How large are changes in protein structure needed to promote aggregation?

Leonardo de Castro Palmieri[1], Luis Mauricio Trambaioli da Rocha e Lima[2], Igor Polikarpov[3], Juliana Batista Barros Freire[1], Débora Foguel[1]

[1]Instituto de Bioquímica Médica, UFRJ; [2]Faculdade de Farmárcia, UFRJ; [3]Instituto de Física de São Carlos, USP, Brazil.

Amyloidogenic diseases, which include Alzheimer's and Parkinson's, are characterized by the deposition of amyloid aggregates in a specific tissue resulting in cellular damage and death. Hitherto, besides amyloid deposition, the other characteristic shared by most of the amyloidogenic proteins is the presence of a misfolded intermediate, which seem to be the raw material for aggregation. For several of these proteins, this intermediate has yet been structurally characterized; for others, this intermediate is too ephemeral, what compromise further structural characterization. In this work, we have studied wild-type transthyretin (WT-TTR), a homotetrameric protein of ~55kDa involved in thyroxin and retinol transport in plasma and in cerebrum-spinal fluid and a monomeric, engineered mutant of this protein (M-TTR) in an attempt to characterize the structure of the aggregation-prone intermediate formed under mild-acidic condition. We have been using denaturant agents such as temperature, high pressure, urea, guanidine and pH shift to show that the aggregation prone intermediate of TTR has only subtle changes on its tertiary and secondary structures as compared to the native protein as seen by fluorescence and circular dichroism. These small changes in the structure were also evaluated by comparing the crystal structure of WT-TTR and M-TTR at pH 4.6 with that observed at pH 7.0. In conclusion, at least in the case of TTR, aggregation uses an intermediate which is almost native-like.