The accurate measurement of selenium in twelve diverse reference materials using on-line isotope dilution with the 8800 Triple Quadrupole ICP-MS in MS/MS mode

Application note

Environmental

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Introduction

Selenium (Se) is an important element in environmental and agricultural studies and in human health, as it is an essential trace nutrient but is also toxic in excess. The role of certain chemical forms of Se is also the subject of significant research into cancer prevention. ICP-MS is the analytical method of choice for both total and speciated Se measurements, but Se is difficult to quantify accurately at trace levels by ICP-MS for several reasons:

- Se has a high first ionization potential (IP = 9.75 eV), which means it is poorly ionized in the plasma and therefore its signal intensity is low.

- Because Se is poorly ionized, it suffers signal suppression in high matrix samples, an issue that is further compounded by the lack of a suitable internal standard element with similar mass and IP.

- All the analytically useful Se isotopes suffer from multiple spectral interferences in real world samples, as summarized in Table 1.

- The resolution required to separate all the spectral interferences is beyond the capabilities of sector-type high resolution ICP-MS.

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Table 1. Spectral interferences on major Se isotopes

| Se isotope | | | | | Interference | | | |
m/z	Abundance %	Isobaric	Argide	Oxide	Hydride	Chloride	Doubly-charged	Dimer
77	7.63		$^{39}K^{38}Ar^+$	$^{61}Ni^{16}O^+$, $^{59}Co^{18}O^+$	$^{76}GeH^+$, $^{76}SeH^+$	$^{40}Ar^{37}Cl^+$, $^{40}Ca^{37}Cl^+$	$^{154}Sm^{++}$, $^{154}Gd^{++}$	
78	23.77	$^{78}Kr^+$	$^{40}Ca^{38}Ar^+$	$^{62}Ni^{16}O^+$	$^{77}SeH^+$	$^{41}K^{37}Cl^+$	$^{156}Gd^{++}$, $^{156}Dy^{++}$	$^{38}Ar^{40}Ar^+$, $^{39}K^{39}K^+$
80	49.61	$^{80}Kr^+$	$^{40}Ca^{40}Ar^+$	$^{64}Ni^{16}O^+$, $^{64}Zn^{16}O^+$, $^{32}S_2^{16}O^+$, $^{32}S^{16}O_3^+$	$^{79}BrH^+$	$^{45}Sc^{35}Cl^+$	$^{160}Gd^{++}$, $^{160}Gd^{++}$	$^{40}Ar^{40}Ar^+$, $^{40}Ca^{40}Ca^+$
82	8.73	$^{82}Kr^+$	$^{42}Ca^{40}Ar^+$	$^{66}Zn^{16}O^+$, $^{32}S^{34}S^{16}O^+$, $^{34}S^{16}O_3^+$	$^{81}BrH^+$	$^{45}Sc^{37}Cl^+$	$^{162}Dy^{++}$, $^{162}Er^{++}$	

While quadrupole ICP-MS with collision/reaction cell is used routinely to measure Se down to the levels that are relevant for environmental legislation, ultratrace (sub-ppb) quantification of Se in high and variable sample matrices remains challenging for the reasons mentioned above.

This application note describes how a new instrument, the Agilent 8800 Triple Quadrupole ICP-MS, has solved the problem of ultratrace Se measurement in variable high matrix samples. The new 8800 ICP-MS Triple Quad (commonly abbreviated to ICP-QQQ) features an additional quadrupole mass filter (Q1), situated in front of a conventional collision/reaction cell (CRC) and quadrupole mass filter (Q2) to give an MS/MS (also called tandem MS) configuration. In ICP-MS/MS, Q1 operates as a mass filter, allowing only the target analyte mass to enter the cell, and rejecting all other masses. This means that ions from the plasma and sample matrix are prevented from entering the cell, so the cell conditions remain consistent even if the sample matrix varies. This improves interference removal efficiency in collision mode (using He cell gas) compared to conventional quadrupole ICP-MS (ICP-QMS).

The greatest benefit of MS/MS, however, is the dramatic improvement of performance in reaction mode. Inside the cell, the analyte and any on-mass interferences are separated by their different reaction rates with an appropriate reactive gas. Because plasma and matrix ions are eliminated by Q1, the reaction efficiency is greatly enhanced and the reaction pathways and product ions remain consistent across a wide range of sample matrices. This unique aspect of MS/MS greatly simplifies method development and routine operation in reaction mode, because the same cell conditions give effective interference removal for multiple sample types.

As with established reaction mode methods in conventional ICP-QMS, a range of cell gases may be used, depending on the analyte being measured. However, the unique capability of MS/MS and what makes the 8800 ICP-MS so powerful is that the chosen reaction gas gives reliable interference removal for each target analyte *regardless of sample matrix*, because in MS/MS mode, the sample matrix ions are rejected by Q1.

Depending on the analyte and reaction mode used, the second quadrupole (Q2), is set to measure either the analyte ion at its original mass (on-mass measurement), or a cell-formed product ion (formed by reaction of the analyte ion with the cell gas) at a different mass. In the case where the analyte ion is more reactive than its interferences, separation of the overlapping ion is achieved by reacting the analyte away from the interference and measuring the analyte at a new product ion mass that is free from overlap. This alternate approach is referred to as indirect or mass-shift measurement.

This application note compares the effectiveness of different reaction gases and measurement strategies for multiple Se isotopes to highlight the capabilities of the 8800 ICP-MS. However, in normal routine use, standardized cell conditions are used for the same analyte across a wide variety of sample types, and are predefined in turnkey, preset methods.

While conventional ICP-QMS reaction mode methods are typically applicable to a single, preferred isotope for each analyte, the consistent and reliable removal of multiple matrix interferences by MS/MS on the 8800 can make multiple isotopes available for measurement. This allows the use of isotope dilution (ID) analysis, which is the most accurate quantification technique

for ultratrace measurement in high and variable matrix samples. On-line isotope dilution analysis (OIDA) [1] is a very powerful and useful development of traditional isotope dilution, since it removes the time consuming step of spiking enriched-isotope standards into each separate sample. To demonstrate the power of the 8800 in MS/MS mode for accurate, ultratrace Se measurement in a range of different sample types, OIDA was used to quantify Se in a range of certified reference materials (CRMs).

Experimental

Instrumentation
An Agilent 8800 ICP-QQQ equipped with a standard sample introduction system was used for the study. The standard sample introduction system consists of a glass concentric nebulizer, a quartz double-pass Scott-type spray chamber, and a quartz torch with 2.5 mm id injector.

The configuration of the Agilent 8800 instrument is similar to the Agilent 7700 Series ICP-QMS, except for the addition of a quadrupole (Q1) in front of the 3rd generation Octopole Reaction System (ORS[3]) cell. Q1 is typically operated as a mass filter (MS/MS mode) ensuring that all non-target ion masses are rejected prior to the cell. In the cell gas studies described in this note, both on-mass measurement (where the analyte is measured directly at its original mass) and mass-shift mode (where the analyte is reacted away from the interference and measured indirectly as a cell-formed product ion) were evaluated. Although the use of indirect (mass-shift) measurement is not unique to the 8800, the product ion spectra generated by MS/MS are far simpler than those generated by traditional ICP-QMS, because MS/MS mode ensures that any co-existing ions that might overlap the new product ion mass are rejected by Q1. This means that indirect measurement is much more accurate and more widely applicable and, as with MS/MS direct on-mass measurement, little or no method development is required when an existing mode is applied to a new sample type.

Cell gas and measurement mode investigation
As stated above, with MS/MS the reaction gas and measurement mode remains consistent for a given analyte, regardless of sample type. However, for the purpose of demonstrating the interference removal power of the 8800 ICP-MS, a range of different gases and gas mixes was used, and both direct and indirect measurement modes were investigated for Se measurement in four different synthetic sample matrices. Data was acquired for three Se isotopes, 78, 80 and 82, since isotope dilution requires two isotopes free from interference. Instrument operating parameters are shown in Table 2. Cell gases evaluated were He (which is inert, and therefore a collision gas), and three reaction gases/gas mixes H_2, O_2/H_2 and *NH_3/H_2.

Table 2. Agilent 8800 ICP-MS operating parameters

Parameter	Value			
RF power (W)	1550			
Sampling depth (mm)	8			
Carrier gas flow rate (L/min)	1.05			
Operating mode	MS/MS			
KED (V)	5	-6		
Octopole bias (V)	-95	-18		
Cell gas	He	H_2	O_2/H_2	NH_3/H_2
Cell gas flow rate (mL/min)	10	6	O_2 = 0.4 H_2 = 2.0	*NH_3 = 6.0 H_2 = 2.0

*10% NH_3 in He

The 8800 ICP-MS was operated in MS/MS mode in all cases, where Q1 functions as a unit mass filter, allowing the Se isotopes at mass to charge (m/z) 78, 80 and 82 to pass sequentially to the cell. For He, H_2 and NH_3/H_2 cell gas modes, the Se isotopes were measured directly, on-mass at m/z 78, 80 and 82 (Q2 set to the same mass as Q1). On-mass interferences entering the cell were removed by collision and kinetic energy discrimination (KED) in the case of He, or by reaction in the other gas modes.

To investigate the effectiveness of mass-shift measurement for Se, O_2/H_2 cell gas was also used. Se^+ reacts reasonably quickly with O_2 to form SeO^+, allowing the Se 78, 80 and 82 isotopes to be measured indirectly as SeO^+ at m/z 94, 96 and 98 respectively. Shifting

the Se^+ to SeO^+ moves it away from the on-mass interferences, which do not react with the cell gas and so remain at their original masses, and are subsequently rejected by Q2. O_2 cell gas alone can be used for Se measurement at levels down to 10s of ppt, but the use of a mixed O_2/H_2 cell gas permits even lower levels of Se to be measured — detection limits of single-ppt can be achieved reliably. H_2 and O_2 were added to the ORS[3] separately using independent mass flow controllers. The 8800 ICP-MS features four separate cell gas lines, so gases can be mixed in the cell, reducing the need for expensive premixed gas blends.

Figure 1 illustrates the indirect method using the measurement of ^{80}Se as an example. Q1 is set to m/z 80, rejecting all the ions at masses other than m/z 80. Only the analyte ion $^{80}Se^+$, plus any interfering ions at the same m/z (for example, $^{40}Ar^{40}Ar^+$, $^{160}Gd^{++}$ and $^{160}Dy^{++}$) are allowed to pass through to the cell. Q2 is set to measure the analyte reaction product ion ($^{80}Se^{16}O^+$) at m/z 96. The original interfering ions $ArAr^+$, Gd^{++} and Dy^{++} do not react with O_2, so these interferences remain at mass 80 and so are rejected by Q2. The SeO^+ product ion at m/z 96 would be interfered by isobaric overlaps from $^{96}Zr^+$, $^{96}Mo^+$ and $^{96}Ru^+$ when measured using ICP-QMS, but these isotopes are all rejected by Q1 on the 8800 ICP-MS and so the SeO^+ product ion can be measured at its new mass, completely free from any interference. This is the unique benefit of MS/MS and what makes it much more accurate and powerful than using reaction mode with ICP-QMS.

The sample matrices evaluated in the cell gas mode test were:

- 1% HNO_3
- 500 ppb Br
- A mix containing 1 ppm of each rare earth element (REE)
- 200 ppm Ca in 0.5% HCl

The Br, REE and Ca matrices were chosen since these elements all give rise to severe interferences on Se, as shown in Table 1. Background equivalent concentrations (BECs) for the Se isotopes 78, 80 and 82 were measured in each gas mode, and the results are shown in Figure 2.

In the simplest matrix, 1% HNO_3, all gas modes eliminated the interferences on ^{78}Se, but not the other two Se isotopes. Since ID requires two interference-free masses, He mode could not be used, even in 1% HNO_3. The other gas modes all worked well for at least two Se isotopes in HNO_3. When 500 ppb Br was present, only NH_3/H_2 and O_2/H_2 worked well for all isotopes. With 200 pm Ca in 1% HCl, the situation was the same — only NH_3/H_2 and O_2/H_2 worked well for all Se isotopes. When the 1 ppm mixed REE matrix was tested however, only O_2/H_2 could remove the interferences on all the Se isotopes. It can be seen therefore that using O_2/H_2 cell gas and indirect measurement, Se can be measured at ultratrace levels in any of the test sample types, giving a choice of three interference-free isotopes. Even doubly-charged REE interferences on Se, which cannot be removed effectively by any other ICP-MS technique, are
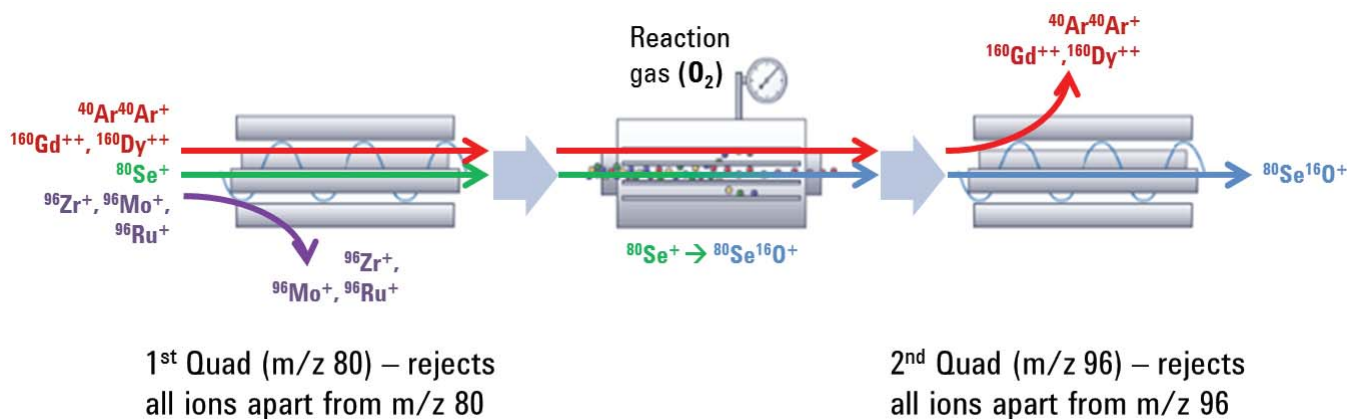

Figure 1. Illustration of indirect measurement of Se using mass-shift with O_2/H_2 reaction gas. $^{80}Se^+$ is reacted away from the interference and measured as SeO^+ at m/z 96. Zr, Mo and Ru isotopes at m/z 96 do not interfere with SeO^+ measurement since they are rejected by Q1.
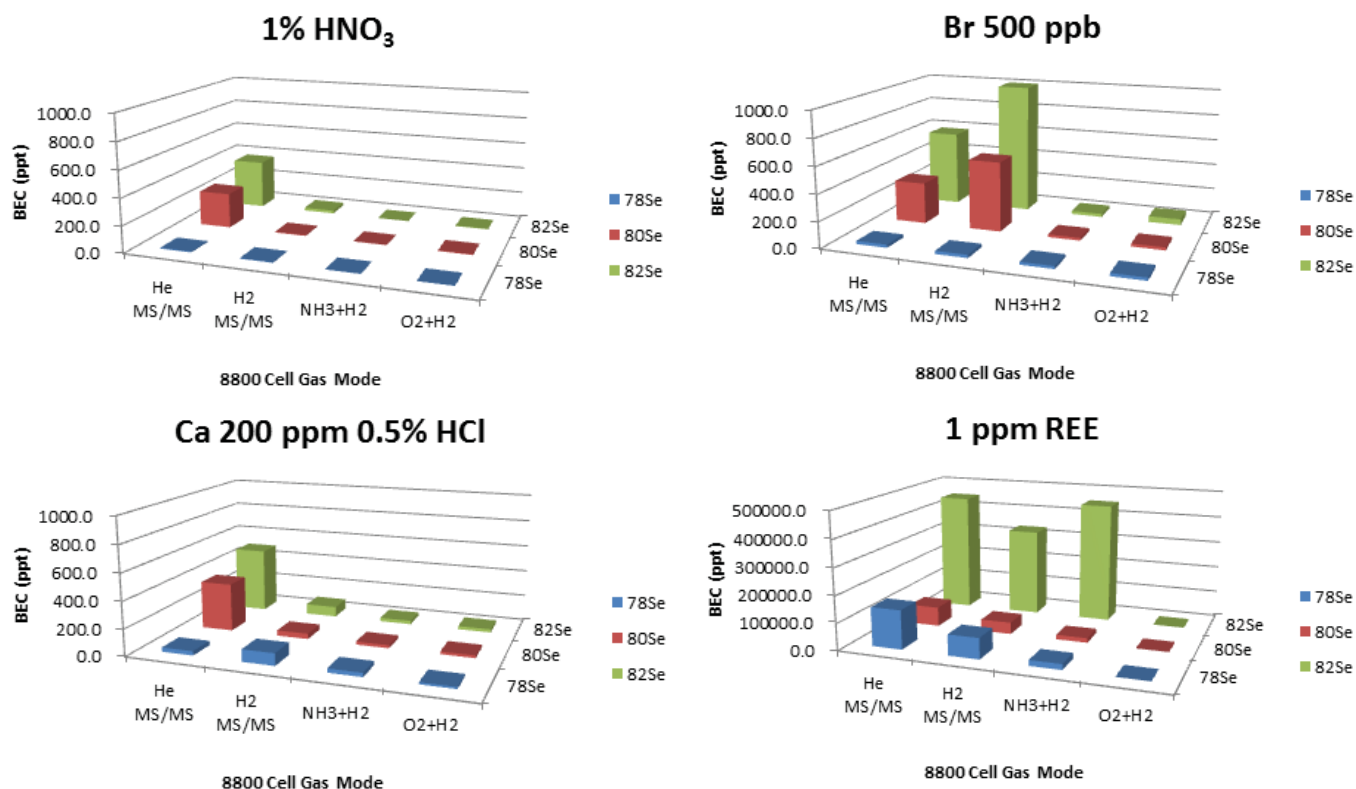
Figure 2. Se BECs (all in ppt) in four synthetic matrices using four different cell gases

removed by the 8800 using MS/MS mode with indirect measurement. The ability of the 8800 ICP-MS to remove all these interferences under a single set of operating conditions permits accurate analysis of multiple Se isotopes in practically any natural sample matrix. This consistent and reliable performance ensures that no prior knowledge of sample matrix is required and matrix-specific method development is no longer necessary.

Selenium on-line isotope dilution analysis (OIDA)
A ^{82}Se-enriched standard (^{82}Se 97.43%, ^{80}Se 1.65% and ^{78}Se 0.51%), Oak Ridge National Laboratory (USA), was dissolved in ultrapure HNO_3 and diluted to make an isotopic spike solution. This spike solution was then added on-line to all samples using the standard Agilent ISTD mixing kit. The presence of carbon enhances the ionization of Se in the plasma, increasing sensitivity, so isopropyl alcohol (IPA) was added to the spike solution to give a final concentration of about 1% IPA in the sample. The Se concentration in the samples was calculated using the measured isotope ratios of two Se isotopes according to the formula presented in Figure 3. No internal standard is required for OIDA, since the

change in the Se ratio is measured and not the absolute Se signal. As a result, errors due non-spectroscopic effects such as signal drift are eliminated. For an explanation of the theory and practical application of OIDA, see Reference [1]. Figure 3 shows the isotope dilution formula used to calculate the Se concentrations in each sample.

$$C_x = C_r \left(\frac{^rR_m - R_s}{R_m - R_n} \right) \left(\frac{R_s - R_m}{R_s - ^rR_m} \right) \quad [1]$$

R_m: Measured isotope ratio of mixed reference standard and spike solution

^rR_m: Measured isotope ratio of mixed unkown sample and spike solution

C_x: Concentration of unknown sample

C_r: Concentration of natural reference standard

R_n: Natural isotope ratio

R_s: Isotope ratio of spike

Figure 3. Isotope dilution formula used to calculate analyte concentration

Quantification error study using OIDA

For accurate quantification using ID, the measured isotope ratios must be corrected for mass bias by bracketing the unknown samples with a mass bias correction standard. However, with the OIDA method, good accuracy can be achieved without the need for mass bias correction, if the concentration of the unknown sample (Cx) is close to the concentration of the natural reference standard (Cr) — see Reference [2]. Theoretically, there is zero error when Cx = Cr. The accuracy of the Se ratio measurement with no mass bias correction was investigated. Natural Se solutions at 0.025 ppb to ~20 ppb were analyzed using a 2 ppb natural Se solution as the reference standard. Figure 4 shows that measurement accuracy of 95–105% without correction should be achievable at Se concentrations ranging from 100 ppt to 5 ppb.
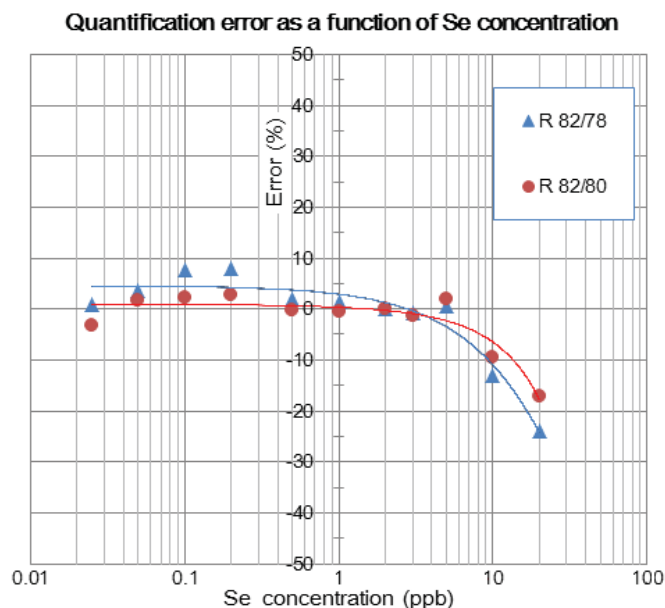

Figure 4. Quantification error analysis of the OIDA method

Results

The concentration of Se was determined in twelve different CRMs using the OIDA method (with no correction for mass bias). The CRMs consisted of a wide range of different sample matrices obtained from NIST (Gaithersburg MD, USA), GSJ Geochemical Reference Samples (Tokyo, Japan), Japan Society for Analytical Chemistry (Tokyo, Japan), and National Institute of Metrology (Beijing, China), as follows:

- NIST 1643e Environmental Water
- JASC 0302-3 River Water
- JB-3 Basalt Rock
- JSI-1 Sediment
- NIST 1646a Estuarine Sediment)
- JSAC0411 Volcanic Ash Soil
- NIST 1566a Oyster Tissue
- NCSZC 81002 Human Hair
- NIST 2976 Mussel Tissue
- NIST 1575a Pine Needles
- NIST 1515 Apple Leaves
- NIST1573a Tomato Leaves

The CRMs were prepared either by simple dilution (for the water samples) or by digestion in a mixture of HNO_3, HCl and H_2O_2 using a Milestone ETHOS closed vessel microwave digestion system (Milestone, Sorisole, Italy) and following the manufacturer's recommended procedures. The tested samples were all diluted to give an expected Se concentration between 100 ppt and 5 ppb, although for unknown samples this would not be necessary if mass bias correction was applied. An integration time of 1 s was used for each of the three Se isotopes measured and ten replicates were acquired. This provided the measured isotope ratio value ^{r}Rm. The Se concentration was then calculated by using the formula in Figure 3.

Figure 5 shows the Se results for each CRM expressed as % recovery relative to the certified value. The measured results for Se were in good agreement with the CRM values (90–112%), using both Se isotope pairs: 78/82 and 80/82. The accurate measurement of Se in this wide range of sample matrices, some including high levels of Ca and REE, demonstrates the consistency and matrix-independence of the interference removal offered by the 8800 in MS/MS mode, using O_2/H_2 cell gas with indirect measurement. Good recoveries for both isotope pairs quantified by OIDA demonstrate that the 8800 ICP-MS was able to remove multiple interferences from ^{78}Se, ^{80}Se and ^{82}Se.
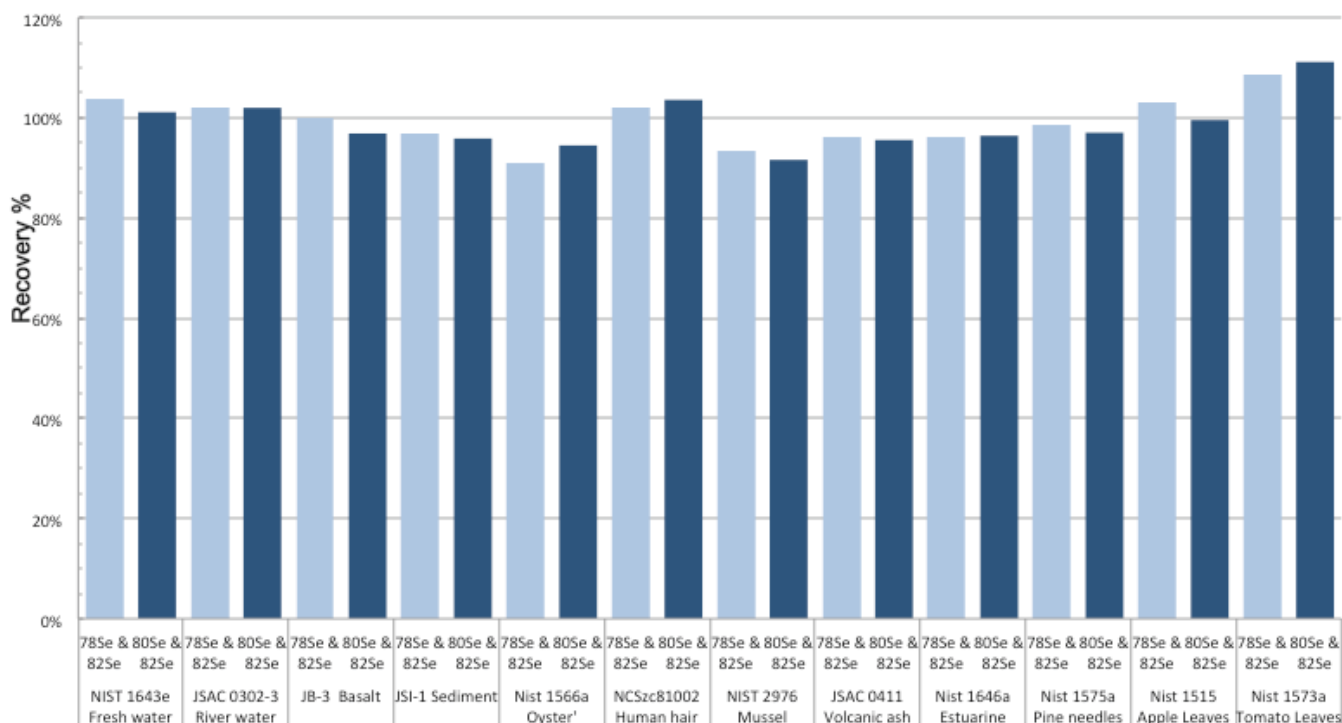
Figure 5. Measurement of Se in twelve CRMs using the Agilent 8800 in MS/MS mode with OIDA. Data shown for both Se 78/82 and Se 80/82 ratios.

Conclusions

Until now it has been difficult to accurately measure Se at ultratrace levels in high matrix samples. The poor ionization and low signal sensitivity of Se, the absence of a good internal standard and the presence of multiple interferences on all Se isotopes makes this one of the most difficult applications in ICP-MS.

However, it has been shown that the new Agilent 8800 ICP Triple Quad operating in MS/MS mode is able to remove all interferences on the three major Se isotopes, giving accurate Se measurement regardless of sample type. By effectively eliminating matrix-derived and plasma-based interfering ions, Q1 ensures that the cell conditions remain consistent with widely different sample matrices, delivering accurate analysis for unknown and variable samples. By mass-shifting Se^+ by reaction with O_2/H_2, and performing indirect measurement of Se as the SeO^+ product ion, all potential interferences are removed, including the challenging polyatomic and doubly-charged interferences from Ca, Br and REE matrices. In contrast with ICP-QMS, the 8800 with MS/MS also eliminates the elemental isobaric overlaps from Zr, Mo and Ru, which would interfere with the measurement of the SeO^+ product ions.

Importantly, MS/MS enables the use of a single set of operating conditions for each element, regardless of the sample matrix. This eliminates the matrix-specific method development that is required for reaction modes on ICP-QMS, making the 8800 easier to use than conventional quadrupole ICP-MS.

The ability of the 8800 ICP-MS to make multiple isotopes available for measurement also enables the use of ID for quantitation. OIDA, a variant of ID more suited to routine labs, was successfully used to measure twelve widely different CRMs with no internal standard and without the need for mass bias correction.

The 8800 brings the power of MS/MS to ICP-MS to improve accuracy and ease of use, and to expand the applicability of ICP-MS to the most challenging sample types.

References

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