A Nuclear Polyhedral Virus of Catopsilia Pomona in Papua and New Guinea.

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ABSTRACT.

A virus located at Popondetta in the Northern District of Papua affecting the larvae of Catopsilia pomona *is a new record and the virus belongs to the nuclear polyhedral group of insect viruses. The virus appears to be restricted to* C. pomona *and readily infects with resulting death all larval stages of* C. pomona. *The polyhedra range from* 1,000 *to* 2,000 *mμ in diameter with an average diameter of* 1,170 *mμ. A prepared suspension was still highly infectious after seven months when stored in suspension in the laboratory, but persistence in the field could not be maintained for a long period.*

INTRODUCTION.

IN the Territory of Papua and New Guinea many ornamental *Cassia* spp. which are grown in household gardens are periodically infested with larvae of *Catopsilia pomona* F. The larvae feed on the young emerging leaves and often continually destroy the leaf flushes of young trees, resulting in their death. At Keravat, in the New Britain District of New Guinea larvae of *C. pomona* were observed feeding on the leaves of *Cassia grandis* L., *C. fistula* L., *C. alata* L. and *C. moschata* L. In January, 1966, *Catopsilia pomona* larvae were found infected with a polyhedral virus at Popondetta, in the Northern District of Papua by Mr. T. Bourke. Investigations were carried out at Keravat to determine the effectiveness of the virus in reducing larval populations of *C. pomona.* The experimental data and the results of the investigations are described in this paper.

DESCRIPTION OF THE DISEASE.

The pigmentation of healthy larvae of *Catopsilia pomona* varies from a pale green to dark metallic blue on the dorsal surface, with a narrow white longitudinal stripe along the length of the body on both sides. The larvae of *C. pomona* infected with the nuclear polyhedral virus within 24 hours preceding death exude a dark brown fluid containing many polyhedral particles from the mouth and anal orifice. At death the body contents are a pale yellow to light brown fluid containing numerous polyhedra and the skin is readily ruptured. The skin is a yellowish-green in the case of the green coloured larvae and the dark blue larvae are usually pale yellow on the ventral surface.

In the field, infected larvae move to mature leaves and usually cease to feed within 24 hours preceding death. At death, the larvae appear flattened on the upper leaf surface surrounded

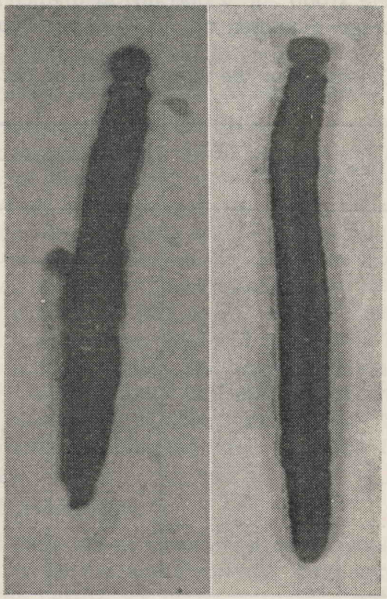

Plate I.—Photograph of the fourth instar larvae of *Catopsilia pomona.* The larva on the left is dead due to the nuclear polyhedrosis, with the skin ruptured and brown fluid oozing from the break. The larva at the right is healthy.

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Table 1.—The susceptibility of seven larval species to the nuclear polyhedral virus of *Catopsilia pomona* at a concentration of 135,000 particles per ml.

Insect Species.	Number of Larvae Fed with Virus.	Number of Deaths	
		Bacteria.	Virus.
Catopsilia pomona	50	4	46
Acheae janata L.	50	3	0
Spodoptera litura F.	50	0	0
Orgyia postica Walker	50	0	0
Ectropis sabulosa Warr.	30	0	0
Hyposidra talaca Walker	30	4	0
Euproctis sp	30	3	0

by a pale yellow to light brown exudate (*Plate* I). In laboratory feeding trials no adults emerged from pupae formed from larvae which had ingested polyhedra. Four to six days after pupation, the pupal cases were found to contain a yellowish to light brown fluid containing masses of polyhedral particles.

The polyhedral particles were observed in the nuclei of the cells of fat bodies and the blood and were 1,000 and 2,000 mμ in diameter, with an average diameter of 1,170 mμ. From the observations of infected larval cells and differential staining, the polyhedral virus is of the nuclear group.

Table 2.—The effect of virus concentration of the nuclear polyhedral virus on six-day-old larvae of *Catopsilia pomona*.

Particles per ml.	Number of Deaths due to.	
	Virus.	Bacteria.
24×10^6	58	2
2×10^6	57	3
2.8×10^5	53	7
3.4×10^4	56	4
9×10^3	50	10
1,250	46	14
100	51	9
CONTROL	0	11

EXPERIMENTAL STUDIES.

Host range.

Seven different larval species from two different families were fed the *Catopsilia* nuclear polyhedral virus at a concentration of 135,000

Table 3.—Effect of the age of larvae of *Catopsilia pomona* on susceptibility to *Catopsilia* nuclear polyhedra virus.

Day of Count.	Age of Larvae in days.						
	1	3	5	7	9	11	13
0	0a	0a	0a	0a	0a	0a	0a 15p
1	0	0	0	0	0	0	0c 38p
2	0	0	0	0	0	18 12p*	0 6p
3	0	0	0	0	16	15 6p*	....
4	0	0	12	4	27	5	....
5	33 2b	12	19	24	16	3	....
6	9	38 2b	13	25	....	....	....
7	14	7	16	5	....	....	....
8	....	1	....	....	....	....	....
9	....	....	....	....	....	....	....

Undesignated numbers refer to the number of larval deaths due to virus infection.
a—Day on which 60 larvae were fed the virus suspension.
b—Deaths due to bacterial infection.
c—Pre-pupal larvae did not feed on the treated leaves.
p—Number of pupal cases.
*—Pupae died four to seven days later with virus infection.

Table 4.—The effect of storage at 25 degrees C. on the infectivity of the nuclear polyhedral virus of *Catopsilia pomona.*

Storage period of virus in Months.	Days after Treatment.								
	1	2	3	4	5	6	7	8	9
1	0*	0	0	5	27 5b	6 3b	2 2b	....	....
3	0	0	0	3 1b	23 3b	14 1b	3 2b	....	....
5	0	0	0	1	25 2b	12 2b	7 1b	....	....
7	0	0	0	0	21 3b	17 2b	6 1b	....	....

Undesignated numbers refer to the number of larval deaths due to virus infection.

*—Fifty six-day-old larvae were treated with the virus applied to young leaves of Cassia fistula.

b—Number of larvae dead due to bacterial infection.

particles per ml., applied to young cacao leaves at the rate of one ml. per 100 square cm. of leaf area. For *Catopsilia pomona* larvae, the virus suspension was applied to young *Cassia fistula* leaves. The larvae were fed the virus treated foliage when five days old and maintained until they pupated and adults emerged, or until death. Control colonies were also set up. The results given in *Table* 1 show that only the larvae of *Catopsilia pomona* were found susceptible to the virus.

Effect of virus concentration on infectivity.

Eggs of *C. pomona* were collected from stock colonies and hatched in petri dishes on young *Cassia fistula* leaves in the laboratory. When the larvae were six days old, they were fed *C. fistula* leaves treated with the nuclear polyhedral virus at various concentrations, applying the virus suspension at the rate of one ml. per 100 square cm. of leaf surface. The larvae were counted every 24 hours and the dead larvae checked for the presence of the virus. Control populations were also maintained and remained virus-free throughout the experiment. From the results in *Table* 2, the populations of *Catopsilia pomona* were killed with a concentration of 100 particles per ml.

Effect of larval age on susceptibility.

C. pomona larvae of ages ranging from 1 to 13 days old, hatched from a stock colony, were fed *Cassia fistula* leaves which had been treated with nuclear polyhedra at the rate of one ml. per 100 square cm. at a virus concentration of 9,000 particles per ml. The larvae were fed and checked every 24 hours and any dead larvae were checked for virus infections. Control cages were also set up for each age group.

It is evident from the results given in *Table* 3 that the age of the larvae of *Catopsilia pomona* has no influence on the susceptibility of the larvae to virus infection. However, the incubation period, that is the time from ingestion to death, was shorter in the larvae of 11 days old compared with one-day-old larvae. It is interesting to note that pupae developed from infected larvae died due to viral infections.

Longevity of the virus in suspension.

A suspension of *Catopsilia* nuclear polyhedra of a concentration of 9,000 particles per ml., was prepared three weeks after field collection and stored at 25 degrees C. in water. Every two months after the date of field collection six-day-old larvae of *C. pomona* were fed the suspension applied to young *Cassia fistula* leaves, at the rate of one ml. per 100 square cm. of leaf tissue. Daily counts were made of the larvae and any dead larvae checked for virus infections. The results are presented in *Table* 4, and show that the infectivity of the preparation has not apparently deteriorated over a period of seven months.

Field persistence.

As there are no extensive areas of young cassia trees in the Gazelle Peninsula, field spraying to test the persistency of the virus in the field was carried out on scattered individual trees. A virus suspension at a concentration of 200,000 particles per ml. was applied to young leaf flush when eggs and young larvae of *Catopsilia*

Table 5.—The efficiency of the nuclear polyhedral virus of *Catopsilia pomona* in controlling infestations of *Catopsilia pomona* on five cassia trees in the field. The virus suspension applied had a concentration of 200,000 particles per ml.

Tree No.	Days after Spray Application.																													
	0		4		8		12		16		20		24		28		32		36		40		44		48		52		56		60	
	H	V	H	V	H	V	H	V	H	V	H	V	H	V	H	V	H	V	H	V	H	V	H	V	H	V	H	V	H	V
1	30	0	2	22	7	6	12	5	10	3	7	2	8	0	15	0	31	0	48	0	46	0	33	0	35	0	28	0	17	0	21	0
2	16	0	0	12	3	1	5	3	2	4	3	1	2	0	3	0	3	0	3	0	2	0	2	0	1	0	5	0	8	0	15	0
															←								→									
															No flush present																	
3	10	0	0	6	3	0	8	2	3	3	9	2	7	0	4	0	2	0	2	0	2	0	0	0	3	0	6	0	12	0	17	0
															←										→							
															No flush present																	
4	13	0	1	9	5	3	8	2	7	3	0	0	0	0	0	0	0	0	0	0	0	0	1	0	2	0	8	0	13	0	43	0
											←										→											
											Ants present (a)																					
5	15	0	0	9	3	0	8	2	11	5	8	6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	15	0	28	0
															←										→							
															No flush present																	
Rainfall in points			34		16		146		11		4		4		34		85		8		248		69		307		80		291			

H—Figures in this column are the number of healthy larvae of C. pomona.
V—Figures in this column are the number of virus infected larvae of C. pomona.
(a) Oecophylla smaragdula.

pomona were present. The trees were checked every four days for a period of 60 days recording the number of feeding larvae present and, if any, dead larvae containing viral polyhedra. The results are given in *Table* 5.

In the first three weeks after the virus application, larvae infected with viral polyhedra were located on the treated trees, with the number of infected larvae decreasing with time after spraying. In the remaining six weeks no diseased larvae were located on the trees. However, the results are limited in that three trees had no flush for the larvae to feed on for a period of 20 to 24 days and another tree became infested with kurukum ants (*Oecophylla smaragdula*) which preyed on any *Catopsilia* larvae present on the tree. During this observation period 13.27 in. of rain were recorded, and on examining leaf material from the treated trees, it was evident that a concentration of viral polyhedra was not being maintained on the young emerging leaves. Thus it is considered that field control of *Catopsilia pomona* larvae by *Catopsilia* nuclear polyhedrosis virus can only be obtained by regular application of the virus material to infested trees.

CONCLUSIONS.

The virus located at Popondetta affecting *Catopsilia pomona* is a new record and belongs to the nuclear polyhedral group of insect viruses, and appears from the limited host range test conducted to be restricted to *C. pomona.* The virus readily infects with resulting death all laval stages of *C. pomona.* A prepared suspension was still highly infectious after seven months when stored in suspension in the laboratory, but persistence in the field could not be maintained for a long period.

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