SHIFTING THE PERFORMANCE BARRIERS OF HPLC USING MICRO-FABRICATED CHROMATOGRAPHIC COLUMNS.

Gert Desmet[1], Wim De Malsche[1,2], Hamed Eghbali[1], Joris Vangelooven[1], David Clicq[1] and Han Gardeniers[2].
[1]Department of Chemical Engineering, Vrije Universiteit Brussel
[2] Research Programme Mesofluidics, MESA+ Institute for Nanotechnology, MESA+ Research institute, Enschede, The Netherlands
tel.: (+)32/ 2.629.32.51, fax (+)32/ 2.629.32.48, e-mail: gedesmet@vub.ac.be

The present contribution aims at illustrating and demonstrating how micro-machining technology can give a boost to High performance Liquid Chromatography (HPLC). Currently, HPLC is routinely used in nearly every chemical analysis lab. Despite its high degree of maturity, the technique however suffers from serious performance limitations when faced to the complex samples that need to be separated to solve the current state-of-the-art problems in the biological and pharmaceutical research (e.g., proteomics and metabolomics), the food and environmental analysis, etc…..

To cope with these highly complex samples, the recent years have witnessed a strong increase in research efforts focusing on the development of novel chromatographic supports, purposely designed to yield less flow resistance and less band broadening than the packing of porous spherical micro-beads that is currently used in HPLC columns. Many of the efforts in this direction can be categorized under the name "monolithic support columns", using advanced chemistries to in-situ synthesize flow-permeable skeletons of micro-porous material. Although yielding significantly smaller flow resistances, the band broadening in these columns is still too large, mainly because of the relatively broad and heterogeneous distribution of the flow-through pores.

To solve this heterogeneity problem, the present contribution will focus on the possibilities of advanced photolithographic etching techniques such as the Bosch-process to produce perfectly ordered porous support columns with optimized hydrodynamic shape and optimized external porosity. As the resolution of chromatographic separations is extremely sensitive to even the smallest heterogeneities of the flow-through pores of the packing, the gain in separation speed and resolution that can be achieved by working in perfectly ordered porous supports is really dramatic (easily up to a factor of 10 in separation speed).

The work presented here in fact aims at putting to work an original idea of Fred Regnier [1] wherein he advocated the use of micromachining to produce perfectly ordered chromatographic supports. Since then, the technological possibilities have clearly evolved and it is now possible to fabricate pillar array columns with a very tight (i.e., sub-micron) control of the pillar to pillar distances and the pillar dimensions.

At the conference, we will demonstrate the possibility of rapid multi-component separations and the possibility to achieve very low plate heights with a microfabricated column in pressure-driven LC. In addition, we will also report on the possibility to apply the shear-driven flow principle [2] to produce low-dispersion flows through nano-channels filled with ordered micro-pillar arrays.

References

[1] B. He, N. Tait and F.E. Regnier, Analytical Chemistry, 70 (1998) 3790.
[2] Clicq, D, Pappaert, K., Vankrunkelsven, S., Vervoort, N., Baron, G. V. and Desmet, G., 2004, Shear-driven Flows: A New Approach to LC and Macro-molecular Separations, Anal. Chem. 76(23); 430A-439A.

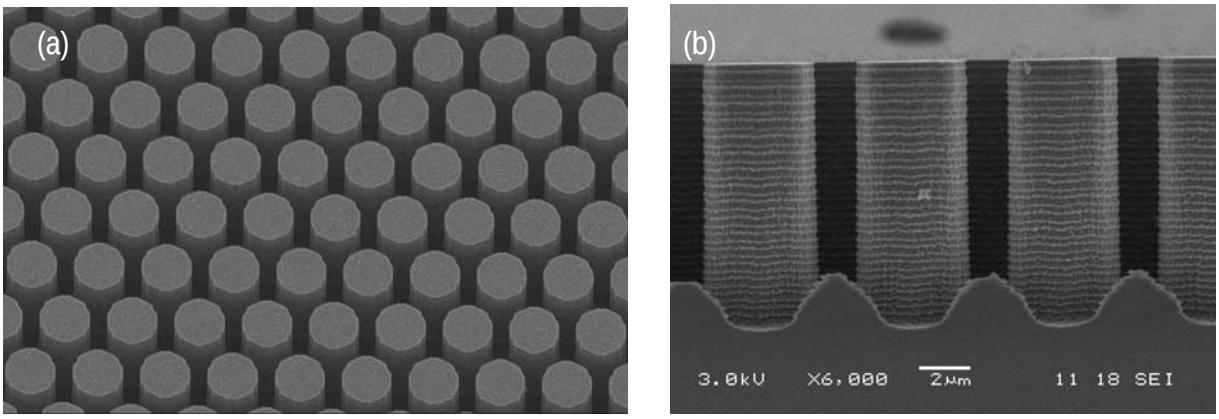

Figure 1. SEM picture of a detail of the micro-pillar packing. (a) Bird's eye view and (b) front view of pillars after cutting the chip.

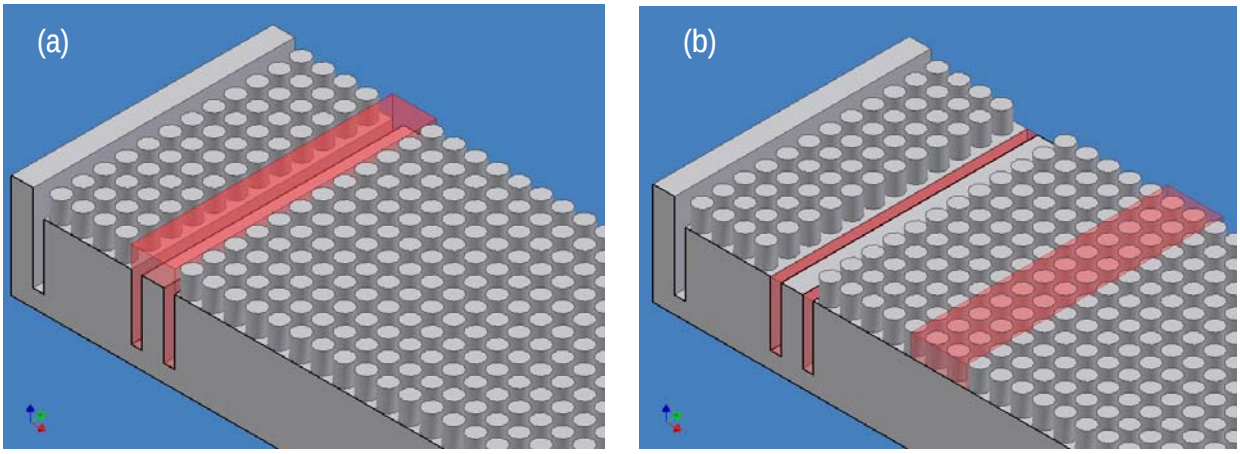

Figure 2. Schematic representation of the employed on-chip detection scheme. (a) Sample loading step and (b) elution step feeding the mobile phase from the top of the column.

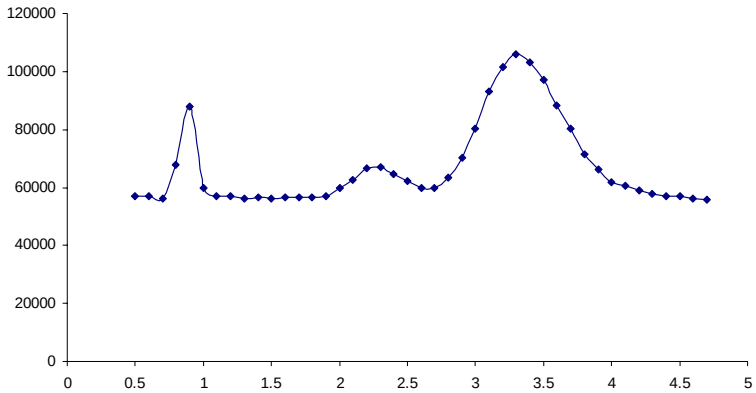

Figure 3. Breakthrough profile of a three component separation at L=1 cm. Sample components= Coumarin 440, 460 and 480. Mobile phase = 30v%/70v% MeOH/water.