EFFECTS OF PESTICIDES APPLICATION ON HONEY BEES

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ABSTRACT

Pollination through honeybees is one of the valuable ecosystem services that we are availing. Bees are pollinators of our important crops like brassicas, sunflower, safflower, apple, pear, blackberry, raspberry, okra, grapes etc. Growing evidence for declines in bee populations has caused great concern because of the valuable ecosystem services they provide. Neonicotinoid insecticides have been implicated in these declines because they occur at trace levels in the nectar and pollen of crop plants. Neonicotinoids are a relatively new class of insecticides that share a common mode of action that affect the central nervous system of insects, resulting in paralysis and death. They include e*imidacloprid, acetamiprid, clothianidin, dinotefuran, nithiazine, thiacloprid and thiamethoxam*. Neonicotinoids are systemic pesticides. Unlike contact pesticides, which remain on the surface of the treated foliage, systemics are taken up by the plant and transported to all the tissues (leaves, flowers, roots and stems, as well as pollen and nectar). Like nicotine, the neonicotinoids act on certain kinds of receptors in the nerve synapse. They are much more toxic to invertebrates, like insects, than they are to mammals, birds and other higher organisms. Recent research has suggested a potential toxicity to honey bees and other beneficial insects even with low levels of contact. Neonicotinoids may impact bees' ability to forage, learn and remember navigation routes to and from food sources.

Key words: Neonicotinoids, CCD,Toxicity

1.1 Introduction:

Pollination is the one of the valuable ecosystem services that benefits mankind in various ways. Insects are one of the important pollinators among biotic agencies. Bees are pollinators of our important crops like brassicas, sunflower, safflower, apple, pear, blackberry, raspberry, okra, grapes etc. Growing evidence for declines in bee populations has caused great concern because of the valuable ecosystem services they provide.There are several reasons for decline in bees populations that leads to the Colony Collapse Disorder(CCD).

CCD is an unusual decrease and disappearance in their honeybee colonies. It seemed as if thousands of honeybees were vanishing into thin air, including many worker bees that are vital to the colonies' survival and prosperity. As more and more of the worker bees disappear, their colonies become weak and soon, they are no longer able to function. Due to the collapse of the colonies, this phenomenon is properly named the ***Colony Collapse Disorder (CCD)*.**

There are many proposed causes for this syndrome:

- The use of pesticides and insecticides, such as neonicotinoids
- The influx of the varroa mite
- The spread of diseases and viruses
- Poor nutrition
- Habitat loss; and stress factors, such as migratory stress.

1.2 What are Neonicotinoids?

Neonicotinoids – first introduced in the mid-1990s - are the fastest growing and most heavily used class of insecticides in the United States. As of 2011 (the most recent year for which there is publicly available data), approximately 3.5 million pounds of neonicotinoids were applied annually to nearly 127 million acres of agricultural crops compared to about 1.6 million pounds used in 2005. Having experienced a two-fold increase in use in a five-year period, and accounting for roughly 25% of the global agrochemical market, neonicotinoids comprise the most widely used class of insecticides in the world today. The neonicotinoid family of includes acetamiprid, clothianidin, imidacloprid, nitenpyram, nithiazine, thiacloprid and thiamethoxam.

Table 1. Cross-reference list of common, trade, and chemical names of neonicotinoid insecticides.

Common name	**Trade names**	**Chemical name**
Acetamiprid	Acetamiprid®, Assail®, Tristar®	(E)-N-(6-chloro-3-pyridinyl)methyl)-N1-cyano-N-methylacetamidine
Clothianidin	Acceleron®, Arena®, Belay®, Celero®, Clutch®, Nipsit Inside®, Poncho®	(E)-1-(2-chloro-1,3-thiazol-5-ylmethyl)-3-nitroguanidine
Dinotefuran	Alpine®, Dinotefuran®, Safari®, Scorpion®,	N-methyl-N'-nitro-N"-[(tetrahydro-3-furanyl) methyl]guanidine
Imidacloprid	Admire®, Advantage®, Gaucho®, Merit®, Premise®, Touchstone®	1-(6-chloro-3-pyridin-3-ylmethyl-N-nitroimidazolidin-2-ylidenamine
Thiamethoxam	Cruiser®, Platinum®	3-(2-chloro-1,3-thiazol-5-ylmethyl)-1,3,5-oxadiazinan-4-ylidenene(nitro)amine

1.3 Mode of action of neonicotinoids

In recent years, neonicotinoids have been the fastest-growing class of insecticides in modern crop protection, with widespread use against a broad spectrum of sucking and certain chewing pests. Due to their systemic nature, neonicotinoids are taken up by the roots or leaves and trans-located to all parts of the plant, making the treated plant toxic to insects, as well as causing lethal and sublethal adverse impacts on other invertebrates and some vertebrates. (van der Sluijs et al 2014; Pisa et al 2014).

The mode of action of neonicotinoid pesticides is modeled after the natural insecticide, nicotine. They act on the central nervous system of insects. Their action causes excitation of the nerves and eventual paralysis, which leads to death. Because they bind at a specific site (the postsynaptic nicotinic acetylcholine receptor), they are not cross-resistant to the carbamate,

organophosphate, or synthetic pyrethroid insecticides, which was an impetus for their development. As a group, they are effective against sucking insects, but also chewing insects such as beetles and some Lepidoptera, particularly cutworms.

There is disturbing evidence that these neonics are making their way into our food and water supply. A study by the U.S. Geological Survey found that neonics are widespread contaminants of surface and groundwater that could be a source of drinking water. Limited testing from the U.S. Department of Agriculture has found neonics in fruits and vegetables, where the pesticide's systemic nature means it cannot be washed off the surface of these foods.

1.4Toxicity

Neonicotinoids are classified by the EPA as both toxicity class II and class III agents and are labeled with the signal word "Warning" or "Caution." Because the neonicotinoids block a specific neuron pathway that is more abundant in insects than warm-blooded animals, these insecticides are more selectively toxic to insects than mammals.

The most available toxicity data of the neonicotinoids is with imidacloprid. These data indicate that it is less toxic when absorbed by the skin or when inhaled compared to ingestion. It causes minor eye reddening, but is non-irritating to the skin. Signs of toxicity in rats include lethargy, respiratory disturbances, decreased movement, staggering gait, occasional trembling, and spasms. There are no accounts of human poisoning, but signs and symptoms of poisoning would be expected to be those similar for rats.

Table 2. Neonicotinoid pesticide wildlife toxicity ranges

Common name	**Bird acute oral LD**50 **(mg/kg)***	**Fish LC**50 **(ppm)****	**Bee LD**50†
Acetamiprid	PNT	PNT	MT
Clothianidin	PNT	PNT	HT
Dinotefuran	PNT -MT	PNT	HT

Imidacloprid	MT	MT	HT
Thiamethoxam	ST	PNT	HT

*Bird LD50: Practically nontoxic (PNT) = > 2,000; slightly toxic (ST) = 501 – 2,000; moderately toxic (MT) = 51 – 500; highly toxic (HT) = 10 – 50; very highly toxic (VHT) = < 10.
**Fish LC50: PNT = > 100; ST = 10 – 100; MT = 1 – 10; HT = 0.1 – 1; VHT = < 0.1.
†Bee: HT = highly toxic (kills upon contact as well as residues); MT = moderately toxic (kills if applied over bees); PNT = relatively nontoxic

1.5 Residues in bee-collected pollen, bees, honey and wax

Neonicotinoid residues in plants and plant parts only become of importance for bees once they are exposed. The most relevant measures of exposure are the concentrations in bee-collected plant materials, such as pollen, bee products like bee bread, honey and beeswax, and in the bees themselves.

Several studies were performed across Europe as well as North America (one study). An extensive inventory of imidacloprid in bee-collected pollen, honey and bees was performed by Chauzat et al. (2006, 2009, 2011), involving five sites across France with sampling of bee hives of five beekeepers in each area for 3 years and with four sampling events per year. Imidacloprid was found in 40.5 and 21.8% of the pollen and honey samples, respectively. The metabolite 6-chloronicotinic acid was present in 33.0 and 17.6% of the respective samples. The sampling took place in four agricultural areas and one natural area. Using a χ^2 test, frequency of imidacloprid + metabolite detection in pollen was shown to be significantly higher in 2003 compared to 2005; there was no difference for honey samples (Chauzat et al. 2011). No significance difference was found in the frequency of pesticide residue detection in pollen and honey between the different sampling areas (Chauzat et al. 2006, 2009). It is not known at what scale imidacloprid was applied in the agricultural areas where sampling took place. Neither is known what were the main plant species represented by the pollen samples collected.

In a study in northern America, thiacloprid and acetamiprid were present in 5.4% of the pollen samples, while thiacloprid was also measured in 1.9% of the beeswax samples (Mullin et al. 2010). Also in Germany, thiacloprid was the most abundant neonicotinoid as it was detected in 33% of the pollen samples at concentration levels up to 199 μg kg^{-1} (Genersch et al. 2010)

The best measure of exposure and bioavailability are concentrations in honey bees. The study of Chauzat et al. (2011) found imidacloprid in 11.2% of the honey bee samples, while the main metabolite 6-chloronicotinic acid was detected in 18.7% of the samples. Average concentrations were 1.2 (>0.3–11.1) and 1.0 (>0.3–1.7) μg kg^{-1}, respectively.For honey bees, other studies did not detect imidacloprid in the bees. Only in the study of Bacandritsos et al. (2010) higher imidacloprid concentrations were measured in honey bees. This study however, concerned only five samplesThe low residue levels in honey bees probably are best explained from the fast imidacloprid metabolism by the honey bee *A. mellifera*.

1.6 Effects of neonicotinoids on Honeybees

1.6.1 Sublethal effects on reproduction

Reproduction is an important process to assure the further existence of the colony. Indeed, a loss of reproduction (brood) might be more detrimental for the colony than the loss of older bees (foragers) (Decourtye and Devillers2010). A few studies have demonstrated the adverse effects on larval development following exposure to imidacloprid (Tasei et al. 2000, 2001; Decourtye et al. 2005; Abbott et al.2008; Gregorc and Ellis 2011). Decourtye et al. (2005) reported a delay in the time needed for honey bee larvae to hatch or develop as an adult when fed with food contaminated with imidacloprid at 5 μg kg^{-1}. Also for bumble bees (*B. terrestris*) a reduction of the brood (larvae) was seen in micro-colonies orally exposed to contaminated sugar water. (10 μg kg^{-1} imidacloprid) + pollen (6 μg kg^{-1} imidacloprid) (Tasei et al. 2000)

1.6.2 Sublethal effects on behaviour

Sublethal effects which interfere with the process of food collection and subsequent social colony life and pollination need to be considered (Thompson and Maus 2007; Desneux et al. 2007; Mommaerts and Smagghe2011.

Neonicotinoid insecticides act as neurotoxic agents and affect the mobility of bees by inducing symptoms such as knockdown, trembling, uncoordinated movements, hyperactivity and tremors (Lambin et al. 2001; Nauen et al.2001; Suchail et al. 2001; Medrzycki et al. 2003; Colin et al. 2004). These symptoms are easy to observe at high exposure levels, while the effect of a lower dose might be more difficult to see.

Another sublethal endpoint affected by neonicotinoids (acetamiprid and thiamethoxam) is the proboscis extension reflex (PER) following perception of sucrose and water (El Hassani et al. 2008; Aliouane et al. 2009). The effect was demonstrated to be dependent on the route, duration and dose of exposure (El Hassani et al. 2008; Aliouane et al. 2009). In addition, by conditioning of the PER using an odor, various studies demonstrated changes in the olfaction learning of bees upon exposure to neonicotinoids. Learning was reduced after chronic (up to 11 days) exposure to imidacloprid (winter bees: 48 $\mu g\, kg^{-1}$; oral), the metabolite 5-hydroxy-imidacloprid (winter bees: 120 $\mu g\, kg^{-1}$; oral) and thiamethoxam (0.1 ng bee^{-1}; contact) (Decourtye et al. 2003; El Hassani et al. 2008; Aliouane et al. 2009). By expanding the PER test also more information was gained on how neonicotinoids interfere with the memory process. Oral uptake of 0.1 $\mu g\, bee^{-1}$ acetamiprid induced long-term memory impairments, whereas chronic contact to 1 ng bee^{-1} thiamethoxam (corresponding with 1/5 of the LD_{50}) did not cause long-term effects as recovery of memory was seen after 48 h (El Hassani et al. 2008; Aliouane et al. 2009).

Neurotoxic compounds such as neonicotinoids were also reported to interfere with the orientation process of honey bees. Associative learning between a visual mark and a reward (sugar solution) in a complex maze showed that only 38% of the bees found the food source after oral ingestion of thiamethoxam at 3 ng bee^{-1} compared to 61% in the control group (Decourtye and Devillers 2010). In another study using marked foragers that were first trained to forage on artificial feeders, Bortolotti et al. (2003) noticed that a 500 m distance between the hive and the feeding area resulted in no foragers at the hive/feeding area up to 24 h after treatment when foragers were fed with imidacloprid at 500 and 1,000 $\mu g\, l^{-1}$ (Table 4). The latter authors also found that a lower concentration (100 $\mu g\, l^{-1}$ imidacloprid) caused a delay in the returning time (to hive or feeding area) of the foragers. This was confirmed by Ramirez-Romero et al. (2005)

and Yang et al. (2008). Based on these results it is obvious that neonicotinoids interfere with the foraging capacity of bees.

1.7 Conclusion

Neonicotinoids do have a negative effect on honey bees and other insect pollinators including important species of native bees such as bumble bees, mason bees, and others. However, it is unclear whether neonicotinoids have a significant lethal or sub-lethal effect on bees at realistic field levels. The best means of minimizing adverse effects may be by increasing people's awareness of the potential issues through educational forums and via improvements in the instructions on the pesticide label.

Ongoing research is increasing our understanding of the impact of these types of pesticides on bees. For now, the best recommendation is to carefully follow the product label, be judicious in your application, and avoid applying any insecticide product when bees are actively foraging or near the target area.

1.8 Refrences

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