## **Electronic Supplementary Information**

# Regulation of Lysozyme Activity Based on Thermotolerant Protein/Smart Polymer Complex Formation

Sumon Ganguli<sup>†</sup>, Keitaro Yoshimoto<sup>†, ‡, \$</sup>, Shunsuke Tomita<sup>†</sup>, Hiroshi Sakuma<sup>†</sup>, Tsuneyoshi Matsuoka<sup>†</sup>, Kentaro Shiraki<sup>†</sup>, and Yukio Nagasaki\* <sup>†, ‡, \$, \$, ¶</sup>

†Graduate School of Pure and Applied Science, University of Tsukuba, 1-1-1 Ten-noudai, Tsukuba, Ibaraki 305-8573, Japan. ‡Center for Tsukuba Advanced Research Alliance (TARA), University of Tsukuba, Ten-noudai 1-1-1, Tsukuba 305-8577, Japan. <sup>\$</sup>Tsukuba Research Center for Interdisciplinary Materials Science (TIMS), University of Tsukuba, 1-1-1 Ten-noudai, Tsukuba, Ibaraki, 305-8571, <sup>\$</sup>Master's School of Medical Science, Graduate School of Comprehensive Human Sciences, University of Tsukuba, 1-1-1 Ten-noudai, Tsukuba, Ibaraki 305-8573, Japan. <sup>∏</sup>Satellite Laboratory, International Center for Materials Nanoarchilectonics (MANA), National Institute of Materials Science (NIMS), 1-1-1 Ten-noudai, Tsukuba, Ibaraki 305-8573, Japan.

\*To whom correspondence should be addressed. Yukio Nagasaki, Ph.D., Graduate School of Pure and Applied Science, University of Tsukuba, Ten-noudai 1-1-1, Tsukuba, Ibaraki 305-8573, Japan.

Phone and FAX: +81-29-853-5749 e-mail: yukio@nagalabo.jp

#### **Supplementary figures and tables**

Figure S1 Size exclusion chromatogram (SEC) of synthesized polymers.

Figure S2 <sup>1</sup>H-NMR spectrum of PEG-MA.

Figure S3 <sup>1</sup>H-NMR spectrum of purified PEAMA-g-PEG.

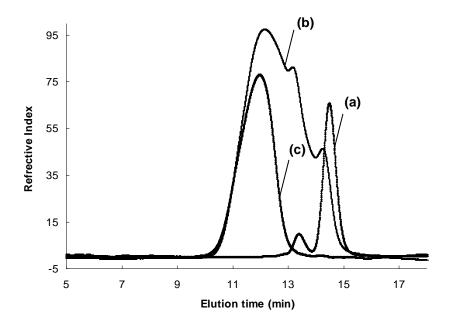
Table S1 Characterization of PEG-MA.

Table S2 Characterization of purified PEAMA-g-PEG.

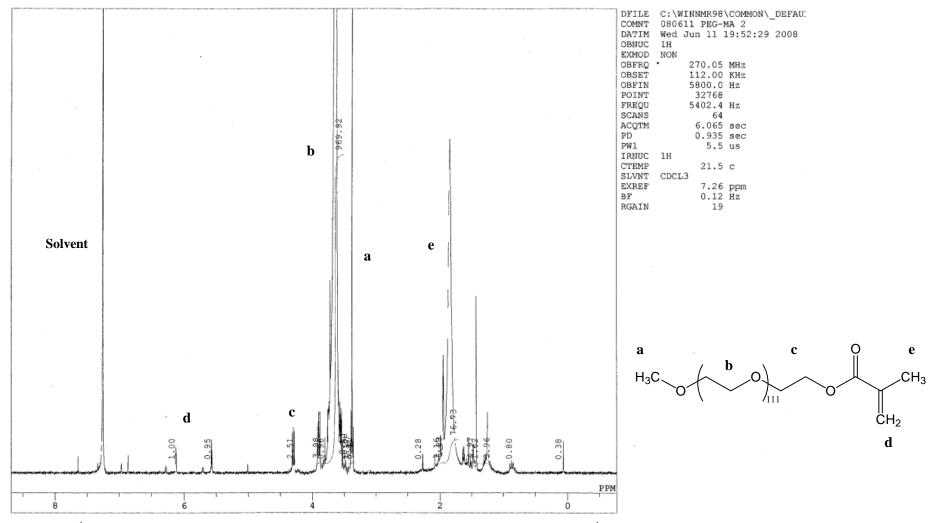
#### Supplementary data

Figure S4 Changes in the normalized enzymatic activity of lysozyme in the presence of PEG-OH(5k)

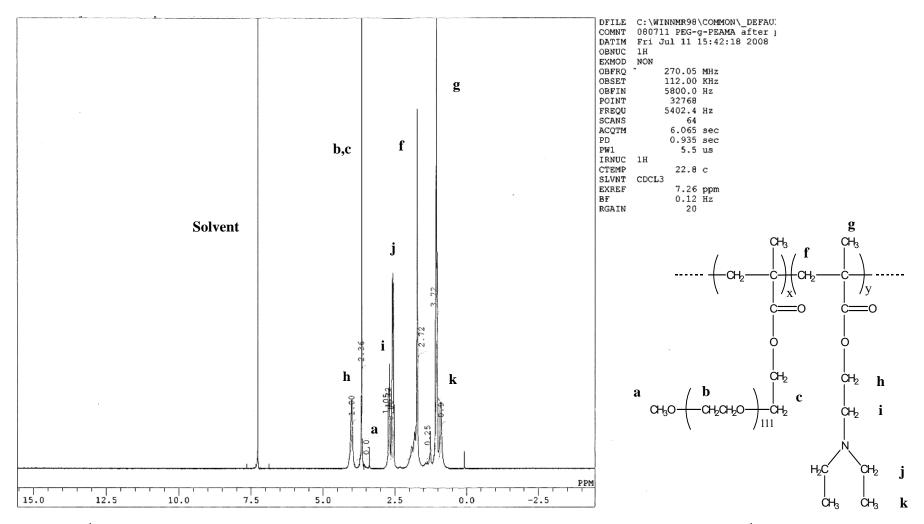
### Supplementary figures and tables



**Figure S1:** Size exclusion chromatogram (SEC) of synthesized polymers. (a) PEG-MA macromonomer, (b) PEAMA-*g*-PEG before HPLC purification, and (c) PEAMA-*g*-PEG after HPLC purification. The molecular weight and the molecular weight distribution were determined by Gel permeation chromatography (TOSOH HLC-8120, TOSHO Co., Tokyo, Japan) with TSK gel columns (TSKgel SuperHZ3000 + HZ4000) and an internal refractive index (RI) detector (TOSO HHLC-8020RI) using the calibration curve of PEG. THF containing 0.5 wt % triethylamine was used as the eluent at a flow rate of 0.35 mL/min at 35°C.



**Figure S2:** <sup>1</sup>H NMR spectrum of PEG-MA. The characterization of the PEG-MA was carried out by <sup>1</sup>H-NMR (JEOL EX-270 spectrometer, JEOL Ltd., Tokyo, Japan) at 270 MHz in CDCl<sub>3</sub> solution at room temperature.



**Figure S3:** <sup>1</sup>H NMR spectrum of purified PEAMA-*g*-PEG. The characterization of the PEAMA-*g*-PEG was carried out by <sup>1</sup>H-NMR (JEOL EX-270 spectrometer, JEOL Ltd., Tokyo, Japan) at 270 MHz in CDCl<sub>3</sub> solution at room temperature.

**Table S1:** Characterization of PEG-MA.

$^{\mathrm{a}}M_{\mathrm{n}}$	$^{ m a}M_{ m w}$	<sup>a</sup> PDI	<sup>b</sup> Functionality
4,700	4,900	1.04	78 %

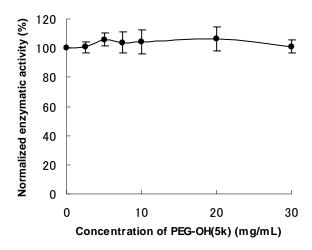
<sup>&</sup>lt;sup>a</sup>Number-averaged molecular weight  $(M_n)$ , weight-averaged molecular weight  $(M_w)$ , and polydispersity index (PDI) were obtained from SEC analysis. <sup>b</sup>End group functionality was determined from <sup>1</sup>H-NMR analysis.

**Table S2:** Characterization of purified PEAMA-*g*-PEG.

$^{\mathrm{a}}M_{\mathrm{n}}$	$^{\mathrm{a}}M_{\mathrm{w}}$	<sup>a</sup> PDI	$^{ m a}M_{ m n}$ of PEG segment (PEG-MA)	<sup>b</sup> Molecular weight of PEAMA segment (Number of EAMA units)	Number of PEG chains
28,000	39,000	1.38	4,700	19,000 (104)	2

 $<sup>^{</sup>a}M_{n}$ ,  $M_{w}$ , PDI of PEAMA-g-PEG, and  $M_{w}$  of PEG segment were obtained from SEC analysis.  $^{b}$ Molecular weight of PEAMA segment in PEAMA-g-PEG were determined by  $^{1}$ H-NMR analysis.

## **Supplementary data**



**Figure S4** Changes in the normalized enzymatic activity of lysozyme in the presence of PEG-OH(5k). The experimental conditions were same as those shown in Figure 1, which details were described in the experimental section.