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Evaluation of *Bacillus thuringiensis* microcapsules for the management of tobacco caterpillar, *Spodoptera litura* (F.) in castor

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Abstract
Castor is a non-edible oilseed crop of major industrial significance in India. The crop is attacked by a number of insect pests which impose a great limitation in realizing the potential productivity. Among the insect pests infesting castor, *Spodoptera litura* (F.) (Lepidoptera: Noctuidae), is the most notorious pest. Although synthetic chemical insecticides remain indispensable to avert the crop losses due to the insect pest, the over reliance on chemical insecticides is being viewed with concern due to the increasing problem of detrimental effect on non-target organisms and environmental pollution. Hence in recent years considerable efforts are being made worldwide to find eco-friendly pest management options substitutes for these synthetic insecticides. Hence, the present study was conducted to evaluate the efficacy of *Bacillus thuringiensis* microcapsules against the polyphagous pest, *viz*., tobacco caterpillar (*Spodoptera litura*) in castor. A caged pot culture experiment on castor revealed that *Bacillus thuringiensis* microcapsules, *viz*., CC-1+Btk (Chitosan + lignosulphonate + manganous sulphate + Btk) and CC-2+Btk (Chitosan + lignosulphonate + carboxymethylcellulose + manganous sulphate + Btk) were found effective and recorded higher percent larval mortality (81.00 to 81.20%) of *S. litura* as compared to Bt-technical @ 1 g/l (70.00% larval mortality), while no mortality was observed in the untreated control. The present study will be useful for the eco-friendly management of *S. litura* infesting agricultural and horticultural crops.

Keywords: *Bacillus thuringiensis*, bio-efficacy, microcapsules, pest management, *Spodoptera litura*

Introduction
India has the largest area and highest productivity of castor (*Ricinus communis* L.) in the world. Among the biological constraints limiting castor production, insect pests undoubtedly dominate the scenario. The avoidable yield losses due to insect pests on castor ranged from 17 to 63% (Madhuri *et al*., 2022; Lakshminarayana and Duraimurugan, 2014; Duraimurugan and Sujatha, 2023) [7, 5, 2]. Among them, the tobacco caterpillar [*Spodoptera litura* (F.)] is of greater economic importance. The larvae damage crops through defoliation (Duraimurugan, 2018) [1]. The newly hatched larvae feed gregariously on a single leaf on the same plant for a short period before dispersal. Gregarious form while feeding on the underside of leaves gives a mesh-like appearance to leaves. The full-grown larvae voraciously feed on castor leaves and cause complete defoliation. Sometimes it also damages capsules during a severe outbreak of the pest (Duraimurugan and Srinivas, 2017) [3]. Chemical insecticides have become indispensable in crop protection owing to their immediate and spectacular effects. On many occasions, they have been used as the principal method. However, the indiscriminate and injudicious use of broad-spectrum insecticides is causing many environmental and health hazards. In order to enhance production and also address the growing concern over safe food and the environment, there is an urgent need to circumvent insect pests' damage by following eco-friendly pest management strategies (Varaprasad and Duraimurugan, 2018) [1]. Microbial biocontrol agents, particularly entomopathogenic bacteria, offer considerable promise for eco-friendly insect pest management (Vimala Devi *et al*., 2021) [10]. There are many commercially available biopesticides for crop protection, especially those based on *Bacillus thuringiensis* (Bt) formulations, which have been widely used as topical insecticides to safeguard crops from insect pests throughout the world (Pradeep Kumar *et al*., 2021) [9]. Microencapsulation of microbials using natural polymers has recently gained importance in order to reduce dose requirements and increase formulation stability and efficacy (Jhones *et al*., 2021) [6]. Keeping in view of the above, the present research work was conducted to study the bioefficacy of *B. thuringiensis* microcapsules for the management of *S. litura* in castor.

Materials and Methods

A pot culture experiment was carried out at ICAR-Indian Institute of Oilseeds Research, Rajendranagar, Hyderabad to assess the efficacy of different doses of two *Bacillus thuringiensis* var. *kurstaki* (Btk) microcapsules, *viz*., CC-1+Btk and CC-2+Btk along with Btk technical, commercial Btk formulations, and conventional insecticides against tobacco caterpillar (*S. litura*). The experimentation was conducted in a completely randomized design with thirteen treatments and replicated three times. The treatments included three doses each for the two Btk microcapsules *viz*., CC-1+Btk and CC-2+Btk along with biopesticide checks [Btk-technical, Btk-WDG, Btk SC, Btk (Delfin®)], insecticide checks (flubendiamide profenophos (2 ml/l) and untreated control (water spray) (Table 1).

Seeds of the castor hybrid ICH-66 were sown in earthern pots, and the treatments were imposed when the plants were 45 days old. The plants were sprayed thoroughly using a Ganesh hand compression sprayer. After the spray fluids had dried, 5-day-old larvae of tobacco caterpillar (*S. litura*) in the respective experiments were released onto the plants at a rate of 20 larvae per potted plant using a camel's hairbrush. To prevent the larvae from escaping, the plant and pot were covered with a nylon net cage, which provided proper ventilation and aeration. Observations on larval mortality were recorded at 24-hour intervals for up to 5 days after treatment. The percent mortalities were transformed to arcsine percentages and subjected to statistical interpretations adopting a completely randomized design (CRD). The mean values of treatments were then separated by least significant difference (LSD).

Results and Discussion

The efficacy of promising Btk microcapsules along with Btk-technical and commercial Btk formulations, was evaluated against *S. litura* in a castor under caged pot experiment, and the results are presented in Table 1. All the treatments were found effective against *S. litura* larvae as compared to the untreated control. Among the different doses of Btk microcapsules tested, CC-1+Btk @ 1.5 g/l and CC-2+Btk @ 1.5 g/l recorded higher percent *S. litura* larval mortality (81.20 and 81.00%, respectively) (Fig. 1 and Fig. 2), while the microcapsules at lower doses (0.75 and 1.0 g/l) recorded 71.00 to 79.00% larval mortality. Among the commercial Btk formulations, Btk-WDG showed 86.66% larval mortality, followed by Btk (Delfin) and Bt-127 SC (81.66%), while the larval mortality in Btk-technical was 70.00%. The conventional insecticides, *viz*., flubendiamide and profenophos, recorded 93.33% and 91.66% larval mortality, respectively. There was no larval mortality was observed in untreated control.

The search for new techniques and formulations that can increase Bt's efficiency has grown in popularity in recent years. Advances in micro/nanotechnology have opened up new possibilities for the development of more potent formulations that can circumvent some of the difficulties associated with employing them in the field (Vimala Devi *et al*., 2021) [10]. Encapsulation, a safe and practical approach, provides the best protection for biopesticides. It accomplishes this by polymer-coating the Bt spores and crystals, increasing their UV radiation resistance (Jhones *et al*., 2021; Mohammad, 2022) [6, 8]. Our experiments indicated that encapsulated Btk with reduced active components was still effective and that it resulted in increased larval mortality when compared to commercial formulations. Microencapsulated formulations have proven to enhance the firmness of Bt and protect it from photodegradation (Vimala Devi *et al*., 2019; Khorramvatan *et al*., 2014) [13, 4]. In our study, the encapsulated Btk (CC-1+Btk and CC-2+Btk @ 1.5 g/l) showed higher effectiveness against *Spodoptera litura* over non-encapsulated Btk-technical. Further, the present study will be useful to develop encapsulated formulations of microbial pesticides with increased efficacy and field persistence for the eco-friendly management of lepidopteran pests.

Table 1: Effectiveness of *B. thuringiensis* var. *kurstaki* (Btk) microcapsules against tobacco caterpillar (*S. litura*) in castor

Treatments	Dose	Percent larval mortality
T1: *CC-1+Btk	0.75 g/l	71.66 (57.84)^{e}
T2: CC-1+Btk	1.0 g/l	78.20 (62.33)^{d}
T3: CC-1+Btk	1.5 g/l	81.20 (64.52)^{c}
T4: #CC-2+Btk	0.75 g/l	71.00 (57.64)^{f}
T5: CC-2+Btk	1.0 g/l	79.00 (63.73)^{d}
T6: CC-2+Btk	1.5 g/l	81.00 (64.41)^{c}
T7: Btk technical	1.0 g/l	70.00 (56.87)^{g}
T8: Btk WDG	1.0 g/l	86.66 (68.59)^{b}
T9: Btk SC	1.0 g/l	81.66 (64.65)^{c}
T10: Btk (Delfin®)	1.0 g/l	81.66 (64.65)^{c}
T11: Flubendiamide 480SC 39.35% w/w	0.2 ml/l	93.33 (75.04)^{a}
T12: Profenophos 50% EC	2.0 ml/l	91.66 (73.22)^{a}
T13: Untreated Control	-	0.00 (0.00)^{h}
CD (P=0.01)		5.58
SEm (±)		2.00
CV (%)		4.10

*CC-1+Btk: Chitosan + Lignosulphonate + Manganous sulphate + Btk

#CC2+Btk: Chitosan + Lignosulphonate + Carboxy Methyl Cellulose + Manganous sulphate + Btk

The values in parenthesis are angular transformed values; Figures with the same letter did not differ significantly

A) CC-1 + Btk **B) Untreated Control**

Fig 1: Effectiveness of *B. thuringiensis* var. *kurstaki* (Btk) microcapsules against tobacco caterpillar (*S. litura*)

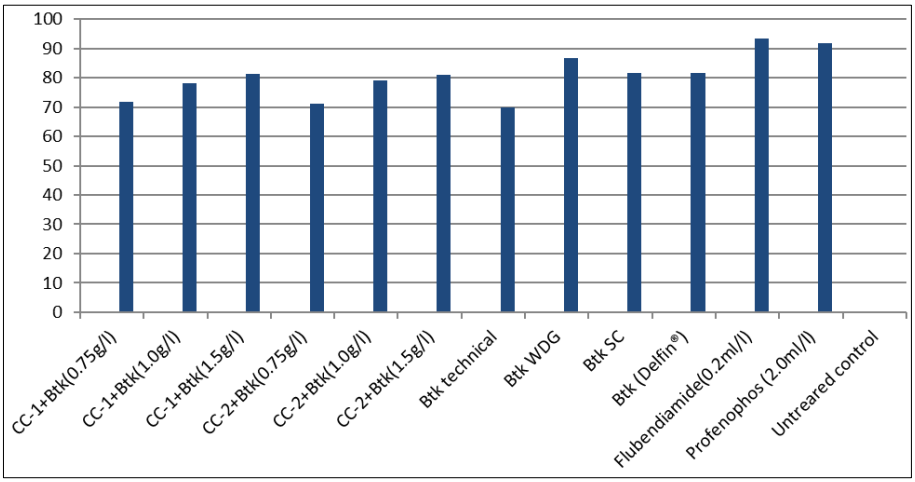

Fig 2: Effectiveness of *B. thuringiensis* var. *kurstaki* (Btk) microcapsules on 5-days old larvae of tobacco caterpillar, *S. litura*

Conclusion

Entomopathogenic bacteria provide great potential for the environmentally benign control of insect pests. *Bacillus thuringiensis* (Bt) has been utilised successfully as an alternative to chemical insecticides in the management of pests. The use of Bt is restricted in field settings due to its susceptibility to environmental factors. Microencapsulation has become a great option to solve this problem. To assess the bio-efficacy of various doses of two promising Btk microcapsules (CC-1+Btk and CC-2+Btk) against the tobacco caterpillar, *S. litura*, a pot culture experiment on castor was carried out. The CC-1+Btk @ 1.50 g/l Btk microcapsule proved to be highly effective and recorded noticeably increased percent larval mortality of *S. litura*. The current results unambiguously show that *Bacillus thuringiensis* encapsulated in biopolymers is more likely to be successful when used in challenging environmental circumstances. The results of the current study will help with the environmentally friendly management of *S. litura* infestations in horticultural and agricultural crops.

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Conflict of Interest

The authors claim that there are no conflicts of interest for this work.

Ethical approval

This article does not contain any studies with human participants or animals performed by any of the authors.

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