

Preparation of Monodisperse Magnetic Polystyrene Microspheres and Its Surface Chemical Modification

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ABSTRACT: Monodisperse magnetic polystyrene (PS) microspheres were prepared in the presence of PS seed particles and styrene-based magnetic colloid by the method of magnetic colloid swelling polymerization. The PS seed particles were prepared in advance by soap-free emulsion polymerization. Styrene-based magnetic colloid was used for swelling the PS seed particles in the magnetic colloid swelling polymerization process. After polymerization, functional amino groups were introduced onto the surface of the magnetic PS microspheres by surface Friedel-Crafts acylation reaction. The morphology, size distribution, and magnetic properties of magnetic PS microspheres were characterized with scanning electron microscopy (SEM) and vibrating sample magnetometer

(VSM), respectively. SEM showed that the magnetic PS microspheres had an average size of 1078 nm with a narrow size distribution. VSM showed that the magnetic PS microspheres were superparamagnetic, and saturation magnetization was found to be 5.714 emu/g. The concentration of functional amino groups on the surface of magnetic PS microspheres was measured by atomic absorption spectroscopy and UV-Vis spectroscopy, and the concentration of amino groups was found to be 0.168 mmol/g. © 2011 Wiley Periodicals, Inc. *J Appl Polym Sci* 120: 3278–3283, 2011

Key words: magnetic polymers; nanoparticle; polystyrene; surface modification; swelling

INTRODUCTION

Owing to superparamagnetic properties, magnetic polymer microspheres with functional groups have been widely used in the fields of enzyme immobilization,¹ protein separation,^{2–4} cell isolation,^{5,6} targeting drug,⁷ and immunoassay.⁸ The magnetic polymer microspheres used in these fields must be provided with such properties as follows: narrow size distribution, high magnetite content, and a high density of surface functional groups.

Many approaches have been used to prepare superparamagnetic polymer microspheres including miniemulsion polymerization,^{9,10} suspension polymerization,^{11,12} dispersion polymerization,¹³ emulsion polymerization,¹⁴ and seed swelling polymerization.^{15,16} These methods need further improvement for poor monodispersity, low magnetite content, or density of surface functional groups. Many types of organic monomer have been used to

prepare magnetic polymer microspheres, methyl methacrylate (MMA), styrene (St), vinyl acetate (VAc)^{12,17} are the most frequently used monomer. Monomer swelling polymerization is an effective approach to prepare monodisperse polymer microspheres.^{18–20} Frequently, the method is used to prepare nonmagnetic or porous polymer microspheres. Until now, there was no report on the preparation of magnetic polymer microspheres by the method of magnetic colloid swelling polymerization.

Amino groups, hydroxyl groups, or other groups were introduced onto the surface of the magnetic polymer microspheres by the method of surface modification. Generally, it is very difficult to introduce the surface functional groups onto the surface of the magnetic polystyrene (PS) microspheres because these surface functionalization reaction are carried out under a strong acidic circumstance, which causes the rapid degradation of magnetic property within the magnetic polymer microspheres. Recently, Yang et al.²¹ reported a method for the surface functionalization of magnetic PS microspheres without destroying the magnetic property by surface chemical reaction.

In this study, we describe an effective method to prepare the highly functionalized, monodisperse magnetic PS microspheres. The magnetic PS microspheres were prepared in the presence of PS seed

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particles and styrene-based magnetic colloid by magnetic colloid swelling polymerization. The functional amino groups were introduced onto the surface of the magnetic PS microspheres by surface Friedel-Crafts acylation reaction. The morphology, size distribution, magnetic properties, and surface amino groups of magnetic PS microspheres were characterized by scanning electron microscopy (SEM), vibrating sample magnetometer (VSM), and atomic absorption spectroscopy (AAS), respectively.

EXPERIMENTAL

Materials

Styrene (St) and divinylbenzene (DVB) were distilled under reduced pressure to remove the inhibitor prior to use. Unless stated otherwise, all other materials were of analytical grade and used without any further purification including aqueous ammonia (25%), ferric chloride hexahydrate ($\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$), ferrous chloride tetrahydrate ($\text{FeCl}_2 \cdot 4\text{H}_2\text{O}$), oleic acid, sodium dodecylbenzene sulfonate (DBS), benzoyl peroxide (BPO), ethylenediamine, chloroacetic acid, dibutylphthalate (DBP), 1,2-dichloroethane, anhydrous aluminum chloride (AlCl_3), EDTA disodium salt (EDTA-2Na), potassium persulfate (KPS), and cupric sulfate ($\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$).

Preparation of oleic acid-coated magnetic Fe_3O_4 nanoparticles

The oleic acid-coated magnetic Fe_3O_4 nanoparticles were prepared by a coprecipitation method with some modifications.²² Under nitrogen gas, 23.4 g of $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ and 8.6 g of $\text{FeCl}_2 \cdot 4\text{H}_2\text{O}$ were dissolved in 500 mL deionized water. When the solution was heated to 80°C, 28 mL of $\text{NH}_3 \cdot \text{H}_2\text{O}$ were added. Then 15 mL oleic acid was added dropwise within 20 min. The Fe_3O_4 gel was cooled to room temperature and washed several times with deionized water. The resultant was dissolved in styrene to form styrene-based magnetic colloid.

Preparation of polystyrene seed particles by soap-free emulsion polymerization

PS seed particles were prepared by soap-free emulsion polymerization.²³ In a 250-mL three-neck flask equipped with a stirrer and a condenser, 9.75 mL styrene and 0.545 mL DVB were mixed in 100 mL deionized water. 40 mL KPS aqueous solution containing 0.05 g KPS was added to the solution which was heated to 70°C. The polymerization reaction proceeded for 12 h. The particles were separated by centrifugation, and washed with deionized water several times.

Preparation of magnetic polystyrene microspheres by magnetic colloid swelling polymerization

In 50 mL of 0.25% DBS aqueous solution, 5 mL of styrene containing oleic acid-coated magnetic Fe_3O_4 nanoparticles was added. In another 50 mL of 0.25% DBS aqueous solution, 15 mL of seed PS particles were added. The two solutions were stirred for 3 h at room temperature, respectively, and then mixed in a 250-mL three-neck flask equipped with a stirrer and a condenser for 12 h at room temperature for the purpose of monomer swelling. When the temperature increased to 70°C, 0.1 g of BPO was added to initiate the polymerization for another 12 h. The resultant magnetic PS microspheres were separated by magnetic decantation and washed three times with deionized water.

Surface functionalization of magnetic polystyrene microspheres with amino groups

Amino groups were introduced to magnetic PS microspheres by the method of Friedel-Crafts acylation reaction²¹. 4 g of magnetic PS microspheres were mixed in 60 mL of 1,2-dichloroethane in a 250-mL three-necked flask equipped with a condenser and a stirrer. 6.3 g of anhydrous aluminum chloride (AlCl_3) and 4.4 g of chloroacetyl chloride were dissolved in 60 mL 1,2-dichloroethane to form a complex for 1 h at 30°C. Then the complex was dropped into the magnetic PS microspheres solution at the rate of 1 drop/3 s. The resultant magnetic PS microspheres were washed with ethanol and deionized water for several times.

The acylated magnetic PS microspheres were mixed with ethylenediamine at the ratio of 1:4. The reaction was carried out at 60°C for 5 h after the pH of the complex was adjusted to 11.0 with sodium hydrocarbonate, and the magnetic PS microspheres with amino groups were obtained.

Characterization

The morphology, size distribution was characterized with SEM. Through the evaluation of the SEM photographs, the number-average diameter (D_n) of the magnetic PS microspheres was calculated according to eq. (1), where N_i is the number of microspheres with diameter D_i (μm):

$$D_n = \frac{\sum N_i D_i}{\sum N_i} \quad (1)$$

The magnetic curves of the magnetic PS microspheres were recorded by VSM.

The concentration of the amino group introduced onto the surface of magnetic PS microspheres were calculated by the concentration of the Cu^{2+}

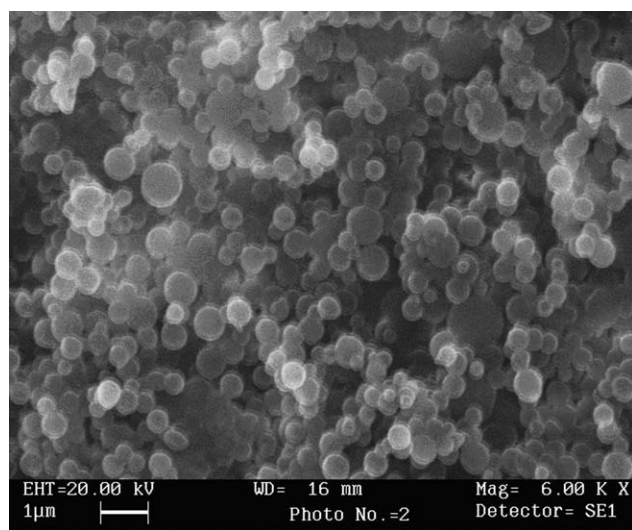


Figure 1 SEM photograph of the PS seed particles prepared by the soap-free emulsion polymerization.

immobilized on the magnetic PS microspheres, where the amino groups were transferred to imino-diacetic acid (IDA) groups by the reaction with sodium salt of chloroacetic acid and the Cu^{2+} can be coupled to. The concentration of immobilized Cu^{2+} was analyzed with AAS and UV-vis spectroscopy (UV-vis).

RESULTS AND DISCUSSION

Morphology and size distribution of magnetic polystyrene microspheres

The PS seed particles were prepared by the soap-free emulsion polymerization. Figure 1 shows the morphology of the nonmagnetic PS seed particles, the particles had a smooth surface and uniform shape, the particle size distribution was calculated

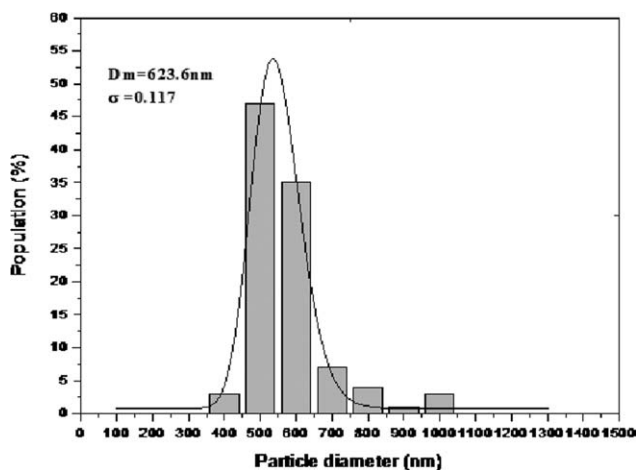


Figure 2 Particles size distribution of PS seed particles obtained from SEM photographs compared to the log-normal size distribution ($D_m = 623.6 \text{ nm}$, $\sigma = 0.117$).

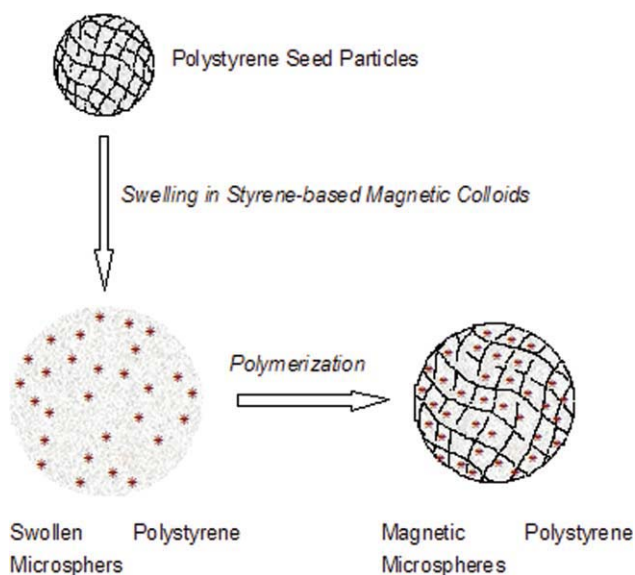


Figure 3 Illustration of procedures for preparing magnetic PS microspheres by magnetic colloid swelling polymerization. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com]

by statistically measuring the diameter of more than 100 individual particles from different regions of the SEM photographs, and the average-number diameter (D_n) of the particles was 623.5 nm with a standard deviation of 0.117 shown in Figure 2.

The magnetic PS microspheres were prepared by the method of magnetic colloid swelling polymerization. The schematic representation of the magnetic colloid swelling polymerization process was shown in Figure 3.

The PS seed particles are mainly made from styrene with DVB as crosslinker, and PS is prone to be swollen with nonpolar organic reagent while the crosslinker DVB is not. When the PS seed particles are swollen with styrene-based magnetic colloids containing Fe_3O_4 nanoparticles, the PS seed particles were more likely to absorb styrene-based magnetic colloids, which results in the expansion of the whole seed particles, while the crosslinker DVB restrains the whole seed particles from collapse. When equilibrium is achieved, the PS seed particles stop expansion, and Fe_3O_4 nanoparticles also penetrate into the seed particles in the course of swelling to form a swollen PS microsphere. After polymerization, magnetic PS microspheres are formed.

Figure 4(a,b) shows SEM photographs at different amplification of the magnetic PS microspheres prepared by the magnetic colloid swelling polymerization. The magnetic PS microspheres turned out to be uniform, spherical and had a smooth surface. The average-number diameter (D_n) was 1078 nm with a standard deviation of 0.192 shown in Figure 5.

The ratio of PS microsphere size after and before swelling was calculated and found to be 1.73, which

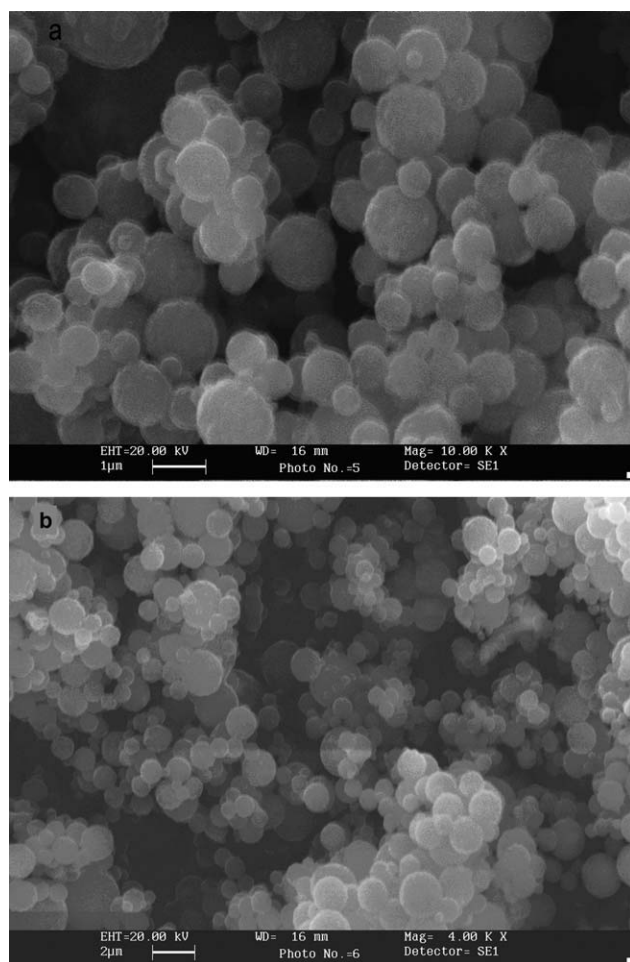


Figure 4 (a,b) SEM photographs at different amplification of the magnetic PS microspheres.

proved the fact that the PS seed particles was swollen with styrene-based magnetic colloid during swelling polymerization process. The PS seed particle grew to some extent and formed an independent

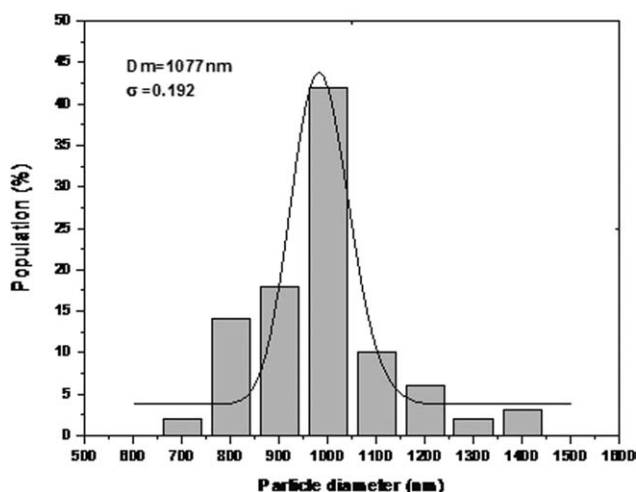


Figure 5 Size distribution of magnetic PS microspheres obtained from SEM compared to the log-normal size distribution.

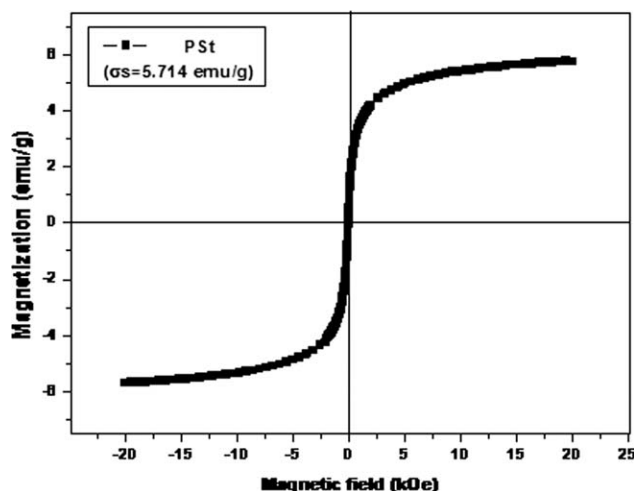


Figure 6 Magnetization curve of the magnetic PS microspheres obtained by VSM at room temperature.

microsphere after polymerization. The crosslinker of DVB is to pretest the seed particles from collapsing. In accordance with TEM image analysis in our previous work,²⁴ the magnetite nanoparticles is dispersed homogeneously throughout the polymer network of the magnetic PS microspheres.

Magnetic properties of the magnetic polystyrene microspheres

For ultrafine magnetic particles, there exists a critical size below which the particles are superparamagnetic.²⁵ It is reported the critical size of superparamagnetism for magnetite is about 25 nm. The average particle size of magnetite prepared in this study was 8 ± 0.5 nm, which has been characterized by TEM in our previous work,^{26,27} and has demonstrated the superparamagnetism by VSM.

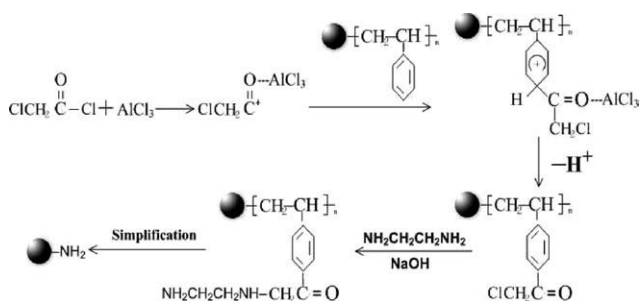
The magnetization curve of the magnetic PS microspheres at room temperature was recorded by VSM as shown in Figure 6. There is no hysteresis in the magnetization curve and both remanence and coercivity are zero. The results suggest the magnetic PS microspheres are typically superparamagnetic. The magnetite nanoparticles were incorporated throughout into the three-dimensional network structure of the PS microspheres, the superparamagnetism of magnetic PS microspheres results from superparamagnetic magnetite nanoparticles.

The saturation magnetization is strongly dependent on the content of magnetite in the magnetic PS microspheres, this can be controlled by adjusting the swelling degree of the polymer microspheres and the concentration of magnetite in the magnetic colloids. The saturation magnetization of the resulting magnetic PS microspheres was found to be 5.714 emu/g, representing a magnetite content of 7 wt % by using the reference value for the previously

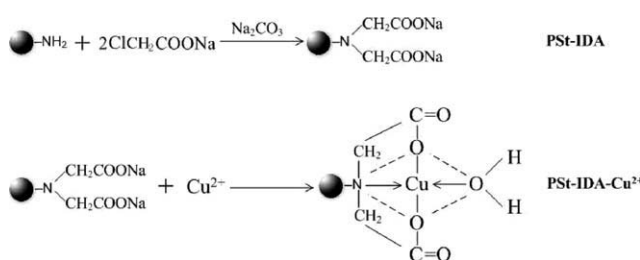
synthesized pure magnetite nanoparticles (81 emu/g). The magnetic PS microspheres could rapidly respond to a magnetic field and could readily be separated by decantation.

Surface functionalization reaction of magnetic polystyrene microspheres

In many cases, the magnetic PS microspheres with surface amino groups ($-\text{NH}_2$) are used as magnetic carriers in the field of enzyme immobilization,¹ protein separation,²⁻⁴ and so on. Proteins or enzymes can more easily be coupled to the amino groups ($-\text{NH}_2$) on the surface of magnetic PS microspheres by glutaraldehyde method,²⁷ and therefore, the amino groups ($-\text{NH}_2$) were introduced onto the surface of the magnetic PS microspheres by Friedel-Crafts acylation reaction.²¹ The schematic representation of the surface functionalization reaction was shown as:



Because it is very difficult to measure the concentration of the amino group on the surface of magnetic PS microspheres directly by instrumental analysis, the concentration of the amino group introduced onto the surface of the magnetic PS microspheres was quantitatively measured by chemistry reaction as is shown as:



The magnetic PS microspheres with amino groups were carboxymethylated by sodium salt of chloroacetic acid. Amino groups were transferred to IDA groups, where metal ions such as Cu^{2+} can be coupled to the surface of magnetic PS microspheres. The concentration of the amino group were calculated by the concentration of the Cu^{2+} immobilized on the magnetic PS microspheres, the

Cu^{2+} could be stripped easily by EDTA. In a series of experiments on surface functionalization reaction of magnetic PS microspheres under different conditions, the maximum concentration of amino group was found to be 0.168 mmol per gram determined by quantitatively by AAS and UV-vis, which was much higher than that of the traditional copolymerization method.²²

CONCLUSIONS

A new approach of seed swelling polymerization was introduced to the preparation of monodisperse magnetic PS microspheres. SEM observation and morphology analysis showed that the average diameter of the resultant magnetic PS microspheres was 1078 nm, with a standard deviation of 0.192. VSM results revealed the magnetic PS microspheres were superparamagnetic, and saturation magnetization was found to be 5.714 emu/g. Friedel-Crafts acylation reaction was used to introduce the amino groups on the surface of the magnetic PS microspheres. The amino groups existed on the surface of magnetic PS microspheres were confirmed by AAS and UV-vis, and the concentration of the amino groups was found to be 0.168 mmol per gram.

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