

INVESTIGATION ON THE CAPABILITIES OF A NEW PARALLEL PRESSURIZED LIQUID EXTRACTION (PLE) SYSTEM FOR DIOXINS AND PCBs IN BIOLOGICAL SAMPLES

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Abstract

Preparing biological samples for dioxin and PCB measurement is a task that is challenging many research centers and routine laboratories. For either human or food-related matrices, the implementation of proper procedures requires time, know-how, and money. Among other techniques, pressurized liquid extraction (PLE) has been used for several years to save time and to reduce solvent consumption, while maintaining good extraction efficiency. Samples are normally sequentially extracted and extraction sequences can be performed unattended overnight. The unavoidable clean-up procedure that follows the extraction step has also evolved over the last 15 years to offer automated parallel sample processing capabilities, based on multi-column set-ups and computer piloted solvent deliveries. Recently, a novel PLE instrumental design has been proposed to allow parallel extraction of several samples in order to better fit with sample throughput requirements of the parallel clean-up procedure. We investigated the use of both parallel PLE and clean-up procedures for sample preparation of reference and quality control food and serum samples. Recovery rates, extraction and transfer efficiency, accuracy, precision, robustness, and usability of the system have been estimated.

Introduction

The ‘Quest for the Holy Grail’ in the dioxin analysis area is dedicated to the development of reliable procedures that can offer congener-specific results on a short time scale, at a low cost, while avoiding down time issues. Such a procedure obviously has to fulfil strict QA/QC requirements such as the ones listed in Eurachem analytical guidelines and EU Directives, but also has to comply with ISO17025 and GLP procedures. Each part of such a procedure, namely extraction, clean-up, fractionation, chromatographic separation, and physico-chemical measurement, has to be fine tuned to their optimum capabilities (one could name this approach the ‘Analytical Dioxinomics’, since ‘omics’ refers to ‘research that studies entities in aggregate’...). The present paper focuses on the testing of the parallel capability of the PLE approach for extraction of selected PCDD/Fs and PCBs in biological matrices. This can potentially be a simple alternative to the extraction-clean-up-fractionation (ECF) approach we reported earlier¹ in terms of the potential use of the method.

Materials and Methods

Extraction system

The extraction system set-up is illustrated in Figure 1. The HPLC pump max flow rate is 40mL/min. The maximum pressure is 2500 psi, the maximum practical temperature is 200°C. Transducers permit pressure and temperature on-line monitoring as well as the recording of their values for archive of extraction parameters. A pressure relief valve

ensures safe operation and the collection of relief valve effluent in the sample vial avoids losing analytes. Nitrogen is used to purge the cell at the end of the extraction cycle.

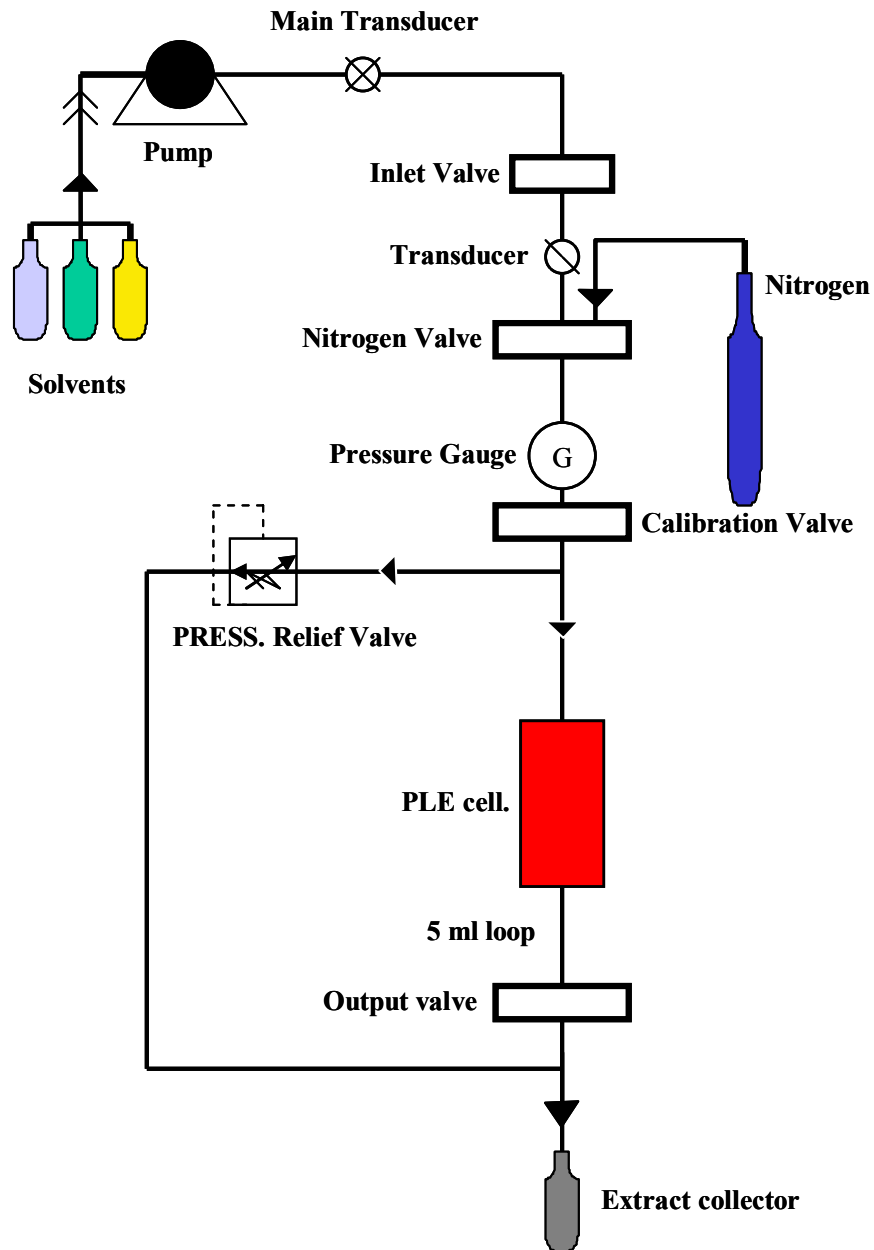


Figure 1 : Plumbing diagram of one extraction line of the parallel PLE system.

Extractions were performed using hexane at 1500psi for 15min, extraction cell of 90mL, at 120°C, 2 extraction cycles, flush with nitrogen.

Test Material

BCR607, a spray dried milk powder, and CDC Serum QC samples were used for testing. The samples (10g milk powder or 15mL serum) were mixed with sodium sulphate and inserted inside the extraction cell between two plugs of pure sodium sulphate. The internal standards were added at the entrance of the cell.

Target compounds

The seventeen 2,3,7,8 toxic PCDD/Fs, and 40 PCBs were measured by GC-HRMS.

Sample clean-up and GC-HRMS measurement

The sample extracts were reduced by evaporation prior to clean-up using the Power-Prep system from Fluid Management Systems (Waltham, MA, USA). PCDD/Fs and non-ortho PCBs were fractionated from other PCBs. GC-HRMS was performed on a MAT95XP under a validated method for human biomonitoring used at the CDC.

Results and discussion

Figure 2 and 3 illustrates the extraction efficiency for the tested matrices.

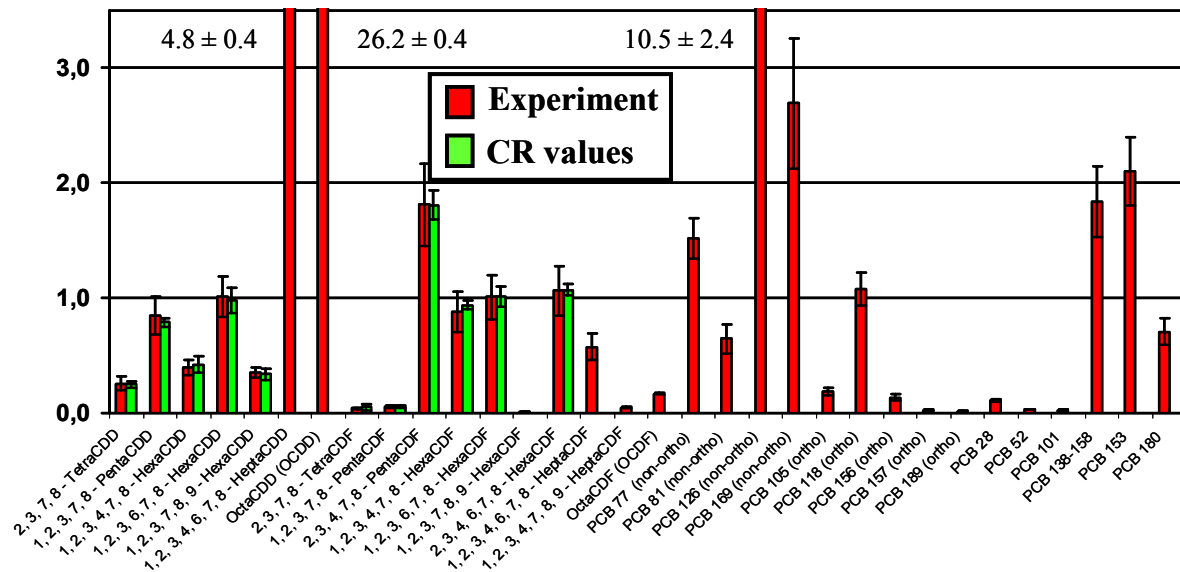


Figure 2 : BCR607 (n=3) in ppt for PCDD/Fs and non-ortho-PCBs, in ppb for other PCBs.

For BCR607, only a few congeners have a certified value available for comparison. RSDs ranged from 3-8% for the certified values and below 20% for all others. Setting the certified value as 100%, all measured concentrations were between 100% and 116% (all overestimations). For all analytes, the recovery rates ranged between from 60-110%. For serum QC samples, all congeners have a reference value available for comparison. RSDs were from 2-10% for all others (except OCDD 14%). Setting the certified value as 100%, most measured concentrations were between 90% and 110% (CB-126 was 50%, still not clearly elucidated). For most analytes, the recovery rates ranged between 60-110% .

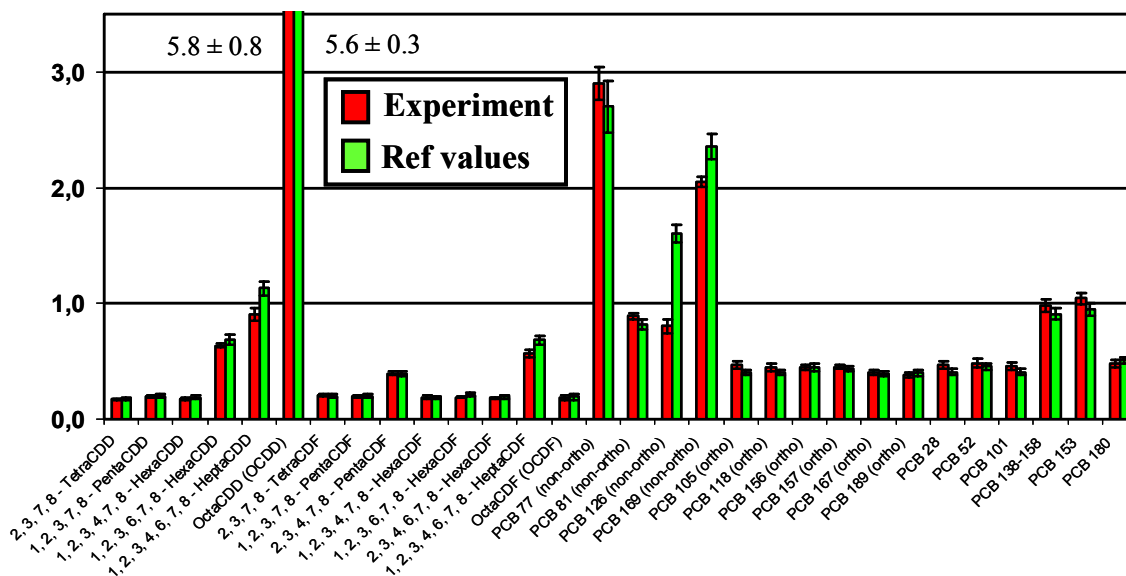


Figure 3 : QC serum pool (n=4) in ppt for PCDD/Fs and non-ortho-PCBs, in ppb for other PCBs.

The extraction stainless steel cell costs 15 EUR and each cap costs 8 EUR. Both are reusable, if needed. The extraction cell volumes range from 1 to 250mL. The modular system allows the extraction of 6 samples in 1.5h. The decontamination of the system between runs is easily carried out by flushing and pressurising a low volume of solvent inside the lines and in the pressure relief line. So far, no cross-contaminations have been observed. Compared to the ASE, one can expect less tubing clogging as the inner diameters are significantly larger. The system is characterized by a good sample throughput-cost effective ratio, even for limited size laboratories because a single line design is available. Finally, an interesting feature of the PLE system is to be able to accommodate a sulphuric acid impregnated silica column right after the extraction column, allowing 'on-line' partial clean-up without the disadvantage of producing highly corrosive fluids under high pressure, as is the case when adding such sorbent inside the ASE cell.

References

1. Focant J.-F., Pirard C., Massart A.-C., Scholl G., Eppe G., De Pauw E., *Organohalogen Comp* 2005; 67:261.