Simultaneous and Sensitive Determination of As and Sb in Coffee and Tea With Double-Channel HG-AFS

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INTRODUCTION

"Coffee or tea?" One may meet this question every day. It is hard to say which one is better for us, but undoubtedly coffee and tea are the two most popular beverages in the world. In fact, the drinking of coffee and tea has become part of everyday life because of its excellent taste and aroma. These beverages are also known to contain phytochemical properties, in addition to being a stimulant due to the presence of caffeine. The health benefits [e.g., prevention of cancers (1) and chronic liver diseases (2)] have been demonstrated in recent years. However, when consumption of coffee and tea becomes excessive and habitual, it can also represent a significant source of potentially hazardous chemicals. Therefore, the chemical composition of coffee and tea, namely the presence of essential, non-essential, and toxic elements, has to be known and kept under control in terms of its safety, quality, nutritional value, and certain sensorial properties. Some elements are toxic even at low concentrations. Both the producers and consumers are now paying more and more attention to coffee and tea safety and quality.

Generally, the selective analysis of most elements in coffee (3) and tea (4) is carried out by flame atomic absorption spectrometry (FAAS). FAAS is the most widespread

ABSTRACT

Coffee and tea are two of the most popular beverages in the world. Thus, it is of great importance to develop simple and efficient analytical methods to determine their chemical composition. In this study, a simple analytical procedure was developed for the sensitive and simultaneous determination of trace arsenic and antimony in coffee and tea samples by hydride generation atomic fluorescence spectrometry (HG-AFS). The conditions for the generation of volatile As and Sb species, such as pre-reduction conditions, carrier gas and shield gas flow rates, and reaction media (KBH_4 and HCl), were investigated in detail. The precision for five replicates at the 20 ng mL^{-1} As level and the 10 ng mL^{-1} Sb level was 1.3% and 2.1% (relative standard deviation, RSD), respectively.

Under the optimized experimental conditions, the limits of detection (LOD) were 0.072 ng mL^{-1} and 0.078 ng mL^{-1} , with linear ranges of 0–80 ng mL^{-1} ($R^2 = 0.9992$) and 0–40 ng mL^{-1} ($R^2 = 0.9965$), for As and Sb, respectively. The proposed method was successfully applied for the simultaneous determination of As and Sb in several coffee and tea samples from a local market, and the spike recoveries were in the 92–104% range. The accuracy of the method was validated by analyzing certified reference materials and the results were in good agreement with the certified values.

traditional atomic spectrometry technique for the determination of trace elements because of its robustness and relatively low cost, but it often suffers from low sensitivity for some important trace elements. Alternatively, atomic fluorescence spectrometry (AFS) has been widely used for the determination of chemical vapor generation (CVG) elements (i.e., As, Sb, Bi, Se, Te, Ge, Sn, Pb, Zn, Cd, and Hg) because of its high sensitivity, wide linear dynamic range, speed of analysis, ease of use, and low cost (5–7). The detection ability of AFS is superior to FAAS and comparable to graphite furnace atomic absorption spectrometry (GFAAS) and inductively coupled plasma mass spectrometry (ICP-MS) (8–10) with regard to sensitivity and linear calibration range, with further advantages (simplicity, low acquisition, and running costs) for chemical vapor-forming elements in routine analysis (4,5,11,12). Holak reported the pioneering work of chemical vapor generation (in this case, hydride generation) combined with atomic absorption spectrometry in 1969 (13). Tsujii et al. first applied hydride generation (HG) to atomic fluorescence spectrometry in 1974 (14). The first simultaneous multi-element determination of HG elements was performed in combination with inductively coupled plasma optical emission spectrometry (ICP-OES) in 1978 (15). Double-channel HG-AFS, whereby two elements could be detected simultaneously by HG-AFS, was first reported by Guo et al. in 1983 (16). However, HG-AFS is mainly applied for single-element measurement. A literature survey

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revealed that most of such work concentrated on single-element analysis, whereas, to the best of our knowledge, no HG-AFS procedure for the simultaneous determination of As and Sb in coffee and tea has been published previously (17-26).

In this work, the simultaneous determination of multi-elements (As and Sb) in coffee and tea samples is presented by HG-AFS. Arsenic and antimony are treated as potential chemical hazards by the World Health Organization. There is much epidemiological evidence that shows that arsenic in drinking water causes diseases such as skin cancer and several internal cancers, especially lung, bladder, and kidney cancers. Exposure to antimony may cause diarrhea, joint and/or muscle pain, vomiting, prob-

lems with the blood (anemia), and heart problems (altered electrocardiograms). Arsenic and antimony levels in coffee and tea are widely used as biological indicators for the estimation of their exposure (3, 4). The experimental conditions, including pre-reduction conditions, carrier gas and shield gas flow rates, reaction medium (KBH_4 and HCl) concentrations, analytical figures of merit, and method validation were investigated in this study and are discussed in detail.

EXPERIMENTAL

Instrumentation

A model AFS-2202 double-channel non-dispersive atomic fluorescence spectrometer (Beijing Haiguang Instrument Co., Ltd., Beijing, P.R. China) was used

throughout this work. The instrument is equipped with a programmable intermittent reactor and two gas-liquid separators to facilitate hydride generation and separation. High-intensity hollow cathode lamps (HCL, Research Institute of Non-Ferrous Metals, Beijing, P.R. China) of arsenic and antimony were used as the radiation sources. The working parameters of HG-AFS are summarized in Table I, and the setup of the HG-AFS is shown in Figure 1.

Reagents and Standard Solutions

All reagents were of analytical reagent grade. Deionized water (DIW) was used throughout this study. Standard stock solutions of arsenic and antimony (100 mg L^{-1}) were purchased from the National Center for Reference Material (Beijing, P.R. China). The mixed working standard solutions in the $\mu\text{g L}^{-1}$ and sub- $\mu\text{g L}^{-1}$ range were obtained by stepwise dilution from the stock solution with 5% (v/v) HCl solution. Solution of KBH_4 , 25 g L^{-1} , was stabilized with 5 g L^{-1} KOH . A mixed pre-reduction solution was composed of 5% (m/v) thiourea and 5% (m/v) ascorbic acid. High purity argon gas (99.99%) was used as both the carrier and shield gas.

Procedure

The raw coffee bean and tea leave samples were purchased from local markets. The coffee beans or tea leaves were put in a drying oven and desiccated at 80°C for one hour. After cooling, the dried samples were ground to a grain size of 200 mesh using the agate mortar. And then, approximately 1.0000 g of coffee or tea samples were accurately weighed into polytetrafluoroethylene (PTFE) vessels. A 10 mL amount of nitric acid and perchloric acid mixture (4:1) was added to the PTFE vessels. After overnight room temperature digestion, the sealed vessels were then placed in

TABLE I
AFS Instrumental Parameters

Parameters	As	Sb
Hollow cathode lamp	193.7 nm, 60 mA	217.6 nm, 60 mA
Negative voltage of PMT		280 V
Atomizer temperature		200°C
Observation height		8 mm
Read mode		Peak area
Read time		10 s
Carrier gas flow rate		800 mL min^{-1}
Shield gas flow rate		1100 mL min^{-1}

Fig. 1. Photo of the setup of HG-AFS. (PMT: photomultiplier tube).

an electric oven and heated at 125 °C for five hours. After cooling, the vessels were placed on an electric hot plate to heat and brought to a final volume of 2 mL, then 4 mL 50% HCl was added. The vessels were heated until the volume of the solutions was reduced to around 0.5 mL. The resultant solutions were transferred into a 25-mL volumetric flask and finally diluted to volume with deionized water. The solutions for the determinations were prepared by diluting 5 mL of sample solutions to a final volume of 25 mL with hydrochloric acid solution (final concentration of 5% v/v) and pre-reductive solution (final concentration of 5%, m/v, thiourea and 5%, m/v, ascorbic acid). To validate the accuracy of the proposed method, two certified reference materials (CRMs) were purchased from the National Center for Reference Material (Beijing, P.R. China): GBW07605 (tea) and GBW10016 (tea). Hydride generation was carried out by mixing the sample solutions with 2.5% (m/v) KBH_4 solution using an intermittent flow system.

RESULTS AND DISCUSSION

Pre-reduction Conditions

As and Sb can both exist in two different oxidation states: trivalent and pentavalent. It is well known that As(V) and Sb(V) have much slower reaction kinetics for hydride formation. This is dependent on the hydride generation system used and the concentration of reagents. With a continuous flow manifold, the hydride generation efficiencies are around 40% compared to their trivalent oxidation state. The most common pre-reduction procedure involves the addition of thiourea and ascorbic acid reagent. Preliminary results indicate that a mixture of thiourea and ascorbic acid provides an effective reduction to the trivalent oxidation states after a pre-reduction time of 30 minutes. In addition, thiourea and ascorbic acid as masking reagents could reduce the interference of coexisting elements.

Carrier Gas and Shield Gas Flow Rate

Pure argon was used as both the carrier gas and the shield gas. Argon transports hydrides from the liquid-gas separator to the atomizer, acts as analyte diluent, and stabi-

lizes the flame by forming an appropriate mixture with H_2 . Various flow rates, which influence the sensitivity and stability of the instrument, were tested in this study. The effects of carrier and shield gas on the signal of As and Sb are shown in Figures 2 and 3. The carrier gas at lower flow rate could not quickly sweep the hydride of the analyte into the inner tube of a quartz furnace, and at higher flow rates it would dilute the analyte introduced in the furnace tube. The amounts of vapor generated were different for different analytes. Different flow rates of extraneous gas were required for completely carrying the different amounts of hydrides into the atomizer, which led to a difference in the optimal flow rates for As and Sb. The shield gas was used to prevent extraneous air from entering the flame. A carrier gas flow rate of 800 mL min^{-1} resulted a maximum signal intensity for both As and Sb. On the other hand, a shield gas flow rate of 1100 mL min^{-1} for both As and Sb gave the highest signal intensity. Consequently, a carrier gas flow rate of 800 mL min^{-1} and a shield gas flow rate of 1100 mL min^{-1} were employed for the simultaneous determination of As and Sb.

Fig. 2. Effect of carrier gas flow rate on the fluorescence signal of As and Sb.

Fig. 3. Effect of shield gas flow rate on the fluorescence signal of As and Sb.

Reaction Medium (KBH₄ and HCl)

In the HG-AFS system employed, KBH₄ is used not only as the reductant, but also as the hydrogen supply to sustain the argon-hydrogen flame. The concentration of KBH₄ had a large impact on the hydride generation process and the stability of the argon-hydrogen flame. The optimization of KBH₄ concentration was carried out between 1% and 3.5% (m/v). Figure 4 illustrates the effect of the KBH₄ concentration on the signal intensities of As and Sb. Generation of both AsH₃ and SbH₃ vapor could be carried out

with KBH₄ concentrations ranging from 1% to 3.5%, but the sensitivities decreased gradually when the KBH₄ concentrations were below 2.5%. The fluorescence intensity decreased a little when the KBH₄ concentration was over 3%. It is also well recognized that some additional drawbacks such as causing very noisy background, taking more water vapor to the atomizer, and achieving a higher standard deviation, hamper the performance of the HG-AFS system when the concentrations of KBH₄ are too high. Thus, 2.5% KBH₄ was chosen for further experiments for As and Sb hydride generation.

The generation efficiency of the volatile species greatly depends upon the acidity at which the reaction is performed. Using 2.5% KBH₄ as the reductant with the flow rate of the sample being approximately equal to that of the potassium tetrahydroborate flow rate, the effect of various HCl concentrations on the signal intensities was investigated within the range of 2–20%, and the results in Figure 5 show that an HCl concentration of 5–10% yielded a response plateau wherein the best results were obtained for As and Sb. However, the reagent background was increased with an increase in HCl concentration. Therefore, a concentration of 5% HCl, which was near the lower edge of the optimization plateau, was chosen as the final working value.

Analytical Figures of Merit

Under the optimal conditions, a series of mixed standard solutions of As and Sb were prepared and detected by two-channel HG-AFS. The fluorescence intensity of the analyte was found to be linearly proportional to the concentration of the analyte. Table II shows the analytical figures of merit of the proposed simultaneous determination method for As and Sb. The

Fig. 4. Effect of KBH₄ concentration on the fluorescence signal of As and Sb.

Fig. 5. Effect of HCl concentration on the fluorescence signal of As and Sb.

Analytical Figures	Sb	As
Dynamic Range	0–40 ng mL ⁻¹	0–80 ng mL ⁻¹
Equation of Linear Fit	$y = 35.17x + 135.3$	$y = 43.91x + 125.3$
Linear Correlation Co-efficient (R ²)	0.9965	0.9992
Relative Standard Deviation /% n=5	2.1 (10 ng mL ⁻¹)	1.3 (20 ng mL ⁻¹)
Limit of Detection	0.078 ng mL ⁻¹	0.072 ng mL ⁻¹

Fig. 6 (a and b). The calibration curves for As and Sb.

calibration curves are shown in Figure 6 (a and b) and exhibited good linearity. The limits of detection were 0.072 ng mL⁻¹ and 0.078 ng mL⁻¹ for As and Sb, respectively, suitable for the sensitive analysis of biological samples.

Validation

In order to validate the applicability of the proposed method for trace arsenic and antimony determination, a series of coffee and tea samples collected from local markets were analyzed. As shown in Table III, the recoveries of spiking 5–60 ng mL⁻¹ of arsenic and antimony were in the range of 92–104%.

In order to further verify the accuracy and precision of the method, two certified reference materials of GBW07605 (tea) and GBW10016 (tea) were analyzed for each element. The samples were prepared in triplicate and analyzed in triplicate. The results shown in Table IV demonstrate excellent recoveries in comparison to the certified values.

CONCLUSION

This work demonstrates the feasibility for the simultaneous determination of trace arsenic and antimony in coffee and tea samples by HG-double-channel-AFS. The

proposed method is simple and sensitive and is promising for the routine analysis of trace As and Sb in botanic samples and other biological materials.

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TABLE III
Determination of As and Sb in Coffee and Tea Samples

Samples	Added (ng mL ⁻¹)		Found (ng mL ⁻¹) ^a		Recovery (%)	
	Sb	As	Sb	As	Sb	As
Coffee 1	0	0	2.345 (0.293 µg g ⁻¹)	1.241 (0.155 µg g ⁻¹)	-	-
	20	20	20.821	20.353	92%	96%
	60	60	57.470	59.165	92%	97%
Coffee 2	0	0	^b	2.130 (0.266 µg g ⁻¹)	-	-
	5	5	5.222	7.124	104	100
	10	10	9.642	11.745	96	96
Tea 1	0	0	2.457 (0.307 µg g ⁻¹)	1.299 (0.162 µg g ⁻¹)	-	-
	20	20	20.974	20.640	93%	97%
	60	60	57.490	59.956	93%	98%
Tea 2	0	0	0.590 (0.074 µg g ⁻¹)	1.089 (0.136 µg g ⁻¹)	-	-
	5	5	5.677	6.003	102	98
	10	10	10.773	11.242	102	102

^a Mean of three determinations.

^b Not detectable.

TABLE IV
Results of the Analysis of Certified Reference Materials (n = 3 for each measurement)

CRM No.	Certified (µg g ⁻¹)		Found (µg g ⁻¹)	
	As	Sb	As	Sb
GBW07605 Tea	0.28 ± 0.04	0.056 ± 0.006	0.26 ± 0.02	0.052 ± 0.004
GBW 10016 Tea	0.09 ± 0.01	0.022 ± 0.006	0.092 ± 0.005	0.021 ± 0.003

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