Fabrication of Water-soluble Fluorescent Polymeric Micelles for Selective Detection of Hg²⁺ in Blood Serum

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In this study, amphiphilic diblock copolymers were designed and synthesized *via* the incorporation of reversible addition-fragmentation chain transfer radical polymerization (RAFT) and a subsequent grafting technique. Subsequently, Hg²⁺-sensitive water-soluble fluorescent polymeric micelles (FNs) were prepared by a reprecipitation strategy. The spectroscopic characteristics demonstrate that the fluorescein isothiocyanate (FITC) was successfully linked into the polymer. Due to the promoted reaction of desulfurization cyclization by Hg²⁺, the fluorescence of fluorescein in FNs was obviously quenched. The as-prepared FNs showed admirable Hg²⁺-sensitivity (detection limit: 54 nM), excellent water-solubility and high selectivity. In addition, FNs were successfully used to determine Hg²⁺ in blood serum. We expected that the as-prepared FNs could perform potential applications in imaging, sensing, and bioanalytic chemistry.

Keywords Fluorescence polymeric micelles, Hg²⁺, blood serum, RAFT

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Introduction

Obtaining simple and convenient nanosensors, which were cost-effective and applicable to imaging and sensing, became a sought-after goal.^{1–5} Mercury ion (Hg²⁺) posed a tremendous menace owing to its bioaccumulation, such as permanent damage and slow accumulation in the central nervous and endocrine systems.⁶ Moreover, it can easily give rise to a wide variety of diseases, including prenatal brain damage, serious cognitive problems, motion disorders and Minamata disease.^{7,8} In addition, once introduced into the marine environment, some bacteria transform inorganic Hg²⁺ into methylmercury.^{9,10} As we know, this is implicated as a cause of mercury pollution related to serious irreversible neurological damage due to its highly neurotoxic property.^{9–11} The Hg²⁺ also caused the deterioration of water quality due to its notorious water and environmental pollution property.^{12,13} Hence, highly sensitive and selective detection of Hg²⁺ was of significance for organisms and environment.

A number of methods have been used to detect Hg²⁺, such as atomic absorption/emission spectroscopy,¹⁴ gas chromatography,¹⁵ cold vapor atomic fluorescence spectrometry,¹⁶ voltammetry,¹⁷ *etc.* However, these conventional methods are not commendably applied in living biosystems due to several common faults, including being time-consuming, meaning complicated sample preparation and a sophisticated process.^{18,19} In contrast, fluorescence spectroscopy reveals a promising way owing to its

ease of operation, being non-invasive, and having highly sensitive and selective properties.^{20–23}

To date, many fluorescent sensors for Hg²⁺ have been reported.^{24–32} However, many of these sensors are small molecules, which can only work in organic or mixed solvents, may restrict their applications in the environmental or bio-related fields due to their low water solubility and high toxicity.^{33,34} To overcome these limitations, polymeric micelles-based sensors have recently attracted considerable interest owing to their excellent properties, including improved water solubility and longer *in vivo* circulation times, effective distribution of fluorescent dyes, *etc.*^{35,36} Also, several polymeric particles-based fluorescence nanosensors have been developed for Hg²⁺ detection.^{36–38} So far, an excellent fluorescent sensor for Hg²⁺ in a totally aqueous solution is still rare. Therefore, designing water-soluble (dispersible) fluorescent nanosensors for detecting Hg²⁺ is of great significance.

Herein, we firstly prepared amphiphilic diblock copolymers (PEO-*b*-P(St-co-VBA-co-VBF) *via* reversible addition-fragmentation chain transfer radical polymerization (RAFT) and a subsequent grafting technique (Scheme 1). Then, the amphiphilic diblock copolymers could be easily turned to form polymeric micelles in aqueous solution by a reprecipitation strategy, as illustrated in Scheme 2. With the addition of Hg²⁺, the fluorescence of the fluorescein units could be effectively quenched through desulfurization cyclization of the thiourea moiety, resulting in the selectively fluorescent sensing of Hg²⁺ in a totally aqueous media and selectively detecting Hg²⁺ in blood serum (Scheme 2).

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Scheme 1 Synthetic route of PEO₁₁₃-b-P(St₁₉-co-VBA₁-co-VBF₃). Scheme 2 Schematic mechanism of fluorescent polymeric micelles for Hg²⁺.

Experimental

Reagents and chemicals

Fluorescein isothiocyanate (FITC, 96%, Aldrich), 1-dodecanethiol (Aldrich) and poly(ethylene glycol) methyl ether (PEO-OH, $M_w = 5000$, Alfa) were used as received. Tetrahydrofuran (THF, A.R.), *N,N*-dimethylformamide (DMF,

A.R.), *N,N*-diisopropylethylamine (DIEA, A.R.), and ethylenediamine (A.R.) were distilled over CaH₂. Styrene (St, 99%, Aldrich) was purified by vacuum distillation. 4-Vinylbenzyl chloride (VBC, 99%, Aldrich) was purified by silica-gel column chromatography to remove any polymerization inhibitor. Azobisisobutyronitrile (AIBN, 99.99%, Aldrich) was recrystallized from ethanol and dried under vacuum. The water used throughout this work was the double-distilled water, which was further purified with a Milli-Q system. Petroleum ether and diethyl ether, other reagents were analytical reagents and used without further purification. Glutathione (GSH), cysteine (Cys), nitrate salts of metal anions (Ca²⁺, Co²⁺, Cu²⁺, Fe³⁺, Ag⁺, Hg²⁺, K⁺, Mg²⁺, Mn²⁺, Na⁺, Ni²⁺, Zn²⁺) and sodium salts of (Cl⁻, I⁻, SO₄²⁻, NO₃⁻, NO₂⁻, HCO₃⁻, and H₂PO₄²⁻) were obtained from Aldrich. The PEO macro-RAFT agent was available from our previous work (PEO₁₁₃-TTC, end-functionality >95%, Fig. S1).³⁹

Apparatus

¹H NMR spectra were measured on a Bruker Avance 500 MHz NMR spectrometer. The nanoparticle diameters were determined by a Malvern Nano-ZS90 instrument, and their morphology was observed with an atomic force microscope (AFM, Seiko SII 400) in the tapping mode and a JEM-2100F transmission electron microscope (TEM, JEOL USA, Inc.). FTIR spectra were obtained by using a Nicolet Avatar 360 FTIR spectrophotometer. The number-average molecular weights (M_n) and PDI were determined by a Waters 2410 gel permeation chromatograph (GPC) at 30°C and using THF as the eluent (1.0 mL/min). The calibration curve was obtained by using polystyrene (PS) as the standard. UV-Vis absorption spectra were recorded on a Shimadzu UV-2501PC spectrophotometer at room temperature (298 K). Fluorescence spectra were recorded on a Shimadzu RF-5301PC fluorescence spectrophotometer at room temperature.

Synthetic route of PEO₁₁₃-b-(St₁₉-co-VBC₄). PEO₁₁₃-TTC (375 mg, 0.075 mmol), St (1.296 g, 12 mmol), VBC (0.428 g, 3 mmol) and AIBN (3.36 mg, 0.02 mol) were dissolved in 3 mL of deputed DMF. After three times of the freeze-pump-thaw cycle, the mixture was stirred under 90°C for 4 h. The copolymer was precipitated in a diethyl ether (20 mL) and

Fig. 1 (A) GPC trace of PEO₁₁₃-TTC and the diblock copolymer PEO₁₁₃-b-(St₁₉-co-VBC₄). (B) Absorption spectra of fluorescence polymeric micelles formed by the coprecipitation of PEO₁₁₃-b-P(St₁₉-co-VBA₄) (without FITC) and (PEO₁₁₃-b-P(St₁₉-co-VBA₁-co-VBF₃)) (with FITC).

diethyl ether (10 mL) mixture solution to remove any unreacted small molecule compounds. After being filtered and dried at 30°C under a vacuum for 48 h, the polymer PEO₁₁₃-b-(St₁₉-co-VBC₄) was obtained (pale-yellow powder; 610 mg; yield, 60%; M_n , 13.44 kDa; PDI, 1.10; Fig. S2, Fig. 1A and Scheme 1).

¹H NMR (500 MHz, CDCl₃) δ 7.24 – 6.22 (m, 108 H), 4.51 (s, 8 H), 3.76 – 3.44 (m, n H), 3.38 (s, 3 H), 2.24 – 1.67 (m, 37 H), 1.63 – 1.21 (m, 56 H), 0.86 (ddd, J = 10.1, 6.4, 3.2 Hz, 3 H).

Synthetic route of PEO₁₁₃-b-P(St₁₉-co-VBA₄). PEO₁₁₃-b-(St₁₉-co-VBC₄) (200 mg, 0.028 mmol), *N,N*-diisopropylethylamine (512 mg, 3.96 mmol) and ethylenediamine (532 mg, 8.8 mmol) were dissolved in 10 mL of dehydrated DMF. The mixture was stirred under 70°C for 8 h. The solvent was removed using a rotary evaporator. Residues were precipitated in diethyl ether (20 mL). After being filtered and dried at 40°C under a vacuum for 48 h, PEO₁₁₃-b-P(St₁₉-co-VBA₄) was obtained (yellow powder, 156 mg, yield: 75%, Fig. S3 and Scheme 1).

¹H NMR (500 MHz, CDCl₃) δ 8.22 (d, J = 8.2 Hz, 8 H), 7.27 (s, 10 H), 7.23 – 5.96 (m, 106 H), 3.64 – 3.40 (m, n H), 3.38 (s, 3 H), 2.88 (d, J = 4.5 Hz, 17 H), 2.22 – 0.77 (m, 99 H).

Synthetic of PEO₁₁₃-b-P(St₁₉-co-VBA₁-co-VBF₃). PEO₁₁₃-b-P(St₁₉-co-VBA₄) (100 mg, 0.008 mmol) and fluorescein isothiocyanate (10 mg, 0.026 mmol) were dissolved in 5 mL of dehydrated DMF. The mixture was stirred under the ambient temperature for 48 h. The solvent was removed using a rotary evaporator. Residues were precipitated in water (10 mL, pH 8.0) to remove any unreacted FITC. After being filtered and dried at 40°C under a vacuum for 48 h, PEO₁₁₃-b-P(St₁₉-co-VBA₁-co-VBF₃) was obtained (red powder, 84 mg, yield: 76%, Fig. S4 and Scheme 1).

¹H NMR (500 MHz, CDCl₃) δ 8.02 (s, 2 H), 7.65 – 7.24 (m, 28 H), 7.20 – 6.16 (m, 122 H), 3.69 – 3.43 (m, n H), 3.38 (s, 3 H), 2.92 (d, J = 36.5 Hz, 20 H), 2.51 – 0.36 (m, 138 H).

Preparation of Hg²⁺-sensitive fluorescent polymeric micelles (FNs)

Typical procedures employed for the preparation of polymeric micelles solution were as follows: 0.2 mg of PEO₁₁₃-b-P(St₁₉-co-VBA₁-co-VBF₃) amphiphilic diblock copolymer was dissolved in 1 mL of THF. The 1 mL of solution was quickly added into 10 mL of deionized water under vigorous sonication by ultrasound. The ultrasonic process was continued for 10 min to obtain a polymeric micelles solution. THF was allowed to completely evaporate using a rotary evaporator at room temperature. The solution was filtered through a 0.22- μ m

cellulose membrane filter and adjusted to the appropriate concentration.

Fluorescence assay of Hg²⁺ in water

Fluorescence measurements were performed in water, according to the following procedure. In a quartz cell, 2.7 mL of deionized water and 0.3 mL of FNs were mixed. Subsequently, this solution was added to various concentrations of Hg²⁺ (0, 2, 5, 10, 15, 20, 40, 60, 80, 100, 130, 150 μ M), respectively. Also, the mixture was equilibrated for 10 min before any measurement. The fluorescence intensity was measured synchronously ($\lambda_{ex/em}$ = 90/517 nm).

Fluorescence assay of Hg²⁺ in tap water and blood serum

Firstly, tap water was ten-fold diluted. A blood-serum solution was 200-fold diluted before use. In a quartz cell, 2.7 mL of tap water (or blood serum) and 0.3 mL of FNs were mixed. Subsequently, this solution was added to various concentrations of Hg²⁺ (5, 10, 15 μ M), respectively. Also, the mixture was equilibrated for 10 min before any measurement. The fluorescence intensity was measured synchronously ($\lambda_{ex/em}$ = 490/517 nm).

Results and Discussion

Characterizations of FNs

For the preparation of FNs, RAFT polymerization was firstly used to synthesize the amphiphilic diblock copolymer PEO-b-(St-co-VBC) with hydrophobic units of St and functional units of VBC from the beginning hydrophilic PEO₁₁₃-TTC macro-RAFT reagent. GPC analysis showed that the amphiphilic diblock copolymer possessed a relatively narrow polydispersity (M_n = 13.44 kDa, M_w/M_n = 1.10, Fig. 1A). The degrees of polymerization (DPs) of the St and VBC units were found to be 19 and 4, which were determined by a ¹H NMR calculation. In detail, they were achieved *via* comparing the integral area of the end methyl group (peak b) of PEO with the identifiable repeat units of St (peak e) or VBC (peak d, Fig. S2). Thus, the diblock copolymer was denoted as PEO₁₁₃-b-(St₁₉-co-VBC₄). The structural parameters of the PEO₁₁₃-TTC precursor and the PEO₁₁₃-b-(St₁₉-co-VBC₄) diblock the copolymer are summarized in Fig. 1A and Table 1. Subsequently, the chlorine group in the VBC units in the copolymers was fully transformed into an amine group, as displayed in Scheme 1 and Fig. S3. For the ethylenediamine-functionalized the polymer of PEO₁₁₃-b-

(St₁₉-co-VBC₄), the conspicuous band at 1672 cm⁻¹ of the amine group (-NH₂) can be seen in Fig. 2A, indicating that the PEO₁₁₃-b-(St₁₉-co-VBC₄) was functionalized with ethylenediamine. Then, FITC was covalently linked to the copolymer, and the grafted content of the FITC moiety in the chain of PEO₁₁₃-b-P(St₁₉-co-VBA₄) could be estimated by ¹H NMR calculation (Fig. S4), and further confirmed by spectrophotometric titration (Fig. 1B). The final copolymer was denoted as PEO₁₁₃-b-P(St₁₉-co-VBA₁-co-VBF₃). However, compared with the amine group (1672 cm⁻¹) of the PEO₁₁₃-b-P(St₁₉-co-VBA₄), the band (-NH₂) at 1672 cm⁻¹ of PEO₁₁₃-b-P(St₁₉-co-VBA₁-co-VBF₃) obviously varied (Fig. 2A). This result further demonstrated that the PEO₁₁₃-b-(St₁₉-co-VBC₄) was functionalized with FITC. Then, as depicted in Fig. S5, the glass transition temperature (*T_g*) of all polymers was determined, respectively. However, no obvious decomposition temperature (*T_d*) could be found from 123.15 to 473.15 K. The results indicated that the polymer was stable, and could be applied to be sensed and imaged in biosystems. Finally, well-defined FNs were prepared by a reprecipitation

strategy, as shown in Scheme 2. The critical micelle concentrations (CMC) were estimated *via* absorbance based on the different behaviors of Nile Red in the hydrophobic and aqueous phases. As shown in Fig. S6, the CMC values of PEO₁₁₃-b-P(St₁₉-co-VBA₁-co-VBF₃) were 11.7 μg/mL in aqueous solution. Moreover, Fig. 2C displays an atomic force microscopy (AFM) image for a typical sample (FNs), which showed that most of the polymeric micelles were discrete, smooth, and spherical shape with an average diameter of 60 nm. This was consistent with that determined by dynamic light scattering (DLS) (Fig. 2B). In addition, it can be seen from the TEM image that the polymeric micelles were almost spherical in shape with an averaged diameter of 56 nm (Fig. 2D).

Sensitive and selective recognition of Hg²⁺ in water

Firstly, the spectroscopic properties of FNs were investigated. As shown in Fig. 3A, the FNs have a prominent emission peak at 517 nm. As the Hg²⁺ concentration increased, the fluorescence intensity (517 nm) gradually decreased, accompanying the fluorescence of FNs changed from bright green to weak green (Scheme 2). Furthermore, the change of the fluorescence intensity (*I-I₀*) *versus* Hg²⁺ concentration showed an excellent linear fitting (the correlation coefficient, *R*² = 0.9770), as displayed in Fig. 3B. Also, the Hg²⁺ detection limit could be determined to be ~54 nM, based the signal-to-noise ratio (*S/N* = 3), which is sensitive enough to sense the Hg²⁺ levels in the natural environment (Fig. S7A).^{1,40,41} In addition, there is also a very good linear relationship that at low concentrations the quenching of fluorescence follows to the classic Stern-Volmer relationship, as shown in Fig. S7B.

The selectivity is a crucial parameter for estimating the performance of a sensing system. The selectivity assay for FNs towards Hg²⁺ was evaluated by putative interference, such as Ca²⁺, Co²⁺, Cu²⁺, Fe³⁺, Ag⁺, Hg²⁺, K⁺, Mg²⁺, Mn²⁺, Na⁺, Ni²⁺ and

Table 1 Molecular weight distribution data of starting linear polymers

Polymer	<i>M_{n, GPC}</i> ^a	<i>M_{w, GPC}</i> ^a	PDI	<i>M_{n, NMR}</i> ^b
PEO-TTC	8334	8728	1.05	5336
PEO-b-P(St-co-VBC)	13443	14757	1.10	7922
PEO-b-P(St-co-VBA)	12493	13560	1.12	8015
PEO-b-P(St-co-VBA-co-VBF)	12793	13810	1.10	9153

a. The data were acquired using SEC based on a polystyrene calibration curve and obtained from GPC analysis was using THF as eluent at a flow rate of 1.0 mL/min.

b. *M_{n, NMR}* values were determined by ¹H NMR.

Fig. 2 (A) FTIR spectra of the PEO-b-P(St-co-VBC)₂₃ (a), PEO₁₁₃-b-P(St₁₉-co-VBA₄) (b), (PEO₁₁₃-b-P(St₁₉-co-VBA₁-co-VBF₃)) (c). (B) Particle sized distribution obtained by using DLS (B) for the FNs sample. (C) AFM image for the PN sample. (D) TEM image of the PN sample.

Fig. 3 (A) Fluorescence emission spectra (excited at 490 nm) of FNs upon different concentrations of Hg^{2+} (concentration: 0, 2, 5, 10, 15, 20, 40, 60, 80, 100, 130, 150 μM). (B) Relationship between the fluorescence intensity (I_{517}) for typical FNs and the concentration of Hg^{2+} (concentration: 0, 2, 5, 10, 15, 20 μM).

Fig. 4 (A) Fluorescence intensity change (I/I_0) of FNs versus various metal ions (Hg^{2+} , 10 μM ; other, 100 μM); I and I_0 were the fluorescence intensities of FNs at 517 nm in the absence and presence of various cation. (B) Influence of various coexisting metal ions on the selective recognition of Hg^{2+} by FNs (other competitive ion concentration, 100 μM ; λ_{ex} , 490 nm; λ_{em} , 517 nm).

Zn^{2+} ; the results are given in Fig. 4A. It was noted that the intensity ratio (I/I_0) for FNs was significantly reduced upon the addition of Hg^{2+} . In comparison, for other metal ions, there were no obvious changes of I/I_0 in the presence of other analytes. Furthermore, a competition experiment with the interferents was also carried out (Fig. 4B). It is clear that the co-existence of metal cations has a negligible effect on Hg^{2+} sensing. These results indicated that the nanosensors system can be potentially used to detect Hg^{2+} in a complex sample matrix containing interfering metal ions.

In addition, the selectivity and anti-interference performance of the FNs system toward Hg^{2+} was further investigated, as shown in Fig. 5. Various other general bio-molecules and anions including GSH, Cys, Cl^- , I^- , SO_4^{2-} , NO_3^- , NO_2^- , HCO_3^- , and $\text{H}_2\text{PO}_4^{2-}$, were picked out together with Hg^{2+} to study the possible response on the FNs system. After joining 10 μM of Hg^{2+} and then added 100 μM of different analyte, respectively. It is clear that only Hg^{2+} induces an obvious fluorescence quenching, whereas the introduction of other bio-molecules and anions resulted in almost no fluorescence change. Moreover, the interference of the above analyte (GSH, Cys, Cl^- , I^- , SO_4^{2-} , NO_3^- , NO_2^- , HCO_3^- , and $\text{H}_2\text{PO}_4^{2-}$) coexisting with Hg^{2+} was also recorded. Figure 5B displays the fluorescence response of the

sensor toward Hg^{2+} under the addition of other competitive anions. Apparently, it was obvious that the co-existence of a competitive analyte had negligible effect on Hg^{2+} sensing. These results further indicated that the system can serve as a highly selective sensor for Hg^{2+} .

For practical applicability, the suitable pH range for the FNs system was further investigated. As illustrated in Fig. S8, in the absence and presence of Hg^{2+} , the fluorescence intensity (I_{517}) of the FNs dispersion maintained the same tendency of changes, which firstly increased, and then remained fairly stable with increasing pH. However, in the presence of Hg^{2+} , a remarkable reduction in I_{517} was observed. It could also be found that the FNs exhibited a relatively placid fluctuation from pH 7 to 10. These results indicated that the FNs could act as excellent fluorescent sensors for Hg^{2+} .

The influence of the degree of polymerization of a polymer on the size of a nanoparticle in water was further investigated. As described in Fig. S9, the change of in the degree of polymerization of a polymer displays no obvious influence on the size of the nanoparticle in water, ranging from $M_n = 8.36$ to 17.34 kDa. Furthermore, the fluorescence lifetimes of FITC and FNs were determined. As shown in Fig. S10, the fluorescence lifetimes of FITC and FNs were 3.89 to 3.81 ns,

Fig. 5 (A) Fluorescence intensity change (I/I_0) of FNs versus various analytes (Hg^{2+} , 10 μM ; other, 100 μM); I and I_0 were the fluorescence intensities of FNs at 517 nm in the absence and presence of various analytes. (B) Influence of various coexisting metal ions on the selective recognition of Hg^{2+} by FNs (other competitive ion concentrations, 100 μM ; λ_{ex} , 490 nm; λ_{em} , 517 nm).

Table 2 Determination of Hg^{2+} in tap water and blood serum

Sample ^a	Added amount of $Hg^{2+}/\mu M$	Found amount of $Hg^{2+}/\mu M$	RSD ^b , % ($n = 5$)	Recovery, %
Tap water	5.00	5.61	0.21	112.20
	10.00	11.78	0.26	117.80
	15.00	14.66	0.19	97.73
Blood serum	5.00	4.38	0.34	87.60
	10.00	10.88	0.22	108.80
	15.00	16.06	0.25	107.00

a. In the test solutions, tap water was ten-fold diluted. Blood serum solution was 200-fold diluted before used. The measurements were conducted instantly after the addition of Hg^{2+} .

b. Full name of RSD is relative standard deviation.

respectively. These results indicated that polymeric nanoparticles had no obvious aggregation-induced energy/electron transfer (AIET) or solution-induced energy/electron transfer.

It has been well revealed that thiourea can be easily transformed into guanidine derivatives, as shown in Scheme 2.^{2,33,42-51} Most importantly, Hg^{2+} can easily promote desulfurization to transform a cyclic compound, as an imidazole (1-R-2-R'-4,5-dihydro-1H-imidazole), resulting in a change of the spectroscopy properties.^{2,42,44,50,51} Also, this phenomenon can be ascribed to changes of the electronic delocalization in the chromophore.⁴⁹ Thiourea moiety in VBF, as an electron-donating group, has little influence on the spectroscopy properties from FITC because of the ethylene spacer. Also, the formation of the imidazoline moiety caused a decrease in the electron-donating ability of the receptor portion due to loss of the spacer unit.^{33,42,50} This effect can cause an overt reduction in electron delocalization on the fluorophore (fluorescein derivative). Therefore, the Hg^{2+} can induce a reaction of desulfurization cyclization.

To further test the practicability of the developed sensor, the FNs were used to detect the concentrations of Hg^{2+} ions in several tap-water and blood serum samples. As exhibited in Table 2, the RSD for all assays were less than 0.34%, and the recoveries were in the range of 87.60 to 117.8%. These results indicated that this method is suitable for the recognition of Hg^{2+} ions in real aqueous samples.

Conclusions

In summary, PEO_{113} -P(St_{19} -co-VBC₄) was successfully synthesized *via* reversible addition-fragmentation chain transfer radical polymerization (RAFT). Subsequently, the chlorine group in VBC units in copolymers was transformed into an amine group by ethylenediamine. The amphiphilic diblock copolymers (PEO_{113} -P(St_{19} -co-VBA₁-co-VBF₃)) were facily prepared by covalently linking FITC to the copolymer (PEO_{113} -P(St_{19} -co-VBA₄)). Finally, Hg^{2+} -sensitive water-soluble fluorescence polymeric micelles were developed *via* a reprecipitation strategy. The as-prepared FNs showed excellent water-solubility, highly selective detection of Hg^{2+} over other interfering analytes (Ca^{2+} , Co^{2+} , Cu^{2+} , Fe^{3+} , Ag^+ , K^+ , Mg^{2+} , Mn^{2+} , Na^+ , Ni^{2+} , Zn^{2+} , GSH, Cys, Cl^- , I^- , SO_4^{2-} , NO_3^- , NO_2^- , HCO_3^- , and $H_2PO_4^-$) in 100% aqueous media and a low detection limit (~ 54 nM). Moreover, the prepared FNs can also be further used for the selective detection Hg^{2+} in blood serum and real aqueous samples. This strategy could provide a new approach for detecting of Hg^{2+} in biological applications, imaging and sensing, and could be applied to construct other nanosensors for other metal ion detection.

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Supporting Information

The corresponding NMR analysis and determination of the detection limit was listed in the Supporting Information. This material is available free of charge on the Web at <http://www.jsac.or.jp/analsci/>.

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