Rapid Fabrication of High-Quality Microfluidic Solid Phase Chromatography Columns: Supporting Information

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Device Fabrication

Multilayer devices were cast from two moulds fabricated using photolithography. Both moulds simultaneously hold control and flow channel features.

The first mould consisted of a multilayer structure having three different resist heights: Firstly, SU8-5 (MicroChem Corp., MA) was used to create column bypass channels. SU8-5 was spun at 1700rpm to a thickness of 5µm, and softbaked in three consecutive 1min long steps at 65 °C, 95 °C, and 65 °C. Following exposure for 5 s using an MA6 mask aligner (Karl Suss) at 42.5 mW/cm², a bake step was performed using the same three-step softbake protocol described above. Wafers were then immersed in SU8 developer (MicroChem Corp., MA) for 2 min. AZ50XT (AZ Electronic Materials, NJ), spun at 4100 rpm for 50 s, was then applied to create the column waste flow channels. Following prebake at 115 °C for 10 min, this layer was exposed for 45 s, developed in 33% AZ400K, and hardbaked at 190 °C for 2 h, resulting in 13 µm high rounded channels. Finally, chromatography columns were created using SU8-3025 spun at 650 rpm to a thickness of 100 µm. Following softbake at 65 °C, 95 °C, 65 °C for 10 min, 45 min, 2 min, respectively, wafers were exposed for 7 s and postbaked at the same temperatures for 3 min, 10 min, 3 min.

For the second wafer, a layer of SU8-3025 was spun at 2500 rpm for 30 s resulting in 30 µm high control channel features. The softbake was carried out at 65 °C, 95 °C, 65 °C for 2 min, 12 min, 2 min, respectively, followed by a 6.5 s exposure and a post exposure bake at the same temperatures for 1 min, 5 min, 1 min. A thicker layer of AZ50XT, spun at 500 rpm for 45 s, was then applied to create the bead inlet channels. This layer was prebaked at 115 °C for 10 min, exposed for 60 s, and hardbaked at 190 °C for 2 h, resulting in 55 µm channels with a rounded cross-section. After fabrication all wafers were coated with a 400 nm thick layer of Parylene C to facilitate release of the cured PDMS (1).

Microfluidic devices were fabricated by replica moulding from the microfabricated masters using a variation of the MSL process (2,3) wherein devices were assembled by oxygen plasma bonding of PDMS layers rather than off-ratio bonding. Briefly, PDMS prepolymer RTV 615 (General Electric, NY), mixed at a ratio of 10:1 (part A: part B) was used for each of the three layers of the microfluidic device. 60 g and 20 g of the uncured PDMS was degassed in a vacuum chamber for 1 h and cured on the first and second wafer, respectively, at 80 °C for 60 min. A featureless membrane was spun at 3000 rpm for 60 s and cured together with the two other layers. Following oxygen plasma treatment for 12 s at 600 mTorr the layers were aligned with the featureless membrane separating the channels of the two thicker PDMS layers. Interlayer connections were made using laser ablation as previously described (4). Finally, completely assembled devices were bonded to a microscope slide for mechanical support.
Figure S-1. The resin inlet of the microfluidic device mates with the Luer-Slip fitting of a 10 mL syringe (BD, NJ) containing a magnetic stir bar and the resin slurry. After flushing the microfluidic port, the syringe is pressurized with 30 psi (207 kPa) to force the slurry into the microfluidic device. During the packing process a rotating magnetic field generated by spinning rare earth magnets (Magcraft, VA) drives the stir bar which keeps the immobile phase in suspension.
**Optimization of Channel Dimensions**

*Figure S-2.* (a) The column packing time increases with the impedance of the bypass ($R_{bp}$) and waste flow channels ($R_w$). The locations of the three packing regimes as depicted in Figures 2b.1 through 2b.3 of the main text are indicated. (b) At 90% column fill factor, the aspect ratio of the void (width at the resin inlet : height) increases below a bypass impedance of $1 \times 10^{16}$ Pa s/m$^3$ and above a waste channel impedance of $1 \times 10^{12}$ Pa s/m$^3$. Inferior column uniformity and voids are expected for an aspect ratio of zero, i.e. the back of the column is clogged before the remainder of the column can be completely packed. For the simulation, column dimensions of 200 $\mu$m $\times$ 100 $\mu$m $\times$ 20 mm ($w \times h \times l$) are assumed with 260 equidistant bypass channels on either side of the column.
Curve Fitting

Figure S-3. Elution profiles of a 50bp FAM-labelled ssDNA fragment (blue) for four individual columns (a-d) produced at a reduced velocity of \( v = 6.3 \) (8 psi). A Gaussian function of the form \( f(x) = a \cdot e^{-\frac{1}{2}(\frac{x-b}{c})^2} + d \) was fitted to each of the four profiles using the Trust-Region method (red). The standard deviations and \( R^2 \) for each curve are 0.03 min and >0.97, respectively. The retention times in each column were determined to be 4.97 min, 4.99 min, 5.00 min, and 4.99 min, respectively.

References


