

One-step synthesis of reactant-product dual-templates imprinted capsule as phosphotriesterase mimetic enzyme for pesticide elimination

Yong Guo, Rui-yu Wang, Wen-hao Chi, Shuai Liu, Huguang Shi and Tian-Ying Guo *

The Key Laboratory of Functional Polymer Materials (Ministry of Education), Institute of Polymer Chemistry, College of Chemistry, Nankai University, Tianjin, 300071 (P. R. China)

tyguo@nankai.edu.cn

Supporting information

Materials.

Azodiisobutyronitrile (AIBN), divinylbenzene (DVB), vinyltriethoxysilane, ammonium hydroxide (24%), *p*-nitrophenol, HF solution (35%) were all purchased from Tianjin Chem-reagent Institute (China). Zinc dimethacrylate was purchased from Aldrich. Tetraethylorthosilicate (TEOS) was purchased from Beijing InnoChem Science and Technology Co., Ltd (China). γ -methacryloxypropyl trimethoxysilane (KH570) was purchased from Nanjing Shuguang (China). Paraoxon (purity>98%) was purchased from Shanghai Quandao Co. (China), DVB were distilled under vacuum before used. AIBN were recrystallized in ethanol before used. Other chemicals were analytical grade used as received.

Methods.

Synthesis of 400 nm v-SiO₂ seed: 360 ml water was mixed with 40 ml ethanol, then added 10 ml vinyltriethoxysilane in it, and the solution was stirred at room temperature for 2 h. Then 24 ml NH₃.H₂O (24%) was added slowly, kept it reacted 0.5 h. The silica seed was separated by centrifugation, washed five times with ethanol and water respectively, then dried in vacuum at room temperature for 24 h. Before used, the v-SiO₂ seed were treated with toluene at room temperature for 24 h.

Synthesis of 200 nm v-SiO₂ seed: Monodisperse pure silica seed was synthesized by a modified Stober method. The pure silica seed was then modified with KH570 to introduce polymerizable

double bonds. Briefly, 1 g of the silica seed was dispersed in 100 ml of ethanol, after adding 10 g of KH570, the mixture was allowed to react at 50 °C for 24 h with stirring under dry nitrogen. The silica seed was separated by centrifugation, washed five times with ethanol and water respectively, then dried in vacuum at room temperature for 24 h. Before used, the v-SiO₂ seed were treated with toluene at room temperature for 24 h.

Typical synthesis of silica/polymer core/shell microspheres: 0.0571 g p-nitrophenol and 0.0282 g paraoxon were dissolved in 200 ml acetonitrile, then 0.2412 g MAA-Zn was added slowly. After stirring gently for 2 h, 0.9348 g DVB, 0.1347 g AIBN and 0.168 g silica seed were added. This mixing solution was purged with nitrogen for 30 min while cooled in ice bath with stirring, then dispersed in an ultrasonic bath for 1 min. The polymerization was done at 80°C for 21 h. The result silica/polymer core/shell microspheres were separated from the mixed solution by centrifugation, and then washed with acetonitrile for 3 times and ethanol for 3 times and dried in vacuum overnight at 30°C. Then, 0.01g of the resulted microspheres were treated with 5/1(V/V) ethanol/ water containing 1.0M of NaOH (totally 30ml) for 15h to remove the template. The microspheres were cleaned by ethanol/water mixture (5/1 V/V). After removal the template, Zn²⁺ was also disappeared. To upload Zn²⁺, the resulted microspheres were treated with 4/1(V/V) ethanol/ water containing 0.2M of ZnCl₂ (totally 25ml) for 15h and then dried in vacuum overnight at 30°C.

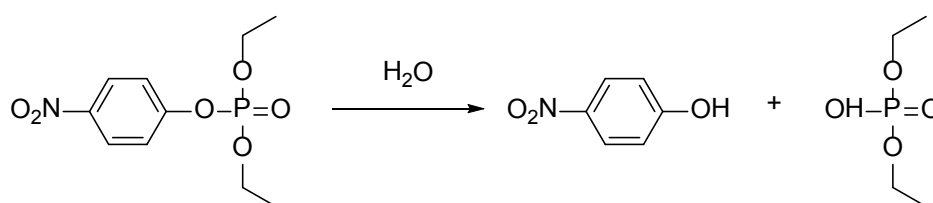
Typical preparation of hollow microspheres: 0.1 g of the obtained core/shell microspheres were dispersed in 25 ml ethanol, then 5 ml HF solution(totally 2 wt%) was added, after stirring gently for 7h, the products were separated from the solution by centrifugation, and then washed with ethanol/water mixture (5/1 V/V) for 3 times. Then the removal of template and upload of Zn²⁺ was the same as the core/shell microspheres.

Characterization.

The morphology of microspheres were observed by transmission electron microscopy (TEM, Tecnai G2 F20)

Evaluation of hydrolytic activity of microspheres: The activity of capsule was assayed using 1 mM of paraoxon as substrate. The hydrolysis activity of capsule was measured spectrophotometrically by monitoring the decrease of paraoxon. An certain amount of capsules were dispersed in 363μL ethanol with the help of ultrasound, then 4.5ml (20 mM) Tris-HCl buffer

(pH 9.0) were added. The dispersion system under a constant temperature 30°C for 10 min and 137μL paraoxon in ethanol was added. Then, the activity of the capsules was determined in batch mode using magnetic agitation at 30°C. An amount of 120 μL of the reaction solution was sampled and centrifuged (9000 rpm for 5 min), then 60μL of the supernatant was diluted with 240μL of 20 mM Tris-HCl buffer (pH 9.0) and the absorbance at 275 nm was determined using UV spectrophotometry.



Scheme S1. illustration of paraoxon hydrolysis reaction.

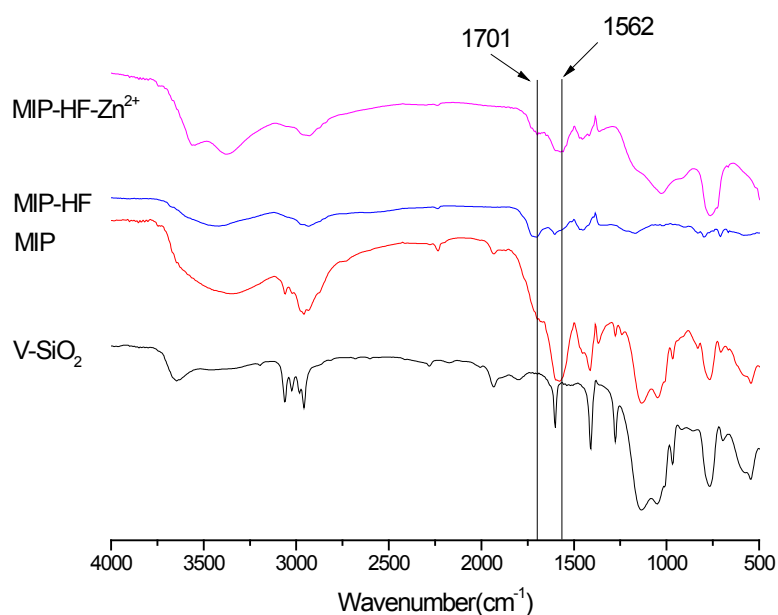


Figure S1 The FTIR spectra of v-SiO₂, MIP, MIP-HF and MIP-HF-Zn²⁺.

As shown in Figure S1, the peak at 1562 corresponds to the asymmetric vibration of carboxylate group, the peak at 1701 corresponds to the protonated carboxylic group. Compared MIP-HF to MIP, 1701 appeared and 1562 almost disappeared, which means that carboxylate was transferred into protonated carboxylic group. So as shown in experimental section, after treated with HF, we did experiment for uploading the Zn²⁺, as shown in Figure below, after upload the

Zn²⁺, the peak at 1701 almost disappeared and the peak at 1562 reappeared. [1, 2]

- [1] Zhou C L, Zhang Z Q, Jiang M. Studies on ionomers and the related blends I. Compatibilization by introducing like ions into component polymers. *Acta Polymerica Sinica*, 1992, 1 (4): 463-471.
- [2] Wang J-Y, Chen W, Liu A-H, *et al.* Controlled Fabrication of Cross-Linked Nanoparticles/Polymer Composite Thin Films through the Combined Use of Surface-Initiated Atom Transfer Radical Polymerization and Gas/Solid Reaction. *J. Am. Chem. Soc.*, 2002, 124 (45): 13358-13359.