Determination of Trace Amounts of Boric Acid in Cosmetics

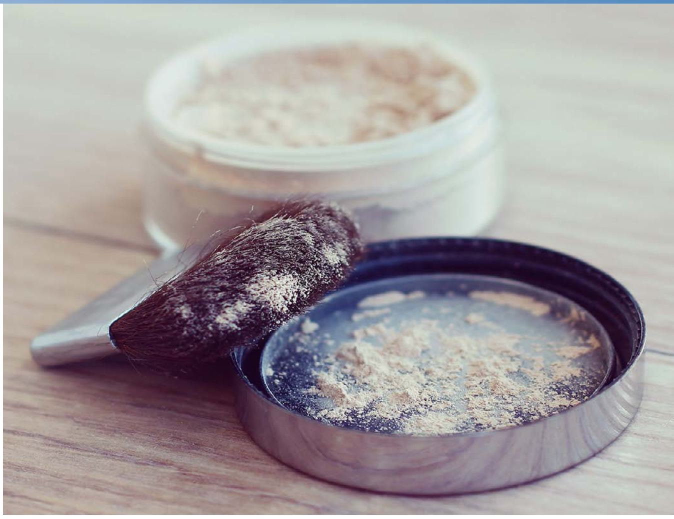
Zheng Hongguo,[1] Li Renyong,[1] Xu Qun,[1] Jeffrey Rohrer[2]
[1]Thermo Fisher Scientific, Shanghai, People's Republic of China
[2]Thermo Fisher Scientific, Sunnyvale, CA, USA

Key Words

Dionex IonPac ICE-Borate Column, Borate, Conductivity Detection, Ion Chromatography (IC), Personal Care Product Analysis

Goal

To develop an efficient IC method for the sensitive determination of boric acid in cosmetics



Introduction

Boric acid and borates (e.g., metaborate, borate, and tetraborate) are added to some cosmetics as a preservative and antibacterial agent. But once inhaled or absorbed through skin cuts, boric acid and borates can cause intoxication.[1] Therefore, the amount of boric acid and borates in different kinds of cosmetics have been strictly regulated. For example, the European Union (EU) and China regulations permit the same amount of boric acid and borates to be added in cosmetics, and use the detected amount of boric acid to state the content as a weight percent of the cosmetic based on the conversion of borates to boric acid during sample preparation (e.g., a solution of perchloric acid and hydrogen peroxide is used to convert borates to boric acid in the China restriction).[2,3] Table 1 lists the requirements of the restrictions in the EU and China. The cosmetics for children under three years of age must not contain these substances, demonstrating that sensitive detection methods are necessary. However, the sensitivity of the spectrophotometry method recommended by the Hygienic Standard for Cosmetics (China)[3], which uses azomethine-H as a colorimetric reagent and has a method detection limit (MDL) of >10 mg/L, cannot meet the strict requirement. Therefore, an IC method that has been applied to the sensitive determination of borate (boric acid) in water samples[4-7] is applied to the determination of borate in cosmetics.

Table 1. Regulations concerning boric acids in cosmetics in the EU and China.

Regulations	Substance	Restrictions		
		Cosmetic Product	Maximum Authorized Concentration	Other Limitations and Requirements
Commission Directive 76/768/EEC (EU), and Hygienic Standard for Cosmetics (China)	Boric acid, borates, and tetraborates	Talcum powder	5% (equivalent to 50 g/Kg, by mass/mass as boric acid)	Not to be used in products for children under 3 years of age
		Products for hygiene	0.1% (equivalent to 1 g/Kg, by mass/mass as boric acid)	
		Other products (excluding bath products and hair waving products)	3% (equivalent to 30 g/Kg, by mass/mass as boric acid)	

Equipment

- Thermo Scientific™ Dionex™ ICS-2100 Integrated Reagent-Free™ Ion Chromatography (RFIC™) system, including:
  - – Isocratic Pump
  - – Vacuum Degasser
  - – Conductivity Cell and Detector
- Thermo Scientific™ Dionex™ AMMS™ ICE 300 Anion Ion-Exchange MicroMembrane Suppressor (P/N: 067527)
- Thermo Scientific Dionex AS-AP Autosampler with 100 µL sample loop (P/N 030391) and 5 mL sample syringe (P/N 074308)
- Thermo Scientific™ Dionex™ Chromeleon™ Chromatography Data System (CDS) software, version 7.1
- Thermo Scientific™ Sorvall™ ST16 Centrifuge (P/N 75004240)
- T 18 Digital ULTRA-TURRAX® Disperser (IKA P/N 3720000)

Consumables

- Thermo Scientific™ Target2™ Nylon Syringe Filter, 0.45 µm, 30 mm (P/N F2500-1)
- Thermo Scientific Sun-SRi Luer-Lock Syringe (Fisher Scientific P/N 14-823-261)
- Thermo Scientific™ Dionex™ OnGuard™ II RP Cartridge (P/N 057084)

Reagents and Standards

- Deionized (DI) water, 18.2 MΩ-cm resistivity (generated from a Thermo Scientific™ GenPure Pro UV-TOC™, P/N 50131948)
- Acetonitrile (CH_3CN), HPLC Grade (Fisher Scientific P/N AC610010040)
- Methanesulfonic acid (MSA), HPLC Grade (Acros Organics P/N AC43297-0010)
- D-Mannitol, 98% (Acros Organics P/N AC12534-1000)
- Tetramethylammonium hydroxide pentahydrate (Reagent), 98% (Fisher Scientific P/N O4645-25)
- Boric acid, 99.5%, crystalline, certified ACS (Fisher Scientific P/N A73500)

Preparation of Standard Solutions

Stock Standard 1

Dissolve 0.01 g of boric acid in 10 mL of DI water. The concentration of Stock Standard 1 is 1000 mg/L.

Stock Standard 2
(also used as a standard solution for calibration)

Dilute 200 µL of Stock Standard 1 to 10 mL with DI water. The concentration of Stock Standard 2 is 20 mg/L.

Standard Solutions for Calibration

For calibration, prepare eight working standard solutions with different concentrations by diluting the proper amount of the Stock Standards with DI water. The volumes of each solution needed to make the calibration standards are shown in Table 2.

Sample Preparation

Ten cosmetic samples (two talcum powders, three creams, and five liquids) were purchased in a supermarket in Shanghai, China.

Cartridge Activation

Use the Dionex OnGuard II RP cartridge for sample pretreatment to remove hydrophobic organic compounds. The cartridge requires activation before use. Pass 10 mL of methanol through the cartridge followed by 15 mL of DI water, and then allow it to stand for 15 min before use.

Cream Sample

Add 5 mL of acetonitrile to 2.0 g of cream sample in a 10 mL centrifuge tube. Mix for 5 min to extract, centrifuge the extract for 10 min at 6000 rpm, remove the supernatant, then add 5 mL of acetonitrile to the residue and extract a second time in the same manner. Combine the two supernatants (total volume ~10 mL) in a 100 mL volumetric flask, and bring to the volume with DI water. After filtering the sample solution through a 0.45 µm syringe filter, pass the filtrate through a Dionex OnGuard II RP cartridge. Discard the first 3 mL of solution and collect the remaining solution. In practice, dilution will be performed with DI water if necessary.

Table 2. Preparation of calibration standards.

Stock Standard for Boric Acid Calibration	Volume of Stock Standard for Boric Acid Calibration (mL)	Volume of DI Water (mL)	Final Volume of Calibration Standard (mL)	Final Concentration of Calibration Standard (µg/L)
Stock Standard 1 (1000 mg/L)	0.20	9.80	10.0	20000
Stock Standard 2 (20 mg/L)	2.50	7.50		5000
	1.00	9.00		2000
	0.50	9.50		1000
	0.25	9.75		500
	0.10	9.90		200
	0.05	9.95		100
	0.04	9.96		80
	0.025	9.975		50

Add 5 mL of acetonitrile and 1500 µL of 20 µg/mL of boric acid standard solution (Stock Standard 2) to 2.0 g of the cream sample in a 10 mL centrifuge tube. Sample preparation is completed using the procedure above. The spiked concentration of boric acid in the cream sample will be 15 mg/Kg (equivalent to 1.2 mg/L in the well-prepared samples).

Add 5 mL of acetonitrile and 300 µL of 20 µg/mL of boric acid standard solution (Stock Standard 2) to 2.0 g of cream sample in a 10 mL centrifuge tube. Sample preparation is completed using the procedure above. The spiked concentration of boric acid in the cream sample will be 3 mg/Kg (equivalent to 0.24 mg/L in the well-prepared samples). This spiked sample is used for the determination of cosmetic products for children under 3 years of age.

Powder Sample

Add 5 mL of DI water to 1.0 g of powder sample (e.g., talcum powder) in a 10 mL centrifuge tube. Mix for 10 min to extract, then centrifuge for 10 min at 6000 rpm. Transfer the supernatant to a 10 mL volumetric flask and bring to the volume with DI water. After filtering the sample solution through a 0.45 µm syringe filter, pass the filtrate through a Dionex OnGuard II RP cartridge. Discard the first 3 mL of solution and collect the remaining solution. Some samples will require dilution with DI water prior to analysis.

Add 5 mL of DI water and 600 µL of 20 µg/mL of boric acid standard solution (Stock Standard 2) to 1 g of talcum powder sample in a 10 mL centrifuge tube. Sample preparation is completed using the procedure above. The spiked concentration of boric acid in the powder sample will be 12 mg/Kg (equivalent to 1.2 mg/L in the well-prepared samples).

Add 5 mL of DI water and 150 µL of 20 µg/mL of boric acid standard solution (Stock Standard 2) to 1 g of talcum powder sample in a 10 mL centrifuge tube. Sample preparation is completed using the procedure above. The spiked concentration of boric acid in the powder sample will be 3 mg/Kg (equivalent to 0.3 mg/L in the well-prepared samples). This spiked sample is used for the determination of cosmetic products for children under 3 years of age.

Liquid Sample

Dilute 1.0 g of a liquid sample (e.g., shampoo, body wash, and body lotion) to the volume with DI water to in a 10 mL volumetric flask. After filtering the sample solution through a 0.45 µm syringe filter, pass the filtrate through a Dionex OnGuard II RP cartridge. Discard the first 3 mL of solution and collect the remaining solution. Some samples will require dilution with DI water prior to analysis.

Dilute 1.0 g of a liquid sample (e.g., shampoo and body wash) and 600 µL of 20 µg/mL of boric acid standard solution (Stock Standard 2) to the volume with DI water in a 10 mL volumetric flask. Sample preparation is completed using the procedure above. The spiked concentration of boric acid in the powder sample will be 12 mg/Kg (equivalent to 1.2 mg/L in the well-prepared samples).

Dilute 1.0 g of a liquid sample (e.g., shampoo and body wash) and 150 µL of 20 µg/mL of boric acid standard solution (Stock Standard 2) to the volume with DI water in a 10 mL volumetric flask. Sample preparation is completed using the procedure above. The spiked concentration of boric acid in the powder sample will be 3 mg/Kg (equivalent to 0.3 mg/L in the well-prepared samples). This spiked sample is used for the determination of cosmetic products for children under 3 years of age.

Conditions	
Columns:	Thermo Scientific™ Dionex™ IonPac™ ICE-Borate, 7.5 µm, 250 × 9 mm (P/N 053945)
Eluent:	3 mM MSA/60 mM mannitol (dissolve 0.2 mL of MSA and 11 g of mannitol in 200 mL of DI water, and bring the total volume to 1000 mL with DI water)
Flow Rate:	1.0 mL/min
Injection Volume:	100 µL
Column Temperature:	30 °C
Detection:	Suppressed conductivity, with Dionex Anion-ICE Suppressor (P/N: 067527), connected to external regenerant (15 mM mannitol and 25 mM Tetramethylammonium hydroxide. Dissolve 2.8 g of mannitol and 4.6 g of tetramethylammonium hydroxide pentahydrate in 100 mL of DI water, and bring the total volume to 1000 mL with DI water) at 3 mL/min flow rate for suppression
System Backpressure:	900 psi
Background Conductance:	0.3 µS

Results and Discussion

Cosmetics commonly can be classified as creams, liquids, or powders. For liquid and powder products, such as shampoo and talcum powder, DI water can be used to extract borate from these samples, and a reversed-phase cartridge (Dionex OnGuard II RP cartridge) can then be used to remove the hydrophobic sample components. For water-insoluble cream products, extraction with DI water and agitation can easily cause emulsion formation, resulting in insufficient analyte extraction.[1] Using an organic solvent (e.g., acetonitrile) not only prevents and disrupts the emulsion efficiently, but also effectively precipitates water-insoluble sample components. Therefore, acetonitrile was used to treat the cream cosmetic samples.

In an acidic environment (~ pH 2.6), borates will be present as boric acid (H_3BO_3), which is the reason that restrictions in the EU and China state using "by mass/mass as boric acid".[2,3] For example, tetraborate ($Na_2B_4O_7$) reacts with MSA (CH_3SO_3H) instantaneously due to the strong acidity of MSA, and boric acid is generated. Moreover, boric acid and mannitol ($C_6O_6H_{14}$) can form a stable monovalent anionic complex with pKa ≈ 4.5 in the acidic environment,[10] allowing it to be more easily detected by conductivity detection (reactions shown in Figure 1). Therefore, MSA eluent was used to separate boric acid, and mannitol was added to the eluent to get higher detection sensitivity by ion-exclusion chromatography (IEC).

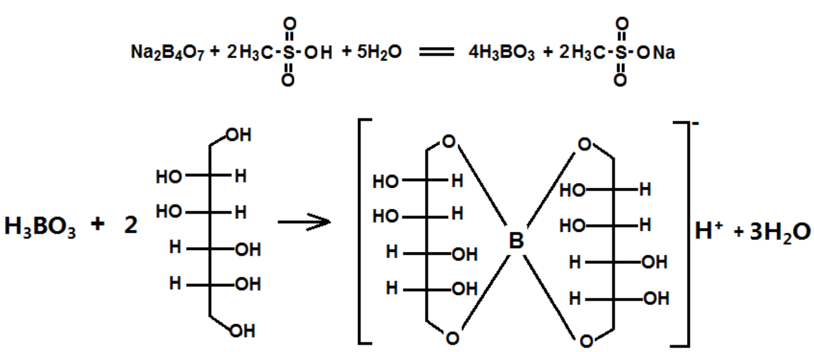


Figure 1. Reactions of tetraborate, boric acid, and mannitol in an acidic environment (~ pH 2.6, 3 mM MSA).

Experiments show that retention of boric acid increases with increasing MSA concentration. As the MSA concentration exceeds 3 mM, the retention of boric acid only changes slightly, whereas conductivity background increases significantly. Therefore, a 3 mM of MSA eluent was used. However, because glycerol and some organic acids such as malic acid and citric acid that are commonly added to cosmetics may interfere with the determination of borate using typical ion-exclusion columns;[8] the Dionex IonPac ICE-Borate, an ion-exclusive column with high selectivity for borate,[9] was used for this determination.

Method precision was estimated by making nine consecutive 100 µL injections of a calibration standard with concentration of 1.0 mg/L of boric acid. Retention time reproducibility (RSD) was ≤0.1 and peak area reproducibility (RSD) was ≤0.8, demonstrating good short-term precision for this method. Calibration linearity of the analyte was investigated by making five consecutive 100 µL injections of the borate Standard prepared at eight different concentrations (i.e., 40 total injections). A linear relationship was observed when plotting concentration versus peak area in the range of 80–5000 µg/L (Figure 2, the 5000 µg/L point is not shown). The linear regression equation was $A = 0.022c - 0.002$, where, A represents peak area, and *c* represents concentration of analyte; and the coefficient of determination was 0.999. This calibration curve was used to quantify boric acid in cosmetic samples. Seven replicate injections of a boric acid standard with concentration 50 µg/L were used for estimating the method detection limit (MDL) using a signal-to-noise ratio (S/N) = 3. The measured MDL of boric acid was 15 µg/L. Figure 3 shows a chromatogram of a boric acid standard solution with concentration of 50 µg/L.

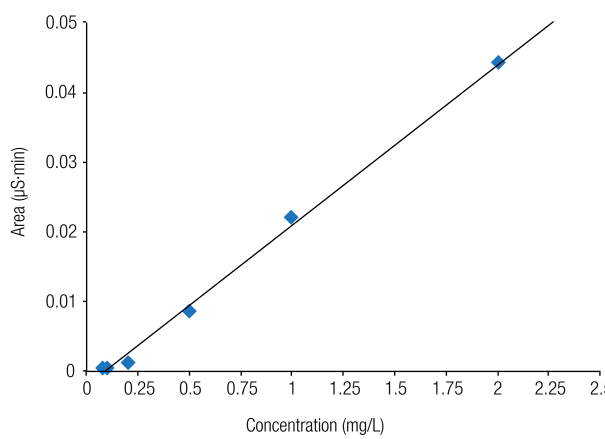


Figure 2. Calibration curve of boric acid.

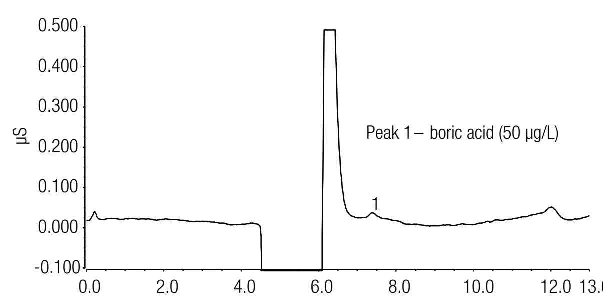


Figure 3. Chromatogram of a boric acid standard solution.

Sample Analysis

Figure 4 shows the chromatograms of a cosmetic cream sample, and Table 3 shows the analysis results for ten cosmetic samples. Boric acid was not detected in samples 2#, 5#, and 7# that are for children under 3 years of age; and was detected in the other seven samples with amounts all less than allowed in the EU and China[2,3] restrictions, indicating these cosmetics are safe with respect to added boric acid. Method accuracy was investigated by determining the recoveries in the spiked cosmetic samples. As summarized in Table 3, the recoveries ranged was from 79 to 108%, demonstrating that this IC method provides good selectivity and sensitivity, and is suitable for the determination of boric acid in cosmetic products.

Conclusion

This work describes an IC method with suppressed conductivity detection for sensitive determination of boric acid in cosmetics. The determination is performed on a Dionex ICS-2100 Integrated RFIC system controlled by Chromeleon CDS software and equipped with a Dionex IonPac ICE-Borate column. Compared to the spectrophotometry method, this IC method provides good selectivity, and a rapid and accurate determination of boric acid in cosmetic products.

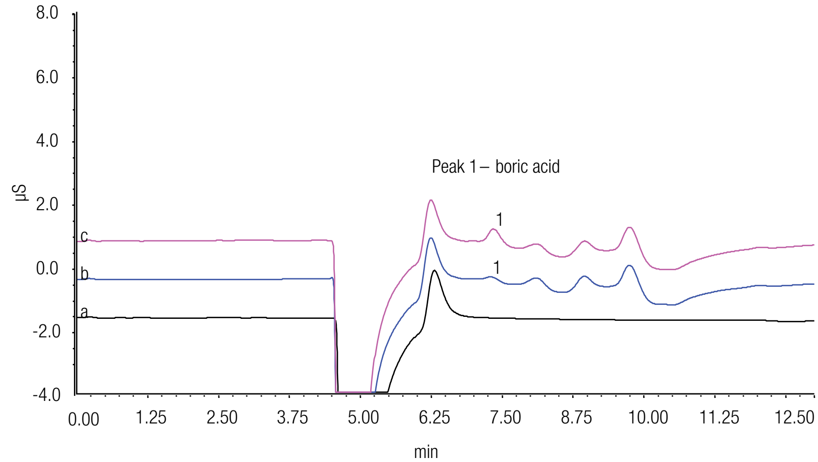


Figure 4. Chromatograms of (a) blank, (b) a cream cosmetic sample, and (c) the same sample spiked with 15 mg/Kg (equivalent to 1.2 mg/L in the well-prepared sample solution) of boric acid standard.

Table 3. Sample analysis results.

Type	Serial No.	Detected (mg/Kg)	Spiked Sample		
			Added (mg/L)	Found (mg/L)	Recovery
Cream	1#	33.2	1.2	1.1	92%
	2#*	ND**	0.24	0.19	79%
Talcum (Powder)	3#	14.1	–	–	–
	4#	60.2	1.2	1.3	108%
	5#*	ND**	0.3	0.26	87%
Liquid	6#	7.70	1.2	1.1	83%
	7#*	ND**	0.3	0.32	107%
	8#	27.0	–	–	–
	9#	40.3	–	–	–
	10#	18.5	–	–	–

** For children under 3 years of age*
*** Not detected*

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