SELECTIVE MAPPING OF LIPIDS IN THE BRAIN USING DESORPTION ELECTROSPRAY IONIZATION COUPLED WITH MULTIPLE REACTION MONITORING

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INTRODUCTION

Alzheimer's Disease (AD) is the most common type of dementia and represents a significant cause of death and reduced quality of life for large proportions of the elderly population.[1,2] Along with the formation of amyloid plaques and neurofibrillary tangles, lipid metabolism was identified as a hallmark of the disease already by A. Alzheimer.[3] Lipid dysregulation is widespread in the AD brain and reflected in the AD patients biofluids (CSF/blood). [4]

Several changes to the brain's lipid distribution and composition during the progression of AD and other neurological conditions have been determined using a variety of imaging and analytical techniques including shotgun lipidomics, where lipid classes including phosphatidylcholines, ethanolamine plasmalogens and a number of sphingosine based lipids (ceramides, cerebrosides, and sphingomyelin) are dysregulated relative to non-Alzheimer's brain tissue.[5-7]

Desorption Electrospray Ionization (DESI) is an imaging mass spectrometry technique that is used to profile the spatial distribution of lipids and other analytes directly from the surface of a biological sample with minimal sample preparation, thus maintaining the integrity of the analyte distribution.[8]

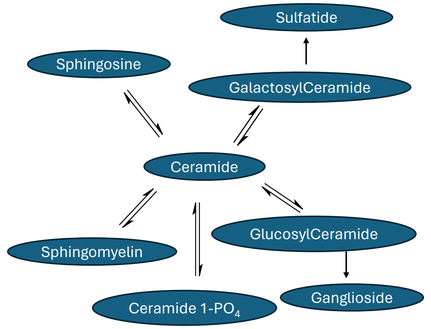

Figure 1. Overview of ceramide metabolism.[9]

METHODS

Flash frozen brain slices (20 micron) from human primary visual cortex from low and high AD subjects (see Figure 4) were mounted on glass microscope slides.

Waters DESI XS Source operated at 50 x50 micron spatial resolution
95% MeOH, 5% H2O, 0.01% Formic Acid with 100 pg/µL Leucine Enkephalin support liquid at 2 µL/min
Nebulizing gas 1 atm Nitrogen
Sprayer Voltage +0.8 kV

Waters Xevo™ TQ Absolute
Operated at unit mass resolution with Argon collision gas.
MRMs optimized on authentic standards.
Data acquisition at 5 pixels per second.
Data processing and visualization using Waters High Definition Imaging (HDI™) v1.8.

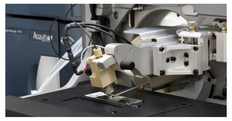

Figure 2. DESI XS ion source mounted on Waters Xevo TQ Absolute mass spectrometer.

Compound Name	Precursor m/z	Fragment m/z	Dwell(sec)	Cone (V)	Collision (eV)
Cer dC18:1 C16:0	538.5	264.3	0.004	15	25
LeuEnk	556.3	397.3	0.004	35	34
Cer dC18:1 C18:0	566.5	264.3	0.004	15	25
Cer dC18:1 C20:0	594.5	264.3	0.004	15	25
Cer dC18:1 C22:0	622.5	264.3	0.004	15	25
Cer dC18:1 C24:1	648.5	264.3	0.004	15	25
Cer dC18:1 C24:0	650.5	264.3	0.004	15	25
HexCer dC18:1 C16:0	700.6	264.3	0.004	30	25
SM dC18:1 C16:0	703.5	184.2	0.004	35	34
HexCer dC18:1 C16:0 +O	716.6	264.3	0.004	30	34
HexCer dC18:1 C18:0	728.6	264.3	0.004	30	34
SM dC18:1 C18:0	731.5	184.2	0.004	35	34
HexCer dC18:1 C18:0 +O	744.6	264.3	0.004	30	34
HexCer dC18:1 C20:0	756.6	264.3	0.004	30	34
PC 34:2	758.6	184.2	0.004	35	25
SM dC18:1 C20:0	759.5	184.2	0.004	35	34
HexCer dC18:1 C20:0 +O	772.6	264.3	0.004	30	34
HexCer dC18:1 C22:0	784.6	264.3	0.004	30	34
SM dC18:1 C22:0	787.5	184.2	0.004	35	34
HexCer dC18:1 C22:0 +O	800.6	264.3	0.004	30	34
HexCer dC18:1 C24:1	810.6	264.3	0.004	30	34
HexCer dC18:1 C24:0	812.6	264.3	0.004	30	34
SM dC18:1 C24:1	813.5	184.2	0.004	35	34
SM dC18:1 C24:0	815.5	184.2	0.004	35	34
HexCer dC18:1 C24:1 +O	826.6	264.3	0.004	30	34
HexCer dC18:1 C24:0 +O	828.6	264.3	0.004	30	34

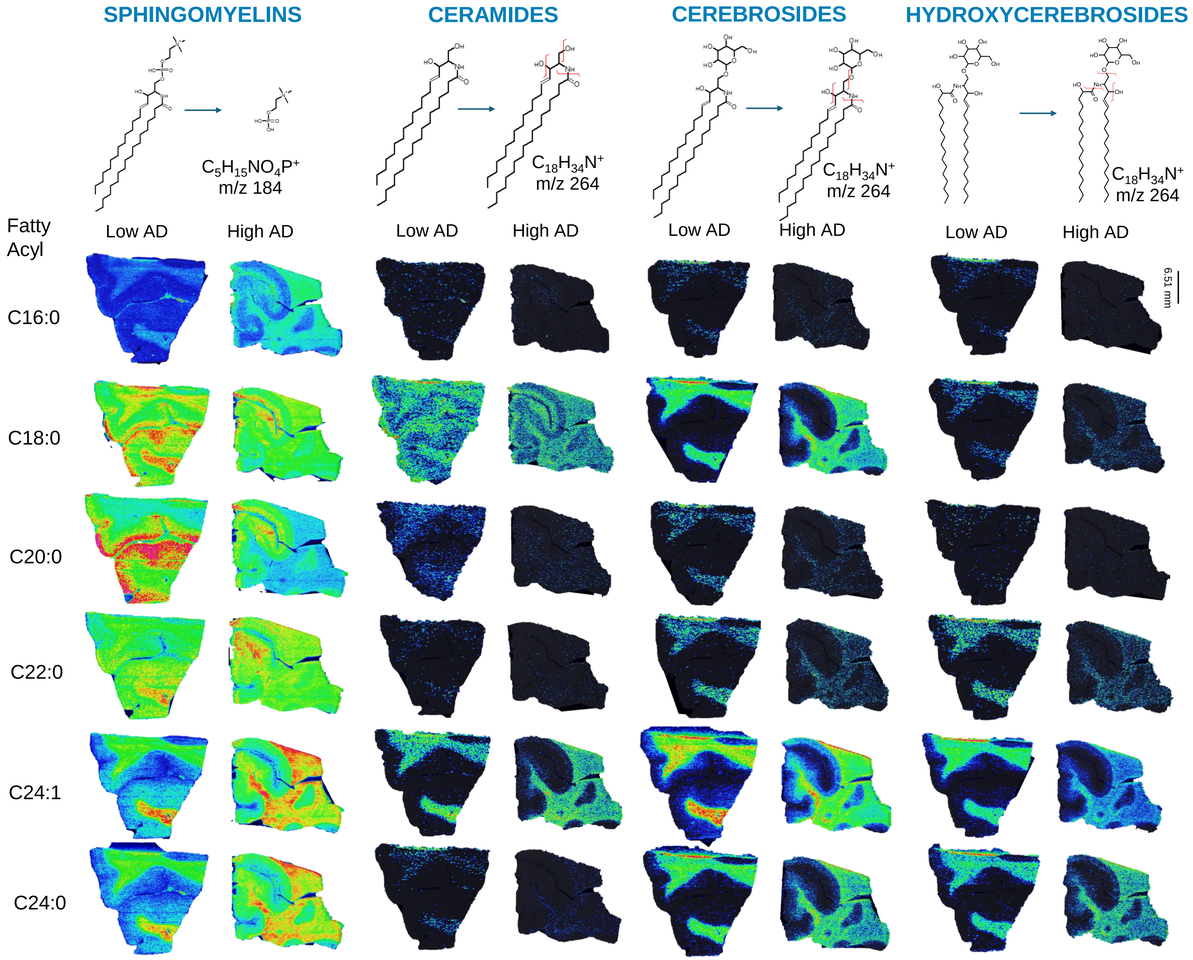

Figure 3. Reconstructed ion images of selected lipids. For all panels, tissue from low AD brain is on the left and high AD brain tissue is on the right. Top to bottom: Lipids with ceramide backbone dC18:1 C16:0, dC18:1 C18:0, dC18:1 C20:0, dC18:1 C22:0, dC18:1 C24:1 and dC18:1 C24:0.

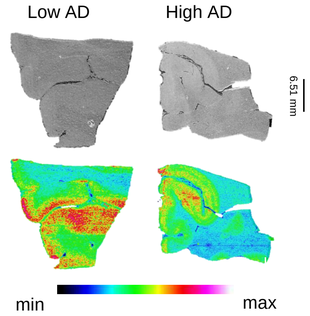

Figure 4. Top panel: Optical image of human brain tissues from the primary visual cortex, Left low AD brain, Right, high AD brain. Bottom panel: Reconstructed ion image for phosphatidylcholine 34:2.

CONCLUSIONS

- **Development of a rapid, sensitive targeted (MRM) assay for brain lipids.**
- **Distribution of phosphatidylcholine C34:2, sphingomyelins, and ceramide dC18:1 C18:0 across brain tissue.**
- **Localization of other ceramides and cerebrosides in the white matter.**
- **First results on human AD brain sections demonstrate sensitivity to detect tissue lipid changes with spatial resolution, paving the way to detailed spatial analyses and correlations with other disease pathology markers.**
- **Extension of study to investigate additional lipid classes, including plasmalogens, lysophosphatidylcholines, sulfated cerebrosides and gangliosides.**

References

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