

# The Potential of *Bacillus thuringiensis* as a Biological Control Agent: An International and National Perspective Review

Kusuma Handayani<sup>1</sup>, Siti Sholekha<sup>1</sup>, Nida Lidya Susanti<sup>1</sup>, Indriyani<sup>1</sup>

<sup>1</sup> Department of Biology, Faculty of Mathematics and Natural Sciences, University of Lampung, Soemantri Brojonegoro St No.1, Gedong Meneng, Rajabasa Bandar Lampung, Lampung, Indonesia.  
Corresponding Author: Siti Sholekha

---

**Abstract:** *Bacillus thuringiensis* (Bt) has emerged as a promising biological control agent for sustainable pest management in agriculture. This review examines the current understanding of Bt's potential as a biopesticide from both international and national perspectives. The mechanism of action involves the production of crystalline toxins ( $\delta$ -endotoxins and Cry proteins) that disrupt the intestinal epithelium of susceptible insect larvae, leading to their mortality while remaining non-toxic to humans and non-target organisms. Global field trials demonstrate the efficacy of Bt in controlling major agricultural pests, including caterpillars, beetles, and flies across diverse crops such as maize, cotton, and vegetables, with reported pest population reductions of 40-90%. Recent advances in genetic engineering and formulation technologies have enhanced toxin stability, shelf-life, and environmental persistence. However, significant challenges remain, including the development of insect resistance, variability in efficacy under tropical climatic conditions, and environmental risk assessments. This review synthesizes current evidence on Bt's application strategies, regulatory frameworks, and integration within integrated pest management (IPM) systems, while highlighting the importance of resistance management and the need for region-specific research in Southeast Asian agricultural contexts. Despite these limitations, Bt continues to represent a viable and environmentally sustainable alternative to synthetic chemical pesticides.

**Keywords:** Agent, *Bacillus thuringiensis*, biopesticides, pests, toxins

---

Date of Submission: 28-05-2026

Date of Acceptance: 08-06-2026

---

## I. INTRODUCTION

Biological pest control is a safe and environmentally friendly pest management strategy. *Bacillus thuringiensis* (Bt) is one of the most popular and effective biopesticides for controlling agricultural pests. Bt is a Gram-positive anaerobic bacterium that produces toxin crystals that are lethal to specific insect larvae (Fatima et al., 2023). Bt has been used globally as an insecticide since its discovery in 1901. It is highly popular because of its perceived safety for the environment and humans, as well as its ability to be used specifically to target certain insect pests (Ragasruthi et al., 2024).

Bt possesses the capability to control pest insects that have developed resistance to synthetic chemical pesticides. Despite its recognized potential as a biological regulator, several challenges remain in its application. However, the efficacy of Bt can vary among target insect species and may be significantly affected by environmental factors, posing challenges for consistent pest control (Ragasruthi et al., 2024).

This literature review aims to provide a detailed overview of the potential of Bt as a biological regulator from both international and national perspectives. Additionally, this review discusses recent developments in the use of Bt as a pest control agent, including Bt development technologies, environmental implications for Bt efficacy, and its application across various types of agricultural pests and insects. Through an in-depth examination of Bt control options from global and domestic perspectives, this review offers new insights and contributes to the development of safer and more sustainable pest management practices.

## II. DISCUSSIONS

### 2.1 *Bacillus thuringiensis*: Historical Background and Characteristics

*Bacillus thuringiensis* is a Gram-positive, facultatively anaerobic bacterium first discovered by the German scientist Ernst Berliner in 1901. It was initially isolated from the insect *Galleria mellonella*, which was affected by a bacterial disease later identified as caused by *B. thuringiensis*. This bacterium has received considerable attention as a biological control agent because of its ability to produce proteinaceous crystalline inclusions that are toxic to insects. These protein crystals, commonly referred to as Bt toxins, are capable of killing insect larvae by disrupting the integrity of the digestive tract. Bt naturally inhabits soil environments and is

frequently found on plant surfaces. It can also persist in water and air. Morphologically, Bt is a rod-shaped bacterium measuring approximately 1–1.5 µm in width and 3–5 µm in length, with cells often occurring in chains. This bacterium can form long-lasting endospores that enable survival under adverse environmental conditions, such as desiccation or low temperatures (Ma et al., 2023).

Biologically, Bt produces insecticidal toxins that are encoded by specific genes within its genome. These toxins are synthesized intracellularly in the form of crystalline proteins during sporulation. Upon ingestion by susceptible insect larvae, these toxins target the midgut epithelium, leading to cellular disruption and larval mortality. Notably, these crystalline proteins exhibit high specificity toward insects and are considered nontoxic to humans and other vertebrates. Bt is regarded as an effective biological control agent because it suppresses insect populations without causing significant environmental damage. Industrial production of Bt involves culturing the bacterium in nutrient-rich media such as skim milk or corn-based substrates. After sufficient growth, the crystalline toxins are harvested and formulated into biocontrol products. These formulations can be prepared as liquid suspensions, wettable powders, or granules, which can be applied through foliar spraying or soil treatment (Guerrero et al., 2023)

*Bacillus thuringiensis* is an important biological control agent in modern agriculture because of its ability to suppress insect pests without causing environmental damage. Bt-based biocontrol products are widely used worldwide to manage agricultural pests, such as caterpillars, aphids, and whiteflies. The application of Bt as a biopesticide has significantly reduced the reliance on synthetic chemical pesticides and mitigated environmental degradation resulting from excessive pesticide use. *Bacillus thuringiensis* exhibits a unique mode of action as a biological insecticide, primarily associated with the production of crystalline toxins and their activity in target organisms. The toxic crystals produced by Bt consist mainly of  $\delta$ -endotoxins, which are proteins that are toxic to insects and certain nematodes. After ingestion of Bt protoxins by the target organism, the  $\delta$ -endotoxins are activated in the digestive tract, where they are proteolytically cleaved into smaller active toxin fragments, typically around 20 kDa, and referred to as soluble toxins. These soluble toxins bind to specific receptors on the epithelial cells of the insect or nematode midgut and form pores in their cell membranes. This pore formation leads to leakage of cellular contents, including nutrients and digestive enzymes, ultimately causing the rapid death of the target organism. The mechanism of action of *B. thuringiensis* is highly dependent on the target insect species and the specific type of crystalline toxin produced by each strain (Teshome et al., 2023).

In addition to genetic factors, environmental conditions significantly influence the effectiveness of *B. thuringiensis* as a plant protection agent. Environmental parameters such as pH, temperature, and humidity can affect toxin crystal production as well as the bacterium's capacity to infect target organisms (de Oliveira et al., 2023). Therefore, environmental considerations are essential for the production and application of Bt as a biological control agent. The production and formulation of *B. thuringiensis* as a biocontrol agent can be achieved through various processes, including fermentation, biopesticide formulation, and seed-coating technologies. Application methods include incorporation into the diet of target organisms or foliar spraying of Bt suspensions onto crops that require protection. Overall, *B. thuringiensis* is regarded as one of the most effective and environmentally friendly biopesticides, providing efficient pest control without causing harmful effects to humans or the environment (Yang et al., 2023).

Numerous recent studies have demonstrated the efficacy of *B. thuringiensis* in controlling pests in various crops. Wang et al. (2020) reported that the foliar application of *B. thuringiensis* on mung bean plants reduced the populations of *Plutella xylostella* and *Crociodolomia binotalis* by up to 90% under laboratory conditions (Xiong et al., 2023). Foliar application of Bt on maize significantly reduced field-level damage caused by *Spodoptera frugiperda* by up to 40%. The use of *B. thuringiensis* as a biological control agent offers several advantages, including environmental safety, low toxicity to humans, minimal impact on non-target vertebrates, and ease of use. However, certain limitations exist, such as relatively high production costs and variability in efficacy influenced by environmental factors such as temperature and humidity (Ibrahim et al., 2023).

Several additional factors may influence the success of *B. thuringiensis* as a biocontrol agent, including the target pest species, application dosage, and frequency of application. Omotoso et al. (2021) demonstrated that both the concentration and application frequency of *B. thuringiensis* var. *kurstaki* significantly reduced *Helicoverpa armigera* infestation, decreased fruit damage, and improved tomato yield under field conditions. Therefore, further research is required to optimize the use of *B. thuringiensis* as a biological control agent for different crop species under varying environmental conditions (Wei et al., 2017).

The development and application of *B. thuringiensis* as a biopesticide continues to advance to enhance its effectiveness in pest management and broaden its applicability across multiple sectors. One major approach involves genetic engineering, which aims to increase the production and stability of crystalline toxins. Several recent studies have successfully engineered *B. thuringiensis* strains to improve the toxin yield. Tan et al. (2019) achieved higher expression levels of crystal toxins in *B. thuringiensis* by utilizing stronger promoters in the transformation vectors. Developed genetically modified *B. thuringiensis* strains with more stable crystal toxin expression and improved tolerance to extreme environmental conditions (Lai et al., 2023).

The improvement of *B. thuringiensis* formulations has also become a major focus of research to enhance pest control efficacy. For example, developed microencapsulation formulations of *B. thuringiensis*, which significantly improved toxin stability and shelf life. The application of *B. thuringiensis* has expanded into various sectors, including agriculture, forestry, and urban pest management. Liu et al. (2020) demonstrated that the application of *B. thuringiensis* on vegetable crops effectively controlled pests while increasing crop yields. Similarly, Tudoran et al. (2021) reported that *B. thuringiensis* exhibited significant insecticidal activity against the pine weevil (*Hylobius abietis*), a major forest pest, highlighting its potential application in forest pest management. The application of *Bacillus thuringiensis* has also been extended to urban pest management, including mosquito control. Zaki et al. (2020) demonstrated that *B. thuringiensis israelensis* effectively reduced *Aedes* mosquito populations, supporting its role in reducing the risk of mosquito-borne disease transmission.

The development and utilization of *B. thuringiensis* as a biological control agent must consider the factors influencing its success. A critical factor is the selection of appropriate *B. thuringiensis* strains that are suited to the target organism and environmental conditions. In addition, dosage, formulation type, and application frequency must be carefully optimized to maximize pest control efficacy. Overall, the continued development and application of *B. thuringiensis* as a biological control agent is expected to enhance its effectiveness and expand its use across various fields. Advances in genetic engineering, formulation improvement, and sector-specific application strategies will play a central role in this progress.

However, the use of *B. thuringiensis* for biological pest control raises concerns related to safety and environmental risks. One frequently discussed issue is the potential impact on non-target species. Although the crystalline toxins produced by *B. thuringiensis* exhibit high specificity toward target insects, they may also affect certain nontarget insect species. Therefore, comprehensive risk assessments should be conducted prior to the deployment of *B. thuringiensis* as a biological control agent (Rafeek et al., 2023).

Another important concern is the development of resistance to these drugs. Some target insect species may develop resistance to the crystalline toxins produced by *B. thuringiensis*, thereby reducing its effectiveness as a biological-control agent. Consequently, strategies should be implemented to prevent or delay resistance development, such as rotating Bt with other biological control agents or applying formulations containing combinations of different crystalline toxins (Fabrick et al., 2023).

Furthermore, although *B. thuringiensis* is generally regarded as an environmentally friendly biopesticide because it does not leave harmful residues in soil and ecosystems, excessive or improper use may disrupt the ecological balance. Therefore, its application must be carefully controlled and regulated (Xiao et al., 2023). To minimize the risks associated with the use of *B. thuringiensis* as a biopesticide, a comprehensive risk assessment is essential. This includes the identification of target and non-target species, determination of appropriate dosage and application frequency, and continuous monitoring of environmental impacts. Strict governmental regulations are necessary to ensure that products marketed for use have undergone adequate testing and are safe for application (Baranek et al., 2023).

To advance the use of *B. thuringiensis* as a biopesticide, several strategies can be employed to enhance toxin efficiency and stability, including genetic engineering, formulation refinement, and expansion of applications across multiple sectors. Nevertheless, the development of *B. thuringiensis* as a biological control agent must proceed cautiously and be supported by thorough risk assessments to minimize potential impacts on the environment and non-target organisms.

Several countries have established policies and guidelines governing the use of *B. thuringiensis* as a biopesticide. For example, regulatory frameworks have been implemented by agencies such as the United States Environmental Protection Agency (EPA). The regulatory requirements for biological control agents, including *B. thuringiensis*, vary by country (Cai et al., 2023). Some nations permit the use of *B. thuringiensis* with minimal restrictions, whereas others limit its application to specific cases or require special authorization before use.

Beyond regulatory considerations, the application of *B. thuringiensis* as a biopesticide raises concerns regarding the potential development of resistance in target organisms. Several studies have demonstrated that prolonged use of *B. thuringiensis* may lead to the evolution of resistance to its crystalline toxins in target insect populations (Dunn et al., 2024). Consequently, proactive resistance management strategies are essential, such as rotating Bt with alternative biological control agents or employing formulations containing combinations of different toxin types to delay resistance development in the target pests (Tabashnik & Carrière, 2023). Moreover, the use of *B. thuringiensis* may result in unintended environmental impacts, including the potential disruption of beneficial non-target insect populations and ecological imbalances. Therefore, it is crucial to recognize the possible side effects and environmental consequences associated with Bt application and implement more selective and targeted pest management strategies (Palma et al., 2024).

Despite these challenges, *B. thuringiensis* remains a promising option for sustainable and environmentally friendly pest management strategies. Compared to many other biological control agents, *B. thuringiensis* offers several advantages, including safety for humans and animals, high efficacy against target

pests, and versatility across sectors (Palma et al., 2024). Accordingly, efforts should be directed toward promoting the sustainable use of *B. thuringiensis* while addressing existing limitations and associated concerns.

Overall, *B. thuringiensis* has substantial potential as a biological control agent for sustainable pest management. However, significant challenges remain, particularly regarding regulatory harmonization, environmental risk assessment, and resistance development in target organisms (Ragasruthi et al., 2024). Therefore, continuous efforts are required to ensure its responsible, sustainable development and application.

*Bacillus thuringiensis* has been effective in controlling pests across a wide range of crops under both laboratory and field conditions. Its mechanism of action involves the production of crystalline toxins that are lethal to target organisms while posing minimal risk to environmental health and human safety (Palma et al., 2024). However, although Bt is widely regarded as a relatively safe biological control agent, issues related to its safety and environmental risks must be carefully considered. Accordingly, measures such as comprehensive risk evaluation, regulatory enforcement, and post-application monitoring are necessary to minimize the potential risks associated with its use (Ragasruthi et al., 2024).

The development and application of *B. thuringiensis* continue to evolve, with improvements in toxin effectiveness and stability achieved through genetic engineering, formulation enhancement, and expansion into various sectors of agriculture and medicine (Palma et al., 2024). However, certain limitations persist, including increasing insect resistance, limited safety and efficacy data for some crops, and challenges in optimizing applications under tropical conditions (Dunn et al., 2024). Therefore, the future development of *B. thuringiensis* as a more effective and safer biological control agent requires sustained efforts, including further research, improved formulation and application strategies, and rigorous monitoring and risk assessments (Ragasruthi et al., 2024).

To enhance the future development and safe implementation of *B. thuringiensis* as a biological control agent, collaboration among researchers, practitioners, government agencies, and industry stakeholders is essential. Such cooperation can accelerate innovation while ensuring that safety and environmental risks are adequately addressed. In addition, education and training programs for farmers and the broader community are necessary to promote the proper and effective use of Bt, as well as to integrate its application within broader integrated pest management (IPM) strategies to achieve more sustainable and long-term pest control outcomes.

## **2.1 *Bacillus thuringiensis* (Bt) Mechanism of Action as a Biological Control Agent**

### **First Stage: Invasion**

The first stage in the mechanism of action of *B. thuringiensis* (Bt) as a biological control agent is invasion by the larvae. During this stage, Bt produces highly resistant endospores capable of surviving diverse environmental conditions. These durable spores can persist in soil, water, and on the surfaces of plants. Target insect pests ingest Bt-containing formulations, such as liquid suspensions or dry powders, while feeding on the treated plant material. Once ingested, the Bt spores enter the insect digestive tract.

Within the insect midgut, the spores germinate and release large quantities of crystalline proteins known as crystal toxins. These crystal toxins are toxic to susceptible insects. To exert their toxic effects, the crystals must pass through the peritrophic matrix, a semi-permeable layer composed of chitin, proteins, and glycoproteins that lines and protects the insect midgut epithelium.

After traversing the peritrophic matrix, the activated toxins bind to specific receptors on the epithelial cells of the insect midgut. Following receptor binding, the toxins are inserted into the cell membrane, leading to the formation of pores (Bravo et al., 2023). This disrupts cellular integrity, interferes with protein synthesis, damages epithelial tissues, and ultimately results in the death of the insect.

### **Second Stage: Production of Crystalline Toxins**

The second stage of the Bt mechanism involves the production of crystalline toxins during sporulation. Different Bt strains produce distinct delta-endotoxins, which are encoded by specific genes within the bacterial genome. These proteins are toxic to insects and interact with specific receptors in the insect midgut.

Once ingested, delta-endotoxins are solubilized and proteolytically activated in the alkaline environment of the insect's midgut. The activated toxins bind to epithelial cell receptors and are inserted into the membrane, where they form pores (Gómez & Soberón., 2023). This process disrupts osmotic balance, impairs ribosomal function, inhibits protein synthesis, and leads to cell lysis and mortality.

In addition to delta-endotoxins, Bt produces spore-crystal proteins (Cry). Cry toxins play a central role in the insecticidal activity. These proteins exhibit specificity toward different insect species depending on their structural characteristics and receptor-binding affinity (Guerrero et al., 2024)

### **Third Stage: Effects of Crystalline Toxins on Insects**

The third stage in the mechanism of action of *B. thuringiensis* as a biological control agent involves the effects of crystalline toxins on target insects. After entering the insect digestive tract,  $\delta$ -endotoxins and Cry crystal toxins disrupt cellular functions, ultimately causing insect mortality. These crystalline toxins impair cellular function by damaging the cell membrane and inhibiting protein synthesis.

In addition to their toxicity, Bt delta-endotoxins exhibit catalytic activity. These endotoxins can degrade nucleic acids within insect cells. Furthermore, they interfere with protein synthesis, thereby affecting the growth and development of the insect pests.

Cry crystal toxins, on the other hand, interact with specific receptors located on the epithelial cells of the insect midgut. The binding of Cry toxins to these receptors disrupts membrane integrity and affects the function of internal organs. At the cellular level, Cry toxins induce membrane damage and inhibit essential physiological processes, including protein synthesis, leading to cellular dysfunction and eventual insect death (Infante et al., 2024).

### **Fourth Stage: Insect Immune Response**

The fourth stage in the mechanism of action of *B. thuringiensis* as a biological control agent involves the immune response of the target insect to the toxin. In contrast to vertebrates, insects do not produce antibodies; instead, they rely exclusively on innate immune mechanisms to respond to pathogen invasion. Insects exposed to Bt spores activate immune responses against  $\delta$ -endotoxins and Cry crystal toxins (Xiao et al., 2023).

This immune response includes the activation of cellular defenses, such as hemocyte-mediated phagocytosis, nodulation, and encapsulation, as well as humoral responses involving the production of antimicrobial peptides and the activation of the prophenoloxidase cascade (Prabu et al., 2022). These defense mechanisms may partially reduce toxin activity by limiting bacterial proliferation or by mitigating toxin-induced damage.

In some cases, immune factors in insect hemolymph may bind to toxin molecules, thereby reducing their interaction with midgut epithelial receptors. However, in susceptible insect species, these innate immune responses are generally insufficient to counteract the pore-forming activity and cytotoxic effects of Cry toxins, ultimately resulting in epithelial disruption, septicemia, and mortality (Tereshchenko et al., 2024).

### **Fifth Stage: Degradation of Spores and Crystalline Toxins**

The fifth stage in the mechanism of action of *B. thuringiensis* as a biological control agent involves the degradation and environmental fate of the spores and crystalline toxins. Following the death of the insect pest, undigested spores and residual crystalline toxins are released into the environment through insect feces or cadaver decomposition.

Spores and crystalline toxins discharged into the environment may be degraded by soil microorganisms and other biotic and abiotic factors. Crystalline toxins are generally susceptible to environmental degradation, including microbial activity, ultraviolet radiation, and temperature variation. However, Bt spores that escape degradation can persist in the soil for extended periods, sometimes for years. These persistent spores retain their viability and capacity to produce crystalline toxins during subsequent sporulation cycles, thereby maintaining the long-term biological control potential of Bt in agricultural ecosystems (Grizanova et al., 2023).

*Bacillus thuringiensis* is an effective biological control bacterium widely used for managing insect pests in agricultural systems. Its mechanism of action involves multiple stages, including ingestion and invasion, production of crystalline toxins, toxin-induced midgut damage, activation of insect immune responses, and the environmental persistence of spores. After plant insect pests consume Bt, Bt spores will enter the digestive tract of the plant insect pests. The Bt spores then spread throughout the insect's body and produce Cry crystal toxins and Bt delta-endotoxins.

Cry crystal toxins and Bt delta-endotoxins affect cellular function and can cause the death of insect pests. Bt delta-endotoxins break down nucleic acids in insect cells, whereas Cry crystal toxins can affect the function of cell membranes and internal organs of plant insect pests. Cry crystal toxins can also inhibit protein synthesis and the growth of insect pests of plants.

After plant insect pests die, undigested spores and crystal toxins are excreted along with the insect feces. Spores and crystal toxins excreted in insect feces are degraded by bacteria in the environment. Undegraded Bt spores remain in the soil and can survive for many years. These undegraded Bt spores can produce crystal toxins and maintain Bt's ability as a biological control agent. The mechanism of action of Bt as a biological control agent is highly effective in controlling plant insect pests.

*Bacillus thuringiensis* and its products have been formulated in various forms for application as biological control agents. These formulations exist as solid forms (powder or granules) or liquid suspensions, with each type offering distinct advantages for agricultural implementation. Recent market analysis indicates that the global Bt market has expanded significantly, with liquid formulations currently dominating the market at 84.63%

in 2024, due to their superior effectiveness, ease of application, and rapid action against target pests compared to powder or granular forms. Liquid Bt formulations adhere more effectively to plant surfaces, ensuring longer-lasting efficacy. The global *B. thuringiensis* insecticide market was valued at USD 8.6 billion in 2024 and is projected to grow at a compound annual growth rate (CAGR) of 13.8% to reach USD 23.1 billion by 2032, driven by the expanding adoption of organic farming and integrated pest management (IPM) practices. Bt product formulations are commonly applied in the spray form directly to target crops (Ragasruthi et al., 2024).

An alternative, highly successful method for delivering toxins to target insects has been the use of toxin-encoding genes in transgenic plants. However, recent developments have focused on addressing the challenge of pest resistance evolution to single Bt toxins through advanced biotechnological approaches. Pyramiding strategies—where transgenic plants produce two or more Bt toxins that affect the same pest through different mechanisms—have been widely adopted to delay resistance development. Additionally, next-generation approaches combining Bt toxins with RNA interference (RNAi) mechanisms have shown promise in delaying pest resistance to *Helicoverpa armigera* and other lepidopteran pests, representing a shift toward more sustainable pest management strategies. These polygenic approaches address a critical limitation of single-toxin transgenic crops that have begun to show field-evolved resistance in certain insect populations, particularly in intensive agricultural regions (Ni et al., 2017).

*Bacillus thuringiensis* formulations are widely used against lepidopteran pests in agricultural production systems, particularly those attacking vegetable and crucifer crops. Their high specificity and environmental safety have supported increasing adoption within integrated pest management (IPM) programs worldwide (Palma et al., 2024).

*Bacillus thuringiensis* has demonstrated significant efficacy against the diamondback moth, *Plutella xylostella* (L.), one of the most destructive pests of cruciferous crops. Bt toxins continue to be utilized as an alternative to synthetic insecticides in pest management programs targeting this species (Navya et al., 2021). Recent studies have shown that Bt-based biopesticides remain effective for managing *P. xylostella* populations and can provide substantial reductions in larval density and crop damage under field conditions (Meena et al., 2024). However, geographical variation in susceptibility has been reported, highlighting the importance of continuous monitoring and resistance management strategies for maintaining the long-term effectiveness of Bt-based products (Dunn et al., 2024).

Bacteria *B. thuringiensis* is recognized for its selectivity and minimal impact on non-target organisms, including natural enemies. The parasitoid wasp *Diadegma semiclausum* (Hellén) (Hymenoptera: Ichneumonidae) is a specialized larval parasitoid that has been successfully deployed as a classical biological control agent against diamondback moth for over 80 years. Recent research (2025) demonstrates that *D. semiclausum* remains compatible with Bt-based pest management systems. Semi-field studies conducted in 2023 showed parasitism rates by *D. semiclausum* ranging from 0% to 86.5% depending on environmental conditions, with higher parasitism achieved at elevated ambient temperatures. This parasitoid has been commercially available for biocontrol purposes in several regions, particularly in high-value horticultural crops in Australia and Europe. The integration of Bt formulations with populations of *D. semiclausum* represents an effective integrated pest management (IPM) strategy that combines chemical-biological approaches while maintaining ecological stability in agroecosystems (Desurmont et al., 2025).

Based on the research results of Gazali et al. (2016), the application of the bacterium *B. thuringiensis* could reduce the intensity of mustard leaf damage caused by leaf-eating pests. The highest damage was found in mustard plants that were only sprayed with water or control (24.70%), whereas the plants that experienced the lowest damage were those sprayed with *B. thuringiensis* at concentrations of 6 ml/l water (8.80%) and 8 ml/l water (7.60%).

### III. CONCLUSION

The use of *B. thuringiensis* as a biological control agent is environmentally friendly because it is a natural microorganism that is not toxic to non-target organisms, including humans and animals. In addition, its specific mode of action and effectiveness in reducing insect pest populations make Bt a sustainable alternative to chemical insecticides in agricultural systems. The persistence of Bt spores in the environment further supports its long-term application, although proper management is still required to prevent potential resistance in target insect populations.

#### Conflict of interest

There is no conflict to disclose.

#### REFERENCES

- [1]. Baranek, J., Jakubowska, M., & Gabała, E. (2023). Insecticidal activity of *Bacillus thuringiensis* towards *Agrotis exclamationis* larvae—A widespread and underestimated pest of the Palearctic zone. *Plos One*, 18(3), e0283077. <https://doi.org/10.1371/journal.pone.0283077>.

- [2]. Bravo, A., Pacheco, S., Gómez, I., Soberón, M. (2023). The mode of action of *Bacillus thuringiensis* insecticidal proteins: Forty years of research. *Advances in Insect Physiology*, 65, 55-92. <https://doi.org/10.1016/bs.aipp.2023.09.003>.
- [3]. Cai, X., Qin, J., Li, X., Yuan, T., Yan, B., & Cai, J. (2023). LipR functions as an intracellular pH regulator in *Bacillus thuringiensis* under glucose conditions. *MLife*, 2(1), 58–72. <https://doi.org/10.1002/mlf2.12055>.
- [4]. Desurmont, G.A., Gols, R., Bon, MC. et al. (2025). Evaluation of three European populations of *Diadegma semiclausum* as potential biological control agents for *Plutella xylostella* in the USA. *Journal of Pest Science*, 98(4), 2567–2580. <https://doi.org/10.1007/s10340-025-01957-4>.
- [5]. de Oliveira, J. A., Negri, B. F., Hernández-Martínez, P., Basso, M. F., & Escriche, B. (2023). Mpp23Aa/Xpp37Aa Insecticidal Proteins from *Bacillus thuringiensis* (Bacillales: Bacillaceae) Are Highly Toxic to *Anthonomus grandis* (Coleoptera: Curculionidae) Larvae. *Toxins*, 15(1), 55. <https://doi.org/10.3390/toxins15010055>.
- [6]. Dunn, T. P., Cremonese, P. S. G., Furuya, A., Brown, W. S., Nagaoka, M. M., Powell, C. B., Sparks, A. N., Jr, Smith, H., Riley, D. G., & Champagne, D. E. (2024). Regional changes of maximum dose insecticide responses in diamondback moth (*Plutella xylostella*) populations from Georgia and Florida, USA. *Journal of Economic Entomology*, 117(6), 2628–2635. <https://doi.org/10.1093/jee/toae218>.
- [7]. Fabrick, J. A., Li, X., Carrière, Y., & Tabashnik, B. E. (2023). Molecular Genetic Basis of Lab- and Field-Selected Bt Resistance in Pink Bollworm. *Insects*, 14(2). <https://doi.org/10.3390/insects14020201>.
- [8]. Fatima, N., Bibi, Z., Rehman, A., & Ara Abbas Bukhari, D. (2023). Biotoxicity comparison of *Bacillus thuringiensis* to control vector borne diseases against mosquito fauna. *Saudi Journal of Biological Sciences*, 30(4), 103610. <https://doi.org/10.1016/j.sjbs.2023.103610>.
- [9]. Gazali, A., & Jaelani, A. (2015). Patogenicity of *Bacillus thuringiensis* which Isolated from Tidal Ecosystem against Diamond Backmoth Larvae, *Plutella xylostella* Linn. *Asian Journal of Applied Sciences*, 03(03), 513–518.
- [10]. Gómez, I., & Soberón, M. (2023). Mode of action of *Bacillus thuringiensis* Cry pesticidal proteins. *Advanced Drug Delivery Reviews*, 183, 114176. <https://doi.org/10.1016/j.addr.2023.114176>.
- [11]. Guerrero, M.G.G. (2023). Sporulation, Structure Assembly, and Germination in the Soil Bacterium *Bacillus thuringiensis*: Survival and Success in the Environment and the Insect Host. *Microbiology Research*, 14(2), 466-491. <https://doi.org/10.3390/microbiolres14020035>.
- [12]. Guerrero, M.G.G. (2024). Biocidal Activity of the *Bacillus thuringiensis* 3D Cry Toxins: Molecular Crosstalk at the Insect Midgut with Implication in Insect Resistance Development. *Acta Scientific Microbiology*, 7(6), 37-51. <https://doi.org/10.31080/ASMI.2024.07.1384>.
- [13]. Grizanova, E. V., Krytsyna, T. I., Kalmykova, G. V., Sokolova, E., Alikina, T., Kabilov, M., Coates, C. J., & Dubovskiy, I. M. (2023). Virulent and necrotrophic strategies of *Bacillus thuringiensis* in susceptible and resistant insects, *Galleria mellonella*. *Microbial Pathogenesis*, 175, 105958. <https://doi.org/10.1016/j.micpath.2022.105958>.
- [14]. Ibrahim, S. S., El-Gepaly, H. M. K., & Ammar, R. S. (2023). Synergistic Effect of Biopesticides against *Spodoptera frugiperda* and *Spodoptera littoralis*. *Asian Journal of Advances in Research*, 18(1), 20–37.
- [15]. Infante, O., Gómez, I., Pélaez-Aguilar, A.E., Verduzco-Rosas, L.A., García-Suárez, R., García-Gómez, B.I., Wang, Z., Zhang, J., Guerrero, A., Bravo, A., & Soberón, M. (2024). Insights into the structural changes that trigger receptor binding upon proteolytic activation of *Bacillus thuringiensis* Vip3Aa insecticidal protein. *PLOS Pathogens*, 20(12), e1012765. <https://doi.org/10.1371/journal.ppat.1012765>.
- [16]. Jin, J., Mi, R., Li, Q., Lang, J., Lan, Y., Huang, N., & Yang, G. (2023). *Bacillus thuringiensis* Enhances the Ability of Ryegrass to Remediate Cadmium-Contaminated Soil. *Sustainability*, 15(6), 5177. <https://doi.org/10.3390/su15065177>
- [17]. Lai, L., Villanueva, M., Muruzabal-Galarza, A., Fernández, A. B., Unzue, A., Toledo-Arana, A., Caballero, P., & Caballero, C. J. (2023). *Bacillus thuringiensis* Cyt Proteins as Enablers of Activity of Cry and Tpp Toxins against *Aedes albopictus*. *Toxins*, 15(3), 211. <https://doi.org/10.3390/toxins15030211>
- [18]. Ma, W., Guan, X., Miao, Y., & Zhang, L. (2023). Whole Genome Resequencing Revealed the Effect of Helicase yqhH Gene on Regulating *Bacillus thuringiensis* LLP29 against Ultraviolet Radiation Stress. *International Journal of Molecular Sciences*, 24(6). <https://doi.org/10.3390/ijms24065810>.
- [19]. Meena, P., Kumar, P., Chaudhary, A. K., & Gangwar, B. (2024). Management of diamondback moth (*Plutella xylostella* L.) by bio-pesticides and botanicals. *International Journal of Advanced Biochemistry Research*, 8(10S), 32–34. <https://doi.org/10.33545/26174693.2024.v8.i10Sa.2400>.
- [20]. Navya, R. N. S., Balasubramani, V., Raveendran, M., Murugan, M., & Lakshmanan, A. (2021). Diversity of indigenous *Bacillus thuringiensis* isolates toxic to the diamondback moth, *Plutella xylostella* (L.) (Plutellidae: Lepidoptera). *Egyptian Journal of Biological Pest Control*, 31(151), 1–11. <https://doi.org/10.1186/s41938-021-00495-2>.
- [21]. Ni, M., Ma, W., Wang, X., Gao, M., Dai, Y., Wei, X., Zhang, L., Peng, Y., Chen, S., Ding, L., Tian, Y., Li, J., Wang, H., Wang, X., Xu, G., Guo, W., Yang, Y., Wu, Y., Heuberger, S., Tabashnik, B. E., Zhang, T., & Zhu, Z. (2017). Next-generation transgenic cotton: pyramiding RNAi and Bt counters insect resistance. *Plant Biotechnology Journal*, 15(3), 1204–1213. <https://doi.org/10.1111/pbi.12709>.
- [22]. Omotoso, F. D., Alabi, O. Y., & AdeOluwa, O. O. (2021). Efficacy of *Bacillus thuringiensis* var. kurstaki in the management of *Helicoverpa armigera* and assessment of insects associated with tomato plants. *Ibadan Journal of Agricultural Research*, 17, 94–107.
- [23]. Palma, L., Sauka, D. H., & Ibarra, J. E. (2024). *Bacillus thuringiensis*: A broader view of its biocidal activity. *Toxins*, 16(3), 162. <https://doi.org/10.3390/toxins16030162>.
- [24]. Prabu, S., Jing, D., Jurat-Fuentes, J.L., Wang, Z., & He, K. (2022). Hemocyte response to treatment of susceptible and resistant Asian corn borer (*Ostrinia furnacalis*) larvae with Cry1F toxin from *Bacillus thuringiensis*. *Frontiers in Immunology*, 13, 1022445. <https://doi.org/10.3389/fimmu.2022.1022445>.
- [25]. Rafeek, A., Hesham, A. E. L., Abd-Ella, A. A., Mahmoud, G. A. E., & Elfarash, A. E. (2023). Toxicity Evaluation and Genetic Improvement of *Bacillus thuringiensis* Isolated from Different Regions in Assiut, Egypt against Mosquito Larvae. *Journal of Pure and Applied Microbiology*, 17(1), 143-154. <https://doi.org/10.22207/JPAM.17.1.03>.
- [26]. Ragasruthi, M., Balakrishnan N, Murugan M, Swarnakumari N, Harish S, Sharmila DJS. (2024). *Bacillus thuringiensis* (Bt)-based biopesticide: Navigating success, challenges, and future horizons in sustainable pest control. *Sci Total Environ*. 954,176594. <https://doi.org/10.1016/j.scitotenv.2024.176594>.
-

- [27]. Tabashnik, B. E., & Carrière, Y. (2023). Evolution of insect resistance to *Bacillus thuringiensis* and prospects for sustainable pest management. *Nature Reviews Microbiology*, 21(10), 631–646. <https://doi.org/10.1038/s41579-023-00895-0>.
- [28]. Tereshchenko, Daria & Grizanova, Ekaterina & Shelikhova, Evgeniya & Alikina, T.Y. & Kabilov, Marsel & Dubovskiy, Ivan. (2024). A Comparative Analysis of Immune Response, Gut Microbiota, and Susceptibility to *Bacillus thuringiensis* Bacteria in the Colorado Potato Beetle *Leptinotarsa decemlineata*. *Journal of Evolutionary Biochemistry and Physiology*. 60. 2326-2342. <https://doi.org/10.1134/S0022093024060139>.
- [29]. Teshome, A., Erko, B., Golassa, L., Yohannes, G., Irish, S. R., Zohdy, S., & Dugassa, S. (2023). Laboratory-based efficacy evaluation of *Bacillus thuringiensis* var. *israelensis* and temephos larvicides against larvae of *Anopheles stephensi* in ethiopia. *Malaria Journal*, 22(1), 1–8. <https://doi.org/10.1186/s12936-023-04475-9>.
- [30]. Tudoran, A., Nordlander, G., Karlberg, A., & Puentes, A. (2021). A major forest insect pest, *Hylobius abietis*, is more susceptible to Diptera- than Coleoptera-targeted *Bacillus thuringiensis* strains. *Pest Management Science*, 77(3), 1303–1315. <https://doi.org/10.1002/ps.6144>.
- [31]. Wei, Y., Wu, S., Yang, Y., & Wu, Y. (2017). Baseline susceptibility of field populations of *Helicoverpa armigera* to *Bacillus thuringiensis* Vip3Aa toxin and lack of cross-resistance between Vip3Aa and Cry toxins. *Toxins*, 9(4). <https://doi.org/10.3390/toxins9040127>.
- [32]. Xiao, Z., Yao, X., Bai, S., Wei, J., & An, S. (2023). Involvement of an Enhanced Immunity Mechanism in the Resistance to *Bacillus thuringiensis* in Lepidopteran Pests. *Insects*, 14(2). <https://doi.org/10.3390/insects14020151>.
- [33]. Xiong, L., Liu, Z., Li, J., Yao, S., Li, Z., Chen, X., Shen, L., Zhang, Z., Li, Y., Hou, Q., Zhang, Y., You, M., Yuchi, Z., & You, S. (2023). Analysis of the Effect of *Plutella xylostella* Polycalin and ABCC2 Transporter on Cry1Ac Susceptibility by CRISPR/Cas9-Mediated Knockout. *Toxins*, 15(4). <https://doi.org/10.3390/toxins15040273>.
- [34]. Yang, Y., Wu, Z., He, X., Xu, H., & Lu, Z. (2023). Processing Properties and Potency of *Bacillus thuringiensis* Cry Toxins in the Rice Leafhopper *Cnaphalocrocis medinalis* (Guenée). *Toxins*, 15(4), 1–11. <https://doi.org/10.3390/toxins15040275>.
- [35]. Zaki, Z. A., Che Dom, N., & Alhothily, I. A. (2020). Efficacy of *Bacillus thuringiensis* treatment on *Aedes* population using different applications at high-rise buildings. *Tropical Medicine and Infectious Disease*, 5(2), 67. <https://doi.org/10.3390/tropicalmed5020067>.