







Article

Impact of Thiamethoxam in Papaya Cultivation (*Carica papaya* Linnaeus) in Rotation with Watermelon (*Citrullus lanatus*) Crops

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Abstract: Thiamethoxam is a neonicotinoid with systemic and contact action, used in Mexico for the care of different traditional fruit crops, mainly in the cultivation of papaya. Soils of agroecosystems with papaya in the Gulf of Mexico area, the main producers of this fruit, are currently characterized as 45% of the producers organize papaya cultivation in rotation with watermelon at different sowing dates. The objective of this study was to determine the presence and concentration of thiamethoxam in soil and water during the rotation of papaya crop with watermelon culture in the central zone of Veracruz, Mexico. An analysis was carried out to know the management of thiamethoxam in different types of soil and in the region; and in an experimental plot. Soil and water samples were taken at different stages during the rotation of both crops. Those samples were taken systemically, starting with the watermelon culture cycle and then, during the phenological stages in the papaya crop cycle. Soil and water samples were analyzed using HPLC-UV equipment for its determination. The design was random blocks with six repetitions and the software used for data analysis was the Statistica 2007 program. Thiamethoxam was concentrated in amounts of ≥ 0.40 mg/L in 79% of the samples in water and ≥ 0.55 mg/kg in 75% of the samples in soil. The highest values of thiamethoxam in soil were in the stage of watermelon culture with 0.4 mg/kg and in the soil preparation of the papaya crop with concentrations of 0.8 mg/kg. Whereas irrigation water from the watermelon cultivation and the soil preparation for the papaya showed concentrations of 0.5 and 0.7 mg/L, respectively. The presence of thiamethoxam was identified in 100% of the samples analyzed in the stage of preparation of soil and water of the cultivation area, concluding a possible risk of residuality of thiamethoxam in fruits that exceed the maximum limits of tolerance established by the EPA, EFSA and FAO.

Keywords: neonicotinoids; pesticides; agroecosystem

1. Introduction

Agroecology is fundamental for the study of sustainable agroecosystems, uses ecological principles and concepts as tools to understand land degradation processes, such as erosion, desertification, salinization, compaction and the decrease of fertility, plus the impact of pesticides in agriculture [1]. Carrying out agricultural cultivation practices to help preserve adequate physical and chemical characteristics of soil and water, while taking into account the eminent growth of human population that demands safe food, goods and services, becomes essential [2].

Agricultural activities require the reduction of a massive use of pesticides and anthropogenic activities that contribute to pollute ground and surface waters [3,4]. This phenomenon limits and prevents the enjoyment of the benefits of quality water, causing negative impacts on aquatic organisms and destruction of soil macrofauna [5]. The consumption of agricultural foods contaminated with neonicotinoid pesticides can be a public health risk due to the residual compounds they generate [5].

The use of pesticides in agriculture began its activity in 1940, since then more than 70,000 chemical substances have been generated for crop management. The sales volume of pesticides is around 2800 million kilograms of active ingredients a year and more than 50 thousand commercial formulations [6]. Thiamethoxam is recommended for 116 agricultural species and is present in 64 countries worldwide [7]. Thiamethoxam was recorded in 2004 and is used to control whitefly (*Bemisia tabaci*) in tomatoes, chili peppers and fruit trees such as papaya [8]. It is also used for whitefly control in the USA., The European Union, Brazil and India. Due to its excessive use, insect pests have become resistant to applications of neonicotinoid insecticides, which is the case of Imidacloprid. For this reason, it has been considered necessary to use of new chemical molecules such as thiamethoxam. This molecule acts in a systemic and contact form, being used for the control of aphids, thrips, mites and soil pests [9]. In addition, there have been positive results on the control of some pests such as *Mahanarva fimbriolata* in perennial crops [10,11]. It is also recommended, in Venezuela, for the control of *Togodes orizicolus* Muir in rice cultivation, with doses of 75 and 100 g ha⁻¹ [12].

Thiamethoxam is also used for the treatment of seeds, recommending doses of 200 to 300 mL per 100 kg, improving its vigor and germination rate [13]. The contamination of surface and underground water can be generated through runoff processes, accumulating in bodies of water large quantities of pesticides such as thiamethoxam. There are studies of thiamethoxam discharges to natural currents or to the ground that is generating focalized contamination [5]. This pesticide has high mobility in the C horizon of soils; in the stream El Cardillo in Argentina, there is a potential risk of leaching pesticides in groundwater by the increase of its net recharge to a value of 2.3 mm day⁻¹, since the presence and residuality of thiamethoxam in soil depends on the frequency of its use in vegetables such as tomato, parsley, cauliflower, bulb onion, and broccoli [14].

The management of tropical agroecosystems with the use of neonicotinoid insecticides can generate a risk of ecological damage such as the death of aquatic organisms, birds and mammals, and hence to public health [9]. The cultivation of the papaya fruit tree, which due to its organoleptic characteristics has acceptance in the national and international market, presents numerous phytosanitary problems, such as the presence of virosis, phytoplasmas, mites and aphids, for which the producers use pesticides as neonicotinoids massively for its control. The instability of the market allows the papaya crop to be associated with vegetables such as watermelon, with the purpose of obtaining additional income during a year. For the aforementioned, this research was conducted with the purpose of knowing the presence and residuality of thiamethoxam in soil and irrigation water as a result of the environmental impact generated by the management and application of thiamethoxam in the control of pests in papaya (*Carica papaya*) crops in rotation with watermelon (*Citrullus lanatus*) in the municipality of Cotaxtla, Veracruz, located in the center of the country in the Gulf of Mexico, where 45% of the producers organize their papaya cultivation in rotation with watermelon crops in different sowing dates [9,15–17].

2. Materials and Methods

The study was developed in the municipality of Cotaxtla, Veracruz, in an experimental plot of papaya Maradol-watermelon (Figure 1), located at the coordinates of 18°53'51" north latitude and 96°22'37" west latitude.



Figure 1. Location of the papaya producing area in the town of Cotaxtla, Veracruz Mexico.

In the study area, papaya producers use thiamethoxam during a papaya crop cycle, with a total volume of 500 mL ha^{-1} for the control of whitefly, thrips and aphids. In this study area it is reported that thiamethoxam insecticide has been commonly used in the papaya crop over the last five years.

Method

To know the types of soils in the area where the insecticide was applied, a diagnostic study was carried out based on the local knowledge of the farmer called the “classification of peasant lands” [18]. An experimental plot of papaya cultivation rotated with watermelon, with a population density of 2778 plants/ha, was established in the study area. Samples were taken from the experimental plot during four phenological stages in the year to know the presence and concentrations of thiamethoxam. The phenological stages in the papaya-watermelon crop, correspond to the following study treatments, which are mentioned below as T1: relay cultivation (watermelon); T2: soil preparation for papaya; T3: stage of papaya transplant; T4: papaya in production. Six repetitions were established for each treatment and five samples were obtained from each repetition, of which a composite sample was taken for analysis, the block corresponded to the phenological stage of the agroecosystem with papaya.

The soil sampling technique was as established by the Official Mexican Standard NOM-AA-105-1988 and the International Standard ISO/TC 190 (UNE-EN ISO 15175:2018-Soil Quality). The sampling consists of using the zig-zag method, with strains of 20 by 20 and 25 cm deep, collecting a sample of 2 kg for the determination. For the sampling in irrigation water, the Mexican Standard NMX-AA-003-1980, the Official Mexican Standard NOM-AA-104-1988, and the International Standard ISO 5667-10-1992 [19] were used. The total of composite samples analyzed were 48, of which 24 corresponded for irrigation water and 24 for soil, these were taken in the field using also the zig-zag sampling method according to national and international regulations. In the process of growing papaya in rotation with watermelon, the samples composed of soil and irrigation water were collected 100 days after sowing watermelon crop; the preparation of the land continued and after 200 days the sampling was carried out again. Later in the papaya transplant, the samples were taken at 300 days; and finally, after completing 450 days that corresponded to the production of papaya, the last sampling was made. The soil and water samples were from April, 2016 to March, 2017. The samples collected were transferred for analysis to the laboratory of the Instituto Tecnológico de Boca del Río, to the laboratory of Investigación de Recursos Agropecuarios (LIRA).

The HPLC-UV is integrated of a quaternary pump, autosampler of 200 vials, with syringe of $250 \mu\text{L}$, a fluorescence and visible UV detector, contains four solvents and a separation column of 10 cm by 4.6 of diameter of 5μ and loop of $20 \mu\text{L}$ sample with a capture system. The instrumental conditions of the chromatographic column is C18 RP $5 \mu\text{m}$, with a temperature of $25 \text{ }^\circ\text{C}$, a mobile

phase of 45/55 (AC/AG), with a flow of 1 mL min^{-1} and $\lambda = 254 \text{ nm}$, the working range was 10 to $100 \mu\text{g}$ of thiamethoxam L^{-1} , with a linearity of $R \geq 0.9$ and a recovery of $\geq 90\%$.

To determine thiamethoxam in soil the following procedure was carried out, in which samples were macerated, then 10 mL of Acetonitrile (CH_3CN) was added, allowing it to sit for 5 min, then it was shaken vigorously for 1 min using a vortex mixer. Afterwards, 3 mL of HPLC water, 6 mL of hexane (C_6H_{14}), 4 g of Magnesium Sulphate (MgSO_4), 1 g of N-acetylcysteine (NAC), and 500 mg of sodium citrate ($\text{Na}_3\text{C}_6\text{H}_5\text{O}_7$) were added. The sample was subjected to manual shaking for 20 min, and vortexed for 1 min, then centrifuged for 10 min. Later, 6 mL of excess was taken and deposited in tube II with 90 mg of magnesium sulfate, 150 mg of Primary and Secondary Amina (PSA), 150 mg of Octadecyl Carbon Chain (C18). The sample was vortexed then for 1 min and centrifuged for 10 min. Next, 3 mL of the excess was taken and dried at 40°C with N_2 , afterwards $200 \mu\text{L}$ of methanol (CH_3OH) was added, measuring $100 \mu\text{L} + 900 \mu\text{L}$ of mobile phase and then filtered with $0.22 \mu\text{L}$ disks, finally it was recovered in vials. Samples that had concentrations greater than 0.1 mg/kg or mg/L presented eluted peaks of thiamethoxam. Method validation tests were performed on each batch, with a fortified control at the same conditions of the samples analyzed. The recovery tests for water were 87% while for soil were 92%.

The statistical design for soil and irrigation water was randomized complete blocks with six repetitions. The statistical software was with the program statistica 2007, with parametric statistical analysis (ANOVA) and non-parametric Kruskal-Wallis, and graphs categorization with pie charts.

3. Results

The values of thiamethoxam concentrations in soil of the experimental plot that were found at a depth of 30 cm in the area of the macrofauna and microflora were $\geq 0.55 \text{ mg/kg}$, these values are higher than those found in the irrigation water of $\leq 0.45 \text{ mg/L}$ (Figure 2). Thiamethoxam was present in 79% of the analyzed samples of irrigation water during the phase of papaya and watermelon cultivation. Whereas in soil, 75% of the total samples analyzed showed average concentrations of thiamethoxam of 0.57 mg/kg , this response is due to its use and management when applied directly to the soil and in the irrigation water system. Irrigation water showed statistically significant differences ($p \leq 0.05$) in the total samples analyzed with respect to soil samples of the papaya crop in rotation with watermelon (Figure 3).

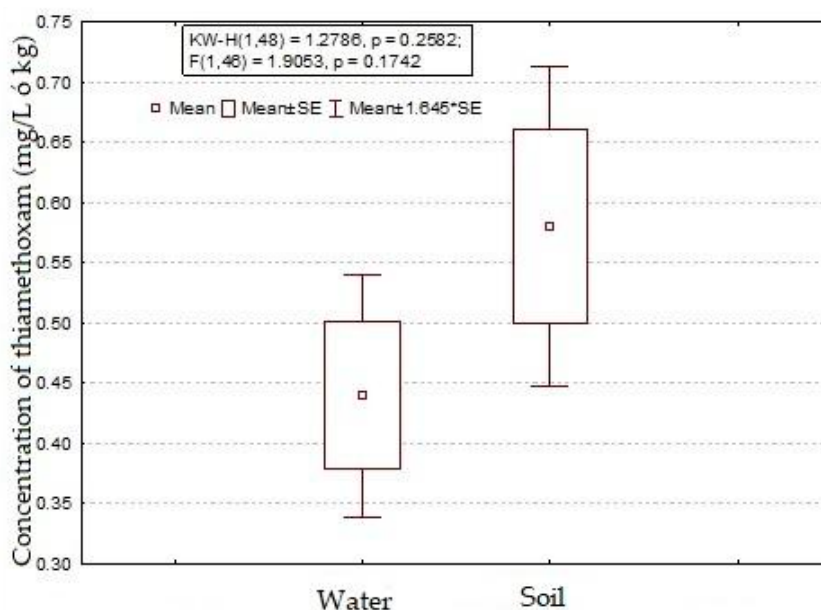


Figure 2. Concentration of thiamethoxam in irrigation water (mg/L) and soil (mg/kg) in a papaya crop rotation with watermelon in the area of Cotaxtla, Veracruz.

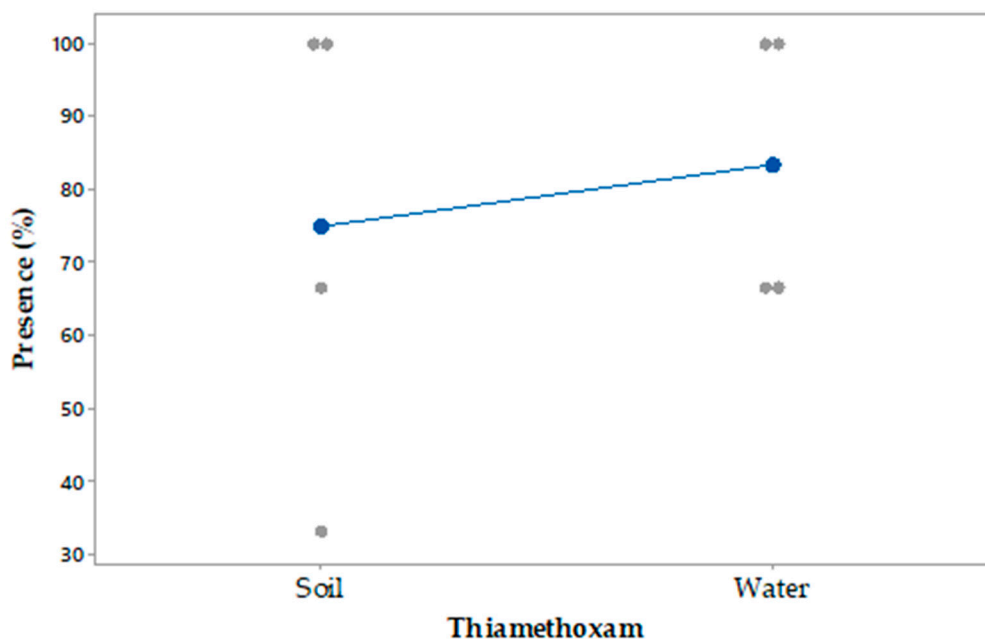


Figure 3. Presence of thiamethoxam in soil and irrigation water of the papaya crop in rotation with watermelon, where water presented significant statistical differences ($p \leq 0.05$) in the area of Cotaxtla, Veracruz.

The highest residues of thiamethoxam in different stages of cultivation of papaya in rotation with watermelon were found in the soil. These corresponded to soil preparation (T2) and papaya transplanted (T3) with values higher than 0.8 mg/kg (Figure 4). The lowest concentrations were present in the papaya fruiting stage, which is when the papaya crop demands more nutrients for its development and fruit production; the concentration values of the insecticide were less than 0.2 mg/kg. Values higher than 0.7 mg/L of thiamethoxam were found in the samples of irrigation water in the stage of the soil preparation, which is when the soil of the papaya crop is without any vegetal cover, which allows thiamethoxam to be retained by clay particles. Because of the above, it is important to mention that when comparing the relay crop that corresponds to watermelon with the papaya crop in fruit production, the greater presence of thiamethoxam in irrigation water and soil corresponded to the watermelon with values higher than 0.5 mg/L in irrigation water and 0.4 mg/kg in soil (Figures 4 and 5). In the soil preparation stage, thiamethoxam concentrations are retained by soil particles. Some particles of thiamethoxam are mobilized into the shallow waters when there are no established crops that extract the thiamethoxam particles through the roots or stomata of the plants.

Samples obtained from different types of soil and irrigation water in watermelon cultivation showed the presence of thiamethoxam in 100% of the irrigation water. In the opposite case, at the beginning of the papaya crop in the stage of seedling production, it was found that 100% of the samples had thiamethoxam in soil and irrigation water. Also, the crop in production showed thiamethoxam particles in 33% of the samples analyzed in soil and 50% in irrigation water. This can be explained, as the rest of thiamethoxam particles were mobilized towards the plant and papaya fruits via symplastic and apoplastic (Figures 6 and 7). In addition, this agrees with the results of the diagnosis made to papaya producers in the municipality of Cotaxtla, Veracruz, they apply the highest volume of thiamethoxam to papaya cultivation mainly in free soils, while in sandy soils there is no application. Therefore, significant differences ($p < 0.05$) were found with the volume of thiamethoxam used in clay soils when compared with the volume of thiamethoxam used in sandy and loamy soils (Figure 8).

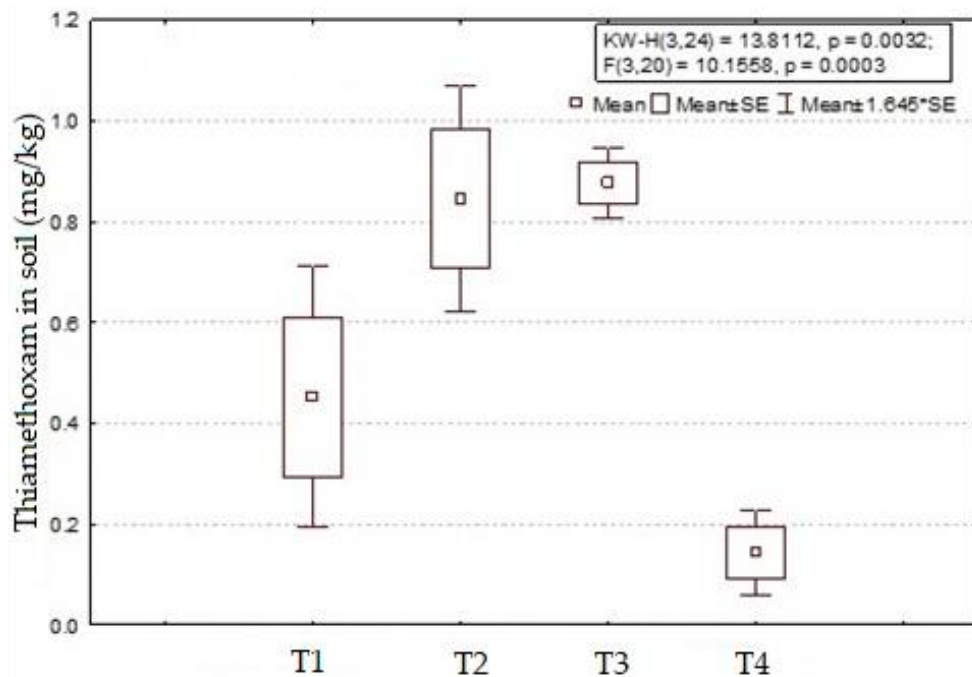


Figure 4. Concentration of thiamethoxam in soil (mg/kg) in different stages of the papaya crop rotation with watermelon. Abbreviations: T1 = Watermelon cultivation; T2 = Soil preparation for papaya; T3 = Stage of papaya transplant; T4 = Papaya in production.

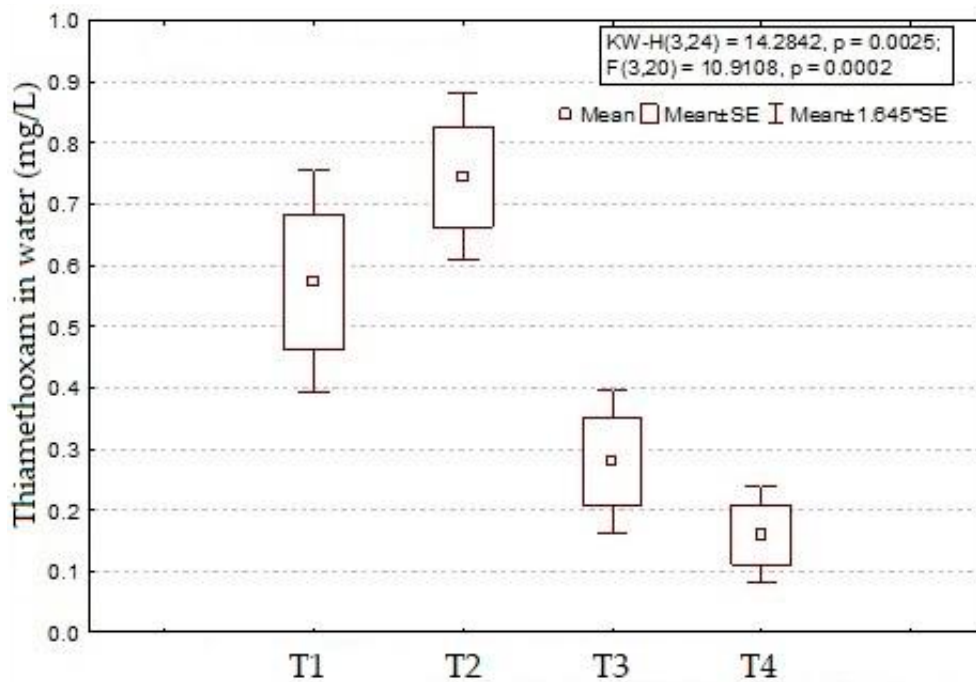


Figure 5. Concentration of thiamethoxam in irrigation water (mg/L). Abbreviations: T1 = Watermelon cultivation; T2 = Soil preparation for papaya; T3 = Stage of papaya transplant; T4 = Papaya in production.

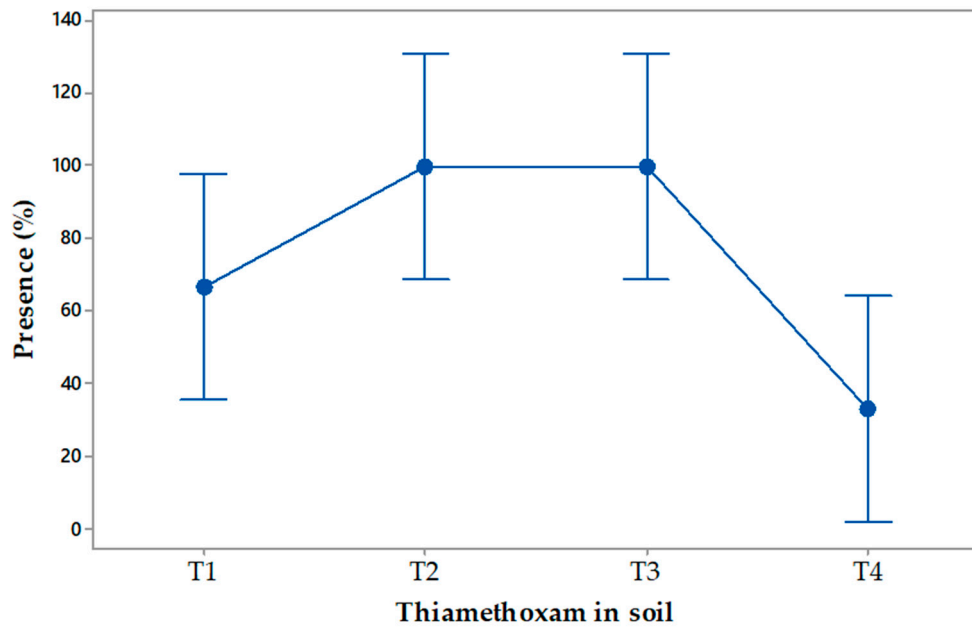


Figure 6. Presence percentage of thiamethoxam in soil in the papaya crop rotation with watermelon in an experimental plot. T4 presented significant statistical differences ($p < 0.05$) with respect to T2 and T3, these differences were not evident with T1.

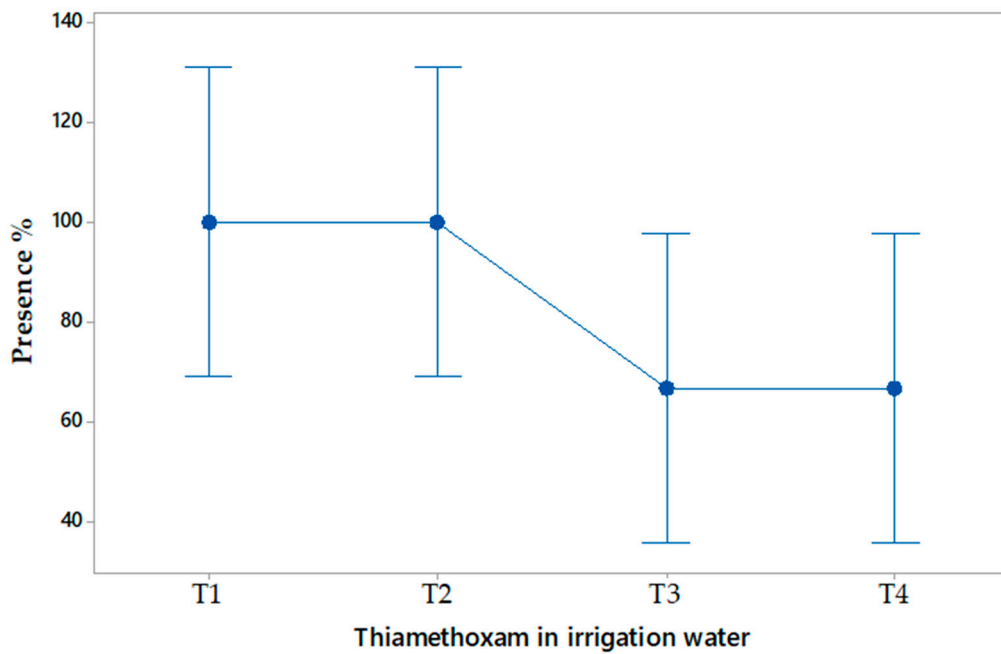


Figure 7. Presence percentage of thiamethoxam in irrigation water in the papaya crop rotation with watermelon in an experimental plot. There were no significant statistical differences ($p > 0.05$) in all the treatments analyzed.

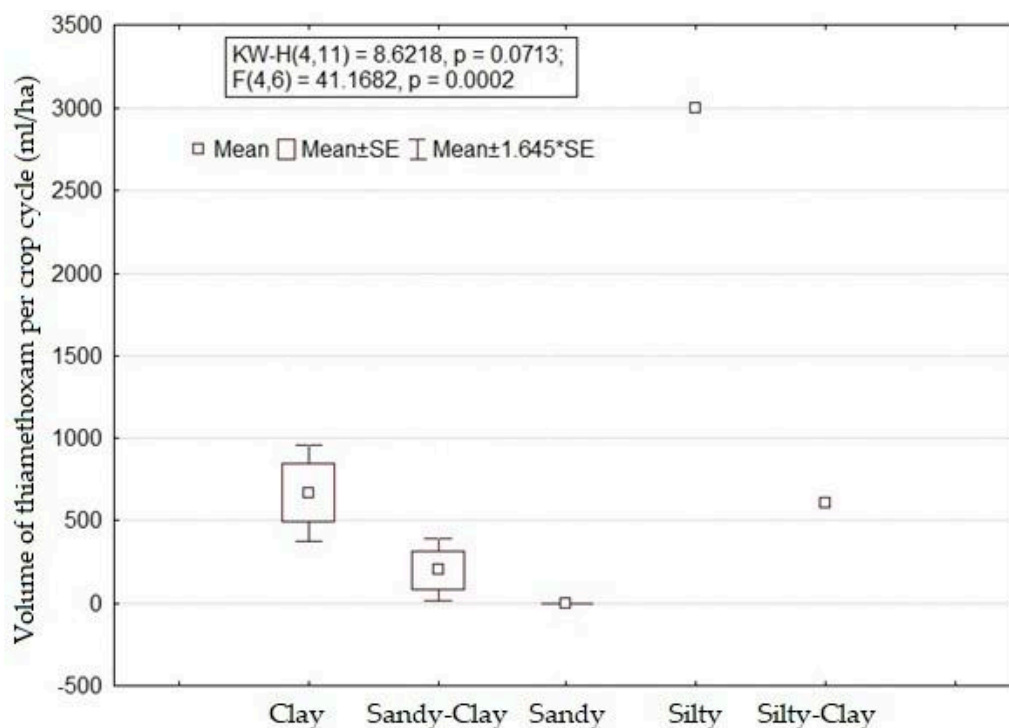


Figure 8. Volume of thiamethoxam per crop cycle managed in different types of soil in the municipality of Cotaxtla, Veracruz.

4. Discussion

In the main papaya producing areas in Mexico, 27% of the producers have orchards under a monoculture scheme and 45% of them associate papaya orchards with different vegetable species, which allows for a greater diversity of pests and diseases. Under the risk of causing economic and ecological damage, producers use thiamethoxam in doses of 3 L ha⁻¹ to control agricultural pests and a minimum dose of 300 L ha⁻¹ in the papaya cultivation cycle. It is estimated that the amount of thiamethoxam used during the papaya crop cycle in the main papaya producing area in Cotaxtla, Veracruz, is 220.5 kg/year [19], comparing these results with the valley of Culiacán Sinaloa, Mexico, the total volume of thiamethoxam applied there is 692.26 kg/year and for the group of neonicotinoids the amount is 1682.25 kg/year of active ingredient [20].

One of the characteristics of thiamethoxam is its mobility capacity, which is why it is found in wells and deep wells that are used for domestic and agricultural use. It is a similar case in Argentina, where there is a presence of thiamethoxam in the C horizon soil (mineral layers that have been affected by pedogenetic processes in a minimal way) with high leaching capacity to groundwater [14]. In addition, the presence of thiamethoxam in water and soil (Figure 6) reinforce the theory of chemical contaminants migration, where the highest concentrations of pesticides are present at lower depths of soil, mainly in shallow waters [3,21]. On the other hand in soils of loamy sandy texture, with guava plantations in Venezuela, good aeration, medium permeability and little retention of water and nutrients was observed, which allowed for thiamethoxam absorption isotherms with values of 2.95 ± 0.83 mg/kg. This phenomenon depends on the characteristics of the soils, such as the clay content, because thiamethoxam has the capacity to protonate, due to its size and polarity. The percolation of thiamethoxam to groundwater depends on the texture of the soil. In soils with high sand content thiamethoxam tends to percolate, considering that thiamethoxam in soil degrades slowly in aerobic conditions, but in concentrations of 4100 mg L⁻¹ at temperatures of 20 °C is highly soluble in water, according to the above there is a risk that thiamethoxam particles may infiltrate into groundwater [18,22]. Studies show that concentrations of 10 µg/L in water cause death of aquatic species, and in turn, 10 µg/kg of thiamethoxam in topsoil damages its macrofauna and microflora.

In addition, if we consider that soil degradation (LD_{50}) is moderately persistent with 39 days in the field, it is important to mention that some studies show that they can persist in soil for up to three years [18,23]. Thiamethoxam has been found with drinking water values of 0.06, 0.2 and 1.0 mg/L, this is the result of applications made in crops; furthermore, thiamethoxam is considered an effective contaminant when applied to the foliage of plants. Another feature of thiamethoxam, is its ability to evaporate in less than 30 s when it reaches a drop diameter of 337 μm . This is an important factor regarding the relative humidity, when there are values of 60%, as this favors the evaporation of thiamethoxam to the environment [24,25]. Thiamethoxam is absorbed by the plant through the roots, stems and leaves, that the largest thiamethoxam particles are concentrated in the stage of fruit production. When the management of thiamethoxam is towards the canopy of the trees, these particles can enter through stomata and lenticels and translocate to the different organs of the plants [22]. The fruits of these crops have to be analyzed to know the residuality of thiamethoxam and to define whether they meet the tolerance levels required for their consumption. According to the above, the Environmental Protection Agency (EPA) recommends that studies be conducted to address the safety of food [26]. For the cultivation of papaya, the EPA (2012) established the tolerance levels with concentrations of 0.4 mg/kg of thiamethoxam, and 0.05 mg/kg for the European Union (EFSA, 2012) [27]; while the FAO establishes maximum residual limits for thiamethoxam at 0.01 mg/kg.

In the phase of cultivation of papaya in soil, the concentrations of thiamethoxam show its residuality in the stage of fruit production. These results are related to studies reported in Ecuador where irrigation water contaminated surface water with thiamethoxam, the following values were found there: 0.32 $\mu\text{g/L}$ in Río Chico, 0.17 $\mu\text{g/L}$ in Río Portoviejo, 0.48 $\mu\text{g/L}$ in Río El Ceibal; while in drinking water the concentration was 0.27 $\mu\text{g/L}$ [28]. Based on this, it is necessary to transfer technical knowledge to producers, with the purpose of raising awareness about the importance of the quality of the soil where the papaya is grown, mainly the type of texture, pH and content of organic matter. This will allow them to raise strategies that will improve the production and not depend on agrochemicals due to the low quality of their soils. New technologies propose the production of fruit trees and annual crops, using sustainable technologies [29]. Currently, an alternative is the use of nanoparticles, to reduce agrochemicals in agricultural crops. It is noteworthy that companies like Bayer and Syngenta are conducting research in this area of nanotechnology, aimed at finding a lower dose of active ingredient, lower residuality and with it, a lower load of contaminants in the environment. With these characteristics it is possible to generate fertilizers, growth regulators and more powerful and efficient pesticides for the control of pests and weeds, which respond to extreme weather conditions, probably leading to the disadvantage of generating contamination to soils and water bodies [30]. Organic agriculture such as the use of insecticides of vegetable origin could be an alternative for the control of agricultural pests, it is also important to consider the use of low doses of chemicals, or insecticides of biological origin, as an alternative for the control of specific pests [18,31]. Such is the case for Mexico where the optimum dose of thiamethoxam of 0.3 kg/ha was evaluated for papaya cultivation, spraying every 15 days for the control of pests such as *Diabrotica* sp., *Laspeyresia siapomonella*, *Frankliniella occidentalis*, *Myzus persicae*, *Bemisia tabaci*, *Nezara viridula*, *Anthocoris nemoralis*, *Planococcus* sp., *Anastrepha ludens* Loew and *Toxotrypana curvicauda* [27].

5. Conclusions

It is necessary to establish planning criteria in the management and use of thiamethoxam in papaya agroecosystems in order to reduce its contamination. The training and transfer of new friendly technologies to the environment are necessary to achieve the implementation of good management practices during the use of chemical compounds and with this, to reduce impacts and conserve soil and water characteristics in the ecosystem. A rational handling of pesticides should include a relationship between the dosage of thiamethoxam and soil type in the growing area, which results in the reduction of the impact on the environment. Crop rotation will have the capacity to achieve the recovery of the quality of the soils and reduce damages caused by runoff to the groundwater layer. With the

implementation of these mitigation strategies in the use of agrochemicals will also decrease the dose of its application and harm to the agroecosystem.

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