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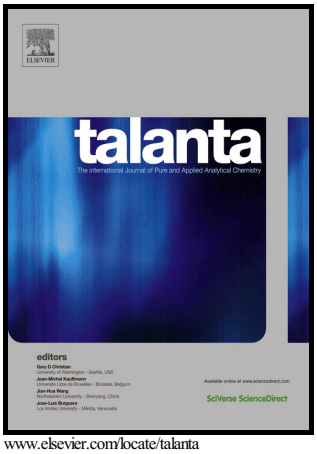

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Gold Nanoparticle Aggregation: Colorimetric Detection of the Interactions between Avidin and Biotin

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Abstract: This paper reported a novel colorimetric assay strategy for avidin and biotin interactions based on terminal protection of the biotinylated single-stranded DNA and the surface plasmon resonance adsorption of gold nanoparticles (AuNPs). In this assay, it was firstly found that biotin-ssDNA specifically bound to the target protein avidin with strong affinity could be protected from hydrolysis by exonuclease I (Exo I). Furthermore, a colorimetric strategy was designed for the detection of avidin and biotin interactions. In the process, in the presence of avidin, the interaction of avidin and biotin protected the digestion of Exo I towards the biotin-ssDNA. The biotin-ssDNA with negatively charged would attach to the surface of AuNPs with positively charge in high salt solution through electrostatic interactions, which prevented AuNPs to aggregate. With the increased addition of avidin, the absorbance of AuNPs in 520 nm increased gradually and the color showed gradually wine red. By taking advantage of terminal protection, the developed strategy could offer high sensitivity for detecting small molecule-protein interactions. The results revealed that the developed strategy was highly sensitive for detecting avidin in the concentration ranging from 0.01-0.2 μg/mL with the detection limit of 4×10^{-3} μg/mL.The developed assay also showed highly specific, cost-efficient and convenient. Moreover, this strategy only required labeling the small molecule on a single-stranded DNA, circumventing protein modifications that might be harmful for activity. In view of these advantages, this new colorimetric method could have potential to become a universal, sensitive, and selective platform for detection of small molecule−protein interactions.

Keywords: biotin, avidin, Au nanoparticles, colorimetric detection