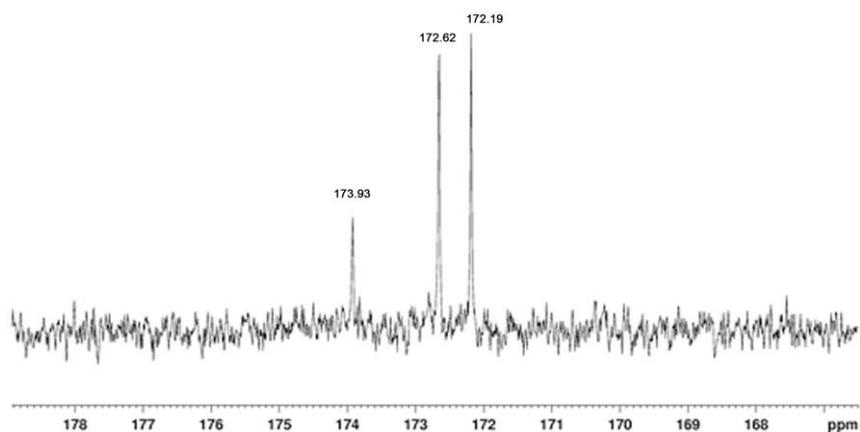


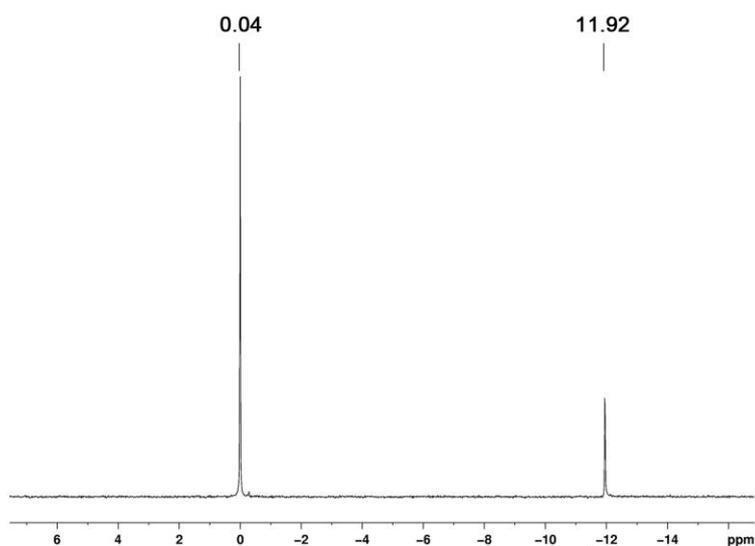
# Synthesis of Biomimetic Co-Polypeptides with Tunable Degrees of Phosphorylation

Vitali Lipik, Zhang Lihong and Ali Miserez

## Supplementary information



**Supplementary Figure 1.**  $^{13}\text{C}$  NMR spectra of the co-polypeptides after ring-opening polymerization. The three peaks from left to right correspond to the signal of carbonyl groups of Tyr-O-Bzl, Ser-OH, Ser-O-Cbz, respectively.

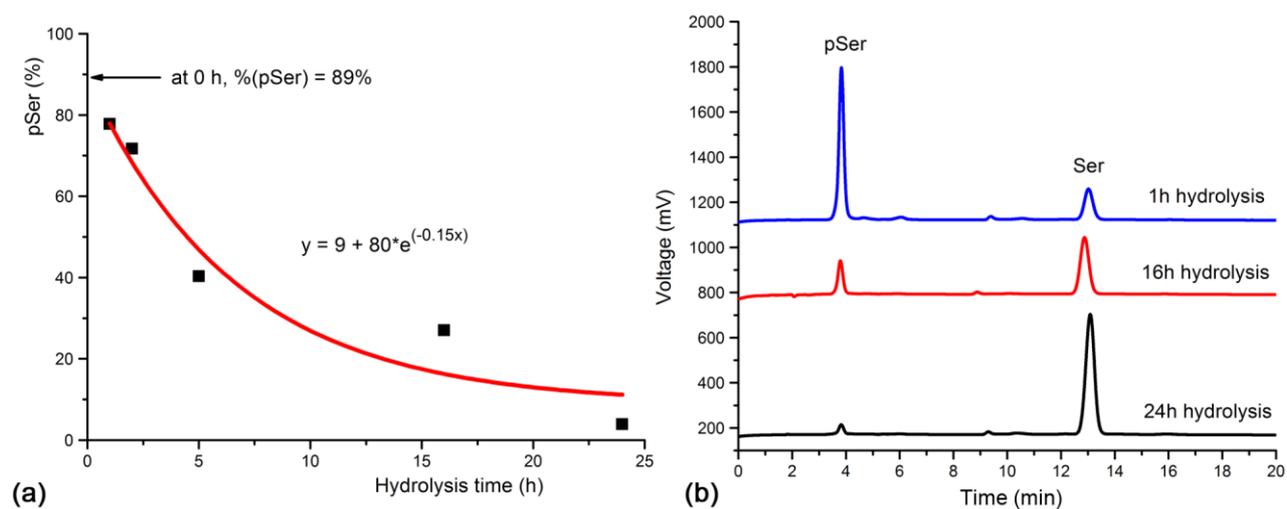


**Supplementary Figure 2.**  $^{31}\text{P}$  NMR spectra of the co-polypeptides after phosphorylation. The two peaks from left to right correspond to the signal of mono-phosphate and di-phosphate, respectively.

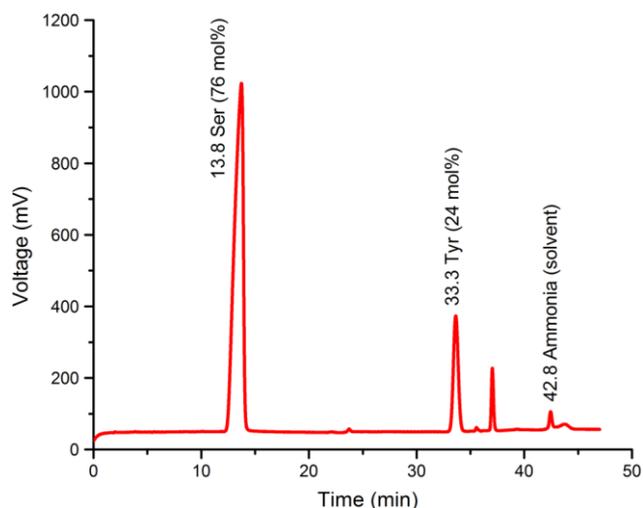
**Supplementary Table 1** - Characteristics of synthesized polypeptides before phosphorylation.

Polypeptides with Ser unprotected –OH groups (target), [%]	Molecular weight by SLS, [kDa]	Yield of polypeptide, [%]	Melting enthalpy (serine), [J/g]	PDI*
25	22.8±2.2	85	83.9	0.476
50	19.3±2.0	52	23.2	0.605
75	19.2±5.7	64	38.8	0.647
100	15.7±4.5	50	67.3	0.845

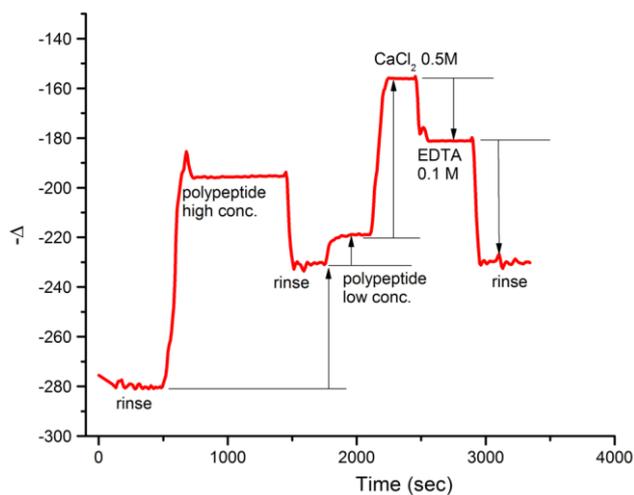
\*PDI values were obtained by Dynamic Light Scattering measurements.



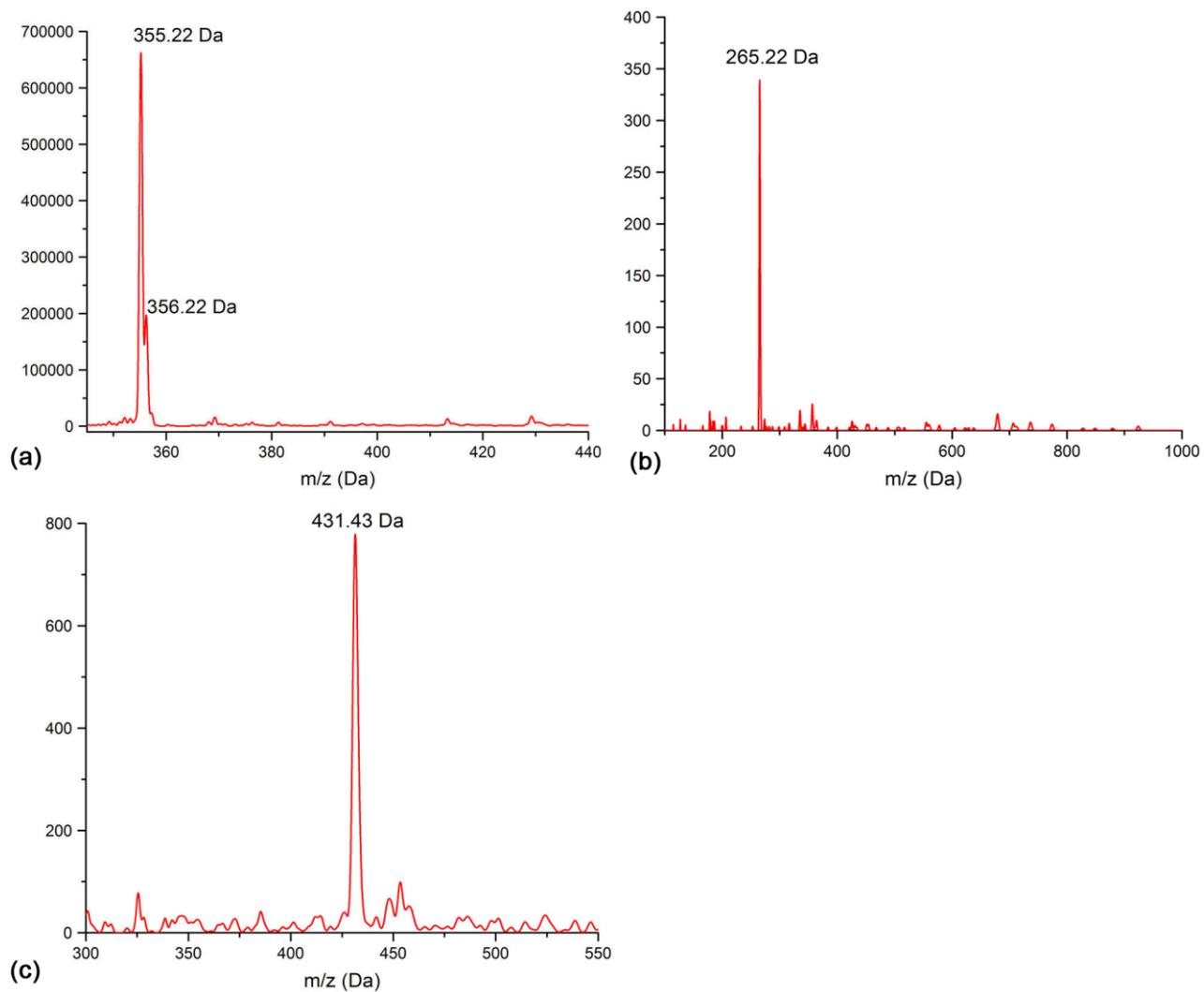
**Supplementary Figure 3.** Phosphorylation degree of serine in polypeptide A4 as quantified by Ninhydrin-based amino acid analysis (AAA) with different hydrolysis time. **(a)** pSer content in polypeptide A4 decreases with increasing the hydrolysis time and the pSer content in the polypeptide is obtained by extrapolation at time zero. **(b)** Amino acid analyzer profiles of phosphorylated polypeptide A4 at various hydrolysis times, showing the decay in pSer and concomitant increase in Ser with hydrolysis time.



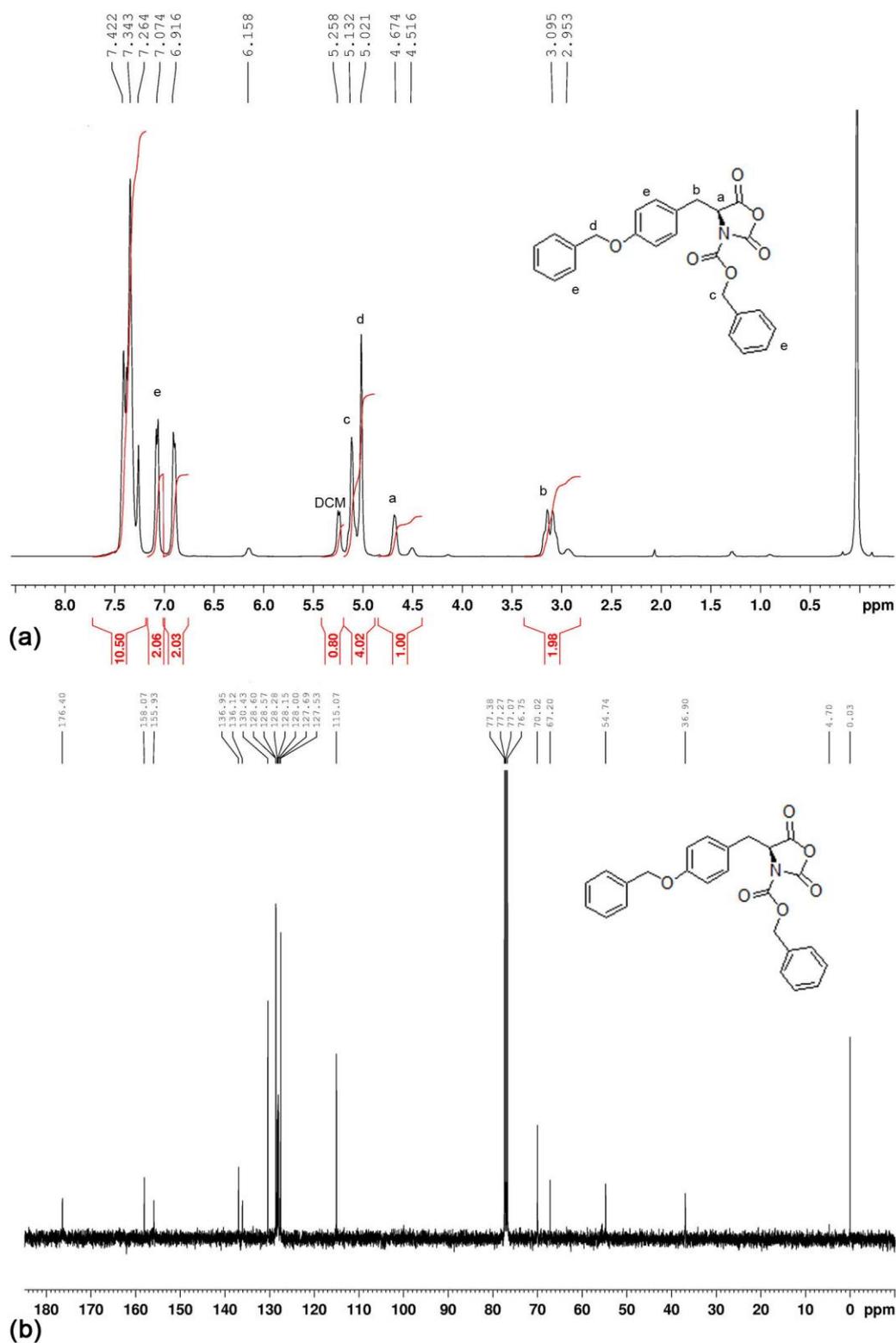
**Supplementary Figure 4.** Ninhydrin-based amino acid analysis for co-polypeptide A2 with 24 h hydrolysis time.



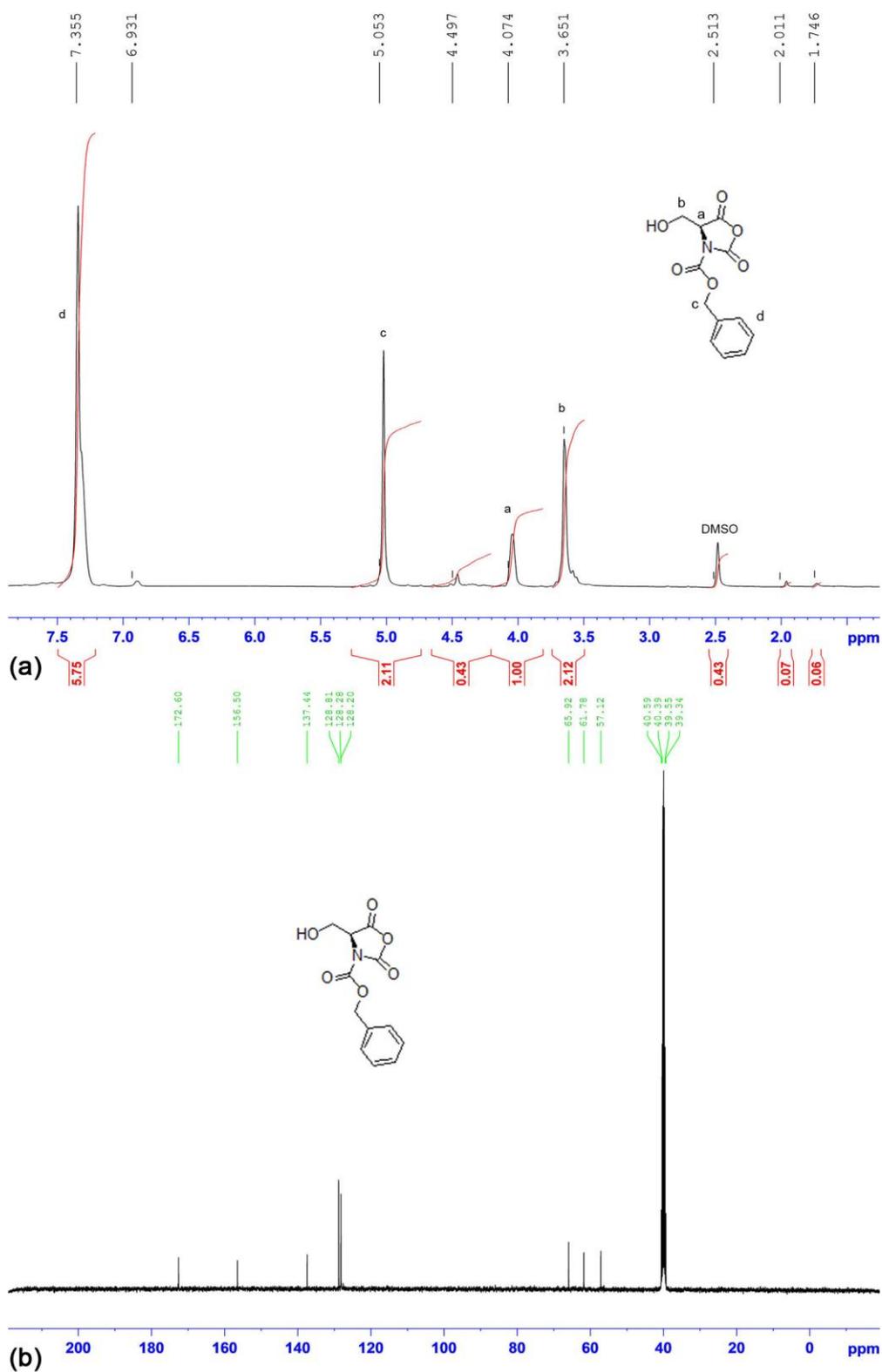
**Supplementary Figure 5.** SPR sensograms of the polypeptides A4, showing the influence of EDTA on the affinity between the polypeptides and Ca<sup>2+</sup> ions. Note that in comparison to Fig. 5, the  $\Delta$  signal returns to its value before the polypeptide (low conc.) injection.



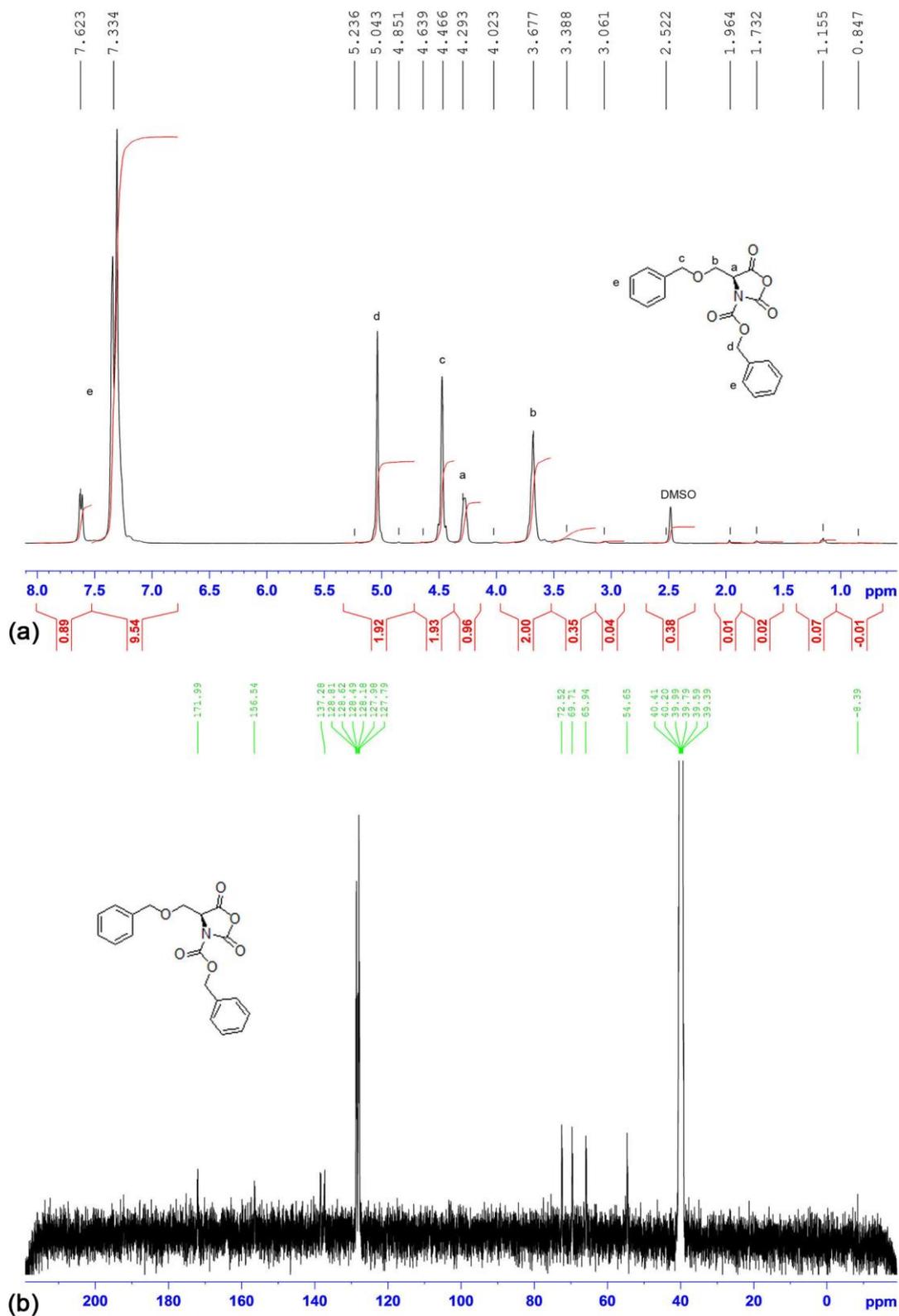
**Supplementary Figure 6.** MALDI-ToF spectra for (a) Z-Ser(Bzl)-NCA, (b) Z-Ser-NCA, (c) Z-Tyr(Bzl)-NCA.



**Supplementary Figure 7.** (a)  $^1\text{H}$  NMR spectrum of Z-Tyr(Bzl)-NCA. (b)  $^{13}\text{C}$  NMR spectrum of Z-Tyr(Bzl)-NCA.



**Supplementary Figure 8.** (a)  $^1\text{H}$  NMR spectrum of Z-Ser-NCA. (b)  $^{13}\text{C}$  NMR spectrum of Z-Ser-NCA.



**Supplementary Figure 9.** (a)  $^1\text{H}$  NMR spectrum of Z-Ser(Bzl)-NCA. (b)  $^{13}\text{C}$  NMR spectrum of Z-Ser(Bzl)-NCA.