Influence of sublethal exposure to triflumuron on the biological performance of *Tetranychus urticae* Koch (Acari: Tetranychidae)

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**Abstract**

Effects of sublethal exposure to triflumuron on the biological performance of the two-spotted spider mite *Tetranychus urticae* Koch were analysed under laboratory conditions. Survivorship was affected by the compound. Triflumuron caused a reduction both in the percentage of eggs that developed to adults and in the survival of adult stage. Triflumuron also affected the fecundity. The net reproductive rate (*R₀*), the intrinsic rate of increase (*rₘ*), and the finite rate of increase (*λ*) of treated females were lower than in those non treated, resulting in a reduction of population growth. These results suggest that triflumuron could be a valuable addition in integrated pest management programs of *T. urticae*, although more laboratory, semi-field and field testing is required.

**Additional key words:** benzoylphenyl ureas, intrinsic rate of increase *rₘ*, life-table, triflumuron, two-spotted spider mite.

**Introduction**

The two-spotted spider mite, *Tetranychus urticae* Koch (Acari: Tetranychidae) has been recorded on more than 150 hosts of some economic value throughout the world (Jeppson *et al.*, 1975), being the most polyphagous spider mite of the tetranychids and a key pest for many kind of crops in temperate regions.

Currently, great efforts are directed towards a reduction in the use of traditional pesticides and towards an increase in the use of Integrated Pest Management (IPM) techniques. Therefore, the search for pesticides that are compatible with IPM programs, such as benzoylphenyl ureas (BPUs) is an interesting approach.

BPUs inhibit chitin synthesis in a wide range of insect groups, resulting in abortive moulting. They act mainly as larvicides and ovicides (Retnakaran and Wright, 1987). Effects on adults fecundity, fertility, and longevity have also been reported (Marco *et al.*, 1998; Perveen, 2000; Medina *et al.*, 2002). Acaricidal activity
has been also mentioned by several authors for various spider mites, including *T. urticae* (Grosscurt *et al.*, 1988; Scheltes *et al.*, 1988; Ahn *et al.*, 1993; Sáenz-de-Cabezón *et al.*, 2002, 2003), but little information is available about their effects on this kind of pests.

In ecotoxicology, the ability to predict the effects of toxicants on the dynamics of natural populations can be seen as a major objective (Moe *et al.*, 2001). Standardized test procedures have been formulated to estimate critical-effect levels (LCx, ECx) from concentration-response relationships for single life cycle variables such as mortality, growth, or reproduction (Kammenga *et al.*, 1997). Then, demographic studies allow the integration of several critical life cycle traits into a single variable (Van Leeuwen *et al.*, 1985). Several authors have argued that the best approach for evaluation of the total effect of a xenobiotic is life table analysis or demographic toxicology (Stark and Wennegren, 1995). Then, the use of the intrinsic rate of increase (r<sub>n</sub>) has been recommended (Allan and Daniels, 1982), because it is based on both survivorship and fecundity. This parameter is also a measure of the ability of a population to increase in an unlimited environment (Stark and Wennegren, 1995).

Triflumuron (like other BPUs) can be slow acting and exert sublethal as well as lethal effects. Then, the compound can affect more than one life stage and pesticide persistence is an important consideration when trying to estimate their effects. Therefore, in this work, the effects of a sublethal dose of triflumuron against *T. urticae* were evaluated using demographic toxicological analysis. The possibility of incorporating this compound in the management of the spider mite was discussed.

**Material and Methods**

**Colony source**

A laboratory colony was used for the bioassay. The colony was collected from a natural population on ornamental crops in 2000 and maintained since then on young pesticide-free green bean plants (*Phaseolus vulgaris*, cv. Garrafal). The plants were introduced in acrylic cages (40 by 40 by 55 cm), placed in a climatic chamber at 24 ± 1°C, 65 ± 5% RH, and 16:8 (L:D). The bioassay was performed under the same conditions.

**Chemical**

The commercial formulation of BPU triflumuron, namely Alsystin® ([wettable powder] 250 g [ai] kg<sup>-1</sup>), Bayer Hispania (Spain), was used for the bioassay.

**Application of insecticide**

One newly ecdysed adult female was placed on each one of the green bean leaf discs (2 cm diameter) used in the bioassay and provided with two adult males. Eight leaf discs with the mites were introduced in each rearing unit consisting of wet filter paper inside a Petri dish (9 cm diameter). Dish top had two holes of 6 mm diameter each to prevent moisture. The rearing units were treated using a Potter Precission Laboratory Spray Tower (Potter, 1952), with 5.5 ml of an aqueous solution of 1g L<sup>-1</sup> of Alsystin®. An air compressor (Burkard 0523-703 Q-R32X; 50 kPa) was used to apply the insecticide. This resulted in a homogeneous spray coverage of 5 ± 0.5 μl (mean ± SE) fluid per square centimeter. Controls were treated with distilled water alone. The concentration was chosen based on previous bioassays (Sáenz-de-Cabezón *et al.*, 2002), because it has a lower short term effect than the LC<sub>50</sub> against the more susceptible developmental stage of the mite.

**Life-table parameters**

Adults were maintained in the same leaf discs throughout the whole bioassay. Daily records for preoviposition and oviposition periods and fecundity were made. Life table parameters were taken until the death of the last individual. In order to obtain a non detectable error doubt to the bioassay, 13 rearing units were sprayed with the compound and 13 with distilled water according with Rejman and Jesiort (1977).

To examine any possible carry-over and residual activity of triflumuron on the offspring of treated adults, 100 eggs from randomly selected females of each treatment were transferred individually to green bean leaf discs treated with the same methodology as described above. Percentage hatchability, mortality,
time of development of each stage, and days to adulthood were monitored once a day.

Statistical methods

Once determined the preoviposition period, the prereproductive survival and the emergence matrixes, rm 2.0 program (Taberner et al., 1993) was used to establish the natural parameters. The data analysis was carried out using Bootstrap technique doing 1,000 replicates as suggested by Meyer et al. (1986). The chosen sex ratio was 1 male to 3 females, according with Helle and Pijnaker (1985). Nine life table parameters were compiled (Table 1).

Results

Examination of the \( l_x \) and \( m_x \) (as defined in Table 1) in untreated mites revealed low mortality in the immature stages (20% of eggs and 10% of larval and nymphal stages) with a 70% chance of reaching adulthood. In contrast, treated females showed higher mortality in the immature stages (40% of eggs and 16% of larval and nymphal stages) with a 44% chance of reaching adulthood (Fig. 1). The untreated mites spent half of their lives in the adult stage and did not reach their peak of reproduction (\( mx = 6.0 \)) until the adults were 9 days old, but the probability of reaching this age was only 8.8%. No delay in time reaching reproductive maturity was observed in treated mites. The effects of mortality on the population growth can be demonstrated by simply comparing the net reproductive rate (\( R_0 \)) (Table 2).

Due to mortality effects, the average treated female only produced 5.0 females, almost three-fold less than untreated mites (14.4). For untreated mites, the mean generation time (\( T \)) was 13.97 days, and the population increased daily by 1.22 times the previous day’s total number (finite rate of increase, \( \psi \)). Every 3.51 days (doubling time, \( DT \)) the population doubled. Generation time of the treated mites did not differ much from generation time of the untreated ones. The population increased daily by 1.13 times. Every 5.73 days the population doubled. Untreated mites had significantly higher \( rm \) values (0.198 \pm 0.005) than the treated ones (0.121 \pm 0.021), with no overlap of their confidence intervals at 95% (Table 2).

Discussion

The \( rm \) value integrates age at first reproduction, survivorship, brood size and frequency, and longevity. However, it is also worth examining how chronic exposure affects the individual components of \( rm \). In our experimental conditions, the probability that individuals can reach the peak of reproduction and the physiological maximum of reproduction (\( l_x \) and \( m_x \) values in that maximum, respectively) was lowered by triflumuron. This caused a reduction in the number of
daughters that replace an average female in course of a generation \((R_0)\). At this point, it is interesting to arrange that the physiological reproductive stage of the females treated seems to be a key factor in order to determine the effect on fecundity (Sáenz de Cabezón et al., 2002).

It is appropriate to underline the importance of the mortality in eggs laid by treated females. Egg hatch inhibition after adult treatment with triflumuron has been reported for different insect species, especially Diptera as *Ceratitis capitata* (Wiedemann) (Casana-Giner et al., 1999). Nevertheless, this activity has been rarely studied for BPUs and spider mite pests; Grosscurt et al. (1988) observed an ovicidal activity of PH 70-23 (flucycloxuron) against *T. urticae* as a consequence of a transovarial transmission of the compound; Ahn et al. (1993) also observed a substantial reduction in egg viability when treating *T. urticae* deutonymph females with flufenoxuron; other kinds of insect growth regulators such as azadirachtin also caused a reduction in the percentage of eggs

![Figure 1. Survivorship \((l_x)\) and fecundity \((m_x)\) curves for *Tetranychus urticae* reared on green bean leaf discs treated with distilled water (control) and with a low mortality concentration (1 g L\(^{-1}\)) of Alsystin® ([wettable powder] 250 g of triflumuron kg\(^{-1}\)), at 24 ± 1°C, 65 ± 5% RH, and 16:8 (L:D).

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Control</th>
<th>Triflumuron</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gross reproductive rate ((GRR))</td>
<td>85.88</td>
<td>25.20</td>
</tr>
<tr>
<td>Net reproductive rate ((R_0))</td>
<td>14.40</td>
<td>5.016</td>
</tr>
<tr>
<td>Mean generation time ((T, \text{days}))</td>
<td>13.97</td>
<td>13.61</td>
</tr>
<tr>
<td>Intrinsic rate of increase ((r_m))</td>
<td>0.198 ± 0.005</td>
<td>0.121 ± 0.021</td>
</tr>
<tr>
<td>Confidence interval 95%</td>
<td>0.188 – 0.208</td>
<td>0.061 – 0.179</td>
</tr>
<tr>
<td>Finite rate of increase ((\psi))</td>
<td>1.219</td>
<td>1.129</td>
</tr>
<tr>
<td>Doubling time ((DT, \text{days}))</td>
<td>3.51</td>
<td>5.78</td>
</tr>
<tr>
<td>% of eggs surviving to adults</td>
<td>70</td>
<td>44</td>
</tr>
</tbody>
</table>
hatched when the compound was applied to *T. urticae* adult females (Dimetry *et al.*, 1993). However, Sáenz de Cabezón *et al.* (2002) observed no effect on the fertility when a sublethal concentration of triflumuron (0.75 g L\(^{-1}\) of Alsysistin \(^{8}\)) to *T. urticae* adults was applied. Therefore, it seems that the concentration is essential to determine the effect level of triflumuron on adult fertility.

Although it is of value to examine how chronic exposure affects the individual components of \(r_m\), the comparison of this parameter values provides insight beyond that available from independent analysis of several life-history parameters (Petitt *et al.*, 1994). In our bioassay, the untreated mites had a significantly higher \(r_m\) value than the treated ones. Therefore, the compound reduced the ability to the populations of *T. urticae* to increase.

In conclusion, more semi-field and field testing is required before the acaricidal potential of triflumuron can be considered completely investigated, but the estimation of critical-effect levels from concentration-response relationship for single life cycle variables as mortality, fecundity and fertility performed by Sáenz de Cabezón *et al.* (2002), together with our estimation of the produced sublethal effects could lead to the incorporation of this compound in IPM programs against *T. urticae*.

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**References**


