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Article

Lethal and sublethal effects of cyflumetofen and clofentezine on life table parameters of *Tetranychus urticae* (Acari: Tetranychidae)

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ABSTRACT
Effects of cyflumetofen and clofentezine were evaluated on *Tetranychus urticae* Koch, under laboratory conditions. LC_{50} values of cyflumetofen and clofentezine against eggs were 0.8 µg a.i./mL and 0.42 µg a.i./mL, respectively. Sublethal effects of these compounds on the life table parameters of *T. urticae* were evaluated by exposing mites to LC_{50} and LC_{25} concentrations. The developmental time of male and female *T. urticae* were significantly affected by both acaricides. Moreover, exposure to LC_{25} and LC_{50} concentrations of cyflumetofen or clofentezine significantly reduced net reproductive rate (R_0), finite rate of increase (λ), and intrinsic rate of increase (r). The LC_{25} and LC_{50} treatments showed strong effects on both survivorship, fecundity, and intrinsic rate of increase. Our findings indicated that lethal and sublethal exposure to cyflumetofen or clofentezine result in significant alteration of life-history characteristics of *T. urticae*. Our findings suggest that these acaricides are promising candidates for use in management programs of this major pest.

KEYWORDS: Acaricide, biological effect, bioassay, herbivorous mites, two-spotted spider mite.

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INTRODUCTION

The two-spotted spider mite, *Tetranychus urticae* Koch has been reported to feed on more than 1,200 plant species in a wide range of ornamental and vegetable crops, and orchard trees (Zhang 2003; Naher *et al*. 2005). Despite alternative control methods available for use in integrated pest management programs, the application of acaricides still is considered the most common used method for controlling the phytophagous mite pests (Devine *et al*. 2001). Unfortunately, the repeated application of synthetic pesticides can lead to persistence and accumulation of non-biodegradable toxic compounds in the ecosystem which subsequently lead to serious health hazards to humans and animals (Pimentel *et al*. 1992; Calmasur *et al*. 2006). In addition to assessing lethal effects of pesticides, the evaluation of their sublethal effects on life table parameters is an effective route to assess the overall impact of a pesticide on a target pest (Ozkara *et al*. 2016). Among several acaricides, clofentezine and cyflumetofen are mostly used to control spider mites in field and greenhouse crops (Marcic 2003; Hayashi *et al*. 2013).

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Cyflumetofen, a novel benzoylacetonitrile acaricide derivative, has recently been introduced as a compound that inhibits complex II in the mitochondrial electron transport chain (Takahashi *et al*. 2012). Mitochondrial complex II (succinate dehydrogenase [SDH]) catalyzes the oxidation of succinate to fumarate with the reduction of ubiquinone to ubiquinol. It was commercialized as an acaricide that is highly effective against *T. urticae* and *T. kanzawai* Kishida which are resistant to conventional acaricides. Cyfleumetofen is safer compound compared to conventional acaricides toward non-target organisms and mammals (Takahashi *et al*. 2012).

Clofentezine [3, 6-bis-(2-chlorophenyl)-1, 2, 4, 5-tetrazine] is an acaricide belonging to the tetrazine group. It is an environment friendly acaricide that is compatible with integrated pest management (IPM). Clofentezine has shown lower side effects on beneficial insects and predatory mites, and reduced toxicity to humans (Aveyard *et al*. 1986; Marcic 2003).

Two types of study methods are commonly used in toxicology to evaluate pesticide efficiency on pests. An acute toxicological study is used to estimate median lethal dose (LD_{50}) or median lethal concentration (LC_{50}); whereas chronic exposure study is used to provide a complete picture of the population-level consequences of individual responses to pesticides over longer time periods (Robertson and Worner 1990; Stark and Banks 2003). The analysis of demographic toxicology or the study of life table parameters is considered as the best method to evaluate the sublethal effects of a pesticide. These types of analyses take into account the various stages or ages of a pest upon pesticide sensitivity (Stark and Banken 1999; De França *et al*. 2017). On the other hand, exposure to a sublethal dose/concentration may result in a population increase from reproduction stimulation or a population decrease or extinction. Therefore, it is necessary to understand the sublethal effects and hazards of acaricide application on a pest population (Wang *et al*. 2016).

In order to promote sustainable management of populations of *T. urticae* in protected areas and fields, understanding of the total effects of acaricides is absolutely important and necessary. There are several studies using demographic techniques to quantify the sublethal effects of pesticides on spider mites (Marcic 2005; Martinez-Villar 2005; Pozzebon *et al*. 2011; Bernardi *et al*. 2013; Marcic and Medo 2014; Li *et al*. 2017; Havasi *et al*. 2022; Mokhtari *et al*. 2022). Here, we evaluated the lethal effects of cyflumetofen and clofentezine against the egg stage of *T. urticae*, and their sublethal effects using demographic toxicological analyses. The primary aim of this study was to assess the lethal and sublethal effects of cyflumetofen and clofentezine on population growth and life-table parameters of *T. urticae*.

MATERIAL AND METHODS

Mite rearing

Tetranychus urticae was collected from leaves of the bean, *Phaseolus vulgaris* L. at the agricultural research greenhouse of the University of Maragheh, Northwestern Iran. The *T. urticae* were reared on bean *P. vulgaris* in a growth chamber at the University of Tabriz for three months. To the best of our knowledge prior to experimentation, the mites were never exposed to clofentezine or cyflumetofen or any other acaricides. The mites were reared at 25 ± 1 ℃, 60 ± 10% RH, and a photoperiod of 16:8 h (light: dark) for at least three generations before experimental use. Bioassays were performed under the same conditions.

Bioassays

Bioassay tests similar to those used by Wang *et al*. (2016) and Musa *et al*. (2017) were used to determine the insecticidal efficacy of the acaricidal compounds against eggs of *T. urticae*. Ten same age female-male pairs of *T. urticae* from the laboratory colony were transferred onto a 2 cm-diameter bean leaf discs and allowed to oviposit. After 24 h, 15 eggs were kept on each leaf disc, and the adults and extra eggs were removed. Each leaf disc was subsequently sprayed with each acaricide

concentration and allowed to air dry for 1 hour under laboratory conditions. The leaf discs were placed on top of the moistened cotton in a plastic Petri dish (6 cm diameter) with a mesh-covered ventilation hole (1 cm diameter). Egg mortality was evaluated daily following the start of larval emergence (about 6 days after treatment) and continued for five consecutive days. Eggs were scored as dead if larva did not emerge at a time when more than 98% of control eggs had hatched (the maximum egg hatch rate in control group experiments was 98%). Each bioassay consisted of five acaricide concentrations and a control group. Control leaf discs were dipped in distilled water only. The concentrations used for the bioassay were selected based on preliminary dose setting experiments. The concentrations tested were 2.00, 1.43, 0.89, 0.59, and 0.40 µg a.i./mL for cyflumetofen and 1.50, 1.00, 0.50, 0.25, and 0.125 µg a.i./mL for clofentezine. Each test was replicated 10 times. Data were subjected to probit analysis (Finney 1971). Concentration–mortality curves were estimated by probit analysis (SAS Institute 2007).

Life-table bioassay

According to the methods of Li *et al.* (2017), Mohammadi *et al*. (2016), and Havasi *et al*. (2019), 100 pairs of young female and male individuals of *T. urticae* (< 1 day old) were placed on each leaf arena and allowed to oviposit. After 24 h, each leaf disc was subsequently sprayed with LC_{25} or LC_{50} concentrations of each acaricides (i.e., 0.45 and 0.80 µg a.i./ml for cyflumetofen, and 0.21 and 0.42 µg a.i./ml for clofentezine, respectively). The leaf discs were allowed to air dry for 1 hour under laboratory conditions. Subsequently, one egg was placed on each leaf disc. Each experimental unit consisted of a 2 cm-diameter bean leaf disc that was placed on moistened cotton pad in a plastic Petri dish (the moistened cotton was exchanged when necessary to maintain moisture levels). The control group consisted of 80 eggs that were treated with distilled water. Development and survival in control and acaricide-treated discs were checked daily. When a female nymph developed into the late stationary phase, one male of *T. urticae* was introduced into the Petri dish for mating. The daily number of eggs laid by each female was recorded. The number of deposited eggs per female, the development time of subsequent stages that reached to adulthood, adult longevity, and adult survival was recorded daily until the death of all individuals. The results provided the basic elements for both the construction and calculation of life table parameters. All experiments were done at 25 ± 1 ℃, 60 ± 10% RH, and a photoperiod of 16:8 h (light: dark).

Data analysis

Bioassay data were subjected to probit analysis (Finney 1971). Estimates of the LC_{10}, LC_{50}, and LC_{90} values and their fiducial limits (95%) were determined using the probit procedure of the SAS program (SAS institute 2007). Life history data were analyzed by the age-stage, two-sex life table theory (Chi 2017). TWOSEX-MS Chart software was used to analyze the data (Chi 2017). The age-specific parameters including: age-stage survival rate (s_{xj}, where x is age and j is the insect stage), age-specific reproduction (l_xm_x), age-specific survival rate (l_x), age-specific fecundity (m_x), and population parameters including: r (intrinsic rate of increase), λ (finite rate of increase), R_0 (net reproductive rate), GRR (gross reproductive rate), and T (mean generation time) were estimated based on age-stage and two-sex life tables as described previously (Chi and Liu 1985; Chi 1988). The bootstrap method (Chi 2017) was used to estimate mean and standard error of the population parameters, longevity, and variances of the life table parameters. Differences among the treatments were compared using the paired bootstrap test (Chi 2017). The age-specific survival rate (l_x) was calculated as:

$$l_x = \sum_{j=1}^{\infty} s_{xj}$$

Where ∞ is the number of life stages (Chi and Liu 1985; Chi and Yang 2003), x = age, j = stage, and

s_{xj} = age-stage specific survival rate, i.e., the probability that a newborn egg will survive to age x and stage j. The age-specific fecundity (m_x) was calculated as:

$$m_x = \frac{\sum_{j=1}^{\infty} s_{xj} f_{xj}}{\sum_{j=1}^{\infty} s_{xj}}$$

The intrinsic rate of increase (r) was estimated using the iterative bisection method from the Euler–Lotka equation:

$$\sum_{x=0}^{\omega} e^{-r(x+1)} l_x m_x = 1$$

with the age indexed from zero (Goodman 1982). The gross reproductive rate was calculated as:

$$GRR = \Sigma m_x$$

The net reproductive rate demonstrates the mean number of offspring that an individual can produce during its lifetime and was calculated as:

$$R_0 = \sum_{x=\alpha}^{\beta} l_x m_x$$

The mean generation time is defined as the length of time that a population requires to increase to the R_0-fold of its size at the stable age-stage distribution. It was calculated as follows:

$$T = \frac{\ln R_0}{r}$$

The finite rate of increase was calculated as:

$$\lambda = e^r$$

The age-specific life expectancy (e_x) was calculated according to Chi and Yang (2003) and defined as the time that an individual of age x is expected to live. The life expectancy for individuals in different age-stage-sex units was calculated as:

$$e_{(x)} = \sum_{i=x}^{n} \sum_{j=y}^{m} s'_{ij}$$

The age-stage-specific reproductive value (v_x) of female *T. urticae* was calculated for an individual of age x and stage j to the future population according to Tuan *et al*. (2016).

RESULTS

Eggs of *T. urticae* were exposed to each acaricide by the contact method, and LC_{50} values were determined based on concentration-response analyses. Based on the obtained LC_{50} values, cyflumetofen and clofentezine were highly toxic against the egg stage of *T. urticae* (Table 1). Clofentezine (LC_{50} = 0.42 µg a.i./mL) was significantly more toxic than cyflumetofen (LC_{50} = 0.80

µg a.i./mL) against the egg stage of *T. urticae* (based on non-overlapping 95% confidence limits of the LC_{50} values).

Table 1. Toxicity of cyflumetofen and clofentezine against eggs of *Tetranychus urticae*.

Treatment	df	Slope ± SE	X^2	Lethal concentration ((µg a.i./mL) 95%FL*)		
				LC_{25}	LC_{50}	LC_{90}
clofentezine	3	2.20 ± 0.26	70.33	0.21 (0.15–0.26)	0.42 (0.34–0.50)	1.59 (1.2–2.44)
cyflumetofen	3	2.68 ± 0.37	50.04	0.45 (0.33–0.55)	0.80 (0.68–0.93)	2.42 (1.86–3.70)

* FL = Fiducial Limits
The values inside the parenthesis are fiducial limits.

Table 2. Life history traits of offspring from *T. urticae* eggs treated with clofentezine and cyflumetofen.

Sex	Stage	mean developmental time (± SE)*				
		Control (day)	Clofentezine LC_{25} (day)	LC_{50} (day)	Cyflumetofen LC_{25} (day)	LC_{50} (day)
Female	**egg**	2.66 ± 0.11c	4.0 ± 0.12b	4.02 ± 0.21b	3.99 ± 0.13b	5.1 ± 0.2a
	nymph	4.0 ± 0.12c	4.97 ± 0.21b	4.84 ± 0.27b	5.0 ± 0.11b	5.85 ± 0.2a
	adult	9.0 ± 0.3a	5.21 ± 0.39b	3.78 ± 0.4c	3.99 ± 0.22c	4.07 ± 0.32c
	TPA[a]	7.01 ± 0.17c	9.02 ± 0.25b	9.44 ± 0.38b	9.60 ± 0.21b	10.91 ± 0.34a
	APOP[b]	3.4 ± 0.05a	0.3 ± 0.15b	0.33 ± 0.01b	0.26 ± 0.01b	0.29 ± 0.09b
	TPOP[c]	7.03 ± 0.2c	9.33 ± 0.32b	9.96 ± 0.4b	9.86 ± 0.28b	10.99 ± 0.20a
	total longevity	17.2 ± 0.38a	14.90 ± 0.53b	13.69 ± 0.56c	15.44 ± 0.26b	13.99 ± 0.34c
	fecundity	14.82 ± 0.75a	4.37 ± 0.39b	4.16 ± 0.81bc	2.76 ± 0.29c	2.17 ± 0.49c
Male	**egg**	2.03 ± 0.16c	3.95 ± 0.19b	4.95 ± 0.15a	4.0 ± 0.11b	4.99 ± 0.2a
	nymph	4 ± 0.12c	5.0 ± 0.2b	5.95 ± 0.15a	5.0 ± 0.23b	5.06 ± 0.2b
	adult	11.83 ± 0.5a	7.33 ± 0.43b	5.02 ± 0.34c	4.97 ± 0.3c	4.23 ± 0.32c
	TPA	6.83 ± 0.23c	9.38 ± 0.32b	10.97 ± 0.24a	9.87 ± 0.25b	10.43 ± 0.39ab
	total longevity	18.51 ± 0.51a	17.26 ± 0.61b	15.27 ±0.33c	16.76 ± 0.37b	15.13 ± 0.39c

* Each treatment was estimated with the bootstrap technique using 100,000 replications, the SEs were estimated using 100,000 bootstraps and compared by paired bootstrap test (comparison of 95% CL.). a: Total pre-adult; b: adult pre-oviposition period; c: total pre-oviposition period.

Sublethal (LC_{25} and LC_{50}) exposure studies revealed that both clofentezine and cyflumetofen had significant negative effects on the life table parameters of *T. urticae*. The sublethal effects of both acaricides on the developmental stages of *T. urticae* are summarized in Table 2. Compared to the control, the number of eggs laid per female and adult longevity were significantly reduced following sublethal exposure. The highest longevity of both female and male mites was observed in control mites (17.2 and 18.5 days, respectively), while the shortest longevity was recorded for *T. urticae* exposed to LC_{50} concentrations of clofentezine or cyflumetofen (13.7 and 14.0 days for females, respectively, and 15.3 and 15.1 days for males, respectively). The mean total pre-adult developmental times of female and male mites were significantly increased following treatment with an LC_{25} or LC_{50} concentration of either acaricide. While the mean development time of adults (both female and male) exposed to sublethal concentrations of each acaricide was significantly reduced.

When the mites were exposed to LC_{25} or LC_{50} concentration of cyflumetofen or clofentezine, the duration of the egg and nymphal (for both female and male mites) stages were significantly increased. Compared to untreated females, the pre-oviposition period (APOP) of *T. urticae* exposed to these acaricides significantly increased (0.30- and 0.33-day following exposure to LC_{25} and LC_{50}

concentrations of clofentezine, respectively, and 0.26- and 0.29-day following exposure to LC_{25} and LC_{50} concentration of cyflumetofen, respectively) (Table 2). The total pre-oviposition period (TPOP) of mites treated with clofentezine or cyflumetofen was significantly higher than control mites; however, there were no significant differences among *T. urticae* treated with sublethal concentrations of clofentezine or cyflumetofen.

The probability that an egg will survive to age *x* and develop to stage *j* was estimated on the basis of the age-stage specific survival rate (S_{xj}) (Fig. 1). An obvious overlap phenomenon was observed in the curves that resulted from the various developmental rates among the individuals. Nymphs that emerged from the treated eggs by acaricides had considerably lower S_{xj} than those from the control mites (Fig. 1). Importantly, an extended developmental time and prolonged overlap between immature and adult stages in acaricide-exposed individuals was observed. The survival rate of female mites was 0.41 and 0.22 following exposure to LC_{25} and LC_{50} concentrations of clofentezine, respectively, 0.44 and 0.33 following exposure to LC_{25} and LC_{50} concentrations of cyflumetofen, respectively, and 0.53 for control-treated mites. Age-specific survivorship (l_x) (Fig. 2) indicates with more clarity the significant reduction in survivorship of the acaricide-treated (LC_{25} and LC_{50} concentrations) mites in comparison to control mites. Age-specific fecundity (m_x) (Fig. 3) was also significantly reduced by acaricide exposure in comparison to the control insects.

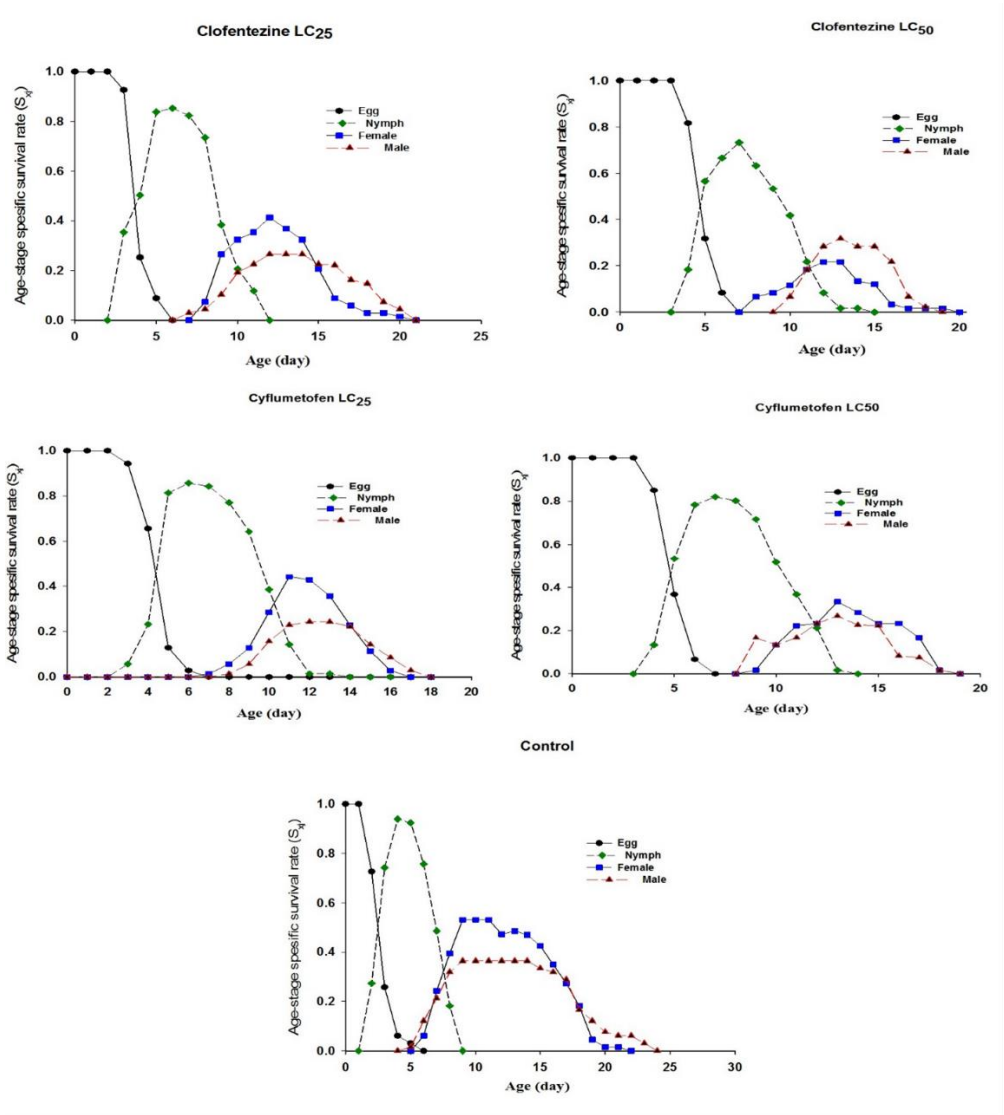

Figure 1. Age-stage-specific survival rate (S_{xj}) of offspring from *T. urticae* eggs treated with LC_{25} and LC_{50} of cyflumetofen and clofentezine.

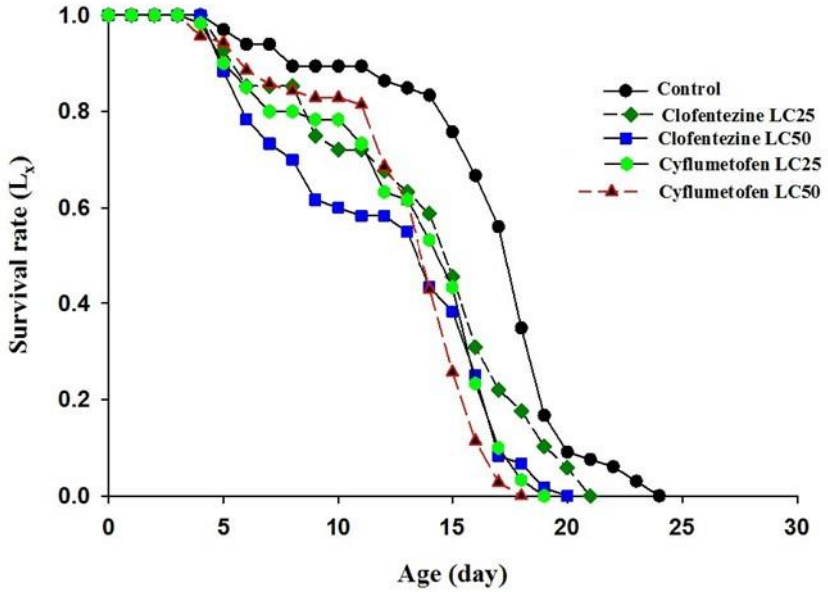

Figure 2. Age-specific survival rate (l_x) of offspring from *T. urticae* eggs treated by LC_{25} and LC_{50} of cyflumetofen and clofentezine compared with control.

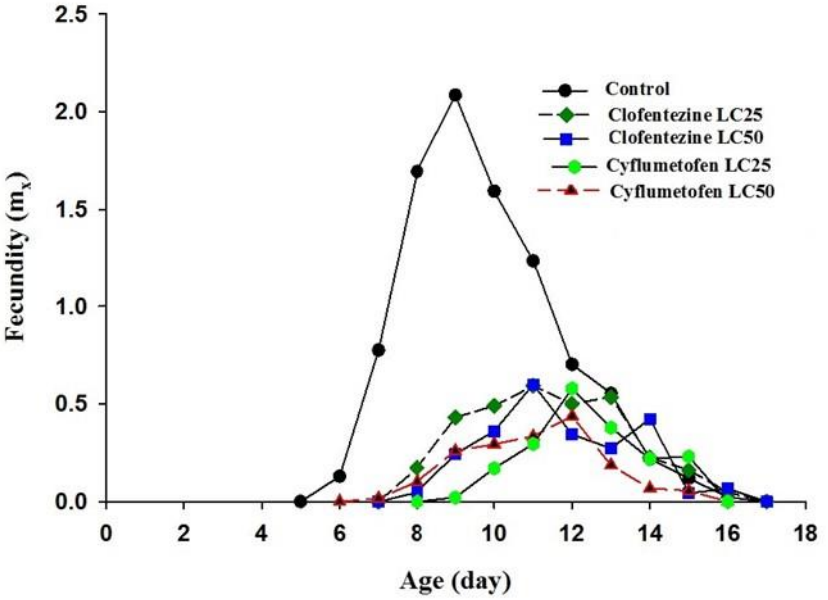

Figure 3. Age-specific fecundity (m_x) of offspring from *T. urticae* eggs treated by LC_{25} and LC_{50} of cyflumetofen and clofentezine compared with control.

Age-specific life expectancy (e_x) gives the expected life span of an individual of age x based on the age-stage and two-sex life table. Overall, the life expectancy of *T. urticae* treated with an LC_{50} concentration of cyflumetofen was shorter in all age and stage groups compared to clofentezine-treated and control mites. For example, at age three days, the life expectancy of mites exposed to LC_{25} and LC_{50} concentrations of clofentezine was 10.89 and 9.26 days, respectively, and 10.29 and 10.08 days, respectively, following exposure to cyflumetofen, whereas the curve of e_x for control mites indicated a life expectancy of 13.72 days (Fig. 4). The peak of the reproductive values (v_x) of mites that were exposed to clofentezine or cyflumetofen at LC_{25} or LC_{50} concentrations was 2.30 and

1.96 days (clofentezine), respectively; and 1.44 and 1.42 days (cyflumetofen), respectively; and 5.71 days for control mites. Reproductive values of *T. urticae* exposed to an LC_{25} concentration of acaricide were significantly lower than that of the control mites (Fig. 5).

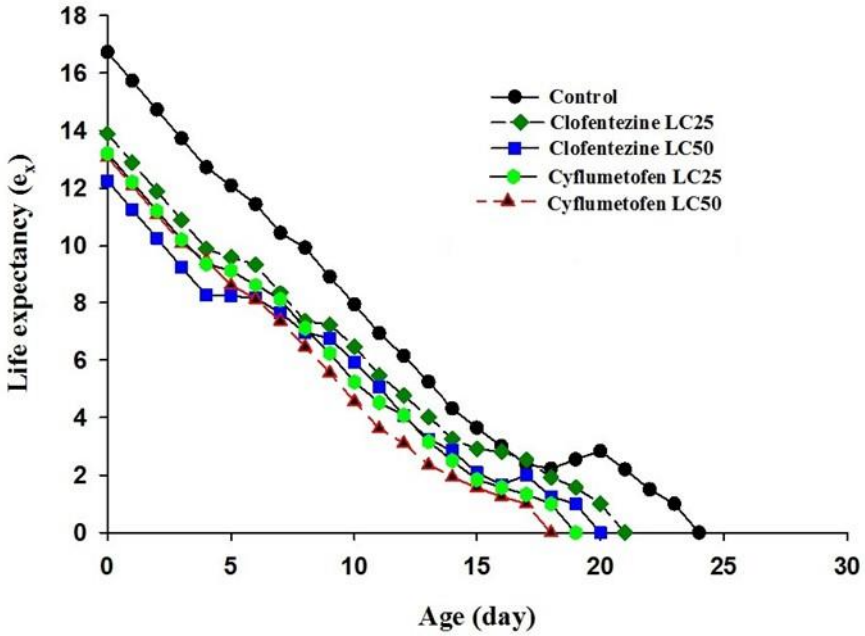

Figure 4. Age-specific life expectancy (e_x) of offspring from *T. urticae* eggs treated by LC_{25} and LC_{50} cyflumetofen and clofentezine compared with control.

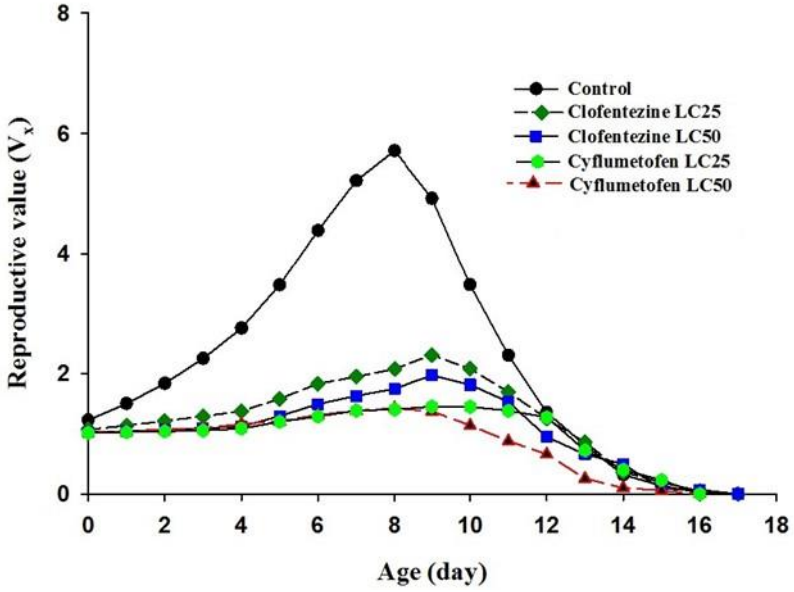

Figure 5. Age- reproductive value (v_x) of offspring from *T. urticae* eggs treated with LC_{25} and LC_{50} of cyflumetofen and clofentezine compared with control.

The demographic parameters of *T. urticae* exposed to LC_{25} and LC_{50} concentrations of cyflumetofen or clofentezine as well as controls are shown in Table 3. The paired bootstrap test revealed that there are significant differences between the acaricide-treated and control mites for all

of the parameters. The intrinsic rate of increase (*r*), the net reproductive rate (R_0), the finite rate of increase (λ), and gross reproduction rate (*GRR*) of acaricide-treated (LC_{25} and LC_{50}) mites were significantly decreased compared to the control mites. Mites exposed to an LC_{50} (0.012 d^{-1}) concentration of cyflumetofen showed the lowest intrinsic rate of increase, while the mites in the control group showed the highest intrinsic rate of increase (0.202 d^{-1}). The mean generation time (T) of acaricide-treated (LC_{25} and LC_{50}) mites increased; however, the progeny population of cyflumetofen-treated mites showed no significant difference from the control mites (Table 3).

Table 3. Life-table parameters of offspring from *T. urticae* females treated with acaricides.

Treatment	Concentration	Population parameter (means ± SE)*				
		r female offspring/female/day	***λ*** (rate of increase/time/individual)	***R₀*** (female offspring/female)	***T*** (days)	***GRR*** (total offspring/female)
Control	0.0	0.202 ± 0.01a	1.82 ± 0.01a	8.12 ± 0.85a	10.34 ± 0.14b	8.62 ± 1.0a
clofentezine	LC_{25}	0.041 ± 0.01b	1.04 ± 0.01b	1.61 ± 0.25b	12.36 ± 0.32a	2.66 ± 0.37b
	LC_{50}	0.021 ± 0.02c	1.022 ± 0.02b	1.3 ± 0.37b	12.26 ± 0.82a	1.90 ± 0.59b
cyflumetofen	LC_{25}	0.016 ± 0.01c	1.016 ± 0.01b	1.24 ± 0.20b	13.19 ± 0.51a	1.35 ± 0.27b
	LC_{50}	0.012 ± 0.01c	1.01 ± 0.01b	1.16 ± 0.18b	11.76 ± 0.68a	1.14 ± 0.25bc

* Means followed by different letters in the same columns are significantly different between treatments by using the paired bootstrap test at 5% significance level.

CONCLUSIONS AND DISCUSSION

In this study, the effects of acute and chronic exposure to the acaricides cyflumetofen and clofentezine were determined on *T. urticae*. Our results were in agreement with previous studies that report that clofentezine is highly toxic to eggs of *Amphitetranychus viennensis* (Zacher) (Marcic 2003; Li *et al*. 2006). There is little published information about the effects of cyflumetofen on mites. Hayashi *et al*. (2013) showed that cyflumetofen is the most toxic to phytophagous mites such as *T. urticae* (with LC_{50} = 1.1 mg/L against eggs) and *Panonychus citri* (McGregor) (LC_{50} = 2.9 mg/L against eggs). They also reported that cyflumetofen does not affect *Apis mellifera* L. under maximum dose (100 µg $female^{-1}$) conditions following OECD test guidelines (TG214) (Hayashi *et al*. 2013). Similarly, Abdel-Rahman and Ahmed (2018) showed that cyflumetofen has significant negative effects on *T. urticae*. The evidence from our study indicated that cyflumetofen and clofentezine are severely toxic against *T. urticae*.

Ecotoxicological analyses based on population growth rate parameters provide accurate predictions of the impacts of pesticides and other toxicants. Demographic toxicology measurements or life table analyses combine lethal and sublethal effects of a toxicant (Stark and Banks 2003). There is little research on the sublethal effects of novel acaricides, such as cyflumetofen on the two-spotted spider mite. This study advances the limited knowledge of sublethal and lethal effects of cyflumetofen and clofentezine on two-sex life table parameters of a progeny generation of *T. urticae*. These acaricides affected the developmental time of female and male mites, indicating that low concentrations of these compounds had a significant negative effect on *T. urticae* life history. These acaricides affected developmental time, adult female and male longevity, and fecundity. Similarly, Abdel-Rahman and Ahmed (2018) found that the fecundity, fertility, and longevity of *T. urticae* are significantly affected following exposure to cyflumetofen. Our results showed that treatment of eggs of *T. urticae* with LC_{25} and LC_{50} concentrations of either acaricide decreased the intrinsic rate of increase (*r*). However, cyflumetofen showed a greater effect on *r* compared with clofentezine. These findings are in agreement with the studies of Havasi *et al*. (2018), Wang *et al*. (2014), Martinez-Villar *et al*. (2005), and Marcic (2003, 2007) for *T. urticae* treated with diflovidazin, bifenthrin, azadirachtin, spirodiclofen, and clofentezine, respectively. The intrinsic rate of increase (*r*) is based

on the overall effects of both survivorship (l_x) and fecundity (m_x), and is considered the best parameter in terms of an ecologically significant bioassay. Similar effects on life table parameters have been reported by Li *et al*. (2006) and Havasi *et al*. (2018). Li *et al*. (2006) showed the effects on life table parameters following treatment of immature stages of *A. viennensis* with several concentrations of clofentezine. Havasi *et al*. (2018) reported that diflovidazin (tetrazine group acaricide) affects the *r* of *T. urticae* at sublethal concentrations. The latter study indicated that exposure to LC_{25} and LC_{50} concentrations cause a greater reduction in the intrinsic rate of increase (*r*). These results agree with those Abdel-Rahman and Ahmed (2018) who demonstrated that the *r* and λ of *T. urticae* are significantly reduced following exposure to an LC_{50} concentration of cyflumetofen. Whereas only moderate or minor effects are found in *Phytoseiulus persimilis* Athias-Henriot.

Because of differences in developmental rates among individual *T. urticae*, the survival rate curves usually show differences depending upon insect stage and significant stage overlap. Based on our data, lethal and sublethal concentrations of clofentezine or cyflumetofen played negative roles during all pre-adult developmental stages (i.e., egg and nymphal stages) in both male and female mites. Our findings are in agreement with the studies of Li *et al*. (2017) and Alinejad *et al*. (2015). Li *et al*. (2017) reported a significant difference in both males and females that were treated by sublethal concentrations of bifenazate during pre-adult stages of *T. urticae*. Similarly, significantly longer development times for both sexes were reported following treatment with fenazaquin (Alinejad *et al*. 2015). While, Havasi *et al*. (2018) showed that pre-adult stages of *T. urticae* are not influenced by sublethal concentrations of diflovidazin. Differences in the mode of action of the acaricides may be a significant contributory factor in these differences. In our study, exposure to LC_{25} and LC_{50} concentrations of clofentezine or cyflumetofen resulted in significant negative effects on the longevity and mean developmental time of both female and male adults. Similarly, in the studies of Martinez-Villar *et al*. (2005) and Wang *et al*. (2014), a significant decrease occurred in the longevity for mites treated with azadirachtin and bifenthrin, respectively. Our current results showed that exposure to lethal and sublethal concentrations of each acaricide significantly affects the APOP and TPOP of *T. urticae*. In contrast, Bozhgani *et al*. (2018) reported that there are no significant effects on APOP among different experimental treatments (LC_{10}, LC_{20}, and LC_{30}) of chlorfenapyr.

Compared to control mites, we found that the net reproductive rate (R_0) and the gross reproductive rate (*GRR*) were significantly reduced in spider mites treated with LC_{25} and LC_{50} concentrations of clofentezine and cyflumetofen. These findings are in line with those of Havasi *et al*. (2018) and Marcic (2003), who studied sublethal concentrations of diflovidazin (LC_{10} and LC_{20}) and clofentezine on the two-spotted spider mite, respectively. Our results are also in agreement with those of Alinejad *et al*. (2015). Compared to control mites, the mean generation time (*T*) of *T. urticae* treated with LC_{25} and LC_{50} concentrations of cyflumetofen or clofentezine significantly increased. Alinejad *et al*. (2015) found that the doubling time (*DT*) of spider mites treated by an LC_{20} concentration of diflovidazin is higher in comparison to control mites. Based on the results of the present study, mites treated with cyflumetofen or clofentezine showed significantly reduced age-specific survival rate (l_x) values and age-specific maternity ($l_x m_x$). Our observations were consistent with the results reported by Li *et al*. (2017) which examined the sublethal concentrations (LC_{10} and LC_{20}) of bifenazate on *T. urticae*. The life expectancy (e_x) of *T. urticae* was significantly decreased when it was exposed to a sublethal concentration of cyflumetofen or clofentezine. A similar decrease in e_x (along with ovicidal effects) was found for other acaricides, such as flufenzine and cofentezine (Marcic 2003; Abdel-Rahman and Ahmed 2018). The reproductive value (v_x) of *T. urticae* treated with LC_{25} and LC_{50} concentrations of clofentezine or cyflumetofen was significantly decreased in comparison to control mites. These outcomes are in agreement with those of Marcic (2003), who showed that clofentezine significantly reduces mite reproductive value and life expectancy. The analytical techniques applied in this study allow thorough quantification of the significance of a pesticide's impact on life table parameters at the population level of a species such as *T. urticae*. This level of analysis and understanding would not be possible without population-based approaches.

According to our results, survivorships and fecundity were significantly different between spider mites exposed to a sublethal concentration of clofentezine or cyflumetofen and control mites. Demonstrative changes in life table parameters of the two-spotted spider mites in this study showed that the acaricides cyflumetofen and clofentezine can influence life-history traits of *T. urticea* at low lethal rates.

In conclusion, using ecotoxicological approaches is improving the evaluation of acaricides and other pesticides in integrated pest control programs. Study of sublethal effects on life table parameters of pests allows us to have the most complete description of the population-level responses to pesticides. The relationships between sublethal effects and the corresponding life history traits can be examined by performing analyses of life table parameters. The effects of cyflumetofen and clofentezine on *T. urticae* suggest that a combination of toxic and sublethal effects could lead to the incorporation of these compounds in management programs against this important pest.

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اثرهای کشندگی و زیرکشندگی سایفلومتوفن و کلوفنتزین بر فراسنجه‌های جدول زندگی کنهٔ تارتن دو لکه‌ای *Tetranychus urticae* (Acari: Tetranychidae)

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چکیده

اثرهای سایفلومتوفن و کلوفنتزین روی *Tetranychus urticae* Koch در شرایط آزمایشگاهی بررسی شد. مقادیر LC_{50} سایفلومتوفن و کلوفنتزین در برابر تخم به ترتیب ۰/۸ و ۰/۴۲ میکروگرم ماده موثر بر میلی لیتر بود. اثرهای زیرکشنده این ترکیبات بر فراسنجه‌های جدول زندگی *T. urticae* با قرار دادن کنه‌ها در معرض غلظت‌های LC_{50} و LC_{25} ارزیابی شد. مدت رشد و نمو *T. urticae* نر و ماده به مقدار زیادی تحت تاثیر هر دو کنه‌کش قرار گرفت. افزون بر این، قرار گرفتن در معرض غلظت های LC_{50} و LC_{25} سایفلومتوفن و یا کلوفنتزین به مقدار فراوانی باعث کاهش نرخ خالص تولیدمثل (R_0) ، نرخ متناهی افزایش (λ) و نرخ ذاتی افزایش (r) شد. تیمارهای LC_{50} و LC_{25} اثرهای قوی بر زنده‌مانی، باروری و نرخ ذاتی افزایش نشان دادند. یافته‌های این پژوهش نشان داد که قرار گرفتن در معرض غلظت‌های کشنده و زیرکشنده سایفلومتوفن و یا کلوفنتزین منجر به تغییر بسیاری در ویژگی‌های تاریخچهٔ زندگی *T. urticae* می‌شود. یافته‌های این پژوهش نشان می‌دهد که این کنه‌کش‌ها نامزدهای امیدوارکننده‌ای برای استفاده در برنامه‌های مدیریتی این آفت مهم هستند.

واژگان کلیدی: کنه‌کش، اثر زیستی، زیست‌سنجی، کنه‌های گیاهخوار، کنهٔ تارتن دو لکه‌ای.

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