# Temperature mediated Aggregation of Reduced Lysozyme at physiological pH and its Luminescence Feature

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**Abstract**:

At non native state proteins monomer have a critical tendency to self-associate and forms dimer/pseudodimer, which further drives the aggregation and form an ordered structures in the range of nanoscale known as amyloid fibril[1]. From the past protein aggregation has been considered as consequences of neurodegenerative disease. However recent research has been shown that protein aggregates can be act as biomaterials. Here, we report the characteristic features of aggregates of reduced HEWL at physiological pH 7.2 while incubating at 57 °C for 60 days[1,2,3,4]. The intrinsic fluorescence at longer wavelengths confirms the presence of excited state dimer (excimer) form of aggregates[2]. As the incubation time increases the excimer induces higher degree of aggregation which was evident from the existence of a boarder fluorescence peak at longer wavelength[2,3]. After adding SDS to the reduced HEWL, we have not observed the disassembly of dimer, the nature of excimer peak was being stabilized. The excitation dependent emission spectra confirm the presence of different degree of aggregates. Additionally, Circular Dichroism (CD) spectroscopy experiment reveals that the protein loses its secondary structure upon reduction induced by Dithiothretol (DTT) and regains a part of the secondary structure upon subsequent treatment with SDS. Our ThT binding assay confirms the presence of ordered aggregates brought in by the added SDS which were otherwise amorphous in nature due to the reduction of the protein[1,4]. AFM micrographs conclusively reveal the morphology of these ordered aggregates which were visualized as marquise and bundle of fibre like structures, primarily induced by SDS. Our steady-state fluorescence results suggest that the π-π stacking interactions within Tryptophan moieties helps in formation of excited state dimer, which in turn leads to the luminescent characteristics of the self-assembled aggregates.

**References and Notes:**

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