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Screening of some medicinal plant extracts for toxic and repellent potential against adult stage of rust red flour beetle, *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae)

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Abstract

Plant based insecticides are effective control agents against the stored grain insect pests especially for *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae). Due to the problems related with the indiscriminate use of artificially made insecticides like insect resistance and effect on non-target organisms, many scientists in whole have focused on the search for active naturally occurring essential plant products. The red flour beetle, *Tribolium castaneum* (Herbst) is one of the severe pests of stored grain commodities The botanicals are have no toxic effects on our surrounding but the bioactivities of these plant extracts may induce certain detrimental effects on the biology of the certain insects causing infestation in stored grains The toxic and growth disruptive effects extracts from five medicinal plants viz; *Azadirachta indica, Murraya exotica, Eucalyptus comeldulensis, Trachspermum ammi* and *Teminalia chebula* were evaluated at a rate of 5, 10 and 15 % concentrations against the *Tribolium castaneue*. Extract of *Trachspermum ammi* gave maximum mean repellency (83.70%) at 15% while minimum repellency was observed in case of *Teminalia chebula* at 15% concentration. Maximum mortality was 16.11% in case of *A. indica* at 15% concentration while minimum (2.78%) was observed against *T. chebula* at 5%.

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Introduction

Post-harvest losses can reach up to 9-20% due to the attack of stored product insects (Philips and James, 2010) and among these stored grain insect pests Tribolium castaneum is the major secondary pest of stored food products and processed foods throughout world (Lee et al., 2002). Synthetic insecticides pose harmful effects on insect pests of stored commodities so there is a need to utilize the plant extracts (Negahban et al., 2007) which pose less danger on the surroundings (Abbasipour et al. 2011). Also, in order to combat the resistance problem in stored grain insect pests, we should move towards other control measures which are socially acceptable, ecologically reliable and economically feasible (Kim et al., 2003; Tapondjoua et al., 2005). Plant extracts are best substitute methods for control of stored commodities insects' pests. Use of plant extracts of Azadirachta indica is found to be most effective against the Callosobruchus chinensis. Botanical extracts can also be used in coordination with other control measures (Hasan et al., 2012). Fumigation is the important way for control of stored grain insect pests. At present, most frequently used fumigants are methyl bromide and phosphine. But their use is restricted due to their toxic effects on environment. An alternative method for control of insect pests of stored commodities is the use of plant oils as fumigents (Tayoub et al., 2012). Essential oils can also be utilized as fumigant s(Li et al., 2010) and can be used as alternative compounds which can control insect pests. These compounds are naturally found in many plants and these are used for the defense and are rich of secondary metabolites (Paranagama et al., 2003).Present study was designed so as to evaluate the effectiveness of new plant extracts along with some older ones so as to compared the bio-efficacy of these extracts as quick knockdown agents.

Materials and methods

The current study will be steered in the Grain Research, Training and Storage Management Cell of

the Department of Agri. Entomology, University of Agriculture Faisalabad during the year 2012-2013.

Insects collection

T. castaneum was collected from grain market and godowns which are located in Bahawalnagar.

Insects rearing

Insect culture were reared on sterilized wheat flour in sterilized jars which will be kept in the incubator at temperature $30\pm2^{\circ}$ C and $70\pm5\%$ relative humidity to get the homogeneous population. Thirty insects were released in each jar containing 250gm flour. The jars were covered with muslin cloths to avoid insect escape. After five days adults were sieved out from the flour and the flour containing eggs were kept again in the incubator to get another generation. The flour containing adults were transferred again into jar to get homogenous population.

Collection of plant material

Plant material *Azadirachta indica, Murraya exotica, Eucalyptus comeldulensis, Trachspermum ammi, Teminalia chebula* and *Nicotiana tabacum* were collected from the different places.

Preparation of extracts

The plant materials were washed in water and then after drying in shade were ground to bring these in form of powder. The extracts were prepared using rotary shaker by shaking 1:2 ratio, following the procedure which is being pronounced by Hasan *et al.* **2012**. The plant extract which is obtained thus put in clean bottles and stored in refrigerator at 4°C.

Bioassay for mortality (%)

The mortality of *T. castaneum* adults was checked using Petri dishes and Whatmans filter paper. Different concentrations of plant extracts were applied on filter papers with the help of pipette and allowed to get dry before the start of bioassay. Treated filter papers were transferred in petri-dishes. One petri dish was kept as a control dor each treatment

Int. J. Biosci.

containing untreated flour. Thirty adults of *T. castaneum* were released in each petri dish containing treated flour and then the Petri dishes were covered with a perforated lid. Three replications of each treatment were used. Mortality of the adults will be recorded four times after equal intervals of 24, 48, 72 and 96 hours.

Bioassay for growth inhibitory effects of plant Extracts

The growth inhibitory effects of the plant extracts were evaluated in small plastic vials. Different concentrations were applied on wheat flour and then the flour was allowed to dry. Thirty adults were introduced in each vial. Data were collected for larval duration, % pupal emergence and adult formation in F_1 progeny were recorded after two day interval, till the F_1 adult emergence.

Bioassay for evaluation of repellent effects of plant extracts

To evaluate repellent effect of plant extracts against *T*. *castaneum*, Whatman no.1 filter papers were used. Different concentrations of the extracts were applied with the help of micropipette on the half filter papers while the other haves were treated with acetone only. In order to evaporate the solvent from treated and untreated halves, the filter papers will be air dried. Then these halves were attached with each other by using sqash tape and were kept in petri dishes. Thirty insects will be released in the center of each Petri dish. The petri dishes then were covered and placed

in incubator. Three replications were used for each concentration. Observations for number of insects on both halves were recorded after 24, 48 and 72 hours.

Statistical analysis

After the completion of the experiment the data was recorded periodically. Abbott's formula was used to calculate corrected mortality and data so obtained was analyzed by Analysis of Variance using STATISTICA 7.0 software and the corrected. Tuckey-HSD test was used comparison of means of significant treatments.

Results

The current study was conducted to evaluate the insecticidal, repellent and growth disrupting effects of different concentrations of extracts of Murraya Azadirachta indica Eucalyptus exotica, comeldulensis, Trachspermum ammi and Teminalia chebula against Tribolium castaneum (Herbst) (Coleoptera: Tenebrionidae) at various intervals of time and their interactions. Three concentrations viz; 5, 10 and 15% were applied to evaluate the mortality of T. castaneum for different exposures of time viz; 24, 48, 72 and 96 hrs. Repellent action of these five plants was evaluated against T. castaneum (Herbst) (Coleoptera: Tenebrionidae) at different cocentrations.

Table 1. Effect of different concentration	ns on repellency of <i>Tribolium castaneum</i> .
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% Mortality ± S.E					
Conc.	E. comeldulensis	M. exotica	T. ammi	T. chebula	A. indica
5%	73.33 ± 4.30 abc	77.037 ± 6.94 ab	74.44 ± 4.44ab	45.93 ± 3.45 de	73.33 abc± 5.18
10%	70.00 ± 4.30 abc	80.37± 4.55ab	$77.78 \pm 4.48 \mathrm{ab}$	$58.89 \pm 6.67 \mathrm{cd}$	$70.74\pm2.20\mathrm{abc}$
15%	$77.78 \pm 4.75 \mathrm{abc}$	$79.26\pm5.23\mathrm{ab}$	$83.70 \pm 2.80a$	38.51± 5.36e	$67.78 \pm 5.98 \mathrm{bc}$

Repellent action of extracts of *M. exotica, E. comeldulensis, A. indica, T. ammi* and *T. chebula* was evaluated against the *T. castaneum* and maximum repellency (83.70%) was observed at highest concentration 15%, followed by *M. exotica*

(80.37%) at 10% and *T. chebula* (38.51%) respectively (Table No 1).

Maximum percent repellency (90.37%) was observed after 24 hrs. Least percent repellency (52.51%) after 96 hrs (Table No 2).

The concentration \times plant interaction also gave maximum repellency of (83.70%) at 15% concentration by *T. ammi*. This shows that repellency is directly affected by concentration.

% Mortality ± S.E					
Exposure period	E. comeldulensis	M. exotica	T. ammi	T. chebula	A. indica
24	$86.67{\pm}4.44\mathrm{abc}$	$81.48 \pm 4.37 \mathrm{abcd}$	90.37 ± 4.44 ab	$37.78 \pm 2.45g$	81.48 ± 4.18 abcd
48	72.59 ±3.44cde	96.30 ± 4.66a	79.26 ± 4.34bcd	$53.33 \pm 3.47 f$	72.59 ± 344cde
72	$61.85 \pm 3.31 \mathrm{abc}$	$58.89 \pm 3.23 ef$	66.30 ± 2.80def	52.51± 3.47fg	57.78 ± 3.23.98ef

Table 2. Effect of different exposure periods on repellency of Tribolium castaneum.

The results further revealed that the mortality of *Tribolium castaneum* increased by increasing the concentration of *A. indica. Tribolium cataneum* gave highest mortality (16.11%) at 15%, while 10% gaved 7.77% mortality. Least mortality was observed in case in the control. By comparing the relative efficacy of

extracts of *M. exotica, E. comeldulensis, A. indica, T. ammi* and *T. chebula*. Highest mean percent mortality (16.11%) was observed in case of *A. indica* at 15% as compare to the extract of *Murraya exotica* which gave high mortality of (15.00%) (Table No 3).

% Mortality ± S.E					
Conc.	E. comeldulensis	M. exotica	T. ammi	T. chebula	A. indica
5%	5.00 ± 0.30 efg	$8.61 \pm 1.10 \mathrm{de}$	$8.89 \pm 1.10 \mathrm{de}$	$2.78\pm0.10\mathrm{g}$	5.28 ± 0.38 efg
10%	7.78 ± 1.10 def	11.11± 1.12bcd	8.61 ± 1.48 de	3.33 ± 0.67 g	7.78 ± 1.10def
15%	9.72 ± 1.11cd	15.00 ±1.13ab	13.33 ± 1.13abc	4.44± 1.05fg	$16.11 \pm 1.15a$

Mean percent value of mortality regarding time was maximum (22.59%) after 96hrs by, while minimum percent mean mortality (0.00%) was observed after 24 hrs. (Table No 4).

Table 4. Effect of different exposure periods on Mortality of *Tribolium castaneum*.

% Mortality ± S.E						
Exposure period	E.comeldulensis	M. exotica	T. ammi	T. chebula	A. indica	
24	1.85 ± 0.30 jkl	1.11 ± 0.24 kl	2.59 ± 0.44ijkl	0.00 ± 0.00 l	5.27 ± 1.14	
48	4.81 ± 0.50hijk	8.15 ± 1.25 fgh	8.52 ± 1.26 fgh	1.11 ± 0.24 kl	7.78 ± 1.20	
72	7.78 ± 1.20fgh	14.44 ±1.28cde	$10.37 \pm 1.27 \mathrm{efg}$	6.66 ± 1.19ghi	16.11 ± 1.30	
96	15.56 ± 1.27 bcd	$22.59 \pm 1.33a$	19.59 ±1.32ab	6.29 ± 1.18hij	$18.51 \pm 1.29 \mathrm{abc}$	

Discussion

277

Plant extracts proved to be effective repellents and toxicants. Iqbal et al. (2010) also done work on the repellent action of Neem and it was concluded that it ranged from 63% to 75%. Gradual decrease in repellency was noted over a longer period 8 weeks but in the present experiments, rapid effects were noted and the insect behavior showed lower repellency towards the treated half with the passage of time. Islam and Talukder (2005) also conducted similar studies by using A. indica powder and it was found that 53.13% progeny was affected but in our experiments the mortality at the highest

Sagheer *et al*.

concentration was 16.11%. The present results are much lower as compared to Islam and Talukder (2005) as they utilized the seed powder and the present studies were based acetone based extracts of leaves. Resistance against A. indica (Neem) also seems to be one of the reasons. Results are also in accordance with Haq et al. (2005) who experimented on different extracts. According to the above mentioned authors, the plant extracts have toxic, fumigant, and sterilizing, anti-feeding and repellent effects against the insect pests of stored commodities. In view of above results, it is recommended that should we pay more

consideration to use of botanical insecticides. The botanical insecticides are safe for our environment as well as our health. As storage is our main objectives so these botanical insecticides not only prevent the insect pests of stored commodities and these are also safe to our health and environment. This information relating to use of plant extracts will act as guide for planning economical control methods and using these plant extracts against the management of T. castaneum. Plant extracts are rich in active metabolites that can play an important role for the integrated pest management programs. Rigorous testing and screening of the plant extracts along with the biochemical studies should be promoted for the identification and synthetic production of active ingredient present in potent plant extracts. Our results and those reported earlier clearly indicate variations in the activity of essential extracts regarding the difference in the plant species, exposure period and concentration used. Bioactivity of these natural extracts can be increased by extracting the natural ingredients of respective plants and by synthetically manufacturing these bio-actives at commercial scale.

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