

Evaluation of some Plants Extracts in Reducing Density of Two Spotted Spider Mite *Tetranychus Urticae* on Eggplant

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ABSTRACT

This study was aimed to evaluating the role of Acramite acaricide in normal and nanosized in addition to extracts of sour orange peels *Citrus aurantium*, harmal seeds *Peganum harmala* and sidr leaves *Ziziphus spina-christi* in reducing the population density of two-spotted spider mite (TSSM) *Tetranychus urticae* and the effect of prior treatments in decreasing the yield loss percentage in the eggplant crop. The cumulative relative efficiency for Acramite nano size 0.4 g/L was 87.7% with significantly outperformed from all other treatments, followed by Acramite nano size 0.2 g/L and Acramite normal size that was 82% and 73.2%, respectively, while was 70%, 67% and 66.8 for alcoholic harmal extract, essential oil of sour orange and alcoholic sidr extracts, respectively

Key words: cumulative relative efficiency, harmal, sidr, nanosized, sour orange



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INTRODUCTION

Eggplant is a vegetable crop that is grown all over the world and can provide nutritional benefits because of its content of vitamins, phenols, and antioxidants (Sharma, M., & Kaushik, P. 2021). In addition, eggplant has potential therapeutic applications that are only recently being identified (Gürbüz et al., 2018). Based on data from 2022, the global production of eggplant is around 59 million tons annually, with a net value of more than 12 billion US\$ a year (FAO, 2022). Several pests significantly limit eggplant output; the most common are the leafhopper, whitefly, aphid, and two-spotted spider mite (Srinivasan, 2009). The two-spotted spider mite *Tetranychus urticae* Koch, 1836, is one of the most important agricultural pests (Jakubowska et al., 2022), not only because of the harm it does, but also because of its extensive host range (Tsagkarakou et al., 2002). The TSSM's rapid developmental pace and strong reproductive capacity allow it to establish destructive population levels quickly when growth conditions are favorable, resulting in a rapid decline in host plant quality (Razmjou et

al., 2009) The use of synthetic acaricides is primarily relied upon to control *T. urticae*. On the other hand, *T. urticae*, has rapidly developed resistance to the majority of the acaricides available for control (Abdelgaleil et al., 2019) due to its very broad host plant range, extremely short lifecycle, high fecundity, arrhenotokous parthenogenesis, and overwintering strategy (Adesanya et al., 2021). Choi et al. (2004) Observed that the use of 53 essential oils extracted from various plants, including clove, eucalyptus, citrus, lemongrass, and other plants, resulted in high mortality rates in the two-spotted spider mite under laboratory conditions after these oils were applied to the mites. It has been found that the use of sandalwood oil had a significant effect on the two-spotted spider mite, with a mortality rate reaching 87.2% under laboratory conditions, additionally, the use of the oil prevented egg-laying, with an egg-laying inhibition rate of 89.3% on the oil-treated leaves (Roh et al., 2011). Nano pesticides are formulations composed of active ingredients and excipients at the nanometer scale, the aim of preparing and using nanopesticide

formulations is to increase the solubility of active ingredients, enhancing their ability to penetrate and enter the bodies of target organisms and resist the defenses of the pest, as well as controlling their slow release towards the target pest, additionally, they protect the active ingredients from degradation factors such as temperature, light, rain, and others (Reeves, 2014). In this study, we employed aqueous and alcoholic extracts of Harmal seeds and Sidr leaves, an aqueous extract and essential oil of sour orange, and Acramite acaricide in normal and nano volumes to assess their ability to reduce TSSM population density on eggplant in a greenhouse.

MATERIALS AND METHODS

We chose Jawaher eggplant cultivar F1 from Enza Zaden, Netherlands, to conduct the experiment. The experiment took place in a greenhouse in the College of Agricultural Engineering Sciences, University of Baghdad, Baghdad Province, Jadriya, in 2024.

Extracts preparation

Aqueous extracts=100 grams each of sour orange peels, harmal seeds, and sidr leaves were ground using an electric grinder, then placed in separate 2-liter triangular glass flasks. Each was added to one liter of distilled and boiled water with continuous stirring. The flasks were then sealed with glass stoppers and wrapped in cellophane to reduce evaporation. They were left for 6 to 8 hours, then filtered through a sieve with 100-microliter diameter holes, followed by another sieve with 56-microliter diameter holes. The filtrate was then poured into test tubes to separate the clear liquid using a centrifuge at 5000 RPM for 10 minutes. The clear liquid was carefully poured into Pyrex glass dishes and left for 48 hours in a vacuum dryer to obtain the extract in its dry form. The dishes are scraped with a sterile metal spatula and stored in airtight glass bottles in the refrigerator (Al-Manhel & Niamah, 2015).

Alcoholic extracts

100 grams of both harmal seeds and sidr leaves were taken and ground using an electric grinder. They were then placed in separate 2-liter triangular glass flasks, each with one liter of 70% ethyl alcohol added, with continuous

stirring. The flasks were sealed with glass stoppers and wrapped in cellophane to reduce evaporation. They were left for 24 hours, then filtered using a sieve with 100-microliter diameter holes, and then filtered again with another sieve with 56-microliter diameter holes. The filtrate was then poured into test tubes for separation using a centrifuge at 5000 RPM for 10 minutes. The clear liquid was carefully poured into Pyrex glass dishes and left for 48 hours in a vacuum oven to obtain the extract in its dry form. The dishes are scraped with a sterilized metal spatula and stored in airtight glass bottles in the refrigerator (Wendakoon et al., 2012; Zamin et al., 2014).

Extraction of essential oil

100 grams of dried sour orange peels were ground using an electric grinder and the powder was placed in a round glass flask with one liter of distilled water. The solution was mixed regularly to ensure the powder blended with the water and did not stick to the bottom of the flask. It was then placed on an electric heater within a Clavenger apparatus until boiling. The oil that forms and separates from the water at the top was collected using an opaque and airtight glass bottle with a metal cap to avoid the oil reacting with the cap, and it is stored in the refrigerator until it is used (Gómez-Velázquez et al., 2021).

Formulation of Nano capsule

75 g of polyethylene glycol 4000 (Loba chemie Pvt, India) was dissolved at 65° C, added 25 ml of Acramite acaricide (Bifenazate 480 g/L, Arysta, Belgium) with continuously heating and stirring by using a heating magnetic stirrer, then put the solution in the freezer until stiffened, then grind it by grinder to obtain a white powder (Hammed et al., 2023).

Characterizations of Nano capsule

Nano capsule shape and size were measured by Mira 3 field emission scanning electron microscopy (FE-SEM), Tescan, Korea. To determine the stability and activity of Nano capsule Zeta potential analysis was made by a HORIBA Zetasizer Nano-SZ-100V2 (HORIBA Scientific Instruments, Japan) (Al Hilfy et al., 2022).

Field experiment

Aqueous and alcoholic extracts of Harmal seeds and Sidr leaves, an aqueous extract and essential oil of sour orange, and Acramite acaricide in both size normal and Nano were used to evaluate its roles in reducing infestation of TSSM on eggplant cultivar Jawaher. To assess the effect of previous extracts and Acramite acaricide six plants were selected for each treatment in addition to the control treatment, depending on primary tests (for extracts) and company recommendations (for Acramite acaricide) concentrations used, aqueous and alcoholic extracts of Harmal seeds and Sidr leaves concentrations was 2g/100 ml and 1g/100ml for both plant, respectively, an aqueous extract and essential oil of sour orange concentrations was 2g/100 ml, and 1ml/100ml, aqueous and alcoholic extracts of Harmal seeds and Sidr leaves concentrations was 2g/100 ml and 1g/100ml for both plant, respectively, Acramite acaricide concentrations was 0.2 ml/L for normal volume, 0.4 g/L and 0.2 g/L for nano volume. The experimental units (plant) of each treatment were sprayed with a extracts and acaricide solution and control treatments sprayed by water only, the treatment plants were observed 1, 3, 7 and 14 day after application to determine the population density of TSSM.

Loss percentage of eggplant yield assessment: To calculate the economic loss in eggplant yield caused by two-spotted spider mite infestation, take eggplant yield every three days when they reach the appropriate

size for marketing from every previous treatment's experimental units (plant) in addition to the exclusion treatment (without infestation by applying acaricides to prevent infestation). The yield was weighed, and the average production rate per plant was calculated by dividing the total weight by the total number of plants. After weighing the eggplant harvest for healthy and infested plants, the proportion of economic loss owing to mite infestation in the crop was calculated using the analytical procedures developed by Judenko (1973) under field conditions.

Statistical analysis: The experiments were carried out according to factorial experiments using a completely randomized block design (CRBD), and the differences between the means of the treatments were tested according to the value of the least significant difference (L.S.D) at the probability level of 0.05. The results were analyzed by the statistical program Genstat (2009). Corrected relative efficiency calculated by Henderson and Tilton (1955) formula.

RESULTS AND DISCUSSION

Scanning Electron Microscopy (SEM) analysis: Analysis in (Figure 1), shows the diameter and shapes of Acramite Nano capsule. The diameter average of Acramite Nano capsule was 75 nm with oval shape.

Zeta potential analysis: The zeta potential analysis in (Figure 2), reveal that zeta potential mean of Acramite Nano capsule was -78.8 mV.

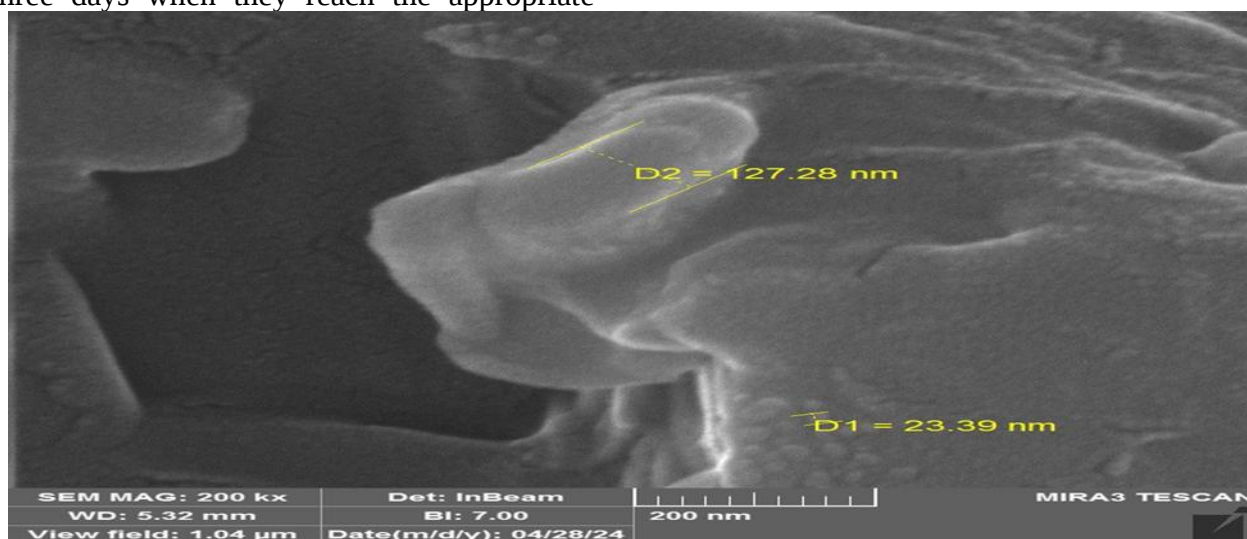


Figure 1. SEM images of Acramite Nano capsule

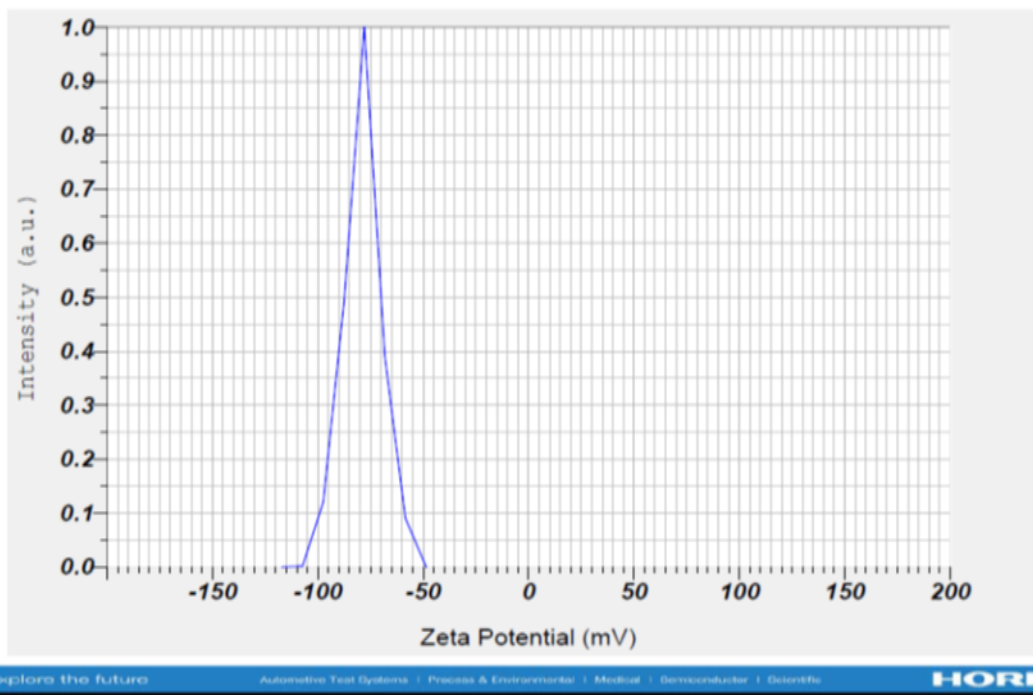


Figure 2. Zeta potential means of Acramite Nano capsule

Effect of extracts and Acramite on TSSM population density: Table (1) shows that the cumulative relative efficiency of Acramite nano volume in concentration 0.4 g/L was 87.7% and significantly outperformed all other treatments, followed by Acramite nano volume in concentration 0.2 g/L, which had a cumulative relative efficiency of 82%. The cumulative relative efficiency of the remaining treatments was as follows: the normal volume of Acramite was 73.2%, and that of alcoholic harmal was 70%, and there is no significant difference between them. However, the normal volume of Acramite significantly exceeds essential oil of sour orange 67% and alcoholic sidr 66.8%, and all previous treatments significantly outperformed all aqueous extracts. The use of pesticides in nano volume increases its efficiency because of the role of the nanocapsule in protecting active ingredients from harsh environmental conditions and the slow release of pesticides; hence, the pesticide in nano volume stays longer than normal volume. In this case, the effect of it remains longer than normal volume (Kumar et al., 2019; Wang et al., 2022). The nano size (75 nm) and zeta potential value of -78.8 mV contribute to Acramite nano volume's great efficiency and stability (Clogston &

Patri, 2011; Rai & Ingle, 2012). The harmal *Peganum harmala* has a number of active compounds that impact arthropods, including harmine, harmaline, vasicinone, and deoxyvasicinone, these chemicals have demonstrated substantial action, including impacts on arthropods such as ticks and mites, and methanol extracts of *P. harmala* seeds showed acaricidal properties against *Rhipicephalus microplus* (Zhu et al., 2022). Previous compounds gave harmal extracts the ability to lower TSSM population density in both aqueous and alcoholic forms. Limonene accounts for 90.25% of the volatile compounds in sour orange peel (Karoui & Brahim, 2013). Limonene was found to have high acaricidal activity against adult females of the two-spotted spider mite, *Tetranychus urticae*, through fumigant and contact effects (Abdelgaleil et al., 2019). This explains why the aqueous extract and essential oil of sour orange peel were effective in lowering TSSM population density. The main phenolic compounds of sidr leaves are tyrosol, hydroxytyrosol, P-hydroxybenzoic acid and vanillic acid (AL-Marzooq, 2014). Previous compounds in sidr leaf extracts may affect TSSM and lead to reducing its density.

Table 1. Relative efficiency of extracts and Acramite acaricide

	Treatments	concentra tions	% Corrected relative efficiency				%Cumulative relative efficiency
			After 1 day	After 3 days	After 7 days	After 14 day	
1	Aqueous Harmal	2 g/100 ml	81.1	76.3	22.0	0	44.9
2	Alcoholic Harmal	1 g/100 ml	98	95.8	83.8	3.2	70
3	Aqueous Sidr	2 g/100 ml	57	51.8	4.8	0	28
4	Alcoholic Sidr	1 g/100 ml	97.2	91.4	78.7	0	66.8
5	Aqueous sour orange	2 g/100 ml	89	72.9	54.2	0	54
6	Essential oil of sour orange	1 ml/100 ml	97.8	92.7	77.4	0	67
7	Acramite normal volume	0.2ml/L	98.1	95.6	92.6	6.4	73.2
8	Acramite nano volume	0.4g/L	99.5	97.9	95.4	58	87.7
9	Acramite nano volume	0.2g/L	99	97.8	94	37	82
Least significant difference (L.S.D) under significance level 5%							
For treatment: 1.06 for duration: 0.67 for interaction: 2.01							
For Cumulative relative efficiency: 3.9							

Effect of extracts and Acramite acaricide on loss percentage of eggplant yield: Table (2) shows that all treatments, except the aqueous sidr treatment, significantly reduced the percentage loss in eggplant yield; the loss rate reached 12.4%, 7.9%, 15.7%, 16.3%, 20.0%, 22.5%, 26.8%, and 27.4% for Acramite normal volume, Acramite nano volume 0.4 g/L, Acramite nano volume 0.2 g/L, essential oil of sour orange, alcoholic harmal, aqueous sour orange, aqueous harmal, and alcoholic sidr, respectively, while the control loss rate was 41%. All preceding treatments significantly exceed the control treatment. The loss percentage for aqueous sidr treatment was 38.9% with no significant difference from the control treatment. Acramite normal volume and Acramite nano volume 0.4 g/L

significantly exceed all other treatments with no significant difference between them. No significant difference between Acramite nano volume 0.2 g/L and essential oil of sour orange with significant difference from alcoholic harmal, aqueous sour orange, aqueous harmal, alcoholic sidr and aqueous sidr treatments. There is no significant difference between alcoholic harmal and aqueous sour orange treatments, but both of them significantly exceed aqueous harmal, alcoholic sidr, and aqueous sidr treatments. These findings represent the efficacy of acaramite acaricide and other plant extracts, demonstrating that when the population density of TSSM declines, so does its harm, resulting in a reduction in yield loss.

Table 2. Economic loss in eggplant yield after treatment with plant extracts and Acramite acaricide

	Treatments	concentrations	Expected production gm/plant	Expected production Kg/donum	Actual production gm/plant	Actual production Kg/donum	Actual loss gm/plant	Actual loss Kg/donum	loss Percentage %
1	Aqueous harmal	2 g/100 ml	1372	10976	1005	8040	367	2936	26.8 %
2	Alcoholic harmal	1 g/100 ml	1372	10976	1097	8776	275	2200	20.0%
3	Aqueous sidr	2 g/100 ml	1372	10976	839	6712	533	4264	38.9%
4	Alcoholic sidr	1 g/100 ml	1372	10976	996	7968	376	3008	27.4%
5	Aqueous sour orange	2 g/100 ml	1372	10976	1064	8512	308	2464	22.5%
6	Essential oil of sour orange	1 ml/100 ml	1372	10976	1149	9192	223	1784	16.3%
7	Acramite normal volume	0.2ml/L	1372	10976	1202	9616	170	1360	12.4%
8	Acramite nano volume	0.4g/L	1372	10976	1264	10112	108	864	7.9%
9	Acramite nano volume	0.2g/L	1372	10976	1157	9256	215	1720	15.7%
10	Control		1372	10976	810	6480	562	4496	41%
LSD values					37.5	69.2	15.7	58.0	3.2

Least significant difference (L.S.D) under significance level 5%

CONCLUSION

It could be concluded that the application of Acramite acaricide and plant extracts can reduce the population density of TSSM and decrease yield loss; the treatments effect begins high and decreases with time.

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AUTHOR/S DECLARATION

We confirm that all Figures and Tables in the manuscript are original to us. Additionally, any Figures and images that do not belong to us have been incorporated with the required permissions for re-publication, which are included with the manuscript.

Author/s signature on Ethical Approval Statement.

Ethical Clearance and Animal welfare

Funds:

AUTHOR'S CONTRIBUTION STATEMENT

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كفاءة بعض المستخلصات النباتية في خفض كثافة الحلم ذي البقعتين *Tetranychus urticae* على الباذنجان

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المستخلص

هدفت هذه الدراسة لتقييم فاعلية مبيد الحلم اكراميت بالحجمين العادي و النانوي بالاضافة الى مستخلصات قشور النارنج *Citrus aurantium* ، بذور الحرمل *Peganum harmala* واوراق السدر *Ziziphus spina-christi* في خفض الكثافة السكانية للحلم ذي البقعتين و تاثير المعاملات السابقة في تقليل الخسارة الاقتصادية في الحاصل لنبات الباذنجان. بلغت الكفاءة النسبية التراكمية للمبيد اكراميت بالحجم النانوي للجرعة 0.4 غم/ لتر 87.7% اذ تفوقت معنويا على جميع المعاملات الاخرى، تلتها معاملة المبيد بالحجم النانوي للجرعة 0.2 غم/لتر و معاملة المبيد اكراميت بالحجم العادي اذ بلغت الكفاءة النسبية التراكمية 82% و 73.2% على التوالي، بينما بلغت الكفاءة النسبية التراكمية 70%، 67% و 66.8% لكل من مستخلص بذور الحرمل الكحولي، الزيوت الطيارة لقشور النارنج و المستخلص الكحولي لاوراق السدر على التوالي.

الكلمات المفتاحية: الكفاءة النسبية التراكمية، الحرمل، السدر، الحجم النانوي، النارنج.