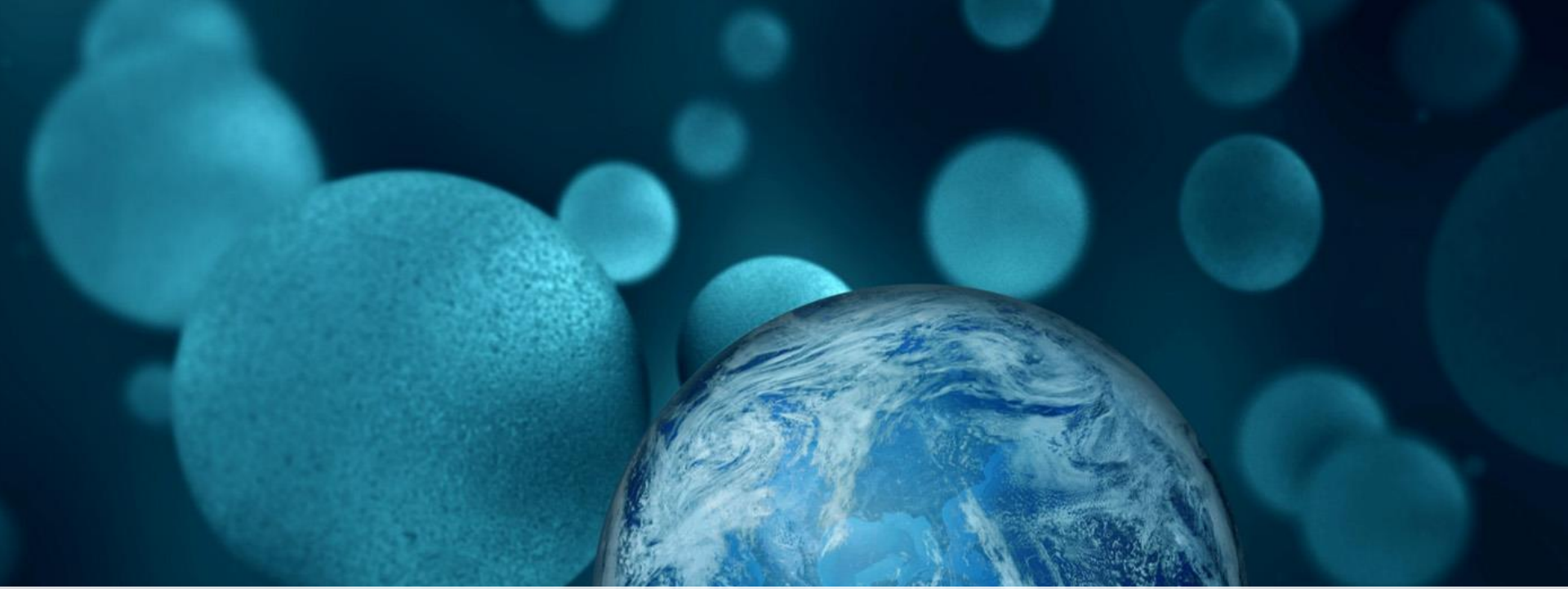

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Meeting the demands of the EU Water Framework Directive for trace element analysis

Simon Nelms, Regional Marketing Manager, TEA, EMEA

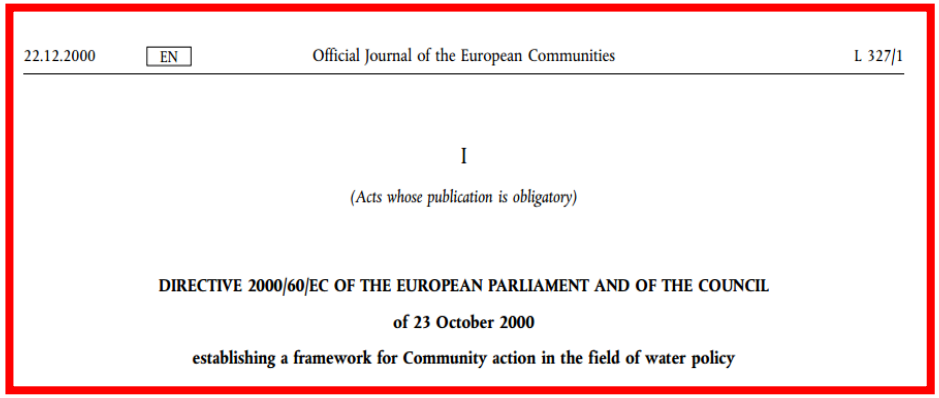
simon.nelms@thermofisher.com

Overview

- What is the EU Water Framework Directive (EU WFD)?
- Why has this legislation been developed?
- Environmental quality standards and the EU WFD
- What trace element levels need to be measured and in what samples?
- What techniques are available to make the measurements?
- Speciation and the WFD
  - What is speciation?
  - Principle steps of speciation analysis
- Application examples
  - Application 1: Multi-element analysis in drinking water using ICP-MS
  - Application 2: Chromium speciation in drinking water using IC-ICP-MS
- Resources available for learning more about trace element analysis and the WFD

What is the Water Framework Directive?

- The Water Framework Directive (Directive 2000/60/EC of the European Parliament and of the Council, implemented on 22nd December 2000) is a European Union (EU) directive which commits EU member states to:
  - Achieve good qualitative and quantitative status of all water bodies (including marine waters up to one nautical mile from shore) by 2015
  - It prescribes steps to reach the common goal, rather than adopting a limit value approach
  - In 2013, Directive 2013/39/EU was issued as an amendment to the WFD and to the intermediate Directive 2008/105/EC. This Directive defines the priority substances and their required limits



22.12.2000 EN Official Journal of the European Communities L 327/1

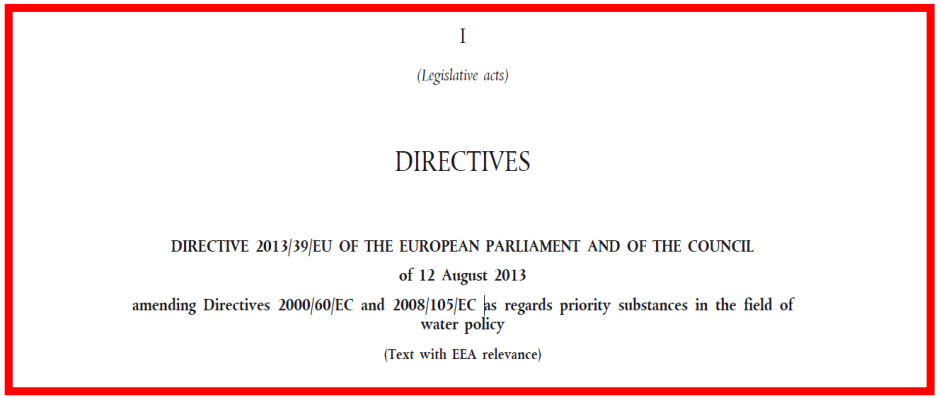
I

(Acts whose publication is obligatory)

DIRECTIVE 2000/60/EC OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL

of 23 October 2000

establishing a framework for Community action in the field of water policy



I

(Legislative acts)

DIRECTIVES

DIRECTIVE 2013/39/EU OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL

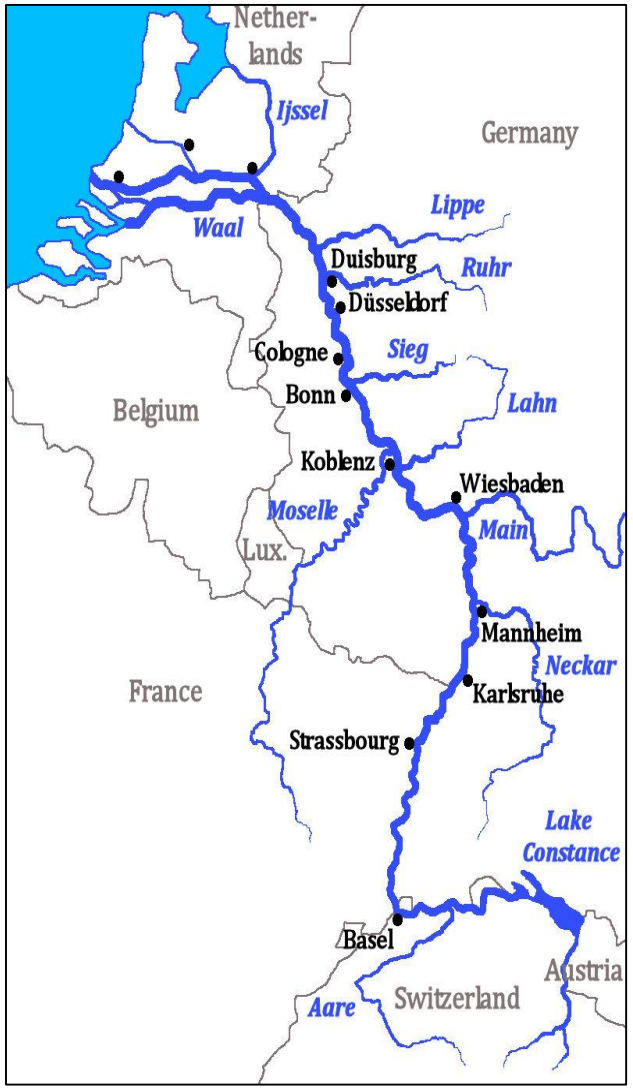
of 12 August 2013

amending Directives 2000/60/EC and 2008/105/EC as regards priority substances in the field of water policy

(Text with EEA relevance)

Why has this legislation been developed?

- To establish a legal framework to protect and restore clean water across Europe and ensure its long-term, sustainable use.
- Based on river basin districts.
- Many river basins cover more than a single country
- Where this is the case, cooperation between member states is required to ensure the quality of the river basin
- For individual water bodies, the directive defines:
  - Good ecological status
  - Good chemical status



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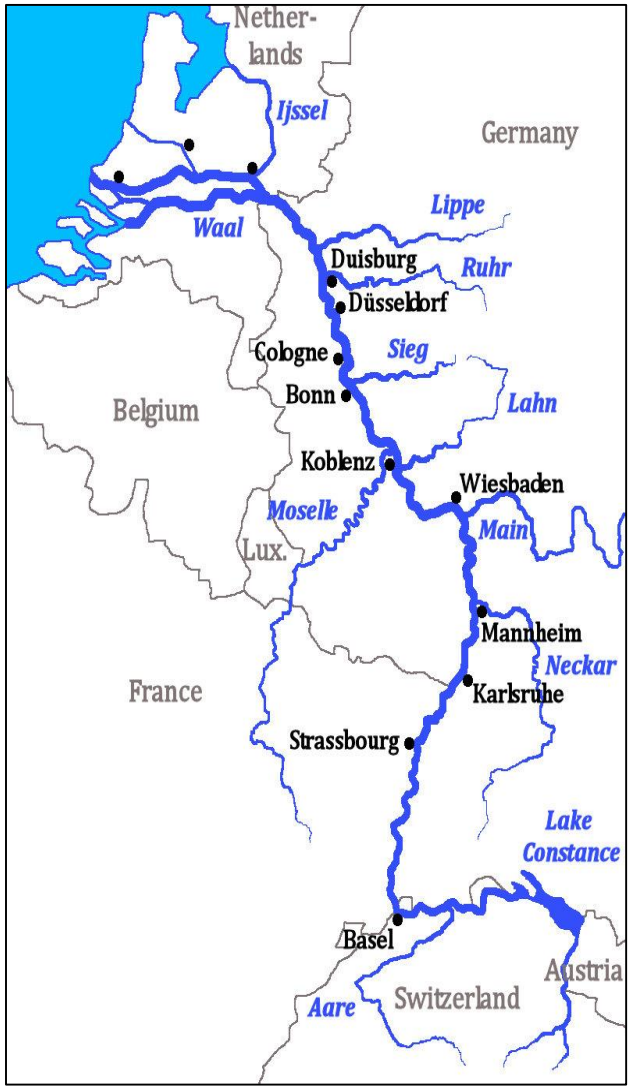
Environmental quality standards and the WFD

- EU WFD amended, together with the Directive on Environmental Quality Standards (2008/105/EC), by the Priority Substances Directive (2013/39/EU)
    - Good chemical status defined by environmental quality standards (EQSs)
    - EQSs are now set in the Priority Substances Directive
- Three different Environmental Quality Standards (EQSs)
    - AA-EQS: Annual Average
    - MAC-EQS: Maximum Allowable Concentration
    - Biota – EQS: set primarily in fish, although some are in crustaceans and molluscs
    - Where prescribed, biota-EQS take precedence over water concentrations



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What trace element levels need to be measured and in what samples?

- Priority Hazardous Substance (PHS) metals and Specific Pollutants

Element	Hardness as $CaCO_3$ (where applicable)	Annual Average (AA)		Maximum Allowable Concentration (MAC) (µg/l)
		All inland surface waters (µg/l)	All other surface waters (µg/l)	
Cd (PHS)	0 – 40	<0.08	0.2	<0.45
	40 – 50	0.08		0.45
	50 – 100	0.09		0.6
	100 – 200	0.15		0.9
	>200	0.25		1.5
Hg (PHS)	n/a	0.05		0.07
Ni	n/a	4	8.6	34
Pb	n/a	1.2	1.3	14

Element	Hardness as $CaCO_3$ (where applicable)	Annual Average (AA)	
		Rivers and fresh water lakes (µg/l)	Transitional and coastal waters (µg/l)
As	n/a	50	25
Cr III	n/a	4.7	n/a
Cr VI	n/a	3.4	0.6
Cu	0-50	1	5
	50-100	6	
	100-250	10	
	>250	28	
Fe	n/a	1000	1000
Zn	0-50	8	40
	50-100	50	
	100-250	75	
	>250	125	

What techniques are available to make the measurements?

Atomic absorption spectrometry (AA)

Flame AA

Graphite furnace AA

Inductively coupled plasma – optical emission spectrometry (ICP-OES)

Axial / radial view ICP-OES

Inductively coupled plasma – mass spectrometry (ICP-MS)

Quadrupole ICP-MS

High resolution ICP-MS

Increasing performance and investment, decreasing detection limit capability

Tributyltin and Cr(III)/Cr(VI) are specified in the EU WFD. Tributyltin can be measured using GC-MS or GC-ICP-MS, Cr(III)/Cr(VI) can be measured colorimetrically, but IC-ICP-MS is more sensitive and accurate

SPECIES SEPARATION

DETECTION

Liquid Chromatography
(Ion Chromatography or HPLC)

Gas Chromatography

ICP-MS

Tributyltin species species – GC-ICP-MS

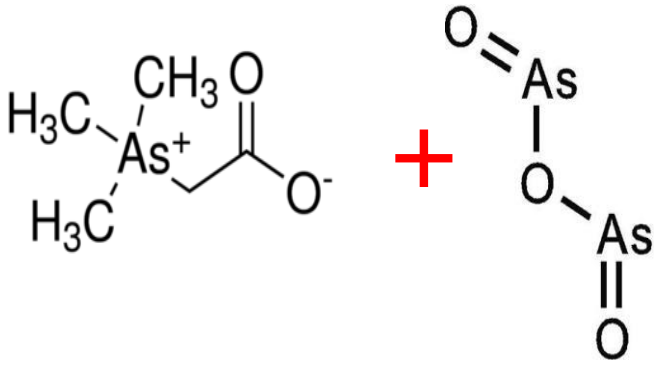
Cr(III) and Cr(VI) species – IC-ICP-MS

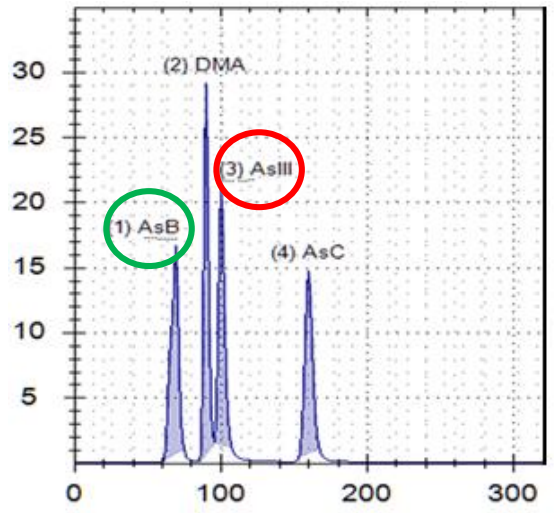
What is speciation?

- **Definition** - 'the analytical activity of identifying and/or measuring the quantities of one or more individual chemical species in a sample' **IUPAC**
- **That means** – chemical species of an element are determined rather than the total element concentration
- **Example** – Arsenobetaine and As (III) species separated by ion chromatography (IC) and detected by ICP-MS

Dimethylarsinic acid (DMA) and arsenocholine (AC) peaks also shown

Arsenobetaine (AsB) **As (III)**





- **Speciation analysis is important as it reveals valuable information about elemental bioavailability, mobility, metabolism and toxicity**

Principle steps of speciation analysis

Extraction → **Separation** → **Detection**

Sample containing various compounds

✓Preservation of original species distribution

Challenges:

✘Loss of species during sample preparation

✘Transformation of species

Different separation mechanisms

Chromatography:

Analyte retention is achieved by interaction of charges (IC) or molecules (LC, GC) with stationary phase, separation by passage of mobile phase through the column

ICP-MS detects the element enclosed in a species

Accurate and reliable quantification of different compounds containing the same element

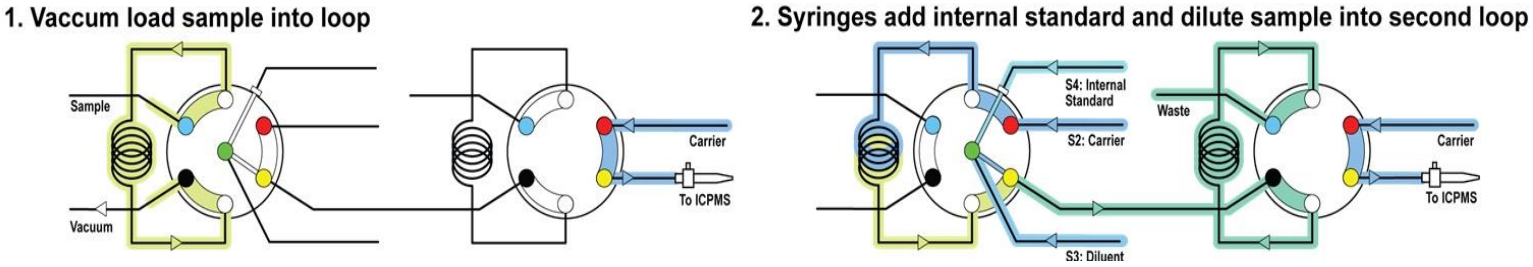
Application 1: Multi-element analysis in drinking water using ICP-MS

- Tap water samples collected in an HDPE tank, acidified to 1% v/v HNO_3
- Standards and quality control (QC) solutions prepared according to the requirements of EPA 200.8
- 320 tap water samples analyzed according to method EPA 200.8, with a Continuing Calibration Verification (CCV) QC sample and a Laboratory Fortified Blank (LFB) analyzed every 10 samples
- ^{6}Li, ^{45}Sc, ^{71}Ga, ^{89}Y, ^{115}In and ^{159}Tb internal standards used, added on-line
- Au added to all blanks, standards and samples at 100 µg/L to aid Hg washout
- ICP-MS coupled with an auto-dilution system and an autosampler; autodilution sample loop volume = 1.5 mL
- Thermo Scientific™ iCAP™ RQ ICP-MS used. Operated in KED collision cell mode for all analytes, using pure He cell gas
- **Analysis time = 66 s per sample**

Data collection and autodilution control

- Thermo Scientific™ Qtegra™ Intelligent Scientific Data Solution™ (ISDS) software used for quantitative evaluation of the data
- Pre-defined EPA 200.8 method template employed - user only needs to enter the number of samples to be analyzed
- All parameters required to comply with EPA 200.8 automatically checked
- Samples that do not meet all criteria, e.g. internal standard recovery or over-range analyte concentrations, automatically diluted to an appropriate level as calculated or defined within the software and the measurement automatically repeated

Autodilution process



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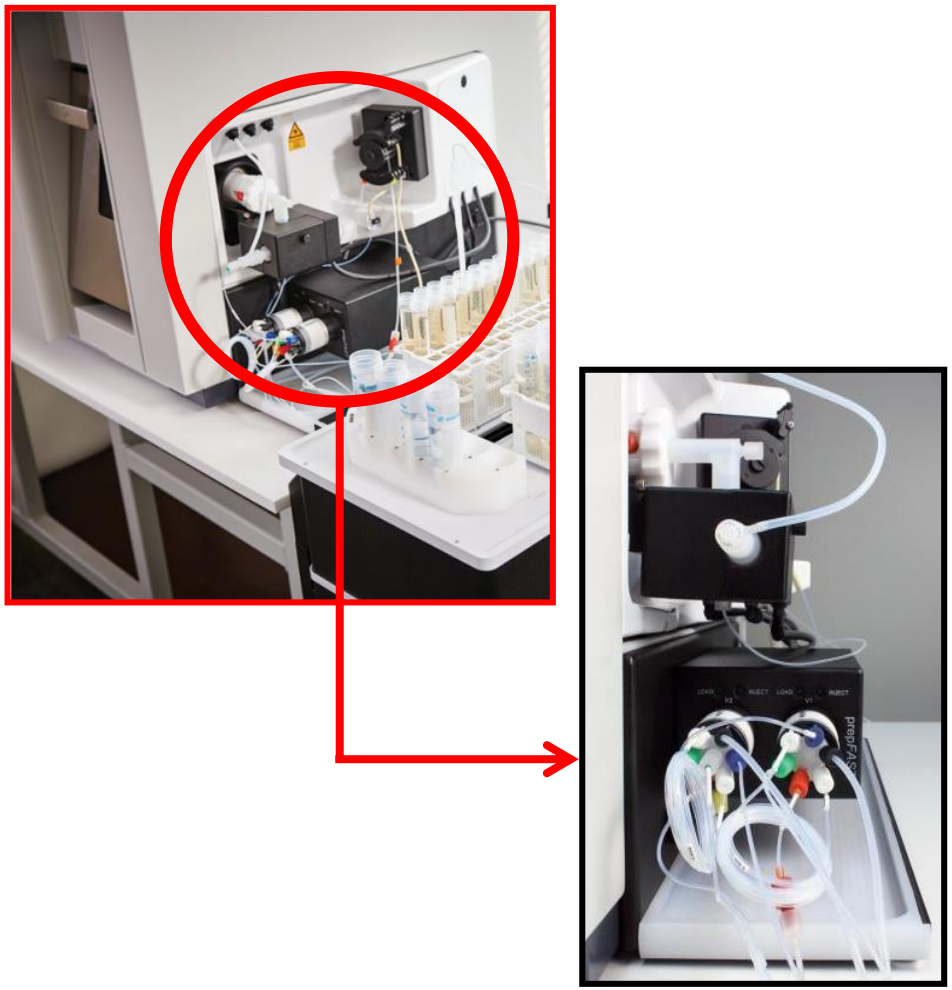
Auto-dilution system – principle of operation



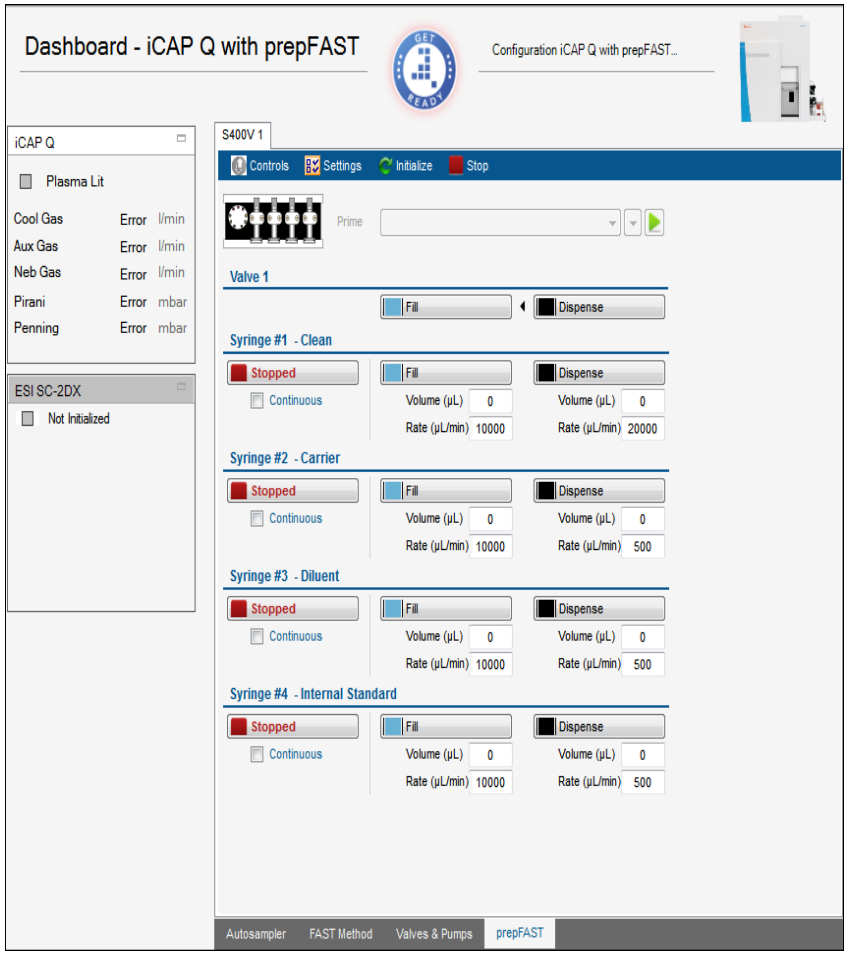
- Elemental Scientific prep*FAST* used
- It is a dual FAST valve system:
  - Samples are loaded on to the loop in the first valve where syringe pumps accurately dilute / add internal standard to the sample
  - Dilution factors of up to 400x can be accurately and reproducibly applied
  - The diluted sample is then passed to the second loop for FAST valve transfer to the ICP-MS
  - Single standalone system, ICP-MS supplier independent
- Control of the prep*FAST* is provided in the ICP-MS's software

Qtegra ISDS prep*FAST* auto-dilution capabilities

- **Qtegra ISDS Software provides complete software control of the prep*FAST***

- Automated ***prescriptive*** dilution for preparation of:
  - Samples
  - Standards

- Automated ***intelligent*** dilution:
  - Over calibration range auto-dilution
  - Internal standard range auto-dilution

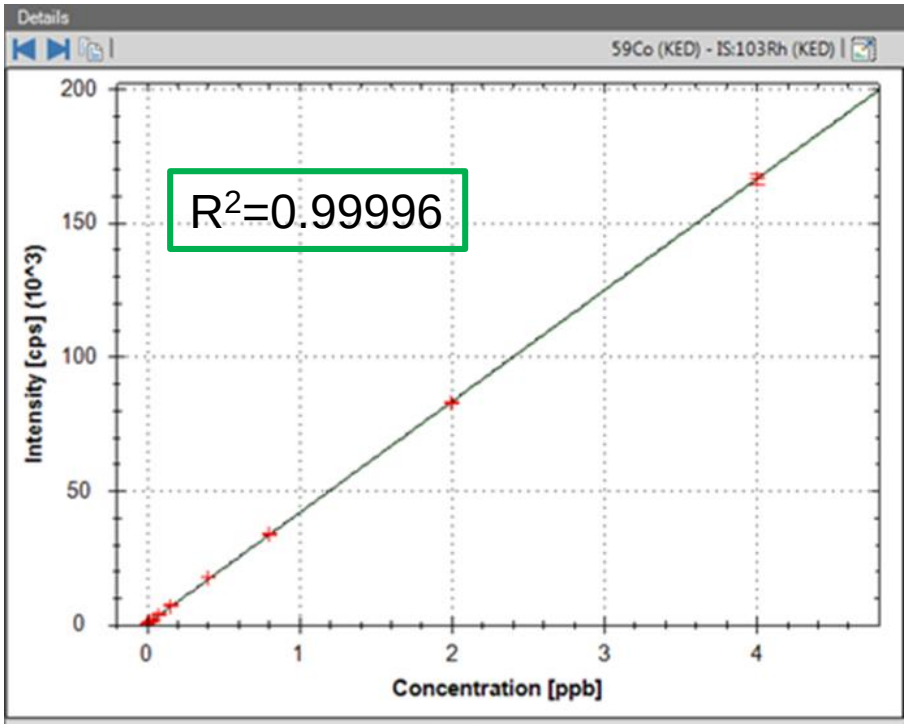


Standard preparation using auto-dilution

- A single standard stock (Rack 3, Vial 2) is used to generate a 10 point calibration (from 0.01 to 4 ppb) with dilution factors from 1 to 400

- Dual stocks can be used to extend automated calibration ranges even further

	Label	Sample Type	Standard	Rack	Vial	prepFAST DF
1	Blank	AVERAGE BLK		3	1	1
2	Blank	AVERAGE BLK		3	1	1
3	Blank	AVERAGE BLK		3	1	1
4	0.010 ppb	STD	4 ppb Stock	3	2	400
5	0.013 ppb	STD	4 ppb Stock	3	2	300
6	0.020 ppb	STD	4 ppb Stock	3	2	200
7	0.040 ppb	STD	4 ppb Stock	3	2	100
8	0.080 ppb	STD	4 ppb Stock	3	2	50
9	0.160 ppb	STD	4 ppb Stock	3	2	25
10	0.4 ppb	STD	4 ppb Stock	3	2	10
11	0.8 ppb	STD	4 ppb Stock	3	2	5
12	2 ppb	STD	4 ppb Stock	3	2	2
13	4 ppb	STD	4 ppb Stock	3	2	1



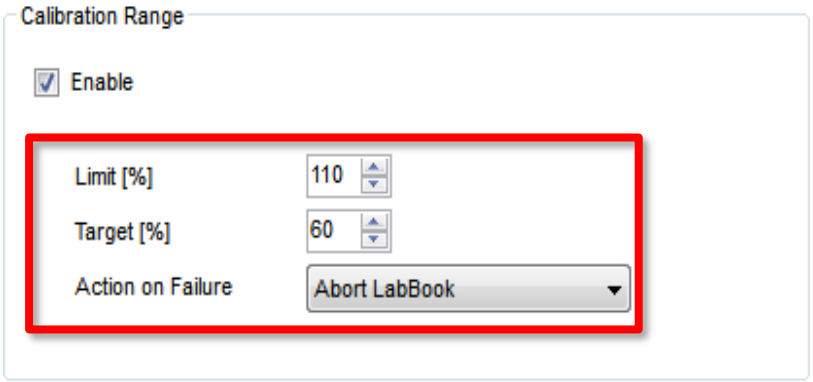
f(x) = b*x + a

Parameter	Value	Confidence Delta at 90%	StdErr	Rel. StdErr [%]
b	41,540.313	+/-103.5	75.423	0.182
a	141.069	+/-0.0	0.000	0.000
R^2	0.99996			
BEC	0.0034 ppb			
IDL(LOD)	0.0001 ppb			

Advanced auto-dilution: Calibration over-range

- The user defines:
  - The maximum allowed over-range limit as a percentage of the top calibration standard, e.g. 110%
  - The target concentration of the analyte after auto-dilution, e.g. 60%

✓ Everything else is done by the software



	Label	Status	Sample Type	Rack	Vial	prepFAST DF	Standard	Total Dilution Factor
1	Blank		BLK	1	1	1		
2	Level 1		STD	1	2	100	Tune B	
3	Level 2		STD	1	2	10	Tune B	
4	Level 3		STD	1	2	2	Tune B	
5	Over-range		UNKNOWN	1	2	1		
6	Over-range		UNKNOWN	1	2	3.367		
7	Washout		QC	1	1	1		1

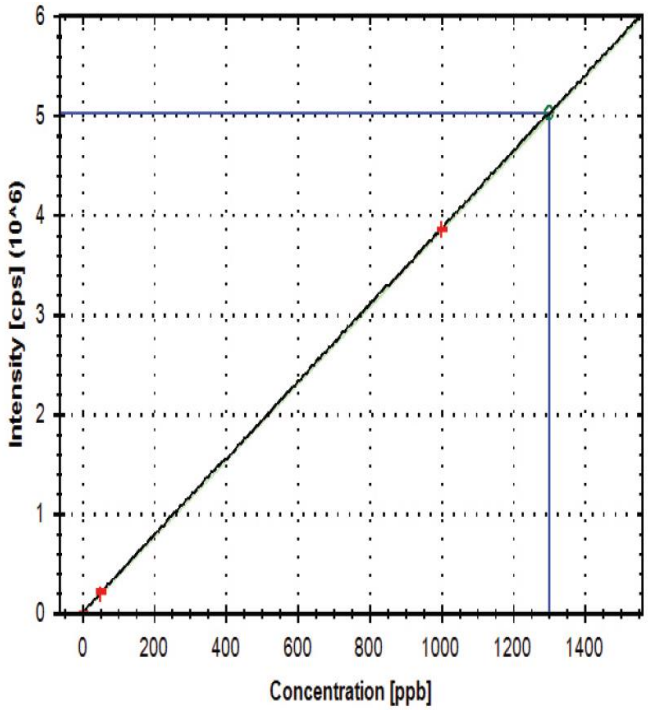
Calibration line generated by auto-dilution from a stock solution in the autosampler rack

Automatically added Analysis

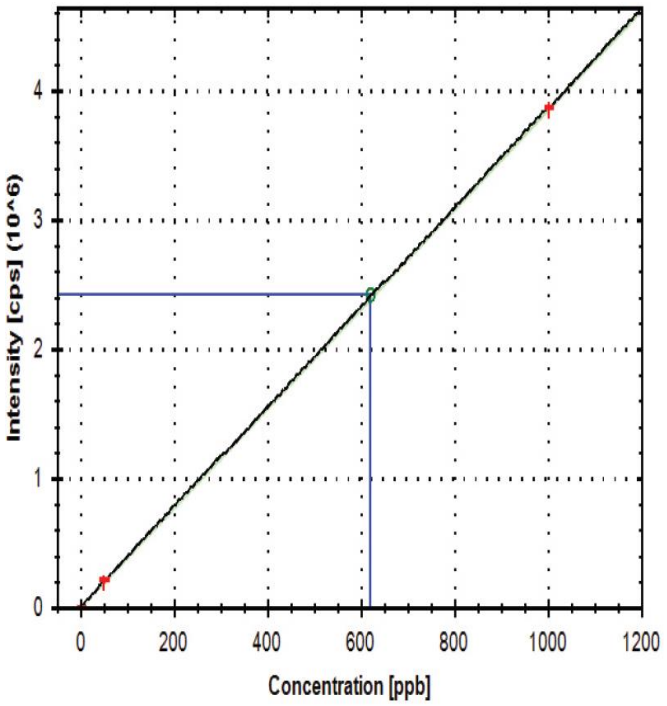
Automatically defined dilution factor

Automatic dilution of an over-range sample in the analysis

Original measured result – above the top calibration standard (over-range)



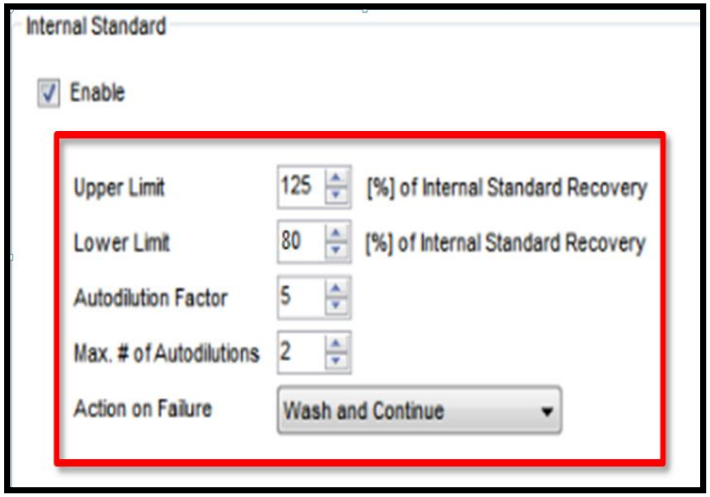
Automatically re-measured with calculated dilution factor of 2.2



- When an analyte exceeds the calibration range, intelligent auto-dilution calculates the required dilution factor as defined by the criteria in the software
- Then dilutes the sample and re-measures only the affected analytes, without manual interaction

Advanced auto-dilution: Internal standard recovery

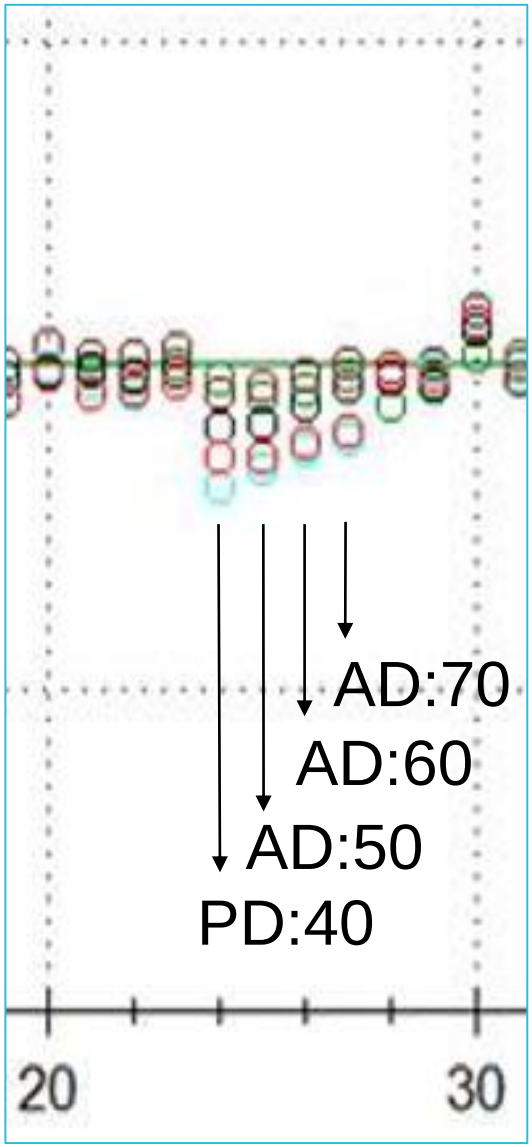
- The user defines:
  - The range of acceptable internal standard recovery, e.g. 80 - 125%
  - The autodilution factor step and maximum number of dilutions before moving on to the next sample
  - ✓ Everything else is done by the software



No	Time	Sample Type	Label	89Y	115In	175Lu
2	6/17/2014 7:37:54 PM	BLK		100.0%	100.0%	100.0%
3	6/17/2014 7:39:10 PM	STD				
22	6/17/2014 8:00:57 PM	UNKNOWN	3% NaCl + 10 ppb Spike	13.5%	10.2%	7.1%
23	6/17/2014 8:01:46 PM	UNKNOWN	3% NaCl + 10 ppb Spike	76.0%	68.2%	62.8%
26	6/17/2014 8:04:32 PM	UNKNOWN	3% NaCl + 10 ppb Spike	90.9%	84.7%	85.2%

A sample of 3% NaCl is analyzed directly

The 3% NaCl sample is automatically diluted until the Internal Standard Recovery is within the defined limits



Practical example of autodilution - analysis of variable matrix saline samples

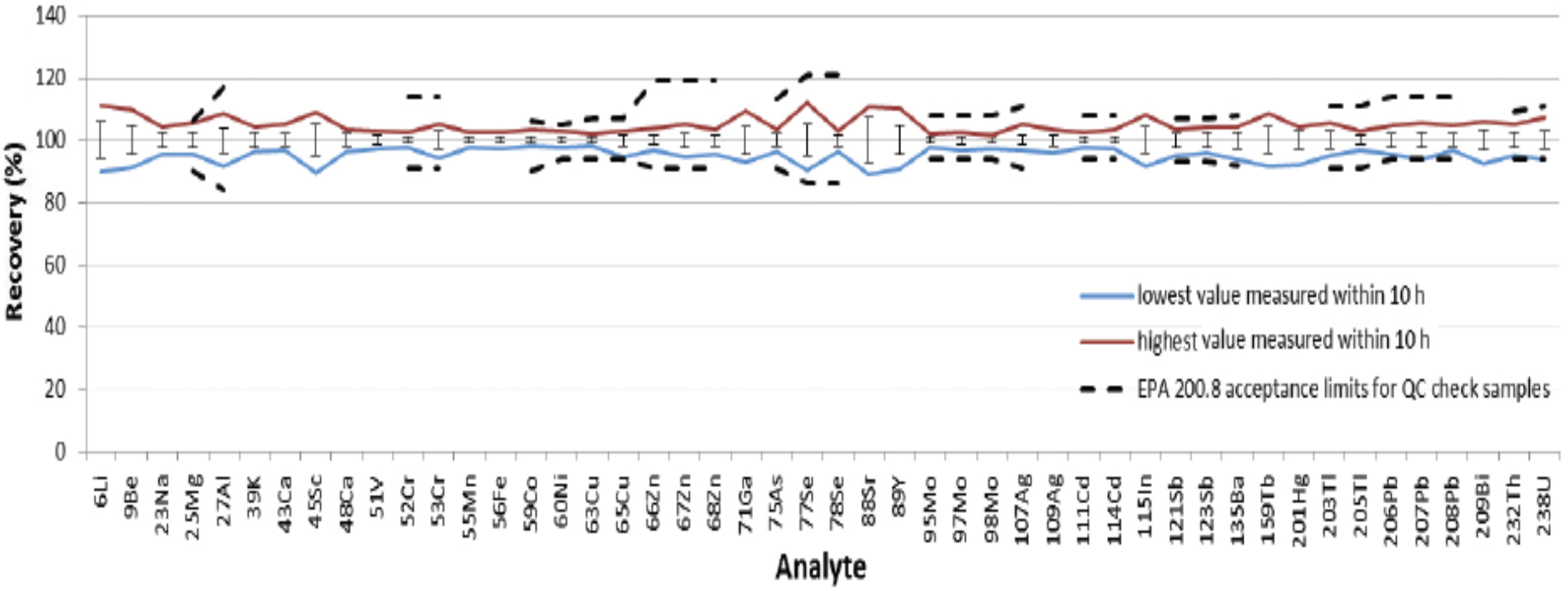
- A close look at sample 24
  - > 8 % NaCl in this sample!
  - Required internal standard recovery was 85-125%
  - Prescriptive dilution (PD) at 40 fold
  - Autodilution (AD) steps:
    - 1 → autodilution to 50 fold
    - 2 → autodilution to 60 fold
    - 3 → autodilution to 70 fold
  - After the 3rd dilution, the internal standard recovery is within the specified range → analysis can continue

Method detection limits (MDL's) vs required annual average maximum concentration

Element	Isotope	MDL (µg/L)	Required annual average maximum concentration (µg/L)
As	75	0.011	25 - 50
Cd	111	0.017	< 0.08 – 0.25
Cr	52	0.022	Cr(III): 4.7, Cr (VI): 0.6 – 3.4
Cu	63	0.119	1 - 28
Fe	56	0.125	1000
Hg	**202**	**0.110**	**0.05**
Ni	60	0.075	4.0 – 8.6
Pb	206+207+208	0.007	1.2 – 1.3
Zn	66	0.175	8 - 125

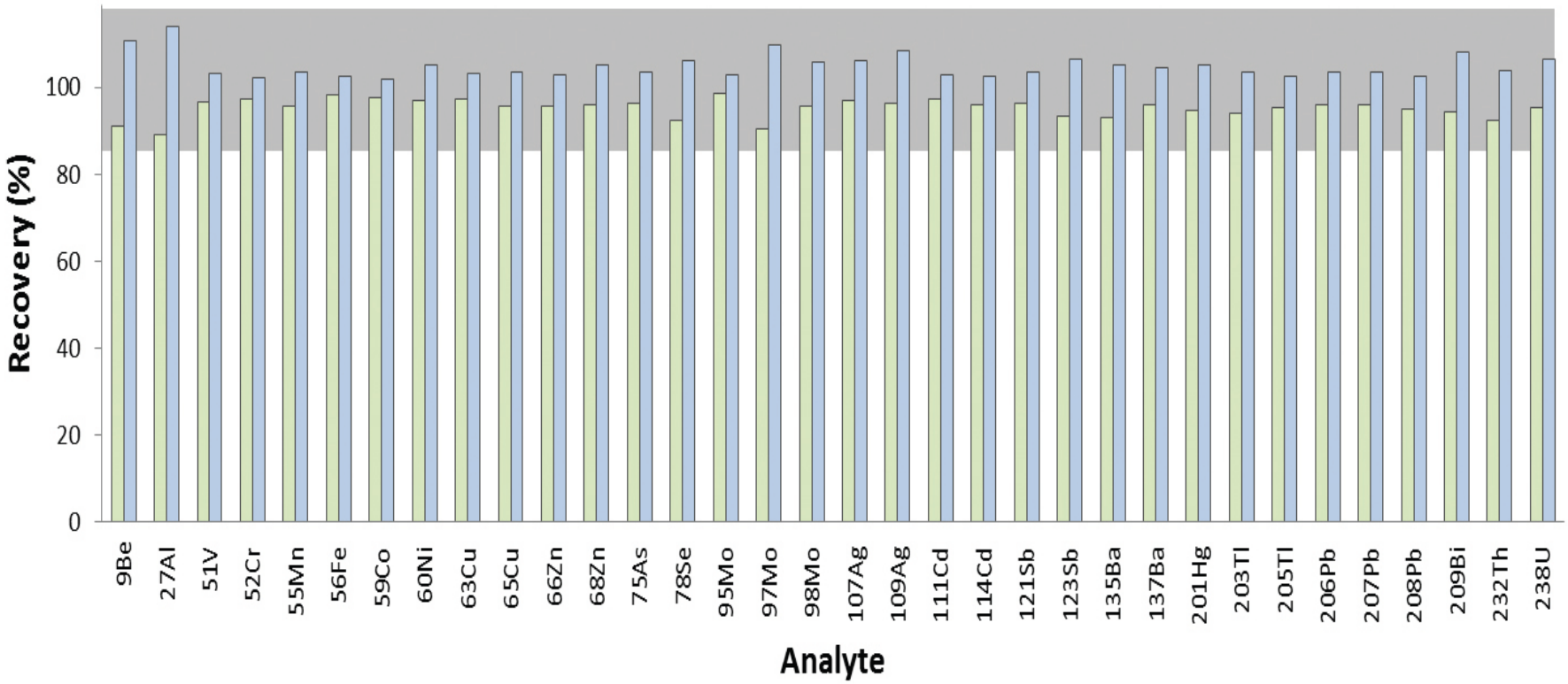
- During the analysis run, a Laboratory Fortified Blank (LFB) QC sample was analyzed a total of 32 times
- Spike level of the LFB = 5x the estimated detection limit for each analyte
- Average value of the LFB results determined together with the standard deviation
- MDL's shown above calculated as 3.14 x the standard deviation, all except Hg lower than the required limit
- **Hg exceeded required detection limit due to memory effects, even with Au added to improve washout. Likely that a dedicated Hg analysis would be required to routinely achieve the required detection limit**

Average recovery and stability of the continuous calibration QC samples over the run



- Continuing Calibration Verification (CCV) QC sample analyzed every 10 samples
- Total number of CCV QC analyses = 32
- EPA 200.8 requires that this QC must be within +/- 10% or within the acceptance limits of the method (EPA 200.8, rev 5.5)
- All elements found to be accurate to within ± 10% of the known concentration, as well as the acceptance criteria and were stable over the whole run

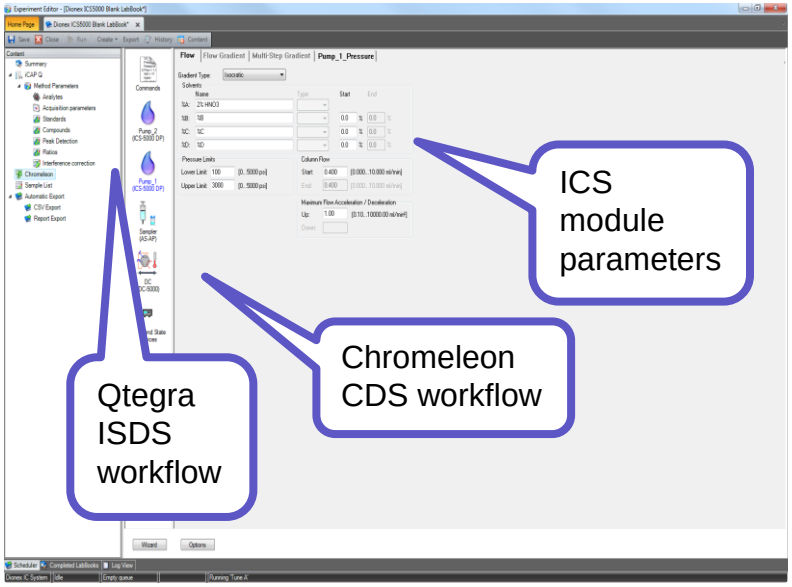
Average recovery and stability of the laboratory fortified blank QC samples over the run



- During the analysis run, a Laboratory Fortified Blank (LFB) QC sample was analyzed every 10 samples
- Total number of LFB QC analyses = 32
- All elements spiked at 5 µg/L except Fe, Zn, Cu and Ba (50 µg/L) and Hg (1 µg/L)
- EPA 200.8 requires that this QC must provide recoveries of 85 to 115%; all elements met this criteria throughout the whole run

Application 2: Chromium speciation in drinking water using IC-ICP-MS

- Thermo Scientific ™ Dionex™ ICS-5000+ Capillary HPIC™ coupled to an iCAP RQ ICP-MS
- Cr(III) and Cr(VI) mixed calibration standards prepared from blank to 15 ng/g
- Detection limit determined for each species
- Spike recovery in a tap water matrix evaluated
- Local (Bremen) potable water sample measured
- All data collected and processed using Qtegra ISDS software with an integrated Chromeleon plug-in
- The Chromeleon plug-in allowed control of the ICS-5000 and configuration of the IC method directly from Qtegra



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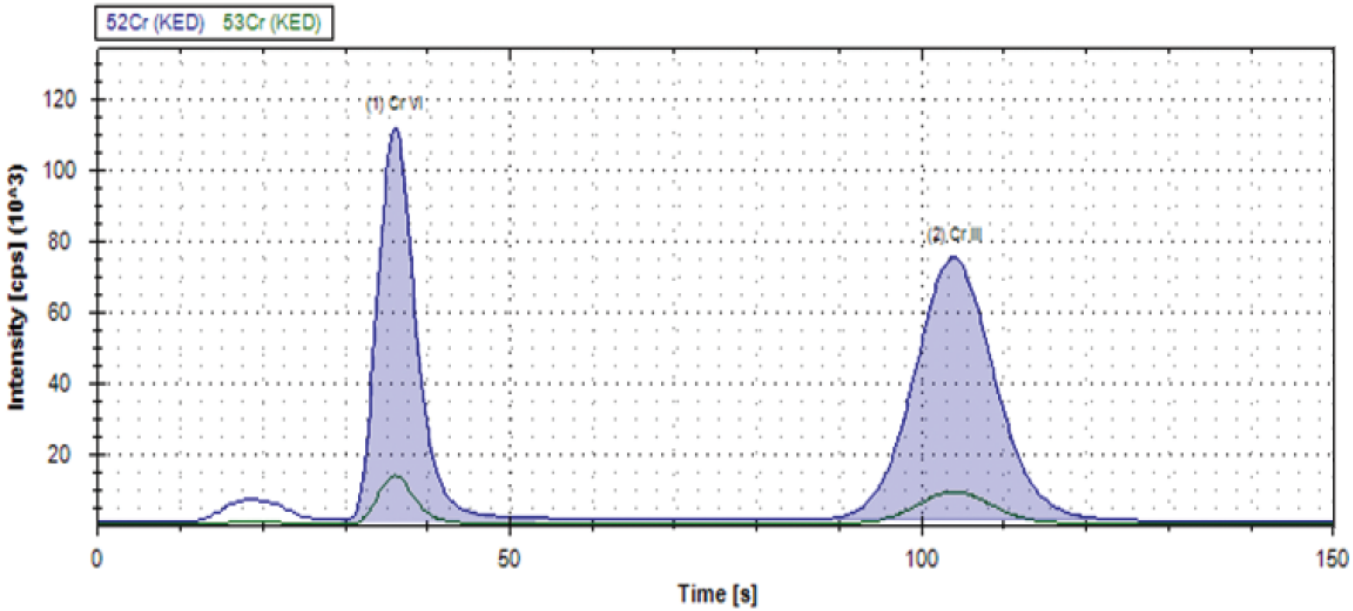
ICP-MS

Parameter	Value
Forward power	1550 W
Nebulizer gas	0.80 L/min
Injector	2 mm I.D.
Cell gas flow / KED voltage	4.8 mL/min He / 2V
Dwell time	100 ms

Ion Chromatograph

Column	Dionex AG-7 (2 mm i. D., 50 mm length)
Elution	Isocratic
Mobile phase	0.4 mol/L HNO_3
Flow rate	400 µL/min
Injection volume	20 µL
Duration	150 s



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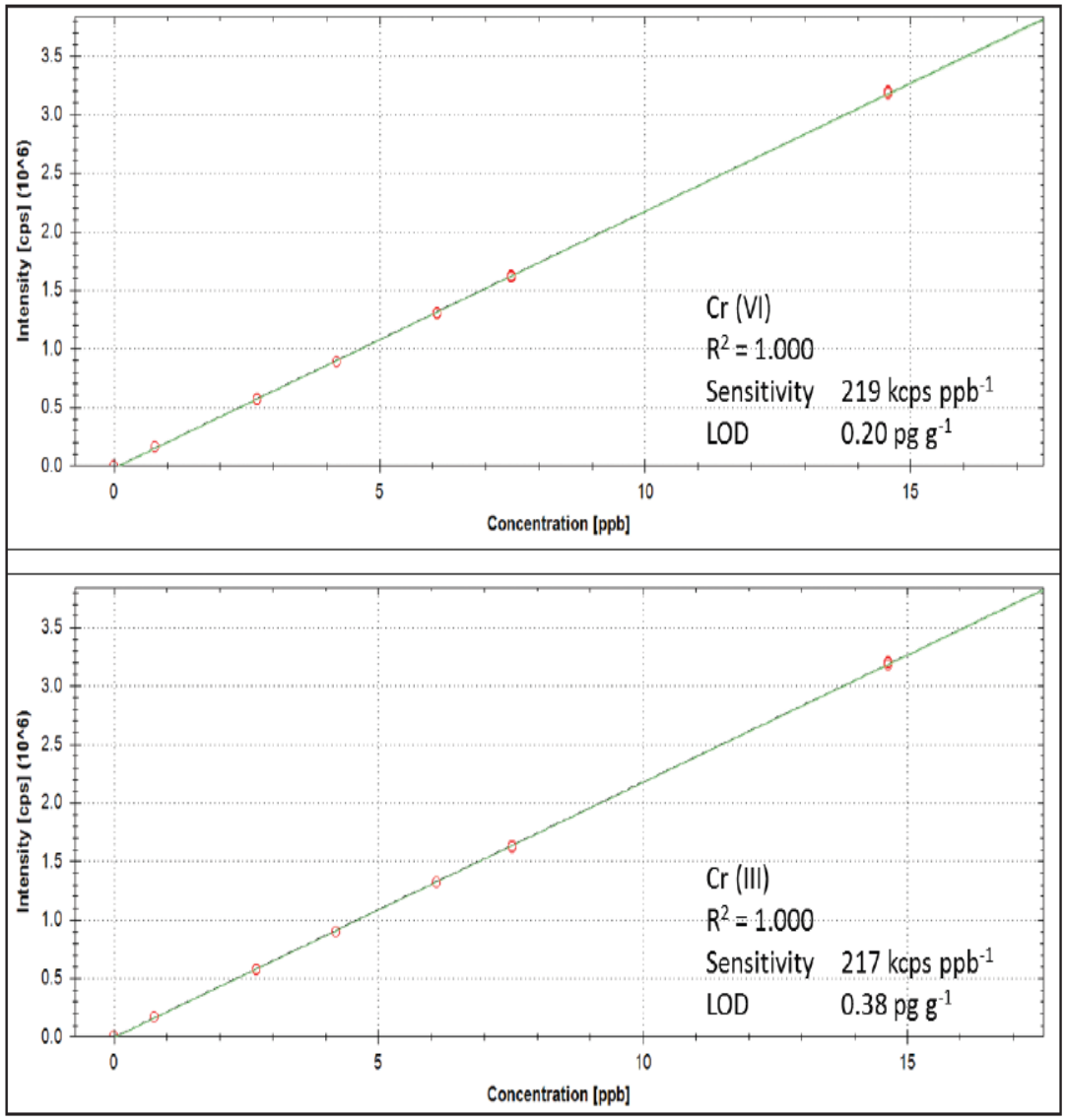
Cr species calibration



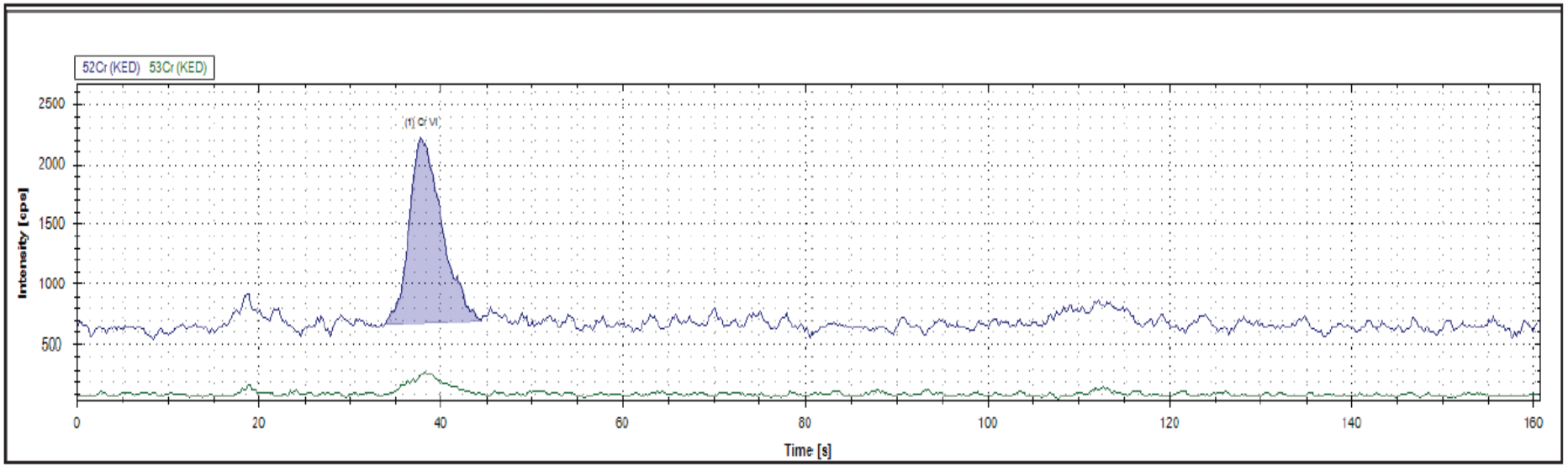
- Good linearity obtained
- Detection sensitivity determined to be 220 kcps / ng/g for both Cr species
- Proves that Cr(VI) stability is unaffected by the 0.4 mmol/L HNO_3 matrix used
- <0.5 pg/g detection limits achieved for both species
- Lowest level required for EU WFD = 600 pg/g for Cr(VI) in transitional and coastal waters

Cr(III) and Cr(VI) spike recovery results

- Mixtures of both Cr species were quantified against the calibrations in a spike recovery test
- Three spike levels tested:
  - 2.34 ng/g of each species
  - 6.03 ng/g Cr(VI) and 1.90 ng/g Cr(III)
  - 1.87 ng/g Cr(VI) and 6.20 ng/g Cr(III)
- Each sample analyzed in triplicate; quantitative recoveries obtained for both species in all samples

Conc. spiked [ng/g]	Cr (VI)		Cr (III)	
	Found (ng/g)	Recovery (%)	Found (ng/g)	Recovery (%)
2.34 of each	2.31 ± 0.01	99 ± 1	2.35 ± 0.02	100 ± 1
6.03 Cr (VI); 1.90 Cr (III)	6.01 ± 0.02	100 ± 1	2.00 ± 0.01	105 ± 1
1.87 Cr (VI); 6.20 Cr (III)	1.85 ± 0.01	99 ± 1	6.15 ± 0.03	99 ± 1

Analysis of a local (Bremen) potable water sample



Species	Measured concentration (pg/g))
Cr (III)	not detected
Cr (VI)	42.5 ± 1

Conclusions

- Quadrupole ICP-MS shown to meet the requirements of the EU Water Framework Directive for all the specified elements, except Hg
  - The required maximum annual average concentration for Hg (0.05 µg/L) is challenging due to memory effects even with Au added to improve washout
  - Dedicated separate analysis required to routinely achieve the required Hg level
- IC-ICP-MS shown to be effective for the speciation of Cr(III) and Cr(VI) in water samples at the levels required for the EU WFD

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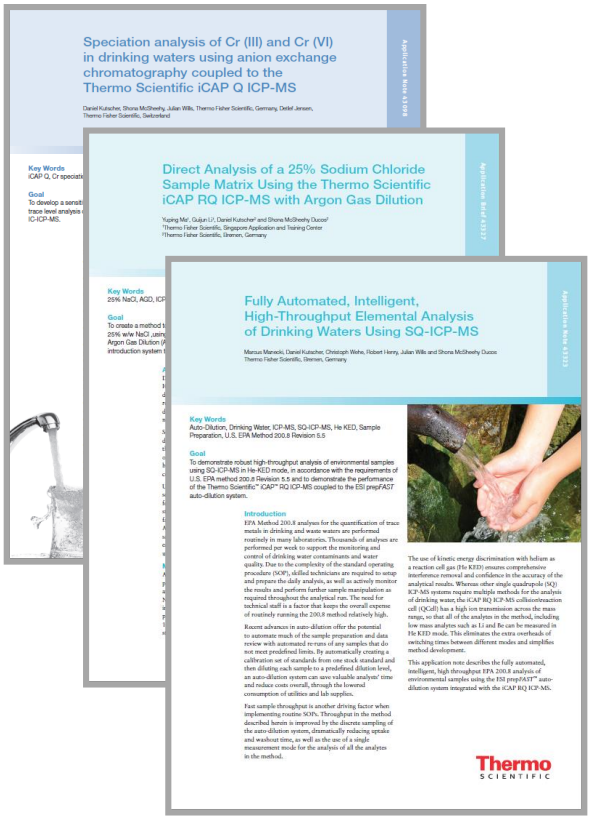
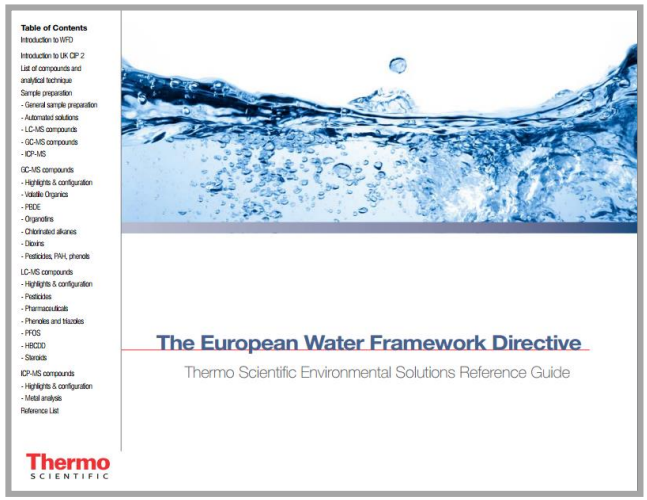
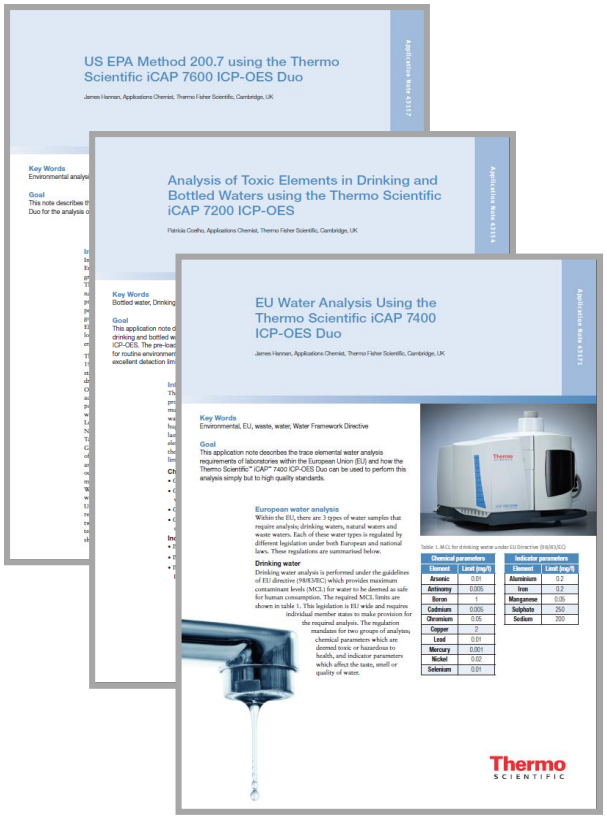
Available environmental analysis application notes and resources







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