Talanta 76 (2008) 575–579

Contents lists available at ScienceDirect

Talanta

journal homepage: www.elsevier.com/locate/talanta

Simultaneous determination of Cd, Cu, Mn, Ni, Pb and Zn in nail samples by inductively coupled plasma mass spectrometry (ICP-MS) after tetramethylammonium hydroxide solubilization at room temperature: Comparison with ETAAS

Bruno L. Batista[a], Jairo L. Rodrigues[a], Juliana A. Nunes[a], Luciano Tormen[b], Adilson J. Curtius[b], Fernando Barbosa Jr.[a,*]

[a] *Laboratório de Toxicologia e Essencialidade de Metais, Depto. de Análises Clínicas, Toxicológicas e Bromatológicas, Faculdade de Ciências Farmacêuticas de Ribeirão Preto-USP, Avenida do Café s/n, Monte Alegre 14040-903, Ribeirão Preto-SP, Brazil*
[b] *Universidade Federal de Santa Catarina, Depto. Química, Campus Trindade, 88040900 Florianópolis, SC, Brazil*

ARTICLE INFO

Article history:
Received 14 February 2008
Received in revised form 24 March 2008
Accepted 26 March 2008
Available online 7 April 2008

Keywords:
ICP-MS
ETAAS
Nail samples
Trace elements
Rapid determination
Tetramethylammonium hydroxide

ABSTRACT

A simple method is described for the determination of Cd, Cu, Mn, Ni, Pb and Zn in nails by using inductively coupled plasma mass spectrometry (ICP-MS) or electrothermal atomic absorption spectrometry (ETAAS). Prior to analysis, 10–20 mg of nail samples were accurately weighed into (15 mL) conical tubes. Then, 1 mL of 25% (w/v) tetramethylammonium hydroxide (TMAH) solution was added to the samples, incubated at room temperature overnight and then further diluted to 10 mL with 1% (v/v) HNO_3. After that, samples were directly analyzed. Rhodium was used as internal standard for ICP-MS analysis. Method detection limits (3 s, $n = 20$) were 0.1, 3.0, 1.0, 4.5, 1.5, 5.0 ng g^{-1} for Cd, Cu, Mn, Ni, Pb and Zn, respectively for ICP-MS, and 24, 26, 30, 143, 130 and 1000 ng g^{-1}, respectively for ETAAS. The key issue addressed here is the elimination of the acid digestion prior to analysis. Moreover, with the use of the proposed method there is a considerable improvement in the sample throughput comparing to the traditional methods using microwave-assisted acid sample digestion prior to analysis. For validation purposes, six ordinary nail samples were solubilized and then directly analyzed by ICP-MS and ETAAS, with no statistical difference between the two techniques at 95% level on applying the *t*-test.

1. Introduction

Nail is a biological specimen that has some advantages as a biomarker for trace elements exposure, especially because its collection is noninvasive and simple and because nail samples are very stable after collection, not requiring special storage conditions [1,2]. Trace elements in nails reflect long-term exposure because this compartment remains isolated from other metabolic activities in the body [2,3]. Because toenails are less affected by exogenous environmental contamination than fingernails, they have been preferred for exposure to toxic metals [3]. Toenails have a slower growth rate than fingernails (up to 50% slower, especially in winter) and thus may provide a longer integration of the exposure [2–4].

Atomic absorption spectrometry (AAS) [5,6] and inductively coupled plasma emission spectrometry (ICP-OES) [7] are still the dominant analytical techniques used for trace element analysis in nail samples. However, the use of inductively coupled plasma mass spectrometry (ICP-MS) is becoming much more common in clinical laboratory analysis [3,8–12]. Compared to electrothermal atomic absorption spectrometry (ETAAS) or ICP-OES, this technique has some distinct advantages, including simultaneous multielement measurement capability coupled with very low detection limits [7]. Moreover, it offers a wider linear dynamic range which allows the determination of major and trace elements at same sample injection [8–10]. Additionally, compared to ICP-OES, ICP-MS provides simpler spectral interpretation and isotopic information [8].

Sample preparation is a critical and laborious step for chemical elements determination in biological samples [13–16]. In the case of nails, usually, samples are digested in acid medium prior to analysis. Wet digestion procedures in open [3] or closed vessels with microwave-assisted acid digestion [6,10] has been used for this purpose. However, both methods may be time-consuming for laboratories operating in routine with large amount of samples to be analyzed and require the use of concentrated acids and careful monitoring of digestion [11]. On the other hand, in clin-

* Corresponding author. Tel.: +55 16 36024701.
E-mail address: fbarbosa@fcfrp.usp.br (F. Barbosa Jr.).

0039-9140/$ – see front matter © 2008 Elsevier B.V. All rights reserved.
doi:10.1016/j.talanta.2008.03.046

ical laboratories, more attention has been given to methods of sample preparation with minimal handling and time consumption [17,18].

Nail is composed of keratin proteins. The sulfur in cysteine molecules in adjacent keratin proteins link together in disulfide chemical bonds. These disulfide bonds are very strong and very difficult to break apart. These disulfide chemical bonds linking the keratins together are the key factor in the durability and resistance of nail fiber. They are largely resistant to the action of acids but the disulfide bonds can be broken apart by alkali solutions, making the nail weak. Tetramethylammonium hydroxide (TMAH), an alkaline solution, has been previously used for sample pre-treatment as an attractive alternative to microwave-assisted acid digestion of biological materials for the determination of trace elements by different atomic spectrometry techniques [19–26]. For example, Pozebon et al. [24] used TMAH to dissolve biological samples for the determination of Tl, Pb, Ag, Hg and Cd by electrothermal vaporization inductively coupled plasma mass spectrometry (ETV-ICP-MS) and Moreton and Delves [25] used TMAH in one of their diluents for the determination of total Hg in whole blood by ICP-MS.

The aim of this study was to develop a simple method for the determination of five elements in nail samples by ICP-MS, that can also be applied for ETAAS analysis, using a sample solubilization with TMAH and then a dilute-and-shoot sample analysis.

2. Experimental

2.1. Reagents

All reagents used were of analytical-reagent grade (Sigma, St. Louis, MO, USA), except HNO_3 (Sigma, St. Louis, MO, USA) which was previously purified in a quartz sub-boiling stills (Kürner) before use. A clean laboratory and laminar-flow hood capable of producing class 100 were used for preparing solutions. High purity de-ionized water (resistivity 18.2 mΩ cm) obtained using a Milli-Q water purification system (Millipore, Bedford, MA, USA) was used throughout. All solutions were stored in high-density polyethylene bottles. Plastic bottles and glassware materials were cleaned by soaking in 10% (v/v) HNO_3 for 24 h, rinsing five times with Milli-Q water and dried in a class 100 laminar flow hood before use. All operations were performed in a clean bench.

For the ETAAS method, stock solutions containing 1000 mg L^{-1} of each element were obtained from PerkinElmer (PerkinElmer, Norwalk, CT, USA). Analytical calibration standards were prepared daily over the range of 5–40; 0.5–5; 1.0–50; 1.0–10; 5–50; 0.5–3.0 μg L^{-1} for Pb, Cd, Cu, Mn, Ni and Zn respectively by suitable serial dilutions of each stock solution in 2.5% (w/v) TMAH + 1% (v/v) HNO_3. For Cd and Pb determination, graphite tubes were previously coated with iridium according to the method proposed by Borges et al. [27]. The modifier solution was prepared by the suitable dilution of the 1000 mg L^{-1} Ir stock standard (PerkinElmer, Norwalk, CT, USA).

For the ICP-MS method, multielement stock solutions containing 1000 mg L^{-1} of each element were obtained from PerkinElmer (PerkinElmer, Norwalk, CT, USA). Analytical calibration standards were prepared daily over the range of 0–20 μg L^{-1} for all elements by suitable serial dilutions of multielement stock solution in 2.5% (w/v) TMAH + 1% (v/v) HNO_3. Rhodium was used as internal standard at the concentration of 10 μg L^{-1} Rh. The internal standard was diluted from 1000 mg L^{-1} stock standard (PerkinElmer, Norwalk, CT, USA). A rinse solution consisting of 0.005% (v/v) Triton X-100® in 2% (v/v) nitric acid was prepared before each run.

Table 1
Optimal heating programs for the determination of Cd, Cu, Mn, Ni, Pb and Zn in nails after TMAH solubilization (30 μL injection volume)

Temperature (°C)	Ramp (°C s^{-1})	Hold (s)	Gas flow rate (L min^{-1})
90	5	15	250
120	10	15	250
600[a]/700[b]/800[c]/1000[d,e,f]	20	15	250
20	1	3	250
1500[a]/1600[b,c]/2200[d,e]/2400[f]	0	5	0
1900[b,c]/2000[a]/2500[d,e,f]	1	5	250

[a] Zn.
[b] Cd.
[c] Pb.
[d] Cu.
[e] Ni.
[f] Mn.

2.2. Instrumentation

2.2.1. Electrothermal atomic absorption spectrometric method

Cadmium, copper, lead, manganese, nickel and zinc were determined in nails by using a AAnalyst 100 atomic absorption spectrometer (PerkinElmer, Norwalk, CT, USA), equipped with an HGA 800 longitudinally heated graphite tube atomizer and an AS-72 autosampler (PerkinElmer). Deuterium-arc background correction was employed to correct for non-specific absorption. All measurements were performed using integrated absorbance (peak area). Hollow cathode lamps for Cd, Pb, Ni, Mn, Cu and Zn (PerkinElmer) were operated at 4, 10, 25, 20, 15 and 15 mA, respectively, with a spectral bandwidth of 0.7, 0.7, 0.2, 0.2, 0.7 and 0.7 nm, respectively. The selected wavelengths were 228.8, 283.3, 232.0, 279.5, 324.8 and 213.9 nm for Cd, Pb, Ni, Mn, Cu, Zn, respectively. Argon 99.996% (White Martins, São Paulo, SP, Brazil) was used as protective and purge gas. Pyrolytic graphite coated polycrystalline electrographite tubes with total pyrolytic graphite platforms (PerkinElmer) were used throughout. Then, 30 μL of diluted samples was directly deposited onto the L'vov platform. The used heating programs used are given in Table 1.

2.2.2. Inductively coupled plasma mass spectrometry method

We used a PE ELAN DRC II ICP-MS instrument (PerkinElmer Life and Analytical Sciences) for the determination of elements in nails. Typical daily instrumental parameters are given in Table 2. Although this instrument can be used in the DRC mode to remove polyatomic interferences, we operated it solely in standard mode, i.e., with the DRC valve vented, for the determination of metals.

Table 2
ICP-MS operating conditions

PerkinElmer Elan DRC II	
Spray chamber	Cyclonic
Nebulizer	Meinhard®
RF power (W)	1100
Ar nebulizer gas flow (L min^{-1})	0.7–0.9 (optimized daily)
Measures	
Scan mode	Peak hopping
Resolution (amu)	0.7
Replicate time (s)	1
Dwell time (s)	50
Sweeps/reading	20
Integration time (ms)	1000
Replicates	3
Isotopes	^{63}Cu, ^{111}Cd, ^{208}Pb, ^{60}Ni, ^{55}Mn, ^{64}Zn
Correction equations	
Cadmium = $^{114}Cd - (0.027250 \times {}^{118}Sn)$	
Lead = $(^{204}Pb - [0.230074 \times {}^{202}Hg]) + {}^{206}Pb + {}^{207}Pb + {}^{208}Pb$	