Scanning x-ray microdiffraction mapping of fibrillar polymorphs in Alzheimer's disease

Lee Makowski[1], Abdullah Al Bashit[2], Prakash Nepal[3]

[1]Northeastern University [2]Northeastern University, [3]Dept. of Bioengineering

l.makowski@northeastern.edu

Alzheimer's disease (AD) is defined by the progressive formation and spread of fibrillar aggregates of Abeta peptide and tau protein throughout the brain. Polymorphs of these aggregates appear to contribute to disease in different ways since different neuropathologies - that follow distinct pathological trajectories - are associated with different sets of fibrillar structures. The molecular mechanisms driving the spread of these aggregates are unknown, but may include nucleation, replication and migration. The relative importance and speed of these processes may vary with polymorphic form, disease stage or region of brain. Different mechanisms of progression are likely to give rise to different spatial distributions of these aggregates. For instance, if fibrillar spread is driven by a prion-like migration, the fibrillar structures seen in an individual brain tissue should be invariant. If independent nucleation events underlie fibrillar spread, then different lesions may contain distinct polymorphs. Therefore, mapping the distribution of polymorphs in situ has the potential to discriminate between different mechanisms of progression. There are few biophysical tools capable of carrying out this mapping. Optical microscopy is incapable of discriminating between polymorphs in situ, and higher resolution imaging modalities such as ssNMR and cryoEM generally require isolation of fibrils from tissue, obscuring relevant spatial information.

To collect information on polymorph structure in a way that preserves information on location of fibrils, we are using scanning x-ray microdiffraction (XMD) of thin sections of human brain tissue. X-ray scattering from fibrillar aggregates fixed in thin tissue sections includes contributions from fibrils; the polymeric matrix in which the fibrils are embedded; other constituents of the tissue; and cross-terms between fibrils and neighboring molecules. This complexity severely limits the information that can be extracted from scattering studies. However, micro- and nano-beams make possible measurement of x-ray scattering from very small volumes which, in neuropathological lesions, may be dominated by a single fibrillar species. In those cases, it may be possible to derive information about the predominant species from the scattering. Coordinated examination of serial sections by immunohistochemistry on serial sections can be used to confirm the identity of the predominant lesion species - Abeta or tau, aiding the interpretation of scattering patterns. Information on the molecular organization within a scattering volume can be derived from small angle scattering (SAXS) whereas information on the structure of individual fibrils can be gleaned from interpretation of the wide-angle scattering (WAXS).

The spatial correlations between the positions of fibrils and other constituents of a scattering volume have significant impact on the SAXS data from these samples and analysis of this scattering can provide information on the organization of fibrils in these tissues. By contrast, WAXS is dominated by scattering from individual fibrils and is not significantly impacted by cross-terms due to correlation between fibril positions and the surrounding polymeric matrix. This makes WAXS a potential source of information about the identity of polymorphs within a lesion. Lesions rich in either Abeta fibrils or tau proteins exhibit wide-angle scattering with a prototypical 4.7 Å cross-beta peak. But the shapes of these peaks are different for Abeta-containing lesions compared to tau. In many cases, the shape of the 4.7 Å peak corresponds well with that predicted from high resolution structures, be it from Abeta or tau, making possible positive identity of polymorph.

In summary, these observations demonstrate that XMD is a rich source of information on the organization and spatial distribution of fibrillar polymorphs in diseased human brain tissue. When used in coordination with neuropathological examination it has the potential to provide novel insights into the molecular mechanisms underlying disease.