Efficacy of Bacillus thuringiensis, Berliner
on Prodenia litura, Fabricius, an important
polyphagous pest of certain cultivated crops

A Thesis submitted to the

Mahatma Phule Krishi Vidyapeeth
(Agricultural University)
Rahuri, Dist. Ahmednagar (Maharashtra)
in partial fulfilment of the requirements for the degree of

Master of Science (Agriculture)
In
Agricultural Entomology

By
M. K. Wali
B. Sc. (Agri.) Hons.

Division of Agricultural Entomology,
College of Agriculture, Poona-5.

October, 1971
EFFICACY OF BACILLUS THURIGIENSIS BERLINER ON
PRODENIA LITURA PABRICIUS, AN IMPORTANT
POLYPHAGOUS PEST ON CERTAIN
CULTIVATED CROPS.

by

M.K. Weli
B.Sc.(Agri)Hons.

A THESIS
submitted to the
MAHATMA PHULE KRISHI VIDYAPEETH, RAHURI,
Dist. Ahmednager (Maharashtra)
in partial fulfilment of the requirements
for the degree of
MASTER OF SCIENCE (Agriculture)
in
AGRICULTURAL ENTOMOLOGY
October, 1971

Approved by the Advisory Committee

Chairman and
Research Guide.

Members

(G.M. Talgeri )

(S. M. Munde )

(A. M. Pokharkar )
This is to certify that the thesis entitled "EFFICACY OF BACILLUS THURINGIENSIS BERLINER ON PORDENA LITURA FABRICIUS AN IMPORTANT POLYPHAGOUS PEST ON CERTAIN CULTIVATED CROPS" submitted to the Faculty of Agriculture Mahatma Phule Krishi Vidyapeeth, Rahuri, District Ahmednagar (Maharashtra) in partial fulfillment of the requirements for the degree of MASTER OF SCIENCE (Agriculture) in Agricultural Entomology embodies the results of a piece of bona fide research work carried out by Shri M.K. Mali under my guidance and supervision and that no part of thesis has been submitted for any other degree or publication.

College of Agriculture, POONA- 5.

30th September 1971.

[Signature]

(G.M. Talgeri)
Professor of Entomology, and Specialist to the
M.P.K.V. Agriculture College, Poona.
The author is greatly indebted to Prof. G.M. Talgeri, M.Sc. (Agri), F.S.S.I., Professor of Entomology and Specialist to Mahatma Phule Krishi Vidyapeeth, College of Agriculture, Poona under whose valuable guidance, the present research programme was carried out. In addition the prompt suggestions and constant encouragement given by Prof. Talgeri and the keen interest taken by him in providing all facilities to carry out the assignment are sincerely acknowledged.

He gratefully acknowledges the help rendered by the University authorities of Mahatma Phule Krishi Vidyapeeth, Rahuri, District, Ahmednagar in deputing the author for undertaking the post-graduate studies.

He also expresses his deep sense of gratitude to Shri G.W. Dhande, M.Sc. (Agri), D.I.C. (London) Professor of Plant Pathology and Specialist to the Mahatma Phule Krishi Vidyapeeth, Agriculture College, Poona and Shri A.N. Pokharkar, M.Sc. (Agri) Assistant Professor of Entomology, College of Agriculture, Poona for the timely guidance and advice during the course of his work.

Lastly, the author would like to express his sincere thanks to the staff members and his colleagues in the Entomology Division, College of Agriculture, Poona, for their kind co-operation during his work.

Division of Entomology
College of Agriculture
Poona-5, (India).

30th September, 1971.

M. K. Wali.
## CONTENTS

<table>
<thead>
<tr>
<th>Section</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>I INTRODUCTION</td>
<td>1</td>
</tr>
<tr>
<td>II REVIEW OF LITERATURE</td>
<td>5</td>
</tr>
<tr>
<td>III MATERIAL AND METHODS</td>
<td>17</td>
</tr>
<tr>
<td>IV DATA AND DISCUSSION</td>
<td>21</td>
</tr>
<tr>
<td>V SUMMARY</td>
<td>33</td>
</tr>
<tr>
<td>VI REFERENCES</td>
<td>(i-viii)</td>
</tr>
</tbody>
</table>
Owing to the trend of the proposed policy of restrictions on the use of persistent synthetic insecticides and the growing problem of insect resistance, there has been renewed interest in biological methods of control including the use of microbes causing insect diseases or producing metabolites toxic to insects. The use of pathogenic micro-organisms is an integral part of applied insect control and of special value when and where other methods have been found inadequate. Any degree of insect damage is no longer tolerable and attempts to develop evermore effective insecticides have been found to some degree of self defeat. The entomogenous micro-organisms such as bacteria, viruses, fungi, protozoans, nematodes and rickettsiae can be used for the control of insect crop pest. Among these pathogens, the bacteria is of prime importance due to the possibility of its becoming as an ideal insect pathogen, because of the ease with which it can be produced on large quantities.

In the bacteria, a number of mechanisms aiding persistence have been evolved, for which the most successful one is the ability to form an endospore. The entomogenous spore forming bacteria have been received increased interest because of their great potentiality
for insect control. The most widely utilised group includes the crystal bearing form, *Bacillus thuringiensis* (Berliner) and its close varieties like *sakai* and *saito*. The type species of the group *B. thuringiensis* var. *thuringiensis* was originally isolated from diseased larvae of *Anagasta kuhniella* (Zeller) by Berliner in 1911 and later used against the insects like *Ostrinia* (*Fyrausta*) *nubilalis* (Hubn) (Huss, 1928; Metalnikov and Toumanoff, 1928, Metalnikov and Chorine, 1929; Metalnikov, 1930). The bacterium was further tested in field trials against *Pestinophora* (*Gelechiia*) *gossypiella* (Saunders), *Prodenia litura* (*Fabricius*), *Sporanothia pilieriana* (Schiffermuller) (Metalnikov and Metalnikov, 1932); *Olius eurytheme* (Boisd) (Steinhaus, 1951). More number of susceptible insects have been added to the list of Krieg (1961) and at present this pathogen has been tested successfully against more than 137 insect species from the orders Lepidoptera, Hymenoptera, Coleoptera and Diptera. *B. thuringiensis* and related crystalliferous bacteria have been produced in many countries under various trade names like 'Thuricide' 'Parasporin' 'Biotrol BX, Bakthane L-69', etc. *B. thuringiensis* is also available in different forms like wettable powder, dust, stabilised emulsion and granules. The pathogen has been used in the field on crops like alfalfa, cabbage, cauliflower, banana, lettuce, tomato, potato, cotton, celery and forest trees and was found very effective against the insects pests.
The mode of action of the crystal bearing 
pathogen is mostly as a stomach poison. When the spores 
of the bacteria are ingested by the larvae of the 
insects, the crystal protein acts on epithelium of midgut 
when the epithelium becomes permeable and the contents of 
midgut and haemocoel get mixed. But the mode of 
action, *Bacillus thuringiensis* has been studied in a 
very few insects and more so in case of *Prodenia litura*, 
which is commonly known as 'tobacco caterpillar' in 
India and cotton leaft worm in Egypt. It is widely 
distributed in India and other countries. It is polypha-
egous in habit causing considerable damage to tobacco, 
castor, maize, tomato, jute, lucern, brinjal, cabbage, 
cotton, groundnut, mulbery, berseem, citrus, bhendi and 
cauliflower. The prolific and gregarious habits of 
*Prodenia litura* have also been causing great concern to 
the farmers though its chemical control with organochlorine 
and organophosphorous compounds has shown great promise. 
Fortunately, previous workers like Metalnikov and 
Metalnikov (1932), Yen D. Fung-yean (1962) and Venkataraman 
and Ramesh chander (1967) have reported the susceptibility 
of *Prodenia litura* to the spores of *B. thuringiensis*
and indicates a new line of research. As such, the present investigations are directed more towards the efficacy of *B. thuringiensis* on *Prodenia litura* and also certain aspects of mode of action in *Prodenia litura* larvae particularly with reference to mortality symptomatology, hydrogen-ion concentration and histological studies.
REVIEW OF LITERATURE
REVIEW OF LITERATURE

As the present research programme is directed towards the susceptibility of *Prodenia litura* to the spores of *Bacillus thuringiensis* in the laboratory, the literature pertaining to its mortality, symptomatology, hydrogen-ion concentration and histological studies are only reviewed and presented in this chapter.

(a) Mortality:

McConnell and Jutkomp (1954) observed cent percent mortality with suspension containing 1.5 million spores/ml of *B. thuringiensis* spray to the first instar larvae of *O. nubilalis* and 50 percent mortality at one million and 25 percent to 0.5 million spores per ml of spray; while with the increasing toxicity to 4 million spores decreased susceptibility was observed in case of the 4th instar larvae.

Sprays with 0.25 gm *B. thuringiensis* per gallon gave complete mortality of *Pieris rapae* and *Trichoplusia ni* within 2 to 4 days (Tanada 1956), while Lemiogne et al. (1956) reported effective control with a spray of 1.25 pounds w.p. or 27 lbs of 5 percent dust per acre against *P. brassicae* on cabbage crop.
Rabb et al. (1957) observed that 2 grams of *B. thuringiensis* containing 40 and 50 billion spores per gallon gave good control of second instar larvae at *P. sexta* and *Protonemus quinquemaculatus* respectively and mortality to the extent of 70 - 80 percent was obtained in *Heliothis virescens* within 4 - 5 days by Chamberline and Dutky (1958) by using DD 135-2 and DD 133-6 cultures of *B. thuringiensis*; whereas 94 billion spores /100 plants gave 100 percent mortality in *P. sexta*.

Laboratory experiments conducted by Hall and Dunn (1958) revealed 0.25 gm to 2 gm powder containing 100 billion spore per gram gave 28 to 100 percent mortality in several leaf chewing insects.

Stern et al. (1959) obtained satisfactory control of *Spilaena strymon* on alfalfa by ground application of spore material at 1.00 - 18 ounces per acre containing 40 billion spores per gm. Complete mortality was however, obtained in the *Plutella maculipennis* (Menn 1960) and *Parnassius dispar* (Cantwell et al. 1961) when *B. thuringiensis* was used at the rate of $47 \times 10^5$ / 100 ml of and 700 million spores /ml respectively.

While using commercial preparation of *B. thuringiensis* in laboratory and field, Ayyer (1961) observed high susceptibility to all the stages of *Plutia neponia*;
Hercyronia indica, Glyphodes sp. etc., but Spodoptera mauritia and Heliothis armigera showed resistance at later stages.

Venkatraman et al. (1962) observed 100 percent mortality of Spilachna ceollata and Athalia proxima with 70 x $10^7$ and 30 x $10^7$ spores per gram within 24 hours and mortality to the extent of 80 percent in G. cephalonica after 30 days with 75 x $10^7$ spores/gas and Schistocera gregaria required high concentration to get 90 percent mortality within 3-6 days. Yen (1962) tested Biston marginata, Prodenia litura, T. ni in field and pyrausta venosatana, Chilo infuscatellus in laboratory for the susceptibility to B. thuringiensis.

Harfs and Krieg (1963) used Be-threne L. 69 (Gala, Minoc), and Hitrol B78, available three commercial preparations in the form of sprays at 0.1 to 0.4 percent and 22.25 lb dust per acre against Pieris brassicae in which the sprays proved to be inferior to the dust on account of sedimentation.

Atwal and Puri (1964) carried out several experiments in the laboratory and field against Helicoverpa, steniallus on sugarcane with Be-threne and thuricide in which laboratory experiment with 0.2 percent spray on split cane gave good control and addition of 1.5 percent
molasses to both preparations on whole cane doubled their effectiveness; whereas 0.4 to 0.8 percent thuricide along with molasses gave varying degree of control.

Chatterji (1965) reported that the 6 kg wettable powder of thuricide containing 30 billions spores/ga was the most effective treatment than 4 and 5 kg per hectare against the larvae of *Anomoia sabulifera* on jute; while the mortality to the extent of 80 percent in 3rd instar larvae of *Neodiprion lecontei* was observed in laboratory after exposing them to previously dipped leaves in 5 mg/ ml of spore suspension of *Be*-4 of t aren'to (Sheikh and Morrison, 1965). However, 88.39 percent mortality of 3rd, 4th, and 5th instar larvae of *Achroa jejata* within 76 hours were obtained with thuricide dust containing $30 \times 10^8$ spores/ga by Kulshreshta et al. (1965).

Jaimarao et al. (1966) found out that aqueous suspension of *B. thuringiensis* containing $1 \times 10^8$ spores/ga gave complete mortality of 4th and 5th instar larvae of *Prayâ altri* in the laboratory; while in the field suspension containing $2.5 \times 10^8$, $2.5 \times 10^9$, $3.3 \times 10^9$ spores/ga gave 77.8, 91.2 and 100 percent mortality respectively.

Venkatraman et al. (1967) observed moderate to high pathogenicity for several lepidopterous pests in which cent percent mortality to all stages of *Pamilla*
demolish at four different concentration was noticed; whereas in 1st instar larvae of Prodenia litura, the mortality was to the extent of 98 percent within 7 days. High pathogenic mortality was noticed at high concentration in 2nd instar larvae of Plutia orichalcea F; while it was moderate in early larval stages of L. orbisonia and G. partellus.

Buftan et al. (1969) reported the higher kill in thuricide 90 Ts than sitrol 25 w with Hyphentria sine. Thuricide at 0.1 and 1 lb (a.i.)/acre containing 30 billion spore gave 14 and 100 percent kill as against 16 and 67 percent in Sitrol 25 w at 0.3 and 3 lb (a.i.)/acre containing 25 billion spores/gm respectively.

Bullock and Duleage observed only 40 and 10 percent infestation in cotton bolls with P. gossypiella when it was treated with 1 and 2 quart of microbial insecticides as against 80 percent in the untreated fields.

(b) Symptomatology

In the larvae of *Bt. thuringiensis*, Steinhaus (1951) observed the symptoms of cessation of feeding, sluggishness, diarrhoea, regurgitation resulting in ultimate death.
Hannay (1955, 56) identified the crystal shaped parasporal bodies of \( B. \) thuringiensis culture which were responsible for the production of toxic substances causing septicemia in insects. Similar toxic substances were extracted from the varieties of \( B. \) alesti, \( B. \) thuringiensis and \( B. \) sotto of \( B. \) thuringiensis where these crystals were mainly responsible for paralytic effect on silk worm (Angus 1956) and toxemia in lepidopterous larvae.

Heimpel (1956) also described the cessation of feeding, diarrhea, and vomiting in \( B. \) aethiopoides and becoming flaccid and changing the colour after death which Steinhaus (1949) referred to as melonosis; while the larvae of \( B. \) mori showed sluggishness, distension, swelling at the abdominal region, oral and anal discharges along with loss of curling ability of larvae after the ingestion of \( B. \) sotto (Angus and Heimpel 1956). Moreover the prolegs were unable to clasp the leaf edges. Symptoms of immobility and darkening were noticed by Stephens (1957) in the \( B. \) capsae pomonella larvae.

Heimpel and Angus (1959) classified the insects into three categories as:

- **Type-I**: an increase of pH to the extent of 1 to 1.5 in blood and although suffered gut paralysis as if it was marked by general paralysis.
Type-II: occurred gut paralysis within a few minutes after ingestion followed by cessation of food. No increase in blood pH but gut pH decreased so that the rapid growth of bacteria took place.

Type-III: neither gut nor general paralysis occurred but death is due to septicemia. Burgerjor and de Barjac (1960) reported that infected tent caterpillar died after 10 days of feeding, showing the symptoms of toxemia and not the paralysis; however, Yamvrias (1961) observed that varieties of *B. thuringensis* caused diarrhoea coupled with chronic toxemia. Ramkrishnan and Pant (1967) noticed death of *larvae fabia* larvae within 30 hours after ingestion showing the symptoms of cessation of feeding, hyperactivity, sluggishness, oral and anal discharges and paralysis; while in *P. damoleus*, it was observed by Venkatraman and Ramesh Chander (1967) to cease their feeding after 90 minutes and general paralysis 150 minutes after ingestion followed by regurgitation, diarrhoea and gut paralysis apparently masked by general paralysis; while the *Prodenia litura* did not show any sign of paralysis and change in blood pH, but the larvae stopped feeding in 24 hours followed by septicemia in 72 hours and death which indicated the type III suggested by Heimpel and Angus.
(c) Hydrogen-ion concentration:

As gut pH in insect is one of the limiting factor for the infection of *B. thuringiensis*, the studies on gut pH in various insects in relation to the development of spores of strains of *Bacillus* were carried out by several workers.

Stephens (1952) for the first time observed that the *B. cereus* strains had grown in the range of 5. to 8 pH in the gut of *Carpocapsa rosanella* whereas the larvae of codling moth was killed by the spores which had developed in midgut of 7.5 to 8 pH. (Phospolipase C ) Foumanoff (1953) observed the production of lecithinase of *B. cereus* which kept optimum pH activity of 6.6 to 7.4. Hannay and Fitz (1955) noticed that the toxic crystals dissolved in alkaline solution and under favourable condition of gut pH, varieties of *B. thuringiensis* secreted lytic exoenzymes and caused the damage to the gut which was noticed by Young and Fitz (1959).

Angus (1956) classified 3 types of Lepidopterous and Hymenopterous insect on the basis of reaction to the pH. Type I species showing the paralysis in the pH from 9.6 to 10.4, Type II species not showing paralysis in the pH from 9.4 to 9.9 and Type III species not susceptible in pH of 8.1 to 9.2.
Amongst the 14 species of Lepidoptorous and Hymenopterous insect, the larvae of hymenoptera were observed to have lower pH than that of Lepidopterious larvae (Heimpel 1956).

Angus and Heimpel (1959) reported that the species infected with crystalliferous bacteria showed very slight change in blood pH.

Haimpel and Angus (1959) observed the drop of pH in the larval gut of A. mori, Chlostridion glumea- maculatus and A. pernyi and was correlated with the progressive development of paralysis.

Martouret et al. (1965) carried out several experiments regarding gut pH in the larvae of Pieris brassicae, which indicated that the intoxicated larval midgut had a drop of pH of 0.15 and 1.57 units after 24 and 45 hours after cessation of feeding respectively and caused the gut epithelium damaged. Sutter et al. (1966) however observed the reduction of blood pH along with the gut pH in the larvae of O. nubilalis and gut pH in Spodoptera frugiperda only.

Vidzova (1966) studied changes of hydrogen ion concentration in the haemolymph of larvae of Heliothis oritana by using two strains as Czechoslovak and strain no 88 of A. thuringiensis at 5 to 6 million spores per ml
which showed the changes in the haemolymph and qualitative and quantitative damage to the haemocytes after the entry of bacteria in to the blood; while Ramkrishnan and Pant (1967) had not observed any change in the blood pH of *caricae fahia* but only noticed reduction of gut pH.

Venkatraman and Ramesh Chander (1967) reported the rise of blood pH in the infected larvae of *P. demoleus* whereas *prodenia litura* did not show any apparent change in blood pH.

(d) **Histological studies**:

Histological investigations carried out by several workers had shown that paralysis was followed by breakdown of gut epithelium showing the visible appearance of erosion, disintegration and spongy degeneration in the gut of several species of insects with *bacillus spores*.

Berliner (1915) found out that after germination in the larvae of *A. Kuhniaea*, the spores of *B. thuringiensis* var. *thuringiensis* developed eroded area at the posterior midgut through which bacteria entered in the haemocoel and caused fetal septicaemia. The same was supported by Mattes (1927) andimpel (1954) by observing the movement of bacteria from gut cells to haemocoel.

In a critical studies on histopathology of
*Pieris rapae*, Tanada (1953) observed the dis-organisation of midgut epithelium by separation of cells through which *B. thuringiensis* spores penetrated the muscular layer and entered haemocoel; while rapid degeneration of midgut epithelium due to *B. thuringiensis* var *alesti* was reported in silk worm larvae by Toumanoff and Vago (1953).

Heimpel and Angus (1959) observed extreme damage in *B. illusor* due to *B. entomocidus var entomocidus* at the anterior two third portion of midgut cells by separation from one another and from the basement membrane; relaxation of musculature and fenestration of generative muscles in haemocoel, where as sloughing of midgut epithelium into the lumen of gut in the 6th instar larvae of *G. mellonella* was noticed by Hoopingarner and Materu (1964) when fed with *B. thuringiensis*. In their experiments, they observed no change in the anterior and of midgut initially in the cell type I -larvae of *P. brassicae* at LD 100 dose; while bulk of epithelial cells having fragments of cells released into lumen of gut and appearance of irregular stratification.

Jutter and Raun (1966) observed the loosening of epithelium at first in anterior portion and progressing towards posterior in the midgut of European corn borer showing the intercellular changes as disappearance of
lipid like bodies, break down of micro villii of globlet cells causing vacuolation, and destruction of endoplasmic reticulum. Ramkrishnan and Pant (1967) showed the changes like shrinkage, loosening degeneration of epithelial cells, and relaxation of muscle layer in the larval midgut of Drosophila. Videnova (1966) reported that normal intestinal bacteria showed no defence reaction in Heliothis orvitrina but when spores gained entry in blood, there was rapid break down of haemocytes due to secretion of toxins causing septicaemia and death.
Test Insect: *Prodenia litura* F. Family - Noctuidae order - Lepidoptera.

Test Material:
Thuricide containing 30 billion viable spores per gram supplied by International Minerals and Chemicals Corporation, Bioferm Division, Wasco, California.

Rearing of the Insects:
Rearing technique of test species, as suggested by Venkatraman and Ramesh chander (1967) was used for the experimental work. The initial culture of *Prodenia litura* was obtained from the naturally infested castor plants from the Division of Entomology. The larvae with the infested leaves were kept in a petridish (6" diameter) and were fed with clean and tender leaves of castor.

When the moths emerged, they were transferred to a breeding chamber consisting of glass jar covered inside with the rough paper, so as to create a dark space inside the jar. A cotton swab dipped in 20 percent glucose solution, served as food for the moths. A folded crumpled papers were also kept inside the jar for egg laying. The glass jar top was covered with the muslin cloth.

When the eggs were laid the duster on the crumpled folded paper, as well as on the paper inside the glass jar,
the moths were transferred to new breeding chamber. The eggs were collected after every 24 hours interval and kept separately. When the colour of the eggs changed to bluish black in 4 to 5 days they were transferred to petridish containing tender castor leaves for feeding the newly hatched larvae. Approximately the same age group larvae were obtained from this culture for conducting the experiments.

*Hemicitus diet:

"Thuricide HP containing 30 billion spores per gram of B. thuringiensis (Berliner) was used. A standard solution was prepared for each experiment. It was stirred well and leaves were dipped in so as to get even distribution of the spores. Then the leaves were air dried and fed to insects. The experiments were conducted at a constant temperature of 27 ± 2°C. The technique was more or less similar to those used by Hall and Dunn (1958) and Venkatraman et al. (1967).

In order to study symptomatology a standard solution containing 10 percent "Thuricide" by weight was prepared. The leaves were dipped and air dried taking care that the solution was constantly stirred. Treated leaf was kept in each petridish and 8 days old larva of Prodenia litura was allowed to feed. The larvae were
starved for 12 hours before the commencement of the experiment. Larvae were transferred from treated leaves to untreated after 12 hours. These experiments were replicated three times and observations were noted at an interval of 6 hours.

As regards pH studies, the alimentary canal of the larva was taken out through the longitudinal incisions and washed in distilled water. It was kept on clean glass slide and dried with the help of filter paper. The alimentary canal was then cut into different regions like gizzard, anterior midgut, median midgut, posterior midgut and hindgut. With the help of clean forceps each region was squeezed out on to a strip of pH paper (pH Indicator Paper). The pH values of different regions of alimentary canal of healthy larvae, infected larvae and starved larvae were recorded. The age of larvae was 8 days. The blood was collected by snipping off the abdominal proleg and the pH of the various treatments were made by using pH papers.

With regard to histological studies, however, about 8 days old larvae of _Prodenia litura_ previously starved for 12 hours were fed on leaves treated with bacillus spores. The larvae were sampled at the interval of 9, 13, 27 and 36 hours. The alimentary canal of the treated larvae was dissected and washed in distilled water.
It was fixed in Bouin's solution and was embedded employing ethyl alcohol series, sections were cut at 8 \mu \text{m} thickness and stained with eosin and hematoxylin.

While studying the mortality of *Podena litura*, young larvae of 4 day and 3 day old were used. Different concentrations of *bacillus* spores i.e. 10 percent to 10 percent were prepared. Castor leaves were dipped, dried and placed in petridish. A single larva was exposed to treated leaf for 12 hours and then provided with fresh leaves, till death. Two replications of 25 larvae of two age groups were exposed on treated leaves. In the control, the leaves were treated with distilled water. The larval mortality in different treatments was recorded at 12 and 24 hours interval for both the age groups. All larvae were starved for 12 hours prior to exposure.
DATA AND DISCUSSION

I. Mortality studies:

The observations on the mortality in different larval stages of *Prodenia litura* with different concentrations of thuricide HP are presented in Table I.

### Table I

Percentage mortality in 4 day old larvae of *Prodenia litura* fed with treated leaves at different concentrations of thuricide-HP.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Concentration</th>
<th>No. of larvae</th>
<th>Progressive percentage mortality after 12</th>
<th>24</th>
<th>36</th>
<th>48</th>
<th>60</th>
<th>72</th>
</tr>
</thead>
<tbody>
<tr>
<td>Thuricide-HP (Containing 3.26 active ingredient having 30 billion viable spores/gm)</td>
<td>6%</td>
<td>25</td>
<td>2</td>
<td>20</td>
<td>68</td>
<td>84</td>
<td>90</td>
<td>90</td>
</tr>
<tr>
<td>6%</td>
<td>25</td>
<td>2</td>
<td>12</td>
<td>60</td>
<td>64</td>
<td>68</td>
<td>68</td>
<td></td>
</tr>
<tr>
<td>4%</td>
<td>25</td>
<td>2</td>
<td>12</td>
<td>54</td>
<td>68</td>
<td>68</td>
<td>68</td>
<td></td>
</tr>
<tr>
<td>3%</td>
<td>25</td>
<td>2</td>
<td>16</td>
<td>50</td>
<td>60</td>
<td>60</td>
<td>60</td>
<td></td>
</tr>
<tr>
<td>2%</td>
<td>25</td>
<td>2</td>
<td>6</td>
<td>32</td>
<td>42</td>
<td>46</td>
<td>46</td>
<td></td>
</tr>
<tr>
<td>1%</td>
<td>25</td>
<td>2</td>
<td>4</td>
<td>20</td>
<td>34</td>
<td>46</td>
<td>46</td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>25</td>
<td>2</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>
Graph 1: Percentage mortality in 4 day old larvae of B. littoralis fed with leaves treated with "Thuricide-HP."
It was observed from the table I and graph 1 that none of the doses gave complete mortality even after 72 hours through the treatments with 10 percent of thuricide-HP gave 90 percent mortality within 36 hours after feeding; whereas that with 9 and 8 percent gave 98 percent and 7 and 6 percent gave 90 percent mortality with 60 hours after feeding as against 24 percent with 1 percent thuricide-HP. The larvae survived after 60 hours of feeding under went pupation in all the treatments. These observations were more or less similar to those noticed by Venkatraman and Namasch Chander (1967).

### Table II

Percentage mortality in 8 day old larvae of *Prodenia litura* fed with the treated leaves at different concentrations of thuricide-HP.

<table>
<thead>
<tr>
<th>Treatment (concentration)</th>
<th>No. of larvae</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
</tr>
</thead>
<tbody>
<tr>
<td>thuricide -HP, 10 ppm</td>
<td>25</td>
<td>54</td>
<td>80</td>
<td>86</td>
<td>86</td>
<td>86</td>
<td></td>
</tr>
<tr>
<td>(Contain 8 ppm active ingredient)</td>
<td>25</td>
<td>48</td>
<td>76</td>
<td>80</td>
<td>80</td>
<td>80</td>
<td></td>
</tr>
<tr>
<td>having 3.2 billion viable spores/gm</td>
<td>25</td>
<td>44</td>
<td>58</td>
<td>72</td>
<td>74</td>
<td>74</td>
<td></td>
</tr>
<tr>
<td>control</td>
<td>25</td>
<td>18</td>
<td>36</td>
<td>42</td>
<td>44</td>
<td>44</td>
<td>44</td>
</tr>
<tr>
<td>2 ppm</td>
<td>25</td>
<td>10</td>
<td>24</td>
<td>28</td>
<td>28</td>
<td>28</td>
<td>28</td>
</tr>
<tr>
<td>1 ppm</td>
<td>25</td>
<td>4</td>
<td>8</td>
<td>16</td>
<td>16</td>
<td>16</td>
<td>16</td>
</tr>
</tbody>
</table>
| Control                  | 25            | 0 | 0 | 0 | 0 | 0 | 0
Graph II: Percentage Mortality in 8-day old larvae of *P. litura* fed with leaves treated with "Thuricide-HP."
Observations in table II and graph II on the mortality of 3 day old larvae also indicated that none of the concentrations of thuricide-HP gave complete mortality. The treatments with 10 percent thuricide-HP gave mortality to the extent of 86 percent within 4 days after feeding. The treatments with 9 and 8 percent thuricide-HP however, gave 80 and 76 percent mortality within 4 and 5 days respectively; while it was 74, 66 and 52 percent in the treatments with 7, 6, and 5 percent at 5 days though the rest of the treatments gave less than 50 percent mortality but extremely meagre mortality was noticed in the treatments with 2 and 3 percent thuricide-HP, while the treatments with 1 percent thuricide had not shown any mortality and the larvae survived underwent pupation in all the treatments. These observations were more or less similar to those noticed by Venkatraman and Ramesh chander (1967).

II. Symptomatology:

The symptoms observed during the present investigations in the infected larvae of *Prodenia litura* with *Bacillus thuringiensis* are furnished in table III.

As soon as the treated leaves were fed to the 12 hour starved larvae, they began feeding within 15 minutes time; however their feeding was not of a continuous nature. Cessation of feeding was noticed which was followed by regurgitation.
### Table III
Symptomatological observations on 12 hour starved 8 day old larvae of *Prodenia litura* (30 larvae) fed with Thuricide-HP

<table>
<thead>
<tr>
<th>Symptoms</th>
<th>Observations after hours of feeding</th>
<th>First hour</th>
<th>3 hrs.</th>
<th>6 hrs.</th>
<th>12 hrs.</th>
<th>18 hrs.</th>
<th>24 hrs.</th>
<th>36 hrs.</th>
<th>48 hrs.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Feeding</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Regurgitation</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Diarrhoea</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Sluggishness</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Paralysis</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Death</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Change in body</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
</tr>
</tbody>
</table>

* Colour and size changed after 8 hours of death.*
After 6 hours of feeding on the treated leaves of castor, the larvae moved slowly and gave no response to the sense of cold needle touch. They continued to feed but feeding was extremely slow by chewing the leaves and taking minute quantity of food up to 24 hours.

After cessation of feeding within 24 hours of feeding infected larvae showed high susceptibility within 36 to 48 hours indicating the high percentage mortality.

Observations recorded immediately after death in infected larvae showed no change in general body colour but the darkening was noticed 8 hours after death and became shrunken with hardened carcass.

As such, the observations on symptomatology in Prodenia litura larvae showed cessation of feeding, regurgitation of food, slow activity indicating sluggishness and finally death with shrunken and darkened body and hard carcass after 8 hours of death. These were in conformity with those recorded by Steinhaus (1949), Angus and Heimpel (1956) and Ramkrishnan and Pant (1967). Cessation of feeding in Prodenia litura was also observed after 24 hours ingestion of treated food which was in agreement with those observed by Venkatraman and Ramesh chander (1967), when compared with normal feeding habit, the quantity of treated food ingested was rather extremely less. As such
the less feeding activity and the cessation of feeding would be of great importance in preventing the defoliation.

Ramkrishnan and Pant (1967) reported the hyperactivity up to 10 hours in the infected larvae of *Satia flava*, however, there was no such activity noticed in case of *Prodenia litura*. Whereas immobility observed by Stephens (1950) in *Carpocapsa pomonella* was also not observed in the infected larvae of *Prodenia litura*.

Another important characteristic symptom of sluggishness was observed in many lepidopterous insect (Steinhaus, 1949; Heimpel and Angus, 1959; and Ramkrishnan and Pant, 1967), the present studies also revealed the sluggishness in the infected larvae.

As regards the regurgitation and diarrhoea observed in *P. leperda* by Ramkrishnan and Pant (1967) larvae of *Prodenia litura* showed only regurgitation but not the diarrhoea.

Most of the lepidopterous insects were observed to show paralytic effect after ingestion of bacillus spores; however, the present investigations revealed that there were no symptoms of paralysis as also observed by Venkatraman and Kamesh chander (1967) in *P. leperda*. 
No change was observed in the colour of the integument before and after the death; however the observations recorded 8 hours after death, showed the change of colour to complete black. Such type of symptom was described as melanosis by Steinhaus (1949). In addition, shrinkage in body with hard carcass was also observed after death.

III Hydrogen-ion concentrations studies:

The observations recorded on pH studies are presented in table IV. It was noticed that the pH values differed from one region to another of the alimentary canal and the three distinct regions were observed on the basis of pH values. It was neutral in foregut, slightly acidic in the midgut, while hindgut was acidic in normal larvae. The proventriculus showed maximum of 7 pH in the alimentary canal.

As regards the blood pH, the table IV indicated that there was no change in the normal, starved, and infected with bacillus spores; while slight change was noticed in the gut pH. It was 6.76 at proventriculus in the normal larva as against 6.72 in starved and infected; indicating of a fall of 0.04 in pH; while in the midgut there was a drop of 0.04, 0.03 and 0.05 in pH at anterior, middle and posterior part of midgut respectively. No change of pH in hindgut was noticed in normal, starved and infected larvae.
Table IV
The hydrogen-ion concentrations of the blood and gut of normal starved and infected 8 day old larvae of *Pseudana litura*.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Age of larva (days)</th>
<th>No. of larvae</th>
<th>Blood pH after 24 hrs</th>
<th>pH of the gut after 24 hours feeding</th>
<th>Proventriculus</th>
<th>Anterior</th>
<th>Medium</th>
<th>Posterior</th>
<th>Hindgut</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>A. Normal larvae.</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>i) Maximum value</td>
<td>8</td>
<td>15</td>
<td>6.4</td>
<td>7.00</td>
<td>6.7</td>
<td>6.4</td>
<td>6.4</td>
<td>6.1</td>
<td>6.28</td>
</tr>
<tr>
<td>ii) Minimum value</td>
<td></td>
<td></td>
<td></td>
<td>6.7</td>
<td>6.4</td>
<td>6.4</td>
<td>6.4</td>
<td>6.4</td>
<td>6.4</td>
</tr>
<tr>
<td>iii) Average value</td>
<td></td>
<td></td>
<td></td>
<td>6.76</td>
<td>6.70</td>
<td>6.33</td>
<td>6.40</td>
<td>6.28</td>
<td></td>
</tr>
<tr>
<td>iv) Value occurred maximum times</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>v) Change from normal average</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>B. Starved larvae.</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>i) Maximum value</td>
<td>8</td>
<td>15</td>
<td>6.4</td>
<td>7.00</td>
<td>6.7</td>
<td>6.4</td>
<td>6.4</td>
<td>6.1</td>
<td>6.28</td>
</tr>
<tr>
<td>ii) Minimum value</td>
<td></td>
<td></td>
<td></td>
<td>6.7</td>
<td>6.3</td>
<td>6.4</td>
<td>6.35</td>
<td>6.28</td>
<td></td>
</tr>
<tr>
<td>iii) Average value</td>
<td></td>
<td></td>
<td></td>
<td>6.72</td>
<td>6.86</td>
<td>6.50</td>
<td>6.35</td>
<td>6.28</td>
<td></td>
</tr>
<tr>
<td>iv) Value occurred maximum times</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>v) Change from normal average</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C. Infected larvae.</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>i) Maximum value</td>
<td>8</td>
<td>15</td>
<td>6.4</td>
<td>7.00</td>
<td>6.7</td>
<td>6.4</td>
<td>6.4</td>
<td>6.4</td>
<td></td>
</tr>
<tr>
<td>ii) Minimum value</td>
<td></td>
<td></td>
<td></td>
<td>6.700</td>
<td>6.50</td>
<td>6.4</td>
<td>6.4</td>
<td>6.4</td>
<td></td>
</tr>
<tr>
<td>iii) Average value</td>
<td></td>
<td></td>
<td></td>
<td>6.72</td>
<td>6.66</td>
<td>6.50</td>
<td>6.35</td>
<td>6.28</td>
<td></td>
</tr>
<tr>
<td>iv) Value occurred maximum times</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>v) Change from normal average</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
It was thus noticed from the pH studies that no change occurred in blood and hindgut while slight fall in pH was noticed in foregut and hindgut.

Thus pH values presented in Table IV were similar in observations to those observed in Pristophora erythrogonia, Dipyron hercyniae and Malosoma disstria by Heimpel (1955) and *Sericis fabia* by Ramkrishnan and Pant (1967). Moreover, the gut pH was also observed to vary in innaniation, ecdysis and pupation by Hoeder (1955). The reduction in the pH units of the midgut in infected larvae of *P. litura* although is very slight, could be attributed to the condition of innaniation since the larval feeding on treated food stopped after an initial feeding.

Mannay and Fitz-James (1955) have reported that the crystal toxin dissolved only at higher pH i.e. 11.8. However, Ramkrishnan and Pant (1967) pointed out that the pH was not only the factor which dissolved the crystal toxin. But the midgut epithelium was quite sensitive to the crystal toxin (Fanada 1953; and Angus 1956). Moreover addition of thioglycolic acid made the crystal toxin to go into the solution at lower pH (Young and Fitz-James 1938) and enzymatic contents acting on the crystal toxin to yield two fractions both of which are toxic by ingestion and injection. But present studies of the gut pH did not show pH to that extent of 11.8 in case of *P. litura* though the
susceptibility to this pathogen was similar to that of many insect species observed by Tanada (1953), Angus (1956) and Ramkrishna and Pant (1967).

The blood pH studies in *P. litura* did not reveal any change during the period of infection as recorded by Ramkrishna and Pant (1967) in *A. sabia* and by Venkatraman and Ramesh chander (1967) in *Prodenia litura*.

The *Prodenia litura* larvae thus did not show any paralysis and change of blood pH and therefore, were in confirmation with the Venkatraman and Ramesh chander (1967). As such, *P. litura* could be placed under Group-III, as suggested by Heimpel and Angus (1939).

IV. Histological studies:

Transverse sections of the midgut of healthy and infected larvae after 9, 18, 27 and 36 hours respectively are presented in plates No. 1 to 5.

The plate No.1 exhibited the transverse section of midgut of normal larva with epithelial cells packed with cytoplasmic material and clearly seen nuclei.

The plates 2 and 3 showed the transverse section of midgut in infected larvae after 9 and 18 hours after feeding respectively in which no change in the arrangement of epithelial cells were noticed, indicating no damage to the gut cells up to 18 hours after feeding.
The plate No. 4 indicated the effect of thuricide-HP in the midgut of larvae showing loose epithelial cells detached from the basement membrane.

The plate No. 5 revealed the progressive loosening of epithelial cells from one another and moving towards the lumen indicating the sloughing in bunches with filling of lumen entirely with lysed cells.

The gross histological changes observed in *P. litura* were therefore in confirmation with the findings of Xanada (1933) Joumanoff and Vago, (1953), Hoopingarner and Materu (1964) and Ramkrishnan and Pant (1967), however explained that the spores of crystalliferous bacteria caused the breakdown of gut epithelium. In *A. fabae* (Ramkrishnan and Pant 1967) observed the changes in midgut epithelium within 24 hours; while in *A. mori* (Heimpel and Angus 1959) extensive damage was observed in 60 minutes, after feeding and 45 minutes in *P. brassicae* (Martouret et al. 1965). The present studies however indicated extensive damage to the midgut epithelial cells within 36 hours after feeding in *P. litura*.

The effect of bacterial spores on epithelium suggested a very rapid intoxication and complete destruction which were in line with the observations made by Hoopingarner and Materu (1964), though the time
required for the progressive damage could vary according to the age, species of insect and spore concentration as pointed out by Taneda (1963) and Heinzel and Angus (1959). Studies on histochemical changes would therefore reveal the specific target of action of these toxic crystals in *Prodenia litura*.
By and large, all the chemicals used in the plant protection for the insect control are highly toxic to the worm blooded animals and leave residue on the treated plants and plant products hereby restricting their use for human consumption. These have also created many more problems in the field of human welfare. In recent years, the limitations of chemical control reported to include hazards of persistant insecticides and inefficacy of chemical treatment due to sublethal dose or even the quality of the chemical. With a view to overcoming these problems, the adoption of integrated approach with the use of microbial insecticides has opened a new line of research.

The present studies therefore, were undertaken to find out the effectiveness of _B. thuringiensis_ on _Prodenia litura_ pest of many cultivated crops like castor, cotton, groundnut, tobacco and vegetables etc. The efficacy of _B. thuringiensis_ was judged on the basis of mortality studies, supplemented with histological and hydrogen-ion concentration studies. The symptoms produced in the larvae were also noticed.

Among the concentrations used, 10 percent of thuricide-HP gave 96 percent kill within 36 hours.
Symptoms produced in the *P. litura* due to the ingestion of thuricide-HP containing 30 billion viable spores of *B. thuringiensis* per gram showed that cessation of feeding within 24 hours. Regurgitation of the food after initial feeding. The other symptoms were slow activity indicating the sluggishness resulting into the ultimate death, shrunken body with change of colour and hard carcass after 8 hours of death was noticed.

The hydrogen-ion concentration showed variable pH values in different regions of alimentary canal and blood. No change in pH was observed in the blood and hindgut in normal, starved and infected larvae; whereas a slight fall in pH was noticed in foregut and midgut.

Histological studies revealed that the break down of gut epithelium showing the visible appearance of loosening of epithelial cells from one another and from basement membrane occurred in the larvae of *Prodenia litura* with the fragments of degenerated cells released into the lumen.

The present findings would be of immense value in controlling the *Prodenia litura* occurring on various cultivated crops.
REFERENCES


Berliner, S. 1911. Über die Schlafsucht der Mehlmottenraupe. 

Berliner, S. 1915 Über die Schlafsucht der Mehlmottenraupe und ihren Erreger Bacillus thuringiensis. 
Z. Angew Botanik. 3 : 29 - 56.

* sushi, P.S.; flake (Jr) H.W.; Germain G.J. 1969. Laboratory insecticide tests against the fall webworm. 
J. econ. Entom. 62 : (3) 585.

* Bullock, H.A.; Duleage H.T. 1969. B. thuringiensis against pink bollworms on cotton in field cages. 


* Cantwell, G.S.; Dutky, S.A.; Keller, J.C. and Thompson, C.G. 1961. Results of tests with Bacillus thuringiensis Berliner against Gypsy moth larvae. 
J. Insect Pathol. 3 : 143-147.


Chatterjee, P.B. 1965 The utilisation of Bacillus thuringiensis Berliner in control of A. sabulifera on jute plant. 
J. Insect pathol. 7 : 457-460

Protocheta, 11: 211 - 212.

Hall, I.M. and Dunn P.H. 1968. Susceptibility of some insect pests to infection by Bacillus thuringiensis Berliner in laboratory tests. 
J. Econ. Entomol., 51: 296-298.

Hanny, C.L. 1953. Crystalline inclusions in aerobic 
Spore forming bacteria. 

Hanny, C.L. and Fitz-James, P.C. 1955. The protein 
crystals of Bacillus thuringiensis Berliner. 

Heimpel, A.M. 1954. 
A strain of Bacillus cereus Fr. and Fr. pathogenic for larch 
sawfly, Pristiphora erichsonii 
(Htg). 

The pH in the gut and blood of 
the larch sawfly, Pristiphora erichsonii (Htg) and other 
insects with reference to the 
pathogenicity of Bacillus cereus 
Fr. and Fr. 

Heimpel, A.M. 1956. Further studies of the pH in the gut and blood of Canadian forest insects.

J. Insect Pathol. 1: 152 - 170.


Hoopingarner, R. and Materu, M.E.A. 1964. The toxicology and histopathology of Bacillus thuringiensis Berliner on Galleria mellonella L.
J. Insect pathol. 4: 26 - 30.

* Huss, B. 1938. Bacillus thuringiensis Berliner a bacterium pathogenic to corn borer larvae.


Lemoine et al. 1956. Essais d'utilisation de Bacillus thuringiensis Contre - Plut's brassicae L. Entomophage, 11: 19-34.


Japt. rend acad. ser. France. 18: 203 - 207


J. Econ. Entomol. 60: 259-262.

Ramakrishnan, N. and Pent, N.C. 1967. Some aspects of the mode of action of Bacillus thuringienensis Berliner in Spintusa tabula (Stoll)
Indian J. Entomol. 29: 149 - 153.


Shaikh, H.V. and Morrison, F.J. 1965. Notes on the susceptibility of Nasonia lecontei (Fitch) to 'Sarahene L-69' a commercial preparation of Bacillus thuringiensis var. thuringiensis.

Steinhaus, F.A. 1951  Possible use of Bacillus thuringiensis Berliner as an aid in the biological control of the alfalfa caterpillar.
    *Hilgardia*, 20 : 339 - 381.

Stephens, J.M. 1952  Diseases in Codling moth larvae produced by several Strains of Bacillus cereus Fr. and Fr.

Stephens J.M. 1957. Spore Coverage and persistence of Bacillus cereus Fr. and Fr. sprayed on apple trees against codling moth.


Tanada, Y. 1953  Susceptibility of the imported cabbage worm to Bacillus thuringiensis Berliner.

Tanada, Y. 1956  Microbial control of some Lepidopterous pests of crucifer.

* Toumanoff, C. and Vago, C. 1953. Etude histopathologique des vers a Soie atteints de Bacillus cereus Var. kloeti.

Venkatraman T.V. and Ramesh Chander 1967. Experiments on the possible use of *Bacillus thuringiensis* Berliner in the control of crop pests - II suscep-tibility of some Lepidopterous pests to *Bacillus thuringiensis* *Proc. Indian acad. sci.* 66(B) 331-336.

Videnova, E. 1966 The pathogenesis of the disease caused by *Bacillus thuringiensis* Berliner in the lucerne cootyledon. Pathology of the haeaplymph (in Bulgarian). *Biol. Vuz.* 3(7) 79-86.

