Dust drift during sowing of maize - effects on honey bees

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considerable route of exposure. Consequently, improvements have been proposed for seed dressing quality regarding dust abrasion taking into account residue content of dust and for the sowing techniques as possible risk mitigation measures. To assess potential effects on honey bee colonies following insecticidal dust drift on adjacent non-target areas, in 2010 and 2011 two large-scale drift experiments were carried out during maize sowing using seed batches from two different years (2010: seed batch from 2008; 2011: seed batch from 2011).

Results: Despite improvements of seed dressing quality regarding dust abrasion comparing the two seed batches (Heubach value 0.86 in 2010 and 0.45 in 2011) and the use of a precision air seeder with drift reducing deflector, in both experiments bee mortality was clearly increased, especially in semi-field conditions.

Conclusions: Drift of insecticidal dusts during sowing of maize may result in a risk for honey bees in field conditions. To exclude adverse effects on bees, especially during sowing of maize further improvements of seed treatment quality and machinery is needed.

Keywords: maize, seed treatment, clothianidin, dust drift, honey bee poisoning

Introduction

Systemic insecticides like neonicotinoids are commonly used as seed treatments in important crops like maize (*Zea mays* L.), sunflower (*Helianthus annuus* L.) and winter oil seed rape (= WOSR; *Brassica napus* L.)⁶. Neonicotinoids are used in maize for control of wireworms (*Agrotes* spp.), cutworms (*Agrotis* spp.), Western Corn Rootworm (*Diabrotica virgifera* Le Conte) and of aphids and leafhoppers¹.

In 2008, in parts of southern Germany treatment of maize seeds with a high application rate of clothianidin (125 g a.s./ha⁷) was used to control the larval stages of the emerging quarantine pest Western Corn Rootworm. During sowing of maize, more than 11.500 honey bee colonies from about 700 beekeepers in the Upper Rhine valley, Baden-Wuerttemberg and approximately 460 colonies from 36 beekeepers near Passau, Bavaria showed symptoms of insecticide poisoning⁷.

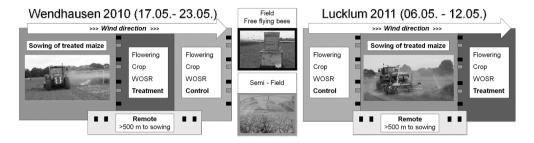
The temporal and spatial link between affected bees and sowing of maize was soon confirmed by residue analyses of samples of dead bees and of flowering bee forage plants from adjacent areas⁷.

Poor seed treatment quality and the use of precision air seeders led to drift of dust containing insecticides which contaminated flowering winter oil seed rape, fruit trees and weeds (e.g. *Taraxacum* sp.). Contaminated flowers are considered to be the most important route of exposure of bees to toxicants².

Since 2010, two large-scale drift experiments during sowing of clothianidin-treated maize were carried out to investigate the link between abrasion potential of treated maize seed (determined by Heubach-values⁴), dust drift and resulting residues in adjacent flowering crops as well as the effects on honey bees exposed to these contaminated flowering crops.

Experimental methods

Both drift experiments, 2010 and 2011, were performed on wind-exposed areas near Braunschweig (Wendhausen and Lucklum). Due to the strong dependence on local wind conditions and a limited availability of test fields, two different field set-ups were used (Fig. 1).





In the preceding autumn 2009 and 2010 at both plots, winter oilseed rape (*Brassica napus* L.) was sown for the drift trials. In spring, this WOSR served as a flowering bee attractive crop in the drift experiments during sowing of maize. The test site Wendhausen in 2010 was located in an area without notable alternative bee attractive crops, while the test site nearby Lucklum in 2011 was in a region with a high portion of flowering WOSR.

In both drift experiments a total of 18 colonies were used (Hohenheimer Einfachbeute, Zander, 10 frames). Each contained at least six combs with brood in all stages (eggs, larvae and sealed brood) including sufficient honey and pollen storage (Tab. 1).

| Variants | Wendhausen 2010 | Lucklum 2011 |
|------------------------------------|-----------------|--------------|
| Field / Remote (n = 4) | ~17.000 | ~8.500 |
| Field / Control (n = 4) | ~21.500 | ~11.500 |
| Field / Treatment ($n = 4$) | ~16.000 | ~11.000 |
| Semi-Field / Control (n = 3) | ~10.00 | ~7.500 |
| Semi-Field / Treatment ($n = 3$) | ~11.500 | ~9.500 |

 Tab. 1
 Average numbers of bees in colonies in the two drift experiments before the start

Two days before set-up of hives at the trial site (five days before start of the experiments), an assessment of the colony and brood nest size was carried out using the Liebefelder evaluation method⁵ to determine the development status of the experimental colonies. Further assessments of colony and brood development were made between the 14 -17th day and 27-30th day after the initial evaluation.

Since highest risk for foragers is expected in adjacent flowering crops next to the sown acreage, in both 'worst case' scenarios bee colonies were placed in semi-field and field and set-up along the edges of the sown area.

In the field four colonies each were placed at distances of 0 m (='Treatment'), 50 m (= 'Control') and more than 500 m (='Remote') to the windward directed border of the WOSR field (Fig. 1) In the semi-field approach in both variants 'Treatment' and 'Control' three gauze-covered tunnels (16 x 6 m) were set up at the field edge on the flowering WOSR crop and each provided with a bee colony (Fig. 1, 'Semi-Field'). Due to the limited food supply in the tents, smaller bee colonies were used in semi-field, while in field larger colonies were used (Tab. 1).

In order to assess bee mortality, each hive was fitted with a modified dead bee trap³ (Type 'Gary', W x L x H: 435 x 400 x 300 mm; mesh size: 8 x 8 mm; Fig. 1, 'Field').

Before bee flight activity, the hive entrances of semi-field colonies were closed and the gauze removed from the tents. After sowing, the tents were covered again and the hives reopened. The hive entrances of the bee hives in the field experiment were not closed during sowing. In both experiments treated maize of different quality was sown using a precision air seeder with drift reducing deflector (Tab. 2).

| Experiment | Wendhausen 2010 | Lucklum 2011 | | | | |
|---|---|--|--|--|--|--|
| Arable crop | Maize (<i>Zea mays</i> L.) | Maize (Zea mays L.) | | | | |
| Seed treatment with application rate | Poncho Pro (clothianidin: 125 g/ha) | Poncho 600 FS (clothianidin: 50 g/ha) | | | | |
| Content of active substance clothianidin in Heubach filter dust [%] | 10.6 | 19.1 | | | | |
| Average Heubach-Value [g/100 000 kernel] | 0.86 | 0.45 | | | | |
| Sowing technology | pneumatic precision air seeder, type Kverneland Accord Optima NT e- drive, converted and drift reduced | | | | | |

 Tab. 2
 Seed treatment of maize, abraded dust and sowing technology

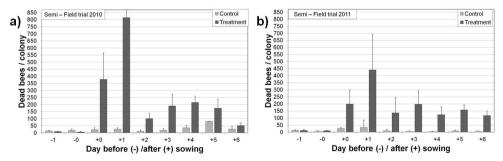
As maize sowing was conducted during bee flight activity, in the field experiments foragers leaving and entering the hives were continuously exposed to contaminated dust, both directly to dust in the air and indirectly through contaminated pollen and nectar of flowers of adjacent downwind WOSR as well as to uncontaminated flowers in the upwind area. As the colonies in the semi-field experiments were not actively foraging during sowing, the foragers were only exposed to contaminated nectar and pollen and dusts deposited on flowers after reopening the entrances of the hives but not directly exposed to the dust drift.

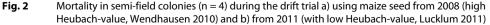
In both drift experiments sampling and assessment of dead bees (workers) from bee traps (Type Gary) was conducted once a day starting the day before sowing (= DBS) until the 6th - 7th day after sowing 5 times a day on the day of sowing date and 3 times the first day after sowing. Samples of dead bees were separately packed and stored at -20 °C until transport to the analytical laboratory of the JKI in Berlin-Dahlem.

Results and discussion

Semi-field trials

The semi-field variant is a 'worst-case' scenario, which allows bees to forage only on limited crop inside the tent.





The semi-field variants 'Treatment' of both experiments showed similar trends of mortality within the experimental period compared to the field approaches, but at a much higher level (Fig. 2 and Fig. 3). After a peak in mortality was detected in the dead bee traps at the assessments in the morning of the first day after sowing (2010: 815 (Fig. 2a); 2011: 440; (Fig. 2b)), the mortality in 'Treatment' colonies was lower in the following days but still showed a treatment related, clearly increased mortality until the end of the experiment.

The mortality in the 'Control' variant was on a low level throughout the study. In 2010, a slightly increased mortality on DAA+5 was likely caused by climatic conditions. In 2011 (Fig. 3b.), on the day of sowing and on the first day after sowing the mortality was slightly increased; this can probably be attributed to slightly increased stress for colonies due to enclosure until sowing process was finished in the late forenoon. Due to the availability of uncontaminated forage in control and worst case exposure to only contaminated forage in the semi-field experiment, the differences in mortality between control and treatment are considerably pronounced. A higher increase of mortality was observed in 2010 compared to 2011.

But also the semi-field study does not allow any differentiation in the proportion of bees poisoned by contact toxicity due to gathering of contaminated pollen and bees poisoned by the consumption of contaminated nectar and pollen. However, it may be concluded that the majority of dead bees in the hives and dead bee traps have been not caused by direct exposure to contaminated dust during forage flights, but occurred after foraging of nectar and pollen on the treated crop and after consumption of contaminated food by hive bees

Field trials

The mortality of the colonies before sowing was on a comparable level for all colonies in 2010. In 2011, on the day of sowing before start of sowing a comparably high mortality was observed with a mean of 125.5 bees/colony, caused by a highly increased mortality of single colony. Although a multi-residue analysis was conducted with the sample, it was not possible to conclude on the reason for the increased mortality. After sowing, the mortality of this colony returned to a mortality level comparable to other treatment colonies. Especially during the first 24 hours after sowing, the 'Treatment' of both experiments revealed considerably higher mortality compared to the control. Thereafter, the mortality was lower but still showed a treatment related increase for several days after sowing (Fig. 3b).

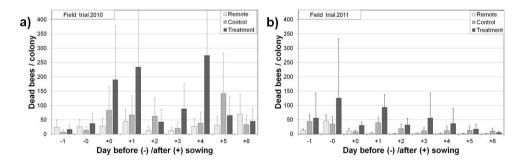


Fig. 3 Mortality in field colonies (3 variants: Treatment directly bordering to exposed flowering WOSR, Control in 50 m distance, Remote < 500 m distance; each n = 4) during the drift trial a) using maize seeds from 2008 (high Heubach-value, Wendhausen 2010) and b) from 2011 (low Heubach-value, Lucklum 2011)

Due to the short distance of the 'Control' (ca 50 m) to the contaminated WOSR area, also a peak in mortality was observed after 24 hours, but mortality was lower than in the 'Treatment'. As the free

flying colonies of control and treatment could forage both on the contaminated but also on the uncontaminated side of the field, it is not possible to conclude if the higher mortality was caused due to exposure of bees to toxic amounts taken up during flying or by a increased portion of bees foraging on the contaminated side of the field. Only the mortality of the 'Remote' colonies with little chance to forage on contaminated WOSR always remained at a low level which was considered as the range of natural mortality.

In general, across all field variants in 2010 (Fig. 3a) much higher increase of mortality was observed compared to the experiment in 2011 (Fig. 3b), in line with the results from the semi-field studies Fig. 2a and 2b). To some extent, this may be attributed to the improvement of seed quality of maize but may also to be due to slightly different sizes of hives used in the experiments (in average: 2010 > 2011; Tab. 1). However, the differences in the field experiments are also caused by differences in the vegetation surrounding the trial sites and the available alternative forage.

While the drift experiment 2010 at Wendhausen was carried out in an area with low alternative nectar availability, the experimental site of the drift experiment 2011 at Lucklum was located in an agricultural region with a high portion of WOSR and other alternative forage (e.g. other WOSR crops, maple trees and dandelion). Thus, in 2011 the foragers of the field colonies may have been partially distracted from the contaminated WOSR crop, which is in line with local observations of lower flying activity in the WOSR crop during the test period. Furthermore, it was confirmed by analyses of randomly selected pollen loads from samples of both experiments that bees have also been collecting pollen e.g. from maple trees.

In field trials, a differentiation of the proportion of bees which died from direct or indirect contact exposure during flying, collecting toxic dust particles and contaminated pollen or the consumption of contaminated nectar and pollen was not possible.

Conclusion

The results of both experiments showed a clear treatment related increase of bee mortality, especially in the worst-case semi-field trials, but also in the field trials. The bee mortality of both variants in 2011 was slightly lower than in 2010, but still on a high level. Presumably the exposure of bees to residues on the crop may have been on a similar level, taking into account the Heubach-values and the residues in the dusts. The experiment 2010 was located in a region with few alternative bee attractive crops, whereas the experiment 2011 was in an area with a wide range of flowering. Thus, in the field experiments, one reason for smaller effects observed at the bee colonies in 2011 was sufficient alternative forage in the surrounding of the fields, distracting foragers from the contaminated field site. In both experiments there was a clear impact of abraded dust from clothianidin-treated maize seeds on the mortality of bees. This indicates a necessity for further improvements of the seed treatment quality of maize and/or of the sowing technique to exclude adverse effects on bees.

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