# ANALYTICAL REQUIREMENTS FOR DETERMINATION OF NON DIOXIN-LIKE PCBS IN FOOD (PCB # 28, 52, 101, 138, 153, 180)

Malisch R[1], Kotz A[1] and Wahl K[1]

[1] *EU Community Reference Laboratory (CRL) for Dioxins and PCBs in Feed and Food, State Laboratory for Chemical and Veterinary Analysis (Chemisches und Veterinäruntersuchungsamt, CVUA), Bissierstr. 5, D-79114 Freiburg, Germany*

## Abstract

The EU Commission has initiated steps to set maximum levels for non dioxin-like PCBs expressed as sum of the concentrations of six indicator PCBs (PCB 28, PCB 52, PCB 101, PCB 138, PCB 153 and PCB 180) in certain food samples. Therefore, the EU Community Reference Laboratory (CRL) for Dioxins and PCBs in Feed and Food was asked by the EU Commission to develop a working document in cooperation with the network of National Reference Laboratories (NRLs) of the EU Member States containing criteria for the analysis of non-dioxinlike PCBs (ndl-PCBs) as basis for possible future EU legislation. This working document proposes criteria for analytical methods for determination of PCB concentrations for control of the maximum levels under discussion (= levels of interest). The following aspects are covered: 1. Applicable detection methods, 2. Identification and confirmation of analytes of interest; 3. Demonstration of performance of method; 4. Limit of quantification; 5. Quality control; 6. Control of recoveries; 7. Requirements for laboratories; 8. Upper bound calculation; 9. Criteria for the sum of the six indicator PCBs at the level of interest (Trueness - 30 to + 30 %; Intermediate precision (RSD%) ≤ 20 %; Difference between upper and lower bound calculation ≤ 20 %).

## Introduction

The EU Commission has initiated steps to set maximum levels for non dioxin-like PCBs expressed as sum of the concentrations of six indicator PCBs (PCB 28, PCB 52, PCB 101, PCB 138, PCB 153 and PCB 180) in certain food samples. Therefore, the EU Community Reference Laboratory (CRL) for Dioxins and PCBs in Feed and Food was asked by the EU Commission to develop a working document in cooperation with the network of National Reference Laboratories (NRLs) of the EU Member States containing criteria for the analysis of non-dioxin like PCBs (ndl-PCBs) as basis for possible future EU legislation.

For orientation, the analytical performance criteria for residues of dioxins and dioxin-like PCBs in food [1], pesticides in food [2] and residues of several groups in food of animal origin [3] were used.

This working document proposes criteria for analytical methods for determination of PCB concentrations for control of the maximum levels under discussion (= levels of interest). The working document was discussed at the workshop of the CRL and NRLs for dioxins and PCBs in feed and food, held on 13 November 2008 in Freiburg, and agreed by all participants.

## Results: Requirements for analytical procedures

1. **Applicable detection methods:**
   GC/ECD, GC/LRMS, GC/MS-MS, GC/HRMS or equivalent methods

2. **Identification and confirmation** of analytes of interest:
   - ➢ Relative retention time in relation to internal standards or reference standards (acceptable deviation of +/- 0.25 %)
   - ➢ Gas chromatographic separation of all six indicator PCBs (PCB 28, PCB 52, PCB 101, PCB 138, PCB 153 and PCB 180) from interfering substances, especially co-eluting PCBs, in particular if levels of samples are in the range of legal limits and non-compliance is to be confirmed.
     [Congeners often found to co-elute are e.g. PCB 28/31, PCB 52/69 and PCB 138/163/164. For GC/MS also possible interferences from fragments of higher chlorinated congeners have to be considered.]
   - ➢ For GC/MS techniques:
     - ✓ Monitoring of at least
       - two specific ions for HRMS,
       - two specific ions of m/z > 200 or three specific ions of m/z > 100 for LRMS,
       - 1 precursor and 2 product ions for MS-MS
     - ✓ Maximum permitted tolerances for abundance ratios for selected fragment masses:

       Relative deviation of abundance ratio of selected mass fragments from theoretical abundance or calibration standard for target ion (most abundant ion monitored) and qualifier ion(s):

| Relative intensity of qualifier ion(s) compared to target ion | GC-EI-MS (relative deviation) | GC-CI-MS, GC-MS$^n$, (relative deviation) |
|---|---|---|
| > 50 % | ± 10 % | ± 20 % |
| > 20 % to 50 % | ± 15 % | ± 25 % |
| > 10 % to 20 % | ± 20 % | ± 30 % |
| ≤ 10% | ± 50 % *) | ± 50 % |

*) Sufficient number of mass fragments with relative intensity > 10 % available, therefore not recommendable to use

   - ➢ For GC/ECD:
     Confirmation of results exceeding the tolerance with two GC columns with stationary phases of different polarity

3. **Demonstration of performance of method:**
   Validation in the range of the level of interest (0.5 to 2 times the level of interest) with an acceptable coefficient of variation for repeated analysis (see requirements for intermediate precision in no. 9)

4. **Limit of quantification:**
   Limit of quantification shall be in the range of about one fifth of the level of interest (for sum of six indicator PCBs).

5. **Quality control:**
   Regular blank controls, analysis of spiked samples, quality control samples, participation in interlaboratory studies on relevant matrices

6. **Control of recoveries:**

- ➢ Use of suitable internal standards with physico-chemical properties comparable to analytes of interest
- ➢ Addition of internal standards:
  - ✓ Addition to products (before extraction and clean-up process);
  - ✓ Addition also possible to extracted fat (before clean-up process), if maximum level on fat basis.
- ➢ Methods using all six isotope-labelled indicator PCB congeners:
  - ✓ Correction of results for recoveries of internal standards,
  - ✓ Generally acceptable recoveries of isotope-labelled internal standards between 50 and 120 %;
  - ✓ Lower or higher recoveries for individual congeners with a contribution to the sum of six indicator PCBs below 10 % are acceptable.
- ➢ Methods using not all six isotope-labelled internal standards or other internal standards:
  - ✓ Control of recovery of internal standard(s) for every sample,
  - ✓ Acceptable recoveries of internal standard(s) between 60 and 120 %,
  - ✓ Correction of results for recoveries of internal standards.
- ➢ The recoveries of unlabelled congeners should be checked by spiked samples or quality control samples with concentrations in the range of the level of interest. Acceptable recoveries for these congeners are between 70 and 120 %.

7. **Requirements for laboratories:**
In accordance with the provisions of Regulation (EC) No 882/2004, laboratories shall be accredited by a recognised body operating in accordance with ISO Guide 58 to ensure that they are applying analytical quality assurance. Laboratories shall be accredited following the EN ISO/IEC 17025 standard *[Commission Regulation (EC) No 1883/2006 of 19 December 2006, Annex II, No. 4].*

8. **Upper bound calculation:**
Calculation of results of the sum of the six indicator PCBs should be performed by the upper bound approach.
The concept of ‘upperbound’ requires using the limit of quantification (LOQ) for the contribution of each non-quantified congener to the sum of the six indicator PCBs. The concept of ‘lowerbound’ requires using zero for the contribution of each non-quantified congener to the sum of the six indicator PCBs *[Commission Regulation (EC) No 1883/2006 of 19 December 2006, Annex I, No. 5, footnote 2].*

9. **Criteria for the sum of the six indicator PCBs at the level of interest:**

| | |
|---|---|
| Trueness | - 30 to + 30 % |
| Intermediate precision (RSD%) | ≤ 20 % |
| Difference between upper and lower bound calculation | ≤ 20 % |

**References:**

[1] Commission Regulation (EC) No 1883/2006 of 19 December 2006 laying down methods of sampling and analysis for the official control of levels of dioxins and dioxin-like PCBs in certain foodstuffs (OJ L364, 20.12.2006, p. 32-43)

[2] Method Validation and Quality Control Procedures for Pesticide Residues Analysis in Food and Feed (Document N° SANCO/2007/3131 of 31/October/2007)

[3] Commission Decision of 12 August 2002 implementing Council Directive 96/23/EC concerning the performance of analytical methods and the interpretation of results (OJ L221, 17.8.2002, p. 8 - 36