Article AR2416

DOI: HTTPS://DOI.ORG/10.23910/1.2021.2416

International Journal of Bio-resource and Stress Management

December 2021

Crossref

Print ISSN 0976-3988

Online ISSN 0976-4038

IJBSM 2021, 12(6):679-686

Research Article *Stress Management*

Evaluation of Some Botanicals against *Callosobruchus chinensis* L. Infesting Stored Chickpea Seeds and Bio-Chemical Analysis of Used Botanicals

Tumma Mounika, S. K. Sahoo* and D. Chakraborty

Dept. of Entomology, Uttar Banga Krishi Vishwavidyalaya, Pundibari. Coochbehar, West Bengal (736 165), India

Open Access

Corresponding Author

S. K. Sahoo

e-mail: shyamalsahoo@gmail.com

Citation: Mounika et al., 2021. Evaluation of Some Botanicals against *Callosobruchus chinensis* L. Infesting Stored Chickpea Seeds and Bio-Chemical Analysis of Used Botanicals. International Journal of Bio-resource and Stress Management 2021, 12(6), 679-686. HTTPS://DOI.ORG/10.23910/1.2021.2416.

Copyright: © 2021 Mounika et al. This is an open access article that permits unrestricted use, distribution and reproduction in any medium after the author(s) and source are credited.

Data Availability Statement: Legal restrictions are imposed on the public sharing of raw data. However, authors have full right to transfer or share the data in raw form upon request subject to either meeting the conditions of the original consents and the original research study. Further, access of data needs to meet whether the user complies with the ethical and legal obligations as data controllers to allow for secondary use of the data outside of the original study.

Conflict of interests: The authors have declared that no conflict of interest exists.

Abstract

A study was carried out to evaluate the bio-efficacy of some botanicals against *Callosobruchus chinensis* L. in stored chickpea (Variety: Anuradha) in the year 2018–20. Different botanicals like Neem (*Azadirachta indica*), Melia (*Melia azedarach*), Datura (*Datura stramonium*) and Tulsi (*Ocimum sanctum*) were used. Among all the botanicals Neem leaf powder @ 6% performed better with minimum egg laying (64 eggs 5 females^{-1}) compared to the other botanicals except the standard check with Deltamethrin @ 0.04%. Thereafter, 6% Melia leaf powder and 8% Datura leaf powder recorded 87.50 eggs 5 females^{-1} and 91.25 eggs 5 females^{-1}, respectively. With regard to adult mortality, the best result was also obtained from the treatment Neem leaf powder @6% with 96.67% mortality in 5th day. Following the same trend, 6% Melia leaf powder and 8% Datura leaf powder exhibited 90% and 83.33% adult mortality, respectively, in 5th day. The estimation of total phenols and total antioxidants (IC-50) contents in Neem leaf powder (345.69 mg g^{-1} and 207.77 µg ml^{-1}), Melia leaf powder (273.40 mg g^{-1} and 383.68 µg ml^{-1}) and Datura Leaf Powder (213.62 mg g^{-1} and 405.77 µg ml^{-1}) also confirms the findings of the bio-efficacy trial of the botanicals. Tulsi leaf powder @5% was least efficacious both in terms of egg laying by the females as well as adult mortality. These botanicals are locally available, economic, bio-degradable and safe to the environment. Therefore, they may be fitted in the Integrated Pest Management strategies against stored grain pests as seed protectants.

Keywords: Adult mortality, antioxidants, botanicals, *Callosobruchus chinensis*, chickpea, egglaying, phenols

1. Introduction

Chick pea (*Cicer arietinum*) is an annual legume crop which belongs to the family, Fabaceae (Anonymous, 2018a and 2018b). It was originated from South East Turkey. It is ranked third in terms of production all over the world with a mean annual production of over 14.25 million tons where 9.94 million tons of the production was recorded from India. India was responsible for 70% of global chickpea production (Anonymous, 2020). Matured raw seeds of Chick pea consist of 20.47 g of proteins, 62.95 g of carbohydrates and 6.04 g of fats per 100 g of seed weight (Anonymous, 2019). It is included in several recipes using its tender plant parts as vegetables as well as the seeds (both the green and the matured ones) for preparing spicy and tasty dishes. The seeds are ground to prepare 'Sattu' and 'Besan' which are also very popular healthy diets in our country. Being a pulse crop, it fixes atmospheric nitrogen in the root

Article History

RECEIVED on 10th June 2021 RECEIVED in revised form on 29th October 2021 ACCEPTED in final form on 17th December 2021

system which enables to meet its own nitrogen requirement (Sowmya and Kumar, 2017). Besides, byproducts of this pulse crop were used as fuel and cattle feed. Naturally, Chickpea is considered as one of the important components for sustainable agriculture. Chickpea crop as well as seeds are subjected to various stresses, of which insect pests and diseases are the major hindrances in achieving its optimum yield. Among the insect-pests damaging chickpea, pod borer is the most severe in case of field level while, bruchids cause damage in storage (Gahukar and Reddy, 2018). *Callosobruchus* spp. (Fam. Bruchidae) belonging to the order Coleoptera, are well known primary and most destructive pest of stored legume seeds. In India, these are commonly named "pulse beetle" (Rahman et al., 1942 and Vasu and Nikita, 2020). They cause damage to grains which are stored at a temperature of 30°C and 70% RH as these conditions favour the development of this pest (Raina, 1970). Chickpea suffers high qualitative and quantitative losses due to the attack of pulse beetle. Mookherjee et al. (1970) reported 32-64% infestation of *Callosobruchus* spp. in leguminous seeds as well as 3% in oilseeds. This pest was found to damage 50-60% grains in stored condition after 6 months of traditional storage (Caswell, 1973). Sometimes, even in severe cases of infestation, the damage can reach even up to 100% (Pruthi and Singh, 1950). Generally, the storage insect pests are controlled by the application of fumigants and synthetic insecticides. Their indiscriminate use in storage has led to many problems like environmental pollution, insect resurgence, toxic residues on food grains (Alemayehu and Getu, 2015) and increased cost of application. As the pulse beetles are internal feeders, mixing insecticides with the seeds as well as fumigating the seeds increase the chance of residual toxicity. While, the botanical insecticides not only possess bioactive chemicals (Wink, 1993) but they are target specific, non-toxic to human and beneficial organisms and less prone to insect resurgence and resistance also. That is why these insecticides are the alternative to chemical insecticides in relation to the management of storage insects. The use of these plant-based insecticides in the storage, therefore, offers a desirable solution. Therefore, the present study was carried out to examine the bio-efficacy of different plant powders on the egg laying capacity and adult mortality of *C. chinensis* infesting chickpea seeds in storage and to estimate the total phenols and antioxidants present in the botanicals.

2. Materials and Methods

2.1. Study site

A laboratory experiment was conducted in the Department of Agricultural Entomology, Uttar Banga Krishi Viswavidyalaya, Pundibari, West Bengal, India to study the efficacy of different botanicals on egg laying and adult mortality of *Callosobruchus chinensis* on chickpea during 2018–20. Besides, the study to estimate the phenols and antioxidants in the botanicals was conducted in the Department of Biochemistry, Uttar Banga Krishi Viswavidyalaya, Pundibari.

2.2. Maintenance and mass rearing of host insect

Mass culturing of *C. chinensis* was done in heat sterilized (hot air oven at 80°C for 2 hrs) and disinfected (formalin 1%) chickpea seeds (Variety: Anuradha) of weight 1 kg in a glass jar. To obtain the pure mass culture of the desired species, *Callosobruchus chinensis* adults from the infected stock were collected and released in the glass jar. The mouth of the container was covered by muslin cloth. After 7 days all the adults were removed and egg laid seeds were maintained at the required temperature and humidity.

The newly emerged adults were used for the experimental purpose. Fresh culture on new seeds of chickpea was raised regularly for multiplication of the culture and to avoid the fungal infection. During winter, the rearing jars were kept in a BOD incubator at 27–30°C temperature and 70–75% relative humidity for maintenance of culture.

2.3. Preparation of the seed for bio-efficacy study

The chickpea seed (Variety: Anuradha) was collected from the Pulses and Oilseeds Research Station, Government of West Bengal, Berhampore, Murshidabad, West Bengal. The seeds were heated and disinfected with formalin (1%). Afterward the chickpea seeds of 25 g was weighted and kept in plastic container for the experiment of adult mortality and egg laying capacity of *Callosobruchus chinensis*.These experiments each were replicated thrice.

Then selected botanicals were mixed with the seeds properly and 5 pairs of newly emerged adult (males and females) beetles which were reared were released in each container based on the experiments carried out. While releasing, the male and females were identified by the antennae which are often pectinate in males and serrate in females and the size of the male was small as compared to female.

2.4. Selection and preparation of treatment material

Four botanical plants namely Neem (*Azadirachta indica* A. Juss), Dharek (*Melia azedarach* L.), Datura (*Datura stramonium* L.) and Tulsi (*Ocimum sanctum* L.) were selected and different plant parts of the botanicals were used for evaluating the efficacy against pulse beetle, *Callosobruchus chinensis*. All the Botanical parts were collected in the university premises. The fresh leaves from Neem, Dharek, Datura and Tulsi and the barks of Neem and Dharek were collected and dried in shade for about 10–15 days. In the case of Datura and Tulsi, the ripened fruits and the matured perianth were also collected for separating the seeds. All the plant parts were powdered with the help of a grinder. Fine powders, after passing through mesh sieves, were utilized by mixing with the chickpea seeds for the bio-efficacy experiment (Alemayehu and Getu, 2015).

2.5. Evaluation of botanicals at different doses against pulse beetle

The experiment was carried out in the laboratory with 14

treatments including untreated control and three replications. Botanical powders were taken in the container having sterilized chickpea seeds at different doses according to the treatment (Table 1) and thoroughly mixed as seed protectant by shaking the plastic containers. Deltamethrin 2.5 WP was utilized in one treatment as a standard check for comparison. Then freshly emerged five pairs of pulse beetle adults were released in each plastic container. The containers were closed by muslin cloth tightly and secured by a rubber band (Satyavir, 1983).

Table 1: Treatment details for the management of pulse beetle

Treatment	Name of the botanicals	Parts used	Conc. (%)
T_1	Neem (*Azadirachta indica*)	Leaves	6%
T_2	Neem (*Azadirachta indica*)	Bark	2%
T_3	Neem (*Azadirachta indica*)	Leaves+Bark	6%+2%
T_4	Dharek (*Melia azedarach*)	Leaves	6%
T_5	Dharek (*Melia azedarach*)	Bark	2%
T_6	Dharek (*Melia azedarach*)	Leaves+Bark	6%+2%
T_7	Datura (*Datura stramonium*)	Leaves	8%
T_8	Datura (*Datura stramonium*)	Seeds	8%
T_9	Datura (*Datura stramonium*)	Leaves+Seeds	8%+8%
T_{10}	Tulsi (*Ocimum sanctum*)	Leaves	5%
T_{11}	Tulsi (*Ocimum sanctum*)	Seeds	2%
T_{12}	Tulsi (*Ocimum sanctum*)	Leaves+Seeds	5%+2%
T_{13}	Standard Check (Deltamethrin 2.5 WP)	-	0.04%
T_{14}	Untreated control	-	-

2.6. Evaluating the efficacy of different botanical powders on different parameters

2.6.1. Efficacy of different botanical powders on egg laying capacity of C. chinensis

The efficacy of different botanical powders was evaluated in terms of the egg laying capacity of adult females of pulse beetle. Chick pea seeds amounting 25 g were taken in each plastic container and treated with botanicals as per the treatments (Table 1) and each treatment was replicated thrice. Then 5 pairs of freshly emerged pulse beetle adults were released in each plastic container. The oviposition period of pulse beetle generally ranges between 7 to 10 days (Ghosal, 2003). Therefore, seven (07) days after the treatment all the dead and alive adults were removed from all the treatments. In each treatment 100 seeds were taken randomly and the total number of eggs were counted and recorded.

2.6.2. Efficacy of different botanical powders on percentage of adult mortality of C. chinensis:

Likewise, 25 g of chickpea seeds in plastic containers were taken and another methodology was followed. The biology of *C. chinensis* has indicated that the adults are short lived for about 6-10 days (Ghosal, 2003; Alemayehu and Getu, 2015) and therefore the observation for adult mortality was recorded upto 5 days. Number of dead adults in chickpea seeds were counted at an interval of 24 hours upto 5 days and then the dead adults were removed from the containers. The percent adult mortality was calculated by the following formula (Laizu, 2009).

% Adult mortality =(No. of dead adults/ Total No. of adults)×100

2.7. Estimation of the total antioxidants as well as phenols present in the selected botanicals

The efficacy of the botanicals depends on the total phenol compounds and total antioxidants present in them. This was analysed in each of the treatments from (T_1 to T_{12}) following Folin-Ciocalteu reagent and DPPH antioxidant assay method by using Spectro-photometer. The carrying capacity of antioxidants and phenols is directly proportional to the efficacy of the botanical formulations (Ho, 1992).

2.8. Statistical analysis

The experiments for the research work were formulated and the data was recorded and statistically analysed according to the Completely Randomized Design (CRD). Duncan Multiple Range Test (DMRT) was also done after transformation of the data to study the difference among the treatments used for evaluation of botanicals against pulse beetle. The biochemical analysis was performed twice on different botanicals and the total phenols content was quantified by using a linear regression formula i.e., "Y=0.0189X- 0.0658", R^2=0.9991 which was derived from different concentration of gallic acid which implicates good linearity. For the Antioxidants activity, linear regression formula was obtained by plotting the percentage of DPPH scavenging versus concentration of samples in Microsoft excel 2007. The concentration of the sample necessary to decrease the DPPH concentration by 50% was obtained by interpolation from linear regression curve and denoted IC-50 value (µg ml^{-1}).

3. Results and Discussion

3.1. Efficacy of different botanical powders on egg laying capacity of C. chinensis

The number of eggs laid by five pairs of pulse beetle,

Callosobruchus chinensis in different botanicals treated chickpea seeds was studied during July 2019 and mentioned in Table 2. The results revealed that minimum eggs (64 eggs 5 females^{-1}) were laid by the adult females of *C. chinensis* in (T_1) Neem leaf powder@6% treated chickpea seeds. Neem leaf powder@ 6% performed better result compared to the other botanicals except for the (T_{13}) Standard check, Deltamethrin @0.04% where the number of eggs reported was 22.83 eggs 5 females^{-1}. Thereafter, (T_4) Melia leaf powder @ 6% resulting 88.67 eggs 5 females^{-1} and (T_7) Datura leaf powder @ 8% 92.00 eggs 5 females^{-1}. All the other botanicals also performed well compared to (T_{14}) untreated control where the number of the eggs was 403.00 eggs 5 females^{-1}.

This experiment on the efficacy of botanicals on the egg laying of pulse beetle was repeated once again during August 2019 for validation of the result found in the aforesaid trial and

Table 2: Efficacy of different botanicals in terms of impact on the egg laying of pulse beetle, *Callosobruchus chinensis* in chickpea seeds during July, 2019

Treatments	Dose	Eggs laid by five pairs of *C. chinensis* in 7 days		Pooled mean value
		First week observation	Second week observation	
T_1- Neem LP#	6%	63.67* (7.98)^g	64.33 (8.01)^i	64.00
T_2- Neem BP	2%	189.33 (13.76)^d	188.67(13.73)^{de}	189.00
T_3- Neem LP+BP	6%+2%	104.33 (10.21)^f	109 (10.43)^g	106.67
T_4-Melia LP	6%	90.67 (9.52)^f	86.67 (9.30)^h	88.67
T_5-Melia BP	2%	281.67(16.78)^{bc}	277.67(16.65)^{bc}	279.67
T_6-Melia LP+BP	6%+2%	176.00 (13.26)^d	172.67 (13.14)^e	174.34
T_7-Datura LP	8%	95.33 (9.76)^f	88.67 (9.41)^h	92.00
T_8-Datura SP	8%	138.33 (11.76)^e	142.33 (11.93)^f	140.33
T_9-Datura LP+SP	8%+8%	129.33 (11.37)^e	134.00 (11.57)^f	131.67
T_{10}-Tulsi LP	5%	297.33 (17.23)^b	301.33 (17.35)^b	299.33
T_{11}-Tulsi SP	2%	250.67 (15.83)^c	257.67 (16.05)^c	254.17
T_{12}-Tulsi LP+SP	5%+2%	203.00 (14.18)^d	210.33 (14.46)^d	206.67
T_{13}- Deltamethrin 2.5WP	0.04%	22.33 (4.72)^h	23.33 (4.82)^j	22.83
T_{14}- Untreated Control	-	405.00 (20.12)^a	401 (20.02)^a	403.00
SEm±	-	0.34	0.333	-
CD (*p*=0.05)	-	0.986	0.966	-

*Mean of all the three replications; Figures in parenthesis are square root transformed values; Parentheses followed by same letters are statistically at par by DMRT; LP: Leaf Powder; BP: Bark Powder, SP: Seed powder

found that among all the botanicals minimum eggs were laid in the chickpea seeds treated with the neem leaf powder@6% with 64 eggs 5 females^{-1} (Table 3).

From the aforementioned results it can be concluded that among the botanicals, (T_1) Neem leaf powder @6% was best which registered minimum number of eggs laid by *Callosobruchus chinensis.* The tested botanicals as per their efficacy are ranked as NLP >MLP >DLP >NLP+BP >DLP+SP > DS P>MLP+BP>NBP>TLP+SP>TSP>MBP>TLP.

Although the lowest oviposition was recorded in the chickpea seeds treated with Deltamethrin, to avoid residual hazards of chemical pesticides, botanicals should be used for management of the insect-pests particularly in seed storage. The results of the present research works are in good conformity with Kaur (2017) who reported that neem leaf powder reduced egg laying effectively. Alemayehu and Getu (2015) noted that the number of eggs laid on the seeds treated with Neem leaf powder was 63 eggs 50 grains^{-1} after 8 days and it was much effective to reduce the egg laying. The effect of the leaf powder of Neem and Datura on the reduction of egg laying capacity of pulse beetle was also recorded by Misra (2000).

The reduction of egg laying by the pulse beetle in chickpea seeds treated with Neem leaf powder might be due to the secondary metabolites i.e., Azadiractin present in neem (Koul, 2004). Whereas the chemical substance like tetranortriterpeniods, melianone, meliantriol and nimbolidin-A present in Dharek (Melia) might be responsible for the reduction of oviposition by the pulse beetle (Saxena, 1998; Isman, 2001).

3.2. Efficacy of different botanical powders on adult mortality of C. chinensis

The results of the experiment to study the efficacy of different botanicals on adult mortality clearly indicated that all the treatments revealed a wide variation in mortality compared to untreated control. It is revealed from Table 4 that among all the botanicals higher percentage of mortality (96.67%)

Table 3: Efficacy of different botanicals in terms of impact on the egg laying of pulse beetle, *Callosobruchus chinensis* in chickpea seeds during August, 2019

Treatments	Dose	Eggs laid by five pairs of *C. chinensis* in 7 days		Pooled mean value
		First week observation	Second week observation	
T_1- Neem LP#	6%	62.33 (7.89)^{i}	65.67 (8.07)^{j}	64.00
T_2- Neem BP	2%	190.67 (13.81)^{de}	191.00 (13.82)^{f}	190.84
T_3- Neem LP+BP	6+2%	111.67 (10.57)^{g}	121.00(11.00)^{h}	116.34
T_4-Melia LP	6%	87.00 (9.32)^{h}	85.67 (9.25)^{i}	86.34
T_5-Melia BP	2%	279.33 (16.70)^{bc}	280.00(16.73)^{c}	279.67
T_6-Melia LP+BP	6+2%	175.67 (13.25)^{e}	179.33 (13.39)^{f}	177.50
T_7-Datura LP	8%	91.33 (9.55)^{h}	89.67 (9.46)^{i}	90.50
T_8-Datura SP	8%	140.67 (11.86)^{f}	147.33 (12.14)^{g}	144.00
T_9-Datura LP+SP	8+8%	135.33 (11.63)^{f}	135.33 (11.63)^{gh}	135.33
T_{10}-Tulsi LP	5%	304.00 (17.43)^{b}	314.33 (17.73)^{b}	309.17
T_{11}-Tulsi SP	2%	257.67 (16.04)^{c}	256.00 (16.00)^{d}	256.84
T_{12}-Tulsi LP+SP	5+2%	213.67 (14.59)^{d}	210.00 (14.49)^{e}	211.84
T_{13}- Deltamethrin 2.5WP	0.04%	24.33 (4.93)^{j}	21.60 (4.65)^{k}	23.00
T_{14}- Untreated Control	-	426.33 (20.64)^{a}	417 (20.42)^{a}	421.67
SEm±	-	0.32	0.232	-
CD (*p*=0.05)	-	0.928	0.671	-

*Mean of all the three replications; Figures in parenthesis are square root transformed values; Parentheses followed by same letters are statistically at par by DMRT; LP: Leaf Powder; BP: Bark Powder, SP: Seed powder

Table 4: Efficacy of different botanicals in terms of mortality of *Callosobruchus chinensis* adult beetles in chickpea seeds during July, 2019

Treatments	Dose	Adult mortality (%) of *Callosobruchus chinensis* after					Pooled mean
		Day 1	Day 2	Day 3	Day 4	Day 5	
T_1	6%	*26.67 (31.00)^{ab}	40.00 (39.23)^{ab}	60.00 (50.85)^{ab}	80.00 (63.93)^{ab}	96.67 (83.00)^{ab}	60.67
T_2	2%	13.33 (21.14)^{bcd}	23.33 (25.50)^{bcd}	43.33 (41.07)^{bc}	63.33 (52.78)^{bcd}	73.33 (59.00)^{cd}	43.33
T_3	6%+2%	13.33 (21.14)^{bcd}	30.00 (33.21)^{ab}	50.00 (45.00)^{abc}	63.33 (52.86)^{bcd}	80.00 (63.93)^{bc}	47.33
T_4	6%	20.00 (26.07)^{bc}	36.67 (37.14)^{ab}	53.33 (46.92)^{abc}	66.67 (55.07)^{bc}	86.67 (68.86)^{bc}	52.67
T_5	2%	6.67 (11.56)^{de}	13.33 (18.57)^{bcd}	33.33 (35.01)^{bc}	46.67 (43.08)^{bcd}	63.33 (52.78)^{cd}	32.67
T_6	6%+2%	10.00 (18.43)^{bcd}	23.33 (28.78)^{bc}	43.33 (41.07)^{bc}	63.33 (52.78)^{bcd}	76.67 (61.22)^{c}	43.33
T_7	8%	6.67 (13.64)^{cde}	26.67 (30.29)^{ab}	40.00 (35.20)^{bc}	63.33 (53.36)^{bcd}	80.00 (68.43)^{bc}	43.33
T_8	8%	20.00 (25.37)^{bc}	26.67 (27.42)^{bc}	46.67 (42.99)^{bc}	40.00 (35.20)^{cd}	76.67 (61.22)^{c}	42.00
T_9	8%+8%	20.00 (26.07)^{bc}	23.33 (25.50)^{bcd}	30.00 (29.43)^{bcd}	63.33 (52.78)^{bcd}	83.33 (66.14)^{bc}	44.00
T_{10}	5%	0 (4.05)^{e}	3.33 (8.85)^{cd}	3.33 (8.85)^{d}	30.00 (33.21)^{cd}	56.67 (48.85)^{cd}	18.67
T_{11}	2%	3.33 (8.85)^{de}	13.33 (21.14)^{bcd}	26.67 (30.79)^{bcd}	33.33 (31.35)^{d}	63.33 (52.78)^{cd}	28.00
T_{12}	5%+2%	10.00 (18.43)^{bcd}	16.67 (21.28)^{bcd}	20.00 (23.49)^{cd}	53.33 (46.92)^{bcd}	46.83 (39.21)^{de}	29.37
T_{13}	0.04%	40.00 (39.15)^{a}	60.00 (50.85)^{a}	83.33 (66.14)^{a}	96.67 (83.00)^{a}	100 (88.72)a	76.00
T_{14}	-	0 (4.05)^{e}	0 (4.05)^{d}	3.33 (8.85)^{d}	3.33 (8.85)^{d}	23.33 (28.78)^{e}	6.00
SEm±		3.95	6.72	7.05	6.97	6.37	-
CD (*p*=0.05)	-	11.43	19.48	20.42	20.18	18.45	-

was observed in the case of the seeds treated with (T_1) Neem leaf powder followed by (T_4) Melia leaf powder with 86.67%, (T_9) Datura leaf powder+seed powder 83.33%. The same experiment was conducted once again to compare the results in the month of August 2019 and it was found that maximum percentage of mortality was observed in the case of (T_1) Neem leaf powder treated seeds with 96.67% followed by (T_4) Melia leaf powder and (T_7) datura leaf powder with 93.33% and 86.67%, respectively (Table 5).

The results of the present study in both (Tables 4 and 5) indicates that, among all the tested botanicals, Neem leaf powder (NLP) had promising effects on adult mortality, followed by Melia leaf powder (MLP) and Datura Leaf powder (DLP). In terms of adult mortality of pulse beetle the botanicals were ranked as NLP>MLP>DLP>NLP+BP>DLP+SP>DSP> MLP+BP>NBP>TLP+SP>TSP>MBP>TLP.

The aforementioned findings are in agreement with Parmar et al. (2018) who reported that Neem leaf powder was more effective against the adults of C*. chinensis*. Karar (1999) also found that Neem leaf powder causing 90% mortality of pulse beetle within 72 hrs.

3.3. Estimation of the total antioxidants as well as phenols present in the selected botanicals

The biochemical analysis was done to study the total phenol and antioxidant properties of the used botanicals to establish the outcome of the bio-efficacy trials against adult pulse beetles and found that higher amount of phenol content and antioxidant activity (IC-50) were measured in the Neem leaf powder with (345.69 mg g^{-1} and 207.77 µg ml^{-1}) followed by Melia leaf powder with (273.40 mg g^{-1} and 383.68 µg ml^{-1}) and Datura leaf powder with (213.62 mg g^{-1} and 405.77 µg ml^{-1}), respectively.

The higher content of phenol and lower IC-50 values indicates the higher antioxidants properties in the botanicals, thereby the respective botanical performed well against pulse beetles. The order of the botanicals in view of phenolic content and antioxidant activity (IC-50) value was NLP>MLP>DLP>NLP+BP >DLP+SP>DSP>MLP+BP>NBP>TLP+SP>TSP>MBP>TLP.

Botanicals represents the insecticidal properties through repellence, antifeedant effect, oviposition deterrent and metamorphosis inhibition (Dhaliwal and Koul, 2007).

Generally, the cells in the insect body release harmful free radicals at the time of normal metabolism. These cells simultaneously also produce antioxidants in the body to neutralise the free radicals and maintain the balance of antioxidants and free radicals. The botanicals used as grain protectants against pulse beetles have a higher amount of phenols and antioxidants. This higher dose of the antioxidants may lead to oxidative stress in the insect body which may give rise to an imbalance of the free radicals and antioxidant levels (Chaitanya et al., 2016). This triggers the antifeedant effect which finally causes starvation and death of the insects (Bartosz and Bartosz, 2014).

Table 5: Efficacy of different botanicals in terms of mortality of *Callosobruchus chinensis* adult beetles in chickpea seeds during August, 2019

Treatments	Dose	Adult mortality (%) of *Callosobruchus chinensis* after					Pooled mean
		Day 1	Day 2	Day 3	Day 4	Day 5	
T_1	6%	*30.00 (33.00)^{a}	53.33 (46.92)^{ab}	66.67 (54.78)^{ab}	83.33 (66.14)^{b}	96.67 (83.00)^{ab}	66.00
T_2	2%	0 (4.05)^{e}	16.67 (23.86)^{def}	30.00 (33.00)^{cde}	46.67 (42.99)^{de}	60.00 (50.85^{ghi}	30.67
T_3	6%+2%	10.00 (18.43)^{bc}	30.00 (33.00)^{cd}	40.00 (39.15)^{cd}	60.00 (50.77)^{cd}	83.33 (66.14)^{de}	44.67
T_4	6%	16.67 (23.86)^{b}	36.67 (37.22)^{bc}	46.67 (43.08)^{bc}	73.33 (59.00)^{bc}	93.33 (77.28)^{bc}	53.33
T_5	2%	0 (4.05)^{e}	3.33 (8.85)^{gh}	16.67 (23.86)^{ef}	36.67 (37.22)^{ef}	46.67 (43.08)^{ij}	20.67
T_6	6%+2%	3.33 (8.85)^{de}	16.67 (23.86)^{def}	20.00 (23.49)^{ef}	46.67 (43.08)^{de}	66.67 (54.78)^{fgh}	30.67
T_7	8%	13.33 (21.14)^{bc}	30.00 (33.00)^{cd}	43.33 (41.15)^{cd}	70.00 (56.79)^{bc}	86.67 (68.86)^{cd}	48.67
T_8	8%	6.67 (13.64)^{cd}	13.33 (19.06)^{efg}	36.67 (37.22)^{cde}	60.00 (50.85)^{cd}	73.33 (59.00)^{efg}	38.00
T_9	8%+8%	10.00 (18.43)^{bc}	26.67 (31.00)^{cde}	43.33 (41.15)^{cd}	63.33 (52.78)^{cd}	76.67 (61.22)^{def}	44.00
T_{10}	5%	0 (4.05)^{e}	0 (4.05)^{h}	10.00 (16.35)^{fg}	30.00 (33.21)^{ef}	43.33 (41.15)^{j}	16.67
T_{11}	2%	0 (4.05)^{e}	6.67 (13.64)^{fgh}	23.33 (28.08)^{def}	23.33 (25.50)^{f}	50.00 (45.00)^{ij}	20.67
T_{12}	5%+2%	6.67 (13.64)^{cd}	13.33 (19.06)^{efg}	26.67 (31.00)^{cde}	36.67 (37.22)^{ef}	56.67 (48.85)^{hij}	28.00
T_{13}	0.04%	40.00 (39.15)^{a}	63.33 (52.86)^{a}	83.33 (66.64)^{a}	96.67 (83.00)^{a}	100 (88.72)^{a}	76.67
T_{14}	-	0 (4.05)^{e}	0 (4.05)^{h}	0 (4.05)^{g}	0 (4.05)^{g}	16.67 (23.86)^{k}	3.33
SEm±		2.78	4.04	4.27	3.94	2.98	-
CD (p=0.05)	-	8.07	11.69	12.36	11.40	8.62	-

The result of the present study reveals that the highest phenol content and lowest IC-50 value was recorded in Neem leaf Powder which reflects its good insecticidal properties against the test insect. Whereas, the lowest phenol content and highest IC-50 value were found in Tulsi leaf powder, which means it was least effective against the pulse beetle.

It is revealed from Table 6 that many botanical plants contain phenolics compounds and it corroborates the findings of the bio-efficacy trial of the botanicals. The phenolic compounds present in Neem and Melia are azadirachtin (Koul, 2004) melianone, meliantriol and nimbolidin A (Dhaliwal and Koul, 2007) which shows their defensive role due to allelopathic effect on insects.

Table 6: Total phenols and antioxidants content in different botanicals used for the bio-efficacy experiment against pulse beetle, *Callosobruchus chinensis*

Name of the botanicals	Phenols (mg g^{-1})	Antioxidants (IC-50 value) (µg ml^{-1})
T_1- Neem LP	345.69	207.77
T_2- Neem BP	149.86	880.62
T_3- Neem LP+BP	177.90	432.11
T_4- Melia LP	273.40	383.68
T_5- Melia BP	131.34	1248.46
T_6- Melia LP+BP	154.36	616.88
T_7- Datura LP	213.62	405.77
T_8- Datura SP	159.38	523.11
T_9- Datura LP+SP	173.93	492.97
T_{10}- Tulsi LP	127.11	1315.80
T_{11}- Tulsi SP	146.16	1156.88
T_{12}- Tulsi LP+SP	149.07	1141.58

LP: Leaf powder; BP: Bark powder; SP: Seed powder

4. Conclusion

6% NLP was the most efficacious botanical followed by 6% MPL and 8% DLP against *Callosobruchus chinensis* in terms of adult mortality and reduction of egg laying. Although it was found that Deltamethrin 2.5 WP performed the best, botanicals should be promoted as alternative to the chemicals. These botanicals can be fitted with the IPM strategies for long term storage of pulse seeds because these are locally available, economic and safe to the environment**.**

5. References

Alemayehu, M., Getu, E., 2015. *Callosobruchus chinensis* (L.) (Coleoptera: Bruchidae) Management on stored chickpea using botanicals in Amhara Region. American Journal of Experimental Agriculture 8(3), 167–177.

Anonymous, 2018a. Kew Science. "*Cicer arietinum* L. – Plants of the World Online". Retrieved 26 February 2018.

Anonymous, 2018b. Feedipedia. "Chickpea (*Cicer arietinum*)". www.feedipedia.org. Retrieved 26 February 2018.

Anonymous, 2020. Chickpea production in 2019, Crops/ Regions/World list/Production Quantity (pick lists)". UN Food and Agriculture Organization, Corporate Statistical Database (FAOSTAT). 2020. Retrieved 25 April 2021.

Bartosz, I.S., Bartosz, G., 2014. Effect of antioxidants supplementation on aging and longevity. Biomed research international 1–17.

Caswell, G.H., 1973. The storage of cowpea. Samaru Agricultural Newsletter 15, 20–73.

Chaintanya, R.K., Shashank, K., Sridevi, P., 2016. Oxidative stress in invertebrate systems, Free radicals and Diseases, Rizwan Ahmad, Intech Open, DOI:10.5772/64573.

Dhaliwal, G.S., Koul, O., 2007. Bio pesticides and pest management: conventional and biotechnological approaches. Kalyani Publishers, New Delhi.

Gahukar, R.T., Reddy, G.V.P., 2018. Management of insect pests in the production and storage of minor pulses. Annals of the Entomological Society of America 111(4), 172–183.

Ghosal, T.K., 2003. Bio-ecology of pulse beetles, *Callosobruchus* spp. (Coleoptera: Bruchidae) and potentiality of their safe management in storage at terai agro climate conditions. Ph.D. thesis submitted to the University of North Bengal, West Bengal.

Ho, C.T., 1992. Phenolic compounds on food: an overview. In: Ho, C.T., Lee, C.Y., Huang, M.T. (Eds.), Phenolic compounds in food and their effects on health I: analysis, occurrence, and chemistry. American Chemical Society, New York, 2–7.

Isman, M.B., 2001. Biopesticides based on phytochemicals. In: Koul, O., Dhaliwal, G.S. (Eds), Phytochemical biopesticides. Harwood Academic Publishers, Amsterdam, 1–12.

Karar, I.N.G.E., 1999. The efficacy of neem (*Azadirachta indica*) and kafur (*Eucalyptus camaldulensis*) extract on the control of *Sinoxylon senegalense* Karsch in (Acacia seyal Del) wood. A thesis submitted to the University of Khartoum.

Kaur, K.D., 2017. Evaluation of plant powders against pulse beetle (*Callosobruchus chinensis* L.) infesting pea seeds. M.Sc. (Ag) thesis submitted to Dr. Y.S. Parmar University of Horticulture and Forestry, Nauni-173 230, Solan (H.P).

Koul, O., 2004. Neem: A global perspective. In: Koul, O., Wahab, S. (Eds), Neem: today and in the new millennium. Kluwer Academic Publishers, Dordrecht, The Netherlands, 1–19.

Laizu, M.S.T., 2009. Effect of some botanicals on mortality, adult longevity and oviposition of pulse beetle on Mungbean. M.Sc. (Ag) thesis submitted to Sher-e-Bangla Agricultural University, Dhaka-1207, Bangladesh.

Misra, H.P., 2000. Effectiveness of indigenous plant products against the pulse beetle, *Callosobruchus chinensis* on stored black gram. Indian Journal of Entomology 62(2), 218–220.

Mookherjee, P.B., Jotwani, M.G., Yadav, T.D., Sircar, D., 1970. Studies on the incidence and extent of damage due to

insect pests in stored seeds. Leguminous and Vegetable seeds. Indian Journal of Entomology 32 (4), 350–355.

Parmar, V.R., Patel, M.V., Patel, S.R., Patel, B.H., 2018. Evaluation of botanicals as grain protectants against pulse beetle *Callosobruchus chinensis* (L). in mung bean. Indian Journal of Entomology 80(1), 78–84.

Pruthi, R.S., Singh, M., 1950. Pests of stored grain and their control. Indian Journal of Agricultural Sciences 18(4), 1–87.

Rahman, K.A., Sohi, G.S., Sapra, A.N., 1942. Studies on stored grain pest in Punjab II. Biology of *Bruchus analis* Fab. and *Bruchus chinensis* L. (Bruchidae: Coleoptera). Indian Journal of Agricultural Sciences 12, 851–864.

Raina, A.K., 1970. *Callosobruchus* spp. infesting stored pulses (grain legumes) in India and comparative study of their biology. Indian Journal of Entomology 32(4), 303–310.

Satyavir, P., 1983. Varietal resistance and susceptibility of cowpea to *Callosobruchus maculates* Fab. Indian Journal of Entomology 45(3), 213–217.

Saxena, R.C., 1998. Botanical pest control. In: Dhaliwal, G.S., Heinrichs, E.A. (Eds), Critical issues in insect pest management. Common wealth Publishers, New Delhi, 155–179.

Soumya, P.R., Kumar, P., 2017. Optimization of paclobutrazol dose for foliar and drenching applications under water deficit stress in chickpea (*Cicer arietinum* L.). International Journal of Bio-resource and Stress Management 8(4), 566–573.

Vasu, M., Nitika, N., 2020. Biology of pulse beetle *Callosobruchus chinensis* (L.) on three pulses. Indian Journal of Entomology 82(4), 631–632.

Wink, M., 1993. Production and application of phytochemicals from an agricultural perspective. In: Beck, T.A.V., Breteler, H. (Eds.), Photochemistry and Agriculture 34. Claredon, Oxford, UK.