MEASUREMENT OF DIOXINS AND WHO PCBs IN FOODSTUFFS USING GCXGC-IDTOFMS

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INTRODUCTION

Together with gas chromatography quadrupole ion storage mass spectrometry operated in tandem mode (GC-QISTMS/MS), comprehensive two–dimensional gas chromatography coupled to time-of-flight mass spectrometry (GCxGC-TOFMS) represent potential mass spectrometric alternative to the use of GC high resolution mass spectrometry (GC-HRMS) based on sector instruments for the measurement of dioxins and PCBs [1]. The present work was carried out to evaluate the capability of GCxGC-TOFMS performing in isotope dilution (ID) for the measurement of dioxins and PCBs in various foodstuffs. A comparison with GC-HRMS is presented.

MATERIAL AND METHODS

Sample preparation

Solid samples are extracted with hexane using PLE (ASE200^{TM}, Dionex, Sunnyvale, CA, USA) after overnight lyophilization. Fat QC samples are directly processed through the clean-up step. The commercially available automated Power-Prep^{TM} system (FMS Inc., Waltham, MA, USA) was used for further sample clean-up. In its basic form, this modular system is made of PC-controlled valves and pumps that manage solvents and samples through various types of disposable columns. Column sizes and sorbent types vary depending applications but classical sets are usually made of multi-layer silica, basic alumina and carbon sorbents. Details on sample preparation procedures are available elsewhere [2-4].

Measurements

Gas chromatography-magnetic sector high resolution mass spectrometry (GC-HRMS)

For PCDD and PCDF analyses, an Agilent 6890 GC was connected to an Autospec Ultima HR mass spectrometer (Micromass, Manchester, UK). It operated at a resolution of 10,000 in the selected ion monitoring mode (SIM) with a RTX-5MS 40m x 0.18mm ID x 0.18µm f.t. (Restek, Interscience, Belgium). For PCBs, a Finnigan MAT95XL (Finnigan, Bremen, Germany) HR mass spectrometer was used. It operated at a resolution of 10,000 in SIM with a HT-8 25m x 0.22mm ID x 0.15µm f.t. (Achrom, Interscience, Belgium). Helium was used as the carrier gas at constant flow rate of 1.2 ml/min. 2 µl of the final extract in nonane were injected into a split/splitless injector held at 275°C in splitless mode. For PCDD/Fs, the oven temperature was 140°C for 1 min, followed by an increase to 200°C at 52°C/min, then increase to 235°C at 2.9°C/min and hold for 10 min, finally

increase to 300°C at 6.9°C/min and hold for 6 min. HRMS parameters conditions, quantification and insurance quality control for measurements were described previously [2].For PCBs, the oven temperature was 140°C for 2 min, followed by an increase to 220°C at 15°C/min for 7.5 min, then increase to 250°C at 6°C/min, then increase to 260°C at 2°C/min, then increase to 320°C at 12°C/min

Gas chromatography-comprehensive two-dimensional gas chromatography-time-of-flight mass spectrometry (GCxGC-TOFMS)

The instrument was the Pegasus 4D (Leco Corp., St Joseph, MI). The 'quad jet' modulator was mounted in an Agilent 6890 GC oven and liquid nitrogen was used to create the cold jets (Figure 1). Details regarding the system have been reported elsewhere [5,6].

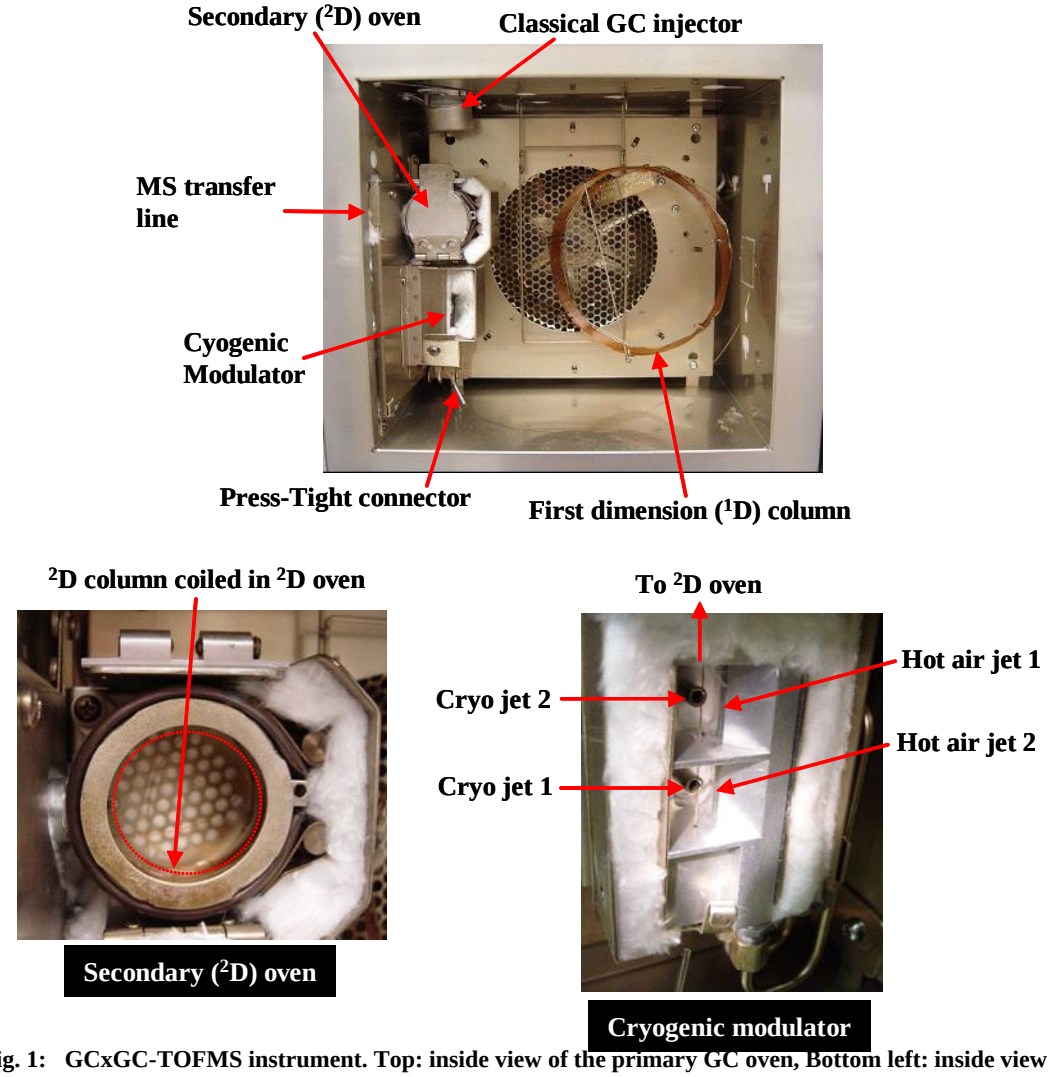

Fig. 1: GCxGC-TOFMS instrument. Top: inside view of the primary GC oven, Bottom left: inside view of the secondary oven, Bottom right: inside view of the cryogenic modulator.

Splitless 1.5 µl injections were performed at 280°C. Carrier gas was helium and a constant flow of 0.8 ml/min was used. The GC column set was a RTX-500 40m x 0.18mm ID x 0.11µm f.t. (Restek) as ^{1}D and a BPX-50 1.5m x 0.10mm ID x 0.10µm f.t. (SGE, Austin, TX, USA) as ^{2}D. Deactivated universal presstight connectors (Restek Corp., Bellefonte, PA) were used. ^{1}D temperature program was 140°C for 1 min, followed by an increase to 240°C at 10°C/min, then increase to 260°C at 1°C/min and hold for 5 min, finally increase to 330°C at 10°C/min and hold for 5 min. ^{2}D temperature program was 160°C for 1 min, followed by an increase to 260°C at 10°C/min, then increase to 350°C at 2.8°C/min and hold for 5 min. The temperature of the modulator had an offset of 40°C compared to the temperature of ^{1}D oven. The modulator period was 4 s with a hot pulse duration set at 750ms and a cool time between stages of 1250ms. The MS transfer line temperature was 275°C and the MS source temperature was 250°C. The data acquisition rate was set at 60 scans/s for a mass range of 100 to 550 *m/z*.

RESULTS AND DISCUSSION

Although coplanar PCBs (CB-77, CB-81, CB-126, CB-169) are commonly isolated in the PCDD/F fraction during the sample clean-up, we modified the fractionation process of the automated system to get all the PCBs collected in the same fraction (Figure 2). The reason was the improvement of the LODs for those cPCBs by reducing the quantity of solvent to be used and simplify the global fractionation procedure. In addition, due to the increased peak capacity of GCxGC, larger number of analytes can be measured simultaneously while maintaining good separation efficiency.

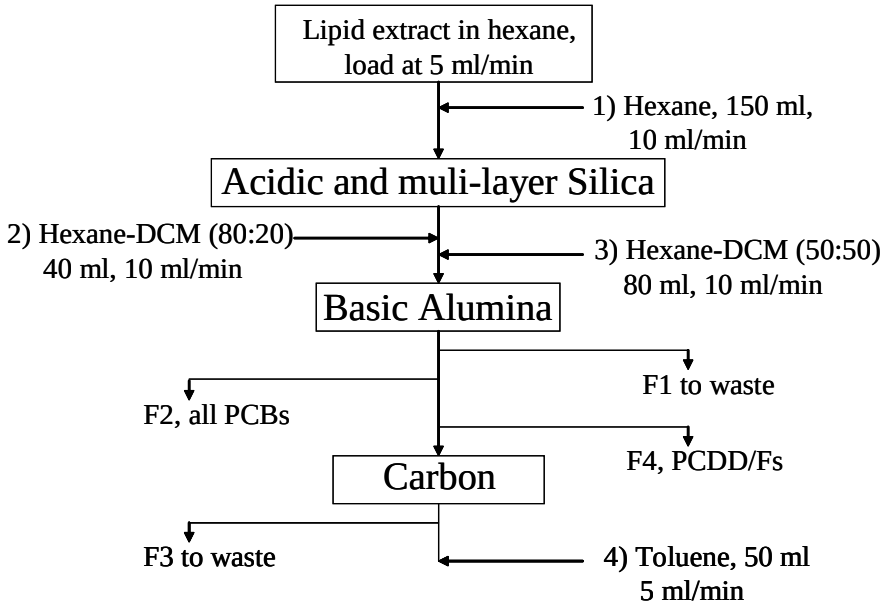

Fig. 2: Modified flow chart for the automated sample preparation procedure.

Several GCxGC column sets were tested to optimize the separation of the 17 PCDD/Fs, the 12 WHO PCBs, as well as the 6 marker PCBs (Aroclor 1260) from a single injection (DB-5/DB-17, DB-1/HT-8, HT-8/BPX-50, RTX-500/HT-8, RTX-500/RTX-CLPesticides, RTX-500/SP-2331 and RTX-500/BPX-50). Among them, the RTX-500/BPX-50 column set offered the best separation of critical congeners such as the HxCDDs and HxCDFs (Figure 3).

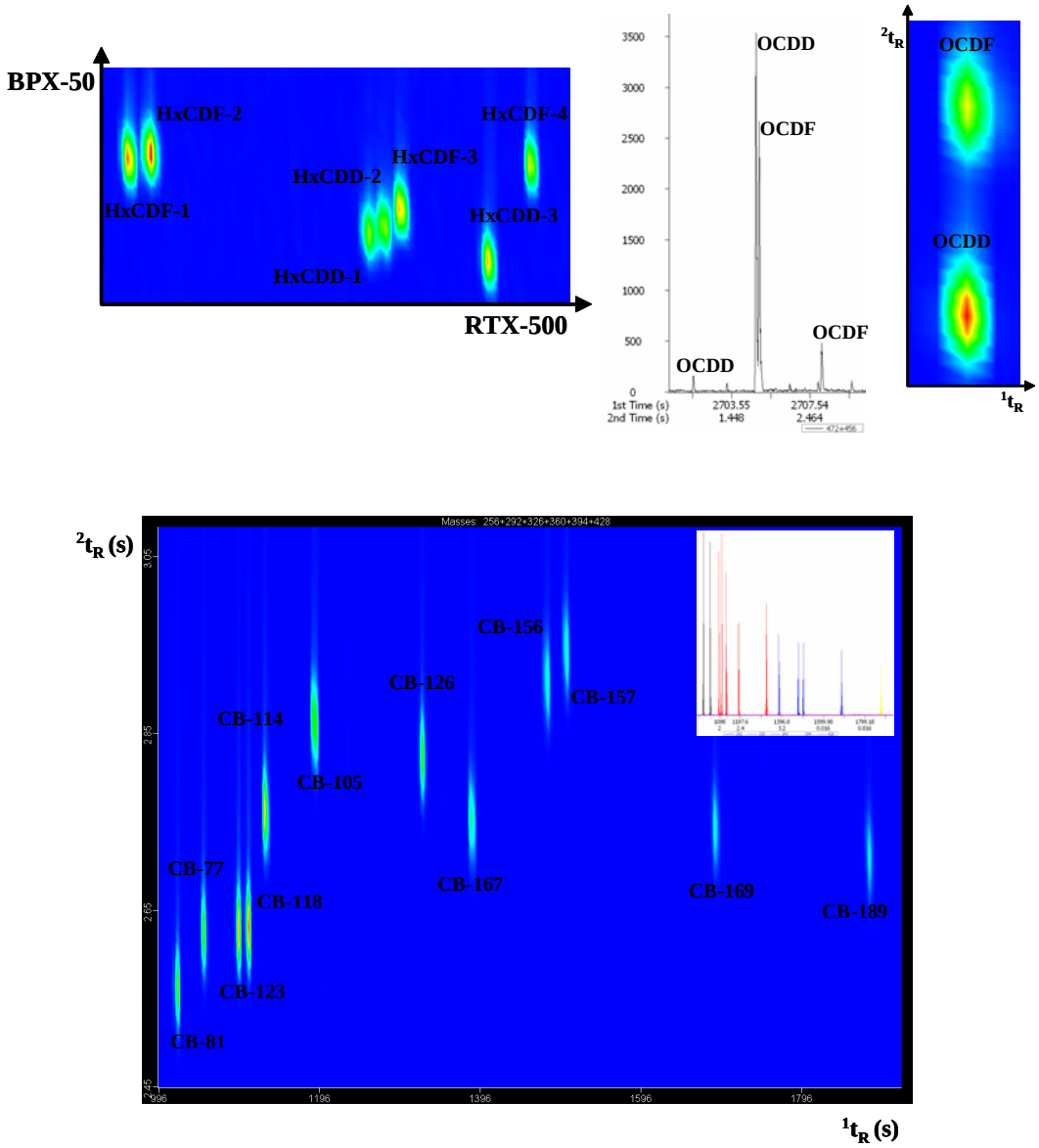

Fig. 3: Separation of the HXCDD/Fs (top left), the OCDD/F (top right), and the PCBs on the RTX-500/BPX-50 column set

The RTX-500 column appeared to have an excellent capability to nicely separate CB-118 from CB-123. Those two congeners are generally not quite well separated, even using the PCB-specific HT-8 phase. All the congeners eluted out of the column set after 45min (CB-189 was out of ^{2}D column after 31 min and OCDD was out after 45 min).

The calibration curves of all analytes were carried out by isotope dilution. The concentration range for PCBs was 2 to 280 pg/µl and the calibration range for PCDD/Fs was 0.2 to 50 pg/µl (2,3,7,8-TCDD). Figure 4 illustrates a calibration curve obtained after 1.5µl of calibration standard (0.3 pg on column for the lowest end of the curve).

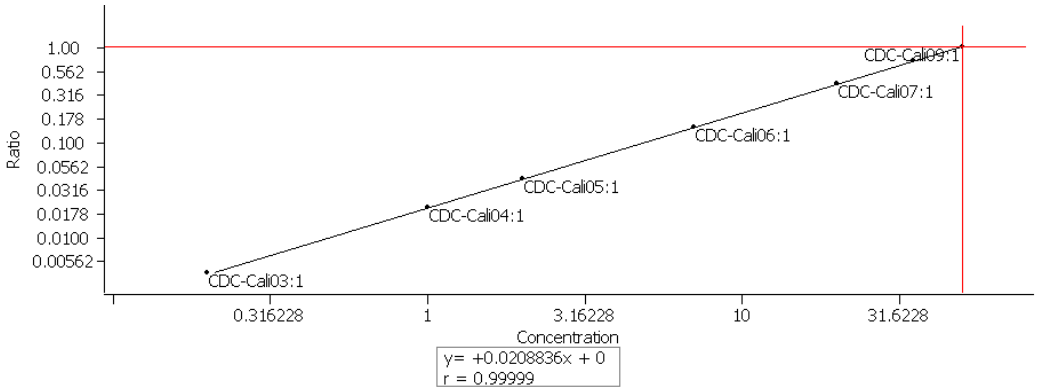

Fig. 4: Calibration curve of 2,3,7,8-TCDD by GCxGC-IDTOFMS.

Excellent correlation coefficients were calculated for all the PCDD/Fs and PCBs. Because the GC conditions have been optimized for the resolution of all analytes in the same run, the PCDD/F fraction and the PCB fraction issued from the clean-up step can be either injected separately or recombined prior injection. This results in a single injection for both fraction and a reduction of the analytical time. The method is being evaluated for the analysis of various food types and compared to reference HRMS method. Such a comparison was recently shown to be successful for higher level matrices [7]. Figure 5 shows some of the preliminary data obtained for the measurement of the WHO PCBs in a beef fat QC pool. Rather good correlation is observed although a significant difference was recorded for CB-189. This is currently investigated and more statistical data are being produced. Recovery rates ranged between 60 and 90% and 5g of QC samples were processed.

ACKNOLEDGEMENT
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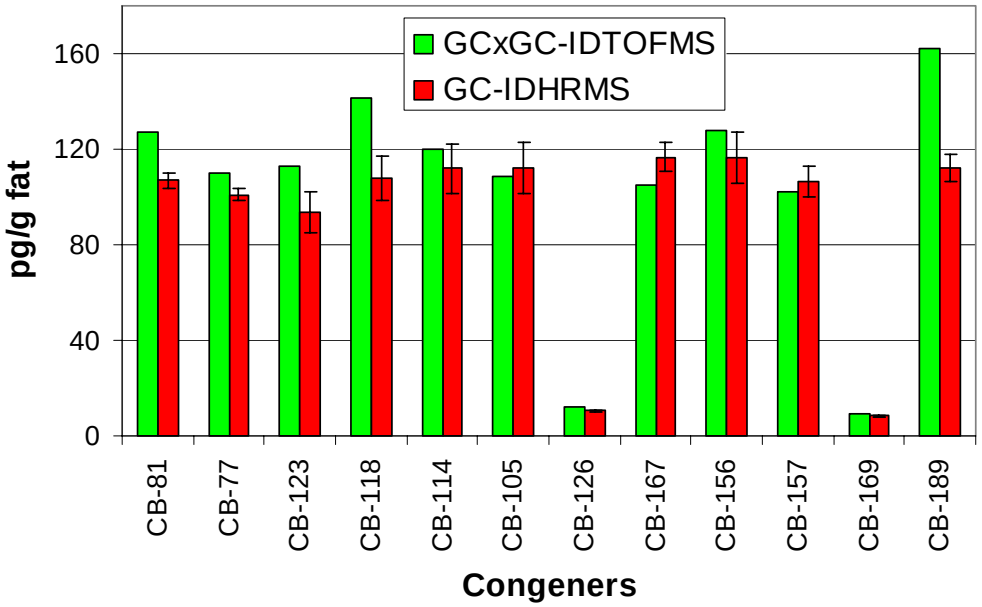

Fig. 5: Comparison between GCxGC-IDTOFMS and GC-IDHRMS measurement of WHO PCB levels in beef fat QC samples.

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