EURL Proficiency Study on the Determination of PCDD/Fs, PCBs, PBDEs and HBCDDs in Dried Citrus Pulp 2021

EURL-PT-DPB_2105-DCP

FEED

Report

PCDD/Fs and PCBs

(Version 1.0)

04 April 2022

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Summary

Test sample	**FEED:** Dried Citrus Pulp [2105-DCP]
Analytes of interest	**PCDD/Fs** (17 2,3,7,8-substituted PCDD/Fs) **PCBs** (12 DL-PCBs, 6 NDL-PCBs) **PBDEs** (BDE-28, -47, -49, -99, -100, -153, -154, -183, -209) **HBCDDs** (α-HBCDD, β-HBCDD, γ-HBCDD or total HBCDD)
Methods	**PCDD/Fs, DL-PCBs:** GC-HRMS, GC-MS/MS and alternative methods; Bioanalytical screening methods **Indicator PCBs, PBDEs and HBCDDs:** Any kind of method
Participants	NRLs, OFLs, other official laboratories, commercial laboratories performing the analysis of samples taken by feed business operators
Statistical evaluation	ISO 13528:2020, IUPAC Protocol
Report of final results	04 April 2022
Publication	EURL POPs reserves all rights to publish and present the anonymised results of the interlaboratory study in scientific journals and/or during conferences.

1. Structure of the ILS, test material and analytes

This proficiency test (PT) on the determination of **PCDD/Fs, PCBs, PBDEs** and **HBCDDs** in **dried citrus pulp** was organized by the European Union Reference Laboratory (EURL) for halogenated persistent organic pollutants (POPs) in Feed and Food to be performed between September and November 2021. The objective was to assess analytical performance of laboratories and the interlaboratory comparability of results from analyses of PCDD/Fs, PCBs, PBDEs and HBCDDs in one sample of **dried citrus pulp.**

National Reference Laboratories (NRLs) for halogenated POPs in Feed and Food from EU member states were requested to participate as part of their work programme for 2021. NRLs were invited to encourage the participation of Official Laboratories (OFLs) from their member states as part of their duties following Article 101 of regulation (EU) 2017/625 of the European Parliament and of the Council of 15 March 2017. Furthermore, participation of OFLs allowed the extension of the data basis for calculation of assigned values and evaluation of results.

Other official laboratories and **commercial laboratories** performing the analysis of samples taken by feed business operators were invited to participate in this proficiency test. The evaluated results were discussed by representatives of European Commission, NRLs and the EURL at the COM/EURL/NRL workshop on 23 and 24 November 2021.

1.1. Samples and coding

The test sample was prepared from commercially available feed and with PCDD/Fs naturally contaminated material. Additionally it was fortified with analytes of interest using standards and technical mixtures of PCBs, PBDEs and HBCDDs.

Dried citrus pulp	**Sample no. 2105-DCP-xxx**

Each participant received about **80 g** of the test sample in a HDPE bottle.

1.2. Analytes of interest

Participants were requested to determine the following parameters:

- 17 2,3,7,8-substituted PCDD/Fs
- WHO-PCDD/F-TEQ (using WHO2005-TEF)
- 12 dioxin-like PCBs
- WHO-PCB-TEQ (using WHO2005-TEF)
- WHO-PCDD/F-PCB-TEQ (using WHO2005-TEF)
- Six non-dioxin-like PCBs (indicator PCBs): PCB 28, 52, 101, 138, 153, 180
- Sum of six non-dioxin-like PCBs (indicator PCBs)
- PCDD/F-PCB-BEQ, PCDD/F-BEQ and/or PCB-BEQ, if applicable (using bioanalytical screening methods)

1.3. Methods

One or more of the following **detection methods** could be applied:

- GC-HRMS-, GC-MS/MS-methods or other alternative methods for PCDD/Fs and dioxin-like PCBs
- Bioanalytical screening methods for PCDD/Fs and dioxin-like PCBs
- Any kind of method for indicator PCB

1.4. Coding of laboratories and confidentiality

The laboratory code of the participating laboratories will be kept confidential and will not be revealed to other participants.
For NRLs, the “Protocol for management of underperformance in comparative testing and/or lack of collaboration of National Reference Laboratories (NRLs) with Community reference laboratories (CRLs) activities” will be observed. The confidentiality of NRLs will be kept according to this protocol.
The confidentiality between NRLs and their OFLs will be kept unless a Member State initiated a cooperation between the NRL, OFLs and the EURL.

1.5. Results of PCDD/Fs and PCBs

1.5.1. Results of PCDD/Fs and PCBs determined by physico-chemical methods

Laboratories should

- use their own reference standards for identification and quantification,
- report results for each analyte,
- report the limit of quantification (LOQ), at least for each non-quantified analyte,
- report upper, middle and lower bound results for WHO-PCDD/F-PCB-TEQ, WHO-PCDD/F-TEQ, WHO-PCB-TEQ and sum of six indicator PCBs,
- report if sample exceeds respective EU maximum levels or action thresholds for WHO-PCDD/F-PCB-TEQ, WHO-PCDD/F-TEQ and/or WHO-PCB-TEQ or the maximum level for the sum of six indicator PCBs beyond reasonable doubt taking into account the measurement uncertainty,
- report the measurement uncertainty, applied for checking of compliance, for WHO-PCDD/F-PCB-TEQ, WHO-PCDD/F-TEQ, WHO-PCB-TEQ and the sum of six indicator PCBs,
- give method information and
- give information about the accreditation of the laboratory according to ISO/IEC 17025 *(for metrological traceability of consensus values of participants used as assigned values).*

Results had to be reported in **ng/kg, relative to a feed with a moisture content of 12 %**, for PCDD/Fs and dioxin-like PCBs, and in **µg/kg, relative to a feed with a moisture content of 12 %**, for indicator PCBs. TEQ-based results had to be calculated using the WHO-TEFs of 2005[1].

1.5.2. Results of PCDD/Fs and PCBs determined by bioanalytical screening methods

Laboratories should

- use their own reference standards,
- report if the samples are suspected to be noncompliant with EU legal limits and confirmation is required,
- report PCDD/F and/or PCB results in BEQ, if applicable,
- report the reporting limit, maximum level / action threshold, which the evaluation is based on, and the bioassay cut-off, if applicable,
- give method information
- and give information about the accreditation of the laboratory according to ISO/IEC 17025.

Results had to be reported in **ng BEQ/kg, relative to a feed with a moisture content of 12 %**, for PCDD/Fs and dioxin-like PCBs.

[1] [1] Martin van den Berg et al., The 2005 World Health Organization Re-evaluation of Human and Mammalian Toxic Equivalency Factors for Dioxins and Dioxin-like Compounds. Toxicological Sciences 93(2), 223-241 (2006)

2. Participating laboratories

This proficiency test was open for participation of:

- National Reference Laboratories (NRLs) of EU member states
- National Reference Laboratories of other European countries
- Official laboratories
- Commercial laboratories

83 laboratories registered for this proficiency test, reporting 80 sets of results for at least one parameter.

Table 1: Participating laboratories

Participating laboratories	Region	No. of participants
National Reference Laboratories	European Union Other Countries	26 2
Official Laboratories	European Union Other European Countries Africa Americas Asia Oceania	35 1 0 1 0 1
Commercial Laboratories	European Union Other European Countries Africa Americas Asia Oceania	14 0 0 2 1 0
	Total	**83**

2.1. Number of reported results

Table 2: Reported results for PCDD/F and PCB sum parameters and moisture content

Reported results	WHO-PCDD/F-PCB-TEQ	WHO-PCDD/F-TEQ	WHO-PCB-TEQ	Sum of six indicator PCBs	PCDD/F-PCB-BEQ [Bioanalytical screening methods]	Moisture content
All laboratories	62	64	62	71	4	76
NRLs	19	19	19	23	2	23

Table 3: Reported accreditation according to ISO/IEC 17025 by participants for PCDD/Fs and PCBs

Dried Citrus Pulp	PCDD/Fs, PCBs [Physico-chemical methods]	PCDD/Fs, PCBs [Bioanalytical screening methods]
yes	68	4
no	5	0

2.2. Detection methods

The following detection methods were applied:

- GC-HRMS-, GC-MS/MS-, GC-LRMS-methods for PCDD/Fs and non- ortho PCBs
- GC-HRMS-, GC-MS/MS-, GC-LRMS-, GC-ECD-methods for mono-ortho-PCBs and indicator PCBs
- Bioanalytical screening methods for PCDD/Fs and dioxin-like PCBs

Table 4: Overview of physico-chemical detection methods for PCDD/Fs and PCBs applied by participants

Detection methods	PCDD/Fs	non-ortho-PCBs	mono-ortho-PCBs	Indicator PCBs
HRMS	51	52	51	42
MS/MS	12	11	10	17
LRMS	2	2	2	7
ECD	0	0	0	2

3. Homogeneity and stability of the test material

The test for sufficient homogeneity was performed according to ISO 13528:2020 [2] and the International Harmonized Protocol for the Proficiency Testing of Analytical Chemistry Laboratories [1].

Therefore, 10 portions of the test samples 2105-DCP were analysed in duplicate for PCDD/Fs and PCBs. The test for sufficient homogeneity was performed for the sum parameters WHO-PCDD/F-PCB-TEQ, WHO-PCDD/F-TEQ, WHO-PCB-TEQ, the sum of six indicator PCBs and individual congeners. The test materials showed sufficient homogeneity for this proficiency test. The stability check of the analytes of interest applying room temperature storage was performed according to ISO 13528:2020 [2]. The test material showed sufficient stability for this proficiency test.

4. Determination of the assigned value

Statistical evaluation of the PT results was performed by the EURL for halogenated POPs in feed and food according to ISO 13528:2020 [2] and the International Harmonized Protocol for the Proficiency Testing of Analytical Chemistry Laboratories [1].

The determination of the assigned value was performed according [1] by estimating of the assigned value as the consensus of participants' results (using only results of physico-chemical methods). The Huber robust mean was taken as assigned value after excluding extreme outliers (outside the range of ± 50% of the median of all reported results) and examination of the distribution of the remaining results using histogram and Kernel density estimation, if necessary.

Assigned values were calculated for WHO-PCDD/F-PCB-TEQ, WHO-PCDD/F-TEQ, WHO-PCB-TEQ, the sum of six indicator PCBs and individual PCDD/F and PCB congeners (including limits of quantification (LOQs)), if possible. Additionally the median of all values was calculated.

For individual congeners (including LOQs) assigned values were only calculated according to the above mentioned procedure, if more than 2/3 of all results are above the LOQ and less than 1/3 of all results (including LOQs) are outside the range of ± 50% of the median of all reported results. Levels for individual congeners were only used for evaluation and calculation if these levels are equal to or above the LOQ; otherwise the LOQ was used instead.

Due to high variation of participants' results, no assigned values could be calculated for:

- 1,2,3,7,8,9-HxCDF
- PCB 169

Since there are no traceable reference values available, the assigned values in this PT were calculated based on the Huber robust mean of the participants' results. Therefore, the assigned values are only traceable to these submitted results. Additionally the results of all participants reporting results and the results of participants having accreditation according ISO/IEC 17025 were compared for PCDD/F and PCB sum parameters. No significant differences between the assigned values calculated for both data sets were observed (Table 5).

Table 5: Comparison of assigned values for all participants and participants with reported accreditation according to ISO/IEC 17025 for PCDD/F and PCB sum parameters in dried citrus pulp 2105-DCP

Sum parameters	Assigned value All participants	Assigned value ISO/IEC 17025 accreditation	Deviation
	ng/kg, µg/kg product (12 % moisture content)	ng/kg, µg/kg product (12 % moisture content)	%
WHO-PCDD/F-PCB-TEQ ub rep	0.980	0.974	<1
WHO-PCDD/F-TEQ ub rep	0.513	0.508	1
WHO-PCB-TEQ ub rep	0.462	0.461	<1
Sum Indicator PCBs ub rep	10.7	10.6	<1

4.1. PCDD/Fs and PCBs – Sum parameters

The assigned values for the test sample 2105-DCP were calculated as consensus of participants' results for the PCDD/F and PCB sum parameters, taking into account the calculation criteria described above.

Table 6: Assigned values for physico-chemical methods for PCDD/Fs and PCBs (rounded to three significant figures)

Test sample	WHO-PCDD/F-PCB-TEQ (ub)	WHO-PCDD/F-TEQ (ub)	WHO-PCB-TEQ (ub)	Sum Indicator PCBs (ub)
	ng/kg product (12 % moisture content)			µg/kg product (12 % moisture content)
Dried Citrus Pulp (2105-DCP)	0.980	0.513	0.462	10.7

Table 7: Assigned values for PCDD/Fs and DL-PCBs for comparison with BEQ results of bioanalytical screening methods (rounded to two significant figures)

Test sample	WHO-PCDD/F-PCB-TEQ (ub)	WHO-PCDD/F-TEQ (ub)	WHO-PCB-TEQ (ub)
	ng/kg product (12 % moisture content)		
Dried Citrus Pulp (2105-DCP)	0.98	0.51	0.46

4.2. PCDD/Fs and PCBs – Individual congeners

The assigned values for the test sample 2105-DCP for individual congeners were calculated as a consensus of the participants' results, taking into account the calculation criteria described above (Figure 1; tabular summary see annex 1).

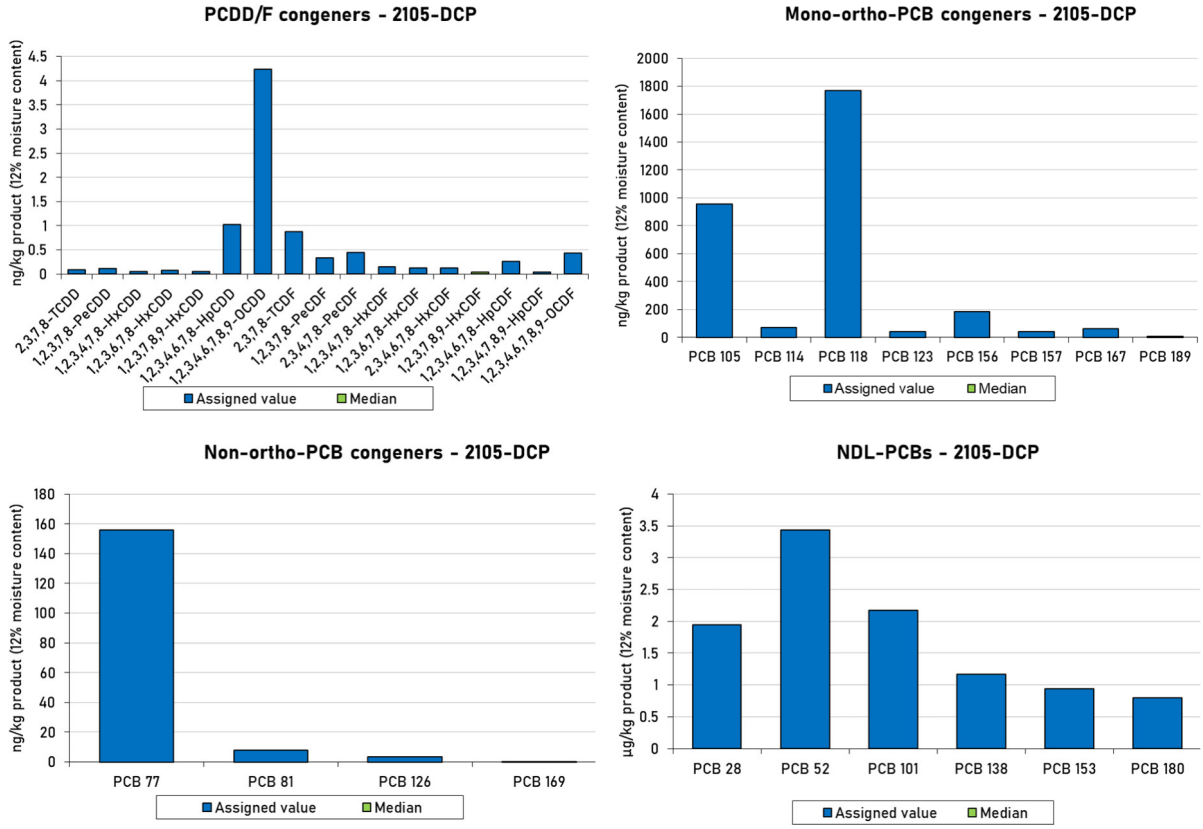

Figure 1: Assigned values (blue) and median values (green) for PCDD/F and PCB congeners for dried citrus pulp (2105-DCP) [ng/kg or µg/kg product (12% moisture content)]

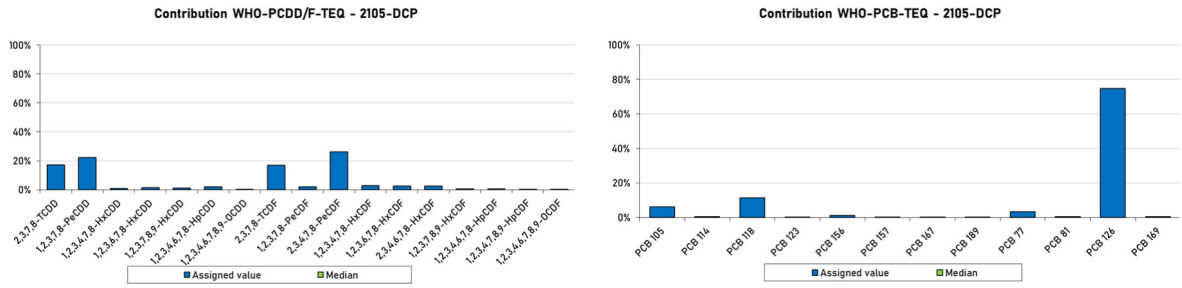

Figure 2: Contributions in % to WHO-PCDD/F-TEQ and WHO-PCB-TEQ for PCDD/F and PCB assigned (blue) and median (green) values for dried citrus pulp (2105-DCP)

4.3. Moisture content

For the moisture content an assigned value of 8.71 % for the test sample 2105-DCP was calculated as a consensus of the participants' results, taking into account the calculation criteria described above. 10 labs reported values for **dry matter** instead of **moisture content.**

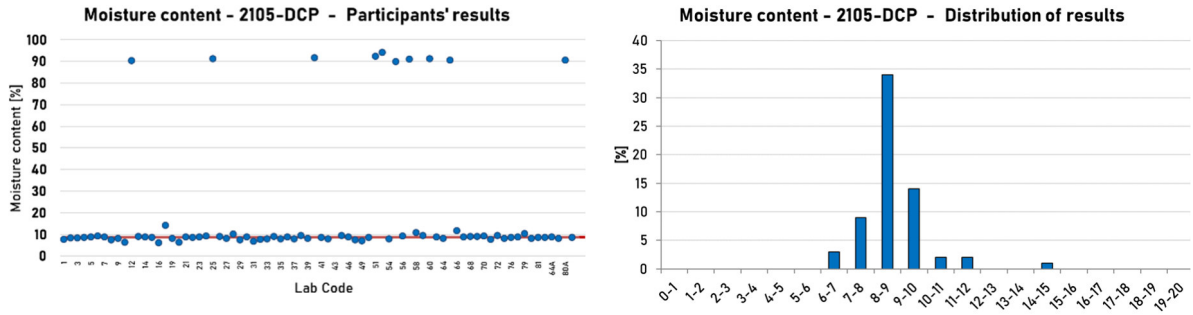

Figure 3: Participant's results (red line assigned value) and distribution of participant's results of the moisture content in % for dried citrus pulp (2105-DCP)

4.4. Comparison of assigned values with legal limits

Maximum levels and action thresholds for feed are defined in the Directive 2002/32/EC of the European Parliament and of the Council of 7 May 2002 on undesirable substances in animal feed (Annex I and II).

Table 8: Maximum levels and action thresholds for feed materials of plant origin (Annex I and II):

Undesirable Substances **Feed materials of plant origin**		**Maximum level**	**Action threshold**
WHO-PCDD/F-PCB-TEQ	ng/kg product*	**1.25**	
WHO-PCDD/F-TEQ	ng/kg product*	**0.75**	**0.5**
WHO-PCB-TEQ	ng/kg product*		**0.35**
Sum of 6 NDL PCBs (sum of PCB 28, 52, 101, 138, 153, 180)	µg/kg product*	**10**	

* *relative to a feed with a moisture content of 12 %*

For the dried citrus pulp test sample 2105-DCP the assigned values for the sum parameters WHO-PCDD/F-PCB-TEQ, WHO-PCDD/F-TEQ and the sum of indicator PCBs were the range of 0.5 to 4 of the respective maximum levels and action thresholds (Figure 4).

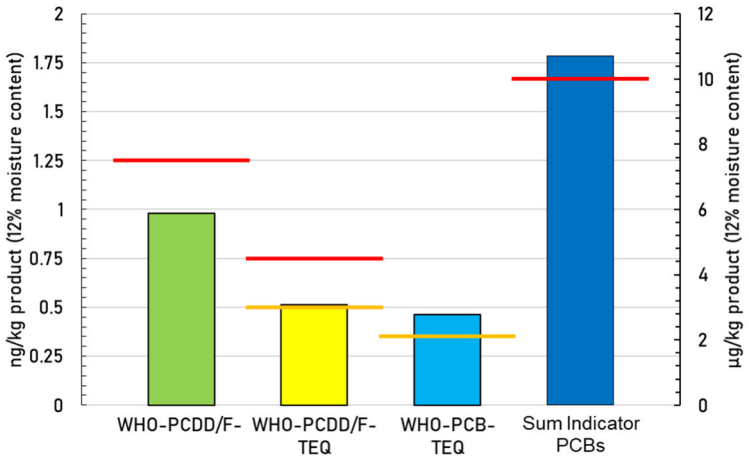

Figure 4: Comparison of the assigned values for sum parameters for dried citrus pulp (2105-DCP) with maximum levels (red lines) and action thresholds (yellow line) [ng/kg and µg/kg product (12% moisture content)]

5. Evaluation of results

5.1. Participants' results for physico-chemical methods

5.1.1. Z-score calculation

Criteria for successful participation of laboratories using physico-chemical methods were based on the evaluation of the results of the sum parameters WHO-PCDD/F-TEQ, WHO-PCB-TEQ, WHO-PCDD/F-PCB-TEQ and the sum of six indicator PCBs and evaluated individual congeners. The criteria will be applicable for sum parameter concentrations in the range (about 0.5 to 4 times) of the level of interest (maximum level or action threshold).

For evaluation of results of physico-chemical methods the z-scores were calculated according to the following formula:

$$z = \frac{(x - x_a)}{\sigma_p}$$

x: *participant's result*
x_a: *assigned value*
σ_p: *fitness-for-purpose-based standard deviation for proficiency assessment*

For WHO-PCDD/F-TEQ, WHO-PCB-TEQ and WHO-PCDD/F-PCB-TEQ the standard deviation for proficiency assessment σ_p was defined as 10 %, for the sum of six indicator PCBs (PCB 28, 52, 101, 138, 153, 180) as 15 % and for evaluated individual PCDD/F, PCB as 20 %.

Z-scores for individual congeners were only calculated and reported if levels for these congeners are equal to or above the LOQ. Otherwise no z-scores will be given.

Interpretation of z-scores:

$\lvert z\text{-score} \rvert \leq 2$	*satisfactory performance*
$2 < \lvert z\text{-score} \rvert < 3$	*questionable performance (warning signal)*
$\lvert z\text{-score} \rvert \geq 3$	*unsatisfactory performance (action signal)*

5.1.2. PCDD/Fs and PCBs - Participants' z-scores

The concentrations of the sum parameters for the test samples 2105-DCP were in the range (about 0.5 to 4 times) of the respective maximum levels (tabular summaries of participants' results and z-scores see annex 2 and 3).

Table 9: Distribution of all participants' and NRLs only z-scores for sum parameters

Dried citrus pulp (2105-DCP)	WHO-PCDD/F-PCB-TEQ	WHO-PCDD/F-TEQ	WHO-PCB-TEQ	Sum of six indicator PCBs
all Participants				
|z-score| ≤ 2	89 %	77 %	92 %	93 %
2 < |z-score| < 3	6 %	15 %	2 %	7 %
|z-score| ≥ 3	5 %	8 %	6 %	0 %
NRLs				
|z-score| ≤ 2	79 %	68 %	89 %	83 %
2 < |z-score| < 3	16 %	21 %	0 %	17 %
|z-score| ≥ 3	5 %	11 %	11 %	0 %

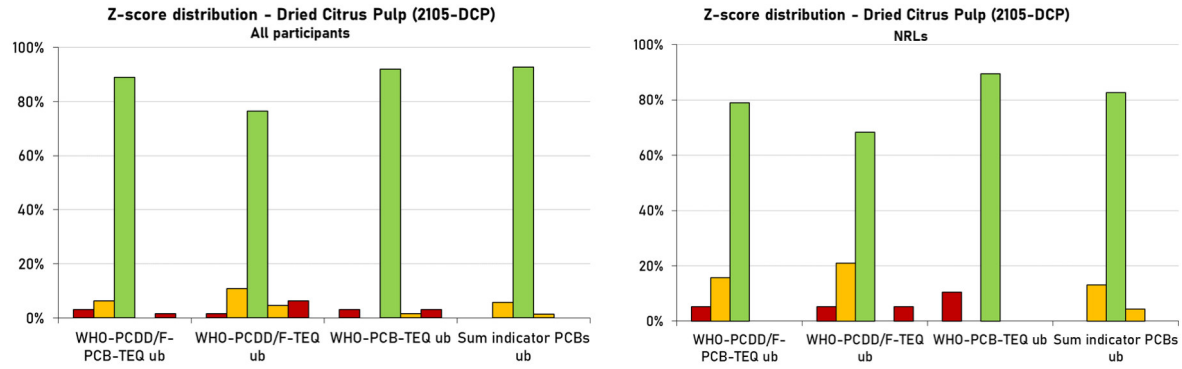

Figure 5: Distribution of all participants' z-scores and NRLs only for sum parameters for dried citrus pulp (2105-DCP) [Green bars: -2 ≤ z-score ≤ 2, orange bars: -3 < z-score < -2, 2 < z-score < 3, red bars: z-score ≤ -3, z-score ≥ 3]

5.1.3. Comparison of reported and calculated sum parameters

In addition to the calculation of the sum parameters for reported individual PCDD/F and PCB congener values, the calculated sum parameters for PCDD/Fs and PCBs by the EURL were compared with the ones reported by each participant. As the reported sum parameters are decisive to compare the results with the legal limits, an incorrect calculation might lead to a wrong assessment of a sample. In case of a significant deviation of the reported sum parameter value from the (EURL) calculated one (deviation > 10 %) the laboratory has therefore not successfully participated in the PT according to the positive scoring system (see 5.1.5).

Table 10: Difference between reported and calculated sum parameters for PCDD/Fs and PCBs for dried citrus pulp (2105-DCP) given in percentage of participants' results

Dried Citrus Pulp (2105-DCP)	WHO-PCDD/F-PCB-TEQ	WHO-PCDD/F-TEQ	WHO-PCB-TEQ	Sum of six indicator PCBs
Deviation ≤ 10 %	98%	98%	98%	99%
Deviation > 10 %	2%	2%	2%	1%

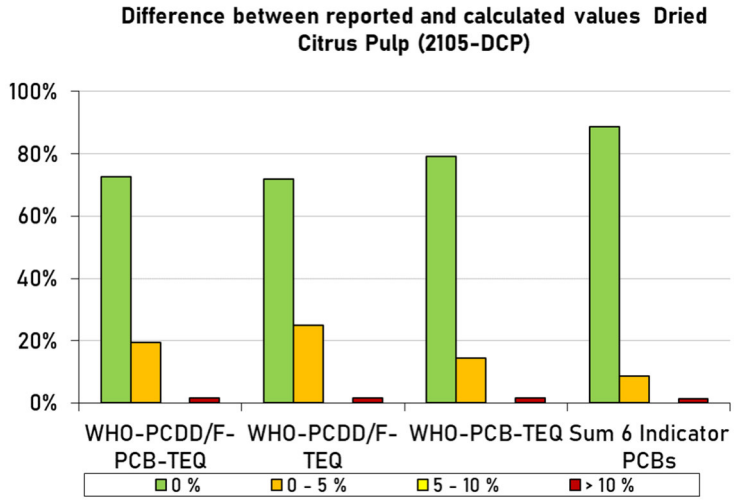

Figure 6: Difference between reported and calculated sum parameters for PCDD/Fs and PCBs [Green bars: 0 %, orange bars: 0-5 %, yellow bars: 5-10 %, red bars: > 10 %] for dried citrus pulp (2105-DCP) given in percentage of participants' results

5.1.4. Difference between upper and lower bound calculation

According to Commission Regulation (EC) No 152/2009 the difference between upper bound level and lower bound level shall not exceed 20 % for confirmation of exceedance of maximum level or in case of need of action thresholds for PCDD/Fs and DL-PCBs. For indicator PCBs the difference between upper bound and lower bound levels for the sum of six indicator PCBs shall be ≤ 20 % at the level of interest. Participants with a larger deviation should review their analytical methods, especially with regard to sensitivity and limit of quantification.

For the test samples 2105-DCP the assigned values for all sum parameters were below the respective maximum levels.

Table 11: Difference between upper and lower bound calculation for dried citrus pulp (2105-DCP) given in percentage of participants' results

Dried Citrus Pulp (2105-DCP)	WHO-PCDD/F-PCB-TEQ	WHO-PCDD/F-TEQ	WHO-PCB-TEQ	Sum of six indicator PCBs
0 – 10 %*	90%	87%	98%	100%
10 – 20 %*	8%	8%	2%	0%
20 – 50 %*	2%	3%	0%	0%
> 50 %*	0%	2%	0%	0%

* Difference between upper and lower bound calculation

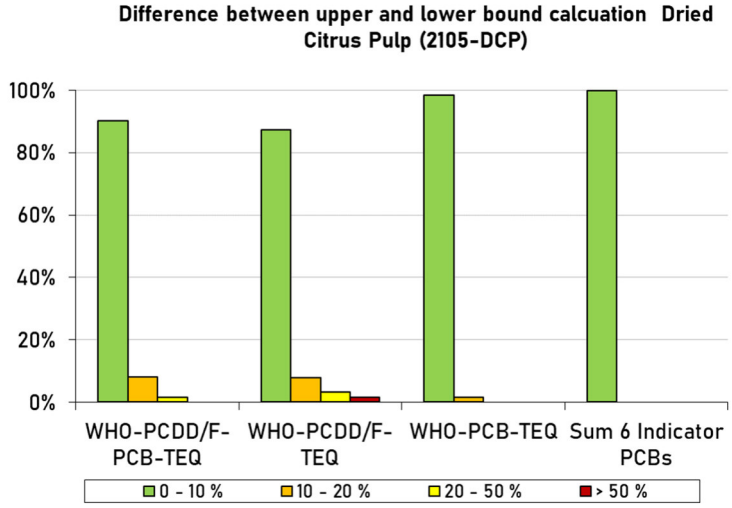

Figure 7: Difference between upper and lower bound calculation for dried citrus pulp (2105-DCP) given in percentage of participants' results [Green bars: 0 – 10 %, orange bars: 10 – 20 %, yellow bars: 20 – 50 %, red bars: > 50 %]

5.1.5. Positive scoring system

The “positive scoring system” gives one assessment for each PT sample covering all relevant PCDD/F and PCB sum parameters and congeners.

The total score for the positive scoring system was calculated according to the following general principles:

- Calculation of z-scores for sum parameters and evaluated individual congeners
- Calculation of the positive scores according to the following table:

Positive scoring system	I z-score I ≤ 2	2 < I z-score I < 3	I z-score I ≥ 3
Individual congeners	**Positive score**	**Positive score**	**Positive score**
Contribution to sum parameter* > 10 %	12	6	0
Contribution to sum parameter* 3-10 %	8	4	0
Contribution to sum parameter* < 3 %	6	3	0
Not evaluated congeners	0	0	0

* separately for the respective sum parameters WHO-PCDD/F-TEQ, WHO-PCB-TEQ and the sum of six indicator PCBs

- Calculation of maximum achievable scores (I z-score I ≤ 2) for PCDD/F and DL-PCB and indicator PCB congeners separately:

$$Maximum\ Score = \sum_{i=1}^{n} Max.Score_{(>10\%)i} + \sum_{i=1}^{m} Max.Score_{(3-10\%)i} + \sum_{i=1}^{p} Max.Score_{(<3\ \%)i}$$

- Calculation of the participant’s scores for PCDD/F and DL-PCB and indicator PCB congeners separately:

$$Participant's\ Score = \sum_{i=1}^{n} Score_{(>10\%)i} + \sum_{i=1}^{m} Score_{(3-10\%)i} + \sum_{i=1}^{p} Score_{(<3\ \%)i}$$

- Calculation of achieved scoring percentage for each participant:

$$Participant's\ Scoring\ Percentage = \frac{Participant's\ score}{Maximum\ score} \cdot 100$$

- Criteria for successful participation:

Sum parameters:	≤ 1 parameter with | z-score | > 2, no parameter with | z-score | ≥ 3
PCDD/F congeners:	≥ 75 % of maximum score
DL-PCB congeners:	≥ 75 % of maximum score
Indicator PCB congeners:	≥ 75 % of maximum score
Difference between reported and calculated results for sum parameters	≤ 10 %

The assessment based on the positive scoring system was performed for the PT test sample dried citrus pulp 2105-DCP. A laboratory participated successfully in an EURL PT for PCDD/Fs and PCBs, if all above mentioned criteria for the reported analytes are met.

Table 12: Successful participation rate according to positive scoring system for dried citrus pulp (2105-DCP)

Scoring system	Successful participation		Reason for non-successful participation		
2105-DCP	yes	no	Sum parameters	Individual congeners	Calculation of sum parameters
Percentage of participants' results	74%	26%	65%	40%	20%

5.2. Participants' results for bioanalytical screening methods

According to Commission Regulation (EC) No 152/2009, "a screening method in principle classifies a sample as compliant or suspected to be non-compliant. For this, the calculated BEQ level is compared to the cut-off value […]. Samples below the cut-off value are declared compliant, samples equal or above the cut-off value as suspected to be non-compliant, requiring analysis by a confirmatory method".

Therefore, the main criterion for evaluation of results from bioanalytical screening methods is their ability to reliably identify compliant samples and samples suspected to be non-compliant with established legal limits.

For further evaluation of the performance of bioanalytical screening methods, bioassay-scores are applied: The reported BEQ-values derived from bioanalytical screening methods are compared with the WHO-TEQ assigned values calculated on basis of the results of physical-chemical methods for the concentration range of 0.5 to 2 times the level of interest.

Because bioanalytical screening methods focus mainly on distinguishing between compliant and potentially non-compliant samples, a direct comparison of bioassay-scores and z-scores is not possible. However, bioassay scores may serve as a tool to assess method performance within the scope of external quality control measures of the respective laboratory.

Bioassay-scores are calculated according to the following formula:

$$bioassay - score = \frac{(x - x_a)}{\sigma_{bioassay}}$$

x: *participant's result (BEQ from bioanalytical screening method)*
x_a: *assigned value (physical-chemical methods)*
$\sigma_{bioassay}$: *bioassay target deviation*

For PCDD/F-BEQ, PCB-BEQ and PCDD/F-PCB-BEQ the bioassay target deviation $\sigma_{Bioassay}$ was defined as 20%.

5.2.1. Assessment of analytical results

As a consequence of the comparison of the assigned values of the test sample 2105-DCP with legal limits, the assessment of the analytical results using bioanalytical screening methods should read "compliant with the maximum level for WHO-PCDD/F-PCB-TEQ and WHO-PCDD/F-TEQ", "suspected to be non-compliant with the action threshold for WHO-PCB-TEQ" and "compliant with the action threshold for WHO-PCDD/F-TEQ".

Table 13: Evaluation of assigned values for dried citrus pulp

	WHO-PCDD/F-PCB-TEQ	WHO-PCDD/F-TEQ	WHO-PCDD/F-TEQ	WHO-PCB-TEQ
2105-DCP	< ML	< ML	< AL	> AL

Four laboratories reported results using CALUX bioassay for Total-BEQ and hereof three also for PCDD/F-BEQ and/or PCB-BEQ.

Table 14: Participants' assessment of analytical results using bioanalytical screening methods for 2105-DCP

Laboratories' assessment of analytical results	WHO-PCDD/F-PCB-TEQ Maximum level	WHO-PCDD/F-TEQ Maximum level	WHO-PCDD/F-TEQ Action threshold	WHO-PCB-TEQ Action threshold
Suspected to be non-compliant	1	2	2	2
Compliant	3	1	1	1

5.2.2. Participants' bioassay-scores

Concentrations for WHO-PCDD/F-PCB-TEQ and WHO-PCDD/F-TEQ in the test sample 2105-DCP are in the range (about 0.5 to 2 times) of the respective maximum levels.

Table 15: Distribution of participants' bioassay-scores for BEQ parameters for dried citrus pulp (2105-DCP)

Percentage of participants' results	PCDD/F-PCB-BEQ	PCDD/F-BEQ	PCB-BEQ
| **bioassay-score** | ≤ 2	75 %	50%	100 %
2 < | **bioassay-score** | < 3	-	-	-
| **bioassay-score** | ≥ 3	25 %	50 %	-

6. Participants' feedback

A questionnaire for feedback from participants of this EURL proficiency test was available as online survey between 03 December 2021 and 21 January 2022. The survey was anonymous, but participants could also give their laboratory name. The identity of the laboratories is kept confidential. The survey included several questions related to different topics (participants' information, organization of the proficiency test, PT test samples and evaluation of results and summary of data) and a possibility to include comments and further suggestions.

In total, 12 laboratories (15 % of all PT participants) replied to this survey.

Table 16: Participating laboratories in the feedback survey

Type of laboratory	Answers
National Reference Laboratory (NRL)	6
Official Laboratory (OFL)	3
Commercial laboratory	3
Other (e.g. research and development)	0
No Answer	0

General aspect

How satisfied are you with the organization of this proficiency test in general? Please rate the parts below according to your experience, with 0 stars meaning "no opinion" and 5 stars meaning "full satisfaction".

Announcement	
Instructions	
Sample shipment	
Reporting of results	
Preliminary report	

Did the proficiency test meet expectations?

Specific aspects of this proficiency test

We would like to know a bit more about specific aspects of this proficiency test. Please rate the aspects below according to your experience, with 0 stars meaning "no opinion" and 5 stars meaning "full satisfaction".

Was all necessary information for participation and performance of the PT provided in an understandable way?	
Was the time frame acceptable?	
Was the handling of EUSurvey as webtool for reporting and source of instructions manageable?	
Was the evaluation of participant's results and the information in the preliminary report clear and comprehensible?	

Was the selected sample adequate for the goal to assess analytical performance of laboratories in relevant matrices?

Choice of matrix	
Level of contamination	

The following comments or suggestions for improvements were submitted:
"The matrix did not correspond to the routinely tested samples. It was the first citrus pulp sample past 10 years."

7. Quality control

The Deutsche Akkreditierungsstelle GmbH attests that the provider of proficiency testing Chemisches und Veterinäruntersuchungsamt Freiburg, EU Reference Laboratory (EURL) for halogenated persistent organic pollutants (POPs) in feed and food is competent under the terms of DIN EN ISO/IEC 17043:2010 to carry out proficiency testing in the testing field of determination of halogenated persistent organic pollutants (POPs) in food and feed (Accreditation number: D-EP-18625-01-00).

8. Results of participants

An overview of the PCDD/F and PCB results for the PT test sample dried citrus pulp (2105-DCP) is given in the following annexes. Laboratories are coded according to the laboratory codes sent after registration.

9. References

[1] M. Thompson, S.L.R. Ellison, R. Wood: The International Harmonized Protocol For The Proficiency Testing Of Analytical Chemistry Laboratories, Pure Appl. Chem., Vol. 78, No. 1, pp. 145-196, 2006.

[2] ISO 13528:2020, Statistical methods for use in proficiency testing by interlaboratory comparisons, International Organization for Standardization

9. Annex

(Please double click on the pdf-icons to open the annexes.)

Dried Citrus Pulp – 2105-DCP		
1	Assigned values – PCDD/F, PCB	
2	Participants' results – Tables – PCDD/F, PCB	
3	Participants' z-scores / bioassay-scores – Tables – PCDD/F, PCB	
4	Participants' z-scores – Charts – PCDD/F, PCB	
5	Scoring system – PCDD/F, PCB	
6	Homogeneity and stability test – PCDD/F, PCB	
7	Participants' methods – PCDD/F, PCB	

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