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Repellent activity of *Argemone mexicana* L. extracts against *Aphis gossypii* Glover and *Tribolium castaneum* (Hbst.) adults

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Abstract
Petroleum ether (Pet. ether), chloroform ($CHCl_3$) and methanol (CH_3OH) extracts of aerial part roots and seeds of *Argemone mexicana* L. were tested for their repellent potentials against *Tribolium castaneum* (Hbst.) and *Aphis gossypii* Glover adults. Against *T. castaneum* the $CHCl_3$ extracts of the aerial part and roots offered moderate repellency at 1% level of significance ($P<0.01$), and the Pet. ether extract of the aerial part, root and seed gave mild repellency at 5% level of significance ($P<0.05$). While, against *A. gossypii* CH_3OH extract of the aerial part showed repellency at 1% level of significance ($P<0.01$), while the Pet. ether and $CHCl_3$ extracts of the aerial part and root; and the CH_3OH extract of the root showed repellency at 5% level of significance ($P<0.05$); and none of the seed extracts gave repellency of *A. gossypii* at all.
According to intensity of repellency the extracts of aerial part is more active than root extracts, and the seed extracts have very less repellent potential against both the test insects.

Keywords: *Argemone mexicana*, *Aphis gossypii* and *T. castaneum*, Repellency

1. Introduction
Argemone. mexicana L. (Papaveraceae) is an annual herb, grows up to 150cm in height. The stem is erect, branched, usually prickly and pale bluish-green in color [1]. It is a weed of most cropping systems [2], especially in Tanzania, Australia and India and Pakistan [3] and in Bangladesh it grows in wheat, sugarcane, potato, pulses and tea fields [4]. In southern India it occurs up to an altitude of 800 m [5]. It is commonly called Mexican poppy, prickly poppy or yellow thistle in English. *A. mexicana* form a leiocarpa which is a form found in West Africa has few or no prickles on the stem, leaves and capsule [6].
This plant is used as a medicinal plant in several countries, and for this purpose the whole plant, seeds, seed oil, flowers, latex, roots and leaves are the organs that are used in the traditional system of medicine [7]. In the folk medicine it is used as an antidote to various poisons. Edible vegetable oil either accidentally contaminated with *A. mexicana* or intentionally adulterated by unscrupulous traders has resulted in epidemic dropsy. Such an epidemic occurred in 1998 in Delhi (India) and epidemic dropsy has also been reported from Nepal [8]. Extracts of *A. mexicana* readily kill the snail *Biomphalaria glabrata* which is the vector for *Schistosoma* spp. and thus have potential as a molluscicide for the relatively cheap control of human schistosomiasis [9].
The present investigation was designed to find out the repellent potential of the crude extracts of the test plant species on two test organisms, *Tribolium castaneum* and *Aphis gossypii*. *T. castaneum* (Hbst.) (Coleoptera: Tenebrionidae) commonly known as 'Rust-red flour beetle, which is the commonest and worldwide pest of wheat-flour [10]. It also attacks pulses, millets and cereals [11]. The red flour beetle is reddish brown in color and has complete metamorphosis (egg, larva, pupa and adult) in their life cycle. In severe infestation, the flour turn grayish and moldy and has a pungent and disagreeable odor making it unfit for human consumption [12]. *A. gossypii* Glover (Hompotera: Aphididae), is an important pest in agriculture, horticulture and greenhouse crops [13-14]. Hence, it can successfully harbor a vast diversity of host plants [15]. Over 320 plant species belonging to 46 families have been documented as suitable host plants for *Aphis gossypii* worldwide [16]. It is pear shaped small, soft-bodied and slow moving yellow or dark green small creatures. It ranges from 0.5-7 mm in length [17]. Most common agricultural insect pest is the aphid, *A. gossypii* and it reduces the yield and quality of crops.

2. Materials and Methods

2.1 Collection and preparation of test materials: *A. mexicana* plants were collected from Chapai Nawabganj, Bangladesh and identified by the Department of Botany, University of Rajshahi where voucher specimens are kept in the Herbarium. Accordingly aerial part, roots and seeds were arranged in separate manner; and the materials were collected separately and excess soil form the roots were removed without washing. The aerial parts and roots portion of plant materials then cut into small pieces using a knife and spread out to dry under shade in a well-ventilated room without heaping the material together. Then materials were powdered by grinder avoiding excess heat (up to 40 ºC) during grinding. The dried ground plant materials were extracted with sufficient amount of solvents (Pet. ether, $CHCl_3$ and CH_3OH) (200g×500ml×3 times). Extracts, thus obtained were filtered and concentrated on a rotary evaporator at 40 ˚C and only as residues (extracts) left were collected in a glass vial and kept in a cool place after labeling.

2.2 Collection and culture of test insects: To carry on tests for repellent activity *T. castaneum,* used in the present experiment were taken from the stock cultures of the Crop Protection and Toxicology Laboratory, University of Rajshahi, Rajshahi-6205, Bangladesh; and *A. gossypii* were collected from the crop field of Institute of Biological Sciences, University of Rajshahi, Bangladesh.

2.3 Repellent activity: The method of repellency test used in this investigation was adopted from the method (No.3) of McDonald *et al*., (1970) [18]. The average of the counts was converted to percentage repellency (PR) using the formula of Talukder and Howse (1993, 1995) [19, 20]: PR = (Nc-5) × 20; where, Nc is the average hourly observation of insects on the untreated half of the disc. A general concentration for each of the extracts (of Pet.E., $CHCL_3$ and CH_3OH) was selected as stock dose for repellency applied against *T. castaneum* adults to make other successive doses by serial dilution to give 0.314, 0.157, 0.079, 0.039 and 0.019mg/cm^2. For *A. gossypii* the doses were established as 0.0831, 0.0416, 0.021, 0.0103, 0.0052mg/cm^2. For the application of *T. castaneum* half filter paper discs (Whatman No. 40, 9cm diam.) were prepared and selected doses of all the extracts separately applied onto each of the half-disc and allowed to dry out as exposed in the air for 20 minutes. Each treated half-disc was then attached lengthwise, edge-to-edge, to a control half-disc with adhesive tape and placed in a Petri dish (9cm diam.). For each of the test samples three replicates were maintained. Being volatile the solvent was evaporated out within a few minutes. Then ten insects were released in the middle of each filter paper circles. Repellency was observed for one-hour interval and up to five successive hours of exposure, just by counting the number of insects from the non-treated part of the filter paper spread on the floor of the 90mm Petri dish. The values in the recorded data were then calculated for percent repellency, which was again developed by arcsine transformation for the calculation of analysis of variance (ANOVA).

For the *A. gossypii* ten adult females from the maintained rearing group on a host plant were taken in one Petri dish (9cm in diam.). Individuals of same were collected for application. Eggplant leaves were trimmed into a particular size (3.5cm diam.) and a piece of water soaked cotton pad was placed at the base of the footstalk of the leaf. Prepared and selected doses of all the extracts separately applied onto each of the pure and fresh half leaf and allowed to dry out as exposed in the air for 20min. The water of the dissolved sample was dry out within a few minutes. On each of the treated a protector transparent plast cap was placed after releasing 10 aphids on the middle. All tests were maintained in three replicates. The tests were carried out in growth chambers set at 25±2 °C, relative humidity (RH) at 70±10%, and a photo phase of 5h. Insects that settled on each half of the leaf were counted after 1h and then at hourly intervals up to 5h.

Table 1: ANOVA results of the repellency of *T. castaneum* by the Pet. ether, $CHCl_3$ and CH_3OH extracts of *A. mexicana* (aerial parts, root and seed)

Name of the test organism	Name of the plant parts of A. mexicana	Name of the extracts of A. mexicana	Sources of variation			F-ratio with level of significance		P- value	
			Between doses	Between time interval	Error	Between doses	Between time interval	Between time interval	Between time interval
T. castaneum	Aerial part	Pet. ether	4	4	16	13.69*	1.651	4.93E-05	0.211
		$CHCl_3$	4	4	16	25.03**	1.481	1.02E-06	0.255
		CH_3OH	4	4	16	1.592	1.952	0.225	0.151
	root	Pet. ether	4	4	16	12.17*	2.897	9.84E-05	0.056
		$CHCl_3$	4	4	16	23.65**	4.455	1.5E-06	0.013
		CH_3OH	4	4	16	1.812	0.766	0.175	0.563
	Seed	Pet. ether	4	4	16	9.033*	0.098	0.0005	0.982
		$CHCl_3$	4	4	16	6.494	2.008	0.003	0.142
		CH_3OH	4	4	16	2.248	1.051	0.109	0.412

* = (P <0.05) **= (P <0.01)

Table 2: ANOVA results of the repellency of *A. gossypii* by the Pet. ether, $CHCl_3$ and CH_3OH extracts of *A. mexicana* (aerial parts, root and seed)

Name of the test organism	Name of the used parts of *A. mexicana*	Name of the extracts of *A. mexicana*	Sources of variation			F-ratio with level of significance		P- value	
			Between doses	Between time interval	Error	Between doses	Between time interval	Between time interval	Between time interval
A. gossypii	Aerial part	Pet. ether	4	4	16	13.68*	1.651	4.93E-05	0.211
		$CHCl_3$	4	4	16	9.259*	6.033	0.0004	0.004
		CH_3OH	4	4	16	23.635**	3.785	1.51E-06	0.024
	root	Pet. ether	4	4	16	12.29*	3.265	9.27E-05	0.0387
		$CHCl_3$	4	4	16	12.84*	2.633	7.2E-05	0.073
		CH_3OH	4	4	16	12.01*	5.305	0.0001	0.006
	Seed	Pet. ether	4	4	16	2.445	0.516	0.089	0.725
		$CHCl_3$	4	4	16	2.445	0.516	0.089	0.725
		CH_3OH	4	4	16	5.315	3.439	0.0064	0.033

* = (P <0.05) **= (P <0.01)

3. Results and Discussion

The Pet. ether, $CHCl_3$ and CH_3OH extracts of *A. mexicana* showed repellency to *T. castaneum* and *A. gossypii* adults. Against *T. castaneum* the $CHCl_3$ extracts of the aerial part and roots offered moderate repellency at 1% level of significance (P<0.01), and the Pet. ether extract of the aerial part, root and seed gave mild repellency at 5% level of significance (P<0.05). However, the $CHCl_3$ extract of seeds and the CH_3OH extracts of the aerial part, roots and seeds didn't show any repellency. According to intensity of repellency against *T. castaneum* the result could be arranged in a descending order: aerial part ($CHCl_3$), root ($CHCl_3$) > aerial part (Pet. ether), root (Pet. ether), seed (Pet. ether) extracts of *A. mexicana*. While, against *A. gossypii* CH_3OH extract of the aerial part showed repellency at 1% level of significance (P<0.01), while the Pet. ether and $CHCl_3$ extracts of the aerial part and root; and the CH_3OH extract of the root showed repellency at 5% level of significance (P<0.05); and none of the seed extracts gave repellency of *A. gossypii* at all. According to intensity of repellency *A. mexicana* extracts to *A. gossypii* the result could be arranged in a descending order: aerial part (CH_3OH)> root (Pet. ether, $CHCl_3$, CH_3OH), aerial part (Pet. ether, $CHCl_3$) extracts.

Though works on repellency by *A. mexicana* is scanty it was found tested on *T. castaneum* [21] and the plant powder was found repellent. The present work was carried out against *T. castaneum* and A. gossypii to yield promising repellency. Bhumij tribes in Odisha, India use seed oil of *A. Mexicana* as mosquito repellent [22]. Since malaria is very common in most of the tribal dominated areas of India and Bangladesh, development of low cost herbal mosquito repellents will save the precious life of many poor peoples. Repellency is the system tends to dissuade pests away from a susceptible crop (repellent) [23] what can be called as a push approach. Thus, plants are natural source of these repellent agents, reported innumerou ethnobotanical information. Plant-derived repellents usually do not pose hazards of toxicity to humans and domestic animals, and are easily biodegraded and compared to synthetic compounds, natural products are presumed to be safer for humans. This study was attempted to highlight *A. mexicana* claimed to be used or associated with insect repellent activity, and it was found considerable. However, test result on other attributes also support the present finding, such as oviposition altering and ovicidal efficacy of the root extracts of *A. mexicana* against mosquito was also established [24, 25].

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