



HAL
open science

Focus on the GC-HRMS based analytical development for characterization of human internal exposure to pesticides, in a multiplex inter-laboratory approach

Tarek Moufawad, Ingrid Guiffard, Elodie Mirmont, Fatima-Zahra Alem, Emilien Jamin, Arthur David, Laurent Debrauwer, Jean-Philippe Antignac, Emmanuelle Bichon

► **To cite this version:**

Tarek Moufawad, Ingrid Guiffard, Elodie Mirmont, Fatima-Zahra Alem, Emilien Jamin, et al.. Focus on the GC-HRMS based analytical development for characterization of human internal exposure to pesticides, in a multiplex inter-laboratory approach. Congrès Francophone sur les Sciences Séparatives et les Couplages, Mar 2023, Paris, France. 48, pp.2097 - 2098, 2023. hal-04357639

HAL Id: hal-04357639

<https://hal.science/hal-04357639v1>

Submitted on 21 Dec 2023

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

Focus on the GC-HRMS based analytical development for characterization of human internal exposure to pesticides, in a multiplex inter-laboratory approach

MOUFAWAD Tarek¹, GUIFFARD Ingrid¹, MIRMONT Elodie², ALEM Fatima-Zahra³, JAMIN Emilien L.², DAVID Arthur³, DEBRAUWER Laurent², ANTIGNAC Jean-Philippe¹, BICHON Emmanuelle¹

¹ONIRIS, INRAE, LABERCA, Route de Gachet, CS 50707, 44307 Nantes, France

²TOXALIM – INRAE, UMR 1331, 180 chemin de Tournefeuille, BP 93173, 31027 Toulouse Cedex 3, France

³Univ Rennes, Inserm, EHESP, Irset (Institut de Recherche en Santé, Environnement et Travail) - UMR_S 1085, F-35000, Rennes, France

tarek.moufawad@oniris-nantes.fr



Introduction

Pesticides exposure assessment

External exposure

Internal exposure

Questionnaire

Targeted Screening

Suspect Screening

Unknown Screening

Indirect

Estimation

Quantitative

Focused

Semi-Qualitative

Medium Scale

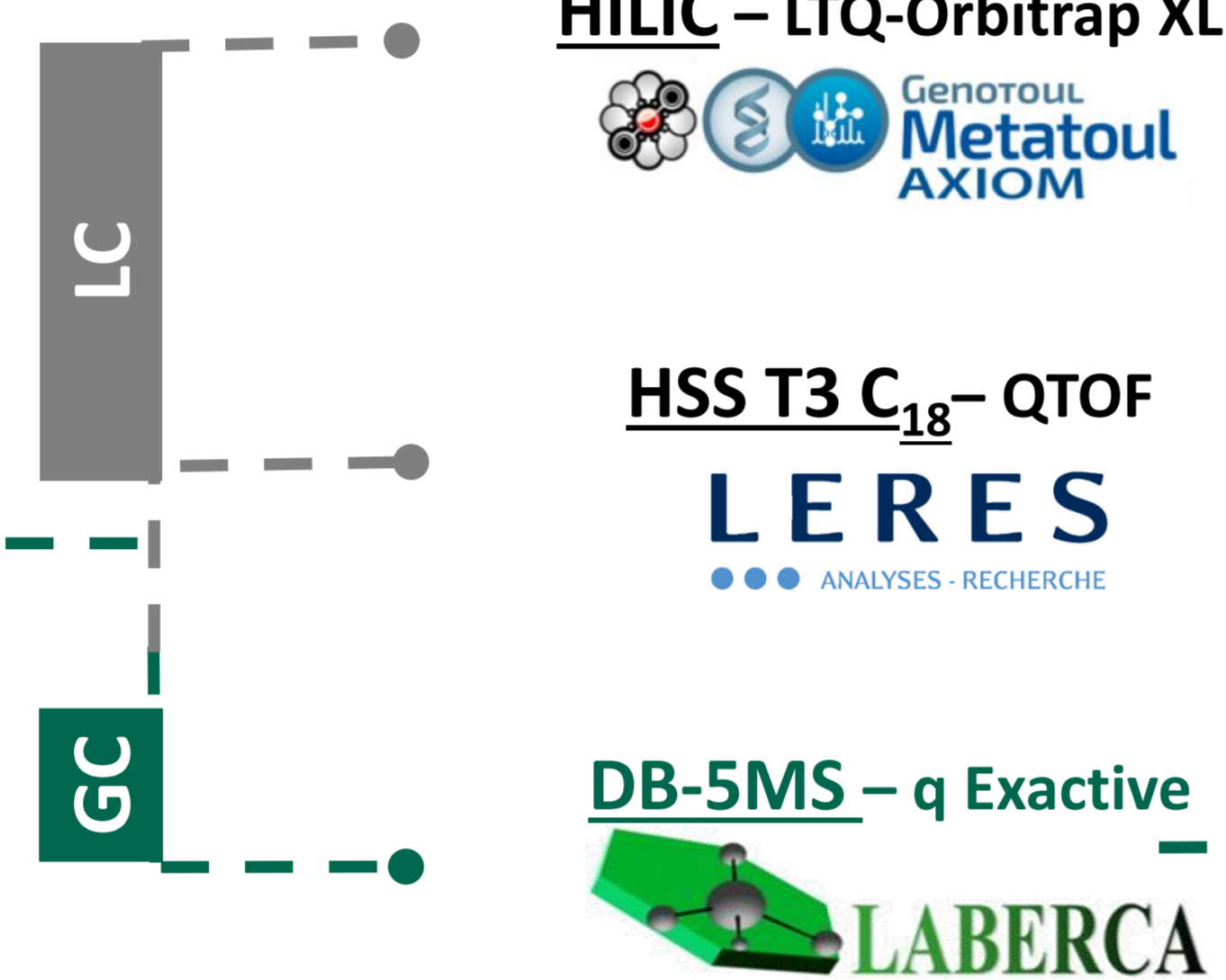
Qualitative

Large scale

The Screenpest Project

Providing a methodology for broad assessment of human internal exposure to pesticides, via suspect screening, based on urine samples.

Development of a unique protocol for analysis of a single urine sample by 3 complementary analytical platforms providing extensive picture of the internal exposure to pesticides, including both parent compounds and metabolites.

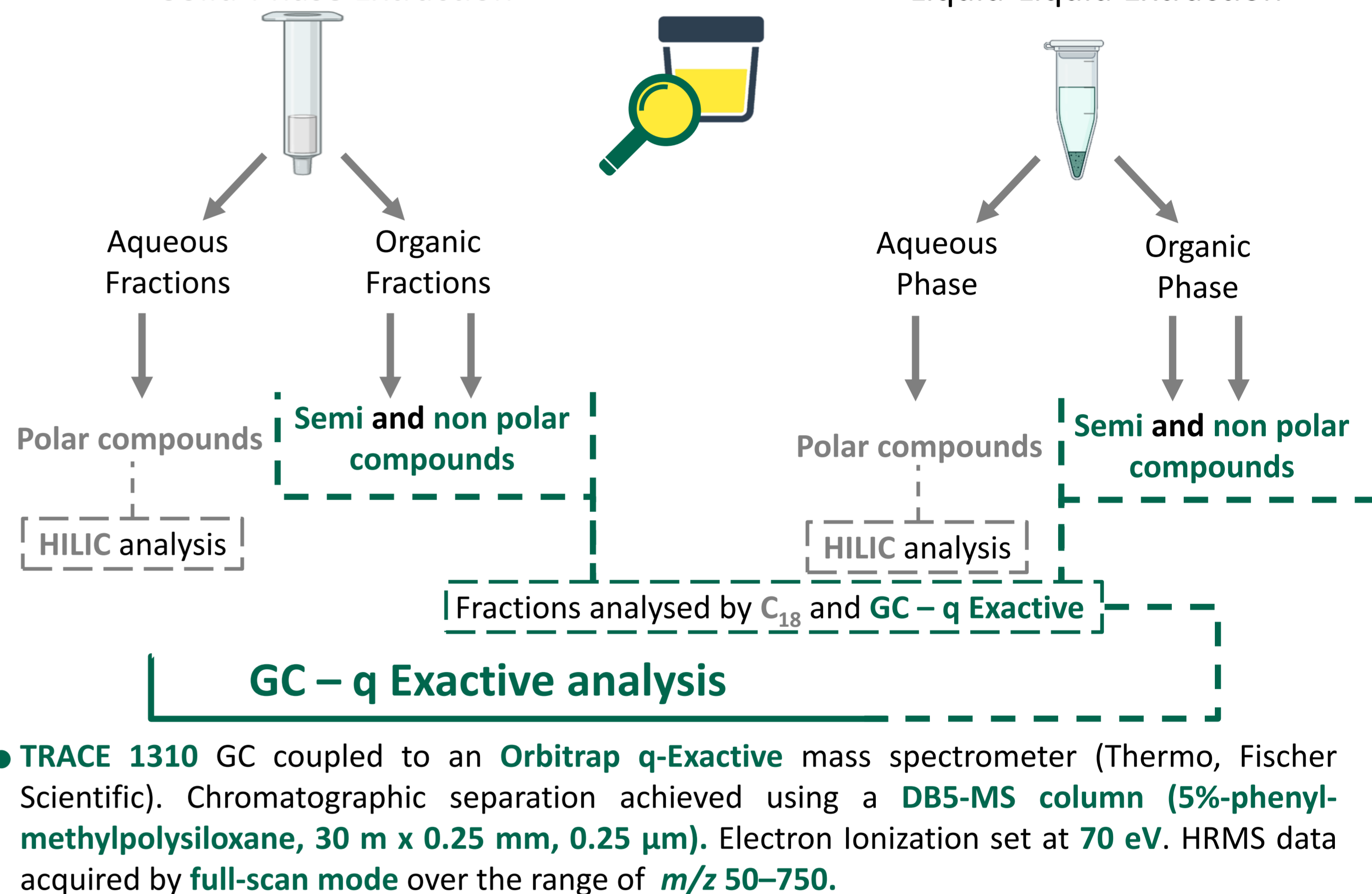


Experimental

Extraction Techniques

Solid-Phase Extraction

Liquid-Liquid Extraction

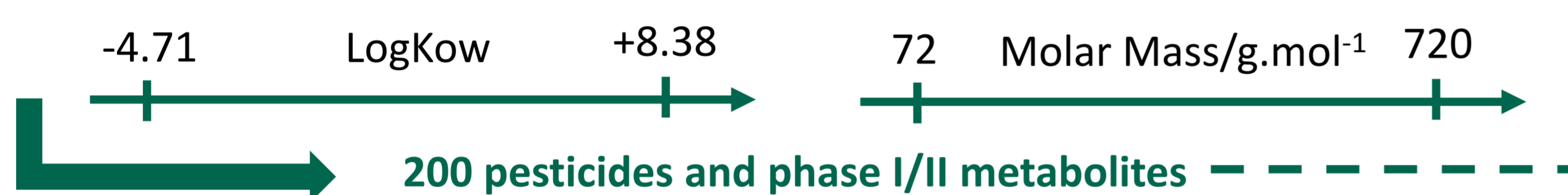


Results and Discussion

Suspects List

Pesticides **suspect list** elaborated based on commercially available compounds, known pesticide usage in France, epidemiological expertise and extensive database elaborated from the HMB4EU project.

Novel multiplexed approach allowing to cover a wide structural diversity.



HRMS spectral libraries

1-For Suspect Screening

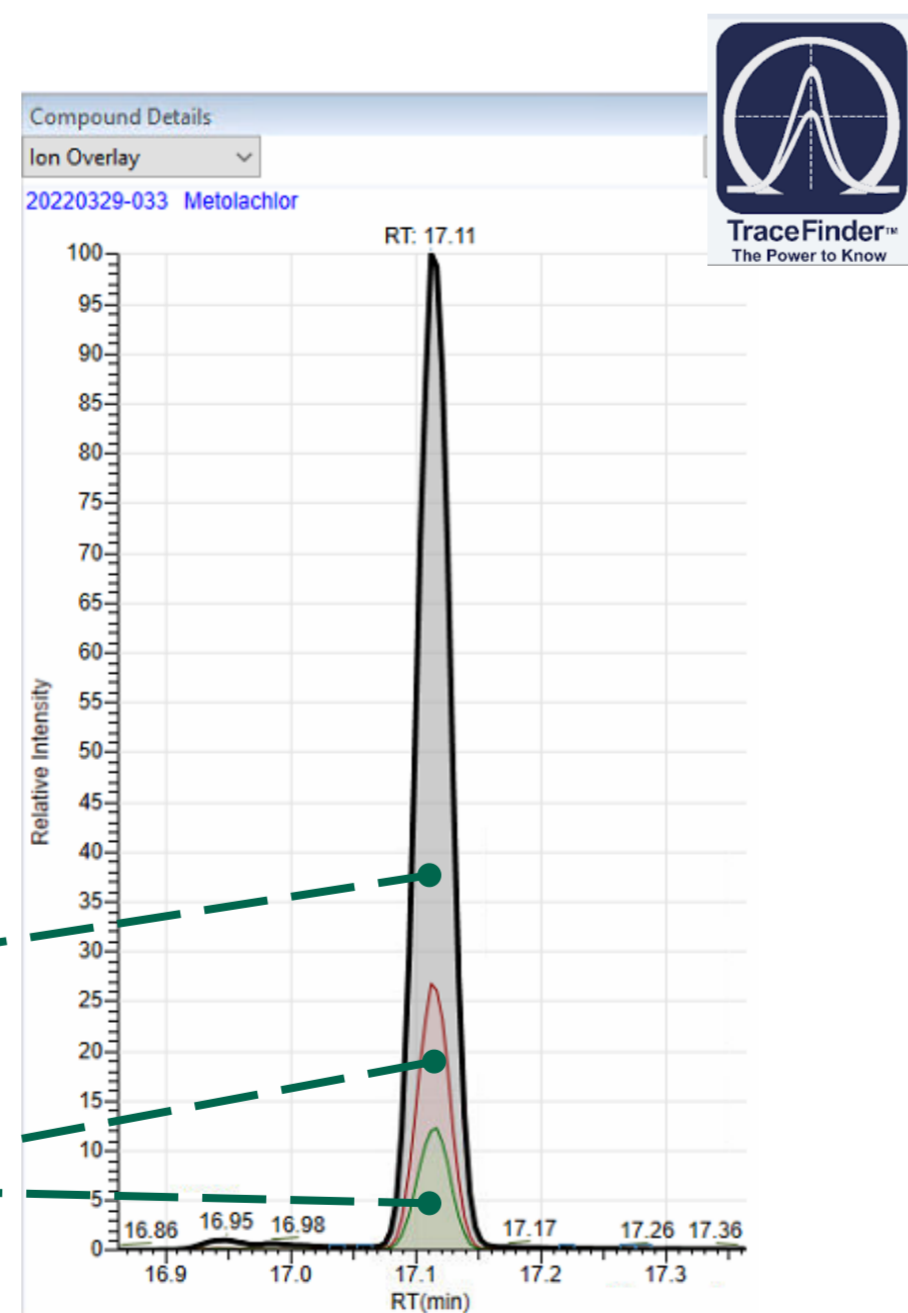
A first **experimental spectral library** was built using TraceFinder (ThermoFisher), allowing the **annotation** of the detected **exposure markers** with a **high confidence level**, using the following criteria:

- Retention time
- Base peak (most abundant fragment ion)
- 2 additional fragment ions
- 2 m/z abundance ratios between the base peak ion and the 2 additional fragment ions

Highest annotation/ Identification confidence level of - Level 1/2 (based on Shymanski scale)
doi.org/10.1021/es5002105

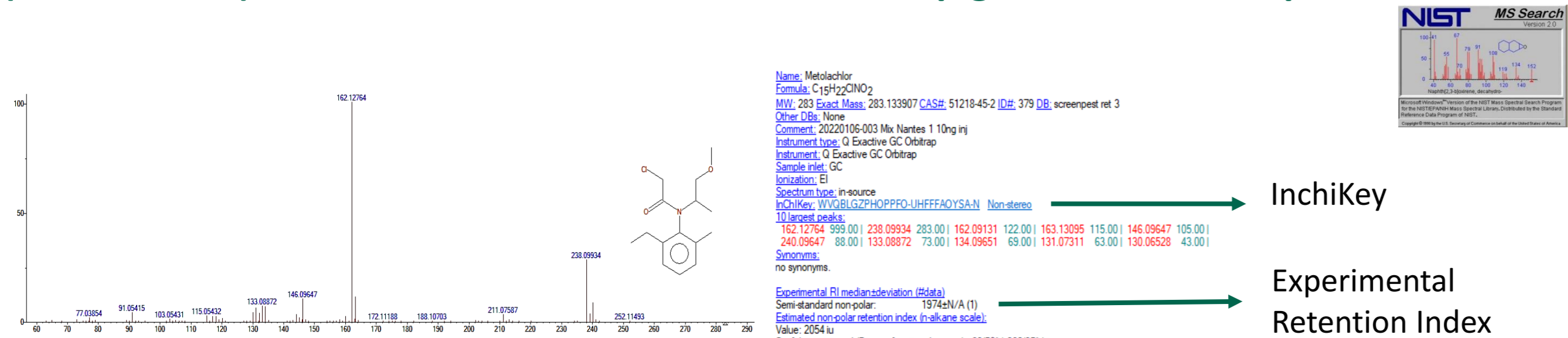
Base peak used for detection and quantification

Additional fragments used for the confirmation of the detected analytes, with an acceptable ion ratios



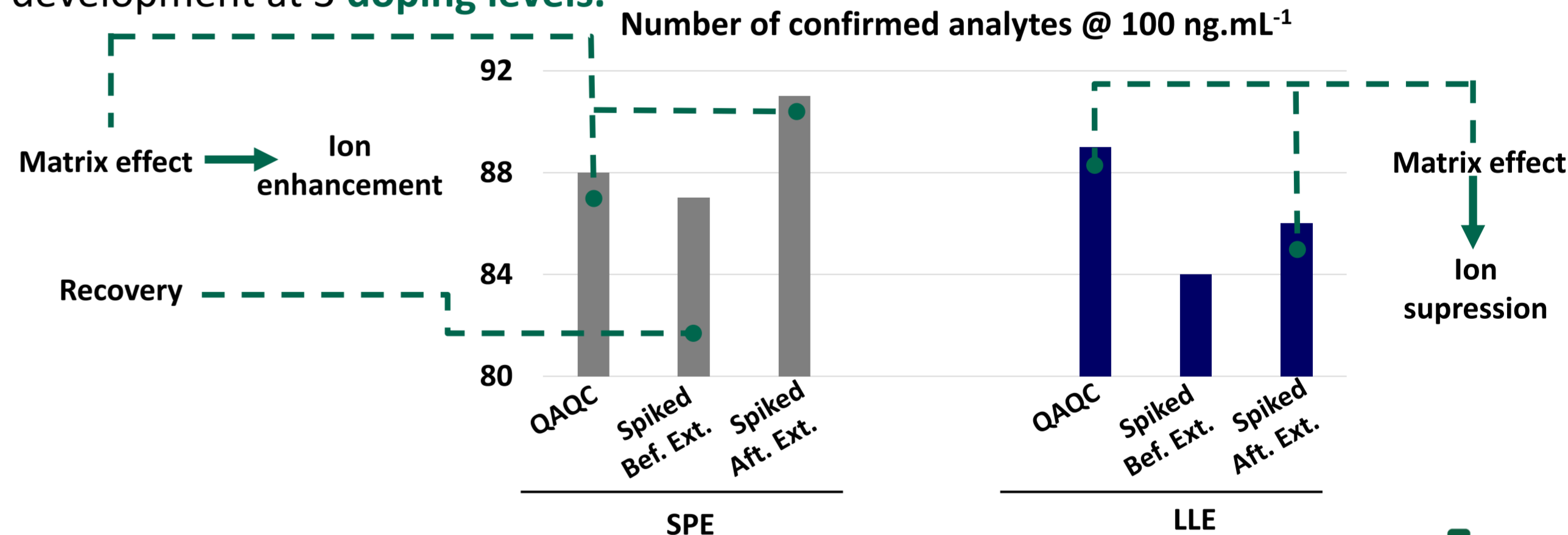
2-For Unknown Screening

A second **experimental spectral library** was built using **MS search NIST software**. The use of the NIST database gives access to a **well documented, interchangeable and universal HRMS library**, which can be **integrated** into in house annotation softwares (Peakforest...) or **enrich online libraries** with Orbitrap generated mass spectra.



Method development

103 compounds were used as **QA/QC markers** as **positive control (pooled urine samples)** for GC method development at **3 doping levels**.



GC-MS performance maintenance

- Boosting GC performances :** → Using Focus Inert Liner, pure cyclohexane & septa 100% PTFE without siloxane
- Handling and Monitoring** the q-Exactive system, focused on the **Ion source** : → Repeller and the different lens voltage, throughout cleaning and adjustment.
- QA/QC** → On cue system **pre-check & control chart** from QC all along the batch

Exposure markers detection

11 pesticides were successfully detected and confirmed in **real human pooled urine** → **Confirmation of the applicability of the developed technique for suspect and unknown screening approaches**

Conclusion & Perspectives

- Elaboration of a **suspect list** covering **200 substances** with a **wide range of physicochemical properties**
- Elaboration of an **experimental extended HRMS spectral library** of reference pesticides and their metabolites
- Evaluation of **different extraction techniques** for pesticides extraction from urine samples
- Development of a **GC-HRMS based workflow** for characterising **human internal pesticides exposure**
- Detection and confirmation of **11 pesticides related exposure markers** in **real pooled human urines**
- Application of a multiplexed approach on real urine samples from (PELAGIE / NutriNet-Health cohorts) → Linking **pesticides exposure** with possible **adverse health effects**
- Increase the knowledge of the **human exposome to pesticides**
- Early warning on **emerging substances** & Weighing in the **decision making**