



Rapid Speciation and Determination of Vanadium Compounds in Water Samples

Speaker 1:

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Research Triangle Institute

Outline

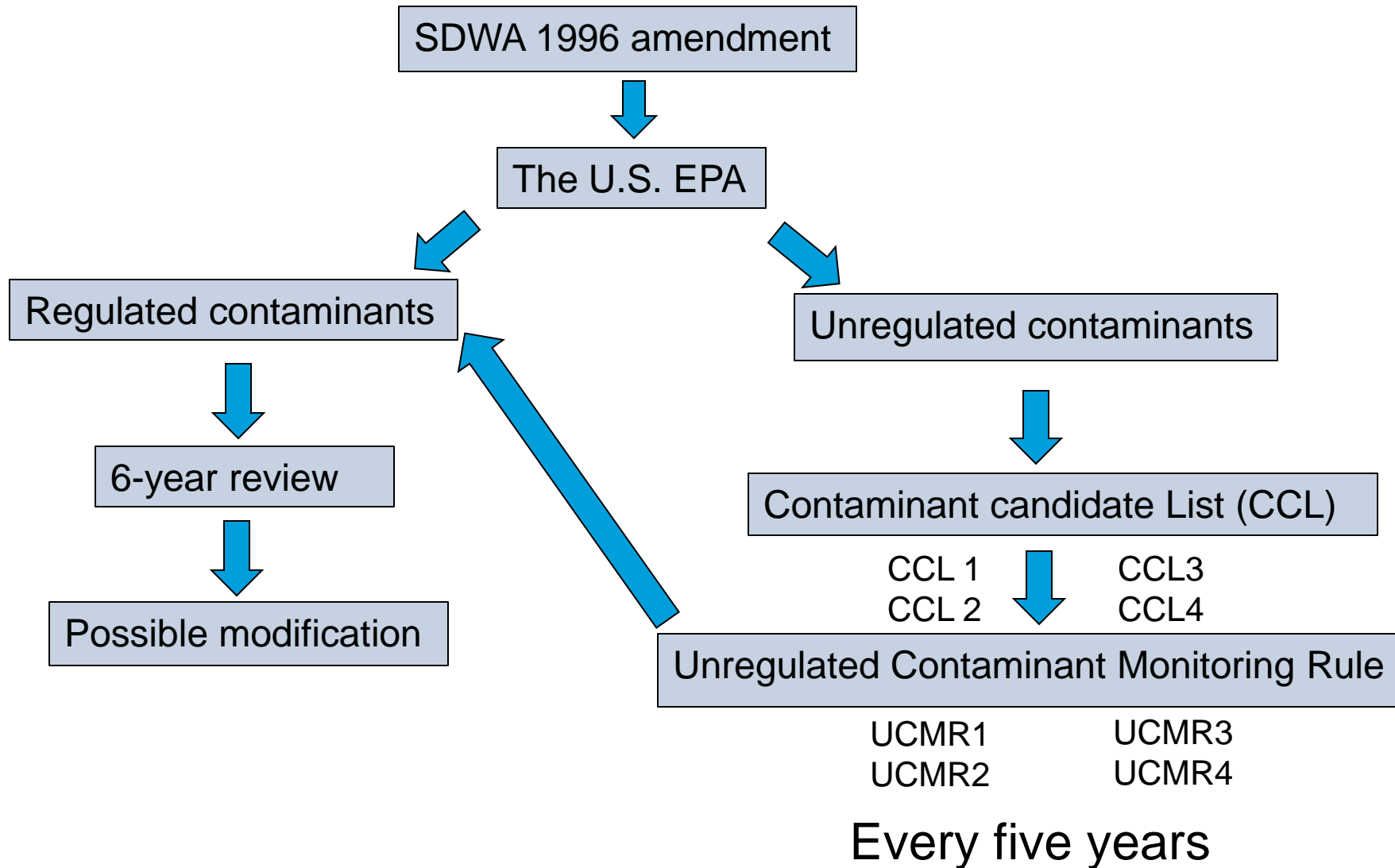
- U.S. drinking water regulations
- Metals regulations in drinking water
- Metals speciation and methods
- IC-ICP-MS as a tool for metals speciation
- Speciation resources

Safe Drinking Water Act and EPA Drinking Water Regulations

- Safe Drinking Water Act (SDWA):
 - Public Health Service Act (PHSA) in 1912 and revised to become SDWA in 1974
 - Amended in 1986 and 1996
 - Authorizes EPA to regulate drinking water to protect the public health
- EPA drinking water regulations:
 - Started to regulate drinking water contaminants in 1912
 - Currently regulate more than 90 contaminants including 17 metal elements
 - Evaluate unregulated contaminants

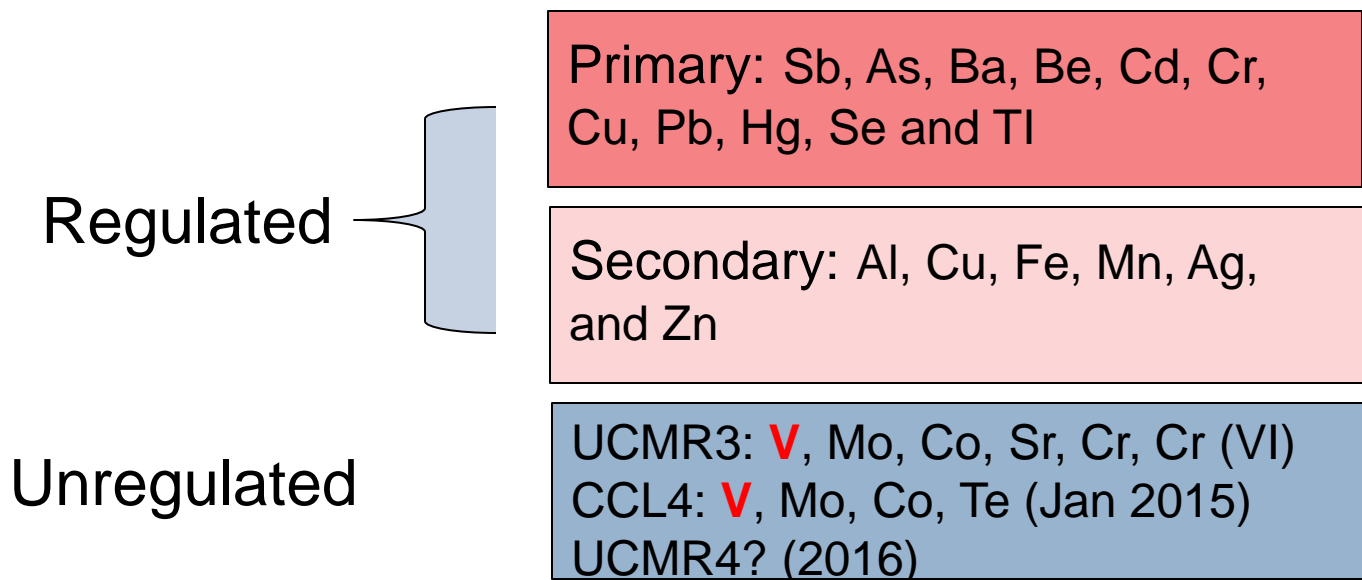


Regulated Contaminants and Unregulated Contaminants



Metals (and Metalloid) Regulations in Drinking Water

- National Primary Drinking Water Regulations: legally enforceable standards that apply to public water systems.
- Secondary list: non-enforceable guidelines to regulate contaminants:
 - May cause cosmetic effects (such as skin or tooth discoloration), or
 - Aesthetic effects (such as taste, odor, or color) in drinking water



Metals Speciation

- Speciation analysis: to identify and quantify different species (chemical forms) of a particular metal element

Metals	Species 1	Species 2	Species 3
Antimony	Sb (III)	Sb (V)	
Arsenic	As (III)	As (V)	Organoarsenic
Chromium	Cr (III)	Cr (VI)	
Mercury	Hg ²⁺	Methylmercury	Ethylmercury
Selenium	Se (IV)	Se (VI)	Others
Vanadium	V (IV)	V (V)	

Metals Speciation and its Importance

- Why is speciation important?
 - Study of the health effects: toxicity
 - Safety purpose: bioavailability and reactivity
 - Understanding of geochemical changes: mobilization and transformation
- Truth about speciation:
 - A toxic species is more relevant than the total concentration (California Cr (VI))
 - Speciation analysis is important but not required in new regulations yet
 - A few speciation analytical methods are in draft form, very few got approved by EPA (321.8 for bromine speciation) and ASTM (D6994-04 for cyanide speciation)



Principles of Speciation Analysis

Sample containing various compounds

Separation



Individual detection of compounds

- Preservation of original species distribution during sample preparation and storage:

- Loss of species
- Transformation of species

- Different separation mechanisms:
 - Charges
- Different species must be well separated
- Ability to tackle several analytes with same instrumentation

- ICP-MS accurately determines the total element concentration within each separated fraction of the species

Commonly Used Techniques for Speciation

- **GC-ICP-MS**
 - Gas chromatography for separation and ICP-MS for detection
 - Example: mercury speciation
- **IC-ICP-MS (IC sometimes is called HPIC or even HPLC)**
 - Ion chromatography for separation and ICP-MS for detection
 - Example: arsenic speciation
- **HPLC-ICP-MS**
 - HPLC or UHPLC for separation and ICP-MS for detection
 - HPLC-Q-ICP-MS or HPLC-HR-ICP-MS
 - Example: vanadium speciation
- **CE-ICP-MS**
 - Capillary electrophoresis for separation and ICP-MS for detection
 - Example: arsenic speciation

Arsenic Speciation: CE-ICP-MS and IC-ICP-MS

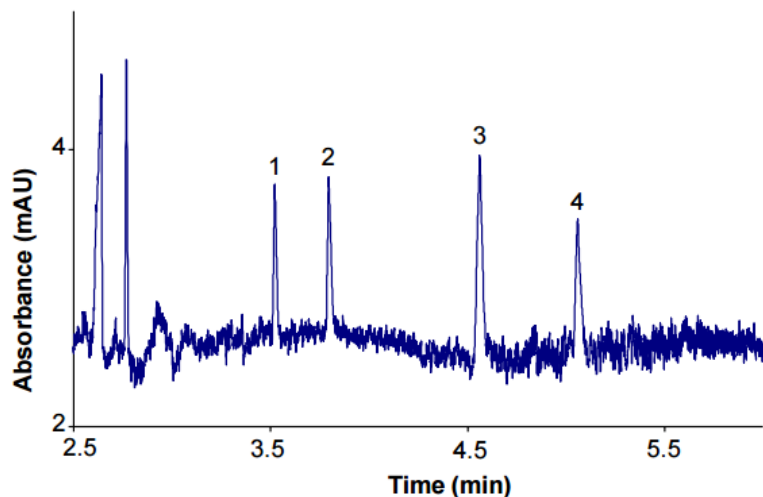


Fig. 4. Determination of arsenic species in a spiked marine sediment sample. Spiked concentration: 10 mg/kg each. Conditions: as for Fig. 2a. Peaks: (1) As(V), (2) MMA, (3) As(III) and (4) DMA.

Capillary electrophoresis
 Sun et al. *Journal of Chromatography A*,
 1039 (2004) 201–208

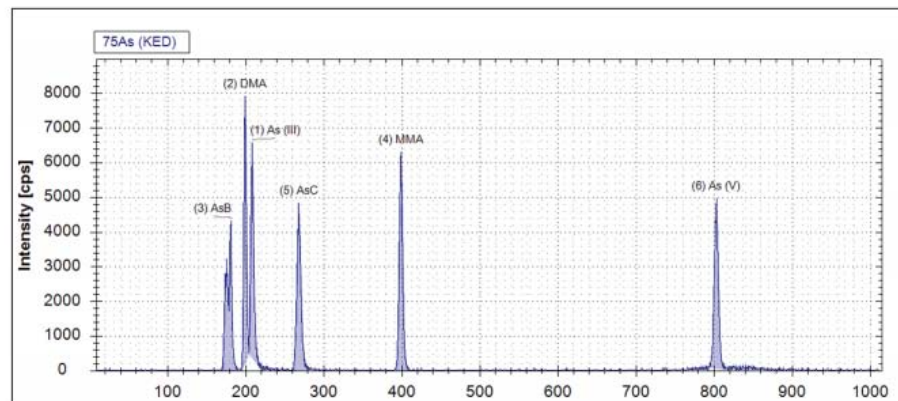


Figure 1: Model separation of six arsenic species, each at 0.45 ng/g in dilute HNO₃.

Column	Dionex AS-7 (2 mm I.D., 250 mm length)
Elution	Gradient
Mobile phase	A: 20 mmol/L Ammonium carbonate B: 200 mmol/L Ammonium carbonate Gradient from 20 to 200 mmol/L in 15 minutes
Injection volume	20 µL
Duration	15 minutes

Table 2: ICS-5000 operating parameters.

IC-ICP-MS for arsenic speciation
 Thermo Scientific AN43099

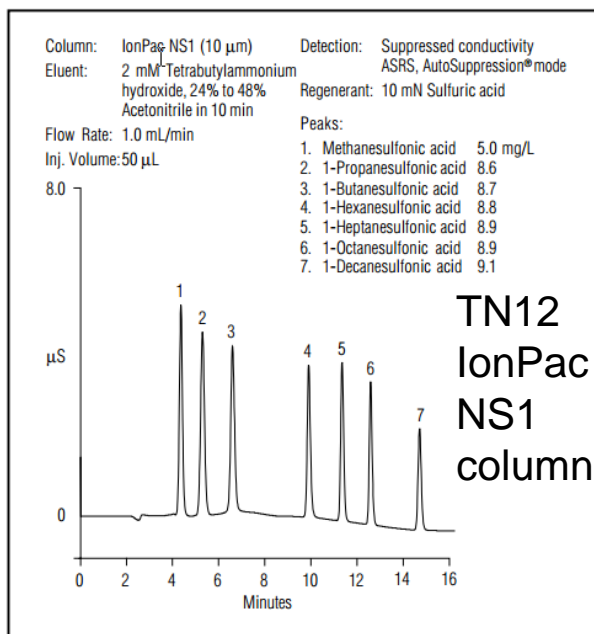
- Advantage:
 - short analysis time, high separation efficiency, and low operating cost
- Disadvantages:
 - High detection limit (100x higher detection limit than IC), high RSDs (can be >30%)

Speciation: Ion Pair and Ion Exchange Chromatography

Both are ion chromatography

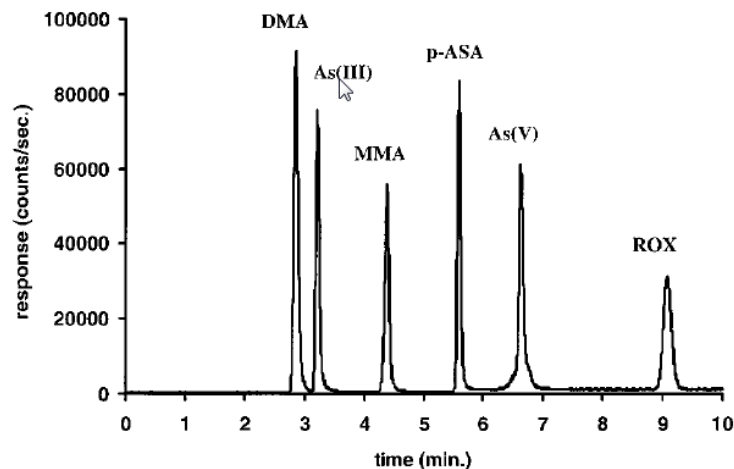
- Ion Pair(ing) Chromatography

- Anionic, neutral and cationic species are separated in only one chromatographic run



- Ion (Exchange) Chromatography

- Low pH: Thermo Scientific™ Dionex™ AS7 or AS4 column with HNO₃ as eluent
- High pH: Dionex AS 7,11, 14, 16 column with eluent of carbonate and hydroxide

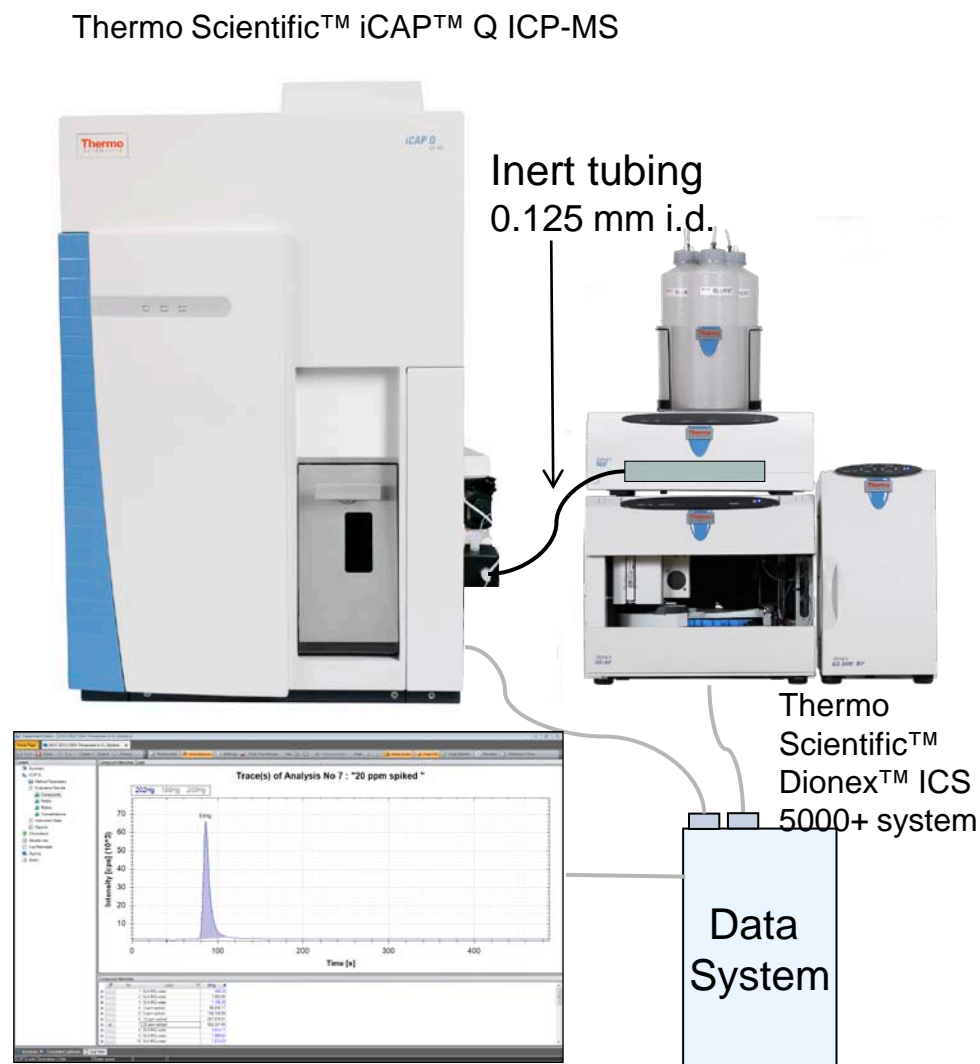


Jackson and Bertsch (2001) AS16 column for arsenic species separation

Ammann (2011) review on arsenic speciation

A Complete Integrated IC-ICP-MS System

- IC-ICP-MS is a fully integrated technique
 - One software and simple hardware connection
- The completely metal-free IC system eliminates contamination by the system
- Large range of IC columns
 - Tailored IC applications
 - Flexibility and support to develop applications
- For many elemental speciation applications IC is the preferred chromatographic choice for ICP-MS:
 - Mobile phases are simpler/cleaner for lower interferences, lower background and lower limits of detection
 - Narrower bore columns (2 mm ID) for higher efficiency separation and lower mobile phase consumption

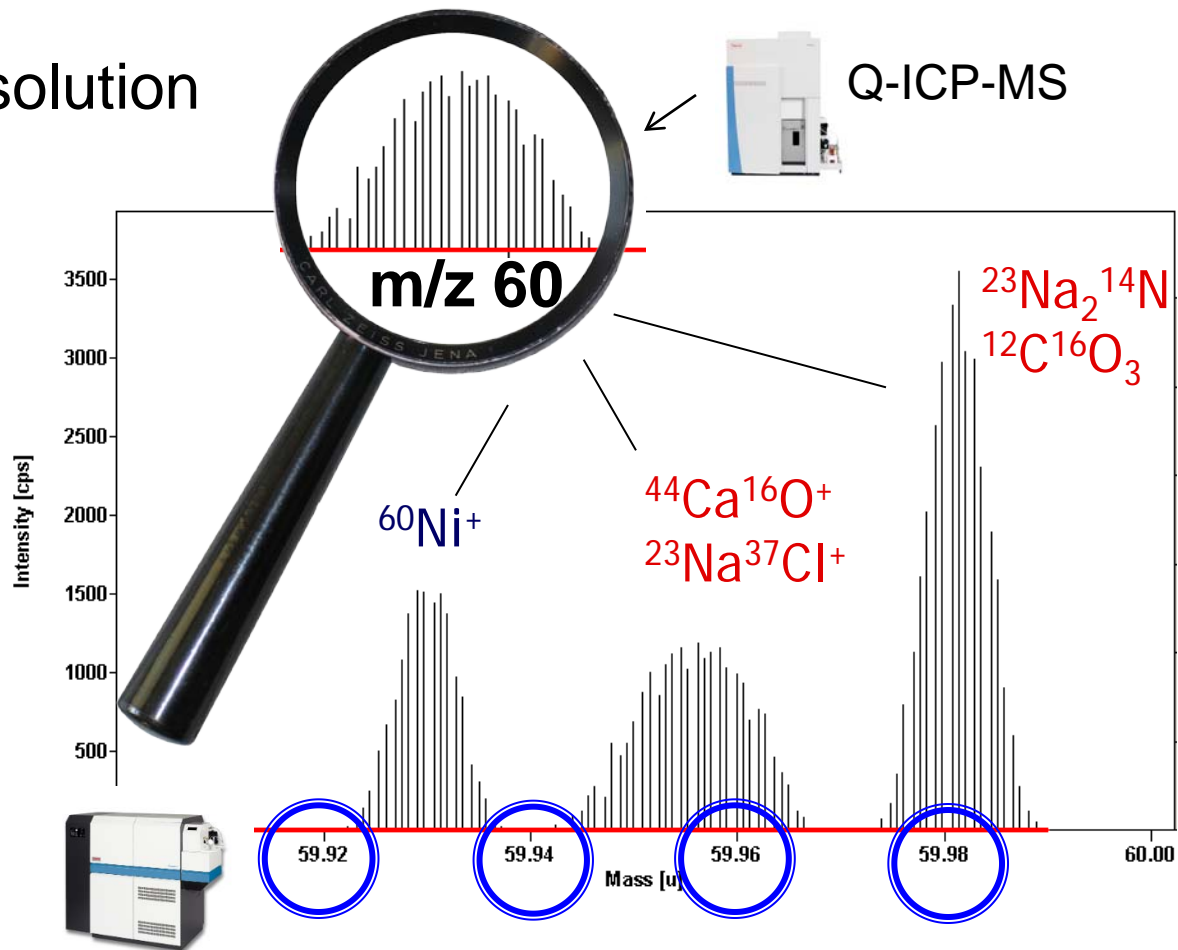


Thermo Scientific™ Qtegra™ Intelligent Scientific Data Solution™ software

The difference between Q-ICP-MS and ICP-SF-MS

The power of high resolution

- ICP-MS with quadrupole analyser:
 - No separation between the analyte (m/z 59.93) and the interferences (m/z 59.958, 59.982)
- ICP-MS with magnetic sector field analyzer:
 - separation of the analyte from the interferences by their small mass difference (High Resolution)



ICP-SF-MS: medium resolution
R=4000

Comparison of ICP-MS and SF-ICP-MS in Speciation

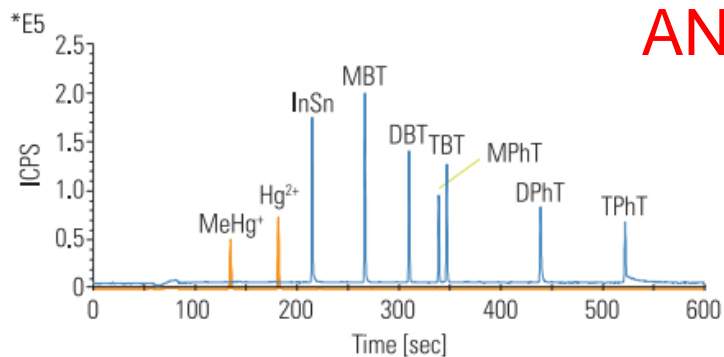


Figure 8a: Multi-element chromatographic data of a 0.5 ng/mL standard acquired with GC-XSERIES 2.

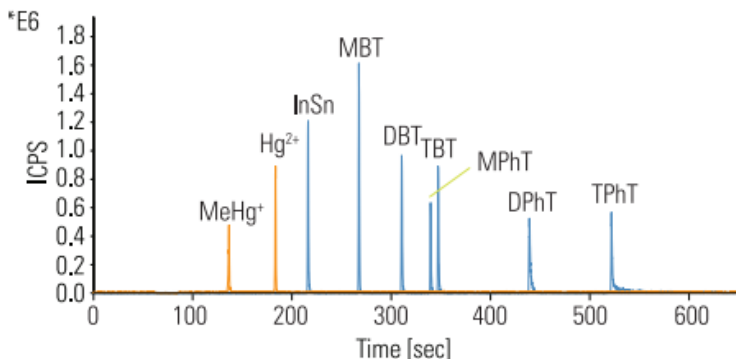


Figure 8b: Multi-element chromatographic data of a 0.5 ng/mL standard acquired with GC-ELEMENT 2.

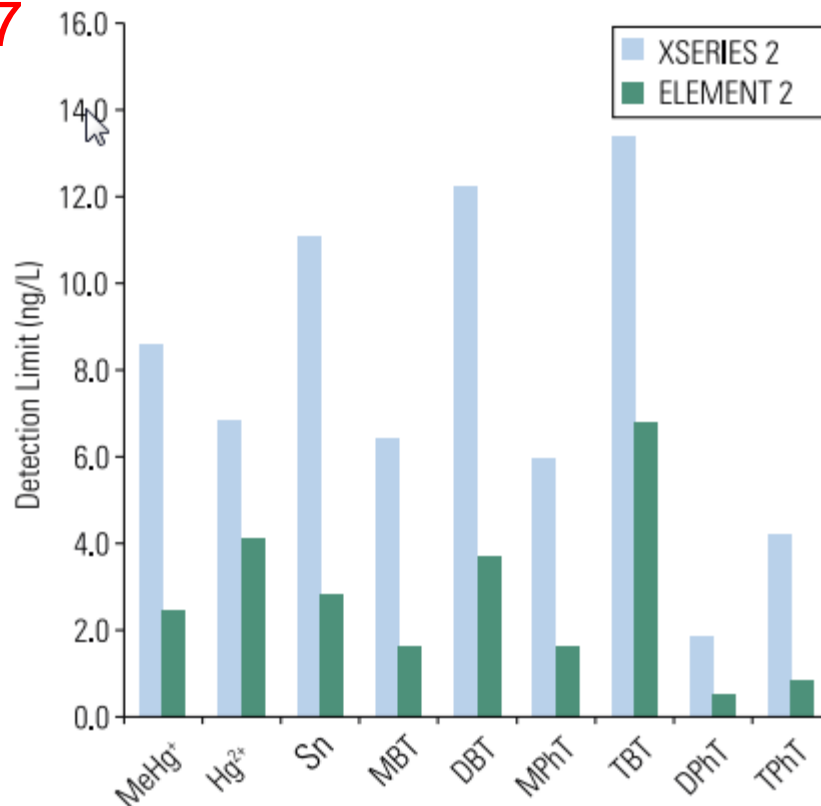


Figure 9b: Limits of Detection in ng/L for XSERIES 2 and ELEMENT 2.

- The chromatograms show identical peak shapes and retention times

- SF-ICP-MS has much lower detection limit compared to ICP-MS

Resources for Speciation

- **Webinars:**

- Complete Inorganic Elemental Speciation Solutions for Environmental Applications
- Bromine Speciation in Ozonated Drinking Water Using Ion Chromatography and Inductively Coupled Plasma Mass Spectrometry (June 9)

- **Speciation web site:**

- <http://www.thermoscientific.com/en/about-us/videos/speciation-using-ic-icp-ms.html>
- European Virtual Institute for Speciation Analysis:
 - www.speciation.net

- Application notes/notebook/white papers:

- Application notebook: Speciation Application Summary Ion Chromatography: Trace Elemental Species Separation and Detection
- Application note 43098: Speciation Analysis of Cr(III) and Cr(VI) in Drinking Waters
- Application note 30127: Speciation of Trace Elemental Species using GC-ICP-MS and GC-HR-ICP-MS
- White Paper: Coupling of an Inert Ion Chromatographic System with ICP-Q-MS for Robust and Accurate Elemental Speciation
- White paper: Benefits of Coupling IC with ICP/MS

Thank You for Your Attention!

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Questions?

Speciation of Vanadium by IPRP-UPLC-ICP-SF-MS

- Introduction
 - Vanadium in the environment
 - Vanadium chemistry and environmental implications
- Chromatographic Method development
 - Mechanism of separation
 - Variation of mobile phase components
- Spectrometric analysis
 - Separation vs resolution
- Calibration and quantification
 - Application to an environmental sample
- Conclusions and Future Directions

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\$800M FY14 revenue, reinvested in programs, facilities, capabilities.

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History, Facts and Figures



- Established in 1958
- Collaboration among state government, area universities, and business leaders
- Over 3,700 staff in 75 countries
- Over 130 academic disciplines spanning social and natural sciences, international development, etc.
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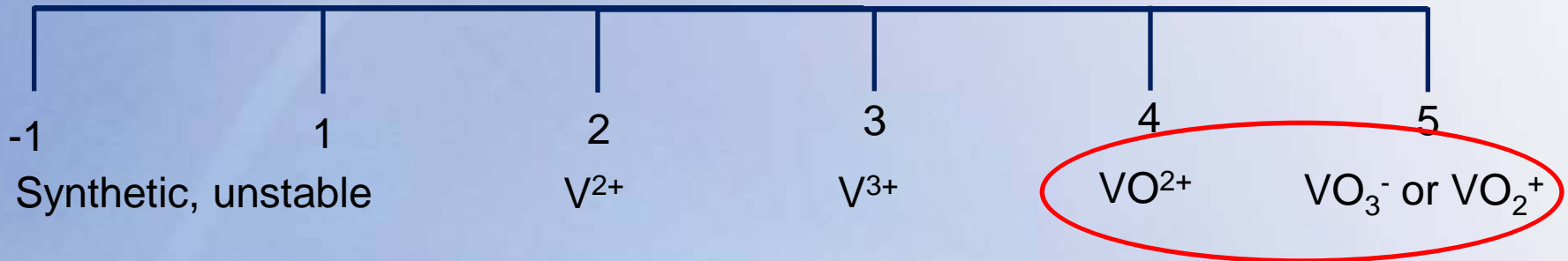
Vanadium in the environment

- Included on EPA Drinking Water Contaminant Candidate List.
- Concentration in the environment
 - 3-310 $\mu\text{g/g}$ (ppm) in soil
 - 0.2-100 $\mu\text{g/L}$ (ppb) in fresh water
 - 0.2-29 $\mu\text{g/L}$ in seawater
- Sources of environmental emission
 - Coal combustion emissions
 - Fly ash
 - Vehicle emissions
 - Metallurgic production(WHO Air Quality Guidelines, 2000)



Vanadium chemistry

- Oxidation states

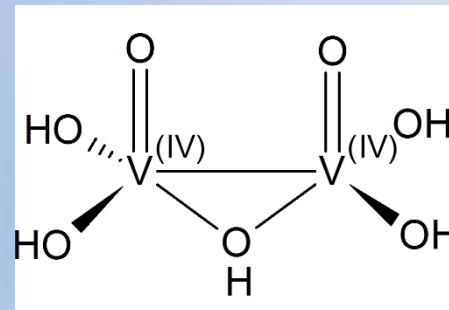


- Isotopes

- ^{51}V (99.75%) and ^{50}V (0.25%)

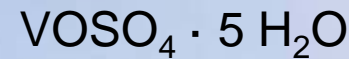
- Chemistry

- Resembles phosphate in environment/biology
- Can form polymeric species
- V(V) is an oxidizing agent, more toxic
- V(IV) has antidiabetic properties



Vanadium Speciation - Toxicity

- Listed as an NTP Priority Contaminant
- Differential acute toxicity of Vanadium species



Rat oral administration
LD₅₀ – 98.0 mg/kg BW

Rat oral administration
LD₅₀ – 448.0 mg/kg BW

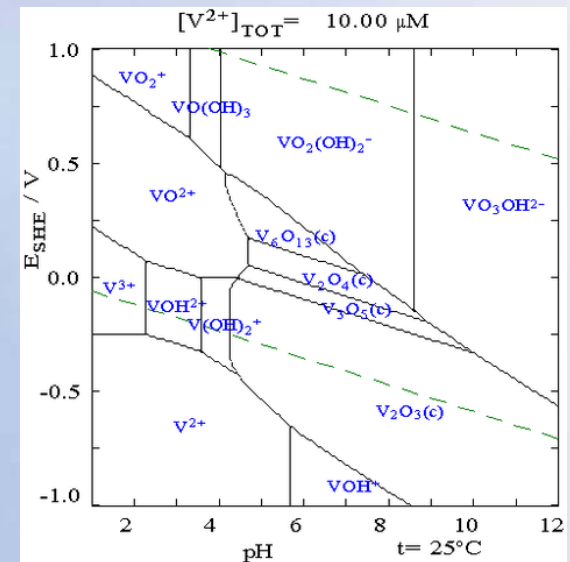
- Symptoms
 - Diarrhea, irregular respiration, decreased locomotor activity.

Speciation of Vanadium compounds

- Speciation varies with conditions
 - pH, redox chemistry, species stability
 - Low pH – III, IV; High pH – V
- Liquid chromatographic separation can separate based on charge
- Method must stabilize speciation, provide accuracy at low concentration
- Detection methods:
 - UV-Vis, AA, ICP-OES: Insufficient sensitivity for low (ppb-ppt) concentrations
 - ICP-MS – low (ppb-ppt) level quantification

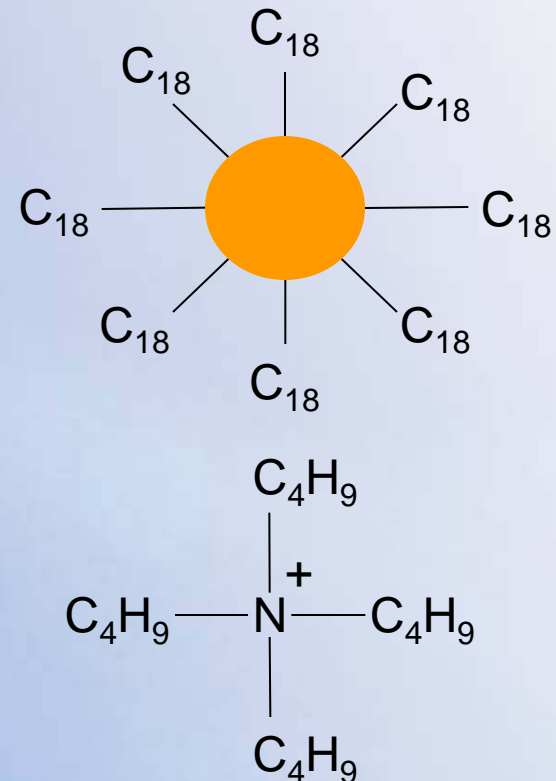


Photo courtesy of Wikipedia



Ion pairing reverse phase chromatography

- RP chromatography
 - Organic mobile phase
 - Requires O_2 addition, interferes with plasma
 - Carbon deposits on cone
- Ion pairing RP chromatography
 - Non-polar ion pairing agents separate ions on non-polar stationary phase
 - Less organic mobile phase.
 - Separates based on polarity and charge
 - Easy to modify separation by varying pH
 - Previously applied to As, V in other matrices, etc.



Experimental

■ UHPLC – Waters® ACQUITY system

- Column: Acquity BEH C₁₈ (1.7 μm), 2.1x50 mm
- Mobile phase: 18 mM EDTA, 0.50 mM tetrabutylammonium hydroxide (TBAOH), 20 mM H₃PO₄
- pH – 7.00 w/ NH₄OH, 4% MeOH
- Temp: Amb, 0.60 mL/min, isocratic (P ~ 10,000 psi)



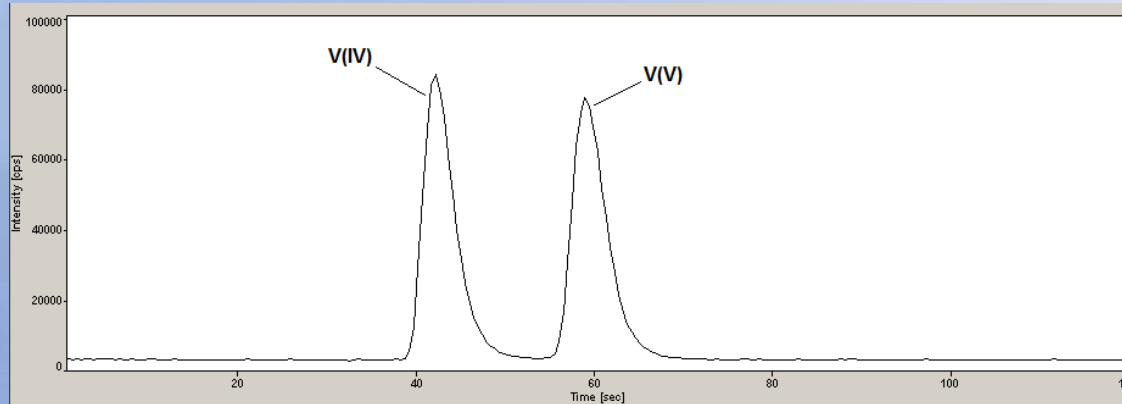
■ ICP-SF-MS – Thermo Scientific™ Element 2™ ICP-MS system

- Ni cones, ESI PC3 peltier-cooled cyclonic spray chamber
- 1475 W FP, Pt guard electrode
- ⁵¹V isotope
- Low resolution (M/ΔM ~ 350)
- Medium resolution (M/ΔM ~ 6000)



Ultra-high performance liquid chromatography

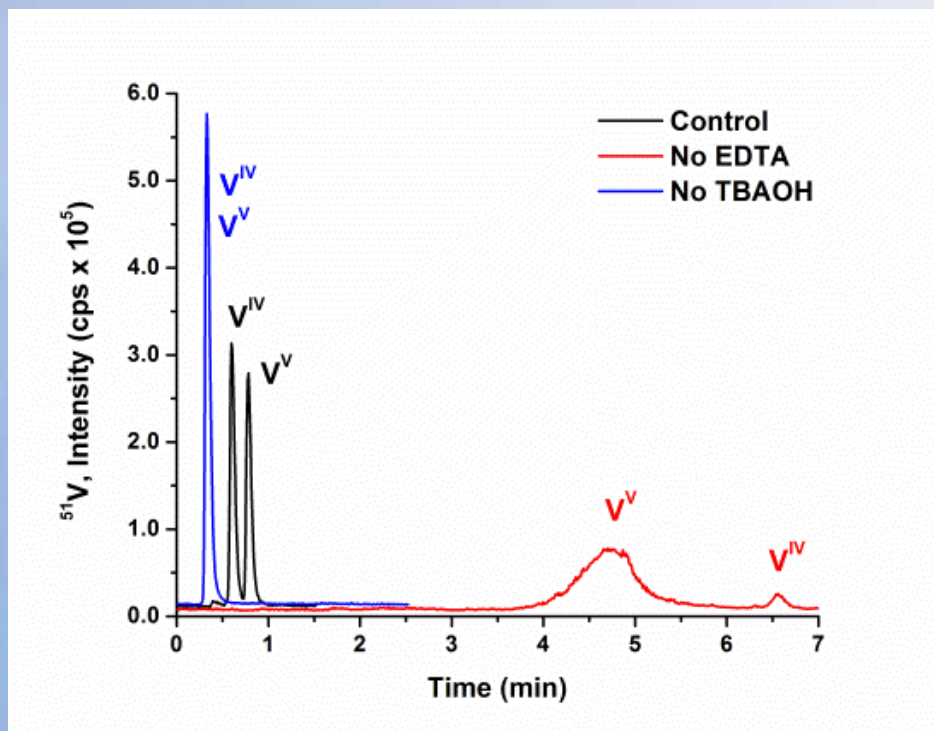
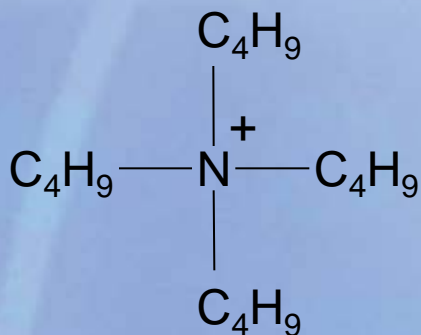
- Why UHPLC?
 - Higher resolution – more theoretical plates
 - Faster separation – higher throughput



- Literature HPLC/CE methods – Retention time - ~5-10 min.
- CE – drifting retention times
- IEC-ICP-AES – Takaya and Sawatari, Indus. Health (1994) 32, 165-178 use; retention ~10 minutes for V(V), LOQ – 1 ppm
- Our method – complete separation ~1 min

Method development – Mobile phase

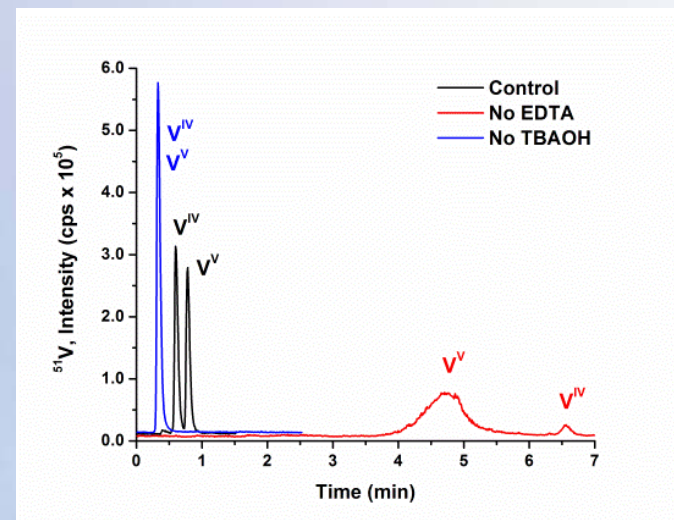
- Omitting mobile phase constituents varies selectivity
 - TBAOH – No ion pairing agent
 - No retention for either charged species, elutes with the void
 - No resolution



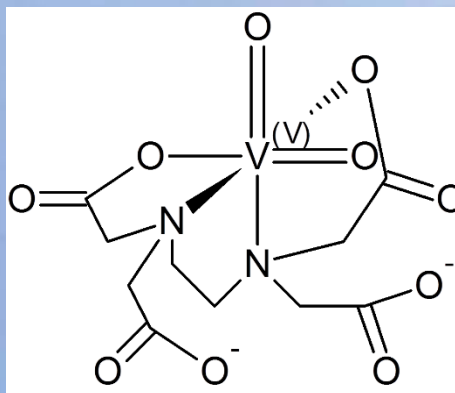
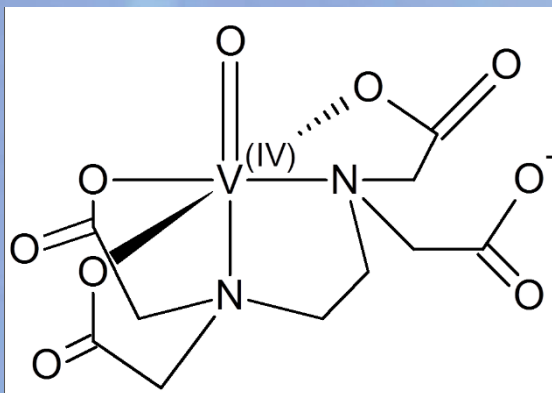
Kilibarda, Afton, Harrington, Yan, and Levine, J. Chrom. A 1304 (2013) 121-126.

Method development – mobile phase

- EDTA – No complex, no displacement
 - No nonpolar core
 - No complex, ions higher charge, reverse order
 - No EDTA to displace V ions
- Decreased oxidative stability
 - EDTA controls rate of oxidation
 - Controls inner sphere
 - Minimizes inner sphere electron exchange



Kilibarda, Afton, Harrington, Yan, and Levine,
J. Chrom. A 1304 (2013) 121-126.



Alternate eluent components

- Other complexing agents for V speciation

- 2,6-pyridinedicarboxylic acid w/ lithium hydroxide

- Decreased species stability – degradation after 2 hours
- Less stable complexation, less able to control inner coordination redox
- Different column, so different selectivity/separation mechanism.

M.J. Tomlinson, J. Weng, J.A. Caruso, JAAS (1994) 9, 957-964.

- 4-(2-Pyridylazo)resorcinol

- Chromophore, UV-Vis detection
- Non-vanadium-specific chelation
 - Interference by other metal ions
- Pre-treatment step and developing step, highly susceptible to pre-treatment process and pH

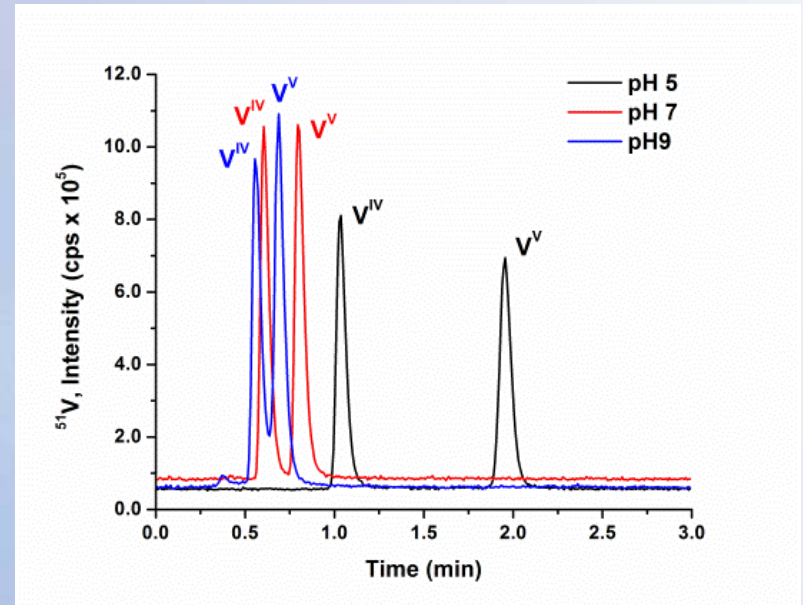
S.J. Tsai, S. Hsu, Analyst (1994) 119, 403-407.

Method development – Mobile phase

- Varying pH varies selectivity
 - Varies EDTA protonation, charge

$$pK_{a3} = 6.16$$

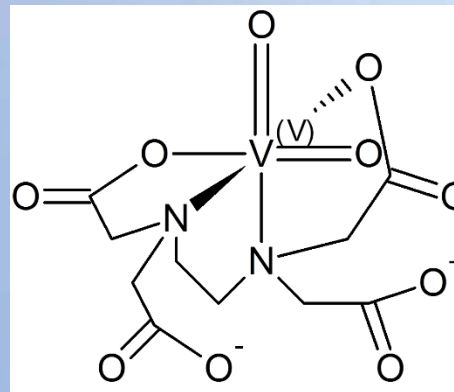
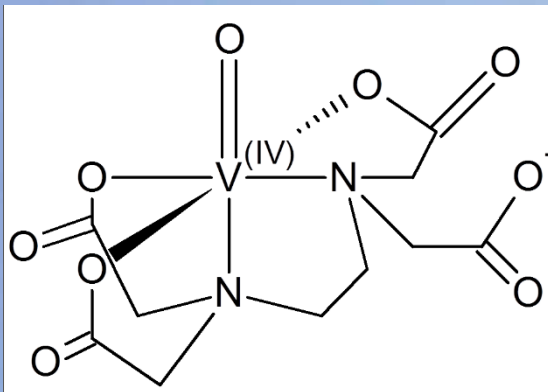
- Phosphate - $pK_{a2} = 7.21$
- Complex protonation does not vary
- Displacement by free EDTA varies
- Order of elution unchanged
- pH ↓, Resolution and RT ↑
- pH 7 – full resolution ($R_s = 1.63$), short RT



Kilibarda, Afton, Harrington, Yan, and Levine,
J. Chrom. A 1304 (2013) 121-126.

Method development – Mobile phase

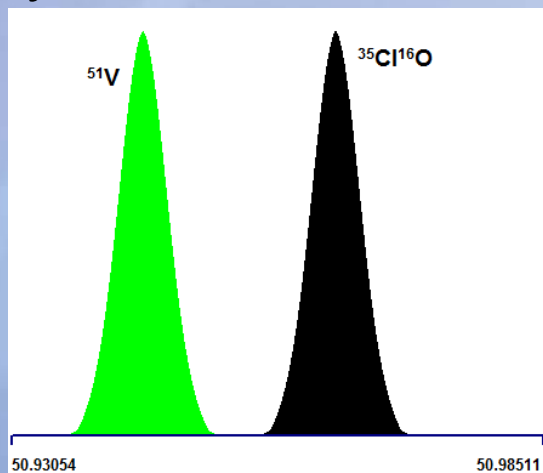
- Must balance mobile phase composition w/ ionic strength
 - ↑ ionic strength, ↓ instrumental sensitivity
 - Aureli, et al., JAAS (2008), 23, 1009-1016 reported decreasing sensitivity at higher concentrations of carbonate buffer, EDTA salt
 - Increase in EDTA → higher carbon content
- Requires organic (MeOH), but may affect complex speciation
 - Previous reports - >4% MeOH



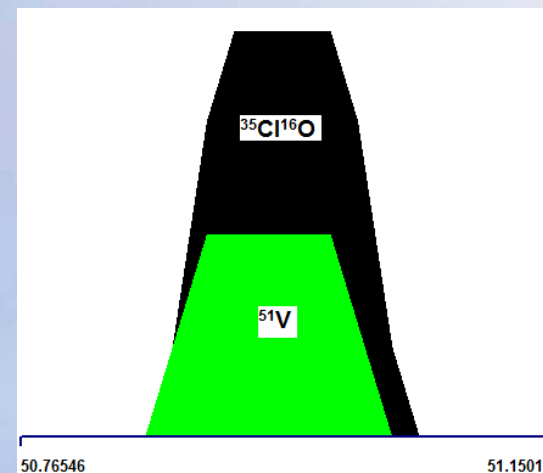
Separation vs Resolution

- ^{51}V polyatomic interference - $^{35}\text{Cl}^{16}\text{O}$

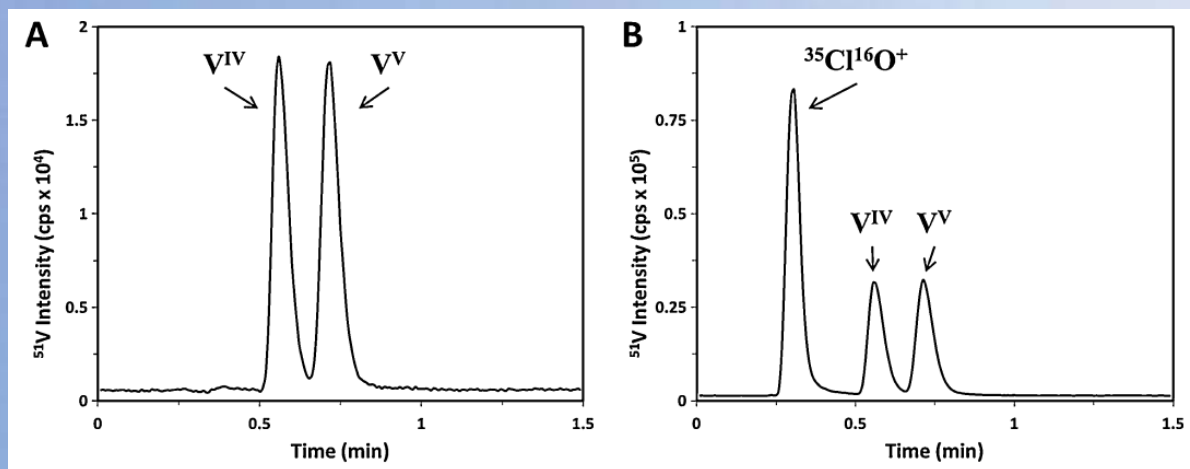
Medium
Res



Low
Res

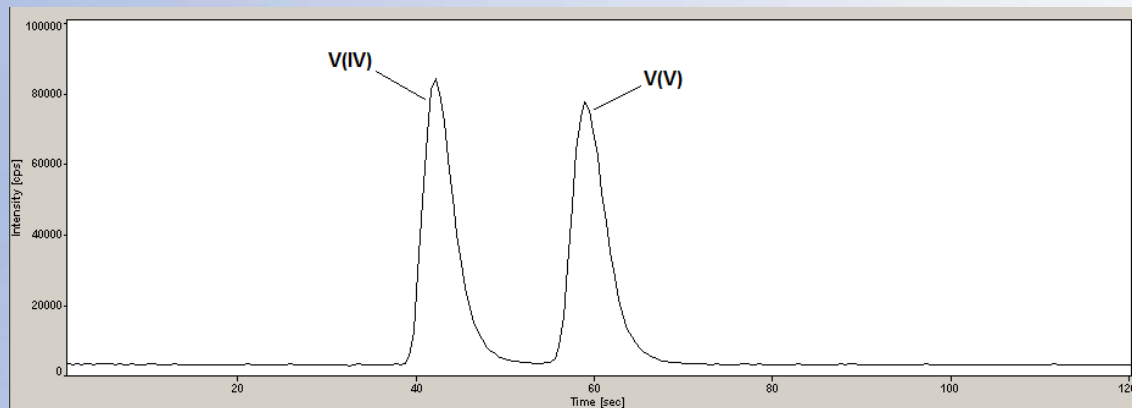


- Complete resolution of vanadium species – 20 mM NaCl

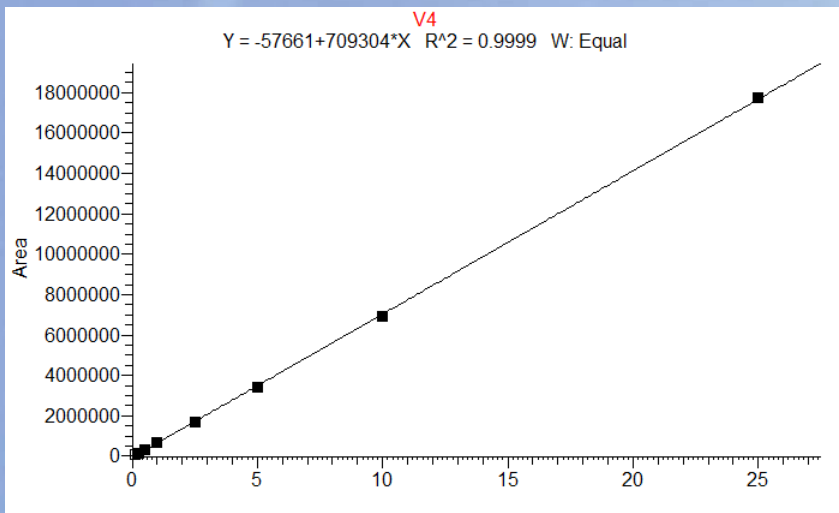


Calibration and quantification

- Calibration range
 - 0.25 – 25.0 ppb
 - Spiked eluent stds
- LOD
 - V(IV) – 0.008 ppb
 - V(V) – 0.013 ppb



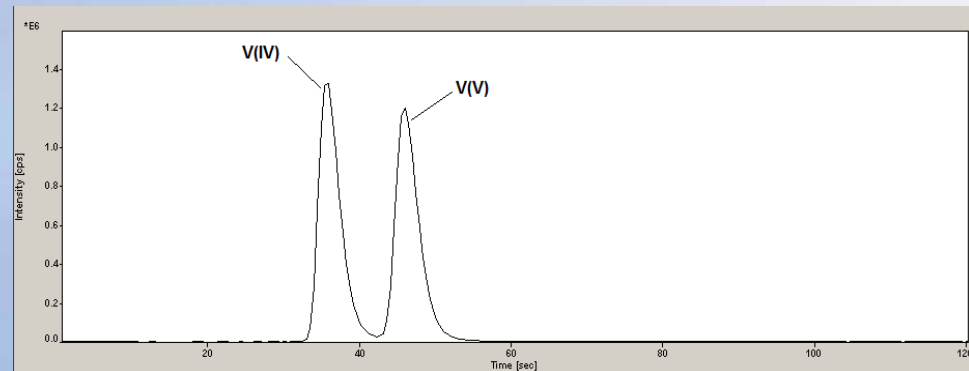
Nominal [V(IV)] = [V(V)] = 2.50 ppb



Standard	Nominal	V(IV)		V(V)	
		Calc Amt	% Diff	Calc Amt	% Diff
Cal1	0.100	0.181	80.7	0.192	92.2
Cal2	0.250	0.317	26.7	0.309	23.7
Cal3	0.500	0.557	11.4	0.550	10.07
Cal4	1.000	1.036	3.60	1.024	2.44
Cal5	2.500	2.509	0.36	2.506	0.23
Cal6	5.000	4.923	-1.54	4.932	-1.37
Cal7	10.000	9.828	-1.72	9.863	-1.365
Cal8	25.000	25.080	0.32	25.065	0.260

Species Stability

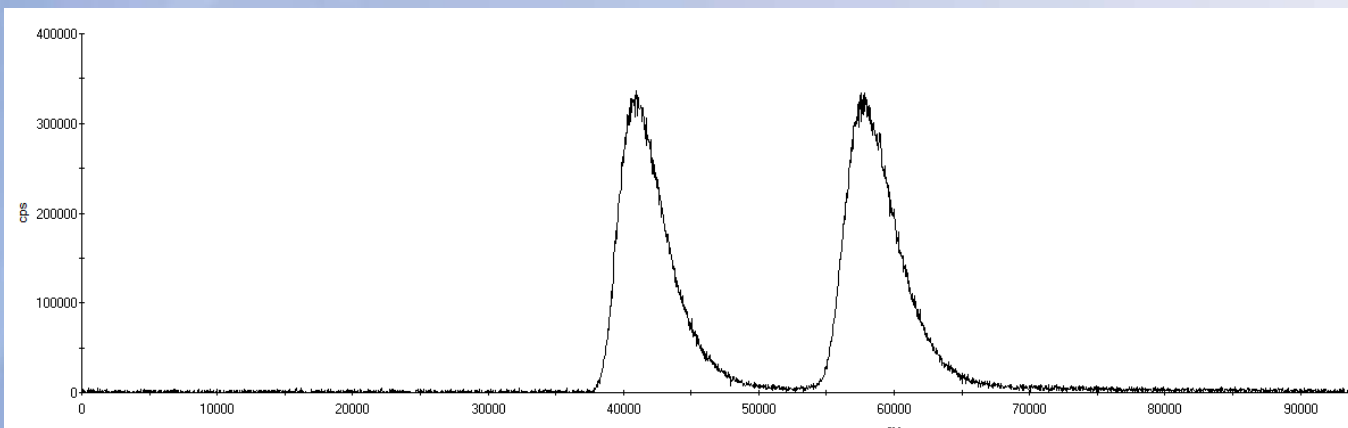
- Good intraday (same day) stability
 - Solvent standards measured within 4 hrs
 - No conversion, but variation in recovery due to instrumental drift
- Interday (different day) stability
 - 7-day autosampler stability
 - Nominal 5.0 ppb
 - Variation artefact of cal fit



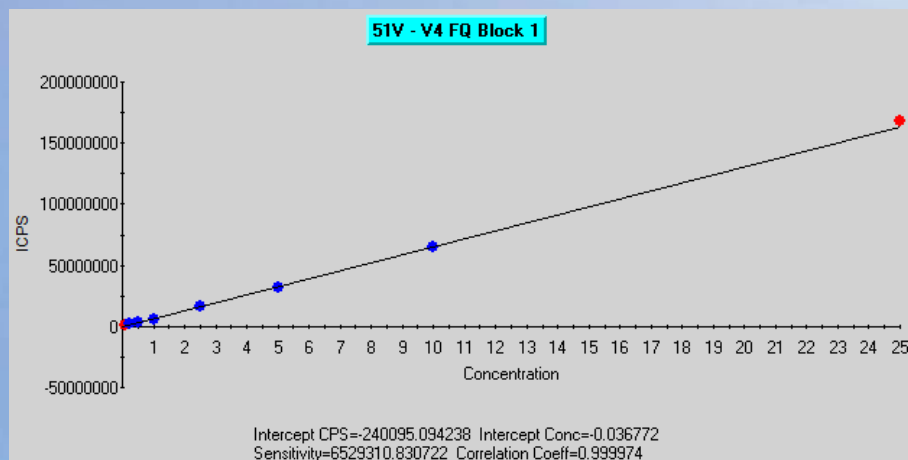
		V4		V5	
Standard	Nominal	Calc Amt	% Diff	Calc Amt	% Diff
Cal1	0.100	0.121	20.6	0.147	47.5
Cal2	0.250	0.272	8.71	0.295	17.9
Cal3	0.500	0.523	4.57	0.538	7.65
Cal4	1.000	1.009	0.926	0.995	-0.472
Cal5	2.500	2.475	-0.990	2.496	-0.180
Cal6	5.000	5.171	3.42	4.880	-2.39
Cal7	10.000	9.947	-0.529	10.001	0.0128
Cal8	20.000	19.873	-0.633	19.919	-0.404

Comparison of other instruments

- X-Series II analysis
 - Ni cones, NH₃ CCT gas – 8.0 mL/min (nominal [V] = 5.0 ppb)



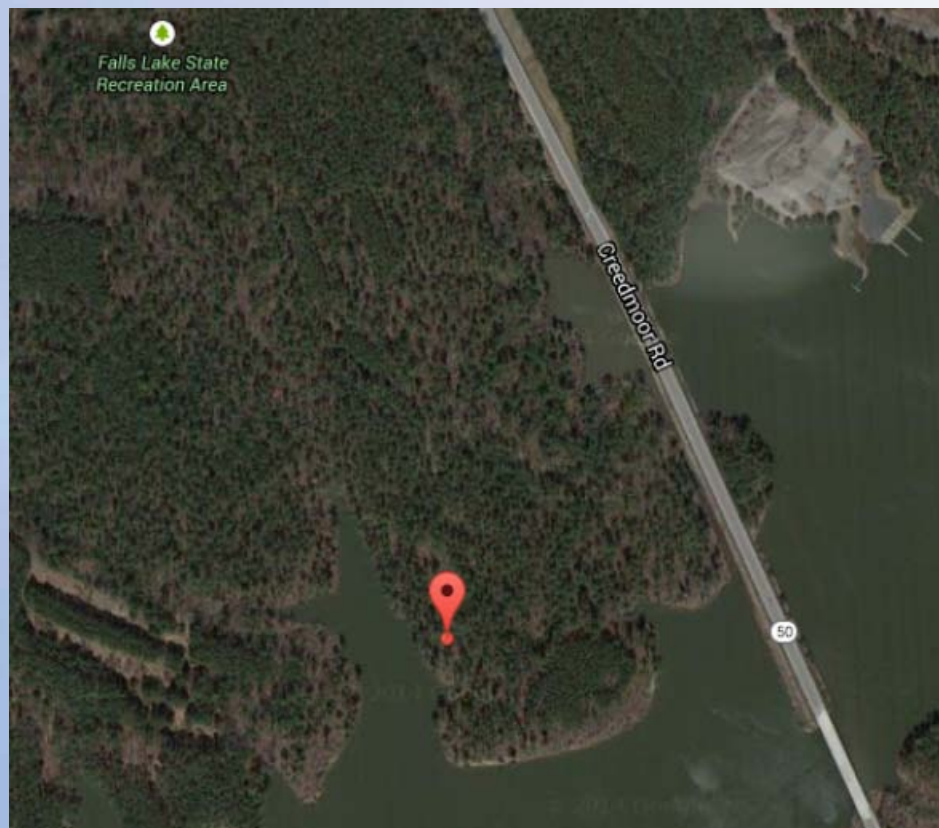
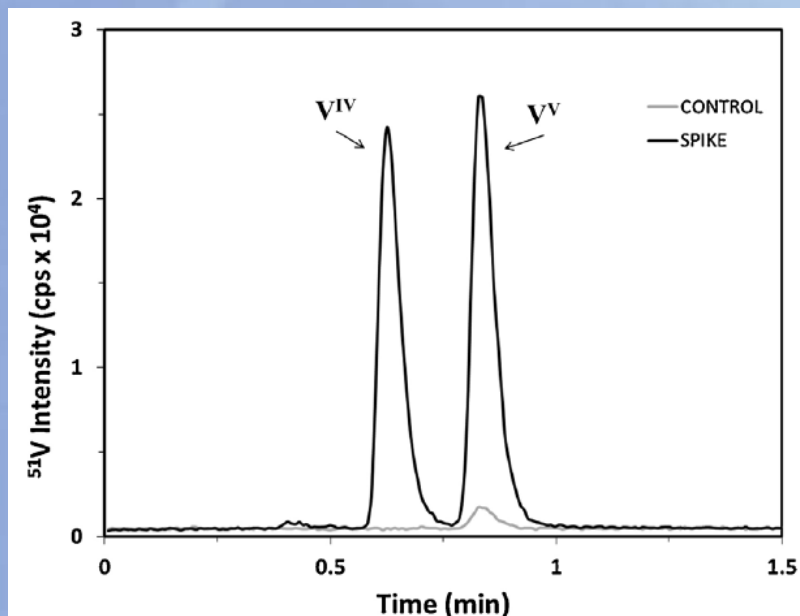
- Separation comparable, S/N lower, comparable cal range.



ELOQ
~1.00 ppb w/o
smoothing

Environmental application

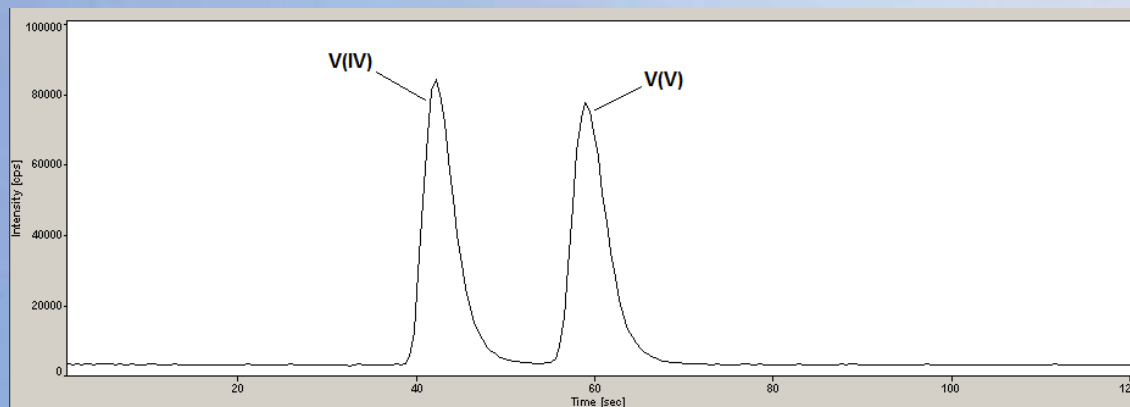
- Lake water sample
 - Collected at Falls Lake
 - Filtered, diluted with mobile phase
 - Analyzed within 24 hrs
 - Spiked w/ nominal 5 ppb
 - Measured ~ 1 ppb V(V)



Kilibarda, Afton, Harrington, Yan, and Levine, J.
Chrom. A 1304 (2013) 121-126.

Conclusions

- Vanadium speciation is possible through application of chromatographic separation
- Ion pairing reverse phase separation is appropriate for environmentally relevant species
- Mobile phase composition can be varied to optimize selectivity
- Analysis of environmental samples allows quantification to 50 ppt
- Minimal matrix effect in natural fresh water on spike recovery

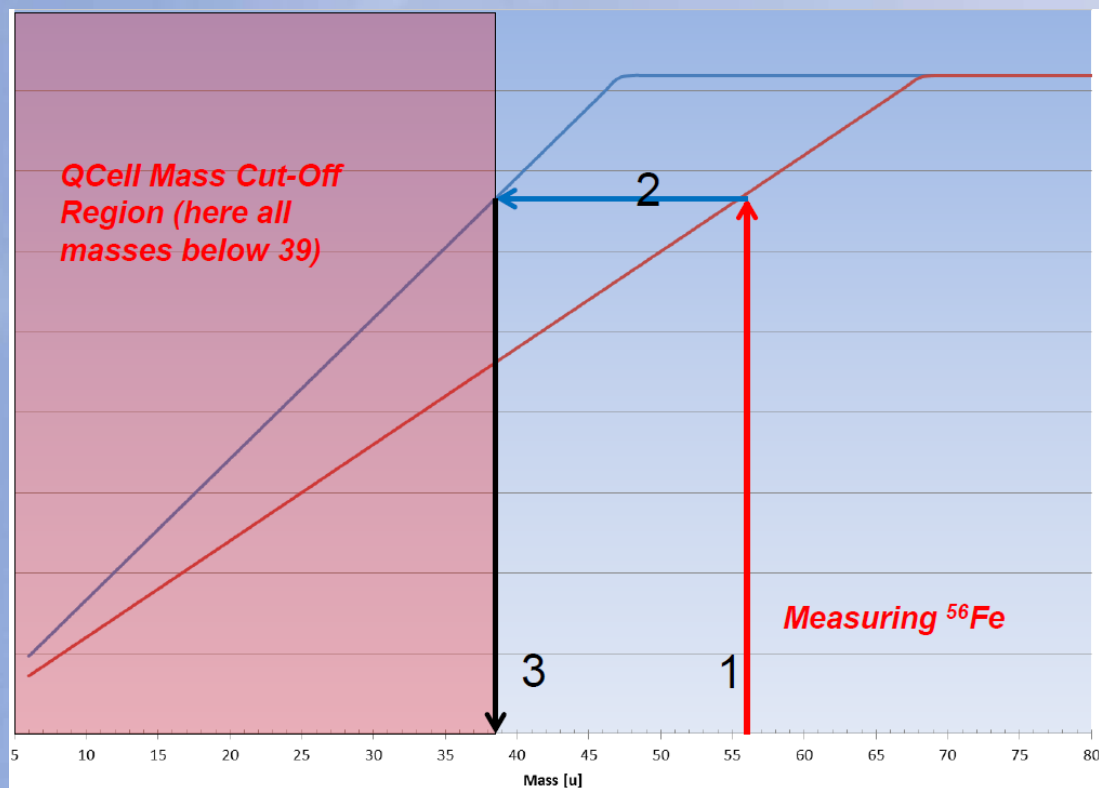


N. Kilibarda, S. Afton, J. Harrington, F. Yan, and K. Levine, "Rapid speciation and determination of vanadium compounds using ion-pair reversed-phase ultra-high-performance liquid chromatography inductively coupled plasma-sector field mass spectrometry," *J. Chrom. A*, 1304 (2013) pg 121-126.

Future directions

Future applications

- Verification of analysis on other ICP systems – X Series II, iCAP Q MS, etc



Qcell Low Mass Cutoff –
Excludes O to prevent ClO formation

- Analysis of vanadium speciation in biological fluids – urine, serum, etc
- Application of separation method to other elements - Cd, Fe, etc.

Image courtesy of: ThermoFisher Scientific

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Resources for Speciation

- **Webinars:**

- Complete Inorganic Elemental Speciation Solutions for Environmental Applications
- Bromine Speciation in Ozonated Drinking Water Using Ion Chromatography and Inductively Coupled Plasma Mass Spectrometry (June 9)

- **Speciation web site:**

- <http://www.thermoscientific.com/en/about-us/videos/speciation-using-ic-icp-ms.html>
- European Virtual Institute for Speciation Analysis:
 - www.speciation.net

- Application notes/notebook/white papers:

- Application notebook: Speciation Application Summary Ion Chromatography: Trace Elemental Species Separation and Detection
- Application note 43098: Speciation Analysis of Cr(III) and Cr(VI) in Drinking Waters
- Application note 30127: Speciation of Trace Elemental Species using GC-ICP-MS and GC-HR-ICP-MS
- White Paper: Coupling of an Inert Ion Chromatographic System with ICP-Q-MS for Robust and Accurate Elemental Speciation
- White paper: Benefits of Coupling IC with ICP/MS