

Quantification of bacterial lipids produced by the gut microbiota: role in visceral pain



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Chronic abdominal pain associated with transit modification and alteration of the quality of life

5-10 % of the world's population affected

Irritable bowel syndrome (IBS)

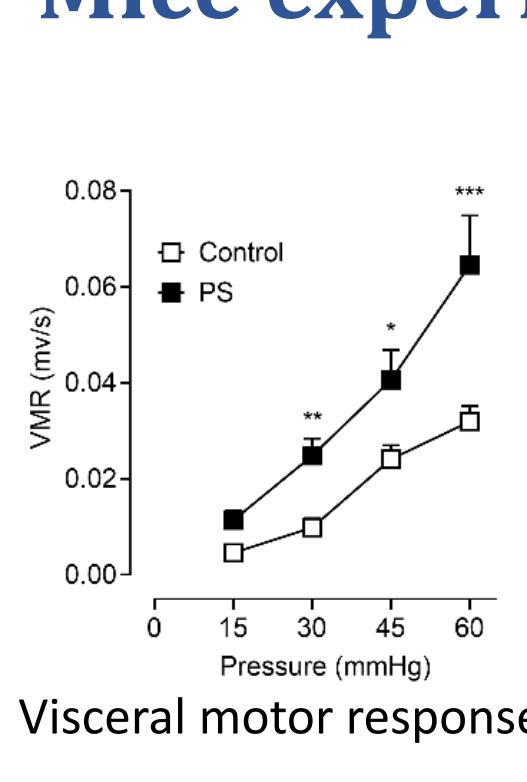
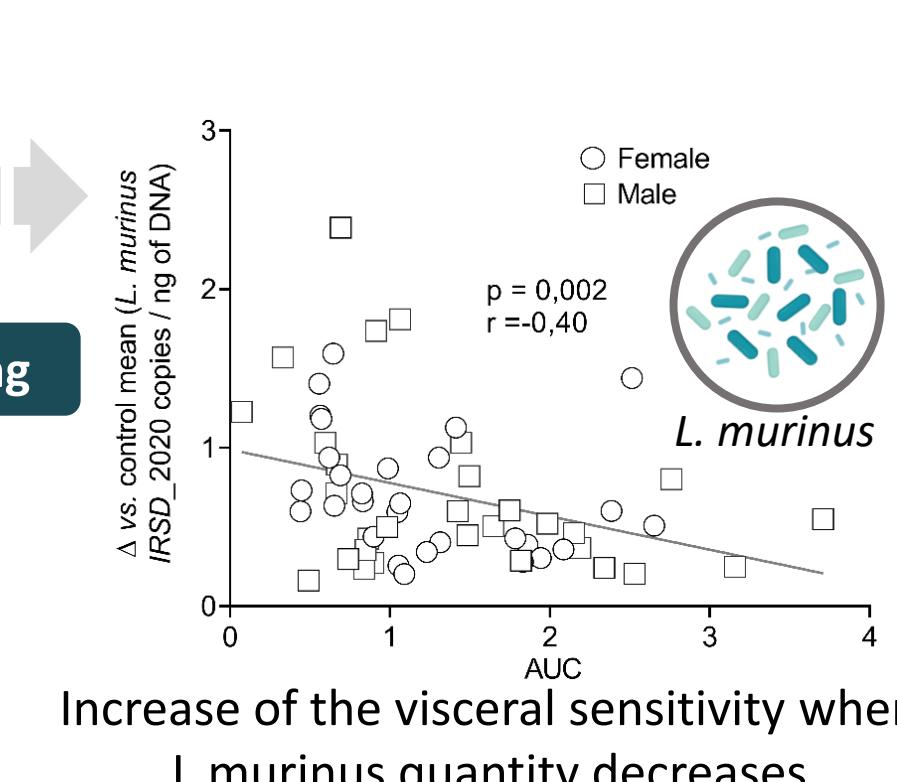
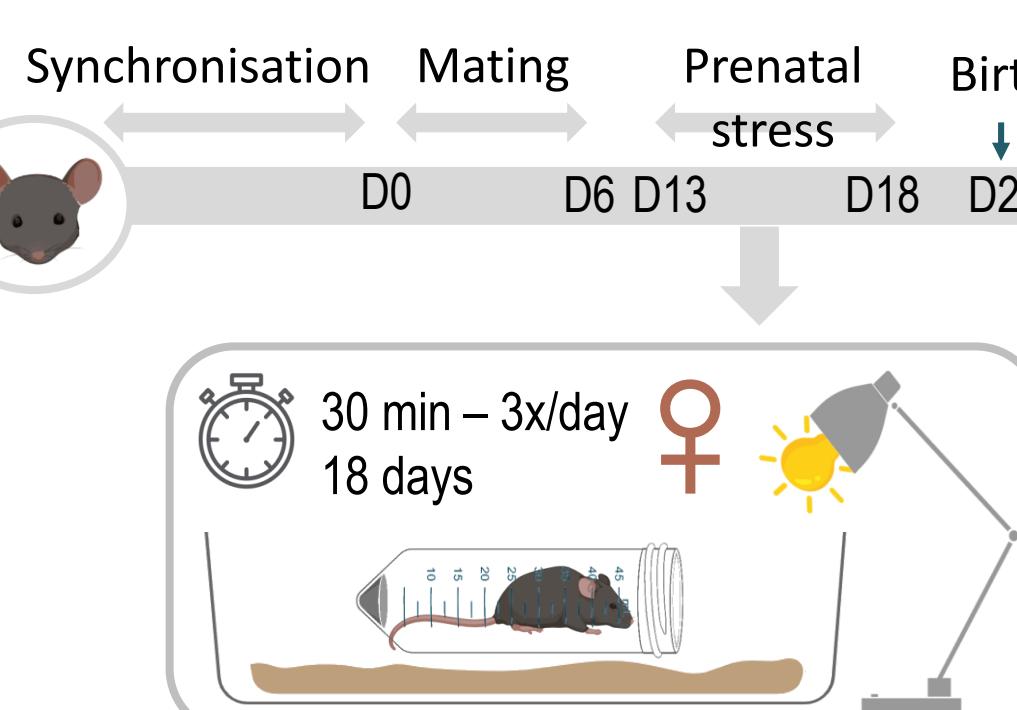
Comorbidity : stress, anxiety and depression aggravates and induces symptoms of IBS

Altered quality of social life

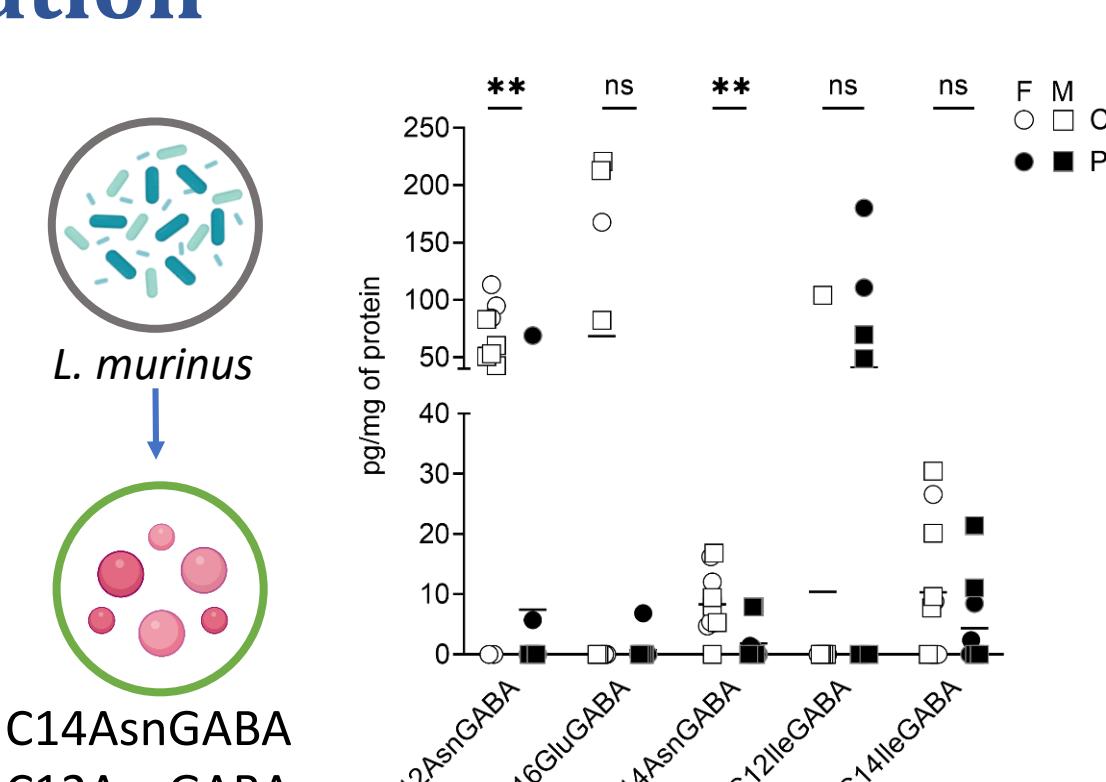


Impairment of the microbiota-gut-brain axis

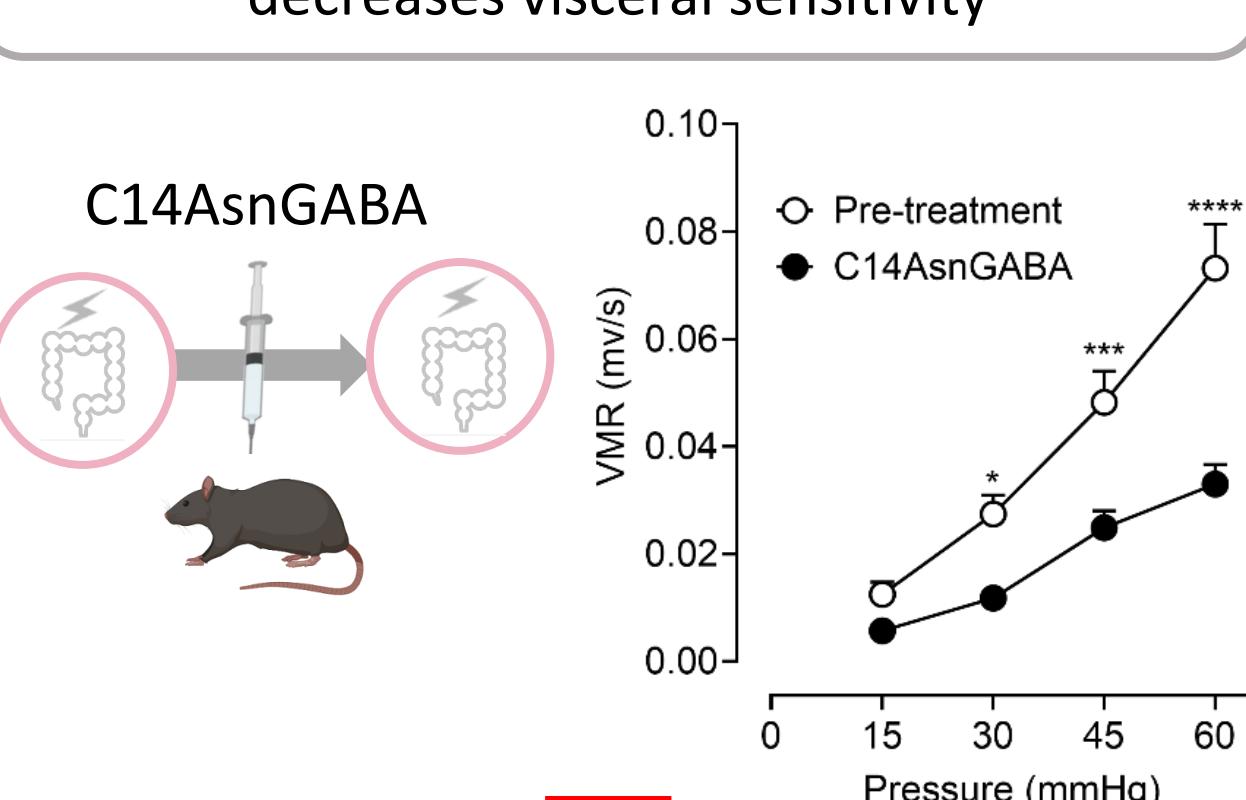
Mice experimentation 1



Increase of visceral sensitivity and taxonomic dysbiosis of prenatal stressed mice offspring



GABA-lipoproteptides decreased visceral pain in IBS mouse model → C14AsnGABA treatment decreases visceral sensitivity



What about IBS humans ?

Visceral pain

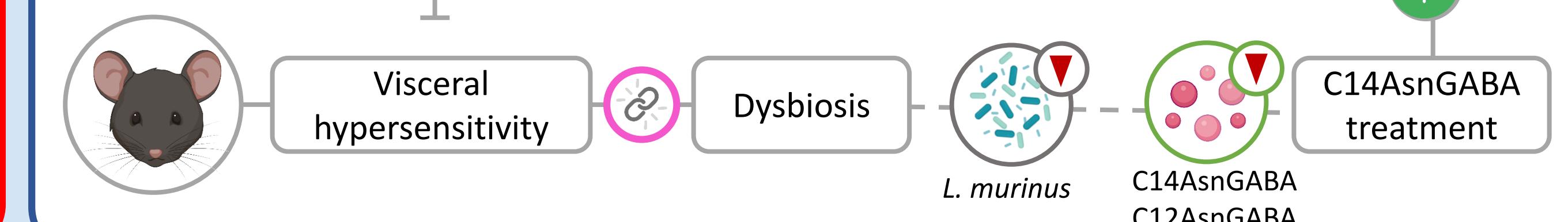


Fonctionnal dysbiosis

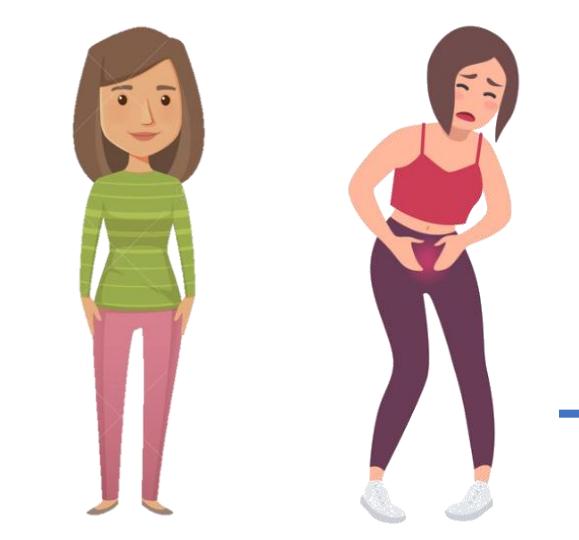


Is there a link between visceral pain and functional dysbiosis ?

Analysis of bacterial lipids in human cohort:
GABA-Lipopeptides, lipoamino acids, short-chain fatty acids and bile acids



Analysis workflow



18 HV 43 IBS-D

15 (75)	32 (78)	Female, N (%)	
31.9 ± 12.1	29.6 ± 8.6	Age, mean ± SD	
7 (16)	Mild IBS, N (%)	75 < IBS-SSS < 149	
18 (42)	IBS-moderate, N (%)	150 < IBS-SSS < 299	
18 (42)	IBS-severe, N (%)	IBS-SSS > 300	

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Lipoaminoacids and GABA-Lipopeptides analysis - UHPLC-MS/MS



Column: C18 CSH, Waters
Solvent A: H₂O, 0.1% formic acid
Solvent B: ACN, 0.1% formic acid
Flow rate: 100 µL/min

Bile acids analysis - UHPLC-MS/MS



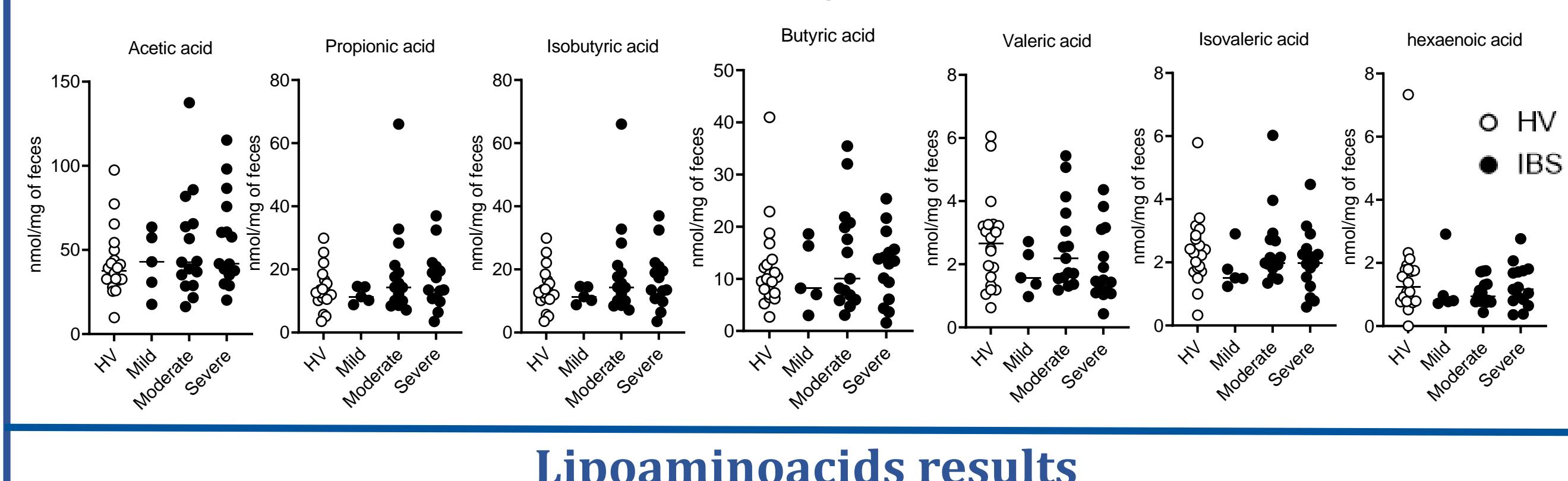
Column: BEH C18, Waters
Solvent A: H₂O, 0.1% formic acid
Solvent B: ACN, 0.1% formic acid
Flow rate: 500 µL/min

Short-chain fatty acids analysis - GC-FID

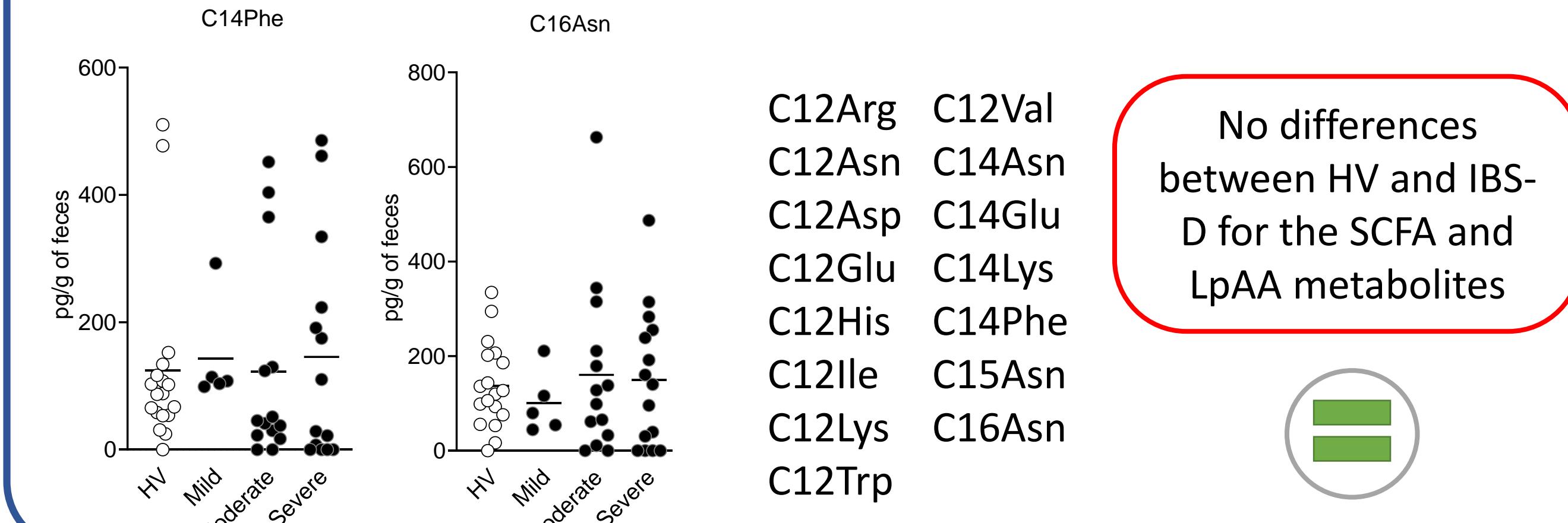


Column: Stabilwax-DA, Restek
Mobil phase: Hydrogen
Flow rate: 1,5 mL/min

Short-chain fatty acids results

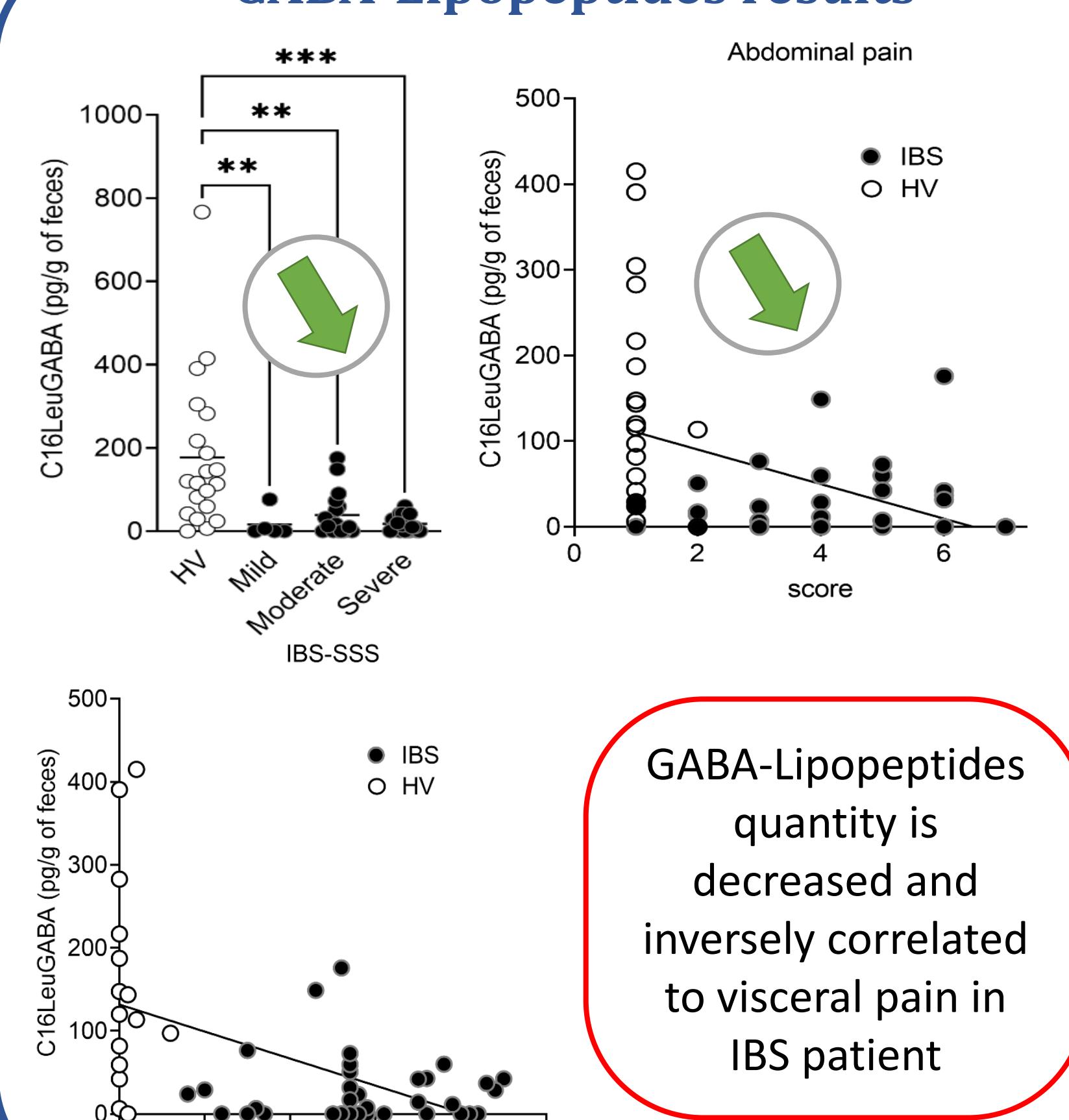


Lipoaminoacids results

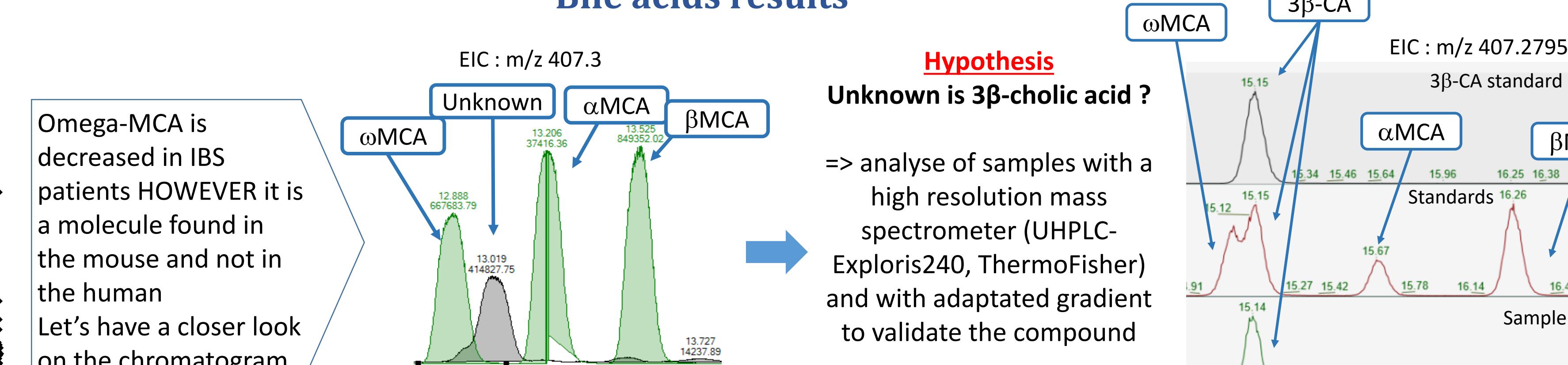
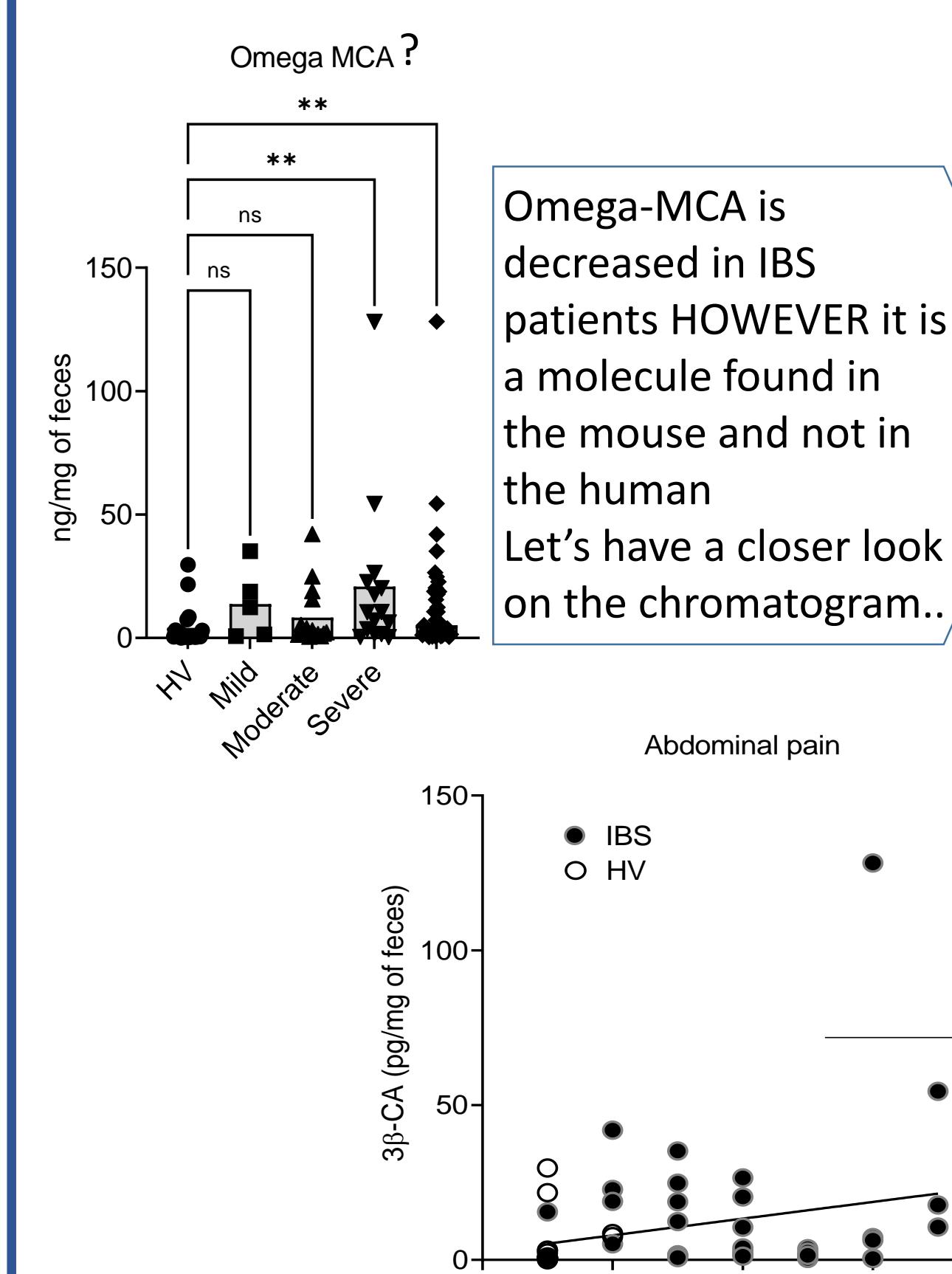


No differences between HV and IBS-D for the SCFA and LpAA metabolites

GABA-Lipopeptides results¹

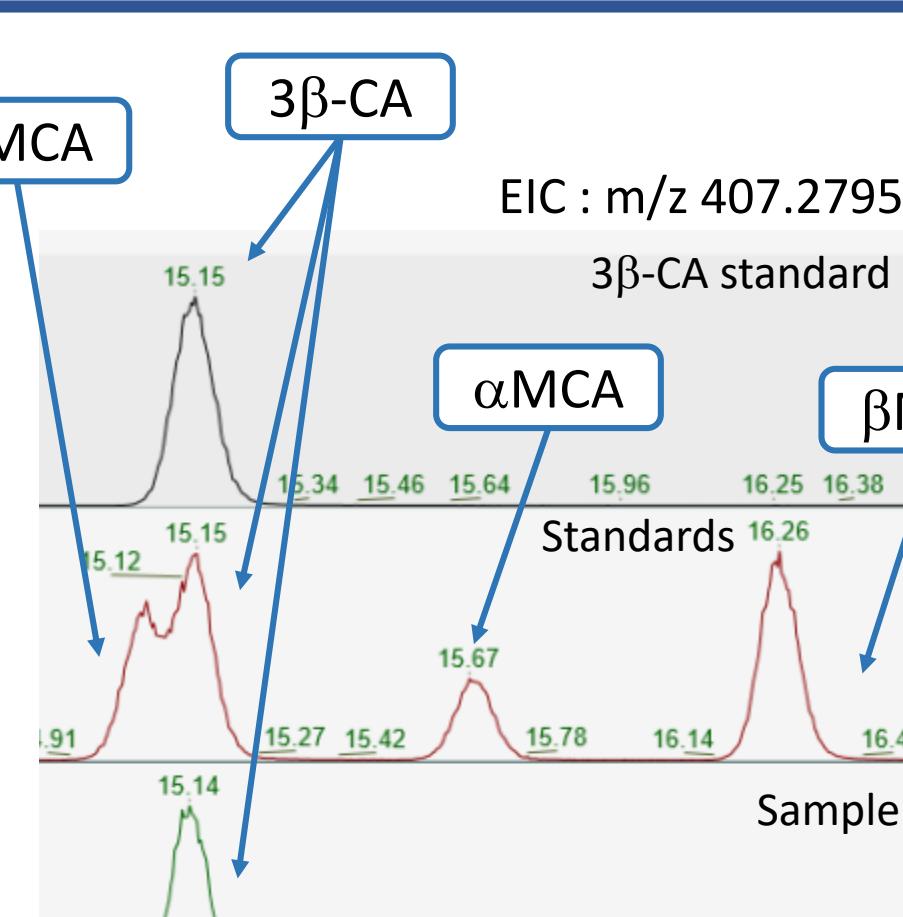


GABA-Lipopeptides quantity is decreased and inversely correlated to visceral pain in IBS patient



Hypothesis
Unknown is 3β-cholic acid ?

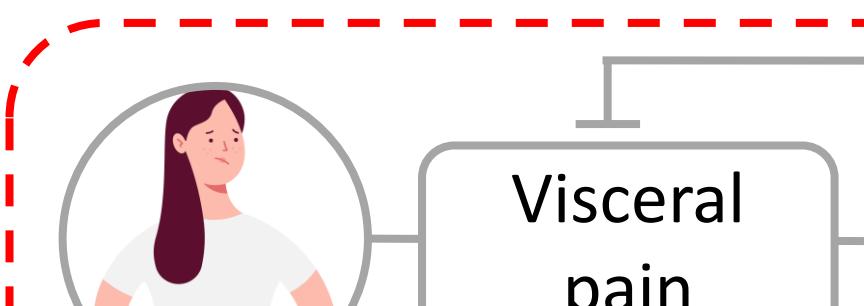
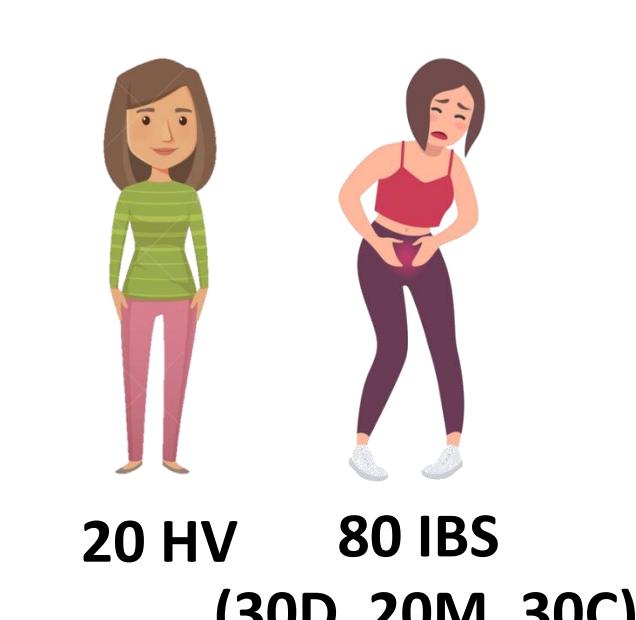
=> analyse of samples with a high resolution mass spectrometer (UHPLC-Exploris240, ThermoFisher) and with adapted gradient to validate the compound



No differences between IBS and HV for primary bile acids and most of secondary bile acids

β-cholic acid quantity is correlated to abdominal pain in IBS patient

Conclusion and Perspectives

20 HV 80 IBS
(30D, 20M, 30C)

- Analysis of SCFA, LpAA, GABA-Lipopeptides, bile acids on diarrhea, constipated and mixed patients
- Addition of oxo-LCA to the bile acids quantification
- Study of the taxonomic dysbiose

References

¹ Petitfils C. et al. Gut 2022

Abbreviations

IBS-D : Intestinal bowel syndrome – Diarrhea
HV : healthy volunteers
LpAA : Lipoaminoacids
SCFA : Short chain fatty acids

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