Highly selective, colorimetric detection of Hg^{2+} based on three color changes of AuNPs solution from red through sandy beige to celandine green

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Abstract: A new colorimetric method of detecting Hg^{2+} in aqueous solutions was proposed, based on the three color changes of gold nanoparticles (AuNPs) with the assistance of ascorbic acid (AA) and multi-sulfhydryl functionalized hyperbranched polyethylenimine (SH-HPEI). The mechanism of sensing is based on the formation of Au-Hg alloy and its aggregation, along with the corresponding three color changes from red (AuNPs) through sandy beige (Au-Hg alloy) to celandine green (larger Au-Hg alloy). Compared with usual colorimetric assays with only one color change progress from red to blue or red to purple of reported AuNPs, this assay is highly selective for Hg^{2+} detection due to the fact that the appearing probability of identical three color changes triggered by interfering agents or the possibility of false positive results is very, very low. The detection limit of 8.76×10^{-9} M is lower than the guideline value of Hg^{2+} (30 nM) in drinking water defined by the World Health Organization (WHO). Meanwhile, the concentration range (from 8.76×10^{-9} to 1.27×10^{-4} M) of this new detection method is the widest one that has been reported. Moreover, this method is successfully applied to detecting Hg^{2+} in real water sample. More importantly, this novel colorimetric sensor based on multiple color changes may open a new avenue for the development of Hg^{2+} sensing probes.

Keywords: colorimetric method, gold nanoparticles, Au-Hg alloy, multi-sulfhydryl functionalized hyperbranched polyethylenimine

1. Introduction

At present, mercury contamination is of great concern all over the world because of its severe hazardous effects to the environment and human health. Hg^{2+} is considered as one of the most toxic heavy metal ions because of its strong affinity to sulfur atoms in proteins and enzymes, which causes dysfunction of brain, liver, kidney, gastrointestinal tract, etc, resulting in many different diseases.^[1, 2] Therefore, there is a strong need to detect Hg^{2+} accurately and timely, in order to solve the problem of early forecast and determination of mercury contamination.

To date, a variety of approaches, such as inductively coupled plasma mass spectrometry,^[3] fluorescence spectroscopy,^[4, 5] atomic absorption spectrometry^[6] and electrochemical sensor^[7] have been developed for Hg^{2+} detection. But all of these analytical methods are carried out by means of expensive instrument and complicated sample preparation processes in certain cases. Recently, rhodamine^[8] and metal nanoparticles^[9-11] based colorimetric sensors without resorting to expensive equipment or complicated operations have attracted increasing attention in detection of Hg^{2+} . Compared with other metal nanoparticles, gold nanoparticles (AuNPs) are preferred for colorimetric sensors due to its high absorption extinction coefficient, strong surface plasmon resonance absorption and colour-tuneable optical properties. For example, Willner et al. developed two different optical methods to analyze Hg^{2+} ions using aggregated AuNPs and a DNA-based machine with a limit of detection (LOD) of 10 nM and 1 nM, respectively.^[12] This aggregation can trigger a color change from red to blue owing to the interparticle coupled plasmon excitons in the aggregated states. Fan et al. reported a oligo-T/AuNPs-based gold nanoprobe that could respond to Hg^{2+} within microfluidic channels with an obvious color change from red to purple and a LOD of 5 μM .^[13] Chen et al. developed a colorimetric sensor composed of oligopeptide and AuNPs for detecting Hg^{2+} on the basis of the thiophilic nature of Hg^{2+} and the color change of the AuNPs solution from red to purple.^[14] Tseng et al. found that the citrate of the AuNPs surface could reduce Hg^{2+} to Hg^0 which can be deposited onto AuNPs surface and displace the Tween 20 stabilizer, resulting in the aggreration of AuNPs with a red to blue color change.^[15] Inspired by these, Chen et al. developed a colorimetric method with a blue-to-red color change for detecting Hg^{2+} facilitated by ascorbic acid (AA) and N-acetyl-L-cysteine.^[16] The detection mechanisms of most reported colorimetric sensors are based on the color change from red-to-blue (purple) or blue-to-red induced by the aggregation-to-dispersion or dispersion-to-aggregation of AuNPs in the presence of Hg^{2+} . However, the aggregated state of the AuNPs is unstable and the aggregated AuNPs will be precipitated out over time. Therefore, the persistence time of the color of the AuNPs aggregates is short, which means that the color change phenomenon can only be observed during a certain time period, limiting their sensing applications. In addition, it is worthwhile to point out that many other external factors, such as highly acidic environment, high-salt medium and so on, can trigger the aggregation of AuNPs with appearing in blue or purple, that

is, the selectivity and tolerance of the assay is poor.

To achieve higher selectivity, the colorimetric sensors based on the color change of AuNPs from red to other color become good candidates for Hg^{2+} detection. Very recently, several groups including ours proposed that modified AuNPs could serve as colorimetric sensors for Hg^{2+} detection by the formation of Hg–Au alloys in the presence of AA. Han et al. presented a colorimetric method of detecting Hg^{2+} based on the color change of CTAB-coated nanogold from red (purple) to sandy beige in the presence of AA, with a LOD of 1 μM by the naked eye and 30 nM by UV–visible light spectroscopy.^[17] Lee et al. reported that graphene–gold nanocomposite (G–AuNPs) can be used as colorimetric sensor for the determination of Hg^{2+} in the presence of AA. The color of the G–AuNPs suspension changed from purple–red to light brown due to the formation of gold amalgam.^[18] By virtue of the specific amalgam of reduced Hg^{2+} with thermoresponsive hyperbranched polymer capped AuNPs, our group found that the color change of this system from red to fawn color can be used as colorimetric probe to detect Hg^{2+} .^[19] Although these methods based on the change from red to other color rather than red to blue or purple show good anti-interference in acidic environment or high-salt medium, and some other problems remain. For example, the assays could also show an obvious response to other metal ions besides Hg^{2+} . Reported colorimetric detection of Ag^+ based on AuNPs stabilized by hyperbranched polyethylenimine derivative also showed similar color change (red to brown).^[20] Let's imagine if one colorimetric probe for Hg^{2+} testing occur successively three color changes during the detecting process. The specificity and selectivity of this colorimetric sensor should be greatly improved owing to the extremely low possibility of false positive results. In other words, the possibility of occurrence of identical three color changes induced by interfering agents is quite low and almost zero. Therefore, the design and fabrication of AuNPs-based colorimetric sensors toward Hg^{2+} with three color changes and high selectivity are very meaningful and useful for promoting the development of detection technique.

In this study, we developed a new colorimetric method of Hg^{2+} detection based on the color change of AuNPs in the presence of AA and multi-sulfhydryl functionalized hyperbranched polyethylenimine (SH–HPEI). In the presence of AA, Hg^{2+} is reduced to Hg^0 which can deposit onto the AuNPs surface, resulting in the formation of Hg–Au alloy with a red to sandy beige color change. Importantly, when multi-sulfhydryl functionalized hyperbranched polyethylenimines with a large number of thiol and amine groups are added to the solution, Hg–Au alloys aggregate together along with a color change from sandy beige to celandine green, realizing the colorimetric detection of Hg^{2+} . As far as we are aware, this is the first colorimetric sensor for Hg^{2+} detection with high-selectivity and high-sensitivity based on three color changes during the sensing process.

2. Experimental Section

2.1 Materials.

Hyperbranched polyethylenimine (HPEI) with a $\overline{M}_n$ of 10^4 g/mol was purchased from Aldrich. Hydrogen tetrachloroaurate (III) tetrahydrate ($\text{HAuCl}_4 \cdot 4\text{H}_2\text{O}$) was purchased from Tianjin Yingda chemical company. Methyl mercaptoacetate, ascorbic acid (AA), sodium citrate and other chemicals were purchased from Shanghai Sinopharm Chemical Reagent Corporation. The water used in all of the experiments was doubly distilled.

2.2 Apparatus and instruments.

^1H NMR spectra of the SH-HPEI were obtained from a Varian INOVA 500MHz spectrometer. Ultraviolet–visible (UV–vis) absorption spectra were recorded on a T6 UV/Vis Spectrophotometer (Purkinje General, China). Transmission electron microscopy (TEM) analysis was carried out on a Philips TECNAI G2 F20 electron microscope operating at an accelerating voltage of 200 kV. The hydrodynamic size of the particles was measured by dynamic light scattering (Zetasizer Nano-ZS90, Malvern Instruments). The concentration of Hg^{2+} was determined by an Inductively Coupled Plasma spectrometer (ICP-9000, Shimadzu).

2.3 Synthesis of multi-sulfhydryl functionalized hyperbranched polyethylenimine (SH-HPEI).

Under the atmosphere of nitrogen, 8 mL of 0.73 M methyl mercaptoacetate solution was added to 5 mL of 0.01 M HPEI solution. Vigorous stirring of the above solution was continued for 24 h at 25 °C. And then the mixture was reacted for 1.5 h at 40 °C to finalize the reaction. The desired SH-HPEI was purified by dialysis against water using a benzoylated cellulose membrane (MWCO 1000 g/mol). Finally, the solvent was removed under vacuum at 50 °C, and the solid product with a pale yellow color was obtained.

2.4 Preparation of AuNPs.

AuNPs were synthesized by the classical citrate reduction of HAuCl_4 with necessary modification.^[19, 20] Under vigorous stirring, after 121 mL of 3.1 mM HAuCl_4 aqueous solution was heated to reflux in a three-necked flask, 10 mL of 30.6 mM sodium citrate was added to the boiled solution. After continuous boiling for 30 min, the mixture became wine red and was cooled to ambient temperature. The average size and the concentration of AuNPs determined by TEM and Lambert-Beer's law according to the reported molar extinction coefficient ($2.7 \times 10^8 \text{ M}^{-1} \text{ cm}^{-1}$ at 520 nm)^[21] were about 14 nm and 2.7 nM, respectively.

2.5 Colorimetric sensing of Hg^{2+} .

For Hg^{2+} determination, AA (0.5 mL, 1.2-50.1 mM) was added to a solution containing AuNPs (0.125-1.3 mL, 2.7 nM) and Hg^{2+} (1.0 mL, 39.4 nM-77.4 mM) with the first color change from red to sandy beige. Then, 1 mL of SH-HPEI (0.3 mg/mL) was added to the above solution with the second color change from sandy beige to celandine green. The total volume of each sample was fixed to 4.5 mL. After equilibrating at room temperature for 5 min, the color changes and the absorption spectra were recorded and measured by a digital camera and a UV-vis spectrophotometer, respectively.

All detecting experiments were performed in triplicate. For TEM testing, the resulting mixture was subjected to centrifugation at 17 000 rpm for 10 min, removing the free SH-HPEI polymer.

2.6 Selectivity of this colorimetric sensor.

To examine the detection selectivity, other ions including Na^+ , Sr^{2+} , Cs^+ , Co^{2+} , Mg^{2+} , Cd^{2+} , Ni^{2+} , Mn^{2+} , Ag^+ , Ba^{2+} and Cu^{2+} were tested under the same experimental conditions.

2.7 Hg^{2+} detection in tap water.

This colorimetric sensor was used for the detection of Hg^{2+} in tap water obtained locally to evaluate its practicality. A series of samples were prepared by adding standard solutions of Hg^{2+} to the tap water. These prepared samples were added to a mixture containing 0.5 mL AuNPs and 0.5 mL ascorbic acid (39.8 mM). After the first color change, 1 mL of SH-HPEI (0.3 mg/mL) was added to the above solution, and the mixed solution was incubated for 5 min before taking photo and spectra measurement.

3. Results and discussion

3.1 Synthesis and Characterization.

Scheme 1. Synthesis of SH-HPEI and proposed Hg^{2+} sensing mechanism based on the formation of Au-Hg alloys

SH-HPEI was obtained from the aminolysis reaction between HPEI and methyl mercaptoacetate (Scheme 1). The final SH-HPEI polymer containing thiol groups was characterized via ^1H NMR (Fig. S1) and FTIR spectroscopies (Fig. S2). As shown in Fig. S1A, SH-HPEI exhibits a new signal located at 3–4 ppm coming from the methylene protons of the thiol ethyl amide groups (f in Fig. S1A, $-\text{NH}-\text{CO}-\text{CH}_2-\text{SH}$) and the methylene protons of amides (g in Fig. S1A, $-\text{CH}_2-\text{NH}-\text{CO}-$), along with the disappearance of $-\text{NH}_2$ and NH peaks (a and b in Fig. S1B) of HPEI reactant. Meanwhile, the substitution degree of the SH groups relative to the amine groups of HPEI is about 26.95% calculated from the integral of the NMR data,^[22] which indicates that the HPEI-SH has not only a large number of thiol groups but also many unreacted amine groups. Importantly, the thiol and amine groups are all strong binding groups to Au and can improve the stability of AuNPs and AuNPs composites. The successful synthesis of SH-HPEI was further confirmed by the comparison of FTIR spectra of HPEI and SH-HPEI. In comparison to HPEI, SH-HPEI exhibits new bands at around 1638 and 2570 cm^{-1} corresponding to the characteristic C=O stretching vibrations of the $-\text{CONH}-$ groups and $-\text{SH}$ stretching vibration, respectively. These characterization results indicate that the SH-HPEI containing a great number of thiol and amine groups is successfully prepared by the reaction of HPEI with methyl mercaptoacetate.

3.2 Sensing mechanism.

The proposed sensing mechanism for Hg^{2+} is illustrated as shown in Scheme 1. Originally, the AuNPs prepared by classical citrate reduction method exhibits red (image a in Fig. 1A) and an obvious surface plasmon resonance (SPR) peak occurs at around 525 nm (curve a in Fig. 1B). With the addition of Hg^{2+} , the color (image b in Fig. 1A) and the spectrum (curve b in Fig. 1B) are about the same as those of pure AuNPs. After the addition of AA, the solution immediately turns to sandy beige (image c in Fig. 1A) along with the emergence of a short wavelength band at 500 nm (curve c in Fig. 1B). Interestingly, upon the addition of SH-HPEI, the solution then changes from sandy beige to celandine green (image d in Fig. 1A) by following the decrease in absorbance of the spectral line (curve d in Fig. 1B). For the first color change process, an obvious red to sandy beige color change is observed as AA is added to the AuNPs solution in the presence of Hg^{2+} . It has been reported that AA can reduce Hg^{2+} to $\text{Hg}(0)$, which can deposit onto the AuNPs surface to form the Hg-Au alloys because of the high affinity between Hg and Au.^[15, 16, 19] The sandy beige color (image c in Fig. 1A) and UV-vis absorption spectra (curve c in Fig. 1B) of this system are all similar to that of the Au-Hg alloys.^[17, 23] Therefore, we deduce that this phenomenon is attributed to the formation of Au-Hg alloys. For the second color change process, the sandy beige to celandine green color change induced by SH-HPEI was seen. The SH-HPEI has not only a great number of amine groups, but also many thiol groups. Importantly, mercury has a high thiophilicity,^[24] thus mercury can combine with SH-HPEI through Hg-S bond.^[25] Moreover, there is a strong interaction between thiol (amine) and Au via forming strong Au-S covalent bonds^[26] and weak Au-NH₂ non-covalent bonds^[27], respectively. Each SH-HPEI molecule having multiple binding groups can interact with several nanoparticles around it. Therefore, these interactions can reduce the distance between nanoparticles, which promotes the aggregation of Au-Hg alloys and formation of larger Au-Hg alloys with a color change.

Fig. 1. (A) Photographs and (B) UV-vis absorption spectra of (a) 0.6 nM citrate-capped AuNPs; (b) 0.6 nM AuNPs with 4.38 μM Hg^{2+} ; (c) 0.6 nM AuNPs with 6.3 mM AA in the presence of 4.38 μM Hg^{2+} with a red to sandy beige color change; (d) SH-HPEI (66.7 mg/L) was added into the mixture of 0.6 nM AuNPs, 6.3 mM AA and 4.38 μM Hg^{2+} with a sandy beige to celandine green color change.

To test the proposed sensing mechanism above, we used TEM to examine the particles during the sensing process. As shown in Fig. 2A, the pure AuNPs with an average diameter of about 12.5 nm display a regular spherical or spherical-like structure. Compared to AuNPs, a slight increase in size and distribution (19.5 ± 4.0 nm) for Au-Hg alloys was observed (Fig. 2B). The TEM images showed that the AuNPs and Au-Hg alloys can be well-separated. After the addition of SH-HPEI, there are an aggregation and a significant increase in size and distribution (41.5 ± 17.0 nm) for the

particles (Fig. 2C). Moreover, the change in particle sizes was also determined by DLS analysis. From Fig. 2D-F, it can be found that the typical dynamic sizes of AuNPs, Au-Hg alloys and the larger Au-Hg alloys are 15.0-18.5, 20.7-38.1 and 78.8-105.7 nm, respectively. Although these results are slightly larger than that obtained from TEM (as known, DLS measured the hydrodynamic diameter of nanoparticles dispersed in water with a looser nanostructure), **the regularity of size change of nanoparticles in the process of sensing is in agreement with that of TEM**. These findings strongly support the idea that SH-HPEI can trigger the aggregation of Au-Hg alloys with obvious color change, which realizes the colorimetric detection of Hg^{2+} .

Fig. 2. TEM images of (A) AuNPs, (B) Au-Hg alloys and (C) larger Au-Hg alloys stabilized by SH-HPEI. DLS diagram of (D) AuNPs, (E) Au-Hg alloys and (F) larger Au-Hg alloys stabilized by SH-HPEI.

3.3 Effect of AA concentration on detection of Hg^{2+} .

Fig. 3. Effect of AA on (A) color changes (B) absorption spectra and (C) the response ($A_{400\text{ nm}}$) of 0.6 nM AuNPs and 4.38 μM Hg^{2+} in the presence of HPEI-SH (66.7 mg/L).

As mentioned above, AA played an important role for the reduction of Hg^{2+} in this detection system. In the absence of AA, the color (image b in Fig. 1A) and the absorption spectra (curve b in Fig. 1B) of AuNPs solution have no significant change upon the addition of Hg^{2+} . It can be seen from Fig. 3A and 3B that the larger Au-Hg alloys show an obvious color change and a broad resonance plasmon band at 350 to 450 nm as the concentration of AA changes. We choose the absorption value at 400 nm ($A_{400\text{ nm}}$) to express the sensor responses. According to Fig. 3C, the $A_{400\text{ nm}}$ value was maximized at the AA concentration of 4.42 mM (pH 4.6). Below the concentration of 4.42 mM, the $A_{400\text{ nm}}$ values went up as the AA concentration increased. This was because the deposition of Hg^0 on AuNPs surface increased, which could increase the $A_{400\text{ nm}}$ values. However, the concentration of AA higher than 4.42 mM was detrimental to the stability of the larger Au-Hg alloy solution, decreasing the $A_{400\text{ nm}}$ value.^[17] Therefore, the concentration of AA of 4.42 mM (i.e., pH 4.6) was selected for use in the following experiments.

3.4 Effect of AuNPs concentration on detection of Hg^{2+} .

Fig. 4. Photographic images of 4.42 mM AA containing different concentration of AuNPs in the (A) presence and (B) absence of 4.38 μM Hg^{2+} . UV-vis absorption responses of 4.42 mM AA containing different concentration of AuNPs in

the (C) presence and (D) absence of $4.38 \mu\text{M Hg}^{2+}$. ($[\text{AuNPs}]/[\text{HPEI-SH}]=1 \text{ nM}/111.2 \text{ mg/L}$).

The color change and UV-vis absorption response to the concentration of AuNPs should be chosen, which is important for the sensitivity of colorimetric sensor. Obviously, in the presence of Hg^{2+} of a given concentration, the lower concentration of AuNPs, the more Hg^0 will be deposited on the surface of each single Au particle and account for the higher sensitivity of the sensor. However, the linear range of the detection of Hg^{2+} would be narrow as the concentration of AuNPs was too low. Meanwhile, at relatively lower concentration of AuNPs, the red is not more obvious. Hence, it is inconvenient to observe the color change before and after sensing. As shown in Fig. 4, it is readily to see a marked difference in color (Fig. 4A and B) and absorption spectra (Fig. 4C and D) when the concentration of AuNPs is above 0.3 nM. Therefore, the concentration of 0.3 nM AuNPs was chosen as the optimum concentration for sensing Hg^{2+} in the following experiments, which was beneficial to obtaining the widest linear response range and the lowest detection limit.

3.5 Sensitivity of the colorimetric sensor for Hg^{2+} .

Fig. 5. (A) Photographs and (B) Absorption spectra of 0.3 nM AuNPs with different Hg^{2+} concentrations. (C) The $A_{400 \text{ nm}}$ values versus the concentrations of Hg^{2+} . ($[\text{AuNPs}]/[\text{HPEI-SH}]=1 \text{ nM}/111.2 \text{ mg/L}$)

Under the optimal conditions, the sensitivity of AuNPs for Hg^{2+} was evaluated. The change in solution color and UV-vis absorption responses of different concentrations of Hg^{2+} were shown in Fig. 5. As the concentrations of Hg^{2+} ranged from 8.76×10^{-9} to $1.27 \times 10^{-4} \text{ M}$, the value of $A_{400 \text{ nm}}$ of the system increased gradually (Fig. 5B) and the color of the AuNPs solution changed from red through sandy beige to celadine green. According to the gradual visible color change (Fig. 5A), the existing of Hg^{2+} in AuNPs solution can be detected qualitatively by the naked eye. A linear relationship was obtained over the concentration range from 8.76×10^{-9} to $1.27 \times 10^{-4} \text{ M}$, with a correlation coefficient of 0.989 (Fig. 5C). The limit of detection (LOD) for Hg^{2+} was estimated to be $8.76 \times 10^{-9} \text{ M}$, which was lower than the guideline value of $0.006 \text{ mg/L} \approx 30 \text{ nM}$ in drinking water defined by the World Health Organization (WHO).^[28] Moreover, we compared the LOD with other AuNPs based sensors reported by the literature (Table S1), our new sensing method shows lower LOD than those above. Therefore, this new colorimetric sensor based on the successive three color changes during the detecting process has favorable potential for ultra-sensitive detection of mercury ions.

3.6 Selectivity of the colorimetric sensor for Hg^{2+} .

Fig. 6. Selectivity of the sensor: (A) Photographs and (B) Absorption spectra of 0.3 nM AuNPs and 4.42 mM AA solutions containing Hg^{2+} (4.38 μM) and several other metal ions (44 μM) except Ag^+ (4.4 μM). ([AuNPs]/[HPEI-SH]=1 nM/111.2 mg/L)

To assess the selectivity of SH-HPEI/AuNPs to Hg^{2+} , many other different metal ions including Na^+ , Sr^{2+} , Cs^+ , Co^{2+} , Mg^{2+} , Cd^{2+} , Ni^{2+} , Mn^{2+} , Cu^{2+} , Ba^{2+} and Ag^+ were tested under identical conditions. Fig. 6 shows the color (Fig. 6A) and UV-vis absorption spectrum changes (Fig. 6B) on all metal ions. It can be seen that only Hg^{2+} and Ag^+ caused obvious changes in the color (Fig. 6A) and absorption spectra (Fig. 6B), which illustrated that the addition of 10-fold higher concentrations of other metal ions to this system did not interfere with the detection of Hg^{2+} . It should be noted that Hg^{2+} and Ag^+ are quite easily interfere with each other in AuNPs based colorimetric detection system. To improve the selectivity of probes, masking agents must be chosen, which complicate the detection process.^[15, 16] However, there are completely different changes in color and UV-vis absorption spectrum upon the addition of Hg^{2+} and Ag^+ , which can avoid the use of masking agents. After the successive addition of 4.38 μM Hg^{2+} and SH-HPEI, the color of the AuNPs solution changed from red through sandy beige to celandine green due to the formation of large Au-Hg alloys, which is different from the color change from red through brown to green for Ag^+ (Fig. S3A). Compared with that of Hg^{2+} sensing system, the UV-vis absorption spectra of Ag^+ sensing system showed a broad resonance plasmon band at 550 to 700 nm (Fig. S3B). Obviously, two UV-vis absorption spectra are quite different from each other. Therefore, this new sensing strategy based on three successive color changes during the detecting process showed great selectivity for Hg^{2+} owing to the lower possibility of false positive results.

3.7 Hg^{2+} detection in tap water.

Fig. 7. (A) Photographs and (B) Absorption spectra of 0.3 nM AuNPs with different Hg^{2+} concentrations in tap water. (C) The $A_{400\text{ nm}}$ values versus the concentrations of Hg^{2+} . ([AuNPs]/[HPEI-SH]=1 nM/111.2 mg/L). Inset: the linear relationship between the $A_{400\text{ nm}}$ and Hg^{2+} concentration range of 4.38×10^{-7} to 1.27×10^{-4} M.

To evaluate the practicality of this new colorimetric method, the SH-HPEI/AuNPs system was applied in the detection of Hg^{2+} in tap water. When the concentration of Hg^{2+} is above 4.38×10^{-7} M, the color (Fig. 7A) and UV-vis absorption spectra (Fig. 7B) all exhibit obvious changes. As shown in Fig. 7C, the $A_{400\text{ nm}}$ of the system increased linearly within the Hg^{2+} concentration of 4.38×10^{-7} to 1.27×10^{-4} M (Fig. 7C) in tap water, with a correlation coefficient of 0.9922 (Inset Fig. of Fig. 7C). And the LOD of 4.38×10^{-7} M for Hg^{2+} was obtained by the naked eye and UV-vis spectra. The responding sensitivity to Hg^{2+} in tap water was lower than that in pure water because of the formation of organic complex constructed by Hg^{2+} and organic matter existed in tap

water.^[16] Although the sensitivity in tap water was different from that in pure water, this colorimetric sensor was potentially applicable for Hg^{2+} detection in real water sample.

4. Conclusions

We developed a novel colorimetric sensor for the detection of Hg^{2+} with highly sensitivity and selectivity by using AA and multi-sulphydryl functionalized hyperbranched polyethylenimine (i.e., SH-HPEI) as reducing agent and triggering aggregation reagent, respectively. The mechanism of sensing includes two stages. The first stage is Au-Hg alloying and the second stage is Au-Hg alloy aggregation, along with the corresponding three color changes from red (AuNPs) through sandy beige (Au-Hg alloy) to celandine green (larger Au-Hg alloy). The selectivity of this colorimetric sensor for Hg^{2+} detection is extremely high because the appearing probability is extremely low to have the identical changes in 3 color triggered by interfering agents or the possibility of false positive results. Meanwhile, this new colorimetric sensor shows high sensitivity with a LOD of 8.76×10^{-9} M. There is a good linear relationship ($R^2=0.989$) between the $A_{400 \text{ nm}}$ values and the Hg^{2+} concentration ranging from 8.76×10^{-9} to 1.27×10^{-4} M. Moreover, this simple and rapid method is useful for detecting Hg^{2+} in real water sample. In conclusion, based on the two stage of color change in AuNPs solution, the sensing system has great potential for high sensitive and selective detection of Hg^{2+} in various fields.

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Graphical Abstract

The novel red through sandy beige to celandine green colorimetric sensor for Hg^{2+} detection based on multi-sulfhydryl functionalized hyperbranched polyethylenimine stabilized Au-Hg alloys showed high sensitivity and selectivity.

Biographies

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