In-process phase growth measurement technique in the fabrication of monolithic porous layer open tubular (monoPLOT) columns using capacitively coupled contactless conductivity

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Abstract

A technique for the in-process measurement of polymer stationary phase growth inside fused silica capillaries during the fabrication of monolithic porous layer open tubular (monoPLOT) columns is presented. In this work, capacitively coupled contactless conductivity detection (C4D) was applied as an online measurement tool for porous polymer layer growth within fused silica capillaries. The relationship between effective capillary diameter and C4D response was investigated for two polymers, butyl methacrylate–ethylene dimethacrylate (BuMA–EDMA) and polystyrene–divinylbenzene (PS–DVB) over a range of capillary diameters and layer thicknesses. The described technique can be used with both thermal and photo-initiated approaches for monoPLOT fabrication and provides an accurate, realtime measurement of the porous layer growth within the capillary, which should vastly improve columnto-column reproducibility. The technique was shown to be very precise, with a measured %RSD < 10%.

1. Introduction

Monolithic porous layer open tubular (monoPLOT) columns are a type of open tubular (OT) column which possess a monolithic porous polymeric phase covalently bonded to the inner wall of the capillary. Compared with standard open tubular formats, such as wall-coated open tubular capillaries (WCOT), monoPLOT capillary columns are expected to exhibit a much higher capacity due to the high surface area of their porous structure. Crucially, monoPLOT columns are a type of bonded phase and are chemically attached to the wall of the capillary, making the stationary phase much more resilient and less prone to column bleed under stressed conditions. This property of monoPLOT columns is of particular importance in gas chromatography (GC) where the column is subjected to high temperatures.

Several techniques for the manufacture of PLOT columns exist [1–9] and include both thermal and photo-initiated methods. However, in the majority of these techniques, it was difficult to precisely control layer thickness and morphology. Recently, a work has been published by the authors on a method which allows the ☐ne control of both layer thickness and morphology.[2] In this case, the capillary was subjected to repeated exposures to UV light, slowly building up the required layer thickness while also maintaining the desired morphology. However, in this method, the authors used experimental or empirical data to manufacture monoPLOT columns of the required characteristics.

Although recent works have shown excellent results in column to column reproducibility there is still a significant amount of experimental work which must be done to achieve the

required layer thickness and pore morphology. Changing the pore structure and/or layer thickness usually means the development of new procedures, and so the manufacture of monoPLOT columns in different capillary sizes, with different layer thickness and structure creates a lot of additional experimental work. In addition to this, there is a significant need for in-process checks during the fabrication process simply as a quality control measure.

In this work, a non-invasive, in-process measurement of the layer growth is presented and its application to existing methods for the fabrication of monoPLOT columns through both photo and thermal initiation is discussed. For the First time, C4D is examined as a possible on-line technique which can be used to measure layer growth during laminar flow polymerisation. Additionally, the advantages, disadvantages, and suitability of the method are discussed, particularly in relation to the fabrication method which is being employed.

2. Theory

2.1 C4D response

Works on contactless conductivity detection were first published in 1980 by Gas et al.,[10,11] however, C4D as we know it today was developed by two independent research groups in 1998[12,13] as a detection method for small inorganic ions in capillary electrophoresis. Two radial electrodes (which are placed around the capillary at approximately 2–5 mm apart) supply and detect an induced current in the solution within the capillary. The magnitude of the detected signal is proportional not only to the magnitude of the excitation signal, but also to the concentration and mobility of the ionic charge carriers within the solution. A complete explanation of the fundamental concept behind C4D is beyond the scope of this work but can be found in the excellent review by Kuban and Hauser.[14]

C4D has been previously used to characterise stationary phase homogeneity of packed columns[15,16] polymer monoliths formed within capillaries[17] and layer thickness of preformed monoPLOT columns in off-line approaches.[1,2] The application of C4D in what has been termed capillary 'scanning mode' can be used to differentiate between monoliths of different morphologies, with Connolly et al.[15–17] having shown that C4D can be used successfully as a non-invasive method for the determination of monolith longitudinal homogeneity, determination of coating stability and location, as well as identification of monolith or packing material voids.

The application of C4D to the detection of polymer layer growth with a capillary is technically relatively straightforward; however, there are some considerations which must be taken into account. First of all the polymerisation mixture must contain a charged species in order to generate any kind of useful signal and so a 1 wt% solution of 10 mM NaCl prepared in H_2O was added to the polymerisation mixture. Secondly, the type of polymerisation is important – for photo-initiated polymerisation the C4D cell must be placed outside the reaction chamber, or else the electrodes should span the chamber so as not to mask any area of the capillary. For thermally initiated polymerisation the C4D cell must be immersed in the

air or water bath, and so some additional considerations must be made for this setup, these will be discussed later.

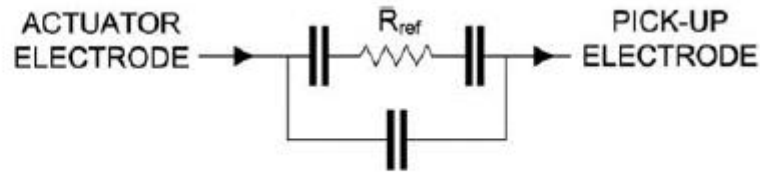An equivalent circuit diagram of the C4D cell is shown in Fig. 1.

Fig. 1 Equivalent circuit diagram of a typical C4D cell.

In this model, R_{ref} represents the resistance of the solution between the electrodes. When a charged species passes through the detector the resistance of R_{ref} decreases due to the increased conductivity and thus the corresponding voltage drop across the cell, V_{Rref} will change. There is an inverse relationship between resistance and conductivity:

$$\sigma = \frac{1}{\rho} \tag{1}$$

where, σ = conductivity of the conductor material, ρ = resistivity of the conductor material.

Resistance is not only a function of the electrical resistivity of the conductor material (in this case the polymerisation solution) but also depends on the geometry of the conductor.

The resistance, R, of a conductor of uniform cross-section is given by:

$$R = \rho \frac{L}{A} \tag{2}$$

Where, L = length of conductor, A = cross-sectional area of conductor.

As can be seen from eqn (2), the resistance of a conductor is inversely proportional to its cross-sectional area. The volume of polymerisation mixture that exists between the excitation and pick-up electrodes (separated by a distance, L) can be viewed as the resistor in the case above. At the start of the polymerisation process, no porous polymer layer exists within the capillary and so the effective cross-sectional area, A_{eff}, of the resistor is equal to the capillary cross-sectional area (see Fig. 2).

As the layer grows, the effective capillary cross-sectional area, A_{eff}, will get smaller, increasing the resistance, R_{ref}, and causing a larger voltage drop across the C4D cell. By measuring the change in voltage drop, DV_{Rref}, it is possible to measure the porous layer build up inside the capillary. Since the conductivity (and hence the resistance) of the volume between the electrodes is proportional to the ionic mobility of the solution, pumping the mixture through the capillary during the polymerisation process results in a very stable response
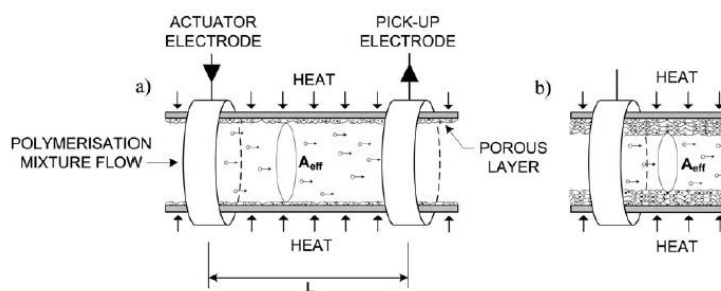
Fig. 2 (a) Section view of capillary and C4D electrodes during the polymerisation process and (b) the reduction in the cross-sectional area, A_{eff}, due to layer growth.

3.Experimental

3.1 Reagents and materials

All chemicals were reagent or analytical grade purity. Ethylene dimethacrylate (EDMA), butyl methacrylate (BuMA), styrene, divinylbenzene, 4-vinylbenzyl chloride, 1-decanol, toluene, benzophenone, 3-methoxysilylpropyl methacrylate, sodium hydroxide (NaOH) and hydrochloric acid (HCL) were all purchased from Sigma-Aldrich (Gillingham, UK). The thermal initiator, azobisisobutyronitrile (AIBN), was obtained from DuPont (Le Grand Sacconex, Switzerland). All solvents which were used for the synthesis and washing of prepared monoliths, namely, acetonitrile (ACN), acetone, and methanol (MeOH), were purchased from Lab Scan (Gliwice, Poland). Deionised water purified by a Milli-Q system (Millipore, Bedford, MA, USA) was utilised throughout the experiments. Polyimide coated (15 μm thickness) fused silica capillary, 25, 50, 75, and 100 μm ID, 0.375 mm OD was purchased from Composite Metal Services Ltd, Charlestown, United Kingdom.

3.2 Instrumentation

Capillaries were filled with monomer mixture and washed using a KDS-100-CE syringe pump (KD Scientic, Inc., Holliston, MA, USA). Fabrication of thermally initiated monoPLOT columns for off-line C4D characterisation was carried out in a water bath, using a Yellow Line MST Basic hotplate with TC1 temperature controller and glassware (VWR Ltd, Dublin, Ireland). Thermally initiated formation of monoPLOT columns using C4D as an inprocess measurement was carried out in the column oven of a Dionex Ultimate 3000 nano-HPLC system (Dionex, Sunnyvale, California, United States).

A PHD2000 syringe pump, purchased from Harvard Apparatus (Holliston, MA, United States) was also used to pump polymerisation mixture during the fabrication process. A Rheodyne 6-port switching valve (Rheodyne, California, United States) was used to switch between the polymerisation mixture and MeOH flows. For the measurement of layer growth by C4D during thermal polymerisation, a TraceDec Capacitively Coupled Contactless Conductivity Detector (Innovative Sensor Technology GmbH, Strasshof, Austria) was used. Settings used during the measurement were as follows: frequency, 3 high; voltage, 6 dB; gain, 50% and offset, 0. A purpose built, C4D sensor head, designed to operate at elevated temperatures was specially made for these experiments by Innovative Sensor Technology

GmbH. Data acquisition was done via TraceDec Monitor V. 0.07a so☐ware, also supplied from Innovative Sensor Technology GmbH. A SputterCoater S150B (BOC Edwards, Sussex, United Kingdom) was utilised for coating capillary monolithic stationary phase samples with a 60 nm gold layer prior to subjecting samples to scanning electron microscopy (SEM) analysis. SEM was performed on a S-3400N instrument (Hitachi, Maidenhead, United Kingdom).

3.3 Procedures

3.3.1 Capillary pre-treatment and monomer mixtures. Fused silica capillaries were initially pretreated through activation of the surface silanol groups of the inner walls by sequential flushing with 1 M NaOH, deionised water, 0.1 M HCl, deionised water, and acetone. The pretreated capillary was silanised using a 50 wt% solution of trimethoxysilylpropyl methacrylate in toluene at 60 C for 24 hours.

The monomer mixture for the fabrication of methacrylatebased polymers consisted of 24 wt% BuMA, 16 wt% EDMA, 60 wt% 1-decanol, and 0.4 wt% AIBN (with respect to monomers) and for polystyrene-based it consisted of 12 wt% styrene, 12 wt% 4-vinylbenzyl chloride, 16 wt% divinylbenzene, 18 wt% toluene, 41.6 wt% 1-decanol, and 0.4 wt% AIBN (with respect to monomers). The monomer mixtures were prepared as per the procedure described by Collins et al.[2]

3.3.2 Measurement of monoPLOT layer thickness by C4D. C4D measurements can be applied to both thermal and photoinitiated polymerisation, however it was noted that in both cases the S/N ratio of the output signal was dramatically improved by pumping the polymerisation mixture through the capillary during the fabrication process. For studies carried out with C4D, a solution of 1 wt% 10 mM NaCl prepared in H_2O was added to the PS–DVB polymerisation mixture to increase its conductivity.

3.3.3 C4D measurements during thermally initiated polymerisation. Two inlet ports of the switching valve were connected to two syringes filled with polymerisation mixture and with MeOH, respectively. Both syringes were placed in a syringe pump. The desired length of silanised capillary was coiled and one end connected to an outlet port on the switching valve which was mounted outside an air oven. The coiled capillary was mounted in the air oven and the other end was left open so that the polymerisation mixture could flow through it to waste. The C4D cell was attached to the capillary so that it was also mounted within the air oven.

The polymerisation mixture was pumped through the capillary at a linear ☐ow rate of 0.5 mm s[1]. After flow was established and the C4D response had stabilised, the air oven temperature was increased to 60 ^{0}C. The C4D response rose with an increase in temperature and once the oven temperature stabilised the C4D response also became steady state. A☐er

some time, formation of the porous polymer layer began and thus the effective internal diameter of the capillary reduced causing the C4D response to fall. The formation of the porous polymer layer was allowed to continue until the desired C4D output was reached, after which the capillary was removed from the oven. The switching valve was also switched over to flush the capillary with MeOH in order to remove all unreacted monomer. Once the capillary had been thoroughly washed it was removed and dried with nitrogen.

4   Results and discussion

4.1 In-process measurement of monoPLOT layer using C4D

Prior to using C4D response as an in-process measurement for layer thickness, it was necessary to show that the C4D cell was stable during repeated long exposures to elevated temperatures. Initial studies carried out by the authors had shown that after some time the C4D response displayed excessive baseline drift and decreased S/N ratio. Further exposure to elevated temperatures (60 C) resulted in a permanently damaged C4D cell which exhibited poor S/N ratio and sensitivity. Using a purpose built C4D cell designed to operate at 60 ^{0}C the stability tests were repeated. Fig. 3 compares stability of the standard and high temperature C4D cells over a 20 hours period.

As can be seen from Fig. 3, the C4D cell designed to operate at high temperatures remained stable even after 20 hours of repeated exposure to 60 ^{0}C. Secondly, it was necessary to demonstrate that the technique was repeatable, regardless of the position of the C4D cell on the capillary. In order to achieve this, polymerisation mixture without the thermal initiator (AIBN) was pumped through a 30 cm length of empty Ø 100 μm ID capillary at a temperature of 60 ^{0}C.
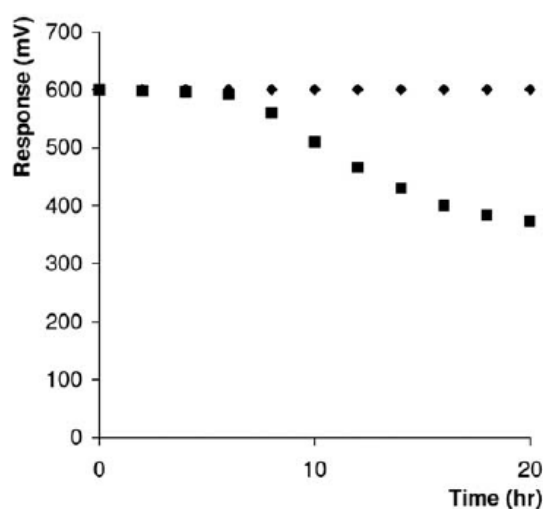

Fig. 3   Comparison of baseline drift for a standard TraceDec C4D cell (■) and a purpose built high temperature C4D cell (◆) over a 20 hours period at 60 °C.
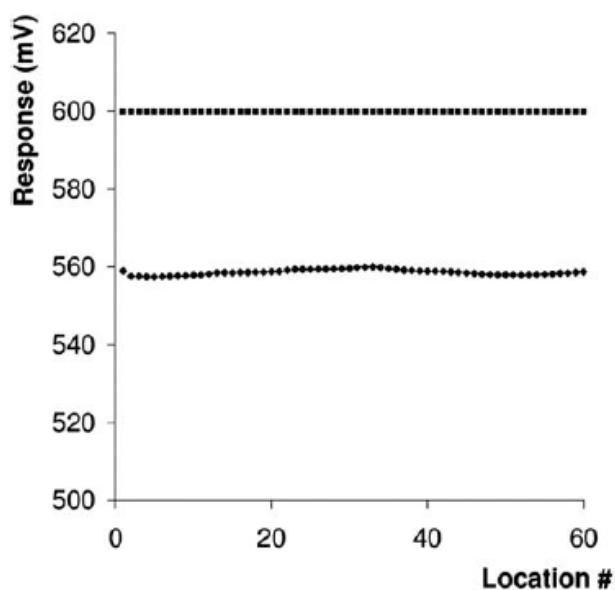
Fig. 4 C4D response recorded at 60 locations for an empty Ø 100 μm ID capillary (■) and for a Ø 100 μm ID capillary with a 2 μm porous layer (◆). Output is approximately 601 and 557 mV respectively.

The C4D response was measured every 5 mm (60 locations) along the column. This was repeated for another 30 cm length of Ø 100 μm ID capillary with a known layer thickness of ~2 mm, again at a temperature of 60 ^{0}C. The linear flow rate was varied between 0.25 and 1.5 mm s^{1} with no change in the response observed. Fig. 4 represents the C4D response for both capillaries at each location, showing a very stable signal for both scans. The difference in the magnitude of signal between empty capillary and monoPLOT with 2 μm layer is clear.

During in-process measurements, the C4D cell was placed inside the air oven and the output was recorded during the polymerisation process. Fig. 5 shows two examples of layer formation and C4D response during polymerisation of BuMA– EDMA and PS–DVB layers at flow rates of 1.0 and 0.5 mm s^{1}, respectively.

C4D response fluctuations due to oven temperature instability at the start of the process can be clearly seen in Fig. 5(a). Fig. 5(c) shows a more stable C4D response, with only slight overshoot, again due to some variation in oven temperature. It is interesting to note that the porosity of the layer has only a slight, but negligible impact on the measured output, however in this instance it is due to the low porosity of the examples which greatly reduce ion mobility in the polymer layer. The polymerisation mixture will flow through larger pores much more freely, and so it is expected that there would be a smaller reduction in ion mobility as the porosity of the layer increases, meaning the technique would be less sensitive to layers with high porosity.

For the example shown in Fig. 5(a), the drop in C4D response (DV_{Rref}) was recorded at 3.3 mV which corresponds to a theoretical layer thickness of 370 nm. The actual average layer thickness (see Fig. 5(b)) was measured at 400 nm, %RSD 7.5%. For Fig. 5(c), the DV_{Rref} value was 63.8 mV which corresponds to a theoretical layer thickness of 2.98 mm. The actual layer thickness (see Fig. 5(d)) was measured at 3.3 mm, %RSD 9.6 %.
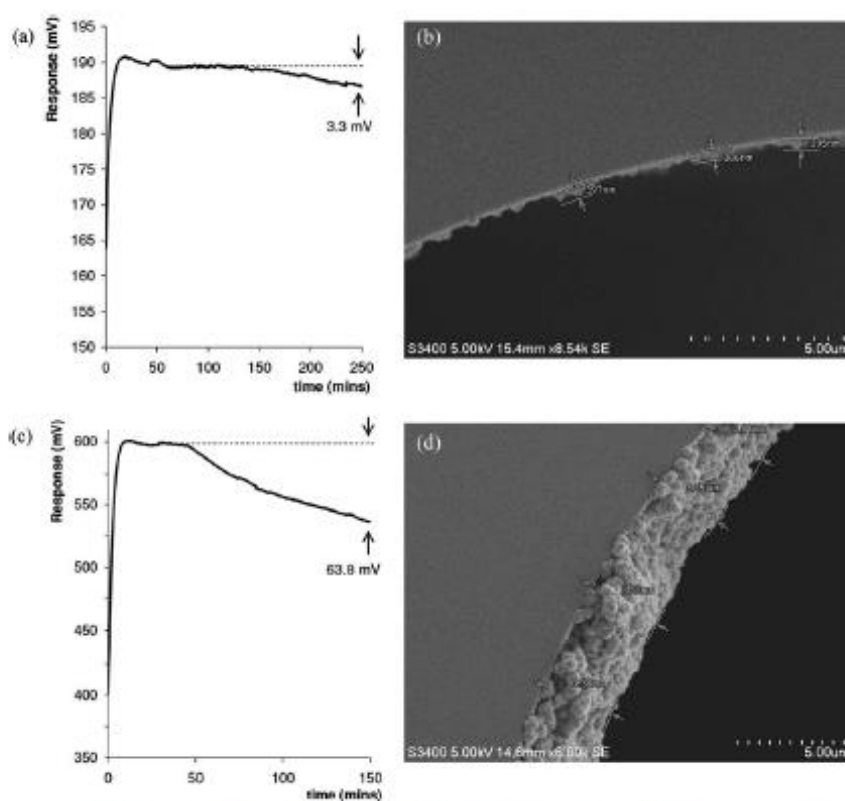
Fig. 5 (a) C4D response recorded during polymerisation of a ~400 nm layer inside a Ø 50 μm ID capillary and (b) SEM image of fabricated layer. Polymerisation conditions: mixture — BuMA–EDMA with 1% 10 mM NaCl, temperature — 60 °C, linear flow rate — 1.0 mm s⁻¹. Change in C4D response was recorded as 3.3 mV, corresponding to a theoretical layer thickness of 370 nm. (c) C4D response recorded during polymerisation of a ~3.3 μm layer inside a Ø 100 μm ID capillary and (d) SEM image of fabricated layer. Polymerisation conditions: mixture was PS–DVB with 1% 10 mM NaCl, temperature — 60 °C, linear flow rate — 0.5 mm s⁻¹. Change in C4D response was recorded as 63.8 mV, corresponding to a theoretical layer thickness of 2.98 μm.

A further study of ten samples was carried out for capillaries of both Ø 50 and Ø 100 μm ID with layers of thickness 400 nm up to 15 μm. The measured C4D response for each capillary was plotted against the average measured layer thickness in each case and Fig. 6 shows the linear relationship between C4D response and effective capillary cross-sectional area, $A_e$ff . Compared with the added electrolyte (NaCl) in the polymerisation mixture, silica, Teflon, and polyimide have very low electrical conductivities, and so the measured C4D response is highly dependant on the effective ID of the capillary.

Although this technique has been shown to be very precise, demonstrating a good level of stability and relatively easy to implement, it has some limitations. Firstly, static polymerisation within the capillary is not possible as no useful C4D response is obtained – the polymerisation mixture must be allowed to ☐ow through the capillary. Secondly, it is necessary to 'spike' the mixture with some charged species. For the two polymerisation mixtures presented within this work, the addition of 1 wt% of 10 mM NaCl had no noticeable effect on the layer morphology or pore structure, however that is not to say that other types of monomer mixture would not be affected. Indeed, initial work in the development of this technique used 2 wt% of 10 mM NaCl, which resulted in the formation of smaller pores and thus lower porosity for the same polymerisation conditions (see Fig. 7).
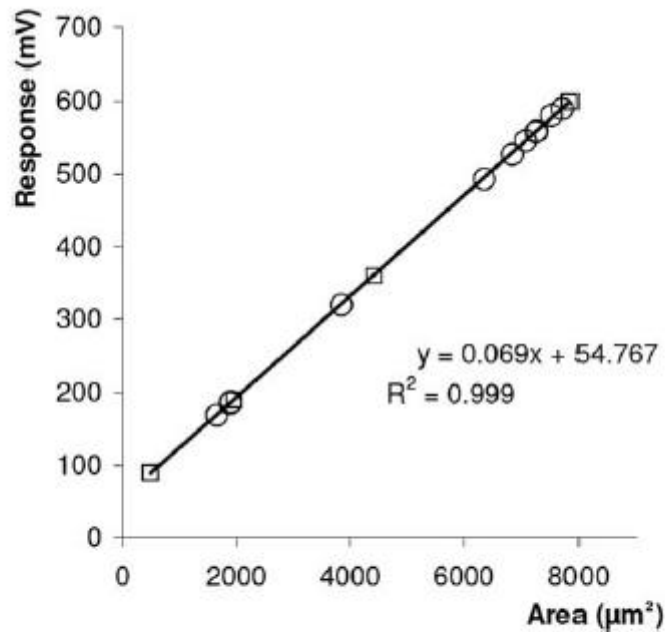
Fig. 6 Relationship between effective capillary cross-sectional area and C4D response, showing both bare capillary (□) and capillary with monoPLOT layer (○).
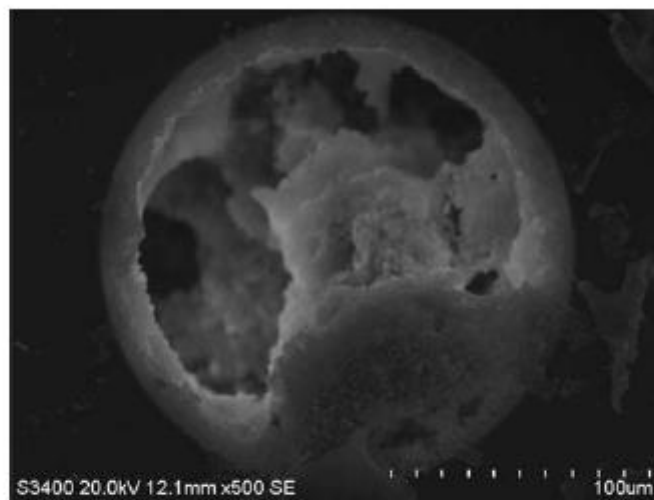

Fig. 7 SEM image of overpolymerised BuMA–EDMA monoPLOT column fabricated from a polymerisation mixture containing 2 wt% of 10 mM NaCl.

Average pore and globule sizes for the BuMA–EDMA polymerisation mixture were measured from SEM images. For the polymerisation mixture containing 1 wt% of 10 mM NaCl, the pore and globule sizes were found to be 0.69 _+0.24 μm and 0.97 0.27 mm respectively, while using 2 wt% of 10 mM NaCl the values were 0.26 _+ 0.10 μm and 0.60 _+ 0.14 mm. The observation correlates well with the theory for the formation of

methacrylate polymers where the introduction (or increase in %) of a polar solvent in the porogen mixture leads to the formation of smaller pores and globules.[18]

The amount and concentration of electrolyte that should be added to the polymerisation mixture is a trade off between C4D sensitivity and affecting the porosity (or in the worst case, undermining the integrity) of the polymer layer. Addition of an electrolyte solution at a high concentration would result in high background C4D signal and as a result, low sensitivity of measurements. On the other hand, the addition of a large volume (in relation to other porogens/monomers in the mixture) of the electrolyte solution at low concentration may have severe impact on the morphology of the polymer layer. However, in cases where conducting monomers are used, there may be no need for the addition of electrolyte.

Perhaps the most difficult aspect of applying C4D as an inprocess tool in the fabrication of monoPLOT columns lies with the long term stability of the electronics in the cell itself. From Fig. 3 it can be seen that for a standard TraceDec C4D cell operated at 60 C, the device will begin to deteriorate rapidly after approximately 8 hours use. Even for the high temperature cell, extended use above 60 ^{0}C was not advised by the manufacturer.

Lastly, temperature has a large effect on the output signal and it is imperative that the oven is very temperature stable. Ion mobility increases with temperature and so any temperature fluctuations within the oven will cause signal fluctuations and reduce the overall accuracy of the technique.

5 Conclusions

A novel method for the in-process measurement of layer growth during fabrication of monoPLOT columns is described, showing a linear relationship between C4D and effective crosssectional area of the capillary. This method allows precise, noninvasive, real time measurement of the growth of the polymer layer, enabling the user to finely control the fabrication process. It can also be used as a quality control test, allowing the user to characterise the monoPLOT column during production. The proposed method was demonstrated with two types of polymer, namely PS–DVB and BuMA–EDMA, on two column diameters, and over a wide range of layer thicknesses, from 400 nm up to 15 mm. The method was also tested for stability and considerations for its implementation were discussed. The authors show that the technique is extremely precise, with measured error levels of <10%. Furthermore, the authors show that the response is highly dependant on temperature, concentration of the charge carrier present in the mixture, and also on the thickness of the polymer layer while layer porosity does not appear to have a significant impact on the measured response. While no change in response was observed for different linear flow rates the authors found that at extremely low flow rates, or during no-□ow conditions, the response was poor with a very low S/N ratio. Although the presented technique was demonstrated using thermally initiated polymerisation it could also be easily applied to photo-initiated polymerisation.

Acknowledgements

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