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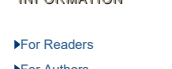


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CHARACTERIZATION OF ORGANIC POLYMER MONOLITH COLUMNS CONTAINING AMMONIUM QUARTERNARY AS INITIAL STUDY FOR CAPILLARY CHROMATOGRAPHY

Aster Rahayu^{*}, Siti Jamilatun^{*}, Joni Aldilla Fajri^{**}, Lee Wah Lim^{***}

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Abstract: The polymerization process with a simple step has become the centre of attention of several researchers. Various polymers have been developed, although in general, they use polymerization with a post-modification method. A quaternary ammonium monolith organic polymer has been prepared using a simple single thermal method in this research. 2-(Methacryloyloxy)-N,N,N-trimethylethanaminium chloride was as the monomer, and ethylene dimethacrylate was as crosslinker. The polymerization proceeded in fused-silica capillary (100 mm, 0.32 mm i.d. x 0.45 mm o.d.) using a one-pot approach method. To achieve the perfect macropores, isopropyl alcohol, PEG 400, and ethanol were used as porogen. Characterization of the surface morphology was carried out using a Scanning Electron Microscope (SEM), and the existence of an amine group was characterized by Fourier Transform Infrared Spectroscopy (FTIR). The distribution size of pores in the polymer was in the range of 1.29 to 3.33 μm .

Keywords: Monolith, Organic polymer, One-pot approach, Ammonium quarternary, Polymerization, Capillary

Abstrak: Polimerisasi dengan proses yang sederhana dan simpel menjadi pusat perhatian beberapa peneliti. Berbagai macam polimer telah dikembangkan, akan tetapi pada umumnya menggunakan polimerisasi dengan metode *post-modification*. Pada penelitian ini, polimer organik yang mengandung amonium kuartener dalam bentuk monolit dengan polimerisasi yang menggunakan suhu tunggal dan sederhana telah dilakukan. 2-(Methacryloyloxy)-N,N,N-trimethylethanaminium chloride digunakan sebagai monomer dan ethylene dimethacrylate sebagai crosslinker. Polimerisasi dilakukan dengan metode *one-pot approach* di dalam kapiler silika (100 mm, 0,32 mm i.d. x 0,45 mm o.d.). Untuk mendapatkan makropori yang sempurna, isopropil alkohol, PEG 400 dan etanol digunakan sebagai porogen. Karakterisasi morfologi permukaan dilakukan dengan menggunakan *Scanning Electron Microscope* (SEM), dan *Fourier Transform Infrared Spectroscopy* (FTIR) untuk mengidentifikasi gugus fungsi gugus amina yang terdapat pada polimer. Ukuran distribusi pori pada polimer berkisar antara 1,29 sampai 3,33 μm .

Kata kunci: Monolitik, Polimer organik, *One-pot approach*, Amonium kuartener, Polimerisasi, kapiler

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Introduction

Polymers are large molecules composed of a series of monomers that are interconnected by the presence of crosslinkers. Monolith polymers can be broadly divided into two types, namely organic polymers and inorganic polymers (Rahayu *et al.*, 2015). Inorganic polymers are commonly based on silica and are widely used as a stationary phase. Several kinds of research have been reported in terms of application of inorganic polymers, such as separation and detection of cation levels in drinking water (Rahayu *et al.*, 2015), protein (Zhao *et al.*, 2012), tryptic digests of bovine serum albumin (BSA) (Zhang *et al.*, 2011), levodopa, carbidopa, benserazide, dopamine, and 3-O-methyldopa in plasma samples (Grecco *et al.*, 2020), polar compounds (Yang *et al.*, 2013) and other macromolecular compounds (Iwasaki *et al.*, 2012). However, time-consuming manufacturing and a limited variety of polymer-forming materials were obtained as disadvantages of inorganic polymer (Zhong *et al.*, 2010, Lalli *et al.*, 2020).

Organic polymers have been developed and applied as a stationary phase for separating compounds or ions in the chromatography system. The existence of pore in the monolith polymer can be served as an adsorbent. Some polymers have been widely used in the adsorption of several heavy metals and organic compounds. The Cr (VI) ion could be adsorbed using a magnetic polymer containing an amine group (Zhao *et al.*, 2013, Zhao *et al.*, 2010), removal of Pb²⁺, Cu²⁺, and Zn²⁺ (Mokadem *et al.*, 2020), trace Cd (II) in biological samples (Chen *et al.*, 2020), and removal of Pb(II) from water medium (Naushad *et al.*, 2020). Methacrylate-based polymers with the epoxy ring-opening method have been successfully prepared and applied for separation of organic compounds such as Bisphenol A (BPA) (Zhao *et al.*, 2013), Bovine Serum Albumin (BSA) (Shamim *et al.*, 2007), alkylbenzene (Wang *et al.*, 2014), pentachlorophenol (Pan *et al.*, 2014), and enantioseparation (Angga *et al.*, 2019). Furthermore, it is also applied for the adsorption of some ions, such as Cu (II) (Shen *et al.*, 2012) and Hg (II) ions (Pan *et al.*, 2014, Pan *et al.*, 2012). Overall, many of these polymers are applied as the stationary phase in the chromatography system.

Polymerization uses two-stage processes, namely the polymerization stage and the advanced modification stage. This advanced modification stage aims to add the desired active side group according to the intended purpose and application. However, a few organic polymers are synthesized using a one-pot approach method. This method was taking the attention of several researchers due to the simple step of the polymerization process.

The synthesizing of monolithic polymers with quaternary ammonium groups using a straightforward one-pot approach as a stationary phase in capillary system liquid chromatography will be the objective of this research. The monolithic polymers containing quaternary amine groups will be polymerized from 2-(Methacryloyloxy)-N,N,N-trimethylethanaminium chloride (META) and ethylene dimethacrylate (EDMA) with a short, simple and straightforward method, namely one-pot approach. The morphology surface of the polymer will be characterized before applying as the stationary phase in the capillary system liquid chromatography to separate organic compounds.

Material And Methods

Materials

The materials are purchased from a different source. 2-(Methacryloyloxy)-N,N,N-trimethylethanaminium chloride (META), Ethylene dimethacrylate (EDMA), Isopropyl alcohol (IPA), ethanol were purchased from Wako 1st Grade, Japan, 2,2'-azobisisobutyronitrile (AIBN) was purchased from Trade TCI Mark, Japan, poly(ethylene oxide) Mn. 400 (PEG 400), 3-(trimethoxysilyl)-propyl methacrylate (γ -MAPS), uracil, toluene, 0.1 M NaOH, 0.1 M HCl were purchased from Nacalai Tesque, Kyoto, Japan, deionized water (water that has a known conductivity and is specifically used for ion chromatography) from GS-590 water distillation system (Advantec, Tokyo, Japan).

Apparatus

The surface morphology of quaternary ammonium organic polymer monoliths was characterized using Scanning Electron Microscope (SEM) S-4800 (Hitachi, Tokyo, Japan) mainly, the size of single unions macropores formed. Furthermore, identifying the functional group in the polymer was carried out using Fourier Transform Infrared (FTIR) Spectrum 400 Series (Perkin Elmer).

The chromatographic system was performed to separate uracil and toluene using a capillary LC system constructed by an L.TEX-8301 Micro Feeder (L.TEX Corporation, Tokyo, Japan) equipped with an MS-GAN 050 gas-tight syringe 0.5 mL (Ito, Fuji, Japan) as a pump, a Model 7520 (Rheodyne, Cotati, CA, USA) injector with an injection volume of 0.2 μ L, a 100 mm x 0.32 mm i.d. of microcolumn and a UV-1575 intelligent UV/Vis detector, (JASCO, Tokyo, Japan) that was operated at 254 nm. The data was acquired using a data processor CDS-Lite ver.5.0 (LA soft, Chiba, Japan).

Synthesis of quaternary ammonium organic polymer monoliths

Synthesize of organic polymer monoliths was started with pre-treatment of the inner wall of capillary silica. The capillary column was rinsed with 0.1 M NaOH, 0.1 M water, 0.1 HCL and flowed with acetone into the column flow rate of 2.5 μ L/min for 60 minutes sequentially. The column was dried by flowing

nitrogen gas for 30 minutes. 30% (γ -MAPS) in acetone filled into the column, and then both column ends sealed. The anchor inner wall of the capillary column is allowed at 60 °C for 24 hours in the water bath. The capillary column was rinsed with acetone at a flow rate of 2.5 μ L/min for 60 minutes and dried with nitrogen gas for 30 minutes.

Initially, the polymerization stage was carried out by preparing a mixture solution according to previous research with major modification (Rahayu *et al.*, 2015); 1.25 mL of META, 0.375 mL of EDMA, 1.75 mL of isopropyl alcohol, 0.35 mL of ethanol, 1.4 mL of PEG 400, and 2 mg 2,2'-azobisisobutyronitrile. The mixture solution was homogenized before filling into the pretreated capillary column. Subsequently, the polymerization was allowed at 60 °C for 24 hours in the water bath. The obtained polymer was rinsed with methanol to remove the residual reaction. Monolith polymers were applied in capillary system liquid chromatography for *the* separation of uracil and toluene.

Discussion

Synthesis of polymeric organic monoliths

The organic polymer monolith containing quaternary ammonium was synthesized directly in the capillary column (100 mm, 0.32 mm i.d. x 0.45 mm o.d.) with a single thermal polymerization one-pot approach method. The synthesis was carried out using a monomer containing a quaternary ammonium functional group. Quaternary ammonium groups in monomers act as strong anion exchangers and work well to separate either anion or organic compounds. The scheme of the expected reaction polymerization one-pot approach method is shown in Figure 1. The functional monomer introduced the quaternary ammonium in the organic polymer, which quaternary ammonium is the active group for the anion exchange.

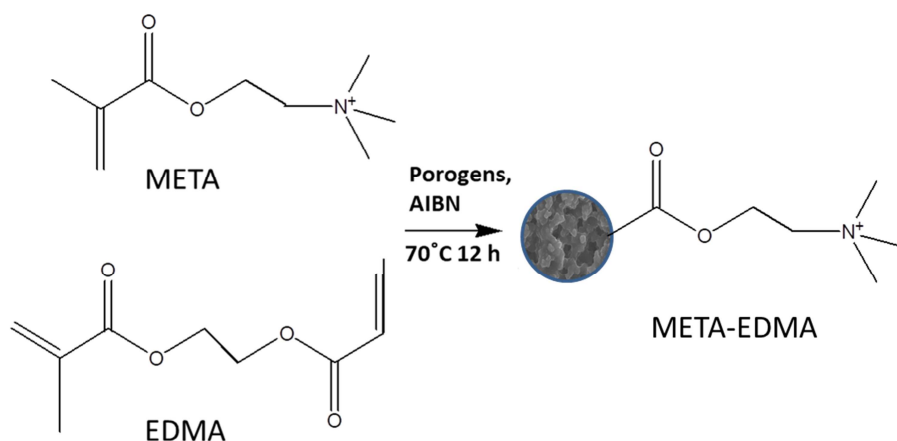


Figure 1. Scheme of the expected reaction of organic polymer

In the polymerization of organic polymer monolith, the ratio between monomer and porogen ratio was 20:80. Meanwhile, 20% was a mixture of monomer and crosslinker, and 80% was porogen. Interm of 20% for monomer and crosslinker, 50% of monomer, and 50% crosslinker were used. Three organic compounds acted as porogen, such as Isopropyl alcohol, polyethylene glycol 400, and ethanol. These three solutions worked to form macropores on the surface. Visually the polymer formed was solid white, as shown in Figure 2.



Figure 2. The visual organic polymer contained quaternary ammonium.

Characterization of organic polymer monoliths Using Fourier Transform Infrared (FTIR)

Fourier Transform Infrared (FTIR) was carried out to determine the presence of amine compounds. The amine group itself was used as a strong anion-exchanger. The FTIR spectrum of organic polymer monolith was shown in Figure 3. In Figure 3, it can be seen that detected five peaks in the organic polymer monolith column. These five peaks were the first peak at wave number 1020.3 cm^{-1} , the second peak at wave number 1352.2 cm^{-1} , the third peak at wavenumber 1723.6 cm^{-1} , the fourth at wave number 2875.3 cm^{-1} , and the fifth peak at wavenumber 3433.6 cm^{-1} . The absorption area in the wavenumber between $1150\text{-}1085\text{ cm}^{-1}$ refers to the C-O (stretching) absorption area. The high intensity of the principal peak in organic polymer monolith describes many oxygen-containing groups after the polymerization process. The number of oxygen-containing groups came from either ethylene glycol dimetharyate or 2-[(methacryloyloxy) ethyl] trimethylammonium. The absorption area in the wavenumber between $1250\text{-}1020$

cm⁻¹ refers to C-N (amine) absorption area. Subsequently, the absorption area in the wavenumber around 1650-1580 cm⁻¹ refers to N-H (amine) absorption area. These amine groups are contributed by functional monomers that are used. The absorption area in the wavenumber between 2853-2962 cm⁻¹ refers to the C-H (stretching) absorption area. Then, in the wavenumber between 3000-3600 cm⁻¹, it relates to O-H functional group (Silverstein *et al.*, 2005). It can be concluded that existing bonds and functional groups are thought to come from the presence of a polymerization reaction involving an initiator, monomer, crosslinker, and porogen.

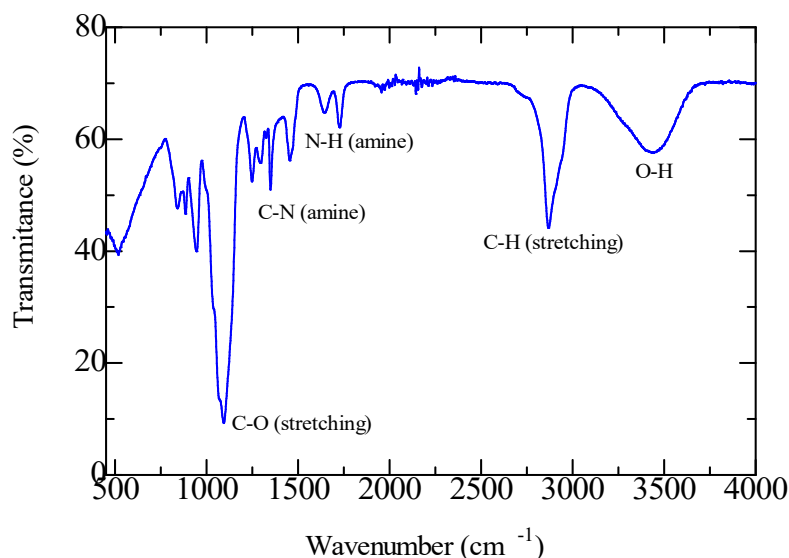
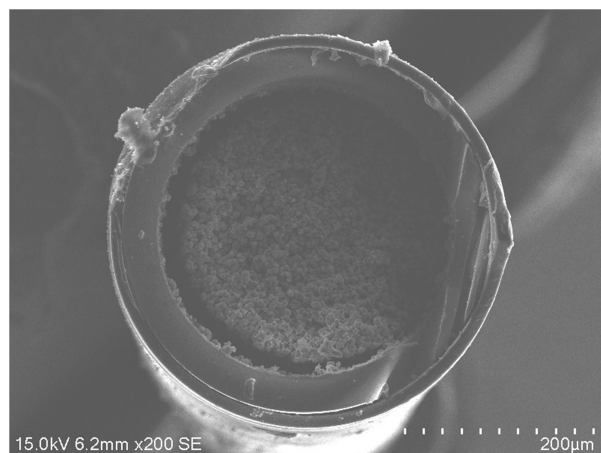


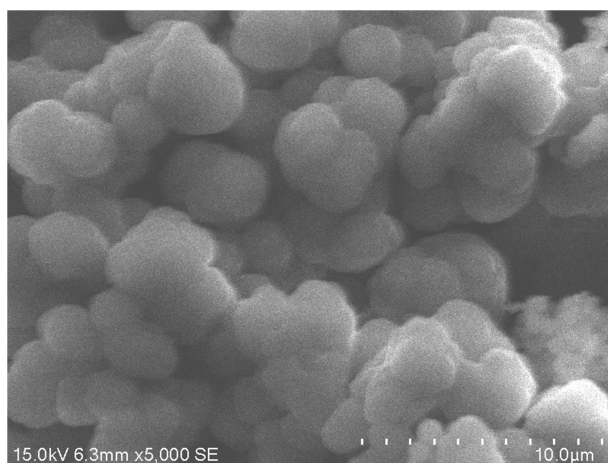
Figure 3. Fourier Transform Infrared (FTIR) spectrum of organic polymer monoliths.

Scanning Electron Microscope (SEM)

The morphology of the organic polymer monolith is one of the essential parameters to determine the adhesion condition of the polymer with an inner wall of the column. Furthermore, the range distribution of pores formed by using porogen in polymer solutions also can be observed. The porogen-created macropores as the distance between one polymer union and another. The morphology photo of the organic polymer monolith can be seen in Figure 4.



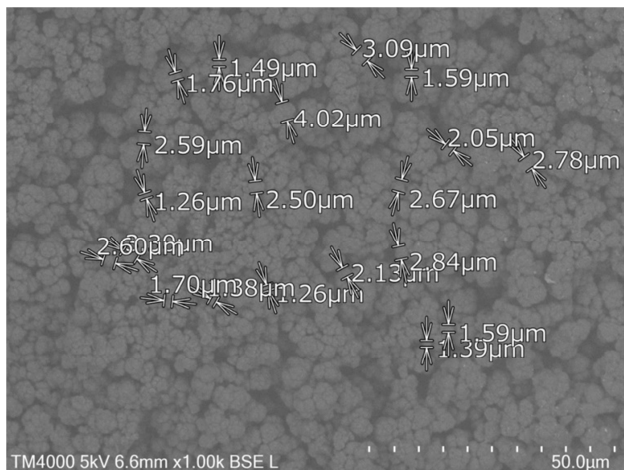
(a)



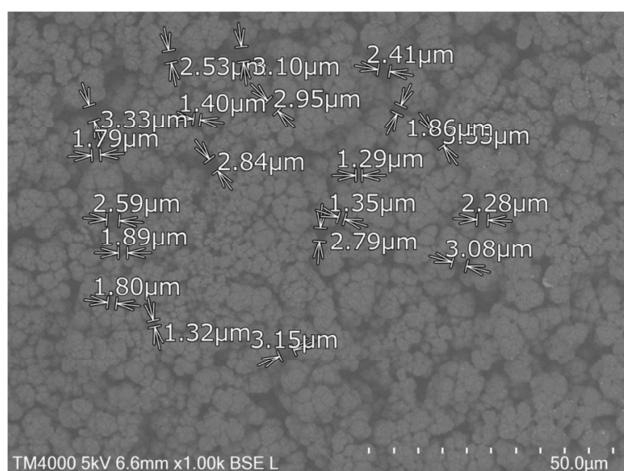
(b)

Figure 4. The morphology of the organic polymer monolith. (a). The magnification of 200x. (b). The magnification of 5000x

Figure 4 shows the performance of the organic polymer attaching into the inner wall of the capillary column by enlarging 200 and 5000 times. In Figure 4.(a). there is a sufficient optimal adhesion between the inner wall surface of the capillary and the organic polymer monoliths. The pre-treatment process in the capillary column using γ -MAPS dramatically affected the adhesion results for attaching the polymer to the inner wall of the capillary. The surface capillary column coated by the silanol group was perfectly laxative by γ -MAPS. Furthermore, Figure 4.(b). shows the distribution of unions and macropores in the organic polymer monolith. The macropores were significantly formed and evenly distributed. Subsequently, the size distribution of the union and macropores are shown in Figure 5. The range size for the union and macropore is 1.26-3.09 μm and 1.29-3.33 μm , respectively.



(a)



(b)

Figure 5. The morphology of organic polymer monolith. (a). Distribution size of the union. (b). Distribution size of macropore

According to the distribution size union and macropore range, the organic polymer could be applied as a stationary phase for the capillary chromatography system. It is promoting good results with a reasonably good system pressure below 2 MPa.

Separation of Uracil and Toluene

The separation of organic compounds was conducted to observe the ability of organic polymer monolith as the stationary phase in the liquid chromatography capillary system. Uracil and toluene were used as analytes for the initial study. The separation of uracil and toluene by the liquid chromatography capillary system is shown in Figure 6. It can be seen that the toluene and uracil could be appropriately separated. The retention time of toluene and uracil are 1.9 minutes

and 2.4 minutes, respectively. Toluene was eluted first and then continued with uracil when 90% acetonitrile was used as eluent. These show that the organic polymer monolith column could work in non-polar compounds by reverse-phase mode. This result is promising for applying organic polymer monolith ammonium quaternary as a stationary phase in the liquid chromatography capillary system.

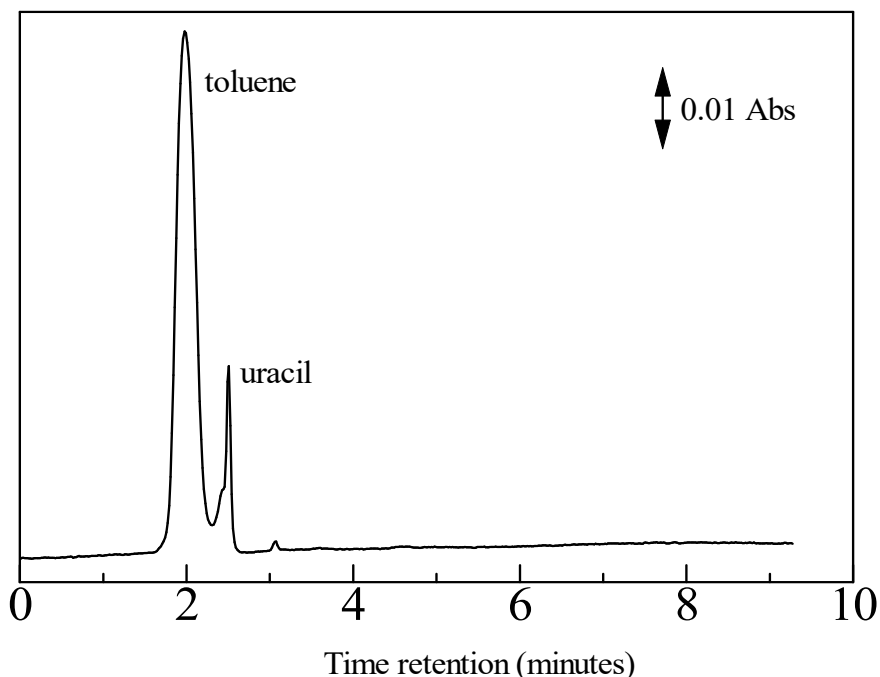


Figure 6. Separation of uracil and toluene by liquid chromatography capillary system. Experimental conditions: column: organic polymer containing ammonium quaternary monolithic (100 mm, 0.32 mm i.d. x 0.45 mm o.d.); mobile phase: 90% acetonitrile; flow rate: 3 μ l/min; detection wavelength: 254 nm, injection volume: 0.02 μ l.

Conclusions

An organic polymer monolith containing quaternary ammonium was successfully synthesized in the capillary column using a one-pot approach method. 2-(Methacryloyloxy)-N,N,N-trimethylethanaminium chloride acted as a monomer, Ethylene dimethacrylate as a crosslinker, and three kinds of solution as porogen; isopropyl alcohol, PEG 400, and ethanol. The organic polymer monolith also contained an amine functional group (C-N) derived from the quaternary ammonium active group for separating uracil and toluene. The range distribution size of the union and macropore is 1.26-3.09 μ m and 1.29-3.33 μ m, respectively.

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