Quality control in PCDD/PCDF analysis by comprehencive examination of GC-MS analytical data

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PCDD/PCDF and relative compounds trace level analysis includs a number of operations i.e. sampling, extraction, clean-up, GC-MS analysis, data treatment, and each of these may distort analysis results due to analytes losses, artefacts appearance, uncomplete elimination of interferences and bulk material. Therefore quality control (QC) should provide a method vavidation procedure for the entire analytical train. Comprehencive QC usually includes labelled surrogate standards spiking a sample before extraction, internal standard adding to cleaned extract before injection, replications, field and laboratory blank and fortified samples analysis, selective multichannel detection, data set examination for some criteria to be fulfilled. GC-MS analysis should provide several isotope ions monitoring from molecular and principal fragment ions clusters for analytes and standards, some ions ofprobable interferences, e.g. biphenyls, for addition confirmation of analyte identity.

Data treatment is a final part of the analytical train and some experimental errors can be smoothed by rational data treatment procedure. The current state of the art in data evaluation is that some simple criteria should be fullfilled: simultaneous responce of all characteristic ions, retention time within the window, signal to noise ratio should be large enough, ratio of simultaneously eluted peak areas on various mass chromatograms should be in tolerance interval near to the theoretical derived from natural isotopes distribution. All these criteria to be fullfilled is a demand of positive identification.

In the attempt to fulfill all these criteria sofisticated clean-up procedures and appropriate GC-MS techniques are used. But even such complex, time and labour conouming clean-up chemes often do not provide correct inentification when these criteria fulfilled achieving especially if analyte concentration is near the detection limit.

A distortion of analytical sygnal may be either when analyte is absent and interferences yield a sygnal similar to that of the analyte, or when the extract contains the analyte but its sygnal is disturbed by background, interferences or instrument parameters variations (selfinduced CI, resolving

power reducing, etc.). Retention times also may fluctuate due to column deteriration, run parameters altering or coeluted compounds (e.g. when analyte peak in on the tail of larger peak).

A possible way is widening of the criteria tolerance intervals if only for sygnals near detection limit. These intervals depend on confidence probability.

A comprehencive data set treating allow to estimate confidence probability based on data available by some more sofisticated mode of criteria using. The procedure includs MS and GC data evaluation.

1. MS data analysis allows to estimate how large is a difference between analyte and reference mass spectra. After compairing retention times and selection of simultaneously eluted peaks in the ion mass chromatograms we have a set of peak areas which distribution should be compaired with that of standard or with the theoretical distribution. This may be done using $\chi 2$-test:

$$\chi^2 = \sum_k \frac{(A_k - A_k^o)^2}{A_k^o} \qquad (1)$$

where A_k - measured peak area, A_k^o - expectes peak area,

If calculated $\chi^2 < \chi^2_{k-1,\alpha}$, where $\chi^2_{k-1,\alpha}$ - critical value for χ^2 from the table for k-1 degrees of fridom and significance level α, then hypotesis may be adopted that the distribution of measured peak areas to be the same as theoretical or standard with confidence probability $P = 1 - \alpha$.

If calcucated $\chi 2$ is larger than critical value or near to it the one or more terms can be found in (1) that yields the most contribution in $\chi 2$ and compairing with theoretical or standard distribution be made for the sum of these and for the rest terms in (1) using $\chi 2$ test. So this term is tested if it is significant for estimating of similarity of peaks distributions measured and theoretical.

For example peak areas A_i in mass chromatograms for 2,3,7,8-TCDD differ from theoretical values A_i^o:

m/z	A	A^o	$(A-A^o)^2/A^o$
320	87	75	1,8
322	103	100	0,1
324	36	50	4,2
χ^2	=		6,1 > 5,99 = $\chi^2_{2,0,05}$

Calculated χ^2 value is larger then tabulated. One of the terms is much more then others and if it is eliminated calculated $\chi 2$ becomes less then tabulated:

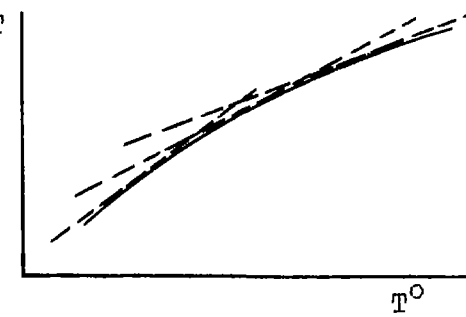
2. GC data analysis. If analysing mixture contains a labelled analog of analyte retention time of the analyte may be determined easily: it should not differ from that of the labelled standard more then 1-2 s of 1-2 scans. If appropriate internal standard is not available data obtaned former in similar conditions or known from literature may be used. Analyte RT may be predicted using a well known way of correlation of two data sets. Comparision of two data sets is carried out using a plot log T versus log T^{O} (or T versus T^{O}), where T and T^{O} are RT in the analysed mixture and in reference data sets. To make this plot some known components in the analysing mixture (standards, known admixtures etc.) corresponding to components in another data set should be found for regression parameters calculating, in simplest case a linear regression:

$$T = c + d\,T^{O} \qquad (6)$$

Then RT of all components in the reference data set may be recalculated for the analysing mixture data set to check if there is coincidence of the recalculated RT with measured within a confidence interval. If a coincidence is real the component may be identified on the base of the reference RT.

When some components in analysing mixture are identified by such a way their RT may be used for regression improving due to increasing a number of points. So final identification is achieved by successive approximation.

In various parts of the plot linear regression parameters may be different, and relation of two RT sets may be represented as nonlinear regression or a number of linear parts or by cubic spline smoothing. A confidence probability may be determined for each analyte as above and used to overall confidence probality determination with MS data.



GC identification complets MS data when appropriate standards are not available. The complete data treatment allows to smooth some ion intensities fluctuations to improve identification reliability near detection limit.

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