Using Surrogate Matrices for the Determination of Sodium and Potassium in Human Urine using ICP-MS

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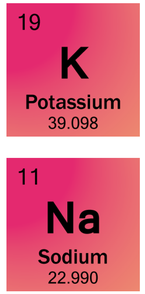
Introduction

Sodium and potassium are essential endogenous electrolytes. Their endogenous levels can significantly vary between individuals, thus the necessity to have methods with adequate quantification limits in order to assess baseline levels. As sodium and potassium are present in relatively high concentrations in human urine, it is difficult to find suitable matrices for the preparation of calibrators. As both elements cannot be stripped out easily from urine using conventional techniques, the use of surrogate matrices that adequately mimic human urine is therefore necessary.



Method

Although deionized water could be used to prepare calibrators, regulatory agencies usually favor the use of matrices which are as close as possible to the study samples. Human urine is primarily composed of water, urea, chloride, sodium, potassium and creatinine. For the development and validation of ICP-MS assays for the determination of sodium and potassium, solutions of the upper mentioned urine components were used for the preparation of calibrators. Surrogate matrices compositions were adjusted in relation to the analyte of interest, omitting salts containing the ion of interest for obvious reasons. Hence, the surrogate matrix used for potassium analysis was composed of urea (9.3 g/L), sodium chloride (3 g/L) and creatinine (0.67 g/L). For sodium analysis the same solution was used replacing sodium chloride by potassium chloride (3 g/L). For the validation of the potassium method, the calibrators and one set of QC samples were prepared in surrogate matrix while a second set of QC samples were prepared in regular human urine. The sodium method was developed using the same approach.

Extraction Procedure

	Sodium	Potassium
Matrix	Human Urine	Human Urine
Analytical Range	100-10000 µg/mL	100-10000 µg/mL
Internal Standard	Beryllium	Strontium
Sample Volume	0.050 mL	0.050 mL
Extraction Type	Dilute and Shoot	Dilute and Shoot
Dilution Factor	10	400

Analytical Conditions

Inductively Coupled Plasma-Mass Spectrometer	Agilent 7500 ce
Autosampler	ASX-570
Acquisition Mode	Spectrum analysis
Number of Masses	2 per methods
Masses	Sodium 23, Beryllium 9 Potassium 39, Strontium 88
Number of Points per Mass	3 (full quant)
Number of Repetition	5
Stabilization Time	5 sec
Uptake Time	30 sec
Total Acquisition Time	10.5 sec
Injector Probe Rinsing Solution	HNO_3 5% in water
Chromatographic Integration	Chemstation G183B
Acquisition data system	Agilent

Table 1. Surrogate Matrix Preparation for the Analysis of Sodium

Component	Concentration in Water
Urea	9.3 g/L
Creatinine	0.670 g/L
Potassium Chloride	3 g/L

Table 2. Surrogate Matrix Preparation for the Analysis of Potassium

Component	Concentration in Water
Urea	9.3 g/L
Creatinine	0.670 g/L
Sodium Chloride	3 g/L

Results

The method for potassium was validated under the analytical range of 100-10000 µg/mL. Mean recovery was similar between the surrogate matrix and human urine. Between-run results were similar between matrices with combine mean biases ranging from 0.21% to 6.62% and %CV ranging from 2.42% to 13.27%. The method was used for study sample analysis with a mean ISR confirmation rate of 95%. Similar performance is expected for the sodium method which is pending validation. However, the between-run accuracy and precision was assessed in a pre-validation run and results are tabulated in **Tables 7 and 8**.

Table 3. Between-Run Accuracy and Precision of Potassium in Human Urine

	Low QC 300.80 µg/mL		Middle QC 5000.80 µg/mL		High QC 7500.80 µg/mL	
	Measured Conc. (µg/mL)	% Bias	Measured Conc. (µg/mL)	% Bias	Measured Conc. (µg/mL)	% Bias
N	36		36		36	
Mean	308.46	2.55	5229.73	4.58	7774.96	3.66
SD(±)	8.62		141.91		188.47	
CV(%)	2.80		2.71		2.42	

Table 4. Between-Run Accuracy and Precision of Potassium in Surrogate Matrix

	Low QC 300.00 µg/mL		Middle QC 5000.00 µg/mL		High QC 7500.00 µg/mL	
	Measured Conc. (µg/mL)	% Bias	Measured Conc. (µg/mL)	% Bias	Measured Conc. (µg/mL)	% Bias
N	42		42		42	
Mean	300.63	0.21	5178.59	3.57	7996.80	6.62
SD(±)	12.17		126.78		1060.98	
CV(%)	4.05		2.45		13.27	

Table 5. Matrix Effect at Low Concentration for Potassium in Human Urine

	Untreated Standard (MFQC1)			Reference Solution (RSQC1)		Calculated Matrix Factor (Analyte)	Calculated Matrix Factor (IS)	IS-Normalized Matrix Factor
	Analyte Signal Count Responses	Corrected Signal Count Responses	Internal Standard Signal Count Responses	Analyte Signal Count Responses	Internal Standard Signal Count Responses			
	901970	122684	714388	153084	724939	0.818887	1.005176	0.814670
	747978	127554	727867	155442	745673	0.851394	1.024142	0.831324
	1023952	120692	675723	147717	689521	0.805591	0.950774	0.847300
	486909	127999	725659	151536	727610	0.854364	1.021035	0.836763
	387267	133910	738906	144867	685166	0.893818	1.039674	0.859710
	865424	120802	725728	146262	691345	0.806326	1.021133	0.789639
Mean				149817.9	710709.0			0.8299010
SD(±)								0.02487338
CV(%)								3.00

Table 6. Matrix Effect at High Concentration for Potassium in Human Urine

	Untreated Standard (MFULOQ)			Reference Solution (RSULOQ)		Calculated Matrix Factor (Analyte)	Calculated Matrix Factor (IS)	IS-Normalized Matrix Factor
	Analyte Signal Count Responses	Corrected Signal Count Responses	Internal Standard Signal Count Responses	Analyte Signal Count Responses	Internal Standard Signal Count Responses			
	5748281	4830229	725636	4818060	705346	1.021029	1.039636	0.982102
	5406628	4719758	703561	4903503	722060	0.997677	1.008008	0.989751
	5591937	4568790	695838	4652968	687823	0.965765	0.996944	0.968725
	5255561	4848846	714715	4889572	728546	1.024964	1.023989	1.000952
	4996683	4729091	684950	4594097	675217	0.999650	0.981344	1.018654
	5300887	4475447	681705	4526285	668834	0.946034	0.976695	0.968607
Mean				4730747.5	697971.1			0.9881318
SD(±)								0.01945654
CV(%)								1.97

Table 7. Between-Run Accuracy and Precision of Sodium in Human Urine

	Low QC 311.73 µg/mL		Middle QC 5011.73 µg/mL		High QC 7511.73 µg/mL	
	Measured Conc. (µg/mL)	% Bias	Measured Conc. (µg/mL)	% Bias	Measured Conc. (µg/mL)	% Bias
Mean	314.57	0.96	4869.44	-2.84	7089.36	-5.62
SD(±)	17.18		126.07		137.51	
CV(%)	5.46		2.59		1.94	

Table 8. Between-Run Accuracy and Precision of Sodium in Surrogate Matrix

	Low QC 100.00 µg/mL		Middle QC 1000.00 µg/mL		High QC 10000.00 µg/mL	
	Measured Conc. (µg/mL)	% Bias	Measured Conc. (µg/mL)	% Bias	Measured Conc. (µg/mL)	% Bias
Mean	100.20	0.20	1031.58	3.16	9837.38	-1.62
SD(±)	1.77		27.49		186.37	
CV(%)	1.77		2.67		1.89	

Spectrums

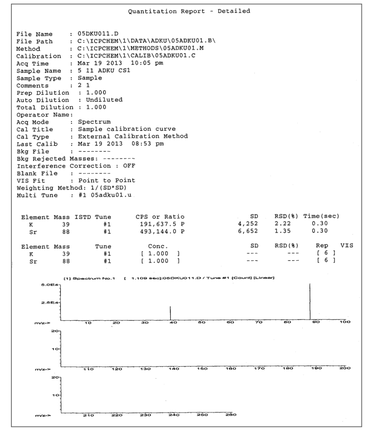


Figure 1. Representative Scan of the Lower Limit of Quantitation Sample in Blank Solution (100 µg/mL) for Potassium

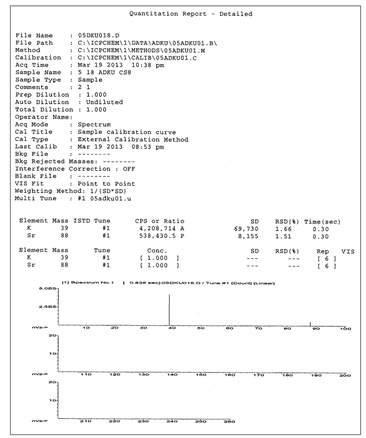


Figure 2. Representative Scan of the Upper Limit of Quantitation Sample in Blank Solution (10000 µg/mL) for Potassium

Conclusion

Reliable ICP-MS quantification methods can be developed for endogenous analytes using surrogate matrices for urine which adequately mimic the study samples matrix. It was demonstrated with the validation of the method for the determination of potassium in human urine that surrogate matrices are adequate to quantify study samples in biological matrices.

TRANSFORMING PROMISING IDEAS INTO COMMERCIAL REALITY