Cadmium and mercury exposure over time in children in southern Sweden (1986 – 2013)

Report to the Swedish Environmental Protection Agency

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Summary

The variation in exposure to toxic metals over time is still not well-characterized, especially in children, the most vulnerable group. The aim of this study was to evaluate whether a reduction in environmental pollution with cadmium (Cd) and mercury (Hg) caused a change in exposure over time in two Swedish cities.

For this, 1,257 children between the ages of 4 and 9 from Landskrona and Trelleborg, in the south of Sweden, were sampled once during 1986-2013. Cd (n=1120) and Hg (n=560) concentrations were determined in blood (b-Cd, b-Hg). The median b-Cd was 0.10 (geometric mean 0.10; range 0.01-0.61) μg/L and b-Hg was 0.91 (geometric mean 0.83; range 0.02-8.2) μg/L.

No sex or age difference was seen in b-Cd or b-Hg. Regarding the cities, children living in Landskrona close to a smelter had higher b-Cd and b-Hg than those living in urban and rural areas. Over the studied time, b-Cd slightly decreased (0.7% per year, p<0.001) while b-Hg showed a marked decrease (3% per year, p<0.001).

In conclusion, exposure to Cd was low during the ages 4 to 9, but the risk of disease might increase later in life since b-Cd only showed a minor decrease over time. This indicates, that Cd pollution should be further restricted. Regarding Hg, b-Hg concentration in children was low and decreased over time. The decrease is probably due to the reduced use of dental amalgam and the lower Hg intake from fish.

Background

Assessing risk and prioritizing preventive actions towards toxic metals require information on the levels of exposure and their time trends, such information is also useful to assess how effective preventive actions are. While metals like lead have been largely monitored in several countries (Skerfving and Bergdahl, 2014), limited information on exposure over time is available for cadmium (Cd) and mercury (Hg), especially for children.

Cd is a ubiquitous element and its exposure mainly occurs through food and smoking in adults (Nordberg et al., 2014). Cd may cause toxic effects on the proximal tubuli of the kidney (Åkesson et al., 2005; Suwazuno et al., 2006) and skeleton (Åkesson et al., 2006 and 2014), even at exposure levels that are present in the general population, and even in areas, such as Sweden, that have low environmental exposure, compared with most other sites worldwide (WHO, 2007). Women accumulate more Cd than men, making them more vulnerable to these effects. Furthermore, low to moderate levels of exposure to Cd during pregnancy may impair child growth (Kippler et al., 2012a; Johnston et al., 2014), kidney development (Hawkesworth et al., 2013), and cognitive function during early childhood (Ciesieski et al., 2012; Kippler et al., 2012b).

The Cd concentration in blood (b-Cd) provides a combined index of recent exposure and the body burden. The body burden of Cd has extremely slow turnover, which attenuates any change in Cd uptake. Thus, the blood concentrations in adults may, to a large extent, reflect exposure from decades ago (Welinder et al., 1977; Liu et al., 2001). Hence, b-Cd in children is likely to be a valid biomarker for monitoring trends in exposure over time. Still, there is almost no information on time trends in children (Schulz et al., 2007), especially young ones, partly because their b-Cd levels are extremely low and earlier studies primarily focused on health effects in adults.

Hg is also a ubiquitous element and its exposure mainly comes from food as organic methylmercury (MeHg) from fish, in particular from Hg-contaminated waters and large marine predatory species (Berlin et al., 2014). MeHg may cause toxic effects on the developing central nervous system (WHO, 2010; Berlin et al., 2014), and probably also causes coronary heart disease (Wennberg et al., 2012). Hg exposure occurs as well from dental amalgam fillings (Berlin et al., 2014). In this case, the exposure is to inorganic Hg, in particular elemental Hg vapour (Hg⁰), which evaporates from the surface of the filling. Hg⁰ passes the blood-brain barrier, and may cause toxic effects but the toxic threshold is not known (Berlin et al., 2014). The Hg concentration in blood (b-Hg) provides a combined index of MeHg and Hg⁰ exposure during the last months.

We here report b-Cd from 1986-2013 and b-Hg from 1990-2013, as measured in large groups of children living in two towns in southern Sweden. These children have participated in long-term monitoring of blood-lead concentrations (b-Pb; children sampled yearly since 1978; Strömberg et al., 1995, 2003, 2008; Skerfving and Bergdahl, 2014; Skerfving et al., 2015).
Material and Methods

**Study area**

Landskrona and Trelleborg, two towns in southern Sweden, were studied (Strömberg et al., 1995). Landskrona, with a population of 39,801 in mid-2006, has a secondary smelter established in 1944. The smelter mainly handles scrap car batteries which may cause limited emissions of Cd and Hg (Landskrona Environmental Management 2014). The smelter is located about 1 km from the town center and annually extracts lead from approximately four million car batteries and about 70,000 tonnes of other types of lead-containing batteries. There are no homes within 0.5 km from the smelter. The town of Trelleborg (population 40,136) has no metal-emitting industries.

**Study subjects**

All children in the selected school classes of 1st to 3rd year (generally 7-10 years of age) were invited and about 60% participated. Each year of sampling, different children were invited and sampled (cross-sectional samples). Also, in 1986, 53 pre-school children (4-7 years of age) from Landskrona participated. Each child did a structured interview and trained nurses did the blood sampling. The interview and blood sampling took place at the schools.

In total, 1,257 children were studied in 1986-2013 (one sample was analysed in each child). Children from Landskrona, sampled in 1986, 1990, 1991, 2004, 2006, 2007, 2009, 2011, and 2013, were divided according to three residential areas: (1) those living 0.5-1 km from the smelter, (2) other urban children, and (3) children living in rural areas. Children from Trelleborg were sampled in 1991, 2003, and 2005, and were divided into urban and rural children. Information on parental smoking habits was obtained through a questionnaire.

The studies were approved by the Ethics Committee at Lund University; informed oral (children) and written (parents) consents were given.

**Sampling**

Blood (4 mL) was obtained once from each child. Blood was drawn from a cubital vein into evacuated heparinized tubes with no detectable amounts of Cd or Hg (Venoject VT-100SH; Treumo Europé, Leuven, Belgium 1986-2006 and Vacuette 4 mL; Lithium Heparin; Greiner-Bio One Gmbh, Frickhausen, Germany 2007-2013). The samples were refrigerated until analysis. All samples collected in one year were analyzed in duplicate in one experiment.

**Cadmium determinations**

Three different analytical methods were employed at different times:

1986: Cd was determined by electrothermal atomic absorption spectrometry (ETAAS, Perkin–Elmer HGA500/AA5000) and a deuterium background correction (Erhart et al., 1985; Willers et al., 1988). In 0.5 mL blood, the proteins were precipitated by the addition of 0.5 mL deionized water and 0.5 mL ultrapure nitric acid. The technique of standard addition was
used and included the addition of two different concentrations to each sample. The limit of detection (LoD) was 0.05 µg/L and the method imprecision, calculated as the coefficient of variation (CV) for duplicate preparations measurements of 30 samples, was 18%. Accuracy was checked by analysis of external quality control (QC) samples.

1991: Samples were analyzed by ETAAS (Varian Spectra AA-40; 283.3 nm; Zeeman background correction). One mL blood was deproteinized by addition of 2 mL of 1.4 M nitric acid (Stoeppler and Brandt 1980, slightly modified). Each sample was prepared in triplicate and Cd standard was added to one of the triplicate samples before deproteinization. The LoD was 0.01 µg/L and the CV calculated from samples without standard added was 11% in the range 0.08 – 0.30 µg/L, and 5% in the range 0.31 – 0.88 µg/L. The accuracy was checked by analysis of external QC samples.

2003-2013: Cd levels were determined by inductively coupled plasma-mass spectrometry (ICP-MS; Thermo X7, Thermo Elemental, Winsford, UK). Samples were diluted ten-fold with an alkaline EDTA solution, according to Bárány et al. (1997). The Cd content was quantified in peak-jumping mode, using 114Cd corrected for the spectral overlap of tin and 115Indium as an internal standard. The LoD was 0.01 – 0.03 µg/L. All samples were prepared in duplicates and the method imprecision (CV) ranged 7.9-20%. QC samples were analyzed along with the samples in each analysis year. In 2003-2007, we participated in the United Kingdom External Quality Assessment Service, with good results.

Mercury determinations
Determinations were made in acid-digested samples by cold vapour atomic fluorescence spectrophotometry. In 1991, Hg was detected by the method described by Einarsson et al. (1984); and the LoD for the method was 0.21 µg/L. The accuracy was checked by analysis of external reference samples. From 2003-2013 the method described by Sandborg-Englund et al. (1998) was used. The LoD ranged 0.04–0.10 µg/L. All samples were prepared in duplicates and the method imprecision (CV) ranged 7.2-8.2%. QC samples were analyzed along with the samples in each analysis year. During the period 2003-2013, we also participated in the Centre de Toxicologie du Quebec International Comparison program with good agreement between obtained element concentrations and expected values.

Statistics
The analysis was based on measurements on Cd and Hg in cross-sectional samples from different years. The predictive values of sample year, sex, school year, residential area, and parents’ smoking habits on b-Cd and b-Hg were examined by generalized linear models (GENLIN in SPSS 22). The outcome variable was In-transformed, as the distribution of residuals indicated that this resulted in a better fit. Thus, in temporal analyses, the exponentiated slope [Exp(B)] indicates the yearly percentage of decrease. P-values below 0.05 were considered “statistically significant”. The measured values of b-Cd and b-Hg were used even when below the formal LoD. This is because such values, though uncertain, contain more information than the usually employed LoD/2 (Helsel 1990).
Results

**Cadmium**

The median b-Cd was 0.10 (n=1120; geometric mean 0.10; range 0.01-0.61) μg/L. The children living near the smelter in Landskrona had higher levels (Table 1) suggesting differences in b-Cd between residential areas in this town. However, in Trelleborg no within-city variations were observed. In addition, the pre-school children had the highest levels of b-Cd compared to other ages. No statistically significant effects of sex or parents’ smoking were found (Table 1).

**Table 1.** Characteristics of Swedish children studied 1986-2013 for concentrations (μg/L) of cadmium and mercury in blood.

<table>
<thead>
<tr>
<th></th>
<th>Cadmium</th>
<th>Mercury</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>N</td>
<td>GM</td>
</tr>
<tr>
<td>Sex</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Boys</td>
<td>594</td>
<td>0.10</td>
</tr>
<tr>
<td>Girls</td>
<td>526</td>
<td>0.10</td>
</tr>
<tr>
<td>Neck smelter</td>
<td>147</td>
<td>0.11</td>
</tr>
<tr>
<td>Urban</td>
<td>464</td>
<td>0.10</td>
</tr>
<tr>
<td>Rural</td>
<td>295</td>
<td>0.090</td>
</tr>
<tr>
<td>Trelleborg</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Urban</td>
<td>95</td>
<td>0.091</td>
</tr>
<tr>
<td>School class</td>
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<td></td>
</tr>
<tr>
<td>0</td>
<td>56</td>
<td>0.12</td>
</tr>
<tr>
<td>1</td>
<td>225</td>
<td>0.093</td>
</tr>
<tr>
<td>2</td>
<td>496</td>
<td>0.093</td>
</tr>
<tr>
<td>3</td>
<td>143</td>
<td>0.10</td>
</tr>
<tr>
<td>Parental smoking</td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>283</td>
<td>0.092</td>
</tr>
<tr>
<td>Yes</td>
<td>1120</td>
<td>0.10</td>
</tr>
</tbody>
</table>

GM = Geometric Mean.

\( ^a \) No blood mercury was analyzed in samples from Trelleborg.

\( ^b \) At least one parent reporting smoking.

We observed significant fluctuations of b-Cd over the sampling years (Figure 1). The exponential slope was 0.993 (95% confidence interval 0.990, 0.995). The estimate did not change when adjusted for residence area or school year.
Figure 1. Blood-cadmium concentrations (b-Cd; geometric means) in 1,120 Swedish children during 1986-2013. An exponential regression line is shown.

**Mercury**

The median b-Hg was 0.91 (n=560; geometric mean 0.83; range 0.02-8.2) μg/L. Like for Cd, we found that children living near the smelter in Landskrona had higher levels of b-Hg than in the other areas (Table 1). We found no statistically significant effects of sex, parents’ smoking or school class on b-Hg.

Figure 2. Blood-mercury concentrations (b-Hg; geometric means) in 560 Swedish children during 1991-2013. An exponential regression line is shown.
We observed a decrease of b-Hg over time (Figure 2). The exponential slope was 0.967 (95% confidence interval 0.961, 0.973). The estimate did not change when adjusted for residence area or school year.

**Discussion**

The main findings were that the children in these two towns had very low b-Cd and that their levels might have slightly decreased (0.7% per year) during 1986-2013. These children also had very low b-Hg. However, b-Hg showed a much more pronounced decrease (3% per year) during 1991-2013. In addition, in Landskrona there was an effect of residential area near the smelter for both metals. The preschool children had higher b-Cd than the older ones, but no statistically significant effects of sex or parents’ smoking was found on b-Cd or b-Hg.

*Cadmium concentrations and time trend*

The children sampled in the present study had lower b-Cd (0.10 µg/L) than the observed in five other European countries (0.13-0.17 µg/L), Ecuador (0.26 µg/L) and Morocco (0.21 µg/L) (Hrubá et al., 2012). We did not see any increase of b-Cd with age. However, since adults in the same geographic area have much higher b-Cd (Åkesson et al., 2005 and 2006; Suwasono et al., 2006; Pawlas et al., 2013), increase of Cd in the body will probably occur later.

Moreover, no effect of sex on b-Cd was observed most probably due to the low age of the children. In adults, females tend to accumulate more Cd than men (Pawlas et al. 2013) because of low iron pools (caused by blood loss during menstruation), which causes an increase in gastrointestinal absorption of Cd (Nordberg et al., 2014).

Regarding time trends of Cd exposure in children, limited information is currently available. There might have been a decrease of b-Cd in German children (age 6-14) from 1990/1992 (geometric mean 0.14 µg/L) to 2003/2006 (<0.12 µg/L), but the concentrations were close to (or below) the detection limit of the analytical method (Schulz et al., 2007).

The slight decrease in b-Cd over time observed in this study should be interpreted cautiously since any decrease is dependent on the single observation from 1986. In fact, during 2003-2013 there was no decreasing trend over time. Nevertheless, considering the dramatic decrease to one-third of Cd deposition during the same period in Sweden (WHO, 2007), the country of Scania where both studied cities are located (IVL, 2007) and in Landskrona specifically (Nordell, 2007), the lack of change in b-Cd is still remarkable. However, this limited effect of the declining pollution is probably because vegetables are the dominant source of exposure (Ohlsson et al., 2005). The fact that Cd in plants comes from the absorption through the root and the Cd content of agricultural soil only changes very slowly after a change of deposition (Clemens et al. 2013), may explain why no decrease over time is seen in this population.
Another finding is the positive association between living near the smelter and b-Cd concentrations. In areas where Cd pollution is higher than in our environment, there is an effect on b-Cd of industrial emissions (Hogervorst et al., 2007; Schroijen et al., 2008). We found a similar pattern in Landskrona despite the fact that the factory is a secondary lead smelter, which mainly emits lead (Nordell, 2007).

Also, no effect of parents’ smoking status was seen on b-Cd, in accordance with a previous report on the same group from 1986 (Willers et al., 1988). Despite tobacco containing high concentrations of Cd, the air levels of Cd in smoker’s dwellings are low (Willers et al., 1993). However, dust in smoker’s homes contains Cd (Willers et al., 1993) and an association between b-Cd and house dust has been established (Hogervorst et al., 2007). Nevertheless, it might be that the exposure in our children is lower.

According to current knowledge, the Cd levels in the present children are not toxic, but it might be that Cd accumulation over time will result in adverse effects on kidneys and bone later in life, particularly in women. Such effects have been shown in elderly women in the same area (Åkesson et al., 2005, 2006, 2014; Suwasono et al., 2006). Thus, even if the children’s exposure was low, there are strong reasons to reduce Cd pollution.

**Mercury concentrations and time trend**

In the children in the present study, b-Hg (0.83 µg/L) was lower than in one European country (Slovenia 0.94 µg/L), China (2.45 µg/L) and Ecuador (3.23 µg/L) (Hrubá et al., 2012). The levels were also lower than in adult women from the same area (1.37 µg/L), as well as from northern Sweden (1.38 µg/L) (Pawlas et al., 2013).

There was a clear decrease of b-Hg over time by about 3% per year. This decrease should be compared with the decrease of 5.8% per year for erythrocyte Hg concentrations reported earlier in Swedish (Wennberg et al., 2006) and no apparent decrease in Czech (Cerna et al., 2012) adults. Since fish consumption is such an important determinant of b-Hg (Bárány et al., 2003; Hrubá et al., 2012), the decay of b-Hg is most likely due to a decreasing intake of MeHg. Though MeHg in Swedish freshwater fish has increased over time (Åkerblom and Johansson, 2008), 85% of the fish intake in Swedish children is as fish sticks, i.e. whitefish and cultured salmon (Becker and Engelhardt Barbieri, 2004), which contain minor amounts of MeHg (EFSA 2012, Kelly 2008). Also, dental amalgam is an important source of b-Hg in European countries, though presently not in Swedish children, in whom there are extremely few such fillings, without any association with b-Hg (Hrubá et al., 2012). However historically, amalgam was very widely used in Swedish children, until a prohibition in 1995 (National Swedish Board of Health and Welfare, 2006). Thus, it is likely that a major fraction of the decrease of b-Hg seen in our study was due to the abolishment of amalgam.

Further, the Landskrona smelter had an impact on b-Hg although it officially does not emit Hg, a fact which warrants follow-up to reduce emissions of Hg from the smelter. Parental
smoking did not affect b-Hg, which is in accordance with earlier findings, that not even active smoking causes an increase (Wennberg et al., 2006).

The current b-Hg levels in children are low but it is not known whether they may cause adverse effects in the future. The major concern at the moment is the central nervous system effects in foetuses (Berlin et al., 2014) and cardiovascular disease (Wennberg et al., 2012), areas in which there is a need for much more information.

**Methodological considerations**

About 40% of the eligible children refrained from donating blood. However, there is no reason to suspect that willingness to participate was associated with Cd or Hg exposure.

Since we consecutively used three different analytical techniques, the quality of the analyses is crucial. To guarantee this, we participated in external quality control schemes and frequently analyzed external quality-control material. Particular efforts were paid to any effects of changes of analytical method. However, we cannot exclude the possibility that varying CVs for blood Cd may account in part for null results.

One b-Cd result (9.7 µg/L) was excluded from the statistical analyses. It was an extreme outlier, 16 times higher than the next highest one (0.61 µg/L). This was probably due to contamination, since a later sampling of the child showed a b-Cd of 0.26 µg/L.

**Conclusions**

Children from southern Sweden have very low exposure to Cd and their b-Cd decreased not at all or only slightly, from 1986 to 2013. Since the present exposure might mean a risk later in life for girls, Cd pollution should still be further restricted.

In the case of Hg, b-Hg was relatively low and there was a clear reduction over time by 3% per year. This decrease was probably because of lower fish intake and reduced use of dental amalgam. At the moment, the b-Hg does not pose any great health risks for future.

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