Zagazig J. Agric. Res., Vol. 47 No. (1) 2020 65-70

Plant Protection and Pathology Research

http:/www.journals.zu.edu.eg/journalDisplay.aspx?Journalld=1&queryType=Master

ACUTE TOXICITY OF SOME INSECTICIDES ON HONEYBEE, *Apis mellifera* L.

Mona H.I. Radwan[1*], R.E. Sand[2] and M.A. Hendawy[3]

1. Nat. Res. Dept., Inst. Asian Studies and Res., Zagazig Univ., Egypt

2. Plant. Prot. Res. Inst., ARC, Giza, Egypt

3. Plant. Prot. Dept., Fac. Agric. Zagazig Univ., Egypt

Received: 03/09/2019; Accepted: 14/11/2019

ABSTRACT: This work was conducted to evaluate the toxicity of some insecticides against honeybee workers under laboratory conditions. Among the tested insecticides, Deltamethrin was the most toxic compound followed by Methomyl while Chlorpyrifos was the least one followed by Profenfos after 24, 48, and 72 hr., of topical exposure. The results indicated that there was negative relationship between the time post treatment and lechal concentration (LC_{50}) values of all the tested insecticides. The LD_{50} values were 9.5, 10.45, 3.68, 18.33 and 12.55 µg/bee for Methomyl, Cyhalothrin, Deltamethrin, Chlorpyrifos and Profenfos, respectively after 24 hr., of exposure. In this respect, the toxicity of the insecticide Deltamethrin was 2.58 times more than Methomyl, 2.83 times than Cyhalothrin, 4.98 times than Chlorpyrifos and 3.41 times than Profenfos. On the other hand, the toxicity of Methomyl was 1.93 times, more than Chlorpyrifos 1.37 times than Profenfos and 1.1 times than Cyhalothrin. The toxicity of the tested insecticides against the workers of honeybee after 48 hr., of topical exposure showed LD_{50} values range from 2.44 to 12.32 µg/bee. The insecticide Deltamethrin was the highest toxic compound where as the Chlorpyrifos was the least toxic one. The other insecticides occupied intermediate degree of toxicity. Deltamethrin toxicity in comparision to the other tested insecticides revealed that it was more toxic than Methomyl by 2.21 times, 2.93 times than Cyhalothrin, 5.01 times than Chlorpyrifos and 3.82 times than Profenfos. The LD_{50} of Deltamethrin was 1.01 mg/l, 0.1mg/l to Methomyl, 5.21 µg/bee to Cyhalothrin, 7.58 mg/l to Chlorpyrifos and 6.11 mg/l to Profenfos. On the other hand, the corresponding LD_{90} were 2.31 to Deltamethrin, 8.71 to Methomyl, 9.98 to Cyhalothrin, 13.75 to Chlorpyrifos and 15.73 µg/bee to Profenfos. The tested insecticides could be arranged descendingly as follows: Deltamethrin > Methomyl > Cyhalothrin > Profenfos > Chlorpyrifos.

Key words: Insecticides, toxicity, honeybee, *Apis mellifera*.

INTRODUCTION

Pesticides are used to protect agricultural crops, however, some may affect bee health, with their toxicity depending on the active substance used and the formulation of different pesticide products. The risk of influencing bee health also increases when, amongst other potential factors, legal requirements or good practice for pesticide application have not been followed. Bees can be exposed to pesticides when spray drifts onto non target fields of crops in bloom, or nearby wildflowers or beehives, contaminating pollen and nectar. In cases where exposure is suspected, it is necessary to analyse dead bee samples and identify the cause of death and the pesticides possibly involved (**Kozowicka, 2013).**

Insecticides are important for ensuring both crop quality and quantity in today's integrated crop management for sustainable agricultural production. The use of insecticides is one of the most effective practices to control pests.

***Corresponding author: Tel. :** +201008322384
E-mail address: mona_hir2005@yahoo.com

However, what concerning us most is how residual levels of sub-lethal dosages of those insecticides being used lead to detrimental effects on non-target pollination species of honeybee and its development, foraging behavior and colony conditions. Either wild or domesticated honeybee, *Apis mellifera*, has been recognized and used as a major pollinator in the agricultural system **(Kevan, 1999)** and by beekeepers to produce valuable products such as honey, royal jelly and pollen. However, honeybee rely on flower plants while foraging and collecting its food sources of nectar and pollen and thus at risk endangering exposing to various levels of chemical residues of pesticides while they are collect nectar and pollen **(Peach *et al.,* 1993)**.

Honeybee workers may be poisoned by the residual pesticides on the nectar and pollen they collect. In addition, the workers may take the pesticide-contaminated nectar and pollen back to their hive. This will expose the larvae, drones and queen to these pesticides, and eventually poison them and causes high mortality.

Feeding honeybee larvae on contaminated nectar and pollen transmitted from the sprayed fields to the hive may also be considered another destructive agents to the honeybee colony, there is scarle data about insecticides toxicity in honey bee brood of neem oil and some insect growth regulators as well as a few number of canceled pesticides **(Erickson, 2013; Pashte and Patil, 2017)**.

From this stand point the present work was designed to assess the topical, as well as the initial and residual activity of some pesticides against honeybee workers.

MATERIALS AND METHODS

Chemicals Used

Carbamate compound

Methomyl [Lannate 90% WP]. Ethanimidothioic acid, N-[methylamino carponyloxy] thio metyl.

The recommended field rate is 300 grams/ faddan.

Organophosphorus compounds

A- Chlorpyrifos [Dursban EC 48%].

Phosphorothioic acid, O.O-diethyl O-[3,5,6-trichloro -2-pyridinyl] ester.

The recommended field rate is 1 liter/faddan.

B- Profenofos [Selecron EC 72%].

O-[4-bromo-2-chlorophenyl] O-ethyl –S-propyl phoshphorothioate.

The recommended field rate is 750 ml/faddan.

Synthetic Pyrethroid Compounds

A- Cyhalothrin (Lambda EC 5%). cyano-3-phenoxybenzy1-3-(2-chloro-3,3,3-trifluoropropenyl) -2,2dimethylcyclopropane-carboxylate.

The recommended field rate is 750 ml/faddan.

B- Deltamethrin (Decis EC 2.5%). (S)-a-cyano-m-Phenoxybenzyl (1R 3R)-3-(2,2 dimethyl cyclopeopane carboxylate).

The recommended field rate is 750 ml/faddan.

Rearing of Insect

The 2nd of honeybee workers needed for laboratory tests were collected from the peripheral combs of the colony. To minimize the genetic variations as possible, tested workers were collected from the honey chamber (hive) of one colony headed by open mated F1 Carniolan queen from the educational Apiary of Faculty of Agriculture, Zagazig University

Laboratory Experiments

Acute toxicity of the tested insecticides

Topical application technique

The toxicity of the tested insecticides against honeybee workers was evaluated using the topical application method (**Stevenson**, **1968**). Bees of approximately identical weight and age were slightly anaesithized by chilling (5 minutes in deep freezer). One µl acetone solution of insecticidal dilution was administered individually on the thoracic mesonotum using the syringe of micro applicator. The control workers were treated with acetone only. The uoses used in this study were 1, 10 and 30 µg/ bee for Methomyl and Cyhalothrin, 0.1, 1 and 10 µg/bee for Deltamethrin and 1, 20 and 50 µg/bee for Chlorpyrifos and Profenofos.

At least three concentrations and 30 workers were used for each insecticide after application, bees of each concentration were placed together in small feeding cages of 10×10×15 cm and fed on 1:1 sugar syrup. The tests were carried out during late summer (2017) at room temperature (25 – 27ºC), and RH (60-68%).

The corrected mortality of bees was carried out using Abbott's formula **(Abbott, 1925)**. The LC_{50}, LC_{90} and slop values of the tested compounds were calculated using Finney equation **(Finney, 1971)** through soft ware computer program.

Toxicity index and relative potency calculated according to Sun equations **(sun, 1950)** as following:

Toxicity index =

$$\frac{LC_{50} \text{ or } LC_{90} \text{ of the most efficient compound}}{LC_{50} \text{or } LC_{90} \text{of the tested compound}} \times 100$$

Relative potency =

$$\frac{LC_{50} \text{ or } LC_{90} \text{ of the tested compound}}{LC_{50} \text{or } LC_{90} \text{of the most efficient compound}} = \text{fold}$$

RESULTS AND DISCUSSION

Efficacy of the Tested Insecticides Against Honeybee Workers Under Laboratory Conditions

After 24 hour.

The results presented in Table 1 reveald that the toxicity of five insecticides; *i.e.* Methomyl**,** Cyhalothrin**,** Deltamethrin, Chlorpyrifos and Profenfos against the 2nd honeybee workers, *Apis mellifera* L for 24 hr., exposure time. Among the tested insecticides Deltamethrin was the most toxic compound followed by methomyl while Chlorpyrifos was the least one followed by Profenfos.

The results indicated that there was negative relationship between the time post treatment and $\mathbf{LD_{50}}$ values of all the tested insecticides. The $\mathbf{LD_{50}}$ values were 0.95, 1.045, 0.36, 1.83 and 1.25 µg/bee for Methomyl, Cyhalothrin, Deltamethrin, Chlorpyrifos and Profenfos after 24 hr., of exposure.

In this respect, the toxicity of the insecticide Deltamethrin was more than Methomyl by 2.58 times, 2.83 times than Cyhalothrin, 4.98 times than Chlorpyrifos and 3.41 times than Profenfos. On the other hand, the toxicity of Methomyl was more than Chlorpyrifos by 1.93 times, 1.37 times than Profenfos and 1.1 times than Cyhalothrin.

After 48 hour

The toxicity of the tested insecticides against the workers of honeybee after 48 hr., were recorded in Table 2. The $\mathbf{LD_{50}}$ values ranged between 0.24 and 1.23 µg/bee. The insecticide Deltamethrin was the highest toxic compound while the Chlorpyrifos was the lowest toxic one. Other insecticides occupied intermediate toxicity. Results showed that toxicity of Deltamethrin was more than Methomyl by 2.21 times, 2.93 times than cyhalothrin, 5.01 times than chlorpyrifos and 3.82 times than Profenfos.

After 72 hour

Rasults presented in Table 3 show that the LD_{50} of Deltamethrin was 0.1 µg/beel, 0.62 µg/ bee to Methomyl, 0.51 µg/bee to Cyhalothrin, 0.75 µg/l to Chlorpyrifos and 0.61 µg/l to Profenfos. The corresponding LD_{90} were 0.23 to Deltamethrin, 0.81 to Methomyl, 0.99 to Cyhalothrin, 1.37 to Chlorpyrifos and 1.57 to Profenfos.

The tested insecticides could be arranged descendingly as follows: Deltamethrin > Methomyl > Cyhalothrin > Profenfos > Chlorpyrifos.

In connection to our finding, **Benedek (1983)** found that the direct toxicity to honeybee against 5 synthetic pyrethroid insecticides, penthrin, chinthnn, cypermethrin, fenvalerate and deltamethrin was high. **Kasamatsu and Kawachi (1985), Erickson (2013) and Pashte and Patil (2017)** reported that the LD_{50} value of deltamethrin recorded 0.004 µg/bee.

Wilkinson *et al.* (1986) reported that cyhalothrin (Kur.) is toxic to honeybees in laboratory tests. **Mayer *et al.* (1990)** found that the LD_{50} value of fenvalerate for honeybees was 0.471 µg/g body weight and **El-Ansary and El-Zogby (1992)** stated that the LD_{50} of deltamethrin (Decis) was 0.014 µg /bee being greatly higher than that recorded in the present work. This variation may be due to the varied sensitivity of the varied races treated. Also, lab. conditions surely are different. In addition they did not give any information about the formulation they tested.

Table 1. Acute toxicity of the tested insecticides on honeybee workers 24 hr., after treatment

Insecticides	LD_{50} ug/bee	LD_{90} ug/bee	Slope	Toxicity index	Relative potency
Methomyl	0.95	1.63	1.66	38.47	2.58
Cyhalothrin	1.045	2.21	1.01	35.22	2.84
Deltamethrin	6.36	7.8	1.53	100	1
Chlorpyrifos	1.83	2.8	0.89	20.08	4.98
Profenfos	1.25	3.3	1.22	29.32	3.41

Table 2. Acute toxicity of the tested insecticides on honeybee workers 48 hr., after treatment

Insecticides	LD_{50} ug/bee	LD_{90} ug/bee	Slope	Toxicity index	Relative potency
Methomyl	0.54	1.221	1.06	45.19	2.21
Cyhalothrin	0.71	1.51	1.55	34.17	2.93
Deltamethrin	0.24	0.46	1.88	100	1
Chlorpyrifos	1.23	1.73	2.11	19.81	5.01
Profenfos	0.93	0.71	1.78	26.18	3.82

Table 3. Acute toxicity of the tested insecticides to honeybee workers 72 hr., after treatment

Insecticides	LD_{50} ug/bee	LD_{90} ug/bee	Slope	Toxicity index	Relative potency
Methomyl	0.62	0.81	1.33	30.79	3.25
Cyhalothrin	0.51	0.99	0.98	19.39	5.16
Deltamethrin	0.1	0.23	1.38	100	1
Chlorpyrifos	0.75	1.37	1.21	13.32	7.50
Profenfos	0.61	1.57	1.55	16.53	6.05

In this respect, **Matar (1996)** recorded LD_{50} and LD_{90} values (µg/bee) of fenvalerate (0.0030 and 0.029). **Ebadah (1998)** recorded LD_{50} and LD_{90} values (µg/bee) as following cyhalothrin (Karate) (0.026 and 0.082), cyhalothrin (Karate super) (0.0207 and 0.0858) and fenpropathrin (0.0345 and 0.1844), respectively.

Also, **Wael and Van (1989)** reported that fenvalorate (Sumicidin), alpha cypernelhrin (Pastac) and tenpropathrin (Danitol) were highly toxic to honybee in feeding. Moreover, **Ebadah (1998)** found that the LC_{50} for cyhalothrin (Karate) and tenpropathrin by ingestion on honybee workers were 25.68 and 37.38 ppm., respectively. Similarly, **Gromisz and Cromisz (1996)** reported that Bulldock (beta-cyfluthrin) is highly toxic to bees when ingested the lethal dose of the active ingridient a.i. 0.2-0.3 ppm/bee.

The severe toxicity of synthetic pyrethroids by ingestion to honeybee was reported by many authors (**Arzone and Patctta, 1982; Arzone and Vidano, 1985; Atallah *et al.,* 1989; Matar, 1996; Ebadah, 1998**).

It is obvious that within the same group there are highly toxic and less toxic compounds. Moreover, the two formulations (Karate and Kendo) of the same compound (Cyhalothrin) showed varied oral toxicity to honeybee workers. The differed toxicity of the formulations was reported by **Atallah *et al.* (1989)**. Their variations could be attributed to the synergistic, dispersing and solved materials added to the formulations which increased or decreased toxicity.

Resutts of the present work are in agrement with those of **Arzone and Patctta (1982)** who reported that, fenpropathrin was highly toxic stomach poisons to honeybee. Also, **Arzonc and Patctta (1987, 1991)** reported that flucythrinate was moderately toxic on ingestion and cyfluthrin was markedly toxic in feeding to honeybee.

REFERENCES

Abbott W.S. (1925). A method of computing the effectiveness of an Insectied. J. Entomol., 18 265-267.

Arzone, A. and A. Patctta (1982). Studies on the action of chlorpyrifos methyl.dimethoate and fenvalerate on honeybee. Cooperative Libraria Univ. Hditrice Bologna, 89 -96. Univ. Torino, Italy.

Arzone, A. and A. Patctta (1987). Research on the action of flucythrinate. propiconazole and quinalphos on honeybees. \picoltore Moderno, 179 -186.

Arzone, A. and A. Patetta (1991). Effects of clofentazin, cyfluthrin, fentinaccetate, methamidophos and penconazol on the honeybee. Noticiaro Sulle Malatie delle Piante, 112: 68 -75.

Arzone, A. and C. Vidano (1985). Research on the action of three pyrethroid insecticides on the honeybee. Apicoltore Moderno 76, (6): 203 - 207. (A.A. 660/87).

Atallah, M.A., Z.A. Zeitoun and A.R. Hassan (1989). The relative toxicity of some synthetic pyrethroid and carbamate insecticides on worker honeybees *(Apis mellifera* L.) in the laboratory. J. of the Entomological Society of British Colombia 84, 39-451.

Benedek, P. (1983). Toxicity of synthetic pyrethroid insecticides to honeybees. 10th International Congress of Plant Protection. Vol. 2. British Crop Protection Council; Croydon, UK.

Ebadah, I.M.A. (1998). Effect of some insecticides on honeybee *(Apis mellifera* L.). Ph.D. Thesis, Fac. Agric., Zagazig Univ., Egypt.

El-Ansary, O. and F. El-Zogby (1992). Toxicity and residual hazards of some insecticides to honeybees. Alex. J. Agric. Res., 37 (3): 259-275.

Erickson, B. (2013). Regulation: Europe bans three neonicotinoids linked to honeybee population declines. Chem. Eng. News Archive 91, 11. (doi: 10.1021/cen-09118-notw9).

Finney, D.J. (1971). Probit Analysis 3rd Ed. Cambridge Univ., 3330.

Gromisz, Z. and M. Gromisz (1996). Laboratory investigations on the sensitivity of honeybees to harmful effects of the Bulldock formulation. Pszczelnic Zeszyty Naukwe, 40 (1): 161 -162. A.A. 275/98.

Kasamatsu, K. and K. Kawachi (1985). Effects of fenvalerate on honeybees in flowering plants. In Proc. 3rd Int. Cong. Apiculture, Nagoya 1985. Nagoya, Japan; Apimondia, 233-237 (A.A.277/88).

Kevan, P.G. (1999). Pollinators as bioindicators of the state of the environment: species, activity and diversity. Agric., Cosystems and Environ., 74: 373-393.

Kozowicka, B. (2013). Pesticides responsible for bee poisoning: new screening technique proposed. Ecotoxicol. and Environ. Safety, 97: 10–22.

Matar, A.M.M. (1996). Effect of bollworms control on honeybees. *Apis mellifera* L. M. Sc. Thesis, Fac. Agric., Zagazig Univ., Egypt.

Mayer, D.F., E.R. Muticzky and J.D. Lunden (1990). Esfenvalerate insecticide and domesticated pollinators. Bee Sci., 1 (1): 33-39, A.A. 645/91.

Pashte, V.V. and G.S. Patil (2017). Evaluation of persistence of insecticide toxicity in honey bee (*Apis mellifera* L). Indian J. Biochem. and Biophysics, 54: 150 – 155.

Peach, M.L., V.J. Tepedino, D.G. Alston and T.L. Griswold (1993). Insecticide treatments for rangeland grasshoppers: potential effects on the reproduction of Pediocactus sileri (Englem.) Benson (Cactaceae). Proceedings of the Southwestern Rare and Endangered Plant Conf., 1993: 309–313.

Stevenson, M.D. (1968). Laboratory studies on the acute and cral toxicities of insecticides to honeybees Ann Appl. Biol., 61(3): 476- 472.

Sun, Y.P. (1950). Toxicity index. An improved method of comparing the relative toxicity of insecticides. J. Econ. Entomol., 43: 45- 53.

Wael, L. and L.O. Van (1989). Toxicity and the repellent activity of synthetic pyrethroids towards the honeybee *(Apis mellifera* L.). Proceeding of the XXXI[st] International Congress of Apic. Warsaw, Poland. August 19-25. 1987. 209- 216.Bucharest, Apimondia House Romania.

Wilkinson, W., H.J. Cough and I.G. Collins (1986). PP32 1-Safety to honeybees *(Apis mellifera)* when used in cereals.

السميـــة الحـــــاده لبعض المبيـــدات الحشـــــريـــة علـــي نحـــل العســل

منى حسن إبراهيم رضوان[١] ـ رضا عليوة سند[٢] ـ محمد عبد العال هنداوى[٣]

١- قسم الموارد الطبيعية – معهد الدراسات والبحوث الآسيوية – جامعة الزقازيق – مصر

٢- معهد بحوث وقاية النباتات – مركز البحوث الزراعية – مصر

٣- قسم وقاية النبات – كلية الزراعة – جامعة الزقازيق – مصر

يهدف هذا البحث إلى دراسة سمية بعض المبيدات الحشرية علي شغالات نحل العسل تحت الظروف المعملية وهذه المبيدات هي المبيد الكارباماتى ميثوميل والمبيدات الفسفورية كلوريرفوس وبروفينوفوس ومبيدات البيرثرويدات لمبادا سيهالوثرين ودلتا مثرن وذلك بعد فترات تعرض ٢٤، ٤٨، و ٧٢ ساعة من المعاملة وذلك عن طريق المعاملة الموضعية. أظهرت النتائج أن المبيد الحشري دلتامثرين كان اكثر المبيدات سمية متبوعا بمبيد الاجرنت والبستبان والتي كانت أقل سمية وفى المركز الاخير مبيد الاكتلك وذلك بعد ٢٤، ٤٨ ، ٧٢ ساعة كانت قيم جرعة التصفية القاتلة LD_{50} هي ٠,٩٥، ١٠,٤٥، ٠,٣٦، ١,٨٣ و١,٢٥ ميكروجرام/ النحلة على التوالى لمبيدات اجرنت واللمبادا دلتامثرين وبستبان وبرفينوفوس بعد ٢٤ ساعة من التعرض، طبقا لما سبق كانت سمية المبيد الحشري دلتامثرين اكثر من مبيد بستبان بمقدار ٤,٩٨ ضعف و ٤,٤١ ضعف مبيد البرفينوفوس، بعد ٤٨ ساعة فأن قيم جرعة التصفية القاتلة تراوحت ما بين ٢,٤٤ – ١٢,٣٢ جزء فى المليون كان المبيد الحشري دلتامثرين اكثر المبيدات سمية في حين كان المبيد بستبان اقل المبيدات كفاءة وباقي المبيدات احتلت مراكز وسطية بين المبيد الأعلي والأقل. يمكن ترتيب المبيدات المختبرة تنازليا طبقا لسميتها كما يلى دلتامثرين > ميثوميل > سيهالوثرين > برفينوفوس > كلوريرفوس.

المحكمـــــون:

١- أ.د. حمزه محمد السيد الشرقاوي أستاذ الحشرات الاقتصادية – كلية التكنولوجيا والتنمية – جامعة الزقازيق.

٢- أ.د. علـــى أحمـــد علـــى أيـوب أستاذ وقاية النبات – كلية الزراعة – جامعة الزقازيق.