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Organic monolithic materials: preparation and applications

KEYWORDS: Monolithic materials; polymer-based monoliths; characterisation; solid phase extraction.

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ABSTRACT *Monolithic materials have been shown to possessmany advantages compared with packed particles. For example, they are easy to prepare, they have high surface area and high stability, and they are highly permeable to liquid flow compared with the packed bed since porous monoliths contain interconnected macro- and mesopores. Theporous polymer-based monolithshave been fabricated by using a wide range of monomers and crosslinking agents enabling the porous properties of the monolith to be controlled. In the recent years, they have found widespread use in many different applications such as liquid chromatography, solid phase extraction, and enzyme immobilisation in capillary and microfluidic chip formats. This review focuses on the various studies outline routes for fabrication of the organic monolithic materials. Moreover, this review focuses on the applications of polymer-based monoliths in solid phase extraction.*

1.1.Definition of the monolithic materials

Monolithic materials were introduced by Hjerten*et al*.[1] when compressed soft gels, called "continuousbeds" were developed in 1989 and utilised in chromatographic separation. A further innovation bySvec and Frechet in the early 1990s was fabrication of rigid macroporous polymer monoliths, which were fabricated by a very simple "moulding" process and used as high performance liquid chromatography separation media[2, 3]. Due to their unique properties, these organic monolithic materials have been used in a variety of applications[4]. Later, inorganic silica-based monolith was fabricated by several groups starting in 1996[5, 6].

The word "monoliths" is from the Greek, "mono", which means 'one', and "lithos", which means 'stone'. It defines a geological or technological feature such as a mountain or boulder, consisting of a single massive rock; for example, a rock, which was collected by a famous Chinese Empress and used to decorate the access to one of her palaces,[7] as shown in Figure 1.

Fig. 1 Photograph of the porous monolith erected at the entrance of the Summer Palace Park, Beijing, China[7].

The monolithic column can be defined as a single piece of a continuous rigid porous polymer that possess an interconnected skeletal structure and contains pores. Based on the size of the pores, they can be divided into three types; micropores (< 2 nm), mesopores (2-50 nm), and macropores or through-pores (> 50 nm). Each of these pores has a special benefit; micropores are the most important pores in terms of separation. However, in some cases, the molecules are too big to diffuse through these micropores; therefore, they will interact with mesopores. The main benefit of the macropores is to control the column permeability resulting in reducing the backpressure of the column[8]. For extraction, besides the macropores, the mesopores are the important pores since they can increase the surface area of the monolith, resulting in increased loadability of the monolith[9].

Figure 2 shows the structural differences between a conventional chromatographic column, tightly packed with particles, Figure 2 (A), and a monolithic column fabricated of a single piece of a porous solid with relatively large channels for convective flow, Figure 2(B). The comparison between the scanning electron micrographs of the packed and monolithic chromatographic beds illustrates that the monolithic bed contains a much greater number of channels penetrating the chromatographic bed compared with the column packed with particles.

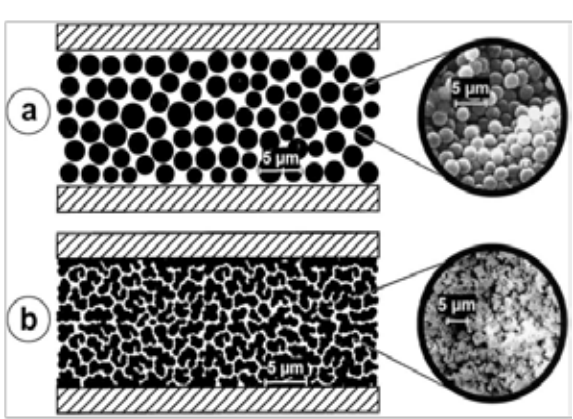

Fig. 2 Structural characteristics of (A) packed, and (B) monolithic chromatographic beds[10].

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1.2. Advantages of the monolithic materials

Monolithic materials have been shown to possessmany advantages compared with packed particles. For example, they are easy to prepare, they have high surface area and high stability, and they are highly permeable to liquid flow compared with the packed bed since porous monoliths contain interconnected macro- and mesopores. This can increase the efficiency of extraction or separation and decrease the backpressure[11, 12]. Furthermore, the monolithic stationary phase does not need frits in comparison with the packed particles[13], which have mechanical problems related to the fragility of the columns and clogging of the column during use [14].

1.3. Characterisation of the monolithic materials

Several methods are commonly used for physical characterisation and measuring the porous properties of monolithic materials. For surface characterisation methods, they can be studied using optical methods such as scanning electron microscopy (SEM), atomic force microscopy (AFM), and transmission electron microscopy (TEM)[12]. These can give information about the morphology of the monolithic materials, which is closely related to their porous properties. Moreover, they are used to estimate the size of the pores, which will in turn determine the hydrodynamic properties and mechanical strength of the column[14]. The physical properties of monolithic materials, such as the porosity and pore size distribution, are studied using mercury intrusion porosimetry (MIP), which has the ability to measure large pores ranging from 10 nm to 150 μm. For measuring small pores (less than 50 nm), inverse size exclusion chromatography (ISEC) is commonly used. Moreover, the specific surface area and the pore size distribution can be studied using the Brunauer-Emmett-Teller (BET) method, which involves measuring the volume of N_2 adsorbed on the surface of the monolithic materials and the surface area can then be calculated from the adsorbed volume of an N_2 molecule[4, 14].

1.4. Applications of the monolithic materials

Due to the unique properties of monolithic materials, they are finding their place in a variety of fields. For example, they have been used in various types of chromatography, such as gas chromatography (GC), high performance liquid chromatography (HPLC), and capillary electrochromatography (CEC). These applications have been described in many reviews[4]. However, less common applications of monoliths are as carriers for immobilisation of enzymes, static mixers, thermally responsive gates and valves, as well as a solid phase support for extraction and preconcentration[15].

2. Organic polymer-based monoliths

According to their components, monolithic materials can be divided into organic polymer-based monoliths and inorganic silica-based monoliths. This review focuses on organic polymer-based monoliths only. An organic monolith is a single block of highly porous material that consists of polymer globules separated by numerous interconnected cavities (pores), and held together through extensive crosslinking[16].

2.1. Fabrication of the organic monoliths

The preparation of a polymer-based monolith is produced by a "moulding" process, which is relatively simple and straightforward compared with silica-based monoliths[12]. Before fabrication of the polymer-based monolith inside a capillary or a microchip, its inner walls are silanised in order to prevent the movement of the monolith during the procedure. Commonly, the capillary column or the microchip is rinsed with a strong basic solution in order to hydrolyse the siloxane groups at the inner surface resulting in increased density of the silanol groups. The silanisation of the inner walls is carried out using a bifunctional reagent solution, typically 3-(trimethoxysilyl) propyl methacrylate (γ-MAPS), which is allowed to react for a period of time. As a result, trimethoxysilane functional groups of the silanising agent will be anchored to the silanol groups while the methacrylate groups of the silanising agent will participate in the polymerisation reaction, causing the monolith to be chemically bound to the inner walls of the capillary or the microchip[4]. On the other hand, a further increase in silanisation reaction can cause a heterogeneous porous structure and form a less porous layer at the inner walls[17].

The silanised capillary or microchip is filled with the polymerisationmixture, which commonly consists of monovinyl monomer, divinyl crosslinking monomer, porogenic solvents, and free radical initiator[18]. The polymerisation reaction consists of three steps; the first is the initiation step, when the initiator produces free radicals that will react with the monomer. Following initiation, the process continues with the successive addition of monomer units to the chains. This is known as propagation step. The last step is the termination step and this happens when two free radicals react together and are no longer available to catalyse polymerisation; as a result, the polymerisation reaction will stop[19, 20].

Generally, the initiation of a polymerisation reaction can be performed by thermal initiation (heat), or photoinitiation(light), which can only be carried out in a transparent mould such as a glass tube, fused silica capillary, or glass microchip[12]. Thermally initiated polymerisation was a standard method for fabrication of the organic monolith in a completely filled closed column such as a column for HPLC[21]. However, adjustment of the length and position of the organic monolith is difficult to achieve using thermal initiation. Fabrication of organic monolithic material using photoinitiatedpolymerisationemerged in 1997 when Viklund*et al.*[22] fabricated a polymer-based monolith using glycidyl methacrylate and trimethylolpropanetrimethacrylate inside a quartz tube (i.d. 2.4 mm). The results of this study showed that using a light initiated polymerisation process is faster than the thermally initiated polymerisation process. Photoinitiationhas been used for fabrication of organic monoliths inside a capillary column or a microfluidic device for CEC, which requires formation of the organic monolith in a specific location using a mask that prevents conversion of monomers to polymers in those areas that are not irradiated[23].

After the polymerisation reaction, the organic monolithic material is washed in order to remove unreacted materials, such as residual monomers, the initiator, and porogenic solvents that remain in the pores of the monolith. Typically, the washing procedure is carried out using a mechanical pump or applying a voltage for a period of time[24]. After washing the monolithic material, it is ready to use. The scanning electron micrograph in Figure 3 represents an example of the porous structure of a polymer-based monolith. The fundamental building units of this network are nuclei, which grow and agglomerate into globules, which will further agglomerate into larger clusters[25].

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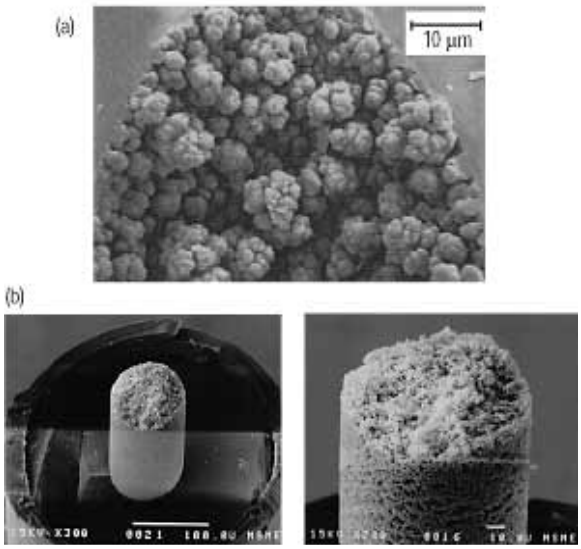

Fig.3 Scanning electron micrographs of polymer-based monoliths by (A) Horvath and coworkers,[26] and (B) Svec and coworkers [27] in 1999.

Polymeric monoliths can be fabricated by using a wide range of monomers and crosslinking agents enabling the porous properties of the monolith to be controlled; for example, acrylate/methacrylate-, styrene-, and acrylamide-based stationary phases[28]. Figure 4 shows the common monomers and their crosslinking agents that have been used for fabrication of polymer-based monoliths. The monolith can be functionalised by co-polymerisation or post-polymerisation in order to convert it into a sorbent with the desired chromatographic binding properties[26].

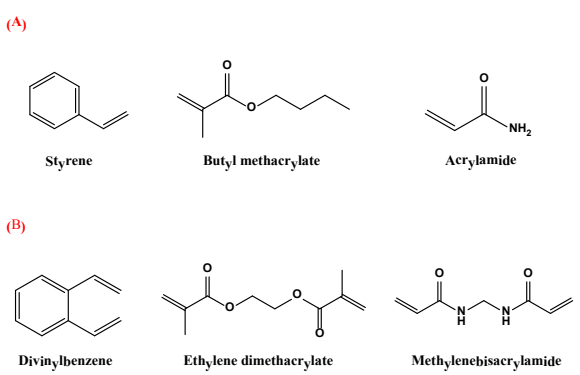

Fig.4 Examples of monomers (A), and their crosslinking agents (B) utilised for the fabrication of the polymer-based monoliths.

2.2. Optimisation preparation of the organic monoliths

The most effective parameters in the fabrication of a polymer-based monolith that can affect its properties are the proportion of monomer to crosslinker, the polymerisation temperature or time of exposure to UV light, concentration of the initiator, and the percentage of the porogenic solvent system in the polymerisation mixture. The specific surface area and the pore size distribution are very sensitive to any variations of the listed parameters [25].

Changing the ratio of monomer to crosslinker (monovinyl/divinyl monomer ratio) can be utilised to control the porous properties of the organic monolith[31]. As the ratio of crosslinker to monomer increases, the monolith will be more dense and highly microporous, resulting in increase of the surface area of the monolith due to the shift of the pore size distribution to the small pore diameter as a result of early formation of highly cross-linked globules [32].

The polymerisation temperature can affect the kinetics of the polymerisation reaction, the pore size distribution, and the specific surface area. As the polymerisation temperature decreases, the reaction rate is slower and larger pores will be formed, resulting in a decrease in the surface area, while if the polymerisation temperature is increased, the reaction rate is quicker and smaller pores will be obtained, resulting in an increase in the surface area[23].

Careful optimisation of the exposure to the UV light is required since the polymerisation time is responsible for conversion of monomers. As the polymerisation reaction continues, the degree of branching increases resulting in growing the polymer chains and making the monolith dense while reducing the irradiation time can lead to formation of less monolithic material in the capillary or the microchip, which can affect the performance of the fabricated monolith[33]. Figure 5 shows the effect of the irradiation time on the growth of the polymer chains. By increasing the irradiation time, the degree of branching increases resulting in forming a dense crosslinked polymer network[34].

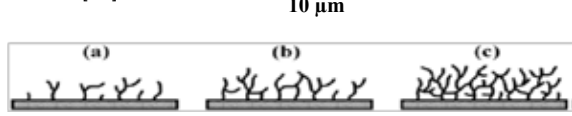

Fig. 5 Schematic representation of the growing polymer chains during photografting with increasing irradiation time from (A) to (C)[34].

The effect of the concentration of initiator on the properties of the polymer-based monolith was firstly studied by Xie *et al.*[35], who mentioned that using a higher concentration of the initiator can increase the number of radicals resulting in an increase in the number of nuclei, which can lead to formation of small pores.

Increasing the percentage of the porogenic solvent system in the polymerisation mixture can lead to increase in pore volume, which can affect the rigidity of the polymer. A useful tool for preparation of a polymer-based monolith that can control the pore size without changing the chemical composition of the polymerisation mixture is the type and composition of the porogenic solvent system[23]. The polymerisation reaction starts from an initially homogeneoussolution until the polymer precipitates, and further polymerisation and crosslinking continues both in the swollen nuclei and in solution[36]. As a rule, using a poorer solvent affects the solvation of the polymer chains leading to formation of large pores due to an earlierpolymer phase separation[12, 35], while if a good solvent is used, the phase separation happens later and this can lead to formation of small pores[36].

2.3. Properties of the organic monoliths

The important property of typical monolithic materials is high surface area, which can be increased by increasing the number of micropores. On the other hand, the permeability of the monolith requires macropores in order to

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allow liquid to flow through the monolith at a reasonable pressure[23]. Both the surface area and the hydrodynamic properties of the monolith depend on the pore size distribution of the monolith; therefore, a balance between the requirement of low flow resistance and high surface area must be found, and the ideal monolith should have both macropores in order to achieve sufficient permeability of the monolith, and micropores for high capacity[12, 29]. Many studies have been carried out to increase the surface area as well as the permeability of flow through the monolith by optimising the composition of the polymerisation mixture and the reaction conditions[30].

There are many advantages of organic polymer-based monoliths since they contain micropores, which can provide the desired surface area, and macropores, which can allow a high flow rate at moderate pressures. Moreover, these polymeric monolithic materials are fritless, and they are covalently bonded to the inner walls of the capillary or microchip[37]. Another advantages of polymer-based monoliths are they can be washed with caustic mobile phase, are stable over a wide range of pH values, and they are easy to prepare compared to silica-based monoliths because they are prepared in a single step by in situ polymerisation. Moreover, the desired length and shape to be exposed to the light source can be controlled easily by using electrical masking tape or foil, and this will help the fabrication of the monolith inside a microchip[38]. However, it can be difficult to ensure the pores are large enough to reduce the backpressure, and that the mesopores are distributed over the desired size range. In addition, organic monolithic materials are not mechanically stable since they are affected by temperature and/or organic solvents causing shrinking or swelling and this can affect the performance of the monolith[39, 40]. Commonly, organic polymer-based monoliths are prepared on a small scale because the fabrication of a large size monolith is quite difficult. The reason for that is the unstirred nature of the polymerisation within the mould can cause a decrease in the capacity to dissipate the heat of polymerisation and creation of heterogeneity in the pore structure[4, 41]. Furthermore, polymer-based monoliths have low binding capacity, which is attributed to the low specific surface area. Although some attempts have been made to increase the surface area of organic monoliths, the fabricated monoliths in previous reports showed relatively low surface areas[42, 43].

Applications

Porous polymer-based monoliths have found widespread use in many different applications such as liquid chromatography, solid phase extraction, and enzyme immobilisation in capillary and microfluidic chip formats[17]. This review will focus on the applications of polymer-based monoliths in solid phase extraction. The history of using monolithic materials as sorbents is not very long. The first paper reporting the use of a porous polymer monolith for solid phase extraction was by Xie*et al*. in 1998[42, 44]. The porous poly (ethylstyrene-*co*-divinylbenzene) monolith was prepared using commercial 80 % divinylbenzene, 20 % ethylstyrene, and dodecanol with toluene as a porogenic solvent. The monolith was fabricated inside a threaded polyetheretherketone (PEEK) tube (20 mm × 1 mm i.d.) using thermal initiation at a temperature of 70 °C for 24 hours. The result showed that by increasing the percentage of divinylbenzene (crosslinking monomer) to ethylstyrene in the polymerisation mixture, the surface area was increased. Excellent properties of this fabricated monolith were demonstrated since it had excellent hydrodynamic properties (average pore size of 6 µm) and a very high surface area (400 m^2 g^{-1}). Its performance was checked by preconcentration of polar organic compounds and quite high recoveries of about 85 % were achieved.

Lee *et al*.[45] have fabricated an organic monolith using glycidyl methacrylate (GMA), and trimethylolpropanetrimethacrylate (TRIM). After fabrication of the poly (GMA-*co*-TRIM) monolith, protein G was immobilised on the monolith for preconcentration and capillary zone electrophoresis (CZE) of immunoglobulin G (IgG) from human serum. The monolithic material (1.5-2 cm) was fabricated inside the inlet end of a fused silica capillary (75 µm i.d. and 365 µm o.d.). The method was able to preconcentrate and clean up IgG from a human serum sample that was diluted to 500 and 65,000 times (Figure 6). However, there was no true CZE separation since only one protein was investigated. The authors stressed that the fabricated organic polymeric monolith is applicable to preconcentrate any protein for which an antibody is available.

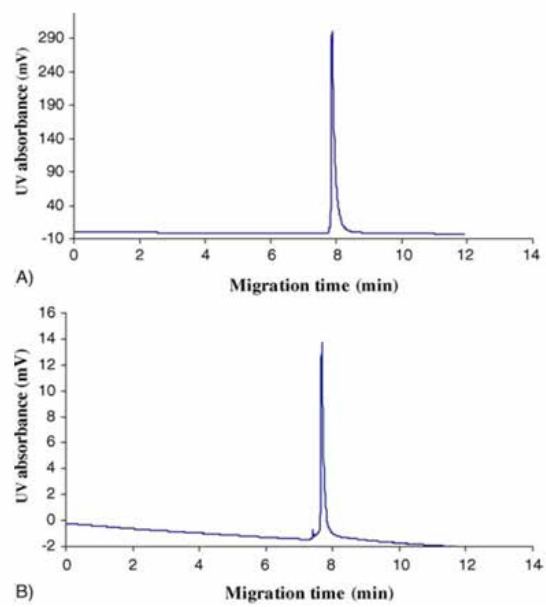

Fig.6Electropherograms of on-line preconcentration-CE of IgG from human serum. Experimental conditions: 1.5 cm monolithic material inside a 64 cm (53 cm to detector) fused silica capillary (75 µm i.d.); 50 mM formic acid (elution buffer); 12.5 mM ammonium formate-formic acid (pH 7.6) separation buffer; 15 kV applied separation voltage; UV detection at 214 nm. (A) 500 times diluted human serum, and (B) 65,000 times diluted human serum[45].

Later the same group fabricated a poly (butyl methacrylate-*co*-ethylene dimethacrylate) (BuMA-*co*-EDMA) monolith inside the inlet end of a polyvinyl alcohol-coated silica capillary, followed by coupling this hydrophobic monolith with the previous monolith via a zero dead volume union for on-line removal of IgG, extraction of standard proteins (lysozyme, cytochrome C, and trypsinogen A), and separation by CZE[46]. The coupled monoliths were equilibrated with 50 mM ammonium formate-formic acid (pH 7.6), followed by injection of the protein solution containing IgG for 50

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min. The hydrophobic monolith was detached from the affinity monolith (the protein G column) and mounted in the CE instrument. The monolith was washed with ammonium formate-formic acid buffer to remove unbound protein, and finally the preconcentrated proteins were eluted using 70 % ACN containing 0.1 % TFA. A good preconcentration and separation were obtained by this method, with RSD for peak area below 3 %, and 1 % for migration time. The authors suggested that this system could be a valuable means of sample preparation for preconcentration of low abundance proteins in a complex sample such as human serum and removal of high abundance proteins such as IgG, and human serum albumin (HSA). Although this work showed the possibility of coupling two types of organic polymeric monoliths (affinity and hydrophobic) for sample clean-up and preconcentration, the technique required disconnection of the two monoliths before washing and elution of the preconcentrated proteins, which can cause loss of the preconcentrated proteins. Additionally, the technique would require affinity for a wide range of high abundance proteins.

Schley*et al.*[47] fabricated poly (styrene-*co*-divinylbenzene) (PS-*co*-DVB) monolith inside a fused silica capillary for both desalting and preconcentration of peptides and proteins (in 10 mm × 0.20 mm i.d. format), and for analytical separation (in 60 mm × 0.20 and 0.10 mm i.d. format). The fabricated monoliths were coupled with HPLC and the detection method was UV absorbance at 214 nm. The hydrophobic monolith showed its ability to preconcentrate and separate seven standard proteins, which were ribonuclease A, cytochrome C, lysozyme, transferrin, myoglobin, α-lactoglobulin B, and carbonic anhydrase. The effect of sample volume and concentration on the extraction recovery were investigated by injection of constant amounts of proteins dissolved in different volumes of water containing 0.050 % TFA (ranging from 1 to 100 µL). As can be seen in Figure 7, reducing the concentration of the standard proteins by a factor of 100 accompanied with an increase in the injection of the sample volume did not affect protein recovery since there was no difference in the peak areas obtained for different concentrations and injection volumes.

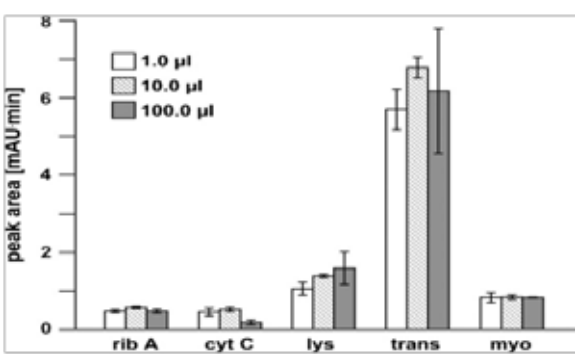

Fig.7Preconcentration of a constant amount of proteins dissolved in different volumes of water containing 0.050 % TFA (1, 10, 100 µL). The plotted peak areas represent the average of two replicates[47].

Extraction using organic monolithic material on a microfluidic device was first demonstrated by Yu *et al.*[48, 49] in 2001. The organic polymer monolith (7 mm long monolith) was fabricated within a simple straight microchannel (100 µm wide, 40 µm deep, and 6 cm long) of a glass microchip using photoinitiated polymerisation (UV irradiation at room temperature for 3 hours) in order to use it for on-chip solid phase extraction and preconcentration of small molecules, peptides, and proteins. Two types of organic polymer monolith were prepared in the chips. The first one was a hydrophobic monolith using butyl methacrylate with ethylene dimethacrylate, and the second one was ion-exchange (IE) using two monovinyl monomers, 2-hydroxyethyl methacrylate (HEMA) and [2-(methacryloyloxy)ethyl]trimethylammonium chloride (META) with ethylene dimethacrylate. Both polymeric monoliths were prepared using 2,2′-azobis(2-methylpropionitrile) (AIBN) as photoinitiator and a binary porogenic solvent, hexane and methanol. Both types of monoliths were able to preconcentrate a small organic acid (coumarin 519). Moreover, they were utilised to preconcentrate a recombinant green fluorescent protein (GFP), and a fluorescently labelled tetrapeptide. GFP was preconcentrated using the hydrophobic monolithic concentrator up to an enrichment factor of 1000 with elution of the protein using a 1:1 water/acetonitrile mixture at a flow rate of 0.53 µL min^{-1} (Figure 8). Although the fabricated hydrophobic and ion-exchange monoliths met the specific requirements for formation of macroporous monoliths (pore sizes were 19.5 and 13.2 µm, respectively), the specific surface areas of both fabricated monoliths were relatively low (0.7 and 1.3 m^2 g^{-1}, respectively).

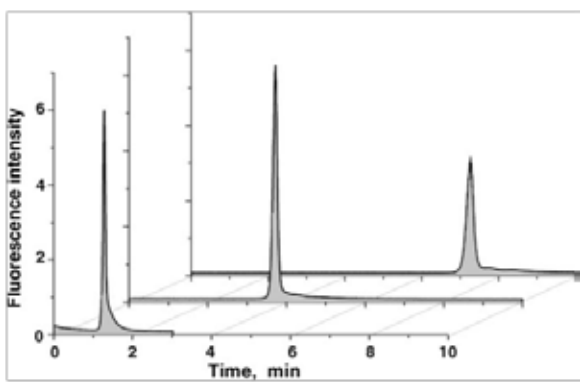

Fig. 8 Elution of green fluorescent protein from hydrophobic monolithic concentrator. Conditions: loading 200 µL of protein solution in tris-HCl buffer solution (pH 8.0) containing 0.95 mol L^{-1} ammonium sulfate. The solution was pumped at a flow rate of 3 µL min^{-1}, elution with 1:1 acetonitrile/water at a flow rate of 3 (1), 1.03 (2), and 0.53 µL min^{-1} (3)[48].

Hua *et al.*[50] in 2011 fabricated a monolithic bed with two different surface chemistries by co-polymerisation of BuMA with META to form a monolith for solid phase extraction that supports anodal electroosmotic flow. The 1 mm long organic monolith (20 µm deep and 140 µm wide) was fabricated at the centre of a 3.5 cm long channel of a glass microchip, Figure 9. The ends of the channel (20 µm deep and 56 µm wide) were sharp to reduce band-broadening effects. The microchip was coupled to electrospray mass spectrometry (ESI-MS) detection and its performance was checked by preconcentration of cytochrome C and myoglobin, as can be seen in Figure 10, which shows the ion electropherograms and mass spectra of the preconcentratedwevee peak of th

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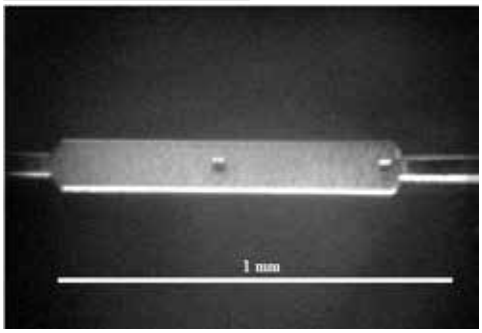

Fig.9 Optical micrograph of an enlarged monolith polymer bed (1 mm long), prepared in a 20 µm deep and 140 µm wide microchannel that has sharp edges [50].

Li *et al*.[51] fabricated a poly (GMA-*co*-TRIM) monolith inside a channel of a glass microchip, which was fabricated using traditional photolithography and wet etching techniques. Figure 11 illustrates the extraction channel (2 cm long) containing the monolithic material and the injection arms (1.5 cm long). The width and depth of the channels were 100 µm and 25 µm, respectively. All flow in the microchip was carried out using a syringe pump (pressure driven).

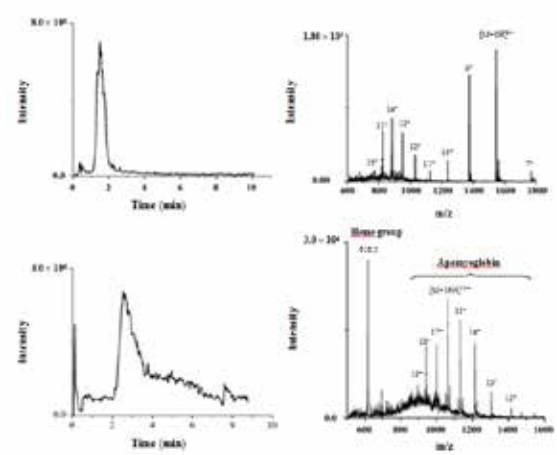

Fig.10 Total ion electropherograms and mass spectra of the preconcentrated cytochrome C (upper trace) and myoglobin (lower trace). Loading buffer (5 mM formic acid), and elution buffer (5 mM formic acid with 60% ACN). Samples were loaded and eluted electrokinetically with −2.5 kV at the sample reservoir and 3.2 kV on the electrospray tip[50].

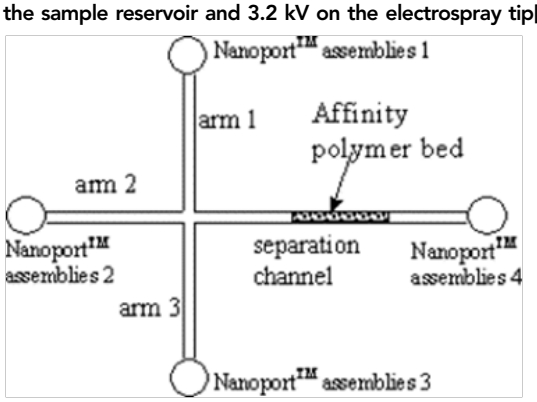

Fig.11 Schematic diagram of the glass microchip used for preconcentration of the protein, the extraction channel contained the polymeric bed, which was poly (GMA-co-TRIM) monolith[51].

After fabrication of the monolith inside the extraction channel, it was derivatised with Cibacron-blue-3G-A, which is a triazine dye that has group specificity for some proteins such as albumin, dehydrogenase, interferon, lysozyme, and related proteins [52]. The extraction of proteins was carried out by injection of the protein sample solution using a syringe pump from nanoport assemblies 1 while the tubing connected to nanoport assemblies 3 was open and the others were closed. When the fluorescent sample was seen in arms 1 and 3 using a fluorescence microscope, nanoport assemblies 3 was closed while nanoport assemblies 4 was open. After preconcentration of the protein, the arms 1, 2, and 3 were flushed in order to wash the polymer bed. The fabricated Cibacron-blue-3G-A-modified polymeric monolith was tested to preconcentrate a standard protein (lysozyme). Cytochrome C and lysozyme were derivatised with fluorescein-5-isothiocyanate (FITC), and then the FITC-labelled lysozyme and FITC-labelled cytochrome C were preconcentrated individually using the fabricated device and fluorescence images were taken. Figure 12 (A) illustrates the fluorescence image of the preconcentrated FITC-labelled lysozyme, indicating the ability of the fabricated monolith to preconcentrate lysozyme, while Figure 12 (B) shows no significant fluorescent signal was observed for preconcentration of FITC-labelled cytochrome C, which indicated that there was no significant adsorption of cytochrome C. A mixture of unlabelled lysozyme and FITC-labelled cytochrome C was investigated and the fluorescence signal was similar to the detected fluorescent signal of FITC-labelled cytochrome C, Figure 12 (C). The main drawback of this work is that the porous properties of the fabricated monolith were not reported. Furthermore, the fluorescent signal of the preconcentrated FITC-labelled lysozyme was high at the end of the polymer monolith, which indicates nonuniformity of the fabricated monolith. In addition, the benefit of the injection channel (arms 1 and 3) was not clear.

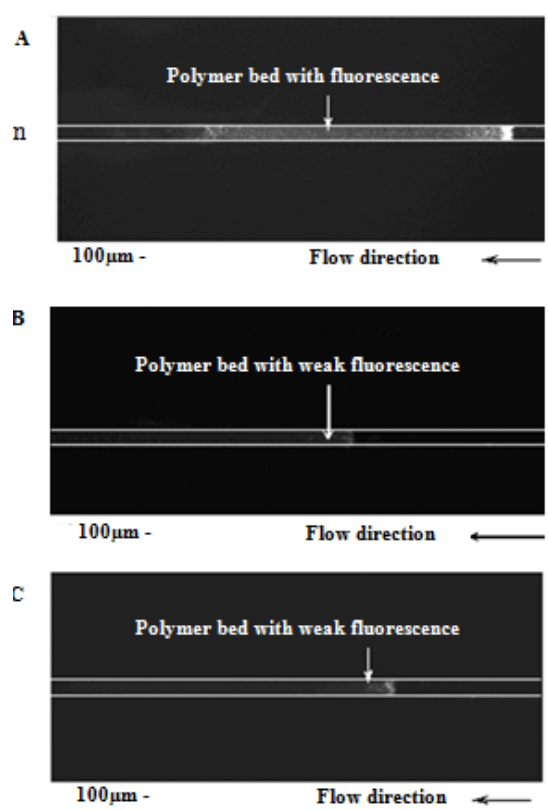

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Fig.12 Fluorescence images of the polymer bed in a glass microchip: (A) the monolithic bed with captured FITC-labelled lysozyme, (B) capturing FITC-labelled cytochrome C, and (C) capturing unlabelled lysozyme and FITC-labelled cytochrome C mixture[51].

Conclsions

This review reports the contributions of several groups working in the preparation of the polymer-based monoliths. Many studies have been carried out to increase the surface area as well as the permeability of flow through the monolith by optimising the composition of the polymerisation mixture and the reaction conditions. In addition, this review highlights the recent applications of the oganic monolithic materials as sorbentmaterials for analytes preconcentration.

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