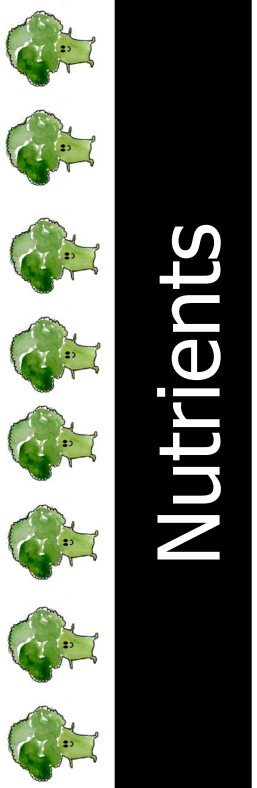
N-Protein and Nutrition

Chayabodee Sae-Jea
Product Specialist

The important of nutrition

- Nutrition is the nutrients in food and how they nourish the body
- **Nutrients** are component of food that are needed for the body to function



Macronutrients


Protein

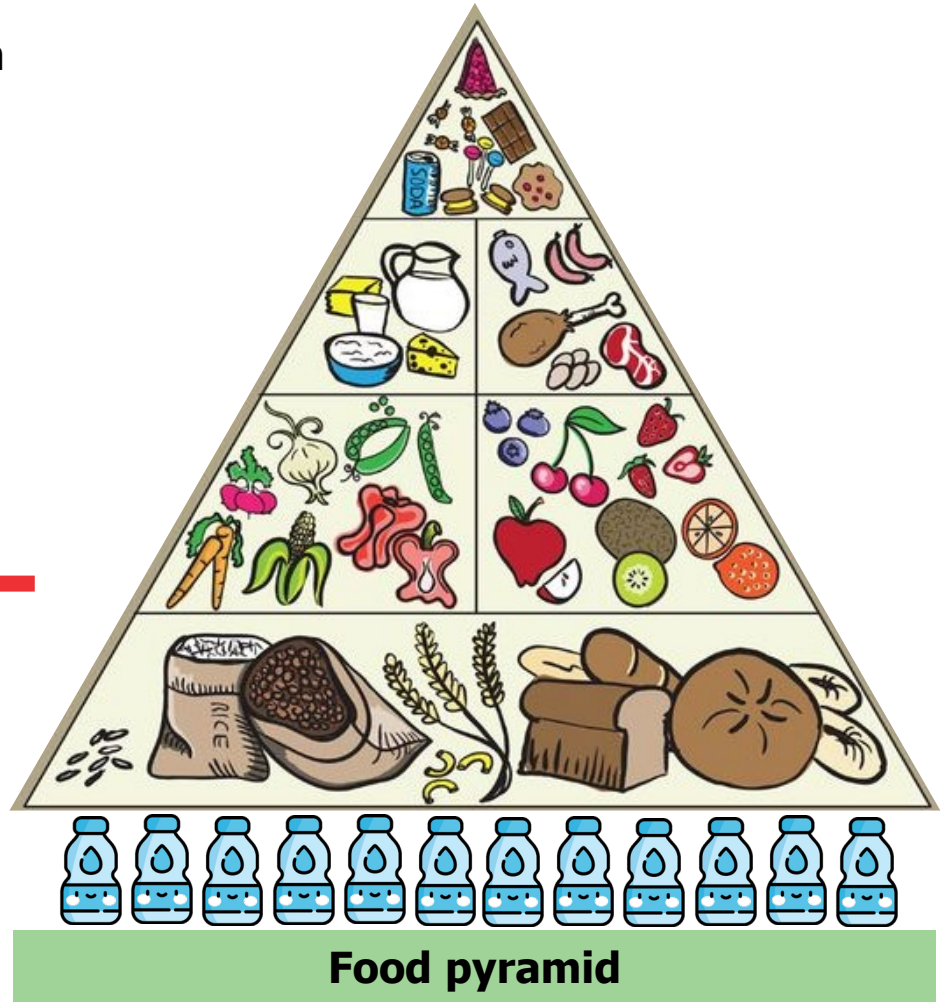

Lipid


Carbs

Micronutrients

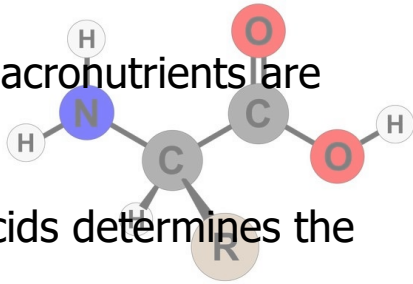

Vegetable


Fruit



Food pyramid

Protein



- Protein is one of three macronutrients, which are nutrients the body needs in larger amounts. The other macronutrients are fat and carbohydrates.
- Protein is made up of long chains of amino acids. There are 20 amino acids. The specific order of amino acids determines the structure and function of each protein.

Amino acid in body

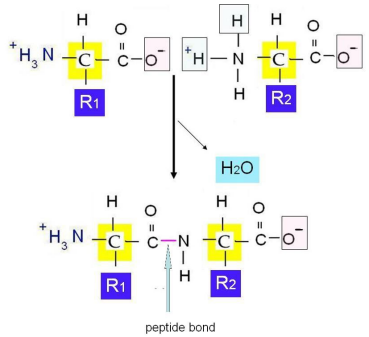
Non-essential amino acid	Essential amino acid
• Alanine	• Histidine
• Arginine	• Isoleucine
• Asparagine	• Leucine
• Aspartic acid	• Lysine
• Cysteine	• Methionine
• Glutamic acid	• Phenylalanine
• Glutamine	• Threonine
• Glycine	• Tryptophan
• Proline	• Valine
• Serine	
• Tyrosine	

- Protein Foods include all foods made from seafood; meat, poultry, and eggs; beans, peas, and lentils; and nuts, seeds, and soy products. Beans, peas, and lentils are also part of the vegetable Group

Protein Analysis (Crude Protein ; CP)

- Protein is organic substances that contain C, N, O, H

Foodstuff	Nitrogen conversion factor
Wheat meal	5.83
Flour	5.70
Pasta	5.70
Bran	6.31
Rice	5.95
Rye/Barley/Oats	5.83
Ground Nuts	5.46
Soybean/Seeds/Flour/Product	5.71
Milk	6.38
Cheese	6.38
Whey cheese	6.38
Other foodstuff not listed	6.25
Mixed protein source (foodstuff)	6.25

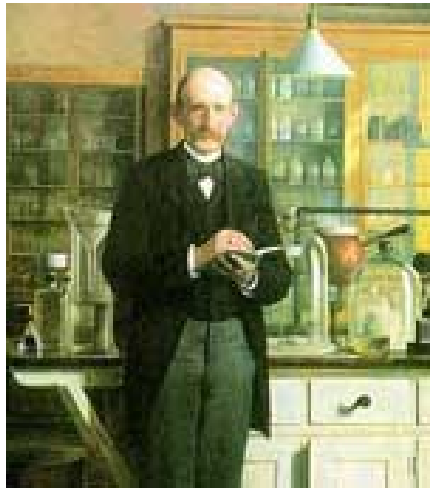


%Crude protein = %N x N conversion factor

Protein Method	Advantages	Disadvantages
Kjeldahl method	- Standard method globally - Easy to compare results with other laboratories	Does not measure true protein and overestimations of protein can result due to use of standard nitrogen correction factor 6.25
Dumas method	- Fast and does not use chemicals - Can measure several samples at a time	Costly to set up and is not very accurate
UV spectroscopy methods	Simple, does not require any assay agents	Highly error prone due to other compounds that absorb at the selected absorbance wavelength (280 nm)
Biuret methods	- Less protein–protein variation than the Coomassie dye-based assays - Compatible with most surfactants used for protein extraction	Incompatible with copper-reducing surfactants and reducing agents including DTT
Bradford Coomassie Blue assay method	- Fast, performed at room temperature, compatible with most solvents	High protein–protein variation; incompatible with detergents
Fluorescent dye methods	- Very sensitive and uses less protein	Requires a fluorescence detector

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7597951/

Kjeldahl method



Dane Johan

Principle : Kjeldahl method protein degradation which consists of amino acids (amino acids) that contain nitrogen as a component in amino group protein degradation nitrogen will be released and is converted to ammonia.

Step I : Digestion

$N(food) + H_2SO_4 \longrightarrow (NH_4)_2SO_4 + CO_2 + H_2O$

Step II : Distillation

$(NH_4)_2SO_4 + 2NaOH \longrightarrow 2NH_3 + 2H_2O + Na_2SO_4$

$NH_3 + H_3BO_3$ (boric acid) $\longrightarrow NH_4H_2BO_3$

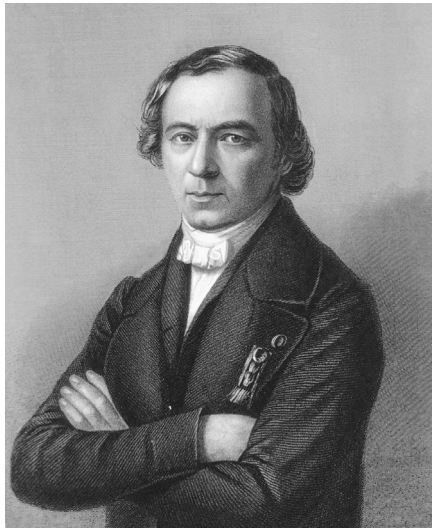
Step III : Titration

$2NH_4H_2BO_3 + H_2SO_4 \longrightarrow (NH_4)_2SO_4 + 2H_3BO_3$

Step IV : Calculation

$$\%\text{Total Nitrogen} = \frac{(A - B) \times C \times 0.014 \times 100}{D}$$

Dumas method



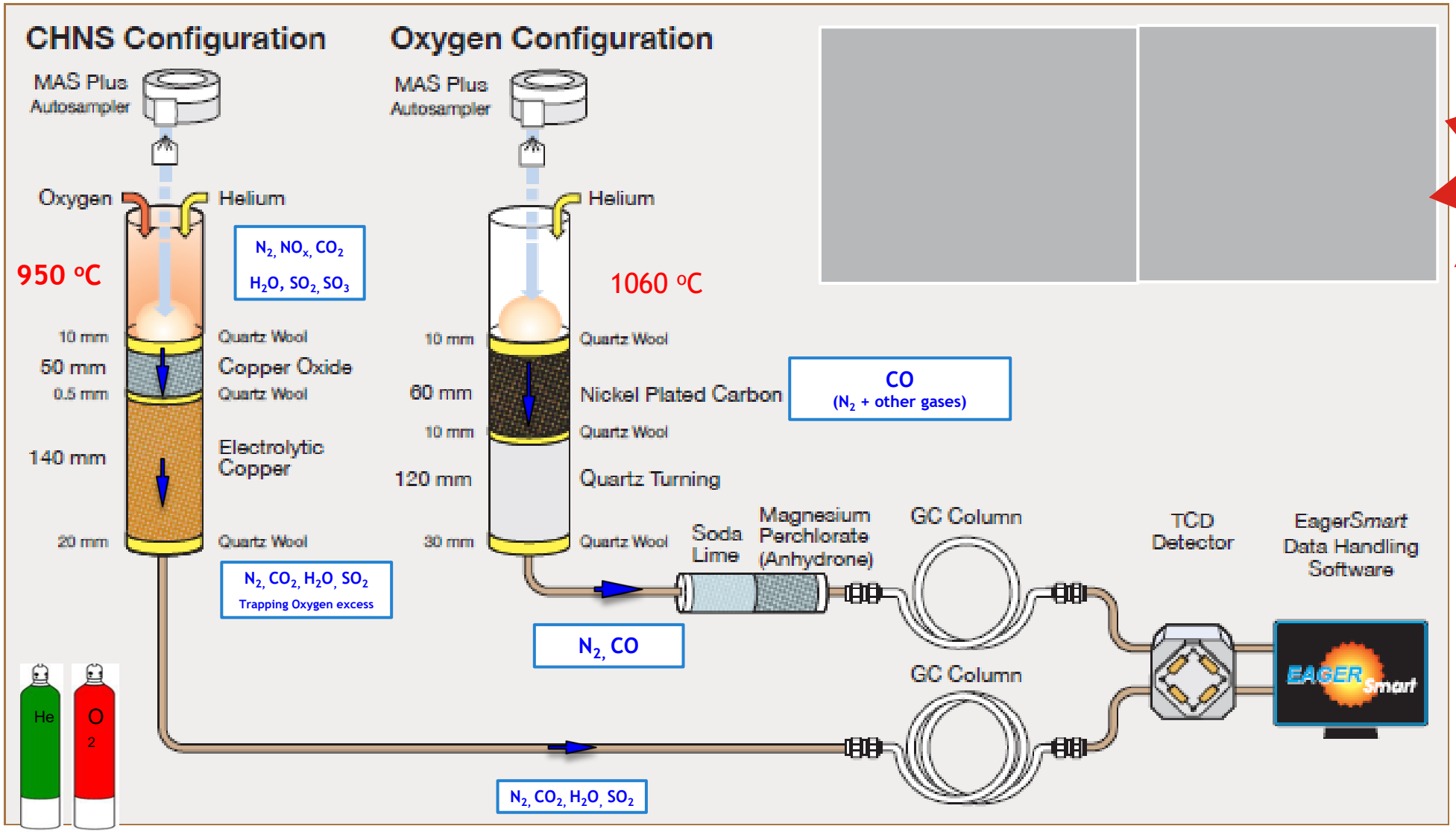
Jean Baptiste Andre Dumas

Principle

- The sample is weighed in an appropriate amount into a container such as a Tin capsule
- The sample is then burned in a combustion tube with a temperature of not less than 850 °C under a pure oxygen atmosphere.
- Carrier gas carries the entire substance into the Reduction tube
- Other compounds will be removed with adsorbents that have different specifications depending on the type of substance. The resulting water is removed by the sorbent.

Organic Elemental Analyzer

CHNS/O Analyzer principle



NEW

99.995% purity $O_2 + Sn \rightarrow SnO_2 + 1800\ ^oC$

N/Protein and S determination in soy and fish sauce

- The Total Nitrogen (TN) and Amino-type Nitrogen (AN) contents are generally used as the quality indices for soy sauce products.

- Sulfur is also an essential component of living matter. Sulfur deficiency has a negative influence in the quality of proteins as it is essential for the synthesis of amino acids such as cysteine, cystine, methionine and the synthesis of vitamins.

Sample name	Double Reactor System								Single Comb./Red. Reactor			
	Helium carrier gas				Argon carrier gas				Helium carrier gas			
	N%	RSD%	Prot.%	RSD%	N%	RSD%	Prot.%	RSD%	N%	RSD%	Prot.%	RSD%
Soy sauce 1	0.91 0.91 0.90 0.91 0.91	0.49	5.69 5.66 5.66 5.66 5.69	0.29	0.91 0.91 0.91 0.90 0.90	0.60	5.66 5.67 5.69 5.64 5.65	0.34	0.91 0.90 0.89 0.91 0.91	0.99	5.68 5.65 5.59 5.69 5.68	0.72
Soy sauce 2	0.48 0.48 0.47 0.48 0.47	1.15	2.99 2.99 2.93 3.03 2.92	1.55	0.46 0.47 0.46 0.46 0.46	0.97	2.89 2.92 2.85 2.90 2.91	0.93	0.47 0.48 0.46 0.47 0.47	1.50	2.96 2.98 2.90 2.94 2.95	1.01
Soy sauce 3	2.23 2.21 2.21 2.22 2.22	0.38	13.92 13.83 13.80 13.90 13.85	0.36	2.20 2.20 2.22 2.20 2.21	0.41	13.74 13.77 13.89 13.73 13.84	0.50	2.20 2.21 2.20 2.20 2.21	0.25	13.72 13.83 13.76 13.75 13.81	0.33
Fish sauce 1	1.40 1.40 1.39 1.42 1.40	0.73	8.74 8.74 8.69 8.86 8.74	0.72	1.38 1.38 1.38 1.38 1.41	0.81	8.63 8.60 8.62 8.59 8.78	0.90	1.140 1.40 1.38 1.39 1.40	0.64	8.77 8.77 8.61 8.70 8.77	0.81
Fish sauce 2	0.85 0.84 0.85 0.85 0.85	0.45	5.31 5.25 5.29 5.30 5.29	0.43	0.83 0.82 0.82 0.82 0.83	0.66	5.18 5.12 5.10 5.11 5.19	0.81	0.83 0.84 0.84 0.84 0.84	0.49	5.19 5.23 5.23 5.24 5.26	0.49

Authors
Dr. Liliana Krotz,
Dr. Francesco Leone,
and Dr. Guido Giazzi
Thermo Fisher Scientific,
Milan, Italy

Keywords
Combustion, Fish sauce, Food quality, Nitrogen, Protein, Soy sauce, Sulfur

Goal
Demonstrate the performance of the Thermo Scientific Flash*Smart* Elemental Analyzer for food quality and labeling purposes for Asian sauces.

Introduction
Asian sauces are used in food preparation to enhance flavor and are an important part of Asian cuisine. Soy sauce, a traditional fermented product, is widely used. Its main ingredients are salt and protein hydrolisates (amino acids and peptides), using steamed defatted soybean flakes and baked wheat grains as the main starting materials. Its quality varies greatly with the raw materials used and the method of manufacture.

The Total Nitrogen (TN) and Amino-type Nitrogen (AN) contents are generally used as the quality indices for soy sauce products. According to the national standard in Taiwan, the first-grade soy sauce products should contain more than 1.4 TN% and more than 0.56 AN%. In Japan, the Japanese Agricultural Standard (JAS) specifies three grades of soy sauce: special, upper and standard.

Other traditional sauces, such as fish sauce, are used as condiments and sometimes as substitutes for soy-bean ones.

ThermoFisher SCIENTIFIC

Benefit

Performance

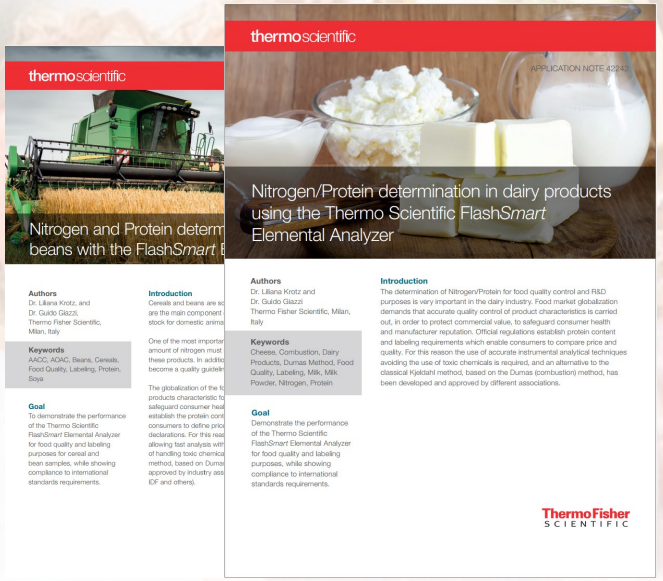
- All type of samples for all application fields
- No memory effects, no matrix (halogens, salts, organometallics etc.) problem
- From 1 to 5 elements: CHNS by combustion, oxygen by pyrolysis
- Total Organic Carbon (TOC) determination
- High sensitivity (few ppm to 100%)
- High accuracy and precision
- High reproducibility: EFCt (Electronic Flow Control), no often calibrations needed
- Low helium and oxygen consumption
- Compliance with most recognized Official Methods

Robustness

- Future proof, modular system
- GC separation column operating for years without the need for replacement: it is not a consumable
- Unattended operation: analyses can run 24/7
- Minimized downtime
- High lifetime of gases and consumables
- Reduced cost per analysis
- Greater productivity
- Long term investment for the lab, which guarantees reliability and accuracy of analysis

Simplicity

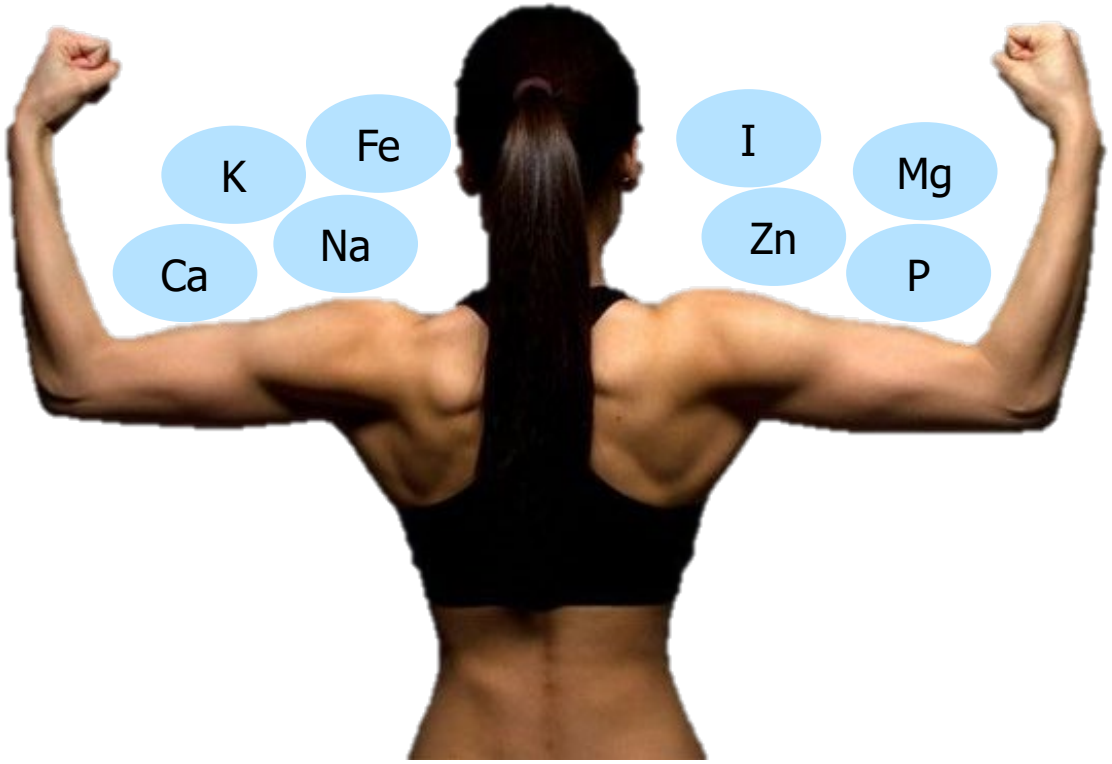
- Fully automated analytical workflows
- No sample digestion or toxic chemicals required
- No need for fume hood or anti-acid table
- Easy-to-use
- Easy maintenance
- GC separation method for real-time view of the analytical process and pathway: anytime, anywhere
- Streamline sample preparation without the need of dangerous chemicals as Kjeldahl method



Mineral your body needs

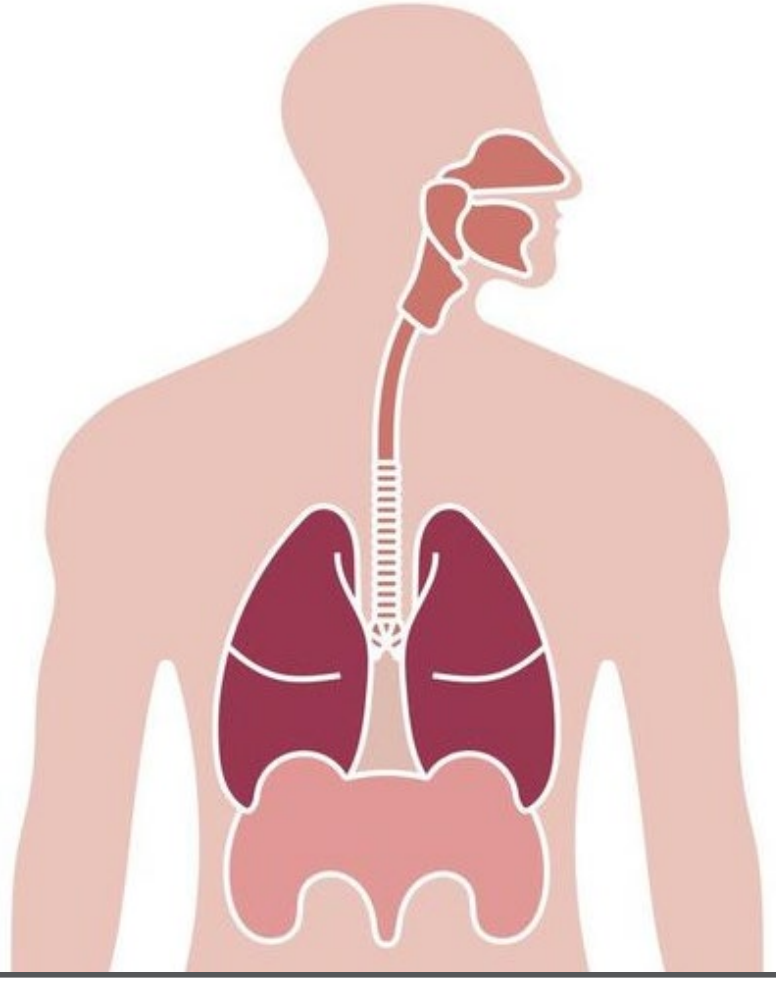
Food is important for life. To be healthy and active, we should certainly have enough food. But the foods we eat should also be safe and rich in all the nutrients our body needs.

- Minerals are important to overall health, and each serves a purpose within the body. Here is the low down on five important minerals and how to make them work for you.



Heavy metal poisoning

- Heavy metal poisoning (toxicity) is the result of exposure to heavy metals. Its bind to parts of your cells that prevent your organs from doing their job. Symptoms of heavy metal poisoning can be life threatening and they can cause irreversible damage



Sn

Zn

Cu

Hg

As

Cd

Pb

Where ?

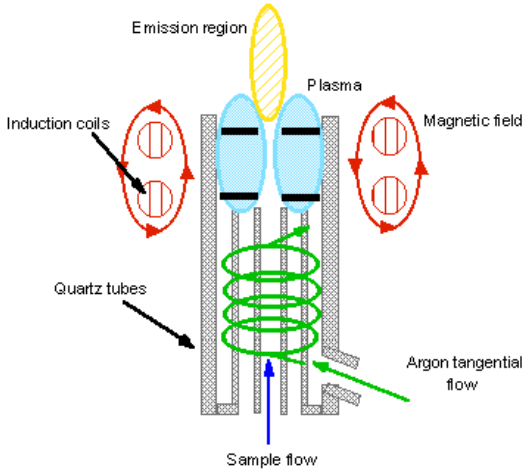
Industry

Raw material

Food packaging

How do I know?

Inductively Coupled Plasma – Optical Emission Spectrometry (ICP-OES)



1 H 1.008																	2 He 4.003
3 Li 6.941	4 Be 1.008											5 B 10.81	6 C 12.01	7 N 14.01	8 O 16.00	9 F 19.00	10 Ne 20.18
11 Na 22.99	12 Mg 24.31											13 Al 26.98	14 Si 28.09	15 P 30.97	16 S 32.07	17 Cl 35.45	18 Ar 39.95
19 K 39.10	20 Ca 40.08	21 Sc 44.96	22 Ti 47.88	23 V 50.94	24 Cr 52.00	25 Mn 54.94	26 Fe 55.85	27 Co 58.47	28 Ni 58.69	29 Cu 63.55	30 Zn 65.39	31 Ga 69.72	32 Ge 72.59	33 As 74.92	34 Se 78.96	35 Br 79.90	36 Kr 83.80
37 Rb 85.47	38 Sr 87.62	39 Y 88.91	40 Zr 91.22	41 Nb 92.91	42 Mo 95.94	43 Tc (98)	44 Ru 101.1	45 Rh 102.9	46 Pd 106.4	47 Ag 107.9	48 Cd 112.4	49 In 114.8	50 Sn 118.7	51 Sb 121.8	52 Te 127.6	53 I 126.9	54 Xe 131.3
55 Cs 132.9	56 Ba 137.3	57 La 138.9	72 Hf 178.5	73 Ta 180.9	74 W 183.9	75 Re 186.2	76 Os 190.2	77 Ir 190.2	78 Pt 195.1	79 Au 197.0	80 Hg 200.5	81 Tl 204.4	82 Pb 207.2	83 Bi 209.0	84 Po (210)	85 At (210)	86 Rn (222)
87 Fr (223)	88 Ra (226)	89 Ac (227)															
			58 Ce 140.1	59 Pr 140.9	60 Nd 144.2	61 Pm (147)	62 Sm 150.4	63 Eu 152.0	64 Gd 157.3	65 Tb 158.9	66 Dy 162.5	67 Ho 164.9	68 Er 167.3	69 Tm 168.9	70 Yb 173.0	71 Lu 175.0
			90 Th 232.0	91 Pa (231)	92 U (238)	93 Np (237)	94 Pu (242)	95 Am (243)	96 Cm (247)	97 Bk (247)	98 Cf (249)	99 Es (254)	100 Fm (253)	101 Md (256)	102 No (254)	103 Lr (257)	

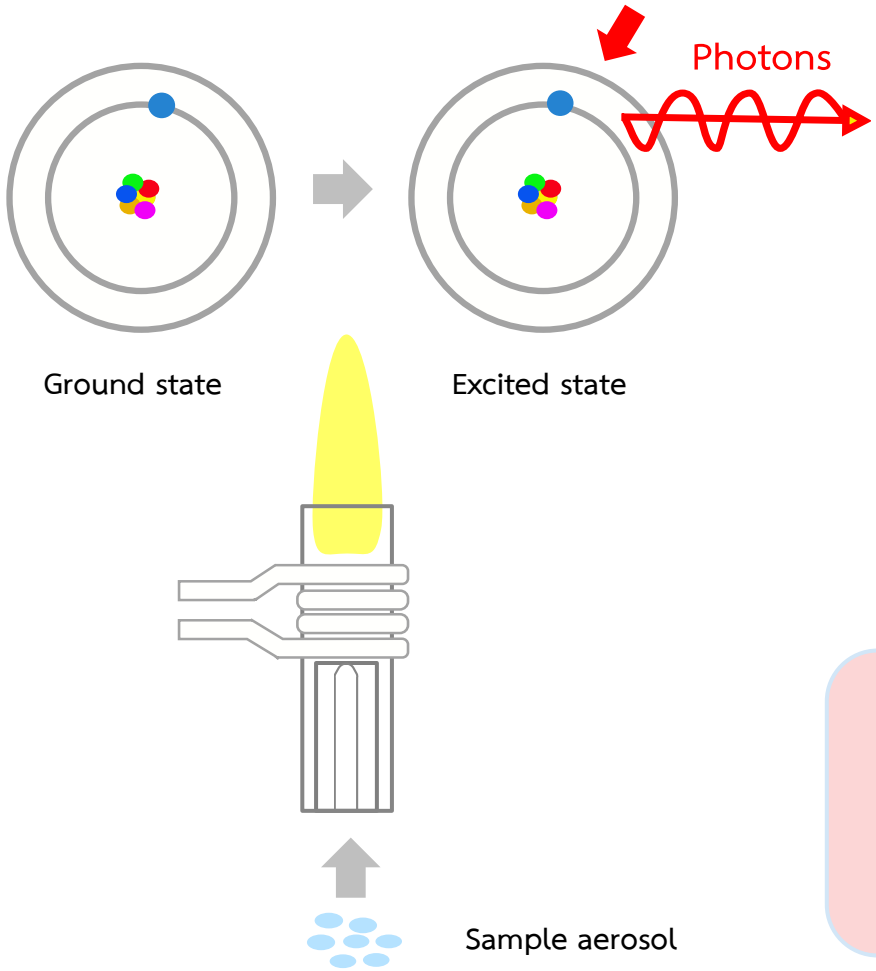
ICP/ICP-MS/AA

ICP-MS

ICP/ICP-MS

ICP

Inductively Coupled Plasma – Optical Emission Spectroscopy

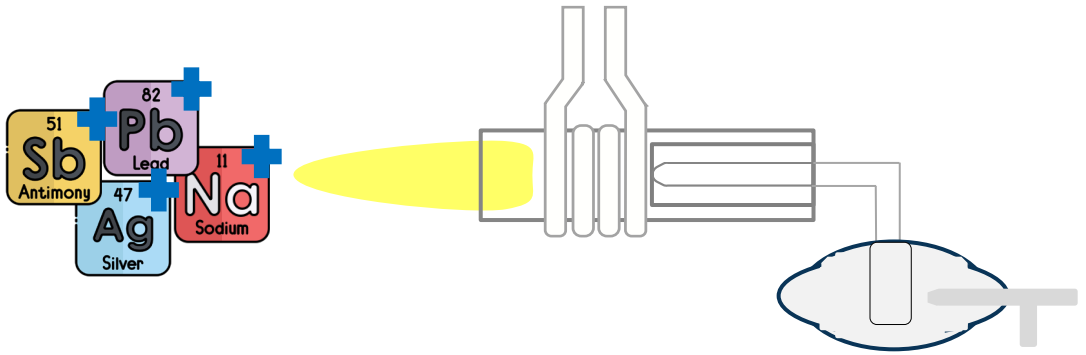


- Electrons in element are excited by plasma
- They jump to higher energy levels (excited state)
- the electrons fall back down (ground state or lower)
- Energy is emitted in the form of photons
- A detector measures the intensity of the emitted light, and calculates the concentration.

The ultraviolet (UV) / visible region (160 - 800 nm) of the electromagnetic spectrum is the region most commonly used for analytical atomic spectrometry.

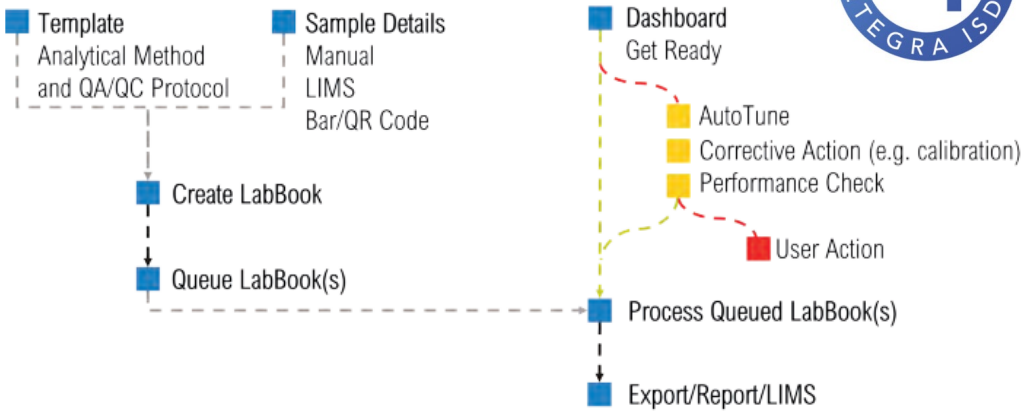
Inductively Coupled Plasma – Mass Spectrometry

- Electrons in the element are ionized by plasma and left out from the atom.
- Generated ions are extracted by the plasma interface into the mass spectrometer.
- The ICP-MS system will screen out the interference to improve sensitivity.
- Ion after passing the interference removal will enter the selective mass analyzer (Quadrupole).
- The screened analyte will count the signal by a Secondary Electron Multiplier (SEM) detector.



ICP-OES and ICPMS capabilities

- ✓ Multi-elements analysis: Impurities, Trace elements, Essential elements, Nutritional labeling
- ✓ Screening for unknown
- ✓ QA/QC
- ✓ Low detection limits
- ✓ Measurement at concentrations from parts per trillion to percent (sub ppb to %)
- ✓ High sample throughput
- ✓ Fast warm-up and standby mode
- ✓ Suitable for all sample types
- ✓ Short-time analysis
- ✓ Good matrix tolerance



THANK YOU