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Infestation dynamics of mealybug vectors (Hemiptera : Pseudococcidae) of the Cocoa swollen shoot virus (CSSV) in young cocoa plots surrounded by barrier crops

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ABSTRACT

The Cocoa swollen shoot disease probably represents today the greatest threat to cocoa production in Côte d'Ivoire. The disease is caused by the Cacao swollen shoot virus (CSSV), a badnavirus transmitted to cocoa by about 15 mealybug species in West Africa. At local scale, the propagation of CSSV outbreaks is closely related to mealybug dispersal behaviors. The ability of barrier crops to stop the disease spread has been demonstrated in Togo and Ghana, yet mechanisms involved are still not elucidated. The present study aims at better understanding these mechanisms by studying mealybug population dynamics in young plantations surrounded by barrier crops. A set of twelve 0.25 ha cocoa plots surrounded by 10 m large hedges of coffee (*Coffea robusta*) or acacia (*Acacia auriculiformis*), or without hedges (control plots), was implemented within large CSSV outbreaks in mature cacao plantations, in July and August 2019, near Soubré (South-West Côte d'Ivoire). First counts of mealybug colonies on cocoa and barrier crops were done in September 2019 and February, March and May 2020. From November 2020 to August 2022, populations were assessed monthly using a scoring scale, a score from 0 to 3 being given to each cocoa according to the size of the hosted population. *Pseudococcus longispinus* and *Ferrisia virgata* were the first species present on cocoa, but populations remained small until May 2020 with a maximum of 0.5% of trees infested by few mealybugs. These two species have long waxy hairs that allow them to be transported by wind. Although very common in neighboring mature plantations, *Formicococcus njalensis* populations remained small in young plots. During the period November 2020 - August 2022, *F. njalensis* was the dominant species on cocoa with 63.8% of the total number of infested trees. *F. virgata*, *Planococcus citri* and *P. longispinus* represented 10.8%, 4.7%, 2.9% of infested trees, respectively. Following results were obtained for the most infested plantation, with coffee barriers. Weighted infestation rate for *F. njalensis* showed significant variation throughout the period, but without clear seasonal trend, suggesting that other factors than climate may have affected mealybug populations. Maps of infestation and semivariograms showed that *F. njalensis* population was globally aggregated, meaning that cocoa trees hosting mealybugs were usually grouped. In addition, borders of the plot were more infested than center. suggesting that plot colonization by mealybugs started from coffee barriers. These results were discussed and recommendations given for more efficient barrier crops.

Keywords: Population dynamics, dispersion, distribution,

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1. Introduction

Mealybugs are the only known vectors of the Cocoa swollen shoot virus (CSSV), a disease that threatens cocoa production in West Africa. In Côte d'Ivoire, about a dozen species would be responsible of the quick spread of the disease over most of producing regions of the country and in only two decades (N'Guessan et al., 2019). Among these species, two are particularly common and widely distributed, namely *Formicococcus njalensis* Laing and *Planococcus citri* (Risso). Due to the white waxy secretion covering their body, these insects are easily detected on cocoa pods and shoots, where they thrive in close interaction with ants. Waxy protection also makes them difficult to control using conventional contact insecticides (Venkatesan et al., 2016). Spatial and temporal dynamics of CSSV outbreaks are strongly related to mealybug dispersion, which is of limited duration and rarely exceeds few meters (Campbell, 1983). That's why CSSV disease is usually considered as an archetypal crowd disease, namely a disease that spreads slowly and does not affect very large areas of plantations in a short time (Jeger & Thresh, 1993). But what remains clear is that the disease incurably progresses over large areas of plantations and that cocoa farmers are poorly equipped to face the threat. Full-sun cocoa monoculture are often found responsible of CSSV expansion in Côte d'Ivoire. Different mechanisms may be involved: i) micro-climate of full-sun plantations is favorable to mealybug development, especially *P. citri* (Risso), which became dominant in some areas (Campbell, 1984); ii) full-sun cocoa leads to stress related to high light and low soil moisture that may exacerbate CSSV symptoms (Andres et al., 2018); iii) in adjacent monocultures, mealybugs disperse without meeting obstacles leading to large infected areas in few years. Based on this last hypothesis, a control strategy involving the planting of barrier crops around cocoa plantations has been tested with some promising results especially in Ghana (Domfeh et al., 2016). However, much remains to be learned about the processes involved in barrier effects of hedges toward mealybugs. Especially our knowledge of mealybug population dynamics on cocoa is poor, including dispersal behaviour and factors involved in mealybug distribution. The present paper presents preliminary results on the colonization of young plots surrounded by barrier crops by different species of mealybug. Population spatio-temporal dynamics are described in most infested plots for a period of two years.

2. Materials and methods

2.1. Experimental set-up

A set of twelve 0.25 ha cocoa plots surrounded by 10 m large barriers of coffee (*Coffea robusta*) or acacia (*Acacia mangium*) (Figure 1), or without barriers (control plots), was implemented within large CSSV outbreaks in mature cacao plantations, in July and August 2019, near Soubré, South-West Côte d'Ivoire (5°47'N, 6°35'O). Before planting, old infected cocoa trees were cut and tree stumps devitalized using a high dose of herbicide, as recommended by government. Plantains and barrier crops were planted first to shade and protect young cocoa plants. Cocoa was planted according to recommended good practices.

2.2. Assessment of mealybug populations

Mealybug colonies were first recorded as presence/absence data for each cocoa tree of the plots in September 2019 and February, March and May 2020. From November 2020 to August 2022, populations were assessed monthly on each cocoa tree using a scoring scale ranging from 0 for no population to 3 for very abundant population. In both cases, presence/absence data were used for computing infestation rates for the different mealybug species.

2.3. Temporal variation

Temporal variation of mealybug populations was characterized for the most infested plot by plotting the weighted infestation rate for the different dates of observation. The weighted infestation rate was calculated for each observation date using the following equation:

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$$Tx = \frac{100}{3n} \times (1x_1 + 2x_2 + 3x_3)$$

Where n is the number of cocoa trees observed in the plot (where there are no missing trees, n = 144) and x_1, x_2 and x_3 are the number of trees with score 1, 2 and 3, respectively.

2.4. Spatial analysis

The most infested plot was chosen for spatial analysis. First, mealybug population data was mapped for the different observation dates using the ggplot2 package on R (version 4.2.1). Then, the spatial relationship between individual counts was assessed through semivariogram analysis using the GeoR package on R. Semivariogram plots semivariance as a function of distance, where the semivariance is given by the following equation:

$$\gamma(\mathrm{h}) = \left[\frac{1}{2N(h)}\right] \times \sum [Z_i - Z_{i+h}]^2$$

where Z_i was the log$_e$-transformed mealybug score plus one for cocoa tree i, Z_{i+h} was the log$_e$-transformed mealybug score plus one for cocoa tree $i+h$ and $N(h)$ was the total number of pairs of cocoa trees observed for the distance interval h, expressed in meters.

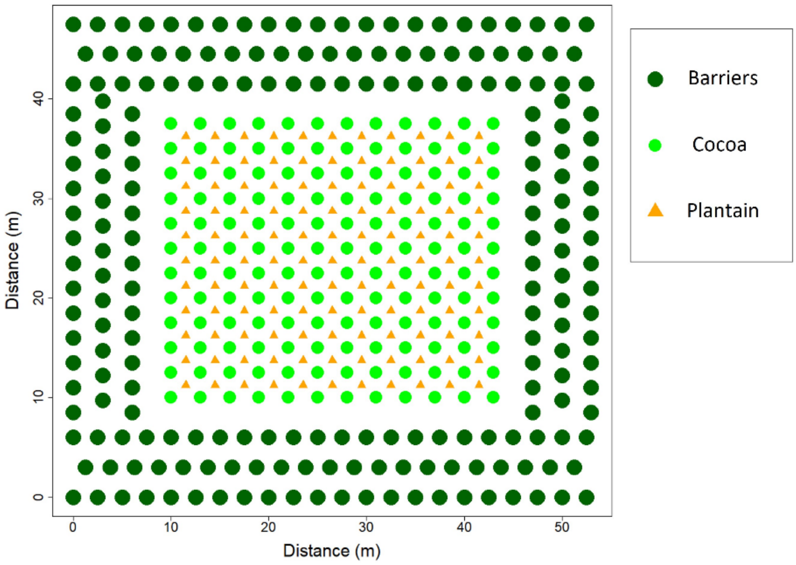

Figure 1 Diagram of the experimental set-up

In our study, the semivariance was calculated for distances between 2 and 20 meters (≈ 50% of the maximum distance between any two cocoa trees of the plot). The number of distance classes was set up at 12. Then parametric variogram models were fitted to sample semivariograms. The following parameters were used to describe parametric variograms: the model type, the range, which is the value x at which the curve reaches the sill, and the nugget variance, which is the value y at which the curve of the model cuts the Y-axis. The range is the distance (in meter in our study) for which spatial dependence is detected. The nugget variance gives the part of the variance not spatially dependent over the range.

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3. Results and discussion

3.1. Infestation by mealybugs

Early counting in September 2019 and February, March and May 2020 revealed that *Pseudococcus longispinus* and *Ferrisia virgata* were the first species present on cocoa, although populations remained small with a maximum of 0.5% of cocoa trees infested by mealybugs. These results suggest that, on one hand, barriers of coffee and Acacia did not keep mealybugs from reaching young cocoa plants. On the other hand, whatever the mealybug species, they failed to colonize plots the first six months after plantation at least. Although very common in neighboring mature plantations, *Formicococcus njalensis* was not early recorded in young plots. However, *P. longispinus* and *F. virgata* may have benefited from their long waxy hairs that could have helped them to be transported by wind over longer distances. In any case, no significant mealybug population has settled in young plots, probably because of a lack of food on young cocoa seedlings. By contrast, observations from November 2020, i.e., 16-17 months after planting, revealed the presence of well-established mealybug colonies, with *F. njalensis* as the dominant species on cocoa, representing 63.8% of the total number of infested trees of the period, while *F. virgata*, *P. citri* and *P. longispinus* represented 10.8%, 4.7%, 2.9% of the total number of infested trees, respectively.

3.2. Temporal evolution of infestation

The results presented here and in the following section are for the most infested plot for the period from November 2020 to August 2022. *Formicococcus njalensis* was the dominant species and weighted infestation rate for the species showed significant variation throughout the period (Figure 2), with peaks in April 2021 (4.8%), September 2021 (7,1%), January 2022 (6.6%) and August 2022 (11,0%). There is no clear seasonal trend, suggesting that other factors than climate, such as insecticide sprayings or exceptionally heavy rains may have affected mealybug populations. Fruit production, that started in February 2022 in this plot, may also be an important factor explaining increasing infestation from May 2022.

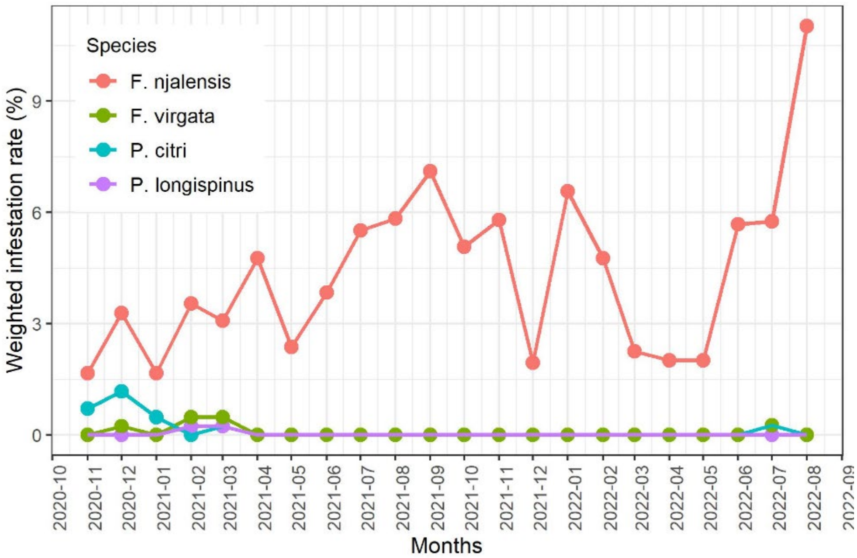

Figure 2 Variation of weighted infestation rate (%) of cocoa trees by mealybugs in the most infested plot of the study

3.3. Spatial distribution of infestation

Maps of infestation show that *F. njalensis* population was globally aggregated, meaning that cocoa trees hosting mealybugs were usually grouped (Figure 3). Semivariograms also reveal that infestation distribution was spatially structured in patches, with spatial dependance between 5 and 10 meters, at

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least for the two first dates of observation. From the third date, distribution in patches was not so clear, as infestation tended to move from the bottom to the top left of the maps. What is clear is that borders of the plot, that is, cocoa trees in contact with coffee barriers were more infested than those in the plot center. Knowing that coffee also hosts *F. njalensis,* this result suggests that plot colonization by mealybugs started from coffee barriers. The spatial distribution in patches suggest an aggregated population and a dispersion based on short distance movements, from one tree to its neighbors.

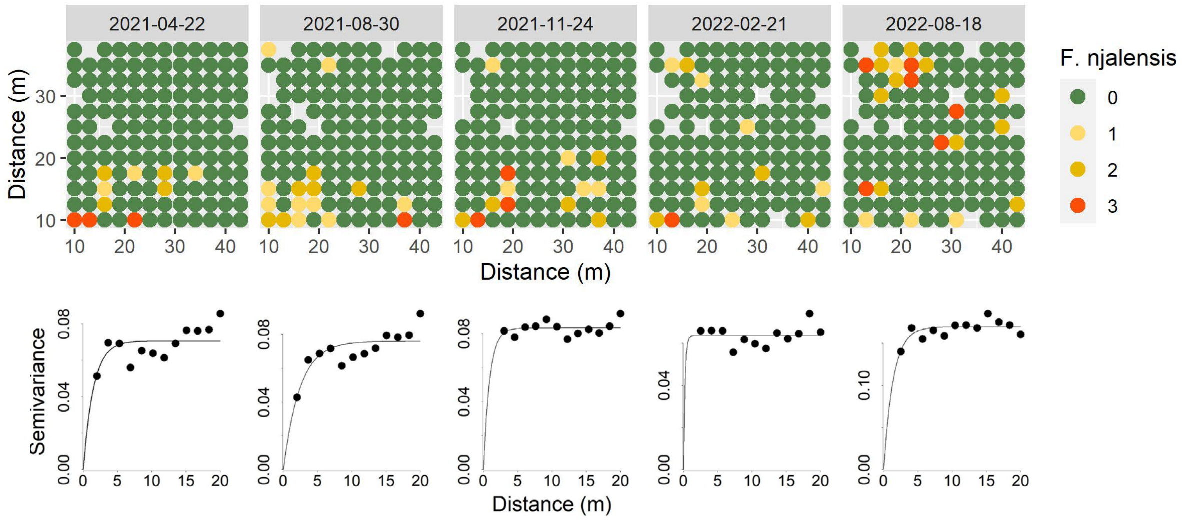

Figure 3 Infestation maps (top) and related semivariograms (bottom) for the most infested plot of the study, for the mealybug Formicococcus njalensis and for 5 selected dates of evaluation. Empty spaces between cacao trees on maps show missing (dead) trees.

The role that interactions between coffee barriers and adjacent cocoa trees may play in plot colonization by mealybugs and CSSV disease is worth studying in more details. Coffee, as host of most of vector mealybugs and non-host of CSSV may protect cocoa from infection by acting as a biological barrier. CSSV-infectious mealybugs may get free from the virus by feeding on coffee and become harmless before reaching adjacent cocoa. In the present study, the coffee barriers included 3 lines of coffee for a total width of 10 m. It is likely that such a barrier is wide enough to allow mealybugs such as *F. njalensis*, which usually slowly disperse by moving from a tree to its neighbor (Campbell, 1983), to get free from CSSV. A different problem arises with mealybugs that disperse using wind. With their long waxy hairs, *P. longispinus* and *F. virgata* are well equipped for transport by wind over longer distances (Barrass et al., 1994). Coffee barriers that hardly exceed cocoa trees in height may not be high enough to protect cocoa from these mealybugs, which may reach young cocoa directly from old CSSV-infected trees located several meters away. In our study, early colonization of plots by *P. longispinus* and *F. virgata*, even if these plots were surrounded by barriers of *Acacia auriculiformis*, suggest insufficient size of barriers that were enable to break wind. These results highlight the importance of both the plant species chosen for barrier and the timing of plantation, which should allow barrier plants to grow enough before planting cocoa.

4. Conclusions and recommendations

In conclusion, our results clearly show that coffee barriers did not prevent cocoa plots to be infested by mealybugs. In a certain way, on the contrary, coffee barriers may have facilitated *F. njalensis* penetration in young plots from surrounding old cocoa plantations. This should not favor the emergence of CSSV disease in the plot, as coffee acts as a biological barrier, allowing mealybugs to get free from virus before reaching young cocoa. Another result is that, for the first few months, none of the barriers have been able to keep young plots from mealybugs dispersed by wind, such as *Ferrisia virgata* or *Pseudococcus longispinus*.

In terms of recommendations, the variety of dispersal behaviors among mealybugs suggests that multispecies barriers, with fast-growing windbreak plants associated to plants hosting mealybugs but not CSSV (as coffee), would be better than monospecific barriers. Another recommendation is that new cocoa plantation should not start before windbreak plants have reached a good height, that is, at least, the height of surrounding mature cocoa.

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