APPLICATION OF ACCELERATED SOLVENT EXTRACTION (ASE) AND SOLVENT CUT LARGE VOLUME (SCLV) INJECTION SYSTEM FOR DETERMINATION OF DIOXINS IN FOODS

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Introduction

Most laboratories have difficulty monitoring dioxins in foods, as a large volume of sample (generally 100g) must be treated collectively in order to attain the desirable limit of detection (LOD) at low ppt levels. The conventional method, including liquid partitioning with a separatory funnel, demands much labor and time. In order to establish small-scale extraction, it is necessary to improve the sensitivity of HRGC/HRMS systems. On the other hand, semi-automated extraction is expected to improve the efficiency of laboratory performance. In the present study we examine the applicability of an accelerated solvent extraction (ASE) and solvent cut large volume (SCLV) injection system to the isomer-specific determination of dioxins in foods. Spinach is used as a representative plant food.

Materials and Methods

Conventional and tentative analytical procedures are summarized in Table 1. Accelerated extraction was performed by ASE-300 (Dionex, USA) under the conditions of 1500psi, 150°C. Extracts of dioxins were prepared from homogenates of spinach that was purchased at a market in Japan. Analyses were performed using an HP-6890 Plus gas chromatograph (Hewlett-Packard, USA) coupled to an Autospec-Ultima mass spectrometer (Micromass, UK). We employed an Rtx-2330 (0.18mm x 40m) capillary column (Restek, USA) on the SCLV injection system (SGE, Australia) to determine tetra- and pentaCDD/DFs. The details of the operating conditions for the SCLV injection system are described in another paper[1]. The LOD for each congener in the present study was decided according to the guidelines for food analysis of dioxins issued by the Ministry of Health and Welfare of Japan in 1999 ('Guideline'): An absolute quantity corresponding to S/N = 3 is evaluated on HRGC/HRMS using verification standards.

Results and Discussion

The SCLV injection system was developed to detect dioxins on the order of low femtograms in human blood[2]. In order to apply the system to food analysis, the applicability of a cyanopropyl-phase capillary column on the system was examined. Extracts from 100g of spinach by conventional liquid partitioning were analyzed under the ordinary GC conditions and under the SCLV injection technique. The chromatograms obtained from these different methods are shown in Figure 1. It was found that both chromatograms were extremely similar in appearance, and that peak separations around 2,3,7,8-substituted isomers compared well with the conventional injection method. Data regarding quantification of 2,3,7,8-substituted isomers are shown in Table 2. The ratios of estimated concentrations from the SCLV system to those from the conventional method ranged from 91 to 118%. We could not demonstrate the quantification of 2,3,7,8-tetraCDD using spinach because it was not

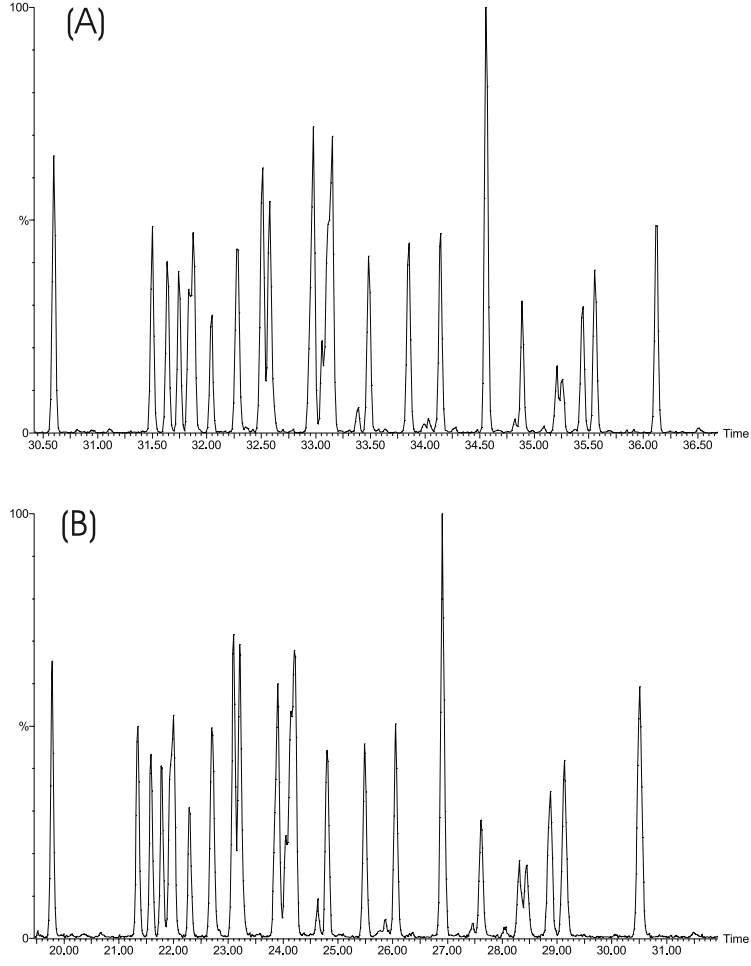

Figure 1. HRGC/HRMS chromatogram of a spinach extract (tetraCDFs). (A) SCLV injection system coupling to an Rtx-2330 column (B) Conventional splitless injection system coupling to an SP-2331 column

detected (< 0.01pg/g) in either sample. However, the fine quantification of 2,3,7,8-tetraCDD was observed in the SCLV system when a flue gas sample was applied[1]. Considering the above results, it is considered that the SCLV injection system coupled to an Rtx-2330 column is useful for the determination of dioxin isomers in food. On the other hand, the SCLV system has at least five times higher sensitivity than the conventional injection technique, a result of multiple analyses of calibration verification standards. Accordingly, it is concluded that sample volume could be commonly reduced from 100 g to 20 g (from 50 g to 10g in case of fish) when the SCLV injection system is applied to food analysis.

ASE has already been applied to the extraction of dioxins from environmental and tissue samples. Its high efficiency has been demonstrated, and the data it provides are equivalent to those provided by the conventional extraction method[3, 4]. We applied a combination of ASE and the SCLV injection

Table 1. Analytical procedures for determination of dioxins in food.

		Conventional method	Tentative method
Extraction		Liquid partitioning Sample weight: 100g Solvent: acetone/n-hexane (1:1, v/v)	ASE Sample weight: 20g Solvent: acetone/n-hexane (1:1, v/v)
Clean-up		Sulfuric acid treatment ↓ Silver nitrate-impregnated silica gel column ↓ Active carbon-dispersed silica gel column	
HRGC/ HRMS analysis	PCDD/DFs and non-ortho PCBs	Splitless injection Injection volume: 1-2 μL / 20μL Combination of columns: SP-2331(0.32mm x 60m) BPX-5 (0.25mm x 60m)	SCLV injection Injection volume: 4 μL / 20μL Combination of columns: Rtx-2330 (0.18mm x 40m) BPX-5 (0.15mm x 30m)
	Mono-ortho PCBs	Splitless injection Injection volume: 1μL/20 μL Capilaly column: HT-8 (0.22mm x 50m)	

Table 2. Comparison of concentrations of tetra- and pentaCDD/Fs determined by ordinary injection and SCLV method.

	LOD (pg/g)	Concentraton ratio (%)*	
		Spinach-1	Spinach-2
2,3,7,8-tetraCDD**	0.01	-	-
1,2,3,7,8-pentaCDD	0.02	93.7	118.0
2,3,7,8-tetraCDF	0.01	90.9	97.1
1,2,3,7,8-pentaCDF	0.02	108.6	99.4
2,3,4,7,8-pentaCDF	0.02	98.7	97.4

Extracts from 100g of spinach were prepared by conventional liquid partitioning.
*Presented by the ratio of concentration from SCLV to that from ordinary injection.
**It is not detected in both samples.

technique for the determination of dioxins in spinach. Our analysis of 20 g of spinach according to tentative procedures, including the ASE and SCLV injection technique, showed recovery rates for 29 kinds of isomers ranging from 44.9 to 103 %, within the range recommended by the Guideline (40-120 %). On the other hand, equivalency of the data to those obtained from the conventional method was not confirmed: Concentrations derived from the tentative method were generally lower than those from the conventional method (data not shown). This is probably due to some inhomogeneities in the stocked spinach sample rather than to some difference in the extraction efficacy between the methods.

Therefore, exhaustive homogeneous tissue (for example, dried milk) must be examined in order to evaluate the equivalency of the methods. Our results suggest that the SCLV injection technique is applicable to dioxin analysis in food and provides good performance in the laboratory, especially when it is coupled to an automated extraction method.

References

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