Assessment of yield and economic losses caused by pests and diseases in a range of management strategies and production situations in coffee agroecosystems

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FOREWORD

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THESIS SUMMARY

Crop losses due to pests and diseases are a major threat to incomes and food security of thousands of rural families worldwide. The assessment of crop losses (yield and economic losses) and their causes is needed to improve the development of agroecosystems capable to offer good crop yields, regulation of pests and diseases, and other ecosystem services. This doctoral research aimed to contribute to the research field of crop losses, by providing experimental and modeling approaches that could be used in perennial crops to estimate primary and secondary losses and analyze their causes. We worked in a perennial crop such as coffee, in Turrialba, Costa Rica, where coffee is grown in plantations from monocultures at full sun exposure to highly diversified agroforestry systems, and under a range of production situations (topoclimate, soil fertility, types of shade) and management strategies (agricultural practices and inputs). The three main research questions were: What is the impact of management strategies and production situations on pests and diseases and coffee yields? How do coffee yield losses caused by injury profiles vary in function of management strategies and production situations? Which types of coffee agroecosystems are capable to obtain the lowest coffee losses (yield and economic) and highest overall benefits (ecosystem services)?

This research was developed through two experimental designs. The first was an experimental coffee parcel under controlled conditions (three-year experiment) to quantify primary and secondary yield losses by comparison of treatments, and to identify the main predictors of yield losses by structural equation modeling. The second experimental design was based on surveys in a coffee research plot network (coffee plots of smallholder farmers), where, during two years, we measured indicators of yields and indicators of four ecosystem services: regulation of pests and diseases, provisioning of agroforestry products, maintenance of soil fertility, and carbon sequestration. Yield losses in this network were estimated through modeling using the predictors identified in the experimental coffee parcel. Analyses of data included several statistical techniques, from analysis of variances, linear regressions to multivariate techniques.

The results were organized in four manuscripts, and then discussed integrally. The main findings were: i) Both production situations and management strategies determine coffee yield and pest and disease injuries, effects of interactions altitude x management x types of shade must be considered; ii) Injury profiles depend on particular combinations of production situations and management strategies, with impacts on yield losses especially in a year of high coffee production (primary yield losses), but compromising also the yields of the next year (secondary yield losses); iii) Diversified agroforestry systems have better chances to regulate pests and diseases (reduce yield and economic losses), and simultaneously provide goods for family benefits, maintain soil fertility, and increase carbon sequestration, without implying trade-offs among these ecosystem services.

The main prospects of this research are related to perform similar studies in coffee and other perennials at regional levels, develop an injury profile simulator for coffee, and prototyping of coffee agroforestry systems to optimize the provision of multiple ecosystem services.

PART I. INTRODUCTION

Chapter 1. Problematic/Definitions/Research questions

1.1 Crop losses worldwide

Crop losses are a major threat to the wellbeing of rural families, to the economy of traders and governments, and to food security worldwide (Zadoks and Schein, 1979; Savary and Willocquet, 2014; Avelino *et al.*, 2015). Crop losses due to pests and diseases for major food and cash crops (rice, wheat, barley, maize, potatoes, soybeans, cotton, and coffee) were estimated between 20 and 40% at country and regional levels in different continents (Oerke *et al.*, 1994; Oerke, 2006). Attacks of pests and diseases can occur in the agricultural land during the production cycle (pre-harvest) and/or during the storage (post-harvest). In both stages, the yield of the crop product as well as its quality can be reduced (Savary *et al.*, 2006b), which implies that financial returns will be also compromised because of less production to sell and/or less quality to offer to buyers (Nutter *et al.*, 1993). Furthermore, implications of crop losses can reach levels far beyond farms, given that a reduced production can affect entire rural communities and regions, national markets and exportations, and at broadest level, the food availability for the world population.

Given the negative implications of crop losses, strategies and measures at different levels (from farms to governments) are needed to reduce them, and must be based on reliable assessments. Quantification of crop product losses and a better understanding of their drivers have been mentioned as essential to (i) evaluating the efficacy of crop protection practices (Oerke, 2006), (ii) making better decisions for integrated pest management (Savary et al., 2006a), (iii) assessing the sustainability of agricultural production systems (Cooke, 2006), (iv) evaluating the effectiveness of pest and disease regulation as an ecosystem service (Avelino et al., 2011; Allinne et al., 2016), and (v) guiding government agencies and other potential donors about where, how and when allocate resources for better control of pests and diseases, and therefore avoid crop losses (Cooke, 2006). The results of these evaluations and guidance could contribute to the design of better practices to reduce the incidences of pests and diseases, as well as to the design of agroecosystems with characteristics (structure-composition-management) aimed to reduce crop losses (Avelino *et al.*, 2015).

Despite the importance of the assessment of crop losses, efforts to quantify them and analyze their causes are considered still scant (Cheatham *et al.*, 2009; Avelino *et al.*, 2011; Savary and Willocquet, 2014). This is an issue for all crops in general, and specially for perennial crops. The most remarkable efforts in the last two decades were concentrated in the quantification of relative yield losses due to pests and diseases through experiments and surveys. In annual crops, for instance, yield losses of: rice ranged from 24% to 41% in Asia (Savary *et al.*, 2000a), potatoes from 5% to 96% in France (Rakotonindraina *et al.*, 2012), cotton up to 100% in Thailand (Castella *et al.*, 2007). In perennial crops, such estimations are really scarce: on apple and other stone fruits yield losses reached up to 5% in the Netherlands (van Leeuwen *et al.*, 2000), in coffee yield losses ranged from 13% to 45% in Brazil (Barbosa *et al.*, 2004). Explicit monetary valorizations of yield losses due to pests and diseases is even scarcer; at least in the scientific literature, they cannot be found.

The scarcity of quantifications of crop losses and analyses of their causes is related mainly to the difficulty of their assessment. Most attempts to measure yield reductions (losses) were based on relationships between yields and an indicator of a given pest or disease. However, farmers usually do not face only one pest or disease, and their crops can be exposed to different conditions (Savary *et al.*, 2006a). Such relationships therefore can be masked by several confounding factors such as interactions with other pests and diseases, and interactions with other factors such as environment (temperature, rainfall), soil fertility and others (Avelino et al., 2006; Cooke, 2006). Apart from the yield reduction of a specific year, there can be reduction of the yielding capacity in the future, which in annual crops, for instance, is given by the negative effects of pathogens inoculum that remains in soil; whereas in perennial crops is given by the death of tissues and/or reduction of reserves (Zadoks and Schein, 1979). The economic valorization of losses needs basically the data of yield losses, but also other variables such as the costs of production, prices and other economic drivers (Nutter *et al.*, 1993; Avelino *et al.*, 2011) which may make difficult the task. To overcome these difficulties an holistic view is needed, involving multidisciplinary research (ecology, epidemiology, biology, agronomy, economics and others) (Savary *et al.*, 2012).

1.2 Coffee crisis in America

The coffee sector in Latin America and the Caribbean is currently suffering one of its worse crises ever, due to the combination of negative climatic, biophysical and socioeconomic factors. Since 2012, there has been a noticeable reduction of coffee production caused especially by the outbreak of coffee leaf rust (*Hemileia vastarix* Berkeley and Broome); considered as a result of the combination of economic and climatic factors: the decreasing coffee prices and increasing production costs lead to a suboptimal management of coffee plantations, and at the same time, an important reduction of the diurnal thermal amplitude apparently favored the development of the pathogen (Avelino *et al.*, 2015). Other reported problems were the use of susceptible coffee varieties to the disease; inadequate policies and lack of investment; and the lack of efficient research and extension services, which resulted in a deficient technical assistance and training to farmers (PROMECAFE, 2013). The decrease of production was estimated from 2% to 28% for the year 2012-2013, around 30% for 2013-2014, and the production continued low the following years since many plantations were stumped (Baker, 2014). This situation has reduced incomes of household economies for several years in a row and still threatens the food security of rural families direct and indirectly involved in coffee production, and almost broke several industries (FEWS-NET, 2014; Avelino *et al.*, 2015).

Pests and diseases represent one of the main problems of coffee production, as the unexpected outbreak of coffee leaf rust. Although this disease is considered the most important in American coffee growing regions nowadays, there are also other diseases such as American leaf spot (*Mycena citricolor* Berk. and Curtis), brown eye spot (*Cercospora coffeicola* Berk. and Curtis), anthracnoses/dieback (*Colletotrichum spp.*) and others, and pests such as coffee berry borer (*Hypothenemus hampei* Ferrari) and leaf miner (*Leucoptera coffeella* Guérin-Mèneville), which constantly threaten the coffee production and even more since they were forgotten in order to attend in urgency coffee leaf rust (PROMECAFE, 2013).

Along with the decrease of coffee production, there is also a risk of degradation of biodiversity and ecosystem services in coffee growing areas. Important ecosystem services that agroforestry systems with perennial crops are capable to provide are: provision (coffee, fruits, timber, others), carbon sequestration, soil conservation, and regulation of pests and diseases (Beer *et al.*, 1998b; Rice, 2011; Jha *et al.*, 2014; Pumariño *et al.*, 2015). However, since 1990 to the present, globally the areas for coffee have decreased in 8% and the intensification of coffee production has increased, involving the elimination of shade trees for simplification of agroforestry systems (Jha *et al.*, 2014). For instance, 50% of coffee areas have disappeared in the Volcan Central Talamanca Biological Corridor in Costa Rica (Bosselmann, 2012), and 35% in southern Guatemala (Haggar *et al.*, 2013). These reductions includes also the loss of trees and other vegetation, consequently involving the loss of biodiversity and ecosystem services (De Beenhouwer *et al.*, 2013).

1.3 Research needs

From the scientific point of view, first there is a need to increase the knowledge on crop losses and their causes for making better decisions for integrated pest management (IPM), and for developing agroecosystems capable to balance good yields and regulate the impacts of pests and diseases through ecosystem services (Savary *et al.*, 2006a; Avelino *et al.*, 2011). The recent coffee rust crisis also highlighted the difficulty to assess the seriousness of the crisis due to the lack of knowledge on losses due to diseases; this lack of knowledge can explain that the responses in aid for farmers were slow in coming, particularly economic responses from international funding agencies (Avelino *et al.*, 2015).

The main regional coffee organizations, technicians and scientific community in Latin America have demanded several actions to cope with the production decrease due to the current coffee crisis. Among them, there is a necessity to analyze the long term viability of the coffee crop, based on new studies on risks, costs, benefits and alternative production models, from full sun coffee to diverse coffee agroforestry systems; with the aim to develop adequate strategies for coffee production in the long term (PROMECAFE, 2013). There is also an urgent need to ensure that farming systems not only provide high yields, but also provide ecosystem services on which agriculture and farmer households depend (Cheatham *et al.*, 2009; Vignola *et al.*, 2015).

1.4 Definitions to state the scope of this research

The study of crop losses and their causes involves several key terms and concepts, which need to be clearly defined in order to facilitate the understanding of the objectives, the main results and their implications. In this research, the most recognized definitions in the literature related to crop losses were used; in case of important variations in the definition of a given term from different sources, the author established the most suitable definition according to the research questions and purposes. The key definitions for this research are described below.

1.4.1 Injury profile

An injury is any symptom or sign caused by a pathogen or pest, then, is any observable alteration of the normal healthy crop development (Nutter *et al.*, 1993). An injury profile is a given combination of injury levels caused by a set of pests and diseases in a crop cycle (Savary *et al.*, 2006b).

1.4.2 Crop loss

For some authors, crop loss is the reduction in quantity and/or quality of the crop yield (yield loss) due to biotic or abiotic factors, which can occur in the field (pre-harvest) or in the storage (post-harvest) (Oerke, 2006). Such reductions are also known as crop damage (Savary *et al.*, 2012). For others, crop loss also includes the reduction in value and/or financial returns due to yield loss (Nutter *et al.*, 1993).

This research is focused on pre-harvest losses caused mainly by biotic factors. Here therefore, crop loss is considered to involve yield losses caused by pests and diseases, as well as the monetary valorization of such losses.

1.4.3 Yield loss

Yield loss is the quantitative decrease of the crop yield caused by a single injury or by an injury profile. The yield loss is the difference between attainable yield and actual yield, and can be expressed in terms of weight or volume, or as relative yield loss (%) with respect to the attainable yield (Nutter *et al.*, 1993; Savary *et al.*, 2006b).

1.4.4 Attainable yield

Attainable yield is the yield without the negative effects of yield reducing factors (especially pests and diseases), limited only by yield defining factors (radiation, temperature, crop phenology and physiology) and limiting factors (water and soil nutrients) (Zadoks and Schein, 1979; Rabbinge, 1993; Savary and Willocquet, 2014). Under this broad definition, some authors define the attainable yield as the maximum yield registered at community or regional levels (Mueller *et al.*, 2012; Wang *et al.*, 2015). Others define attainable yield as the site-specific yield achieved under the environmental conditions of the site and with the best available production techniques to avoid biotic stress caused by pests (Nutter *et al.*, 1993; Oerke *et al.*, 1994). Others define the attainable yield as the yield achieved in a given production situation when the crop was not exposed to reducing factors such as pests, taking into account that for these authors the production situation is site-specific and includes a specific management (Savary *et al.*, 2006b). Textual definitions of production situations are given in Table 1.1.

In this research, it is considered that the definitions of attainable yield given by Nutter *et al.* (1993) and Oerke *et al.* (1994), have two important similitudes: both consider that attainable yield is site specific and is achieved with the local production techniques, and both consider that it should be achieved in absence of pests. These definitions are considered the most suitable for the approaches and objectives of the present research, and therefore are accepted.

Thus, for this research, an adapted definition of attainable yield is: the site-specific yield achieved under the local environmental conditions in combination with the available cropping practices (labors and inputs), trying to minimize the negative effects of pests and diseases. This definition implies that each plantation has its own attainable yield.

It is important to mention that the achievement of an attainable yield can involve high costs to control any pest or disease, and thus, would not be always the best economic yield; that is why this yield is considered to be theoretically independent from economic factors (Avelino *et al.*, 2011).

Table 1.1. Definitions of attainable yield

Definition	Source
"The attainable yield is the site-specific maximum yield that can be obtained under the geographic and ecological conditions at a location, using the best production techniques to avoid biotic stress. It is determined by, among other factors, climate, latitude and the variety grown. For the purposes of this study, it is defined as the yield attainable in the absence of pests using the site-specific methods of cultivation ('no loss scenario')"	(Oerke *et al.*, 1994)
Attainable yield: "the site-specific yield obtained when crops are using all available pest control technologies to minimize biotic stress"	(Nutter *et al.*, 1993)
"Attainable yield: a reflection of a given production situation; the yield performance of a crop that has not been exposed to yield-reducing factors, especially pests"	(Savary *et al.*, 2006b)
"Attainable yield (Yatt)—the highest yield in the surveyed region obtained from on-farm surveys"	(Wang *et al.*, 2015)

1.4.5 Actual yield

The definition of actual yield is consistent in the most important literature on crop losses, and so is also accepted in this research: the actual yield is the site-specific yield achieved using the available resources and current practices (labor and inputs) of the farmer, generally affected by pests and diseases (Nutter *et al.*, 1993; Savary *et al.*, 2006b; Savary and Willocquet, 2014). In this research, as well as for attainable yield, it is considered that each plantation has its own actual yield.

1.4.6 Economic loss

Economic loss, basically, is any reduction in economic benefits due to crop damage; the economic loss should also consider the costs of labor, materials and inputs for the control of pests and diseases applied to reduce damages (Savary *et al.*, 2006b; Cheatham *et al.*, 2009; Avelino *et al.*, 2011). It is also mentioned that the economic loss should be calculated as the difference between the maximum economic profit and the economic profit obtained with the actual yield (Zadoks and Schein, 1979; Avelino *et al.*, 2011). However, the assessment of the maximum economic profit (economic yield) is rather difficult because it is not necessarily determined by a maximum yield or by an attainable yield.

In this research, the scope of the estimation of economic losses goes up to the monetary valorization of the reduced yields (yield losses) due to pests and diseases.

1.4.7 Primary and secondary crop losses

Primary crop loss is the reduction of yield and/or quality of the crop product caused by pests and diseases in the current crop cycle; this loss may result in a loss of income and/or in increased production costs (Zadoks and Schein, 1979; Nutter *et al.*, 1993).

Secondary crop loss is the “loss in yielding capacity of future crops” (Zadoks and Schein, 1979). In annual crops, secondary losses are caused by inoculums of pathogens that remained in soil, seeds or tubers which are going to reduce the yields of the new crop sowed in the same terrain; whereas in perennial crops, secondary losses are caused by defoliations and other negative physiological effects caused by pests and diseases, which lead to loss of vigor of plants and therefore reduce the production in the next years (Zadoks and Schein, 1979). For instance, in a perennial crop such as coffee, dead branches resulting from the attack of pests and diseases in a given year, are not going to bear fruits anymore, representing that way secondary losses (Avelino *et al.*, 2015). In economic terms, secondary losses should include the loss of income and/or the costs to manage soil and seeds, costs of production and renewal of plantations if necessary (Nutter *et al.*, 1993).

In this research, therefore, for a perennial crop such as coffee, it is considered that each year there are primary and secondary crop losses, with these definitions: primary crop loss is the loss caused by pest and disease injuries in the current harvesting year of coffee; and secondary crop loss is the loss resulting from negative impacts of pests and diseases injuries caused in the previous year.

1.4.8 Production situations

The production situation is a broad concept because it involves several conditions under which the crop is grown, which leads to some differences in its definition. Some authors consider that the production situation includes the social, physical, biological, economic and technical context where an agricultural production takes place; this definition includes the management (cropping system), in the sense that farmer´s skills are part of the production situation (Savary *et al.*, 2006b). That means that a specific biophysical context may comprise several production situations and, as a consequence, that there is no necessarily a geographic continuity among plots belonging to the same production situation. Other authors consider that a production situation is defined by the biological, chemical, physical, socioeconomic and environmental conditions under which a crop is grown, excluding the management from the definition [(Aubertot and Robin, 2013) adapted from (Breman and de Wit, 1983)] but recognizing that in a given context of production, farmers do not apply the same management. Textual definitions of production situations are given in Table 1.2. Although including or excluding management from the definition of production situation may appear as being mostly part of a semantic debate, there are some key implications, particularly when defining the attainable yield. The attainable yield actually is the yield obtained in a given production situation with no pests and diseases. Depending on authors, crop management will be included or not when calculating attainable yield. In addition, pest and disease attack levels which are the result of the interaction between the physical and the biological environment (including the host plant and the pest or pathogen) and actions conducted by the farmer (Zadoks and Schein, 1979), a reflection of socio-economic conditions, are then dependent on a production situation, or on a combination of a production situation and a particular management, depending on authors.

We retained the second meaning of production situation, i.e. with no crop management included, as used recently is studies related to the topic (Aubertot and Robin, 2013; Robin *et al.*, 2013; Aouadi *et al.*, 2015). The definition of production situation given by Aubertot and Robin (2013) adapted from Breman and de Wit (1983), enabled us to put the same emphasis in both production situations and management strategies. Besides, we considered that injury profiles, physiological conditions of coffee plants, yield and yield losses, are affected by a given combination of a production situation and a management strategy in each plantation. However, we maintained the idea that the attainable yield is dependent on crop management, which seems particularly true in perennial crops as coffee, where some characteristics of the plantation (plantation distances, shade type, aged the coffee tree) cannot or are difficult to be changed.

Thereby, in this research, the production situation is defined as the set of biological, chemical and physical components of the agroecosystem, and the socioeconomic and environmental (climate and topography) conditions under which the agroecosystem takes place. Soil fertility was considered as a chemical component. And, given that this research dealt with agroforestry systems, the type of shade (shade canopy) was included as an important biological component. A type of shade includes the botanical composition (species richness of herbaceous plants, bushes, palms and trees) and the structure (abundances, basal areas and shade cover) of the coffee shade canopy in this case. Given that this research was focused on the study of the crop (yields) and of injury profiles, both were excluded from the biological component of production situations. This latter decision is convenient when such components are the objects of study in order to put more emphasis on them, and to well differentiate them from other components in the study (Savary *et al.*, 2006b).

Table 1.2. Definitions of production situation

Definition	Source
"Production situation: the physical, biological, technical, social, and economic context in which agricultural production takes place"	(Savary *et al.*, 2006b)
We characterize the production situation of each field, that is, the set of factors—physical, biological, and socioeconomic—that determine agricultural production (7,22). The latter group of (socioeconomic) variables is assumed to be indirectly reflected by several characteristics of the patterns of cropping practices, such as inputs (fertilizers and pesticides), the method of crop establishment, weed control practices, and crop rotation	(Savary *et al.*, 2000b)
"A production situation has been defined as the bio-physical and socio-economic environment where agricultural production takes place (Penning de Vries & Van Laar 1982; Breman & De Wit 1983; Rabbinge & De Wit 1989). A production situation may thus be decomposed into components which account for the physical, biological, economic, social, and technological environment of agriculture"	(Savary *et al.*, 2016)
"Production situation is defined by the physical, chemical and biological components, except for the crop, of a given field (or agroecosystem) and its environment, as well as socio-economic drivers that affect farmer's decisions (adapted from Breman & De Wit 1983)" "In a given production situation, a farmer can design several cropping systems according to his goals, his perception of the socio-economic context and his environment, farm organization, knowledge and his cognition"	(Aubertot and Robin, 2013)
"The concept of production situation (PS) was defined as "the physical, chemical and biological components of a given field and its environment that are not directly managed by the farmer, as well as socio-economic drivers that affect his decisions (Aubertot and Robin, 2013 adapted from Breman and de Wit, 1983)"	(Aouadi *et al.*, 2015)

1.4.9 Management strategies

Each farmer makes decisions on what components he will establish/maintain in his plantation, and what cropping system he will apply according to his preferences, objectives and socioeconomic conditions. Therefore, in this research, it is considered that a management strategy is composed mainly by the agricultural practices and inputs (cropping system) applied to the agroecosystem; and that a management strategy can affect components of the system such as the crop, injury profile, soil, and the type of shade in the case of agroforestry systems. The management strategy can easily change from one year to another in terms of agricultural practices and inputs applied to the main crop of the system (coffee in this case); whereas it is difficult that the management affects the type of shade from one year to another. The strategy of maintaining a given type of shade usually is a long-term decision made by the farmer.

Related to management, the term management intensity was also used in this research. It was considered that: the more practices and inputs applied to the main crop (coffee), the more intensive the system.

1.5 The importance of injury profiles for crop loss assessments

It is assumed that each cropping system, which depends on the farmer´s decisions under a given production situation, leads to a particular injury profile (Aubertot and Robin, 2013); and each injury profile can have a specific impact on crop losses (Avelino *et al.*, 2011). Both important statements mention the injury profile and not individual injuries, suggesting that analyzing the incidences or severity of only one pathogen and its impact on yields separately from other pathogens would not be enough for comparing different agroecosystems and determining which the best is for avoiding crop losses. For instance, an agroecosystem A could have less attack of a given disease than an agroecosystem B, but its crop loss could be higher due to other set of pathogens. Therefore, it is necessary to identify and analyze all the pests and diseases involved (injury profile), the management strategy and the production situation; and then, translate the impact of that injury profile in a crop loss data.

The study of injury profiles involves multiple pathosystems, and requires the intervention of several disciplines to be successful on giving the scientific bases for designing and implementing IPM (Savary *et al.*, 2006a). For understanding and explaining injury profiles, it is necessary to study a range of cropping systems and production situations. Pest and disease outbreaks strongly depend on rainfall patterns and altitude which determines temperatures. In coffee agroecosystems, outbreaks also depend on important practices such as fertilization, pruning and regulation of shade, which are capable to modify the microclimate and the physiology of the crop (Avelino *et al.*, 2004). Biophysical factors such as topography (inclination and orientation of the slope, altitude), distance between coffee rows, coffee tree height, type of shade (structure and composition) and shade percentage must be also measured, because they have effects on the development of pathogens (Avelino *et al.*, 2007). The soil fertility can affect the nutritional status and vigor of coffee plants, and consequently has relationships with injury levels (Avelino *et al.*, 2006). The relationship between biodiversity and pest and disease regulation is particularly important since there can be positive effects but also negative;

for this matter, it is important to determine both the functional diversity and the botanical composition of the system (Cheatham *et al.*, 2009).

1.6 How to assess crop losses

Currently, there are several approaches and methods suggested to quantify, estimate, model, and to generate different types of knowledge on crop losses; they can be combined and applied for coffee losses research. Several types of knowledge are empirical environment-disease relationships, empirical disease-crop loss relationships, mechanistic simulation models–single disease, mechanistic simulation models–multiple diseases, risk zoning–disease prevalence, and production situation-based models; these types of knowledge are useful for making decisions of different categories: tactical–during season decisions, strategic–between season decisions, strategic–domains for management, and strategic–research priorities (Thornley and Johnson, 1990; Cooke, 2006; Savary *et al.*, 2006b). Important tools for modeling such as the Injury Profile Simulator (IPSIM) using qualitative data (Aubertot and Robin, 2013), or the XPEST platform for modeling yield losses caused by injury profiles with quantitative data (Aubertot *et al.*, 2014) are currently available for generating knowledge and predictions useful for designing innovative cropping systems as part of strategies for a better control of pests and diseases.

The assessment of crop losses becomes more complex in perennial crops such as coffee due to indirect effects of several diseases and their outbreaks impacts on the yields of following years. Crop losses caused by leaf diseases such as coffee leaf rust, American leaf spot or brown eye spot for instance, are harder to quantify because they do not affect directly the fruits like other pathogens do (e.g. coffee berry borer, anthracnose), but they do affect indirectly the production of fruits through alterations of the plant physiology (Avelino *et al.*, 2006).

Besides, when quantifying crop losses in perennial crops, there can be involved together primary and secondary losses due to the effect of a pest or disease outbreak in previous years (Zadoks and Schein, 1979). Therefore, several methods should be utilized in order to assess the overall direct and indirect effects of injury profiles and to determine correctly what primary or secondary losses are.

Surveys, experiments and modeling are the most important methods to quantify crop losses. Results combining these methods could be stronger and more reliable than individual results of each one. Results of surveys and experiments can be incorporated in the modeling. The modeling of yield losses involving multiple pests and diseases rises as one of the main methods for a full understanding of the system and to hierarchize pathogens according to the damage they cause; such understanding is a key aspect for developing long-term strategies in order to determine where an IPM is needed, and for prioritizing the pests and/or diseases which needs further research (Savary *et al.*, 2006b; Savary and Willocquet, 2014). That kind of models for assessing losses of tropical crops, including coffee, does not exist at the moment.

1.7 Regulation of pests and diseases and other ecosystem services

The regulation of pests and diseases is an ecosystem service that biodiversity can provide (MEA, 2005). However, in the case of coffee agroforestry systems, the associated plant biodiversity could have either positive or negative effects on the regulation of pathogens. On one hand, shade trees can hamper noxious pathogens by favoring their natural enemies, by forming barriers to the movement of infectious propagules or pests, and by modifying microclimate of the understory (Staver *et al.*, 2001). Trees could also be capable to increase nutrients in soil, which are important to increase plant resistance to pests and diseases (Avelino *et al.*, 2011). On the other hand, shade conditions (especially high dense shade) could favor the development of given pathogens including fungi and insects, and some trees and plants could be hosts of other pathogens (Avelino *et al.*, 2007; López-Bravo *et al.*, 2012).

The positive or negative effects of shade therefore could depend on the type of pathogen and on the structure of the shade canopy. The management of trees/bushes/palms could become a key aspect depending on the injury profile that is aimed to prevent or control. The effects of the heterogeneity of different trees or plant species on the presence of pathogens and their severities, can be determined by comparing the ecosystem services provision between coffee agroforestry systems and intensive coffee monocultures (Cheatham *et al.*, 2009). Furthermore, the regulation of pests and diseases should be assessed through the quantification of avoided crop losses, which includes both yield losses and economic losses (Avelino *et al.*, 2011).

Using biodiversity for pests and diseases control can provide other benefits crucial for future perspectives. For instance, the environment health and water quality of farms and communities would be better due to the reduction of the use of pesticides, along with better conditions for farm workers, animal biodiversity and even increase in carbon sequestration (Avelino *et al.*, 2011). The reduction of farmer´s dependence on pesticides could also be achieved through the “vertical integration” (use of several control methods) and “horizontal integration” (management of a set of pests and diseases simultaneously) (Aubertot and Robin, 2013); regulation of pests and diseases can be part of both types of integration. Another reason for understand the role of biodiversity and ecosystem services is related to the situation of coffee farmers in the face of climate change. It is suggested that, among the adaptation measures of rural families’ livelihoods, it would be necessary to implement highly diversified coffee agroforestry systems, in order to regulate microclimates, to protect coffee plants from strong winds, high temperatures and long dry periods, and to diversify the source of products and incomes (Schroth *et al.*, 2009; Jha *et al.*, 2014).

Apart from the regulation of pests and diseases, there are other major ecosystem services of interest for farmers and for the society in general. For instance, diversified production in agroforestry systems become an important provisioning service, given that such systems, apart from the main crop product, can provide fruits, timber, firewood and others (Rice, 2008). The maintenance of soil fertility is another service of interest for the land owners, given that their production depends in great part on the state of this resource. For the society in general, the carbon sequestration is an essential service to contribute to the mitigation of climate change (MEA, 2005; Müller *et al.*, 2015)

1.8 Justification of the research

This doctoral research has developed and combined methods original or suggested by the scientific literature in order to quantify and analyze coffee yield and economic losses, caused by injury profiles, in a range of management strategies and production situations. Experiments, surveys and modeling were combined to achieve that goal. The research took place in Turrialba, Costa Rica, an important coffee growing area where it is possible to find coffee being cultivated under different conditions of topography and altitudes, types of shade and management intensities. The main results were expected to be useful for a better understanding on how coffee losses occur, and to identify the combinations of management strategies and production situations in which coffee agroecosystems are capable to regulate pests and diseases, while providing other ecosystem services for smallholder farmers. This knowledge is necessary to design and manage sustainable agroecosystems.

1.9 Research questions and hypotheses

Major research question: What is the impact of injury profiles on coffee yield and economic losses under different management strategies and production situations?

Major hypothesis: Pests and diseases can be the same in coffee agroecosystems, but the management strategies and production situations affect the injury profile and its impact on yield and economic losses.

The main research questions and intermediary questions are listed here and they can also be located in a scheme showing the variables and relationships studied in this research (Fig. 1.1):

Q1 What is the impact of management strategies and production situations on pests and diseases and coffee yields?

The following hypothesis and intermediary questions are addressed in chapters 3 and 5

Hypothesis: The levels of pests and diseases injuries, and coffee yields, depend mainly on the interactions between management intensity and the type of shade of coffee agroecosystems

- **Q1.1** What is the impact of management strategies and production situations on pests and diseases injuries? *chapter 3*
- **Q1.2** What is the impact of management strategies and production situations on actual yields and attainable yields? *chapters 3 and 5*

Q2 How do coffee yield losses caused by injury profiles vary in function of management strategies and production situations?

The following hypothesis and intermediary questions are addressed in chapters 4 and 5

Hypothesis: The impacts of injury profiles on coffee yield losses differ from one year to another, depending on various combinations of management strategies and production situations

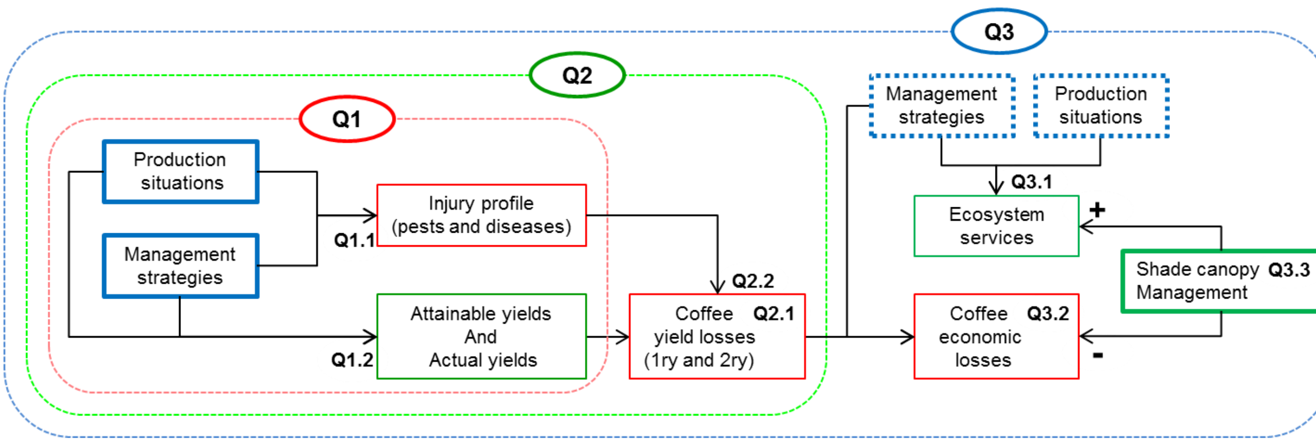
- **Q2.1** How to estimate, by experimentation and modeling, primary and secondary coffee yield losses caused by pests and diseases? *chapter 4*

- **Q2.2** What is the impact of injury profiles on primary and secondary coffee yield losses from one year to another, under different management strategies and production situations? *chapter 5*

Q3 Which types of coffee agroecosystems are capable to obtain the lowest coffee losses (yield and economic) and highest overall benefits (ecosystem services)?

The following intermediary questions are addressed in chapters 3 and 6

- **Q3.1** What is the effect of management strategies and production situations on the provision of ecosystem services (provisioning of agroforestry products, carbon sequestration, soil fertility, regulation of pests and diseases)? *chapter 3*
- **Q3.2** What are the economic losses caused by pests and diseases? *chapter 6*
- **Q3.3** What are the shade canopy and management characteristics of the coffee agroecosystems capable to obtain the lowest coffee losses and highest ecosystem services? *chapter 6*



See the main questions and intermediary questions (Q) in section 1.9

Fig. 1.1. Scheme of the variables and relationships studied in order to respond the main and intermediary research questions

PART II. MATERIALS AND METHODS

Chapter 2. Materials and methods

2.1 General strategy to respond the research questions

This research was developed through two experimental designs whose results were articulated in order to respond the research questions. The first was an experimental coffee parcel at full sun, with different controlled treatments to control pests and diseases during three years. This experiment was funded by the project DAMAGE (INRA, SMaCH metaprogram) and was called Coffee Losses (CoLosses). The second experimental design was based on surveys in different coffee plots of small farmers under a range of production situations and management strategies during two years. Such plots conformed a coffee research plots network, which was called CASCADE, given that it was part of the CASCADE project ("Ecosystem-based Adaptation for Smallholder Subsistence and Coffee Farming Communities in Central America") funded by the International Climate Initiative (ICI). Results of both experimental designs derived from several analyses and modeling enabled to respond the research questions one and two **(Q1 and Q2)**; analysis of ecosystem services in CASCADE permitted to respond research question three **(Q3)**.

CoLosses was dedicated to:

- Calculate primary and secondary yield losses jointly and separately through the comparison of different treatments, and determine the proportions of primary and secondary losses (**Q2.1**).
- Identify, through modeling techniques, the most important and easily measurable predictors of yields and yield losses (**Q2.1**).

Articulating the findings in CoLosses with the analyses in CASCADE:

- CoLosses offered key insights and recommendations to develop models for the estimation of primary and secondary coffee yield losses caused by pests and diseases. The most important predictors of yield and yield losses identified in Colosses, were used in CASCADE to develop statistical models for the estimation of primary and secondary yield losses in each plot of this network.

- Observations and learnings from the field work in both CoLosses and CASCADE, combined with information in the scientific literature and with the knowledge of the author and advisors of this research, permitted to develop conceptual models to guide the analysis of data to respond all research questions. Such conceptual models (in chapters 4 and 5) also became an important contribution to the field of crop loss research.

CASCADE was dedicated to:

- CASCADE was useful to determine, in conditions of small farmers, the relationships of a range of production situations and management strategies with individual injury levels of pests and diseases, injury profiles, coffee yields and yield losses. Results were useful to discuss and respond Q1.1, Q1.2.and Q2.2.

- Data on cropping practices (labor, inputs, costs), overall agroforestry production (coffee, bananas, other fruits, timber, and their prices) and biophysical measurements were used to assess several ecosystem services as overall benefits (provisioning of products for families, carbon sequestration, maintenance of soil fertility, regulation of pests and diseases). Then, analysis of effects of management strategies and production situations on ecosystem services were performed to respond **Q3.1.**
- Based on yield losses and the local prices of coffee, it was possible to estimate the economic yield losses in conditions of small farmers, in order to respond **Q3.2.**
- Using the data of yield and economic losses, and on ecosystem services, the most promising coffee agroecosystems were identified and described, in order to respond **Q3.3.**

2.2 Coffee Losses Experiment (CoLosses)

2.2.1 Location and establishment

A coffee experimental parcel at full sun exposure (960 m^2 in total) was established in a flat terrain in the farm of the Tropical Agricultural Research and Higher Education Center (CATIE), Turrialba, Costa Rica (Lechevallier, 2013). The parcel was planted in 2010, and the experiment began in year 2013 and lasted until 2015. The parcel was located in the coordinates 9°53’11.24’’N and 83°40’07.94’’O, at 648 meters above sea level (m.a.s.l.). Turrialba is a rainy area all year long, with slightly dry periods in March and April, and highly rainy periods in June and July. In the last ten years, the mean total annual rainfall was 2781 mm and the mean annual temperature was 22.2°C. In the years on the experiment, there were more noticeable differences in the amounts of rainfall (especially in 2015), while temperatures and relative humidity were similar (Table 2.1).

Table 2.1. Climate characteristics in Turrialba, Costa Rica (2011-2015)

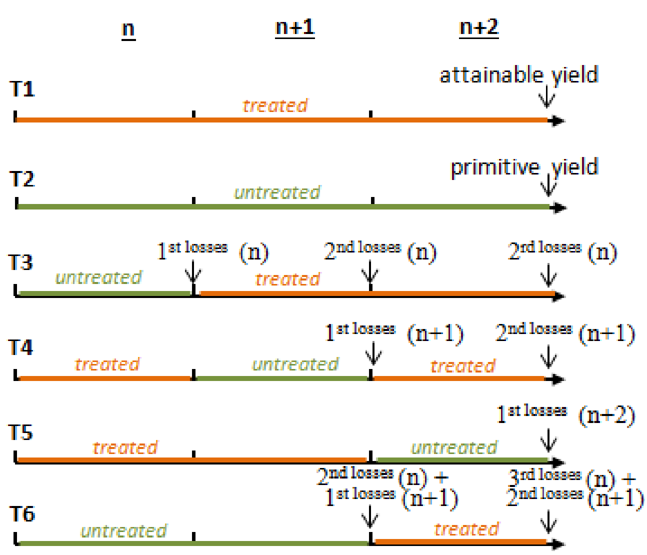
Year	Total Rainfall (mm)	Monthly Rainfall (mm) Mean ± SD	Relative Humidity (%) Mean ± SD	Monthly temperature (°C) Mean ± SD	Monthly max temperature (°C) Mean ± SD	Monthly min temperature (°C) Mean ± SD
2011	2740	223.3 ± 126.8	93.0 ± 2.0	22.0 ± 1.0	27.2 ± 1.2	18.5 ± 1.0
2012	2639	219.9 ± 173.9	93.7 ± 1.4	21.9 ± 1.0	27.1 ± 1.3	18.3 ± 1.2
2013	1945	162.1 ± 93.9	93.7 ± 1.3	22.3 ± 0.8	27.5 ± 0.8	18.7 ± 1.2
2014	2659	221.6 ± 139.7	88.5 ± 3.2	23.0 ± 1.0	27.7 ± 0.8	20.0 ± 1.6
2015	3248	270.7 ± 155.4	91.2 ± 3.2	22.8 ± 0.4	27.5 ± 0.8	19.7 ± 0.5

SD: standard deviation; max: maximum; min: minimum. Source: Weather station of the Tropical Agricultural and Higher Education Center (CATIE).

2.2.2 Experimental design

All coffee plants (*Coffea arabica* L.) in the experimental parcel were of the dwarf variety *Caturra*, with a density of 5000 plants per hectare (2m between coffee rows and 1m between plants). The first two years of coffee plants growth, the entire parcel received the same adequate management: three fertilizations, three applications of chemical fungicides and three applications of herbicides yearly. During the experiment, only the fungicide applications varied, the fertilizations and herbicide applications were always the same, and one pruning per year was done (leaving between two and four productive stems each year).

The experiment had six treatments, each one consisting in the application (treated) or not application (untreated) of chemical pesticides yearly, during three years (Fig. 2.1). The chemical pesticides, the dosages and times of applications each year are described in Table 2.2. The experimental design was composed by 24 plots in total (40 m^2 each plot): six randomized treatments and each one with four replicates.



Expected information	$Y_{(n)}$= observed yield on year *n*; $I_{(n)}$= observed injuries on year *n*
Attainable yield	
$Y_{(n)}=f(I_n)$; $Y_{(n+1)}=f(I_n+I_{n+1})$; $Y_{(n+2)}=f(I_n+I_{n+1}+I_{n+2})$	
$Y_{(n)}=f(I_n)$: → primary losses *(n)*; $Y_{(n+1)}=f(I_n)$:→ secondary losses *(n)*; $Y_{(n+2)}=f(I_n)$:→ secondary losses *(n)*	
$Y_{(n+1)}=f(I_{n+1})$: → primary losses *(n+1)*; $Y_{(n+2)}=f(I_{n+1})$:→ secondary losses *(n)*	
$Y_{(n+2)}=f(I_{n+2})$:→ primary losses *(n+2)*	
$Y_{(n+1)}=f(I_n+I_{n+1})$; $Y_{(n+2)}=f(I_n+I_{n+1})$	

Treatments consisted in pesticide applications in order to avoid injuries caused by pests and diseases. $Y_{(n)}$ is the observed yield in year n, and $I_{(n)}$ is the observed injuries in year n

Fig. 2.1. Treatments (T1 to T6) for the primary and secondary yield losses assessment

Table 2.2. Characteristics of the chemical pesticides, dosages and applications during the experiment CoLosses

Characteristics of the chemical fungicides and dosages utilized				
	Symbol	**Active components**	**Action mode**	**Dosage utilized in the experiment**
Fungicide				
SOPRANO C25SC	SO	Triazole, Epoxiconazole, Carbendazim, Benzimidazole	Systemic; protective, curative and eradicant; inhibits sterol biosynthesis	1.25cc/liter of water
HACHERO 6,6SL	HA	Copper sulfate pentahydrate	Systemic; preventive and curative; causes protein denaturation	5.0g/liter of water
AMISTAR 50WG	AM	Azoxystrobim, Difeconazole	Systemic; preventive; prevent respiration	2.0cc/liter of water
OPERA 18.3 SE	OP	Pyraclostrobij, Epoxiconazole	Systemic; protective, curative and eradicant	2.0cc/liter of water
Insecticides				
Sumithion 50EC®	SU	Fenithrotion	Contact	4.0cc/liter of water
Solver 48EC®	SOL	Chlorpyrifos	Contact	2.5.0cc/liter of water

Applications during the experiment only in plots assigned to be treated each year												
Year	**Ene**	**Feb**	**Mar**	**Apr**	**May**	**Jun**	**Jul**	**Aug**	**Sep**	**Oct**	**Nov**	**Dic**
2013							SO	SO	HA	AM		
2014					OP		OP		SO	OP		
2015			OP		OP		OP		SO	OP		

2.2.3 Measurement of the studied variables

Each plot had 30 plants (five rows and six plants per row), the external rows and plants were defined as borders. Therefore, useful plots have 12 plants in the three central rows, where six plants were marked for measurements (Fig. 2.2). On each marked plant, three branches were marked for measurements of pests and diseases: one in the low stage, one in the middle stage and one in the upper stage (Fig. 2.3); these branches were changed every year. Besides, Tinytag PLUS2 devices (to measure microclimate: temperature and humidity) and sensors from Campbell Scientific stations (to measure leaf temperatures and leaf wetness) were installed in 12 coffee plots, two plots per treatment (Fig. 2.2).

Measurements were done during three years (2013-2015): healthy and infected/infested leaves with pests and diseases, differentiating young and old leaves, and severity were measured in marked branches monthly; dead branches were counted at the end of the harvest period each year; yield components (productive branches, fruiting nodes, fruits per node) were counted once, and coffee cherries were harvested in the entire marked plants each 15 days; samples of leaves in the middle stage of several plants in each plot were taken for analysis of nutrient contents; soil sub-samples were taken at 50cm from the trunk of marked plants, and a composite sample per plot for chemical fertility analysis; temperature and relative humidity of microclimate, and leaf temperatures and leaf wetness were measured in four key periods each year. In Chapter 4 a Table is also provided with more details of the list of the variables, the methods of measurements, calculation of additional variables when needed, and frequencies of measurements.

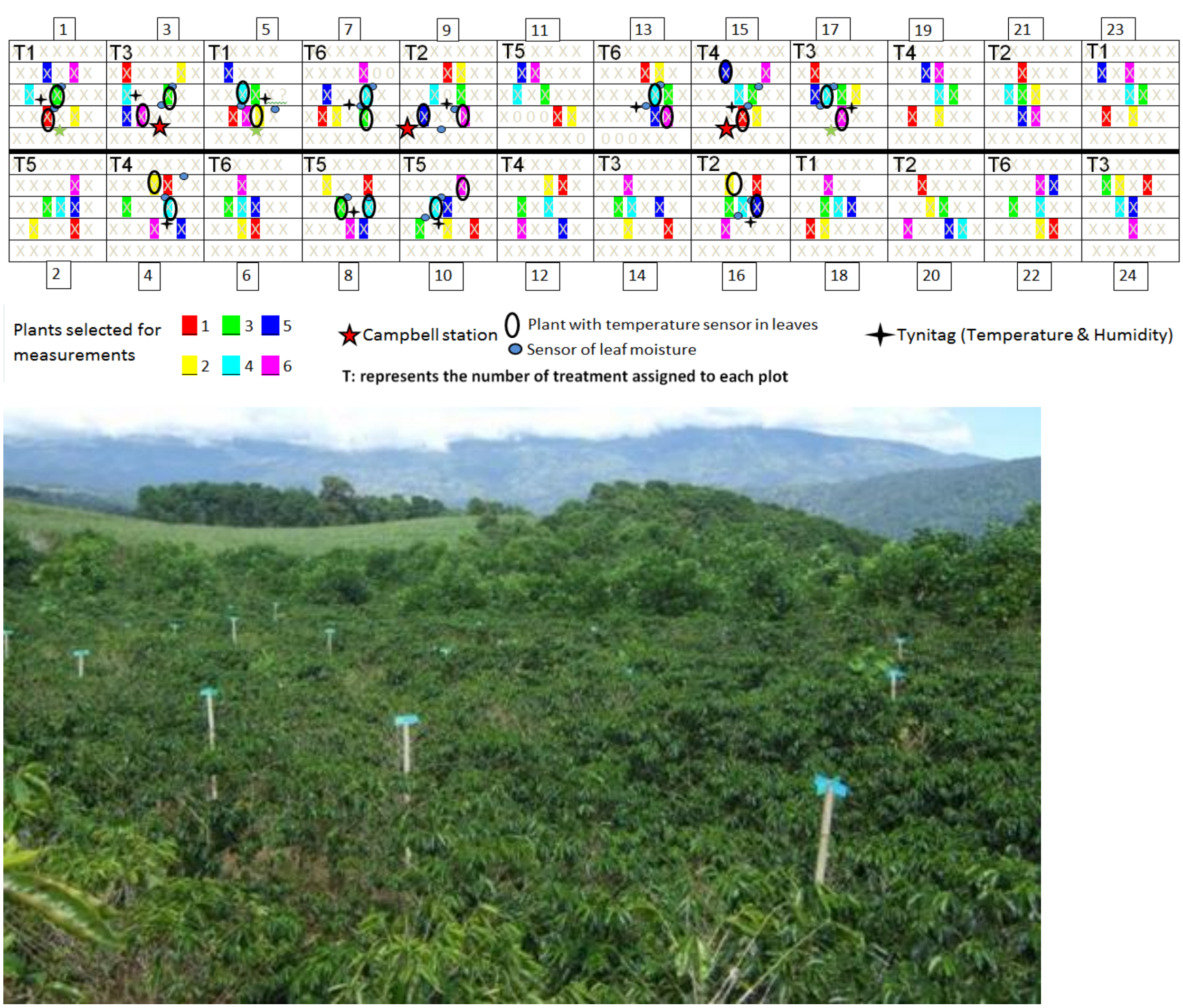


Fig. 2.2. Sketch and picture of the Coffee Losses experiment in CATIE farm, Turrialba, Costa Rica

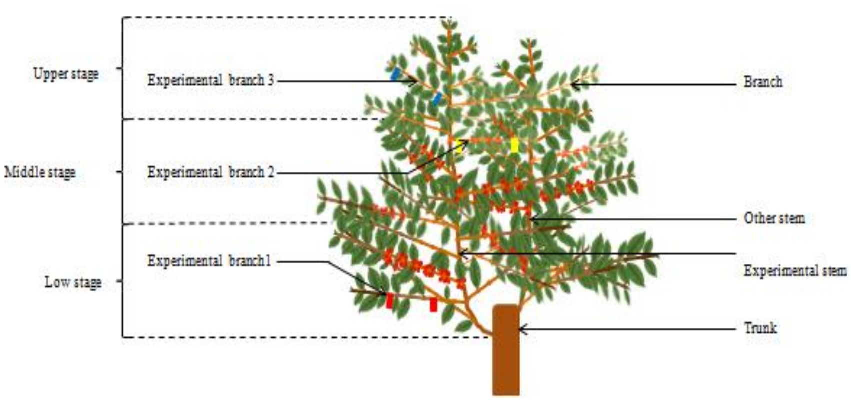
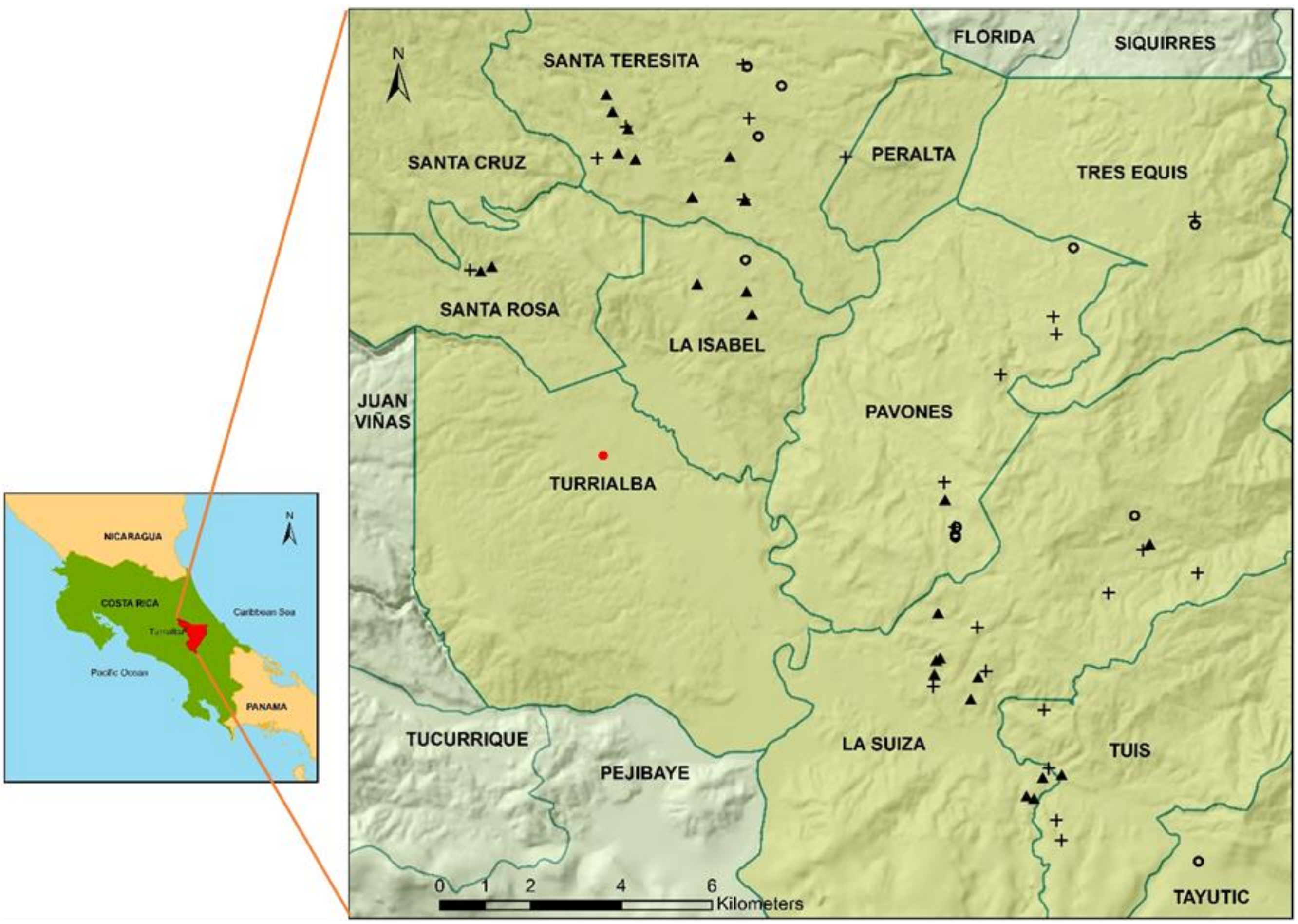


Fig. 2.3. Illustration of marked branches in coffee plants for measurements

2.3 Coffee Research Plot Network (CASCADE)

2.3.1 Location and establishment

A coffee research plot network (69 plots) for two years of research (2014-2015) was established in coffee growing communities of the canton Turrialba, Costa Rica. All of the selected coffee plots were in farms of smallholder coffee farmers. The 69 coffee plots were distributed in 27 communities of eight districts of Turrialba, covering 300 km^2 (=30.000 ha) approximately (Fig. 2.4). Plots were chosen from a database build by the project CASCADE, where 150 coffee farms were described regarding crop management, shade type and topography. The general weather conditions in Turrialba were already described in section 2.1.1 and Table 2.1. However, rain gauges were also installed in several communities to have a better estimation and representation of the rainfall in different areas of the network; these data were used especially in Chapter 5, where climate was an important production situation to take into account.



O CFS: Coffee in full sun + CLD: Coffee agroforestry systems with low diversification ▲ Coffee agroforestry systems with high diversification

The coverage area of the Coffee Research Plot Network were among: 9°56'16.63''N 83°43'24.09''O; 10°02'27.93''N 83°39'40.60''O; 9°48'33.85''N 83°34'12.08''O; 9°54'11.25''N 83°29'47.31''O

Fig. 2.4. Location of coffee plots in the Coffee Research Plot Network (CASCADE), Turrialba, Costa Rica

2.3.2 Objective and strategy for the selection of coffee plots

The objective for the establishment of CASCADE was to involve a wide range of production situations and management strategies in order to respond the major research question of this Ph.D. research. That is why the strategy was to select coffee plots in different conditions of: topo-climate (different topography, soil and altitude), type of shade (diversity-structure-shade cover) and management intensities of cropping systems. The intention was to select coffee plots according to the following criteria:

- Topo-climate:
  - Half of the coffee plots under 850 m.a.s.l.
  - Half of the coffee plots above 850 m.a.s.l.
- Type of shade: a quarter of coffee plots under each of these four types of shade canopy:
  - Type 1: Full sun coffee
  - Type 2: Coffee + service trees (mainly Poró -*Erithrina poeppigiana*-)
  - Type 3: Coffee + service trees + bananas
  - Type 4: Coffee + service trees + productive woody perennials (fruit and timber trees)
- Management intensity:
  - Half of coffee plots with high management intensity (more than one application of fertilizers, herbicides and pesticides, plus one pruning and one or more weedings the last year)
  - Half of coffee plots with low management intensity (lack of application of one or more of these inputs: fertilizers, herbicides or pesticides, plus and few weedings –less than two- the last year)

That strategy meant 16 combinations (2x4x2 criteria), and with four coffee plots replication per each one, it was expected 64 coffee plots. But, it was not possible to find five coffee plots for all combinations due to some of them are rare in Turrialba, for instance, coffee plots in low lands, in full sun and with low management are very scarce. That is why, there were 59 coffee plots selected, one per farm; but in 10 of that farms, an additional coffee plot was installed in order to generate data for the models to estimate yield losses (explained in the section 2.3.3). Thus, 10 more plots were added and CASCADE was composed by 69 coffee plots in total.

Furthermore, coffee plots initially were classified by observation in a given type of shade; but after doing a corroboration with a simple cluster hierarchical multivariate analysis with variables of number of species and densities per species (which were measured in a subplot, see the following sections), turned out that coffee plots classified by observation in types 3 and 4 were classified in the same group, and therefore it was decided to work only with three main types of shade.

It is important to state that the intention of this strategy was not to have strictly the same number of coffee plots for each combination of the criteria, but to generate a variability of conditions. Besides, it is also important to remember that each plot is unique by nature, which means that even within each combination of criteria the coffee plots can also vary. As a consequence, this strategy of selection of coffee plots was useful and gave a good range of production situations and management strategies.

Thus, CASCADE covered an important range of production situations and management strategies, involving: coffee plots in different altitudes (as representative of environmental conditions); with different botanical composition and structure of shade canopies represented by three types of shade (Fig. 2.5); with different cropping practices for the agronomic and agroforestry management, in terms of type and frequency of labors and quantity of inputs; and also with different characteristics of soil, given that coffee plots were located in different communities and altitudes, and influenced by particular fertilizations in each plot. All of these quite different conditions also permitted the observation of a wide range of injury levels of pest and diseases, and coffee yields. Only the type of shade was maintained as a quantitative variable, and all the other variables were used as quantitative variables in order to construct indexes and/or typologies representing production situations, as it can be seen in chapters 3, 5 and 6.

A common characteristic maintained for all coffee plots was to have coffee plants (*Coffea arabica* L.) of the dwarf variety *Caturra* as the unique or dominant variety. *Caturra* is the most common variety in Costa Rica, and is widely cultivated in countries of Central and South America (McCook and Vandermeer, 2015). This was an important criterion in order to ensure that pests and diseases which were evaluated, were attacking the same variety, avoiding that way possible biases if we had different varieties.

2.3.3 Experimental design

In each coffee plot, an experimental plot composed of 12 coffee rows with 19 plants each, was demarcated in a representative place of the plot, with the following characteristics (Fig. 2.6):

- The area of each experimental plot depended on the distances between rows and between plants. It was estimated that the sizes were between 221 m^2 and 595 m^2
- On each of the four sides of the experimental plot, the edges of two plants were considered as borders. Therefore, a useful experimental subplot for measurements was composed by eight rows and 15 plants per row in the center of the experimental plot
- For the measurement of characteristics of the shade canopy (e.g. species richness and abundances, shade cover), which could have influence on the plants (yield components, pests and diseases) in the experimental subplot, a circular area of 1000 m^2 was demarcated from the center of the experimental subplot (17.8 m radius). This was useful for analysis in chapters 3 and 5

For estimating coffee yield losses, we choose ten coffee farms where two experimental plots were established (20 plots in total). These coffee farms were located in altitudes above 850 m.a.s.l. with low management intensity. In each farm, in one of the experimental plots, chemical fungicides were applied several times during two years (2014-2015) to control coffee diseases (Table 2.3), and in the other one there were no applications; other than that, the management of both experimental plots (labors, fertilizers, weedings, etc.) were the same, applied by the coffee farmer. Coffee yield losses was intended to be estimated by modeling using data of coffee yields, yield components, and yields reducing factors (pests and diseases, dead branches) of these ten plots.

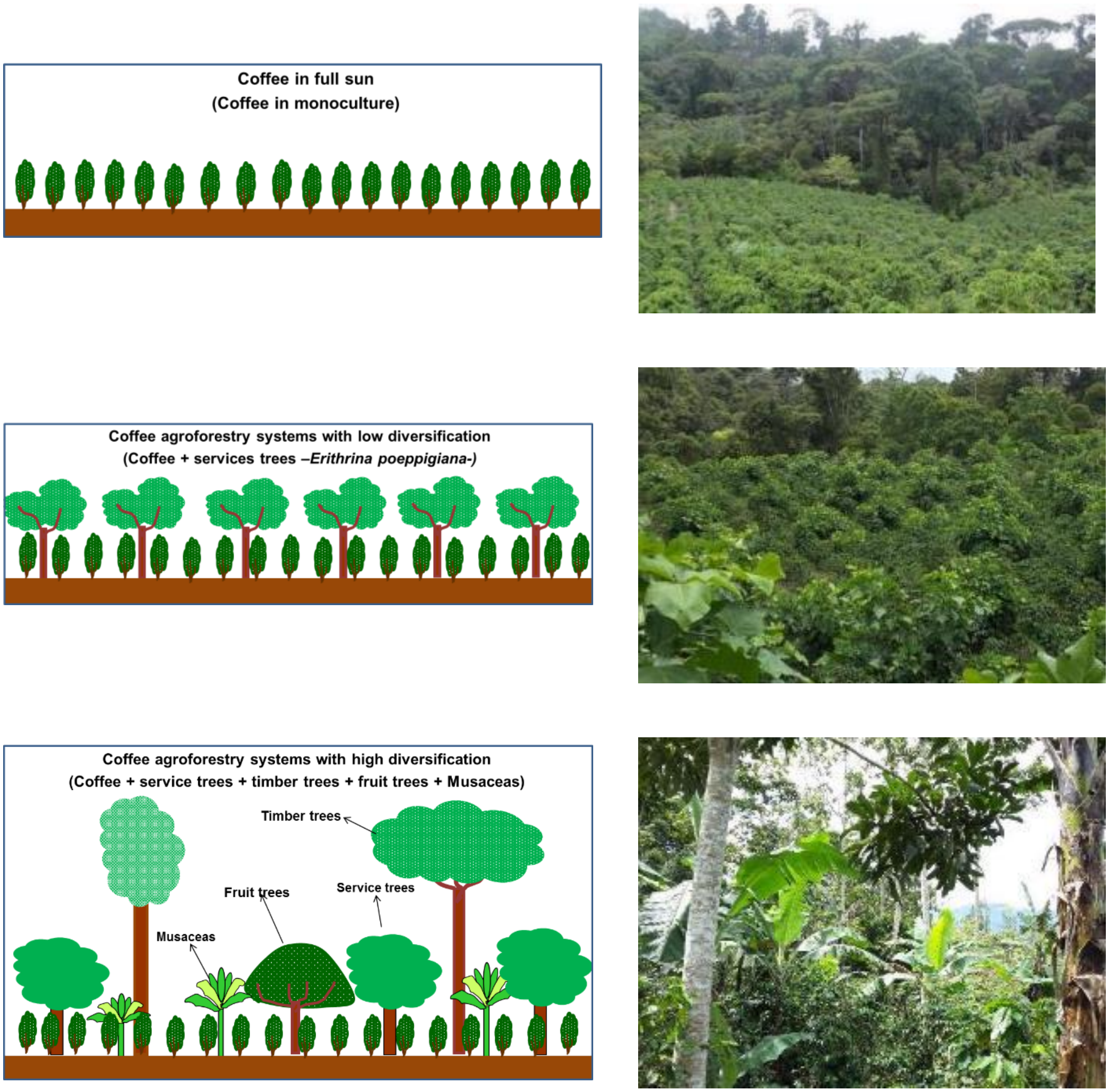


Fig. 2.5. The final main three types of shade considered in the Coffee Research Plots Network, Turrialba, Costa Rica

Table 2.3. Characteristics of the chemical fungicides, dosages and applications during the experiment in ten coffee plots of CASCADE

Characteristics of the chemical fungicides and dosages utilized				
Fungicide	**Symbol**	**Active components**	**Action mode**	**Dosage utilized in the experiment**
SOPRANO C25SC	SO	Triazole, Epoxiconazole, Carbendazim, Benzimidazole	Systemic; protective, curative and eradicant; inhibits sterol biosynthesis	1.25cc/liter of water
OPERA 18.3 SE	OP	Pyraclostrobij, Epoxiconazole	Systemic; protective, curative and eradicant	2.0cc/liter of water

Applications during the experiment only in ten experimental plots assigned to be treated each year												
Year	**Ene**	**Feb**	**Mar**	**Apr**	**May**	**Jun**	**Jul**	**Aug**	**Sep**	**Oct**	**Nov**	**Dic**
2014					OP		OP		SO		OP	
2015			OP		OP		OP		SO		OP	

2.3.4 Measurements of the studied variables

Each experimental subplot had 120 useful plants in total (eight rows and 15 plants per row), where eight coffee plants were marked for measurements, one plant per row (Fig. 2.6). On each marked plant, three branches were marked for measurements of pests and diseases: one in the low stage, one in the middle stage and one in the upper stage (the same as in CoLosses, Fig. 2.3); these branches were changed every year. Besides, iButton devices (DS1921G and DS1923) to measure microclimate (temperature and humidity) were installed in two of the marked plants (Fig. 2.6).

Measurements were done during two years following the same protocol than in CoLosses (section 2.2.3) for variables related to pests and diseases, dead branches and yield components, samples of leaves for analysis of nutrient contents and samples of soil for chemical fertility analysis. Besides, given that CASCADE represents different production situations and management strategies, many other variables were measured: topographic characteristics of coffee plots (slope, orientation of slope, altitude); distances between coffee rows and between coffee plants; growth characteristics of coffee plants (trunk diameter, plant height); dieback scales and symptoms of nutrient deficiencies of coffee plants in the entire useful plot; soil cover; characteristics of shade canopies (species richness, abundances, trunk diameters, shade cover); temperature and relative humidity of microclimate from October 2014 to December 2015.

Furthermore, all the information on management labors and costs (cropping practices and inputs) and prices of agroforestry products (coffee, fruits, timber) were collected through interviews with the coffee farmers for each year. In Chapters 3, 5 and 6, several Tables are also provided with more details of the list of the variables, the methods of measurements, calculation of additional variables when needed, and frequencies of measurements.

Ripe coffee cherries were harvested in marked plants of the 20 plots (160 plants) located in the same ten farms dedicated to the yield estimation. Yield in the other coffee plot had to be estimate using statistical models built from these observed data.

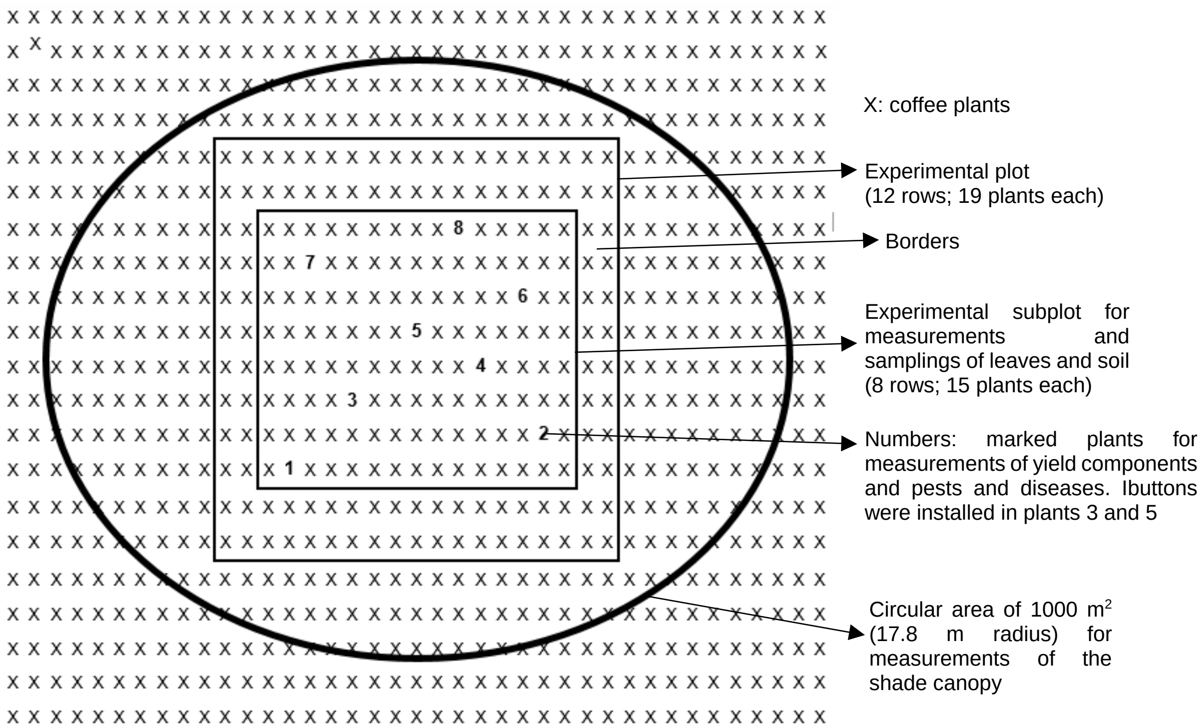


Fig. 2.6. Sketch of the coffee plots in the coffee research plots network

2.4 Statistical methods

Six main types of statistical analysis were performed in order to respond the research questions. These statistical analyses were defined along with the advisors of the Thesis, as well as with the help of Master students who participated in the field work and tested different statistical techniques (Krolczyk, 2014; Clement, 2015; Mathiot, 2015). In the following paragraphs, a general description of the statistical analysis is presented:

1) **Linear models to assess effects of production situations and management strategies:** Linear models were used to estimate response variables (yields, pests and diseases, ecosystem services) in function of variables of production situations and management strategies (individual effects and interactions). Variables with significant effects in the estimations were those considered to have effects on response variables. This technique was useful to test effects of altitude, shade and management on the provision of ecosystem services (chapter 3).

2) **Linear regressions:** Linear regressions were performed between indicators of four ecosystem services (regulation of pests and diseases, provisioning of agroforestry products, maintenance of soil fertility, and carbon sequestration), and between indicators of ecosystems services and indicators of plant biodiversity. These analyses were useful to assess possible trade-offs or synergies between such indicators, and then identify the most promising coffee agroforestry systems (chapters 3 and 6).

3) **Linear mixed models (randomized) to assess effects of treatments on yields components and pests and diseases variables in CoLosses:** This analysis was applied to the data of CoLosses, in order to assess significant differences especially of coffee yields among treatments. The models had the treatments as fixed factors, and some variables such as soil acidity and year as random effects. Differences of means among treatments were analyzed with the Fisher's LSD test ($p<0.05$). These analyses were necessary because only finding differences of coffee yields among treatments, it was possible to calculate primary and/or secondary yield losses by experimentation (chapter 4).

4) **Structural equation modeling:** we applied piecewise structural equation modeling (PiecewiseSEM), which is a confirmatory path analysis that works with two or more linear models in order to test direct and indirect effects of given variables for the estimation of a final response variable—in our case, yields. This modeling approach was used to identify the most important predictors of coffee yields in CoLosses (chapter 4), which then were used in linear mixed models to estimate attainable yields, actual yields, and primary and secondary yield losses in CASCADE (chapter 5).

5) **Linear mixed models (randomized) for estimations of coffee yields and yield losses:** these estimations were especially useful for CASCADE. Linear mixed models were run with the predictors identified by PiecewiseSEM, using plots as random effects. To check the models, graphs of linear regressions with the estimated yield as a function of real harvested coffee yield were traced, displaying the respective R^2. In addition, indicators of the predictive quality of the models were calculated: mean of residues (Bias), mean absolute error (MAE), root-mean-squared error (RMSE), relative root-mean-squared error (RRMSE), modeling efficiency (EF), index of agreement (IA) (chapter 5).

6) **Multivariate analysis to assess the associations between typologies of injury profiles and production situations, management strategies, and categories of yield and yield losses**

This was the most challenging analysis given that we had to test several sets of multivariate techniques (methods), trying to identify the most suitable method that best reflected the associations. In Fig. 2.7, we show, just to illustrate, the test of five methods, from which, we finally selected the methods 5 (which was used in chapter 5). This method is described below:

- **Construction of injury profiles**: i) introduce variables of pest and diseases in a principal component analysis (PCA); ii) consider only the components which represent more than 85% of the cumulative variability for the clustering; iii) hierarchical clustering with that components, using Euclidian distance between plots and a Ward method for the clustering.
- **Construction of typologies:** i) perform the Partial Least Square Discriminant Analysis (PLS-DA) which put together injury profiles and variables of a given type of variables, in order to obtain several components; ii) consider only two or three components resulting from the analysis for the clustering; iii) hierarchical clustering with that components (Euclidian distance; Ward method). With this

methodology, we constructed several typologies to represent production situations, management strategies and physiological characteristics of coffee plants.

- **Construction of categories of yields:** we did a frequency distribution of each yield (attainable, actual, loss) and divide it in three groups composed by the same number of coffee plots. That way, we constructed three categories for: attainable yields, actual yields, yield losses (primary and secondary).
- **Description of typologies:** we described the profiles (groups) of each typology based on the means of the variables used for their clustering. Significant differences of each variable among profiles were assessed with the Kruskal-Wallis non-parametric test ($p<0.05$).
- **Test significant associations between injury profiles and each typology:** we used the exact test of Fisher; which permits to test the null hypothesis that injury profiles and a given typology in a contingency table are independent.
- **Association among injury profiles, yields and all the typologies:** analysis of correspondences in which attainable yields, actual yields and yield losses were illustrative variables, and injury profiles and all the other typologies were active variables. This analysis permitted to obtain a graphic in which was possible to deduce the clustering of several types, thus representing several different situations and their association with given levels of injury profiles and yields.

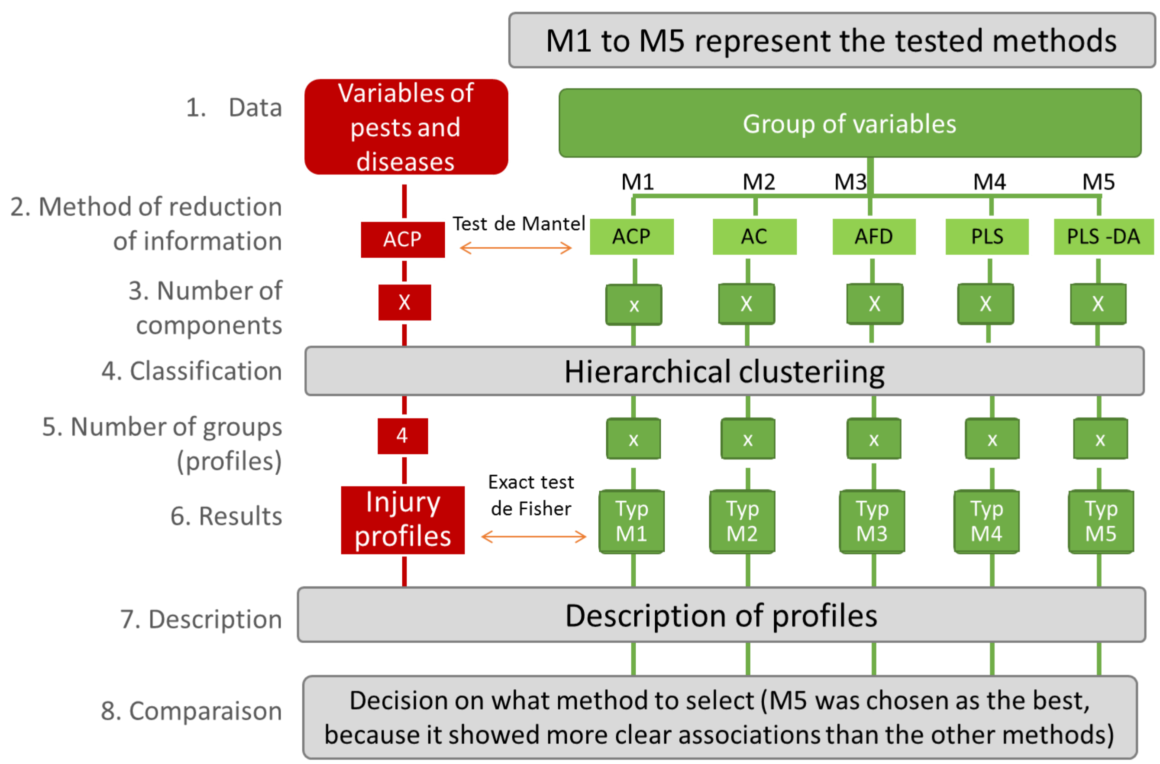


Fig. 2.7. Simple illustration of five methods (multivariate techniques) tested for the assessment of associations between typologies of injury profiles and production situations, management strategies, and categories of yield and yield losses.

PART III. RESULTS

Chapter 3. Multiple ecosystem services provided by coffee agroecosystems

Manuscript 1: Effects of shade, altitude and management on multiple ecosystem services in coffee agroecosystems

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Effects of shade, altitude and management on multiple ecosystem services in coffee agroecosystems

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Abstract

Agroforestry systems provide diverse ecosystem services that contribute to farmer livelihoods and the conservation of natural resources. Despite these known benefits, there is still limited understanding on how shade trees affect the provision of multiple ecosystem services at the same time and the potential trade-offs or synergies among them. To fill this knowledge gap, we quantified four major ecosystem services (regulation of pests and diseases; provisioning of agroforestry products; maintenance of soil fertility; and carbon sequestration) in 69 coffee agroecosystems belonging to smallholder farmers under a range of altitudes (as representative of environmental conditions) and management conditions, in the region of Turrialba, Costa Rica. We first analyzed the individual effects of altitude, types of shade and management intensity and their interactions on the provision of ecosystem services. In order to identify potential trade-offs and synergies, we then analyzed bivariate relationships between different ecosystem services, and between individual ecosystem services and plant biodiversity. We also explored which types of shade provided better levels of ecosystem services. The effectiveness of different types of shade in providing ecosystem services depended on their interactions with altitude and coffee management, with different ecosystem services responding differently to these factors. No trade-offs were found among the different ecosystem services studied or between ecosystem services and biodiversity, suggesting that it is possible to increase the provision of multiple ecosystem services at the same time. Overall, both low and highly diversified coffee agroforestry systems had better ability to provide ecosystem services than coffee monocultures in full sun. Based on our findings, we suggest that coffee agroforestry systems should be designed with diversified, productive shade canopies and managed with a medium intensity of cropping practices, with the aim of ensuring the continued provision of multiple ecosystem services.

Key words: Agroforestry; Carbon sequestration; Coffee yields; Disservices; Incomes; Pests and diseases; Soil fertility; Trade-offs

1. Introduction

Agroforestry systems in tropical landscapes provide a series of ecosystem services that help sustain crop production, improve farmers´ livelihoods and conserve biodiversity (Jose, 2009; Tscharntke *et al.*, 2011). Shade trees and other companion plants in agroforestry systems can produce fruits (Rice, 2011; Cerda *et al.*, 2014), timber, firewood and other products for sale or household use (Somarriba *et al.*, 2014), thereby diversifying the sources of income for farmers and contributing to food security. The roots and leaf litter of shade trees, especially leguminous trees, improve nutrient recycling and soil quality (Beer *et al.*, 1998a) and can help reduce soil erosion (Gómez-Delgado *et al.*, 2011b). Shade trees are also useful for protecting crops from strong winds, high temperatures and extended dry periods (Schroth *et al.*, 2009; Jha *et al.*, 2014). Shade trees and other woody perennials contribute to the conservation of animal and plant biodiversity, and sequester carbon from the atmosphere, thereby contributing to climate mitigation (Jha *et al.*, 2011; Somarriba *et al.*, 2013; Deheuvels *et al.*, 2014).

Yet agroforestry systems can also result in disservices and antagonistic effects (Zhang *et al.*, 2007; Power, 2010). A known drawback of agroforestry systems is that the yields of the main crop are often lower than those in full sun systems (López-Bravo *et al.*, 2012), at least in the short term. With increasing shade cover, the relative yield of the main crop tends to decrease (Zuidema *et al.*, 2005) due to greater competition for light, water and nutrients in soil between trees and the main crop. Another potential drawback of agroforestry systems is the higher labor requirements to manage trees and other plants. Agroforestry systems can favor or disfavor the attack of pathogens and insects depending on the composition, structure and management of the shade canopies (Staver *et al.*, 2001; Avelino *et al.*, 2006; Cheatham *et al.*, 2009; Pumariño *et al.*, 2015).

Despite the recognition that agroforestry systems can potentially provide diverse ecosystem services, there is still limited understanding on how shade trees affect the provision of multiple ecosystem services and about the potential trade-offs or synergies among them. Most studies in agroforestry systems have focused on a single ecosystem service (Jose, 2009), and have not examined relationships among various ecosystem services. In addition, most studies have only considered the individual effect of shade on ecosystem services, underestimating other factors, such as management practices and environmental conditions, which may interact with shade to provide ecosystem services (Staver *et al.*, 2001; Avelino *et al.*, 2006). However, a good understanding of different factors, including their interactions, affecting the provision of ecosystem services, and the analysis of relationships (trade-offs or synergies) among ecosystem services, are needed to design high performing agroforestry systems (Rapidel *et al.*, 2015).

Understanding the provision of ecosystem services by agroforestry systems is particularly important for the coffee sector in Central America which is currently under severe stress. A chain of events, including decreasing coffee prices, increasing production costs, and an outbreak of coffee leaf rust (*Hemileia vastarix* Berkeley and Broome) since 2012, has significantly reduced coffee production. Following the coffee rust outbreak, farmers were forced to stump their impacted coffee plantations to rejuvenate coffee trees or to renew them with new coffee varieties, or even to replace them with new crops (Baker, 2014; Avelino *et al.*, 2015; McCook and

Vandermeer, 2015). For instance, 50% of coffee areas have disappeared in the Volcan Central Talamanca Biological Corridor in Costa Rica between 2000 and 2009 (Bosselmann, 2012), and 35% of the coffee areas in southern Guatemala between 2000 and 2004 (Haggar *et al.*, 2013). The conversion of coffee plantations to other land uses results in the loss of shade trees and other vegetation and negatively affects plant biodiversity (Zhang *et al.*, 2007; De Beenhouwer *et al.*, 2013). Information on the potential benefits provided by shade trees associated with coffee plantations could encourage decision makers, technicians, and farmers to maintain and/or increase land uses under coffee agroforestry systems, and stem the ongoing loss of these systems (Cheatham *et al.*, 2009; Jose, 2009).

The objectives of this study were i) to assess the effectiveness of different types of shade of coffee agroecosystems in providing multiple ecosystem services under different environmental and management gradients, and ii) to understand the relationships (trade-offs or synergies) across different ecosystem services and plant biodiversity. We quantified indicators of four major ecosystem services: 1) regulation of pests and diseases; 2) provisioning of agroforestry products (coffee, bananas, other fruits, timber); 3) maintenance of soil fertility; and 4) carbon sequestration, in coffee agroecosystems belonging to smallholder farmers under a range of altitudes (as representative of environmental conditions), management practices and types of shade. We hypothesized that the effectiveness of different types of shade in providing ecosystem services depends on their interaction with coffee management and altitude where coffee is grown, and that trade-offs or synergies could occur among certain ecosystem services. Based on our findings, we highlighted key aspects that should be considered for the design and management of coffee agroecosystems to ensure the continued provision of multiple ecosystem services.

2. Materials and methods

2.1. Coffee plot network and experimental design

A coffee plot network (69 plots) was established in the canton of Turrialba, Costa Rica. Turrialba is located in a premontane wet forest life zone, with an mean annual rainfall of 2781 mm and a mean annual temperature of 22.2°C (averages of the last 10 years), with small variations among months. In this area, coffee is grown from 600 to 1400 m.a.s.l. (meters above sea level). Farms in higher elevations experience slightly wetter and cooler temperatures compared to farms at lower elevations.

The plot sampling strategy had the objective to select coffee plots of different types of shade across altitudinal and management intensity gradients. Plots were selected with contrasting characteristics in the botanical composition and structure of shade canopies (in terms of species richness, abundances and trunk basal areas), with contrasting coffee cropping practices (in terms of different types of practices and frequency of applications), and at different altitudes. However, in order to limit variations and avoid confounding effects of different factors (Clermont-Dauphin *et al.*, 2004), we chose coffee plots that shared three main characteristics: i) they were owned by smallholder farmers, ii) had coffee plants (*Coffea arabica* L.) of the dwarf variety *Caturra* as the unique or dominant variety, which is the most common variety in Costa Rica and in other countries of Central and South America (McCook and Vandermeer, 2015), and iii) were located on soils belonging to the order

Inceptisols, suborder Udepts. These soils in Turrialba are considered to have moderate fertility but with problems of acidity (CIA, 2016).

In each coffee plot of the network, an experimental subplot composed of eight coffee rows with 15 plants each was demarcated in a representative place of the plot. Eight coffee plants (and three branches per plant) were marked, one plant per row, inside the experimental subplot. These eight plants were used for measurements of pests and diseases and coffee yields, and for sampling soil subsamples near them. For the measurement of characteristics of the shade canopy, a circular area of 1000 m^2 was established in the center of the experimental subplot (17.8 m radius).

2.2. Measurements and calculations of the factors studied

2.2.1. Altitude

The altitude of each coffee plot was measured with a GPS. The mean altitude ± standard deviation of all coffee plots was 877 ± 126 m.a.s.l., ranging from 646 to 1107 m.a.s.l.

2.2.2. Management intensity index

Data on the management were obtained through semi structured interviews with farmers. A management intensity index was calculated for each coffee plot. The calculations were based on existing indices of management intensity used in coffee studies (Mas and Dietch, 2003; Philpott *et al.*, 2006). In the present study, the calculations included 11 cropping practices commonly applied in Turrialba (Table 3.1). First, for each cropping practice, the number of times per year that this practice was applied was transformed to a value *IH* or *IL* between 0 to 1 reflecting the practice intensity, the higher the value, the higher the intensity:

$$IH = \frac{value\ - minimum}{maximum - minimum} \qquad IL = 1 - \frac{value - minimum}{maximun - minimum}$$

where IH is the transformed value for cropping practices for which a higher number of applications denotes a higher management intensity (e.g. number of weedings, application of fertilizers, fungicides, etc.) and IL is the transformed value for cropping practices for which a lower value denotes a higher management intensity (distances between coffee rows and between coffee plants); value was the annual number of applications of a given cropping practice for a given plot; and minimum and maximum were the minimum and maximum values registered for that cropping practice in the data set, respectively. Then, the transformed values obtained for all cropping practices were summed to obtain the management intensity index of each coffee plot (maximum possible =11, since we had 11 cropping practices); the higher the index, the higher the management intensity.

Table 3.1. Management descriptors of coffee agroecosystems in the coffee plot network (n= 69 plots) in Turrialba, Costa Rica

Cropping practices	Mean	SD	Minimum	Maximum	Median
Distance between coffee rows (cm)	173.0	23.5	100.2	233.5	174.3
Distance between coffee plants (cm)	119.7	17.7	76.6	168.8	118.9
Pruning of coffee plants (number yr^{-1})	0.8	0.4	0	1	1
Machete weedings (number yr^{-1})	1.3	1.3	0	5	1
Shovel weedings† (number yr^{-1})	0.4	0.7	0	3	0
Applications of fertilizers (number yr^{-1})	1.4	0.9	0	3	1
Applications of fungicides (number yr^{-1})	2.1	1.8	0	6	2
Applications of herbicides (number yr^{-1})	1.8	1.3	0	5	2
Harvest rounds of coffee (number yr^{-1})	10.3	2.4	5	14	11
Diversity of practices	5.2	1.1	3	7	5
Overall (number of practices yr^{-1})	18.1	4.3	8	26	18
Management intensity index††	5.0	1.3	2.1	7.7	5.1

SD: standard deviation;Diversity of practices: number of different practices applied per year
† Shovel weedings are done with a shovel, scraping at the ground level trying to better eliminate the weeds.
†† See text in section 2.2.2 for calculation of management intensity index

2.2.3. Types of shade

In the circular area of 1000 m^2, the shade canopy was characterized by identifying and measuring all plants >2.5 m in height. Plants were classified as: *Musaceae* (bananas and plantains), service trees (i.e., nitrogen fixing trees), fruit trees or timber trees. The trunk diameters and *Musaceae* stems were measured at 1.3 m from ground (breast height); fruit tree diameters were measured at 0.30 m. For service trees, such as *Erythrina poeppigiana,* which are pollarded once or twice a year, the height of the main trunk was also measured. In addition, the trunk diameters of the eight marked coffee plants were measured at 0.15 m from the ground. Shade cover (%) was measured with a spherical densiometer in the four corners and in the center of the experimental subplot, and then averaged. Finally, basal areas of tree trunks and the Shannon diversity index were calculated.

These variables were analyzed by using cluster analysis (Ward method and Euclidean distance) which classified coffee plots in three types of shade: i) Coffee monocultures in full sun (CFS); ii) Coffee agroforestry systems with low diversification (CLD), where the shade canopy was dominated by *Erythrina poeppigiana*, a leguminous tree; and iii) Coffee agroforestry systems with high diversification (CHD), where the shade canopy included a mix of service trees, bananas, fruit trees, timber trees and other plants (Table 3.2). These systems were equally distributed in the range of altitude, and were not related to the age of the plantation, the planted area, nor the management intensity index; that is why the means and standard deviations of these data were similar across the types of shade (Table 3.2).

Table 3.2. Structure and plant diversity, age, planted area, management intensity and altitude of the three types of shade (CFS: Coffee monocultures in full sun; CLD: Coffee agroforestry systems with low diversification; CHD: Coffee agroforestry systems with high diversification) in the coffee plot network, Turrialba, Costa Rica

	Variable	CFS (n=13) Mean ± SD	CLD (n=27) Mean ± SD	CHD (n=29) Mean ± SD
Structure and plant diversity	Density of coffee plants (Ind ha^{-1})	4864 ± 612	4868 ± 978	5286 ± 1367
	Density of fruit trees (Ind ha^{-1})	N/A	12 ± 27	59 ± 80
	Density of timber trees (Ind ha^{-1})	N/A	18 ± 29	63 ± 69
	Density of *Musaceae* plants (Ind ha^{-1})	N/A	98 ± 123	401 ± 340
	Density of service trees (Ind ha^{-1})	N/A	265 ± 136	173 ± 136
	Total density of shade canopy (Ind ha^{-1})	N/A	392 ± 180	695 ± 367
	Basal area of coffee plants (m^2 ha^{-1})	47.9 ± 32.1	52.0 ± 25.5	43.9 ± 25.7
	Basal area of fruit trees (m^2 ha^{-1})	N/A	0.1 ± 0.3	0.9 ± 1.1
	Basal area of timber trees (m^2 ha^{-1})	N/A	0.5 ± 0.9	3.4 ± 3.2
	Basal area of *Musaceae* plants (m^2 ha^{-1})	N/A	1.6 ± 2.2	6.5 ± 5.9
	Basal area of service trees (m^2 ha^{-1})	N/A	8.2 ± 5.3	5.0 ± 4.0
	Total basal area of shade canopy (m^2 ha^{-1})	N/A	10.4 ± 5.8	15.7 ± 6.7
	Total species richness (number)	N/A	4 ± 2	7 ± 3
	Shannon diversity index	N/A	0.7 ± 0.4	1.2 ± 0.4
	Shade cover (%)	N/A	14 ± 9	29 ± 10
Other data	Age of the cropping system (years)	15 ± 9	20 ± 10	15 ± 9
	Planted area (ha)	1.5 ± 1.0	1.0 ± 1.0	1 ± 0.9
	Management intensity index	5.4 ± 1.0	4.9 ± 1.3	5.0 ± 1.3
	Altitude (m.a.s.l.)	873 ± 109	885 ± 129	872 ± 133

SD: standard deviation; Musaceae: includes mainly bananas but also plantains; Citrus: includes mainly oranges (Citrus sinensis) and mandarin lemons (Citrus aurantifolia); Other fruits: includes mainly avocados (Persea americana), cas (Psidium friedrichsthalium), arazá (Eugenia stipitata) and peach palm (Bactris gasipaes); Service trees: includes mainly poro trees (Erythrina poeppigiana); Timber trees: includes mainly Cordia alliodora and Cedrela odorata.

2.3. Measurements and calculations of ecosystem service indicators (response variables)

The choice of ecosystem service indicators must meet several conditions in order to ensure that they are relevant. It should take into account the following criteria: i) be easily measured; ii) be sensitive to changes in the system; iii) respond to change in a predictable manner; iv) be anticipatory (be an early warning indicator); v) predict changes that can be averted by management actions; vi) be integrative (able to reflect the features of the system along with other indicators; vii) have known variability in response (Dale and Polasky, 2007). Each one of the indicators used in this study (see Table 3.3 for description and measurements) is considered to meet at least the majority of these criteria. In the following paragraphs, a brief overview and justification for the selection of indicators is provided.

The regulation of pests and diseases was represented by the area under the disease progress curve (AUDPC) of each pest or disease and the number of dead branches. The AUDPC was deduced from the incidences which were measured five times, and then standardized (sAUDPC) by the total number of days of observation.

The attack of pests and diseases can cause the defoliation of branches leading to their death; dead branches therefore can be an overall reflection of such attack (Avelino *et al.*, 2015).

For the provisioning services, the indicators were coffee yield and economic indicators such as the gross income, net income, cash flow and family benefit. This information was obtained through semi structured interviews with farmers. Economic indicators included coffee income and also incomes and/or the value of the domestic consumption of other agroforestry products (bananas and other fruits, timber). Including several agroforestry products and not only the product of the main crop is an appropriate way to reflect the real overall contribution of the coffee system to the provisioning of agroforestry products to farmer families (Rice, 2011; Cerda *et al.*, 2014).

Coffee yields were estimated for the 69 coffee plots through an empirical model. This model was calibrated from the data obtained by harvesting a subsample of three coffee plants in 20 out of the 69 coffee subplots. Out of the 20 harvested subplots, 8 were of the type CHD, 8 of CLD and 4 of CFS. Based on these data, a general linear model was developed to estimate coffee yields in all plots, including the 49 non-harvested plots. The model was initialized with coffee yield per plant as a function of coffee yield components (number of productive stems per plant; number of productive branches; number of fruiting nodes per plant; number of fruits per node), sAUDPC of pests and diseases, and number of dead branches after harvest as fixed effects. Plots were considered as random effects. The model, including only significant variables ($p<0.05$) to estimate the coffee yield, is shown in Table 3.3, and a figure showing the regression between predicted and observed (harvested) yield, and other indicators (R^2, mean of residues, mean absolute error, root-mean-squared error, relative root-mean-squared error, modeling efficiency, index of agreement) to support the model reliability, are shown in a supplementary material.

For the maintenance of soil fertility service, the indicators used were soil acidity, C, N, P and K contents. These are also considered as key indicators of soil quality and soil fertility and are key elements for crop growth (Beer *et al.*, 1998a; Swinton *et al.*, 2007).

For the carbon sequestration service, the main indicator was the total carbon stored in the aboveground biomass. However, the amounts of carbon stored per type of plant or tree were also considered, in order to show the particular contribution of different types of shade to carbon sequestration. This indicator is useful to compare the carbon storage (sequestered) among different types of agroecosystems. It is also helpful to guide better management practices to increase the carbon sequestration (Somarriba *et al.*, 2013).

Table 3.3. List of indicators of ecosystem services (ES) and the methods used for measuring them in the coffee plot network, Turrialba, Costa Rica

ES	Indicators of ES	Methods/Formulas	Data sources/times of measurement
Regulation of pests and diseases	sAUDPC of pests and diseases (%)	$AUDPC = \sum_{i=1}^{n-1} \frac{I_i + I_{i+1}}{2} \times (t_{i+1} - t_i)$ $sAUDPC = \frac{AUDPC}{Nd}$ where *AUDPC*: area under the disease progress curve; I_i: incidence of a given pest or disease at the *i*th measurement; t_i: time (in days) of the *i*th measurement; n: total number of measurements *sAUDPC*: standardized AUDPC; *Nd*: total number of days in which the plants were measured (from the first to the last measurement) Source: (Simko and Piepho, 2012)	Incidences where measured five times: 1st Fruit formation (slightly dry period); 2nd Beginning of fruit ripening (beginning of rainy period); 3rd Just before the harvest (rainy period); 4th During the peak of harvest (slightly dry period); 5th End of coffee harvest period (highest rainy period)
	Dead branches (Number MS^{-1})	The number of dead branches in the main stem (MS) of the marked coffee plants was counted	End of the coffee harvest period
Provision of agroforestry products	Coffee yield (kg fresh coffee cherries ha^{-1} yr^{-1})	*Coffee yield = (8.58 + 3.88xNPS + 1.95xNFNode + 0.03xNFNPlant - 0.18 DeadB)²* where *NPS*: number of productive stems per plant; *NFNode*: number of fruits per node; *NFNPlant*: number of fruiting nodes per plant; *DeadB*: number of dead branches in the main stem per plant. R^2 = 0.78 The model estimates the yield expressed in grams of fresh coffee cherries per plant; estimated yield in kg per hectare was deduced by using the coffee plant density of each coffee plot	Coffee yield components were counted in three coffee plants in 20 subplots, during the period of fruit formation
	Gross income (USD ha^{-1} yr^{-1}) Cash flow (USD ha^{-1} yr^{-1}) Family benefit (USD ha^{-1} yr^{-1})	*GI = AS x MP* *CF = GI – CC* *VDC = ADC x MP* *FB = CF + VDC* where: *GI*: gross income from sale of agroforestry products; *AS*: amount of agroforestry products for sales; *MP*: market price; *CF*: cash flow; *CC*: cash costs; *VDC*: value of domestic consumption; *ADC*: amount of agroforestry products for domestic consumption; *FB*: family benefit Sources: (Ambrose-Oji, 2003; Cerda *et al.*, 2014)	Data on management practices of coffee plots, costs of labor and inputs, and agroforestry production (fruits, bananas, plantains, etc.) were obtained through semi-structured interviews with the owners of the coffee plots, at the end of the coffee harvest period
Maintenance of soil fertility	Soil acidity (mg kg^{-1}) Phosphorus (mg kg^{-1}) Potassium (mg kg^{-1}) Carbon (%) Nitrogen (%)	Acidity was obtained with chemical titration with standardized solution of NaOH 0.01 Normal. The extraction of P and K was done with the method of Olsen modified; the quantification of P was done with the colorimetric method and K was determined by spectroscopy with atomic absorption C and N were determined with the method of combustion in auto-analyzer equipment Sources: (Briceño and Pacheco, 1984)	Eight subsamples of soil at a depth of 0-20 cm were taken near the trunk of eight coffee plants (at 50cm approximately) in each experimental subplot, during the peak of harvest (slightly dry period). The subsamples were mixed to obtain a composite sample for the chemical analyses
Carbon sequestration	Aboveground biomass carbon (Mg ha^{-1})	Allometric equations for the estimation of biomasses: *Coffea arabica:* *B = 10^(-1.113 + 1.578 * Log₁₀(d₁₅) + 0.581 * Log₁₀(h))* (Segura *et al.*, 2006) *Cordia alliodora: B = 10^(-0.755 + 2.072 * Log₁₀(dbh))* (Segura *et al.*, 2006) *Inga spp.: B = 10^(-0.559+2.067* Log₁₀(dbh))* (Segura *et al.*, 2006) *Bactris gasipaes: B = 0.74 * h²* (Szott *et al.*, 1993) Fruit trees: *B = 10^(-1.11 + 2.64 * Log₁₀(dbh))* (Andrade *et al.*, 2008) Other trees: *B = (21.3 - 6.95 *(dbh)+ 0.74*(dbh²))* (Brown and Iverson, 1992) *Musaceae*: *B = 0.030 * dbh ^2.13* (van Noordwijk *et al.*, 2002) where *B*: biomass (kg); Log_{10}: Logarithm base 10; d_{15}: trunk diameter (cm) at 15 cm from soil; *dbh*: trunk diameter (cm) at breast height (1.3 m from soil); *h*: height of the plant The biomass was multiplied by a 0.47 fraction to obtain the carbon content	The trunk diameters were measured as described in section 2.2.3, during the coffee fruit formation (slightly dry period) Note: for *E. poeppigiana* trees, given that they are pollarded once or twice a year, only the volume of the trunk was used for C sequestration assessment to avoid overestimations; the trunk was considered as a cylinder; the volume was then multiplied by 0.00045 kg cm^{-3} (density of the wood) to obtain the biomass

2.4. Statistical data analyses

2.4.1. Analysis of the effects of shade, altitude and management on indicators of ecosystem services

A linear model was used to estimate the effects of altitude, management intensity (quantitative data for both variables), type of shade (qualitative data), and their interactions on each specific ecosystem service indicator. Ecosystem service indicators were first checked for normality. Then, model selection procedure for each indicator was run several times. Each time, non-significant factors or interactions were removed from the model. Factors retained in the final model were those that were considered to have effects on the specific ecosystem service indicator. Analysis of variance and Fisher's LSD test ($p<0.05$) were also used to compare the effects of types of shade on ecosystem service indicators. Significant effects of double and triple interactions among the factors on ecosystem service indicators were represented graphically.

2.4.2. Analysis of relationships between ecosystem services and biodiversity

Bivariate linear regressions were performed between indicators of the four ecosystem services studied, and between indicators of ecosystem services and plant biodiversity. Significant negative relationships denote trade-offs, while significant positive relationships denote synergies (Rapidel *et al.*, 2015).

For simplification and in order to highlight only the most important relationships between ecosystem service indicators, only one indicator which better represents each ecosystem service was used in the bivariate linear regressions. The number of dead branches was chosen as representative of the regulation of pest and disease service, given that it is considered as a general indicator of the plant illness. Coffee yield was used as the indicator of provisioning services since this indicator is of interest both to smallholder farmers (looking for diversification of incomes, but always with coffee as the main source of revenue) and to medium or big farmers (with coffee as their unique product of interest). Soil acidity was chosen as representative of the maintenance of soil fertility service, considering that soil acidity is a general problem in tropical areas, and reducing acidity involves high extra costs (labor and inputs) for farmers. Total aboveground biomass carbon was used to represent carbon sequestration. Finally, the Shannon index of plant diversity was used as an indicator of biodiversity.

Regressions were performed with all the observations as a whole, and also per type of shade, with the aim of verifying if ecosystem services are related to a particular type of shade. The regressions were represented with figures, where a "desirable area" was demarcated. The "desirable area" was defined as the area where the best values of the ecosystem service indicators could be found (Rapidel *et al.*, 2015); this area was demarcated to identify which type of shade achieved those values. This type of approach and analysis has been demonstrated to be useful to evaluate and design agroecosystems (Groot *et al.*, 2012; Rapidel *et al.*, 2015).

The procedure to demarcate the "desirable area" was: 1) outlier points were removed from the figures for not taking into account very uncommon observations that could difficult the demarcation of "desirable areas"; 2) the cloud of points was divided into four quadrants based on the values of the mid-point in each axis (same distance to zero and to the maximum value, outliers excluded); and 3) the best quadrant, representing the "desirable area", was demarcated. In order to compare the abundance of different types of coffee plots in the "desirable areas", the percentage of coffee plots per type of shade in the "desirable areas" was calculated.

Statistical analysis were performed with the software R version 3.1.1 (R_Core_Team, 2014).

3. Results

3.1. Effects of altitude, shade and management intensity on indicators of ecosystem services

The factors affecting the provision of ecosystem services varied across services. In some cases, individual factors (i.e., type of shade, management intensity or altitude) had significant single effects, while in others the double or triple interactions between these factors were important (Table 3.4). We first explored the impacts of individual factors, then explained the significant double interactions and triple interactions. We have broken down the triple interactions into two double interactions *altitude x type of shade* and *type* of *shade x management intensity* to make it easier for the reader to understand the results.

3.1.1. Effects of altitude, shade and management on regulation of pest and diseases

The most common pests and diseases found in Turrialba included coffee leaf rust (*Hemileia vastatrix* Berkeley and Broome), the brown eye spot (*Cercospora coffeicola* Berk. and Curtis), anthracnosis (*Colletotrichum spp*.) and leaf miner (*Leucoptera coffeella* Guérin-Mèneville). Coffee leaf rust was the most important disease in terms of the mean sAUDPC; the sAUDPC of other pests and diseases did not exceed 5% on average (Table 3.4). Dead branches were useful to reflect the attack of pests and diseases, especially for its positive significant relationship with the sAUDPC of coffee leaf rust ($p = 0.0005$; $R^2 = 0.17$).

The effects of shade, altitude and management on pest and disease regulation were variable across different pests and diseases. Both altitude and management intensity had significant single positive effects on leaf miner insect and brown eye spot, with attack levels increasing with increasing altitudes and management intensities, irrespective of the shade type (Table 3.4). In contrast, neither altitude nor management nor shade had any effect on the prevalence of Anthracnosis. The most important disease, coffee leaf rust, was affected significantly by the double interaction *altitude x type of shade*, and was not affected in any form by management intensity. With increasing altitudes (meaning increasing rainfall and decreasing temperatures), the sAUDPC of coffee leaf rust decreased in CFS and CHD, but was not affected in CLD. These results suggest that coffee leaf rust responded more to environmental conditions than to management intensities under different types of shade (Fig. 3.1A). Interestingly, the disease behaved the same in the two most contrasting systems (full sun versus highly diversified agroforestry system). The triple interaction *altitude x type of shade x management intensity* was significant for dead branches: the number of dead branches was lower in higher altitudes (Fig. 3.1B1), and increased with increasing management intensity in CFS only, while remaining practically stable in CLD and slightly decreasing in CHD (Fig. 3.1B2).

Table 3.4. General statistical measures and effects of altitude (A), type of shade (S) and management intensity (M) on indicators of four ecosystem services provided by coffee agroecosystems in Turrialba, Costa Rica (n=69 plots)

Ecosystem service	Indicators	General statistical measures			Effects of altitude, type of shade and management intensity						
					Single effects			Interactive effects			
		Mean ± SD	Min	Max	A	S	M	AxS	AxM	SxM	AxSxM
					P	*p*	*p*	*p*	*p*	*p*	*p*
Regulation of pests and diseases	sAUDPC Rust (%)	60.8 ± 14.3	12.8	88.2				*Fig.3.1A			
	sAUDPC Cerc (%)	4.9 ± 3.3	0.7	13.8	***(+)		*(+)				
	sAUDPC Antra (%)	1.6 ± 1.2	0.0	5.3							
	sAUDPC Miner (%)	1.1 ± 0.9	0.0	4.7	**(+)		*(+)				
	Dead Branches (number MS^{-1})	8.4 ± 6.9	0.1	35.4							*Fig.3.1B1,B2
Provision of agroforestry products	Coffee Yield (kg ha^{-1} yr^{-1})†	6407 ± 4132	259	22864			**(+)				
	Gross Income (USD ha^{-1} yr^{-1})	3206 ± 1969	123	10828							*Fig.3.2B1,B2
	Cash Costs (USD ha^{-1} yr^{-1})	858 ± 772	12	3572						*Fig.3.2A	
	Cash Flow (USD ha^{-1} yr^{-1})	2348 ± 1808	-680	9229			*(+)				
	Family Benefit (USD ha^{-1} yr^{-1})	2469 ± 1798	-680	9229			*(+)				
Maintenance of soil fertility	Soil Acidity (mg kg^{-1})	169.3 ± 125.6	9.0	577.8		*Fig.3.3A					
	Soil Carbon (%)	4.8 ± 2.8	2.1	14.1							**Fig.3.4A1,A2
	Soil Nitrogen (%)	0.4 ± 0.2	0.2	1.2							*Fig.3.4A1,A2
	Soil Phosphorus (mg kg^{-1})	7.0 ± 6.4	1.5	26.8							
	Soil Potassium (mg kg^{-1})	77.6 ± 64.8	23.4	347.1		*Fig.3.3B					
Carbon sequestration	TAGB Carbon (Mg ha^{-1})	20.2 ± 10.5	2.8	51.1				*Fig3.5			

sAUDPC: standardized area under the disease progress curve; Rust: coffee leaf rust; Cerc: brown eye spot; Antra: anthracnosis; Miner: leaf miner insect; #: number; MS: main stem of coffee plants; †Fresh coffee cherries; TAGB Carbon: total aboveground biomass carbon

SD: standard deviation; Min: minimum; Max: maximum

*p-values: *p < 0.05; **p < 0.01; ***p < 0.001; (+) indicates that the effect was positive: with increasing value of the factor, the value of the response also increased; Fig: indicates that the effect was significant and the reader is directed to the indicated figure to appreciate the effect on the indicator*

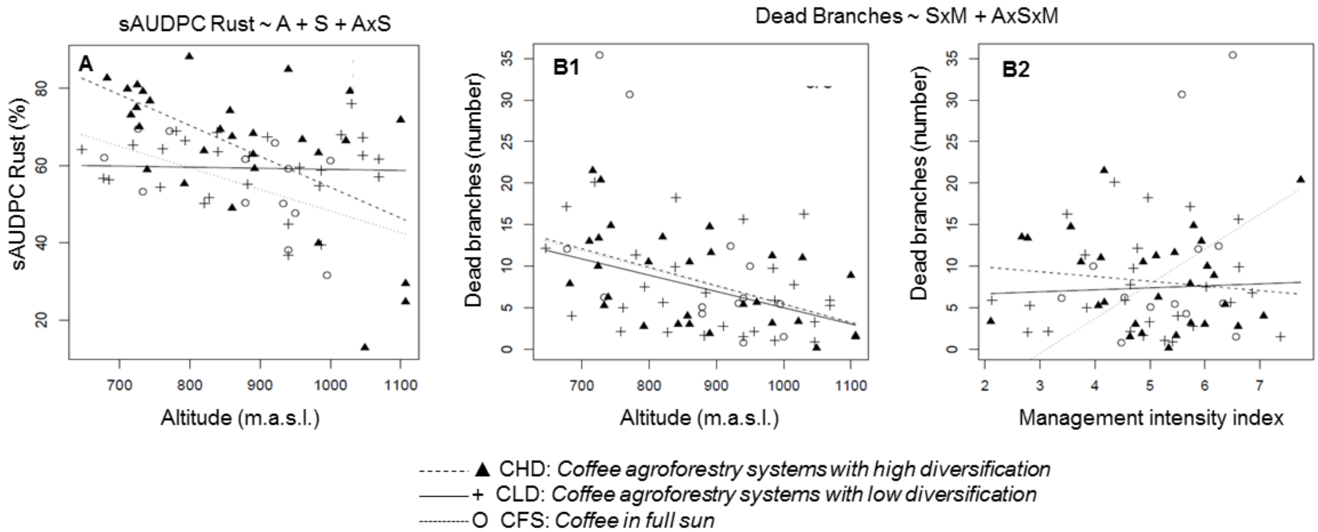


Fig. 3.1. Effects of the double interaction altitude x type of shade on the sAUDPC of coffee leaf rust (A), and of the triple interaction altitude x type of shade x management intensity on the number of dead branches (B1 and B2). The two double interactions altitude x type of shade (B1) and shade x management intensity (B2) are represented for simplification.

3.1.2. Effects of altitude, shade and management on indicators of provisioning of agroforestry products

Interestingly, neither the type of shade nor the altitude affected coffee yields. Coffee yields were only positively affected by management intensity. Management intensity also had positive significant effects on cash flow and family benefits (Table 3.4). However, there was a significant double interaction effect of *type of shade x management* on cash costs, and a significant triple interaction effect on gross income. The increasing management intensity strongly increased the cash costs for CFS and slightly for CHD, but did not have any effect on the costs of CLD (Fig. 3.2A), Gross income was always higher in coffee plots with greater management intensity (Fig. 3.2B2); but with increasing altitudes, it increased in CFS and decreased in CLD (Fig. 3.2B1). These results suggest that the costs of increasing management intensity (and therefore increasing coffee yield, cash flow and family benefits) were clearly higher in monocultures than in agroforestry systems, independent of the altitude of the plot.

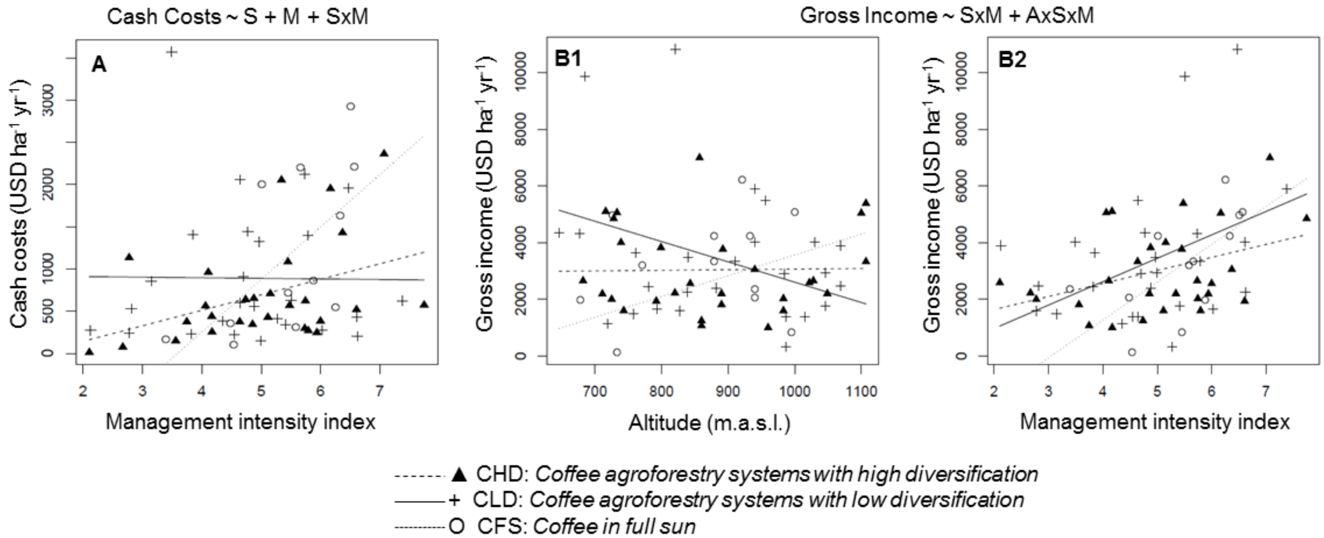


Fig. 3.2. Effects of the double interaction *type of shade x management intensity* on cash costs (A), and of the triple interaction *altitude x type of shade x management intensity* on gross income (B1 and B2). The two double interactions *altitude x type of shade* (B1) and *shade x management intensity* (B2) are represented for simplification

3.1.3. Effects of altitude, shade and management on indicators of maintenance of soil fertility

The types of shade had significant single effects on soil acidity and K; whereas there was a significant triple interaction effect of *altitude x type of shade x management intensity* on soil C and N. With increasing complexity of the shade canopy (from CSF, passing through CLD, until CHD) the acidity was lower (Fig. 3.3A) and the soil K content was higher (Fig. 3.3B). Soil C was always higher at higher altitudes (Fig. 3.4A1), but with increasing management intensity, soil C was higher in CHD than in CFS and CLD (Fig. 3.4A2). The effects on soil N were practically the same as on soil C. So, agroforestry systems maintained less acidity than monocultures in full sun conditions independent on the management intensity and altitude; highly diversified agroforestry systems had more soil K, and soil C and N contents were not affected by management intensity, in comparison to the other types of shade.

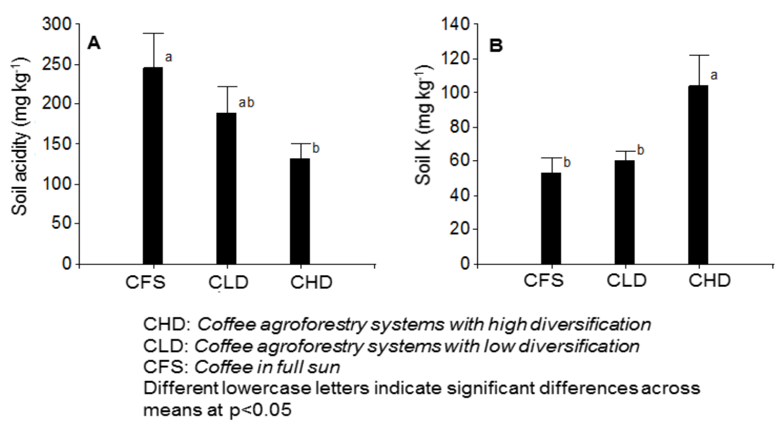


Fig. 3.3. Single effects of type of shade on soil acidity (A) and soil K (B)

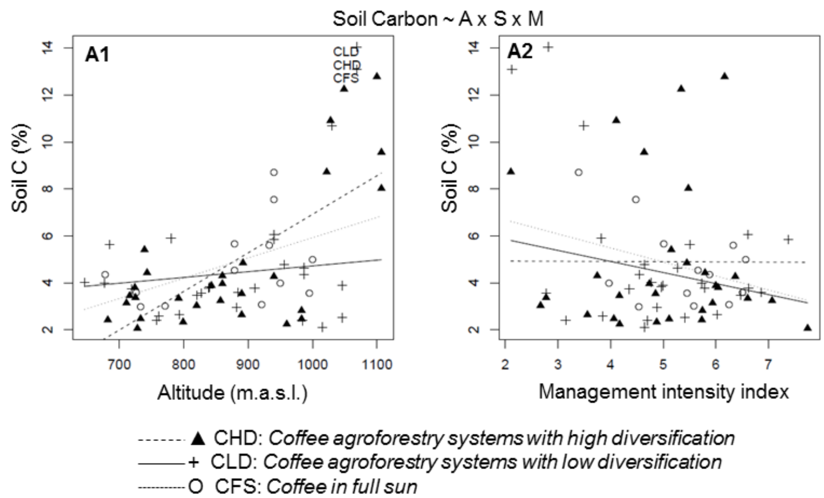


Fig. 3.4. Effects of the triple interaction *altitude x type of shade x management intensity* on soil C. The two double interactions *altitude x type of shade* (A1) and *shade x management intensity* (A2) are represented for simplification

3.1.4. Effects of altitude, shade and management on indicators of carbon sequestration

There was a significant double interaction *altitude x type of shade* effect on carbon sequestration. At higher altitudes the total aboveground biomass carbon increased in CFS and CLD compared to lower altitudes while the opposite occurred in CHD. However, in this case, it is more relevant to show that agroforestry systems on average had more than the twice the carbon stock than CFS, due to the carbon of the fruit trees, timber trees and service trees present in their shade canopies (Fig. 3.5).

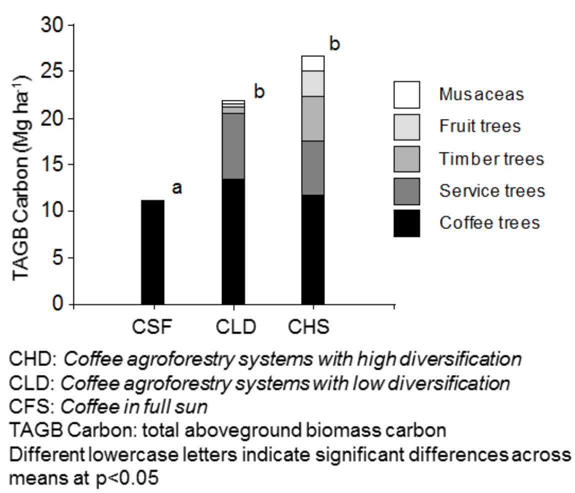


Fig. 3.5. Effects of the types of shade on the total aboveground biomass carbon

3.2. Relationships between ecosystem services, and with biodiversity

The pairwise comparisons of different ecosystem services provided by different types of shade systems showed no clear trade-offs across the provision of different ecosystem services; i.e. there were no clear patterns where the provision of one service came at the expense of another. Similarly, pairwise comparisons of the provision of different ecosystem services relative to the biodiversity present in the coffee plots showed no clear trade-offs (Fig. 3.6). Only in the case of CHD, one synergy ($p = 0.012$; $R^2 = 0.22$) was found between biodiversity and carbon sequestration; i.e. as biodiversity of the system increased, so did the amount of carbon stored in aboveground biomass (Fig. 3.6).

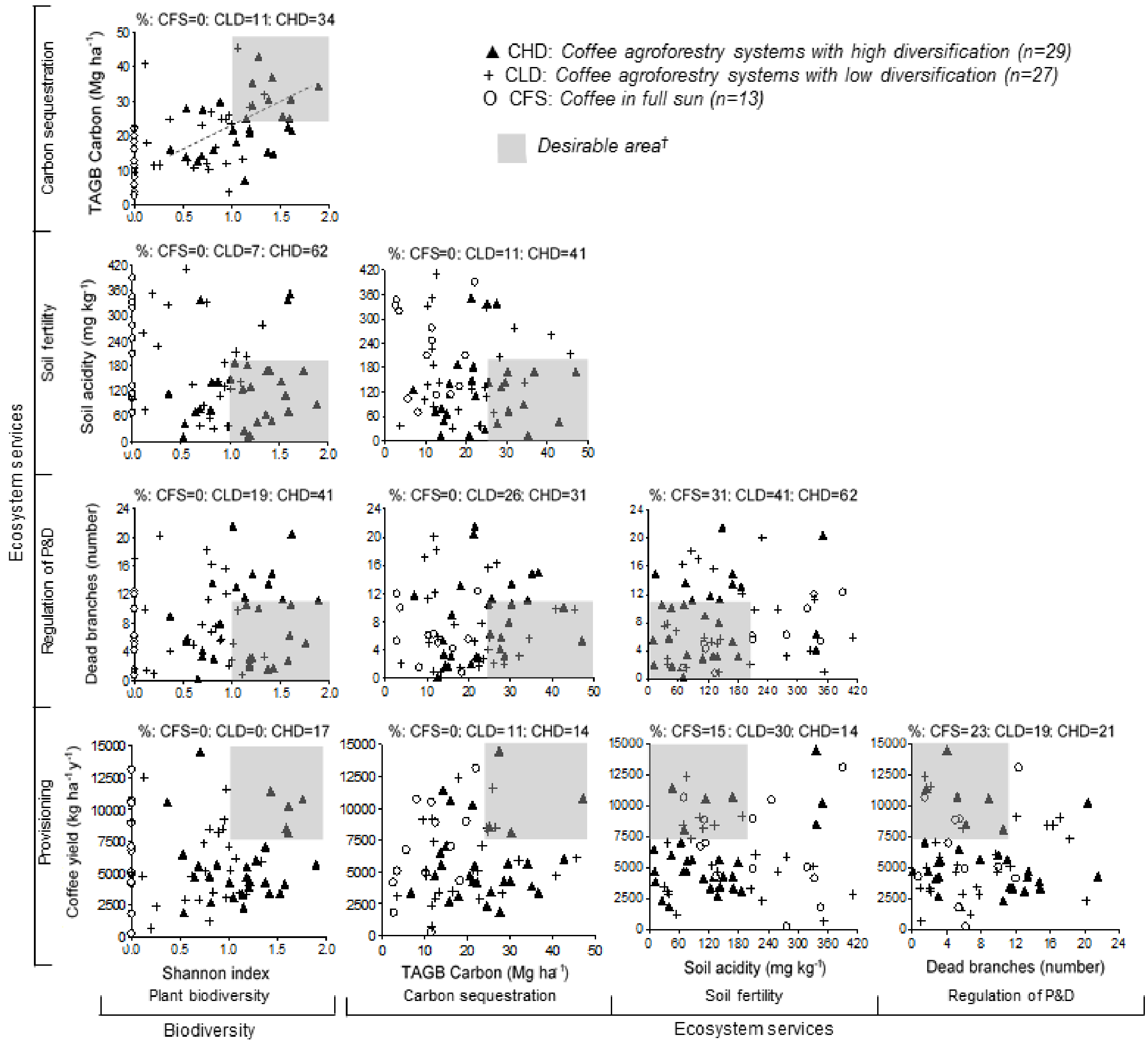
The graphs showing ‘desirable’ levels of ecosystem services indicate that plots of agroforestry systems (CLD and CHD) were more likely to be located in the ‘desirable’ quadrants than CFS (Fig. 3.6). Coffee plots of the type CHD were present in all the desirable areas, and with more plots of this type than CLD and CFS. CLD plots were present in almost all the desirable areas (except one); while CFS plots were present only in three desirable areas (Fig. 3.6).

4. Discussion

In our study, the indicators of each type of ecosystem service (e.g., regulation of pests and diseases, provisioning of agroforestry products, soil fertility, and carbon sequestration) responded differently to the effects of altitude, shade and management. In addition, there were no clear trade-offs among different ecosystem services or across ecosystem services and biodiversity. The fact that at least one indicator of three major ecosystem services was affected by the triple interaction *altitude x type of shade x management intensity* indicates that the combination of these three factors should be always considered in studies aiming to understand the provision of ecosystem services by the cropping systems under study.

4.1. Effects of types of shade on the provision of ecosystem services

The combined knowledge of single and/or interactive effects of shade with altitude and management intensity on ecosystem services is useful for understanding how to manage coffee agroecosystems to obtain the ecosystem services of interest. For instance, coffee leaf rust, the most important disease in our study, was affected by the interaction *types of shade x altitude*, but not by management intensity. This suggests that efforts to manage coffee leaf rust need to consider both the type of shade and the altitude which determines environmental and microclimatic conditions (Avelino *et al.*, 2006). Highly diversified coffee systems will be better at reducing coffee leaf rust incidences in higher altitudes; whereas in lower altitudes less diversified agroforestry systems will be more suitable. We hypothesize that the less diversified canopies maintain low moisture in lower altitudes, while in higher altitudes the highly diversified canopies maintain low temperature; both effects could reduce the development of the pathogen.



TAGB Carbon: total aboveground biomass carbon; P&D: pests and diseases

†The Desirable area is the quadrant in the figure where the most desirable values of both indicators can be found. Examples: in the combination of carbon sequestration and plant biodiversity, the desirable area is the quadrant in the upper right corner of the figure, because this was the quadrant with higher TAGB Carbon and higher Shannon index; in the combination of provision and regulation of P&D, the desirable area is the quadrant in the upper left corner of the figure, because plots in this quadrant had higher coffee yields but fewer dead branches.

Percentages (%) above each figure: represent the number of coffee plots of a given type of shade in the desirable area with respect to the total coffee plots of that type of shade. For instance, in the figure of carbon sequestration vs biodiversity, 10 coffee plots of the type CHD were identified in the desirable area, representing ~34% of the total of 29 CHD coffee plots. The only one significant relationship (a synergy) was found between biodiversity and carbon sequestration for CHD plots

Fig. 3.6. Relationships between ecosystem services and biodiversity; and percentage of different types of coffee plots achieving the most desirable values of ecosystem services.

We did not find any effect of types of shade on coffee yields. That means that agroforestry systems, besides providing several ecosystem services, did not reduce coffee yields within the studied range of shade cover (<30%). In addition, under shade, yields are more stable over time, ensuring also more stable incomes to coffee farmers (DaMatta, 2004). In contrast, coffee plantations in full sun had more dead branches, especially with high management intensities. Consequently, reduction and higher variability in yields in subsequent years can be expected (Avelino *et al.*, 2015). The reduction of yields, i.e. yield losses, is also considered as a key indicator of the regulation of pests and diseases; therefore, it should be explicitly quantified to reinforce the assessment of this ecosystem service in further studies (Avelino *et al.*, 2011; Allinne *et al.*, 2016).

We found that agroforestry systems can have lower cash costs than full sun systems. This indicates that the management intensity of these agroforestry systems can still be increased without incurring necessarily high cash costs. The shade canopy management would not mean more costs either. For instance, the cutting of banana leaves, pruning of trees and the harvesting of fruits are usually done by family members and together with operations applied to coffee plants (pruning of coffee plants, weedings, coffee harvests, etc.); that way, those activities do not imply to hire external workers nor many extra labor.

Contrary to what was expected, no interaction was found between types of shade and management on cash flow and family benefit. This reflects that coffee farmers in Turrialba rarely harvest agroforestry products (such bananas, other fruits and timber) either for sale or home consumption. In other regions where socioeconomic conditions are worse, the contribution of agroforestry products is more important to farmers. For instance, Guatemalan coffee farmers harvest fruits for sale, and Peruvian farmers use fruits for home consumption (Rice, 2011). In remote areas of different countries of Central America, the agroforestry products contribute significantly more than the main crop (cocoa) to family benefit (Cerda *et al.*, 2014). Other studies demonstrate that sustainable high timber harvest ratio (>4 m^3 ha^{-1} $year^{-1}$ ~ 265 USD ha^{-1} $year^{-1}$) in agroforestry systems is possible (Somarriba *et al.*, 2014). The important point is that plants and trees that are present in the coffee agroforestry systems can be harvested when farmers need products for consumption or for sale, which is not possible in coffee in full sun. This is especially important during crises of low coffee prices or low coffee production (Rice, 2008).

In our study, coffee agroforestry systems (~25 Mg C ha^{-1}) had more than twice the amount of aboveground carbon than coffee in full sun. In other parts of the world, coffee agroforestry systems can store even more carbon due to their more diverse and denser shade canopies. For instance, the stock of aboveground biomass can reach ~40 Mg ha^{-1} in Guatemalan, Nicaraguan and Mexican coffee agroforestry systems (Soto-Pinto *et al.*, 2010; Haggar *et al.*, 2013; Rahn *et al.*, 2013), ~43 Mg ha^{-1} in Indonesia (van Noordwijk *et al.*, 2002), and ~67 Mg ha^{-1} in Togo (Dossa *et al.*, 2008). Carbon sequestration within the coffee agroforestry systems could be enhanced by the establishment and management of trees with tall and coarse trunks, which can store high amounts of carbon vertically, also increasing the shade canopy production without reducing the yields of the main crop (Somarriba *et al.*, 2013).

Agroforestry systems had better soil fertility than coffee in full sun, considering single effects of types of shade or in interaction with management intensity. Other studies have also documented the importance of shade for soil fertility in coffee agroecosystems. More trees means less loss of nitrogen (Tully *et al.*, 2012). The use of bananas can help improve the cation exchange capacity (Tully *et al.*, 2013). In our study, shade was important for reducing acidity and increasing K independently of the other factors, and was capable of maintaining higher soil C and N levels with increasing management intensity (especially in CHD plots). The use of shade trees and bananas can reduce the need for nitrogen fertilizers and amendments for correcting the soil acidity, and thus, reduce both soil contamination and production costs. In addition, although soil physical indicators that are also important for soil fertility were not measured, it is known that soil C is closely related to organic matter and better soil physical conditions (Swinton *et al.*, 2007).

In summary, overall, agroforestry systems have proven to be more effective than full sun coffee for the provision of ecosystem services, and consequently for improving farmers´ livelihoods. Furthermore, the delivery of multiple ecosystem services can considerably increase the economic value of the land (Dale and Polasky, 2007; Pert *et al.*, 2013), which is important for both current and future generations.

4.2. Relationships between ecosystem services and biodiversity

We did not observe trade-offs between ecosystem services or between individual ecosystem services and biodiversity. Trade-offs reported in the scientific literature on agroforestry systems such as the ones between yields and carbon sequestration, yields and biodiversity (Wade *et al.*, 2010), and yields and regulation of diseases (López-Bravo *et al.*, 2012) were expected, but did not happen. The lack of trade-offs among the studied ecosystem services is a novel result. This can be explained by the fact that the provision of ecosystem services is the consequence of both the composition and the management of the system (Rapidel *et al.*, 2015), i.e. of the interaction between these factors. Thus, highly diversified systems should be able to produce high levels of ecosystem services without trade-offs with the appropriate management. Several studies have documented that management of the system can strongly affect coffee pollination and production (Boreux *et al.*, 2013), provision of other tree products (Ango *et al.*, 2014), regulation of diseases (Avelino *et al.*, 2006), soil fertility (Méndez *et al.*, 2009) and carbon sequestration (Häger, 2012). In Turrialba, many different combinations of types of shade and cropping practices can be found, with varying responses in terms of ecosystem services provision. Some coffee plots had low values of an ecosystem service and other coffee plots of the same type of shade had high values of the same ecosystem service; that is why no trade-offs between ecosystem services were found.

On the other hand, not all the synergistic relationships between ecosystem services and plant biodiversity reported in the literature were observed. The well-known synergy between carbon sequestration and plant biodiversity (Häger, 2012; Richards and Mendez, 2014) was also confirmed in our study, but only for CHD plots. There are recent studies in other crops that report synergies between plant biodiversity and economic indicators, which were not found in our study, apparently because coffee farmers in Turrialba did not take advantage of agroforestry products as mentioned earlier. Such studies demonstrate that synergies are found in multistrata agroforestry systems with a notable presence of productive species in the shade canopy and adequate management, including frequent harvests of agroforestry products (Cerda *et al.*, 2014; Cardozo *et al.*, 2015). Although we did not find significant synergies between ecosystem services, the fact that most of coffee systems that achieved high levels of ecosystem services were highly diversified agroforestry systems (desirable areas in Fig. 3.6), indicates that the use of shade trees and other plants in combination with the coffee crop is essential for providing multiple ecosystem services simultaneously. These specific agroforestry systems should be studied in more detail to identify the specific set of practices which makes these systems more successful (Rapidel *et al.*, 2015).

4.3. Important caveats on the results of ecosystem service indicators

There are several important caveats related to our results. First, we have used a specific set of indicators (Table 3.3) to measure ecosystem services. While these indicators were carefully selected and have been used in other studies on ecosystem services, it is possible that the use of other ecosystem service indicators might lead to different results. As more research on the provision of multiple ecosystem services by coffee systems becomes available, the extent to which our results can be generalized across ecosystem services will become clear. Second, we used a specific method to identify the desirable levels of ecosystem services and to explore which systems were more likely to provide these desirable levels (Rapidel *et al.*, 2015). While this method has been proven to be useful in identifying systems with different levels of ecosystem service provision, the boundaries of the desirable area are subjective and the use of different boundaries or thresholds for desirability could lead to slightly different results. Finally, our results are applicable only to the range of altitudes, shade cover and management intensity that we studied (Tables 1 and 2). In other regions where coffee is grown at much higher elevations or with higher shade, the relationships between altitude, management practices, shade type, and ecosystem services could vary.

4.4. Key aspects for the design and management of coffee agroforestry systems

We have shown that agroforestry systems with low or high diversification allow the delivery of multiple ecosystem services without compromising coffee yields in smallholder farms. Therefore, smallholder farmers should be able to design and manage shade trees without undermining their productive and economic objectives, and at the same time ensure the delivery of other ecosystem services.

Key aspects that should be considered in the design and management of coffee agroecosystems are described below.

Efforts to ensure that coffee systems deliver ecosystem services need to consider not only the type of shade, but also the altitude where the plantation occurs and the management practices implemented, as demonstrated in the present and in other studies (Boreux *et al.*, 2013). The altitude must be especially considered regarding pests and diseases (Allinne *et al.*, 2016); the decision on the type of shade and management practices to implement at a given altitude, must be in accordance with the expected pest or pathogen and their responses to environment conditions. In case that various altitudes also involve types of soil with different characteristics, the types of shade and management practices should aim at controlling soil fertility.

Farmers manage trees mainly according to their objectives and socioeconomic conditions, in order to obtain the most beneficial services and avoid the most problematic disservices (Haggar *et al.*, 2013; Ango *et al.*, 2014). When designing new coffee systems, farmers should integrate tree and plant species that have good prices in the local market and require little management. The inclusion of bananas, plantains and productive (fruit and timber) tree species can help ensure products for farmer households as well as providing other services (e.g. shade, protection, N fixation, etc.). The decision on how many trees and which tree species to use, must also consider the soil fertility, because higher tree densities than those shown in this study (CHD in Table 3.2) could cause disservices such as cost increments and competition for soil water and nutrients (Dale and Polasky, 2007; Zhang *et al.*, 2007). High tree densities are associated with less N, K and soil C; in contrast, tree species richness was positively related to soil pH, CEC, Ca, and Mg (Méndez *et al.*, 2009; Tully *et al.*, 2013). In areas with low rainfall and well-marked dry periods, the densities of trees and plants in the shade canopy should be lower than those in Turrialba where water availability is not limiting.

The significant effects of management intensity on indicators of provisioning service, reported in this and in other recent studies (Cerda *et al.*, 2014; Cardozo *et al.*, 2015), suggest that both low and highly diversified shade canopies require specific management strategies to deliver multiple ecosystem services (Power, 2010). In areas where severe attacks of diseases occur, such as coffee leaf rust, and where soils are considered of medium fertility (like in Turrialba), we suggest that the best option for smallholder farmers is to use medium management intensity under shade. We define this management as two fungicide sprays per year against diseases, at least one soil fertilization, at least one coffee plant pruning, the necessary controls of weeds (hopefully manual weedings), the necessary labors of harvest according to the ripening of coffee fruits, and maintaining the shade cover around 30% during the year. Such management should also include resistant coffee varieties to pests and diseases (Avelino *et al.*, 2015), avoid unnecessary excessive quantities of inputs (McCook and Vandermeer, 2015) and use family labor to reduce cash costs. In order to avoid or reduce disservices like poisoning of the family members, death of non-target organisms, contamination of soil and emissions of greenhouse gases (Dale and Polasky, 2007; Zhang *et al.*, 2007; Power, 2010), farmers should try to use low toxic preventive fungicides and traps for insects instead of chemical insecticides. When possible, organic fertilizers (composts, manures) should be applied with the aim to also improve soil physical characteristics.

Agricultural extension and training of farmers, as well as adequate certifications, market based incentives and payment of ecosystem services, can help promote the adoption of well-designed, sustainable coffee agroforestry systems that provide both economic and ecological benefits. There is a lack of agroforestry education and extension to farmers (Lasco *et al.*, 2015), which can be attended through trainings with participatory methodologies (Tscharntke *et al.*, 2011). Certifications and market based incentives to increase prices (Williams-Guillén and Otterstrom, 2014), the economic valuation of ecosystem services (Zhang *et al.*, 2007; Avelino *et al.*, 2011), and payment of ecosystem services (Le Coq *et al.*, 2011) are needed to encourage better management of agroforestry systems in general.

5. Conclusions

The effectiveness of different types of shade to provide major ecosystem services in coffee plantations depends both on the altitude where the coffee is grown and how the system is managed. In our study, no trade-offs were found between different ecosystem services, and between ecosystem services and biodiversity. This indicates that it is possible to increase the provision of ecosystem services without decreasing the provision of other ecosystem services (at least under the types of coffee shade, management and altitudes studied here). More ecosystem services are provided by coffee agroforestry systems than by coffee systems in full sun. Coffee agroforestry systems should be designed with diversified, productive shade canopies and managed with a medium intensity of cropping practices, in order to ensure the continued provision of multiple ecosystem services.

6. Acknowledgements

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7. Supplementary material of manuscript 1

Supplementary material for the model of estimation of coffee yield, explained in section 2.3 and presented in Table 3.3.

Measures of the yield observed values:

Mean of yield observed values = 1201 g fresh coffee cherries per plant

Min = 4 ; Max = 4145

Model:

$$Coffee\ yield = (8.58 + 3.88xNPS + 1.95xNFNode + 0.03xNFNPlant - 0.18\ DeadB)^2$$

where *NPS*: number of productive stems per plant; *NFNode*: number of fruits per node; *NFNPlant*: number of fruiting nodes per plant; *DeadB*: number of dead branches in the main stem per plant.

Indicators for evaluation of the model:

Bias = 25.4

R^2 = 0.78

MAE = 290.0

RMSE = 426.4

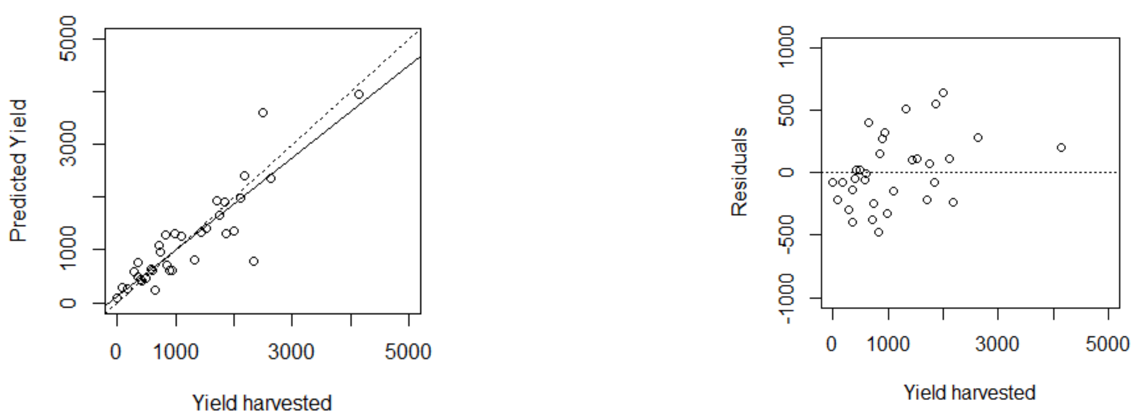
RRMSE = 35.5

EF = 0.77

IA = 0.94

Where Bias: mean of residues; MAE: mean absolute error; RMSE: root-mean-squared error; RRMSE: relative root-mean-squared error; EF: modeling efficiency; IA: index of agreement

Figure of the adjustment of the model



Dashed line in the graphic is the relation 1:1; continuous line is the predictions

All the indicators of evaluation in general indicated that the model is acceptable for the estimation of coffee yields; considering also that the range of observed yield was high.

Chapter 4. Modelling coffee yield losses caused by pests and diseases

Manuscript 2: Primary and secondary yield losses caused by pests and diseases: assessment and modeling in coffee

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Primary and secondary yield losses caused by pests and diseases: assessment and modeling in coffee

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Abstract

The assessment of crop yield losses is needed for the improvement of production systems that contribute to the incomes of rural families and food security worldwide. However, efforts to quantify yield losses and identify their causes are still limited, especially for perennial crops. Our objectives were to quantify primary yield losses (incurred in the current year of production) and secondary yield losses (resulting from negative impacts of the previous year) of coffee due to pests and diseases, and to identify the most important predictors of coffee yields and yield losses. We established an experimental coffee parcel with full-sun exposure that consisted of six treatments, which were defined as different sequences of pesticide applications. The trial lasted three years (2013-2015) and yield components, dead productive branches, and foliar pests and diseases were assessed as predictors of yield. First, we calculated yield losses by comparing actual yields of specific treatments with the estimated attainable yield obtained in plots which always had chemical protection. Second, we used structural equation modeling to identify the most important predictors. Results showed that pests and diseases led to high primary yield losses (26%) and even higher secondary yield losses (38%). We identified the fruiting nodes and the dead productive branches as the most important and useful predictors of yields and yield losses. These predictors could be added in existing mechanistic models of coffee, or can be used to develop new linear mixed models to estimate yield losses. Estimated yield losses can then be related to production factors to identify corrective actions that farmers can implement to reduce losses. The experimental and modeling approaches of this study could also be applied in other perennial crops to assess yield losses.

Key words: attainable yield, actual yield, conceptual model, dead branches, biennial behavior of production, structural equation modeling

1. Introduction

Crop losses due to pests and diseases are a major threat to incomes of rural families and to food security worldwide (Savary and Willocquet, 2014; Avelino *et al.*, 2015). Quantitative information on crop losses and a better understanding of their drivers have been mentioned as essential to (i) evaluating the efficacy of crop protection practices (Oerke, 2006), (ii) assessing systems sustainability (Cooke, 2006), (iii) making better decisions for integrated pest management (Savary *et al.*, 2006a), and (iv) evaluating the effectiveness of pest and disease regulation as an ecosystem service (Avelino *et al.*, 2011; Allinne *et al.*, 2016).

For some authors, crop loss is the reduction of the crop yield, defined both in terms of quantity and quality, that can occur in the field (pre-harvest) or in the storage (post-harvest) due to biotic or abiotic factors (Oerke, 2006; Savary *et al.*, 2006b). For others, crop loss also includes the decrease in the value and financial returns of the crop (Nutter *et al.*, 1993). Furthermore, crop losses comprise primary and secondary losses. Primary crop losses are those caused in the specific year when pest and disease injuries occur; secondary crop losses are those resulting from negative impacts of pests and diseases of the previous year (Zadoks and Schein, 1979). In annual crops, the inoculum accumulation of pathogens in soil or in seeds and tubers remaining from the previous year, can cause secondary losses. These losses however can be avoided by implementing crop rotations or chemical treatments. In perennial crops, premature defoliation or the death of stems and branches caused by leaf injuries lead to loss of vigor and decreased production (secondary losses) in subsequent years. In this case, such secondary losses cannot be avoided since they come from already damaged plants (Zadoks and Schein, 1979).

A first step to crop-loss assessment is the quantification of yield losses, defined as the difference between attainable yield and actual yield (Nutter *et al.*, 1993). The attainable yield is the site-specific yield achieved under the geographic and ecological conditions (radiation, temperature, water, and soil nutrients) of the location, with the best available production techniques and without the influences of any yield-reducing factors such as pests and diseases (Oerke *et al.*, 1994; Savary *et al.*, 2006b). The actual yield is also a site-specific yield, achieved with the available practices at farm level and resulting from the influence of yield-reducing factors such as pests and diseases (Nutter *et al.*, 1993; Oerke *et al.*, 1994).

Despite the importance of information on crop losses, the main reviews on the topic agree that efforts to quantify yield losses and analyze their causes have been scant (Oerke, 2006; Savary *et al.*, 2006b; Cheatham *et al.*, 2009; Savary and Willocquet, 2014). Estimated yield losses for major food and cash crops (rice, wheat, barley, maize, potatoes, soybeans, cotton, and coffee) at country and regional levels have been mainly based on information from literature and trials conducted by chemical companies (Oerke *et al.*, 1994; Oerke, 2006). However, in addition to being limited, yield-loss assessments normally do not consider secondary yield losses, which means that the real situation is not addressed. This is an issue for all crops, and even more so for perennial crops such as coffee, for which we assume that secondary losses may have far-reaching consequences.

Quantification of yield losses in perennial crops is particularly complex because of (i) the typical biennial pattern of production in these crops, characterized by a repetitive cycle of high production one year and low production the following year (DaMatta *et al.*, 2007; Smith and Samach, 2013), and (ii) the sustained presence of pests and diseases along years, which is an issue specially under tropical climates, which have no marked cold season to disrupt the life cycles of pests and diseases, and where susceptible organs of plants, particularly leaves, are almost always available.

The ongoing coffee crisis in Latin America and the Caribbean highlights the need for reliable assessments of coffee yield loss for governments, banks, and development agencies in order to implement appropriate economic responses (Avelino *et al.*, 2015). This crisis has been triggered by a severe outbreak of coffee leaf rust (*Hemileia vastarix* Berkeley and Broome) combined with the inefficient management of coffee plantations, which have led to a decrease in production estimated between 30% and 50% in some countries, negatively affecting the incomes and food security of families involved directly or indirectly with coffee activities (Baker, 2014; Avelino *et al.*, 2015; McCook and Vandermeer, 2015). The quantification or reliable estimation of yield losses is a major challenge and requires trustworthy methods (Cooke, 2006; Savary and Willocquet, 2014).

Our research was a first attempt aimed at (i) quantifying primary and secondary coffee yield losses through field experimentation, taking into account a three-year dataset on foliar pests and diseases of coffee, and (ii) identifying the most important predictors of coffee yields and yield losses, through structural equation modeling.

2. Materials and methods

2.1. Conceptual model for coffee yield losses

In order to guide the understanding of the experimental design and the statistical modeling, we developed a conceptual model for the assessment of coffee yield losses caused by pests and diseases. The model was built based on the scientific literature and the expert knowledge of the authors of this research. The model reflects effects over several years to assess secondary losses and take into account the already mentioned biennial behavior of coffee production (DaMatta, 2004; DaMatta *et al.*, 2007). The model includes yield components, pests and diseases, and dead productive branches as the main yield drivers; attainable and actual yields are the outputs for the estimation of yield losses. Coffee yield is determined by the shoot growth of the previous year since fruits appear on the one-year-old wood (DaMatta *et al.*, 2007); pests and diseases can affect that shoot growth or cause the death of branches, compromising the yields of current and future years (Avelino *et al.*, 2015). Here we consider that dead productive branches are those branches which had fruits but died mainly due to the impacts of pests and diseases (e.g. defoliation and dieback). Yield loss is derived from the difference between attainable and actual yield (Nutter *et al.*, 1993). According to the effects of pest and disease injuries and dead branches, different actual yields, and therefore, different types of yield losses (primary and secondary) can be seen each year (Fig 4.1).

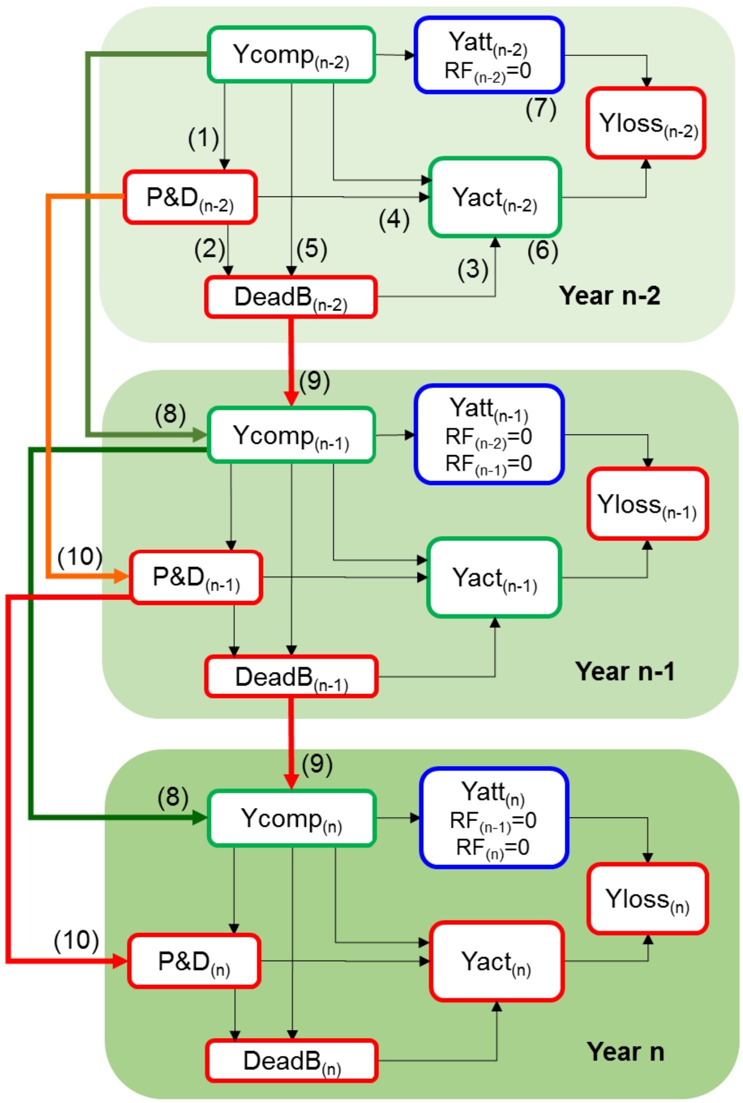

Fig. 4.1. Conceptual model for the assessment of coffee yield losses caused by pests and diseases

Ycomp: yield components (productive stems and branches, fruiting nodes and fruits); P&D: pests and diseases; DeadB: dead productive branches; RF: reducing factors (P&D, DeadB); Yatt: attainable yield; Yact; actual yield; Yloss: yield loss; n: a given year.

Within each year: (1) Yield components can affect pests and diseases, since it is known that high fruit loads can make the plant more susceptible to pathogens. (2) Pests and diseases can cause defoliation and thus contribute to the death of branches. (3) In turn, this will cause the drying and death of the fruits that were growing on them. (4) Pests and diseases can also reduce the photosynthetic capacity of the branch without causing its death but negatively affecting the development of fruits. (5) Yield components also influence the death of branches, because high fruit loads could cause the exhaustion of plant tissues, especially when the plant does not have enough nutrients to sustain growth and production. (6) The actual yield, then, as an output of the system, depends on yield components, pests and diseases, and dead productive branches. (7) The attainable yield would be the output of the system if there were no influences of pests and diseases (P&D = 0) nor dead branches (DeabB = 0), i.e., reducing factors = 0. Yield loss for a specific year (primary yield loss) is the difference between this attainable yield and the actual yield.

Across years: (8) Yield components depend on the yield components of the previous year. A year with yield components achieving high values will be followed by a year with low values, and vice versa (biennial behavior of coffee production). (9) Yield components of the current year also depend on the number of dead branches of the previous year, as those branches will no longer be able to bear fruits. (10) Pest and disease abundance of the previous year will influence their abundance in the current year through its effect on primary inoculum.

Secondary yield loss is the difference between the attainable yield (i.e., the yield with no yield-reducing factors in the previous and current years) and the actual yield obtained with reducing factors >0 in the previous year and reducing factors = 0 in the current year.

2.2. Location and experimental design

We established an experimental coffee parcel with full-sun exposure on flat terrain at 648 masl (meters above sea level) on the farm of the Tropical Agricultural Research and Higher Education Center (CATIE) in Turrialba, Costa Rica. Turrialba is considered a rainy area, with slightly dry periods from March to April and rainiest periods in June and July. In the past 10 years, the mean annual rainfall was 2781 mm and the mean annual temperature was 22.2°C, with small variations among months.

The experimental parcel was planted in 2010 and the experiment lasted until 2015. All coffee plants (*Coffea arabica* L.) were of the dwarf variety Caturra, with planting distances of 2 m between coffee rows and 1 m between plants within the row (5,000 plants ha^{-1}). From 2010 to 2013, all coffee plants received the same management protocol, provided by the farm with three yearly applications of fertilizers, three of fungicides and other pesticides, and three of herbicides. From 2013 to 2015 only fungicide applications varied, depending on the treatments defined for this study. The experiment had six treatments, each one consisting in a particular sequence of chemical pesticide applications, including fungicides and insecticides (Fig. 4.2). Insecticides were applied only in 2013. Given that the incidence of pests was low, insecticide applications were suspended the following years, which, in addition, allowed us to avoid the use of highly toxic products. Control of coffee berry borer (*Hypothenemus hampei* Ferrari) in the entire experimental parcel was achieved by using traps with chemical attractants. Each treatment had four randomized plots, representing a completely randomized design. Each plot had five rows and six plants per row, where external rows and plants were defined as borders. In the three central rows, we marked six plants and three branches per plant for measurements. One coffee plant pruning per year was performed in 2014 and 2015 to remove exhausted orthotropic stems, leaving two to four productive orthotropic stems per plant.

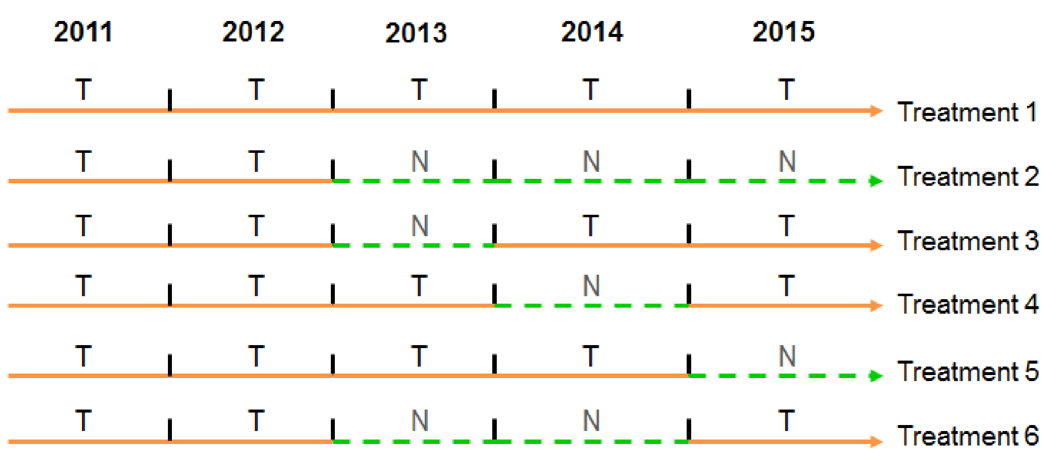

Fig. 4.2. Scheme of the treatments applied in the coffee experimental parcel

T: Treated with pesticides. Each year T consisted in: Fungicides: three applications of Opera® (13.3% Pyraclostrobin and 5% Epoxiconazole) with doses = 2 ml lt^{-1} ; one of Soprano 25SC® (12.5% Carbendazim and 12.5% Epoxiconazole) with doses: 1.25 ml lt^{-1}; **Insecticides** (only in 2103)**:** three applications of Sumithion 50EC® (50% Fenithrotion) with doses = 4 ml lt^{-1} ; one of Solver 48EC® (48% Chlorpyrifos) with doses = 2.5 ml lt^{-1}
N: No pest and disease control

Treatments were designed for two specific objectives: (i) to generate different levels of injuries in different years in order to calculate yield losses through comparisons of treatments, i.e., by experimentation; and (ii) to generate variability of data among coffee plants regarding yield components, pest and disease injuries, and yields, to be used in the statistical modeling.

2.3. Measurement and calculations of the variables studied from 2013 to 2015

We quantified several different yield components, yields, pest and disease injuries, and dead productive branches over all the years of the study, except for the latter, which were not measured in 2013 (Table 4.1). Basic statistical measures were calculated for these variables for each year.

Yield losses were expressed in units of the product or in percentages (Savary and Willocquet, 2014):

$$Yloss = Yatt - Yact \qquad Yloss\ (\%) = \frac{Yatt - Yact}{Yatt} \times 100$$

where Yloss is yield loss in grams of fresh coffee cherries per plant (g plant^{-1}), Yloss (%) is yield loss in percentage, Yatt is attainable yield, and Yact is actual yield.

Table 4.1. Variables characterized in the coffee experimental parcel, Turrialba, Costa Rica

<table>
<tr><th></th><th>Variables</th><th>Descriptions</th><th>Time of assessments</th></tr>
<tr><td rowspan="5">Yield components</td><td>Stems</td><td>Number of productive orthotropic stems per plant</td><td rowspan="4">All these variables were quantified before the beginning of the harvest season, when small fruits were already visible</td></tr>
<tr><td>Branches</td><td>Number of productive branches per plant</td></tr>
<tr><td>Fruiting nodes</td><td>Number of fruiting nodes per plant</td></tr>
<tr><td>Fruits per node</td><td>Number of fruits per node each 25 fruiting nodes; then averaged</td></tr>
<tr><td>Coffee yield</td><td>Grams of ripe fresh coffee cherries per coffee plant</td><td>Harvests every 15 days</td></tr>
<tr><td rowspan="8">Pests and diseases of leaves</td><td>sAUDPC of:</td><td rowspan="6">Incidences for each pest and disease per plant were calculated based on the accumulated number of infected/infested leaves with respect to the total of accumulated leaves at each assessment date
$$AUDPC = \sum_{i=1}^{n-1} \frac{I_i + I_{i+1}}{2} \times (t_{i+1} - t_i)$$
where AUDPC: area under the disease/pest progress curve; I_i: incidence of a given pest or disease at the ith measurement; t_i: time (in days) of the ith measurement; n: total number of measurements
$$sAUDPC = AUDPC/Nd$$
where sAUDPC: standardized area under the disease/pest progress curve; Nd: total number of days of the assessment period (Simko and Piepho, 2012)

sAUDPC P&D: represents the sAUDPC of all pests and diseases together</td><td rowspan="6">Infected/infested/healthy leaves were counted monthly</td></tr>
<tr><td>Coffee leaf rust</td></tr>
<tr><td>Brown eye spot</td></tr>
<tr><td>Anthracnose</td></tr>
<tr><td>Coffee leaf miner</td></tr>
<tr><td>sAUDPC P&D</td></tr>
<tr><td>sAUPC of severity</td><td>A scale from 0 to 6 (the higher the number, the higher the severity), based on the size and number of symptoms on leaves, was used to assign a level of overall severity to marked branches. Averages of severities were calculated per plant, and then standardized areas under the progress curve (sAUPC) of severity were calculated.</td><td>Severity was assessed monthly for all visible injuries</td></tr>
<tr><td>Dead branches</td><td>Number of dead productive branches per plant</td><td>Quantified at the end of the coffee harvest period</td></tr>
</table>

Coffee leaf rust (*Hemileia vastarix* Berkeley and Broome); brown eye spot (*Cercospora coffeicola* Berk and Curtis); anthracnose (*Colletotrichum spp.*); coffee leaf miner (*Leucoptera coffeella* Guérin-Mèneville).

2.4. Effects of treatments on coffee yields and calculation of yield losses through experimentation

Three-year sequences of chemical treatments ($T_{(n-2,\ n-1,\ n)}$) starting from 2011, 2012, and 2013, constructed based on the scheme of treatments (Fig. 4.2) and shown in Table 4.2, were used to estimate primary and secondary yield losses. Yield losses were obtained by comparing the yield obtained in the third year (*n*=2013, 2014 or 2015*)* of the completely protected treatment (TTT), i.e., the attainable yield, with the yield obtained in the third year of other sequences of chemical treatments, i.e., different actual yields.

Table 4.2. Number of plots and plants considered in the analysis, according to different three-year sequences of chemical treatments and quantification of yield losses

Three-year sequence of chemical treatments $T_{(n-2,\ n-1,\ n)}$	Last year of the three-year sequence of chemical treatments						Total plots	Total plants	Quantification of yield losses in the third year
	2013		2014		2015				
	plots	plants	plots	plants	plots	plants			
TTT^{Yatt}	12	72	8	48	4	24	24	144†	
TTN	12	72	4	24	4	24	20	120†	TTT-TTN → 1ry Yloss[I]
TNT			4	24	4	24	8	48†	TTT-TNT → 2ry Yloss[II]
TNN			8	48			8	48†	TTT-TNN → 1ry + 2ry Yloss[III]
NTT					4	24	4	24	
NNT					4	24	4	24	
NNN					4	24	4	24	
Total	24	144	24	144	24	144	72	432	

n: year; T: treated with pesticides; N: no pest and disease control.
A treated plot (T) in a given year in the experimental parcel implies that reducing factors (RF) = 0 (Fig. 4.1), because no pests and diseases or dead branches were expected; on the contrary, a no-treated plot (N) implies that reducing factors >0. For instance, the three-year sequence TNT implies that $RF_{(n-2)} = 0$, $RF_{(n-1)} > 0$, $RF_{(n)} = 0$. The difference between the attainable yield (Yatt, obtained in TTT) and the actual yield in TNT in year *n* represents a secondary yield loss (Yloss) in year *n* caused by pest and disease injuries of year *n-1*.
†Number of plants used to test the effects of sequences of chemical treatments with the Model T.
All losses are quantified on year n; [I]: primary losses resulting from the year *n* injuries; [II]: secondary losses resulting from the year *n-1* injuries; [III]: primary and secondary losses resulting from the years *n* and *n-1 injuries*.

We applied a linear mixed model for the analysis. The sequence of chemical treatments in the last three years ($T_{(n-2,\ n-1,\ n)}$) was declared as the fixed effect, which included the sequences TTT, TTN, TNT, and TNN. Given that these sequences were present during several years (submitted to the biennial behavior of coffee production) and there were soil heterogeneity conditions among plots in the experimental parcel (especially the acidity of soil), the year and the plot were declared as random effects:

$$Yield\_Plant \sim T_{(n-2,\ n-1,\ n)} + (1\,|\,\mathrm{Year}) + (1\,|\,\mathrm{Plot}) \qquad \text{(Model T)}$$

where $T_{(n-2,\ n-1,\ n)}$ is the statistical treatment (a three-year sequence of chemical treatments) and Yield_Plant is the coffee yield per plant in the third year.

The sequences NTT, NNT, and NNN were not included in the analysis because yields for the third year of these sequences were available only in the last year of the experiment (2015). The year 2015 was a year of low production in all of the experimental parcel (similar yields for all treatments). We checked that the inclusion of such sequences with the proposed Model T would have introduced a statistical unwanted bias in the results.

Model T was run in the program R (R_Core_Team, 2014) with the package 'lme4' (Bates *et al.*, 2015), using the chi-squared test to determine significant effects of the three-year sequence of chemical treatments ($P<0.05$) on coffee yields of the third year. The normal of the data was verified by inspecting a histogram of the residuals. Significant differences between yields of the third year were analyzed using the LSD test, with the package 'predictmeans' (Welham *et al.*, 2004; Luo *et al.*, 2014).

2.5. Statistical modeling for the identification of the most important predictors of primary and secondary coffee yield losses

We used all variables showed in Table 4.1 as predictors of coffee yield. According to our conceptual model (Fig 1), the actual yield of a specific year is a direct function of yield components, pest and disease injuries, and dead productive branches for that year, and also the result of the indirect effect of the same predictors of the previous year. To take into account the chain of effects described in Fig 1, we applied piecewise structural equation modeling (PiecewiseSEM) (Lefcheck, 2016). PiecewiseSEM is a confirmatory path analysis that works with two or more linear models in order to test direct and indirect effects for estimation of the final response variable—in our case, yields. PiecewiseSEM permits testing whether the equations used to explain intermediate and final response variables are independent and a reflection of the true paths involved, for which *P*-value must be higher than the significance threshold (we chose $\alpha=0.05$) to ensure a good fit of the modeling. PiecewiseSEM also provides the Fisher's C statistic and the likelihood degrees of freedom (K) to calculate the Akaike's information criterion (AIC = C + 2K) (Lefcheck, 2016).

Given that this modeling approach considers the influence of predictors of the previous year, it was necessary to use data of two consecutive years for each plant. We used data from 2014 and 2015 only, as no information was available for 2011 and 2012, and the number of dead branches was not assessed in 2013. Contrary to previous analysis, no treatments were excluded. However, since the estimation of yield losses makes sense only if plants have yield components >0, only plants with yield components >0 were used. Models were then constructed with data of only 82 plants out of the original 144.

We used linear mixed models to estimate the actual yield for 2015 and to model intermediate predictors influenced by those of the previous year (2014); in all cases, the plot was declared as random effect. The list of models included in PiecewiseSEM was as follows:

$Yact_{(n)} \sim Yield\ components_{(n)} + sAUDPC_{(n)} + DeadB_{(n)} + (1 \mid \text{Plot})$ (Main model)

$Yield\ components_{(n)} \sim Yield\ components_{(n-1)} + DeadB_{(n-1)} + (1 \mid \text{Plot})$

$sAUDPC_{(n)} \sim sAUDPC_{(n-1)} + Yield\ components_{(n)} + (1 \mid \text{Plot})$

$DeadB_{(n)} \sim sAUDPC_{(n)} + Yield\ components_{(n)} + (1 \mid \text{Plot})$

where Yact is actual coffee yield per plant; sAUDPC is the standardized area under the disease progress curve of pests and diseases (we included the sAUDPC of each pest and disease individually and also the sAUDPC P&D -all pests and diseases together-); DeadB is the number of dead productive branches; (n) represents the year of interest (2015 in this case), and (n-1) represents the previous year (2014).

We ran the PiecewiseSEM on this list in the software R (R_Core_Team, 2014) with the package ‘piecewiseSEM’ (Lefcheck, 2016). Different combinations of predictors in the models were tested, verifying that the *P*-value was $P > 0.05$ to ensure a good fit, and suppressing predictors with no significant effects ($P > 0.05$) or with very low effects (i.e., low coefficients <0.001) in each test. Which means that different lists of models were run until we selected the best list, based on two criteria: the list that showed the lowest values of AIC and the highest values of R^2 provided by the PiecewiseSEM. Finally, based on the significance and on the coefficient of each predictor involved in the best list, also provided by the PiecewiseSEM procedure, we identified the most important predictors of yields, and of primary and secondary yield losses.

3. Results

3.1. Statistical description of the variables studied from 2013 to 2015

The most observable differences among variables across years happened in 2014 when the yield components, coffee yields, and dead productive branches were noticeable higher, especially in comparison with 2015, illustrating the biennial behavior of coffee production (Table 4.3). The most important diseases were coffee leaf rust and brown eye spot, according to sAUDPC values. Both diseases had wide ranges during the three years of measurements, especially in 2015 when some plants reached the 100% of sAUDPC. The other disease (anthracnose) and the insect coffee leaf miner had low levels (<7% on average). The overall severity of pest and disease attacks appeared to be important especially in 2014 and 2015, when the maximum values were between 5 and 6, which means that branches had most of the leaves with large symptoms caused by pests and diseases (scale 5) or were already dead (scale 6).

Table 4.3. Basic statistics of the variables studied in the coffee experimental parcel, Turrialba, Costa Rica

Variable	2013		2014		2015	
	Mean±SD	Range	Mean±SD	Range	Mean±SD	Range
NS (number plant^{-1})	3±1	1–5	3±1	1–5	3±1	0–5
NPB (number plant^{-1})	145±77	64–433	97±42	0–209	25±32	0–134
NFN (number plant^{-1})	421±238	9–1158	500±329	0–1901	129±181	0–796
NF (number node^{-1})	4±1	2–7	3±1	0–6	1±1	0–4
Coffee yield (g plant^{-1})	2172±1354	5–7295	2416±1859	0–11600	680±1038	0–4862
sAUDPC_Rust (%)	28±14	3–71	36±14	0–71	34±15	0–100
sAUDPC_Cerc (%)	26±10	5–58	22±9	3–46	23±12	0–100
sAUDPC_Ant (%)	5±4	0–19	6±5	0–23	3±4	0–17
sAUDPC_Min (%)	1±1	0–8	4±4	0–15	2±3	0–12
sAUDPC_All (%)	59±10	32–88	67±9	44–96	58±12	24–100
sAUPC_Sev (scale)	2.7±0.6	1.4–4.4	3.6±0.7	2.1–5.5	2.9±0.8	1.5–5.8
DeadB (number plant^{-1})	46±26	14–114	26±31	0–193	8±6	0–29

SD: standard deviation; NS: number of productive orthotropic stems; NPB: number of productive branches; NFN: number of fruiting nodes; NF: number of fruits; Coffee yield: grams of fresh coffee cherries; sAUDPC: standardized area under the disease progress curve; Rust: coffee leaf rust; Cerc: brown eye spot; Ant: anthracnose; Min: coffee leaf miner; All: all pests and diseases; sAUPC_Sev: standardized area under the progress curve of severity; DeadB: number of dead productive branches

3.2. Effects of three-year sequences of chemical treatments on yield losses

The effects of the sequences TTT, TTN, TNT, and TNN (Model T) on coffee yield in the third year were significant (*P*-value = 0.0009) (Fig. 4.3). The yield of the sequence TTT (the attainable yield), with 2235 g fresh coffee cherries plant^{-1}, was the highest and significantly different from the other yields, which allowed us to quantify yield losses. The largest yield loss was the total yield loss (primary + secondary yield losses = 57%) as a consequence of no pest and disease control in the current and in the previous year (TNN). Primary yield losses (TTN) were high (26%), and secondary yield losses, due to previous-year injuries (TNT), resulted in even higher losses (38%).

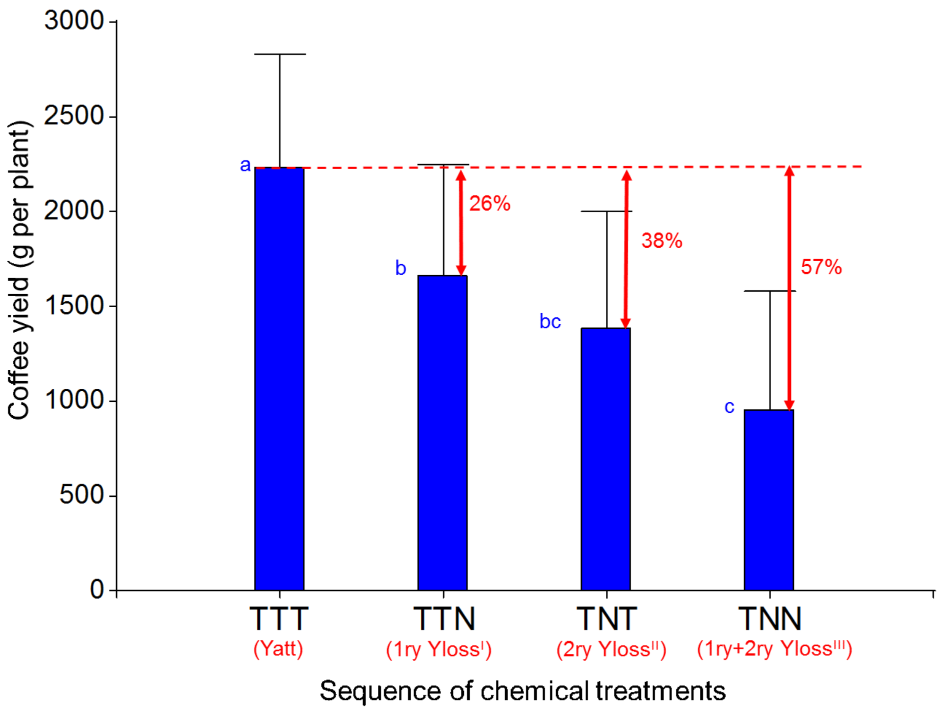

Fig. 4.3. Yields and primary and secondary yield losses resulting from the sequences of chemical treatments TTT, TTN, TNT, TNN

T: treated with pesticides; N: no control of pests and diseases. Different lowercase letters between bars indicate significant differences (*P*<0.05) between coffee yields. Red arrows and numbers in percentages represent the yield losses.
Yatt: attainable yield; Yloss: coffee yield loss; ^{I}: primary losses resulting from the current-year injuries (year n); ^{II}: secondary losses resulting from year n-1 injuries; ^{III}: primary and secondary losses (total losses) resulting from years n and n-1 injuries.

3.3. The most important predictors of primary and secondary coffee yield losses

We found two lists of linear mixed models that fitted well in the PiecewiseSEM (*P* >0.05). The first list (List 1) was the largest, with models for all the predictors that direct or indirectly affected actual yield, including yield components, pest and disease injuries, and severity and number of dead productive branches of current and previous years. The second list (List 2) was a simplified modeling, including only yield components and number of dead productive branches in the models (Table 4.4). The significance of the variables and the magnitude of their influences (coefficients), for both List 1 and List 2, are shown in Fig.4.4. From List 1, it can be deduced that effects within a specific year and across years explained in the conceptual model (Fig 1) are validated. This modeling however used several predictors (variables) which are not easily measurable in the field (e.g., sAUDPC of pests and diseases), and which were not significant in the modeling. Therefore, we used the List 2 to determine the most significant and easily measurable yield predictors. In addition, List 2 exhibited a lower AIC value than List 1 (Table 4.4).

Thereby, the most important and useful predictors of yield, considering influences of at least two years, were: the number of fruiting nodes of the current year characterizing yield components, and the number of dead productive branches of both the current year and the previous year, characterizing reducing factors of yield (List 2 in Fig. 4.4). Dead productive branches of the previous year were considered as predictors of secondary yield losses since they had negative effects on the number of branches bearing fruiting nodes of the following year, representing a one-year delayed indirect effect on the actual yield. Dead productive branches of the current year were considered as predictors of primary yield losses since they had direct negative effects on the number of fruiting nodes remaining at the harvest time, i.e. the actual yield.

Table 4.4. Models for the estimation of actual coffee yields in 2015 (g of fresh coffee cherries per plant) with data of 2014 and 2015, through Piecewise structural equation modeling

Number of list	Linear mixed models for the PiecewiseSEM	Indicators for the PiecewiseSEM			
		Fisher's C	K	AIC	*P*-value
List 1	$Yact_{(n)}$ ~ $NFN_{(n)}$+$DeadB_{(n)}$+sAUDPC $P\&D_{(n)}$+(1|Plot) **Main model**	36.03	23	82.03	0.21
	$NFN_{(n)}$ ~ $NFN_{(n-1)}$+$DeadB_{(n-1)}$+(1|Plot)				
	$sAUDPC\_All_{(n)}$ ~ sAUDPC $P\&D_{(n-1)}$+$sAUPC\_Sev_{(n-1)}$+$NFN_{(n)}$+(1|Plot)				
	$DeadB_{(n)}$ ~ sAUDPC $P\&D_{(n)}$+$sAUPC\_Sev_{(n)}$+$NFN_{(n)}$+(1|Plot)				
List 2	$Yact_{(n)}$ ~ $NFN_{(n)}$+$DeadB_{(n)}$+(1|Plot) **Main model**	5.20	13	31.20	0.27
	$NFN_{(n)}$ ~ $DeadB_{(n-1)}$+(1|Plot)				
	$DeadB_{(n)}$ ~ $NFN_{(n)}$+(1|Plot)				

Yact: actual coffee yield per plant; NFN: number of fruiting nodes per plant; DeadB: number of dead productive branches per plant; sAUDPC P&D: standardized area under the disease progress curve of all pests and diseases; sAUPC_Sev: standardized area under the progress curve of severity; $_{(n)}$: current year (2015); $_{(n-1)}$: previous year (2014); K: likelihood degrees of freedom; AIC: Akaike's information criterion.

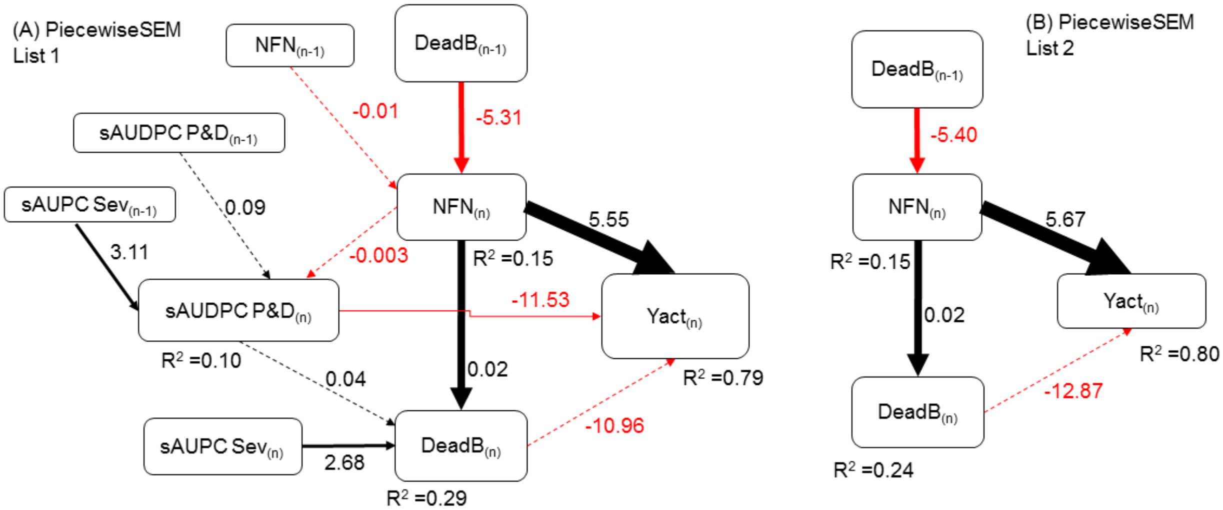

Fig. 4.4. Structural equation models for the estimation of actual coffee yield for Piecewise List 1 (A) and Piecewise List 2 (B) presented in Table 4.4

Yact: actual coffee yield per plant; NFN: number of fruiting nodes per plant; DeadB: number of dead productive branches per plant; sAUDPC P&D: standardized area under the disease progress curve of all pests and diseases together; sAUPC Sev: standardized area under the progress curve of severity; $_{(n)}$: current ye
ar (2015); $_{(n-1)}$: previous year (2014).
Arrows represent relationships among variables; black arrows denote positive relationships and red arrows denote negative relationships; arrows for no significant paths (*P* >0.05) have dashed lines; arrows with significant paths (P <0.05) have continuous lines.
The numbers near the arrows are the regression coefficients. The thickness of the significant paths was scaled based on the magnitude of the standardized regression coefficients (not shown in the figures).

4. Discussion

4.1. Primary and secondary yield losses

Our study shows that both primary (26%) and secondary yield losses (38%) caused by foliar pests and diseases can be severe in a perennial crop. Efforts to estimate yield losses have increased in the last decades, but most of them concentrated in annual crops and focused on primary yield losses. On annual crops, yield losses due to pests and diseases were estimated from 24% to 41% in rice in Asia (Savary *et al.*, 2000a), 5% to 96% in potatoes in France (Rakotonindraina *et al.*, 2012), and up to 7 t ha^{-1} in wheat in France (Zhang *et al.*, 2006). In perennial crops, studies on apple and other stone fruits reported yield losses that reached up to 5% in the Netherlands (van Leeuwen *et al.*, 2000). In Thailand yield losses of cotton were estimated up to 100% (Castella *et al.*, 2007). In coffee, yield losses were reported from 13% to 45% in Brazil (Barbosa *et al.*, 2004). At a global scale, yield losses ranging from 20% to 40% were quantified in rice, wheat, barley, maize, potatoes, soybeans, cotton, and coffee in different countries and regions (Oerke *et al.*, 1994; Oerke, 2006). In all these cases, yield losses were assessed in the current year, and therefore we assumed that they were primary yield losses. The ranges of these primary yield losses are wide, depending on the type of crop/variety and pests and diseases. In our study, we provided new quantitative data indicating that primary yield losses on a perennial crop such as coffee can reach 26% due to a set of foliar pests and diseases.

To our knowledge, our study was the first attempt to quantify secondary yield losses, which could have important implications for perennial crops if they are as high as the one we estimated for coffee (38%). In accordance with the hypothesis proposed by Avelino and colleagues (Avelino *et al.*, 1991; Avelino *et al.*, 1993;

Avelino *et al.*, 1999; Avelino *et al.*, 2015), our study confirms that the attacks of foliar pests and diseases have delayed impacts in coffee. We show that secondary yield losses are an important issue in perennial crops whose production depends on the growth of organs in the previous years. In annual crops, secondary yield losses are principally related to the inoculum of pathogens in the soil or to infected/infested seeds (Zadoks and Schein, 1979). However, annual-crop farmers normally select or buy the best and healthiest seeds, do rotations, or sometimes disinfect soil before sowings; therefore, expected secondary losses should be quite low. On the opposite, in perennial crops, secondary yield losses, resulting from previous year damages, cannot be avoided. Losses over several consecutive years are even expected, which can only be reduced by implementing appropriate practices to recover plant growth.

The finding that the number of dead productive branches of a previous year is the most important reducer of the number of fruiting nodes of the next year (Fig. 4.4) has implications for the management of coffee plant architecture. In areas under disease pressure, it would be better to promote more developing branches (for the next year) and more productive branches without many fruiting nodes per branch than to have few productive branches and many fruiting nodes per branch. That way the risk of losses due to the death of branches would be reduced.

Our findings can also contribute to the explanation of severe reductions in productivity in perennial crops, as happened during the coffee crisis in Central America. This crisis started in 2012 as a combination of the outbreak of coffee leaf rust and inefficient management of coffee plantations (Avelino *et al.*, 2015). From that year, the production decreased in two successive years in most of the countries. By analyzing the evolution of coffee production from the year prior to the crisis to the present, we deduced that: the total production in Central America in the harvest year 2012-13 decreased about 10% with respect to 2011-12, which could be attributed in part to the primary losses as a consequence of the first impacts of the disease; but the most severe impacts (defoliation and/or death of branches or plants) were seen in the harvest year 2013-14, with a 20% of reduction with respect to 2011-12, reflecting the seriousness of secondary losses (Fig. 4.5). The production started to recover in the harvests 2014-15 and 2015-16, in part due to control actions conducted at national and regional levels and to pruning operations as stumping (total cut of coffee plants at 30-40 cm from the ground level) conducted by farmers in 2013, with production recovering two or three years later (Baker, 2014; Avelino *et al.*, 2015).

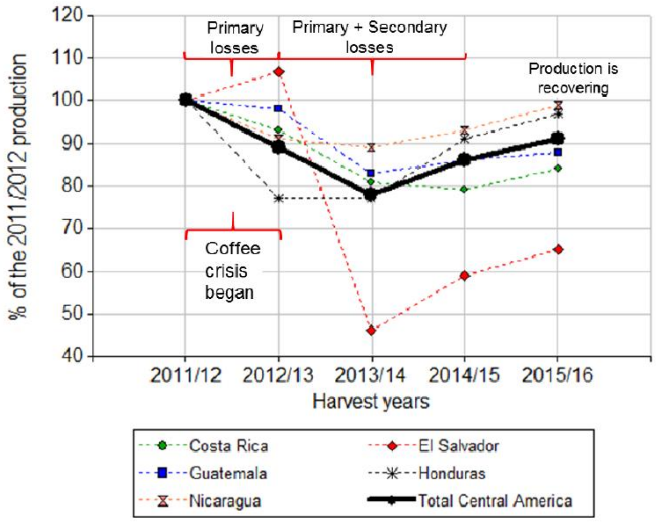

Fig. 4.5. Evolution of coffee production in Central America as a percentage of the production in the harvest year 2011/2012

Curves were constructed with data of the International Coffee Organization (ICO, 2016); the production in 2011/2012 (before coffee crisis) was taken as the reference, corresponding to 100%.

4.2. Methodological aspects of experimentation and modeling

Our experimental approach was useful in overcoming the difficulties of quantifying yield losses. Most attempts to assess yield losses were based on statistical relationships between yields and descriptors of specific pests or diseases in empirical models. However, these relationships can be masked or weakened by several confounding factors such as interactions among pests and diseases, interactions among these pests and diseases and other factors impacting on production, such as physiological effects of high temperatures, soil acidity and overbearing disease (dieback of plants) (Avelino *et al.*, 2006; Cooke, 2006). For instance, in the case of coffee leaf rust, researchers assumed that the disease did not cause primary yield losses because its incidence was positively related to fruit load and even to the yield of the current year (the higher the yield, the higher the coffee rust incidence) (Avelino *et al.*, 1993). Our experiment, based on treatment sequences, enabled us to estimate both primary and secondary yield losses.

In addition, our modeling approach was useful to identify the number of dead productive branches as a predictor of the impact of pests and diseases on yield and at the same time as a pathway (driver) that leads to yield losses. Models to estimate yield losses should be improved by adding this kind of variables that represent mechanistic links (Kropff *et al.*, 1995; Cooke, 2006; Savary *et al.*, 2006b). The modeling also revealed that pests and diseases were not important individually for the estimation of coffee yield. Only sAUDPC of all pests and diseases together and their overall severity fitted well in the PiecewiseSEM, confirming the relevance of the assessment of injury profiles, defined as a given combination of injury levels caused by a set of pests and diseases in a crop cycle (Savary *et al.*, 2006b).

For coffee, there are few but important physiological process-based (mechanistic) models that could benefit from our findings. One of these models is capable of simulating plant growth and production under agroforestry systems in Central America. Another model was developed in Brazil and predicts growth of vegetative parts and yield components, and incorporates dynamics of the coffee berry borer and its natural enemies (Gutierrez *et al.*, 1998). These mechanistic models have a general value and can be used in different conditions, as was done in Colombia using the last model (Rodríguez *et al.*, 2013). Our findings on the importance of dead productive branches as yield-reducing factors can possibly be incorporated into such models to assess yield losses due to pests and diseases.

4.3. Practical applications considering management and other production factors

Similar approaches of experimental design and modeling used in this study, could be applied to other perennial crops to assess primary and secondary yield losses. The data of yield losses can be used to quantify economic losses in different agroecosystems, as well as to assess the effectiveness of pest and disease regulation as an ecosystem service, in terms of reduced or avoided losses (Cheatham *et al.*, 2009; Avelino *et al.*, 2011; Allinne *et al.*, 2016).

Furthermore, once yield losses are quantified, then the main causes can be analyzed, shedding light on how to improve production systems. Several studies, for instance in wheat (Zhang *et al.*, 2006; Robin *et al.*, 2013), rice (Savary *et al.*, 2000b), and coffee (Allinne *et al.*, 2016), highlight the importance of identifying production factors (environment, topography, soil, associated plant biodiversity) that influence injury profiles, but few studies related production factors directly to yield losses, with very limited exceptions—such as a yield loss assessment due to pests and diseases in rice (Savary *et al.*, 2000a). In the case of coffee and other crops that can be grown in agroforestry systems, the associated plant biodiversity and shade cover are probably important production factors that need to be considered in crop-loss assessments due to their influence on crop growth and fruit load , pest and disease injury levels (Avelino *et al.*, 2006), and branch dieback (DaMatta, 2004).

There are also several studies on the reduction of crop yields, referred as “yield gap”. The yield gap, in a broad definition, is “the difference between two levels of yield,” which can be chosen according to particular objectives (FAO and DWFI, 2015). Some studies quantified the yield gap as the difference between the site-specific potential yield (obtained with no limitations of nutrients and water, or pest or disease attacks) and the observed actual yield (Lobell *et al.*, 2009; van Ittersum *et al.*, 2013; Schierhorn *et al.*, 2014; Pradhan *et al.*, 2015). Others quantified the yield gap as the difference between the maximum attainable yield identified in a whole region and the actual yield on a given farm (de Ponti *et al.*, 2012; Mueller *et al.*, 2012; Wang *et al.*, 2015). In both cases, yield gaps can be considered as yield reductions caused not only by pests and diseases but also by other production factors. Like yield losses, yield gaps were estimated mainly for annual crops, and few for perennial ones. One study quantified coffee-yield gaps from 45% to 57% in Uganda, identifying poor management practices, poor soil fertility, and the coffee twig borer as the main causes (Wang *et al.*, 2015). From this study, we can hypothesize that coffee yield losses also vary according to different production factors and management.

The most important and useful predictors identified in this study could be considered as indicators of yield losses, and used to estimate yield losses in farms of different coffee-growing areas. Measurements can be easily done at three critical times: dead productive branches just after the current harvest, fruiting nodes after the next flowering, and finally, the dead productive branches at the end of the next harvest. The data needed to estimate primary and secondary yield losses can therefore be obtained in the lapse of one year. These predictors could be included in linear mixed models to estimate actual yields, using random effects to consider the influence of local production factors. The random effects can improve the estimations, especially when working in large areas (with diverse production factors). Once the equations are obtained, the predictors representing reduction factors (dead productive branches in this case) can be set as "zero" to estimate attainable yields, and then calculate yield losses (attainable yield – actual yield). Such attainable yields are the yields without negative impacts of the current and the previous year, making possible to calculate both primary and secondary yield losses, as suggested in the conceptual model (Fig 1).

Once the models and equations with their respective coefficients are defined for a given coffee-growing area, they can be used to estimate the yield losses that have already occurred as well as to predict what to expect in the next harvest. Such predictions, along with the identification of the main causes of yield losses, can act as motivators for improvement of the production system, because the user (farmer, technician, or researcher) will know how much losses could be avoided and how much could be gained by implementing the necessary measures in the system (Savary and Willocquet, 2014).

5. Conclusions

Our work demonstrates that foliar pests and diseases in a perennial crop can lead to high primary yield losses and even higher secondary yield losses. To our knowledge, this research is the first to quantify secondary yield losses in a perennial crop, revealing the importance of its assessment since high secondary yield losses, as in the case of coffee, can have severe consequences for crop production over several years.

Fruiting nodes and dead productive branches were the most important and useful predictors of coffee yields and yield losses. Both predictors had significant effects in the modeling and are indicators easy to measure in the field.

Our research contributes to the field of crop losses, providing experimental and modeling approaches that could be used in perennial crops to estimate primary and secondary yield losses. The usefulness of this contribution for further studies is that once the yield losses are estimated, economic losses can be deduced and the main causes of losses identified, allowing the farmer to take corrective actions.

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Chapter 5. Coffee yield losses due to injury profiles under different management strategies and production situations

Manuscript 3: Primary and secondary yield losses caused by injury profiles under different management strategies and production situations in coffee agroecosystems

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Primary and secondary yield losses caused by injury profiles under different management strategies and production situations in coffee agroecosystems

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Abstract.

Knowledge on crop losses and their causes is needed to improve the regulation of pests and diseases in agroecosystems. In this study, we worked on the question: what is the impact of injury profiles on primary and secondary yield losses of coffee under different management strategies and production situations? We worked with two-year data of 61 coffee plots to construct typologies representing injury profiles (set of injuries caused by pests and diseases), management strategies (cropping practices and inputs), production situations (topoclimate, soil fertility, types of shade) and physiological characteristics of coffee plants (productive characteristics and nutrition of coffee plants) in each year, though multivariate techniques. We estimated attainable yields, actual yields, and primary and secondary yield losses through linear mixed models and divided each one in three categories. We tested significant associations of injury profiles with all the other typologies, and with categories of yields and losses with Fisher´s exact test; and we used correspondence analysis and clustering in order to determine groups of profiles most linked among each other. The main finding of this study was that gradients of yield losses (from low to high losses) were positively related to gradients of injury profiles (from slight to highly severe profiles), which depended on particular combinations of production situations, management strategies and physiological characteristics of coffee plants. Injury profiles had impacts on yield losses especially in a year of high coffee production (primary yield losses), but compromising also the yields of the next year (secondary yield losses). We found that the most severe injury profiles (high injury levels of coffee leaf rust and dieback) resulted in high primary yield losses (~30%) and even higher secondary yield losses (~70%). Furthermore, primary and secondary yield losses were positively associated; indicating that by assessing the first it is possible to deduce the second. The analytical approach of this study enabled us to reveal the serious consequences of injury profiles for coffee farmers and support the necessity to monitor them during several continued years. Key insights for the management of coffee agroecosystems were derived in order to better regulate injury profiles.

Key words: Agroforestry; Attainable yield; Climate; Modeling; Nutrient deficiencies; Pests and diseases; Shade; Soil

1. Introduction

Knowledge on how to reduce crop losses caused by pests and diseases is a major need for thousands of rural families worldwide. Based on this knowledge, the decisions for integrated pest management can be improved, and agroecosystems capable to offer high crop yields based on regulation of pests and diseases can be developed (Savary *et al.*, 2006a; Avelino *et al.*, 2011). However, information on crop losses is still scarce, possibly because their assessment is difficult, particularly on perennial crops (Cerda *et al*., 2016, Chapter 4). In addition, crop losses may be affected by management and production situations (Savary *et al.*, 2000a; Willocquet *et al.*, 2002). And injury levels caused by pests and diseases may also vary, the injuries themselves can be different and the impact of these injuries on yield may also differ according to management and production situations (Savary *et al.*, 2000a; Allinne *et al.*, 2016). As a consequence, crop loss assessments require first to determine injury levels caused by pests and diseases, and then to assess their impacts on crop losses under given management strategies and production situations.

The first step is to assess the levels of pests and diseases injuries (symptoms or signs) and determine injury profiles. An injury profile is a given combination of injury levels caused by a set of pests and diseases in a crop cycle (Savary *et al.*, 2006b). Farmers never face only one pest or disease. Generally, several of them attack the crop. The levels of injuries depend on the production situation and the management strategy. In this study, we assumed that a production situation is the set of biological, chemical and physical components of the agroecosystem, and the socioeconomic and environmental (climate and topography) conditions under which a crop is grown, excluding crop management (Breman and de Wit, 1983; Aubertot and Robin, 2013). It is considered that a management strategy, which involves a set of cropping practices and inputs, depends on the farmer´s decisions under a given production situation (Aubertot and Robin, 2013).

The second step is to quantify crop losses and relate them to the injury profiles in an integrated approach which considers management strategies and production situations. Crop loss is the reduction of the quantity and/or quality of the crop product due to biotic or abiotic factors (Oerke, 2006). In this study we focused on pre-harvest yield losses as representative of crop loss. Yield loss is the difference between attainable yield and actual yield (Nutter *et al.*, 1993). The attainable yield is the site-specific yield obtained under the production situations of the site, with the best available cropping practices, and without the influences of pests and diseases (reducing factors) (Oerke *et al.*, 1994; Savary *et al.*, 2006b). The actual yield, which is also site-specific, is the result of the cropping practices that farmers actually apply and of the impacts of pests and diseases (Nutter *et al.*, 1993; Oerke *et al.*, 1994). Furthermore, there can be primary and secondary yield losses; the first one is caused by the impact of pests and diseases on the yield of the current year, while the second is the loss resulting from negative impacts of pests and diseases of the previous year (Zadoks and Schein, 1979). In perennial crops, both should be considered, because secondary yield losses can be even higher than primary yield losses (Cerda *et al*., 2016, Chapter 4).

Despite the relevance of studying the injury profiles in an integrated approach, studies on their relationships with management strategies and production situations, and their impacts on yield losses are still scarce. In annual crops this approach was developed in rice (Savary *et al.*, 2000a) and in wheat (Zhang *et al.*, 2006) where relationships among production situations-injury profiles-yield losses were explicitly assessed and led to develop crop loss models (Willocquet *et al.*, 2002). In perennial crops, such as coffee, which can be grown under many types of systems (e.g. from monocultures at full sun exposure to highly diversified agroforestry systems) and have a biennial production cycle, characterized by a high production one year and a low production the next one (DaMatta, 2004), that kind of studies are even scarcer. Several studies on coffee show relationships between different biophysical factors (shade, soil, management, etc.) and specific pests and diseases, and deduced control options (Soto-Pinto *et al.*, 2002; Avelino *et al.*, 2006; Avelino *et al.*, 2007; López-Bravo *et al.*, 2012). However, only one recent study analyzed coffee injury profiles in their globality, i.e. a set of injury levels, in relation with yield under different production situations and management systems (Allinne *et al.*, 2016). The authors concluded to the necessity of explicitly relating these yields with yield losses in order to propose more performant management systems.

In this study, we raised and worked on the following question: what is the impact of injury profiles on primary and secondary yield losses of coffee under different management strategies and production situations? We aimed to contribute to the research field of crop losses with several methods of analysis and with new knowledge on yield losses and their drivers in perennial crops.

The objectives of this study were: i) to explain the associations between injury profiles and different management strategies and production situations, and ii) to assess associations between such injury profiles and primary and secondary yield losses of coffee. These associations were analyzed within the same year and from one year to another, in order to take into account the biennial production cycle. We hypothesized that the impacts of injury profiles on coffee yield losses differ from one year to another, depending on combinations of management strategies and production situations. Based on our findings, we provided insights on management strategies for regulating pests and diseases injuries under different production situations, and thus to conduct to low yield losses in coffee agroecosystems.

2. Materials and methods

We worked in a coffee plot network where we determined different typologies of management strategies, production situations, injury profiles, physiological characteristics of coffee plants, and categories of yields and yield losses. Each typology was composed by two or more groups of coffee plots that shared similar characteristics. Each group was then considered as a profile. Each profile can then be associated with profiles of other typologies. Below, we first present a conceptual model on how yield losses occur and then explain our methods to test the associations between injury profiles and yields, on one side, and between injury profiles and physiological characteristics of coffee plants, management strategies and production situations, on the other side.

2.1 Conceptual model

In order to illustrate how management strategies and production situations can influence injury profiles and yield losses, we developed a conceptual model based on expert knowledge and literature. In this model, we defined the environment and the management strategies as the two main factors affecting the system (the coffee plot). The coffee plot comprises four main components: shade canopy, injury profiles, coffee plants and soil. Such a system can produce several outputs as agroforestry products. However, this study mainly focused on the attainable and the actual yield of coffee, which are both necessary for the estimation of yield losses. The model was built for two years to take into account primary and secondary yield losses, and the biennial production cycle of coffee. We assumed that each year had particular production situations and management strategies, but also that some components of one year may influence components of the next one (Fig. 5.1). Based on this model, we tested associations between different components (typologies and their profiles, as explained later), within the same year and from one year to another, with the aim to identify and explain which ones are significantly leading to specific injury profiles and primary and secondary coffee yield losses. This model is quite complete and reflects many associations, in this study we analyzed only the associations with respect to injury profiles, according to the objectives stated.

2.2 Sampling strategy and location of the coffee plot network

To gather a gradient of management strategies and production situations we selected a network of coffee plots with contrasting types of shade, cropping practices and altitudes. To avoid undesirable effects caused by other factors (components) different from those considered in the conceptual model (Fig. 5.1), we ensured that all the selected coffee plots belonged to smallholder farmers, had coffee plants (*Coffea arabica* L.) of the variety *Caturra* as the most common in the plot, and were located on soils of the same order (Inceptisols). Following this strategy, we established a coffee plot network in the canton of Turrialba, Costa Rica, where coffee is grown from 600 to 1400 m.a.s.l. This area is catalogued as a premontane wet forest life zone (mean annual rainfall = 2781 mm and mean annual temperature = 22.2°C; 10 year averages); the climate is rainy most of the year, but a most rainy period is considered from May to July, and a slightly dry period is considered from August to October. We worked in 61 coffee plots of this network where measurements were taken during two years (2014-2015). More details on the sampling strategy, experimental design and measurement of different variables (described in the next subsection) can be found in a previous study which took place in the same coffee plot network (Cerda *et al.*, 2017).

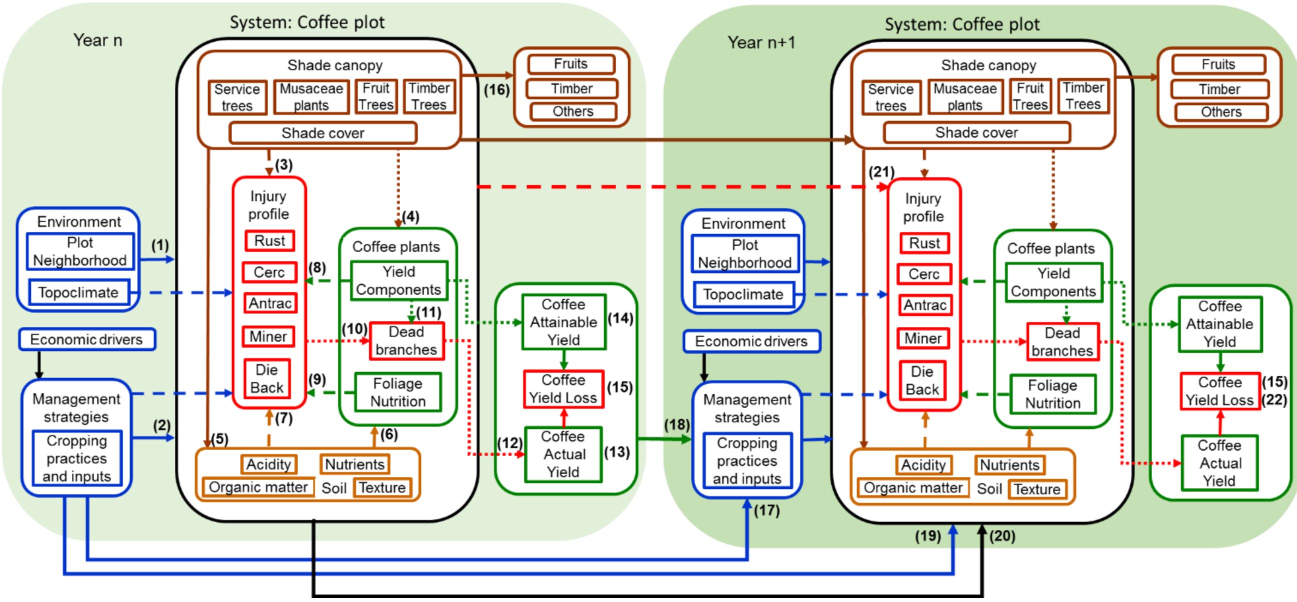


Fig. 5.1. Conceptual model describing the associations between components of production situations and management strategies with injury profiles leading to coffee yield losses

Arrows with dashed lines represent the associations analyzed through multivariate analysis in this study. Arrows with dotted lines represent effects tested with linear mixed models to estimate yields also analyzed in this study. Arrows with continued lines represents associations that certainly exist but were not analyzed in this study.

Within each year: (1) The environment (topoclimate) is considered to be associated with all the components of the system, since altitude, rainfall and temperature affect the growth of coffee plants and of other plants/trees in the shade canopy, the development of pathogens and insects, and the processes in soil that modify its chemical properties. (2) Management strategies can be associated with all the components of the system too, e.g. practices like application of fertilizers and control of weeds affect soil chemical properties and soil cover; pruning of coffee plants affects the plant architecture and yield components; practices applied to the plants/trees of the shade canopy can affect its structure and composition, and its shade cover; application of pesticides affects the level of pests and diseases injuries, and therefore the injury profile.

Within the system, shade canopy is associated with the other three components: the shade cover modifies the light interception and the microclimate, and thus affects (3) The development of pests and diseases (injury profile), and (4) The growth of coffee plants; (5) Shade cover, leaf litter and roots of trees affect soil chemical properties by influencing nutrient cycles. Soil is associated with both the coffee plants and the injury profile: soil fertility affects the (6) Growth/nutrition of coffee plants and thus, indirectly affects (7) Injury profiles also. The physiological characteristics of coffee plants are associated with injury profiles, since (8) Coffee yield components e.g. high fruit loads can make the plant more susceptible to pathogens, and (9) Certain leaf nutrient contents (foliage nutrition) can affect the attack of pests and diseases. (10) The injury profile can cause severe defoliations and/or lead to the death of branches. (11) Yield components can also cause the death of branches, i.e. high fruit loads could cause the exhaustion of plant tissues. (12) Defoliated branches (reduction of the photosynthetic capacity) and/or dead branches will cause the drying and death of the fruits that were growing on them. (13) The actual yield, then, as an output of the system, depends on and is associated with yield components, injury profiles and dead productive branches. (14) The attainable yield would be the output of the system if there were no influences of pests and diseases or dead branches, i.e., reducing factors = 0. (15) Yield loss for a specific year (primary yield loss) is the difference between the attainable yield and the actual yield. (16) Plants and trees of the shade canopy can also produce agroforestry products as other outputs of the system.

Across years: (17) management strategies of one year are considered to be associated with the management strategies of the next one, considering that some farmers can maintain or only slightly modify their cropping practices every year. (18) The outputs of the system of one year could be associated with the management strategies of the next year, since farmers could modify cropping practices according to the previous yields and yield losses. (19) Management strategies could also be associated with components of the system the next year, especially when cropping practices were strong enough (e.g. high amounts of pesticides or severe pruning) to modify drastically some of them. (20) It is also considered that all the components of the system of one year can be associated with their pairs in the next year. Therefore, (21) components of the previous year could be associated with the injury profile of the next one. (22) Secondary yield loss is the yield lost due to effects of reducing factors (injury profile) in the previous year, i.e. the yield that is not produced by the branches that died in the previous year. This means that in a given year, both primary and secondary yield losses should be considered.

2.3 Measurement and calculation of the studied variables

We demarcated an experimental subplot in each coffee plot, composed of eight rows with 15 coffee plants each. Eight coffee plants (one per row) were identified for sampling soil, and three branches on each chosen coffee plant were marked for measurements related to pests and diseases and coffee yields. We also demarcated a circular area of 1000 m^2 (17.8 m radius) from the center of the experimental subplot, for measurements related to the shade canopy. The variables used in this study are listed in Table 5.1, along with the methods for measurements and calculations.

2.4 Modeling and estimation of coffee yields and yield losses

Based on the measurement of coffee yield and plant productive characteristics (Table 5.1), we developed models and equations to estimate yields and yield losses. We used linear mixed models to estimate coffee actual yields as a function of fruiting nodes (yield component) and dead productive branches (yield reducing factor) from the current year and from the previous one (Cerda *et al.*, 2016, Chapter 4). In this study, given that we had agroforestry systems, we added the mean annual shade cover as another predictor of coffee yields.

For the year 2014, we estimated only primary yield losses, as no data were available for 2013, and for year 2015 we estimated both primary and secondary yield losses. Models and equations for these estimations are shown in Table 5.2. For 2014, we estimated primary yield losses based on a specific attainable yield of that year. For 2015, we estimated both primary and secondary yield losses, but based on an attainable yield that considers no yield reductions from at least two years (2014 and 2015). The equations along with indicators of the quality of their prediction are presented in a supplementary material of this manuscript.

Table 5.1. List of variables per typology, and methods used for measurements during two years (2014-2015) in the coffee plot network, Turrialba, Costa Rica

Typology: Variables	Methods/Formulas	Times of measurements (each year)
Injury profiles:		
sAUDPC (%) of: Coffee leaf rust Brown eye spot Anthracnose Coffee leaf miner	$AUDPC = \sum_{i=1}^{n-1} \frac{I_i + I_{i+1}}{2} \times (t_{i+1} - t_i)$ $sAUDPC = \frac{AUDPC}{Nd}$ where *AUDPC*: area under the disease progress curve; I_i: incidence of a given pest or disease at the *i*th measurement; t_i: time (in days) of the *i*th measurement; n: total number of measurements; *sAUDPC*: standardized AUDPC; *Nd*: total number of days (from the first to the last measurement). Source: (Simko and Piepho, 2012)	Incidences, severity and dieback index were measured in several key periods of coffee plants development: 1st Fruit formation; 2nd Beginning of fruit ripening; 3rd Just before the harvest; 4th During the peak of harvest; 5th End of coffee harvest period
sAUDPC (%) of severity	A scale from 0 to 6 (the higher the number, the higher the severity), based on the size and number of symptoms on leaves, was used to assign a level of severity to marked branches. Averages of severities were calculated per plant, and then sAUDPC (see above) of severity were calculated	
Maximum Dieback Index	Each plant of the subplot was classified in a scale (1-4): 1: plant with few defoliated branches; 2: several defoliated and already dead branches; 3: a lot of defoliated and already dead branches, and with withered dry fruits; 4: almost all dead branches, almost dead plants. Dieback index were calculated with the formula below (where N: number of plants registered in each scale); and the maximum index during the year was identified $Dieback\ index = \frac{1xN_1 + 2xN_2 + 3xN_3 + 4xN_4}{4xNtotal} \times 100$	
Dead productive branches (Number plant^{-1})	The number of dead productive branches in the main stem and in whole plant of the marked coffee plants was counted	End of the coffee harvest period
Coffee plants productive characteristics:		
Age of plants (years) Number of: Productive stems Productive branches Fruiting nodes Fruits per node	The age of coffee plants was asked to the farmer We quantified: Number of productive orthotropic stems per plant Number of productive branches per plant Number of fruiting nodes per plant Number of fruits per node each 25 fruiting nodes; then averaged	All these variables were quantified before the beginning of the harvest season, when small fruits were already visible
Nutrient deficiencies:		
Maximum deficiencies (%) of: N, P, K, Ca, Fe, Zn, Mg in coffee plants	The number of coffee plants with observable deficiency symptoms of each nutrient was counted in the subplot; then, percentages of plants with deficiencies were calculated; and the maximum percentages during the year were identified	These measurements were done in the same periods for sAUDPC measurements (see above)
Nutrients in leaves:		
N, P, K, Ca, C and Mg (%); Fe, Mg, Zn (mg kg^{-1})	Subsamples of leaves were taken from four plants in each coffee row of the subplot. Then a composite sample of leaves was obtained and sent to a laboratory for chemical analysis	Samples were obtained during the peak of harvest. Only in 2014
Topoclimate:		
Altitude (m.a.s.l.) Rainfall (mm)	Altitude was measured in the center of each coffee plot with a GPS device Rain gauges were installed in the communities were coffee plots were located. Data were registered daily and rainfall was divided in periods (next column)	Rainfall was quantified for the slightly dry periods and most rainy periods (see section 2.2)
Type of shade:		
Densities (plant ha^{-1}) Basal areas (m^2 ha^{-1}) Species richness of: Musaceae plants, and service, fruit and timber trees	In the circular area of 1000 m^2, all plants >2.5 m in height were identified. The trunk diameters of service and timber trees, and Musaceae stems were measured at 1.3 m from ground (breast height); fruit tree diameters were measured at 0.30 m. Densities and trunk basal areas were calculated per type of plant and per hectare.	These measurements were done during the dry period
Shade cover (%)	Shade cover was measured with a spherical densitometer in the four corners and in the center of the subplot, and then averaged. This variable was measured in four key times per year (next column) and an average of shade cover for all the year was (Lemmon, 1957)	Measurements in the most dry period, most rainy period, slightly dry period and slightly rainy period
Soil fertility:		
pH; soil acidity, P, K, Ca, Mg, Zn, Fe, Mn, (mg kg^{-1}); C and N (%); Sand, clay, silt (%)	Eight subsamples of soil at a depth of 0-20 cm were taken near the trunk of the eight marked coffee plants (at 50cm approximately) in each experimental subplot. The subsamples were mixed to obtain a composite sample, and sent to a soil laboratory for the chemical and texture analyses. Source: (Briceño and Pacheco, 1984)	Samples were obtained during the peak of harvest. Only in 2014
Management (Cropping practices):		
Distances between coffee rows and between plants (cm)	Distances were measured from each of the eight coffee plants to the neighboring coffee trees in the same row and between rows, and then averaged.	Measurements with the establishment of the coffee network
Number of: pruning, harvests, weeding, and applications of fertilizer, herbicide, fungicide	Data on how many times each cropping practice was applied to the coffee plot during the year were obtained through semi structured interviews with farmers	Farmers were asked, at least three times during the year, on cropping practices they were applying
Coffee yields and yield losses:		
Coffee actual yield (kg ha^{-1}) of ripe fresh coffee cherries Primary yield losses (%) Secondary yield losses (%)	We harvested coffee cherries of marked plants of 20 subplots, and based on these data we developed models to estimate yields and yield losses in the other 41 coffee plots (see section 2.4). Eight harvested subplots were under diversified shade canopies, eight under simple shade canopies and four in full sun, to consider the influence of shade on yields. Ten of these subplots were protected with fungicides in order to have plants with almost no pests and diseases.	Coffee cherries were harvested every two weeks during the harvest season

Coffee leaf rust (*Hemileia vastatrix* Berkeley and Broome); brown eye spot (*Cercospora coffeicola* Berk. and Curtis); anthracnose *(Colletotrichum spp.)*; leaf miner (*Leucoptera coffeella* Guérin-Mèneville)

Table 5.2. Models and equations to estimate attainable yields actual yields, and primary and secondary yield losses of coffee

Explanations	Models and equations	Eq.
(Model for 2014)	$Y_{(14)} \sim NFN_{(14)} + DeadB_{(14)} + ShadeC_{(14)} + (1 \mid \text{Plot})$	
Based on the results of the Model for 2014, we obtained an equation to estimate the actual yield of 2014 ($Y_{(14)}$)	$Y_{(14)} = I + a \times NFN_{(14)} + b \times DeadB_{(14)} + \text{c} \times ShadeC_{(14)}$	1
By setting the dead branches to “zero”, we estimated the specific attainable yield of 2014 ($Ya_{(14)}$)	$Ya_{(14)} = I + a \times NFN_{(14)} + \text{c} \times ShadeC_{(14)}$	2
We estimated the primary yield loss as a percentage of the specific attainable yield of 2014	$\%PYL_{(14)} = \frac{Ya_{(14)} - PYL_{(14)}}{Ya_{(14)}} \times 100$	3
(Model for 2015-1)	$Y_{(15)} \sim NFN_{(15)} + DeadB_{(15)} + ShadeC_{(15)} + (1 \mid \text{Plot})$	
Based on the results of the Model for 2015-1, we obtained an equation to estimate the actual yield of 2015 ($Y_{(15)}$)	$Y_{(15)} = I + a \times NFN_{(15)} + b \times DeadB_{(15)} + \text{c} \times ShadeC_{(15)}$	4
By setting the dead branches as “zero”, we estimated the specific attainable yield of 2015 ($Ya_{(15)}$)	$Ya_{(15)} = I + a \times NFN_{(15)} + \text{c} \times ShadeC_{(15)}$	5
We estimated the primary yield loss of 2015 in weight	$PYL_{(15)} = Ya_{(15)} - Y_{(15)}$	6
(Model for 2015-2)	$NFN_{(15)} \sim DeadB_{(14)} + ShadeC_{(14)} + (1 \mid \text{Plot})$	
Based on the results of the Model for 2015-2, we obtained an equation to estimate the actual fruiting nodes of 2015 ($NFN_{(15)}$)	$NFN_{(15)} = I + a \times DeadB_{(14)} + \text{b} \times ShadeC_{(14)}$	7
By setting the dead branches to “zero”, we estimated the fruiting nodes that would have been attainable ($NFNa_{(15)}$) without reducing factors of 2014	$NFNa_{(15)} = I + \text{b} \times ShadeC_{(14)}$	8
Then, we estimated the loss of fruiting nodes for 2015 ($NFNl_{(15)}$)	$NFNl_{(15)} = NFNa_{(15)} - NFN_{(15)}$	9
Thereby, we considered that estimating a yield with this $NFNl_{(15)}$ using the equation (5) represented the secondary yield losses	$SYL_{(15)} = I + a \times NFNl_{(15)} + \text{c} \times ShadeC_{(15)}$	10
To estimate an attainable yield without negative impacts of 2014 and 2015 ($Ya_{(14-15)}$), we replaced equation 7 in equation (4) and then set as zero the dead branches of both years to assume that there were no reducing factors	$Ya_{(14-15)} = I + a \times I + a \times \text{b} \times ShadeC_{(14)} + \text{c} \times ShadeC_{(15)}$	11
We calculated the primary yield loss of 2015 $\%PYL_{(15)}$ as a percentage of the $Ya_{(14-15)}$	$\%PYL_{(15)} = \frac{PYL_{(15)}}{Ya_{(14-15)}} \times 100$	12
We calculated the secondary yield loss of 2015 $\%SYL_{(15)}$ as a percentage of the $Ya_{(14-15)}$	$\%SYL_{(15)} = \frac{SYL_{(15)}}{Ya_{(14-15)}} \times 100$	13

Y: actual coffee yield per plant; NFN: number of fruiting nodes per plant; DeadB: number of dead productive branches per plant; ShadeC: mean annual shade cover; $_{(14)}$: represents the variables in the year 2014; $_{(15)}$: represents the variables in the year 2015; I: represents the intercepts in the equations; a, b, c represent the coefficients in the equations

2.5 Construction of typologies and description of the resulting profiles

We chose to use an analytical approach based on the building of typologies. This approach has already been successfully used in previous studies with large dataset describing production situations and crop management, whose goal was to identify pest and disease drivers (Savary *et al.*, 1995; Savary *et al.*, 2000b; Avelino *et al.*, 2006; Allinne *et al.*, 2016).

We constructed one typology (a group of profiles) for each group of variables mentioned in Table 5.1. The typology of injury profiles represented the set of the injuries caused by the most important pests and diseases; the typologies of coffee plant productive characteristics, nutrient deficiencies and nutrient in leaves represented the physiological characteristics of coffee plants; the typology of management represented the management strategies; and the typologies of topoclimate, type of shade and soil fertility represented the production situations. We also determined categories of attainable yield, actual yield, and primary and secondary yield loss.

All of these typologies were constructed for two years separately (2014 and 2015), because since several factors changed from one year to another (e.g. rainfall, cropping practices, and others), we considered that each year had particular management strategies and production situations.

For each typology, we described the profiles obtained based on the means of the variables used for their clustering. Although most typologies were constructed taking into account many variables, we described their profiles based on maximum five variables which showed the most contrasting means among profiles. Significant differences of each variable among profiles were assessed with the Kruskal-Wallis non-parametric test ($p<0.05$), given that several of the variables did not follow a normal distribution.

2.5.1 Typologies of injury profiles

The construction of these typologies followed three steps: i) we ran principal component analysis (PCA) with the standardized areas under the progress curve of the main pests and diseases and the maximum dieback index of the coffee plots; ii) we identified the components which represented more than 85% of the cumulative variability from the PCA; iii) we performed a hierarchical clustering with those components (Euclidian distance; Ward method) to determine the number of profiles.

2.5.2 Typologies of physiological characteristics of coffee plants, production situations, and management strategies

These typologies were constructed by maximizing their relationships with the injury profile as proposed by Avelino *et al.*, (2006) in order to avoid the loss of information related with the studied pests and diseases. We followed three steps to construct each typology: i) we ran partial least square discriminant analysis (PLS-DA) by putting the variables of a given typology in relation to the already constructed injury profiles; ii) we identified the first two components resulting from the PLS-DA; iii) we performed a hierarchical clustering with those components (Euclidian distance; Ward method) to determine the number of profiles within the typology.

2.5.3 Categorization of yields and yield losses

We performed a frequency distribution of the estimated yields and yield losses (primary and secondary) per plot, and divided each of them in three groups composed by the same number of coffee plots. That way, we determined three categories for each attainable yield, actual yield, primary yield loss and secondary yield loss.

2.6 Associations between profiles of typologies

We used a descriptive analysis whose basis has been developed by Savary and collaborators (1995, 2000) and applied with success to coffee pests and diseases (Avelino *et al.*, 2006; Allinne *et al.*, 2016). The finality of the analysis is to graphically represent the significant relationships between different typologies. To test the significance of the associations between the typology of injury profiles and each of the other typologies we used the Fisher's exact test ($p<0.05$). Then, using a contingency table with the injury profiles and the profiles of yields and yield losses in columns, and all the profiles of the other typologies in rows, we ran a simple correspondence analysis in order to represent graphically the associations among different profiles. In the correspondence analysis, the attainable yields, actual yields and yield losses were declared as additional variables; i.e. the axes of the correspondence analysis were derived only from the inertia of injury profiles and the profiles located in the rows of the contingency table, and yields and yield losses were just super-imposed on this system of axes. In order to highlight groups of profiles most linked among each other, we performed a hierarchical clustering with the coordinates of the profiles in the two first axes resulting from the correspondence analysis (Allinne *et al.*, 2016). We performed the Fisher's exact test and correspondence analysis for the profiles of each of the two years separately (2014 and 2015), and for the injury profiles, and yields and yield losses of 2015 in relation with profiles of 2014, with the aim to explain associations from one year to another.

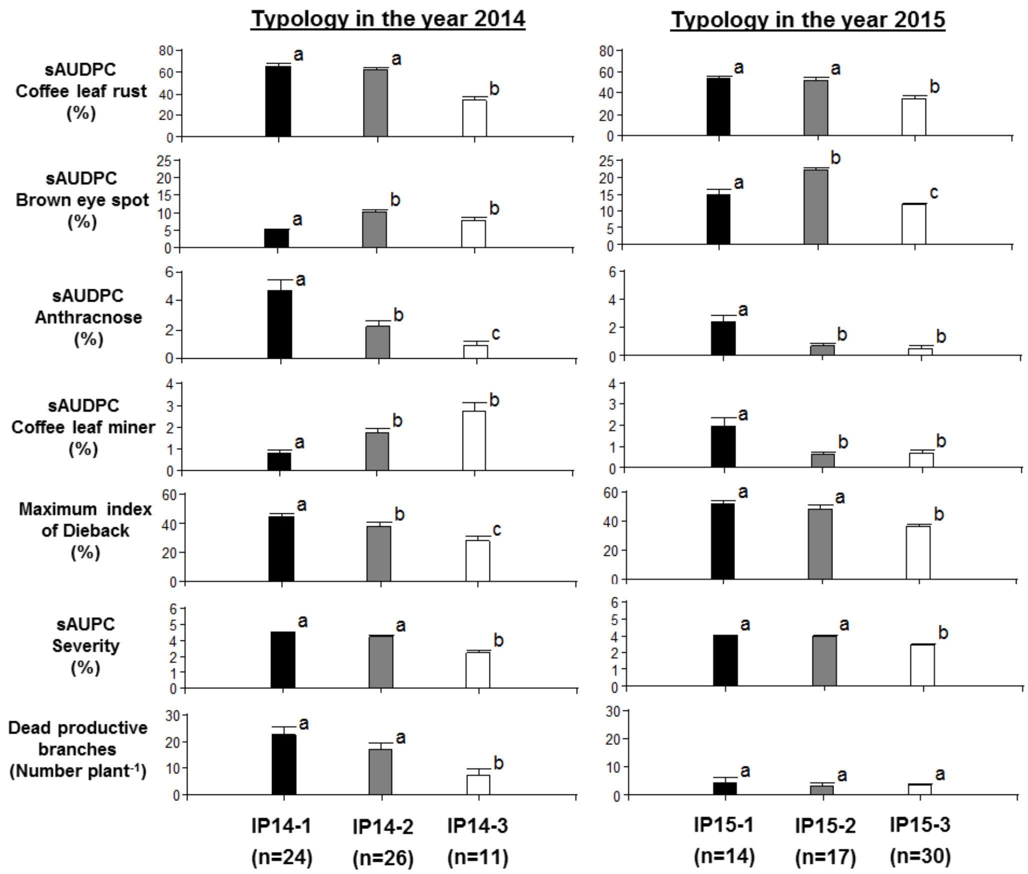
All statistical analyses were carried out with the program R (R_Core_Team, 2014), with packages lme4 (Bates *et al.*, 2015) and FactoMineR (Husson *et al.*, 2012).

3. Results

3.1 Description of typologies and their resulting profiles

3.1.1 Injury profiles

Three injury profiles (IP) were identified in each year. Coffee leaf rust and brow eye spot were the most important diseases according to their high standardized area under the disease progress curve (sAUDPC), as well as the maximum index of dieback which attained high values; while anthracnose and leaf miner presented sAUDPC lower than 5%. In 2014: IP14-1 was characterized by high levels of coffee leaf rust and dieback, resulting in high severity values and a high number of dead productive branches; IP14-2 had similar levels of coffee leaf rust as IP14-1, but had lower levels of dieback and number of dead productive branches; IP14-3 showed the lowest levels of the most important diseases, as well as the lowest severity and number of dead productive branches. In 2015: IP15-1 and IP15-2 were similar in coffee leaf rust, dieback and severity, but IP15-2 had highest levels of brown eye spot; while IP15-3 was considered as the one with the lowest injury levels. There were no differences in dead productive branches among profiles in 2015 (Fig. 5.2).



sAUDPC: standardized area under the disease progress curve

Different lowercase letters between bars indicate significant differences determined by Kruskal-Wallis non-parametric test ($p<0.05$)

Fig. 5.2. Description of the typologies of injury profiles obtained in 2014 and 2015. Typologies in 2014 and in 2015 were built independently.

3.1.2 Physiological characteristics of coffee plants

In the typology of coffee productive characteristics (PC), three profiles were identified in each year. In 2014: PC14-2 had the highest number of fruiting nodes and productive branches; PC14-1 was considered the second in productive characteristics because it had similar characteristics as PC14-2 but a lower number of fruiting nodes; while PC14-3 had the lowest productive characteristics in general. In 2015: PC15-3 had the highest values in all productive characteristics; PC15-2 was considered as the second; and PC15-1 had the lowest productive characteristics, and the highest age. In 2014 there were noticeably higher productive characteristics than in 2015 in general (Fig. 5.3A).

In the typologies of nutrient deficiencies (De), it was possible to differentiate some profiles with deficiencies of macro nutrients and other of micro nutrients. In 2014 there were two profiles: De14-1 had lower deficiencies of N, P, Ca and Mg than De14-2. In 2015 there were three profiles: De15-3 had the highest deficiencies overall; De15-2 had the lowest deficiencies of Zn and Mg; and De15-1 had the lowest deficiencies of N (Fig. 5.3B).

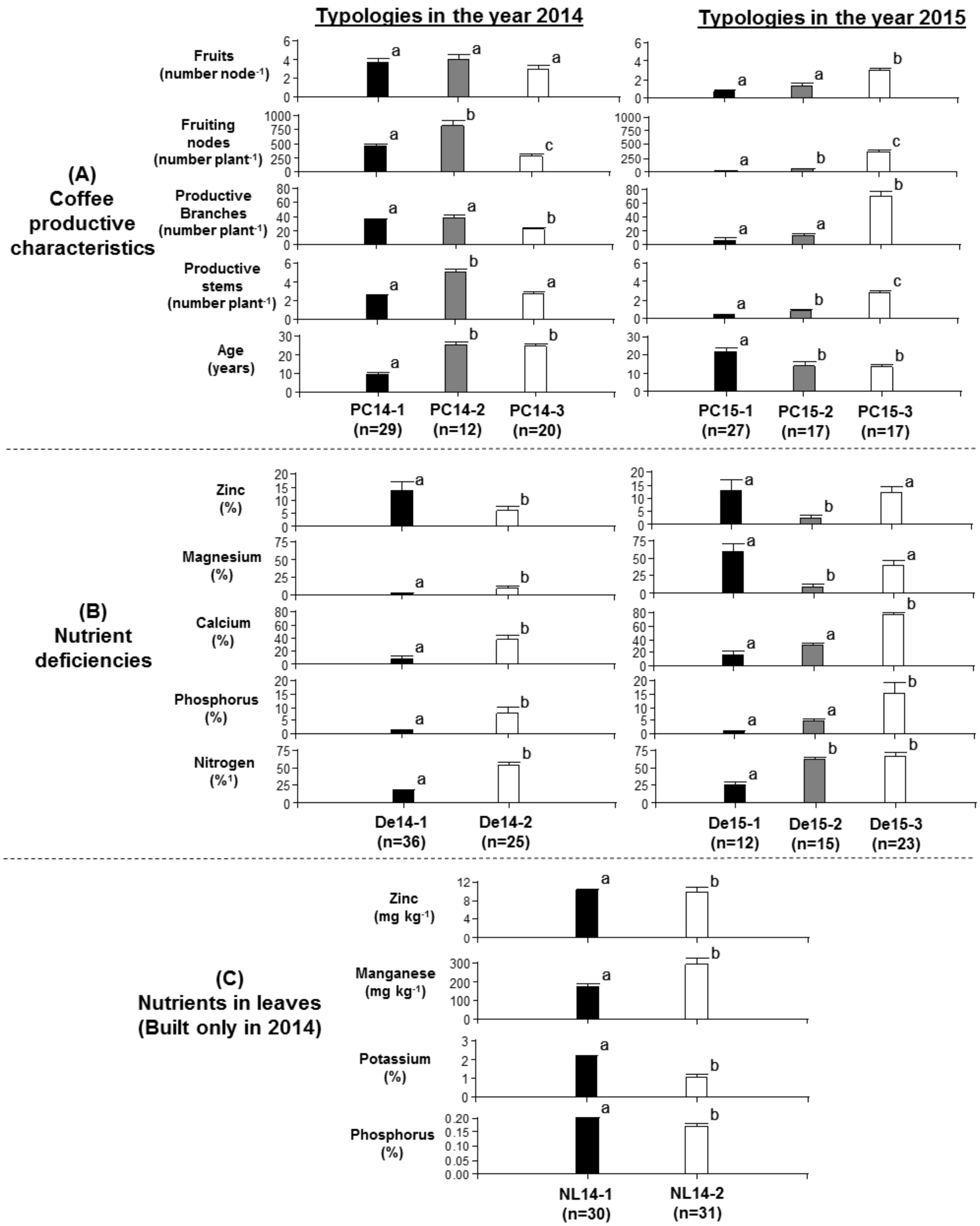
In the typology of nutrient in leaves (NL), two profiles with contrasting characteristics especially in macro nutrients were defined in 2014. The profile NL14-1 had higher P, K and Zinc but lower Mn than the other profile NL14-2 (Fig. 5.3C).

3.1.3 Production situations

There were three profiles in the typology of topoclimate, in 2014: TC14-1 had the lowest altitude and the lowest rainfall in the most rainy period; while TC14-2 and TC14-3 had similar altitudes but TC14-3 had the highest rainfall in the most rainy period and the lowest rainfall in the slightly rainy period. In 2015, we obtained only two profiles with TC15-1 having lower altitude and higher rainfall in the slightly rainy period than TC15-2 (Fig. 5.4A).

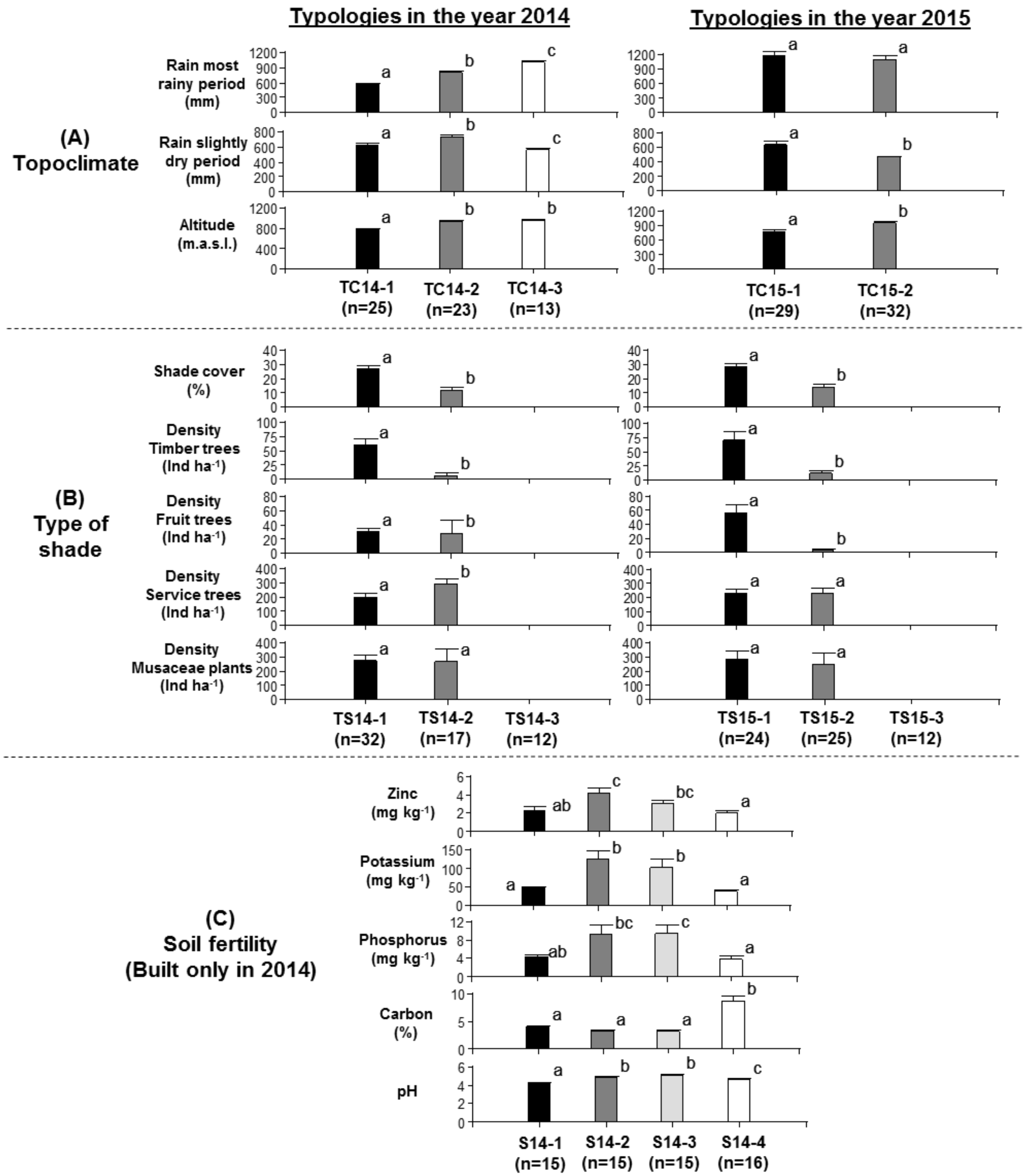
In typologies of type of shade, three profiles were found each year, representing well defined agroecosystems. In 2014, TS14-1 had high densities of timber and fruit trees, and the highest shade cover representing a highly diversified agroforestry system; TS14-2 had few timber and fruit trees but more services trees and lower shade cover representing a simple agroforestry system; and TS14-3 represented a coffee monoculture at full sun exposure. In 2015, the same description applied: TS15-1 represented a highly diversified agroforestry system; TS15-2 represented a simple agroforestry system; TS15-3 represented a coffee monoculture at full sun (Fig. 5.4B).

In the typology of soil, there were four profiles in 2014: S14-1 had the lowest pH; S14-4 had the highest content of C (organic matter); while S14-2 and S14-3 were quite similar between each other, both were characterized for having higher contents of P, K and Zn than the other two profiles (Fig. 5.4C).



Different lowercase letters between bars indicate significant differences determined by Kruskal-Wallis non-parametric test ($p<0.05$)

Fig. 5.3. Description of the typologies and profiles representing the physiological characteristics of coffee plants: (A) Coffee productive characteristics; (B) Nutrient deficiencies; and (C) Nutrient in leaves, obtained in 2014 and 2015. Typologies in 2014 and in 2015 were built independently.

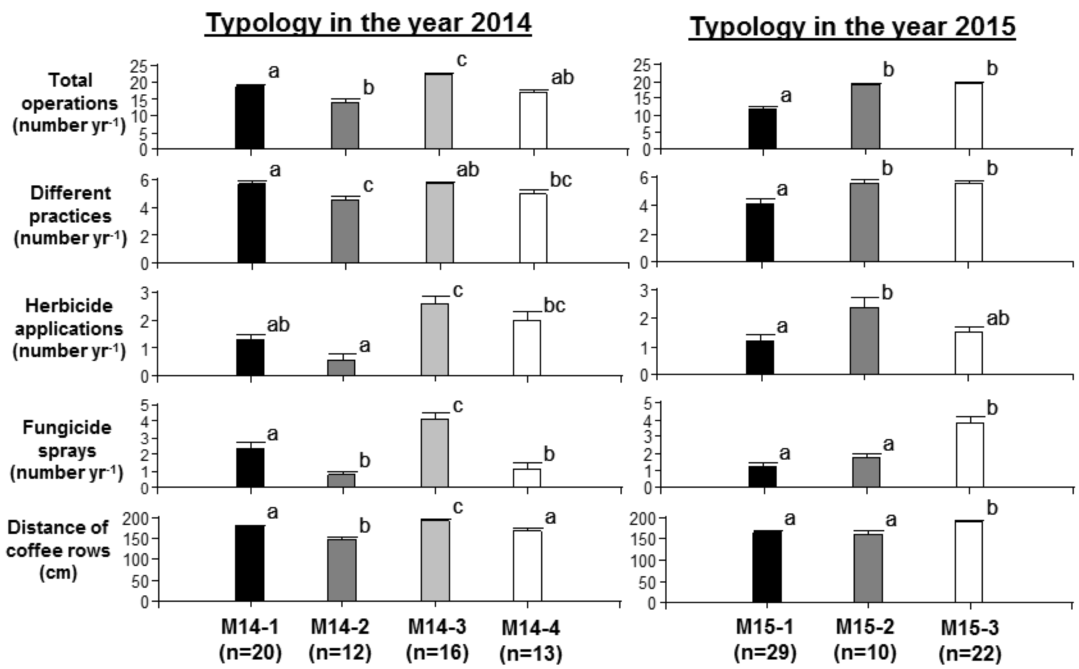


Different lowercase letters between bars indicate significant differences determined by Kruskal-Wallis non-parametric test ($p<0.05$)

Fig. 5.4. Description of the typologies and profiles representing production situations (A) Topoclimate; (B) Type of shade; and (C) Soil fertility, obtained in 2014 and 2015. Typologies in 2014 and in 2015 were built independently.

3.1.4 Management strategies

In typologies of management, there were profiles with more cropping practices than others mainly based on the number of fungicide sprays, the diversity of cropping practices and total operations applied per year. In 2014, four profiles were found: M14-3 had the highest number of fungicide sprays, herbicide applications and total operations per year, as well as the highest distances between coffee rows; M14-1 could be considered as the second in the intensity of cropping practices, and M14-4 as the third one; while M14-2 were the last due to its lowest number of fungicide sprays and total operations, as well as the lowest distances between coffee rows. In 2015, M15-3 had the highest number of fungicide sprays, as well as the highest distances between coffee rows; M15-2 had similar number of total operations and herbicide applications than M15-3; while M15-1 had the lowest number of cropping practices in general (Fig. 5.5).

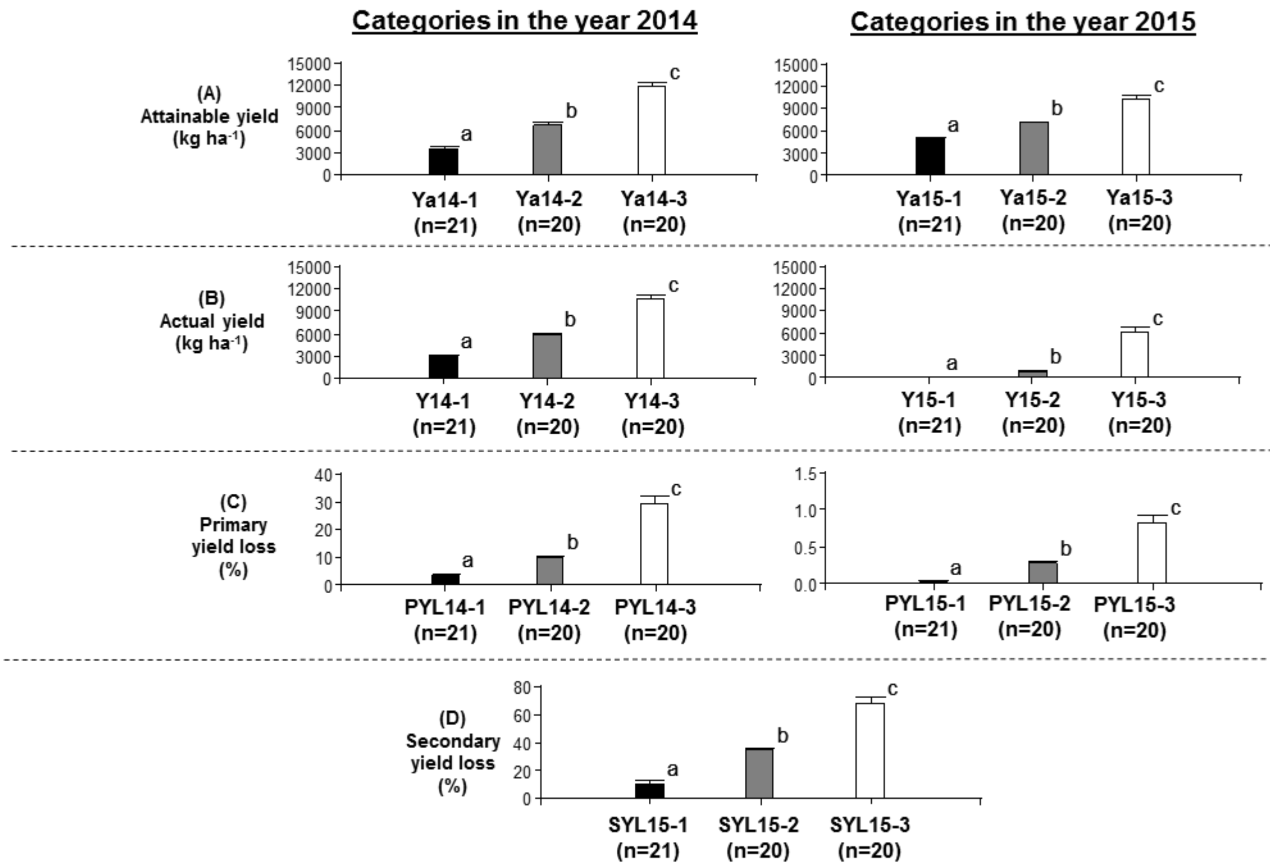


Different lowercase letters between bars indicate significant differences determined by Kruskal-Wallis non-parametric test ($p<0.05$)

Fig. 5.5. Description of the typologies and profiles of management strategies, obtained in 2014 and 2015. Typologies in 2014 and in 2015 were built independently.

3.1.5 Yields and yield losses of coffee

Each estimated attainable yield, actual yield, primary yield loss, and secondary yield loss had three well differentiated categories. Attainable yields and especially actual yields were higher in 2014 than in 2015 (Fig. 5.6A and 5.6B), reflecting the biennial production cycle of coffee. In 2014, the means of primary yield losses of the profiles ranged from 4 to 29%, while they did not surpass 1% in 2015 (Fig. 5.6C). The means of secondary yield losses ranged from 11 to 68% in 2015 (Fig. 5.6D), indicating that these losses after a year of high production year can be severe and noticeably higher than primary yield losses.



kg ha^{-1} are kg of fresh coffee cherries per hectare (using a factor 1/5.6, they can be transformed to green coffee)
Different lowercase letters between bars indicate significant differences determined by Kruskal-Wallis non-parametric test ($p<0.05$)

Fig. 5.6. Categories of (A) Attainable yield; (B) Actual yield; (C) Primary yield loss; and (D) Secondary yield loss, obtained in 2014 and 2015. Typologies in 2014 and in 2015 were built independently.

3.2 Associations between injury profiles and yields, yield losses, management strategies, production situations and physiological characteristics of coffee plants

3.2.1 Associations in 2014

In 2014, injury profiles were significantly associated ($P<0.05$) with primary yield losses, management strategies, physiological characteristics of coffee plants, and production situations, but not with the type of shade (Table 5.3). The correspondence analysis representing these associations produced two dimensions. The first dimension, where most of the profiles, and particularly injury profiles, were well represented, explained 84.88% of the inertia. Primary yield losses were negatively associated with this first dimension. Soil profiles S14-1 and S14-3 contributed more to the second dimension (15.12% of the inertia); and actual yields were positively associated with this dimension.

Cluster analysis on these two dimensions produced five groups of associated profiles (Fig. 5.7):

In the group G14-1, the injury profile characterized by the highest levels of injuries (IP14-1) was associated with the highest primary yield losses (PYL14-3). This group comprised plots located at the lowest altitudes and under the lowest rainfalls in the most rainy period (TC14-1), with the most suboptimal management (M14-2), on soils with the highest contents of P, K, and Zn, and lowest acidity (S14-2), planted with old and high yielding coffee plants (PC14-2); showing the lowest deficiencies of nutrients (De14-1), with the highest leaf contents of P, K and Zn, and the lowest of Mn and Mg (NL14-1).

In the group G14-2, the medium injury profile (IP14-2) was associated with the lowest attainable (Ya14-1) and actual yields (Y14-1). This group comprised plots with the second most suboptimal management (M14-4); on soils that had good contents of K and P (S14-3); and planted with old and very low yielding coffee plants (PC14-3).

The group G14-3 comprised plots with the lowest levels of injuries (IP14-3) and with the most intensive management, i.e. the highest number of fungicide sprays and herbicide applications, total practices, and the largest distance between coffee rows (M14-3). According to its position along the first axis, we can deduce that IP14-3 was related with the lowest values of primary yield losses.

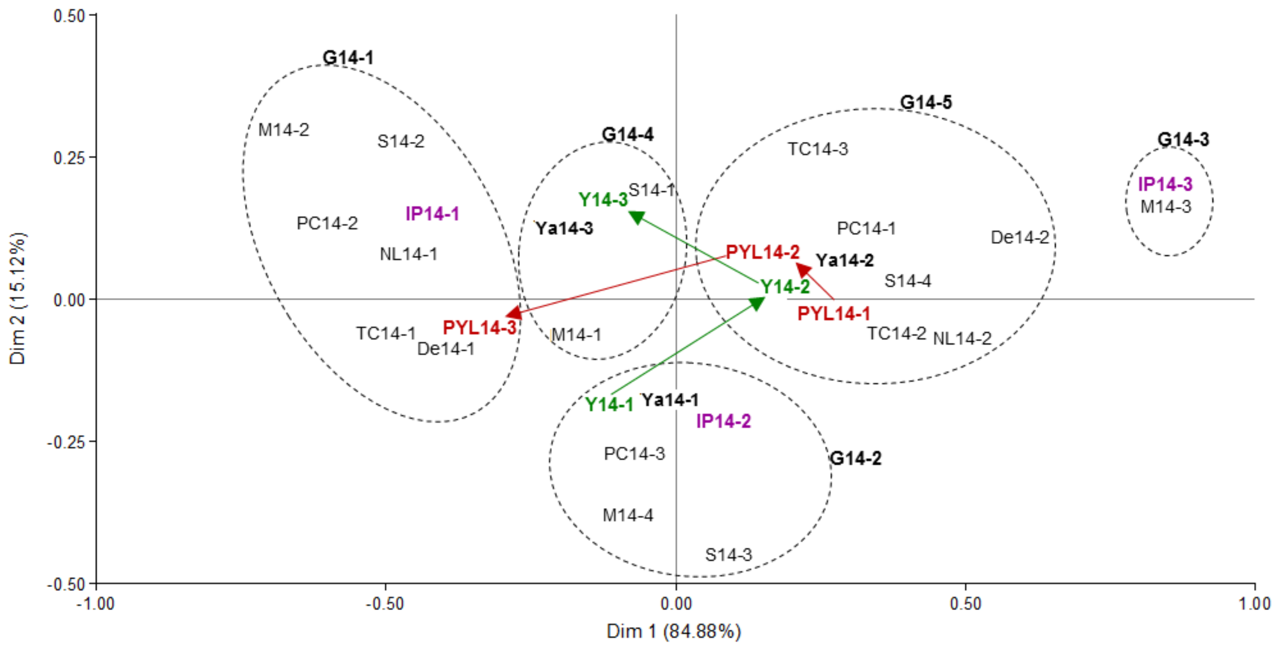
The other two groups (G14-4 and G14-5) did not involve any injury profile. The group G14-4 clustered the plots with the highest attainable (Ya14-3) and actual yields (Y14-3), located on soils with the lowest contents of P and K (S14-1), and under a management which can be considered the second in intensity (M14-1). Group G14-5 comprised the plots with the lowest primary yield losses (PYL14-1 and PYL14-2), medium attainable (Ya14-2) and actual yields (Y14-2), planted with young and medium yielding plants, showing severe deficiencies of macronutrients (PC14-1), and located at the highest altitudes (TC14-2 and TC14-3).

Table 5.3. Associations between injury profile and typologies of yields, yield losses, management strategies, production situations and physiological characteristics of coffee plants in 2014

	Y14	Ya14	PYL14	M14	TC14	TS14	De14	NL14	PC14	S14
IP14	NS	NS	*	**	***	NS	***	***	*	*

**p < 0.05; **p < 0.01; ***p < 0.001; NS: not significant*

IP: injury profiles; Y: categories of actual yields; Ya: categories of attainable yields; PYL: categories of primary yield losses; M: typology of management strategies; TC: typology of topoclimate; TS: typology of type of shade; De: typology of coffee plants nutrient deficiencies;; NL: typology of nutrient in leaves of coffee plants; PC: typology of coffee plants productive characteristics; S: typology of soil fertility; 14: represents the year 2014



IP14-X: injury profiles; Ya14-X: categories of attainable yields (estimated yields with no pests and diseases in 2014); Y14-X: categories of actual yields; PYL14-X: categories of primary yield losses; TC14-X: profiles of topoclimate; M14-X: profiles of management strategies; PC14-X: profiles of coffee plants productive characteristics; De14-X: profiles of coffee plants nutrient deficiencies; NL14-X: profiles of nutrient in leaves of coffee plants; S14-X: profiles of soil fertility. Dotted circles represent groups of associated profiles. See Figures 2-4 for characteristics of each profile. Arrows indicate increasing values of yields and yield losses.

Fig. 5.7. Graphical representations of the simple correspondence analysis of injury profiles, estimated yields and yield losses, physiological characteristics of coffee plants, management strategies and production situations in 2014.

3.2.2 Associations in 2015

In 2015, injury profiles were significantly associated ($P<0.05$) with actual yields and secondary yield losses, but not with primary yield losses. As for 2014, injury profiles had associations with most of the typologies, except with the type of shade and topoclimate (Table 5.4). The first dimension of the correspondence analysis explained 91.90% of inertia. Most of the profiles were well represented on this dimension. Actual yields and secondary yield losses were negatively and positively linked with this dimension respectively.

Three groups of associated profiles were identified through cluster analysis on these dimensions (Fig. 5.8):

In the group G15-1, the two injury profiles with the highest levels of injuries (IP15-1 and IP15-2) were associated with the highest secondary yield losses (SYL15-2 and SYL15-3), highest attainable yield (Ya15-3), and lowest actual yields (Y15-1 and Y15-2). This group comprised plots with the lowest distances between coffee rows and the least number of fungicide sprays (M15-1 and M15-2), planted with the oldest and lowest yielding plants (PC15-1), exhibiting the lowest deficiencies of P and K (De15-2).

The group G15-2, which did not include any of the injury profiles, involved the lowest attainable yields (Ya15-1 and Ya15-2) and the highest primary yield losses (PYL15-2 and PYL15-3). It comprised plots with low yielding coffee plants (PC15-2).

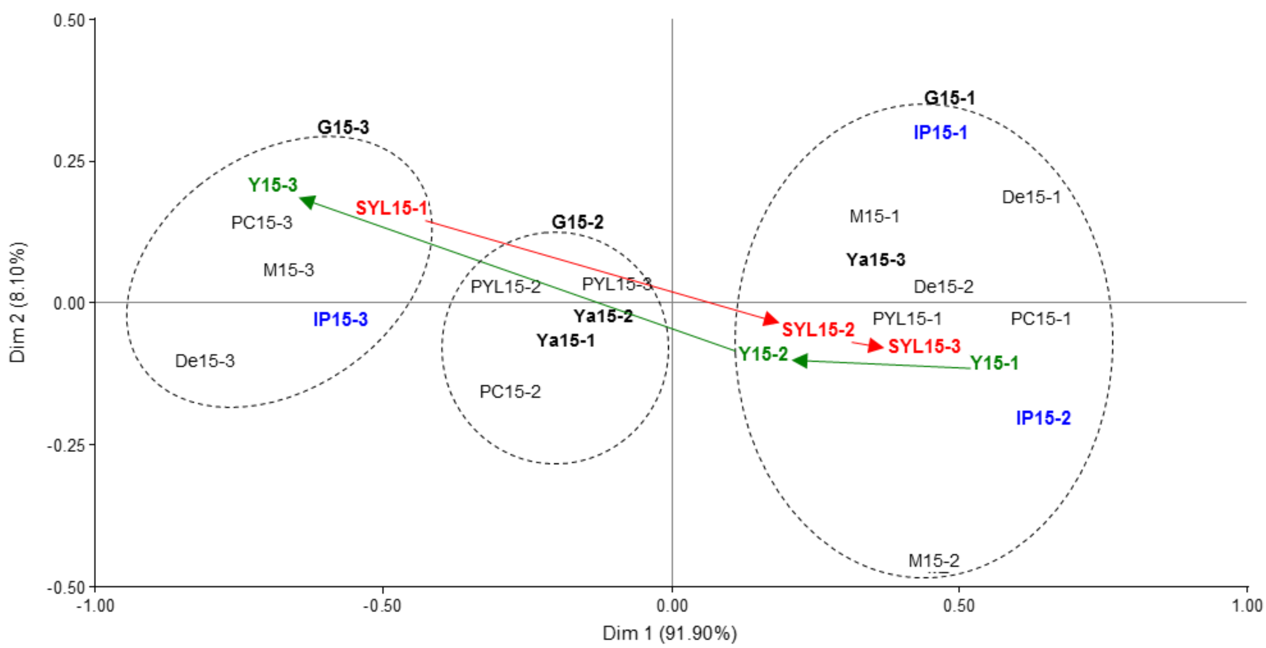
In the group G15-3, the injury profile with the lowest levels of injuries (IP15-3) was associated with the highest actual yields (Y15-3) and the lowest secondary yield losses (SYL15-1). This group clustered plots with the largest distances between coffee rows and the highest number of fungicide sprays (M15-3), planted with very high yielding coffee plants (PC15-3) showing severe deficiencies of nutrients, especially of P and Ca (De15-3).

Table 5.4. Associations between injury profile and typologies of yields, yield losses, management strategies, production situations and physiological characteristics of coffee plants in 2015

	Y15	Ya15	PYL15	SYL15	M15	TC15	TS15	De15	PC15
IP15	**	NS	NS	NS	***	NS	NS	***	***

**p < 0.05; **p < 0.01; ***p < 0.001; NS: not significant*

IP: injury profiles; Y: categories of actual yields; Ya: categories of attainable yields; PYL: categories of primary yield losses; SYL: categories of secondary yield losses; M: typology of management strategies; TC: typology of topoclimate; TS: typology of type of shade; De: typology of coffee plants nutrient deficiencies; PC: typology of coffee plants productive characteristics; 15: represents the year 2015



IP15-X: injury profiles; Ya15-X: categories of attainable yields (estimated yields with no pests and diseases in 2014 and 2015); Y15-X: categories of actual yields; PYL15-X: categories of primary yield losses; SYL15-X: categories of secondary yield losses; M15-X: profiles of management strategies; PC15-X: profiles of coffee plants productive characteristics; De15-X: profiles of coffee plants nutrient deficiencies. Dotted circles represent groups of associated profiles. See Figures 2-4 for characteristics of each profile. Arrows indicate increasing values of yields and yield losses.

Fig. 5.8. Graphical representations of the simple correspondence analysis of injury profiles, estimated yields and yield losses, physiological characteristics of coffee plants, management strategies and production situations in 2015

3.2.3 Associations of injury profiles of 2015 with profiles of 2014

Injury profiles of 2015 were significantly associated ($P<0.05$) with injury profiles, primary yield losses, management and nutrient deficiencies of 2014 (Table 5.5). The correspondence analysis representing these associations produced two dimensions. The first dimension, comprised 89.44% of the inertia, where injury profiles of 2014 were well represented; and actual yields and secondary yield losses of 2015 were negatively and positively linked with this dimension respectively.

Three groups of associated profiles were identified through cluster analysis on these dimensions (Fig. 5.9):

In the group G14,15-1, the two injury profiles with highest levels of injuries in 2015 (IP15-1 and IP15-2) were associated with a similar injury profile of 2014 (IP14-1), which together were associated with the lowest actual yields of 2015 (Y15-1 and Y15-2), highest secondary losses of 2015 (SYL15-3), and highest primary yield losses of 2014 (PYL14-3); as well as with suboptimal managements of 2014 (M14-2 and M14-4), and with lowest nutrient deficiencies in 2014 (De14-1).

The group G14,15-2 which did not include none of injury profiles of 2015, gathered the lowest and medium attainable yields of 2015 (Ya15-1 and Ya15-2), and medium secondary yield loss of 2015 (SYL15-2); as well as the most intensive management of 2014, and the medium injury profile of 2014 (IP14-2).

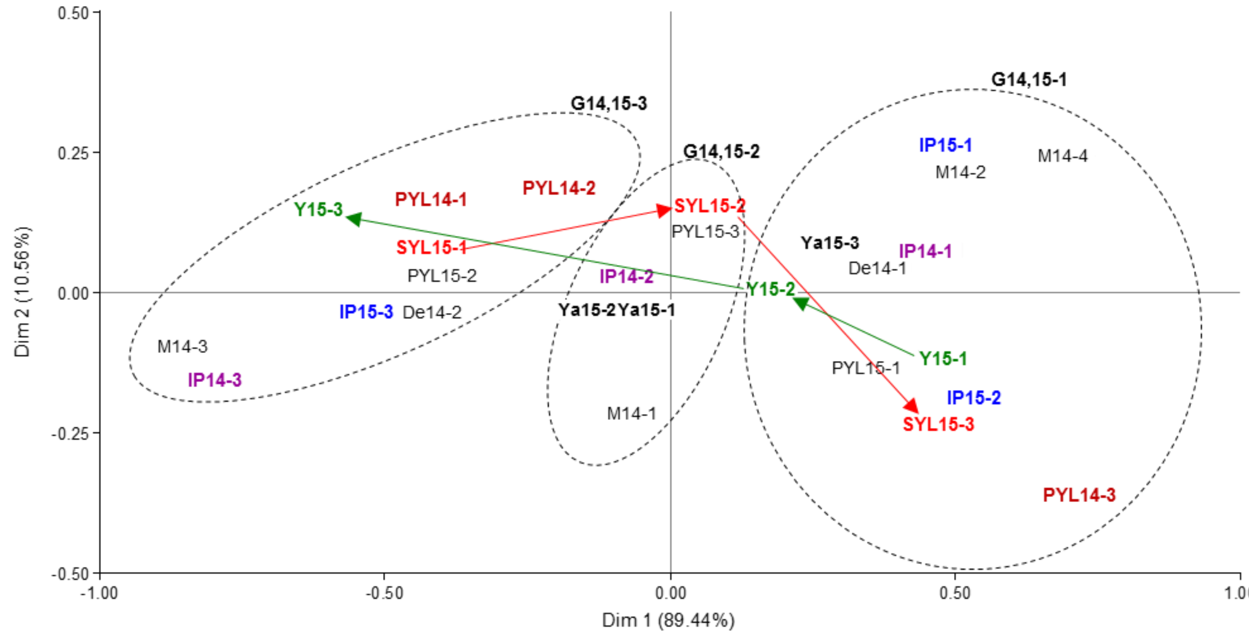
In the group G14,15-3, the injury profile with lowest injury levels in 2015 (IP15-3) was associated with a similar injury profile of 2014 (IP14-3), with the lowest primary yield losses in 2014 (PYL14-1) and lowest secondary yield losses in 2015 (SYL15-1), with the highest actual yield in 2015 (Y15-3), as well as with the most intensive management (M14-3) and the highest nutrient deficiencies in 2014 (De14-2).

Table 5.5. Fisher test between injury profile of 2015 and typologies of 2014

	IP14	Y14	Ya14	PYL14	M14	TC14	TS14	De14	S14	PC14	NL14
IP15	*	*	NS	***	***	NS	NS	*	NS	NS	NS
PYL15	***	NS	**	*	NS	NS	NS	*	*	*	NS
SYL15	**	NS	NS	***	NS	NS	NS	*	*	NS	*

**p < 0.05; **p < 0.01; ***p < 0.001; NS: not significant*

IP: injury profiles; Y: categories of actual yields; Ya: categories of attainable yields; PYL: categories of primary yield losses; SYL: categories of secondary yield losses; M: typology of management strategies; TC: typology of topoclimate; TS: typology of type of shade; De: typology of coffee plants nutrient deficiencies; S: typology of soil fertility; PC: typology of coffee plants productive characteristics; NL: typology of nutrient in leaves of coffee plants; 14: represents the year 2014; 15: represents the year 2015



IP15-X: injury profiles of 2015; Ya15-X: categories of attainable yields (estimated yields with no pests and diseases in 2014 and 2015); Y15-X: categories of actual yields in 2015; PYL15-X: categories of primary yield losses in 2015; SYL15-X: categories of secondary yield losses in 2015; IP14-X: injury profiles in 2014; PYL14-X: categories of primary yield losses in 2014; M14-X: profiles of management strategies in 2014; De14-X: profiles of coffee plants nutrient deficiencies in 2014. Dotted circles represent groups of associated profiles. See Figures 2-4 for characteristics of each profile. Arrows indicate increasing values of yields and yield losses.

Fig. 5.9. Graphical representations of the simple correspondence analysis of injury profiles, yields and yield losses of 2015 in association with profiles of 2014

4. Discussion

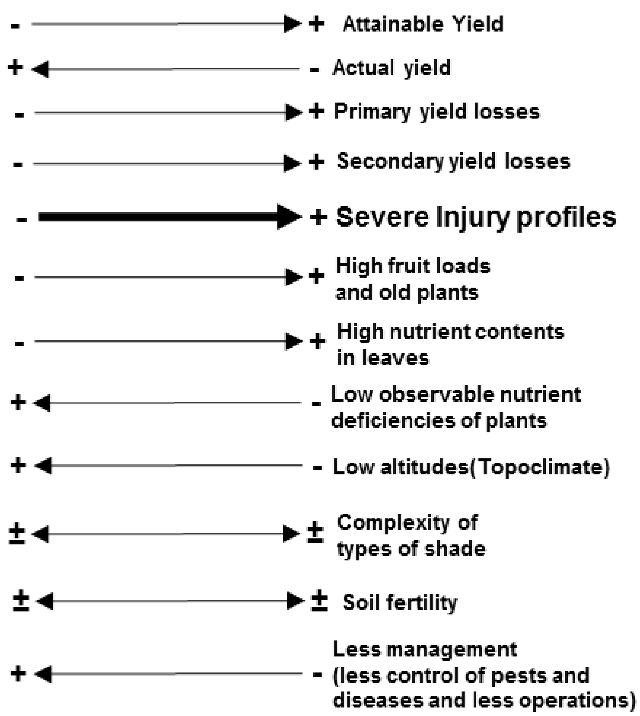
4.1 Primary and secondary yield losses depend on injury profiles, on management strategies, and on production situations

The main finding of this study was that gradients of yield losses (from low to high losses) were positively related to gradients of injury profiles (from slight to highly severe profiles), which depended on production situations, management strategies and physiological characteristics of coffee plants. The analytical approach used in this study enabled us to conclude on such finding, which is valid as long as associations between typologies are significant (test on contingency tables) and when correspondence analysis shows clear relationships among profiles (Savary *et al.*, 1995), as happened in our results.

The associations found in this study permit to explain how management strategies and production situations can affect yield losses through their influences on injury profiles. In Fig. 5.10 we present a summary, derived from results of the correspondence analyses, on the main relationships leading to severe injury profiles (high injury levels of pests and diseases) and then to high primary and secondary yield losses, and low actual yields. A high dependence of injury profiles on topoclimate, soil and management was documented in previous studies in rice (Savary *et al.*, 2000b) and also in coffee recently (Allinne *et al.*, 2016). With this study, we confirm the relevance of topoclimate and management, but we also highlight the importance of the physiological characteristics of coffee plants in relation with injury profiles.

Based on the patterns detected in the correspondence analyses, we can deduce that increasing fruit loads in old coffee plants, increasing contents of nutrients in coffee leaves, decreasing altitudes, and suboptimal management were the main drivers leading to severe injury profiles. Whereas, regarding the other two representatives of production situations (soil and types of shade): there were no gradients of soil fertility related to severe injury profiles; and types of shade were not associated with injury profiles (Fig. 5.10).

From the results that injury profiles were related to yield losses, and that injury profiles of the two years were not associated with types of shade, it can be deduced that types of shade were not associated with yield losses. The role of shade in regulating pests and diseases has been controversial. Some studies suggest that shade trees can hamper noxious pathogens by favoring their natural enemies or by modifying the microclimate of the system (Staver *et al.*, 2001), whereas other studies indicate that shaded conditions could favor the development of fungi and insects, particularly because some trees and plants could be their hosts (Avelino *et al.*, 2007; López-Bravo *et al.*, 2012). The findings of this study suggest that types of shade do not affect the development of injury profiles nor the reduction of yields (losses), in other words, good yields and low yield losses should be achieved independently of the type of shade. Similarly, previous studies demonstrated that shade is not good nor bad by itself, but interactions among shade, altitude and management must be considered to enhance the regulation of pests and diseases and the provision of other ecosystem services (Cerda *et al.*, 2017).



One-way arrows from (-) to (+) indicate the increase or decrease of a given variable with respect to the injury profiles. For instance, when the severity (high level injuries) of the injury profile increases, the primary and secondary yield losses also increases, or when the altitude decreases (toploclimate), the severity of the injury profile increases. Whereas two-way arrows with (±) in the extremes, indicate that the variable is not associated with the injury profiles. For instance, the severity of the injury profile can increase or decrease independently of the complexity of the type of shade

Fig. 5.10. Summary of the management strategies and production situations leading to severe injury profiles and then to primary and secondary yield losses

4.2 Primary and secondary losses are linked between each other, implying serious consequences

Secondary yield losses were worse than primary yield losses. A previous study, demonstrated similar results under experimental controlled conditions in a coffee parcel at full sun (see Cerda *et al*., Chapter 4). To our knowledge, this was the first study to quantify both primary and secondary yield losses in a perennial crop, and analyze their causes in the actual field conditions that smallholder farmers face. We contribute to the field of crop loss research with data indicating that mean primary yield losses in the worst cases can reach up 30%, and secondary losses up to 70%. This finding highlights the usefulness, in a perennial crop, to assess not only the losses occurring in a current year, but also the losses which may occur in later years (Zadoks and Schein, 1979; Avelino *et al.*, 2015). In the case of coffee, the severe secondary yield losses indicate that farmers should be helped with priority to recover the growth and vigor of their coffee plants, and thus recover yields faster.

Primary yield losses in a year of high production were positively associated with secondary yield losses given that they had similar drivers (similar injury profiles, management, and others), indicating that by assessing the first it is possible to deduce the second. For instance, on one hand, in our results the primary yield losses of the category PYL14-3 (~30% in a year of high production) were associated with the secondary yield losses of the category SYL15-3 (~70%) (Fig. 5.6 and Fig. 5.9), indicating that the secondary yield losses after a year of high production could be more than the double of the primary yield losses of such year. On the other hand, the primary yield losses (PYL15-1 - PYL15-1) in a year of low production (2015) were very low (<1%), indicating that secondary losses in the next year (2016) would be also low. This become a novel finding which can serve as a warning about what to expect in terms of reduced production in the next year, especially after a year of high production. This type of warning can also serve as motivator for improving the crop management in order to reduce losses, and therefore, increase gains (Savary and Willocquet, 2014).

Injury profiles and the resulting primary and secondary losses confirm the polyetic behavior of the epidemics. In tropical areas, where there are not cut breaks between growing seasons, polyetic epidemics can be the consequence of the conservation or even the accumulation of the inoculum from one growing season to the next one; that way, epidemics can be continuous during many years (Arneson, 2001). This helps to understand why injury profiles were stable along time. For instance, coffee leaf rust maintained its high incidence and brown eye spot even increased during the two years of study, whereas this relationship was apparently not driven by the topoclimate. An outbreak of coffee leaf rust began in 2012 in Central America (Avelino *et al.*, 2015), causing reductions of production up to 30% until 2014 (Baker, 2014), and its effects are still damaging coffee plantations. This finding becomes an important argument to support that coffee farmers actually need aid to recover their plantations for more than two continued years after an important outbreak of diseases.

4.3 Implications for the management of coffee agroecosystems

We have shown the main drivers leading to different injury profiles, according to the associations found in the correspondence analyses. Based on that, we provide the following key recommendations to avoid high injury levels caused by pests and diseases, and therefore to avoid high yield losses.

One of the most important factors to consider is the topoclimate. The altitude should be the first to be considered given that it is strongly related to temperatures and rainfall, which cannot be modified by farmers, and therefore management should be adapted to the climate conditions of the site (Allinne *et al.*, 2016). This implies, for instance, that in low lands the control of pests and diseases should be more careful than in high lands. Thus, coffee leaf rust and dieback would need more preventive cropping practices and/or more fungicide sprays in low lands. Furthermore, shade could help to regulate microclimate against pathogens as long as it is well adapted to site conditions (Ratnadass *et al.*, 2012). For instance, as suggested by a previous study (Cerda *et al.*, 2017), unfavorable conditions for fungi could be generated in low lands with low shade cover (~15%) in simple agroforestry systems by maintaining low moisture in the system; whereas in high lands they could be generated by higher shade cover (~30%) that would generate low temperatures, also unfavorable for fungi.

The avoidance of high injury levels could be achieved not only by fungicide sprays, but also considering other aspects of management. For instance, the profiles of management associated with the lowest injury levels, had a higher distance between coffee rows than other management profiles. This suggests that coffee plots managed with broader alleys between rows of coffee plants could promote the ventilation of the microclimate, and therefore disfavor fungal diseases. Of course, distances of coffee rows are hard to change in already established plantations; this finding is more useful for the design and management of new coffee agroecosystems.

The knowledge of physiological characteristics of coffee plants becomes also important to avoid severe injury profiles. For instance, high fruit load makes plants more susceptible to pests and diseases (DaMatta *et al.*, 2007). Our results suggest that when coffee plants are old (>25 years), high fruit loads per plant should especially be avoided, given that such conditions were related to severe injury profiles. High fruit loads can be easily avoided by regulating the number of productive stems per plant through pruning. If the farmer wants high fruit loads, then he should better think in renewing coffee plants. With respect to plant nutrition, the fact that profiles of plants with the highest nutrient contents in leaves were associated with the most severe injury profiles, suggests that an excessive foliar nutrition should be avoided, because such condition is more attractive to pathogens and insects.

The confirmation of polyetic epidemics in this study represents a hard challenge in the search of management options to reduce their negative impacts. Stop the transfer of inoculum from one growing season to the next one would be practically impossible, given that the hosts (coffee plants) are permanently in the field; whereas trying to reduce the accumulation of inoculum would be more feasible. For instance, the renewing of old coffee plants can be an alternative, as well as the use of varieties tolerant or resistant to the main pathogen (Avelino *et al.*, 2015; McCook and Vandermeer, 2015). The enhancement of antagonist microorganisms to pathogens, such as *Lecanicillium lecanii*, under diversified agroforestry systems (Staver *et al.*, 2001; Jackson *et al.*, 2012), could also contribute to the reduction of the pathogen inoculum accumulation.

4.4 Caveats

The associations found in this study can be applicable to other regions with similar characteristics of production situations that we studied (reflected in section 2.2. and Figs. 2 to 5). In regions with very different production situations, and/or where coffee plantations have even more complex shade canopies with much higher shade cover, for instance, there can be different pests and diseases, and therefore different injury profiles, and different associations with production situations and management strategies.

We demonstrated the usefulness of the analytical approaches used in this study to reveal important implications of injury profiles and their resulting yield losses, and to give lights to reduce negative impacts. Thereby, it would be worth to perform similar studies for the most important food and cash crops at regional levels, in order to derive more generalizable conclusions, as it was done with studies in rice in tropical Asia (Savary *et al.*, 2000a; Savary *et al.*, 2000b).

4.5 Prospects

This study has produced data on primary and secondary yield losses, and explanations on how they occur. A next step would be the estimation of the economic impact of such yield losses. The valuation of economic losses is useful both for improving management decisions and for the valuation of ecosystem services (Klein *et al.*, 2007). The regulation of pests and diseases is a major ecosystem service of global interest, whose effectiveness should be evaluated with indicators such as yield losses and their resulting economic losses (Avelino *et al.*, 2011). Furthermore, given that there are also many other ecosystem services of interest, indicators of the regulation of pests and diseases should be evaluated along with indicators of other ecosystem services in order to find the best ways to provide several ecosystem services at the same time without trade-offs (Cheatham *et al.*, 2009; Rapidel *et al.*, 2015).

The assessment of relationships between indicators of different ecosystem services can give lights for a better design and management of coffee agroecosystems. Previous studies, as well as the present one, have provided general recommendations or insights on how to better manage coffee agroecosystems in order to reduce impacts of pests and diseases or injury profiles (Staver *et al.*, 2001; Avelino *et al.*, 2006; Avelino *et al.*, 2007; Allinne *et al.*, 2016; Cerda *et al.*, 2017). However, it is necessary to identify the most promising agroecosystems to provide multiple ecosystem services simultaneously, in order to learn and derive from them more precise recommendations on the structure, composition and management that should be applied to the system for being successful (Rapidel *et al.*, 2015; Cerda *et al.*, 2017).

5. Conclusions

This study demonstrated that gradients of primary and secondary yield losses (from low to high losses) were positively related to gradients of injury profiles (from slight to highly severe profiles). Each injury profile depended on particular combinations of profiles of management strategies, production situations and also physiological characteristics of coffee plants. Injury profiles had impacts on yield losses especially in a year of high coffee production (primary yield losses), but compromising also the yields of the next year (secondary yield losses). These findings are important, because by knowing particular combinations of production situations and management strategies in a given site, it is possible to know what injury profiles and what levels of yield losses to expect. For instance, we deduced that increasing fruit loads in old coffee plants, increasing contents of nutrients in coffee leaves, decreasing altitudes, and suboptimal management were the main drivers leading to severe injury profiles, and therefore to the highest yield losses.

Primary yield losses resulting from the most severe injury profile were high (~30%) and secondary yield losses even higher (~70%). Furthermore, they were positively associated given that they have similar drivers; indicating that by assessing primary yield losses, it is possible to deduce secondary yield losses.

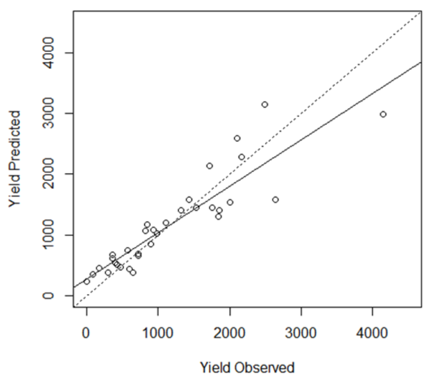
The analytical approach used in this study was useful to identify the main causes leading to severe injury profiles and high yield losses, and therefore it also permitted to derive important insights for the management of coffee agroecosystems in order to reduce the negative impacts of injury profiles (losses).

6. Acknowledgements

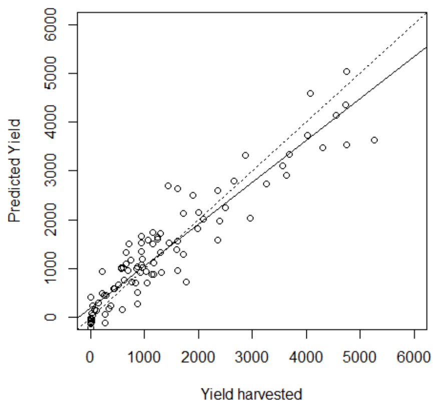
This research was supported by CASCADE project “Ecosystem-based Adaptation for Smallholder Subsistence and Coffee Farming Communities in Central America”, funded by the International Climate Initiative (ICI). The German Federal Ministry for the Environment, Nature Conservation, Building and Nuclear Safety (BMUB) supports this initiative on the basis of a decision adopted by the German Bundestag.

7. Supplementary materials of manuscript 3

Supplementary table. Models and equations, and indicators of prediction quality, to estimate attainable yields actual yields, and primary and secondary losses of coffee, mentioned in section 2.4

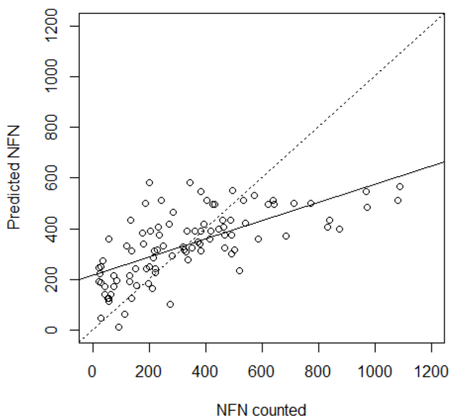
Eq.	Models and equations	Indicators of prediction quality
(Model for 2014)	$Y_{(14)} \sim NFN_{(14)} + DeadB_{(14)} + ShadeC_{(14)} + (1 \mid \text{Plot})$ Y: actual coffee yield per plant; NFN: number of fruiting nodes per plant; DeadB: number of dead productive branches per plant; ShadeC: mean annual shade cover; $_{(14)}$: represents the variables in the year 2014 Shade cover had no significant effect on yield in 2014	
1	$Y_{(14)} = 215.91 + 2.70{\times}NFN_{(14)} - 24.95{\times}DeadB_{(14)}$  Dashed line in the graphic is the relation 1:1; continuous line is the prediction	Mean of yield observed values = 1167 g plant^{-1} (grams of ripe fresh coffee cherries per plant) Bias = 1.35 x 10^{-13} R^2 = 0.81 MAE = 280.48 RMSE = 384.44 RRMSE = 32.94 EF = 0.81 IA = 0.94 Where Bias: mean of residues; MAE: mean absolute error; RMSE: root-mean-squared error; RRMSE: relative root-mean-squared error; EF: modeling efficiency; IA: index of agreement
2	$Ya_{(14)} = 215.91 + 2.70{\times}NFN_{(14)}$ $Ya_{(14)}$: specific attainable yield in 2014	
3	$\%PYL_{(14)} = \frac{Ya_{(14)} - PYL_{(14)}}{Ya_{(14)}} \times 100$ $\%PYL_{(14)}$: primary yield losses in percentage in 2014	

(Continued)

Eq.	Models and equations	Indicators of prediction quality
(Model for 2015-1)	$Y_{(15)} \sim NFN_{(15)} + DeadB_{(15)} + ShadeC_{(15)} + (1 \mid \text{Plot})$ Y: actual coffee yield per plant; NFN: number of fruiting nodes per plant; DeadB: number of dead productive branches per plant; ShadeC: mean annual shade cover; $_{(15)}$: represents the variables in the year 2015	
4	$Y_{(15)} = -328.50 + 3.96 \times NFN_{(15)} - 1.56 \times DeadB_{(15)} + 21.96 \times ShadeC_{(15)}$  Dashed line in the graphic is the relation 1:1; continuous line is the prediction	Mean of yield observed values = 1343 g plant^{-1} (grams of ripe fresh coffee cherries per plant) Bias = 2.54 x 10^{-13} R^2 = 0.87 MAE = 353.98 RMSE = 465.65 RRMSE = 34.68 EF = 0.87 IA = 0.96 Where Bias: mean of residues; MAE: mean absolute error; RMSE: root-mean-squared error; RRMSE: relative root-mean-squared error; EF: modeling efficiency; IA: index of agreement
5	$Ya_{(15)} = -328.50 + 3.96 \times NFN_{(15)} + 21.96 \times ShadeC_{(15)}$ $Ya_{(15)}$: specific attainable yield in 2015	
6	$PYL_{(15)} = Ya_{(15)} - Y_{(15)}$ $PYL_{(15)}$: primary yield loss (in weight) in 2015	

(Continued)

<table>
<tr><th>Eq.</th><th>Models and equations</th><th>Indicators of prediction quality</th></tr>
<tr><td>(Model for 2015-2)</td><td>$$NFN_{(15)} \sim DeadB_{(14)} + ShadeC_{(14)} + (1 \mid \text{Plot})$$
NFN: number of fruiting nodes per plant; DeadB: number of dead productive branches per plant; ShadeC: mean annual shade cover; $_{(15)}$: represents the variables in the year 2015

Shade cover had no significant effect on NFN of 2015</td><td></td></tr>
<tr><td>7</td><td>$$NFN_{(15)} = 365.93 - 16.1 \times DeadB_{(14)}$$


Dashed line in the graphic is the relation 1:1; continuous line is the predictions</td><td>Mean of yield observed values = 337 NFN plant^{-1}

Bias = 4.90 x 10^{-13}
R^2 = 0.44
MAE = 141.98
RMSE = 190.35
RRMSE = 56.40
EF = 0.43
IA = 0.73
Where Bias: mean of residues; MAE: mean absolute error; RMSE: root-mean-squared error; RRMSE: relative root-mean-squared error; EF: modeling efficiency; IA: index of agreement</td></tr>
<tr><td>8</td><td>$$NFNa_{(15)} = 365.93$$
$NFNa_{(15)}$: fruiting nodes that would have been attainable</td><td></td></tr>
<tr><td>9</td><td>$$NFNl_{(15)} = NFNa_{(15)} - NFN_{(15)}$$
$NFNl_{(15)}$: lost fruiting nodes</td><td></td></tr>
<tr><td>10</td><td>$$SYL_{(15)} = -328.50 + 3.96 \times NFNl_{(15)} + 21.96 \times ShadeC_{(15)}$$
$SYL_{(15)}$: secondary yield loss (in weight) in 2015</td><td></td></tr>
<tr><td>11</td><td>$$Ya_{(14-15)} = -328.50 + 3.96 \times 365.93 + 21.96 \times ShadeC_{(15)}$$
$Ya_{(14-15)}$: attainable yield without negative impacts of 2014 and 20154</td><td></td></tr>
<tr><td>12</td><td>$$\%PYL_{(15)} = \frac{PYL_{(15)}}{Ya_{(14-15)}} \times 100$$
$\%PYL_{(15)}$: primary yield losses in percentage in 2015</td><td></td></tr>
<tr><td>13</td><td>$$\%SYL_{(15)} = \frac{SYL_{(15)}}{Ya_{(14-15)}} \times 100$$
$\%SYL_{(15)}$: secondary yield losses in percentage in 2015</td><td></td></tr>
</table>

Chapter 6. Reduction of coffee losses and provision of multiple ecosystem services

Manuscript 4: Coffee agroforestry systems for reducing crop losses while providing multiple ecosystem services

This manuscript is complete, the discussion however needs to be improved and enlarged. Afterwards, the manuscript will be submitted to the journal Agriculture, Ecosystems and Environment or to the journal Agroforestry Systems

Coffee agroforestry systems for reducing crop losses while providing multiple ecosystem services

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Abstract

The regulation of pests and diseases is one of the ecosystem services of most interest for agriculture worldwide, given that crop losses caused by harmful organisms threaten the food security and the incomes of farmers and their families. Apart from that, there is also an urgent need to ensure that farming systems provide multiple ecosystem services simultaneously. For thousands of farmers and workers, whose wellbeing depend on the coffee crop, it is important to know: what are the shade canopy and management characteristics of the coffee agroecosystems able to supply the lowest coffee losses and the highest ecosystem services? To contribute to answer this question, we first assessed the relationships between primary and secondary coffee losses (as indicators of regulation of pests and diseases) and indicators of other three ecosystem services: provisioning of agroforestry products, maintenance of soil fertility and carbon sequestration. Such indicators were measured during two years in 61 farmers' coffee plots of a research network. Based on the results of 48 analyzed relationships, we then defined five criteria to identify the most promising coffee agroecosystems capable to offer the lowest losses and provide multiple ecosystem services at the same time. We found 20 significant relationships between crop losses and ecosystem services (denoting slight trade-offs). In those 20 and in the other 28 not significant relationships, it was noticed that coffee agroforestry systems accomplished better levels of ecosystem services than coffee monocultures in full sun. Finally, we identified six coffee agroforestry systems (CAF) as the most promising ones to reduce crop losses and provide other ecosystem services. One of these CAF was a simple agroforestry system (coffee-service trees); three were medium diversified coffee agroforestry systems (coffee-musaceaes-service trees-timber trees); and two were highly diversified coffee agroforestry systems (coffee-service trees-timber trees-fruit trees-musaceas). We described in detail their characteristics of structure and composition in the shade canopy, management and costs, and the levels of ecosystem services they provide. The six CAF represent several options to choose (imitate) for the design of new plantations, or for transforming the current characteristics of existing plantations to similar characteristics of one of them.

Key words: Carbon sequestration; Incomes, Management; Pests and diseases; Primary losses; Secondary losses; Shade; Soil; Yields

1. Introduction

There is an urgent need to ensure that farming systems not only provide high yields, but also the provision of multiple ecosystem services on which farmer households and other stakeholders depend (Cheatham *et al.*, 2009; Vignola *et al.*, 2015). Among ecosystem services, the regulation of pests and diseases is of the highest interest for agriculture worldwide. Pests and diseases cause severe losses of crop products, threatening the food security and the incomes of farmers and their families (Oerke *et al.*, 1994).

The effectiveness of the regulation of pests and diseases should be measured in terms of reduced crop losses (Avelino *et al.*, 2011). Traditionally, the performance of a cropping system for regulating pests and diseases is measured in terms of reduction of the presence (incidences) or severity of symptoms/signs of specific pathogens or insects; but these measurements do not fully guarantee that crop losses are low, as other harmful but not assessed pests or pathogens can arise. On one hand, data of quantified crop losses are necessary to reinforce that the reduction of specific pests and diseases is actually accompanied with reduced crop losses. On the other hand, data on quantified crop losses could demonstrate that it is possible to live with certain levels of pests and diseases, and then limit pesticide use, without incurring high crop losses. For the latter, the assessment of crop losses should be related to particular characteristics of the environment, structure, biodiversity and management under which the crop is grown (Staver *et al.*, 2001; Ratnadass *et al.*, 2012).

The assessment of crop losses must quantify the loss in quantity and/or quality of the crop product due to pests and diseases (Oerke, 2006), and quantify also the economic losses (Nutter *et al.*, 1993). Furthermore, there can be primary and secondary crop losses. Primary crop losses are those caused in the specific year when pest and disease injuries occur; secondary crop losses are those resulting from negative impacts of pests and diseases of the previous year (Zadoks and Schein, 1979; Avelino *et al.*, 2015). In the case of perennial crops such as coffee, secondary losses can be higher than primary losses, as demonstrated in experimental conditions (Cerda *et al.*, 2016a, Chapter 4), and in coffee plots of smallholder farmers (Cerda *et al.*, 2016b, Chapter 5). That is why the assessment of the regulation of pests and diseases must consider both types of losses, which means that at least two years of production must be considered, which is also the shortest period of time for coffee yield assessments, considering its biennial production cycle.

Apart from the regulation of pests and diseases, there are other ecosystem services of interest for farmers and for the society in general. For instance, for farmers and their families, the provisioning of crop products is of course of great interest; diversified production systems such as agroforestry systems, apart from the main crop product, can provide fruits, timber, firewood and others (Rice, 2008). The maintenance of soil fertility is another service of interest for the land owners, given that their production depends in great part on the state of this resource. For the society in general, the carbon sequestration is an essential service which contributes to the mitigation of climate change (MEA, 2005; Müller *et al.*, 2015). All of these services can be provided individually or together simultaneously, depending on the type of agroecosystem. For instance, agroforestry systems are considered to provide more ecosystem services than monocrop systems (Jose, 2009).

Given that the reduction of crop losses by the regulation of pests and diseases would not be the only service of interest, it becomes important to know what are the relationships between crop losses and indicators of other ecosystem services. The knowledge on these relationships would allow us to know which trade-offs among ecosystem services should be avoided, and what synergies could be best used (Rapidel *et al.*, 2015).

The knowledge on ecosystem services and relationships among them can also help to identify which types of agroecosystems and management decisions are needed to provide multiple ecosystem services (Lamarque *et al.*, 2014). This type of information, for instance, is of common interest of scientists, technicians and farmers who work with perennial crops in agroforestry systems, given that these systems are capable to provide multiple ecosystem services that could be improved (De Beenhouwer *et al.*, 2013).

In the case of coffee, an important question for the wellbeing of farmers´ families is: what are the shade canopy and management characteristics of the coffee agroecosystems able to supply the lowest coffee losses and the highest ecosystem services? A recent study, based on bivariate analyses of relationships between ecosystem service indicators, contributed with general recommendations for the design and management of coffee agroecosystems to provide several ecosystem services simultaneously (Cerda *et al.*, 2017). In order to reinforce this latter contribution, the present study followed similar analytical approaches, but using two-year data on ecosystem services, including regulation of diseases, with two objectives: i) assess the relationships between crop losses (as indicators of regulation of pests and diseases) and indicators of other three ecosystem services: provisioning of agroforestry products, maintenance of soil fertility and carbon sequestration, and ii) identify the most promising coffee agroecosystems capable to offer the lowest crop losses and provide multiple ecosystem services at the same time. We described and discussed these systems to give lights on several options of structure, composition, and management that coffee farmers can use to increase ecosystem services.

2. Materials and methods

2.1 Location, data collection and indicators of ecosystem services

We worked with data collected during two years in 61 coffee plots of a research network established in Turrialba, Costa Rica. Turrialba is catalogued as a premontane wet forest life zone (mean annual rainfall = 2781 mm and a mean annual temperature = 22.2°C; 10 year averages), where coffee is grown from 600 to 1400 m.a.s.l. The plot sampling strategy aimed to gather plots where coffee is grown under different management strategies and production situations; therefore, the strategy consisted in the selection of coffee plots with contrasting characteristics of types of shade and cropping practices in different altitudes. All coffee plots had the coffee variety *Caturra* as the dominant one, and were owned by smallholder farmers. In each coffee plot, we demarcated a subplot composed of eight coffee rows with 15 plants each; where eight coffee plants (one per row) were marked, and used for measurements related to diseases and coffee yields, and for sampling soil near them. We also demarcated a circular area of 1000 m^2, from the center of the subplot, for measurements related to the shade canopy.

In this network, we measured different variables to calculate indicators of four ecosystem services: maintenance of soil fertility, carbon sequestration, provisioning of agroforestry products and regulation of diseases. We also measured yield components of coffee plants (number of productive branches, fruiting nodes, and fruits per node) and yield reducing factors (number of dead productive branches) in the eight marked coffee plants. The composition and structure (abundances and trunk diameters) of trees and other plants in the shade canopies were measured as well. A simple clustering analysis with such variables permitted to group the coffee plots in three main types of coffee plantations: coffee in full sun (CFS), coffee agroforestry systems with low diversification (CLD), and coffee agroforestry systems with high diversification (CHD). Through interviews with the owners, we obtained the data on cropping practices and costs for the management of coffee plots, and the production of agroforestry products apart from coffee (bananas, plantains, other fruits). Equations to estimate yields and yield losses, and to estimate economic losses are shown in Table 6.1. Ecosystem service indicators and characteristics of coffee plots, along with the main methods to obtain them, are listed in Table 6.2.

Table 6.1. Equations to estimate coffee yields and primary and secondary losses

Explanations	Models and equations	Eq.
For the estimation of primary yield losses in 2014		
We used equation 1 to estimate the actual yield of 2014 ($Y_{(14)}$)	$Y_{(14)} = 215.91 + 2.70 \times NFN_{(14)} - 24.95 \times DeadB_{(14)}$	1
By setting the dead branches as "zero", we estimated the specific attainable yield of 2014 ($Ya_{(14)}$)	$Ya_{(14)} = 215.91 + 2.70 \times NFN_{(14)}$	2
The previous equations estimated yields and yield losses in grams of ripe coffee cherries per plant, and before applying the following equations, they were transformed to kg per hectare, by using the density of coffee plants per hectare of each plot		
We estimated the primary yield loss as the difference between $Ya_{(14)}$ and $Y_{(14)}$	$PYL_{(14)} = Ya_{(14)} - Y_{(14)}$	3
We estimated the primary yield loss as a percentage of the specific attainable yield of 2014	$\%PYL_{(14)} = \frac{PYL_{(14)}}{Ya_{(14)}} \times 100$	4
For the estimation of secondary yield losses in 2015		
We used equation 5 to estimate the actual yield of 2015 ($Y_{(15)}$)	$Y_{(15)} = -328.50 + 3.96 \times NFN_{(15)} - 1.56 \times DeadB_{(15)} + 21.96 \times ShadeC_{(15)}$	5
We used the equation 6 to estimate the actual fruiting nodes of 2015 ($NFN_{(15)}$)	$NFN_{(15)} = 365.93 - 16.1 \times DeadB_{(14)}$	6
By setting the dead branches as "zero", we estimated the fruiting nodes that would have been attainable ($NFNa_{(15)}$) without reducing factors of 2014	$NFNa_{(15)} = 365.93$	7
Then, we estimated the loss of fruiting nodes for 2015 ($NFNl_{(15)}$)	$NFNl_{(15)} = NFNa_{(15)} - NFN_{(15)}$	8
Thereby, we considered that estimating a yield with this $NFNl_{(15)}$ using the equation (5) represented the secondary yield losses	$SYL_{(15)} = -328.50 + 3.96 \times NFNl_{(15)} + 21.96 \times ShadeC_{(15)}$	9
To estimate an attainable yield without negative impacts of 2014 and 2015 ($Ya_{(14-15)}$), we replaced equation 7 in equation (4) and then set as zero the dead branches of both years to assume that there were no reducing factors	$Ya_{(14-15)} = -328.50 + 3.96 \times 365.93 + 21.96 \times ShadeC_{(15)}$	10
The previous equations estimated yields and yield losses in grams of ripe coffee cherries per plant, and before applying the following equations, they were transformed to kg per hectare, by using the density of coffee plants per hectare of each plot		
We calculated the secondary yield loss of 2015 $\%SYL_{(15)}$ as a percentage of the $Ya_{(15)}$	$\%SYL_{(15)} = \frac{SYL_{(15)}}{Ya_{(14-15)}} \times 100$	11
For the estimation of economic primary and secondary losses		
Primary Economic loss (PEL) in 2014 was calculated with the price of coffee in that year	$PEL = PYL_{(14)} \times Price\ of\ coffee_{(14)}$	12
Secondary Economic loss (SEL) in 2015 was calculated with the price of coffee in that year	$SEL = SYL_{(15)} \times Price\ of\ coffee_{(15)}$	13

Source: Equations from 1 to 11 come from Cerda *et al*. (2016b, Chapter 5).
Y: actual coffee yield per plant; NFN: number of fruiting nodes per plant; DeadB: number of dead productive branches per plant; ShadeC: mean annual shade cover; $_{(14)}$: represents the variables in the year 2014; $_{(15)}$: represents the variables in the year 2015
The price of coffee in 2014 was 0.48 USD per kg of ripe fresh coffee cherries; in 2015 it was 0.39 USD per kg of ripe fresh coffee cherries. These prices were the same for all coffee plots, given that the farmers sell the coffee to the same company "Compañía Santa Rosa de Turrialba".

Table 6.2. List of indicators of ecosystem services (ES) measured during two years (2015-2015) in the coffee plot network, Turrialba, Costa Rica

Indicators of ES	Methods/Formulas		Data sources/times of measurement
Regulation of pests and diseases			
sAUDPC (%) of: Coffee leaf rust and Brown eye spot	$AUDPC = \sum_{i=1}^{n-1} \frac{I_i + I_{i+1}}{2} \times (t_{i+1} - t_i)$ $\quad sAUDPC = \frac{AUDPC}{Nd}$ where *AUDPC*: area under the disease progress curve; I_i: incidence of a given disease at the *i*th measurement; t_i: time (in days) of the *i*th measurement; n: total number of measurements; *sAUDPC*: standardized AUDPC; *Nd*: total number of days in which the plants were measured Source: (Simko and Piepho, 2012)		Incidences where measured five times: 1st Fruit formation (slightly dry period); 2nd Beginning of fruit ripening (beginning of rainy period); 3rd Just before the harvest (rainy period); 4th During the peak of harvest (slightly dry period); 5th End of coffee harvest period (highest rainy period)
Maximum dieback index (%)	Each plant of the subplot was classified in a scale (1-4): 1: plant with few defoliated branches; 2: plants with several defoliated and dead branches; 3: plants with a lot of defoliated and dead branches, and with withered dry fruits; 4: plants with almost all dead branches, almost dead plants. Dieback index were calculated with the formula below (where N: number of plants registered in each scale of dieback); and the maximum dieback index during the year was identified $Dieback\ index = \frac{1xN_1 + 2xN_2 + 3xN_3 + 4xN_4}{4xNtotal} \times 100$		The same as above
Primary and secondary yield losses (%) Primary and secondary economic losses (USD ha^{-1})	These indicators were estimated through several equations. See the explanations and equations in Table 6.1		
Provisioning of agroforestry products			
Cash flow (USD ha^{-1}) Value DC (USD ha^{-1})	*GI = AS x MP* *CF = GI – CC* *Value DC = ADC x MP*	where: *GI*: gross income from sale of agroforestry products; *AS*: amount of agroforestry products for sales; *MP*: local market price; *CF*: cash flow; *CC*: cash costs; *Value DC*: value of domestic consumption; *ADC*: amount of agroforestry products for domestic consumption Sources: (Ambrose-Oji, 2003; Cerda *et al.*, 2014)	Data on management practices of coffee plots, costs of labor and inputs, and agroforestry production (fruits, bananas, plantains, etc.) were obtained through interviews with the owners of the coffee plots
Maintenance of soil fertility*			
Acidity (mg kg^{-1}) K (mg kg^{-1}) Carbon (%)	Eight subsamples of soil at a depth of 0-40 cm were taken near the trunk of eight coffee plants (at 50cm approximately) in each experimental subplot. The subsamples were mixed to obtain a composite sample, and sent to a soil laboratory for the chemical and texture analyses Source: (Briceño and Pacheco, 1984)		The subsamples and composite samples were obtained during the peak of harvest in 2014
Carbon sequestration*			
Carbon (Mg ha^{-1}) in: Fruit trees Timber trees Service trees	Biomasses of plants and trees in the shade canopy were estimated through allometric equations, which use the trunk diameters at breast height (1.3 m from soil) as predictor. Detailed equations can be found in Source (Cerda *et al.*, 2017). Then, biomasses were multiplied by a 0.47 fraction to obtain the carbon content		The trunk diameters were measured the coffee fruit formation (slightly dry period)

Coffee leaf rust (*Hemileia vastarix* Berkeley and Broome); Brown eye spot (*Cercospora coffeicola* Berk and Curtis)
*: these indicators were measured only in the year 2014, the year of high production according to the biennial cycle of coffee production.
Source: adapted from (Cerda *et al.*, 2017)

2.2 Analytical methods

2.2.1 Representation of the relationships between crop losses and ecosystem services

Bivariate linear regressions were performed between indicators of coffee crop losses and indicators of presence of diseases (both indicators of regulation of diseases), and between indicators of crop losses and indicators of the other ecosystem services studied. The dependent variables were: primary yield losses, economic primary losses, secondary yield losses, and economic secondary losses; and the regressor variables were the indicators of presence of diseases, provisioning of agroforestry products, maintenance of soil fertility and carbon sequestration listed in Table 6.2. Given that low levels of crop losses are desirable, the interpretation of relationships were: negative relationships between crop losses and a desirable high level of another indicator (e.g. carbon sequestration) denote synergies, and positive relationships denote trade-offs; whereas, negative relationships between crop losses and a desirable low level of other indicator (e.g. soil acidity) denote trade-offs, and positive relationships denote synergies.

2.2.2 Identification of the “desirable areas”

A total of 48 regressions were performed and represented graphically, and a “desirable area” was demarcated in each graphic. The “desirable area” was defined as the area where the desirable levels of both ecosystem service indicators can be found; this area was demarcated to identify which type of coffee plots achieved those levels. The demarcation of the “desirable areas” followed three steps: i) the outlier points of indicators of crop losses were removed from the analysis and graphics, to avoid considering very uncommon observations; ii) the cloud of points in graphics between crop losses and indicators of presence of diseases were divided into two zones according to the medians of crop losses (absence of regulation of specific diseases with low crop losses is considered as an acceptable situation), while the cloud of points in graphics between crop losses and indicators of the other ecosystem services were divided into four quadrants based on the data of the medians in each axis (in these cases both indicators in the axis are of interest); and, iii) the best quadrant, representing the “desirable area”, was demarcated (for instance, in a graphic between yield loss and carbon sequestration, the desirable area would be located in the lower-right corner). Given that medians were the most important data to define desirable areas, such medians were considered as boundary values in this study, which meant that desirable levels of ecosystem service indicators were those located at least within the 50% of the best levels.

2.2.3 Identification and characterization of the most promising coffee agroecosystems

The most promising coffee agroecosystems were identified based on five criteria: i) the coffee plots should be located in at least 32 desirables areas in general (two thirds of the total 48 desirable areas); ii) the coffee plots should be located in at least one desirable area between primary crop losses and indicators of each ecosystem service; iii) the coffee plots should be located in at least one desirable area between secondary crop losses and indicators of each ecosystem service; iv) the coffee plots should be within the 50% plots with lower economic secondary losses; and v) the coffee plots should not present negative values of cash flow in none of the two studied years. We considered that the coffee agroecosystems complying with the five criteria, were the most promising ones for the regulation of diseases (reduced crop losses), the provisioning for farmers´ families, the maintenance of soil fertility and the sequestration of carbon. Once the most promising coffee agroecosystems were identified, we described their characteristics of structure and composition of their shade canopies, the characteristics of management that farmers apply to these systems, the resulting costs, and finally the indicators of ecosystem services that they provide.

3. Results

We found 20 significant relationships between crop losses and ecosystem services (out of the total 48 regressions), and we found six most promising coffee agroforestry systems to provide multiple ecosystem services. Figures of relationships are presented and interpreted in the first section of results. In each figure, the main types of coffee plantations considered in this study can be differentiated, and the six most promising coffee agroforestry systems can be already identified. Then, the six most promising coffee agroforestry systems are described in detail in the second section of results.

3.1. Relationships between indicators of crop losses and indicators of ecosystem services

Coffee leaf rust and dieback were positively related to crop losses, although the R^2 were not high; whereas no significant relationships were found between crop losses and brown eye spot. In the case of coffee leaf rust, low primary losses were found within the complete range (10-80%) of the standardized area under the disease progress curve (sAUDPC), but low secondary losses were not found above 70% of sAUDPC. In the case of dieback, both low primary and secondary losses were found below 60% of the maximum index of the disease. In the case of brown eye spot, both low primary and secondary losses were found within the range 0-14% of the sAUDPC. Coffee plots of the three main types of coffee plantations (CFS, CLD and CHD), and the six most promising coffee agroforestry systems were located in desirable areas (Fig. 6.1).

Positive relationships were found between the value of domestic consumption and secondary losses; whereas negative relationships were found between cash flow and primary yield loss, and between coffee yield and both primary and secondary yield losses (Fig. 6.2). Yet the R^2 were low in general (<0.10). It was interesting to note that none of the coffee plots with more than 900 USD ha^{-1} of value of domestic consumption in 2015 were in the desirable areas. Most of the coffee plots in the desirable areas were agroforestry systems (CLD and CHD). The six most promising coffee agroforestry systems were in desirable areas involving secondary losses, but not all of them in desirable areas regarding primary losses (Fig. 6.2).

Positive relationships were found between soil K and secondary losses; whereas a negative relationship was found between soil C and primary yield losses. All of them with low R^2 (<0.13) (Fig. 6.3). Coffee plots of the three main types of coffee plantations (CFS, CLD and CHD) were in most desirable areas, except in desirable areas involving crop losses and soil K, where most plots were agroforestry systems. The six most promising coffee agroforestry systems were in desirable areas involving crop losses and soil acidity, but not all of them in desirable areas regarding soil K and soil C (Fig. 6.3).

Carbon in timber and fruit trees were related positively with secondary losses, and C in fruit trees were also related to primary economic losses; although the R^2 were low in general (<0.16) (Fig. 6.4). Only coffee plots of the type agroforestry systems (CLD and CHD) were in desirable areas as expected, given that coffee plots in full sun did not have shade trees. None of the coffee plots with carbon in timber trees higher than 8 Mg ha^{-1}, and none of the coffee plots with carbon in fruit trees higher than 4 Mg ha^{-1} were in the desirable areas. In the case of carbon in service trees, it was interesting to see that plots with carbon until 27 Mg ha^{-1} were still located at least in the desirable areas with respect to secondary losses. Most of the six most promising coffee agroforestry systems were in desirable areas involving crop losses and timber trees, but few of them (two as maximum) were in desirable areas regarding fruit trees and service trees (Fig. 6.4).

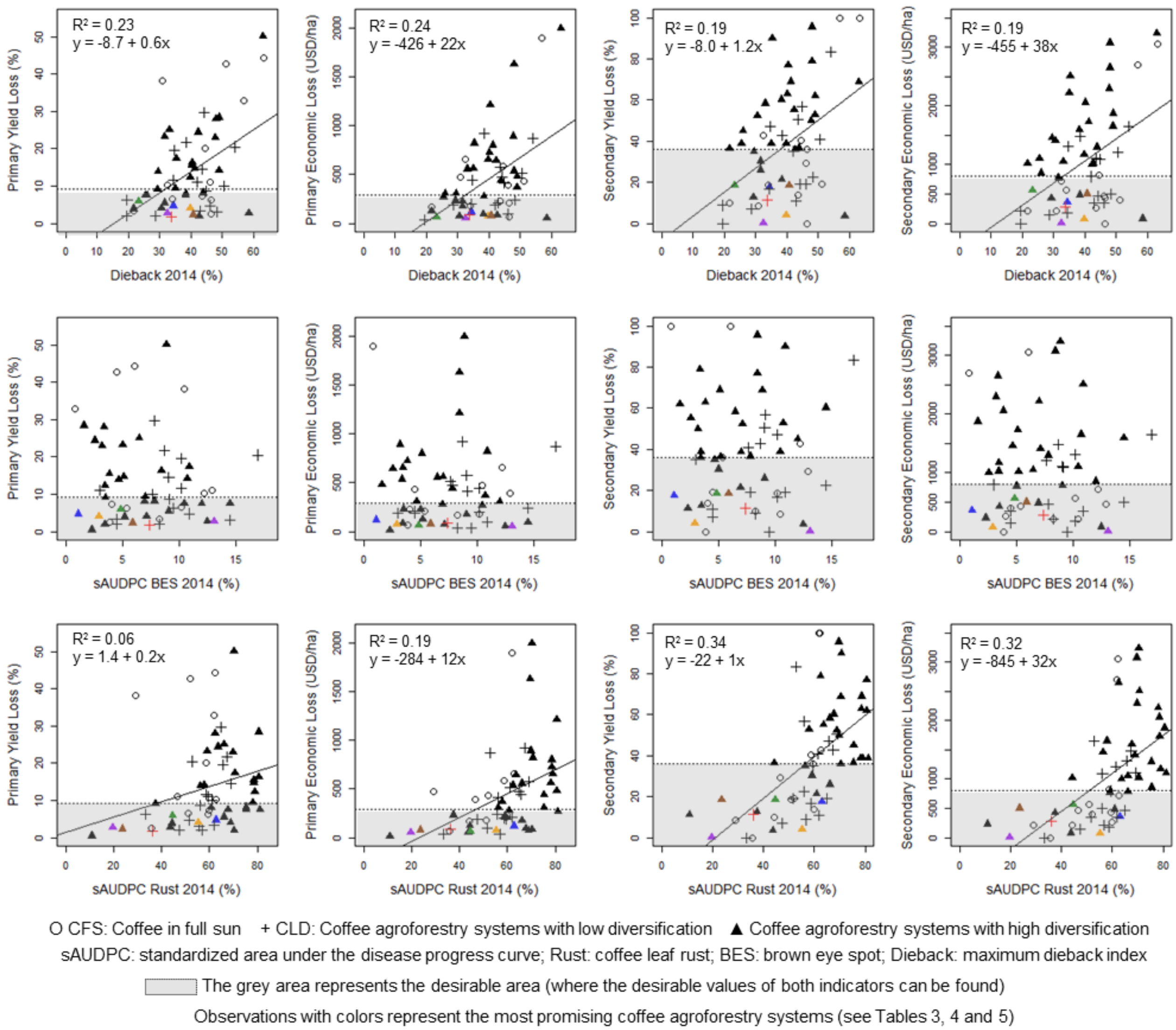

Fig. 6.1. Relationships between indicators of crop losses and indicators of presence of diseases; and different types of coffee plots achieving the most desirable low levels of crop losses.

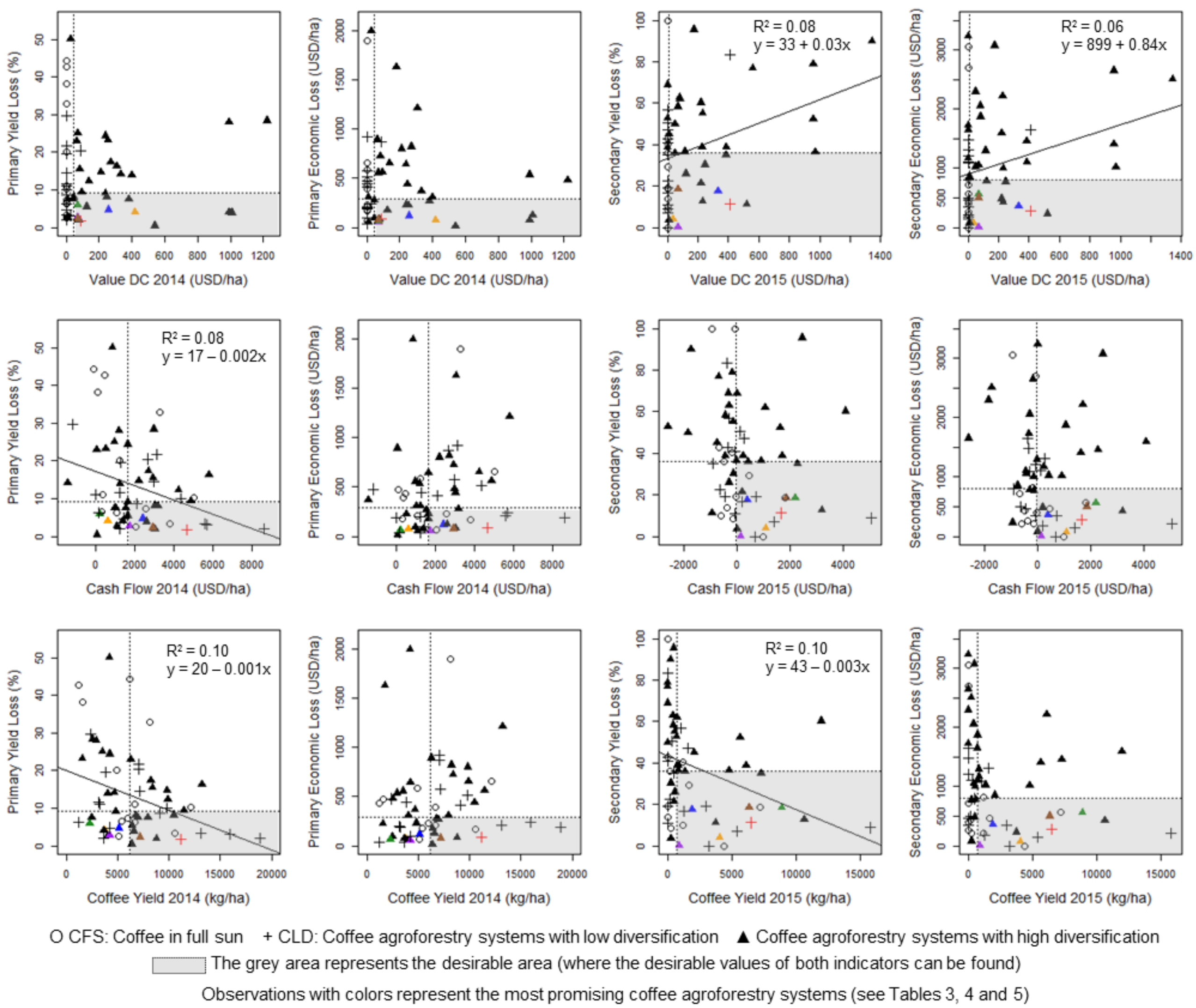

Fig. 6.2. Relationships between indicators of crop losses and indicators of provisioning of agroforestry products; and different types of coffee plots achieving the most desirable levels of ecosystem services.

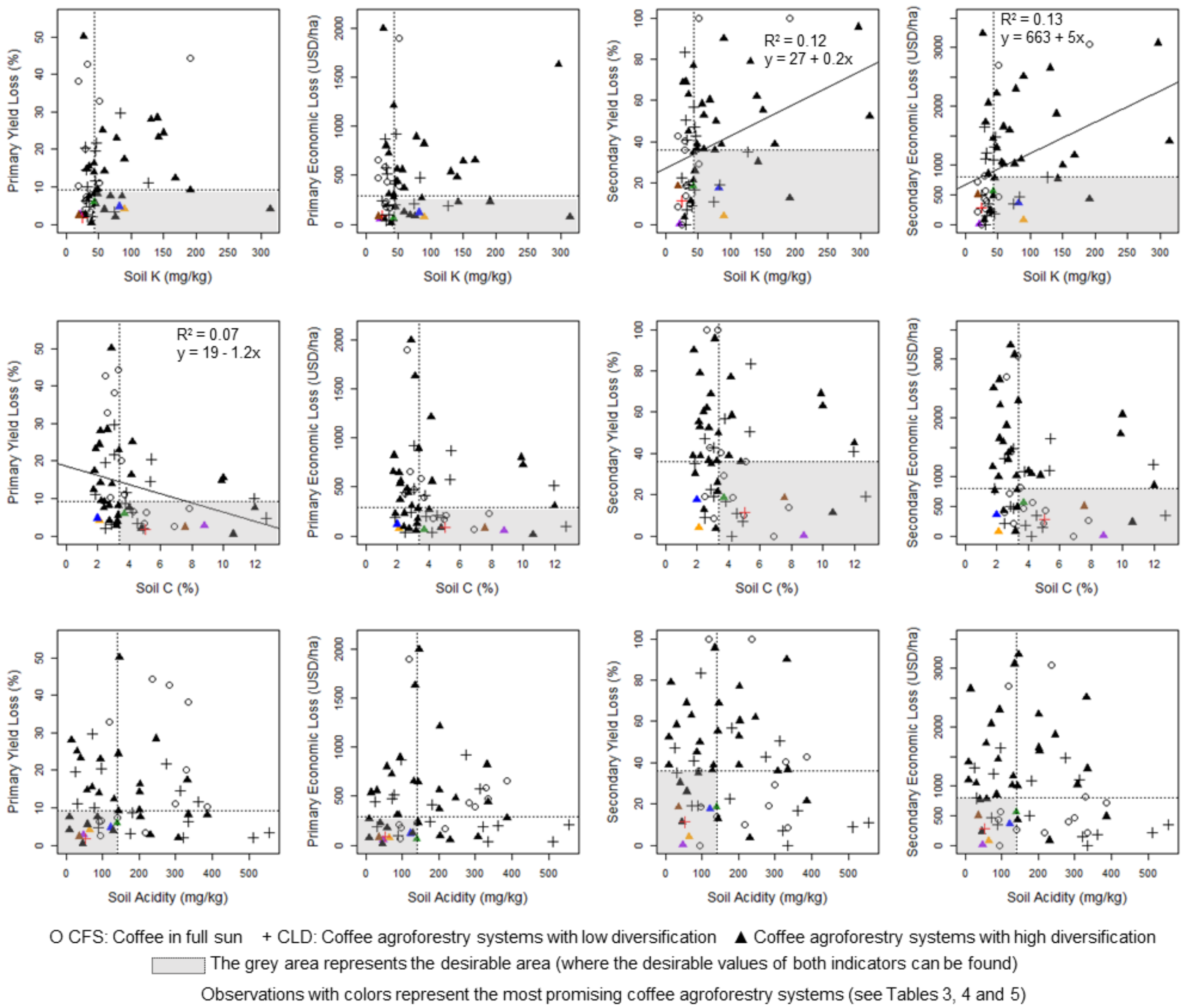

Fig. 6.3. Relationships between indicators of crop losses and indicators of maintenance of soil fertility; and different types of coffee plots achieving the most desirable levels of ecosystem services.

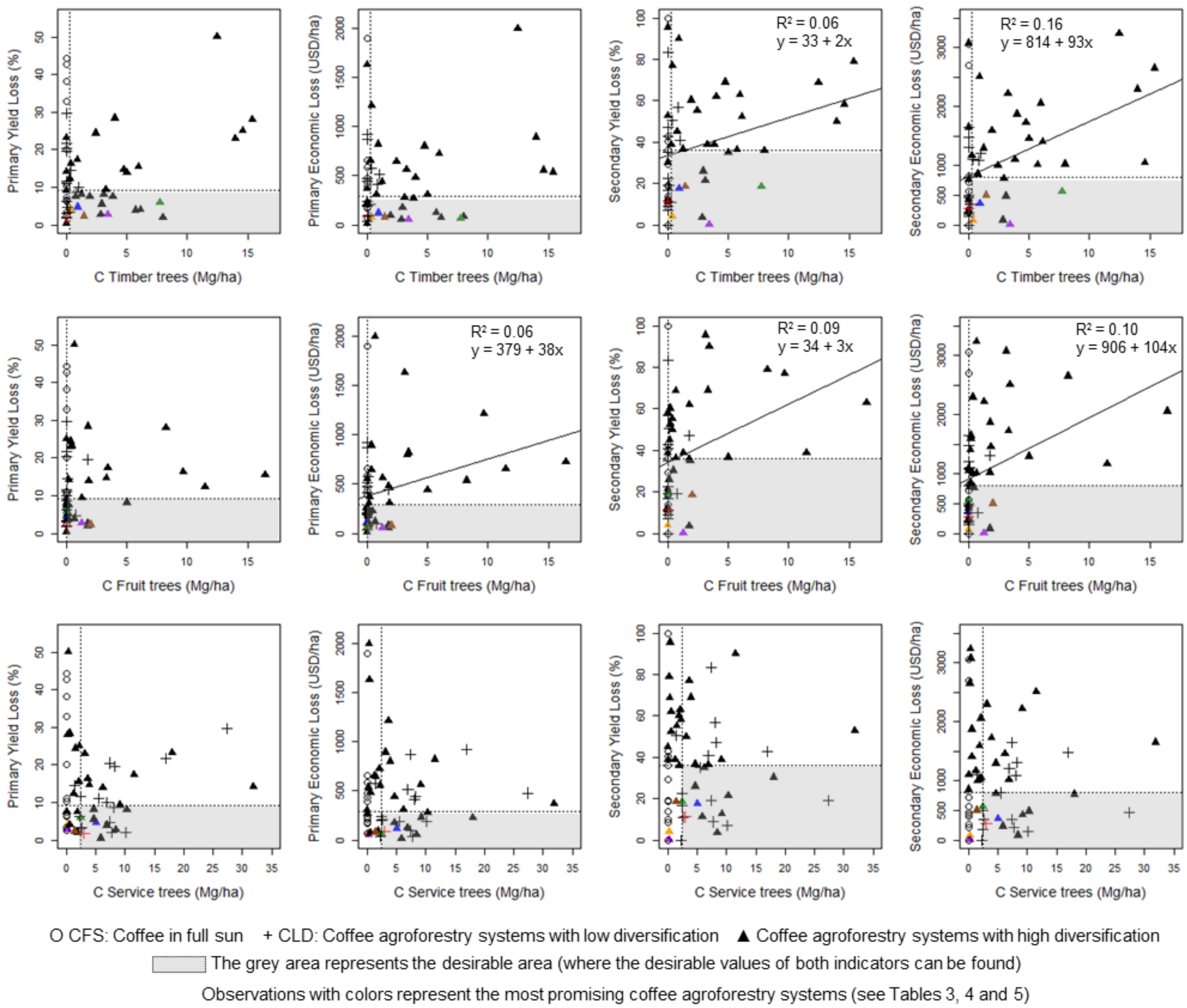

Fig. 6.4. Relationships between indicators of crop losses and indicators of carbon sequestration; and different types of coffee plots achieving the most desirable levels of ecosystem services.

3.2. Identification of the most promising coffee agroecosystems to provide ecosystem services

A total of six coffee agroforestry systems (CAF), a 10% of the 61 coffee plots studied, were identified as the most promising ones to reduce crop losses and provide other ecosystem services. They were numerated from 1 to 6 according to the complexity of their structure and composition (Table 6.3), and described as follow:

CAF1: simple coffee agroforestry system, basically a combination of coffee and service trees. This system has the higher density of coffee plants among the six most promising coffee agroforestry systems.

CAF2: medium diversified coffee agroforestry system, coffee-service trees-musaceaes-timber trees, where service trees dominate the shade canopy and timber trees are big and coarse according to their basal area.

CAF3: medium diversified coffee agroforestry systems, coffee-musaceaes-service trees-timber trees, where musaceas dominate in the shade canopy.

CAF4: medium diversified coffee agroforestry systems, coffee-musaceaes-service trees-timber trees, where musaceas dominate in the shade canopy. This CAF 4 is similar in plant densities than to CAF3, but has more than the double of basal areas of musaceas than CAF3.

CAF5: highly diversified coffee agroforestry systems, coffee-service trees-timber trees-fruit trees-musaceas, where service trees dominate in the shade canopy.

CAF6: highly diversified coffee agroforestry systems, coffee-service trees-timber trees-fruit trees-musaceas, where timber trees dominate in the shade canopy.

The management of the most promising coffee agroforestry systems mostly differed in the number of fungicide and herbicide applications, and in both cost of hired labor and cost of inputs. In general, in 2014 more practices and higher costs were applied than in 2015 (Table 6.4).

Finally, values of the indicators of the ecosystem services that the six most promising coffee agroforestry systems can provide, and the percentage of desirable areas in which each of them was located, are shown in Table 6.5.

Table 6.3. Characteristics of the structure, diversity and shade cover of the most promising coffee agroforestry systems (CAF) to provide multiple ecosystem services

Characteristics	**CAF1**	**CAF2**	**CAF3**	**CAF4**	**CAF5**	**CAF6**
Color for being identified in Figs. 1-4	red	green	orange	blue	brown	purple
Size of coffee plantation						
Plot area (ha)	0.22	0.40	0.37	0.22	1.40	1.40
Densities of coffee plants						
Density of coffee plants (individuals ha^{-1})	5079	3358	3864	4341	4480	4652
Distances between coffee rows (cm)	175	204	200	199	196	193
Distances between coffee plants (cm)	113	146	129	116	114	111
Densities in the shade canopy						
Density of fruit trees (individuals ha^{-1})	0	0	0	0	80	70
Density of timber trees (individuals ha^{-1})	0	30	10	20	30	170
Density of *Musaceae* plants (individuals ha^{-1})	10	60	260	300	20	20
Density of service trees (individuals ha^{-1})	340	280	20	30	180	120
Total density in shade canopy (individuals ha^{-1})	350	370	290	350	310	380
Basal (BA) areas in the shade canopy						
BA of fruit trees (m^2 ha^{-1})	0.00	0.00	0.00	0.00	0.87	0.60
BA of timber trees (m^2 ha^{-1})	0.00	5.53	0.29	0.71	1.11	2.67
BA of *Musaceae* plants (m^2 ha^{-1})	0.05	0.66	3.32	8.83	0.17	0.22
BA of area of service trees (m^2 ha^{-1})	6.52	6.43	0.12	5.16	1.27	0.61
Total BA in shade canopy (m^2 ha^{-1})	6.58	12.62	3.72	14.70	3.42	4.10
Species richness						
Species richness	2	4	6	5	8	7
Shannon index	0.13	0.81	0.97	0.61	1.43	1.40
Shade cover						
Shade cover in 2014 (%)	6	34	7	20	30	20
Shade cover in 2015 (%)	7	29	8	10	23	16

Musaceae plants: include mainly bananas but also plantains; Service trees: include mainly poro trees (Erythrina poeppigiana); Fruit trees: include mainly oranges (Citrus sinensis) and mandarin lemons (Citrus aurantifolia), but also avocados (Persea americana), cas (Psidium friedrichsthalium), arazá (Eugenia stipitata) and peach palm (Bactris gasipaes); Timber trees: include mainly Cordia alliodora and Cedrela odorata

Table 6.4. Characteristics of cropping practices and costs for the management of coffee plants in the most promising coffee agroforestry systems (CAF) to provide multiple ecosystem services

Characteristics	**CAF1**	**CAF2**	**CAF3**	**CAF4**	**CAF5**	**CAF6**
Color for being identified in Figs. 1-4	red	green	orange	blue	brown	purple
Cropping practices in 2014						
Machete weeding (number yr^{-1})	2	0	3	1	1	1
Harvests of coffee (number yr^{-1})	12	10	8	12	13	13
Applications of fertilizers (number yr^{-1})	2	1	1	2	0	0
Applications of fungicides (number yr^{-1})	6	4	1	1	5	1
Applications of herbicides (number yr^{-1})	2	3	1	1	2	2
Pruning of coffee plants (number yr^{-1})	1	1	0	0	1	1
Cash costs in 2014						
Cost of hired labor (USD ha^{-1})	0	465	907	2512	226	226
Cost of inputs (USD ha^{-1})	624	391	245	682	253	42
Total cash cost (USD ha^{-1})	624	856	1152	3194	479	268
Cropping practices in 2015						
Machete weeding (number yr^{-1})	1	0	1	3	1	1
Harvests of coffee (number yr^{-1})	12	9	12	10	12	12
Applications of fertilizers (number yr^{-1})	1	1	2	1	0	0
Applications of fungicides (number yr^{-1})	6	4	1	1	6	2
Applications of herbicides (number yr^{-1})	2	2	1	0	2	2
Pruning of coffee plants (number yr^{-1})	1	1	1	1	1	1
Cash costs in 2015						
Cost of hired labor (USD ha^{-1})	0	418	0	0	121	121
Cost of inputs (USD ha^{-1})	626	526	353	283	277	62
Total cash cost (USD ha^{-1})	626	943	353	283	399	184

Table 6.5. Indicators of ecosystem services of the most promising coffee agroforestry systems (CAF) to provide multiple ecosystem services

Indicators of ecosystem services	**Boundary value* (median)**	**CAF1**	**CAF2**	**CAF3**	**CAF4**	**CAF5**	**CAF6**
Color for being identified in Figs. 1-4		red	green	orange	blue	brown	purple
Regulation of diseases in 2014							
Primary yield loss (%)	9	2	6	4	5	2	3
Primary economic loss (USD ha^{-1})	291	90	66	76	120	79	55
sAUDPC Coffee leaf rust (%)	-	36	45	55	63	24	20
sAUDPC Brown eye spot (%)	-	7	5	3	1	6	13
Maximum index of dieback (%)	-	34	23	40	34	41	32
Provisioning of agroforestry products in 2014							
Coffee yield (kg ha^{-1})	6174	11185	2257	3742	5137	7189	4241
Cash Flow (USD ha^{-1})	1645	4673	213	620	2400	2926	1740
Value DC (USD ha^{-1})	47	85	70	419	256	70	70
Regulation of diseases in 2015							
Secondary yield loss (%)	36	11	18	4	17	18	0
Secondary economic loss (USD ha^{-1})	807	284	564	70	357	504	0
sAUDPC Coffee leaf rust (%)	-	21	41	45	48	19	23
sAUDPC Brow eye spot (%)	-	15	14	5	9	14	16
Maximum index of dieback (%)	-	40	43	49	50	31	46
Provisioning of agroforestry products in 2015							
Coffee yield (kg ha^{-1})	712	6481	8892	4056	1926	6347	917
Cash Flow (USD ha^{-1})	-21	1657	2190	1076	396	1838	139
Value DC (USD ha^{-1})	8	413	68	37	331	68	68
Maintenance of soil fertility							
Potassium (mg ha^{-1})	42	25	43	90	82	20	21
Carbon (%)	3	5	4	2	2	8	9
Acidity (mg ha^{-1})	139	53	139	65	122	36	47
Carbon sequestration							
Carbon in fruit trees (Mg ha^{-1})	0	0	0	0	0	2.0	1.3
Carbon in timber trees (Mg ha^{-1})	0.3	0	7.8	0.4	0.9	1.5	3.4
Carbon in service trees (Mg ha^{-1})	2.4	3.0	2.5	0.2	5.1	1.4	0.3
Percentage of desirable areas (out of 48) in which the CAF were located (%)		75	67	67	79	83	79

*The boundary values used to determine desirable areas in this study (See Figs. 1-4) were the medians of ecosystem service indicators, which meant that the desirable levels of ecosystem services indicators were those located at least within the 50% of the best levels.
sAUDPC: standardized area under the disease progress curve

4. Discussion

4.1. The usefulness of the analysis of relationships between indicators of ecosystem services

The analysis of the relationships between crop losses and indicators of ecosystems services, and the determination of desirable areas demonstrated to be useful, whether the relationships resulted significant or not. On one hand, the analysis permitted to identify trade-offs that, although their R^2 were not strong, become important warnings to avoid crop losses. For instance, the finding of slight trade-offs between carbon in trees and crop losses, warns that excessive presence of fruit and timber trees should be avoided. In the case of relationships which could reflect synergies, increasing the provision of a specific ecosystem service, such as soil C, could help to reduce crop losses, i.e. not necessarily by regulating diseases but by reducing their impact on yield (Avelino *et al.*, 2006). On the other hand, if no significant relationships were found, that means that a given ecosystem service can be increased without increasing (but without reducing neither) crop losses. Independently of the significance or type of relationship between ecosystem service indicators and crop losses, the desirable area permits to identify which type of agroecosystems are achieving the most desirable levels of ecosystem services (Rapidel *et al.*, 2015).

Results on the relationships between diseases and crop losses were useful, on one hand, to reveal damage and loss functions, and in the other hand, that it is possible to live with certain levels of diseases without incurring necessarily in high losses. These were novel findings demonstrated with data. The damage function is the relationship between injury level (caused by a pest or disease) and yield loss, and the loss function is the relationship between injury level and economic loss (Zadoks, 1985). For instance, per each increased unit of dieback index, the secondary yield loss increased in 1.2%; and per each increased unit of coffee leaf rust (sAUDPC), the secondary economic loss increased in 32 USD ha^{-1} (Fig. 6.1). Apart from these relationships, in the desirable areas it was also possible to note the presence of coffee plots which despite high levels of coffee leaf rust and dieback, registered low coffee losses. This offers the possibility to explore the rationalization of the use of pesticides by not looking at reduce pest incidences at the minimum. Both findings became an important contribution for crop loss research, given their complexity to be assessed (Savary and Willocquet, 2014).

Some relationships, such as the ones involving domestic consumption and soil K, should be interpreted carefully. The positive relationship between the value of domestic consumption of agroforestry products and secondary losses of coffee could be seen as an undesirable relationship (trade-off). However, a high domestic consumption of agroforestry products is more probably the result of high secondary losses, becoming an important alternative resource for families when coffee yield is low. Farmers use agroforestry products (fruits, timber and others) different to the product of the main crop in order to increase the self-consumption of in-farm products, increase incomes, and therefore reduce financial risk (Ramírez *et al.*, 2001; Rice, 2011). The slight positive relationship between soil K and secondary losses could suggest that it should be preferable to have low K content in soil, which is not actually correct. It would be better to interpret that extremely high contents of K in soil should be avoided.

4.2. Detailed recommendations derived from the characteristics of the most promising coffee agroforestry systems

The characteristics of structure of shade canopy and management of the six most promising coffee agroforestry systems (Tables 6.3 and 6.4) represent by themselves recommendations that users could follow to improve other coffee systems. However, it is important to highlight some of them and warn about others to avoid misinterpretations, based on the range of characteristics of such six coffee agroforestry systems:

About coffee densities: coffee plant density should not surpass 5000 plants ha^{-1}, and should maintain at least 2 m between coffee rows. This latter is important to avoid high incidences of pests and diseases, also demonstrated in a previous study (Cerda *et al.*, 2016b, Chapter 5).

About service trees: they could be managed with densities up to 350 trees ha^{-1} in simple agroforestry systems; in systems were bananas are important, then service trees should not surpass 30 trees ha^{-1}; and in diversified agroforestry systems, service trees could reach up to 200 trees ha^{-1}. It is important to say that we are referring to service trees of the species *Erythrina poeppigiana* or similar species, which are easily pruned to maintain low shade cover with crows of low height. If the user is planning to manage different service trees, for instance *Inga spp,* then the densities should be much lower.

About musaceae plants: they could reach up to 300 plants ha^{-1} in medium diversified agroforestry systems, but much lower (~30 plants ha^{-1}) in highly diversified agroforestry systems.

About fruit trees: in CAF 5 and CAF 6 they reached up 80 trees ha^{-1}, but they were small trees according to their basal areas, they were mainly fruit trees of the types *cas (Psidium friedrichsthalium)* and *arazá (Eugenia stipitata)* which have low shade cover. If the farmer wants to use fruit trees like oranges or avocados, which have denser crowns, then we believe that density should be between 20 and 30 trees ha^{-1} the maximum.

About timber trees: they should not surpass 30 trees ha^{-1} when they are trees similar to *Cordia alliodora* or *Cedrela odorata* as found in this study. One of the CAF had 170 trees ha^{-1} but these were very young and small according to their basal areas; farmer surely will eliminate many of them across years. Low densities of timber trees can be managed in agroforestry systems concentrating the sylvicultural management in the thickness of the trunk to sequester carbon and/or produce timber (Somarriba *et al.*, 2013).

About shade cover: shade cover was very variable among the most promising coffee agroforestry systems, but none of them surpassed the 35%. This should be the maximum percentage of shade cover to manage in a climate such as the one of Turrialba: rainy most part of the year, with mean annual rainfall = 2781 mm.

About management: control of weeds (herbicides and machete weeding) are applied when necessary, however, at least three controls per year will be ideal. At least one application of fertilizers per year is needed, better if two applications are made. Harvests should be every two weeks in an area like Turrialba were ripe fruits can be seen frequently during the six months of the harvest season. The application of fungicides per year was very variable among the most promising agroforestry systems, from one to six; in case of small farmers we believe the ideal is two applications per year, and could be three or four if it is noticed an extremely high incidence of diseases accompanied by defoliation. More than four applications of fungicides can be justifiable, but is not always feasible for small farmers due to the investment needed.

4.3. How to conduct other coffee agroecosystems towards being similar than the most promising coffee agroforestry systems

The six most promising coffee agroforestry systems, which were different in terms of structure-composition and management, represent several and varied options that farmers could choose for the design of new plantations, or for transforming the current characteristics of existing plantations to similar characteristics. It is known that farmers do not apply radical changes to their systems when they want to do improvements in the structure or management, they will apply changes step by step according to their objectives and resources (Mussak and Laarman, 1989). Therefore, one way to encourage farmers to do improvements in order to generate multiple ecosystem services, would be to identify which type of coffee plantation they have, and then to identify which type of the most promising agroforestry systems they want to move to. For instance, monocultures in full sun could more easily move to simple agroforestry systems coffee-service trees than to diversified agroforestry systems. By taking into account the main types of coffee agroecosystems in tropical areas (Toledo and Moguel, 2012), in Fig. 6.5, for each of them, we show which would be the most suitable promising coffee agroforestry system to move to.

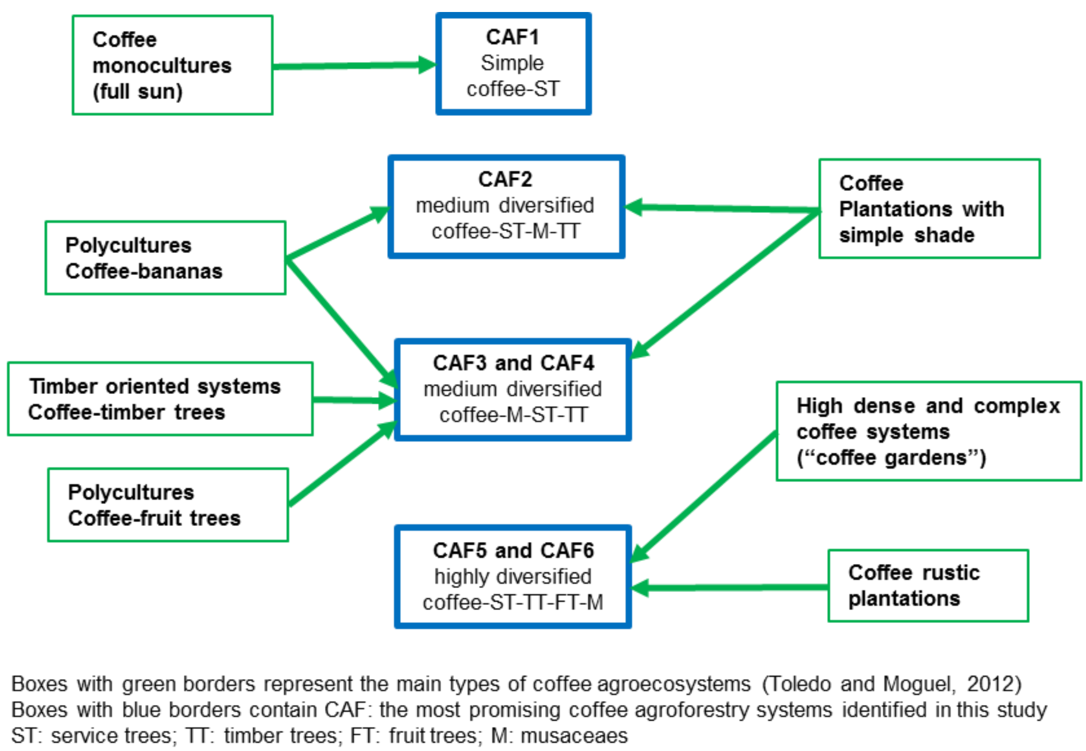

Fig. 6.5. Options for the main types of coffee agroecosystems in the tropics to move to one of the most promising agroforestry systems identified in this study

4.4. Limitation

The boundaries (medians) to demarcate the desirable areas used in this study are not generalizable for studies on coffee in other areas. In this study, the boundaries are dependent both on the indicators of ecosystem services of the sampled coffee plots and on the number of coffee plots. If similar studies were developed in other areas, their boundaries should be the medians registered in the very study. Alternatively, the boundaries could be established according to the specific boundaries desired by the user (according to particular objectives of farmers or researchers), as also stated in a previous study (Cerda *et al.*, 2017). To define boundaries for a given whole region, for instance, the sampled plots should represent a huge variability of production situations and management strategies within the studied region, avoiding replications of similar plots to keep the same weight by situation; that way, the medians obtained could be considered as "threshold boundaries" for the region.

4.5. Prospects

In this study, the analysis was focused mainly in crop losses as an indicator of the regulation of diseases, and on their bivariate relationships with indicators of other ecosystem services. This means that more weight was put on the service of regulation of diseases for the identification of the most promising agroforestry systems. If the same importance were wanted for each ecosystem service, then studies with multidimensional (multivariate) analysis of ecosystem services would be needed, which would be a major challenge taking into account that there are several indicators per each type of ecosystem service.

5. Conclusions

This study allowed us to identify the most important relationships between indicators of crop losses and indicators of other ecosystem services, which gives lights for a better management of coffee agroecosystems. The control of diseases must be concentrated on reducing coffee leaf rust and dieback, as they were the main diseases that increased crop losses. The increasing carbon in timber and fruit trees implied slight trade-offs, because such increment also increased crop losses, indicating that densities of these types of trees, for caution, should be low. The finding that increasing carbon in soil decreased primary losses, indicates that it will worth to do efforts to maintain or increase organic matter in the system.

The six coffee plots identified as the most promising ones to reduce crop losses and provide multiple ecosystem services simultaneously, belonged to different types of agroforestry systems and management strategies. This is an important finding for farmers, technicians and researchers, because they have several options to choose (imitate) for the design of new plantations, or for transforming the current characteristics of existing plantations to similar characteristics of one of the six most promising coffee agroforestry systems.

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PART IV. GENERAL DISCUSSION

Chapter 7. General discussion

This general discussion has four objectives: i) highlight the main scientific outputs to respond the three main questions of this Ph.D. Thesis, discussing also methodological points (7.1); ii) discuss conceptual contributions in order to guide the assessment of crop losses in relation with ecosystem services in perennial crops (7.2); iii) summarize the implications of the Thesis findings for the design and management of tropical agroecosystems with perennial crops (7.3); and iv) expose the prospects for further research (7.4).

7.1 Scientific contributions of this Ph.D. Thesis in the fields of crop losses research and assessment of ecosystem services

This Ph.D. Thesis provides both novel contributions and confirmation of important findings in previous studies, for the research on crop losses and ecosystem services in perennial crops. Such contributions are highlighted and discussed along with methodological points in the following three subsections, from 7.1.1 to 7.1.3, as responses to the three main research questions of the Thesis, from Q1 to Q3, respectively [Q1: What is the impact of management strategies and production situations on pests and diseases and coffee yields?; Q2: How do coffee yield losses caused by injury profiles vary in function of management strategies and production situations?; Q3: Which types of coffee agroecosystems are capable to obtain the lowest coffee losses (yield and economic) and highest overall benefits (ecosystem services)?].

7.1.1 Both production situations and management strategies determine coffee yield and pest and disease injuries

The effects of shade, altitude and management on pest and disease regulation were variable across different pests and diseases (Chapter 3). In other study such factors were also demonstrated to affect individual diseases such as coffee leaf rust (Avelino *et al.*, 2006) and American leaf spot (Avelino *et al.*, 2007). In a recent previous study, the importance of these factors for the development of injury profiles was also demonstrated (Allinne *et al.*, 2016). In our study, the importance of such factors was confirmed, but additionally, it was demonstrated, by using inferential statistics, that the epidemic risk of coffee leaf rust, the most serious disease nowadays, was the result of the interaction altitude x management. This result suggests that in order to improve coffee leaf rust control, by applying methods adapted to the risk and to the management strategy employed by the farmer, this interaction needs to be taken into account. We also demonstrated that dead productive branches (a yield reducing factor) depended on the triple interaction altitude x management x type of shade (Chapter 3). Other diseases such as brown eye spot, and pests such as coffee leaf miner depended only on individual effects of altitude and management.

There were no effects of types of shade on coffee yields, only management intensity was the most important factor affecting coffee yields (Chapters 3 and 5). This was an unexpected result. The types of shade were derived from cluster analysis involving the structure (densities and basal areas of shade trees), composition (species richness) and shade cover of the shade canopies. However, the shade cover alone did have a positive and significant effect in the models to estimate coffee yields (Chapter 5). Therefore, on one hand, it can be deduced that shade cover is probably more important than the structure and composition of shade trees in agroforestry systems. On the other hand, the coffee yields in the study area were not as high as the ones in other studies (Avelino *et al.*, 2006; Allinne *et al.*, 2016) where shade type partially explained coffee rust intensity. These low yields were probably not propitious to highlight a significant effect of types of shade

7.1.2 In perennial crops such as coffee, injury profiles affect yield losses not only during the same year but also during the following year(s)

Injury profiles had impacts on yield losses especially in a year of high coffee production (primary yield losses), but compromising also the yields of the next year (secondary yield losses). This was a novel finding documented with data, demonstrating also that secondary losses were higher than primary yield losses (Chapters 4 and 5). This supports hypotheses regarding the impacts of the severe coffee rust epidemic which occurred in 2012 in Central America (Avelino *et al.*, 2015). Furthermore, yields were possibly impacted more than one year after the damage occurred. However, these delayed impacts were not assessed in our trial. This was observed in the production trends after the year 2012, when the epidemic of coffee leaf rust began. There were actually two consecutive years of low production (2013, and 2014), and then production started to recover just in 2015 (Chapter 4, Fig. 4.5). Knowing that fruit load is positively related to coffee rust incidence and severity (López-Bravo *et al.*, 2012), it can be stated that a specific coffee rust epidemic has polyetic effects, i.e. affects the development of several consecutive epidemics. Furthermore, in our research, we found polyetic effects with injury profiles, not necessarily related to the physiology of coffee plants nor with the topoclimate (Chapter 5). This is a novel finding, revealing that polyetic effects influence the development of injury profiles, and that they can be the consequence of both the physiology of coffee plants and the accumulation of inoculum in the plantation.

The gradients of injury profiles (from slight to highly severe profiles), depended on particular combinations of production situations, management strategies and physiological characteristics of coffee plants (Chapter 5). Some previous studies reported this dependences before, in rice (Savary *et al.*, 2000b), in wheat (Willocquet *et al.*, 2002), and recently in coffee (Allinne *et al.*, 2016). In this research, we confirm such dependencies for coffee, but adding also a novel finding that gradients of injury profiles (from slight to highly severe profiles) were also positively related to gradients of both primary and secondary yield losses (from low to high yield losses); in other words, both injury profiles and their resulting yield losses are dependent on the same drivers of productions situations and management strategies. For instance, in the range of conditions of this study (Turrialba, Costa Rica), it was deduced that increasing fruit loads in old coffee plants, increasing contents of nutrients in coffee leaves, decreasing altitudes, and suboptimal management were the main drivers leading to the most harmful injury profiles, and therefore to the highest yield losses.

The first major challenge we had to address to achieve such findings was the estimation of primary and secondary yield losses through experimentation and modeling, and then, the analysis with several multivariate techniques to identify relationships (associations) among such yield losses, injury profiles, production situations and management strategies. The methods used are discussed regarding their usefulness and limitations.

Our experimental design (Chapter 4) followed most of the traditional recommendations but we added innovations for the estimation of primary and secondary yield losses in a perennial crop. The calculation of yield reduction can be done by comparing yields between plots treated and untreated with pesticides. The experiments must have an adequate design to permit statistical analysis, and they should be repeated in several seasons (Teng, 1990; Cooke, 2006). Our innovations were: i) the generation of different sequences of chemical treatments during several years, which were the studied statistical treatments (with replications), allowing to estimate primary and secondary yield losses, individually or together, only by direct comparison of mean yields; ii) the inclusion of the plot (characteristics of the location) and year as random effects in the statistical analysis, which demonstrated the importance of these effects in perennial crops to improve the quality of the estimations; iii) the measurement of not only symptoms/signs of pests and diseases, but also lost organs that could reflect a mechanism leading to yield losses, such as the dead productive branches.

Nonetheless, an improvement to the experimental design (Chapter 4) to calculate yield losses can be proposed. It was actually found that the sum of primary and secondary losses individually assessed was similar but not equal to the sum of primary and secondary losses in treatments where both were cumulated and assessed together. The reason, we believe, is that after the first year of the trial, coffee trees are not totally equal anymore, since injury profiles developed on unprotected coffee trees will influence plant physiological aspects and subsequent injury profiles (polyetic effects), which adds a factor of variation leading to not exact results in the comparisons of treatments. The only way to avoid such variations is to, each year, subdivide each plot in two subplots, and have one of these subplots with fungicides and the other with no fungicides. This is a new step that can be applied in future experiments to assess yield losses in perennial crops.

Our models to estimate yield losses (Chapter 5) could, on one hand, be considered as statistical empirical models only, but on the other hand, they could also be considered as models that involve mechanistic links. Empirical models show the response of a given variable as a direct function of one or more variables (or components) of the system, without explaining what happened to obtain the given response. Mechanistic models follow not a direct but a circuitous route in which processes are assigned to the components of the system; thus, several equations are generated and finally integrated to obtain the response of a given variable, making possible to know the mechanisms that led to such response (Thornley and Johnson, 1990). For the modeling of yield losses, both empirical and mechanistic models can be used. There are detailed process-based mechanistic models that include agrophysiological variables, modifiers (k) and injury mechanisms, and also simplified mechanistic models based on the use of intercepted radiation and radiation use efficiency (Savary *et al.*, 2006b). Our models are simpler than the latter ones, and although they cannot be considered as mechanistic, they include the dead productive branches, which can be considered as indicators (predictors) of both primary and secondary yield losses.

Results of the modeling showed that shade cover do not affect yields in a year of high production, but it does, positively, in a year of low production. This is an important finding along with the fact that the types of shade were not associated with injury profiles, because it indicates that the composition and structure of shade canopies does not affect injury profiles nor yield losses, and the shade cover can help to obtain better yields in a year when low production is expected. This result can also be related with the biennial production cycle of coffee, which is more marked in coffee plantations at full sun exposure than in coffee agroforestry systems, because under shaded conditions the fruit load and dieback are better regulated (Avelino *et al.*, 1999; DaMatta, 2004). These positive findings regarding shade could be generalized with more certainty within the ranges of shade cover of the types of shade considered in this study, up to ~30% of shade cover, where shade would not be at least a limiting factor of yields. Higher shade cover ranges could lead to negative effects on pests and diseases, yields and yield losses.

The multivariate methods used to achieve the findings described above were inspired in previous studies which used correspondence analysis to relate different typologies and thus identify the main drivers of injury profiles or individual pests or diseases (Savary *et al.*, 1995; Savary *et al.*, 2000b; Avelino *et al.*, 2006; Allinne *et al.*, 2016). The usefulness of this holistic approach, consisting in the analysis of the influences of several production situations and management strategies at the same time, is confirmed in this study. But furthermore, in this study, we improved an additional intermediary step in the analysis, which was already proposed by Avelino *et al*. (2006) for the case of coffee rust, and extended it to the case of injury profiles: the maximization of the relationships between injury profiles and typologies representing production situations, management strategies and physiological characteristics of coffee plants. For that purpose, we built these typologies on the first two components of a PLS-DA model relating injury profiles with all the variables involved in the typologies to be built. This important additional step helped to preserve the useful information, i.e. the information (mainly specific variables) related to injury profiles, giving less weight to the information with no relationship with the injury profiles. It can be said that the typologies were discriminated according to the characteristics of the injury profiles. Despite the loss of information that the building of typologies implies, our procedure helped to save the most important information and to highlight production situations, management strategies and physiological characteristics of coffee plants related with injury profiles.

It is important to remember that another important indicator of crop losses, such as the loss of quality of the product due to pests and diseases, was not assessed in this study. In coffee, the quality of the product is very relevant. On one hand, pests and diseases can decrease the quality of coffee in different ways, for instance: direct impact on physical aspect of fruit/bean, and indirect impact on chemical content of beans through physiological disturbances; on the other hand, shade can increase the coffee quality, for instance, by its indirect impact on physical and chemical characteristics of beans through yield regulation (the higher the yield, the lower the quality, which is found mainly at full sun conditions) (Ribeyre and Avelino, 2012). Therefore, the assessment of quality loss and also their resulting economic losses certainly deserves to be addressed in future studies.

7.1.3 Diversified agroforestry systems have better chances to regulate pests and diseases and provide multiple ecosystem services simultaneously

Both low and highly diversified coffee agroforestry systems had better ability to provide ecosystem services (including pest and disease regulation) than coffee monocultures in full sun. This finding was first revealed in Chapter 3 where effects of altitude, shade and management on ecosystem services were assessed, and then confirmed in Chapter 6 where the most six promising coffee agroforestry systems were identified. In the results of Chapter 3, it was evident that more coffee plots representing coffee agroforestry systems were located in desirable areas of ecosystem service provision than coffee plots in full sun. In Chapter 6, five out of the six most promising coffee agroforestry systems had from medium to high diversification in the shade canopy. Several authors have already concluded in review articles or in research studies the high capacity of agroforestry systems to provide ecosystem services (Beer *et al.*, 1998a; Jose, 2009; Gómez-Delgado *et al.*, 2011a; Tscharntke *et al.*, 2011; Somarriba *et al.*, 2013). Others reviews also concluded that systems based on enhancing plant species diversity can contribute to a sustainable control of pests and diseases in several crops (Ratnadass *et al.*, 2012) and in coffee (Staver *et al.*, 2001). With the results of this research, it is confirmed that certain types of coffee agroforestry systems are capable to regulate incidences of pests and diseases; with the novel finding, based on two-year data analyses, that these systems are also capable to reduce yield and economic losses, and simultaneously provide other major ecosystem services such as provisioning of agroforestry products, maintenance of soil fertility and carbon sequestration.

Results of the analysis of relationships among indicators of ecosystem services also support that coffee agroforestry systems can provide multiple ecosystem services without trade-offs. In Chapter 3 we have shown that there were no trade-offs between indicators of four ecosystem services, nor between such indicators and biodiversity, regardless the type of shade; and in Chapter 6 we found few significant relationships denoting slight trade-offs, but with very low R^2, which indicates that more than actual trade-offs, they are warnings to avoid possible undesirable relationships. Some examples of that, were the positive relationships between crop losses and carbon in fruit trees and in timber trees ($R^2<0.16$), which just suggests that excessive densities of that type of trees should be avoided for caution. Trade-offs reported in agroforestry systems such as the ones between yields and carbon sequestration, yields and biodiversity (Wade *et al.*, 2010), and yields and regulation of diseases (López-Bravo *et al.*, 2012) did not happen in this research, at least in the ranges of production situations studied. In a recent study on relationships involving carbon sequestration, yields and family incomes in coffee agroforestry systems in Nicaragua, trade-offs were not found either (Pinoargote *et al.*, 2016). Although this latter study and the present research did not cover all existing variability of production situations, their results still become important findings to encourage the use of agroforestry systems. The fact that trade-offs were not found regardless the type of shade, means that management provides leeway for reaching good combinations of ecosystem services.

7.2 Conceptual models and definitions regarding crop losses adapted to perennial crops

Another important contribution of this Ph.D. Thesis were two conceptual models that we made available as guides for studies in perennial crops. We believe these models can be used not only in coffee, but adapted for other perennial crops. One of the models is useful to guide the assessment of crop losses in a perennial crop, explaining effects of yield components and yield reduction factors leading to primary and secondary yield losses across years (Fig. 4.1). The other model is useful to guide the assessment of relationships among different components, or to guide the assessment of effects of given components on others across years (Fig. 5.1). The construction of this model followed a detailed protocol for the conceptualization of components affecting a given agroecosystem, the establishment of components inside the system and possible relationships among them, and the outputs of the system (Lamanda *et al.*, 2012). In this latter conceptual model, we also shown associations (relationships) which were not tested in this research, which represents opportunities for further studies to contribute to the research field of crop losses and for the research field of interactions and outputs of agroforestry systems.

The definitions are crucial in the assessment of crop losses. Our work contributed with definitions more adapted to perennial crops, especially when these are managed in agroforestry systems. The most important concepts to which we contribute, based on our findings, are the production situation and the attainable yield, explained in the following paragraphs.

There are different points of view about including management within the production situation (Savary *et al.*, 2006b) or excluding it (Aubertot and Robin, 2013; Aouadi *et al.*, 2015). In our research management was composed by agricultural practices and inputs applied to the main crop (coffee). On one hand, in our results management intensity had several individual significant effects on important indicators of ecosystem services (Chapter 3), which indicates that management can change the provision of certain ecosystem services (e.g. coffee yields, family benefits, regulation of certain pests and diseases) regardless conditions of topoclimate or types of shade. On the other hand, in the integral analysis of associations of injury profiles with different typologies (soil, types of shade, topoclimate, physiological characteristics of coffee plants and management), we found that only one specific profile of management was associated with the injury profile with the lowest levels of injuries (Chapter 5, Fig. 5.7); and the typology of management of one year was the only one significantly associated with the injury profiles of the next year (Chapter 5, Table 5.5). These findings indicate that management by itself is capable to influence the development of injury profiles, their resulting yield losses, and the provision of ecosystem services. Given such importance of management, it is worth to keep it excluded from the definition of production situation. For instance, there can be two production systems in similar production situations (regarding topoclimate, soil fertility and type of shade), but the outputs of such systems (yields for instance) can be different depending on the management applied.

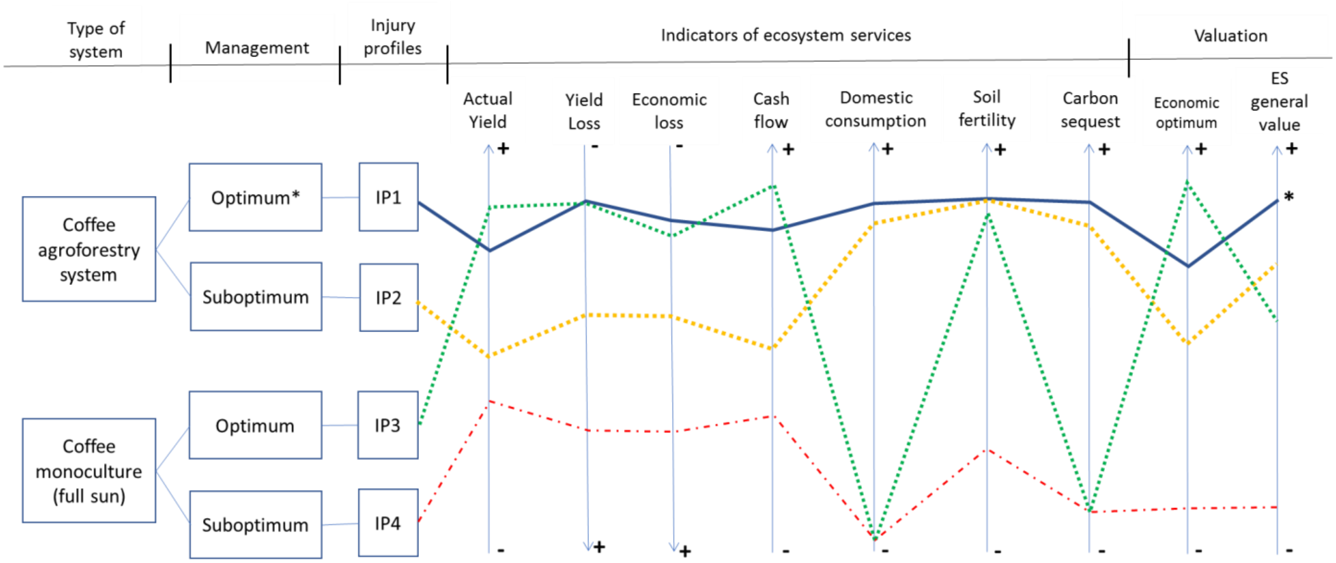
However, some characteristics of the agroecosystem which also result from human decisions and actions such as the type of shade, deserve to be maintained within the production situation concept. We propose that the structure and nature of shade are stable enough in time to be considered as an intrinsic characteristic of the production situation, as topoclimate and soil fertility, for the case of agroforestry systems.

Working with perennial crops incite us to re-examine the definition of attainable yield. By definition, an attainable yield is a yield achieved without the negative influence of pests and diseases (Nutter *et al.*, 1993; Oerke *et al.*, 1994). This seems easy to assess in an annual crop. However, in a perennial crop, where injuries profiles have polyetic effects (Chapter 5) and where yield is determined by the growth of the current year but also by the growth of previous years (DaMatta, 2004), the attainable yield cannot be assessed in only one specific year with no pests and diseases. We propose that attainable yield should be assessed at least after a period of a minimum of two years with no negative impacts (losses) caused by pests and diseases (Chapters 4 and 5). This period of two years is also the minimum period for considering the biennial rhythm of production of coffee.

7.3 Implications for the design and management of tropical agroecosystems oriented towards the reduction of crop losses and provision of multiple ecosystem services

As a broad summary, the technical recommendations derived from the results of this research, are oriented to promote agroforestry systems with medium to high diversification in the shade canopy and managed with a medium intensity, with the aim to reduce yield and economic losses of the main crop, and simultaneously provide goods for family benefits, improve or at least maintain the soil fertility, and increase carbon sequestration in aboveground biomass, without implying trade-offs among these ecosystem services. Our recommendations evolved from Chapter 3 to Chapter 6, from general recommendations based on the main findings to specific recommendations derived from the most promising coffee agroforestry systems identified in this research. In Chapter 3, there are general recommendations oriented to promote the provision of four ecosystem services (regulation of pests and diseases, provisioning of agroforestry products, maintenance of soil fertility, and carbon sequestration); in Chapter 5, the recommendations are oriented specifically to avoid severe injury profiles and therefore avoid high yield losses; and, in Chapter 6, the recommendations are detailed regarding the structure of shade canopies and management strategies, oriented towards both reduce crop losses and provide multiple ecosystem services simultaneously. Each of the mentioned Chapters have a short section in their discussion with practical recommendations supported also by literature, and therefore it is not worth to repeat all of them again here.

Our recommendations for improving the design and management of tropical agroecosystems can be applied to crops different from coffee such as cocoa, banana and others which are managed in agroforestry systems. Our recommendations for the design and management of the shade canopy are oriented to provide ecosystem services, but also to avoid undesirable interactions between shade trees and other components of the system (disservices); therefore, such recommendations should be suitable and of interest of farmers growing other crops in shaded systems. In Fig. 7.1, based on our findings, we show a conceptual scheme on the ecosystem services that can be achieved by successful agroforestry systems, in comparison with other agroecosystems.



Optimum management: represents a medium management intensity suitable for smallholder farmers (see recommendations in Chapters 3, 5 and 6)
Suboptimum management: represents a management with deficiencies especially in the control of pests and diseases
*Represents a coffee agroforestry system with the recommendations of structure, composition and management derived from the present research
The other combinations of types of system and management represents other types of coffee agroecosystems which can be found in coffee growing areas
Each combination of type of system and management will produce a particular injury profile and achieve particular levels of ecosystem services
Domestic consumption: agroforestry products including coffee, bananas, other fruits, and other goods (timber, materials) that the family can consume from the system
One-way arrows from (-) to (+) indicate the increase of the provision of a given ecosystem service indicator
Economic optimum: the maximum economic profit (the maximum relation between benefits and costs)
ES general value: represents a general value of the set of ecosystem services provided by the system

Fig. 7.1. Qualitative representation of the ecosystem services provided by successful agroforestry systems in comparison with other coffee agroecosystems.

Recommendations to improve the management of diversified agroecosystems should be based both on characteristics of target groups of agroecosystems and on characteristics of individual successful agroecosystems. When data is analyzed with contingency table and correspondence analysis, the variability of situations originated by each plantation, is limited to few combinations of production situations and injury profiles, which could be considered as different targets for management of pests and diseases (Savary *et al.*, 2016). We confirmed this in our correspondence analysis and we derived recommendations based on characteristics of associated profiles (Chapter 5). We think, however, that such recommendations could be sufficient for monocultures but not necessarily sufficient for mixed cropping systems (such as agroforestry systems). The variability of conditions in mixed cropping systems would be higher than in monocultures, because they have more biological components. For coffee agroecosystems, for instance, we showed great variability in their structure and management (Chapters 3 and 6). Therefore, we believe that it is also worth to identify the most successful cases within such variability, which become as individual targets to follow, as we did in Chapter 6. Such successful cases are not possible to identify in correspondence analysis. That way, findings obtained from both analytic approaches can be integrated to offer more precise and detailed recommendations for farmers.

In the scientific literature, there is an important question on “whether intensive or extensive agriculture best optimizes the various trade-offs associated with ecosystem services provision” that deserves research (Dale and Polasky, 2007; Pert *et al.*, 2013). The findings of this study, for the field of agroforestry, contribute with hints that intensive agroforestry systems (in terms of agricultural practices and inputs) can be better than extensive ones. It was shown that increasing management intensity index, in both lowly and highly diversified coffee agroforestry systems, increased the provisioning service indicators without causing negative effects on indicators of the other ecosystem services studied here; and that these shaded systems by themselves do not involve trade-offs between those ecosystem services (Chapter 3). Extensive agroforestry systems, which can be represented by low management intensity index, did not cause negative effects either, but their disadvantage would be the lower provisioning services; reflecting that way a lesser optimization than intensified agroforestry systems.

The dissemination of better agroforestry systems could derive in the provision of multiple ecosystem services as demonstrated in this research, which also implies better chances to face climate change. If farmers orient their plantations to produce several ecosystem services at the same time, then the adaptation of farming systems, farms and families to climate change would be also improved. The implementation of diversified agroforestry systems has already been suggested as adaptation measures of rural families' livelihoods, because, for instance, in the face of extreme negative events caused by climate change, the shade cover is useful to protect plants from strong winds, high temperatures and long dry periods (Schroth *et al.*, 2009; Jha *et al.*, 2014). Furthermore, given that the ecosystem services provided by agroforestry systems contribute to conservation of natural resources, adaptation and mitigation of climate change, and livelihoods security; the use of shade trees is also considered as one of the most promising ecosystem-based adaptation practice, especially for smallholder farmers (Vignola *et al.*, 2015).

7.4 Main prospects

Our results are applicable to production situations and management strategies similar to the ones of the study area of the present research, as stated in Chapters 3 and 5; studies in regions with wider ranges or with very contrasting characteristics are needed to corroborate the findings of this research. The approaches and methods used in this research demonstrated to be useful to accomplish the objectives, and could be applied not only in coffee agroecosystems, but also in agroecosystems with cocoa, bananas and other crops which are also managed in perennial plantations, and from perennial plantations in monocultures to perennial plantations in agroforestry systems. The most ideal would be to develop similar research on the assessment of crop losses and ecosystem services at regional levels, as it was done for crop losses in rice in tropical Asia (Savary *et al.*, 2000a; Savary *et al.*, 2000b). If it were possible, based on the experience of this research, the main recommendations would be: determine the most representative and contrasting areas of the crop production in the region, i.e. areas with different production situations (especially in terms of altitude, soil fertility and botanical structure/composition of the target agroecosystem) and management strategies (agricultural practices and inputs); sample plots well distributed in such areas for representative purposes; identify the most important indicators (predictors) of crop attainable and actual yields, and predictors of crop yield reduction, for which the PiecewiseSEM is recommended (Chapter 5); identify predictors of crop product quality and predictors of quality reduction to assess quality loss; identify and assess the most important ecosystems services for the region; and, perform measurements and work with data of two years (at least), especially in perennial crops which has a biennial production cycle and/or are affected by polyetic epidemics.

The findings of this research about factors affecting coffee pests and diseases, along with findings on the same topic in other studies, could be useful to develop an injury profile simulator for coffee agroecosystems. Our results provided the most important relationships between individual pests and diseases and variables representing management strategies and production situations, how different combinations of pests and diseases produce different injury profiles, and estimations of the yield losses that each injury profile can cause. Furthermore, there are also many other studies on coffee showing more relationships of pests and diseases with production situations and management (Soto-Pinto *et al.*, 2002; Avelino *et al.*, 2004; Barbosa *et al.*, 2004; Avelino *et al.*, 2006; Avelino *et al.*, 2007; Avelino *et al.*, 2009; Rocha *et al.*, 2010; López-Bravo *et al.*, 2012; Allinne *et al.*, 2016). Based on all of this information, it should be possible to construct a qualitative aggregative modelling framework to predict (simulate) injury profiles for coffee. This model can be constructed in the software DEXI (v.4.0), as already was constructed for wheat pests for instance (Robin *et al.*, 2013; Aubertot *et al.*, 2014). In this software, we can establish the scales and functions (rules) that are going to determine given levels of each pest or disease; then the rules to indicate what combinations of levels of pests and diseases injuries lead to given injury profiles; finally, each injury profile could be associated with an amount of yield loss.

The findings of this study could also be used for the prototyping of coffee agroforestry systems with the aim to optimize the provision of ecosystem services. The prototyping is a methodology to design sustainable farming systems which is possible to apply to both monocultures and mixed cropping systems (Sterk *et al.*, 2007; Rapidel *et al.*, 2015). This methodology can have variations in their application, but the main steps are: i) to define and hierarchize the objectives of the farming systems, ii) to identify the limitations for achieving the objectives, iii) to design prototypes of the desired systems, iv) to test and evaluate the prototype in field conditions, v) to disseminate the most successful prototypes (Lançon *et al.*, 2007; Sterk *et al.*, 2007; Blazy *et al.*, 2009; Rapidel *et al.*, 2009). Based on the relationships between crop losses and ecosystem services, and on the recommendations for shade and management derived from the most promising coffee agroforestry systems identified in this research (Chapter 6), several prototypes could be designed and tested in different conditions of topoclimate (altitudes, temperatures and rainfall). The prototypes could be improved based on the results of multidimensional relationship analysis integrating several ecosystem services at the same time. In this research the analysis of relationships between ecosystem services were only bivariate. The test of prototypes of coffee agroforestry systems would need big financial efforts, but stakes and expected impacts are high considering the multiple ecosystem services that could be generated in hand of coffee smallholders, as demonstrated in this study.

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