Electronic Supplementary Material (ESI) for journal name
This journal is © The Royal Society of Chemistry 2022

Electronic Supplementary Information (ESI)

Confinements of Thermoresponsive Dendronized Polymers to Proteins

Yi Yao,[a] Jintao Yang,[a] Wen Li,*[a] and Afang Zhang*[a]

[a] International Joint Laboratory of Biomimetic and Smart Polymers, School of Materials Science and Engineering, Shanghai University, Nanchen Street 333, Shanghai 200444, China. *Email: wli@shu.edu.cn (W.L.) and azhang@shu.edu.cn (A.Z.)

Table of Contents

Fig. S1. (A) Molecular structure of OEG-based dendronized polymethacrylate homopolymer **PG1** and dendronized chitosans (**DCS**). (B) Synthetic procedure for copolymer **PG1S**. Through free radical polymerization of **MS=S** with dendritic monomers (**M3**) in the presence of AIBN, DMF, at 65 °C for 3 h S3

Table S1. Conditions for and results from the polymerizations S3

Fig. S2. ^{1}H NMR spectra of **PG1S** in DMSO-d_6 at 60 °C, C = 10 mg·mL^{-1} S4

Fig. S3. (A) Absorbance of solutions for **PG1S** with Mb at 420 nm over reaction time. Heat effects by ITC in the titration of (B) **PG1** and (C) **DCS** to Mb in traditional mode at 25 °C S4

Fig. S4. (A) Cloud point T_{cp}s for **PG1/proteins** mixture with different concentrations of proteins, $C_{\mathbf{PG1}}$ = 2.5 mg·mL^{-1}, $C_{\mathbf{PG1}}$ = 2.5 mg·mL^{-1}. (B) Plots of transmittance *vs* temperature for **DCS**, C = 2.5 mg·mL^{-1}. (C) Hydrodynamic radii (R_h, intensity radii) of **PG1/BSA**, **PG1/LYZ**, and **DCS** between 25 °C and 45 °C, respectively. C = 0.05 mg·mL^{-1} S5

Fig. S5. R_h of (A) proteins and **PG1/proteins**, and (B) **PG1S-Mb**, **DCS/Mb**, and **DCS** at different temperatures. (C) Fluorescence spectrum of ANS (420−650 nm) in samples at 25 °C and 40 °C, respectively S6

Fig. S6. FT-IR spectra of (A) **PG1S**, **PG1S-Mb**, **PG1**, and **PG1/Mb**, at 25 °C, and (B) **PG1S** at different temperatures. All samples were prepared in D_2O S6

Fig. S7. Changes of frequency Δf (red curves) and dissipation ΔD (blue curves) in QCM cell over time, after the injection of (A) Mb and PBS solution at 25 °C (below: schematic representation for immobilization process of proteins on QCM cells), and (B) **DCS** at 25 °C and 60 °C, respectively S7

Fig. S8. Far-UV CD spectra of (A, C) BSA and (B, D) LYZ in presence of **PG1** with different concentrations and at different temperatures (polymer/protein = 40, w/w), respectively S8

Fig. S9. Absorbance spectra of Mb from **PG1/Mb** (A) with different concentrations of polymer (polymer/protein, from 0 to 50, w/w) and (B) at different temperatures (polymer/protein = 40, w/w). (C) Maximum absorbance (A_{max}) at 410 nm of Mb from **PG1/Mb** solution after heating and cooling between 25 °C and 37 °C for 8 cycles S9

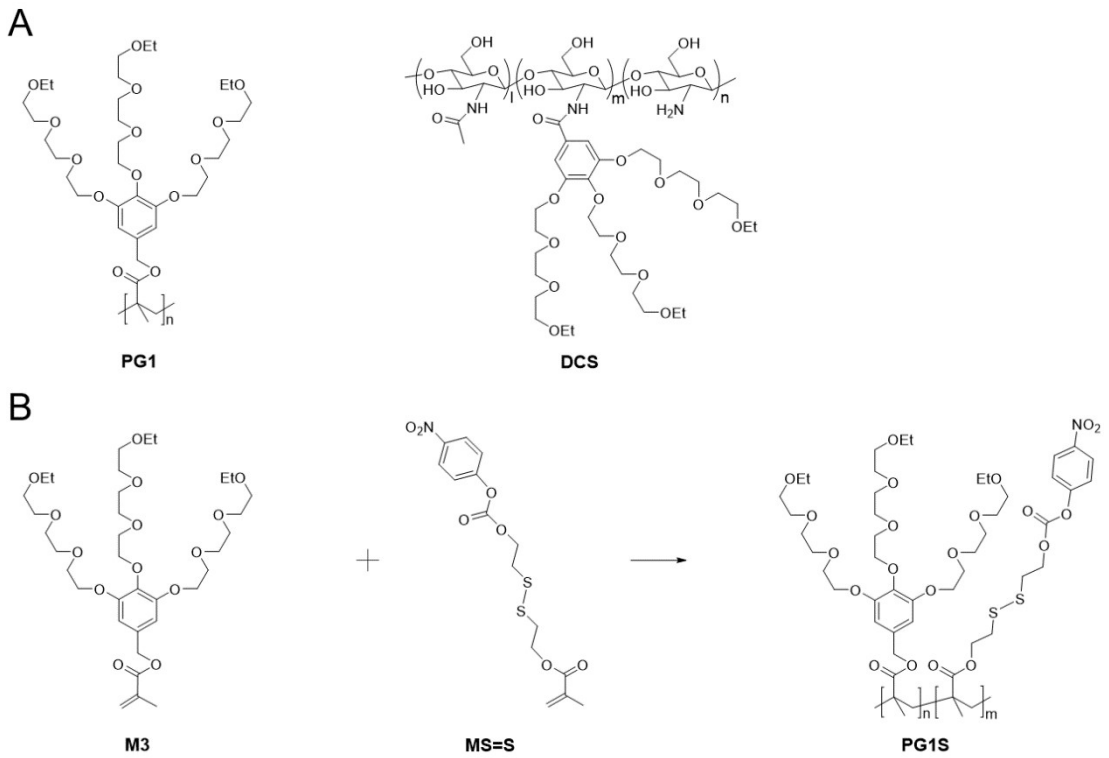

Fig. S1 (A) Molecular structures of OEG-based dendronized polymethacrylate homopolymers **PG1** and dendronized chitosans (**DCS**). (B) Synthetic procedure for the copolymer **PG1S** through free radical copolymerization of **MS=S** with dendritic monomer **M3** in the presence of AIBN at 65 °C for 3 h in DMF.

Table S1 Conditions for and results from the polymerizations

sample	feed ratio [a]	actual copolymerization ratio [b]	GPC results [c]	
	[**M3**]/[**MS=S**]	[**M3**]/[**MS=S**]	$M_n \times 10^{-5}$	Đ
PG1	\	\	2.3	1.5
PG1S	20:1	21.5	2.2	2.3

[a] Polymerizations were performed with AIBN as the initiator at 65 °C. [b] Copolymer compositions were determined through calculation according to proton integrations from ^{1}H NMR spectra in Fig. S2. [c] Determined by GPC with DMF as the eluent containing 0.1 wt% LiBr, M_n and Đ represent number-average molecular weight and polydispersity, respectively.

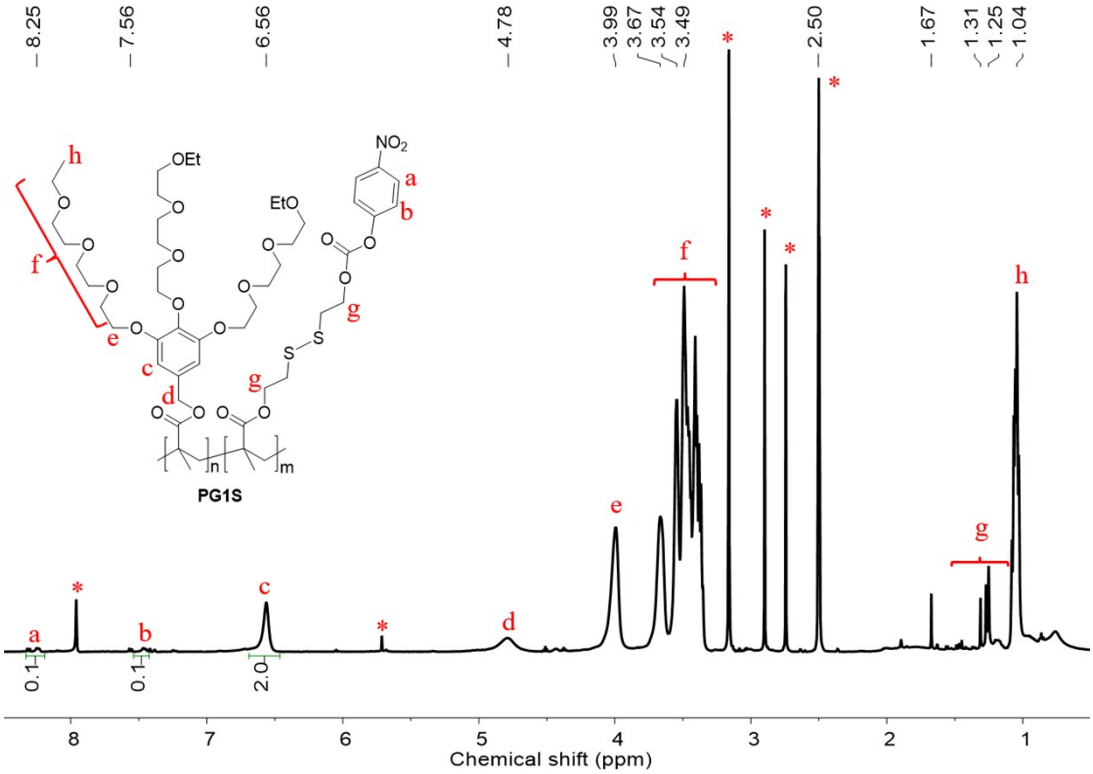

Fig. S2 ^{1}H NMR spectra of **PG1S** in DMSO-d_6 at 60 °C, C = 10 mg·mL^{-1}.

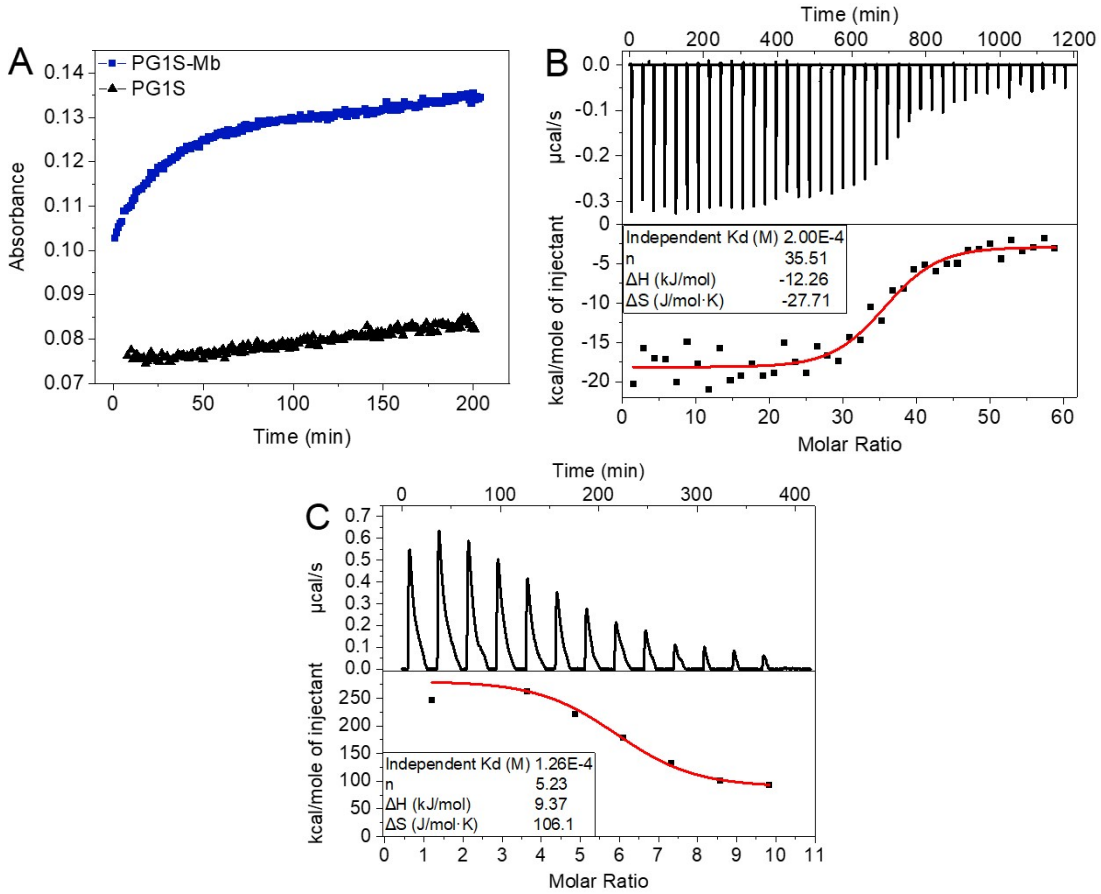

Fig. S3 (A) Absorbance of solutions for **PG1S** with Mb at 420 nm over reaction time. Heat effects by ITC in the titration of (B) **PG1** and (C) **DCS** to Mb in traditional mode at 25 °C.

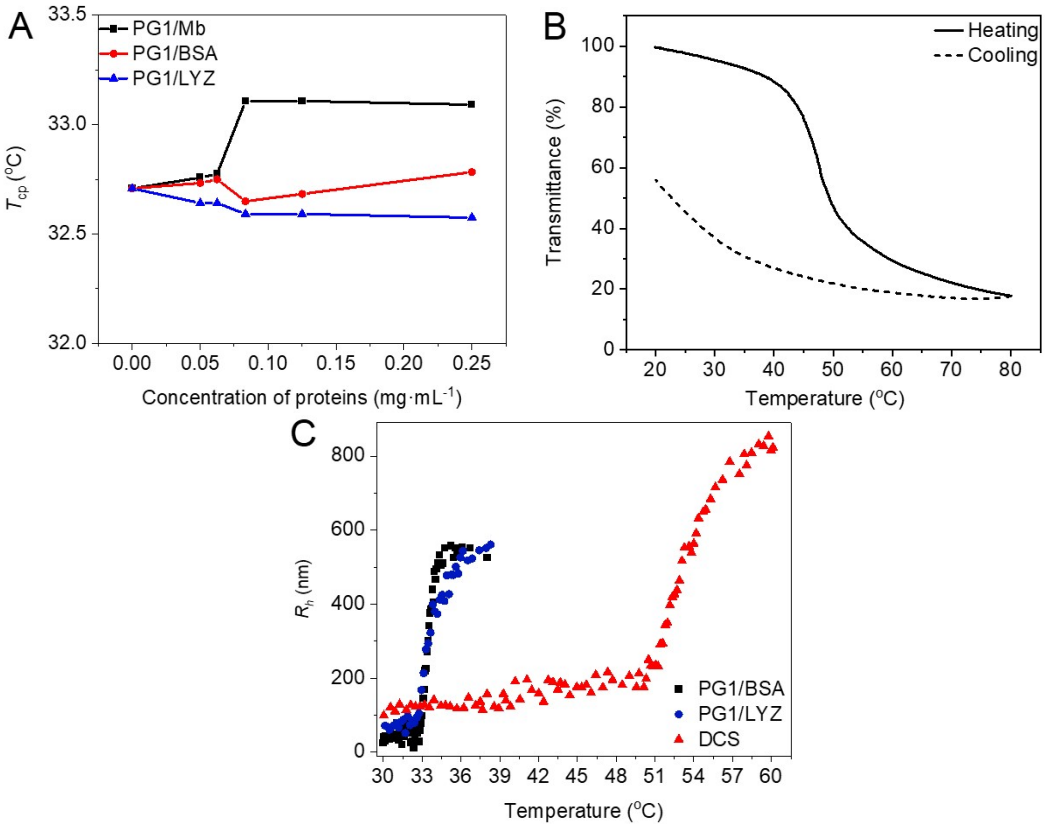

Fig. S4 (A) Cloud point (T_{cp}s) for **PG1/proteins** mixture with different concentrations of proteins. [**PG1**] = 2.5 mg·mL^{-1}. (B) Plots of transmittance *vs* temperature for **DCS**, C = 2.5 mg·mL^{-1}. (C) Hydrodynamic radii (R_h, intensity radii) of **PG1/BSA**, **PG1/LYZ**, and **DCS** between 25 °C and 45 °C, respectively. C = 0.05 mg·mL^{-1}.

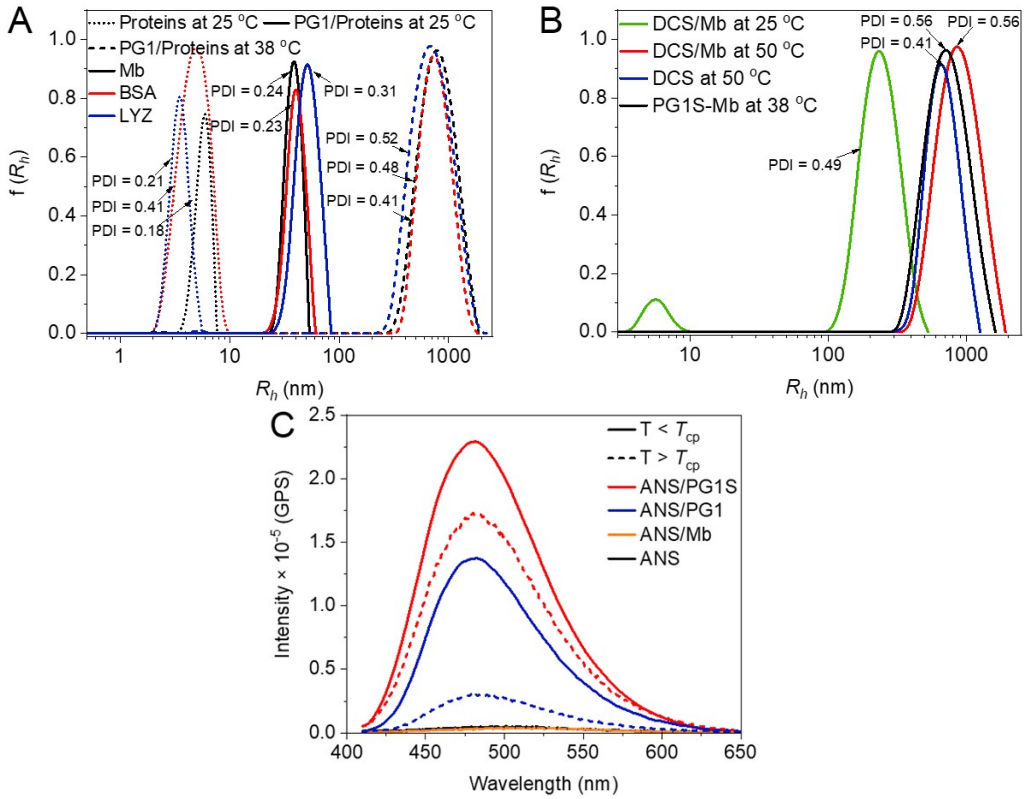

Fig. S5 R_h of (A) proteins and **PG1/proteins**, and (B) **PG1S-Mb**, **DCS/Mb**, and **DCS** at different temperatures. (C) Fluorescence spectrum of ANS (420−650 nm) in samples at 25 °C and 40 °C, respectively. PDI in Figure A and B represents the polydispersities of the aggregates.

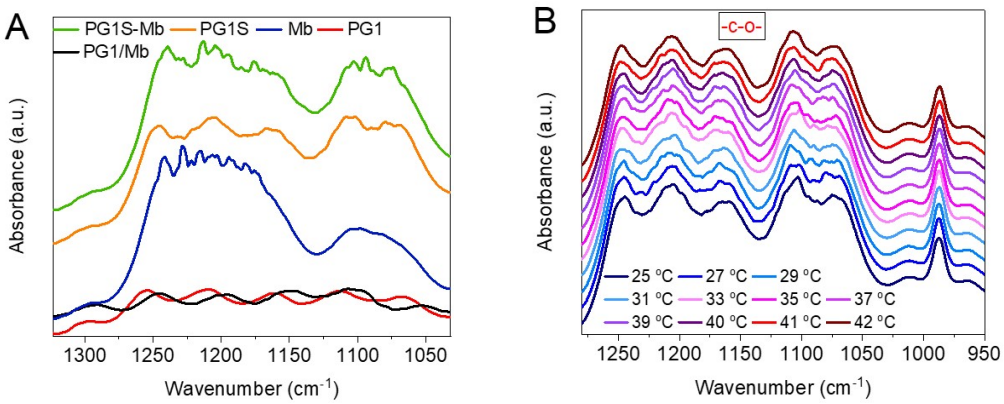

Fig. S6 FT-IR spectra of (A) **PG1S**, **PG1S-Mb**, **PG1**, and **PG1/Mb**, at 25 °C, and (B) **PG1S** at different temperatures. All samples were prepared in D_2O.

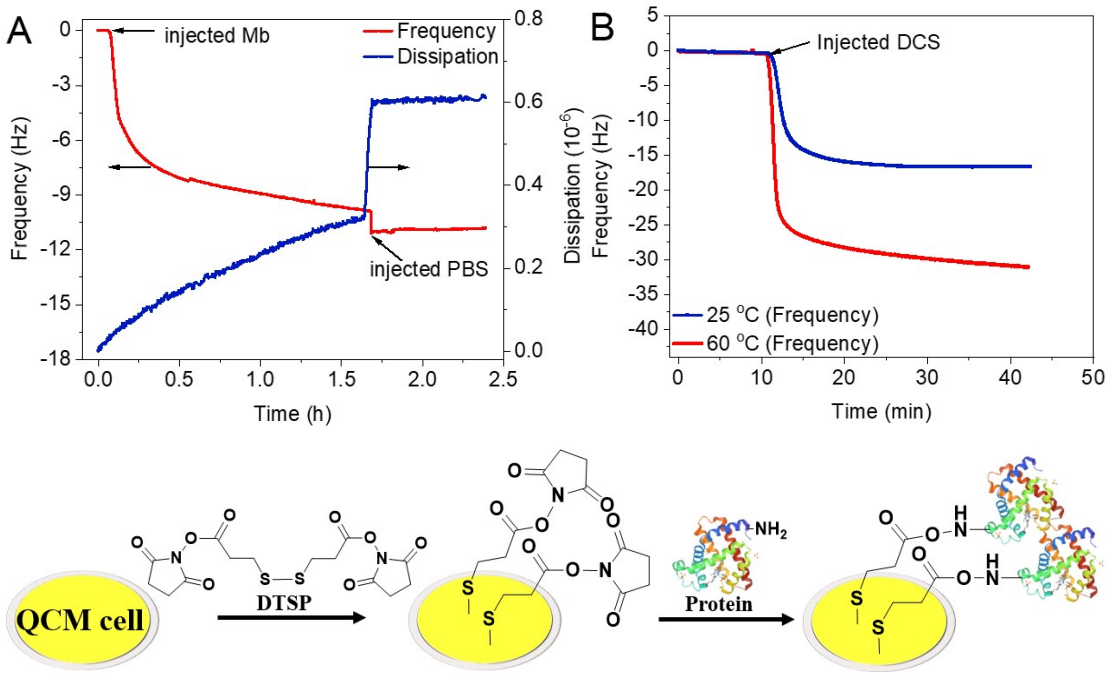

Fig. S7 Changes of frequency Δ*f* (red curves) and dissipation Δ*D* (blue curves) in QCM cell over time, after the injection of (A) Mb and PBS solution at 25 °C (below: schematic representation for immobilization process of proteins on QCM cells), and (B) **DCS** at 25 °C and 60 °C, respectively.

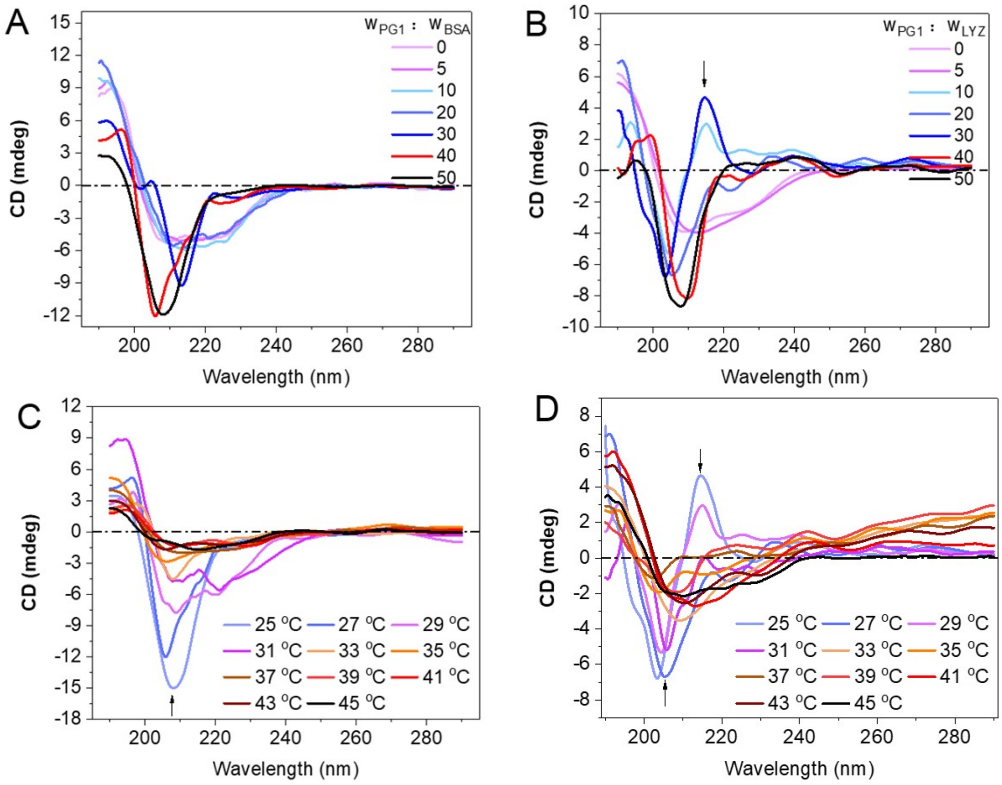

Fig. S8 Far-UV CD spectra of (A, C) BSA and (B, D) LYZ in presence of **PG1** with different concentrations and at different temperatures (polymer/protein = 40, w/w), respectively.

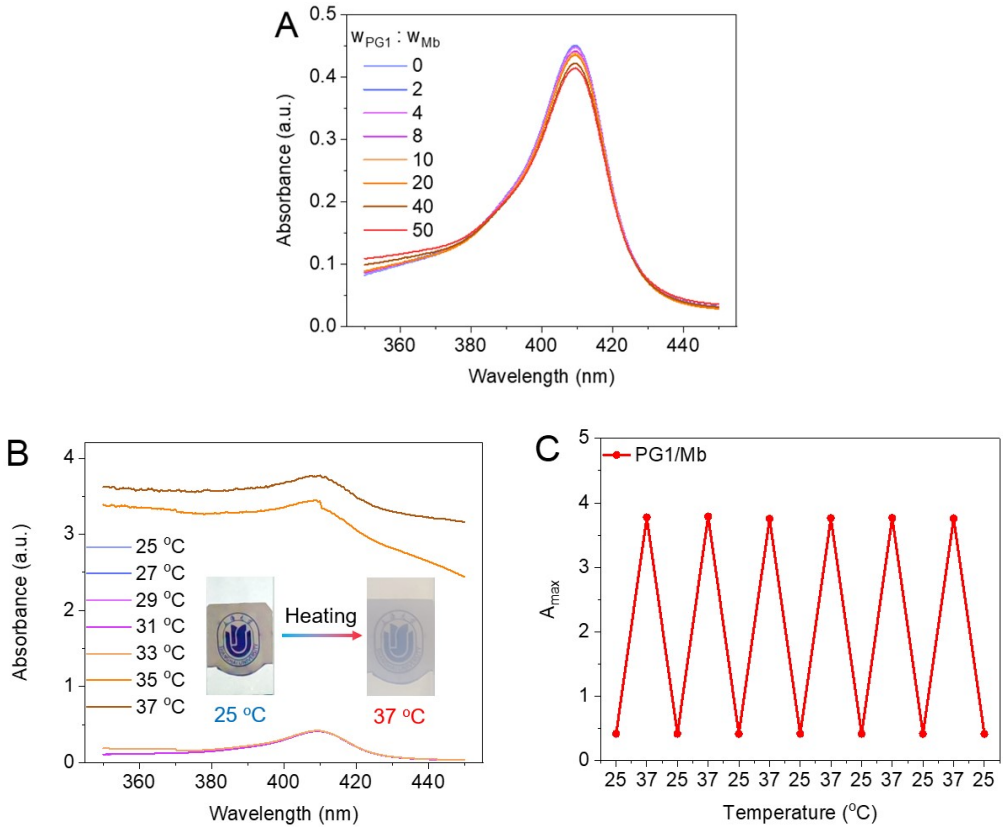

Fig. S9 Absorbance spectra of Mb from **PG1/Mb** (A) with different concentrations of polymer (polymer/protein, from 0 to 50, w/w) and (B) at different temperatures (polymer/protein = 40, w/w). (C) Maximum absorbance (A_{max}) at 410 nm of Mb from **PG1/Mb** solution after heating and cooling between 25 °C and 37 °C for 8 cycles. Inset in Figure B: photographs of the solutions below and above the T_{cp}.