

***In situ* preparation of programmable curli nanomaterials as fine-tuned sustainable supports enabling selective and oriented incorporation of enzymes**

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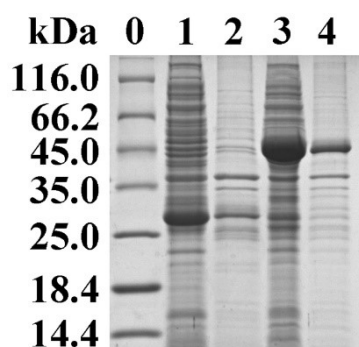
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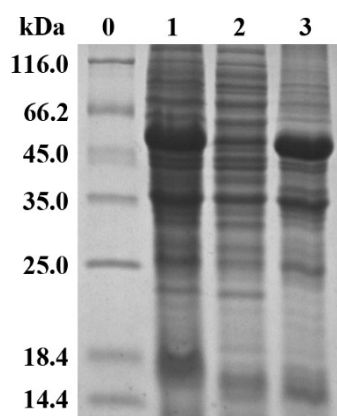
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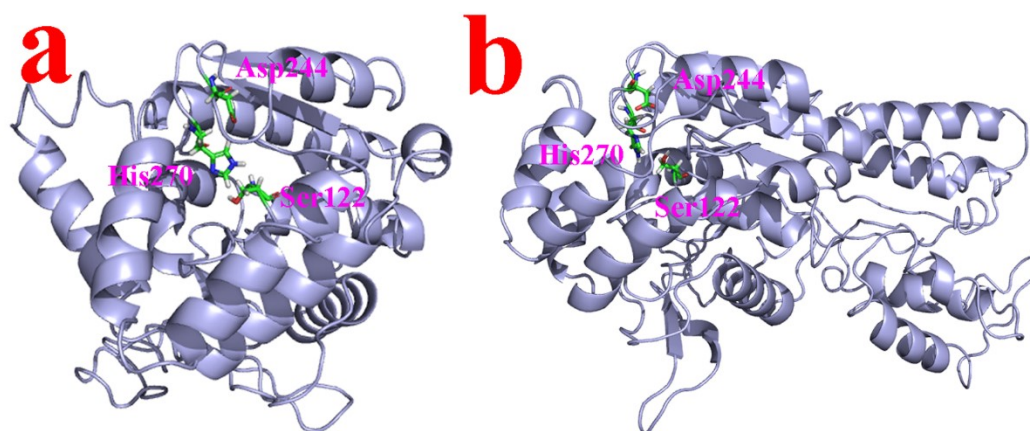
## Supplementary figures and tables



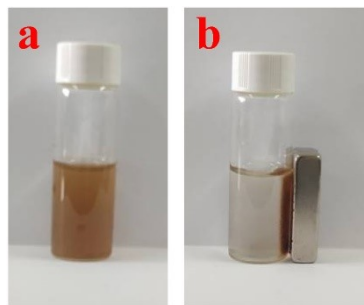
**Figure S1.** SDS-PAGE analysis of the expression of fluorescent proteins. Lane 0, protein marker; lane 1, the supernatant of the cell lysis of induced IN01; lane 2, the precipitation of the cell lysis of induced IN01; lane 3, the supernatant of the cell lysis of induced IN02; lane 4, the precipitation of the cell lysis of induced IN02.



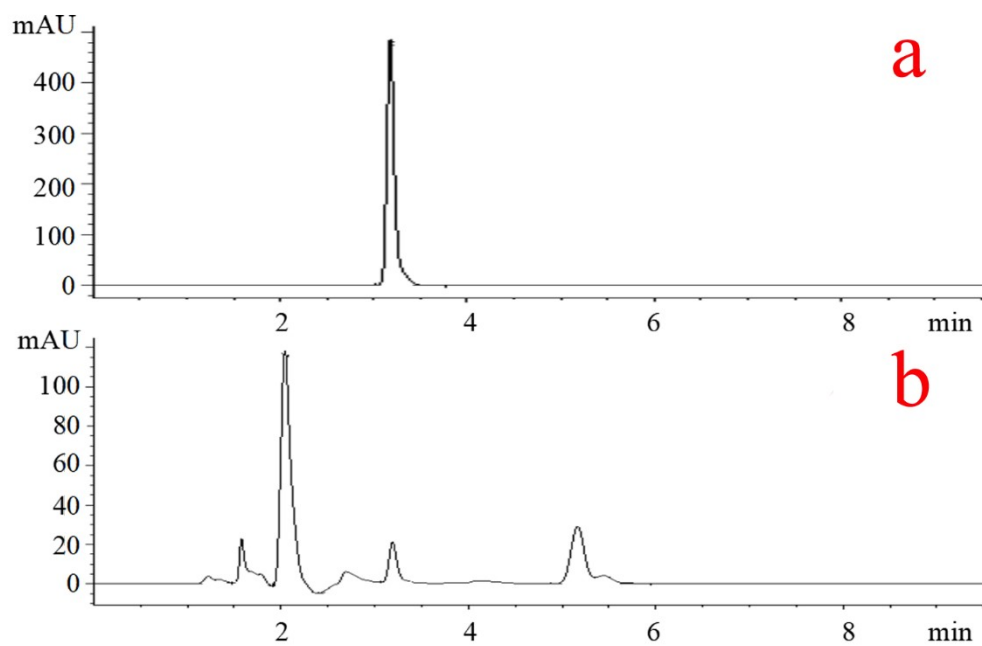
**Figure S2.** SDS-PAGE analysis of the expression of Est3-14(SpyCatcher). Lane 0, protein marker; lane 1, the cell lysis of induced EX02; lane 2, the supernatant of the cell lysis of induced EX02; lane 3, the precipitation of the cell lysis of induced EX02.



**Figure S3.** 3D structures and the catalytic triad of Est3-14 (a) and Est3-14(SpyCatcher) (b).



**Figure S4.** Before (a) and after (b) recovery of immobilized esterases using a magnet.



**Figure S5.** HPLC results of dibutyl phthalate before (a) and after (b) enzymatic hydrolysis.

## Supplementary tables

**Table S1** Amino acid sequences of functional proteins

Name	Amino acids sequence	Length (aa)
CsgA(SpyTag) <sup>1</sup>	MKLLKVAIAAIVFSGSALAGVVPQYGGGGNHGGGG NNSGPNSELNIYQYGGGNSALALQTDARNSDLTITQH GGGNGADVGGSDSSIDLTRGFGNSATLDQWNG KNSEMTVKQFGGGNGAAVDQTASNSSVNVTVQVGF NNATAHQYGGGGSSGGGSAHIVMVDAYKPTK	174
CsgB <sup>2</sup>	MAGYDLANSEYNFAVNELSKSSFNQAAIIGQAGTNS AQLRQGGSKLLAVVAQEGSSNRAKIDQTGDYNLAYI DQAGSANDASISQGAYGNTAMIIQKGSNGKANITQYG TQKTAIVVQRQSQMAIRVTQRHHHHHHH	137
GFP <sup>3</sup>	MGHHHHHHMVSKGEEDNMASLPATHELHIFGSINGV DFDMVGQGTGNPNDGYEELNLKSTKGDLQFSPWILV PHIGYGFHQYLPYDPGMSPFQAAMVDGSGYQVHRT MQFEDGASLTVNYRYTYEGSHIKGEAQVKGTGFPAD GPVMTNSLTAADWCRSKKTYPNDKTIISTFKWSYTTG NGKRYRSTARTTYTFAKPMAANYLKNQPMYVFRKT ELKHSKTELNFKEWQKAFTDVMGMDELKLAALAE EEEEAYGWMDF	261
SpyCatcher <sup>4</sup>	MSYYHHHHHHHDYDIPTTENLYFQGAMVDTL SGLSSE QGQSGDMTIEEDSATHIKFSKRDEDGKELAGATMELR DSSGKTISTWISDGQVKDFLYPGKYTFVETAAPDGY EVATAITFTVNEQGQVTVNGKATKGDAHI	139
Est3-14(SpyCatcher) <sup>4,5</sup>	MGHHHHHHSVKPTSVM DIPLLPGR LISLPGRGEIFVR HHQHVNPDAPTLLLLHGWTASSDLQFFTAYEELSRN YSIVGVDHRGHGRGLRPNHTFSLEDCADDAAAVVRA LGIRNVITVGYSMGGPISLLVWQRHSDLVTGMVLQAT ALEWSGTRQERNKWRVMHVIDPLFRRINSPRLTRWY VRLIPRGHEINRYLPWITGELRRNDSWMISEAGRAIS RFDARGFAHTVNVPTSFVLTTLDKLVLP HKQQALAD AVRAEVVELEGDHLAPMQQPREFSWATARAVEIVVR QTNQKLG GGGSSGGGSSMSYYHHHHHHHDYDIPTTENL YFQGAMVDTL SGLSSEQGQSGDMTIEEDSATHIKFSK RDEDGKELAGATMELRDSSGKTISTWISDGQVKDFYL YPGKYTFVETAAPDGYEVATAITFTVNEQGQVTVNG KATKGDAHI	448

**Table S2** Strains and plasmids used in this study

Strains	Description	Source
DH5 $\alpha$	Cloning	Stored in our laboratory
BL21(DE3)	Protein expression	Stored in our

		laboratory
BL21:: $\Delta$ CsgA	Protein expression; with chloramphenicol resistance gene	Constructed in our previous work <sup>1</sup>
<b>Plasmids</b>	<b>Description</b>	<b>Source</b>
pET21a (+)	Ampicillin; pBR322 origin	Stored in our laboratory
pET28a (+)	Kanamycin; pBR322 origin	Stored in our laboratory
<b>Recombinant strains (Abbreviations)</b>	<b>Description</b>	<b>Source</b>
<b>EX01</b> (Amp)	BL21(DE3) pET21a- <i>CsgB</i>	This work
<b>EX02</b> (Kan)	BL21(DE3) pET28a- <i>Est3-14(SpyCatcher)</i>	This work
<b>EX03</b> (Amp+Chl)	BL21:: $\Delta$ CsgA pET21a- <i>CsgA(SpyTag)</i>	Previous work <sup>1</sup>
<b>EX04</b> (Kan)	BL21(DE3) pET28a- <i>Est3-14</i>	This work
<b>IN01</b> (Kan)	BL21(DE3) pET28a- <i>GFP</i>	Previous work <sup>6</sup>
<b>IN02</b> (Kan)	BL21(DE3) pET28a- <i>GFP-SpyCatcher</i>	Previous work <sup>6</sup>

## References

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