



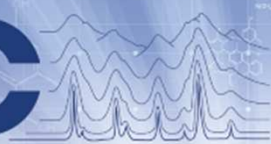
***DETERMINATION OF Pb, Cd,
Ni, Cr, Hg and As AT TRACE
LEVELS IN GEOLOGICAL
MATERIALS USED IN THE
FOOD INDUSTRY***

María Fernanda Gazulla



Invited speaker.
Denver X-Ray
Conference

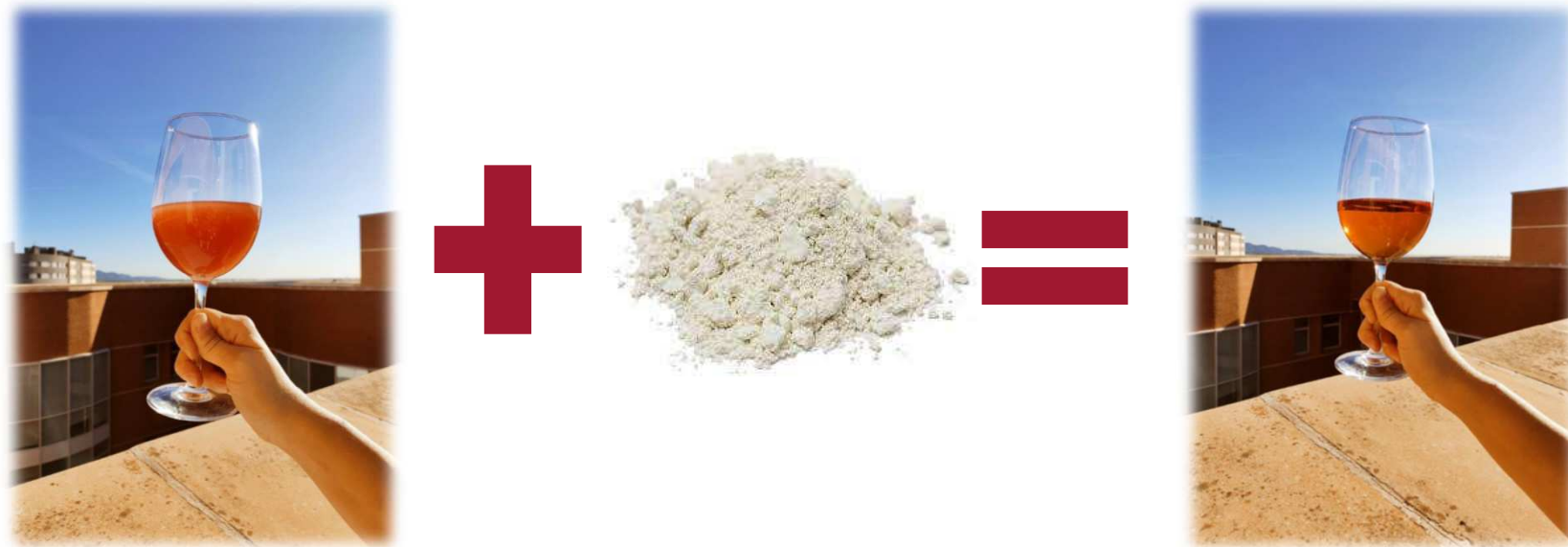
DXC



2 - 6, August, 2021

1. INTRODUCTION

- Certain geological materials are used in the food industry to enhance or modify the food properties as additives or processing aids.



1. INTRODUCTION

For instance, bentonites are used to clarify wines and vegetable oils

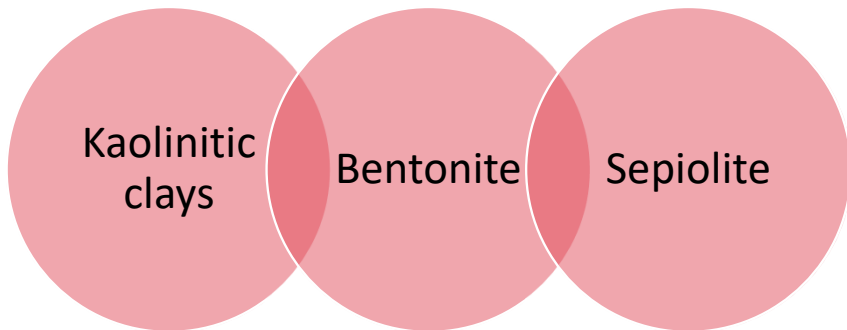
[1] N. Worasith, B.A. Goodman, N. Jeyashoke, P. Thiravetyan, *J. Am. Oil Chem. Soc.* **2011**; *88*, 2005.

[2] L.F. Londoño-Franco, P.T. Londoño-Muñoz, F.G. Muñoz-García, *Biotecnol. Sect. Agropecu. Agroind.* **2016**; *14(2)*, 145.

[3] Jaeckels, N.; Tenzer, S.; Meier, M.; Will, F.; Dietrich, H.; Decker, H.; Fronk, P.; *LWT – Food Science and Technology*, **2017**, *75*, 335

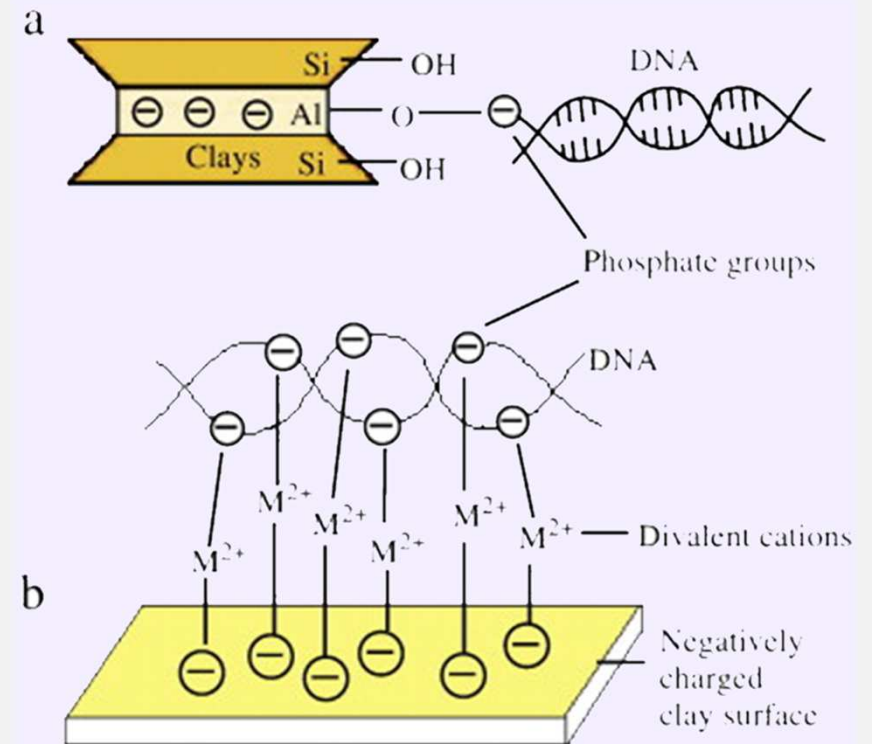


1.1 PROCESSING AIDS



The high specific area and the negative surface electrical charge allow these materials to adsorb positively charged compounds like proteins [1, 2]

Mecanism of absorbption

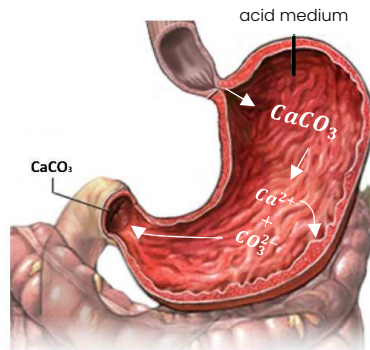
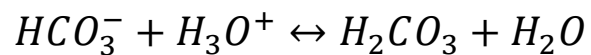
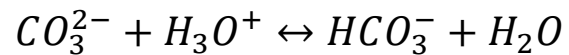
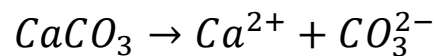


[4] S. Servagent-Noinville, M. Revault, H. Quiquampoix, M. H. Baron, *J. Colloid Interface Sci.* (2000), 221, 273
[5] W. A. Yu, N. Li, D. S. Tong, C. H. Zhou, C. X. Lin, C. Y. Xu, *Appl. Clay Sci.* (2013), 80-81, 443

1.2 ADDITIVES

• Calcium carbonate (CaCO_3)

- Used in bakery, calcium-rich drinks, breakfast cereals or canned fruit.
- Its behavior in the stomach improves the digestibility of some food.



• Potassium nitrate (KNO_3)

- Used to prevent bacteria and fungus in the meat industry, cheese, and some pizzas



Maximum permitted levels for some heavy metals present in food additives and processing aids.



Law Regulation	Additive	As (mg kg ⁻¹)	Cd (mg kg ⁻¹)	Cr (mg kg ⁻¹)	Hg (mg kg ⁻¹)	Ni (mg kg ⁻¹)	Pb (mg kg ⁻¹)
Regulation (EU) no. 231/2012	CaCO ₃	3	1	5	-	5	3
	KNO ₃	3	-	5	1	5	2
	Bentonite	-	-	-	-	-	-
Directive 2008/84/CE	CaCO ₃	3	-	-	1	-	5
	KNO ₃	3	-	-	1	-	5
	Bentonite	2	-	-	-	-	20
FAO and WHO	CaCO ₃	3	-	-	-	-	3
	KNO ₃	-	-	-	-	-	2
	General limits	Indicated by the manufacturer	1	-	1	-	2 (1 for high consumption)

- FAO and WHO Explanatory note evidences the need to develop a method that avoids the dry-ashing procedure, due to the potential loss of metals and arsenic with high temperatures.

[6] Commission Regulation (EU) No 231/2012 of 9 March 2012 laying down specifications for food additives listed in Annexes II and III to Regulation (EC) No 1333/2008 of the European Parliament and of the Council Text with EEA relevance

[7] Joint FAO/WHO expert committee on food additives (JECFA), *Limit test for heavy metals in food additive specifications. Explanatory note*, FAO Joint Secretariat, 2002

State of art

ICP-MS



Advantages:

- Extremely sensitive

Disadvantages:

- Time consuming
- Requires highly-specialized technicians
- Requires a special sample preparation

HG-AAS



Advantages:

- Recommended for As and Hg determination

Disadvantages:

- Time consuming
- Requires highly-specialized technicians
- Requires a special sample

ICP-OES



Advantages:

- Low quantification limits for all the analytes

Disadvantages:

- Requires a special sample preparation

WD-XRF

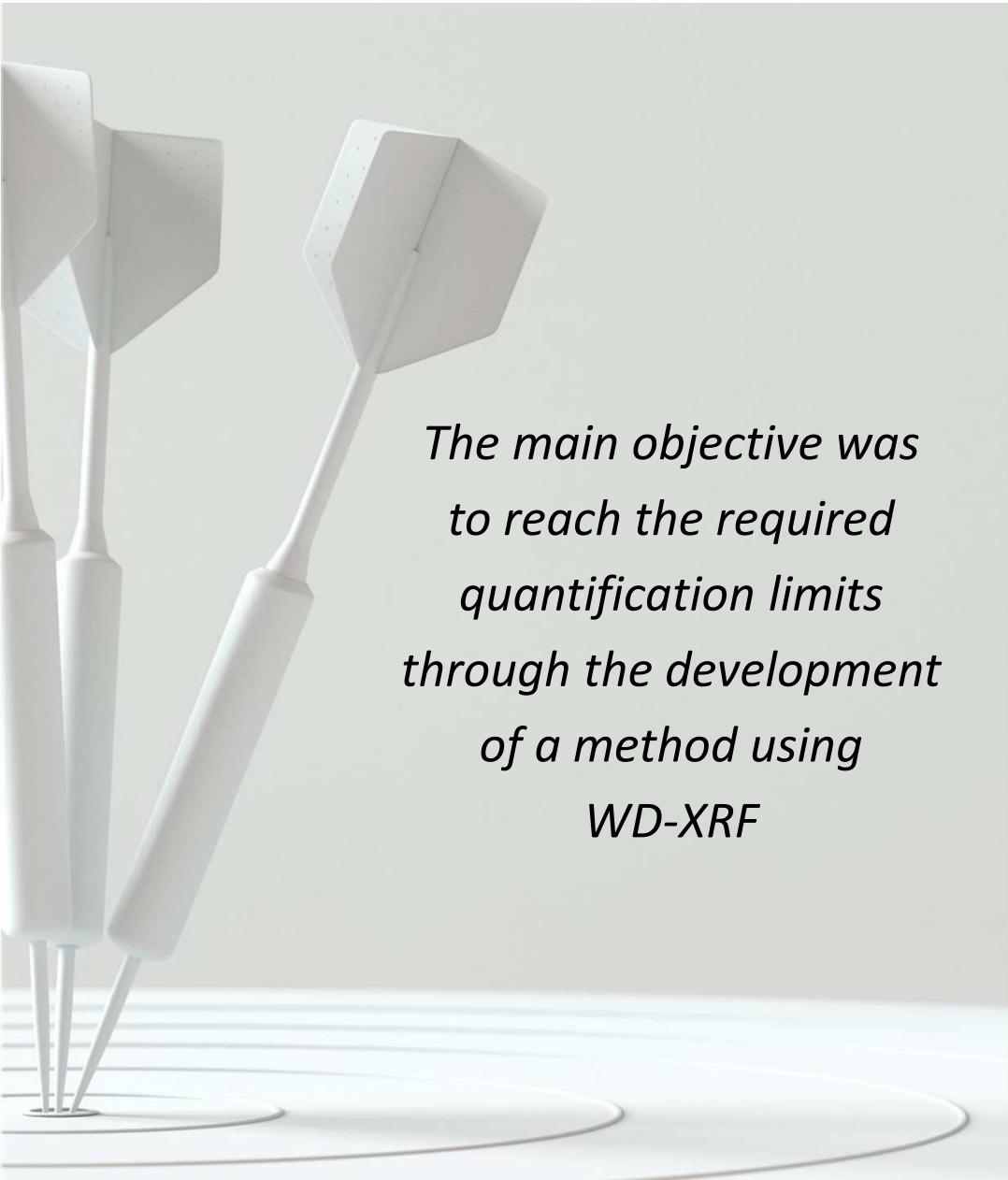


Advantages:

- Short time of analysis
- Easy to operate

Disadvantages:

- QL sometimes are higher than the required



*The main objective was
to reach the required
quantification limits
through the development
of a method using
WD-XRF*

2. OBJECTIVES



To reach the quantification limits required



To decrease the time of analysis

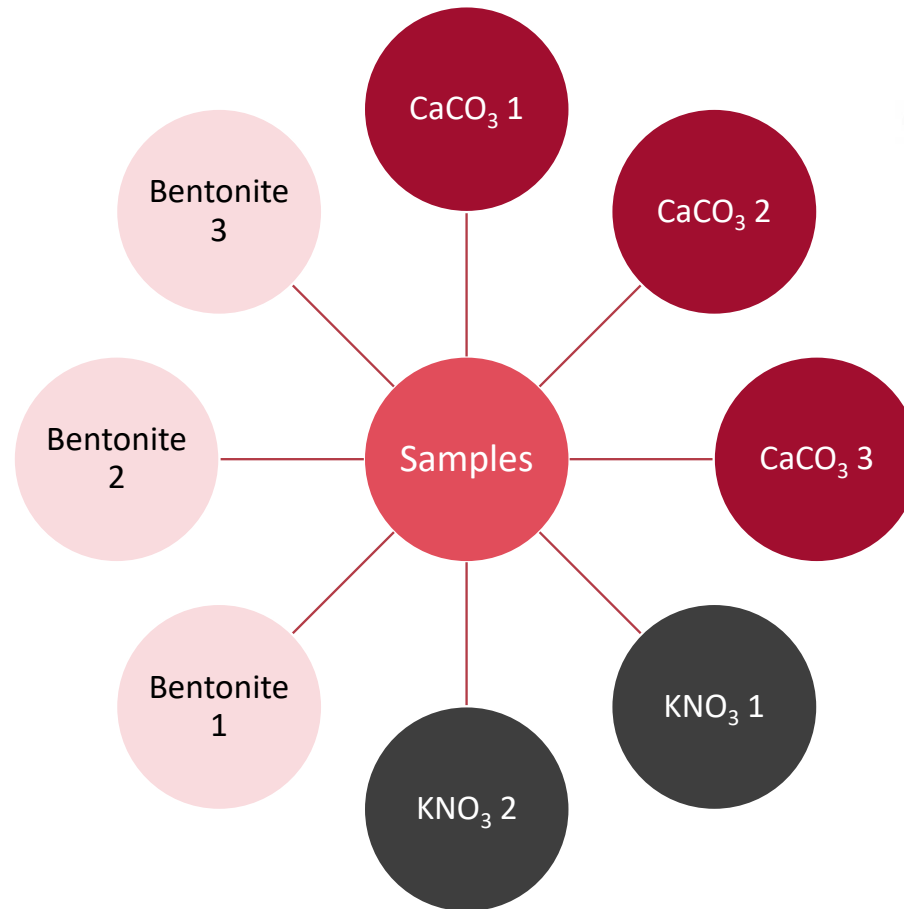


To develop an environmentally friendly control method

3. EXPERIMENTAL PART

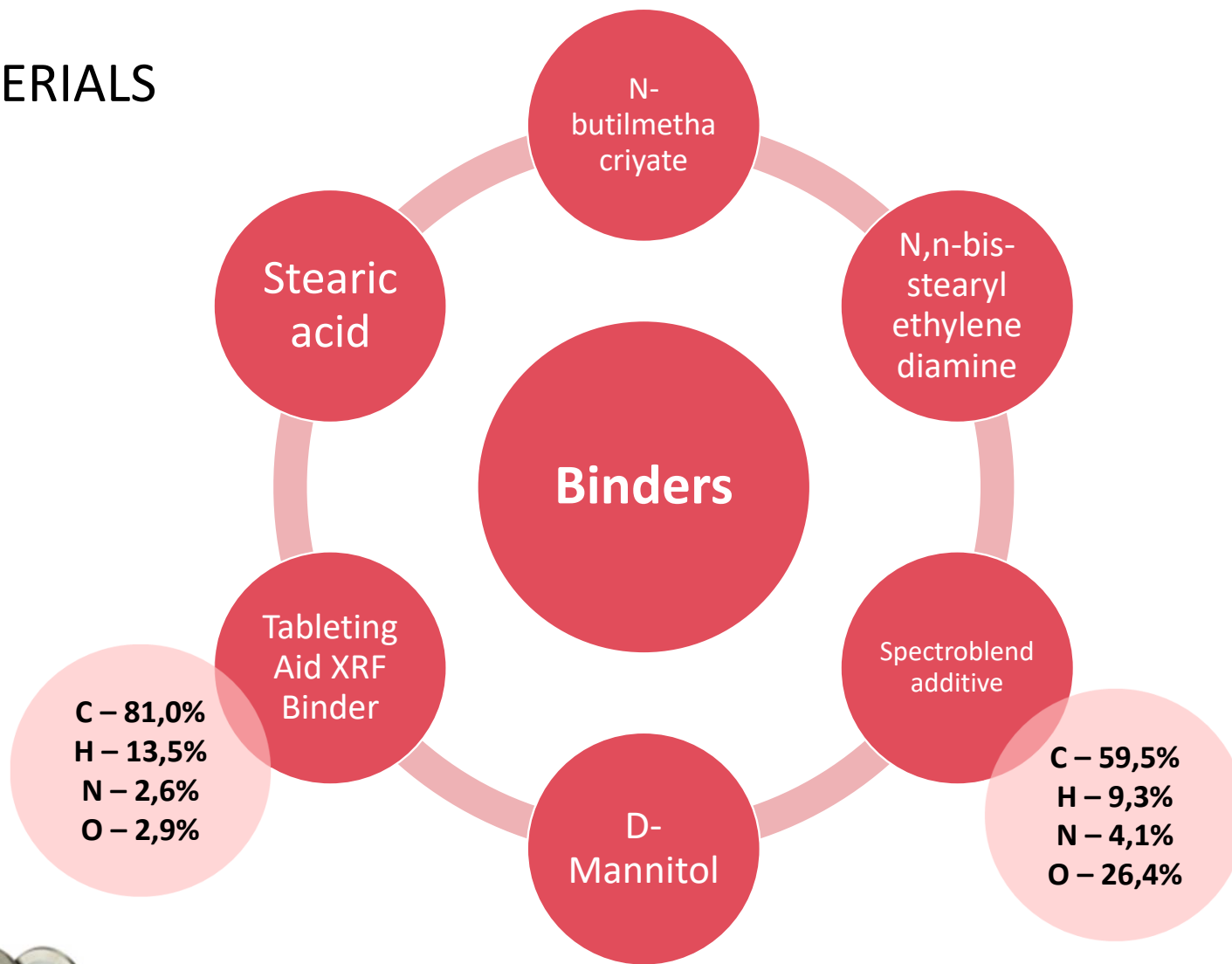
3.1. MATERIALS

- Samples



3.1. MATERIALS

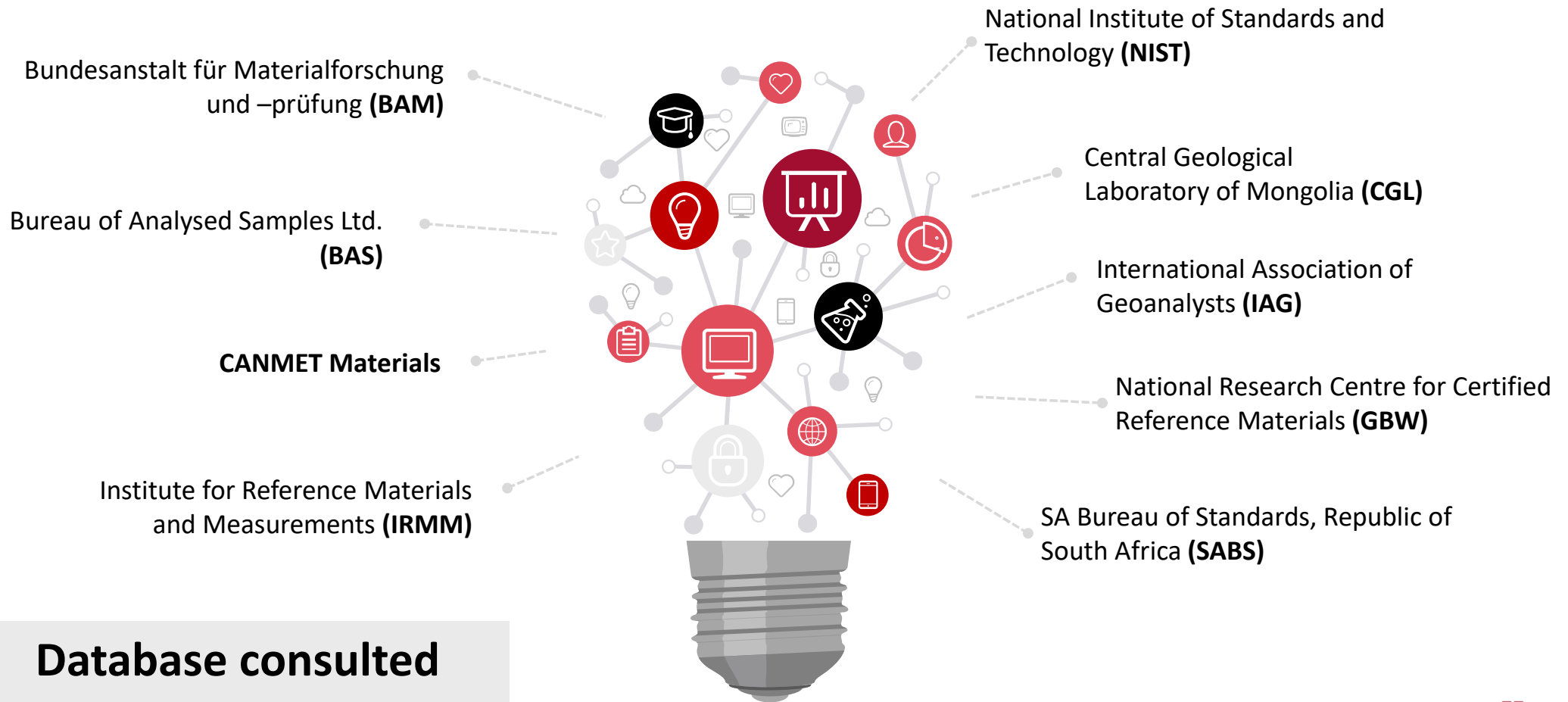
- Binders



[8] Gazulla, M.F.; Rodrigo, M.; Ventura, M.J.; Orduña, M.; Andreu, C.; *X-Ray Spectrometry*, (2021), 50, 197-209

3.1 MATERIALS

- Certified Reference Materials



3.1 MATERIALS

- Certified Reference Materials

Certified Reference Materials		Pb (mg·Kg ⁻¹)	Ni (mg·Kg ⁻¹)	Cr (mg·Kg ⁻¹)	Cd (mg·Kg ⁻¹)	Hg (mg·Kg ⁻¹)	As (mg·Kg ⁻¹)
Interlaboratory Test for the Analysis of geological samples (GeoPT) organised by IAG (International Association of Geoanalysts) (United Kingdom)	GeoPT-11 (OU-5 Leaton Dolerite)	4.6±0.3	15.00±1.22	38.40±2.32	0.20±0.04	-	2.45±0.16
	GeoPT-13 (UoK LOESS)	11.34±0.22	42.71±0.82	105.7±5.4	-	-	6.7±0.3
	GeoPT-40A (Calcareous organic-rich shale, ShTX-1)	6.05±0.58	74.92±2.56	29.65±0.26	2.02±0.12	-	15.05±0.84
Central Geological Institute of Mongolia (CGL) (Mongolia)	Mercury Soil-2 (MS-2)	-	-	-	-	1.52±0.08	-
	Mercury Soil-3	-	-	-	-	2.75±0.19	-
National Research Centre for Certified Reference Materials GBW (China)	GBW 07103 GSR-1	31.0	2.3	3.6	0.029	0.0041	2.1 ± 0,4
	GBW 07401 Soil	98±6	20.4±1.8	62±4	4.3±0.4	0.032±0.004	34 ± 4
	GBW 07402 Soil	20±3	19.4±1.3	47±4	0.071±0.014	0.0015±0.003	13.7±1.2
	GBW 07403 Soil	26±3	12±2	32±4	0.060±0.009	0.060±0.004	4.4±0.6



3.2 INSTRUMENTATION

PROCEDURE 1

Hydraulic press
+
WD-XRF

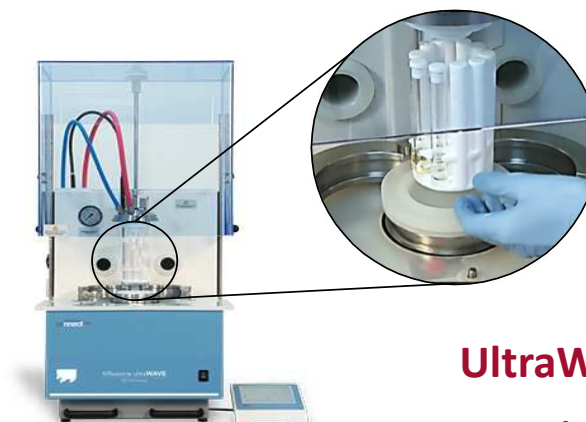


PROCEDURE 2

HG-AAS

or

ICP-OES



UltraWAVE

+



3.3 DEVELOPMENT OF THE METHOD

WD-XRF

Studied variables

Optimization of the sample preparation



- Type of binder
- Sample/binder rate
- Mixing process
- Pressure
- Size (27 mm ϕ , 37 mm ϕ)

Optimization of the measurement conditions



- Tube power
- Crystal
- Analyte line
- Detector
- Measurement time

3.3 DEVELOPMENT OF THE METHOD

ICP-OES

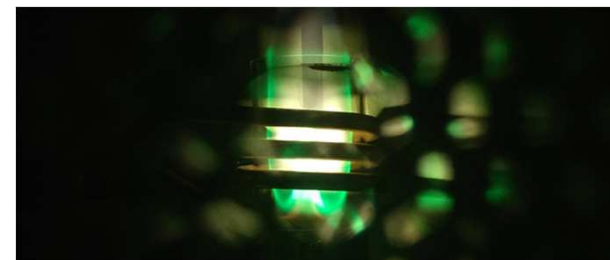
Studied variables

Optimization of the sample preparation



- Sample weight
- Type of acid
- Acid volume ratio
- Maximum temperature
- Digestion time

Optimization of the measurement conditions



- Plasma power
- Plasma flow
- Nebulizer flow
- Peristaltic pump speed

4. RESULTS

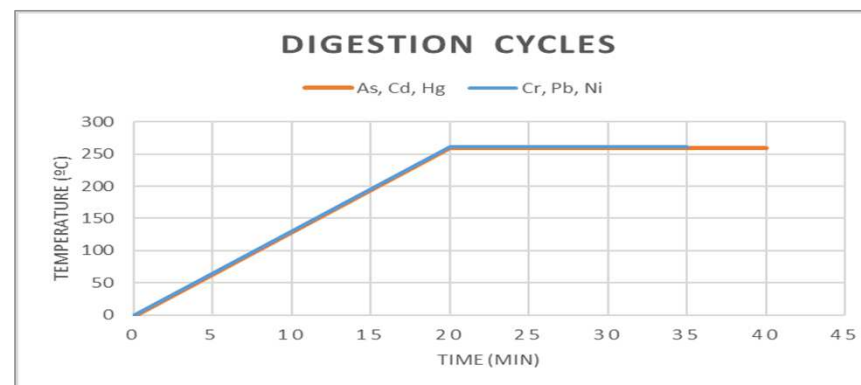
4.1 OPTIMISATION OF SAMPLE PREPARATION

Optimized conditions for pellets (WD-XRF)

Material	Binder	Ratio sample: binder (g)	Mixing			Total sample preparation time (min)
			Device	Time (min)	Speed	
Potassium nitrate	D-Mannitol	12:3	WC ring mill	1	II	20
Bentonite	D-Mannitol	12:1.5	WC ring mill	1	II	20
Calcium carbonate	D-Mannitol	12:2	WC ring mill	1	II	20

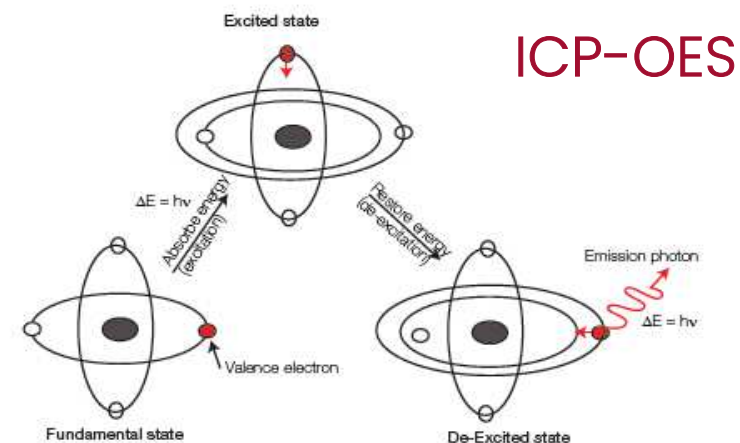
Optimized conditions for digestion (ICP-OES)

Elements	Sample weight (g)	Acid mixture	Final weight (g)
As, Cd and Hg	1,0	12 ml of inverse aqua regia	40
Pb, Cr and Ni	0,2	2 ml HNO ₃ + 6 ml HCl + 1 ml HF	30

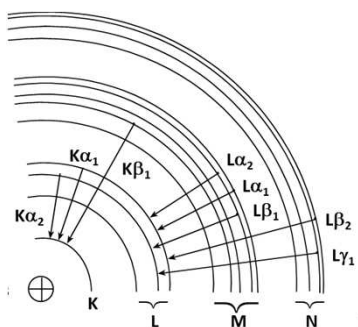


4.2 OPTIMISATION OF MEASUREMENT CONDITIONS

	As, Cd	Hg	Cr, Ni, Pb
Plasma Power (kW)	1,4	1,2	1,5
Plasma Flow (L/min)	13,5	15,0	12,0
Nebulizer Gas Flow (L/min)	0,70	0,85	0,80
Peristaltic Pump Rate (rpm)	12	15	10



WD-XRF



Electron shells

Element	Analyte line	Measurement angle 2θ (°)	Detector	Crystal	Voltage (kV)	Intensity (mA)	Time (s)
Pb	L_{β_1}	40.3754	Scintillation	LiF220	60	60	100
Ni	K_{α}	71.2382	Scintillation	LiF220	60	60	100
Cr	K_{α}	107.1520	Flow	LiF220	50	72	100
Cd	K_{α}	21.6540	Scintillation	LiF220	60	60	100
Hg	L_{α}	51.6746	Scintillation	LiF220	60	60	150
As	K_{β}	43.5876	Scintillation	LiF220	60	60	100

4.3 VALIDATION

Detection limit (DL)	Quantification limit (QL)
$L_D = 3,29s$	$L_Q = 10s$
$s = \text{standard deviation}$	



Uncertainty (U)	Method uncertainty (u_{method})
$U = k \cdot u_{method}$	$u_{method} = u_{V_R}^2 + u_{V_L}^2 + u_R^2$
$k = 2$	u_{V_R} = uncertainty of the certified value u_{V_L} = uncertainty of the measurement of the CRM u_R = uncertainty of the measurement of the sample



4.3 VALIDATION

- To compare the results obtained either with the certified value of the CRM or with values obtained by an independent technique, the difference between both (Δ_m) with the related uncertainty (U_{Δ_m}) were compared

$$\Delta_m = |c_m - c_{cert}|$$

$$u_{\Delta_m} = \sqrt{u_m^2 + u_{cert}^2}$$

$$U_{\Delta_m} = 2u_{\Delta_m}$$

Δ_m = absolute value of the difference between the measured and the known value

c_m = measured value by WD-XRF or ICP-OES

c_{cert} = certified value or value measured by an independent technique

u_{Δ_m} = combined uncertainty of the measured value and of the certified/measured by other technique value

u_m = uncertainty of the measured value by WD-XRF or ICP-OES

u_{cert} = uncertainty of the certified value or value measured by an independent technique

Goodness of the method \longrightarrow $\Delta_m \leq U_{\Delta_m}$

[9] T. Linsinger. Comparison of a Measurement Result with the Certified Value, https://ec.europa.eu/jrc/sites/jrcsh/files/erm_application_note_1_en.pdf (accessed: Sept 2020)

4.3 VALIDATION

4.3.1 Measurement of the CRM by the new WD-XRF method

	As		Cd		Cr		Hg		Ni		Pb	
	Δ_m	$U_{\Delta m}$	Δ_m	$U_{\Delta m}$	Δ_m	$U_{\Delta m}$	Δ_m	$U_{\Delta m}$	Δ_m	$U_{\Delta m}$	Δ_m	$U_{\Delta m}$
GeoPT-11 (OU-5 Leaton Dolerite)	0,6	2,0	-	-	2,6	4,6	-	-	1,0	2,3	0,4	1,0
GeoPT-13 (UoK LOESS)	1,6	0,8	-	-	5,6	12,1	-	-	2,7	3,0	0,3	1,3
GeoPT-40A (Calcareous organic-rich shale, ShTX-1)	1,8	6,6	0,1	1,0	4,4	5,2	-	-	2,8	4,1	10,0	19,8
Mercury Soil-2 (MS-2)	-	-	-	-	-	-	0,113	0,120	-	-	-	-
Mercury Soil-3	-	-	-	-	-	-	0,117	0,278	-	-	-	-
GBW 07103 GSR-1	0,30	0,40	-	-	0,31	0,94	-	-				
GBW 07401 Soil	2,24	4,13	0,313	0,406	3,99	5,00	-	-	1,30	2,14	3,05	6,36
GBW 07402 Soil	14,67	16,44	-	-	6,00	7,81	-	-	3,00	4,56	34,00	41,42
GBW 07403 Soil	0,8	1,34	-	-	4,8	8,94	-	-	2,51	4,12	3,00	6,46

Goodness of the method \longrightarrow $\Delta_m \leq U_{\Delta m}$

4.3 VALIDATION

4.3.2 COMPARISON BETWEEN THE RESULTS OBTAINED BY WD-XRF AND ICP-OES

There were no significant differences between the values obtained

The main difference between the two techniques was the quantification limit

	CaCO ₃ 1		CaCO ₃ 2		CaCO ₃ 3	
	WD-XRF	ICP-OES	WD-XRF	ICP-OES	WD-XRF	ICP-OES
As (mg kg ⁻¹)	<2	<1	2	2,3	<2	<1
Cd (mg kg ⁻¹)	<1	<1	<1	<1	<1	<1
Cr (mg kg ⁻¹)	4	3,6	6	6,1	5	4,2
Hg (mg kg ⁻¹)	<3	<1	<3	<1	<3	<1
Ni (mg kg ⁻¹)	<3	<1	<3	<1	<3	<1
Pb (mg kg ⁻¹)	<1	<1	<1	<1	<1	<1

4.3 VALIDATION

4.3.2 COMPARISON BETWEEN THE RESULTS OBTAINED BY WD-XRF AND ICP-OES

	KNO ₃ 1		KNO ₃ 2	
	WD-XRF	ICP-OES	WD-XRF	ICP-OES
As (mg kg ⁻¹)	<2	<1	<2	<1
Cd (mg kg ⁻¹)	<1	<1	2	2
Cr (mg kg ⁻¹)	<5	<1	<5	<1
Hg (mg kg ⁻¹)	<3	<1	<3	<1
Ni (mg kg ⁻¹)	<3	<1	<3	<1
Pb (mg kg ⁻¹)	2	2	<1	<1

There were no significant differences between the values obtained

4.3 VALIDATION

4.3.2 COMPARISON BETWEEN THE RESULTS OBTAINED BY WD-XRF AND ICP-OES

	Bentonite 1		Bentonite 2		Bentonite 3	
	WD-XRF	ICP-OES	WD-XRF	ICP-OES	WD-XRF	ICP-OES
As (mg kg ⁻¹)	6	5,4	2	2,3	13	12,3
Cd (mg kg ⁻¹)	3	2,3	3	3,1	2	2,3
Cr (mg kg ⁻¹)	24	21,5	<5	4,2	28	26,8
Hg (mg kg ⁻¹)	<3	1,5	<3	2,2	<3	1,4
Ni (mg kg ⁻¹)	10	8,5	13	13,4	12	12,3
Pb (mg kg ⁻¹)	49	48,0	29	27,5	17	15,4

There were no significant differences between the values obtained



4.4 QUANTIFICATION LIMITS ACHIEVED FOR EACH METHOD

Element	Strictest legislation limits (mg kg ⁻¹)	WD-XRF quantification limits(mg kg ⁻¹)	ICP-OES quantification limits(mg kg ⁻¹)
As	2	2	1
Cd	1	1	1
Cr	5	2	1
Hg	1	3	1
Ni	5	3	1
Pb	5	1	1

WD-XRF

- ⊗ Hg – WD-XRF method presents higher limits than the required
- ✓ As, Cd, Cr, Ni and Pb – WD-XRF method presents the same or lower quantification limits than the required

ICP-OES

- ✓ ICP-OES method reaches the quantification limits required for all the studied elements.

5. CONCLUSIONS

- A sample preparation method for WD-XRF was optimized, by using D-Mannitol as binder in a variable ratio, depending on the sample type.
- A sample preparation method for ICP-OES was optimized, depending on the element to be measured.
 - As, Cd and Hg: 1g of sample + 12 ml of inverse aqua regia
 - Cr, Ni and Pb: 0,2 g of sample + 8 ml of aqua regia + 1 ml of HF
- The analysis time involving the WD-XRF methodology was 40 min, while the analysis time of the ICP-OES methodology was about 240 min.

5. CONCLUSIONS

- The WD-XRF method is suitable for all the selected elements when the requirement is higher than the quantification limit of each element (Cd and Pb > 1 ppm, As and Cr > 2 ppm and Hg and Ni > 3 ppm)
- The ICP-OES method is suitable whenever the requirements are 1 ppm for all the elements. However, the sample preparation process needs to be the adequate one for each element, as it has been described before.
- The WD-XRF methodology developed is a fast and exact method that allows to be used as a control method. In addition, it is environmentally friendly, as it avoids the use of acids and toxic compounds to carry out the sample preparation, so it is the best one when the limit required is higher than the quantification limit.

6. ACKNOWLEDGEMENTS



6. ACKNOWLEDGEMENTS

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**GENERALITAT
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**THANKS FOR
YOUR ATTENTION**

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