# Omics for clinicians

**Christophe Junot**

**Université Paris-Saclay, CEA, INRAE, MetaboHUB**
**Department of Medicine and Healthcare Technologies**
**christophe.junot@cea.fr**

# A personal view on systems medicine and the emergence of proactive P4 medicine: predictive, preventive, personalized and participatory

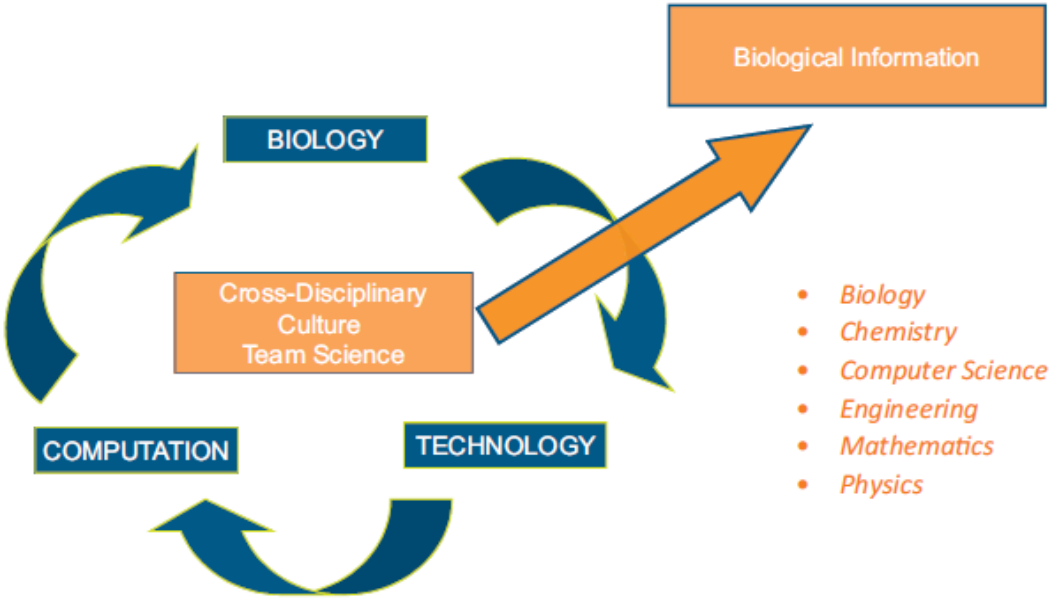
New Biotechnology • Volume 29, Number 6 • September 2012

**Leroy Hood[1], and Mauricio Flores[2]**

[1] Institute for Systems Biology, 401N. Terry Ave, Seattle, WA 98121, USA
[2] P4 Medicine Institute, 401N. Terry Ave, Seattle, WA 98121, USA

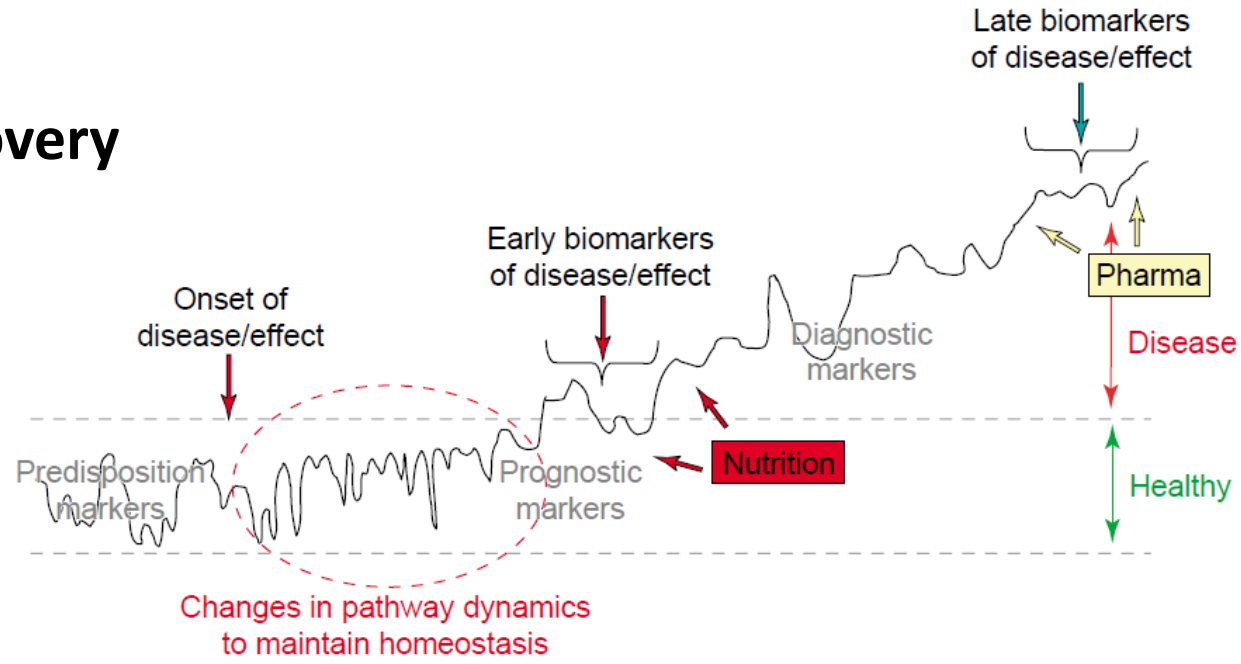

**FIGURE 1**

In 10 years a virtual cloud of billions of data points will surround each patient. These data will be of many different types and, accordingly, multistage. The challenge will be to convert these data into simple hypotheses about health and disease for the individual.



# Why measuring proteins and metabolites?

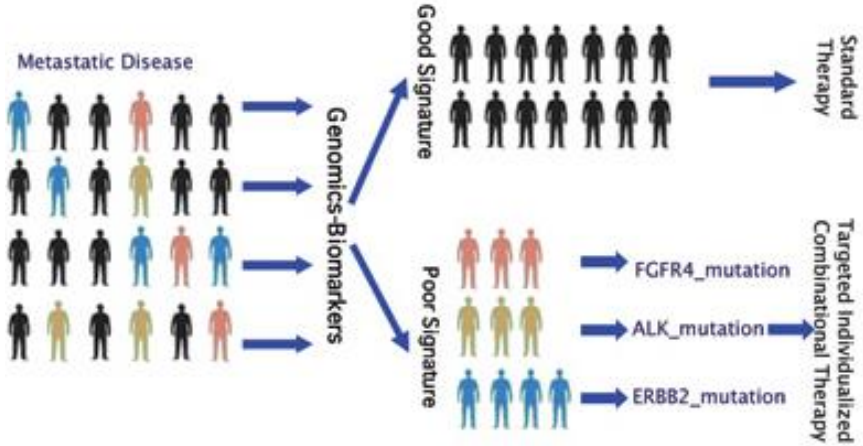
## Biomarker discovery



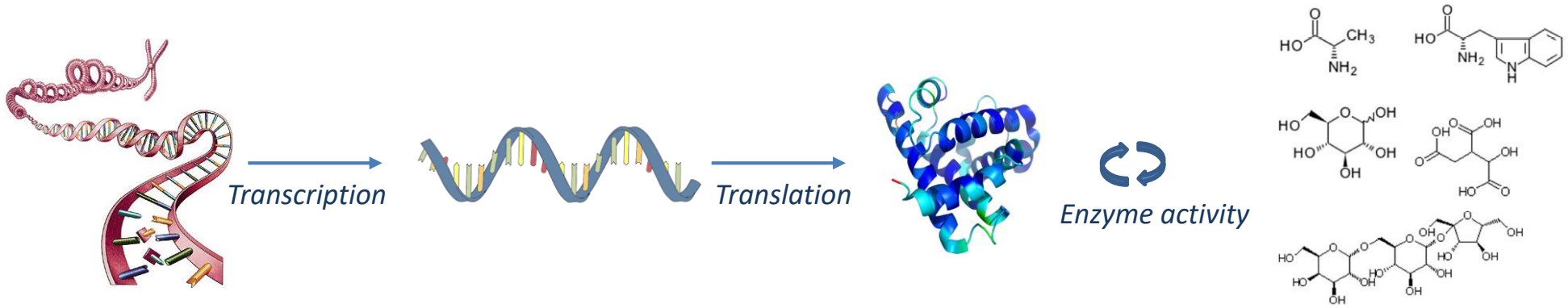

van der Greef et al, Curr Opin Chem Biol 2004

## Systems biology

## Personalized medecine



# Omics as key technologies for personalized medicine




Genes → Genome → **GENOMICS**

mRNAs → Transcriptome → **TRANSCRIPTOMICS**

Proteins → Proteome → **PROTEOMICS**

Metabolites → Metabolome → **METABOLOMICS**

(Genotype) ⟶ (Phenotype)

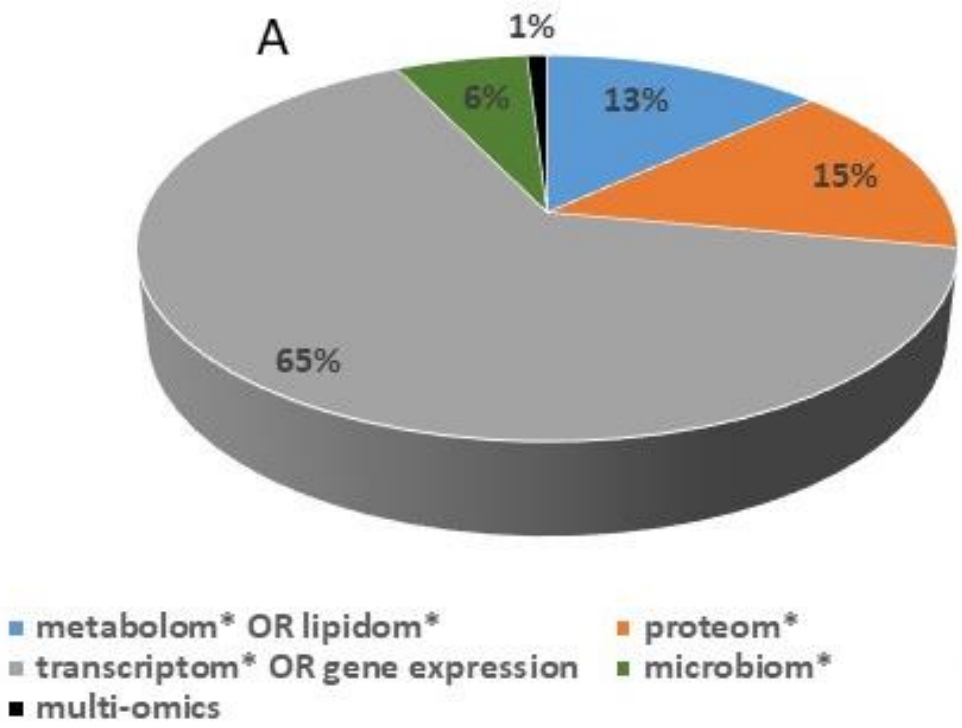
« What might happen »
« How it is regulated »

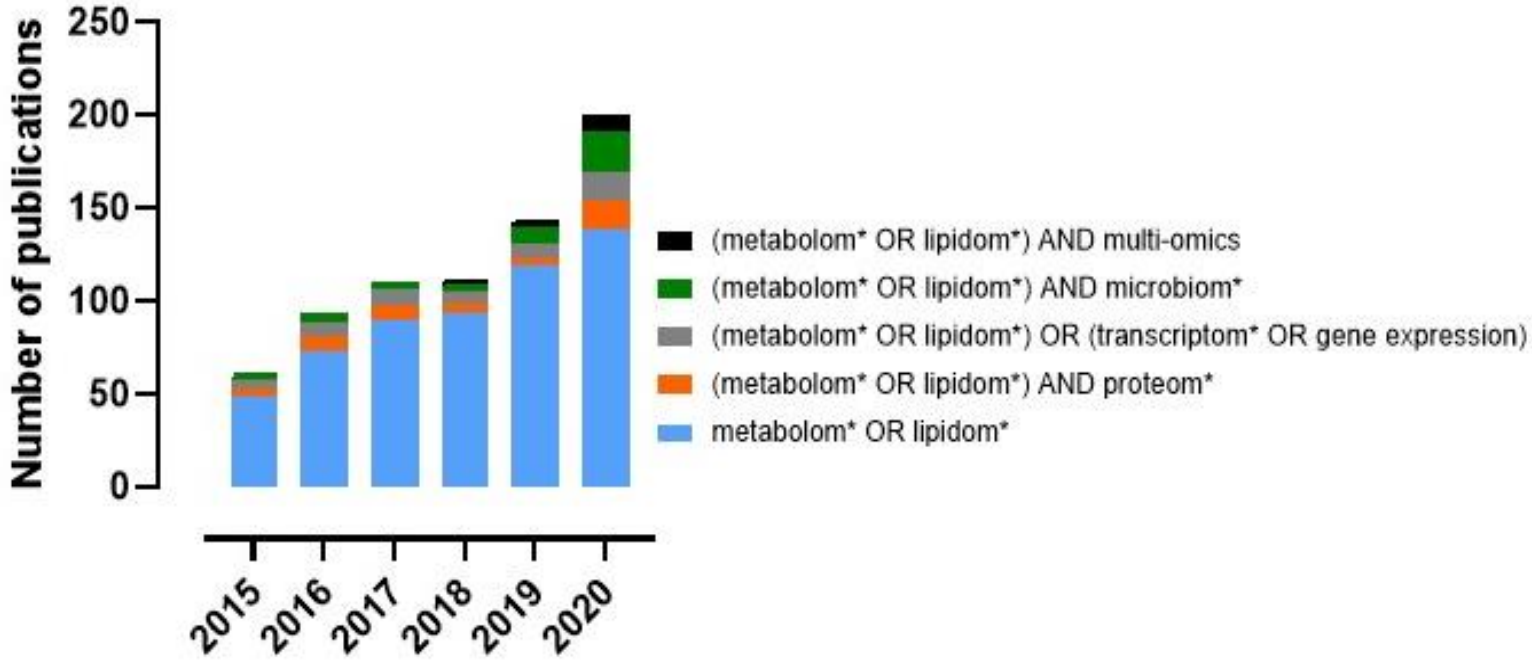
« What is happening »

*Genotype: part of the genetic makeup of an organism which determines one of its characteristics*

*Phenotype: observable characteristics or traits of an organism*

# Omics based technologies for biomarker discovery in the medical field




(Castelli et al., Anal. Bioanal. Chem., 2021)

# After having been mostly developed by analytical chemists during the 2000's, metabolomics is now part of the tools available for clinical phenotyping

| NATURE |

## Succinate is an inflammatory signal that induces IL-1β through HIF-1α

G. M. Tannahill[1], A. M. Curtis[1], J. Adamik[2], E. M. Palsson-McDermott[1], A. F. McGettrick[1], G. Goel[3], C. Frezza[4,5], N. J. Bernard[1], B. Kelly[1], N. H. Foley[1], L. Zheng[4], A. Gardet[6], Z. Tong[7], S. S. Jany[1], S. C. Corr[1], M. Haneklaus[1], B. E. Caffrey[8], K. Pierce[6], S. Walmsley[9], F. C. Beasley[10], E. Cummins[11], V. Nizet[10], M. Whyte[9], C. T. Taylor[11], H. Lin[7], S. L. Masters[12], E. Gottlieb[4], V. P. Kelly[1], C. Clish[6], P. E. Auron[2]*, R. J. Xavier[3,5]* & L. A. J. O'Neill[1]

| NATURE |

## Metabolites produced by commensal bacteria promote peripheral regulatory T-cell generation

Nicholas Arpaia[1,2], Clarissa Campbell[1,2], Xiying Fan[1,2], Stanislav Dikiy[1,2], Joris van der Veeken[1,2], Paul deRoos[1,2], Hui Liu[3], Justin R. Cross[3], Klaus Pfeffer[4], Paul J. Coffer[1,2,5] & Alexander Y. Rudensky[1,2]

## Personal Omics Profiling Reveals Dynamic Molecular and Medical Phenotypes

Cell

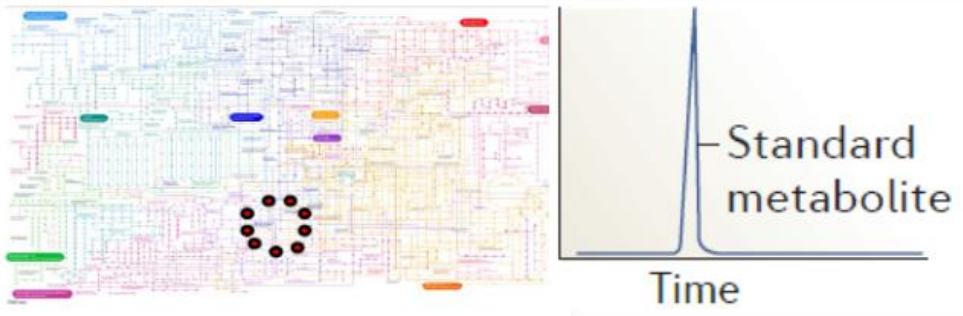
Rui Chen,[1,11] George I. Mias,[1,11] Jennifer Li-Pook-Than,[1,11] Lihua Jiang,[1,11] Hugo Y.K. Lam,[1,12] Rong Chen,[2,12] Elana Miriami,[1] Konrad J. Karczewski,[1] Manoj Hariharan,[1] Frederick E. Dewey,[3] Yong Cheng,[1] Michael J. Clark,[1] Hogune Im,[1] Lukas Habegger,[6,7] Suganthi Balasubramanian,[6,7] Maeve O'Huallachain,[1] Joel T. Dudley,[2] Sara Hillenmeyer,[1] Rajini Haraksingh,[1] Donald Sharon,[1] Ghia Euskirchen,[1] Phil Lacroute,[1] Keith Bettinger,[1] Alan P. Boyle,[1] Maya Kasowski,[1] Fabian Grubert,[1] Scott Seki,[2] Marco Garcia,[2] Michelle Whirl-Carrillo,[1] Mercedes Gallardo,[9,10] Maria A. Blasco,[9] Peter L. Greenberg,[4] Phyllis Snyder,[1] Teri E. Klein,[1] Russ B. Altman,[1,5] Atul J. Butte,[2] Euan A. Ashley,[3] Mark Gerstein,[6,7,8] Kari C. Nadeau,[2] Hua Tang,[1] and Michael Snyder[1,*]

## Metabolic Phenotypes of Response to Vaccination in Humans

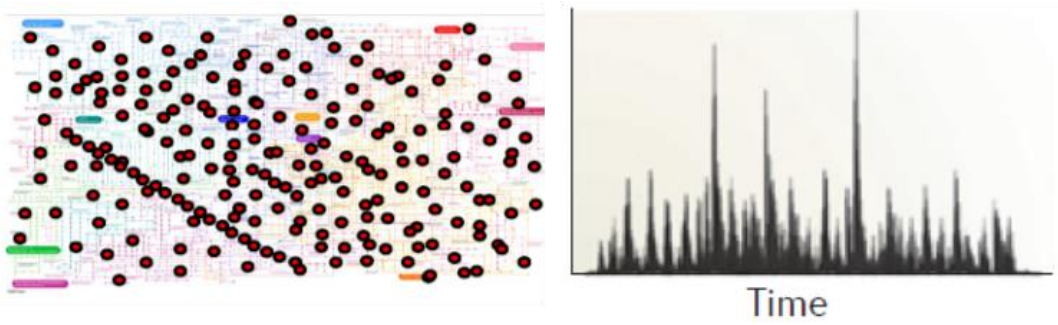
Cell

Shuzhao Li,[1,12] Nicole L. Sullivan,[2,12,14] Nadine Rouphael,[1,3] Tianwei Yu,[4] Sophia Banton,[1] Mohan S. Maddur,[2] Megan McCausland,[2] Christopher Chiu,[2] Jennifer Canniff,[5] Sheri Dubey,[6] Ken Liu,[1] ViLinh Tran,[1] Thomas Hagan, Sai Duraisingham,[2] Andreas Wieland,[2] Aneesh K. Mehta,[1] Jennifer A. Whitaker,[1,13] Shankar Subramaniam,[7] Dean P. Jones,[1] Alessandro Sette,[8] Kalpit Vora,[6] Adriana Weinberg,[5] Mark J. Mulligan,[1,3] Helder I. Nakaya,[9,10] Myron Levin,[5] Rafi Ahmed,[2,11] and Bali Pulendran[2,10,15,*]

**Targeted metabolomics**
(Quantification)

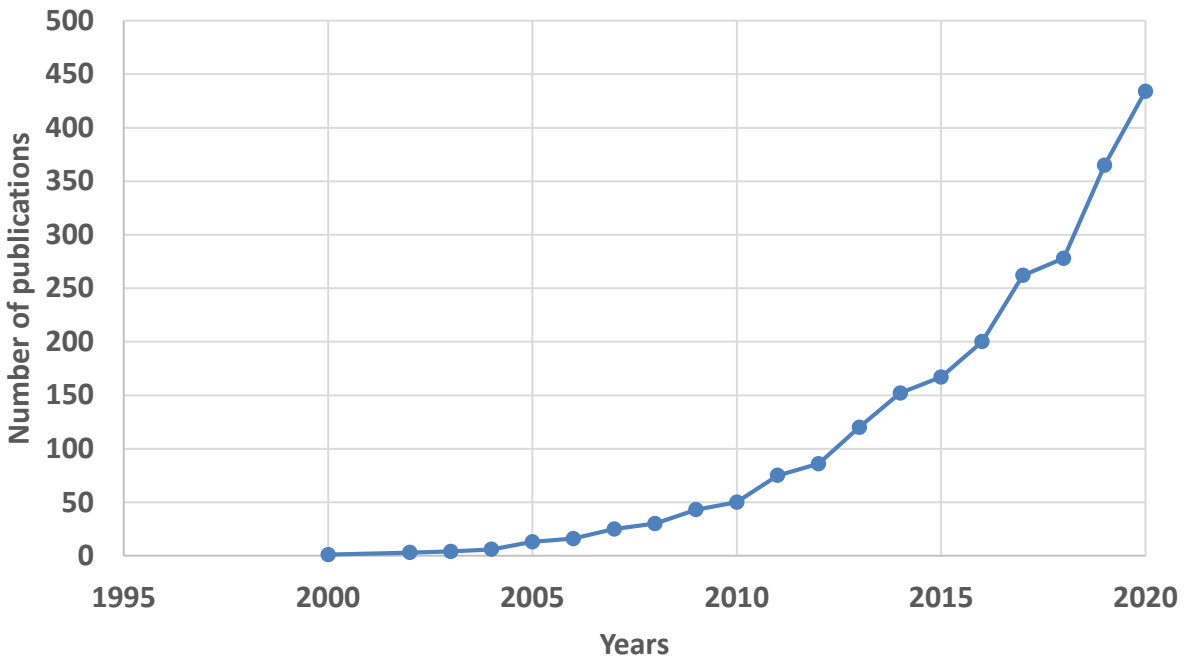


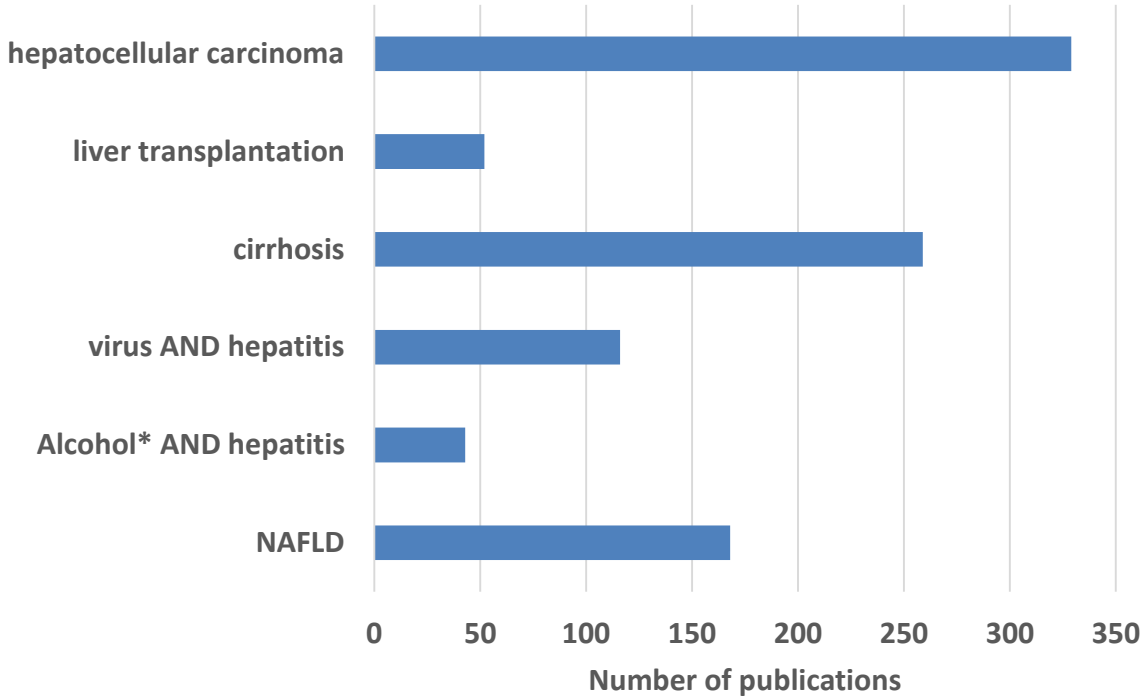

**Untargeted metabolomics**
(Metabolic profiling)



# Metabolomics in the field of hepatology

**> 2000 publications (Pubmed database)**




Search query: (metabolom*[Title/Abstract] OR metabonom*[Title/Abstract] OR metabotyp*[Title/Abstract]) AND ((liver disease)[Title/Abstract] OR hepatitis[Title/Abstract] OR cirrhosis[Title/Abstract] OR (liver transplantation)[Title/Abstract] OR (hepatocellular carcinoma)[Title/Abstract])

**Recent developments:**

**multi-omics integrated studies ++**
**impact of the gut microbiota ++**

ARTICLES
https://doi.org/10.1038/s41591-018-0061-3
nature medicine

# Molecular phenomics and metagenomics of hepatic steatosis in non-diabetic obese women

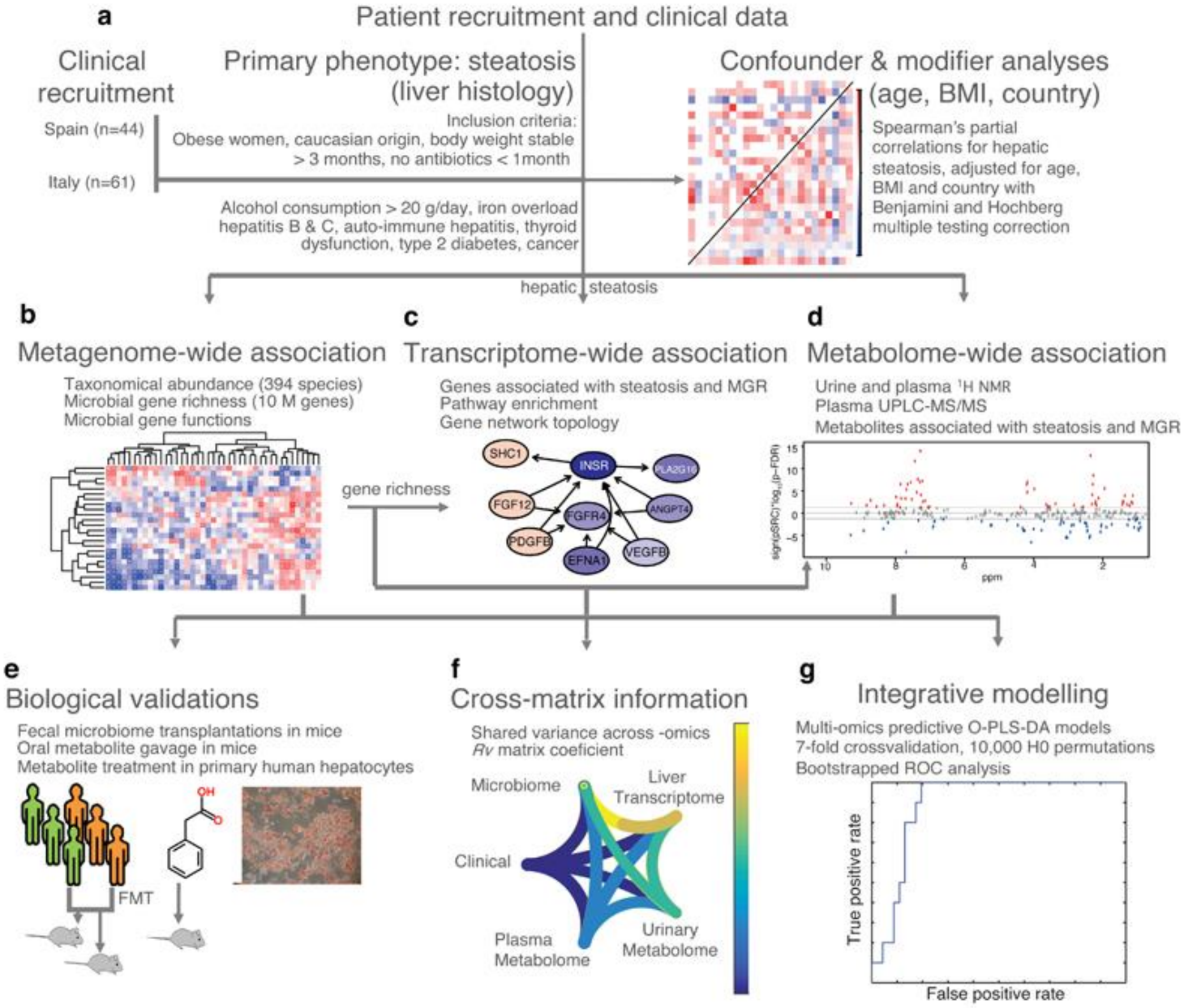
Lesley Hoyles[1,10], José-Manuel Fernández-Real[2,10*], Massimo Federici[3,10*], Matteo Serino[4,5], James Abbott[1], Julie Charpentier[4,5], Christophe Heymes[4,5], Jèssica Latorre Luque[2], Elodie Anthony[6], Richard H. Barton[1], Julien Chilloux[1], Antonis Myridakis[1], Laura Martinez-Gili[1], José Maria Moreno-Navarrete[2], Fadila Benhamed[6], Vincent Azalbert[4,5], Vincent Blasco-Baque[4,5], Josep Puig[2], Gemma Xifra[2], Wifredo Ricart[2], Christopher Tomlinson[1], Mark Woodbridge[1], Marina Cardellini[3], Francesca Davato[3], Iris Cardolini[3], Ottavia Porzio[7,8], Paolo Gentileschi[7], Frédéric Lopez[4,5], Fabienne Foufelle[9], Sarah A. Butcher[1], Elaine Holmes[1], Jeremy K. Nicholson[1], Catherine Postic[6], Rémy Burcelin[4,5*] and Marc-Emmanuel Dumas[1*]

**Shotgun sequencing of fecal metagenome**
**Host phenome (hepatic transcriptome, plasma and urine metabolomics)**

**Molecular networks linking the gut microbiome and the host phenome to hepatic steatosis.**

**Steatosis: low microbial gene richness, hepatic inflammation, dysregulation of aromatic and branched chain AA metabolism.**

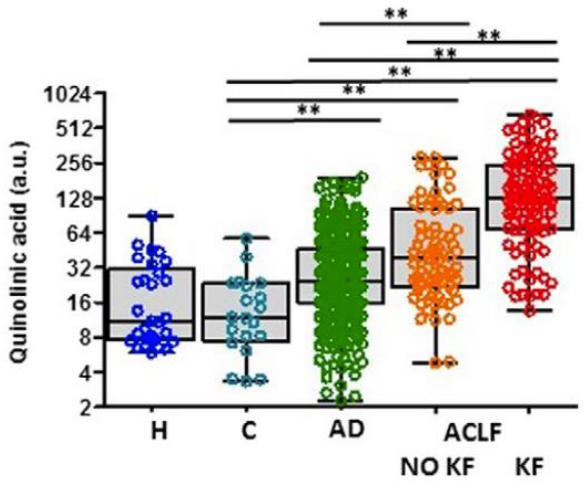
**Microbiota transplants and chronic treatment with phenylacetic acid trigger steatosis and dysregulation of BCAA metabolism**

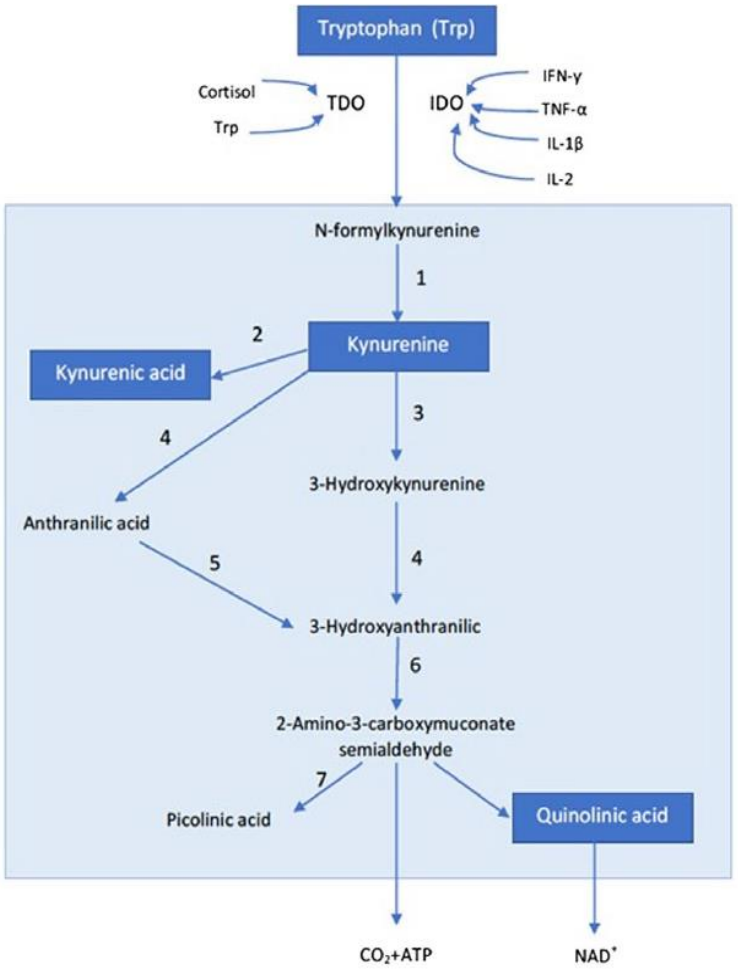


# Orchestration of Tryptophan-Kynurenine Pathway, Acute Decompensation, and Acute-on-Chronic Liver Failure in Cirrhosis

Joan Clària,[1,2*] Richard Moreau,[1,3*] François Fenaille,[4] Alex Amorós,[1] Christophe Junot,[4] Henning Gronbaek,[5] Minneke J. Coenraad,[6] Alain Pruvost,[7] Aurélie Ghettas,[7] Emeline Chu-Van,[4] Cristina López-Vicario,[2] Karl Oettl,[8] Paolo Caraceni,[9] Carlo Alessandria,[10] Jonel Trebicka,[1,11,12] Marco Pavesi,[1] Carme Deulofeu,[1] Agustin Albillos,[13] Thierry Gustot,[14] Tania M. Welzel,[12] Javier Fernández,[1,2] Rudolf E. Stauber,[8] Faouzi Saliba,[15] Noémie Butin,[4] Benoit Colsch,[4] Christophe Moreno,[14] François Durand,[3] Frederik Nevens,[16] Rafael Bañares,[17] Daniel Benten,[18] Pere Ginès,[2] Alexander Gerbes,[19] Rajiv Jalan,[20] Paolo Angeli,[1,21] Mauro Bernardi,[9] and Vicente Arroyo[1]; for the CANONIC Study Investigators of the EASL Clif Consortium, Grifols Chair and the European Foundation for the Study of Chronic Liver Failure (EF Clif)

HEPATOLOGY, VOL. 69, NO. 4, 2019



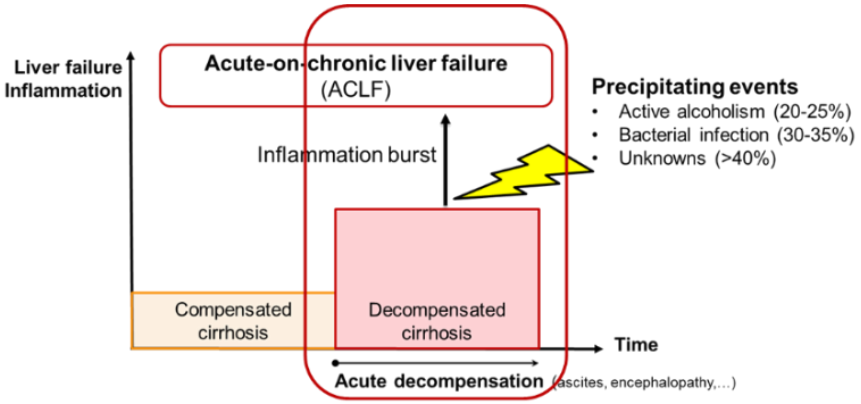

**Kynurenine pathway is activated in patient with acute decompensation and acute-on-chronic liver failure**

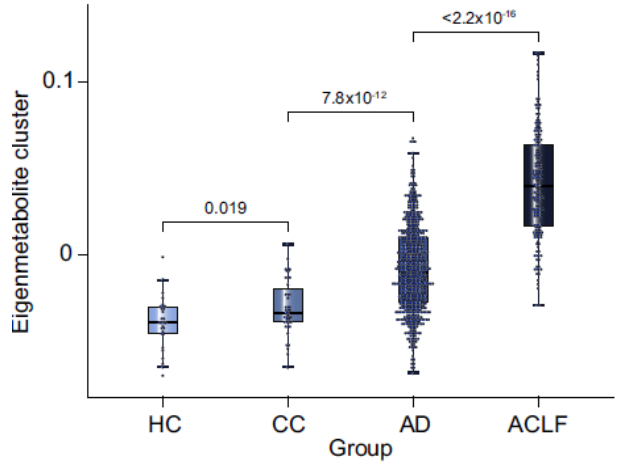
Confirmation by a quantitative LC-MS/MS assay for kynurenic acid, quinolinic acid, kynurenine and tryptophan on 234 samples (validation cohort)

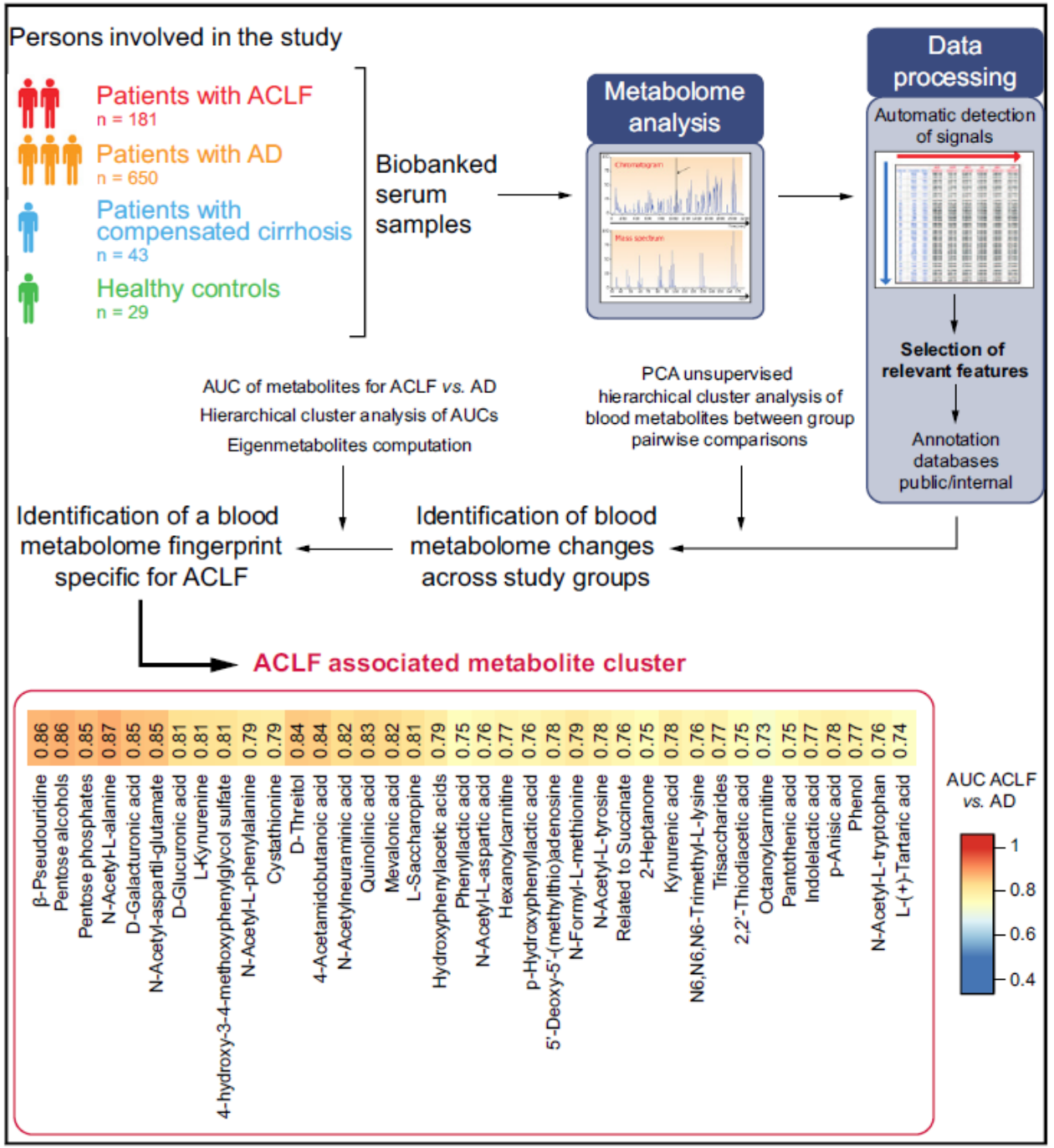
JOURNAL
OF HEPATOLOGY

# Blood metabolomics uncovers inflammation-associated mitochondrial dysfunction as a potential mechanism underlying ACLF

Richard Moreau[1,2,*,†], Joan Clària[1,3,4,†], Ferran Aguilar[1,†], François Fenaille[5,†], Juan José Lozano[4], Christophe Junot[5], Benoit Colsch[5], Paolo Caraceni[6], Jonel Trebicka[1,7], Marco Pavesi[1], Carlo Alessandria[8], Frederik Nevens[9], Faouzi Saliba[10], Tania M. Welzel[7], Agustin Albillos[11], Thierry Gustot[12], Javier Fernández[1,3,4], Christophe Moreno[12], Maurizio Baldassarre[6], Giacomo Zaccherini[6], Salvatore Piano[13], Sara Montagnese[13], Victor Vargas[14], Joan Genescà[14], Elsa Solà[3,4], William Bernal[15], Noémie Butin[5], Thaïs Hautbergue[5], Sophie Cholet[5], Florence Castelli[5], Christian Jansen[16], Christian Steib[17], Daniela Campion[8], Raj Mookerjee[18], Miguel Rodríguez-Gandía[11], German Soriano[19], François Durand[2], Daniel Benten[20], Rafael Bañares[21], Rudolf E. Stauber[22], Henning Gronbaek[23], Minneke J. Coenraad[24], Pere Ginès[3,4], Alexander Gerbes[17], Rajiv Jalan[1,18], Mauro Bernardi[6], Vicente Arroyo[1], Paolo Angeli[1,13], for the CANONIC Study Investigators of the EASL Clif Consortium, Grifols Chair and the European Foundation for the Study of Chronic Liver Failure (EF Clif)



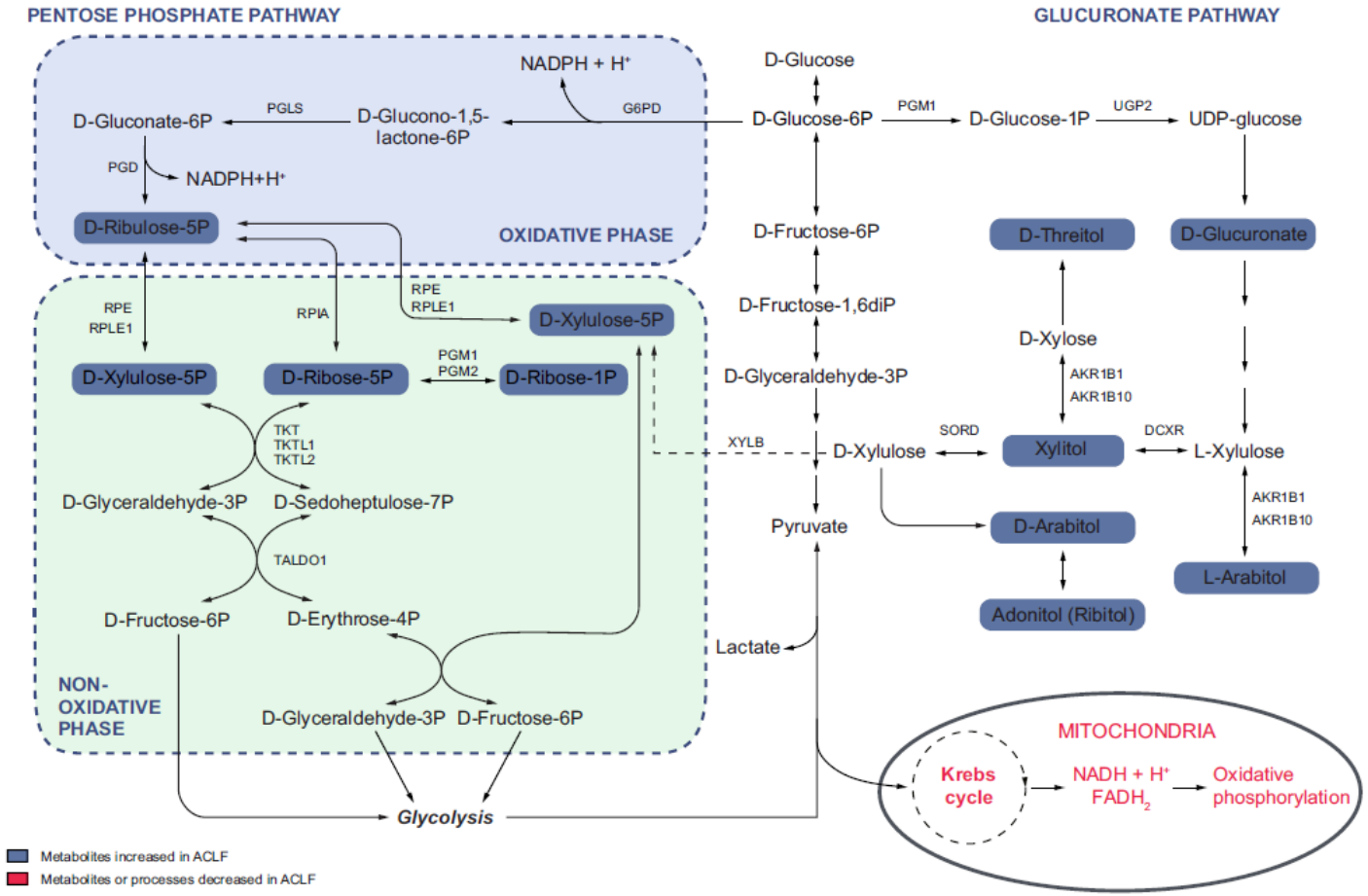
JOURNAL
OF HEPATOLOGY

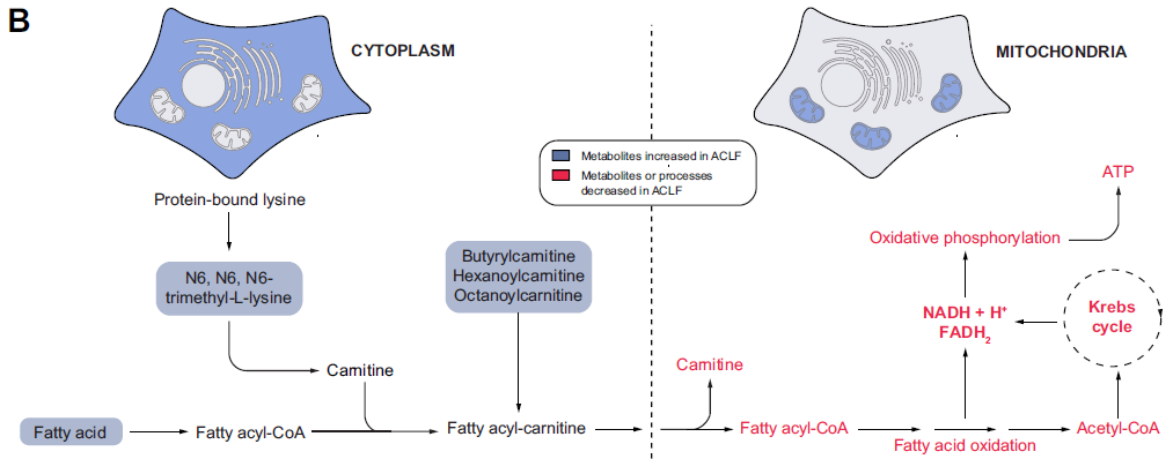
# Blood metabolomics uncovers inflammation-associated mitochondrial dysfunction as a potential mechanism underlying ACLF

The ACLF associated metabolite cluster correlates with systemic inflammation.

It also reflects:

- increased proteolysis and lipolysis,
- changes in intracellular metabolism in response to the proliferation of innate immunity cells,
- Reduced mitochondrial oxydation,



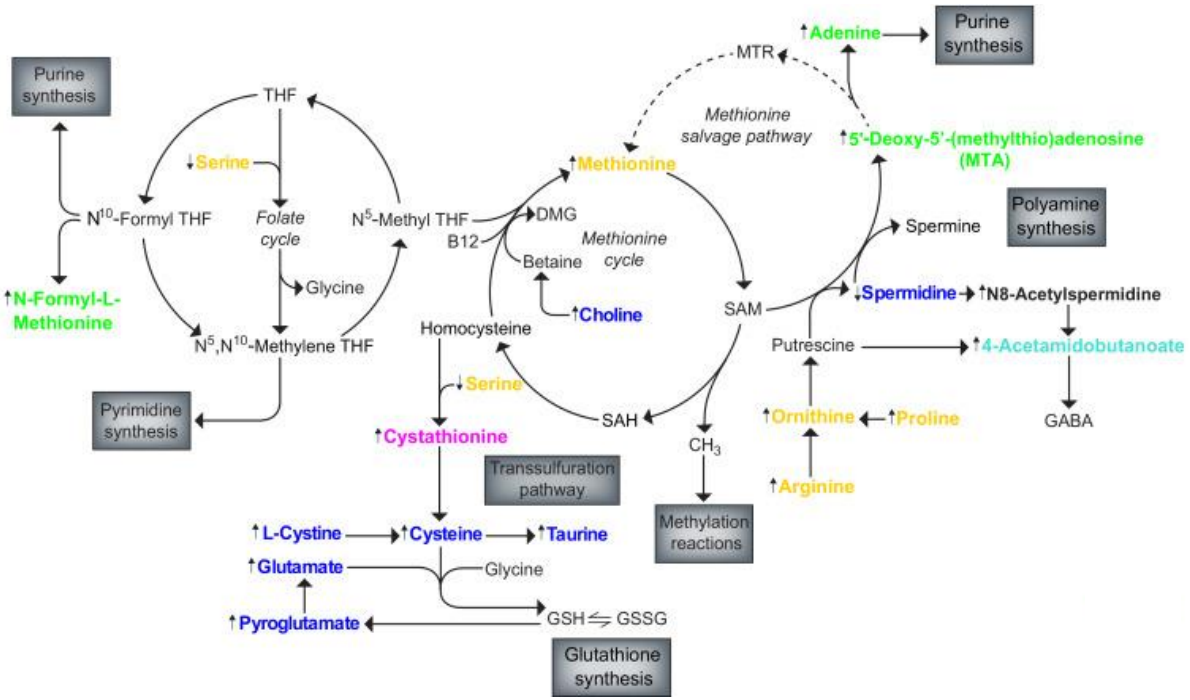
JOURNAL OF HEPATOLOGY

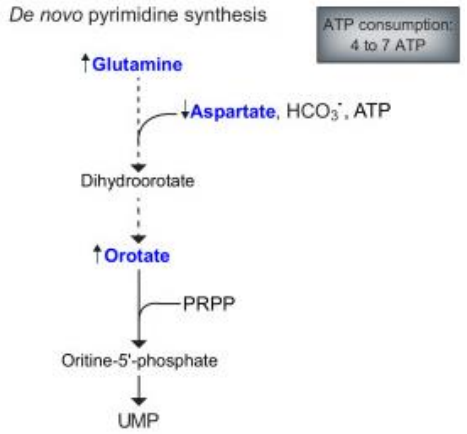
# Assessing the role of amino acids in systemic inflammation and organ failure in patients with ACLF

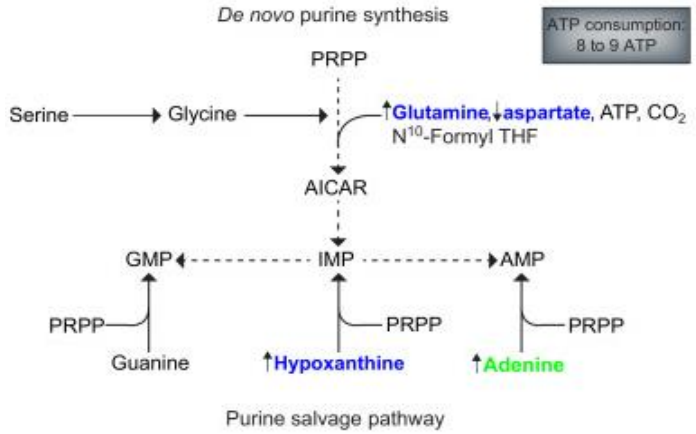
Giacomo Zaccherini[1,2,†], Ferran Aguilar[1,†], Paolo Caraceni[2], Joan Clària[1,3,4], Juan José Lozano[4], François Fenaille[5], Florence Castelli[5], Christophe Junot[5], Anna Curto[1], Chiara Formentin[6], Emmanuel Weiss[1,7], Mauro Bernardi[2], Rajiv Jalan[1,8], Paolo Angeli[1,6], Richard Moreau[1,9,10,*,‡], Vicente Arroyo[1,‡]

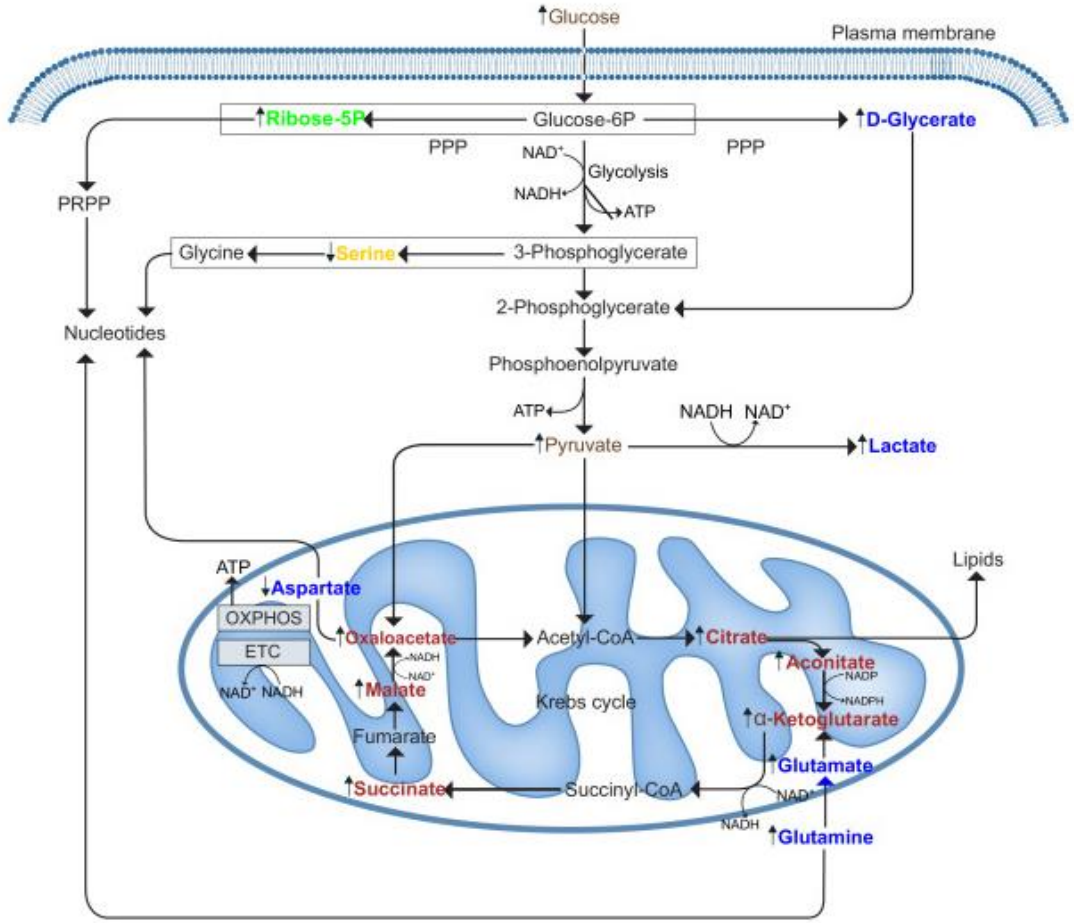
## Reanalysis of the blood metabolomic data of the CANONIC study

- Blood AA fuel protein and nucleotide synthesis required for intense systemic inflammation.
- Extensive catabolism of ketogenic AA to produce energy substrates in peripheral organs



# Which questions do we now address?

- Improve stratification power regarding the decompensation of cirrhosis
- Provide new insights into pathophysiology of ACLF (biomarkers of precipitants and organ failure)
- Response to treatment
- Pronostic biomarkers that could be translated into clinics

# Serum Levels of Metabolites Produced by Intestinal Microbes and Lipid Moieties Independently Associated With Acute on Chronic Liver Failure and Death in Patients With Cirrhosis

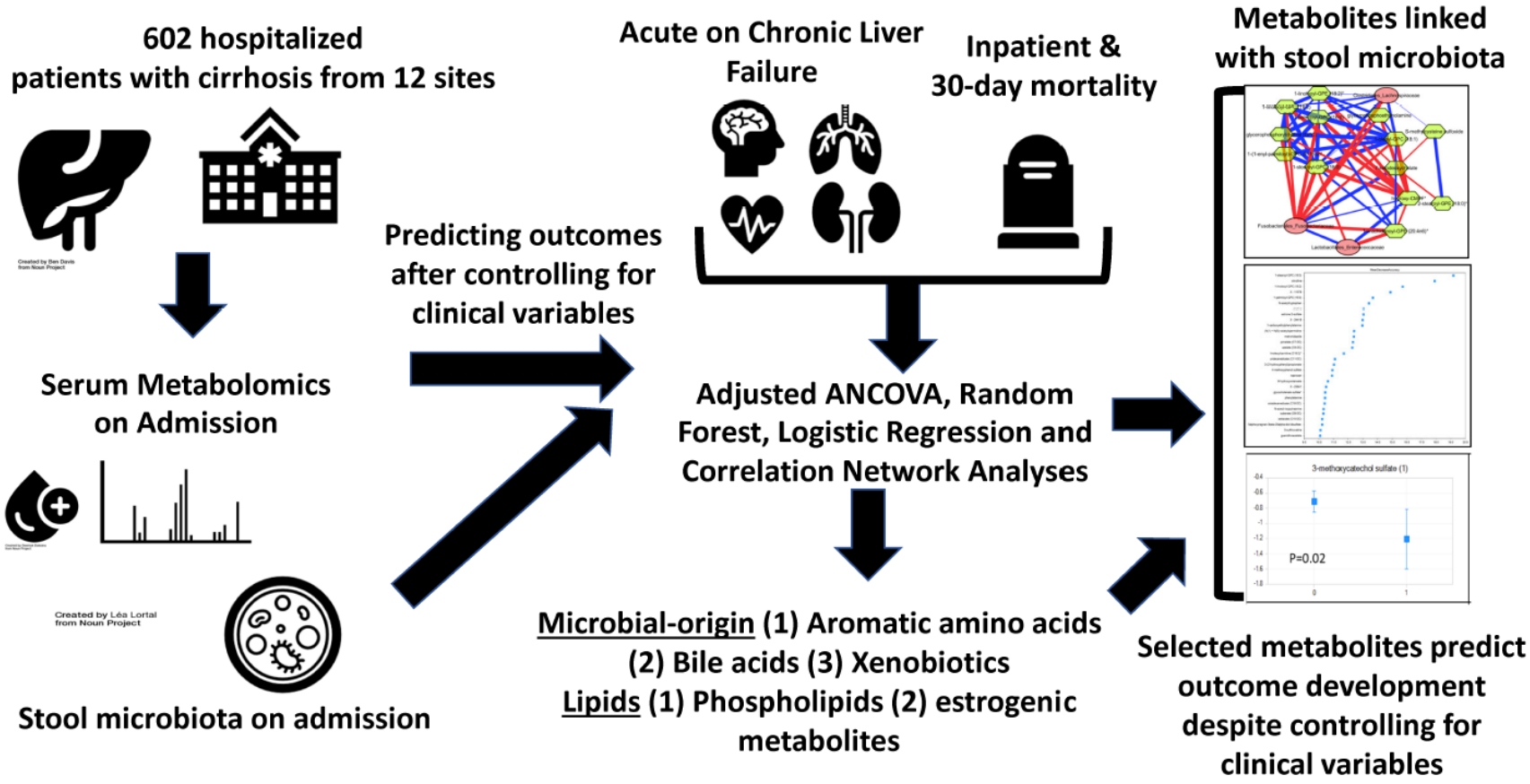
**Bajaj J.S. et al. Gastroenterology, 2020**

Serum samples of 602 patients collected at admission

88 developed ACLF (15%)
43 died in the hospital (7%)
72 died within 30 days (12%)

**Metabolomic signature associated with ACLF and death:**

- ✓ Increased levels of metabolites of microbioal origin (aromatic compounds, secondary biles acids, benzoic acid)
- ✓ Increased levels of estrogen metabolites
- ✓ Decreased levels of phospholipids



**Over the last ten years, the number of biomarkers derived from omics based approaches, approved by regulatory agencies and used in clinical settings remains far from expectations!**

Clinical Chemistry 60:10
1256–1257 (2014)

Perspective

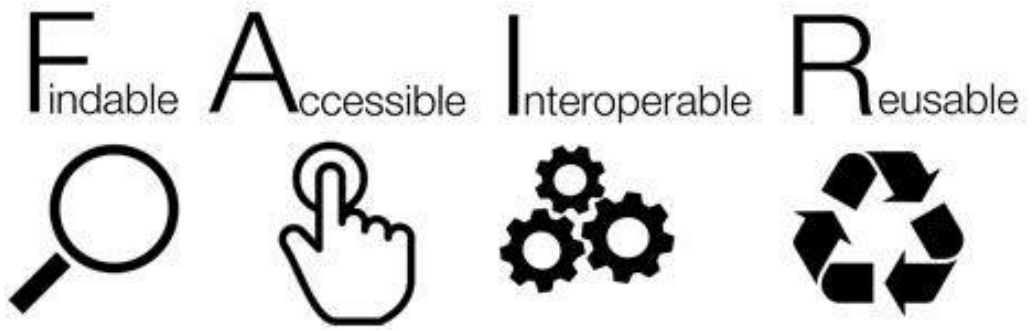
# Where Are All the New Omics-Based Tests?

Patrick M. Bossuyt[1*]

Several challenges need to be overcome to foster the implementation of omics in personalized medicine and in clinical practice:

- data production for biomarker discovery and validation,
- Translation of omics signatures into assays for medical laboratories
- Point of care tests.

# The main challenges related to omics data production




- ✓ Large multi-center cohorts as well as validation cohorts to increase statistical power and biomarker specificity and avoid confounding factors.
- ✓ Standardized data production workflow with improved robustness and capability of automated interpretation of the huge amount of data generated.
- ✓ Linked untargeted and targeted quantitative approaches for proper analytical validation of biomarker candidates.
- ✓ Improved metabolome coverage and metabolite identification confidence level.

# The metabolomic workflow

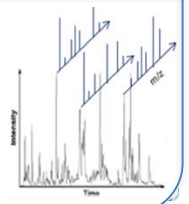
## 1 Sample preparation

## 2 Metabolome analysis

GC/MS

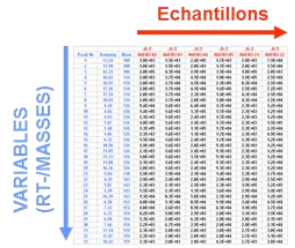
LC/MS

RMN



## 3 Data processing

**Automatic detection of signals**

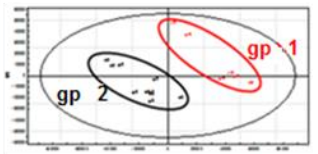



**Selection of relevant features**

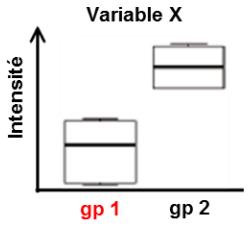
**Annotation** :
Databases
public / internal

## 4 Statistical analyses

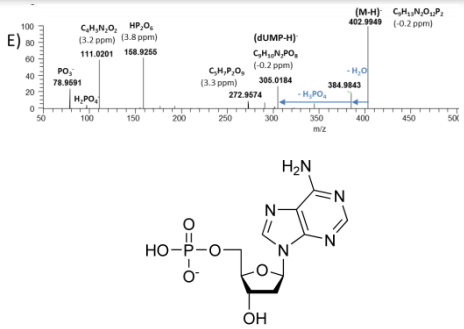
**Multivariate**




**Univariate**

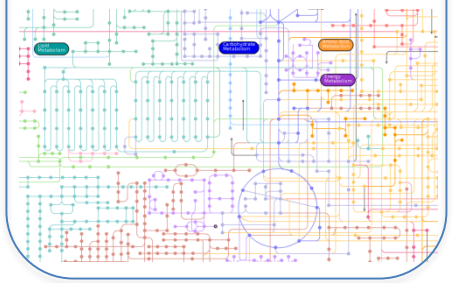



## 5 Metabolite identification



- To confirm annotations
- To characterize unknowns

## 6 Data visualization



# Batch to batch effects may compromise data re-use in the frame of untargeted metabolomics approaches

***part 1*** *(Dec. 2016)*

| | B | C | G | H | I | J | DE |
|---|---|---|---|---|---|---|---|
| 1 | | Group | 1 | 1 | 1 | 1 | |
| 2 | | SubGroup | 11 | 11 | 11 | 11 | |
| 3 | mz | RT (mi | 2-C1 | 2-C4 | 2-C9 | 2-E1 | Annotation |
| 862 | 203.0541 | 7.49 | 99362.53 | 9722.862 | 18559 | 20087.36 | NA |
| 863 | 203.0559 | 6.41 | 71015.29 | 68043.2 | 8875.421 | 51001.99 | NA |
| 864 | 203.0673 | 8.48 | 65797.6 | 83563.82 | 50967.94 | 45883.1 | NA |
| 865 | 203.0673 | 5.19 | 70188.11 | 247774.6 | 37803.5 | 36702.89 | NA |
| 866 | 203.0824 | 3.81 | 9972380 | 12998564 | 7235510 | 14344017 | L-Tryptophan _M-H_Mono_3. |
| 867 | 203.0825 | 1.66 | 481963.5 | 153078.8 | 249169.7 | 376764.5 | NA |
| 868 | 203.0924 | 2.06 | 178718.7 | 50073.22 | 98091.84 | 123049.2 | NA |
| 869 | 203.0943 | 2.83 | 27412.25 | 9703.278 | 16533.15 | 23487.23 | NA |
| 870 | 203.1037 | 9.02 | 6071.539 | 3296.272 | 3397.401 | 4249.104 | NA |
| 871 | 203.9972 | 2.25 | 154.3777 | 4005.641 | 392.0382 | 24765.69 | NA |
| 872 | 204.0137 | 6.68 | 11301.86 | 3702.277 | 15789.83 | 8371.775 | NA |
| 873 | 204.0334 | 9.44 | 4253.019 | 1663.416 | 2842.252 | 6606.748 | NA |
| 874 | 204.0512 | 7.80 | 21974.09 | 4484.383 | 15518.31 | 12721.81 | NA |
| 875 | 204.0665 | 2.44 | 928595.4 | 1092329 | 599208.1 | 2164618 | Indolelactic acid_M-H_Mono |
| 876 | 204.0706 | 8.49 | 2447.438 | 2527.517 | 1229.98 | 3372.858 | NA |
| 906 | 207.0773 | 3.34 | 757032.3 | 51676.82 | 32755.66 | 228438.6 | L-Kynurenine_M-H_Mono_3. |
| 907 | 207.0871 | 3.41 | 723737.9 | 46186.98 | 31498.62 | 216209.4 | NA |
| 908 | 207.3543 | 3.52 | 19762.05 | 27778.62 | 26139.36 | 18982.6 | NA |
| 909 | 207.3544 | 7.76 | 13739.32 | 18924.93 | 14243.27 | 21149.81 | NA |
| 947 | 212.0023 | 2.32 | 2892421 | 2778146 | 11108.61 | 679913.5 | Indoxyl sulfate_M-H_Mono_ |
| 948 | 212.0557 | 7.07 | 5683.415 | 19101.76 | 4070.753 | 11798.26 | NA |
| 969 | 247.0992 | 2.09 | 14123.6 | 5849.136 | 9831.569 | 10916.39 | N-acetyl-DL-tryptophan_M-H |
| 970 | 247.1264 | 1.74 | 12202.12 | 23413.05 | 25843.08 | 32935.61 | NA |
| 971 | 247.1357 | 1.49 | 104914.7 | 31345.87 | 82394.97 | 60224.55 | NA |

***part 2*** *(Oct. 2017)*

| 8 | ACLF_degree | | ACLF 3 | ACLF 3 | ACLF 3 | ACLF 3 | |
|---|---|---|---|---|---|---|---|
| 9 | mz | RT (min) | 593 | 624 | 686 | 697 | Annotation |
| 10 | 75.0085 | 7.51 | 730206.665 | 1779855.63 | 785311.344 | 924867.7302 | NA |
| 360 | 163.0397 | 1.62 | 57444.1392 | 738962.101 | 30500.6157 | 73436.71297 | 1018_Phenylpyruvic acid_M-H_Mono_1.36_TRU |
| 367 | 148.0435 | 7.80 | 149481.729 | 936508.169 | 1805640.76 | 1894619.59 | NA |
| 391 | 150.0015 | 1.80 | 915555.931 | 1902309.56 | 716501.972 | 631370.9258 | NA |
| 393 | 245.0928 | 2.24 | 42270.0609 | 245910319 | 608540.702 | 321494.5451 | 1107_N-acetyl-DL-tryptophan_M-H_Mono_2.31 |
| 394 | 150.0485 | 7.25 | 239159.356 | 205628.626 | 249180.691 | 142341.5824 | NA |
| 413 | 152.0017 | 4.37 | 1745103.39 | 1928931.36 | 60546.13 | 73318.36111 | NA |
| 414 | 152.0116 | 6.81 | 46162.0195 | 116316.458 | 376170.208 | 685457.7719 | NA |
| 415 | 152.0196 | 4.07 | 268228.281 | 103151.907 | 38430.337 | 35133.68502 | NA |
| 531 | 165.0764 | 4.87 | 701913.889 | 762910.962 | 8867.34299 | 283935.9458 | NA |
| 532 | 165.0764 | 5.86 | 254769.039 | 1463467.8 | 852096.783 | 644437.517 | NA |
| 533 | 165.0766 | 6.24 | 88175.1329 | 298668.137 | 154191.629 | 249462.7123 | NA |
| 534 | 188.0348 | 1.89 | 135752.201 | 5227741.67 | 236966.038 | 2591189.663 | 265_Kynurenic acid_M-H_Mono_2.28_TRUE |
| 536 | 166.0175 | 3.58 | 1673558.71 | 3379409.17 | 436535.286 | 971624.4962 | NA |
| 537 | 166.0433 | 8.07 | 506969.679 | 1734812.08 | 785166.425 | 438652.4626 | NA |
| 539 | 167.0710 | 1.52 | 395591.379 | 1063366.32 | 484457.517 | 76032.43282 | NA |
| 1314 | 257.9980 | 6.36 | 89169.1019 | 7857.49759 | 28222.1929 | 5918.639736 | NA |
| 1315 | 203.0821 | 4.13 | 10084428.2 | 15041486.1 | 1152936.58 | 46837793.8 | 289_DL-Tryptophan_M-H_Mono_3.86_TRUE/29 |
| 1316 | 258.0438 | 1.75 | 3758.99818 | 654560.305 | 1815.21326 | 18535.86882 | NA |
| 1317 | 258.0555 | 6.00 | 276484.614 | 1955110.89 | 75532.966 | 419095.6514 | NA |
| 1318 | 258.0557 | 5.53 | 267705.802 | 645093.463 | 89281.7804 | 1538628.442 | NA |
| 1319 | 258.0559 | 6.86 | 264893.194 | 754727.558 | 1015059.22 | 211103.4957 | NA |
| 1320 | 258.0615 | 8.20 | 552664.559 | 662226.708 | 581167.156 | 256808.8165 | NA |
| 1706 | 207.0771 | 3.29 | 343693.494 | 2439568.27 | 180077.537 | 527768.1569 | 50_L-Kynurenine_M-H_Mono_3.17_TRUE |
| 1707 | 326.0990 | 2.26 | 1847.3073 | 2723.35557 | 12463.513 | 275505.442 | NA |
| 1708 | 326.1090 | 5.54 | 464325.502 | 52313.5659 | 342254.98 | 86492.86135 | NA |
| 1709 | 326.1871 | 1.27 | 122658.306 | 454841.904 | 157195.949 | 67211.46772 | NA |

***Data fusion on annotated compounds after normalization***

**300 annotated metabolites included for analysis (2016, 100 samples)**

**156 annotated metabolites included for analysis (2019, 900 samples)**

# Batch to batch effects may compromise data re-use in the frame of untargeted metabolomics approaches

| Injection Order | Sample |
|---|---|
| 1 | Blank |
| 2 | Blank |
| 3 | QC |
| 4 | QC |
| 5 | QC |
| 6 | QC |
| 7 | QC |
| 8 | Blank |
| 9 | 8x dil. QC |
| 10 | 4x dil. QC |
| 11 | 2x dil. QC |
| 12 | QC |
| 13 | Blank |
| 14 | QC |
| 15 | Sample 1 |
| 16 | Sample 2 |
| 17 | Sample 3 |
| … | … |
| 24 | Sample 10 |
| 25 | Blank |
| 26 | QC |
| 27 | Sample 11 |
| … | … |
| 36 | Sample 20 |
| 37 | Blank |
| 38 | QC |
| … | … |

QCs for equilibration

Diluted QCs for data treatment (n=3 each)

10 biological samples

## 1. Repeatability filter

- CV of Internal Standards < 30%
- CV of QC Samples < 30%

## 2. Intensity and linearity filters

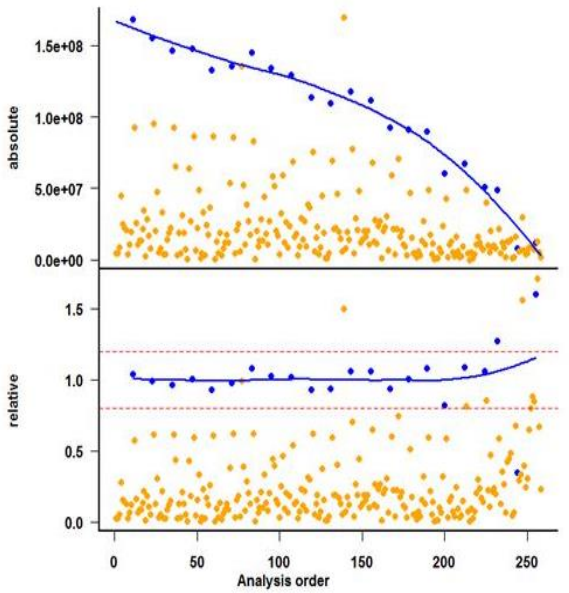
- Biological/blank>3
- Correlation to dilution <0.7

## 3. Drift correction

- Correct feature specific drift within a batch

## 4. Batch correction

- Correct drift across batches



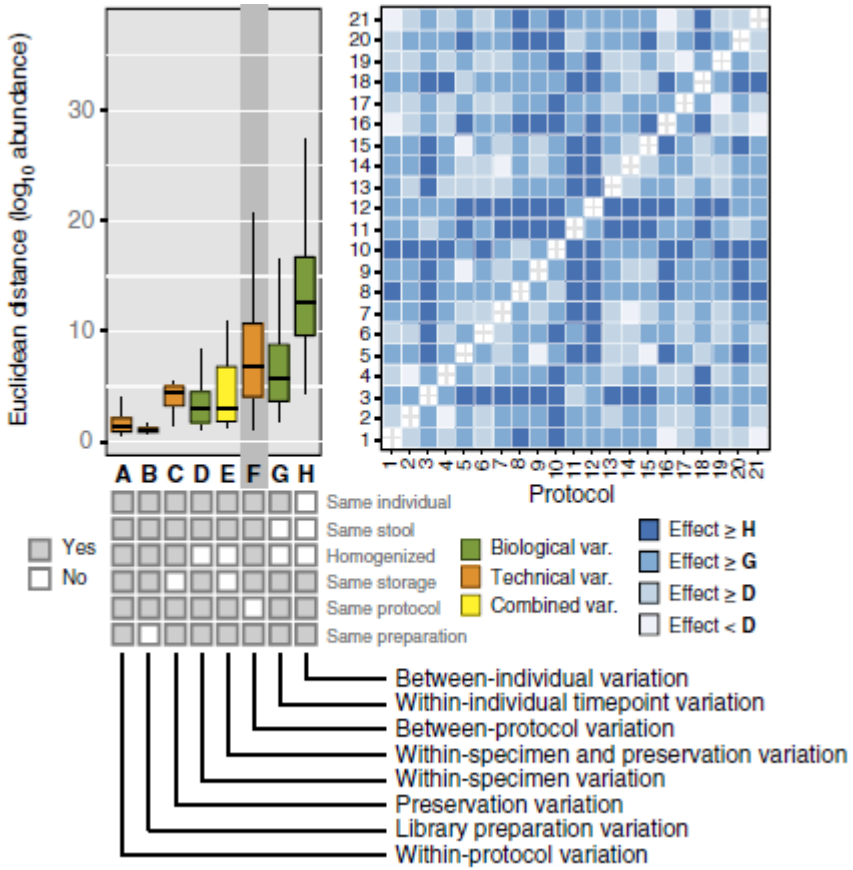

**LOESS Algorithm**
(Low Order non linear locally Estimated Smoothing Function)


Cleveland, J. Am. Stat. Assoc. 1979
Dunn WB, Nat. Protoc. 2011

# Data sharing: the need for standardization

Towards standards for human fecal sample processing in metagenomic studies

NATURE BIOTECHNOLOGY VOLUME 35 NUMBER 11 NOVEMBER 2017




## NIST plasma samples (SRM 1950) for metabolomics and lipidomics


Journal of Lipid Research: Full Article

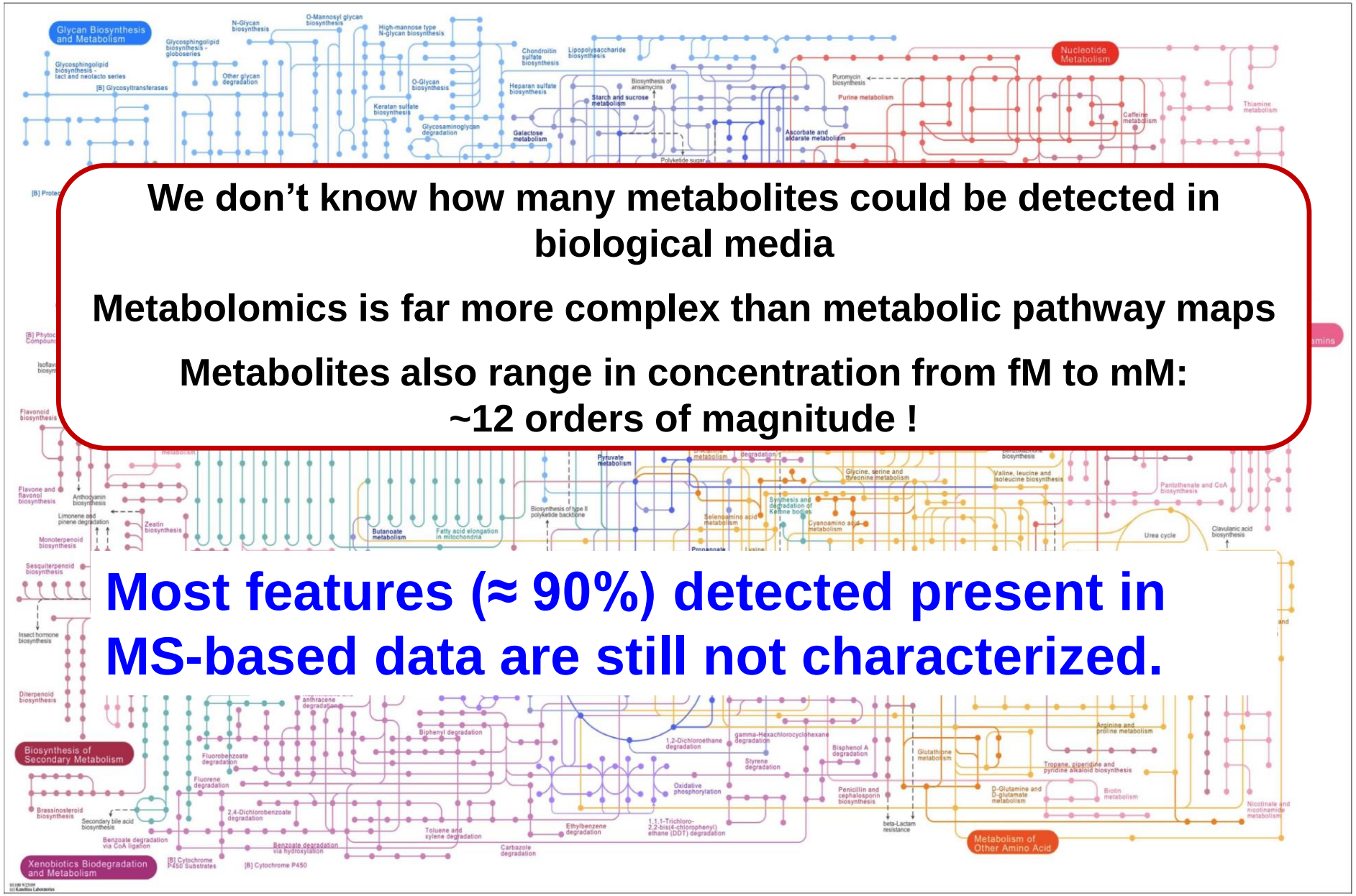
**Harmonizing Lipidomics: NIST Interlaboratory Comparison Exercise for Lipidomics using Standard Reference Material 1950 – Metabolites in Frozen Human Plasma**

John A. Bowden[*1], Alan Heckert[2], Candice Z. Ulmer[1], Christina M. Jones[1], Jeremy P. Koelmel[3]

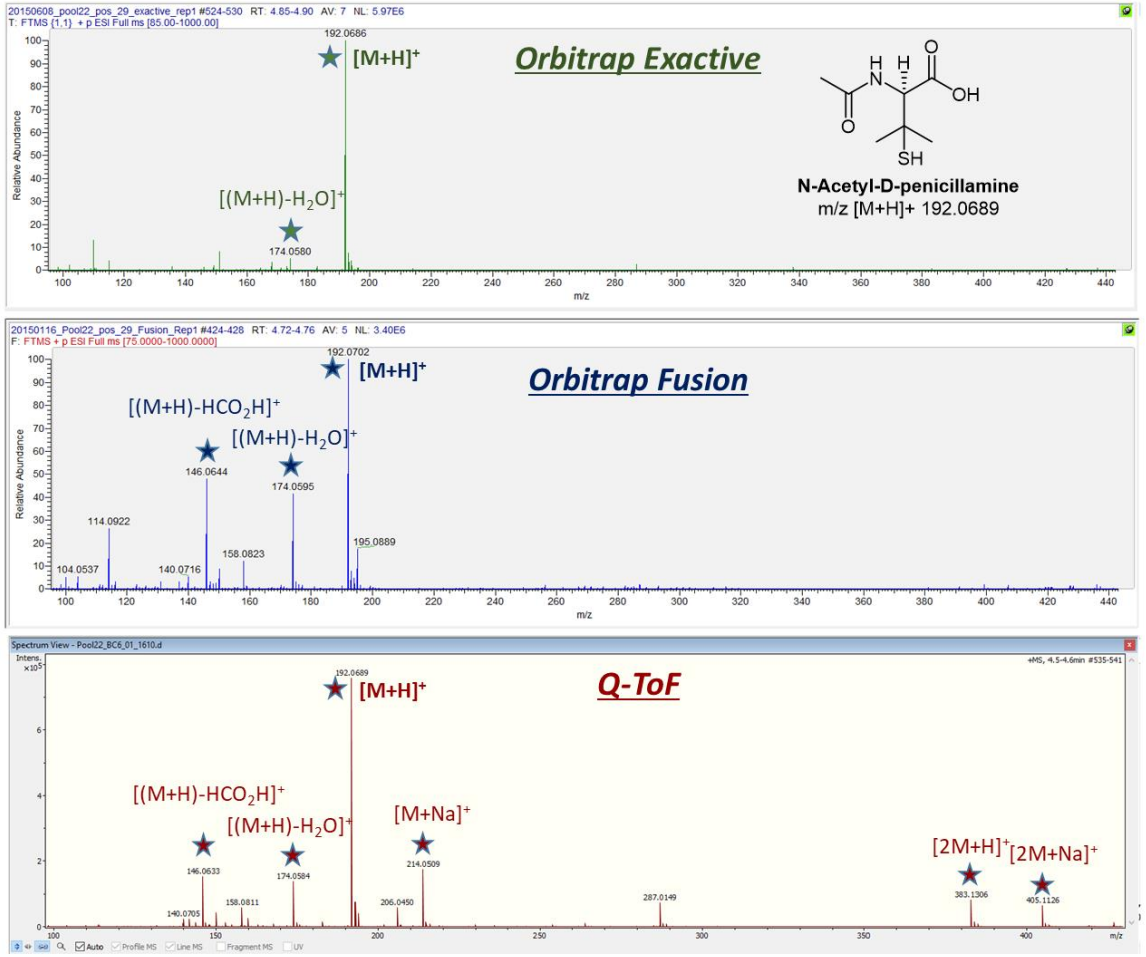
**Validating Quantitative Untargeted Lipidomics Across Nine Liquid Chromatography–High-Resolution Mass Spectrometry Platforms**

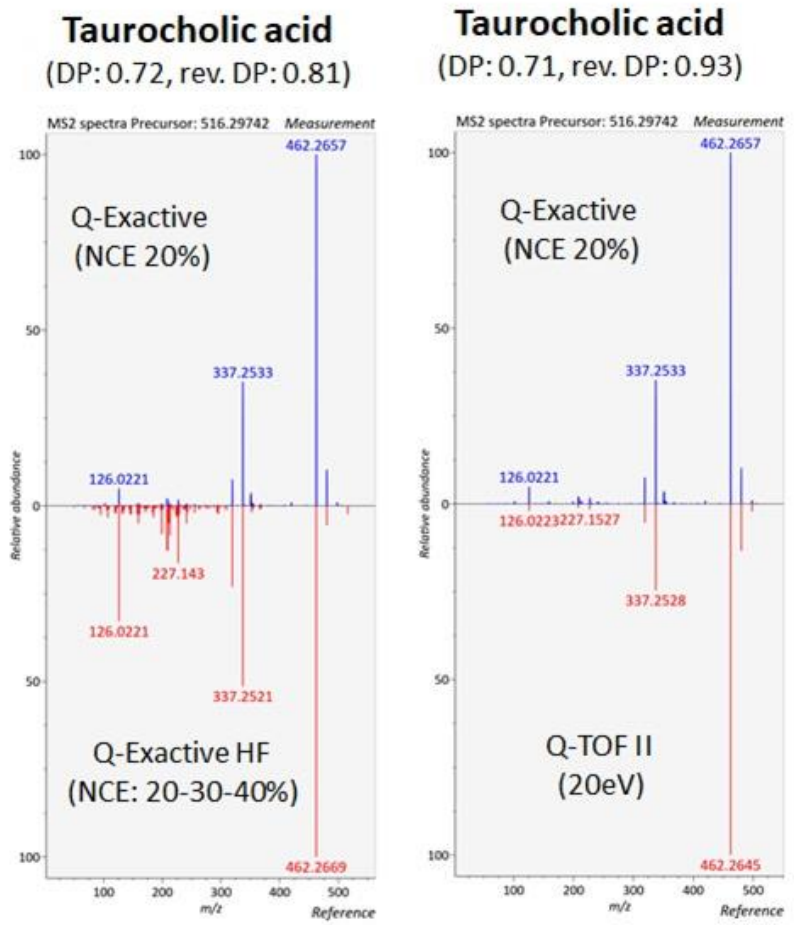
Tomas Cajka,[†] Jennifer T. Smilowitz,[‡,§] Oliver Fiehn[*,†,⊥]

analytical chemistry


We don’t know how many metabolites could be detected in biological media
Metabolomics is far more complex than metabolic pathway maps
Metabolites also range in concentration from fM to mM:
~12 orders of magnitude !
Most features (≈ 90%) detected present in MS-based data are still not characterized.
Glycan Biosynthesis and Metabolism
Nucleotide Metabolism
Biosynthesis of Secondary Metabolism
Xenobiotics Biodegradation and Metabolism
Metabolism of Other Amino Acid

# Annotation: we don't exactly produce the same kind of data and we don't process it in the same way




Damont A et al., unpublished

Castelli et al., Anal. Bioanal. Chem., 2021

# The need of metabolite identification/characterization reporting standards…

Metabolomics (2007) 3:211–221
DOI 10.1007/s11306-007-0082-2

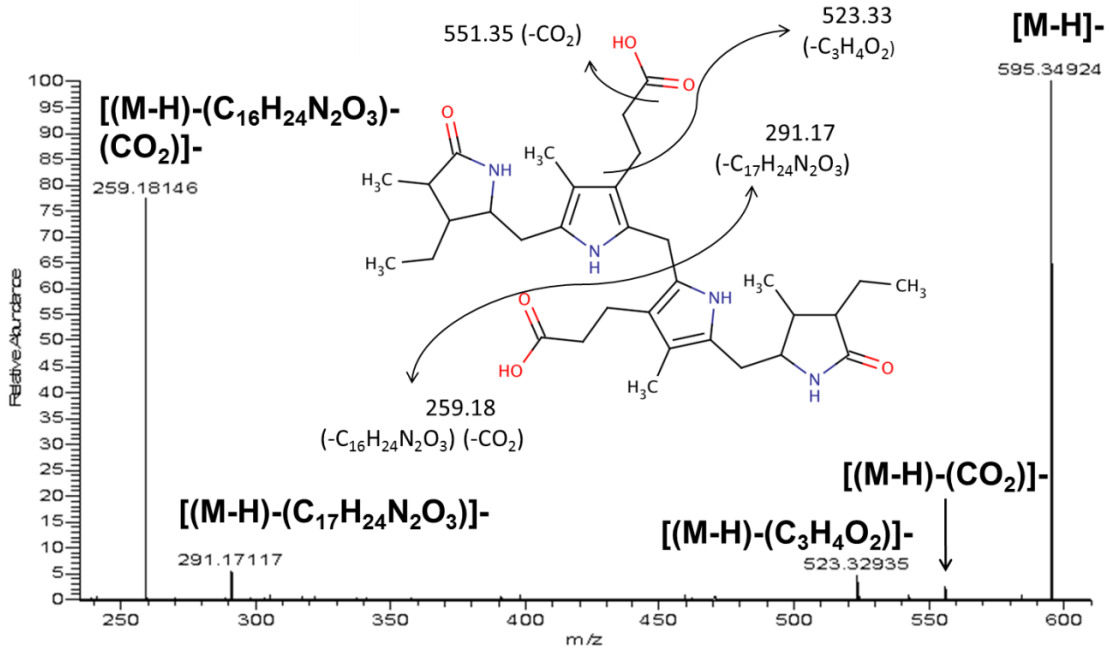
ORIGINAL ARTICLE

## Proposed minimum reporting standards for chemical analysis

**Chemical Analysis Working Group (CAWG) Metabolomics Standards Initiative (MSI)**

Lloyd W. Sumner · Alexander Amberg · Dave Barrett · Michael H. Beale · Richard Beger · Clare A. Daykin · Teresa W.-M. Fan · Oliver Fiehn · Royston Goodacre · Julian L. Griffin · Thomas Hankemeier · Nigel Hardy · James Harnly · Richard Higashi · Joachim Kopka · Andrew N. Lane · John C. Lindon · Philip Marriott · Andrew W. Nicholls · Michael D. Reily · John J. Thaden · Mark R. Viant

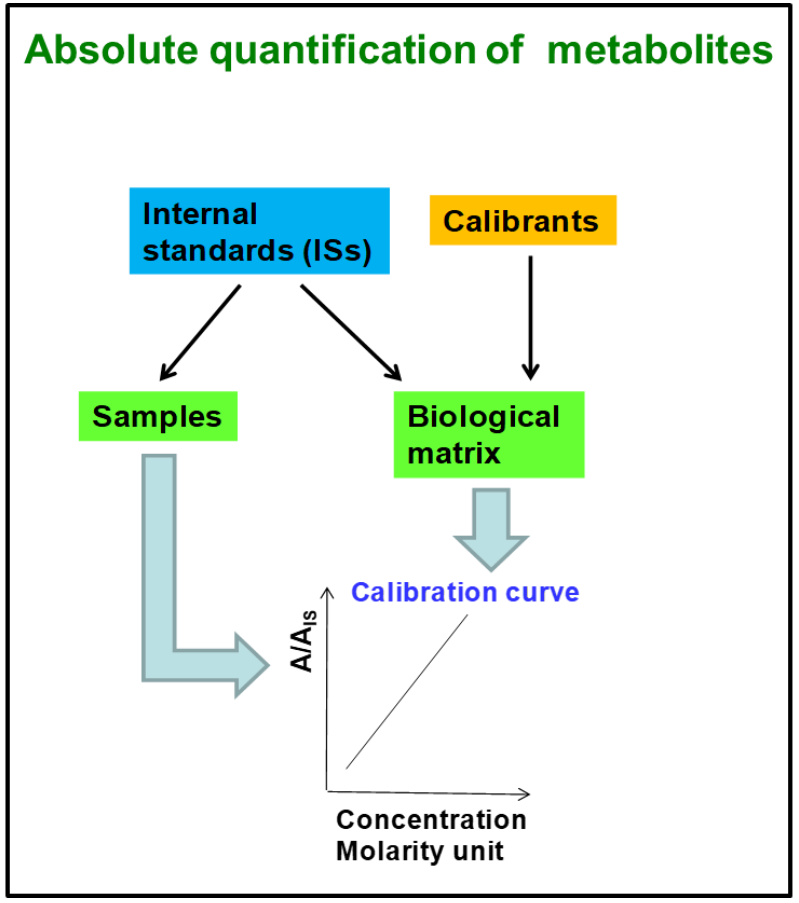
1. Identified compounds (see below).
2. Putatively annotated compounds (e.g. without chemical reference standards, based upon physicochemical properties and/or spectral similarity with public/commercial spectral libraries).
3. Putatively characterized compound classes (e.g. based upon characteristic physicochemical properties of a chemical class of compounds, or by spectral similarity to known compounds of a chemical class).
4. Unknown compounds—although unidentified or unclassified these metabolites can still be differentiated and quantified based upon spectral data.




Metabolite identification task group…

**Update under progress**

# Analytical validation of biomarker candidates: the need for targeted quantitative metabolomics approaches.

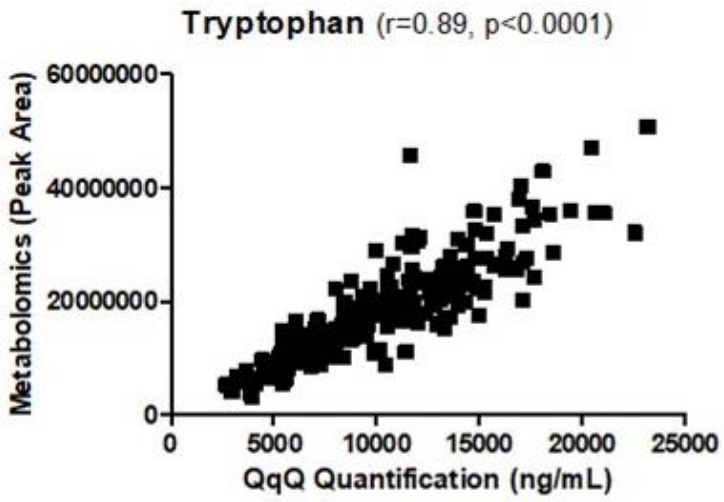


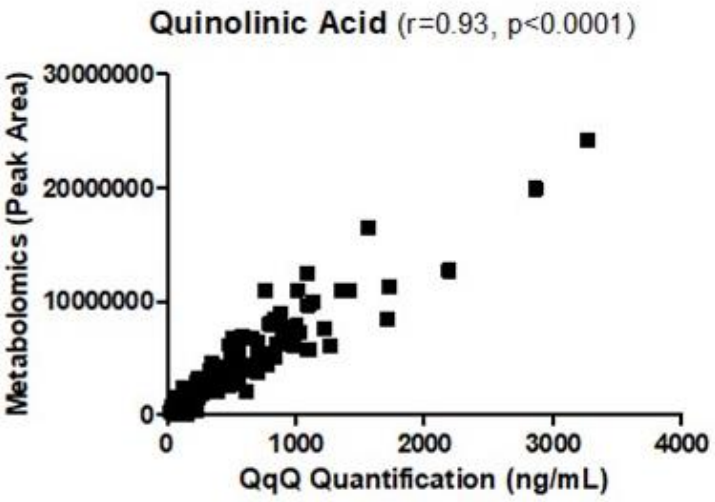

- $^{13}C$- and/or $^{15}N$ based Metabolic labeling (Mashego, J. Biotechnol. Bioeng., 2004; Lafaye, anal. Chem., 2005…)

- Chemical labeling

- Derivatization: dansyl chloride…

(Guo K, Anal Chem., 2009…)

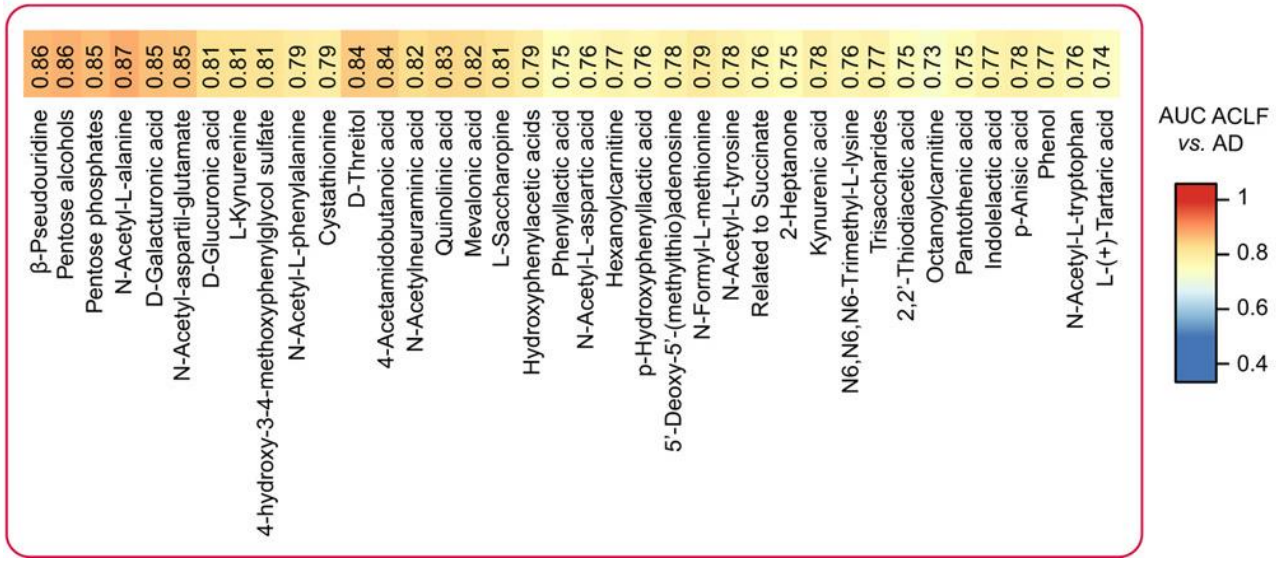



Castelli et al., Anal. Bioanal. Chem., 2021

# How to transfer metabolomics signatures from the research laboratories to the field?

- ✓ Omics signatures are complex and need to be simplified.
- ✓ There are small concentration variations from one group the other



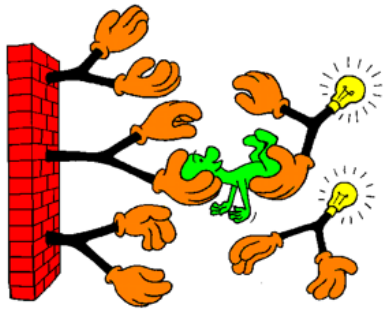

- ✓ Metabolomics in clinical chemistry laboratories

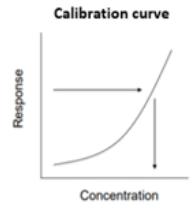
- ✓ Toward point of care tests for metabolomics

# Metabolomics and proteomics in clinical chemistry laboratories

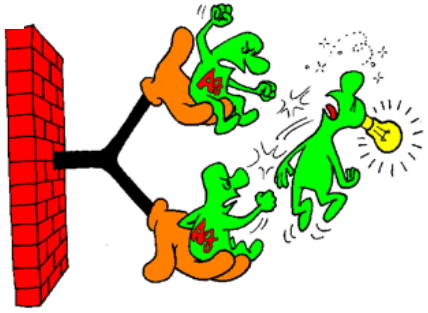
- ✓ **Quantitative liquid chromatography coupled to mass spectrometry assays**
- ✓ **Enzyme assays**
- ✓ **Immunoassays**

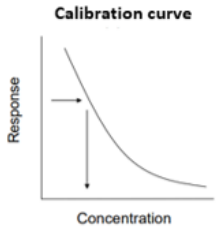
Immunometric format





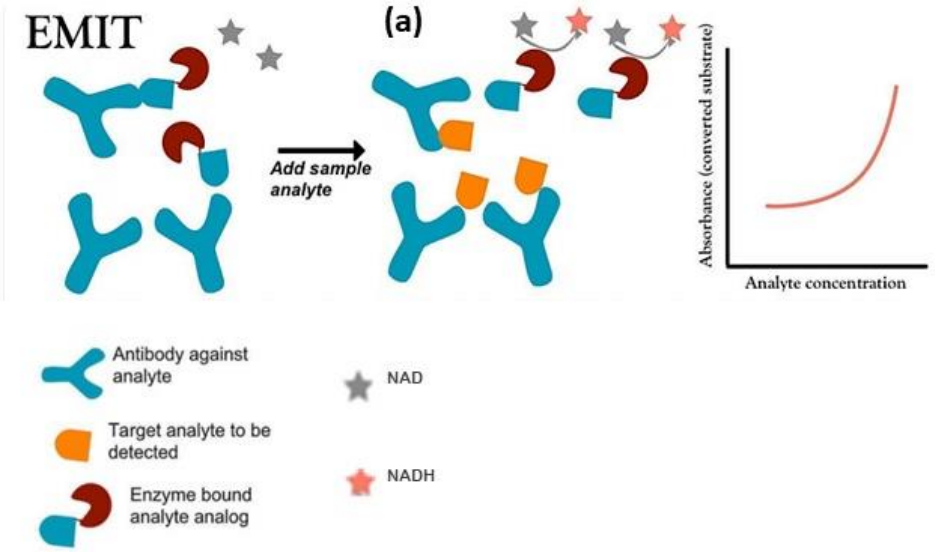

Competitive format (small molecules)



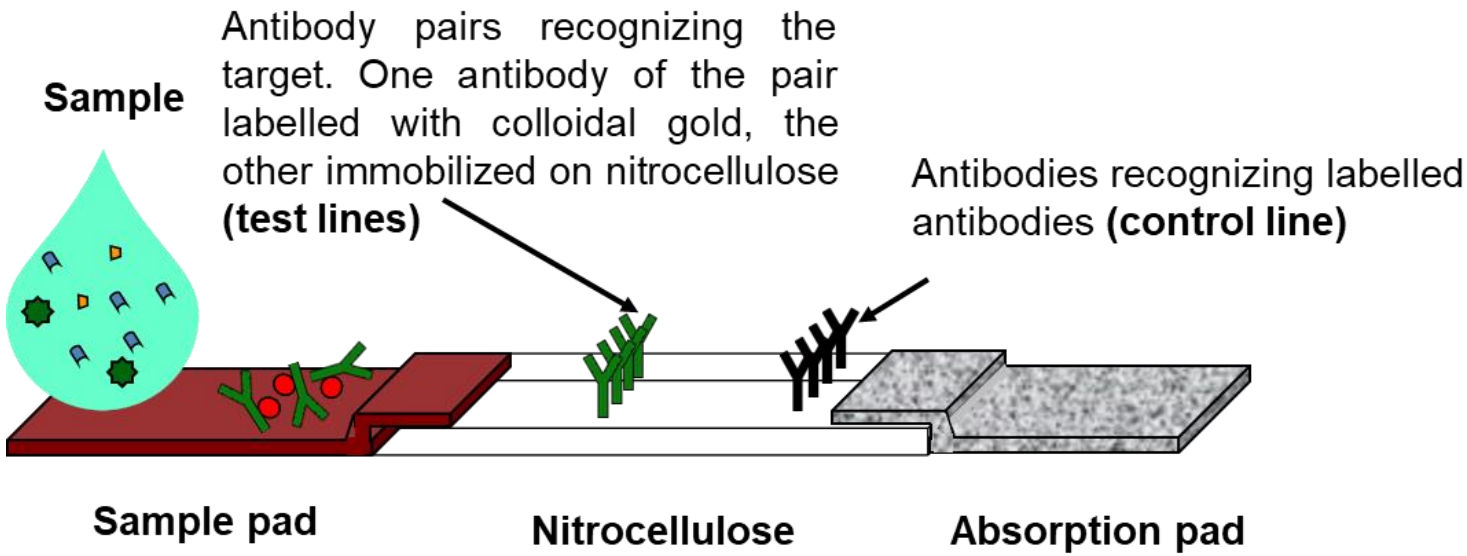



Drawing from Jean-Claude Lardrot

Immunoassays dedicated to small molecules



# Toward point of care tests for omics: lateral flow immunoassays



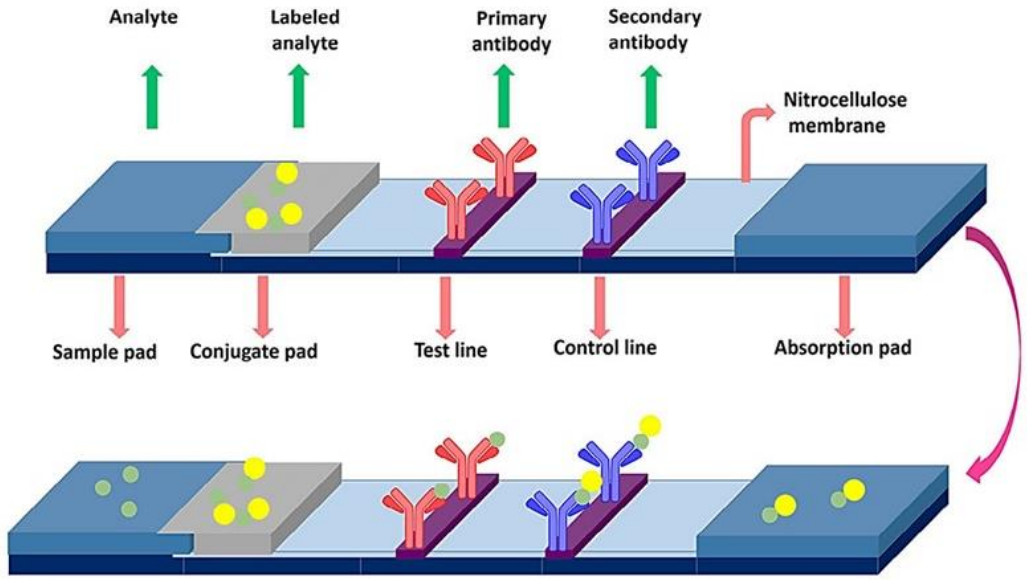

**Low price, easy to use**

**BUT**

**No absolute quantification**

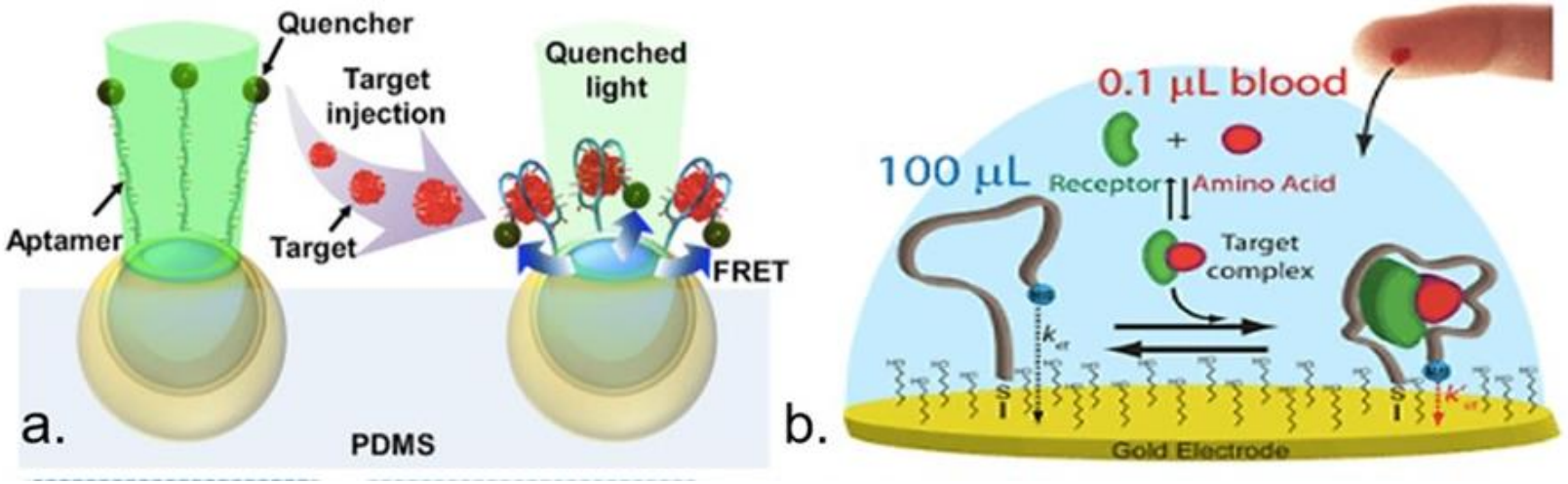
**Low multiplexing capabilities**

Non competitive lateral flow immunoassay for small molecules



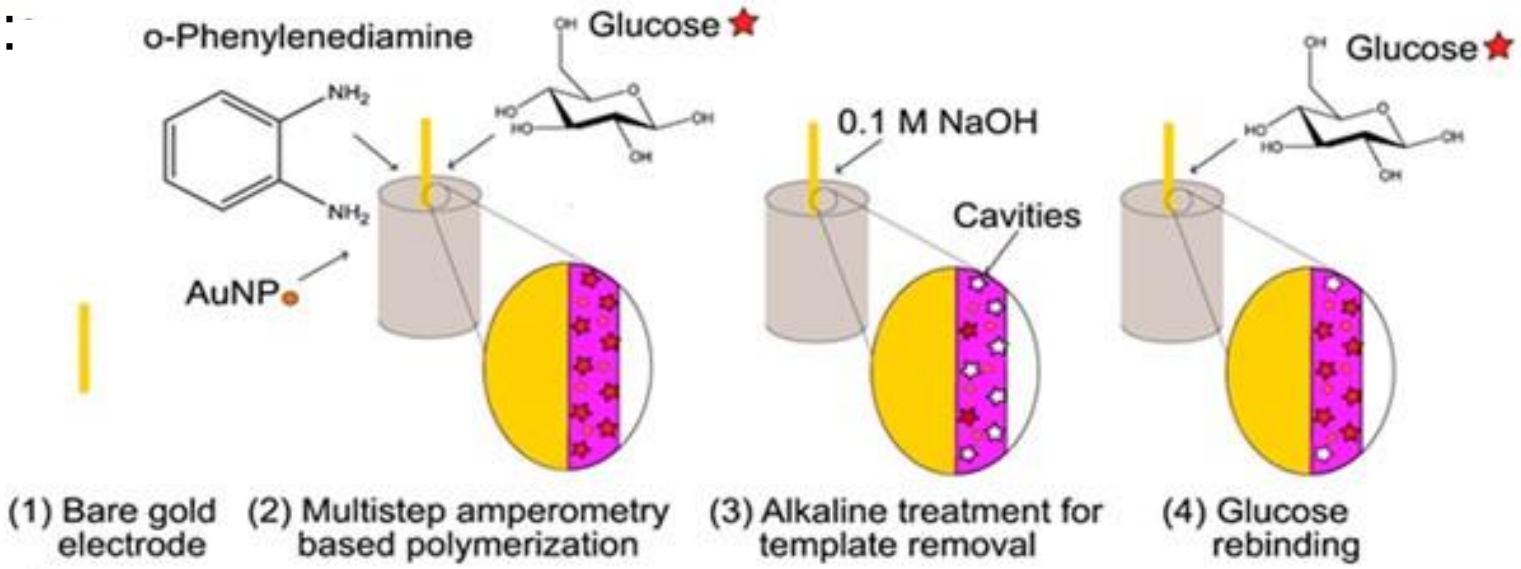
# Toward point of care tests for metabolomics: biosensors

Aptamers




Transduction/detection:
Electrochemistry +++

MIP: Molecularly Imprinted Polymers

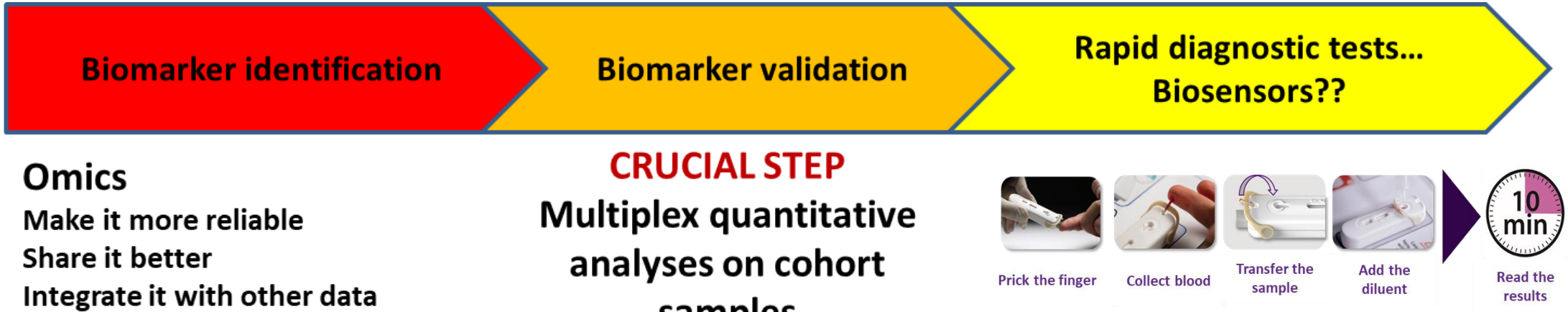



(Castelli et al., Anal. Bioanal. Chem., 2021)

# Take home message

**Metabolomics tools** (data acquisition and treatment) **are constantly improving** and are nowadays used in the field of personalized medicine

Improvement of data interoperability and reusability of untargeted metabolomic data is a key priority, which has to be addressed in a context of permanent and rapid technological evolution.



# Many thanks to my colleagues





# And to you for your attention