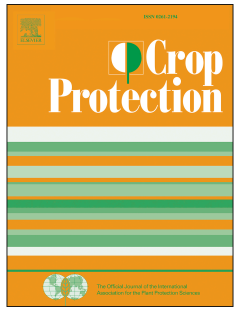

Integrated pest management for yard-long bean (*Vigna unguiculata* subsp. *Sesquipedalis*) in Cambodia

Antonino Malacrinò, Kim Hian Seng, Chanratha An, Socheath Ong, Megan E. O'Rourke

PII: S0261-2194(19)30145-0

DOI: https://doi.org/10.1016/j.cropro.2019.05.005

Reference: JCRP 4811

To appear in: *Crop Protection*

Received Date: 16 August 2018

Revised Date: 2 May 2019

Accepted Date: 4 May 2019

Integrated pest management for yard-long bean (*Vigna unguiculata* subsp. *sesquipedalis*) in Cambodia

Antonino Malacrinò[1], Kim Hian Seng[2], Chanratha An[2], Socheath Ong[3], Megan E. O'Rourke[4]

[1] Department of Physics, Chemistry and Biology (IFM), Linköping University, Linköping (Sweden)
[2] iDE Cambodia, Phnom Penh (Cambodia)
[3] Royal University of Agriculture, Phnom Penh (Cambodia)
[4] School of Plant and Environmental Sciences, Virginia Tech, Blacksburg, VA (USA)

Corresponding author: Megan E. O'Rourke, megorust@vt.edu

Abstract

Pesticides are widely used to protect crops against insect pests and diseases. However, current conventional pest management strategies can severely impact environmental and human health. Therefore, it is timely to find an alternative to conventional chemical control in order to counteract the negative effects of pesticides. Integrated Pest Management (IPM) represents a promising alternative in vegetable production to reduce pesticide use while also maintaining acceptable yields. In this study, we compared an IPM strategy with conventional management and a no-spray control for managing arthropod pests and pathogens in yard-long bean (*Vigna unguiculata* subsp. *sesquipedalis*), an important crop in Southeast Asia. We conducted the study during two crop cycles (rainy and dry season) on 24 farms (12 per season) spread across the Soutr Nikum district of Siem Reap, Cambodia. In both seasons, our IPM strategy controlled overall pest levels better than conventional pest management, which exhibited better pest control than the no-spray control. This pattern was not reflected by yields, however, since dry season yields were similar across all three treatments (conventional = 1.68, IPM = 1.73, control = 1.52 kg m^{-2}), and rainy season yields were similar between conventional and IPM treatments, which were higher than yields in the no-spray control treatment (conventional = 1.74, IPM = 1.71, no-spray control = 1.33 kg m^{-2}). The costs of IPM were higher than conventional management, which contributed to lower profits in the IPM treatments in both seasons despite yields being as high as in conventional production. Therefore, future efforts should focus on increasing the profitability of IPM production.

Keywords: IPM; on farm trial; *Bacillus subtilis*; *Bacillus thuringiensis*; *Beauveria bassiana*; *Trichoderma*

Highlights

- IPM outperformed conventional pest management on yard-long bean in Cambodia
- Pest management strategy did not influence yard-long bean yields
- Costs for IPM were higher than for conventional management

1. Introduction

Agricultural pests, including arthropods, diseases, and weeds can lead to complete crop failures (Matson et al., 1997). Over the past century, farmers have increasingly turned to chemical pesticides to help control pest problems. In fact, the use of chemical pesticides has quickly become widespread worldwide and it is estimated that 35 billion kg of pesticides are applied annually (FAOSTAT, 2014). However, it is also estimated that pests still cause the loss of 37% of potential crop yield (Pimentel, 2005). Pesticides can also have many negative side effects including: human health impacts, water contamination, killing of non-target species, residues in food, and the onset of pesticide resistance (Carvalho, 2006; Chagnon et al., 2015).

Integrated Pest Management (IPM) provides a potential alternative to conventional pest management. It was originally introduced as "applied pest control which combines and integrates biological and chemical control" (Stern et al., 1959). During the last 60 years, IPM has garnered a great deal of research attention, and we now have a strong foundational knowledge of how to implement it for most major crops (Way and van Emden, 2000). We still lack sufficient knowledge, however, of IPM in minor crops. Furthermore, IPM needs to be continuously studied and tailored to specific situations, which can be a major obstacle to implementing IPM in developing countries (Parsa et al., 2014). Another barrier to the spread of IPM in developing countries is that conventional pesticides are perceived as a solid investment to protect crops and guarantee production (Timprasert et al., 2014).

In Cambodia, agricultural production plays a pivotal role in the economy and in ensuring food security. Indeed, 80% of the population lives in rural areas, and the land dedicated to agriculture is estimated to be 5.5 million hectares (FAOSTAT, 2014), or about one third of the country. Although policy makers are pushing towards the implementation of IPM in the field, conventional pest management is still widespread along with the associated negative impacts on environmental and human health (Jensen et al., 2011). Examples of IPM strategies implemented in Cambodia are also nearly absent from the scientific literature. For example, a former meta-analysis by Pretty and Bharucha (2015), spanning 85 IPM projects over 24 developing countries, reports just one example for Cambodia (Van den Berg and Jiggins, 2007).

The current challenge is to develop successful IPM strategies tailored to Cambodia's diet and growing conditions. *V. unguiculata* subsp. *sesquipedalis* Verdc. (yard-long bean) is one of the most important legumes for human food consumption cultivated in Cambodia (Tantasawat et al., 2010). This species can be grown year-round, and the long pods can be collected many times per crop cycle (Kongjaimun et al., 2012). Insects and diseases, however, pose a major challenge to successful yard-long bean cultivation. In a survey or farmers in Thailand and Vietnam, leaf rust (*Uromyces vignae*) and bean pod borer (*Maruca vitrata*) were consistently ranked as the top disease and insect pest, respectively (Schreinemachers et al., 2014). Other insect pests were more variable between Thailand and Vietnam with armyworm (*Spodoptera litura*), bean butterfly (*Lampides boeticus*) and the American bollworm (*Helicoverpa armigera*) being major insect pests in Vietnam while aphids (*Aphis craccivora* and *Aphis glycines*) were more important in Thailand. For diseases, leaf spot, root rot, and yellow mosaic virus were major challenges in Vietnam, while damping off was more important in Thailand (Schreinemachers et al., 2014).

Due to the importance of yard-long bean as a staple food in Southeast Asia and Cambodia, and with the aim to favor the implementation of IPM to counteract the negative effects of pesticide overuse, we evaluated the use of IPM strategies to protect yard-long bean against both its major arthropod pests and pathogens. In this research, we took an "IPM package" approach whereby we tested a suite of management strategies simultaneously (Dinakaran et al., 2013). Specifically, we focused on the substitution of conventional pesticides with biocontrol agents targeting insects (*Beauveria bassiana*, *Bacillus thuringiensis*) and diseases (*Bacillus subtilis*, *Trichoderma viride* and *Trichoderma harzianum*), and products of natural origin (orange essential oil). These products have been shown to be effective in controlling agricultural pests while being safe for mammals, having low toxicity for natural enemies or non-target species, and having an overall low environmental impact (Amin et al., 2010; de Boer Sietske and Diderichsen, 1991; Koul et al., 2008;Siegel, 2001; Srinivasan et al., 2019; Zimmermann, 2007). We specifically did not use neem in our IPM package because its use as a pesticide would compete with human consumption in Cambodia (Seng, pers. comm.). We hypothesized that our IPM strategy could control arthropod pests and pathogens in yard-long bean at least as well as conventional management.

2. Materials and methods

2.1 Experimental design

The study was carried out over a rainy and dry season in the Soutr Nikum district of Siem Reap, Cambodia. This area is flat with a sandy loam soil (pH 4.2–5.9). During the rainy season (July 2017 – November 2017), the total rainfall was 1,200 mm, with the temperatures ranging from 27 to 33°C with 45-76% relative humidity. During the dry season (November 2017 – March 2018), the total rainfall was 230 mm, with the temperatures ranging from 24 to 33°C with 35-62% relative humidity.

The research was designed with farmers using a farming systems research approach that emphasized participatory, on-farm research (Darnhofer et al., 2012; Norton et al., 1999). During each season, we established on-farm trials at 12 unique sites (24 farms total). Each farm was considered one experimental replicate and contained two treatments for pest control of yard-long bean: IPM, and conventional farmer practices. While the IPM treatment protocol was standardized across farms, the conventional treatment included a mix of farm practices; each farmer managed their conventional plot according to their usual practices (Dinakaran et al., 2013). There was also a small, no-spray control area on each farm that was not treated with any pesticides and that served as a check for pest pressures. Each farm devoted around 300m^2 to the experiment, with treatments encompassing entire rows of production and treatments separated only by the regular inter-row space with an approximate ratio of the treatment areas of 3:3:1, IPM to conventional to no-spray control, respectively. The locations of the IPM and conventional management treatments were randomized within the experimental area and the no-spray control was always an outside row. While the relatively small size or placement of the no-spray control could bias results in that treatment, its placement was decided in conjunction with farmers in order to minimize spray drift and farmers accidentally spraying those plots (Bingen and Gibbon 2012).

2.2 Agronomy

Field plots were prepared about two weeks before transplanting by roughly ploughing the soil before the application of compost/manure (5 t ha^{-1}) and lime (320 kg ha^{-1}). Soil was then ploughed a second time, and hilled (80 cm wide × 30 cm high) with a 70 cm walkway between rows. Before transplanting seedlings, the drip irrigation system was placed lengthwise in the middle of each bed and covered with silver plastic mulch. Yard-long bean seedlings were transplanted one week after seeding and were planted at 10 cm of depth with 30 cm between plants within the same row (4.76 plants/m^2). Plants were grown vertically on a two meter high trellising system of nets supported by small bamboo or wooden poles, and irrigated daily according to crop needs. In the rainy season, fields were fertilized with 50 kg ha^{-1} N, 40 kg ha^{-1} P, and 80 kg ha^{-1} K, and during the dry season with 45 kg ha^{-1} N, 45 kg ha^{-1} P, and 220 kg ha^{-1} K. Fertilization was divided between a basal fertilizer application during the first plow, which included 40% of total N and P and 19% of total K during the rainy season, and 44% of total N and P and 7% of total K during the dry season, with the remaining fertilizer applied as weekly fertigation applications.

2.3 Pest management

The IPM treatment included a focus on growing healthy seedlings followed by weekly spray treatments with biological control agents as outlined in Table S1. Seedlings were planted in seed trays in coco peat derived from coconut husks (Jiffy DecoGro, Jiffy International AS, Norway) and were inoculated with *Trichoderma viride* (Royal University of Agriculture, Cambodia) at 0.5 g L^{-1} of coco peat and protected with insect exclusion nets (32 mesh $inch^{-2}$). After one week, seedlings were transplanted into the field and watered in with a *T. viride* solution. Thereafter, plants were treated weekly with three biocontrol agents: *Bacillus thuringiensis* subsp. *aizawa* (Ladda Co., Ltd., Thailand) at 0.4 kg ha^{-1}, *Bacillus subtilis* (Tab Innovation Co., Ltd., Thailand) at 0.25 kg ha^{-1}, and *Beauveria bassiana* (Tab Innovation Co., Ltd., Thailand) at 0.4 kg ha^{-1}, which were mixed together and sprayed onto foliage weekly. During only the rainy season, farmers sprayed *Trichoderma harzianum* (Royal University of Agriculture, Cambodia) at 0.6 kg ha^{-1} two times onto foliage in weeks four and five after transplanting seedlings. Orange essential oil (Prev-Am, ORO Agri Inc, USA), with the active ingredient sodium tetraborohydrate decahydrate, was also used periodically as an insecticide when needed. Conventional pesticides were used sparingly in the IPM treatment as follows: metalaxyl (Kalaxy 35WP, Papaya Trade Co., Cambodia) was sprayed one or two times as needed at a rate of 590 g ha^{-1}for disease control during the rainy season, and imidacloprid (Confidor 200SL, Bayer, Germany) was sprayed once by each farmer at a rate of 667 ml ha^{-1} during the dry season targeting thrips. Yellow sticky traps (35 x 40 cm) and two pheromone lures were also placed throughout each experimental field for monitoring.

Applying a farming systems research approach, plants cultivated under conventional management were grown according to the discretion of each individual farmer using pest management practices summarized in table S2 (Darnhofer et al., 2012; Dinakaran et al., 2013). Seedlings were typically grown in seed trays using field soil and without any netting protection. After transplanting, farmers used a wide range of different chemicals to control pests, which included eleven different insecticides and four different fungicides over the two seasons. Seedlings for no-spray control plots were grown with the same procedure used for IPM, but without using *Trichoderma viride*, and after transplanting they did not receive any pest management treatment.

2.4 Data collection and analysis

After transplanting seedlings into the field, we scouted all experimental fields each week and selected five random plants per treatment on which we evaluated the damage by arthropod pests and pathogens (Table 1). The pest and disease levels on each plant were scored on a simple scale from 0 to 3, which was granularly calibrated so that it was meaningful and replicable for farmers to differentiate: 0 nothing, 1 little damage/infestation, 2 moderate damage/infestation, and 3 severe damage/infestation (Table S3). We also recorded yield, input costs (not including labor), income from sales, and profit. Input costs included all the expenses for fertilizers, conventional or IPM inputs, and gasoline for pumping water; input costs also included seed trays, insect exclusion net, plastic mulch, drip irrigation system, and trellis system, adjusted for depreciation. Yield related parameters, including marketable pod weight (kg) and price of marketable pods per kg (USD), were collected by farmers and reported to researchers for each treatment. Costs and benefits were calculated for each treatment by subtracting all the expenses from all the income from selling marketable pods.

Table 1. Pests and diseases scored during surveys

Pest Category	**Common Name**	**Scientific Name**
Insect	Armyworm	*Spodoptera exigua* (Hübner) and *Spodoptera litura* (F.)
	Bean fly	*Ophiomyia phaseoli* (Tryon)
	Bean pod borer	*Maruca vitrata* (F.)
	Cowpea aphid	*Aphis craccivora* Koch
	Green stink bug	*Acrosternum hilare* Say
	Leafhopper	*Empoasca kerri* Singh-Pruthi

	Spider mite	*Tetranychus* spp.
	Sweet potato whitefly	*Bemisia tabaci* (G.)
	Thrips	*Megalurothrips usitatus* (Bagnall)
	Vegetable leaf miner	*Liriomyza sativae* Blanchard
Disease	Downy mildew	*Peronospora parasitica*
	Fusarium	*Fusarium oxysporum*
	Powdery mildew	*Sphaerotheca fuliginea*
	Pythium	*Pythium* spp.
	Rhizopus	*Rhizopus* spp. including *Rhizopus stolonifer* and *Rhizopus solani*
	Rust	*Uromyces vignae*
	Viruses	especially *Bean common mosaic virus*

Due to very different growing conditions between the wet and dry seasons, all analyses were carried out separately for each season using R 3.5 (R Core Team, 2013). The global analyses of overall pest scores were performed by fitting a Generalized Linear Mixed-Effects Models (GLMM) using the treatment (IPM, conventional, and no-spray control) as a fixed effect, and pest, farm, and sampling week as random variables. Analyses of each pest's infestation scores were performed similarly but with pest removed as a random variable. Models were fitted using the *glmer* function under the *lme4* package (Bates et al., 2015) specifying a poisson error distribution and log link function. The effect of treatment was assessed using Type II Wald χ^2 from the package *car* and function *Anova*, and the package *emmeans* was used to infer pairwise contrasts using Tukey's multiple comparison procedure. Pests for which at least one treatment did not have an average infestation score of at least 0.02 (i.e. at least one of 50 plants was scored as 1 or higher per week) were not statistically analyzed due to lack of data and model convergence issues. Data about yields, costs, incomes and profits were analyzed through a Linear Mixed-Effects model (LME) using the *lmer* function and specifying treatment as a fixed effect and farm as a random variable, and pairwise contrasts were inferred using Tukey's multiple comparison procedure, as described above.

3. Results

3.1 Infestation

A general view over the infestation scores of the entire dataset suggests differences between all the experimental treatments (χ^2 = 61.8; P <0.001 for rainy season, and χ^2 = 45.1; P <0.001 for dry season). Overall pest and disease infestation scores were higher in the conventional than the IPM management plots (P <0.05), while both strategies had lower overall infestation scores than the no-spray control plots (P < 0.05) (Fig. 1).

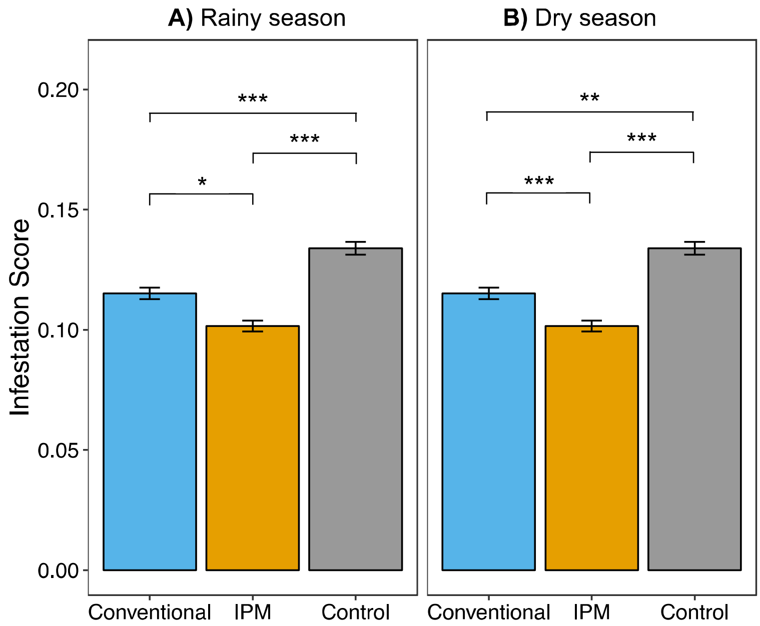

Fig 1. Global infestation scores and standard errors for yard-long bean under conventional management, IPM and no-spray control during both rainy (A) and dry (B) seasons. Significant pairwise differences are denoted by asterisks (*** = $P < 0.001$, ** = $P < 0.01$, * = $P < 0.05$).

A closer analysis of individual pathogens of yard-long bean indicated variability in the severity of different diseases across the two seasons of study. In the rainy season, powdery mildew and *Rhizopus* were the main pathogens, while rust was the main pathogen in the dry season. *Fusarium* was not detected at all during the rainy season, while powdery mildew and *Pythium* were detected at much lower levels in the dry season than in the rainy season. The efficacy of management strategies also differed across the two seasons. Specifically, during the rainy season, rust symptoms were reduced in IPM compared to conventional plots while powdery mildew was lower in IPM compared to no-spray control plots but not compared to conventional management. During the dry season, *Rhizopus* scores were lower under IPM than conventional management, while symptoms of viruses were less in IPM compared to the no-spray control treatment but not compared to conventional management. Downy mildew was not affected by management and was detected at similarly low levels across both seasons of study. Pathogen scores for each season were never higher, on average, in the IPM treatment than the conventional treatment (Tab. 2).

The arthropod analysis revealed that aphids, armyworms, bean flies, and thrips were the most common pests with armyworms having higher scores in the dry season than the rainy season, while bean flies and thrips had higher scores in the rainy season than the dry season, and aphids reached similarly high levels in both seasons. The effect of pest management was consistent across both seasons against armyworms, with lower values under IPM than conventional management. It was also consistent across both seasons that aphids were controlled equally well by IPM and conventional management compared with the no-spray control. Damage by bean fly and leafhopper was controlled equally well by both IPM and conventional methods compared to no-spray control during the rainy season but these pests were detected much less in the dry season with no apparent management effects. Thrips were also equally controlled by both IPM and conventional methods during the dry season, but no effect of treatment was observed during the rainy season. There were no management effects on bean pod borer in either season and the pheromone lures did not appear attractive as no adult *M. vitrata* were captured. Spider mites were not observed in the wet season and stink bugs were not observed in the dry season. Leaf miner, and whitefly pest levels were very low in both seasons. Arthropod pest scores for each season were never higher, on average, in the IPM treatment than the conventional treatment (Tab. 3).

Table 2. Infestation scores for each pathogen recorded during both seasons. Different letters indicate significant pairwise differences between values in the same row within the same season at the $P < 0.05$ level.

	Rainy season					Dry season				
Pest	χ^2	**P**	**Conventional**	**IPM**	**No-spray control**	χ^2	**P**	**Conventional**	**IPM**	**No-spray control**
Downy mildew	4.5	0.1	0.02±0	0.02±0	0.01±0	1.7	0.4	0.02±0	0.01±0	0.02±0.01
Fusarium	N/A[1]	N/A	0	0	0	15.4	**<0.001**	0.038±0.013 (b)	0.003±0.002 (a)	0.013±0.007 (ab)
Powdery mildew	8.86	**0.01**	0.43±0.03 (ab)	0.41±0.03 (a)	0.51±0.03 (b)	N/A	N/A	0.008±0.004	0.003±0.002	0.003±0.002
Pythium	0.7	0.6	0.04±0.01	0.04±0.01	0.03±0.01	N/A	N/A	0.002±0.002	0.002±0.002	0.003±0.002
Rhizopus	4.2	0.1	0.34±0.02	0.28±0.02	0.31±0.02	13.1	**0.001**	0.04±0.01 (b)	0.01±0.007 (a)	0.01±0.01 (ab)
Rust	7.57	**0.02**	0.09±0.01 (b)	0.06±0.01 (a)	0.09±0.01 (b)	0.4	0.8	0.56±0.03	0.53±0.03	0.56±0.03
Virus	ND[2]					10.1	**0.006**	0.07±0.01 (ab)	0.05±0.01 (a)	0.09±0.02 (b)

[1] Statistical analyses were not applicable due to low infestation scores.

[2] Virus levels were not determined during the dry rainy season.

Table 3. Infestation scores for each pest recorded during both seasons. Different letters indicate significant pairwise differences between values in the same row within the same season at the $P <$ 0.05 level.

	Rainy season					Dry season				
Pest	χ^2	**P**	**Conventional**	**IPM**	**No-spray control**	χ^2	**P**	**Conventional**	**IPM**	**No-spray control**
Aphids	42.6	**<0.001**	0.3±0.02 (a)	0.32±0.02 (a)	0.48±0.02 (b)	26.02	**<0.001**	0.32±0.02 (a)	0.27±0.02 (a)	0.43±0.02 (b)
Armyworms	9.06	**0.01**	0.1±0.01 (ab)	0.08±0.01 (a)	0.13±0.01 (b)	20.2	**<0.001**	0.22±0.02 (b)	0.16±0.01 (a)	0.28±0.02 (c)
Bean flies	11.8	**0.002**	0.31±0.02 (a)	0.28±0.02 (a)	0.38±0.02 (b)	N/A[1]	N/A	0.017±0.005	0.015±0.005	0.015±0.005
Bean pod borers	4.1	0.12	0.11±0.01	0.11±0.01	0.14±0.01	0.2	0.8	0.06±0.01	0.05±0.01	0.05±0.01
Leaf miners	N/A	N/A	0.004±0.002	0.001±0.001	0	1.6	0.4	0.02±0.01	0.01±0	0.02±0.01
Leafhoppers	7.52	**0.02**	0.08±0.01 (ab)	0.06±0.01 (a)	0.1±0.01 (b)	2.3	0.3	0.05±0.01	0.03±0.01	0.04±0.01
Spider	N/	N/A	0	0	0	0.7	0.6	0.09±0.	0.08±0.	0.09±0.

mites	A							01	012	01
Stink bugs	N/A	N/A	0.009±0.004	0	0.004±0.002	N/A	N/A	0	0	0
Thrips	4.7	0.09	0.18±0.01	0.16±0.01	0.21±0.01	11.2	**0.003**	0.07±0.01 (a)	0.05±0.01 (a)	0.09±0.02 (b)
Whiteflies	N/A	N/A	0.005±0.002	0.002±0.001	0.001±0.001	N/A	N/A	0.005±0.003	0.005±0.002	0

[1] Statistical analyses were not applicable due to low infestation scores.

3.2 Productivity

Yard-long bean production using conventional and IPM management strategies resulted in statistically the same yields and subsequent incomes during both seasons of study (Tab. 4). In the dry season, these yields and incomes were statistically higher than the no-spray control treatment (but not during the rainy season). The costs of IPM practices were much higher than the conventional treatments that farmers typically use and this resulted in significantly lower profits for IPM management compared with the other treatments. In fact, during the dry season, the high costs of IPM actually resulted in a net income loss.

Table 4. Yield, income, expenses and profit from yard-long bean cultivated under conventional, IPM and no-spray control treatments during a rainy and dry season in Cambodia. Different letters indicate significant pairwise differences between values within the same column at the $P < 0.05$ level.

Season	Treatment	Yield (kg m^{-2})	Income (\$ m^{-2})	Expenses (\$ m^{-2})	Profit (\$ m^{-2})
Rainy	Conventional	1.68±0.1	0.5±0.03	0.34±0.003 (b)	0.16±0.03 (b)
	IPM	1.73±0.13	0.52±0.04	0.66±0.01 (c)	–0.14±0.04 (a)
	No-spray control	1.52±0.1	0.46±0.03	0.31±0 (a)	0.15±0.03 (b)
	F	3.5	3.6	1706.8	110.3
	P	0.05	0.05	**<0.001**	**<0.001**
Dry	Conventional	1.74±0.11 (b)	0.75±0.05 (b)	0.36±.003 (b)	0.39±0.05 (c)
	IPM	1.71±0.12 (b)	0.74±0.06 (b)	0.66±0.01 (c)	0.07±0.06 (a)
	No-spray control	1.33±0.12 (a)	0.58±0.06 (a)	0.31±0 (a)	0.27±0.06 (b)
	F	20.2	23.9	2597	64.1
	P	**<0.001**	**<0.001**	**<0.001**	**<0.001**

4. Discussion and conclusions

The cultivation of yard-long bean in Cambodia represents an important source of income for local farmers, and our results suggest that IPM practices can produce yields comparable to conventional management practices. Global analyses across all insect pests and diseases indicate that IPM controlled them better than farmers' conventional management practices. This result supports previous IPM research on *Vigna unguiculata* that showed high performance of IPM practices (Nabirye et al., 2003; Srinivasan et al. 2019). The success of our IPM package for yard-long bean in Cambodia is especially important because pesticide imports have been increasing by 61% yearly, and banned or restricted chemicals are still widespread (Jensen et al., 2011;

Schreinemachers et al., 2017, 2015), which can have serious negative impacts on farmers' health (Kesavachandran et al., 2009; Krstevska-Konstantinova et al., 2001).

Depending on the season of study, disease pressures on yard-long bean and the efficacy of IPM and conventional management varied. During the rainy season, powdery mildew and *Rhizopus* spp. were the most serious diseases while rust the was most serious disease during the dry season (Schreinemachers et al., 2014). In all cases, the IPM controlled these diseases as well as conventional farmer practices. For powdery mildew in the rainy season, the IPM treatment controlled the disease significantly better than the no-spray control treatment. Powdery mildew has also been controlled in previous IPM studies by using cultural practices (Nathaniels et al., 2003; Oliva et al., 1999), and the same biological pesticides used in this study, including *T. harzianum* (Elad et al., 1998) and *B. subtilis* (Garcia-Gutierrez et al., 2013). Compared to the no-spray control, neither IPM nor conventional management better controlled *Rhizopus* spp. This is in contrast to previous studies in which *B. subtilis* induced disease resistance against *Rhizopus* spp. (Wang et al. 2013). Rust infestation was also high across all treatments during the dry season, despite similar expectations that *B subtilis* treatments would induce resistance (Mishra et al. 2018). In the future, seed priming with *B. subtilis* may enhance its effectiveness for combatting diseases (Mishra et al. 2018). Downy mildew, *Fusarium*, *Pythium*, and virus symptoms were all detected at very low infestation levels (< 0.1) and while our research did not show any significant differences among treatments for downy mildew or *Pythium*, previous research has highlighted the feasibility of using *Pseudomonas* spp. for biocontrol (Georgakopoulos et al., 2002), or to trigger plant systemic resistance (Monot et al., 2002).

Aphids, bean flies, and armyworms were the insect pests with the highest infestation scores in this study with aphids consistently present across both seasons, bean flies more abundant in the rainy season, and armyworms more abundant in the dry season. Each of these pests was controlled at least as well by the IPM treatment as by farmers' conventional treatments. Aphid infestations were more severe in the no-spray control plots than in either IPM or conventional treatments during both seasons of study. *Beauvaria bassiana* (Abdou et al., 2017) and orange essential oil (Mossa 2016) have both been found to be able to control aphids in previous studies. Controlling aphids may have also contributed to lower virus infestation during the rainy season in IPM compared to the no-spray control plots since aphids are important virus vectors (Damayanti et al., 2009). For bean flies, which were controlled significantly better by the IPM treatment than the no-spray control, recent research indicates that both the *Trichoderma* spp. and *B. bassiana* treatments may have been effective control strategies. *Trichoderma* spp. inoculation may stimulate the production of feeding deterrents (Mutune et al., 2016), while *B. bassiana* is directly entomopathogenic (Mascarin and Jaronski 2016). The armyworm was controlled better by the IPM treatment than the conventional treatment, which may be attributable to *B. thuringiensis* sprays, which can be highly toxic to early-instar lepidopteran larvae (Herrero et al. 2016). Leaf miners, spider mites, stink bugs and whitefly populations were controlled equally well across all treatments, with quite low infestation scores (< 0.10), and likely did not contribute much to yield loss.

Yields and income for our IPM treatment package were just as good as for conventional management with yields comparable to those obtained in commercial production systems (Coker et al., 2007). Unfortunately, IPM may come with higher costs than conventional pesticides, especially in countries where products for biocontrol are not widespread. These high input costs produced the gap in profitability between IPM and conventional management that we observed in this study. This gap could be reduced by decreasing the costs of inputs, the quantity of inputs, and/or by obtaining a premium for IPM produce. In many cases, IPM inputs are very difficult to obtain outside the research community in Cambodia and are only imported in small quantities at high prices. Increasing demand for biopesticides has the potential to lower input costs (Glare et al. 2012). The amount of IPM inputs could also be reduced in a number of ways. The pheromone traps used to attract bean pod borer were not effective in the study region of Cambodia (Srinivasan et al. 2012), and can be dropped from an IPM package, which would reduce its price by about 20%. Fewer sprays of expensive biopesticides like *B. thuringiensis* may also be warranted. *B. thuringiensis* was sprayed weekly about six or seven times during each growing season and was the single most expensive IPM input accounting for nearly one third of IPM costs. Careful scouting for young bean pod borer larvae may be able to reduce this spray window and cut expenses. Furthermore, our IPM package includes multiple biocontrol agents with redundant protection against various insects and diseases. Research has shown that deploying multiple biocontrol agents simultaneously may actually have antagonistic effects and may not control target pests and diseases better than fewer products (Xu et al. 2011). Thus, removing relatively expensive biological control products such as the orange oil from the IPM package may not significantly reduce the overall efficacy of the management plan while reducing expenses. As an alternative to reducing the costs of an IPM package, creating market premiums for IPM produce could also increase the sales price of IPM produce and the profitability of using IPM packages (Ahuja et al., 2015).

The farming systems research approach employed in this study (Darnhofer et al., 2012; Norton et al., 1999) should be considered carefully when interpreting the results of this study. In our approach, farmers were responsible for spraying the plots, harvesting their beans, and reporting marketable yields. To minimize the likelihood that farmers accidentally sprayed no-spray control plots, we systematically located them on the outer edges of fields. Some insect pests, including some aphids (Nguyen and Nansen 2018), are known to be more abundant at the edges than centers of fields. Thus, the infestation rates in no-spray control plots may be biased by them always being located along the edges of plots. Given the small size of no-spray control plots, it is also impossible to rule out possible effects of drift on the health of those plots over the course of the growing season. Spray drift may have decreased the pest and disease pressures in our small, no-spray control plots compared to if those plots had been larger. Farmers harvesting their own beans may also explain the relatively low infestation rates recorded for bean pod borers in this study compared to others in Southeast Asia (Yule and Srinivasan 2014; Srinivasan et al. 2019). While our researchers surveyed fields once per week, farmers harvested beans, including beans infested by bean pod borers, multiple times per week, which may have decreased the infestation rates apparent to researchers. Overall, while it is important to examine any systematic biases that can be introduced with a participatory, farming systems research approach, it is also important to acknowledge its benefits, which include: farmers gaining direct experience with IPM technologies, the ability to economically evaluate a relatively large number of field sites, and robust comparisons between IPM and conventional famer practices.

In conclusion, our IPM package appears to be at least as efficacious as farmers' conventional pest management practices for controlling insect pests and diseases in yard-long bean in Cambodia, and potentially in other countries in Southeast Asia with similar growing conditions. We showed that our IPM strategy had a higher efficacy than conventional pesticides overall across all arthropod pests and pathogens. However, this did not appear to have a significant impact on yields as they were similar between conventional and IPM treatments over the two seasons of study. This research represents an important step in bringing new IPM technologies to the market and field in Cambodia, and future efforts should focus on trying to minimize IPM inputs, reduce IPM input costs, and develop markets for IPM produce in order to increase its profitability. It has been suggested that the dominance of pesticides in crop protection locked agriculture into a "pesticide trap", where the switch to other management techniques represents an economic barrier (Wilson and Tisdell, 2001). Therefore, commitments by policy makers and stakeholder to promote IPM and reduce its costs are needed to unlock agriculture from conventional pesticides in Cambodia, similar to the successes achieved in Indonesia, Sweden, Norway, Denmark, Netherlands and Guatemala (Pimentel, 1997; Wilson and Tisdell, 2001).

5. Acknowledgements

Funding for this research was provided by the United States Agency for International Development under Cooperative Agreement No. AID-OAA-L-15-00001 to Virginia Tech for the Feed the Future Innovation Lab for Collaborative Research on Integrated Pest Management (IPM IL). Contents of this paper are the sole responsibility of the authors and do not necessarily reflect the views of USAID or the United States Government. The authors would like to thank Soth Sereyboth for his support of the entomological work and the many farmer cooperators without whose commitment this work would not have been possible.

References

Abdou W.L., Abdel-Hakim, E.A. and Metwally, M.M., 2017. Influence of entomopathogenic fungus *Beauveria bassiana* on the mortality, reproduction and enzyme activity of the aphid adults *Aphis craccivora* (Koch). Middle East J. Appl. Sci. 7,567-573.

Amin, F., Razdan, V.K., Mohiddin, F.A , Bhat, K. A. and Banday, S., 2010. Potential of *Trichoderma* species as biocontrol agents of soil borne fungal propagules. J. Phytol. 2, 38-41. https://updatepublishing.com/journal/index.php/jp/article/view/2179.

Ahuja, D.B., Ahuja, U.R., Singh, S.K., Singh, N., 2015. Comparison of integrated pest management approaches and conventional (non-IPM) practices in late-winter-season cauliflower in Northern India. Crop Prot. 78, 232–238. https://doi.org/10.1016/J.CROPRO.2015.08.007.

Bates, D., Mächler, M., Bolker, B., Walker, S., 2015. Fitting linear mixed-effects models using lme4. J. Stat. Softw. 67. https://doi.org/10.18637/jss.v067.i01

Bingen J., Gibbon D., 2012. Early Farming Systems Research and Extension experience in Africa and possible relevance for FSR in Europe. In: Darnhofer I., Gibbon D., Dedieu B. (Eds.) Farming Systems Research into the 21st Century: The New Dynamic. Springer, Dordrecht, pp. 49-72. https://doi.org/10.1007/978-94-007-4503-2_3.

Carvalho, F.P., 2006. Agriculture, pesticides, food security and food safety. Environ. Sci. Policy. 9, 685–692. https://doi.org/10.1016/J.ENVSCI.2006.08.002

Chagnon, M., Kreutzweiser, D., Mitchell, E.A.D., Morrissey, C.A., Noome, D.A., Van der Sluijs, J.P., 2015. Risks of large-scale use of systemic insecticides to ecosystem functioning and services. Environ. Sci. Pollut. Res. 22, 119–134. https://doi.org/10.1007/s11356-014-3277-x.

Coker, C., Ely, M., Freeman, T., 2007. Evaluation of yardlong bean as a potential newcrop for growers in the Southeastern United States. HortTechnology 17, 592–594.

Damayanti, T.A., Alabi, O.J., Naidu, R.A., Rauf, A., 2009. Severe outbreak of a yellow mosaic disease on the yard long bean in Bogor, West Java. HAYATI J. Biosci. 16, 78–82. https://doi.org/10.4308/HJB.16.2.78.

Darnhofer, I., Gibbon, D., Dedieu, B., 2012. Farming systems research: an approach to inquiry. In: Farming Systems Research into the 21st Century: The New Dynamic. Springer, Dordrecht, pp. 3-31.

de Boer Sietske, A., Diderichsen, B., 1991. On the safety of *Bacillus subtilis* and *B. amyloliquefaciens*: a review. Appl. Microbiol. Biotechnol. 36, 1–4. https://doi.org/10.1007/BF00164689.

Dinakaran, D., Gajendran, G., Mohankumar, S., Karthikeyan G., Thiruvudainambi, S., Jonathan, E.I., Samiyappan, R., Pfeiffer, D.G., Rajotte, E.G., Norton, G.W., Miller, S., Muniappan, R., 2013.

Evaluation of integrated pest and disease management package for shallots in Tamil Nadu, India – a farmer's participatory approach. J. Integr. Pest Manag. 4, 1–9. https://doi.org/10.1603/IPM12019.

Elad, Y., Kirshner, B., Yehuda, N., Sztejnberg, A., 1998. Management of powdery mildew and gray mold of cucumber by *Trichoderma harzianum* T39 and *Ampelomyces quisqualis* AQ10. BioControl. 43, 241-251. https://doi.org/10.1023/A:1009919417481.

FAOSTAT, 2014. FAOSTAT Statistics Database. FAO, Rome.

García-Gutiérrez, L., Zeriouh, H., Romero, D., Cubero, J., de Vicente, A., Pérez-García, A., 2013. The antagonistic strain *Bacillus subtilis* UMAF6639 also confers protection to melon plants against cucurbit powdery mildew by activation of jasmonate- and salicylic acid-dependent defence responses. Microb. Biotechnol. 6, 264-274. https://doi.org/10.1111/1751-7915.12028.

Georgakopoulos, D.G., Fiddaman, P., Leifert, C., Malathrakis, N.E., 2002. Biological control of cucumber and sugar beet damping-off caused by Pythium ultimum with bacterial and fungal antagonists. J. Appl. Microbiol. 92, 1078–1086. https://doi.org/10.1046/j.1365-2672.2002.01658.x.

Glare, T., Caradus, J., Gelernter, W., Jackson, T., Keyhani, N., Köhl, J., Marrone, P., Morin, L., Stewart, A., 2012. Have biopesticides come of age? Trends Biotechnol. 30, 250-258. https://doi.org/10.1016/j.tibtech.2012.01.003.

Herrero, S., Bel, Y., Hernández-Martínez, P., Ferré, J., 2016. Susceptibility, mechanisms of response and resistance to *Bacillus thuringiensis* toxins in *Spodoptera* spp. Curr. Opin. Insect Sci. 15, 89-96. https://doi.org/10.1016/j.cois.2016.04.006.

Jensen, H.K., Konradsen, F., Jørs, E., Petersen, J.H., Dalsgaard, A., 2011. Pesticide use and self-reported symptoms of acute pesticide poisoning among aquatic farmers in Phnom Penh, Cambodia. J. Toxicol. 2011, 639814. https://doi.org/10.1155/2011/639814.

Kesavachandran, C.N., Fareed, M., Pathak, M.K., Bihari, V., Mathur, N., Srivastava, A.K., 2009. Adverse Health Effects of Pesticides, in: Whitacre, D.M., (Ed.), Agrarian Populations of Developing Countries. Springer, Boston, MA, pp. 33–52. https://doi.org/10.1007/978-1-4419-0028-9_2.

Kongjaimun, A., Kaga, A., Tomooka, N., Somta, P., Shimizu, T., Shu, Y., Isemura, T., Vaughan, D.A., Srinives, P., 2012. An SSR-based linkage map of yardlong bean (*Vigna unguiculata* (L.) Walp. subsp. *unguiculata* Sesquipedalis Group) and QTL analysis of pod length. Genome 55, 81–92. https://doi.org/10.1139/g11-078.

Koul, O., Walia, S., and Dhaliwal, G.S., 2008. Essential oils as green pesticides: potential and constraints. *Biopestic. Int.* 41, 63–84.

Krstevska-Konstantinova, M., Charlier, C., Craen, M., Caju, M., Heinrichs, C., Beaufort, C., Plomteux, G., Bourguignon, J.P., 2001. Sexual precocity after immigration from developing countries to Belgium: evidence of previous exposure to organochlorine pesticides. J. Pathol. Microbiol. Immunol. 109, S135–S143. https://doi.org/10.1111/j.1600-0463.2001.tb05759.x.

Mascarin, G.M. and Jaronski, S.T. 2016. The production and uses of *Beauveria bassiana* as a microbial insecticide. World J. Microbiol. Biotechnol. 32: 177. https://doi.org/10.1007/s11274-016-2131-3.

Matson, P.A., Parton, W.J., Power, A.G., Swift, M.J., 1997. Agricultural intensification and ecosystem properties. Science 277, 504–9. https://doi.org/10.1126/SCIENCE.277.5325.504

*Mishra, R.K., Bohra, A., Kamaal, N., Kumar, K., Gandhi, K., Sujayanand, G.K., Saabale, P.R., Naik, S.J.S., Sarma, B.K., Kumar, D., Mishra, M., Srivastava, D. K., Singh, N.P., 2018. Utilization of biopesticides as sustainable solutions for management of pests in legume crops: achievements and prospects. Egypt J. Biol. Pest Control. 28, 3. https://doi.org/10.1186/s41938-017-0004-1

Monot, C., Pajot, E., Le Corre, D., Silué, D., 2002. Induction of systemic resistance in broccoli (*Brassica oleracea* var. botrytis) against downy mildew (*Peronospora parasitica*) by avirulent isolates. Biol. Control. 24, 75–81. https://doi.org/10.1016/S1049-9644(02)00006-3.

Mossa, A-T. H. 2016. Green pesticides: essential oils as biopesticides in insect-pest management. J. Environ. Sci. Technol. 9, 354-378. DOI: 10.3923/jest.2016.354.378.

Mutune, B., Ekesi, S., Niassy, S., Matiru, V., Bii, C., Maniania, N.K., 2016. Fungal endophytes as promising tools for the management of bean stem maggot *Ophiomyia phaseoli* on beans Phaseolus vulgaris. J. Pest Sci. 89, 993–1001. https://doi.org/10.1007/s10340-015-0725-4.

Nabirye, J., Nampala, P., Ogenga-Latigo, M.., Kyamanywa, S., Wilson, H., Odeke, V., Iceduna, C., Adipala, E., 2003. Farmer-participatory evaluation of cowpea integrated pest management (IPM) technologies in Eastern Uganda. Crop Prot. 22, 31–38. https://doi.org/10.1016/S0261-2194(02)00094-7.

Nathaniels, N.Q.R., Sijaona, M.E.R., Shoo, J.A.E., Katinila, N., 2003. IPM for control of cashew powdery mildew in Tanzania. I: Farmers' crop protection practices, perceptions and sources of information. Int. J. Pest Manag. 49, 25–36. https://doi.org/10.1080/713867836.

Nguyen, H.D.D. & Nansen, C., 2018. Edge-biased distributions of insects. A review. Agron. Sustain. Dev. 38, 11. https://doi.org/10.1007/s13593-018-0488-4.

Norton, G.W., Rajotte, E.G. & Gapud, V. 1999. Participatory research in integrated pest management: Lessons from the IPM CRSP. Agric. Human Val. 16, 431. https://doi.org/10.1023/A:1007608019218.

Oliva, J., Navarro, S., Navarro, G., Cámara, M.., Barba, A., 1999. Integrated control of grape berry moth (*Lobesia botrana*), powdery mildew (*Uncinula necator*), downy mildew (*Plasmopara viticola*) and grapevine sour rot (*Acetobacter* spp.). Crop Prot. 18, 581–587. https://doi.org/10.1016/S0261-2194(99)00064-2.

Parsa, S., Morse, S., Bonifacio, A., Chancellor, T.C.B., Condori, B., Crespo-Pérez, V., Hobbs, S.L.A., Kroschel, J., Ba, M.N., Rebaudo, F., Sherwood, S.G., Vanek, S.J., Faye, E., Herrera, M.A., Dangles, O., 2014. Obstacles to integrated pest management adoption in developing countries. Proc. Natl. Acad. Sci. 111, 3889–94. https://doi.org/10.1073/pnas.1312693111.

Pimentel, D., 2005. Environmental and economic costs of the application of pesticides primarily in the United States. Environ. Dev. Sustain. 7, 229–252. https://doi.org/10.1007/s10668-005-7314-2.

Pimentel, D., 1997. Techniques for Reducing Pesticide Use : Economic and Environmental Benefits. Wiley, Hoboken, NJ.

Pretty, J., Bharucha, Z., 2015. Integrated pest management for sustainable intensification of agriculture in Asia and Africa. Insects 6, 152–182. https://doi.org/10.3390/insects6010152

R Core Team, 2013. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.

Schreinemachers, P., Srinivasan, R., Wu, M., Bhattarai, M., Patricio, R., Yule, S., Quang, V.H., Hop, B., 2014. Safe and sustainable management of legume pests and diseases in Thailand and Vietnam: A situational analysis. Int. J. Trop. Insect Sci. 34, 88-97. doi:10.1017/S174275841400023X.

Schreinemachers, P., Afari-Sefa, V., Heng, C.H., Dung, P.T.M., Praneetvatakul, S., Srinivasan, R., 2015. Safe and sustainable crop protection in Southeast Asia: status, challenges and policy options. Environ. Sci. Pol., 54, 357-366. https://doi.org/10.1016/j.envsci.2015.07.017.

Schreinemachers, P Chen, H., Nguyen, T.T.L., Buntong. B., Bouapao, L., Gautam, S., Le, N.T., Pinn, T., Vilaysone, P., Srinivasan, R. 2017. Too much to handle? Pesticide dependence of smallholder vegetable farmers in Southeast Asia. Sci. Total Environ. 593-594, 470-477. https://doi.org/10.1016/j.scitotenv.2017.03.181.

Siegel, J.P., 2001. The mammalian safety of *Bacillus thuringiensis*- based insecticides. J. Invertebr. Pathol. 77, 13–21. https://doi.org/10.1006/JIPA.2000.5000.

Srinivasan R, Yule S, Chang J-C, Malini P, Lin MY, Hsu YC, Schafleitner R. 2013. Towards developing a sustainable management strategy for legume pod borer, *Maruca vitrata* on yard–long bean in Southeast Asia. In: Holmer R, Linwattana G, Nath P, Keatinge JDH, (Eds.) Proceedings of the Regional Symposium on High Value Vegetables in Southeast Asia: Production, Supply and Demand (SEAVEG2012), Publication No. 12-758. AVRDC – The World Vegetable Center, Taiwan. pp. 76-82.

Srinivasan**,** R. Paola, S., Lin, M-y., Hy, H.C., Sareth, K. , Sor, S. 2019. Development and validation of an integrated pest management strategy for the control of major insect pests on yard-long bean in Cambodia**.** Crop Prot**.** 116, 82-91, 2019. https://doi.org/10.1016/j.cropro.2018.10.015.

Stern, V.M., Smith, R.F., van den Bosch, R., Hagen, K.S., 1959. The integration of chemical and biological control of the spotted alfalfa aphid: The integrated control concept. Hilgardia 29, 81–101. https://doi.org/10.3733/hilg.v29n02p081.

Tantasawat, P., Trongchuen, J., Prajongjai, T., Seehalak, W., Jittayasothorn, Y., 2010. Variety identification and comparative analysis of genetic diversity in yardlong bean (*Vigna* *unguiculata* spp. *sesquipedalis*) using morphological characters, SSR and ISSR analysis. Sci. Hortic. 124, 204–216. https://doi.org/10.1016/J.SCIENTA.2009.12.033

Timprasert, S., Datta, A., Ranamukhaarachchi, S.L., 2014. Factors determining adoption of integrated pest management by vegetable growers in Nakhon Ratchasima Province, Thailand. Crop Prot. 62, 32–39. https://doi.org/10.1016/J.CROPRO.2014.04.008.

Van den Berg, H., Jiggins, J., 2007. Investing in Farmers—The impacts of farmer field schools in relation to integrated pest management. World Dev. 35, 663–686. https://doi.org/10.1016/J.WORLDDEV.2006.05.004.Wang, X., Wang, P., Jin, P., Zheng, Y., 2013. Investigating the efficacy of *Bacillus subtilis* SM21 on controlling *Rhizopus* rot in peach fruit. Int. J. Food Microbiol. 164, 141-147. https://doi.org/10.1016/j.ijfoodmicro.2013.04.010.

Way, M.., van Emden, H.., 2000. Integrated pest management in practice — pathways towards successful application. Crop Prot. 19, 81–103. https://doi.org/10.1016/S0261-2194(99)00098-8.

Wilson, C., Tisdell, C., 2001. Why farmers continue to use pesticides despite environmental, health and sustainability costs. Ecol. Econ. 39, 449–462. https://doi.org/10.1016/S0921-8009(01)00238-5.

Xu, X.-M., Jeffries, P., Pautasso, M., Jeger, M.J., 2011. Combined use of biocontrol agents to manage plant diseases in theory and practice. Phytopathology. 101, 1024–31. https://doi.org/10.1094/PHYTO-08-10-0216

Yule, S., Srinivasan, R., 2014. Combining bio-pesticides with chemical pesticides to manage legume pod borer (*Maruca vitrata*) on yard-long bean in Thailand. Int. J. Pest Manag. 60, 67–72. https://doi.org/10.1080/09670874.2014.900707.

Zimmermann, G., 2007. Review on safety of the entomopathogenic fungi *Beauveria bassiana* and

Beauveria brongniartii. Biocontrol Sci. Technol. 17, 553–596.
https://doi.org/10.1080/09583150701309006.