Ecole de printemps de la Société Française de Spectrométrie de Masse

XXII^{èmes} Rencontres du Club Jeune de la SFSM

Club Jeune SFSM

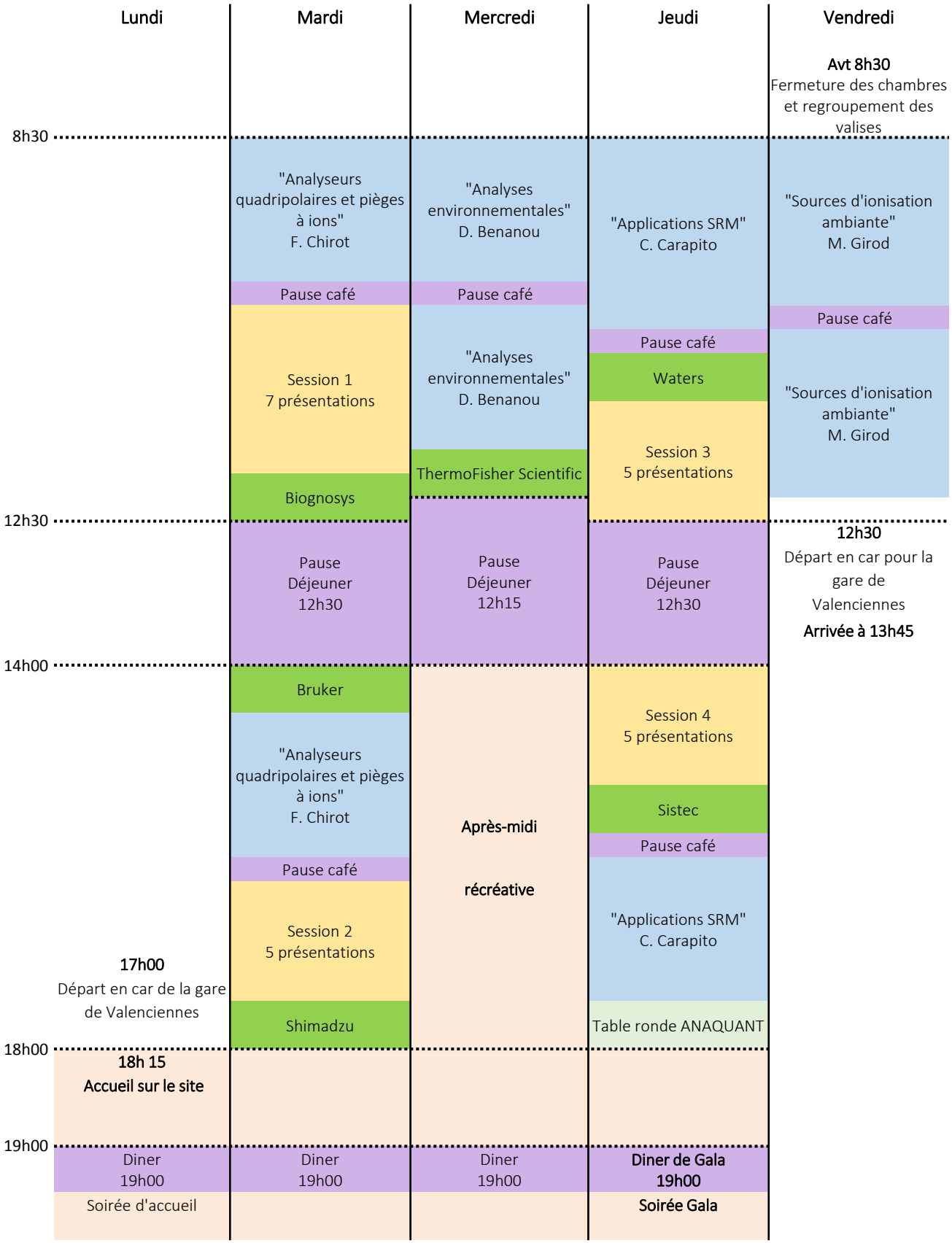
20 au 24 mars 2017

Le Bol Vert

Trélon (59), Région Hauts-de-France

Parc Naturel Régional de l'Avesnois

Programme détaillé



Mardi 21 mars

Modérateur: Jasmine Hertzog

08h30-10h00 : "Guider, filtrer, piéger et plus… avec des RF"

Fabien Chirot
Institut des Sciences Analytiques (ISA), UMR 5280 CNRS-Université Lyon 1

10h15-12h00 : Session 1

10h15 **O1 -** "Effet de la sulfatation du domaine extracellulaire du récepteur CXCR4 sur son interaction avec la chimiokine SDF-1α analysé par électrophorèse capillaire d'affinité couplée à la spectrométrie de masse"
Maud Fumex, Denis Servent, Gilles Mourier, Régis Daniel, Florence Gonnet

10h30 **O2 -** "Nouvelles sources d'ionisation pour le couplage de la chromatographie gazeuse et de la spectrométrie de masse à haute résolution : intérêt pour les molécules liées à la défense"
Valentin Baillet

10h45 **O3 -** "Analysis of bile acids by LC-MS/MS"
Noélie Bossut

11h00 **O4 -** "Tandem Mass Spectrometry to Decipher Messages Encoded in Polyurethanes Used as Molecular Barcodes in Anti-Counterfeiting Technology"
Jean-Arthur Amalian, Benoit Eric Petit, Denise Karamessini, Abdelaziz Al Ouahabi, Christophe Chendo, Didier Gigmes, Jean-François Lutz, Laurence Charles

11h15 **O5 -** "Lipid profiling of cardiac tissue following a myocardial infarction on a mouse model: MALDI MS Imaging and LC-MSMS Identification"
Anela Dorbane, Diane Kyriazis, Ganesh Halade, Jean-Marie Schmitter, Boutayna Rhourri-Frih

11h30 **O6 -** "Correlation between the Shape of the Ion Mobility Signals and the Stepwise Folding Process of Polylactide Ions "
Quentin Duez, Thomas Josse, Vincent Lemaur, Fabien Chirot, ChangMin Choi, Philippe Dubois, Philippe Dugourd, Jérôme Cornil, Pascal Gerbaux and Julien De Winter

11h45 **O7 -** "Développement du couplage chromatographie d'exclusion stérique - spectrométrie de masse native (SEC-native MS) "
Anthony Ehkrich, Sarah Cianferani

12h00-12h30 : Intervention de Florian Marty, Biognosys

"Best of both worlds: Combining quantitative targeted proteomics with high-content discovery proteomics"

Mardi 21 mars

Modérateur: Maud Fumex

14h00-14h30 : Intervention de Sabine Jourdain, Bruker

"Solutions en spectrométrie de masse pour le criblage et la quantification en routine "

14h30-16h00 : "Guider, filtrer, piéger et plus… avec des RF"

Fabien Chirot

Institut des Sciences Analytiques (ISA), UMR 5280 CNRS-Université Lyon 1

16h15-17h30 : Session 2

16h15 **O8 -** "Monitoring of catalytic deoxygenation of pyrolysis bio-oil by ESI and APPI FT-ICR mass spectrometry"
Jasmine Hertzog, Vincent Carré, Anthony Dufour, Ondřej Mašek, Frédéric Aubriet

16h30 **O9 -** "Action-FRET and works"
Geoffrey Knight

16h45 **O10 -** "Analysis by MALDI-TOF imaging and LC-MS/MS of the effects of probiotics on bowel inflammation "
Stefania Pozzan, Maria Urdaci, Mohamed Mokrani, Marc Bonneu, Jean-Marie Schmitter, Boutayna Rhourri-Frih

17h00 **O11 -** "Caractérisation de polymères énergétiques fluorés par couplage ASAP-Orbitrap™ : exploitation des défauts de masse de Kendrick "
Gabriel Gaiffe, Richard B. Cole, Sabrina Lacpatia, Maxime Bridoux

17h15 **O12 -** "Temporally & Spatially-Resolved Microproteomics Analysis for Traumatic Brain Injury Biomarkers Identification"
Khalil Mallah, Firas Kobeissy, Michel Salzet, Kazem Zibara, Isabelle Fournier

17h30-18h00 : Intervention de Etienne Maout, Shimadzu

"SFE SFC MSMS principes et applications"

Mercredi 22 mars

Modérateur: Laurent Laboureur

08h30-10h00 : “25 ans de spectrométrie de masse pour un meilleur environnement”

David Benanou

Veolia Environnement, Paris

10h15-11h45 : “25 ans de spectrométrie de masse pour un meilleur environnement”

David Benanou

Veolia Environnement, Paris

11h45-12h15 : Intervention de Bénédicte Duretz, ThermoFisher Scientific

“Potentialités du couplage IC/MS pour l’analyse de pesticides polaires ”

Mercredi 22 mars

Après-Midi

Récréative

Jeudi 23 mars

Modérateur: Jasmine Hertzog

08h30-10h30 : "Développement de méthodes de quantification ciblée de peptides (SRM, PRM et DIA) et applications"

Christine Carapito
Laboratoire de Spectrométrie de Masse BioOrganique (LSMBO)
Université de Strasbourg, IPHC

10h45-11h15 : Intervention Christophe Siroit, Waters

"Innovations technologiques au service de la sensibilité, avec le nouveau triple quadripôle, le Xevo TQXS "

11h15-12h30 : Session 3

11h15 **O13 -** "Inflammatory bowel diseases and the contribution of OcSILAC strategies to the quantification of the oxidative and inflammatory effects of lipopolysaccharide on human colon cells' proteome"
Nicolas Eskenazi, Shakir Shakir, Emilie Mathieu, Clotilde Policar and Joelle Vinh

11h30 **O14 -** "Study of changes in the kidney's lipid profile in the case of obesity using MALDI-ToF imaging and ESI-LC-MS/MS analysis"
Gianluca Sighinolfi, Daniela Cota, Marc Bonneu, Jean-Marie Schmitter, Boutayna Rhourri-Frih

11h45 **O15 -** "Caractérisation du site cible anti-métastases de deux protéines par des techniques originales de spectrométrie de masse"
Teddy Bijot

12h00 **O16 -** "Caractérisation d'assemblages anioniques de polyoxométallates par spectrométrie de masse couplée à la mobilité ionique"
Hupin S., Lavanant H., Piot M., Izzet G., Afonso C.

12h15 **O17 -** "Contribution of peptidomics to the study of Streptococcus thermophilus peptide utilization in complex media"
Lucas Proust, Sophie Liuu, Eloi Hautebourg, Véronique Monnet, Vincent Juillard

Jeudi 23 mars

Modérateur: Laurent Laboureur

14h00-15h15 : Session 4

14h00 **O18 -** "Caractérisation d'anticorps immunoconjugués site spécifiques par spectrométrie de masse couplée à la mobilité ionique"
Thomas Botzanowski, Oscar Hernandez Alba, Stéphane Erb, Anthony Ehkirch, David Rabuka, Alain Beck, Penelope Drake, Sarah Cianferani

14h15 **O19 -** "Étude de la phase gazeuse et particulaire de la fumée de cigarette par spectrométrie de masse"
Adama Kamissoko, Vincent Carré, Frédéric Aubriet

14h30 **O20 -** "SpiderMass, a high potential tool using in-vivo and real-time mass spectrometry for the guided surgery of cancer"
Saudemont P., Fatou B., Wisztorski M., Ziskind M., Focsa C., Salzet M., Fournier I.

14h45 **O21 -** "Heme-binding as biodereplication method for search antimalarial drugs in complex extracts mixtures: active compounds from Piper coruscans (Miq.) C.D. (Piperaceae)"
Pedro Vásquez-Ocmín, Billy Cabanillas, Sandrine Cojean, Agathe Martinez, Jean-Marc Nuzillard, Mehdi Beniddir, Bruno Figadère, Alexandre Maciuk

15h00 **O22 -** "Tracking Biomarkers in Cerebral Malaria by Mass Spectrometry Imaging and Spatially-Resolved Microproteomics"
Tristan Cardon, David Koffi, Jusal Quanico, Hélène Yapo Etté, Michel Salzet, Joseph Djaman, Sylviane Pied, Isabelle Fournier

15h15-15h45 : Intervention de Sandra Beghi et Guy Lacour, Sistec

"Analyse de composés organiques dans l'air ambiant par spectrométrie de masse en ligne EXTREL"

16h00-17h30 : "Développement de méthodes de quantification ciblée de peptides (SRM, PRM et DIA) et applications"

Christine Carapito
Laboratoire de Spectrométrie de Masse BioOrganique (LSMBO)
Université de Strasbourg, IPHC

17h30-18h00 : Table ronde animée par Christine Carapito – LSMBO – et Quentin Enjalbert – ANAQUANT.

"Applications SRM : Problèmes rencontrés en quantification"

Repas

&

Soirée de Gala

…Carnaval…

Tenue correcte exigée!

Vendredi 25 mars

Modérateur: Maud Fumex

08h30-10h15 : “Ambient Ionization Mass Spectrometry”

Marion Girod

Institut des Sciences Analytiques (ISA), UMR 5280 CNRS-Université Lyon 1

10h30-12h15 : “Ambient Ionization Mass Spectrometry”

Marion Girod

Institut des Sciences Analytiques (ISA), UMR 5280 CNRS-Université Lyon 1

12h30 : Départ en car du Bol Vert

Arrivée prévue à la gare de Valenciennes vers 13h45

Paniers repas distribués dans le car

Présentations Sponsors

Florian Marty, Ph.D.
Product Support Manager
(florian.marty@biognosys.com)

Best of both worlds: Combining quantitative targeted proteomics with high-content discovery proteomics

Biognosys is the leading next generation proteomics company. Using our proprietary HRM-MS approach we have recently identified over 9'000 proteins in a single shot data independent acquisition (DIA) measurement of mouse cerebellum tissue. This is to date the highest number of proteins identified in one single measurement. Apart from the discovery pipeline Biognosys has a long history in targeted proteomics using technologies such as multiple reaction monitoring (MRM) and parallel reaction monitoring (PRM). Here we present a workflow that combines the quantitative power of stable isotope-labelled reference peptides with the discovery potential of HRM-MS. Using the reference peptides from our PlasmaDive and PlasmaDeepDive multiplexed panels we have obtained absolute quantitative information of 173 proteins in native human plasma. Additionally, over 300 plasma proteins were identified in a single shot experiment. These proteins can be quantified over a large cohort with high reproducibility to obtain relative quantitative information. Further, the quantification values obtained for the SIS peptides can be used to extrapolate the absolute quantitative values of all proteins identified. This principle can be applied to any predefined panel of identified biomarkers for any sample type.

Sabine Jourdain
Sales Manager Life Science & Applied Markets France
(Sabine.Jourdain@bruker.com)

Solutions en spectrométrie de masse pour le criblage et la quantification en routine

Etienne Maout
Account Manager Biotech & MS technologies
(em@shimadzu.fr)

SFE SFC MSMS principes et applications

Cette présentation vous propose de découvrir la SFC, ses notions de base, le système Nexera ainsi que le configuration SFE - SFC - MS Online. Le principe de préparation et analyse d'échantillons en ligne sera détaillé et des exemples d'applications vous seront présentés

Bénédicte Duretz
Sales Support Manager
(benedicte.duretz@thermofisher.com)

Potentialités du couplage IC/MS pour l'analyse de pesticides polaires

L'analyse de composés polaires dans les échantillons environnementaux est une problématique particulièrement délicate qui requiert des détecteurs sensibles et capables de s'affranchir des effets matrices. La chromatographie ionique (CI) couplée avec la spectrométrie de masse (IC-MS) est une alternative de choix pour l'analyse de nombreux contaminants polaires tels que le glyphosate et ses métabolites. L'IC présente en effet dans ce cadre une spécificité et une sélectivité bien supérieure à la chromatographie en phase inverse. Le couplage de cette technologie avec la spectrométrie de masse (Example : Triple Quadrupole et Instrument équipé de technologie OrbiTrap) constitue donc une approche de choix pour l'analyse de cette classe de composés. Afin de rendre la phase mobile IC compatible avec la source d'ionisation du spectromètre de masse, on place en sortie de l'instrument, un suppresseur qui aura pour objectif de « déssaler » l'éluant et le rendre compatible avec la source HESI du spectromètre de masse. On pourra également ajouter un éluant de phase organique à l'entrée de la source de manière à améliorer le processus de désolvatation des ions et par voie de conséquence la sensibilité. Nous présenterons ici les résultats obtenus pour l'analyse du glyphosate et de ses métabolites dans les eaux et les matrices alimentaires.

Christophe Siroit
MS Sales Specialist
(christophe_siroit@waters.com)

Innovations technologiques au service de la sensibilité, avec le nouveau triple quadripôle, le Xevo TQXS

Les exigences imposées aux laboratoires ne cessant d'évoluer, ces derniers doivent impérativement s'adapter au changement de plus en plus vite et aux challenges toujours plus poussés. Pour répondre à ces attentes, Les équipes R&D de Waters ont développé le spectromètre de masse triple quadripôle le plus sensible jamais construit, le Xevo® TQ-XS.

Voici les quelques innovations technologiques qui seront présentées :

- Une sonde sans maintenance, qui réduit le temps nécessaire aux différents entretiens et assure une reproductibilité parfaite, quel que soit l'utilisateur.
- Le guide d'ions StepWave XS™, qui assure une sensibilité accrue pour les composés les plus complexes.
- Un système de détection amélioré, doté d'une gamme dynamique de 6 ordres de grandeur, pour une sensibilité toujours accessible et un transfert de méthode aisé.
- **Une nouvelle technologie de source, Unispray™.** Repoussant toutes les limites, UniSpray™ est capable d'ioniser une gamme de composés bien plus vaste que l'ESI standard. Couvrant un large éventail de composés et incroyablement simple à utiliser, la technologie UniSpray permet à votre laboratoire d'analyser un nombre sans précédent de composés, sans même devoir changer de source ni de sonde.

Guy LACOUR, Sandra BEGHI

Ingénieur commercial et Ingénieure d'affaires

(glacour@sistec-instrumentation.com

SBeghi@sistec-instrumentation.com)

Analyse de composés organiques dans l'air ambiant par spectrométrie de masse en ligne EXTREL

L'analyse en ligne, sélective et rapide de contaminants type composés organiques volatils (COV) dans l'air ambiant est à l'heure actuelle un défi de santé publique que peu de techniques analytiques peuvent relever. Les spectromètres de masse quadripôles en ligne EXTREL MAX 300, fonctionnant hors couplage, satisfont aux exigences des analyses environnementales des composés organiques volatils par leur rapidité de réponse (de l'ordre de 400 ms/composé), leur haute précision et leur limite de détection de l'ordre de 10 ppb sans préconcentration. En effet, l'innovation d'EXTREL vient de l'utilisation d'un multiplicateur d'électrons en complément du détecteur Faraday.

Cette rapidité et cette sensibilité permettent aussi de réaliser des mesures séquentielles sur de nombreux points de mesure, offrant un temps global « de boucle » sur plusieurs dizaines de points égal à ce qu'offre un couplage chromatographique sur un point unique.

L'EXTREL MAX 300 a fait ses preuves pour l'analyse en ligne (24/24 ; 7/7) des Benzène Toluène Xylène (BTX, 10 unités installées), pour l'analyse de monomère de chlorovynil et dichloroéthylène et Acrylonitrile (34 Unités installées) à l'air ambiant. La gamme EXTREL MAX 300 se décline aussi pour des applications procédés, industries chimie, pétrochimie, pharmacie, etc.

ANAQUANT

SOLUTIONS FOR BIOANALYSIS

Quentin Enjalbert
Business development
(quentin.enjalbert@anaquant.com)

ANAQUANT is a private Contract Research Organization (CRO) specialized in targeted protein quantification by Mass spectrometry for Biologics and Biomarkers analysis.
Founded in 2014 by Tanguy Fortin (PhD), ANAQUANT has established a strong partnership with the Institute for Analytical Sciences (ISA / CNRS UMR 5280) headed by the Professor Jérôme Lemoine. Thus, the company benefits of leverages proprietary technology and more than 15 years of experience of the laboratory's team with industrial companies.
As expert and trusted partner, ANAQUANT provides customized services and standards to pharmaceutical and biotech companies during early-Nonclinical phases of biotherapeutics development.
Our mission is to bring our partners reliable data which contribute to the selection of their best drug candidates and guarantee a better control of their biologic to limit failure during further clinical phase.

YOUR PARTNER FOR LABELLED COMPOUNDS

(^{13}C, ^{15}N, D, ^{18}O)

Renseignements:

Romain Hubert

Responsable Marketing et Communication

(rhubert@eurisotop.com)

Fort d'une expérience de 25 ans, Euriso-top synthétise et commercialise la plus large gamme de composés marqués par des isotopes (D, ^{13}C, ^{15}N, ^{18}O).

C'est ainsi que depuis sa création en plein cœur du CEA de Saclay, que l'entreprise Euriso-top s'est imposée comme le leader européen en solvants deutérés et en molécules marquées, en proposant plus de 15.000 références produits, couvrant de multiples domaines de la recherche.

Elle confirme ainsi sa vocation d'être, pour toute la communauté scientifique, le partenaire privilégié de leurs recherches.

Euriso-top®

Parc des Algorithmes

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Renseignements:
Pieter Dekocker
Responsable Marketing et Communication
(pieter.dekocker@ionbench.com)

Manufacturer of laboratory furniture for mass spectrometry (LC/GC/MS) & Elevating UHPLC benches. Mass Spec IonBench products integrate MS peripherals, a built-in vacuum pump noise reduction enclosure and protect turbo molecular pumps by reducing vibration by 99%. There is up to 30% savings in laboratory space allocation. Solidly built lockable casters simplify moving the system. Our integrated vacuum pump enclosure reduces noise emissions by 80% down in perception. LC Elevating IonBench, on caster wheels, can be easily lifted up or down by commuting a switch, for a convenient & safe access to the top of your UHPLC.

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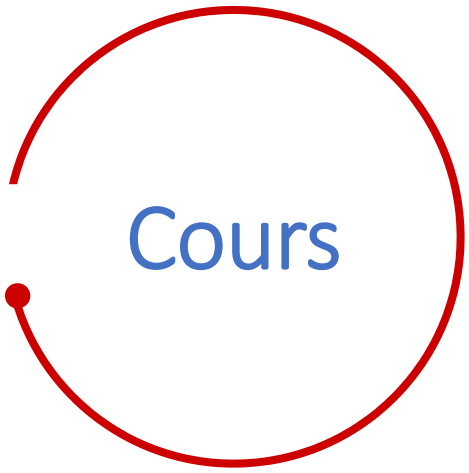


Ambient Ionization Mass Spectrometry

Marion Girod
Institut des Sciences Analytiques (ISA), UMR 5280 CNRS-Université Lyon 1

22èmes rencontres du club jeune SFSM - 20 au 24 mars 2017 - Trélon

Ambient Sampling/Ionization Mass Spectrometry

Marion Girod

Institut des Sciences Analytiques, UMR 5280, CNRS , Université Lyon 1 et ENS Lyon

Overview

I- Introduction

II- ElectroSpray Ionization-based ambient ionization techniques
- II-1) Direct Desorption : DESI, EASI
- II-2) Sampling and Transferring through a gas stream: EESI, ND-EESI, FD-ESI
- II-3) Laser Desorption/ablation : ELDI, MALDESI, LAESI, LIAD-ESI
- II-4) Thermal Desorption : ESA-Py, TPD/ESI
- II-5) Direct ionization: PSI , LSI

III- Atmospheric Pressure Chemical Ionization-based ambient ionization techniques
- III-1) Direct Desorption : DAPCI, DBDI, LTP, PADI , DCBI
- III-2) Laser Desorption/ablation : LD-APCI, LDTD
- III-3) Thermal Desorption : DART, ASAP

IV- Atmospheric Pressure Photo-Ionization-based ambient ionization techniques : DAPPI

V- Comparaison of techniques

VI- Conclusions

I- Introduction

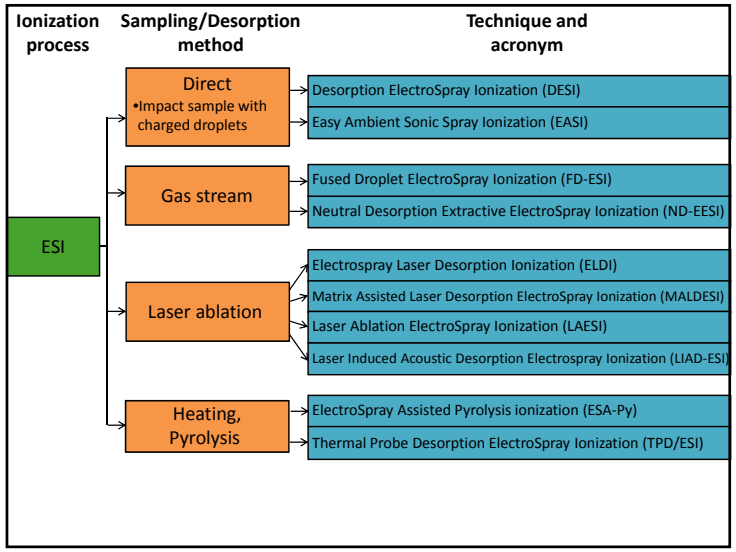
- Ambient ionization mass spectrometry allows the rapid analysis of samples or objects in their native state in the open environment with no or little prior preparation.

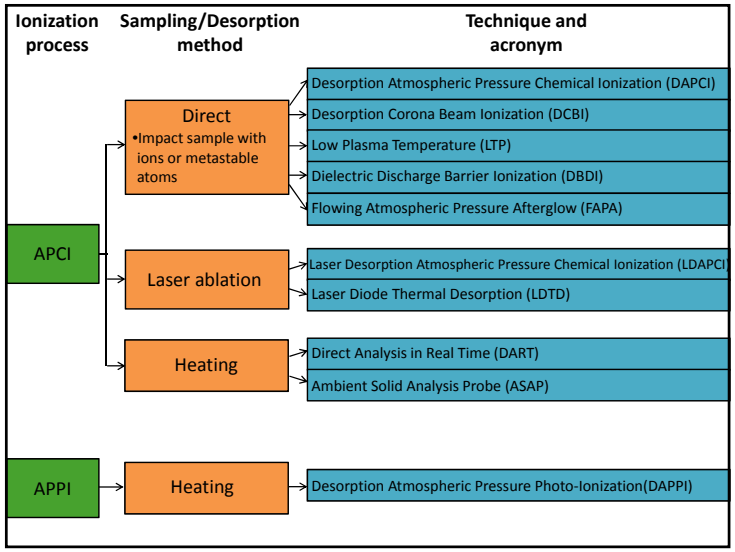
- Ambient ionization techniques combine a **desorption process** of the analytes, such as laser, thermal desorption, or direct exposure of the analyte surface to the charged species, with their **ionization**. Electrospray, atmospheric pressure chemical ionization and photo-ionization are the most used techniques for the ionization in ambient MS.

- These techniques allow the analysis of a wide range of substances (polar/non-volatile and non-polar/volatile) from various surfaces and matrices.
Broad application areas, both qualitative and quantitative in nature, including pharmaceutical analysis, process chemistry, biological imaging, in vivo analysis, proteomics, metobolomics, forensics and explosives detection.

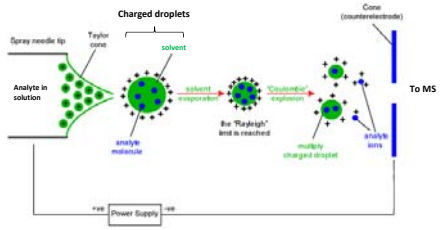
I- Introduction

-Since the early 2000's, many ambient ionization techniques have been developed (~ 30 ambient ionization sources are reported in the literature) due to the large number of possible combinations of desorption and ionization processes, giving rise to the proliferation of many acronyms always more complicated.





II- ESI-based ambient ionization techniques



In ambient ionization techniques using this mode of ionization, the **solvent charged droplets** formed by ESI allow the **post-ionization** of the desorbed analytes.

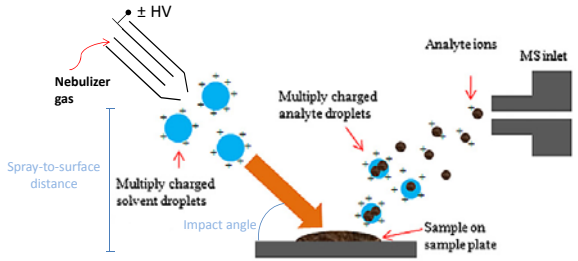
- Direct desorption/ionization
- Two-step ionization: desorption/sampling first and then contact with the ESI plume

Samples can be gaseous, liquid or solid. The use of ESI for the post-ionization produces singly and multiply charged ions, which allows analysis of small species as well as biological macromolecules.

II-1) Direct Desorption

Analytes are directly desorbed from the surface of the sample by the ionization agent

Desorption ElectroSpray Ionization (DESI)



- Electrospray dissolves/extracts analytes on the surface: « droplet pick-up ».
- Secondary droplets are transported into inlet.
- Classical evaporation/fission of analyte droplets.

II-1) Direct Desorption

Desorption ElectroSpray Ionization (DESI)

- Instrumentation and operating parameters:

Parameter	Range of settings	
Solvent flow rate	3–5 µl.min^{-1}	Spot size
Nebulizer gas pressure	8–12 bar	
Spray voltage	2–6 kV	
Spray-to-surface distance	1–5 mm	
Spray-to-surface angle	30–70°	
Surface-to-MS inlet	1–3 mm	
Temperature of desolvation capillary	200–300°C	

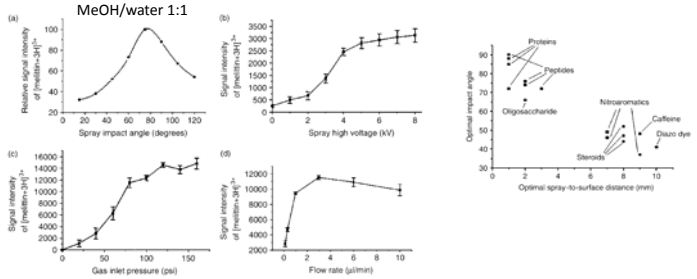
Surface = glass, PTFE, PMMA, TLC paper… → importance of the wettability

Solvent composition = organic solvent (EtOH, MeOH, ACN)/water ± acid or base; in some cases $CHCl_3$/MeOH or ACN; + specific reagent in ReactiveDESI → needs to desolve analytes, spray stability issues

II-1) Direct Desorption

Desorption ElectroSpray Ionization (DESI)

- Instrumentation and operating parameters:



Spray size	~ 1.24 mm	~ 1.38 mm	~ 1.59 mm	~ 2.01 mm
Gas Pressure:	150 psi	150 psi	200 psi	150 psi
Spray-to-surface distance:	5 mm	5 mm	5 mm	3 mm
H_2O :	20 %	50 %	50 %	20 %

Spot size ↗ : signal ↗ but spatial resolution ↘

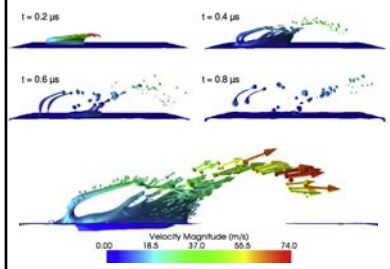
II-1) Direct Desorption

Desorption ElectroSpray Ionization (DESI)

- Characterization:

Chemical Physics Letters 464 (2008) 1–8

Multiphase computational fluid dynamics simulations

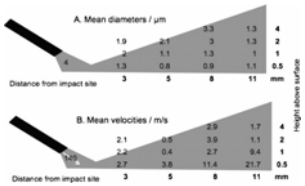


Anal. Chem. 1999, 71, 4111-4113

Phase Doppler Anemometry

MeOH/water 1:1

PTFE surface



Primary droplets size: 4 µm
Primary droplets velocity: 140 m/s

Secondary droplets size: ~ 1 µm
Secondary droplets velocity: 2 - 30 m/s

→ Surface-to-MS inlet angle important = 10-20°

II-1) Direct Desorption

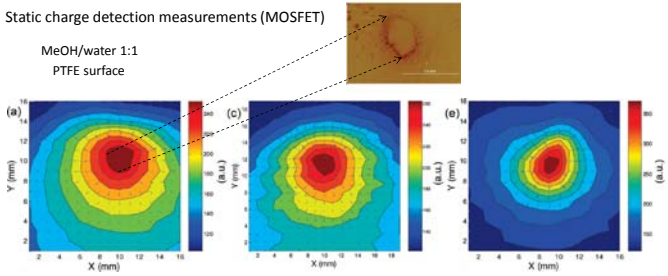
Desorption ElectroSpray Ionization (DESI)

- Characterization: *J. Phys. Chem. C, Vol. 114, No. 12, 2010*

Static charge detection measurements (MOSFET)

MeOH/water 1:1

PTFE surface



(a) 55° spray angle, 130 psi sheath gas pressure, and 2 µL/min flow rate;
(c) 30° spray angle, 100 psi sheath gas pressure, and 2 µL/min flow rate;
(e) 55° spray angle, 100 psi sheath gas pressure, and 1 µL/min flow rate

→Highest charge density located in the small desorption area (~1mm²)

II-1) Direct Desorption

Desorption ElectroSpray Ionization (DESI)

- Mechanisms of ion formation:

- For large molecules : mostly droplet «pick-up»

→ Large charged solvent droplets making contact with the surface, dissolving the analyte and then producing ions through ESI like mechanisms (fission/evaporation)

$$[M + mS + nH]^{n+}_{(l)} \rightarrow [M + mS + nH]^{n+}_{(g)} \rightarrow [M + nH]^{n+}_{(g)}$$

Solvated ion in liquid phase — Solvated ion in gas phase — Ion in gas phase

- For low molecular weight molecules: Droplet «pick-up» can occur but other mechanisms are proposed:

→ Direct charge transfer between the ionized solvent and the solid analyte at the surface which is removed through static repulsion or sputtering into the gas phase

$$S^+_{(aq)} + A_{(s)} \rightarrow S_{(aq)} + A^+_{(s)} \rightarrow A^+_{(g)}$$

→ Neutral volatization i.e. volatile analyte molecules leaving the surface, interacting with the spray itself, being ionized via charge transfer

$$S^+_{(g)} + A_{(g)} \rightarrow S_{(g)} + A^+_{(g)}$$

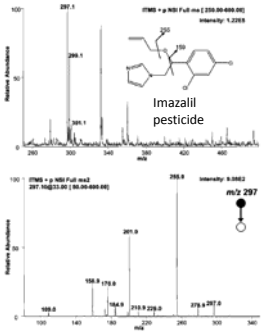
II-1) Direct Desorption

Desorption ElectroSpray Ionization (DESI)

- Applications:

Testing in food industry

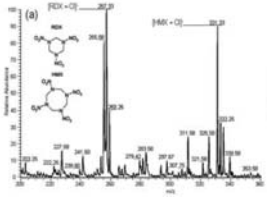
Anal. Chem. 2009, 81, 820–829



DESI-MS(/MS) on lemon peel

Homeland security

Analyst, 2010, 135, 1953–1960



DESI negative ion MS of a transfer wipe used to concentrate 33.3 ng of explosives spotted over a 10 cm² area onto a rough plastic surface

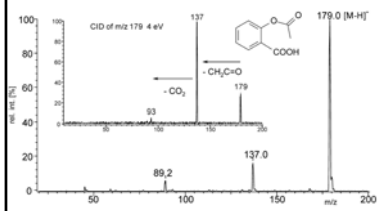
II-1) Direct Desorption

Desorption ElectroSpray Ionization (DESI)

- Applications:

Pharmaceutical industry

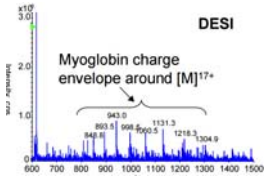
Anal. Chem. 2005; 77, 6915



Negative DESI-MS(/MS) on Aspirin tablet

Proteomics

Anal Chem, 86, 9603–9611



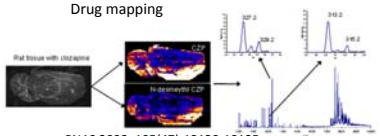
Positive DESI MS of myoglogin spotted onto a glass plate

II-1) Direct Desorption

Desorption ElectroSpray Ionization (DESI)

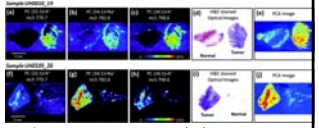
- Imaging: spatial resolution 35 µm

Drug mapping



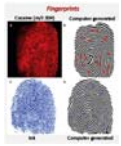
PNAS 2008; 105(47):18120-18125

Tumor Diagnosis



Chemistry-A Eur. J. 2010; 17(10), 2897- 2902

Chemical fingerprint

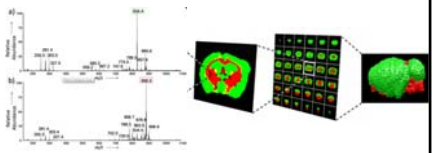


Science, 2008 , 321, 805 -805

Analysis of inks

Analyst 2007;132(5):461-467

3D molecular images



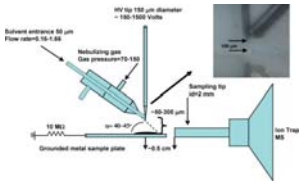
Angew Chem Int Ed Engl. 2010 ; 49(5): 873–876

II-1) Direct Desorption

Techniques closely related to DESI

Electrode-Assisted DESI (EADESI)

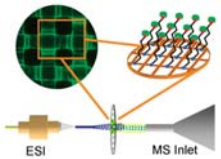
J. Mass. Spectrom. 2010, 45, 1203–1211



Sharp-edge tip subjected to a high applied voltage

Transmission mode DESI (TM-DESI)

Anal. Chem. 2010, 82, 16–18



Passing ESI plume through a sample pre-deposited on a mesh substrate

Desorption Ionization by Charge Exchange (DICE)

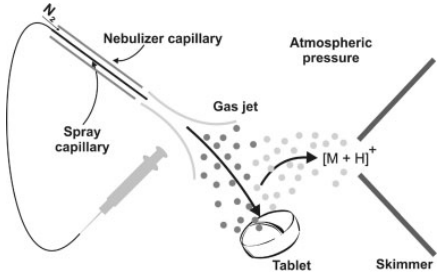
J Am Soc Mass Spectrom 2010, 21, 1554 –1560

Use toluene as solvent for the spray to ionize low polarity analytes by charge exchange

II-1) Direct Desorption

Easy Ambient Sonic spray Ionization (EASI)

previously referred to Desorption Sonic Spray Ionization (DeSSI)



- Neutral (super)sonic spray impinges on surface.
- Spray dissolves analytes.
- Secondary droplets are transported into inlet.
- Ionization via non-statistical charge distribution.

II-1) Direct Desorption

Easy Ambient Sonic spray Ionization (EASI)

- Instrumentation and operating parameters:

Nebulizer gas pressure = 200 psi (~15 bar)
Solvent mixture = MeOH/water 1:1 + 0.1 % formic acid
No heating and voltage are required

- Mechanisms of ion formation:

* High-velocity sprayer forms small droplets.
* Ionization occurs due to statistical imbalanced distribution between cations and anions in the spray.
* Then, desorption and ionization of the analytes from the surface follow similar mechanisms as DESI.

→ No electrochemical or oxidation processes, low charge density of droplets

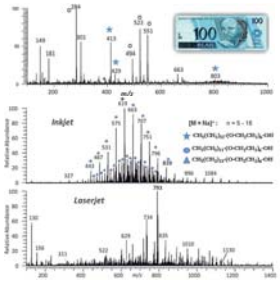
II-1) Direct Desorption

Easy Ambient Sonic spray Ionization (EASI)

- Applications:

Forensic

Analyst, 2010, 135, 2533–2539

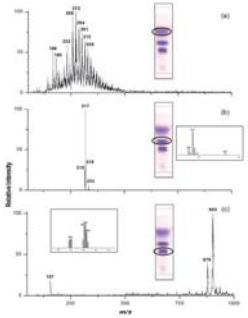


EASI(+) MS spectra of (a) authentic Brazilian R$ banknote and counterfeit banknotes made by using (b) inkjet and (c) laserjet printers

Biodiesels analysis

Analyst, 2009, 134, 1652–1657

Coupled with TLC separation

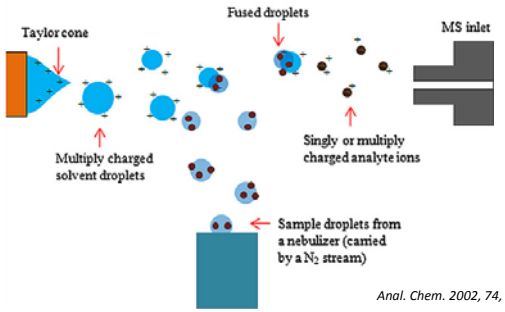


On-spot HPTLC-EASI-MS of: (a) petrodiesel; (b) soybean biodiesel; and (c) soybean oil

II-2) Sampling/Transferring through a gas stream

Gas is used to transport the analytes and put them in contact with the charged droplets

Fused Droplet ElectroSpray Ionization (FD-ESI)



Anal. Chem. 2002, 74, 2465-2469

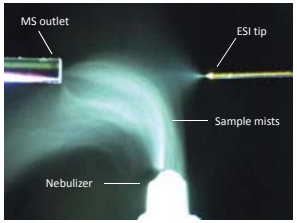
- Analyte droplets generated from a nebulizer.
- Analyte droplets are delivered in N_2 gas stream to the ESI plume .
- Analyte droplets fusing with the charged solvent droplets.

II-2) Sampling/Transferring through a gas stream

Fused Droplet ElectroSpray Ionization (FD-ESI)

- Characteristics:

For liquid samples
Ultrasonic, pneumatic or piezoelectric nebulizer (~1µm neutral droplets)
Analyte solution in « native » conditions
ESI solvent mixture adapted to the analyte polarity



- Mechanisms of ion formation:

* Fusion of analyte droplets with ESI droplet.
* Analytes are selectively extracted into the charged droplets.
  → Low salts solubility in MeOH → FD-ESI has a higher salt tolerance than ESI
* Charge transfer between the analytes and the charged solvent.
* Then, classical evaporation/fission of analyte droplets.

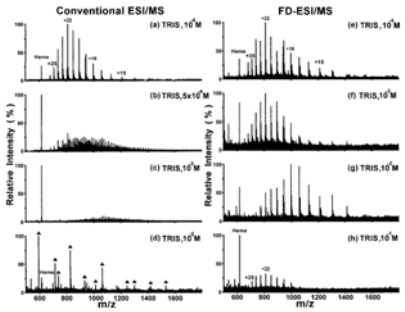
II-2) Sampling/Transferring through a gas stream

Fused Droplet ElectroSpray Ionization (FD-ESI)

- Application:

Proteomics

J. Proteome Res., 2005, 4 (2), pp 606–612



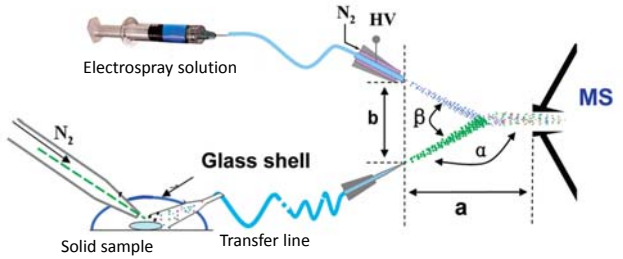
Conventional ESI (left side, a-d) and FD-ESI (right side, e-h) mass spectra of myoglobin/TRIS solutions.

→ No interference from salts using FD-ESI

II-2) Sampling/Transferring through a gas stream

Neutral Desorption Extractive ElectroSpray Ionization (ND-EESI)

J. Mass Spectrom. 2007; 42: 1123–1135

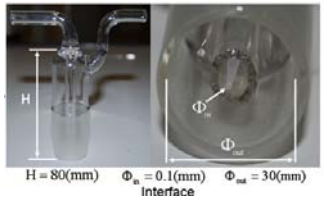


- Analyte desorption by N_2 stream.
- Desorbed neutrals are transported to the ESI plume.
- Analytes merge with the charged droplets for post-ionization by charge transfer.

II-2) Sampling/Transferring through a gas stream

Neutral Desorption Extractive ElectroSpray Ionization (ND-EESI)

- Characteristics:
  - For solid samples
  - Unheated gas stream (20°C)
  - Gas flow of 200 mL/min
  - Gas-to-surface angle = 30-90°
  - Glass enclosure to cover the sample
  - Transfer line length up to 10 m
  - Classical ESI solvent mixture; + specific reagent for ion/molecule reactions
  - Angle between the ESI plume and the neutral analyte plume β = 50-60°



- Mechanisms of ion formation:

  Sames mechanisms as FD-ESI; high matrix salt tolerance
  Difficulty to desorb large molecules with an unheated gas stream: limit < *m/z* 500

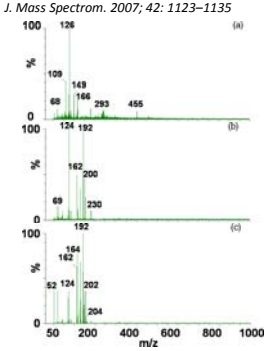
II-2) Sampling/Transferring through a gas stream

Neutral Desorption Extractive ElectroSpray Ionization (ND-EESI)

- Applications:

Food

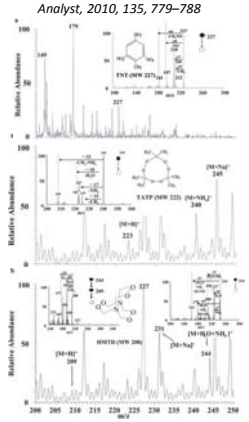
J. Mass Spectrom. 2007; 42: 1123–1135



ND-EESI spectra of frozen beef samples: (a) without exposure to room temperature; (b) after exposure to room temperature for 1 day; (c) after exposure to room temperature for 2 days.

Homeland Security

Analyst, 2010, 135, 779–788



LOD 5pg

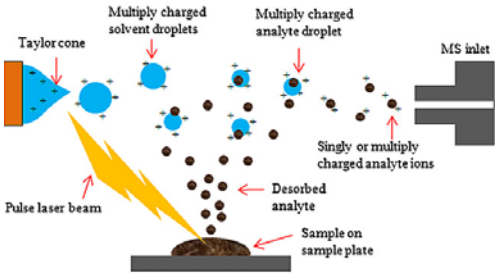
ND-EESI spectra of explosives on human skin

II-3) Laser Desorption/ablation

Analytes are desorbed from the surface by a laser pulse

Electrospray Laser Desorption Ionization (ELDI)

Rapid Commun. Mass Spectrom. 2005; 19: 3701–3704

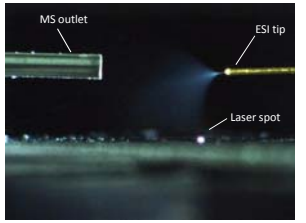


- Laser desorbs/ablates neutrals from sample surface.
- Neutral plume is entrained by the ESI droplets cloud.
- Analytes are desolved in the droplets and ionized through ESI mechanisms.

II-3) Laser Desorption/ablation

Electrospray Laser Desorption Ionization (ELDI)

- Characteristics:
  - UV wavelength (Nd:YAG 266 nm or N_2 337 nm) for nanosecond duration @ 10 Hz
  - Pulse energy = 150-300 µJ
  - Incident laser beam angle to the surface = 45°
  - Surface = glass, PTFE, paper, TLC paper
  - ESI solvent adapted to the analyte polarity
  - For small organic molecules and large biomolecules



- Mechanisms of ion formation:

  * Absorption of the laser energy by the analytes.
  * Excitation and sublimation of the analytes.
  * Formation of gaseous plasma plume of neutral analytes.
  * Dissolution of the analytes in the ESI droplets.
  * Ionization via charge transfer, then, classical evaporation/fission of analyte droplets.

II-3) Laser Desorption/ablation

Techniques closely related to ELDI

Laser Ablation ElectroSpray Ionization (LAESI)

Mid-IR wavelength (2.94 µm) for 5 ms duration @ 100 Hz
Pulse energy = 100 µJ
Incident laser beam angle to the surface = 90°

InfraRed Laser Desorption ElectroSpray Ionization (IR-LDESI)

IR wavelength (10.6 µm) for 5 ms duration @ 5 kHz
Pulse energy = 100 µJ
Incident laser beam angle to the surface = 90°

Matrix-Assisted Laser Desorption ElectroSpray Ionization (MALDESI)

Mid-IR wavelength (2.94 µm) or UV (349 nm) for 5 ms duration @ 100 Hz
Pulse energy = 100 µJ
Incident laser beam angle to the surface = 90°
Use of matrix for desorption

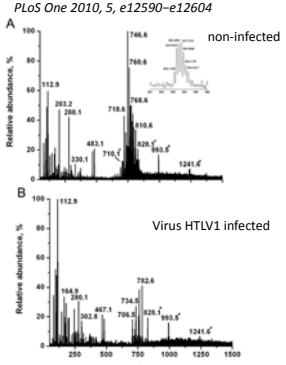
II-3) Laser Desorption/ablation

Electrospray Laser Desorption Ionization (ELDI) and others….

- Applications:

Metabolomics

PLoS One 2010, 5, e12590–e12604

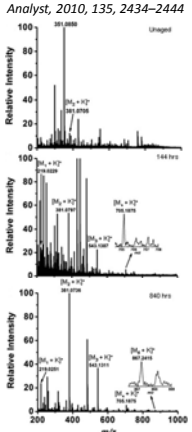


(+) LAESI MS spectra of T-lymphocytes

Q-ToF

Forensics

Analyst, 2010, 135, 2434–2444



cellulose degradation

(+) LAESI MS on paper

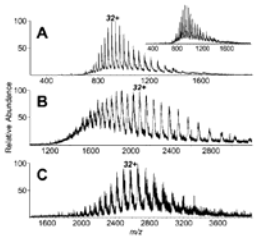
II-3) Laser Desorption/ablation

Electrospray Laser Desorption Ionization (ELDI) and others….

- Applications:

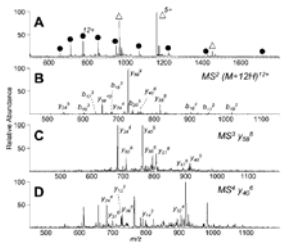
Proteomics

Analyst, 2010, 135, 767–772



ELDI-MS spectra (A) 1 mL of 200 mM bovine carbonic anhydrase, (B) 1 mL of 500 mM bovine serum albumin, and (C) 1 mL of 500 mM transferrin applied on-target. 1.06 µm; solvent mixture: ACN/water 1:1 + 0.2 % FA

Top-down analysis



ELDI-MS and MS^n of bovine ubiquitin. (A) IR-ELDI-MS. (B) MS^2 analysis of the 12+ ubiquitin ion (*m/z* 714). (C) MS^3 of y_{58}^8 ion (*m/z* 817). (D) MS^4 analysis of y_{40}^6 ion (*m/z* 761)

LTQ-Orbitrap

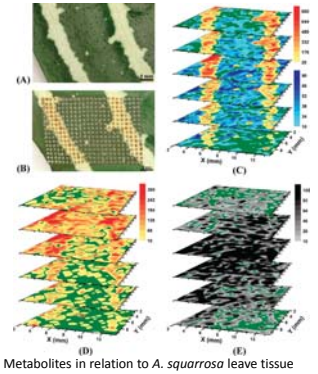
II-3) Laser Desorption/ablation

Electrospray Laser Desorption Ionization (ELDI) and others….

- Imaging – spatial resolution = 100 µm:

Plant slice

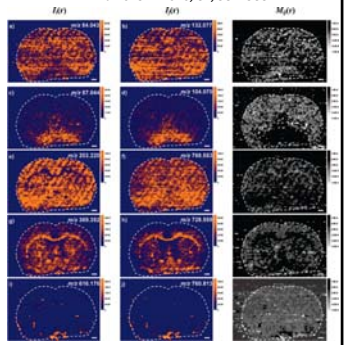
Anal. Chem. 2009, 81, 6668–6675



Metabolites in relation to *A. squarrosa* leave tissue architecture captured by LAESI 3D imaging MS

Biological sample

Anal. Chem. 2010, 82, 982–988

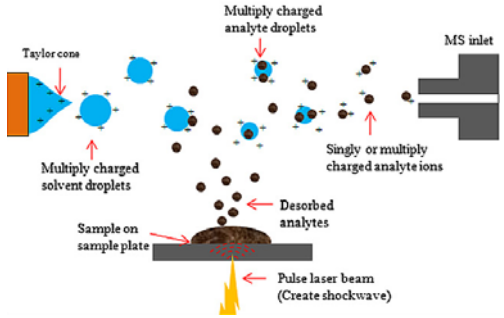


LAESI MS imaging of rat brain: (a) ethanolamine (b) creatine; (c) *γ*-butyrolactone (d) *γ*-aminobutyric acid; (e) spermine (f) PC(35:4); (g) cholesterol-H2O (h) plasmalogens PC(O-33:3); and (i) heme (j) R chain of hemoglobin.

II-3) Laser Desorption/ablation

Laser-Induced Acoustic Desorption ESI (LIAD-ESI)

Anal. Chem., 2009, 81 (3), pp 868–874



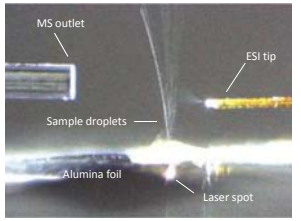
- Analytes are desorbed or aerosolized by laser-induced acoustic wave through special substrate.
- Neutral plume is entrained by the ESI droplets cloud.
- Analytes are desolved in the droplets and ionized through ESI mechanisms.

II-3) Laser Desorption/ablation

Laser-Induced Acoustic Desorption ESI (LIAD-ESI)

- Characteristics:

Nd:YAG 532 nm or 10.64 µm laser for nanosecond duration @ 10 Hz
Pulse energy = 100 mJ
Laser beam backside (45°)
Surface = titanium or aluminum foil (10-15 µm thick)
ESI solvent adapted to the analyte polarity



- Mechanisms of ion formation:

* Absorption of laser energy by the thin foil produces mechanical tension and generate an acoustic pulse, that propagates toward the other side and desorbs analytes.
* Fusion of analyte droplets with ESI droplet.
* Charge transfer between the analytes and the charged solvent.
* Then, classical evaporation/fission of analyte droplets.

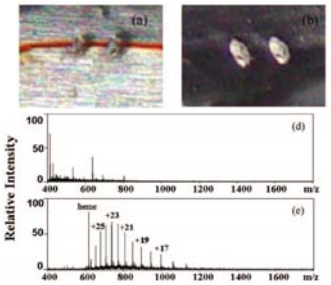
II-3) Laser Desorption/ablation

Laser-Induced Acoustic Desorption ESI (LIAD-ESI)

- Applications:

Proteomics

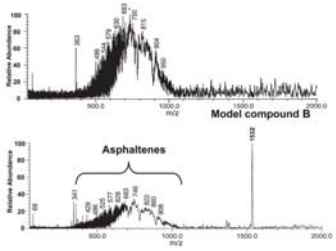
Anal. Chem. 2009, 81, 868–874



Photographs of the (a) front and (b) rear sides of an aluminum foil, revealing the path of desorption of a dry myoglobin solution line after irradiation of the rear side of the foil using an IR laser.
(d and e) Positive-ion LIAD-ESI mass spectra of the background and myoglobin

Fuel analysis

Energy Fuels 2009, 23, 5564–5570



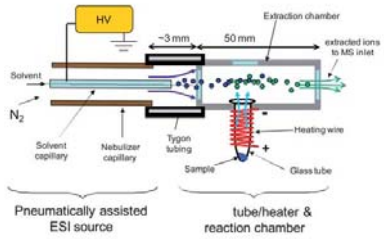
Top: LIAD-ESI (60 eV, 18 mJ, 30 laser pulses) mass spectrum of an asphaltene sample.
Bottom: asphaltene sample spiked with model compound B (MW 1532)

II-4) Thermal Desorption

Analytes are desorbed from the surface by heating or pyrolysis

ElectroSpray-Assisted Pyrolysis Ionization (ESA-Py)

Anal. Chem. 2005, 77, 7744-7749



- Analytes are desorbed by pyrolysis/heating.
- Desorbed gaseous analytes are in contact with the charged droplets in the extraction chamber.
- Charge transfer with the charged solvent.

II-4) Thermal Desorption

ElectroSpray-Assisted Pyrolysis Ionization (ESA-Py)

- Characteristics:

Pyrolysis up to 940°C
Other classical temperature = 350°C
For volatile thermally stable analytes
ESI solvent adapted to the analyte polarity

- Mechanisms of ion formation:

* Analytes are put into the gas phase by thermal desorption.
  → Possible thermal decomposition products
* Analytes are in contact with the charged droplets in the reaction chamber.
* Charge transfer between the analytes and the charged solvent.
* Then, classical evaporation/fission of analyte droplets.

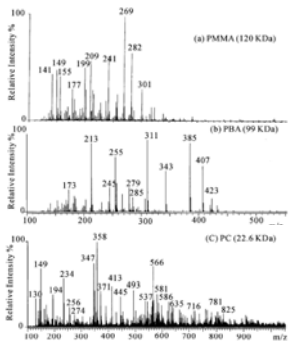
II-4) Thermal Desorption

ElectroSpray-Assisted Pyrolysis Ionization (ESA-Py)

- Applications:

Polymer analysis

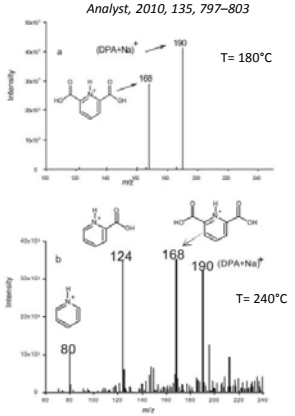
Anal. Chem. 2005, 77, 7744-7749



Positive-ion ESA-Py MS spectra of polymers
T= 590°C

Bacteriology

Analyst, 2010, 135, 797–803

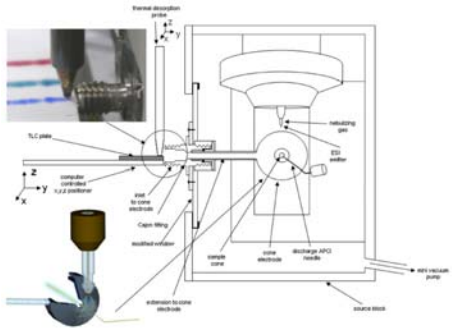


ESA-Py MS of *Bacillus* spores

II-4) Thermal Desorption

Thermal Probe Desorption ElectroSpray Ionization (TPD-ESI)

Rapid Commun. Mass Spectrom. 2010; 24 : 1721–1729



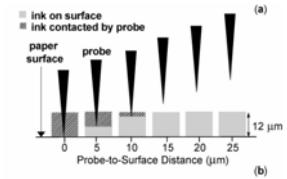
- Thermal probe desorbes the analytes from the surface.
- Desorbed gasous analytes are in contact with the charged droplets in a special cone electrode.
- Charge transfer between the analytes and the charged solvent.

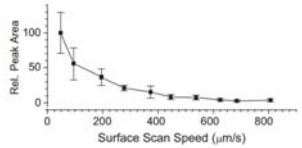
II-4) Thermal Desorption

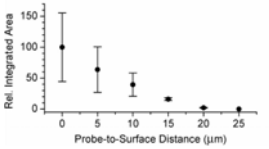
Thermal Probe Desorption ElectroSpray Ionization (TPD-ESI)

- Characteristics:

Classical temperature = 350°C
Probe tips diameter of 50 µm
Surface = Glass slide, TLC paper
Probe-to-surface distance ~ 5 µm
Classical ESI solvents adapted to the analytes
Imaging capability







- Mechanisms of ion formation:
Similar to ESA-Py

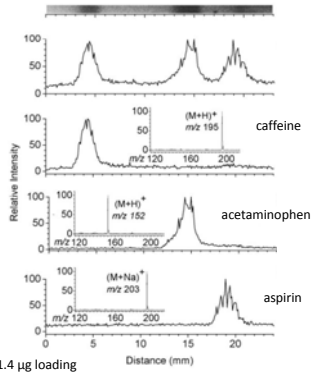
II-4) Thermal Desorption

Thermal Probe Desorption ElectroSpray Ionization (TPD-ESI)

- Applications:

Analysis on TLC plate

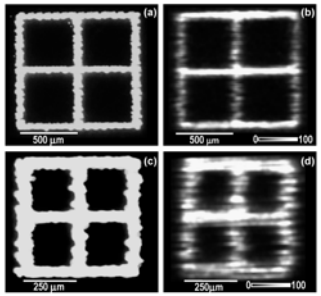
Rapid Commun. Mass Spectrom. 2010; 24: 1721–1729



1.4 µg loading
T= 350°C , scan speed 700 µm/s

Imaging of printed inks

Anal. Chem. 2011, 83, 598–603

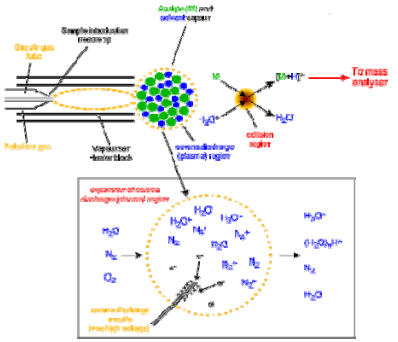


Optical images

Interpolated TPD-ESI MS chemical images (*m/z* 387)

T= 350°C , scan speed 100 µm/s
→ Spatial resolution 50 µm

III- APCI-based ambient ionization techniques



In ambient ionization techniques using this mode, the **charged species** formed by the APCI allow the **post-ionization** of the desorbed analytes (direct or in two steps).
Different methods are used to generate this plasma as a corona discharge, dielectric barrier discharge or glow-to-arc discharge.
Samples can be gaseous, liquid or solid. Widely used for the analysis of volatile and low polar compounds.

III- APCI-based ambient ionization techniques

Ionization of analyte molecules (M) by reactants (R)

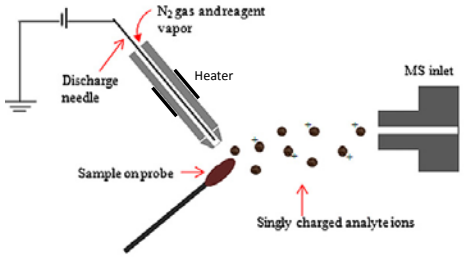
Ionization mechanism	Reaction pathway
Proton transfer	$M+[(H_2O)_n+H]^+ \rightarrow [M+H]^+ + nH_2O$
Charge transfer	$M+R^+ \rightarrow M^+ + R$ $R^+ = N_2^+, NO^+, O_2^+$ $M+R^- \rightarrow M^- + R$ $R^- = O_2^-$ (from $O_2 + e^- \rightarrow O_2^-$), OH^-
Penning ionization	$M+R^* \rightarrow M^{+\bullet} + R + e^-$ $R^* = He^*, N_2^*$
Electron attachment	$M+e^- \rightarrow M^{-\bullet}$
Ion attachment	$M+R^- \rightarrow [M+R]^-$ / $M+R^+ \rightarrow [M+R]^+$ $R^- = NO_2^-, NO_3^-$ / $R^+ = NH_4^+$
Proton/hydride abstraction	$M+R^-/R^+ \rightarrow [M-H]^-/[M-H]^+ + RH$ $R^- = OH^-$; $R^+ = NO^{+\bullet}$

III-1) Direct Desorption

Analytes are directly desorbed from the surface by the reagent species

Desorption Atmospheric Pressure Chemical Ionization (DAPCI)

Rapid Commun. Mass Spectrom. 2006; 20: 3130–3138



- Corona discharge is established between 2 electrodes.
- Reactive plasma desorbs analytes from the surface.
- Desorbed analytes are carried by the gas.
- Analytes ions are formed via ion/molecule reactions with plasma species.

III-1) Direct Desorption

Desorption Atmospheric Pressure Chemical Ionization (DAPCI)

- Characteristics:

Heated N_2 nebulizer gas (0.15~0.2 MPa, 250~450 mL/min)
Reagent solution = water, MeOH, ACN
Corona discharge voltage = ±4.5 kV
Discharge needle-to-surface distance = 2-3 mm
Discharge needle-to-surface angle = 30-45°
Surface = glass, PTFE, foodstuffs, skin…

- Mechanisms of ion formation:

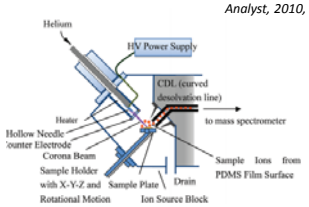
* Gas phase solvent vapors are ionized by corona discharge ionization.
* Reagent species (electrons, protons, metastable atoms, solvent ions = H_3O^+, N_2^+, OH^-, CN^-, CH_2CN^-) impact the surface.
* Analytes are desorbed by charge buildup on the surface : « chemical sputtering ».
* Charge or proton transfers with reagent species allow ionization of the desorbed analytes. Formation of singly charged analyte ions and complexes.

III-1) Direct Desorption

Technique related to DAPCI

Desorption Corona Discharge ionization (DCBI)

Analyst, 2010, 135, 688–695



No solvent needed
He reactive gas → ions and metastable He atoms
Low current (10-40 µA)

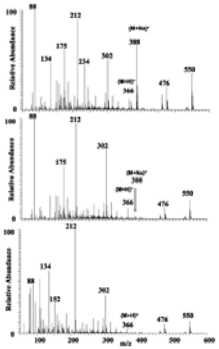
III-1) Direct Desorption

Desorption Atmospheric Pressure Chemical Ionization (DAPCI)

- Applications:

Pharmaceutical

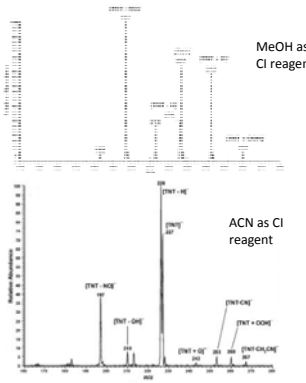
Anal. Chem. 2010, 82, 8060–8070



DAPCI(+) MS spectra of amoxicillin capsules from different origins

Homeland Security

Rapid Commun. Mass Spectrom. 2006; 20: 3130–3138

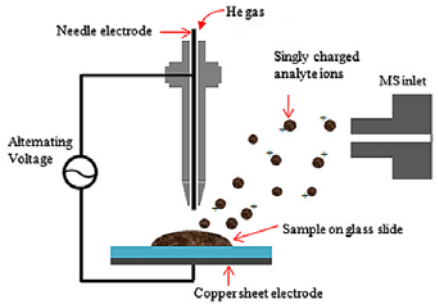


DAPCI(-) MS spectra TNT on cotton applicator

III-1) Direct Desorption

Dielectric Barrier Discharge Ionization (DBDI)

J Am Soc Mass Spectrom 2007, 18, 1859–1862



- Dielectric barrier discharge between the needle electrode and the sheet electrode generates plasma species.
- Reactive plasma desorbs analytes from the surface.
- Analyte ions are mainly formed via electron transfer with metastable atoms.

III-1) Direct Desorption

Dielectric Barrier Discharge Ionization (DBDI)

- Characteristics:

He gas (<0.2 L/min)
Alternative voltage of 3.5-4.5 kV @ 20 kHz
Copper sheet as counter electrode
Glass slide that served as both the discharge barrier and the sample plate
Discharge needle-to-surface distance = 5-10 mm

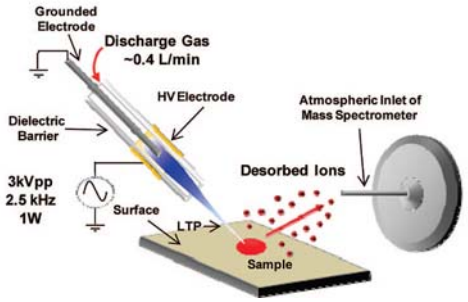
- Mechanisms of ion formation:

* Plasma is formed by dielectric barrier discharge.
* Reagent species (electrons, protons, metastable atoms, gas reagents) impact the surface.
* Analytes are desorbed by charge buildup on the surface : « chemical sputtering ».
* Ion/molecules reactions allow ionization of the desorbed analytes. Penning ionization by the bombardment of metastable gas may occur.

III-1) Direct Desorption

Low-Temperature Plasma (LTP)

Anal. Chem. 2008, 80, 9097–9104



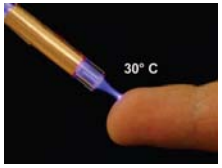
- Dielectric barrier discharge to create a low-temperature plasma.
- The counter electrode is placed within the probe.
- Direct interaction of the plasma with the sample.
- Analytes are ionized through proton or charge transfers.

III-1) Direct Desorption

Low-Temperature Plasma (LTP)

- Characteristics:

Glass tube (o.d. 6.35 mm and i.d. 3.75 mm) with an internal grounded electrode (stainless steel; d. 1.57 mm)
Outer electrode (copper tape) surrounding the outside
He, Ar, N_2, air gas (<0.4 L/min)
Alternative voltage of 2.5-5 kV @ 2-5 kHz
Probe-to-sample surface distance: 1 mm to 2 cm



- Mechanisms of ion formation:

Similar to DBDI (wide range of ionization processes)
Formation of $[M+H]^+$, $[M+H+(H_2O)_n]^+$, $[M+NO_2]^-$, $[M]^-$, $[M-NO_2]^-$

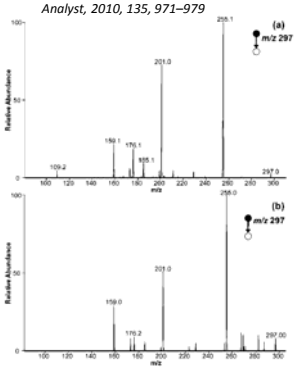
III-1) Direct Desorption

Low-Temperature Plasma (LTP)

- Applications:

Food Analysis

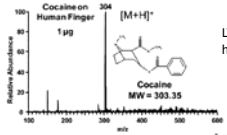
Analyst, 2010, 135, 971–979



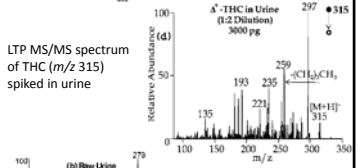
LTP MS/MS spectra of fungicide imazalil on market-purchased (a) orange and (b) lemon peel.

Drug abuse analysis

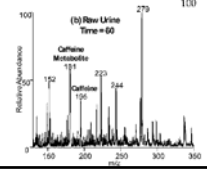
Analyst, 2010, 135, 927–933



LTP MS spectrum of humain skin



LTP MS/MS spectrum of THC (*m/z* 315) spiked in urine

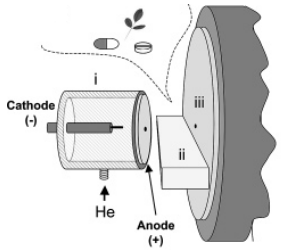


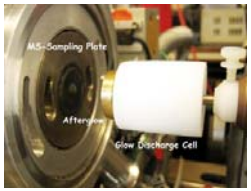
LTP MS spectrum after intake of a caffeinated (*m/z* 195) soft drink

III-1) Direct Desorption

Flowing Atmospheric Pressure Afterglow (FAPA)

Anal. Chem. 2008, 80, 2646-2653





- Glow-to-arc discharge generates metastable atoms.
- Gas stream is heated in discharge.
- Analytes ions are formed via proton transfer or charge exchange with metastable atoms.

III-1) Direct Desorption

Flowing Atmospheric Pressure Afterglow (FAPA)

- Characteristics:

Two electrodes (a cathode : 1.5 mm diameter tungsten pin mounted into a 5 mm diameter steel rod, and an anode: 10 mm diameter, 2 mm thick brass disk with a 1 mm diameter orifice in its center) tightly mounted in a discharge chamber outer electrode (copper tape) surrounding the inside
The body of the cell (T) is Teflon and has a suitable entrance orifice for the discharge gas (He, <0.4 L/min)
Direct current (DC) of 10-50 V

- Mechanisms of ion formation:

* Heating of the gas stream through Joule heating within the electrical discharge.
* Reagent plasma species impact the surface.
* Analytes are desorbed by charge buildup on the surface.
* Ion/molecules reactions allow ionization of the desorbed analytes.

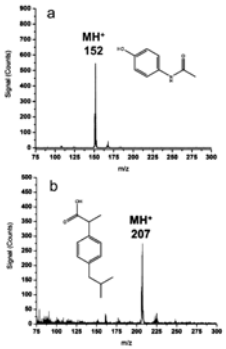
III-1) Direct Desorption

Flowing Atmospheric Pressure Afterglow (FAPA)

- Applications:

Pharmaceutical

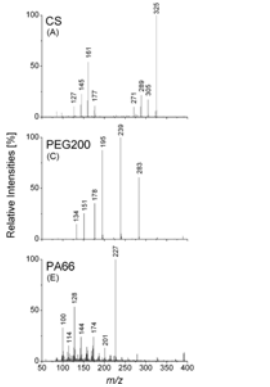
Anal. Chem. 2008, 80, 2654-2663



FAPA(+) MS spectra of (a) Tylenol tablet (acetaminophen), (b) Ibuprofen tablet

Polymer Analysis

Rapid Commun. Mass Spectrom. 2006; 20: 3130–3138



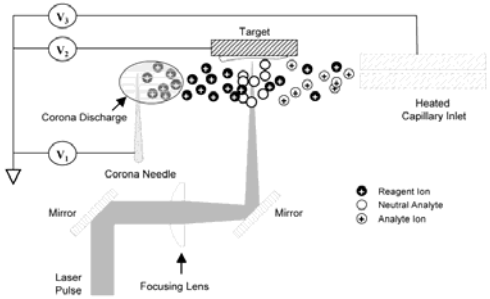
FAPA MS spectra of solid polymers

III-2) Laser Desorption/ablation

Analytes are desorbed from the surface by a laser pulse

Laser Desorption Atmospheric Pressure Chemical Ionization (LDAPCI

Anal. Chem. 2002, 74, 5600-5605



- Laser desorbs/ablates neutrals from sample surface.
- Corona discharge generates charged species.
- Reagent ions stream merges with neutral plume, reacting with analytes through ion/molecule reactions.

III-2) Laser Desorption/ablation

Laser Desorption Atmospheric Pressure Chemical Ionization (LDAPCI)

- Characteristics:

IR wavelength (CO_2 10.6 µm)
Incident laser beam angle to the surface = 90°
Spot diameter ~ 0.5 mm
Sample on stainless steel surface held at an offset potential of 2 kV.
The corona needle positioned ~3 cm from the inlet of the heated capillary
Corona discharge potential of +8.1 kV

- Mechanisms of ion formation:

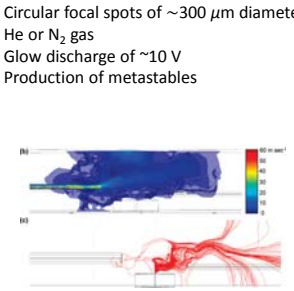
* Absorption of the laser energy by the analytes.
* Excitation and sublimation of the analytes.
* Formation of gaseous plume of neutral analytes.
* Neutral analytes react with plasma reagent species
* Formation of singly charged analyte ions and complexes.

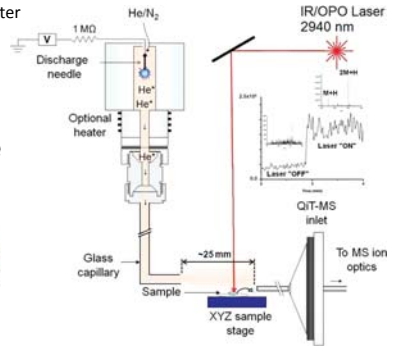
III-2) Laser Desorption/ablation

Techniques related to LD-APCI

InfraRed Laser Ablation Metastable-Induced Chemical Ionization (IR-LAMICI)

IR wavelength (2.94 µm) OPO Nd:YAG laser for 4 ns duration @ 20 Hz
Pulse energy = 2 mJ
Incident laser beam angle to the surface = 90°
Circular focal spots of ~300 μm diameter
He or N_2 gas
Glow discharge of ~10 V
Production of metastables





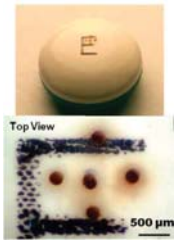
Anal. Chem. 2010, 82, 2178–2181

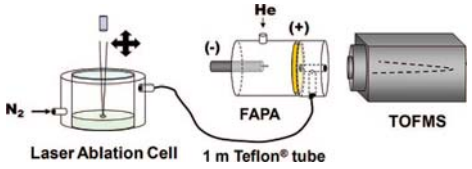
III-2) Laser Desorption/ablation

Techniques related to LD-APCI

Laser Ablation Flowing Atmospheric Pressure Afterglow (LA-FAPA)

UV 266 nm Nd:YAG laser operating at 20 Hz
Laser spot sizes between 10 and 300 μm in diameter.
Aerosol generated by the ablation event carried in a stream of N_2 at 0.3 L/min
1 m Teflon transfer tube to the FAPA chamber
Afterglow discharge. Direct current (DC) of 10-50 V





Anal. Chem. 2008, 80, 8308–8313

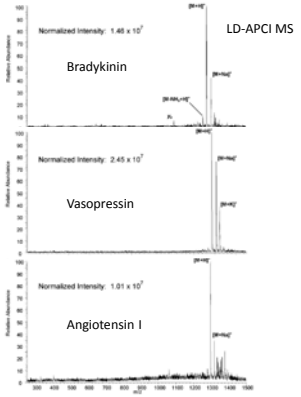
III-2) Laser Desorption/ablation

LD-APCI, IR-LAMICI and LD-FAPA

- Applications:

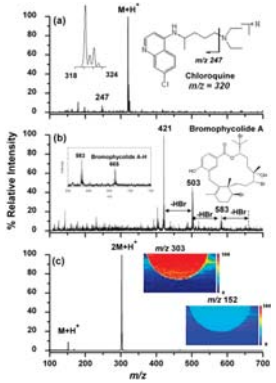
Peptideomics

Anal. Chem. 2002, 74, 5600-5605



Pharmaceutic

Anal. Chem. 2010, 82, 2178–2181



IR-LAMICI MS spectra of (a) a counterfeit artesunate antimalarial drug tablet; (b) a red macroalga, *Callophycus serratus*, and (c) a Tylenol tablet (325 mg of acetaminophen).

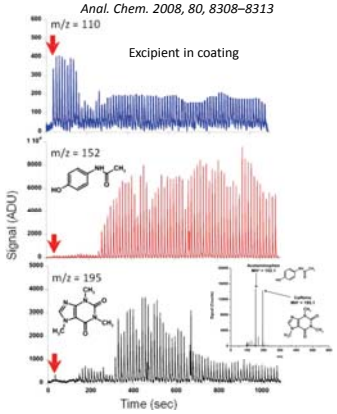
III-2) Laser Desorption/ablation

LD-APCI, IR-LAMICI and LD-FAPA

- Applications:

Pharmaceutical

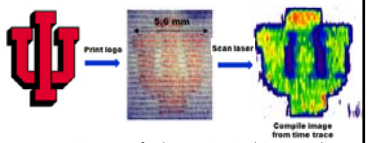
Anal. Chem. 2008, 80, 8308–8313



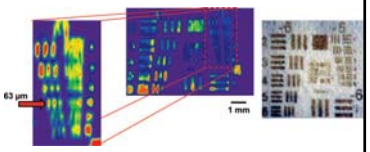
LA-FAPA depth profiling of an Excedrin tablet

Imaging analysis

Anal. Chem. 2008, 80, 8308–8313



LA-FAPA MS imaging of Indiana University logo printed on paper with caffeine-doped ink.



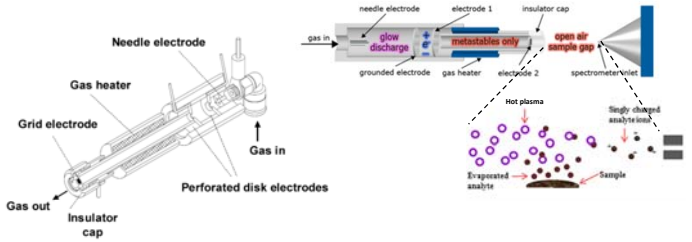
IR-LAMICI chemical images of the 1951 USAF resolution target (shown on right) printed with caffeine at 1200 dpi onto paper. Vertical and horizontal resolution are 178 and 63 µm.

III-3) Thermal Desorption

Analytes are desorbed from the surface by heating or pyrolysis

Direct Analysis in Real Time (DART)

Anal. Chem., 2005, 77, 2297–2302

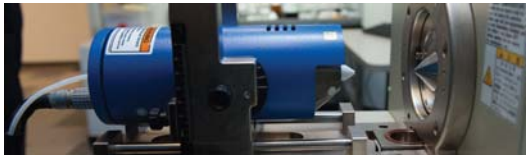


- Corona discharge generates plasma species. Ions from the plasma are removed; only metastable atoms are kept.
- Resistively heated gas stream thermally desorbs analytes.
- Analytes are ionized directly by metastables or via undirect ion/molecule reactions.

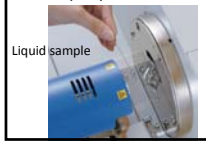
III-3) Thermal Desorption

Direct Analysis in Real Time (DART)

- Instrumentation and operating parameters:



He or N_2 nebulizer gas (~ 1L/min)
Corona discharge voltage = ± 1 to 5 kV
The potential of the second electrode (to remove ions) typical ± 100 V to ± 250 V
Gas temperature up to 250 °C
Typical DART/Sample/Orifice distance is 5 to 25 mm
Sample placed in «transmission» or «glancing» geometry


Liquid sample


Solid sample

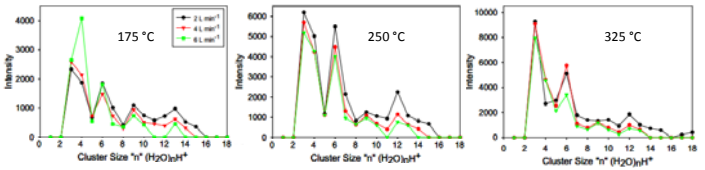

Powder

III-3) Thermal Desorption

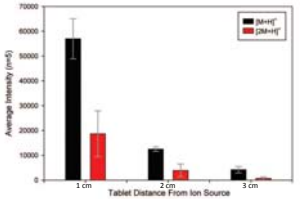
Direct Analysis in Real Time (DART)

- Characterization:

J Am Soc Mass Spectrom 2010, 21, 855– 863



Influence of DART temperature and discharge gas flow rate



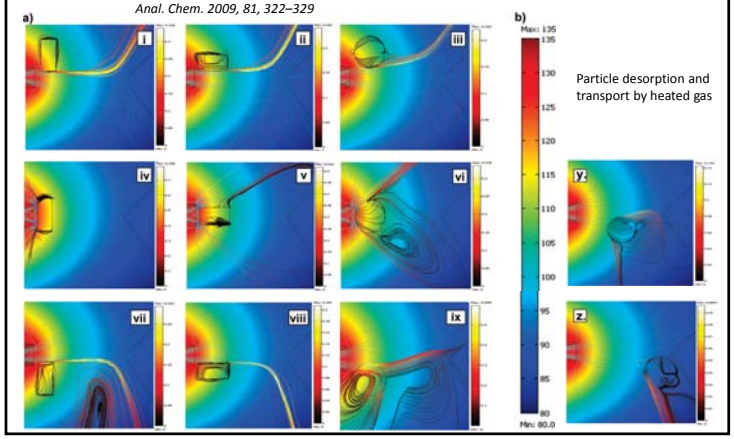
Distance-dependent sensitivity of a Tylenol tablet on both the acetaminophen protonated molecule and dimer

III-3) Thermal Desorption

Direct Analysis in Real Time (DART)

- Characterization:

Anal. Chem. 2009, 81, 322–329



Particle desorption and transport by heated gas

III-3) Thermal Desorption

Direct Analysis in Real Time (DART)

- Mechanisms of ion formation:

i) Penning ionization: $M + R^* \rightarrow M^{+\bullet} + R + e^-$

ii) When He is used, positive ion formation involves formation of ionized water clusters followed by proton transfer reactions:

$$He^*_{(g)} + nH_2O_{(g)} \rightarrow He_{(g)} + (H_2O)_{n-1}H^+_{(g)} + OH^-_{(g)}$$

$$(H_2O)_m H^+_{(g)} + M_{(s/g)} \rightarrow MH^+_{(g)} + nH_2O_{(g)}$$

iii) Charge exchange reactions with oxygen molecular ions:

positive mode $O^{+\bullet}_{2(g)} + M_{(g)} \rightarrow M^{+\bullet}_{(g)} + O_{2(g)}$

negative mode $e^- + O_{2(g)} \rightarrow O^-_{2(g)}$

$O^-_{2(g)} + M_{(g)} \rightarrow M^-_{(g)} + O_{2(g)}$

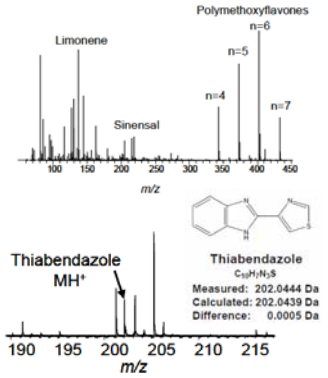
III-3) Thermal Desorption

Direct Analysis in Real Time (DART)

- Applications:

Food Analysis

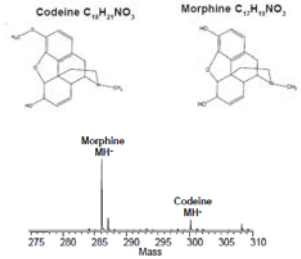
Rapid Commun. Mass Spectrom. 2011, 25, 127–139



DART MS spectra of orange peel

Food Analysis

J Forensic Sci. 2008, 96, 29-38



DART MS spectrum of poppy seed showing traces of opiate

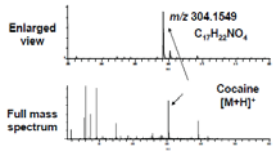
III-3) Thermal Desorption

Direct Analysis in Real Time (DART)

- Applications:

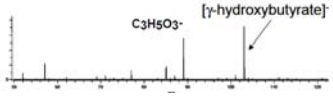
Forensics

J Am Soc Mass Spectrom 2009, 20, 891– 899



DART MS spectrum of 1$ bill

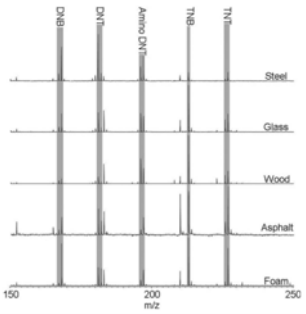
J Forensic Sci. 2009, 54, 370-375



DART(-) MS spectrum of GHB spiked in Ocean Spray Cranberry Juice

Homeland Security

Propellants Explos. Pyrotech. 2010, 35, 446 – 451



DART MS spectra of explosives on different surfaces

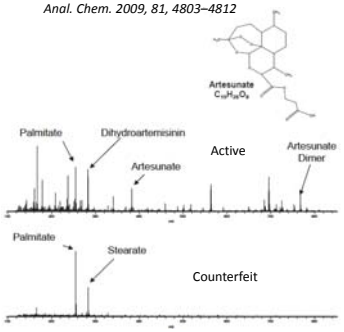
III-3) Thermal Desorption

Direct Analysis in Real Time (DART)

- Applications:

Pharmaceutical

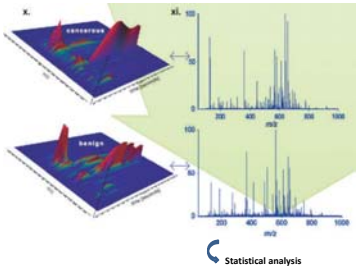
Anal. Chem. 2009, 81, 4803–4812



DART(-) MS spectra of anti-malarial drugs "Guilin B" pills

Bioanalysis

Cancer Epidemiol. Biomarkers Prev. 2010, 19, 2262–2271

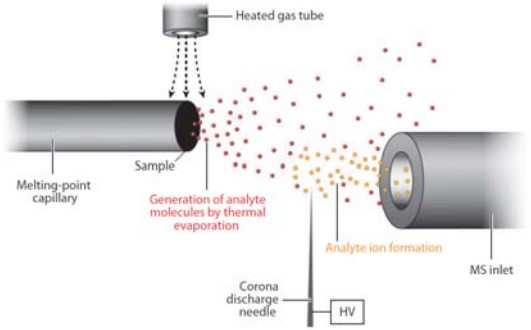


DART MS spectra of serum from women with ovarian cancer or healthy women

III-3) Thermal Desorption

Atmospheric Pressure Solids Analysis Probe (ASAP)

Anal. Chem. 2005, 77, 7826-7831



- Analytes are evaporated from the capillary surface by a heated gas.
- Stream of hot gas directs desorbed analytes to the MS inlet.
- Analytes are ionized by corona discharge by applying HV on a needle.

III-3) Thermal Desorption

Atmospheric Pressure Solids Analysis Probe (ASAP)

- Characteristics:

Classical APCI source used without solvent
Hole to plug the melting-point capillary with sample on it
N_2 gas flow rate : 0.4 L/min
Gas temperature ~ 300-400°C
Corona discharge of ± 3-6 kV

- Mechanisms of ion formation:

* Analytes are put into the gas phase by thermal desorption.
* Corona discharge on the N2 gas stream containing analytes allows ionization of the analytes molecules.
* Processes similar to APCI.

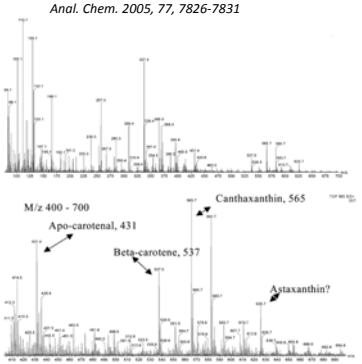
III-3) Thermal Desorption

Atmospheric Pressure Solids Analysis Probe (ASAP)

- Applications:

Food Analysis

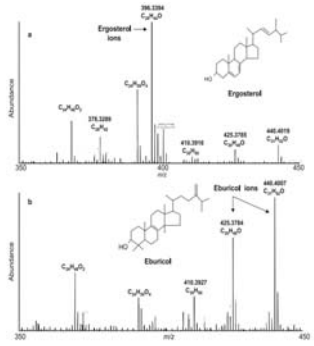
Anal. Chem. 2005, 77, 7826-7831



ASAP MS spectra of fresh spinach leaf

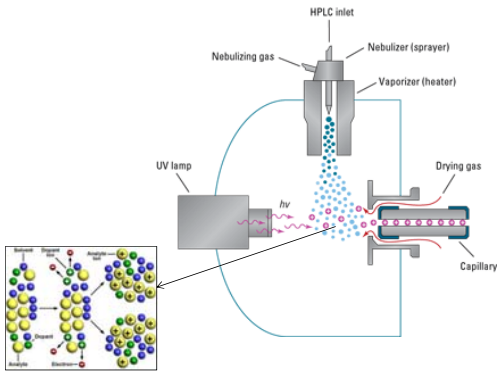
Cell analysis

J Am Soc Mass Spectrom 2007, 18, 1274 –1278



ASAP MS spectra of (a) untreated fungal cells (b) cells treated with the triazole inhibitor of C14-demethylase (4 M) added during growth.

IV- APPI-based ambient ionization techniques

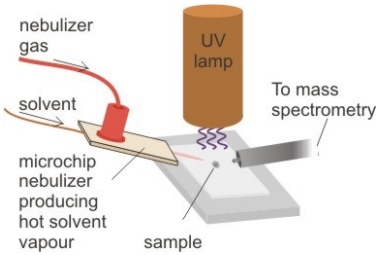


In ambient ionization techniques using this mode of ionization, **UV light** allows the **post-ionization** of the desorbed analytes (APPI processes).
Direct analysis of volatile sample species with a wide range of polarities from surface.

IV- 1) Thermal/chemical desorption

Desorption Atmospheric Pressure PhotoIonization (DAPPI)

Anal. Chem. 2007, 79, 7867-7872



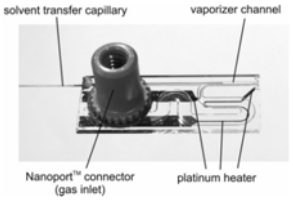
- Heated nebulizer gas evaporates solvent.
- Neutral hot vapor solvent jet impinges on the surface.
- Analytes are extracted from the surface by thermal desorption.
- Gas phase neutral analytes ionize directly by photon absorption or react with ionized solvent species.

IV- 1) Thermal/chemical desorption

Desorption Atmospheric Pressure PhotoIonization (DAPPI)

- Characteristics:

A nebulizer µchip for heated solvent/gas mixture. 50 µm diameter vaporizer channel
Microchip nebulizer is at an angle of ~45° and ~10 mm above the sampling surface



Solvent flow rate : 10 µL/min
Solvent mixture: adapted to the analyte polarity (± dopant : toluene or acetone)
N_2 nebulizer gas temperature ~ 150-300°C, and gas flow rate 180 mL/min
UV krypton lamp (10 eV) placed at 90° and ~10 mm above the sampling surface

IV- 1) Thermal/chemical desorption

Desorption Atmospheric Pressure PhotoIonization (DAPPI)

- Mechanisms of ion formation:

* Analytes are thermally desorbed by the hot solvent vapor jet.
* Photon absorption by the analyte molecule, leading to electron ejection, forming a molecular radical cation $M^{\bullet+}$

$$M + h\nu \rightarrow M^{\bullet+} + e^-$$

* Subsequent reaction of abstraction of a hydrogen atom from the abundant solvent to form the stable $[M+H]^+$ cation.

$$M^{\bullet+} + S \rightarrow [M+H]^+ + [S-H]^{\bullet}$$

* Dopant (D) is first photo-ionized and then $D^{\bullet+}$ ionized analytes via proton or electron transfers.

$$D^{\bullet+} + M \rightarrow [M+H]^+ + [D-H]^{\bullet}$$

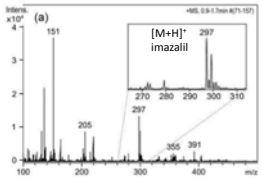
IV- 1) Thermal/chemical desorption

Desorption Atmospheric Pressure PhotoIonization (DAPPI)

- Applications:

Food Analysis

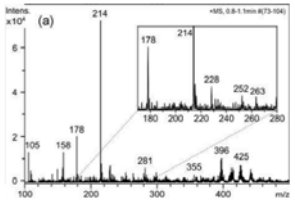
Rapid Commun. Mass Spectrom. 2010; 1343–1350



DAPPI MS spectra of orange peel with acetone dopant

Environmental Analysis

Rapid Commun. Mass Spectrom. 2010; 1343–1350



DAPPI MS spectra of soil spiked with PAHs peel with toluene dopant → $M^{\cdot+}$ species

LOD 100 pg

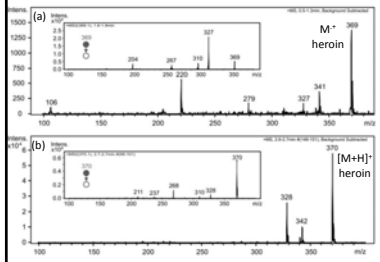
IV- 1) Thermal/chemical desorption

Desorption Atmospheric Pressure PhotoIonization (DAPPI)

- Applications:

Forensics

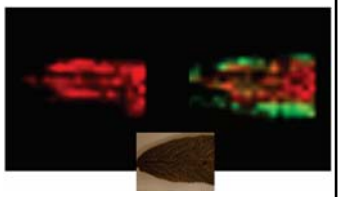
Rapid Commun. Mass Spectrom. 2009; 1401–1404



DAPPI MS spectra of illegal drugs with (a) toluene and (b) acetone dopant

Imaging

Anal. Chem. 2009, 81, 8479–8487



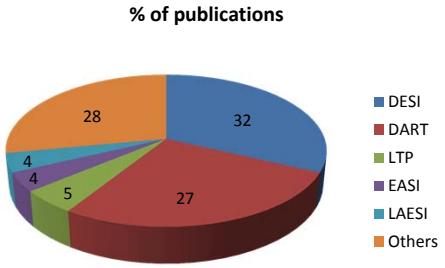
DAPPI MS chemical images of sage leaf

Spatial resolution 1 mm

V- Comparison of techniques

Technique	Surface sampling process	Ionization process	Highest mass	Analyte polarity	Sample state	Detection limit/Sample	Dynamic range
ASAP	Thermal	APCI	700 Da	Polar, non-polar	Solid, liquid	N/A	N/A
DAPCI	Direct	APCI	600 Da	Polar	Solid, liquid	1 ng.cm^{-2}/TATP	10^3
DAPPI	Thermal	APPI	600 Da	Polar, non-polar	Solid	56-670 fmol/Verapamil	N/A
DART	Thermal	APCI	~ 1kDa	Polar, non-polar	Solid, liquid, gas	7 fmol/Ethylpalmitate	10^3
DBDI	Direct	APCI	400 Da	Polar, non-polar	Solid, liquid	3.5 pmol/Alanine	10^2
DCBI	Direct	APCI	600 Da	Polar, non-polar	Solid, liquid	10 pg/Atrazine	10^3
DESI	Direct	ESI	66 kDa	Polar	Solid, liquid	100 pg/PETN	10^5
EADESI	Direct	ESI	24 kDa	Polar	Solid, liquid	10 pmol/Maltoheptaose	N/A
EASI	Direct	ESI	1 kDa	Polar	Solid, liquid	0.01 ppm/Nicotine	10^3
ELDI	Laser	ESI	66 kDa	Polar	Solid, liquid	30 fmol/Cytochrome C	10^4
ESA-Py	Thermal	ESI	1.5 MDa	Polar	Solid, liquid	1 ppm/dimethylated PDA	N/A
FAPA	Direct	APCI	38 kDa	Polar, non-polar	Solid, liquid	60 fmol/Acetophenone	N/A
FD-ESI	Gas stream	ESI	17 kDa	Polar	Liquid, gas	N/A	N/A
IR-LAMICI	Laser	APCI	665 Da	Polar, non-polar	Solid, liquid	30 pg/Acetaminophen	N/A
LAESI	Laser	ESI	66 kDa	Polar	Solid, liquid	8 fmol/Verapamil	10^4
LD-APCI	Laser	APCI	1.5 kDa	Polar, non-polar	Solid, liquid	N/A	N/A
LIAD-ESI	Laser	ESI	66 kDa	Polar	Solid, liquid	N/A	N/A
LTP	Direct	APCI	500 Da	Polar, non-polar	Solid, liquid, gas	500 fg/TNT	10^4
MALDESI	Laser	ESI	20 kDa	Polar	Solid, liquid	13 fmol/Angiotensin I	N/A
ND-EESI	Gas stream	ESI	1 kDa	Polar	liquid, gas	N/A	N/A
TPD-ESI	Thermal	ESI	379 Da	Polar	Solid, liquid	0.1 nmol/TNT	N/A

V- Comparison of techniques



2009-2016

VI- Conclusions

- Ambient ionization MS techniques make use of well-established ionization principles such as ESI, chemical ionization or photo-ionization but in an open air direct ionization format which allows unique experiments to be performed with no or little sample preparation.

- These techniques employ various methods for the sampling of solids, liquids and gas through desorption (thermal evaporation, laser ablation , pneumatic nebulization or direct impinging on the surface with charged and metastable species).

- There is a considerable overlap between various ambient ionization techniques because most of them combine a limited number of sample-introduction, desorption and ionization processes; nomenclature is confusing. Unifying naming may be in place for future work.

- Ambient ionization has already been an important part of modern MS, but much remains to be learned about the fundamental mechanisms.


Résumés

O1 - Effet de la sulfatation du domaine extracellulaire du récepteur CXCR4 sur son interaction avec la chimiokine SDF-1α analysé par électrophorèse capillaire d'affinité couplée à la spectrométrie de masse

Maud Fumex[1], Denis Servent[2], Gilles Mourier[2], Régis Daniel[1], Florence Gonnet[1,*]

1. Université d'Evry Val d'Essonne, CNRS UMR 8587, Laboratoire Analyse et Modélisation pour la Biologie et l'Environnement, 91025 Evry Cedex, France
2. Commissariat à l'Energie Atomique, iBiTecS, Service d'Ingénierie Moléculaire des Protéines, 91191Gif-sur-Yvette.

La sulfatation fait partie des plus de 300 modifications post-traductionnelles répertoriées. A l'instar de la phosphorylation, elle affecte les fonctions de nombreuses biomolécules telles que les glucides et les protéines, mais ses mécanismes d'action sont encore méconnus, alors que 1% des tyrosines des protéines seraient sulfatées.

Les protéines connues pour être sulfatées sont majoritairement des récepteurs cellulaires, parmi lesquels le récepteur CXCR4, l'un des plus étudiés en raison de son implication dans de nombreux phénomènes physiopathologiques. Ce récepteur possède un domaine extracellulaire de 38 acides aminés, comportant trois résidus tyrosine (Y7, Y12 et Y21) connus pour être sulfatés, qui est primordial dans le processus d'interaction avec son ligand spécifique, la chimiokine SDF-1/CXCL12 (*Stromal cell-derived factor-1*).

Afin d'évaluer l'impact de la sulfatation sur les propriétés d'interaction entre le domaine extracellulaire du récepteur CXCR4 et SDF-1, un peptide modèle correspondant aux 38 premiers acides aminés de la partie N-terminale du récepteur, appelé P38, a été synthétisé chimiquement, puis sulfaté sur différentes positions (Y7, Y12 et/ou Y21) et à différents degrés (mono-, di- ou tri-sulfatés) conduisant à une bibliothèque de peptides P38 différemment sulfatés.

L'analyse de l'interaction de ces peptides P38 variablement sulfatés est effectuée par électrophorèse capillaire d'affinité (ACE) couplée ou non à la spectrométrie de masse (MS). L'ACE permet la détermination des constantes de dissociation de ces complexes noncovalents formés librement en solution et sans immobilisation de l'un des partenaires. Le couplage ACE-MS en conditions non dénaturantes permet la caractérisation des complexes formés, à savoir la détermination de leur stoechiométrie. Dans ces conditions, nous observons la formation de complexes entre les peptides P38 différemment sulfatés et SDF-1, dont l'affinité dépend de la position et du nombre des groupements sulfate.

O2 - Nouvelles sources d'ionisation pour le couplage de la chromatographie gazeuse et de la spectrométrie de masse à haute résolution : intérêt pour les molécules liées à la défense

Valentin Baillet

Laboratoire COBRA

La chromatographie en phase gazeuse couplée avec la spectrométrie de masse (GC/MS) reste actuellement une des techniques de référence pour l'analyse des produits chimiques liés à la Convention sur les armes chimiques (CAC). L'ionisation électronique (EI) est la technique d'ionisation la plus utilisée et la plus ancienne. Cependant cette méthode conduit à de nombreuses fragmentations ce qui peut compliquer voir empêcher l'attribution des spectres. Ces dernières années, de nouvelles méthodes d'ionisation dédiées au couplage GC/MS se sont développées sur des instruments commerciaux telles que l'ionisation par champ (FI) et les interfaces à pression atmosphériques APGC. Ces deux modes d'ionisation sont universels et permettent de conduire à des espèces ioniques intactes. De telles sources d'ionisation sont particulièrement pertinentes quand elles sont associées à un analyseur à haute ou très haute résolution. Ces instruments sont intrinsèquement plus sensibles et permettent donc la mise en évidence de composés à l'état de trace. Au cours de ce travail de thèse, nous chercherons à évaluer l'intérêt de ces différentes interfaces associées à la spectrométrie de masse à haute résolution (TOF) et très haute résolution (FT-ICR) pour l'analyse de molécules liées à la défense telles que des impuretés de synthèse d'agents de guerre chimique et leurs composés de dégradation ainsi que des produits simulant leurs propriétés physiques et chimiques. Nous chercherons en particulier à mettre à profit les mesures de masses précises obtenues avec ces instruments pour établir des cartographies moléculaires comme des diagrammes de Kendrick ou de Van Krevelen pour mettre en évidence des composés inconnus ou inattendus dans des matrices complexes tels que les sols ou certains échantillons biologiques comme le sang ou l'urine.

O3 - Analysis of bile acids by LC-MS/MS

Noélie Bossut

Gustave Roussy

The human body is composed of billion bacteria allowing the metabolism of nutrients. Now, research is focused on the understanding of the microbiota for therapeutic interest that goes beyond the nutritional framework. The goal of biologists is to better understand the microbial field in the efficiency of cancer treatments such as chemotherapy. For this, Gustave Roussy's metabolomics platform implements various targeted and non-targeted analytical methods analyzing families of molecules that may be of interest in this study. A method of targeted analysis of bile acids is part of the implemented projects. These products are the final products of cholesterol catabolism. Stored and concentrated in the gallbladder, these metabolites are known to facilitate the digestion and absorption of nutrients in the small intestine. This development of analytical method highlights two main problems: the separation of isotopomers and the elimination of carry over in the chromatographic system. We will also see the advantage of transferring a method on a high-resolution device.

O4 - Tandem Mass Spectrometry to Decipher Messages Encoded in Polyurethanes Used as Molecular Barcodes in Anti-Counterfeiting Technology

Jean-Arthur Amalian,[1] Benoit Eric Petit,[2] Denise Karamessini,[2] Abdelaziz Al Ouahabi,[2] Christophe Chendo,[1] Didier Gigmes,[1] Jean-François Lutz,[2] Laurence Charles[1]

[1] *Aix Marseille Université – CNRS, UMR7273, Institut de Chimie Radicalaire – Marseille, France*
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Inspired by the storage of genetic information in DNA using a molecular code based on four different bases, information can be implemented in monodisperse synthetic polymers using a set of two different co-monomers, arbitrarily defined as the 0-bit and 1-bit of the ASCII code.[1,2] To date, tandem mass spectrometry (MS/MS) is the most efficient methodology to read information contained in such sequence-controlled polymers.[3-5] To do so, fragmentation rules (as well as their robustness) have to be established based on a clear understanding of dissociation reactions, known to highly depend on the chemistry of the polymer backbone.

Here, we report on dissociation mechanisms of sequence-controlled polyurethanes (PUs) in the negative ion mode. In these molecules, the code is contained in small alkyl segments substituted (1-bit) or not (0-bit) by a methyl group and linked to a carbamate function. Because their starting group comprises a carboxylic acid function, sequence-defined PUs were readily produced as deprotonated species when subjected to electrospray ionization (ESI). Their collision-induced dissociation gave rise to very simple MS/MS spectra, with a single fragment series in which the distance between two consecutive peaks is equal to the mass of one or the other co-monomer. In spite of this very simple dissociation pattern which allows the 0/1 sequence to be literally read from the spectrum, fragmentation processes at work were found to be puzzling. By combining breakdown curves established in MS^2, H/D exchange and MS^3 experiments, both competitive and consecutive charge-remote reactions were demonstrated for the cleavage of all carbamate bonds but the first one, which dissociation was evidenced to involve the nearby deprotonated group.

[1] R.K. Roy et al., Design and synthesis of digitally encoded polymers that can be decoded and erased, Nat. Commun. 6 (2015), 7237.
[2] A. Al Ouahabi et al., Synthesis of non-natural sequence encoded polymers using phosphoramidite chemistry, J. Am. Chem. Soc. 16 (2015), 5629-5635.
[3] L. Charles et al., MS/MS sequencing of digitally-encoded poly(alkoxyamine amide)s, Macromolecules 48 (2015), 4319-4328.
[4] L. Charles et al., MS/MS sequencing in the negative ion mode to read binary information encoded in sequence-defined poly(alkoxyamine amide)s, Rapid Commun. Mass Spectrom. 1 (2016), 22-28.
[5] J.-A. Amalian et al., MS/MS digital readout: analysis of binary information encoded in the monomer sequences of poly(triazole amide)s, Anal. Chem. 88 (2016), 3715-3722.

O5 - Lipid profiling of cardiac tissue following a myocardial infarction on a mouse model: MALDI MS Imaging and LC-MSMS Identification

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Myocardial infarction (MI) is one of the most serious challenges of contemporary cardiology. It is the major source of morbidity and mortality worldwide, with over 07 million people suffering infarction each year. Heart muscle damaged during myocardial infarction is replaced by a collagenous scar over a period of several weeks. Recent research provides new insight into the integrative biology of inflammation as it contributes to ischaemic cardiovascular disease; thus large infarct induce a process of cardiovascular remodeling which includes gross morphological, histological and molecular changes of both infarcted and the residual non infarcted myocardium.

The aim of this work is to study a new category of biomarkers involved in cardiac remodeling and determine the localization of lipids in the failing and control heart using mass spectrometric imaging (MSI).
MSI was conducted on cardiac tissue following 24h left coronary ligation to analyze multiple compound classes. The samples were prepared from sections of mouse left ventricle. After matrix deposition, samples were analyzed by MALDI-TOF in both negative (using 9AA matrix) and positive mode (using HCCA matrix) . A multicariate analysis was made using Scils Lab Software. MALDI tissue imaging revealed spatially-resolved lipids signals within heart sections that are specific to structures or regions of the heart, and allows to establish the differences of lipid distribution in the failing and non-failing heart. The lipids present in the area of difference were then analyzed using tandem mass spectrometry (LC MSMS) in order to get an identified lipid mapping.

O6 - Correlation between the Shape of the Ion Mobility Signals and the Stepwise Folding Process of Polylactide Ions

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Ion Mobility Mass Spectrometry (IMMS) is an elegant approach that measures the drift time of ions flying through a cell pressurized with a neutral buffer gas under the influence of an electric field. When analyzing multiply charged polymer ions by IMMS, the most striking feature is the observation of breaking points in the evolution of the average collisional cross sections with the number of monomer units. Those breaking points are associated to stepwise folding process of the polymer chain around the cationizing agents. [1,2]

In the context of the present work, we monitored the shape of the arrival time distribution (ATD) of multiply cationized polylactide ions along the polymer distribution. The observation of broad and asymmetric ATDs has been associated to the coexistence of different stable structures in the gas phase, namely *opened, extended* and *folded* structures. Since these structures appear around the breaking points previously reported, a correlation between the stepwise folding process of polylactide ions and the coexistence of opened and folded structures is then proposed. In order to get information on the possible interconversion between the different observed populations upon ion activation, IM-IM-MS experiments (tandem ion mobility measurements) were performed in collaboration with Dr. Ph Dugourd (University Lyon I). Upon selection of one conformer and collisional activation, the interconversion between the folded to the opened structures has not been observed (and vice versa). This interconversion appears to be very energy-demanding in the gas phase and it seems therefore that the observed conformational heterogeneity originates from the desolvation/ionization Electrospray processes.

[1] De Winter, J. et al, Chem. Eur. J 17 (2011)
[2] Larriba, C.; Fernandez de la Mora, J. J. Phys. Chem. B 116. (2012)

O7 - Développement du couplage chromatographie d'exclusion stérique – spectrométrie de masse native (SEC-native MS)

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L'approche de la spectrométrie de masse native permet de transférer en phase gazeuse des assemblages de macromolécules biologiques sans dissocier les liaisons faibles. En effet, l'activité biologique d'une protéine est fortement liée à sa structure quaternaire et c'est pourquoi l'information structurale est très précieuse.

Les tampons de purification classiquement utilisés sont en général incompatibles avec le mode d'ionisation ESI de par la présence de sels, de détergents et autres molécules non-volatils. Il convient de procéder à un échange de tampon dit "dessalage" afin de solubiliser l'échantillon dans un tampon compatible avec l'ionisation ESI.

Un des verrous de l'analyse par spectrométrie de masse native est cette étape de dessalage qui reste manuelle, délicate et chronophage. Elle présente une limitation importante pour l'automatisation et de la démocratisation de la MS native, notamment dans l'industrie pharmaceutique. Dans ce contexte, le développement d'un couplage SEC-MS native est particulièrement attractif en particulier pour l'automatisation de l'échange de tampon ou la séparation de protéines au sein d'un mélange avant l'analyse en ligne par MS native.

Nous avons ainsi développé un couplage SEC-MS native. Des optimisations basées sur la phase mobile, faisant directement office de tampon d'échange, ont été réalisées ainsi que sur le débit. Pour l'ensemble des protéines ou complexes non-covalents analysés, la qualité du dessalage s'est avérée meilleure en SEC qu'en dessalage manuel. Ainsi, le couplage SEC-MS native permet soit l'automatisation du dessalage en ligne de manière assez rapide (5-10 min) ou une séparation/dessalage des protéines en mélanges en amont de l'analyse par spectrométrie de masse native.

O8 - Monitoring of catalytic deoxygenation of pyrolysis bio-oil by ESI and APPI FT-ICR mass spectrometry

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The bio-oils produced by the pyrolysis of the biomass are very complex mixture involving thousands of species with different chemical properties. Moreover, the significant amount of oxygen contained in this material makes it unsuitable for a direct use as a bio-fuel. To overcome this hurdle, catalytic treatments are developed to decrease the oxygen content and increase the energy density. In order to improve the production and the upgrading processes, the comprehensive description of the bio-oil before and after the catalytic treatment is required by applying the non-targeted petroleomic approach. Therefore, we performed (+/–) ESI and (+) APPI FT-ICR MS analyses on a raw and an upgraded bio-oil.

It is demonstrated that close to 90 % of the signal is attributed to CxHyOz compounds for the raw bio-oil. The distribution of the observed features in respect with the oxygen amounts is very close whatever the used analytical conditions. The compounds contain from 1 to 16 oxygen atoms with O4 to O7 predominant and present a double bound equivalent (DBE) ranging from 2 to 20 depending on the used experimental condition. In (+) ESI, DBE ranges from 2 to 10 whereas in (+) APPI, more unsaturated compounds are identified with DBE values between 5 and 20. Van Krevelen diagrams are used to represent each CxHyOz formula by its O/C and H/C ratio. It enables to evidence some biomass component derivatives such as lipids (O/C < 0.4 and H/C ~ 2) and lignin-linked compounds (0.2 < O/C < 0.8 and H/C ~1), which are common to all measurements. Sugaric derivatives (O/C > 0.6 and H/C ~ 2) are generally observed in (+) ESI and in a lesser extent in (–) ESI. These results demonstrate that ESI is suitable for the analysis of less apolar compounds (low DBE value and important oxygen amounts) whereas APPI is more suited for the detection of more apolar compounds (high DBE and in a lesser extent less oxygenated species).

This methodology was also applied to an upgraded (deoxygenated) bio-oil. In each experiment, close to 95% of the signal is relative to CxHyOz and CxHy compounds. In (+) APPI, the decrease of the oxygen amounts is observed for compounds of treated bio-oil which evidences the efficiency of the deoxygenation process. Furthermore, ESI gives information on the refractory bio-oil components which are still observed in these conditions.

O9 - Action-FRET and works

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The yearning to observe conformational changes in biomolecules in gas phase isolation has fuelled the recent development of transposing Förster Resonance Energy Transfer (FRET), a common solution-phase technique, to mass spectroscopic settings. It has been widely admitted that the technique has led to a useful understanding of molecular proximity relationships. Our lab has made further inroads by setting out a methodology for performing gas-phase FRET experiments that circumvents the need to monitor fluorescence as a reporter of FRET efficiency, instead the latter is converted from the detection of specific photo-fragmentation – a technique coined as action-FRET. Here, we report the unprecedented work of action-FRET measurements on a protein – a cysteine containing mutant of Ubiquitin. We show that FRET efficiency responds in accordance to an increase in charge state and methanol concentration, setting out an emerging picture of the initial distribution of structures in solution and how these structures are charged during electrospray ionization. In a second part, a focus is given on how action-FRET has been used to scrutinise amyloid-beta systems and how it has aided in piloting an understanding on its conformational complexity and evolution. The application of action-FRET to a protein macromolecule represents a pivotal step in this technique becoming a 'gold standard' structural biology tool. So has been our motive to attempt and test perspective of action-FRET working in the negative mode of the coupled MS instrument. These most recent experimental efforts are presented completing the third part of this presentation. We are hopeful that both action-FRET techniques, although still in its infancy, will develop into a new research tool ready to glean insight on molecular machinery within the scope of native mass spectrometry and pave a way to a better understanding of molecular and supramolecular chemical relationships.

O10 - Analysis by MALDI-TOF imaging and LC-MS-MS of the effects of probiotics on bowel inflammation

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Introduction

Nowadays inflammatory bowel diseases are considered as an issue for the global population because the rate of affected people is increasing very fast every year, especially in western countries. Furthermore, this situation is aggravated by the poor understanding of the causes of these troubles and the absence of efficient treatments.

In this work, taking into account the importance of microbiota for the regulation of intestinal tractus, we used a mouse model to find out if there was any protection effect of the intestinal barrier during an inflammatory bowel disease associated to the administration of probiotics.

For this study, 18 mice have been used, divided in 3 groups of 6 mice each (control, sick and sick pretreated with probiotics). After sacrifice, the lipid profiles of their guts were compared using MALDI-ToF imaging.

Methods

18 Albinos mice were divided in three groups ; the first one was the control, the second group was submitted to a treatment causing bowel inflammation and the third one was pretreated by administration of a lactobacillus bacterial strain before inducing the inflammation. Colons and ileum of each group were collected after mice sacrifice, and stored at -80°C before analysis.

Sample preparation for MALDI-ToF imaging consisted in tissue sectioning at 10 micrometer thickness using a cryostat (Leica), before deposition on ITO slides, drying and matrix application using an ImagePrep device (Bruker). The image acquisition was achieved by means of a MALDI-ToF UTRAFLEX III mass sepctrometer (Bruker). Images were treated by FlexImaging (Bruker) and SCiLS-LAB softwares. For the LC-MS-MS analysis the samples submitted to extraction of regions of interest and analyzed by LC-MS/MS in reversed phase mode using a Shimadzu HPLC and a Q-TRAP 5500 mass spectrometer (Sciex).

Preliminary Data

The study was focused on lipids, with the objective of identification of species involved in the inflammation process ; results obtained for the 3 groups were confronted.

The comparison between the bowels of each group of mice showed different levels of phospholipids between control, sick and mice pretreated with probiotics. In particular, higher amounts of these compounds were observed in sick mice compared to control mice. Importantly, a significant reduction of inflammatory reaction intermediates was revealed for mice pretreated with the probiotic strain, confirming the protective role of probiotics against inflammation.

O11 - Caractérisation de polymères énergétiques fluorés par couplage ASAP-Orbitrap™ : exploitation des défauts de masse de Kendrick

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La capacité de caractériser de manière globale, à l'échelle moléculaire, des échantillons présents à l'état de traces (collectés sur frottis, par exemple) est fondamentale dans des domaines allant de l'environnement à l'industrie. La plupart des formulations industrielles sont constituées d'un composé ou plusieurs composés d'intérêt dilués dans une matrice de liants plastifiants et polymériques, d'huiles, de cires, et d'additifs organiques divers. La caractérisation fine de ces substances dites « connexes », peut révéler de précieuses informations portant sur la nature, la source et l'origine géographique des échantillons étudiés, ainsi que sur les voies de synthèse mises en œuvre [1].

Une source ASAP (Atmospheric Solid Analysis Probe) couplée à un spectromètre de masse haute résolution Orbitrap™ a été utilisée pour caractériser des polymères et copolymères fluorés. Afin de simplifier la lecture des spectres de masse obtenus et d'augmenter la confiance dans la bonne attribution des compositions des pics, les données ont été représentées sous forme de cartes de Kendrick, permettant ainsi de définir une empreinte spectrale spécifique à chaque polymère qui amènera à une identification non-ambiguë.

[1] Mahoney C. M., Fahey A. J., Steffens K. L., Benner B. A., Lareau R. T., Anal. Chem. 82, 7237-7248 (2010).

O12 - Temporally & Spatially-Resolved Microproteomics Analysis for Traumatic Brain Injury Biomarkers Identification

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Traumatic brain injury (TBI) is a major cause of mortality and morbidity affecting nearly 2 million persons, with approximately 500,000 hospitalizations annually. The major cause of TBI is the direct mechanical damage to the brain tissue leading to an axonal disruption, along with a wide spread neural dysfunction. TBI is characterized by a primary acute phase lasting from few hours up to few days, followed by a secondary phase due to the activation of the immune system and recruitment of neuro-inflammatory cells. Several studies have been conducted to assess potential TBI biomarkers such as GFAP and αII-spectrin. However, these studies have assessed these markers in surrounding brain tissues as well as in the biofluids (serum and CSF). In this work, we investigated TBI impact at the injury site, as well as the surrounding tissues, in order to connect TBI microenvironment dynamics along with the underlying biological processes associated with the location and proximity of TBI lesion. This work is achieved using an experimental TBI rat model of mild/moderate Controlled Cortical Impact (CCI) injury by combining large scale proteomics identification and relative quantification using Spatially-Resolved Microproteomics coupled to MALDI MS Imaging. Direct on-tissue micro-digestion followed by micoextraction from 0.25-1 mm2 surface area are then subjected to an LC-MS & MS/MS analysis using HR MS. Data will involve protein regulation dynamics specific to the lesion area, or to the close environment, coupled with the related biological networks. Our analysis is focused on studying and identifying the different proteins and pathways involved in a spatial and temporal manner. Spatially by identifying unique markers prior and post TBI injury in a coronal view of the brain as well as the injury site itself, and temporally by better understanding the acute phase through focusing at the following time points post injury: 1, 3, 7, and 10 days. This will help in the identifying TBI markers as well as determining the time points which could be targeted for therapy.

O13 - Inflammatory bowel diseases and the contribution of OcSILAC strategies to the quantification of the oxidative and inflammatory effects of lipopolysaccharide on human colon cells' proteome

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Crohn's disease and Ulcerative colitis are two notable affections among inflammatory bowel diseases (IBDs). Their rapidly increasing incidence rate, hampering symptoms and severe complications make IBDs primary public health concern, but are very challenging to apprehend due to their multicausal nature. Oxidative homeostasis is a precisely regulated process in every cell and a disruption in this equilibrium leading to oxidative stress had been identified as a key factor for developing IBDs. Specifically a deficit in mitochondrial Mn-superoxide dismutase (Mn-SOD) has regularly been observed in patients' cells.

Manganese peptidomimetic complex *Mn1* was screened for its activity analogous to native Mn-SOD, making *Mn1* a possible candidate as anti-IBD therapeutic molecule. To study the complex and its effects on proteins, a bottom-up proteomics approach was carried out for its high sensitivity and the expression and cysteine-oxidation quantification protocol OcSILAC.

We measured the expression level of mitochondrial Mn-SOD and found that it was increased under the effect of bacterial lipopolysaccharide in our model cell culture, but that the cytosolic Cu/Zn-SOD expression was not affected. This suggests that the modified cell line used is a fitting model to oxidative stress response in colorectal epithelium and IBDs in which this expression profile was observed. We were also able to quantify global change in cysteine oxidative-state occurring under the effect of stress-inducing LPS.

The main goal of this project was to quantify the effect of the drug-candidate *Mn1* on the proteome of LPS-stressed cells at two distinct levels. We first assessed the evolution of the expression of redox-involved proteins in these conditions, and then at the individual cysteine level we measured the induced shift of their oxidation state.

O14 - Study of changes in the kidney's lipid profile in the case of obesity using MALDI-ToF imaging and ESI LC-MS-MS analysis

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Introduction

Obesity is a social and public health problem that is in the origin of several diseases touching different organs.

In this study, we analyzed modifications of the kidney occurring when mice fed with a fat diet become overweighted or obese. These modifications were related to lipids involved in the metabolic reactions e.g.inflammation.

For a better understanding of the link between obesity and kidney disease, we established a kinetic study of obesity in mice and compared healthy and obese mouse organs.e also oberved the effects of

Methods

For MALDI-ToF imaging, mouse kidneys, previously stored at -80 °C, were cut in slices (10 micrometers thickness) using a Cryostat (Leica) and slices were dried on ITO slides. Matrix (alpha-cyano-hydroxy-cinnamic acid) was deposited on the slides by means of an Imageprep automated device (Bruker). The mass spectrometer was an Ultraflex III (Bruker) ; Fleximaging (Bruker) and SCILS softwares were used for image analysis.

Samples for ESI LC-MS/MS were extracted from regions of interest revealed by the MALDI-ToF imaging appoach. These sample extracts were then analyzed by LC-MS/MS (C18 column, Shimadzu chromatograph; QTrap 5500 from Sciex) in full scan MS/MS and MRM modes. Results were analysed by means of LipidView and LipidQuant softwares (Sciex).

Preliminary Data

The kinetic study was split in 3 stages. The first one (days 1-3), did not lead to any significative difference in the spatial distribution and composition of phospholipids between control and obese mice.

The second stage was up to week 16. At that time, mouse fed with a fat diet were obese and differences in distribution and relative concentration of phospholipids in the kidney were clearly observed by MALDI-ToF imaging and confirmed by LC-MS/MS analysis.

For the last stage, after week 16, mice were submitted to a restricted diet, provoking a weight loss.

We observed then that previous modifications in both spatial distribution and relative concentration of phospholipids in obese mice were only partly restored. This observation is used to help establishing a link between obesity and occurrence of kidney disease.

O15 - Caractérisation du site cible anti-métastases de deux protéines par des techniques originales de spectrométrie de masse

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Le cancer est une maladie qui ne cesse de s'étendre au cours de ces dernières années. La physiopathologie cancéreuse n'est actuellement que partiellement élucidée, notamment la transition d'un foyer localisé (tumeur primaire) vers le stade avancé des métastases. Or, il est nécessaire de garder à l'esprit que la principale cause de mortalité lié au cancer n'est pas la tumeur primaire mais bien les métastases, pour lesquelles il n'existe aucune thérapie efficace à ce jour. Notre projet consiste à fournir les bases moléculaires de traitements futurs des métastases. Le projet de thèse se focalise sur 2 cibles cellulaires reconnues comme des éléments impliqués dans le phénomène de métastases et dont les propriétés naturelles inhibitrices (protéine hPEBP1) ou promotrices de métastases (canaux ioniques SK) constituent une opportunité. Le but de ce travail est de localiser des ligands actifs sur la protéine cible à travers des méthodologies différentes. Dans le cas de hPEBP1, nous appliquerons une démarche en 3 étapes : identification par approche qualitative, identification par approche indirecte SSPaQ (Gabant et al., JASMS 2016) et développement d'une nouvelle méthode de criblage rapide de complexe protéine/ligand en LC-MS/MS avec fragmentation ETD. Dans le cas du canal SK, tétramère activé par l'entrée de calcium dans la cellule, la localisation de ligands lipidiques inhibiteurs sur le canal sera effectuée par une méthode de footprinting (protéolyse ménagée). Ainsi ces travaux ouvrent la porte à la conception raisonnée de ligands sur la base de la structure du site actif, apportant une amélioration significative du champ thérapeutique actuel.

O16 - Caractérisation d'assemblages anioniques de polyoxométallates par spectrométrie de masse couplée à la mobilité ionique

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Les polyoxométallates (POM) sont une classe remarquable d'oxoclusters nanoscopiques avec une diversité inégalée de structures et de propriétés. Ils sont construits à partir de la connexion de polyèdres [MOx], M étant un élément du bloc d dans un état d'oxydation élevé, tel que MoVI ou WVI. Ils forment ainsi des structures compactes et fortement chargées. Ces POM peuvent être fonctionnalisés avec des molécules organiques pour donner des POM hybrides organique / inorganique. La spectrométrie de mobilité ionique couplée à la spectrométrie de masse (IMMS) est une méthode intéressante pour la caractérisation de ces dérivés de POM,1 car elle permet de résoudre les problèmes de superposition des ions de même rapport m/z provenant d'espèces multimériques.
Huit isopolyoxométallates sous forme de sels de tetrabutylammonium (TBA) ont été analysés par ionisation electrospray sur un tube de dérive couplée à la spectrométrie de masse (DTIMS). Ces analyses ont permis de déterminer à partir des temps de dérive, les sections efficaces de collision (CCS) dans l'azote de quinze ions avec ou sans contre-ions TBA.

Ces valeurs de CCS ont été utilisées comme référence pour l'étalonnage d'un SYNAPT G2 (Waters), doté d'une cellule de mobilité ionique de type TWIMS, selon la méthode de Smith et al.2 Cet étalonnage nous a permis de mesurer les CCS expérimentales d'un POM hybride similaire à celui d'une étude récente,3 et pouvant former un auto-assemblage de différentes stœchiométries en solution via ses fonctions pyridines et en présence de sels de palladium.

Parallèlement, nous avons optimisé des structures de POM avec et sans TBA par calculs de la théorie de la fonctionnelle de la densité (DFT). Les CCS théoriques de ces structures ont été calculées via une version modifiée du logiciel MOBCAL4, en utilisant des paramètres de Lennard-Jones adaptés du champ de force universel (UFF) pour les atomes Mo et W. Ces calculs de CCS théoriques ont ainsi pu fournir une comparaison avec les résultats obtenus expérimentalement sur les POM utilisés pour la calibration.

1.Robbins, P. J., Surman, A. J., Thiel, J., Long, D.-L. & Cronin, L. Use of ion-mobility mass spectrometry (IMS-MS) to map polyoxometalate Keplerate clusters and their supramolecular assemblies. Chem. Commun. 49, 1909 (2013).
2.Smith, D. et al. Deciphering drift time measurements from travelling wave ion mobility spectrometry-mass spectrometry studies. Eur. J. Mass Spectrom. 15, 113 (2009).
3.Izzet, G. et al. Metal-Directed Self-Assembly of a Polyoxometalate-Based Molecular Triangle: Using Powerful Analytical Tools to Probe the Chemical Structure of Complex Supramolecular Assemblies. Chem. - Eur. J. 21, 19010–19015 (2015).
4.Mesleh, M. F., Hunter, J. M., Shvartsburg, A. A., Schatz, G. C. & Jarrold, M. F. Structural information from ion mobility measurements: effects of the long-range potential. J. Phys. Chem. 100, 16082–16086 (1996).

O17 - Contribution of peptidomics to the study of *Streptococcus thermophilus* peptide utilization in complex media

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Peptidomics is an emerging and promising field derived from proteomics and allowing the comprehensive analysis of endogenous peptides in biological samples. It appeared in the early 2000s and has been up to now primarily used for the discovery of disease biomarkers, hormones, toxins, or for the characterization of food proteolysis dynamics. Here, we present a novel and innovate use of peptidomics in the context of the growth of the lactic acid bacterium *Streptococcus thermophilus* in complex media where amino nitrogen is solely supplied by cellular hydrolysates (CH). Those hydrolysates represent a rich source of peptides, as well as other nutrients, and are routinely used for the massive production of dairy starters. However, to date, not much is known about their peptide fraction. The objectives of the present work were 1) to characterize and compare the peptide fraction of two different CH based media, and 2) to identify dynamics of peptide consumption throughout the culture of *S. thermophilus* in bioreactor scaled fermentations. To do so, a specific analytical pipeline was developed including peptide isolation by solid phase extraction, two RP-HPLC separation steps, MS/MS detection on an ESI-LTQ-Orbitrap mass spectrometer, and peptide identification by the X!TandemPipeline software. Through this analysis process, we were able to map the peptide content of the two CHs. They showed significant differences regarding the richness, diversity and biochemical properties of their respective individual peptides. With the help of an agglomerative hierarchical clustering strategy, we also showed that only specific classes of peptides were consumed throughout the growth of *S. thermophilus*. The biochemical properties of the consumed peptides matched the peptide transporter specificities of the strain.

O18 - Caractérisation d'anticorps immunoconjugués site spécifiques par spectrométrie de masse couplée à la mobilité ionique

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Les anticorps monoclonaux immunoconjugués (Antibody Drug Conjugates, ADCs) constituent une nouvelle génération de protéines thérapeutiques pour traiter de nombreuses maladies, dont les cancers. Ce sont des molécules tripartites constituées d'un agent cytotoxique (drogue) lié de manière covalente à un anticorps monoclonal (mAb) via un linker.

Les chimies de couplage les plus classiquement utilisées ciblent les lysines ou cystéines, engendrant ainsi une hétérogénéité de greffage au niveau de l'ADC. Afin de mieux contrôler l'homogénéité des sites de fixation de la drogue sur le mAb, de nouvelles stratégies de couplage chimique pour cibler spécifiquement certaines positions de l'anticorps et maîtriser le nombre de molécules cytotoxiques greffées ont été développées. Ces nouvelles chimies de couplage ont récemment permis de faire émerger une troisième génération d'anticorps appelée site specific. Dans cette présentation, nous montrons l'intérêt de la mobilité ionique couplée à la spectrométrie de masse native (IM-MS) pour la caractérisation d'un anticorps de troisième génération site-spécifique. Dans un premier temps, l'intégrité de l'ADC ainsi que le profil de greffage et le nombre de moyen de drogues greffées ont été déterminés par MS native, permettant ainsi d'évaluer l'hétérogénéité de greffage et de valider l'efficacité de la nouvelle chimie de couplage utilisée. D'autre part, les expériences de mobilité ionique ont permis de mesurer les sections efficaces de collision (CCS) du mAb non conjugué et de son immunoconjugué associé. Pour la première fois, des expériences Collision Induced Unfolding ont été réalisées sur un ADC et mettent en évidence des profils de déploiement (unfolding) différents pour le mAb et l'ADC. L'ensemble des résultats obtenus est comparé aux précédentes études réalisées sur deux anticorps immunoconjugués de référence à lysine (T-DM1) et à cystéine (Brentuximab Vedotin). Cette étude démontre l'intérêt des approches de spectrométrie de masse en conditions natives pour la caractérisation de protéines thérapeutiques de dernière génération.

O19 - Étude de la phase gazeuse et particulaire de la fumée de cigarette par spectrométrie de masse

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La fumée de cigarette est un aérosol (particulaire et gazeux), issu de la combustion et de la pyrolyse du tabac et du papier d'une cigarette. Certains composés présents dans cet aérosol sont toxiques.

La composition de cet aérosol est très dépendante de la nature de la fumée considérée (fumées inhalées, exhalées,…).

L'objectif de mon travail est d'abord, la mise en place et la validation d'une méthode de prélèvement de particules émises lors du fumage d'une cigarette.

Une machine à fumer a été conçue pour permettre le prélèvement des particules et les composés organiques volatils (COV) du courant principal de la fumée de cigarette. Les particules sont récupérées sur un filtre de quartz alors que les COV sont piégés sur du charbon actif. Après le prélèvement, les COV sont extraits puis analysés par GC/MS et les particules du filtre sont analysées par LDI-FTICRMS. Les cigarettes d'une même marque mais vendues dans trois pays différents (Égypte, Italie et France) ont été étudiées.

L'étude par GC/MS a permis la quantification de certains COV en particulier, les BTEX (Benzène, Toluène, Ethyl-benzène et Xylènes). Les limites de détection et de quantification sont comprises entre 0,06 et 0,25 µg/cig et entre 0,13 et 0,85 µg/cig, respectivement. Les variations observées, de l'ordre 20%, relatives aux étapes de fumage, de collecte, de prétraitement sont jugées satisfaisantes pour qualifier la méthode. Des différences significatives ont alors été observées dans la composition gazeuse des fumées issues des trois types de cigarettes.

L'analyse LDI-FTICRMS de la phase particulaire a été réalisée à 355nm pour les cigarettes italiennes et françaises. De nombreux composés CxHyOzNo sont détectés. Les principaux composés détectés sous la forme d'ions protonés (ou déprotonés) ou d'ions radicalaires sont de type C11-30H7-28N1-4. Les espèces C13-23H6-28N1-2 O1-3 sont observées dans une moindre mesure.

Le mode de détection négatif renseigne sur un plus grand nombre de composés. Ils sont plus insaturés (DBE ~ 15) que ceux identifier en mode de détection positif (DBE ~ 11,4). Bien qu'un nombre significatif de composés soient communs aux types de cigarettes, des différences apparaissent nettement.

O20 - SpiderMass, a high potential tool using in-vivo and real-time mass spectrometry for the guided surgery of cancer

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The uptake of cancerous patients depends on several criteria among which the earliness of diagnosis, the chemotherapy efficiency and when it is needed, the complete tumour removal during surgery. To date, the surgeon principally uses macroscopical criteria like interventional MRI or fluorescent dye during the operation to know whether tumoral tissues remains. Even if he can be helped by the per-operatory analysis of the resection margins by pathologists, this procedure takes 30 to 45min while the patient is still in the operation room. In these conditions, there is a clear need to obtain a precise and fast diagnosis directly in the surgical theatre and allowing in-vivo and real time analysis of the tumour margins and the local metastasis sites. Cancerous cells show particular metabolic features that induce significant changes in the classes of biomolecules that they express and it is now an evidence that molecular signatures, generated from tissues by mass spectrometry, can be correlated with pathological stage. Since a decade, mass spectrometry has been widely used in clinical application to get untargeted molecular information close to the tumour environment, that's why we wanted to use an MS-based instrument which aims to obtain tissue characterization inside the operating room in real-time conditions with low invasiveness. This instrument, called SpiderMass, is based on surface tissue sampling by laser ablation using a laser tuned in the IR wavelength which targets the stretching OH bonds of H2O molecules. To perform real-time analysis, the ablated material is aspirated through a tubing connected to the inlet of the mass spectrometer. A software compare the obtained molecular profiles to a databank with a previously-build classification model. In this project we focus on ovarian cancer as the laboratory works since several years with gynaecological surgeons. These cancers are often diagnosed at late stage so the complete removal of tumoral tissue is critical to avoid metastasis. With a cohort containing 25 high grade serous carcinoma and 25 normal samples, our first results demonstrate the ability of the system to discriminate tissues between these two classes and to use the obtained profiles for databanks building. We are also working in collaboration with veterinarians on dog sarcomas for the future preclinical studies. This class of cancer is really complicated to cure for surgeon as it often infiltrates surrounding tissues. With the SpiderMass, we built a first model with 36 pet patients and are currently using it for margin analysis. These preliminary data demonstrate the potential of the future instrument we are developing for in vivo real-time MS analysis. SpiderMass is a new invasiveness and painless technology that will find various applications such as clinical surgery, dermatology or microbiology.

O21 - Heme-binding as biodereplication method for search antimalarial drugs in complex extracts mixtures: active compounds from *Piper coruscans* (Miq.) C.D. (Piperaceae)

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Malaria remains the most important global public health problem, with an estimated 214 million cases and 438 000 deaths in 2015, principally among children under 5[1]. Antimalarial drugs resistance is permanently rising and calls for the research of new active molecules against *Plasmodium*. The principal antiplasmodial drugs mechanism consists in interrupting heme crystallization in parasite erythrocytic phase, thus perturbating his waste-sorting strategy[2]. With such a target-based approach, we developed a method which identifies heme-binding molecule in a complex extract (*m/z* for heme = 616) by mass spectroscopy (ESI-Q-TOF instrument). This *in vitro* miniaturized biodereplication is based on a medium mimicking the parasite digestive vacuole (pH= 4.8)[3].

Standardization of the method was performed with the identification of adducts (M + heme) for principal antimalarial drugs, synthetic compounds and natural methoxyflavones and showed reproducible results. Our screening method was applied on an ethanolic extract of *Piper coruscans*, used in the Peruvian traditional medicine as antimalarial (IC50 of extract on *P. falciparum* 3D7 chloroquine-sensible strain = 1.36 ± 0.06 µg/mL). Potentially active compounds were identified by *m/z* of adducts [two compounds with *m/z* = 298, one compound with *m/z* = 284] and visualized using molecular networking[4]. Subsequent fractionation was performed by several chromatography methods (centrifugal partition, flash and preparative chromatography) to isolate the target natural compounds. Structural identification was performed by 600 MHz NMR (COSY, HSQC and HMBC). Three molecules were isolated, giving an example of biodereplication method for the fast targeting of active compounds against malaria.

1. WHO. World Malaria Report 2015. 2015.

2. Tekwani BL, Walker LA. Targeting the hemozoin synthesis pathway for new antimalarial drug discovery: technologies for in vitro beta-hematin formation assay. Comb Chem High Throughput Screen. 2005, 8(1):63–79.

3. Wunderlich J, Rohrbach P, Dalton JP. The malaria digestive vacuole. Front Biosci Sch Ed. 2012, 4:1424–48.

4. Yang JY, Sanchez LM, Rath CM, Liu X, Boudreau PD, Bruns N, et al. Molecular Networking as a Dereplication Strategy. J Nat Prod. 2013, 76(9):1686–99.

O22 - Tracking Biomarkers in Cerebral Malaria by Mass Spectrometry Imaging and Spatially-Resolved Microproteomics

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438,000 cases of death by malaria were reported in 2015 [1] in large majority due to Cerebral malaria (CM) induced by Plasmodium falciparum infection. Death is in part due to the sequestration of parasite infected erythrocytes within brain microvessels that lead to the rupture of the blood-brain barrier and a strong neuroinflammatory process. However, the molecular modifications associated with CM are not well understood and represents a lack for both prognosis as well as therapies. In order to get information about molecular pathways associated to the neurological syndrome, we have studied biopsies of brain of patients who died from CM using a combined approach of MALDI MS Imaging and Spatially-Resolved Microproteomics.

The study was realized on fresh tissues of brain cortex from autopsies of 2 CM patients and 3 controls. All samples were cut in 7 serial sections of 12 µm thickness which were subsequently mounted on ITO glass slides. These sections were submitted to MALDI MS Imaging (MALDI MSI) for the study of lipids and proteins distribution and the identification of Region of Interest (ROI) of specific molecular content between controls and malaria samples. Selected ROIs were used as targeted area for micro-digestion assisted by micro-spotting and micro-extraction by liquid extraction surface analysis (LESA) [2]. The extracts were further analyzed using HR LC-MS & MS/MS to allow for large scale identification and relative quantification of the proteins associated to the ROIs. Diagnosis was confirmed by histological staining on a consecutive tissue section.

Statistical analysis of MSI data allow us to identify candidate zones and guided micro-extraction on gray and white matter in order to characterize specifics metabolites distinguishing CM samples. Significant differences were observed in the proteomic profiles between CM samples and controls. These preliminary data obtained from a small number of biopsies highlight the interest of combined MSI-Microproteomics strategies to identify biomarker of CM and for better understanding of the pathological process associated to CM.

[1] MONDIALE DE LA SANTÉ, Organisation. Rapport sur le paludisme dans le monde 2015
[2] Wisztorski, M., Desmons, A., Quanico, J., Fatou, B., Gimeno, J. P., Franck, J., … Fournier, I. (2016). Spatially-resolved protein surface microsampling from tissue sections using liquid extraction surface analysis. Proteomics, 16(11–12), 1622–1632. http://doi.org/10.1002/pmic.201500508