It was later refitted and renamed *Foudroyant* and subsequently sold[20].

Teak is one of the best timbers used for various purposes. The shipwrights industry also used teak in the shipbuilding industry for its superior qualities. Anatomical analysis indicates that *L. microcarpa* grown in the Western Ghats of India was used for building boats and ships. Indian as well as Portuguese shipwrights used both *L. microcarpa* and *T. grandis* species in the construction of ships. The timber off St George's Reef corroborates this.

The Portuguese understood the qualities of teak and used it extensively in shipbuilding industry and for various other purposes such as furniture, and house and church constructions. The East India Company also built several ships both on the east and west coast of India for the Royal Navy and overseas trade and commerce. Use of Indian teak for construction of ships by the Portuguese, British and Indian boatbuilders has been proved from the shipwreck findings in Indian and foreign waters. The use of Indian teak by Portuguese for making anchors is confirmed from the finding of the wooden stock of iron anchor at Aguada Bay. The use of teak for other purposes in the shipbuilding industry and its provenance could be known from the findings of shipwreck explorations in India in the coming years.

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Combined effect of gamma radiation and Azadirachtin on the growth and development of *Spodoptera litura* (Fabricius)

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Spodoptera litura **is an economically serious and polyphagous pest in the Indian subcontinent. This pest is reported to attack a wide range of food plants belonging to diverse origin 112 cultivated food plants belonging to 44 families all over the world. In order to explore an eco-friendly strategy that could be coupled up with this radio genetic method, i.e., F-1 sterility technique, the present attempt was made to study the effect of a safe chemical (azadirachtin) for the effective management of lepidoteran pest, *S. litura*. The studies show that insecticidal toxicity can be altered if the insects are exposed to radiation prior to insecticidal treatment.**

Keywords: Azadirachtin, gamma radiation, *Spodoptera litura*.

THE efforts of man in combating agricultural pests by the use of toxic chemicals are frequently thwarted by the development of insecticide resistance by the insects, and/or by the establishment of new pests when predators and parasites

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are killed by the insecticides. Extensive use of chemicals for pest control has raised tremendous concern regarding environmental hazards too. In view of the limitations associated with chemical control, pest managers have been forced to seek some alternatives which must be ecologically compatible. One such ecosafe control method is radiation-based sterile insect technique (SIT) (F-1 sterility technique) for the management of lepidopteran pests. This technique has been used since 1968 as an autocidal method to protect 0.4 million hectares of cotton crop in the San Joaquin Valley, California, USA, from the pink bollworm[1]. In India, the bioefficacy of sterilizing radiation was assessed in order to understand the feasibility of SIT in the potato tuber moth, *Phthorimaea operculella*[2] and the tobacco caterpillar *Spodoptera litura*[3,4].

There are reports on the combined effects of gamma radiations with other treatments like infrared[5], microwaves[6] and insecticides[7–10]. Combination of these treatments is likely to be economical and complementary in their effectiveness against insect pests. The studies on the combined effects of ionizing radiations and insecticides conducted so far, show that insecticidal toxicity can be altered if the insects are exposed to radiation prior to insecticidal treatment.

In this communication we report the combined effect of the radiogenetic method, i.e. F-1 sterility technique and a biopesticide, azadirachtin ($\cong 0.03\%$) for the effective management of the lepidopteran pest, *S. litura* (Fabricius) (Lepidoptera: Noctuidae), the common cutworm, an economically serious and polyphagous pest in the Indian subcontinent.

S. litura was reared in the laboratory under optimum environmental conditions. The moths were paired in cages made of perspex and nylon. Larvae were fed on castor (*Ricinus communis* L.) leaves and the same leaves were introduced in the cage to serve as ovipositional arena. Individual pairing of moths was done in perspex jars (20 cm diameter × 30 cm height), whereas in group pairing, cages of different sizes were used. Cotton swabs saturated with 15–20% (w/v) honey solution were placed in small plastic containers, on which the adults were fed. The eggs laid were collected and treated with 0.2% sodium hypochlorite solution and distilled water. Newly emerged (0–24 h old) male adult moths were subjected to the required doses of gamma irradiation using a ^{60}Co source at the rate of 7.26 KGy (1 rad = 100^{-1} Gy) per hour in the gamma cell at the Institute of Nuclear Medicine and Allied Sciences (INMAS), Delhi. Two radiation doses, i.e. 100 and 130 Gy were used. An eco-friendly, neem-based insecticide containing $\cong 0.03\%$ azadirachtin was used to study its bioefficacy on *S. litura*, which was already subjected to radiation. Insecticide containing $\cong 0.03\%$ azadirachtin was treated as 100% stock solution. Four desired concentrations of 0.25, 0.50, 0.75 and 1.00% of the 100% stock solution were made in distilled water. These were applied on leaves by dipping once in one concentration followed by air-drying. Thereafter these treated leaves were fed to the F-1 larvae (L-3 stage) for two days, followed by their feeding on untreated diet (castor leaves). Distilled water was applied similarly to act as control. The data collected in the experimental evaluations were subjected to appropriate biostatistics to assess the significance of results[11,12].

In the first phase, for each concentration 45 larvae of irradiated and non-irradiated P-1 males were reared and the effect on the growth and development in response to radiation (given to moths) and azadirachtin (applied on leaf and fed to larvae) was observed. Castor leaves treated with different concentrations of azadirachtin were fed to larvae at L-3 stage for 48 h followed by provisioning of normal diet onwards. The growth index of Raulston[13] was used here, i.e. Growth index = percentage adult formation/developmental period. For each concentration, three replicates were taken. Each replicate contained a group of 15 larvae.

It was observed that azadirachtin delayed the development of F-1 larvae of *S. litura* at both the radiation doses. For instance, at 100 Gy the developmental period from L-3 to adult formation was 15.45 days at 0% azadirachtin, whereas it was further prolonged to 17.66 and 21 days at 0.75 and 1% azadirachtin respectively. Similarly, azadirachtin delayed the development of F-1 larvae at 130 Gy (Table 1).

There are reports on the dose-dependent effects of azadirachtin in the inhibition of post-embryonic development of *S. litura* and for other insect species[14–19]. Inhibition of larval development of *S. litura* might have resulted, at least in part, from a reduced physiological age brought about by reduced food intake and less weight gain. This might be due to disruption of the neuroendocrine regulation of moulting by azadirachtin[15,17,18].

In the present study, azadirachtin treatment (1%, equivalent to 3 ppm of azadirachtin), when given to L-3 for 48 h caused 87.3% reduction in adult emergence, which was further reduced to 92.5 and 94.2% for F-1 insects from 100 and 130 Gy-treated parents respectively. It showed an enhanced impact of radiation and azadirachtin application on insect ecdysis, growth and adult eclosion. The adult formation of F-1 progeny of irradiated male moth was adversely affected due to azadirachtin treatment, which was significantly evident at 0.5% concentration onwards.

The F-1 adult formation was also found to be adversely affected when azadirachtin treatment was combined with radiation treatment. For instance, F-1 adult formation was 88.88% in control (0 Gy and 0% azadirachtin) and it was decreased to 53.33% at 100 Gy and 48.88% at 130 Gy in case of 0% azadirachtin. In case of 1% azadirachtin treatment, per cent adult formation was 11.26, 6.66 and 5.15 at 0, 100 and 130 Gy respectively (Table 2).

Further the growth index was computed from ratio of per cent adult formation at developmental period. At each radiation dose, the growth index was decreased in a dose-dependent fashion with respect to azadirachtin treatment. For instance, at 130 Gy the growth index value was 2.90 at 0% azadirachtin and decreased to 2.09, 1.63, 0.46 and 0.37 at 0.25, 0.50, 0.75 and 1% azadirachtin respectively (Figure 1).

Table 1. Effect of feeding azadirachtin-treated diet on developmental period of F-1 progeny of irradiated male moths of *S. litura* (Fabr.)

Gamma dose (Gy) to male parent	Concentration of azadirachtin (%)				
	0.00	0.25	0.50	0.75	1.00
0	$15.52^{Aa} \pm 0.07$	$15.25^{Aa} \pm 0.07$	$15.86^{Ab} \pm 0.07$	$17.50^{Ac} \pm 0.42$	$18.60^{Ac} \pm 0.40$
100	$15.45^{Aa} \pm 0.10$	$16.50^{Bb} \pm 0.15$	$16.85^{Bb} \pm 0.34$	$17.66^{Ab} \pm 0.33$	$21.00^{Bc} \pm 0.00$
130	$16.68^{Ba} \pm 0.15$	$17.00^{Cab} \pm 0.15$	$16.75^{Bab} \pm 0.17$	$17.50^{Ab} \pm 0.28$	$21.00^{Bc} \pm 4.00$

Mean (± SE) followed by same small letters within a row, and means (± SE) followed by same capital letters within a column are not significantly different at $P = 0.05$ level (Duncan's multiple range test).

Table 2. Effect of feeding azadirachtin-treated diet on adult formation of F-1 progeny of irradiated male moths of *S. litura* (Fabr.)

Gamma dose (Gy) to male parent	Concentration of azadirachtin (%)				
	0.00	0.25	0.50	0.75	1.00
0	$88.88^{Aa} \pm 2.22$	$70.47^{Ab} \pm 1.98$	$52.37^{Ac} \pm 3.06$	$15.17^{Ad} \pm 1.85$	$11.26^{Ad} \pm 2.06$
100	$53.33^{Ba} \pm 3.85$	$35.95^{Bb} \pm 0.80$	$23.09^{Bc} \pm 2.37$	$9.78^{Bd} \pm 0.61$	$6.66^{Be} \pm 0.00$
130	$48.88^{Ba} \pm 2.22$	$35.54^{Bb} \pm 3.19$	$21.84^{Bc} \pm 2.43$	$8.14^{Bd} \pm 1.48$	$5.15^{Bd} \pm 2.60$

Mean (± SE) followed by same small letters within a row, and means (± SE) followed by same capital letters within a column are not significantly different at $P = 0.05$ level (Duncan's multiple range test).

Table 3. Effect of feeding azadirachtin-treated diet on sex ratio in F-1 progeny of irradiated male moths of *S. litura* (Fabr.)

Gamma dose (Gy) to male parent	Sex	Concentration of azadirachtin (%)				
		0.00	0.25	0.50	0.75	1.00
0	M	45	41.93	47.82	71.42	60
	F	55	58.06	52.17	28.57	40
100	M	40.90	50	57.86	66.6	100
	F	59.09	50	42.13	33.3	0
130	M	58.33	56.25	50	75	100
	F	44.46	43.75	50	25	0

M, Male, F, Female.

Tanzubil and McCaffery[20] reported that the larvae of *S. exempta* emerging out of treated (with azadirachtin) eggs were considerably less viable, probably as a consequence of consuming azadirachtin, with the egg shell eaten after hatching. However, delayed development and reduced growth index were noticed in the case of *S. litura* in the present study when 3rd instar larvae were fed on azadirachtin-treated diet, but notable adverse effect on their viability was not observed by Tanzubil and McCaffery.

Similarly, in the second phase of the experiment, the reproductive behaviour in response to radiation (given to moths) and azadirachtin (fed to larvae) was observed. For this experiment, F-1 adult males formed at 130 Gy as a result of the above experiment (i.e. by feeding azadirachtin-treated diet to F-1 larvae) were crossed with normal females. Two different concentrations, viz. 0.25 and 0.50% of azadirachtin were fed to F-1 larvae through diet. Four different cages were set up. (i) Normal males were crossed with normal females. (ii) F-1 adult males (130 Gy) fed with 0% azadirachtin diet (control) were crossed with normal females. (iii) F-1 adult males (130 Gy) fed with 0.25% azadirachtin were crossed with normal females. (iv) F-1 adult males (130 Gy) fed with 0.50% azadirachtin were crossed with normal females.

The results show that the sex ratio was almost 1 : 1 at control and changed towards male at 130 Gy at 0% azadirachtin, whereas within each radiation treatment azadirachtin concentration of 0.75% and above evidently increased males in the F-1 adults formed. For instance, at 0.75% azadirachtin the sex ratio was 66.6% males and 33.33% females at 100 Gy and 75% males and 25% females at 130 Gy, whereas 100% males were formed at 1% azadirachtin concentration at both the doses (Table 3).

Azadirachtin treatment was evaluated on the mating %, oviposition and fertility of F-1 male moths at 130 Gy when crossed with normal female along with control (0 Gy and 0% azadirachtin) treatment having normal parents. Mating percentage was 90.1 at control, 73.33 at 0% azadirachtin and decreased to 46.66 at 0.5% azadirachtin in case of 130 Gy treatment. Similarly, the eggs oviposited were decreased significantly when azadirachtin concentration was high. For instance, eggs laid were reduced by 13.9, 21.3 and 30.8% at 0, 0.25 and 0.50%, azadirachtin respectively in case of 130 Gy treatment with respect to control treatment. Further, F-1 fertility was 82.7% at control and 23, 17.6 and 12% at 0, 0.25 and 0.50% azadirachtin in case of 130 Gy treatment (Table 4).

Azadirachtin-treated diet was found to show protracted growth and low percentage of adult formation, leading to

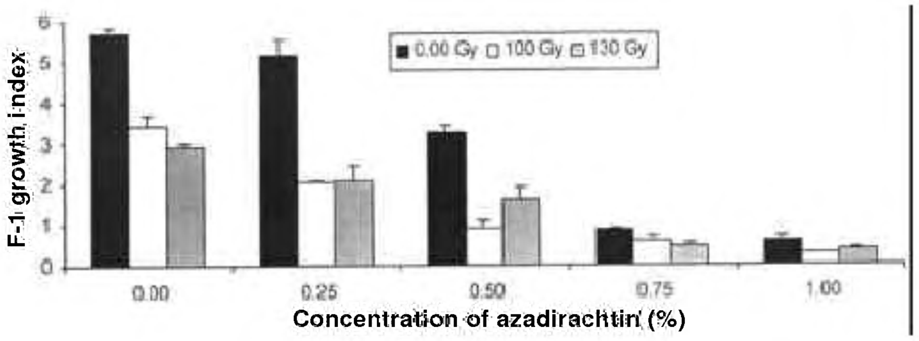

Figure 1. Effect of feeding azadirachtin-treated diet on growth index of F-1 progeny of irradiated male moth, *Spodoptera litura* (Fabr.).

Table 4. Effect of feeding azadirachtin-treated diet on mating percentage[I], eggs laid per mated female[II] and fertility[III] of F-1 male moths crossed with normal females of *S. litura* (Fabr.) treated as P-1 male moths with 130 Gy

Radiation to P-1 males and azadirachtin concentration	Mating (%)	Eggs laid per mated female	F-1 fertility (%)
0 Gy and 0% azadirachtin	90.1	1308^{a}	82.7^{a} ± 4.1
130 Gy and 0% azadirachtin	73.33	1125^{b} ± 66	23.00^{b} ± 0.60
130 Gy and 0.25% azadirachtin	66.66	1029^{bc} ± 51	17.60^{c} ± 0.53
130 Gy and 0.50% azadirachtin	46.66	904^{c} ± 45	12.00^{d} ± 0.45

[I] Mating was confirmed on third day by presence of spermatophore in bursa copulatrix and observations conducted in 15 pairs in each regime. [II] Eggs laid recorded for first three days of oviposition, $n = 5$. [III] Determined as egg hatchability, $n = 20$; each replicate comprising 50 eggs.
Means (± SE) followed by same letters within a column are not significantly different at $P = 0.05$ level (Duncan's multiple range test).

adversely affected growth index. It is presumed that the differentiation of adult organs during pupal development must be affected, which would influence reproduction in adults. Keeping this in mind, the effect of feeding of azadirachtin (0.03%)-treated diet was assessed on the mating percentage, eggs laid per mated female and fertility of F-1 males of *S. litura*, which were treated as P-1 male moths with 130 Gy. Mating percentage reflected the negative correlation with azadirachtin treatment to F-1 progeny. Similarly, oviposition was reduced due to azadirachtin treatment, and the reduction was more in case higher concentration, i.e. at 0.5% azadirachtin. Further, fertility of F-1 male moths (ascertained by the per cent egg hatch of eggs laid by female mated to F-1 males) showed a marked reduction with an increase in azadirachtin concentration fed to F-1 larvae.

Various neem products exert a dose-dependent influence on the reproduction of female insects[21], whereas the literature regarding the effect of neem on male reproduction is scanty. However, effects are also apparent in males as subnormal mating behaviour in locusts[22]. In an *in vitro* study of spermiogenesis in diapausing *Mamestra brassicae* males, 3 ppm azadirachtin caused spermatocysts to degenerate, even in the presence of 20-hydrozyecdysone, suggesting a direct effect on the testicular membrane, rendering the tissue incapable of developing spermatocysts[23].

Fecundity of female mated to male treated with 0.125 µg per insect was severely reduced[24]. Similar response was observed in case of *S. litura*, when normal female was mated with F-1 male (130 Gy) fed on azadirachtin-treated diet. Fecundity was reduced by 21.3% at 0.25% azadirachtin and 30.8% at 0.5% azadirachtin, whereas fertility was reduced by 78.7% at 0.25% azadirachtin and 85.4% at 0.5% azadirachtin. These reductions in fecundity and fertility of azadirachtin-treated males of *S. litura* were in addition to the effect induced by substerilizing doses alone. Combined effects of gamma radiations with other treatments, like infrared, microwave and insecticides have been reported to be more effective and complementary in certain cases, instead of using control tactics alone against insect pests. The studies indicate that insecticidal toxicity can be altered to varying degree if the insects are exposed to radiation along with insecticidal treatment[5,7–10].

A combination consisting of 0.1% (w/v) reserpine incorporated in the diet plus treatment of the resulting pupae with 75 Gy gamma radiation induced complete sterility in females. However, fecundity and fertility of normal females mated with treated males were reduced by 24 and 49% respectively[25]. In the present findings, fecundity and fertility of normal *S. litura* females mated with F-1 males were reduced by 21.3 and 78.7% at 0.25% azadirachtin and reduced by 30.8 and 85.4% at 0.5% azadirachtin respectively, at 130 Gy. While studying the effect of neem seed kernel suspension (NSKS), on egg parasite of *S. litura*, Joshi *et al.*[26] reported that NSKS in water sprayed on eggs of *S. litura*, before and after parasitization, did not adversely affect the emergence of egg parasite, *Telonomus remus*. This indicated that a schedule of NSKS sprays can be integrated with *T. remus* for the control of the tobacco caterpillar *S. litura* in tobacco nurseries. Likewise, the feasibility of combining inherited sterility and other biocontrol strategies using different parasitoids for suppression of noctuid moths as serious pests, has been demonstrated by Mannion *et al.*[27].

Azadirachtin feeding synergistically enhanced the adverse effect on growth and reproduction, that reflect the combination of irradiation and azadirachtin treatment might be compatible for lepidopteran pest suppression; it could also increase the efficacy of F-1 sterility technique. This integration of control methods could be feasible in the field application of these techniques for the control of this pest. Further studies are required to combine this azadirachtin treatment with F1 sterility technique for pest management. Certainly this indicates that there exists a possibility to use azadirachtin in combination with F-1 sterility to enhance the bioefficacy of inherited sterility for the suppression of pest population of *S. litura*.

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