UNIT-V

BIO-PESTICIDES

BACTERIAL ORIGIN

Bacterial Insecticides

Bacteria are mostly associated with causing disease in humans as well as plants. However, there are several bacteria that work as pathogens to several insects. These bacteria play an important role in the development of bacterial insecticides. According to several studies, bacteria that can control several insects are highly specific towards its hosts and it makes it efficient enough to develop as an alternative to chemical insecticides. There are different types of bacterial insecticides available commercially, some of those are described as following

I. *Pseudomonas* as Bacterial Insecticide

Along with plant growth promotion, several Pseudomonas species are reported to have pathogenicity against several insects. *Pseudomonas aeruginosa* is one of the species which is mostly reported as a bacterial insecticide. Apart from that *P. taiwanensis* was reported for its insecticidal activity against agricultural pests such as *Plutellaxylostella, Spodoptera exigua, Spodoptera litura,* etc. These bacteria mostly carry toxin complex genes that have specificity towards insects. *P. fluorescence* is known for its plant growth-promoting activity. However, studies supported the fact that *P. fluorescence* involves bacterial antagonists to control fungal pathogens. Moreover, *P. cepacia* was also reported for its activity towards suppression of plant-pathogen by secretion of siderophores.

II. *Bacillus* as Bacterial Insecticide

a) Bacillus thuringiensis

B. thuringiensis (Bt) is widely used bacilli which can control insects such as moths, beetle, flies, aphids, butterflies, and even some pathogenic fungi like *Pythium ultimum.* The mechanism of Bt to control the insects is depending on its toxins. The endotoxin is a crystallized protein that is soluble in alkaline conditions. The pH in the gut of an insect is mostly alkaline and as it ingested the toxin, the toxin easily dissolves in the midgut region. Then proteases come into action by digesting the toxin which leads to the production of small active fragments. These active fragments then bind to the gut epithelial membrane and create pores which leads to disturbance in osmotic equilibrium. This results in the death of the insects. Several agricultural plants were genetically modified with the gene responsible for toxin production in Bt., such plants are Bt. Brinjal, Bt. Tomato etc.

b) Other Bacillus species

B. thuringiensis is one of the bacillus species which is used widely to control insects. However, there are other *Bacillus* species such as *B. papilliae* and *B. lentimorbus* which play an important role in controlling insects such as Japanese beetle.

***Bacillus thuringiensis* :**

Bacillus thuringiensis (*Bt*) is a rod shaped, gram-positive bacteria that forms a spore and is found in the soil. Classification of this bacteria includes Bacteria (domain); Eubacteria (kingdom); Firmicutes (phylum); Bacilli (class); Bacillales (order); Bacillaceae (family). *Bt* was isolated in 1901 and named in 1911. It was used as a commercial biopesticide for the first time in the United States in 1958. It is placed in IRAC group 11, microbial disruptors of insect midgut membranes. *Bt* is toxic to caterpillars, some fly larvae, and some beetle larvae but not toxic to other organisms. A few strains of *Bt* are available in products used in the United States. *Bt* var. kurstaki is toxic to lepidopteran (butterfly, skipper, and moth) larvae; *Bt* var. aizawai is toxic to wax moth larvae; *Bt* var. israelensis is toxic to mosquito, midge, fungus gnats, and blackfly larvae; *Bt* var. galleriae is toxic to larvae of May or June beetles (white grubs); *Bt* var. tenebrionis (or var. San Diego) is toxic to Colorado potato beetle, elm leaf beetle, and willow leaf beetle larvae. However, *Bt* var. tenebrionis does not kill all leaf beetles.

Bt strains are very specific to the insects they kill. Therefore, identification of the injurious insect is very important. The correct strain must be applied to susceptible insects. Applications of *Bt* to insects that are not susceptible will be ineffective. *Bt* is most effective against young larvae and usually does not kill adults or other stages of an insect. Insects must eat *Bt* for it to be effective, and good coverage is important. Some insects do not eat the outside of the plant part they attack and applications of *Bt* on the surface of the plant are ineffective against them. For example, the pecan nut casebearer bites the outside of nutlets and spits it out. This insect eats the inside of nutlets and does not eat the *Bt*. *Bt* as a biopesticide applied to plants is not systemic or translaminar and does not kill on contact. It is not toxic to beneficials and is listed as an organic insecticide.

Bt is rapidly deactivated by ultraviolet radiation. Applications made in the evening, on cloudy, or on rainy days last longer. However, heavy rains wash *Bt* off the plant. Applications become inactivated in one to a few days and may need to be reapplied in 3 to 7 days. Applications for leaf beetles may be effective for only one day. Applications of *Bt* do not result in continuous management of insects by reproduction of bacterial cells, and *Bt* is applied similar to chemical insecticides. Once a solution of *Bt* is prepared it should be used immediately; especially, if the water used to make the solution has a pH greater than 7 (basic).

The effectiveness of *Bt* may be reduced after two or three years of storage. Dry formulations last longer than liquid formulations. *Bt* products should be stored out of sunlight and in cool, dry conditions.

A crystalline toxin and spore is usually produced by *Bt* cells. The toxin is called a delta endotoxin. *Bt* products usually contain the toxin and spores (environmental resistant stage of the bacterium) but some products do not contain spores. Spores may become bacterial cells inside the insect.

Once the insect eats the *Bt* the delta endotoxin is activated in the insect's gut by enzymes and alkaline (basic) conditions of the gut. A specific pH is required to activate the endotoxin. The endotoxin disrupts the cell walls of the gut. Bacterial cells enter the body of the insect. Infected insects stop feeding in a few hours and die in a few hours to weeks (frequently 2-3 days). Different strains of *Bt* have different endotoxins and kill different insects. The endotoxin is not activated in the gut of humans.

MECHANISM AND MODE OF ACTION OF *Bacillus thuringiensis* :

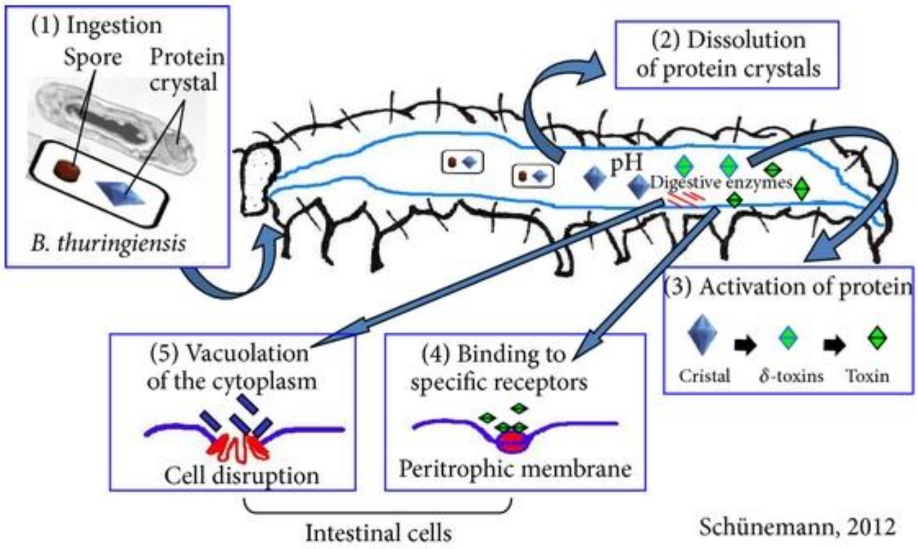
Histological studies, using the insects gut, have been the focus of researches to control agricultural pests. This is due the fact that changes in the gut can not only affect their development, but also cause major physiological events, such as changes in nutrient absorption, degenerative transformation, appetite loss and abandonment of food, gut paralysis, physiological disorders, and total paralysis. These are the most common symptoms observed from the moment the susceptible insects ingest the *Bt* spores and crystals, leading to insect death, when larvae show a blackened color, a characteristic symptom of infections caused by this microorganism [24].

In lepidopterans, the chewing mouthparts promote the ingestion of *Bt* toxins both in a product form and in the form of a toxin-containing GMP. Their digestive tract is divided into three regions: preintestine (front), midgut, and hindgut and is one of the most important areas of contact between the insect and the environment. This has been the subject of research aiming to develop alternative control methods [71].

In caterpillars, the midgut epithelium, pseudostratified columnar, consists of four types of cells, which are involved in the processes of absorption and secretion of enzymes, which are columnar and caliciform cells with ionic homeostasis function, endocrine cells with endocrine function, and regenerative cells involved in the renewal of the epithelium [71]. All of them are coated by a peritrophic membrane, which serves to protect the epithelium from mechanical damage and also as a barrier against harmful chemicals and toxins.

According to Levy et al. [71], the columnar cells are in larger quantities, tall, and present long and numerous microvilli on the apical portion. The basal portion, invaginations form the basal labyrinth. The caliciform cells have a large central cavity delimitated by cytoplasmic projections filled with mitochondria. The regenerative cells present an electron-dense cytoplasm and few organelles. The endocrine cells are characterized by the presence of electron-dense secretory granules concentrated in the cytoplasm of the basal cell.

Several histopathological and ultrastructural studies have investigated the interaction between *Bt* toxins in the midgut of larvae of these insects [72–77]. In Figure 2, the series of events that occur after ingestion of *Bt* in Lepidoptera are shown.



ADVANTAGES OF BIO-PESTICIDES:

1. Accept**How they affect non target species**

When a pesticide is applied to counter a specific pest, this pest is referred to as the target species. Biopesticides products usually fight their intended pests while chemicals end up affecting non target species which include; other insects, birds and mammals.

2. **Pollution**

Due to the toxic ingredients contained in the conventional pesticides, their pollution levels are so high that they cause serious and most of the times fatal effects to the environment. These negative effects can be experienced from the production of the product to the consumption of the plants that these products have been applied on.

3. **Cost**

Most biological pesticide products occur naturally which reduces the cost of production resulting in relatively cheaper prices compared to chemical pesticides whose manufacturing cost is high. This results in the consumer footing the bill at a relatively costlier price.

4. **Pest resistance**

Records have shown that pests tend to become resistance to conventional pesticides thus proving that it is not a long term solution, something that never happens with the use of organic pesticides.

5. **Market**

As the ordinary consumer became aware of the dangers posed by synthetic chemicals, demand for farm products that have undergone organic treatments rose. This makes the use of these chemicals a potential risk as there’s a glaring possibility of incurring huge losses due to the consumer shunning your product.