

Thallium in Hair by ICP/MS

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Thallium in Hair by ICP/MS

1 INTRODUCTION

Thallium is a toxic metal that may cause gastrointestinal, neurological or dermatological symptoms. Thallium can be detected in hair following chronic exposure or following survival of an acute exposure. This procedure is used to detect and quantitate thallium (Tl) in hair specimens.

2 SCOPE

Analyses	<input checked="" type="checkbox"/> Screening <input checked="" type="checkbox"/> Confirmation <input checked="" type="checkbox"/> Quantitation
Matrices	Hair
Analytes	Thallium
Personnel	This document applies to authorized personnel who perform the described tasks, singly or in combination.

3 PRINCIPLE

Hair specimens are weighed, completely digested in tetramethylammonium hydroxide (TMAH), mixed with the Iridium Internal Standard Working Solution, diluted up to 10 mL in Dilute Nitric Acid in Water Solution and analyzed contemporaneously with a complete matrix matched calibration curve, a Negative Control Hair sample and duplicate sets of Positive Control Hair samples.

4 SPECIMEN CRITERIA

This procedure is validated for hair. Two 5.5 mg samples are analyzed. However, if it is suspected that the thallium concentration is above the procedure's linear range, smaller sample sizes or further dilutions of the samples may be analyzed.

5 EQUIPMENT

5.1 Equipment

10 mL and 25 mL volumetric flasks, class A, PMP (VITLAB® or equivalent)

5 mL 12 x 75 mm round bottom tubes with caps, PP (Falcon® or equivalent)

100 mL volumetric flask, class B, PP (Nalgene® or equivalent)

1 L volumetric flask, class B, PP (Nalgene® or equivalent)

25 mL plastic graduated cylinder (Nalgene® or equivalent)

Balance capable of measuring ± 0.1 mg

Routine laboratory supplies including but not limited to: pipettes, disposable pipettes, forceps, hand shears, etc.

Cryogrinder (optional)

Vortexer

Heating block

5.2 Consumables

15 mL and 50 mL conical tubes with screw tops, PP (Falcon® or equivalent)

5.3 Instruments

- A. Inductively Coupled Plasma-Mass Spectrometer with a collision cell installed (Thermo-Fisher iCAP Q or equivalent)
- B. Autosampler (CETAC ASX-260 or equivalent)

5.4 Software

Component	Software	Version
Operating System	Microsoft Windows	7 Pro SP 1
Mass Spectrometer	Chromeleon	7.2 SR5
	Qtegra ISDS	2.10 SR1

5.5 Chemicals/Reagents

Stability and storage provided by supplier unless otherwise noted.

5.5.1 Purchased

Concentrated nitric acid (Optima grade)

Tetramethylammonium hydroxide (TMAH), 25% w/w aqueous solution (Electronic grade)

Acetone (Reagent grade or better)

Methanol (HPLC grade or better)

Water (Deionized, 18 MΩ)

5.5.2 Prepared

Dilute Nitric Acid in Water Solution (v:v)

To a 1 L Nalgene® volumetric flask, add approximately 800 mL of deionized water. Add 40 mL of Optima grade concentrated nitric acid, fill to the mark with deionized water and mix well. Store at room temperature in plastic. Stable for at least one year.

5.6 Standards/Controls

Stability and storage provided by supplier unless otherwise noted.

5.6.1 Purchased

Iridium Internal Standard Stock Solution (1 µg/mL)

Purchased from SPEX CertiPrep or an equivalent supplier.

Thallium Calibrator Stock Solution (1000 mg/L in Dilute Nitric Acid in Water Solution)

Purchased from SPEX CertiPrep or an equivalent supplier.

Thallium Control Stock Solution (10,000 mg/L in Dilute Nitric acid solution)

Purchased from High Purity Standards or an equivalent supplier.

5.6.2 Prepared

5.6.2.1 *Internal Standard*

Iridium Internal Standard Working Solution (10 µg/L)

To a 100 mL Nalgene® volumetric flask that has been rinsed with Dilute Nitric Acid in Water Solution, add about 80 mL of Dilute Nitric Acid in Water Solution. Add 1 mL of Iridium Internal Standard Stock Solution, fill to the mark with Dilute Nitric Acid in Water Solution and mix well. Store at room temperature in plastic. Stable for at least one year.

5.6.2.2 *Calibration*

Thallium Calibrator Working Stock Solution - High (5 mg/L in Dilute Nitric Acid in Water Solution)

To a 25 mL class A volumetric flask that has been rinsed with Dilute Nitric Acid in Water Solution, add about 15 mL of Dilute Nitric Acid in Water Solution. Add 0.125 mL of the Thallium Calibrator Stock Solution (1000 mg/L), fill to the mark with Dilute Nitric Acid in Water Solution and mix well. Store at room temperature in plastic. Stable for at least one year.

Thallium Calibrator Working Stock Solution - Low (0.5 mg/L in Dilute Nitric Acid in Water Solution)

To a 10 mL class A volumetric flask that has been rinsed with Dilute Nitric Acid in Water Solution, add about 7 mL of Dilute Nitric Acid in Water Solution. Add 1.0 mL of the Thallium Calibrator Working Stock Solution High (5 mg/L), fill to the mark with Dilute Nitric Acid in Water Solution

5.6.2.3 *Calibration Scheme*

Thallium Calibrator Working Solutions (1 µg/L – 250 µg/L)

The following table shows the preparation of the individual Calibrator Working Solutions. The Calibrator Working Solutions are prepared in individual 25 mL class A volumetric flasks that have been washed with Dilute Nitric Acid in Water Solution. Store at room temperature in plastic. Fill to the mark with Dilute Nitric Acid in Water Solution and mix well. Stable for at least one year.

Calibrator Working Solutions ($\mu\text{g/L}$) (Prepared in 25-mL volumetric flasks)	Volume of Thallium Calibrator Working Stock Solution - High (5 mg/L) (μL)	Volume of Thallium Calibrator Working Stock Solution - Low (0.5 mg/L) (μL)	Corresponds to xx ng Thallium per mg of hair as prepared
1		50	0.02
2.5		125	0.05
5		250	0.10
10	50		0.20
25	125		0.50
50	250		1.00
75	375		1.50
100	500		2.00
175	875		3.50
250	1250		5.00

5.6.2.4 Controls

Thallium Intermediate Control Solution High (10 mg/L in Dilute Nitric Acid in Water Solution)

To a 25 mL class A volumetric flask that has been rinsed with Dilute Nitric Acid in Water Solution, add about 15 mL of Dilute Nitric Acid in Water Solution. Add 0.025 mL of the Thallium Control Stock Solution (10,000 mg/L), fill to the mark with Dilute Nitric Acid in Water Solution and mix well. Store at room temperature in plastic. Stable for at least one year.

Thallium Intermediate Control Solution Low (0.5 mg/L in Dilute Nitric Acid in Water Solution)

To a 10 mL class A volumetric flask that has been rinsed with Dilute Nitric Acid in Water Solution, add about 15 mL of Dilute Nitric Acid in Water Solution. Add 0.5 mL of the Thallium Intermediate Control Solution High (10 mg/L), fill to the mark with Dilute Nitric Acid in Water Solution and mix well. Store at room temperature in plastic. Stable for at least one year.

Thallium High Control Working Solution (200 $\mu\text{g/L}$ in Dilute Nitric Acid in Water Solution)

To a 25 mL class A volumetric flask that has been rinsed with Dilute Nitric Acid in Water Solution, add about 15 mL of Dilute Nitric Acid in Water Solution. Add 0.5 mL of the Thallium Intermediate Control Solution High (10 mg/L), fill to the mark with Dilute Nitric Acid in Water Solution and mix well. Store at room temperature in plastic. Stable for at least one year.

Thallium Low Control Working Solution (7.5 $\mu\text{g/L}$ in Dilute Nitric Acid in Water Solution)

To a 25 mL class A volumetric flask that has been rinsed with Dilute Nitric Acid in Water Solution, add about 15 mL of Dilute Nitric Acid in Water Solution. Add 0.375 mL of the Thallium Intermediate Control Solution Low (0.5 mg/L), fill to the mark with Dilute Nitric Acid

in Water Solution and mix well. Store at room temperature in plastic. Stable for at least one year.




Negative Control Hair

Prepared from in-house anonymous donations. Collected negative specimens are screened for thallium. If thallium is present, the level must be below 0.02 ng/mg (1 µg/L). Store at room temperature in paper. Stable indefinitely.

6 PROCEDURE

Prepare Negative Control Hair Digests		Note/Reference
	A. To individually labeled 15 mL Falcon® tubes:	
<input type="checkbox"/>	1. Add a minimum of 100 mg of negative hair.	
<input type="checkbox"/>	2. Accurately record the mass to the nearest 0.1 mg. <i>Alternatively, a larger hair sample may be cryoground to mix it well. (The negative control hair digest will be prepared in the same manner as the unknown sample.)</i>	
<input type="checkbox"/>	3. Based upon the recorded mass, add enough TMAH to establish a solution of 5 mg of hair per 100 µL of TMAH. (For example, 2000 µL TMAH is needed for 100 mg of hair.)	
<input type="checkbox"/>	4. Allow the hair to completely digest, vortexing occasionally. (This process typically takes at least 8 hours, and the process may be left to proceed overnight.)	
	Calibration	
	B. To individual 10 mL Vitlab® volumetric flasks that have been washed with 2% Nitric Acid in Water Solution add:	
<input type="checkbox"/>	1. Approximately 5 mL 2% Nitric Acid in Water Solution	
<input type="checkbox"/>	2. 100 µL Negative Hair Digest (step A)	
	3. 100 µL of the appropriate Calibrator Solution (µg/L)	
<input type="checkbox"/>	i. Cal 0 (deionized water)	
<input type="checkbox"/>	ii. Negative Control (deionized water) <i>(One Negative Control needed before each Positive Control, as well as one Negative Control prior to each unknown replicate)</i>	
<input type="checkbox"/>	iii. Cal 1	
<input type="checkbox"/>	iv. Cal 2.5	
<input type="checkbox"/>	v. Cal 5	
<input type="checkbox"/>	vi. Cal 10	
<input type="checkbox"/>	vii. Cal 25	
<input type="checkbox"/>	viii. Cal 50	
<input type="checkbox"/>	ix. Cal 75	
<input type="checkbox"/>	x. Cal 100	
<input type="checkbox"/>	xi. Cal 175	

<input type="checkbox"/>	xii. Cal 250		
<input type="checkbox"/>	4. 100 µL Iridium Internal Standard Working Solution		
<input type="checkbox"/>	5. QS with 2% Nitric Acid in Water Solution mix well		
<input type="checkbox"/>	C. Transfer calibrators to labeled 15 mL Falcon® tubes		
<input type="checkbox"/>	D. Additionally, prepare a Blank (no matrix added) with Internal Standard by adding 100 µL Iridium Internal Standard Working Solution to 10 mL with Dilute Nitric Acid in Water Solution. (See step 3-ii also)		
	Control (prepared in duplicate for quantitative analysis)		
	E. To individual 10 mL Vitlab® volumetric flasks that have been washed with 2% Nitric Acid in Water Solution add:		
<input type="checkbox"/>	1. Approximately 5 mL 2% Nitric Acid in Water Solution		
<input type="checkbox"/>	2. 100 µL Negative hair digest (step A)		
	3. 100 µL of the appropriate Control Solution (µg/L);		
<input type="checkbox"/>	i. Control 7.5 – Low Control W/S		
<input type="checkbox"/>	ii. Control 200 – High Control W/S		
<input type="checkbox"/>	4. 100 µL Iridium Internal Standard Working Solution;		
<input type="checkbox"/>	5. QS with 2% Nitric Acid in Water Solution mix well;		
<input type="checkbox"/>	F. Transfer controls to labeled 15 mL Falcon® tubes.		
	Note: As prepared, the Low Control corresponds to 0.15 ng thallium / mg hair and the High Control corresponds to 4 ng thallium / mg hair.		
	Unknowns		
<input type="checkbox"/>	Optional: If specimen size allows, positive specimens are repeated following an external wash. Submerge the hair specimen in acetone, and mix for 10 minutes. Repeat the process three times with deionized water for ten minutes each, and finally for 10 more minutes with acetone. When the unknown hair specimen is washed in this manner, a negative control should also be contemporaneously washed and analyzed alongside the unknown. Allow the specimen to dry in a fume hood or in a heating block at 50°C.		
	G. To individually labeled 5 mL Falcon® tubes:		
<input type="checkbox"/>	1. Add a minimum of 5.5 mg of specimen hair in duplicate		
<input type="checkbox"/>	2. Accurately record the mass to the nearest 0.1 mg.		

	<i>Note: 5.5 mg is a small amount of hair. In order to ensure that a representative hair sample is analyzed, a larger amount of hair may be cut into small snippets and mixed before removing the 5.5 mg sample. Alternatively, a larger hair sample may be cryoground to mix it well.</i>		
<input type="checkbox"/>	3. Based upon the recorded mass, add enough TMAH to establish a solution of 5 mg of hair per 100 μ L of TMAH. (For example, 110 μ L TMAH is needed for 5.5 mg of hair.)		
<input type="checkbox"/>	4. Allow the hair to completely digest, vortexing occasionally. (This process typically takes at least 8 hours, and the process may be left to proceed overnight.)		
	H. To individual 10 mL Vitlab [®] volumetric flasks that have been washed with 2% Nitric Acid in Water Solution add:		
<input type="checkbox"/>	1. Approximately 5 mL 2% Nitric Acid in Water Solution		
<input type="checkbox"/>	2. 100 μ L of specimen hair digest (step G, above)		
<input type="checkbox"/>	3. 100 μ L of deionized water		
<input type="checkbox"/>	4. 100 μ L Iridium Internal Standard Working Solution;		
<input type="checkbox"/>	5. QS with 2% Nitric Acid in Water Solution mix well;		
<input type="checkbox"/>	I. Transfer samples to labeled 15 mL Falcon [®] tubes.		
<input type="checkbox"/>	J. Centrifuge all samples at 3500 rpm for 15 minutes and transfer to new labeled 15 mL Falcon [®] tubes before analysis by ICP/MS.		
<input type="checkbox"/>	K. When setting up the sequence, run a nitric acid blank four times followed by a Blank with Internal Standard, followed by a Negative Control. Then run the calibrators, followed by the unknowns bracketed by the Positive Controls. A Blank with Internal Standard followed by a Negative Control may be analyzed between specimens and before the Positive Controls.		

7 ANALYTICAL PARAMETERS

The following conditions are written to follow Thermo-Fisher's LabBooks software package.

Analytes: TI (Thallium) and Ir (Iridium)

7.1 Acquisition Parameters

Identifier	Dwell time (s)	Channels	Spacing (u)	Measurement mode	Resolution
205TI (KED)	0.05	1	0.1	KED	Normal
191Ir (KED)	0.05	1	0.1	KED	Normal
				# sweeps = 20	

7.2 Monitor Analytes

	Uptake	Wash
Minimum	30	30
Maximum	300	300

7.3 Survey Scan Settings

Start mass (u)	End mass (u)	Dwell Time (s)	Spacing (u)	Resolution	Measurement mode
4.60	245.00	0.01	0.2	Normal	KED

7.4 Interference Correction

Not applicable

7.5 Standards

Covered in calibration/control section

7.6 Quantification

Analyte	Measurement mode	Quantify	Internal Standard	Fit type	Weighting	Forcing	Use for Semi-Quant
205TI (KED)	KED	Yes	193 Ir (KED)	Linear	None	Blank	Yes
191Ir (KED)	KED	No	Use as Internal Standard	Linear	None	Blank	Yes
IS Recovery		Low warning limit: 80%		Low failure limit: 75%			
		High warning limit: 120%		High failure limit: 125%			

7.7 Ratios

Not applicable

7.8 Quality Control

7.8.1 Calibration Tests / Laboratory Control Standard (LCS)

Analyte	Low failure limit	Low warning limit	High warning limit	High failure limit
205Tl (KED)	75%	80%	120%	125%

7.8.2 Continuous Tests / Relative Stability Verification (RSV)

Analyte	Verify	Ignore concentration below	unit	Concentration warning limit	Concentration failure limit
205Tl (KED)	concentration	10	ppb	5%	10%

7.9 Autosampler

Time Settings:	Wash Time (s):	200	Take up Time (s):	45
Rack Settings:	Rack 1 Type:	60-vials (12x5)	Rack 2 Type:	60-vials (12x5)
Autotune Settings:	Autotune rack:	Standard	Autotune vial:	1
Rinse settings	Rinse Rack:	Rinse Station		

8 DATA ANALYSIS

8.1 Decision Criteria

8.1.1 Batch Acceptance Criteria

Thallium should not be detected in the Negative Control Hair specimen above a level of 0.02 ng/mg.

Thallium in the Positive Control Hair specimens should quantitate within $\pm 20\%$ of the target value. See TOX-101 for more information.

8.1.2 Sample Acceptance Criteria

The Iridium (Internal Standard) response in the unknown specimen(s) should fall within 80% and 120% of the Internal Standard response for the initial blank in the run.

8.2 Calculations

Quantitation is performed by constructing a multi-point calibration curve based on the ratio of the intensity for each calibrator level and the internal standard. The curve is forced through a Negative Control / Zero Calibrator specimen with no weighting. See TOX-101 for acceptable practices in calculating quantitative results.

For hair specimens, 5 mg of hair is used in the place of 0.1 mL of liquid. Therefore, results as received from the instrument for hair should be divided by 50. For example, if a result of 100 $\mu\text{g/L}$ of Tl is obtained for a hair specimen, that corresponds to 2 ng Tl per mg of hair.

When a hair specimen contains thallium above the method's lower limit of quantitation, it may be analyzed again using a method of standard addition to verify the Tl concentration.

9 REPORTING

9.1 Measurement Uncertainty

Refer to CHEM-100 and TOX-101.

9.2 Interpretation

Normal hair contains thallium levels less than 0.02 ng/mg. Chronic thallium hair levels in occupational workers have ranged from 0.020 to 0.57 ng/mg. During acute thallium toxicity, thallium hair levels can be greater than 1 ng/mg.

10 CORRECTIVE MEASURES

Refer to TOX-101 for guidance.

11 PERFORMANCE CHARACTERISTICS

11.1 LOD

Limit of Detection = 0.02 ng/mg (1 $\mu\text{g/L}$)

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11.2 LOQ

Limit of Quantitation = 0.02 ng/mg (1 µg/L)

11.3 Linearity

0.02 – 5.00 ng/mg (1 µg/L - 250 µg/L)

11.4 Bias / Precision

Hair:	@ 0.15 ng/mg (7.5 µg/L)	@ 2.4 ng/mg (1200 µg/L)	@ 4 ng/mg (200 µg/L)
% Bias	-3.63%	-3.68%	-2.91%
% Repeatability	1.83%	1.63%	1.16%
% Intermediate Precision	2.49%	2.79%	2.63%

11.5 Carryover

No carryover was observed when a negative control hair specimen was analyzed immediately following a 5 ng/mg (250 µg/L) calibrator.

12 LIMITATIONS

12.1 Interferences

No endogenous material/matrices interfered with the analysis of thallium. Additionally, elemental standards of gallium, arsenic, indium, rhodium, cadmium, lead, mercury, bismuth, holmium, lithium, scandium, terbium, yttrium, barium, cerium, cobalt and uranium did not interfere with the thallium signal.

12.2 Processed Sample Stability

Sample stability was evaluated up to four days after the initial analysis and remained within ± 20% with refrigerated storage.

13 SAFETY

Take standard precautions for the handling of chemicals and biological materials. Refer to the *FBI Laboratory Safety Manual* for guidance.

14 REVISION HISTORY

Revision	Issued	Changes
01	02/11/2022	Document reformat. Minor updates to wording.

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