# EFSA's RISK ASSESSMENT ON THE PRESENCE OF HIGH LEVELS OF DIOXINS AND DIOXIN-LIKE PCBs IN LIVER FROM SHEEP

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## Introduction

The European Food Safety Authority (EFSA) was established in January 2002 as an independent body providing scientific advice and communication on risks associated with the food chain (Regulation (EC) No 178/2002). As a risk assessor, EFSA produces scientific opinions and advice to provide a sound foundation for European policies and legislation and to support the European Commission (EC), European Parliament and EU Member States in taking effective and timely risk management decisions.

Comprehensive monitoring programmes conducted worldwide during the past two decades showed that human exposure to dioxins and PCBs has decreased significantly over time. In this respect, the term "dioxins" refers to polychlorinated dibenzo-p-dioxins (PCDDs) and dibenzofurans (PCDFs) together. The monitoring programmes also demonstrated that certain food commodities, such as sheep liver and deer liver, can have high dioxin levels even when not affected by specific contamination sources. In 2008/2009 a number of sheep samples were analysed in Germany for dioxins and PCBs. Most sheep meat samples were below the respective maximum levels set by Regulation (EC) No 1881/2006. However, the corresponding liver samples from the same sheep in almost all cases exceeded the respective maximum levels considerably. Investigations indicated that these high levels were not due to poor husbandry practices or high localized contamination but were much more likely to be associated with the physiology of the animals.

Following a request from the EC, EFSA was asked to deliver a scientific opinion on the risk to public health related to the presence of high levels of dioxins and dioxin-like polychlorinated biphenyls (DL-PCBs) in liver from sheep and deer. The opinion should also explore possible reasons for the findings of high levels of dioxins and PCBs in liver from sheep and deer, and to provide scientific elements on the appropriateness to establish in future regulatory levels in liver on a product basis rather than on a fat basis. This task was allocated to the EFSA Panel on Contaminants in the Food Chain (CONTAM Panel) and a Working Group was established for this purpose[1].

## Materials and methods

EFSA collected and evaluated the dioxin and DL-PCB results from 332 sheep liver, 175 sheep meat and 9 deer liver samples submitted by eight European countries. The data were either submitted directly to EFSA by the European countries or sent to the European Commission and then forwarded to EFSA. More than 60 % of the results were from Germany. Occurrence data for NDL-PCBs were submitted by eight European countries for 257 sheep liver, 146 sheep meat and 9 deer liver samples.

For the exposure assessment the occurrence data on dioxins and PCBs in sheep liver were combined with the data on human consumption of sheep liver. Consumption data on sheep liver are scarce in Europe. Data extracted from the EFSA's "Comprehensive European Food Consumption Database" indicate that only a very small fraction of the European population consumes sheep liver. The available data for adults from six countries show that less than 3 % are consumers of "mutton and lamb liver". The methodology used to collect the food consumption data differ between surveys affecting comparability. In three cases the surveys reported data taken on a 7-day food record basis, while the remaining three are either reported as 24 hour recall or as a 3-day food record, with consumption data reported either "as raw", "as consumed" or "as cooked". Consequently, calculation of average consumption of sheep liver expressed on a weekly basis for the small number of sheep liver consumers in the total adult population is subject to a high degree of uncertainty. A viable alternative is to use the portion size distribution assuming that an arbitrary frequency of one eating occasion in a week can be taken as a conservative estimate. For adults, the results show an average sheep liver portion size of 106 g (or 1.5 g/kg b.w.) across six countries with the highest average value being in the UK of 141 g (or 1.9 g/kg b.w.). The average sheep liver portion size recorded in the UK has been used for the exposure assessment with the assumption that it represents a weekly amount. Assuming that sheep liver consumption in children is similar to consumption of "all liver", the average weekly amount of 2.8 g/kg b.w. from a Bulgarian survey was selected as a conservative estimate for the exposure assessment for children. Because of the small fraction of the European population that consumes sheep liver, the CONTAM Panel did not evaluated the exposure through consumption of sheep liver for the total European population but for "consumers only". As it was assumed that sheep liver is not consumed daily, the evaluation was based on a potential weekly intake.

Two different concentrations for the sum of dioxins and DL-PCBs in sheep liver were used in this evaluation. These are the mean levels calculated from the results reported by the submitting European countries and the maximum level laid down in Regulation (EC) No 1881/2006. The dietary intake was estimated using upper bound (UB) concentrations as UB and lower bound (LB) values were coinciding when rounded to the first decimal. An average fat content of 5.1 % for sheep liver was calculated from the submitted occurrence data and used in the exposure assessment.

**Results and discussion:**

For sheep liver, the mean UB concentrations for dioxins and the sum of dioxins and DL-PCBs (expressed as $WHO\text{-}TEQ_{1998}$) were 14.9 pg WHO-TEQ/g fat (range: 0.27-116) and 26.1 pg WHO-TEQ/g fat (range: 0.47-279), respectively. The corresponding levels in sheep meat were considerably lower: 0.70 pg WHO-TEQ/g fat (range: 0.08-5.1) and 2.0 pg WHO-TEQ/g fat (range: 0.16-11.9), respectively. The mean value for the sum of dioxins and DL-PCBs in deer liver was almost 2.5-fold the mean value in sheep liver.

For sheep liver and sheep meat the mean UB concentrations for the sum of the six NDL-PCBs (PCB-28, -52, -101, -138, -153 and -180) identified as indicator-PCBs by the CONTAM Panel in 2005, were 26.8 ng/g fat (range: 0.41-350) and 13.1 ng/g fat (range: 0.51-162), respectively.

The mean NDL-PCB values for sheep liver and deer liver were comparable. In addition to higher levels of dioxins in sheep liver, also a distinct difference was observed in the relative contributions of PCDDs, PCDFs and DL-PCBs to total TEQ. In general, the relative contribution of PCDFs in sheep liver was considerably higher compared to sheep meat.

The average weekly exposure to the sum of dioxins and DL-PCBs based on the mean concentration calculated from the occurrence data submitted by the European countries is 2.5 pg WHO-TEQ/kg b.w. for adults (consumers only) (**Table 1**). When using the maximum level laid down in Regulation (EC) No 1881/2006 the average exposure is 1.2 pg WHO-TEQ/kg b.w. For children, the average weekly exposure is 3.7 pg WHO-TEQ/kg b.w. and 1.7 pg WHO-TEQ/kg b.w. considering the occurrence data submitted by the European countries and the maximum level in the legislation, respectively. For NDL-PCBs, the exposure estimations were performed with the mean concentration for the sum of the six indicator NDL-PCBs (PCB-28, -52, -101, -138, -153 and -180) calculated from the occurrence data submitted by the eight European countries. Harmonized maximum levels for the sum of these six congeners in various food categories are foreseen to be set soon. The average weekly dietary exposure for adults (consumers only) is 2.6 ng/kg b.w. For children, the average weekly exposure to NDL-PCBs based on the consumption value derived by the "generic liver" data is 3.8 ng/kg b.w. (**Table 2**).

In order to characterize the risk of chronic consumption of sheep liver, calculations were made to compare how much the consumption of sheep liver would add to the total human exposure and how this compares with the tolerable weekly intake (TWI) for dioxins and DL-PCBs. As TWI the CONTAM Panel used the value of 14 pg TEQ/kg b.w. per week established by the SCF in 2001. As a starting point, a human background daily intake was derived from the literature. The median dietary intake of dioxins and DL-PCBs across European countries for which data were reported as $WHO_{1998}$-TEQs is 1.53 (0.51-3.2) pg/kg b.w. per day or converted to a weekly basis around 11 (3.6-23) pg WHO-TEQ/kg b.w. The CONTAM Panel noted that data on dietary exposure was only available from a limited number of European countries and these might not reflect the most recent exposure. The data based on the $WHO\text{-}TEF_{1998}$s were used because a considerable number of occurrence data was only reported as TEQs calculated with these TEFs without giving the raw data which made a conversion with the most recent $WHO\text{-}TEF_{2005}$s not possible. Applying the latter TEFs may lead to 10-15 % lower values.

For adults, consumption of about 140 g (or 1.9 g/kg b.w.) sheep liver with the mean value of 26.1 pg WHO-TEQ/g fat would result in a weekly intake of 2.5 pg WHO-TEQ/kg b.w. and in a total weekly intake of 13.5 pg WHO-TEQ/kg b.w., taking a median background exposure of 11.0 pg WHO-TEQ/kg b.w. into account (**Table 1**). This is 96 % of the TWI. As a number of sheep liver samples showed considerably higher concentrations for dioxins and DL-PCBs, it can be assumed that frequent consumers may be exposed on individual occasions to much higher values.

Due to lack of data, the consumption of sheep liver for children was based on an age range up to 18 years old. As respective current data on background exposure to dioxins and DL-PCBs for children are sparse, the CONTAM Panel decided not to estimate a median dietary background exposure for children. The assessment indicates that for children the exposure to dioxins and DL-PCBs through consumption of sheep liver is approximately 50 % higher as compared to adults because of the higher food consumption relative to body weight.

Assuming a weekly background exposure of 35-161 ng/kg b.w. for the sum of the six indicator NDL-PCBs[2], the additional contribution through consumption of sheep liver for adults amounts to 1.6-7.4 %. The respective evaluation for children would result in an additional weekly intake between 2.2 and 11 % (**Table 2**).

**Table 1.** Additional and total weekly intake of dioxins and DL-PCBs (pg $WHO_{1998}$-TEQ/kg b.w.) for adults (consumers only) and additional weekly intake for children up to 18 years old through consumption of sheep liver. A median weekly background exposure for adults of 11 pg WHO-TEQ/kg b.w. was taken into account.

| Population | Weekly consumption of sheep liver (g/kg b.w.) | Concentration of dioxins and DL-PCBs in sheep liver (pg WHO-TEQ/g fat) | Weekly intake of dioxins and DL-PCBs from sheep liver (pg WHO-TEQ/kg b.w.) | Total median weekly intake of dioxins and DL-PCBs (pg WHO-TEQ/kg b.w.) |
|---|---|---|---|---|
| **Adults** | 1.9 | 26.1 [(a)]<br>12.0 [(b)] | 2.5<br>1.2 | 13.5<br>12.2 |
| **Children** | 2.8 | 26.1 [(a)]<br>12.0 [(b)] | 3.7<br>1.7 | - |

**Table 2.** Additional and total weekly intake of the sum of six indicator NDL-PCBs (ng/kg b.w.) for adults (consumers only) and children through consumption of sheep liver assuming a weekly background exposure of 35-161 ng/kg b.w. for adults and 35-175 ng/kg b.w. for children up to 18 years old.

| Population | Weekly consumption of sheep liver (g/kg b.w.) | Concentration in sheep liver (ng/g fat) | Weekly intake of NDL-PCBs from sheep liver (ng/kg b.w.) | Total additional weekly intake of NDL-PCBs (ng/kg b.w.) (% of background exposure) |
|---|---|---|---|---|
| **Adults** | 1.9 | 26.8 | 2.6 | 1.6-7.4 |
| **Children** | 2.8 | 26.8 | 3.8 | 2.2-11 |

In conclusion, regular consumption of sheep liver would result on average in an approximate 20 % increase of the background exposure to dioxins and DL-PCBs. On individual occasions, consumption of sheep liver could result in high intakes exceeding the TWI. The CONTAM Panel concluded that the frequent consumption of sheep liver, particularly by women of child-bearing age and children, may be a potential health concern.

For deer liver only nine results were reported on concentrations for dioxins, DL-PCBs and NDL-PCBs. This number of samples is too small to perform a meaningful risk assessment. However, as the reported concentrations for dioxins and DL-PCBs were generally higher than the concentrations for sheep liver (with an almost 2.5-fold higher mean value), the CONTAM Panel concluded that frequent consumption of deer liver, especially for high consumers, may be of health concern.

The European Commission also asked EFSA to provide scientific elements on the appropriateness to establish in future regulatory levels in liver on a product basis rather than on a fat basis. In general, the expression of results on a product basis would be preferable from a dietary exposure point of view as this would better reflect the exposure to the consumed products. The CONTAM Panel noted that the fat content of sheep liver reported in literature and submitted by the European countries generally range from 3 to 8 % fat with a mean content of 5.1 %. Comparable fat contents are found for liver samples of other terrestrial animals, such as bovine, pigs and chicken. These ranges of fat content are considerably narrower than for a number of other food categories regulated in Regulation (EC) No 1881/2006 such as dairy products which cover a range from 1 to >80 %. Even if there would be a possible hepatic sequestration and the dioxins and PCBs would not be totally associated with the fat fraction of the liver, this would have no influence on the result, whether based on lipid or fresh weight basis, as all dioxins and PCBs are extracted during the analytical procedure irrespective of the liver compartment where they are present. Therefore, the CONTAM Panel sees no need to change the basis for expression of results for liver only. A change of the expression of maximum levels seems only meaningful if all food categories would be considered.

The CONTAM Panel also explored possible reasons for the findings of high levels of dioxins and PCBs in liver from sheep and deer. Similar to other mammalian species, sheep are able to metabolize DL-PCBs and probably dioxins to hydroxy-derivatives, very likely through cytochrome P450 (CYP) 1A enzymes. Studies *in vitro* and *in vivo* with prototype substrates for CYP1A enzymes indicate a lower CYP1A1 activity in sheep than in cattle and suggest that differences in metabolism might be possible explanations for the marked differences in the liver storage of dioxins and related compounds between the two species. The differences in liver CYP1A expression between the two species, however, remain to be confirmed. Furthermore, as demonstrated in rodents, it cannot be excluded that other mechanisms such as the sequestration of dioxins and dioxin-like compounds by hepatic CYP1A2 or their biotransformation by other enzymes, may affect their accumulation in the liver of ruminants.

**Acknowledgements:**
EFSA wishes to thank the members of the EFSA CONTAM Panel. EFSA and the CONTAM Panel acknowledge all the European countries that provided occurrence data of dioxins and PCBs in food and supported the consumption data collection for the Comprehensive European Food Consumption Database.

**References:**
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