HDX-Mass Spectrometry Study of Amyloid beta (Aβ 1-42) Peptide Oligomerization

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Overview

Purpose
- To study the oligomerization of amyloid beta (Aβ 1-42) peptide by using Hydrogen Deuterium Amide Exchange (HDX) coupled with mass spectrometry.

Methods
- HDX kinetic studies show dramatic differences in the state of Aβ at 4 and 25 ºC.
- Variations in deuterium uptake were observed on the peptide level as a function of Aβ oligomerization.

Results
- Several diseases involve the amyloid deposition of proteins and peptides. The Aβ peptide is implicated in Alzheimer’s disease.
- There are several forms of Aβ peptide, of which Aβ1-42 is the most toxic and disease-related.1
- Aβ1-42 sequence (hydrophobic residues in red): DAEFRHDSGYEVHHQKLVFF AEDVGSNKGAIGLMVGVVIA

Introduction

Overview

Material:
- Aβ1-42 peptide was from nPeptide (Bogart, GA); other chemicals from Sigma-Aldrich (St. Louis, MO).

H/DX Exchange (H/DX):
- Aβ1-42 monomer was obtained by dissolving peptide in DMSO at 5 mg/mL. Different forms of oligomers were achieved by varying the incubation time in PBS buffer (pH 7.4, 4 ºC vs. 25 ºC). HDX was done at 4 ºC.
- HDX was quenched by adding 3 M urea and 1% TFA. Peptide-level analysis was achieved by using on-line pepsin digestion at 4 ºC. Resulting peptides eluted by a 9.5 min gradient.

Instrumentation:
- Experiments (# 4 ºC) were on Bruker MaXus Q-TOF while the other set of experiments (25 ºC) were with Thermo LTQ-FT in positive-ion ESI mode, interfaced to Agilent 1100 and 1200 HPLC.

Data analysis:
- All the spectra were processed by MagTran, and results were reported as % Deuterium uptake (%δ vs. δ0).

Results

Aggregation at 4 ºC

Aggregation at 25 ºC

Discussion

- No difference in HDX observed at 4 ºC over long times, indicating that Aβ1-42 does not undergo significant aggregation at this temperature.
- At 25 ºC, however, HDX extent decreases dramatically as time increases, indicating that Aβ1-42 undergoes aggregation with accompanying conformational changes or precursors to protein. We do not know whether Aβ is aggregated in a single conformation or single oligomeric state.

Future Work

- More analysis will be conducted at different temperatures.
- The effects of concentration on Aβ aggregation will be investigated.
- HDX experiment will be coupled with ECD/ETD to provide the residue level information.
- Use HDX to investigate the interaction of Aβ with various compounds.

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References