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Impact of certain insecticides with different mode of action on the California red scale *Aonidiella aurantii* (Hemiptera-Diaspididae) on orange under local conditions in Egypt

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Abstract:

The California red scale (CRS) (*Aonidiella aurantii* (Maskell). (Hemiptera-Diaspididae) is one of the most important pests infested citrus trees, which come second to grapes of fruit trees worldwide. In Egypt, It is threatening the citrus trees that are important either in exporting or national consumption. The California red scale is able to develop resistance, so five insecticides with different modes of action were tested in this study. These insecticides are: Pyriproxyphen (Admiral), 10% EC; imidacloprid (Best), 25% WP; spirotetramat (Movento), 10% SC; sulfoxaflor (Isoclast), 50% WG and a mineral oil **(**Kz oil**)** 95% EC. The used insecticides were applied using 20 Liter Sprinkler using randomized complete blocks design (RCBD) design on fruit full navel orange trees. The tested insecticides reduced the treated insect stages population in a function of the treated stage, the insecticide mode of action and the time after treatment. Among the tested insecticides, Kz oil was the most effective, reducing the treated population in systemic arrangement through suffocation effect. Pyriproxyphen (Admiral) appeared too low effective to cause 50% adult population reduction with multiplied effect against the other stages to as maximum as 97.9% because of its IGR mode of action. Sulfoxflor (Isoclast) and spirotetramat (Movento) exceeded the imidaclobrid (Best) in their lethal effects against all stages of almost the checked time after treatment in both 2018 and 2019 seasons. Different effects of the tested insecticides are explained. These insecticides are known as integrated pest management compatible with little to no effect against CRS natural enemies, low toxicity to mammals and man. They are also classified as non-carcinogenic, non-mutagenic and non-reprotoxic under the European Chemicals Agency (ECHA) classification.

Introduction

Citrus fruits come second to grapes of fruit trees worldwide (Grafton-Cardwell, 2010). According to 2020 statistics of the Egyptian Ministry of Agriculture, Egypt is the 1st citrus exporting country exporting 1.8 million ton to 111 countries all over the world. Scale insects are one of the problematic phenomena threatens the citrus trees that are important in Egypt either in exporting or national consumption. Imidacloprid, clothianidin and sulfoxaflor strongly caused significant repellency, reduction in feeding and adults body weight of *Schizaphis graminum* (Rondani) (Hemiptera: Aphididae) and *Coccinella septempunctata* L. (Coleoptera: Coccinellidae) at sub-lethal doses (Bilal *et al*., 2020). Imidacloprid is used to control sucking insects, some chewing insects, including termites, soil insects, and fleas on pets. In addition to its topical use on pets, imidacloprid may be applied to crops, soil, and as a seed treatment (Tomlin, 2006).

The California red scale (CRS) (*Aonidiella aurantii* Maskell, Family, Diaspididae is one of the most important pest infested citrus trees in different parts of the world (Sorribas *et al*., 2010). This pest causes leaf drop (Defoliation), fruit, dry out and fall off and trunk heavy infestation (Bedford, 1998). It decreases the tree viability and the fruits cosmetic damage, resulting in its downgrading. In severe infestations, leaf yellowing and dieback of branches occur, reducing the tree productivity and health to death (Flint *et al*., 1991). Biological control is not always sufficient to keep it below an economic threshold (Forster *et al*., 1995). Fresh market fruit, results in a pest management challenge that often requires insecticides. California red scale has developed resistance to the used insecticides. CRS was controlled in late in the 18 century with hydrogen cyanide (HCN), but its resistance was detected **(**Quayle, 1938**).**

In the 1940s, citrus growers next relied on organophosphate and later carbamate insecticides (Carman, 1977), which the CRF developed their resistance in 1970s in South Africa (Nel *et al*.,1979), Australia (Abdelrahman, 1973) and in the 1990s in California (Grafton-Cardwell and Vehrs, 1995). Several generic 350 SC imidacloprid formulations are used in drip or micro sprinkler irrigation systems, but CRS control was inconsistent depending on the quality of the irrigation system. Insect growth regulators (IGRs) were used to interfere with insect metamorphosis, growth or reproduction. However, some are non-selective and may detrimentally affect natural enemies. Buprofezin and pyriproxyfen IGRs were registered for CRS control in California in the late 1990s.

Pyriproxyfen was used for 90% of the IGR applications in the San Joaquin Valley in 2005-2010 because of its low cost and great efficacy [California Department of Pesticide Regulation (CDPR)] (2000-2010) (Grafton-Cardwell *et al*., 2006). Pyriproxyfen and buprofezin are low toxic on the primary parasitoid of CRS, *Aphytis melinus* DeBach (Hymenoptera: Aphelinidae) (Rill *et al*., 2008), increasing the scale control. Although pyriproxyfen was the primary CRS treatment for more than a decade, its resistance monitoring has not revealed significant levels (Ouyang and Grafton-Cardwell, 2010). Because of the ability of California red scale to develop resistance, it is important to introduce insecticides with alternative modes of action into the treatment regime. So, in this study, some insecticides with different modes of action were tested against the CRS different stages on navel orange trees on October 2018 and October 2019 as *A. aurantii* has two peaks in April and October.

This study aimed to differentiate among the used toxicants (Pyriproxyphen (Admiral), 10% EC; imidacloprid (best), 25% WP; spirotetramat (Movento), 10% SC;

sulfoxaflor (Isoclast), 50% WG and a mineral oil (Kz oil) 95% EC), regarding their reduction of each CRS stage as well as its total population.

Material and methods

1. Treated Insect:

The California Red Scale (CRS), *A. aurantii* as the most spreader, the highest census and the most effective in the treatment region. The all treated insect stages were treated *in situ* on the fruit full navel orange trees in Rashid City, Behira Governorate, Egypt.

2. Tested insecticides:

Five commercial insecticides belong to different chemical groups were tested for their lethality against the treated insect (CRS) different stages. These insecticides were applied at the recommended rates. The tested insecticides common and trade names, formulations, application rates, chemical classes and chemical structures as well as their basic manufacturers are arranged in the following Table (1).

3. Experimental procedure:

Treating the CRS insect stages was carried out on navel fruit full orange host plant trees using the foliar application. The host plant trees were not chemically treated two years before this study. The treatment experiment was designed in randomized complete block design (RCBD). Four host plant trees were taken for each replicate and four replicates were used for each treatment. The navel orange host plant trees were sprayed once with the tested compounds at the application rates (Table **1**) using the 20 liter sprinkler machine. Fifty (50) leaves of each replicate were randomly taken directly before spraying (Pre-spraying). The treated insect (CRS) stages were counted and the infestation limit was determined. This step (Taking 50 leaves of each replicate) was repeated again four times in 2, 4, 6 and 8 weeks after treatment in both treatment and control. These leaves were transferred in paper bags to the laboratory and each stage of the treated insect was individually counted at each time interval. The total studied stage number was calculated. This count and discrimination of each stage alive number were carried out using the stereomicroscope. Counting was repeated four times in each replicate and its mean± SD number was considered. The four mean± SD numbers of the four replicates were averaged for each treatment. Control was concurrently conducted. The reduction percentage in each studied stage population and the total population number was calculated according to (Henderson and Tilton, 1955) formula.

$$\text{Reduction \%} = 100\ [1- (T_2 / T_1 * C_1 / C_2)]$$

T_2 Population in Treatment after spray

T_1 Population in Treatment before spray

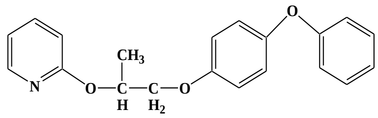
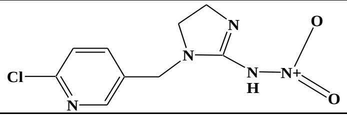
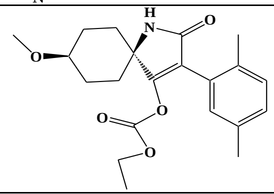
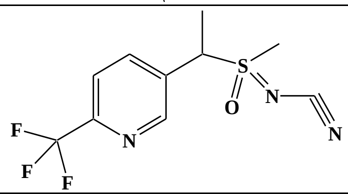
C_1 Population in control before spray

C_2 Population in control after spray

4. Statistical analysis:

The split-plot system in randomized complete block design (RCBD) with five treatments and control, four replicates in each treatment were designed according to Steel and Torrie (1981). The analysis of variance (ANOVA) was performed , Costat Software version 6.311 (Cohort Soft Ware , 2005) at 0.05 probability level. Effective time that reduces 50% of the treated stage (ET_{50}) value was calculated according to probit analysis (Finney, 1971).

Table (1): The tested insecticides used against the *Aonidiella aurantii* (CRS) insect stages.

Common name (**Trade name**)	**Chemical class**	**Basic manufacturer**	**Application rate**	**Chemical structure**
Pyriproxyphen (Admiral) 10% EC	Phenoxyphenylpyridyloxy) propyl ether	Sumitomo Chemical Australia Pty Ltd A.B.N. 21 081 096 255	50 ml/ 100 L	
Imidacloprid (Best) 25% WP	Neonicotinoids	El-Helb Pesticides and Chemicals Industries, Egypt	100 gm/ 100 L	
Spirotetramat (Movento) 10% SC	Tetramic acid derivative (Ketoenol)	Bayer Crop Science LP, Research Triangle Park, NC	40 ml/ 100 L	
Sulfoxaflor (Isoclast) 50% WG	Sulfoximines	Dow AgroSciences, LLC, Indianapolis, IN	125 gm/ 100 L	
Mineral oil (Kz oil) 95% EC		Kz Company for Pesticides and Chemical Industries, Kafr-Elzayat, Egypt	1.5 L/ 100 L	

Results and discussion

The tested insecticides killed the treated insect stages differently in a function of the treated insect stage, the insecticide mode of action and the time after treatment. These effects are shown in Tables (2, 3 and 4), from which it could be said that the untreated adult population increased systematically with the time after spraying until 8 weeks in both 2018 and 2019 treatments. The other untreated stages population density was fluctuated through the test period; however, the average total population count was increased from 1193 to 1591 and from 936.5 to 1207 in 2018 and 2019 studies, respectively (Table **2**).

Among the tested insecticides, the used mineral oil (Kz oil) was the most effective with lethal effect against the CRS stages increased with increasing the time in systemic arrangement. It harshly reduced the treated populations from 234.5 to 10.8, from 408.8 to 12.0, from 235.5 to 7.3 and from 394 to 12.3 in case of adult, crawler, 1st instar nymph and the 2nd instar nymph stages, respectively in the 2018 season, comparing with the reduction from 188.3 to 0.75, from 294.8 to 4.0, from186.8 to 0.8 and from 273.5 to 4.0 in the same arrangement in 2019 experiment.

The average total stages population number was reduced with its treatment from 1272.8 to 42.3 and from 243.3 to 9.5 (Table **2**) in the two seasons, respectively. It caused reduction percent increased along the experiment time ranged from 89.0 to 97.1, from 80.4 to 97.3, from 93.3 to 98.1, from 64.4 to 97.1, from 83.5 to 97.5 in case of adult, crawler, 1st instar nymph, 2nd instar nymph and the total stages population in 2018 test, respectively (Table 3).

In 2019 experiment, it achieved reduction ranges from 91.2 to 99.8, from 80.6 to 98.7, from 94.1 to 99.7, from 68.6 to 98.4 and from 84.9 to 99.2 in the same order in the 2019 experiment (Table 3). Our obtained results agreed with Helmy *et al*. (1991) who proved the sensitivity of *A. aurantii* to Kz oil and the more susceptibility of the nymphal stage than adult females and with Khalil *et al*. (1996) who reported the satisfactory results of Kz oil against different *A. aurantii* (CRS) stages at 2, 4 and 6 weeks after summer spray on balady orange trees. From the obtained results, Kz oil was the most effective emphasizing that the use of oils in crop protection is a good alternative to conventional synthetic insecticides. Oils have good control of some pests and plant pathogens at low doses (1–2%), no resistance in target pathogens, low cost, excellent spreading on leaf surface and low environmental impact. Its relatively identical results in 2018 and 2019 seasons emphasize its similar strong effect.

Imidacloprid (Best) reduced the treated insect stages populations nearly similar in both 2018 and 2019 treatments. Its reduction percent ranges were 77.6 – 87.7, 64.7 – 82.8, 74.8 – 80.9 and 54.0 – 80.1 against adult, crawler, 1st instar and 2nd instar, respectively in 2018 treatment, comparing with 79.9 – 84.4, 72.1 – 87.7, 75.8 – 83.9 and 53.0 – 84.4 in the same arrangement in 2019 treatment emphasizing its toxicity precision. Low fluctuation in its effect with the time after spraying can be referred to the fluctuation in control population or to its weak residual effect. It decreased the total treated insect stages population with 70.7, 82.7, 85.4 and 78.5 in 2018, comparing with 74.9, 85.9, 85.9 and 79.4 in 2019, respectively at 2, 4, 6 and 8 weeks after treatment (Table **3**).

These results agreed with Tomlin (2006) as he reported that imidacloprid is designed to be effective by contact or ingestion controlling sucking insects, some chewing insects, including termites, soil insects, and fleas on pets as a systemic insecticide that rapidly translocate through plant tissues following application and it can be applied to crops, soil, and as a seed treatment (Tomlin, 2006). Spirotetramat (Movento) affected the treated insect stages in a function of both treated stage and time after treatment. It killed the treated population exhibiting 82.3 – 92.5, 75.9 – 90.6, 85.5 – 93.4, 57.1 – 86.6 and 78.8 – 92.8 reduction ranges in population of adult, crawler, 1st instar and 2nd instar as well as total population, respectively in 2018 treatment. These values were 87.0 – 94.6, 78.0 – 93.5, 90.7 – 94.8, 59.1 – 89.2 and 80.5 – 94.1, respectively in the same order in 2019 treatment insuring the insecticide effect precision (Table 3). These results agreed with several researchers who proved spirotetramat, a foliar systemic tetramic acid insecticide activity against sucking insects as white mites, psyllids, and aphids was registered in California citrus in 2008 (Frank and Lebude, 2011; Jamieson *et al*., 2010 and Page-Weir *et al*., 2011). Its unique two-way systemic action (Moving via the phloem and xylem) potentially allows its application at lower water volumes comparing with other foliar insecticides (Bruck *et al*., 2009). 1st and 2nd instars male and female *A. aurantii* were more susceptible to spirotetramat than early and late 3rd instar females because of reduction in feeding (Forster *et al*., 1995) as it has limited contact toxicity and its main effect is achieved through ingestion (Bruck *et al*., 2009). Spirotetramat also reduced the fecundity of California red scale but did not eliminate it (Cruz *et al*., 2013). Sulfoxaflor (Isoclast) appeared more effective than imidacloprid achieving 83.6 – 92.8, 77.3 – 92.2, 87.3 – 93.7, 59.4 – 88.6 and 80.5 – 93.4

population reduction percent ranges in 2018 treatment, comparing with 88.8 – 96.5, 78.0 – 94.5, 92.4 – 96.4, 64.3 – 90.7 and 82.3 – 95.9 in 2019 treatment in adult, crawler, 1st instar, 2nd instar and total stages population, respectively (Table 3). Pyriproxyphen (Admiral) behaved different from the other tested insecticides as its reduction in the treated adult population was as high as 41.6% at 4 weeks after treatment and decreased after that time. Its effect was multiplied to 97.9, 96.1 and 75.1% against crawler, 1st instar and 2nd instar stages, respectively in 2018 treatment, comparing with 89.7, 97.6 and 76.4 in the same order in 2019 treatment at 8 weeks after treatment. Its low reduction in average total population might be due to its low adult stage population reduction in agreement with Mohamed (2002) who proved the toxicity of admiral pyriproxyfen (Admiral) on CRS (*A. aurantii*). Sulfoxflor (Isoclast) and spirotetramat (Movento) exceeded the imidaclobrid (Best) in its lethal effect against all treated insect stages at all the checked time after treatment in both 2018 and 2019 seasons. Pyriproxyphen (Admiral) behaved different from the other tested insecticides as its mortal effect appeared too low to reach 50% of the treated adult population (As high as 41.6%) at 4 weeks after treatment and decreased after that time. Its lethality was multiplied on the other stages to as maximum as 97.9, 96.1 and 75.1% against crawler, 1st instar and 2nd instar stages, respectively in 2018 treatment, comparing with 89.7, 97.6 and 76.4 in the same order in 2019 treatment at 8 weeks after treatment. Its low reduction effect on total population might be due to the low reduction against the adult stage population. Mohamed (2002) proved the toxicity of Kz oil and admiral (Pyriproxyfen) on *A. aurantii* and proved that Admiral was more effective in summer than spring. The difference among the used insecticides activities against the treated California red scale (CRS), *A. aurantii* may be due to their different mode of action. KZ oil controls the California red scale (CRS), *A. aurantii* through blocking effect on the respiratory system openings resulting in suffocation effect (Cook *et al*., 2004 and Martín *et al*., 2004). In general, oils suffocate the full range of scale developmental stages on leaves or wood more than on fruits because the mature scale seals down their scale cover more securely on the comparatively smooth, uniform fruit surface than leaves or wood. However, imidacloprid (best) acts as a nicotinic acetyl choline receptors (nAChRs) agonist (Kayser *et al*., 2016). It acts on several types of post-synaptic nicotinic acetylcholine receptors in the nervous system (CNS) (Matsuda and Sattelle, 2005), which are located only within the CNS in insects. Its binding to the nicotinic receptor causes nerve impulses spontaneously discharging at first, followed by the neuron failure to propagate any signal (Sheets, 2001).This binding process is irreversible (Ware and Whitacre, 2004). Sulfoxaflor (Isoclast) has a novel mode of action as a new nAChRs modulating insecticide (Cutler *et al*., 2013). Bacci *et al*. (2018) added that it binds to nAChR in place of acetylcholine and acts as an allosteric activator of nAChR. Its binding to receptors caused uncontrolled nerve impulses followed by muscle tremors, paralysis and finally death. Sulfoxaflor binds differently from neonicotinoids and so, it causes a high efficacy degree against a wide range of insects including resistant to neo-nicotinoids exhibiting structure activity relationship that are different from other nAChRs agonists such as imidacloprid (Sparks *et al*., 2013). Sulfoxaflor passes cross-resistance of many pest species because of some monooxygenase as Cytochrome P450 and CYP6G1are able to degrade some neonicotinoids as imidacloprid, but are incapable of metabolizing sulfoxaflor (Zhu *et al*., 2011).

Table (2): Effect of the tested insecticides on *Aonidiella aurantii*; shown as mean ±SD.

Tested Compounds	Season	Treated Stage	Number of the treated insect stages after different times				
			Pre-Spraying	2 weeks	4 weeks	6 weeks	8 weeks
Control	2018	Adult	223.5±6.61	272.3±9.74	303.5±4.8	336.3±14.8	351.8±8.58
		Crawler	384.3±14.80	402.8±8.77	412.0±4.99	416.3±6.02	412.0±7.40
		1^{st} instar	226.8±5.19	429.0±10.2	367.3±8.66	383.5±4.8	375.3±5.1
		2^{nd}instar	358.5±14.6	230.5±8.10	289.5±6.40	100.0±2.9	379.0±2.50
		Total	**1193.0±32.03**	**1334.5±21.76**	**1372.0±18.57**	**1498.0±7.04**	**1591.0±11.64**
	2019	Adult	184.0±4.69	233.5±5.56	253.3±5.12	278.3±12.87	300.0±11.52
		Crawler	292.5±5.80	308.3±1.26	318.0±2.16	304.5±4.80	299.8±10.24
		1^{st} instar	175.5±3.87	319.5±3.87	252.3±5.44	250.5±2.65	246.0±5.60
		2^{nd}instar	284.5±6.35	195.3±5.44	247.8±3.95	82.5±2.65	254.3±4.50
		Total	**936.5±13.92**	**1061.8±9.00**	**1071.0±10.70**	**1196.0±10.6**	**1207.0±20.90**
Pyriproxyfen (Admiral)	2018	Adult	225.8±5.38	194.5±4.93	189.5±5.0	211.5±5.8	222.3±6.19
		Crawler	384.3±12.04	119.5±9.04	41.5±2.08	9.75±2.22	8.75±2.21
		1^{st} instar	224.5±4.94	113±4.43	49.3±2.75	10.5±1.73	14.8±1.71
		2^{nd}instar	368.0±9.4	191.8±9.7	124.3±3.60	77.5±3.40	92.3±2.99
		Total	**1202.5±19.64**	**618.3±11.64**	**404.5±7.42**	**309.3±9.29**	**338.0±7.12**
	2019	Adult	172.5±4.66	164.0±6.37	150.5±1.92	162.0±2.94	178.5±4.44
		Crawler	294.8±5.68	86.0±4.55	26.0±2.83	4.8±2.50	4.0±1.63
		1^{st} instar	190.3±2.99	99.8±2.99	28.0±4.83	4.5±1.29	6.0±1.63
		2^{nd}instar	284.0±4.97	137.8±4.03	89.0±1.83	52.0±3.56	60.0±2.58
		Total	**941.5±7.60**	**487.5±14.25**	**293.5±7.60**	**223.3±6.40**	**248.5±7.00**
Imidacloprid (Best)	2018	Adult	224.5±5.69	61.3±6.1	49.5±2.65	41.5±2.1	73.0±3.65
		Crawler	390.3±10.87	144.0±5.16	57.3±3.30	72.5±5.80	91.8±8.34
		1^{st} instar	225.0±4.97	81.3±5.74	75.0±7.39	77.2±1.9	94.0±3.56
		2^{nd} instar	374.5±10.5	110.8±5.9	60.3±3.59	21.3±1.7	89.3±8.30
		Total	**1214.3±16.58**	**397.3±13.94**	**242.0±11.28**	**222.0±4.24**	**348.0±16.75**
	2019	Adult	179.5±3.70	36.3±3.30	26.3±0.96	38.8±4.35	58.8±5.91
		Crawler	294.8±12.12	86.8±2.75	39.5±1.29	57.8±2.87	63.0±3.65
		1^{st} instar	178.8±3.50	52.3±3.30	46.5±1.91	52.0±3.16	60.8±3.78
		2^{nd} instar	278.5±6.81	89.8±2.22	37.8±1.71	18.8±1.71	64.8±4.99
		Total	**931.5±21.00**	**265.0±4.90**	**150.1±0.82**	**167.3±6.90**	**247.3±12.60**
Spirotetramat (Movento)	2018	Adult	229.8±5.32	49.5±3.42	32.0±2.58	26.0±3.74	56.0±2.16
		Crawler	389 ±9.89	98.5±5.57	48.25±2.5	39.5±1.29	69.5±3.7
		1^{st} instar	230.0±2.16	39.3±4.50	32.3±2.22	25.8±4.86	55.25±4.79
		2^{nd} instar	377.8±8.99	104.3±8.6	40.8±1.50	19.3±1.7	68.8±7.80
		Total	**1226.5±16.90**	**291.5±9.40**	**153.3±5.62**	**110.5±9.26**	**249.5±14.80**
	2019	Adult	177.0±2.58	29.8±0.96	21.3±1.71	14.5±1.29	28.0±0.82
		Crawler	303.8±5.62	70.5±1.29	30.3±0.96	20.5±1.29	40.3±3.78
		1^{st} instar	180.0±3.74	29.5±1.29	17.8±0.96	13.5±1.29	23.5±0.58
		2^{nd} instar	277.3±4.92	77.8±2.75	26.0±1.41	22.0±1.83	41.0±1.41
		Total	**938.0±14.20**	**207.5±2.60**	**95.3±3.60**	**70.5±2.60**	**138.8±4.50**
Sulfoxaflor (Isoclast)	2018	Adult	230.0±2.94	39.3±4.86	30.3±1.98	25.0±1.83	59.3±1.71
		Crawler	395.0±5.72	93.8±5.44	39.0±1.83	33.3±1.71	70.8±1.71
		1^{st} instar	228.5±1.30	37.0±3.20	28.8±2.60	24.3±0.96	48.0±4.20
		2^{nd} instar	380.3±5.30	99.3±3.10	35.0±2.90	20.5±1.30	56.5±8.7
		Total	**1233.8±11.33**	**269.3±5.19**	**133.0±7.57**	**103.0±1.41**	**234.5±14.20**
	2019	Adult	180.5±3.0	26.3±1.71	15.3±0.96	10.5±1.29	22.0±1.41
		Crawler	298.0±7.62	69.0±3.16	25.5±2.38	17.0±1.41	35.8±2.22
		1^{st} instar	183.0±5.48	25.3±1.50	13.0±0.82	9.5±1.29	16.8±1.71
		2^{nd} instar	280.5±5.80	68.8±2.22	22.8±3.30	12.8±1.26	29.3±1.71
		Total	**942.0±17.80**	**189.3±4.90**	**76.5±3.50**	**49.8±1.90**	**96.0±13.6**
Mineral oil (Kz oil)	2018	Adult	234.5±8.74	31.5±1.91	15.3±1.26	14.0±1.83	10.8±1.71
		Crawler	408.8±2.63	83.8±7.14	16.0±0.82	15.0±2.16	12.0±1.41
		1^{st} instar	235.5±4.80	29.8±4.20	21.3±2.50	11.0±0.82	7.3±.096
		2^{nd} instar	394.0±5.60	90.3±4.60	21.3±1.70	18.3±1.50	12.3±0.96
		Total	**1272.8±10.81**	**235.3±7.14**	**73.8±6.13**	**58.3±2.99**	**42.3±2.75**
	2019	Adult	188.3±2.75	21.5±2.08	7.3±0.96	4.5±1.29	0.75±0.96
		Crawler	294.8±9.91	60.3±2.99	12.3±1.71	6.0±1.83	4.0±1.83
		1^{st} instar	186.8±5.74	20.0±1.41	5.5±1.29	3.5±1.29	0.8±0.96
		2^{nd} instar	273.5±3.11	59.0±2.94	10.3±1.50	6.0±1.41	4.0±0.82
		Total	**943.3±12.70**	**160.8±4.10**	**35.3±1.50**	**20.0±3.90**	**9.5±3.00**

Table (3): Effect of the tested insecticides on *Aonidiella aurantii* (CRS) different stages; shown as reduction % in population.

Tested insecticide	Season	Treated Stage	Reduction % in different CRS insect stages after different times			
			2 weeks	4 weeks	6 weeks	8 weeks
Pyriproxyfen (Admiral)	2018	Adult	29.2±3.53	37.5±2.41	37.0±4.23	36.1±1.54
		Crawler	70.3±3.10	89.9±0.51	97.7±0.56	97.9±0.57
		1st instar	73.5±0.43	86.6±0.90	97.3±0.42	96.1±0.48
		2nd instar	19.0±3.50	57.1±0.57	22.5±1.90	75.7±0.73
		Total	**54.0±1.40**	**70.5±0.73**	**79.4±0.65**	**78.8±0.59**
	2019	Adult	26.7±2.85	40.6±1.43	41.7±3.49	40.4±3.75
		Crawler	72.0±1.51	91.8±0.91	98.4±0.84	98.7±0.55
		1st instar	71.2±0.52	88.9±2.04	98.2±0.52	97.6±0.66
		2nd instar	29.3±2.75	64.1±1.14	36.9±4.55	76.4±1.21
		Total	**54.3±0.80**	**72.6±0.90**	**81.3±0.70**	**79.4±0.90**
Imidacloprid (Best)	2018	Adult	77.6±2.94	83.8±1.09	87.7±0.96	79.3±0.83
		Crawler	64.7±2.91	86.3±1.24	82.8±2.65	78.1±1.91
		1st instar	80.9±1.40	79.4±2.30	77.2±1.90	74.8±0.54
		2nd instar	54.0±1.80	80.1±1.60	79.7±0.79	77.5±0.74
		Total	**70.7±1.58**	**82.7±1.20**	**85.4±0.86**	**78.5±0.61**
	2019	Adult	84.4±1.71	89.4±0.48	85.7±1.92	79.9±2.37
		Crawler	72.1±1.33	87.7±0.29	81.2±1.39	79.1±1.90
		1st instar	83.9±1.23	81.9±1.24	79.6±1.78	75.8±1.47
		2nd instar	53.0±1.00	84.4±0.73	76.7±2.56	74.0±2.34
		Total	**74.9±0.70**	**85.9±0.20**	**85.9±0.80**	**79.4±1.30**
Spirotetramat (Movento)	2018	Adult	82.3±1.38	89.7±1.06	92.5±1.14	84.5±0.83
		Crawler	75.9±1.31	88.4±0.79	90.6±0.68	83.3±0.77
		1st instar	91.0±0.90	91.3±0.80	93.4±1.40	85.5±1.60
		2nd instar	57.1±3.50	86.6±0.69	81.7±1.87	82.9±1.30
		Total	**78.8±0.35**	**89.1±0.49**	**92.8±0.60**	**84.8±0.51**
	2019	Adult	87.0±1.10	91.3±0.71	94.6±0.50	90.3±0.32
		Crawler	78.0±0.75	90.8±0.30	93.5±0.59	87.0±1.49
		1st instar	91.0±0.44	93.1±0.63	94.8±0.39	90.7±0.29
		2nd instar	59.1±0.64	89.2±0.28	72.6±2.87	83.5±0.54
		Total	**80.5±0.60**	**91.1±0.10**	**94.1±0.20**	**89.0±0.40**
Sulfoxaflor (Isoclast)	2018	Adult	86.0±1.98	90.3±0.67	92.8±0.82	83.6±0.64
		Crawler	77.3±1.73	90.8±0.79	92.2±0.91	83.3±1.53
		1st instar	91.4±0.85	92.2±0.67	93.7±0.37	87.3±1.40
		2nd instar	59.4±1.98	88.6±1.01	80.7±0.94	85.9±2.50
		Total	**80.5±0.62**	**90.6±0.60**	**93.4±0.17**	**85.7±1.25**
	2019	Adult	88.8±1.07	93.9±0.27	96.2±0.45	92.5±0.65
		Crawler	78.0±0.42	92.1±0.57	94.5±0.44	88.3±0.93
		1st instar	92.4±0.45	95.1±0.40	96.4±0.36	93.5±0.85
		2nd instar	64.3±1.77	90.7±1.47	84.3±1.67	88.3±0.95
		Total	**82.3±0.40**	**92.9±0.10**	**95.9±0.20**	**92.1±1.10**
Mineral oil (Kz oil)	2018	Adult	89.0±0.56	95.2±0.25	96.0±0.44	97.1±0.46
		Crawler	80.4±2.0	96.4±0.26	96.6±0.51	97.3±0.34
		1st instar	93.3±0.97	94.4±0.45	97.2±0.31	98.1±0.30
		2nd instar	64.4±2.0	93.3±0.76	83.4±1.99	97.1±0.24
		Total	**83.5±0.66**	**95.0±0.43**	**96.4±0.27**	**97.5±0.22**
	2019	Adult	91.2±1.01	97.2±0.43	98.4±0.45	99.8±0.31
		Crawler	80.6±0.54	96.2±0.45	98.0±0.66	98.7±0.60
		1st instar	94.1±0.75	97.9±0.62	98.7±0.54	99.7±0.37
		2nd instar	68.6±1.06	95.7±0.64	92.4±1.76	98.4±0.36
		Total	**84.9±0.40**	**96.7±0.20**	**98.3±0.40**	**99.2±0.30**

Data are averages of four replicates means± SD   Data are comparing with zero% reduction in control

Table (4): Statistical analysis of the tested insecticides effects on *Aonidiella aurantii* (CRS) stages.

Tested insecticide	Season	Treated Stage	ET_{50} (Days)	95% Conf. Limit	Slope ±SE	γ^2
Pyriproxyfen (Admiral)	**2018**	Adult	> 8			
		Crawler	1.27	0.90 – 1.74	2.68 ± 0.18	0.73
		1st instar	1.04	0.62 - 1.64	2.11 ± 0.15	2.40
		2nd instar	5.25	4.69 – 5.89	2.72 ± 0.11	15.13
		Total	**1.58**	**0.92 – 2.59**	**1.26 ± 0.09**	**0.09**
	2019	Adult	> 8			
		Crawler	1.27	0.91 – 1.72	2.90 ± 0.21	0.55
		1st instar	1.24	0.86 – 1.72	2.60 ± 0.17	1.7
		2nd instar	4.23	3.68 – 4.86	2.13 ± 0.09	7.12
		Total	**1.54**	**0.90 – 2.50**	**1.31 ± 0.09**	**1.61**
Imidacloprid (Best)	**2018**	Adult	0.16	0.006 -2.44	0.81 ± 0.12	0.77
		Crawler	0.43	0.05 – 2.56	0.77 ± 0.094	8.25
		1st instar	0.06	0.0002 – 5.23	0.70 ± 0.14	0.19
		2nd instar	1.39	0.80 – 2.31	1.37 ± 0.091	7.89
		Total	**0.15**	**0.014 – 6.01**	**0.56 ± 0.10**	**4.02**
	2019	Adult	0.05	0.0001 – 6.52	0.69 ± 0.14	0.38
		Crawler	0.43	0.06 – 2.24	0.84 ± 0.10	0.64
		1st instar	0.003		0.46 ± 0.16	0.37
		2nd instar	1.70	1.11 – 2.53	1.48 ± 0.09	0.89
		Total	**0.25**	**0.014 – 2.75**	**0.73 ± 0.10**	**0.39**
Spirotetramat (Movento)	**2018**	Adult	0.16	0.006 - 2.45	0.81 ± 0.12	0.77
		Crawler	0.42	0.08 -1.77	1.02 ± 0.11	6.94
		1st instar	5.96	0.0003 – 5.24	0.70 ± 0.14	0.19
		2nd instar	1.39	0.85 -2.18	1.56 ± 0.10	2.22
		Total	**0.35**	**0.06 – 1.7**	**1.03 ± 0.12**	**0.33**
	2019	Adult	0.05	0.0007 -6.50	0.69 ±0.14	0.38
		Crawler	0.46	0.11 – 1.56	1.21 ± 0.12	0.02
		1st instar	0.002	..	0.43 ± 0.16	0.30
		2nd instar	1.52	0.99 – 2.27	1.63 ± 0.10	0.62
		Total	**0.34**	**0.05 – 1.58**	**1.12 ± 0.13**	**0.12**
Sulfoxaflor (Isoclast)	**2018**	Adult	0.12	0.002 - 2.99	0.76 ± 0.13	0.51
		Crawler	0.47	0.11 – 1.63	1.14 ± 0.12	0.61
		1st instar	0.02	0.0001 - 27.5	0.58 ± 0.15	0.20
		2nd instar	1.37	0.85 – 2.10	1.66 ± 0.10	0.67
		Total	**0.31**	**0.04 – 1.69**	**1.03 ± 0.12**	**0.35**
	2019	Adult	0.07	0.0005 – 3.83	0.84 ± 0.17	0.18
		Crawler	0.54	0.17 – 1.50	1.36 ± 0.13	0.006
		1st instar	0.006		0.56 ± 0.19	0.18
		2nd instar	1.13	0.65 – 1.88	1.65 ± 0.11	0.76
		Total	**0.36**	**0.07 – 1.44**	**1.28 ± 0.15**	**0.44**
Mineral oil (Kz oil)	**2018**	Adult	0.15	0.007 – 1.78	1.12 ± 0.20	0.18
		Crawler	0.69	0.31 – 1.39	1.99 ± 0.19	2.14
		1st instar	0.49	0.0001 – 5.33	1.00 ± 0.23	0.59
		2nd instar	1.45	1.05 – 1.95	2.41 ± 0.13	0.63
		Total	**0.52**	**0.17 – 1.34**	**1.72 ± 0.19**	**0.47**
	2019	Adult	0.38	0.08 – 1.35	1.85 ± 0.33	0.75
		Crawler	0.87	0.49 – 1.46	2.48 ± 0.24	0.61
		1st instar	0.22	0.02 – 1.68	1.63 ± 0.41	0.23
		2nd instar	1.31	0.94 – 1.77	2.74 ± 0.18	0.52
		Total	**0.72**	**0.34 – 1.38**	**2.36 ± 0.29**	**0.17**

ET_{50} (Days) is effective time in days for 50% reduction in each stage.

Sparks *et al*. (2013) added that its effect as nAChRs agonist is in a mannar distinct from other insecticides acting at nAChRs. Spirotetramat insecticide affects the treated CRS different stages as a phloem-mobile systemic insecticide targeting acetyl-CoA carboxylase interrupting the lipid biosynthesis that reduces the fecundity of sucking insects upon foliar applications (Ke *et al*., 2010). So it affects all of the treated insect stages nearly similar as a lipid biosynthesis inhibitor. Pyriproxyphen (admiral) was completely different against adult than the other nymphal and crawler stages because of its mode of action as an insect growth regulator (IGR), which is highly active against California red scale (CRS), and is currently the product of choice for abatement efforts. It sterilizes the adults and causes nymphal mortality. It is fairly selective with 12 hours restricted entry interval. It disrupts the molting process through chitin synthesis inhibition. Since this material affects molting, treatment should be made during peak crawler (1st instar) emergence (Cruz *et al*., 2013). So, this fact explains why its effect was so low against the adult stage comparing with the other treated crawler and nymphal stages.

From the obtained results, Kz oil achieved its effect in systemic arrangement with the time after treatment and its highest effect was continued to 8 weeks after treatment. However, the highest effect was achieved at 6 weeks after treatment in case of the other tested insecticides in both 2018 and 2019 treatments. So the tested insecticides can be arranged according to their effect against the total stages population reduction as Kz oil, sulfoxaflor (Isoclast), spirotetramat (Movento), imidacloprid ((Best) and pyriproxyphen (Admiral), respectively in both treatment seasons. Spirotetramat is an important rotational insecticide with pyriproxyfen for *A. aurantii* control and is an integrated pest management compatible insecticide, effective in reducing *A. aurantii* stages (Cruz *et al*., 2013). However, some of IGR have been shown to be non-selective and are not considered to be IPM friendly but may nevertheless be useful when red scale populations are high and out of biological control.

Worth mentioning, the used insecticides had little or no effect against *A. melinus*, the natural enemy of CRS as spirotetramat at 75 ppm had no negative effect on its egg stage. Residues of spirotetramat; pyriproxyfen and imidacloprid on leaves and twigs collected from a treated citrus orchard allowed 61%, 83% and 95%; 20%, 78% and 95% and 30%, 45% and 94% survival of *A. melinus,* during 1, 2 and 3 weeks after treatment, respectively. Sulfoxaflor also shows a trans-laminar activity and is able to protect plant canopy and undersides leaves (Casida, 2018).

Sulfoxaflor binds to insects nAChRs more strongly than to mammals' ones, so it is much less toxic for mammals and man with a low environmental impact and less aggressive against non-target species (Bacci *et al*., 2018). Sulfoxaflor is classified as non-carcinogenic, non-mutagenic and non-reprotoxic under the European Chemicals Agency (ECHA) classification with no hepatocellular proliferation induction in humans and therefore would not be a human liver carcinogen (LeBaron *et al*., 2013). It is non-volatile, rapidly absorbed in crop leaves, degradable in a few days in soil, not persistent in water and not transferable to groundwater. Its rapid dissipation with absence of residual toxicity makes isoclast a good partner in IPM programs. However, the binding affinity of imidacloprid at the nicotinic receptors in mammals and other vertebrate groups including birds is much less than in insect nicotinic receptors, which cause its less toxicity against human (Tomizawa and Casida, 2005). The blood-brain barrier in vertebrates blocks access of imidacloprid to the central nervous system, reducing its toxicity (Sheets, 2001), which improve its ecosystem communication.

In conclusion from this study, it could be summarized that the used insecticide succeeded in population reduction of the California red scale treated stages (adult, crawler, 1st instar nymphs and 2nd instar nymphs as well as against the total population number in different manners according to their different mode of actions. The tested insecticides were found less toxic on mammals, predators and other non-target biota encouraging us to stress on their entrance the insecticides clique against the examined insect.

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