

Analytical, Nutritional and Clinical Methods Section

Determination of lead in cookies by electrothermal atomic absorption spectrometry with various chemical modifiers

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Abstract

The determination of lead in some Turkish cookies has been studied in the presence of some chemical modifiers. In order to decrease the interferences of sample matrix and to stabilize the atomic absorption signal of lead, W, Pd, W + Pd and W + Pd + tartaric acid (TA) have been tested as matrix modifier. Modifiers have been compared in terms of pyrolysis temperature, atomization and background profiles, precision and accuracy. The W + Pd + TA modifier mixture was found to be preferable for the determination of lead in cookies. The recovery and limit of detection of lead were about 99% and 1.2 µg/l, respectively. © 2000 Elsevier Science Ltd. All rights reserved.

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1. Introduction

The presence of contaminants in the food may result from various sources such as soil, environmental pollution and agricultural practices. Heavy metal contaminants may present a potential hazard for human health if exposure exceeds tolerable levels (Baht & Moy, 1997; Cabrera, Lorenzo & Lopez, 1995). Lead is one of the most extensively studied toxic metals that has aroused considerable concern (Cabrera et al., 1995; Zong, Parsons & Slavin, 1996). However, processing, packaging and other technological processes used to bring foods to the consumer can significantly increase the total concentration of lead (Creaser & Purchase, 1991). Because large amounts of wheat flour products are consumed, even small amounts of contaminants can lead to considerable concentrations of trace elements in a human being organism. The accumulation of toxic metals such as lead in the body can have middle-term and long-term health risks and adversely affect the physiological functions (Cabrera et al.). Inorganic lead (Pb²⁺) is a general metabolic poison and is accumulated in different organs of the human body. It inhibits enzyme systems necessary for the formation of hemoglobin

through its strong interaction with –SH groups. Lead alkyls such as tetraethyl-lead are even more poisonous than Pb²⁺, and are handled quite differently in the body (Stalikas, Pilidis & Karayannis, 1996). Determination of food contaminations is necessary for labeling and for food composition databases that are of interest to consumers as well as nutritionists and health professionals doing epidemiological studies (Miller-Ihli, 1996). FAO/WHO (1983) expounded the dietary intake of lead by adults, infants and children that the determined provisional tolerable weekly intake of lead was 50 µg/kg for adults, 25 µg/kg for infants and children (Baht & Moy, 1997).

Electrothermal atomic absorption spectrometry (ETAAS) is one of the suitable methods for the determination of lead in food, biological, environmental, etc., samples due to its inherent high sensitivity and good selectivity (Bin & Zhe-Ming, 1996; Lynch & Littlejohn, 1989; Zong et al., 1996). But, due to chemical interferences and premature loss of analyte as volatile compounds during the pretreatment and atomization steps, the determination of Pb in such samples by ETAAS is highly difficult. Because of the low concentration of lead in cookies which contain relatively large amounts of metals such as Ca, Cu, Fe, K, Mn, Zn and organic matrix and the lack of a suitable modifier which can stabilize the analyte elements to a pyrolysis temperature, the determination of Pb in such samples by

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ETAAS causes considerably trouble. In order to reduce interferences and increase accuracy, the use of a chemical modifier has become indispensable for the stabilization of volatile elements such as lead during the pyrolysis stage. For this purpose, many metal salts and some organic substances have been used as chemical modifiers in ETAAS (Acar, Türker & Kiliç, 1997; Havezov, Detcheva & Rendl, 1995; Slaveykova & Tsalev, 1990; Tsalev & Slaveykova, 1992; Tsalev, Slaveykova & Mandjukov, 1990; Welz, Schlemmer & Mudakavi, 1992).

In this study, the stabilizing effects of W, Pd, W + Pd and W + Pd + TA in the determination of Pb in Turkish cookies by ETAAS have been investigated systematically. Also, performances of modifiers in terms of pyrolysis temperature, background absorption and interference effects on the atomization profiles of Pb in samples after decomposition with a mixture of HNO₃–HCl were compared. Limit of detection (LOD), characteristic mass (m_0), precision and accuracy were evaluated. The proposed method has been applied for the determination of Pb in a standard reference material (IAEA/V-10 Hay (Powder) and in Turkish cookies.

2. Materials and methods

2.1. Apparatus

All atomic absorption measurements at 283.3 nm resonance line of lead were performed by using a Hitachi Model 180-80 atomic absorption spectrometer, equipped with a graphite furnace (Hitachi 180/078), Zeeman-effect background corrector and an automatic data processor. A 20 µl volume of sample solution was injected by an autosampler (P/N-170/0126). A lead hollow cathode lamp was employed as the radiation source and operated at 7.5 mA. A Varian Model 9176 recorder was used to obtain atomization signal profiles in a 20 mV/FS span. Hitachi pyrolytically coated graphite tubes (P/N-180/7444) were used in tube-wall atomization to measure absorbance values by using integrated mode throughout. Argon served as a carrier gas. The optimized graphite furnace temperature program for pyrolysis and atomization is given in Table 1.

2.2. Reagents

All solutions were prepared from analytical grade reagents in deionized water distilled in an Elgastat Type C114 distillation unit. Stock standard solution (2.0 mg/ml) of Pd in 15% HCl (Merck) were prepared from palladium chloride, PdCl₂ (Merck). 0.4% (m/v) acidic solution of W (V) was prepared from W powder dissolved in H₂O₂. Modifier solutions were diluted as required. 4% (m/v) tartaric acid (TA) solution was prepared daily before measurements. 1.0 mg/ml of Pb standard solution

Table 1
Optimized graphite furnace temperature program

Step	Temperature (°C)	Ramp time (s)	Hold time (s)	Ar flow (ml/min)
Drying-I	50–120	30	–	300
Drying-II	120–300	20	20	300
Pyrolysis	300-Var. ^a	30	30	300
Atomization	2000	–	7	0
Cleaning	2650	–	3	300

^a Var.: varied according to modifier.

(BDH chemical) was used and diluted with 0.2% HNO₃ to prepare working standard solutions.

2.3. Collection and preparation of samples

Twenty four cookie samples were collected from different manufactures. Some of them were in different packs of the same brand. Samples 8–11 were the same brand and samples 2, 6 and 22 were the other brand. Other samples were different brands and different packs. Some of the samples including the above mentioned were also salty and others were sugared. All samples were grounded to small particles and homogenized.

2.4. Decomposition of samples

A portion (6.0–10.0 g) of the cookie sample was accurately weighed into a special crucible which is suitable for the determination of trace elements. The crucible was put into a cold muffle furnace and heated up to 110°C and kept at this temperature for 2 h in order to dry the sample. After that, the temperature was gradually increased up to 450°C (Turkish standard, TS 3606, 1997) and left 4 h for the sample to ash completely. The cooled sample was taken from the furnace and a minimal volume of 0.02 mol/l nitric acid was added to moisten it. The sample was transferred from the crucible into a 50 ml Teflon beaker by washing the interior surface of the crucible with a mixture of 10 ml of conc. HNO₃ and conc. HCl (3 + 1; v/v). The Teflon beaker was covered with a Teflon cover and heated on a hot-plate at about 120°C until the total volume of the sample was reduced to 5 ml. After removing and cooling the beaker, interior surface of beaker and cover were washed with 3 ml of 0.02 mol/l nitric acid and heated at 100°C for 1 h, then raised to 120°C with gentle heating until complete decomposition of the sample was achieved. When the residue was left, a further 2 ml of the same acid mixture was added and the above procedure was repeated. After cooling, the sample solution was transferred into a 50 ml volumetric flask. The interior surface of the Teflon beaker was washed with a sufficient volume of 0.02 mol/l nitric acid twice and the final solution was diluted up to the mark with deionized water.

Standard reference material (1.22 g) (IAEA/V-10, Hay powder) was weighed into a Teflon beaker and moistened with a minimal volume of 0.02 mol/l nitric acid. 3 ml of acid mixture (HNO₃ + HCl; 3 + 1, v/v) was added to the sample and heated at a temperature of about 150°C on a hot-plate, until the total volume was nearly 1.5 ml. To achieve the complete decomposition, a further 1.5 ml of the same acid mixture was added and the above procedure was repeated. The sample solution was transferred into a 25 ml volumetric flask by washing the interior surface of the Teflon and diluted up to the mark with deionized water.

In all instances, blanks were run with the reagents used under the same conditions in order to check for possible contamination from the digestion procedure.

2.5. General procedure

1 ml of Pb standard solution (100 ng/ml) for matrix modification was added to 1 ml of the modifier solution (0.4 mg/ml of Pd, 2.0 mg/ml of W or 2.0 mg/ml of W + 0.4 mg/ml of Pd) (Acar et al., 1997) and injected into the pyrolytic graphite coated tube in order to obtain sufficient absorbance signal (above 0.05 abs. units) by initiating the optimized graphite furnace temperature program as shown in Table 1. The optimum modifier mass and mass ratio of the mixed modifier components obtained in our previous paper (Acar et al., 1997) as 20 µg of W, 4 µg of Pd and 20/4 (µg/µg) of W/Pd were used. Two hundred micrograms of TA were used together with W + Pd. In order to determine pyrolysis temperature of lead for the standard and sample solution, 0.5 ml of the standard solution (analyte concentration 200 µg/l) or 0.5 ml of the sample solution and 0.5 ml of water were added to 1 ml of modifier solution (2.0 mg/ml of W, 0.4 mg/ml of Pd or 2.0 mg/ml of W + 0.4 mg/ml of Pd mixture) separately. Half a milliliter of TA was used instead of 0.5 ml of water when it was used as a reducing agent. 20 µl of standard solution or sample solution diluted by a factor 4 (0.5 ml of sample solution + 1.5 ml of modifier mixture or 1.5 ml of water) was injected into the graphite tube in the presence or absence of modifiers.

Thermal pretreatment curves of Pb in aqueous standard and in the cookie sample with or without of modifiers were then plotted by using absorbance versus pyrolysis temperature and given in Fig. 1. Pyrolysis temperatures varied between 600°C and 1500°C for Pb.

3. Results and discussion

3.1. Thermal stabilizing effects of modifiers

The optimum temperatures and times for drying, pyrolysis and atomization steps were studied in the

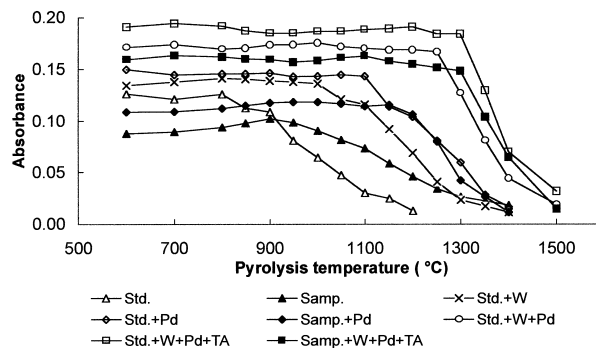


Fig. 1. Comparison of matrix modification studies for Pb in standard (50 ng/ml of Pb) and in sample with modifiers: 20 µg of W, 4 µg of Pd, 20 µg of W and 4 µg of Pd, 20 µg of W and 4 µg of Pd and 200 µg of TA.

absence or presence of modifiers by using aqueous Pb standard. The results are shown in Table 1.

The main aim of using a modifier in an ETAAS is to stabilize volatile elements so that they tolerate higher pyrolysis temperatures and to volatilize the matrix components in the sample efficiently, prior to atomization of the analyte. By using the higher tolerable pyrolysis temperature, minimum matrix interferences on the analyte are encountered in the atomization step (Bin & Zhe-Ming, 1996). Thermal pretreatment curves of Pb in aqueous standard (Std.) and the sample (Samp.) in the presence or absence of modifiers were obtained at the same conditions and compared (Fig. 1). It was observed that pyrolysis temperature for Pb in the sample was higher than aqueous standard in the absence of a modifier or presence of Pd because of the behavior of the sample matrix as a modifier. When W + Pd + TA was used, the same pyrolysis temperature was obtained for the sample and standard. Thermal pretreatment curves and maximum pyrolysis temperatures (T_{max}) for aqueous Pb standard and the sample with W, Pd and W + Pd were compared with the literature (Acar et al., 1997; Havezov et al., 1995; Slaveykova & Tsalev, 1990; Tsalev & Slaveykova, 1992) and similar stabilizing effects were observed.

It is desirable to use a higher pyrolysis temperature in order to remove the matrix efficiently and a temperature of at least 1000°C should be aimed at for many analytes in food, biological and environmental samples (Bin & Zhe-Ming, 1996; Lynch & Littlejohn, 1989; Zong et al., 1996). As can be seen in Fig. 1, maximum pyrolysis temperature for Pb (~1300°C) was obtained with W + Pd + TA by using aqueous Pb standard and sample.

It was found that the addition of tartaric acid (TA) as a reducing agent together with W + Pd has an additional effect on pyrolysis temperature of Pb. In the presence of 200 µg of TA together with W + Pd, pyrolysis temperature for Pb is higher by 50°C. The addition of reducing agents has proved very efficient in reducing the modifiers and analytes to a highly dispersed and reactive

metallic form, particularly in the presence of halides (Byrne, Chakrabarti, Gilchrist, Lamoureux & Bertels, 1993; Gilchrist, Chakrabarti, Byrne & Lamoureux, 1990). The effect of tartaric acid has been discussed in our previous papers (Acar et al., 1997; Acar, Kiliç & Türker, 1999). TA may increase the reduction of modifiers and analytes and prevent the loss of analyte elements as their chloride forms by removing the chlorides as HCl (g) at low temperatures.

3.2. Effects of modifiers on the atomization profiles of Pb in the sample

One of the advantages of chemical modification is that the atomization signals of analytes become fairly symmetrical and shifted to higher pretreatment temperatures and times (Acar et al., 1999; Shan & Wen, 1995; Welz, Schlemmer & Mudakavi, 1992). Shan and Wen (1995) demonstrated this behavior by comparing the shapes of atomization signals of various elements in the absence or presence of Pd, Pd-ascorbic acid and Pd-Mg modifiers.

In order to demonstrate how the W, Pd, W+Pd, W+Pd+TA affect the atomization profiles and background absorbance values of Pb in samples, a comparative study was conducted by using integrated absorbance mode. The atomization and background profiles of Pb in the cookie sample obtained in the absence or presence of modifiers were investigated. As can be seen in Fig. 2, the atomic signals appeared at an earlier time in the absence of a modifier. When W, W+Pd or W+Pd+TA was included, the appearance times of atomization signals were shifted to a later time while increasing pyrolysis temperature and no reduction in atomic absorption signals was observed at maximum pyrolysis temperature obtained from Fig. 1. A small analyte signal and higher background absorbance were obtained in the absence of a modifier. When W+Pd+TA was used as a matrix modifier, it was observed that the absorbance values of Pb in samples increased while the background absorbance was decreasing. As can be seen in Fig. 2, higher signal/noise ratio of Pb was obtained in the presence of W+Pd+TA. It may be due to early reduction of analyte and modifiers to reactive metallic forms. Tartaric acid may reduce the modifiers and analytes to their free metals at pyrolysis temperatures less than 800°C (Acar et al., 1999; Shan & Wen, 1995) and, therefore, the stabilizing effect of the modifier is increased.

3.3. Analytical performance of the method

For the determination of analytical performance of the proposed method, accuracy, precision, limit of detection (LOD), characteristic mass (m_0), correlation coefficient of calibration curve (r) and dynamic analytical range have been evaluated.

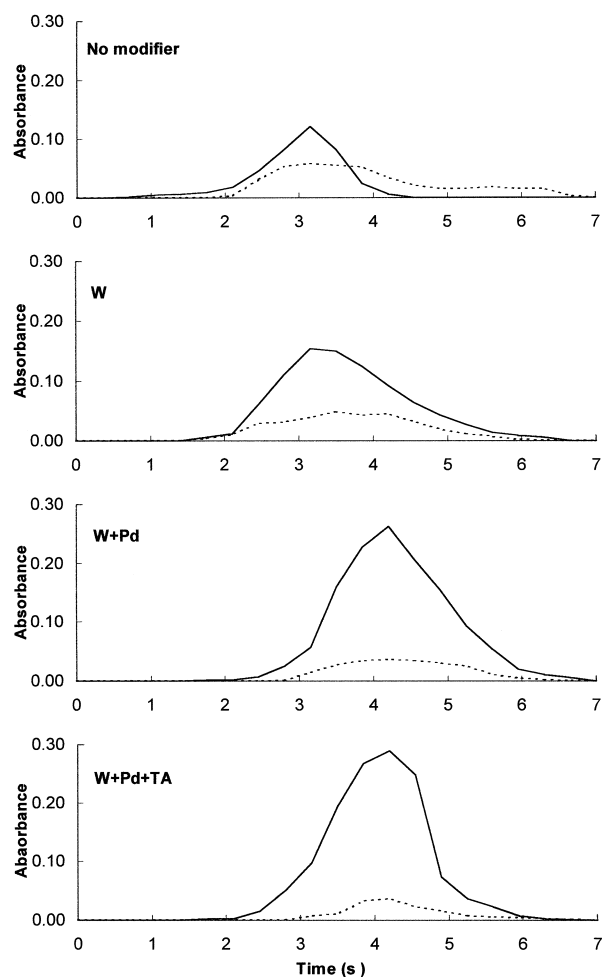


Fig. 2. Comparison of atomization and background profiles and appearance times for Pb in sample by using integrated mode (—, atomization; , background).

Characteristic mass is defined as mass of sample corresponding to 0.0044 abs. unit. LOD is also defined as concentration of solution corresponding to the three folds of standard deviation of the absorbance measurements. Limits of detection and characteristic masses were determined in the presence or absence of modifiers and given in Table 2. As can be seen in Table 2, the lowest detection limit and characteristic mass were obtained by using W+Pd+TA mixed modifier.

Table 2
Detection limits (LOD, 3 σ -criterion) and characteristic masses (m_0) for Pb determination using some modifiers^a

Matrix modifiers	LOD, $\mu\text{g/l}$	M_0 , pg
No modifier	5.34	38.91
W	4.23	32.50
W+Pd	2.57	29.71
W+Pd+TA	1.20	23.72

^a Mean of 10 measurements with 95% confidence level.

The determination of Pb in sample solutions was performed with W + Pd + TA using the calibration on the base of a single element solution. Calibration against working standard solution of Pb in the presence of W + Pd + TA was obtained by using optimum parameters such as a furnace temperature program (Table 1), mass ratio of mixed modifier as described in Acar et al. (1997). The calibration graph was linear ($r > 0.999$) in the range of 5–80 $\mu\text{g/l}$ of Pb.

To demonstrate accuracy of the method, the recovery studies have been carried out with a cookie sample solution and a standard reference material (V-10, Hay powder). The cookie sample solution was analyzed before and after the addition of standard lead solution. Each determination has been repeated eight times in the sample solution in order to determine repeatability of the method. Results are given in Table 3.

The accuracy and the precision of the proposed method were highly good. Relative error was about 3% and the relative standard deviation was lower than 5%.

3.4. Application

The recommended modifier mixture, W + Pd + TA, was applied to determine the lead in Turkish cookies by using the optimized furnace programme (Table 1) and pyrolysis temperature (1300°C) obtained with W + Pd + TA (Fig. 1). The results were given in Table 4. Especially W + Pd + TA, which allows the use of a pyrolysis temperature up to 1300°C for lead could be enough to volatilize many matrix constituents without loss of analyte elements (Acar et al., 1997). The results of Pb in samples found were compared with maximum permissible values given by Joint FAO/WHO standard programme (Joint FAO/WHO, 1983) and Turkish Standards (TS 2383, 1991; TS 12299, 1997). As can be seen in Table 4, four of 24 cookie samples (8–11) are within 2% of the legal limit. It can be said that this brand contains a high level of lead. It was generally observed that the results obtained were not higher than the maximum permissible values.

Table 3
Precision and accuracy of the method (in the presence of W + Pd + TA)

Sample	Unit	Pb concentration			Recovery (%)
		Certified value	Added	Found ^a	
Sample solution of a cookie	$\mu\text{g/l}$	–	–	35.4±1.2	–
	$\mu\text{g/l}$	–	20.0	53.6±2.6	97
	$\mu\text{g/l}$	–	37.5	72.4±3.0	99
Hay powder (V-10)	$\mu\text{g/g}$	1.6	–	1.61±0.08	101

^a Mean of eight replicate measurements of one sample solution with 95 % confidence level.

Table 4
Determination of lead in Turkish cookies^a

Sample no.	Pb concentration ^b , ng/g, $\bar{X} \pm \frac{ts}{\sqrt{n}}$
1	181±6
2	265±9
3	194±8
4	145±4
5	122±4
6	255±10
7	192±6
8	305±11
9	303±12
10	298±10
11	294±9
12	141±5
13	112±4
14	172±8
15	194±9
16	210±9
17	220±10
18	240±11
19	210±8
20	200±12
21	221±9
22	264±11
23	196±9
24	232±9

^a Maximum permissible values of lead given in Turkish Standards (TS 2383, 1991; TS 12299, 1997) is 300 ng/g.

^b Mean of eight replicate measurements of one sample solution with 95% confidence level.

4. Conclusions

Determination of lead in some cookies by ETAAS using W + Pd + TA modifier mixture was examined. In general terms, the analytical problems arising from sample matrix can be controlled by using W + Pd + TA modifier mixture and ETAAS with Zeeman background correction. Recovery and detection limit of lead were about 99% and 1.2 $\mu\text{g/l}$, respectively. W + Pd + TA modifier mixture with ETAAS might be applied for the determination of lead at lower concentrations in food and biological samples.

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