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EFFECT OF NUCLEAR POLYHEDROSIS VIRUS ON THE LARVAE OF DIAMONDBACK MOTH, *PLUTELLA XYLOSTELLA* L.

K. Padmavathamma[1] and G.K. Veeresh
University of Agricultural Sciences, GKVK, Bangalore 560 065

Abstract: A laboratory experiment was conducted to study the efficacy of nuclear polyhedrosis virus (NPV) with nine concentrations on larval mortality, incubation period, rate of pupation, pupal mortality and emergence of malformed and normal adults of all instars of *Plutella xylostella* L. The incubation period of the virus increased and mortality decreased with advancement in age of the caterpillars. Pupation rate and mortality decreased with increase in polyhedral inclusion bodies in all instars. Higher percentage of malformed adult emergence was observed with lower concentration of virus infection in early instars. Normal adult emergence was higher in lower concentrations of virus fed to the early instars.

INTRODUCTION

Nagarakatti and Jayanth (1982) reported for the first time the occurrence of a nuclear polyhedrosis virus (NPV) on the larvae of *Plutella xylostella* L. in Bangalore, but no work was carried out on this virus in India. During March 1987, diseased larvae of diamondback moth were found in a cabbage field of Sriramapura village near Bangalore and on examination, the infected individuals revealed the presence of nuclear polyhedral inclusion bodies (PIB). The possibility of using this viral agent for the control of diamondback moth was found promising one. Hence, detailed studies were undertaken on the effect of NPV on larval and pupal mortality, incubation period and adult emergence of this insect; the results of which are reported in this paper.

MATERIALS AND METHODS

The purified concentrated suspension of polyhedra isolated from the diseased dead larvae of P. *xylostella* was used as infective material as described by Bakwad and Pawar (1981). A standard haemocytometer was used for counting the polyhedra and serial dilutions were prepared from the stock suspension.

Larvae of P. *xylostella* of uniform age and size from the laboratory culture were used for this study. Nine concentrations of virus viz., 1.7×10^9, 1.7×10^8, 1.7×10^7, 1.7×10^6, 1.7×10^5, 1.7×10^4, 1.7×10^3, 1.7×10^2 and 1.7×10^1 PIB/ml suspensions were used for infecting the larvae. The suspensions had 0.1 per cent teepol (used as wetting agent). Fifty larvae from each instar starved for 6 hours were placed in paper cups (3 x 6 cm) individually. The cabbage leaves cut into bits of 3 cm^2 area each were uniformly smeared with each of the above mentioned virus suspensions. The leaf bits were dried in shade and fed to the larvae (one leaf bit/larva) for 24 hours. Fresh uncontaminated leaves were supplied daily thereafter. The experiment was conducted at 28.5± 2°C and 89+2 per cent relative humidity.

Another set of 50 larvae in each instar fed with leaf bits smeared with sterile distilled water served as control. Daily observations were made on larval mortality, incubation period, rate of

Present address: 1. S.V. Agricultural College, Tirupati 517 502, A.P., India

pupation, pupal mortality and emergence of malformed and normal adults pertaining to each instar. Microscopic examination was conducted in doubtful cases by preparing smears from the dead larvae.

RESULTS AND DISCUSSION

Incubation period and larval mortality:

There was an increase in the mean incubation period and a decrease in mortality as the larvae advanced in age, indicating a decrease in susceptibility. The incubation period was less and larval mortalities were higher at higher concentrations of virus (Tables 1-4). The mean incubation period of 3.43 days at the highest concentration of 1.7×10^9 PIB/ml was recorded along with 100 per cent larval mortality in the case of first instar larvae; while in the fourth instar larvae the mean incubation period was 6.29 days and the larval mortality was 74.67 per cent at the same concentration (Tables 1 and 4). The results are in close agreement with the observations of Legacion and Gabriel (1978) in *Spodopteralitura* (Hb.). The virus at 17.5×10^6, 12.1×10^7 and 90.05×10^8 PIB/50 ml concentrations fed to larvae caused mortality of 100 percent in the case of first instar, whereas in the fifth instar, these three concentrations caused 18, 45 and 43 per cent mortality, respectively. Higher concentration of the virus caused early death. The above mentioned concentrations caused death after application in 13.5, 12.5 and 11 days in the first instar larvae and in 12.81, 13.24 and 14 days in the fifth instar larvae, respectively. Lathika and Jacob (1974) observed decreased larval susceptibility and longer incubation period of the virus with advancement of the age of the larvae of *Spodoptera mauritia* (Boisduval).

Pupation and adult emergence:

Higher percentage of pupation was observed with lower viral concentrations and the pupal formation decreased as the dosage of PIB/ml increased in the first, second and third instars. But in the fourth instar, the pupation occurred even in higher concentrations of virus showed that these individuals were somewhat resistant to the virus unlike the early instars (Tables 1-4). Similar findings were observed by Smirnoff (1961) in *Neodiprion swainei* (Middleton) which showed that the occurrence of death was rapid in young larvae than in old larvae and also when the infection occurred in third or later instars, the progress of the disease was slower and a larger population survived to spin cocoons.

Pupal mortalities decreased with the decrease in the viral concentrations and increase in the age of the host in the present study. Jacob *et al.* (1978) also found that the NPV of *Nymphula depunctalis* (Gn.) caused 60 to 70 per cent mortality of some fresh larvae in 5 to 6 days and those infected in the later instars were able to complete larval development, but died in the pupal stage.

At low concentrations, greater degree of malformed adult emergence occurred in the first, second and third instar infections compared to fourth instar NPV infected larvae. The latter was probably due to the sublethal dose of inoculum of NPV and less duration of exposure to virus as well as maturation immunity. Legacion and Gabriel (1978) observed similar trend of survival of some infected larvae in the larval stage which gave rise to pupae or malformed adults in *S. litura*.

Table 1. Effect of nuclear polyhedrosis in the first instar larvae of P. *xylostella*

Viral concentration PIB/ml	Inoculation period days		% larval mortality	% pupation	% pupal mortality	% malformed adult emergence	% normal adult emergence
	Range	Mean					
1.7×10^9	3-4	3.43	100.00	0.00	0.00	0.00	0.00
1.7×10^8	3-4	3.45	100.00	0.00	0.00	0.00	0.00
1.7×10^7	3-5	3.57	100.00	0.00	0.00	0.00	0.00
1.7×10^6	3-5	3.87	100.00	0.00	0.00	0.00	0.00
1.7×10^5	3-6	4.98	05.33	4.67	100.00	0.00	0.00
1.7×10^4	3-6	5.13	84.67	15.33	65.20	34.80	0.00
1.7×10^3	4-7	5.58	80.00	20.00	33.33	66.67	0.00
1.7×10^2	5-7	6.31	58.00	42.00	13.21	52.83	33.96
1.7×10^1	5-8	6.65	40.00	60.00	5.56	3.33	91.11

Table 2. Effect of nuclear polyhedrosis in second instar larvae of P. *xylostella*

Viral concentration PIB/ml	Inoculation period days		%larval mortality	% pupation	% pupal mortality	% malformed adult emergence	% normal adult emergence
	Range	Mean					
1.7×10^9	4-5	4.38	100.00	0.00	0.00	0.00	0.00
1.7×10^8	4-5	4.45	100.00	0.00	0.00	0.00	0.00
1.7×10^7	4-6	4.55	100.00	0.00	0.00	0.00	0.00
1.7×10^6	4-7	4.90	95.33	4.67	100.00	0.00	0.00
1.7×10^5	4-7	5.52	86.00	14.00	100.00	0.00	0.00
1.7×10^4	4-7	5.70	66.00	34.00	27.45	72.55	0.00
1.7×10^3	5-8	6.46	58.00	42.00	15.87	38.10	46.03
1.7×10^2	6-9	7.12	46.00	54.00	8.64	22.22	69.14
1.7×10^1	6-10	8.28	26.00	74.00	0.00	10.81	89.10

Table 3. Effect of nuclear polyhedrosis in third instar larvae of P. *xylostella*

Viral concentration PIB/ml	Inoculation period days Range	Mean	% larval mortality	% pupation	% pupal mortality	% malformed adult emergence	% normal adult emergence
1.7×10^9	5-6	5.31	100.00	0.00	0.00	0.00	0.00
1.7×10^8	5-7	5.43	100.00	0.00	0.00	0.00	0.00
1.7×10^7	5-7	5.65	100.00	0.00	0.00	0.00	0.00
1.7×10^6	5-8	5.84	93.33	6.67	100.00	0.00	0.00
1.7×10^5	5-8	6.00	78.67	21.33	75.00	25.00	0.00
1.7×10^4	5-9	6.62	60.67	39.33	23.73	57.63	18.64
1.7×10^3	6-9	7.45	50.00	50.00	6.67	22.67	70.66
1.7×10^2	6-10	7.58	38.00	62.00	0.00	21.51	78.49
1.7×10^1	6-10	7.65	16.67	83.33	0.00	7.20	92.80

Table 4. Effect of muclear polyhedrosis in fourth instar larvae of P. *xylostella*

Viral concentration PIB/ml	Inoculation period days Range	Mean	% larval mortality	% pupation	% pupal mortality	% malformed adult emergence	% normal adult emergence
1.7×10^9	5-8	6.29	74.67	25.33	55.26	44.74	0.00
1.7×10^8	5-8	6.68	70.67	29.33	34.10	65.90	0.00
1.7×10^7	6-9	7.38	51.33	48.67	19.18	60.27	20.55
1.7×10^6	7-10	8.08	45.33	54.67	10.98	35.37	53.65
1.7×10^5	7-10	8.10	36.67	63.33	5.26	18.95	75.79
1.7×10^4	7-10	8.20	30.00	70.00	0.00	17.14	82.86
1.7×10^3	8-11	9.56	26.00	74.00	0.00	9.01	90.99
1.7×10^2	8-11	10.08	8.67	91.33	0.00	0.00	100.00
1.7×10^1	-	-	0.00	100.00	0.00	0.00	100.00

Table 2. Mean values for the ratios in different layers

Elevation	Depth (cm)	Ratios						
		OC:N	OC:P	OC:S	N:P	N:S	P:S	OC:N:P:S
E1	0-15	10.93	125.89	62.67	11.52	5.73	0.50	141:12.90:1.12:2.25
	15-50	14.12	528.00	63.46	37.40	4.50	0.12	132:9.35:0.25:2.08
	50-100	13.11	420.00	53.16	32.08	4.05	0.13	101:7.70:0.24:1.90
E2	0-15	13.40	120.00	46.05	8.96	3.44	0.38	198:14.78:1.65:4.30
	15-50	12.51	226.56	43.15	18.11	3.45	0..19	145:11.59:0.64:3.36
	50-100	11.55	423.08	37.80	36.62	3.27	0.09	110:9.52:0.26:2.91
E3	0-15	16.23	139.04	48.61	8.57	2.99	0.35	349:21.50:2.51:7.18
	15-50	11.74	245.45	32.99	20.91	2.81	0.13	162:13.80:0.66:4.91
	50-100	1081	394.74	41 90	36.53	3.88	0 11	150:13 88:0 38:3 58

in E, the decomposition is rapid when compared with E, and E_3 which has a warmer superhumid climate.

Similar to OM, total N also was found to increase with elevation and it showed close correlation with OM. The relatively higher proportions of N at E_2 and E, can he adduced to the ability of N forming a variety of compounds in association with other constituent parts of OM, such as fats and waxes, carbohydrates, lignin and various humus fractions.

Organic P was also found to be governed by and associated with elevation. As OM content increases with elevation, the higher organic P at E, and E_3 can be traced back to an increase of OM.

The organic S content increased with elevation. The relatively higher content of organic S in E_2 can be partly due to the retention by finer fractions which increase with elevation.

The OC:N ratio of soils in E_1, E_2 and E, revealed that this ratio remarkably maintained itself at a constant ratio of 12:1. This is a unique phenomenon in the sense that with large additions of fresh OM which has a widening effect on the OC:N ratio and a rapid rate of decomposition of OM which has a narrowing effect on the ratio, OC:N ratio is observed to be near constant at 12:1.

The ratios of OC:P manifested that in E, with low rainfall, the values were found to be lowest. As elevation increased, the P contents also increased. Soil conditions such as aeration, temperature, texture and reaction would affect both OC and organic P. At lower altitude the above effect was found to be more pronounced on OC than on organic P. The higher values for OC:P ratio in E_2 and E_3 could be attributed to higher OM content and relatively lesser proportion of organic P. The microbial activity in E_2 and E, was also found to be less as a result of which OC:P ratio widened.

There was a progressive reduction in the OC:S ratio with elevation. It can be inferred that this narrow ratio results from the accumulation of S compounds which form strong linkages with humic substances and therefore resist decomposition. This can also be explained by the fact that under extreme acidic conditions in the higher elevations, the activity of the bacteria, especially S oxidising ones is very much inhibited and this causes greater accumulation of S and consequently lowering of OC:S ratio (Misra *el al.*, 1990). On the contrary, with decrease in acidity, the conditions become favourable for S oxidising bacteria and hence a high ratio at higher pH.

The N:P ratios decreased with elevation. Regular changes in N:P ratio in accordance with OC:P ratio is due to the constancy in the