

## **SECTION 1**

---

# **FUNDAMENTALS AND METHOD DEVELOPMENT**

COPYRIGHTED MATERIAL



---

# 1

---

## IMPROVEMENT IN PRETREATMENT AND ANALYSIS WITH SPECTROMETRIC METHODS: A TYPICAL APPLICATION TO ROUTINE ANALYSIS

KHALID BOUTAKHRIT, FABIAN BOLLE, JEAN-MARIE DEGROODT,  
AND LEO GOEYENS

*Scientific Institute of Public Health, Section of Food, rue Juliette Wytsman 14, 1050  
Brussels, Belgium, E-mail: Khalid.Boutakhrif@iph.fgov.becc*

1.1	Preparation of Sample in Routine Analysis	4
1.1.1	Purity of Reagents	4
1.1.2	Contamination from Contact Materials	4
1.1.3	Sample Manipulation	5
1.2	Originality and Applicability of the Procedure	5
1.2.1	Routine Analysis of Trace Element	5
1.2.2	Advantage of Zeeman ET-AAS Over Inductively Coupled Plasma Mass Spectrometry	6
1.2.3	Simplicity and Efficacy at the Same Time	8
1.3	Domain of Application	8
1.3.1	Council Directive 96/23	8
1.3.2	Applications in Control Laboratories	9
1.4	Materials and Methods	9
1.4.1	Mineralization and Analytical Blank	9
1.4.2	Determinations	12
1.5	Validation	13
1.5.1	Precision (Repeatability and Reproducibility)	13
1.5.2	Accuracy	14
1.5.3	Internal Quality Control	14
1.6	Conclusions	16
	References	17

---

*The Determination of Chemical Elements in Food: Applications for Atomic and Mass Spectrometry.*

Edited by Sergio Caroli

Copyright © 2007 John Wiley & Sons, Inc.

## 1.1 PREPARATION OF SAMPLE IN ROUTINE ANALYSIS

In most cases, sample preparation includes several stages, such as drying, homogenization and mineralization. Some of these steps, as performed in the laboratory, may be the source of contamination phenomena, essentially due to the type of vessels and purity of reagents used. They become more and more troublesome as the concentrations to be determined decrease.

### 1.1.1 Purity of Reagents

The purity of the reagents used in the laboratory, mainly  $\text{HNO}_3$  and  $\text{H}_2\text{O}_2$ , is permanently checked when quantifying two procedural blanks. In the ranges used for calibrating, reagent purity has never been a problem.

For instance,  $\text{HNO}_3$  is guaranteed at a purity of better than  $0.4 \mu\text{g l}^{-1}$  in lead and  $0.08 \mu\text{g l}^{-1}$  in cadmium. These values must be compared with dilution or concentration factors resulting from sample preparation. The water used in the laboratory ( $>18 \text{ M}\Omega$ ) has always yielded to totally insignificant blanks. Therefore, if one has 2.5 mL  $\text{HNO}_3$  for a total volume of 10 mL, the maximum contamination can be  $0.1 \mu\text{g l}^{-1}$  Pb. As the range used goes from 1 to  $40 \mu\text{g l}^{-1}$ , the potential contamination from acid can be considered negligible. It is worth noting that the use of a lower range could pose such problems. Considering the nature of routine analyses, the range adopted is the best compromise.

The organization of the analytical process is another point. A systematic contamination is unveiled when analyzing the procedural blanks. Because of the systematic nature of the procedure, one can reasonably assume that, given an equal treatment of all the samples, the same error occurs for every sample. In this way, the error can be brought under control and corrected.

### 1.1.2 Contamination from Contact Materials

In this chapter, procedures are outlined for defining and showing the lowest reliable determination of Pb in food by electrothermal atomization atomic absorption Spectrophotometry (ET-AAS), especially as regards milk and infant formulae, where the maximum acceptable level is  $20 \mu\text{g kg}^{-1}$  fresh weight as defined by Directive 92/46/EEC and Directive 91/321/EEC, respectively [1].

However, the lowest possible measurement of Pb is limited by the contribution of the element from several sources of contamination [2], for example, reagents, vessels and laboratory environment. The contamination coming from ambient air is hard to control unless it is at the price of substantial investment, which sometimes cannot be afforded. The most important contribution of Pb found in this work was the one due to leaching of Pb traces from the walls of materials, mainly digestion tubes and equivalent. This contribution increases with temperature, pressure and time contact between the digested solution and the wall of the digestion tubes. The last parameter (wall of the tube) may not be reduced and the use of wet digestion depends strongly on it. The only way out

is to look for other materials of vessel digestion that release less trace Pb from the walls.

This kind of contamination was carefully evaluated by using various labware: glass and quartz tubes, polytetrafluoroethylene (PTFE) vessels, fluorinated ethylene propylene (FEP) and perfluoroalkoxy (PFA) tubes. Ideally, all trace element determinations should be carried out in “clean” rooms in which the incoming air is filtered and special clothing is worn.

### 1.1.3 Sample Manipulation

The key steps in the analysis of foodstuffs for their trace metal content are basically the acquisition of a representative sample, destruction of the organic matter and end determination. Each of these steps is vital to the success of the overall analysis. Foodstuffs are generally heterogeneous and sample preparation is obviously very important to ensure the quality of the entire analysis. The preparation of the sample depends on its nature (matrix, fresh or dry foods, solid or liquid, fatty compounds, etc.) and on the destruction of the organic matter (wet digestion or dry ashing). To reduce the contamination risks during these steps, a number of rules must be respected [3], for example, ensure a clean environment in the laboratory, minimize handling, avoid filtration and transfer of solutions unless they are absolutely necessary, and carefully clean all vessels by soaking them in acid. In this chapter, sample contamination during sampling and storage is not addressed, although it plays a crucial role.

In the case at hand, samples are first homogenized by mechanical mixing in a mortar for dry foods or a high-speed blender for wet foods. Then 0.5 g of wet product or 0.2 g of dry product is placed directly in PFA digestion tubes equipped with Teflon-lined screw caps. Liquids are probably the easiest material to sample, provided that they have been thoroughly mixed. All materials used are immersed overnight in 10 percent  $\text{HNO}_3$  before use and PFA digestion tubes are decontaminated by introducing 5 mL concentrated  $\text{HNO}_3$ , then heated at  $180^\circ\text{C}$  for 2 h and rinsed three times with Milli Q water.

It is well known that the extreme hydrophobic and anti-adhesive properties of PFA allow quick and simple acidic cleaning procedures to be adopted when preparing labware for trace analysis [4]. On the other hand, preparing conventional quartz vessels for trace analysis requires considerably longer, more powerful and more costly cleaning methods (e.g., boiling or streaming for many hours with  $\text{HNO}_3$ ) [5].

## 1.2 ORIGINALITY AND APPLICABILITY OF THE PROCEDURE

### 1.2.1 Routine Analysis of Trace Element

For the routine analysis of food contaminants, atomic spectrometry has an obvious advantage over chromatographic methods in terms of detection time.

However, the mineralization of sample is time-consuming for application in routine analyses. The analysis duration can be reduced by investigation of the sample digestion. Table 1.1 shows that it is possible to reduce the analysis duration by resorting to wet digestion with PFA tubes.

In ET-AAS, the electrothermal atomization is an obvious advantage, considering the complexity of some matrices. Thanks to the capabilities of this technique, one can better control the effects of the matrix. As sample preparation is less difficult, the time between mineralization and actual measuring out is reduced, which in turn reduces contamination risks.

The control of matrices in ET-AAS also enables a major application for many types of samples. Previous experiments revealed that the behaviors of vegetal and animal matrices were not significantly different. Therefore, at least in principle, one can use the same procedure for both. In practice, and for many determinations in organic chemistry, one and the same operator for monitoring the entire analytical chain is preferable. However, in routine analysis, the tasks of preparing the sample can be differentiated from those of measuring out. As long as the digestion process is under control, the level of knowledge needed is not the same as for the handling of spectrometers.

### **1.2.2 Advantage of Zeeman ET-AAS Over Inductively Coupled Plasma Mass Spectrometry**

The use of low volumes in ET-AAS is an obvious advantage in routine analysis. The risk of an insufficient quantity of digested sample is almost nil. In the concrete case of control during analysis, the mode of the sample injection has several advantages; it allows the apparatus calculation of the recovery rate on each sample without supplementary sample preparation. The addition being directly done by the automatic sampler, the sample does not need further handling. On the other hand, depending on the injection technique, in the case of vegetal samples with a large amount of insoluble silica, filtering is certainly useful. Manipulation is therefore reduced and this is all the more convenient since filtration is a potential source of contamination.

It should be also pointed out that the robustness of electrothermal atomization enables one to avoid the use of high dilution factors. The sensitivity of ET-AAS to a high percentage of dissolved salts, major elements and/or acids is relatively controllable. Manipulation can be also reduced. Calibration is therefore possible at concentrations where contamination phenomena can be better mastered. Multi-element atomic spectrometers have additional advantages of saving time and resources by quantifying simultaneously Cd and Pb.

On the other hand, multi-element ET-AAS shows some weak aspects, namely:

- (1) Linear ranges are element dependent.
- (2) Electrothermal programs may differ widely depending on the elements so that it is impossible to use them in the simultaneous mode.

**TABLE 1.1. Sample Analysis Duration from Digestion to End of the Analysis Depends Strongly on the Mineralization Procedures and the Labware Used**

	Digestion (h)	Comparison of Time and Digestion for Various Procedures		Digestion Rate (%)	Samples/Run	Comparison of Sample Run Capacities	
		Digestion (h)	Analysis Time (min)			Digestion (h)	Time/Sample (min)
Glass or quartz tubes	7	10	10	98	40	7	10.5
Microwave oven	2	10	10	92	10	2	12
Dry ashing	12	10	10	99	10	12	72
Bomb	3	10	10	94	20	3	9
PFA tubes	2	10	10	92	40	2	3

- (3) There is an increase in the difficulties of checking parameters with the number of elements for analyses.

### 1.2.3 Simplicity and Efficacy at the Same Time

The search for efficiency in the analysis process can often coincide with the search for simplification of the analytical process. It is understandable that some requirements must be respected, such as the digestion level of the samples and the control of the matrix effects. This leads to:

- Greater execution speed because fewer operations are required.
- Greater flexibility because the reaction speed is greater.
- Greater analytical pace because analyses are quicker.
- Better control of the parameters because they are less numerous.
- Fewer risks of contamination, because the analysis time and the number of manipulations are reduced.
- Better task assessment.
- Better quality control because processes are shorter and hence better defined.

## 1.3 DOMAIN OF APPLICATION

### 1.3.1 Council Directive 96/23

The aim of the Council Directive 96/23 of 29 April 1996 is to keep the percentage of food contamination at an affordable level. An effective reduction in the levels of residues and contaminants can guarantee a better level of health protection, especially when sensitive groups of population are taken into account.

Another major aim of this Directive is to harmonize legal provisions regarding maximum percentages of contaminants in some foodstuffs to avoid possible barriers to trade caused by differences in standards among the Member States.

Lead can be a serious danger to public health as quoted in the Council Directive 96/23 of 29 April 1996. Lead can slow down cognitive development, reduce the intellectual abilities of children and increase cardiovascular disease and high blood pressure in adults. One should note that, over the last few years, the lead percentage in foodstuffs has considerably decreased thanks to public awareness of the problem and efforts to reduce lead levels. A significant role was played in this context by the quality and reliability of chemical analyses.

The Scientific Committee on Food (Directorate General, "Health and Consumer Protection," European Commission) concluded, in its statement of 19 June 1992, that the average percentage of lead in foodstuffs did not seem alarming, but that longer term action was necessary to further reduce it.

As far as cadmium is concerned, this metal can accumulate in the human body and cause kidney, bone, and reproduction pathologies. Carcinogenic effects are also possible. The Scientific Committee for Food, in its statement of 2 June 1995,

recommended further reduction of food exposure to cadmium because food is the main exposure mode in humans. Hence, for cadmium, maximum affordable percentages should be set at the lowest possible level.

### 1.3.2 Applications in Control Laboratories

The Belgian Food Agency is in charge of the control, analysis, and appraisal of raw materials at every step of the food chain, that is, production, transformation, storage, transport, trade, import, and export. Within this control body there are various units composed of all the external departments entrusted with the control of animals, which was previously the responsibility of several administrations or entities (Veterinary Appraisal Institute, General Inspection of Foodstuffs). While effective control is ensured by the Federal Agency for Foodstuffs, the development and implementation of control measures and programs in order to ensure the protection of consumer health is the task of another administration depending on the Ministry of Public Health.

In their control role, laboratories must ensure quick analysis of the samples taken by the control bodies, aiming at a prompt reaction in cases of crisis and possessing flexibility in their analytical capacity in terms of rapidity (acute crisis), throughput (large number of determinations), and reliability (different types of determinations). Measurement accuracy must obviously be guaranteed.

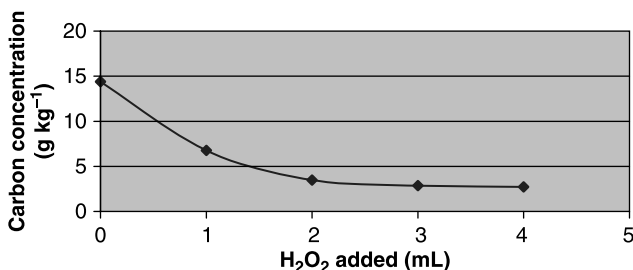
In the authors' laboratory the goal is to submit results on the same day, as long as the sample is brought in the morning. The speed in processing samples has increased substantially in order to be able to cope with crisis situations, and the laboratory can now manage 60 samples a day for simultaneous determination of Cd and Pb with ET-AAS.

## 1.4 MATERIALS AND METHODS

### 1.4.1 Mineralization and Analytical Blank

Most samples require a procedure to get the matrix into solution before analysis by atomic absorption spectrometry. The two most commonly used techniques to accomplish this task are dry ashing at a defined temperature and wet digestion with mineral acids. Dry ashing is generally rather time-consuming: It usually takes a day or more, but very little attention from the analyst is necessary. Contamination and loss of analyte during ashing are possible. Wet digestion methods are generally more rapid; analyses may be completed within a few hours and the methods are less sensitive to contamination, volatilization, or analyte loss.

In the authors' laboratory many acidic mixtures are used to digest organic matter in food. Containers of various materials are also adopted for the digestion of the samples so as to minimize the analytical blank. The magnitude of the latter, in fact, and the degree of uncertainty associated with it, usually limits the ability to perform reliable quantitative determinations at low concentration levels [6].



**Figure 1.1.** Dependence of the digestion process on the amount of H<sub>2</sub>O<sub>2</sub> (from 0 to 4 mL) after digestion of the sample with HNO<sub>3</sub> only for 2 h at a temperature of 180°C.

The combination of HNO<sub>3</sub> and HClO<sub>4</sub> followed by H<sub>2</sub>O<sub>2</sub> is the most commonly used method for food sample digestion. HClO<sub>4</sub> is an excellent oxidizing medium, but sometimes it produces violent explosions, mainly with food powder. It should not be used alone and the mixture should not be evaporated to dryness since inorganic perchlorates are explosive [7]. Therefore, HNO<sub>3</sub> is often preferred and increasing amounts of H<sub>2</sub>O<sub>2</sub>, from 0 to 4 mL, are generally added. After a rapid digestion, the residual carbon can range from 14.4 g kg<sup>-1</sup> (0 mL H<sub>2</sub>O<sub>2</sub>) to 2.7 g kg<sup>-1</sup> (4 mL H<sub>2</sub>O<sub>2</sub>), which is equivalent to 97 percent of destruction (see Fig. 1.1). In the case of skim milk powder (a BCR Certified Reference Material), for example, use was made of HNO<sub>3</sub> + H<sub>2</sub>O<sub>2</sub> and the ratio of organic matter digestion was estimated to be 99.1 percent.

Plant tissues were tested as well to understand the effect of HF in the digestion mixture. There is good evidence that dissolving silica with HF does not improve the quality of the Pb analysis (see Table 1.2). The use of HF should be discouraged because of the risks it brings, all the more so in the case of routine analysis.

To achieve more reliable determinations at lower concentration levels by reducing the analytical blank, several types of labware (glass, quartz, PTFE, FEP, and PFA) were tested to determine their leachability and cleanliness. All materials were kept in 10 percent HNO<sub>3</sub>, then different digestion procedures were carried out using the same amounts of reagents (5 mL HNO<sub>3</sub> + 3 mL H<sub>2</sub>O<sub>2</sub>) and with the use of the heating block reactor, microwave, electric oven, or Bombs. The results obtained for the analytical blanks are summarized in Table 1.3.

**TABLE 1.2. Comparison of Lead Values in Certified Reference Materials Obtained with and Without Adding HF in the Wet Digestion Process**

Materials	Matrix	Certified Value ( $\mu\text{g kg}^{-1}$ )	Value Without HF	Value with HF
SRM 1570a	Spinach leaves	200 <sup>a</sup>	198.7 $\pm$ 4.6	200.6 $\pm$ 3.7
BCR 62	Olive leaves	25000 $\pm$ 1500	26149 $\pm$ 345	26388 $\pm$ 410
IMEP 19	Rice flour	415.9 $\pm$ 7.1	444.4 $\pm$ 9.3	438.3 $\pm$ 10.7

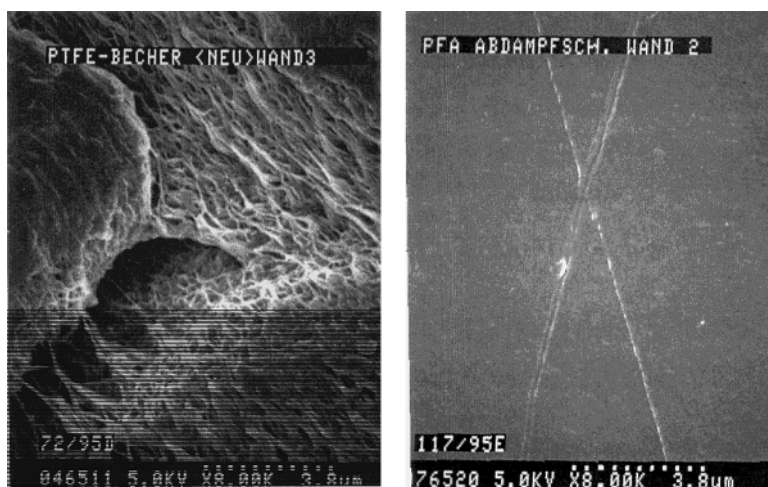
<sup>a</sup>No certified value.

**TABLE 1.3. Estimation of the Detection Limit by Determining the Blank Level for Each Digestion Labware Piece**

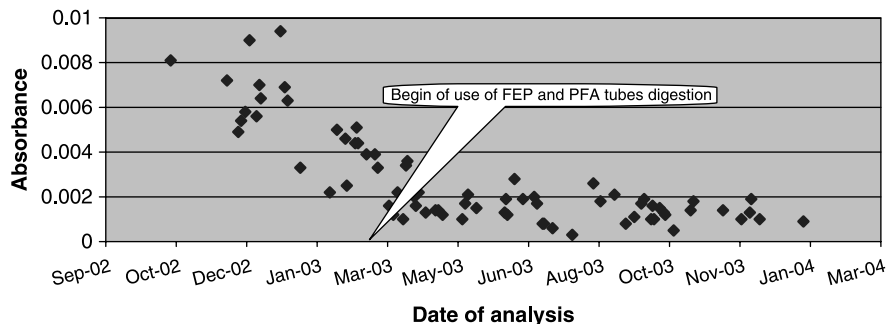
Vessels	Blank Absorbance Value Due to Adsorption	[Blank] ( $\mu\text{g l}^{-1}$ )	L.D ( $\mu\text{g kg}^{-1}$ )
Glass tubes	$0.0073 \pm 0.0009$	$2.47 \pm 0.30$	34
Quartz tubes	$0.0026 \pm 0.0018$	$0.88 \pm 0.61$	27
PTFE (microwave)	$0.0056 \pm 0.0010$	$2.27 \pm 0.34$	33
PTFE (bomb)	$0.0038 \pm 0.0008$	$1.19 \pm 0.27$	20
FEP tubes	$0.0012 \pm 0.0010$	$0.40 \pm 0.34$	14
PFA tubes	$0.0012 \pm 0.0004$	$0.40 \pm 0.14$	8

The results confirmed that the ancillary materials used in an analytical procedures might contribute to blank values more than reagents themselves do, especially glass-ware and microwave digestion systems. Although the latter have been extensively employed to shorten the time required for sample dissolution, some problems have been ascribed to this procedure, as reported by Lima et al. [8].

Digestion bombs attain satisfactory blank values if properly handled, although experimental data turn out to be less accurate. Recovery rates are in general exceedingly high due to lead migration in the Teflon cup walls. In fact, PTFE shows some degree of inhomogeneity, a very rough surface with deep pores and protrusions [9], which renders decontamination more difficult. On the other hand, PFA and FEP wares have a smooth surface with the lowest roughness (Fig. 1.2).



**Figure 1.2.** Scanning Electron Microscopy (SEM) image of surface of PTFE beaker (left) and a PFA evaporating dish (right). Reproduced from [9] with kind permission of VITLAB GmbH, Grossostheim, Germany.



**Figure 1.3.** Improvement in the procedural blank response due to adsorption by using different vessels. In the order: glass tubes, quartz tubes, FEP tubes, and PFA tubes.

The improvement of the procedural blank signal using FEP and PFA tubes instead of glass and quartz tubes is shown in Fig. 1.3. The last shows a better decrease of the signal (blank absorbance) and therefore a lower concentration level will be reached. Fluoromaterials possess a series of unique properties such as wide operational temperature range ( $-200$  to  $+260^{\circ}\text{C}$ ) and high chemical resistance, thus being particularly useful in trace and ultratrace work. Needless to say, a low analytical blank can substantially improve the limits of detection (LoDs) and the accuracy of the method [10].

#### 1.4.2 Determinations

The development of analytical methods for reliable lead determinations in food-stuffs requires the utmost care in consideration of the extremely low levels of this metal. Each single sample preparation steps deserves attention because contamination must always be strictly controlled [11], and problems related to inadequate LoDs can impair the analysis. In this framework, an analytical method was set up for the determination of lead at the trace level and its application to baby food was investigated where the maximum accepted level for the metal is  $20 \mu\text{g kg}^{-1}$  fresh weight. Samples (about 0.5 g) were digested using  $\text{HNO}_3 + \text{H}_2\text{O}_2$  (5 mL + 3 mL) in FEP and PFA tubes. For the determination of Pb, a Perkin–Elmer atomic absorption spectrometer model SIMAA 6000 was used. It is equipped with longitudinal Zeeman effect background correction and a transversely heated graphite atomizer (THGA). The best matrix modifier was a mixture of  $\text{NH}_4\text{H}_2\text{PO}_4$  (0.5 percent w/v) and  $\text{Mg}(\text{NO}_3)_2$  (0.03 percent w/v). The furnace conditions were  $500^{\circ}\text{C}$  ash temperature and  $1650^{\circ}\text{C}$  atomization temperature. Further details about the GFAAS program are given in Table 1.4.

Calibration was performed using aqueous standard solutions containing  $40 \mu\text{g l}^{-1}$  Pb in 2 percent  $\text{HNO}_3$  prepared by diluting a Perkin–Elmer Standard of  $1.0 \text{ g l}^{-1}$ . The measuring cycle was as follows:  $20 \mu\text{L}$  of sample solution and  $10 \mu\text{L}$  of matrix modifier solution were pipetted by the auto-injector on the L'vov platform in the graphite tube and the THGA program was started. All

**TABLE 1.4. Programme for ET-AAS Analysis**

Steps	$T$ ( $^{\circ}\text{C}$ )	Ramp Time (s)
Injection	20	—
Drying	110–130	10
Ashing	500	10
Atomization	1650	0
Clean out	2400	3

measurements were run in duplicate. Sample content calculations are based on peak areas of the background corrected absorption signal and the calibration graph was drawn up for each series of determinations, up to  $40 \mu\text{g l}^{-1}$  Pb.

According to IUPAC, the LoD is defined as three times the standard deviation of the mean of the blank determinations added to the mean of the blank measures [12]. The LoD obtained for the PFA procedure (Table 1.3) is  $8.2 \mu\text{g kg}^{-1}$  Pb. This value is low enough to quantify lead in the vast majority of foodstuffs. This method was successfully applied for various food samples, vegetable and animal tissues. No interferences from other elements have been observed. Matrix effects were seen with some samples, but were eliminated simply by diluting the samples.

## 1.5 VALIDATION

The selection of an analytical technique and the development of procedures and operating conditions are only one part of the process for achieving reliable measurements. Among the first steps of the validation figure the assessment of the method precision; this parameter must be assessed between analysts and days. Appropriate precision, although necessary for reliable work, is not sufficient to guarantee accurate results: Evaluation of sample matrix effect, development, and analysis of spiked samples, analysis of Certified Reference Materials (CRM) of similar composition for which the Pb concentration has been established by other laboratories and comparison of results between laboratories (proficiency testing) are equally important [13].

### 1.5.1 Precision (Repeatability and Reproducibility)

The precision measures include the determination of the repeatability ( $Sr$ ) and the reproducibility ( $SR$ ). A number of determinations were made on spiked meat and vegetable matrix samples. As shown in Table 1.5, on the basis of six runs in six consecutive analytical series (different days and calibrations) and for different analytical levels of Pb, average values of  $Sr = 1.9$  percent,  $SR = 5$  percent and  $Sr = 1.9$  percent,  $SR = 4$  percent; were obtained for meat and vegetables, respectively.

From these results, it seems that, as the concentration of analyte in a sample approaches the LoD, the relative standard deviation associated with that determination increases, as reported in the literature [14]. It is worth noting that these

**TABLE 1.5. Precision of the Procedure for Meat and Vegetable Matrices**

Lead ( $\mu\text{g kg}^{-1}$ )	Repeatability ( <i>Sr</i> )		Reproducibility ( <i>SR</i> )	
	SD ( $\mu\text{g kg}^{-1}$ )	CV (%)	SD ( $\mu\text{g kg}^{-1}$ )	CV (%)
<i>Meat</i>				
50	1.98	3.96	4.88	9.76
100	2.86	2.86	8.80	8.80
300	3.98	1.33	9.42	3.14
500	5.28	1.06	12.90	2.58
750	9.04	1.21	22.25	2.97
1000	8.79	0.88	29.40	2.94
<i>Vegetables</i>				
50	1.98	3.96	2.84	5.69
100	1.64	1.64	3.13	3.13
200	2.92	1.46	6.41	3.20
300	1.61	0.54	14.84	4.95

results were obtained by digestion in FEP tubes. In glass tubes, random contamination at low levels is the major factor of variation, which can be 5 times higher for  $50 \mu\text{g kg}^{-1}$  Pb.

Further improvement of precision at low levels would require the reduction of blank variation; in practice, an additional source of variation may arise from sample inhomogeneity.

### 1.5.2 Accuracy

CRMs are often the best tools for assessing accuracy. The results obtained for FEP and PFA tubes digestion are in agreement with the certified contents for various types of materials, while with other digestion vessels, the lead content was higher than the certified value (Table 1.6).

Checking the FAPAS (S7R40), the largest analytical chemistry proficiency testing scheme in the food sector, lead content with the PFA tube procedure provided the result expected, whereas that obtained previously with glass tubes turned out to be higher.

### 1.5.3 Internal Quality Control

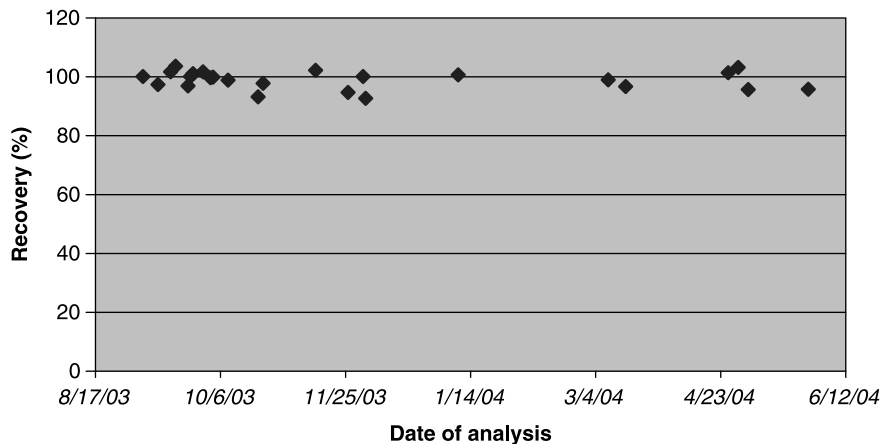
Once the analytical parameters have been determined from the method development and the method has proven suitable for routine measurements, internal quality control (IQC) procedures must be established to maintain the validity of the analytical scheme and to better monitor potential sources of errors. The IQC used includes pre- and post-digestion controls, blank determination, half range of the calibration graph checking, and recovery rate of the samples. The stability of the recovery rate with time (Fig. 1.4) shows that the method is robust after using

**TABLE 1.6. Certified and Found Values for Reference Materials Obtained with Different Vessels**

Certified Reference Material	Code	Certified Concentration ( $\mu\text{g kg}^{-1}$ )	Pb Content Found with Different Vessels ( $\mu\text{g kg}^{-1}$ )					
			Glass Tubes	Quartz Tubes	PTFE <sup>a</sup>	PTFE <sup>b</sup>	FEP Tubes	PFA Tubes
Bovine muscle	BCR 184	239 ± 11	330 ± 8	280 ± 16	351 ± 20	317 ± 52	250 ± 3	245 ± 5
Bovine liver	BCR 185R	172 ± 9	344 ± 33	191 ± 41	260 ± 30	259 ± 42	167 ± 10	191 ± 3
Pig kidney	BCR 186	306 ± 11	413 ± 37	—	464 ± 41	476 ± 165	334 ± 6	330 ± 4
Mussel tissue	BCR 278R	2000 ± 40	2390 ± 160	2240 ± 126	—	2089 ± 121	2109 ± 76	2079 ± 63
Cod muscle	BCR 422	85 ± 15	209 ± 23	143 ± 8	193 ± 14	219 ± 28	124 ± 18	—
Tuna fish	FAPAS 740	9.21	21 ± 2	—	—	—	—	8.8 ± 2

<sup>a</sup>Microwave digestion.

<sup>b</sup>Bomb digestion.



**Figure 1.4.** Recovery of Pb in foodstuff routine samples.

it for about 6 months for the analysis of various foodstuffs matrices. IQC also includes CRMs and PT analyses, control charts, internal and external audit. Therefore IQC comprises the routine practical procedures which enable the analytical chemist to accept either a result or a group of results as fit for purpose or to reject them and repeat the analysis.

In the method described, the procedural blank determination is essential for the analytical procedure since it allows the estimation of all kinds of contaminations. To assess the matrix effect for each sample, recovery rates were determined. The checking of the half range of the calibration graph for each 10 runs of samples allowed the evaluation of calibration graph deviation.

## 1.6 CONCLUSIONS

For foodstuff matrices, the use of PFA labware and  $\text{H}_2\text{O}_2$  reagent improved the conventional analytical parameters (LoDs, level of blanks, stability, recovery rate, and also the speed of analyses. The quantification of lead at low concentration was possible down to  $9 \mu\text{g kg}^{-1}$ , whereas in the recent past the LoD in the same laboratory was at about  $35 \mu\text{g kg}^{-1}$ , thus making participation in the previous FAPAS S7R40 PT impossible. Thanks to the improved procedure, more parameters of quality control could be integrated in the program, that is, recovery rate is calculated for each run of sample and this without lengthening analysis time.

The method described was included among those covered by the accreditation granted by BELAC, that is, the official organization in charge of quality management in Belgium. It can be concluded that reliable results in trace lead analysis may be affected by the purity and surface properties of labware, which can impact on the quality and accuracy of the method in ultra-trace analysis.

**REFERENCES**

1. T. Berg, D. Licht, International legislation on trace elements as contaminants in food, *Food Addit. Contam.*, **19** (2002), 916–927.
2. G. Capar, A. Subjoc, Defining a lowest level of reliable measurement for lead in foods, *J. Assoc. Off. Anal. Chem.*, **65** (1982), 1025–1029.
3. M. Hoenig, Preparation steps in environmental trace element analysis, facts and traps, *Talanta*, **54** (2001), 1021–1038.
4. K. Englert, G. Giebenhain, H.-J. Mosh, N. Muller, Stability of diluted standard reference solution of mercury (Hg) and precious metal in PFA-vessels, *GIT Fachz. Lab.*, **1997**, 32.
5. P. Tschöpel, L. Kotz, S. Schulz, M. Veber, G. Tölg, Causes and elimination of systematic errors in the determination of elements in aqueous solutions in the ng/mL and pg/mL range, *Fresenius' Z. Anal. Chem.*, **302** (1980), 1–14.
6. R. K. Skogerboe, The analytical blank: sources and effects on lead analyses, *J. Assoc. Off. Anal. Chem.*, **65** (1982), 957–964.
7. N. T. Crosby, Determination of metals in foods, *Analyst*, **102** (1977), 222–268.
8. C. Lima, F. Barbosa, J. Krug, Comparison of ultrasound-assisted extraction, slurry sampling and microwave-assisted digestion for cadmium, copper and lead, *J. Anal. Atom Spectrom.*, **15** (2000), 995–1000.
9. J. Dahmen, K. Englert, G. Giebenhain, Properties and production of labware from fluorinated hydrocarbons and their advantages for ultratrace analysis, *Int. Lab. (Pacific Rim Ed)*, **12** (1997 April/May), 30–32.
10. M. J. Baxter, J. A. Burrell, H. M. Crews, R. C. Massey, D. J. McWeeny, A procedure for determination of lead in green vegetables at concentrations down to 1 µg/kg, *Food Addit Contam.*, **6** (1989), 341–349.
11. Y. Schmitt, Influence of preanalytical factors on the atomic absorption spectrometry determination of trace elements in biological samples, *J. Trace Elem. Electrolytes Health Dis.*, **1** (1987), 107–114.
12. *IUPAC Compendium of Chemical Terminology*, 1997, 2nd Ed; <http://www.chemsoc.org/chembytes/goldbook/>.
13. D. A. Bass, D. Hickok, D. Quig, K. Urek, Trace element analysis in hair: Factors determining accuracy, precision, and reliability, *Alternative Med. Rev.*, **6** (2001), 472–482.
14. R. Satzger, E. Bonnin, L. Fricke, Chemical contaminants monitoring, *J. Assoc. Off. Anal. Chem.*, **67** (1984), 1138–1140.

