



E-ISSN: 2320-7078

P-ISSN: 2349-6800

JEZS 2018; 6(5): 2251-2254

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Received: 05-07-2018

Accepted: 10-08-2018

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Assessment of leaf damage in *Rabi* groundnut treated with oil based formulations of *Nomuraea rileyi* against *Spodoptera litura*

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Abstract

The liquid formulations of *Nomuraea rileyi*, an important entomopathogenic fungus were prepared by using two vegetable oils and two mineral oils viz., olive oil, rice bran oil, liquid paraffin oil, heavy grade mineral oil. *N. rileyi* spore mass was harvested from culture plates and mixed to autoclaved test oils in the proportions of 0.1g (0.5 x 10⁸ spores/0.1 g) and 0.2g (0.1 x 10⁹ spores/0.2 g) per 100ml. Triton-X 100, a wetting agent was also used in two different concentrations i.e., 0.05% and 0.1% for all four test oils. The pathogenicity of *N. rileyi* conidia was studied at monthly intervals up to 5 months and mortality percentages of third instar larva of *S. litura* was calculated.

Among the 16 oil based formulations of *N. rileyi* tested in the laboratory, based on the better performance, eight were selected for field evaluation against *S. litura* in *Rabi* groundnut. In all the plots in which treatments are imposed by spraying lab selected oil formulations of *N. rileyi*, the defoliation was significantly less when compared to untreated control treatment. The lower percentages leaf damage of 26, 25.87 and 21.61 with respect to *S. litura* at 5, 10 and 20 DAT was recorded with rice bran oil with 0.2g spores and 0.1ml triton-X 100 oil formulation. In other formulations also leaf damage was considerably lower at 20 DAT. The leaf damage by *S. litura* was 22-25 per cent in the formulations against 49 per cent in the untreated control at 20 days after treatment.

Keywords: *Nomuraea rileyi*, oil formulations, *Spodoptera litura*, groundnut, leaf damage

1. Introduction

The cultivated groundnut (*Arachis hypogaea* L.) is an important oilseed crop of tropical and subtropical areas of the world. It is considered as 13th most important food crop, 4th important source of edible oil and 3rd substantial source of vegetable protein around the world [1]. The seeds are rich source of edible oil (43-45%) and protein (25-28%) and also a valuable source of vitamins namely B, E and K. Groundnut cake, after the oil extraction is a high protein animal feed and haulm provides quality fodder. The cake is used as cattle and poultry feed and also serves as organic manure with high nitrogen content. Among several pests attacking groundnut, *Spodoptera litura* (F.) is the major defoliator causing considerable yield loss. In India, *S. litura* has become a major pest and created a serious threat to agricultural industry due to the development of resistance towards commonly used insecticides.

An indiscriminate use of chemical pesticides is posing threat to the environment and human health. Many species of insect pests have significantly developed resistance to different group of chemical insecticides. So, works on alternate ecofriendly strategies have been initiated, that reduces the negative influence of chemical pesticides.

One line of such strategies is the use of microbial agents/microbial pesticides such as bacteria, virus, fungi, nematodes, protozoa etc.

Usage of entomopathogenic fungi against insect pests gained importance from the last few decades. More than 750 species of fungi, mostly deuteromycetes and entomophthorales, are pathogenic to insects.

Species that have been most intensively investigated as mycoinsecticides in the crop pest control include *Beauveria bassiana*, *Lecanicillium lecanii*, *Metarhizium anisopliae*, *Nomuraea rileyi*, *Paecilomyces fumosoroseus*, *P. farinosus*, *Entomophthora* sp., *Fusarium* sp. and *Aspergillus* sp. They are specific to insects and do not infect host plants. These fungi are cosmopolitan in their distribution and diversity.

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Due to their eco-friendly and bio-persistence behavior and easily preference to kill pest species at different developmental stages, their utilization increases day-by-day [11].

Nomuraea rileyi (Farlow) Samson is a deuteromycetous fungus of cosmopolitan nature. *N. rileyi* is an important mortality factor for many lepidopteran insects throughout the world. It has the potential to cause spectacular epizootics under favorable environmental conditions. In India, epizootics of *N. rileyi* were recorded on lepidopteran insect pests in field crops and forest trees. In Andhra Pradesh also regular occurrence of *N. rileyi* is being observed on *Helicoverpa armigera*, *Spodoptera litura*, *Plusia sps etc.*, in crops like groundnut, cotton under favorable ecosystem.

2. Material and Methods

2.1 Preparation of oil based formulations of *N. rileyi*

The test oils used for the preparation of *N. rileyi* formulations are commonly and commercially available vegetable and mineral oils viz., Olive oil, rice bran oil, liquid paraffin oil, heavy grade mineral oil. The selected oils manufactured by standard companies were purchased. The oils were poured into sterilized conical flasks/blue cap bottles of 250 ml and autoclaved at 15 psi pressure at 121°C for 15 min. Each oil was considered as a treatment and three replications were maintained (100ml/replication). The harvested spores of *N. rileyi* were mixed to the test oils in the proportions of 0.1g and 0.2g per 100 ml of test oil. Triton-X 100, a wetting agent was also used in two different concentrations i.e., 0.05% and 0.1% for all four test oils for uniform mixing of spores under aseptic conditions.

2.2 Details of the field experiment

The oil based formulations of *N. rileyi* that recorded considerably higher larval and pupal mortalities under laboratory conditions were used for evaluating per cent leaf damage in *Rabi* groundnut crop against *S. litura*, at dry land farm, S.V. Agricultural College, Tirupati. A Randomized Block Design (RBD) was laid with three replications of 5m x 4m plot size. Seeds of the Narayani variety of Groundnut treated with mancozeb @ 3 g kg⁻¹ seeds were sown in rows at 22.5 cm apart and 10 cm spacing was maintained between plants. All the recommended package of practices was followed to raise successful crop except plant protection measures. When considerable damage of *S. litura* was noticed reaching above ETL, then the sprayings of *N. rileyi* was done (at 40 DAS). The experiment includes eight treatments and an untreated control with three replications each.

2.3 Preparation of spray suspension from oil formulations for field spraying

For field spraying, from each oil based formulation, 5ml was taken with the help of measuring cylinder and dissolved in a litre of water and mixed thoroughly. A quantity of 3.5 lit of this spray fluid was used to sufficiently wet the groundnut foliage (20 m² area) with the help of foot sprayer.

2.4 Observations

2.4.1 Assessment of leaf damage of *S. litura*

In each replication, in all the treatments the tagged plants were observed for healthy and damaged leaves at 5, 10 and 20 days after treatment. A pre-count was also taken and per cent leaf damage was calculated by using the following formula [10].

$$\text{Per cent leaf damage} = \frac{\text{Total no. of damaged leaves}}{\text{Total no. healthy leaves}} \times 100$$

2.5 Statistical Analysis

The data obtained on leaf damage was subjected to statistical analysis (ANOVA). Per cent values were transformed to arc-sin values before subjecting to statistical analysis. Means were separated by Duncan's Multiple Range Test (DMRT). The statistical analysis carried out through SPSS.20.00.

3. Results and Discussion

A day before application, the leaf damage was above ETL (30-34%) and uniform (Plate 1.). The per cent leaf damage values due to *S. litura* at 5, 10 and 20 days after treatment are presented in Table 1. and Fig 1.

The results indicate that rice bran oil with 0.2g spores and 0.1ml triton-X 100 oil formulation treated plot recorded the lower percentages leaf damage of 26, 25.87 and 21.61 with respect to *S. litura* at 5, 10 and 20 DAT. The treatments liquid paraffin oil with 0.2 g spores and 0.1 ml of triton-X 100, heavy grade mineral oil with 0.2 g spores and 0.1 ml of triton-X 100 recorded 28-23 per cent leaf damage up to 20 DAT.

All the other oil formulations of *N. rileyi* recorded the lowest leaf damage of 29-24 per cent compared to untreated control which recorded highest leaf damage of 40, 46 and 49 per cent at 5, 10 and 20 DAT.



Plate 1: Skeletonization of groundnut leaflets by early instar *S. litura*

Table 1: *Spodoptera litura* damage in *Rabi* groundnut when different oil based formulations of *N. rileyi* applied

Treatments	Mean per cent leaf damage			
	Pre-treatment	5 DAT	10 DAT	20 DAT
T ₁ :	31.76	26.00 ^{ab} (32.52)	25.87 ^{ab} (30.55)	21.61 ^a (27.68)
T ₂ :	30.57	27.75 ^a (31.76)	25.18 ^a (30.10)	22.82 ^{ab} (28.52)
T ₃ :	30.86	27.86 ^{ab} (31.85)	25.73 ^{ab} (30.46)	23.28 ^{abc} (28.83)
T ₄ :	30.69	28.38 ^{ab} (32.17)	25.79 ^{ab} (30.50)	24.06 ^{bc} (29.36)
T ₅ :	33.03	28.72 ^{ab} (32.39)	26.26 ^{ab} (30.81)	24.18 ^{bc} (29.44)
T ₆ :	32.00	29.03 ^{ab} (32.58)	27.40 ^b (31.55)	24.46 ^{bc} (29.62)
T ₇ :	31.73	29.36 ^{ab} (32.79)	27.39 ^b (31.54)	24.49 ^{bc} (29.63)
T ₈ :	32.05	29.73 ^b (33.02)	27.39 ^b (31.54)	25.20 ^c (30.11)
T ₉ :	33.24	40.02 ^c (39.23)	45.87 ^c (42.45)	49.06 ^d (44.47)
SE(m) ±		0.55	0.51	0.63
C.D.(p = 0.05)		1.67	1.55	1.90

Figures in parenthesis indicate angular transformed values.

Means in the column followed by same letter(s) are not significantly different by DMRT

DAT = Days after Treatment

Data are the means of three replications

T₁: Rice bran oil with 0.2g spores and 0.1ml triton-X 100,

T₂: Liquid paraffin with 0.2g spores and 0.1ml triton-X 100,

T₃: Heavy grade mineral oil with 0.2g spores and 0.1ml triton-X 100,

T₄: Rice bran oil with 0.2g spores and 0.05ml triton-X 100,

T₅: Liquid paraffin with 0.2g spores and 0.05ml triton-X 100,

T₆: Heavy grade mineral oil with 0.2g spores and 0.05ml triton-X 100,

T₇: Rice bran oil with 0.1g spores and 0.1ml triton-X 100, T₈:

Olive oil with 0.2g spores and 0.1 ml triton-X 100, T₉:

Untreated control

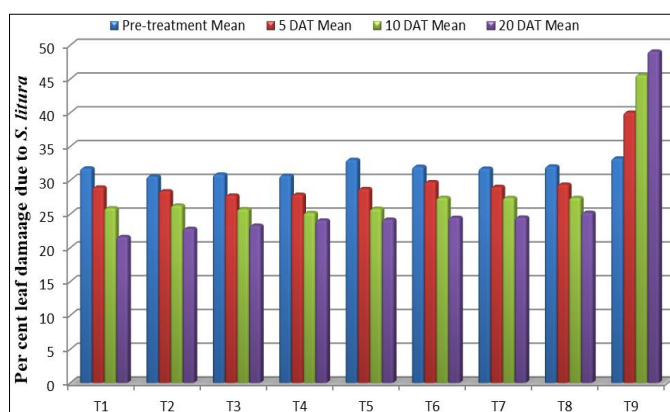


Fig 1: Groundnut leaf damage due to *S. litura* recorded with different oil based formulations of *N. rileyi* in Rabi

T₁: Rice bran oil with 0.2g spores and 0.1ml triton-X 100,

T₂: Liquid paraffin with 0.2g spores and 0.1ml triton-X 100,

T₃: Heavy grade mineral oil with 0.2g spores and 0.1ml triton-X 100,

T₄: Rice bran oil with 0.2g spores and 0.05ml triton-X 100,

T₅: Liquid paraffin with 0.2g spores and 0.05ml triton-X 100,

T₆: Heavy grade mineral oil with 0.2g spores and 0.05ml triton-X 100,

T₇: Rice bran oil with 0.1g spores and 0.1ml triton-X 100, T₈:

Olive oil with 0.2g spores and 0.1 ml triton-X 100, T₉:

Untreated control

The effectiveness of oil based formulations (Diesel: Sunflower oil 7:3) of conidia of fungal isolate *N. rileyi* N812 against *H. armigera* has been evaluated. The conidia were found to be most effective in controlling *H. armigera*. The per cent efficacy was 61 per cent. Pod damage was 15.48 and yield was 12.62 per ha in *N. rileyi* treated plots [8].

It has been reported that the conidial suspension of *B. bassiana* in oil was effective for field application because of its non-drying properties. The oil formulation of *B. bassiana* exhibited the additional advantage of prolonged conidial survival [9].

B. bassiana was tested under field conditions to control *H. armigera* infesting chickpea for two crop seasons and was found very effective. At a spore concentration of 2.68×10^7 spores per ml, the average pod damage was 6.8 per cent and yield of 2377 kg ha⁻¹. Where untreated control recorded 16.3 per cent pod damage with a yield of 1344 kg h⁻¹ [4].

A Field experiment has been carried out to determine the

effectiveness of *N. rileyi* against *S. litura* on beet root at IIHR, Bangalore. Five applications of *N. rileyi* @ 3.2×10^6 spore/ml along with Triton x -100 (0.01%) at weekly interval effectively controlled the larval population of *S. litura* (0.28 larvae/plant) compared to mean larval population of 2.03 larvae/plant in untreated control plot. They also recorded minimum % root damage in *N. rileyi* treated plot (7.21%) whereas 51.91% in untreated plot. Marketable yield was also significantly higher in fungus treated plot which recorded 4.38 tonnes/acre followed by 3.81 tonnes/acre in endosulfan as against 2.33 tonnes/acre in untreated plot [2].

The effectiveness of *N. rileyi* against *S. litura*, in potato has been studied. The fungus when sprayed @ 2×10^8 conidia/liter thrice on potato at 15 days interval from 50 days after sowing proved as effective as SNPV and *Bt* and reduced up to 32% defoliation [5].

Formulations like oil based (sunflower oil), wettable powder (talc) and crude formulation of *N. rileyi* were evaluated against *H. armigera* under chickpea ecosystem. The results clearly indicated that significantly higher per cent mycosis (26.56%) was recorded in oil formulation of *N. rileyi* @ 2×10^{11} conidia per ha which reflected on least pod damage (26.43%) which in turn resulted in getting higher pod yield (9.97 q ha⁻¹) as compared to other two formulations and control [6].

It has been found that the oil based formulation of *M. anisopliae* recorded minimum of 5.25 leafhopper per three leaves followed by *B. bassiana* oil based (6.88) and *V. lecani* oil based (7.75) after the second spray in okra. The yield of okra was significantly higher in oil based formulation of *M. anisopliae* (38.80 q ha⁻¹) and *V. lecanii* (38.50 q ha⁻¹) with monetary returns of Rs. 14720 and Rs. 14480 ha⁻¹, respectively followed by *B. bassiana* [3].

It has been evaluated that the oil formulations of *M. anisopliae* (M 34412), *B. bassiana* (B 3301) and *N. rileyi* (N 3.12) under field conditions in pigeonpea against *H. armigera*. After two sprays, per cent efficacy of various treatments ranged from 55.51 to 70.93. The treatment with *M. anisopliae* was found to be effective with maximum efficacy of 70.93 per cent, *N. rileyi* with 62.95 per cent. The per cent pod damage in *M. anisopliae*, *N. rileyi* was 8.76 and 10.24, respectively [7].

4. Conclusion

The present results proved that storage of *N. rileyi* in best suited vegetable oils such as rice bran oil and mineral oils such as liquid paraffin would be more advantages for maintenance of satisfactory viability and virulence up to 5 months. In field evaluation also rice bran oil with 0.2g spores and 0.1ml triton-X 100 formulation treated plot recorded lowest leaf damage due to *S. litura* (21%) at 20 days after treatment.

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