However, at 30%, at 30%, growth inhibition of strains of *F. oxysporum* is variable. Higher growth was obtained after eight days in *F. oxysporum* strains collected in $L_3$ (6.5 ± 0.03cm) and the highest record of inhibition was with strain collected in $L_2$ (73,36%) after 2 days. The results also show that the percentage of inhibition decreased over time. At 60%, the inhibition of radial growth is very high for all the strains. The low growth values were observed in *F. oxysporum* isolated from

*X. sagittifolium* of $L_4$ varying between day 2 and day 8, from 1.2 ± 0.04 to 1.5 ± 0.01 cm. This radial growth is constant in the strain harvested in $L_3$ (1.5 cm) (Table II). In the strain harvested at $L_5$, the inhibition of growth does not vary in the presence of 30 and 60% of ethanol leaves extract concentrations. This inhibition is 4.0 ± 0.02 and 4.0 ± 0.05 cm respectively (Table III). Inhibition of rate of growth was more with 60% ethanol leaf extracts of *P. guajava*. Thus, high inhibition (83.33%) was observed with strains isolated from *X. sagittifolium* of $L_3$ and $L_4$, after 8 days of growth. This percentage of inhibition is lower at 30% (Table IV).

**Virulence test of each strain of *F. oxysporum* harvested and inoculated to young plants of *X. sagittifolium* obtained *in vitro***

The test of virulence of different strains of *F. oxysporum* showed that plants inoculated with strains isolated from *X. sagittifolium* roots harvested from $L_1$, $L_2$, $L_4$ $L_5$ and $L_6$ presented an overall yellowing of the leaves (Fig. 3). *X. sagittifolium* plants inoculated with strains from $L_7$ and $L_8$ presented wilting of the leaves of the basal part of the plant (Fig. 4 A and B). However, 14 days after, plants inoculated with the *F. oxysporum* strains isolated from *X. sagittifolium* harvested from $L_3$ showed both yellowing and wilting of leaves (Fig.5). All inoculated plants had several dry roots in the rhizosphere. The severity of the disease evaluated varies significantly at 5% with all the strains of *F. oxysporum* isolated (Fig. 7 A). It is maximum 64.00±0.86% with the *F. oxysporum* strain isolated at $L_3$. The severity score was zero in the control (Fig. 7 A and B). The lowest severity percentage was recorded with the *F. oxysporum* strain isolated at $L_5$. This severity is identical in strains of $L_4$ (56.00 ± 1.00%), $L_6$ (56.00 ± 0.86%) and $L_8$ (56.00 ± 0.50%). After treatment with ethanol extract of *P. guajava* in twice day 7 and day 14, there is a resumption of growth (Fig. 6). The leaves were greener and wider and the petioles more vigorous. The reduction in severity was noted in plants treated with the solution of ethanol leaf extracts from *P. guajava* (Fig. 7 B). This reduction is 67.85% in the presence of the strain of *F. oxysporum* isolated at $L_6$. It is greater than 50% in plants of *X. sagittifolium* inoculated with $L_2$ (64.06%), $L_4$ (51.78%) $L_7$ (54.16%) and $L_8$ (56.25%), then treated with ethanol leaf extract of *P. guajava*

**Comment [A8]:** virulence assay of ***F. oxysporum*** against ***X. sagittifolium***

**Discussion**

The objective of this work was to determine the different morphotypes of *Fusarium* sp. present in the roots of *Xanthosoma sagittifolium* L. Schott. The results highlight the presence of *Fusarium oxysporum*, based on the identification keys of *Fusarium* Interactive Key (Agr &

**Comment [A9]:** need more causes to explanation of the results with comparison with other studies

Agri-Food Canada) and Simplified Fungi Identification Key (2001). The morphological analyzes of the different strains show that the cottony appearance of the hyphae is the most abundant in *F. oxysporum*. This suggests that cottony form of *F. oxysporum* may be the most abundant in the soil. Similar results were obtained by Bhimani et *al*. (2018), who observed an abundance of the cottony form of *F. oxysporum* in *Trigonella foenum-graecum* L. (Fenugreek). The histological analysis of the conidial structure of the different strains isolated, showed macroconidia, microconidia and chlamydospores after eight days of growth. Strains isolated from *X. sagittifolium* harvested from $L_1$ and $L_5$ are rich in macroconidia and chlamydospores, while those of $L_2$, $L_4$, $L_6$, $L_7$ and $L_8$ are rich in microconidia.

Growth inhibition tests with the ethanol extracts of *Psidium guajava* showed significantly reduced growth in *F. oxysporum* in all isolated strains, being at the initial phase with 30% and decrease with time. This suggests that the amount of secondary metabolites released into petri dish and responsible for inhibiting the fungus decreases over time. Therefore, they are volatile compounds. Higher concentration of the ethanol extract cause low growth. This growth reduction is very high under 60% of ethanol extract of *P. guajava* leaves. This inhibition of growth could probably be attributed to the richness of the secondary metabolites of the leaves which have been released and which would have antifungal properties vis-à-vis *F. oxysporum*. Djeuani et *al*. (2014), Rongai et *al*. (2015), attribute this antifungal property of the leaves *P. guajava* to their richness in flavonoids, phenol, tannins and alkaloids. Moreover, Pelczar et *al*. (1998) evidence shows that during the mechanism of antimicrobial substances to inhibit the growth of microbes, phenolic are able to change permeability of the cytoplasmic membrane which causes the leakage of nutrients from within the cell. In addition, the highest inhibition percentages of 83.33%, obtained in strains harvested from $L_3$ and $L_4$ localities after 8 days. This suggests that both strains are the most sensitive to the concentration of 60% ethanol leaf extract used. This concentration of 60% of ethanol leaf extract of *P. guajava* used would stop these strains of *F. oxysporum* to obtain in the PDA medium nutrients necessary for their growth.

The virulence tests carried out showed that most *F. oxysporum* strains used cause dry roots and leaf yellowing in *X. sagittifolium*. Strains of $L_3$ caused both wilting and yellowing of the leaves. Similarly, $L_7$ and $L_8$ strains cause wilting of the leaves. This suggested that *F. oxysporum* is a pathogen of *X. sagittifolium* plants and their aggressiveness observed depends on the strain used and maybe the age of the plant. At three months of age, *X. sagittifolium* plants used are in the active growth stage, this would explain the susceptibility observed towards *F. oxysporum* strain collected. After spraying the plant with ethanol extract from *P.*

*guajava* leaves, growth was accelerated, leaves were greener and wider and the petioles more were vigorous. This suggests that the ethanol leaves extract of *P. guajava* stimulates growth while inhibiting the action of different strains of *F. oxysporum*. The greener leaves observed implies stimulation of the photosynthesis mechanism. Similarly, Hanafy et *al*. (2012) showed that, foliar application of aqueous garlic bulb extract accelerated plant growth through the stimulation of photosynthetic pigments and soluble sugar content in *Schefflera arboricola* Plants.

## CONCLUSION

Most of the *F. oxysporum* strains isolated cause leaf yellowing and dry root in *Xanthosoma sagittifolium*. It appears that leaf extracts of *P. guajava* inhibit the mycelial growth of the various strains isolated. The higher inhibition was recorded under 60% of ethanol extract of *P. guajava* leaves. In addition, the use of *P. guajava* extracts not only appears to inhibit the pathogen but can facilitate the recovery of growth in *X. sagittifolium*.



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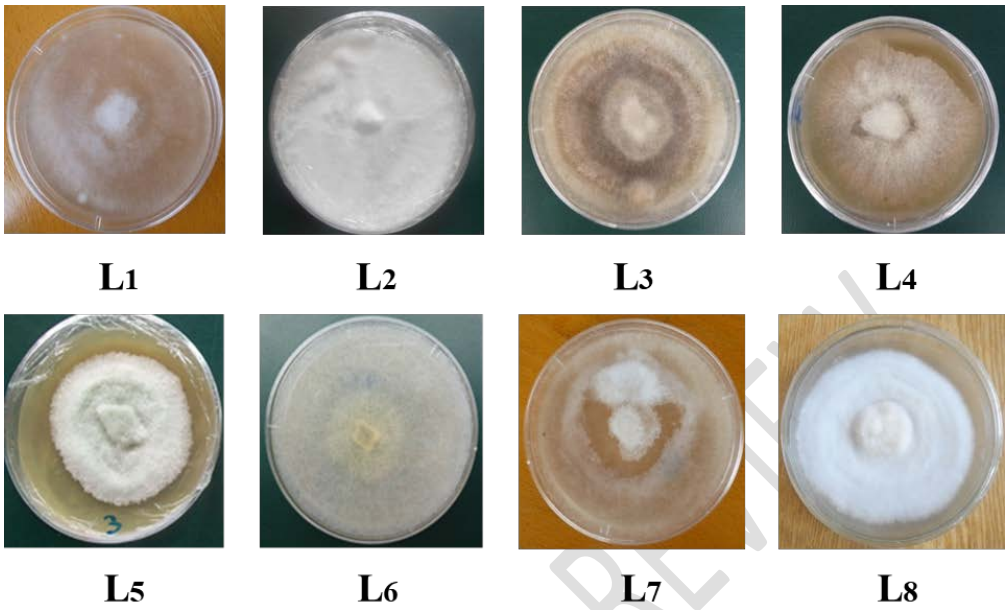


Fig.1. Aspect of growth of *Fusarium oxysporum* strains in Petri dishes. *F. oxysporum* strain harvested in different localities: $L_1$ (Soa), $L_2$ (Banda), $L_3$ (Loum) and $L_4$ (Bangoua), $L_5$ (Abong-Mbang), $L_6$ (Bansoa), $L_7$ (Santa) and $L_8$ (Ekona).

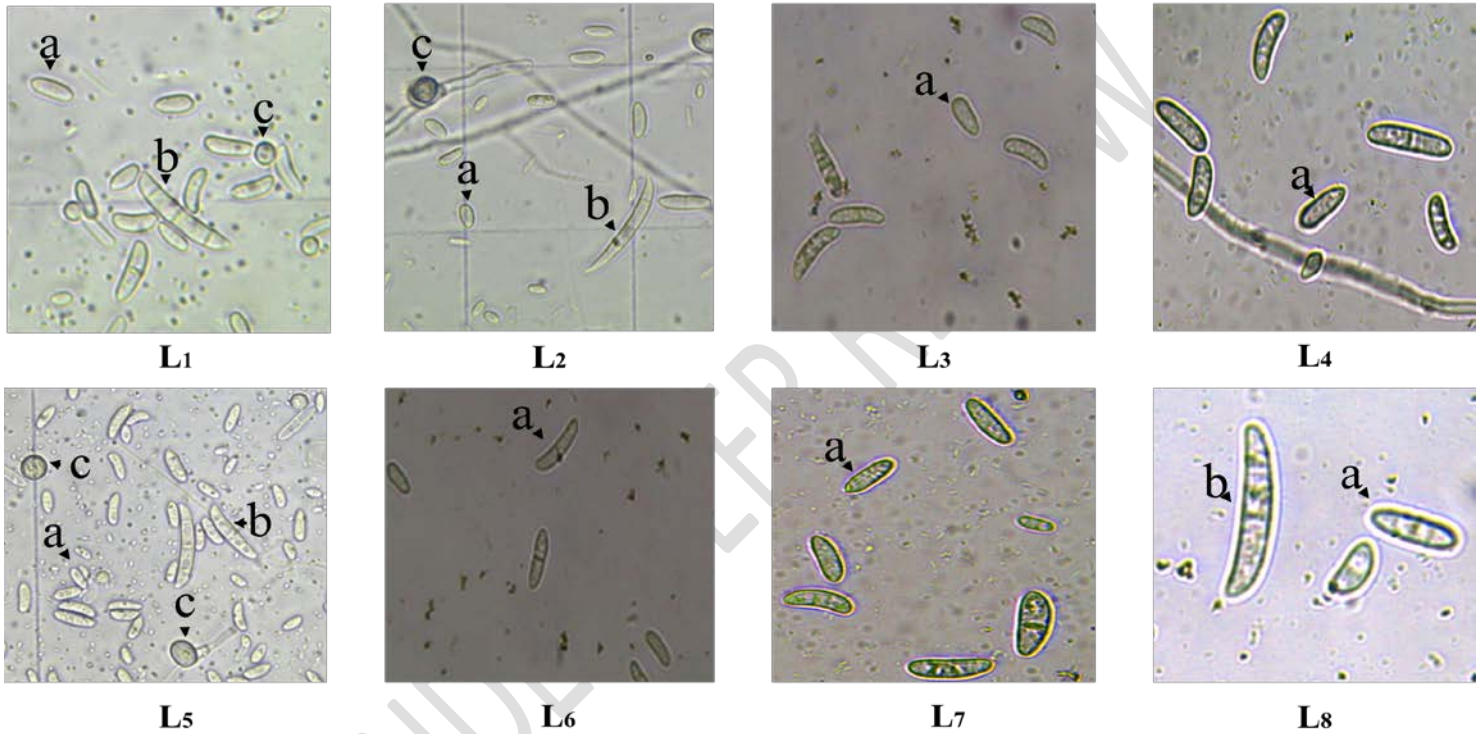


Fig. 2. Structures of the conidia of the different strains of *Fusarium oxysporum* according to the harvest locality observed under an optical microscope (Brand) at 400X. Microconidia (a), macroconidia (b) and chlamydospores (single) (c). *F. oxysporum* strain harvested in different localities: $L_1$ (Soa), $L_2$ (Banda), $L_3$ (Loum) and $L_4$ (Bangoua), $L_5$ (Abong-Mbang), $L_6$ (Bansoa), $L_7$ (Santa) and $L_8$ (Ekona).

**Comment [A10]:** all images need scale par

Table I: Abundance of strains in roots following localities

| | harvest locality | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | **$L_1$** | **$L_2$** | **$L_3$** | **$L_4$** | **$L_5$** | **$L_6$** | **$L_7$** | **$L_8$** |
| Fluffy | + | - | - | - | - | - | - | - |
| Woolly or cottony | + | + | + | + | + | + | + | + |
| Sclerotal | - | - | + | - | - | - | - | - |
| Senescent Ras | - | - | - | + | - | - | - | - |
| Woolly or cottony | + | + | + | + | + | + | + | + |
| Pionnotal | - | - | - | - | - | + | - | - |
| Senescent Ras | - | - | - | - | - | - | + | - |
| Woolly or cottony | + | + | + | + | + | + | + | + |

*F. oxysporum* strain isolated from *X. sagittifolium* roots in different localities: $L_1$ (Soa), $L_2$ (Banda), $L_3$ (Loum) and $L_4$ (Bangoua), $L_5$ (Abong-Mbang), $L_6$ (Bansoa), $L_7$ (Santa) and $L_8$ (Ekona). Absent (-) and present (+)

Table II: The colony characters and sporulation of different strains of *F oxysporum* collected in roots following localities

| Strains | Texture | Color | Density | Aerial mycelium | Growth habit | Form | Sporulation |
|---|---|---|---|---|---|---|---|
| **$L_1$** | Fluffy | White | Low | Regular | Moderate | Radial | Abundant |
| **$L_2$** | Woolly | White | Regular | Abundant | Abundant | Radial | Profuse |
| **$L_3$** | Sclerotal | Purplish pink | Regular | Abundant | Abundant | Radial | Moderate |
| **$L_4$** | Senescent Ras | Pinkish | Abundant | Abundant | Moderate | Radial | Moderate |
| **$L_5$** | Woolly | White | Abundant | Abundant | Slow | Radial | Abundant |
| **$L_6$** | Pionnotal | Beige | Regular | Regular | Abundant | Radial | Poor |
| **$L_7$** | Senescent Ras | Beige | Regular | Abundant | Moderate | Radial | Moderate |
| **$L_8$** | Woolly | White | Abundant | Abundant | Moderate | Radial | Moderate |

*F. oxysporum* strain harvested in different localities: $L_1$ (Soa), $L_2$ (Banda), $L_3$ (Loum) and $L_4$ (Bangoua), $L_5$ (Abong-Mbang), $L_6$ (Bansoa), $L_7$ (Santa) and $L_8$ (Ekona).

Table III. Average radial growth of the different strains of *Fusarium oxysporum* in petri dish according to concentrations *Psidium guajava* applied.

| Concentration % | Inhibition time (Days) | Average radial growth of the different strains | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Strains harvested according to localities | | | | | | | |
| | | **$L_1$** | **$L_2$** | **$L_3$** | **$L_4$** | **$L_5$** | **$L_6$** | **$L_7$** | **$L_8$** |
| **0** | 0 | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ |
| | 2 | 6.0±0.02$^{b}$ | 6.3±0.01$^{b}$ | 6.0±0.06$^{b}$ | 5.5±0.05$^{b}$ | 5.9±0.01$^{b}$ | 6.5±0.01$^{b}$ | 4.9±0.11$^{b}$ | 6.5±0.08$^{b}$ |
| | 4 | 7.8±0.07$^{c}$ | 7.8±0.14$^{c}$ | 7.8±0.10$^{c}$ | 7.8±0.07$^{c}$ | 7.5±0.02$^{c}$ | 7.9±0.05$^{c}$ | 7.6±0.13$^{c}$ | 7.5±0.10$^{c}$ |
| | 6 | 8.7±0.01$^{d}$ | 8.8±0.15$^{d}$ | 8.6±0.05$^{d}$ | 8.9±0.06$^{d}$ | 8.7±0.00$^{d}$ | 8.8±0.04$^{d}$ | 8.5±0.09$^{d}$ | 8.6±0.11$^{d}$ |
| | 8 | 8.9±0.01$^{d}$ | 9.0±0.00$^{d}$ | 9.0±0.05$^{d}$ | 9.0±0.02$^{d}$ | 9.0±0.05$^{d}$ | 9.0±0.04$^{d}$ | 9.0±0.01$^{d}$ | 9.0±0.00$^{d}$ |
| **30** | 0 | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ |
| | 2 | 1.7±0.06$^{b}$ | 1.3±0.01$^{b}$ | 5.5±0.01$^{b}$ | 2.2±0.01$^{b}$ | 1.6±0.10$^{b}$ | 2.2±0.03$^{b}$ | 1.7±0.06$^{b}$ | 1.9±0.01$^{b}$ |
| | 4 | 2.0±0.05$^{c}$ | 1.8±0.05$^{b}$ | 5.5±0.01$^{b}$ | 3.4±0.00$^{c}$ | 2.6±0.09$^{c}$ | 2.7±0.12$^{b}$ | 2.3±0.01$^{b}$ | 3.0±0.05$^{c}$ |
| | 6 | 3.2±0.01$^{d}$ | 2.6±0.09$^{c}$ | 5.7±0.02$^{b}$ | 3.9±0.11$^{cd}$ | 3.3±0.06$^{d}$ | 3.0±0.01$^{c}$ | 3.4±0.01$^{c}$ | 3.2±0.05$^{c}$ |
| | 8 | 4.0±0.01$^{e}$ | 3.3±0.08$^{d}$ | 6.5±0.03$^{c}$ | 4.4±0.10$^{d}$ | 4.0±0.02$^{e}$ | 3.3±0.05$^{c}$ | 3.4±0.05$^{c}$ | 4.0±0.05$^{d}$ |
| **60** | 0 | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00$^{a}$ | 0.0±0.00 |
| | 2 | 2.1±0.03$^{b}$ | 2.2±0.01$^{b}$ | 1.5±0.08$^{b}$ | 1.6±0.04$^{b}$ | 2.3±0.01$^{b}$ | 1.5±0.04$^{b}$ | 0.0±0.00$^{a}$ | 1.5±0.01 |
| | 4 | 2.5±0.05$^{b}$ | 3.3±0.00$^{c}$ | 1.5±0.10$^{b}$ | 1.5±0.00$^{b}$ | 3.2±0.01$^{c}$ | 1.7±0.05$^{b}$ | 2.2±0.05$^{b}$ | 2.5±0.02 |
| | 6 | 2.5±0.08$^{b}$ | 3.3±0.05$^{c}$ | 1.5±0.01$^{b}$ | 1.5±0.05$^{b}$ | 3.7±0.03$^{d}$ | 2.0±0.05$^{c}$ | 2.3±0.09$^{b}$ | 2.5±0.02 |
| | 8 | 2.7±0.01$^{b}$ | 3.3±0.05$^{c}$ | 1.5±0.11$^{b}$ | 1.5±0.01$^{b}$ | 4.0±0.05$^{d}$ | 2.0±0.06$^{c}$ | 2.3±0.10$^{b}$ | 2.5±0.04 |

*Data sharing the same letter in the same column were not significantly different at 5% level (Duncan's multiple range tests).*

*F. oxysporum* strain harvested in different localities: $L_1$ (Soa), $L_2$ (Banda), $L_3$ (Loum) and $L_4$ (Bangoua), $L_5$ (Abong-Mbang), $L_6$ (Bansoa), $L_7$ (Santa) and $L_8$ (Ekona).

Table IV. Evaluation of the average percentage of inhibition in the different strains of *Fusarium oxysporum* according to the concentrations of *Psidium guajava* applied.

| Concentration (%) | Inhibition time (days) | **Percentage inhibition (%)** | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Strains harvested according to localities | | | | | | | |
| | | **$L_1$** | **$L_2$** | **$L_3$** | **$L_4$** | **$L_5$** | **$L_6$** | **$L_7$** | **$L_8$** |
| **30** | **0** | 00.00 | 00.00 | 00.00 | 00.00 | 00.00 | 00.00 | 00.00 | 00.00 |
| | **2** | 71.66 | 79.36 | 08.33 | 60.00 | 72.88 | 66.15 | 65.30 | 70.76 |
| | **4** | 74.35 | 76.92 | 29.48 | 56.41 | 65.33 | 65.82 | 69.73 | 64.00 |
| | **6** | 63.21 | 70.45 | 33.72 | 56.17 | 62.06 | 65.90 | 60.00 | 62.79 |
| | **8** | 55.05 | 63.33 | 27.77 | 51.11 | 55.55 | 63.33 | 62.22 | 55.55 |
| **60** | **0** | 00.00 | 00.00 | 00.00 | 00.00 | 00.00 | 00.00 | 00.00 | 00.00 |
| | **2** | 65.00 | 65.07 | 75.00 | 78.18 | 61.01 | 76.92 | 00.00 | 76.92 |
| | **4** | 67.79 | 57.69 | 80.76 | 83.31 | 57.33 | 78.84 | 71.05 | 66.66 |
| | **6** | 71.26 | 62.50 | 82.55 | 83.14 | 57.47 | 77.27 | 72.94 | 70.93 |
| | **8** | 69.66 | 63.33 | 83.33 | 83.33 | 55.55 | 77.77 | 74.44 | 72.22 |

*F. oxysporum* strain harvested in different localities: $L_1$ (Soa), $L_2$ (Banda), $L_3$ (Loum) and $L_4$ (Bangoua), $L_5$ (Abong-Mbang), $L_6$ (Bansoa), $L_7$ (Santa) and $L_8$ (Ekona).

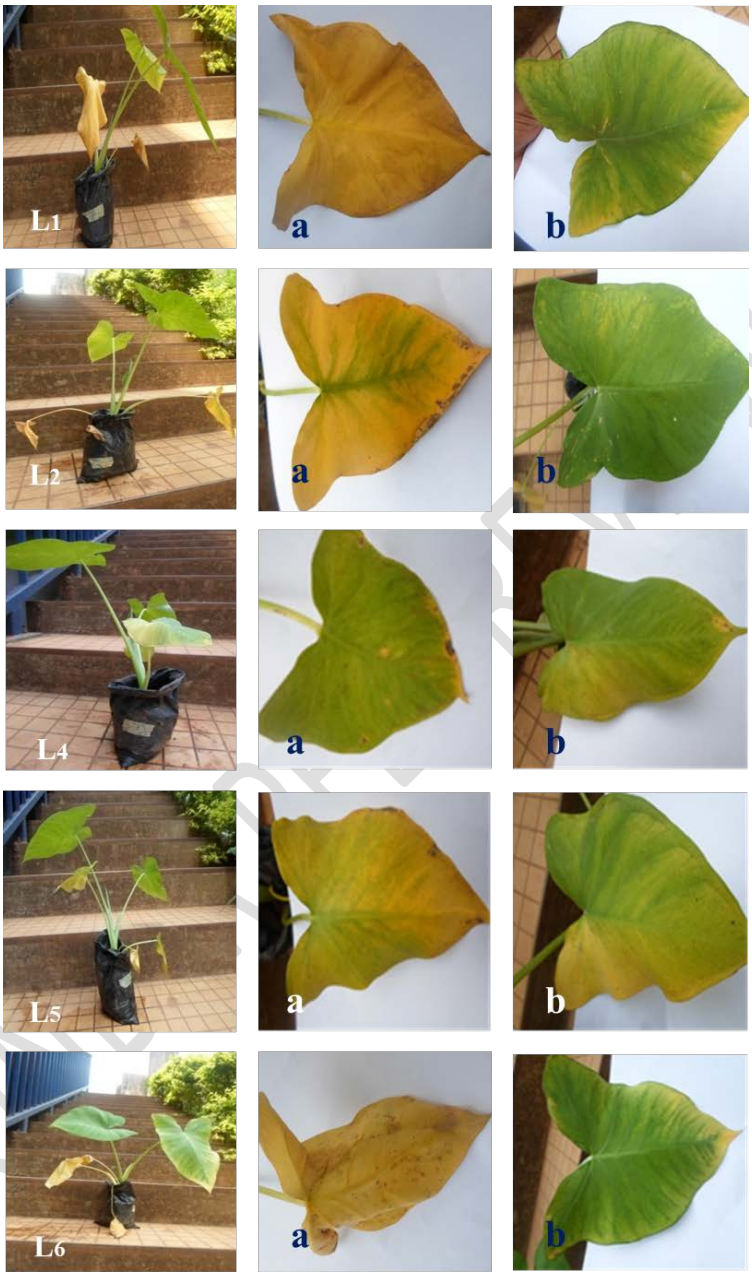


Fig. 3. Aspect of *X. sagittifolium* leaves at Day 14 after inoculation. *F. oxysporum* strain responsible of the yellowing of leaves according to the different harvesting locations: $L_1$ (Soa), $L_2$ (Banda), and $L_4$ (Bangoua), $L_5$ (Abong-Mbang) and $L_6$ (Bansoa). Leaves almost or completely yellow (a) and leaf showing yellowing (b).

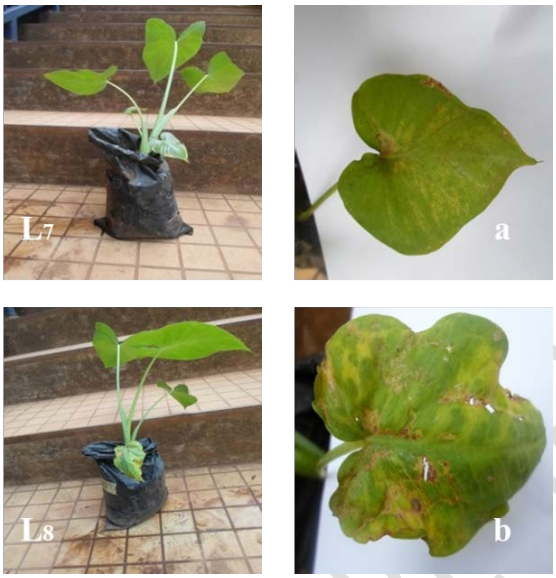


Fig. 4. Plantlets with wilting of the leaf due to the action of *Fusarium oxysporum* after 14 days of inoculation. *F. oxysporum* strain responsible of the wilting of leaves according to the different harvesting locations: $L_7$ (Santa) and $L_8$ (Ekona). Withered leaves (a and b).


Fig. 5. Plantlets of *X. sagittifolium* with both yellowing (a) and wilting (b) leaves after 14 days of inoculation with *F. oxysporum* strain harvested in $L_3$ (Loum).

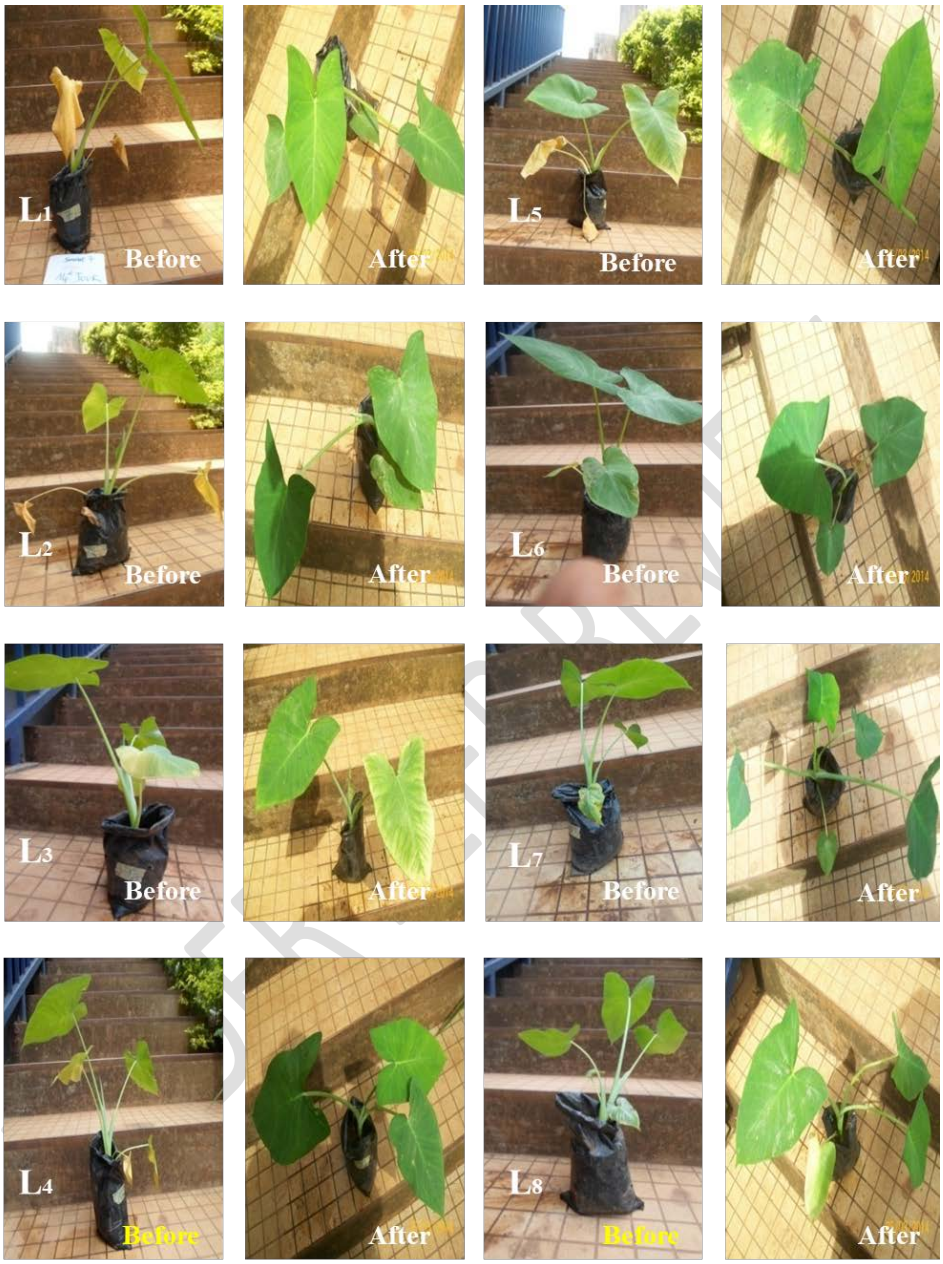


Fig. 6. Aspect of leaves of inoculated plants of *X. sagittifolium* with *F. oxysporum* strains after treatment with ethanol leaves extracts of *P. guajava* at day14. Locations: $L_1$ (Soa), $L_2$ (Banda), $L_3$ (Loum) and $L_4$ (Bangoua), $L_5$ (Abong-Mbang), $L_6$ (Bansoa), $L_7$ (Santa) and $L_8$ (Ekona).

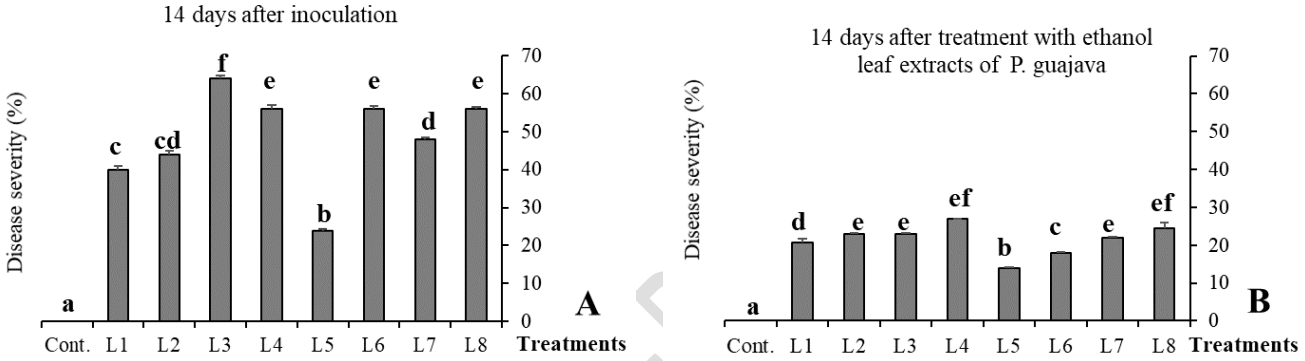


Fig. 7. Disease severity in *X. sagittifolium*. Day 14 after inoculation (A) and day 14 after treatment of plants with ethanol leaf extract from *P. guajava* (B). Locations: $L_1$ (Soa), $L_2$ (Banda), $L_3$ (Loum) and $L_4$ (Bangoua), $L_5$ (Abong-Mbang), $L_6$ (Bansoa), $L_7$ (Santa) and $L_8$ (Ekona).