



Levels of PCDD/F in adipose tissue on non-occupationally exposed subjects living near a hazardous waste incinerator in Catalonia, Spain

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ARTICLE INFO

Article history:

Received 12 September 2008

Received in revised form 18 November 2008

Accepted 19 November 2008

Available online 17 January 2009

Keywords:

Hazardous waste incinerator
Tarragona County (Catalonia, Spain)
Polychlorinated dibenzo-*p*-dioxins and furans (PCDD/F)
Adipose tissue
Non-occupationally exposed subjects

ABSTRACT

The potential adverse effects on human health of hazardous waste incinerators (HWIs) are a subject of concern. The construction of the first and till now only HWI in Spain finished in 1999. In 2007, after approximately 9 years of regular operations in the facility, the concentrations of polychlorinated dibenzo-*p*-dioxins and dibenzofurans (PCDD/F) were determined by HRGC/HRMS in samples of adipose tissue of 15 autopsied subjects living in the area under potential impact of the HWI (Constantí, Tarragona County, Catalonia). These data were compared with the levels found in two previous surveys performed in 1998 (baseline) and 2002. The current mean PCDD/F concentration in adipose tissue was 14.6 pg WHO-TEQ/g of fat (range: 3.3–55.4 pg WHO-TEQ/g of fat). It means significant changes (a reduction of 64% and an increase of 47%) in the mean PCDD/F levels in comparison to those found in 1998 and 2002: 40.1 and 9.9 pg WHO-TEQ/g of fat, respectively. Women showed higher levels of PCDD/F than men (23.8 vs 11.2 pg WHO-TEQ/g of fat). The increase in the last 5 years would not be directly attributable to exposure to PCDD/F emitted by the HWI, as other biological monitors such as human plasma and milk decreased during the same period.

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1. Introduction

Polychlorinated dibenzo-*p*-dioxins and dibenzofurans (PCDD/F) are semi-volatile substances and undergo long-range transport, which are generated as unintentional by products in a number of thermal and chemical-industrial processes (Fiedler, 2006, 2007; Kulkarni et al., 2008). Until recent years, waste incinerators (municipal, hazardous and medical) have been among the most important emission sources of PCDD/F (Glorennec et al., 2005; Kim et al., 2008; UNEP, 2008). These compounds are persistent and accumulate in the environment and organisms, some congeners such as the 2,3,7,8-TCDD being highly toxic for humans (Kogevinas, 2001; Steenland et al., 2004; Schecter et al., 2006).

In 1999, the first, and up to now only, hazardous waste incinerator (HWI) in Spain began to operate in Constantí (Tarragona County, Catalonia). It is well known that these facilities can emit a considerable number of pollutants including PCDD/F, which is a cause of considerable concern for the public opinion. For this reason, three years before the HWI initiated regular operations, a wide surveillance program, consisting on biological and environmental monitoring studies, was initiated. This program was mainly focused on determining the human health risks derived from the emissions of metals and PCDD/F (Ferré-Huguet et al., 2006). In the baseline

study (1996–1998), samples of plasma, milk, and adipose tissue were collected from individuals living in the vicinity of the facility (Schuhmacher et al., 1999a–c). In 2002, after approximately 3 years of regular operations, a new survey was again carried out using samples of the same biological monitors. Notable reductions in the levels of PCDD/F in plasma, milk, and adipose tissue were noted (Schuhmacher et al., 2004a,b; Agramunt et al., 2005). This decline was hypothesized to be mainly due to a reduction of the dietary intake of PCDD/F (Bocio and Domingo, 2005). It was concluded that PCDD/F emissions from the HWI were not being the main source of exposure to these pollutants for the population of the zone.

Recently, and continuing with this specific surveillance program, a new survey focused on determining the temporal trend in the concentrations of PCDD/F in the same biological tissues was performed. The PCDD/F concentrations in samples of human adipose tissue collected in 2007 are presented here. The results are compared with the levels obtained in the baseline (1998) and 2002 surveys, as well as with data of recent studies from different countries.

2. Materials and methods

2.1. Sampling

Information about the HWI, as well as the characteristics of the area in which it is placed were previously described (Schuhmacher

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et al., 1999a–c). In 2007, adipose tissue samples of 15 autopsied subjects living in zones of Tarragona County (Catalonia, Spain) under potential impact of the HWI were collected. At the time of death, all the individuals had lived for at least 10 years in the area under evaluation. All samples were obtained from the same body compartment (abdominal adipose tissue). Samples were classified according to sex (11 men and 4 women) and specific residential zone, urban or industrial. Nine subjects had lived in urban zones (Tarragona downtown), while the remaining individuals had been residing near an industrial area (chemical and petrochemical facilities, and a municipal solid waste incinerator). The mean age of the subjects was 52 years, ranging from 28 to 83. Individual and medical information of the participants were collected by passing a questionnaire to the relatives. According to that, no occupational exposure to PCDD/F was found for any of the subjects.

2.2. Analytical procedure

PCDD/F analysis was done according to the US EPA method 8290. Samples were dissolved in hexane, and a mixture of $^{13}\text{C}_{12}$ -PCDD/F standards was spiked in order to control potential losses during the extraction and clean-up processes. Lipids were destructed by adding acid silica. In order to remove interfering components, a multi-step clean-up was performed. The first step consisted on a multilayer silica column clean-up. Further, the extract was eluted on a basic alumina column by passing different solvents in order to separate PCDD/F from other compounds. The PCDD/F fraction was collected and concentrated to near dryness with a nitrogen flux. Finally, 25 μL of $^{13}\text{C}_{12}$ -PCDD/F injection standards were added.

Analysis of PCDD/F was carried out by high resolution gas chromatography/high resolution mass spectrometry (HRGC/HRMS). The extract obtained after extraction and clean-up was injected on an Agilent 6890 gas chromatograph equipped with a ZB5-MS capillary column and coupled to a Waters Autospec Ultima mass spectrometer. The chromatographic process separated the 17 toxic 2,3,7,8-substituted congeners from each other. The mass spectrometer measured, via “selected ion recording” at a resolution of >10000, two selected ions per congener group for both the native and labeled compounds. The calculation of the concentrations was done by using the corresponding C13 congener level, automatically correcting according to the recovery percentage specific for each congener (mean: 62.2%). In addition, the relative standard deviation (RSD) was also calculated as a measure of the uncertainty. In all cases, the RSD of the control sample was lower than 10%.

2.3. Data treatment

The SPSS 15.0 statistical software package was used for data analysis. The total PCDD/F concentrations were calculated according to the 2005 WHO-TEFs (van den Berg et al., 2006). The levels of our previous surveys (baseline and 2002) were again recalculated by using these factors. The Levene test was applied to study the equality of variances. Furthermore, the ANOVA or Kruskal–Wallis tests were executed. A probability of 0.05 or lower ($p < 0.05$) was considered as statistically significant. For calculations, when a PCDD/F congener presented a level below the respective limit of detection (LOD), the concentration was assumed to be one-half of that limit ($\text{ND} = 1/2 \text{ LOD}$).

3. Results and discussion

The individual PCDD/F concentrations of the 15 adipose tissue samples corresponding to individuals who had lived in Tarragona County are summarized in Table 1. Personal informations such as

Table 1

Individual PCDD/F concentrations (pg/g fat) in adipose tissue samples of individuals of Tarragona County.

Sample	Area	Age (years)	Sex	WHO-TEQ
1	Urban	75	M	23.8
2	Urban	35	F	7.5
3	Urban	28	M	6.2
4	Urban	83	M	27.7
5	Industrial	29	M	3.4
6	Urban	29	M	5.9
7	Urban	60	M	17.4
8	Urban	50	M	7.5
9	Urban	45	M	7.3
10	Industrial	70	F	28.9
11	Industrial	77	F	55.4
12	Industrial	61	M	11.4
13	Industrial	53	M	5.7
14	Urban	72	M	7.1
15	Industrial	35	F	3.3

M: male; F: female.

residential zone, age, and sex are also given. The mean PCDD/F concentration was 14.6 pg WHO-TEQ/g fat, with minimum and maximum values of 3.3 and 55.4 pg WHO-TEQ/g fat. The highest concentrations corresponded to two sisters (77 and 70 years old) residing in a small village located in the industrial area.

The mean levels of the PCDD/F congeners, as well as the total concentration of the 15 adipose tissue samples collected in the baseline (1998), previous (2002) and current (2007) surveys are shown in Table 2. In the baseline (1998) study, a mean PCDD/F concentration in adipose tissue of 40.1 pg WHO-TEQ/g fat (range: 24.2–72.1 pg WHO-TEQ/g fat) was found (Schuhmacher et al., 1999c), while in the 2002 survey, a mean PCDD/F level of 9.9 pg WHO-TEQ/g fat (range: 1.4–36.1 pg WHO-TEQ/g fat) was observed (Schuhmacher et al., 2004a). In the period 2002–2007, a non-significant increase of 47% in the mean PCDD/F concentrations was noted. OCDD and 1,2,3,7,8-PeCDF were the individual congeners showing the most remarkable increase (183% and 181%, respectively). However, if the median PCDD/F concentrations are compared (39.6, 6.8 and 7.5 pg WHO-TEQ/g fat in 1998, 2002 and 2007, respectively), the increase between the second and third surveys was notably lower, 10.3%. On the other hand, the reduction (64%) between the baseline (1998) and current (2007) studies, was still statistically significant ($p < 0.001$). Age may be an important confounding factor when determining the human accumulation of PCDD/F (Kiviranta et al., 2005). In order to establish its influence on the TEQ values, the age of the participants was studied for each survey. Mean ages of 60 (range: 28–83), 58 (range: 19–94) and 53 (range: 28–83) years were found in 1998, 2002 and 2007 studies, respectively. Therefore, in principle, a decrease in the mean age could derive in a diminution of the PCDD/F levels in the human body. Notwithstanding, a slight increase of these TEQ values were observed. The box plot of the PCDD/F concentrations in adipose tissue found in 1998, 2002 and 2007, according to age subgroups, is depicted in Fig. 1. In the two last surveys, a clear increase of the PCDD/F levels was appreciated for the older people. The PCDD/F congener profiles obtained in the three surveys are presented in Fig. 2. All samples showed considerably higher levels of PCDDs than PCDFs. As in our previous studies (Schuhmacher et al., 1999c, 2004a), OCDD was the predominant congener, followed by 1,2,3,6,7,8-HxCDD and 1,2,3,4,6,7,8-HpCDD.

PCDD/F concentrations in adipose tissue, classified according to sex and specific place of residence (urban or industrial) of the individuals, are given in Table 3. Women showed remarkable higher levels of PCDD/F than men (23.8 vs 11.2 pg WHO-TEQ/g fat). However, the difference did not reach the level of statistical significance. The temporal trend indicated a notable (but not significant) increase of PCDD/F levels between 2002 and 2007, in both males

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