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DOTT.SSA GIULIA RAMPAZZO
TUTOR: PROF.SSA TERESA GAZZOTTI

Part 1 - Period abroad in the laboratory of Food Chemistry and Toxicology, Faculty of Pharmacy, University of Valencia.
Multi-residue method based on HPLC Q-TOF-HRMS for the determination of pesticides residues and mycotoxins in honey samples

OBJECTIVE

Considering the shortage of available works on multi-residual methods on mycotoxins and pesticides residues in honey, the aim of the work was: (1) to develop and validate a multi-residue method using QuEChERS approach in combination with high pressure liquid chromatography coupled with quadrupole-time of flight-high resolution mass spectrometry (HPLC–Q-TOF-HRMS), (2) to apply the developed method evaluating the presence of pesticide residues and mycotoxins in real honey samples collected from local beekeepers, (3) to detect possible nontargeted compound and their metabolites in samples.

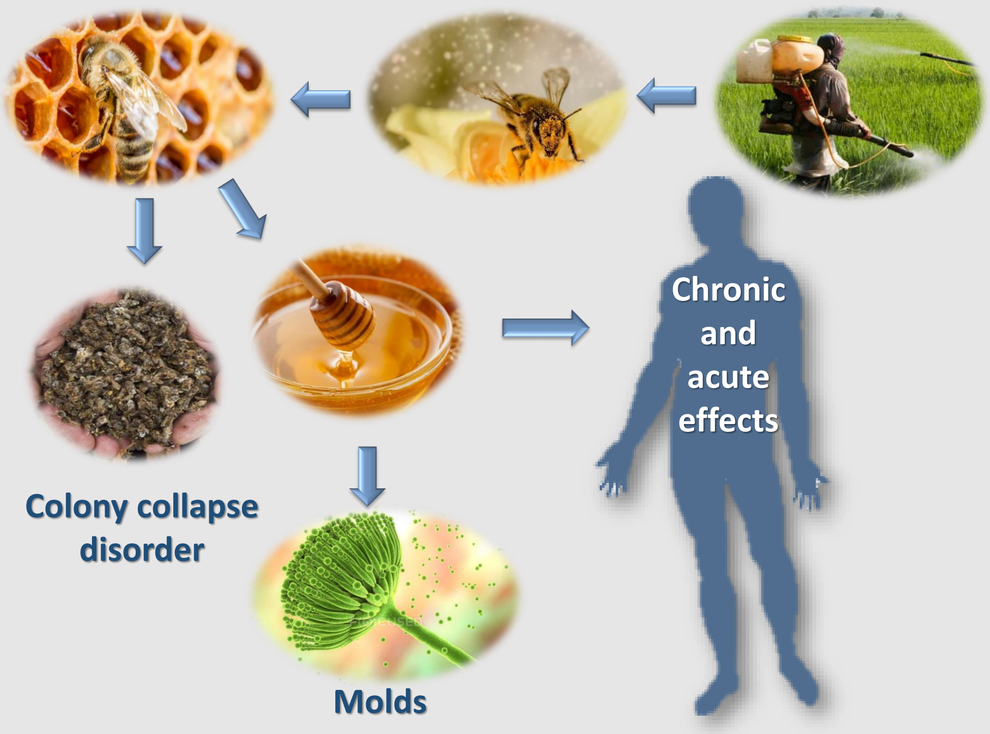

Fig.1 Sources of exposure and risks for humans and bees

MATERIALS AND METHODS

A simple and rapid method for simultaneous determination of pesticide residues (*n=6*) and mycotoxins (*n= 17*) in honey was developed. Sample preparation consisted of a QuEChERS-based methodology and the analysis of extracts were performed by high pressure liquid chromatography coupled with quadrupole-time of flight-high resolution mass spectrometry (HPLC Q-TOF-HRMS).

RESULTS

The extraction method was optimized in order to obtain the best sample clean-up and higher level of analites recovery: two mix of different d-SPE sorbents were tested. The C18 sorbent mix showed a better recovery for most of compounds (70-120%). The method was finally validated for linearity, recovery, precision and sensibility. The validated protocol was successfully applied to 18 honey samples acquired from local Spanish beekepers.

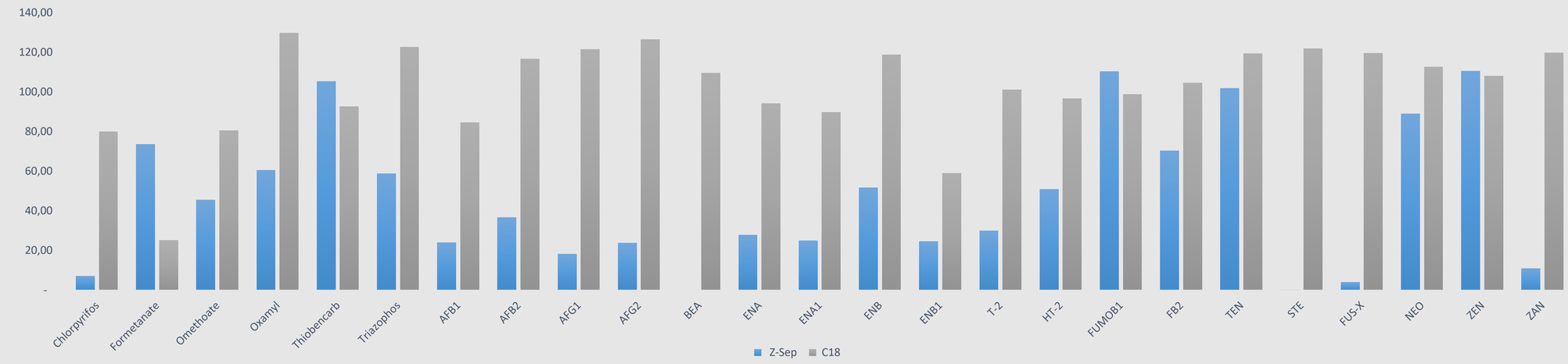

Fig. 2 - Effect of two different mixtures of d-SPE sorbents on honey samples clean-up fortified at level of 100 ng/g for studied analytes.

CONCLUSIONS

Occurrence of mycotoxins was not detected in analyzed honey samples. However, detectable levels of chlorpyrifos and oxamyl were found in 2 out of the 18 analyzed samples but at levels not exceeding the maximum limit fixed by the European Union for pesticide residues in honey.

Part 2 - Department of Veterinary Medical Sciences, University of Bologna, *BSBT, CABA-Lab.*
Development of a LC-MS/MS method for the analysis of polar anionic pesticides in honey

OBJECTIVE

To complement the work carried out in Spain, it was decided to proceed with the development of a further method of analysis focusing on polar anionic pesticides in honey, in particular glyphosate (GLY) and its main metabolite aminomethylphosphonic acid (AMPA).
GLY is a broad-spectrum herbicide, currently with the highest production volumes of all herbicides. GLY is currently approved for use in the EU until 15 December 2022. In September 2021, EFSA and ECHA launched public consultations and several NGOs expressed their concerns about the renewal assessment process.

MATERIALS AND METHODS

The first approach was to test the method (QuPPe-PO-Method) proposed by the EU Reference Laboratories for Residue of Pesticides (EURL-SRM). Since it is stated in the procedure itself that “strong matrix-effects are frequently observed”, tests were conducted for its evaluation.

PRELIMINARY RESULTS

Preliminary results show good recovery and little matrix effect for GLY and its presence has been detected in some honey samples available on the Italian market. In contrast, AMPA showed strong ion suppression of the signal.

FUTURE PROPOSAL

Further tests are in progress to improve sample purification. In addition to this it is intended to extend the method to other polar pesticides (ethephon, glufosinate, fosetyl-Al , phosphonic acid) and their metabolites.

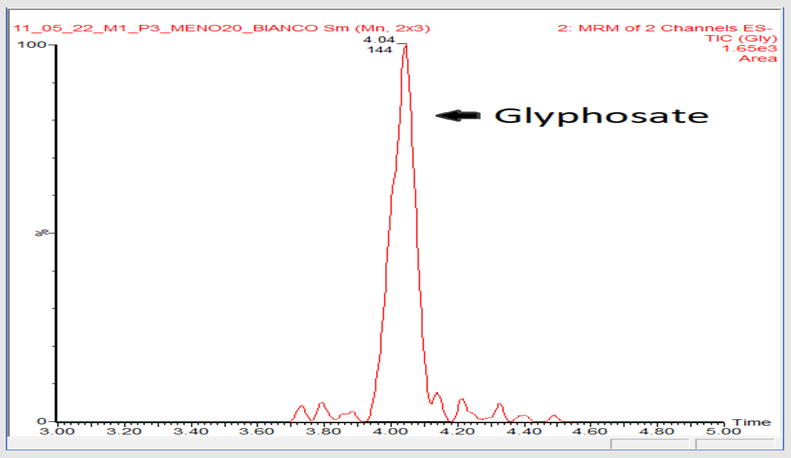

Fig. 1 – Chromatogram on Italian honey sample