



Untargeted metabolomic approach by GC-QTOF : From low to high resolution

Carole Migné, Nils Paulhe, Yann Guittot, Franck Giacomoni, Mélanie Pétéra,
Stéphanie Durand, Estelle Pujos-Guillot

► To cite this version:

Carole Migné, Nils Paulhe, Yann Guittot, Franck Giacomoni, Mélanie Pétéra, et al.. Untargeted metabolomic approach by GC-QTOF : From low to high resolution. SMMPA 2017 (Spectrométrie de Masse, Métabolomique et Analyse Protéomique), Oct 2017, Marne-La-Vallée, France. , pp.476, 2017, SMMPA 2017-Livre des résumés. hal-01674479

HAL Id: hal-01674479

<https://hal.science/hal-01674479>

Submitted on 2 Jan 2018

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

UNTARGETED METABOLOMIC APPROACH BY GC-QTOF :

FROM LOW TO HIGH RESOLUTION

Carole Migné^a, Nils Paulhe^a, Yann Guittot^b, Franck Giacomoni^a, Mélanie Peter^a, Stéphanie Durand^a, Estelle Pujos-Guillot^a

^a Université Clermont Auvergne, INRA, UNH, Plateforme d'Exploration du Métabolisme, MetaboHUB Clermont, F-63000 Clermont-Ferrand, France
^b LUNAM Université, Oniris, Laboratoire d'Etude des Résidus et Contaminants dans les Aliments (LABERCA), Nantes, F-44307, France

Untargeted metabolomic approach aims to gather information on as many metabolites as possible in biological systems by taking into account all informations present in the data sets. This approach is essential to pinpoint modifications of metabolic pathways associated to nutritional, health or environmental status for identifying biomarkers.

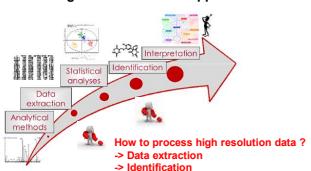
GC is often coupled to single quadrupole analyzers, which offer high sensitivity, good dynamic range but operate with slower scan rates and lower resolution compared to Time of Flight (ToF) systems. GC coupled with ToF Mass Spectrometry is increasingly used for metabolic profiling because of fast acquisition rates, particularly useful for an accurate deconvolution of overlapping peaks obtained from complex mixtures.



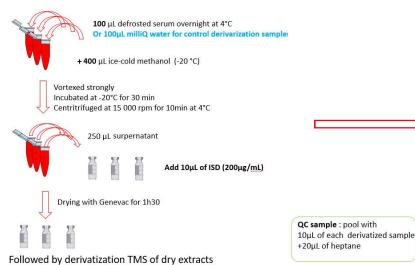
	Low Resolution	High Resolution
Sensitivity	-	+
Resolution	-	+
Acquisition Rate	+	++
Databases	NIST, GOLM, FIEHN and in-house databases	MassBank and in-house databases



Untargeted metabolomic approach

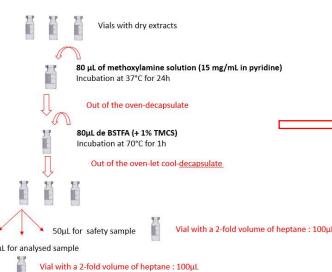


Sample preparation*



* Adapted from Gao X. et al. Anal Chem 2010

Trimethylsilylation Derivatization*



GC Conditions :

Injection system: 2 µL split/20, T = 250 °C.
Column: HPS-MS (30 m x 0.25 mm x 0.25 µm)
Carrier gas: He 5.1, mL / min constant debit.

GCMS Analysis*

Run	Time /min	Value %/full	Initial Time /min	Final Time /min
Kamp_1	20	145	0	2
Kamp_2	20	140	0	30
Kamp_3	20	160	0	30

Total run time : 49 min

MS Conditions: EI ion source temperature : 230°C, full scan mode (m/z 50-800)

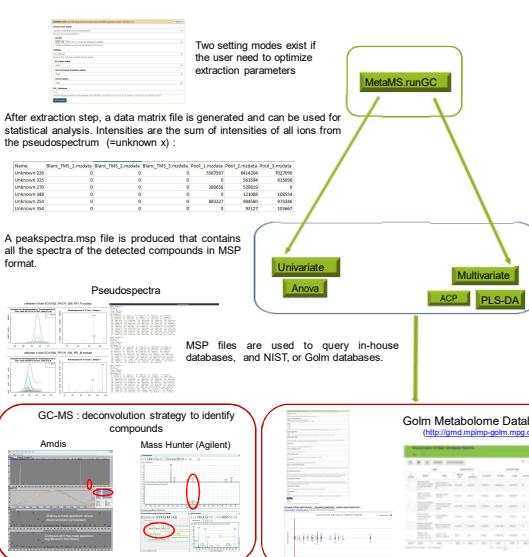
Quad parameters
EI ion source temperature: 230°C
Quadrupole Temperature: 150°C
Electron energy: 70eV

ToF parameters
Acquisition 2GHzEDR with N2 (1.5mL/min)
Aqc rate: 5 spectra/s
Aqc time: 200ms/spectrum
Transients by spectrum: 2712
Limits for average PPM error: 3.0 and maximum error: 8.0

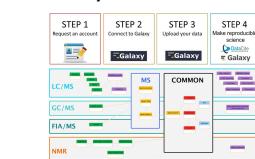
Bioinformatic tools : Available workflow to process GS-MS data

Raw data files are processed and converted to mzdata format using the Agilent MassHunter software.

Data are processed under the Galaxy web-based platform Worflow4metabolomics, using MetaMS or XCMS packages :

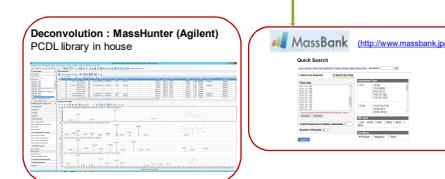
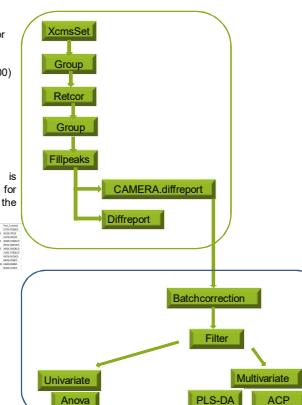


Workflows : Workflow4Metabolomics 3.0 Galaxy online infrastructure



Extraction method for peak detection :
Matched filter (R= 6000)

Data matrix file is generated; it represents for each ion, the value of the intensity in each sample

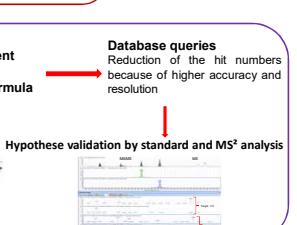
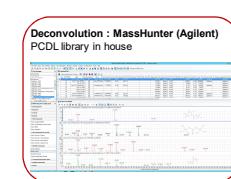


Preprocessing

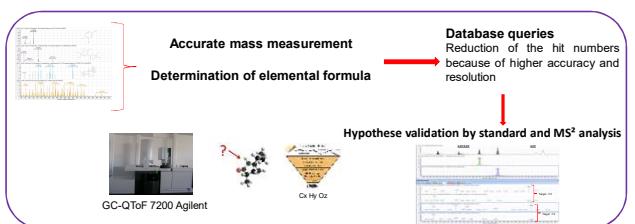
Statistics

Annotations

Structural identification



Complementary analysis in low resolution are necessary to elucidate chemical structure of unknown compounds :



Development of new tools

Data extraction :

To process high resolution data using MetaMS package it is necessary to optimize extraction parameters. Therefore, a tool was developed in the lab to realize this step. Several parameters can be considered (e.g.: FWHM, similarity, threshold,...) one by one or in combination.

Identification

Reference mass spectra are acquired in high resolution in order to implement Peak Forest database.

NIST plasma samples are also analyzed by GC-QToF (MS, MS²) to annotate reference compounds in the matrix.



Conclusion and perspectives

Annotated spectra of reference compounds and biological matrices will be implemented in the metaboHUB national reference database (Peakforest). Accurate mass measurements, as well as the development of new tools for data processing should allow solving the actual bottleneck in biomarker discovery, concerning the identification of metabolites.