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Evaluation of eight plant oils against pulse beetle, *Callosobruchus chinensis* Linnaeus (Bruchidae: Coleoptera) in chickpea under storage conditions

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Abstract
Experiment was conducted under storage conditions to evaluate the efficacy of eight plant oils *viz.*,. *Neem* oil (*Azadirachta indica*), Sesame oil (*Sesamum indicum*), Clove oil (*Syzium aromaticum*), Castor oil (*Ricinus communis*), *Mahua* oil (*Madhuca longifolia*), Coconut oil (*Cocos nucifera*), Mustard oil (*Brassica juncea*), and *Karanj* oil (*Pongamia pinnata*) at 0.25, 0.50, and 1.0 per cent concentrations against pulse beetle, *Callosobruchus chinensis* L. in chickpea. Altogether five parameters namely parent mortality, fecundity, emergence of F_1 progeny, developmental period and weight loss were considered to determine the efficacy. The results revealed that the efficacy of oils was directly proportional to the concentration applied and all the treatments were significantly superior to control. Amongst various oils, the maximum adult mortality (63.89 & 61.89%), minimum oviposition (10.00 & 22.44 eggs/20g seed) and F_1 adult emergence (12.20 & 15.43%), and delayed developmental period (67.67 & 48.84 days) have been observed in the seeds treated *mahua* and *neem* oils, respectively. All the oils have extended protection to chickpea seeds up to 3 months after treatment. Minimum weight loss was found in the chickpea seeds treated with *mahua* (1.78%) and mustard (2.81%) oils followed by *neem* oil (3.15%).

Keywords: *Callosobruchus chinensis*, chickpea, plant oils, pulse beetles, storage conditions

1. Introduction
Amongst the various pulses grown in India, chickpea, *Cicer arietinum* L. ranks first in area and production and accounts for more than 65 per cent of the total global output (around 17 million metric tons). During 2018-19, the production volume of chickpeas in India amounted to over eleven million metric tons followed by Australia (Shahbandesh, 2020) [23]. Major seven states to contribute more than 90 per cent in gram production have been Madhya Pradesh (4.60 Mt), Maharashtra (1.78 Mt), Rajasthan (1.67 Mt), Karnataka (0.72 Mt), Andhra Pradesh (0.59 Mt), Uttar Pradesh (0.58 Mt) and Gujarat (0.37 Mt) (Crops division, 2018) [5].

More than 50 species of insects infest chickpea in field and under storage conditions. Amongst the stored grain pests of pulses, the pulse beetle, *Callosobruchus chinensis* L., is the most important and economic pest (Ahmed *et al*., 2003) [1] and had caused 99.33 per cent of seed damage after 120 days of storage (Venkatesham *et al.*, 2015) [28]. Most of the pulse beetles infest the grains from the field and their hidden infestation could be detected only during storage. Presence of white egg cemented to the surface of grain is the earlier sign of infestation. The newly hatched larva enters the seed and feeds on the inner content throughout its development. The adults emerge through windows in the grain, leaving round holes that are the main evidence of damage. This results in seed-damage and weight-loss making seed unfit for consumption.

Being an internal feeder, it is not advisable to treat the grains with synthetic insecticides. Usage of chemical insecticides under storage is not safe for consumption and insects may develop resistance towards those insecticides (Prasanthi *et al.* 2017) [13]. Pulse beetle had already developed resistance against malathion (Singh and Srivastava, 1983) [25]. Keeping in view of these negative effects of chemicals, many alternatives have been developed to manage these bruchids. These include seed coating by plant extracts/oils (Schoonhoven, 1978) [22], seed drying by bio-electrical thermal device (Monica and Natarajan, 2016) [13], integrating host plant resistance (Pedigo, 1996) [17], controlled atmosphere, release of natural enemies (Doumma *et al*., 2010) [6], irradiation techniques leading to male sterility (Faruki *et al.*, 2007) [7], usage of pheromone traps and insect growth regulators (IGR) (Ukeh *et al*., 2013) [27], etc.

Besides being eco-friendly, the use of plant oils as grain protectants is affordable as the

farmers can make use of locally available raw materials. The bioactive compounds of various plant oils, *viz.*, tetranortriterpenoids (*Neem*), Furanoflavonoid (*Karanj*), ricin and ricinine (castor), sesamin and sesamolin (sesamum), Eugenol (clove), saponin and sapogenin (*Mahua*), Allyl isothiocyanate (Mustard), *etc*. have various effects on insects *i.e*., anti-feedant action, insect growth regulatory activity, inhibits juvenile hormone synthesis, oviposition deterrent, repellant action, reduction of life span of adults and intermediates are formed giving rise to larval-pupal, nymphal-adults, and pupal-adult intermediates (Prasanthi *et al*., 2017) [13]. Keeping in view of these advantages, present study was designed to evaluate the efficacy of different plant oils against pulse beetles.

2. Materials and Methods

2.1 Test insect

Adults of pulse beetles were collected from the pulses godown of Birsa Agricultural University. The insect species was confirmed as *Callosobruchus chinensis* as per the report of Raina (1970) [19]. These initial cultures of bruchids were further multiplied in a big sized jar containing mung bean (*Vigna radiata*) grains. The jar was then covered with a muslin cloth and allowed for free mating and oviposition by the beetles for seven days. The parent beetles were removed and the grains containing eggs were left in that container.

2.2 Collection of chickpea seeds and oils

For the current study, insecticide-free, undamaged seeds of susceptible chikpea variety, C-235 were used. Eight various plant derived oils *viz*., *Neem* oil (*Azadirachta indica*), Sesame oil (*Sesamum indicum*), Clove oil (*Syzium aromaticum*), Castor oil (*Ricinus communis*), *Mahua* oil (*Madhuca longifolia*), Coconut oil (*Cocos nucifera*), Mustard oil (*Brassica juncea*), and *Karanj* oil (*Pongamia pinnata*) were chosen and were procured from the local market and were stored in room temperature maintained at 25±2°C and 65-70 per cent R.H.

2.3 Seed treatment

This study was conducted during 1st August, 2015 to 30th October, 2015 in the laboratory of Department of Entomology, Birsa Agriculture University, Ranchi, India. The experiment has been conducted under uncontrolled conditions intended for domestic storage. Three concentrations of oils *viz*., 0.25, 0.5 and 1 per cent were tested for the experiment. For this, a quantity of 0.25, 0.50 and 1.0 ml of each oil was taken out separately from the bottle with the help of a micro pipette and mixed homogeneously by proper shaking with 100 g of undamaged chickpea grains. Three replicates of 20 gm seeds of each treatment and concentration were measured with the help of a sensitive balance and then transferred separately into 200 ml plastic jars. Ten pairs of less than one day old beetles were separated from the culture and were introduced into each treatment. The mouth of each of the jars was covered with a piece of thin muslin cloth which was held tightly in position by a rubber band. In a similar process ten pairs of freshly emerged adults were introduced into three jars containing untreated seeds which served as the control.

2.4 Observations recorded

i. The cumulative parental mortality up to 7 days was recorded and converted into percentage by the following formula:

$$\text{Per cent adult mortality} = \frac{\text{Total number of adults died}}{\text{Total number of adults introduced}} \text{ X } 100$$

ii. The number of eggs laid was counted twice i.e. at an interval of 7 and 14 days after the introduction of adults. The per cent reduction in oviposition over control was calculated by using the following formula:

$$\text{Per cent reduction in oviposition over control (ROC)} = \frac{E_c - E_t}{E_c} \text{ X } 100$$

E_c = number of eggs laid in control
E_t = number of eggs laid on treated seed

iii. In both the sets of experiments, the F_1 adults emerged in each jar containing the treated seed were counted at the intervals of 60and 90 DAI (Days after Introduction of adults). Adult emergence percentage was calculated by the formula:

$$\text{Per cent adult emergence} = \frac{\text{Total number of adults emerged}}{\text{Total number of eggs deposited}} \text{ X } 100$$

iv. The days taken for the emergence of the first adult in each treatment from the day of oviposition were recorded as the developmental period *i.e*., time taken for egg laying to the adult emergence.

v. The weight loss in the seed was calculated by subtracting the final weight from the initial weight. Then the weight loss was converted into percentage by the following formula:

$$\text{Per cent weight loss} = \frac{\text{Initial weight of grains} - \text{Final weight of grains}}{\text{Final weight of grains}} \text{X } 100$$

2.4 Statistical analysis

The experiment was laid out in a factorial randomized block design with a single factor (oil) at three different levels *viz.* 0.2, 0.50, and 1.0 per cents. Three replicates of each concentration of oil were maintained under storage conditions. Per cent adult mortality, F_1 adult emergence, seed damage and weight loss were subjected to angular transformation whereas number of eggs laid by adults and days taken for the emergence of F_1 adults were subjected to percentage transformation. The transformed data were analyzed using ANOVA and subjected to DMRT (Duncan's Multiple Range test).

3. Results and Discussion

3.1 Parent/Adult mortality

Significantly (p=0.05) higher mortality in adults was recorded (Table 1) in all the treatments when compared to control. Both *mahua* (63.89 %) and *neem* oils (61.89%) were equally capable in bringing about highest parental mortality up to 7 days of treatment followed by mustard oil (51.67%) and this is in conformity with the results of Khaire *et al.* (1987) who reported that mustard and neem oils at higher concentrations were more toxic against parental mortality of pulse beetles. Borthakur (1992) [4] also found that oils like mustard, coconut, groundnut and sunflower oil could totally prevent infestation by *C. analis* in green gram seeds up to 45 days after storage. The least (37.22%) per cent of adult mortality was observed in the seed treated with clove oil but it also

maintained its superiority over control (15%). The effectiveness of clove was reported by Ratnasekhara and Rajpakshee (2012) [21]. No literatures could be come across on effect of *mahua* oil on adult mortality of pulse beetle.

3.2 Fecundity

All the oils tested significantly reduced oviposition by the female beetle on treated seeds and were able to extend its toxicity against the pulse beetle up to 3 months after treatment (Table 2). Clove oil was least effective which might be due to non-sticking and quick drying nature of this oil over the seed coat. *Mahua* oil was the most effective and significantly superior to *neem*, *karan*j, mustard, castor, coconut, and sesame oils. Efficacy in oils increased with the increase in their level of application in case of all oils. The least oviposition was noticed in 1.0 per cent concentration for all oils tested. The variation in effectiveness of various oils in reducing oviposition appears to be due to number of factors like chemical composition, viscosity, smell, *etc*. Slippery seed surface due to oil treatment and odour produced by oils probably inhibit egg laying on the treated seeds.

Earlier, Singh (2003) [24] also reported that there was no oviposition in coconut, mustard, sesamum, *mahua*, *neem*, *karanj*, and castor oils even after 9 months after the release of pulse beetles in pigeon pea. Ali *et al*. (1983) [2] reported that neem, coconut, *mahua*, and sesame at 1.00 ml/ 100 g seeds did not permit adult beetle to lay eggs. More or less similar findings about the bio-efficacies of various oils used were reported by many workers including Bhargava and Meena (2000) [3].

3.3 Emergence F_1 Adult/Progeny

The emergence of adult beetles varied in different treatments (Table 3). All the treatments showed significantly lesser emergence of adult beetles in comparison to control and restricted the adult emergence up to 6 months after treatment. Not even a single adult was noticed in the jars containing the seeds treated with *mahua, neem*, castor, and *karanj* oils even after 60 days after treatment at 1.0 per cent concentration. This might be due to the larvicidal/ ovicidal properties of oils that resulted in the failure of developed adult to emerge out from the seed hole.

The present findings draw considerable support from the work of Ali *et al.* (1983) [2] who reported that *mahua, neem*, mustard, and sesame @ 1.00 per cent had brought 100 per cent grub mortality accounting to zero per cent adult emergence in green gram. Similarly, Singh (2003) [23] observed that there was no adult emergence in *mahua*, coconut, mustard, sesamum, *neem*, *karanj*, and castor oils even after 9 months after the release of pulse beetles in pigeon pea. Insecticidal properties of various vegetable oils on adult emergence of pulse beetles were earlier highlighted by Ibrahim (2012) [8]; Meghwal *et al.* (2007) [12]; and Rani *et al.* (2000) [20].

3.4. Developmental period

Under the present study (Table 4), exposure of *mahua* and *neem* oils could successfully restrict adult emergence completely up to 3 months of storage. Treatment of chickpea seed with *karanj*, castor, mustard also lead to significant delay in the developmental period of pulse beetles. The oils selected for the experiment could statistically enhance the developmental period over control up to three months of storage at all the levels. This may be due to the unfavorable conditions created by the oils inside the jars containing the treated seeds.

The present findings coincides with the work of Ali *et al.* (1983) [2] who observed that *neem*, mustard, *mahua*, and sesame @ 1.00 per cent had brought 100 per cent grub mortality leading to zero per cent adult emergence in green gram and hence there was no question of developmental period. In another study Lal and Raj (2012) [11] observed that there was no adult emergence at 3 ml/ kg dosage of *neem* and castor; hence the developmental period taken by the pulse beetles was zero. Namdev *et al.* (2014) [14] found neem oil as an effective grain protectant and had shown maximum increase in developmental period at 3 ml/ kg seed dose followed by mustard oil.

3.5 Weight loss

All the oils could bring down the weight loss significantly over control (Table 5) all throughout the experimental period of 3 months in storage. Minimum weight loss was noticed in mustard and *mahua* oils which was statistically followed by other oils. Maximum weight loss was recorded in untreated control.

The present observations are in accordance with views of Kumari *et al.* (1990) [10] who revealed that mustard, *til*, *neem*, and *mahua* oils as grain protectants at one per cent concentration proved equally effective for reduction in the percentage of damaged grains by number as well as weight bases. Parsai *et al.*, (1990) [16] have also reported lower weight loss with mustard at 1.0 % concentration. Srinivasan (2008) [26] also observed that *neem* oil @ 5 and 10 ml/kg are highly effective against pulse beetle on pigeon pea seeds up to 9 months. Bhargava and Meena (2000) [3] reported that castor and neem oils @ 3 ml/kg had given 100 per cent control in terms of reduction of weight loss after 120 DAT. Results showed that the oils do effect the protoplasm of the egg and larval feeding, hence oils proved to be most effective and safest method to control the pulse beetle for stored pulses.

4. Conclusion

The results of the storage experiment have shown that the chickpea grains/seeds can effectively be saved from the ravages of the pulse beetles, *C. chinensis*, by mixing the plant oils at 1.0 per cent (mg/ kg seed) concentration. For this to be achieved, proper admixing of oil and the grains should be done to ensure that uniform coating. The use of plant oils should be encouraged on small scale storage, as the cost of application of these oils is low when compared with the losses incurred in untreated seeds.

5. Acknowledgement

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Table 1: Effect of various oils on adult mortality of pulse beetle up to 7 DAI

Sl. No.	Treatment	Per cent adult mortality at the dosage (%)			Mean
		0.25	0.50	1.00	
1	Neem oil	51.67 (45.97)*	60.00 (50.77)	74.00 (59.40)	61.89a
2	Sesame oil	33.33 (35.11)	35.00 (36.24)	51.67 (45.97)	40.00d
3	Clove oil	31.67 (34.23)	35.00 (36.24)	45.00 (42.12)	37.22d
4	Castor oil	40.00 (39.21)	41.67 (40.18)	56.67 (48.84)	46.11c
5	Mahua oil	53.33 (46.92)	63.33 (51.76)	75.00 (60.08)	63.89a
6	Coconut oil	38.33 (38.24)	40.00 (39.21)	55.00 (47.88)	44.44c
7	Mustard oil	45.00 (42.11)	51.67 (45.97)	58.33 (49.83)	51.67b
8	Karanj oil	43.33 (41.16)	50.00 (45.00)	58.33 (49.80)	50.55b
9	Mean	42.08	47.08	59.25	
	Control				15.00e (22.60)
	Oil (O)	Dose (D)	T X D	Control vs Treatment	
S.Em±	1.12	0.68	-	1.98	
CD (P=0.05)	3.18	1.95	-	3.97	
CV (%)	7.65				

*Figures in the parenthesis are angular transformed values

Table 2: Effect of various oils on fecundity of pulse beetle

Sl. No.	Treatment	Average fecundity per 20 gm seed at the dosage (%)									
		7 DAI					14 DAI				
		0.25	0.50	1.00	Overall mean	ROC (%)	0.25	0.50	1.00	Overall mean	ROC (%)
1	Neem oil	11.23 (3.36)*	9.00 (3.00)	7.66 (2.75)	9.30d	85.98	32.66d (5.71)*	24.33d (492)	10.33d (3.20)	22.44d	78.14
2	Sesame oil	17.66 (4.20)	14.00 (3.74)	12.33 (3.50)	14.67g	77.88	48.00g (6.92)	40.00g (6.32)	32.66 g (5.71)	40.22f	60.82
3	Clove oil	19.00 (4.35)	15.67 (3.95)	14.66 (3.82)	16.44h	75.21	59.33h (7.70)	50.66h (7.11)	45.33h (6.73)	51.77g	49.57
4	Castor oil	9.66 (3.10)	7 .00 (2.64)	5.66 (2.37)	7.44b	88.78	30.00c (5.47)	26.66f (5.16)	18.00 f (4.24)	24.89e	75.75
5	Mahua oil	9.00 (3.00)	6.00 (2.44)	4.00 (2.00)	6.33a	90.46	15.00a (3.86)	9.33a (3.04)	5.66a (2.36)	10.00a	90.26
6	Coconut oil	14.33 (3.78)	12.00 (3.46)	11.66 (3.41)	12.66f	80.91	35.00f (5.91)	25.00e (5.00)	14.33e (3.78)	24.78e	75.86
7	Mustard oil	12.67 3.55)	10.00 (3.15)	9.00 3.00)	10.55e	84.09	33.00e (5.74)	22.33c (4.72)	9.66c (3.10)	21.66c	78.90
8	Karanj oil	11.67 (3.41)	8.33 2.88)	6.33 (2.50)	8.77c	86.77	29.33b (5.41)	19.33b (4.39)	8.66b (2.93)	19.11b	81.38
	Mean	13.15	10.25	8.91			35.29	27.21	18.07		
	Control (C)				66.33i (8.14)					102.66 h (10.13)	
		Oil (T)	Dose (D)	T X D	C vs T		T	D	T X D	C Vs T	
S.Em ±		0.07	0.04		0.84		0.06	0.04	0.11	0.12	
CD(P=0.05)		0.20	0.12	NS	1.69		0.19	0.12	0.33	0.24	
CV (%)		6.06					3.95				

*Figures in parenthesis are square root transformed values

Table 3: Effect of various oils on adult emergence of pulse beetle on chickpea

Sl. No.	Treatment	Average per cent adults emerged at the dosage (%)							
		60 DAI				90 DAI			
		0.25	0.50	1.00	Mean	0.25	0.50	1.00	Mean
1	Neem oil	8.19b (16.55)*	5.53 (13.40)	0.00a (0.00)	4.57b	30.83b (33.60)	15.47ab (22.74)	0.00a (0.00)	15.43 a
2	Sesame oil	26.84d (31.13)	22.77 (28.6)	23.47c (28.88)	24.36ef	64.22d (52.92)	63.15e (52.86)	56.02e (48.65)	61.13 e
3	Clove oil	34.24e (35.81)	30.88 (33.71)	25.10c (29.97)	30.07	80.34e (63.72)	79.02f (64.00)	73.65f (59.25)	77.67 f
4	Castor oil	22.37d (28.10)	10.15 (18.46)	0.00a (0.00)	10.84c	48.59c (44.18)	36.60c (37.13)	24.11c (28.59)	36.43 c
5	Mahua oil	0.00a (0.00)	0.00 (0.00)	0.00a (0.00)	0.00a	22.66a (28.24)	13.93a (21.90)	0.00a (0.00)	12.20 a
6	Coconut oil	26.39d (30.71)	18.44 (25.35)	11.39b (19.6)	18.74d	54.81c (49.69)	52.17d (46.32)	41.69d (46.09)	49.56 d
7	Mustard oil	17.22c (24.49)	14.99 (22.70)	27.73c (31.73)	19.98d	65.00d (53.94)	46.65d (43.05)	45.45d (42.36)	52.37 d
8	Karanj oil	11.32b (19.66)	8.50 (16.82)	0.00a (0.00)	6.61b	37.49b (37.76)	22.91b (28.29)	15.50b (20.92)	25.30 b
	Mean	18.32	13.91	10.96		50.49	41.24	32.05	
	Control (C)				78.62 (62.51)	-	-	-	91.25 (72.95)
		Oil (T)	Dose (D)	T X D	C vs T	T	D	T X D	C Vs T
S.Em ±		0.95	0.58	1.64	1.67	1.54	0.94	2.67	2.70
CD(P=0.05)		2.69	1.65	4.67	3.35	4.37	2.68	7.58	5.43
CV (%)		13.72				11.60			

*Figures in parenthesis are angular transformed values

Table 4: Effect of various oils on developmental period of pulse beetles on freshly treated chickpea seed

Sl. No.	Treatment	Average number of days taken for emergence of F_1 adults at the dosage (%)			Mean
		0.25	0.50	1.00	
1	Neem oil	45.67b (6.75)*	52.00b (7.21)	0.00a (0.00)	48.84b
2	Sesame oil	30.33g (5.51)	33.33g (5.77)	39.00f (6.24)	34.22g
3	Clove oil	28.00h (5.29)	32.67h (5.71)	37.00g (6.08)	32.55h
4	Castor oil	34.67e (5.89)	39.67e (6.30)	63.00c (8.06)	45.78d
5	Mahua oil	65.00a (8.06)	70.33a (8.39)	0.00a (0.00)	67.67a
6	Coconut oil	34.33f (5.86)	38.67f (6.22)	41.33e (6.43)	38.11f
7	Mustard oil	37.67c (6.14)	40.00d (6.32)	43.67d (6.61)	40.45e
8	Karanj oil	36.33d (6.03)	42.33c (6.50)	65.00b (8.06)	47.00c
	Mean	39.00	43.63	48.17	
9	Control				24.66 (4.97)
	Oil (T)	Dose (D)	T X D	Control vs Treatment	
SEm±	0.05	0.03	0.08	0.09	
CD (P=0.05)	0.14	0.08	0.24	0.17	
CV %	2.6				

*Figures in parenthesis are square root transformed values

Table 5: Effect of various oils on per cent weight loss by pulse beetles on chickpea at 90 DAI

Sl. No.	Treatment	Per cent weight loss at the dosage (%)			Mean
		0.25	0.50	1.00	
1	Neem oil	4.58 b (12.37)*	3.24 b (10.37)	1.63 a (7.30)	3.15 bc
2	Sesame oil	6.92 d (15.25)	4.65 c (12.45)	2.79 (9.59)	4.78 d
3	Clove oil	9.02 e (17.45)	8.97 d (17.42)	5.24 (13.19)	7.74 e
4	Castor oil	2.97 a (9.94)	1.97 a (8.07)	1.20 a (6.28)	2.05 a
5	Mahua oil	2.58 a (9.25)	1.79 a (7.45)	0.97 a (5.62)	1.78 a
6	Coconut oil	6.22 c (14.46)	3.58 bc (10.91)	1.65 a (7.36)	3.82 c
7	Mustard oil	4.63 b (12.39)	2.92 ab (9.83)	0.88 a (5.34)	2.81 b
8	Karanj oil	5.23 bc (13.22)	3.97 bc (11.48)	2.20 (8.51)	3.80 c

	Mean	5.27	3.88	2.07	
9	Control				25.79 (30.51)
	Oil (T)	Dose (D)	T X D	Control vs Treatment	
SEm±	0.25	0.15	0.43	0.44	
CD (P=0.05)	0.70	0.43	1.22	0.87	
CV (%)	6.50				

*Figures in parenthesis are arcsine √percentage transformed values

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