Application of a Plackett- Burman Statistical Design in the Determination Method of the Environmentally Hazardous Microelements (Ni, Pb, Cd) from Polyether Polyols

ALEXANDRU PANA[1], ION TANASE[2], DANA ELENA POPA[2]*
[1] QAFCO Company, Mesaieed , Qatar, PO Box : 50001
[2] University of Bucharest, Faculty of Chemistry, Department of Analytical Chemistry, 90-92 Panduri Road, 050663, Bucharest, Romania,

Quantifying environmentally hazardous elements content of polyether polyols used to obtain polyurethane foam based products, before selling them on the market, is regulated by the international laws so that the products have no negative impact on the environment. For the determination of hazardous elements there are not any standardized methods of analysis. For these reasons, to obtain a thorough characterization of the polyether polyols regarding the content of hazardous metals (Ni, Pb, Cd) a validated method is required. In the method development stage, a Plackett-Burman statistically designed experiment was used to identify which factors have the greatest impact on the performance parameters of the analytical method: the ashing time in the case of Pb, the atomization time for Cd determination and the bandpass in the case of Ni. The half-normal probability plots were constructed. Based on the estimated effects, the experimental error (SE) and the margin of error (ME) were also calculated. For Ni and Cd the non-significance intervals were estimated.

Keywords: Plackett-Burman experimental design, Atomic absorption spectrometry, hazardous metals, polyether polyols

This study aims to assess the performance of the method used for some of the environmentally hazardous elements (Ni, Pb, Cd) quantification from polyether polyols, considering the analyzed matrix is one with special properties (products with variable viscosity and high molecular mass). The quantitative determination in polyols of hazardous substances such as volatile unreacted monomers (using Headspace Gas Chromatography/ Mass Spectrometry) and metals such as Ni, Pb, Cd, Co, Hg, As is important regarding the performing of the foaming process and also from the point of view of the end user. The quantification of environmentally hazardous elements content in polyether polyols (used in the preparation of the polyurethane foams based products: coating materials, thermal and acoustic insulation materials, adhesives for various packaging, flooring, furniture, auto parts) before selling these products on the market is strictly regulated by the European environmental legislation [1-4].

It should be noted that for the determination of Ni, Pb, Cd in polyether polyols there is no standardized analytical method. Obtaining a reliable method involves a previous validation. The lack of the environmentally hazardous elements control or using non-validated analytical methods may lead to banning the products selling or to claims for compensation from the beneficiaries. It is well known that an extension of the concentration range or adapting the method to another totally different matrix can easily lead to incorrect measurement results.

The development and validation of an analytical method for the quantitative determination of the hazardous elements from polyether polyols will provide a safe instrument for certifying the quality of such products allowing their free movement on the market according to the international requirements.

Since the method development is the first step of the validation process we intended to perform an evaluation study of the method process parameters significance and to calculate the critical effects associated to the factors. Setting the nominal process parameters of the method is very important. Inappropriate choice of the nominal values can ultimately lead to obtaining the unsuitable method performance parameters. For this reason we decided that the establishment of the nominal values of method process parameters to be done using a Plackett - Burman experimental statistical design.

It is internationally recognized that the validation is necessary in the analytical laboratories. As the definition says, validation always concerns a particular “analytical system”. This means that for a specific type of material and a certain range of concentrations, the method must be able to solve a particular analytical problem [5]. The method validation is closely related to the method development. When a new method is developed, some parameters were already evaluated during the “development stage” which in fact is a part of the “method characterization” [6].

The first major stage of the analytical method validation, namely method development, is devoted to the creation of a method with good performance parameters which will allow it to be validated. In this stage, to identify the potentially responsible factors a robustness test is used [7]. Screening designs are commonly used when little is known about the system or the process. In general, these designs are fractional factorial of a 2^n design that can identify main factors from a large number of suspected variables. So the number of trials can be reduced down to the absolute minimum. The plan has the goal of screening out numerous factors in order to choose the ones that have the biggest impact on the results [8, 9]. The designs applied in the robustness tests are the fractional factorial [10-12] or the Plackett-Burman designs [12, 13]. Because in a robustness test the main concern relates to the main effects of the

---

* *email: dana_lena1978@yahoo.com*

factors and the interactions between two factors can however be considered negligible [14] in this method development study a Plackett-Burman experimental design was selected. The literature data focused on the determination of different hazardous metals in the polyether polyols matrix is lacking. It is also worth mentioning that the application of the experimental projects in both stages of the validation method to evaluate a key parameter as robustness is less frequently used in industrial laboratories practice.

In 1946, R.L. Plackett and J.P. Burman published a paper [13] that described the construction of very economical designs with the experimental run number multiple of four (rather than of power of 2). Theoretical developments, especially the construction of the fractional factorial designs, may be found in literature [15].

Performing a robustness test in the analytical method development stage is an approach to examine the potential sources of variability, to evaluate the quantitative aspects of the method, to define the system suitability test (SST) limits and to predict the reproducibility or intermediate precision estimates [16].

The first step is the identification of the factors to be tested. The second step consists in the selection of the factors levels. Usually, when an analyst applies a well established method all the factors are at the nominal level. In a robustness test performed in the method development stage the factors are checked at extreme levels, taking into consideration the widest ranges of variation of these factors. The levels can be defined by the analyst according to his personal experience (in most case studies) or can be based on the precision or the uncertainty [17] with which a factor can be set. When the uncertainty is used to select the extreme levels in a robustness test experiment, this interval (value of nominal factor + uncertainty) is enlarged to represent possible variations between instruments and laboratories by multiplying the uncertainty with a coefficient *k*. The value *k* = 5 is proposed as the default value. The introduction of the coefficient *k* should also compensate for the occasional sources of variability which were not taken into account in the estimation of the absolute uncertainty. Once the factors are selected and the levels are established the analyst should use an experimental design selected as a function of the number of factors to investigate. A Plackett- Burman design with N experiments (N must be multiple of four) can examine up to N-1 factors. An example of a Plackett-Burman design for N = 12 is shown in table 1.

Table 1

PLACKETT-BURMAN DESIGN FOR 11 FACTORS (N = 12) [20]

Exp. No.	Factors											y_i
	A	B	C Dum 1	D	E	F	G Dum 2	H	I	J	K Dum 3	
1	+1	+1	-1	+1	+1	+1	-1	-1	-1	+1	-1	y_1
2	-1	+1	+1	-1	+1	+1	+1	-1	-1	-1	+1	y_2
3	+1	-1	+1	+1	-1	+1	+1	+1	-1	-1	-1	y_3
4	-1	+1	-1	+1	+1	-1	+1	+1	+1	-1	-1	y_4
5	-1	-1	+1	-1	+1	+1	-1	+1	+1	+1	-1	y_5
6	-1	-1	-1	+1	-1	+1	+1	-1	+1	+1	+1	y_6
7	+1	-1	-1	-1	+1	-1	+1	+1	-1	+1	+1	y_7
8	+1	+1	-1	-1	-1	+1	-1	+1	+1	-1	+1	y_8
9	+1	+1	+1	-1	-1	-1	+1	-1	+1	+1	-1	y_9
10	-1	+1	+1	+1	-1	-1	-1	+1	-1	+1	+1	y_{10}
11	+1	-1	+1	+1	+1	-1	-1	-1	+1	-1	+1	y_{11}
12	-1	-1	-1	-1	-1	-1	-1	-1	-1	-1	-1	y_{12}

The first row in the design is described by Plackett-Burman [13]. After the determination of the real number of the factors to be examined, the remaining columns in the design are defined as dummy factors. A dummy factor is an imaginary factor for which the change from one level to the other has no physical meaning. For every studied factor the effect on the result of the response (E_X) can be calculated from the measured response results. The effects of the factors on the responses can be significant or non-significant. The identification of the important effects is done graphically when an estimate of the experimental error of the effects is not available and with a statistical test when this estimate exists. The graphical identification of the important effects is applied using a normal probability plot [18] or a half-normal plot [19]. For normal probability plot construction should be used a statistical software package and for a half-normal plot the effects from a robustness test design are plotted against the corresponding rankits [20]. Both in a normal probability and in a half-normal plot the no-significant effects tend to fall on a straight line through zero, while the significant effects deviate from it. The statistical interpretation provides the user a numeric limit value that can be plotted on the graphical representation and that allows to define, in a less subjective way than the visual one, what is significant and what is not. This limit value is derived from the t-test statistic [7, 21]:

$$E_{critical} = t_{critical}(SE)_c \qquad (1)$$

where: $(SE)_e$ is the estimated standard error, $t_{critical}$ is a tabled t-value at R freedom degrees (R= n-1 for n replicated nominal experiments and R = N for duplicate design experiments).The critical effect $E_{critical}$ is calculated at a significance level $\alpha = 0.05$. An effect is considered significant at a given α level if $|E_X| > |E_{critical}|$. For the statistical interpretation of the effects three different ways of estimating the experimental standard error of an effect can be applied: from intermediate precision estimate, from dummy effects and from the distribution of the effects. Dong's algorithm [22] is a suitable tool to identify the significant effects for the small designs. After the calculation of the initial error of an effect (s_0) and of the experimental standard error (s_1), the margin of error (ME) can be determined. According to the literature, an effect that exceeds the margin of error (ME) is potentially significant and is recommended to be used as the decision criterion for all the effects calculated from the robustness test. It is recommended that the analysts apply Dong's method for the interpretation of the graphical plots. When a factor has a significant effect on a response, the interval in which the factor levels should be controlled to eliminate the effect is also estimated.

Experimental part

In order to carry out a robustness test in the method development stage, using Plackett-Burman statistically designed experiment (with 12 experiments and 11 factors) a standard polyether polyol of type three-ol (commercially known as Petol 48-3MB, produced by SC Oltchim SA company from glycerine, ethylenoxide and propylenoxide, mostly used industrially to obtain polyurethane foams) was used. Since there are no reference materials with a similar matrix with those of polyol a commercially polyol sample was used. This sample was analysed 6 times in terms of repeatability. After the exact determination of Ni, Pb, Cd metal content this sample was enriched in each metal up to a level well established to be situated in the middle of the calibration curve. According to the Safety Data Sheet of the product, the polyether polyol is partially soluble in water. Thus by enrichment of the actual sample with the aqueous standard relatively homogeneous mixtures were obtained. Then, the concentrations of Ni (10 μg × kg^{-1}), Pb (10μg × kg^{-1}), Cd (2.5 μg × kg^{-1}) in this enriched sample were determined according to the statistically designed experiment. To check if the results are influenced by the drift because of the time effect and the sample is analyzed under identical experimental conditions, from time to time two supplementary injections of the blank and the standard solutions were performed. Because of the organic nature of the sample the measurement procedure involves the samples (5g) calcination (4 h at 380 °C) and the transfer of the calcination residue into the aqueous acid solution 0.5% HNO_3 (23). The analysis of these elements was carried out over a period of 1 week. During this period the samples were sealed and kept at controlled temperature between 15-30 °C, to prevent their contamination or degradation. To enrich the commercial sample of polyol, (S) CertiPUR stock solutions of metals with concentration of 1000 mg/L, produced by Merck, were used. The used analytical instrument was an Atomic Absorption Spectrometer, SOLAAR 969 type, manufactured by UNICAM. Because the content of the metals of interest is at the μg × kg^{-1} level, the measurements have been carried out by Electrothermal Atomization Technique using a Massman graphite furnace equipped with a graphite cuvette of ELC (Extended Lifetime Cuvettes) type (table 2) [23, 24].

Table 2
WORKING NOMINAL PARAMETERS OF AAS DETERMINATION

AAS parameters for Ni: wavelength: 232 nm; bandpass: 0.2 nm; lamp current: 80%					
Sample volume: 10 μL. Furnace parameters					
The phase	Temp (°C)	Time (s)	Ramp (°C × s^{-1})	Gas flow (L × min^{-1})	The action
1	100	30	5	0.2	-
2	250	10	5	0.2	-
3	500	10	10	0.2	-
4	1000	20	150	0.2	-
5	2500	3	0	Off	RD TC*
6	2600	3	0	0.2	TC
AAS parameters for Pb: wavelength: 217 nm; bandpass: 0.5 nm; lamp current: 90%					
Sample volume: 20 μL. Furnace parameters					
The phase	Temp (°C)	Time (s)	Ramp (°C × s^{-1})	Gas flow (L × min^{-1})	The action
1	90	30	10	0.2	-
2	250	20	10	0.2	-
3	600	10	10	0.2	-
4	700	10	150	0.2	-
5	1200	3	0	Off	RD TC
6	2500	3	0	0.2	TC
AAS parameters for Cd: wavelength: 228.8 nm; bandpass: 0.5 nm; lamp current: 80%					
Sample volume: 5 μL. Furnace parameters					
The phase	Temp (°C)	Time (s)	Ramp (°C × s^{-1})	Gas flow (L × min^{-1})	The action
1	100	30	5	0.2	-
2	200	10	10	0.2	-
3	300	20	150	0.2	
4	900	3	0	Off	RD TC
5	2500	3	0	0.2	TC

* TC is the optical wire temperature control and RD is the read data.

Table 3
FACTORS AND LEVELS INVESTIGATED IN THE ROBUSTNESS TEST FOR Ni

Crt. No.	Factor	Units	Limits	Level (-1)	Level (+1)	Nominal
1	Ashing temperature of the organic sample	°C	± 10	370	390	380
2	Flask volume	mL	± 0.25	24.75	25.25	25
3	Ashing time	sec	± 0.5	19.5	20.5	20
4	Atomization time	sec	± 0.5	2.5	3.5	3
5	Lamp current	%	± 5	75	85	80
6	Furnace temperature	°C	± 10	2490	2510	2500
7	Bandpass	nm	0.2; 0.5	0.2	0.5	0.2
8	Sample mass	g	± 0.001	4.999	5.001	5

Table 4
FACTORS AND LEVELS INVESTIGATED IN THE ROBUSTNESS TEST FOR Pb

Crt. No.	Factor	Units	Limits	Level (-1)	Level (+1)	Nominal
1	Ashing temperature of the organic sample	°C	± 10	370	390	380
2	Flask volume	mL	± 0.25	24.75	25.25	25
3	Ashing time	sec	± 0.5	9.5	10.5	10
4	Atomization time	sec	± 0.5	2.5	3.5	3
5	Lamp current	%	± 5	85	95	90
6	Furnace temperature	°C	± 10	1190	1210	1200
7	Bandpass	nm	0.2; 1.0	0.2	1.0	0.5
8	Sample mass	g	± 0.001	4.999	5.001	5

Table 5
FACTORS AND LEVELS INVESTIGATED IN THE ROBUSTNESS TEST FOR Cd

Crt. No.	Factor	Units	Limits	Level (-1)	Level (+1)	Nominal
1	Ashing temperature of the organic sample	°C	± 10	370	390	380
2	Flask volume	mL	± 0.25	24.75	25.25	25
3	Ashing time	sec	± 0.5	19.5	20.5	20
4	Atomization time	sec	± 0.5	2.5	3.5	3
5	Lamp current	%	± 5	75	85	80
6	Furnace temperature	°C	± 10	890	910	900
7	Bandpass	nm	0.2; 1.0	0.2	1.0	0.5
8	Sample mass	g	± 0.001	4.999	5.001	5

To comprise the entire method, the factors studied in the experimental design were selected from the sample preparation phase and from the AAS measurement phase. Where it was possible the factor levels were chosen symmetrically around the nominal level. For the factors for which the measurement uncertainty was known the selected extreme levels were obtained by multiplying the uncertainty by a coefficient $k = 5$ (tables 3-5).

For example, the ± 0.25 mL extreme levels for the flask volume were obtained by multiplying the value from the calibration certificate with the $k = 5$ coefficient. For the factors for which the manufacturer of AAS instrument has not indicated the uncertainty, the lower and upper values closest to the nominal values permitted by instrument software multiplied by 5 were selected as extreme levels. For the sample mass weighing a typical balance with four decimals was used, the tolerance permitted between two weighings being 0.0002 g.

No additional nominal experiments were added to the experimental set-up. In order to minimize the influence of an occasional time effect on the real factor effects and to have the possibility to perform a statistical interpretation of the effects, to those 8 quantitative factors were added 3 dummy factors.

Results and discussions

After the Plackett-Burman statistically designed experiment was carried out, a set of concentration values for metals of interest was obtained. The uncorrected analytical data obtained from the measurements are showed in the table 6.

Table 6

RESULTS OF THE EXPERIMENTS FOR THE STUDIED RESPONSE

	Responses (concentrations)		
Exp. No.	C_{Pb} ($\mu g \times kg^{-1}$)	C_{Ni} ($\mu g \times kg^{-1}$)	C_{Cd} ($\mu g \times kg^{-1}$)
1	10.12	9.84	2.59
2	8.79	9.39	2.30
3	10.03	10.17	2.22
4	10.70	8.97	2.43
5	9.28	8.73	2.00
6	8.96	8.92	2.12
7	9.19	10.84	2.53
8	9.29	8.70	2.15
9	9.02	9.63	2.23
10	9.47	9.70	2.37
11	9.20	9.34	2.48
12	8.75	9.24	2.25
Mean	9.40	9.46	2.31
RSD %	6.30	6.65	7.74

Using the measured responses the effects of studied factors on the results were calculated. Each effect is calculated with the formula:

$$E_X = \frac{\sum Y(+)}{N/2} - \frac{\sum Y(-)}{N/2} \qquad (2)$$

where: X is the factor, E_X is the effect of X on response Y, $\Sigma Y(+)$ and $\Sigma Y(-)$ are the sums of the responses at the extreme levels, N is the number of experiments.

Since no additional nominal experiments were performed no normalized effect values were calculated. Table 7 shows the calculated effects of the different factors on the considered response.

Because a statistical software package was not available the normal probability plots were not constructed. The half-normal probability plots were constructed using the rankits given in literature for the most frequently executed Plackett-Burman designs (table 8) [20].

To create the half-normal plots the calculated effects were ranked according to the absolute increase of the effect size (fig. 1). The median values obtained were 0.168, 0.203, and, respectively, 0.078 $\mu g \times kg^{-1}$ for Ni, Pb and Cd.

Table 7

EFFECTS OF THE FACTORS ON THE CONSIDERED RESPONSES

	Effects of the factors ($\mu g \times kg^{-1}$)		
Factors	Ni	Pb	Cd
Ashing temperature of the organic sample (A)	0.595	0.150	0.121
Flask volume (B)	-0.168	0.330	0.075
Dummy 1 (C)	0.075	-0.203	-0.078
Ashing time (D)	0.068	0.693	0.125
Atomization time (E)	0.125	0.293	0.166
Lamp current (F)	-0.328	0.023	-0.152
Dummy 2 (G)	0.395	0.097	-0.004
Furnace temperature (H)	0.125	0.520	-0.044
Bandpass (I)	-0.815	0.017	-0.143
Sample mass (J)	0.308	-0.120	0.001
Dummy 3 (K)	0.051	-0.500	0.039

According to the theory, both in a normal probability and in a half-normal probability plot non-significant effects tend to fall on a strait line through zero (fig. 1). The points on the curves that exceed the horizontal line which represent the critical effect (ME - margin of error) are suspected to be significant factors.

Table 8

RANKITS FOR THE MOST FREQUENTLY DESIGNS [20]

Effect	Design size		
	N = 8	N = 12	N = 16
1	0.09	0.06	0.04
2	0.27	0.17	0.12
3	0.46	0.29	0.21
4	0.66	0.41	0.29
5	0.90	0.53	0.38
6	1.21	0.67	0.47
7	1.71	0.81	0.57
8		0.98	0.67
9		1.19	0.78
10		1.45	0.89
11		1.91	1.02
12			1.18
13			1.36
14			1.61
15			2.04

The interpretation of these plots is not straightforward and for this reason graphical interpretation was combined with a statistical one. Because the number of experiments was big enough, having a proper number of freedom degrees to perform a statistical interpretation the estimated experimental standard error $(SE)_e$ and from dummy factors and the critical effects (ME) from Dong's criterion were calculated as follows.

The estimated experimental standard error $(SE)_e$ from the intermediate precision estimation is calculated according to the formula:

$$(SE)_e = \sqrt{\frac{4s^2}{N}} \qquad (3)$$

where: s^2 is the variance of the replicated measurements at nominal level or of the duplicate design experiments and N is the number of experiments.

An estimate of the experimental standard error can be also obtained from the dummy effects according to the formula:

$$(SE)_e = \sqrt{\frac{\sum E_{error}^2}{n_{error}}} \qquad (4)$$

where: ΣE^2_{error} is the sum of squares of the n_{error} dummy effects. According to Dong (22) an initial estimate of the error on an effect (s_0) is obtained from the formula:

$$s_0 = 1.5\,\mathrm{median}|E_i| \qquad (5)$$

where: E_i is the value of the i effect. From s_0 the final estimation of the standard error (s_1) can be calculated:

$$s_1 = \sqrt{m^{-1}\sum E_i^2} \qquad (6)$$

where: m is the number of the absolute effects smaller than $2.5s_0$ $(|E_i| \langle 2.5\, s_0)$. The s_1 value is used to calculate the margin of error (ME) which is a critical effect:

Table 9

CRITICAL EFFECTS OBTAINED FROM THE DIFFERENT STATISTICAL INTERPRETATION METHODS

	Critical Effects		
	Ni	Pb	Cd
The estimated experimental standard error $(SE)_e$ from dummy factors			
$\alpha = 0.05$	0.744	1.007	0.160
$\alpha = 0.1$	0.550	0.745	0.119
The estimated margin of error (ME) from Dong's criterion			
$\alpha = 0.05$	0.799	0.624	0.227
$\alpha = 0.1$	0.652	0.509	0.185

Table 10

THE SIGNIFICANCE OF THE FACTOR EFFECTS ON THE DIFFERENT RESPONSES WHEN CRITICAL EFFECTS ARE ESTIMATED USING DUMMY EFFECTS (• SIGNIFICANT) OR DONG'S CRITERION (•• SIGNIFICANT)

	From dummy effects			From Dong's criterion		
Factors	Ni	Pb	Cd	Ni	Pb	Cd
Ashing temperature of the organic sample (A)	-	-	-	-	-	-
Flask volume (B)	-	-	-	-	-	-
Dummy 1 (C)	-	-	-	-	-	-
Ashing time (D)	-	-	-	-	••	-
Atomization time (E)	-	-	•	-	-	-
Lamp current (F)	-	-	-	-	-	-
Dummy 2 (G)	-	-	-	-	-	-
Furnace temperature (H)	-	-	-	-	-	-
Bandpass (I)	•	-	-	••	-	-
Sample mass (J)	-	-	-	-	-	-
Dummy 3 (K)	-	-	-	-	-	-

$$ME = t_{(1-\alpha/2,df')}s_1 \qquad (7)$$

where: $1-\alpha/2 = 0.975$ and df = m. All the obtained results are presented in table 9.

Based on the critical effects estimation from dummy effects and from the effects distribution (Dong's algorithm) the significant effects were identified (table 10). It can be observed that in the case of Pb determination the significant factor is the ashing time (D), in the case of Cd is the atomization time (E) and in case of Ni determination the significant factor is the bandpass (I).

Because the factors identified as significant are quantitative factors and the extreme levels are symmetrically situated around the nominal one using equation 7, in the case of Ni and Cd, non-significance intervals (table 11) were estimated as:

$$\left[X_{(0)} - \frac{|X_{(1)} - X_{(-1)}|E_{critical}}{2|E_X|}, X_{(0)} + \frac{|X_{(1)} - X_{(-1)}|E_{critical}}{2|E_X|}\right] \qquad (8)$$

In the case of Ni if the bandpass is controlled within the interval 0.11 – 0.29 nm and in the case of Cd if the atomization time is controlled within the interval 2.99 – 3.01 s no significant effects of the bandpass and of the atomization time on the concentrations of Ni and Cd will be found anymore.

Table 11

NON-SIGNIFICANCE INTERVALS (NSI) ESTIMATED FOR THE ATOMIZATION TIME AND BANDPASS FACTORS IN THE CASE OF Ni AND Cd

Metal	Factors	$E_{critical}$	E_X	X_0	$X_{(-1)}$	$X_{(+1)}$	$NSI_{(-1)}$	$NSI_{(+1)}$
Ni	Bandpass (nm)	0.744	0.815	0.2	0.2	0.5	0.11	0.29
Cd	Atomization time (sec)	0.160	0.166	3	2.5	3.5	2.99	3.01

Conclusions

Using a Plackett-Burman statistically designed experiment allowed the identification of the significant effects and to conclude that the nominal levels of the factors were well established in the method development stage and that they will ensure good performance parameters in the method characterization stage. Starting from an AAS method indicated in the literature, using a statistically designed experiment, a well suited method for the quantitative determination of environmentally hazardous metals (Ni, Pb, Cd) in polyether polyols was obtained, according to the intended purpose, avoiding unnecessarily extensive experiments that imply laboratory resources consumption.

In the case of Ni, Pb and Cd three significant factors were identified and for this reason when a robust considered method is used special attention should be paid to the selection of factor values (the ashing time, the atomization time and the bandpass).

Unlike the application of a statistical experimental design to the robustness study in the method characterization phase (for which, according to the International Conference on Harmonization, the method capacity to remain unaffected by the small variations of the method parameters is studied), in the method development phase the k = 5 used coefficient for multiplying the uncertainty has been considered to compensate the occasional sources of variability that could not have been taken into account in the estimation of the absolute uncertainty. However, it is obvious that using well maintained and calibrated equipment, in the case of these three factors it is unlikely for the variation levels to be so high. This fact led to the utilization of a k = 2 factor in the robustness study in the method characterization phase, which is much closer to the real conditions of the laboratory. In these conditions the factors could become insignificants.

References

1.*** Directive 76/769/EEC on the approximation of the laws, regulations and administrative provisions of the Member States relating to restrictions on the marketing and use of certain dangerous substances and preparations, O.J., L 262, 1976, p. 201

2.*** European Association of Flexible Polyurethane Foam Blocks Manufacturers, http://www.europur.com/uploads/DocumentsLibrary/documents/CertiPUR_Technical_Paper_11.05.2011.pdf, 2012

3.*** Directive 94/62/EC of the European Parliament and of the Council on packaging and packaging waste, O.J., **L 365**, 1994, p. 10

4.*** Directive 2000/53/EC of the European Parliament and of the Council on end-of life vehicle, O.J., **L 269**, 2000, p. 34

5.THOMPSON, M., ELLISON, S.L.R., WOOD, R., Pure Appl. Chem.**, 74**, 5, 2002, p. 835.

6.*** EURACHEM Guide, The fitness for purpose of analytical methods. A laboratory guide to method validation and related topics. LGC, Teddington, 1998

7.YOUDEN, W.J., STEINER, E.H., Statistical Manual of the Association of Official Analytical Chemists (AOAC), AOAC, Arlington, 1975

8.JAIN, S.P., SINGH, P., JAVEER, S., AMIN, P.D., AAPS PharmSciTech, **11**, 2, 2010, p. 936

9.WOYNA-ORLEWICZ, K., JACHOWICZ, R., Acta Pol. Pharm., **68**, 5, 2011, p. 725

10.MORGAN, E., Chemometrics: Experimental design. Analytical Chemistry by Open Learning, Wiley, Chichester, 1991, p. 118-188

11.BOX, G., HUNTER, W., HUNTER, S., Statistics for Experimenters: An Introduction to Design, Data analysis and Model Building, Wiley, New York, 1978, p. 306-418

12.VANDER HEYDEN, Y. MASSART, D.L., in Robustness of Analytical Chemical Methods and Pharmaceutical Technological Products, A. Smilde, J. de Boer & M. Hendricks (Eds), Elsevier, Amsterdam, 1996, p. 79-147

13.PLACKETT, R.L., BURMAN, J.P., Biometrika, **33**, 4, 1946, p. 305

14.VANDER HEYDEN, Y., LUYPAERT, K., HARTMANN, C., MASSART, D.L., HOOGMARTENS, J., DE BEER, J., Anal. Chim. Acta, **312**, 3, 1995, p. 245

15.PRVAN, T., STREET, D.J., J. Stat. Plan. Infer., **106**, 1-2, 2002, p. 245

16.*** The United States Pharmacopeia 23th edition, National Formulary 18, United States Pharmacopeial Convention, Rockville, 1995, p. 1983

17.*** EURACHEM/CITAC Guide, Quantifying Uncertainty in Analytical Measurement, 2nd edn,. EURACHEM Secretariat, BAM, Berlin, 2000

18.VANDER HEYDEN, Y., QUESTIER, F., MASSART, D.L., J. Pharm. Biomed. Anal., **17**, 1, 1998, p. 153

19.NIJHUIS, A., VAN DER KNAAP, H.C.M., DE JONG, S., VANDEGINSTE, B.G.M., Anal. Chim. Acta, **391**, 2, 1999, p. 187

20.VANDER HEYDEN, Y., NIJHUIS, A., SMEYERS-VERBEKE, J., VANDEGINSTE, B.G.M., MASSART, D.L., J. Pharm. Biomed. Anal., **24**, 5-6, 2001, p. 723

21.JONES, K., Int. Lab., **16**, 9, 1986, p. 32

22.DONG, F., Stat. Sinica, **3**, 1, 1993, p. 209

23.CORDOS, E., MANOLIU, C., Spectrometria de absorbtie si de fluorescenta atomica, Editura Academiei, Bucureşti, 1984

24.TĂNASE, I., Analiză instrumentală. Tehnici şi metode spectrometrice. Editura Universităţii din Bucureşti, Bucureşti, 2007

Manuscript received: 21.06.2013