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Effect of intercropping on cassava bacterial blight caused by *Xanthomonas axonopodis* pv. *manihotis* in Togo

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ABSTRACT

Bacterial blight is one of the biotic constraints to cassava production, affecting yield and cuttings. To contribute to preventing farmers from yield losses, the present study investigated the effect of intercropping cassava with common staple crops in Togo on the development of cassava bacterial blight under field conditions in four agroecological zones of the country. Cassava was intercropped with maize (*Zea mays* L.) in the forest-savanna transition zone at Davié, in the forest lowland zone at Adéta, in the forest highland zone at Danyi, and in the wet savanna zone in Sotouboua. Additionally, cassava was intercropped with taro (*Colocasia esculenta* (L.) Schott.) in the forest highland zone at Danyi. Border plants of cassava for each treatment were inoculated with a bacterial suspension of 10^7 cfu/ml. Bacterial blight severity was significantly reduced compared to sole cassava in the forest highland in cassava-taro and cassava-maize intercropping ($p < 0.05$ and $p < 0.01$ in 2000 and 2001, respectively), in the forest lowland in cassava-maize intercropping ($p = 0.002$ in 2001), and in cassava-maize intercropping in the wet savanna zone ($p < 0.001$ and $p < 0.01$ in 2000 and 2001, respectively), with generally no significant negative effect on cassava root yield. Though significant, disease reductions by intercropping generally were low (10 to 24%). Since no varieties with complete resistance to the disease had been identified among local and local improved varieties across ecozones in Togo, the combination of medium resistant varieties and an intercropping system, both adapted to the respective ecozone, can be recommended to farmers as part of integrated control measures against bacterial blight of cassava.

Keywords: Cassava, bacterial blight, intercropping, control, Togo.

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INTRODUCTION

Cassava production is largely reduced due to the attack by pests and diseases (Zinsou et al., 2004; Fanou et al., 2017, 2018), with bacterial blight caused by *Xanthomonas axonopodis* pv. *manihotis* (Vauterin et al., 1995), former *X. campestris* pv. *manihotis* (Bondar, 1915), is one of the major constraints (Fanou et al., 2017). Symptoms include angular leaf spots, blighting, wilting, defoliation, vascular necrosis of the stem, exudation and dieback. The vascular symptoms affect the quality and quantity of planting material, while root yield losses due to bacterial blight of more than 50% (Wydra and Rudolph, 1999) and 77% in some cassava varieties in the dry savanna zone (Fanou et al., 2018) were reported. Due to the unstable and environmentally dependent nature of resistance to bacterial blight, an integrated control system combining host plant resistance with agronomic and cultural measures could significantly reduce bacterial blight epidemics since intercropping was described to reduce diseases in several crops (Zinsou et al., 2004; Wydra et al., 2007; Banito et al., 2008; Fanou et al., 2018).

Intercropping can affect disease and pest incidence and severity (Van Rheenan et al., 1981; Ofuya, 1991; Trenbath, 1993). Intercropping cassava with cowpea reduced egg populations of *Aleurotrachelus socialis* and *Trialeurodes variabilis*, compared to those in monoculture (Gold et al., 1990). The combination of improved genetic resistance with the benefits of intercropping should result in a more sustainable control of diseases and pests (Davis and Woolley, 1993). Larios and Moreno (1976, 1977) and Moreno (1979) observed incidence and severity reduction of mildew and anthracnose in intercropping cassava and common bean association in Costa Rica, while Ghosh et al. (1986) reported a reduction of brown leaf spots (*Cercosporidium henningsii*) in cassava associated with *Eucalyptus* sp. and *Leucaena* sp.

The use of intercropping was proposed as means to reduce cassava bacterial blight in the dry savanna and humid forest (Tabot, 1995). A significant reduction of cassava bacterial blight severity in cassava intercropped with cowpea and maize was observed in the forest-savanna transition zone of Nigeria (Fanou et al., 2018). To develop a control strategy against the bacterial blight of cassava reported in Togo (Banito et al., 2007) the present study aimed to evaluate the effectiveness of intercropping cassava with maize and taro in controlling cassava bacterial blight under field conditions in various agroecological zones in Togo.

MATERIALS AND METHODS

Experimental sites

Intercropping trials were conducted in four sites in stations of the Institut Togolais de Recherche Agronomique (ITRA), Togo, during the growing seasons 1999-2000 and 2000-2001: in the forest-savanna transition zone at Davié, in the forest lowland zone at Adéta, in the forest highland zone at Danyi, and the wet savanna zone in Sotouboua.

The vegetation in the forest-savanna transition zone (littoral zone) in the South part of the country is characterized by shrubby vegetation with few trees, in the forest zone in the South-West by rainforest vegetation, and in the wet savanna in the Center part of the country by more shrubby vegetation.

The forest-savanna transition and the forest zones are characterized by a sub-equatorial climate with one long rainy season (March – June), one short dry season (July - August), one short rainy season (September – October) and one long dry season (November – March), whereas the wet savanna is characterized by a tropical climate with one long rainy season (April – September) and one long dry season (October – March) (Lamouroux, 1979). The average annual rainfall is about 1,200 mm in the forest-savanna transition zone and 1,400 mm in the forest and wet savanna zones, with an average temperature of 28, 24 and 27°C, respectively (DMN, 2002).

Planting materials and experimental design

The bacterial blight-susceptible variety Ben86052 from IITA was used in this intercropping experiment. Cassava stem cuttings of 20 cm deriving from apparently healthy field plants were single planted at a spacing of 1 × 1 m on well-prepared flat ground in June 1999 and 2000 for the growing seasons 1999-2000 and 2000-2001, respectively. Each treatment consisted of three plots (20 m^2 per plot) of 20 plants.

Cassava was intercropped with maize (IKENNE variety) in a row intercropping system. Additionally in the forest highland, cassava was row-intercropped with taro, a common tuber crop in this area. Maize plants within rows were 40 cm spaced apart, while cassava and taro (local cultivar) were 1 m spaced. The three crops were planted at the same time. In this study, block design was not used due to the accessity of land. Each treatment was in three plots non-replicated. Weeding was conducted when necessary and no watering was applied.

Bacterial suspension and inoculation

X. axonopodis pv. *manihotis* strain X27 was grown on GYCA medium (glucose 5 g/l, yeast 5 g/l, $CaCO_3$ 10 g/L, agar 15 g/L) in Petri dishes. A 48-hour-old culture of the bacterial culture was harvested and suspended in 0.01 M $MgSO_4$ solution. One-month-old cassava plants were inoculated with a bacterial suspension of 10^7 cfu/ml by spraying the abaxial surface of cassava leaves with a motorized sprayer. For each of the treatments, only the border plants (outer rows of each plot) were inoculated. A total of three inoculations were performed at 3-weekly intervals.

Disease and yield assessment

Disease symptoms were evaluated 3 weeks after each inoculation and after six and twelve months, by counting leaves with angular leaf spots, blight or wilt on ten randomly selected rows between the inoculated outer rows of each. When leaves showed more than one symptom type, they were recorded under the more severe symptom type. The total remaining leaves, dropped leaves (number of scarifications remaining on the stem) and number of shoot tips with dieback were also recorded. The percentages of leaves with spots, blight, wilted/dropped leaves and shoots with dieback were calculated for each plant.

The severity index (Si) was calculated for each plant at each evaluation date as follows:

$$Si = (1xS + 2xB + 1xW + 2xD)/6$$

where S, B, W and D represent the percentage of leaves with spots, blight, wilt and shoots with dieback, respectively. The weight attributed to the symptoms of blight and dieback is an estimation resulting from regression analysis of symptom and plant growth data, revealing blight as the most important factor influencing root yield, and dieback with the highest influence on overall plant growth (leaf and stem weight) (unpublished data).

The effect of the cropping system on cassava bacterial blight severity was assessed by calculating the area under the severity index progress curve (AUSiPC) for each plant at six evaluation dates, by the trapezoidal integration (Shaner and Finney, 1977; Jeger and Viljanen-Rollinson, 2001) according to ecozones.

In the forest and forest-savanna transition zones:

$$AUSiPC = [(Si1+Si2)\times 21/2 + (Si2+Si3)\times 21/2 + (Si3+Si4)\times 60/2 + (Si5+Si6)\times 120/2]/275$$

In the wet savanna zone:

$$AUSiPC = [(Si1+Si2)\times 21/2 + (Si2+Si3)\times 21/2 + (Si3+Si4)\times 30/2 + (Si5+Si6)\times 90/2]/215$$

where Si1, Si2, Si3, Si4, Si5 and Si6 represent the severity index at the evaluation dates 1, 2, 3, 4, 5 and 6, respectively. Si4 and Si5 correspond to the severity index during the dry season and are equal to zero. The AUSiPC in days over the growing period was divided by the evaluation period of 275 or 215 days, corresponding to 365 days minus the dry season period of 90 days in the forest and forest-savanna transition zones and 150 days in the wet savanna zone to receive an average comparable between ecozones. Thus, all AUSiPC values are standardized.

Cassava roots were harvested 12 months after planting by uprooting ten plants randomly selected within rows between the inoculated outer rows of each plot. Plant height was measured, and the roots of each plant were counted and weighed. All the roots of each plot were mixed and a sub-sample was cut into small pieces, weighed and dried in an oven at 105°C for 72 h for dry weight determination.

Statistical analysis

Standardized area under severity index progress curve (AUSiPC) and dry root weight values were log-transformed to stabilize variances and the analysis was performed using the Linear Mixed Model ANOVA (Bernardo, 1994). Values and standard errors in tables are real, non-transformed values. Analysis of variance was performed on AUSiPC and root dry weight values using the General Linear Model (GLM) procedure in the SAS system (SAS, 1990; 1997). The Student-Newman-Keuls (SNK) test at a 5% level was used to compare the means of AUSiPC and root dry weight values (Danielie, 1975), and to discriminate between the intercropping patterns.

RESULTS

Effect of intercropping on cassava bacterial blight severity

Cassava bacterial blight severity expressed as area under severity index progress curve (AUSiPC) varied from 4.9 in cassava-taro intercropping in the forest highland to 8.1 in cassava monocropping in the forest lowland and the wet savanna zone. The lowest disease severity (4.9 to 5.2) of all ecozones and across the two years of the experiment. The cassava-maize intercropping recorded a disease severity of 5.2 to 7.5, while the single cassava treatments generally recorded the highest disease severity of up to 8.1 (Table 1). The AUSiPC was significantly reduced in the forest highland in cassava-taro and cassava-maize intercropping treatments ($p = 0.03$ and $p = 0.002$ in 2000 and 2001, respectively) compared to sole cassava, in the forest lowland in cassava-maize intercropping in one growing season ($p = 0.002$ in 2001), and in the wet savanna zone in cassava-maize intercropping in the inoculated treatment ($p < 0.0007$ and $p = 0.005$ in 2000 and 2001, respectively). However, the reduction of the disease severity in cassava-taro and cassava-maize intercropping did not differ significantly. Though significant in some treatments, disease severity reductions by intercropping were generally low (about 10 to 24%).

In the forest highland, the disease develops during the rainy season, with the highest values at 2 and 4 months after planting (Figure 1). Symptoms disappear during the dry season and reappear in the rainy season of the following year.

Table 1. Effect of intercropping cassava-maize and cassava-taro on cassava bacterial blight severity expressed as area under the severity index progress curve (AUSiPC) in four ecozones of Togo.

Crop system	Forest lowland		Forest highland (Plateaux)	
	2000	2001	2000	2001
Cassava	7.5[a]	8.1[a**]	5.8[a*]	6.4[a**]
Cassava-maize	7.5[a]	7.0[b]	5.2[b]	5.3[b]
Cassava-taro	nd	nd	5.2[b]	4.9[b]

Crop system	Forest savanna transition		Wet savanna	
	2000	2001	2000	2001
Cassava	6.3[a]	7.5[a]	7.8[a***]	8.1[a**]
Cassava-maize	6.2[a]	7.2[a]	6.0[b]	7.3[b]

* = significant (SNK test) at probability level p < 0.05; ** = significant at probability level p < 0.01;
*** = significant at probability level p < 0.001; nd = not done.

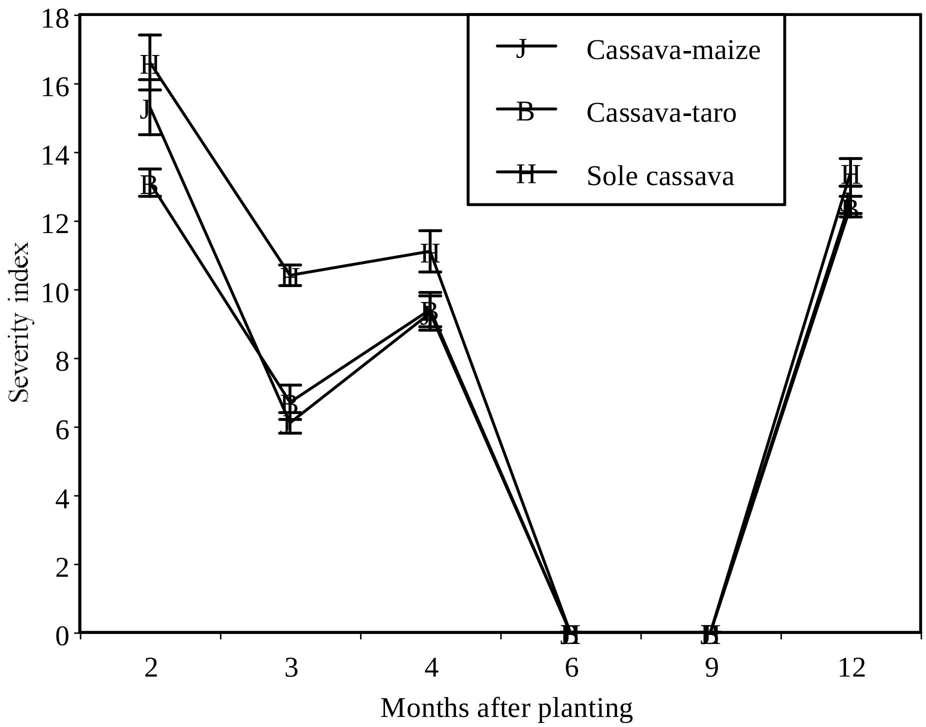

Figure 1. Example of the development of severity index in intercropping patterns in the susceptible genotype Ben86052 in the inoculated treatment in the forest highland in growing season 1999-2000 (dates of inoculation: 30, 51 and 72 days after planting).

Effect of intercropping on cassava yield

Generally, no significant differences in yield (root dry weight) were observed between cassava monoculture and cassava intercropping plots across environments and treatments, except in the forest-savanna transition zones, where intercropping cassava-maize, significantly reduced root yield in 2000 and 2001 (p = 0.007 and p = 0.02, respectively) (Table 2).

DISCUSSION

The effect of intercropping cassava-maize and cassava-taro on cassava bacterial blight was investigated. Significant, but relatively low reductions of cassava bacterial blight severity (AUSiPC) were observed in cassava-maize intercropping in the wet savanna zone for the two years of experiments, in cassava-maize intercropping in the forest lowland in 2001, and also in

Table 2. Effect of intercropping on cassava yield (root dry weight in t/ha) in four ecozones of Togo.

Crop system	Forest lowland		Forest highland (Plateaux)	
	2000	2001	2000	2001
Cassava	26.1[a]	25.0[a]	19.5[a]	21.9[a]
Cassava-maize	23.1[a]	24.4[a]	15.3[a]	16.9[a]
Taro-Cassava	nd	nd	20.0[a]	19.5[a]

Crop system	Forest savanna transition		Wet savanna	
	2000	2001	2000	2001
Cassava	26.3[a]**	21.1[a]*	17.3[a]	15.6[a]
Cassava-maize	15.8[b]	14.6[b]	12.4[a]	13.4[a]

* = significant (SNK test) at probability level $p < 0.05$; ** = significant at probability level $p < 0.01$; nd = not done.

cassava-maize and cassava-taro intercropping in the forest highland zone. Intercropping had been reported to influence disease incidence and severity (Van Rheenan et al., 1981; Ofuya, 1991; Davis and Woolley, 1993). A reduction of bacterial blight incidence and severity in cassava intercropped with maize (Moreno, 1979), with melon Tabot (1995) has been reported.

Also, Sikirou (1999) did not observe clear effects on cowpea bacterial blight when cowpea was intercropped with maize or cassava in the forest-savanna transition zone of West Africa. Similar studies reported a significant reduction of bacterial blight of cassava severity by intercropping cassava with sorghum and also with cowpea (Zinsou et al., 2004). Cassava bacterial blight reduction due to cassava-maize and cassava-cowpea intercrops in the forest-savanna transition zone and cassava-maize intercrop in the dry savanna zone in Nigeria was reported by Fanou et al. (2018) Intercropping system was to reduce potato bacterial wilt and tomato bacterial wilt caused by *Ralstonia solanacearum* (Autrique and Potts, 1987; Kloos et al., 1987; Michel et al., 1997). The reduction of the disease severity observed in intercropping might be due to the barriers provided by maize or taro plants, which could reduce plant-to-plant dissemination of the disease through rain splash or drops of water carried by wind (Fanou et al., 2018).

Concerning the impact of the intercropping systems on cassava yield, generally, no significant yield loss occurred in intercropped cassava compared to monocropping in the present study, except in the forest-savanna transition zone where a significant yield reduction in intercropping cassava with maize was observed. Intercropping cassava with maize in the forest-savanna transition zone of Nigeria did not cause a reduction in yield (Fanou et al., 2018). However, Zuofa et al. (1992) reported cassava yield reduction in intercropping cassava with maize in the rainforest zone of Nigeria.

Since intercropping cassava-maize and cassava-taro reduced disease severity slightly, but in most cases significantly in the forest highland, the forest lowland and the wet savanna zone, with no significant negative yield effect due to the cropping system, these cropping systems can be recommended in these ecozones as part of an integrated control strategy for cassava bacterial blight. A suppressive effect of intercropping might be more obvious when a medium-resistant variety is used.

Since no resistant varieties were identified among local and local improved varieties across ecozones in Togo (Wydra et al., 2007; Banito et al., 2008), the combination of medium resistant varieties and an intercropping system, both adapted to the respective ecozone, could be recommended to farmers as part of a cassava bacterial blight control strategy.

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