A Nuclear Polyhedral Virus Disease Affecting the Larvae of Tiracola Plagiata Walk.

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ABSTRACT.

A nuclear polybedral virus of Tiracola plagiata was located at a plantation at Popondetta in Papua. The larvae are readily susceptible up to 18 days old at a virus concentration of 20,650 particles per ml. The incubation period was found to be up to eight days for 12-day-old larvae. From the evidence of field trials at a plantation near Lae, the virus is of limited use for the control of T. plagiata due to the prevailing climatic conditions and the preference of the larvae to feed on flush growth.

INTRODUCTION.

ROM 1962 to 1965, Tiracola plagiata (commonly termed cacao army worm in Papua and New Guinea) caused considerable damage to cacao (Theobroma cacao) plantings in the Popondetta-Sangara area of the Northern District of Papua and at a plantation at Lae. The larvae of T. plagiata appeared regularly during this period every six weeks devouring the young cacao flush and damaging the terminal buds resulting in the loss of vigour and growth of the trees. In September, 1965, investigations were carried out with several noctuid polyhedral viruses on T. plagiata and also a suspect polyhedral virus of T. plagiata collected by Mr. T. Bourke at a plantation at Popondetta. A search on this plantation by the author resulted in the findings of seven specimens of T. plagiata with nuclear polyhedral infection. This paper gives the investigations carried out on the polyhedral virus of T. plagiata found at the plantation at Popondetta.

DESCRIPTION OF THE DISEASE.

T. plagiata are highly susceptible to this disease during the early larval stages up to 18 days old. The disease is highly infectious and fatal with an incubation period of up to eight days. The larvae cease to feed within 24 hours of death showing little movement, becoming soft and limp. The skin appears to be thin and readily ruptures when touched liberating large quantities of white to pale pink fluid containing large numbers of polyhedra. Infected larvae, within 48 hours of death, appear pale brown in colour compared with the greenish-brown colour of the

healthy larvae. Diseased larvae in the field are usually found flattened on the upper leaf surface surrounded by whitish liquid containing the polyhedra (*Plate I*). Older larvae are not readily susceptible to the disease.

The polyhedra produced are approximately 1100 to 2400 mp in diameter and angular in outline (*Plate II*).



Plate I.—Photograph of third instar larvae at death with surrounding whitish fluid which contains the polyhedral particles.

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EXPERIMENTAL STUDIES.

Effect of age of larvae on susceptibility.

Several egg masses collected in the field at the plantation near Lae, were used to obtain a stock colony of larvae of *T. plagiata*. Every two days, 50 larvae were collected from the colony and fed with young cacao leaves on which 5 ml. of virus suspension had been smeared. The virus suspension contained 20,650 polyhedra per ml. Each day the larvae were counted and any dead larvae examined for virus infection. The larvae were kept under observation until they had pupated. The results are given in *Table* 1.

From these results, it is evident that the larvae from 2 to 18 days old are highly susceptible to infection, after which susceptibility rapidly decreases.

Effect of virus concentration on infectivity.

A stock suspension of virus material was diluted with distilled water and applied to the surface of young cacao leaves at the rate of 1 ml. per 15 sq. in. of leaf area. Larvae, four days old, from a stock colony were fed on the prepared leaves and maintained until they pupated. Larval counts were made daily and any dead ones examined for virus infection. The results are given in Table 2.

It is evident that a virus particle count of 20,650 particles per ml. will result in approximately 100 per cent. death of larvae, while a count of 13,400 particles per ml. yielded approxi-

Table 1.—The influence of the age of larvae of Tiracola plagiata on susceptibility of Tiracola nuclear polyhedral virus.

| Age of larvae when fed on virus in days. | Deaths due to virus infection.* | Control.* | Deaths due to bacterial infection. | Control. | | |
|--|---------------------------------------|-----------|---|----------|--|--|
| 2 | 50 | .0 | 0 | 5 | | |
| 4 | 48 | 0 | 2 | 3 | | |
| 6 | 49 | 0 | 1 | 2 | | |
| 8 | 47 | 0 | 3 | 4 | | |
| 10 | 48 | 0 | 2 | 1 | | |
| 12 | 50 | 0 | 0 | 3 | | |
| 14 | 47 | 0 | 3 | 2 | | |
| 16 | 45 | 0 | 5 | 2 | | |
| 18 | 47 | 0 | 3 | 3 | | |
| 20 | 16 | 0 | 7 | 5 | | |
| 22 | 12 | 0 | 6 | 6 | | |
| 24 | 3 | 0 | 2 | 1 | | |
| 26 | 2 | 0 | 8 | 5 | | |

^{*} Fifty larvae were used for each test.

Table 2.—The effect of virus concentration in respect to the minimum lethal dosage for four-day-old larvae of Tiracola plagiata.

| (Control) | | | |
|-------------|-----------|-------------------------|-------------------------|
| (COMMETON) | 0 | | 2 |
| | 0 | | 4 |
| | 3 | | 3 |
| | 8 | | 4 |
| | 21 | | 3 |
| | 49 | | 1 |
| | 46 | | 4 |
| | (Control) | 0 3 8 21 49 | 0 3 8 21 49 |

* Fifty larvae were used for each test.

mately 50 per cent. Thus a concentration of 20,650 particles per ml. was used for the remaining experiments and the field trial.

Period of incubation of the virus in the larvae.

Thirty larvae from the stock colony, four and twelve days old, were fed with the virus suspension (20,650 particles per ml.) on cacao leaves and the period of incubation determined. The results are given in *Table 3*.

For four-day-old larvae, the incubation period was found to be from three to six days, and for the 12-day-old larvae, three to eight days.

Field Trial.

In November, 1965, and January, 1966, field trials were laid down at the plantation near Lae to determine the efficiency of the virus in controlling *T. plagiata*, in the field. A limited trial was laid down in November and January; 100 cacao seedlings (18 months old) were sprayed with virus suspension (concentration 20,650 particles per ml.) with a "Motoblo" superseventy, powered knapsack sprayed with a nozzle setting of No. I and minimum engine power. Five litres of spray were used for 100 trees.

Table 3.—Determination of the incubation period, in four and twelve-day-old larvae of Tiracola plagiata, of the Tiracola nuclear polyhedral virus.

| Age of lar in days. | | | | fter | | | on | | Total virus deaths. |
|------------------------|----|----|----|------|----|---|----|---------|---------------------|
| | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | RATE OF |
| 4a | 1b | 0 | 2* | 17 | 9 | 1 | 0 | 0 | 29 |
| 12 | 0 | 2b | 3 | 7 | 8 | 6 | 4 | 1 1b | 27 |
| Control | | | | | | | | | |
| 4 | 0 | 0 | 1b | 0 | 0 | 0 | 0 | 0 | 0 |
| 12 | 0 | 0 | 0 | 0 | 2b | 0 | 0 | 0 | 0 |

a. Thirty larvae used in each age group.

b. Death due to bacterial infection.

^{*} Numbers in table refer to the number of larvae found to be dead on that day.

Table 4.—Recordings in the field trial carried out at a plantation near Lae, in January, 1966, on the number of dead larvae of *Tiracola plagiata* infected with *Tiracola* nuclear polyhedral virus.

| | | | | | virus. | | | | | |
|------------------|---------|-------|-----------|------|---------|---------------|------|------|------|-------|
| Tree Number * | 13/1 | 16/1 | 17/1 | 18/1 | 19/1 | 20/1 | 21/1 | 22/1 | 24/1 | 25-28 |
| 3 | 0 | Т | (lateral) | 11 | Т | 0 hf | 0 | T | 2 | 0 |
| 11 | 0 | T | 43 | 17 | 0 | 0 hf | 0 | T | 0 | 0 |
| 12 | 0 | T | T | 3 | 0 hf | 0 | 0 | T | T | 0 |
| 16 | T | T | T | 6 | 4 | 3 | 7 | 1 | T | 0 |
| 18 | 0 | T | T | T | 0 | T hf | 6 | T | 0 | 0 |
| 23 | 0 | T | T hf | T | 0 | T | 1 | 1 | 1 | 0 |
| 26 | 0 | T | 1 | 18 | 0 | T hf | 2 | T | 1 | 0 |
| 27 | T | T | T | T | T | T | 16 | 0 | T | 0. |
| 29 | T | T | T | T | T | T hf | 23 . | T | 2 | 0 |
| 31 | 0 | Т | 1 | 1 | T | T | 1 | 2 | 6 | 0 |
| 33 | 0 | Т | T | T | 6 | T | 9 | T | T | 0 |
| 35 | 0 | T | T | T | 3 | T | 5 | Т | 1 | 0 |
| 37 | T | T | T | 3 | 0 | 2 hf | 5 | Т | 0 | 0 |
| 38 | T | Т | T | T | 0 | T | 9 | Т | T | 0 |
| 52 | T | Т | 3 | 16 | 2 | T | 17 | T | 0 | 0 |
| 55 | 0 | T | Т | T | 4 | 3 hf | 11 | 2 | T | 0 |
| 56 | 0 hf | T | T | Т | Т | T | 5 | 0 | 3 | 0 |
| 57 | 0 | T | T | Т | 3 | T | 2 | T | T | 0 |
| 63 | Т | Т | 2 | 29 | T | 6 hf | 0 | 2 | 0 | 0 |
| 68 | T | T | 3 | 16 | 0 | 1 hf | T | 0 | T | 0 |
| 70 | T | out T | 3 | 2 | 3 | 2 hf | 1 | Т | 0 | 0 |
| 74 | Т | Т | 27 | 7 | 1 | T hf | T | Т | Т | 0 |
| 84 | 0 | Т | T | 3 | 7 | 2 hf | 6 | Т | 3 | 0 |
| 97 | Т | T | T | 1 | 6 | T hf | 7 | 4 | Т | 0 |
| 99 | T | T | T | 2 | 0 | ht 1 hf | Т | Т | 1 | 0 |

^{*}The trial area consisted of 100 trees and 25 tree records were selected from the random numbers table from 'Statistical tables for biological, agricultural and medical research' by R. A. Fisher and F. Yates (1949).

hf = Hard flush on trees only.

T = Tiracola plagiata larvae present on the tree.

Most of the trees had a heavy infestation of Ophiusa janata larvae which had resulted in considerable flush damage. The cacao trees and smaller shade trees (Leucaena leucocephela) were examined daily for dead Tiracola plagiata larvae. Daily rainfall recordings were also carried out. The results from 25 randomly selected trees are given in Table 4.

Apart from tree II, which had an egg mass present when sprayed, the number of larvae per tree was low and a large number of deaths resulted. The trial was of limited use, due to the fact that on 20th January, 1966, no young flush was present on the trees, and on this date, larvae in plague numbers descended from the taller Leucaena leucocephela, onto the cacao, but few remained to feed. From field observations, the majority of T. plagiata eggs are laid on Leucaena leucocephela, but the larvae feed only for a short time as the young growth of cacao is preferred.

Effect of Tiracola nuclear polyhedral virus on several other species.

Ectropis sp. and Ophinsa janata are also cacao flush eating in the larval stages and can cause considerable damage in the field. Fifty young larvae of each species were fed cacao leaves treated with Tiracola polyhedral virus suspension in the laboratory. None of the larvae in this experiment died as a result of virus infection. Tachinid flies which are predators of Tiracola plagiata were found to be unaffected by the Tiracola polyhedral virus.

DISCUSSION.

From the experimental data obtained, the *Tiracola* nuclear polyhedral virus could be employed as a useful additive for the biological control of *Tiracola plagiata* outbreaks in cacao in Papua and New Guinea. It can be readily applied by conventional spray machinery. How-

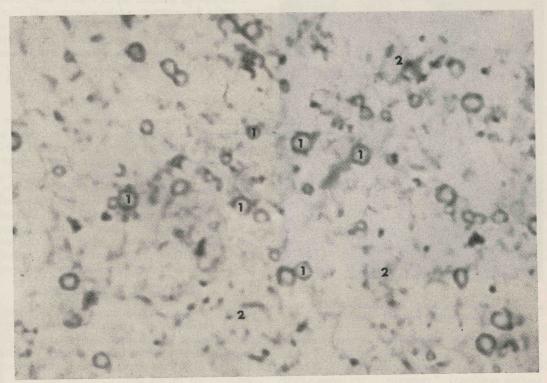


Plate II.—Photomicrograph of Tiracola plagiata viral polyhedra (1) taken at a magnification of 2,700 times, stained with 0.1 per cent, methylene blue. Bacteria (2) are also present in the preparation. (Photograph by Dr. D. E. Shaw.)

ever, the virus suspension would need to be applied in the early larval stages and this is not always possible since *T. plagiata* moths lay their eggs mainly on the cacao shade tree and the larvae descend onto the cacao eight or more days after hatching. The amount of flush on the cacao trees will also influence the effectiveness, as the larvae will move off the trees onto the ground when no flush is present. Only limited persistence in the field was observed at a plantation near Lae over a period of three months due to the regularity of the rainfall. This is an impor-

tant defect in natural field infection, as it does not allow the concentration of virus material to build up.

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