#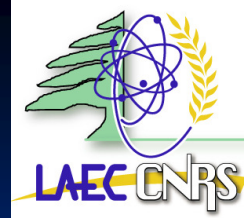 On the Direct Characterization and Quantification of Active Ingredients in Commercial Solid Drugs using PIXE, PIGE and ToF-SIMS techniques

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Content

Introduction

□ Drug

→ Active Ingredient (therapeutic molecule)

→ Excipients (inactive – major role in drug formulation)

Carrier system (to ensure the delivery of the A.I to the site of action),

Control the time of residence of the A.I, physical stability, drug dissolution,

antioxidant, coating material, binder (tablets), desintegrants,…

□ The quantification of active ingredients in drugs is a crucial step in the drug quality control process

□ Classical and universal wet chemical analytical techniques: LC\MS\MS, UV, Voltametry, etc…

➡ **If the A.I contains one or more heteroatom F, S, Cl, Br,…**

Elemental IBA (PIXE, PIGE) can be explored

➡ Advantages of IBA in Drug Quality Control (Rapidity, Precision)
  ✓ Analysis of drugs under solid form: simple sample preparation procedure
  ✓ Analysis time is few minutes/sample
  ✓ Simultaneous determination of different active ingredients
  ✓ Results can be done with good precision(< 5%)

Ability of the TT-PIXE and TT-PIGE for the quantification of heteroatom containing active ingredient in drugs

Potential of the TOF- SIMS for heteroatom free active ingredients quantification

Experimental
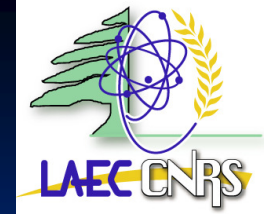
- **Sample category**: capsule, tablet, coated tablet
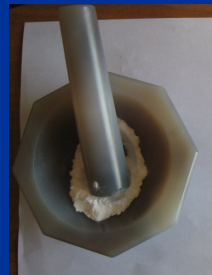

- **Sample preparation for elemental analysis (PIXE-PIGE)**

Decapsulating

Decoating

Milling

Pellet with external binder (0.1-0.2g)

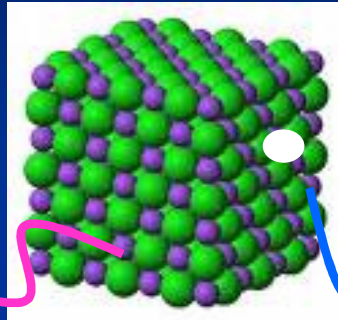Carbon coating

Sample/Al sample holder

Experimental

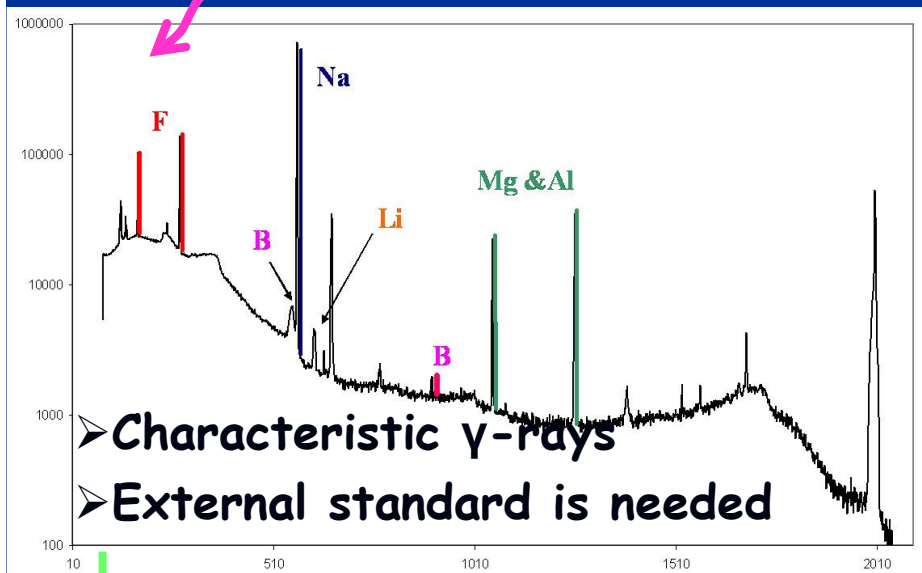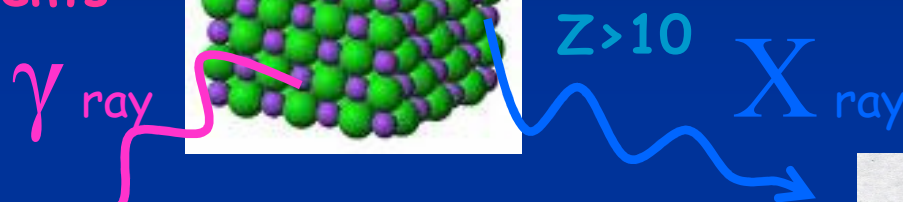NEC 5-SDH 1.7 MV LAEC-Tandem accelerator

Proton 3MeV

- **PIXE, PIGE**

Light elements

Si(Li): 170 eV at 5.9 keV
HPGe: 1.9 keV at 1.33 MeV

γ ray

Z>10

X_{ray}

> **Characteristic γ-rays**
> **External standard is needed**

> **Absolute Analysis (std less)**
> **Relative analysis: external std**

↳ **Matrix correction**

↳ **Relative quantification: If composition sample ~ composition std**

Experimental

- **ToF-SIMS technique**
  - ❖ **Ar³⁺ 9MeV delivered by the 4MV VDG accelerator of the IPNL**

  Surface analysis

  (few Å)

  - ❖ **ToF detection with high transmission**
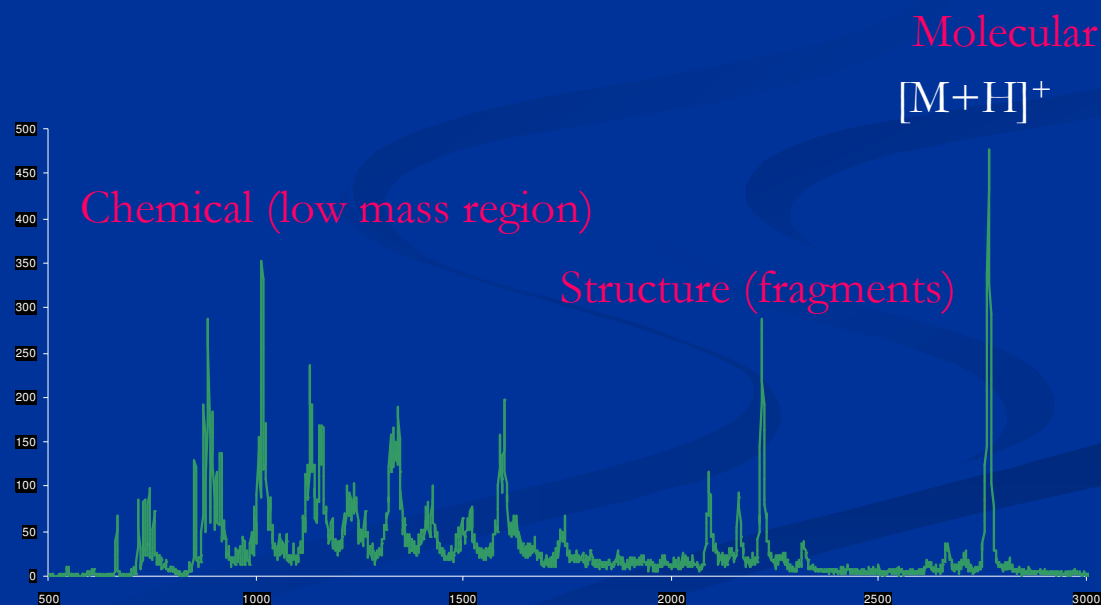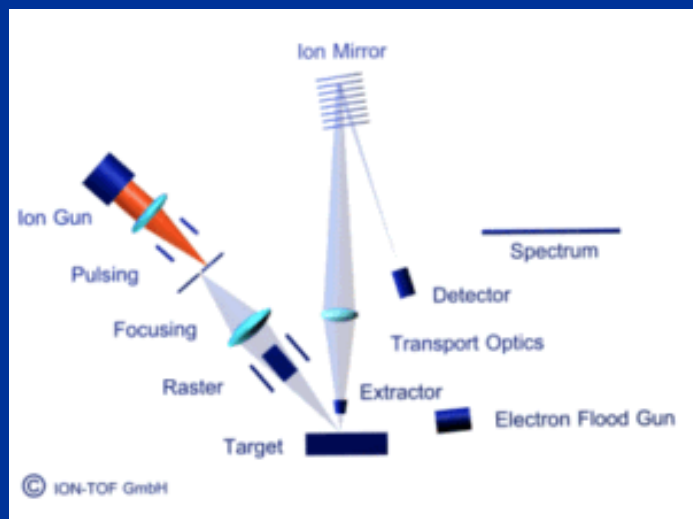 - ❖ **Ion/Ion technique (static mode ~1000-5000 ions/s over ~300 um beam diameter)**
  - ❖ **Negative and positive ion mode**
  - ❖ **Samples (thin film, pellet, ..)**
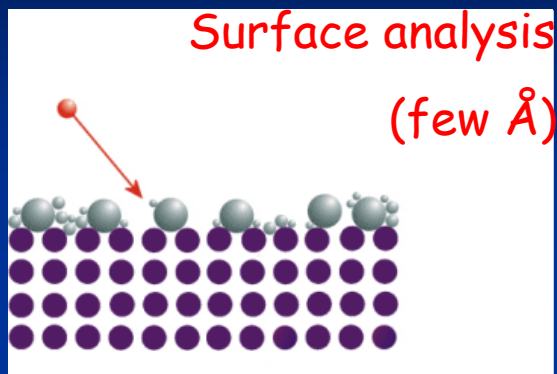
  Emission of charged characteristic secondary species

Molecular

$[M+H]^+$

Chemical (low mass region)

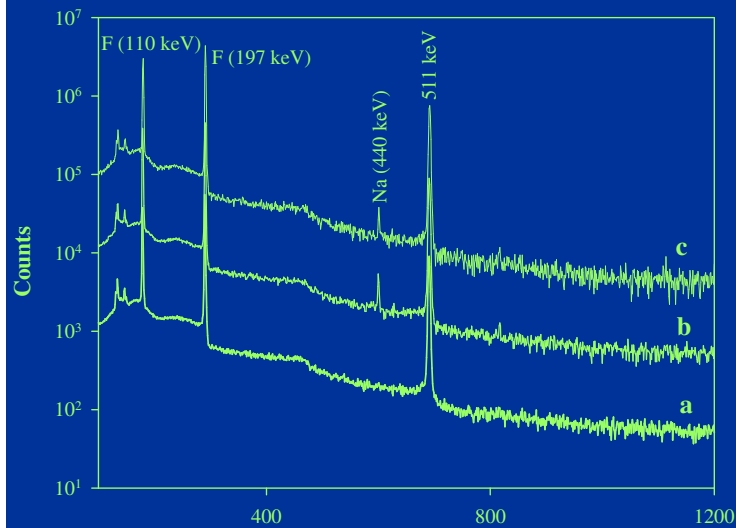Structure (fragments)
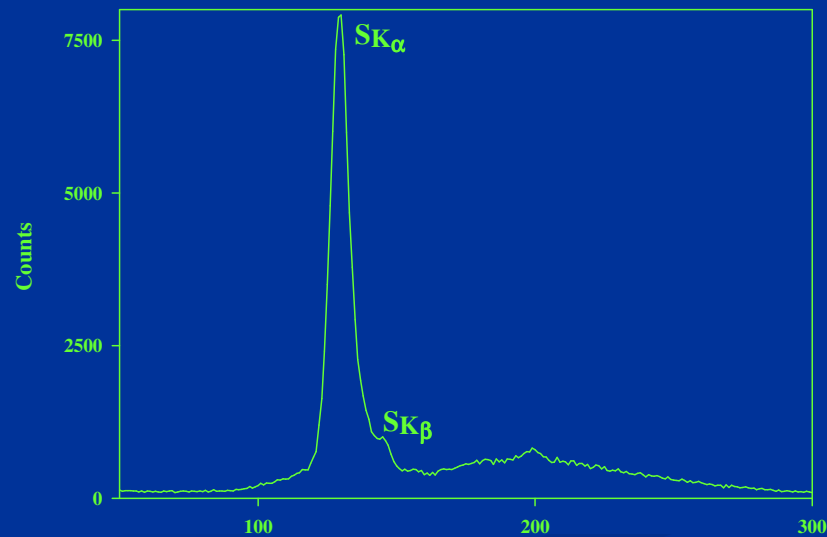
Results : Elemental analysis

CELECOXIB (F, S)
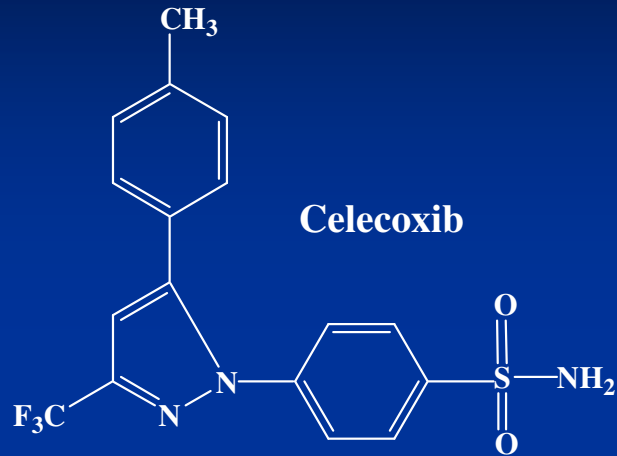
Celecoxib

- Celebrex-pfizer origin A 200mg (A.I ~ 70% - 75%)

- Celebrex-pfizer origin B 200mg

- Celex-alpha 100 mg

- Celexocib Standard (A.I = 100%)

Celecoxib, Celebrex, Celex (PIGE)

Celecoxib (PIXE)

Results : Elemental quantification

CELECOXIB (F, S)

Organic matrix : Method validation !!
-Stability under beam irradiation
-Matrix composition Std and Drugs: Matrix correction?

Stability of the matrix under ion irradiation was checked
3 Mev p+ 0.1 nA-2 nA -10 nA different acquisition time
Accumulated charge during the analysis 0.5-5µC
Dose and dose effect assessment

$$m_{A.I.} = m_{drug} \cdot \frac{I_{samp(F)}}{I_{ref(F)}}$$

→ Count rates of F and S per µC were practically stable

Matrix similarity of the Std and the analysed drugs was confirmed by RBS

Quantification by relative calculation/external standard without matrix correction

	Standard	Drug A		Drug B		Drug C	
	Celecoxib	labeled	measured	labeled	measured	labeled	measured
F	100%	72.5%	72% ± 0.5	72.5%	73% ± 0.5	68.9%	68.2% ± 0.5
S	100%	72.5%	74.7% ± 0.7	72.5%	74% ± 0.7	68.9%	68% ± 0.7

% of Celecoxib in analyzed drug

Results : Elemental analysis
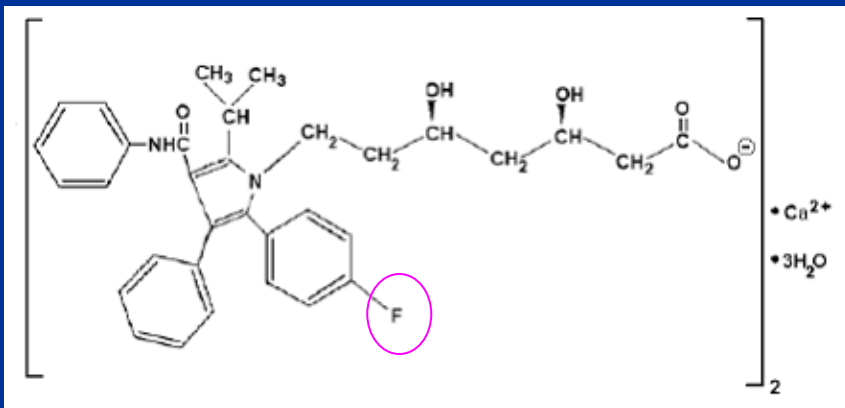
TIEMONIUM METHYLSULFATE (S)
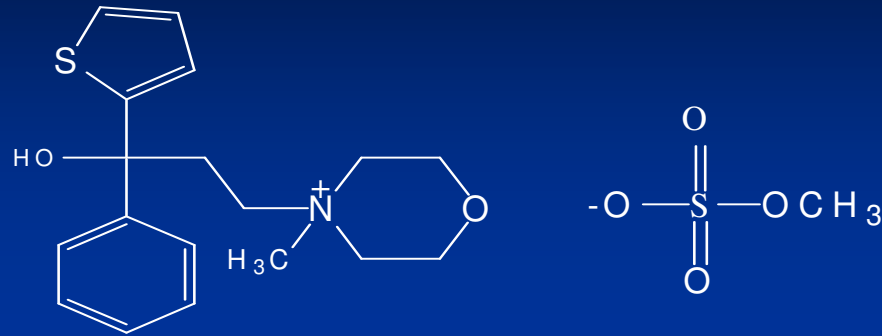
- Timozin 50 mg (A.I ~ 16%)
- TIE Standard (A.I. = 100%)

Labeled

50 mg TIE

PIXE relative quantification: External Standard

60 mg ± 1.2

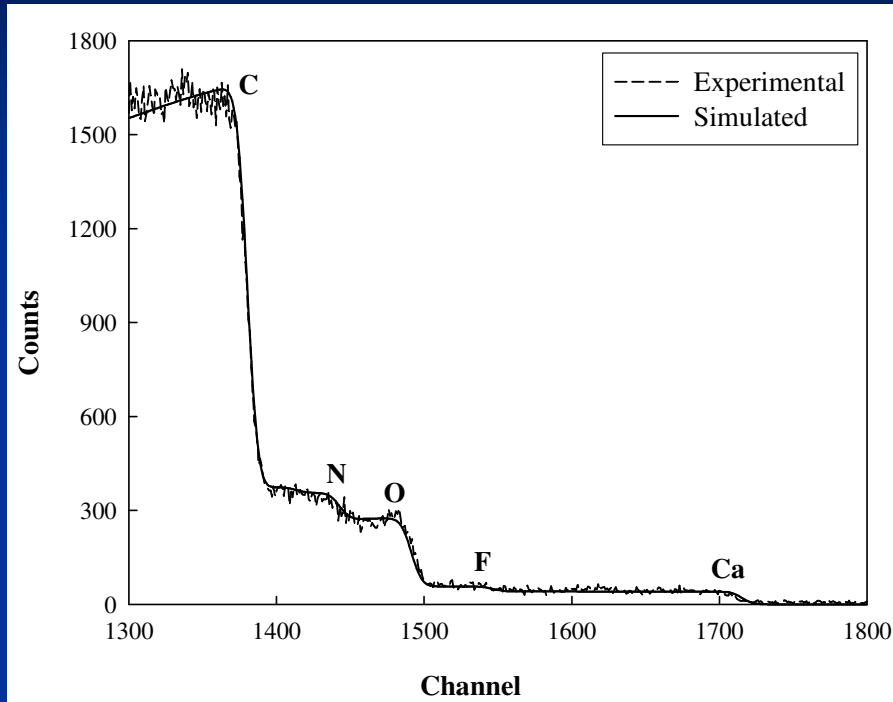Matrix correction (PIXE absolute quantification)

48.7mg ± 1.1

ATORVASTATIN (F)

- Storvas 10 mg
- Lipinorm 10 mg
- Lipitor 10mg

(A.I ~ 6.5%)

Results : Elemental analysis

ATORVASTATIN (F)

Elemental composition of the Atorvastatin standard **(100 % atorvastatin)** is significantly <u>different</u> from the composition of the analyzed drug samples (RBS measurements)
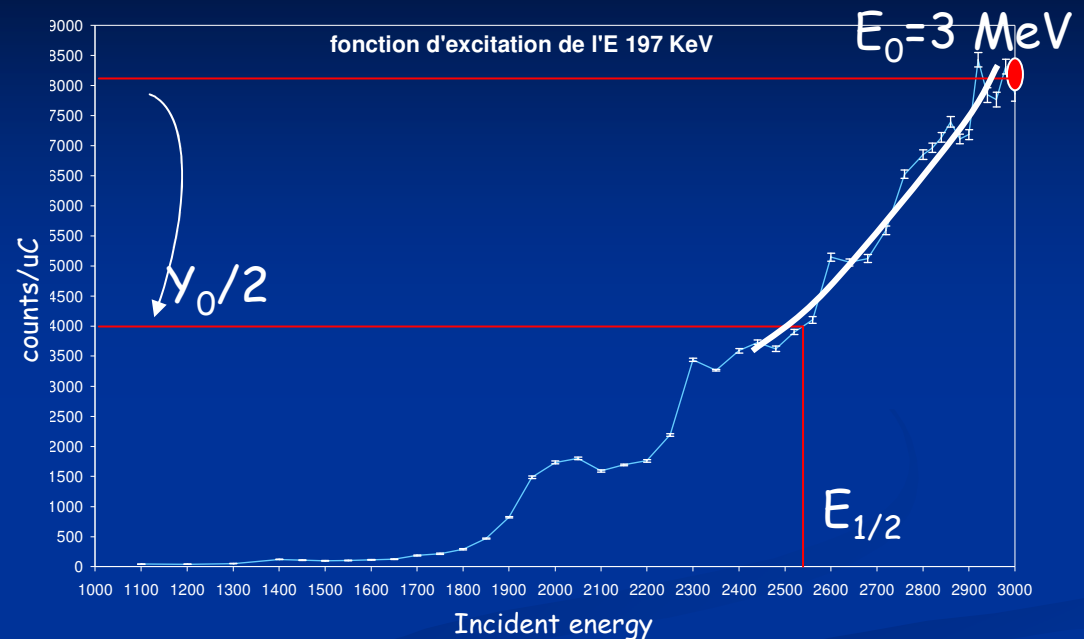
Elemental composition %											
	C_t	C_m	H_t	H_m	F_t	F_m	N_t	N_m	O_t	O_m	Ca_t	Ca_m
Atorvastatin std.	68.61	69.42	5.93	6.00	3.29	3.29	4.85	4.78	13.85	12.82	3.47	3.69
Lipitor®		40.3		6.76		0.25		4.16		35.6		12.9

Results : Elemental analysis

ATORVASTATIN (F)

$$\frac{Y_{Samp}}{Y_{Ref}} = \frac{C_{Samp}}{C_{Ref}} * \frac{S_{Ref}(E_{1/2})}{S_{Samp}(E_{1/2})}$$

fonction d'excitation de l'E 197 KeV

E_0=3 MeV

$Y_0/2$

$E_{1/2}$

counts/uC — Incident energy

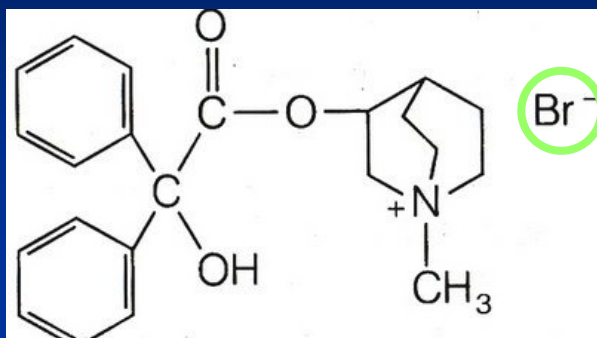| Atorvastatin Active Ingridient (mg)								
Storvas® 10 mg			Lipitor® 10 mg			Lipinorm® 10 mg		
Measured relatively/std	measured with matrix correction	UV	Measured relatively/std	measured with matrix correction	UV	Measured relatively/std	measured with matrix correction	UV
12.5±0.3	11.2±0.3	10.9±0.4	11.9±0.3	11.0±0.3	10.8±0.2	12.1±0.2	11.0±0.3	11.0±0.1

PIGE RESULTS ARE IN A GOOD AGREEMENT WITH UV MEASUREMENTS
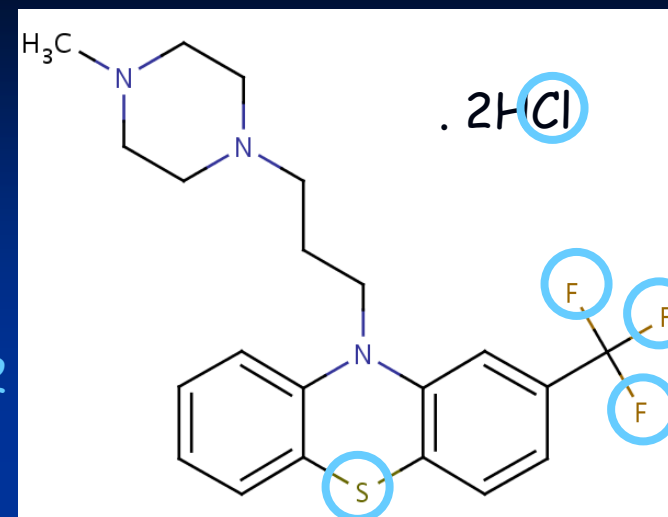
Results : Elemental analysis
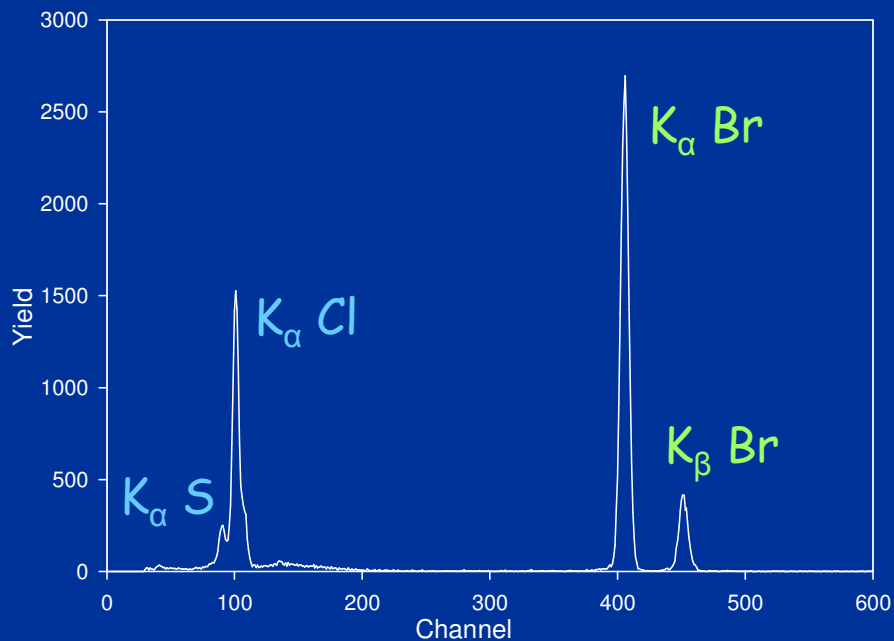
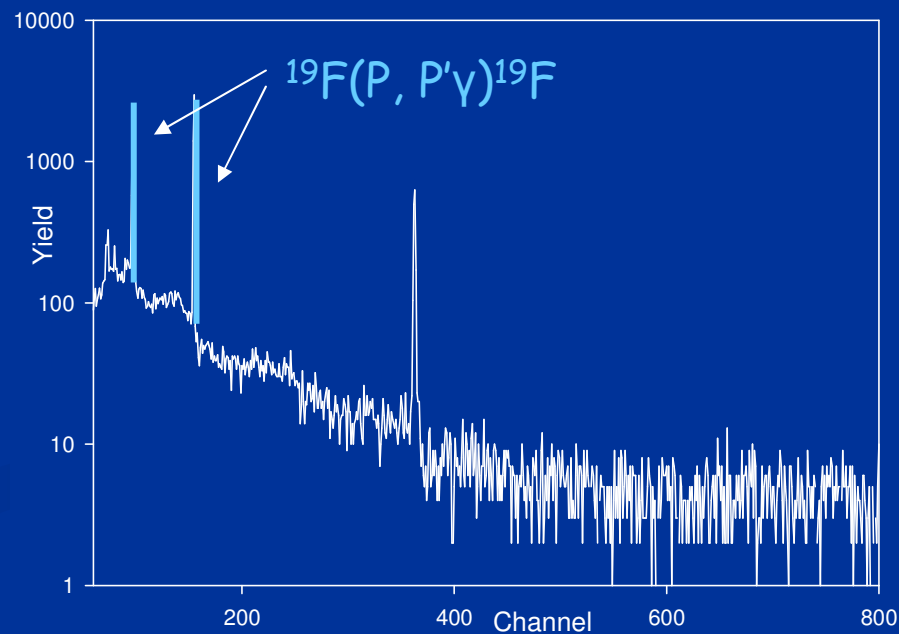**FLUDINIUM (Br, S, Cl, F)**

Binary drug

A.I.1

A.I.2

Clidinium bromide

Trifluoperazine

. 2HCl

Fludinium® spectra

PIXE

K_α Br

K_α Cl

K_β Br

K_α S

Yield

Channel

PIGE

$^{19}F(P, P'\gamma)^{19}F$

Yield

Channel

Results : Elemental analysis

FLUDINIUM (Br, S, Cl, F)

Analysed Samples:
-Standards with different % of the two A.I. and excipients.
-Fludinium commercial drug with different compositions

Stability under irradiation:

→Samples are stable under analysis conditions (3MeV, 0.2 nA, 0.1µC,15 min acquisition)
→Loss of Cl under ion irradiation even under condition of analysis
→Cl is not related to thev A.I by covalent band it is thermo fragile.

Cl will not considered for quantification
S, F: quantification of A.I.2
Br: quantification of A.I.1

Results : Elemental analysis

FLUDINIUM (Br, S, Cl, F)

VALIDITY OF THE ANALYSIS

❑ S direct quantification in drugs/standard is highly affected by matrix composition of the analyzed drugs. Matrix correction is needed (A.I.2)
❑ Br can be quantified directly in drugs/standard (A.I 1)
❑ F can be quantified directly in drugs/standard (A.I 2)

Stopping power

Matrix composition
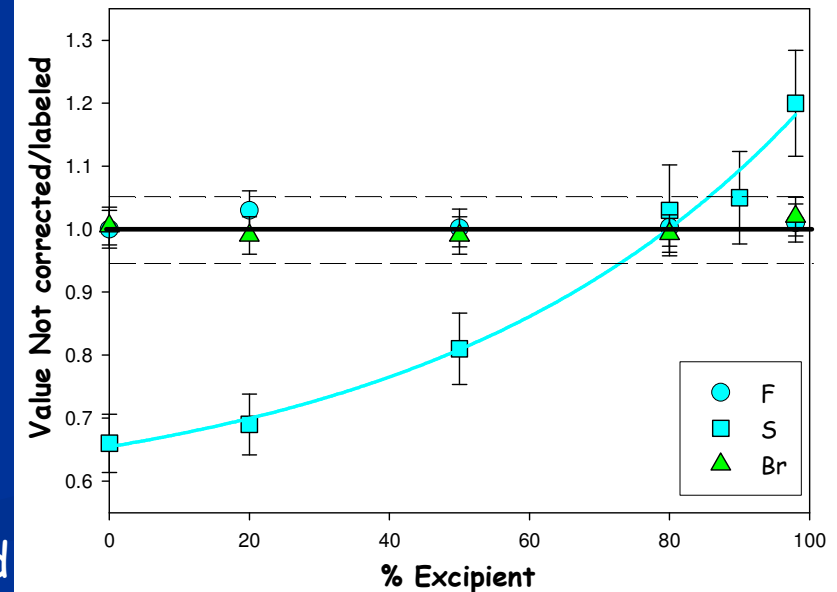
Absorption Coefficient

K_α (S)=2.3 keV: highly affected by variation of matrix absorption coefficient

K_α (Br) =11.9 keV

γ (F)= 197 KeV

Practically not affected

Results : Elemental analysis

FLUDINIUM (Br, S, Cl, F)

Sample labeled composition	S (matrix correction) PIXE-Gupix	F (direct analysis) PIGE	Br (direct analysis) PIXE
50mg A.I.1, 50mg A.I.2 3.33% S, 5.9% F, 9.25% Br	3.37%±0.16	5.7% ±0.2	9.4% ±0.2
40mg A.I.1, 40mg A.I.2, 20 mg Excipient 2.67% S, 4.7% F, 7.4% Br	2.60%±0.12	4.61% ±0.22	7.47% ±0.22
25mgA.I.1, 25mgA.I.2, 50mg Excipient 1.67% S, 2.97% F, 4.6% Br	1.62%±0.03	2.73% ±0.13	4.66% ±0.14
12.2mg A.I.1, 4.9mgA.I.2, 82.9mg Excipient 0.32% S, 0.58% F, 2.25% Br	0.31%±0.01	0.57% ±0.03	2.26% ±0.07
7.9mg A.I.1, 2.6mg A.I.2, 89.5mg Excipient 0.175% S, 0.31% F, 1.46% Br	0.18%±0.01	0.35% ±0.02	1.53% ±0.04
0.58mg A.I.1, 1.45mg A.I.2, 97.2mg Excipient 388ppm S, 691ppm F, 2692ppm Br	434ppm±43	705ppm±35	2943ppm±147

Results : Molecular analysis (ToF-SIMS)

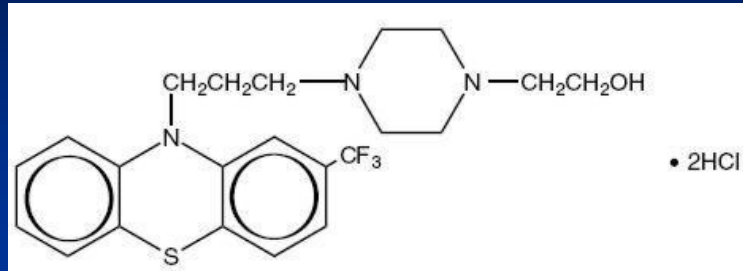➢ **Larger number of A.I. are <u>heteroatom free</u>.**

✧ Molecular characterization in stead of element analysis
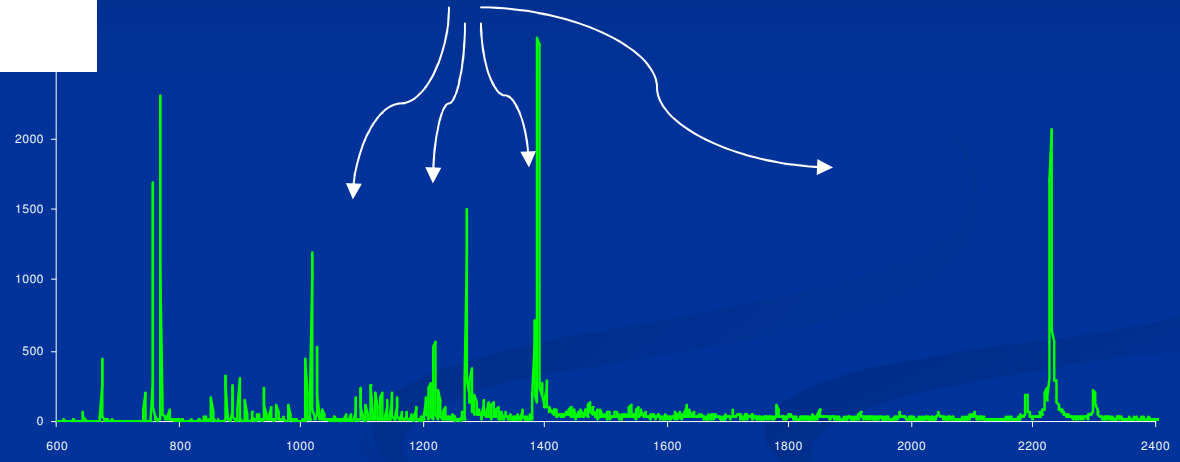
✧ Surface analysis – Semi quantitative

✧ The secondary Ion Emission is highly dependent on the matrix (composition, texture, inter and intra molecular interaction, surface structure)
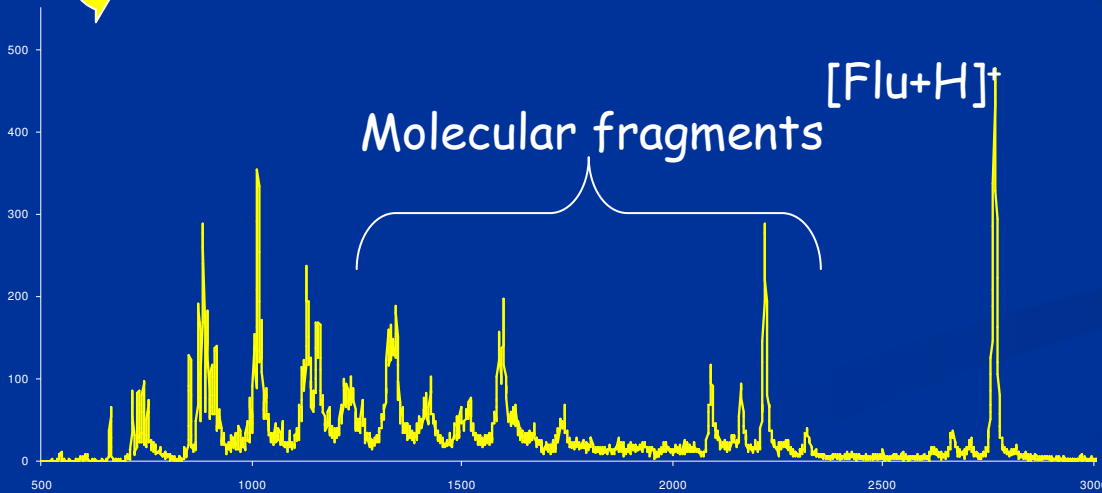
Results : ToF-SIMS

FLUHENAZINE DIHYDROCHLORIDE

· 2HCl

Excipients are trade secret

Positive emission

Fluphenazine
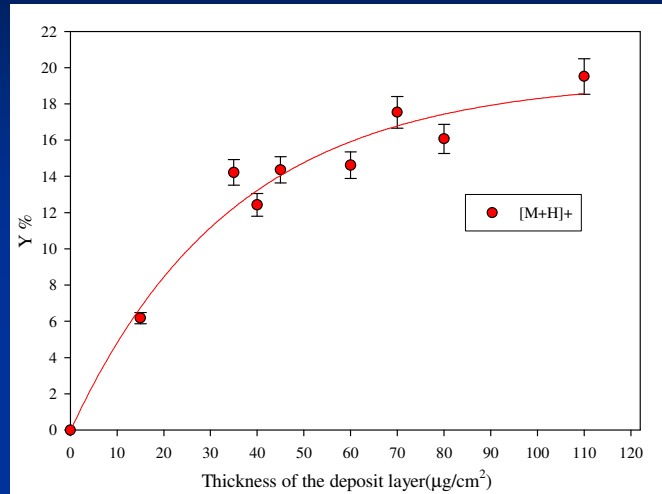Finger print

Molecular fragments

[Flu+H]⁺

Negative emission

High
emission of
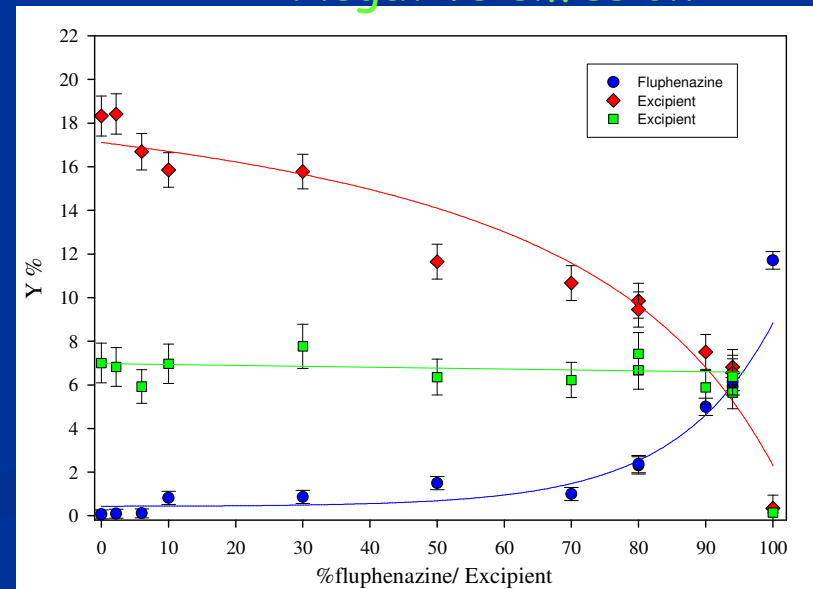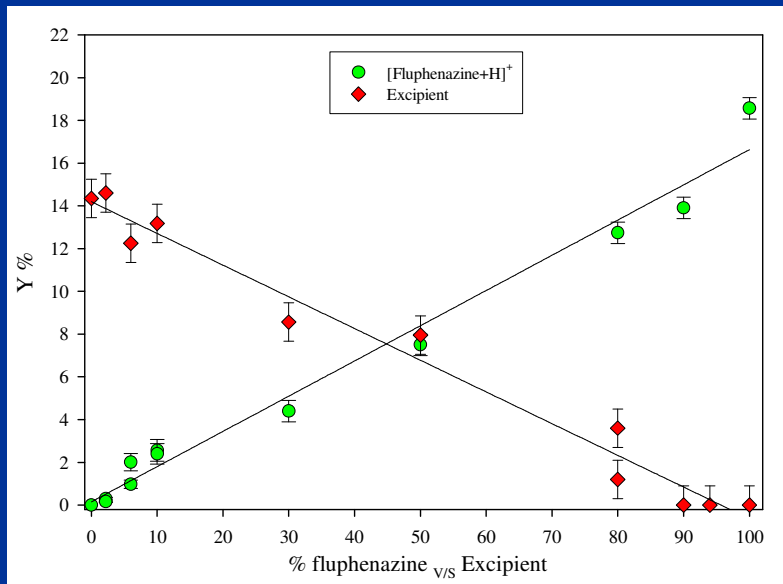Excipient

Results : ToF-SIMS

FLUHENAZINE DIHYDROCHLORIDE

Any semi-quantitation needs that the thickness of the sample > secondary ion escape depth
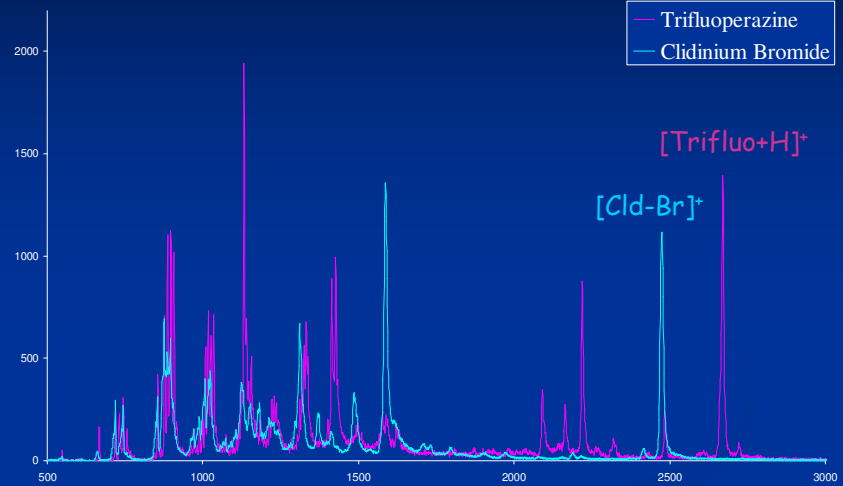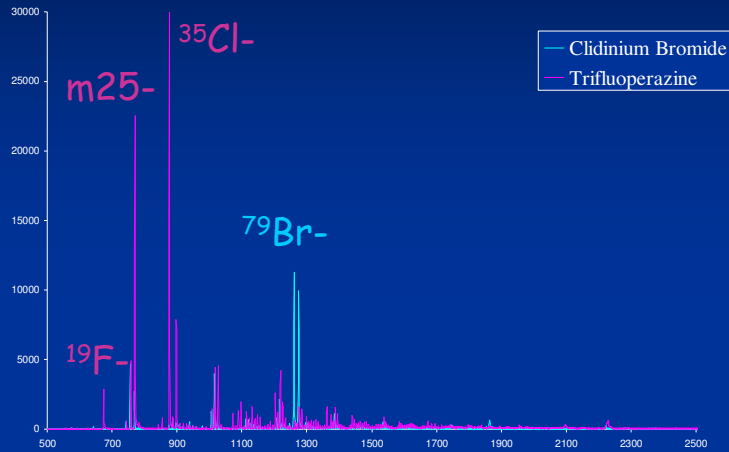
In positive emission semi-quantification can be extracted

Negative emission
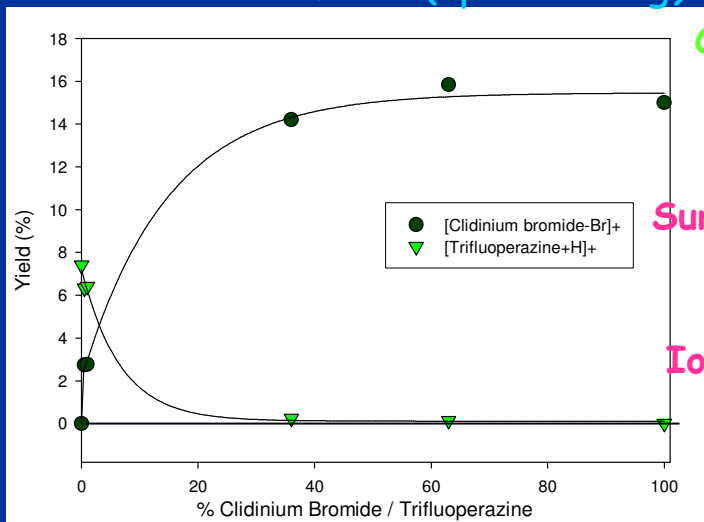
Results : ToF-SIMS

FLUDINIUM (BINARY DRUG)

2.5 mg Clidinium Br, 1 mg Trifluoperazine

Positive emission is rich in molecular and chemical structure

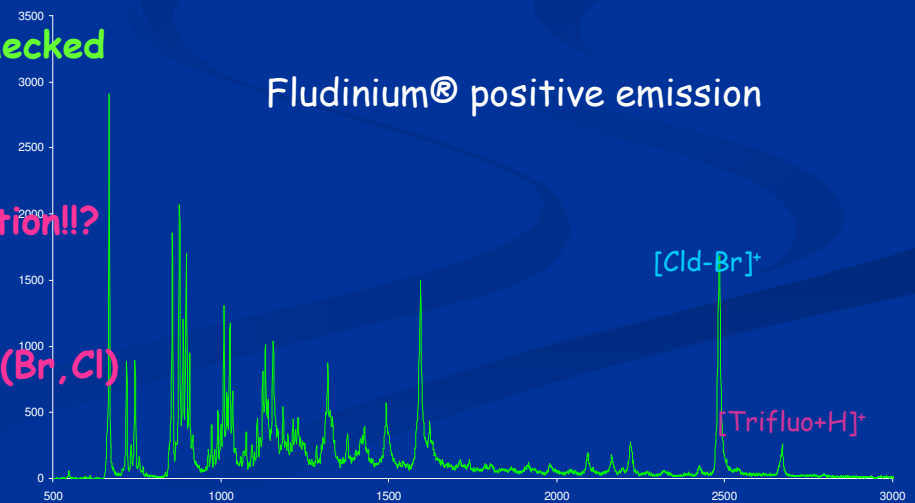Semi-quantification in a prepared mixture as thin films (spin coating)???

Composition checked by RBS

Fludinium® positive emission

Surface segregation!!?

Ion Microscopy (Br,Cl)

ESCA

Conclusion and Overview

- PIXE and PIGE techniques are appropriate for rapid and accurate quantification of A.I. containing heteroatoms like S, F, Cl, Br.

- Methode validation: - Ensure the stability of the sample under ion irradiation, - Elemental composition of the standard and the analyzed drugs. – Quantification was validated with or without matrix correction.

- TOF-SIMS can be promising technique for A.I. heteroatom free characterization

- Effort to be done in the sample preparation techniques to ensure analytical reproducibility and trueness (ToF-SIMS)

- A.I. determination in liquid form (syrup) to be analyzed as thin film

- Elemental and molecular techniques for analysis of solid drug with several A.I. (>3A.I. with low concentration ppm range)

- TOF-SIMS Exploration of the use of internal standards for direct quantification (d-labeled A.I. or chemically similar molecule(s) to A.I.)

Thank you for you attention