The Conjugation of Amyloid Beta Protein on the Gold Colloidal Nanoparticles' Surface

Kazushige Yokoyama
The State University of New York Geneseo College.

Statement of Purpose: The protein coated gold nanoparticles can be used to investigate the conformational change of proteins at a solid-liquid interface. This approach was applied to investigate the conjugation of amyloid β protein (Aβ) over gold nanocolloid in conjunction with a mechanism of fibrillogenesis, which is associated with Alzheimer's disease.

Methods: The ultra-pure Aβ_{1-40} (MW: 4329.9 Da), in the form of lyophilized powder (97% by HPLC) was purchased from American Peptide (Sunnyvale, California, USA), and stored at −12 °C. The stock solution of 100 μM Aβ_{1-40} was prepared at approximately 18 °C using double-distilled deionized and filtered water. Gold colloidal nanoparticles (5 – 100 nm) were purchased from Ted Pella Inc. (Redding, California, USA) The ratio of the particles between the protein and the gold colloidal nanoparticle solution was adjusted to a factor of 1000 more than the gold colloids. The pH of the solution was repeatedly altered between pH 4 and pH 10 for 10 cycles, and the corresponding absorption spectrum was monitored at each pH condition. The temperature of the solution was monitored by a temperature sensor installed in the cuvette holder and was confirmed by a digital thermometer inserted in a sample cuvette for the temperature range between 5 and 50 °C. For TEM (transmission electron microscopy) measurement, Formvar-coated grids were used for preparing samples of Aβ_{1-40} coated gold beads. Formvar was cast onto microscope slides, floated off onto water, and grids were applied to the film. Samples were examined with a Morgagni model 268 TEM (FEI Co., Hillsboro, OR, USA) operated at 80 kV.

Results: Among tested Aβ proteins and various sizes of gold colloids, only Aβ$_{1-40}$ coated 20 ± 1 nm gold colloidal nanoparticles exhibited a reversible color change as the pH was externally altered between pH 4 and 10. (See Figure 1) This size selective reversibility is an important implication for the initial reversible step reported for the fibrillogenesis of Aβ$_{1-40}$. When the process was repeated with ovalbumin, all tested sizes of gold colloidal nanoparticles (10, 20, 30, 40, 50, 60, 80, and 100 nm) showed a quasi reversible color change, implying that the conjugation process is partially dependent on the protein. Temperature dependent features were observed in the reversibility of Aβ$_{1-40}$ conjugated to 20, 30, and 40 nm gold colloidal suspension. While Aβ$_{1-40}$-coated 20 nm gold colloid nanoparticles exhibited a reversible color change under 50 °C except for 5 ± 0.2 °C and lower, Aβ$_{1-40}$-coated 30 and 40 nm colloids exhibited the reversible color change when temperature was lowered to 18 ± 0.2 and 6 ± 0.2 °C, respectively. (See Figure 2)

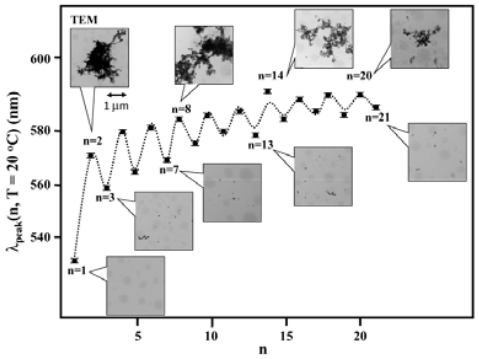

Figure 1. A peak shift (λ_{peak}) as a function of pH changes for Aβ_{1-40}-coated 20 nm gold colloid particles at 20 °C is shown along with the TEM images of Aβ_{1-40}- coated 20 nm gold colloids: n = 1 (pH 7), n = 2 (pH 4), n = 3 (pH10), n = 7 (pH 10), n = 8 (pH 4), n = 13 (pH 10), n = 14 (pH 10), n = 20 (pH 4), and n = 21 (pH 10).

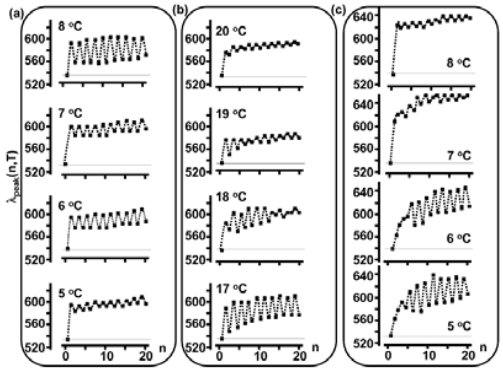

Figure 2. The demonstration of color reversibility of (a) Aβ_{1-40}-coated 20 nm gold particles between 5 and 8 °C, (b) Aβ_{1-40}-coated 30 nm gold particles between 17 and 20 °C, and (c) Aβ_{1-40}-coated 40 nm gold particles between 5 and 8 °C.

Conclusions: Specific and unique size and temperature dependence was observed in reversible color change for Aβ_{1-40} coated gold nanocolloids. This strongly suggests that the noncovalent intrinsic intermolecular potential formed between the nanocolloidal surface and each Aβ$_{1-40}$ monomer conjugated at the surface drives the process. Our series of studies explored conformational changes unique to the surface size of the gold colloidal nanoparticles and opened new insights for protein associated nanomaterials.

References: (Yokoyama K Advances in Nanotechnology, Nova Publisher 2010; 1: 65-104) (Yokoyama K. J Phys Chem A, 2010; 114, 1521-1528) (Yokoyama K. Int J Mol Sci, 2009; 10, 2348-2366) (Yokoyama K. Nano-Tech, 2007;18 105101-105107)