



HAL
open science

New method to characterize epidermal Ceramides by Supercritical Fluid Chromatography (SFC) coupled to High Resolution Mass (Q-Tof) : Application to various samples with skin barrier defect. Conclusion & outlooks Skin ceramides extraction SFC-QTOF analysis Data Processing Ceramides characterization by MS/MS acquisition

Cyrielle Clement, Julia Soullier, Roselyne Gautier, Carla Orlandi, Emilien Jamin, H el ene Duplan, Carine Jacques, Nathalie Jonca, Pauline Le Faouder, Justine Bertrand-Michel

HAL Id: hal-04338851

<https://hal.science/hal-04338851>

Submitted on 12 Dec 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 destin ee au d ep ot et  a la diffusion de documents scientifiques de niveau recherche, publi es ou non,  emanant des  tablissements d'enseignement et de recherche fran ais ou  trangers, des laboratoires publics ou priv es.

► **To cite this version:**

Cyrielle Clement, Julia Soullier, Roselyne Gautier, Carla Orlandi, Emilien Jamin, et al.. New method to characterize epidermal Ceramides by Supercritical Fluid Chromatography (SFC) coupled to High Resolution Mass (Q-Tof): Application to various samples with skin barrier defect. Conclusion & outlooks Skin ceramides extraction SFC-QTOF analysis Data Processing Ceramides characterization by MS/MS acquisition. ILS (International Lipidomics Society) Conference, Aug 2023, Vienne, Austria. hal-04338851

New method to characterize epidermal Ceramides by Supercritical Fluid Chromatography (SFC) coupled to High Resolution Mass (Q-ToF) : Application to various samples with skin barrier defect.

Cyrielle Clément^{1,2}, Julia Soullier^{1,2}, Roselyne Gautier^{1,3}, Carla Orlandi^{1,3}, Emilien Jamin^{1,3}, H el ene Duplan⁴, Carine Jacques⁴, Nuria Pell Vidal⁵, Nathalie Jonca⁵, Pauline Le Faouder^{1,2}, Justine Bertrand-Michel^{1,2}
¹MetaboHUB-MetaToul, National Infrastructure of Metabolomics and Fluxomics, Toulouse, 31077, France, ²I2MC, Universit e de Toulouse, Inserm, Universit e Toulouse III – Paul Sabatier (UPS), Toulouse, France., ³Toxalim (Research Center in Food Toxicology), Universit e de Toulouse, INRAE, ENVT, INP-Purpan, Toulouse, France, ⁴Centre de R&D Pierre Fabre - Centre de Recherche Pierre Fabre, ⁵INSERM, Infinity - Institut Toulousain des Maladies Infectieuses et Inflammatoires

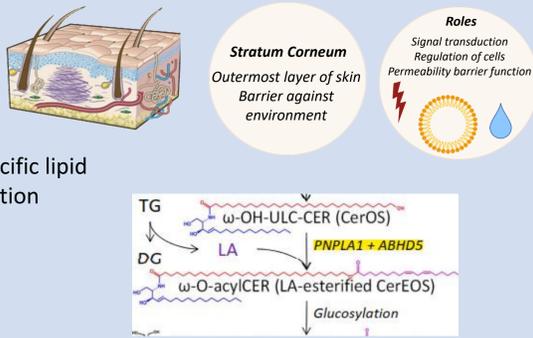
Biological context

✓ Skin ceramide are key partners of epidermal barrier

➢ ω-O-Acylceramide is an epidermis specific lipid with a crucial role for skin barrier function

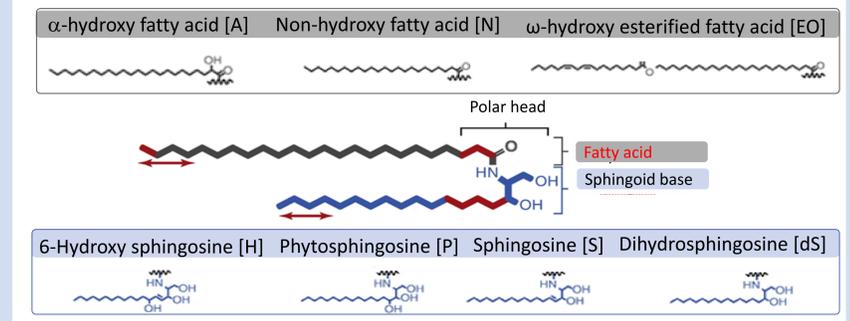
✓ Diseases such as ichthyosis are impacted by ceramide quality

➢ Mutation in patatin-like phospholipase domain-containing 1 (PNPLA1) causes autosomal recessive congenital ichthyosis

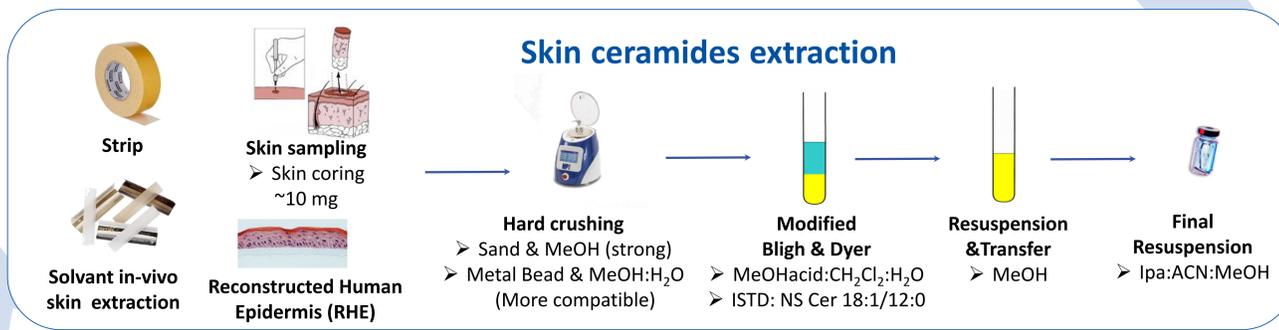


➢ PNPLA1 mutated patients present a default in CerEOS synthesis

Ceramides structures

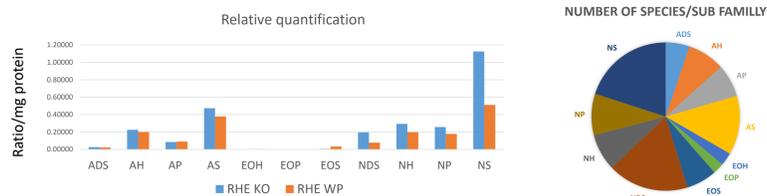


✓ Huge and complex diversity of epidermal ceramides

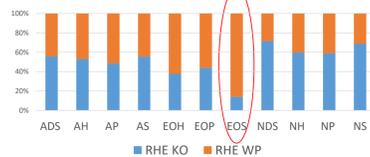


RHE analysis

✓ Analysis of 209 ceramides species in 11 subclasses



✓ Decrease of CerEOS in PNPLA1 KO



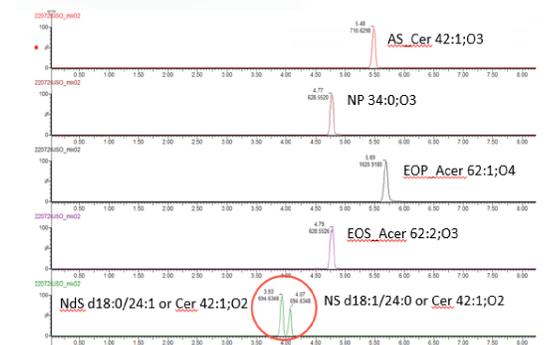
SFC-QTOF analysis

✓ Optimized SFC conditions

Column : Torus DIOL 130Å, 1.7µm, 3 x 100 mm; Waters
A solvent : SCCO₂
B solvent (=Modifier) : IPA:ACN:MeOH 30:30:40 (v/v/v)
 0.1% acid formic
Flow rate : 1.1 mL/min
ISM : Methanol at 0.1 mL/min



✓ Separation by polar heads in 8min

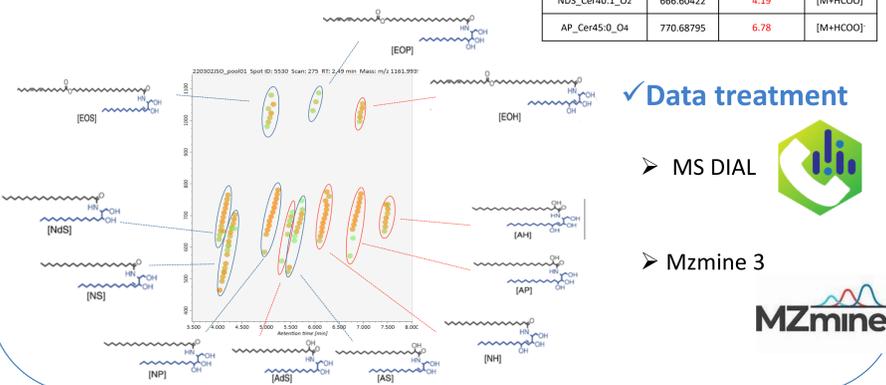


Data Processing

✓ Home made data base:

➢ 280 species for 11 sub-classes

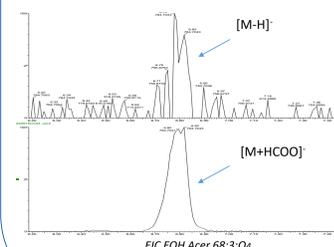
Metabolite	m/z	RT (min)	Adduct
ADS_Cer34:0_O3	600.52088	5.09	[M+HCOO] ⁻
NDS_Cer40:1_O2	666.60422	4.19	[M+HCOO] ⁻
AP_Cer45:0_O4	770.68795	6.78	[M+HCOO] ⁻



Ceramides characterization by MS/MS acquisition

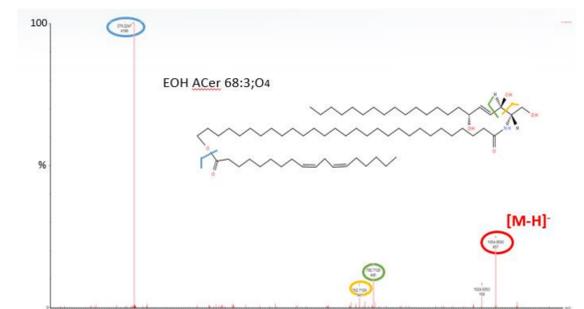
✓ Mass detection

➢ ESI negative mode
 ➢ [M+HCOO]⁻ adduct



✓ MS/MS fragmentation

➢ Validation of MS/MS for all on [M+HCOO]⁻



Conclusion & outlooks

- ✓ Development of a specific and robust method for skin ceramides quantitative profiling using SFC-QTOF
- ✓ Application to RHE sample in the context of ichthyosis diseases
- ✓ Need to develop the analysis of Cer EOS precursor: CerEOS
- ✓ Continue the optimization of extraction on various sample (i.e Strip)

References

- [1] Smeden et al, Biochimica et Biophysica Acta, 2014 (<https://doi.org/10.1016/j.bbali.2013.10.002>)
- [2] Smeden et al, Biochimica et Biophysica Acta, 2014 (<http://dx.doi.org/10.1016/j.bbali.2013.11.006>)
- [3] Jamin et al, EJMS, 2018 (DOI: 10.1177/1469066718815380) Forum lipidomic meeting, ILS, Vienna, 2023

Acknowledgments

R egion Occitanie
 Pierre Fabre Dermo-cosm etique
 Inserm

