Occupational exposure to polychlorinated dibenzo-p-dioxins and dibenzofurans in a magnesium production plant

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Abstract

Objectives—The production of magnesium is a well known source of both aliphatic and aromatic chlorinated compounds, among others the polychlorinated dibenzo-p-dioxins (PCDDs) and polychlorinated dibenzofurans (PCDFs). The aim of this study was to establish whether increased concentrations of PCDDs and PCDFs could be found in the blood of workers in a magnesium plant.

Methods—Blood plasma from 10 workers, employed at a magnesium plant for 10 to 36 years, and from a control group consisting of nine people who had no direct contact with the production were studied. Isomer specific analyses of PCDDs and PCDFs by means of high resolution gas chromatography/high resolution mass spectrometry (HRGC/HRMS) techniques were performed.

Results—A significant increase was found in the concentrations of some of the congeners, mainly PCDFs, in the workers compared with the control group. Octachlorodibenzofuran (OCDF) is the congener that most strongly correlates with occupational exposure in the magnesium plant. Low concentrations of 1,2,3,4,6,8,9-heptachlorodibenzofuran were found in seven of the workers. Such isomers—that is, not 2,3,7,8-substituted—are rarely found in human samples.

Conclusion—Due to the long biological half lives and lipid solubility of PCDDs and PCDFs, blood analyses may serve as an index of past cumulative occupational exposure and a means of assessing a person's exposure situation.

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Environmental and human exposure to polychlorinated dibenzo-p-dioxins (PCDDs) and polychlorinated dibenzofurans (PCDFs) have been the focus of much attention in recent years. These compounds are persistent and highly lipophilic, they bioaccumulate in the food chain and have been shown to cause a variety of biological and toxic responses including immunological and hepatic toxicity, carcinogenic, and teratogenic effects.^{1,2} Humans are exposed through the intake of food such as fish, meat, and dairy products, resulting in a general background of PCDDs and PCDFs that is fairly similar throughout the industrialised world. Higher concentra-

tions of 2,3,4,7,8-pentachlorodibenzofuran are found in Europe than in the United States.³ Human concentrations of PCDDs and PCDFs and major exposure routes are summarised in a review by Ryan and Norstrom.⁴ People exposed accidentally or occupationally often display a different congener pattern than those exposed indirectly through the environment and such exposure can be identified through the analysis of PCDDs and PCDFs in blood. Several occupational studies are focused on exposure to 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD), a congener associated with the production and use of phenoxy herbicides. For example, high concentrations of TCDD among chemical workers involved in the production of 2,4,5-trichlorophenol have been reported by Patterson *et al.*⁵ In a Swedish study of production workers who had been employed in a factory producing chlorophenoxy herbicides and chlorophenols, increased concentrations of TCDD and 1,2,3,7,8-pentachlorodibenzo-p-dioxin were found 16 to 21 years after the workers had left their employment.⁶ Furthermore a study presented by Päpke *et al.*⁷ showed that various kinds of occupational exposure to PCDDs and PCDFs—for example, in the production of tri and pentachlorophenol and at a metal reclamation plant—resulted in a characteristic congener pattern in blood.

Significant amounts of PCDDs and PCDFs are formed as undesired byproducts in various industrial and high temperature processes.⁸ Magnesium production involves several different steps and it is in the chlorination and electrolytic processes that different organic chlorocompounds are formed—for example, considerable amounts of PCDDs and PCDFs.⁹ These compounds have been detected in effluents and exhausts from a magnesium plant at Porsgrunn in the southern part of Norway, as well as in sediment samples and marine biota collected in the vicinity of the plant. The yearly emission to the waste water from the plant has been estimated at 0.3–0.5 kg TCDD equivalents^{10–12} calculated according to the Nordic model,² and the concentration ratio of Σ PCDF/ Σ PCDD is about 10:1. In a study of human milk from Scandinavia, the concentration of PCDFs was found to be slightly higher in the Skien-Porsgrunn area, where the magnesium plant is located, than in other regions in Norway.¹³ As well as organic chlorocompounds, the production of magnesium involves combined occupational exposure to many other potentially harmful substances such as asbestos (before 1980), chlorine gas, and hydrochloric

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aerosols. Strong magnetic fields are also present in the work environment. A higher incidence of different types of cancers in a cohort of magnesium production workers was found in a study by Heldas *et al.*¹⁴ in which the relation between exposure to factors in the working environment and the development of cancer is discussed. Since 1990, there has been a considerable reduction in emissions from the plant. The primary aim of this study was to compare the concentrations of PCDDs and PCDFs in the blood lipids of workers in the magnesium plant with those found in controls who had no connection with the production. Moreover, the study was thought to be of importance to ongoing efforts to improve working conditions at the plant.

Method

SUBJECTS

Hexachlorobenzene (HCB) is formed under conditions similar to those under which PCDDs and PCDFs are formed, and is present in emissions from the plant and in the working environment. The health department at the plant has monitored the concentrations of HCB in blood samples from the workers since the mid-1970s. Although HCB has a much shorter biological half life than PCDDs and PCDFs, it was considered a possible indicator of occupational exposure to organic chlorocompounds. The workers with the highest concentrations of hexachlorobenzene (between 21 to 52 ng/g whole blood) were chosen for this study. They were working either in the electrolytic or the chlorination sector of the plant. Several of the workers had experienced episodes of exposure to chlorine gas resulting in symptoms ranging from minor discomfort to respiratory impairment requiring acute medical treatment. Ten workers, employed for 10 to 36 years, and a control group consisting of nine people working at the same plant were studied. As far as dioxin exposure is concerned, the only difference between the control group and the workers is the fact that the workers are directly involved in the production process. There were two women in each group. The mean age at the time of the study was 48 in the group of work-

ers and 38 in the control group. A general medical examination of all subjects did not show any clinical signs indicative of exposure to polychlorinated substances.

Blood samples were taken from the 19 people, who had been on a low fat diet for 12 hours. Standard hospital procedure was used in the separation of the blood plasma. The plasma bags were immediately frozen and stored at -20°C . All samples were coded before they were shipped to the analysing laboratory and were analysed blind. The samples were taken in late 1988 and analysed in early 1989.

ANALYTICAL METHOD

The analytical method for the determination of trace concentrations of PCDDs and PCDFs is described elsewhere.^{15,16} In short, the procedure involves extraction by means of organic solvents, cleaning up with various adsorbents, and a gravimetric determination of the lipid content. After cleaning up, the extracts were analysed by means of high resolution gas chromatography/high resolution mass spectrometry (HRGC/HRMS) techniques. The mass spectrometric data was obtained by means of a VG-250S high resolution MS operating in electron impact mode. In a quality control study between laboratories organised by the World Health Organisation, in which the concentration range was the same as that of the present study, this analytical method yielded an average coefficient of variation for reproducibility and repeatability of 6.7%.¹⁷

STATISTICAL ANALYSIS

Principal component analysis (PCA), described in detail by Wold *et al.*,¹⁸ is a projection method in which the included variables are combined into a few underlying descriptive dimensions, which makes it possible to study the systematic variation present in a data matrix. The PCA gives an overview of the dominating patterns and major trends in the data. The analysis results in two complementary plots, a score plot and a loading plot. The relation between the objects (samples) is shown on the score plot. The loading plot shows the extent to which each variable contributes to the object separation. Variables located far from zero have a large influence. Cross validation¹⁹ was used to determine the number of significant principal components.

Differences between the two groups were also tested with non-parametric statistics (Mann-Whitney *U* test).

Results and discussion

Table 1 shows the results of the analysis for PCDDs and PCDFs in blood plasma for nine of the workers (one worker was excluded for reasons accounted for later) and the control group, where the abbreviation T denotes tetra, Pe penta, Hx hexa, Hp hepta, and O octa. Concentrations of PCDDs and PCDFs are expressed in pg/g lipid extracted from the blood plasma. Also included are TCDD equivalents calculated according to the Nordic model,² a model for risk assessment of com-

Table 1 Mean concentrations of PCDFs and PCDDs, expressed in pg/g lipid, in blood plasma from workers and controls

	Variable No	Controls (n = 9) mean (range)	Workers (n = 9) mean (range)	P* value
PCDFs:				
2,3,7,8-TCDF	1	2.9 (1.2-4.7)	3.5 (1.5-6.1)	0.5
1,2,3,7,8-PeCDF	2	2.2 (<1-30)	5.4 (<1-11)	0.06
2,3,4,7,8-PeCDF	3	20 (14-29)	51 (19-170)	0.02
1,2,3,4,7,8-HxCDF	4	22 (5.9-94)	59 (23-150)	0.01
1,2,3,6,7,8-HxCDF	5	12 (5.7-31)	59 (19-190)	0.002
2,3,4,6,7,8-HxCDF	6	3.4 (<1-6.8)	5.4 (2-9.6)	0.09
1,2,3,4,6,7,8-HpCDF	7	14 (4.7-32)	85 (21-150)	0.0007
1,2,3,4,7,8,9-HpCDF	8	<2	6.9 (2-16)	0.0005
OCDF	9	7.5 (<3-18)	216 (17-560)	0.0005
PCDDs:				
2,3,7,8-TCDD	10	3.9 (2.8-6.3)	8.2 (2.5-35)	0.1
1,2,3,7,8-PeCDD	11	7.2 (5.0-13)	16 (6.7-51)	0.01
1,2,3,4,7,8-HxCDD	12	2.7 (<1-4.6)	5.6 (2.7-10)	0.01
1,2,3,6,7,8-HxCDD	13	20 (11-32)	32 (17-74)	0.04
1,2,3,7,8,9-HxCDD	14	4.2 (2.1-5.9)	7.3 (3.3-14)	0.02
1,2,3,4,6,7,8-HpCDD	15	51 (26-113)	46 (29-75)	0.9
OCDD	16	372 (150-700)	341 (200-530)	0.7
TCDD equivalents		25 (17-42)	60 (27-190)	0.007

*Mann-Whitney *U* test for comparison between groups (two tailed).

plex mixtures of PCDDs and PCDFs.

The workers' concentrations of chlorinated PCDFs, primarily the Hx, Hp, and O chloro substituted PCDFs, are clearly raised, compared with the control group. The concentrations of some of the PCDD congeners are also increased, except for HpCDD and OCDD, and the Mann-Whitney U test shows a significant difference in TCDD equivalents between the two groups.

An unexpected result of the analysis was that low concentrations (4–17 pg/g lipid) of 1,2,3,4,6,8,9-HpCDF were found in seven of the 10 workers. Isomers that are not 2,3,7,8-substituted are hardly ever detected in human samples.⁴

The mean sum of PCDFs for the workers and the control group was 583 and 85 pg/g lipid, respectively. There is a relation between the number of years in the production and concentrations of PCDFs. Figure 1 shows the ratio of Σ PCDFs/ Σ PCDDs plotted against years of employment at the plant. The ratio eliminates any age related increases in the concentrations of the congeners.²⁰ The correlation coefficient (r^2) was calculated to be 0.84. The mean ratio of Σ PCDFs/ Σ PCDDs of the workers was 1.1 but the control group had a mean ratio of 0.21. In samples from people with no known occupational or accidental exposure, overall PCDD concentrations are normally much higher than PCDF concentrations.

The concentrations of the 16 detected 2,3,7,8-substituted congeners in the blood samples were used as variables in the PCA. They were logarithmically transformed (table 1). Two principal components were significant and together they account for 82% (61 + 21) of the variance in the data set. As shown in the score plot (fig 2(A)), where the first principal component is plotted against the second, the workers and the controls form separate groups. The corresponding loading plot (fig

2(B)) shows that all congeners, except 1,2,3,4,6,7,8-HpCDD (variable number 15) and OCDD (16) which are located close to zero, were to some extent active in this separation. The chlorinated dibenzofurans, 1,2,3,4,7,8 and 1,2,3,6,7,8-HxCDF (4 and 5), 1,2,3,4,6,7,8-HpCDF (7) and particularly OCDF (9) contribute the most strongly to the separation of the groups. These isomers also account for the most significant difference between the two groups according to the Mann-Whitney U test (table 1). The OCDF is a minor component (often not detected) in human samples. In this group of workers, the concentration of OCDF in the blood plasma was in the range of 17–560, with a mean of 216 pg/g lipid. These are unusually high concentrations in human samples and OCDF is undoubtedly the congener that most strongly correlates to occupational exposure in the magnesium plant. Analyses of indoor air from the plant show that OCDF accounts for 10%–40% of the total amount of PCDDs and PCDFs calculated as TCDD equivalents.

Studies have shown that crabs in this area contain very high concentrations of PCDFs and PCDDs¹⁰ and there are restrictions on the consumption of seafood from this area. The worker A in the score plot, declared a very high consumption of crabs (50 crabs a year) from the coastal area near the magnesium plant. As we were not sure to what extent this high consumption of crabs reflects the concentrations of PCDFs and PCDDs in this person's blood plasma, he was excluded from table 1. Worker A has been with the company for 22 years and has primarily been doing maintenance work in the electrolytic sector. Table 2 shows the concentrations of PCDFs and PCDDs for worker A and an additional person who also eats a lot of crabs from this area (a declared intake of 40 crabs a year) but who has no connection at all with the magnesium plant. The blood sample from this crab consumer was analysed in the same manner as the other samples but at a later time. His concentrations of several of the congeners are higher than the mean concentrations of both the workers and the control group, resulting in 101 TCDD equivalents in his blood fat. This indicates that a high consumption of crabs from this area may cause increased concentrations of PCDFs and PCDDs. The ratio of Σ PCDFs/ Σ PCDDs was 1.6 for worker A and 0.4 for the crab consumer. The higher ratio of worker A is likely to be caused by work related

Figure 1 Years of employment plotted v the ratio of Σ PCDFs/ Σ PCDDs.

Figure 2(A) The score plot of principal component 1 v principal component 2 from the 19 objects analysed. (B). Loading plot for the variables listed in table 1. The first loading vector is plotted v the second.

Table 2 Concentrations of PCDFs and PCDDs (pg/g lipid) in blood plasma from worker A and a crab consumer

	Worker A	Crab consumer
Crabs consumed (n/y)	50	40
PCDFs:		
2,3,7,8-TCDF	11	2.7
1,2,3,7,8-PeCDF	30	6.5
2,3,4,7,8-PeCDF	440	94
1,2,3,4,7,8-HxCDF	400	99
1,2,3,6,7,8-HxCDF	400	84
2,3,4,6,7,8-HxCDF	20	11
1,2,3,4,6,7,8-HpCDF	95	43
1,2,3,4,7,8,9-HpCDF	7	<2
OCDF	14	<3
PCDDs:		
2,3,7,8-TCDD	65	11
1,2,3,7,8-PeCDD	110	29
1,2,3,4,7,8-HxCDD	26	13
1,2,3,6,7,8-HxCDD	110	51
1,2,3,7,8,9-HxCDD	26	6.0
1,2,3,4,6,7,8-HpCDD	45	59
OCDD	480	620
TCDD equivalents	442	101

exposure to PCDFs. It is not possible to establish whether worker A's overall increased concentrations (442 TCDD equivalents) were due to the working environment or to a high consumption of crabs as our numbers were too small for any conclusions to be drawn. It is likely that the high concentrations of PCDFs and PCDDs in the blood lipids are attributable to both of these factors. The consumption of contaminated marine biota is believed to play a substantial part in human concentrations of PCDDs and PCDFs, as has been shown by Svensson *et al.*²¹ They found significantly higher concentrations of PCDDs and PCDFs in men with a high intake of fish from the Baltic Sea than in men who consumed less or no fish. An ongoing study of consumers of crabs from the Norwegian coast, performed by the National Institute of Public Health in Norway, will hopefully provide further information.

In two other studies of occupational exposure to PCDDs and PCDFs in a metal reclamation plant⁷ and a municipal waste incinerator,²² higher blood concentrations of PCDFs were found in the workers compared with control groups. In both cases the Pe, Hx, and HpCDF concentrations were raised but the high concentrations of OCDF found in the workers in the magnesium plant were not detected.

Conclusion

Blood samples provide an index of past cumulative exposure to PCDDs and PCDFs, due to the long biological half lives and lipid solubility of these compounds. People exposed accidentally or occupationally often have a different congener pattern than those exposed indirectly through the environment and such exposure can be identified through the analysis of PCDDs and PCDFs in blood.

This report presents findings of increased concentrations of PCDFs and PCDDs in the blood plasma from workers in a magnesium production plant. The dominating congeners in these workers, compared with the control group, are 1,2,3,4,7,8- and 1,2,3,6,7,8-HxCDF, 1,2,3,4,6,7,8-HpCDF and, in particular, OCDF. A non-2,3,7,8-substituted congener, 1,2,3,4,6,8,9-HpCDF, was also

identified in seven of the 10 workers. A comparison of the ratio of Σ PCDFs/ Σ PCDDs shows that the predominance of the Σ PCDFs over Σ PCDDs correlates with years of employment in the magnesium plant.

The present study shows that a combination of isomer specific analysis and principal component analysis provides a means of characterising different kinds of occupational exposure.

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