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#### Chapter

## Integrated Management Approach to Citrus Fungal Diseases by Optimizing Cocoa-Based Agroforests Structural Characteristics

Ndo Eunice Golda Danièle and Akoutou Mvondo Etienne

#### Abstract

The health and productivity of citrus are generally jeopardized by a host of diseases, for which the environmental conditions of the cropping system are critical drivers. Several studies conducted on various diseases of perennial crops have shown the involvement of the structural futures of the cocoa-based agroforestry system (CBAFS) in the spread of pathogens and the epidemics development. This chapter highlights the effect of the CBAFS's structural characteristics on the intensity of three citrus diseases in the humid forest zones of Cameroon. The involvement of CBAFS structural characteristics in diseases regulation is demonstrated. In particular, the spatial structure of citrus in agroforests shows an effect on the spread of diseases. Moreover, distribution of citrus in the CBAFS, with minimum spacing of 12 m between citrus trees, limits the damage caused by Pseudocercospora leaf and fruit spot disease (PLFSD) and citrus diseases caused by Phytophthora (CDP). Dense shading helps to minimize the intensity of diseases such as CDP and PLFSD and Citrus scab disease. This work may make it possible to contribute to the development of an integrated management tool for citrus diseases in an associated crop context.

**Keywords:** Integrated disease management, cultural practices, citrus, fungal diseases, shade trees, spatial structure

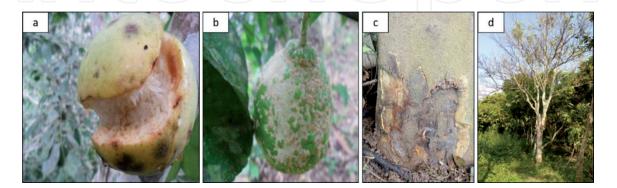
#### 1. Introduction

Conventional and intensive agriculture has enabled a considerable increase in agricultural production since the 1950s. However, the resulting heavy ecological balance sheet discredits this unsustainable model of agriculture [1]. Thus, although the preferred strategies of intensive agriculture have shown undeniable benefits, their use is becoming increasingly worrying both for agriculture itself and for the environment and human health. This is partly due to the excessive use of pesticides and other chemicals [2–6]. The improvement of intensive production systems towards new models of sustainable agriculture, favoring the development of effective means of combating diseases that are sustainable and environmentally friendly, is becoming a matter of urgency. Tropical agroforestry systems, thanks to their high biodiversity and structural diversity, represent a privileged way out of the agroecological transition. Several studies have demonstrated the contribution of the structural characteristics of these systems in the integrated management of pests and diseases of perennial crops, particularly citrus [7–9].

Citrus represent a fruit crop of prime importance in socio-ecological terms in Cameroon [10–12]. Their significance lies in the fact of their high-quality nutritional value and their contribution to the diversification of producers' incomes in rural areas. Citrus are also important in the local pharmacopeia [13–15]. They are also known for their role in restoring ecological balances after deforestation [16]. However, despite the favorable agroecological conditions throughout the country, the number of production basins identified and even the density of trees in farms, production remains poor [12, 17, 18]. A diversity of diseases affecting citrus in the country humid zones is the main constraint to their production [8, 11, 19, 20].

Pseudocercospora leaf and fruit spot disease (PLFSD) caused by *Pseudocercospora angolensis*, citrus scab disease caused by *Elsinoe* spp.; and citrus diseases caused by Phytophthora (CDP) caused by *Phytophthora* spp., are the main soil diseases on citrus in Cameroon (**Figure 1**) [7, 21–25]. Damages caused by these diseases result in heavy crop losses [26]. Sorting deviations of up to 100% of the production can be recorded in the case of PLFSD, if no treatment measures are taken [27]. Concerning citrus scab, severe attacks on young *C. volcameriana* plants, for example in the nursery, result in their death [20, 28]. CDP significantly limits citrus production in the plots where it is present [29–34]. In addition to reduced yields from the beginning of infection, the economic viability of orchards is reduced following tree death [7, 32, 35]. This strong and constantly changing diseases pressure, which not only causes enormous economic losses, but also leads (in the case of PLFSD) to quarantine and a ban on the export of citrus products to other production areas [25].

A variety of strategies are used to control citrus diseases. These include the practice of sanitation measures, the use of resistant cultivars and varieties, grafting, organic or mineral soil amendments, the use of plant extracts, biological control in a systemic approach and, above all, chemical control through fungicides [32, 36–40]. However, the high costs of these methods, development of resistance to chemical inputs, emergence of new diseases and growing concerns about environmental and soil health make these methods inadequate [41–45]. In addition, most of these techniques are unsuitable for the socio-cultural and even technological context of small-scale producers in inter-tropical regions. The development of targeted control protocols, taking into account the local socio-ecological context and existing production systems is therefore imperative. This chapter highlights the effect of the



#### Figure 1.

Symptoms of citrus diseases in Cameroon. C. paradisi fruit torn following a severe attack by Pseudocercospora angolensis (a), C. volcameriana fruit covered with scab spots due to Elsinoe spp. (b), lesions resulting from crown attacks due to Phytophthora spp. on C. sinensis tree(c) and dieback due to various diseases of an C. sinensis (d).

structural characteristics of the cocoa-based agroforestry system on the intensity of the three main citrus diseases in the humid forest zones of Cameroon.

## 2. Complex cocoa-based agroforest systems and structural characteristics

In Cameroon, citrus are mainly grown in cocoa-based agroforestry systems (CBAFS) [7, 46]. These are complex, highly biodiverse, natural forest-like cropping systems (**Figure 2**) [30, 47, 48]. In this system, several interactions of different nature and intensity can take place depending on the species present, their sizes and their positions [9, 49, 50]. One of these interactions is the action of diseases. The structural characteristics of CBAFS can contribute to control of these [20, 30, 51, 52]. Studies in these cropping systems and on various pathosystems have shown that spatial structure of species is important in reducing diseases development [49, 52]. Indeed, spatial structure has a twofold effect on the pathogen: firstly, the high plant biodiversity into CBAFS makes it possible to dilute the pathogens resource and thus reduce their presence and damage [53–55]. Secondly, multi-species agroecosystems are recognized for the high diversity of vertical and horizontal structure affects diseases mainly through the microclimatic weathering mechanism [51, 52].

Previous works have supported interactions between individuals of a host population of pathogen and associated plants within intercropping systems [7, 8]. This type of interaction is likely to influence the presence of diseases. The action of shade trees on the understory microclimate decreases with decreasing distance between trees and pathogen transmission decreases as the distance between host individuals increases [51, 52, 57–59].

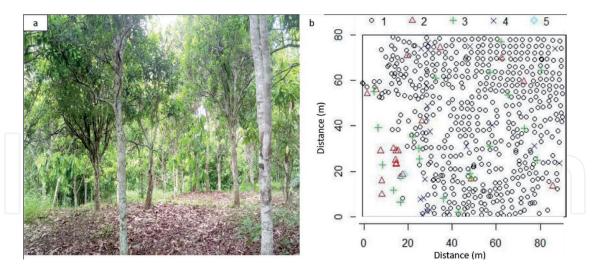


Figure 2.

Illustration of a cocoa-based agroforestry system planted with citrus trees (a) and the horizontal structure of its plant population (b). 1 = cocoa trees, 2 = various forest tree species, 3 = various other fruit tree species, 4 = citrus and 5 = palm trees.

## 3. Effect of spatial structure of citrus into cocoa based agroforest systems on citrus diseases

The spatial structure of a plant community is the vertical and horizontal arrangement of constituent elements [51, 60, 61]. It reflects therefore the local environment around each individual [55, 62]. Within agroforest, non-host plants

mainly perform a physical barrier effect on diseases [27, 51, 54]. The effect of the citrus spatial structure within CBAFS on the three main diseases affecting them in Cameroon has been assessed through various studies. A network of 27 plots in the three study sites was set up in Obala, Muyuka and Bokito sites. These are located in three ecological conditions into humid forest zone of Cameroon. CBAFS with at least 12 citrus trees in the plot area were selected. Each plot area was a square of at least 2500 m<sup>2</sup> (50 X 50 m). Plots were chosen in villages among the most productive areas and also representative of the study zone in terms of system diversity and variability of citrus species produced.

The analysis of the spatial structure of the citrus sub-population was done by the Ripley method [62]. Following the method illustrated in Ngo Bieng [55], a typology of spatial structure was build based on the spatial structure of the citrus trees in the study plots. In a first step, the horizontal spatial structure was characterized on the citrus trees in each plot, using the L(r) modified Ripley function [20, 61, 63]. The L (r) function is based on the calculation of the expected number of neighbor trees (**Figure 2**), within a distance  $\leq$  to r of any point of the study pattern. This method enables to distinguish three types of tree spatial patterns: regular when L (r) is <0, aggregated when L (r) is >0, and random when L (r) =0. This function characterizes the neighborhood structure around a point. It is used for a simple, homogeneous and isotropic point process of density  $\lambda$  [64].

$$\mathbf{K}(\mathbf{r}) = \lambda^{-1} \mathbf{E}(\mathbf{r}) \quad L_{(r)} = \sqrt{\frac{K(r)}{\pi}} - r \tag{1}$$

Subsequently, a hierarchical cluster analysis based on the Euclidean distance between the values of the L(r) function of the citrus trees in the different plots was made. It resulted in clusters of plots with a similar spatial structure, based on their trend to regular, random or aggregated spatial structure. This analysis was done with *ads* and *ade4* package R 3.2.2 software (**Figure 3**). Symptoms of CDP, PLFSD, and citrus scab were assessed by the visual recognition method. The intensity was assessed using a scale from 1 to 4.

From this study it emerges that, the spatial structure has a significant influence on the intensity of the diseases observed. The analysis of variance and the mean comparison test reveal that plots in which citrus have an aggregated spatial structure, have a high intensity of citrus scab disease and PLFSD. On the other hand, plots in which citrus fruits have a regular spatial structure show a significantly low intensity of these same diseases (**Table 1**).

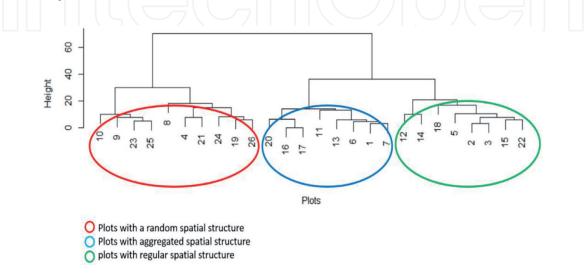


Figure 3.

Hierarchical classification of plots according to the spatial structure of citrus trees in the experimental plots.

Maladies fongiques	Aggregated spatial structure	Random Spatial structure	Regular Spatial structure	Anova/Tukey test	Df	F value	Pr(>F)	
<i>Pseudocercospora</i> leaf and fruit spot disease	1.55 ± 0.36 <sup>b</sup>	$1.43 \pm 0.28^{a}$	$1.41 \pm 0.26^{a}$		2	10.08	5.14e <sup>-7**</sup>	
Citrus scab disease	$1.22 \pm 0.56^{b}$	$1.11 \pm 0.38^{ab}$	$1.08 \pm 0.34^{a}$		2	5.102	0.00638	
Phytophthora foot rot disease	$1.59 \pm 0.93^{a}$	$1.97 \pm 0.94^{\rm b}$	1.78 ± 0.91 <sup>ab</sup>		2	6.297	0.002**	

**Table 1.**Effect of the spatial structure of citrus trees in cocoa based agroforests systems on diseases.

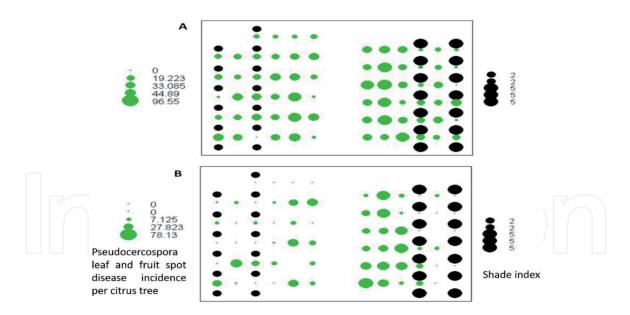
It is thus demonstrated that the aggregate spatial structure of citrus in CBAFS has a negative effect on diseases observed. These results are similar to those obtained by Ndo *et al.* [7, 8, 30] in the particular case of PLFSD. In addition to that, the involvement of spatial structure in the spread of various diseases has been shown [8, 53, 54]. Indeed, the aggregation of host populations favors the dispersion of diseases, while regularity would reduce it [65]. In addition, it is recognized that transmission of the pathogen decreases as the distance between host individuals increases [52, 58]. On the other hand, it has been shown that aggregation of host populations can reduce the incidence of pathogens [66]. Because the transmission of the pathogen between aggregates decreases with increasing distance between aggregates. These aspects would therefore explain the low intensity of CDP observed in plots where citrus fruits have an aggregated spatial structure.

#### 4. Effect of shade intensity management on CDP and PLFSD

Depending on the situation of citrus in the CBAFS and in relation to the upper stratum, three levels of shading (dense, moderate and no shading) were defined. Shade trees play various roles in tropical agroforests. They can improve adverse weather conditions by modulating temperature variations [51, 52, 57, 59]. Shading has been recognized as one of the factors that can influence PLFSD dissemination [54, 67, 68]. Given that shading favors climatic conditions for the development of certain citrus pathogenic fungi such as *P. angolensis* or many *Phytophthora* spp. (high relative humidity (>60%) and cool temperature conditions (<25°C)) [18, 28, 69]. It is assumed that within a plot, trees under shade would have a higher incidence of the disease than those in full sunlight. On the other hand, given the role of shade trees in improving climatic and nutritional conditions, the growth of trees under these conditions can be improved, as well as their vigor and response to disease. In addition, shade trees can act as a barrier against wind and rain (the main factors in the spread of conidia) and slow the progress of the epidemic.

An experiment carried out in a fruit trees orchard in Foumbot in the Western region of the country demonstrated the effect of shade trees on the PLFSD epidemic. The trial was set up in the Institute of Agricultural Research for Development (IRAD) experimental orchard. This orchard comprises collection plots of mango (*Manguifera indica*), avocado (*Persea americana*) and various citrus trees separated from each other by fallow plots often reserved for annual crops. This experimental plot enabled to compare tree shading situations ie dense shade (under mango trees), light shade (under avocado trees) and full sun light (fallow plot). One-year old pomelo seedlings have been placed in three levels of shade i.e.: under mango trees (dense shade), under avocado trees (light shade) and on fallow land (no shade).

The results of this experiment showed that the higher the shade index (under mango trees), the lower the PLFSD severity. When the shade intensity is lower (under avocado trees), disease severity is also lower, however the differences are not significant (**Figure 4**). These results suggest that shading should be sufficient to significantly reduce PLFSD incidence. Otherwise too little shading will not have significant effect on disease severity. But in the meantime, the plant must receive sufficient sun radiation for good growth. So, it is necessary to determine an optimum shading that allows a good compromise between plant growth and reduces PLFSD incidence. This optimum can vary according to the climatic and sanitary conditions of the plantations under consideration [18, 20, 52].



#### Figure 4.

Graphical representation of the percentage of diseased leaves for each pomelo plant and the shade indices of the different shade trees during the first observation date (A) and the second date (B) in the Foumbot plot.

Citrus diseases <sup>—</sup>	Shade conditions			Anova/	Df	F value	<b>Pr(&gt;F)</b>
	Full sunlight	Light shade	Dense shade	Tukey test			
Citrus scab disease	$1.08 \pm 0.34^{a}$	1.11 ± 0.41 <sup>a</sup>	$1.18 \pm 0.50^{a}$		2	2.257	0.106
<i>Phytophthora</i> foot rot disease	$1.92 \pm 0.91^{b}$	1.98 ± 1.00 <sup>b</sup>	$1.56 \pm 0.86^{a}$	_	2	11.18	1.8 e <sup>-6***</sup>

In the same column, values with same letter are not significantly different (Tukey HSD test P < 0.05). <sup>\*\*\*</sup> indicates highly significant.

#### Table 2.

Effect of shade on citrus diseases.

Another experiment conducted in 26 cocoa-based agroforestry systems showed the effect of shading on the spread of CDP and citrus scab disease. In this study, a total set of 476 citrus were observed under three shading conditions. Depending on the tree diversity and population, the various scenarios of shade density have been coded as follows: (1) "*dense shade*" when the citrus tree were placed under a direct and thick shading of the upper stratum; (2) "*light shade*" when the citrus tree received a mean shading of a higher stratum and finally (3) "*full sun*" when the citrus fruit did not receive a shade of a top stratum.

Results showed that, there is a variation in the citrus diseases intensity depending on whether they are located under dense shade, light shade or in full sunlight. The mean comparison test thus revealed significant differences in the intensities of CDP, according to the three citrus trees situations depending on shade. Citrus trees under dense shade are significantly less affected by CDP compared to those under light shade and those in full sunlight (**Table 2**).

Results therefore showed that, the shade had a significant effect on diseases. This shading effect was positive on the intensity of CDP. In general, in CBAFS, shade reduces diseases intensity [52, 70]. Indeed, for pathogens, spore dispersal and germination are the two main phases of their life cycle. Shading promotes spore germination while the sensitivity of the dispersion of pathogen spores to microclimate depends on how it is dispersed [7, 59].

## 5. Mode of dispersal of citrus infectious pathogens and CBAFS structural characteristics

The majority of fungi require moisture for infection and production of conidia [9, 71, 72]. These conidia may be disseminated by wind or soil water runoff for telluric fungi like *Phytophthora*. Local dispersal is primarily favored by rain-splash as well as some insects moving on trees [24]. This mode of dissemination determines the spatial distribution of each disease.

In the case of PLFSD, the analysis of its spatial distribution indicates that the disease is distributed in clusters, and that above 12 m there is no spatial dependency. In fact, the disease spreads from one tree to its closest neighbors depending on wind speed and/or rainfall intensity. Infection will depend on the presence and quality of the host. If neighbors are susceptible hosts, infection continues and the epidemiological cycle continues. Otherwise, the course of the disease can be circumscribed. This may explain the aggregated spatial structure of diseases that usually have this mode of spread. However, Brown and Bolker [65] pointed out that the aggregation of host populations favors the dispersal of the diseases while its regulation reduces it. That is, the further away the trees are from each other, the slower the transmission of the disease [22].

With regard to CDP, the spread of *Phytophthora* in the field is primarily ensured by the use of infected plant material [32]. However, mechanical means of dispersal of *Phytophthora* have been illustrated. Cases of transmission from an infected root to a healthy root following their respective growing zones have been reported. The inoculum can also be spread by run-off water. Splashes can promote the spread of the inoculum from the soil to the aerial parts of the plant. This mode of disease spread may be promoted by aggregation of the host species of this pathogen. This hypothesis was confirmed by Akoutou *et al.* [7, 30]. These studies showed that citrus with an aggregated spatial structure were more attacked by CDP in contrast to those with a regular spatial structure. In fact, root diversity in the rhizosphere could limit contact between roots of the same species. Host trees planted at wide spacings and having non-host trees between them are less likely to come into contact and this would help to limit the spread of the inoculum. This effect of dilution of the pathogen's resource can also be applied to the mode of transmission through diseased fruits and contact with parts of the plant close to the soil.

In addition, it was shown that environmental factors play a critical role in the development, severity, dispersal and conservation of inoculum in the epidemiology of *Phytophthora* disease. The increase in temperature favors population growth of species such as *P. parasitica* and *P. palmivora*. Hot, dry climates are favorable to *P. parasitica*. These observations corroborate the conclusions drawn on the effect of shading on CDP development. Indeed, the cooler environmental conditions in the understory created by shade trees would make the habitat unfavorable for the pathogen. Citrus planted under dense shade would therefore be less exposed to the inoculum, which is therefore more intense in plot areas of the plot where there is no shade.

#### 6. Conclusion

This study highlighted the effect of shading trees on citrus in agroforestry plots. In such plots citrus trees are mixed with plants belonging to different tree. Spatial structure has a significant influence on the observed diseases intensity. Plots in which citrus have an aggregated spatial structure have a high intensity of studied diseases, while plots in which citrus have a regular spatial structure are significantly

less attacked by these diseases. Optimizing the structural characteristics of CBAFS could lead to the development of integrated control strategies against fungal diseases. These management strategies will be adapted to local agroecological contexts, respectful of the environment, and applicable by smallholders.

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