Dissipation Kinetics of Pymetrozine in Tomato Field Ecosystem

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ABSTRACT

Background: The food safety is one of the top priorities in the protection of public health. Pesticide residue has environmental and human health impact so, studding the behavior and dissipation of pesticides on foods is necessary.

Objectives: This paper aimed to study the dissipation kinetics of pymetrozine to assess its effect in the ecosystem of tomato field, to expose residual dissipation behavior.

Materials and methods: In a tomato-field ecosystem, tomato samples were collected after pymetrozine application in different time intervals 0, 1, 3, 5, and 7 days. The dissipation kinetics and residual behavior of insecticide were analyzed based on a QuEChERS method and quantified by high-performance liquid chromatography combined with photodiode array detection (HPLC-DAD). **Results:** The average recovery from tomatoes was 90.75% (88.5 - 93%), and 85 - 87.5% from soil. The coefficient variation of the method (CV %) was less than 7% in tomatoes, and less than 12%. in soil, ranged from 1.27% to 4.77% for repeatability. The low detection limit (LOD) and limit of quantitation (LOQ) of pymetrozine were determined to be 0.01 and 0.03 mg/kg, respectively. The dissipation kinetics of pymetrozine residue was $C = 1.504\ e^{-0.528t}$ in tomato and $C= 11.82\ e^{-0.4082t}$ in soil, in addition, residues half-lives were 1.31, and 2 days, respectively. The pre-harvest interval (PHI) of pymetrozine on tomatoes was 3 days. The recommended dosage could be considered a safe dosage for humans.

Conclusion: This study would contribute to provide information on safe and proper use of the pesticide.

Keywords Dissipation, Pymetrozine, Tomato, QuEChERS, HPLC-DAD.

INTRODUCTION

Pymetrozine is a selectivity insecticide used for insects, such as aphids, whiteflies, and plant hoppers [1-4].It has a novel way by interfering in the neuro-regulation or nerve-muscle interaction and cause the death of insects after a few days through the feeding behavior [5-7].Moreover, pymetrozine has been considered by the US-EPA as a ‘‘likely’’ human carcinogen [8].Dissipation studies are necessary to evaluate the pesticide residue behavior and the degradation kinetics under the open field conditions to determine PHI is below the maximum residue limit (MRL). QuEChERS is a well-known method that was applied in the multi-residues analysis of pesticides in a variety of food[9], in tomato [10], in fruits and vegetables [11]. penconazole in tomato [12], propamocarb-hydrochloride in different matrices [12], QuEChERS method is easy and high throughput compared to conventional analysis methods [14,15]. QuEChERS was recently considered to be the official multi-residues AOAC method for the study of pesticide residues in fruits and vegetables [16].

Tomato is one of Egypt's most important and common vegetables. Approximately 526 feddan; representing 34% of Egypt's cultivation area were used for tomato cultivation. Tomato can be eaten fresh as a salad, or processed as a juice or paste[17]. Tomatoes at various growing stages are infested by several pests. A large number of pesticides are applied in the tomato field to control all these different diseases[18].

To our knowledge, few studies have examined the dissipation of pymetrozine in agricultural products; broccoli [19], rice field [20,21], red pepper [22], and lettuce [23],and there are no published studies about its dissipation on tomatoes. The main aim of this paper was to study the dissipation kinetics of pymetrozine to assess the pymetrozine effect in the ecosystem of tomato field, to expose residual dissipation behavior of pymetrozine. These findings may aid in the creation of regulations for the safe use of pymetrozine in tomato field pest management strategies.

MATERIALS and METHODS

1 - Chemicals and reagents

Analytical standard (purity > 98%), and the formulation (Chess 50% WG) were supplied by Syngenta (Cairo, Egypt), general structure of pymetrozine (figure 1).LC grade organic solvent(acetonitrile, methanol, and analysis grade acetic acid) was purchased from Merck Millipore Ltd. Milli-Q water purification system for ultrapure water. PSA, 40 µm was purchased from Supelco (Supelco, USA). Magnesium sulfate anhydrous, and Sodium chloride were purchased from Merck Millipore Ltd. An Agilent HPLC with a quaternary pump, column thermostat, and photodiode array detector was used.

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Carbon 18(250 × 4.6 mm, 5μm) was used as an analytical column for pymetrozine.

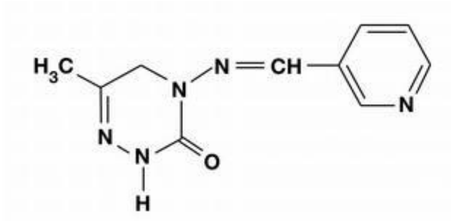

Figure 1. Pymetrozine structural formula

2 - Field experiment

At Syngenta Research Farm Station, El-Kalubia Governorate, Egypt, tomatoes were cultivated in an open field. In this experiment, a complete randomized block design was utilized. Tomatoes were planted in these plots using common agricultural practices. Chess (50% WG) was sprayed with a backpack sprayer motor at the recommended dose (240 g/Acres). Pymetrozine formulation was prepared according to the recommendations of the manufacturer. Untreated plots were left as a control.

3 - Sample preparation

Tomato samples

According to the FAO/WHO (1986) recommendations, tomato samples of approximately 1 kg each was collected from experimental plots in three replicates at different time intervals were 0, 1, 3, 5, and 7 days after application. The samples obtained were subsequently transported to the laboratory in an ice box. Fifteen gram of the homogenized blender sample was placed in a 50 ml polyethylene tube.

QuEChERS methodology was used for sample extraction and cleaned-up according to [24]. Fifteen ml of 1% acetic acid in acetonitrile was added into each tube, then shaking with a vortex mixer. Afterward, 4.0 and 1.0 g of anhydrous magnesium sulfate and sodium chloride respectively were added then centrifuged for 10 min at 5,000 rpm after vigorously shaking. In a clean 15 ml tube containing 100 mg and 500 mg of PSA and anhydrous magnesium sulphate respectively, 4 ml of the supernatant was transferred. To be ready for chromatographic analysis using HPLC, samples were centrifuged for 5min at 6,000 rpm after shaking and 2 ml of supernatant was concentrated to dryness.

Soil samples

Surface soil (0–15 cm depth) was sampled for analysis. The air-dried soil sample was crushed and passed through a sieve (40 – mesh). Ten g of soil was extracted with 150ml dichloromethane for 6h at 60 ºC by Soxhlet unit. The extract was evaporated to dryness by vacuum rotary evaporator at 40ºC.

Clean-up

A C18 SPE cartridge was used for clean-up, after the conditioning with 3 ml of methanol then 3 ml of water, the sample was resolved in the 0.5 ml and applied to the cartridge, then the cartridge was washed with 5 ml of the mixture of methanol/water (1:9 v/v). Pymetrozine residue was eluted from the cartridge with 5 ml of the solution of methanol/water (7:3 v/v). The eluent was centrifuged at 5000 rpm for 5 min, and then 4 ml of supernatant were evaporated to dryness, before the determination of pymetrozine.

4 - Chromatographic conditions

HPLC with photodiode array detector was used for residues analysis following the method described by Shen et al. (2009) [19] with the following modifications: acetonitrile: water (60/40 v/v) at a flow rate of 1.0 ml/min was the mobile phase. Pymetrozine residues were detected at a wave length of 240 nm. The retention time of pymetrozine was about 3.05 min. Estimation of pymetrozine residues was by comparing of peak area of the unknown or spiked samples with standards, under the same conditions. The Statistical Package for Social Sciences (SPSSWIN software) version 16.0 was used to do the statistical analyses.

RESULTS

Method performance

Method performance; Accuracy, precision and Recovery were determined for pymetrozine residue in tomato and soil samples (Table 1). Method selectivity was evaluated by analysis of blank tomato samples; there are no interfering peaks from endogenous matrix components.

Accuracy and precision

Inter-day and intra-accuracy and precision were determined by analyzing three sample replicates. Three QCs were analyzed in five different days to determination the intra-day. While three replicates of QCs were analyzed on the same day for inter-day estimation. Accuracy was estimated as the deviation percentage of the true value mean, and precision was expressed as the relative standard deviation (R.S.D. %) in the concentrations of QCs.

Recovery

Recovery assays were performed in the levels of 0.1, 1.0 and 5.0 mg/kg. Recovery was calculated by comparing the peak area of the analyte added to and extracted from the matrix with the peak area of the pure standard. The working solution of pymetrozine was prepared by diluting pymetrozine stock solution in acetonitrile, for the method of extraction from tomato. Untreated tomato samples were homogenized before begin spiked with pymetrozine. The average recoveries were 90.7% (88.5-93.0) and 86.2% (85 - 87.5), while precision was less than 7% and 12% for tomato and soil, respectively.

Table (1): Results of pymetrozine recovery (%), intra - and inter-day accuracy, and precision from spiked tomato and soil samples.

QC level	Nominal mg/kg	Mean Recovery		Accuracy (%)		Precision (R.S.D. %)	
		Tomato	Soil	Intra-day	Inter-day	Intra-day	Inter-Day
QC H	5	88.5	85.0	7.4	10.4	8.60	9.3
QC M	1	97	87.5	6.9	12.7	8.72	10.3
QC L	0.1	102	91.0	7.6	6.9	9.50	11.7

QC, quality control; H, high; M, medium; L, low; R.S.D., relative standard deviation.

DISCUSSION

Method validation was performed according the single laboratory validation approach and European commission guidelines and also method validation procedures for pesticide residues analysis **[25,26]**.

Kinetic study

The results explained that the dissipation kinetics and behavior of pymetrozine residues in tomato and soil were determined using first-order rate equation: $Ct = C_0 e^{-kt}$,Ct is the pesticide concentration at a certain time t, C_0 is the initial concentration and k is the rate constant/day. Pesticide persistence in the environment can be determined by pesticide half-life. Half-life ($t_{1/2}$) is a period of time that takes the pesticide to decay to half its initial concentration, which provides information on the pesticides dissipation in plant and the environmental sources. The half-life ($t_{1/2}$) was determined from the k value for each experiment, being $t_{1/2} = \ln 2/k$ **[10]**.

Pymetrozine residue in tomato

The estimated pymetrozine residues in tomato after application were 1.54, 0.86, 0.35, 0.11, and 0.03 mg/kg at 0, 1, 3, 5, and 7 days after application, respectively (Fig. 2). As expected, the dissipation of the pymetrozine residue in tomato fruits was observed as a function of time after application. The half-life value ($t_{½}$) for dissipation of pymetrozine on tomatoes was calculated and observed to be 1.31 days, at the recommended dosage. European Union MRL for pymetrozine in tomato was 0.5 mg/kg, while the FAO/WHO has not established MRLs for pymetrozine. The dynamics of pymetrozine residue could be described by the equation $C = 1.504\ e^{-0.528t}$, with $R^2 = 0.998$. There is no any pymetrozine residue detected in tomatoes after 18 days of application. The results showed that the pymetrozine was degraded relatively faster in the first three days of application. This fast dissipation of pymetrozine on tomato fruits could explain due to the nature of the tomato surface or the growth of the tomato fruit which may cause a dilution of the pymetrozine concentration.

Shen *et al*.[19] reported that the half-life of pymetrozine was 3.5, and 1.4 days in broccoli and soil, respectively. **Zhang *et al*. [21]** studied the dissipation of pymetrozine in the rice field and reported that the degradation of pymetrozine was faster in rice $t_{½}$ was ranged from 2.3-2.6 days, comparing to the soil $t_{½}$ was ranged from 5.1 to 5.7 days.

Another study by **Li *et al*. [20]**which also studied the dissipation of pymetrozine in the rice field and their results showed that the half-life was 0.89 and 7.0 days in rice and soil, respectively.

Moreover, the obtained results showed that the pymetrozine residue level on tomato fruits was lower than the MRL after 3 days of the recommended dose application. From these findings, we concluded that the faster degradation rate of pymetrozine on tomato could be due partially to the higher growth rate of tomato fruit relative to other crops.

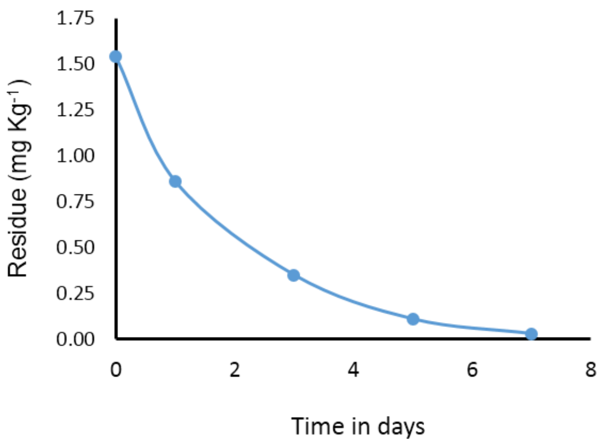

Figure 2. Dissipation kinetics of pymetrozine in tomato fruits.

Pymetrozine residue in the soil

Pymetrozine in the soil was presented in Fig. 3over the experimental period. The residues were 9.6, 7.4, 5.1, 2.53, 1.14, and 0.08 mg/kg at 0, 1, 3, 5, 7, and 14days after application, respectively. Dissipation kinetics of pymetrozine residues could be described by the equation $C = 11.82\ e^{-0.3465t}$, $R^2 = 0.9897$. Pymetrozine half-life in soil was 2 days.

The initial deposit of pymetrozine residue was higher in soil than tomatoes after the application, which demonstrates that pymetrozine residue exists in the surface layer of the soil. While the degradation rate of pymetrozine was slower in soil than in tomatoes. After 20 days of the recommended dose application of pymetrozine, the pymetrozine residues could not be detected.

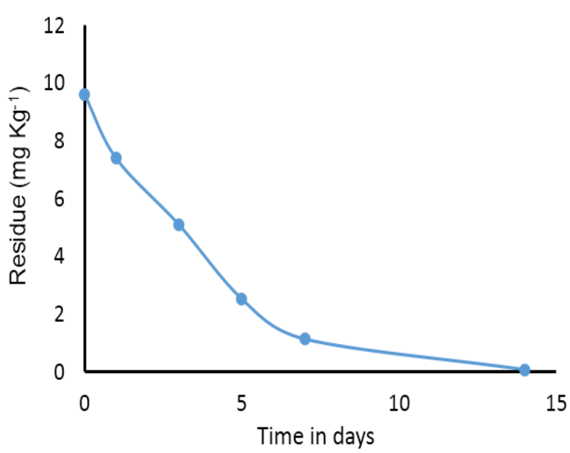

Figure 3. Dissipation kinetics of pymetrozine in soil.

CONCLUSION

Good analytical performance was achieved through using the QuEChERS method, sensitivity (LODs, 0.01mg/kg; LOQs, 0.03 mg/kg), and recovery rates ranged from 88.5-93% and 85-87.5% for tomato and soil, respectively. Thus, this method can be used to determine a very low concentration of pymetrozine residues. Pymetrozine has shown fast dissipation rates: $t_{1/2}$were 1.31 days, and 2 days for tomato and soil, respectively. The residues following application of the recommended dosage were lower than 0.5 mg/kg. Therefore, 3days interval between application and harvest was thought to be safe. This study would contribute to provide information on the safe and proper use of the pesticide.

REFERENCES

1. **Kristinsson H(1990):**US Patent No. 4,931,439.*Ciba-Geigy Corp.www:patentimages.storage.googleapis.com/91/30/fe/5e6878c80cf297/US4931439.pdf.*
2. **Allemann DV, Robinson SW and Flüeckiger CR(1995):**Pymetrozine interferes with transmission of Tomato yellow leaf.In *1st World Cotton Research Conf.*https://link.springer.com/article/10.1007/BF02979742 **3.**
3. **APHA (2005):**National Coordinator for Health Information Technology. *136th Annual Meeting, San Diego*.https://www.medscape.com/viewcollection/17562
4. **Nicholson W, Senn R, Flüeckiger C, Fuog D (1995):** *Pymetrozine: a novel compound for control of whiteflies*. 635–639. (Taxonomy, Biology, Damage, Control and Management, Intercept Ltd.
5. **ASTM (1992):**Standard Practice for Conducting Bioconcentration Tests with Fishes and Bivalve Molluscs. *ASTM Committee E-47.01, p.652.en.wikipedia.org › wiki › ASTM_International*
6. **Kayser H, Kaufmann L and Schurmann F(1994):** In *Proc. 1994 Brighton Crop Protection Conf.* Vol. 2 737–742 (Pests and Diseases).https://books.google.com.eg/books/about/Brighton_Crop_Protection_Conference.html?id=iOtJAAAAYAAJ&redir_esc=y
7. **Lawson D, Dunbar D, White S, Ngo N(2000):***In: Proc. of National Cotton Conference* Vol. 2 1106 (Memphis).http://www.cotton.org/beltwide/proceedings/getPDF.cfm?year=2000&paper=557.pdf
8. **EPA (2000):**<http://www.epa.gov/opprd001/factsheets/pymetrozine.pdf>.
9. **Hong J, Lee C, Lim J, Lee K(2011):**Comparison of analytical methods and residue patterns of pymetrozine in Aster scaber. *Bulletin of environmental contamination and toxicology*,**87**: 649-652, doi:10.1007/s00128-011-0407- 8.
10. **Abd-Alrahman S, Monir M, and Nevin S (2011):**Dissipation of Fungicides, Insecticides, and Acaricide in Tomato Using HPLC-DAD and QuEChERS

Methodology. *Food Analytical Methods,* DOI 10.1007/s12161-011-9279-0.

11. **Lehotay S, de Kok A, Hiemstra M ,Van Bodegraven P (2005):** Validation of a fast and easy method for the determination of residues from 229 pesticides in fruits and vegetables using gas and liquid chromatography and mass spectrometric detection. *Journal of AOAC International,* **88**: 595 - 614.
12. **Abd-Alrahman S, and Ahmed N (2012):**Dissipation of penconazole in tomatoes and soil. *Bulletin of environmental contamination and toxicology,***89**: 873 - 876, doi:10.1007/s00128-012-0776-7.
13. **Abd-Alrahman S, and Almaz M (2012):** Degradation of propamocarb-hydrochloride in tomatoes, potatoes and cucumber using HPLC-DAD and QuEChERS methodology. *Bulletin of environmental contamination and toxicology,* **89**: 302 - 305, doi:10.1007/s00128-012-0703-y.
14. **AOAC (2000):**Official Methods of Analysis 17^{th}ed. *AOAC INTERNATIONAL*, 22, https://www.aoac.org/official-methods-of-analysis.
15. **Luke M, Froberg J and Masumoto H (1975):** Extraction and Cleanup of Organochlorine and determination by Gas-Liquid Chromatography. *J. Assoc. Off. Anal. Chem.,* **58**: 1020.
16. **Lehotay S (2007):** Determination of pesticide residues in foods by acetonitrile extraction and partitioning with magnesium sulfate: collaborative study. *Journal of AOAC Internationa, l***90:** 485 - 520.
17. **EL- Nabarawy I, Abou-Donia M and Amra H (1992):** Determination of profenofos and malathion residues in fresh tomatoes and paste. *Egyptian Journal of Applied Science,* **7:** 106 - 111.
18. **APC (2010):** Pesticides registration, *Agricultural Pesticide Committee*, www: apc.gove.eg.
19. **Shen G, Hu X and Hu Y (2009):** Kinetic study of the degradation of the insecticide pymetrozine in a vegetable-field ecosystem. *Journal of hazardous materials,* **164:** 497 - 501, doi:S0304-3894(08)01227-2[pii]10.1016/j.jhazmat.2008.08.020.
20. **Li C, Yang T, Huangfu W and Wu Y (2010):** Residues and dynamics of pymetrozine in rice field ecosystem. *Chemosphere*, doi:S0045-6535(10)01196-3 [pii]10.1016/j.Chemosphere.2010.10.053.
21. **Zhang Y, Zhang L, Xu P, Li J and Wang H (2015):**Dissipation and residue of pymetrozine in rice field ecosystem. doi:10.1007/s10661-014-4256-x.
22. **Jang J, Rahman M, Ko A, Abd El-Aty M, Park H, Cho K and Shim H (2014):** A matrix sensitive gas chromatography method for the analysis of pymetrozine in red pepper: application to dissipation pattern and PHRL. *Food Chem***., 146:** 448 -454.
23. **Cabizza M, Satta M, Falconi S, Onano M (2007):** Degradation of cyprodinil, fludioxonil, cyfluthrin and pymetrozine on lettuce after differentapplication methods. *J Environ Sci Health B***, 42:** 761 - 766.
24. **Abd-Alrahman S (2014):** Residue and dissipation kinetics of thiamethoxam in a vegetable-field ecosystem using QuEChERS methodology combined with HPLC-DAD. *Food Chem.,* **159:** 1 - 4.
25. **SANTE/11945/(2015):**Guidance document on analytical quality control and method validation procedures for pesticides residues analysis in food and feed. *(Implemented by 01/01/2016)*. European Commission Directorate General for Health and Food Safety, https://ec.europa.eu/food/sites.
26. **Thompson M, Stephen L, and Wood R (2002):**Harmonized Guidelines For Single Laboratory Validation of Methods OF Analysis. *Pure Appl. Chem., 74 (5): 835–855.*