RELIABILITY OF RESULTS OF LABORATORIES FOR SELF-CONTROL IN THE FEED AND FOOD BUSINESS WITH ACCREDITATION ACCORDING TO EN 17025 BUT NOT FOLLOWING SPECIFIC EU REGULATIONS

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1 Introduction

Self-control of industry is an important pillar for safety of feed and food. The question was raised if accreditation according to the EN 17025 standard is sufficient to guarantee a high quality of the results or if Commission Regulation (EU) No 589/2014 (1) and Commission Regulation (EU) No 709/2014 (2) are binding also for self-control of industry. Commission Regulation (EC) No 882/2004 of the European Parliament and of the Council of 29 April 2004 on official controls performed to ensure the verification of compliance with feed and food law, animal health and animal welfare rules, in particular Article 11(4) thereof (with Article 11: Methods of sampling and analysis; sampling and analysis methods used in the context of official controls shall comply with relevant Community rules …) forms the basis for that. Self-control of industry falls within regulatory purposes. For a scientific discussion, this document on requirements „EN 17025 versus COM REG 589/2014 resp. 709/2014“ was prepared showing essential deficiencies if laboratories, beyond being accredited according to the EN 17025 standard do not comply with the above mentioned Commission Regulations.

2 Materials and methods: Requirements of EN 17025 and EU Regulations

2.1 DIN EN ISO/IEC 17025:2005-08 *"*General requirements for the competence of testing and calibration laboratories" specifies parameters to be checked in a rather general way, focusing on:

- management (organization, management system, documentation, contracting, service to customers, complaints, improvement…)
- general technical requirements (qualification of personnel, infrastructure)
- selection and validation of methods *(excerpt of most relevant aspects)*:
  - The hierarchy of analytical procedures to be followed in an accredited lab is prescribed: Preferably methods published in international, regional or national standards shall be used. Non-standard methods shall be subject to agreement with the customer and must be validated appropriately before use.
  - The requirements for **validation** read *(5.4.5 Validation of methods)*:

> The laboratory shall validate non-standard methods, laboratory-designed/developed methods, standard methods used outside their intended scope, and amplifications and modifications of standard methods to confirm that the methods are fit for the intended use. The validation shall be as extensive as is necessary to meet the needs of the given application or field of application. The laboratory shall record the results obtained, the procedure used for the validation, and a statement as to whether the method is fit for the intended use.
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> The range and accuracy of the values obtainable from validated methods (e.g. the uncertainty of the results, detection limit, selectivity of the method, linearity, limit of repeatability and/or reproducibility, robustness against external influences and/or cross-sensitivity against interference from the matrix of the sample/test object), as assessed for the intended use, shall be relevant to the customers' needs.

As a result, the correct use of methods must be demonstrated, **but no explicit analytical requirements for validation are prescribed.**

- The requirements for **measurement uncertainty** read *(5.4.6 Estimation of uncertainty of measurement):*

> A calibration laboratory, or a testing laboratory performing its own calibrations, shall have and shall apply a procedure to estimate the uncertainty of measurement for all calibrations and types of calibrations. Testing laboratories shall have and shall apply procedures for estimating uncertainty of measurement. In certain cases the nature of the test method may preclude rigorous, metrologically and statistically valid, calculation of uncertainty of measurement. In these cases the laboratory shall at least attempt to identify all the components of uncertainty and make a reasonable estimation, and shall ensure that the form of reporting of the result does not give a wrong impression of the uncertainty. Reasonable estimation shall be based on knowledge of the performance of the method and on the measurement scope and shall make use of, for example, previous experience and validation data.
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> When estimating the uncertainty of measurement, all uncertainty components which are of importance in the given situation shall be taken into account using appropriate methods of analysis.

As a result, measurement uncertainty must be derived, **but no explicit analytical requirements and specifications are prescribed.**

2.2 In contrast, **Commission Regulations (EU) No 589/2014 (for food) and 709/2014 (for feed)** prescribe **parameters to be checked plus their specifications**. As a basic requirement of these Regulations, laboratories shall be accredited following the EN 17025 standard. In addition, they specify screening and confirmatory methods and set specific requirements for their application as follows (*selection of important criteria*):

- For **screening methods**, most important is a **false-negative rate below 5 %**. To meet this requirement, a number of specific criteria and constraints are set, the most significant of which are:
  - Cut off-value: It is described in detail how cut-off concentrations must be established for checking of possible exceedance of action and maximum levels, separately for dioxins, dioxin-like PCBs and/or the sum parameter,
  - Use of blank and positive control samples (for verification and correction),
  - Repeatability (< 20 %),
  - Within-laboratory reproducibility (< 25 %),
  - Bioassay apparent recoveries, for PCDD/F: 50 – 130 %, for dl-PCB: 20 – 60 %,
  - Details for establishing a calibration curve with TCDD and/or PCB 126
  - Initial Validation: method performance (matrix-related) must be demonstrated before use,
  - Close cooperation with laboratories applying confirmatory methods,
  - Check of possible suppression of the cell response and cytotoxicity in 20 % of the samples,
  - Check of the results of 2 to 10 % of the compliant samples (minimum of 20 samples per matrix) with confirmatory methods,
  - Quality control on suspected samples: to be checked by confirmatory analysis
  - Determination of false-compliant rates (based on $\geq$ 20 confirmed samples per matrix)
  - Expression of results from bioanalytical screening methods as BEQs, determination of BEQ/TEQ ratio for each matrix as important link between BEQ-based results and TEQ-based maximum and action levels,
  - Reporting level.

- For **confirmatory methods for PCDD/F and dl-PCB,** the most important requirements and criteria are:
  - Use of all relevant ^{13}C-labelled PCDD/F and dl-PCB as internal standards for control of all native congeners of interest in all samples with specified acceptable recoveries and specification of stage of addition;
  - Acceptable trueness between +/- 20 %;
  - Within-laboratory reproducibility of < 15 %;
  - A number of specific criteria for HRMS and MS/MS determination with regard to gas-chromatographic separation, and MS resolution,
  - Acceptable differences between upperbound and lowerbound WHO-TEQ-levels (not exceeding 20 % for control of maximum or action levels),
  - Limit of quantification.

- Similar requirements and criteria are set for **methods for ndl-PCBs.**
- Furthermore, important details on **compliance** are fixed:
  - **Measurement Uncertainty** is derived by calculation of the expanded measurement uncertainty (using a coverage factor of 2) or by establishing the decision limit (CC alpha). For compliance, the measured value minus U must be below the maximum level.
  - **Duplicate analysis** is necessary in cases of non-compliance with specified details.

3 Conclusions

The general requirements of EN 17025 form the basis for laboratory accreditation on which the specific requirements of COM Regulations 589/2014 and 709/2014 are built. Comparability and reliability of results of methods for determination of dioxins, dl-PCBs and ndl-PCBs in feed and food are, therefore, only ensured if the specific requirements of the "lex specialis" are met. **Risk of false-compliant results:** If the criteria and procedures laid down in COM Regulations (EU) 589/2014 and 709/2014 are not followed during screening and confirmatory analyses, underestimation and false-compliant results may be very likely to occur, mainly based on insufficient recovery and/or precision.

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References:

1. Commission Regulation (EU) No 589/2014 of 2 June 2014 laying down methods of sampling and analysis for the control of levels of dioxins, dioxin-like PCBs and non-dioxin-like PCBs in certain foodstuffs and repealing Regulation (EU) No 252/2012. Official Journal of the European Union, L 164/18
2. Commission Regulation (EU) No 709/2014 of 20 June 2014 amending Regulation (EC) No 152/2009 as regards the determination of the levels of dioxins and polychlorinated biphenyls. Official Journal of the European Union, L 188/1