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Carbon nanoparticle for highly sensitive and selective fluorescent detection of mercury(II) ion in aqueous solution

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ABSTRACT

In this article, carbon nanoparticles (CNPs) were used as a novel fluorescent sensing platform for highly sensitive and selective Hg^{2+} detection. To the best of our knowledge, this is the first example of CNPs obtained from candle soot used in this type of sensor. The general concept used in this approach is based on that adsorption of the fluorescently labeled single-stranded DNA (ssDNA) probe by CNP via π–π stacking interactions between DNA bases and CNP leads to substantial dye fluorescence quenching; however, in the presence of Hg^{2+}, T–Hg^{2+}–T induced hairpin structure does not adsorb on CNP and thus retains the dye fluorescence. A detection limit as low as 10 nM was achieved. The present CNP-based biosensor for Hg^{2+} detection exhibits remarkable specificity against other possible metal ions. Furthermore, superior selectivity performance was observed when Hg^{2+} detection was carried out in the presence of a large amount of other interference ions. Finally, in order to evaluate its potential practical application, Hg^{2+} detection was conducted with the use of lake water other than pure buffer and it is believed that it holds great promise for real sample analysis upon further development.

1. Introduction

Mercury(II) ion (Hg^{2+}) is a highly toxic heavy metal ion and considered as one of the most dangerous and ubiquitous pollutants (Renzoni et al., 1998). Its contamination comes from a variety of natural sources as well as human activities (Harris et al., 2003), and an annual release was 4400–7500 metric tons estimated by the United Nations Environment Programme (UNEP) (Liu and Lu, 2007). It have been demonstrated that Hg^{2+} can easily pass through skin, respiratory, and gastrointestinal tissues into the human body and damage the central nervous and endocrine systems (Gutknecht, 1981), which raises serious environmental and health concerns. So there is an ever-growing demand to develop effective analytical methods for sensitive and selective detection of Hg^{2+}. Indeed, the past years have witnessed increasing research efforts in this direction and many detection methods have been established (Chen and He, 2004; Chiang et al., 2008; Li et al., 2009, 2010; Guo et al., 2009; Huang et al., 2007; Kim and Bunz, 2006; Liu and Lu, 2007; Miller and Chang, 2007; Wang et al., 2005, 2007, 2008; Wegner et al., 2007; Yoon et al., 2005; Zhang et al., 2008). Traditionally, Hg^{2+} is detected by atomic absorption/emission spectroscopy, Auger-electron spectroscopy, inductively coupled plasma mass spectrometry, or ion selective electrode or polarography (Guo et al., 1996; Wang et al., 2007); however, these methods require sophisticated instrumentation and/or sample preparation, which limits their practical applications. To solve these problems, alternative methods using small-molecule-based fluorescent probe (Chiang et al., 2008; Descalzo et al., 2003; Miller and Chang, 2007; Nolan and Lippard, 2003; Wang et al., 2005; Yoon et al., 2005; Zhang et al., 2008), DNAzymes (Liu and Lu, 2007; Vannela and Adriaens, 2007; Darbha et al., 2008; Li et al., 2009), protein (Chen and He, 2004; Wegner et al., 2007), conjugated polymers (Kim and Bunz, 2006), gold nanoparticles (Huang et al., 2007; Liu et al., 2008; Wang et al., 2008), and semiconductor quantum dots (Guo et al., 2009) have also been developed in the past years. However, each of these methods has its own drawbacks, such as poor selectivity, insufficient resolution in aqueous media, high detection limit, and/or sophisticated synthesis of the probe materials. Although highly selective and sensitive detection of Hg^{2+} has been reported recently (Chiang et al., 2008; Huang et al., 2007; Liu et al., 2008), it still remains a great challenge to develop other new methods for this kind of detection, which will provide more insights for the development.

On the other hand, oligonucleotide (OND) has also been proven to be a versatile tool for the detection of metal ions due to their specific interactions. The conformation of OND can be induced to fold into a hairpin structure by metal ion such as Hg^{2+} through T–Hg^{2+}–T base pairs (Miyake et al., 2006), providing a rationale to design T-rich OND-based sensor for Hg^{2+}. Indeed, Ono et al.

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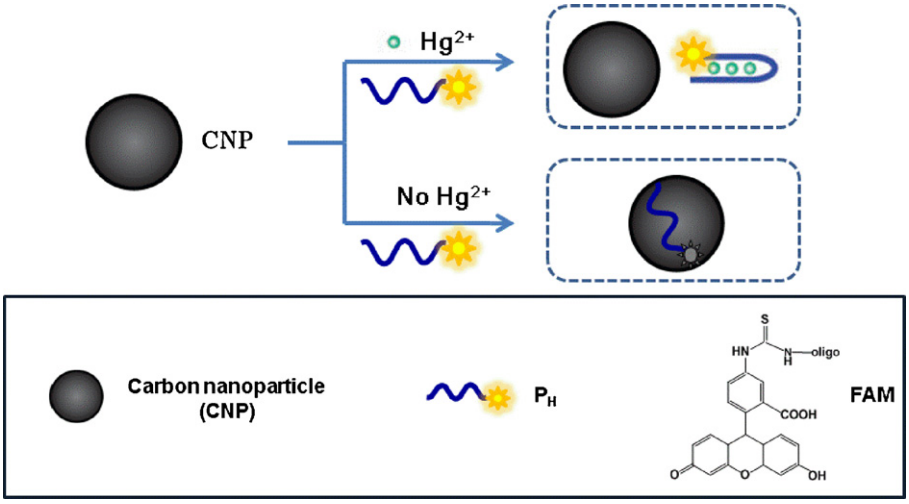

Scheme 1. A schematic (not to scale) illustrating the CNP-based fluorescent Hg^{2+} detection based on conformational change of a Hg^{2+}-specific T-rich OND (P_H). P_H: a FAM-labeled Hg^{2+}-specific OND probe.

have developed a fluorescent Hg^{2+} sensor using a T-rich OND, dual-labeled with a fluorescent dye and a quencher at each terminus (Ono and Togashi, 2004). In the presence of Hg^{2+}, the chelating of Hg^{2+} into mismatched T–T base pair induces the formation of a hairpin structure, which brings both termini close to each other, resulting in significant quenching of the dye fluorescence. More recently, nanostructures including single-walled carbon nanotubes (SWCNTs) (Zhang et al., 2010), graphene oxide (GO) (He et al., 2010) and nano-C_{60} (Li et al., 2011a) have been used to quench the dye of a single-labeled fluorescent OND probe and as an effective fluorescent sensing platform for sensitive and selective Hg^{2+} detection. However, SWCNT, graphite (used for GO preparation) and C_{60} (used for nano-C_{60} preparation) must be purchased from some sources. On the other hand, SWCNT treatment and GO preparation by Hummers method (Hummers and Offeman, 1958) is time-consuming and labor-intensive. Accordingly, new sensing platform which can overcome all these shortcomings is highly desirable. In this article, we demonstrate the first use of carbon nanoparticles (CNPs), relatively inexpensive carbon materials obtained from candle soot, as a fluorescent sensing platform for highly sensitive and selective detection of Hg^{2+}, expecting to extend the use of this emerging kind of nanocarbon (Sheila and Gary, 2010). The general concept used in this approach is based on that adsorption of the fluorescently labeled single-stranded DNA (ssDNA) probe by CNP via π–π stacking interactions between DNA bases and CNP (Li et al., 2011b; Varghese et al., 2009) leads to substantial fluorescence quenching; however, in the presence of Hg^{2+}, T–Hg^{2+}–T induced hairpin structure does not adsorb on CNP and thus retains the dye fluorescence. Scheme 1 presents a schematic to illustrate the CNP-based fluorescent Hg^{2+} detection.

2. Experimental

2.1. Chemicals and instruments

The chemically synthesized oligonucleotide was purchased from Shanghai Sangon Biotechnology Co. Ltd. (Shanghai, China). DNA concentration was estimated by measuring the absorbance at 260 nm. All the other chemicals were purchased from *Aladin* Ltd. (Shanghai, China) and used as received without further purification. Metal ion solutions were prepared from corresponding chloride salt. The water used throughout all experiments was purified through a Millipore system.

Scanning electron microscopy (SEM) measurements were made on a XL30 ESEM FEG scanning electron microscope. Transmission electron microscopy (TEM) measurements were made on a HITACHI H-8100 EM (Hitachi, Tokyo, Japan). Fluorescent emission spectra were recorded on a RF-5301PC spectrofluorometer (Shimadzu, Japan). Zeta potential measurements were performed on a Nano-ZS Zetasizer ZEN3600 (Malvern Instruments Ltd., U.K.).

Oligonucleotide sequence is listed as follows:

P_H (FAM dye-labeled ssDNA probe for Hg^{2+}):
5′-TTC TTT CTT CCC CTT GTT TGT T-FAM-3′

2.2. Carbon nanoparticles preparation

CNPs were prepared as follows: Too small CNPs (below 10 nm) produce strong photoluminescence emission interfering with detection (Sheila and Gary, 2010) and too large CNPs tend to sink during the measurements, so CNPs with suitable size should be selected. In brief, 3-mg candle soot obtained using well-established method (Liu et al., 2007) was suspended in 12-mL water–ethanol mixture (1:1) with the help of ultrasonication for 30 min. After that, the black solution was centrifuged at 3000 rpm for 2 min to separate out large carbon soot particle and the supernatant was collected. Subsequently, the collected supernatant was subjected to centrifugation at 6000 rpm for 2 min. Too small CNPs still suspended in the liquid and were discarded, the black precipitate was collected and redispersed in 12-mL water–ethanol mixture (1:1) for further characterization and sensing application. We repeat the separation and collected the desired CNPs, both of water and ethanol can be removed by lyophilization. According to the mass of the precipitation after drying, the concentration of previously prepared suspension is about 0.15 mg/mL.

For scanning electron microscopy (SEM) characterization, 20 μL of the suspension was placed on an indium tin oxide (ITO) glass slide and air-dried at room temperature. SEM measurements were made on a XL30 ESEM FEG scanning electron microscope at an accelerating voltage of 20 kV. The sample for transmission electron microscopy (TEM) measurements was prepared by placing a dilution of colloidal solution on a carbon-coated copper grid and drying at room temperature. TEM measurements were made on a HITACHI H-8100 EM (Hitachi, Tokyo, Japan) with an accelerating voltage of 200 kV.