Introduction

• Huntington’s disease (HD) is a fatal neurodegenerative disorder resulting from a genetic defect.
• Metabolic studies of HD have had low sensitivity and have not been applied to pharmaceutical therapeutics.
• The use of MALDI as an analytic and imaging technique will allow spatial localization and high-sensitivity imaging in order to formulate the link between metabolic disruption and toxic gain of function.

Objective of the Study
Understanding which cellular metabolic pathways are altered, how they are altered, and which are most important for pathogenesis can help us understand why mutant huntingtin confers HD and how to prevent the disease from progressing to the symptomatic phase.

Results

Method Development
• After testing each MALDI matrix with a base set of 39 standards in a 1:1 ratio with the matrix, NEDC and DAN were the most consistent and thorough for detection of small metabolites, while CHCA was the best matrix for large metabolites.
• The most optimal ratio of NEDC:9AA for the greatest ion detection is 6:4.
• Spraying matrices at a temperature above 80°C provides the most consistent application and smallest crystal size.

Detection of Cholesterol
• Beginning with the method outlined in M. Santivañez-Veliz et al. 2017, a Gas Chromatography coupled Mass Spectrometry (GCMS) method for detecting cholesterol and its precursors was optimized.
• GCMS detection can then be used in parallel with MALDI and immunohistochemistry.

Antibody Markers
• Tested six different antibodies for efficacy in WT mouse brain tissue: SDHA, IDH1, G6PD, TH, FA Synthase, GR, CYP27A1.

Discussion
• With an optimized map of each matrices efficacy and range, we can continue to study important metabolites in the HD phenotype—most prominently cholesterol.

Next Steps
• Image antibody staining for both WT and HD brain tissue for comparison now that optimal antibody concentrations have been studied and selected.
• Test the efficacy and range of each matrix using on-slide tissue with MALDI to confirm the results in standards.
• Optimize a spraying method for all matrices.
• Optimize a ratio of DAN:NEDC:CHCA.
• Continue to discern cholesterol defects in HD and use the data to study potential therapeutic targets in the cholesterol synthesis pathway.

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