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Influence of dietary vegetable oils on the tobacco cutworm, *Spodoptera litura* (Fabricius) and its nuclear polyhedrosis virus production

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ABSTRACT: **Investigations were carried out on the impact of certain vegetable oils, *viz.*, castor, coconut, cottonseed, groundnut, mustard, safflower, sesamum, soybean, sunflower and linseed added at the rate of 0.6 per cent in the semi-synthetic diet on the tobacco caterpillar, *Spodoptera litura* (Fabricius). The results revealed that the vegetable oils (0.6 %) did not have any effect on larval and pupal duration, fecundity and egg period of the insect. However, the yield of NPV was significantly increased by the addition of mustard and coconut oils to the standard diet. When early fifth instar larvae were inoculated with a dose of 1966.18 POB/mm² of diet surface, diet incorporated with 0.6 per cent coconut oil yielded the highest number of 8.7×10^9 POB/larva which was significantly higher than that obtained from larvae fed on standard diet. At the higher inoculum level of 3932.96 POB/mm², mustard oil recorded the highest virus yield of 7.4×10^9 POB per larva. But incorporation of mustard oil in the diet reduced the fecundity in *S. litura* moths significantly and hence mustard oil cannot be used in host culture. Also, by incurring an additional cost of Rs. 0.30/- by adding 0.6 per cent of coconut oil, to the diet, an increase in yield upto 12.9×10^{11} POB/200 larvae (4.3 fold increase) was obtained. The results indicate that the insects can be mass produced in the standard diet, and for virus production mustard and coconut oils can be used as dietary adjuvant at 0.6 per cent to the standard diet.**

KEY WORDS: NPV, semi-synthetic diet, *Spodoptera litura*, vegetable oils

The tobacco caterpillar, *Spodoptera litura* (Fabricius) is a polyphagous pest attacking many crop plants. Chemical control of this pest has often failed as this pest is reported to have developed resistance to many of the commercially available insecticides (Ramakrishnan *et al.*, 1984; Jayaraj and Santharam, 1985; Mehrotra, 1989). Successful control of *S. litura* with the nuclear polyhedrosis virus (NPV) has been reported on crops like tobacco (Santharam and Balasubramanian, 1980; Ramakrishnan *et al.*, 1981), banana (Santharam *et al.*, 1978), cauliflower (Chaudhari and Ramakrishnan, 1980), and cotton (Jayaraj *et al.*, 1980). The virus is mass-produced *in vivo* in *S. litura* larvae. Increased productivity of the host larvae and the virus is a key factor in commercial scale production of NPV. Dietary oils have been found to improve host larval production in many lepidopterous insects (Farror *et al.*, 1992; Sathiah, 2001). In the present study, the effect of incorporation of vegetable oils in the diet on the productivity of both *S. litura* and its NPV was studied.

MATERIALS AND METHODS

Effect of oils on the growth and development of *S. litura*

A healthy laboratory culture of *S. litura* maintained in the Department of Agricultural Entomology was used for the experiments. Larvae of *S. litura* were grown in the standard chick pea-based diet until they reached the second instar, and then transferred to diets incorporated with vegetable oils of castor, coconut, cotton seed, groundnut, mustard, safflower, sesamum, soybean, sunflower and linseed @ 0.6 per cent and reared individually in glass vials. Each treatment was replicated thrice with 25 larvae per replication. A similar number of larvae in three replicates reared on standard diet were maintained for comparison. The larvae were allowed to pupate in the vial itself. The pupae were weighed individually at the rate of ten pupae per replication. The pupae were harvested after hardening for 5 days, washed in running water and rolled over filter paper to remove the moisture. They were then placed in plastic containers with filter paper at the bottom and kept in adult emergence cages (45x45x45cm).

Healthy adults after emergence were sexed and five pairs of adults were released into cylindrical buckets (7 litre) (25x15cm) for mating and oviposition. A bouquet of fresh nerium (*Nerium odoratum* L.) shoots arranged in a glass vial containing water, plugged with cotton was kept in each of the container to encourage oviposition (Santharam, 1985). The adults were fed with sugar solution (10%) fortified with ABDEC® solution (0.3ml/100ml sugar solution) provided in each bucket. The top of the bucket was covered with sterile muslin cloth, which also served as an oviposition substrate. The larval and pupal duration, pupal weight, number of egg masses per pair, fecundity and egg period was recorded.

Effect of oils on NPV production

Early fifth instar larvae were allowed to feed on a semisynthetic diet surface contaminated with 10µl suspension of NPV containing $5x10^6$ POB. The larvae were released 15 minutes after contamination of diet. The vials with the laevae were later incubated at 25 ± 0.5°C. After about 5-6 days, virosed larvae upon death were collected, rinsed and homogenized in sterile distilled water and passed through a double layer of muslin. The filtrate was centrifuged at 500 rpm for one minute and the pellet was discarded. The supernatant was centrifuged at 5000 rpm for about 15 minutes to pellet the virus. The virus pellet was resuspended in distilled water and the concentration of POB was assessed.

Larvae of *S. litura* were reared on semi-synthetic diets containing different oils until they moulted to the fifth instar. At the early fifth instar stage, the larvae were starved for 4h. Diet containing the different oils at 0.6 per cent but lacking formalin were poured into sterile glass vials (5ml) and allowed to cool under a hood for about one hour. Ten microlitres of the virus suspensions ($1x10^6$ and $5x10^5$ POB) were applied on the diet surface uniformly. This resulted in a concentration of 3932.36 and 1966.18 POB/mm^2, respectively of the diet surface. After about 15 minutes, one early fifth instar larva was released into each vial and plugged with sterile cotton. Each treatment had 30 larvae in three replications and incubated at 25 ± 0.5°C in an incubator. Virosed larvae upon death were harvested individually and weighed. The virus was extracted and processed as described earlier and the yield of POB in each cadaver was assessed.

The data were subjected to analysis of variance after suitable transformation, wherever necessary, and means separated by Duncan's new multiple range tests.

RESULTS AND DISCUSSION

Influence of vegetable oils on host culture

The addition of oils @ 0.6 per cent did not increase the larval and pupal duration, pupal weight, fecundity and egg period (Table 1). Decreased pupal weight was noticed in the diets containing castor, sesamum, cottonseed, sunflower, safflower, linseed, soybean, mustard and groundnut oils. The number of egg masses laid by a single female was higher in coconut and sesamum oil-incorporated diet than in

Table 1. Effect of dietary incorporation of vegetable oils on the laboratory production of *S. litura*

Semi-synthetic diet + oils (0.6 %)	Larval period (days)	Pupal duration (days)		Pupal weight (mg)	No.of egg masses/ pair	Fecundity (eggs/ female)	Egg period* (days)
		Male*	Female				
Standard	15.5a	9.9	9.0abc	394a	3.5bc	878.2ab	3.5
Castor	15.8ab	10.4	9.2bc	371b	3.6b	1057.1a	3.5
Groundnut	16.8cd	10.7	8.4a	349cd	3.4bc	791.6bc	3.5
Sesame	16.2abc	10.2	9.4bc	333d	4.0ab	817.4bc	3.6
Cotton	16.0abc	10.8	8.9abc	371b	2.9c	602.7de	3.5
Mustard	15.9abc	11.1	9.5bc	366bc	2.9c	545.6e	3.4
Soybean	17.2d	10.8	9.6bc	366bc	2.8c	670.4cde	3.6
Coconut	16.4a-d	9.8	8.8abc	382ab	4.5a	893.9ab	3.5
Sunflower	16.7bcd	10.4	8.9abc	366bc	3.4bc	725.6bcd	3.7
Linseed	15.7ab	10.8	9.0abc	347cd	2.8c	615.2de	3.6
Safflower	15.6a	10.7	9.7c	332d	3.4bc	797.3bc	3.7

$ In a column, means followed by a common letter are not significantly different (P=0.05) by DMRT
*Differences between the means not significant

other diets. Fecundity per female, however, was not increased due to the addition of oils. No significant difference in the egg period was noticed by the addition of oils to the diet. Oils of corn, cottonseed, linseed, olive, peanut, rapeseed, safflower and sunflower were substituted for essential fatty acids and reported to improve the biological characters of lepidopteron (Farror *et al.*, 1992). Improved growth potential of *Corcyra cephalonica* (Stainton) moths was observed by Senrayan *et al.* (1992) by the addition of lipid source to the cereal based diet.

Influence of vegetable oils on virus production

In the experiment on virus production, mortality caused by *Sl*NPV in larvae reared on diets containing oils, *viz.*, castor, soybean, mustard, groundnut, coconut, sunflower, gingelly, cottonseed and safflower was similar to the mortality observed in standard diet, while linseed oil was found to be inhibitory (Table 2). The cadaver weight was significantly increased by the addition of oils of groundnut, soybean, coconut, sunflower and safflower (all at 0.6%) to the standard diet. This may be due to the phagostimulatory effect of these oils (Rabindra and Jayaraj, 1988). The POB yield per larva, per 100 inoculated larvae or per gram of body weight was however higher only in the diet incorporated with coconut oil followed by mustard oil. At the higher inoculation dose of 3932.36 POB/ mm^2, mustard oil recorded the highest yield of 7.4×10^9 POB/larva, while, at the lower dose of 1196.18 POB/mm^2, coconut oil recorded the highest yield of 8.7×10^9 POB/larva. Increased yield of *Ha*NPV by addition of sunflower, soybean, groundnut, sesamum and coconut oils (8000 ppm) to the semi-synthetic diet was reported by Sathiah (2001). It should be noted that mustard oil reduced the fecundity significantly (37.6 %) and this would reduce the overall productivity of the virus.

Table 2. Effect of inoculum dose and oils on the production of *Sl*NPV

Dose (POB/mm²)	Treatment (oils @ 0.6%)	Larval mortality (%)	Cadaver weight (mg)	Yield/larva $\times 10^9$ POB	Yield/100 inoculated larvae $\times 10^{11}$ POB	Yield/gram of body weight $\times 10^8$ POB
3932.36	Standard	100.0a	432cd	2.4bcd	2.4bc	5.5bc
	Castor	100.0a	531b	3.1b	3.1b	5.9b
	Sesame	96.7a	403d	2.6bc	2.5bc	6.1b
	Soya	100.0a	553ab	1.1f	1.1e	2.0f
	Mustard	100.0a	528b	7.4a	7.4a	14.1a
	Cotton	96.7a	516b	1.7e	1.6d	3.2e
	Groundnut	100.0a	584a	2.3bcd	2.3c	3.9de
	Coconut	100.0a	530b	2.1cde	2.1cd	4.0de
	Sunflower	100.0a	555ab	2.6bc	2.6bc	4.7cd
	Safflower	96.7a	560ab	2.9b	2.8bc	4.9bcd
	Linseed	93.3a	459c	1.8de	1.7d	4.1de
	Mean	98.5A	514B	2.7A	2.7A	5.3A
1966.18	Standard	96.7ab	508c	1.9de	1.9d	3.8cde
	Castor	96.7ab	583ab	2.0cde	2.0d	3.5cde
	Sesame	96.7ab	418d	1.5f	1.4e	3.6cde
	Soya	100.0a	578ab	2.6c	2.8c	4.8c
	Mustard	100.0a	538bc	4.4b	4.4b	8.2b
	Cotton	93.3ab	538bc	1.5ef	1.4e	2.9e
	Groundnut	96.7ab	612a	2.3cd	2.2cd	3.8cde
	Coconut	96.7ab	616a	8.7a	8.4a	14.0a
	Sunflower	100.0a	572ab	2.4cd	2.8cd	3.9cde
	Safflower	96.7ab	571ab	1.9cf	1.9d	3.5de
	Linseed	90.0b	541bc	2.3cd	2.0d	4.2cd
	Mean	96.7B	552B	2.9A	2.8A	5.1A

In a column, means of different oils for individual virus dosage followed by same letters in lower case are not significantly different (P=0.05) by DMRT

In a column means of different dose of inoculum effects irrespective of the oils followed by same capital letters are not significantly different (P=0.05) by DMRT

The findings suggest that the standard diet without the incorporation of any of the oils may be used for growing *S. litura* until they reach the early fifth instar stage and for virus production, the larvae can be transferred to diet incorporated with coconut oil (0.6%). Use of coconut oil (0.6%) in the diet gave 8.4×10^{11} POB/100 inoculated larvae giving the highest productivity of 14.0×10^8 POB/g of cadaver. Comparing the cost: benefit, by incurring an additional cost of Rs. 0.30 (Cost of coconut oil is taken as Rs50/litre) by adding coconut oil (0.6%) to standard diet for a batch of 200 larvae, an increased yield of 12.9×10^{11}POB/200 larvae was obtained. This four-fold increase in the productivity of virus when compared to the standard diet will contribute significantly to economic production of *Sl*NPV. This improved production technique coupled with the use of a highly virulent strain of

*Sl*NPV will contribute to effective control of the pest at reasonable cost.

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