6 The Dioxin function

6.1 Summary

In 1980, Dr William Horwitz and its collaborators published an evaluation of hundreds of performance interlaboratory studies that led them to conclude that there is a fixed relationship between analyte level (c) and s_R whatever the kind of analyte, matrix or analytical method (Horwitz et al., 1980; Horwitz, 1982). It takes the form of RSD_R (%) = $2^{(1-0.5\log C)}$. The relationship is widely accepted but is less appropriate at concentrations lower than about 10 ppb (Thompson, 2000). We revisit the application range of the Horwitz equation for dioxins and DL-PCBs in food and feed and we propose a new empirical function called the *'the Dioxin function'* adapted to ultra-trace analysis of these contaminants in biological samples.

The foregoing discussion in chapter 5 already pointed out the fact that when s_r and s_R standard deviations of PCDD/Fs and DL-PCBs were plotted against their corresponding mean pg g^{-1} result on a log-log scale, striking linear functions were already observed in animal feedingstuffs. The slopes of the lines were statistically not significantly different, i.e. they could be considered as parallel. In addition, whatever the toxic congener analyzed (i.e. data point), we concluded that the GC-ID-HRMS method's precision was clearly independent of the concentration (r^2 >0.98) over a wide range from sub-ppt to ppb levels (Eppe et al., 2004). We wanted to bear out these preliminary results with more data and matrices in order to establish a general relationship. With the European project called DIFFERENCE, we had a remarkable opportunity to complete this work initiated a few years before. A restricted number of expert laboratories with long-standing experience in this field were invited to evaluate five possible certified reference materials for dioxins and DL-PCBs in food and feed. All the raw data were kindly provided by the work package leader. After statistical treatment of data according to ISO 5725-2, striking linear functions in log scale between reproducibility standard deviation and congener's level over a concentration range of 10^{-8} to 10^{-14} g g^{-1} fresh weight were observed. The data fit very well to a Horwitz-type function of the form s_R = $0.153c^{0.904}$, where s_R and c are dimensionless mass ratios expressed in pg g^{-1} on fresh weight, regardless of the nature of the toxic congeners, food and feed matrices, or sample preparation methods.

The dioxin function used in the context of analytical quality assurance (AQA) management

The terms validation and quality assurance (QA) are widely used. However, many analysts and laboratories do not know the exact meaning neither the difference nor the relationship between the two terms. Validating a method is investigating whether the analytical purpose of the method is achieved, which is obtaining analytical results with an acceptable uncertainty level. Analytical method validation forms the first level of QA in the laboratory (Figure 6-1). AQA is the complete set of measures a laboratory must undertake to ensure that it can always achieve high-quality data. Besides the use of validated methods, these measures are: effective internal quality control (IQC) procedures (use of reference materials, control charts, see chapter 7): participation at relevant proficiency testing (PT) schemes: and, accreditation to an international standard ISO/IEC 17025 (Tavernier et al., 2004).

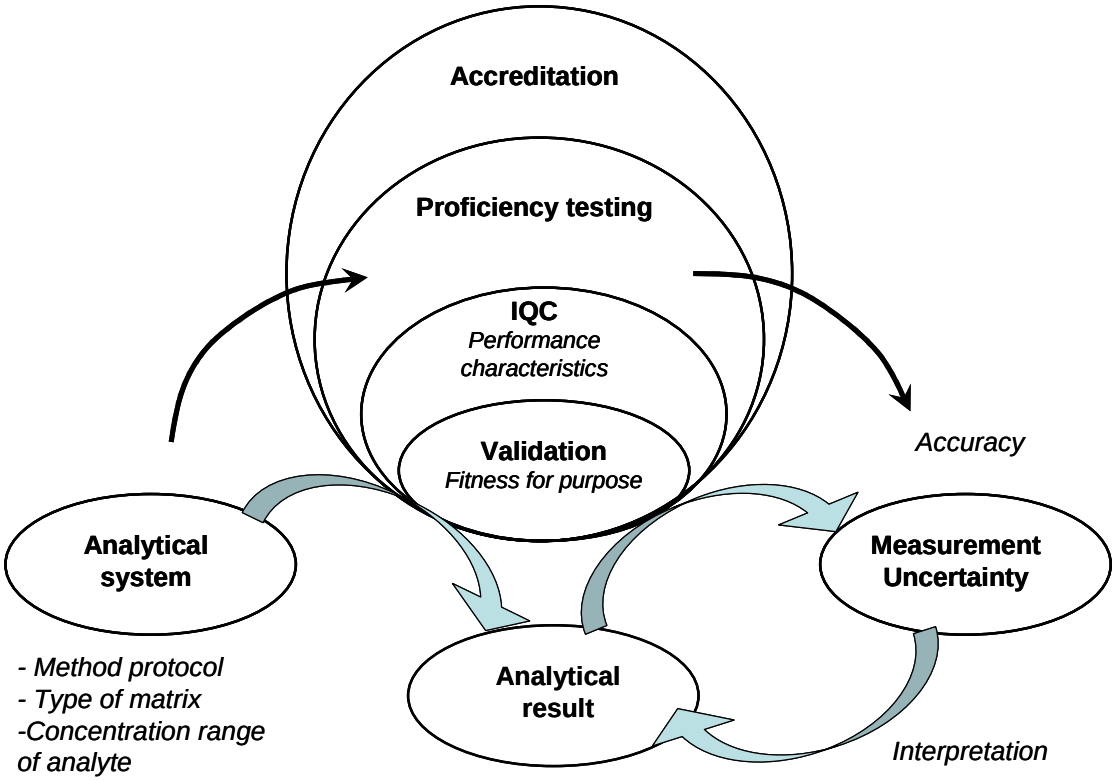

Figure 6-1: Different levels of QA measurements for analytical chemistry in food laboratories (Figure from Tavernier et al., 2004).

One of the main features of the dioxin function could be its use as a suitable fitness-for-purpose criterion for dioxins and related compounds in PT exercises.

Participation in PT schemes is a useful way for assessing the quality of a laboratory's results. In chemical analysis, the statistical evaluation of PT results is based on the scoring system as recommended in the International Harmonized Protocol (Thompson et al., 2006). In this system, the participant's result is converted into a z-score that gives a valuable indication of the performance of the laboratory. The z-scores are calculated as follows:

$$z = \frac{x - X}{\sigma_p}$$

Where x: lab result; X: assigned value, σ_p target value for the standard deviation.

In fact, a z-value is nothing more than the estimate of the error in the result scaled in standard deviation units (σ_p).

In order to properly estimate the performances of a laboratory participating in a PT exercise, we need objective criteria based on sound statistical data. By providing target standard deviation values for scoring systems, the dioxin function could be a useful tool for this purpose. We will illustrate its use with a practical example with data from the largest international PT in this field.

The dioxin function as benchmark precision values for internal validation

Another remarkable aspect of the dioxin function is its use as benchmark precision (repeatability and reproducibility) for internal validation purposes. In practice, internal method validation is done by evaluating a series of method-performance parameters such as precision, trueness, linearity, selectivity, working range, recovery, limit of detection, limit of quantification, robustness. Among these, precision is generally assessed by using reference materials/certified reference materials or spiked materials sufficiently homogeneous and stable to perform these tests. The level should be as close as possible to the level of interest. Repeatability precision (RSD_r) and within-laboratory reproducibility or intermediate precision (RSD) studies are carried out on these materials. The dioxin function derived in the form of RSD_r (%) = $0.57(15.3c^{-0.096})$ and RSD_R (%) = $15.3c^{-0.096}$ can be used as target RSDs. As the levels of the 29 PCDD/Fs and DL-PCBs are generally quite different in naturally contaminated biological samples (i.e. spanning several orders of magnitude depending on the congener), adapted target RSDs according to the equations are therefore available. Calculated RSDs performed during the internal validation in repeatability and intermediate precision should be lower or equal to target RSD_r and RSD_R as acceptance criteria.