Bioactivity of Brown Algal Seaweed, *Turbinaria ornata* (Turner) J. Agardh in Combination with Pungam Oil against *Spodoptera litura* (Fab.)

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(Received: July 22, 2022; Accepted: September 3, 2022)

ABSTRACT

The combined action of the solvent extract of brown algal seaweed, *Turbinaria ornata* with Pungam oil at 3 and 5% at various concentrations viz., 2, 4, 6, 8, 10 and 20% along with standard check was investigated against *Spodoptera litura* for their effectiveness on larval mortality, pupation, pupal malformation, adult emergence and pupa to adult conversion ratio under controlled condition during 2021-22 in Department of Entomology, Faculty of Agriculture, Annamalai University. In comparison with Pungam oil at 3% alone, combination effect of *T. ornata* (20%)+Pungam oil 5% showed the maximum larval fatality (80.00%), reduction in pupation (20.00%), adult emergence (13.33%) and pupal to adult conversion ratio (1 : 0.66) compared to standard check. Pungam oil addition supplemented the toxicity effect of brown algal seaweed and demonstrated critical reduction in the population of test insects.

Key words: Brown algae, Turbinaria ornata, Pungam oil, Spodoptera litura, maximum larval fatality

INTRODUCTION

Spodoptera litura (Fab.) (Lepidoptera : Noctuidae), a serious polyphagous pest (Sun et al., 2019) has caused severe damage to crops and led to huge economic yield losses ranging from 26 to 100% (Datta et al., 2020). It grows throughout the year and mounts nearly 7 to 8 generations per year. The larvae of S. litura feed initially on plant leaves and latterly feed on almost every part of the plant (Kousar et al., 2020). Farmers have used various insecticides belonging to different groups such as organochlorines, organophosphates and synthetic pyrethroids indiscriminately which lead to resistant development, direct impact on non-target organisms (Keswani et al., 2022), etc. The searches for finding out suitable alternatives paved way for new and novel products from natural resources and among them marine algal seaweeds which harboring vast coastal lines with various biogenic molecules and bio-activity played a crucial role in plant production and protection in agriculture (Oktaviani et al., 2019). The brown algal seaweeds, Turbinaria ornata (Turner) J. Agardh is rich in secondary metabolites such as alkaloids, phenolic compounds, flavonoids (Fauziee et al., 2021), etc., which may accelerate or decelerate the metabolic processes in insects and also promote plant

growth and development. In this study, the combined effect of Pungam oil with *T. ornata* was tested against *S. litura* to find out per cent mortality, antifeedant and insect growth activity.

MATERIALS AND METHODS

The brown algal seaweed (*Turbinaria ornata*) was collected from the coastal region of Rameshwaram, Tamil Nadu, India using hand picking method. The collected seaweeds were washed with tap water to remove debris and swabbed with a blotting sheet to take away excess moisture. It was shade-dried for a fortnight for complete drying and stored in an airtight container (Kannan and Bharathkumar, 2016). The seaweeds were identified at CAS Marine biology, Annamalai University.

Egg masses of *Spodoptera litura* were collected from castor plants at Sivapuri village, Cuddalore district of Tamil Nadu. The fieldcollected egg masses were allowed to hatch at room temperature (RH 76 \pm 5%) and newly hatched neonates were allowed to feed on tender castor leaves (*Ricinus communis* L.). To enhance the pupation, surface-sterilized soil was provided and pupae were collected and kept inside the oviposition chamber for adult emergence. Honey solution (10%) was provided as feed for adult moths. After emergence, adult moths were allowed to mate and lay eggs on nerium leaves (petiole immersed in a conical flask containing water). The egg masses were surface sterilized with 0.02% sodium hypochlorite and then dried and kept in plastic trays. Tender castor leaves were given as feed for emerged neonates and reared. The homogenous population of third instar larva was used in laboratory experiments (Gowthish and Kannan, 2019).

The powdered *T. ornata* was separately weighed as 2, 4, 6, 8, 10 and 20 g and soaked in 100 ml of acetone and kept for 12 h incubation at room temperature. The filtered extract was stored and used in experiments. For enhancement, 3% Pungam oil was added to the respective concentrations along with 2.5 ml of teepol as a wetting agent for better effectiveness. A similar procedure was followed in the preparation of 5% Pungam oil where it was added to the solvent extracts of 2, 4, 6, 8, 10 and 20% concentrations of *T. ornate*, respectively.

The enhanced solvent extracts of brown algal seaweed with 3 and 5% Pungam oil at various concentrations: 2, 4, 6, 8, 10 and 20% along with solvent control, standard and untreated check was tested against third instar larva of S. litura using no choice leaf dip method. Surface sterilized castor leaves were cut into pieces (5 cm in diameter) and it was treated with the extracts as per treatments. The treated leaves after drying were placed inside the Petri dish (5 leaves per Petri dish) with filter paper and adequate moisture was provided to avoid early drying of test materials. Four hours pre-starved third instar larva was released into each petri dish (5 larvae per Petri dish) and allowed to feed on treated leaves for 24 h. The experiments were laid under a completely randomized design with eight treatments and three replications. Data on larval mortality, pupation, pupal malformation and adult emergence were collected and the means were pooled and analyzed statistically and antifeedant activity was calculated after 24 h after treatment using Leaf Area Meter, respectively.

RESULTS AND DISCUSSION

The combined action of 3% Pungam oil with solvent extract of *T. ornata* showed that the

larvicidal action commenced from 12 HAT and persistently increased up to 96 HAT wherein the larval death rate ranged from 6.67 to 66.66%. Among various treatments, 20% *T. ornata*+3% Pungam oil recorded the greatest larval fatality of 66.66% pursued by 10% *T. ornata*+3% Pungam oil (53.33%), respectively (Figs. 1 and 2). The least larval death rate was recorded in 2% *T. ornata*+3% Pungam oil (33.33%), whereas in standard check 40.00% larval mortality was recorded. Meanwhile no mortality of larva was noted in solvent and untreated controls.



Fig. 1. Larvicidal effect of *T. ornata* acetone extract combined with Pungam oil against *S. litura* at 24 HAT.



Fig. 2. Larvicidal effect of *T. ornata* acetone extract combined with Pungam oil against *S. litura* at 96 HAT.

The remaining larva was allowed to pupate and a higher pupation rate was exhibited by 2% *T. ornata*+3% Pungam oil (66.66%), whereas 20% *T. ornata*+3% Pungam oil displayed the least pupation rate of 33.33% (Fig. 3). Due to enhanced toxicity, higher pupal malformation was noticed in 10% *T. ornata*+3% Pungam oil (13.33%) which was at par with 8% *T. ornata*+3% Pungam oil (Fig. 4) and per cent adult emergence was very low in 20% *T. ornata*+3% Pungam oil (20.00%; Fig. 5) with pupa to the adult conversion ratio of 1 : 0.60 (Table 1), whereas in the solvent and untreated check, 100% adults emerged successfully.



Fig. 3. Effect of *T. ornata* acetone extract combined with Pungam oil on pupation of *S. litura*.



Fig. 4. Effect of *T. ornata* acetone extract combined with Pungam oil on pupal malformation of *S. litura.*

The presence of phagodeterrents in Pungam oil detered the larva from feeding the treated leaf discs. The highest antifeedant activity was observed in 20% *T. ornata*+3% Pungam oil (18.40 sq. cm), whereas in solvent and untreated checks, leaf area index was 8.30 and 1.40 sq. cm, respectively (Table 2).

The influence of enhanced solvent extract of seaweed with 5% Pungam oil on larval fatality initiated from 12 HAT and continued up to 96 HAT. The uppermost larval mortality was



Fig. 5. Effect of *T. ornata* acetone extract combined with Pungam oil on adult emergence of *S. litura.*

Table 2. Antifeedant effect of enhanced brown algal
seaweed, *T. ornata* solvent extract with 3%
Pungam oil on growth and development of
S. litura under laboratory conditions (Full leaf
area=20 sq. cm)

Treatment	Antifeedant activity (sq. cm) 24 HAT
T ₁ -2% T. ornata+3% Pung	gam oil 13.2
T_2 -4% T. ornata+3% Pung	gam oil 13.7
T ₃ -6% <i>T. ornata</i> +3% Pun	gam oil 14.5
T_{4} -8% T. ornata+3% Pung	gam oil 15.8
$T_{5}-10\%$ T. ornata+3% Pu:	ngam oil 16.1
$T_{6}^{-20\%}$ T. ornata+3% Pu:	ngam oil 18.4
T ₇ –3% Pungam oil	16.3
T _s –Solvent control	8.3
T_9 –Untreated control	1.4

noticed in 20% T. ornata+5% Pungam oil (80.00%) at 96 HAT (Fig. 2) and the lowest per cent in 2% *T. ornata*+5% Pungam oil (40.00%), respectively. Meanwhile the standard check showed 53.33% larval mortality. The remaining larvae were allowed for pupation. The lowest pupation rate of 20.00% was seen in 20% T. ornata+5% Pungam oil and 100% pupation occurred in the solvent and untreated control (Fig. 3). Due to IGR activity, 6% T. ornata+5% Pungam oil showed a higher level of pupal malformation rate (13.33%; Fig. 4), whereas minimal adult emergence was noted in 20% T. ornata+3% Pungam oil (13.33%) (Figs. 5 and 6); with pupa to the adult conversion ratio of 1 : 0.66 (Table 1); and the high antifeedant activity with leaf area index 18.90 sq. cm, respectively (Table 3).

Table 1. The effect of T. ornata solvent extract combined with Pungam oil on pupa to adult

Treatment	т ₁ (2%)	Т ₂ (4%)	Т ₃ (6%)	Т ₄ (8%)	T ₅ (10%)	Т ₆ (20%)	Absolute control	Untreated control	Solvent control
T. ornata+3% Pungam oil	1 : 0.90	1 : 0.88	1 : 0.87	1 : 0.75	1 : 0.71	1 : 0.60	1 : 0.88	1:1	1:1
T. ornata+5% Pungam oil	1 : 0.88	1 : 0.85	1 : 0.83	1 : 0.79	1 : 0.79	1 : 0.66	1 : 0.85	1:1	1:1

Table 3. Antifeedant effect of enhanced brown algal
seaweed, T. ornata solvent extract with
Pungam oil 5% on growth and development
of S. litura under laboratory conditions (Full
leaf area=20 sq. cm)

Treatment	Antifeedant activity (sq. cm) 24 HAT
$T_1-2\%$ <i>T.</i> ornata+5% Pun	gam oil 14.2
$T_2-4\%$ <i>T.</i> ornata+5% Pun	gam oil 14.7
$T_3-6\%$ <i>T.</i> ornata+5% Pun	gam oil 15.1
$T_4-8\%$ <i>T.</i> ornata+5% Pun	gam oil 15.8
$T_5-10\%$ <i>T.</i> ornata+5% Pu	ngam oil 16.7
$T_6-20\%$ <i>T.</i> ornata+5% Pu	ngam oil 18.9
$T_7-5\%$ Pungam oil	15.3
T_8 -Solvent control	8.1
T_9 -Untreated control	1.9



Fig. 6. Effect of *T. ornata* acetone extract combined with Pungam oil on adult malformation of *S. litura*.

The results obtained from the above study confirmed that the addition of Pungam oil enhanced the toxicodynamics properties of T. ornata which exhibited remarkable insecticidal action against S. litura in its various life stages and the result was in accordance with the studies tested against various insects. The result obtained from testing the ethanol-water (3:1) extract of Turbinaria decurrens exhibited uttermost larvicidal action against Aedes aegypti and Anopheles stephensi with LC_{50} values of 0.079 μ g/ml and 0.099 μ g/ml, respectively (Yu et al., 2014). Moorthi et al. (2015) studied the pesticide activity of silver nanoparticles synthesized from Sargassum muticum extract tested against Ergolis merione (Lepidoptera : Nymphalidae). The insecticidal activities of nanoparticles against E. merione recorded the prominent physiological changes such as hemolymph protein profile, hemocytes morphology and deteriorated midgut inclusions such as lumen, basement membrane and gastric caeca. Gowthish and Kannan (2018) investigated methanolic extract of brown algal seaweed, Sargassum cristaefolium against third instar larva of S. litura and recorded maximum mortality of 73.33% at 72 h after treatment, respectively. Gonzalez-Castro et al. (2019) evaluated the insecticidal and repellent activities from ethanolic extracts of seaweeds, *Caulerpa sertularioides, Laurencia johnstonii* and *Sargassum horridum* against *Diaphorina citri* adults. Among them, repellency assay with S. *horridum* extract showed higher repellent activity (Index of Behavioural Tendency, IBT=0.376±0.047) at 24 h after treatment than repellent control Neemix.

The antifeedant and growth retarding activities of PONNEEM (Pungam oil+Neem oil) were studied against the fourth instar larva of *Helicoverpa armigera* and the result stated that antifeedant activity was very high (88.4%) even at lower concentrations (15 μ l/l) of PONNEEM and effective growth inhibitory activities were exhibited at 10 μ l/l concentration (Packiam *et al.*, 2014).

CONCLUSION

It can be concluded that solvent extract of *Turbinaria ornata* in combination with Pungam oil could be used as a better alternative to synthetic chemical pesticides for reducing pest infestation and it can be used in bio-intensive pest management modules.

ACKNOWLEDGEMENT

The authors would like to acknowledge the support of the Head, Department of Entomology, Faculty of Agriculture, Annamalai University for the smooth conduct of the the experiments.

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