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Impact of different groups of pesticides on the population dynamic of honey bee (*Apismelifera* L.)

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Authors'Contribution **Ali, A.** designed research experiments and executed by**G.Parwaiz**. **S. A.Memon,M.A. Roonjho**, **A. H.Mastoi&G. A.Bugti**.**M. S.Arain, J.Zaib, S. Rajput and C.Yasir**collected and analysed the data.

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ABSTRACT

Losses of honey bees and decline in bee colonies are becoming continuous threat to the apicultural industry, such as food security and environmental stability. Decline the population of honey bees in the field of agriculture, insecticides could be the possible factor for global colony losses. The aim of present research was to assess the impact of selected pesticides from different groups. Such as pyrethroid, organophosphate and botanical pesticides was used to compare with control group. Botanical extracts were used with 3 concentrations (5%, 10%, and 20%). The maximum mortality % of honey bees was observed on synthetic pesticides groups (organophosphate and pyrethroid), chlorpyrifos, trichlofon, and imidacloprid on (100%) after that profenofos treatment was associated with a high mortality rate (98.3%), Similarly pyrethroid group of pesticides lambda cyhalothrin (92%), clothianidin (87.3%), cypermethrin (80.3%) and bifenthrin (69.3%) as compared with control and botanical extracts. However on the plant base pesticides a maximum mortality % of honey bees was observed on Azadirachtaindica20% (17.6%) and stramonium leaves 20% (15%) as contrast with control group mortality rate (3.3%). Maximum repellency % of honey bees was observed on both (organophosphate and pyrethroid) groups of pesticides as compared with their respected control groups. While a less repellency % of honey bees was observed on all 3 concentrations of both botanicals plant based pesticides as compared with control. It is concluded that synthetic pesticides have lethal effects on the survival and repellency percentage of honey bees as compard with botanical pesticides.

Keywords: Pyrethroid, organophosphate, botanical plant, mortality, repellency

INTRODUCTION: Insect's as pollinators are important for obtaining maximum production in most cultivated agriculture crops. Bees has been used worldwide agriculture crops pollinations (Garibaldi *et al.*, 2013), and their pollination service yield considerable which gives cost-effective benefits for the crop productions. Unfortunately, the honey bee's population has reduced all over the globe (Lebuhn *et al.*, 2013). Various aspects have been observed as the possible reason of this reduction, such as synthetic insecticides (Van der Sluijs *et al.*, 2013). Honey bees mature and immature stages might be killed numerous toxic influence when located in contact with pollen and nectar infected with pesticides (Desneux *et al.*, 2007). Between the toxic influences caused by pesticides are problems with honey bees life cycles and reproduction (Desneux *et al.*, 2007). Changes of behavioral activities in honey bees such as movement, orientation and communications might be the cause pesticides influence (Belzunces *et al.*, 2012).The impact of pesticides on the honey bees growth and development can be defined as pupal malformation and failure of adults emergence (Koskor *et al.*, 2009). Mainly the problem caused by synthetic pesticides on the ecosystem and non target organisms for example honey bees(Biondi *et al.*, 2012; Decourtye *et al.*, 2013) has stimulated the use of natural products as an substitute insect pest management, these natural products have a minor persistence in the ecosystem and, therefore are calculated eco-friendly and lethal safer than various of the presently used synthetic insecticides (Copping and Menn, 2000; Duke *et al.*, 2010). In addition to toxic influence(Vendramini *et al.*, 2012; Hsu *et al.*, 2013) of natural products such as plant base pesticides, might have repellent effects, inhibit fecundity and vary the feeding and hormonal systems of various arthropod insect pest (Copping and Menn, 2000; Isman, 2006). These characteristics force the use of bio-pesticides attractive for the insect pest managements(Isman, 2006). Botanicals pesticides Neem (azadirachtin) have a range of commercial formulations that show signs of excellent effectiveness more than 400 hundred insect pest species (Ntalli and Menkissoglu-Spiroudi, 2011) and mites(Flamini, 2003). Others plant base pesticides for example andiroba oil, citronella oil, eucalyptus oil, garlic extract, and rotenone has also been suggested for managements (Moreira *et al.*, 2006; Mureithi, 2008). But the authorization still controversial to use these natural products globally. Because the activity of the plant base insecticides on agriculture insect pest is well established, their impact on non target organisms such as honey bees needs to be examined more comprehensively.

OBJECTIVES: Keeping in view above facts, current research aimed is to investigate the effects of different groups of selected pesticides organophosphate (profenofos, chlorpyrifos, trichlorfon, imida-cloprid), pyrethroid groups (clothianidin, lambdacyhalo-thrin, cypermethrin, bifenthrin) and botanical plant extract (neem and dhaturaleaves) on the behavioral activities of honey bees.

MATERIAL AND METHODS: The present research was carried out at Laboratory of Entomology, Faculty of Agriculture, Lasbela University of Agriculture Water and Marine Sciences(LUAWMS), Uthal Baluchistan. Culture of honey bee (*Aphis melifera* L.) was purchased from mianey forest form Hyderabad and established on the artificial diet (sugar solution). Upon their established and acclimatization, bees were released in natural environment for the collection of pollen and nectar from different flowers. About 4 pesticides (clothianidin, lambda-cyhalothrin, cypermethrin, bifen-thrin) were used from pyrethroid groups, similarly 4 pesticides (profenofos, chlorpyrifos, trichlorfon andimidacloprid) were used organophosphate groups and 2botanical pesticides (neem and dhaturaleaves) were used to compare with synthetic group of pesticides. However, in control group, no any pesticide was used except artificial diet. All synthetic pesticides were used according to the recommendation of pesticide manufacturing companies and 2 botanical plant extracts were used with 3 concentrations (5%, 10%, and 20%). The present experiment was conducted at25 ± 2°Ċ with 60 ± 5 relative humidity in arandomized complete design (RCD). Mortality and survival % was recorded after 3h, 6h, 12h, 24h, 48h and 72 h. While the data on repellence % of honey bee were recorded after 24 h. andeach experiment was replicated thrice. Neem and datura leaves (10 kg each) were collected separately and later on these leaves was chopped and boiled in 10L water until 5 L boiled water is left which was drained through muslin cloth and prepared it for further use. Three different concentrations 5%, 10% and 20% of botanical plant extracts was used. Preparation of botanical plant extracts, method suggested by Chang *et al.* (2013);Rajput *et al.* (2017) were used.

About 100 adult bees were collected from colony and transferred in the plastic cups for exposure of 3h, 6h, 12h, 24h, 48h and 72h) along with the provision of pesticide treated artificial diets to find the mortality % and survival % of bee adults honey bees.Yellow colour of flowers were collected from the field and dipped in treated artificial diet with synthetic and botanical pesticides and after that 500 hundred honey bees were collected from the already established culture. Bees were exposed on yellow coloure flowers with pesticides treated to check the repellency % of honey bees. Control was kept individually with each treatment and datea was recorded after 24 hours.

Statistical analysis: One-way analysis of variance (ANOVA) was used with the help of SPSS software and entire means were compared with Tukey Test at level $p<0.05$ level.

RESULTS: A maximum mortality % of honey bees on clothianidin

pesticides was observed after 12h. (16.7±0.17),followed by 48h (16±1.27), 24h (15.3±1.13), 3h (14.7±0.13) and 6h (13.33±0.12), while a minimum mortality % was observed on 72h (11.4±1.37). Similarly, a highest mortality % on lambda cyhalothrin pesticides was observed after 3 h (22±0.18), followed 6h (16±0.2), 12h (15.3±1.17), 48h (14±1.28) and 24h (13±1.80), however a lowest mortality % was observed on 72h (11.7±1.09). A maximum mortality % on cypermethrin pesticides was found after 12h (18±0.87), following by 6h (17.7±1.98), 3h (14.7±1.22) 24h (11±0.19) and 48 h (10.3±0.15), but a minimum mortality % was noticed on 72h (8.7±1.29). Similarly, a peak mortality % was observed by bifenthrin pesticides after 3h (15.3±0.77), followed by 12h (13.7±067), 24h (13±1.29), 6h (12.7±0.87) and 48h (08.7±1.38), while a minimum mortality% was observed on 72h (06±1.27). Statistically significant (P>0.05) difference was found among all given treatments of Pyrethroid group of pesticides on the mortality % of honey bees (table 1).

Treatments	Clothianidin	Lambda Cyhalothrin	Cypermethrin	Bifenthrin
3h	14.66±0.13b	22.00±0.18a	14.66±1.22b	15.33±0.77a
6h	13.33±0.12bc	16.00±0.20b	17.66±1.98a	12.66±0.87b
12h	16.66±0.17a	15.33±1.17b	18.00±0.87a	13.66±067b
24h	15.33±1.13ab	13.00±1.80bc	11.00±0.19c	13.00±1.29b
48h	16.00±1.27a	14.00±1.28c	10.33±0.15c	08.66±1.38c
72h	11.33±1.37d	11.66±1.09d	8.66±1.29d	06.00±1.27d
Mortality %	87.31b	91.99a	80.31c	69.31d

Table 1: Effects of different pyrethroid groups of pesticides on the mortality % of *A. melifera*.
Values (Mean ± SE) in given colum letters are significantly different by Tukey test (P<0.05).

A maximum mortality % on profenofos was observed after 3 h (28.3±0.17), following by 6h (25.7±0.13), 12h (14.7±016), 48h (12.7±1.29) and 24h (10±018), while a minimum mortality % was observed on 72h (7±1.25).Similarly, a highest mortality % on chlorpyrifoswas observed after 6h (28.3±1.27), following by 3h (27±1.26),12h (17±1.29), 48h (15±1.99) and 24h (12.7±1.35), in present result we observed that all honey bee bees has been died before reaching 72h. A maximum mortality % on trichlorfonwas found after 3h (34.3±1.77), followed by 6h (31.7±1.33), 12h (27±1.22) and 24h (7±1.44), we observed that hundred mortality % on trichlorfon just 3 to 24 h. Similarly, a peak mortality % was observed by imidacloprid after 3h (29±1.34), followed by 6h (24.7±0.23), 12h (22±0.17), 24h (21±0.27) and 48h (3.3±0.45), in present result we observed that all honey bee bees has been died before reaching 72 h. Statistically no significant (P<0.05) changes was noticed among all given treatments of (OP) group of pesticides on the mortality % (table 2).

Treatments	Profenofos	Chlorpyrifos	Trichlorfon	Imidacloprid
3h	28.33±0.17a	27.00±1.26a	34.33±1.77a	29.00±1.34a
6h	25.66±0.13b	28.33±1.27a	31.66±1.33b	24.66±0.23b
12h	14.66±016c	17.00±1.29b	27.00±1.22c	22.00±0.17c
24h	10.00±018e	12.66±1.35d	7.00±1.44d	21.00±0.27c
48h	12.66±1.29d	15.00±1.99c	00.00±1.88e	3.33±0.45d
72h	7.00±1.25f	00.00±0.19e	00.00±1.38e	00.00±0.33e
Mortality %	98.31b	100.00a	100.00a	100.00a

Table 2: Effects of organophosphate groups on mortality % of *A. melifera*.
Values (Mean ± SE) in given colum letters are significantly different by Tukey test (P<0.05).

A maximum mortality % of honeybees on neem leaves (5%)was observed after 12h (2.7±0.27), followed by 48h (2.3±1.77), 6h (2±1.99), 72h (1.3±0.15) and 24h (1±0,37), while a minimum mortality % was observed on 3h (0.3±1.22). Similarly, a highest mortality % on neem leaves (10%) was observed after 24h (4.7±0.55), followed by 12h(3.3±0.88), 48 h (3±1.36) and 72h (2.7±1.28), however a lowest mortality % was observed on 3h (1.7±0.33) and 6h (1.7±1.99). A peak mortality % was observed by neem leaves (20%) after 72 h (4.7±0.77), followed by 24h (3.7±0.33), 48h (3.3±0.55), 12h (2.7±0.44) and 6h (2.3±1.56), but a minimum mortality % was observed on 3h (1±1.25). Statistically significant (P>0.05) variation was noticed Between 5 % of neem leave extract as compared with 10 and 20% of neem leaves extract, while no statistically significant (P<0.05) changes was found between 10% and 20% of neem leaves extract on the mortality % of honey bees (table 3). The highest mortality % was observed in plant extract (5%) on 12h (3±0.27) followed by 24h (2.7±0.25), 48h (1.3±0.28) and 6h (1±0.22) , while no mortality was recorded at 3h and 72h. Similarly, the maximum mortality % was found on 48h, (3±1.22) when it is exposed on 10% dhaturaafter that 72h (2.7±1.33) followed by 24h (2.3±1.44), 12h (2±1.34) and 6h (1.3±0.88), while no mortality was noticed on 3h. A peak mortality%was recorded after 72h (4.3±1.340) when honey bees was treated with 20% dhatura leaves, after that 24h (4±0.33), following by 48h (2.7±0.26), 12h (2.3±0.22) and 6h (1±0.01), however no mortality %was observed on 3h. Statistically significant (P>0.05) changes was found among all given 3 concentrations ofdhatura leaves (table 4).The highest mortality % in control group was observed after 72 h (1.7±0.66), followed by 24h (1.3±0.33) and 48 h (0.3±0.10). However no mortality was observed on 3h, 6h, and 12h. Statistically no significant (P<0.05) variation was found among all given times of treatments(table 5).

Treatments	Neem 5%	Neem 10%	Neem 20%
3h	0.33±1.22b	1.66±0.33c	1.0±1.25cd
6h	2.00±1.99a	1.66±1.99c	2.33±1.56bc
12h	2.66±0.27a	3.33±0.88ab	2.66±0.44bc
24h	1.00±0,37ab	4.66±0.55a	3.66±0.33ab
48h	2.33±1.77a	3.00±1.36ab	3.33±0.55ab
72h	1.33±0.15ab	2.66±1.28b	4.66±0.77a
Mortality %	9.65b	16.97a	17.64a

Table 3: Effects of neem on the mortality % of *A. melifera*.
Values (Mean ± SE) in given colum letters are significantly different by Tukey test (P<0.05).

Treatments	Dhatura 5%	Dhatura 10%	Dhatura 20%
3h	00.00±0.01c	00.00±0.01b	0.00±0.01bc
6h	1.00±0.22c	1.33±0.88b	1.00±0.012bc
12h	3.00±0.27a	2.00±1.34ab	2.33±0.22b
24h	2.66±0.25ab	2.33±1.44ab	4.00±0.33a
48h	1.33±0.28bc	3.00±1.22a	2.66±0.26b
72h	00.00±0.01c	2.66±1.33ab	4.33±1.34a
Mortality %	7.99c	11.32b	14.32a

Table 4: Effects of dhatura on the mortality %of *A. melifera*.
Values (Mean ± SE) in given colum letters are significantly different by Tukey test (P<0.05).

Teatments	Artificial diet without any insecticides (Control)
3h	0.00±0.00ab
6h	0.00±0.00ab
12h	0.00±0.00ab
24h	1.33±0.33a
48h	0.33±0.10ab
72h	1.66±0.66a
Mortality %	3.32

Table 5: Effect of control group on the mortality % of *A. melifera*.
Values (Mean ± SE) in given colum letters are significant different by Tukey test (P<0.05).

The highest mortality rates were observed on synthetic pesticides groups (organophosphate and pyrethroid), chlorpyrifos, trichlofon, and imidacloprid on (100%) after that profenofos treatment was associated with a high mortality rate of (98.3%), followed by lambda cyhalothrin (91.9%), clothianidin (87.3%), cypermethrin (80.3%) and bifenthrin (69.3%) as compared with control and botanical plant base pesticides. Similarly a maximum mortality % was observed on plant base pesticides neem 20% (17.6%), neem 10% concentration (16.9%) following by dhatura leaves at a 20% concentration (14.9%), dhatura(10%) concentration (11.3%) neem 5% (9.6%), and dhatura5% (7.9%) as contrast with control group mortality rate (3.3%). Statistically significantly (P>0.05) difference was found among all treatments (organophosphate, pyrethroid and botanicals) when it is compared with control groups, while no statistically significant (P>0.05) difference was observed among organophosphate group (imidacloprid, trichlofon and chlorpyrifos) and neem (20% and 10%)(table6).

Treatments	mortality % of *A. melifera*
Clothianidin	87.31d
Lambda Cyhalothrin	91.99c
Cypermethrin	80.31e
Bifenthrin	69.31f
Profenofos	98.31b
Chlorpyrifos	100.00a
Trichlofon	100.00a
Imidacloprid	100.00a
Neem leaves 5%	9.65j
Neem leaves 10%	16.97g
Neem leaves 20%	17.64g
Dhatura leaves 5%	7.99k
Dhatura leaves 10%	11.32i
Dhatura leaves 20%	14.98h
Control	3.33%l

Table 6: Effect of different pesticide on mortality % of honeybees.
Values (Mean ± SE) in given colum letters are significant different by Tukey test (P<0.05).

The highest survival % was observed in the control group (96.7%)

as compared with different groups of synthetic and non synthetic pesticides. However among 3 groups of pesticides, (organophosphate, pyrethroid and botanicals),neem leaves 5% (90.4), 10% (83), 20% (82.4) and dhatura5% (92), 10% (88.7), 20% (85.7) was found maximum survival % as compared with organophosphate and pyrethroid. A maximum survival % was noticed on pyrethroid group's pesticides, bifenthrin (30.7%) cypermethrin (19.7%), clothianidin (13%) and lambda cyhalothrin (8%) as compared to organophosphate groups of pesticides. No survival % of honey bees was observed on imidacloprid, trichlofon, chlorpyrifos and profenofos(figure 1).

Figure 1: Survival % of honey bees against imidacloprid, trichlofon, chlorpyrifos and profenofos.

Statistically significant (P>0.05) was seen among all given treatments and compared with control groups, while no statistically significant (P>0.05) difference was observed among imidacloprid, trichlofon and chlorpyrifos(table 7).

Treatments	**Survival % of *A. melifera***
Clothianidin	12.96h
Lambda Cyhalothrin	8.01i
Cypermethrin	19.69g
Bifenthrin	30.69f
Profenofos	1.69j
Chlorpyrifos	0.00k
Trichlofon	0.00k
Imidacloprid	0.00k
Neem leaves 5%	90.35bc
Neem leaves 10%	83.03e
Neem leaves 20%	82.36e
Dhatura leaves 5%	92.01b
Dhatura leaves 10%	88.68cd
Dhatura leaves 20%	85.68d
Control	96.68%a

Table: 7. Effect of different groups of pesticides on the survival % of *A. melifera*.

Values (Mean ± SE) in given Colum letters are significant different by Tukey test (P<0.05).

A maximum repellency % of honey bees was observed in trichlorfon (97%) followed by imidacloprid (95%), chlorpyrifos (95%), profenofos (90%), clothianidin (87%),lambda cyhalothrin (85%), cypermethrin (72%), bifenthrin (70%), neem 20% (65%), 10% (63%) and5% (60%), dhatura 20% (60%), 10% (55%), and 5% (51%) as compared with their control groups. Statistically significantly (P>0.05) difference was seen on all given treatments Organophosphate, Pyrethroid and Botanicals base pesticides when it is contrasted with control groups (figure 2).

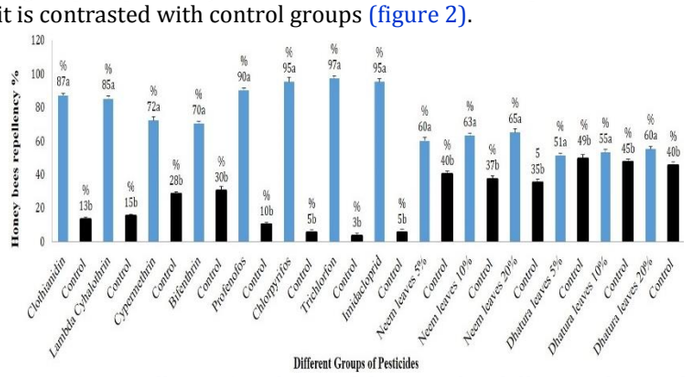

Figure 2: Repellency % of *A. melifera* on the different chemicals group of pesticides.

DISCUSSION: As pollinators of flowering crops, honey bees play a important role regarding primary production system, as well as being an essential primary industry themselves. Uses of agro-chemicals, such as insecticides, make honey bees susceptible to toxic and death. Bothe farmers and beekeepers can take initiative to help encourage harmless pollination and control the risk of bees poisoning. Abay*et al.* (2023) reported that synthetic groups of pesticides have lethal effect on the behavioral and physiological activities of honey bees. This implies that the chemicals mode of application and contact duration of the honey bees to the insecticides incidents were differently affecting the honey bees longevity (Ostiguy *et al.*, 2019). In present research we observed that, honey bees are more significant vulnerable on toxic occurrence of the insecticides when ingested the insecticides through fumes or body contact assessment. The maximum mortality rate of tested bees noticed in present research trial might be indicating that the way of poison occurrence that has been causing colony losses and decline in Pakistan. Highest mortality on organophosphate and pyrethroid group of pesticides indicate that these insecticides incidents reason significant honeybee mortality via spray, which might be attributed to their fumy properties. These consequences are agreed with the results of Dawit Melisie *et al.* (2015), who reported that various substance such as diazinon 60% EC have potential to volatize even at room temperature. The use of plant base pesticides has currently recommended as an substitute pest management technique, mainly in agriculture crop systems where the conventional synthetic pesticides have limited use, for example in agroecological agricultural and natural farming systems (Duke *et al.*, 2010).*Azadirachtaindica* chemicals have lethal effects on the behavioral activates on honey bees. We Noticed that the influence of plant base insecticides such as Dhatura and neem extracts, which had been earlier explained as "harmless" for A. melifera(Naumann and Isman, 1996; Riedl, 2006). In present research we noticed that Dhatura and neem extracts resulted in a less toxicity for the adults of A. melifera, which indicating that use of pesticides should be avoided during flowering stage of crops because at that time A. melifera visited to plants for pollen and nectar. While Melathopoulos *et al.* (2000) did not found any harmful influence of *Azadirachtaindica* on adult honey bees, they found that this pesticides decreased the quantity of larvae in colonies and, at sub lethal doses, differ malformations occurred when the bees emerged from the cocoons. Rembold and Lackner (1981) observed that *Azadirachtaindica* oil extracts were severely lethal to young one of bees. Efrom *et al.* (2012) observed a significant enhance in the death rate of honey bees adult workers honey bee with an augmented exposure time of the bees to various concentrations of Neem extract.Koskor *et al.* (2009) examined that a chronic treatment with sub lethal amount of pesticides influenced the pollen foraging of bumble bees. The repellent impact of plants extracts on honey bees was also found by Nicodemo and Nogueira Couto (2004). Naumann *et al.* (1994) noticed that foragers were deterred from feeding on sugar solutions with extremely low application of neem extract, no significant decline was found in the foraging bees in canola fields sprayed with neem pesticides. Likewise, we found the repellent consequence of plant base pesticides on *A. melifera* foragers. The field trial to assess the foraging behavior of bees is essential to understand the repellent result of pesticides on honey bees. The use of repellent compound may be exploited to decrease the ability of accidental insecticides toxicity in honey bees. Repellent products may be incorporated in prepared solutions with synthetic insecticides to decrease the contact of bees with these insecticides (Solomon and Hooker, 1989). While, it is essential to highlight that these repellent products should not use severe toxicity on honey bees. Otherwise, if the repellent results last for a long time, they can influence the foraging activities and produce a state of starvation or lacking of dietary at the individual or colony (Belzunces *et al.*, 2012). In addition, influence of Pesticides as repellent results the number of bees foraging on flowers, which may lead not only to insufficient nectar and pollen assembling but also to lacking crop pollination. In additionally declined the walking activities of adults honey bees might be the cause of both synthetic and non synthetic pesticides. All the pesticides changed the foraging behavior by being repellent to adult workers of honey bees, So, uses of synthetic and non synthetic insecticides for the management of insects on agriculture crops should be implement with caution. Likewise, the possible hazard of the side effects of these insecticides to non target organisms such as pollinators should be evaluated.

CONCLUSION: Present results indicate that the synthetic groups of pesticides have lethal effects on the survival and repellency % of honey bees in the field as compared to botanical extract. There is

dire need to mitigate the use of pesticides specially flowering stages because at that time honey bees visits for pollen and nectar.

CONFLICT OF INTEREST: The authors declare that they have no conflict of interest that affects the publication of this article.

LIFE SCIENCE REPORTING: In current research article no life science threat was reported.

ETHICAL RESPONSIBILITY: This manuscript is original research, and it is not submitted in whole or in parts to another journal for publication.

INFORMED CONSENT: The author(s) have reviewed the whole manuscript and approved the final version of the manuscript before submission.

REFERENCES:

Abay, Z., A. Bezabeh, A. Gela and A. Tassew, 2023. Evaluating the impact of commonly used pesticides on honeybees (*Apismellifera*) in North Gonderof Amhara Region, Ethiopia. Journal of toxicology, 2023(1): 2634158.

Belzunces, L. P., S. Tchamitchian and J. L. Brunet, 2012. Neural effects of insecticides in the honey bee. Apidologie, 43: 348-370.

Biondi, A., V. Mommaerts, G. Smagghe, E. Vinuela, L. Zappalà and N. Desneux, 2012. The non-target impact of spinosyns on beneficial arthropods. Pest management science, 68(12): 1523-1536.

Chang, A., R. Khuhro, B. Yousufzai, K. Dhiloo and I. Rajput, 2013. Efficacy of neem kernal powder and oil against cauliflower insect pests under field conditions. International journal of life sciences, 7(1): 2848-2852.

Copping, L. G. and J. J. Menn, 2000. Biopesticides: A review of their action, applications and efficacy. Pest management science: Formerly pesticide science, 56(8): 651-676.

DawitMelisie, D. M., T. D. Tebkew Damte and A. Thakur, 2015. Effects of some insecticidal chemicals under laboratory condition on honeybees [*Apismellifera*L.(Hymenoptera: Apidae)] that forage on onion flowers. African journal of agricultural research, 10: 1295-1300.

Decourtye, A., M. Henry and N. Desneux, 2013. Overhaul pesticide testing on bees. Nature, 497(7448): 188-188.

Desneux, N., A. Decourtye and J.-M. Delpuech, 2007. The sublethal effects of pesticides on beneficial arthropods. Annual review of entomology, 52(1): 81-106.

Duke, S. O., C. L. Cantrell, K. M. Meepagala, D. E. Wedge, N. Tabanca and K. K. Schrader, 2010. Natural toxins for use in pest management. Toxins, 2(8): 1943-1962.

Efrom, C. F. S., L. R. Redaelli, R. N. Meirelles, 2012. Side-effects of pesticides used in the organic system of production on *Apismellifera*Linnaeus, 1758. Brazilian archives of biology technology, 55: 47-53.

Flamini, G., 2003. Acaricides of Natural origin, personal experiences and review of literature (1990-2001). Studies in natural products chemistry, 28: 381-451.

Garibaldi, L. A., I. Steffan-Dewenter, R. Winfree, M. A. Aizen, R. Bommarco, S. A. Cunningham, C. Kremen, L. G. Carvalheiro, L. D. Harder and O. Afik, 2013. Wild pollinators enhance fruit set of crops regardless of honey bee abundance. Science, 339(6127): 1608-1611.

Hsu, W.-S., J.-H. Yen and Y.-S. Wang, 2013. Formulas of components of citronella oil against mosquitoes (*Aedesaegypti*). Journal of environmental science health, Part B, 48(11): 1014-1019.

Isman, M. B., 2006. Botanical insecticides, deterrents, and repellents in modern agriculture and an increasingly regulated world. annual review of entomology, 51(1): 45-66.

Koskor, E., R. Muljar, K. Drenkhan, R. Karise, A. Bender, E. Viik, A. Luik and M. Mänd, 2009. The chronic effect of the botanical insecticide neem ec on the pollen forage of the bumble bee *Bombusterrestris*L. Agronomy research, 7: 341-346.

Lebuhn, G., S. Droege, E. F. Connor, B. Gemmill-Herren, S. G. Potts, R. L. Minckley, T. Griswold, R. Jean, E. Kula and D. W. Roubik, 2013. Detecting insect pollinator declines on regional and global scales. Conservation biology, 27(1): 113-120.

Melathopoulos, A. P., M. L. Winston, R. Whittington, H. Higo and M. Le Doux, 2000. Field evaluation of neem and canola oil for the selective control of the honey bee (Hymenoptera: Apidae) mite parasites *Varroajacobsoni* (acari: Varroidae) and *Acarapiswoodi* (acari: Tarsonemidae). Journal of economic entomology, 93(3): 559-567.

Moreira, M. D., M. C. Picanço, E. d. Silva, S. C. Moreno, J. C. Martins, M. Venzon, T. de Paula and A. Pallini, 2006. Uso de inseticidas botânicos no controle de pragas. Controle alternativo de pragas e doençasViçosa: EPAMIG/CTZM: 89-120.

Mureithi, J., 2008. Use plant pesticides to control crop pests. Kenya Agricultural Research Institute, Kenya.

Naumann, K., R. W. Currie and M. B. Isman, 1994. Evaluation of the repellent effects of a neem insecticide on foraging honey bees and other pollinators. The canadianentomologist, 126(2): 225-230.

Naumann, K. and M. B. Isman, 1996. Toxicity of a neem (azadirachta indica a. Juss) insecticide to larval honey bees. American bee journal, 136(7): 518-520.

Nicodemo, D. and R. Nogueira Couto, 2004. Use of repellents for honeybees (*Apismellifera*L.) in vitro in the yellow passion-fruit (*Passifloraedulisdeg*) crop and in confined beef cattle feeders. Journal of venomous animals toxins including tropical diseases, 10: 77-85.

Ntalli, N. G. and U. Menkissoglu-Spiroudi, 2011. Pesticides of botanical origin: A promising tool in plant protection. Pesticides-formulations, effects, fate: 1-23.

Ostiguy, N., F. A. Drummond, K. Aronstein, B. Eitzer, J. D. Ellis, M. Spivak and W. S. Sheppard, 2019. Honey bee exposure to pesticides: A four-year nationwide study. Insects, 10(1): 13.

Rajput, I. A., T. S. Syed, G. H. Abro, I. Khatri and A. M. Lodhi, 2017. Effect of different plant extracts against pink bollworm, *Pectinophoragossypiella*(Saund.) larvae on BT. And non-bt. Cotton. Pakistan journal of agricultural research, 30(4): 373-379.

Rembold, H. and B. Lackner, 1981. Rearing of honeybee larvae in vitro: Effect of yeast extract on queen differentiation. Journal of apicultural research, 20(3): 165-171.

Riedl, H., 2006. How to reduce bee poisoning from pesticides. Oregon State University Extension Service.

Solomon, M. and K. Hooker, 1989. Chemical repellents for reducing pesticide hazard to honeybees in apple orchards. Journal of apicultural research, 28(4): 223-227.

Van der Sluijs, J. P., N. Simon-Delso, D. Goulson, L. Maxim, J.-M. Bonmatin and L. P. Belzunces, 2013. Neonicotinoids, bee disorders and the sustainability of pollinator services. Current opinion in environmental sustainability, 5(3-4): 293-305.

Vendramini, M. C. Mathias, A. U. De Faria, K. C. S. Furquim, L. P. De Souza, G. H. Bechara, 2012. Action of andiroba oil (*Carapaguianensis*) on *Rhipicephalussanguineus* (Latreille, 1806)(acari: Ixodidae) semi-engorged females: Morphophysiological evaluation of reproductive system. microscopy research technique, 75(12): 1745-1754.