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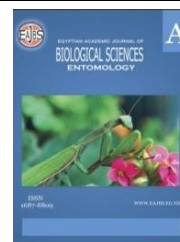
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Field Efficacy of Chlorpyrifos in A Chitosan Nanoformulation Against the Red Palm Weevil, *Rhynchophorus ferrugineus* (Coleoptera: Curculionidae) in Date Palm

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ABSTRACT

The current study aims to test a nanoformulation of an approved RPW-controlling active ingredient in order to optimize its delivery to RPW in infested date palms and protect it against premature degradation. This includes evaluating their ability to deliver the encapsulated insecticide active ingredient in a controlled release manner into the aqueous environment. Chitosan-Chlorpyrifos nanoformulation (Ch-Fos NF) was formulated via sonication. HR-TEM and EDX Mapping were used to characterise the resulting nanoformulation. The toxicity of five distinct dosages of Chitosan-fos NF (50, 100, 150, 200, and 250) against 5th instar RPW larvae was determined via bioassay testing. The observed fatality percentages after 48 hours were 10, 15, 35, 55, and 80, respectively. The nanoformulation was applied on date palm and compared with the active ingredient, the carrier and a control test. The evaluation proved the efficacy of nanoformulation in insecticide delivery. Further study is required regarding phytotoxicity and residual analysis.

INTRODUCTION

The red palm weevil (RPW), scientifically known as *Rhynchophorus ferrugineus*, is an insect that is recognized globally for the destruction it causes. It poses a severe, relentless threat to various palm species around the world, causing significant damage that is difficult to manage. This invasive pest was first identified in the Arabian Gulf region in the mid-1980s, marking the beginning of its destructive path (FAOSTAT 2013). Since its first identification, the red palm weevil has rapidly spread to other parts of the world. It has blazed a path of destruction across regions such as the Middle East, Southern Asia, North Africa, and the Mediterranean basin. This spread has been marked by significant damage to palm species in these regions, causing concern for local ecosystems and economies (FAOSTAT 2015).

Recent findings have added to the growing concern about this pest. The research shows that this pest is now establishing itself in new territories, including East Africa and the Caucasian region (El-Shafie *et al.* 2005). These new establishments pose significant

risks to date palms in East Africa and canary island palms in the Caucasian region (Faleiro *et al.* 2019). It is feared that the damages caused by this invasive pest could have far-reaching effects on these regions' palm populations, posing significant risks to the local ecosystems and the industries that rely on these palms (Kassem *et al.* 2020). This spread underscores the need for increased vigilance and control measures to manage the ongoing threat posed by the red palm weevil (Faleiro *et al.* 2019).

The infestation of RPW carries with it significant and far-reaching implications for the overall production of palm species, particularly in regions where these trees are a primary agricultural commodity (Dembilio and Jaques 2015). It has been documented that the damage caused by pests and diseases, including RPW, can contribute to a staggering loss of up to 30% in date palm production (Faleiro 2006).

The economic impact of such a loss is substantial. It doesn't just result in the loss of millions of dollars that are spent on pest control and management efforts, which include the removal of diseased trees. It also accounts for the direct financial loss brought about by the destruction of infested palms as authorities grapple with the need to control the rampant spread of the invasive RPW (Manee *et al.*, 2023). In a bid to address this growing concern, the United Nations Food and Agriculture Organization, known universally as the FAO, has officially recognized the red palm weevil as a category-1 pest. This classification is especially pertinent in the Middle East and North Africa, where the RPW poses a severe threat. In these regions, the livelihood security of rural date palm producers, many of whom rely on these trees as their primary source of income, is under significant strain due to the relentless advance of the weevil (FAOSTAT 2015).

The situation is so severe that it necessitates the urgent development and implementation of effective methods to control the spread of the red palm weevil. These methods are needed not just for the sake of the palm species themselves, but also for the countless individuals and industries that depend on them for their livelihood and economic stability. The urgency of this situation cannot be overstated, and it underscores the need for concerted efforts in fighting the spread of the destructive red palm weevil (Turusov *et al.*, 2002).

The most common method of controlling the red palm weevil, often shortened to RPW, is through the strategic and systematic application of chemical treatments. These treatments are widely used in both preventive and curative capacities. Preventive treatments aim to deter the weevil from infesting the palm trees in the first place, while curative treatments are designed to eliminate any existing infestations and help the trees recover from the damage (Mafra-Neto *et al.*, 2014).

Despite their proven effectiveness in combating the RPW menace, these chemical treatments are not without their drawbacks. One of the most significant issues is the development of resistance among the pests. Over time, repeated exposure to these chemicals can lead the weevils to develop adaptations that make them resistant, thereby undermining the overall effectiveness of the treatments (Al-Sagheer and Faleiro 2011).

Another major concern associated with the use of chemical treatments is the potential for environmental contamination (Al-Sagheer and Faleiro 2011). The chemicals used in these treatments can seep into the soil, water sources, and the surrounding ecosystem, leading to a range of negative environmental impacts. These impacts can include harm to non-target species, disruption of local ecosystems, and potential health risks for humans and animals (FAOSTAT 2013).

Additionally, there are other challenges that complicate the use of chemical treatments for RPW control (Hallet *et al.*, 1993). These include the premature degradation of active ingredients, which can reduce the effectiveness of the treatments over time (Abbas *et al.*, 2006). The precise targeting of the pests is also a major challenge, as it

requires careful application to ensure it reaches the weevils without causing undue harm to the trees or surrounding environment. Furthermore, the eco-toxicological profile of these pesticides, which refers to their potentially harmful effects on the environment and non-target species, remains a significant area of concern (Faleiro 2006).

Despite these challenges, the application of insecticides remains a crucial component in the fight against the red palm weevil (Faleiro 2006). As we move forward, there's a pressing need for the development of new, environmentally friendly products that can effectively combat the weevil without causing the aforementioned problems. This, coupled with the implementation of more accurate application methods and reliable delivery techniques, could potentially revolutionize the way we manage this destructive pest. Therefore, ongoing research and development in this area is of paramount importance (Falerio *et al.*, 2019).

In recent years, an innovative and promising strategy that has gained considerable attention in the scientific community is the application of nanotechnology in the formulation of more effective and non-persistent insecticides (Iavicoli *et al.*, 2017). This cutting-edge approach utilizes the unique properties of materials at the nanoscale to provide solutions that can potentially revolutionize pest management strategies. In particular, nanoformulations are designed with the primary objective of enhancing the solubility of active insecticidal ingredients. By improving solubility, these ingredients can be better absorbed and utilized, thereby increasing their effectiveness in controlling pests like the red palm weevil (Falerio *et al.*, 2019).

Furthermore, nanoformulations offer the added advantage of controlled release of these active ingredients. This feature is particularly beneficial as it ensures a sustained release of the insecticide over a prolonged period, thereby providing longer-lasting protection against the pest. In addition, nanoformulations also offer protection against premature degradation of the active ingredients. This is a critical aspect, as it helps maintain the potency of the insecticide, ensuring that it remains effective for as long as possible (Iavicoli *et al.*, 2017).

The main focus of this research study is to explore the potential of a specific nanoformulation, namely chitosan nanoformulation, in the delivery of an insecticide known as chlorpyrifos. This insecticide is particularly effective in controlling the red palm weevil, making it a strategic choice for this study. The application of chitosan nanoformulation in delivering chlorpyrifos aims to maximize the impact of this insecticide on the red palm weevil, while also minimizing any potential adverse effects on the date palms themselves and the wider environment. The ultimate goal is to develop a more effective and sustainable strategy for managing the destructive impact of the RPW on date palms. As such, the findings of this study could have far-reaching implications for the future of pest management in the agricultural sector.

This research study is poised to make a substantial contribution to the existing body of knowledge through the exploration of the application of nanotechnology in the realm of pest control. It will delve into the intricacies of using nanoformulations, such as chitosan nanoformulation, to enhance the delivery and effectiveness of insecticides, specifically chlorpyrifos, which is particularly effective against the red palm weevil.

The successful implementation and validation of this innovative method in a real-world context could lead to more effective, sustainable, and environmentally friendly strategies for controlling the red palm weevil. This would not only benefit date palm producers who are directly affected by the damages caused by RPW, but it would also have far-reaching implications for the broader agricultural sector.

In the context of a growing global population and the increasing demand for food, the importance of sustainable and effective pest control measures cannot be overstated.

Therefore, the potential contribution of this study to the agricultural sector is immense, as it could help safeguard vital agricultural commodities, protect ecosystems, and ensure the livelihoods of millions of people who depend on the agriculture sector for their income. In conclusion, the findings from this research study hold the promise of making a significant contribution to our understanding of how nanotechnology can be harnessed to combat destructive pests like RPW. The implications of this could reverberate through the agricultural sector, potentially leading to more efficient, effective, and environmentally responsible pest management strategies.

MATERIALS AND METHODS

The chitosan powder used in the study was obtained from Techno Pharmachem in Bahadurah, Haryana, India, and the chlorpyrifos insecticide (Tafaban 48% EC) was purchased from ElHelb Pesticides & Chemicals in Damietta, Egypt. Deionized water was sourced from the Central Lab of Ain Shams University's Faculty of Science. The purchased chemicals were utilized in their original form without undergoing any additional purification processes.

Lab Application:

A. Tested Population:

Larvae, cocoons, and adult specimens of *R. ferrugineus* were obtained from The Agricultural Research Center for rearing over multiple generations. *R. ferrugineus* was consistently maintained on a diet of gritted sugarcane, which served as both a food source and substrate for oviposition, throughout these generations. The rearing process took place under controlled environmental conditions, with a temperature of 30 ± 2 °C and a relative humidity ranging between 60% and 80%. The photoperiod was set to approximately 12 hours of light followed by 12 hours of darkness (12:12 L:D) (Abbas 2013).

B. Synthesis of Tested Insecticide-Polymer Nanoformulation:

Sonication was used to synthesize a chitosan-chlorpyrifos nanoformulation (Ch-Fos NF) according to (Ahmed *et al.*, 2018) with modifications. To 1 Liter 10^{-4} M chlorpyrifos solution, eight grams of Chitosan nanoparticles (ChNPs) were added. For one hour, the mixture was sonicated. Filtration was used to obtain Ch-Fos NF. It was then allowed to dry at ambient temperature. Finally, it was sieved to prevent agglomeration (Ahmed *et al.*, 2018).

C. Characterization of Chitosan-Chlorpyrifos Nanoformulation:

Micrographs of the Ch-Fos NF were obtained using a high-resolution transmission electron microscope (HR-TEM), specifically the Tecani G20 model manufactured by FEI in the Netherlands. The HR-TEM was equipped with an Energy-Dispersive Spectrum (EDX) at the City of Scientific Research and Technological Applications (SRTA-City) in Borg El-Arab, Egypt, for imaging and revealing the crystal structure. To prepare the samples, 50 mg of the Ch-Fos NF was sonicated in deionized water for 30 minutes. The dispersed samples were then deposited onto copper grids coated with a thin layer to facilitate solvent evaporation. TEM images were analyzed using the HR-TEM analysis software TECNAI G2 (Ahmed *et al.*, 2018).

D. Laboratory Experiment:

Preliminary range-finding dose-mortality bioassays were conducted to determine the lethal range for the tested nanoformulation. A stock solution (1000 ppm) of Ch-Fos NF was prepared by dispersing 0.1 gm in 100 cc of deionized water. The finalized five concentrations (50, 100, 250, 200, 250 ppm) were obtained by dilution with the appropriate volume of deionized water.

Small peeled cylindrical sugarcane pieces (5 cm) were immersed in the prepared solutions for ten seconds before drying in air for two hours. The treated parts were placed in a cylindrical plastic box with a perforated top. Fifth instar larvae were placed in the box (Ten larvae per box), with 50 larvae for each pesticide concentration (represented by 5 repetitions). Each replicate had ten larvae (Elgohary *et al.*, 2015). After 48 hours, mortality records were collected. The percentage mortality was determined using (Finny 1972). Control test was performed using deionized water.

Field Application:

The altitude of the tested location was recorded: Ezbet Nashawyia, Sharkia (30.836733, 31.833996). In the experiment, the infected palm trees were divided into four treatment groups: eleven palms were injected with Ch-Fos NF, nine with commercial insecticide Tafaban (48% chlorpyrifos EC), eight with ChNPs, and eight served as the control group (deio).

The detection of infected palms was conducted using a red palm weevil detection device as well as optical methods. For each treatment, four holes were created 10 cm away from the injured area using an injection instrument, following the method described by El Ezaby (1997). Each palm received an injection of 1 liter of the prepared pesticide concentration. The injection holes were sealed with a combination of fiber and clay, as described by Abbas (2013). The control group was treated with 1 liter of water only.

The treatments were conducted at two separate sites using palm trees with similar characteristics, including an age range of 4 to 15 years, lengths ranging from four to five meters, and widths varying from 30-60 cm. The height of the injuries from the ground ranged from 20 to 140 cm. The pesticides were administered at a temperature of 30°C, with a volume of 1 liter per palm, targeting a 10 cm area using the injection apparatus. Pesticide efficiency was evaluated after 21,30 and 90 days of treatment.

Statistical Analysis:

The statistical analysis was performed using the LDP line program to assess the data on larval mortality. A 95% confidence interval (C.I) was employed to determine the lethal concentration (LC_{50}). The correlation coefficient (r^2) was calculated using the Finney formula, and the goodness of fit was evaluated using the Chi-square test (χ^2) (Finney 1972). Control mortality was adjusted using the Abbott formula (Abbott 1925). For field application, the Chi-square test (χ^2) was used to test the correlation between the dead insect in date palms and the applied formulations.

RESULTS

1 Chitosan-Fos Nanoformulations:

EDX mapping with TEM has shown that chloride was found in the sample of Ch-Fos NF. TEM images indicate the synthesis of chitosan chlorpyrifos nanoformulation in a sphere shape with a diameter of less than 3 nm. The abundance of carbon and hydrogen indicates the presence of chitosan in the sample (Figs. 1 and 2).

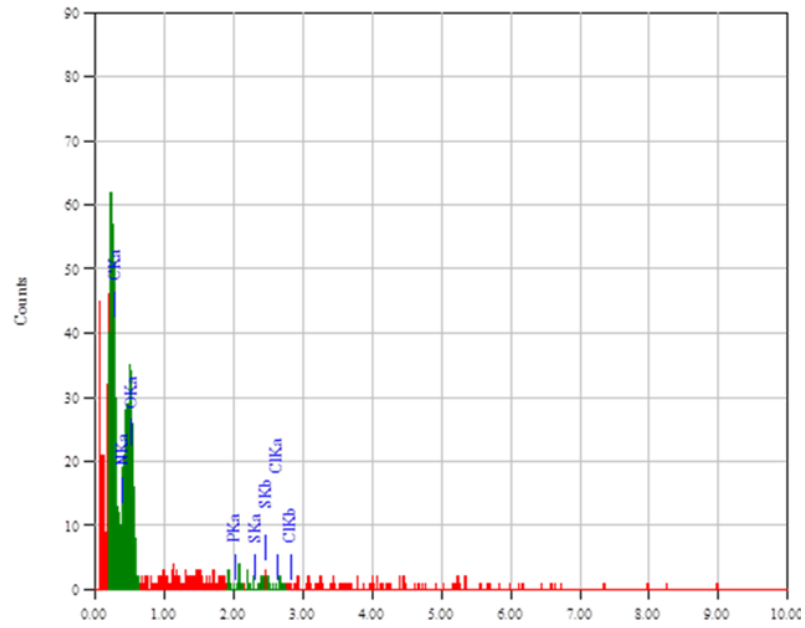


Fig. 1. EDX elemental mapping of Chitosan-Chlorpyrifos Nanoformulation

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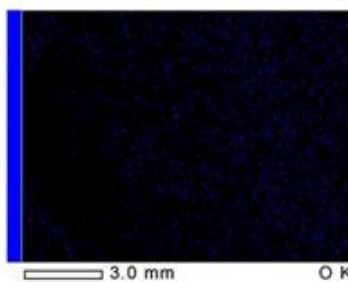
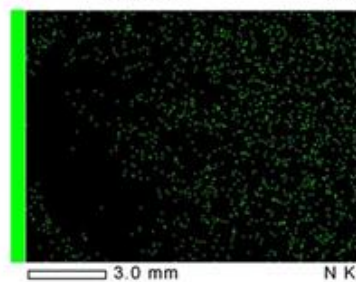
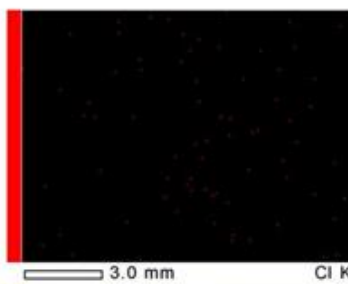
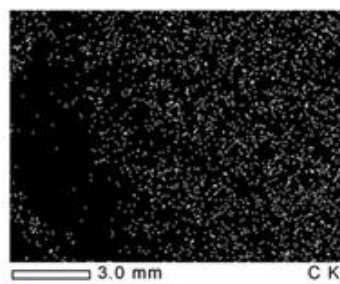
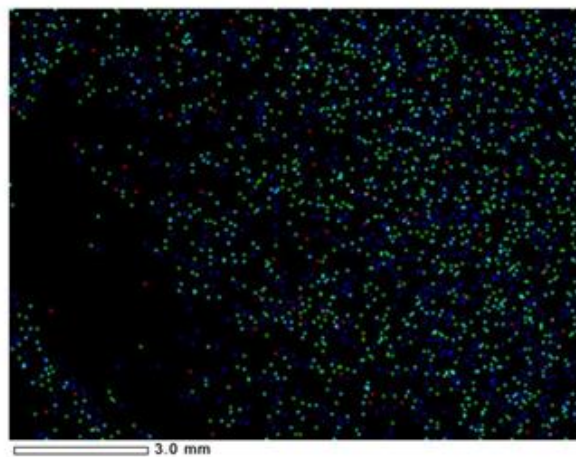


Fig. 2. EDX Mapping analysis Chitosan-Chlorpyrifos Nanoformulation.

2 Lab Application:

The toxicity of five tested concentrations (50, 100, 150, 200, 250) of Chitosan-fos NF against 5th instar larvae of RPW was evaluated by bioassay test. The observed mortality percentage after 48 hrs was 10, 15, 35, 55, 80, respectively. Median lethal concentration (LC50) was calculated using LDP line software and was 172.8369 ppm, as shown in (Table 1) and graphically illustrated in (Fig. 3). Dead larvae were recorded every 12 hours. The survival rate of larvae started to decrease after 36 hrs.

Table 1. Toxicity data of chitosan-fos nanoparticles against 3rd instar larvae of laboratory RPW strain ($p < 0.05$).

Concentrations (ppm)	Observed Mortality \pm SE %
50	10 \pm 0.00
100	15 \pm 0.00
150	35 \pm 3.20
200	55 \pm 3.60
250	80 \pm 1.80
Control	0 \pm 0.00
Slope	3.1345 \pm 0.6987
Chi-Square (χ^2)	3.6546 (tabulated 7.8)
Correlation Coefficient (r)	0.933 (tabulated 0.878)
LC ₅₀ (Its limits at 95%)	172.8369 (140.9869-225.0947)
LC ₉₀ (Its limits at 95%)	443.0977 (306.9969-1059.3909)

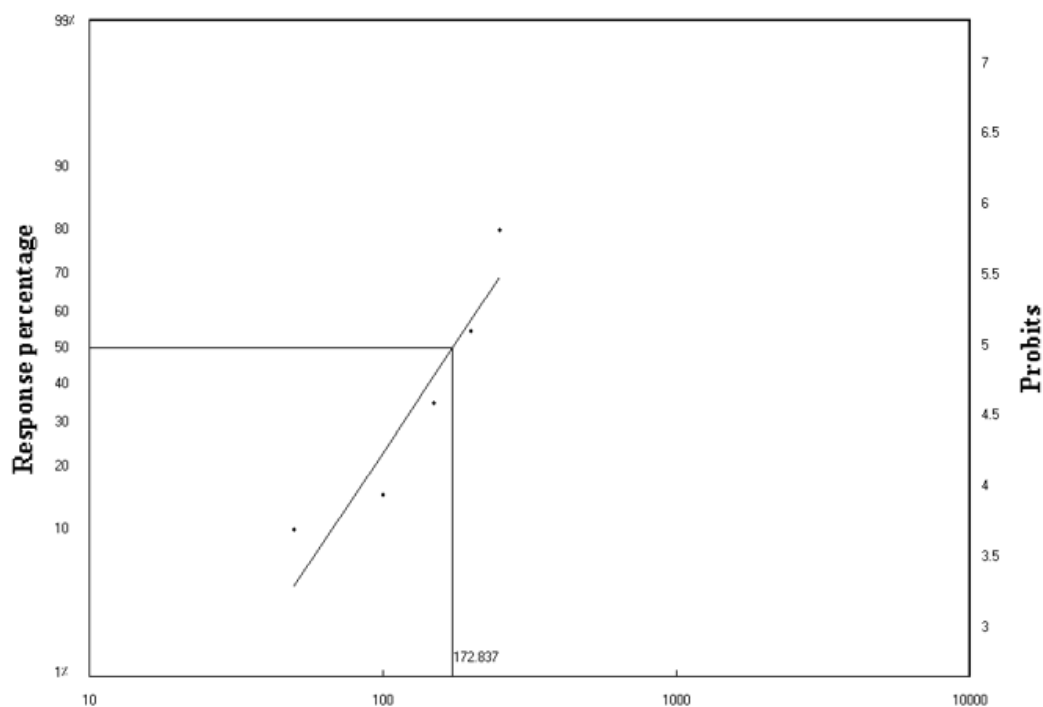


Fig 3. Regression line of chitosan-fos nanoformulations bioassay against 5th instar larvae of RPW

3. Field Application:

From the control group, no palm was cured from RPW infestation after 21, 30 and 90 days. From the group of ChNPs, only one palm was cured. From the group of chlorpyrifos, six palms out of nine were cured without any infection symptoms. From the group of Ch-Fos NF, nine palms out of eleven were cured for three months after injection. The effect of tested insecticides at one concentration against RPW using the injection method was presented in Table 2 and Figure 4.

Table 2. Curation of date palm after three months with control, chitosan nanoparticles (ChNPs), chlorpyrifos, and chitosan-chlorpyrifos nano formulation (Ch-Fos NF).

Test Result	Control	ChNPs	Chlorpyrifos (Insecticide)	Ch-Fos NF	No. of Palms	Statistical values
Uncured palms	8	7	3	2	20	$\chi^2 =$ 0.00267 P= 0.05
Cured palms	-	1	6	9	16	
Total No. of Palms	8	8	9	11	36	

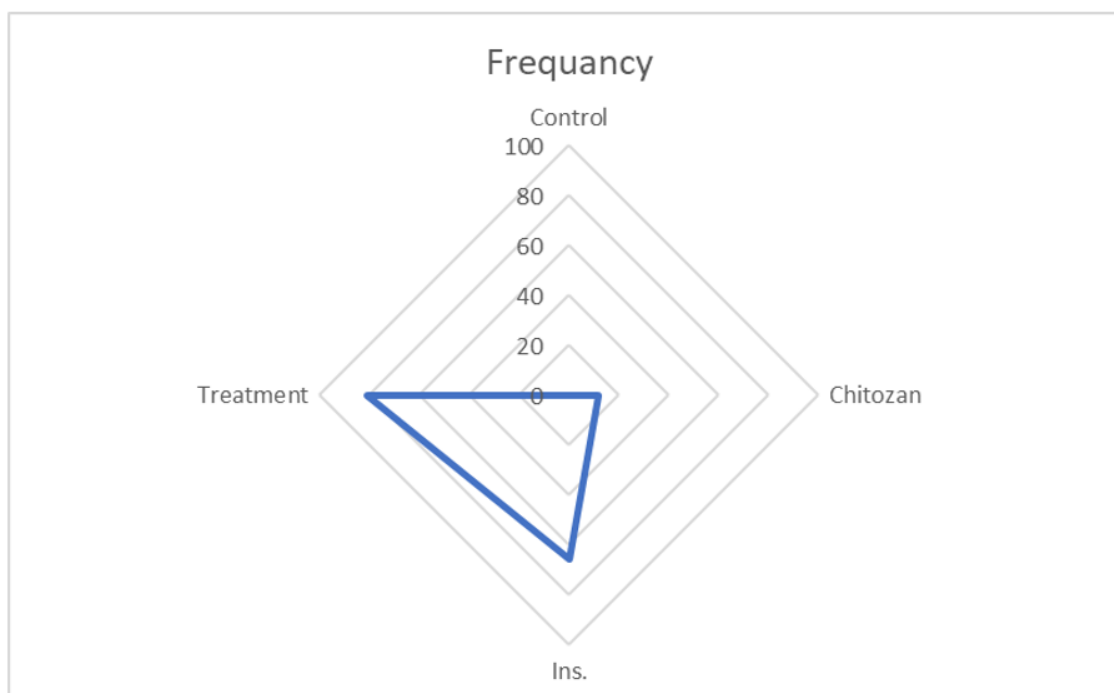


Fig. 4. Frequency of Control, Chitosan, chlorpyrifos (Ins.), Chitosan Fos nanoformulation (Treatment).

DISCUSSION

The red palm weevil is a highly destructive insect that poses a global threat to various palm species, including date palms. Its rapid spread across different regions, including the Arabian Gulf, Middle East, Southern Asia, North Africa, and the Mediterranean basin, has led to substantial losses in date palm production. The control of RPW infestations traditionally relies on chemical treatments, both preventive and curative, but these methods have limitations and drawbacks (FAOSTAT 2013).

Preventive chemical treatments, often employed on a calendar basis, can lead to pest resistance, chemical residues in fruits, resurgence of secondary pests, and environmental contamination (Dembilio and Jaques 2015). Moreover, recent research has shown that RPW is developing resistance to commonly used insecticides (Al-Sagheer and Faleiro 2011). On the other hand, curative insecticidal treatments administered early in the infestation can help mitigate the damage caused by RPW (Al-Sagheer and Faleiro 2011). However, the insecticide delivery methods need to be carefully chosen to avoid damaging the palm tissue.

The present study focused on the synthesis of chitosan chlorpyrifos nanoformulation (Ch-Fos NF) and its evaluation against the red palm weevil (RPW). To address the limitations of conventional insecticide treatments, the study explores the use of nanotechnology in developing a chitosan nanoformulation containing chlorpyrifos. Nanotechnology offers promising strategies for improving the efficiency and controlled release of active ingredients in pesticides (Habood *et al.*, 2022). The chitosan nanoformulation aims to enhance the solubility of chlorpyrifos and protect it from premature degradation, potentially increasing its efficacy in controlling RPW infestations. The primary findings suggest that the diameter of the synthesized Ch-Fos NF was less than 3 nm, indicating that the formulation was indeed in the nano range as per the EDX mapping with TEM results. These results align with the intention to create nanoformulations to increase the apparent solubility of poorly soluble active ingredients, release the active ingredient in a controlled manner, and shield against premature degradation as mentioned in previous research.

In the field application, the efficacy of Ch-Fos NF was compared with chlorpyrifos and chitosan treatments, as well as a control group. The results showed that nine out of nine palms treated with Ch-Fos NF were cured of RPW infestation three months after injection. This was a significant improvement compared to the six out of nine palms cured in the chlorpyrifos treatment group, and the single palm cured in the chitosan treatment group. Also, this method of application was performed and gave very promising results including total cure palms with a Chi-square value equal to 0.00267. Due to the nonhomogeneous data of field application, the Chi-square test was performed to test the efficacy of the tested formulation. The results illustrated that Ch-Fos NF was significantly more effective in controlling RPW infestation compared to other treatments. The present results (Abdelfattah *et al.*, 2019), who reported that nanoformulations of chlorpyrifos are effective insecticidal agents against RPW in date palm.

The results of the study are expected to provide insights into the potential of chitosan nanoformulation as an effective method for controlling RPW infestations in date palm trees. If successful, this approach could offer a more environmentally friendly alternative to conventional chemical treatments. Moreover, the use of nanotechnology in pesticide formulations has the potential for broader applications in pest control, facilitating the development of more efficient and non-persistent insecticides (Jun-Hao *et al.*, 2023).

It is important to note that while the study focuses on the efficacy of chlorpyrifos in the chitosan nanoformulation, the use of chlorpyrifos itself has been subject to regulatory actions (Tudi *et al.*, 2023). In Egypt, the Agricultural Pesticides Committee decided to ban the use of chlorpyrifos by 2023. Therefore, the study serves as an example of the potential effectiveness of a nanoformulation containing chlorpyrifos rather than a recommendation for its use.

Nanoencapsulation, a technique that involves enclosing active ingredients within nanoscale particles, shows great promise in the field of pest control. This discussion focuses on the potential of nanoencapsulation as a strategy for controlling insect pests and highlights its advantages and challenges (Habood *et al.*, 2022). Nanoencapsulation offers

several advantages in the context of pest control. Firstly, it enhances the stability and shelf-life of active ingredients by protecting them from degradation, thereby maintaining their efficacy over an extended period (Madhavi *et al.*, 2020). This is particularly important for insecticides that are prone to degradation due to environmental factors. By encapsulating the active ingredient, nanoencapsulation provides a controlled release mechanism, ensuring a sustained and prolonged effect, which can improve the effectiveness of the treatment (Nasif *et al.*, 2024).

Another advantage of nanoencapsulation is the ability to enhance the solubility of poorly soluble active ingredients. Many insecticides have limited solubility in water, which can affect their dispersal and availability to target pests. Nanoencapsulation allows for the formulation of insecticides in water-dispersible nanoparticles, improving their solubility and facilitating their application (Jun-Hao *et al.*, 2023).

Furthermore, nanoencapsulation can provide targeted delivery of active ingredients to pests. Nanoparticles can be designed to selectively release the encapsulated insecticide when in contact with the target pest or its habitat. This targeted delivery approach minimizes the exposure of non-target organisms to the insecticide, reducing the risk of environmental contamination and adverse effects on beneficial insects (Yuxia *et al.*, 2023).

Nanoencapsulation also offers the potential to reduce the amount of active ingredients required for effective pest control (Yuxia *et al.*, 2023). The encapsulation process enhances the stability and bioavailability of the active ingredient, allowing for lower concentrations to achieve the desired effect. This reduction in active ingredient usage can contribute to minimizing environmental impact and reducing the development of resistance in pest populations (Jun-Hao *et al.*, 2023).

Despite the promising potential of nanoencapsulation in pest control, some challenges need to be addressed (Jun-Hao *et al.*, 2023). One challenge is the scalability of the production process. The synthesis and large-scale production of nanoparticles with consistent quality and properties can be complex and costly. Standardization and optimization of manufacturing methods are necessary to make nanoencapsulated formulations commercially viable (Nasif *et al.*, 2024).

Another challenge is the potential toxicity and environmental impact of nanoparticles themselves. While nanoencapsulation can reduce the overall amount of active ingredient used, the nanoparticles themselves may have unintended effects on non-target organisms and ecosystems. It is crucial to conduct comprehensive toxicity assessments and environmental risk evaluations to ensure the safety and sustainability of nanoencapsulated formulations (Huijuan *et al.*, 2023).

The findings of this study are significant given the global threat posed by RPW to various palm species. The Ch-Fos NF appears to be a promising control measure, offering a more targeted and potentially environmentally friendly alternative to conventional chemical treatments. However, further research is needed to confirm these results under different conditions and to explore the potential environmental impacts of this nanoformulation.

In conclusion, the study presents evidence of the potential effectiveness of Ch-Fos NF in controlling RPW infestation. It adds to the existing body of knowledge by providing a new direction for the development of nanoformulations in pest management. This could have significant implications for the sustainable management of RPW and potentially other agricultural pests.

Declarations:

Ethical Approval: Ethical Approval is not applicable.

Competing interests: The authors declare no conflict of interest.

Authors Contributions: I hereby verify that all authors mentioned on the title page have

made substantial contributions to the conception and design of the study, have thoroughly reviewed the manuscript, confirm the accuracy and authenticity of the data and its interpretation, and consent to its submission

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Availability of Data and Materials: All datasets analysed and described during the present study are available from the corresponding author upon reasonable request.

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ARABIC SUMMERY

الفعالية الحقلية للكلوربيريفوس في تركيب كيتوزان نانوي علي سوسة النخيل الحمراء، رينكوفورس فيروجينيس (غمدية الأجنحة: كركليونيدي) في نخيل البلح

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تهدف الدراسة الحالية إلى اختبار تركيبة نانوية لمكون نشط معتمد لمكافحة سوسة النخيل الحمراء من أجل تحسين توصيله إلى RPW في أشجار النخيل الموبوءة وحمايته من التدهور المبكر. وهذا يشمل تقييم قدرتها على إيصال المكون النشط للمبيدات الحشرية المغلفة بطريقة محكمة في البيئة المائية. تمت صياغة تركيبة كيتوزان-كلوربيريفوس النانوية (Ch-Fos NF) عن طريق الموجات فوق الصوتية. تم استخدام تقنية المجهر الإلكتروني النافذ عالي الدقة وتقنية مطيافية تشتت الطاقة بالأشعة السينية لتوصيف التركيبة النانوية الناتجة. تم تطبيق سمية خمس جرعات منفصلة من تركيبة كيتوزان-كلوربيريفوس النانوية (50, 100, 150, 200, 250) جزء في المليون على يرقات العمر الخامس من سوسة النخيل الحمراء من خلال اختبار المقاييس الحيوية. كانت نسب الوفيات الملحوظة بعد 48 ساعة 10 و 15 و 35 و 55 و 80 على التوالي. تم تطبيق التركيبة النانوية على نخيل التمر ومقارنتها بالمكون النشط والناقل والكنترول. أثبت التقييم فعالية التركيبة النانوية في توصيل المبيدات الحشرية. ويلزم إجراء مزيد من الدراسة بشأن السمية النباتية والتحليل المتبعي.