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#### ACCEPTED MANUSCRIPT

# Selective binding, magnetic separation and purification of histidine-tagged protein using biopolymer magnetic core-shell nanoparticles

Yang Zhou<sup>a</sup>, Dandan Yan<sup>a</sup>, Shaofei Yuan<sup>a</sup>, Yawen Chen<sup>a</sup>, Fletcher Emmanuella E<sup>a</sup>, Haifeng Shi<sup>a</sup>, Bangxing Han<sup>b,\*</sup>

<sup>a</sup> Institute of Life Sciences, Jiangsu University, No. 301 Xuefu Road, Zhenjiang 212013, P. R. China.

<sup>b</sup> College of Biological and Pharmaceutical Engineering, West Anhui University, Moon Island, Lu'an 237012, P. R. China.

ABSTRACT: In previous studies, we synthesized the magnetic core-shell structured Fe<sub>3</sub>O<sub>4</sub>/PMG/IDA-Ni<sup>2+</sup> nanoparticles. The Ni<sup>2+</sup> on the surface of nanoparticles provides abundant docking sites for histidine, and the composite nanoparticles showed potential applications in the separation and purification of histidine-tagged (His-tagged) proteins. Meanwhile, the presence of the superparamagnetic core (Fe<sub>3</sub>O<sub>4</sub>) in the nanoparticles allows them to be quickly separated and purified by an external magnetic field. Herein, the ability of magnetic nanoparticles to purify His-tagged human superoxide dismutase 1 (hSOD1) was verified. SDS-PAGE and activity data showed His-tagged hSOD1 specifically bound to Fe<sub>3</sub>O<sub>4</sub>/PMG/IDA-Ni<sup>2+</sup>, and there was no significant competition for binding between final and three intermediate products. The binding capacity of nanoparticles can reach to 62.0 mg/g (dry weight of hSOD1/nanoparticles). The nanoparticle-bound hSOD1 exhibited better thermal and storage stability compared to free hSOD1. Furthermore, the purification efficiency of the magnetic nanoparticles in the separation and purification of His-tagged proteins

E-mail address: hanbx1978@sina.com (B. Han).

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<sup>\*</sup> College of Biological and Pharmaceutical Engineering, West Anhui University, Moon Island, Lu'an 237012, P. R. China. Fax: +86 564 3305070.

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