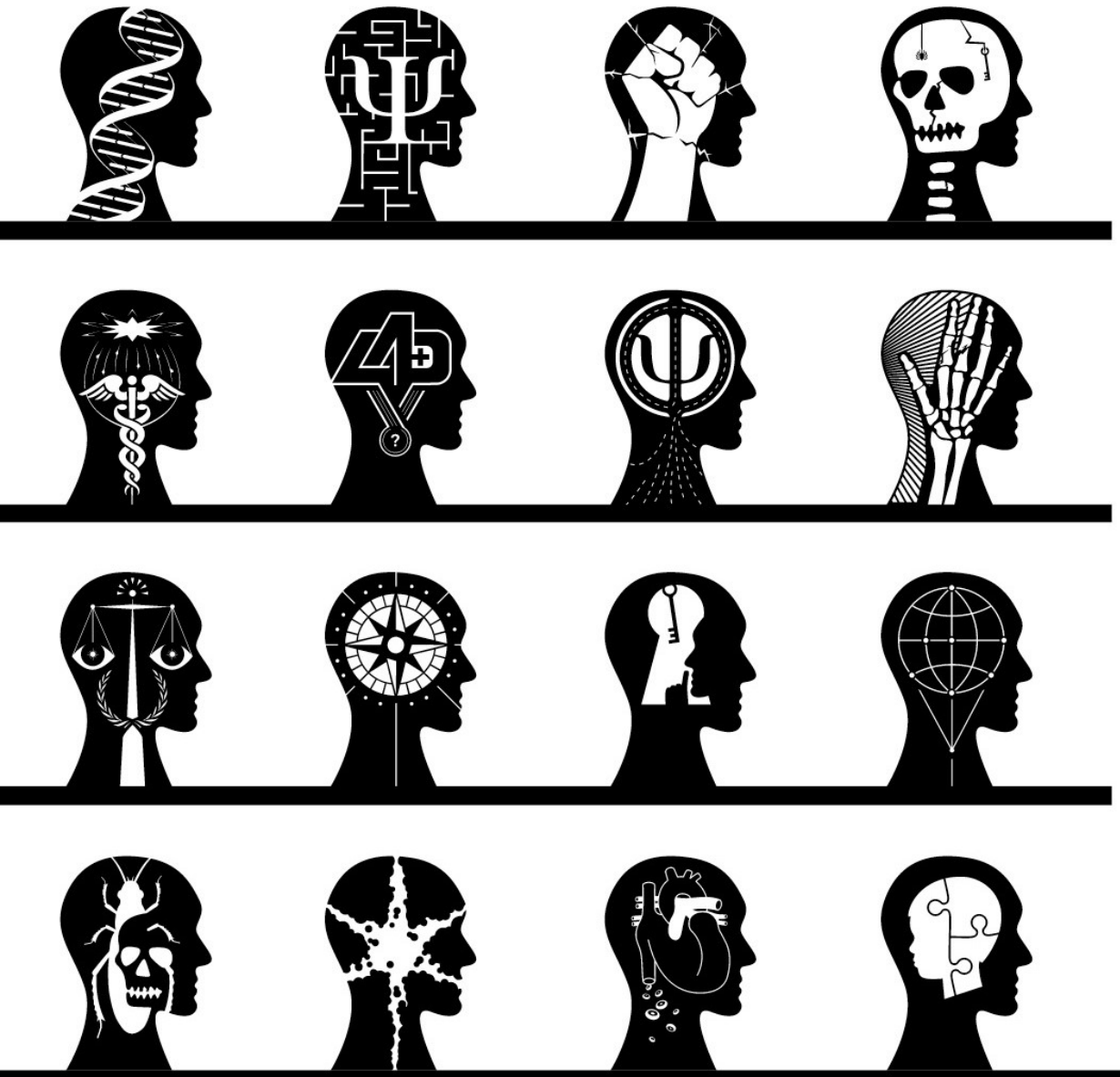


Centre
Universitaire
Romand

Médecine
Légale

Spectrométrie de masse à haute ou basse résolution dans l'analyse antidopage. Est-ce vraiment le point de non-retour ?

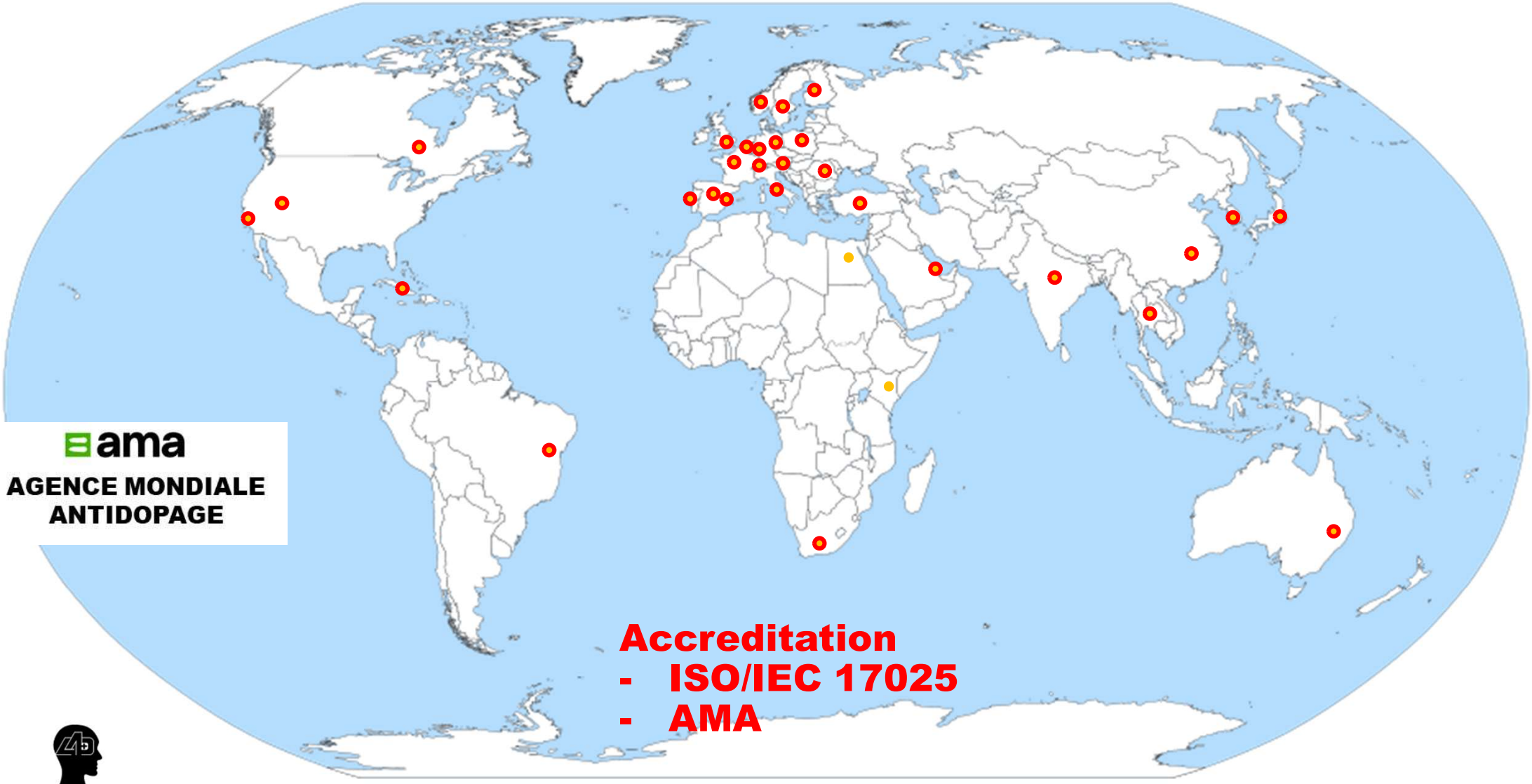
Alessandro Musenga
Laboratoire Suisse d'Analyse du Dopage

02 octobre 2025
Journées Scientifiques - ccCTA

LC-HRMS dans l’antidopage

- **Est-il permis d'utiliser la spectrométrie de masse à haute résolution dans l'analyse antidopage?**
- **La spectrométrie de masse à haute résolution est-elle couramment utilisée dans l’analyse antidopage ?**
- **Pour quelles applications la spectrométrie de masse à haute résolution est-elle utilisée dans l’analyse antidopage ?**
- **La spectrométrie de masse à haute résolution a-t-elle complètement remplacé les instruments à triple quadripôle ?**

Les laboratoires accrédités par l'AMA



ama
AGENCE MONDIALE ANTIDOPAGE

Accreditation
- **ISO/IEC 17025**
- **AMA**

Les règles de l’AMA pour l’utilisation de la spectrométrie de masse

Les règles de l'AMA pour l'utilisation de la spectrométrie de masse

5.3.6.2.1 Confirmation Procedure Methods

Mass spectrometry (MS) coupled to chromatographic separation (*e.g.* gas or liquid chromatography) is the analytical technique of choice for confirmation of most *Prohibited Substances*, *Metabolite(s)* of a

cified in a *Technical Document*, Technical Letter or Laboratory Guidelines

WORLD ANTI-DOPING CODE
INTERNATIONAL STANDARD
LABORATORIES
2021

wada

WADA Technical Document – TD2023IDCR

Document number:	TD2023IDCR	Version number:	1.1
Written by: Reviewed by:	WADA Science / IDCR Working Group *WADA* Laboratory Expert Group	Approved by:	*WADA* Executive Committee
Date:	14 February 2023	Effective date:	14 February 2023

MINIMUM CRITERIA FOR CHROMATOGRAPHIC-MASS

3.2. Requirements

MS criteria for identification by either scanning (*e.g.*, Full Scan, Product Ion Scan) or non-scanning [*e.g.*, Selected Ion Monitoring (SIM), Selected Reaction Monitoring (SRM)] techniques are based on the presence and RA of a number of Diagnostic Ions which has been validated by the Laboratory as diagnostic for the Analyte. Any data processing (*e.g.*, integration, subtraction, averaging, *etc.*) shall be performed consistently across the analytical batch.

Depending on the Analytical Method, the concentration of the Analyte may need to be comparable (*i.e.*, signal of the Analyte within one order of magnitude) in the *Sample* and the Reference Specimen to ensure identification of the Analyte.

The following identification criteria shall be applied:

- Each measured mass used for identification shall be within ± 0.5 Da of the corresponding mass of the same Diagnostic Ion acquired from the Reference Specimen analyzed in the same analytical batch;
- When using single-stage MS, at least three (3) Diagnostic Ions shall be acquired;
- When using multiple-stage MS (*e.g.*, MS/MS), at least two (2) Diagnostic Ions [*i.e.*, two precursor-product ion transitions (SRM transitions)] shall be acquired. The isolation width of the precursor ion shall not be more than (≤) *m/z* 1.3, unless required by its molecular mass and charge state;
- The Signal-to-Noise (S/N) ratio of all Diagnostic Ions shall be greater than (>) three to one (3:1);
- The abundance of Diagnostic Ions shall be determined from the peak area or height in the integrated selected ion chromatograms. This is also applicable when Scan technique only is used for identification;
- The most abundant Diagnostic Ion acquired from a Reference Specimen is the Reference Diagnostic Ion, which shall be applied to the calculation of the RAs. The same ion shall be applied

LC-HRMS dans l’antidopage

- **Est-il permis d'utiliser la spectrométrie de masse à haute résolution dans l'analyse antidopage?**
- **La spectrométrie de masse à haute résolution est-elle couramment utilisée dans l’analyse antidopage ?**
- **Pour quelles applications la spectrométrie de masse à haute résolution est-elle utilisée dans l’analyse antidopage ?**
- **La spectrométrie de masse à haute résolution a-t-elle complètement remplacé les instruments à triple quadripôle ?**

Screening ou confirmation: quelle est la charge majeure pour un laboratoire?

Echantillons négatives

- Tous les échantillons
- (20000/an au LAD)

Initial testing procedure (ITP)

Screening

Echantillons suspects

(1-2% du total)

Confirmatory procedure (CP)

Méthodes de confirmation

Statistiques 2022 - AMA

Sport	Analyzed	*AAFs* [1]	(%)	*ATFs* [2]	(%)	*Total Findings* [3]	(%)
Olympic Sports [4]	218,774	1,222	0.56%	316	0.14%	1,538	0.70%
Non-Olympic Sports [5]	37,995	764	2.01%	78	0.21%	842	2.22%
TOTAL	**256,769**	**1,986**	0.77%	**394**	0.15%	**2,380**	0.93%

Evolution des techniques GC et LC dans l'antidopage

GC-NPD

GC-MS

HPLC

LC-MS

GC-C-IRMS

LC-MS/MS

1972-1976 | 1984 | 1988-1992 | 1994-1998 | 2004-2008

GC-NPD
GC-MS

HPLC

LC-MS
Glucocorticoïdes, diurétiques

GC-C-IRMS
Stéroïdes endogènes

LC-MS/MS

2004 - la LC devient un équipement standard dans les laboratoires

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Analytica Chimica Acta 573–574 (2006) 242–249

ANALYTICA CHIMICA ACTA

www.elsevier.com/locate/aca

Doping control analysis in human urine by liquid chromatography–electrospray ionization ion trap mass spectrometry for the Olympic Games Athens 2004: Determination of corticosteroids and quantification of ephedrines, salbutamol and morphine

M.-H. Spyridaki [a], P. Kiousi [a], A. Vonaparti [a], P. Valavani [a], V. Zonaras [a], M. Zahariou [a], E. Sianos [a], G. Tsoupras [b], C. Georgakopoulos [a,*]

[a] *Athens Doping Control Laboratory, Olympic Athletic Center of Athens "Spiros Louis", 37 Kifissias Ave., 151 23 Maroussi, Athens, Greece*
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Received 30 November 2005; received in revised form 17 April 2006; accepted 20 April 2006
Available online 27 April 2006

Ionisation difficile en GC-MS

Substances polaires

Abstract

A new liquid chromatography–electrospray ionization-mass spectrometry (LC–ESI–MS) (*n*) ion trap method for the determination of corticosteroids in urine has been developed and validated. Some anabolic agents, such as epitrenbolone, trenbolone, 2-hydroxymethylformebolone, tetrahydrogestrinone, gestrinone and formoterol were included in the LC–ESI–MS method. Matrix interference, specificity, identification capability, carry over and robustness were estimated as validation parameters. Recoveries ranged from 74 to 113% at the minimum required performance limit (MRPL), which is 30 ng mL^{-1} for corticosteroids and 10 ng mL^{-1} for anabolic agents. Methods for the confirmation and quantification of norpseudoephedrine, ephedrine, methylephedrine, salbutamol, morphine and morphine glucuronide were also developed and validated and in order to minimize analysis time, direct urine injection was used. These methods proved to be specific, accurate and precise across a calibration range for each substance since matrix interference, specificity, carry over, within and between run precision, limit of detection, limit of quantification, intermediate precision and uncertainty were estimated.

Keywords: Liquid chromatography–electrospray ionization ion trap mass spectrometry; Antidoping; Corticosteroids; Ephedrines; Salbutamol; Morphine

2008 - la MS à haute-résolution fait ses débuts dans l'antidopage

JOURNAL OF MASS SPECTROMETRY
J. Mass Spectrom. 2008; **43**: 949–957
Published online 19 June 2008 in Wiley InterScience
(www.interscience.wiley.com) DOI: 10.1002/jms.1447

JMS

Introduction of HPLC/orbitrap mass spectrometry as screening method for doping control

E. D. Virus,* T. G. Sobolevsky and G. M. Rodchenkov

Moscow Antidoping Center, Elizavetynsky10, 105005, Moscow, Russia

Received 10 April 2008; Accepted 10 May 2008

A new doping control screening method has been developed, for the analysis of doping agents in human urine, using HPLC/orbitrap with in-source collision-induced dissociation and atmospheric pressure chemical ionization. The developed method allows the detection of 29 compounds including agents with antiestrogenic activity, β_2 agonists, exogenous anabolic steroids, and other anabolic agents. The mass accuracy of this method is better at 2 ppm using an external reference. The detection limit for all compounds tested was better than 100 pg/ml. The recoveries of most analytes were above 70%. The measured median repeatability values for doping agents included in the method at concentrations of 1 and 10 ng/ml were 21 and 17%, respectively. The relative standard deviation (RSD) of the intraday precision (n = 6) ranged from RSD = 16–22%, whereas the interday precision (n = 18), ranged from RSD = 17–26%, depending on the solute concentration investigated.

2008 - la MS à haute-résolution fait ses débuts dans l'antidopage

JOURNAL OF MASS SPECTROMETRY
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JMS

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Received 10 April 2008; Accepted 10 May 2008

A new doping control screening method has been developed, for the urine, using HPLC/orbitrap with in-source collision-induced diss chemical ionization. The developed method allows the detection with antiestrogenic activity, β_2 agonists, exogenous anabolic steroids, accuracy of this method is better at 2 ppm using an external reference. T tested was better than 100 pg/ml. The recoveries of most analytes wer repeatability values for doping agents included in the method at conc and 17%, respectively. The relative standard deviation (RSD) of the in RSD = 16–22%, whereas the interday precision (n = 18), ranged from solute concentration investigated.

CONCLUSIONS

Although the sensitivity is similar for the HPLC/MS/MS and HPLC/HRMS techniques, the latter technique has the advantage of acquiring full scan MS spectra with a very good sensitivity. The proposed method also matches the basic requirements of all methods used to analyze drugs or metabolites in an antidoping laboratory, i.e. the sensitivity, selectivity, specificity, and speed of analysis. In conclusion, this system is sufficiently robust to carry out 300 analyses without routine maintenance. More than 1500 analyses have been carried out using this instrument. We

Screening par LC-HRMS - avantages

- **Acquisition full scan : la méthode d'acquisition devient "untargeted"**

- Acquisition de données brutes sans nécessité d'une connaissance préalable des composés cibles

- La sélection des analytes se fait au moment du traitement des données

Il est très simple d'ajouter des nouveaux composés à la méthode

(aucune modification de la méthode d'acquisition)

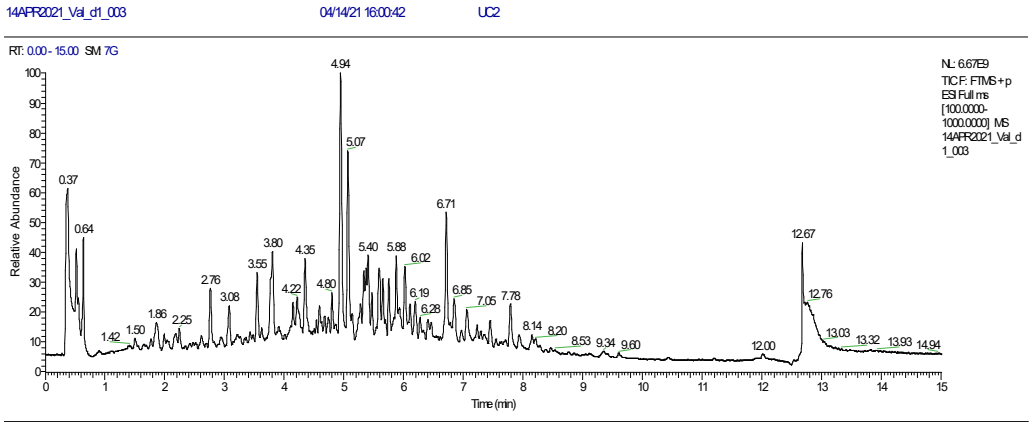
Analyse rétrospective

(traitement des données précédemment acquises pour rechercher de nouveaux composés)

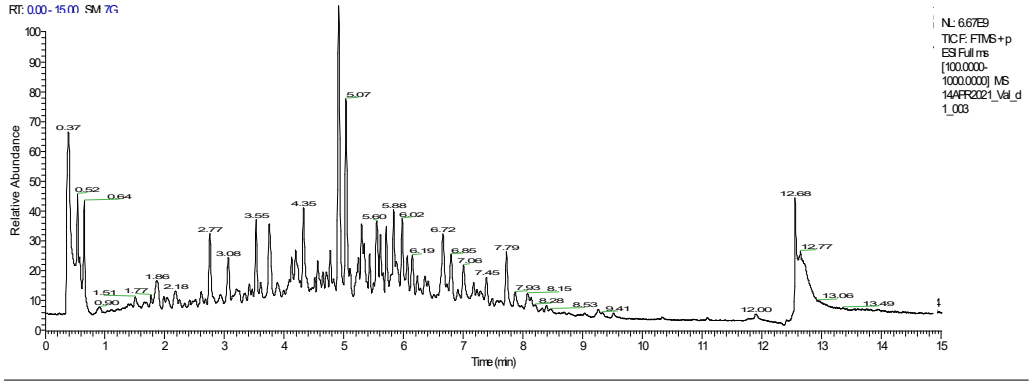
Full scan - trop de pics dans le TIC…

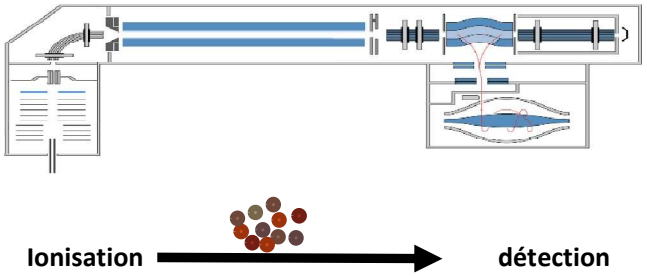
Total ion chromatogram (TIC)

Contrôle positif



Urine négative



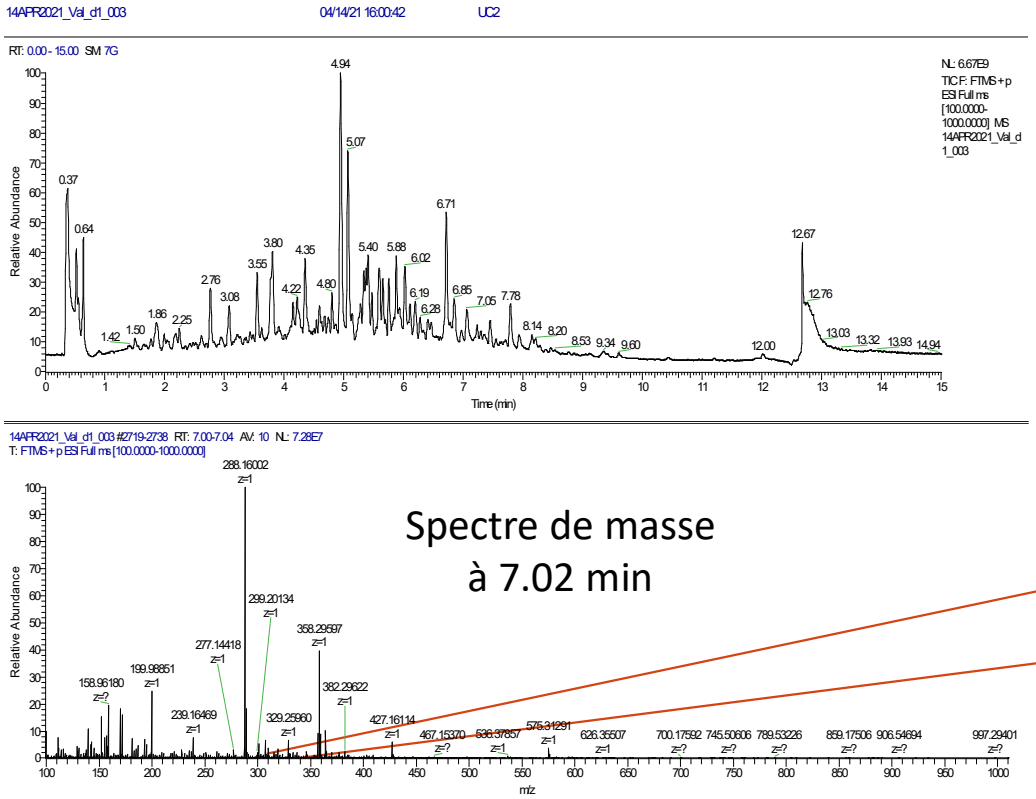


Acquisition : full scan

Exemple: Analyse du stanozolol
m/z : 329.2587, RT: 7.02 min

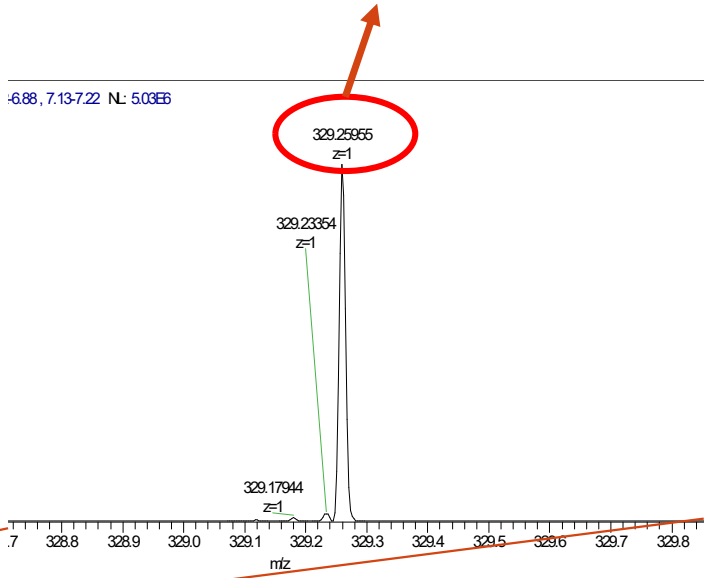
Screening par LC-HRMS

Total ion chromatogram (TIC)



Spectre de masse
à 7.02 min

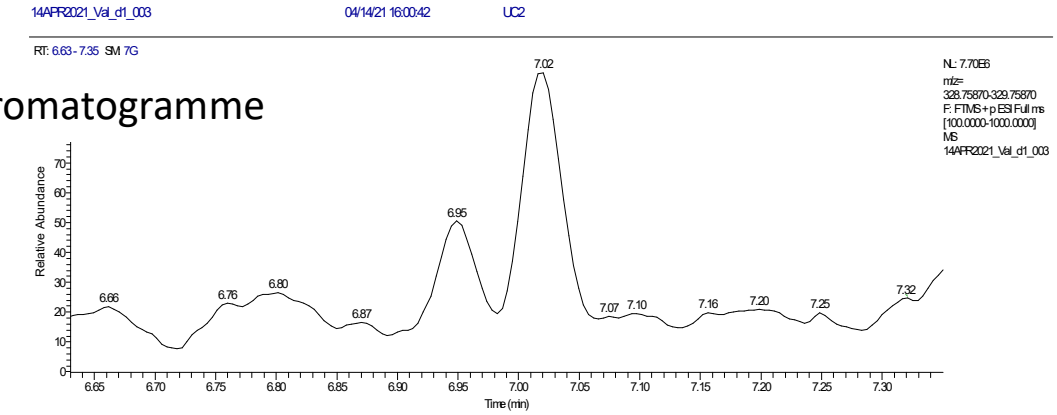
m/z du stanozolol



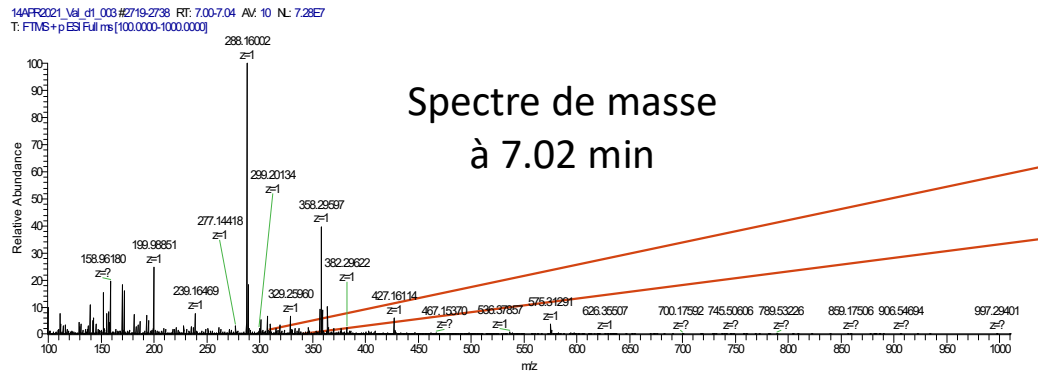
Screening par LC-HRMS

Extracted ion chromatogram (XIC)
m/z range ± 0.5 Da (328.7587 – 329.7587)

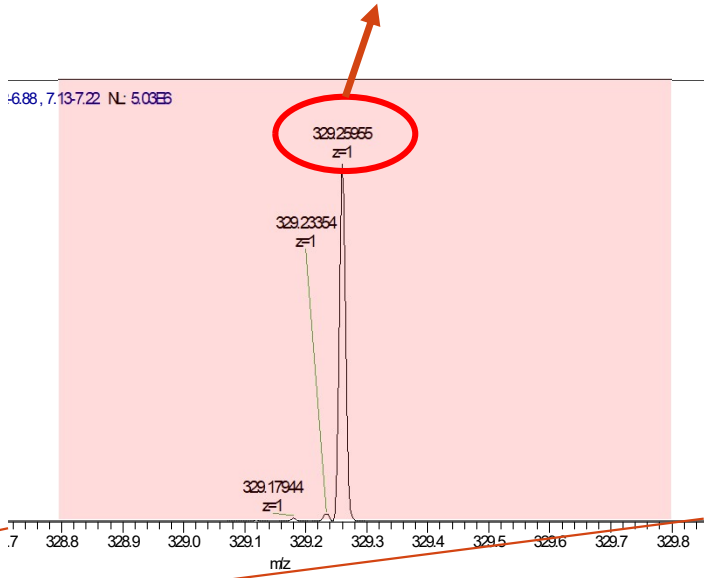
Chromatogramme



Spectre de masse
à 7.02 min



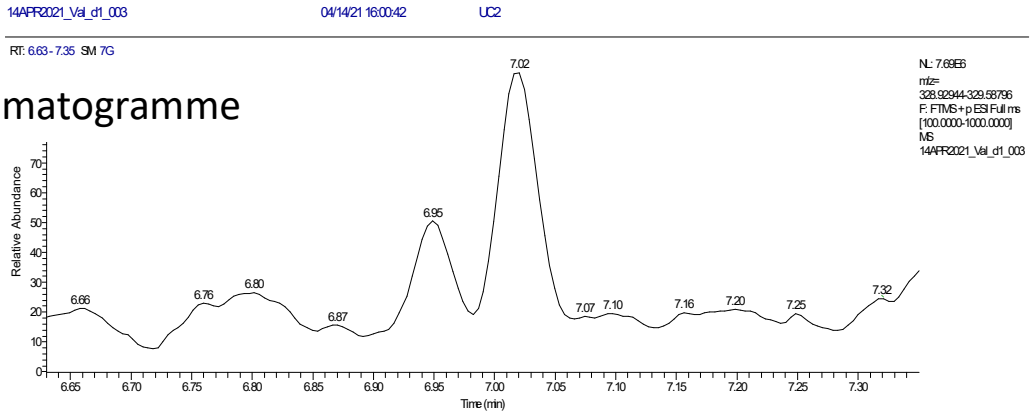
m/z du stanozolol

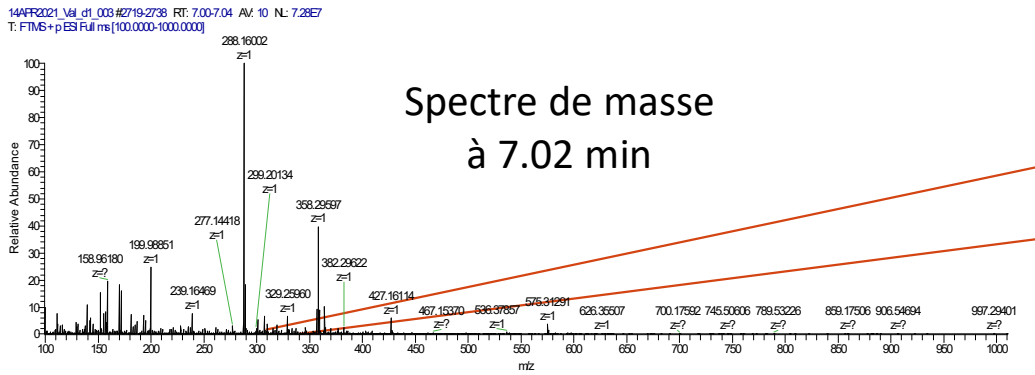


Screening par LC-HRMS

Extracted ion chromatogram (XIC)
m/z range ± 1000 ppm (328.9294 – 329.5880)

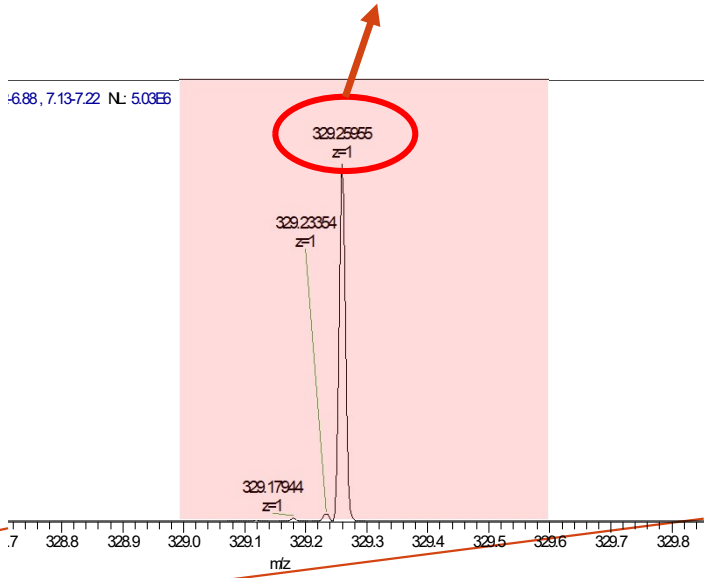
Chromatogramme





Spectre de masse
à 7.02 min

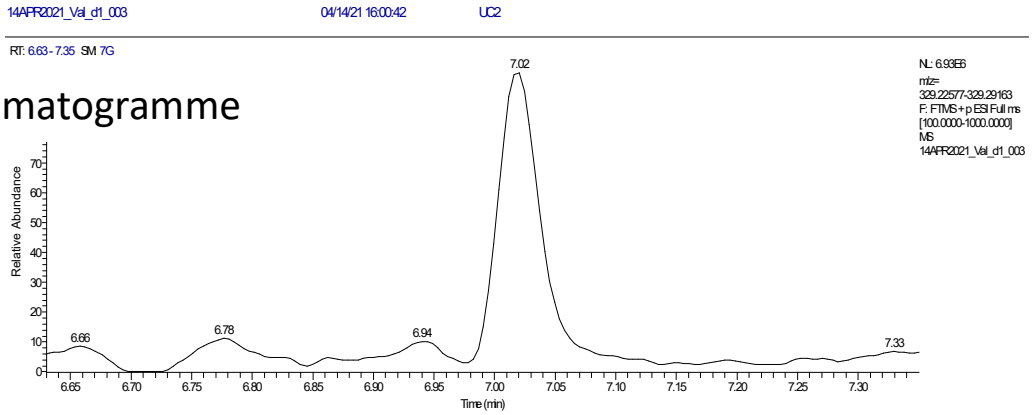
m/z du stanozolol



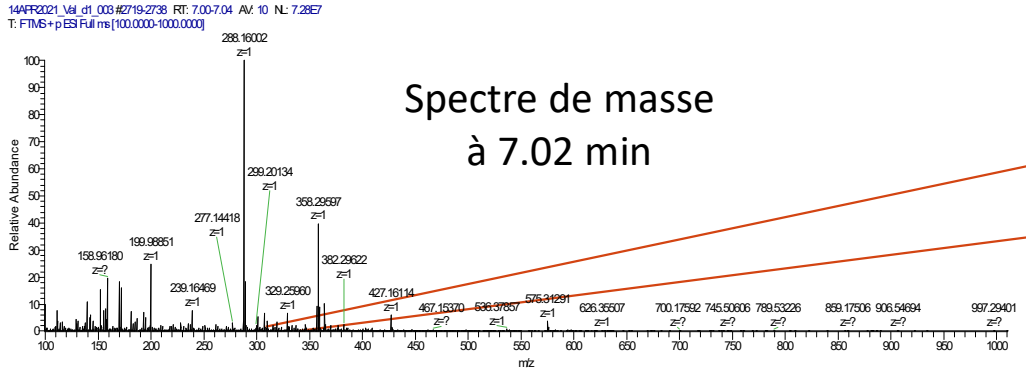
Screening par LC-HRMS

Extracted ion chromatogram (XIC)
m/z range ± 100 ppm (329.2258 – 329.2916)

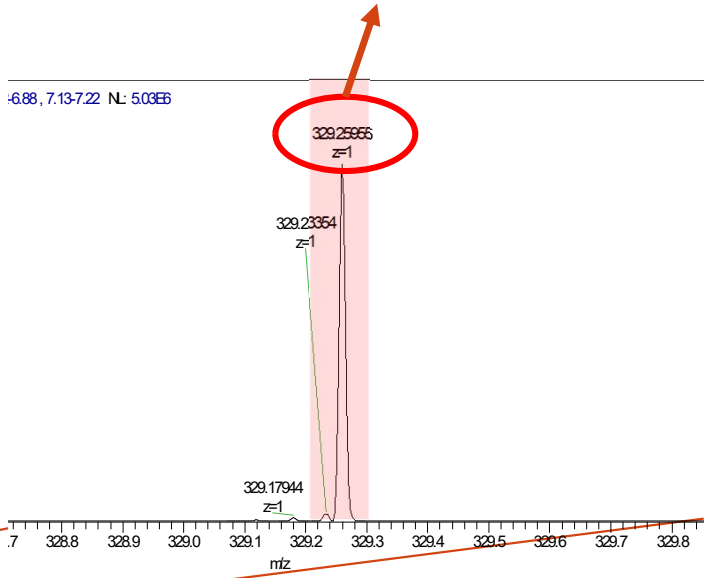
Chromatogramme



Spectre de masse
à 7.02 min



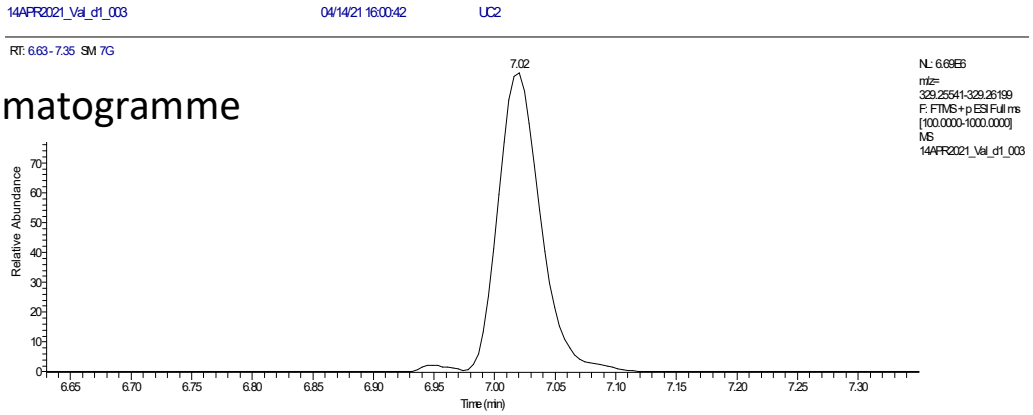
m/z du stanozolol



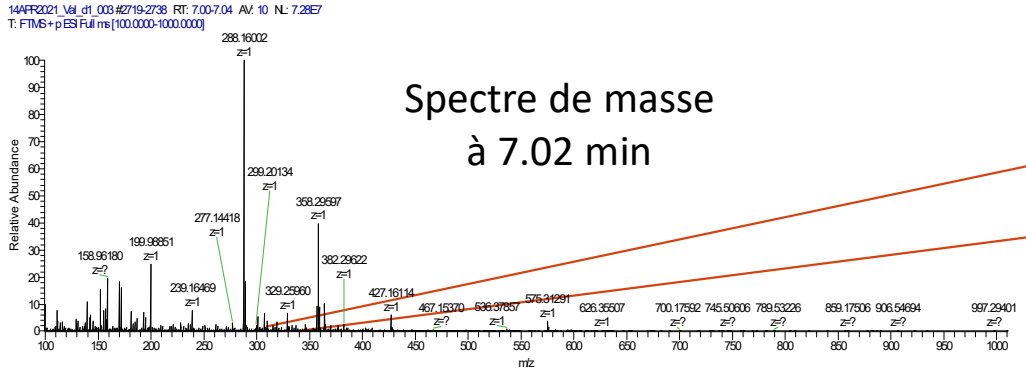
Screening par LC-HRMS

Extracted ion chromatogram (XIC)
m/z range ± 10 ppm (329.2554 – 329.2620)

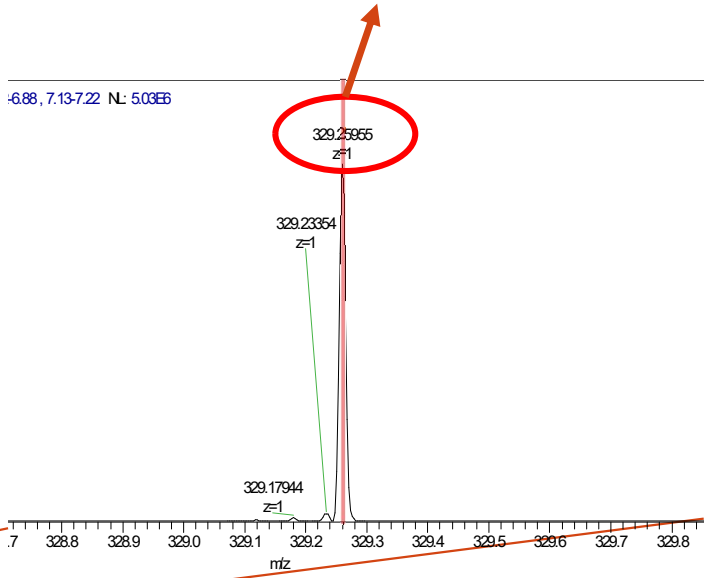
Chromatogramme



Spectre de masse
à 7.02 min

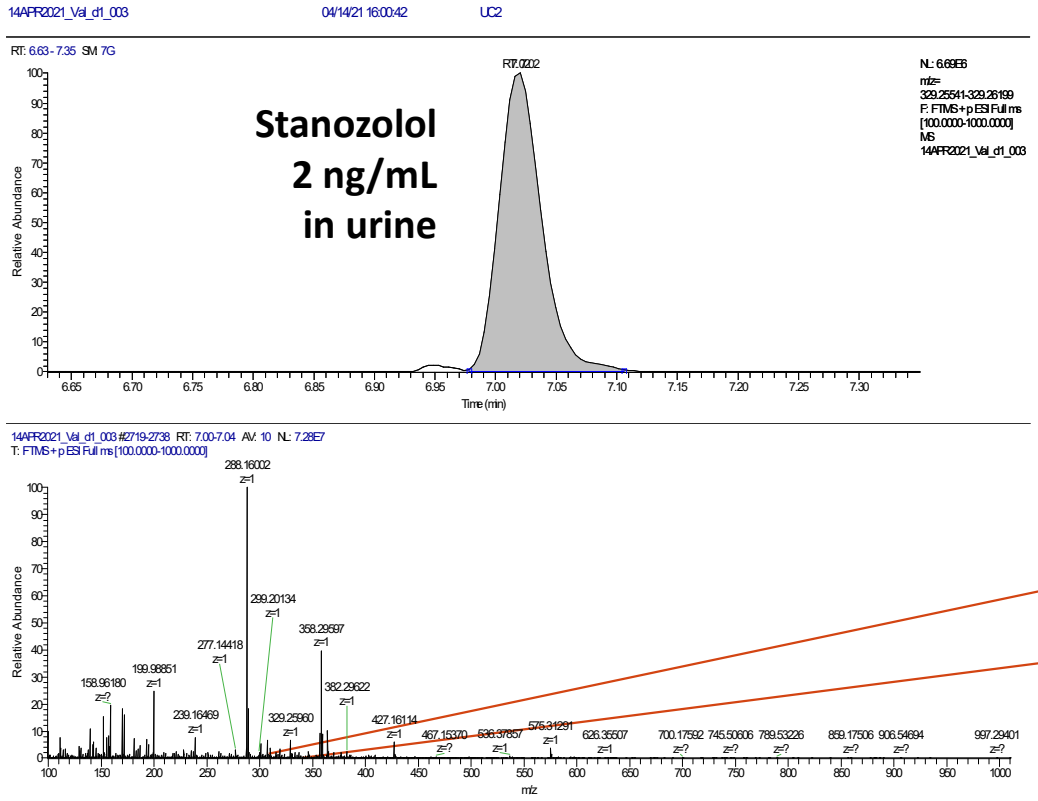


m/z du stanozolol

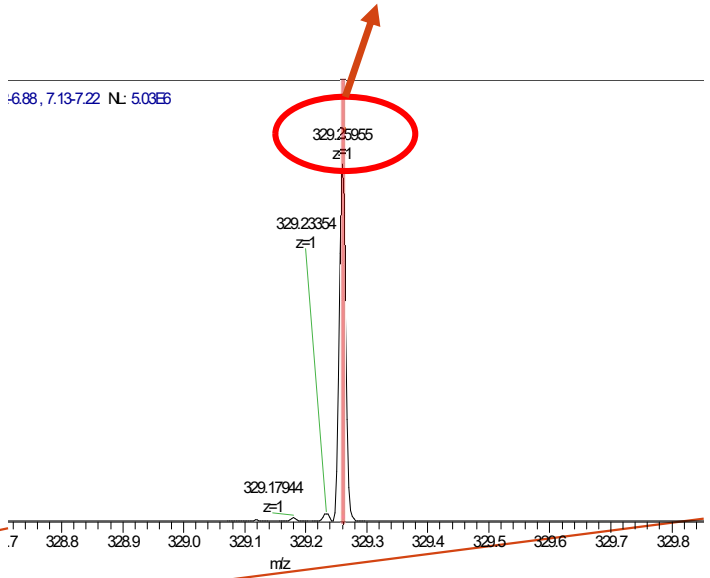


Screening par LC-HRMS

Extracted ion chromatogram (XIC)
m/z range ± 10 ppm (329.2554 – 329.2620)



m/z du stanozolol



Evolution des techniques GC et LC dans l'antidopage

GC-NPD

GC-MS

HPLC

LC-MS

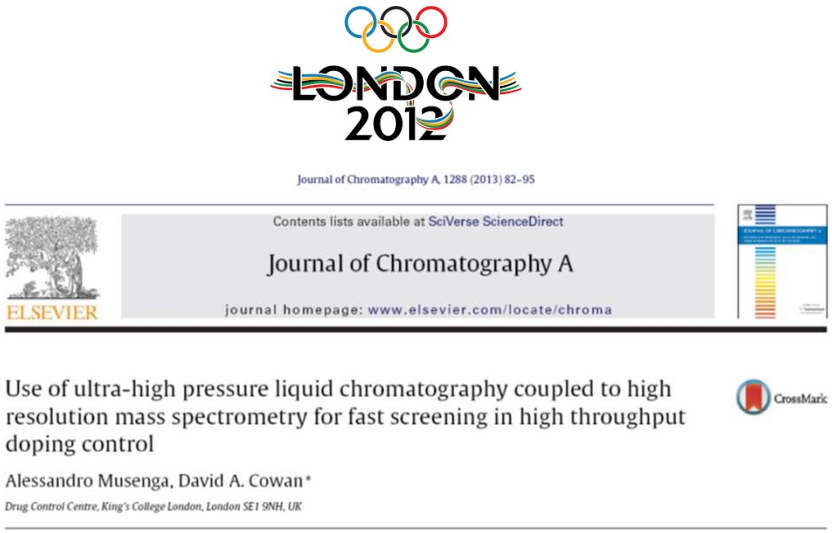
GC-C-IRMS

LC-MS/MS

LC-HRMS

1972-1976	1984	1988-1992	1994-1998	2004-2008	2012	2016
GC-NPD **GC-MS**	**HPLC**	**LC-MS** Glucocorticoïdes, diurétiques	**GC-C-IRMS** Stéroïdes endogènes	**LC-MS/MS**	**LC-HRMS** **LC-MS/MS** "Comprehensive screening"	**LC-HRMS** **LC-MS/MS** Screening (peptides)

2012-2016 – HRMS – «comprehensive screening»



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Journal of Chromatography A

journal homepage: www.elsevier.com/locate/chroma

Use of ultra-high pressure liquid chromatography coupled to high resolution mass spectrometry for fast screening in high throughput doping control

Alessandro Musenga, David A. Cowan*

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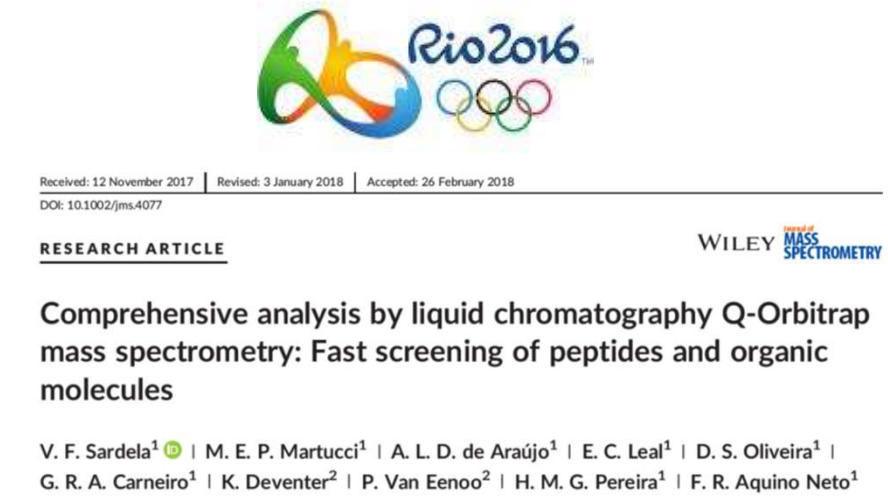
HRMS (Thermo Exactive)

- **Hydrolyse enzymatique**
- **SPE MCX, 60mg**
- **UPLC (10 min)**
- **Full scan (+/-) et AIF**

182 composés

Les petits peptides ne sont pas inclus



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DOI: 10.1002/jms.4077

RESEARCH ARTICLE

WILEY Journal of MASS SPECTROMETRY

Comprehensive analysis by liquid chromatography Q-Orbitrap mass spectrometry: Fast screening of peptides and organic molecules

V. F. Sardela[1] | M. E. P. Martucci[1] | A. L. D. de Araújo[1] | E. C. Leal[1] | D. S. Oliveira[1] | G. R. A. Carneiro[1] | K. Deventer[2] | P. Van Eenoo[2] | H. M. G. Pereira[1] | F. R. Aquino Neto[1]

HRMS (Thermo Q-Exactive)

- **Hydrolyse enzymatique**
- **SPE MCX, 60 mg**
- **Ajout d'une aliquote d'échantillon supplémentaire (10 µL) après extraction pour pour obtenir l'analyse des composés qui ne sont pas extraits**
- **UPLC (10 min)**
- **Full scan (+/-) et AIF + Full-MS/MS^2**

Screening par LC-HRMS

Aliquotage → Hydrolyse enzimatique → Extraction SPE → Evaporation → Analyse LC-HRMS

LONDON 2012 — SPE SCX — Rio2016

Small peptides

LONDON 2012 — Full scan (+/-) AIF

Rio2016 — Full scan (+/-) AIF MS/MS

2025 → SPE WCX (small peptides) — Full scan (+) MS/MS

Screening par LC-HRMS – full scan

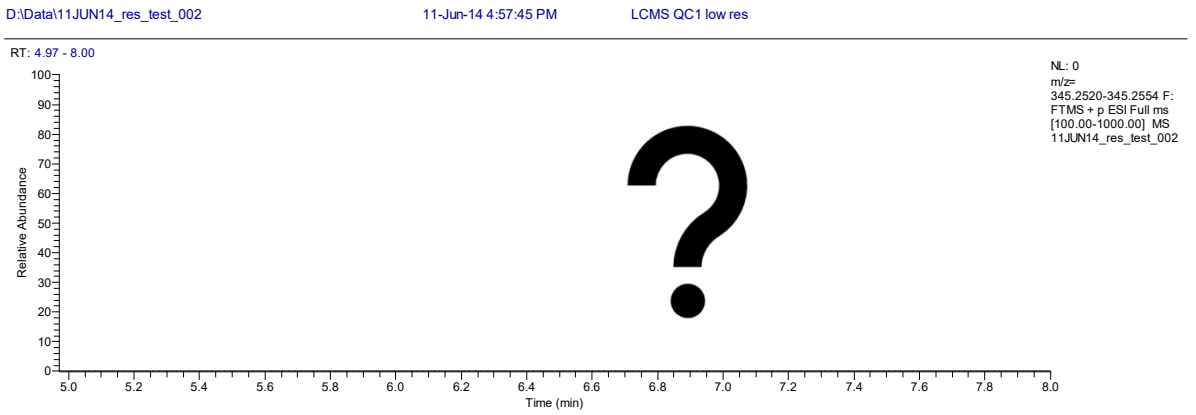
Est-ce que la résolution
(et la séparation chromatographique) est utile pour les petites molécules?

Screening par LC-HRMS – full scan

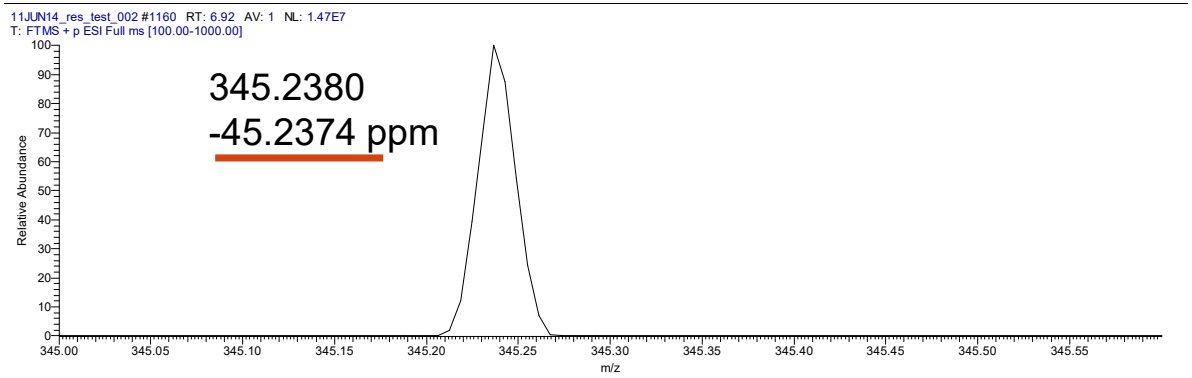
Est-ce que la résolution (et la séparation chromatographique) est utile pour les petites molécules?

Acquisition à **17,500** FWHM

Chromatogramme
XIC à m/z 345.2537 ± 5 ppm



Spectre de masse à
6.9 min



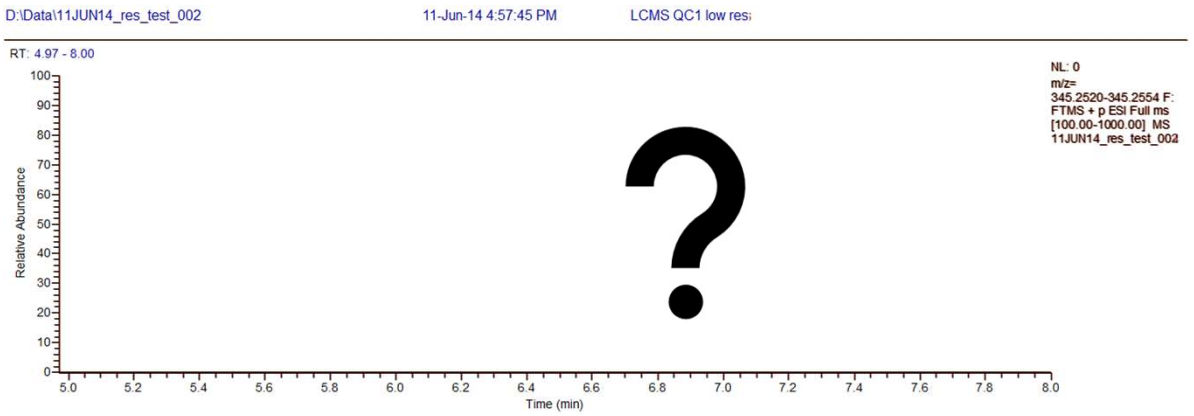
3'OH Stanozolol
2 ng/mL
dans l' urine

Screening par LC-HRMS – full scan

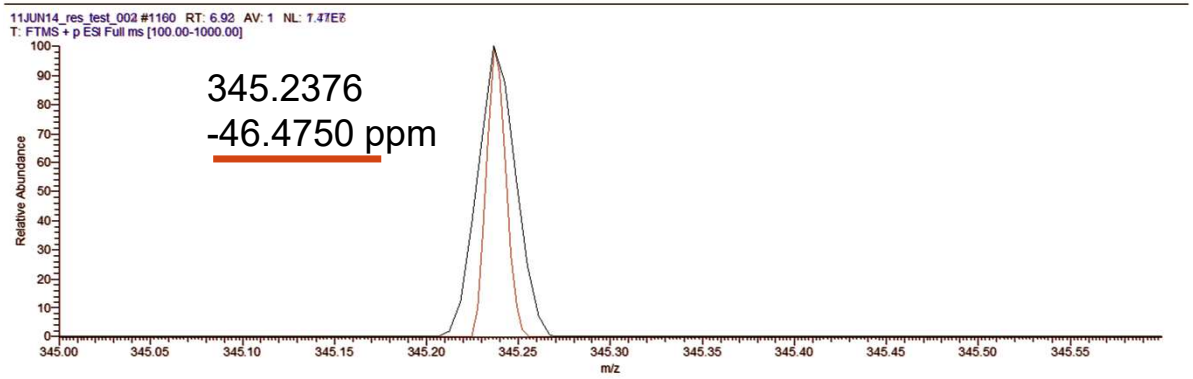
Est-ce que la résolution (et la séparation chromatographique) est utile pour les petites molécules?

Acquisition à **35,000** FWHM

Chromatogramme
XIC à m/z 345.2537 ± 5 ppm



Spectre de masse à
6.9 min



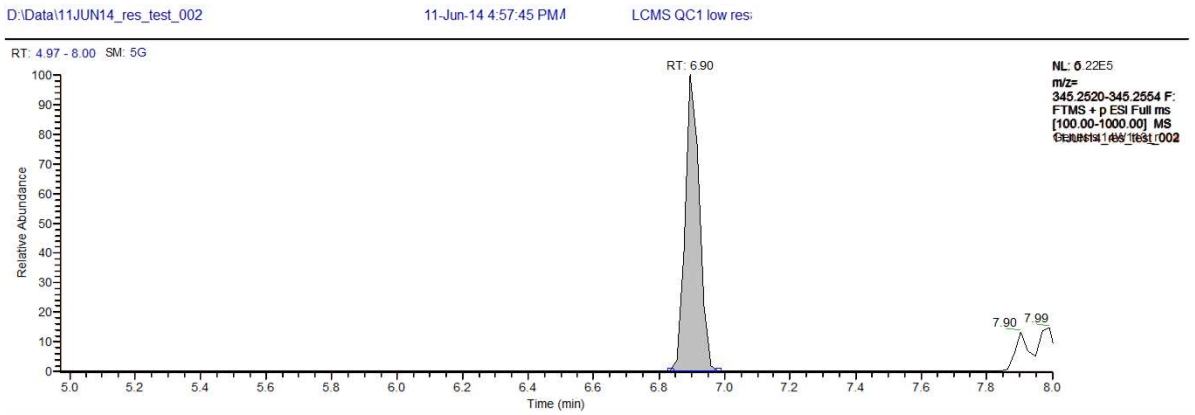
3'OH Stanozolol
2 ng/mL
dans l' urine

Screening par LC-HRMS – full scan

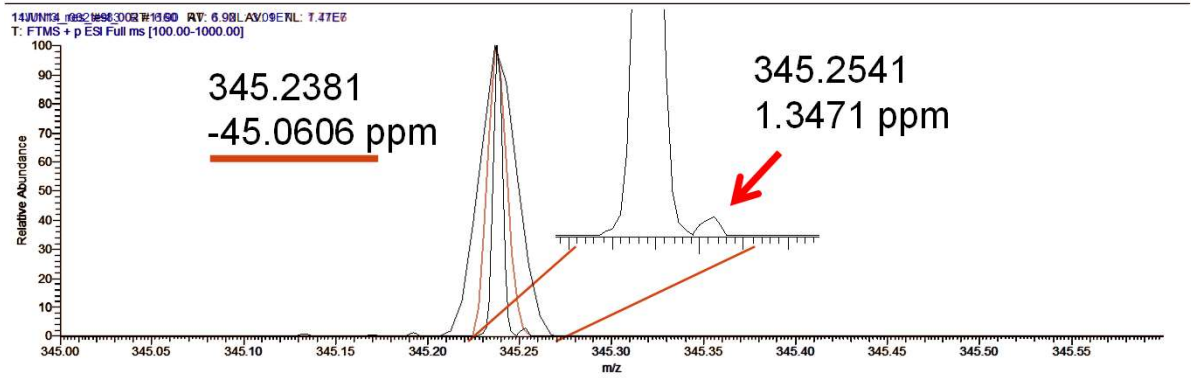
Est-ce que la résolution (et la séparation chromatographique) est utile pour les petites molécules?

Acquisition à **70,000** FWHM

Chromatogramme
XIC à m/z 345.2537 ± 5 ppm



Spectre de masse à
6.9 min



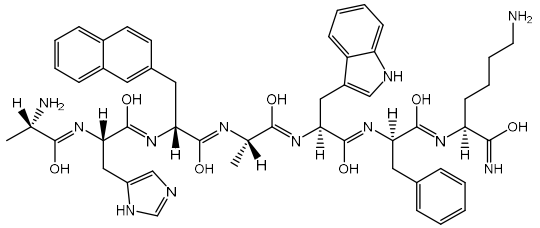
3'OH Stanozolol
2 ng/mL
dans l' urine

Screening par LC-HRMS – full scan

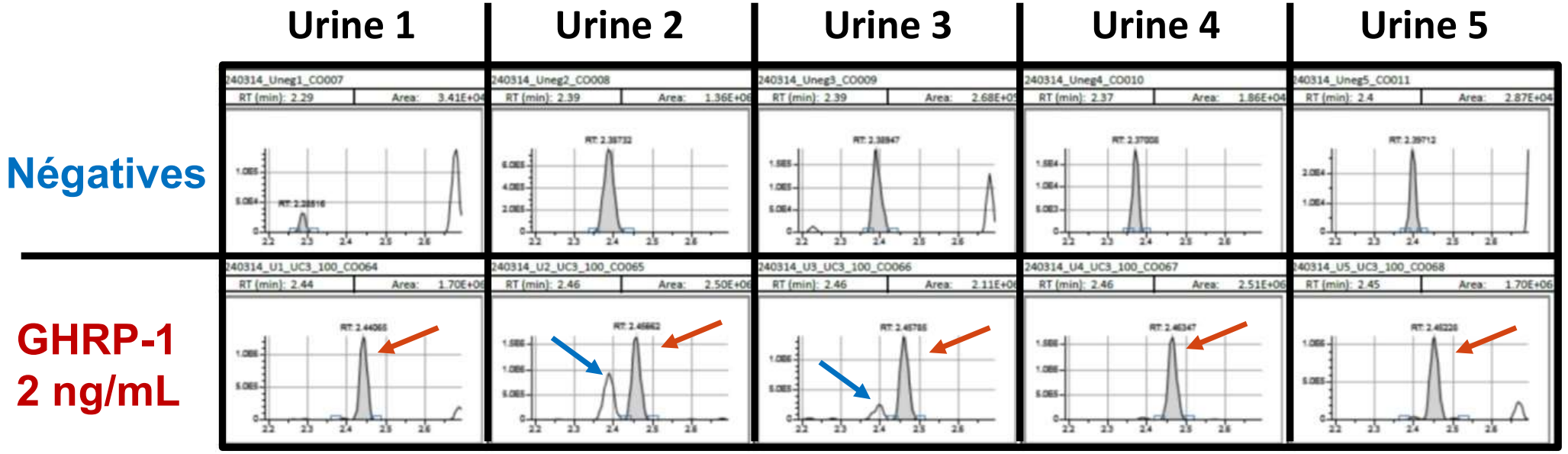
Est-ce que la haute résolution est utile pour les petits peptides et les composés multichargés?

LC-HRMS – détection de peptides

GHRP-1 : Ala-His-(D-β-Nal)-Ala-Trp-(D-Phe)-Lys-NH2
MW: 955.1 g/mol

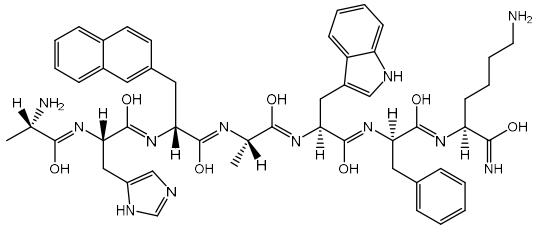


m/z = 319.1694
$[M+3H]^{3+}$

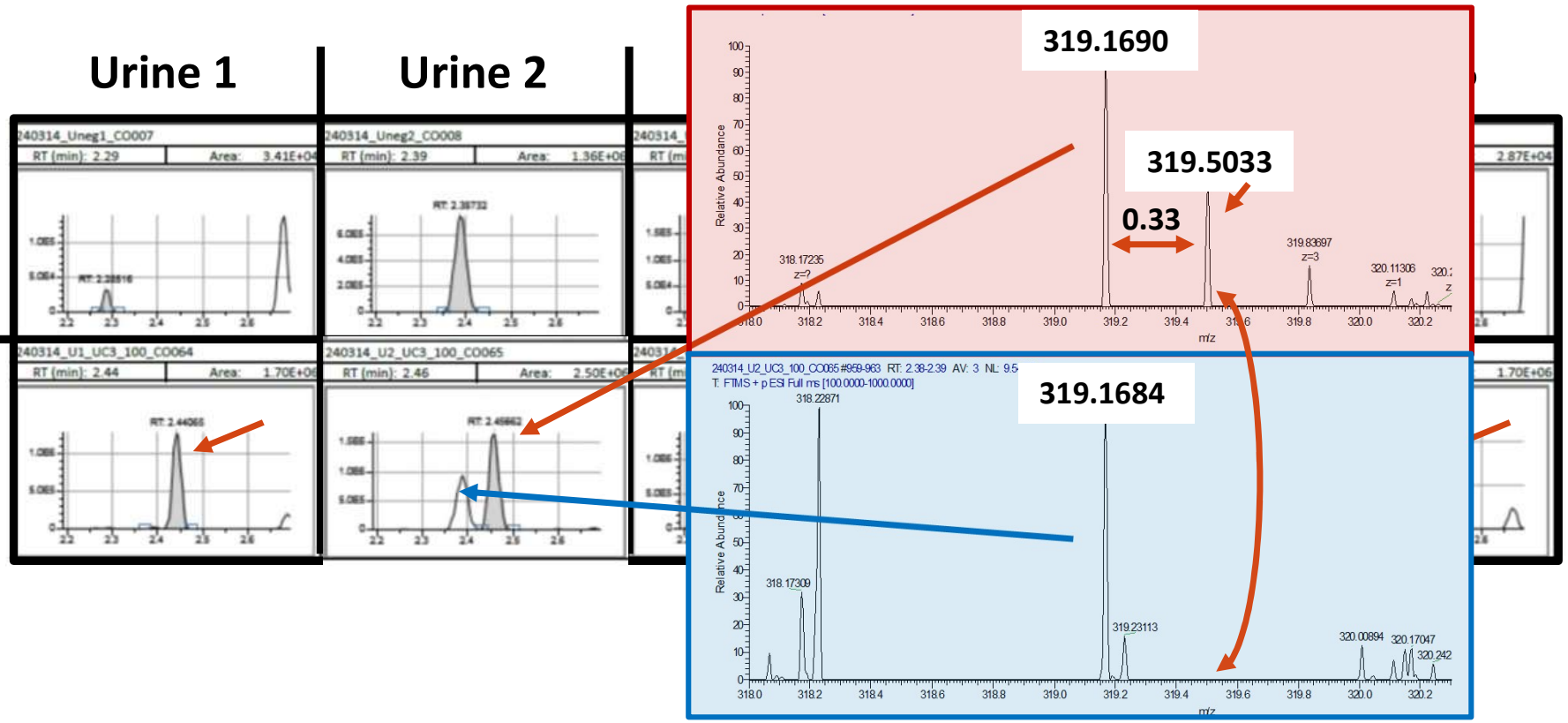


LC-HRMS – détection de peptides

GHRP-1 : Ala-His-(D-β-Nal)-Ala-Trp-(D-Phe)-Lys-NH2
MW: 955.1 g/mol



m/z = 319.1694
$[M+3H]^{3+}$

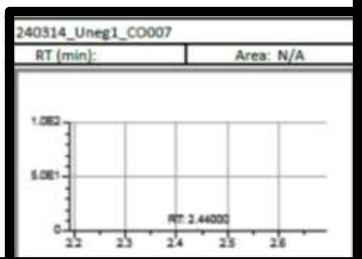
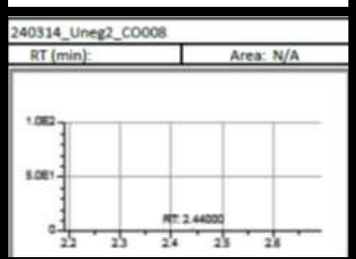
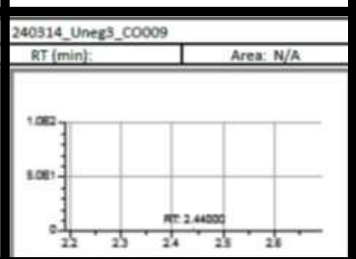
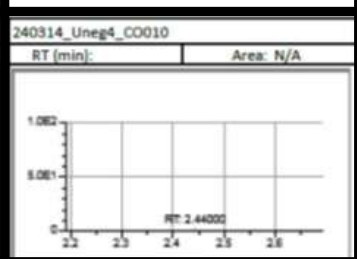
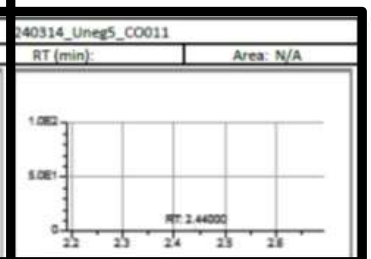
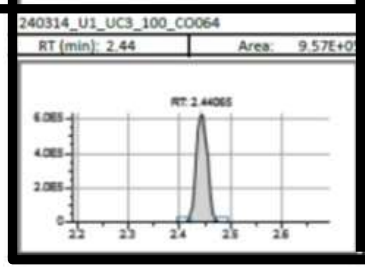
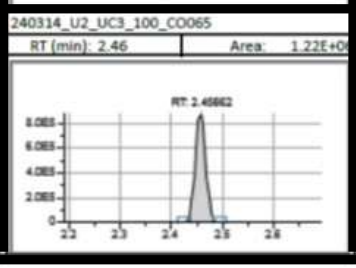
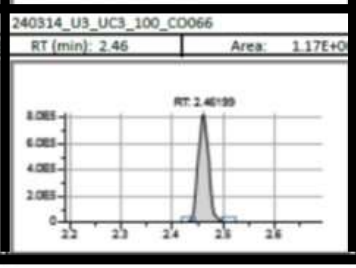
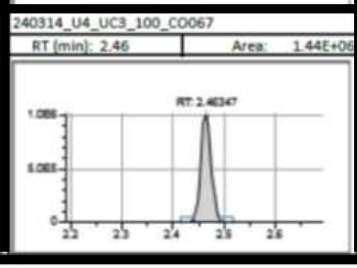
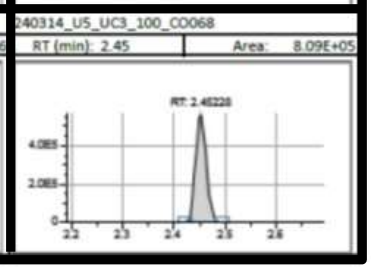


LC-HRMS – détection de peptides

GHRP-1 : (Ala-His-(D-β-Nal)-Ala-Trp-(D-Phe)-Lys-NH2)

m/z = 319.1694
$[M+3H]^{3+}$

m/z = 319.5039
$[M+3H]^{3+}$
^{13}C

	Urine 1	Urine 2	Urine 3	Urine 4	Urine 5
Négatives					
GHRP-1 2 ng/mL					

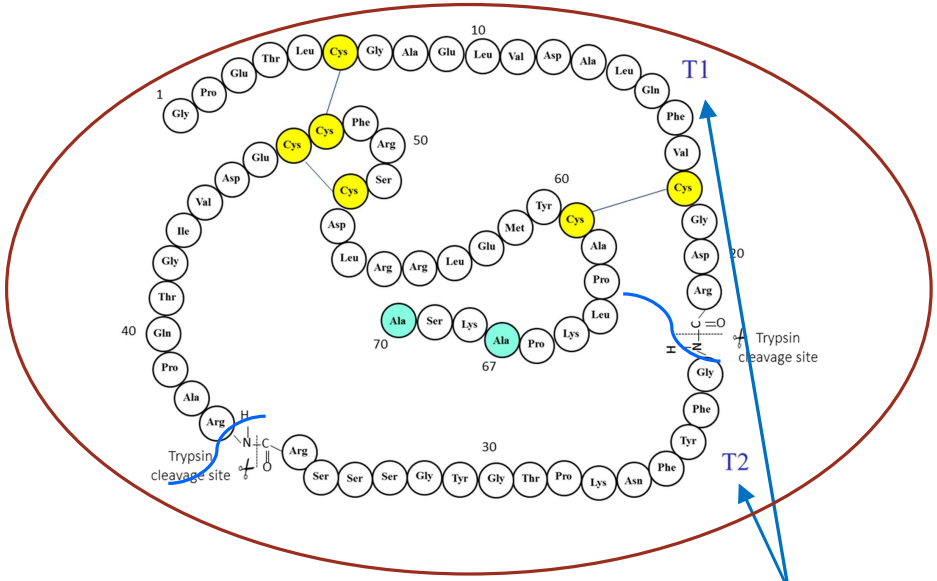
Analyse de l'IGF-1 – bottom up

IGF-1

- 70 acides aminés, mw 7.6 kDa
- Composé endogène
- Stimule la synthèse des protéines et inhibe la dégradation protéique
- Utilisée comme agente dopante pour stimuler l'anabolisme musculaire
- Monitoré dans le cadre du passeport endocrinien comme biomarqueur de la prise de l'hormone de la croissance

Deux approches analytiques

Top down
(Analyse de l'IGF-1 intact)



Bottom-up
(Digestion triptyque suivie de l'analyse de l'un ou des deux peptides obtenus (T1-T2))

Analyse de l'IGF-1 – bottom-up – étude inter laboratoire

Clinical Chemistry 60:3
541–548 (2014)

Interlaboratory Agreement of Insulin-like Growth Factor 1 Concentrations Measured by Mass Spectrometry

Holly D. Cox,[1] Filipe Lopes,[2] Getachew A. Woldemariam,[3] Jessica O. Becker,[4] Mark C. Parkin,[2] Andreas Thomas,[5] Anthony W. Butch,[3] David A. Cowan,[2] Mario Thevis,[5] Larry D. Bowers,[6] and Andrew N. Hoofnagle[4*]

Analyse de l'IGF-1 – bottom-up – étude inter laboratoire

Clinical Chemistry 60:3
541–548 (2014)

Interlaboratory Agreement of Insulin-like Growth Factor 1 Co

Holly D.
Andrea

Bottom-up

Supplemental Data Table 1. Comparison of LC-MS/MS Platforms

Laboratory	A	B	C	D	E
Mass Spectrometer and Conditions					
Instrument	ABI QTOF 5600	Waters Micromass Quattro Premier	AB Sciex QTRAP 5500	Agilent 6490 QQQ	AB Sciex QTRAP 4000
Interface	DuoSpray Ion Source	Nano ESI (New Objective)	Turbo ion-spray	Jet Stream ESI	Turbo ion-spray
Capillary	5.5 kV	3.30 kV	5.5 kV	3 kV	5.5 kV
Nozzle	80 V (declustering potential)	32 V (cone voltage)	---	500 V	---
Nebulizing gas	Gas 1 (N_2) 50 psi Gas 2 (N_2) 70 psi	---	50 psi	20 psi	50 psi
Sheath or curtain gas	N_2 25 psi	---	40 psi	11 L/min	20 psi
Temperature	500°C (heater gas)	85° (Source)	400°C	250°C	---
Drying gas	---	---	50 psi	14 L/min	50 psi
Temperature	---	---	---	200°C	250°C

Analyse de l'IGF-1 – top-down – étude inter laboratoire

Clinical Chemistry 66:4
579–585 (2020)

Inter-Laboratory Agreement of Insulin-like Growth Factor 1 Concentrations Measured Intact by Mass Spectrometry

Danielle Moncrieffe,[a,b,*] Holly D. Cox,[c] Samantha Carletta,[d] Jessica O. Becker,[e] Andreas Thomas,[f] Daniel Eichner,[c] Brian Ahrens,[d] Mario Thevis,[f] Larry D. Bowers,[g] David A. Cowan,[b] and Andrew N. Hoofnagle[e,*]

Analyse de l'IGF-1 – top-down – étude inter laboratoire

Clinical Chemistry 66:4
579-58[illegible] (2020)

Inter-Laboratory Agreement of Insulin-like Growth Factor 1 Concentrations Measured Intact by Mass Spectrometry

Danielle Moncrieffe,[a,b,*] Holly D. [illegible]
Daniel Eichner,[c] Brian Ahrei

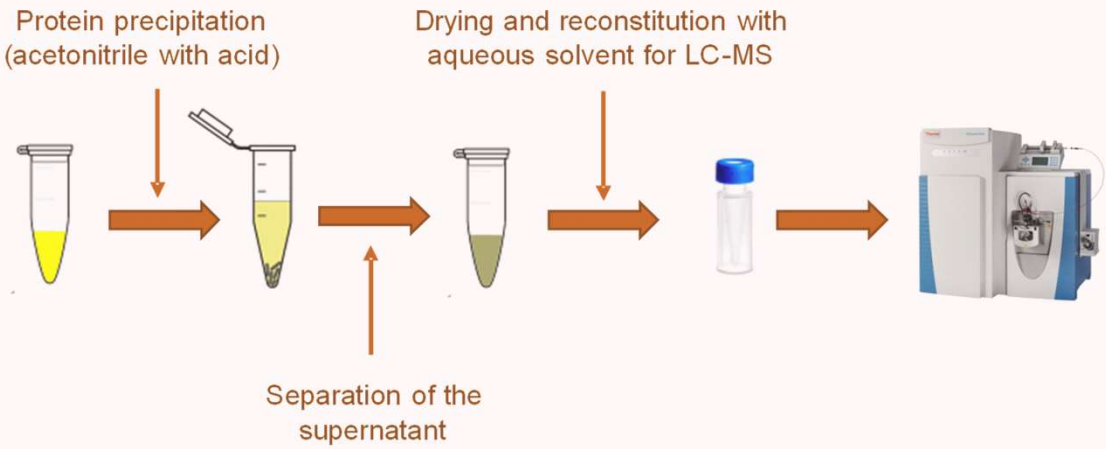
Bottom-up

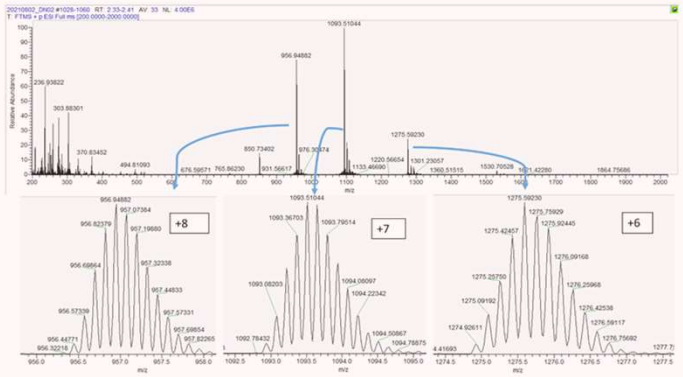
Digest LC-MS instrument conditions					
Laboratory	**A**	**B**	**C**	**D**	**E**
			Mass spectrometer and Conditions		
Instrument	Thermo Q-Exactive HFX	Sciex 5500 TripleQuad	Thermo Q-Exactive Plus	Sciex 6500+	Xevo TQS-micro QQQ
Ion Source mode	HESI (positive)	ESI (positive)	ESI (positive)	ESI (positive)	ESI (positive)
Capillary	4 kV	5.5 kV	3.5 kV	4.25 kV	1 kV
Cone	50 (S-lens RF level)	-	-	(See declustering potential, DP with MRMs below)	35 V
Cone Gas Flow	aux gas 10	Curtain gas 35	aux gas 10	35	0 L/hour
Desolvation Gas Flow	sheath gas 30	GS1: 50, GS2: 50	sheath gas 50	GS1: 45, GS2: 45	1000 L/hour
Source temperature	550 °C	450 °C	350 °C	475 °C	150 °C

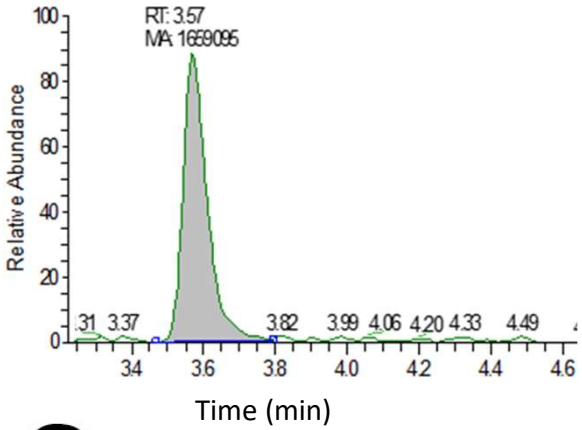
Top down

Intact LC-MS instrument conditions					
Laboratory	A	B	C	D	E
			Mass spectrometer and Conditions		
Instrument	Thermo Q-Exactive HFX	Thermo QE Plus	Thermo Q-Exactive Plus	Sciex 6500+	Xevo TQS-micro QQQ
Ion Source mode	HESI (positive)	ESI (positive)	ESI (positive)	ESI (positive)	ESI (positive)
Capillary	4 kV	4 kV	3.5 kV	5 kV	1 kV
Cone	50 (S-lens RF level)	-	-	(See declustering potential, DP with MRMs below)	30 V
Cone Gas Flow	aux gas 10	aux gas 30	aux gas 10	40	0 L/hour
Desolvation Gas Flow	sheath gas 30	sheath gas 50	sheath gas 50	GS1: 50 GS2: 50	1000 L/hour
Source temperature	550 °C	370 °C	350 °C	600 °C	500 °C

Quantification de l'IGF-1 intact au LAD: LC-HRMS (SIM)

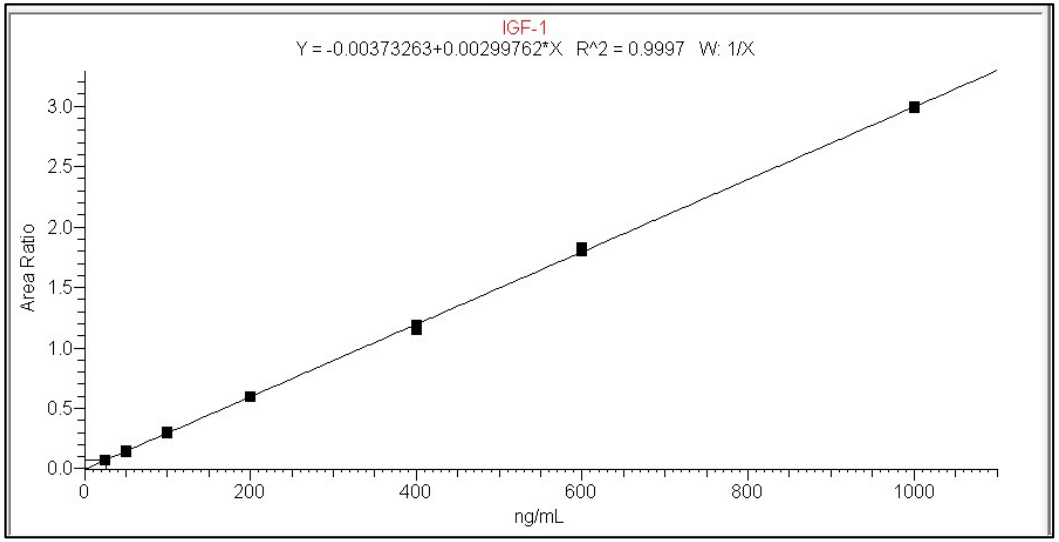






LOQ = 25 ng/mL

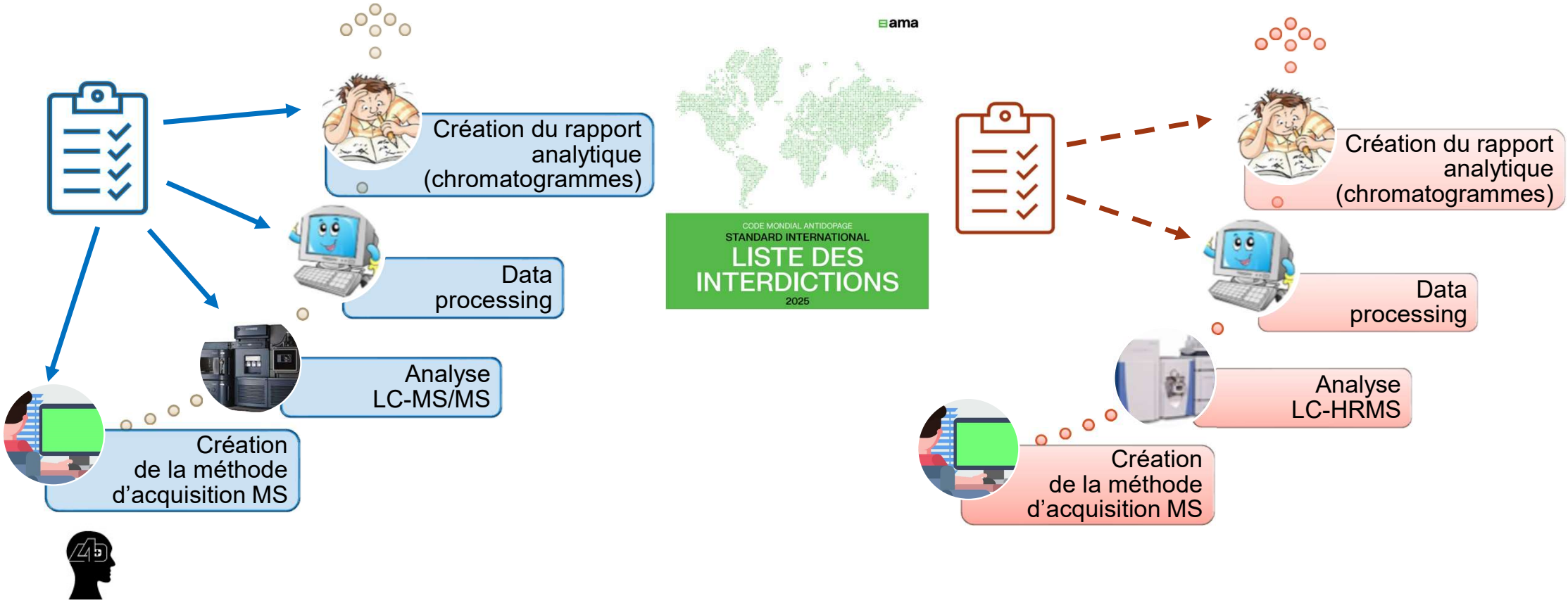
Linéarité:
25-1000 ng/mL



HRMS: targeted ou untargeted?

MS/MS (triple quadripôle)

MS à haute résolution



Investigation sur l'adultération des échantillons

Échantillon urinaire analysé avec les méthodes de screening de routine

- **Profile stéroïdien non mesurable (GC-MS)**
- **Absence du cortisol et cortisone (LC-MS)**

↓

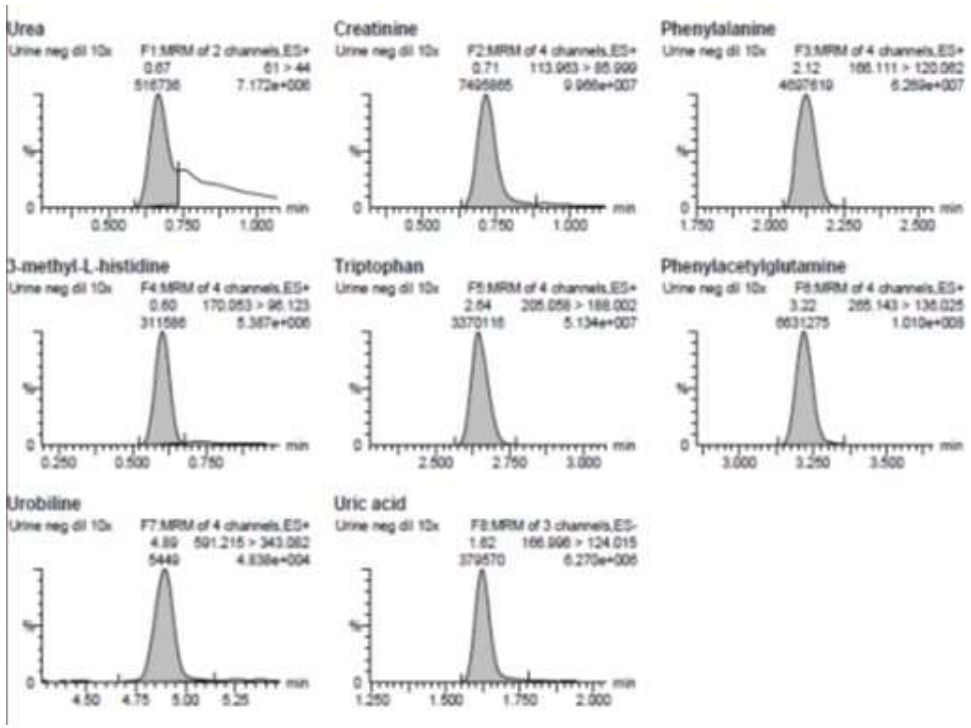
Analyse supplémentaire pour le recherche de markers urinaires

Analyse LC-MS/MS (triple quadrupôle)
pour la recherche de markers urinaires typiques

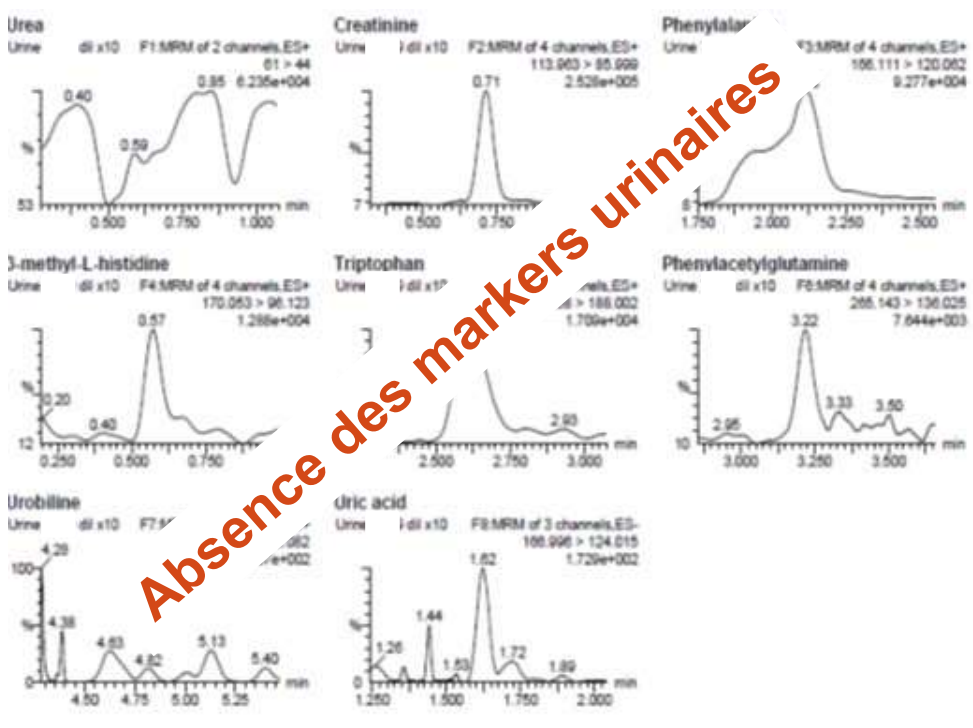
- *urée*
- *acide urique*
- *urobiline*
- *créatinine*
- *phénylalanine*
- *tryptophane*
- *phenylacetylglutamine*
- *3-methylhistidine*

Recherche de markers urinaires – LC-MS/MS

Urine négative



Échantillon suspect



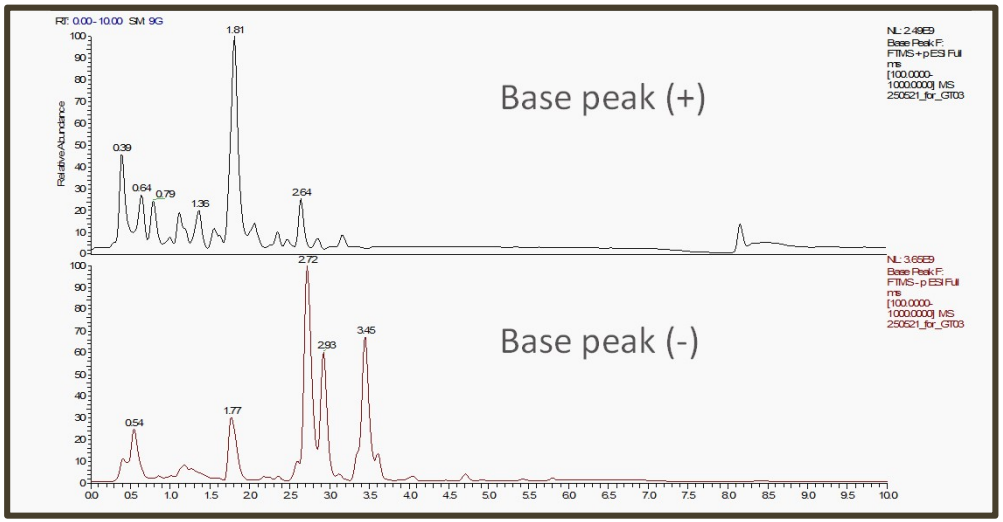
Absence des markers urinaires

100 µL urine
dilution

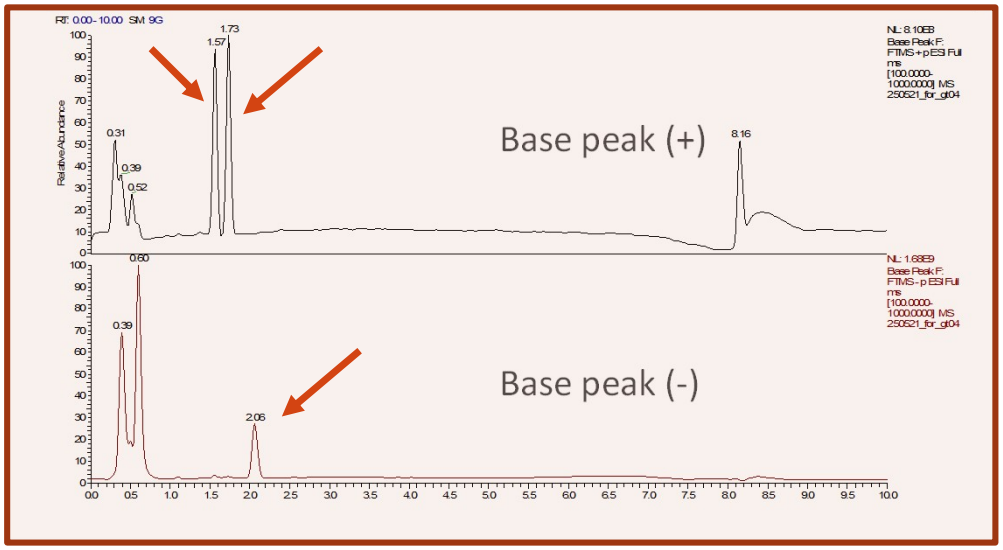
UHPLC-MS/MS
HSS t3
ESI (+/-)

Analyse HRMS full scan – «profil» de l'échantillon

Urine négative



Échantillon suspect

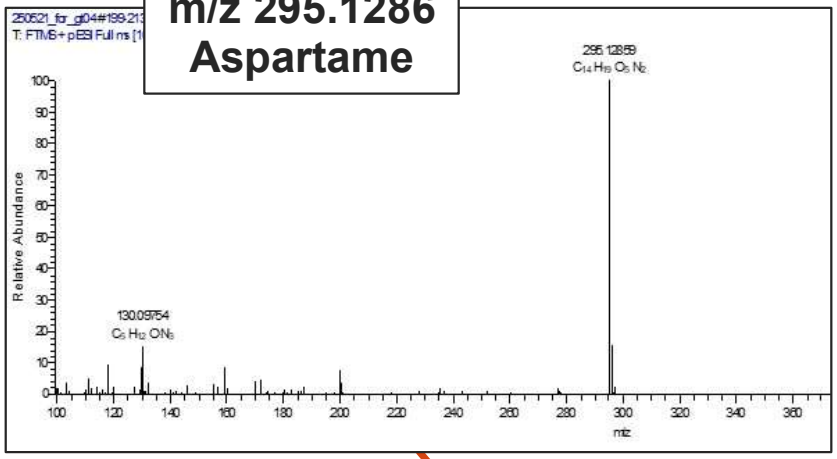


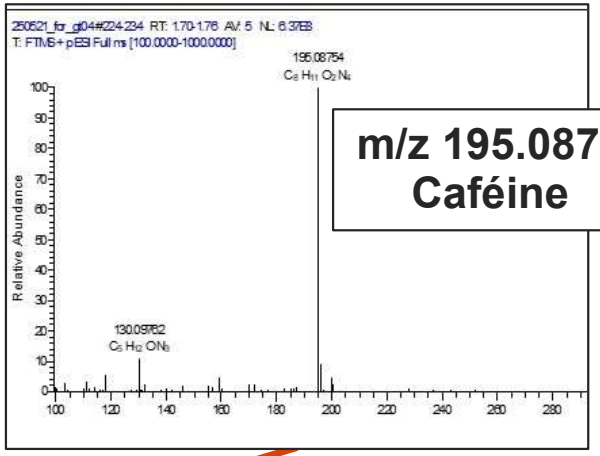
UHPLC-HRMS
Full scan, ESI (+/-)
C18 CSH

- **« profil » diffèrent**
- **absence de pics principaux des échantillons urinaires**
- **présence de trois pics qui sont normalement absents ou plus faibles dans les autres échantillons urinaires**

Analyse LC-HRMS – identification des substances

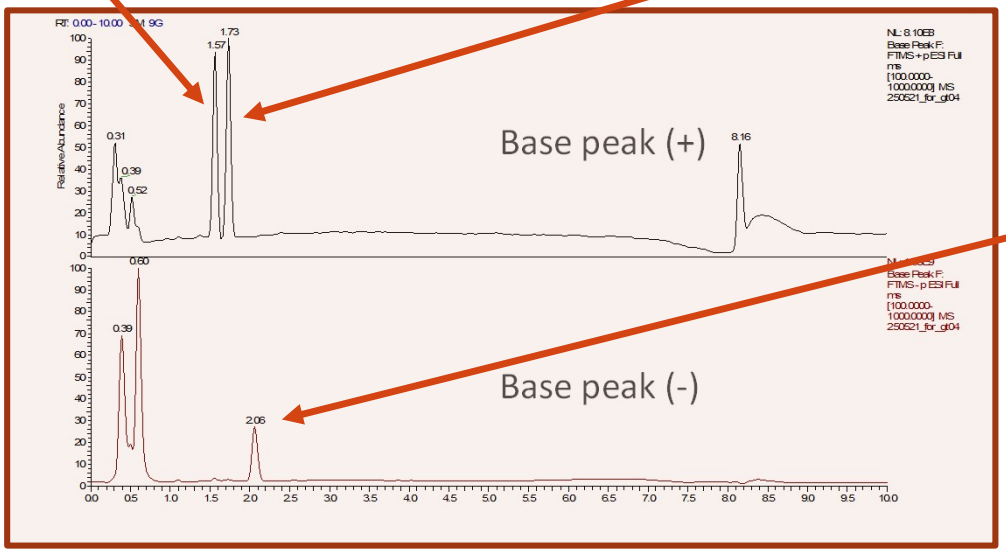
m/z 295.1286
Aspartame

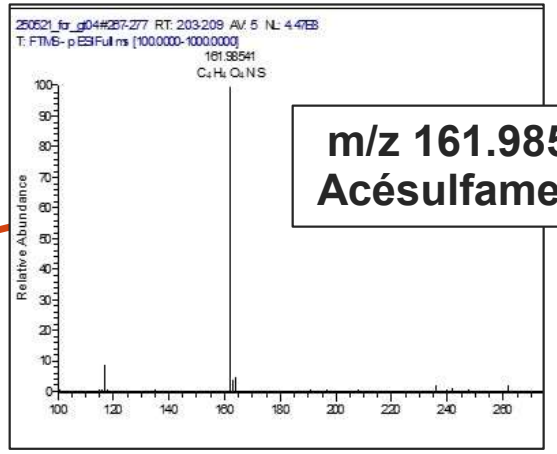




m/z 195.0875
Caféine

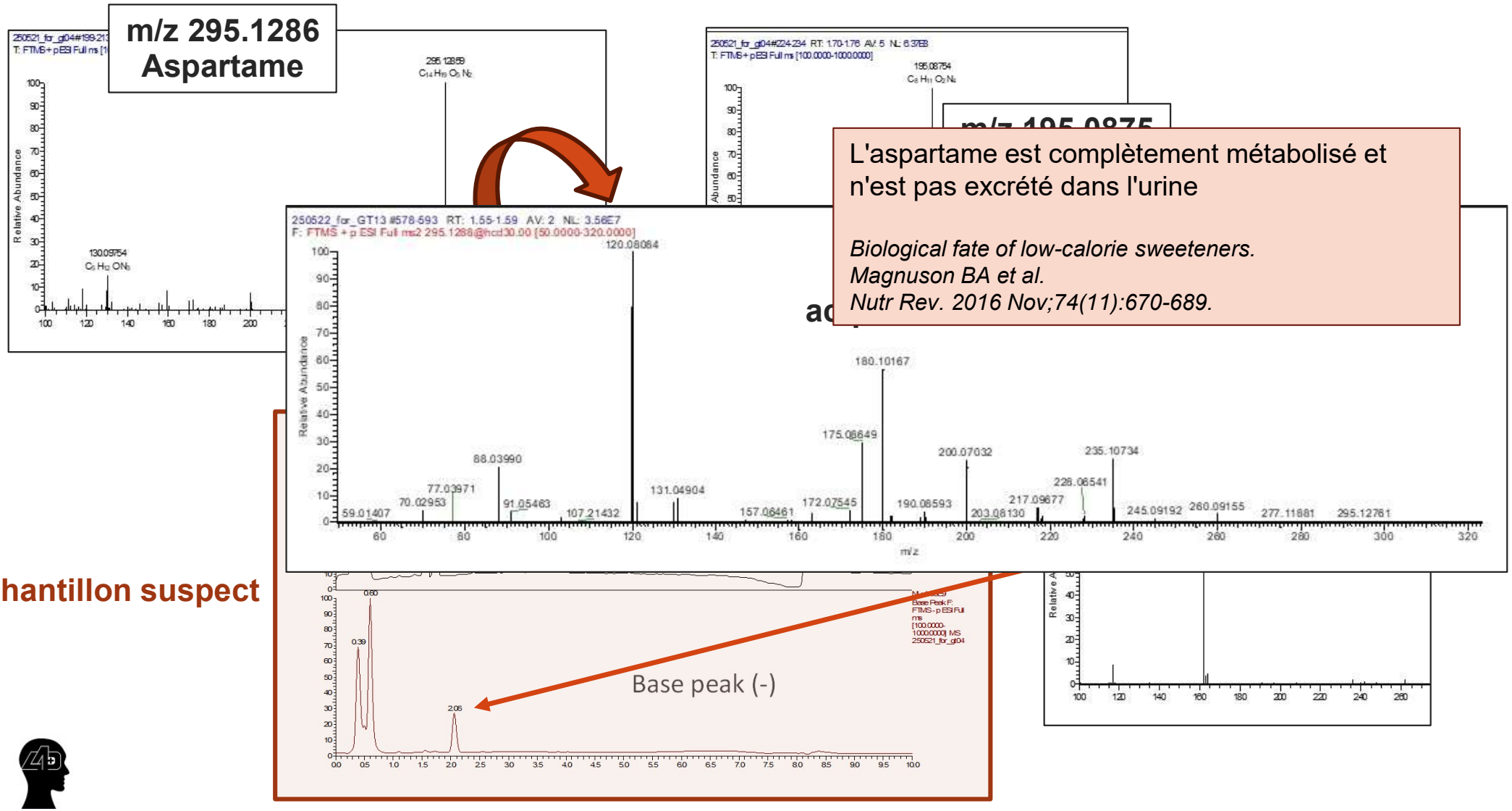
Échantillon suspect





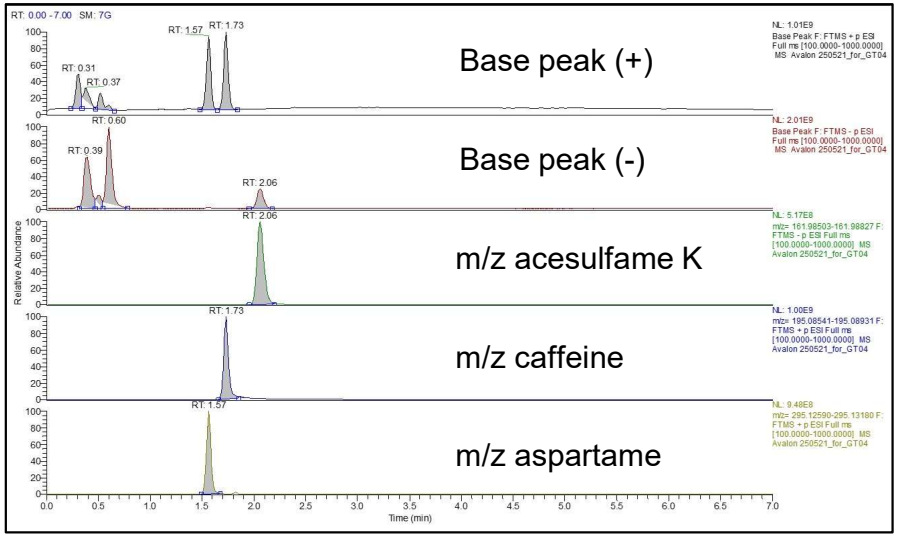
m/z 161.9854
Acésulfame K

Analyse LC-HRMS – identification des substances

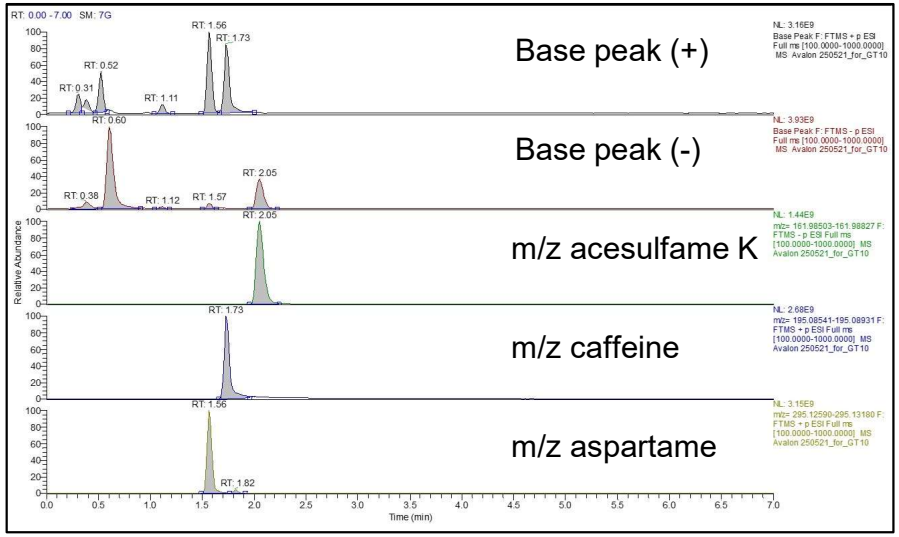


Analyse LC-HRMS – identification d'adultération d'urine

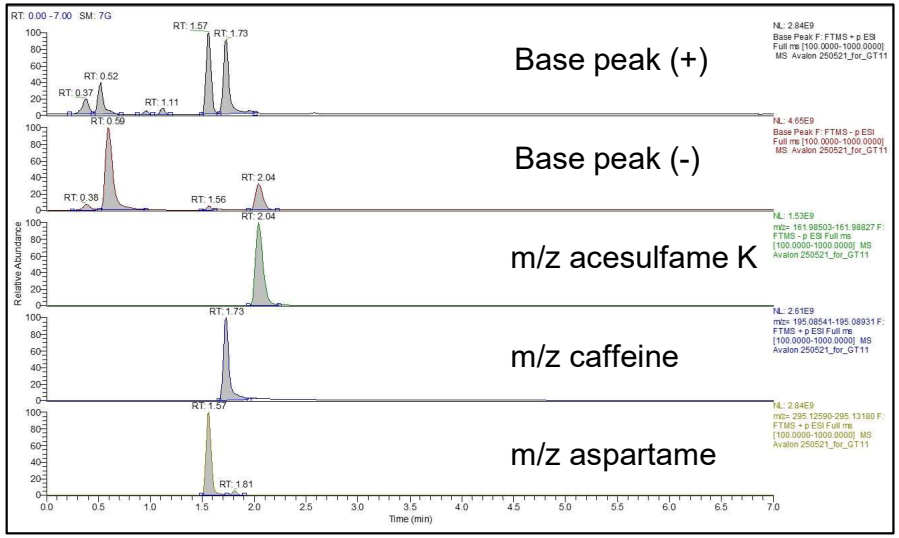
Échantillon suspect



« Energy drink » 2Go



« Energy drink » E-drink



LC-HRMS dans l’antidopage

- **Est-il permis d'utiliser la spectrométrie de masse à haute résolution dans l'analyse antidopage?**
- **La spectrométrie de masse à haute résolution est-elle couramment utilisée dans l’analyse antidopage ?**
- **Pour quelles applications la spectrométrie de masse à haute résolution est-elle utilisée dans l’analyse antidopage ?**
- **La spectrométrie de masse à haute résolution a-t-elle complètement remplacé les instruments à triple quadripôle ?**

Evolution des techniques GC et LC dans l'antidopage

GC-NPD

GC-MS

HPLC

LC-MS

GC-C-IRMS

LC-MS/MS

LC-HRMS

1972-1976	1984	1988-1992	1994-1998	2004-2008	2012	2016	2020-2024
GC-NPD GC-MS	HPLC	LC-MS Glucocorticoïdes, diurétiques	GC-C-IRMS Stéroïdes endogènes	LC-MS/MS	LC-HRMS LC-MS/MS "Comprehensive screening"	LC-HRMS LC-MS/MS Screening (peptides)	LC-HRMS LC-MS/MS (DBS, IGF-1)

Montréal 1976

Barcelona '92

Atlanta 1996

NAGANO

ATHENS 2004

Beijing 2008

LONDON 2012

Rio2016

TOKYO 2020

PARIS 2024

MS et HRMS à Tokyo 2020

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SPECIAL ISSUE - PERSPECTIVE

WILEY

Doping control analyses during the Tokyo 2020 Olympic and Paralympic Games

Masato Okano | Ayako Ikekita | Mitsuhiko Sato | Takeshi Inoue | Shinji Kageyama | Kentaro Akiyama | Arisa Aoi | Asami Miyamoto | Atsushi Momobayashi | Masanori Ota | Miho Ishige | Hikaru Sakurai | Sho Shiomura | Mitsuha Takemine | Yuma Watanabe | Tomohisa Hikota

TOKYO 2020

2.10.16 | High-resolution full scan for future investigations using data retrospective data analysis

For the Olympics, full-scan data from high-resolution mass spectrometers were obtained out of 24-h TAT reporting to find traces of doping substances that are not currently being analyzed in the retrospective digital data processing.[1] All ITP-1a samples were analyzed using LC–HRMS (Table 2a) but with a runtime twice as long as that of ITP-1a (i.e., 16 min). The full-scan experiments were conducted with a resolu-

TABLE 1 Instrumental list equipped at the Tokyo 2020 Games laboratory

	Analytical instrument	Number	Back up power supply	Actual operation
LC–MS	Acquity UPLC I-Class PLUS and M-Class Xevo-TQ-XS (Waters)	17 micro flow (11)	9	ITP-1a (4) ITP-1b (5) ITP-2 (6) HBOCs (2) IGF-I (GH biomarkers (2) DBS (1) CP_Quantification (2) CP (6)
	UltiMate 3000 RSLC and RSLCnano UHPLC/Q Exactive Plus (Thermo Fisher Scientific)	9 capillary flow (5)	3	CP (1) Large peptides (4) Full scan (4)
GC–MS	GC-2030/GCMS-TQ8050 NX (Shimadzu)	12	4	ITP-4 (12) SP_CP (2) CP_Quantification (3) CP (7)
	Trace 1310 GC/Q Exactive (Thermo Fisher Scientific)	4	2	CP (2) Full scan (4)

Screening petits composés

Screening petits peptides (GHRP)

IGF-1

DBS

Confirmations

Insulines et «large peptides»

Screening des agents anabolisants

Confirmations

MS et HRMS à Paris 2024

ThermoFisher SCIENTIFIC

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Inside Laboratoire Anti-Dopage Français (LADF): An Interview with Director Dr. Magnus Ericsson

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At the forefront of global anti-doping efforts, the Laboratoire Anti-Dopage Français (LADF) plays a critical role in ensuring fair play and integrity in elite sports. We spoke with Dr. Magnus Ericsson, Director of LADF and a leading expert in analytical chemistry, to explore how the lab detects banned substances, manages high-throughput workflows, and leverages advanced Thermo Fisher Scientific technologies to stay ahead of constantly evolving challenges in anti-doping science.

Screening

GC-MS/MS

UHPLC-HRMS

https://www.thermofisher.com/blog/analyteguru/inside-laboratoire-anti-dopage-francais-ladf-an-interview-with-director-dr-magnus-ericsson/

Méthodes de screening chromatographique au LAD

Urine

Sang

Sérum — **DBS**

Stéroïdes endogènes et exogènes	Composés conjugués, petits peptides	Stimulants, diurétiques, etc.	IGF-1	Stéroïdes endogènes et exogènes, esters	Stéroïdes exogènes, composés divers, esters
Préparation des échantillons					
Hydrolyse enzymatique LLE TBME	**Hydrolyse enzymatique SPE WCX**	**Dilution**	**Precipitation des proteines**	**SLE**	**Extraction avec MeOH**
Dérivatisation				(Dérivatisation pour les esters)	(Dérivatisation pour les esters)
Analyse					
GC-MS GC-MS/MS	UHPLC-HRMS (Full scan + PRM) C18 CSH ESI (+)	UHPLC-MS/MS BEH C18 ESI (+/-)	UHPLC-HRMS (SIM) C18 CSH ESI (+)	UHPLC-MS/MS BEH C18 ESI (+)	UHPLC-MS/MS BEH C18 ESI (+/-)	UHPLC-HRMS (PRM) BEH C18 ESI (+)

LC-HRMS dans l'antidopage

- ***Est-il permis d'utiliser la spectrométrie de masse à haute résolution dans l'analyse antidopage?***

  Oui, il n'y a pas des règles spécifiques et chaque laboratoire peut choisir entre MS et HRMS

- ***La spectrométrie de masse à haute résolution est-elle couramment utilisée dans l'analyse antidopage ?***

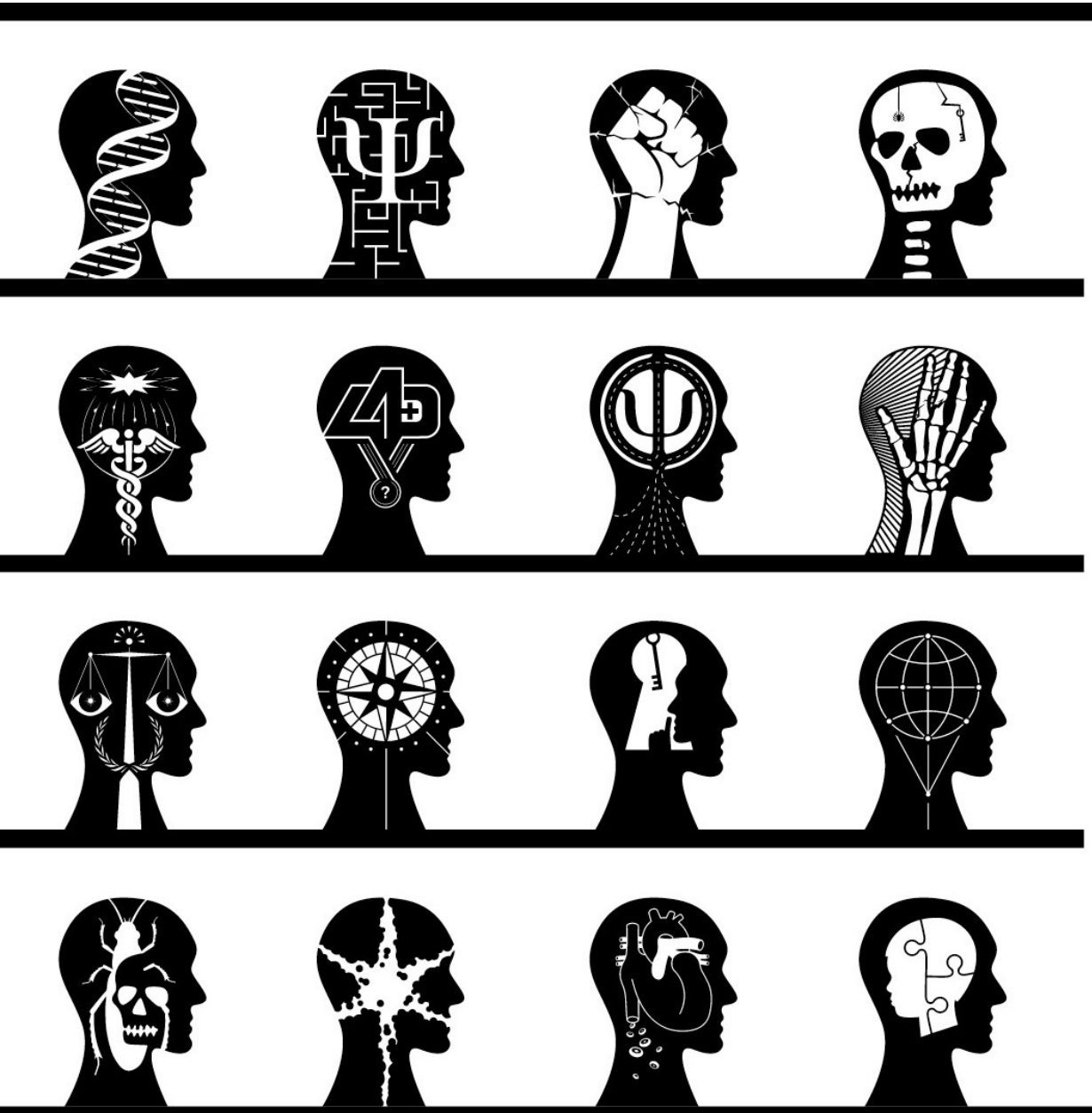
  Oui, depuis 2010-2012, les laboratoires ont commencé à inclure la HRMS dans leur portefeuille et elle est aujourd'hui couramment utilisée dans l'anti-dopage

- ***Pour quelles applications la spectrométrie de masse à haute résolution est-elle utilisée dans l'analyse antidopage ?***

  Surtout pour le screening et pour les grosses molécules, mais chaque laboratoire utilise la HRMS ou la MS en fonction de ses préférences et de ses choix analytiques

- ***La spectrométrie de masse à haute résolution a-t-elle complètement remplacé les instruments à triple quadripôle ?***

  Non, les deux techniques sont toujours utilisées



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Merci!