

Monitoring the effect of imidacloprid under semi-field conditions using electronic bee counters

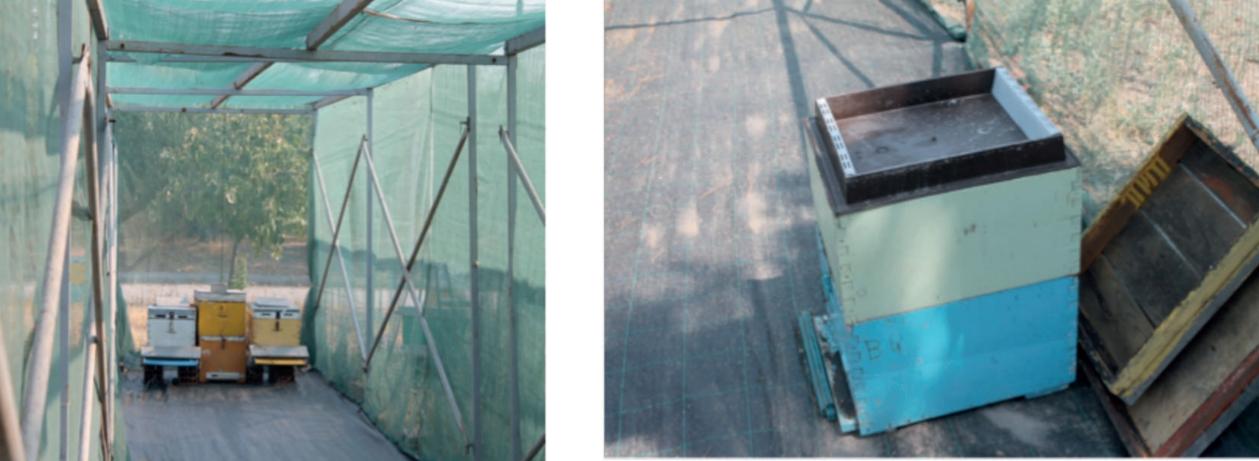
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Abstract

Honeybees exposed to very small doses of neurotoxic substances such as imidacloprid, don't die but they present dysfunction or disorder of their nervous system, their glandular system as well as their cardiac and respiratory rhythms. Laboratory assays provide valuable basic knowledge on the effect of pesticides on honeybees. However, applied research on the field is necessary since this is a reliable way to assess and confirm the laboratory findings. The objective of this work was to determine the impact of imidacloprid on flight activity of honeybees, brood temperature, and *Nosema* prevalence in semi-field conditions.

Two flight tunnels of 12 x 2 x 2 m were used and two colonies were introduced in each tunnel. Imidacloprid was administered in syrup and pollen in dosages of 2 and 3 ppb respectively. Pollen pastries were applied inside the colonies, while syrup was placed in feeders at the opposite end of the tunnels. Electronic bee counters were used at the entrance of the colonies in order to monitor



the daily activity of the honeybees. The results have shown that treated colonies were keeping significantly lower brood temperature compared to controls, they had higher loads of *Nosema* spores at the end of the experiments and they presented higher loses of foragers after about 10 days of imidacloprid administration.

Introduction

Honeybees exposed to very small doses of neurotoxic substances such as imidacloprid, don't die but they present dysfunction or disorder of their nervous system, their glandular system as well as their cardiac and respiratory rhythms (Hatjina et al. submitted). Other studies have demonstrated that low doses of imidacloprid have negative sub-lethal effects on learning and orientation (Decourtye et al. 2004a; b), on foraging activity (Yang et al. 2008) and on homing behavior (Henry et al. 2012), as well as neurophysiological effects (Lambin et al. 2001). Laboratory assays provide valuable basic knowledge on the effect of pesticides on honeybees. However, applied research on the field is necessary since this is a reliable way to assess and confirm the laboratory findings. The objective of this work was to determine the impact of imidacloprid on flight activity of honeybees, brood temperature, and Nosema prevalence in semi-field conditions.

Materials and methods

Two flight tunnels of 12 x 2 x 2 m were used and two colonies were introduced in each tunnel (Fig. 1). Imidacloprid was administered in syrup and pollen in dosages of 2 and 3 ppb respectively. Pollen pastries were applied inside the colonies, while syrup was placed in feeders at the top of empty hives at the opposite end of the tunnels (Fig.2, 3). Electronic bee counters were used at the entrance of the colonies in order to monitor the daily activity of the honeybees (Fig. 4). The bee counters are micro-processor-controlled high precision devices, which count number of bees through 32 bi-directional in/out channels. This way, they continuously monitor the number of outgoing and incoming foragers.

Initially both tunnels were acting as controls and the colonies were fed with control syrup and pollen for 7 days. The next 7 days, the colonies in one tunnel were given syrup and pollen contaminated with imidacloprid for additional 8 days. Finally, for the last 7 days of the experimentation the colonies were continued to be fed with contaminated syrup and pollen but the tunnels were opened from one side, in order the bees to be also able to flight out if they wanted to (see Fig. 3).

Samples of bees for Nosema spores' counting were collected at the beginning and the end of experiment from the outer frames of the colonies. Brood thermometers were also placed in one colony per tunnel and the temperature range (Max-Min) was recorded every second day.

Results-Discussion

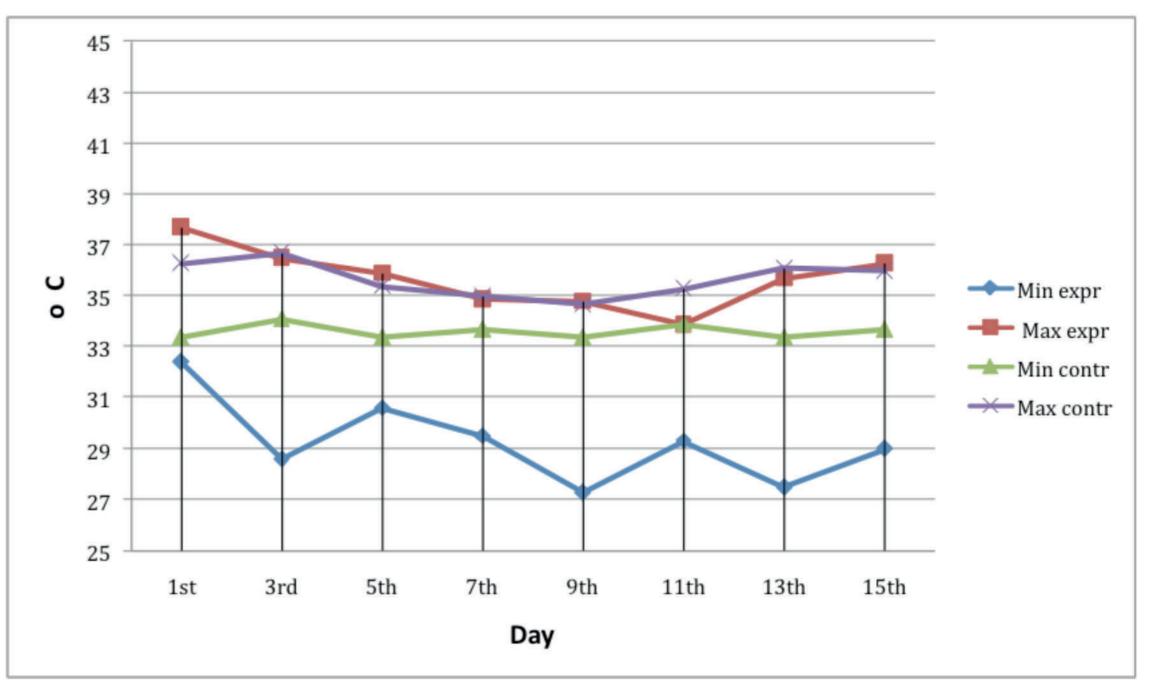
Fig. 1. The tunnels with the colonies

the feeder was 11 m.

Fig. 2. Feeding sugar syrup on top of an empty colony

Fig. 3. The tunnels (opened) with the colonies in one end and the

Fig. 4. The bee counter attached on the entrance of the colony. The solar panel on top of the colony was charging the battery used for the micro-processor.





feeder colony at the other. The distance between the colonies and

The results have shown that treated colonies were keeping lower Min brood temperature by almost 2° C compared to controls, as it is demonstrated in Fig. 5, while not significant difference was found in the Max temperature. This effect could be detrimental to the colonies, as it is known that when brood is reared in 2° C lower temperatures that usual, the result could be higher adult mortality (Medrzycki et al. 2010) or reduced foraging performance (Tautz, et al. 2003).

The difference between outgoing and incoming foragers was noted as 'lost foragers'. Normally, all colonies showed a low number of lost foragers (Fig. 6, first days of experimentation). However, treated colonies presented higher lose of foragers after about 10 days of imidacloprid administration (Fig. 6). Lost foragers could be bees dying naturally, or bees losing their way to the colony and die on the sides of the tunnels. Nevertheless, bee losses were mostly apparent while the tunnels were opened, suggesting that foragers from treated colonies did not find their way back to the colony when they were out of the tunnel, eventually losing their orientation ability.

At the beginning of the experimentation foraging bees from all colonies had a small number of Nosema spores. However, treated colonies developed much higher loads of *Nosema* than control colonies during the 22 days of the experimentation (Fig. 7) confirming that a strong interaction occurs between the microsporidium and this pesticide (Alaux et al. 2009). Possibly, imidaclcoprid affects the immune system of the colonies and increases their sensitivity to parasites.

Conclusions

From the above semi-field study it is aparrent that imidacloprid in sublethal doses has a significant detrimental effect in different aspects of bees' behaviour and health.

- It reduces the average brood temperatute
- It affects foraging performance of the adults, by reducing their orientaion ability
- It reduces their immunity, making them more suceptible to diseases

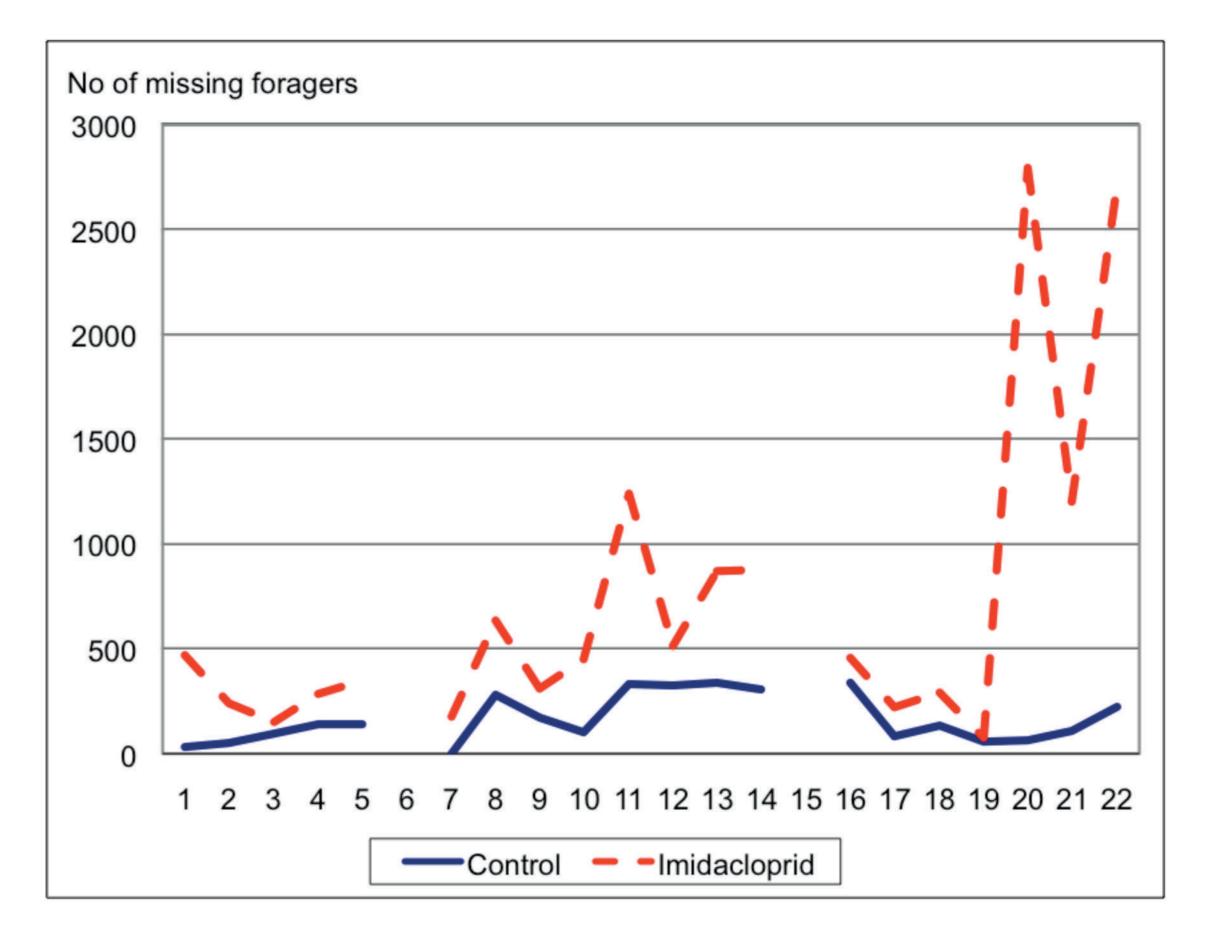
Furthermore, semi-field resuslts confirmed the laboratory results of other researchers.

Awknowlwdgements

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Fig. 6. Number of missing foragers in control and treated colonies colonies as measured by the bee counters (=incoming foragers-outgoing foragers)

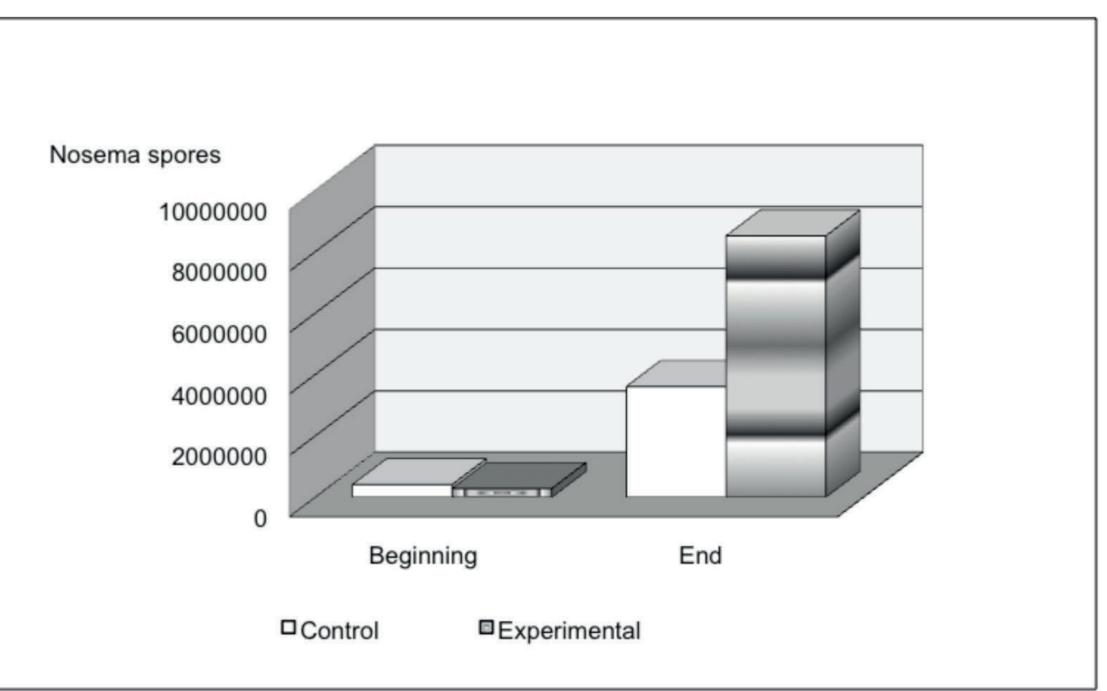


Fig. 7. Absolute counts of Nosema spores in control and experimental (treated) colonies at the beginning and end of experimentation