

## Comparison between EPPO and CEB field methodologies

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### Introduction

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provided that laboratory and semi-field data (tunnel test according to CEB) show an acceptable risk for honeybees.

The French CEB procedure of tunnel tests was revised in 2003 to include the toxic reference dimethoate and to account for a mandatory mitigation measure which is to spray the product out of the presence of bees, during early morning or late evening, on the entire crop area. These conditions are clearly worst-case, more than the previous ones where sprayings were conducted on half of the crop area in the presence of bees. However, applications out of the presence of bees are now considered. This revised procedure is in line with the recommendations of the EPPO guideline n°170 which was recently revised<sup>6</sup>.

The semi-field test was the preferred option since test conditions are homogeneous regarding soil, crop, climatic exposure and bee colonies settled in similar insect-proof tunnels, whereas field studies imply greater heterogeneity of conditions.

However, field trials are the last step of the testing scheme developed by the EPPO guideline n°170 which is the reference frame for regulatory risk assessment for honeybees in Europe<sup>3, 7</sup>. Since assessment for the bee ban waiver in France could not be separated from a full risk assessment according to the EPPO risk assessment scheme, a procedure for field trials was necessary to complete the CEB methods.

Therefore, the French Bee Working Group was commissioned to write a field methodology suitable for the assessment of impact to honeybees when a bee ban waiver is required.

**Keywords:** field, French CEB, EPPO

### Development

The French working group worked during two years under official supervision on the text of a methodology adapted to field studies with honeybees. This working group is composed of members from academia, industry, beekeeping and consultancy. This group presented the result of this work in March 2011 to the Ministry of Agriculture. This field methodology became an official guideline under CEB recognition, compliant with the EPPO guideline<sup>3, 6</sup>.

Although field studies were not recommended in France for a long time because of poor confidence in their reliability to represent worst-case conditions, they became of interest for post-registration monitoring of plant protection products and to complete the testing procedures to apply for a bee ban waiver in France. By extension they became of high interest to specific studies such as the impact of plant protection products on the honeybee brood in field conditions.

The working group focuses at simple parameters in order to manage field studies with honeybees that could also be used for registration at the European level. The objective of this field methodology is to assess the effect of foliar application of plant protection products on honeybee colonies.

Compared to semi-field studies, characteristic of the open field tests are the representative conditions of agriculture, with sufficient surfaces for the foraging activity of honeybees. In these agricultural conditions honeybee colonies are settled in hives suitable for a professional use. They are strong enough to forage large surfaces, comparable to those used by professional beekeepers. By the strength of the colony and the normal bee activity of forager bees there is no experimental stress in

the swarming behavior. These field studies allow mid-term observations and quantitative samples of different matrices for residue analysis (of worker honeybees, brood, flowers, honey and pollen).

Although EPPO guideline n°170 and French CEB n°230 are being harmonized about field studies, there are still some significant differences on the following points:

- According to beekeepers recommendations the French methodology requires surfaces of at least 2 ha per test unit in order to provide sufficient crop plots to forager bees while presenting realistic conditions of agricultural practices. EPPO recommends plots of at least 2500 m<sup>2</sup> of *Phacelia* or about 1 ha for mustard or rapeseed.
- In both methodologies the number of hives required is adapted to provide enough data. Four hives per test unit are recommended in the EPPO guideline when the French CEB proposes 3 hives per hectare that makes a minimum of 6 hives per test unit. In both cases it is mandatory to use dead bee traps and recommended to use pollen traps (for further analysis). These numbered hives are used to limit the variability of bee keeping conditions.
- The timing of applications is also different. For regulatory reasons applications in France have to be realized during flowering and after the flight bee activity, close to or during the night. However the European guideline recommends maximizing the bee exposure with product application during the foraging activity. In this way it is considered as a worst case for the impact to bees. It is therefore possible to add a test modality during bee flight in the French schedule, but this could require a previous authorization for trial if the study treatment is under development or known to have adverse effects to bees.
- Similarly two test modalities only are supposed to be tested in this French guideline (study treatment and water control) whereas it is possible to add a toxic reference in some cases when using the EPPO guideline. The use of a toxic reference such as dimethoate is strictly forbidden during flowering in field trials in the CEB requirements, not only because of the impact to bees but also and mainly because of the effects to all beneficials in the surrounding.
- Mortality and foraging observations are collected with similar timing within both methodologies and bee keeper's visits cover at least one brood cycle (up to 28 days).

### **Discussion and conclusion**

Over the experimental phase in fields with treated and untreated modalities, collected data are supposed to bring a useful tool in the assessment of impact to honeybees. Despite limits to field trials (field surfaces, homogeneity of tested and control plots, only high effects on adult mortality are detectable, impossible statistical validation without replicates)<sup>4</sup> we think it is reasonable to consider the harmonization of both French CEB and EPPO field trials to honeybees.

For the validation of the field study the French Bee Working Group proposes that mortality levels and foraging activity should be similar among treatments before foliar applications. Then the control mortality should normally not increase more than 50% after application, unless it is demonstrated that the increase is not biologically significant. On the other hand the EPPO bee guideline suggests to repeat the test if mortality is too high in the control or too low in the reference modality. The criteria for deciding whether a mortality rate is too low in a toxic reference or too high in a control are currently under discussion by a working group of ICPBR.

Both guidelines require the use of statistical analysis when appropriate. However sufficient available and reliable data are necessary for a significant analysis.<sup>5</sup>

This French CEB guideline for field studies intends to bring an improvement to the EPPO guideline, considering more realistic conditions for the validation of field trials with honeybees. Therefore the Bee Working Group of the French CEB proposes ICPBR to look at potential improvements in the parameters for conducting field trials, in the revision of the field guidance to the EPPO 170 guideline.

## References

1. Arrêté du 28 novembre 2003 relatif aux conditions d'utilisation des insecticides et acaricides à usage agricole en vue de protéger les abeilles et autres insectes pollinisateurs. JORF, 30 mars 2004.
2. AFPP- Commission des Essais Biologiques: Méthode d'évaluation des effets des préparations phytopharmaceutiques sur l'abeille domestique *Apis mellifera* L. Méthode CEB n°230. Edition Mars 2011.
3. Alix A and Lewis G, 2010: Guidance for the assessment of risks to bees from the use of plant protection products under the framework of Council Directive 91/414 and Regulation 1107/2009, EPPO Bulletin, Volume 40, Issue 2, pages 196–203.
4. Borlotti L, Montanari, Marcelino J, Medrzycki P, Maini S and Porrini C: Effects of sublethal imidacloprid doses on the homing rate and foraging activity of honey bees, *Bulletin of insectology* **56**: 63 – 67 (2003).
5. Dechaume-Monchamrion FX, Decourtye A, Hennequet-Hantier C, Pons O and Pham-Délégue MH, 2003: Statistical analysis of Honey bee survival after chronic exposure to insecticides, *Environmental Toxicology and Chemistry* **22 (12)**: 3088–3094 (2003).
6. OEPP/EPPO, 2010: Guideline for the efficacy evaluation of plant protection products – Side effects on honeybees. PP 1/170 (4), Bulletin OEPP/EPPO Bulletin 40, 313–319.
7. OEPP/EPPO, 2010: Environmental risk assessment scheme for plant protection products – Chapter 10: honeybees. PP 3/10 (3), Bulletin OEPP/EPPO Bulletin 40, 323–331.