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Effect of Temperature And Storage Time On The Infectivity of Granulosis Virus of Sugarcane Shoot Borer, *Chilo infuscatellus* Snell.

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ABSTRACT

The susceptibility of sugarcane shoot borer, *Chilo infuscatellus* Snell., to the granulosis virus at four different rearing temperatures *viz*., 20, 25, 30 and 35°C and the infectivity of the virus stored at three different temperatures *viz*., 0,4 and 28°C for three years were studied. The virus caused high and rapid mortality of third instar larvae when the post-treatment temperature ranged from 30 - 35°C. There was a decrease in the mortality rate and an increase in the time taken for kill with decrease in the temperature. Bioassay studies indicated that infectivity of the virus was not reduced significantly even when the virus was stored for three years either at 0, 4 or 28°C.

KEY WORDS: Granulosis virus, *Chilo infuscatellus*, temperature, storage, infectivity

The granulosis virus (GV) infecting the sugarcane shoot borer, *Chilo infuscatellus* Snell., (Easwaramoorthy and David, 1979) is found widely distributed in the sugarcane ecosystem in Tamil Nadu and Pondicherry (Easwaramoorthy and Jayaraj, 1987) causing considerable mortality of the host. The virus, when multiplied and applied is found effective in suppressing the pest population (Easwaramoorthy and Jayaraj unpubl., Easwaramoorthy and Santhanalakshmi, 1988) in the trials conducted in Tamil Nadu and Karnataka. For extensive field testing, large quantity of the virus has to be produced and stored. In this connection, studies have been carried out to determine the optimum post-treatment temperature required for the virus to cause high mortality of the host and the influence of storage temperature and time on the infectivity of the virus to shoot borer larva.

MATERIALS AND METHODS

Production of virus

Field - collected early third instar larvae were orally inoculated with purified GV capsules. Moribund diseased larvae were collected and placed in water. Capsules were released from cadavers by maceration and filtering through two layers of muslin. Purification was carried out using cycles of differential centrifugation and the concentration was determined using a Petroff Hauser and Helber counting chamber with 0.02 mm depth.

Effect of temperature

The susceptibility of shoot borer larvae to the virus was studied under a range of temperatures. Healthy third instar shoot borer larvae were microfed with 1 µl of the virus suspension containing 1.1×10^4 and 1.1×10^5 inclusion bodies (IBs) / larvae using an Agla micrometer syringe. Care was taken to discard those larvae which failed to ingest the entire quantity of the inoculum. The control larvae were fed with an equal quantity of distilled water.

The larvae were placed at the rate of three in a plastic box (7.0 cm dia x 7.5 cm ht) provided with filter paper circle at the bottom to absorb excess moisture and three pieces (5-6 cm length) of sugarcane shoot bits (Variety Co 6304) split open at one end. The larvae were held at four different temperatures *viz*.., 20, 25, 30 and 35°C in constant temperature cabinets (BOD incubators). The treatments were replicated thrice with 30 larvae in each replication. The mortality of larvae due to virus infection and other causes were recorded daily after changing the shoot bits and filter paper.

Effect of storage time and temperature

Aliquots of 10 ml of fresh, purified virus at the concentration of 1-2 x 10^9 IBs/ml were stored at 28, 4 and 0°C temperatures at quarterly intervals for 3 years. At the end of the test period, the activity of the virus suspensions was bioassayed using third instar larvae of shoot borer by micro feeding method at the dose of 1 x 10^6 IBs/larva. Before micro feeding, the concentration of the original stock solution was determined and suitable dilutions made to get approximately uniform concentration of 1×10^6 IBs/µl. The treatments were replicated thrice with 20 larvae / replication and the experiment was conducted at 28 ± 1°C. Data on per cent larval mortality and time taken for kill were collected.

RESULTS AND DISCUSSION

Among the various incubation temperatures tested, 30°C was found to be more favourable for the virus to cause a mortality (82.9 per cent). This was followed by incubation at 35°C in causing increased mortality of 81.1 per cent and the differences observed in the mortality between these two temperatures were not significant. A least mortality of 41.1 per cent was recorded when the post-treatment temperature was 20°C (Table 1). There was no significant variation in the mortality of larvae fed with 1.1×10^4 or 1.1×10^5 IBs of the virus.

The time taken for kill was low at 30°C followed at 35°C. It was significantly more (17.3 days) when the larvae were reared at 25°C and was the maximum (24.1 days) at 20°C. The incubation time was significantly less when the larvae were fed with 1.1×10^5 IBs/larva compared to 1.1×10^4 IBs/larva.

With regard to storage time, mortality of larvae varied from 68.7 to 75.6 per cent in different periods (Fig. 1a) when all the three temperatures *viz.*, 28, 4 and 0°C, were considered together. The differences observed between mortality of larvae fed with virus stored for different periods were not statistically significant. In the same way, when all the periods were considered together, the mean per cent mortality was 72.1, 73.1 and 72.9 at 28, 4 and 0°C respectively and the differences observed between the mortality of larvae fed with virus stored at different temperatures were not statistically significant. Similarly, in the case of time taken for kill, there was no significant variation between different periods and temperatures. It varied from 13.5 to 14.4 days in different periods and from 13.9 to 14.1 days in different temperatures (Fig. 1b).

It is evident that the mortality of GV - infected shoort borer larvae was high and rapid when the post-treatment temperature ranged from 30 to 35°C. Influence of incubation temperature on the rate of GV disease is known in *Pieris brassicae* Linn. (David *et al.*, 1971) and *Plodia interpunctella* (Hb) (Hunter and Hartsell, 1971). In general, the optimum temperature range of the host also favours the disease. The shoot borer is essentially a pest of hot summer months during which time the atmospheric temperature easily crosses 30-35°C and so the virus can cause rapid mortality of the larvae. This is of great practical importance, when quick protection of crops is desired (Falcon, 1971). At low temperatures, food consumption and metabolic activity of the borer may be comparatively less and so the time taken for kill would have been prolonged.

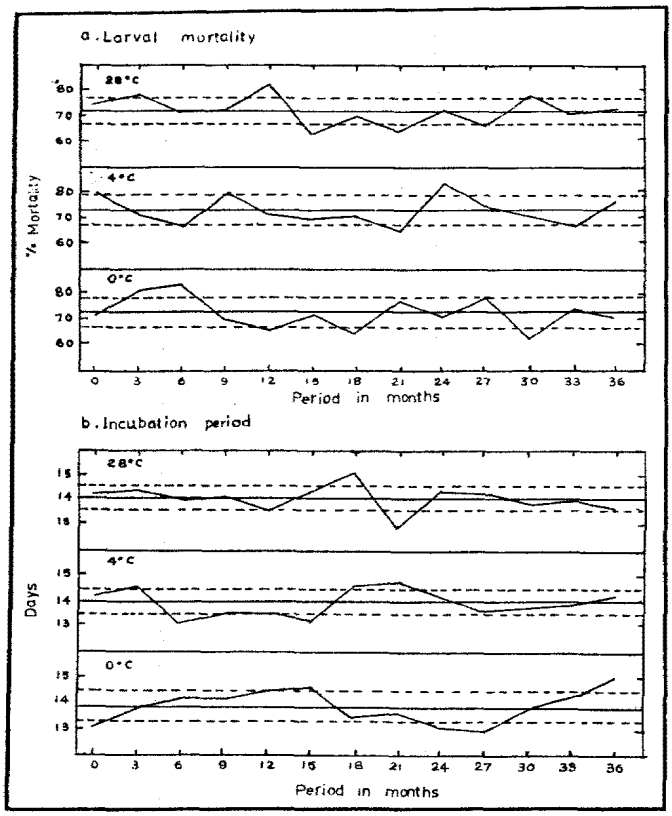
TABLE -1 : Effects of different rearing temperatures on shoot borer virus infection.

Temperature (°C)	Mortality				Time taken for kill (days)			
	10^5 IBs/ larva	10^4 IBs/ larva	Control @	Mean	10^5 IBs/ larva	10^4 IBs/ larva	Control @	Mean
20	48.2 (44.3)	34.5 (35.2)	0.0 (0.57)	41.4 (39.8)	22.7	25.5	0	24.1
25	71.1 (57.5)	61.3 (52.0)	0.0 (0.57)	66.2 (54.8)	15.7	18.9	0	17.3
30	82.0 (68.1)	83.8 (72.6)	0.0 (0.57)	82.9 (70.3)	10.3	11.3	0	10.8
35	85.1 (70.3)	77.1 (65.0)	0.0 (0.57)	81.7 (67.6)	12.0	12.9	0	12.5
Mean	71.6 (60.0)	64.2 (56.2)	0.0 (0.57)		15.2	17.2	0	

Figures in parentheses are arc sine / percentage values.
@ control values were excluded for statistical analysis.

	Mortality		Incubation period	
	S.E.	C.D. (P=0.05)	S.E.	C.D. (P=0.05)
Temperature	5.1**	15.0	0.6**	1.7
Dose	3.6 NS	--	0.4**	1.2
Temperature x Dose	7.2 NS	--	0.8 NS	--

** Significant at 1% level NS = not significant.



There was no reduction in the infectivity of the virus when it was stored for three years either at room or low temperatures. Earlier, several workers have found that many GVs retained their activity for 1 to 5 years (Vago *et al.*, 1961; Huger, 1963; Schmid, 1974). Though there was no difference in the infectivity of virus stored at 0, 4 and 28°C, storage at low temperature may be more advantageous and possibly prolong the period of infectivity as observed in GV infecting *Pieris rapae* (Linn.) (Liu and Liang, 1981).

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