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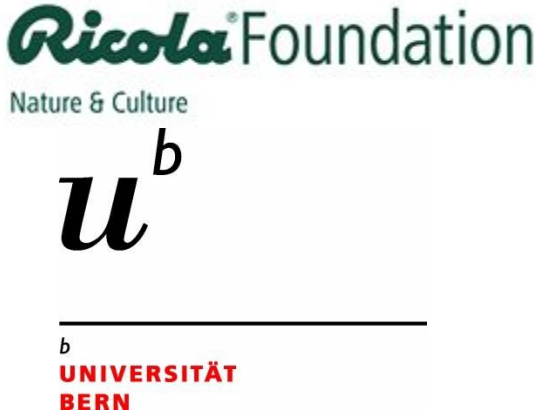

APITOX WORKSHOP

Proceedings

Bologna, Italy, 27/03/2017 – 28/03/2017

APITOX WORKSHOP

Topic

- Field trials
- HPG development
- On-going ring tests
- Chronic toxicity of adult bees

When

- 27 March - from 9:00
- 28 March - until 14:00

Where

CREA-API - Council for Agricultural Research and Economics
Honey Bee and Silkworm Research Unit
Via di Saliceto 80
40128 Bologna
Italy

SCHEDULE

27 03 2017

09:00-09:30	**Medrzycki** - Welcome words + tour de table (with update of EU ring tests (bumblebees, osmia, homing flight))
09:30-10:00	**Giffard** - Current approaches of the FR methodological group about repro-toxicity tests **Fourrier** - Homing flight ring testing **Jeker** - video homing flight ring testing
10:00-11:30	**Hatjina** - Final results of the common assessment of the HPGs after imidacloprid uses. Field vs Lab experiments
11:30-11:50	**Coffee/snack break**
11:50-12:20	**Brandt -** Effects of clothianidin on HPG and brood development in small colonies
12:20 - 12:50	**Brandt -** Imidacloprid degradation and resurrecting bees
13:00–14:00	**Lunch**
14:00-15:30	**Simon Delso -** Field trials on trial: state of play and paper discussion
15:30-16:00	**Coffee/snack break**
16:00-18:30	**Tosi/Simon-Delso** - Chronic toxicity trial. Presentation of results and discussion
20:00-	**Social dinner**

28 03 2017

09:00-10:45	Discussion about 2017 activities - Publications
10:45-11:05	**Coffee/snack break**
11:05-12:35	Discussion about 2017 activities - Testing
12:35-13:00	AOB
13:00-14:00	**Lunch**

ORGANIZER CONTACTS	
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ABSTRACTS

Effects of chronic long-term exposure to pesticides on honey bees: a ring test

Tosi S[1], Brandt A[2], Colli M[3], Fourrier J[4], Giffard H[5], Hernández-López J[6], Williams G[7], Simon-Delso N[8]

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New pesticides enter the market each year, and their effects on non-target species are assessed through standard guidelines that test limited side-effects after limited exposure scenarios. Although official guidelines require testing acute or short-term chronic (10 days) pesticide exposures only, honey bees can be chronically exposed to pesticides for longer periods. We tested the effects of chronic long-term exposure to a pesticide of low acute toxicity and dimethoate (reference item) on honey bee survival, behaviour and consumption. The experiment was separately performed in 7 laboratories, located in EU and USA, in 2016. The experiment was concluded when the LT50 of the control treatment was reached (25 ± 5, mean ± SD).

We present the methodology of the common experiment, together with the preliminary results. We will discuss about variability, reproducibility and interpretation of the results, and discuss about the next steps.

The meaning of life and death: quick-freezing stops detoxification and improves chances of imidacloprid detection in honeybees

Matthias Schott[1,2], Gabriela Bischoff[3], Gerrit Eichner[4], Andreas Vilcinskas[2,5], Ralph Büchler[6], Marina Doris Meixner[6], Annely Brandt[5,6*]

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In cases of acute intoxication, honeybees lay in front of their hives for several days, exposed to sunlight and weather, before a beekeeper can take a sample. Beekeepers send samples to analytical laboratories, but sometimes no residues can be detected. Temperature and sun light could influence the degradation rate of pesticides and thereby the level of residues left for analysis. Additionally, samples are usually sent via normal postal services without cooling. Here we addressed the question whether freezing of honeybee samples helps to prevent the degradation of the neonicotinoid imidacloprid.

Imidacloprid was rapidly degraded, even in immobile, “dead” looking honeybees that surprisingly recovered from paralysis after 48 hours. Detoxification was stopped by quick freezing of samples. The mode of transport did not affect the imidacloprid residue levels. However, UV light significantly reduced imidacloprid residues in honeybees. Additionally, we found that group feeding resulted in increased variance of residue levels, which is relevant for acute oral toxicity tests according to the OECD guideline for testing chemicals.

We conclude that elapsed time after pesticide poisoning is key for detection of neonicotinoids. Freezing samples before mailing significantly reduces the degradation of imidacloprid and may increases the accuracy of laboratory analysis for pesticides with similar degradation behavior.

Quantifying resilience:

Negative effects of neonicotinoid pesticides on individual nurse bees and larvae can be compensated by increased brood rearing of the colony

Matthias Schott[1], Maximilian Sandmann[2], Matthias Becher[3], Rayko Halitschke[4], Gerrit Eichner[5], Stephanie Krueger[6], Rolf-Alexander Düring[7], Gertrud Morlock[5], Andreas Vilcinskas[1], Marina Doris Meixner[2], Ralph Büchler[2], Annely Brandt[2*]

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Honey bees are highly social insects, with labor division and intensive brood care. Nurse bees produce royal jelly in the hypopharyngeal gland (HPG) to feed larvae, young workers, and the queen. In laboratory experiments exposure of nurse bees to the neonicotinoid imidacloprid leads to a reduction of the HPG size. We wanted to study the effect of the neonicotinoid clothianidin on HPG size, royal jelly composition, and brood development in colonies.

Over a period of 7 weeks, twenty small colonies were chronically exposed to different concentrations of clothianidin (0 µg/L, 1 µg/L, 10 µg/L, or 100 µg/L) in sugar syrup. Every treatment was replicated five times. The clothianidin content was quantified in samples of food and worker bees. Each week all eggs, larvae, or pupae were counted for all colonies, and samples of royal jelly, larvae, and adult worker bees were collected.

The HPG size was reduced in age-defined worker bees after 12 days of exposure to clothianidin, even in the lowest concentration. In addition, the composition of the royal jelly was altered in colonies treated with 10 µg/L or 100 µg/L clothianidin. After three weeks of treatment, the number of larvae and pupae were significantly reduced in colonies exposed to 100 µg/L clothianidin. The brood survival was reduced to 40% or lower in clothianidin exposed colonies in the second half of the experiment.

Exposure to clothianidin affected HPG size, royal jelly composition, and brood development in small honey bee colonies.

Final results of the common assessment of the HPGs after imidacloprid uses. Field vs Lab experiments: differences among the testers

Hatjina Fani[1], Balzola Luigi[2], Brandt Annely[3], Bruckner Selina[4], Colli Monica[5], Hernández López Javier[6], Krainer Sophie[7], Laurino Daniela[2], Maistros Anastasios-Damianos[1], Manino Aulo[2], Porporato Marco[2], Renzi Maria Teresa[8], Sgolastra Fabio[8], Simon-Delso Noa[9], Tosi Simone[10], Williams Geoffrey[4]

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The objective of this work was firstly to assess differences in results between laboratory and field studies and secondly to evaluate the operator variability when assessing the effects of stressors on honey bees HPGs development. For the evaluation of HPGs, bee samples from 2 different experiments were used; this way the same operators had the possibility to test more than 2 different conditions (and presumably effects) of the same stressor on the bees' HPGs. In the present study, we studied the impact of sublethal doses of a neurotoxic stressor on the development of hypopharyngeal glands of nurse bees from the same colonies (n=2) both in individual bees kept in small cages under laboratory conditions and in individual bees kept in the original colonies. Bee colonies were fed with 2 ng/kg imidacloprid administered in sugar solution and with 3 ng/kg imidacloprid administered in pollen pasty during one month. Statistical analysis of the results show a different development of the glands in the laboratory and in the field conditions, as well as different impact of the stressor:
the glands have a general high diameter in the field conditions than in the laboratory conditions; bees 15 days old had decreased diameter of acini compared to the 10 days old bees when reared in the laboratory but not in the field conditions; the stressor (imidacloprid) had a significant decreasing effect on the acini in the laboratory as well as in the field conditions on bees at age of 10 days but not at bees of age 15. Some differences were detected among the testers, but the results and the significance of the effects of the stressor were in depended form the tester. The reliability of the tester was always higher of 96%. In conclusion we can state that digital measurement of the acini of the HPGs for detecting changes in the development of the glands is a highly reliable method.

Field trials on trial - Evaluation of the information of honey bee field test for pesticide risk assessment

Simon-Delso Noa[1], Aupinel Pierick[2], Brandt Annly[3], Colli Monica[4], Fourrier Julie[5], Giffard Hervé[6], Gregorc Ales[7], Hatjina Fani[8], Hernández-López Javier[9], Jeker Lukas[10], Laurino Daniela[11], Malagnini Valeria[12], Manino Aulo[11], Medrzycki Piotr[13], Molitor Claire[6], Porporato Marco[11], Renzi Maria Teresa[14], Sgolastra Fabio[14], Tosi Simone[14,15], Williams Geoff[10,16]

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Field trials (FT) have been traditionally employed as tests used to ultimately determine the risk of a pesticide for honey bees in the risk assessment scheme, the so called highest tier. In 2016, test summaries publicly available in the Draft Assessment Reports of different active ingredients (aa.ii., n=5) were randomly allocated and evaluated by bee ecotoxicologists (n=18), each one receiving 2 summaries. The purpose of the exercise was to verify and score the quality of the information included in these summaries according to the criteria proposed by the EFSA guidance document for the risk assessment of pesticides on bees (2013) related to exposure and toxicology assessment, statistical requirements, design of the FT and duration and frequency of assessments. The results and following steps were discussed in the view of improving the quality of information available for risk assessment purposes and public availability of data.

PARTICIPANTS TO THE WORKSHOP

Annely Brandt, Javier Hernandez Lopez, Ulrike Riessberger-Gallé, Lukas Jeker, Hervé Giffard, Fani Hatjina, Piotr Medrzycki, Noa Simon Delso, Daniela Laurino, Valeria Malagnini, Fabio Sgolastra, Julie Fourrier (not in the picture)

OUTCOME OF THE WORKSHOP

PROJECTS 2016:
PROJECT ON FIELD TRIALS - Send to publication
PROJECT ON HPG - Send to publication
PROJECT ON CHRONIC TOXICITY - Send to publication

PROJECTS 2017:
PROJECT ON HPG:
- Evaluation of parameters that may affect HPGs
- Comparison HPG diameters, protein content, weight of the head

PROJECT ON PESTICIDE DEGRADATION - Ring testing
PROJECT ON CHRONIC TOXICITY - Second year of ring testing

Next APITOX activities:
- COLOSS General Assembly (Istanbul, Turkey) - Presentation of bee ecotoxicology
- APITOX Workshop - Valencia (Spain) 16-17/10 or 21-22/10/2017