An ultra-sensitive and colorimetric sensor for copper and iron based on glutathione-functionalized gold nanoclusters

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HIGHLIGHTS

- We developed a simple system for Fe^{3+} and Cu^{2+} sensing with low cost and easy operations.
- Fe^{3+} and Cu^{2+} are simultaneously detected via colorimetric readout and fluorescent absorbance signals.
- The sensor realized the simultaneously detection of Fe^{3+} and Cu^{2+} successively with highly sensitivity and selectivity.

GRAPHICAL ABSTRACT
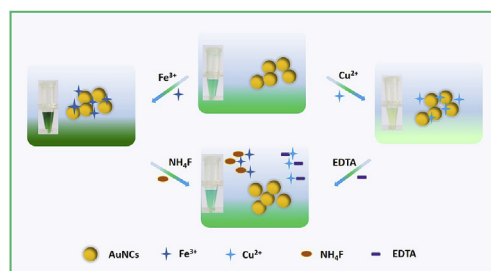
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ABSTRACT

Here, we report an ultra-sensitive and colorimetric sensor for the detection of Fe^{3+} or Cu^{2+} successively using glutathione-functionalized Au nanoclusters (GSH-AuNCs). For GSH-AuNCs can catalytically oxidize peroxidase substrates, such as 3, 3′, 5, 5′-tetramethylbenzidine (TMB), colored products are formed in the presence of H_2O_2. While upon the addition of Fe^{3+} or Cu^{2+} into the GSH-AuNCs-TMB-H_2O_2 system, diverse color and absorbance of the system was obtained due to the self oxidation of Fe^{3+} and the inhibition of peroxidase-like activity of GSH-AuNCs. With the introduction of ethylene diamine tetraacetic acid (EDTA) or ammonium fluoride (NH_4F) to GSH-AuNCs-TMB-H_2O_2+Cu^{2+} system or GSH-AuNCs-TMB-H_2O_2+Fe^{3+} system respectively, a restoration of color and absorbance of system was realized. On the basis of above phenomenon, a colorimetric and quantitative approach for detecting Fe^{3+} and Cu^{2+} was developed with detection limit of 1.25×10^{-9} M and 1.25×10^{-10} M respectively. Moreover, the concentration of Fe^{3+} and Cu^{2+} in human serums was also accurate quantified by this method. So this design strategy realized the simple and simultaneous detection of Fe^{3+} and Cu^{2+}, suggesting significant potential in clinical diagnosis.

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1. Introduction

The detection of heavy transition metal ions is of utmost importance because of their potential applications in biological systems [1,2]. As widely used metals, copper and iron are essential

trace elements in various biological processes. Imbalance of copper and iron homeostasis may leads to a wide variety of health's disease. For example, the deficiency of Cu^{2+} may lead to neurological problems [3–5], such as Alzheimer's and Parkinson's, but there will be liver and kidney damage [6–8] under the excessive uptake of copper due to the potential toxicity to living organisms of free Cu^{2+}. The anomaly of iron ion content will lead to the hemolytic anemia, peptic ulcer and uremia [9]. Therefore, the identification and measurement of copper and iron is highly desired as an important issue in biological fluids and clinical research, which has attracted the sustained attention of scholars.

So far, extensive available methods have been developed for determining of copper and iron, such as atomic absorption spectrometry (AAS) [10,11], inductively coupled plasma mass spectrometry (ICP-MS) [12,13], electrochemical sensors [14], DNA enzymes [15], fluorescence technique [16,17] and so on. Although these methods provide excellent sensitivity and admirable selectivity, they are limited by the strong dependence on large-scale instruments and expensive cost and complicated procedures, which prevents them from being used in outside laboratory applications. More importantly, those detection sensors generally realized the one kind of metal ion detection, two or more kinds of ions successive or simultaneous detection was rarity. Thus, the development of portable but simultaneously detection sensors, which give satisfactory responses at highly sensitivity and selectivity are still of continued interest and urgently needed.

The metal nanoclusters (NCs), with a core size below 2 nm, have been widely studied much in recent years, owing to their promise for diverse applications such as sensing, biological labeling, imaging and therapy [18–23]. The gold nanoclusters have been reported for the possess of intrinsic peroxidase-like activity [24] that can catalyze the reaction of peroxidase substrate 3,3,5,5-tetramethylbenzidine (TMB) in the presence of H_2O_2 to produce a blue color reaction [24]. Herein, we developed GSH-AuNCs−TMB−H_2O_2 system for simple and selective colorimetric sensing of Fe^{3+} and Cu^{2+} according to the self oxidation of Fe^{3+} and the inhibition of peroxidase-like activity of GSH-AuNCs. With the introduction of ethylene diamine tetraacetic acid (EDTA) or ammonium fluoride (NH$_4$F) to GSH-AuNCs-TMB-H_2O_2+Cu^{2+} system or GSH-AuNCs-TMB-H_2O_2+Fe^{3+} system respectively, a restoration of color and absorbance may be found. When the Fe^{3+} and Cu^{2+} simultaneously present in the GSH-AuNCs−TMB−H_2O_2

system, the selectively masking of Fe^{3+} through NH$_4$F realized the detection of Fe^{3+} and the introduction of EDTA subsequently achieved the detection of Cu^{2+}. Moreover, this colorimetric system was applied to the detection of human serums successfully with high accuracy. So we developed a simple, reliable, and selective colorimetric sensing technique of Fe^{3+} and Cu^{2+} by utilizing GSH-AuNCs as a platform (Scheme 1).

2. Experimental

2.1. Materials and chemicals

HAuCl$_4$·3H$_2$O, glutathione (GSH), 3,3′,5,5′-tetramethylbenzidine (TMB) were obtained from Aladdin Industrial Corporation (Shanghai, China) and used without further purification. Ethylene diamine tetraacetic acid (EDTA), ammonium fluoride (NH$_4$F), cupric chloride (CuCl$_2$), ferric chloride (FeCl$_3$), hydrogen peroxide (H$_2$O$_2$, 30 wt %) were purchased from Beijing Dingguo Biotechnology Co., Ltd. The aqueous solutions were prepared with Milli-Q water (18 MΩ, Millipore Co., USA).

2.2. Preparation of GSH-Au NCs

The water-solubility GSH-modified Au NCs were synthesized following the method as described in the literature reported earlier. In a typical protocol, 0.50 mL of 20 mM freshly prepared HAuCl$_4$·3H$_2$O and 0.15 mL of 0.1 M GSH aqueous solutions were added in 4.35 mL of distilled water under gentle stirring in sequence at room temperature in around bottomed flask. Then the reaction was allowed to proceed at 80 °C for 20 h under vigorous stirring followed by cooling to room temperature naturally. The final GSH-AuNCs solution was kept at 4 °C prior to use.

2.3. Analytical procedures of the GSH-AuNCs peroxidase-like activity

In a typical test, firstly, 200 μL of AuNCs solution was mixed with TMB (100 μL, 5 mM) solutions and H$_2$O$_2$ (50 μL, 50 μM). The mixture was incubated at room temperature for 1 h and the absorption measurements were conducted after the full reaction. There was no AuNCs or H$_2$O$_2$ in the control experiments. Secondly, 50 μL of 0.01 M Fe^{3+} or 50 μL of 0.01 M Cu^{2+} solutions was mixed with the AuNCs before the addition of TMB and H$_2$O$_2$. After 1 h incubation,
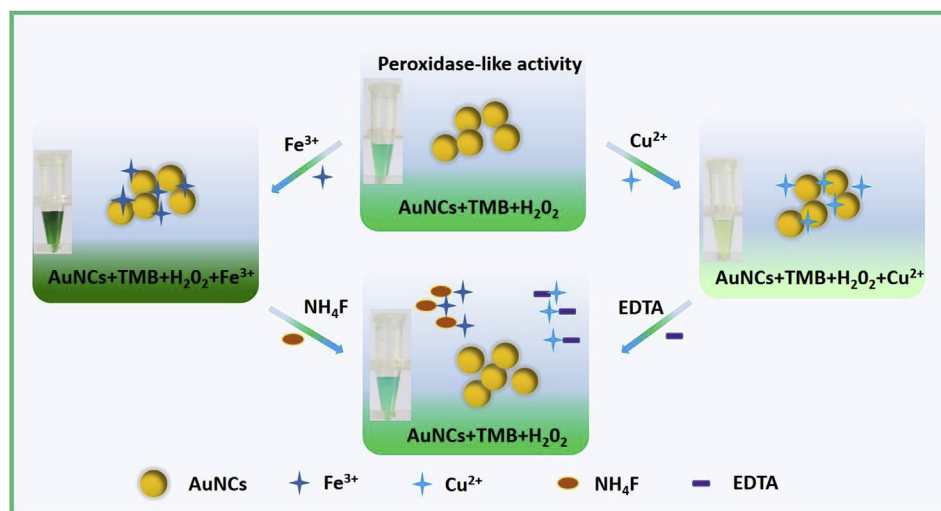
Scheme 1. Schematic diagram of the colorimetric and sensitive detection of Fe^{3+} and Cu^{2+} based on the peroxidase-like activity of GSH-AuNCs.