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Article

Study of the population fluctuations and feeding effects of *Tetranychus urticae* (Acari: Tetranychidae) on three cultivars of *Capsicum annuum* (Solanaceae)

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ABSTRACT

The objectives of this study were to investigate the population fluctuation of *Tetranychus urticae* Koch (Acari: Tetranychidae) on three *Capsicum annuum* L. (Solanaceae) cultivars (red delta star, yellow delta star and chili omega 62). Experimental trials were carried out under high plastic-net house conditions during two seasons, 2021–2022 and 2022–2023. The feeding effects of *T. urticae* on some biochemical compounds of *C. annuum* were estimated. Densities of egg and mobile stages on the three test cultivars increased gradually from initial count in March 15th and reached peak at last week of November of two seasons, except densities of eggs on red delta reached peak at last week of October. In both growing seasons, the highest population densities of *T. urticae* were observed on red delta star, while the lowest densities were on chili omega 62. Correlation coefficient of *T. urticae* densities levels with total carotenoids, carbohydrates and proteins of tested *C. annuum* cultivars showed positive relationship and a reverse one with total lipids, chlorophyll, phenolic, flavonoids and alkaloids compounds.

KEYWORDS: Herbivore-plant interactions, pepper, pest population dynamics, pest feeding behavior, phytochemical analysis.

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INTRODUCTION

Pepper, *Capsicum annuum* L. (Solanaceae) is one of the most popular economically and nutritionally beneficial vegetable crops grown in greenhouses or net houses around the world (El Arnaouty *et al.* 2018; Yankova *et al.* 2021). The Egyptian pepper is ranked as the top 10 products and exported globally (Howard *et al.* 2000; Navarro *et al*. 2006; El-Laithy *et al.* 2013). According to FAO (2021), the total Egyptian pepper cultivated areas reached 381,320 hectares with a total production of more than 600,000 tons.

Tetranychus urticae Koch (Acari: Tetranychidae) known as the two-spotted spider mite (TSSM), is one of the most damaging pest species to pepper, the majority of Solanaceae crops, and other plant families worldwide (Adango *et al.* 2006; Perdikis *et al.* 2008; Dutta *et al*. 2012; El-Saiedy *et al*. 2015; Abou-Elella *et al.* 2021; Barghout *et al.* 2022). The severe attack of the pest mites causes reduction

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of the quantity and quality of production; it feeds on mesophilic tissues of leaves and results in mechanical damage which leads to increases of necrotic cells. Moreover, the feeding activity reduces concentrations of nitrogen, protein and phosphorous, as well as disrupting cell physiology and reducing photosynthesis (Ohtsuka and Osakabe 2009). Furthermore, it possesses multiple biological features that facilitate the pest mite resistance to various pesticides (Grbic *et al.* 2011). The considerable incidence of *T. urticae* has been reported under protected cultivation due to suitable environmental conditions (Haque *et al.* 2011; Tehri *et al.* 2014).

Considering the importance of *T. urticae* under protected houses, its population dynamics should be investigated. Population dynamic is little seen in pest control programs, although its evaluation can provide valuable insight into invasiveness or preservation status in changing habitats as it reflects the different effects among populations' behavioral interactions with environment (Raje *et al.* 2016; Mech *et al.* 2019). Additionally, based on the herbivore-plant interactions, many tactics have been suggested to get plant-triggered defenses enhanced (Bazazzadeh *et al.* 2020; Santamaria *et al.* 2020; Zidan *et al.* 2022). Numerous investigations on the plant resistance to *T. urticae* infestations have been conducted. The assessment of the resistant plants, which is a new trend in the pest management practices, has been considered as an alternative tool in the biological pest control (Ibrahim *et al.* 2008; El-Saiedy *et al.* 2011; Afifi *et al.* 2013).

In view of this, the present work was conducted to monitor the population buildup of *T. urticae* on three pepper cultivars (red delta star, yellow delta star and chili omega 62) during two successive seasons in order to understand suitable time and control management tactics of the pest. In addition, the feeding impact of *T. urticae* on some plant biochemical compounds was estimated.

MATERIAL AND METHODS

Experimental location

The experiment was conducted at at Om Saber, Badr Centre, El Beheira Governorate (30° 28' 59.4" N, 30° 46' 52.9" E) during two successive seasons 2021–2022 and 2022–2023 under high plastic-net house.

Plantation

Capsicum annuum seeds of the three cultivars, red delta star (RDS), yellow delta star (YDS) and chili omega 62(CO62) were propagated on January 10th in the nursery then seedlings were transplanted in the net house on February 20th. There were three equal plots and each one represented a cultivar. The plots were separated completely by plastic sheets. Each cultivar consisted of three rows with 12 m length × 75 cm width, and each row represented one replicate and contained 40 plants within a completely randomized block design. All pepper cultivars received uniform agricultural practices without use of any pesticides. The mean temperature and relative humidity of the high plastic-net house were recorded monthly and averaged 28 ± 3 °C and 70 ± 5 % RH.

Sampling

The pest mite infestation occurred naturally. Samples were taken at regular weekly intervals, starting from March 15th and continuing till February 15th through two successive seasons. Thirty leaves (10/replicate) were randomly gathered from top, middle and bottom of the plant, and then kept inside an ice box in polyethylene bags for a stereomicroscope check. Mobile stages (including active and quiescent larval, nymphal, and adult stages), besides eggs of *T. urticae* were counted and recorded.

Biochemical analysis of the three C. annuum *cultivars leaves*

Chemical analysis of leaf samples was carried out during the peak season of infestation. Leaves of the three pepper cultivars were collected and air dried, then moved to Cairo University's Laboratory of Agriculture Faculty Research Park to estimate the biochemical alterations. The method of Lee and Takabashi (1966) was followed to calculate the caloric value of total protein. The plant leaves' total carbohydrates were removed and prepared for estimation (Crompton and Birt 1967). Total lipids were also calculated (Bligh and Dyer 1959). Chlorophyll and total carotenoids were measured colorimetrically using Holden's method (1965). By using a modified version of Singleton and Rossi's Folin-Ciocateu technic (1965), the amount of total phenol was calculated. Titrimetric analysis was used to determine the alkaloids using Sabri *et al.* (1973). Folin-Ciocalteu technic was used to determine the total flavonoid concentration based on Hung and Morita (2008).

Data analysis

The IBM SPSS statistics computer program, version 20, was used to conduct all statistical analyses. By utilizing Duncan's multiple range tests with a 95% confidence level, one-way analysis of variance was used to compare the means of three replicates of each cultivar (Duncan 1955). The simple correlation coefficient between the pest mite populations and biochemical changes of each pepper cultivar was calculated by Pearson simple correlation coefficient calculator.

RESULTS

Population fluctuations of T. urticae *egg and mobile stages on the three* C. annuum *cultivars*

Data in Figures 1–4 concluded the egg and mobile stages fluctuations on RDS, YDS and CO62 cultivars through two consecutive seasons; the initial population built up from March 15th of two seasons, respectively and gradually fluctuated at determined times. As shown in Figures 1–2, the highest peaks of mobile stages densities on the three pepper cultivars during two consecutive seasons were recorded on November 30th and the lowest peaks after population built up were on January 15th. Data presented in Figure 3 show eggs stage densities during season 2021–2022, in which the density gradually built up to reach the highest peaks on October 30th for RDS cultivar, and November 30th for YDS and CO62 cultivars. It is worth noting that the average temperature and relative humidity were 28 ± 3 °C and 70 ± 5% RH in the months with a growing population rise and 16 ± 3 °C and 65 ± 5% RH in the months with a growing population drop.

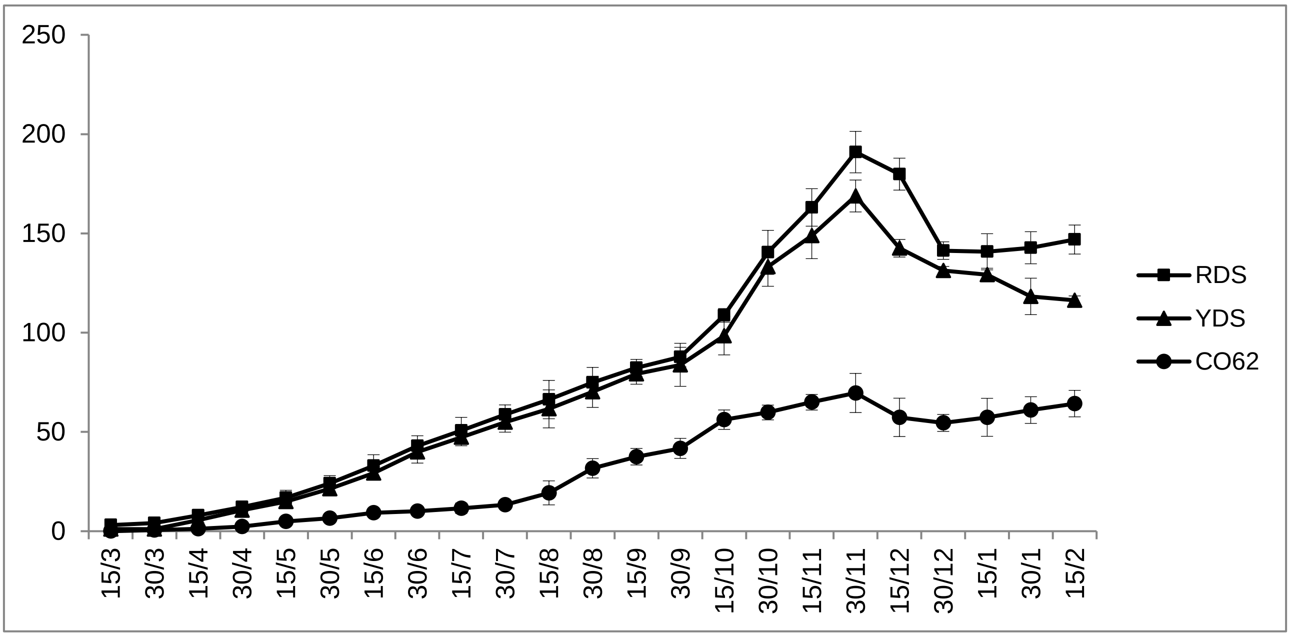

Figure 1. Population fluctuations of *T. urticae* mobile stages on the three *C. annuum* cultivars at El-Beheira governorate during seasons 2021–2022.

The same trend was observed in the second season 2022–2023 (Fig. 4). The yearly average of the egg and mobile stages was estimated and it was observed that the population densities of *T. urticae* are in positive relationship with the host plant sensitivity as confirmed statistically in Table 1; variations of both stages densities on each cultivar were significant ($P < 0.05$). RDS cultivar was the most sensitive one to *T. urticae* infestations, while CO62 cultivar was the least sensitive one during two seasons.

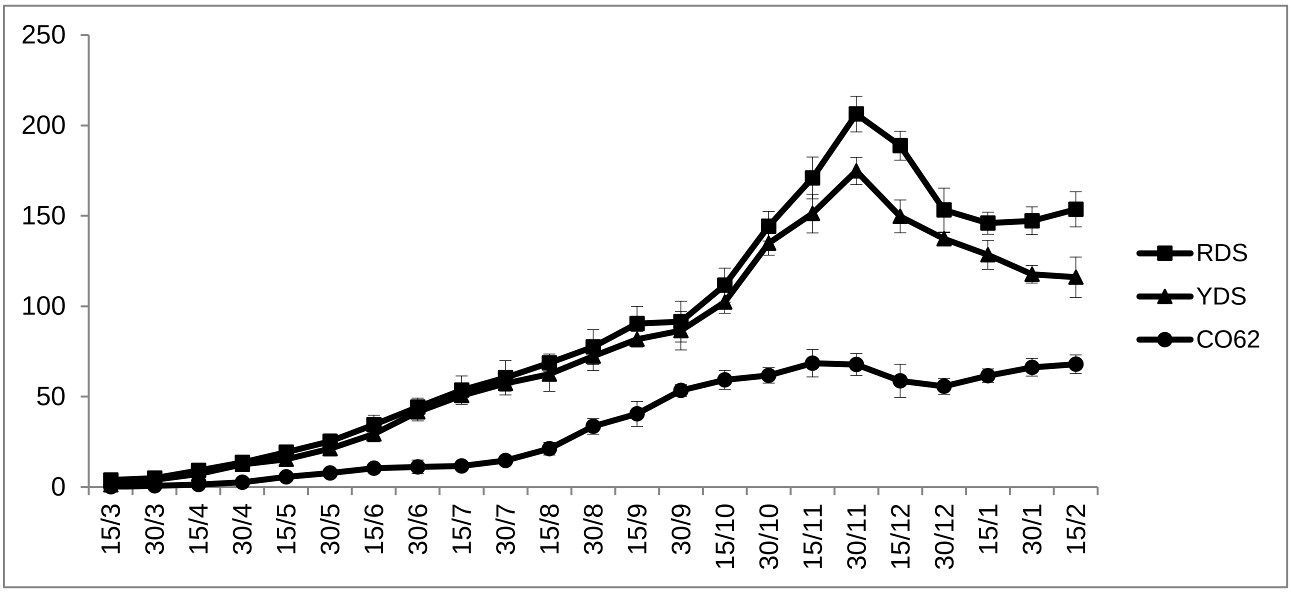

Figure 2. Population fluctuations of *T. urticae* mobile stages on the three *C. annuum* cultivars at El-Beheira governorate during seasons 2022–2023.

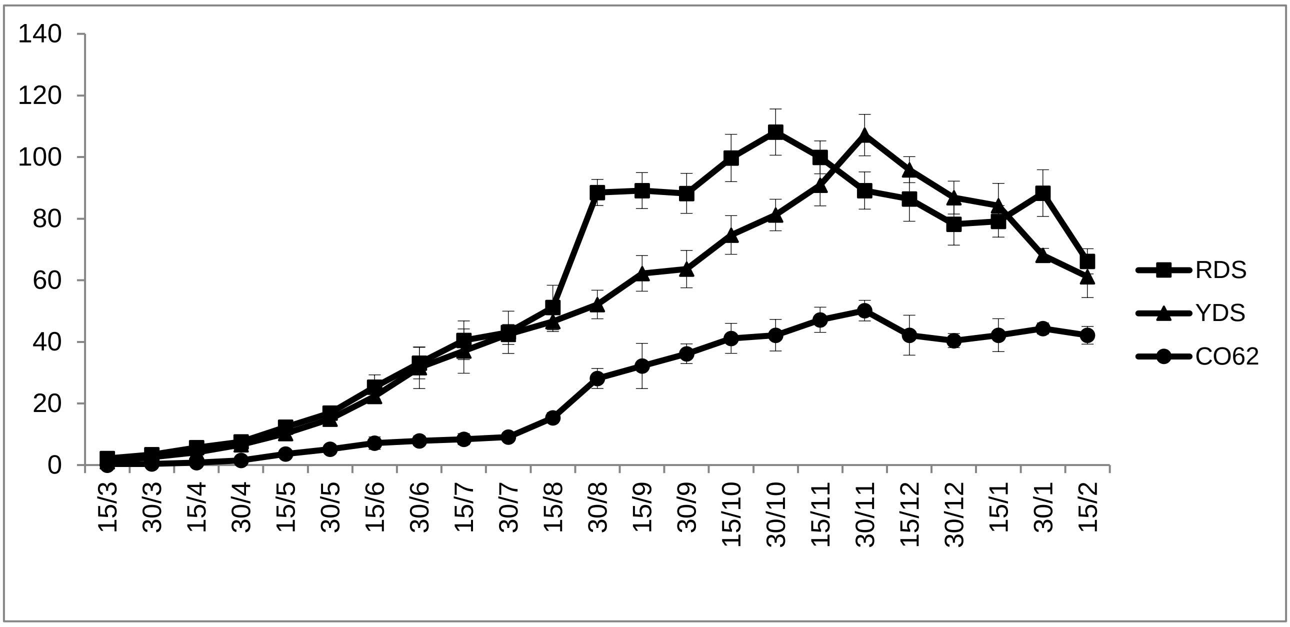

Figure 3. Population fluctuations of *T. urticae* eggs stages on the three *C. annuum* cultivars at El-Beheira governorate during seasons 2021–2022.

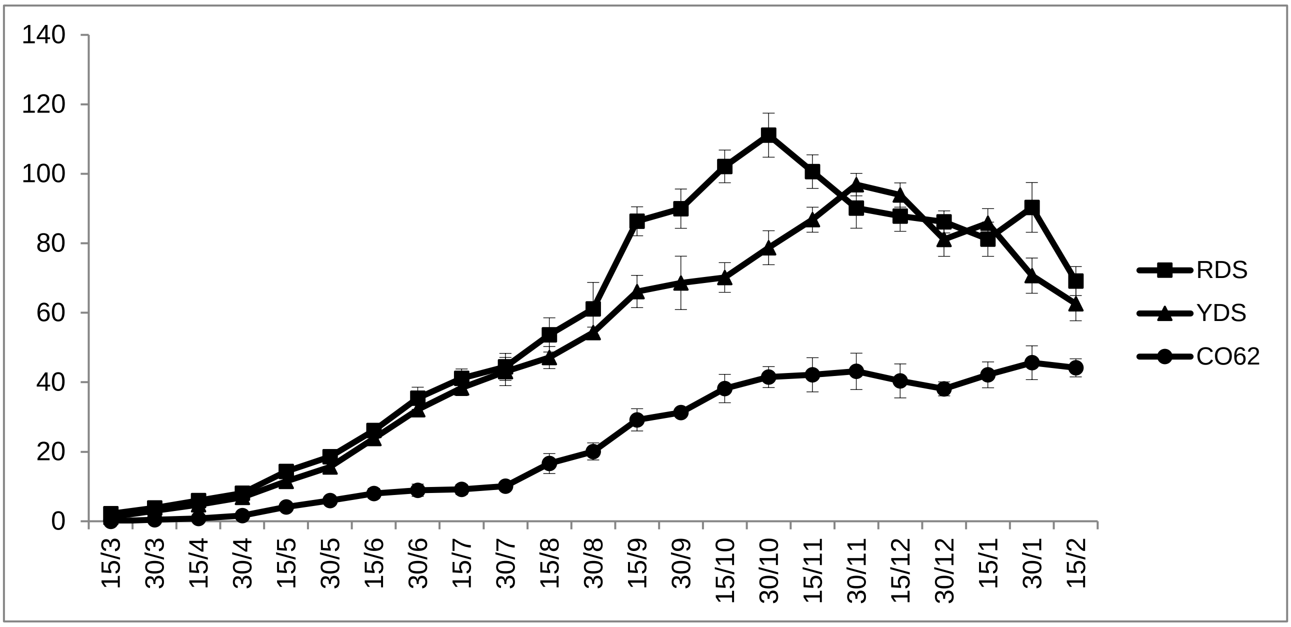

Figure 4. Population fluctuations of *T. urticae* eggs stages on the three *C. annuum* cultivars at El-Beheira governorate during seasons 2022/2023.

Table 1. Population densities of *T. urticae* egg and mobile stages (mean no. of *T. urticae*/leaf ± SE) on the three *C. annuum* cultivars during 2021–22 and 2022–23 seasons.

Cultivar	Season			
	2021–22		2022–23	
	Mobile stages	Eggs	Mobile stages	Eggs
RDS	83.45 ± 12.81[a]	56.60 ± 7.68[a]	87.79 ± 13.46[a]	56.96 ± 7.64[a]
YDS	74.24 ± 11.21[a]	49.89 ± 6.99[a]	76.35 ± 11.39[a]	49.71 ± 6.67[a]
CO62	31.95 ± 5.41[b]	23.81 ±3.92[b]	34.02 ± 5.64[b]	22.69 ± 3.66[b]
F value	7.09	7.35	7.01	8.42
***P* value**	0.002	0.001	0.002	0.001

Different letters within the same column refer to a significant difference among cultivars. $P \leq 0.05$ (significant), $P \leq 0.01$ (highly significant).

Feeding impact of T. urticae *on some biochemical compounds of the three* C. annuum *cultivars*

The estimated biochemical components of each cultivar were considerably altered by the activity of *T. urticae* stages, according to data shown in Table 2, and the degree of influence varied depending on how sensitive the cultivar was to the pest attack ($P < 0.05$); a positive relationship was observed between mobile or egg stages infestation and levels of total carotenoids, total carbohydrates and total proteins in the three tested pepper cultivars (r = 0.990 ; 0.999; 0.966 with mobile stages and 0.992; 0.999; 0.969 with egg stages), respectively; while, a negative relationship was observed with total lipids, total chlorophyll, total phenolic, total flavonoids and alkaloids compounds (r = −0.889; −0.928; −0.921; −0.952; −0.962 with mobile stages and −0.895; −0.923; −0.925; −0.956; −0.965 with egg stages), respectively.

DISCUSSION

In this study, the population fluctuations of egg and mobile stages of *T. urticae* on RDS, YDS and CO62 cultivars varied according to tested time intervals and the host plant sensitivity. The densities increased gradually from March15th and reached the highest peak on November 15th and the reverse was achieved at other tested times.

Table 2. Feeding impact of *T. urticae* on some biochemical compounds of the three *C. annuum* cultivars. CCVm = Correlation of coefficient values with mobile stages; CCVe = Correlation of coefficient values with egg stage.

Cultivar	RDS	YDS	CO62	F value	*P* value	CCVm	CCVe
Mean no. of *T. urticae* mobile stages	85.62	75.29	32.99				
Mean no. of *T. urticae* egg stage	56.78	49.80	23.25				
Total carotenoids (mg/g)	10.93 ± 0.04[a]	9.92 ± 0.02[b]	7.84 ± 0.05[c]	1822.09	0.0001	0.990	0.992
Total carbohydrates (mg/g)	17.44 ± 0.30[a]	15.85 ± 0.14[b]	10.83 ± 0.36[c]	151.98	0.0001	0.999	0.999
Total protein (%)	23.10 ± 0.51[a]	21.22 ± 0.07[b]	18.76 ± 0.17[c]	48.268	0.0001	0.966	0.969
Total lipids (%)	9.16 ± 0.020[c]	10.55 ± 0.12[b]	11.40 ± 0.11[a]	144.617	0.0001	−0.889	−0.895
Total chlorophyll (mg/g)	111.67 ± 0.69[b]	108.93 ± 0.20[b]	122.21 ± 1.56[a]	49.697	0.0002	−0.928	−0.923
Total phenol (mg/g)	0.49 ± 0.009[c]	0.54 ± 0.006[b]	0.58 ± 0.009[a]	27.706	0.001	−0.921	−0.925
Total flavonoids (mg/g)	28.54 ± 0.12[c]	29.44 ± 0.17[b]	30.43 ± 0.15[a]	40.23	0.0003	−0.952	−0.956
Alkaloids (%)	4.8 ± 0.06[c]	5.52 ± 0.13[b]	6.41 ± 0.049[a]	79.645	0.0001	−0.962	−0.965

Different letters within the same row show a significant difference among biochemical level. $P \leq 0.05$ (significant), $P \leq 0.01$ (highly significant).

Hypothetically, to find an explanation, environmental factors, especially temperature and relative humidity play an important role in *T. urticae* biology. In our experimental site, under high plastic-net house conditions 28 ± 3 °C and 70 ± 5% RH, *T. urticae* population developed well and this is compatible with Riahi *et al.* (2013) and Damos *et al.* (2023). Moreover, several population densities of the pest mite were studied on many agricultural crops and all were in positive relationship to temperature (Praslicka and Huszar 2004; Aiad *et al.* 2014; Abou-Awad *et al.* 2017; Kamel *et al.* 2019; Ghongade and Sangha 2020). Thus, the necessity for regularly assessing pest populations and community structure is important to better stand on an IPM tactic or to apply biological control method to make pests below the economic threshold (ETH) and/or the economic injury level (EIL) (Zidan *et al.* 2022).

Additionally, it is concluded that RDS cultivar was the most sensitive to *T. urticae*, in contrast to CO62 cultivar. Variation in host sensitivity may be referred to the genetic variations or differences in the nutrition content and induced chemical defenses, thereby affecting *T. urticae* survival, growth, and reproduction (Kumral *et al.* 2017; Velázquez-Ventura *et al.* 2018; Fahim *et al.* 2020; Santamaria *et al.* 2020; Divekar *et al.* 2022).

Chilies could be characterized by genetic variations resistant to pests more than the other two cultivars. As example, Piquin chili has a diversity of defensive secondary metabolites and it was the most resistant one to *Tetranychus merganser* Boudreaux (Hayano-Kanashiro *et al.* 2016; Chacón-Hernández *et al.* 2020). Also, out of eight pepper cultivars, Roxy red was the most vulnerable one to *T. urticae* infestation (El-Saiedy *et al.* 2013). Within Solanaceae, *C. annuum* was poorly accepted by *T. urticae* (Van den Boom *et al.* 2003), and the less vulnerable than *Solanum melongena* L. and *S. lycopersicum* plants (Awad *et al.* 2018).

In addition, the histological changes in the three tested pepper cultivars in response to *T. urticae* feeding indicated negative effects on the plant's fitness and survival. The empty collapsed cells in mesophilic tissues which were presented in histological transverse section (See Supplementary file) resulted the feeding activity, and it was notably seen in the most susceptible RDS cultivars. Similarly, the feeding impact of *T. urticae* was imparted on the mesophilic tissue of arabidopsis, *Arabis thaliana*

L. (Brassicaceae) and cucumber, *Cucumis sativus* L. (Cucurbitaceae), leaves (Park and Lee 2002; Bensoussan *et al.* 2016). Jeppson *et al.* (1975) illustrated that *Tetranychus* spp. use long stylets of 150 µm for flush and lacerate feeding on mesophilic cells, mainly parenchyma, of which they can empty up to 18–22 cells/minute.

In the present study, the resulted biochemical changes recorded positive relations with total carotenoides, total carbohydrates and total protein, in addition to negative relations with total lipids, total chlorophyll, total phenolic, total flavonoids and alkaloids compounds. These together considered the plant responses and defensive mechanisms against *T. urticae* infestation. The pest mite can tamper with primary metabolites like proteins, lowering their levels. This also leads to decreasing chlorophyll levels because of photosynthetic cells damaged due to pest feeding activity (Zhou *et al*. 2015), as well as, some plant nutrients transferred into the pest's digestive system and turned into ATP compounds that increase the pest's life table parameters (Abou-Elella *et al*. 2021).

Interestingly, the levels of induced secondary metabolites as phenols, flavonoids and alkaloids by the action of *T. urticae* feeding confirmed the cultivars sensitivity levels which contribute to plant's defense by deterring or repelling herbivore attacks (Smith 2005; Divekar *et al.* 2022).

The current results revealed that CO62 cultivar recorded the highest levels of the induced secondary metabolites resulting in its resistance to the pest mite, and vice versa in other two cultivars. El-Saiedy *et al.* (2013) and Abd El- Rahman *et al.* (2016) observations on sweet pepper cultivars and tomato leaves biochemical analysis are similar to our results.

In contrary, insignificant correlation has been recorded between *T. urticae* eggs with the same estimated components on melon, *Cucumis melo* L. (Cucurbitaceae) (Aiad *et al.* 2014), total protein of eggplant, *S. melongena* leaves (Abd-Elsamed *et al.* 2018), carotenoids, and total free amino acids (Helmi and Rashwan 2015).

CONCLUSION

The present study revealed that the population fluctuations of all *T. urticae* stages at the tested time intervals had the same trend in the two successive seasons. Chili omega 62 cultivar was the most resistant one to *T. urticae* infestation. The extensive disruption of mesophyll tissues and the highest levels of biochemical parameters, carotenoids, carbohydrates and proteins were observed in RDS cultivar and the reverse on CO62. The secondary metabolites as phenolics, flavonoids and alkaloids which were induced after the pest mite attack were highly in CO62 cultivar. The resistant cultivar can contribute to the pest management and it could be used as protected fence around other two sensitive cultivars to decrease the incidence or repel the pest mite infestation.

Figuring out the population's build up of the pest mite and the host plant characteristics are important factors to be considered when exploring integrated pest management (IPM) for *T. urticae*; anticipating the initial infestation time can help in early management. Furthermore, the host plant interaction and deep understanding to the biochemical alteration may contribute to pest control as well.

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مطالعهٔ نوسانات جمعیت و اثرهای تغذیه‌ای گیاه *Tetranychus urticae* (Acari: Tetranychidae) روی سه رقم *Capsicum annuum* (Solanaceae)

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چکیده

هدف از این مطالعه بررسی نوسانات جمعیت *Tetranychus urticae* Koch (Acari: Tetranychidae) بر روی سه رقم *Capsicum annuum* L. (Solanaceae) (ستاره دلتای قرمز، ستاره دلتای زرد و فلفل قرمز امگا ٦٢) بود. آزمایش‌ها در شرایط گلخانه بلند با توری پلاستیکی در دو فصل، ٢٠٢١–٢٠٢٢ و ٢٠٢٢–٢٠٢٣ انجام شده‌اند. اثرهای تغذیه‌ای *T. urticae* روی برخی از ترکیبات بیوشیمیایی *C. annuum* برآورد شده است. تراکم تخم و مراحل متحرک در سه رقم مورد آزمایش به تدریج از شمارش اولیه در ١٥ مارس افزایش یافت و در هفته آخر نوامبر دو فصل به اوج خود رسید، به جز تراکم تخم‌ها روی دلتای قرمز در هفته آخر اکتبر به اوج خود رسید. در هر دو فصل رشد، بیشترین تراکم جمعیت *T. urticae* بر روی ستاره دلتای قرمز مشاهده شد، در حالی‌که کمترین تراکم در فلفل قرمز امگا ٦٢ مشاهده شد. ارقام سالانه با ترکیبات کل لیپید، کلروفیل، فنل، فلاونوئید و آلکالوئید رابطه مثبت و معکوس نشان دادند.

واژگان کلیدی: برهم‌کنش گیاهخوار و گیاه، فلفل، پویایی جمعیت آفات، رفتار تغذیه‌ای آفت، آنالیز فیتوشیمیایی.

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