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EVALUATION OF SOME BOTANICALS, THERMAL TREATMENT AND PACKING MATERIALS AGAINST PULSE BEETLE *CALLOSOBRUCHUS CHINENSIS* L. IN CHICKPEA

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ABSTRACT

The pulse beetle *Callosobruchus chinensis* (L.) is a major pest of stored chickpea. In the present study, efficacy of neem (0.5 and 1.0%) and turmeric powders (0.5 and 1.0%) has been evaluated against this pest, in combination with mustard oil (0.3%), thermal treatment @ 60°C for 20 min and packing materials including open, PP (polypropylene) and LD (low density polyethylene). The results revealed increased efficacy with concentration of neem and turmeric powder. Use of neem oil @ 1.0%+ mustard oil @ 0.3% kept in open packing, proved to be significantly superior at four months of storage. Turmeric powder @ 0.5%+ mustard oil @ 0.3% treated grains showed a significantly better germination. Moisture content differed significantly with treated samples. The colour change and myroflora incidence were comparatively minimum with neem leaf powder @ 0.5%+ mustard oil @ 0.3% treated samples. Thermal treated grains proved superior compared to botanicals, whereas, colour change led to significantly lesser effect. Among the packings, PP and LD packed samples significantly influenced the infestation and germination irrespective of the botanicals.

Key words: Chickpea, *Callosobruchus chinensis*, neem, turmeric, thermal treatment, packing materials, mycoflora incidence, moisture content, colour change, germination, protein content

The pulse beetle *Callosobruchus chinensis* (L.) is a major storage pest of chickpea causing 32-64% loss in Asia and Africa (Demnayk et al., 2007; Ketoh et al., 2005). Globally pest control measures in stored grains rely on insecticides and fumigants (Shaheen and Khaliq, 2005; Sharma et al., 2007). These pesticides are hazardous to man and the environment, also residues may remain on the treated grains. To reduce the use of insecticides, there is a need for alternatives such as biopesticides (Yusuf et al., 2011; Ashok et al., 2020; Singh et al., 2021), hot and cold treatment (Alice et al., 2013), and high-density polyethylene bags (Sanon et al., 2011) for the control of *Callosobruchus* spp. All these measures have different mode of action, but comprehensive knowledge that is essential on the use of combination of such methods against *Callosobruchus* spp. is lacking. The present study is an attempt to evaluate few such alternatives against *C. chinensis*.

MATERIALS AND METHODS

The study was done at the laboratory of Department of Processing and Food Engineering, Punjab Agricultural University (PAU), Ludhiana. Freshly harvested grains of chickpea var. GPF 2 were procured from the PAU, Ludhiana Research Farms. The whole lot was subjected to phosphine fumigation (one tablet of 3 gm of aluminium phosphide/ t and exposed for 7 days) to eliminate hidden infestation (Anonymous, 2020-21). The rearing was done by releasing five pairs of *C. chinensis*/ kg grains in an incubator (29± 2ºC, 70± 5% RH); and 10 pairs of *C. chinensis* were released in 10 kg grains and kept for one month for uniform infestation. Then infested grains (500 gm) were treated with- T_1 (neem leaf powder @ 0.5% + mustard oil @ 0.3%), T_2 (neem leaf powder @ 1.0 % + mustard oil @ 0.3%), T_3 (turmeric powder @ 0.5% + mustard oil @ 0.3%), T_4 (turmeric powder @ 1.0% + mustard oil @ 0.3%), T_5 (thermal treatment @ 60°C for 20 min) and T_6 (untreated control). The treated grains were stored in three types of packings- open type (where plastic jars covered with muslin cloth fastened with a rubber band); LD (low density polyethylene); and PP (polypropylene) at ambient conditions and left for four months. Monthly observations on insect infestation (%), germination (%), and colour change were recorded at regular intervals. To record observations on total infestation (%), both the damaged grains (%) and grains with eggs were sorted out and weighed separately. The infestation method given by Indian Standards: 7715 (1975) was followed. Further, to determine the moisture content in grains- hot air oven method (Association of Official Analytical Collaboration, 1984); for

germination test- blotting paper method (International Seed Testing Association,1985); and colour change measurement done by Colour Reader CR-10 (Konica Minolta Sensing Inc.) with the equation given by Gnanasekharan et al. (1992). Whole experiment was replicated thrice using completely randomized design (CRD), and the observations/ data obtained were statistically analysed.

RESULTS AND DISCUSSION

Table 1 depicts the efficacy of botanical treatments packed in three kinds of packings against infestation by *C. chinensis*; these reveal that the infestation decreased with the increase in concentration of neem as well as turmeric powder; and there was significant difference at different doses along with mustard oil in the study conducted in open packings (Table 1); and T_2 (neem oil @ 1.0% + mustard oil @ 0.3%) significantly superior against infestation and mycoflora incidence (%); insect infestation (%) started decreasing after 2 months in all the botanicals, and with mycoflora incidence, T_1 and T_2 showed more effectiveness at one month and later started decreasing. Verma and Anandhi (2010), Nisha and Asaf (2019) reported that neem leaf powder gave maximum mortality of *C. chinensis* in stored mung bean; turmeric and ginger were the least effective. Thakur and Pathania (2013) revealed that mustard oil, powder of black pepper, neem oil proved promising in black gram seeds up to five months. Adarkwah et al. (2017) showed that the combined mixture of plant powders and diatomaceous earth controlled the beetles faster in case of *Acanthoscelides obtectus*, *Sitophilus granarius* and *Tribolium castaneum* in stored grain cereals.

Results on the mycoflora incidence in the present study agree with those of Bhardwaj et al. (2013) in pea

Table 1. Effect of treatments/ packing materials on *C. chinensis* and mycoflora incidence on chickpea grains

Treatments (A)	Packing material (B)	Storage period (months) (C)							
		*Insect infestation (%)				*Mycoflora incidence (%)			
		1st	2nd	3rd	4th	1st	2nd	3rd	4th
T_1	Open	5.33	5.66	3.00	2.00	43.30	40.00	36.70	36.70
	PP	2.66	5.66	2.33	1.16	40.00	36.70	33.30	30.00
	LD	2.33	4.66	2.33	1.93	36.70	36.70	33.30	30.00
T_2	Open	3.00	3.33	1.66	1.26	43.30	36.70	33.30	30.00
	PP	2.00	3.66	1.33	1.00	36.60	30.00	26.60	30.00
	LD	1.66	2.66	1.66	1.43	33.30	26.70	20.00	26.60
T_3	Open	2.00	5.76	3.60	2.20	56.70	60.00	60.00	60.00
	PP	1.66	5.30	3.00	2.20	53.30	56.70	56.60	50.00
	LD	2.16	4.50	2.33	2.46	50.00	56.70	50.00	53.30
T_4	Open	1.66	2.66	1.66	2.70	53.30	56.60	60.00	63.30
	PP	2.00	2.66	2.33	2.33	50.00	50.00	53.30	56.70
	LD	2.33	3.00	1.66	1.00	46.70	43.30	46.70	50.00
T_5	Open	6.33	6.33	5.33	5.33	53.33	56.67	63.33	66.67
	PP	4.66	3.33	3.50	2.33	53.33	53.33	56.67	60.00
	LD	5.00	2.33	2.00	2.33	50.00	50.00	53.33	56.67
T_6	Open	11.66	30.30	30.30	98.66	70.00	83.30	90.00	100.0
	PP	5.66	5.66	5.66	19.00	66.70	80.00	93.30	96.70
	LD	5.66	6.33	6.33	4.33	66.70	76.70	90.00	93.30
CD (p=0.05)		A= 0.48; B= 0.34; C= 0.39; AB= 0.83; AC= 0.96; BC= 0.68; ABC= 1.60				A= 2.77; B= 1.96; C= NS; AB= NS; AC= NS; BC= NS; ABC= 1.60			

T_1: Neem 0.5% + mustard oil 0.3%; T_2= Neem 1.0% + mustard oil 0.3%; T_3= Turmeric powder 0.5% +mustard oil 0.3%; T_4: Turmeric powder 1.0% +mustard oil 0.3%; T_5: Thermal 60^{o}C/20 min; T_6: untreated control; *Means of 3 replications; PP= Polypropylene; LD= Low density polyethylene, initial mycoflora incidence: 42.5%

treated with neem oil. The moisture content in T_2 was significantly high, but may be strong effect of neem powder, the insect and mycoflora incidence was low; also increased significantly with increase in storage period up to first three months and then decreased further (Table 2). Similar results were obtained by Beedi et al. (2018) in gram; and Rathinavel and Raja (2007) in cotton. The botanicals influenced the grain colour significantly; it was observed more at Ist month of storage but gradually decreased. It may be due to the colour of botanicals attached to grains initially, but later the grains reverted back to their original colour. The results of the present study are contradictory to those of Ogendo et al. (2004) which found that the grain colour and odour were unaffected by the botanicals. The effect of botanicals on the germination of chickpea grains was insignificant with open packings (Table 2); it was above 95% in general and it corroborates with the study conducted by Bajiya (2009) on mung bean. Rahman and Talukder (2009) also showed that oil treatment had no adverse effects on germination even after three months of treatment in black gram. Increase in germination of seed was also observed in botanical treated grains in later months in PP and LD packed grains Similar results were observed by Nazareth et al. (2018) who found that the neem and tulsi leaf powders increased seed germination and emergence with increased shoot and root length of oilseeds.

Botanical treated grains showed insignificant differences in protein content (%); in botanical treated grains it was significantly less (18.30 in T_2) as compared to control sample (21.10%) at four months of storage (Table 2). The increase in crude protein content in control samples was directly correlated to the increase grain damage, insect excreta being uric acid as a main constituent and presence of eggs contributed a major part of N_2 estimated in the damaged grains and then converted into crude proteins. It was further observed there was little increase of true constituents in the treated samples also, which may be due to hidden infestation. A similar trend was observed by Hamdi et al. (2017), Bamaiyi et al. (2006) and Mbah and Silas (2007) that protein, moisture and ash contents increased in infested seeds of cowpea. Thermal treatment (T_5) i.e. exposure of grains at 60^{o}C for 20 min showed that the infestation was significantly high in thermal treated grains compared to botanical treated grains in open packings (Table 1). The insect infestation was high in initial months as compared to later months which may be due to the reason that thermal treated grains take time to kill the hidden infestation. Alice et al. (2013) showed the effect of exposure to sunlight on the mung bean grains having eggs of *C. maculatus* and found highest number of eggs laid/ seed was observed in seeds exposed to 4 hr (0.65) while it was the least when exposed to 24 hr (0.02). Renault et al. (2004) and Colinet et al. (2011) found the injurious effect of chill injuries in adult *Alphitobius diaperinus* exposed to low temperature. The incidence of mycoflora was less at Ist month and later started increasing which may be due that effect of thermal treatment reduced in later stages as moisture content also started increasing (Table 2). Increase in moisture content is directly related to fungi incidence (Mohapatra et al., 2017). Germination of thermal treated grains differed insignificantly in open packing and with botanicals. Germination of seeds was not affected by the thermal treatment (Kariluoto et al., 2006). Significantly less colour change was observed in thermal treated grains as compared to botanical treated ones. Hou et al. (2015) also studied effect of thermal treatment on colour change of chestnuts and suggested that the yellowness (b*) was more sensitive to temperature changes than the lightness (L*). Protein content in thermal treated grains were more as compared to other treatments in open packings which may be because roasting saves and improves protein parameters. The similar results were obtained by Jurkovic and Colic (1993) in wheat grains.

About the packings it was observed that infestation increased up to two months and then started decreasing; LD packing was significantly superior compared to that of PP and open packings in 2nd, 3rd and 4th months, showing the least infestation (Table 1). These results are similar to those of Gomaa and Salem (2018) and Sanon et al. (2011). Qasim et al. (2013) studied the penetration ability of the beetle, *Tribolium castaneum* for three packaging types includes polyethylene (PE), polypropylene (PP) and polyvinylchloride (PVC) and found PE as a susceptible medium, while LD packing was more effective against fungi. The moisture content increased in all the treatments with the increase in storage time. The results are similar to those of Yeole et al. (2018) found that air tight packing materials like polypropylene bags and hermetic bag had higher microbial load because the environment inside these bags has high relative humidity with warm temperature. The colour of grains is used as an indicator for the quality of food products. study, significant change in colour of moong grains was observed from 1-4 months of storage period in all the tested packings (Table 2). Grains packed in LD packings showed less change in colour, while maximum colour change found in PP bags. Similar results were found by Yeole et al. (2018). The

Table 2. Effect of treatments/ packing materials on quality parameters of stored chickpea grains

Treatments (A)	Packing material (B)	Storage period (months) (C)															
		*Moisture content (%)				*Colour Change				* Germination (%)				*Protein content (%)			
		1st	2nd	3rd	4th	1st	2nd	3rd	4th	1st	2nd	3rd	4th	1st	2nd	3rd	4th
T_1	Open	11.00	13.05	16.46	15.06	6.59	5.92	4.44	2.20	95.33	95.67	95.00	95.67	18.30	18.50	18.48	18.40
	PP	8.37	9.65	14.12	13.56	6.31	6.09	3.72	2.55	98.00	97.00	97.00	97.67	18.10	18.50	18.45	18.40
	LD	9.49	9.66	14.27	13.78	5.10	3.02	2.57	2.41	98.00	96.67	96.67	97.67	18.14	18.50	18.42	18.39
T_2	Open	10.35	13.81	17.38	15.28	9.34	9.15	3.68	2.52	96.00	95.67	95.67	95.67	18.40	18.45	18.34	18.30
	PP	7.50	10.21	14.69	14.52	6.54	5.59	5.15	2.66	97.33	97.00	97.67	97.67	18.30	18.40	18.44	18.40
	LD	7.21	10.28	14.64	14.27	3.74	3.04	3.13	4.17	98.00	97.33	97.33	98.00	18.30	18.38	18.40	18.40
T_3	Open	9.36	13.10	16.21	15.22	7.07	5.03	4.72	4.39	96.33	95.67	96.00	96.33	18.20	18.75	18.80	18.40
	PP	9.06	9.35	14.71	14.35	6.35	6.19	6.10	6.08	97.00	98.00	98.00	97.67	18.10	18.64	18.60	18.58
	LD	9.26	9.76	14.96	14.77	6.51	5.47	4.72	4.50	97.67	98.00	98.00	98.00	18.25	18.52	18.50	18.49
T_4	Open	9.88	13.48	16.72	15.67	7.07	5.74	5.47	5.35	96.00	96.33	96.00	96.00	18.24	18.44	18.50	18.46
	PP	8.96	9.85	15.11	14.06	6.31	7.82	7.63	6.21	97.67	98.00	98.00	97.33	18.30	18.40	18.40	18.46
	LD	9.12	10.17	13.70	12.79	5.30	10.85	7.61	3.61	97.33	98.33	98.00	97.33	18.30	18.44	18.40	18.36
T_5	Open	9.40	13.52	14.85	17.67	0.49	0.90	1.99	2.69	95.33	95.33	95.33	95.00	18.50	18.56	18.50	18.50
	PP	9.36	11.88	14.03	14.35	1.80	2.91	4.27	6.66	96.33	97.67	97.00	97.67	18.35	18.30	18.42	18.42
	LD	7.77	10.70	13.88	15.04	1.36	1.45	3.48	4.11	97.00	97.67	97.33	97.33	18.40	18.42	18.40	18.45
T_6	Open	11.43	15.04	20.09	19.22	1.14	3.11	3.42	3.66	79.33	66.00	66.00	14.33	18.60	19.60	20.80	21.10
	PP	9.09	11.65	15.23	14.84	2.86	3.28	3.42	7.27	85.00	73.33	73.33	52.67	18.40	18.50	19.20	19.60
	LD	9.78	11.39	15.86	15.03	1.39	1.90	2.18	2.85	88.67	88.67	88.67	86.00	18.40	18.60	18.70	18.70
CD (p=0.05)		A=0.23; B=0.16; C=0.4; AB=0.41; AC=0.53; BC=0.37; ABC=0.91;				A=0.32; B=0.23; C=0.26; AB= 0.56; AC=0.65; BC=0.46; ABC=0.11				A=0.40; B=0.28; C=0.36; AB=0.69; AC=0.89; BC=0.63; ABC=1.55				A=0.13; B=0.95; C=0.12; AB=0.23; AC=0.30; BC=0.21; ABC=0.52.			

T_1: Neem 0.5% + mustard oil 0.3%; T_2= Neem 1.0% + mustard oil 0.3%; T_3= Turmeric powder 0.5% +mustard oil 0.3%; T_4: Turmeric powder 1.0% +mustard oil 0.3%; T_5: Thermal 60^{o}C/20 min; T_6: untreated control;*Means of 3 replications; PP= Polypropylene; LD= Low Density Polyethylene, Initial moisture content: 8.73%, Initial protein content: 17.9%, Initial germination=98.0%

protein content (%) in grains packed in various type of packings differed non-significantly. Also, there is insignificant effect of PP and LD packing on protein content. The results are similar to the studies conducted by Mbah and Silas (2007) which reported an increase in protein contents with severity of infestation.

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