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# Effect of Plain and Nano Emulsions of Tree Borne Oil Seed Protectants on Bruchids (*Callosobruchus Maculatus*) in Blackgram

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**ABSTRACT**

A laboratory investigation was carried out to assess the efficacy of plain and nano emulsions of tree borne oils (neem and pongamia) seed protectants against bruchids, *Callosobruchus maculatus* (Linnaeus) as surface treatment of both uninfested and infested black gram seeds @10ml.kg$^{-1}$ at Tamil Nadu Agricultural University, Coimbatore during 2014. Of the two forms of tested tree borne oils uninfested seeds treated with plain forms (0.0 adults/week/250g seeds) were superior over nano emulsions each of which had significantly higher numbers (0.4 adults/week/250g seeds). Treated uninfested seeds when reintroduced with another 10 bruchids after eight weeks of observation were found dead within a week time indicating the persistence effect of plain oils of neem and pongamia in killing all the introduced adults but nano emulsions of neem and pongamia oils promoted the emergence of 83.5 and 44.7 adults per week per 250g seeds. In case of infested seeds, nano emulsion of neem and pongamia oils were found to favour the insect development. Uninfested seeds treated with plain and nano formulations of neem and pongamia oils had 93 and 92 percentage germination while infested seeds, treated with nano pongamia oil (72%) and nano neem oil (60%) exhibited low germination.

**Key words :** *Callosobruchus maculatus*, Efficacy, Germination, Infestation, Mortality rate, Nano emusions, Neem oil, Pulses, Pongamia oil.

*Callosobruchus maculatus* (Linnaeus) (Bruchidae: Coleoptera) is the most serious insect pest of storage and attacks invariably all the pulses under storage conditions with the mild infestation in the field as well. It is therefore, imperative to save the stored pulses from this serious pest. A single beetle could cause 3.5 per cent weight loss in cowpea seeds (Booker, 1967). Gujar and Yadav (1978) recorded infestation of 55-60 per cent loss in seed weight and 45.50 to 66.30 per cent loss in protein content due to damage by the pulse beetle. Infestations can cause up to 60 per cent loss in seed weight and up to 66 per cent loss in protein content of pulses. The plant materials are potentially suitable for use in integrated pest management (IPM) effectively. Conventional chemicals/ insecticides when used to control stored insects resulted in leaving objectionable residues on treated commodity and generally are hazardous to handle and apply. Besides some of the stored product insects have also developed resistance to these insecticides (Chitra Srivastava *et al.*, 2000). In order to manage the storage pests attacking grains and seeds, newer eco-friendly management methods have to be developed. Azadirachtin has broad mode of activity, working as a feeding deterrent, insect-growth regulator, repellent, and sterilant; and it may also inhibit oviposition. The neem based botanicals are active on a broad range of insects, including stored grain pests, aphids, caterpillars and mealybugs and are easily biodegradable (Freedman *et al*., 1979). Rajapakse *et al*. (1998) reported that neem oil gave highest reduction in oviposition and adult emergence of *C.maculatus* in cowpea and resulted in the highest adult mortality in six days after treatment. Rahman and Talukdar (2006) observed the least number of F1 adult emergence of *C. maculatus* from black gram seeds treated with neem oil @ 10 ml per kg. The oil treatment did not show any adverse effects on germination of seeds even after three months after treatment. Alice *et al*. (2007) found that the lowest *C. maculatus* infestation was observed when black gram was treated with 1% neem oil. Grain protection potential of different plant derivatives including the plant oil against major stored product pests was found very promising and could reduce the risks associated with the use of insecticides (Shaaya *et al*., 1997). The present investigations were carried out on *C. maculatus*

Table 1. Effect of tree borne oils on bruchids of uninfested black gram seed (Live adults).

| Treatments | Live adults (No.)/250g | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | Period | | | | | | | | |
| | 1WAT | 2WAT | 3WAT | 4WAT | 5WAT | 6WAT | 7WAT | 8WAT | Mean |
| Absolute check | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) |
| Standard check | 7.4 (2.8) | 7.0 (2.7) | 7.2 (2.7) | 7.0 (2.7) | 6.8 (2.7) | 8.0 (2.9) | 7.4 (2.8) | 7.4 (2.8) | 7.2 (2.5) |
| Neem oil @10ml.Kg$^{-1}$ | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.0) |
| Nano neem oil @10ml.Kg$^{-1}$ | 1.0 (1.2) | 0.9 (1.2) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.1 (0.7) |
| Pungam oil 10ml.Kg$^{-1}$ | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) |
| Nano pungam oil 10ml.Kg$^{-1}$ | 1.8 (1.4) | 1.4 (1.3) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.4 (0.9) |
| Mean | 1.7 (1.5) | 1.5 (1.4) | 1.2 (1.3) | 1.1 (1.2) | 1.1 (1.2) | 4.0 (2.1) | 1.2 (1.3) | 1.2 (1.3) | 1.6 (1.4) |
| | | SE(d) | CD(0.05) | | | | | | |
| Treatment | | 0.03 | 0.06 | | | | | | |
| Period | | 0.03 | 0.07 | | | | | | |
| Interaction | | 0.09 | 0.18 | | | | | | |

*WAT= Week after treatment Values in the parentheses are square root transformed

to assess the efficacy of tree borne oils of neem and pongamia in plain as well as nano emulsions

## MATERIALS AND METHODS

### Mass culturing of bruchid

Mass culturing of bruchid was done at the Insectary, TNAU, Coimbatore. The bruchid adults collected from black gram seed samples of the Department of Seed Science and Technology, confirmed as *Callosobruchus maculatus* by morphological characters and were utilized for mass culturing. The bruchids were reared on fresh black gram seeds (Plate 1.a, Plate 1.b, Plate 1.c) disinfested by sun drying to kill any existing insect stages. One hundred adult insects were released in 400 g black gram seeds in a 500 ml plastic container covered with muslin cloth to ensure ventilation and the culture maintained at 25+5ºC and 70±5% RH throughout the period of investigation. Adults numbering 20 were released and the jar was left for 25 days to obtain adult uniform aged beetles for the experiment. Sub culturing of the beetle was done at weekly intervals so as to have a continuous supply of insects for experiments proposed in this investigation.

### Preparation of nano emulsions of Neem / Pongamia oil

Commercial neem/pongamia oil, commercial surfactant (Tween 60) and distilled water (1:4:10) were used in the preparation of nano emulsion (Plate 2). Tween 60 was used as the surfactant because non-ionic surfactants are known to be less affected by pH and ionic strength.

### Characterization of nano particles using Particle size analyzer

In the present study, HORIBA nano particle size analyser SZ 100 was used. Accurately, 0.5 mg of sample was dispersed in 10 ml pure water through ultrasonication and the measurements were taken. Nano emulsions of neem and pongamia measured 486.7 and 36.2 nm at the tested effective surfactant concentrations and found stable (Plate 3 and Plate 4).

Table 2. Effect of tree borne oils on bruchids of uninfested black gram seed (Dead adults).

| Treatments | Dead adults (No.)/250g | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | Period | | | | | | | | |
| | 1WAT | 2WAT | 3WAT | 4WAT | 5WAT | 6WAT | 7WAT | 8WAT | Mean |
| Absolute check | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) |
| Standard check | 0.6 (1.0) | 0.6 (1.0) | 1.2 (1.3) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.3 (0.9) |
| Neem oil @10ml.Kg$^{-1}$ | 10 (3.2) | 2.2 (1.6) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 1.5 (1.4) |
| Nano neem oil @10ml.Kg$^{-1}$ | 8.8 (3.0) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 1.1 (1.2) |
| Pungam oil 10ml.Kg$^{-1}$ | 10 (3.2) | 2.2 (1.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 1.5 (1.4) |
| Nano pungam oil 10ml.Kg$^{-1}$ | 4.6 (2.2) | 3.2 (1.9) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.9 (1.1) |
| Mean | 4.0 (2.1) | 1.0 (1.2) | 0.2 (0.8) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.6 (1.0) |
| | | SE(d) | CD(0.05) | | | | | | |
| Treatment | | 0.02 | 0.04 | | | | | | |
| Period | | 0.02 | 0.05 | | | | | | |
| Interaction | | 0.06 | 0.13 | | | | | | |

*WAT= Week after treatment Values in the parentheses are square root transformed

**Efficacy of plain and nano emulsions of neem, pongamia oil on bruchids**

Sample seeds of 250g uninfested and infested seeds taken in polybags (25x20cm) suitably perforated for aeration at the upper half, were added with 2.5ml of plain (Plate 5. a, Plate 5. b) and nano emulsions (Plate 5. c, Plate 5. d, Plate 5. e, Plate 5. f) separately and vigorously shaken manually so as to have uniform coating on the seed surface. In case of the uninfested treated samples 10 bruchids were released using aspirator while the infested samples left without any addition.

Sufficient number of replications (five) were maintained and kept for weekly observations on number of adults emerged (live and dead), dead adults were removed from the released adults and seeds were analysed for germination as per standard protocols prescribed by ISTA (1999).

**Seed germination**

Seed germination was assessed by following towel roll method as recommended by ISTA (1999) in germination room maintained at 25±5°C temperature and 95±3 per cent RH. After eight days, the seedlings were evaluated and the normal seedlings were counted and expressed in percentage (Plate 6).

**Vigour index**

Vigour index (VI) was calculated by using the formula suggested by Abdul–Baki and Anderson (1973) and expressed in whole number.

VI = Germination percentage x [Root length (cm) + shoot length (cm)].

X 100

Germination percentage = Total number of germinated seeds / Total number of seeds x 100

**Statistical analyses**

The data obtained from different experiments were analyzed by analysis of variance as described by Panse and Sukhatme (1985). Per cent values were subjected to arcsine transformation and insect counts were transformed

Table 3. Effect of tree borne oils on re-introduced bruchids after eight weeks of treatment of uninfested black gram seed (Live adults).

| Treatments | Live adults (No.)/250g | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | Period | | | | | | | | |
| | 1WAT | 2WAT | 3WAT | 4WAT | 5WAT | 6WAT | 7WAT | 8WAT | Mean |
| Absolute check | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) |
| Standard check | 7.7 (2.8) | 9.1 (3.0) | 9.5 (3.1) | 11.9 (3.5) | 22.7 (4.8) | 22.9 (4.8) | 24.3 (4.9) | 26.5 (5.1) | 16.8 (4.1) |
| Neem oil @10ml.Kg$^{-1}$ | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) |
| Nano neem oil @10ml.Kg$^{-1}$ | 48.6 (7.0) | 79 (8.9) | 82.4 (9.1) | 85.6 (9.2) | 87.8 (9.4) | 90.6 (9.5) | 95 (9.7) | 99 (9.9) | 83.5 (9.1) |
| Pungam oil 10ml.Kg$^{-1}$ | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) |
| Nano pungam oil 10ml.Kg$^{-1}$ | 39.8 (6.3) | 44.6 (6.7) | 41.2 (6.4) | 45 (6.7) | 43.2 (6.6) | 46 (6.8) | 46.2 (6.8) | 51.8 (7.2) | 44.7 (6.7) |
| Mean | 16.0 (4.0) | 22.1 (4.7) | 22.1 (4.7) | 23.7 (4.8) | 25.6 (5.1) | 26.5 (5.2) | 27.5 (5.3) | 29.5 (5.4) | 24.1 (4.9) |
| | | SE(d) | CD(0.05) | | | | | | |
| Treatment | | 0.05 | 0.11 | | | | | | |
| Period | | 0.06 | 0.13 | | | | | | |
| Interaction | | 0.16 | 0.32 | | | | | | |

*WAT= Week after treatment Values in the parentheses are square root transformed

to square root transformation before subjected to the analysis. The critical differences (CD) were calculated at five per cent probability level.

## RESULTS AND DISCUSSION

Botanicals are non toxic and considered ecofriendly in storage pest management. Plain and nano emulsions of tree borne oils (neem and pongamia), at 10ml.Kg$^{-1}$ was tested as surface treatment of black gram seeds against bruchids. In uninfested seeds treated with neem, pongamia oils and also absolute check had nil population against the standard check (7.2 adults/week/250g seed) in all the periods of observations. Neem and pongamia oils exhibited similar trend wherein plain forms (0.0 adults/week/250g seeds) were superior over nano emulsions each of which had significantly higher numbers ( 0.4 adults/week/250g seeds) though not exorbitantly (Table 1 and Table 2).

After eight weeks of treatment, again reintroduced with another ten adults into neem and pongamia oil treated seeds had no infestation at all during further eight weeks of observations. Surprisingly nano emulsion oils of neem and pongamia treated seeds favoured the reintroduced adults for further development and resulted in 83.5 and 44.7 adults per week per 250g seeds in period of another eight weeks (Table 3). Comparatively emerging adults were more in the seeds treated with nano emulsion of neem oil.

All the reintroduced adults after a week of release were found dead indicating the persistence effect of oils of neem and pongamia in killing all the introduced adults. There was no significant difference between nano emulsions of neem (10.3 adults/week/250g seed) and pongamia (10.1 adults/week/250 g seed) oils (Table 4). In infested seeds, neem oil plain form could cause 30 times reduction in the adult emergence while nano emulsion of neem oil 1.4 times (Table 5) increased over the standard

Table 4. Effect of tree borne oils on re-introduced bruchids after eight weeks of treatment of uninfested black gram seed (Dead adults).

| Treatments | Dead adults (No.)/250g | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | Period | | | | | | | | |
| | 1WAT | 2WAT | 3WAT | 4WAT | 5WAT | 6WAT | 7WAT | 8WAT | Mean |
| Absolute check | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) |
| Standard check | 0.8 (1.1) | 0.4 (0.9) | 1.0 (1.2) | 1.6 (1.4) | 0.8 (1.1) | 0.2 (0.8) | 0.4 (0.9) | 1.2 (1.3) | 1.6 (1.4) |
| Neem oil @10ml.Kg$^{-1}$ | 10.0 (3.2) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 1.2 (1.3) |
| Nano neem oil @10ml.Kg$^{-1}$ | 5.2 (2.4) | 8.2 (2.9) | 9.4 (3.1) | 10.2 (3.2) | 11.4 (3.4) | 12.2 (3.5) | 12.4 (3.6) | 10.2 (3.2) | 10.3 (3.2) |
| Pungam oil 10ml.Kg$^{-1}$ | 10.0 (3.2) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 0.0 (0.7) | 1.2 (1.3) |
| Nano pungam oil 10ml.Kg$^{-1}$ | 5.0 (2.3) | 8.0 (2.9) | 9.0 (3.0) | 10.0 (3.2) | 11.4 (3.4) | 11.8 (3.5) | 12.4 (3.6) | 13.8 (3.7) | 10.1 (3.2) |
| Mean | 5.1 (2.3) | 2.7 (1.7) | 3.2 (1.9) | 3.6 (2.0) | 3.9 (2.0) | 4.0 (2.1) | 4.2 (2.1) | 4.2 (2.1) | 4.0 (2.1) |
| | | SE(d) | CD(0.05) | | | | | | |
| Treatment | | 0.03 | 0.07 | | | | | | |
| Period | | 0.04 | 0.08 | | | | | | |
| Interaction | | 0.10 | 0.20 | | | | | | |

*WAT= Week after treatment Values in the parentheses are square root transformed

check (90.9 adults/week/250g seed) indicating the nano emulsion of neem oil were found to favour the insect development. In case of pongamia oil, plain form could bring 10 times reduction in the adult emergence while nano emulsion of pongamia oil 1.4 times increased over the standard check. Nano emulsions treated seeds had 27.6 (neem) and 21 (pongamia) dead adults per week per 250g seeds in accordance with live adults observed in the seeds.

Comparing the two oils, as far as plain form is concerned, neem oil treatment was found to perform better than pungam whereas in nano emulsion form the reverse performance was observed.

Rahman and Talukdar (2006) proved that the least number of F1 adult emergence of *C. maculatus* from black gram seeds treated with neem oil @ 10 ml per kg. Alice *et al*. (2007) found that the lowest *C. maculatus* infestation was observed when black gram was treated with 1% neem oil. Nano neem oil @10ml.Kg$^{-1}$ had cent per cent mortality of the adult bruchids in cowpea when released immediately after the treatment and until six week period of treatment (Mariam, 2011) while the same effect could not be obtained when adults were reintroduced after six weeks favouring the insect infestation even more than the standard check.

Insecticidal properties of different botanicals against *Callosobruchus spp* reported time to time and adjudge their potential for future commercialization as a biorational alternative to control the bruchids in stored leguminous grains. A neurotoxic mode of action by interrupting the function of a neuromodulator octapamine and thus breakdown of the nervous system of insects is reported (Kostyukovsky *et al*., 2002) due to plant based pesticides.

With a view to assess the impact of tested plant oils on the germination of seeds, the treated uninfested seeds when evaluated for germination

Plate 1 a Healthy black gram seeds (Vamban-5)

Plate 1 b Artificially infested black gram seeds (Vamban-5)

Plate 1 c Mass culturing of black gram bruchids

Plate 2 Nano emulsions of neem and pongamia oil

2013.10.28 10:24:59

**Neem emulsion.nsz**

**Measurement Results**

| | | |
|---|---|---|
| Date | : | Monday, October 28, 2013 10:24:22 AM |
| Measurement Type | : | Particle Size |
| Sample Name | : | Neem emulsion |
| Scattering Angle | : | 90 |
| Temperature of the holder | : | 25.0 °C |
| T% before meas. | : | 24679 |
| Viscosity of the dispersion medium | : | 0.894 mPa·s |
| Form Of Distribution | : | Standard |
| Representation of result | : | Scattering Light Intensity |
| Count rate | : | 1289 kCPS |

**Calculation Results**

| Peak No. | S.P.Area Ratio | Mean | S. D. | Mode |
|---|---|---|---|---|
| 1 | 1.00 | 123.1 nm | 31.8 nm | 112.1 nm |
| 2 | --- | --- nm | --- nm | --- nm |
| 3 | --- | --- nm | --- nm | --- nm |
| Total | 1.00 | 123.1 nm | 31.8 nm | 112.1 nm |

**Cumulant Operations**

| | | |
|---|---|---|
| Z-Average | : | 486.7 nm |
| PI | : | 0.718 |

**Molecular weight measurement**

| | | |
|---|---|---|
| Molecular weight | : | --- |
| Mark-Houwink-Sakurada parameters | : | --- |

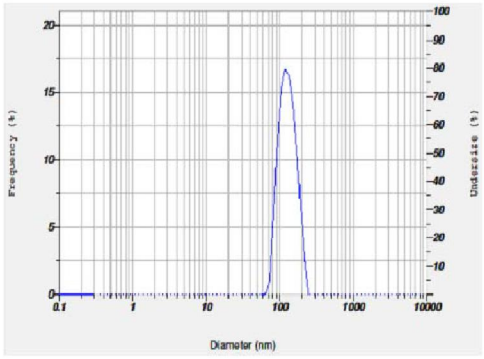


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Plate 3 Nano emulsion of neem oil size using particle size analyzer

2013.10.28 10:32:25

**pumgam emulson 1.nsz**

**Measurement Results**

| | | |
|---|---|---|
| Date | : | Monday, October 28, 2013 10:31:46 AM |
| Measurement Type | : | Particle Size |
| Sample Name | : | pumgam emulson 1 |
| Scattering Angle | : | 90 |
| Temperature of the holder | : | 25.0 °C |
| T% before meas. | : | 18752 |
| Viscosity of the dispersion medium | : | 0.894 mPa·s |
| Form Of Distribution | : | Standard |
| Representation of result | : | Scattering Light Intensity |
| Count rate | : | 3022 kCPS |

**Calculation Results**

| Peak No. | S.P.Area Ratio | Mean | S. D. | Mode |
|---|---|---|---|---|
| 1 | 1.00 | 165.2 nm | 681.5 nm | 15.9 nm |
| 2 | --- | --- nm | --- nm | --- nm |
| 3 | --- | --- nm | --- nm | --- nm |
| Total | 1.00 | 165.2 nm | 681.5 nm | 15.9 nm |

**Cumulant Operations**

| | | |
|---|---|---|
| Z-Average | : | 36.2 nm |
| PI | : | 0.556 |

**Molecular weight measurement**

| | | |
|---|---|---|
| Molecular weight | : | --- |
| Mark-Houwink-Sakurada parameters | : | --- |

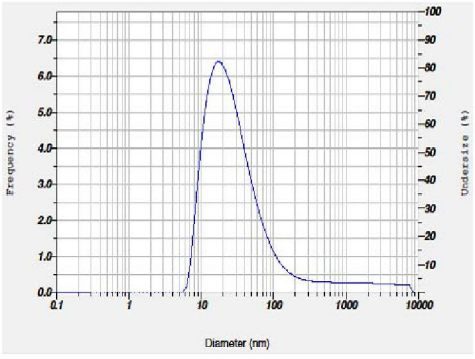


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Plate 4 Nano emulsion of pongamia oil size using particle size analyzer

Plate 5. a Experimental set up for neem oil treatment

Plate 5.b Experimental set up for nano neem oil treatment

Plate 5. c Experimental set up for pongamia oil treatment

Plate 5. d Experimental set up for nano pongamia oil treatment

Plate 5. e Experimental set up for absolute check

Plate 5. f Experimental set up for standard check

Table 5. Effect of tree borne oils on bruchids of infested black gram seed.

| Treatments | Live adults (No.) | | | | | Dead adults (No.) | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Period | | | | | Period | | | | |
| | 1WAT | 2WAT | 3WAT | 4WAT | Mean | 1WAT | 2WAT | 3WAT | 4WAT | Mean |
| Standard check | 64.6 | 84.4 | 97.8 | 116.8 | 90.9 | 78.4 | 83 | 87.2 | 84.6 | 83.3 |
| | (8.06) | (9.2) | (9.9) | (10.8) | (9.5) | (8.8) | (9.1) | (9.3) | (9.2) | (9.1) |
| Neem oil @10ml.Kg$^{-1}$ | 8 | 2.4 | 2 | 0.0 | 3.1 | 88.4 | 59.4 | 10.2 | 0.0 | 39.5 |
| | (2.8) | (1.6) | (1.5) | (0.7) | (1.8) | (9.3) | (7.5) | (3.1) | (0.7) | (6.3) |
| Nano neem oil @10ml.Kg$^{-1}$ | 125.6 | 125 | 137.2 | 141.8 | 132.4 | 23.2 | 27.8 | 26.8 | 32.8 | 27.6 |
| | (11.2) | (11.1) | (11.7) | (11.9) | (11.5) | (4.7) | (5.1) | (5.0) | (5.6) | (5.3) |
| Pungam oil 10ml.Kg$^{-1}$ | 19.4 | 17.4 | 2.2 | 0.0 | 9.7 | 137.4 | 105 | 45.4 | 0.0 | 71.9 |
| | (4.4) | (4.1) | (1.5) | (0.7) | (3.2) | (11.6) | (10.1) | (6.7) | (0.7) | (8.5) |
| Nano pungam oil 10ml.Kg$^{-1}$ | 68.6 | 90.4 | 123.2 | 138 | 105.0 | 25.8 | 18.2 | 19.8 | 20.2 | 21 |
| | (8.2) | (9.4) | (11.0) | (11.7) | (10.2) | (5.0) | (4.2) | (4.4) | (4.4) | (4.6) |
| Mean | 57.2 | 63.9 | 72.4 | 79.3 | 68.2 | 70.6 | 58.6 | 37.8 | 27.5 | 48.6 |
| | (7.5) | (8.0) | (8.5) | (8.9) | (8.2) | (8.4) | (7.6) | (6.1) | (5.2) | (7.0) |
| | SE(d) | CD(0.05) | | | | SE(d) | CD(0.05) | | | |
| Treatment | 0.21 | 0.43 | | | | 0.34 | 0.68 | | | |
| Period | 0.19 | 0.38 | | | | 0.39 | 0.79 | | | |
| Interaction | 0.43 | 0.86 | | | | 0.68 | 1.36 | | | |

*WAT= Week after treatment Values in the parentheses are square root transformed

Table 6. Effect of tree borne oils on germination of uninfested black gram seed.

| Treatments | Germination (%)* | Vigour Index (VI) ** |
|---|---|---|
| Absolute check | 100 | 3142 |
| | (89.71) | (3.49) |
| Standard check | 92 | 3507.3 |
| | (73.57) | (3.54) |
| Neem oil @10ml.Kg$^{-1}$ | 93 | 3684.93 |
| | (74.66) | (3.56) |
| Nano neem oil @10ml.Kg$^{-1}$ | 93 | 3922.7 |
| | (74.66) | (3.59) |
| Pungam oil 10ml.Kg$^{-1}$ | 93 | 3702 |
| | (74.66) | (3.56) |
| Nano pungam oil 10ml.Kg$^{-1}$ | 92 | 3972.66 |
| | (73.57) | (3.59) |
| Mean | 94 | 3655.2 |
| | (75.82) | (3.56) |
| | Treatment | Treatment |
| SE (d) | 1.54 | 0.01 |
| CD (0.05) | 3.35 | 0.02 |

*Values in the parentheses are arcsine transformed

**Values in the parentheses are log transformed

Plate 6 Germination test by Roll towel method

revealed significant reduction when the seeds coated with oils (neem and pongamia) either plain or nano forms (93%) compared to absolute check (100%) (Table 6). In case of infested seeds, after four weeks of treatment revealed reduced germination in seeds treated with neem oil either plain or nano forms (72 and 60 %) unlike uninfested seeds. In infested seeds pongamia oil (72%) treated seed had low germination similar to neem and nano neem forms (Table 7). Considering the difference between the uninfested and infested seeds, germination was reduced in infested seeds due to infestation by the insect. Among the nano formulations, pongamia treatment increased the germination than neem. This might be due to less number of adults observed indicating reduced level of infestation due to pongamia nano oil than it's neem counterpart.

## CONCLUSION

In general the plain forms of oils performed better than the nano formulations. In the case of plain oils, no dilution was effected as the oils as such used for treating the seeds and hence the seeds receiving the oils could have more botanical principles on the seed coat than the nano emulsion forms wherein only one tenth of principle compound would get coated on the seeds as the formuation is diluted 10 times. Concentrations of principle compounds in the tested oils might have gone to the sub lethal doses which might act as a growth promoting factors for insects. Physiological changes in the treated seeds induced by the treatment of nano-particles (Senthilkumar, 2011) might have paved way for the insect to utilize the altered physiological conditions for the insect development thereby leading to more adult population in nano forms than the plain forms.

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Table 7. Effect of tree borne oils on germination of infested black gram seed.

| Treatments | Germination (%)* | Vigour Index (VI) ** |
|---|---|---|
| Absolute check | 92 | 2931.5 |
| | (73.57) | (3.46) |
| Neem oil @10ml.Kg$^{-1}$ | 72 | 2242.7 |
| | (58.05) | (3.35) |
| Nano neem oil @10ml.Kg$^{-1}$ | 60 | 1938.9 |
| | (50.76) | (3.28) |
| Pungam oil 10ml.Kg$^{-1}$ | 72 | 2102.43 |
| | (58.05) | (3.32) |
| Nano pungam oil 10ml.Kg$^{-1}$ | 92 | 2408.54 |
| | (73.57) | (3.38) |
| Mean | 74 | 2324.8 |
| | (59.34) | (3.36) |
| | Treatment | Treatment |
| SE (d) | 3.11 | 0.01 |
| CD (0.05) | 6.94 | 0.04 |

*Values in the parentheses are arcsine transformed
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