MEASURABLE IMPACT OF AN OLD MSWI ON THE LEVEL OF DIOXINS IN FREE-RANGE CHICKENS AND EGGS GROWN IN ITS VICINITY

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Introduction

In the past, eggs from free ranging chicken have already been followed-up and showed relatively high level of dioxins compared to those from commercial battery-farming\textsuperscript{1,2,3}. Soils, and their incorporated organisms appeared to be the main source of dioxin contamination\textsuperscript{1,2,4,5} for such foraging poultries since soils are known to act as a conservative matrix for long term dioxin deposition\textsuperscript{5,6}. Foraging animals, and especially chickens and cows, can therefore be used as efficient bioindicators of potential environmental dioxin contamination\textsuperscript{1}. Monitoring of levels in milk or eggs from such animals raised in the vicinity of known emission sources such as chemical waste incinerator (CWI)\textsuperscript{4}, pentachlorophenol wood treatment facilities\textsuperscript{1,7} or municipal solid waste incinerator (MSWI)\textsuperscript{8,9,10} is thus often carried out. However, very few of these surveys have tried to correlate dioxin levels found in eggs or milk with those found in corresponding soils\textsuperscript{1,7,10}. Most of the works performed to study the feed to animal transfer are usually carried out using feed mixed with contaminated soil in controlled exposure conditions\textsuperscript{2,11,12}. Although experimental conditions are set to reflect real situations, these experiments can not reflect it scrupulously. In the present study, dioxins levels in soil, egg and chicken samples from a potential contaminated area have been measured and compared. This selected area had been under the influence of an old MSWI emissions for more than 20 years.

Methods and Materials

Sampling: Samples were collected from 3 homes in Maincy, a small French village located at about 60km south of Paris. This rural area was subject to the emission of an old MSWI for over 20 years.
years. This MSWI had recently been closed since very high dioxin emission rate, more than 2000 fold higher than the actual European norm of 0.1ngTEQ/Nm³, had been recorded. Figure 1 shows the map locating the 3 sites and the MSWI. Sites selected for soil sampling were located between 1250 and 1500m from the MSWI, under the prevailing wind stream (NE). Soils were sampled to a depth of 10 cm, at 2 distinct randomly selected spots, pooled and kept into polyethylene (PE) sampling bags at room temperature. For each site, abdominal fat from 2 chickens were taken and kept in PE vials at 4°C. Eggs were hard-boiled, yolks separated from white, and placed at –20°C in PE vials. Commercial eggs and chicken were purchased in an hypermarket located in the surrounding area.

**Analytical method:**
The analytical procedures for determination of PCDD/Fs and c-PCBs in eggs and abdominal fat have already been reported\(^1^3\). Soil samples were dried at 100°C overnight, mixed with acidic silicagel and extracted by pressurized toluene extraction (ASE\(^\text{TM}\) 200, Dionex, Sunnyvale, CA, USA). \(^1^3\)C labelled standards (isotopic dilution) were added to the sample in the extraction cell prior to extraction. Extract were evaporated and re-diluted in hexane. Further clean-up was carried out using the Power-Prep\(^\text{TM}\) (FMS Inc., Waltham, MA, USA) system using classical sets of columns\(^1^3\). Analyses were performed on an Agilent (Palo Alto, CA, USA) 6890 Series gas chromatograph coupled to an Autospec Ultima (Micromass, Manchester, UK) high resolution mass spectrometer. The capillary column was a RTX-5SIL-MS (30m x 0.25mm I.D., 0.25µm film thickness) (Restek, Evry, France).

**Results and Discussion**
According to the results of several international studies\(^1^4,1^5,1^6\), soils sampled within 1500m of a MSWI would be under its emission influence and would show specific congener distributions in addition to higher levels.

As shown in Figure 2 (non 2,3,7,8-substituted congeners were found but are not reported), profiles are similar to homologue distributions found in soils collected near incinerators in Spain\(^1^5\), United States\(^1^6\) and Japan\(^1^7\). These are characterized by high level of OCDD which contributes for between 55 and 80% of the toxic congener concentration, HpCDD, HpCDF and OCDF being the next most prominent congener in roughly the same proportions (about 10%). Soil from Site#2
shows a bit different figure with an OCDD contribution of only 25%, and an increased the furan contribution.

<table>
<thead>
<tr>
<th></th>
<th>Soil</th>
<th>Abdominal fat</th>
<th>Eggs</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>sample #1</td>
<td>sample #2</td>
</tr>
<tr>
<td>Home 1</td>
<td>409.4</td>
<td>143.4</td>
<td>244.6</td>
</tr>
<tr>
<td></td>
<td>[13.4]</td>
<td>[37.9]</td>
<td>[74.7]</td>
</tr>
<tr>
<td>Home 2</td>
<td>132.3</td>
<td>317.1</td>
<td>731.6</td>
</tr>
<tr>
<td></td>
<td>[11.1]</td>
<td>[60.7]</td>
<td>[121.1]</td>
</tr>
<tr>
<td>Home 3</td>
<td>1837.1</td>
<td>151.6</td>
<td>256.1</td>
</tr>
<tr>
<td></td>
<td>[12.8]</td>
<td>[34.3]</td>
<td>[55.2]</td>
</tr>
<tr>
<td>Bio-products</td>
<td>-</td>
<td>1.32</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>-</td>
<td>[0.25]</td>
<td>-</td>
</tr>
</tbody>
</table>

Table 1 Total concentration of 2,3,7,8-substituted congeners in different matrices. Results are expressed in pg/g of dry weight for soils and in pg/g of fat for abdominal fat and eggs. In square bracket are reported total concentrations in pgTEQ/g dry weight or pgTEQ/g fat.

As it appears in Table 1, the soil from the Site#3 showed very high total concentration. This was due to the great contribution of OCDD and HpCDD, which did not significantly influence the TEQ value that was similar for the 3 different soils, regardless to the type of sampled surface (Table 2). This is quite surprising because one could have expected that pasture (Site#1) or arable farming soil (Site#2) would show higher background values because more exposed to atmospheric deposition than the manure layer of hens house soil (covered area) from Site#3.

In the present study, measured concentrations are higher than the 0.1 to 6 pgTEQ/g of dry weight range usually reported for soil surfaces sampled close to operating European incinerator 14, 15, 18. On the other hand, these results match with another study reporting levels for soils collected near another very old incinerator 18. Soil concentrations, however, remain inside the range of European soil backgrounds.

Table 2 Information’s on breeding

<table>
<thead>
<tr>
<th>Home</th>
<th>Distance from MSWI</th>
<th>Direction</th>
<th>Type of “accommodation”</th>
<th>Space/hen (meters²)</th>
<th>Eggs/day</th>
<th>Age</th>
<th>Commercial feed</th>
<th>Quantities</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1500 m</td>
<td>NE</td>
<td>pasture land</td>
<td>150</td>
<td>1</td>
<td>1</td>
<td>yes</td>
<td>ad libitum</td>
</tr>
<tr>
<td>2</td>
<td>1250 m</td>
<td>NE</td>
<td>arable land</td>
<td>70</td>
<td>1</td>
<td>3</td>
<td>no</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>1250 m</td>
<td>NE</td>
<td>hens house</td>
<td>3</td>
<td>1/2</td>
<td>5</td>
<td>yes</td>
<td>ad libitum</td>
</tr>
</tbody>
</table>

Regarding chicken and egg samples, although not observed in our previous transfer study where we used feed artificially contaminated with oil 13, small amount of non 2,3,7,8-substituted congeners have been detected in all samples. As it can be seen in Table 1, large variations in concentrations are observed between the animals originated from the different sites, as well as inside the same site. These differences for chickens from the same hen house are quite difficult to interpret. The chicken corresponding to sample #1 from Site#1 was characterized by twice the fat content of the chicken corresponding to sample #2. Assuming an equivalent diet, one could assume a dilution of the dioxin burden in the inverse proportion. Such an hypothesis is, however, not confirmed for chickens issued from Sites #2 and #3, which roughly have the same fat content. Furthermore no differences between hens have been observed in term of race, age, laying rhythm, etc. Despite the inter-individual variations, a common trend emerges from the 3 different sites. The levels recorded in egg and abdominal fat samples issued from Site#2 are higher, even though levels in soil are the lowest. By analysing breeding information’s (Table 2), one can assume that...
this would most probably be due to the absence of commercial feed available for hens from Site#2. Animals would therefore get themselves most of their feed from the environment (maize from farming, vegetation, soil organisms, etc). Hence, the intake from contaminated soil would be bigger than for chickens for which commercial food is provided at libitum. Due to the free-ranging aspect of the breeding, the correlation between dioxin concentration in egg and abdominal fat is very uneasy because eggs could not be traced back to a specific hen. Nevertheless as for abdominal fat, eggs picked up in Site#2 showed markedly higher mean level than those picked up in other sites. The levels in eggs are higher than those observed by Harnly et al.7 (10 pg TEQ/g fat) for eggs issued from hens foraging on a twice more contaminated soils, but are similar to some levels reported in Belgium (20 pg TEQ/g fat) for eggs from free-ranging chickens raised in private gardens characterized by a 2 pg TEQ/g level in soil.

Conclusions
Concentrations of dioxins in egg and free-ranging chicken samples in a small village close to an old MSWI are found to be more than 15 times higher than the European norm set at 3 pg TEQ/g fat and would potentially be harmful for exposed population. Although soil has already been demonstrated to be the primary dioxin exposure source for foraging animal, levels measured in sampled soils are comparable to those recorded in European rural area. Soils therefore can not be the only reason of this worrying contamination case. Moreover, level variations between animals issued from the same sampling site have been observed. This makes difficult the extrapolation of the present results to general situations, in order to establish transfer coefficient from soils to eggs.

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References