

Influence of application methods on Confidor detoxification and tomato fruit quality in greenhouse cultivation

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Abstract

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The detoxification dynamics of Confidor in greenhouse tomato plants were evaluated under two application methods: traditional foliar spraying and drip irrigation (insectigation). The study also assessed the impact of these methods on key fruit quality parameters. Results showed that the application method significantly influenced residue dynamics, fruit quality, and safety. Foliar spraying led to rapid penetration into leaves and fruits, followed by gradual detoxification. The greatest residue reduction occurred within the first three days post-application, reaching 35.3% in leaves and 43.8% in fruits. In contrast, insectigation resulted in lower initial residues but a steady accumulation over time, with a 212.5% increase (threefold rise) by day five. While foliar spraying accelerated surface residue dissipation, insectigation prolonged systemic effects, enhancing sugar and ascorbic acid levels and improving overall fruit quality. By day five, Confidor residues in tomato fruits remained at or below the Maximum Residue Limit (MRL) of 0.5 mg/kg, depending on the application method. These findings highlight the need for careful selection of treatment strategies and Pre-Harvest Intervals to balance regulatory compliance, consumer safety, and fruit quality optimization.

Keywords: foliar spraying; insectigation; Imidacloprid; ascorbic acid; sugars; titratable acidity

Abbreviations: PHI – Pre-Harvest Interval; MRLs – Maximum Residue Levels; LSD – The Least Significant Difference; SAR – sugar-acidity ratio; ROS – reactive oxygen species

Introduction

Environmental and health concerns associated with chemical pesticides necessitate safer and more efficient pest-control strategies.

There have been studies examining the impact of applying insecticides through irrigation systems – a practice sometimes referred to as “insectigation” or more commonly “chemigation” – on crop quality. Research has indicated that this method can offer several benefits, including uniform distribution of insecticides, reduced application costs, and minimized exposure to non-target species. Unlike traditional foliar spraying, chemigation ensures consistent application irrespective of

weather conditions. It protects the entire plant, often achieving equal or superior pest control with fewer applications. However, chemigation can be applied only if the product label allows such application and only according to the label directions and restrictions (Sreeremya, 2017; He et al., 2018; Sujithra et al., 2020; Zheng et al., 2020; Li et al., 2022; Yan et al., 2023).

Confidor (active constituent: 200 g/l imidacloprid) is a systemic insecticide approved for both foliar spraying and root application in greenhouse conditions. It effectively controls pests while minimizing external inputs. However, Pre-Harvest Intervals (PHIs) are typically established for foliar application, necessitating re-evaluation for root-system use to ensure safety and regulatory compliance.

Regarding the impact of insectigation on crop biochemical key parameters, research remains limited, highlighting the need for further studies.

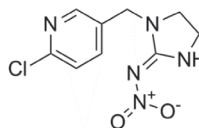
The objective is to determine whether PHIs for foliar applications can be safely extended to root-system methods, promoting sustainable agricultural practices.

Materials and Methods

The experiments were conducted during the period 2022–2024 in the Ararat province, within a greenhouse located in Darakert village, Armenia, using “Cherry Kokta” F1 hybrid tomato plants. The average temperature and relative humidity during the experimental period were 20–26 °C and 65%, respectively. Application rates followed Bayer’s recommendations: 250 mL/ha for foliar spraying and 1 L/ha for drip irrigation. The insecticide Confidor was used in the experiments.

Confidor (Bayer) with imidacloprid as its active ingredient, belongs to the neonicotinoid class, targeting the central nervous system of pests. Its systemic properties and contact-intestinal effects make it effective against various pests, including thrips, flies, moths, and whiteflies. The chemical works by interfering with the transmission of stimuli in the insect nervous system. Specifically, it causes a blockage of the nicotinic neuronal pathway. By blocking nicotinic acetylcholine receptors, imidacloprid prevents acetylcholine from transmitting impulses between nerves, resulting in the insect’s paralysis and eventual death. It is effective on contact and via stomach action (Breaz, 2012).

IUPAC name: *N*-{1-[(6-Chloro-3-pyridyl) methyl]-4,5-dihydroimidazol-2-yl}nitramide:



The experimental plot was divided into three groups (each 70 m² with 175 plants), separated by a buffer zone of 1 m² to avoid cross-contamination:

- Foliar spraying Confidor solution;
- Root application through drip irrigation (insectigation);
- Control group with no treatment.

During the drip application process, plants were initially irrigated with 80% of the planned water volume (1.6 liters). Following the application of the treatment with insecticide, the remaining 20% of water (400 ml) was added.

Leaf and fruit samples (≥ 2 kg) were collected at 1, 3, and 5 days after application. Each sample was divided into two

portions: one was cleaned with wet (a 1:1 volume ratio mixture of water and acetone) cotton to remove surface residues, while the other was left untreated. The amount of pesticide on the fruit and leaf surfaces was calculated as the difference between the total residue (uncleaned samples) and the residue after cleaning (washed samples).

For the drip irrigation treatment, only the total amount of imidacloprid in the uncleaned samples was measured.

Residues were measured using high-performance liquid chromatography (HPLC) with the QuEChERS pesticide analysis method. Recoveries between 85 and 101% and repeatability typically < 5% (Anastassiades et al., 2003).

10 g of homogenized, frozen sample was weighed into a 50 mL centrifuge tube. To this, 10 ml of acetonitrile and 100 µl of an internal standard mixture were added. The tube was sealed and shaken vigorously for 1 minute. The extraction process required the addition of a salt mixture comprising 4 g anhydrous MgSO₄, 1 g NaCl, 1 g trisodium citrate dihydrate, and 0.5 g disodium hydrogen citrate sesquihydrate to the sample. The mixture was sealed in a tube, shaken vigorously by hand for 1 minute, and centrifuged.

Simultaneous water removal and sample cleanup were accomplished using dispersive solid-phase extraction (dispersive-SPE). In this step, 150 mg of anhydrous MgSO₄ and 25 mg of primary-secondary amine (PSA) sorbent were added to 1 ml of the acetonitrile extract. This process efficiently removed polar matrix components, including organic acids, certain polar pigments, and sugars, ensuring a cleaner sample for analysis. Chromatography details are presented in the Figure 1.

At the end of the Pre-Harvest Interval, qualitative fruit indicators in the crop were evaluated using standard biochemical methods (Yermakov et al., 1987). Dry matter content was determined by drying samples in a thermostat at 100–105 °C until a constant weight was achieved. Titratable acidity was measured by titration using phenolphthalein as an indicator with a standard 0.1 N NaOH solution until a faint pink color appeared, and the results were expressed as a percentage of malic acid. Ascorbic acid (vitamin C) content was determined using the Murray titration method. Sugar content was analyzed using Bertrand’s method, which is based on the reducing action of sugars on an alkaline solution of a tartrate complex with CuSO₄.

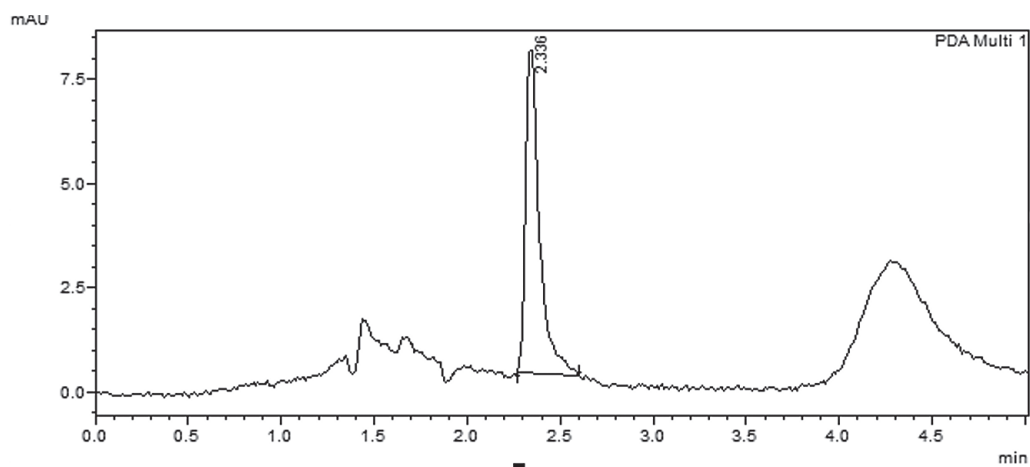
Statistical analysis

All experiments were conducted in four replicates, and the results are presented as the mean ± standard deviation. Statistical comparisons were performed using ANOVA: Single-Factor, with a significance threshold set at *p* = 0.05. Sig-

Fig. 1. Chromatogram of imidacloprid standard (2.33 min)

Analysis was performed using HPLC with a PDA detector at 250 nm. Mobile phase: acetonitrile/water (60:40); flow rate: 1.2 ml/min; injection volume: 20 μ l.

Source: Authors' own elaboration



nificant differences among variants for various traits were assessed using the Least Significant Difference (LSD) test and the Tukey HSD test.

Results

Foliar spraying

The data in Table 1 clearly demonstrate that Confidor effectively penetrates tomato fruits and leaves. One day after application, internal residues account for 64.2% of the total detected in fruits and 70.6% in leaves. Residue levels decline progressively over time.

Within three days, internal residues decrease by 43.8% in fruits and 35.3% in leaves. By day five, they decline further, with an additional 18.5 % reduction in fruits and 27.3% in leaves compared to day three. Surface residues dissipate even faster, decreasing by 60% on fruits and 57.9% on leaves within five days.

Overall, Confidor residues in both leaves and fruits diminish steadily over time following foliar application. This decline results from a combination of surface dissipation and internal metabolic detoxification (Figure 2).

Table 1. Residues of Confidor over time at different time points after foliar spraying ($p < 0.05$)

Residues, mg/kg	Days after the treatment		
	1	3	5
On the surface of the fruit	0.20 ± 0.07	0.12 ± 0.07	0.08 ± 0.03
In the fruit	0.48 ± 0.09	0.27 ± 0.02	0.22 ± 0.02
On the surface of the leaves	0.19 ± 0.10	0.12 ± 0.02	0.08 ± 0.07
In the leaves	0.34 ± 0.04	0.22 ± 0.04	0.16 ± 0.03

Source: Authors' own elaboration

Insectigation

Primarily results in internal residues, as Confidor is absorbed systemically through the plant's roots. One day after drip application, only trace amounts of Confidor are detected in the leaves. By day 3, the leaf residues reached 0.6 ± 0.08 mg/kg and increased by 16.7% to 0.7 ± 0.09 mg/kg on day 5. Residue accumulation was even more pronounced in fruits, where concentrations rose from 0.16 ± 0.03 mg/kg on day 1 to 0.26 ± 0.05 mg/kg on day 3, representing a 62.5 % increase. By day 5, fruit residues reached 0.5 ± 0.02 mg/kg, representing a 92.3 % increase compared with day 3 and a 212.5% increase compared with day 1.

Overall, residue levels in tomato fruits and leaves increased steadily over time compared to day one.

Safety implications

By day 5, at the end of the Pre-Harvest Interval (PHI), Confidor residues in tomato fruits remain at (insectigation) or below (spraying) the Maximum Residue Limit (MRL) of 0.5 mg/kg. This indicates a potential margin of safety for both consumption and exposure (Codex 2009; ER 2005).

However, caution is warranted, as residue levels can be influenced by various factors, including environmental conditions, varietal characteristics, plant development stages, and other endogenous or exogenous variables. These factors may cause fluctuations in residue concentrations during the PHI, potentially leading to deviations from the Maximum Residue Limit (WHO, 2022; Kariyanna et al, 2024).

Impact on fruit quality

We conducted as well biochemical analyses to evaluate the effects of Confidor on key qualitative indicators of tomatoes.

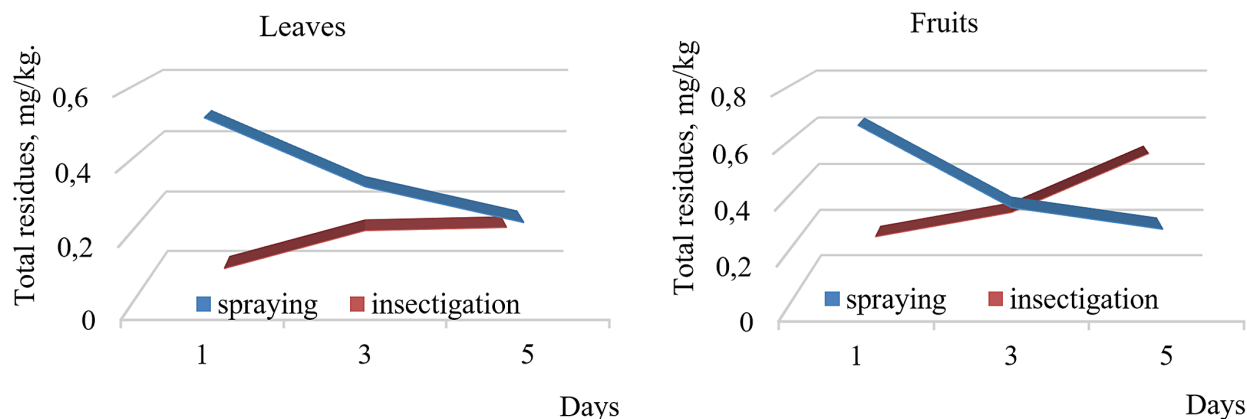


Fig. 2. Total residue dynamics of Confidor in tomato fruits and leaves following foliar and insectigation treatments

Source: Authors' own elaboration

As shown in Table 2, both foliar spraying and insectigation reduced titratable acidity compared to the control ($p = 0.01$), indicating an impact on fruit acidity balance. The values for both application methods were similar, with no statistically significant differences.

The vitamin C (L-ascorbic acid) content increased significantly in both treatments compared to the Control ($p = 0.001$). The differences between foliar spraying and insectigation were minor and fell within the margin of error, suggesting a comparable effect on vitamin C levels.

There was no strong evidence to suggest that monosugar content varied significantly across Control, Spraying, and Irrigation treatments. However, sucrose content showed significant changes in foliar treatments compared to the Control. While both application methods had comparable effects, insectigation exerted a slightly stronger influence ($p = 0.0061$). A similar trend was observed for total sugar content, with both treatments surpassing the Control, and insectigation yielding the highest levels ($p = 0.0082$).

The sugar-acid ratio (SAR) improved in both cases, enhancing fruit sweetness, with insectigation showing a slightly stronger effect.

The statistical analysis revealed significant differences

in residue dynamics between the two application methods ($p < 0.05$). These variations were attributed to the systemic nature of insectigation, which led to sustained residue presence, whereas foliar spraying resulted in more rapid detoxification.

Additionally, the Tukey HSD test conducted on various parameters (sugars, sucrose, vitamin C, dry matter, acidity) indicated that insectigation significantly increased total measurement, suggesting a potential improvement in overall fruit quality or quantity.

Discussion

Residue dynamics in foliar vs. root applications

Foliar spraying

Confidor (Imidacloprid) exhibits lipophilic properties, allowing it to penetrate the cuticle layer of tomato fruits and leaves. Once inside, plant detoxification mechanisms mitigate its toxicity through enzymatic and non-enzymatic pathways.

The rapid decline in Confidor levels within the first three days after application is likely driven by the activation of the plant's physiological defense mechanisms. Upon detecting

Table 2. Effect of Confidor on biochemical quality indicators of tomato fruits when applied by foliar and insectigation methods ($p < 0.05$)

Method of application	Dry matter, %	Titrable acidity, %	Vitamin C, mg %	Sugars, %			SAR
				Monosugars	Sucrose	Total	
LSD	0.16	0.02	0.2	0.08	0.07	0.07	
Control	6.9 ± 0.09	0.28 ± 0.02	17.2 ± 0.18	2.64 ± 0.08	0.32 ± 0.03	2.96 ± 0.15	10.6
Spraying	7.2 ± 0.13	0.25 ± 0.01	18.0 ± 0.10	2.70 ± 0.02	0.42 ± 0.01	3.12 ± 0.02	12.5
Insectigation	7.2 ± 0.06	0.25 ± 0.01	18.5 ± 0.06	2.78 ± 0.02	0.44 ± 0.06	3.22 ± 0.05	12.9

Source: Authors' own elaboration

pesticide residues as xenobiotic compounds, plants initiate metabolic responses, including increased enzyme activity, stimulation of biochemical pathways, and enhanced metabolism. These responses strengthen the antioxidant system, creating a favorable environment for accelerated pesticide breakdown (Pang et al., 2020; Zhang et al., 2024). As pesticide concentrations decrease and toxicity subsides, these detoxification processes gradually slow down.

Research by Mohammed et al. (2017) indicates that imidacloprid is metabolized by cytochrome P450 enzymes into hydroxylated derivatives, facilitating its detoxification.

Surface pesticide residues dissipate more rapidly than internal residues due to various environmental factors, including evaporation, photolysis, volatilization, microbial degradation, and wash-off. Photolysis, for instance, involves the breakdown of pesticides by sunlight, primarily affecting surface residues exposed to light. Volatilization causes pesticides to evaporate from plant surfaces into the atmosphere, while microbial degradation – carried out by microorganisms on plant surfaces – further accelerates dissipation (Pang et al., 2020). Additionally, environmental factors such as rain or irrigation can wash away surface residues, reducing their concentrations more quickly than internal residues.

Insectigation

Conversely, when applied via drip irrigation, Confidor residues accumulate gradually in fruits and leaves, reflecting the systemic nature of root-based uptake. Within five days of application, residue levels in tomato fruits increase threefold compared to day one. This accumulation occurs due to the continuous absorption of imidacloprid from irrigation water, underscoring its systemic action. Unlike the rapid dissipation observed in foliar applications, root uptake results in sustained residue presence, reinforcing the prolonged effect of soil-mediated pesticide absorption.

These findings align with previous studies on the root application of Actara (active ingredient: thiamethoxam), which demonstrated that a portion of residues remains in the soil and is reabsorbed during subsequent irrigation cycles (Atchemyan et al., 2024). Similar trends have been observed with other systemic insecticides, including thiamethoxam, dimethoate, tetrachlorantraniliprole, and cyantraniliprole, which exhibit prolonged persistence and significant upward mobility within plants. This phenomenon, known as the “hysteresis effect,” has been widely documented, highlighting the distinct residue dynamics associated with root-applied pesticides in drip irrigation systems (He et al., 2018; Zheng et al., 2020; Li et al., 2022; Yan et al., 2023).

Impact on fruit quality

Whether applied by foliar spraying or through root uptake, Confidor treatment has been observed to alter tomato fruit acidity while increasing dry matter content, possibly due to metabolic shifts associated with sugar accumulation. Additionally, tomato plants exposed to Confidor exhibited increased vitamin C (L-ascorbic acid) levels.

However, these metabolic changes should not be solely interpreted as improvements in fruit quality, but rather as complex physiological responses to external stimuli, including pesticide exposure.

Ascorbic acid (vitamin C) plays a vital role in plant defense and human nutrition, acting as a major antioxidant that scavenges reactive oxygen species (ROS) and helps maintain cellular redox homeostasis (Mellidou et al., 2021; Wu et al., 2024). In plants exposed to pesticides, changes in ROS balance may directly influence ascorbic acid metabolism and the activation of antioxidant defense systems. In particular, low pesticide doses may induce a hormetic oxidative signal that stimulates antioxidant defenses, including the ascorbate–glutathione cycle, whereas higher doses can cause excessive ROS accumulation, oxidative damage, and metabolic disruption (Kumar et al., 2025).

While sugars are not as potent as dedicated antioxidants such as ascorbic acid, they play an important role in modulating plant oxidative-stress responses. Soluble sugars such as glucose, fructose, and sucrose contribute to ROS homeostasis under abiotic stress, both through antioxidant/ROS-scavenging roles and through broader metabolic and signaling functions (Afzal et al., 2021).

The increased sucrose content in tomato fruits affected by Confidor application could result from active biosynthesis within plant cells, triggered by stress-induced metabolic shifts.

Organic acids are vital for cellular metabolism, redox balance, and stress tolerance. They function as chelating agents and participate in metabolic reactions involving electron and proton transfer (Igamberdiev et al., 2018; Panchal et al., 2021).

Studies by Zhang et al. (2022) demonstrated that imidacloprid exposure in lettuce significantly altered amino acid metabolism and reduced organic acid levels, despite no visible phytotoxicity.

These findings suggest that, beyond their intended pest-control function, pesticides can influence ascorbic acid, sugar, and organic acid levels in plants. However, the effects vary depending on pesticide type, concentration, and plant species. At low doses, pesticides act as mild stressors, inducing ROS production, which in turn stimulates ascorbic acid and sugar biosynthesis as part of the plant’s defense response.

Therefore, a balanced approach to pesticide use – avoiding excessive application and strictly adhering to pre-harvest safety regulations – is essential to maintaining both agricultural productivity and food safety.

Conclusion

Confidor residue dissipation varies significantly between foliar spraying and root application (insectigation). Foliar spraying causes rapid surface residue loss but a slower decline in internal residues, whereas insectigation leads to a gradual and sustained buildup of residues in fruits and leaves, indicating strong systemic uptake and translocation within the plant.

Confidor application induces significant biochemical changes in tomato fruits, including reduced acidity, increased dry matter, higher sugar and vitamin C levels, and an improved sugar-acid ratio (SAR). Insectigation enhances sugar and vitamin C accumulation more effectively than foliar spraying, underscoring its potential for sustainable crop management.

The observed differences in residue dynamics emphasize the need to tailor Pre-Harvest Intervals (PHIs) based on the application method to ensure consumer safety and compliance with regulatory standards.

Optimizing application methods allows growers to enhance pest control efficacy, improve fruit quality, and reduce pesticide residue risks – key steps toward safer and more sustainable agriculture.

Author contributions

Atchemian L. contributed to the conceptualization and methodology of the study, Mirzoyan V. and Petrosian N. contributed to the laboratory analysis and to the preparation of the original draft of the manuscript. All authors have read and approved the final version of the manuscript.

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Conflict of interest and financial disclosure

The authors declare that the research was conducted in the absence of any commercial and financial relationships that could be construed as a potential conflict of interest.

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