Emamectin Benzoate Residues in Fruits and Fronds of Date Palm Trees Following Trunk Injection

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Abstract

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Red palm weevil (RPW) is one of the most dangerous insect pests of the date palm. It primarily infests the palm trunk and, less frequently, the palm top. This study, conducted in the Kingdom of Saudi Arabia, is a continuation of previous work that demonstrated trunk injection with undiluted emamectin benzoate in date palm apically infested with RPW, resulting in approximately 91% complete healing. The same trunk injection achieved a 100% success rate in killing all RPW instars inside the palm trunk. In the present experiment, the same insecticide was injected at various intervals (between 1 and 12 months) into the trunks of healthy date palms, and subsequently, the residues of emamectin benzoate in fruits and fronds of the palm trees were analyzed. RPW larva rearing was conducted in the laboratory inside cut frond bases (karabs) of the injected palms. Results obtained indicated absence of emamectin benzoate residues in the fruits at all maturation stages: early ripening (Bisr) in late-June, mid-ripening (Rotab) in mid-August, and final ripening (Tamr = Date) in late-September. As for the insecticide residues, significant residues of emamectin benzoate (0.054 mg/kg) were detected in karabs after a trunk injection of only one month. In addition, RPW larva rearing inside the karabs, one month after trunk injection, gave a significant high larva mortality rate 83.3%. This study indicated that with the injection of the palm trunk by the undiluted pesticide emamectin benzoate, no pesticide residues were found in the palm fruits, and if the injection was made one month earlier, it provided significant protection to the bases of the fronds against the red palm weevil.
Keywords: Date palm, emamectin benzoate, insecticide residues, red palm weevil, trunk injection.

Introduction

Red palm weevil (RPW), *Rhynchophorus ferrugineus* Olivier (Coleoptera: Curculionidae), is known as one of the most dangerous insect pests that attacks date palm (*Phoenix dactylifera* L.) at the basal part of the trunk and, less frequently, at the head of the date palm tree. In the Kingdom of Saudi Arabia (KSA), the RPW damage on date palms are usually observed at the low part of the trunk, but since few years ago, apical infestations started to be increasingly observed, reaching an average rate of around 11% of the total number of infested palm trees in the KSA during 2022 (Weqaa Center, 2022).

In previous work (Nasraoui *et al.*, 2024), a simple, easy, and low-cost technique of insecticide trunk injection was used to control RPW infestation at the trunk basal part and at the top of date palm. This method was inspired from the trunk injection technique (endotherapy) reported to control RPW in ornamental Canary palm (*Phoenix canariensis* hort. ex Chabaud) commonly infested at the apex. Following injection in the trunk basal part of the Canary palm, the insecticide migrates to the top of the palm tree, killing all RPW instars and protecting the palm head from later RPW infestations (Chihaoui-Meridja *et al.*, 2019; Ferry & Gomez, 2014; Ferry *et al.*, 2019; Gomez & Ferry, 2019). In our previous work during 2020 on date palms (Nasraoui *et al.*, 2024), the insecticide was injected into the trunk at around one meter above the ground level against the apical infestation and at the ground level against the trunk infestation. The insecticide used in this study was emamectin benzoate, since previously published research showed that it killed all RPW instars in the Canary palm top and protected it for a period of one year from any new RPW infestations (Chihaoui-Meridja *et al.*, 2019; Gomez & Ferry, 2019). The same insecticide was also used to control the RPW trunk infestation by injecting the trunk of the date palm (Mashal & Obeidat, 2019; Rasool *et al.*, 2021).

In a previous study, interesting results were obtained with the insecticide emamectin benzoate for RPW control (Nasraoui *et al.*, 2024). However, such a study lacked insecticide residue monitoring in the injected date palm. Thus, this study focused on monitoring the insecticide emamectin benzoate residues in the fruits and fronds of the palm trees over one year period.

Materials and Methods

Experimental site

This study was performed during 2022/23 at Nofa, a site located 100 km west from the city of Riyadh, KSA (24°22'46.5"N, 45°57'49.0"E). This site contains orchards of date palms and other fruit trees. This study was performed on 30 healthy date palm trees (variety "Sagae") with approximately the same age (around 30 years) and the same height (around 6 m).

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Chemical treatment

The insecticide used in this study was emamectin benzoate 50 g/L (Proact 50 EC). Trunk injection of the insecticide into the palm trunk was made by first drilling a hole in the trunk (1 cm diameter × 35 cm long), four opposite 45° downward-inclined holes around the trunk at nearly 1 m above the ground in each tree, and then a 100 ml-syringe (without needle) was utilized to inject the insecticide inside the holes (25 ml/hole). Each treatment and the control were repeated in 5 palm trees, as follows: (1) first injection was made during the first week of October 2022, 12 months before the fruits final ripening; (2) second injection was made during the first week of January 2023; (3) third injection was made during the first week of March 2023; (4) fourth injection was made during the first week of July 2023; (5) the last injection was made during the last week of August 2023. The holes were closed by mud immediately after each injection. The control trees were not injected with the insecticide.

Samples collection

The residues were analyzed in fruits and fronds. Fruits were collected at 3 ripening stages which were early ripening (Bisr) in late-June, mid-ripening (Rotab) in mid-August and final ripening (Tamr = Date) in late-September (Figure 1). The fronds were collected in late-September along with the dates.

Figure 1. Collected date palm fruits at three ripening stages: Bisr (up), Rotab (middle), and Tamr (down).

Bisr, rotab, and date fruits were collected as samples of 250 g from each of the four cardinal directions of each palm tree. The collected four samples were mixed in one composite sample of 1 kg/tree and sent to the laboratory in ice cool boxes to be analyzed for the insecticide residue content.

Fronds were collected by removing one frond from each cardinal direction of each palm tree. Fronds were then cut at 20 cm from their bases, and the pieces obtained (karabs) were mixed in one composite sample of 4 karabs/tree and sent to the laboratory in ice cool boxes to be analyzed for the insecticide residue content. Karab bases are known to be the place where RPW attack the heads of date palm trees.

Insecticide residue analysis

For the analysis of the emamectin benzoate residues, fruits and karabs were sent to an accredited laboratory (ISO 17025) for pesticide residue analysis (IDAC Merieux Nutriscience, Riyadh, KSA) which performed the analyses according to the AOAC 2007.01 method. Each sample was homogenized and weighed, then extracted with acetonitrile containing 1% acetic acid. The sample was treated with Quechers kits and processed by the Instrument Shimadzu-LCMS-8045/LCMSMS. The chemicals and standard company are LGC/Dr. Ehrenstorfer. All analyses were performed with a Limit of Quantification (LOQ) equal to 0.01 mg/kg (or ppm).

Rearing larvae inside basal fronds (karabs)

For rearing RPW larvae, fronds were collected (one frond from each palm tree), cut at 25 cm from their bases and the obtained pieces (karabs) were sent to the laboratory in ice cool boxes. In the laboratory, each karab was dug 2 holes, and one big old RPW larva (all larva average = 3.12 g) was brought from a previous RPW rearing and introduced in each hole and the hole closed with palm fibers. Two weeks later, the karabs were dissected to determine the larva mortality.

Statistical analysis

Since the treated palms were randomly distributed in the orchard, the experimental design of the site was considered as a completely randomized design (CRD) experiment. Likewise, CRD was used for the laboratory experiment, considering that inside the experimental chamber, all conditions are the same: light, temperature and humidity. A one-way ANOVA calculator program was used at P=0.05, that includes Tucky HSD, which is available online (https://www.socscistatistics.com/tests/anova/default2.aspx).

Results

Insecticide residues in fruits

All analyzed fruits, collected at the 3 ripening stages (Bisr, Rotab and Date), were free from the residues of the insecticide emamectin benzoate, previously injected to the date palm trunks (similar to the date palm control without insecticide injection). The LOQ is equal to the maximum residue limit (MRL) in date fruits, which is 0.01 mg/kg (EFSA, 2022).

Insecticide residues in fronds

Non-insecticide-injected date palm trees (control) showed no residues in fronds. Insecticide-injected date palms at 12, 9, 6 and 3 months before collecting fronds showed very low insecticide residues that were not significantly different from the control at P=0.05. The residue average quantities varied between 0.000 and 0.017 mg/kg. The only case with a statistically significant quantity of insecticide residues was obtained in the palms injected 1 month before collecting fronds and reached 0.054 mg/kg (Figure 2-A).

Larva rearing inside basal fronds (karabs)

Rearing RPW larvae inside the karabs of injected palm trees, 3 to 12 months before fronds collection, resulted in a very low mortality rate, which was not significantly different (P=0.05) from the non-insecticide injected palm control (between 8.3% and 16.7%). In contrast, the karabs from palms injected 1 month before, fronds collection showed high mortality rate of larvae (83.3%) (Figures 2-B and 3).

Discussion

Results obtained in this study confirmed that there was no emamactin benzoate insecticide residues detected in all fruit stages, regardless of the time of trunk injection, which is in agreement with previous findings (Hajjar *et al.*, 2018; Mashal & Obeidat, 2019).

In our experiment, even if there are emamectin benzoate residues in the date fruits with a concentration not detectable by the instrumentation used (less than LOQ 0.01 mg/kg), this concentration is also below the MRL (0.01 mg/kg, EFSA, 2022) which means that the dates are safe for human and animal consumption. However, farmers should not inject emamectin benzoate for RPW control during the last month before fruits harvest. This precautionary condition is due to the fact there is no information available on the residues content when injection is made less than one month from harvest.

Compared to the non-injected control, the frond bases (karabs) of the date palms contained non-significant emamectin benzoate residues for all trunk injection times (≤ 0.017 mg/kg), except when the injection was made one month before collecting the samples (0.054 mg/kg). Such result was confirmed by the fact that 83% mortality of RPW larvae was only obtained following a one-month period after injection. In all other samples, including the non-injected control, we obtained less than 17% of dead larvae. This result means that the injected insecticide provided to the fronds, and then the palm head, an almost complete protection against RPW for at least one month. In further work, it will be helpful to check if a maximum protection (100%) can be reached with 0.5, 1.5, 2.0 and 2.5 months after truck injection. The present result also is in agreement with our previous work, where injected emamectin benzoate in the trunk of RPW-infested head palms, produced full control to 91% of the treated date palms (Nasraoui *et al.*, 2024). To the contrary, our present results on date palms are very different from those of some previous studies on ornamental Canary palm where it was mentioned that the emamectin benzoate trunk injection of the Canary palm heads infested with RPW, provided them a long time protection against RPW reaching one year (Chihaoui-Meridja *et al.*, 2019; Gomez & Ferry, 2019).

In the present experiment in the KSA, the injected emamectin benzoate provided highly significant protection to the top of the date palm for one month, whereas in southern Europe (Italy and Spain), this protection lasted a full year for the Canary palm (Chihaoui-Meridja *et al.*, 2019; Gomez & Ferry, 2019).

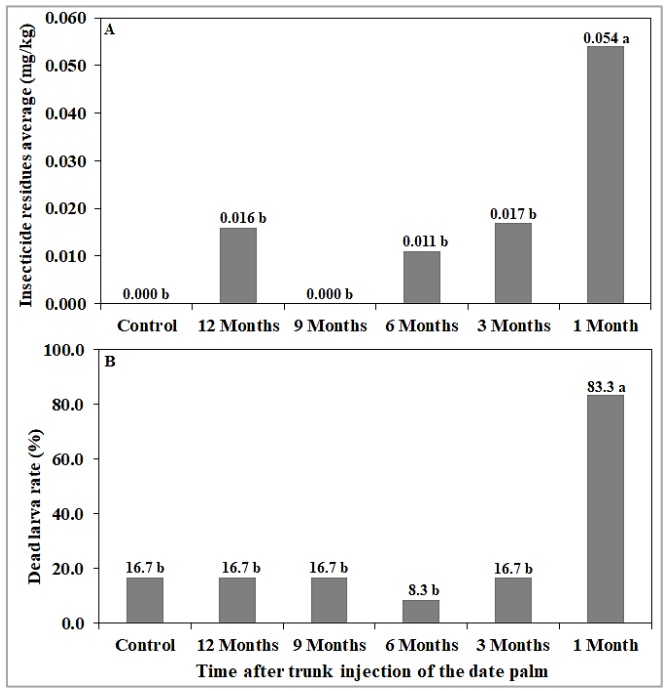

Figure 2. Residue average quantities of emamectin benzoate (A) and states of larvae reared, (B) in frond bases (karabs) of injected date palm trunks. Values followed by the same letter are not significantly different at P=0.032 for residue and P=0.027 for larvae reared.

Figure 3. Dead larvae reared inside frond basis (karab) of date palm injected in the trunk by emamectin benzoate one month earlier (A), and living larva reared inside karab of non-injected date palm control (B), in the laboratory.

This unexpected difference between the two cases might be explained by one or both of the following reasons: (i) the physiology of the date palm is different from that of the Canary palm in the way how to react to the injected pesticide, as it would be degraded in date palm faster than in the Canary palm, (ii) the Saharan climate of Riyadh Province (KSA), with its long dry period and high temperature, would quickly break down the pesticide inside the date palm tree, as compared to the climate of southern Europe, where the temperature is lower and the humidity is higher.

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الملخص

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تعدّ سوسة النخيل الحمراء واحدةً من أخطر الآفات الحشرية التي تصيب نخيل التمر، فهي تهاجم أساساً جذع النخلة، وعلى نحوٍ أقلّ قمّتها أيضاً. تأتي هذه الدراسة التي تمّ إجراؤها في المملكة العربية السعودية كاستمرارٍ لعملٍ سابقٍ أظهر أن حقن المبيد الحشري إيمامكتين بنزوايت غير المخفف في جذع نخيل التمر المصاب قمياً بسوسة النخيل الحمراء، أدى إلى تعافي حوالي 91% منها تماماً. كما أعطى هذا الحقن نفسه نجاحاً بنسبة 100% في قتل جميع أطوار سوسة النخيل الحمراء في جذع النخلة. في هذه الدراسة، تمّ حقن المبيد الحشري نفسه المذكور آنفاً في أوقات مختلفة (بين 1 و 12 شهراً) في جذوع نخيل تمر سليم (صنف "صقعي")، وتمّ تحليل متبقيات المبيد (إيمامكتين بنزوايت) في ثمار وسعف أشجار النخيل. وفي المختبر، تمّ أيضاً تربية يرقات سوسة النخيل الحمراء داخل قواعد السعف المقطوعة (الكرَب) للنخيل المحقون. أظهرت النتائج عدم وجود متبقيات إيمامكتين بنزوايت في الثمار خلال جميع مراحل النضج: النضج المبكر (بسر) في أواخر شهر حزيران/يونيو، منتصف النضج (رطَب) في منتصف آب/أغسطس والنضج النهائي (تمر) في أواخر أيلول/سبتمبر، وذلك مع جميع أوقات حقن المبيد. وفيما يتعلق بمتبقيات المبيد في السعف، تمّ تسجيل وجود متبقيات مرتفعة معنوياً من المبيد إيمامكتين بنزوايت (0.054 مغ/كغ) في الكرَب بعد حقن الجذع بشهر واحدٍ فقط. وتمّ تأكيد هذه النتيجة من خلال تربية يرقات سوسة النخيل الحمراء داخل الكرَب، حيث أعطى الحقن بعد شهر واحد فقط، نسبة مرتفعة معنوياً لليرقات المقتولة (83.3%). تشير مجموع نتائج هذه التجربة أنه مع حقن جذع النخلة بالمبيد إيمامكتين بنزوايت غير المخفف، لم يتمّ العثور على متبقيات المبيد في ثمار النخلة، وإذا تمّ الحقن قبل شهر، فسيوفر هذا حمايةً كبيرةً لقواعد السعف من هجمات سوسة النخيل الحمراء.

كلمات مفتاحية: إيمامكتين بنزوايت، حقن الجذع، سوسة النخيل الحمراء، متبقيات المبيدات، نخيل التمر.

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