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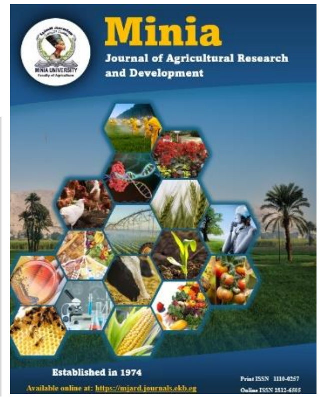

Effectiveness of Spinosad as a biogenic insecticide in controlling desert locust (*Schistocerca gregaria* Forsskål)

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1. ABSTRACT

Desert locust, *Schistocerca gregaria* (Forsskål, 1775), is the most destructive migratory pest worldwide, driven by climate change, land degradation, and regional conflicts. It forms massive swarms which devastating entire crops and causing widespread food shortages. Current control measures depend on traditional chemical insecticides, which have detrimental effects on human health and the environment. In this study, we assessed the efficacy of Spinosad, a biogenic insecticide, against desert locusts. Spinosad (Tracer® 24% SC) was applied at 65 ml/100 L against various stages of desert locust (hoppers and adults). Under field conditions, our observations revealed that Spinosad induced a mortality rate of 71% in adult and 67% in hopper instars (nymphs) at 96 hours post-treatment. Furthermore, we assessed the proportional impact of contact toxicity versus stomach poisoning induced by Spinosad on locust mortality. Our findings indicated that contact toxicity had a more pronounced effect on both adult locusts and hoppers compared to stomach poisoning. Meanwhile, adults demonstrated greater susceptibility to both modes of action than hoppers. Additionally, we evaluated the residual levels of Spinosad on treated vegetation, and the data demonstrated a rapid decrease in Spinosad concentrations on plants, starting at 4.582 mg/kg one hour after application and declining to 0.098 mg/kg by day 6 post-treatment. Considering the results obtained, Spinosad can provide effective control of desert locusts, particularly when utilized as part of a comprehensive integrated pest management program.

Keywords: Desert locust, biological control, Spinosad, CRC

2. INTRODUCTION

Food security became a concern due to population explosion and global climate change (Vermeulen et al., 2012; Molotoks et al., 2021). In this regard, agricultural pests are one of the most important threats facing mankind from ancient times where they threatened food security and human well-being (FAO, 2019). The estimated annual

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loss caused by agricultural pests in global agricultural production is about 35-45% of total production, i.e. almost half of the production (Junaid et al., 2024). Desert locust, *Schistocerca gregaria* (Forsskål, 1775), is the world's most destructive migratory pest, driven by climate change, land degradation, and regional conflicts (FAO, 2020; Tang et al., 2023). It's a highly mobile and famished eaters, where a single swarm can travel more than 90 miles a day, have 80 million locusts, and eat the same amount of food per day as 35,000 people, and as a migratory pest, desert locust invading more than 60 countries (FAO, 2024). In addition, climate change, especially global warming, affects the geographical distribution of species on earth (Meynard et al., 2017), and recent studies showed that over the past five years, the damage caused by desert locust was increased dramatically as a results of climate changes. Also, predictions indicates that the danger of desert locust may increase in the region due to the continues of these environmental consequences (Salih et al., 2020). Thus, numerous strategies are required to reduce the severity of desert locust, specially through promoting of preventive control strategy which provides means of preparation, monitoring, warnings and alerts and early intervention against desert locust infestations (Zhang et al., 2019).

Pesticide application has consistently been an effective method for controlling locusts. However, its use in agricultural systems is associated with several issues, including the mortality of non-target organisms, severe environmental pollution, and adverse effects on human health (Prior and Streett, 1997; van der Valk, 1997 and 1999). Hence, a continuous effort has been made to mitigating these negative effects by developing alternative pesticides with fewer negative impacts on the environment. These efforts include developing of contacting-killing and short-lasting insecticides such as organophosphorus, carbamates, and pyrethroids to replace 'Dieldrin' and other organochlorine pesticides (Ahmad et al., 2020). However, these compounds still have a high toxicity and causing a great damage to the environmental and ecological system (Ore et al., 2023; Gonzales et al., 2024). With increasing awareness of the negative impact of chemical pesticides on environment make it a must to find alternative means of control which are eco-friendly and sustainable, such as bio-pesticides (Van der Valk, 2006; Van Huis, 2006). When applied at their recommended dose, biological pesticides are usually characterized with their high specificity, low adverse impact on the environment, and are nontoxic to humans and livestock (Lomer et al., 2001; Kooyman, 2003). One of the successful biological measures against desert locust is the use of entomopathogenic fungi; *Metarhizium acridum* (Lomer et al., 2001), this entomopathogenic fungus has proven to be a valuable tool in managing locust outbreaks. Also, Spinosad, the biogenic insecticide has been tested to control desert locust and the African migratory locust, *Locusta migratoria migratorioides*, with promising results (Soliman et al., 2019; Mohamad et al., 2020).

Spinosad, consists of spinosyn A and spinosyn D, which are fermentation products of the soil inhabiting actinomycete *Saccharopolyspora spinosa* (Tompson et al., 2000). This naturalities biopesticide is a promising alternative to the conventional pesticides. It characterized by its high effective against many pests, lower toxicity on mammalian compared to the synthetic insecticides, poses minimum risk to humans and wildlife, quickly degradation in the environment, and with little or no effect on beneficial species (Williams and Valle, 2003). Spinosad is a neurotoxin that has both contact and stomach activity, and its mode of action is primarily through altering the function of GABA-gated chloride channels (Salgado et al., 1998; Tompson et al., 2000). However, the relative contribution of oral and contact toxicities to overall susceptibility of stored-grain insects to Spinosad is unknown (fang et al., 2002).

The Commission of Desert Locust Control in the Central Region (CRC), was established in 1967 and is the largest of the three FAO regional commissions, promotes the developing of prevention and cost-effective strategies, and introduces new technology and ecofriendly alternatives such as use of biopesticides instead of synthetic pesticides, which would enable countries to deal efficiently and effectively to save agricultural production and secures food security and human well-being against locust infestations. Therefore, and to support the development of effective alternatives to existing pesticides, in this study we aimed to assess the efficacy of Spinosad (trade name: Tracer® 24% SC), against various stages (hoppers and adults) of desert locust.

3. MATERIALS AND METHODS

3.1. Location of the study

The experiments were carried out in Wadi El-Na'am of Abu-Ramad city (23°30′ 31.4″ N/ 36°12′18 .8″ E), about 650 km South Hurghada city, Red Sea governorate, Egypt (Figure 1). This site has subtropical climate condition characterized by a hot season (May to September) with an average temperature of 31°C and a cool season (December to March) with an average temperature of 23 °C. This area known as a breeding location for desert locust, with winter breeding season extended from October to April every year. The study was conducted during the winter breeding season, from February to April 2024. The ground was >75% covered with green vegetation of *Zygophyllum simplex* herb (Figure 1). The average temperature and relative humidity during the study were 21°C±2 and 68% ± 7 respectively.

Figure 1: The location of the experiment illustrates the condition of the vegetation, with treated plots indicated by flags and wooden cages employed for confining insects post-treatment.

3.2. **Tested insecticide**

Spinosad, a natural insecticide synthesized by the soil bacterium, *Saccharopolyspora spinosa* (Thompson et al 1997 and 2000). Trade Name: Tracer 24 % SC (Suspension Concentrate), used at rate of 65 ml/100L of water.

3.3. **Field Trial**

Spinosad as Tracer® 24% SC (Corteva Agriscience, Indianapolis, USA), was applied at 65ml/100L water. One control plot, and two treatment plots each measuring 50 m× 21 m (1/4 acre), were marked out in the area, with an average distance of 100 m between plots. Control plot was upwind in relation to other plots and sprayed only with water. Plots were sprayed in early morning (wind speed: 3 m/second measured using anemometer, temperature: 23°C and relative humidity: 53%. Spraying was conducted using a backpack sprayer (air pressure Taral port 512 Knapsack mist blower, flow scale graded from 1 to 5; filled volume application rate (var) 101 L/ha. The nozzle number 4 was used with a flow rate (FR) 3.3 L/min and spraying height was 0.5 m, forward speed of 3.9 km/h, and track spacing of 5 m according to wind velocity. Each treated plot was represented by a wooden cage (1.00 m × 1.00 m × 1.00 m) (figure 1), with wire-gauze sides. Approximately 150 hoppers (nymphs) of the 4^{th} and 5^{th} instars, along with 150 adults, were captured and confined in each cage. These cages were positioned in the spray lines during application with their open side facing the spray drift. following treatment, cages were maintained in the treated area under field conditions, and locusts (hoppers and adults) were fed daily on plants from the same plot. Mortality rates were daily recorded at 24, 48, 72 and 96 hours post treatment. Each treatment was replicated twice.

3.4. **Effect of Spinosad as stomach poison**

To evaluate the effect of Spinosad as a stomach poisoning, 200 locusts (100 adults and 100 4^{th} nymphal instar) were confined in wooden cages with wire-gauze sides (50 cm × 50 cm × 60 cm) dimensions. Caged locusts were fed for 24 hours with air dried vegetation collected from treated plot one hour after spraying. Mortality was recorded at 6, 24, 48, 72 and 96 h after treatment. Locusts fed on plants treated with water were used as a control.

3.5. **Effect of Spinosad as a contact (nerve) toxin**

To assess the toxicity of Spinosad as a contact insecticide, locusts (adults and hoppers) were caged as described above. Cages were placed inside the spraying line during the application with its open side facing the spray drift according to the wind direction. Locusts were sprayed as described in the field trial (above) and left for about one hour without food (to allow air dried), before feeding on vegetations treated with water. Mortality was recorded at 6, 24, 48, 72 and 96 hours after treatment. Locusts sprayed with water were used as a control.

3.6. **Spinosad residual analysis**

Vegetative samples (about 1 kg fresh wight) of *Zygophyllum simplex* plants were collected randomly from treated and untreated (control) plots after one hour, 24 hours, 48 hours, 72 hours, and 96 hours post treatment. Samples were kept in a labeled Ziploc bags and stored in the freezer at −20 °C, in dark before sending for further analysis by the Central Laboratory of Residue Analysis and Pesticides and Heavy Metals in Food (Dokki, Giza, Egypt). The residual analysis was performed according to QuEChERS protocol (Lehotay 2005;

Payá 2007) for determination of pesticide residues in foods using LC-MSMS, GC-MSMS, and according to the European standard (EN 15662:2018).

3.7. Statistical Analysis

Mortality percentage was corrected according to Abbott's formula (Abbott, 1925). Data were analyzed by one-way ANOVA using web-based statistical software (http://vassarstats.net/anova1u.html). Means were compared using the Tukey's Honest Significant Difference Test provided by the software. Differences were considered statistically significant at $P < 0.05$.

4. RESULTS

4.1. Effect of Spinosad on Locust mortality

Spinosad Treatment significantly increased the mortality of adults and hoppers of *Schistocerca gregaria* ($p < 0.05$), as compared to water control (Table 1 and Figure 2). Corrected mortality percentage was calculated by using Abbott's formula (1925) as follows: % population reduction = [1- (post treatment population in treatment/ pretreatment population in treatment X pretreatment population in control/ post treatment population in control)] X 100. At 65 ml/ 100 L of water Spinosad caused 10.4 % mortality after 24 hours post treatment reaching to 71.5% after 96 hours post treatment for locust adults. Meanwhile, treated locust hoppers showed 20.26% mortality after 24-hour post treatment reached 66.75 % after 96 hours post treatment (Table 1 and Figure 2). Meanwhile, there were no significant difference ($p < 0.05$) between mortality rates of adult and hopper stages.

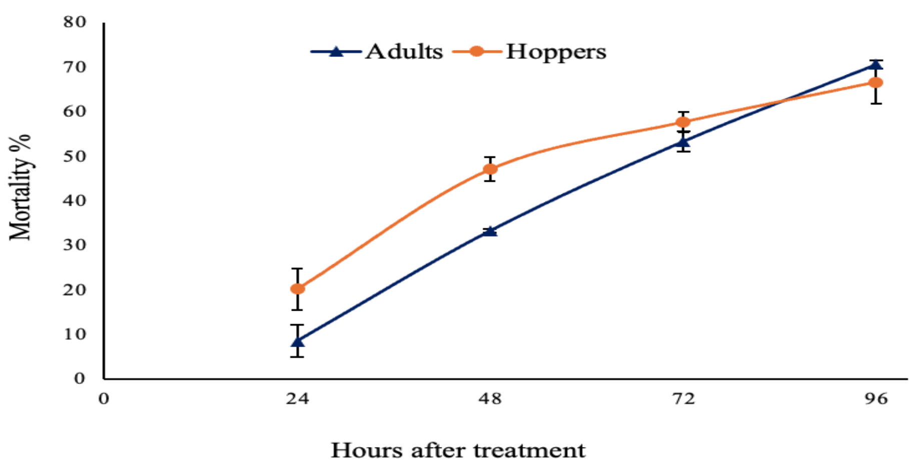

Figure 2. Lethal effect of Spinosad on *Schistocerca gregaria* (adults and hoppers) at 24,48, 72, and 96 h after treatment.

Table 1: Mortality percentage of *Schistocerca gregaria* (adults and hoppers) after 24, 48, 72 and 96 hours of treatment.

Stage	**Abbott-corrected mortality %**			
	24 h	48 h	72 h	96 h
Adults	8.7 ± 3.5	33.3 ± 0.34	53.4 ± 2.25	70.7 ± 0.9
Hoppers	20.26 ± 4.6	47.15 ± 2. 6	57.78 ± 2.28	66.75 ± 4.95

4.2. Spinosad stomach poisoning effects on locust.

Our data demonstrated that the mortality rate of desert locusts (adults and hoppers) fed on plants treated with Spinosad was significantly increased ($p < 0.05$) when compared to water control. Results shows that stomach poisoning with adults caused a mortality percentage of 2.35 %, 46.30%, and 66.3% at 6 h, 48 h, and 96 h post treatment, respectively (Table 2, Figure 3), giving almost the same mortality rate caused by the collective (contact and stomach) poisoning, (Table 1 and Figure 2). While in case of hoppers the mortality percentage recorded was 2.17% and 26.26 %, at 6 h, 48 h, and reached 49.3 % at 96 h post treatment (Table 2, Figure 3), much less than the mortality percentage of hoppers in case of collective poisoning (Table 1, Figure 2)

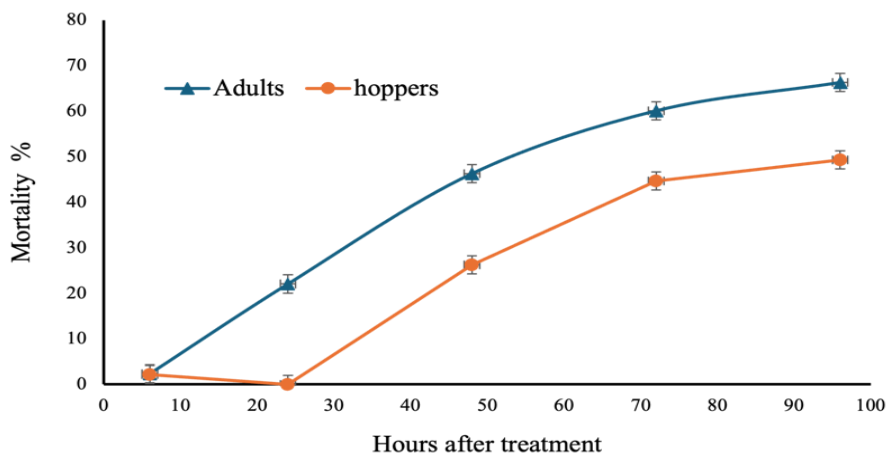

Figure 3. Impact of Spinosad as stomach poison on *Schistocerca gregaria* (adult and hoppers) after 6, 24,48, 72, and 96 h of feeding on treated vegetation.

Table 2: Toxicity of Spinosad insecticide, both as a stomach poison and through contact on *Schistocerca gregaria* (adults and hoppers of 4th and 5th instars).

Time after treatment	Abbott-corrected mortality %			
	Stomach poisoning		Contact poisoning	
	Adults	Hoppers	Adults	Hoppers
6 h	2.35	2.17	1.96	0.00
24 h	22.06	0.00	35.76	12.28
48 h	46.3	26.26	55.31	37.19
72 h	60.09	44.69	64.66	51.58
96 h	66.3	49.3	69.8	72.52

4.3. Toxicity of Spinosad as a contact (nerve) poison

Spinosad has a slower effect on desert locust than other chemical insecticide which showed an immediate effect. Thus, the purpose of this experiment is to separately evaluate the (nerve) toxicity of Spinosad as a contact poison in case treated insects flayed or moved out from the treated area to untreated places directly after treatment (without spending feeding time in the treated plots), which may happen occasionally with adult and late instars. To evaluate the toxicity of Spinosad as a contact poison, locusts (adults and hoppers) were sprayed with Spinosad during the field treatments as described above and left to dry for about 1 hour before feeding them on untreated plants. Results (Table 2, Figure 4) showed that mortality percentage of treated adults reached 69.8 % after 96 h post treatment which is slightly higher than stomach treatment (66.3 %). However, locust hoppers showed a mortality percentage of 72.5 % after 96 h of treatment, which is significantly higher than stomach treatment (49.3 %). Data also showed that there is no comparable difference between the effect of the contact poisoning alone (Table 2) and the collective poisoning (Table 1), on mortality percentage of both adult and hoppers stages.

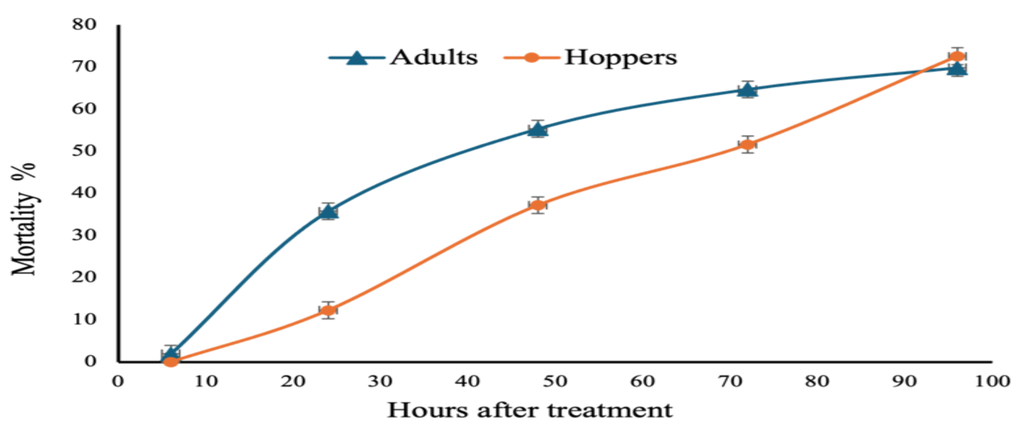

Figure 4. Effect of Spinosad toxicity administered as a contact (nerve) poison on *Schistocerca gregaria* (adults and hoppers) at 6, 24,48, 72, and 96 h after treatment.

4.4. Analysis of Spinosad residues

Spinosad residuals on sprayed vegetation (*Zygophyllum simplex*) were evaluated using QuEChERS protocol according to (Lehotay 2005; Payá 2007). Data in figure 5 showed the level of residuals at 1, 24, 48, 72 hours, and 6 days post application. Results showed a quick decline of Spinosad remains on plants starting with 4.582 mg/kg one hour after application and reaching 0.098 mg/kg at day 6 after treatment.

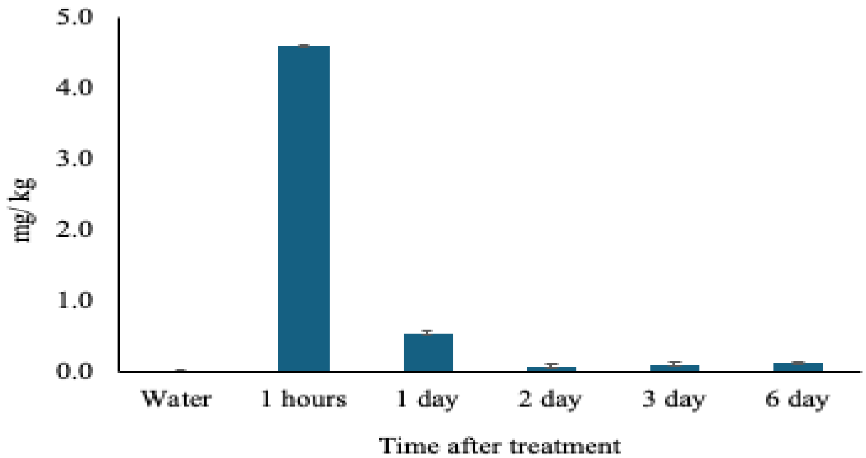

Figure 5: Spinosad residuals (mg/kg), in *Zygophyllum simplex* plants after 1 hour and 1,2, 3 and 6 days of treatment

5. DISCUSSION

Desert locust threat to the agricultural system increases in the Middle East and has extended in 2019 to 2021 to the greater horn of Africa and eastwards to India and Pakistan (FAO, 2022). Conventional insecticides, used for controlling locusts including chlorpyrifos, malathion and fenitrothion (organophosphates), deltamethrin (pyrethroid), as well as the insect growth regulator (IGR) teflubenzuron, always causes a death of non-target organisms, and have a serious negative impact on health and environment (van der Valk, H. 1997 and 1999).

The Commission of Desert Locust Control in the Central Region (CRC) always gives its utmost support for the implementation of alternative control methods that respect the environment. In this study, we run a field evaluation to assess the efficacy of Spinosad, which is a biogenic pesticide on desert locust, *Schistocerca gregaria*. The study was conducted in one of the most important breading areas for desert locust near Abu-Ramad city, Egypt. The efficacy of Spinosad under field conditions was assessed, proportional impact of contact toxicities versus stomach poisoning of Spinosad on locust was evaluated, and residues analysis of Spinosad in treated plants was measured. For all experiments, Spinosad was applied at 65 ml/100 L against various stages of locust including adults and hoppers. Under field conditions, we found that Spinosad showed 71 % mortality for adults, and 67 % for hoppers at 96 h post treatment. Our results are in consistent with some previous studies who reported that Spinosad is effective against several pests (Fang et al., 2002[a], 2002[b]; Toews and Subramanyam, 2003, Toews et al., 2003). Also, our results are comparable to the studies demonstrated by (Hosny et al., 2010), against locust where, they mentioned that Spinosad caused 75 % and 100 % mortality after 24 h and 48 h, respectively.

Obviously, when we compared the effects of Spinosad as direct (contact toxicity) and as stomach poisoning, we found that adults exhibited more sensitivity toward both modes. Data also showed that when we compared the mortality rate of each stage (adults or hoppers) with each mode of poisoning, hoppers were more sensitive to contact toxicity (72.5 %) than stomach poisoning (49.3%) at 96 h after treatment. On the other hand, adults exhibited a comparable sensitivity to both modes of actions with 66 % and 69 % of mortality with stomatic poising and contact toxicity, respectively. This difference of Spinosad efficacy on locust adults and hoppers could be referred to the amount of vegetation eaten by adults, which is also exposed to a higher amount of Spinosad on vegetation as the surface of contact (body size) is significantly higher for adults than hoppers.

In addition, the residual effect of Spinosad was assessed at time intervals starting at 1h after spraying and then daily until six days of treatment. Results indicated that residues of Spinosad on vegetation was declined by approximatively 90 % after 24 h of treatment compared to one hour after spraying. This rapid degradation of Spinosad could be advantageous when it is necessary to run treatment in cultivated areas. Otherwise, with this quick decline it is recommended that treatments should be performed in periods when locust is actively taking food, to combine the contact and stomach effects.

In conclusion, bio-pesticides derived from plants and microorganisms, such as Spinosad, are highly recommended for locust control due to their high specificity, non-toxicity to humans and livestock at recommended doses, and minimal environmental impact. However, one challenge that may limit the effectiveness of Spinosad in the field is the potential for locusts to rapidly leave the treated area during or immediately after application. This movement of locust outside of the treated zone reduces the biopesticide’s effectiveness because locusts may not accumulate a sufficient dose to be lethal. Consequently, the treated area should be sufficiently large to ensure locusts remain on contaminated vegetation long enough to acquire an adequate amount of the treatment. Additionally, as a biopesticide, Spinosad acts more slowly compared to chemical pesticides, making it inappropriate in case of upsurges and outbreaks. Overall, within the context of a preventative strategy, the use of Spinosad could significantly enhance locust control efforts by reducing pesticide applications, lowering economic costs, and minimizing environmental risks. Additional research and investigations under varying conditions in desert locust habitats are recommended to further assess the efficacy of Spinosad as part of an integrated management system for controlling desert locusts.

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الملخص العربي

فعالية المبيد الحشري (سبينوساد) كمركب حيوي،

في مكافحة الجراد الصحراوي (*Schistocerca gregaria* Forsskål)

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يعتبر الجراد الصحراوي (Schistocerca *gregaria* (Forsskål ١٧٧٥ أكثر الآفات المهاجرة تدميراً في جميع أنحاء العالم مدفوعا بتغير المناخ وتدهور الأراضي والصراعات الإقليمية. ويشكل الجراد أسراباً ضخمة تدمر المحاصيل بأكملها وتسبب نقصاً واسع النطاق في الغذاء. وتعتمد تدابير المكافحة الحالية على المبيدات الحشرية الكيميائية التقليدية، والتي لها آثار ضارة على صحة الإنسان والبيئة. في هذه الدراسة، قمنا بتقييم فعالية سبينوساد، وهو مبيد حشري حيوي، ضد الجراد الصحراوي. تم تطبيق سبينوساد تحت الاسم التجاري (Tracer® 24% SC) بجرعة ٦٥ مل/١٠٠ لتر ضد مراحل مختلفة من الجراد الصحراوي (الحوريات والحشرات البالغة). تحت ظروف الحقل، كشفت ملاحظاتنا أن سبينوساد تسبب في معدل وفيات بلغ ٧١% في الحشرات البالغة و٦٧% في أطوار الحوريات وذلك بعد ٩٦ ساعة من العلاج. وعلاوة على ذلك، قمنا بتقييم التأثير النسبي للسمية التلامسيه مقابل التسمم المعوي الناجم عن سبينوساد على معدل وفيات الجراد. وأشارت نتائجنا إلى أن السمية التلامسيه كان لها تأثير أكثر وضوحًا على كل من الجراد البالغ والحوريات مقارنة بالتسمم المعوي. وفي الوقت نفسه، كان الجراد البالغ اكثر حساسية لكلا التأثيرين مقارنة بالحوريات. بالإضافة إلى ذلك، قمنا بتقييم المستويات المتبقية من سبينوساد على النباتات المعالجة، وأظهرت البيانات انخفاضًا سريعًا في تركيزات سبينوساد على النباتات، بدءًا من ٤.٥٨٢ مجم / كجم بعد ساعة واحدة من المعالجة وانخفاضًا إلى ٠.٠٩٨ مجم / كجم بحلول اليوم السادس بعد العلاج. بالنظر إلى النتائج التي تم الحصول عليها، يمكن أن يعطي مركب سبينوساد مكافحة فعالة للجراد الصحراوي، وخاصة عند استخدامه كجزء من برنامج مكافحة متكاملة.