Journal of Agriculture and Environmental Sciences
December 2017, Vol. 6, No. 2, pp. 121-127
ISSN: 2334-2404 (Print), 2334-2412 (Online)

Published by American Research Institute for Policy Development
DOI: 10.15640/jaes.v6n2a14
URL: https://doi.org/10.15640/jaes.v6n2a14

Multiyear (2010 – 2014) Assessment of Bollgard II® Efficacy in Commercial Small Scale Farmer Fields in Burkina Faso

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Abstract

Cotton is the main cash crop in Burkina Faso and more than three millions people are living on this crop. Unfortunately, many insect pests' damages cause low yield and incomes for farmers. Because of chemical overuse, pests have developed resistance to pyrethroids. In the aim to find a solution to insect pests' damages, Bollgard II® was tested from 2003-2008 and commercially released in 2009. Bollgard II® efficacy on leaf-worms and bollworms was monitored in farmers' fields at 8 locations from 2010 to 2014 in the three major cotton production zones of Burkina Faso. The results showed that total number of worms, leaf-worms and bollworms were respectively 4.5, 3.0, and 6.7 times higher in conventional cotton sprayed six times compared to twice sprayed Bollgard II®. The results showed also 23.9% (272.2 kg ha^{-1}) yield increase with Bollgard II®. No indications on Bollgard II® efficacy reduction during the commercial period of 2010 to 2014 were found. Ensuring the implementation of a structured refuge will best maintain Bollgard II® long-term economic value to cotton growers in Burkina Faso.

Keywords: cotton, Bollgard II®, insect pests, refuge.

1. Introduction

Cotton is the main cash crops for Burkina Faso (Traoré *et al.*, 2008). In fact, cotton generates over 60% of Burkina Faso's export earnings (ICAC, 2006) and serves as a vital catalyst to the country development sector with more than three million people earning all or part of their income from the cotton (Vognan *et al.*, 2008).The cotton crop is subject to many pest attacks, the main being lepidopteran larvae. Since the 1960's, different types of insecticides have been used to control these cotton pests in Burkina Faso. During the 1990s, a number of pests (mainly the bollworm *Helicoverpa armigera*) developed resistance to pyrethroids insecticides, the main chemical family used to control caterpillars in cotton fields in West Africa (Héma *et al.*, 2009a, Ochou *et al.*, 1998). The organ chloride endosulfan was introduced to counter pyrethroids resistance but was not able to reduce the level of this resistance in *H. armigera*. To minimize the economic losses due to *H. armigera*, the farmers increased the number of sprays resulting in environmental and health hazards. The cotton sector in Burkina Faso was at the brink of the bankruptcy (poor productivity, credits not reimbursed back by farmers). Investigations from scientists have led to the testing of a genetically modified cotton (Bollgard II® cotton) containing *Bt* proteins Cry1Ac and Cry2Ab2, both of which control selected cotton caterpillar pests (Traoré *et al.*, 2008). This Bollgard II® cotton is a second generation *Bt* product and provides effective control of *H. armigera* in several geographic regions (James, 2012).

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From 2003 to 2008, confined field trials were carried out on INERA research stations to show the efficacy of Bollgard II® cotton in controlling the main lepidopteran pests (Héma *et al.*,2009b). The results were convincing and the Bollgard II® cotton (*Bt* cotton) was commercially released in farmers' fields since 2009. After the commercial release of Bollgard II® cotton, a yearly contract was established between Monsanto and INERA to continue monitoring the technology efficacy in farmers' fields. The current paper summarizes the results obtained during the five first years (2010-2014) of commercial Bollgard II® cotton cultivation in farmers' fields in Burkina Faso.

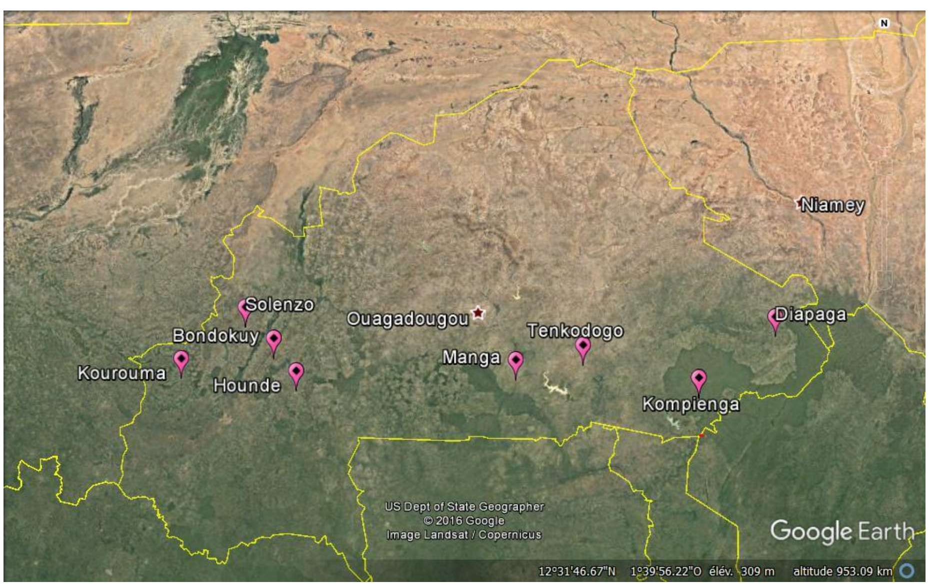
2. Materials and methods

2.1. Trials locations

The monitoring trials were carried out at 8 different locations in farmers' fields in the three cotton companies' area of Burkina Faso (SOFITEX, FASO COTON and SOCOMA). The climate in the trials zone is the Sudanian (south and north Sudanian) with rainfall between 700-1200 mm. The 8 locations were selected in order to cover the diversity of climate and soil conditions encountered in the cotton production zones of Burkina Faso. Soils and climate conditions are more favorable for SOFITEX and SOCOMA zones compared to FASO COTON zone with drier and poor soil. Sowing period was extended from earlier June to mid-July depending to rain precocity. The number of trials per site varied depending on the cropping year. Table 1 and map 1 summarize the trials locations, the number of trials per site during these 5 cropping seasons.

Table 1. Efficacy Trials Locations Characterization (2010-2014)

Cotton company	Climatic zone	Rainfall (mm)	Cropping period	Location	Numbers of trials per location per year				
					2010	2011	2012	2013	2014
SOFITEX	South Sudanian	900-1200	May-October	Houndé	10	10	10	10	10
				Kourouma	10	10	10	08	08
				Bondokuy	-	-	-	08	08
				Solenzo	-	10	10	-	-
FASO COTON	North Sudanian	700-900	June-September	Manga	-	10	-	08	08
				Tenkodogo	10	10	10	08	08
SOCOMA	South Sudanian	900-1100	May-October	Diapaga	10	10	10	08	08
				Kompienga	-	-	10	10	10



Map 1: Trials location map

2.2. Experimental design

The trials were carried out on a non-replicated design composed of 2 plots of 0.5 ha each in farmers' field. In the 2 plots, conventional and Bollgard II® cotton were compared under the same farming conditions.

The planting density was the density adopted by farmers in the area and this varied from 30 to 40 cm within rows and 60 to 80 cm between rows. Seeds were pretreated with imidacloprid 35% + thiram 30% WS before sowing. Two cotton plants are kept by hole. For all the plots and all locations, the following operations were done:

- Organic manure application at 2 t/ha as recommended by the extension.
- Pre-emergence herbicide applied after sowing to avoid any interference between cotton plants and weeds.
- Chemical fertilizer: 100 kg of NPKSB and 50 kg of KCl applied 15 days after emergence and 50 kg urea at 40-45 days after emergence.
- Hand weeding: 2 -3 depending on the weed pressure

2.3. Insecticides spray

The insecticides were applied by the same operator on all plots with the Very Low Volume (VLV) sprayer (10 l/ha) or Low Volume (LV) (60 - 120 l/ha). The insecticides application starts at 30 days after emergence and repeated every 14 days for the conventional plots and at 86 and 100 days after emergence for Bollgard II cotton. Table 2 summarizes insecticides application scheme:

Table 2. Insecticides Sprays Scheme in Efficacy Trials

	T1= 30 DAE	T2=44 DAE	T3=58 DAE	T4=72 DAE	T5= 86 DAE	T6=100 DAE
Transgenic	NONE	NONE	NONE	NONE	IU	IU
Conventional	profenofos orindoxacarb	profenofos orindoxacarb	IU	IU	IU	IU

T1… T6 = Spray Number; DAE = Days After Emergence
Profenofos: dose 1 l/ha; indoxacarb 150 SC: dose 170 ml/ha;
IU = Insecticide Used in the cotton company's area (pyrethroid + organophosphate or pyrethroid + neonicotinoid).

2.4. Field data collection

All observations were carried out on 30 plants taken by groups of 5 consecutive plants on a diagonal (Ochou *et al.*, 1998) once every week from 29 days after emergence to harvest: The following observations were done on the two plots:

- Counting the number of the following bollworms: *Helicoverpa armigera*, *Earias sp.*, *Diparopsis watersi*;
- Herbivore caterpillars (foliar-feeding):
  - *Haritalodes derogata*: counting the number of infested plants with alive insects;
  - *Anomis flava* and *Spodoptera littoralis*: counting the number of individuals;
- Health analysis of the mature bolls on 5 lines of 20 m per plot (counting the number of healthy and damaged bolls);
- Harvest: during the vegetative phase (approximately 100 days after emergence), 3 squares of 100 m² each (10 m x 10 m) were placed in each plot for yields estimation.

2.5. Statistics

All statistical analyses were performed in R! Software (Core, 2015). Restricted-maximum likelihood ANOVA (lme4 package) was used to determine the impact of Bollgard II® cotton on the following variables: cotton yields, number of healthy bolls, cumulative infestation for all lepidopteran pests of cotton, cumulative infestation of the bollworm complex and cumulative infestation of foliar-feeding lepidopterans. For all analyses, cotton variety (Bollgard II® vs. conventional cotton) was considered as a fixed effect; year, location nested within a year, "location (year)", and grower field nested with year-location combinations, "residual", were random effects in the model. Denominator degrees-of-freedom (df) for F-tests were estimated via Satterthwaite's approximation. To enhance normality of the data and reduce heteroscedasticity, the following data transformations were used: square-root transformation for yield and all measures of larval infestation and $\log_{10}$-transformation for the assessment of healthy bolls. All summary statistics presented in Table 3 were calculated on non-transformed data.

3. Results and discussion

3.1. Efficacy of Bollgard II®cotton in farmers' field in Burkina Faso

The results showed significantly lower lepidopteran pests infestation in the Bollgard II® cotton fields compared to the conventional ones (Table 3). Approximately 4.5, 3.0, and 6.7 times more total numbers of larvae, foliar-feeding larvae and bollworm, respectively, were found in the conventional cotton treatment sprayed 6 times when compared to the Bollgard II® cotton sprayed 2 times (Table 3).

Most of the variability in larval counts was site dependent within years. Annual variability in cotton-treatment effects were minimal (Table 4). The results showed 71.2% and 58.8% of Bollgard II® cotton fields with no bollworms and no foliar-feeding worms, respectively, against 11.3% and 19.8% for conventional cotton fields. These results are comparable to those reported by Héma *et al.*, (2009b) in confined field trials in Burkina Faso with the Bollgard II® cotton, Armstrong *et al.* (2011) and Greene (2015) in USA and Wu *et al.* (Wu *et al.*, 2005; Wu *et al.*, 2008; Wu and Guo, 2004) in China. For Udikeri *et al.* (2011) Hallad *et al.* (2014), Onkaramurthy *et al.* (Onkaramurthy *et al.*, 2015; Onkaramurthy *et al.*, 2016), and which concluded that the efficacy of the second generation of the *Bt* cotton (Bollgard II®) on lepidopteran larvae was due to the combined efficacy of the two proteins Cry1Ac and Cry2Ab2 (Downes *et al.*, 2010; Greenplate *et al.*, 2003, Knight *et al.*, 2016). Our results showed that the Bollgard II® cotton is efficient in controlling the main cotton leaves worms (*Haritalodes derogata*, *Spodoptera littoralis* and *Anomis flava*) in Burkina Faso. The Spodoptera gender often reported to be unsusceptible to *Bt* toxins seemed to be well-controlled in our monitoring trials. The second generation of *Bt* cotton (Bollgard II®) containing Cry1Ac and Cry2Ab2 has solved Cry1Ac inefficacy (Bollgard I®) on Spodoptera species often reported (Greenplate *et al.*, 2000; Onkaramurthy *et al.*, 2015). Indeed, larvae fed with the two generations *Bt* cotton tissues got longer life cycle conducting to high mortality (Arshad and Suhail, 2011, Kumar and Grewal, 2015).

Table3. REML-ANOVA forMeasures ofCotton Performance. Means, Standard Errors, And 95% Confidence Intervals for Non-Transformed Data Are Presented For Both Cotton Treatments.

Variable	Numerator df	Denominator df	F	Mean±SE (95% Confidence Interval)	
				Bollgard II	Conventional
Yield[1]	1	330.3	74.5****	1456.2±31.4 (1394.6,1517.8)	1184.0±29.7 (1125.9,1242.1)
Healthy Bolls[2]	1	330.1	59.8****	26.83.6±0.88 (25.11,28.56)	21.63.0±0.73 (20.21,23.05)
Total Insect Infestation[3]	1	332.1	489.6****	0.73±0.12 (0.50,0.96)	3.25±0.33 (2.60,3.90)
Bollworm Infestation[3]	1	332.1	914.8****	0.29±0.06 (0.17,0.41)	1.93±0.19 (1.56,2.29)
Foliar Larval Infestation[3]	1	332.2	138.4****	0.44±0.07 (0.31,0.57)	1.32±0.16 (1.00,1.64)

**** - $P < 0.0001$;[1] - kg ha^{-1}; [2] - bolls m^{-1}; [3] – number of larvae per 30 plants

3.2. Seed Cotton Yield and Healthy Bolls in *Bt*and Conventional Fields.

The fact the lepidopteran larvae were controlled by Cry toxins in Bollgard II® cotton contributed to an increase in cotton yields and better quality bolls. In fact, the results from the Bollgard II® fields showed significantly greater cotton yields compared to the conventional cotton fields sprayed 6 times (Table 3). On average, the Bollgard II® field provided 272.2 kg ha^{-1} (23.9%) more yield than conventional cotton one. The yearly variability of the cotton yields were negligible (Table 4). Most of the variation was due to differences among locations and fields within a location. From the 177 fields, the Bollgard II® fields generated in 87.0% of the side by side comparisons an increased cotton yield compared to the conventional field (Fig. 1). The yield advantage observed for Bollgard II® fields were partially due to higher numbers of healthy bolls (24% more) when compared to the conventional field (Table 3). Similar results were reported in confined field trials carried out in Burkina Faso (Héma *et al.*, 2009b). The effectiveness of these two *Bt* genes to control the bollworms (main cotton pest in cotton in Burkina Faso), is the basis of the yield increase and the higher number of healthy bolls observed in Bollgard II® cotton fields. Hallad *et al.* (2014) in India have reported 39.8% of seed cotton yield increase for the Bollgard II® cotton compared to conventional variety sprayed six times (4 time against bollworms and 2 times against sucking pest). In the same country, Udikeri *et al.* (2011) and Onkaramurthy *et al.* (2016) reported bolls damage reduction and yield increase with 2nd generation *Bt* cotton.

On average in China, the yield increase due to *Bt*toxins effectiveness ranged from 31 to 63% (Witjaksono *et al.*, 2014). In the US the yield increase over the past ten years was approximately 33% (Witjaksono *et al.*, 2014). In terms of income, Sankula *et al.* (2005) reported the net benefit of US$74.29/ha for conventional cotton and US$128.85/ha for *Bt*one. In China, the average profit per ha ranged from $76 to $250 for *Bt* cotton. The *Bt* cotton cultivation led to the reduction of insecticides use and in Burkina Faso, the technology allowed 4 spayssaving.

These results were also reported for China (with 67% reduction) for Australia (with 64% reduction) (Witjaksono *et al.*, 2014). Because the *Bt* genes don't control the sucking pests (aphids, white flies, jassides, and bugs), it was recommended in Burkina Faso to spray two times at 86 and 100 days after cotton emergence. These two insecticides sprays should be done after threshold as recommended by many authors (Freeman and Smith, 1997; Wu *et al.*, 2002; Fitt, 2003; Greene, 2015); but since many factors are involved, thresholds should be flexible (Freeman *et al.*, 1997) in order to enable an increase in the population of useful insects, optimize the natural control of certain sucking pests by beneficial arthropods (Yang *et al.*, 2005), and consider refuge strategy and other tools for technology sustainability (Gould, 2013; Huang *et al.*, 2012, Sankula *et al.*, 2005; Tabashnik *et al.*, 2013; Tabashnik, 1994).

Table 4.Estimates of Variance Due to Random Effects (Percentage of Total, %) for Each Measured Variable In REML ANOVA.

Variable	Year	Location (Year)	Residual
Yield	0.0 (0%)	0.1394 (44.5%)	0.1741 (55.5%)
Healthy Bolls	0.0 (0%)	0.0264 (65.2%)	0.0141 (34.8%)
Total Insect Infestation	0.0 (0%)	0.4903 (72.4%)	0.1872 (27.6%)
Bollworm Infestation	0.0 (0%)	0.2823 (76.9%)	0.0846 (23.1%)
Foliar Larval Infestation	0.0 (0%)	0.2653 (62.9%)	0.1566 (37.1%)

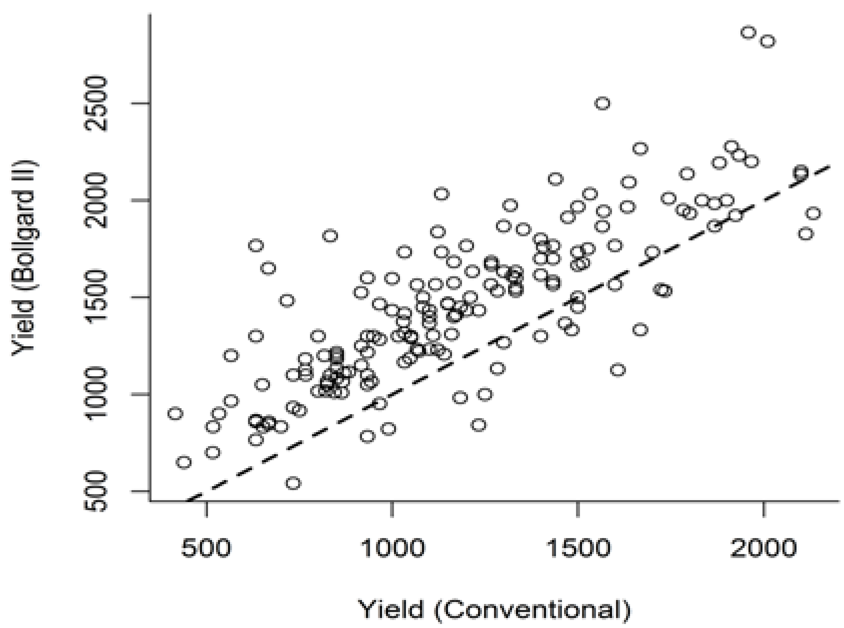


Figure 1. Yields (kg ha^{-1}) in Bollgard II® vs. conventional cotton paired by grower's fields. Dotted line represents expectation if Bollgard II and conventional cotton had a one-one relationship. Data points above the dotted line represent Bollgard II yields that were greater than yields in conventional cotton.

4. Conclusions

The efficacy of Bollgard II® cotton on the main cotton worms (bolls worms and leaves worms) was demonstrated in farmers' fields in Burkina Faso across this study covering from 2010 till 2014. The Cry genes were able to efficiently control these two types of lepidopteran pests leading to healthier bolls and higher cotton yields in Burkina Faso.

No decrease in the efficacy was observed during these five years of monitoring but there is a need to adequately implement the structured refuge to avoid the occurrence of resistance to Bollgard II® cotton. For these reasons, resistance management tools must be deployed to assure sustainability of this technology.

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