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Repellent potential of *Azadirachta indica* A. Juss. and *Glycyrrhiza glabra* L. against cowpea bruchid, *Callosobruchus chinensis* L. (Coleoptera: Bruchidae)

Mansoor-ul-Hasan[1], Muhammad Sagheer[1], Muhammad Farhan[2], Malik Najam-ul-Hassan[2], Shireen Raza Haidri[1], Maimona Bukhari[3], Hafiza Tahira Gul[3], Fawad Zafar Ahmad Khan[4*]

[1]*Department of Entomology, University of Agriculture, Faisalabad, Pakistan*

[2]*Department of Pest Warning and Quality Control of Pesticides, Punjab, Pakistan*

[3]*Bahauddin Zakariya University, Multan, Pakistan*

[4]*College of Agriculture, Bahauddin Zakariya University Multan, Bahadur Sub-Campus, Layyah, Pakistan*

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Abstract

Keeping in view the bio-friendly nature of plant extracts, experiments were designed to determine the toxicological and repellent effects of *Azadirachta indica* and *Glycyrrhiza glabra* against the insect pest of stored beans, *Callosobruchus chinensis* (Coleoptera: Bruchidae). For determination of repellent action, area preference method was adopted. In the method of area preference, half of filter paper was treated with serial concentrations of plant extracts. After the release of beetles and their response against the plant extracts, results highlighted potential of crude extracts towards creating a repellent zone for the storage beetle. After the experiments, it was found that the *A. indica* was more potent as compared to the *G. glabra*.

***Corresponding Author:** Fawad Zafar Ahmad Khan ✉ fawaduca@gmail.com

Introduction

Cicer arietinum L. (chickpea) has high nutritional value in pulses and is cultivated all over the world. It is ranked third in the important leguminous food items in the world. The contents of this pulse are maximum than other pulses such as carbohydrates and proteins are 38-59% and 25.3-28.9% respectively (Salami and Ahmadi, 2010; Shukla *et al*., 2007). Heavy qualitative and quantitative losses occur due to the attack of pulse beetle, *Callosobruchus chinensis* L. in the stored chickpea seeds and other stored grains such as beans, gram and lentil seeds in the developing countries. Invasion of this insect causes reduction in germination of seeds, weight loss and lower market value (Raja *et al*., 2008; Patel, 2011; Sagheer *et al*., 2013; Islam *et al*., 2013; Tesfu and Emana, 2013).

Numerous control methods have been used for the control of *C. chinensis* including the use of larval parasitoids as biological control agents, changes in the temperature of storage house and microwave energy use. Synthetic chemicals use is not promising as this pest feeds internally in the seeds. Similarly, it is impossible to mix synthetic insecticides with food grains due to health threatening chemical residues. The only possible solution is the use of fumigants in the airtight stores. Nevertheless, most of the pulses are stored in homes by the farmers at village level; therefore, fumigants cannot be used as promising agents.

There is a need to find some alternative procedures for the control of *C. chinensis*. These methods should be cheaper, safe to environment and human health and highly effective in use (Park *et al*., 2003; Islam *et al*., 2013; Khan *et al*., 2013; Thein *et al*., 2013). An alternative method found is the use of plant parts and their products as repellents and deterrents such as essential oils and powders of some parts of plants (Sagheer *et al*., 2013; Khan *et al*., 2014). Different spices and medicinal plants have been used for this purpose such as curry, garlic, neem and peppermint (Zia *et al*., 2013; Tuda *et al*., 2014). Extracts of these plants contain volatile compounds that affect insect behavior and physiology (Mbaiguinam *et al*., 2006; Choudhary *et al*., 2014; Sagheer *et al*., 2014).

In this study, experiments have been performed to evaluate the repellent action of *Azadirachta indica* and *Glycyrrhiza glabra* against the control of stored chickpea infesting insect, *C. chinensis*. The experiments were based on the screening of the plant extracts that are potent against the pest of stored commodities. Due to the safe potential of these biocides, these can be utilized as repellents. The basic aim of the experiment was to evaluate the best repellent among the two plant extracts.

Materials and methods

Experiments were performed during the year 2013. All the experiments were conducted under the controlled conditions in Grain Research Training and Storage Management Cell, Department of Entomology, University of Agriculture, Faisalabad.

Insect Collection

Collection of Pulse beetle was made from Grain market, Faisalabad. The attacked pulses were having holes and the upper side of infested bags showed more infestation compared to bottom side. Insects were collected by placing the jar inside the bag and allowing them to move by themselves. In this way, many plastic jars were filled with adults and some infested grains were collected and kept in the jar (Haidri *et al*., 2014).

Insect Rearing

Sterilized chickpea was used as medium for egg laying and rearing. The jars with adults were kept for egg laying. Fifteen pairs were released in every jar. After 5 days, dead as well as alive adults were removed. Eggs were in the form of yellow, shiny and oval structures laid singly on the chickpea. SANYO incubator MIR-254 was used to maintain the uniform conditions of temperature and relative humidity at 27±3°C and 70±5%, respectively. Homogenous population was achieved after 21-30 days (Haidri *et al*., 2014).

Plant Material

Collection of leaves of *Azadirachta indica* and *Glycyrrhiza glabra* was done from the botanical garden, University of Agriculture, Faisalabad.

Preparation of plant powder and extracts

The dried material was grinded in an electric grinder and then sieved to get fine particles. 50 grams of ground sample was allowed to mix with 100 ml of acetone in a conical flask. The covered flasks were then allowed to shake on Rotary Shaker (IRMICO OS-10), adjusted at 120 rpm for 24 hours. After this period, the initial filtrate was obtained by using filter paper. Concentrated stock solution was obtained by subjecting the filtrate to rotary evaporator, as devised by Hasan *et al.* (2005) and Sagheer *et al.* (2013).

Bioassay for percent Repellency

Different concentrations (5, 10, 15, 20%) were made by diluting the concentrated stock with acetone. These dilutions were then applied on the half of filter paper and allowed to dry for 60 minutes. Half of the filter paper was untreated. Thirty adults (1 day old) were released and the plate was covered. Data was taken at 24, 48 and 72 hours. Experiment was replicated three times and Completely Randomized Design (Factorial) was followed. Statistica 7.0 was used for statistical analysis.

Results

Observation lead towards significant repellency of plant extracts towards the adult stage of *Callosobruchus chinensis*.

Repellent effect at different time intervals

Table 1 indicates the effect of plants after three intervals. It was found that *Azadirachta indica* was more repellent in action as compared to *Glycyrrhiza glabra* in all the time intervals.

Table 1. Repellency of two extracts at various exposure periods (24, 48, 72h) against *C. chinensis*

Time	Repellency % (Mean ± Standard error)	
	Azadirachta indica	***Glycyrrhiza glabra***
24	81.67 ± 2.25	14.00 ± 0.39
48	72.50 ± 2.98	12.91 ± 0.34
72	66.67 ± 3.27	11.75 ± 0.28

Repellent effect at different concentrations

Table 2 demonstrates the repellency at four concentrations and it is clearly demonstrated that the repellent action tended to increase with the concentration factor. A highest of 72.77 % repellency was achieved at 20% concentration.

Table 2. Repellency of two extracts at four concentrations (5, 10, 15, 20%) against *C. chinensis*

Concentration	Repellency % (Mean ± Standard error)	
	Azadirachta indica	***Glycyrrhiza glabra***
5	57.22 ± 1.69	11.56 ± 0.37
10	61.11 ± 2.32	13.56 ± 0.29
15	67.77 ± 1.69	14.33 ± 0.47
20	72.77 ± 1.47	17.11 ± 0.42

Discussion

Our experiments proved to be effective in terms of the repellent effect of the plant extracts. It was found the the *Azadirachta indica* was more potent compared to the *Glycyrrhiza glabra*. Kim *et al.* (2003) tested for the effects of some plant extracts against adults of *Sitophilus oryzae* and *Callosobruchus chinensis* and found the definite effect of extracts towards the biology of insects. Our experiments also showed the effectiveness of the plant extracts against the pulse beetle. Elhag (2000) also did experiments towards the deterrent effects of extracts towards the pulse beetle. Evaluation was done for the effect of nine extracts and it was found that the extracts disturbed the biology of insect causing repellent action. In our case, repellent effects of both plant extracts were significant. Chaubey (2013) determined for the effects of essential oils of *Zingiber officinale* (Zingiberaceae) and *Piper cubeba* (Piperaceae) against the cowpea Bruchid. The result suggested the persistence of these extracts towards the biology of storage pest. Further,

the repellent action can be safely used to repel the storage beetles from the stored commodities. Haidri *et al*., (2014) also checked for the effectiveness of *Azadirachta indica* and *Murraya koenigii* towards pulse beetle and found the definite potential of the plants towards causing mortality of test insect. Future research should target the active ingredients that are having less toxicity for humans but have greater repellent action against the insect pests. Our findings predict the potential of plant extracts towards the stored grain insect pest management. Further , there is a need to screen out the active plants that have repellent properties against the insect pests. Various combinations of plant extracts can be used in this regard. Overall, our experiments suggest that *Azadirachta indica* and *Glycyrrhiza glabra* are active plants towards control of stored grain insect pests especially pulse beetle.

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