Agilent

Trusted Answers

ICP-OES Analysis of Nutrient Elements for Labeling Compliance of Dietary Food Supplements

Simultaneous measurement of major to trace elements by Agilent 5900 ICP-OES following US FDA EAM 4.4

Author

Alyssa Stevenson

Agilent Technologies, Inc.

Introduction

Dietary supplements are defined by the US Food and Drug Administration (FDA) as a product for consumption that can contain vitamins, minerals, amino acids, herbs or botanicals, concentrates, extracts, or metabolites (*1*). They are intended to supplement the diet rather than replace a meal and are therefore regulated differently to conventional food products (*2*). Most dietary supplements are available as tablets, capsules, gels, liquids, or powders.

Consumer demand for dietary supplements is expected to remain strong over the next few years, driven by the wide choice, availability, convenience, and perceived health and wellbeing benefits of the products. The global market for dietary supplements is forecast to grow annually at a compound growth rate of 8.9% to reach an estimated value of 327.4 billion US dollars by 2030 (*3*).

The FDA regulates both finished dietary supplement products and dietary ingredients under the Dietary Supplement Health and Education Act of 1994 (DSHEA). Per DSHEA, which is an amendment of the Federal Food, Drug, and Cosmetic Act, manufactures and distributors are prohibited from selling adulterated or misbranded products. Companies must comply with the appropriate labeling of nutrients and health claims for dietary supplements, as specified in DSHEA (2).

The method used in this study for the analysis of elemental nutrients and micronutrients in dietary supplements was adapted from FDA Elemental Analysis Manual (EAM) 4.4 (4). EAM 4.4 outlines how to accurately analyze 23 elements in food by Inductively Coupled Plasma-Optical Emission Spectroscopy (ICP-OES) for regulatory purposes. The ICP-OES method is useful for labeling purposes, as well as providing information on the elemental content of some commonly available dietary supplements.

In this study, an Agilent 5900 Synchronous Vertical Dual View (SVDV) ICP-OES was used in axial view mode to determine 24 elements in range of dietary supplement samples following the EAM 4.4 method. The list of analytes included: aluminum, arsenic, barium, boron, cadmium, calcium, chromium, cobalt, copper, iron, lead, magnesium, manganese, molybdenum, nickel, phosphorus, potassium, selenium, sodium, strontium, thallium, vanadium, zinc, plus sulfur. For maximum flexibility and application coverage, the 5900 SVDV ICP-OES can operate in four different modes: SVDV, VDV, axial, and radial (5), ensuring that a lab can transfer any existing ICP-OES methods to the 5900. Axial mode and the MultiCal software routine were used in this study to highlight the versatility of the 5900. The method described in this study can also be used with the Agilent 5800 VDV ICP-OES.

Experimental

Instrumentation
The 5900 SVDV ICP-OES and Agilent SPS 4 autosampler were used for the analysis of dietary supplement samples. The 5900 ICP-OES was fitted with the standard sample introduction system (SIS), which is ideal for these sample types, and keeps instrument set-up simple and repeatable. The SIS comprised a SeaSpray concentric nebulizer, double-pass glass cyclonic spray chamber, and an Easy-fit one-piece torch with a 1.8 mm injector. Instrument operating parameters are shown in Table 1.

The instrumentation and method were set up, optimized, and controlled using Agilent ICP Expert Pro software (version 7.6), which includes smart tools such as IntelliQuant Screening and Intelligent Rinse. The 5900 also includes the integrated AVS 6/7 Advanced Valve System switching valve as standard. To account for the different sample matrices, the Intelligent Rinse function within the ICP Expert software was used to maximize sample throughput by automatically optimizing the rinse time between samples.

To provide the robustness and stability of the plasma over long analytical runs of varied matrix samples, the 5900 ICP-OES uses a vertical plasma, a solid-state radio frequency (SSRF) generator operating at 27 MHz, the high-speed Vista Chip III CCD detector and a Cooled Cone Interface (CCI). Typically, the cooler temperatures in the plasma tail can cause interferences from self-absorption and recombination, usually from the presence of elements at high concentrations (such as Fe, Ca, Mg, Na, and K). These interferences can cause difficulty when viewing the plasma axially. However, the CCI deflects the plasma's cooler tail, avoiding interferences and allowing optimized emission of light from the central-view channel of the plasma into the optics. So, when reading the vertical plasma in axial view mode the required limits of detection (LODs) for trace elements specified in EAM 4.4 can be measured with minimal interfering effects.

Table 1. Agilent 5900 SVDV ICP-OES instrument and method conditions.

Parameter	Setting
Read Time (s)	10
Replicates	3
Sample Uptake Delay (s)	20
Stabilization Time (s)	15
Intelligent Rinse	Enabled
Pump Speed (rpm)	12
RF Power (kW)	1.50
Aux Flow (L/min)	1.00
Plasma Flow (L/min)	12.0
Nebulizer Flow (L/min)	0.7
Viewing Height (mm)	NA
Viewing Mode	Axial
Sample Pump Tubing	White-white
Waste Pump Tubing	Blue-blue
Internal Standard Pump Tubing	Black-black

Easily ionizable elements (EIEs) such as Ca, K, Mg, and Na are common constituents of many dietary supplements. These elements have low ionization energies and are easily ionized in the plasma. If EIEs are present in a sample at a high enough concentration, the plasma electron density and the atomization-ionization equilibria are affected. These effects cause either an enhancement or suppression of the emission signals, leading to the reporting of false high or false low element concentrations. To compensate for the EIE effects, a customized internal standard mix was matched to the analytes, as detailed in Tables 2 and 5. Yttrium (Y) was used to correct elements that have ionic wavelengths and/or high ionization energies. To correct the EIE effects of K, rubidium (Rb) was used. Rb provides the best internal standard correction for K as it behaves in a similar way in the plasma. To correct Na, lithium (Li) was used. For other atomic lines Bi was used as the internal standard. Bismuth (Bi) 223.061 nm is an atomic line, so is preferable to Y 371.029 nm, which is an ionic line. The internal standards were added on-line using a Y-piece connection to the nebulizer.

Table 2. Concentration of internal standard elements used in the mix.

Element and Wavelength (nm)	Concentration (mg/L)
Y 371.029	5
Rb 780.026	50
Bi 223.061	5
Li 670.783	5

Intelligent Rinse

Intelligent Rinse monitors the intensities of nominated element wavelengths during the rinse period and controls the SPS 4 autosampler rinse times (6). The software automatically ends the rinse when element intensities reach a user-specified threshold. There are three defined washout thresholds within Intelligent Rinse: quick, moderate, and thorough. Monitoring signal intensities means that rinse periods vary, depending on the rinse required for each sample.

In this study, Intelligent Rinse modes were selected based on which mode returned the signal below the detection limit for that element in the blank after rinsing (Table 3). Intelligent Rinse reduces carry-over between samples through sufficient rinsing, while optimizing both sample throughput and argon usage.

Table 3. Intelligent Rinse modes used for each element to achieve a low LOQ for each element.

Mode	Elements
Quick	Al, Ba, Ca, Cd, Co, Cr, Cu, Mo, Na, Ni, Pb, Se, Sr, Tl, V
Moderate	As, B, K
Thorough	Fe, Mg, Mn, P, S, Zn

The rinse time for samples can vary due to the range of elemental concentrations in botanical supplements. Using Intelligent Rinse, the rinse times throughout the study were mostly between 15–75 s, averaging around 45 s per sample. Few samples required close to the full rinse time of 120 s.

Samples such as NIST 3280 Multivitamin/Multielement Tablets standard reference material (SRM) and the plant iron supplement contain relatively high concentrations of K, Mg, Fe, S, B, and Mn. These samples required longer rinsing between samples compared to samples like the juice berry concentrate or Ginkgo biloba, which required shorter rinse times before the elemental concentration thresholds were reached. Short rinse times were also used for the low concentration quality control (QC) and blank solutions, which were read periodically throughout the run.

The time saving benefits of Intelligent Rinse based on the rinse times of 275 samples using the 5900 ICP-OES are shown in Figure 1. The samples included dietary supplement digests, SRMs, spiked samples etc. The rinse time was 25% faster using Intelligent Rinse (orange line) compared to standard rinse conditions (blue line), corresponding to a time difference of over 60 minutes.
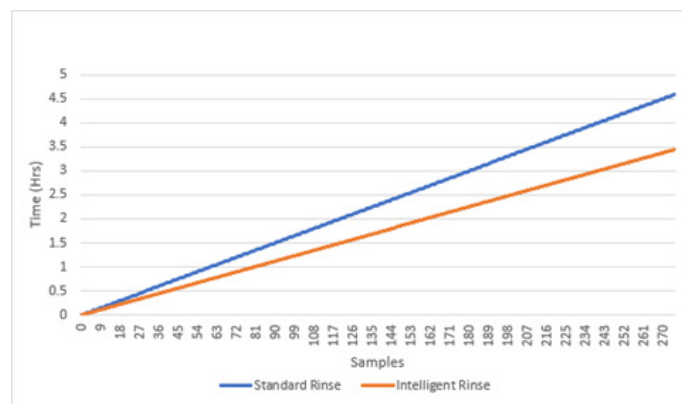

Figure 1. Rinse time for 275 samples using Intelligent Rinse (orange line) with an average of 45 s per sample (3.5 h) compared to the standard rinse with an average of 60 s per sample (4.6 h, blue line).

Method development using IntelliQuant Screening

Samples can be easily screened across a wide wavelength range in a few seconds using the IntelliQuant Screening function in the ICP Expert Pro software (*7, 8*). With 98% coverage of all elemental lines from 167 to 785 nm, the IntelliQuant Screening data provides valuable information for method development such as:

– Identifying the elemental composition of each sample including nutrients, micronutrients, and trace elements. The results can be displayed in a table, as a periodic table heat map, or in a pie chart.

– Obtaining semiquantitative concentration levels of analytes, which can be used to establish the calibration range for the quantitative method.

– Checking spectral interferences on analytes and obtaining interference-free wavelength recommendations for analytes, greatly assisting with wavelength-selection.

Figure 2 shows the semiquantitative data for Na in eight samples. Based on the IntelliQuant data, calibration standards for Na were prepared from 0.1 to 1000 ppm.

Rack:Tube	Label	Na mg/L
4:1	Tomato leaves	4.99420
4:2	Orchard leaves	2.55342
4:3	Apple leaves	1.81070
4:4	Sargasso	377.55430
4:5	Chlorella	4.07862
4:6	Spirulina	70.52695 o
4:7	Acai	11.82115
4:8	Mushroom-Chaga	5.69079

Figure 2. Semiquantitative data showing the concentration of Na in various samples and reference materials, informing the upper calibration concentration point for Na.

Background correction

A combination of Fitted Background Correction (FBC) and Fast Automated Curve-fitting Technique (FACT) were used in this method (*9*), as detailed in Table 5. FBC was selected for elements where there were no direct spectral overlaps, while FACT is most effective when the interferent partially overlaps the analyte peak.

IntelliQuant Screening identified a spectral interference on the V 292.401 line due to the proximity of the Fe peak (Figure 3), requiring the FACT background correction technique. FACT subtracts the contribution to the analyte signal from the interference and the blank, providing an accurate result for vanadium. FACT does not require knowledge of the precise concentration of interferent, which is beneficial for routine applications.
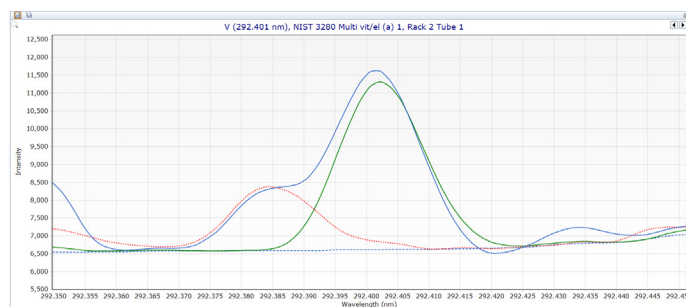

Figure 3. FACT corrects the Fe interfering peak to the left of V 292.401. Key: Raw spectral peak signal (blue solid line), blank (light blue dotted line), interferant peak (red dotted line), corrected analyte peak (green solid line).

Reference materials and samples

EAM 4.4 requires the use of at least one RM for method verification. Given the range of samples included in the study, two NIST SRMs were used: 3280 Multivitamin/Multielement Tablets and 1570a Trace Elements in Spinach Leaves (National Institute of Standards and Technology, Gaithersburg, US). To provide further verification of the method, Fortified Analytical Portions (FAPs) were prepared using a turmeric and a Ginkgo biloba (traditional herb) sample.

All samples, including spirulina, plant iron, juice berry concentrate, St John's wort, and a herbal stress adaptive, were bought online or in local stores in Melbourne, Australia. The samples were in tablet, capsule, or powder form.

Sample preparation

The powders were used as received, the capsules were emptied of their contents into a separate container, and the tablets were ground. Multiple capsules and tablets of each sample were processed and mixed for homogeneity. The SRMs were included to verify the digestion and analytical method.

The samples were digested in accordance with the procedure set out in EAM 4.4 using a CEM MARS 6 microwave digestion system. 0.5 g of each sample was weighed and transferred into a MARS Xpress vessel along with 1 mL of de-ionized water (DIW), 8 mL of HNO_3, and 1 mL of H_2O_2. If the acid reacted with the sample, the vessels were placed under a fume hood for around 20 minutes before capping. Duplicates of the samples, SRMs, and spiked samples (FAPs) were prepared and digested using the heating program shown in Table 4. (A higher temperature digestion was used to recover Al from the SRMs so not to bias low, as noted in EAM 4.4). After digestion and cooling, the solutions were transferred to 50 mL polypropylene centrifuge tubes and diluted to 25 mL with DIW.

Table 4. Microwave digestion program.

Parameter	Setting
Power (W)	1800
Ramp Time (min)	25
Hold Time (min)	15
Control Temperature (°C)	200

The analytical sequence of calibration standards, samples, and QC solutions specified in EAM 4.4 is shown in Figure 4. The sample block was analyzed repeatedly with automatic insertion of the periodic QC block after every 10 samples.
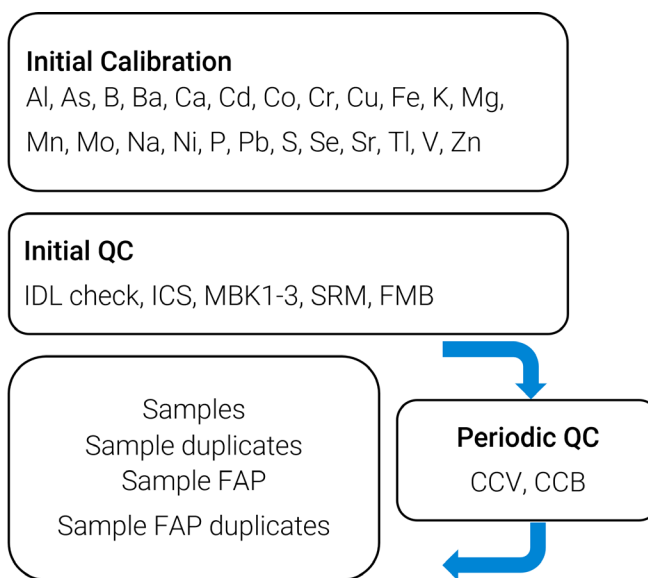

Initial Calibration
Al, As, B, Ba, Ca, Cd, Co, Cr, Cu, Fe, K, Mg, Mn, Mo, Na, Ni, P, Pb, S, Se, Sr, Tl, V, Zn

Initial QC
IDL check, ICS, MBK1-3, SRM, FMB

Samples
Sample duplicates
Sample FAP
Sample FAP duplicates

Periodic QC
CCV, CCB

**Figure 4.** Analytical sequence.
Key: Instrument detection limit (IDL), initial calibration verification (ICV), method blank (MBK), standard reference material (SRM), fortified method blanks (FMB), fortified analytical portion (FAP), continuing calibration verification (CCV), continuing calibration blank (CCB).

Calibration standards

The calibration standards (Table 5) were prepared to reflect the concentration of analytes in a range of typical samples, based on the semiquantitative data obtained by IntelliQuant Screening. The calibration standards and spiked samples were prepared from Agilent single element 10,000 ppm stock standards. The standards were diluted with DIW and HNO_3 to give final solutions at 16% HNO_3.

Calibration

Linear calibrations were obtained for all 24 elements analyzed using the 5900 ICP-OES, as indicated by the calibration coefficients reported in Table 5. Representative calibration curves for major to trace (Ca, Zn, Ni, and Pb) elements are shown in Figure 5.

Figure 5. Representative calibration curves of a major (Ca), a minor (Zn), and two trace elements (Ni and Pb).
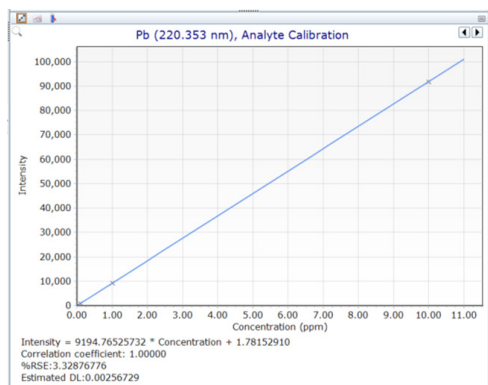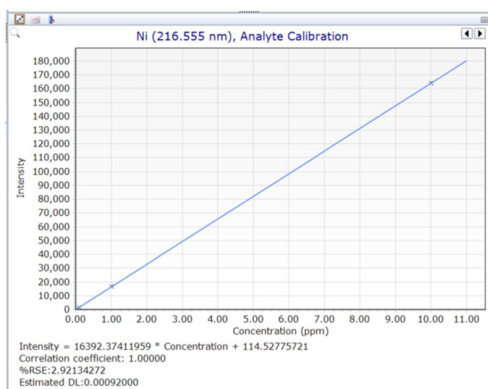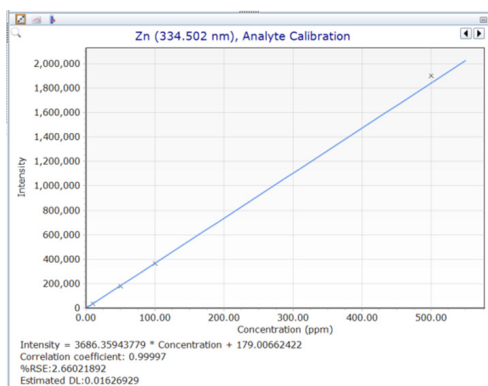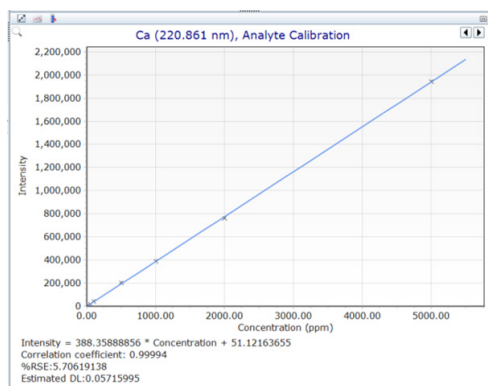
MultiCal

Axial viewing of the plasma increases detectability and precision of low concentration elements, but high-intensity wavelengths, which are normally measured radially, can overrange the detector. However, the Vista Chip III detector featured in the 5900 provides wide wavelength coverage enabling multiple suitable wavelengths to be used for an element. As different wavelengths often have different sensitivities, multiple wavelengths can be used for the same element. The MultiCal feature of the ICP Expert software combines the wavelengths of the element, which is an effective way to extend the linear dynamic range (LDR) in axial view mode. The data obtained from the multiple wavelengths can be automatically reported by the software as a single result.

In this study, the varying ranges of the concentration of Na in different samples (Figure 2) were resolved by calibrating Na 589.592 and Na 568.821 nm lines simultaneously. The IntelliQuant data also showed that multiple calibration ranges were also needed to achieve the upper limit of LDR for K and Zn. MultiCal was used to combine calibration ranges for Na, K, and Zn in axial mode, using two wavelengths per element, as shown in Table 5.

Table 5. Wavelengths, background correction method, and working calibration range of each element.

Element and Wavelength (nm)	Background Correction	Internal Standard	Calibration Range (mg/L)	Correlation Coefficient
Al 394.401	Fitted	None	0.0 to 10	0.99999
As 188.980	Fitted	Bi 223.061	0.0 to 10	1.00000
B 249.678	FACT	Bi 223.061	0.0 to 10	1.00000
Ba 455.403	Fitted	Y 371.029	0.0 to 10	1.00000
Ca 220.861	Fitted	Y 371.029	0.0 to 5000	0.99994
Cd 228.802	FACT	Bi 223.061	0.0 to 10	0.99999
Co 228.615	Fitted	Y 371.029	0.0 to 10	1.00000
Cr 206.550	Fitted	Y 371.029	0.0 to 10	1.00000
Cu 324.754	Fitted	Bi 223.061	0.0 to 50	0.99992
Fe 261.187	Fitted	Bi 190.171	0.0 to 500	0.99983
K 766.491	Fitted	Rb 780.026	0.0 to 50	0.99996
K 404.721	Fitted	Rb 780.026	50 to 2000	0.99981
Mg 279.800	Fitted	Y 371.029	0.0 to 2000	0.99915
Mn 257.610	Fitted	Y 371.029	0.0 to 50	0.99954
Mo 202.032	Fitted	Y 371.029	0.0 to 10	1.00000
Na 589.592	FACT	Li 670.783	0.0 to 10	1.00000
Na 568.821	Fitted	Li 670.783	10 to 1000	0.99985
Ni 216.555	Fitted	Y 371.029	0.0 to 10	1.00000
P 213.618	FACT	Bi 223.061	0.0 to 2000	0.99977
Pb 220.353	FACT	Y 371.029	0.0 to 10	1.00000
S 181.972	Fitted	Bi 223.061	0.0 to 2000	0.99972
Se 196.026	FACT	Bi 223.061	0.0 to 10	1.00000
Sr 216.596	Fitted	Y 371.029	0.0 to 10	1.00000
Tl 190.794	Fitted	Y 371.029	0.0 to 10	1.00000
V 292.401	FACT	Y 371.029	0.0 to 10	1.00000
Zn 213. 857	Fitted	Bi 223.061	0.0 to 50	0.99954
Zn 334.502	Fitted	Y 371.029	50 to 500	0.99997

Results and discussion

Analytical limits

Limit of detection (LOD) and limit of quantification (LOQ) are important indicators of the performance of the analytical method. The LOD represents the lowest concentration that can be detected in a sample, while the LOQ indicates the lowest concentration that can be reliably quantified with an acceptable degree of tolerance. The LODs and LOQs were calculated based on 10 reads of a spiked method blank that was prepared independently by spiking a blank solution with analyte concentrations, as recommended in EAM 3.2 (10). The limits reported are an average from three determinations performed over non-consecutive days.

The LOD is the standard deviation (SD or sigma) of 10 replicate measurements multiplied by 3, with the dilution factor applied. LOQ is 10 sigma times the dilution factor based on 0.5 g of sample in 25 mL.

To provide confidence that the method will meet regulatory requirements, EAM 4.4 provides nominal analytical limits for 23 elements. The 5900 ICP-OES met the requirements outlined in EAM 4.4 (Table 6), demonstrating the suitability of the instrument for the analysis.

Table 6. Agilent 5900 ICP-OES and EAM 4.4 nominal analytical limits.

Element and Wavelength (nm)	Calculated based on EAM 4.4 Analytical Limits		EAM 4.4 Nominal Analytical Limits	
	LOD (mg/kg)	LOQ (mg/kg)	LOD (mg/kg)	LOQ (mg/kg)
Al 394.401	0.15	0.50	0.8	2
As 188.980	0.18	0.6	2	4
B 249.678	0.22	0.72	0.3	0.8
Ba 455.403	0.0028	0.0095	0.05	0.2
Ca 220.861	2.2	7.3	8	30
Cd 228.802	0.024	0.081	0.3	0.9
Co 228.615	0.05	0.17	0.3	0.8
Cr 206.550	0.054	0.18	2	5
Cu 324.754	0.026	0.088	0.1	0.3
Fe 261.187	0.066	0.22	0.2	0.3
K 766.491	0.27	0.89	20	40
Mg 279.800	0.13	0.42	2	6
Mn 257.610	0.003	0.01	0.2	0.4
Mo 202.032	0.032	0.11	0.4	1
Na 589.592	0.13	0.42	2	5
Ni 216.555	0.041	0.14	0.9	3
P 213.618	0.13	0.42	2	6
Pb 220.353	0.11	0.36	3	6
S 181.972	0.55	1.8	-	-
Se 196.026	0.27	0.89	3	9
Sr 216.596	0.021	0.07	0.03	0.07
Tl 190.794	0.17	0.57	2	6
V 292.401	0.054	0.18	0.2	0.5
Zn 213. 857	0.018	0.061	0.3	0.8

Method verification

EAM 4.4 includes a set of QC measures such as the analysis of RMs and spiked samples that analysts should use to verify the method. EAM 4.4 also requires the periodic measurement of QC standards within specified precision limits.

Reference materials

To confirm the accuracy of the sample preparation and quantitative method, the multivitamin and spinach leaves SRMs were analyzed in triplicate using the 5900 ICP-OES. The averaged recoveries for most analytes measured above the LOQ were within ±10% of the certified value (Table 7), meeting the ±20% acceptance criteria specified in the EAM 4.4 method.

The 5900 was able to detect the low concentration of boron in both SRMs. Boron can often present problems with memory effects and cause carryover between samples due to its tendency to stick to the tubes and sample introduction system. To achieve a good recovery for boron when using a clean (or new) sample introduction system, the sample introduction system needs to be conditioned with a high concentration boron rinse solution for 30 minutes. A boron-free rinsing solution should then be aspirated through the instrument for another 30 minutes before analysis.

Due to its tendency to be present in complex molecules, the aluminum recovery was achieved using a higher temperature digestion that may not be suitable for all elements covered in this application.

Table 7. Recovery of elements measured in NIST 3280 Multivitamin/Multielement tablets and NIST 1570a Trace Elements in Spinach Leaves. Blank cells indicate the absence of a certified value.

Element and Wavelength (nm)	NIST 3280 Multivitamin/Multielement Tablets			NIST 1570a Trace Elements in Spinach Leaves		
	Certified (mg/kg)	Measured (mg/kg)	Recovery (%)	Certified (mg/kg)	Measured (mg/kg)	Recovery (%)
Al 394.401				310	343	109
As 188.980	0.132	<LOQ	**	0.068	<LOQ	**
B 249.678	141	142	101	37.7	40.7	108
Ba 455.403						
Ca 220.861	110700	108000	97	15260	16400	107
Cd 228.802	0.08015	<LOQ	**	2.876	2.92	101
Co 228.615				0.393	0.425	108
Cr 206.550	93.7	91.5	98			
Cu 324.754	1400	1383	99	12.22	13.1	107
Fe 261.187	12350	12100	98			
K*	53100	50100	94	29000	28800	99
Mg 279.800	67800	67800	100			
Mn 257.610	1440	1320	91	76	78	103
Mo 202.032	70.7	73.3	104			
Na*				18210	18300	101
Ni 216.555	8.43	8.18	97	2.142	2.05	96
P 213.618	75700	69500	92	5187	5710	87
Pb 220.353	0.2727	<LOQ	**			
S 181.972						
Se 196.026	17.42	18	103	0.1152	<LOQ	**
Sr 216.596				55.54	53.7	97
Tl 190.794						
V 292.401				0.568	0.595	105
Zn*	10150	10100	99	82.3	86.6	105

*Combination of wavelengths used for MultiCal calibration. **Recoveries not calculated as the certified values are close to or below the method LOQ.

Spiked method blank

Two fortified method blanks (FMBs) were prepared by spiking the blank at 50 ppb for most elements and 100 ppb for major elements before digestion. The FMBs were analyzed during the analytical run. All recoveries were within ±10%, as shown in Table 8.

Spike recoveries

To further confirm the performance of the 5900 ICP-OES method, including for elements not certified in the two SRMs, two FAPs were prepared. Elements were spiked in the turmeric and Ginkgo biloba samples at concentrations around their unfortified levels. In accordance with the EAM 4.4 method, any analytes over 100 ppm in either sample were not spiked.

Most elements were present in turmeric at low levels apart from Mg, which was present at 650 mg/L. Since the native concentration matrix of the Ginkgo biloba sample had slightly less variation, spike concentrations between 0.05 and 40 mg/L were used, providing an extra level of verification of the method. The recoveries for all elements in the FAPs were within ±15%, as shown in Table 8, which is well within the EAM 4.4 method QC criteria of ±20%.

The FAP test demonstrates the instrument's ability to accurately measure analytes at analytically relevant concentrations in the sample matrix.

Table 8. FMB and FAP spike recovery data. All elements were spiked before digestion.

Element and Wavelength (nm)	Fortified Blank		Turmeric			Ginkgo Biloba		
	Spike (mg/L)	Recovery (%)	Quantitative Results (mg/L)	Spike (mg/L)	Spike Recovery (%)	Quantitative Results (mg/L)	Spike (mg/L)	Spike Recovery (%)
Al 394.401	0.05	95	0.11	0.5	105	0.18	0.15	98
As 188.980	0.05	94	<LOQ	0.5	97	<LOQ	0.1	93
B 249.678	0.05	96	0.023	0.5	98	0.09	0.20	96
Ba 455.403	0.05	95	0.0066	0.5	100	0.0017	0.05	97
Ca 220.861	0.1	108	0.67	0.5	95	270	40	107
Cd 228.802	0.05	99	<LOQ	0.5	100	<LOQ	0.05	95
Co 228.615	0.05	98	<LOQ	0.5	94	<LOQ	0.05	94
Cr 206.550	0.05	97	0.014	0.5	98	0.015	0.05	96
Cu 324.754	0.05	102	0.0064	0.5	102	0.0087	0.05	97
Fe 261.187	0.05	98	0.18	0.5	99	0.26	0.4	100
K*	0.1	90	0.57	10	102	0.13	1.0	94
Mg 279.800	0.05	95	650	-	-	7	10	101
Mn 257.610	0.05	96	0.026	0.5	97	0.011	0.1	95
Mo 202.032	0.05	97	<LOQ	0.5	99	<LOQ	0.1	92
Na*	0.1	102	1.0	5.0	112	3.0	4	92
Ni 216.555	0.05	96	<LOQ	0.5	92	0.0062	0.05	92
P 213.618	0.05	100	0.026	5.0	107	210	40	103
Pb 220.353	0.05	97	<LOQ	0.5	93	<LOQ	0.1	88
S 181.972	0.1	110	0.65	0.5	110	10	10	110
Se 196.026	0.05	94	<LOQ	0.5	99	<LOQ	0.25	93
Sr 216.596	0.05	97	0.0045	0.5	98	0.052	0.20	90
Tl 190.794	0.1	89	<LOQ	0.5	96	<LOQ	0.20	89
V 292.401	0.05	97	<LOQ	0.5	98	<LOQ	0.1	92
Zn*	0.05	105	0.026	0.5	103	0.0075	0.1	93

Combination of wavelengths used for MultiCal calibration.

Long-term stability

To assess the stability of the 5900, samples including supplement digests, SRMs, FAPs etc. were measured over seven hours without the need for recalibration. A 10 ppm QC solution was measured after every 10 samples during the run.

The QC stability plot in Figure 6 shows the recoveries of all elements over seven hours to be within ±10%. The results demonstrate the excellent robustness and precision of the 5900 SVDV ICP-OES for the routine QC analysis of dietary supplement samples.
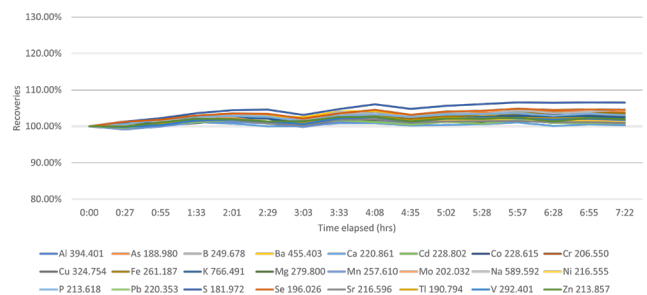

Figure 6. Long-term stability showing recovery of a 10 ppm QC solution analyzed every 10 samples over 7 h.

Quantitative results

The quantitative results of five dietary supplement samples, all with differing compositions, show the range of samples that can be analyzed using the 5900 ICP-OES method (Table 9).

Spirulina is algae that grow naturally or is farmed in mineral-rich waters. The variable growing environment of spirulina will affect its elemental composition. However, all elements measured above the LOQ showed favorable replicability, with relative standard deviations (RSDs) and relative percent differences (RPDs) within the respective EAM 4.4 stipulated limits of 7 and 20%. The RPD results are used to check the reproducibility of the method by measuring the difference between two different readings of a sample relative to their concentrations.

The reported label value of Fe in the plant iron supplement was 12 mg per 0.5 g capsule. The 5900 ICP-OES measured Fe concentration in the sample was 23,900 mg/kg, which represents a 99% recovery of the label value of 24000 mg/kg.

Table 9. Agilent 5900 ICP-OES average quantitative data for five commercially available dietary supplements and RPDs of two measurements.

Element and Wavelength (nm)	Spirulina		Plant Iron		Juice Berry Concentrate		St John's Wort		Herbal Stress Adaptive	
	Quantitative Result (mg/kg)	RPD (%)	Quantitative Result (mg/kg)	RPD (%)	Quantitative Result (mg/kg)	RPD (%)	Quantitative Result (mg/kg)	RPD (%)	Quantitative Result (mg/kg)	RPD (%)
Al 394.401	314	1.	62.4	1.1	38.3	1.1	58.0	0.2	17.4	0.1
As 188.980	0.725	3.4	<LOQ		<LOQ		<LOQ		<LOQ	
B 249.678	6.92	1.6	159	0.1	17.6	0.2	11.4	1.2	8.63	0.0
Ba 455.403	14.9	1.5	65.5	0.9	3.91	1.2	0.327	0.8	5.59	1.2
Ca 220.861	2110	1.8	13800	1.3	3650	1.3	46100	0.8	1910	1.1
Cd 228.802	0.273	5.5	<LOQ		0.146	6.5	<LOQ		<LOQ	
Co 228.615	0.41	8.5	1.52	1.8	0.42	7.3	0.37	1.1	0.30	1.8
Cr 206.550	1.35	0.3	37.1	0.6	1.24	1.1	0.74	0.1	2.53	2.0
Cu 324.754	1.76	1.5	2.51	3.9	12.5	0.8	5.78	0.6	4.18	1.0
Fe 261.187	803	0.3	23900	0.1	76.3	0.5	2620	0.2	62.2	0.3
K*	14500	0.7	40400	0.2	16800	0.3	12200	0.3	14700	0.3
Mg 279.800	3770	1.9	6540	1.2	2070	0.7	1030	1.1	1590	1.0
Mn 257.610	28.1	1.8	199	0.7	39.8	0.9	10.3	0.8	24.6	1.1
Mo 202.032	<LOQ		1.39	1.2	0.17	26.4	0.23	10.6	0.12	2.9
Na*	2430	1.1	5890	0.6	716	1.8	2480	0.9	966	0.0
Ni 216.555	1.14	5.6	21	0.8	3.42	1.8	1.59	0.8	2.21	0.7
P 213.618	10600	0.1	1060	0.4	2710	0.0	34700	0.4	1760	0.6
Pb 220.353	0.42	3.6	0.65	6.2	<LOQ		<LOQ		<LOQ	
S 181.972	7630	1.5	26900	0.4	1480	0.1	529	0.4	1990	0.8
Se 196.026	<LOQ		<LOQ		<LOQ		<LOQ		<LOQ	
Sr 216.596	18.9	1.8	84.7	0.9	6.9	1.2	11.4	1.1	6.36	1.4
Tl 190.794	<LOQ		<LOQ		<LOQ		<LOQ		<LOQ	
V 292.401	0.30	6.1	1.28	1.2	<LOQ		0.39	4.1	<LOQ	
Zn*	15.1	1.7	32.2	1.5	19.3	1.3	15.2	1.0	11.4	1.1

*Combination of wavelengths used for MultiCal calibration.

Conclusion

The study demonstrates the effectiveness of the Agilent 5900 SVDV ICP-OES for the routine analysis of nutrient and other elements in dietary supplements following EAM method 4.4. In this study we used one of the four available viewing modes, axial, to measure all elements, meeting the requirements of the EAM 4.4 method. A range of samples bought as powders, capsules, or tablets were prepared by microwave digestion before analysis by ICP-OES. Smart tools included within the ICP Expert Pro software were used throughout the workflow, providing valuable information on samples and instrument performance.

– IntelliQuant Screening was used to identify or confirm interference-free analyte wavelengths for the quantitative measurement of 24 elements in various dietary supplement samples. The semiquantitative data was used to establish the calibration range for all 24 elements.

– FBC and FACT background correction techniques within the ICP Expert software successfully corrected for background structures and spectral interferences arising from the various sample matrices.

– The MultiCal function within the ICP Expert software was used to combine wavelengths for K, Na, and Zn—a quick and effective way to extend the linear dynamic range in axial mode.

– Sample throughput was maximized using Intelligent Rinse software. Intelligent Rinse automatically optimizes the rinse time between samples, saving time and resources.

The 5900 ICP-OES method was verified in accordance with EAM 4.4 criteria. The LOQs were well below the requirements specified in EAM 4.4. Excellent recoveries were obtained for all certified elements in the NIST 3280 Multivitamin/multielement, NIST 1570a Spinach SRMs, and in two fortified samples, confirming the accuracy of the method. The long-term stability test demonstrated the robustness of the 5900 ICP-OES for the analysis of dietary supplements.

The 5900 SVDV ICP-OES provides developers, manufacturers, and distributors of dietary supplements with a routine and accurate method that meets current testing requirements with added flexibility if requirements change. Intelligent software features equip the analyst with tools to develop an accurate quantitative method for the routine measurement of multiple elements in dietary samples.

References

1. US Food and Drug Administration, Dietary Supplement Labeling Guide: Chapter I. General Dietary Supplement Labeling, April 2005, accessed February 2023, https://www.fda.gov/food/dietary-supplements-guidance-documents-regulatory-information/dietary-supplement-labeling-guide

2. US Food and Drug Administration, Dietary Supplements, June 2022, accessed February 2023, https://www.fda.gov/food/dietary-supplements

3. Grand View Research, Dietary Supplements Market Size Report, 2022–2030, March 2022, accessed February 2023, https://www.grandviewresearch.com/industry-analysis/dietary-supplements-market

4. Mindak, W.R., Dolan S.P., U.S. Food and Drug Administration Elemental Analysis Manual, 4.4 Inductively Coupled Plasma-Atomic Emission Spectrometric Determination of Elements in Food Using Microwave Assisted Digestion, Version 1.1 (FDA, Rockville, MD, 2010), accessed February 2023, https://www.fda.gov/media/95162/download

5. Synchronous Vertical Dual View (SVDV) for High Productivity and Low Cost of Ownership, Agilent 5900 SVDV ICP-OES, Agilent publication, 5994-1513EN

6. Intelligent Rinse for ICP-OES, Increase productivity and reduce errors, Agilent publication, 5991-8456EN

7. Agilent IntelliQuant Screening, Smarter and quicker semiquantitative ICP-OES analysis, Agilent publication, 5994-1518EN

8. Agilent ICP Expert Software: Powerful software with smart tools for ICP-OES, Agilent publication, 5994-1517EN

9. ICP-OES Background and Interference Removal Accurate. Easy. Reliable, Agilent publication, 5991-8452EN

10. US Food and Drug Administration, Elemental Analysis Manual - Section 3.2 Version 3.0 (December 2021), accessed February 2023, https://www.fda.gov/media/89337/download

Consumables

Part Number	Description
G8010-60228	Easy-fit torch one piece for 5000 Series vertical dual view ICP-OES
G8010-60256	Double-pass spray chamber, glass cyclonic design with ball joint socket and UniFit drain outlet, for Agilent 5000 series ICP-OES
G8010-60255	SeaSpray concentric glass nebulizer for Agilent 5000 series ICP-OES
3710034400	Peristaltic pump tubing, PVC, white/white, 12/pk
3710027200	Peristaltic pump tubing, PVC, black/black, 12/pk
3710034600	Peristaltic pump tubing, PVC, blue/blue, 12/pk
G8494-60007	Reagent uptake capillary tube 1.6 mm OD x 1.0 mm id x 1400 mm long with heavy PTFE sinker, 1/pk
2410020500	Nebulizer capillary tubing, per m.
G8010-80036	UniFit drain connector, 2.0 mm od x 0.86 mm id, 3/pk
3710024600	Drain tubing for spray chamber waste, 0.12" id x 0.062" thick, per m.
G8010-40026	Clip to secure the drain tubing
5190-8352	Agilent 10,000 ppm single element stock solution for Al
5190-8356	Agilent 10,000 ppm single element stock solution for As
5190-8364	Agilent 10,000 ppm single element stock solution for B
5190-8358	Agilent 10,000 ppm single element stock solution for Ba
5190-8368	Agilent 10,000 ppm single element stock solution for Ca
5190-8366	Agilent 10,000 ppm single element stock solution for Cd
5190-8376	Agilent 10,000 ppm single element stock solution for Co
5190-8374	Agilent 10,000 ppm single element stock solution for Cr
5190-8378	Agilent 10,000 ppm single element stock solution for Cu
5190-8402	Agilent 10,000 ppm single element stock solution for Fe
5190-8432	Agilent 10,000 ppm single element stock solution for K
5190-8412	Agilent 10,000 ppm single element stock solution for Mg
5190-8414	Agilent 10,000 ppm single element stock solution for Mn
5190-8418	Agilent 10,000 ppm single element stock solution for Mo
5190-8454	Agilent 10,000 ppm single element stock solution for Na
5190-8422	Agilent 10,000 ppm single element stock solution for Ni
5190-8428	Agilent 10,000 ppm single element stock solution for P
5190-8406	Agilent 10,000 ppm single element stock solution for Pb
5190-8209	Agilent 10,000 ppm single element stock solution for S
5190-8448	Agilent 10,000 ppm single element stock solution for Se
5190-8207	Agilent 10,000 ppm single element stock solution for Sr
5190-8217	Agilent 10,000 ppm single element stock solution for Tl
5190-8228	Agilent 10,000 ppm single element stock solution for V
5190-8234	Agilent 10,000 ppm single element stock solution for Zn
5190-8362	Agilent 10,000 ppm single element stock solution for Bi
5190-8408	Agilent 10,000 ppm single element stock solution for Li
5190-8440	Agilent 10,000 ppm single element stock solution for Rb
5190-8232	Agilent 10,000 ppm single element stock solution for Y

www.agilent.com/chem/5900icp-oes