

Efficacy of Extracts of Certain Host Plants as Adjuvants for Nuclear Polyhedrosis Virus of *Helicoverpa armigera* Hbn. and its Dust Formulation*

R.J.RABINDRA and S.JAYARAJ

Centre for Plant Protection Studies

Tamil Nadu Agricultural University, Coimbatore - 641 003

ABSTRACT

Laboratory studies revealed that 10% water extracts of sorghum and pearl millet grains and sunflower seeds (all at dough stage), cotton bolls and squares, and tomato fruits (both green and ripe) when added to nuclear polyhedrosis virus of *Helicoverpa armigera* (Hbn.) (HaNPV) significantly increased the mortality due to virus in second instar larvae of *H. armigera*. Probit analysis of time-mortality response showed that the LT_{50} s were also reduced in these treatments. Dust formulations of HaNPV containing extracts of sorghum grain, cotton seed kernel, chickpea flour and sunflower seed kernel were significantly more effective against *H. armigera* larvae than the virus formulation without any adjuvant. Sorbic acid and methyl parahydroxy benzoate which were used in the formulations were not responsible for the enhanced efficacy of the virus.

KEYWORDS : Nuclear polyhedrosis virus, dust formulations, adjuvants, *Helicoverpa armigera*

The nuclear polyhedrosis virus (HaNPV) has been found to be effective in the control of the gram pod borer *Helicoverpa* (= *Heliothis*) *armigera* Hbn. on several crops particularly the chickpea (*Cicer arietinum* L.) (Rabindra and Jayaraj, 1988a). Several adjuvants like crude sugar (Rabindra and Jayaraj, 1988b), crude larval extracts of certain lepidoptera including *H. armigera*, whole milk, whole egg homogenate, egg white and tender coconut milk (Rabindra and Jayaraj, 1988c) were reported to be effective in increasing the efficacy of the virus against *H. armigera* larvae. These adjuvants were also found to be effective in the field control of the pest (Rabindra *et al.*, 1989). Results on the evaluation of more such adjuvants for the NPV and the efficacy of some of them in the dust formulations of NPV against *H. armigera* larvae are reported in this paper.

MATERIALS AND METHODS

The HaNPV used in the study was propagated in fourth instar *H. armigera* larvae and the concentration of polyhedral occlusion bodies (POB) assessed with the help of a new improved double ruled Neubauer haemocytometer. Water extracts of the different substances (Table 1) were prepared by maceration in an all glass pestle and mortar and filtered through a muslin. In the case of sunflower seeds, the seed coat was removed before extraction. The cotton squares and bolls and tomato fruits were cut into small bits before extraction. Teepol was added at 0.1 per cent as a surfactant. Appropriate quantity of HaNPV was added to 100 ml samples of the extracts to get a concentration of 5×10^4 POB/ml. The leaf-dip bioassay method of Rabindra and Jayaraj (1988b) was adopted using second instar *H. armigera* larvae. After 24 h inoculation feeding time, the

* Part of the research project funded by U.S.D.A / ICAR under U.S. India Funds (PL 480) programme

Table 1. Efficacy of certain aqueous plant extracts as adjuvants for HaNPV against second instar larvae of *H.armigera*

Treatments @	% larval mortality*	Time Mortality Response - Probit Analysis				
		No. of insects	Chi ² (n-2)**	'b' slope	LT ₅₀ (h)	Fiducial Limits
Sorghum (dough stage) 10%	97.1 ^a	74	0.50	10.84	76.21	78.82 - 72.40
Sunflower seed kernel 10%	100.0 ^a	68	0.17	17.09	78.63	80.33 - 76.60
Crude sugar 20%	89.2 ^a	70	0.99	15.13	81.44	83.23 - 79.44
Pearl millet (dough stage) 10%	94.3 ^a	70	0.39	14.70	81.78	83.63 - 79.77
Cotton squares 10%	91.7 ^a	72	1.79	11.79	81.83	88.73 - 70.91
Cotton bolls 10%	97.2 ^a	72	0.61	13.90	80.25	82.18 - 77.97
Tomato (green) 10%	75.7 ^b	74	2.36	11.48	92.48	96.57 - 89.73
Tomato (ripe) 10%	88.2 ^{ab}	68	0.75	12.36	84.59	86.91 - 82.31
Control (NPV alone)	54.2 ^c	68	1.11	9.59	92.99	98.77 - 89.58

@ All treatments carried NPV at 5×10^4 POB/ml

* Means followed by similar letters are not different statistically ($P=0.05$) by DMRT

** All lines are a significantly good fit ($P < 0.05$)

larvae were removed to individual vials containing the semisynthetic diet (Shorey and Hale, 1965) and observed daily for mortality. Suitable controls without virus inoculation were maintained. Once the larvae started dying due to the treatment, observations on the mortality were made every 8 h.

Dust formulations of HaNPV were prepared with talc as the filler. Water extracts of sorghum seeds (dough stage), chickpea flour, cotton and sunflower seed kernels were incorporated individually at 10 per cent in the dust formulations to test their efficacy as adjuvants. Methyl parahydroxy benzoate at 0.2 per cent and sorbic acid at 0.1 per cent were added as mould inhibitors. The formulations were prepared by drying over calcium chloride in a desiccator. When the formulations had dried completely, they were passed through a 100-mesh sieve repeatedly to ensure homogeneous mixing.

Chickpea plants were raised in pots (30 cm dia) and at the preflowering stage, the shoots were dusted with the formulations by a simple polythene bag method. Exactly 100

mg of the dust formulation was placed inside a polythene bag (30 x 20 cm). Then three chickpea shoots, each carrying 6 compound leaves were introduced into the polythene bag with the basal ends of the shoots just protruding out of the bag. Air was blown to the full capacity of the bag, the mouth closed and vigorously tapped so as to ensure complete coverage of the shoots with the dust formulations. The quantity of 100 mg dust formulation was just sufficient to cover the shoots uniformly. Second instar larvae of *H. armigera* were released on treated shoots @ 10/shoot and allowed to feed for 24 h. The treatments were replicated adequately. After 24 h, the larvae were removed to individual vials containing semisynthetic diet and held at room temperature. Mortality was recorded on the seventh day. In another laboratory experiment, sorbic acid 0.1%, methyl parahydroxy benzoate 0.2% and sorbic acid 0.1% + methyl parahydroxy benzoate 0.2% were tested along with other adjuvants to find out their role in increasing the efficacy of the virus.

Table 2. Efficacy of adjuvants in HaNPV dust formulations in increasing the virus mortality in second instar larvae of *H. armigera*

Treatments*	Mean % mortality**
Cotton seed kernel 10% + Talc	89.7 ^a
Sorghum grain 10% + Talc	61.3 ^b
Chickpea flour 10% + Talc	96.4 ^a
Sunflower kernel 10% + Talc	60.0 ^b
Talc alone	46.7 ^c

* Dust formulations containing Ha NPV @ 10⁸ POB /g

** Means followed by similar letters are not different statistically (P=0.05) by DMRT

RESULTS AND DISCUSSION

Results of the bioassay revealed that 10 per cent water extracts of sorghum and pearl millet seeds, sunflower and cotton seed kernels, cotton squares and bolls and tomato fruits significantly increased the larval mortality due to NPV and were as effective as 20 per cent crude sugar (Table 1). Extract of

Table 3. Effect of sorbic acid and methyl parahydroxy benzoate in HaNPV formulations on mortality due to NPV in second instar larvae of *H. armigera*

Treatments*	% mortality**
Sorbic acid 0.1%	36.4 ^d
Methyl parahydroxy benzoate (MPHB) 0.2%	57.1 ^{bc}
Sorbic acid 0.1% + MPHB 0.2%	52.8 ^{bc}
Cotton seed kernel extract 10%	72.6 ^{ab}
Chickpea flour extract 10%	82.5 ^a
Sunflower seed kernel extract 10%	66.0 ^b
Sorghum grain extract 10%	62.7 ^{bc}
Talc alone	46.2 ^{cd}

* Dust formulations containing HaNPV @ 10⁸ POB/g

** Means followed by similar letters are not different statistically (P=0.05) by DMRT

green fruits of tomatoes was however inferior to other adjuvants. Probit analysis of time-mortality responses showed that all the adjuvants + NPV treatments recorded shorter LT₅₀ values than NPV alone, the minimum being recorded by the sorghum seed extract. All these adjuvants should have acted as phagostimulants since they are the natural host plants for *H. armigera*.

Cotton seed flour as the most preferred adjuvant for *Heliothis virescens* (F). has been reported earlier by Bell and Kanavel (1978) and 'Coax' a commercial adjuvant (Traders Oil Mill Co., Texas, USA) consisting mainly (62.3%) of cotton seed flour improved the field efficacy of NPV against *H. virescens* on cotton (Bell and Romine, 1980). Cotton seed oil cake and chickpea flour extracts among others were reported earlier to increase the efficacy of NPV against *H. armigera* larvae by Rabindra and Jayaraj (1988b). The performance of these adjuvants can be enhanced by the addition of whitening agents like Ranipal or Robin blue (commercial whitening agents) which were reported to increase the persistence of *H. armigera* NPV (Rabindra and Jayaraj, 1988c).

Evaluation of the dust formulations with adjuvants showed that the formulations containing 10 per cent extracts of sorghum, cotton seed kernel, chickpea flour and sunflower kernel significantly increased the efficacy of the NPV dust formulations (Table 2). Neither sorbic acid/methyl parahydroxy benzoate nor a mixture of the two had any role in enhancing the efficacy of the virus (Table 3). There are no earlier reports on the evaluation of adjuvants for the dust formulations of NPV of *H. armigera*. Corn meal extract was reported to enhance the efficacy of a dust formulation of NPV against *Heliothis zea* Boddie (Montoya *et al.*, 1966).

The results of the present studies have clearly indicated the scope for the use of phagostimulant adjuvants for increasing the efficacy of NPV and its formulation in the control of *H. armigera*. While recommending such ad-

juvants however, the cost factor should be borne in mind. The consistency and concentration should be such that it is amenable for use with the appropriate plant protection equipment.

REFERENCES

- BELL, M.R. and KANAVAL, R.F. 1978. Tobacco Budworm: Development of a spray adjuvant to increase effectiveness of a Nuclear polyhedrosis virus. *J. Econ. Entomol.*, 71, 350-352.
- BELL, M.R. and ROMINE, C.L. 1980. Tobacco Budworm: Field evaluation of microbial control in cotton using *Bacillus thuringiensis* and a Nuclear polyhedrosis virus with a feeding adjuvant. *J. Econ. Entomol.*, 73, 427-430.
- MONTOYA, E.L., IGNOFFO, C.M. and MCGARR, R.L. 1966. A feeding stimulant to increase the effectiveness of and field test with a nuclear polyhedrosis virus of *Heliothis*. *J. Invertebr. Pathol.*, 8, 320-324.
- RABINDRA, R.J. and JAYARAJ, S. 1988a. Efficacy of nuclear polyhedrosis virus with adjuvants as high volume and ultralow volume applications against *Heliothis armigera* Hbn. on chickpea. *Trop. Pest. Mgmt.*, 34, 441-444.
- RABINDRA, R.J. and JAYARAJ, S. 1988b. Evaluation of certain adjuvants for Nuclear polyhedrosis virus (NPV) of *Heliothis armigera* (Hbn.) on chickpea. *Indian J. Experimental Biol.*, 26, 60-62.
- RABINDRA, R.J. and JAYARAJ, S. 1988c. Larval extract and other adjuvants for increased efficacy of nuclear polyhedrosis virus against *Heliothis armigera* larvae. *J. Biol. Control*, 2, 102-105.
- RABINDRA, R.J., SATHIAH, N., MUTHIAH, C. and JAYARAJ, S. 1989. Controlled droplet application of Nuclear polyhedrosis virus with adjuvants and UV protectants for the control of *Heliothis armigera* Hbn. on chickpea. *J. Biol. Control*, 3, 37-39.
- SHOREY, H.H. and HALE, H.L. 1965. Mass rearing of the larvae of nine noctuid species on a simple artificial medium. *J. Econ. Entomol.*, 58, 522.