Colloids and Surfaces A: Physicochem. Eng. Aspects 468 (2015) 333–338

Contents lists available at ScienceDirect

Colloids and Surfaces A: Physicochemical and Engineering Aspects

journal homepage: www.elsevier.com/locate/colsurfa

Highly sensitive fluorescence detection of copper ion based on its catalytic oxidation to cysteine indicated by fluorescein isothiocyanate functionalized gold nanoparticles

Shasha Wang[a,c], Xiaokun Wang[a,b], Zhiyang Zhang[a,c], Lingxin Chen[a,*]

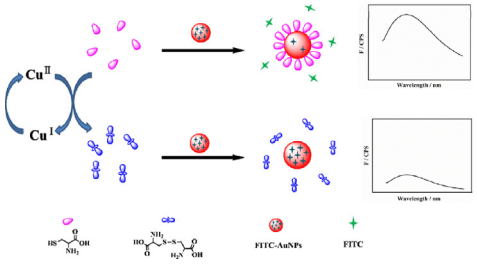
[a] Key Laboratory of Coastal Environmental Processes and Ecological Remediation, Yantai Institute of Coastal Zone Research, Chinese Academy of Sciences, Yantai 264003, China
[b] College of Chemistry and Chemical Engineering, Qufu Normal University, Qufu 273165, China
[c] University of Chinese Academy of Sciences, Beijing 100049, China

HIGHLIGHTS

- A fluorescent probe for Cu^{2+} has been developed by FITC functionalized gold nanoparticles.
- Cysteine could replace FITC from the surfaces of gold nanoparticles and Cu^{2+} could catalyze O_2 oxidation of cysteine.
- The fluorescent probe provides high sensitivity toward Cu^{2+} in drinking water as a real sample.

GRAPHICAL ABSTRACT

A highly sensitive fluorescent sensor for Cu^{2+} based on its catalytic oxidation of cysteine and cysteine-induced increase in fluorescence intensity of FITC-AuNPs.



ARTICLE INFO

Article history:
Received 3 September 2014
Received in revised form 2 December 2014
Accepted 22 December 2014
Available online 8 January 2015

Keywords:
Gold nanoparticles
Fluorescein isothiocyanate
Fluorescence resonance energy transfer
Copper ions
Fluorescence

ABSTRACT

An innovative fluorescence method for sensitive detection of copper ion (Cu^{2+}) was developed based on fluorescein isothiocyanate functionalized gold nanoparticles (FITC-AuNPs). Due to the stronger binding affinity of isothiocyanate functional group to gold, FITC molecules could adsorb on the surface of AuNPs, forming a simple fluorescence resonance energy transfer (FRET) system, and the fluorescence intensity of FITC was remarkably quenched. Upon adding cysteine, FITC could be displaced from the surface of AuNPs because the formation constant (K_f) of Au–S linkage ($K_f(AuS^-) = 4 \times 10^{35}$) was much higher than Au–SCN linkage ($K_f(Au(SCN)^{2-}) = 10^{23}$), leading to the recovery of fluorescence intensity. However, Cu^{2+} could catalyze O_2 oxidation of cysteine, and the generated disulfide cystine could not remove FITC from AuNPs' surface. Therefore, the recovery of fluorescence intensity was much weaker when compared with that of in the absence of Cu^{2+}. And on the basis of this principle the concentration of Cu^{2+} could be detected quantitatively. Under optimal conditions, our method exhibited high selectivity toward Cu^{2+} and provided a good linear relationship in the range of 1.0–17.0 nM with the detection limit of 0.37 nM calculated by $3\sigma/S$. Furthermore, complicated synthetic procedures and poor water solubility could be ignored in this proposed fluorescent sensor.

1. Introduction

As an indispensable trace element in human body, copper ion (Cu^{2+}) plays an important role in the development and function of

* Corresponding author. Tel.: +86 535 2109130; fax: +86 535 2109130.
E-mail address: lxchen@yic.ac.cn (L. Chen).

http://dx.doi.org/10.1016/j.colsurfa.2014.12.050
0927-7757/

internal organs [1], such as brain, liver and heart. And it also activates the formation of hemoglobin and promotes the absorption and utilization of iron [2]. However, the excessive accumulation of copper is confirmed to be poisonous to humans, in that it can lead to neurodegenerative diseases probably by its involvement in the generation of active oxygen species [3,4]. Accordingly, the safe limit of Cu^{2+} is 1.3 ppm (~20 μM) in drinking water recommended by U.S. Environmental Protection Agency (EPA) [5]. The common analytical methods adopted for Cu^{2+} measurement include atomic emission spectrometry (AES) [6], atomic absorption spectrometry (AAS) [7], inductively coupled plasma mass spectrometry (ICP-MS) [8] and electrochemistry assay [9]. In spite of high selectivity and sensitivity, these methods often require professional operational skills, tedious sample pretreatments and a long analysis time. As a consequence, there has been growing interest in developing new methods for monitoring Cu^{2+} with high selectivity and sensitivity.

Recently, numerous chemosensors and biosensors have been designed based on nano-materials, e.g. gold nanoparticles (AuNPs) [10–12], gold nanorods (AuNRs) [13–15], carbon dots (CDs) [16–18] and quantum dots (QDs) [19,20]. Among various nano-materials, AuNPs have attracted a great deal of attention for chemical and biological analysis over past decades. Due to their unique optical properties, especially localized surface plasmon resonance (LSPR) [21], AuNPs were widely applied in colorimetric [10,11], fluorescent [22], surface-enhanced Raman scattering [23] and electrochemistry assays [24]. What's more, possessing higher extinction coefficient than those of common organic dyes in the ultraviolet and visible regions [25], AuNPs are emerging as efficient quenchers for fluorophores.

On account of high sensitivity and relative versatility, fluorescent assay has been gained comprehensive attention. However, complicated synthetic procedures and poor water solubility limit the application of most of existing fluorescent methods, which are only based on the design and synthesis of organic compounds. AuNPs have opened up a new alternative for the fabrication of fluorescent chemosensors. Because of AuNPs' high extinction coefficient, fluorophores attached to the surfaces of AuNPs will be quenched by fluorescence resonance energy transfer (FRET) [26]; when added analytes which have much stronger affinity with AuNPs, fluorophore molecules will be replaced, leading to the increase of fluorescence intensity. So far, AuNPs-FRET-based assay has been used to develop sensors for the detection of metal ions [27], inorganic anions [28], small organic molecules [29] and biomolecules [30].

Herein, a simple and sensitive method for fluorescence detection of Cu^{2+} in aqueous solution at room temperature was proposed based on fluorescein isothiocyanate functionalized gold nanoparticles (FITC-AuNPs). The fluorescence of FITC switched off when attached to the surfaces of AuNPs through forming Au–SCN linkage; upon adding cysteine, FITC was replaced because the Au–S linkage was stronger than Au–SCN linkage, and the fluorescence turned to switch-on. Yet, the oxidation of cysteine by O_2 could be catalyzed with the existence of Cu^{2+}, resulting in the formation of disulfide cystine [31,32]. At this point, the recovery of fluorescence was weakened when adding AuNPs. Based on this principle, the quantitative analysis of Cu^{2+} could be realized and this proposed method also showed high sensitivity toward Cu^{2+} over other metal ions.

2. Materials and methods

2.1. Chemicals

Fluorescein isothiocyanate (FITC, 96%), $Na_2HPO_4{\cdot}12H_2O$ and $NaH_2PO_4{\cdot}2H_2O$ were purchased from Aladdin. Hydrogen tetrachloroaurate (III) hydrate ($HAuCl_4{\cdot}4H_2O$), sodium citrate, cysteine, $CuSO_4{\cdot}5H_2O$, $MgCl_2$, $CaCl_2$, $FeCl_3$, $NiCl_2$, $ZnCl_2$, $Cd(ClO_4)_2$, LiCl, $MnCl_2$, KCl, $BaCl_2$, $AgNO_3$, $Pb(NO_3)_2$ and $HgCl_2$ were obtained from Sinopharm Chemical Reagent Company Limited (Beijing, China). All the reagents were of analytical grade and used without any future purification.

2.2. Apparatus

Solutions were prepared with double-deionized water (18.2 MΩ cm specific resistance) obtained by a Cascada LS Ultrapure water system (Pall Corp., USA). Transmission electron microscope (TEM) images were captured on a JEM-1230 electron microscope (JEOL Ltd., Japan) operating at 100 kV. UV–vis absorption spectra were collected on a Thermo Scientific NanoDrop 2000C spectrophotometer (Gene Company Ltd., USA). The fluorescence spectra were recorded on a Fluoromax-4 spectrofluorometer with a xenon lamp and 0.5 cm quartz cells (HORIBA Scientific, Japan).

2.3. Synthesis and modification of AuNPs

All glasswares used in the following experimental procedure were bathed in freshly prepared 3:1 HCl–HNO_3, rinsed thoroughly with double-deionized water and dried in air. AuNPs were prepared by the citrate-mediated reduction of $HAuCl_4$ according to Frens' method [33]. Typically, 100 mL of 1.0 mM $HAuCl_4$ was introduced into a three-necked flask and heated to reflux with stirring. Then 10 mL of 38.8 mM sodium citrate was rapidly added to the boiling solution, resulting in a color change from pale yellow to deep red. The solution was kept boiling for another 30 min, and then cooled to room temperature with continuous stirring. The concentration of the obtained AuNPs solutions was estimated to be 12 nM according to Beer's law ($A = \varepsilon bc$), where the colorimetric cuvette had a path length (b) of 1 cm and the extinction coefficient (ε) of 13 nm AuNPs at 520 nm is $2.78 \times 10^8\ M^{-1}\ cm^{-1}$.

A stock solution of 1.0 mM FITC was prepared in ethanol absolute. Then, 50 μL of the prepared FITC solution was added to the 50 mL AuNPs solution with stirring, and the mixture was equilibrated in the dark at room temperature.

2.4. Fluorescence detection of Cu^{2+}

A stock solution of $CuSO_4{\cdot}5H_2O$ (0.01 M) was prepared in double-deionized water, and different concentrations of Cu^{2+} solution was acquired by diluting the stock solution accurately. For Cu^{2+} detection, 10 μL various concentrations of Cu^{2+} solution was added to 890 μL Na_2HPO_4–NaH_2PO_4 buffer (10 mM) at a pH of 6.8 containing 3.0 μM cysteine; the mixture solution was incubated at room temperature for 30 min. Then 100 μL FITC-AuNPs was added to the mixture solution. 10 min later, fluorescence spectra were collected at 514 nm with an excitation wavelength of 490 nm.

To measure the selectivity of the developed method, other metal ions instead of Cu^{2+} were detected in a similar way under the same optimized conditions.

2.5. Fluorescence sensing of Cu^{2+} in drinking water

The drinking water was obtained locally and 5-fold diluted, then this water sample was used to prepare 10 mM Na_2HPO_4–NaH_2PO_4 buffer at a pH of 6.8 spiked with standard Cu^{2+} solutions leading to different final concentrations. 10 μL of 300 μM cysteine was dissolved in 890 μL above buffer solution. After incubation for 30 min, 100 μL FITC-AuNPs was added to the solution and the mixed solution was equilibrated for another 10 min before spectra measurement.