Available online at www.sciencedirect.com
SCIENCE DIRECT®
Talanta 64 (2004) 258–263
Talanta
www.elsevier.com/locate/talanta

Determination of lead in vinegar by ICP-MS and GFAAS: evaluation of different sample preparation procedures

Kuria Ndung'u [a,b,*], Sharon Hibdon [a], A. Russell Flegal [a]

[a] *Environmental Toxicology, WIGS University of California, Santa Cruz, CA 95064, USA*
[b] *Institute of Applied Environmental Research (ITM), Stockholm University, Frescativagen 54, S-106 91 Stockholm, Sweden*

Received 4 November 2003; received in revised form 12 February 2004; accepted 12 February 2004

Available online 17 April 2004

Abstract

Lead concentrations of 59 different types of vinegars (15–307 $\mu g\,l^{-1}$ in balsamic vinegars and 36–50 $\mu g\,l^{-1}$ in wine vinegars) were determined using both inductively coupled plasma mass spectrometry (ICP-MS) and graphite furnace atomic absorption spectrometry (GFAAS). Although the precision of direct analyses, following simple aqueous dilutions, with either instrumental method was poor; that precision, following nitric acid and/or hydrogen peroxide digestions, markedly improved with either instrument and the values obtained with the two instruments were in good agreement. The efficacy of different digestions, including (1) nitric acid using a heating block, with or without addition of hydrogen peroxide and (2) mixtures of nitric acid and hydrogen peroxide using ultraviolet (UV) photolysis, were then assessed. The latter procedure was found to be much faster and more efficient, but it was limited by the relatively high levels of contaminant lead in hydrogen peroxide. Consequently, it is recommended that lead concentrations in vinegar be measured following a nitric acid digestion and UV photolysis to oxidize all organic matter before ICP-MS or GFAAS analysis; and it is further recommend that the thermal settings for the latter analyses be adjusted to account for the apparent presence of relatively volatile organolead compounds in vinegar digests.

Keywords: Organolead compounds; Vinegar; ICP-MS

1. Introduction

Exposure to contaminant lead remains a public concern because of its pervasiveness in the environment and increasing evidence of lead's sub-lethal toxicities at exposure levels lower than previously thought harmful [1]. In response to those concerns, there have been orders of magnitude reductions in atmospheric emissions of industrial lead, which have resulted in a pronounced decrease in blood lead levels in the US and elsewhere [2]. Now, the most common route of exposure to the general population, in countries where leaded gasoline has been banned, is through the ingestion of food and water contaminated with lead [3].

Among those foods is vinegar, which can contain relatively high levels of lead [4,5]. It may, like wine, come from the grapes vinegar is made from and it might be of endogenous or anthropogenic origin [6,7]. Conversely, the lead may come from contamination during the vinegar production process [8].

Although there are numerous published studies on the concentration of lead in wine, only a handful of studies have looked at the concentration of lead in vinegar [4,5,9,10]. While some of those studies measured the lead in vinegar or wine directly after simple dilution [10–12], quite often a sample clean-up step was employed prior to the instrumental analysis. This pretreatment is often needed because, in addition to acetic acid and alcohol, both vinegar and wine contain, suspended particles and polymeric organic compounds, particularly sugars, which interfere with GFAAS and ICP-MS measurements. The polymeric organic matter might cause blockage of the injector tube and cones of the ICP, due to incomplete pyrolysis of the sugars in the plasma and formation of residual carbon deposits [13]. During the GFAAS analysis, incomplete pyrolysis of the organic matter produces fumes and accumulation of carbonaceous residue

* Corresponding author. Tel.: +46-86747236; fax: +46-86747636.
E-mail address: kuria.ndungu@itm.su.se (K. Ndung'u).

0039-9140/$ – see front matter © 2004 Elsevier B.V. All rights reserved.
doi:10.1016/j.talanta.2004.02.017

after several graphite tube firings which adversely affect the analysis [11].

Two types of oxidation are most common: acidification and irradiation. Wet digestion using nitric acid is usually employed to oxidize the organic matter, and those oxidative digestions are often accelerated by heating the samples in Teflon or other inert and trace metal clean containers on a heating block or heating plate. The addition of hydrogen peroxide also speeds up the oxidation process, but most peroxides contain relatively high amounts of lead. Alternatively, ultraviolet (UV) and/or microwave energy have also been used to oxidize the organic matter in wine [13,14] which is a precursor of many vinegars. Since UV photolysis has not previously been applied to vinegar digestions, and the relative accuracy and efficacy of the different analytical methods for measuring lead in vinegar have not been previously determined.

2. Background

Vinegar is produced by a two-stage fermentation process of suitable sugar or starch containing agricultural material such as grapes, apples, rice, garlic or even onions [15]. Besides vinegar from red and white wine, there are special products such as vinegar from Jerez (Sherry vinegar) in Spain or balsamic vinegar elaborated from a specific region of Italy, Modena [15] Aceto Balsamico di Modena, a typical Italian product is produced from fresh grape must, which is concentrated up to a third of its original volume by a slow heating process. The traditional method of production requires storage in different wood barrels up to 25 years. Another balsamic vinegar is produced by blending the concentrated must with acetic acid, and the mixture is allowed to mature in wooden barrels to develop the typical organoleptic properties [15].

Consequently, there may be pronounced differences in the organic composition of different types of vinegars, including different balsamic vinegars. There may also be large differences in the lead concentrations of different vinegars, based on the origins of the ingredients and the production process. Both of those variables complicate accurate and precise measurements of lead in vinegar.

3. Experimental

3.1. Reagents

All solutions were prepared with de-ionized water ($18\,M\Omega\,cm^{-1}$) from a Milli-Q® analytical reagent-grade water purification system (Millipore, Bedford, MA). Calibration standard solutions and internal standards were prepared from commercial lead standard solution (Spex Plasma, Edison, NJ). Trace metal grade (TMG) nitric acid and hydrochloric acid (Fisher Scientific, Pittsburgh, PA) were used for cleaning laboratory ware. Optima grade nitric acid (Fisher) was used for the preparation of calibration standard solutions and analytical solutions. High purity hydrogen peroxide 30% (Ultrapur, Bayer, Pittsburg, NJ), together with nitric acid was used for both heat and UV digestions. The matrix modifier used for GFAAS analysis contained 0.05 mg of $NH_4H_2PO_4$ and 0.003 mg of $Mg(NO_3)_2$ per 5 μl of solution (Environmental Express, Mt. Pleasant, SC).

3.2. Instrumentation

3.2.1. ICP-MS

All ICP-MS measurements were made with a Thermo-Finnigan Element magnetic sector high resolution ICP-MS using a Glass Expansion Conikal nebulizer, a Scott-type double pass spray chamber (cooled to 10 °C) and standard nickel cones. Since there were small or no polyatomic interferences for lead, it was analyzed at low resolution ($r = 300$) using ^{209}Bi as an internal standard. The instrument operating parameters and data acquisition details are listed in Table 1.

3.2.2. GFAAS

Graphite furnace atomic absorption spectroscopy (GFAAS) analyses were made on a Perkin-Elmer SIMAA 6000 instrument, fitted with a Zeeman background corrector and AS72 auto sampler. End capped, traversely heated pyrocoated graphite tubes with an integrated L'vov platform (Perkin-Elmer) were used. A lead electrodeless discharge lamp (Perkin-Elmer) was used at the recommended line of 283.3 nm and a lamp current of 450 mA. Magnesium nitrate ($Mg(NO_3)_2$)/ammonium phosphate ($NH_4H_2PO_4$) was

Table 1
ICP-MS operating and acquisition parameters

RF power (W)	1250
Plasma gas flow ($l\,min^{-1}$)	13
Auxiliary gas flow ($l\,min^{-1}$)	0.75
Nebulizer gas flow ($l\,min^{-1}$)	0.85–0.95 (optimized daily)
Sample flow rate ($\mu l\,min^{-1}$)	60

Data acquisition (low resolution, 200 scans)

Isotope	% mass window	Sample time (s)	Samples/peak	Segment duration (s)	Detection mode
^{208}Pb	5	0.001	100	0.050	Count
^{209}Bi	5	0.001	100	0.050	Count

Table 2
Optimized GFAAS program for measuring lead concentrations (ug l^{-1}) in vinegar, following acid, UV and/or microwave digestions

Temperature (°C)	Ramp time (s)	Hold time (s)	Gas flow (ml min^{-1})	Read
110	5	30	250	No
130	15	30	250	No
700	15	30	250	No
1400	0	3	0	Yes
2450	1	3	250	No

used as a chemical modifier. The optimized, based on tests conducted for this report (see following section on GFAAS Program Optimization) GFAAS program used is shown in Table 2.

3.3. Samples

Vinegar samples were purchased from retail stores in California. Fifty-two different types of balsamic vinegar, four wine vinegars, one apple cider vinegar, one rice vinegar and one garlic vinegar were analyzed. The vinegars were mostly in glass bottles, but some were in plastic or ceramic bottles.

3.4. Contamination control

The exteriors of the bottles were rinsed with deionized water before opening in a HEPA filtered (Class 100) trace metal clean laboratory. Aliquots were placed in Teflon digestion vessels that were cleaned with Micro-90 liquid laboratory grade detergent (Cole-Parmer, Vernon Hills, IL) and deionized water when first used or after an incomplete digestion. Subsequently, digestion vessels were re-cleaned by soaking them overnight in 8 M TMG hydrochloric acid followed by at least 8 h in hot TMG nitric acid. The vessels were then rinsed with reagent water and dried under class 100 HEPA-filtered laminar flow air. All other plastic ware (polyethylene or Teflon) used for storing analytical solutions were cleaned using the same procedure, dried, capped, and stored under class 100 HEPA-filtered laminar flow air or double bagged in trace metal clean, self-locking (Zip loc®) plastic bags. The GFAAS was in a HEPA-filtered air room and directly beneath a HEPA-filtered (Class 100) laminar flow canopy within a plastic enclosure.

3.5. Vinegar digestions

3.5.1. Heating block digestions

Analytical portions were weighed (0.5–1.0 g) into Teflon digestion vessels, and 10 ml of TMG nitric acid was added. Vessels were covered loosely with acid cleaned Teflon lids and placed in the heating block (CPI International, Santa Rosa, CA). They were initially digested at 50 °C for 2–3 h to avoid sputtering then the temperature was increased to 90 °C, and then digested to dryness. After cooling, the digests were dissolved in 1 M TMG nitric acid, producing a clear to light yellow analytical solutions. These were then analyzed for their lead concentration by GFAAS or ICP-MS.

3.5.2. UV digestions

The UV digestion unit consisted of a medium pressure mercury vapor discharge tube (1200 W; Hanovia, Union, NJ) positioned on the ceiling of a purpose-built aluminum housing, (36 cm × 29 cm × 23 cm; UVO-cleaner model 342, Jelight Inc., Laguna Hills, CA), which was cooled by a fan. A digital photometer (model JL1400A, Jelight Inc., Irvine, CA) was used to monitor the power of the UV radiation during the oxidation ($x = 9.2 \pm 0.4$ mW cm^{-2} during the continuous operation of the Hg lamp).

The digestions were carried out by placing 16 custom-made PTFE 15 ml digestion cups fitted with quartz glass caps in the UV digestion unit.

Vinegar samples (0.5 g) were weighed in tarred Teflon vials. These and 1 ml of TMG nitric acid and 0.5 ml of 30% hydrogen peroxide were added prior to the UV treatment.

3.6. Quality control

Sample batches consisted of 24 analytical portions including several duplicate samples. Spikes of lead were added (90–150 μg l^{-1}) prior to digestion to several vinegar analytical portions representative of the variety of products. Standard solutions were analyzed after every 10 analytical solutions to ensure instrument performance. Each analytical batch contained at least three method blanks, three spiked analytical samples, and three reference materials. Because there is no commercially available certified reference material for lead in vinegar (or wine), we used the National Institute of Standards and Technology (NIST) 1640 Standard Reference Material (SRM) for trace metals in natural waters (NIST, Gaithersberg, MD) with a lead concentration (where X is the mean ± S.D.) of 27.89 ± 0.14 μg l^{-1} to monitor the extraction efficiency of the digestion process.

4. Results and discussion

4.1. Nitric acid digestion

As previously noted, only a small number of studies have been published on the determination of lead in vinegar [4,5,9,10]. Most of them have employed a sample pretreatment to destroy the organic matter, which might interfere with GFAAS or ICP-MS analyses. In contrast, a few studies have reported direct analysis of lead in wine by GFAAS [16] or ICP-MS [11,17] after a simple aqueous dilution.

However, our attempts to analyze vinegar with or without dilution by either GFAAS or ICP-MS resulted in erroneously high lead concentration values (compared to nitric acid digested vinegar) and relatively poor precision. This analytical variability is illustrated in Table 3, which is a summary of the lead determination in four different types of balsamic

Table 3
Comparison of lead concentrations in four different balsamic vinegars analyzed by GFAAS and ICP-MS with and without nitric acid digestion

Vinegar	Lead concentration[a] ($\mu g\,l^{-1}$)			
	Simple dilution		Digested with nitric acid	
	GFAAS	ICP-MS	GFAAS	ICP-MS
Balsamic-1	595 (18)	447 (7)	319 (9)	306 (6)
Balsamic-2	653 (14)	205 (5)	198 (7)	174 (2)
Balsamic-3	277 (38)	68 (16)	61 (7)	60 (5)
Balsamic-4	349 (4)	109 (17)	99 (9)	95 (4)

[a] Mean and relative standard deviation (values in parenthesis) of at least six determinations.

vinegars (six replicate digestions or analyses). Because of their complex organic content, those vinegars proved to be the most difficult to analyze by either GFAAS or ICP-MS and with and without a prior nitric acid digestion.

Specifically, measurements with both types of instruments yielded significantly ($P \leq 0.05$, paired t test) higher lead concentrations in balsamic vinegars after simple aqueous dilutions compared to measurements after acid digestions. The disparity was greater in direct analyses of undigested diluted vinegars by GFAAS. In addition to vinegar matrix interferences, we noticed irreproducible sample deposition on the graphite tube due to adhesion of vinegar solutions to the Teflon GFAAS deposition tubing. Moreover, this problem persisted after filtering and diluting the vinegars.

The agreement and precision of the analyses between the two instruments was greatly improved ($R = 0.997$, $m = 0.94$, simple linear regression) after nitric acid digestions. These improvements are attributed to the oxidation of organic matter. That destruction eliminates interferences resulting from nonspecific absorption and scattering of light due to concomitant species in the vinegar solutions.

4.2. UV and heat digestion

Nitric acid, and to a lesser extent hydrogen peroxide, are widely used for wet digestions of organic and inorganic matter prior to instrumental analyses of metals. The oxidative digestions are accelerated by heating the samples in Teflon or other inert, trace metal clean containers on a heating block or heating plate. Those thermal energy sources are now often being replaced by microwave and UV radiation in sample preparations where acid digestion is necessary, because they may be faster and may be done within a closed system [13,18].

Comparing the two methods, nitric acid digestions with UV radiation were faster than those with heating blocks and the digestions were more complete. While the addition of hydrogen peroxide further enhanced the degradation of organic material in the vinegars, the amount of contaminant lead in TMG hydrogen peroxide we used was relatively high ($\sim 15\,\mu g\,l^{-1}$) and comparable to the lead concentration in some of the vinegars. Thus, cleaner hydrogen peroxide is necessary for digestion of vinegars with lead concentration in the low to sub $\mu g\,l^{-1}$ level.

4.3. GFAAS analysis

Although the instrument manufacturer (Perkin-Elmer) recommended a maximum ashing and atomization temperatures of 400 and 1400 °C, respectively, in the furnace program for lead determination, the use of chemical modifiers allows much higher ashing and atomization temperatures. Freschi et al. [11] used an ashing temperature of 1000 °C and an atomization temperature of 1800 °C to determine lead in diluted wine samples and nitric acid wine digests using a phosphate/magnesium matrix modifier. Buldini et al. [19] also used a phosphate/magnesium modifier and were able to determine lead in nitric acid wine digests using ashing and atomization temperatures of 900 and 1800 °C, respectively.

In the absence of a vinegar or similar matrix SRM with certified lead concentration, we initially started the optimization of the furnace program using digested vinegar spikes and NIST SRM 1640 (natural water) that had undergone a similar nitric acid digestion process as the vinegars. We used the manufacturers recommended ashing and atomization temperatures with a $Mg(NO_3)_2/NH_4H_2PO_4$ chemical modifier. We found ashing and atomization temperatures of 800 and 1400 °C, respectively, to be optimum for analysis of digested natural water SRM and quantitative recovery. However, the same furnace program produced low lead recoveries of spiked digested vinegar samples.

An investigation of the GFAAS measurements of vinegar digests with similar lead concentrations as the SRM showed a sharp drop in absorbance between 700 and 800 °C of the digested vinegar samples, but not for the SRM. This disparity is shown in Fig. 1. It contains plots of the variation of absorbance during ashing and atomization temperatures steps of the two types of samples.

Curvatures in both plots indicate the digestion of the vinegar samples produced a relatively labile lead compound(s). Their volatilization between 700 and 800 °C markedly altered the measurements of lead concentrations of the vinegar, which was not replicated in the measurements of lead in the SRM. This thermal variability underscores the importance of close investigation of the furnace program optimization for different sample types and matrices.

4.4. Quality control

Process blanks (reagent water) were also analyzed together with the samples. The mean blank lead concentration was $0.03\,\mu g\,l^{-1}$ ($n = 4$) with a standard deviation of 0.04, giving a detection limit of $0.12\,\mu g\,l^{-1}$ analyzed by GFAAS after nitric acid and heat digestion. The spike recovery ($x \pm$ S.D.) of six different vinegars was $96 \pm 5\%$, while the mean recovery of NIST 1640 SRM digests was $97.4 \pm 1.3\%$. The relative standard deviation for duplicate analysis was <8%.

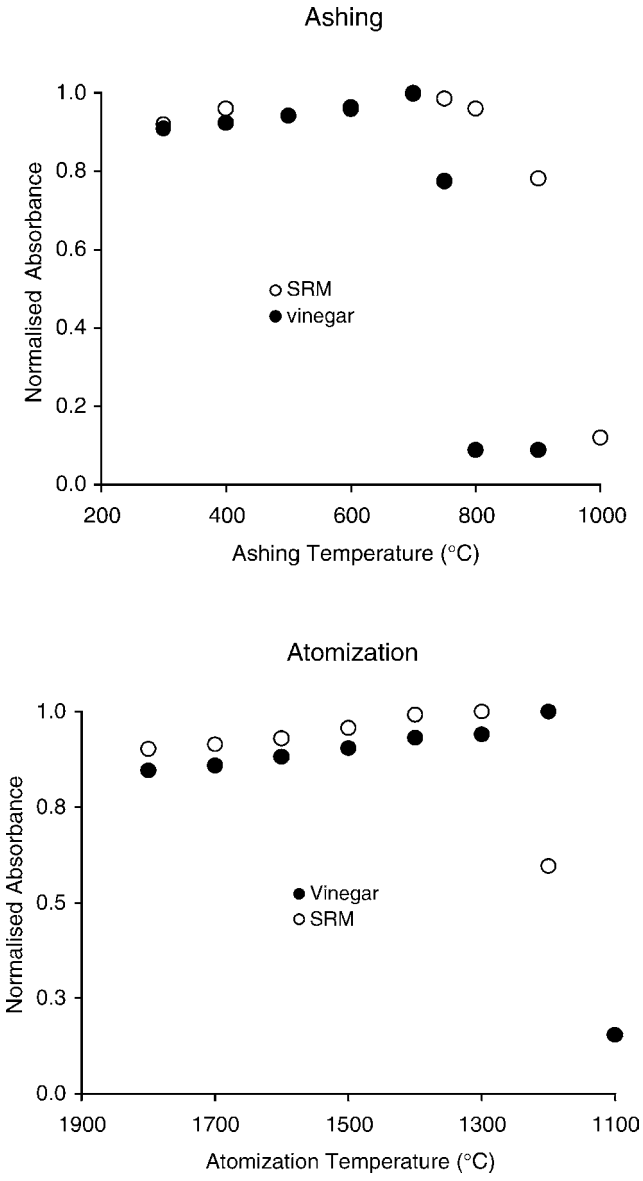

Fig. 1. Variation of absorbance with GFAAS ashing and atomization temperatures for NIST 1640 natural water SRM and a balsamic vinegar after nitric acid (65% m/m) digestion.

4.5. Lead concentrations in vinegars

Table 4 summarizes the results of the measurements. Again, these included analyses of lead concentrations in 52 different types of balsamic vinegars, 4 wine vinegars, 1 apple vinegar, 1 garlic, and 1 rice vinegar. The concentration of lead in the balsamic vinegars ranged from 14.9 to 307 $\mu g\,l^{-1}$, with a mean of $68 \pm 56\,\mu g\,l^{-1}$.

It is notable that the balsamic vinegar ($n = 6$) with the highest lead concentration ($307 \pm 19.5\,\mu g\,l^{-1}$) was, reportedly, produced by the traditional method of production, which involved aging in different vintage wood barrels. Another balsamic vinegar with a high lead concentration ($257\,\mu g\,l^{-1}$) had, reportedly, been aged for 18 years. We, therefore, hypothesize that most of the lead in those vinegars comes from their contact with contaminant lead surfaces during production and storage, possibly metal fittings securing the barrels used to age the vinegars.

Table 4
Lead concentrations in vinegar

Type of vinegar	Number of brands	Lead concentration ($\mu g\,l^{-1}$)		
		Range	Mean	Standard deviation
Balsamic	36	15–68	44	15
	12	73–110	85	12
	2	174–179	173	4
Balsamic (aged)-1	1		276	
Balsamic (aged)-2	1		307	
Wine	4	36–62	50	12
Garlic	1		15	
Apple	1		6.6	
Rice	1		19	
All vinegars	59	7–307	64	54

All measurements are means of at least four or six replicate determinations.

This hypothesis is based on the results of several studies of the sources of lead contamination in grape wine [6,7,20,21], because there are no published studies on the sources of lead contamination in vinegar. Three of the wine studies [7,20,21] found the production and storage process to be the main source of lead contamination rather than the grape or the soil. For example, Almeida and Vasconcelos [20] found that only about a quarter of the total content of lead in Portuguese fortified and red table wines, respectively, came from soil and atmospheric deposition and that the rest of the lead was introduced in the vinification processes. They concluded that marked reductions of the lead content in the wines would occur if the sources of lead were removed from the tubes and containers used in the vinification system, particularly by using welding alloys and lead free fittings. They also observed that wines produced via traditional vinification methods had a higher lead concentration compared to those produced by modern technology.

5. Conclusions

Lead concentrations in different types of vinegar vary from less than 10 to more than $300\,\mu g\,l^{-1}$, and the accuracy and precision of those lead concentration measurements with different protocols may also vary widely. Direct determinations of lead in vinegar by ICP-MS and GFAAS may provide irreproducible measurements and give poor agreement between the two types of instrumental analysis, but there is good agreement in measurements with the two types of instrumental analysis after vinegar is digested with nitric acid. While use of nitric acid and evaporative digestion in heating blocks is an effective and clean method of processing vinegars prior to ICP-MS and GFAAS analysis, nitric acid digestions with UV photolysis reduce the time required

to process the vinegar and increase the oxidation of its organic constituents. Although the digestion times may be further enhanced with the addition of hydrogen peroxide, the amount of contaminant lead in TMG hydrogen peroxide is too high for measurements of lead concentrations in vinegars with concentrations $<50\,\mu g\,l^{-1}$. Therefore, we recommend nitric acid digestion of vinegars before ICP-MS or GFAAS determination, and that the latter measurements use ashing and atomization temperatures of 600 and 1300 °C, respectively, rather than the manufacturer's recommended settings because of the apparent volatilization of relatively labile forms of lead in vinegars above those temperatures.

Acknowledgements

We are grateful to Ana Gonzalez for help with vinegar digestion and Rob Franks for help with the ICP-MS analysis. This research was funded by Environmental Law Foundation.

References

[1] K.D. Rosenman, A. Sims, Z.H. Luo, J. Gardiner, J. Occup. Environ. Med. 45 (2003) 546.
[2] CDC, Second National Report on Human Exposure to Environmental Chemicals (2002), CDC, Centers for Disease control and Prevention, 2003, http://www.cdc.gov/exposurereport/metals/, accessed on 6 October 2003.
[3] CDC, Toxicological Profile Information: Toxic Profile for Lead, CDC, Centers for Disease Control and Prevention, Agency for Toxic Substances and Desease Registry (ATSDR), 1999, http://www.atsdr.cdc.gov/toxprofiles/, accessed on 6 October 2003.
[4] F. Corradini, L. Marcheselli, A. Marchetti, C. Preti, C. Biancardi, J. Aoac. Int. 77 (1994) 714.
[5] A. Acosta, C. Diaz, A. Hardisson, D. Gonzalez, Environ. Contam. Tox. 51 (1993) 852.
[6] V.R. Angelova, A.S. Ivanov, D.M. Braikov, J. Sci. Food Agric. 79 (1999) 713.
[7] V. Orescanin, A. Katunar, A. Kutle, V. Valkovic, J. Trace Microprobe Tech. 21 (2003) 171.
[8] M.I. Guerrero, C. Herce-Pagliai, A.M. Camean, A.M. Troncoso, A.G. Gonzalez, Talanta 45 (1997) 379.
[9] A. Del Signore, B. Campisi, F. Di Giacomo, J. Aoac. Int. 81 (1998) 1087.
[10] Z.J. Suturovic, N.J. Marjanovic, N.M. Dostanic, Nahrung 41 (1997) 111.
[11] G.P.G. Freschi, C.S. Dakuzaku, M. de Moraes, J.A. Nobrega, J.A.G. Neto, Spectrochim. Acta B 56 (2001) 1987.
[12] A.M.T. Gonzalez, M.G. Chozas, Nahrung 32 (1988) 743.
[13] C.M.R. Almeida, M. Vasconcelos, J. Anal. Atom. Spectrom. 14 (1999) 1815.
[14] C.R. Quetel, S.M. Nelms, L. Van Nevel, I. Papadakis, P.D.P. Taylor, J. Anal. Atom. Spectrom. 16 (2001) 1091.
[15] W. Tesfaye, M.L. Morales, M.C. Garcia-Parrilla, A.M. Troncoso, Trends Food Sci. Technol. 13 (2002) 12.
[16] Z.Y. Zuo, M. Zhang, Z.A. Sun, D.S. Wang, Spectrosc. Spectr. Anal. 22 (2002) 859.
[17] C.M.R. Almeida, M. Vasconcelos, M. Barbaste, B. Medina, Anal. Bioanal. Chem. 374 (2002) 314.
[18] Q.H. Jin, F. Liang, H.Q. Zhang, L.W. Zhao, Y.F. Huan, D.Q. Song, Trace Trends Anal. Chem. 18 (1999) 479.
[19] P.L. Buldini, S. Cavalli, J.L. Sharma, J. Agric. Food Chem. 47 (1999) 1993.
[20] C.M.R. Almeida, M. Vasconcelos, J. Agric. Food Chem. 51 (2003) 3012.
[21] J. Kristol, M. Veber, M. Slekovec, Anal. Bioanal. Chem. 373 (2002) 200.