Electronic Supplementary Material (ESI) for Journal of Materials Chemistry B.

Protein-directed approaches to functional nanomaterials: a case study of lysozyme

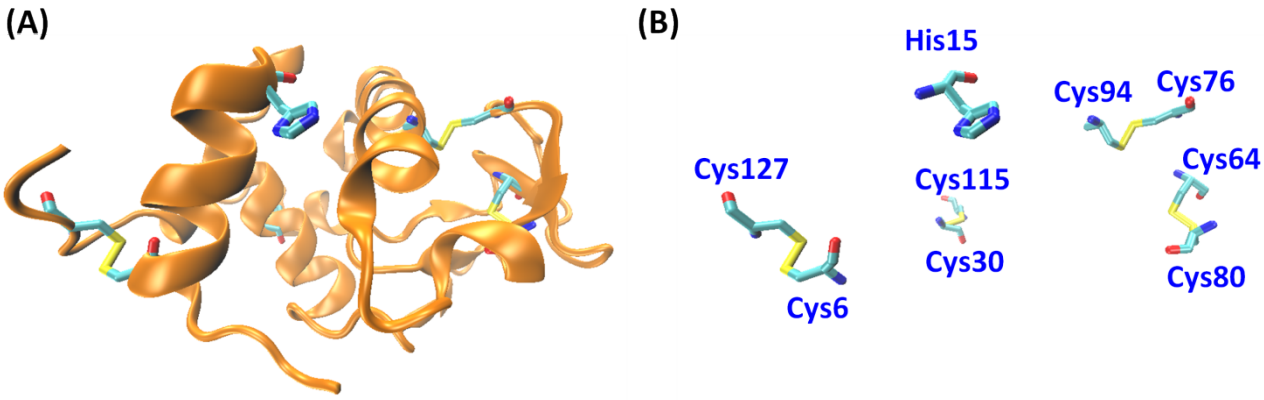

Figure S1. A X-ray crystal structure of lysozyme. (A) Overall structure with highlighted histidine and cysteine residues; (B) the highlighted histidine and cysteine residues with disulfide bonds. The structure was adopted from PDB: 2LYZ.

Lysozyme is the first enzyme to have its X-ray crystal structure solved. As shown in Figure S1, it consists of 7 helices and 1 triple-stranded β-sheet as well as irregular loops. The 8 cysteines form 4 disulfide bonds. 1 hitidine (His15) is exposed on the surface, which is ready to bind to many ligands (usually metal ions or metal complexes).[1-9] Other residues such as asparate may also bind to metal ions or metal complexes.[2, 10-15] The interaction (e.g., coordination) between these residues and metal-based precursors would direct the formation of functional nanomaterials. Lysozyme is stable over wide temperature and pH ranges. For example, the reversible thermal unfolding experiments demonstrated that lysozyme's secondary and tertiary structures were highly stable between 20°C and 64°C.[16] Lysozyme is positively charged at neutral pH due to a pI (isoelectric point) of 11.35, which would interact stronger with negatively charged species than the positively charged or neutral ones. All these factors would affect the lysozyme's roles in directing formation of nanomaterial, as discussed in the current Feature Article.

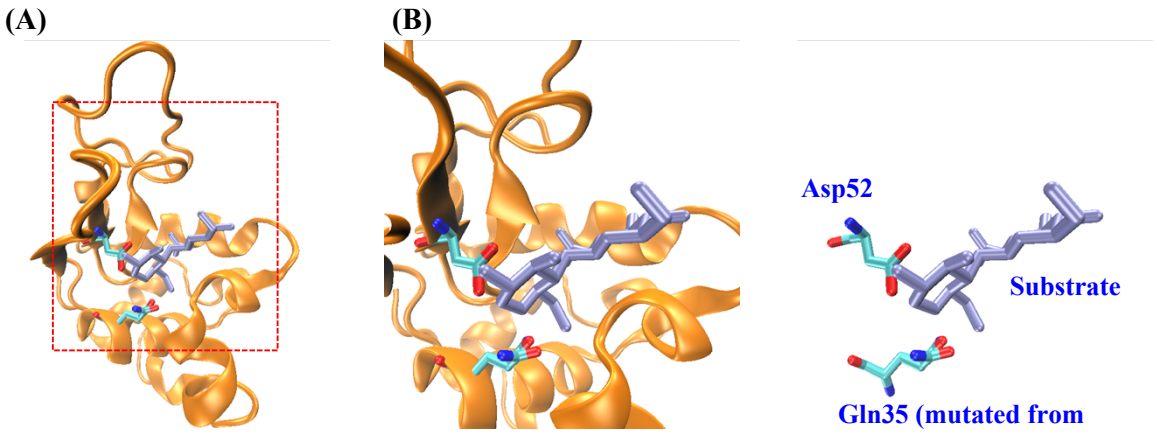

Figure S2. A X-ray crystal structure of lysozyme showing catalytic active sites as well as the binding of a synthetic model substrate. The structure was adopted from PDB: 1H6M.

Though other proteins like BSA have also been widely used to prepare lots of nanomaterials, lysozyme has its own unique features. Besides the small size and the structural characteristics discussed above (and in the main text), the enzymatic activity of lysozyme enables it to prepare nanomaterials with bioactivity. As shown in Figure S2, lysozyme can bind to its carbohydrate substrates (either from chemical synthesis or from the nature, such as cell wall of bacteria). By cleaving the bound carbohydrate substrate, lysozyme exerts its antibiotic and antibacterial functions. The functional nanomaterials with lysozyme may retain the enzymatic activity, endowing the prepared materials with multiple functions.

Table S1. Selected applications with lysozyme-directed functional nanomaterials.

Nanomaterials	**Applications**	**Ref.**	**Notes**
AuNC	Detection of Hg^{2+}	17	
	Detection of Hg^{2+}	18	
	Detection of CH_3Hg^+	18	
	Detection of cyanide ions	19	
	Protein detection	20	Combined with other protein-stabilized AuNCs to form an array for detection.
	Bacteria identification	21	
	Bacteria enrichment	22	
	Tumor cell detection	23	
AgNC	Detection of Hg^{2+}	24	
PtNC	Degradation of methylene blue	25	
Au/Ag alloy NC	Detection of Hg^{2+}	26	
AgNP	Antimicrobial activity	27	
TiO_2-SiO_2-Ag nanocomposites	Degradation of rhodamine B	28	
AuNP in single crystals	Catalysis	29	Catalytic reduction of *p*-nitrophenol to *p*-aminophenol by $NaBH_4$ was used a model reaction.
	Catalysis	30	
AgNP in single crystals	Catalysis	31	

Note: AuNC, lysozyme stabilized gold nanocluster; AgNC, lysozyme stabilized silver nanocluster; PtNC, lysozyme stabilized platinum nanocluster; Au/Ag alloy NC, lysozyme stabilized gold/silver alloy nanoclusters; AgNP, lysozyme stabilized silver nanoparticles.

Compared with other nanomaterials, the functional materials prepared via lysozyme-directed approach have several advantages in applications. First, as nanomaterials formed with assistance by other proteins, these nanomaterials are water soluble and biocompatible. Further, owning to lysozyme's enzymatic characteristics, the obtained nanomaterials may have multiple functions, for example, enabling them to bind and enrich bacteria, or even kill bacteria. In addition, the ease of crystallization makes the as-prepared functional nanomaterials ready to be recycled. And there are other potential advantages, such as lysozyme-mediated assembly, that remain to be explored in the future.

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