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RESEARCH ARTICLE

Evaluation of different insecticides against the citrus leafminer (*Phyllocnistis citrella* Stainton) (Lepidoptera: Gracillaridae) on citrus seedlings in Sudan

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Abstract

The citrus leafminer (*Phyllocnistis citrella*), has become an important pest in orchards and nurseries of citrus fruits throughout the Sudan. Hence, various insecticides were screened in this study against *P. citrella* infesting lemon and grafted grapefruit seedlings in Khartoum State. The experiments were conducted in nurseries during 2011 and 2012. The evaluated chemicals were: foliar sprays of Tracer 480 SC (spinosad), Karate 5% EC (lambda cyhalothrin) and Nembicedine 0.03% EC (azadirachtin) as well as soil drench of Actara 25 WG (thiamethoxam) and Confidor 200 SL (imidacloprid). All the studied insecticides were effective in reducing leafminers infestation at variable levels compared with the untreated control. Regarding foliar sprays, Tracer 480 SC proved the best significant results on lemon seedlings for two weeks, followed by Karate 5% EC and Nembicedine 0.03% EC. Lemon and grafted grapefruit seedlings treated with Actara 25 WG and Confidor 200 SL were almost free from miners attack for more than a month, indicating the superiority of these two soil applied chemicals over those sprayed. Thus, both insecticides can be used for citrus seedlings. However, the performance of such chemicals on orchard trees of different citrus species should be evaluated before concluding on their wide scale usage.

INTRODUCTION

Sudan offers great potentialities for citrus production due to the wide variability in climatic, soil and geographical situations. Although, large scale citrus production is not yet fully established, the recent trend in agricultural sector reveals genuine interests to expand the area of horticultural crops in the near future. The important citrus species grown in different parts of the country may include; baladi (local) lime *Citrus aurantifolia* L., grapefruit *Citrus paradisi* Macf., sweet orange *Citrus sinensis* (L.) Osb., lemon *Citrus limon* (L.) Burm. and mandarin *Citrus reticulata* Blanco. The bulk of production is consumed locally and little amount generally exported to some Arabian and European countries. Various citrus pests and diseases have been reported in different growing areas of the country (Badawy, 1967; Schmutterer, 1969; Giha, 1987). However, among important insect pests of citrus orchards are; Mediterranean fruitfly (*Ceratitis capitata*), citrus leafminer (*Phyllocnistis citrella*), California red scale (*Aonidiella aurantii*), the citrus mealybug (*Planococcus citri*) and lemon butterfly (*Papilio demodocus*).

The citrus leafminer (*Phyllocnistis citrella* Stainton) (Lepidoptera: Gracillariidae, Phyllocnistinae), is a serious pest of citrus and related species of the plant family Rutaceae. It was firstly described from Calcutta, India, in 1856 (Stainton, 1856), and then widely distributed in citrus-growing regions of Asia, Australia, and East Africa (e.g., Sudan and Ethiopia). However, the citrus leafminer is considered as a major pest of almost all citrus orchards in Sudan (Badawy, 1967; Schmutterer, 1969). Although, is listed among alien pest species in the country, the exact

route of entry and detection time of this leafminer is uncertain (Satti, 2011). In the last two decades, such leafminer has invaded most of citrus-producing regions of the world and considered one of the major citrus pests globally (Badawy, 1967; Heppner, 1993; Garcia-Marí *et al*., 1997). The pest damages new leaf flushes and adversely affects plant health and fruit development. The economic impact of direct damage is particularly highest on nursery stock where cosmetic appearance is important, and on young trees three years of age or less and grafted seedlings where significant reduction of photosynthetic surface area can stunt growth (Grafton-Cardwell and Montez, 2009). Severe damage to young trees may result in delayed maturity of 1 to 2 years or kill plants. Moreover, larval feeding reduces the photosynthetic capacity of leaves and increases the susceptibility of leaves to citrus bacterial canker, *Xanthomonas axonopodis* pv. *citri* (Hill, 1918; Gottwald *et al*., 2007).

Economic losses are reflected by the increased cost of protecting nurseries and non-bearing citrus trees. Chemical control is the main tactic adopted for citrus leafminer control in all continents. Broad-spectrum insecticides (pyrethroids, carbamates and organophosphates) are generally used against *P. citrella* in different countries (Zhang *et al.*, 1994). Similarly, in many citrus growing areas in Sudan, containment of this pest depends almost entirely on the application of chemical insecticides. Despite the fact that, biological control is mainly ecologically sound and can also be economically feasible, very meager effort is currently devoted to this approach as compared with that given to insecticidal control, especially in the third world countries. Since encouraging results are available regarding the diversity of agricultural natural enemies in Sudan including some notes on bio-agents of citrus pests (Satti *et al*., 1998; Bilal and Satti, 2012; Satti and Bilal, 2012; Satti and Abdalla, 2014), as well as potent findings on indigenous botanical extracts and entomopathogenic microbial isolates (Satti and Nasr, 2006a,b; Satti *et al*., 2010; Satti and Gorashi, 2013), more emphasis should be paid for proper exploitation of these resources so as to minimize the negative pressures of synthetic chemicals on different traits of the environment. Nevertheless, chemical control of citrus leafminers in Florida was reported to increase yield in 3- to 5- year-old grapefruit or orange trees by 13.1% to 16.9% (Stansly *et al*., 1996). Chemical products that contain natural insecticides like azadirachtin (neem product) or spinosad showed some efficacy against citrus leafminer larvae, whereas insecticides applied to the ground for young trees or to the soil of potted citrus provides the longest period of control (1-3 months) (Hyland, 2009). The objectives of this study were to evaluate the efficacy of some insecticides against the citrus leafminer infesting lemon and grapefruit nurseries in Khartoum State, Sudan.

Material and Methods

The efficacy of some commercial insecticides was tested against the citrus leafminer in nurseries at Shambat Research Station and Horticultural Sector Administration, Khartoum State, Sudan, in seasons 2011 and 2012. Three experiments were firstly followed on lemon (*Citrus limon* L.) seedlings. Seedlings of lemon about 0.5 m tall growing in plastic containers were initially kept in screen cages to protect them from attack by citrus leafminer. All seedlings were fertilized with NPK applied to the soil several weeks before the experiment to encourage new growths. At the time of each experiment, the seedlings were removed from the cages and placed near a citrus orchard infested with citrus leafminer to enable free infestation after treatments. The chemicals sprayed during each season were: two foliar sprays (14 days apart) with Karate 5% EC (lambda cyhalothrin), Tracer 480 SC (spinosad), each at 1 ml and 1.5 ml/liter of water, and Nembicedine 0.03% EC (azadirachtin) at 4 ml/litre of water. Upper and lower leaf surfaces were sprayed to run-off using a 2-liter hand sprayer. These insecticides were compared with Actara 25 WG (thiamethoxam) (at 0.5g and 1g per liter of water applied once as soil drench with 50 ml per seedling from each dosage rate) and the untreated control. Seedlings of lemon were examined for leafminer damage at weekly intervals after each spraying. The percentages of mined leaves per plant were determined for the different treatments.

In another experiment during 2012 the performance of soil drenches of two neonicotinoids, Actara 25WG and Confidor 200 SL (imidacloprid), in combating leafminer on grapefruit grafted seedlings was tested in Horticultural Sector Administration Nursery. Seedlings grafting was done on August 26, 2012 and the insecticides were applied to the soil on September 6, 2012 at 50ml from each dosage rate/liter of water. The first count of mined leaves per grafted seedling (percentage of damage) was recorded on September 26, 2012, and weekly thereafter for three weeks.

All experiments were laid out in a Randomized Complete Block design, with four replicates and ten seedlings per each replicate. Rows were about 2.5 m apart and each 2 m-long plot of seedlings within rows was separated by 0.5 m. Numbers of leaves with advanced mines per seedling were transformed to $\sqrt{x} + 0.5$ before analysis of data (ANOVA) and computation of means separation through Duncan's Multiple Range Test.

Result and Discussion

The results regarding the evaluation of four insecticides in controlling citrus leafminers on lemon seedlings were presented in Tables 1, 2 and 3 as obtained from three experiments conducted during 2011 and 2012 seasons. Variable levels of activities mostly with significant reduction in infestations were reported by all chemicals compared with the untreated control. However, the soil applied Actara 25WG (thiamethoxam) gave the best significant results, followed by the sprays; Tracer 480 SC (spinosad), Karate 5% EC (lambda cyhalothrin) and lastly Nembicedine 0.03% EC (azadirachtin) which revealed the highest leafminer damage. Howard (1994) reported that the azadirachtin prevent damage by citrus leafminers when sprayed on leaves prior to oviposition period. Therefore, further studies on time and frequency of application by such neem product should be conducted.

Table 1. Percentages of mined leaves per lemon seedlings at weekly intervals following two sprays of three insecticides compared with Actara soil drenching, during September – November 2011, at Khartoum State.

Treatments	Mean % of mined leaves/ lemon seedling at two intervals after sprayings			
	1st spray		2nd spray	
	1st week	2nd week	1st week	2nd week
Karate 5% EC, 1ml/l	(12.1)3.1b	(15.1)3.9b	(11.8)3.5b	(8.4)3.0b
Karate 5% EC, 1.5/l	(12.8)4.6ab	(19.6)4.7c	(11.7)3.4b	(10.4)3.3b
Tracer 480 SC, 1ml/l	(8.8)2.7a	(0.0)0.7a	(3.0)1.1a	(2.6)1.9ab
Tracer 480 SC, 1.5ml/l	(6.7)2.4ab	(1.1)1.1a	(1.8)1.1a	(1.5)1.9ab
Nimbecidine 0.03% EC, 4ml/l	(11.9)3.1a	(2.0)1.2a	(2.9)1.6a	(7.4)2.0b
Actara 25WG, 0.5ml/l	(0.0)0.7a	(0.0)0.7a	(0.0)0.7a	(0.0)0.7a
Actara 25WG, 1ml/l	(0.0)0.7a	(0.0)0.7a	(0.0)0.7a	(0.0)0.7a
Untreated control	(46.5)6.8b	(77.4)8.8d	(71.7)8.3c	(67.8)8.3c
SE±	1.0	0.2	0.3	0.4
C.V.%	57	15	22	26

Means followed by the same letter(s) in each column are not significantly different ($P > 0.05$).
Means in parenthesis are actual values transformed to $\sqrt{x} + 0.5$.

Table 2. Percentages of mined leaves per lemon seedlings at weekly intervals following two sprays of three insecticides compared with Actara soil drenching, during September – November 2012, at Khartoum State.

Treatments	Mean % of mined leaves/ lemon seedling at two intervals after sprayings			
	1st spray		2nd spray	
	1st week	2nd week	1st week	2nd week
Karate 5% EC, 1ml/l	(2.8)1.5a	(13.1)3.6b	(9.8)3.0b	(6.3)2.5ab
Karate 5% EC, 1.5/l	(5.6)2.3a	(3.7)1.8ab	(0.8)1.0a	(10.2)3.3b
Tracer 480 SC, 1ml/l	(1.4)1.8a	(3.5)1.8ab	(0.0)0.7a	(2.7)1.7ab
Tracer 480 SC, 1.5ml/l	(2.4)1.4a	(2.6)1.6ab	(1.1)1.1a	(0.0)0.7a
Nimbecidine 0.03% EC, 4ml/l	(2.8)1.4a	(42.6)6.5c	(36.7)6.0c	(40.0)6.2d
Actara 25WG, 0.5ml/l	(2.8)1.4a	(1.2)1.2a	(1.0)1.1a	(0.0)0.7a
Actara 25WG, 1ml/l	(2.2)1.5a	(1.5)1.2a	(0.0)0.7a	(0.0)0.7a
Untreated control	(24.6)5.0b	(52.8)7.3c	(59.5)7.7d	(71.6)8.5e
SE±	0.7	0.7	0.5	0.5
C.V.%	57	37	31	28

Means followed by the same letter(s) in each column are not significantly different ($P > 0.05$).
Means in parenthesis are actual values transformed to $\sqrt{x} + 0.5$.

Table 3. Percentages of mined leaves per lemon seedlings at weekly intervals following two sprays of three insecticides compared with Actara soil drenching, during September – November 2012, at Khartoum State.

Treatments	Mean % of mined leaves/ lemon seedling at two intervals after sprayings			
	1st spray		2nd spray	
	1st week	2nd week	1st week	2nd week
Karate 5% EC, 1ml/l	(13.3)3.5c	(7.9)2.8c	(6.9)2.7c	(11.5)3.4de
Karate 5% EC, 1.5/l	(2.9)1.8b	(9.0)3.0c	(5.7)2.4c	(7.4)2.7cd
Tracer 480 SC, 1ml/l	(0.0)0.7a	(2.4)1.3ab	(2.1)1.5ab	(3.3)1.9b
Tracer 480 SC, 1.5ml/l	(0.0)0.7a	(3.3)1.9bc	(3.4)2.0bc	(4.0)2.1bc
Nimbecidine 0.03% EC, 4ml/l	(19.0)4.4c	(6.1)2.5c	(14.2)3.8d	(14.5)3.9e
Actara 25WG, 0.5ml/l	(0.0)0.7a	(0.0)0.7a	(0.0)0.7a	(0.0)0.7a
Actara 25WG, 1ml/l	(0.0)0.7a	(0.0)0.7a	(0.0)0.7a	(0.0)0.7a
Untreated control	(38.3)6.2d	(36.4)6.1d	(36.6)6.1e	(37.6)6.2f
SE±	0.3	0.4	0.3	0.3
C.V.%	23	27	20	16

Means followed by the same letter(s) in each column are not significantly different ($P > 0.05$).
Means in parenthesis are actual values transformed to $\sqrt{x} + 0.5$.

Moreover, the two soil applied chemicals, Actara 25WG and Confidor 200 SL (imidacloprid), also proved very potent results in combating leafminers significantly on grafted grapefruit seedlings which appeared nearly free of damage (Table 4). The results of the two latter insecticides on grapefruit seedlings were almost similar to what had been obtained by Actara on lemon seedlings, as mentioned above. These findings indicated that soil applied with such neonicotinoid insecticides can prevent damage by citrus leafminers for more than one month, which was longer than that obtained by the Tracer 480 SC spraying. Setamou *et al*. (2010), Sharma and Bhatti (2012) and Stansly *et al.* (2012) stated that drenches of imidacloprid and thiamethoxam can suppress the same pest on citrus seedlings and young trees for one to three months. An advantage is that both spinosad spraying and neonicotinoids incorporation in soil are reported to be safe or with little direct effect on the leafminer's natural enemies (Rogers *et al*., 2014).
As achieved in the present work, soil applications with Actara and Confidor proved effective against the citrus leafminer and gave prolonged control effects for at least one to three months. The soil application technique is seemed to be advantageous over foliar sprays because there is a reduction in the frequency of application in citrus nurseries and orchards resulting in minimum exposure of such broad spectrum insecticides to beneficial insects. Moreover, the applications of foliar insecticidal products which contain natural insecticides like azadirachtin or spinosad proved to show some efficacy against leafminers, and at the same time expected to be non harmful to natural enemies as shown in some literature (Satti *et al*., 2003; Satti and Nasr, 2006a,b). Also, the residues of these insecticides seemed to be easily degraded as they need to be reapplied every 7 to 14 days to ensure good control of leafminers for 15 days. Further tests are to be carried out to confirm the efficacy of neonicotinoids, spinosad and neem products against leafminers on citrus trees. The effects of such chemicals on fruit yields of different citrus species, as well as their impacts on natural enemies of citrus pests, should be emphasized in future works as a prerequisite step for their wide adoption in commercial orchards.

Table 4. Percentages of mined leaves per grapefruit seedlings at weekly intervals starting 20 days after soil application of two neonicotinoid insecticides at different rates, during August – Oct. 2012, at Khartoum State.

Treatments	Mean % mined leaves/ grapefruit seedling at three intervals from treatments		
	20 days	27 days	34 days
Actara 25WG, at 0.5ml/l	(0.0)0.7a	(0.0)0.7a	(0.0)0.7a
Actara 25WG, at 1ml/l	(0.0)0.7a	(0.0)0.7a	(0.0)0.7a
Actara 25WG, at 1.5ml/l	(0.0)0.7a	(0.0)0.7a	(0.0)0.7a
Confidor 200 SL, at 1.5ml/l	(0.0)0.7a	(0.0)0.7a	(0.0)0.7a
Confidor 200 SL, at 2.0ml/l	(0.0)0.7a	(0.0)0.7a	(0.0)0.7a
Confidor 200 SL, at 2.5ml/l	(0.0)0.7a	(0.0)0.7a	(0.0)0.7a
Untreated control	(41.7)b	(38.2)b	(41.1)b

Means followed by the same letter in each column are not significantly different ($P > 0.05$).
Means in parenthesis are actual values transformed to $\sqrt{x} + 0.5$.

Conclusion

The results proved the superior effects of two systemic neonicotinoid insecticides over the tested foliar sprays of spinosad, lambda cyhalothrin and azadirachtin products in controlling leafminers on lemon and grafted grapefruit seedlings. The two former soil applied insecticides were expected to be safe for natural enemies of citrus pests, a character that needs to be confirmed in additional research, together with their efficacy against leafminers on different species of citrus orchards.

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