

## Effect of four commercial preparations of bacillus thuringiensis on *Opisina arenosella* walker

P. KANAGARATNAM, U. PETHIYAGODA\* and M. S. VELU  
Coconut Research Institute, Lunuwila, Sri Lanka

### ABSTRACT

The effectiveness of four commercial preparations of the bacterium, *Bacillus thuringiensis* was studied in the laboratory for the control of *Opisina arenosella* Walker. Fresh coconut leaflets treated with a range of concentrations of the four formulations were fed to larvae over a period of four days. Subsequent feeding was on fresh untreated leaflets. All four preparations caused high mortality of the larvae. Dipel was the most effective formulation. The potential for the use of *Bacillus thuringiensis* for the control of *O. arenosella* is discussed.

### INTRODUCTION

The coconut caterpillar, *Opisina arenosella* Walker (Xylorictidae) is a serious pest of coconut palms in Sri Lanka. It causes heavy damage to coconut leaves by eating the parenchymatous tissue on the ventral surface. In Sri Lanka control of this pest has been attempted by mass breeding and releasing of egg, larval, prepupal and pupal parasites. However these parasites do not establish well enough to bring the pest under effective and lasting control.

Chemical control of insects faces on the one hand mounting concern about environmental pollution by poisons, and on the other, resistance of pests to pesticides. Among the alternatives to chemical control is the potential employment of arthropod diseases. *Bacillus thuringiensis* Berliner, a spore forming bacterium is a "broad spectrum" microbial agent, causing mortality in larvae of several species of moths and butterflies. It is not harmful to humans, other mammals, birds, fish and microflora. This bacterium is now mass produced, formulated as a wettable powder and as an emulsion and is marketed commercially. Although generally lethal to insects, at sub-lethal dosages, this pathogen could also interfere with their development, lower resistance to attack by parasites, predators and other pathogens and influence the susceptibility to chemical insecticides (Falcon 1971 a).

Experiments were carried out to determine whether *O. arenosella* is susceptible to *B. thuringiensis*. Also the effectiveness of four commercial wettable powder preparations containing the bacterium were compared and the results are reported in this paper.

### MATERIALS AND METHODS

Coconut caterpillars used in the experiments were collected from the field. Caterpillars of uniform size in the third instar were selected. These were fed in the laboratory on fresh coconut leaflets before being used in the experiment. Those at immediate pre - and post - moult stages were discarded.

\* Present address - FAO, Box 163, Baghdad, Iraq.

The formulations of *B. thuringiensis* tested were, Dipel, Thuricide, Biotrol and Bactospeine in the wettable powder form. These were thoroughly mixed with distilled water to give uniform suspensions. The concentration range suitable for each formulation was found in preliminary experiments and the concentrations tested, seven each of Dipel, Thuricide and Biotrol and eight of Bactospeine are given in Table 1. The suspensions were constantly agitated to ensure uniform dispersion of spores and applied using a hand sprayer.

Table 1. Concentrations of the formulations in per cent wettable powder in water (Wt/Vol.)

Dipel	Thuricide	Biotrol	Bactospeine
0.12	0.16	0.26	0.20
0.24	0.32	0.52	0.40
0.36	0.48	0.78	0.60
0.48	0.64	1.04	0.80
0.60	0.80	1.30	1.0
0.72	0.96	1.56	1.20
0.84	1.12	1.82	1.40
			1.60

Both surfaces of the coconut leaflets were sprayed to run off. The spray fluid on the leaflets was allowed to dry before feeding to the caterpillars. Control leaflets were sprayed with water. The sprayed leaflets were kept in glass jars and 30 healthy caterpillars were transferred to each jar. The mouth of the jar was covered with thick cloth and perforated polythene sheets to prevent the escape of the caterpillars. The caterpillars were allowed to feed on sprayed leaves for four days. Subsequent feeding was on fresh unsprayed leaflets until the experiment was concluded at the end of two weeks. Mortality counts were made 4, 8, 12 and 16 days after spraying Dipel, Thuricide and Biotrol and 5, 9 and 15 days after spraying Bactospeine.

#### RESULTS AND DISCUSSION

*B. thuringiensis* caused high mortality of the larvae of *O. arenosella*. The early symptoms displayed by affected caterpillars were discoloration, flaccidity and sluggishness. The body contents of the dead larvae were of a soft fluid consistency. Occasionally there was disintegration of the integument but in most cases it remained intact. The dead larvae emitted a disagreeable odour. Limited feeding by the larvae sometimes continued until the last date of observation. The larval mortality increased with post-spray duration and with concentration for all four formulations tested. To compare the effectiveness of the four formulations the data were analysed by the method of Reed-Muench (Woolf, 1968) for the estimation of the median lethal concentration,  $LC_{50}$ . The  $LC_{50}$  for the four formulations, at different intervals after feeding with sprayed leaflets, are given in Table 2. From these results it could be inferred that Dipel was the most effective formulation with the lowest  $LC_{50}$  followed by Thuricide, Biotrol and Bactospeine in that order. The  $LC_{50}$  values decreased with the increase in duration after feeding the sprayed leaves.

#### Four commercial preparations on *Opisina arenosella*

Table 2.  $LC_{50}$  in per cent wettable powder in water (Wt./Vol.) at different intervals after spraying the four formulations

	4th day	8th day	12th day	16th day
Dipel	0.57	0.31	0.25	0.18
Thuricide	0.91	0.61	0.36	0.201
Biotrol	1.4	1.02	0.32	0.26**
	5th day	9th day	15th day	
Bactospeine	1.51	1.07	0.58	

Note. \*\*at the lowest concentration used 0.26%, cumulative mortality was 64%.

\* $LC_{50}$  is the concentration at which 50% mortality was obtained.

*B. thuringiensis*, at the time of sporulation, in addition to forming an endospore, produces a toxic proteinaceous parasporal crystal in the sporangium. During the process of infection, susceptible insects are either killed by the toxic crystals or so weakened that the bacteria can readily invade the haemocoel from the gut and produce a lethal septicæmia (Heimpel and Angus, 1963).

In the present study the initial effects on the caterpillars, following the ingestion of bacteria, was most probably due to the toxic proteinaceous parasporal crystal. The increase in larval mortality with the increase in concentration of the formulations may be due to the consumption of a larger number of toxic crystals and that with the post spray duration is probably due to infection causing septicæmia.

The feeding capacities of the larvae were affected after consuming the bacteria. Hence the damage caused to the leaves by surviving larvae was negligible. The larvae not exposed to *B. thuringiensis* in the control remained healthy over the duration of the experiment and caused severe damage to leaflets.

Studies on several lepidopterous pests have shown that mortality does not always occur in the larval stage but may be delayed until after pupation and even eclosion (Falcon, 1971 b). It is, therefore, likely that had the experiment been continued over a longer period the  $LC_{50}$  values would have been even lower.

Commercial preparations of *B. thuringiensis* are registered in the U.S.A. for use on Agricultural crops, shade trees, ornamentals and forests for the control of several insect species (Falcon, 1971 b). Although promising results were obtained in the laboratory in the present study, there are limitations to using *B. thuringiensis* for the control of *O. arenosella* in the field. This bacterium is short lived. It neither persists nor spreads in the field, and therefore cannot be used as a self perpetuating biological control agent.

This means that repeated applications would be necessary to keep *O. arenosella* under control. This would be both expensive and cumbersome, as spraying tall coconut palms is a difficult task and the commercial preparations of the bacterium are relatively expensive.

There is however a potential for the use of this bacterium for the control of *O. arenosella* on coconut seedlings. Infestations on seedlings have to be controlled quickly and parasites generally bring about control rather slowly.

Chemical insecticides are unsuitable when pasture and other intercrops are grown among seedlings. In such situations *B. thuringiensis* would be acceptable because it is harmless to farm animals, humans and wild life even if ingested from sprayed plants. This bacterium can be applied at any time upto harvest with no residue problem. Also these formulations do not have phytotoxic effects (Falcon, 1971 b).

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