BIPM Capacity Building & Knowledge Transfer Programme

2025 BIPM - TÜBİTAK UME Project Placement

REPORT

Project Name	Determination of per- and polyfluoroalkyl substances (PFAS) in food matrices
Description	The project consisted of several key stages: sample preparation and processing, preparation of stock and spiking solutions, spiking of the samples, and the application of extraction techniques, as well as training focused on method validation and measurement uncertainty evaluation.
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Motivation & Introduction

The main purpose of this project placement was to strengthen my technical skills in method development for PFAS analysis in food matrices and to gain a deeper understanding of the process of preparing certified reference materials (CRMs).

PFAS are a large group of man-made chemicals widely used for their water-resistant properties in everyday products, such as cookware, textiles, and food packaging. Because of their persistence in the environment, ability to accumulate in living organisms, and potential harmful effects, PFAS have become one of the major emerging contaminants of concern. Their presence in food, either through environmental contamination or contact materials, poses a growing risk to food safety.

Given this context, the development of sensitive analytical procedures for detecting and quantifying PFAS in food and the preparation of traceable and highly accurate reference materials for quality control and quality assurance purposes are of great importance for ensuring reliable testing and supporting food safety laboratories.

It is important to highlight that TÜBİTAK UME is participating in the SCREEN FOOD 23IND13 project, and its role within the project is method development and production of certified reference materials of PFAS in food samples. The laboratory work performed during this placement, including carrying out all calculations, preparing stock and spiking solutions, sample preparation, extraction optimization represents a valuable basis for the next stages of CRM production and further method validation.

Working with the experienced staff at TÜBİTAK UME, who shared their expertise in method development, uncertainty evaluation, and CRM production, was a valuable part of this placement. The knowledge and experience gained through this collaboration will help to strengthen the scientific and technical capabilities of IMBIH, especially in the area of PFAS determination and reference material production.

Research

The research involved several stages, beginning with sample and reagent preparation, followed by extraction trials and training in method validation and uncertainty evaluation.

Organic chemistry laboratory

Before my placement began, initial sample processing of tomato and infant food had already been performed. During my time at TÜBİTAK UME, I continued this work by preparing new batches of samples and conducting extraction trials to support future PFAS analysis.

Tomato and infant food samples were selected for this placement based on the decisions made during the project meeting. Food matrices were chosen to represent categories posing different analytical challenges, including high water and high protein content.

For the purpose of method development, all calculations for preparing individual stock solutions, both native and labelled, as well as for the calibration curve, were performed. Mixed stock solutions for spiking and method development were prepared by dilution in methanol: 17 analytes at 500 µg/kg, 4 analytes at 500 µg/kg, and 1 analyte at 250 µg/kg. Additionally, another stock solution of 50 µg/kg was prepared for 22 analytes (originally planned for 24). Two analytes were not included in the stock solutions: one was unavailable, and PFNA was excluded due to its anhydride form.

The four analytes at 500 µg/kg (PFOS, PFOA, PFNA, and PFHxS) were prepared separately, as they require different concentrations in the regulation and candidate reference material, they will be certified, while the remaining analytes will be reported as informative values. The single analyte at 250 µg/kg was also prepared separately because its original concentration was lower than the others.

Extraction trials were performed on infant cereal-based food and tomato matrices. Centrifugation parameters (speed, temperature, and time) were optimized to improve phase separation.

The first stage of the work involved preparing reagents such as tetrabutylammonium (TBA) and sodium carbonate (Na_2CO_3), followed by several pH adjustment trials using sodium hydroxide (NaOH). Extraction trials on both infant food and tomato samples highlighted the importance of pH control and ion pairing for transferring polar PFAS compounds into the non-polar solvent (MTBE).

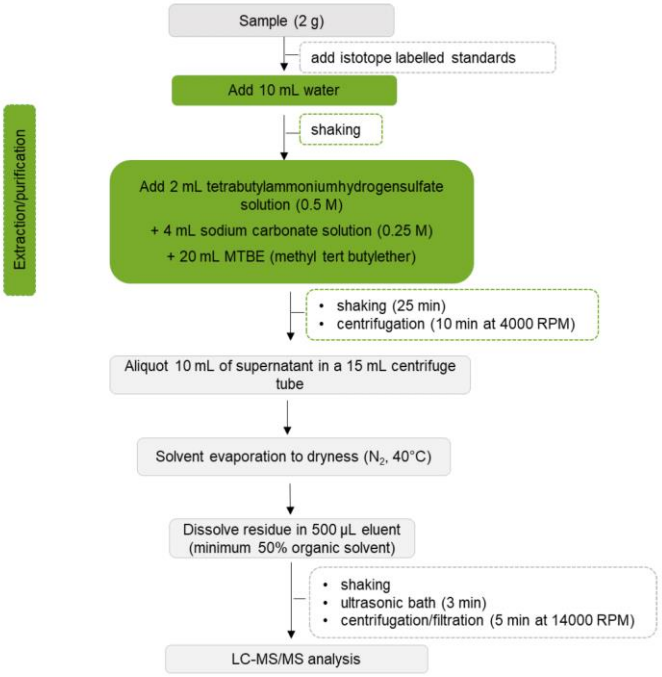

Figure 1. Extraction method used, adopted from Sample Preparation and LC-MS/MS Method for the Determination of 33 Per- and Polyfluoroalkyl Substances (PFAS) in Food of Plant Origin (EURL, 2024).

Figure 2. Sample extraction tomato (left) and infant food (right)

For tomato samples, the method was successful and optimized. However, for infant food sample, this method turned out to be unsuitable, and in the future QuEChERS method will be applied, which should be more suitable than our previous extraction method.

Although the PFAS analyses could not be completed during the placement due to the temporary unavailability of the Orbitrap LC-MS instrument, the preparation phase I carried out forms an important foundation for the next steps of the project. The processed and spiked samples will be used in future analytical measurements, supporting the goal of further method development and production of Certified Reference Materials for PFAS in tomato and infant food samples.

Following this work, I participated in preparing spiking solutions and assisting in the preparation of candidate reference materials of PFAS.

Reference materials laboratory

Work in sample processing for candidate reference materials for 22 PFAS analytes in tomato and infant food samples included the following:

Tomato sample

A total of 70 kg of fresh organic tomatoes was obtained. The process involved manual cutting and removal of green parts, followed by stepwise machine milling in approx. 3 kg batches to achieve a sufficiently homogeneous material. Each batch added to the milling machine was weighed, and the final mass of the processed sample was recorded.

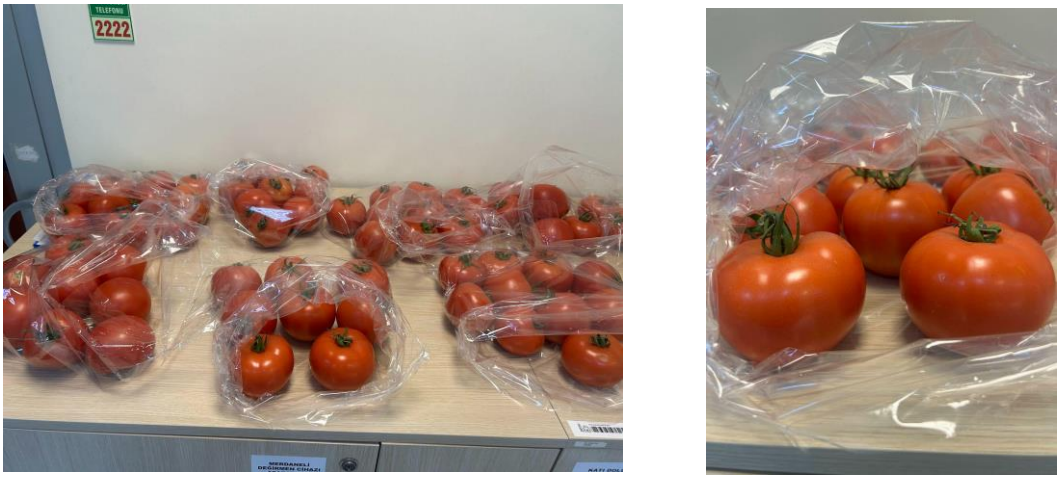

Figure 3: Fresh organic tomatoes

Figure 4: Manual cutting and removal of green parts from fresh tomatoes

Figure 5: Beginning, middle, and final stages of milling fresh organic tomatoes

After processing, a few samples were taken from the main batch: approximately 6 kg for spiking and around 2.3 kg for blank sample to be freeze dried. The remaining material was stored at –20 °C for further use and developing CRM.

Spiking-tomato sample

Spiking solutions were prepared from previously prepared stock solutions. Four levels of spiked samples were made, along with a zero level for blank. Each level contained approximately 1.5 kg of processed tomato sample and was spiked with slightly more than the calculated amount to account for potential analyte loss during processing.

Each 1.5 kg batch was carefully spiked and rinsed with a small amount of methanol to ensure complete transfer, then mixed using a laboratory mixer for 30 minutes. After mixing, approx. 10 g of sample was aliquoted into 15 ml centrifuge tubes (16 tubes per level) for analysis by project partners. This procedure was followed for all levels, including the zero level.

The remaining material from each 1.5 kg batch (after taking approximately 160 g for project partner samples of fresh tomato) was transferred to aluminum containers for freeze-drying. Containers for each level, including zero, were placed in the lyophilizer for several days. After freeze-drying, the samples were ground, sieved, and placed into centrifuge tubes for analysis by project partners. In the end, two types of spiked tomato samples were prepared, both originating from fresh tomato: one was frozen, and the other was freeze-dried.

Figure 6. Filling centrifuge tubes for analysis by partners and the filled tubes

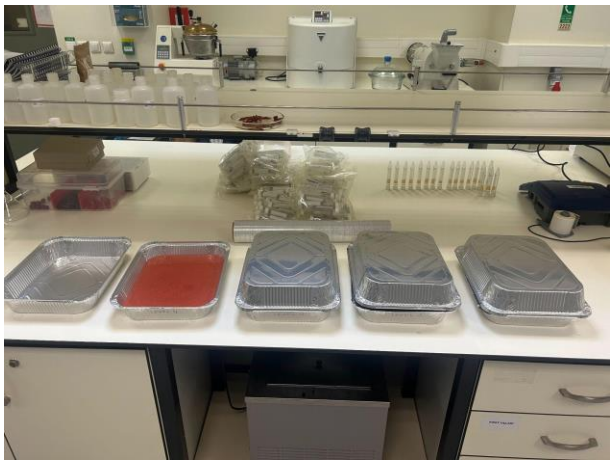

Figure 7: Remaining of the each batch of 1.5kg prepared for freeze-drying

Spiking-infant food sample

For the infant food, the sample was obtained directly from a manufacturer of infant food. First, 40 g of the sample was spiked using the previously prepared stock solutions to create a primary spiked matrix. Approximately 200 mL of methanol was added to ensure complete mixing, and the mixture was processed in a rotary evaporator to remove the solvent and obtain a homogeneous powdered material. This spiked matrix was then used to prepare additional batches of samples at different concentration levels (same as tomato, four spiked levels and zero blank level). In this way, the batches that were later on used to fill centrifuge tubes were not spiked directly with stock solutions, but were instead carefully mixed with the pre-prepared 40 g spiked matrix. This approach ensures better homogeneity and more consistent spiking across all samples that will be used for analysis by project partners.

All calculations, including the preparation of stock and spiking solutions as well as mass recordings, were performed and documented using Microsoft Excel.

Trainings and seminars

The project placement also included training on method validation and calculation of measurement uncertainty, using examples for various methods developed at UME. For this purpose, among other literature the Eurachem Guide *The Fitness for Purpose of Analytical Methods* (2025) was used. All calculations were performed using Microsoft Excel and the Curve Expert program. Dr. Burcu Binici delivered a detailed presentation, provided extensive literature resources, and addressed all questions related to method validation and uncertainty assessment.

Additionally, Dr. İlker Ün gave an insightful presentation on NMR spectroscopy and its future applications in metrology, supported by examples involving different types of samples.

Dr. Taner Gökçen introduced the HPLC technique through practical experiments focused on the analysis of Ochratoxin A (OTA) and shared literature and technical insights on chromatographic analysis.

During this project placement, we also participated in a seminar organized by BIPM and TÜBİTAK UME. The seminar covered topics such as global quality infrastructure and the role of metrology, the CIPM MRA and its importance within the quality infrastructure, CIPM requirements and documentation for QMS, RMOs, comparisons, CMCs, developing uncertainty budgets, and a demonstration of the KCDB. Each participant also presented their work, making the seminar highly interactive and providing valuable opportunities to discuss these topics.

Conclusions and Future Work

I gained expertise in analytical method optimization, validation, and evaluation of measurement uncertainty, along with a deeper understanding of CRM production. These skills will contribute to my professional growth and support IMBIH's ongoing efforts in reference material development. This work also aligns with IMBIH's ongoing ISO 17034 accreditation process and will strengthen quality assurance and service expansion.

Acknowledgements

I would like to express my sincere gratitude to everyone involved in organizing the BIPM–TÜBİTAK UME project placements. This is a great opportunity to expand knowledge, share experiences, and foster multicultural collaboration.

I am truly grateful to have had the chance to participate in such a program at UME, where everyone has been kind and supportive.

In particular, I would like to thank my mentor, Dr. Burcu Binici, for her generous guidance, knowledge sharing, and continuous support throughout this project.

I would also like to express my appreciation to Dr. Taner Gökçen for his insights and technical support. Also I would like to express my gratitude to the Dr. Alper İŞLEYEN and Dr. Şükran AKKUŞ ÖZEN from the Reference Materials laboratory for their knowledge transfer in the field.

Finally, I would like to thank Ms. Müge Atam from the International Relations Department for ensuring that everything went smoothly during my stay.