BIOSPECTRA
100 Majestic Way, Bangor, PA 18013 / www.biospectra.us

ANALYTICAL METHOD VALIDATION REPORT: DETERMINATION OF ICH Q3D ELEMENTAL IMPURITIES BY INDUCTIVELY COUPLED PLASMA MASS SPECTROMETRY (ICP-MS) IN WATER FOR INJECTION

TABLE OF CONTENTS

1. PURPOSE:

1.1. The purpose of this validation report is to establish documented evidence that the validation protocol, BSI-PRL-0554 v. 1.0, for Elemental Impurities in Water for Injection products performs according to USP and BioSpectra requirements.

1.1.1. Elements under USP <232> will be considered and are as follows:

1.1.1.1. Class 1: Hg, As, Cd, and Pb

1.1.1.2. Class 2A: Co, V, and Ni

1.1.1.3. Class 2B: Tl, Au, Pd, Ir, Os, Rh, Ru, Se, Ag, and Pt

1.1.1.4. Class 3: Li, Sb, Sn, Ba, Mo, Cu, and Cr

1.1.1.5. Class 4: Al, Ca, Fe, K, Mg, Na and Zn

2. SCOPE:

2.1. Applies to Water for Injection (WFI) and related products manufactured at BioSpectra.

2.2. Applies to the NexION 350X S/N 85VN5093001 ICP-MS located in the Quality Control (QC) Laboratory at the BioSpectra Bangor, PA facility.

2.3. This report applies to the validation protocol for elemental impurities in Water for Injection by Inductively Coupled Plasma-Mass Spectrometry (ICP-MS) performed at BioSpectra Inc.

3. REFERENCES:

3.1. BSI-PRL-0554: Analytical Method Validation Protocol: Determination of Elemental Impurities in Water for Injection

3.2. BSI-SOP-0303: NexION 350X ICP-MS SOP

3.3. BSI-SOP-0304: NexION 350X ICP-MS Care and Maintenance SOP

3.4. BSI-SOP-0436: Analytical Method Validation Master Plan

3.5. ICH Guideline for Elemental Impurities Q3D

3.6. USP <730> Plasma Spectrochemistry

3.7. USP <1730> Plasma Spectrochemistry—Theory and Practice

3.8. NexION Operation with Syngistix Software Guide

3.9. USP <232>, <233>

4. BACKGROUND:

4.1. This validation was executed using a parenteral PDE (permissible daily exposure) daily dose calculation of 1,000 grams per day and was performed as per ICH Q3D and USP General Chapters <232> and <233>, Elemental Impurities –Procedures, Validation of Quantitative Procedures. (See Table 1)

4.2. The test protocol validation report includes the following parameters:

4.2.1. Specificity

4.2.2. Linearity and Range

4.2.3. Limit of Quantification (LOQ)

4.2.4. Accuracy by "Spiked Recovery"

4.2.5. Precision (Repeatability)

4.2.6. Intermediate Precision (Ruggedness)

4.2.7. Standard and Sample Solution Stability

TABLE 1: LIMITS FOR WATER FOR INJECTION (1,000 GRAM/DAY EXPOSURE)						
Elements	**ICH Class**	**Parenteral PDE Limits (µg/day)**	**0.3J LOQ (µg/kg) in sample**	**0.5J Target (µg/kg) in sample**	**1.0J Target (µg/kg) in sample**	**1.5J Target (µg/kg) in sample**
As	1	15	4.5	7.5	15	22.5
Cd	1	2.0	0.60	1.0	2.0	3.0
Hg	1	3.0	0.90	1.5	3.0	4.5
Pb	1	5.0	1.5	2.5	5.0	7.5
Co	2A	5.0	1.5	2.5	5.0	7.5
Ni	2A	20	6.0	10	20	30
V	2A	10	3.0	5.0	10	15
Tl	2B	8.0	2.4	4.0	8.0	12
Se	2B	80	24	40	80	120
Ag	2B	10	3.0	5.0	10	15
Au	2B	100	30	50	100	150
Pd	2B	10	3.0	5.0	10	15
Ir	2B	10	3.0	5.0	10	15
Os	2B	10	3.0	5.0	10	15
Pt	2B	10	3.0	5.0	10	15
Rh	2B	10	3.0	5.0	10	15
Ru	2B	10	3.0	5.0	10	15
Ba	3	700	210	350	700	1,050
Sb	3	90	27	45	90	135
Li	3	250	75	125	250	375
Mo	3	1,500	450	750	1,500	2,250
Cu	3	300	90	150	300	450
Sn	3	600	180	300	600	900
Cr	3	1,100	330	550	1,100	1,650
Al	4	*25	7.5	12.5	25	37.5
Ca	4	*750	225	375	750	1,125
Fe	4	*200	60	100	200	300
K	4	*500	150	250	500	750
Mg	4	*500	150	250	500	750
Na	4	*500	150	250	500	750
Zn	4	*200	60	100	200	300

*No PDE limits for Class 4 elements; limits derived from other internal product specifications.

5. MATERIALS AND EQUIPMENT:

TABLE 2: EQUIPMENT

Type	Supplier	Model	Serial Number	Cal. Due
Analytical Balance	Sartorius	MSE224S	36707108	10/2022
Automatic Pipette	Rainin	E4-XLS (2-20 µL)	C040200714	06/30/22
Automatic Pipette	Rainin	E4-XLS (20-200 µL)	C016314640	06/30/22
Automatic Pipette	Rainin	E4-XLS (100-1000 µL)	C016314969	06/30/22
Automatic Pipette	Rainin	E4-XLS (0.5-5 mL)	C023506909	06/30/22
ICP-MS	Perkin Elmer	NexION 350X	85VN5093001	01/2023
Deionized water system	Millipore	IQ-7005/ Element POD	F9SA14284H	06/2022

TABLE 3: REAGENTS

Type	Grade	Supplier	Catalog Number	Lot Number	Expiration
70% Nitric Acid	Trace Metal	VWR	87003-261	1121110	12/08/23
36% Hydrochloric Acid	Trace Metal	VWR	87003-253	4121050	06/03/24
Sulfuric Acid	Trace Metal	Fisher	A510-P212	3120012	04/13/24
Deionized water	Type 1 Ultrapure	In-House	N/A	N/A	N/A
Thiourea	99+% Pure	ACROS	220052500	A0407315	10/31/23
ICP-MS Setup Solution	N/A	Perkin Elmer	N8145051	37-147GSX1	03/30/23
ICP-MS KED Setup Solution	N/A	Perkin Elmer	N8145052	36-191GST1	08/30/22
SiliaPrep SPE Filter	Silica-Based AMPA	Silicycle	R85130B	175959	N/A

5.1. Consumable Supplies
- 5.1.1. SCP Digitubes® 15 mL, 50 mL and 100 mL
- 5.1.2. Pipette Tips of various sizes

TABLE 4: REFERENCE STANDARDS				
Identification	**Manufacturer**	**Lot Number**	**Expiration**	**Concentrations / Elements**
Pharma-CAL Standard Parenteral STD# 1 IA 140-131-201	SCP Science	S220310005	03/2023	Ag (10 µg/mL), As (15 µg/mL), Cd (2 µg/mL), Co (5 µg/mL), Hg (3 µg/mL), Ni (20 µg/mL), Pb (5 µg/mL), Se (80 µg/mL), Tl (8 µg/mL), V (10 µg/mL)
USP232/ICH Q3D Parenteral STD# 2 IA 140-131-215	SCP Science	S210811029	11/2022	Au (100 µg/mL); Ir, Os, Pd, Pt, Rh, & Ru (10 µg/mL)
Pharma-CAL Standard Parenteral STD# 3 IA 140-131-221	SCP Science	S210331019	07/2022	Ba (700 µg/mL), Cr (1,100 µg/mL), Cu (300 µg/mL), Li (250 µg/mL), Mo (1,500 µg/mL), Sb (90 µg/mL), Sn (600 µg/mL)
Aluminum Stock Standard N9300184	Perkin Elmer	25-165ALY1	04/30/23	Al (1,000 µg/mL)
Calcium Stock Standard N9303763	Perkin Elmer	25-39CAY1	08/30/22	Ca (1,000 µg/mL)
Iron Stock Standard N9303771	Perkin Elmer	25-59FEY1	08/30/22	Fe (1,000 µg/mL)
Magnesium Stock Standard N9300179	Perkin Elmer	25-11MGY1	08/30/22	Mg (1,000 µg/mL)
Potassium Stock Standard N9303779	Perkin Elmer	25-188KY1	04/30/23	K (1,000 µg/mL)
Sodium Stock Standard N9303785	Perkin Elmer	26-01NAY1	03/30/23	Na (1,000 µg/mL)
Zinc Stock Standard N9300178	Perkin Elmer	25-145ZNY1	11/30/22	Zn (1,000 µg/mL)
Pharma-CAL Custom Standard AQ0-086-125 (Internal Standard)	SCP Science	S210827010	09/2022	Be, Sc, Y, Re (10 µg/mL); Te (25 µg/mL); Ge, Tb, Bi (5 µg/mL)

6. PROCEDURE:

6.1. All standards were prepared volumetrically from stock solutions purchased from certified vendors. If the vendor supplied stock standard was within 2% of the nominal value as per the certificate of analysis, then the nominal value was used to calculate the concentration of the standard. If the stock standard certificate of analysis value was greater than or less than 2% of the nominal value, then the certificate of analysis value was used for the stock standard concentration.

6.2. Acid Digestion Mixture (Acid Mix)
[2:1] Nitric Acid (HNO_3): Sulfuric Acid (H_2SO_4)

6.2.1. Added 50 mL of nitric acid to a 100 mL Digitube® and then slowly added 25 mL of sulfuric acid. Scaled as required.

6.2.2. Solution was placed in a cold-water bath to aid cooling and was prepared day of use.

6.3. Internal Standard/Complexing Solution

6.3.1. Weighed approximately 1.0 gram of Thiourea into a 50 mL Digitube®.

6.3.2. Added approximately 20 mL of deionized water and mixed to dissolve.

6.3.3. Filtered solution through a SiliaPrep Cation Solid Phase Extraction (SPE) cartridge.

6.3.4. Transferred 2.5 mL of Pharma CAL Custom Standard (Internal standard) Stock to the filtered solution and added 25 mL of hydrochloric acid.

6.3.5. Diluted to a final volume of 50 mL with deionized water and mixed well.

6.3.6. Scaled proportionally as needed for use.

6.4. 2% Thiourea Solution

6.4.1. Weighed approximately 1.0 gram of Thiourea into a 50 mL Digitube®.

6.4.2. Added approximately 20 mL of deionized water and mixed to dissolve.

6.4.3. Filtered solution through a SiliaPrep Cation Solid Phase Extraction (SPE) cartridge.

6.4.4. Diluted to a final volume of 50 mL with deionized water and mixed well.

6.4.5. Scaled proportionally as needed for use.

6.5. Intermediate Standard Preparation

6.5.1. Prepared a standard solution containing the elements listed in Table 5 using the standards STD#1 IA, STD#2 IA, STD#3 IA, and individual single source 1,000 µg/mL standards. Prepared by adding stock standards to a 15 mL Digitube®. Added DI water to approximately 8 mL then added hydrochloric acid. Diluted to volume using DI Water.

TABLE 5: INTERMEDIATE STANDARD

Identification	Element	Stock Identification	Amount Added (mL)	HCl (mL)	Final Volume (mL)	Final Concentration (µg/mL)
Intermediate Standard	As	STD# 1 IA 140-131-201*	1.0	1.0	10	1.5
	Cd					0.20
	Hg					0.30
	Pb					0.50
	Co					0.50
	Ni					2.0
	V					1.0
	Tl					0.80
	Se					8.0
	Ag					1.0
	Au	STD# 2 IA 140-131-215*	1.0			10
	Pd					1.0
	Ir					1.0
	Os					1.0
	Pt					1.0
	Rh					1.0
	Ru					1.0
	Ba	STD# 3 IA 140-131-221*	1.0			70
	Sb					9.0
	Li					25
	Mo					150
	Cu					30
	Sn					60
	Cr					110
	Al	1,000 µg/mL Al Std	0.025			2.5
	Ca	1,000 µg/mL Ca Std	0.750			75
	Fe	1,000 µg/mL Fe Std	0.200			20
	K	1,000 µg/mL K Std	0.500			50
	Mg	1,000 µg/mL Mg Std	0.500			50
	Na	1,000 µg/mL Na Std	0.500			50
	Zn	1,000 µg/mL Zn Std	0.200			20

* SCP Science catalog numbers ending in 1 denote 125 mL bottle sizes and catalog numbers ending in 5 denote 500 mL bottle sizes.

6.6. 0.5J Calibration Standard Preparation

6.6.1. Prepared a solution containing the elements listed in Table 6 below in 5.0% HNO_3, 2.5% H_2SO_4, 1.0% HCl, and 0.04% (400 µg/mL) Thiourea. Added intermediate standard to a separate 50 mL Digitube® followed by addition of approximately 35 mL of DI Water. Added acid mixture then diluted to 45 mL using DI Water. Added internal standard/complexing solution and diluted to volume using DI Water.

TABLE 6: 0.5J CALIBRATION STANDARD

Identification	Element	Intermediate Standard (mL)	Acid Mix (mL)	Internal Standard/ Complexing Solution (mL)	Final Volume (mL)	Final Concentration (µg/L)
0.5J Calibration Standard	As	0.050	3.75	1.0	50	1.5
	Cd					0.20
	Hg					0.30
	Pb					0.50
	Co					0.50
	Ni					2.0
	V					1.0
	Tl					0.80
	Se					8.0
	Ag					1.0
	Au					10
	Pd					1.0
	Ir					1.0
	Os					1.0
	Pt					1.0
	Rh					1.0
	Ru					1.0
	Ba					70
	Sb					9.0
	Li					25
	Mo					150
	Cu					30
	Sn					60
	Cr					110
	Al					2.5
	Ca					75
	Fe					20
	K					50
	Mg					50
	Na					50
	Zn					20

6.7. 1.5J Calibration Standard Preparation

6.7.1. Prepared a solution containing the elements listed in Table 7 below in 5.0% HNO_3, 2.5% H_2SO_4, 1.0% HCl, and 0.04% (400 µg/mL) Thiourea. Added intermediate standard to a separate 50 mL Digitube® followed by addition of approximately 35 mL of DI Water. Added acid mixture then diluted to 45 mL using DI Water. Added internal standard/complexing solution and diluted to volume using DI Water.

TABLE 7: 1.5J CALIBRATION STANDARD

Identification	Element	Intermediate Standard (mL)	Acid Mix (mL)	Internal Standard/ Complexing Solution (mL)	Final Volume (mL)	Final Concentration (µg/L)
1.5J Calibration Standard	As	0.150	3.75	1.0	50	4.5
	Cd					0.60
	Hg					0.90
	Pb					1.5
	Co					1.5
	Ni					6.0
	V					3.0
	Tl					2.4
	Se					24
	Ag					3.0
	Au					30
	Pd					3.0
	Ir					3.0
	Os					3.0
	Pt					3.0
	Rh					3.0
	Ru					3.0
	Ba					210
	Sb					27
	Li					75
	Mo					450
	Cu					90
	Sn					180
	Cr					330
	Al					7.5
	Ca					225
	Fe					60
	K					150
	Mg					150
	Na					150
	Zn					60

6.8. 2.0J Calibration Standard Preparation

6.8.1. Prepared a solution containing the elements listed in Table 8 below in 5.0% HNO_3, 2.5% H_2SO_4, 1.0% HCl, and 0.04% (400 µg/mL) Thiourea. Added intermediate standard to a separate 50 mL Digitube® followed by addition of approximately 35 mL of DI Water. Added acid mixture then diluted to 45 mL using DI Water. Added internal standard/complexing solution and diluted to volume using DI Water.

TABLE 8: 2.0J CALIBRATION STANDARD

Identification	Element	Intermediate Standard (mL)	Acid Mix (mL)	Internal Standard/ Complexing Solution (mL)	Final Volume (mL)	Final Concentration (µg/L)
2.0J Calibration Standard	As	0.200	3.75	1.0	50	6.0
	Cd					0.80
	Hg					1.2
	Pb					2.0
	Co					2.0
	Ni					8.0
	V					4.0
	Tl					3.2
	Se					32
	Ag					4.0
	Au					40
	Pd					4.0
	Ir					4.0
	Os					4.0
	Pt					4.0
	Rh					4.0
	Ru					4.0
	Ba					280
	Sb					36
	Li					100
	Mo					600
	Cu					120
	Sn					240
	Cr					440
	Al					10
	Ca					300
	Fe					80
	K					200
	Mg					200
	Na					200
	Zn					80

6.9. Calibration Blank

6.9.1. Prepared a solution containing 5.0% HNO_3, 2.5% H_2SO_4, 1.0% HCl and 0.04% (400 µg/mL) Thiourea as per Table 9 below. Internal Standard Solution was not allowed to contact concentrated acids. To a separate 50 mL Digitube®, added approximately 35 mL of DI Water. Added acid mixture then diluted to 45 mL using DI Water. Added Internal Standard/Complexing Solution anddiluted to volume using DI Water.

TABLE 9: CALIBRATION BLANK

Description	Acid Mix (mL)	Internal Standard/ Complexing Solution (mL)	Final Volume (mL)
Cal Blank	3.75	1.0	50

6.10. Method Blank Preparation

6.10.1. Added 35 mL of deionized water to a 50 mL Digitube®.

6.10.2. Added 3.75 mL of Acid Digestion Mixture.

6.10.3. Added deionized water to approximately 45 mL and then transferred 1.0 mL of Internal Standard/Complexing Agent Solution.

6.10.4. Diluted to a final volume of 50 mL using deionized water and mixed well.

6.11. Sample Preparation

6.11.1. Weighed approximately 10 grams of sample into a 50 mL Digitube®.

6.11.2. Added 3.75 mL of Acid Digestion Mixture and allowed to react with gentle swirling periodically to evolve any gasses produced during the reaction.

6.11.3. Added deionized water to approximately 45 mL and then transferred 1.0 mL of Internal Standard/ Complexing Solution.

6.11.4. Diluted to a final volume of 50 mL with deionized water and mixed thoroughly.

6.12. Isobaric Overlap Corrections

6.12.1. An isobaric interference results from equal mass isotopes of different elements present in the sample solution. Analysis sequences that are processed utilizing multi-element standards will require the use of correction equations to compensate for known isobaric overlaps originating from the elemental standard and sample. The following correction equations should be used.

KED Mode:

$M_c(58) = M_u(58) \times 1 - M_{(rm)}(57) \times 0.13208$
$M_c(98) = M_u(98) \times 1 - M_{(rm)}(99) \times 0.14655$
$M_c(106) = M_u(106) \times 1 - M_{(rm)}(111) \times 0.09766$
$M_c(108) = M_u(108) \times 1 - M_{(rm)}(111) \times 0.06953$
$M_c(120) = M_u(120) \times 1 - M_{(rm)}(125) \times 0.01273$
$M_c(123) = M_u(123) \times 1 - M_{(rm)}(125) \times 0.12588$
$M_c(190) = M_u(190) \times 1 - M_{(rm)}(195) \times 0.00036$
$M_c(192) = M_u(192) \times 1 - M_{(rm)}(195) \times 0.02315$
$M_c(196) = M_u(196) \times 1 - M_{(rm)}(202) \times 0.005023$

The correction equations can be derived from the following equation:
$Mc = M_u - [M_{(rm)} \times (A_{(ie)}/A_{(rm)})]$

Where:

M_c = Corrected Count Rate for the analyte
M_u = Uncorrected count rate for the analyte
$M_{(rm)}$ = Count Rate of Reference Mass (rm) for the Interfering Element
$A_{(ie)}$ = Percent Abundance of Interfering Element (ie) at the analyte mass
$A_{(rm)}$ = Percent Abundance of Interfering Element at the Reference Mass (rm)

Example:

$M_c(58) = M_u(58) \times 1 - M_{(rm)}(57) \times (0.28 / 2.12)$

6.13. All correction coefficients were calculated based on the Agilent Technologies 2016 Relative Isotopic Abundance Table.

6.14. Multiplier used in the correction equation may differ slightly from the multiplier used in the Syngistix instrument method due to rounding.

7. INSTRUMENT PROCEDURE:

7.1. Performed the ICP-MS daily performance check prior to beginning the analytical sequence. Refer to NexION 350X ICP-MS SOP BSI-SOP-0303 for Daily Check procedures.

7.2. A calibration curve of no less than two standards and a blank was used. The calibration correlation coefficient (R) was ≥ 0.99.

7.3. Set up the sequence as per Table 10.

7.4. Confirmed the calibration by analyzing the 1.5J standard after the calibration. The calibration check must recover ± 20% of the calculated theoretical concentration for multi-element analysis and ± 10% for single element determinations.

7.5. The check standard was verified after each calibration. A re-analysis of the check standard was performed a minimum of once every 10 samples and at the end of the analytical run.

7.6. Bracketing standard checks were verified to recover ± 20% of the calculated theoretical concentration for multi-element analysis. Additionally, the drift (calculated as absolute difference) between bracketing standard checks was verified to be NMT 20% for each target element.

7.7. The sample concentration is calculated as:

$$\text{Conc. } (\mu g/g) = \frac{\text{Solution Conc. } (\mu g/L) \times \text{Solution vol. } (L) \times \text{Dilution Factor}}{\text{Sample Mass } (g)}$$

TABLE 10: EXAMPLE SAMPLE ANALYSIS SEQUENCE

ID	Type	Level
Cal Blank	Cal Blank	Level 1
0.5J Cal Std	Cal Std	Level 2
1.5J Cal Std	Cal Std	Level 3
2.0J Cal Std	Cal Std	Level 4
Cal Blank Check	QC Check	N/A
1.5J Check Std 1	QC Check	N/A
Method Blank	Sample	N/A
Sample(s) 10 or less	Sample	N/A
1.5J Check Std 2	QC Check	N/A

7.8. Instrument Setup and Parameters

7.8.1. Instrument settings are only listed as guidelines. Settings may be changed in order to accommodate changes in sample matrix or hardware configurations.

7.8.2. The AMS-II makeup gas was engaged during analysis using a minimum dilution gas ratio of 15%.

7.8.3. The elements aluminum, arsenic, iron, and selenium were analyzed using hydrogen reaction gas in order to remove polyatomic interferences. A hydrogen DRC (Dynamic Reaction Cell) flow rate of approximately 4 mL/min was used.

TABLE 11: ICP-MS PARAMETERS

ICP-MS System	Perkin Elmer NexION 350X Inductively Coupled Plasma Mass Spectrometry (ICP-MS) with Syngistix Software Version 2.4
Sweeps/Readings	20
Replicates	3
Nebulizer Gas	Argon
Collision Cell Gas	Helium
Reaction Cell Gas	Hydrogen
Dilution Gas	Argon
Sample and Skimmer Cone	Platinum
Sample Rinses	Rinse-1: 60 sec at 45 rpm 5.0% HNO_3, 2.5% HCl with 0.04% Thiourea (or as applicable to mitigate carry over)

TABLE 12: LINEAR RANGE AND CORRESPONDING TUNE MODE							
Isotope	**Internal Standard**	**Mode**	**Linear Range (µg/L)**	**Isotope**	**Internal Standard**	**Mode**	**Linear Range (µg/L)**
7Li	9Be	STD	15-100	106Pd	185Re	KED	0.60-4.0
23Na	45Sc	KED	30-200	107Ag	185Re	KED	0.60-4.0
24Mg	45Sc	KED	30-200	108Pd	185Re	KED	0.60-4.0
27Al	45Sc	STD	1.5-10	109Ag	185Re	KED	0.60-4.0
27Al	45Sc	H_2 DRC	1.5-10	111Cd	125Te	KED	0.12-0.80
39K	45Sc	KED	30-200	113Cd	125Te	KED	0.12-0.80
44Ca	45Sc	KED	45-300	118Sn	125Te	KED	36-240
51V	45Sc	KED	0.60-4.0	119Sn	125Te	KED	36-240
52Cr	45Sc	KED	66-440	120Sn	125Te	KED	36-240
53Cr	45Sc	KED	66-440	121Sb	125Te	KED	5.4-36
56Fe	45Sc	H_2 DRC	12-80	123Sb	125Te	KED	5.4-36
57Fe	72Ge	KED	12-80	135Ba	89Y	KED	42-280
58Ni	72Ge	KED	1.2-8.0	137Ba	89Y	KED	42-280
59Co	72Ge	KED	0.30-2.0	138Ba	89Y	KED	42-280
60Ni	72Ge	KED	1.2-8.0	188Os	185Re	KED	0.60-4.0
62Ni	72Ge	KED	1.2-8.0	189Os	185Re	KED	0.60-4.0
63Cu	72Ge	KED	18-120	190Os	185Re	KED	0.60-4.0
65Cu	72Ge	KED	18-120	191Ir	185Re	KED	0.60-4.0
66Zn	72Ge	KED	12-80	192Os	185Re	KED	0.60-4.0
67Zn	72Ge	KED	12-80	193Ir	185Re	KED	0.60-4.0
68Zn	72Ge	KED	12-80	194Pt	185Re	KED	0.60-4.0
75As	72Ge	H_2 DRC	0.90-6.0	195Pt	185Re	KED	0.60-4.0
75As	72Ge	KED	0.90-6.0	196Pt	185Re	KED	0.60-4.0
77Se	89Y	H_2 DRC	4.8-32	197Au	185Re	KED	6.0-40
78Se	89Y	H_2 DRC	4.8-32	199Hg	185Re	KED	0.18-1.2
95Mo	89Y	KED	90-600	200Hg	185Re	KED	0.18-1.2
97Mo	89Y	KED	90-600	202Hg	185Re	KED	0.18-1.2
98Mo	89Y	KED	90-600	203Tl	209Bi	KED	0.48-3.2
99Ru	89Y	KED	0.60-4.0	205Tl	209Bi	KED	0.48-3.2
101Ru	89Y	KED	0.60-4.0	206Pb	209Bi	KED	0.30-2.0
103Rh	125Te	KED	0.60-4.0	207Pb	209Bi	KED	0.30-2.0
105Pd	185Re	KED	0.60-4.0	208Pb	209Bi	KED	0.30-2.0

7.9. Linearity and Range

7.9.1. The ICP-MS linearity study included standards equivalent to the concentrations shown in Table 14 and encompassed the following standards: 30%, 50%, 100%, 150%, and 200% of the Target Concentration. Each standard was prepared in triplicate and analyzed against the calibration curve described in Section 6.6 to Section 6.8. The average standard recovery for each level of the three replicates was then determined.

7.9.2. Linearity study was performed once as the standard preparation in the protocol is the same regardless of the product analyzed.

7.9.2.1. Acceptance Criteria:

7.9.2.1.1. The mean standard recovery for each element at each of the spike levels, as per USP <233> requirement, must be in the range of 70% - 150%.

TABLE 13: LINEARITY STANDARD PREPARATION

Description	Intermediate Standard (mL)	Acid Mix (mL)	Internal Standard/ Complexing Solution (mL)	Final Volume Deionized Water (mL)
Cal Blank Reference	N/A	3.75	1.0	50
0.3J Standard	0.030	3.75	1.0	50
0.5J Standard	0.050	3.75	1.0	50
1.0J Standard	0.100	3.75	1.0	50
1.5J Standard	0.150	3.75	1.0	50
2.0J Standard	0.200	3.75	1.0	50

TABLE 14: LINEARITY STANDARD CONCENTRATIONS					
Element	**0.3J Standard (µg/L)**	**0.5J Standard (µg/L)**	**1.0J Standard (µg/L)**	**1.5J Standard (µg/L)**	**2.0J Standard (µg/L)**
As	0.90	1.5	3.0	4.5	6.0
Cd	0.12	0.20	0.40	0.60	0.80
Hg	0.18	0.30	0.60	0.90	1.2
Pb	0.30	0.50	1.0	1.5	2.0
Co	0.30	0.50	1.0	1.5	2.0
Ni	1.2	2.0	4.0	6.0	8.0
V	0.60	1.0	2.0	3.0	4.0
Tl	0.48	0.80	1.6	2.4	3.2
Se	4.8	8.0	16	24	32
Ag	0.60	1.0	2.0	3.0	4.0
Au	6.0	10	20	30	40
Pd	0.60	1.0	2.0	3.0	4.0
Ir	0.60	1.0	2.0	3.0	4.0
Os	0.60	1.0	2.0	3.0	4.0
Pt	0.60	1.0	2.0	3.0	4.0
Rh	0.60	1.0	2.0	3.0	4.0
Ru	0.60	1.0	2.0	3.0	4.0
Ba	42	70	140	210	280
Sb	5.4	9.0	18	27	36
Li	15	25	50	75	100
Mo	90	150	300	450	600
Cu	18	30	60	90	120
Sn	36	60	120	180	240
Cr	66	110	220	330	440
Al	1.5	2.5	5.0	7.5	10
Ca	45	75	150	225	300
Fe	12	20	40	60	80
K	30	50	100	150	200
Mg	30	50	100	150	200
Na	30	50	100	150	200
Zn	12	20	40	60	80

TABLE 15: LINEARITY PERCENT RECOVERY RESULTS

Isotope	Mode	0.3J Mean	0.5J Mean	1.0J Mean	1.5J Mean	2.0J Mean	Isotope	Mode	0.3J Mean	0.5J Mean	1.0J Mean	1.5J Mean	2.0J Mean
7Li	STD	97	101	101	108	107	106Pd	KED	99	99	100	99	100
23Na	KED	103	102	104	103	105	107Ag	KED	99	100	103	102	102
24Mg	KED	102	99	102	101	102	108Pd	KED	101	98	101	100	99
27Al	STD	101	98	100	101	102	109Ag	KED	100	99	102	101	102
27Al	H_2 DRC	94	98	98	95	96	111Cd	KED	88	99	93	95	91
39K	KED	102	100	100	98	100	113Cd	KED	98	101	99	98	100
44Ca	KED	97	98	98	101	101	118Sn	KED	100	101	102	103	102
51V	KED	101	100	100	96	97	119Sn	KED	100	100	102	102	101
52Cr	KED	99	98	99	97	97	120Sn	KED	100	102	102	103	102
53Cr	KED	100	99	100	97	97	121Sb	KED	99	100	100	98	97
56Fe	H_2 DRC	96	100	104	102	104	123Sb	KED	101	100	101	98	97
57Fe	KED	103	99	105	106	104	135Ba	KED	100	100	101	100	98
58Ni	KED	101	99	101	105	102	137Ba	KED	101	100	100	100	98
59Co	KED	103	98	102	106	104	138Ba	KED	101	100	100	101	99
60Ni	KED	100	99	103	103	102	188Os	KED	101	97	102	99	100
62Ni	KED	99	99	98	107	101	189Os	KED	100	99	102	101	101
63Cu	KED	102	100	103	105	104	190Os	KED	101	100	103	102	101
65Cu	KED	102	100	102	104	103	191Ir	KED	100	99	102	101	101
66Zn	KED	101	99	100	101	101	192Os	KED	100	99	103	100	100
67Zn	KED	101	98	100	100	100	193Ir	KED	101	99	101	100	99
68Zn	KED	100	99	100	101	101	194Pt	KED	101	99	100	98	98
75As	H_2 DRC	98	97	97	92	96	195Pt	KED	99	98	100	99	98
75As	KED	103	100	100	101	95	196Pt	KED	102	99	102	99	99
77Se	H_2 DRC	97	97	95	94	94	197Au	KED	102	99	100	96	96
78Se	H_2 DRC	97	97	97	94	95	199Hg	KED	103	103	104	98	101
95Mo	KED	102	101	101	100	99	200Hg	KED	102	96	95	91	96
97Mo	KED	102	101	100	99	99	202Hg	KED	96	102	101	97	98
98Mo	KED	102	101	101	99	99	203Tl	KED	99	100	102	102	103
99Ru	KED	107	102	102	103	101	205Tl	KED	100	100	102	103	102
101Ru	KED	102	101	100	101	100	206Pb	KED	100	99	101	103	102
103Rh	KED	100	101	103	103	104	207Pb	KED	103	100	102	103	102
105Pd	KED	99	100	102	101	103	208Pb	KED	103	102	104	103	103

All analytes meet Linearity acceptance criteria of 70% - 150%.

7.10. Accuracy

7.10.1. Three (N=3) unspiked samples were prepared for analysis. The unspiked sample preparations were used for spike recovery calculations. Samples were prepared in triplicate at three spiking levels (50%, 100%, and 150% of the 1.0J Target Concentration) as shown in Table 1. The solutions were analyzed by ICP-MS, as per the method, by a single analyst.

$$\% \text{ Recovery} = \frac{(\text{Conc. of spiked replicate} - \text{Average Conc. of 3 unspiked samples}) \times 100}{\text{Expected spiked concentration}}$$

7.10.1.1. Acceptance Criteria

7.10.1.1.1. The mean spike recovery for each element at each of the three spike levels, as per USP <233> requirement, must be in the range of 70% - 150%.

7.10.2. Spiked Reference (Unspiked) Solution Preparation

7.10.2.1. Prepare as per section 6.11.

7.10.3. Spike Recovery Sample Preparation

7.10.3.1. Weighed the appropriate amount of sample as per Table 16 into a 50 mL Digitube®.

7.10.3.2. Pipetted appropriate intermediate standard spike amount as per Table 16 and swirled to mix.

7.10.3.3. Pipetted 3.75 mL of Acid Mixture and allowed to react. Swirled solution to mix.

7.10.3.4. Added deionized water to 45 mL and transferred 1.0 mL of Internal Standard/ Complexing Solution.

7.10.3.5. Diluted to a final volume of 50 mL with deionized water and mixed well.

7.10.3.6. Prepared spiked sample solutions in triplicate and three preparations of unspiked sample solutions.

TABLE 16: ACCURACY SAMPLE SPIKES

Description	Sample Amount (g)	Intermediate Standard Spike (mL)	Acid Mix (mL)	Internal Standard/ Complexing Solution (mL)	Final Volume (mL)
Method Blank	N/A	N/A	3.75	1.0	50
Unspiked	10	N/A	3.75	1.0	50
0.3J Spiked Sample	10	0.030	3.75	1.0	50
0.5J Spiked Sample	10	0.050	3.75	1.0	50
1.0J Spiked Sample	10	0.100	3.75	1.0	50
1.5J Spiked Sample	10	0.150	3.75	1.0	50

TABLE 17: ACCURACY RESULTS FOR WATER FOR INJECTION
(Mean percent recovery of triplicate preparations)

Isotope	Mode	0.5J Mean	1.0J Mean	1.5J Mean	Isotope	Mode	0.5J Mean	1.0J Mean	1.5J Mean
7Li	STD	103	96	101	106Pd	KED	102	99	101
23Na	KED	101	99	99	107Ag	KED	103	98	102
24Mg	KED	101	99	99	108Pd	KED	102	99	101
27Al	STD	101	98	97	109Ag	KED	100	98	100
27Al	H_2 DRC	101	99	95	111Cd	KED	93	99	97
39K	KED	96	99	99	113Cd	KED	98	99	99
44Ca	KED	97	99	101	118Sn	KED	99	101	102
51V	KED	97	98	98	119Sn	KED	100	102	101
52Cr	KED	95	100	100	120Sn	KED	100	101	101
53Cr	KED	95	100	99	121Sb	KED	97	101	101
56Fe	H_2 DRC	98	100	101	123Sb	KED	97	103	104
57Fe	KED	104	102	100	135Ba	KED	99	97	98
58Ni	KED	100	102	100	137Ba	KED	98	98	100
59Co	KED	101	100	99	138Ba	KED	98	97	98
60Ni	KED	100	103	101	188Os	KED	100	97	100
62Ni	KED	99	100	97	189Os	KED	99	99	100
63Cu	KED	101	101	100	190Os	KED	100	98	99
65Cu	KED	100	102	101	191Ir	KED	99	99	101
66Zn	KED	98	101	98	192Os	KED	99	98	101
67Zn	KED	97	99	94	193Ir	KED	99	98	99
68Zn	KED	97	98	96	194Pt	KED	97	96	97
75As	H_2 DRC	91	98	97	195Pt	KED	95	98	99
75As	KED	93	103	100	196Pt	KED	97	96	99
77Se	H_2 DRC	93	98	97	197Au	KED	94	98	99
78Se	H_2 DRC	96	99	97	199Hg	KED	97	106	103
95Mo	KED	94	97	99	200Hg	KED	94	102	98
97Mo	KED	96	97	100	202Hg	KED	92	102	98
98Mo	KED	96	98	100	203Tl	KED	101	100	101
99Ru	KED	98	100	102	205Tl	KED	101	101	102
101Ru	KED	99	98	99	206Pb	KED	102	99	99
103Rh	KED	101	102	103	207Pb	KED	101	101	100
105Pd	KED	101	99	100	208Pb	KED	101	99	100

All elements meet Accuracy acceptance criteria of 70% - 150%.

7.11. Specificity

7.11.1. Specificity was demonstrated by using a calibration blank and spiked calibration blank for ICP-MS analysis. The calibration blank was prepared as per the analytical method protocol. A separate blank was spiked with a mixed standard solution which produced a spiked solution at a concentration equivalent to the 2.0J calibration standard.

7.11.2. The solutions were analyzed as per the analytical method and the intensities for the calibration blank and 2.0J calibration standard are reported.

7.11.2.1. Acceptance Criteria:

7.11.2.1.1. The lack of a significant interference (as demonstrated by the spike recovery of 70% to 150%, as per the Accuracy requirement from USP <233>) or by any other element in the spiked blank solution or the solution matrix itself will indicate the specificity of the method.

TABLE 18: SPECIFICITY RESULTS

Isotope	Blank (CPS)	2.0J STD (CPS)	Isotope	Blank (CPS)	2.0J STD (CPS)
7Li	2041	3060543	106Pd	10	14606
23Na	2961	46122	107Ag	23	24817
24Mg	16	13747	108Pd	8	15197
27Al (STD)	11961	454117	109Ag	19	24969
27Al (DRC)	2301	80445	111Cd	6	660
39K	1626	15193	113Cd	5	633
44Ca	53	1059	118Sn	43	466144
51V	131	4744	119Sn	13	172909
52Cr	34	709158	120Sn	59	662876
53Cr	48	88325	121Sb	14	60413
56Fe	2130	1012933	123Sb	-35	48993
57Fe	18	4384	135Ba	53	140413
58Ni	38	28183	137Ba	74	253892
59Co	11	10566	138Ba	518	1714252
60Ni	13	12259	188Os	40	14658
62Ni	3	1996	189Os	31	18004
63Cu	58	485886	190Os	42	29201
65Cu	40	232431	191Ir	67	40455
66Zn	99	22367	192Os	44	46483
67Zn	24	4079	193Ir	72	67869
68Zn	33	17956	194Pt	59	23266
75As (DRC)	42	6201	195Pt	53	24471
75As (KED)	30	2644	196Pt	48	18021
77Se	46	13575	197Au	160	347121
78Se	36	43673	199Hg	36	961
95Mo	8	1020966	200Hg	36	1372
97Mo	14	636426	202Hg	47	1733
98Mo	17	1629104	203Tl	102	23910
99Ru	9	7155	205Tl	57	58715
101Ru	6	9962	206Pb	238	11894
103Rh	18	58676	207Pb	207	10449
105Pd	79	11631	208Pb	437	25735

7.12. Precision

7.12.1. All solutions for the Precision (Repeatability) test were prepared by a single analyst for Water for Injections samples

7.12.2. The value of the unspiked sample preparations from Section 7.10, "Accuracy," was used for spike recovery calculations. Six sample solutions were prepared at the 1.0J Target Concentration as shown in Table 1. For ICP-MS analysis, the Target Concentration spiked samples and the unspiked samples were used for the accuracy experiment.

7.12.3. Precision and %RSD results are reported to the nearest whole number, but values are calculated from data that contains nine decimal places.

7.12.3.1. Acceptance Criteria:

7.12.3.1.1. The %RSD for the spike recovery concentration must be NMT 20% for each element in each sample.

TABLE 19: PRECISION RESULTS FOR WATER FOR INJECTION
(Mean recovery concentration of 6 preparations)

Isotope	Mode	1.0J Mean Recovery Conc. N=6 (µg/kg)	% RSD N=6	Isotope	Mode	1.0J Mean Recovery Conc. N=6 (µg/kg)	% RSD N=6
7Li	STD	239	1	106Pd	KED	10	1
23Na	KED	496	2	107Ag	KED	10	1
24Mg	KED	495	2	108Pd	KED	10	2
27Al	STD	24	1	109Ag	KED	10	1
27Al	H_2 DRC	25	2	111Cd	KED	2	8
39K	KED	501	2	113Cd	KED	2	3
44Ca	KED	754	3	118Sn	KED	604	2
51V	KED	10	1	119Sn	KED	606	2
52Cr	KED	1093	1	120Sn	KED	601	2
53Cr	KED	1097	1	121Sb	KED	91	1
56Fe	H_2 DRC	201	1	123Sb	KED	92	2
57Fe	KED	201	2	135Ba	KED	685	2
58Ni	KED	20	1	137Ba	KED	688	1
59Co	KED	5	1	138Ba	KED	684	1
60Ni	KED	21	1	188Os	KED	10	1
62Ni	KED	20	4	189Os	KED	10	1
63Cu	KED	302	1	190Os	KED	10	1
65Cu	KED	302	2	191Ir	KED	10	1
66Zn	KED	201	2	192Os	KED	10	1
67Zn	KED	193	3	193Ir	KED	10	1
68Zn	KED	195	1	194Pt	KED	10	1
75As	H_2 DRC	15	1	195Pt	KED	10	1
75As	KED	15	3	196Pt	KED	10	2
77Se	H_2 DRC	79	2	197Au	KED	99	1
78Se	H_2 DRC	79	1	199Hg	KED	3	8
95Mo	KED	1467	2	200Hg	KED	3	4
97Mo	KED	1474	1	202Hg	KED	3	4
98Mo	KED	1481	1	203Tl	KED	8	1
99Ru	KED	10	1	205Tl	KED	8	0
101Ru	KED	10	2	206Pb	KED	5	1
103Rh	KED	10	2	207Pb	KED	5	1
105Pd	KED	10	2	208Pb	KED	5	1

All analytes meet Precision RSD% acceptance criteria of NMT 20%.

7.13. Intermediate Precision (Ruggedness)

7.13.1. A second analyst, on a different day from the performance of the Repeatability experiment, prepared and analyzed the Intermediate Precision solutions. Six sample solutions were prepared at the 1.0J Target Concentration level found in Table 1 for ICP- MS analysis (this fulfilled two events as "different day" and "different analyst").

7.13.2. Ruggedness and %RSD results are reported to the nearest whole number, but values are calculated from data that contains nine decimal places.

7.13.2.1. Acceptance Criteria:

7.13.2.1.1. The %RSD for the spike recovery concentration from both analysts (N=12) must be NMT 25% for each element.

TABLE 20: RUGGEDNESS RESULTS FOR WATER FOR INJECTION
(Mean recovery concentration of 12 preparations)

Isotope	Mode	1.0J Mean Recovery Conc. N=12 (µg/kg)	% RSD N=12	Isotope	Mode	1.0J Mean Recovery Conc. N=12 (µg/kg)	% RSD N=12
7Li	STD	237	2	106Pd	KED	10	1
23Na	KED	491	2	107Ag	KED	10	2
24Mg	KED	491	2	108Pd	KED	10	2
27Al	STD	25	1	109Ag	KED	10	1
27Al	H_2 DRC	24	3	111Cd	KED	2	8
39K	KED	494	2	113Cd	KED	2	3
44Ca	KED	736	4	118Sn	KED	601	2
51V	KED	10	2	119Sn	KED	601	2
52Cr	KED	1083	2	120Sn	KED	599	2
53Cr	KED	1085	2	121Sb	KED	91	2
56Fe	H_2 DRC	199	2	123Sb	KED	91	2
57Fe	KED	200	3	135Ba	KED	681	2
58Ni	KED	20	2	137Ba	KED	682	1
59Co	KED	5	2	138Ba	KED	681	1
60Ni	KED	20	3	188Os	KED	10	2
62Ni	KED	20	4	189Os	KED	10	1
63Cu	KED	299	2	190Os	KED	10	1
65Cu	KED	299	2	191Ir	KED	10	1
66Zn	KED	198	2	192Os	KED	10	1
67Zn	KED	194	3	193Ir	KED	10	1
68Zn	KED	195	2	194Pt	KED	10	2
75As	H_2 DRC	15	2	195Pt	KED	10	1
75As	KED	15	3	196Pt	KED	10	2
77Se	H_2 DRC	79	2	197Au	KED	98	1
78Se	H_2 DRC	78	2	199Hg	KED	3	6
95Mo	KED	1469	1	200Hg	KED	3	3
97Mo	KED	1476	1	202Hg	KED	3	4
98Mo	KED	1469	1	203Tl	KED	8	1
99Ru	KED	10	1	205Tl	KED	8	2
101Ru	KED	10	2	206Pb	KED	5	1
103Rh	KED	10	2	207Pb	KED	5	1
105Pd	KED	10	2	208Pb	KED	5	1

All analytes meet the Ruggedness %RSD acceptance criteria of NMT 25%.

7.14. Limit of Quantitation (LOQ)

7.14.1. The limit of quantitation (LOQ) is demonstrated from spike recovery performed at the 10% and 30% Target Concentration spiking levels as shown in Table 1.

7.14.2. Samples were prepared in triplicate following Section 7.10.3 and using amounts listed for 0.3J spiked samples in Table 16 above.

7.14.2.1. Acceptance Criteria:

7.14.2.1.1. The mean percent spike recovery for each element at the 10% and 30% Target Concentration spiking levels, as per the USP <233> accuracy guideline, must be in the range of 70% - 150%.

TABLE 21: LIMIT OF QUANTITATION RESULTS FOR WFI

(Mean percent recovery of 3 preparations)

Isotope	Mode	0.3J Mean % Recovery	Isotope	Mode	0.3J Mean % Recovery
7Li	STD	101	106Pd	KED	102
23Na	KED	97	107Ag	KED	100
24Mg	KED	99	108Pd	KED	101
27Al	STD	101	109Ag	KED	102
27Al	H_2 DRC	100	111Cd	KED	95
39K	KED	94	113Cd	KED	98
44Ca	KED	94	118Sn	KED	98
51V	KED	95	119Sn	KED	98
52Cr	KED	94	120Sn	KED	99
53Cr	KED	95	121Sb	KED	97
56Fe	H_2 DRC	99	123Sb	KED	96
57Fe	KED	101	135Ba	KED	100
58Ni	KED	99	137Ba	KED	99
59Co	KED	103	138Ba	KED	99
60Ni	KED	98	188Os	KED	101
62Ni	KED	99	189Os	KED	100
63Cu	KED	100	190Os	KED	100
65Cu	KED	100	191Ir	KED	100
66Zn	KED	98	192Os	KED	98
67Zn	KED	93	193Ir	KED	98
68Zn	KED	100	194Pt	KED	98
75As	H_2 DRC	97	195Pt	KED	97
75As	KED	96	196Pt	KED	99
77Se	H_2 DRC	96	197Au	KED	96
78Se	H_2 DRC	99	199Hg	KED	104
95Mo	KED	97	200Hg	KED	102
97Mo	KED	98	202Hg	KED	97
98Mo	KED	97	203Tl	KED	100
99Ru	KED	98	205Tl	KED	100
101Ru	KED	98	206Pb	KED	102
103Rh	KED	98	207Pb	KED	102
105Pd	KED	101	208Pb	KED	102

All analytes meet LOQ acceptance criteria of 70% - 150%.

7.15. Sample and Standard Stability

7.15.1. The 50% and 200% Target Concentration level calibration standards were analyzed as samples against calibration curves constructed from freshly prepared calibration standards at T=1 day from the date of preparation.

7.15.2. A spiked sample solution prepared at the 1.0J Target Concentration level in Table 1 from the Ruggedness experiment was used for sample stability for Water for Injection. The spiked sample solution was analyzed against calibration curves constructed from freshly prepared calibration standards at time points T=0 (day of preparation) and T=1 (1 day from the date of preparation).

7.15.2.1. Acceptance Criteria:

7.15.2.1.1. The recovery of each element must be within the range of 80% to 120% recovery of the T = 0 results for the calibration standard.

7.15.2.1.2. The recovery of each element must be within the range of 80% to 120% recovery of the T = 0 results for the spiked sample solution.

TABLE 22: SAMPLE AND STANDARD STABILITY (% RECOVERY)

Isotope	0.5J Std (%)	2.0J Std (%)	1.0J Spike (%)	Isotope	0.5J Std (%)	2.0J Std (%)	1.0J Spike (%)
7Li	98	101	96	106Pd	105	103	97
23Na	108	102	105	107Ag	103	101	101
24Mg	100	102	101	108Pd	100	101	96
27Al (STD)	103	105	103	109Ag	98	100	98
27Al (DRC)	100	100	103	111Cd	90	103	113
39K	103	103	106	113Cd	82	102	101
44Ca	97	105	103	118Sn	90	102	98
51V	106	101	100	119Sn	93	100	100
52Cr	100	101	102	120Sn	90	99	96
53Cr	100	101	102	121Sb	92	102	98
56Fe	94	99	99	123Sb	92	101	99
57Fe	98	102	97	135Ba	96	99	99
58Ni	100	102	101	137Ba	99	100	101
59Co	107	102	106	138Ba	97	100	101
60Ni	96	102	100	188Os	104	101	102
62Ni	94	100	101	189Os	103	103	99
63Cu	102	103	99	190Os	100	98	97
65Cu	103	104	99	191Ir	99	102	99
66Zn	102	103	98	192Os	102	101	101
67Zn	103	103	99	193Ir	100	101	98
68Zn	101	102	100	194Pt	100	101	102
75As (DRC)	101	97	99	195Pt	101	103	103
75As (KED)	99	101	107	196Pt	101	100	104
77Se	105	99	96	197Au	102	102	99
78Se	98	97	98	199Hg	99	97	93
95Mo	97	101	98	200Hg	96	98	97
97Mo	98	102	102	202Hg	100	104	98
98Mo	99	99	99	203Tl	97	98	98
99Ru	95	100	96	205Tl	101	99	99
101Ru	97	99	98	206Pb	95	101	96
103Rh	96	103	100	207Pb	99	100	97
105Pd	102	102	101	208Pb	98	101	96

All analytes meet Sample and Standard Solution Stability acceptance criteria of 80% - 120%.

8. DEVIATIONS:

8.1. Accuracy (1.0J and 1.5J spikes) and Precision studies were performed a second time due to check standard failure in the final bracketing standard. Following the failure, the instrument was thoroughly cleaned. Residue buildup was noted in the injector, which would lead to increased oxide formation, lower gas flow, and overall system suitability issues. After the injector was cleaned, the Makeup Gas Flow optimized value increased from 0.26 L/min to 0.33 L/min. Second attempt at these studies was performed the following day with no issues; all validation parameters passed. Care will be taken in future analyses to ensure there is no residue buildup in the injector.

9. CONCLUSION:

9.1. The test method for Elemental Impurities in Water for Injection products has been validated. The Method was found to be:

9.1.1. Specific: The method blank did not show any significant interference for all analyzed masses.

9.1.2. Linear: 30% to 200% of working standard solution corresponding to 0.3J to 2.0J. Mean percent recovery ranged from 88% to 108%.

9.1.3. Sensitive: LOQ recoveries were within 93% to 104%. All analytes met acceptance criteria established.

9.1.4. Accurate: From 50% (0.5J) to 150% (1.5J) of working standard concentration level with mean percent recoveries ranging from 91% to 106%. All masses analyzed met acceptance criteria within the specified range.

9.1.5. Precise: Closeness of agreement demonstrated between six sample preparations by percent RSD's ranging from 0% to 8%.

9.1.6. Rugged: Satisfactory precision was demonstrated between two sets of six sample preparations performed on different days and by different analysts. The percent RSDs ranged from 1% to 8% and 1% to 8%.

9.1.7. Stable: With respect to stability of solutions, the sample solutions for Water for Injection are shown to be stable for 24 hours for all elements analyzed using this protocol. The working standard preparations were shown to be stable for all analytes under the protocol as well for 24 hours. The samples and standards are to be noted as stable for 24 hours in the final analytical testing method.

10. NOTEBOOK REFERENCE:

TABLE 23: NOTEBOOK REFERENCE

STUDY		NOTEBOOK REFERENCE
Specificity		EIV-7/ pages 49-53
Linearity and Range		EIV-7/ pages 45-48
LOQ by "Spiked" recovery for Water for Injection		EIV-7/ pages 49-53
Accuracy/ Precision by "Spiked" recovery for WFI		EIV-7/ pages 49-58
Intermediate Precision (Ruggedness)		EIV-7/ pages 59-62
Solution Stability	Day-0	EIV-7/ pages 59-62
	Day-1	EIV-7/ pages 63-65