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CHAPTER 4

A Pressure Driven Injection System for an Ultra-Flat Chromatographic Microchannel

A pressure-actuated on-chip injection system has been developed that is compatible with shallow microchannels of a very large aspect ratio i.e. 1 µm deep and up to 1000 µm wide. Such channels offer potential advantage in the miniaturization of liquid chromatography and other separation methods as they allow high loadability and low sample dispersion at the same time. Computational fluid dynamics simulations were performed to predict the flow profiles and the transport of a sample in the system and to justify the injection principle. Based on these simulations, a prototype injector integrated into a chip for hydrodynamic chromatography has been realized and tested experimentally. The performance of the device is satisfactory and the results are in qualitative agreement with the numerical models.

Introduction

Reproducible injection of a small, well-defined sample zone is a necessary condition for a good performance of any analytical separation device. Especially when such a device is miniaturized into a chip this is a challenging problem. Use of an external injector would in most cases prevent efficient separation because of large zone dispersion in the connection tubing. Integration of mechanical components such as a microvalve or a plunger into a chip often makes the device too complex and expensive. Therefore, passive injector microstructures are preferred, either a so-called cross injector,\textsuperscript{1,2} where the sample delivery channel intersects the separation channel at one position, or a twin-T injector,\textsuperscript{3,4,5} where both channels share a common, short length. This length defines the injection volume. In the majority of cases described in literature both channels have a small aspect ratio (length/width < 10) and an equal depth as a result of a single etching step. Recently, some interesting applications of large-aspect-ratio (shallow but wide) microchannels have been presented.\textsuperscript{6,7,8,9} These channels both allow higher sample loadability (feasible for the detection) and have low peak dispersion because the
nonequilibrium mass transfer over the small channel thickness is fast. In such systems the above mentioned passive injection principles are in essence applicable. However, during the filling the sample can leak considerably into the separation channel because of the large width it has to cross. This would result in peak deformation, overloading and poor separation. In electrodriven devices this could be prevented by so-called pinching,\textsuperscript{10} applying additional voltages at specific positions. An analogous pinching scheme for pressure driven systems is difficult to implement because this would require exact control over the pressure at all the outlets.

Moreover, although the electrodriven approach is probably the most easily applicable one in micro Total Analysis Systems (\(\mu\)TAS), pressure-driven devices offer advantages in many special cases. Notably, when organic solvents are used as carrier liquid, electroosmosis is often difficult to generate or control. Different electrophoretic mobilities or buffer compositions can complicate the injection also in aqueous solutions. Another important application field for pressure driven separation systems is on-line industrial process monitoring where an often wet or explosive environment is at hand and the use of high-voltage electrodriven methods presents a safety risk.

In this work, we show a form of a twin-T injector based on a careful choice of flow resistances inside a chip combined with an external flow actuation. The injector was implemented in a chip\textsuperscript{9} for hydrodynamic chromatography\textsuperscript{11,12} (HDC), where a shallow channel is required not only to preserve efficiency but also for the separation principle itself. This analytical size-separation technique uses the effect of the nonuniform flow profile on the transport of large analytes, such as polymers or particles through a relatively narrow (in at least one dimension) conduit. Differential migration of species with respect to the size is achieved without a stationary phase or an external field. However, a chip of similar parameters (if modified with a stationary-phase moiety) and with the same injector would be suitable for on-chip liquid chromatography of small molecules.

Design of the system involves modeling of flow and species transport. The simplest approach in fluidics is a lumped element model, where a fluidic system is treated as a group of interconnected resistances and capacitors for which average fluxes and pressure drops are obtained from expressions analogous to electric circuits.\textsuperscript{13} Such a model is efficient in complex systems but can neglect some important phenomena, especially in the case of
species transport. In less complicated systems, another approach is a direct, full or partial numerical simulation of flow and other transport phenomena using computational fluid dynamics (CFD).\textsuperscript{14} Recent advances in computer performance allow studying many cases to higher detail and precision in 3D and in acceptable time.\textsuperscript{14,15} However, the computational domain is still limited in size (relative to its smallest dimension to be fully accounted for) by the maximum possible number of discretization elements given by the computer memory and time available.

A combined approach was used here. The overall injection scheme was developed based on a flow resistances model. Subsequently 2D- and later 3D-CFD simulations were performed of the central part of the injector in order to predict in detail the flow profiles and the transport of the sample with its implications for the actual amount injected and injection zone broadening. The presented CFD study is unusual because of the very large aspect ratio of the domain.

A prototype injection system was fabricated and tested. The experimental results show the same features as the CFD predictions. While 2D-CFD already gives a reasonable estimate for an ideal injector, 3D simulations are necessary to verify the conclusions and, in the case of the simplified prototype, to describe the observed initial flow non-uniformity. In separation experiments,\textsuperscript{9} reliability of the injection was shown.

**System Design**

The requirements on the injector for the HDC chip follow the desired parameters of the separation channel, which were obtained through combining chromatographic rules\textsuperscript{9} and technological limits.\textsuperscript{16} The suitable separation channel dimensions were: depth $\sim 1$ $\mu$m, width $\sim 500$–$1000$ $\mu$m and length $\sim 10$ cm. Flow velocities should be $\sim 0.1$–$1$ mm s$^{-1}$, corresponding to a working pressure $\sim 1$–$10$ bar. The injection space will be defined in the short initial part of this channel.

In order to assure that enough sample is injected and at the same time the injection contribution to peak broadening is only $\sim 5\%,$ the injection should produce a rectangular sample plug (or a symmetric peak of equal standard deviation $\sigma$) of length $l,$ given by\textsuperscript{17}
\[ l^* \approx \frac{L_s}{\sqrt{N}} \]  

(1)

where \( L_s \) is the length of the separation channel and \( N \) the maximum number of theoretical plates for this channel. Assuming the above mentioned dimensions and providing that the maximum separation efficiencies for this channel, i.e. 100 000–300 000 theoretical plates,\(^9\) can be achieved, \( l^* \) should be \( \sim 150–300 \, \mu \text{m} \) (implying \( \sigma^*_l \sim 50–100 \, \mu \text{m} \)).

**Figure 1:** (a) The layout of the prototype injection system. The flow direction and valve positions in the injection steps are shown. (b) The photograph of the injection area on the prototype HDC chip. (Resistance \( R_2 \) is not used). The central part (in the frame) was studied in detail.
However, flow splitting will be necessary to avoid peak tailing, and therefore the length of the injection space, \( L_i \), should be larger than \( l_i^* \), because only a part of its content will be injected. For the prototype \( L_i = 300 \mu m \) was chosen and the actual length injected, \( l_i \), will be investigated.

The injection can be realized in 3 steps, using a combination of external flow actuation and internal chip resistances in a system depicted in Figure 1a. The valves A and B are switched simultaneously. In the macrofilling step, fresh sample is introduced directly from the syringe through the valve B into the collector C and thus deposited inside the chip close to the microinjector (Figure 1b). The collector is made as a larger groove of a few \( \mu L \) volume, having a low resistance in order to ease its purge. During the next microfilling step, pressure is applied to this collector to fill the injection space \( R_i \). Direct filling of this part from a syringe would be very difficult because of the auxiliary resistance \( R_R \) (explained below), and the large volume of the connecting tubing to be flushed with the sample. In the final microinjection step, a large part of the sample located in \( R_i \) is transported into the separation channel \( R_S \) by the carrier liquid entering from slit \( S_1 \) through \( R_0 \). The remainder of the sample is "swept" back into the slits \( S_2 \) and \( S_3 \).

In order to avoid complex fabrication procedures, the delivery slits are made as open grooves in the bottom of the channel, entering it from the same side (Figure 1b). Uniform filling can be achieved with such transversal conduits only if their flow resistance is much smaller than that of the shallow channels. In this way, pressure gradients and consequently flow non-uniformities over the channel width are prevented in the shallow part. The fluidic resistance \( R \) of a rectangular conduit is equal to\(^\text{18}\)

\[
R = \frac{\Delta p}{F} = K \frac{\eta L}{ab^3}
\]

where \( \Delta p \) is the pressure drop and \( F \) the volumetric flow rate, \( a \) and \( b \) are the conduit cross-sectional dimensions assigned so that \( a \geq b \), \( L \) is the conduit length, \( \eta \) is the fluid dynamic viscosity and \( K \) a geometrical factor. For channels of large aspect ratio \( K \sim 12 \), for lower aspect ratios \( K \) depends on \( a/b \). Specifically, for \( a/b = 2 \) (as considered below) is \( K = 17.49 \).
In order to restrict the sample to the space between slits S2 and S3 during the microfilling, additional conditions \( R_1 \ll R_5 \) and \( R_0 \gg R_2 \) are imposed. These are satisfied because of the vast difference in the lengths of the corresponding channels (Figure 1b) and because of the shallow depth.

The already mentioned flow splitting has to be implemented at slits S2 and S3 to reduce the convective and diffusive carry-over of the remaining sample from the slits after the microinjection step. The cross-section of the slits should not be too large as this would slow down this cleaning process and also the micro-filling step. The fabrication method easily allows creation of a groove 10 \( \mu m \) wide and 20 \( \mu m \) deep. As will be shown by the simulations, the low-resistance vs. small-volume trade-off is acceptable with this slit size for the 1 \( \mu m \times 500 \mu m \) channels.

As a compromise between flow loss and tailing prevention, a flow splitting ratio 1:1 was adopted in the model for both slits. This would ideally result into an injected length \( l_i = 0.5 L_i \). However, because broader slits have to be used due to the required low resistance and because of microtechnology limits, some carry-over will occur, and \( l_i > 0.5 L_i \) should be expected.

In the real device the flow splitting 1:1 is easily achieved for slit S3 by incorporating an auxiliary channel (R_R in Figure 1b) of the same flow resistance as the separation channel. However, its volume has to be much smaller in order to avoid long injection times. As can be seen from eq. 2, the resistance of a large-aspect-ratio channel remains constant if its width \( (a) \) and length \( (L) \) are reduced by the same factor, while keeping the depth \( (b) \) unchanged. Fast injection is then possible providing the consequent high linear velocity and shearing in the resistance channel is not degrading the species to be analyzed. For the prototype the resistance channel is 20 times smaller in the length and width than the separation channel.

To keep the prototype simple and less prone to possible clogging we have chosen to control the outflow from slit S2 by a macrovalve outside the chip (valve A) instead of implementing resistance \( R_2 \). Compliance induced outflow then occurs shortly, due to the compressibility of the large volume of liquid in the tubing compared to the slit volume. This results in some flow nonuniformity in the start of the microinjection but still assures cleaning and (delayed) closing of this slit.
Using gas-pressurized vessels with the carrier liquid offers a stable flow and no contamination with particles from pump seals. It is therefore preferable for the 1 μm deep channel, where a working pressure below 10 bar is sufficient for efficient HDC separations. However, the external valve system is also suitable for direct connection to an LC pump (with split) if more pressure is required, for example for sub-micrometer channels. A clamped stainless steel system with chemically resistant O-rings was chosen for the external part to allow use of also organic solvents. The silicon and glass based chip is resistant to all chemicals except strong bases and HF solutions.

**Materials and Methods**

**CFD model**

As a part of the design, finite volume 2D- and 3D-CFD models were built for the central part of the injection system (Figure 1) using the FLUENT™ software package (version 5.5.14). The computation domain geometry (Figure 2) was created and meshed in the default pre-processor GAMBIT™ (version 1.3.0). The numerical simulations were performed on a PC platform.

The dimensions of the domains are equal to the values chosen in the preceding section. The 2D domain also contains a longer part of the separation channel to accommodate the whole injected zone and inspect its spatial profile. In 3D such an extension would result into an excessive number of computation elements. Therefore, the injected amount is obtained by monitoring the average analyte concentration on the boundary OUT at each time step. Still a large number of cells is necessary (~ 200 000) because of the large aspect ratios of the domain. A non-uniform grid with wide, thin cells has to be used in the shallow parts to keep the number of cells limited. Such cells generally lead to numerical problems and inaccuracy but could be used in flat conduits in regions where gradients are small. A time step of $10^{-3}$ sec was used in all species transport simulations.

Higher order numerical schemes and double precision were used as recommended for large aspects ratio problems involving long thin conduits. The solutions were found to be practically independent on the spatial and temporal discretization with the grids and time
step used. The amount of analyte, as reported by integration on the boundaries, corresponds to the amount introduced to the domain within \( \sim 1\% \).

**Figure 2:** (a) The geometry of the computation domain in the 2D- and 3D-CFD models of the central part of the injector. (b) The detail of the computation mesh in the 3D model.

All flows are pressure driven, laminar \((\text{Re} \sim 10^3-10^4)\) and modeled as stationary. The flow calculations and species transport calculations were decoupled. First, Navier-Stokes equations were solved for the carrier liquid to obtain the flow field. This field was then fixed and the time-dependent solution of the convection-diffusion equation was performed for an analyte that was specified by a diffusion coefficient and the initial concentration given by the boundary condition.

As another boundary condition, the velocity on each opening is specified. In the microfilling step, the velocities on IN and OUT are set to zero. This reflects the conditions
\( R_S \gg R_i \) and \( R_0 \gg R_2 \). Velocities on S2, S3 are then equal but opposite in signs and determined by the flow rate generated on resistance \( R_R \) (\( \gg R_i \)) by the applied pressure (optional \( R_2 \) is not considered). For the microinjection, first the velocity on OUT is determined from the applied pressure and the separation channel resistance \( R_S \). The remaining velocities are obtained from the continuity and splitting conditions and from the cross-sectional areas of the boundaries.

The carrier liquid is water at 293 K and the sample is a mixture of water and "analyte" with a diffusion coefficient \( D = 5 \times 10^{-10} \text{ m}^2\text{s}^{-1} \) or \( D = 10^{-11} \text{ m}^2\text{s}^{-1} \). For the assumed pressure of 2 bar the velocity in S3 is 0.5 mm s\(^{-1}\) in the microfilling step for the 3D model. The same pressure yields a velocity of - 0.2 mm s\(^{-1}\) on OUT during the microinjection step. In the species simulation, a constant mass fraction 0.001 of analyte in the carrier liquid is supposed in boundary S3 in the microfilling step. For the microinjection, the same mass fraction is uniformly applied as an initial profile in the slits S2 and S3 and everywhere in between them.

**Experimental**

In the prototype setup silicon-glass chips were used containing a separation channel that was 1 \( \mu \text{m} \) deep, 0.5 mm or 1 mm wide and 80 mm long. The shallow channels were created by wet silicon oxide etching of a thermally oxidized silicon wafer. The inlet and outlet slits were defined by Reactive Ion Etching. Inlet and outlet holes of 0.5 mm diameter were created in a CMP-polished 4\" Pyrex glass wafer by powderblasting. The wafer pair was fusion bonded and subsequently annealed at 425 °C. Details of the chip fabrication can be found elsewhere.\(^{16}\) A clamp with O-rings was used as the interface between the chip and the valve system. The injection process was monitored for a fluorescent sample using an upright fluorescence imaging microscope (BX51) equipped with a CCD camera (Colorview 12) and a computer image acquisition and processing system (Olympus). The internal microscope mercury lamp served as the illumination source. All tubing and valves used were stainless steel standard HPLC 1/16\" components (Valco).

Aqueous phosphate buffer (10 mM pH = 7) was used as the eluent and the solvent of the sample. Solutions of fluorescein (0.01 mg/mL) or fluorescent latex particles (180 nm, 0.2 mg/mL) obtained from Molecular Probes (Leiden, The Netherlands) were used as test
samples. All water was deionized and subboil-distilled. The buffer was filtered prior to use through a 0.02 μm membrane filter. Upstream each reservoir, a 0.5 μm in-line filter (Upchurch, USA) was placed. The working pressure was 2 bar.

Results and Discussion

After preliminary 2D-CFD studies, which demonstrated the injection principle, a 3D model was built to prove the feasibility of the design. The flow simulation confirms the assumption that, despite the direction of the flow in the slits, a uniform velocity profile parallel with the direction of the separation channel is obtained in the shallow part (Figure 3).

Figure 3: The flow simulation in the 3D model. Top view (above) and detailed view in slit S3 (below) of the velocity field for the microfilling (left) and the microinjection (right). Only selected vectors in a single plane are plotted.
Figure 4: The sample transport simulation. Microfilling step (left). Microinjection step (right) in an injector with ideal split control and ideal filling.

The subsequent simulations of the species transport (Figure 4) show that the microfilling step can fill the whole injector space within a few seconds. It also shows that the uniformity of an (ideally) filled zone is preserved in the microinjection step.

An HDC-chip prototype injection system of the same dimensions, however without the control of the splitting on slit S2, was fabricated. Figure 5 shows an experiment where an aqueous solution of fluorescein \((D = 5 \times 10^{-10} \text{ m}^2\text{s}^{-1})\) is injected. The displayed time is measured from the moment when the sample front visibly enters the domain from slit S3. Qualitative agreement with the numerical simulations was obtained for the microfilling step (Figs. 4 and 5 left). A detailed comparison is difficult because the simulation assumes a step increase in the sample concentration in the inlet at the start of the filling process. In practice this is a ramp increase, as the front of the sample undergoes dispersion when it moves from collector C to the microinjector. This takes ~10–20 seconds in the prototype because long
interconnecting grooves were used to allow easy access for the microscope objective to the injection structure in order to inspect the species transport. The internal loop is therefore less distinctly filled, as compared to the simulation. In a more advanced device with an integrated detector these grooves will be shortened.

**Figure 5:** The sample inject experiment. The injection is demonstrated on an HDC chip prototype. Fluorescein solution is used as a “sample”, the working pressure is 2 bar. Microfilling (left) and microinjection (right).

In the microinjection step simulation, a rectangular species zone was applied into the injector instead of using the profile obtained for the microfilling. Although this does not represent the real injection properly, the net effect of this step on the zone is better visible. It is also useful not to relate the microinjection to a specifically deformed initial shape. This can be modified e.g. if a higher pressure is used for the microfilling than for the
microinjection (where the pressure is prescribed by the desired optimum velocity in the separation channel) to speed up the process and reduce the diffusive overfilling.

A more apparent difference (Figs 4 vs. 5 right) is the "J" shaped rear end of the zone, which occurs in the experiment. This can be explained by a flow non-uniformity in the shallow part of the injector space, caused, as already mentioned, by the macrovalve actuation of slit S2. For a short time, a relatively high, compression-induced outflow develops in this slit, as the working pressure is opposed only by \( R_0 \). This flow disturbs the uniform velocity field in the adjacent shallow part. To study this effect we have performed additional simulations. The volume of liquid between slit S2 and valve A is \( \sim 25 \mu \text{L} \). Assuming the isothermal compressibility of water to be \( 5 \times 10^{-5} \text{ bar}^{-1} \), the void volume will be \( \sim 2.5 \text{ nL} \) at 2 bar pressure. We consider now an average stationary flow, corresponding to an intermediate situation, when the liquid in the tubing is already partly compressed and has a pressure of 1 bar, while the applied pressure is, as before, 2 bar. The pressure difference over \( R_0 \) is then 1 bar. (The actual outflow decays as the compression proceeds because the pressure difference over \( R_0 \) decreases). This results into an average linear velocity of 8 mm s\(^{-1}\) on boundary IN. The pressure drop over \( R_S \) is 1 bar and gives rise to a velocity of \(-0.1 \text{ mm s}^{-1}\) on OUT. Due to the flow splitting on S3 and because of continuity, the velocity on boundary S2 will be \(-19.5 \text{ mm s}^{-1}\). With this average outflow the compressibility compliance would be filled in \( \sim 0.5 \text{ sec} \). The flow and resulting species profile are solved for those conditions. Then the flow into slit S2 stops, the flow in R becomes uniform and the injection proceeds normally. This is modeled by fixing the obtained species profile, solving the flow for the new boundary conditions and then again solving the species transport.

Although the decay-dynamics of the process is averaged, this simple model does explain the effect. The flow profile deformation is clearly visible (Figure 6 left) and the sample transport solution (Figure 6 right) shows the same feature as the experiment (Figure 5 at 4.4 s).
Figure 6: The simulation of the effect of an initially high outflow in slit S2 if actuated with a microvalve. The resulting nonuniform flow field (left), applied for 500 ms, partly deforms the injection plug (right).

From the monitored concentration profile (Figure 7, curve E) it appears that the injected amount, according to this simple model, is smaller than ideal. Still, enough sample is injected and the initial zone deformation is partly compensated by faster cleaning and elimination of diffusion from slit S2.

In principle it is possible to accurately model the whole injection process as a sequence of flow and species transport calculations with changing boundary conditions to account for the effect of both over-filling (with a slightly larger domain) and the disturbed flow on the injected peak. One can also include time dependent boundary conditions to represent the ramp of concentration, or a non-stationary flow field coupled with the species calculation for the non-ideal injector. More computer power would be needed to do such calculations in a reasonable time. However, because all those effects can likely be suppressed by modification of the design and operating conditions, such detailed simulations are not necessary in our opinion.

The simulated concentration profiles in time are plotted in Figure 7 for both the 2D and 3D models.
Figure 7: The calculated profiles in time of an average mass fraction of analyte at the boundary OUT during the microinjection. Simulation with ideal splitting conditions in 2D for the fast- and the slowly diffusing analyte (A, B respectively) and the same simulation in 3D (C, D respectively). (E) the profile obtained for the fast diffusing analyte in the 3D-model with initial non-uniform flow. (---) a hypothetical zone with $l_i = 0.5 \ L_t$.

The asymmetry of the curves for the fast diffusing compounds is mainly the effect of monitoring the concentration at a fixed position over the time span, during which the actual profile is still developing. Interestingly, the difference between the 2D and 3D simulations for an injector with an ideally controlled splitting is not large, despite the different orientation of the flows in the slits. This allows comparing of the more informative, spatial profiles after injection into the longer part of the separation channel in the 2D domain (Figure 8). Some tailing due to carry-over from the slits is observed but is not significant in our application. For the same reason, 2D simulations can be used to quickly explore the influence of different flow conditions providing those would still result into a uniform flow in the actual device. These simulations (Figure 8) suggest that moderately higher flow rates or higher splitting ratios (higher flow into the slits) can further improve the peak symmetry if necessary.
Figure 8: The injected sample peak shapes in the beginning part of the separation channel, obtained from the 2D simulation. Normal flow (−0.2 mm s⁻¹ at OUT) and splitting (1:1) for the fast (A) and the slowly diffusing analyte (B). Moderately higher flows (C, −0.4 mm s⁻¹ at OUT) and splitting ratios (D, slit/channel 2:1, −0.2 mm s⁻¹ at OUT) further improve the zone shapes for the same analyte as in (A). (---) a hypothetical zone with \( l_i = 0.5 \) \( L_i \).

The amount of injected species \( (m_i) \) can be obtained by integrating the concentration profiles from the simulation, and, in the experiment, by integrating the image intensities. The values can be compared to the amount \( M_i \) located in the shallow part between slits S2 and S3 in the ideally filled injector. Because the spatial profiles of the injected peaks are close to either a rectangular or a Gaussian shape with the maximum equal to the initial concentration, the comparison holds also for the injected lengths (represented by a virtual rectangular plug of the same standard deviation), i.e. \( l_i / L_i \sim m_i / M_i \). In the limiting case with splitting 1:1 and no carry-over from the slits, \( m_i / M_i = 0.5 \). Because of the broader slits, the carry-over cannot be suppressed completely and more sample is injected, both in the CFD model and the experiment. From the CFD simulation (when ideal filling and ideal split control is supposed) \( m_i / M_i \sim 0.65 \) for the slower- and \( \sim 0.85 \) for the faster-diffusing species. In the case of the non-ideal injector (when the above mentioned specific flow conditions are considered), a ratio \( \sim 0.75 \) is obtained with the faster diffusing species.

In experiments, \( m_i / M_i \sim 0.8-1.2 \) for fluorescein and similar injected amounts were observed for the particles. Zones with \( \sigma_i \sim 100 \) µm (i.e. \( l_i \sim 300 \) µm) are typically obtained.
These experimental results on the prototype are, however, inaccurate because of the limitations of the imaging (integration time 100–500 ms) and timing of the manually controlled valves. The latter leads to partial overfilling of the injector. Nevertheless, the zones satisfy the chromatographic criteria. Further reducing the microinjector length to ~150–200 μm would increase the robustness because of more tolerance to possible overfilling.

Separation experiments on the HDC chips have proved that the injection method is suitable for a large range of analytes with different sizes and also in 1 mm wide channels. In experiments with non-filtered latex particles, some pollution from particle clusters has built up in the slits, but injection was still possible (Figure 9).

![Figure 9: Injection of latex microparticles (180 nm diameter) into a 1 mm wide HDC channel.](image)

Long interconnection grooves in the prototype result in slight HDC frontal separation and thus discrimination during the microfilling step. This should be accounted for in the timing of the valves. Shortening of the grooves will eliminate this effect.

Organic carrier liquids (methanol, tetrahydrofuran) used over several weeks in other test experiments have not affected the device either. The injector was also implemented with minor modifications into test prototypes for a microviscodetector.

Further substantial miniaturization and simplification of the presented injection system can be suggested by using sample and carrier liquid reservoirs (10's of μL) created directly on the chip and actuated by gas pressure via gas valves and manifolds, miniaturized in a
compact block clamped above the reservoirs. A small tank or a piston gas pump could serve as a pressure source.

**Conclusions**

The presented prototype injection system shows good performance with wide shallow separation microchannels such as those used in the tested chip for hydrodynamic chromatography. In the design, simulations by computational fluid dynamics were possible despite the large aspect ratio, and provided useful information. This injector can be used in other similar flat channel systems with both organic and inorganic liquids.

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