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## Harnessing Bacteria for Sustainable Pest Management: A Biological Approach

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#### **ABSTRACT**

Microbial pest management in agricultural crops provides an environmentally sustainable alternative to chemical pesticides, aligning with the principles of Integrated Pest Management (IPM). This strategy relies on beneficial microorganisms, particularly entomopathogenic bacteria (EPB), which target and suppress insect pests through specific mechanisms such as toxin production, host colonization, and microbial symbiosis. Among EPBs, *Bacillus thuringiensis* (Bt) is the most widely used, producing Cry and Cyt toxins that disrupt insect gut function, leading to mortality. Other bacterial genera, including *Photorhabdus*, *Xenorhabdus*, *Serratia*, and *Paenibacillus*, also exhibit insecticidal properties and play crucial roles in microbial pest control. These bacteria are classified based on their pathogenic nature (obligate vs. facultative), spore production ability (spore-forming vs.

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non-spore-forming), and cell wall composition (Gram-positive vs. Gram-negative), which influence their effectiveness and application methods. The adoption of bacterial biopesticides within IPM frameworks reduces environmental contamination, mitigates pesticide resistance, and promotes ecological balance. Unlike chemical pesticides, bacterial control agents are highly specific to target pests, minimizing harm to non-target organisms such as pollinators and natural predators. This specificity is a key advantage, ensuring that beneficial insects and the broader ecosystem remain unharmed. Despite their promise, bacterial-based pest management faces challenges, including microbial persistence in the environment, formulation stability, and the feasibility of large-scale application. Continued research in microbial biotechnology, genomics, and improved formulation techniques is essential to overcome these limitations. Furthermore, regulatory support and public acceptance are critical for the successful integration of microbial biopesticides in modern agriculture. Advancements in this field will enhance sustainable agricultural practices, reduce dependence on chemical pesticides, and address the global challenge of food security while preserving environmental health and biodiversity.

Keywords: Biopesticide; Entomopathogenic bacteria; integrated pest management; microbial pest control; and sustainable agriculture.

#### 1. INTRODUCTION

increasing demand for sustainable agricultural practices has led to a shift from chemical-based pest control to environmentally friendly alternatives. Among these, microbial pest management has gained significant attention as an effective strategy within the framework of IPM. Traditional chemical pesticides, though effective, pose several risks, including pesticide resistance, environmental contamination, and harmful effects on beneficial organisms. As a result, there has been a growing emphasis on harnessing microorganisms, beneficial particularly entomopathogenic bacteria (EPB), to control insect pests in a more sustainable manner.

Entomopathogenic bacteria are naturally occurring microorganisms capable of infecting and killing insect pests through various biological mechanisms. These bacteria either act as direct pathogens or produce insecticidal toxins that disrupt the physiological functions of target pests. One of the most widely studied and commercially utilized bacterial species is Bacillus thuringiensis (Bt), known for its production of crystalline (Cry) toxins that specifically target insect larvae. In addition to Bt, other bacteria such as Serratia, Paenibacillus, Photorhabdus, and Xenorhabdus have demonstrated potential for pest control, either independently or in symbiosis with entomopathogenic nematodes (Irshad et al., 2023).

The use of bacterial biopesticides presents multiple advantages, including specificity to target pests, biodegradability, and reduced risk of environmental pollution. Moreover, integrating

bacterial agents into IPM strategies helps maintain ecological balance by preserving beneficial insects and reducing dependency on synthetic chemicals. However, despite these benefits, the large-scale adoption of microbial pest management faces challenges such as formulation stability, environmental persistence, and commercial viability. Continued research into microbial ecology, genetic improvement, and innovative delivery methods will be essential to overcoming these challenges and expanding the role of bacteria in sustainable pest management.

## 2. MICROBIAL CONTROL STRATEGIES FOR INSECT PEST MANAGEMENT

Microbial pest management in agricultural crops embodies an environmentally friendly strategy consistent with the principles of IPM, which hiahliaht sustainable practices to reduce environmental impact. In recent years, there has been a collaborative effort by the academic and industrial sectors to create and implement ecofriendly pest control methods within the IPM frameworks (Ruiu, 2015). This transition towards using microbial-based tactics for pest control demonstrates a growing acknowledgment of the constraints and potential dangers linked to traditional chemical insecticides, propelling the investigation and exploitation of beneficial microorganisms as feasible substitutes.

The adoption of beneficial microorganisms for insect pest regulation has gained momentum as an alternative option to chemical pesticides. The identification and utilization of various entomopathogenic microbial species, such as bacteria, viruses, protozoa, fungi, microsporidia,

and nematodes, have emerged as economically feasible choices for pest management<sup>1</sup>(Irshad et al., 2023). These microbial agents provide precise and specific regulation of populations while minimizing negative effects on non-target organisms and the environment. Notably, pathogenic microorganisms have been recognized as causal factors of diseases in different insect pests, highlighting their potential incorporation into microbial approaches (Rajendran and Gopalan, 1999; Markandeva et al., 2001; Neema et al., 2010). Several of these entomopathogens have been developed and approved as microbial pesticides by commercial enterprises and governmental bodies globally, indicating their increasing acceptance and utilization in agricultural pest management practices (EPA, 2013).

The implementation of microbial-based strategies for insect pest management within IPM systems signifies a fundamental shift towards sustainable farming. By leveraging the inherent capabilities of beneficial microorganisms, such entomopathogens, to suppress pest populations, farmers can lessen their dependence on synthetic chemicals and mitigate the emergence of pesticide resistance. Moreover, the targeted approach of microbial control reduces impacts on non-target organisms, including beneficial insects essential for pollination and ecological equilibrium. Looking ahead, continuous research advancement in microbial technologies, coupled with regulatory backing for their integration into pest management schemes, will be crucial for promoting sustainable agricultural practices and ensuring food security in an evolving environment.

#### 2.1 Bacteria as a Microbial Control Agent

Entomopathogenic bacteria (EPB), derived from the words "entomo" (insect) and "pathogenic" (causing disease), refer to a diverse group of bacteria capable of infecting and killing insects. bacteria have evolved specialized mechanisms to colonize and invade insect hosts, leading to disease and ultimately death (Tanzini et al., 2001). One well-known example of entomopathogenic bacteria is Bacillus thuringiensis (Bt), a Gram-positive bacterium that produces insecticidal proteins known as crystal toxins, these toxins, when ingested by susceptible insect larvae, disrupt gut function and lead to paralysis, ultimately causing mortality (El-Menofoy et al., 2014). The specificity of Bt toxins certain insect species, coupled

their environmentally friendly nature. made B. thurinaiensis а widely used agriculture. Most biopesticide in of entomopathogenic bacteria belong to families as Bacillaceae. Enterobacteriaceae, Pseudomonadaceae, Streptococcaceae, Micrococcaceae (Mampallil, 2017).

The study of entomopathogenic bacteria has revealed a wealth of microbial diversity within this encompassing various genera and species with unique insecticidal properties. Besides Bacillus thuringiensis, other notable entomopathogenic bacteria include Photorhabdus and Xenorhabdus, which are symbiotically associated with entomopathogenic nematodes (Shawer et al., 2018; Mahmood et al., 2020). These bacteria produce a range of toxins and antimicrobial compounds that contribute to mortality. Moreover, advances molecular biology and genomics have facilitated characterization and manipulation entomopathogenic bacteria, allowing for the development of novel biocontrol strategies targeting specific insect pests while minimizing impacts on non-target organisms.

### 2.1.1 Classification of entomopathogenic bacteria

Based on their pathogenicity, bacteria can be classified as either obligate or facultative pathogens. Obligate bacterial pathogens undergo their life cycles solely within the insect host, as demonstrated by species such as *Paenibacillus* spp. In contrast, facultative bacteria possess the capacity to thrive both internally within the host and in the external environment, as observed in species like *Bacillus* and *Serratia*.

Another classification of entomopathogenic bacteria is established on the basis of spore production. These bacteria can be distinguished into spore-producing and non-spore producing categories (e.g., *Pseudomonas* spp.). Within the spore producers, there exist obligate spore-producing bacteria (e.g., *Bacillus papillae*) alongside facultative spore-producing bacteria. The facultative spore producers can be further subdivided into crystalliferous (e.g., *Bacillus thuringiensis*) and non-crystalliferous (e.g., *Bacillus cereus*).

Entomopathogenic bacteria are also classified under Eubacteria, which encompasses three primary divisions based on cell wall characteristics (Vega and Kaya, 2012). These divisions encompass bacteria with Gramnegative type cell walls (Gracilicutes), Grampositive type cell walls (Firmicutes), and Eubacteria lacking a cell wall (Tenericutes). This system of classification, rooted in cell wall structure, offers valuable insights into the diversity and evolutionary connections among entomopathogenic bacteria.

## 2.2 Major Groups of Entomopathogenic Bacteria

Entomopathogenic bacteria belong to a variety of families. with notable representatives residing in the Bacillaceae, Paenibacillaceae, Pseudomonadaceae, and Enterobacteriaceae families. These families harbor a considerable number of entomopathogenic species, such as Bacillus thuringiensis (Bt) in the Bacillaceae family, renowned for its synthesis of insecticidal proteins. The Paenibacillaceae family, on the other hand. includes organisms Paenibacillus. which are obligatory insect pathogens that complete their life cycles inside the host. In the Pseudomonadaceae and Enterobacteriaceae families. species like Pseudomonas and Serratia exhibit entomopathogenic properties, establishing symbiotic associations with entomopathogenic nematodes to infect and eliminate insect hosts. Despite being less conspicuous, members of the Streptococcaceae and Micrococcaceae families display certain dearee entomopathogenicity. comprehensive Α comprehension of the distribution and attributes principal of these categories entomopathogenic bacteria is crucial for the formulation of efficient biocontrol tactics and the progression of sustainable pest management methodologies in agriculture.

#### 2.2.1 Family: Bacillaceae

The Bacillaceae family consists of Gram-positive, heterotrophic, rod-shaped bacteria capable of producing endospores. Notable members of this family include *Bacillus thuringiensis*, *Lysinibacillus sphaericus*, and *Bacillus pumilus*. These bacteria are renowned for their entomopathogenic properties and play key roles in biocontrol strategies targeting insect pests in agriculture and forestry.

#### 2.2.1.1 Bacillus thuringiensis

Bacillus thuringiensis (Bt) is a Gram-positive, rod-shaped bacterium that possesses the

capacity to generate endospores, found widely in various habitats such as soil. environments, deceased insects, silo residues, and diverse plant materials. This bacterium is renowned for its production of insecticidal proteins, specifically Crystal (Cry) and cytolytic (Cyt) toxins, which are formed as parasporal crystalline inclusions during sporulation and stationary growth phases. Moreover, Bt strains have the capability to produce additional insecticidal proteins like vegetative insecticidal proteins (Vip) and secreted insecticidal proteins (Sip) during their vegetative growth stage.

In order to establish a consistent naming and categorization system for toxins, the *Bacillus thuringiensis* Toxin Nomenclature Committee devised a structured four-tier nomenclature based on amino acid sequence similarity for  $\delta$ -endotoxins (Cry and Cyt) and excreted (Vip and Sip) Bt toxins. Several theoretical frameworks, including the "classical," sequential binding, and signaling pathway models, have been postulated to elucidate the mechanism of action of three-domain Cry toxins (Palma *et al.*, 2014).

Scholars such as Yang et al. (2018) have conducted research on the insecticidal properties and cooperative impacts of various Bt toxins against pests like the Oriental armyworm (Mythimna separata). Their results indicated that Vip3 proteins, in conjunction with Cry1A proteins demonstrated Cry2Aa, substantial insecticidal efficacy, with LC50 values ranging from 1.6 to 7.4 µg/g. Another investigation by Plata-Rueda et al. (2020) identified Bt strains such as ABTS-1857 var. aizawai, GC-91 var. aizawai, and HD-1 var. kurstaki as viable substitutes for organophosphate insecticides in managing E. elaeasa in oil palm plantations, with LC50 values spanning from 0.84 to 1.25 mg/mL. These studies underscore the promising potential of Bt toxins in eco-friendly pest control strategies within the agricultural sector.

#### 2.2.1.2 Lysinibacillus sphaericus

Lysinibacillus sphaericus (formerly known as Bacillus sphaericus) is a Gram-positive, mesophilic, rod-shaped bacterium commonly present in soil habitats. Various strains of L. sphaericus produce toxins such as Bin (derived from Binary) and/or Mtx (derived from Mosquitocidal Toxin). The Bin and Cry48/49 toxins are manufactured and contained within parasporal bodies in the sporangium, while Mtx toxins are generated during the vegetative growth phase (Wirth et al., 2014).

Table 1. List of Bt toxins and their role in pest suppression

Bt Toxin	Target Pest(s)	Mode of Action	Reference(s)
Cry1A	Lepidopteran pests ( <i>Spodoptera</i> spp., <i>Helicoverpa armigera</i> )	Damages midgut cells by forming pores, leading to gut disruption and death.	Hoebeke et al., 1997 (Journal of Invertebrate Pathology, 69: 103–114).
Cry2A	Lepidopteran pests ( <i>Plutella xylostella</i> , <i>Trichoplusia ni</i> )	Attacks gut lining by binding to receptors, causing cell rupture and death.	Bravo et al., 2004 (Journal of Invertebrate Pathology, 87: 106–115).
Cry3A	Coleopteran pests ( <i>Diabrotica virgifera</i> , <i>Leptinotarsa</i> <i>decemlineata</i> )	Targets beetle midgut cells, causing membrane disruption and insect mortality.	Huang et al., 2006 (Applied and Environmental Microbiology, 72: 5801–5807).
Cry4B	Dipteran pests (Aedes aegypti, Anopheles gambiae)	Damages the digestive tract of mosquito larvae, leading to their death.	Wirth et al., 2005 (Current Microbiology, 50: 271–278).
Cry1Ab	Lepidopteran pests (Ostrinia nubilalis, Helicoverpa zea)	Disrupts gut epithelium by forming pores, resulting in larval mortality.	Pardo-López et al., 2013 ( <i>Microbiological Research</i> , 169: 609–617).
Cyt2A	Dipteran pests ( <i>Culex</i> quinquefasciatus, <i>Aedes aegypti</i> )	Increases cell membrane permeability, leading to gut cell lysis and death.	Zhang et al., 2008 (Applied and Environmental Microbiology, 74: 1341–1346).
Cry34/35Ab1	Coleopteran pests ( <i>Diabrotica virgifera</i> , Western corn rootworm)	Targets the midgut of beetle larvae, causing cell breakdown and death.	Ellers-Kirk et al., 2003 (Applied and Environmental Microbiology, 69: 3650– 3657).

Recent investigation by Rojas-Pinz et al. (2018) combined illustrated that cultures Lysinibacillus sphaericus show notable larvicidal efficacy against Aedes aegypti larvae, resulting in about 90% mortality. Notably, A. aegypti larvae exhibited greater susceptibility to combined cultures in comparison to individual bacterial strains. The most efficient combined culture attained an LC<sub>50</sub> of 1.21  $\times$  10<sup>5</sup> CFU/mL with Rockefeller larvae and 8.04 x 104 CFU/mL with field-collected larvae. These results underscore the potential of L. sphaericus as a biological agent for controlling mosquito vectors, providing a promising substitute to chemical insecticides in mosquito control initiatives.

#### 2.2.1.3 Bacillus pumilus

The strain 15.1 of *Bacillus pumilus* has been recognized for its harmful effects on the larvae of the Mediterranean fruit fly (*Ceratitis capitata*), a highly damaging pest of fruits and vegetables on a global scale (Molina *et al.*, 2010). Throughout the process of sporulation, this particular strain generates parasporal crystals that bear a resemblance to the cry proteins commonly linked with *Bacillus thuringiensis*. Moreover, the

genome of *B. pumilus* 15.1 harbors genes that encode a variety of entomopathogenic elements, such as chitinases, metalloproteases, and cytolysins (Molina *et al.*, 2010). This revelation highlights the capacity of B. pumilus to serve as a biological control agent against economically important agricultural pests, thereby showcasing its diverse array of insecticidal genes and mechanisms.

#### 2.2.2 Family: Paenibacillaceae

The family Paenibacillaceae encompasses a diverse group of Gram-positive, spore-forming bacteria that are commonly found in soil and various environmental habitats. Members of this family, such as Paenibacillus species, known for their versatile metabolic capabilities and ecological significance. They play important roles in nutrient cycling and plantmicrobe interactions, including plant growth promotion and biocontrol of plant pathogens. Some Paenibacillus species exhibit entomopathogenic properties, producing toxins and enzymes that can target and control insect pests.

#### 2.2.2.1 Paenibacillus spp.

Paenibacillus species are spore-forming obligate pathogens that primarily target larvae of beetles from the Scarabaeidae family (order Coleoptera). In contrast to Bacillus species, growth of Paenibacillus spp. on nutrient media is restricted. These microorganisms are accountable for inducing "milky disease," which is characterized by the milky appearance of infected larvae's hemolymph (Abdelgaffar et al., 2022). Spores of Paenibacillus have the ability to endure in soils for prolonged durations.

Infection by Paenibacillus transpires subsequent to the ingestion of spores by the host larvae. Upon ingestion, the spores germinate, and the vegetative cells infiltrate the midgut epithelium, ultimately reaching the luminal side of the basal membrane. This instigates a process multiplication within the larval host. Toxins generated by the parasporal bodies disrupt the gut epithelial barrier, thus aiding in the invasion of the hemocoel. Subsequent sporulation leads to the prevalence of refractive spores, resulting in the distinctive milky appearance in most Nonetheless, infection instances. by lentimorbus in scarab larvae triggers hemolymph clotting, leading to the brown discoloration of the larvae (Glare et al., 2017).

#### 2.2.2.2 Brevibacillus laterosporus

Brevibacillus laterosporus is frequently encountered in terrestrial and aquatic habitats, displaying aerobic growth alongside the ability to engage in facultative anaerobic metabolism. This particular bacterium has the capacity to infect various insect orders, such as Coleoptera, Lepidoptera, and Diptera, and is characterized by the production of unique parasporal bodies shaped like canoes (CSPB), which are believed to play a crucial role in its insecticidal properties against mosquitoes (Ruiu et al., 2013).

Ormskirk et al. (2019) illustrated that the strain of Brevibacillus laterosporus (Bl 1951), when extracted and utilized as an endophytic bacterium in Brassica plants, demonstrated toxic effects on diamondback moth (DBM) larvae upon consumption. While there was no significant impact on the herbivorous behaviour of DBM larvae, a noticeable decrease in DBM pupation was observed on plants inhabited by Bl 1951.

In a separate investigation conducted by Bedini et al. (2020), it was discovered that spores of

B. laterosporus exhibited substantial insecticidal properties against common house mosquitoes (Culex pipiens) and yellow fever mosquitoes (Aedes aegypti), with LC50 values of 0.10 x 106 spores  $mL^{-1}$  and 0.18 x 10<sup>6</sup> spores  $mL^{-1}$ , respectively. Notably, blow flies, especially Calliphora vomitoria, displayed susceptibility to B. laterosporus spores in comparison to Lucilia caesar, as evidenced by LC50 values of  $78.84 \times 10^6$  spores mL<sup>-1</sup> and  $148.30 \times 10^6$  spores mL<sup>-1</sup>, respectively. The efficacy of B. laterosporus spores was diminished against the house fly (Musca domestica), exhibiting the least susceptibility among fruit flies.

#### 2.2.3 Family: Enterobacteriaceae

The family Enterobacteriaceae is a large family of Gram-negative bacteria that includes many important pathogens and commensals. These bacteria are commonly found in gastrointestinal tract of animals, including humans, and in various environmental niches. Some members of the Enterobacteriaceae are notable as entomopathogens, particularly in the context of IPM. Certain species within this family, such as Photorhabdus and Serratia, form symbiotic associations with entomopathogenic nematodes to infect and kill The insecticidal properties insects. Enterobacteriaceae species have made them contributors to biological control strategies targeting agricultural pests disease vectors.

#### 2.2.3.1 Serratia entomophila

Serratia entomophila is the causative agent of amber disease affecting the New Zealand grass Costelytra zealandica (Coleoptera: grub, Scarabaeidae). This disease only infects the larvae of this specific grub species and was first identified by Trought in 1982. Hurst et al. (2007) conducted studies isolating and characterizing the antifeeding prophage of S. entomophila, evaluating its insecticidal potential against C. zealandica larvae. Their findings revealed that the antifeeding prophage induces cessation of larval feeding and a shift to amber coloration within 48 hours post-inoculation, with increased dose rates correlating with larval mortality. These studies underscore the specific interactions and potential applications of S. entomophila as a biological control agent targeting C. zealandica larvae.

#### 2.2.3.2 Serratia marcescens

Serratia marcescens is a Gram-negative bacterium widely distributed in the environment and classified as aerobic within the family Enterobacteriaceae. This bacterium is known to infect both invertebrates and vertebrates, with a notable reputation as a pathogen of insects, leading to bacteremia and rapid insect mortality. Studies have demonstrated the insecticidal effects of S. marcescens, with findings showing significant mortality rates in various insect species. For example, Aswathy (2015) reported a mortality rate of 93.28% in epilachna beetle (Henosepilachna vigintioctopunctata) isolated from brinjal. Similarly, Pu and Hou (2016) observed a mortality rate of 56.75% in fourth instar larvae of the red palm weevil (Rhynchophorus ferrugineus) caused by S. marcescens. These studies highlight the potential of S. marcescens as an effective biological control agent against insect pests.

#### 2.2.3.3 Yersinia entomophaga

The bacterial species Yersinia entomophaga was initially discovered in infected larvae of the New Zealand grass grub, Costelytra zealandica. Following ingestion, Y. entomophaga efficiently breaks down the larvae's gut epithelial membranes, leading to hemocoel invasion. septicemia, and eventual mortality. The lethal dose (LD50) of purified Tc protein from Y. entomophaga has been determined as 50 ng for C. zealandica larvae and 30 ng for larvae of the diamondback moth (Plutella xylostella) (Hurst et al., 2011). In a study by Hurst et al. (2020), a bait formulation of Y. entomophaga was created to target porina moth (Wiseana spp.) larvae, resulting in a 65% decrease in larval density when applied at a rate of 87 kg per hectare. These results underscore the potential of Y. entomophaga as a viable bioinsecticide for the control of significant agricultural pests.

#### 2.2.4 Family: Pseudomonadaceae

Within the family Pseudomonadaceae, there exist aerobic, Gram-negative bacteria characterized by straight or curved rods with polar flagella. Numerous Pseudomonas species within this family have connections to insects, either as pathogens inducing insect diseases or as commensals inhabiting the insect's gut.

#### 2.2.4.1. Pseudomonas spp.

Pseudomonas entomophila distinguishes itself among Pseudomonas species due to its natural

capacity to infect and kill insects upon consumption. First identified in 2005. P. entomophila has emerged as a key model for investigating insect-microbe interactions. Initially isolated from a lone Drosophila melanogaster female in Guadeloupe. bacterium. this strain designated as L48T, displayed entomopathogenic traits and was subsequently classified as P. entomophila. Ingestion of P. entomophila triggers a systemic immune response in susceptible insects, affecting both larvae and adult stages. Wang et al. (2021) discovered and characterized a strain of P. entomophila (CSHH2) and conducted bioassays that exhibited substantial insecticidal effects against second instar larvae of Monochamus alternatus, with efficacy evident at 10 days posttreatment.

Various strains of Pseudomonas species have been assessed for their entomopathogenic potential against fifth instar larvae of the migratory locust, Locusta migratoria. A week after treatment. nymphs treated Pseudomonas strains B3 (HF911369) and B4 (HF911366) displayed mortality rates of 100% and 98%, respectively (Mohandkaci et al., 2015). Furthermore, Pseudomonas aeruginosa isolated from deceased larvae of the epilachna beetle (Henosepilachna vigintioctopunctata) responsible for mortality in 73.01% of these grubs (Aswathy, 2015). These results emphasize the potential efficacy of diverse Pseudomonas strains as efficient biocontrol agents against insect pests.

#### 2.2.5 Bacteria associated with nematodes

Bacteria associated with nematodes play crucial roles in various ecological interactions, particularly in the context of entomopathogenic nematodes (EPNs). These nematodes form symbiotic associations with specific bacteria, primarily from the genera *Photorhabdus* and *Xenorhabdus*, which are essential for the nematode's ability to infect and kill insect hosts. Upon entering an insect host, the nematode releases its associated bacteria, which then multiply and produce virulence factors that contribute to insect mortality.

#### 2.2.5.1 Photorhabdus luminescens

Photorhabdus luminescens is categorized as a Gram-negative bacterium that engages in a mutualistic relationship and falls under the Gamma proteobacteria group. It is commonly

found in the intestinal tract of entomopathogenic nematodes affiliated with the Heterorhabditidae family. The bacterium, alongside its symbiotic nematodes, exhibits significant pathogenicity towards insects. Following the invasion of an insect host by the nematode, P. luminescens is discharged into the hemocoel, where it releases enzymes and high-molecular-weight complexes (Tc) responsible for decomposing and transforming the insect's body into nourishing substances. This mechanism serves as a source of sustenance for both the nematode and the bacterium. Shawer et al. (2018) conducted a study showcasing the insecticidal efficacy of P. luminescens against Drosophila suzukii at different concentrations, resulting in mortality rates varying from 86.7% to 100% across distinct developmental phases of the insect. These outcomes underscore the potent insecticidal attributes of P. luminescens and its significance in the realm of biological tactics targeting insect pests.

#### 2.2.5.2 Xenorhabdus nematophilus

Xenorhabdus nematophilus is identified as a Gram-negative bacterium primarily inhabiting the digestive system of soil nematodes affiliated with the genus Steinernema, thereby establishing a symbiotic bond. This mutualistic relationship between the bacterium and nematode proves to be highly lethal for numerous insect species, notably inducing mortality in insect larvae. In a research endeavor by Mahmood et al. (2020), the insecticidal potential of chitinase protein discharged by X. nematophilus was assessed concerning its impact on Helicoverpa armigera. Substantial levels of mortality were documented when H. armigera larvae were subjected to chitinase at concentrations of 20 µg g-1 and 40 μg g-1 of their diet. At a concentration of 20 μg g-1 of diet of chitinase protein, a noteworthy 9-fold decrease in larval weight was noticeable in comparison to larvae nourished with 2.5 µg g-1 of diet of chitinase protein. These findings shed light on the potential of X. nematophilus and its associated proteins the context in biological strategies aimed at controlling insect pests.

#### 2.2.6 Plant associated bacteria

Bacteria dwelling within plants in their native settings are denoted by various terms contingent on their precise locations within the plant structure. These designations encompass rhizosphere bacteria, endophytic bacteria, and

phylloplane bacteria, which inhabit the root zone. internal plant tissues, and leaf surfaces. respectively. The induction of resistance in plants, characterized by an augmentation of their defense mechanisms, transpires through two principal pathways: pathogen-induced Systemic and Acquired Resistance (SAR) Induced Systemic Resistance (ISR) facilitated bν beneficial bacteria (Van Loon et al., 1998). The jasmonate (JA) family, inclusive of JA and its analogs, assumes a pivotal role in triggering defensive reactions against insect assaults in plants.

#### 2.2.7 Rhizosphere bacteria

Bacteria thriving in close association with the rhizosphere are commonly known as rhizosphere bacteria. Among these, approximately 2 to 5 percent demonstrate advantageous effects on plant growth, leading to their classification as plant growth-promoting rhizobacteria (PGPR). Research indicates that **PGPRs** like Pseudomonas. Bacillus. and Serratia proficient at establishing themselves in roots and providing a protective function against various crop pests. Remarkably, specific strains of plant growth-promoting Pseudomonas fluorescens, such as Pf1, TDK 1, and PY 15, have shown a significant reduction in leaf folder damage in rice plants. This reduction is linked to the enhanced activity of enzymes like polyphenol oxidase, lipoxygenase, chitinase, and proteinase inhibitors in plants treated with PGPR (Saravankumar et al., 2007). Moreover, cowpea plants treated with PGPR displayed a notable increase in the mortality rate of cowpea aphids compared to untreated controls, with Bacillus subtilis identified as the most potent among the various PGPRs tested.

#### 2.2.8 Endophytic bacteria

Endophytic bacteria are those inhabiting the internal regions of plants, particularly within leaves, branches, and stems, without causing evident harm to their host. The presence of endophytic microorganisms in host plants can enhance plant vigor by offering protection against insect pests and diseases, promoting growth, boosting resistance under stressful conditions. For example, endophytic strains of Bacillus thuringiensis, such as S1905 and S2122, induced 100% mortality in third instar caterpillars of the diamondback moth, Plutella xylostella, while strain S2124 led to 58.33% mortality (Praca, These 2012). findinas underscore the potential of endophytic bacteria in contributing to plant health and strategies for pest management.

#### 2.2.9 Phylloplane bacteria

Phylloplane bacteria flourish on leaf surfaces, often colonizing specific sites like trichomes, stomata, and junctions of epidermal cell walls. While many phylloplane bacteria establish commensal relationships with their host plants, some possess the ability to produce extracellular chitinase enzymes. These enzymes can degrade the peritrophic membrane of chewing insects, rendering these bacteria effective biocontrol agents. For instance, Bacillus thuringiensis, isolated by Gonzalez and Molla (2011) from the phylloplane of tomato plants, exhibited effective control against the tomato leaf miner. Tuta absoluta. This showcases the potential phylloplane bacteria as natural agents for managing insect pests on agricultural crops.

#### 2.2.10 Modified bacteria

The field of genetic engineering presents significant potential for advancing development of engineered entomopathogens with enhanced resilience to environmental challenges. Yang et al. (2018) showcased this potential by introducing the insecticidal gene cry218 from B. thuringiensis into a poplar bacterial endophyte, Burkholderia pyrrocinia JK-SH007, resulting in increased toxicity expression. Similarly, the fusion of B. thuringiensis (Bt) with B. licheniformis and S. marcescens contributed to heightened mortality rates when applied to the larval stage of Tuta absoluta (Atia et al., 2019). studies underscore These how engineering can be utilized to bolster the effectiveness and resilience of entomopathogens against specific insect pests.

## 2.3 Effect of Bacterial Enzymes in the Integrated Pest Management

Bacterial enzymes are emerging as a valuable component of IPM strategies. These enzymes, produced by various bacteria, have the unique ability to target and break down specific molecules essential for pest survival. This targeted approach offers several advantages over traditional chemical pesticides.

The efficacy of entomopathogenic bacteria is heavily reliant on their appropriate deployment

within the field. An illustration of this is seen when utilizing substances that function through ingestion, such as Bt-based products, where precise timing is essential to ensure the stability of bacterial toxins in the surroundings until they are ingested by the targeted insect phase (Vega and Kaya, 2012). Ensuring adequate coverage of surfaces like foliage visited or consumed by insects represents another crucial element. This requirement has stimulated the advancement of specialized techniques for processing formulating bacteria-based bioinsecticides, with the aim of optimizing shelf-life, enhancing dispersion and adhesion, minimizing spray drift, and, most importantly, boosting effectiveness. The sector has introduced a variety of enhancers and supplements for microbial formulations, including dispersants, surfactants, agents, spreaders, drift regulators, pH stabilizers, antifoaming agents, carriers, feeding stimulants, and attractants (Brar et al., 2006). Depending on intended use and environmental circumstances, a variety of solid and liquid formulations are accessible. Solid formulations comprise dusts, granules, briquettes, wettable powders (WPs), whereas liquid suspensions involve suspension may concentrates (SCs), emulsions, and encapsulations. Cutting-edge technologies like microencapsulation's and microgranules are designed to prolong residual impacts.

Bacterial enzymes are often highly specific to their target pest. Unlike broad-spectrum chemical insecticides that can harm beneficial insects and pollinators, bacterial enzymes only affect the targeted pest species. This specificity minimizes collateral damage to the ecosystem and promotes biodiversity (Ruiu, 2015). Additionally, some bacterial enzymes can degrade the protective coatings of insects or disrupt their digestive processes, leading to death (Palma *et al.*, 2014).

Bacterial enzymes are often biodegradable and pose minimal risk of environmental contamination. Unlike chemical pesticides that can persist in the environment for long periods, bacterial enzymes are naturally broken down by microorganisms in the soil. This reduces the risk of long-term environmental damage and ensures safety of food production Furthermore. research ongoing develop new formulations that enhance the shelf life and effectiveness of these enzymes in field applications. Integrating bacterial with other IPM strategies enzymes

monitoring and habitat manipulation holds great promise for sustainable pest control in the future (Rai *et al.*, 2014).

The spectrum of entomopathogenic bacteria-based active ingredients endorsed for commercial purposes, in conjunction with their respective commercial items, is undergoing substantial growth on a global scale. This progression is resulting in the expansion of their market into fresh sectors, thereby broadening their application in pest control, a domain that historically had been confined to specific contexts like forests, public health, and shielded crops (Glare et al., 2012).

## 2.4 Entomopathogenic Bacteria with other Entomopathogens, Botanical and Chemical Pesticides

The combination of Beauveria bassiana with Bacillus thuringiensis subsp. tenebrionis showed increased control efficacy ranging from 6% to 35% against the Colorado potato beetle, Leptinotarsa decemlineata, based on field studies conducted in the United States (Wraight and Ramos, 2005). Similarly, in Kazakhstan, improved mortality rates within a shorter timeframe were achieved by combining B. thuringiensis subsp. morrisoni (Btm) with lower concentrations of Metarhizium anisopliae or B. bassiana against L. decemlineata (Kryukov et al., 2009). Yaroslavtseva et al. (2017) demonstrated that sub-lethal doses of Bt var. tenebrionis significantly suppressed cellular immunity and inhibited detoxification enzymes L. decemlineata, resulting in a synergistic effect when combined with M. robertsii.

In another study by Nouri-Ganbalani et al. (2016), the combination of Bacillus thuringiensis (Bt) and neem at LC50 concentrations (490 µg a.i/mL for Bt and 241 µg a.i/mL for neem) exhibited synergistic effects against Plodia interpunctella, while the LC30 combination showed additive interactions. Niu et al. (2018) reported both additive and synergistic interactions between Serratia marcescens and spirotetramat/thiamethoxam, depending concentration, in laboratory and greenhouse studies against third instar nymphs of the brown plant hopper, Nilaparvata lugens. In India, Singh et al. (2016) found that a chitinase-producing bacterium, Paenibacillus sp. D1, and chitinase extracted from this bacterium exhibited a synergistic interaction with acephate against the cotton bollworm, Helicoverpa armigera, laboratory studies.

#### 3. CONCLUSION

The use of entomopathogenic bacteria in pest management represents a promising and sustainable alternative to conventional chemical pesticides. These microorganisms, particularly Bacillus thuringiensis, Serratia, Paenibacillus, Photorhabdus. Xenorhabdus. and demonstrated significant potential in controlling insect pests through targeted mechanisms, minimizing harm to non-target organisms and reducing environmental pollution. The integration bacterial-based biopesticides into frameworks enhances the sustainability of agricultural practices while addressing concerns related to pesticide resistance and ecological imbalance.

Despite their advantages, the large-scale adoption of bacterial biopesticides faces challenges such as formulation stability, environmental persistence, and commercial feasibility. Advancements in microbial engineering. biotechnology, genetic and innovative application techniques are essential to overcoming these limitations. With continued research and regulatory support, bacterial-based pest control strategies can play a pivotal role in ensuring long-term agricultural sustainability, improving food security, and mitigating the adverse effects of chemical pesticides on the environment. By embracing microbial solutions, modern agriculture can achieve a balance between effective pest management ecological preservation.

#### **DISCLAIMER (ARTIFICIAL INTELLIGENCE)**

Author(s) hereby declare that NO generative Al technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

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#### **COMPETING INTERESTS**

Authors have declared that no competing interests exist.

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