Electronic Supplementary Information (ESI)

Amphiphilic spherical nanoparticles with nitrogen-enriched carbon-like surface by using ß-lactoglobulin as template

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Experimental Section

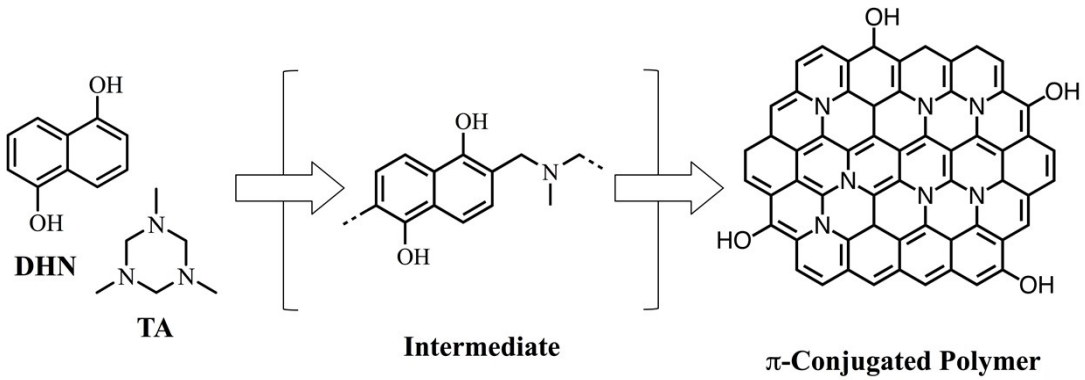
βlg from bovine milk was purchased from Sigma Aldrich (Germany) and was used as received**.** 1,5-Dihydroxynaphthalene (DHN) and 1,3,5-trimethyl-1,3,5-triazinane (TA) were purchased from Tokyo Chemical Industry Co., Ltd. (Tokyo, Japan) and used as received. Ethylalcohol were purchased from Wako Pure Chemical Industries, Ltd. (Osaka, Japan) and used as received.

Synthesis of polymer/carbon-coated nanoparticles*:* 1 wt% βlg protein solution was prepared in 10 ml vials by dissolving the powder in Milli-Q water at room temperature (298 K). Sample vials were gently rotated for 1 h at room temperature to allow the protein to fully dissolve before centrifuged at 10,800g over a period of 15 min at 25˚C. The solution pH was adjusted to 5.0-6.0 with minimal titration (i.e. addition of HCl/NaOH). A relatively high concentration of HCl/NaOH (i.e. 5% w/v) was used in order to minimize the alteration of the desired protein concentration of the solution. Finally, the solution was filtered through a 0.22-µm Millipore filter and distributed in 10-ml vials, which were hermetically sealed and placed in a water bath for heating at 85 ˚C for 15 min. After heat treatment, the vials were immediately cooled by immersion in ice-water mixtures to quench the aggregation process. Solutions of 0.25 wt% ß-lactoglobulin spherical aggregate were prepared by dilution using Milli-Q water. Then 2.5mM of DHN and 2.5mM of TA were added in dilute protein solution at 40˚C temperature and incubated at 25˚ C for 160 min. Color change is a good indicator of reaction completion, which turned to dark brown color from light pink. The pure solid brown colored products were centrifugal separated, then washed by three cycles of centrifugation/washing/redispersion in deionized water and alcohol, and oven-dried at 80˚C for 4 h. The dryed brown particles were dispersed in ethanol and carbonized at 180-200˚C for 2 h under constant N_2 pressure. Then brown to dark grey particles were centrifugal separated, then washed by three cycles of centrifugation/washing/redispersion in alcohol and oven-dried at 80 C for 4 h/ vacuum dry at room temperature.

Phase transfer of *C*-NP: To study the phase transfer behavior of the NP and *C*-NP, we added 1 ml of water to the previously prepared NP-toluene (1 ml) and *C*-NP-toluene suspension (1 ml) respectively. The photographs in Fig 4 c and Fig 4 d were taken with a digital camera (Nikon-D5300).

Characterization: Transmission electron microscopy (TEM) was performed on a JEOL JEM-1400 plus microscope. The morphology and cross sectional analysis of spherical particles were done with the help of FE-SEM (Hitachi SU-8000, JAPAN) equipped with an energy dispersive X-ray analysis system (EDXA). Fluorescence spectra were investigated using a JASCO FP-6500 spectrofluorometer. UV-visible absorption spectra were obtained on a JASCO V-630 spectrophotometer. The particle`s zeta

potentials were evaluated by dynamic light scattering (DLS) with a Zetasizer nano ZS (Malvern Instruments Ltd., Worcestershire, UK).



Scheme S1. Reaction scheme for the synthesis of polymer-coated nanoparticles. 1,5-dihydroxynapthalene and 1,3,5-trimethyl-1,3,5-triazinane polymerized *in situ* and crosslinked on the protein surface with extension of π -conjugated structures at room temperature (298 K).

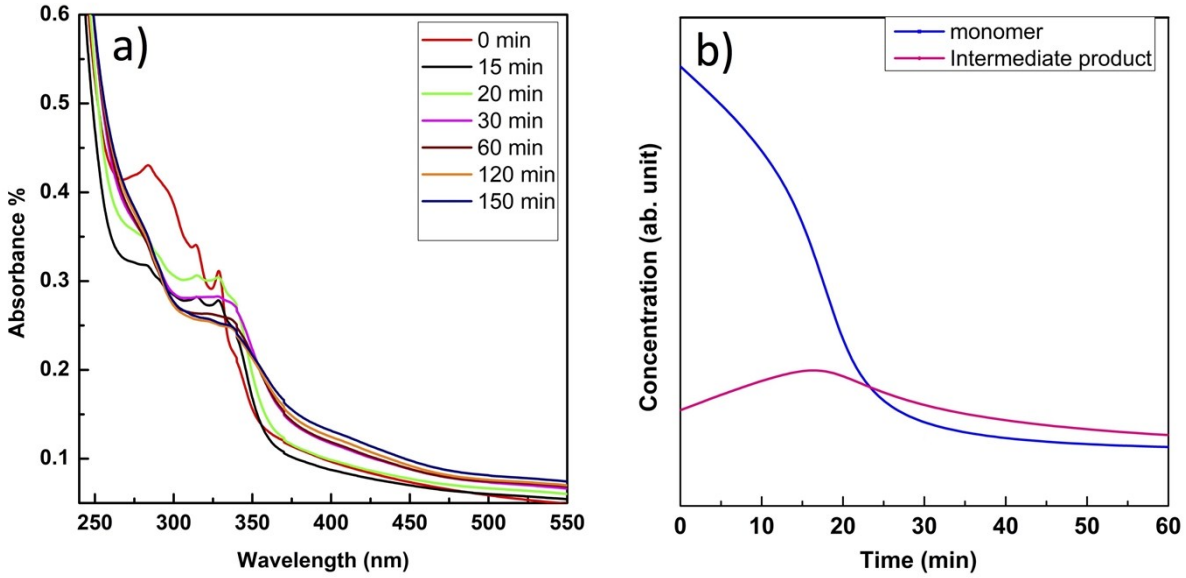


Figure S1. a) UV-Vis spectra of intermediate products in aqueous solution b) Time dependent concentration change of monomer and intermediate species

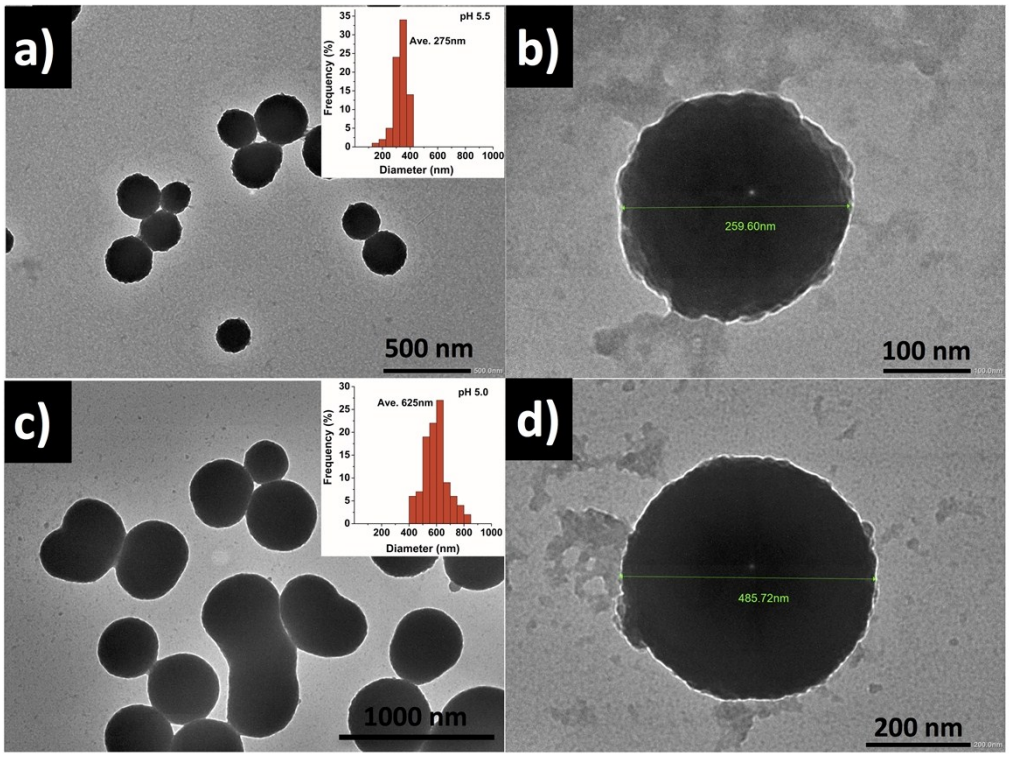


Figure S2**.** TEM micrographs of *P*-NP at varying pH. *P*-NP at pH 5.5 (a, b); *P*-NP at pH 5.0 (c, d); The particle size distribution of each sample is included in each micrograph

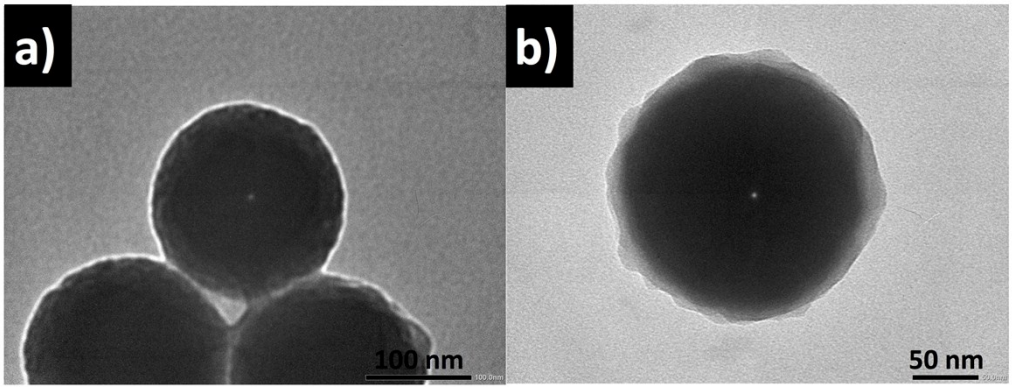


Figure S3. TEM micrographs of carbon-coated silica nanospheres (a), and individual nanosphere (b)

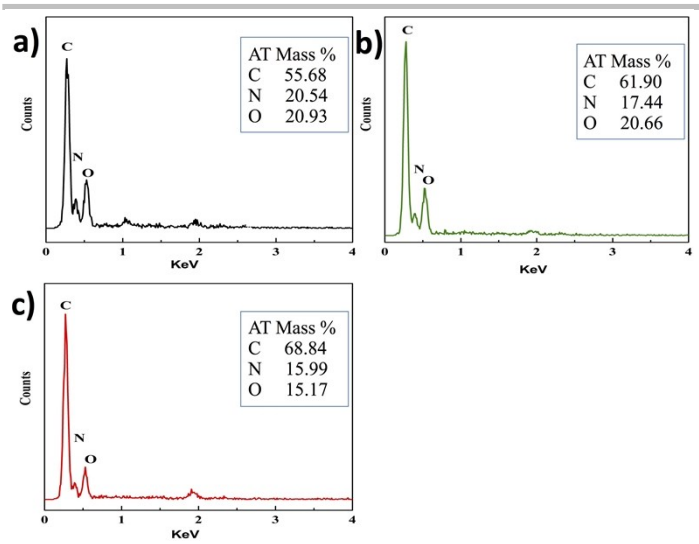


Figure S4. EDX spectra of the prepared NP (a); *P*-NP (b) and *C*-NP (c)

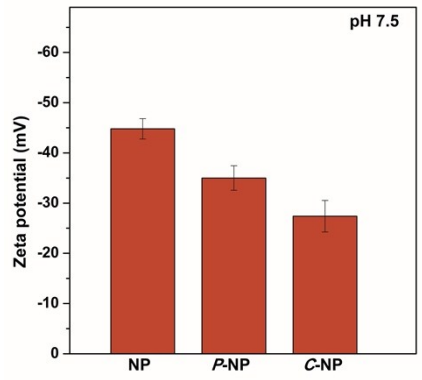


Figure S5. Zeta-potential of NP, *P*-NP and *C*-NP particles at pH 7.5

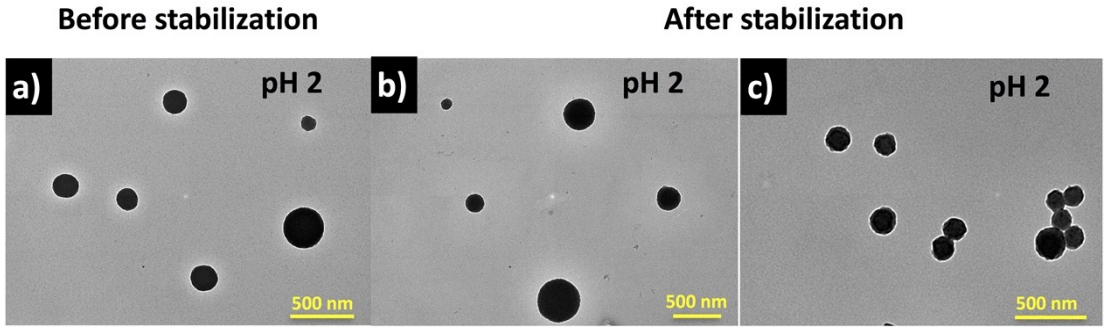


Figure S6. TEM micrographs of NP (a); *P*-NP (b) and *C*-NP (c) in acidic solution, incubated for 3h at 25˚C

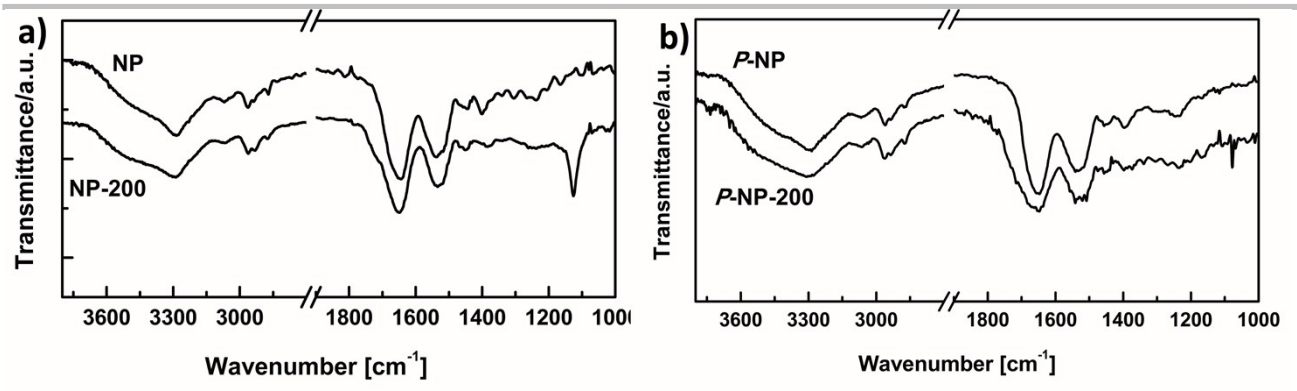


Figure S7. FTIR spectra of protein nanospheres; a) Without polymer coating, b) with polymer coating