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Research Article

Translocation of emamectin benzoate residues in the trunk, leaves, and fruits of date palm, *Phoenix dactylifera*

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Abstract

The red palm weevil, *Rhynchophorus ferrugineus* (Olivier), severely attacks date palm trees and has become a global problem. Due to extreme infestation of *R. ferrugineus*, date farmers use chemical insecticides, which are toxic to humans and animals if residues are present in fruits. In this study, we assessed residues of emamectin benzoate (Aretor®), a biorational insecticide, in the date palm, trunk, leaves, and fruits at different times (0, 3, 9, and 15 months) following trunk injection. Emamectin benzoate was injected into the palm trees using the Syngenta Tree Micro-Injection Devic. Each date palm was treated with 48 ml of insecticide solution, injected in four directions (i.e., 12 ml per direction). The concentrations of emamectin benzoate were monitored at 0, 3, 9, and 15 months for trunk and leaves samples, while fruit samples were taken at 1, 1.5, 2.5, and 3 months after treatment. The concentration of emamectin residues in treated palm trunks was 55.1 ppb immediately after treatment and 213.6 ppb after 15 months. Results indicated that the persistence of emamectin residues in the date palm trunk up to 15 months after injection can be utilized as preventive and curative endotherapy to protect date palm trees from red palm weevil infestation. Throughout the entire analysis period, the emamectin residues were not detected in the leaf or fruit samples. This research indicates that emamectin benzoate is relatively safe for humans and animals and can be a good option for red palm weevil management in date palm orchards.

Keywords: Pesticide, toxicity, monocot, stem, Saudi Arabia

Introduction

The date palm, *Phoenix dactylifera* L. (Arecales: Arecaceae), is cultivated in warm climates across the globe and contributes significantly to the international date industry. It provides highly nutritious fruit, which possesses medicinal value and can serve as a complete diet for humans (Al-Karmadi and Okoh, 2024). Saudi Arabia is one of the warmest countries and produces over 1million tonns of quality dates in 2024. However, several pests are associated with both the date fruit and the date palm tree (Aldawood et al., 2013, Antary et al., 2015). The red palm weevil, *Rhynchophorus ferrugineus* (Olivier) (Coleoptera: Dryophthoridae), attacks date palm trees and has become a global issue for many palm species (Murphy and Briscoe, 1999; Faleiro, 2006; Aziz, 2024; Omer et al., 2025). The *R. ferrugineus* is highly concealed in nature, persisting, reproducing, and feeding within the date palm trunk until the tree dies (Abraham et al., 1998). The larval stages feed vigorously on the succulent tissues of the palm, reducing it to a damaged structure. The *R. ferrugineus* can complete several generations in a single palm tree. Once the internal tissues of the attacked trees are depleted, adults move to another young palm and lay eggs. After hatching, the neonates travel towards the inner tissues of the palm and begin feeding (Faghih, 1996).

Farmers use chemical insecticides such as imidacloprid, deltamethrin, dimethoate, endosulfan, fipronil, and emamectin benzoate (Aretor®) to protect their date palm trees from *R. ferrugineus* infestation (Azam and Razvi, 2001, Kaakeh, 2006, Dembilio and Jacas, 2012, Mashal and Obeidat, 2019, Ali-Bob, 2019, Rasool et al., 2024, Nasraoui et al., 2024). Several studies have examined pesticide residue in fresh fruit (Osaili et al., 2022) and vegetables (Mehta et al., 2025), and they have suggested ways to minimize residues in vegetables (Balah et al., 2024). In contrast, few studies have assessed the fate of pesticides in trees (Liang et al., 2024) and date fruit after they have been sprayed or injected into date palm trees (Rohani et al., 2024).

There are no records of emamectin benzoate contamination in the date palm trunk, leaves, or fruits in Riyadh, Saudi Arabia. As a result, it was necessary to investigate the fate of emamectin benzoate in the date palm trunk and leaves, as well as the possibility of residue accumulation in the fruit. Several well-developed techniques are being used globally for the chemical residue analysis in date fruits such as; QuEChERS method, GC-MS/MS, and UHPLC-MS/MS (Khezri et al., 2022; Morsi et al., 2014). In this regard, the purpose of this study was to examine the accumulation of emamectin benzoate residues in date palm trees,

specifically in the trunk, leaves, and fruits. To execute the experiment, the QuEChERS method was selected.

Material and methods

Apparatus and equipment

A Specialized Tree Micro-Injection (TMI) device (Syngenta, Switzerland) was used to inject the insecticide solution into the date palm trunks. The TMI is a device that is specifically equipped for the injection of pesticides into the trees. This high-precision device is pre-programmed to inject undiluted chemicals into tree trunks.

Tree selection

Experiments were carried out using a completely randomized block design at a date palm farm in Al-Kharj, Riyadh region, Saudi Arabia (24.14.84°N, 15.182°E). In our open field trial, a completely randomized block design is used to compensate for environmental variability such as soil type and microclimate changes, ensuring that treatment effects were reliably assessed. This approach improves the precision of our results by grouping similar experimental units together, allowing for reliable comparisons between treatments. Non-infected date palms of similar age, approximately 10–15 years old, were selected for the experiment. In total, there were three blocks, each containing three date palm trees, and each palm tree was considered a replicate. In each of the three blocks, two date palm trees were injected with insecticide, while one tree received a water injection, for a total of six trees treated with insecticide and three treated with water throughout the experiment.

Initially, the selected trees were labeled with colored strips and carefully inspected for any signs of infestation. No date palm management activities were conducted on the experimental trees by the farm manger and employees. The trunk height of selected trees low crown (canopy) ranged from 80 cm to 430 cm, and the radius of each trunk ranged from 21 cm to 36 cm.

Injection procedure

The experimental trees were drilled in four different directions, (East, North, West, and South) covering the entire trunk. Four holes were made using a drill bit with a diameter 6 mm, drilled to a depth of one-third of the date palm trunk diameter at an angle of 15–20 degrees. Each tree was injected in a spiral manner with 48 mL of emamectin benzoate (Aretor®) 4EC, Syngenta, Switzerland), 12 mL per injection site, at intervals of 50 cm, from a height of 1.0 to 2.5 m from the base. Control trees were injected with a similar amount of distilled water. After

injection of emamectin benzoate, a biodegradable plug was inserted into each drilled hole to prevent backflow of the insecticide solution.

Trunk samples collection

Depending on the trunk height, samples were obtained from two positions on the date palm trunk at heights of 0.5-1 m and 3-3.5 m representing the lower (below injection area) and upper (above injection area) portions of the trunk, respectively. The initial sample (Month 0) was collected on the same day as the treatment, followed by samples taken 3, 9, and 15 months later. The samples were taken using a drill, with a core sample length of 10-12 cm and a diameter of 5-7 cm, to minimize any damage to the trees (Figure 1). Trunk samples were collected from locations other than the injection sites.

Figure 1. Drilling and sampling procedure for trunk samples.

To avoid sample contamination, the drill bit was cleaned and disinfected with acetonitrile (ACN) prior to drilling. The samples were placed in plastic bags or aluminum foil, then stored in an icebox to protect pesticide residues from photo-degradation, and subsequently transported to the laboratory. In the laboratory, trunk samples were stored in amber bottles (dark) or wrapped in aluminum foil and preserved in a cooled ice chest. All samples were analyzed in triplicate.

Leaf and fruit sample collection

Fresh leaves from the treated and control trees were cut randomly, sealed in clean aluminum foil, and placed in an icebox to protect the pesticide residues from photodegradation. The samples were immediately stored in a freezer at -0ºC until preparation and analysis. Fruit samples were collected directly from the treated and control trees, processed immediately by peeling to remove the seeds, thoroughly homogenized, and stored in the freezer at -80ºC until

preparation and analysis. All the samples were received, processed, and stored following European guiding principle (European Commission, 2019).

Chemicals and equipment for quantitative analysis of emamectin benzoate

Emamectin benzoate (Aretor®), was provided by Syngenta, Switzerland. Acetonitrile (High-performance liquid chromatography, (HPLC) grade) and trifluoroacetic acid (HPLC grade) were purchased from Sigma (St. Louis, MO). The Waters 2545 Binary Gradient Module HPLC was equipped with a diode array detector (DAD) and an auto-sampler 2707 ALS automatic injector (Waters Corporation, USA). The Quick, Easy, Cheap, Effective, Rugged, and Safe (QuEChERS) Extraction Tube (Agilent Part No. 5982–5755/50 mL) consisted of 4 g of $MgSO_4$, 1 g of Sodium chloride (NaCl), 1 g of Sodium citrate, and 0.5 g of disodium citrate sesquihydrate. The QuEChERS Dispersive SPE Tube (Agilent Part No. 5982-5122 /2 ml) consisted of 50 mg of PSA, 50 mg of C18, (PSA (Primary Secondary Amine) and C18 (Octadecyl Silane) absorbents) and 150 mg of $MgSO_4$.

Reference stock and standard solutions preparation

A standard sample of emamectin benzoate was used to prepare reference stock solutions of 1000 mg/L and an intermediate solution of 10 mg/L in acetonitrile. Standard solutions were prepared by diluting the reference stock solutions with the blank solution to concentrations of 25, 50, 100, and 500 ng/L.

Sample preparation for analysis

Samples (trunk, leaves, and fruits) were weighed in triplicate and homogenized in a blender for 2 minutes. For each sample, 10 g of homogenized trunk tissue was placed in a 50 mL centrifuge tube and extracted with 20 mL of acetonitrile, then vortexed for 1 minute. The drying agent (Agilent Part No. 5982–5755/50 mL) was immediately added to the extracted solution and mixed in an extraction tube according to the QuEChERS method (Anastassiades et al. 2007; Burkhard et al. 2015). The extraction tube was centrifuged for 5 minutes at 3000 rpm. Ten milliliters of the supernatant were transferred to a TurboVap® tube evaporated, and concentrated to 2 mL using a gentle stream of nitrogen to increase the concentration of emamectin benzoate residue in the samples. The solution was then transferred to a QuEChERS Dispersive SPE tube and mixed. After centrifugation for 5 minutes at 4000 rpm, the clean supernatant was filtered through a 0.2 mm PTFE syringe filter and loaded onto the HPLC auto-sampler vials (Figure 2). For method validation, quality assurance, and quality control

samples, including blanks and standard reference materials, were prepared and analyzed alongside the treatment samples.

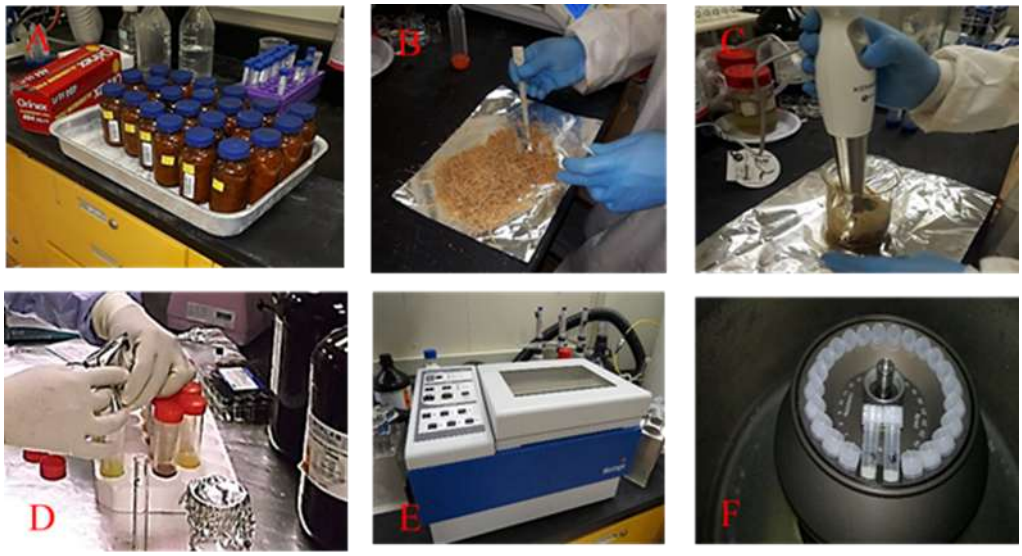

Figure 2. Step-wise description of sample preparation using quechers method Samples (A), Samples preparation, (B), grinding (C), Sample Extraction (D), Concentrations (E), and Centrifugation (F).

Analysis of emamectin benzoate residues

Emamectin benzoate residues in the tested samples were analyzed using high-performance liquid chromatography (HPLC) connected to a DAD. The column used was a Mediterranean Sea 18 (Teknokroma C18 column (150×4.6 mm, 3.5 µm particle size) Part No. TR-010903, Barcelona, Spain). The analysis was carried out at room temperature. The mobile phase consisted of a combination of 0.1% aqueous trifluoroacetic acid–acetonitrile (Isocratic70-30 to 75-٢٥) at a flow rate of 1.2 mL/min for10 minutes, followed by a cleaning phase at a flow rate of 1.2 mL for 2 minutes. The wavelength of light was 244 nm. Retention time for emamectin benzoate was 4.03 minutes (Figure 3).

The recoveries were obtained with the extracted spiked samples. Matrix-matched calibration solutions were prepared by spiking blank fruit samples at three different concentration levels (i.e., limit of quantification: 0.1, 0.5 and 1 ng/kg) of emamectin benzoate, with three replicates for each level (Liu et al. 2014). Before the extraction step, the spiked samples were allowed to stand for 2 hours at room temperature to let the pesticide to distribute evenly and ensure complete interaction with the sample matrix. The limit of detection (LOD) was 100 ppb, and

the limit of quantitation (LOQ) was 500 ppb. The injection volume was 20 µl for both standard and sample solutions.

The analysis to detect any residue of emamectin benzoate in the leaves and fruit samples was performed with high sensitivity. The calibration curve construction and calibration verification checks were performed after every 10 analyses and consistent results were obtained. The limit of detection (LOD) was 100 ppb and the limit of quantitation (LOQ) was 500 ppb. By following this analysis protocol, the available concentration could be calculated from the constructed calibration curve down to a few ppbs.

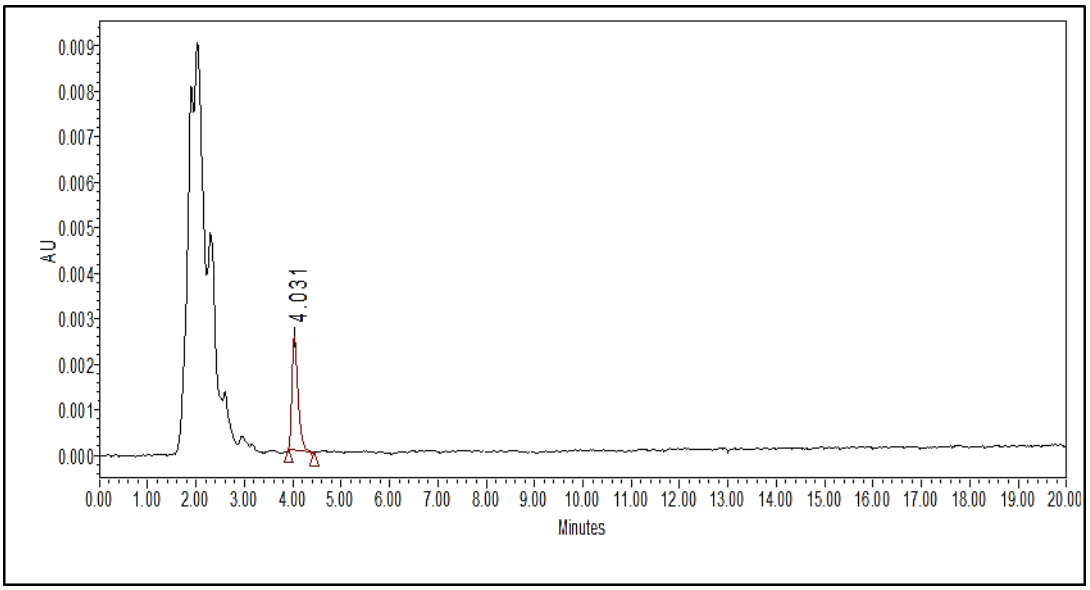

Figure 3. Chromatogram of emamectin benzoate standard (0.025ppb) with matrix.

Statistical analysis

The acquired data on emamectin benzoate (ng/g) in the samples were analyzed using the statistical program SAS (SAS 2009), which used Repeated Measures ANOVA to account for data collected from the same trees at different times. The data were subjected to analysis of variance (ANOVA) technique (Fisher's analysis of variance) to determine the significance level of emamectin benzoate residue data and means were separated using the least significant difference (LSD) at $P < 0.05$.

Results

Results showed the bioavailability of emamectin benzoate (Aretor®) in the trunk samples, which was detected after all exposure periods. The mean concentrations of emamectin benzoate in the whole trunk (additive of the top and bottom samples) are shown in Table 1, whereas the mean concentrations of emamectin benzoate in the top and bottom of the trunk are shown in Tables 2 and 3, respectively. Concentrations of emamectin benzoate were 55.1,

47, 178.2, and 213.6 ng/g after 0, 3, 9, and 15 months of exposure, respectively, in the whole trunk, whereas in the control samples, emamectin benzoate was not detected (ND) (Table 1). These experiments indicate that the pesticide moved upward after injection, with some areas showing no residue.

Table 1. Mean of overall concentrations of emamectin benzoate (ng/g ± SE) in the entire date palm trunk, leaves, and fruit across different time intervals following treatment.

Exposure (months)	Concentration (ng/g)		
	Trunk	Leaves	Fruit
Control	NQ*	NQ*	NQ*
0**	55.10 ± 6.1b	NQ*	NQ*
3	47.00 ± 6.3b	NQ*	NQ*
9	178.20 ± 57.8a	NQ*	NQ*
15	213.60 ± 35.5a	NQ*	NQ*
Statistics (*F*, *P*, df)	*F*= 7.55, *P*= 0.0079 df 3, 12 (Model, corrected total)		

Means followed by the same letter (s) are not significantly different (LSD test at $P < 0.05$)
*present but not quantifiable; **The samples were taken on the same day of the treatment.

The concentrations of emamectin benzoate in the bottom of the date palm trunk were 19.7, 26.4, 75.7, and 125-ng/g after 0, 3, 9, and 15 months of exposure, respectively, whereas in the control samples, emamectin benzoate was not detected (ND) (Table 2).

Table 2. Mean concentrations of emamectin benzoate (ng/g ± SE) in the bottom of the date palm trunk across different time intervals following treatment.

Exposure (months)	Concentration (ng/g)		
	Trunk	Leaves	Fruit
Control	NQ*	NQ*	NQ*
0**	19.70 ± 3.01b	NQ*	NQ*
3	26.40 ± 11.4ab	NQ*	NQ*
9	75.70 ± 44.7ab	NQ*	NQ*
15	125.00 ± 18.7a	NQ*	NQ*
Statistics (*F*, *P*, df)	*F*= 2.43, *P*= 0.1323 df 3, 12 (Model, corrected total)		

Means followed by the same letter (s) are not significantly different (LSD test at P < 0.05) *present but not quantifiable; **The samples were taken on the same day of the treatment.

The concentrations of emamectin benzoate in the top of date palm trunk were 35.5, 16.5, 87, and 88.6 ng/g after 0, 3, 9, and 15 months of exposure, respectively. However, in the control samples, emamectin benzoate was not detected (ND) (Table 3). However, the emamectin benzoate concentration levels in the leaf and fruit samples were not detected at any exposure time.

Table 3. Mean concentrations of emamectin benzoate (ng/g ± SE) at the top of the date palm trunk across different time intervals following treatment.

Exposure (months)	Concentration (ng/g)		
	Trunk	Leaves	Fruit
Control	NQ*	NQ*	NQ*
0**	35.50 ± 6.1a	NQ*	NQ*
3	16.50 ± 6.3a	NQ*	NQ*
9	87.00 ± 57.8a	NQ*	NQ*
15	88.60 ± 35.5a	NQ*	NQ*
Statistics (*F*, *P*, df)	*F*= 2.62, *P*= 0.1036 df 3, 14 (Model, corrected total)		

Means followed by the same letter (s) are not significantly different (LSD test at P < 0.05) *present but not quantifiable; **The samples were taken on the same day of the treatment.

Discussion

The results obtained illustrate the bioavailability of emamectin benzoate (Aretor®) in the date palm trunk up to 15 months into the experiment. The average concentration (88.6 ng/g) of emamectin residue found in the treated palm tree trunk during the trial was below the regulated level of 100 ng/g for the EU and USA, respectively (EFSA, 2020). A trend of emamectin benzoate distribution was observed in the whole trunk, at the bottom, and at the top of the trunk throughout the trial period. A low concentration of emamectin benzoate residues (below LOQ) was detected in the zero-time exposure samples, which we suspect may have been due to cross-contamination during the sampling procedure.

Variability in the distribution of emamectin benzoate across the trunk over different sample times may be related to the sampling location, depth, and timing.

Both the tree structure and physiology play very important roles in residue translocation.

In line with external studies, the absence of detectable residues in fruits is extremely compatible with Nasraoui et al. independent 2025 study, which showed no emamectin residues in date fruits at any maturation stage following trunk injection over a one-year monitoring period. Similarly, not quantifiable residues were reported in date palm fruits 60 days post-injection when 4% of emamectin benzoate was injected into trees using tree micro-injection device (Mashal and Obeidat, 2019). Similar patterns of long persistence in trunk tissues, variable levels in branches or fronds, and low or non-detectable residues in reproductive tissues have been reported in other tree systems (pecan, apple, horse chestnut), supporting the overall conclusion that emamectin is largely restricted to structural tissues after trunk injection (Takai et al., 2004, Burkhard et al., 2015; Coslor et al., 2019).

Previously, the highest amount of chlorpyrifos-ethyl residues in date palms was found with injection treatment (11.5 µg/kg) compared to foliage spray treatment (0.9 µg/kg) in Egypt (El-Tokhy and Amer, 2017). Residues of lambda-cyhalothrin (0.0034 µg/kg) have been reported in date palms from the UAE (Al-Samarrie and Akela, 2011), and Fenazaquin residues (> 0.1 µg. Kg-1) were reported in date fruits from Tunisia (Attia et al., 2019). The studies confirm that trunk injection of emamectin benzoate in date palm trees for red palm weevil control is relatively safe for people because not quantifiable residues were found in the leaves. In contrast, spraying date palm trees to manage pests and mites during fruit development can be hazardous. Recent residue analyses of date fruit samples from local markets in Riyadh and Al-Qassim, Saudi Arabia, revealed that several samples contained pesticide residues exceeding MRLs (El-Saeid and Al-Dosari, 2010, Abdallah et al., 2018). To protect consumers,

implementing an effective control program is essential to reduce pesticide accumulation in date fruits. It is crucial to emphasize that in the present research sample size of three blocks each containing three date palm trees may limit the capacity to capture diversity between trees and within trees. The trees used were well-grown specimens from a working farm, as obtaining fruiting date palms may be expensive.

Future research should employ larger sample sizes to improve the robustness of their findings, as well as investigate potential exposure pathways to nontarget organisms via abscised, decomposing tissues or sap to offer a thorough environmental risk assessment of injected emamectin. Also, trunk core sampling from multiple height zones and radial positions can be helpful to better assess the distribution of emamectin benzoate. This approach will help account for the known axial and radial gradients of xylem mobile compounds, providing a more comprehensive understanding of its distribution within date palm trunks. Incorporating these assessments will help us better understand the ecological implications of its use.

Conclusion

In conclusion, present study findings strongly indicated that trunk injection under the tested conditions is unlikely to produce fruit residues above widely used MRLs, the current analytical LOQ does not exclude residues at lower, but still measurable, levels; and residues were not detected above 0.5 mg/kg. These findings confirm that emamectin benzoate, administered via trunk injection, is considered safe for humans and animals under the tested conditions and effective for sustainable management of red palm weevil.

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