



HAL
open science

Overview of metabolomics directed sample preparation strategies for a range of biological matrices

Anne-Lise Royer, Yann Guitton, Gaud Dervilly, Jean-Philippe Antignac, Antoine Escourrou, Nora Cesbron, Karinne Pouponneau, Fabrice Monteau, Bruno Le Bizec

► To cite this version:

Anne-Lise Royer, Yann Guitton, Gaud Dervilly, Jean-Philippe Antignac, Antoine Escourrou, et al.. Overview of metabolomics directed sample preparation strategies for a range of biological matrices. SMMAP, Oct 2017, Marne-La-Vallée, France. 2017. hal-04262576

HAL Id: hal-04262576

<https://hal.science/hal-04262576v1>

Submitted on 27 Oct 2023

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.

OVERVIEW OF METABOLOMICS DIRECTED SAMPLE PREPARATION STRATEGIES FOR A RANGE OF BIOLOGICAL MATRICES

A.-L. ROYER, Y. GUITTON, G. DERVILLY-PINEL, J.-P. ANTIGNAC, A. ESCOURROU, N. CESBRON, K. POUPONNEAU, F. MONTEAU, B. LE BIZEC






LABORATOIRE D'ÉTUDE DES RÉSIDUS ET CONTAMINANTS DANS LES ALIMENTS (LABERCA)
USC INRA 1329, Oniris, LUNAM Université, BP 50707, 44307 Nantes Cedex 3, France
Fax : +33 (0)2 40 68 78 78 - Tél : +33 (0)2 40 68 78 80
laberca@oniris-nantes.fr




Metabolomic approach can be summarized in the form of a generic workflow from experimental design, sample preparation, generation of metabolic profiles, reprocessing of raw data and statistical analysis to the biological interpretation of data.

In a metabolomic analysis, research question, choice of the matrix are studied however it's also important to examine the detail of the process; the collection technique, the type of tube, etc. These parameters are essential because they can be source of variability and interpretation errors.

My Checklist Experimental design :

- ✓ Research question ?
- ✓ Number of samples : 
- Ensuring statistical power, anticipating realization of a pre-test
- Choice of biological matrix : 
- ✓ Depending on the research question
Invasiveness or non-invasiveness
- Conditions of experimentation : 
- ✓ Animals locale
- Food management, ...
To collect the physicochemical parameters, behaviour, disease, ...
- Collection and storage of samples :
- ✓ Collection technique, homogeneity of collection
- Materials used for collection: contamination, ...
- Make of aliquots : allows limit thawing and freezing cycles
- Quenching ?
- Storage at -20 °C or at -80 °C

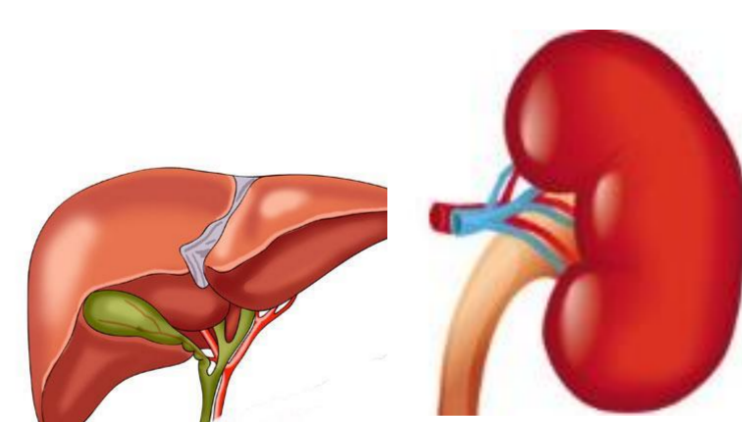
My Checklist Sample preparation :

- Targeted or non-targeted ?
- ✓ . Compromise between the overall view of metabolism and quality of data
- . What fraction of interest?
- . Type of preparation :
Filtration
Extraction with an organic solvent
SPE (Solid Phase Extraction)
- Conditions of preparation : 
- ✓ Control the thawing step
- Glass, plastic container
- Use of filter cones for sampling
- Keeping temperature close to zero during sample preparation
- Preparation of the samples according to the injection sequences
- Ensuring data quality :
- ✓ . Quality Control (QC)
- . Internal standard and/or external standard
- . Blank sample

Specificity according to matrices



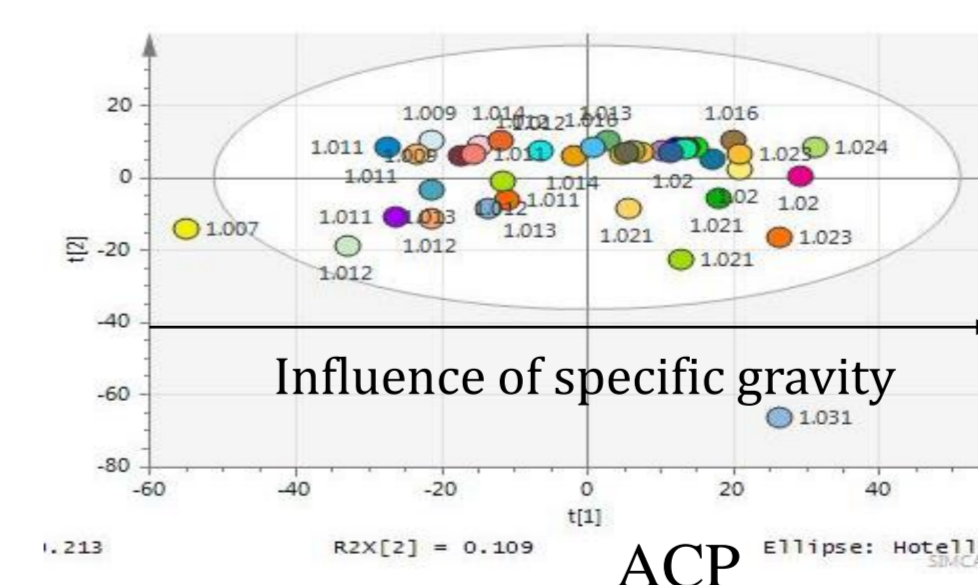
Blood requires collection of anticoagulant such as EDTA, which can increase the analytical variability by transformation of the blood matrix into plasma matrix.^[1]



A solid matrix implies adding a homogenization step of the sample. For this purpose, a grinding device (Precellys, Retsch or other manufacturers) is regularly used before extraction.

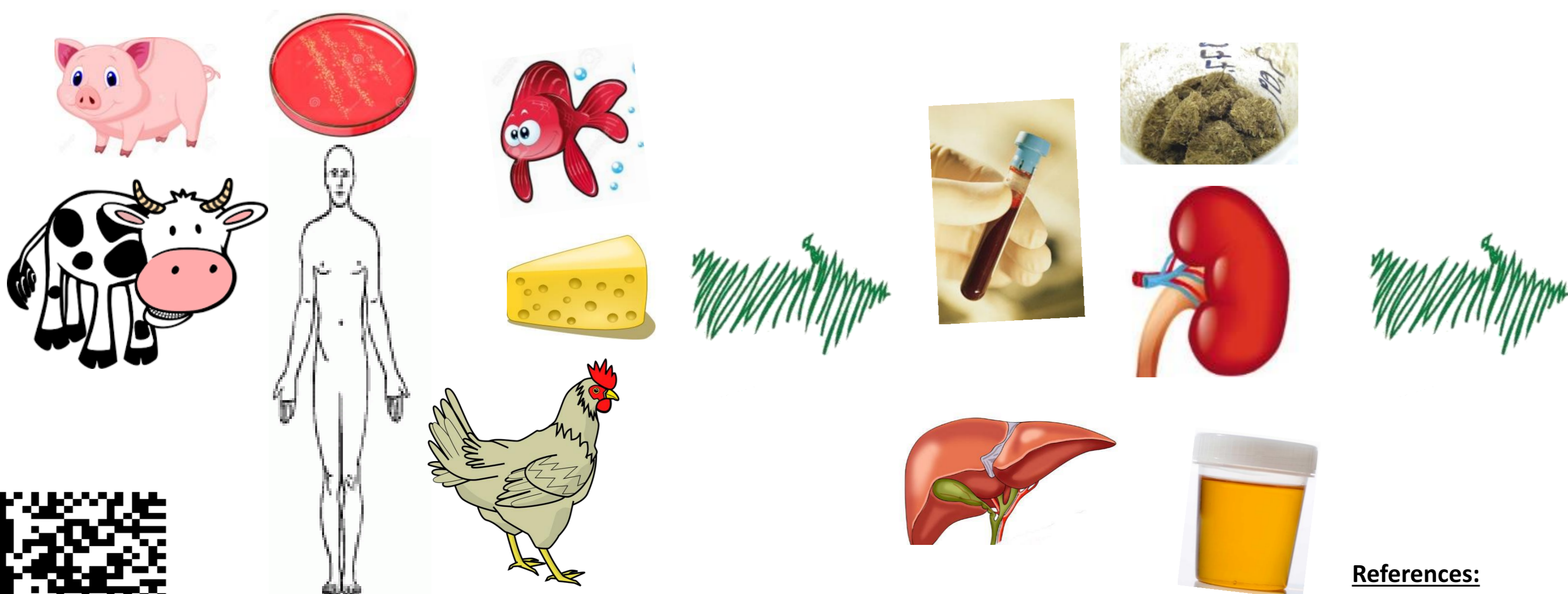


Urine matrix is strongly impacted by the consumption of water and the physiological factors which can cause a more or less dilution of the urine^[2]. In statistical analyzes, this phenomenon may involve separation according to dilution and not according to the research question.



In order to minimize this effect, a standardization step of samples is regularly performed, freeze-drying or specific gravity.

And, what do we analyze in our platform



Extraction for Metabolomic and Lipidomic analysis

- + 190 µL MeOH (vortex 20s)^[3]
- + 380 µL Chloroform (vortex 20s)
- + 120 µL Water (vortex 20s)
- Centrifugation (8000g, 10 min, T 10°C)

Extraction for Lipidomic analysis

- + 220 µL MeOH (vortex 10s)
- + 750 µL MTBE (vortex 10s)
- + 188 µL Water (vortex 20s)
- Centrifugation (1400g, 2 min, T 10°C)

Extraction for Metabolomic analysis

- + 1400 µL MeOH (vortex 15s)
- Centrifugation (15800g, 20 min, T 10°C)

References:

- [1] Dunn, W.B., et al., Nature Protocol, 2011, 1060-1083.
- [2] Royer, A.L. et al, Spectra Analyse, 2017, n°314,
- [3] Bligh, E.G., and Dyer, W.J., Can. J. Biochem. Physiol., 1959, 37(8):911-7.