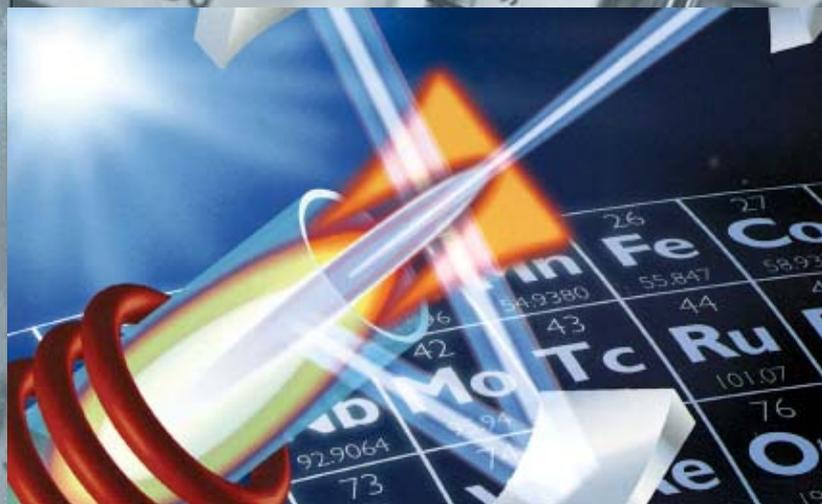


Application of ICP techniques for food analysis

Dr. FUNG Wai Hong
Government Laboratory

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Outline

- Determination of trace elements and minerals in food – an overview
- ICP for food analysis: basic principle, advantage, practical importance
 - ICP-OES
 - ICP-MS
- Future perspectives



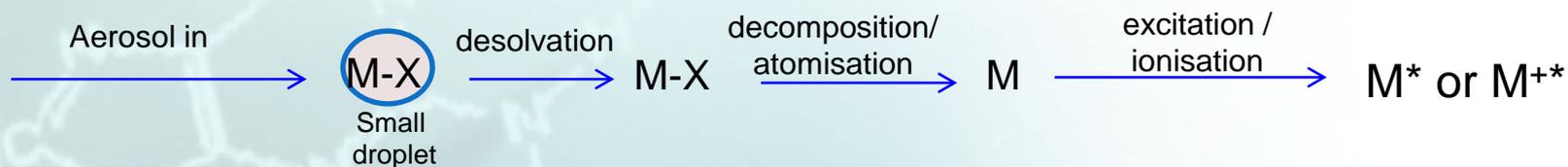
Elements in Food

- Amount of different elements in foodstuff –
 - Nutritional value: essential minerals: Ca, K, P.....
 - Safety: toxic elements: Cd, Hg, Pb.....
- Common measurement protocol:
 - Sample dissolution – e.g. acid digestion, hydrolysis...
 - Spectroscopic analysis
 - Optical spectroscopy: atomic absorption, atomic emission, optical emission spectroscopy (ICP, MIP..)
 - Mass spectrometry (ICP)



Inductively Coupled Plasma

- Inductively Coupled Plasma (ICP) - a plasma energized by inductively heating the flowing gas (Ar), contains a sufficient concentration of ions and electrons to make the gas electrically conductive
- the hot plasma (6000 to 10000K) produces excited atoms and ions



ICP Techniques

- ICP-optical (atomic) emission spectroscopy, ICP-OES
 - excited atoms and ions emit electromagnetic radiation at wavelengths characteristic of a particular element
 - intensity of this emission is indicative of the concentration of the element within the sample.
- ICP-mass spectrometry, ICP-MS
 - Ions from the plasma are extracted into a mass spectrometer
 - The ions are separated on the basis of their mass-to-charge ratio and a detector receives an ion signal proportional to the concentration.



ICP - Application

- ICP-OES / ICP-MS:
 - Examination of elements from % to sub-ppb levels
- Using for the determination of elements in various matrices
 - Environmental analysis – elements in air, water, soil, waste...
 - Material analysis – e.g. toxic elements in plastic
 - Food analysis
- Adopted by many International and National Organizations as Standard Methods for the determination of elements in general and specific food matrices



AOAC (1)

- **AOAC Official Method 984.27 (Final action 1986)** -- Calcium, Copper, Iron, Magnesium, Manganese, Phosphorus, Potassium, Sodium, and Zinc in Infant Formula: Inductively Coupled Plasma Emission Spectroscopic Method
- **AOAC Official Method 2006.03 (Final action 2009)** -- Arsenic, Cadmium, Cobalt, Chromium, Lead, Molybdenum, Nickel, and Selenium in Fertilizers: Microwave Digestion and Inductively Coupled Plasma Optical Emission Spectrometry
- **AOAC Official Method 2011.14 (Final action 2013)** -- Calcium, Copper, Iron, Magnesium, Manganese, Potassium, Phosphorus, Sodium, and Zinc in Fortified Food Products: Microwave Digestion and Inductively Coupled Plasma-Optical Emission Spectrometry



AOAC (2)

- **AOAC Official Method 2011.19 (Final action 2014)** -- Chromium, Selenium, and Molybdenum in Infant Formula and Adult Nutritional Products: Inductively Coupled Plasma-Mass Spectrometry (ISO/IDF–AOAC Method*)
- **AOAC Official Method 2012.15 (Final action 2015)** -- Total Iodine in Infant Formula and Adult/Pediatric Nutritional Formula: Inductively Coupled Plasma-Mass Spectrometry (ISO/IDF–AOAC Method*)
- **AOAC Official Method 2015.01 (First action 2015)** -- Heavy Metals in Food: Inductively Coupled Plasma–Mass Spectrometry
- **AOAC Official Method 2015.06 (First action 2015)** -- Minerals and Trace Elements in Infant Formula and Adult/Pediatric Nutritional Formula: ICP/MS Method



ISO



- **ISO20649:2015** “Infant formula and adult nutritionals -- Determination of chromium, selenium and molybdenum -- Inductively coupled plasma mass spectrometry”
- **ISO 20647:2015** “Infant formula and adult nutritionals -- Determination of total iodine -- Inductively coupled plasma mass spectrometry”
- **ISO 21033:2016** “Animal and vegetable fats and oils -- Determination of trace elements by inductively coupled plasma optical emission spectroscopy”



- **EN 15111:2007** -- Foodstuffs. Determination of trace elements. Determination of iodine by ICP-MS (inductively coupled plasma mass spectrometry)
- **EN 15763:2009** -- Foodstuffs. Determination of trace elements. Determination of arsenic, cadmium, mercury and lead in foodstuffs by inductively coupled plasma mass spectrometry (ICPMS) after pressure digestion
- **EN 15765:2009** -- Foodstuffs. Determination of trace elements. Determination of tin by inductively coupled plasma mass spectrometry (ICPMS) after pressure digestion
- **EN 16801:2016** -- Foodstuffs. Determination of elements and their chemical species. Determination of methylmercury in foodstuffs of marine origin by isotope dilution GC-ICP-MS
- **EN 16802:2016** -- Foodstuffs. Determination of elements and their chemical species. Determination of inorganic arsenic in foodstuffs of marine and plant origin by anion-exchange HPLC-ICP-MS
- **DC EN 16943 (Draft)** -- Foodstuffs. Determination of elements and their chemical species. Determination of minerals by ICP-OES



US FDA

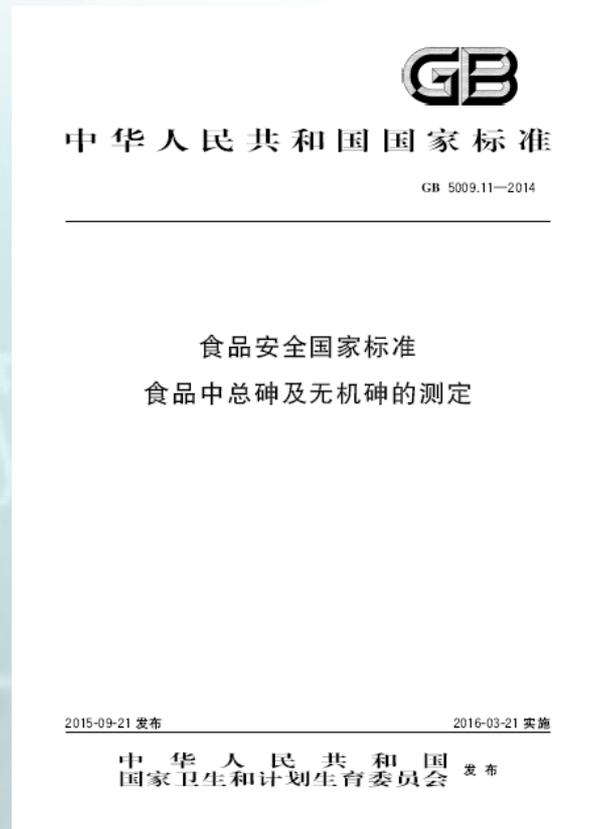
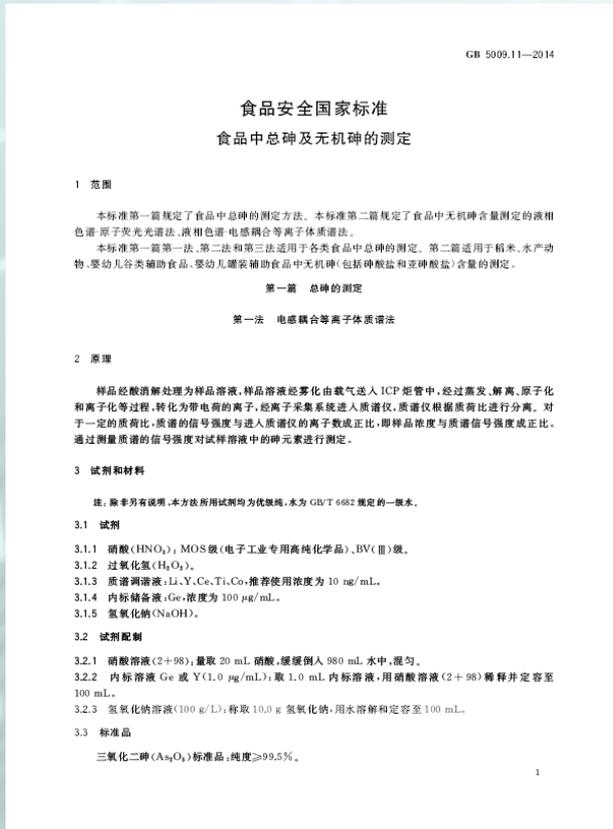


- **EAM4.4** “Inductively Coupled Plasma - Atomic Emission Spectrometric Determination of Elements in Food Using Microwave Assisted Digestion”
- **EAM4.7** “Inductively Coupled Plasma - Mass Spectrometric Determination of Arsenic, Cadmium, Chromium, Lead, Mercury, and Other Elements in Food Using Microwave Assisted Digestion”
- **EAM4.8** “High Pressure Liquid Chromatographic - Inductively Coupled Plasma - Mass Spectrometric Determination of Methylmercury and Total Mercury in Seafood ”
- **EAM4.10** “High Performance Liquid Chromatography - Inductively Coupled Plasma - Mass Spectrometric Determination of Four Arsenic Species in Fruit Juice”
- **EAM4.11** “Arsenic Speciation in Rice and Rice Products Using High Performance Liquid Chromatography - Inductively Coupled Plasma-Mass Spectrometric Determination”

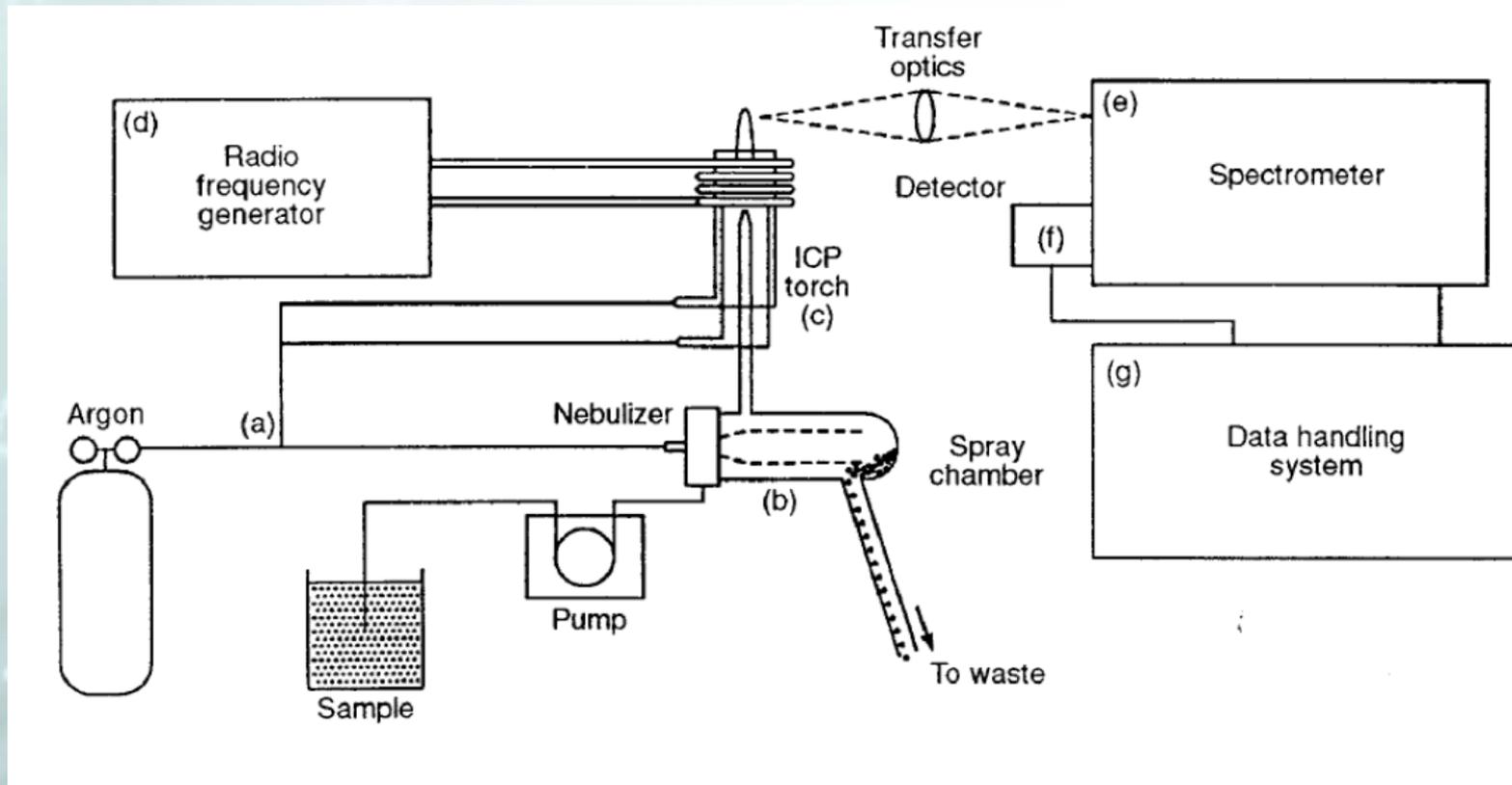


China

- **GB 5009.94-2012** “食品安全国家标准 植物性食品中稀土元素的测定”
- **GB 5009.11-2014** “食品安全国家标准 食品中总砷及无机砷的测定”



ICP-OES System Diagram



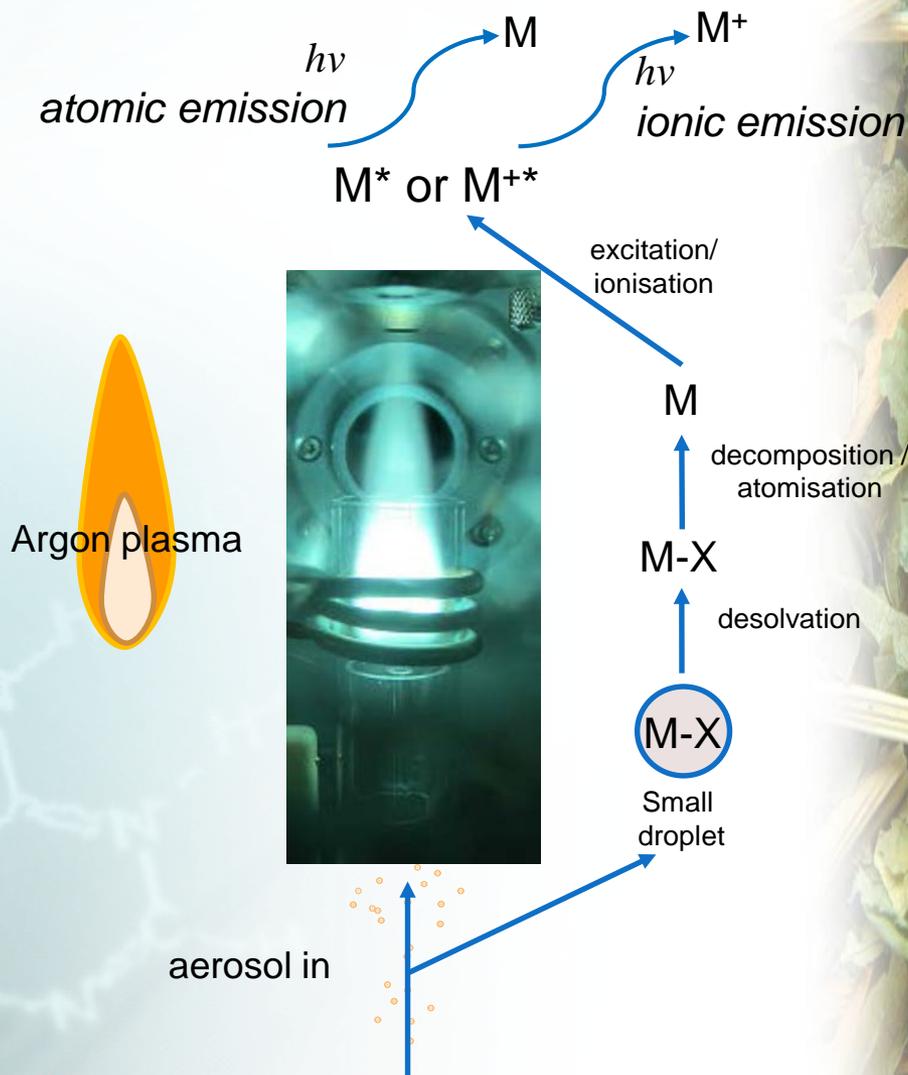
ICP-OES: Basic Principle

- Method basis: the measurement of emission of light by an optical spectroscopic technique.
- Liquid samples are nebulized to produce aerosol, which is transported to the plasma torch where excitation occurs.
- Characteristic emission spectra are produced by a radio-frequency inductively coupled plasma (ICP).
- The spectra are dispersed by a grating spectrometer and the intensities of the lines are monitored by a detector.
- The signals from the detector(s) are processed and controlled by a computer system.



ICP-OES Excitation

- Argon plasma excites the atoms / ions into higher energy levels (i.e., excited states) that subsequently emit light when they return to the ground state.
- The emitted light does not have a continuum therefore is expressed as spectral lines specific for giving element.



ICP-OES spectrometer

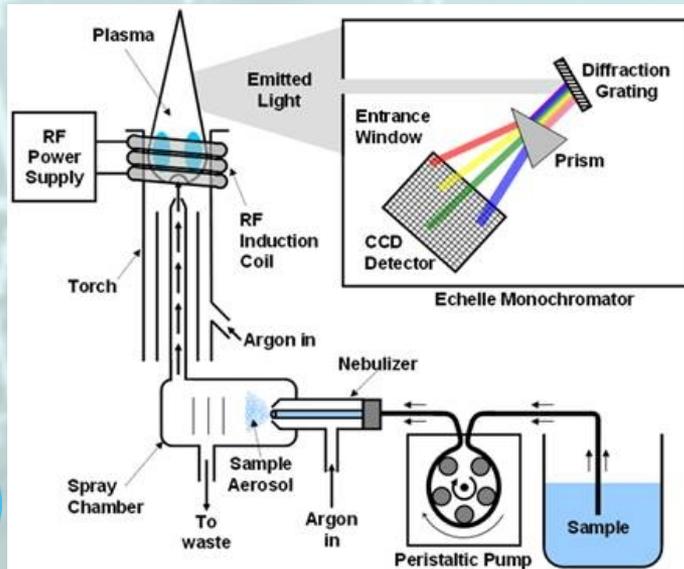
- Capability of determining multiple wavelengths for each element of interest with the required intensity
- Axial, radial, or dual view grating configurations
- 3-channel peristaltic pump, nebulizer, spray chamber, torch
- RF power supply, RF induction coil
- Detector (e.g. PMT, solid state CCD)
- Argon gas supply unit
- Auto-sampler



ICP-OES vs traditional AAS

➤ Advantages

- Multi-element determinations in a single run
- Dual viewing of the plasma design offers linear range from % to sub-ppm level
- Simultaneous background correction can be performed adjacent to the analytical wavelengths



- Solid state detectors (e.g. CCD) usually provide uniform resolution at all wavelengths
- Able to analyse routinely of 10% total dissolved solid (TDS)



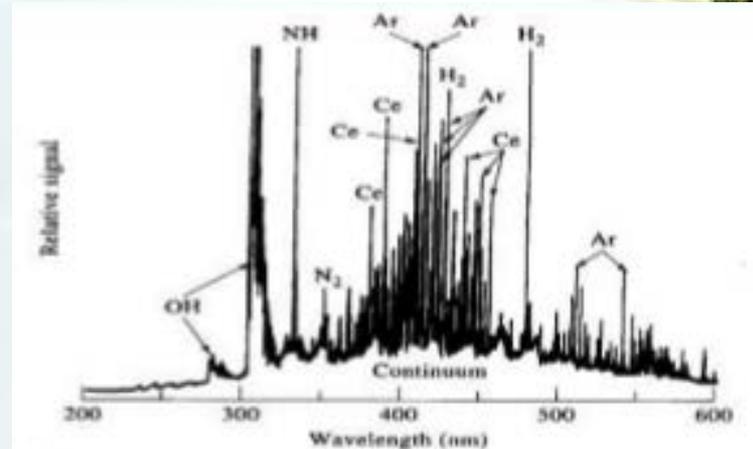
ICP-OES: Food Analysis

- Commonly used for the determination of % to ppm levels of Ca, Na, K, Mg, Fe, P, Zn... and ppm to sub-ppm of toxic elements Cd, Pb ...in food samples
- Digested sample solution or an appropriate dilution is presented to the ICP-OES instrument.
- The instrument is calibrated with acid(s) matched standard solutions at multi-levels
- Internal standard (e.g. indium, strontium, and yttrium) is commonly used for the correction of physical and chemical interferences.



ICP-OES ...wavelengths

- A number of recommended and alternative wavelengths can be used for the elements of interest and internal standards.
- **One** recommended and **at least one** (if available) alternative wavelength for each element
- All responses for both recommended and alternative wavelengths for each element are corrected using **only one internal standard line**.
- Common wavelength (nm): Ca: 317.933 (In: 303.936); Fe: 259.94 (Sr: 338.071); K: 766.491 (Sr: 460.733)...
- Emission lines of appropriate sensitivity, free of interferences or corrected for interferences in specific matrixes, are acceptable.



ICP-OES: Food Analysis...more (2)

- Power settings and nebulizer gas flow better be optimized: e.g. Mg280.271:Mg285.213 ratio (Mermet principle of robust plasma) demonstrates robust operating conditions in accordance with the ratio established by the instrument manufacturer (e.g. >10).
- At least three replicate readings of the same sample are performed, with relatively long integration times to minimize noise.
- Other calibration techniques: matrix-matched bracketing, standard addition



ICP-OES: Food Analysis...more (3)

- Certain food materials contain substantial concentrations of easily ionizable elements (EIEs) such as K, Na, and Ca.
- Presence of EIEs in relatively large quantities in plasma upsets equilibrium conditions
- Both suppressing and enhancing effects in complex ways may take place for analyte signals in ICP-OES
- Examples: measurement of sodium in high Ca milk product; calcium in soy sauce (high Na)...
- Ionization buffer (e.g. cesium) provides a significant source of electrons in the plasma. The presence ionization buffer of in all samples and standards can minimize the effects of varying concentrations of EIEs in the sample.



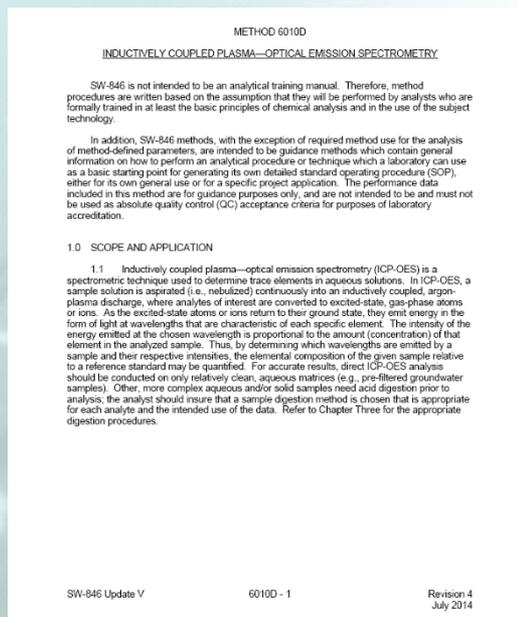
ICP-OES: Interferences

- Spectral interferences
 - Background emission – background correction
 - Molecular spectra overlap of the same target element and optical spectral-line overlaps between target elements - careful selection of alternative emission lines
- Physical interferences due to large difference in viscosity and surface tension between sample solutions and standard solutions – correction includes further sample dilution, using IS for calibration, standard addition
- Chemical interferences includes molecular compound formation, ionization effects and solute vaporization effects – normally not significant with ICP-OES, can minimize by changing the operation condition, matrix-matching and standard addition



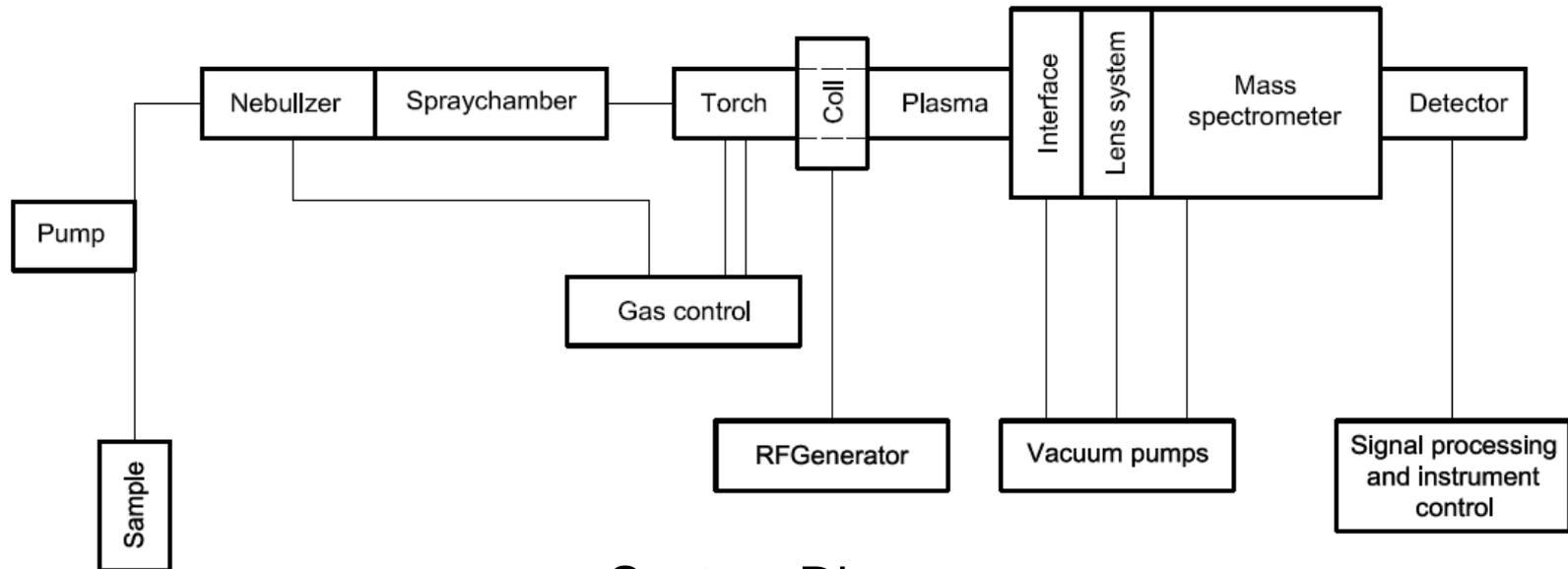
Other Useful References for ICP-OES

- OIML R116: 2006 “Inductively coupled plasma atomic emission spectrometers for the measurement of metal pollutants in water”, International Recommendation by the International Organization of Legal Metrology (OIML)
- USEPA SW-846 Method 6010D “Inductively Coupled Plasma – Optical Emission Spectrometry”, Revision 4, July 2014.



ICP-MS

- Analytical technique whereby molecules are atomized and ionized, providing elemental and isotopic information
- A very sensitive, element selective detection system, (for trace analysis; part per billion level, even part per trillion level)



System Diagram

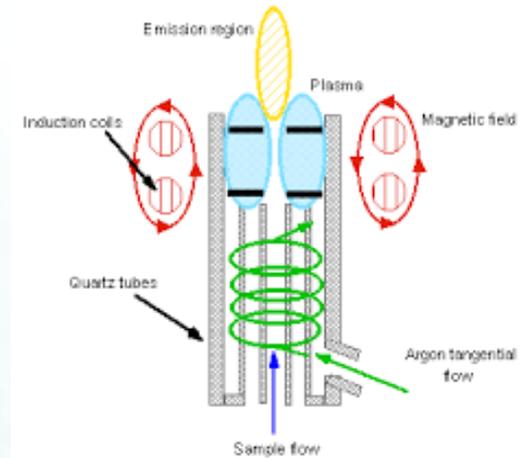
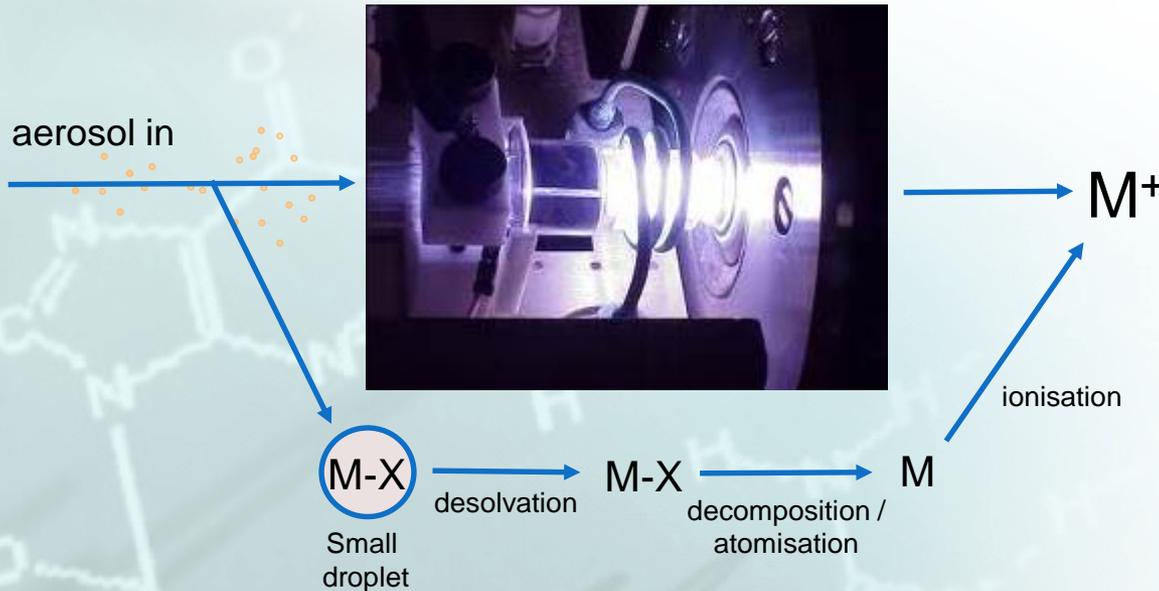
ICP-MS: Basic Principle

- Method basis: the measurement of ions (m/z ratio) generated from ICP using a mass spectrometer to separate and quantify those ions.
- Liquid samples are nebulized to produce aerosol, which is transported to the plasma torch where ionization occurs.
- The ions transfer from the plasma to the mass spectrometer via the interface (a sampling and a skimmer cone)
- The electronic lens system direct the ions travel from the interface to and focused on the entrance of the analyser.
- In the analyser (e.g. quadrupole), the ions are separated according to their mass-to-charge ratio, m/z .
- the ions exiting the analyser will hit the detector conversion dynode resulting in the release of electrons
- The signals from the detector(s) are processed and controlled by a computer system.



ICP-MS Analysis

Argon plasma

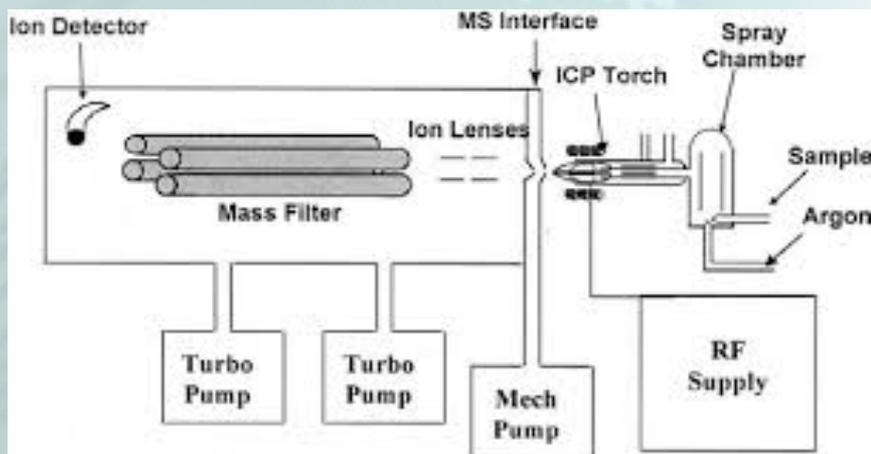


- Extremely **HOT** argon plasma generated at the end of a torch by means of electromagnetic interaction
- Desolvation of the aerosol; fragmentation of the entire molecules to atoms and ionization all happens in the gas phase, providing elemental and isotopic information



ICP-MS spectrometer

- 3-channel peristaltic pump, nebulizer, spray chamber, torch
- Torch, RF power supply, RF induction coil
- Interface, ion lens
- Mass analyzer, ion detector
- Argon gas supply unit
- Auto-sampler



ICP-MS: Food Analysis

- Commonly used for the determination of multi-elements (As, Cd, Hg, Pb...) from ppm to sub-ppb levels in food samples
- Digested sample solution or an appropriate dilution is presented to the ICP-MS instrument.
- The instrument is calibrated with acid(s) matched standard solutions at multi-levels
- Internal standard (e.g. Ge, In, Rh, Ho, Bi...) is commonly used for the correction of physical and chemical interferences.
- For serious matrix effects, apply matrix-matched calibration, or standard addition



ICP-MS: Choice of isotopes

- Measure analyte elements at two different isotopic masses (for elements with more than one isotope): one mass number for quantification and the other mass number for control purposes in case of doubt; e.g. $^{111,114}\text{Cd}$, $^{52,53}\text{Cr}$
- The control mass number is very often subject to more interferences than the quantitative mass number, care has to be taken when judging the results of this procedure.
- Advisable to measure the matrix elements with regards to possible spectral interferences, especially for samples of unknown or widely varying matrix composition.
- Due to variation in isotopic compositions, sum of 206 and 207 and 208 is used for the calculation of Pb



ICP-MS: Choice of internal standard

- Internal standard is used for reducing the matrix effect; assumption behind this approach is that the internal standard signal is influenced by the matrix in the same way as the analyte signal, resulting in a stable ratio
- The best correction is found when an internal standard with a similar 1st ionization potential is used.
- Internal standard should also have a mass similar to the analyte element to minimize the differences in mass dependent matrix effects and mass-dependent drift. Generally, an internal standard should be no more than 50 u removed from the analyte.
- For examples: ^{75}As (^{74}Ge), $^{111,114}\text{Cd}$ (^{103}Rh), $^{200,201,202}\text{Hg}$ (^{193}Ir), $^{206,207,208}\text{Pb}$ (^{209}Bi)



ICP-MS

➤ Advantages

- Can measure up to 70 elements simultaneously in a single sample analysis
- Superior detection capability (ppm to sub-ppb levels), excellent linear range
- Able to obtain isotopic information

<u>Element</u>	<u>Monitoring isotopes, m/z</u>	<u>Quantitation limit, mg/kg</u>
Arsenic	75	0.02
Cadmium	111, 114	0.01
Lead	206, 207, 208	0.005
Mercury	200, 201, 202	0.005



ICP-MS, Interferences (1)

- Isobaric molecular (polyatomic) and doubly charged ion interferences.
 - In many cases molecular ions contain argon (plasma gas) and/or oxygen originating from the water of the solution aspirated. Also chloride plays an important role with respect to the formation of interfering polyatomic ions, e.g. $^{40}\text{Ar}^{35}\text{Cl}^+$ (interference with $^{75}\text{As}^+$) and $^{44}\text{Ca}^{16}\text{O}^+$ (interference with $^{60}\text{Ni}^+$).
 - doubly charged ions, e.g. Ba^{2+} (interference with ^{65}Cu , ^{66}Zn , ^{67}Zn and ^{68}Zn)
 - Minimize the effects first by optimizing the instrumental plasma conditions and matrix components; can apply correction equations, or using the collision/reaction cell technology with a quadrupole instrument, ICP-triple quadrupole-MS, or the ICP-high resolution-MS

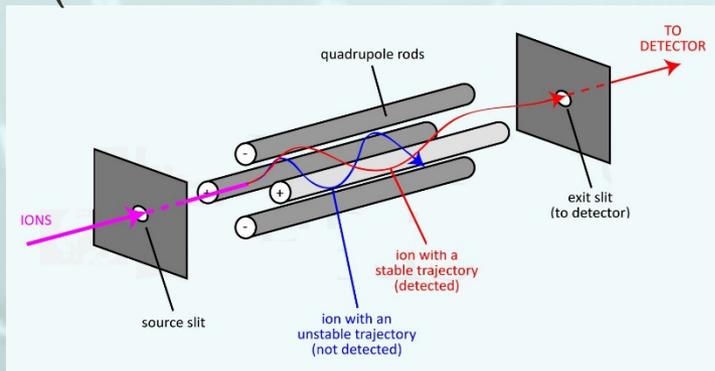


ICP-MS, different mass analyzer

Mass analyzers

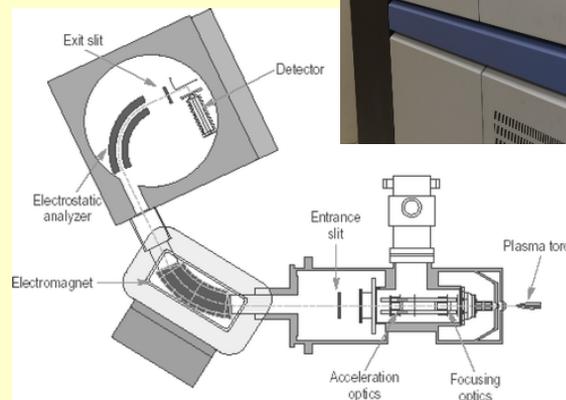
Ion beams resolved by the mass analyzer unit to provide elemental and isotope information

- Most common: Single-Quadrupole MS (w or w/o collision reaction cell)



schematic diagram for quadrupole MS

- Triple-Quadrupole MS



- High resolution model: Sector-Field Double Focusing HRMS

ICP-MS, Interferences (2)

➤ Isobaric elemental interferences

- an isotope of another element having the same mass as the analyte isotope, for example ^{58}Fe (interferant) and ^{58}Ni (analyte)
- Avoided by selection of an alternative analytical isotope, if unavoidable appropriate corrections must be made

➤ Physical Effects

- This can be due to transport process of sample to the plasma, sample conversion processes in the plasma and transmission of ions through the ICP-MS interface. The use of internal standard may compensate for physical interference effects.



ICPMS correction equations

- Based on the assumption that the number of the molecular (and doubly charged) ions formed is a fixed fraction of the number of parent ions present in the plasma, independent of variations of the sample composition and small variations of the plasma conditions.

e.g.

$$\text{Corrected } ^{75}\text{As signal} = (\text{m/z 75 signal}) - 3.13[(\text{m/z 77 signal}) - 0.87(\text{m/z 82 signal})]$$

$$\text{Corrected } ^{115}\text{In signal} = (\text{m/z 115 signal}) - 0.0149(\text{m/z 118 signal})$$

$$\text{Corrected } ^{58}\text{Ni signal} = (\text{m/z 58 signal}) - 0.04825(\text{m/z 54 signal})$$

- If collision/reaction cell technology is applied for the removal of molecular interference, correction equation does not apply!



ICP-MS for food analysis, more...

- For foodstuffs, the digested sample solution often contains variable amount of carbon and other dissolved solids (Na, Ca...)
- In the presence of carbon, large positive matrix effects (signal enhancement) can be observed for As and Se (carbon enhancing effect);
- A deposit, consisting of constituents from the measurement solutions, may formed around the orifices of the cones and influence the analysis (signal drift).
- May be minimized by further diluting the sample before analysis, add carbon source to both calibration standards and the measurement solutions, or using the method of standard addition.



Other Useful References

ICP-MS

- USEPA SW-846 Method 6020B “Inductively Coupled Plasma - Mass Spectrometry”, Revision 2, July 2014.
- ISO 17294-1:2004 “Water quality — Application of inductively coupled plasma mass spectrometry (ICP-MS) — Part 1: General guidelines”



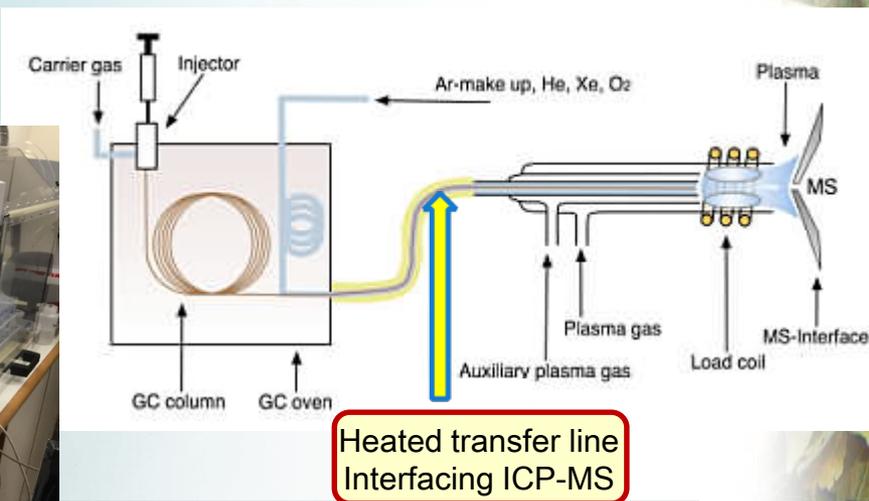
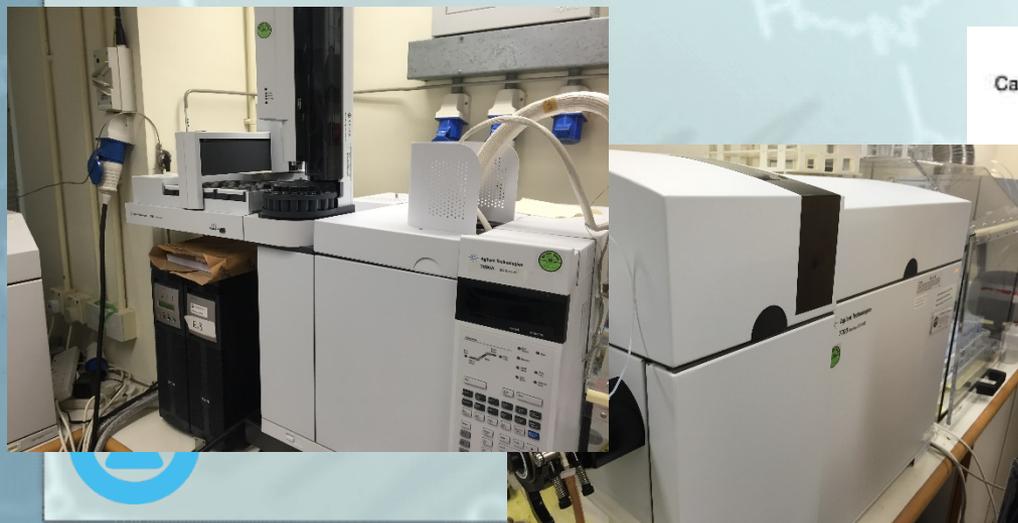
ICP techniques - Development

- Hyphenated technique for speciation analysis
 - GC-ICP-MS
 - organo-mercury, organotin
 - LC(IC)-ICP-MS
 - organo-mercury, organotin
 - Cr(VI)
 - Inorganic arsenic and organo-arsenic



Gas Chromatography (GC)—ICP-MS

- Very sensitive hyphenated system, but
- **applications limited solely** to organometallic compounds which are:
 - volatile and thermally stable in the native form (e.g. $(C_2H_5)_4Pb$), or
 - **convertible** to volatile and thermally stable form by **derivatisation** (e.g. hydride generation or alkylation), such as TBT & CH_3Hg^+
- Various sample introduction principles:
 - ✓ direct injection (split, splitless, on-column) of an organic solution
 - ✓ solid-phase microextraction (SPME),
 - ✓ headspace-injection or purge-and-trap using cryofocussing



Liquid Chromatography (LC)—ICP-MS

Liquid chromatography (LC): much versatile approach for elemental speciation with ICP-MS

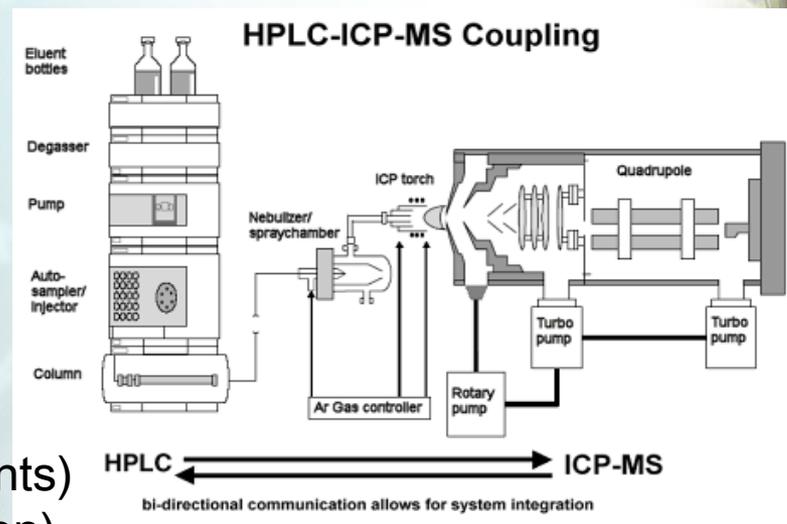
- LC coupling with ICP-MS is **straightforward**: direct connection of the column outlet to nebulizer of ICPMS is feasible
- both the stationary phase and the mobile phase may be altered to achieve the desired separation:

A variety of stationary phase:

- ✓ Reversed phase (RP)
- ✓ Ion pairing (IP)
- ✓ Ion exchange (IE)
- ✓ Size exclusion (SE)

Modification of mobile phase:

- Addition of additives (e.g. buffer, surfactants)
- Gradual change of polarity (gradient elution)



Thank you

