## Abstract

Early detection markers are critical for neurodegenerative diseases such as Alzheimer's disease, Parkinson's disease, and Huntington's disease. These diseases are known to develop over extended periods of time, making early detection and diagnosis critical for timely treatment and management. This study aimed to identify candidate peptide biomarkers in Alzheimer's disease (AD), Parkinson's disease (PD), and Huntington's disease (HD) using a mass spectrometry-based approach.

### Methods

- **Sample Collection**: CSF samples were collected from the Alzheimer's Disease Neuroimaging Initiative (ADNI) database.
- **Sample Preparation**: CSF samples were processed using the BioFIND platform to identify candidate peptide biomarkers.
- **Proteomic Analysis**: LC-MS/MS analysis was performed on the BioFIND samples to identify differentially regulated proteins.
- **Statistical Analysis**: The significance of the identified biomarkers was evaluated using q-values and false-discovery rates.

### Results

- A total of 1,200 proteins were identified and quantified in CSF. After correcting for multiple testing, 18 proteins were determined to be significantly differentially regulated in at least one of the three neurodegenerative diseases.
- These proteins were categorized based on their functional annotations and were found to be involved in various biological processes, such as neuroinflammation, autophagy, and protein metabolism.

### Conclusion

The identification of these biomarkers provides new insights into the pathophysiology of neurodegenerative diseases and could potentially serve as early diagnostic tools, contributing to improved patient outcomes.

## Targeted MRM to Identify CSF Biomarkers in Alzheimer's Disease

### Background and Goals

- Collaborative partnership with the MRM/Biomarker Consortia
- Overall goal: to study a proteomics-based approach for the identification of novel CSF AD markers

### Study Design and Overview

A large list of candidate biomarkers (Table 1) was identified and analyzed as an early diagnostic measure of disease progression and/or a measure of drug efficacy in clinical trials.

### Biomarker Discovery in Parkinson's Disease

**Background and Goals**

- Supported by the Michael J Fox Foundation
  - Study goal: to identify cross-sectional biomarkers that may serve as early diagnostic markers and/or to confirm the initial results.

### Study Design and Overview

The LC-MS/MS assay identified 500 proteins associated with PD. Elevated peptide levels were found in PD, with some proteins unique to PD and others shared with AD.

### Similiries and Differences Across CNS Diseases

A subset of proteins has been analyzed by MRM in AD, PD, and HD and found to be up- or down-regulated in more than one disease. Shared down-regulated proteins in AD and PD are dominated by neuroprotective hormones (NGF, CCK, SCG), while proteins in HD are more related to inflammation and autophagy.

### Conclusions

- Targeted MRM mass spectrometry provides the means to rapidly and confidently quantify over 200 proteins in a single assay, requiring only 50μl of CSF. Reproducibility of the assay is very high, with the use of stable isotope-labeled peptide standards for every protein peptide being used.
- Differentially expressed proteins were identified in each of the three CNS diseases, providing insight into the underlying biology as well as candidate biomarkers of disease and disease progression. Proteins associated with inflammation, coagulation, and complement were typically up-regulated in all diseases, while proteins involved in neuronal signalling, axon guidance and cellular adhesion were often down-regulated.
- The proteomics profiles of AD, PD and HD were distinct overall but commonalities between pairs of diseases were also found. The current MRM assay has been compiled from all studies and includes proteins found to be significant in each disease. Verification studies are currently underway in AD, PD and HD, all using the same unified assay.

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Table 1: Differentially regulated proteins (q-value < 0.001) in AD, PD, and HD.

<table>
<thead>
<tr>
<th>Protein</th>
<th>FC AD vs HC</th>
<th>p-value AD vs HC</th>
<th>FC PD vs HC</th>
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## Conclusions

The study is different for up-regulated proteins with the observation that significant proteins in PD are similarly regulated in HD. They serve more general functions in inflammation, immune response, and coagulation, proteins expected to be up-regulated in multiple diseases. While other similar proteins were up-regulated in AD, these proteins were not. The study thus highlights the unique proteomic profiles of AD, PD, and HD.

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