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Effect of Ultraviolet UV-A Irradiation on the Efficacy of Three Acaricides for Mortality and Egg Hatching of Spider Mite: *Tetranychus Urticae* in the Laboratory

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Abstract: This study examined the influence of three acaricides, Bifenazate, Clofentezine, and Spirodiclofen, exposed to artificially ionizing UV-A irradiation for four duration times: 30, 60, 90, and 120 minutes on mortality of the herbivorous mite *Tetranychus urticae* in the laboratory. Guava leaves were used as a test fruit substrate to assess the lethality effect of acaricides on mites. The study demonstrated that UV-A irradiation reduced the action of active ingredients of tested acaricides and produced less toxic products, decreasing mite individuals' mortality. Lethality in *T. urticae* was reduced to 77.5% compared to the control ($P < 0.05$), after spraying with 30 minutes of UV-A irradiated Bifenazate, while it was 40% in Clofentezine and 27% in Spirodiclofen. After a 120-minute exposure period, Bifenazate demonstrated 62.5% lethality. In the same time interval, the lethality of mites was significantly decreased, as recorded at 22.5% in Clofentezine and 12.5 % in Spirodiclofen ($P < 0.05$). These results suggest that the effectiveness of acaricides in killing adult mites decreases with prolonged exposure to UV-A irradiation [Pearson's correlation coefficient ($R= 0.84$, $P < 0.05$)]. Regarding egg viability, control treatments without UV-A exposure showed a percentage viability of 5-19%, while the treated groups exhibited a viability range of 38-53% during the 30-minute irradiation. Two hours of irradiated acaricides caused highly surviving mite individuals and extremely viable eggs compared to the control. In conclusion, the random application of acaricides in some regions during sunny periods is ineffective for optimal pest control.

Key words: Ionizing radiation, Two-spotted spider mite, Acaricides, Lethality, Egg viability.

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1. Introduction

The two-spotted spider mite, *Tetranychus urticae* (Acari: Tetranychidae), is a worldwide and economically major pest with a short life cycle that causes economic loss in crops, including vegetables and

fruit trees. This mite tends to infest the leaves of various trees, including plum, guava, apricot, and apple trees (**Awad *et al.*, 2018 and Eman *et al.*, 2022).**

Electromagnetic radiation, known as UV irradiation, has wavelengths ranging from 10 to 400 nm. This radiation can be categorized into several types: UV-A (315–400 nm), UV-B (280–315 nm), and UV-C (200–280 nm). Importantly, UV-C and much UV-B radiation are absorbed by the atmosphere and ozone layer, meaning that the solar UV radiation reaching the ground predominantly consists of UV-A, with only a small amount of UV-B **(Ballare´ *et al.*, 2001).**

Few studies have addressed the effect of radiation on pesticides, and no studies were done on acaricides. Findings of previous studies suggest that ultraviolet irradiation can reduce the toxicity of agrochemical compounds, diminishing their persistence in agricultural products. These studies have shown that UV irradiation can disrupt the chemical bonds of active ingredients in these chemicals (**Bustos *et al.*, 2019; Ahmed, 2022 and Lakshmipathy *et al.*, 2024)**. Our study focuses on the influence of UV-A irradiation that is not absorbed by the atmosphere and the ozone layer on acaricides sprayed on host leaves. This effect is particularly evident when certain farmers in some regions apply these chemicals randomly around noon. The study investigated three acaricides used to control pest populations in agriculture to determine whether this irradiation affects the lethality and egg hatchability of *T. urticae* when reared on guava leaves in the laboratory.

2. Material and Methods

2.1. Culturing technique

The susceptible strain of *Tetranychus urticae* was obtained from the Plant Protection Research Institute Acarology laboratory. The two-spotted spider *T. urticae* mite was reared in the laboratory following the methods outlined by **Awad *et al.* (2018)**. For at least 15 days, mites were transferred using a camel hairbrush to fresh guava leaf trees and placed on moistened cotton. The cultures were maintained at a temperature of 25 ± 5 ºC and a relative humidity of 65 ± 5% under a light-dark cycle of L12:D12. Adult females of uniform age were collected from these cultures for the experiments.

2.2. Tested compounds

Three compound formulations according to their active ingredients were**:**

- Bifenazate (Abidex SC 48%): isopropyl 3-(4-methoxybiphenyl-3-yl) carbazate: Acaricide with long residual action. Used for control of phytophagous mites (both eggs and motile stages) on citrus crops, tree fruits, nuts, ornamentals, and cotton. Use rates are 40cm³/100 liters of water.

- Clofentezine (Sadio SC 20%) 3, 6-bis (2-chlorophenyl)-1, 2, 4, 5-tetrazine: Acaricide with long residual activity. Inhabit embryo development, used to control eggs and young motile stages of *Tetranychus* species on pome fruit, stone fruit, and citrus fruit, has no effect on predatory mites or beneficial insect species. Use rates are 50cm³ /100 liters of water (**Mann, 2003–2004**).

- Spirodiclofen (Marvil SC 24%) 3-(2,4-dichlorophenyl)-2-oxo-1- oxaspiro [4.5] dec-3-en-4yl 2,2-dimethyl butanoate: Acaricide used to control mite pests such as *Tetranychus* species at 50 ml/100 liter of water, for uses in citrus, pome fruit, stone fruit, grapes and nuts.

2.3. Experimental design

Discs were cut from guava leaves using a cork borer and placed on a moist sponge covered with moist tissue paper in 9 cm Ø Petri dishes. All experiments were performed in the laboratory during spring.

2.4. Irradiation methods

The acaricides were exposed to UV-A light generated by standard UV lamp type EMITA VP- 60 (made in Poland) with 180 watts, mercury lamp, 220 V, 50 Hz equipped with a monochromatic filter (ʎ=320nm), installed at National Center for Radiation Research and Technology (NCRRT), Egypt. In this study, the sample was placed at a constant distance of (5 cm) from the lamp for four intervals at a dose rate of 23.7 KJ/m²/hr generated by the device**.**

2.5. Toxicity of tested irradiated acaricides on adults of *T. urticae*

To evaluate the toxicity of the irradiated acaricides on adult *T. urticae*, we prepared four 2.5 cm diameter discs from guava leaves for each treatment, while control discs received non-irradiated acaricide. Ten female mites were placed on each disc using a camel hairbrush. Each treatment involved three sprays from a consistent distance, applying the recommended agricultural doses for each acaricide. Mortality counts for the female mites were recorded at 24, 48, and 72 hours.

2.6. Ovicidal effect of irradiated acaricides on eggs of *T. urticae*

To assess the hatching effects of UV-A irradiation, we placed ten adult female two-spotted spider mites on Guava leaf discs. After removing the adult females, the next day, we counted the eggs deposited on the leaves. Each tested acaricide was sprayed on Guava leaves containing 20-25 eggs per leaf. On the fourth day after oviposition, we evaluated the viability of both the control and treated eggs. Following the methodology proposed by **Yanar *et al.* (2011)**, the eggs that did not hatch after this time were considered non-viable.

2.7. Statistical analyses

Data collected from the experiments were analyzed with a one-way ANOVA analysis of variance using SPSS. To examine the relationship between the irradiation period and mite mortality, we employed Pearson's correlation coefficient. Our experiments specifically investigated how irradiation acaricides affected the hatchability of mite eggs; we considered the numbers of emerging living juveniles as viable eggs.

3. Results and Discussion

The impact of solar radiation has emerged as a significant concern, particularly due to the depletion of stratospheric ozone and the consequent increase in UV-A and UV-B radiation reaching the Earth's surface. Our study aimed to investigate whether the random application of agrochemicals in specific areas during sunny periods could mitigate the toxic effects of acaricides on mites.

The study was conducted in April 2024; the results revealed notable changes in the color and texture of the chemical substances tested. Figures (1-5) showed the changes in color and texture of tested acaricides after exposure to different periods of artificial irradiation compared to the un-irradiated ones, a dense dark layer formed on the surface of the liquid indicating that the chemical bonds were breaking down and the compounds were being altered under UV-A exposure.

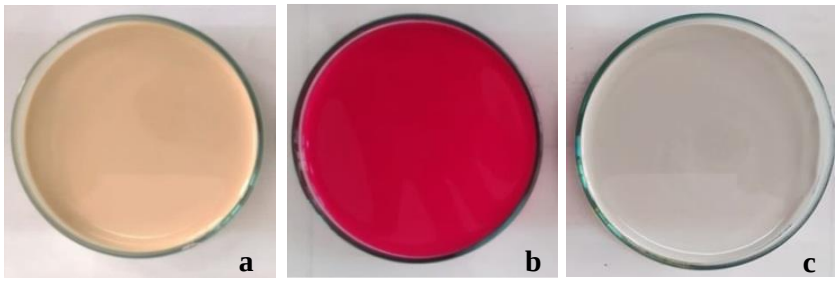

Fig. (1). Tested non-irradiated acaricides, photos are **a**) Bifenazate, **b**) Clofentezine, and **c**) Spirodiclofen

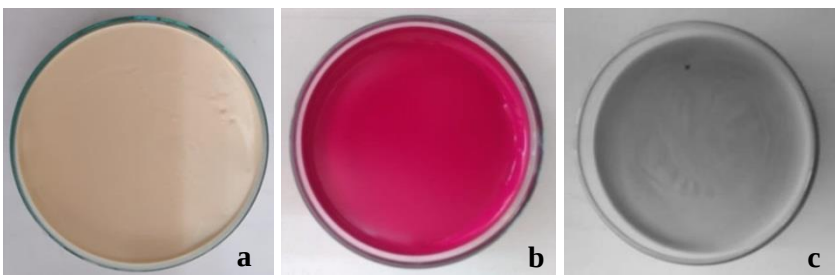

Fig. (2). Tested acaricides after 30 min. exposure with UV-A irradiation, the formation of a hard solid layer on top of the dish. Photos are **a**) Bifenazate, **b**) Clofentezine, and **c**) Spirodiclofen

Fig. (3). Tested acaricides after 60 min. exposure to UV-A irradiation, formation of a hard solid layer on top of the dish and darkness of the liquid. Photos are **a**) Bifenazate, **b**) Clofentezine, and **c**) Spirodiclofen

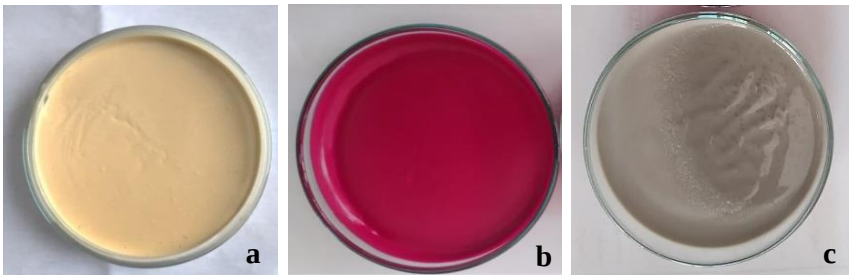

Fig. (4). Tested acaricides after 90 min. exposure to UV-A irradiation, formation of a hard solid layer on top of the dish and darkness of the liquid. Photos are **a**) Bifenazate, **b**) Clofentezine, and **c**) Spirodiclofen

Fig. (5). Tested acaricides after 120 min. exposure to UV-A irradiation and breaking of hard solid layer on top of the liquid. Photos are **a**) Bifenazate, **b**), Clofentezine, and **c**) Spirodiclofen

Table (1) shows the percentage lethality (%) of *T. urticae* following the application of UV-A irradiated Bifenazate, Clofentezine, and Spirodiclofen across four different irradiation exposure durations: 30, 60, 90, and 120 minutes. Data of (0) min. Indicates mortality in control, which were not exposed to UV-A irradiation. All three unirradiated acaricides demonstrated a maximum lethality rate of 100% against *T. urticae* after 24 hours. The mortality rate in *T. urticae* after spraying with UV-A irradiated Bifenazate dropped to 77.5 %, which was statistically significant compared to the control group ($P < 0.05$). Moreover, Clofentezine and Spirodiclofen showed notable decreases in lethality, with values of 40% and 27%, respectively, after the initial 30 minutes of exposure. For the second exposure group, which lasted 60 minutes, Bifenazate displayed a further decreased lethality rate of 72.5% after 24 hours. During this same interval, significant reductions ($P < 0.05$) were observed in Clofentezine and Spirodiclofen, resulting in 35% and 25% mortality rates. Following the 90-minute exposure, the mortality rates slightly declined compared to the 60-minute group; however, these changes did not reach statistical significance after 24 hours for any of the tested acaricides. Finally, in a 120-minute exposure period, Bifenazate resulted in a lethality rate of 62.5% in female *T. urticae* after 24 hours. However, Clofentezine and Spirodiclofen exhibited significantly lower rates ($P < 0.05$) of 22.5% and 12.5%, respectively. Following a 48-hour observation period, lethality rates across all acaricides were markedly reduced. Observation of the effect of UV-A irradiation on tested acaricides after 48 and 72 showed more lethality of *T. urticae* but did not achieve a complete mortality of 100%.

Table (1). Mean mortality percentage of female *T. urticae* exposed to UV-A irradiated acaricides under four exposure times and three days observations

24 hr.

Acaricide	Time of exposure	Mean mortality percentages of *T. urticae* %	*P value*	LSD
	(0)min	100.0±0 a	.0028 **	1.662
	30min	77.5±5 b		
Bifenazate	60min	72.5±15 b		
	90min	70±14.14 b		
	120min	62.5±12.58 b		
	(0)min	100.0±0 a	.0000***	1.481
	30min	40±14.14 b		
Clofentezine	60min	35±12.9 bc		
	90min	27.5±9.57 bc		
	120min	22.5±5 c		
	(0)min	100.0±0 a	.0000***	1.305
	30min	27.5±9.57 b		
Spirodiclofen	60min	25±12.9 bc		
	90min	22.5±9.57 bc		
	120min	12.5±5 c		

Data are the mean of four replicate discs ± standard deviation. (0) min. indicates mortality for the controls, which were not exposed to UV-A radiation. Means in a column with the same letter doesn't differ significantly at $P < 0.05$, LSD test.

48 hr.

Acaricide	Time of exposure	Mean mortality percentages of *T. urticae* %	*P value*	LSD
	(0)min.	100.0±0 a	.0089 **	1.815
	30min.	85±5.77 ab		
Bifenazate	60min.	75±12.9 bc		
	90min.	72.5±15 bc		
	120min.	65±17.32 c		
	(0)min.	100.0±0 a	.0000***	0.912
	30min.	52.5±9.57 b		
Clofentezine	60min.	45±5.77 b		
	90min.	35±5.77 c		
	120min.	22.5±5 d		
	(0)min.	100.0±0 a	.0000***	1.246
	30min.	42.5±9.57 b		
Spirodiclofen	60min.	37.5±9.57 bb		
	90min.	32.5±9.57 b		
	120min.	20.00 ±8.16 c		

72hr.

Acaricide	Time of exposure	Mean mortality percentages of *T. urticae* %	*P value*	LSD
	(0)min.	100.0±0 a	.0016**	1.362
	30min.	87.5±5 ab		
Bifenazate	60min.	75±9.57 bc		
	90min.	72.5±9.57 c		
	120min.	70±14.14 c		
	(0)min.	100.0±0 a	.0000***	1.866
	30min.	52.5±12.58 b		
Clofentezine	60min.	47±17.07 b		
	90min.	42±15 bc		
	120min.	27.5±9.57 c		
	(0)min.	100.0±0 a	.0000***	1.246
	30min.	42.5±9.57 b		
Spirodiclofen	60min.	37.5±9.57 b		
	90min.	32±9.57 b		
	120min.	20±8.16 c		

We conclude that, the degree of photodegradation in the field is determined by several factors based on the chemical composition of the pesticide formulation and their application methods on crops. Our findings indicate that applying insecticides during sunny periods does not necessarily lead to the highest levels of pest mortality. The deadly effect of tested acaricides varied depending on their exposure period to solar radiation in the field. This aligns with the research study conducted by **Papagiannaki *et al.*, (2020)**, which examined the biological effects of UV irradiated glyphosate on aquatic organisms in groundwater. Their results indicated that irradiated glyphosate was less harmful to the aquatic test organisms than controls, suggesting that UV irradiation may lead to less toxic products, ultimately reducing overall toxicity to these organisms.

The relationship between the duration of irradiation and the average mortality of adult *T. urticae* across three different acaricides is illustrated in (Fig. 6). A notable positive correlation was found for the acaricide Bifenazate, with a Pearson's correlation coefficient of $R = 0.84$ ($P < 0.05$). Mortality of mites decreased significantly with more prolonged exposure periods to irradiation. These results are consistent with the findings of **Shayeghi *et al.* (2012)**, who reported that longer irradiation durations reduced the toxicity of malathion; their ANOVA analysis revealed significant time-related variations. Furthermore, **Sulzberger *et al.* (2019)** also contributed valuable insights on this subject. Photodegradation from solar UV radiation can diminish the effectiveness of pesticides.

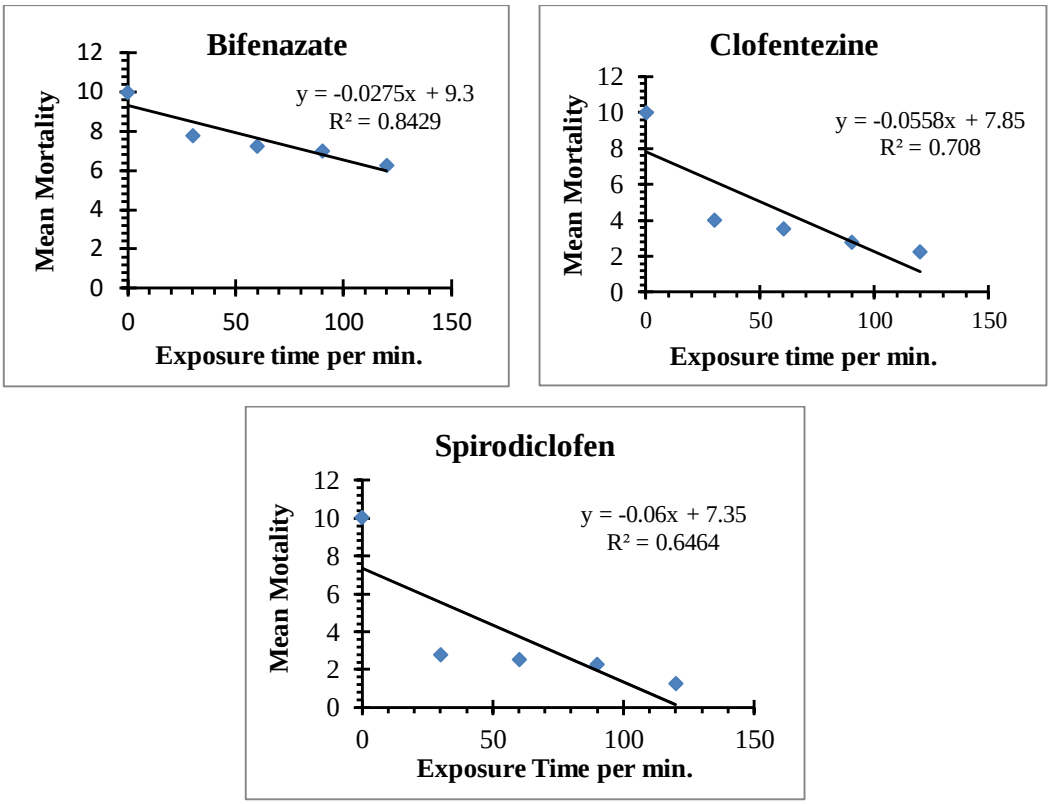

Fig. (6). Correlation between exposure time to irradiation and mean mortality of *T. urticae* [Pearson's correlation coefficient]

Several studies explore how pesticides interact with UV light. Initially, UV radiation can cause direct photochemical alterations in pesticides by absorbing photons. This leads to various chemical reactions, including bond cleavage and oxidation-reduction processes. Additionally, UV light interactions with water and oxygen can produce photoproducts such as hydrogen peroxide (H_2O_2), hydroxyl radical (OH), and superoxide anion (O_2^{-}), which can further degrade insecticides. Furthermore, factors such as the functional groups in the water matrix of the original compound and the specifics of UV exposure (like UV dose and wavelength) play crucial roles in the degradation process (**Wols & Hofman-Caris, 2012 and Katagi, 2018).**

The data shown in Table (2) illustrates how irradiated acaricides impacted the eggs of *T. urticae* depending on four different exposure times. Notably, as the irradiation exposure time increased, the viability of eggs increased. In the control treatments not subjected to UV-A radiation, egg viability showed a significant decrease ranging from 5-19%. Conversely, the treated eggs with an irradiation time of 30 min caused 38-53 % of viable eggs in tested acaricides. After treatment with a 120-minute exposure, the viability of the eggs increased to between 50-69%. This finding compatible with the research study conducted by **Gala *et al.* (2021)**, who examined the impact of irradiation on egg mortality. Their study reported nearly 100% viability for each of exposure duration, even at the lowest energy density.

Table (2). Effect of UV-A irradiated acaricides on viability of eggs of *T. urticae* under four exposure times

Exposure time	Acaricide	No.of eggs	Viable eggs	Viability (%)
	Bifenazate	90	5	5.55
(0) min.	Clofentezine	88	27	13.63
	Spirodiclofen	91	34	19.78
	Bifenazate	86	33	38.37
30 min.	Clofentezine	95	50	52.63
	Spirodiclofen	90	48	53.33
	Bifenazate	91	37	40.66
60 min.	Clofentezine	94	50	53.19
	Spirodiclofen	95	61	64.21
	Bifenazate	97	39	40.21
90 min.	Clofentezine	96	56	58.33
	Spirodiclofen	92	60	65.22
	Bifenazate	92	46	50.00
120 min.	Clofentezine	92	55	59.78
	Spirodiclofen	91	63	69.23

4. Conclusions

The indiscriminate application of chemical acaricides during sunny periods by some farmers has prompted an investigation into the effects of UV-A irradiation that is not absorbed by the atmosphere and ozone layer on the toxicity of these compounds on the herbivorous mite *Tetranychus urticae*. Notably, there is a lack of research addressing the influence of UV irradiation on the active ingredients of acaricides and their toxicity to pest organisms. The effectiveness of acaricides in killing adult mites decreases with more prolonged exposure to UV-A irradiation [Pearson's correlation coefficient ($R= 0.84$, $P < 0.05$)]. Furthermore, the viability of mite eggs treated with UV light increased, with viable eggs ranging from 38-53% after just 30 minutes of irradiation. Two hours of irradiated acaricides caused highly surviving mite individuals and extremely viable eggs compared to the control. These results suggest that the random application of acaricides during sunny hours is not an effective strategy for pest control.

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