Chemiluminescent magnetic nanoparticles with good catalytic activity and rapid separation capability and sensitive sensing for H₂O₂

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Abstract
It is of considerable importance to develop chemiluminescent functionalized nanomaterials (CF-NMs) with good catalytic activity, high chemiluminescence (CL) efficiency and good stability, and rapid magnetic separation capability, achieving excellent performance in CL biosensing. In this study, N-(4-aminobutyl)-N-ethylisoluminol (ABEI)-functionalized CuFe₂O₄ magnetic nanomaterial (ABEI/CuFe₂O₄) with high catalytic activity was synthesized by virtue of a solvothermal and post-functionalization method. ABEI/CuFe₂O₄ showed outstanding CL properties, superior to ABEI-CuFe₂O₄ in liquid phase. This reveals that the immobilization of ABEI on the surface of CuFe₂O₄ exhibits unique heterogeneous catalytic property. The catalytic ability of CuFe₂O₄ was better than that of CoFe₂O₄, ZnFe₂O₄, MnFe₂O₄, and NiFe₂O₄. It is suggested that the peroxide-like activity as well as Cu²⁺ and Cu⁰ enriched on the surface of ABEI/CuFe₂O₄ opened up a dual route for synergistic catalysis of H₂O₂. ABEI/CuFe₂O₄ also demonstrated good superparamagnetism and magnetic separation could be carried out in 2 min, which is advantageous for the separation and purification of ABEI/CuFe₂O₄ during the synthetic procedures and bioassays. Owing to the sensitive response of ABEI/CuFe₂O₄ to H₂O₂, an enzyme-free sensor was developed for the detection of H₂O₂ with a wide line range over 5 orders of magnitude of H₂O₂ concentrations and a low detection limit of 5.6 nM. The as-developed sensor is sensitive, stable, and convenient. This work provides a new family member of nanomaterials with good magnetism and CL activity as well as good stability. The developed ABEI/CuFe₂O₄ shows great prospects in biocatalysis, bioassays, biosensing, and bioimaging, etc.

Keywords Chemiluminescence · Magnetic nanomaterials · Catalytic activity · Sensor

Introduction
Over the past decade, chemiluminescent functionalized nanomaterials (CF-NMs) have received great interests in bioassays due to their excellent CL properties, ease-to-assemble, and good biocompatibility [1–4]. They have been used as nanoprobe and nanointerfaces to develop various methodologies for immunoassays and nucleic acid assays, which have greatly improved the analytical performance [5–9]. Nevertheless, some CF-NMs do not have high CL efficiency on their own and usually need to modify extra catalysts to improve their performance. In that case, not only the modification steps are required, but also the stability of CF-NMs cannot be guaranteed. In addition, conventional CF-NMs often require complicated purification procedures, such as filtration, dialysis, or centrifugation, which greatly prolong the analysis time and may also influence the analytical accuracy. Therefore, it is highly desired to develop CF-NMs with good catalytic activity, high CL efficiency, and good stability, achieving excellent performance in CL biosensing.

It is worth noting that magnetic nanomaterials (MNNs) deploy extensive applications in the fields of bioassay, biomedicine, catalysis, and energy storage [10–13]. Benefiting from their tunable chemical composition, intrinsic magnetism, and large specific surface area, MNNs can be considered as...
nanoparticles in CL bioassays [14–16]. To date, various magnetic materials as carriers have been used for the development of CF-NMs, such as magnetic mesoporous carbon materials [17], Fe₃O₄ magnetic beads [18], magnetic graphene materials [19], and magnetic hydrogels [20]. Among them, CF-NMs without catalysts often possess relatively low CL efficiency, while others loaded with catalysts exhibit improved CL performance. However, it is tedious to modify the catalyst, which is also easy to leak. On the other hand, some MNMs have catalytic properties due to their special chemical composition. Among them, MFe₂O₄ (M = Cu, Co, Mn, Ni, and Zn) with a spinel structure have received wide attention. The catalytic effect of MFe₂O₄ on luminol CL reactions has been reported [21–23]. To the best of our knowledge, MFe₂O₄ has not been used as a carrier to synthesize CF-NMs. Considering inherent catalytic activity and superparamagnetism of MFe₂O₄, MFe₂O₄ may be utilized directly loading CL reagents to develop highly efficient CF-NMs without additional loading of catalysts.

Herein, we synthesized ABEI-functionalized CuFe₂O₄ magnetic nanoparticles (ABEI/CuFe₂O₄) by using a solvothermal and post-functionalization method. The as-prepared ABEI/CuFe₂O₄ was characterized by high-resolution transmission electron microscopy (HRTEM), scanning electron microscopy (SEM), X-ray photoelectron spectroscopy (XPS), X-ray diffraction (XRD), inductively coupled plasma-atomic emission spectrometry (ICP-AES), and ultraviolet-visible (UV-vis) absorption spectroscopy. The as-synthesized ABEI/CuFe₂O₄ showed good CL performance and superparamagnetism. The effect of various factors on CL properties of ABEI/CuFe₂O₄ was examined. The CL behavior of various ferrite/ABEI with different transition metal element was compared, including CuFe₂O₄, CoFe₂O₄, ZnFe₂O₄, MnFe₂O₄, and NiFe₂O₄. A dual route pioneered by CuFe₂O₄ as well as surface-rich Cu²⁺ and Cu⁰ for synergistic catalysis mechanism has been proposed. Moreover, based on sensitive CL response of ABEI/CuFe₂O₄ to H₂O₂, an enzyme-free sensor was constructed for detection of H₂O₂. The applicability of the H₂O₂ sensor in human serum samples was explored.

**Experimental section**

**Chemicals and materials**

Copper chloride dihydrate (CuC₁₂·2H₂O), iron chloride hexahydrate (FeC₁₃·6H₂O), sodium acetate (NaAc), trisodium citrate dihydrate (Na₃Cit·2H₂O), and ethylene glycol were purchased from Sinopharm Chemical Reagent Co. (Shanghai, China). Four millimolars of ABEI stock solution was obtained by dissolving ABEI (TCI, Japan) in 0.1 M NaOH solution and stored in a dark environment at 4 °C. H₂O₂ (30%, v/v) was provided by Xinke Chemical Reagent Factory (Bengbu, China) and diluted freshly before CL measurement. The reagents involved in the experiments were of analytical grade. The ultrapure water was obtained from a Milli-Q system (Millipore, France). Human serum samples were obtained from Anhui Chest Hospital.

**Synthesis of ABEI/CuFe₂O₄**

CuFe₂O₄ was prepared by referring to a previous work [24] with some modifications as follows. First, 0.2556 g CuC₁₂·2H₂O and 0.81 g FeC₁₃·6H₂O were dissolved in 20 mL ethylene glycol solution at room temperature. After the solution was clear, 1.64 g NaAc and a certain amount of Na₃Cit·2H₂O were added and stirred for 2 h. The obtained solution was transferred to an autoclave and subsequently placed at 200 °C for 16 h. After the reaction, the product was washed several times using ultrapure water and ethanol alternately. The precipitate was dried under vacuum for 6 h and stored at 4 °C. Next, 10 mg of the obtained powder was dispersed in ultrapure water by ultrasonication. Then, CuFe₂O₄ dispersion, ultrapure water, and ABEI solution were mixed by the volume ratio of 2:1:1. After shaking for 12 h, the mixture was magnetically separated. The resulted ABEI/CuFe₂O₄ was washed three times and then dispersed in ultrapure water and placed at 4 °C for further use.

**Characterizations**

The morphology of ABEI/CuFe₂O₄ was characterized by HRTEM (Talos F200X, FEI, USA). The SEM image of the nanomaterials was recorded by a cold field emission scanning electron microscopy (SU8220, Hitachi, Japan). The XPS data was obtained using X-ray photoelectron spectroscopy (MiniFlex600, Rigaku, Japan) with Cu Kα radiation (λ = 0.15406 nm). The elemental composition was measured by ICP-AES (OPTIMA 7000DV, PerkinElmer, USA). The loading efficiency of ABEI was measured by UV–vis spectrophotometry (Agilent 8453, Agilent Technologies, USA). The CL spectra was performed on a fluorescence spectrophotometer with the lamp on (F-7000, Hitachi, Japan). The data of the hysteresis curve was measured in the magnetic measurement system (MPMS3, Quantum Design, USA). The chemical bonds in the nanomaterials were analyzed with Fourier-transform infrared spectroscopy (FTIR) (EQUINX55, Bruker, Germany).
CL measurements

The measurements of CL kinetics, condition optimization, and stability were performed on a Centro LB 960 microplate luminometer (Berthold, Germany). The investigation of the CL mechanism of the nanomaterials was carried out on a CL detector (LUMIstar Omega, BMG LABTECH, Germany). Typically, 50 μL of ABEI/CuFe$_2$O$_4$ dispersion was injected into a 96-well microplate, and 50 μL of H$_2$O$_2$ alkaline solution was introduced as a co-reacting reagent. Generally, the CL signal was recorded every 0.1 s within 30 s, and the integrated intensity of the nanomaterials was recorded.

Procedures for serum samples

The human serum samples were processed as follows. Firstly, 20 μL of human serum was diluted to 2 mL with ultrapure water. The diluted sample was then centrifuged with an ultrafiltration tube at 4000 rpm for 5 min at 4 °C to thoroughly remove precipitates. After purification, CL measurements were performed immediately. In a recovery experiment, a certain concentration of H$_2$O$_2$ was added to samples, and processed according to the above steps.

Results and discussion

Synthesis and characterizations

ABEI/CuFe$_2$O$_4$ was synthesized by a solvothermal method with post-functionalization. The synthesis steps are briefly shown in Scheme 1. During the synthesis of CuFe$_2$O$_4$, the molar ratio of CuCl$_2$·2H$_2$O to FeCl$_3$·6H$_2$O was 1:2. Na$_3$Cit·2H$_2$O was used as an electrostatic stabilizer, and NaAc was used to adjust the pH of the reaction solution. Ethylene glycol was used as a reducing agent, which also provided a stable solvent environment to prevent from agglomeration at high temperature and generate CuFe$_2$O$_4$. After purification and drying, the product was redispersed in water, followed by shaking with ABEI for 12 h. Under neutral conditions, the final product ABEI/CuFe$_2$O$_4$ was obtained.

The size and morphology of ABEI/CuFe$_2$O$_4$ nanomaterials were observed by HRTEM and SEM. HRTEM (Fig. 1A) demonstrated that the nanomaterials were well-dispersed quasi-spheres with a relatively uniform particle size and an average diameter of about 97.5 ± 7.2 nm, which was roughly consistent with the SEM image (see Supplementary Information (ESM) Fig. S1). In addition, the surface of the nanoparticles was slightly rough to provide more adsorption sites. In Fig. 1B, the measured spacings of two adjacent planes in different directions were 0.30 nm and 0.25 nm, which corresponded finely to the lattice spacings in the (220) and (311) planes of CuFe$_2$O$_4$, respectively. To further investigate the elemental composition and distribution of ABEI/CuFe$_2$O$_4$, elemental mapping was performed. It could be seen in Fig. 1D–H that Cu, Fe, O, and N elements existed on the nanomaterials, which confirmed the successful preparation of CuFe$_2$O$_4$ and the subsequent effective adsorption of ABEI.

The crystalline phase structure of the nanomaterials was further determined by XRD measurements (Fig. 2). The positions and relative intensities of the diffraction peaks appearing in the synthesized CuFe$_2$O$_4$ (a) and ABEI/CuFe$_2$O$_4$ (b) did not change significantly, which showed that the addition of ABEI did not destroy the crystalline structure of CuFe$_2$O$_4$. The peaks at 2θ = 18.3°, 30.2°, 35.6°, 43.3°, 57.0°, and 62.7° in the spectra represented the crystallographic planes of cubic crystals CuFe$_2$O$_4$ at (111), (220), (311), (400), (511), and (440), respectively. Strikingly, it was also observed that the peaks at 2θ = 43.3°, 50.4°, and 74.1° corresponded to the (111), (200), and (220) diffraction planes of fcc Cu crystals. This was due to the excess presence of ethylene glycol as a strong reducing agent during the synthesis of CuFe$_2$O$_4$ by the solvothermal method at high temperatures, which made it possible that Cu$^{2+}$ could be reduced to Cu$^0$ [25, 26].

XPS was taken to further confirm the composition and elemental valence state in the nanomaterials. As shown in ESM Fig. S2, after the addition of ABEI, Fe, Cu, O, C, and N elements were found in the nanomaterials, which indicated the successful modification of ABEI. The component and content of the nanomaterials were determined by UV-vis as well as ICP-AES. CuFe$_2$O$_4$ had a wide absorption band between 200 and 1000 nm, while two characteristic absorption peaks of ABEI were located near 291 nm and 320 nm (ESM Fig. S3A). The UV-vis curve of ABEI/CuFe$_2$O$_4$ demonstrated broad absorption band of CuFe$_2$O$_4$ and two characteristic absorption peaks of ABEI, which proved again that ABEI had been successfully modified to the surface of CuFe$_2$O$_4$.
nanospheres. In addition, ABEI loaded on the nanomaterials was about 52.0%. Moreover, the contents of Cu and Fe in ABEI/CuFe$_2$O$_4$ were calculated to be 272.5 mg/g and 304.0 mg/g by ICP-AES, respectively.

The assembly mechanism of ABEI/CuFe$_2$O$_4$ was discussed in detail. As shown in FTIR (ESM Fig. S4), the peak position at 3446 cm$^{-1}$ was a typical -OH peak [24], indicating the presence of a large number of oxygen-containing functional groups on the surface of CuFe$_2$O$_4$, which facilitated the subsequent functionalization of the molecule. Since the ABEI molecule contained -NH$_2$ and CuFe$_2$O$_4$ possessed abundant -OH, the hydrogen bonding interactions were likely to occur between the two groups [27, 28]. Furthermore, the surface of CuFe$_2$O$_4$ contained Cu$^{2+}$ and Cu$^0$, which allowed ABEI to coordinate with Cu elements. Therefore, it was suggested that ABEI could be connected on the surface of the nanomaterials by the hydrogen bonding between the hydroxyl group of CuFe$_2$O$_4$ and the amino group of ABEI and the coordination between Cu element and the amino group of ABEI.

**Magnetic property of ABEI/CuFe$_2$O$_4$**

The hysteresis test curves of CuFe$_2$O$_4$ and ABEI/CuFe$_2$O$_4$ collected at room temperature are shown in Fig. 3. The above nanomaterials were both free of hysteresis and showed excellent superparamagnetism. The saturation magnetization strengths of CuFe$_2$O$_4$ and ABEI/CuFe$_2$O$_4$ are 39.01 emu/g and 38.36 emu/g, respectively, which did not change. Under the force of an applied magnet, ABEI/CuFe$_2$O$_4$ could be separated simply from the aqueous phase within 2 min.

**CL behavior of ABEI/CuFe$_2$O$_4$**

The CL properties of ABEI/CuFe$_2$O$_4$ were studied on a microplate luminometer. As shown in Fig. 4A, when ABEI/CuFe$_2$O$_4$ reacted with H$_2$O$_2$, strong light emission was observed, which was 397 times that of ABEI-H$_2$O$_2$ system. When the mixture of ABEI and CuFe$_2$O$_4$ reacted with H$_2$O$_2$, much weaker CL emission was generated, but it was
also 220 times that of ABEI-H$_2$O$_2$ system. The results implied that the immobilization of ABEI on the surface of CuFe$_2$O$_4$ was beneficial for the CL emission. Furthermore, instead of CuFe$_2$O$_4$, Fe$_3$O$_4$ was used to prepare ABEI/Fe$_3$O$_4$. The CL intensity of ABEI/Fe$_3$O$_4$-H$_2$O$_2$ system was only 13% of that of ABEI/CuFe$_2$O$_4$-H$_2$O$_2$ system. This result demonstrated that copper played an important role in the CL emission. The CL spectra of ABEI/CuFe$_2$O$_4$ and ABEI reacting with H$_2$O$_2$ are presented in Fig. 4B. It was found that maximum CL emission of ABEI/CuFe$_2$O$_4$ was around 445 nm, which was consistent with that of ABEI-H$_2$O$_2$ CL system, indicating that the CL emission of the nanomaterials was generated by the reaction of ABEI with H$_2$O$_2$ [29].

In addition, since transition metal elements have a significant catalytic effect on the CL reaction, the CL behavior of various ABEI/ferrite with different transition metal element (M) was also examined. CuFe$_2$O$_4$, CoFe$_2$O$_4$, ZnFe$_2$O$_4$, MnFe$_2$O$_4$, and NiFe$_2$O$_4$ were mixed with equal amounts of ABEI and reacted under the same conditions to obtain the corresponding functionalized nanomaterials ABEI/MFe$_2$O$_4$. As shown in Fig. 5A and B, ABEI/CuFe$_2$O$_4$, ABEI/CoFe$_2$O$_4$ showed higher CL signal than that of ZnFe$_2$O$_4$, MnFe$_2$O$_4$, and NiFe$_2$O$_4$. Moreover, ABEI/CoFe$_2$O$_4$ was a typical flash-type emission, while ABEI/CuFe$_2$O$_4$ had a slower kinetic curve, which was advantageous for analytical accuracy.

The effects of various factors on the CL properties of the nanomaterials were investigated, including the concentration of electrostatic stabilizer Na$_3$Cit·2H$_2$O and ABEI/CuFe$_2$O$_4$, the pH of the ABEI solution, and the pH and concentration of H$_2$O$_2$. It was not difficult to find that the particle size of CuFe$_2$O$_4$ became more uniform and better dispersed with increasing Na$_3$Cit·2H$_2$O concentration (ESM Fig. S5). Meanwhile, the CL signal increased with the increase of Na$_3$Cit·2H$_2$O concentration. Among them, the strongest signal was observed with 2.0 mM Na$_3$Cit·2H$_2$O (ESM Fig. S6). The highest CL intensity of ABEI/CuFe$_2$O$_4$ was obtained when the ABEI solution was neutral (ESM Fig. S7). Furthermore, 0.25 mg/mL ABEI/CuFe$_2$O$_4$ and 0.1 mM H$_2$O$_2$ (pH = 12.0) was selected for the subsequent detection (ESM Figs. S8–10).

The stability of the nanomaterials was studied in a day and in a month as well as in different batches. As seen in ESM Fig. S11, ABEI/CuFe$_2$O$_4$ maintained a stable CL signal within 24 h and 30 days. Moreover, the signals were examined for the five batches of nanomaterials synthesized at different times and excellent reproducibility between batches was
obtained. Minor fluctuations in the measured signal values around 1,150,000 A.U. were observed. The relative standard deviations (R.S.D.) of the measured CL signals were calculated to be 1.2%, 1.3%, and 1.2% for repeated measurements within a day and within a month and between 5 batches, respectively. The above results indicated that the synthesized ABEI/CuFe$_2$O$_4$ possessed good CL stability.

**CL reaction mechanism of ABEI/CuFe$_2$O$_4$ with H$_2$O$_2$**

The as-prepared ABEI/CuFe$_2$O$_4$ nanomaterials exhibited superior CL properties, which might be related to the surface catalytic properties of CuFe$_2$O$_4$ nanospheres. It was reported that the enhancement of the ABEI CL system by catalysts was due to the catalysts could facilitate the formation of CL intermediates such as superoxide radicals ($O_2^{•−}$) and hydroxyl radicals (OH$^{•}$). Previously, it was also reported that CuFe$_2$O$_4$ with peroxidase-like catalytic activity could catalyze the decomposition of H$_2$O$_2$ to generate OH$^{•}$, which was beneficial to enhance the efficiency of CL reactions [30]. Besides, Cu$^{2+}$ and Cu$^0$ was able to produce OH$^{•}$ indirectly in the presence of H$_2$O$_2$ [31–33]. It was speculated that CuFe$_2$O$_4$, Cu$^{2+}$, and Cu$^0$ may facilitate the generation of $O_2^{•−}$ and OH$^{•}$ during ABEI CL reaction, leading to high CL efficiency.

To determine whether or not the dissolved oxygen, $O_2^{•−}$, and OH$^{•}$ were involved in the CL reaction, the effects of nitrogen (N$_2$), oxygen (O$_2$), and two free radical scavengers SOD and thiourea, suggesting that $O_2^{•−}$ and OH$^{•}$ took part in the CL reaction. Accordingly, CuFe$_2$O$_4$, Cu$^{2+}$, and Cu$^0$ did facilitate the generation of $O_2^{•−}$ and OH$^{•}$ during ABEI CL reaction, resulting in strong light emission. The CL mechanism is suggested as shown in Fig. 6.

**Detection of H$_2$O$_2$**

Hydrogen peroxide (H$_2$O$_2$) is not only a by-product of active oxygen metabolism in organisms, but also a key regulator of various oxidative stress-related states. It is closely related to the occurrence and development of some diseases [34, 35]. Therefore, it is very important to develop stable and efficient H$_2$O$_2$ detection methodologies. In view of the excellent CL behavior of ABEI/CuFe$_2$O$_4$ and its sensitive response to H$_2$O$_2$, a non-enzymatic H$_2$O$_2$ sensor was constructed. Under the optimal experimental conditions, H$_2$O$_2$ solution was diluted at different concentrations. As shown in Fig. 7A, the CL intensity was linear with H$_2$O$_2$ concentration in the range of 10.0 nM to 0.1 mM. The linear regression equation was fitted as $I_{\text{integrated}} = 661746.0 \times C(\text{H}_2\text{O}_2) + 33355.5$, and the correlation coefficient ($R^2$) was 0.999. The limit of detection (LOD) could be as low as 5.6 nM when the signal-to-noise ratio (S/N) was 3. Table S1 (see ESM) summarized several representative non-enzymatic H$_2$O$_2$ assays developed in recent years. Compared to the previously reported methods, the proposed sensor demonstrated more sensitive detection capabilities with improved linear range and detection limits.

To verify the selectivity of this sensor, seven common interferents were selected instead of H$_2$O$_2$ in CL assay, including glucose, glycine, NADH, sodium hypochlorite, sodium nitrite, uric acid, and lactic acid. The concentration of these...
species \( (1.0 \times 10^{-5} \text{ M}) \) was set to 10 times the concentration of \( \text{H}_2\text{O}_2 \) to be measured. As shown in Fig. 7B, the strong CL response of our nanomaterials occurred only in the presence of \( \text{H}_2\text{O}_2 \), which clearly illustrated the excellent selectivity of the proposed sensor.

\( \text{H}_2\text{O}_2 \) has played an important role in biological signal transduction pathways. It has been closely associated with the occurrence and development of tumors as well as some inflammatory diseases. Generally, \( \text{H}_2\text{O}_2 \) expresses at a high level in some cancers or during chronic inflammation [36–38]. Clinically, the detection of \( \text{H}_2\text{O}_2 \) is helpful for tumor diagnosis. Therefore, lung cancer was chosen as a model to explore the applicability of the sensor in human serum samples. Before the assay, human serum samples, including healthy human serum samples and lung cancer patient serum samples, were collected and diluted 100 times, and then purified by centrifugation using an ultrafiltration tube to thoroughly remove precipitates. The results are summarized in Table 1. The sensor could efficiently discriminate healthy and lung cancer patients. The sample recoveries ranged from 99.4 to 105.2\%, confirming that the sensor might be applied for the detection of real human serum samples.

**Conclusion**

In this work, a new kind of CL functionalized magnetic nanomaterials with unique catalytic activity was synthesized. Firstly, the \( \text{CuFe}_2\text{O}_4 \) nanomaterials were prepared by a simple solvothermal method, and then reacted with ABEI to form ABEI/\( \text{CuFe}_2\text{O}_4 \). The synthesized ABEI/\( \text{CuFe}_2\text{O}_4 \) was quasi-spherical with a rough surface, and the average particle size was 97.5 ± 7.2 nm. The ABEI/\( \text{CuFe}_2\text{O}_4 \) nanomaterials without additional catalysts showed unique CL properties, superior to ABEI-CuFe\(_2\)O\(_4\) in liquid phase. This reveals that the immobilization of ABEI on the surface of CuFe\(_2\)O\(_4\) exhibits unique heterogeneous catalytic property. Moreover, the catalytic ability of CuFe\(_2\)O\(_4\) was better than that of CoFe\(_2\)O\(_4\), ZnFe\(_2\)O\(_4\), MnFe\(_2\)O\(_4\), and NiFe\(_2\)O\(_4\). CuFe\(_2\)O\(_4\) with peroxidase-like activity exposed a large amount of Cu elements on the surface, which opened up a \( \text{H}_2\text{O}_2 \) dual-route

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**Table 1. Results of \( \text{H}_2\text{O}_2 \) assay in human serum samples**

| Sample                | Initial \( (\times 10^{-9} \text{ mol/L} ) \) | Added | Found \( \pm \text{SD} \) | Recovery (%) |
|-----------------------|-----------------------------------------------|-------|--------------------------|--------------|
| Healthy \( (\times 10^{-9} \text{ mol/L} ) \) | 1 -                                           | 50.0  | 52.0 ± 0.4               | 104.0        |
|                       | 2 -                                           | 50.0  | 51.6 ± 0.6               | 103.1        |
|                       | 3 -                                           | 50.0  | 49.9 ± 0.3               | 99.8         |
| Lung cancer \( (\times 10^{-6} \text{ mol/L} ) \) | 1.0 ± 0.1                                    | 50.0  | 52.0 ± 3.8               | 102.1        |
|                       | 6.3 ± 0.3                                    | 50.0  | 56.0 ± 0.4               | 99.4         |
|                       | 8.1 ± 0.3                                    | 50.0  | 61.1 ± 1.1               | 105.2        |

*a Mean value ± SD of three independent experiments, \( n = 3 \)*
synergistic catalysis, resulting in strong light emission. In addition, ABEI/CuFe$_2$O$_4$ demonstrated good superparamagnetism, which may be used to separate the sample matrix from target in bioassays. Finally, an enzyme-free sensor for H$_2$O$_2$ was developed. H$_2$O$_2$ could be detected from 10.0 nM to 0.1 mM with a low detection limit of 5.6 nM and good selectivity. The sensor could be applied to determine H$_2$O$_2$ in real human serum samples. It is of great application potential in clinical diagnosis.

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Author contribution Yuyang Wu designed the nanomaterial, conducted experimental work, and wrote the original draft. Jue Wang contributed to the design of the study and revision of the manuscript. Hua Cui directed the study and wrote the manuscript.

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Declarations

Ethics approval The research was approved by the Ethics Committee of Anhui Chest Hospital. All volunteers were informed of and agreed with the objectives of the study.

Conflict of interest Hua Cui is an editor for Analytical and Bioanalytical Chemistry, but was not involved in the peer review of this work. The authors declare no competing interests.

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