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Efficacy of Eco-Friendly Products Against S*podoptera litura* (Fab) (Lepidoptera: Noctuidae) in Cabbage: A Field Trial in Nepal

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ABSTRACT

The tobacco caterpillar (*Spodoptera litura*) is one of the most economically devastating insect pests in Nepal, where it preys on cabbage and other crops. This study evaluated the efficacy of two entomopathogens (*Beauveria bassiana* and *Bacilus thuringiensis*), two plant-based commercial products (Karanjin and Azadirachtin), chinaberry leaf extract, and Jholmal (mixture of fermented animal urine and herbs) against *S. litura* in cabbage under field conditions in Chitwan, Nepal, and compared the efficacy with that of a commercial toxic product "All Killer" (Cypermethrin 10%). Karanjin and Jholmal were found to be significantly more effective against *S. litura* larvae than the entomopathogen-based products and chinaberry leaf extract; fewer cabbage heads were damaged by *S. litura* when Karanjin, Jholmal, and All Killer were applied. Cabbage yields in plots treated with Karanjin, Jholmal, and All Killer were similar and were significantly higher than those in plots treated with other tested items. This study reveals that Karanjin and Jholmal are effective eco-friendly alternatives to All Killer in controlling *S. litura* on cabbage in the field.

Key words: Cabbage, yield, *Spodoptera litura,* pest,population

Introduction

Cabbage (*Brassica oleracea* var. *capitata*) is the most widely cultivated leafy vegetable in Nepal, where the total area cultivated and the amount of cabbage produced are 27,445 ha and 468,284 mt, respectively. The area of cabbage cultivation represents 11% of the total amount of vegetables cultivated by area in Nepal, and the crop contributes to 14% of the total amount of vegetables produced (Anonymous, 2013). However, several factors currently limit cabbage production in Nepal, and insect pests contribute to a 20% yield loss (Neupane, 1999). Of the various economically important cabbage insect pests, the tobacco caterpillar (*Spodoptera litura*) is the most serious polyphagous pest. It attacks numerous crop species in Nepal (Neupane, 1999, 2000), but it damages cabbage throughout all growth stages by causing irregular bore holes on

the developing cabbage head and producing larval excrement (Neupane, 2000).

S. litura is known to be a serious pest affecting many agricultural crops in Asia and is widely distributed throughout the tropical and temperate regions of Asia, Australasia, and the Pacific Islands (Mushtaq *et al.*, 2007; Meagher *et al.*, 2008). To reduce its population, several pest control methods, including chemical pesticides, are currently practiced in Nepal. However, chemical pesticide application is a public health and environmental concern. Therefore, researchers are searching for alternative strategies for managing this pest that also prevent economical losses. This study, therefore, evaluated the efficacy of natural products, two entomopathogens and two biopesticides against *S. litura* under field conditions in Chitwan, Nepal.

Materials and Methods

Cabbage field

Thirty-day old cabbage seedlings (variety: Green Coronet) were transplanted in 32 plots, each measuring 3 × 3 m (with a row spacing of 60 cm and plant spacing of 45 cm), by using recommended cultural practices (Anonymous, 2011). Fertilizer was applied at a rate of 50:80:60 kg NPK/ha with farm yard manure at a rate of 20 T/ha. Each plot contained 5 rows of 15 cabbages, and yield data of the middle row were recorded. The experiment was conducted using the randomized complete block design, eight treatments, and four replications. A spacing of 1 m was set between all plots.

Jholmal

Jholmal is a fermented mixture comprising various plant materials, water, and cattle urine and is made using 500 g of leaves from each of: *Azadirachta indica*, *Melia azaderach*, *Adhatod avasica*, *Osimum sanctum*, *Lycopersicon esculentum*, *Artemisia vulgaris*, *Acorus calamus*, *Tagete* sp., *Sapium insigne*, *Chrysanthemum* sp., and *Vitex negundo*. In addition, prepared using 500 g of leaves from *Allimum sativum*, *Capsicum annuum*, and *Zingiber officinale* tubers was added to the mixture.

In this study, plant parts were washed with normal tap water after collection, left to dry at room temperature for 24 h, then chopped finely (approximately 1 cm in length), mixed, and placed in a black plastic container. Eight liters of cattle urine collected 24 h earlier were then added to the container, together with 10 L of tap water, and the mixture was mixed well. The container was then tightly closed with a lid and left for 30 days under room conditions to allow the mixture to decompose and ferment.

Other test items

With the exception of Jholmal and chinaberry (*Melia azaderach*) leaf extract, all other test items (All Killer, Derisom, Racer, Bilop, and Margosom) were bought from the local market. Details of the test items used in this study are listed in Table 1.

To prepare chinaberry leaf extract, 500 g of leaves were washed and left to dry at room temperature for 24 h. They were then chopped finely (approximately 1 cm in length), mixed with 1 L of water, and left for 24 h. The mixture was subsequently filtered with a fine cotton cloth, and the extract used for further applications of the testes.

Preparation and application of test items

A total of 4 mL of All Killer and 10 mL of Margosom were added to 1 L of water in separate containers and stirred thoroughly; a further 1 L of water was then added to the containers separately to obtain the final spray. In addition, 10 g of *B. bassiana* and 4 g of *Bt.* var *kurstaki* were added to 2 L of water in separate containers and mixed through stirring. Furthermore, 500 mL of both chinaberry leaves and Jholmal were added to 2.5 L of water to prepare the final spray mixture.

The test items were sprayed on cabbage plants in the evening with a portable hand compression sprayer (Agricart, India), and a total of four spray sessions were conducted at the rate of 540 mL/plot at 10-day intervals.

Cabbage head damage and yield

Head damage at harvest was estimated using the method reported by Fail *et al.* (2008). The cabbage heads were observed by peeling off leaves until no damage was seen on four consecutive leaves. The extent of damage was noted by examining the proportion of damage to the underside surface of the leaf, with scores ranging from 0 to 1. Plants without damaged

Table 1. Details of test items used in the study

Trade name (Common name)	Chemical/scientific name	Manufacturer	Formulation	Concentration
Racer (Entomopathogenic fungus)	*Beauveria bassiana*	Agrilife, India	1.15% spores w.p.	5 gm/L
Biolep (Entomopathogenic bacteria)	*Bacillus thuringiensis* var *kurstaki*	Agrilife, India	5-8% δ-endotoxin	2 gm/L
Derisom (Karanjin biopesticide)	Karanjin 20,000 ppm from *Derris indica*	Agrilife, India	20,000 ppm	2 mL/L
Margosom (Neem biopesticide)	Azadirachtin	Agrilife, India	0.15% EC	5 mL/L
Chinabery leaf extract	*M. azadirach*	Local botanical	Liquid	1:5
Jholmal	Mixture of herbs and cattle urine	Locally made product	Liquid	1:5
All Killer	Cypermethrine 10EC	Nepal Agro Industries	10 % EC	2 mL /L
Control (Water)	Water			

leaves were weighed, and their total yields were converted into tons per hectare.

Data recording and analysis

Each treatment tested was applied four times at 10-day intervals (20, 30, 40, and 50 days after transplantation [DAT]). Furthermore, changes in *S. litura* population after each treatment were recorded 10 days after treatment application. Insect population data were taken from five randomly selected plants in each plot, each of which was marked with a bamboo stick, and the *S. litura* population was counted over the entire plant. The data collected were analyzed using SAS (2015), and means separation was conducted using the *Student-Newman-Keuls* method at a significance level of 5%.

The average temperature, humidity, and rainfall during the study period were 24.35°C, 83.54%, and 1.7 mm, respectively.

The numbers of insects killed by the test items over control were calculated using the following equation:

$$Mortality\ (\%) = \frac{\text{No. of insects died at Control} - \text{No. of insects died at test item}}{\text{No. of insects died in Control}} \times 100$$

Results

S. litura population

At 30 DAT, the numbers of *S. litura* larvae observed on plants sprayed with each of the treatments were significantly different (P=0.001, F=358.82). However, no significant differences in the numbers of *S. litura* larvae were observed between the Margosom and Derisom plots and the Derisom and All Killer plots. The least and most significant numbers of *S. litura* were observed in the All Killer-treated and control plots, respectively. Compared with the control, at 30 DAT, percentages of *S. litura* larvae controlled by Racer, Biolop, Derisom, Margosom, chinaberry leaf extract, Jholmal, and All Killer were 46.12%, 25%, 73.71%, 69.83%, 61.21%, 54.31%, and 75.00%, respectively (Table 2).

At 40 DAT, the numbers of *S. litura* larvae observed on plants sprayed with each of the treatment solutions were significantly different (P<0.001, F=465.21). However, no significant differences in the numbers of *S. litura* larvae were observed between the Racer and Margosom plots and the Derisom and chinaberry leaf extract plots. Compared with the control, at 40 DAT, the percentages of *S. litura* larvae controlled by Racer, Bilop, Derisom, Margosom, chinaberry leaf extract, Jholmal, and All Killer were 51.75%, 44%, 65.5%, 51.50%, 65.50%, 61.75%, and 72.50%, respectively (Table 2).

Table 2. Effect of different products on the population of *S. litura* observed under field conditions

Treatments	No. of *S. litura* larvae observed (mean ± SE)[a]			
	30 DAT[b]	40 DAT	50 DAT	60 DAT
Racer	31.25 ± 0.63cD	48.25 ± 0.48cB	73.25 ± 0.48bA	44.25 ± 0.85bC
Bilop	43.5 ± 0.65bB	56 ± 0.58bA	37.25 ± 0.85cC	39.75 ± 1.11bC
Derisom	15.25 ± 0.48gfD	34.5 ± 0.65eB	38.25 ± 0.63cA	24.5 ± 0.96dC
Margosom	17.5 ± 0.65fD	48.5 ± 0.65cA	38.5 ± 1.04cC	44.25 ± 1.11bB
Chinaberry leaf extract	22.5 ± 1.32eD	35.5 ± 0.65eB	27.5 ± 0.65dC	41.75 ± 1.03bA
Jholmal	26.5 ± 0.65dC	38.25 ± 1.25dA	17.5 ± 0.65eD	30 ± 0.91cB
All Killer	14.5 ± 0.65gD	27.5 ± 0.65fC	30.5 ± 0.65dB	34.25 ± 1.31cA
Control	58 ± 1.08aB	100 ± 2.27aA	102 ± 3.08aA	98.25 ± 3.12aA

[a] Mean in the same column (lower case) and same row (upper case) with same letter are not significant at a significance level of 5%.
[b] DAT = days after transplantation.

At 50 DAT, the numbers of *S. litura* larvae observed on plants sprayed with each of the treatments were significantly different (P<0.001, F=473.6). However, no significant differences in the numbers of *S. litura* larvae were observed between the Biolop, Derisom, and Margosom plots and the chinaberry leaf extract and All Killer plots. Compared with the control, at 50 DAT, the percentages of *S. litura* larvae controlled by Racer, Bilop, Derisom, Margosom, chinaberry leaf extract, Jholmal, and All Killer were 28.19%, 63.48%, 62.5%, 62.25%, 73.04%, 82.84%, and 70.10%, respectively (Table 2).

At 60 DAT, the numbers of *S. litura* larvae observed on plants sprayed with each of the treatments were significantly different (P<0.001, F=273.92). However, no significant differences in the numbers of *S. litura* larvae were observed between the Racer, Biolop, Margosom, and chinaberry leaf extract plots and the Jholmal and All Killer plots. Compared with the control, at 60 DAT, the percentages of *S. litura* larvae controlled by Racer, Bilop, Derisom, Margosom, chinaberry leaf extract, Jholmal, and All Killer were 54.96%, 59.54%, 75.06%, 54.96%, 57.51%, 69.47%, and 65.14%, respectively (Table 2).

A comparison of the numbers of *S. litura* larvae observed for each test item on different observation dates revealed that the most significantly high numbers of larvae were observed as follows: at 40 DAT: in Biolop (P<0.001, F=102.31), Margosom (P<0.001, F= 240.39), and Jholmal (P<0.001, F=91.48) plots; at 50 DAT: in Racer (P<0.001, F=779.79) and Derisom (P<0.001, F=219.28) plots; at 60 DAT: in chinaberry leaf extract (P<0.001, F=77.18) and All Killer (P<0.001, F=98.87) plots (Table 2).

Cabbage head damage and yield

At the end of the study period (60 DAT), significant differences were observed in the numbers of cabbage leaves damaged by *S. litura* larvae with respect to the various treatments (P<0.001, F=12.12). However, there were no significant differences in the numbers of cabbage leaves damaged by *S. litura* larvae between the Racer, Biolop, Margosom, and chinaberry leaf extract plots and the Derisom, Jholmal, and All Killer plots (Table 3).

At harvest, the average cabbage yields from the different treatment plots were significantly different (P<0.001, F=441.74); however, the differences in the yields between the plots within the following groups were nonsignificant: Derisom, Jholmal, and All Killer plots; Margosom and chinaberry leaf extract plots; and Racer and Biolop plots (Table 3).

Table 3. Effect of different products on the numbers of leaves damaged and cabbage yield under field conditions

Treatments	No. of leaves damaged (mean ± SE)[a]	Yield (T Ha^{-1}) (mean ± SE)[a]
Racer	5.00 ± 0.41b	29.75 ± 0.63c
Bilop	4.75 ± 0.48b	30.75 ± 1.03c
Derisom	3.25 ± 0.25c	61.50 ± 0.87a
Margosom	4.50 ± 0.29b	41.00 ± 0.71b
Chinaberry leaf extract	5.00 ± 0.41b	38.75 ± 0.85b
Jholmal	3.00 ± 0.41c	63.25 ± 1.11a
All Killer	2.75 ± 0.25c	62.00 ± 0.08a
Control	6.75 ± 0.48a	16.75 ± 0.63d

[a] Mean in the same column with same letter are not significant at a significance level of 5%.

Discussion

The numbers of larvae were consistently higher in the control plot than in the other treatment plots. However, numbers of *S. litura* larvae first increased in the Racer applied plots, reached a maximum at 50 DAT, then decreased at 60 DAT, at which point the larva population became statistically similar to that of other treatment plots. Racer is a *B. bassiana* product, and its larvae-killing speed was found to be substantially slower than that of the other toxic products; a similar result was previously reported in other studies (Sood *et al.,* 2001; Asi *et al.,* 2013). Studies have also reported variations in the susceptibility of *S. litura* larvae against *B. bassiana* with respect to environmental factors, particularly temperature and humidity.

The numbers of *S. litura* larvae observed in the Bilop-treated plots reached a maximum at 40 DAT and then decreased at 50 DAT, at which point the larva population became statistically similar to that of other treatment plots. This study found that *B. thuringiensis* was faster in controlling *S. litura* larvae than *B. bassiana*. Alotaibi (2013) reported that *B. thuringiensis* was more effective against lepidopteran pests in early stages, which supports the results with respect to *B. thuringiensis* in this study.

The numbers of *S. litura* larvae observed in the Derisom-treated plots were consistently fewer than those in other treatment plots in this study, with the exception of the All Killer-treated plot. Reena and Singha (2012) reported the toxicity of *D. indica* extract to *Helicoverpa armigera* larvae, and we observed that Karanjin was also toxic to *S. litura* larvae, which resulted in the decreased numbers in the Derisom-treated plots.

Although fewer larvae were observed at the start of the observation period, the numbers of *S. litura* larvae observed in Margosome-treated plots were similar to those observed in the Biolep-treated plots. This study found that Margosom was faster in controlling *S. litura* larvae than *B. bassiana.* Pandey and Summarwar (2016) reported that the leaf extract of *A. indica* caused *S. litura* larval and pupal mortality under laboratory conditions. They also reported that Azadirachtin, a bioactive compound of *A. indica*, is highly toxic to the larvae and pupae of *S. litura*.

The numbers of larvae observed in the chinaberry leaf extract-treated plot were consistently fewer than those in the control plot. Limonoids, a class of tetranortriterpenes, in the leaves and seeds of *M. azedarach* are the major toxic compounds of this plant, causing stomach poisoning, damage to the midgut epithelium, and high larval mortality of *S. litura* (Kumar *et al.,* 2006). Similar results were reported by Carpinella *et al.,* (2003) when testing against the *Epilachnapa enulata* beetle.

The numbers of *S. litura* larvae observed in the Jholmal-treated plots were lower than those

in most other treatment plots. Jholmal is a mixture of fermented cattle urine, the leaves of *A. indica*, *M. azaderach*, *A. avasica*, *O. sanctum*, *L. esculentum*, *A. vulgaris*, *A. calamus*, *Tagete s*sp., *S. insigne*, *Chrysanthemum* sp., and *V. negundo*, and the fruits of *A. sativum*, *C. annuum* and *Z. officinale*. Many studies have reported the insecticidal value of parts of these plants or of the essential oils extracted from them (Catherine 1997, Ananda *et al*. 2006, Dadji *et al*. 2007, Sahaf *et al*. 2008, Kumar *et al*. 2012, Okwute, 2012, Shabir *et al*. 2013, Mya *et al*. 2016, Pandey and Summarwar 2016). In addition, Katuwal *et al.* (2012) reported that urine-mix was efficient against *Plutella xylostella* under field conditions and determined that fermented urine-mix contained a number of chemical compounds (3-and 4-methyl phenol, 3-ethylphenol, 3-n-propylphenol, and 2-methoxyphenol). These compounds may have had an effect on the morality of *S. litura* in this study; however, details of the mode of action of Jholmal is yet to be investigated.

The Cypermethrin-based product "All Killer" was very efficient in controlling the numbers of *S. litura* larvae. However, later observations revealed that its efficacy was similar to that of Jholmal and Derisom.

Yields from the Jholmal, All Killer, and Derisom plots were statistically similar. This result indicates that the application of biopesticide is effective in providing the same cabbage yield as using chemical controls.

In conclusion, *S. litura* is the most critical cabbage pest in commercial cabbage growing areas of Nepal, which attacks cabbage in all growth stages, from seedlings to head formation. The results of this study indicate that Jholmal and Derisom can be effectively used in controlling this pest because these eco-friendly products were as effective as All Killer in controlling populations of *S. litura* and providing similar cabbage yields.

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環保產品對尼泊爾高麗菜的斜紋夜盜蟲 ***Spodoptera litura* (Fab)** 防治功效

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摘　　要

在尼泊爾 (Nepal)，斜紋夜盜蟲 (*Spodoptera litura*) 是危害高麗菜和其多種農作物最嚴重的雜食性害蟲。本研究主要在尼泊爾奇旺 (Chitwan) 地區的高麗菜田進行試驗，評估了兩種蟲生病原體，白僵菌 (*Beauveria bassiana*) 和蘇力菌 (*Bacilus thuringiensis*)、兩種植物為主的商業產品 (Karanjin 和 Azadirachtin)、苦楝樹葉萃取物及 Jholmal (發酵的動物尿液和草藥混合物) 等針對斜紋夜盜蟲的防治功效，並和商業農藥產品"All Killer" (Cypermethrin 10%) 做比較。結果發現 Karanjin 和 Jholmal 對斜紋夜盜蟲幼蟲的防治效能明顯優於蟲生病原體與植物和苦楝樹葉萃取物。使用 Karanjin、Jholmal 和 All Killer 時，高麗菜被斜紋夜盜蟲損害的數量有減少，且三者間產量相似，均明顯地高於其他測試項目。這項研究顯示，Karanjin 和 Jholmal 是更有效的環保替代產品，可以控制高麗菜田的斜紋夜盜蟲。

關鍵詞：高麗菜、生產量、斜紋夜盜蟲、族群