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EXTRACT OF BITTER MELON *CITRULLUS COLOCYNTHIS* LEAVES AS A PROMISING INSECTICIDE AGAINST THE GRAIN WHEAT APHID *SCHIZAPHIS GRAMINUM*

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ABSTRACT

This study aimed to evaluate the biological effect of bitter melon *Citrullus colocynthis* leaf extracts on the mortality using hot water and organic solvent extracts (ethyl alcohol, hexane) effects on the nymph, winged, and nonwinged adults of the wheat grain aphid *Schizaphis graminum* were evaluated. Results indicated that the ethyl alcohol extract is more effective than the hexane and hot water extracts on mortality rates. The highest mortality rate of the nymphs was caused by the ethyl alcohol extracts (75.63, 59.20, and 53.49%) at a concentration of 0.7 g/ ℓ, respectively.

Key words: *Citrullus colocynthis, Schizaphis graminum,* grain wheat aphid, plant extract, ecofriendly pesticides, hot water and solvent extracts, mortality, nymphs, winged adults, alale adults

The wheat is one of the major cereal crops and its yield is affected by insect pests (Mohanty et al., 2016). The insect primarily resides on the lower surface of leaves and leading to wilting and drying of leaves and consequently, the yield decreases. Additionally, aphids are significant carriers of various viruses, with the most notable one being the virus that causes stunting and yellowing in barley (Oelbermann and Scheu, 2009; Zhang et al., 2022). Pesticides have been used to control this pest however, these pesticides have several harmful effects. This has prompted researchers to seek out more appropriate, environmentally friendly, easy-to-use, and inexpensive alternatives, such as plant extracts (Zhang et al., 2022; Hussain et al., 2024). This medicinal plant in large areas in the Western Badia in the province of Muthanna southwest of Iraq as a natural desert plant. This study evaluates leaf extracts of this plant occurs as alternative to chemical pesticides.

MATERIALS AND METHODS

Nymphs and mature individuals of the wheat grain aphid *Schizaphis graminum* were obtained from wheat and barley fields in the Wadi Farz and Al-Rehab areas of the Badia of Muthanna Governorate in Iraq. Aphids were carefully collected from plants and sorted based on their developmental stages before being placed in polyethylene bags. The insect was identified as *S. graminum*, using the taxonomic key (Toba, 1964). It was reared in plastic pots on wheat plants with three or more leaves, in a greenhouse. The pots were covered with tulle cloth. This ensured the production of multiple generations. The adult aphids from these plants in an incubator (27 ±2 °C, 65± 5% RH, lighting schedule of 12 hr of light, followed by 12 hr of darkness) (Pourya et al., 2020). The bitter melon plant's leaves were gathered from the Al-Nadwa region in the Western Samawa Badia in April 2023. The plant's leaves were washed with sterile water and cut and dried in a shaded area. They were then finely powdered using a domestic grinder and stored in polyethylene bags were stored at 4°C. The plant *C. colocynthis*, was identified by the Iraqi National Herbarium/ Seed Inspection and Certification Department of the Iraqi Ministry of Agriculture, as documented in the Iraqi Botanical Encyclopedia.

Preparation of leave extract of *C. colocynthis* was done in the laboratory using hot distilled water, where 100 g of dried leaf powder was taken and placed in a glass flask of 500 g containing 200 ml of hot distilled water, mixing the leaf powder with hot water by an electric mixer for 20 min and then leaving the mixture for another 30 min to settle down, then filtering the fresh filtrate to a volume of 200 ml and storing at 4 °C until use. 0.5 ml of Surfix was added as a diffuser and adhesive to 100 ml of pre-prepared hot water extract, and the concentration was prepared (0.7, 1.4, and 2.1%).

The control treatment was sterile distilled water only. Leaf extract using organic solvents was done with two organic solvents; ethyl alcohol, a polar solvent, and hexane, a non-polar solvent. 50 g of bitter melon plant powder in a Soxhlet extraction device was used. To this powder, 200 ml of each organic solvent (ethyl alcohol and hexane) were added. The extraction process lasted for 8 hr. After completing the extraction process, extract was concentrated using rotary evaporator. Then the concentrations (0.7, 1.4 and 2.1%) were prepared. For the control treatment, 1.5 ml of each (ethyl alcohol and hexane) was used, and the volume was completed to 100 ml by adding 98.5 ml of sterile distilled water. Laboratory study was conducted to examine the impact of these extracts on different stages of *S. graminum*, including nymphs, winged pupae, and non-winged-adults. Ten aphids were placed in a 9 cm dia petri dish, which was perforated with a needle to allow for ventilation. The insects were subjected to the predetermined concentrations with three replicates for each treatment. These data were sujected to a two-way ANOVA using Genstat 12th Edition. Tukey's honestly significant difference (HSD) and the least significant difference (LSD) were used to evaluate means, after applying Abbott's correction.

RESULTS AND DISCUSSION

Study evaluated the effectiveness of *C. colocynthis* leaf extract on the mortality rate of the nymphal stage and winged, and nonwinged adults, stages. Results showed that all concentrations were effective. Mortality rate of the first-stage nymphs exhibited variation in response to the concentrations; 2.1% gave superior efficacy. Mortality was 52.27, 66.85, and 75.63% for hot water extract, hexane, and ethyl alcohol, respectively, 0.7% exhibited the lowest mortality rate (Table 1). Exposure time to the leaf extracts showed significant variations across the three types of extracts (hot water, hexane, and ethyl alcohol). The interaction between the duration of exposure and concentration exhibited a substantial variations. There was a significant effect of *C. colocynthis* leaf extracts on the cumulative mortality of wingless adults of *S. graminum* at different concentrations. The concentration had a significant effect on the mortality. The concentration of 2.1% gave maximum mortality (47.88, 53.49, and 59.20%) with hot water extract, hexane, and ethyl alcohol, respectively. Also mortality increased with exposure time and was 59.07% after 96 hr with ethyl alcohol extract, compared to 40.73% after 24 hr. Similarly, the trend was noted with hexane and hot water extracts. Significant interaction between the concentrations and the exposure time was also observed.With winged adult ethyl alcohol extract performed better than others (53.49, 44.68 and 38.05%), respectively at 2.1% after 96 hr of treatment with maximum mortality. The results revealed significant difference in the time of exposure to the extracts. Statistical analysis indicated significant differences between the time of exposure and the concentrations.

The motile may be due to their sensitivity to toxic substances found in the leaves of *C. colocynthis*, or due to poisoning of the digestive system. Poisoning the digestive tract responsible for absorption and decreased efficiency of food conversion, as phenolic compounds bind with proteins, forming complexes with proteins by hydrogen bonds that are difficult to digest (Farooq et al., 2016) or that the immature stages refrain from feeding because of their exposure to the extract and then die (Stevenson et al., 2017). The death may be a result of the effect of the active compounds on the body wall or digestive system (Kumar and Singh, 2015; Foster et al., 2014; Khan et al., 2009. The plant extracts showed some phytochemical effect on *S. graminum* (Abdullah et al., 2024).

Thus, use of alternative active substances as pesticides help in control of pests resisting various pests. The use of *C. colocynthis* medicinal extracts, which are known to contain various parts of the plant on many effective compounds with a medicinal effect and biological effect in increasing the rates of insect mortality, may contribute a lot.

AUTHOR CONTRIBUTION STATEMENT

ANA, MKA and MHH conceived of the original idea. ANA and MKA developed the theoretical and performed the statistical analysis for experimental data. MKA and MHH verified the analytical methods. MHH, MKA and ANA worked for lab analysis and supervises the project. MHH, MKA and ANA discussed the results and contributed to the wrote the manuscript.

CONFLICT OF INTEREST

No conflict of interest

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Table 1. Effect of concentrations of *C. colocynthis* leaf extract *S. graminum*

Extract type	Concentration g/ l	Exposure time (hr) 24	48	72	96	Concentration
First nymphal instar						
Hot water	0.0	0.0	0.0	0.0	0.0	0.00
	0.7	37.97	41.91	44.81	47.43	47.43[a]
	1.4	38.79	43.14	46.86	51.18	49.31[b]
	2.1	46.83	52.02	54.92	58.20	52.27[c]
Exposure time		41.79[a]	45.69[b]	48.86[c]	52.27[d]	
LSD (p<0.05)	Exp. x Cons = 0.549		Exp. Time = 0.289		Cons. = 0.209	
Hexane	0.0	0.0	0.0	0.0	0.0	0.0
	0.7	42.64	57.17	59.13	62.72	55.41[a]
	1.4	49.24	59.24	62.46	65.17	59.03[B]
	2.1	53.42	67.97	71.62	74.39	66.85[C]
Exposure time		48.43[a]	61.46[b]	64.40[c]	67.43[D]	
LSD (p<0.05)	Exp. x cons = 0.730		Exp. time = 0.378		Cons. = 0.332	
Ethyl alcohol	0.0	0.00	0.00	0.00	0.00	0.00
	0.7	50.58	67.97	72.53	76.08	66.79 [a]
	1.4	54.86	72.99	77.50	80.24	71.40b
	2.1	57.45	75.43	83.40	86.25	75.63 [c]
		54.30[a]	72.13[b]	77.81[c]	80.86[d]	
LSD (p<0.05)	Exp. x cons = 1.0790		Exp. time = 0.6170		Cons. = 0.5337	
Apterous adult						
Hot water	0.0	0.0	0.0	0.0	0.0	0.00
	0.7	25.95	31.03	37.25	43.95	34.55[a]
	1.4	34.85	38.94	42.27	47.43	40.87[b]
	2.1	40.65	46.99	50.74	53.12	47.88[c]
Exposure time		33.82[a]	38.99[b]	43.42[c]	48.21[d]	
LSD (p<0.05)	Exp. x cons = 2.7259		Exp. time =1.5587		Cons. = 1.3482	
Hexane	0.0	0.00	0.00	0.00	0.00	0.0
	0.7	40.04	46.80	53.00	59.88	0.7
	1.4	42.97	59.93	63.91	66.13	1.4
	2.1	50.94	67.16	70.99	73.53	2.1
Exposure time		26.78[a]	29.32[b]	36.07[c]	41.00[d]	
LSD (p<0.05)	Exp. x cons = 0.385		Exp. time =0.2792		Cons. = 0.249	
Ethyl alcohol	0.0	0.0	0.0	0.0	0.0	0.0
	0.7	35.75	38.96	44.92	48.54	42.04[a]
	1.4	39.55	55.74	58.89	62.56	54.18[b]
	2.1	46.91	59.87	63.91	66.13	59.20[c]
		40.73[a]	51.52[b]	55.90[c]	59.07[d]	
LSD (p<0.05)	Exp. x cons = 1.088		Exp. time = 0.2792		Cons. = 0.249	
Winged adult						
Hot water	0.0	0.0	0.0	0.0	0.0	0.00
	0.7	22.18	21.87	30.27	34.88	27.3[a]
	1.4	27.22	31.88	37.00	42.03	34.53[b]
	2.1	30.94	34.21	40.96	46.09	38.05[c]
Exposure time		26.78a	29.32b	36.07c	41.00d	
LSD (p<0.05)	Exp. x cons = 2.7259		Exp. time =1.5587		Cons. = 1.3482	
Hexane	0.0	0.0	0.0	0.0	0.0	0.0
	0.7	24.86	27.10	33.33	36.93	30.55a
	1.4	32.46	33.53	40.65	48.01	38.66b
	2.1	37.82	32.93	44.98	63.02	44.68c
Exposure time		26.78[a]	29.32[b]	36.07[c]	41.00[d]	
LSD (p<0.05)	Exp. x Cons = 2.7259		Exp. Time =1.1161		Cons. = 0.9654	
Ethyl alcohol	0.0	0.00	0.00	0.00	0.00	0.00
	0.7	29.60	34.04	39.50	56.23	39.84[a]
	1.4	35.55	44.23	43.82	60.98	46.14[b]
	2.1	42.76	55.91	47.97	67.35	53.49[c]
		26.98[a]	32.82[b]	33.54[c]	46.14[d]	
LSD (p<0.05)	Exp. x Cons = 0.6814		Exp. time = 0.3896		Cons. = 0.3370	

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