Insulin stability and fibrillation with surfaces: Role of surface wettability

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A molecular explanation of the structural path from a native protein to an amyloid fibril consisting of antiparallel β-sheets in a cross-β-sheet arrangement has remained elusive due to the difficulty in obtaining intermediates for crystal X-ray analysis. Using a recently developed algorithm with attenuated total reflection infrared spectroscopy, circular dichroism and UV-Vis absorbance assays, and human insulin hormone as a model protein, we have followed the loss of α-helix and gain in β-sheet as a result of exposure to a series of model surfaces differing in wettability.

First, we investigate the secondary structural conformational stability of native insulin adsorbed from solution under physiological conditions onto several synthetic membrane surfaces for 12 h, and for different times on a hydrophobic model membrane surface - poly(tetrafluoroethylene) (PTFE). The results demonstrate that native insulin unfolds when adsorbed onto solid substrates, especially hydrophobic surfaces such as PTFE. Two-phase kinetics is observed, where conversion of α-helix to random and turns occurs at very short times (< 1 min) and then, after many hours, to β-sheet. Then, we investigate the fibrillation process of insulin (pH 1.6 and 65ºC) in the presence of several materials, characterized by their surface energies, and commonly used as catheters or other invasive devices. We also show that materials like poly(tetrafluoroethylene), polyethylene, polypropylene commonly used for microfiltration and in catheters speed-up fibril formation. Hydrophilic membrane materials such as regenerated cellulose exhibit the opposite effect. These hydrophobic-induced fast transitions provide a starting point for a molecular explanation of the structural path from native protein to amyloid fibril based on aromatic stacking and charge attraction of the di-peptide FV (amino terminus) with the hexa-peptide RGFFYT (carboxyl terminus) in insulin.