Synthesis, Characterization and Cytotoxicity of Novel Multifunctional Fe$_3$O$_4$@SiO$_2$@GdVO$_4$:Dy$^{3+}$ Core-Shell Nanocomposite as a Drug Carrier

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Academic Editor: Dusan Losic
Received: 22 January 2016; Accepted: 24 February 2016; Published: 3 March 2016

Abstract: In this study, multifunctional Fe$_3$O$_4$@SiO$_2$@GdVO$_4$:Dy$^{3+}$ nanocomposites were successfully synthesized via a two-step method. Their structure, luminescence and magnetic properties were characterized by X-ray diffraction (XRD), scanning electronic microscope (SEM), transmission electron microscopy (TEM), photoluminescence (PL) spectra and vibrating sample magnetometer (VSM). The results indicated that the as-prepared multifunctional composites displayed a well-defined core-shell structure. The composites show spherical morphology with a size distribution of around 360 nm. Additionally, the composites exhibit high saturation magnetization (20.40 emu/g) and excellent luminescence properties. The inner Fe$_3$O$_4$ cores and the outer GdVO$_4$:Dy$^{3+}$ layers endow the composites with good responsive magnetic properties and strong fluorescent properties, which endow the nanoparticles with great potential applications in drug delivery, magnetic resonance imaging, and marking and separating of cells in vitro.

Keywords: Fe$_3$O$_4$@SiO$_2$@GdVO$_4$:Dy$^{3+}$; nanocomposites; core-shell structure; magnetic property; luminescence property

1. Introduction

In recent years, controlled drug delivery systems for modern drug therapy have been attracting increasing attention because they exhibit low toxicity, a wide therapeutic window, and ideal drug efficacy as compared to conventional drug delivery systems [1,2]. The multifunctional nanocomposites combine with magnetic and luminescent properties in one entity, and they have attracted great attention in recent years owing to their potential application in the biotechnology and nanomedicine fields including magnetic resonance imaging (MRI), cell separation, drug delivery agents, cell separation, labeling, and optical probes [3–5]. In the choice of luminescent nanomaterials for labeling, targeting and imaging, lanthanide-doped nanomaterials possess many of advantages such as high fluorescence quantum yields, low toxicity, long lifetimes, and high stability in comparison to quantum dots and organic dyes [5–8].

So far, there have been some reports of constructing multifunctional nanomaterials that were made up of Fe$_3$O$_4$ and lanthanide-doped nanomaterials. In these reports [8–12], if the lanthanide-doped nanomaterials are chosen as cores, their luminescent intensity may be suppressed to some extent due to the coating of the outer layers. Meanwhile, if the lanthanide-doped nanomaterials are in direct contact with Fe$_3$O$_4$, their luminescence may be decreased as the direct contact can cause fluorescence-quenching [13–15]. Therefore, a SiO$_2$ mid-layer between Fe$_3$O$_4$ and lanthanide-doped nanomaterials is needed.

To the best of our knowledge, there are no previous reports on the combination of magnetic properties with gadolinium vanadate nanophosphors. The previous investigation results indicated that
nanosized GdVO₄:Ln³⁺ phosphors have a significant application in a high definition flat display panels and potential applications in biology [16–19]. Compared with Ln³⁺-activated YVO₄, GdVO₄:Ln³⁺ exhibits highly efficient emitting phosphors, in which the energy transfers from the GdVO₄ host to the incorporated Ln³⁺ ions through V⁵⁺–O²⁻ charge transfer (CT), yielding an efficient luminescence of Ln³⁺ activators.

Herein, we develop, for the first time, a novel and simple route to prepare Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ core-shell microspheres with excellent magnetic and luminescence properties. The good aqueous colloidal stability, low toxicity and excellent self-heating efficacy make these novel magnetic, luminescent nanomaterials suitable for the hyperthermia treatment of cancer, and the luminescent entity helps us to identify the location of magnetic nanoparticles during in vitro cellular imaging [20–24].

2. Results and Discussion

Figure 1 depicts the X-ray diffraction (XRD) patterns of as-synthesized Fe₃O₄, Fe₃O₄@SiO₂ and Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ nanoparticles. From Figure 1, we can find that there are characteristic diffraction peaks of Fe₃O₄, with a face-centered-cubic structure in all curves according to JCPDS card No. 65-3107. Besides the corresponding peaks of Fe₃O₄, SiO₂ (JCPDS card No. 29-0085) and GdVO₄ (JCPDS card No.86-0996) can be detected in Figure 1a–c, respectively. No peaks corresponding to impurities are detected, showing the adequate purity of the Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ composites.

![Figure 1. X-ray diffraction (XRD) patterns of pure Fe₃O₄ (a); Fe₃O₄@SiO₂ (b) and Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ (c). The diffraction peaks that are indexed in 1c correspond to GdVO₄.](image)

The morphology and size details of the composites were characterized by SEM (scanning electronic microscope) and TEM (transmission electron microscopy) images. SEM investigations, as displayed in Figure 2a, reveal that the magnetic cores of Fe₃O₄ particles are of a rough appearance and have an average size of 290 (±20) nm. Once coated with one layer of silica, the composite microspheres are slightly larger in diameter and have a relatively smooth surface, with their size increased up to 320 (±30) nm, as shown in Figure 2b. The average size of the core-shell nanocomposites finally increased up to 360 (±25) nm, as illustrated in Figure 2c. The representative TEM images in Figure 2e,f indicate that the nanocomposites exhibit a core-shell structure.

To estimate the magnetic sensitivity, the room temperature magnetization hysteresis loops of the as-prepared cores and core-shell nanocomposites were collected and displayed in Figure 3. The magnetic hysteresis loops in Figure 3 indicate that they have saturation magnetizations of 83.9 emu/g (Fe₃O₄), 27.8 emu/g (Fe₃O₄@SiO₂) and 20.4 emu/g (Fe₃O₄@SiO₂@GdVO₄:Dy³⁺) as well as negligible coercivity at room temperature, implying characteristics of their strong magnetism. The reduction of saturation magnetization could be attributed to the nonmagnetic shells (SiO₂ and GdVO₄:Dy³⁺). Our study revealed that, though the magnetism of the core-shell nanocomposites is
less than that of the bare magnetic cores, it still possesses enough magnetic response for biomedical applications such as MRI, which is effectively magnetic separation.

![Figure 2](image-url)  
**Figure 2.** Scanning electronic microscope (SEM) images of Fe₃O₄ (a); Fe₃O₄@SiO₂ (b); Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ (c); and transmission electron microscopy (TEM) images of Fe₃O₄ (d); Fe₃O₄@SiO₂ (e); Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ (f).

![Figure 3](image-url)  
**Figure 3.** The magnetic hysteresis loops of pure Fe₃O₄ (a); Fe₃O₄@SiO₂ (b); and Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ (c).

The photoluminescence spectra of Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ are shown in Figure 4. In the excitation spectra (Figure 4A), the excitation band at 300–350 nm monitored with a 571 nm emission of ⁴F₉/₂→⁶H₁₅/₂ electronic transition of Dy³⁺ can be attributed to a charge transfer through the V–O bond overlay of the Dy–O charge transfer band. The emission spectra of GdVO₄:Dy³⁺ are shown in Figure 4B. The main emission peaks at 481 nm and 571 nm are results of the ⁴F₉/₂→⁶H₁₅/₂ transition and ⁴F₉/₂→⁶H₁₃/₂ transition of Dy³⁺ ions. Moreover, Figure 4 shows the excitation spectra and emission spectra of Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ composites with different doped concentrations of Dy³⁺ ions. It is shown that the optimum doped concentration of Dy³⁺ ions in the Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ composites is 1 mol %.

To investigate the porous structure of the Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ nanocomposites, the N₂ adsorption-desorption isotherms were investigated and are shown in Figure 5. This isotherm profile can be categorized as type IV, with a small hysteresis loop observed at a relative pressure of 0.05–1.0, indicating the mesoporous features. The inset in Figure 5 is the pore size distribution. As calculated by the Brunauer-Emmett-Teller (BET) method, Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ nanocomposites’ core-shell structure gives rise to a BET area of 30.21 m²·g⁻¹, with a relatively high pore volume of 0.212 cm³·g⁻¹,
and the average pore diameter is 17.46 nm. The BET indicated the potential of such nanostructures for drug delivery applications.

To evaluate the cytotoxicity of the Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ nanoparticles, in vitro cytotoxicity tests against HeLa cells were carried out. From the 3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyltetrazolium bromide (MTT) viability histogram, shown in Figure 6, we can find that the Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ causes insignificant damage to the HeLa cells when the sample concentration increases to 200 μg mL⁻¹ for 24 h, and the cell viability remains at 90.38% even at the highest concentration, which indicates that Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ nanoparticles are biocompatible.

**Figure 4.** Excitation spectra (A) and emission spectra (B) of Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ with different doped concentrations of Dy³⁺ (a: 0.5%, b: 1%, c: 2%, d: 3% and e: 4%).

**Figure 5.** The N₂ adsorption/desorption isotherms and pore size distribution (inset) of Fe₃O₄@SiO₂@GdVO₄:Dy³⁺.

**Figure 6.** The viability histograms of HeLa cells incubated with different concentrations of Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ nanoparticles for 24 h measured by a 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay.
3. Materials and Methods

3.1. Materials

All reagents are of analytical reagent grade and used without further purification. Gd₂O₃ (99.9%) and Eu₂O₃ (99.9%) were purchased from Jinan Camolai Trading Company, Ferrous chloride hexahydrate (FeCl₂·6H₂O) (99%), tetraethyl orthosilicate (TEOS, 99.0%), sodium acetate (NaAc), Citrate acid monohydrate were purchased from Beijing Chemicals Corporation. Nitric acid, ethanol, ethylene glycol (EG), and ammonia aqueous (25%) were purchased from Tianjin Chemicals Corporation. Deionized water obtained from the Milli-Q system (Millipore, Bedford, MA, USA) was used in all experiments. The magnetic Fe₃O₄ nanoparticles were prepared using a modified solvothermal reaction.

3.2. Synthesis of Fe₃O₄

The magnetic Fe₃O₄ nanoparticles were prepared according to a previously reported synthetic process [19]. Typically, FeCl₃·6H₂O (1.3495 g) and NaAc (7.1926 g) were dissolved in EG solution (40 mL). Then PEG-10000 (1.0015 g) was added with vigorous stirring and the mixture was stirred for 30 min to form a homogeneous russet solution. The obtained solution was transferred to a Teflon-lined autoclave (50 mL capacity) and heated at 200 °C for 10 h. Subsequent cooling to room temperature yielded black magnetite particles, which were washed with ethanol and deionized water three times, respectively, and dried at 60 °C for 12 h.

3.3. Synthesis of Fe₃O₄@SiO₂

Fe₃O₄@SiO₂ nanoparticles were prepared according to the modified by the Stöber method. In brief, 1.0 g of Fe₃O₄ nanoparticles were homogeneously dispersed in a mixture of 160 mL of ethanol, 40 mL of deionized water, and 3.0 mL of 28 wt % concentrated ammonia aqueous solution, followed by the addition of 3.0 mL of tetraethyl orthosilicate (TEOS). After vigorous stirring at 40 °C for 6 h, the obtained Fe₃O₄@SiO₂ microspheres were separated with a magnet and washed repeatedly with ethanol and deionized water to remove nonmagnetic by products.

3.4. Synthesis of Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ Nanoparticles

Functionalization of GdVO₄:Dy³⁺ on the template Fe₃O₄@SiO₂ was achieved according to the reported process with a doping concentration of Dy³⁺ of 0.5–4 mol % to Dy³⁺ in GdVO₄:Dy³⁺. The typical procedure for synthesis is described as follows: stoichiometric amounts of Gd₂O₃, Dy₂O₃ and citric acid were dissolved in dilute nitric acid with heating followed by the addition of NH₄VO₃ in distilled water. Then PEG-10000 was added with a concentration of 0.05 g·mL⁻¹. After stirring for 0.5 h, a homogenous sol was formed. Then the desired amount of Fe₃O₄@SiO₂ nanoparticles was added into the gel, after further stirring for another 3 h, the resulting material was dried at 120 °C for 12 h to obtain the precursors. Then the precursors were calcined at 700 °C for another 4 h. The obtained nanoparticles were denoted as Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ (Scheme 1).

![Scheme 1. Illustration for the synthesis process of the spherical Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ nanocomposite.](image)

3.5. Cytotoxicity Study of Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ Nanoparticle

Cell viabilities of Fe₃O₄@SiO₂@GdVO₄:Dy³⁺ nanoparticles at different concentrations were tested by MTT assay on HeLa (human cervical cancer cells). In the experiment, the corresponding untreated
cells were used as control. First, the HeLa cells were pre-incubated in a 96-well plate (about 3000 cells per well) for 24 h. Second, 2 mg of the Fe$_3$O$_4$@SiO$_2@$GdVO$_4$:Dy$^{3+}$ nanoparticles were added into 10 mL of 0.01 M phosphate buffered saline (PBS, pH = 7.4) to form a stable orange solution. Third, the above solution at concentrations of 6.25, 12.5, 25, 50, 100 or 200 µg mL$^{-1}$ was added to the cells. Six parallel-group experiments were simultaneously conducted for each concentration. After 24 h, the viability of HeLa cells was examined by a MTT assay.

3.6. Characterization

The purities of all the nanoparticles were checked by X-ray diffraction (XRD) measurements at room temperature using Cu Kα radiation (Kα = 1.54059 Å). The morphology and microscope structure of all the nanocomposites were characterized by a scanning electronic microscope (SEM, NoVa™ Nano SEM 430, FEI Co., Ltd., Hillsboro, OR, USA) and transmission electron microscopy (TEM, JEOL JEM-2100F, JEOL Co., Ltd., Tokyo, Japan). The room temperature magnetic hysteresis (M-H) loops were measured using a superconducting quantum interference device vibrating sample magnetometry (SQUID-VSM, Quantum Design Co., Ltd., San Diego, CA, USA). Luminescence spectra were recorded on a FluoroMax-4 spectrophotometer (HORIBA Jobin Yvon Co., Ltd., Paris, France). The specific surface area was determined by the Brunauer-Emmett-Teller (BET) method. The HeLa cells were assayed for viability by using a microplate reader (Bio-Rad 680, Bio-Rad Co., Ltd., Hercules, CA, USA).

4. Conclusions

In summary, we report a novel magnetic/luminescence multifunctional nanocomposite, Fe$_3$O$_4$@SiO$_2@$GdVO$_4$:Dy$^{3+}$, with a core-shell structure from a combination of hydrothermal reaction and the sol-gel process. The as-prepared nanocomposites, combining the merits of the good magnetic response of the assembled Fe$_3$O$_4$@SiO$_2$ microspheres and the fluorescence property of GdVO$_4$:Dy$^{3+}$, displayed high surface area and biocompatibility. Therefore, our study may provide new insight and useful information for the design of diverse, functional nanocomposites as drug carriers.

Acknowledgments: This work is supported by the National Natural Science Foundation of China (Nos. 21401112, 21301100).

Author Contributions: Huitao Fan, and Bo Li were involved in designing the aim of this manuscript, performed the experiments and prepared the manuscript. Qiang Zhao performed the room temperature magnetic hysteresis (M-H) loops measurements and analyzed the data. Congcong Wang helped with several analyses. Each contributor was essential to the production of this work.

Conflicts of Interest: The authors declare no conflict of interest.

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