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Insecticidal and Repellent effect of Botanicals against Red Flour Beetle *Tribolium castaneum*, (Herbst) Pest of Stored Grains

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ABSTRACT: Red flour beetle, *Tribolium castaneum* (herbst), occurs worldwide, is a devastating pest of stored grains particularly the previously damaged cereals. Grub feed on embryo and germ portion of seeds and grains with release of quinines which gives unpleasant odour in serve infestation. To control this pest, chemical pesticides and fumigants are widely employed, which lead to the development of insect resistance. Nowadays, phosphine resistance is a major concern and it also affects human health and the environment. Therefore, it must need to implement an alternative management strategy for pest control. In present study the insecticidal and repellent effect of three botanicals viz. sweet flag (*Acorus calamus*), black pepper (*Piper nigrum*), and clove (*Syzygium aromaticum*) has been evaluated against red flour beetle under laboratory conditions with different exposure time that were replicated thrice. The highest insecticidal effect was observed in black pepper after fifteen days of treatment followed by sweet flag and clove at 88.66%, 85.34%, and 76.56% respectively. Botanical extract exhibited repellent activity against red flour beetle with highest repellency was observed in sweet flag rhizomes extract after 48 hours of treatment 68.42% followed by black pepper and clove at 64.76%, 61.29% respectively. These findings show that sweet flag, black pepper, and clove possess essential components which impart insecticidal and repellent activity to control insect pests.

Keywords: *Acorus calamus*, red flour beetle, repellency, insecticidal effects, botanicals.

INTRODUCTION

Wheat (*Triticum aestevum*) is an important staple food and significant cereal crop of many diets around the world. According to ICAR-IIWBR, karnal estimated production of India is 106.4 million tonnes of wheat in year 2021-2022, with national productivity of 3484 kg per hectare. Storage of food grains is important task in order to maintain the good health and to protect it from insect infestation. Out of the total post harvest losses alone insect can cause 2 to 4.2 per cent loss every year as per IGMRI, Hapur, U.P., 2019 report (Ali *et al.*, 2016).

Most destructive pest of wheat includes Red flour beetle *Tribolium castaneum* (Tenebrionidae: Coleoptera). It is a serious secondary pest of stored grains such as cereals, pulses, oilseeds as well as agriculture products. Adult can live for one to one and half year, female may lay upto 1,000 eggs during their life span. Both adult and the grub feed on damaged grains as well as milled products which causes unpleasant odour and reduces quality and quantity of food products. Repeated consumption of such contaminated grains will cause hazardous effects to human health and the environment (Alsudani *et al.*, 2021). Fumigation and synthetic pesticides has long been used regularly to control this pest (Attia *et al*., 2020). Primarily phosphine and methyl bromide employed globally for controlling stored grain pests but nowadays these control strategies are losing effectiveness because of resistance development among storage grain pests. Additionally, the use of chemical pesticides has been linked with adverse effects on non-target organisms, human health and environment (Nicolopoulou-Stamati *et al.*, 2016) while causing soil,

water and air pollution in agriculture (Hamel *et al.*, 2020; Sonu Kumar *et al.*, 2022).

In order to prevent resistance development among the pests it is necessary to use pest control strategies that are safe alternatives which are derived from the sources that are not dangerous to people, such as plant components, botanical oils, spices etc. Plant extracts have been shown to be a safe substitute to chemical pesticides because the presence of secondary metabolites such terpenoids, phenolics, flavonoids, and alkaloids which provide insecticidal and repellent properties (Rakha and Mousa 2017). Botanicals are more reliable, biodegradable, economically cheap in production and less likely to provoke resistance development (Jaleel *et al.*, 2020). Therefore present study was conducted to evaluate the insecticidal and repellent effect of plant species viz. black pepper (*Piper nigrum*), clove (*Syzygium aromaticum*) and sweet flag (*Acorus calamus*) on adults of red flour beetles.

MATERIALS AND METHODS

Study was carried out at Biocontrol Laboratory, Department of Entomology and Laboratory of Biotechnology, Biotechnology Centre. Post Graduate Institute Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola.

Rearing of red flour beetles (*Tribolium castaneum*): Red flour beetles of mixed age were collected from store house Dr. PDKV Akola. Collected adults were introduced into a sterilized plastic jar of size 500 ml containing 200 g diet which contains 95 parts of wheat flour and five parts of brewer's yeast. Jar was covered with muslin cloth and tightens up by rubber for proper aeration and to prevent the escape of newly emerged adults. Culture was maintained at 27±2°C and relative humidity 70±5 % (Perez-Mendoza *et al.*, 2011). Adult used for experiment were obtained from stock culture established (Rehman *et al*., 2020).

Preparation of botanical extracts. Botanical extract were prepared according to Odey *et al.* (2012) with some modifications. Seeds of black pepper, rhizomes of sweet flag and unopened bud of clove were purchased from local market. Botanicals were washed and dried under electric oven at 50°C for 24 hr and ground into a fine power with the help of mixture girder. These powders were used for mortality test. For extract preparation, Ten gram of power was added in 100 ml of distilled water and heated at 65°C for 20 min. filter the solution by using whatman no. 1 filter paper and then centrifuged the extract at 1200 rpm for 10 min. Stored the extract in refrigerator for further experiment.

Mortality test: Prepared botanicals powder were mixed with twenty gram of sterilized wheat grains in plastic jar (20 ml) and shaken well for proper application to each grain. In each jar twenty newly emerged adults were released separately. Number of insect died after treatment was counted in regular intervals, treatment was repeated thrice (Ahmad *et al.*, 2019). Moribund insects were considered as dead. A control set was maintained without application of botanical. Mortality per cent was calculated by using formula,

$$\text{Mortality (\%)} = \frac{\text{Number died insects}}{\text{Total number of released insects}} \times 100$$

Repellency assay: Repellency assay was carried out using filter paper of size 9 cm. Filter paper was cut into two equal parts, half part treated with botanical extracts and another half part treated with distilled water as a control. Rejoined the two pieces together using tape carefully so that attachment do not prevent the free movement of insects from one side to the other. Twenty adults of red flour beetle was released in to a petri plate containing treated filter paper and covered with lid. Number of insect moves toward control area from treated area of filter paper was counted (Ramsha *et al.*, 2019; Iqbal *et al*., 2010). Repellency per cent was calculated using formula,

$$\text{Repellency (\%)} = \frac{(\text{Nc - Nt})}{(\text{Nc + Nt})} \times 100$$

Where, Nc = number of insects on the control (untreated) area;

Nt = number of insects on the treated area.

Data Analysis. The collected data were analyzed in Completely Randomized Design (CRD) with four treatments and three replications at five per cent level of significance using Web Based Agricultural Statistical Package (WASP) developed by ICAR Research Complex, Goa, India.

RESULTS AND DISCUSSIONS

Insecticidal effect of botanicals on red flour beetle. Experiment was conducted to know the insecticidal activity of three botanicals. Results computed in Table 1 indicate that, highest mortality of 88.66% was observed in black pepper after 15 days of treatment followed by 85.34%, 76.56% in sweet flag and clove respectively. Initially after 3 days of exposure higher mortality 27.75% was observed in sweet flag followed by black pepper, 25.84% and clove, 21.07% as the time of exposure increases mortality also increases. After 5 days of treatment 39.82%, 38.67% and 35.46% mortality was observed in sweet flag, black pepper and clove respectively. Similar trends of mortality were also observed after 10 days of treatment 77.03%, 76.02% and 69.63% in sweet flag, black pepper and clove. Comparison of mortality showed by botanicals for different time periods revealed that, as the time of exposure increases from 3 to 10 days mortality per cent also increases in sweet flag than black pepper and clove. Although, black pepper had lower value than sweet flag yet it was more persistence as highest value of mortality was observed after 15 days of exposure. Mortality of adults depends on botanicals and their exposure time. It is due to presence of many active phyto-chemicals present in natural botanicals. In previous studies many researchers have already studied

the potency of botanicals against red flour beetle. The finding of this study are supported by More *et al.,* 2015 who investigated toxicant effect of nine spices and he found that black paper showed significantly elevated adult mortality of red flour beetle and germination viability of wheat grains compared to other tested spices turmeric, chili, coriander, garlic, fennel, ginger, fenugreek and cumin. Saxena (2016) reported that black pepper (*Piper nigrum*) shows maximum mortality recorded 60.00% than clove powder (*Syzygiumn aromaticum*) (55.86%). AL-joboory (2019) studied insecticidal property of black (*Piper nigrum*) in powder form against red flour beetle on wheat grains shows that black pepper powder@ 6% causes maximum adult mortality than cinnamon, coriander and syphilis. Ushasri *et al.* (2022) reported that 100% mortality was observed in black pepper@ 1.5g/kg than other tested botanicals with minimum seeds damage.

Table 1: Effect of the botanicals powder on mortality of red flour beetle adult.

Botanical extract	Application to wheat grains (% mortality)			
	3 DAT	5 DAT	10 DAT	15 DAT
Sweet flag @ 0.5%	27.75 (31.79)[a]	39.82 (39.13)[a]	77.03 (61.36)[a]	85.34 (67.49)[b]
Black pepper @ 0.3%	25.84 (30.55)[b]	38.67 (38.45)[b]	76.02 (60.68)[a]	88.66 (70.32)[a]
Clove @ 0.3%	21.07 (27.32)[c]	35.46 (36.55)[c]	69.63 (56.56)[b]	76.56 (61.04)[c]
Control	0.00 (0.28)[d]	0.00 (0.28)[d]	0.00 (0.28)[c]	0.00 (0.28)[d]
SE(m)±	0.53	1.00	0.65	0.76
CD@5%	1.07	0.80	1.30	2.25
CV	3.03	1.49	1.24	1.90

*Figures in the parentheses are angular transformed values; *Data represented in the tables are mean of three replications

Repellent effect of botanicals on red flour beetles. Data presented in Table 2 shows that, among the all tested botanicals extract, maximum repellency was observed in sweet flag@ 0.5% after 48 hours of treatment 68.42% followed by black pepper@ 0.3% which at par with clove@ 0.3 % recorded 64.76% and 61.29% respectively. Initially after 12 hour of exposure 40.56% repellency was observed in sweet flag followed by black pepper and clove recorded 38.21% and 35.16%. Repellency increasing after 24 hours of exposure 61.69%, 59.73% and 57.19% in sweet flag, black pepper and clove respectively. However, significant decrease in repellency was observed after 72 hr of treatment recorded minimum repellency in sweet flag 37.21% followed by black pepper extract 32.32% which at par with clove 31.07%. The result demonstrated that all the tested botanicals have tendency to repel the adults of red flour beetle trough filter paper assay. Sweet flag shows increasing repellency from 12 hr to 48 hr after treatment and then after 72 hr there is gradual decreased in repellency than other two botanicals black pepper and clove which indicate that repellent effect is depend on type of botanical used in the experiment and time of exposure. Present study in accordance with earlier studies Chen *et al.* (2015) noticed that rhizomes of sweet flag (*Acorus calamus*) showed insecticidal and repellent activity evaluated against *Tribolium castaneum*. Essential oils particularly of *A. calamus* contained β–asarone, shyobunone, isoshyobunone which exhibited insecticidal activity varies from genotype to genotype. Similarly, Iqbal *et al.* (2010) reported the highest repellency of sweet flag @ 1600 µg/cm^3 in first week of treatment then repellency decreases gradually from fourth to eight week compared to other botanicals tested which are neem, turmeric, balchar, harmal, kuth and ner. Jilani and Saxena (1990) observed greater repellent effect of sweet flag oil through filter paper strips experiment against lesser grain borer. Hossain *et al.* (2008) confirmed that extract of sweet flag rhizomes in petroleum ether was more effective against red flour beetle than rice weevil. Madavi *et al.,* 2022 reported that sweet flag extract shows maximum repellency 90.07% against pulse beetle (*Callosobruchus chinesis*) than black pepper and clove.

Table 2: Repellency effects of botanicals on red flour beetle adults.

Botanical extract	Repellency (%)			
	After 12 hrs	After 24 hrs	After 48 hrs	After 72 hrs
Sweet flag @0.5%	40.56 (39.56)[a]	61.69 (51.76)[a]	68.42 (55.81)[a]	37.21 (37.59)[a]
Black pepper @ 0.3%	38.21 (38.18)[b]	59.73 (50.61)[b]	64.76 (53.58)[b]	32.32 (32.65)[b]
Clove @ 0.3%	35.16 (36.37)[c]	57.19 (49.13)[c]	61.29 (51.52)[c]	31.07 (33.88)[b]
SE(m)±	0.36	0.45	0.53	0.43
CD@5%	1.09	1.47	1.61	1.29
CV	1.44	1.19	1.24	1.92

*Figures in the parentheses are angular transformed values; *Data represented in the tables are mean of three replications

CONCLUSION

It is concluded that all the three natural plant botanicals have tendency to control *T. castaneum* population in wheat grains. Black paper and sweet flag shows the highest insecticidal and repellent effect on *T. castaneum*. These powders can be used as potential agent for the plant protection strategies which are easily available and biodegradable without any risk to human health. Hence further studies should conduct to evaluate the potential of tested botanicals to control the pests of stored grains.

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