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Supporting information for article:

Crystallographic analysis and biochemical applications of a novel penicillin-binding protein/ β -lactamase homologue from a metagenomic library

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Table S1 Specific activity of Est-Y29 and its mutants toward *p*-nitrophenyl acetate.

Protein	Specific activity (U/mg protein)
Est-Y29	27.461 ± 1.736
Est-Y29-K73A	0.090 ± 0.004
Est-Y29-Y182A	0.410 ± 0.018

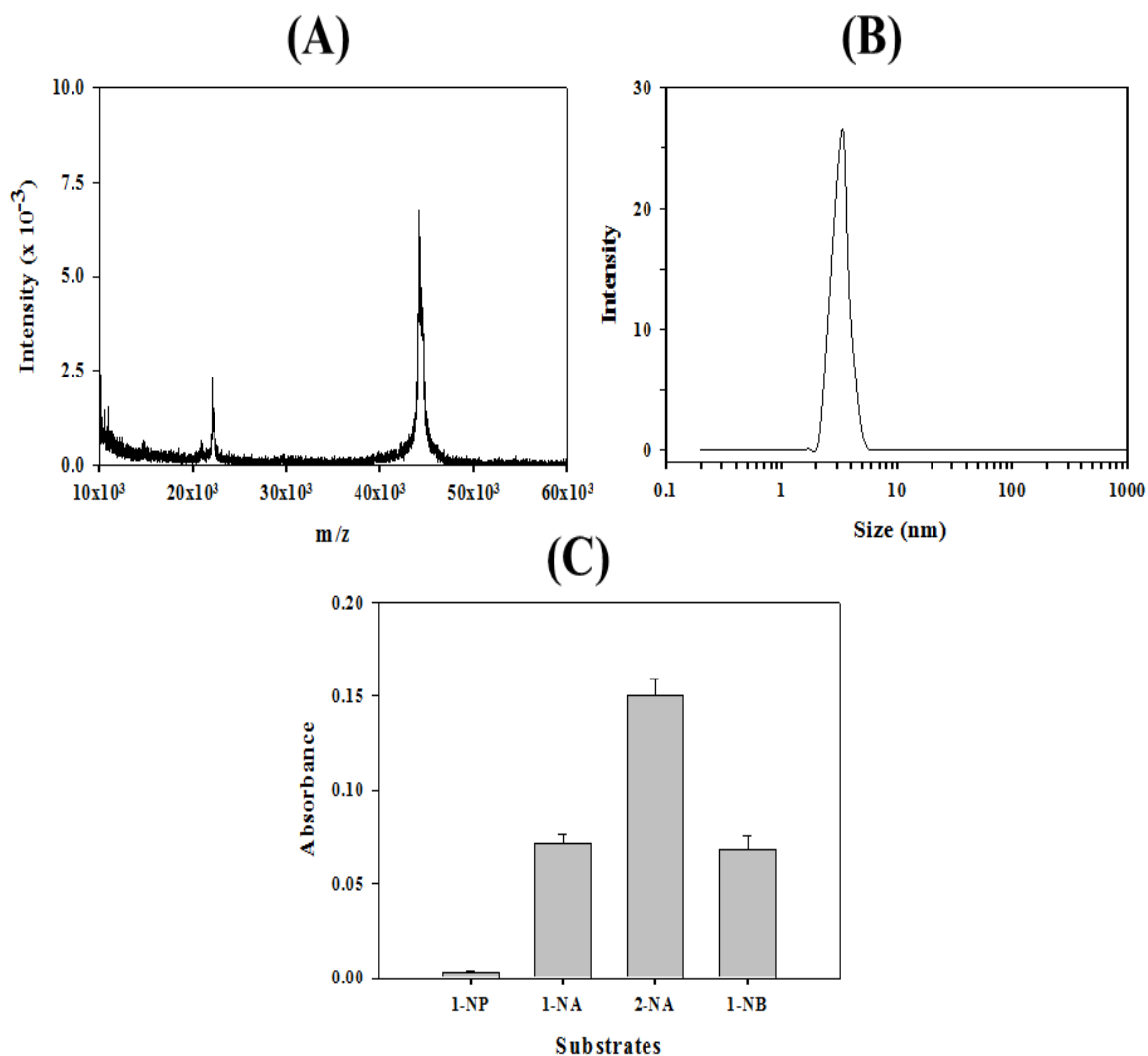


Figure S1 Biochemical characterization of Est-Y29. (A) MALDI-TOF mass spectra of Est-Y29. The $[M+H]^+$ and $[M+2H]^{2+}$ ion peaks are located at m/z values of 42.5 kDa and 21.3 kDa, respectively. (B) Dynamic light scattering results of Est-Y29. Note the single peak of Est-Y29. (C) Substrate specificities of Est-Y29 were determined for 1-naphthyl acetate (1-NA), 2-naphthyl acetate (2-NA), 1-naphthyl butyrate (1-NB), and 1-naphthyl phosphate (1-NP).

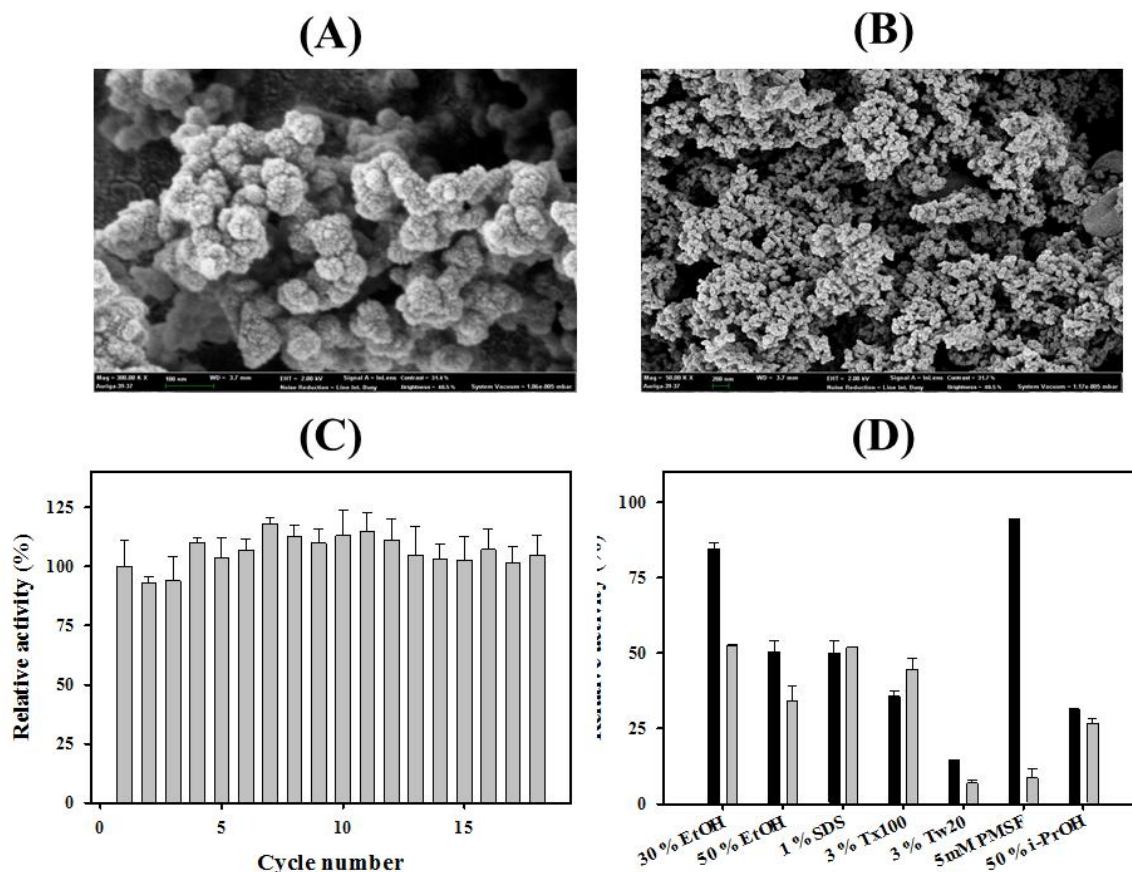


Figure S2 Cross-linked enzyme aggregates (CLEAs) of Est-Y29. Field emission scanning electron microscopic (FE-SEM) images of CLEA-Est-Y29 are shown at magnification ratios of (A) 50,000 and (B) 300,000. (C) The stability of CLEA-Est-Y29 activity was investigated for 18 cycles of reuse. (D) The chemical stabilities of CLEA-Est-Y29 and soluble Est-Y29 were compared by measuring residual activities after 1 h incubations with various chemical compounds.