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RESEARCH ARTICLE

Toxicity and side effects of some insecticides applied in cotton fields on *Apis mellifera*

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Abstract

Honeybee (*Apis mellifera* L.) provides not only bee products of immense value but also render invaluable free service as cross-pollination and propagation of several cultivated and wild species, thereby, maintaining biological diversity. Bee larvae and adults might be killed or suffer various sublethal effects when placed in contact with pollen and nectar contaminated with insecticides. The present work was conducted to investigate the toxicity of seven insecticides on laboratory using oral toxicity test and their side effects on *A. mellifera* in cotton fields. Results indicated that lambda-cyhalothrin was the most toxic-tested pesticide, recording the lowest LC_{50} and LC_{90} values at all tested periods and the lowest LT_{50} and LT_{90} at all tested concentrations, followed by abamectin, spinosad, chlorpyrifos, and emamectin benzoate. On the other side, dipel and pyridalyl recording the highest LC_{50} and LC_{90} at all tested periods and the highest LT_{50} and LT_{90} at all tested concentrations. As for the application of pesticides in cotton fields, the tested pesticides significantly increased the number of dead workers in comparison with control. The tested pesticides significantly decreased bee foraging activities, i.e., number of foraging workers, number of worker collecting nectar, number of worker gathering pollen grains, area of broad workers, and honey bee yields. Dipel and pyridalyl were the most safety pesticides on honey bee workers in laboratory and field, so it could be introduced as a component in IPM programs of cotton pests.

Keywords Toxicity · Side effects · Insecticides and honey bee workers · *Apis mellifera*

Introduction

Honey bee, *A. mellifera*, is the most important pollinator of agricultural crops. In addition, honey bee products are very important to humans. Honey bees, like other living organisms, are exposed continuously to a wide range of biological and non-biological pressures, such as environmental factors, which may interact with each other and affect the health or survival of the insects (Holmstrup et al. 2010; Gonza'lez-Varo et al. 2013).

Spraying of pesticides at the flowering period causes the extreme probability of acute exposure for bees. This can induce direct contamination of flower nectar (Alex and Miles 2011; Thompson 2003), leading to direct poisoning or death of bees, affect the bee larvae, division of labor, and foraging as well as the development of bee colonies.

Dead bees either on the hive bottom boards or on the ground in front of the hives assured the negative effects of chemicals (Winston 1987), thus pesticides in the environment potentially could be transmitted to bees' brood through pollen, wax, or brood food contamination (Villa et al. 2000; Chauzat et al. 2006).

The lethal doses of different pesticides show harmful effects on the behavior of the insect during its return flight, and they affect the insects' sense of orientation and foraging efficacy (Colin et al. 2004). The bee larvae and adults might be killed or suffer many sublethal effects problems with the growth, development, reproduction, and behavior when it was in contact with pollen and nectar contaminated with insecticide (Desneux et al. 2007; Koskor et al. 2009; Garibaldi et al. 2013).

Pyridalyl have low toxicity to the honey bee *A. mellifera*, and it can be safely applied to crops during flowering periods of low or no honey bee activity (Hassona and Kordy 2015; Badawy et al. 2015). The lowest doses of emamectin benzoate showed mortality higher than 90% to bees, 48 h after application Zoclanclounon et al. (2016). Abamectin has an adverse effect and was very toxic on honeybees (*Apis mellifera jemenatica*) (Aljedani 2017). Cypermethrin was harmful to

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the honey bee (*A. mellifera*) and exhibited different poisoning symptoms of bees, which could be useful for beekeepers for the identification of the cause of colony mortality (Pashte and Patil 2018).

From the previous review, this study was conducted to throw the light on the toxicity and side effect of recommended insecticides applied in cotton fields on honey bee colonies.

Materials and methods

Honey bee

This study was carried out at the laboratory of Pesticides Department at the Faculty of Agriculture, Menoufia University. The foraging young worker bees (3–7 days after emergence) used in this experiment were obtained from the apiary of the Economic Entomology Department of the Faculty of Agriculture, Menoufia University and transported to the laboratory in plastic boxes covered with screen, kept at 25 ± 2 °C and 65 ± 5% relative humidity and they were fed on 50% sugar cane solution until used.

Tested insecticides

Microbial insecticides: dipel (B.t. 6.4% WP), abamectin (Vertemic 1.8% EC), spinosad (Tracer 24%SC) and emamectin benzoate (Excellent 1.9% EC).

Pyrethroids: lambda-cyhalothin (Lambada 5% EC).

Oranophosporous: chlorpyrifos as positive control, (Dorsil 48% EC).

New developed insecticides: pyridalyl (Pleo 50% EC).

Laboratory experiments

Honey bee (*Apis mellifera*) adult workers toxicity test, single exposure

Foraging young adult worker bees (3–7 days after emergence) were used for oral toxicity test, where five concentrations of each tested insecticide were prepared in 50% (*w*/*v*) sucrose (sugar cane). Prior to treatment with insecticide, young adult worker bees (10/jar) were anesthetized by exposure to carbon dioxide gas for 1 min. A piece of cotton was saturated with 5 ml of each concentration and attached to the upper side of the jar, then the jars were covered with a piece of muslin and tied with a rubber band. Bees were left to feed on a single dose of each tested insecticide for 24 h, then cotton was removed and replaced with other one contain only 50% sugar cane solution and replaced daily until the end of the test. Control was fed with 50% sugar cane solution. All treatments were replicated three times. Experiments were carried out by incubating bees at 25 ± 2 °C and 65 ± 5% relative humidity, 12:12 (L:D) photoperiod. The bees were considered dead if unable to walk when prodded with a fine hair brush. Mortality percentage were recorded 3, 6, 9, and 12 h and 1, 2, 3, 4, 5, 6, and 7 days of treatment and corrected according to Abbotts formula (1925) then analyzed by probit analysis to calculate the lethal concentrations (LC_{50} and LC_{90}) and (LT_{50} and LT_{90}) with their corresponding fiducial limits at 95% (FL).

Control

Two controls were used in this study, namely, negative and positive control. Negative control (untreated insects) was fed with 50% sugar solution and used in three replicated. Chlorpyrifos was used as a positive control in our study. Chlorpyrifos is well known of its toxicity to honey bees and is classified as highly toxic by direct contact exposure. In addition, consumption of polluted pollen and nectar poses a risk to honey bees (Cutler et al. 2014). Positive control insects were treated exactly as other treatments with tested compounds.

Field experiments

Side effects of tested insecticides on honey bees activities

To study the side effects of seven insecticides on the numbers of dead honey bee and some activities, i.e. foraging workers, number of worker collecting nectar, number of worker gathering pollen grain, area of broad workers, and honey yield under field conditions, eight areas at different locations of El Behira governorate during summer of 2016, each of 12 karat (about 2000 m^2) cultivated with cotton plants. Giza 81 was sprayed with the recommended rate of each tested insecticides at the flowering period to control cotton bollworms using a dorsal sprayer (20 L), while control treatment was sprayed with water only.

For each treatment including control, three bee colonies (each representing a replicate), similar in its strength, size, fed, healthy (i.e., as far as possible disease- and parasite-free), with known history and physiological status, obtained from private apiary, were transferred to about 50 m from the cotton field to study the effect of sprayed pesticides on bee activities.

One, 3, 5, and 7 days after insecticides application, the foraging honeybee workers of the three colonies was recorded, where observation was done at 9 am for 3 min per cotton

flower. Mean numbers of foraging workers were calculated for each treatment and control.

The decrease of foraging percentages was computed for each insecticide using the formula:

$$\text{The decrease of foraging percentages} = ((\text{no.of foraging in control} - \text{no.of foraging in treatment})/(\text{no.of foraging in control})) \times 100 \quad (1)$$

One, 3, 5, and 7 days after insecticide application, the dead honeybee workers in front of the three colonies, also the collecting nectar honey bee workers, in addition, worker gathering pollen were recorded, observation was done at 6 pm, and mean numbers of dead workers were calculated for each treatment.

Gathering pollen honey bee workers were differentiated than collecting nectar honey bee workers by having two baskets full of pollen grain and clear on each of the tibia of the hind leg of bees.

Death percentage was computed for each insecticide using the formula:

$$\text{Death}\% = ((\text{dead workers in treatment} - \text{dead workers in control})/(100 - \text{dead workers in control})) \times 100 \quad (2)$$

$$\text{The decrease of worker collecting nectar percentages} = \left(\begin{array}{r}(\text{no.of worker collecting nectar in control} - \text{no worker collecting nectar in treatment})/\\ (\text{no.of worker collecting nectar in control})\end{array}\right) \times 100 \quad (3)$$

$$\text{The decrease of workers gathering pollen grain percentages} = \left(\begin{array}{r}(\text{no.of workers gathering pollen in control} - \text{no.of workers gathering pollen in treatment})/\\ (\text{no.of workers gathering pollen in control})\end{array}\right) \times 100 \quad (4)$$

At the end of the season (August month), scale of worker brood/colony, pollen grains/trap, and honey production/colony were measured, where the scale of brood/colony ($in.^2$) was determined using a ruler (the wood ruler was used to measure the width and length of the sealed broom area, and the width was multiplied by length to obtain the incubation scale), while pollen grains/trap (g) was weighted, and honey production/colony (kg) was weighted and calculated for each treatment .

The decrease percentages in the scale of worker brood/colony, pollen grains/trap, and honey production/colony were computed for each insecticide using the following formula:

$$\text{The decrease percentages of worker brood scale} = \left(\begin{array}{r}(\text{no.of worker brood scale in control} - \text{no.of worker brood scale in treatment})/\\ (\text{no.of worker brood scale in control})\end{array}\right) \times 100 \quad (5)$$

$$\text{The decrease percentages of pollen/trap} = ((\text{no.of pollen/trap in control} - \text{no.of pollen/trap in treatment})/(\text{no.of pollen/trap in control})) \times 100 \quad (6)$$

$$\text{The decrease percentages in honey yield} = ((\text{no.of honey yield in control} - \text{no.of honey yield in treatment})/(\text{no.of honey yield in control})) \times 100 \quad (7)$$

Table 1 Toxicity of dipel, emamectin benzoate, spinosad, abamectin, chlorpyrifos, pyridalyl, and lambda-cyhalothrin on the honey bee, *Apis mellifera*, workers

Treatments	Periods (h)	LC_{50} (ppm)	Fiducial limits (95%)	LC_{90} (ppm)	Fiducial limits (95%)	Slope ± SE
Dipel	3	3638.9	1450.3–66,539.3	71,032.3	11,065.8–1.5E+8	0.6 ± 0.3
	6	2425.1	1145.1–16,163.8	32,888.9	7449.7–7.7E+6	1.1 ± 0.3
	9	701.8	390.6–1228.0	4397.1	2194.7–19,708.6	1.6 ± 0.4
	12	385.2	206.4–667.3	2595.1	1347.5–8479.8	1.5 ± 0.3
	24	125.4	–	2400.1	–	0.9 ± 0.4
	48	45.8	19.5–99.6	1143.1	435.2–5546.9	0.9 ± 0.2
	72	31.94	12.8–71.4	938.7	345.4–4885.4	0.9 ± 0.1
	96	21.3	8.8–46.1	478.7	188.4–2164.5	0.9 ± 0.2
	120	10.2	3.4–23.8	337.7	122.4–1869.2	0.8 ± 0.1
	144	5.5	1.6–13.4	192.1	68.8–1136.8	0.8 ± 0.2
	168	3.6	0.9–9.3	136.9	48.3–872.2	0.8 ± 0.2
Emamectin benzoate	3	217.5	70.4–6.6E+10	1284.6	194.3–1.09E+19	1.7 ± 0.8
	6	80.1	27.1–1649.2	2359.8	290.8–4.3E+6	0.9 ± 0.3
	9	7.2	2.8–26.1	502.1	97.8–12,466.7	0.7 ± 0.2
	12	1.5	0.1–40.3	92.4	7.8–5.4E+6	0.7 ± 0.2
	24	0.5	0.2–1.3	35.7	9.8–300.2	0.7 ± 0.1
	48	0.5	0.01–21.5	44.1	2.7–1.9 E+8	0.6 ± 0.2
	72	0.3	0.1–0.7	18.4	5.3–135.2	0.7 ± 0.1
	96	0.2	0.004–3.1	23.1	1.6–3.1	0.6 ± 0.2
	120	0.05	0.02–0.1	3.8	1.1–28.5	0.6 ± 0.1
	144	0.03	0.01–0.1	1.9	0.6–12.9	0.7 ± 0.1
	168	100% dead	100% dead	–	–	–
Spinosad	3	153.8	44.7–1346.9	30,242.5	2700.1–7.2E+6	0.6 ± 0.1
	6	61.1	19.4–338.3	13,443.9	1547.9–1.1E+6	0.5 ± 0.1
	9	5.7	2.0–18.0	1082.7	215.5–16,567.8	0.6 ± 0.09
	12	0.8	0.2–2.3	198. 6	43.7–2263.4	0.5 ± 0.1
	24	0.13	0.05–0.3	7.8	2.5–43.3	0.7 ± 0.1
	48	0.04	0.0001–0.2	1.2	0.2–220.9	0.9 ± 0.2
	72	0.02	0.0001–0.2	1.3	0.1–16,822.8	0.7 ± 0.2
	96	0.005	0.001–0.02	0.5	0.1–4.01	0.6 ± 0.1
	120	0.0002	4.5E−7–0.001	0.01	0.003–0.2	0.7 ± 0.2
	144	0.0001	1.2 E−10–0.001	0.01	0.001–0.2	0.7 ± 0.3
	168	100% dead	–	100% dead	–	–
Abamectin	3	119.42	21.7–7607.7	41,912.2	1431.3–7.3E+8	0.5 ± 0.1
	6	42.1	8.2–1228.9	36,185.1	1235.6–2.0E+8	0.4 ± 0.1
	9	2.09	0.6–9.6	851.46	99.7–58,401.9	0.5 ± 0.09
	12	0.6	0.21–1.2	143.7	25.7–3184.3	0.5 ± 0.09
	24	0.1	0.01–1.4	2.66	0.3–9780.4	0.9 ± 0.2
	48	0.03	0.01–0.07	0.8	0.3–4.3	9 ± 0.1
	72	0.03	0.01–0.08	1.2	0.4–6.7	0.8 ± 0.1
	96	0.003	0.001–0.01	0.06	0.02–0.4	0.9 ± 0.1
	120	0.0001	3.9E−13–0.001	0.005	0.001–0.2	0.8 ± 0.3
	144	100% dead	–	100% dead	–	–
	168	100% dead	–	100% dead	–	–
Chlorpyrifos	3	170.6	73.3–2878.1	2855.2	485.9–4.9E+6	1.0 ± 0.3
	6	73.7	30.9–348.4	2724.4	500.5–1.9E+5	0.8 ± 0.2
	9	6.6	0.7–111.1	497.5	44.9–6.2E+6	0.7 ± 0.2
	12	0.9	0.03–15.2	53.7	4.9–5.2E+5	0.7 ± 0.2
	24	0.2	0.01–0.4	6.0	2.2–26.5	0.824 ± 0.111
	48	0.2	0.02–2.01	8.4	1.3–1357.5	0.864 ± 0.197
	72	0.2	0.01–0.4	5.4	2.0–22.7	0.870 ± 0.119
	96	0.1	0.03–0.2	4.3	1.5–20.6	0.768 ± 0.106
	120	0.01	0.004–0.04	1.4	0.4–10.1	0.653 ± 0.107

Table 1 (continued)

Treatments	Periods (h)	LC_{50} (ppm)	Fiducial limits (95%)	LC_{90} (ppm)	Fiducial limits (95%)	Slope ± SE
	144	0.01	0.001–0.02	0.9	0.3–7.2	0.601 ± 0.104
	168	170.6	73.3–2878.1	2855.2	485.9–4.9E+6	1.0 ± 0.3
Pyridalyl	3	2997.7	842.8–1.1E+6	29,915.63	3685.8–1.9E+9	1.3 ± 0.4
	6	1743.7	611.6–1.9E+8	12,489.83	1943.4–1.3 E+14	1.4 ± 0.7
	9	367.1	150.1–2532.5	12,111.5	1950.7–2.4E+6	0.8 ± 0.2
	12	207.8	89.1–1002.9	8989. 7	1554.01–9.9E+5	0.8 ± 0.2
	24	92.9	43.50260.4	3162.0	784.5–80,757.6	0.8 ± 0.2
	48	23.3	0.001–303.7	188.3	43.2–3.5E+18	1.4 ± 0.4
	72	10.7	–	47.4	–	1.9 ± 0.8
	96	7.6	–	49.5	–	1.6 ± 0.5
	120	0.4	0.01–1.2	7. 7	2.8–41.1	0.9 ± 0.3
	144	100% dead	–	100% dead	–	–
	168	100% dead	–	100% dead	–	–
Lambda cyhalothrin	3	38.3	19.6–135.1	522.8	144.1–22,286.5	1.1 ± 0.3
	6	14.3	7.9–3.5	127.7	57.6–695.1	1.4 ± 0.3
	9	1.9	0.7–4.8	135.9	36.2–1881.2	0.7 ± 0.1
	12	0.4	0.07–1.2	62.8	15.1–1340.8	0.6 ± 0.1
	24	0.08	0.01–0.2	2.3	0.9–12.9	0.9 ± .2
	48	0.02	0.0004–0.01	0.8	0.3–6.7	0.8 ± 0.2
	72	0.003	1.0E−38–0.03	0.11	1.6E−7–11.3	0.8 ± 0.4
	96	0.001	–	0.06	–	0.7 ± 0.4
	120	100% dead	–	100% dead	–	–
	144	100% dead	–	100% dead	–	–
	168	100% dead	–	100% dead	–	–

The obtained data were subjected to statistical analysis of variance (ANOVA) at 5% probability, where the measurements were separated using Duncan's multiple range test (DMRT) through CoStat software program (version 6.400 copyright © 1998–2008, n. d).

Results and discussion

Toxicity of tested insecticides against honey bee workers

The toxicity of seven insecticides against honey bee workers after 3, 6, 9, 12, and 24 h and 3, 4, 5, 6, and 7 days of oral treatment are presented in Table 1. The data show that lambda-cyhalothrin was the highest toxic compound, where LC_{50} was calculated as 38.3 and 0.001 ppm after 3 and 96 h of treatment, while, the values of LC_{90} was 522.8 and 0.06 ppm.

After 5, 6 and 7 days of treatment, all treated honeybee workers were dead.

As for abamectin, LC_{50} was 119.4 and 0.0001 ppm after 3 h and 5 days of treatment, while LC_{90} was 41,912.2 and 0.005 ppm. After 6 and 7 days of treatment, all treated honeybee workers were dead.

As for spinosad, LC_{50} was 153.8 and 0.0001 ppm after 3 h and after 6 days of treatment, while LC_{90} was 30,242.5 and 0.01 ppm. After 7 days of treatment, all treated honey bee workers died.

Regarding chlorpyrifos as the positive control, LC_{50} was 170.6 and 0.01 ppm after 3 h and 6 days of treatment, while LC_{90} was 2855.2 and 0.9 ppm. After 7 days from treatment, all treated honeybee workers were dead. Emamectin benzoate achieved LC_{50} value 217.5 and 0.03 ppm after 3 h and 6 days of treatment, while LC_{90} were 1284.6 and 1.9 ppm. After 7 days from treatment, all treated honeybee workers were dead.

Dipel and pyridalyl were less toxic for honey bee workers, where LC_{50} was 3638.9 and 3.6 ppm after 3 h and 7 days of treatment, while LC_{90} was 71,032.3 and 136.9 ppm for dipel, while, LC_{50} of pyridalyl was 2997.7 and 0.4 ppm after 3 h and 5 days from treatment and LC_{90} was 29,915.6.3 and 7.7 ppm. After 6 and 7 days after treated with pyridalyl, all treated honeybee workers were dead.

The lethal times of tested pesticides for orally treated honey bee workers (LT_{50} and LT_{90}) presented in Table 2. For dipel, the LT_{50} and LT_{90} were 323.3 and 2163.9 h at 1 ppm and 4.8 and 10.1 h at 2000 ppm; for emamectin benzoate, are 152.7 and 478.1 h at 0.001 ppm and 5.9 and 11.2 h at 50 ppm; these were 22 and 244 h at 0.001 ppm and 4.4 and 14.5 h at 50 ppm for abamectin.

As for spinosad, LT_{50} and LT_{90} were 73.7 and 258.9 h at 0.001 ppm and 3.5 and 9.4 h at 200 ppm, where these were 171.1 and 839.7 h at 0.001 and 3.2 and 11.7 at 50 ppm for chlorpyrifos; as

Table 2 lethal times of tested insecticides on *Apis mellifera* workers as LT_{50} and LT_{90} (h) together with the corresponding 95% fiducial limits (FL 95%)

Treatments	Conc. (ppm)	LT_{50} (h)	Fiducial limits (95%)	LT_{90} (h)	Fiducial limits (95%)	Slope ± SE
Dipel	1	323.3	192.1–1158.9	2163.9	738.6–39,772.5	1.5 ± 0.4
	10	104.5	83.9–136.7	377.4	250.9–785.3	2.3 ± 0.4
	100	61.7	44.5–90.3	596.9	316.7–1735.6	1.3 ± 0.2
	1000	9.2	7.0–11.7	31.5	23.4–48.9	2.4 ± 0.2
	2000	4.8	3.7–5.8	10.1	8.1–14.6	3.9 ± 0.7
Emamectin benzoate	0.001	152.7	96.5–6220.5	478.1	212.4–1.8E+8	2.6 ± 0.2
	0.01	157.4	78.1–2025.6	1194.9	318.3–2.2E+8	1.5 ± 0.5
	0.1	144.2	62–420.3	929.1	299.4–49,868.8	1.4 ± 0.4
	1	42.8	27–67.8	254.6	138.3–833.7	1.7 ± 0.3
	10	18.9	8.2–13.9	40.8	29.9–61.1	2.2 ± 0.3
	50	5.9	4.8–6.9	11.2	9.2–15.6	4.6 ± 0.8
Abamectin	0.001	22	1.8–42.6	244	126–4659.6	2.5 ± 0.8
	0.01	45.2	26–79.4	215.4	112–985.9	1.9 ± 0.4
	0.1	28.2	18.6–40.8	98.3	69.4–197	2.4 ± 0.4
	1	10.6	8–13.6	39.3	29.1–60.2	2.3 ± 0.3
	10	6.1	4.5–7.8	18.3	13.7–29.8	2.7 ± 0.5
	50	4.4	2.8–5.8	14.5	10.7–24.5	2.5 ± 0.5
Spinosad	0.001	73.2	59–91.4	259	186.3–435.2	2.4 ± 0.3
	0.01	59.6	47.1–75.4	244.2	180–409.9	2.1 ± 0.3
	0.1	42.8	25.2–75.8	222.2	112.6–1035.5	1.8 ± 0.4
	1	12	9.4–14.9	36.3	27.4–54.6	2.7 ± 0.3
	10	7.2	5.5–9	21.3	15.9–34	2.7 ± 0.4
	100	4.4	2.8–5.8	14.8	10.9–25	2.4 ± 0.5
	200	3.5	2.1–4.6	9.4	7.2–15.4	2.9 ± 0.7
Chlorpyrifos	0.001	171.1	87.6–2.4E+5	839.7	252.6–5.02E+12	1.9 ± 0.7
	0.01	124.6	67.9–640.2	670.7	239.2–1.3E+5	1.8 ± 0.5
	0.1	88.7	53.8–192	483	213.7–5119.6	1.7 ± 0.4
	1	31.7	24.3–40.8	158.6	112.1–259	1.8 ± 0.2
	10	8	6.5–9.7	18.3	14.2–28	3.6 ± 0.6
	50	5.9	4.6–7.1	12.8	10.1–19	3.8 ± .7
	100	3.2	1.9–4.5	11.7	8.9–16	2.3 ± 0.3
Lambda-cyhalothrin	0.05	23.6	18.9–29.1	70.9	55.1–99.2	2.7 ± 0.3
	0.5	17.8	14.2–22.1	54.8	41.9–78.6	2.6 ± 0.3
	5	7.2	5.5–8.9	20.2	15.2–32.1	2.9 ± 0.5
	25	4.1	2.5–5.6	14.7	10.8–25.2	2.3 ± 0.5
	50	3.2	1.7–4.3	9	6.8–15	2.8 ± 0.7
Pyridalyl	1	86.2	39.3–237.09	277.9	138.6–30,379.1	2.5 ± 0.9
	10	74.8	33.8–175.9	254.9	127.2–8299.8	2.4 ± 0.8
	50	24.3	19.9–29.5	55.9	44.6–75.9	3.6 ± 0.4
	100	18.6	14.9–22.9	51.5	39.9–73.0	2.9 ± 0.3
	500	8.4	6.5–10.6	25.9	19.3–40.6	2.6 ± 0.4

for lambda-cyhalothrin, these were 23.6 and 70.9 h at 0.05 ppm and 3.2 and 9 h at 50 ppm and these were 86.2 and 277.9 h at 1 ppm and 8.4 and 25.9 h at 500 ppm for pyridalyl.

From the previous results, it could be concluded that lambda-cyhalothrin was the most toxic tested compound showing the lowest LC_{50} and LC_{90} values at all tested periods and lowest LT_{50} and LT_{90} at all tested concentrations, followed by abamectin, spinosad, chlorpyrifos, and emamectin benzoate. In contrast, dipel and pyridalyl were more safe for honey bee workers, where these were the lowest toxic-tested compounds recording the highest LC_{50} and LC_{90} values all tested periods and the highest LT_{50} and LT_{90} at all tested concentrations.

Table 3 The side effect of some insecticides applied on the cotton fields on the number of dead honey bee workers

Treatments	Pre treatment	Post treatment (no. of dead bees/colony)								Mean	
		1 day		3 day		5 day		7 day			
		No.	*D* %	No.	*D* %	No.	*D* %	No.	*D* %	No.	*D* %
Dipel	18	15	10.5	18	11.8	22	17	13	8.4	17.0d	11.9
Emamectin benzoate	9	13	8.4	45	40.9	35	30.9	18	13.7	27.8c	23.5
Abamectin	17	78	76.8	53	49.5	45	41.5	38	34.7	53.5b	50.6
Spinosad	15	72	70.5	61	58.1	52	48.9	45	42.1	57.5 b	54.9
Lambda-cyhalothrin	14	84	83.2	74	72	65	62.8	52	49.5	68.8a	66.9
Chlorpyrifos	10	15	10.5	44	39.7	37	32.9	20	15.8	29.0c	24.7
Pyridalyl	10	20	15.8	25	19.4	35	30.9	12	7.4	23.0cd	18.4
Control	8	5	–	7	–	6	–	5	–	5.75e	–

D % = dead bee worker percentage

The same letters in the column mean no significant difference at 5% level (LSD 5% = 6.3)

Results in (Table 3) presented the number of dead honey bee workers per colony as a side effect of cotton field applications of some insecticides for 7 days.

Statistical analysis of the obtained data revealed that there were significant differences in the number of dead bee workers among all insecticides and control. The highest dead bee worker percentages, after 7 days of application were recorded with lambda-cyhalothrin (66.9%), followed by spinosad and abamectin (54.9 and 50.6%), while the least percentages were recorded at dipel treatment (11.9%) and pyridalyl (18.4%).

Results in (Table 4) show the side effect of insecticides applied to cotton fields on the number of foraging workers for 7 days.

Statistical analysis of the obtained data revealed that there were significant differences in the number of foraging bee workers among all tested insecticides and control except dipel treatment. The highest decrease in foraging bee worker percentages, after 7 days of application, were recorded with lambda-cyhalothrin and spinosad (48.89 and 46.67%), followed by abamectin and chlorpyrifos (37.78 and 35.56%), while the least percentage was recorded at dipel treatment (11.11%).

Results in (Table 5) show the side effect of some insecticides applied on the cotton fields on the number of nectar collecting workers for 7 days.

Statistical analysis of the obtained data revealed that there were significant differences in the number of nectar collecting workers between abamectin, spinosad, and lambada-cyhalothrin and control, while there were no significant differences between dipel, emamectin benzoate, chlorpyrifos, pyridalyl, and control. The highest decrease in nectar collecting worker percentages, after 7 days of application, were recorded with abamectin, spinosad, and lambda-cyhalothrin (21.73%), while the least percentage was recorded at dipel and pyridalyl treatments (4.35%).

Results in (Table 6) show the side effect of some insecticides applied on the cotton fields on the number of workers gathering pollen grains for 7 days.

Table 4 The side effect of some insecticides applied on the cotton fields on the number of foraging workers

Treatments	Pre treatment	No foraging bee workers/colony/3 min						% decrease in bee foraging
		Days post treatment				Total	Mean	
		1 day	3 day	5 day	7 day			
Dipel	9	8	9	10	13	40	10.0ab	11.11
Emamectin benzoate	8	7	7	9	8	31	7.75cd	31.11
Abamectin	6	6	7	8	7	28	7.0cd	37.78
Spinosad	7	7	7	6	4	24	6.0cd	46.67
Lambda-cyhalothrin	5	5	6	7	5	23	5.75d	48.89
Chlorpyrifos	7	7	8	6	8	29	7.25cd	35.56
Pyridalyl	9	9	9	8	6	32	8.0bc	28.89
Control	10	11	10	11	13	45	11.25a	

The same letters in the column mean no significant difference at 5% level (LSD 5% = 2.2)

Table 5 The side effect of some insecticides applied on the cotton fields on the number of nectar collecting workers

Treatments	Pre treatment	Mean no. nectar collecting workers/flower/3 min						% decrease in bee collecting nectar
		Days post treatment				Total	Mean	
		1 day	3 day	5 day	7 day			
Dipel	6	6	5	5	6	22	5.5ab	4.35
Emamectin benzoate	6	6	5	5	4	20	5.0ab	13
Abamectin	4	5	4	4	5	18	4.5b	21.73
Spinosad	5	5	4	4	5	18	4.5b	21.73
Lambda-cyhalothrin	6	5	4	5	4	18	4.5b	21.73
Chlorpyrifos	7	6	5	4	4	19	4.7ab	17.39
Pyridalyl	5	6	5	4	7	22	5.5ab	4.35
Control	6	6	5	5	7	23	5.75a	

The same letters in the column mean no significant difference at 5% level (LSD 5% = 1.1)

Statistical analysis of the obtained data revealed that there were significant differences in the number of workers gathering pollen grains between abamectin, spinosad, lambda-cyhalothrin, and chlorpyrifos and control, while there were no significant differences between dipel, emamectin benzoate, and pyridalyl and control. The highest decrease percentages in workers gathering pollen grains, after 7 days of application, were recorded with lambda-cyhalothrin (50.0%), while the least percentage was recorded at dipel and pyridalyl treatments (11.11 and 16.66%).

Results in (Table 7) show the side effect of some insecticides applied on the cotton fields on the area of worker brood/colony, pollen grains/trap, and honey production/colony.

Statistical analysis of the obtained data revealed that there were significant differences in the area of worker brood/colony between abamectin, spinosad, and lambda-cyhalothrin and control, while there were no significant differences between dipel, emamectin benzoate, chlorpyrifos, and Pyridalyl and control. The highest decrease percentages in the area of worker brood/colony were recorded with lambda-cyhalothrin (22.92%), while the least percentage was recorded at dipel treatment (3.13%).

As for the statistical analysis of the obtained data of collecting pollen grains per trap, there were significant differences in weight of pollen grains between abamectin, spinosad, and lambda-cyhalothrin and control, while there were no significant differences between dipel, emamectin benzoate, chlorpyrifos as positive control, pyridalyl, and control. The highest decrease percentages in pollen grain weights per trap were recorded with lambda-cyhalothrin and spinosad (36.7 and 30.0%), while the least percentage was recorded at dipel treatment (10.0%).

As for the statistical analysis of the obtained data of honey yield per colony, there were significant differences in weight of honey yield between emamectin benzoate, abamectin, spinosad, lambda-cyhalothrin, chlorpyrifos as the positive control, and control, while there were no significant differences between dipel, pyridalyl, and control. The highest decrease percentages in honey yield weights per colony were recorded with lambda-cyhalothrin, spinosad, and abamectin

Table 6 The side effect of some insecticides applied on the cotton fields on the number of workers gathering pollen grain

Treatments	No. of workers gathering pollen grain/flower/3 min							% decrease in bee gathering pollen
	Pre treatment	Days post treatment				Total	Mean	
		1 day	3 day	5 day	7 day			
Dipel	4	6	3	3	4	16	4.0ab	11.11
Emamectin benzoate	2	5	3	3	3	14	3.5ab	22.22
Abamectin	4	5	2	3	3	13	3.25bc	27.78
Spinosad	4	4	3	2	3	12	3.0bc	33.33
Lambda-cyhalothrin	3	3	2	1	3	9	2.25c	50
Chlorpyrifos	4	3	3	4	3	13	3.25bc	27.7
Pyridalyl	3	4	3	4	4	15	3.75ab	16.66
Control	4	6	3	4	5	18	4.5a	–

The same letters in the column mean no significant difference at 5% level (LSD 5% = 1.18)

Table 7 The side effect of some insecticides applied on the cotton fields on some honey bee activities

Treatments	Area of worker brood/colony in.^{2}	% decrease	Pollen grains trap (g)	% decrease	Honey production/ colony (kg)	% decrease
Dipel	465ab	3.13	27ab	10.0	2.2ab	18.52
Emamectin Benzoate	415abc	13.54	24abc	20.0	2.0bc	25.93
Abamectin	405bc	15.63	23bc	23.3	1.8bc	33.33
Spinosad	398bc	17.1	21bc	30.01.73	1.7bc	37.0
Lambda-cyhalothrin	370c	22.92	19c	36.7	1.6c	40.74
Chlorpyrifos	410abc	14.58	24abc	20.0	1.9bc	29.63
Pyridalyl	425abc	11.46	25abc	16.67	2.2ab	18.52
Control	480a	–	30a	–	2.7a	–
LSD 5%	70.1	–	6.1	–	0.5	–

The same letters in each column mean no significant difference at 5% level

(40.74, 37.0, and 33.33%), while the least percentages were recorded at dipel and pyridalyl treatments (18.52%).

Discussion

The obtained results exhibited that lambda-cyhalothrin, abamectin, spinosad, chlorpyrifos as the positive control, and emamectin benzoate was very toxic on *Apis mellifera* compared with pyridalyl and dipel, which were less toxic on *Apis mellifera*. These results are in harmony with those of (Miles et al. 2002; Farooqi et al. 2016) which demonstrated that spinosad was highly toxic to honeybees under laboratory conditions and oral exposure induced the great risk. Costa et al. (2014) and Aljedani (2017) found that abamectin has an adverse effect and was very toxic on honey bees (*Apis mellifera jemenatica*). Zoclanclounon et al. (2016) recorded that the lowest doses of emamectin benzoate, showed mortality higher than 90% to bees, 48 h after application. Li et al. (2017) found that chlorpyrifos was toxic for *A.mellifera*, where the LD_{50} was 103.4 ng/bees. Also, Dai et al. (2017) found that the toxicity of the tested pesticides to honey bee larvae was ranged from most to least toxic as chlorpyrifos (as positive control) > fluvalinate > coumaphos = imidacloprid > amitraz. Pashte and Patil (2018) found that cypermethrin was harmful to the honey bee (*A. mellifera*) and exhibited different poisoning symptoms of bees, which could be useful for beekeepers for the identification of the cause of colony mortality.

Natural products have a lower persistence in the environment; therefore, they were considered environmentally and toxicologically safer than several of the currently used organosynthetic pesticides (Copping and Menn 2000; Duke et al. 2010). Hassona and Kordy (2015) found that tested biopesticides, achook and dipel, 2× exhibited low toxicity on the honey bee, *Apis mellifera* L. Badawy et al. (2015) suggested that pyridalyl have low toxicity to the honey bee *A. mellifera*, and it can be safely applied to crops during flowering periods of low or no honey bee activity.

Bee larvae and adults might be killed or suffer by various sublethal effects when placed in contact with pollen and nectar contaminated with insecticides (Desneux et al., 2007; Koskor et al. 2009). Among the sublethal effects caused by insecticides are problems with the growth, development, reproduction, behavior, on the environment and non-target organisms such as bees (Desneux et al. 2007; Biondi et al. 2012; and Decourtye et al. 2013).

Conclusion

The obtained results exhibited that lambda-cyhalothrin, abamectin, spinosad, chlorpyrifos, and emamectin benzoate were very toxic to honey bee and showed harmful side effects on the number of dead honey bee workers, number of foraging workers, number of nectar collecting workers, nectar collecting workers, area of worker brood/colony, pollen grains/trap, and honey production/colony workers gathering pollen grains. In contrast, pyridalyl and dipel were safe on honey bee worker and showed less harmful side effects compared with other tested insecticides.

Author contribution I am a single author, so, I conceived this research and designed experiments; design and interpretation of the data; carried out experiments and analysis; wrote the paper; and participated in the revisions.

The author read and approved the final manuscript.

Compliance with ethical standards

Conflict of interest The author declares that there is no conflict of interest.

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