

Supporting Information

Protein-inorganic hybrid system for efficient his-tagged enzymes immobilization and its application in L-xylulose production

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Table S1. Synthesis of recombinant enzyme nanoflowers as enzyme-Cu(PO₄)₃·H₂O hybrids.

Method (Reference)	Synthesis conditions	Enzyme			
		HjLAD		SpNox	
		EY ^a (%)	RA ^b (%)	EY (%)	RA (%)
1 (3)	Cu (0.8 mM) and incubation of 3 days at 25 °C	66.8 ± 4.3	14.4 ± 2.1	64.2 ± 4.0	17.1 ± 1.2
2 (28)	Cu (6 mM) and incubation of 24 h at 4 °C	51.2 ± 4.1	90.0 ± 3.3	44.9 ± 3.7	71.6 ± 4.7
3 (This study)	Cu (2 mM) and incubation of 24 h at 4 °C	63.4 ± 5.8	246 ± 21	58.9 ± 5.7	144 ± 16

^a Encapsulation yield; ^b Relative activity.

Relative activity of the free enzyme was considered as 100%.

Table S2. Comparison of his-tagged enzymes immobilization through organic-inorganic hybrid systems.

Enzyme	Organic-inorganic hybrid system		RA ^a (%)	Reference
	Complex	Morphology		
Bs-ADH	Cu ₃ (PO ₄) ₂ -enzyme	Nano-flower	1.74	3
	Co ₃ (PO ₄) ₂ -enzyme	Sponge	44.0	
Ll-ADH	Cu ₃ (PO ₄) ₂ -enzyme	Nano-flower	42.0	3
	Co ₃ (PO ₄) ₂ -enzyme	Sponge	94.0	
Epoxide hydrolase	Fmoc-diphenylalanine- polyethyleneimine- silicate-enzyme	Capsule	60.0	40
ADP-dependent glucokinase	Diatom silica-enzyme	Film	90.0	41
Cellulase	Diatom silica-enzyme	Film	75.0	41
Glucose-6-phosphate dehydrogenase	Diatom silica-enzyme	Film	80.0	41
HjLAD	Cu ₃ (PO ₄) ₂ -enzyme	Nano-flower	246	This study
SpNox	Cu ₃ (PO ₄) ₂ -enzyme	Nano-flower	144	

^a Relative activity.

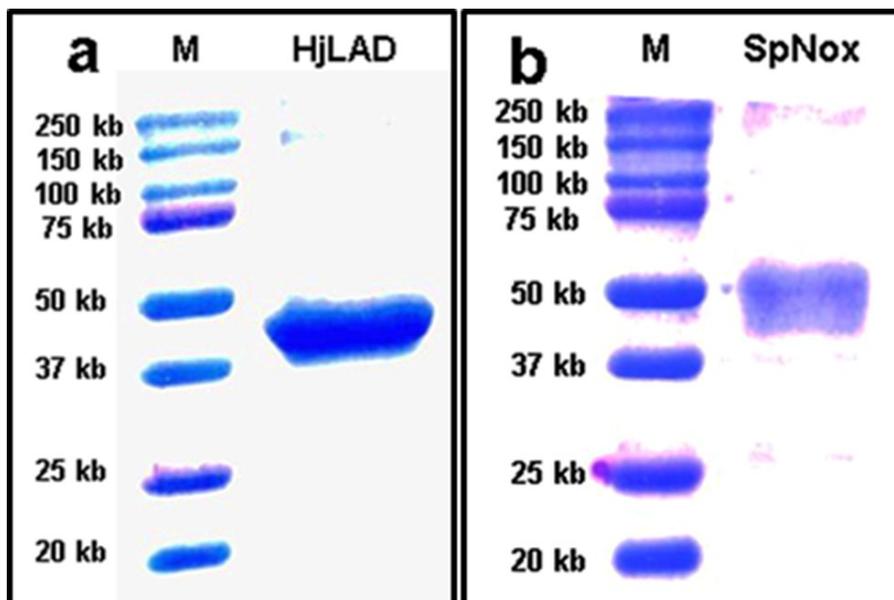


Fig. S1 SDS-PAGE analysis of purified enzymes (Lane M contains the protein markers). a) HJLAD (~41 kDa) and b) SpNox (50 kDa).

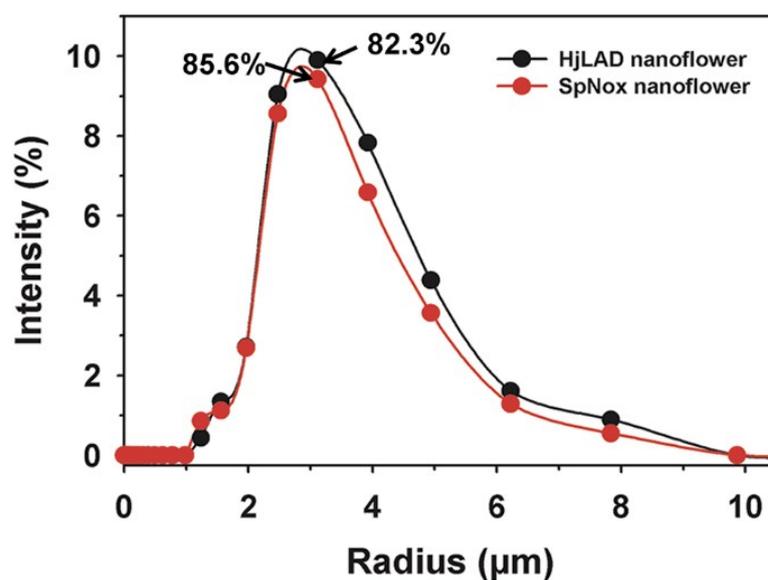


Fig. S2 DLS analysis of the particle distribution of synthesized nanoflowers.

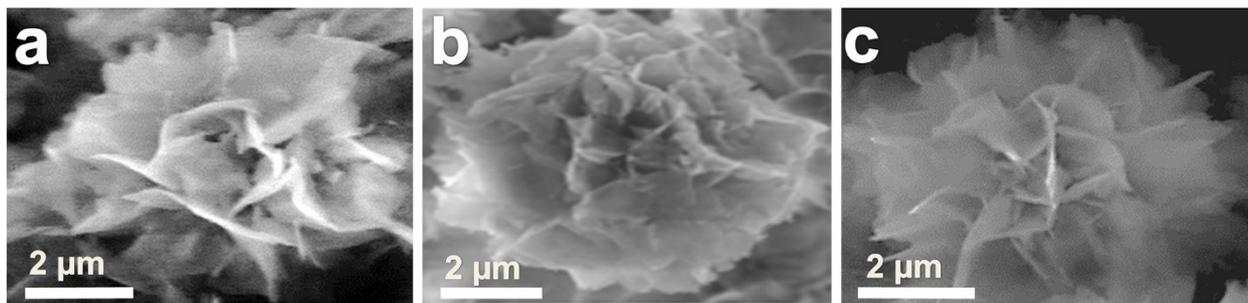


Fig. S3 FE-SEM analysis of the synthesized HJLAD nanoflower at pH 6.5 (a), 7.4 (b), and 8.0 (c) in phosphate buffer (10 mM).

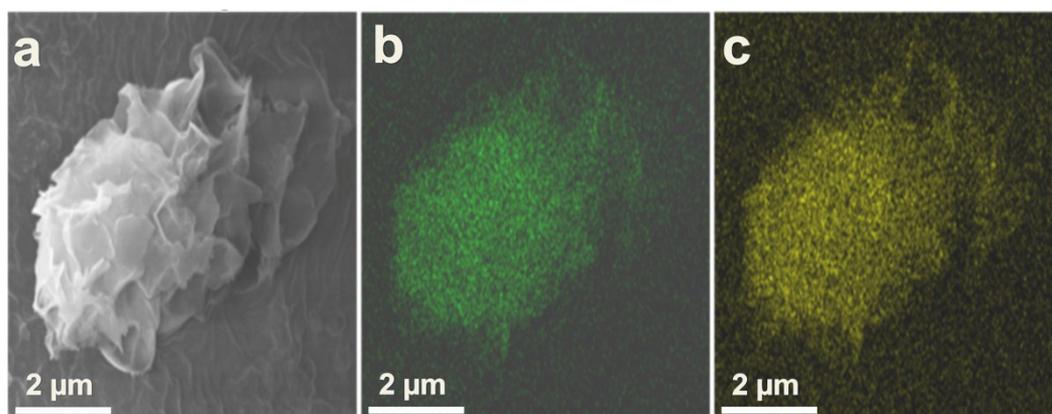


Fig. S4 FE-SEM analysis of the synthesized HJLAD nanoflower (a), and elemental mapping images of copper (b) and phosphorous (c).