

Efficiency of Different Application Techniques of Three Neonicotinoids Insecticides against Florida Wax Scale, *Ceroplastes floridensis* Comstock and Its Parasitoid, *Coccophagus scutellaris* (Dalman)

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Abstract: Field experiments were conducted to evaluate the efficiency of three different application techniques of three neonicotinoids insecticides against the Florida wax scale (FWS), *Ceroplastes floridensis* Comstock (Hemiptera: Coccidae) and its parasitoid, *Coccophagus scutellaris* (Dalman). The experiments were achieved in a private farm at Nobaria district, Beheira Governorate, Egypt during the spring seasons of 2019 and 2020. For ten weeks, the leaves samples were taken weekly. The results show that, among the different methods of insecticide applications, trunk injection technique achieved the highest reduction percentages for both adults and nymphs stages of FWS, followed by soil drench and finally foliar spray technique. However among different methods of application, trunk injection application maintained more *C. scutellaris* population (low reduction percentages %) than the two other methods (soil drench and foliar spray). In another hands, among insecticides, imidacloprid was the most effective and caused significant reduction in the population of the FWS and its parasitoid followed by acetamiprid and finally dinotefuran. Therefore, these results concluded that the trunk injection was the most appropriate technique for control *C. floridensis* and could be recommended in integrated pest management programs.

Keywords: Trunk injection, soil drench, foliar spray, neonicotinoids insecticides, *Ceroplastes floridensis*

INTRODUCTION

Florida wax scale, *Ceroplastes floridensis* Comstock (Hemiptera: Coccidae) is one of the most dangerous citrus pests (Ben-Dov, 1993; Eserkaya and Karaca, 2016; Zaki, *et al.*, 2019). The different stages of this insect feed on the leaves and twigs of its hosts. Severe infestation leads to dropping of leaves and drying the twigs. The parasitoid, *Coccophagus scutellaris* (Dalman) (Hymenoptera: Aphelinidae) is a cosmopolitan parasitoid of various species of soft scale insects including *C. floridensis* (Rosen, 1967; Nikol'skaja, 1952; Abd-Rabou, 2001). Chemical control of scale insects is relatively difficult, because the waxy layer which cover and protect them from contact insecticides. (Radwan, 2003; Elekçioğlu and Şenal, 2007; Abo-Shanab, 2012; Siam and Othman, 2020). Furthermore many studies show that the conventional application methods of chemical insecticides such as foliar spray are relatively inefficient techniques of delivering insecticides to their target. The deposited of solution of the applied spray on the tree canopy ranged from 29 to 56 % of the total solution. The remaining product wasted, either to drift, run off, or because it can not be applied precisely to where it is needed. (Perry *et al.*, 1991; Zhu *et al.*, 2006). Despite of the rapidly evolved of the insecticides groups which used in fruit production, the application techniques of these insecticides have changed very little (VanWoerkom, 2012). Trunk injection is a method of target precise application of insecticides. The trunk injection system directly injects the insecticides into the trees vascular system without exposed it to the environment conditions. As compared with traditional methods (such as foliar sprays) the trunk injection which also known stem injection is relatively simple, quick and require less active ingredient (Norris 1965; Doccola *et al.* 2009;

VanWoerkom, 2012; Doccola and Wild, 2012; Bhandari and Cheng, 2016). Soil drench methods involve placing insecticides in liquid formulation near the roots in the soil for root uptake. Keeping in view the above mentioned facts, the objective of this work was to evaluate the efficacy of different applications techniques of three neonicotinoid insecticides against FWS, *C. floridensis* and its parasitoid *C. scutellaris* in navel orange trees (*Citrus sinensis* L.).

MATERIALS AND METHODS

The study was carried out in a private farm of navel orange *Citrus sinensis* (L.) grafted on Rangpur lime (*Citrus limonia* L. Osbeck) at Nubaria district, Beheira Governorate, Egypt during two consecutive seasons, 2019 and 2020. The trees were 15 years old and spaced 5 by 5 m apart. Three different application techniques of the selected insecticides (acetamiprid, imidacloprid, dinotefuran) were evaluated against the citrus wax scale, *C. floridensis* and its parasites, *C. scutellaris*.

The methods of applications of the selected insecticides:

1. Foliar spray

The trees were sprayed with a rate of 5-6 liters / tree with conventional hand knapsack sprayer to ensure complete coverage of all parts of the trees with selected insecticides.

2. Trunk injection

The trunk injections were made with the help of a metal drill on approximately 25 cm of the average trees diameter (in areas of healthy and active wood), 30 cm above ground level and on a hole of 5 cm deep were bored into the trunk at an angle of 45°. The injection volume was made up to 10 ml using tap-water

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3. Soil drench

Soil drenching was the processed of adding diluted insecticides directly to the soil at the base of the trees. The rate of application was of 5-6 liters of each tested insecticides dilution / tree. The specific (recommended) doses of the different insecticides were diluted with water in a container and drenched around

the navel orange trees within 20-50 cm of the tree trunk and then the trees were irrigated.

Insecticides:

Tested insecticides trade names, percentage of active ingredients, formulation types and application rates are listed in Table 1. The insecticides concentrations used in this study were based on the labeled recommendation rate.

Table (1): Descriptions of the insecticides used against the Florida wax scale and its parasitoids.

Active ingredient	Trade Name	Rate	(a.i.) % and formulation	Recommended rate	Manufacturer
Acetamiprid	Mospilan®	350 gm./ Fad.	20% SP	25mg L ⁻¹	Nippon Soda Ltd.
Dinotefuran	Ochin®	25gm./100 L	20% SG	50mg L-1	Mitsui Chemicals
Imidaclopride	Confidor®	30 cm / 100L	20% SC	0.5ml L-1	Bayer CropScience

Insects Count

Randomly samples consisted of 100 leaves from each treatment were divided into two parts. The 1st portion (ten leaves from each tree or replicate) was inspected before and weekly for 10 weeks post treatment. The scales were carefully examined under binocular microscope and the number of living and dead adult females and immature stages were recorded. Dead scales were discolored, shriveled, gradually fall off the leaves. Squishing the live scales will produce a liquid, while dead scales will be dry when crushed. In the 2nd portion, after the examined of leaves, the sample scales were counted, the samples placed in glass jars and kept in the laboratory for securing any emerging parasitoids. For ten weeks, the samples were weekly taken. For each application technique, the efficiency of the insecticides as the general means of reduction percentages of the adults and nymphs of FWS and its parasite was calculated according to the formula of Henderson and Tilton (1955):

$$\text{Reduction percentage} = 100 * [1 - (T2 R1/T1 R2)]$$

Where:

R1 = number of alive scales (or its parasite) before treatment in untreated plots.

R2 = number of alive scales (or its parasite) after treatment in untreated plots.

T1 = number of alive scales (or its parasite) before treatment in treated plots.

T2 = number of alive scales (or its parasite) after treatment in treated plots.

Statistical analysis:

The experiment was performed as a factorial in the form of Randomized Complete Block design (RCBD), with five replicates (trees) per treatment and untreated control (fifty trees). Water was used as control treatment. Data were subjected to the statistical analysis according to the technique of analysis of variance (ANOVA) as published by Gomez and Gomez (1984) by using "MSTAT-C" computer software package. Least significant of difference

(LSD) method was used to test the differences between treatment means at 5% level of probability as described by Snedecor and Cochran (1980).

RESULTS

Effect of application methods

The results indicated that there are significant differences between the different application methods as well as among the insecticides treatments. As shown in Figs (1 - 6), the trunk injection technique was the most effective application method for combat both of adults and nymphs of *C. floridensis* by using each of imidacloprid, dinotefuran and acetamiprid. By using the imidacloprid, the trunk injection method led to reduction percentages of 82.34 and 84.73% for the adults of *C. floridensis* during the consecutive seasons, 2019 and 2020, respectively. While these percentages recorded 79.64 and 81.92% for the nymphs for the same seasons, respectively. On the other hand, the least effective method in combat this scale insect with imidacloprid was the foliar spray method (54.26 and 57.54% in 1st season and 49.62 and 58.47% in 2nd season for adults and nymphs, respectively). Statistically analysis of variance of data histogramatically illustrated in Figs. (3 and 4) revealed that the highest mortality rates of adults and nymphs of *C. floridensis* were achieved using the acetamiprid insecticide throughout trunk injection technique (73.27 and 76.03% in the 1st season, 2019 and 73.69 and 76.7 in the 2nd season, respectively). Otherwise, the foliar spray methods achieved the least reduction percentages of adults (45.36 and 43.49%) and nymphs (49.87 and 52.75%) during the two successive seasons. The same trends were observed by using the dinotefuran insecticide whereas the reduction percentages recorded 65.15 and 75.27% in the 1st season and 67.47 and 71.92% in the 2nd season of the adults and nymphs of *C. floredensis* when the trunk injection was applied. While the soil drench ranked the second and finally foliar spray method.

To study the appropriate insecticide applied method against the populations of the parasitoids, *C. scutellaris* (Dalman), the reduction percentages of the parasitoid, *C. scutellaris* population were compared. The results revealed that the most effective appropriate method (the lowest reduction percentages) was the

trunk injection method (Figs 1-6). Statistically analysis of variance of data histogrammatically illustrated in Figs (1 and 6) revealed that the foliar spray was the highly effective methods in reduction *C. scutellaris*. Therefore, this method is not recommended in the integrated pest management programs of *C. floridensis*.

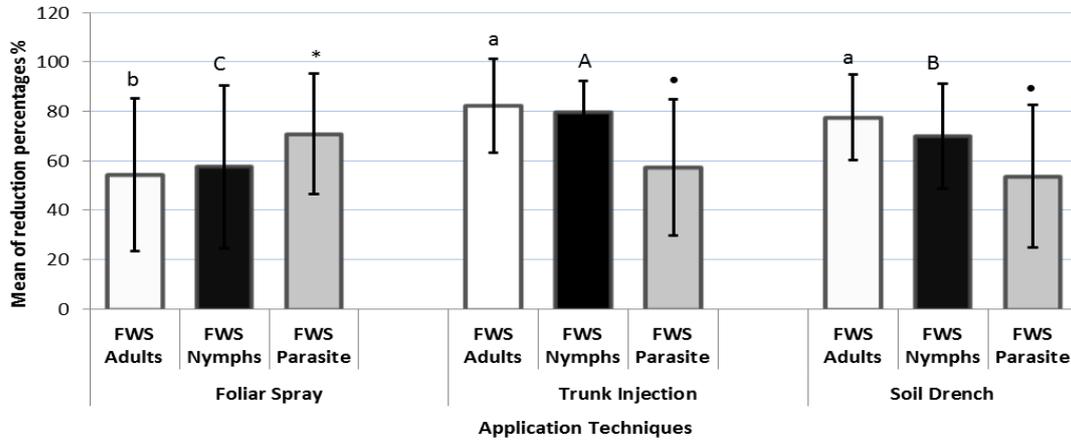


Fig. (1): The general means of reduction percentages of adults and nymphs of Florida wax scale (FWS) and its parasitoid by Imidacloprid insecticide throughout different application techniques in the 1st season, 2019. (F adult = 21.016, df = 149, L. S. D. = 9.17095; F nymphs = 10.811, df = 149, L. S. D. = 9.41315; F parasite = 5.606, df = 149, L. S. D. = 10.713)

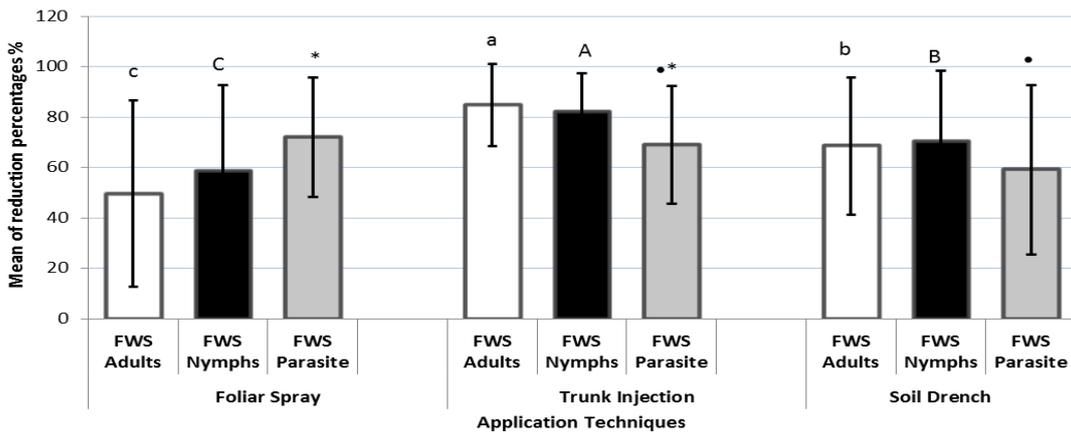


Fig. (2): The general means of reduction percentages of adults and nymphs of Florida wax scale (FWS) and its parasitoid by Imidacloprid insecticide throughout different application techniques in the 2nd season, 2020. F adult = 19.531, df = 149, L. S. D. = 11.11245 F nymphs = 9.381, df = 149, L. S. D. = 10.69875 F parasite = 3.061, df = 149, L. S. D. = 10.77475

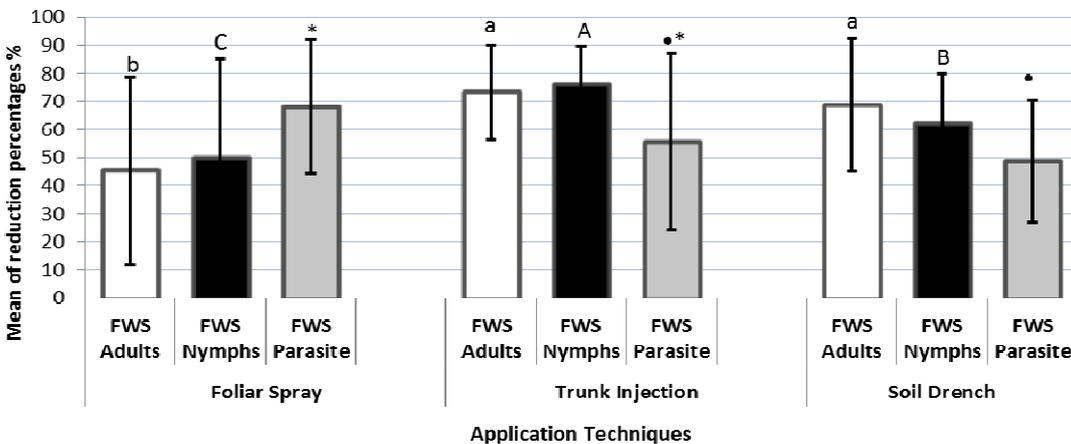


Fig. (3): The general means of reduction percentages of adults and nymphs of Florida wax scale (FWS) and its parasitoid by acetamiprid insecticide throughout different application techniques in the 1st season, 2019. F adult = 17.190, df = 149, L. S. D. = 10.09545 F nymphs = 14.779, df = 149, L. S. D. = 9.52055 F parasite = 7.034, df = 149, L. S. D. = 10.26425

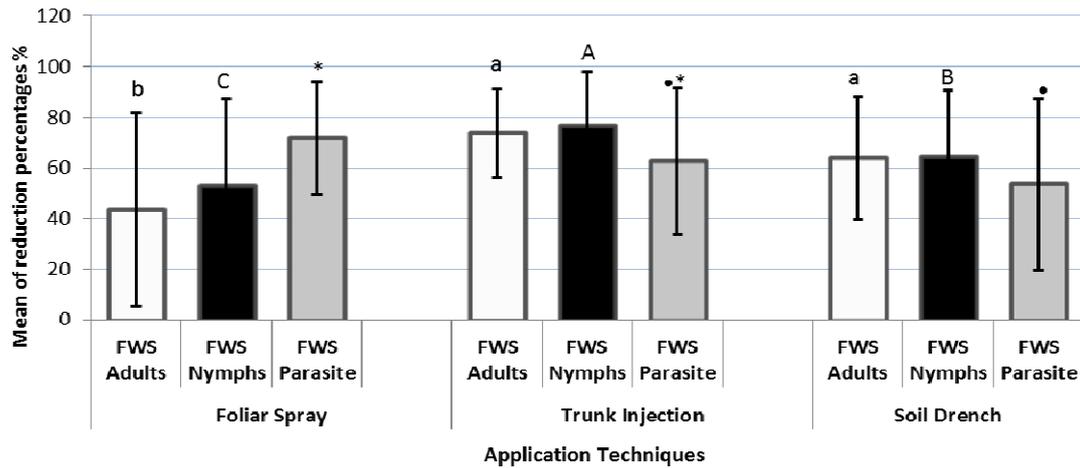


Fig. (4): The general means of reduction percentages of adults and nymphs of Florida wax scale (FWS) and its parasitoid by acetamiprid insecticide throughout different application techniques in the 2nd season, 2020. F adult = 15.185, df = 149, L. S. D. = 11.05395 F nymphs = 9.191, df = 149, L. S. D. = 11.04185 F parasite = 5.022, df = 149, L. S. D. = 11.3292

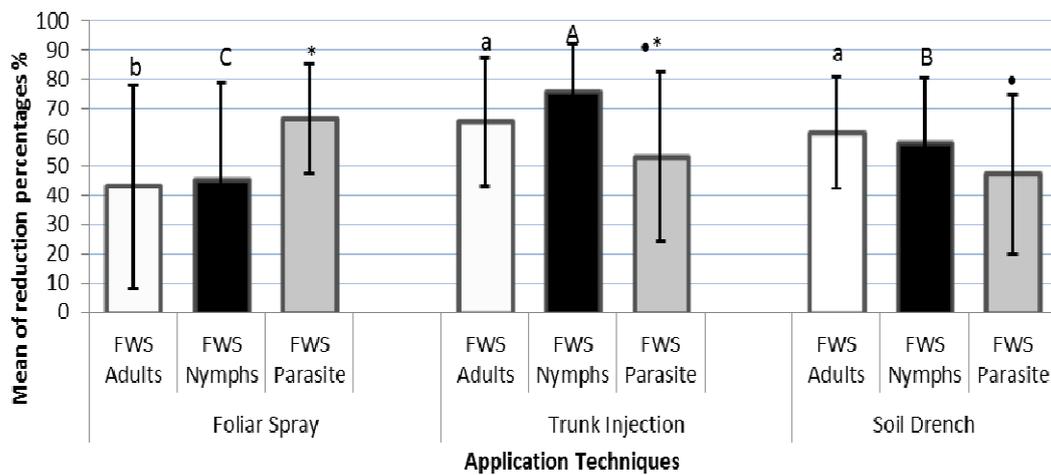


Fig. (5): The general means of reduction percentages of adults and nymphs of Florida wax scale (FWS) and its parasitoid by dinotefuran insecticide throughout different application techniques in the 1st season, 2019. F adult = 10.147, df = 149, L. S. D. = 10.3732; F nymphs = 17.807, df = 149, L. S. D. = 9.9333; F parasite = 7.221, df = 149, L. S. D. = 10.06515

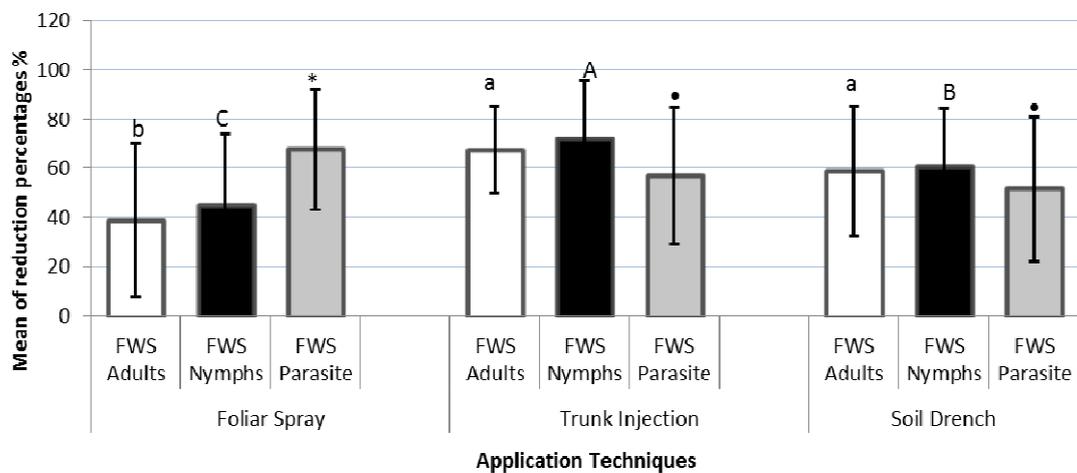


Fig. (6): The general means of reduction percentages of adults and nymphs of Florida wax scale (FWS) and its parasitoid by dinotefuran insecticide throughout different application techniques in the 2nd season, 2020. F adult = 16.186, df = 149, L. S. D. = 10.1701 F nymphs = 13.937, df = 149, L. S. D. = 10.1585 F parasite = 4.616, df = 149, L. S. D. = 10.7567

Effect of the insecticides treatments:

According to the general means of reduction percentages, the Florida wax scale populations were significantly reduced by the insecticide treatments (Figs. 7 and 8). Results indicated that, imidaclopride achieved the highest reduction% of adults and nymphs of *C. floridensis* with general mean 71.42% and 69.01% in 1st season, 67.65% and 70.21% in 2nd

season, respectively. On the other hand, dinotefuran achieved the least reduction with general mean 56.61% and 59.55% in 1st season, 55.05% and 59.13% in 2nd season. Imidaclopride gave the highest toxicity against parasite, *C. scutellaris* (60.63% and 66.71%) followed by acetamiprid (57.53 and 62.59%) which is followed by dinotefuran (55.53 and 58.79) in 1st and 2nd season, respectively.

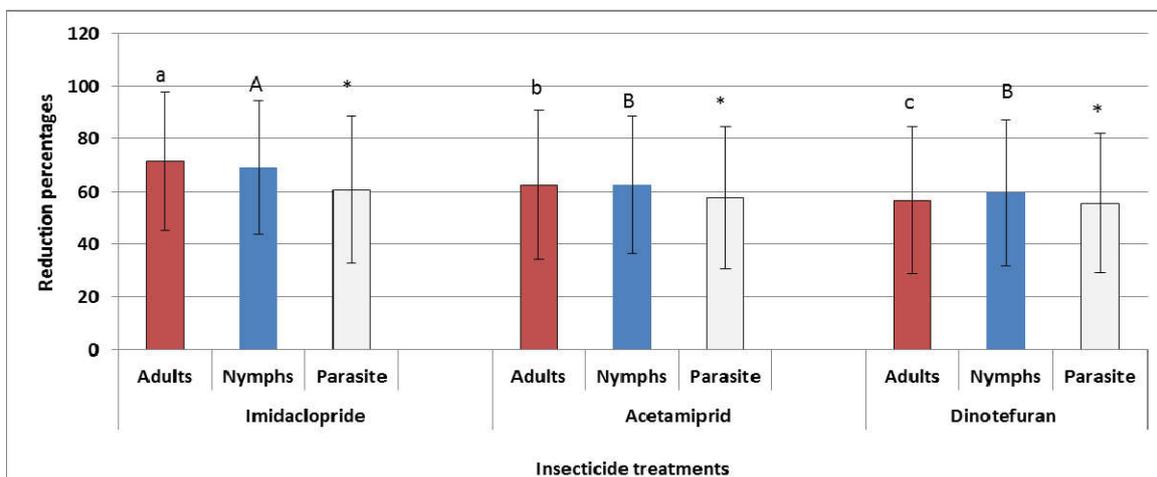


Fig. (7): The Efficacy of three Neonicotinoids insecticides against Florida wax scale and its parasitoid during 2019 season under field conditions as a general means of three different application methods. F adult = 11.125, df = 449, L. S. D. = 6.21555. F nymphs = 4.978, df = 449, L. S. D. = 6.00905 F parasite = 1.341, df = 449, L. S. D. = 6.16175

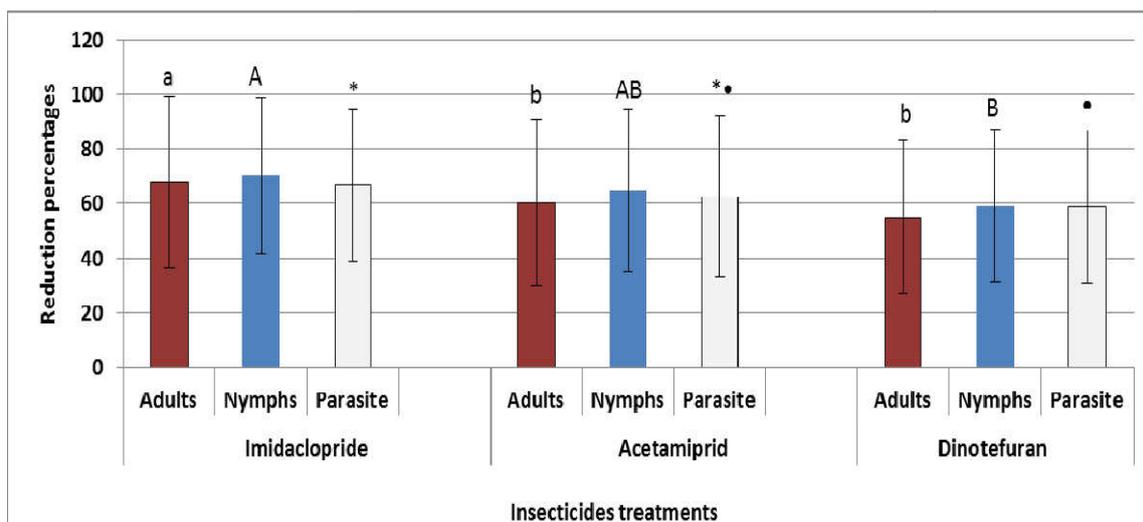


Fig. (8): The Efficacy of three Neonicotinoids insecticides against Florida wax scale and its parasitoid during 2020 season under field conditions as a general means of three different application methods. F adult = 6.635, df = 449, L. S. D. = 6.8271. F nymphs = 5.622, df = 449, L. S. D. = 6.49395, F parasite = 2.931, df = 449, L. S. D. = 6.427

Effect of the interactions between the application techniques and insecticides:

There are many significant effects of the interactions among studied factors (application techniques and insecticides treatments) on studied characters. The three Neonicotinoids insecticides and the three types of application methods had significant differences between one of them with the other. Data presented in Tables 2 and 3 revealed that the best results of reduction percentages (highest percentages) of adults and nymphs of FWS obtained from samples treated with imidaclopride throughout trunk injection

method. The least effective method was the foliar spray. In all cases of the foliar spray the nymphs of FWS were more affected by the three insecticides than the adults. On the contrary, the least suitable technique for the parasite of FWS was the foliar spray with imidaclopride insecticides which causing 70.91 and 72.09 % reduction percentages in the 2019 and 2020 seasons, respectively. While the most suitable technique (the least reduction percentages) was achieved using the soil drench with the dinotefuran insecticide.

Table (2): The general means of reduction percentages of adults and nymphs of Florida wax scale (FWS) and its parasitoid by three insecticide throughout three different application techniques in the 1st season, 2019

Application techniques	Insecticides	Adults of FWS	Nymphs of FWS	The parasitoids, <i>C. scutellaris</i>
Foliar spray	Imidaclopride	54.26±30.93 ^e	57.54±33.03 ^{cd}	70.91±24.43 ^a
	Acetamiprid	45.36±33.38 ^f	49.87±35.21 ^{de}	68.1±23.71 ^a
	Dinotefuran	43.12±34.79 ^f	45.4±33.42 ^c	66.15±18.6 ^{ab}
	Mean	47.58±33.2	50.94±34.04	68.39±22.33
Soil drench	Imidaclopride	77.66±17.24 ^{ab}	69.84±21.32 ^b	53.72±29.03 ^{cd}
	Acetamiprid	68.72±23.7 ^{cd}	62.02±17.83 ^c	48.91±21.747 ^{cd}
	Dinotefuran	61.57±19.15 ^{de}	57.97±22.4 ^c	47.214±27.25 ^d
	Mean	69.32±21.14	63.28±21.06	49.95±26.16
Trunk injection	Imidaclopride	82.34±19 ^a	79.64±12.49 ^a	57.25±27.65 ^b
	Acetamiprid	73.27±16.77 ^{bc}	76.03±13.51 ^{ab}	55.59±31.43 ^{cd}
	Dinotefuran	65.15±22.12 ^{cd}	75.27±16.63 ^{ab}	53.23±29.27 ^{cd}
	Mean	73.59±20.54	76.98±14.35	55.35±29.34
L.S.D.		8.473	7.7751	9.4792

In each column the mean followed by the same letter(s) are not significantly different as indicated by L.S.D. test (P=0.05)

Table (2): The general means of reduction percentages of adults and nymphs of Florida wax scale (FWS) and its parasitoid by three insecticide throughout three different application techniques in the 2nd season, 2020

Application techniques	Insecticides	Adults of FWS	Nymphs of FWS	The parasitoids, <i>C. scutellaris</i>
Foliar spray	Imidaclopride	49.62±36.87 ^c	58.47±34.12 ^{de}	72.09±23.61 ^a
	Acetamiprid	43.49±38.11 ^{ef}	52.75±34.64 ^{ef}	71.7±22.17 ^a
	Dinotefuran	38.91±31.23 ^f	44.89±29.24 ^f	67.79±24.41 ^{ab}
	Mean	44 ±35.56	52.04±33.01	70.53±23.34
Soil drench	Imidaclopride	68.6±27.29 ^{bc}	70.22±28.09 ^{bc}	59.14±33.6 ^{cd}
	Acetamiprid	63.93±24.29 ^{cd}	64.43±26.22 ^{cd}	53.54±33.78 ^d
	Dinotefuran	58.76±26.45 ^d	60.56±23.96 ^{de}	51.52±29.24 ^d
	Mean	63.76±26.18	65.07±26.27	54.7315±32.22146
Trunk injection	Imidaclopride	84.73±16.35 ^a	81.92±15.63 ^a	68.89±23.31 ^{ab}
	Acetamiprid	73.69±17.45 ^b	76.7±21.31 ^{ab}	62.52±28.84 ^{bc}
	Dinotefuran	67.47±17.64 ^{bc}	71.92±23.51 ^{bc}	57.07±27.77 ^{cd}
	Mean	75.29±18.48	76.85±20.69	62.83±27.01
L.S.D.		7.60983	8.78354	7.892

In each column the mean followed by the same letter(s) are not significantly different as indicated by L.S.D. test (P=0.05)

DISCUSSION

Results of the current study revealed significant differences in the reduction percentages (mortality) of FWS, *C. floridensis* and its parasitoid, *C. scutellaris* between the different application techniques and insecticide treatments in all sampling sessions. By this study, we compared the efficiency of three different application methods comprised of foliar spray, trunk injection and soil drench against FWS and its parasitoid. The results revealed a high efficacy of the trunk injection and soil drench in suppressing FWS population which agrees with findings of Bhandari and Cheng (2016) for control stem and leaf gall wasps, *Josephiella* species (Hymenoptera: Agaonidae), on Chinese banyan. The results also are in agreement with Arbab Tafti *et al.* (2014) on the high efficacy of trunk injection in control of Dubas bug, *Ommatissus lybicus* (Hemiptera: Tropicuchidae) on date palms. Additionally, the results also refer to fewer side effects of trunk injection and soil drench on the parasite, *C.*

scutellaris compared with foliar application may be due to the less direct contact. Similar results were obtained by many other studies such as Arbab Tafti *et al.* (2014) who found fewer side effects of trunk injection on natural enemies of *O. lybicus* on date palms compared with foliar application. Also, the results of Wise (2014), Paula Alayón Luaces *et al.* (2018), Majeed Askari Seyahoei (2019) and Darwish (2020) refer to the use of trunk injections and soil drench would be a more efficient and more environmentally friendly than the foliar spray method. They concluded that the use of trunk injection and soil drench reduces the drift of insecticides and environmental risks and consequently also safe for farmers, agricultural workers. In this study we test three of the neonicotinoid insecticides viz. acetamiprid, dinotefuran and imidaclopride. Fernandes de Cordova and Gallego (1997) cured the oak scale insect which infested oak trees by injection the trees with imidacloprid solution; moreover, they added that more

than 79% control of this insect was obtained when imidacloprid was injected at rates of 0.8 ml per tree. The results also are consistent with Paula Alayón Luaces *et al.* (2018) who reported that to control the snow scale of citrus trunk under the field conditions only one application of imidacloprid injections per campaign is enough. The least effective insecticide was dinotefuran, especially when applied as a foliar spray. It achieved only 43.12 and 45.4% reduction percentages in the 1st season and 38.91 and 44.89% in the 2nd season for adults and nymphs of FWS, respectively. Quesada *et al.* (2018) found that the insecticide dinotefuran was non effective (as a foliar spray) on the two soft scale insects, calico scale (*Eulecanium cerasorum*) and striped pin scale (*Toumeyella pini*). While this insecticide was effective when applied as a soil drench against the soft scale, *Eulecanium cerasorum* (Quesada Machigua, 2013).

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تقييم كفاءة طرق تطبيق مختلفة لثلاث مبيدات نيكوتينية مختلفة علي حشرة الموالح الشمعية *Coccophagus scutellaris* والطفيل الخاص بها *Ceroplastes floridensis*

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تعتبر حشرة الموالح الشمعية *Ceroplastes floridensis* واحده من أهم الآفات الحشرية التي تصيب أشجار الموالح في العالم. في هذه الدراسة تم إجراء تجارب حقلية لتقييم كفاءة ثلاثة طرق مختلفة لتطبيق ثلاثة مبيدات نيكوتينية جهازية مختلفة علي حشرة الموالح الشمعية وعلي الطفيل غشائي الأجنحة الخاص بها *Coccophagus scutellaris*. تمت التجارب في مزرعة خاصة في منطقة النوبارية، محافظة البحيرة، جمهورية مصر العربية في موسم الربيع لعامي ٢٠١٩ و ٢٠٢٠. العينات تم أخذها أسبوعيا ولمدة عشر أسابيع. أوضحت النتائج أنه من بين الثلاث طرق مختلفة للتطبيق، حققت طريقة حقن الجذع بالمبيد Trunk injection أعلى نسب خفض للحوريات والحشرات الكاملة للحشرة القشرية. وأتبعته طريقة حقن الجذع بطريقة التطبيق عن طريق التربة Soil drench وأخيرا طريقة الرش الورقي Foliar spray. علي العكس من ذلك كانت طريقة حقن الجذع أكثر حفاظا علي الطفيل *C. scutellaris* محققة أقل نسب خفض لأعداد الطفيل عن الطرق الأخرى للتطبيق. ومن جانب آخر، كان أكثر المبيدات كفاءة في خفض عشيرة الحوريات والحشرات البالغة للحشرة القشرية والطفيل هو مبيد الايميداكلوبريد Imidacloprid، يليه مبيد الاسيتامبريد Acetamiprid وأخيرا مبيد الداينوتروفوران Dinotefuran. وعلي ذلك فإن هذه النتائج تشير إلي أن طريقة حقن الجذع بالمبيد كانت أكثر الطرق مناسبة لمكافحة الحشرة ويمكن التوصية بهذه الطريقة في برامج مكافحة المتكاملة.