PREPARATION AND EVALUATION OF POLY(PENTAERYTHRITOL TRIACRYLATE-CO-TRIALLYL ISOCYANURATE) MONOLITHIC COLUMN FOR SMALL MOLECULES SEPARATION

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Abstract

A monolithic stationary phase was prepared for liquid chromatography by in situ copolymerization of pentaerythritol triacrylate (PETA) and triallyl isocyanurate (TAIC). The morphology of monolithic column and pressure drop across the column were characterized. The effect of the porogen on the pore structure was characterized by SEM. Under the comparison condition, the monolith was successfully prepared and its mechanical strength had been

studied. Furthermore, the poly (PETA-co-TAIC) monolithic column was applied to the separation of aromatic compounds. These successful application demonstrated that the purposed monolith was promising for HPLC separation of small molecules.

1. Introduction

In the recent years, the monolith was used as a novel separation media, the techniques and methods of its preparation became to be mature [1-5]. According to the type of material, the monolith could be divided into organic monolith and silica-based monolith [6-9]. The organic monolith was widely used in liquid chromatography because they had many advantages, such as good permeability, easily preparation, and low pressure [10-14]. The polymer monolithic column possessed a whole network skeleton with strong rigidity and low back pressure, which could be carried on a fast and efficient analysis at higher flow rate. While with the development of science and technology, there have been many new monomers and crosslinker, such as epoxy resin, polyhedral oligomeric silseaquioxanes, trimethylpropane triacrylate (TMPTA), pentaerythritol triacrylate (PETA), and triallyl isocyanurate (TAIC) etc. PETA was often used as a crosslinker because of its more double bonds, which could increased the number of crosslinking points and the density of the network. Recently, Huang et al. [15] synthesized the polymeric monolith by copolymerization of 4-vinylphenylboronic (VPBA) and pentaerythritol triacrylate (PETA) for capillary liquid chromatography (CLC). Xie's group [16] had prepared a series of hydrophilic interaction/cation-exchange polymeric monolithic stationary phase by copolymerization of PETA with various ionizable monomers. The results demonstrated that these monoliths had high column efficiency, high selectivity, and high permeability.

In this paper, a novel poly(PETA-co-TAIC) monolith which was prepared by a single polymerization step, was developed as a separation column in HPLC. The influence of the ratio of TAIC to PETA, as well as the composition of the porogenic solvent on the morphology, permeability and selectivity of the monolith was evaluated. The designed polymeric media was applicated to separate a series of typical low-molecular-weight organic compounds.

2. Experimental

2.1. Materials

PETA and TAIC were purchased from Energy Chemical Technology Co. Ltd. (Shanghai, China). 2,2'-azobisisobutyronitrile (AIBN) was obtained from Shanghai Chemical Plant (Shanghai, China) and purified before use. Cetylalcohol was provided by Tianjin Guangfu Fine Chemical Research Institute (Tianjin, China). Methanol was purchased from Tianjin Kemiou Com (Tianjin, China). Ultrapure water was prepared from a Millipore-Q water-purification system (Taiwan, China) and solutions were filtered through a 0.45um membrane before use throughout.

2.2. Synthesis of monolith

The polymerization mixture for the monolithic column was prepared as follows: A mixture containing PETA, TAIC, methanol, cetylalcohol, and AIBN was ultrasonicated for 10 min to get a homogeneous suspension. Then the stainless-steel column (50mm × 4.6mm i.d.) sealed at the bottom was filled with the polymerization mixture and then sealed at the top. The column was submerged into water bath at 60°C for 24h. After polymerization was complete, the monolithic column was connected to HPLC pump and extensively washed with methanol to remove the unreacted reagents and porogenic solvents. The polymerization reaction equation was shown in Figure 1.
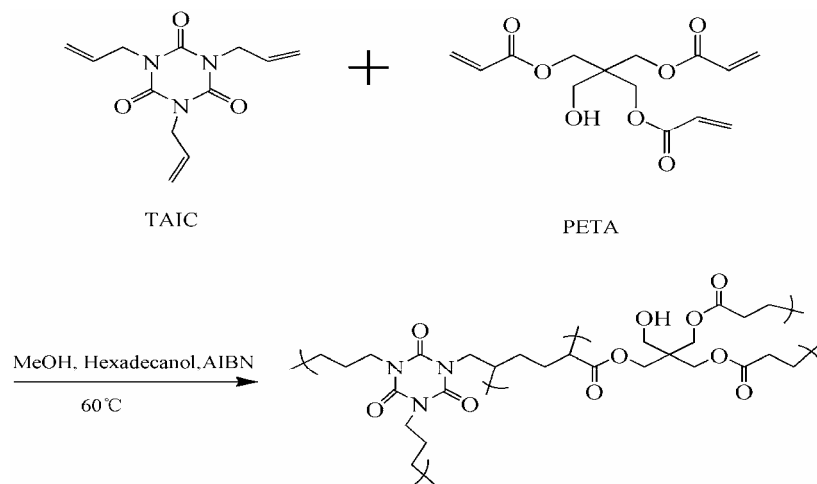

Figure 1. Synthesis scheme of the poly (TAIC-co-PETA) monolithic column.

2.3. Characterization of the monolithic column

The prepared monolith was cut into little pieces that were dried in vacuo at 60ºC for 24h. The morphological properties of the monolith were photographed using scanning electron microscope (SEM) by Hitachi (Hitachi High Technologies, Tokyo, Japan) S-4300 SEM instrument. Chemical group of the monolith was detected by Fourier transform infrared spectroscopy (FT-IR) (Shimadzu, Kyoto, Japan) instrument. The pressure drop across the column at different flow rate was also analyzed.

2.4. HPLC analysis

A HPLC system (Beijing Heng Tong Innovation) equipped with P-3000 pumps and a UV-3000 detector was carried out in all the chromatographic experiments. The observed data were processed by CXTH-3000 chromatography software. The flow rate was 1mL/min and injection volume was 2uL. The elution was monitored at 254nm.

3. Results and Discussions

3.1. The structural characterization of the monolith

Monomer, porogen, and crosslinker played important roles in the preparation of monolith. They had significant effects not only on the rigidity and porosity, but also on the selectivity and column efficiency of the monolith. To get the best state of the monolith, the preparation conditions have been optimized, such as the types and the amounts of monomer, crosslinker, solvent, and porogen. A series of monoliths were prepared with different compositions of the polymerization mixture, which were described in Table 1. The SEM images of the cross-section of prepared monoliths were shown in Figure 2. Firstly, when the concentration of TAIC or PETA increased (column B and C), the pore structure became more dense which led to high back pressure and detached (Figure 2(B) and 2(C)). In addition, analytes were not retained on column (column C in Figure 3). Secondly, when the proportion of TAIC

or PETA decreased (column E and G), the pore structure became loose (Figure 4(E) and 4(G)). Moreover, the selectivity and column efficiency of the column were poor (Figure 3). When the amount of methanol increased (column D) or decreased (column A), the polymer was slack (Figure 2(D)) or dense (Figure 2(A)). By comparison, column F had better pore structure which caused low back pressure and good permeability. Moreover, better peak shape could be obtained on column F (Figure 3). So column F was used for further experiment.

Table 1. Optimization of compositions for preparation of the monolithic column

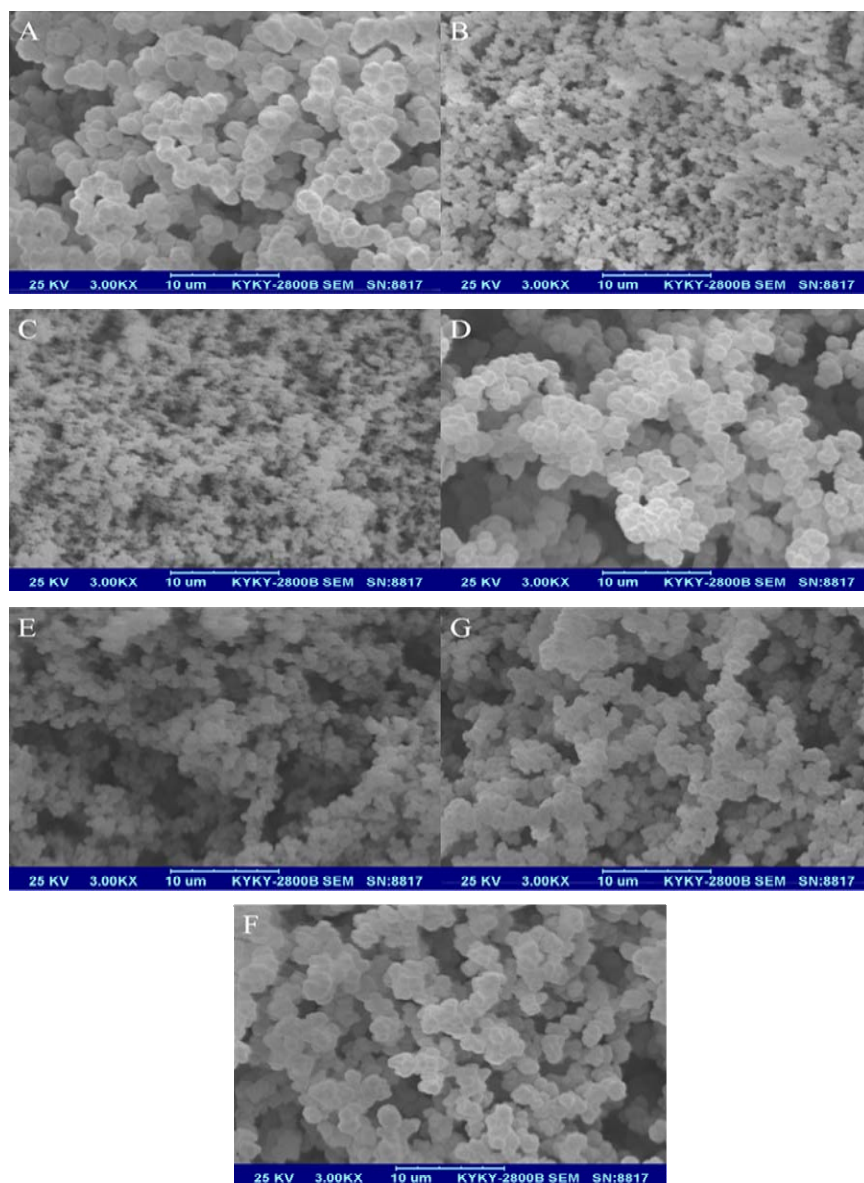| Column	PETA (mL)	TAIC (mL)	MeOH (mL)	Hexadecanol (g)	AIBN (g)	Mechanical
A	0.3	0.3	0.8	0.5076	0.0051	Hard
B	0.4	0.3	1.0	0.5080	0.0050	So hard
C	0.3	0.4	1.0	0.5078	0.0048	So hard
D	0.3	0.3	1.2	0.5082	0.0053	Slack, flexible
E	0.2	0.3	1.0	0.5084	0.0051	Slack, flexible
F	0.3	0.3	1.0	0.5080	0.0049	Hard
G	0.3	0.2	1.0	0.5079	0.0052	Slack, flexible

Figure 2. Scanning electron micrographs of the poly (TAIC-co-PETA) monolith.
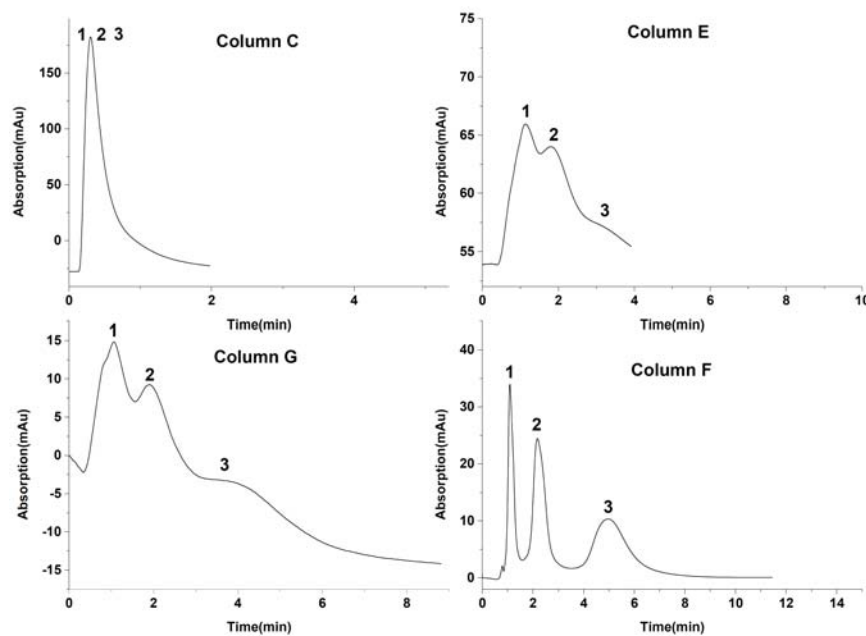
Figure 3. Chromatograms for separation of alkkylbenzenes on poly (TAIC-co-PETA) monolith benzene 1; diphenyl 2; phenanthrene 3. Mobile phase: 75% methanol. Flow rate: 1.0mL/min. Detection wavelength: 254nm.

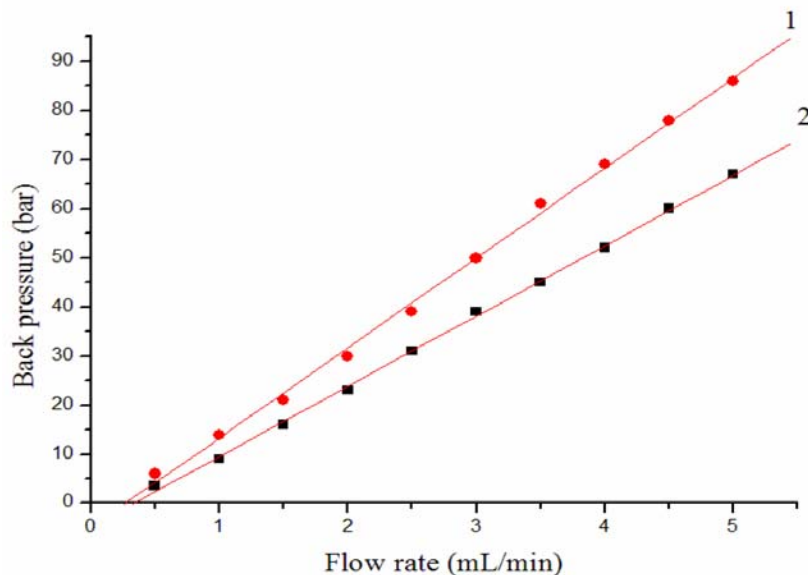**3.2. Permeability and mechanical stability of the monolith**

Figure 4. Back pressures of the poly (TAIC-co-PETA) monoliths at different flow rates. Mobile phase: (1) water and (2) methanol.

3.3. IR spectrum of monolith

The IR spectrum of monolith was shown in Figure 5. The spectra at 2965cm^{-1} and 2872cm^{-1} was caused by C-H symmetrical stretching vibration and asymmetrical stretching vibration. The broad bands at $3420\text{-}3432\text{cm}^{-1}$ was due to O-H. The absorption observed at 1678cm^{-1} was caused by C=O amide band I. The stretching band at 1731cm^{-1} was due to C=O ester. The band at 1152cm^{-1} was due to C-O-C.
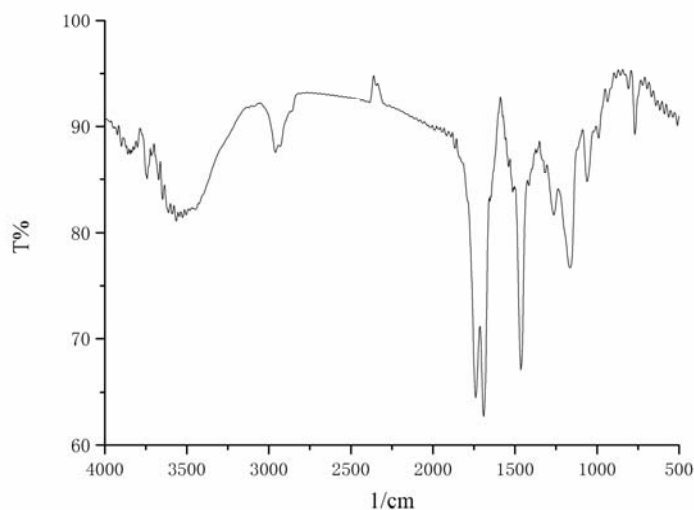
Figure 5. The FT-IR spectrum of the poly (TAIC-co-PETA) monolith.

3.4. Application of poly (PETA-co-TAIC) monolithic column

The poly (PETA-co-TAIC) was applied to the separation of aromatic compounds. Figure 6 represented the baseline separation of three aromatic compounds within 6 min. The compounds were eluted in the order of benzenes, biphenyl, phenanthrene (Figure 6(a)) and aniline, diphenylamine, anthracene (Figure 6(b)) according to their hydrophobicity which confirmed a typical reversed-phase separation mechanism.
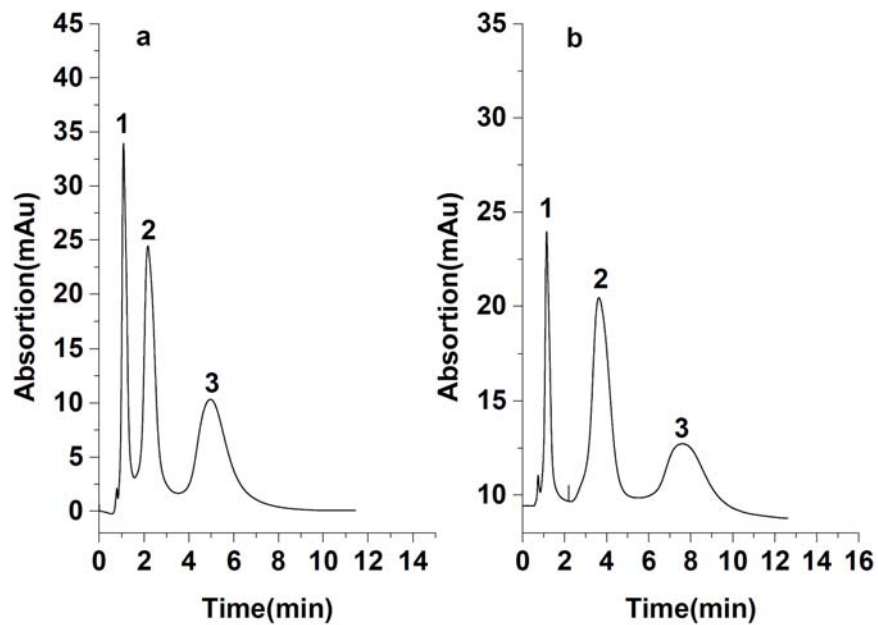
Figure 6. Chromatograms for separation of alkkylbenzenes on poly (TAIC-co-PETA) monolith. (a): benzene 1; diphenyl 2; phenanthrene 3; (b): aniline 1; diphenylamine 2; anthracene 3. Mobile phases: (a) 75% methanol; (b) 80% methanol. Flow rate: 1.0mL/min. Detection wavelength: 254nm.
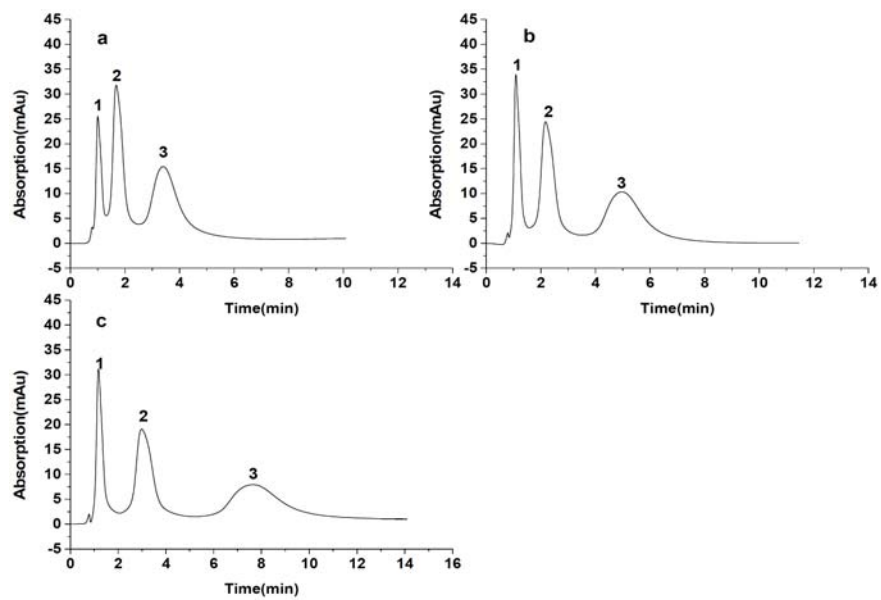
Figure 7. Chromatograms for separation of alkkylbenzenes on poly (TAIC-co-PETA) monolith benzene 1; diphenyl 2; phenanthrene 3. Mobile phases: (a) 80% methanol; (b) 75% methanol; (c) 70% methanol. Flow rate: 1.0mL/min. Detection wavelength: 254nm.
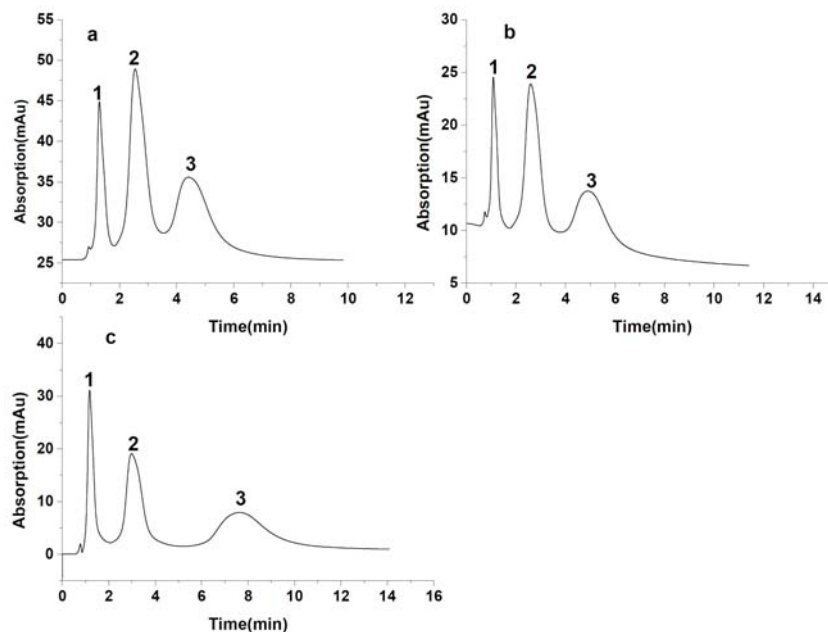
Figure 8. Chromatograms for separation of alkkylbenzenes on poly (TAIC-co-PETA) monolith aniline 1; diphenylamine 2; anthracene 3. Mobile phases: (a) 85% methanol; (b) 80% methanol; (c) 75% methanol. Flow rate: 1.0mL/min. Detection wavelength: 254nm.

4. Conclusion

In this paper, a novel porous poly (PETA-co-TAIC) monolithic column was prepared by in situ free-radical polymerization. The morphology characterized by SEM showed a well-ordered skeleton structure. The synthetic method was simple, convenient, and time-saving. The prepared poly (PETA-co-TAIC) monolith exhibited good stability and successful separation capacity for the aromatic compounds.

Acknowledgement

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