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REVIEW

Polymer microfluidic devices

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Abstract

Since the introduction of lab-on-a-chip devices in the early 1990s, glass has been the dominant substrate material for their fabrication (J. Chromatogr. 593 (1992) 253; Science 261 (1993) 895). This is primarily driven by the fact that fabrication methods were well established by the semiconductor industry, and surface properties and derivatization methods were well characterized and developed by the chromatography industry among others. Several material properties of glass make it a very attractive material for use in microfluidic systems; however, the cost of producing systems in glass is driving commercial producers to seek other materials. Commercial manufacturers of microfluidic devices see many benefits in employing plastics that include reduced cost and simplified manufacturing procedures, particularly when compared to glass and silicon. An additional benefit that is extremely attractive is the wide range of available plastic materials which allows the manufacturer to choose materials' properties suitable for their specific application. In this article, we present a review of polymer-based microfluidic systems including their material properties, fabrication methods, device applications, and finally an analysis of the market that drives their development.

Keywords: Microfabrication; Polymer; Microfluidics; Chip

1. Materials properties

In microfluidic applications [1,2], properties of the material that may be of fundamental importance include machinability, surface charge, molecular adsorption, electroosmotic flow mobility, optical properties, and many others. When choosing a polymer-based substrate, the properties of the material are critical for both the fabrication process and the successful application of the device.

1.1. Materials properties and fabrication

For many applications in the literature, plastics rather than pure polymers, are used to fabricate microfluidic devices. Plastics can contain a num-

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ber of additives that impact their processing and shelf life and should be considered. These may include fillers (e.g. mica, talc, calcium carbonate), plasticizers (e.g. dioctyl phthalate in PVC), heat stabilizers (e.g. organo-tin compounds in PVC), antioxidants (e.g. phenols, amines), and UV stabilizers (e.g. benzophenones, salicylates).

Different fabrication protocols have different and very specific constraints with regard to the properties of the material. For instance, with hot embossing and injection molding methods, the glass transition temperature, melt temperature, and thermal expansion coefficient are some of the most critical parameters for successful fabrication. The glass transition temperature is the temperature range where the polymer substrate changes from a rigid glassy material to a soft (not melted) material, and is usually measured in terms of the stiffness, or modulus. The degree to which a thermoplastic material softens, its stiffness, is dependent on the crystallinity. The melt temperature is the temperature at which the polymer flows and is generally much higher than the glass transition temperature. Crosslinked polymers and thermoplastics that contain very long polymer chains with strong intermolecular attractions do not melt and flow but remain soft until they decompose. Finally, the thermal expansion coefficient refers to a change in length or volume resulting from a specified change in temperature. This parameter is not only important in several fabrication processes, but also in the sealing process where different materials are thermally bonded.

In room temperature imprinting or stamping methods, hardness is an important parameter. This parameter can be measured in terms of an indentation that is produced in a material due to a specified applied force. For molding by soft lithography, elasticity, or the ability of the polymer to retain original shape after deformation, and shelf life are important properties to consider. Table 1 provides a list of some common polymer-based materials and properties associated with each material that are critical for fabrication [3–5]. Common microfluidic channel fabrication protocols and materials' issues associated with each method will be discussed in greater detail in the following section.

1.2. Materials properties and application

Electroosmotic pumping is the most common method used to propagate flow in microfluidic systems. In electroosmotically driven systems, it is critical that the substrate material exhibit good electrical insulating properties so that the electric field will drop across the fluid-filled channel and not through the substrate. This effect is evaluated by several parameters including the dielectric strength, and electrical resistance. The dielectric strength is the voltage that can be applied across an insulator before breakdown occurs. The dielectric strengths of several common polymers are given in Table 1.

A second consideration when using electrically driven pumping is heating. It is well established that Joule heating can be substantial in systems employing electroosmotic flow. If heat is not effectively dissipated in microchannel systems, elevated local temperatures can dramatically impact the efficiency of chemical separations, and induce solution degassing and eventually boiling. With plastics having a low melt temperature, high localized heating can also cause significant channel deformation. Therefore, heat dissipation in the substrate material is a very important consideration when electroosmotic pumping is utilized. Heat dissipation is characterized by the thermal conductivity of the material. The thermal conductivities of several polymers are also given in Table 1. For comparison, fused quartz has a higher thermal conductivity (33×10^{-4} g cal cm s^{-1} cm^{-2} °C [6]), and much higher melting point (1665 °C) than most plastics.

A third consideration when using electrically driven pumping is microchannel charge. Electroosmotic flow (EOF) is generated by the surface charge on the microchannel walls in combination with an electric field along the microchannel. Because polymer materials exhibit a wide range of charge and charge densities, EOF in microchannels made from different polymer materials is highly variable. EOF has been measured in various polymer microchannels fabricated by laser ablation [7], and imprinting [8], and in poly(methylmethacrylate) (PMMA) channels fabricated by LIGA methods [9]. These measurements

Table 1
Material properties

	PMMA	PC, high viscosity	PETG, poly-ethylene tere-phthalate glycol	PE, poly-ethylene, branched homo-polymer	Polyimide	Styrene copolymer	Silicone
Melt Flow (g/10 min)	1.4–2.7	3–10		0.25–27.0	4.5–7.5	1.4	
Melt T (°C)	85–105	150	81	98–115	388	100–200	
Mold (linear) shrinkage	0.001–0.004	0.005–0.007	0.002–0.005	0.015–0.05	0.0083	0.003–0.005	0.0–0.006
Process T, (°C) njection	163–260	294	249	149–232	390–393	182–288	
Molding P, 10^7 (Pa)	3.4–13.8	6.9–13.8	0.69–13.8	3.5–10.0	2.0–13.8	3.5–13.8	
Hardness, Rockwell	M68–105	M70–M75	R106		E53–99, R129, M95	M80, R83, 75	
Coef. Linear thermal expansion, 10^{-6} (°C)	50–90	68		100–220	45–56	65–68	10–19
Thermal conductivity, 10^{-4} g cal-cm s^{-1} cm^{-2} °C	4.0–6.0	4.7		8	2.3–4.2	3.0	3.5–7.5
Dielectric strength, 0.003175 m specimen, MV m^{-1}	16–20	15–16		18–39	16–22	17	16–22
Vendors	American Polymers, AtoHaas, Continental Acrylics, Cyro, DuPont, ICI Acrylics, LG Chemical, Network Polymers, Plaskolite, RTP	Albis, American Polymers, Ashley Polymers, Bayer Corp., Dow Plastics, Ferderal Plastics, GE Plastics, MRC, Network Polymers, Polymers Int'l, Polymer Resources, RTP, Shuman	Eastman	American Polymers, Bamberger Polymers, Chevron, Dow Plastics, DuPont, Eastoman, Equistar, Exxon, Huntsman, Mobil, Network Polymers, MOVA Chemicals, RSG Polymers, A. Schulman, Union Carbide, Wash. Penn, Westlake	Ciba Specialty Chemicals, DuPont, Mitsui Chemicals, Solutia	Albis, American Polymers, BASF, Bamberger Polymers, Bayer Corp., Dow Plastics, Federal Plastics, Huntsman, LG Chemical, Network Polymers, RSG Polymers, A. Schulman	Bayer Corp., Dow Corning, Emerson and Cuming, GE Silicones

indicate that the fabrication method, as well as the material itself, can affect the surface charge density and therefore can have a profound effect on the EOF. For instance, laser ablated channels support higher EOF than channels that have been hot embossed in a similar material due to the fact that reactive species are incorporated into the microchannel surface during the ablation process [10]. The location of charged groups on PMMA substrates was also determined to be dependent on the fabrication procedure; for example, room temperature imprinted channels can have highly charged walls while hot embossed channels fabricated in a similar material have a low surface charge density on the walls [11]. Finally, surface charge and EOF can also be modulated in polyethyleneterephthalate (PETG) by alkaline hydrolysis [12,13] and in polydimethylsiloxane (PDMS) by plasma treatment [14]. Charge density, charge location, and electroosmotic flow can therefore be controlled by several parameters including (1) choice of polymer material, (2) fabrication protocol, and (3) various surface treatments.

Other material parameters that are critical to successful application of microfluidic devices include auto-fluorescence (when using optical detection), permeability (when using living cells), chemical resistance (when using non-aqueous solutions), and analyte adsorption. Table 2 lists the chemical resistances of several common polymeric materials [15]. Analyte adsorption is a parameter that is highly dependent on several material characteristics including hydrophobicity and surface charge. The biocompatibility of many plastic materials is associated with both of these parameters and has been evaluated and characterized extensively in the biomedical engineering literature.

1.3. Surface modification of plastic microchannels

Very few methods for chemically modifying plastic microfluidic channels have been reported in the literature. Henry et al. have reported on the covalent modification of PMMA channels to impart an amine functionality [9]. The amine group can be further derivatized to create a variety of stable surface chemistries on PMMA channels. At NIST, we have used a non-specific coating method, polyelectrolyte multilayer deposition, to treat plastic microchannels [16]. This coating method is generic and stable for a variety of plastic materials provided that the material has a significant surface charge to promote electrostatic interactions between the polyelectrolyte and the surface. Others have reported the non-specific coating of plastic microchannels with proteins for biochemical assays [17]. Non-specific adsorption

Table 2
Chemical resistance of common polymer substrates

	Methyl-methacrylate	Poly-carbonate	Poly-ester (styrene alkyd)	Poly-styrene	Poly-vinylchloride	Silicones
Mineral Acids						
Weak	Good	Excellent	Good	Excellent	Excellent	Fair–good
Strong	Fair–poor	Fair	Poor	Excellent	Good–excellent	Poor–good
Oxidizing Acids	Attacked	–	Poor	Poor	Fair–good	–
Bases, weak	Good	Poor	Good	Excellent	Excellent	Fair
Bases, strong	Poor	Poor	Poor	Excellent	Good	Poor
Alcohols	–	Poor	Good	Excellent	Excellent	Poor
Ketones	Dissolves	Poor	Poor	Dissolves	Poor	Poor
Esters	Dissolves	Poor	Good	Poor	Poor	–
Hydrocarbons						
Aliphatic	Good	Poor	Good	Poor	Excellent	Fair–good
Aromatic	Softens	Poor	Poor–fair	Dissolves	Poor	Poor
Oils, Vegetable, animal, mineral	Good	Poor	Good	Excellent–poor	Excellent	Good

of proteins has also been shown to modulate EOF [8]. These methods, both covalent and non-covalent, can be used to modulate EOF, prevent non-specific adsorption, or as a means to attach active molecules (such as antibodies) to the surface.

2. Fabrication

By the mid-1990s, there were few attempts to fabricate devices in alternative materials like plastics, ceramics or metals. Most of the early reports of polymer micromachining for microfluidic applications were patented [18] and not published in the open literature. One example of an all-plastic microfluidic system was reported in 1996 by Ver-Lee et al. from Abbott Laboratories [19]. Channels were fabricated using a computer-controlled milling machine with the smallest channel diameter on the order of 800 μm. Although the plastic channel network, called a fluid circuit, was a complete system, channel dimensions were still in the range of conventional tube diameters in which turbulent flow could be readily achieved. By 1997, when commercial companies were beginning to invest heavily in the developing microfluidic technology, interest in the use of polymers and plastics as substrate materials for microfluidic systems began to increase. With the introduction and development of new microfabrication technologies for plastics in the late 1990s, plastic microchannels with dimensions in the range of 15–30 μm were realized using techniques such as hot embossing or imprinting, injection molding, laser ablation, soft lithography, or X-ray photolithography.

Most of the activity in micromachining of polymers for this application resides in the industrial sector as is evidenced by the existence of hundreds of patents or pending patents in this field. The following is a review of some of the fabrication methods that have been described over the past 5 years for polymer microfluidic systems.

2.1. Imprinting and hot embossing

Hot embossing or imprinting techniques for plastic microchannel fabrication were first described in the late 1990s by several groups [20–24]. Some of the first methods described the use of wires for imprinting plastic substrates [23], methods that are still used today [25–28]. Recently, however, a silicon stamp is more commonly used as the imprinting tool for the fabrication of polymer microfluidic devices. To make a silicon stamp, a drawing of the channels is first created using a CAD tool, and the image may be transferred to a photomask or alternatively to a high contrast resolution transparency if features greater than 20 μm are desired. A silicon wafer with a crystal orientation ⟨100⟩ is coated with a masking material such as silicon dioxide or silicon nitride, and then coated with a layer of photoresist. The transparency is placed on top of a silicon wafer, and upon exposure to the UV light source, the photoresist is developed revealing the transferred image. The image is then transferred to the exposed masking layer by etching in a solution of hydrofluoric acid (HF) or potassium hydroxide (KOH) for silicon dioxide or silicon nitride, respectively. The exposed silicon is then etched anisotropically using tetramethylammonium hydroxide (TMAH), ethylenediaminepyrocatechol (EDP) or KOH, and the result is a raised three-dimensional inverted image of the channels as shown in Fig. 1A [23]. If the wafer is anisotropically etched, the resulting microstructure has a trapezoidal shape. The height and the width of the positive image may be controlled by the amount of time the wafer is etched. The etched silicon stamp may then be used to imprint microchannels in plastic materials at room temperature [29], or at elevated temperature (Fig. 1B) [23].

Alternatively, a micromachined silicon wafer may be used to fabricate a stamp in metal [30]. In this process, a metal electroform is produced (typically using Ni) using the micromachined silicon wafer as the master. The first metal electroform is the mirror image of the master. Then, a second metal electroform is created using the first electroform as a template. The second electroform is then a replica of the original silicon master. Thus, micrometer features are transposed to the more robust metal substrate. The metal stamp may be used to fabricate microchannels in plastic substrates by imprinting or injection molding.

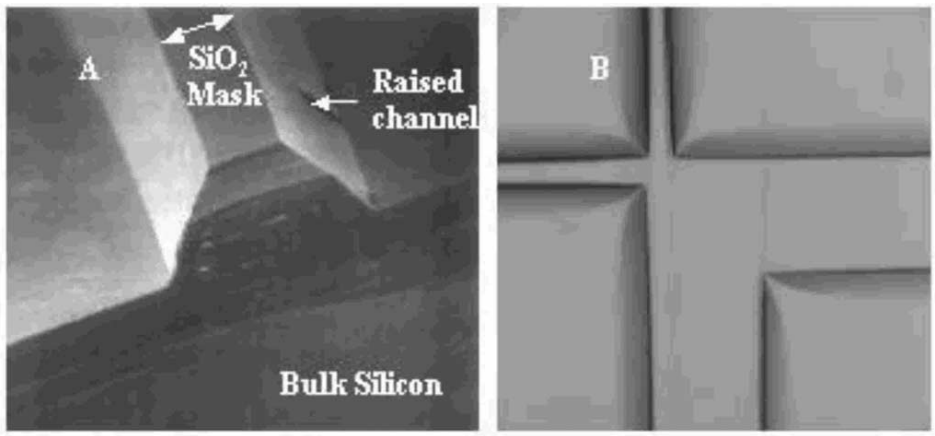


Fig. 1. A. Scanning electron micrograph of silicon template fabricated by wet chemical etching in TMAH. B. Imprinted channel in PMMA.

To imprint or emboss microchannels, a hard plastic material is generally cleaned thoroughly, dried, and then placed on top of the silicon or metal stamp. The stamp and plastic are then placed in a hydraulic press and pressure is applied for a time that is typically less than 10 min. Hot embossing is performed at temperatures close to the softening temperature of the plastic and at lower pressures. Alternatively, plastic devices can be imprinted at room temperature with elevated pressures. When devices are hot embossed, the resulting plastic microchannel dimensions are the exact mirror of the silicon stamp. When devices are imprinted at room temperature, the microchannel dimensions are much more dependent on several parameters including imprinting pressure, imprinting time and properties of the plastic itself [29]. An advantage of room temperature imprinting, however, is that fabrication time is reduced as compared to hot embossing. Reproducible imprints can be made at room temperature in less then 2 min.

With hot embossing methods, both the metal and silicon stamps have very long lifetimes and may be used repeatedly to fabricate hundreds of plastic microfluidic devices. The metal and silicon stamps can both withstand elevated temperatures and fast temperature cycling used for the replication and release processes. With room temperature imprinting of softer plastics (including PETG and PVC), the silicon stamps can also survive many imprinting cycles. With harder plastics (i.e. PMMA and polycarbonate) the silicon stamp is subject to fractures, therefore, it may be necessary to first transfer the image to a metal stamp to increase the lifetime of the stamping tool.

Many common plastics have been successfully imprinted or hot embossed with excellent device-to-device reproducibility. These include polystyrene (PS), polyethylenetetraphthalate glycol (PETG), polymethylmethacrylate (PMMA), polyvinylchloride (PVC), polystyrene, and polycarbonate [10,21,31–34]. Instruments for automated hot embossing were developed and are sold commercially by Jenoptik Mikrotechnik GmbH in Germany.

2.2. Injection molding

Injection molding techniques for fabricating microchannels were first described in the open literature by researchers at ACLARA (formerly Soane Biosciences) [30]. Silicon masters are generally fabricated using wet-chemical etching procedures as previously described, however, the silicon wafers may also be processed by deep reactive ion etching to provide structures with higher aspect ratios [35]. The method uses a nickel electroform

produced from a silicon master, as previously described, to transfer micrometer features to a substrate suitable for injection molding. The nickel electroforms from a single silicon master can be used to produce hundreds of thousands of injection molded parts. In the injection molding process, the nickel electroform is mounted onto a mold insert and the copolymer resin is introduced to produce the microfluidic parts. During the injection, the polymer solution is of relatively low viscosity which should lead to good contact with the mold resulting in well-defined features. In some of the early reports on injection molding of microchannels, acrylic injection molded channels had features that differed significantly from the features on the nickel mold. Some of the variables that can impair the replication of micrometer-sized features include mold temperature and relaxation of the polymer after release from the mold among many others. By adjusting the process time and temperature, injection molded parts can be fabricated with excellent precision.

Injection molding has now been used to reproducibly fabricate plastic microfluidic channels in plastics such as PMMA [30], and polycarbonate [36,37]. Researchers from Amersham Pharmacia Biotech and Gamera Bioscience Corporation described the replication of plastic microfluidic devices using common CD molding equipment. Advantages of injection molding over imprinting or hot embossing include the ability to create three-dimensional structures and the fact that preformed elements can be embedded into the plastic during the molding process.

2.3. Soft lithography (3-D)

A significant advancement in microfluidic systems development was the introduction of elastomeric polymer molding techniques, known as soft lithography [14,38–40]. As with all other methods described thus far, a positive relief master may be fabricated in silicon. An elastomeric polymer is then cast onto the silicon stamp and allowed to cure. After curing at room temperature or at a slightly elevated temperature to speed the curing process (generally 40–70 °C for PDMS), the elastomeric polymer is peeled off the stamp. Again, the silicon stamp may be used repeatedly to replicate hundreds of polymer microfluidic devices. Since the stamp is not exposed to excessive pressure (as with imprinting), or excessive heat (as with injection molding), fabrication of a metal electroform is not necessary. Also, stamps may be made from softer materials such as photoresists.

A great advantage of this fabrication technique is that elastomeric polymers may be easily bonded to each other or to plastic or glass substrates by conformal contact. This will be discussed in more detail later, but the simplicity associated with the sealing procedure has made this fabrication technology one of the most widely used for prototyping microfluidic systems.

Recently, there have also been reports describing the use of soft lithography for fabricating three-dimensional microfluidic devices [41–43]. The fabrication of these devices requires the design of several silicon stamps, which are then used to individually fabricate the different layers of a multi-layer three-dimensional structure. In one method, the first elastomeric layer is cast as a thick sheet and, upon curing and removal from the silicon stamp, serves as the support for the remaining system. All other elastomeric layers are all cast as very thin sheets on the silicon stamps. These thin layers are lifted off the silicon stamps in successive fashion using the supporting first layer. The excellent adhesion between successive layers promotes the use of this method for fabricating multi-layer three-dimensional devices.

Thus far, the vast majority of reports in the literature have used polydimethylsiloxane (PDMS) as the elastomeric polymer for fabrication of microfluidic devices by soft lithography [44–48]; however, other elastomeric polymers should also be suitable for molding by this method.

2.4. Laser photoablation

Photoablation was introduced in the literature as a method for fabricating polymer microfluidic channels in 1997 by Roberts et al. [7]. In the photoablation process, a polymer is exposed to a pulsed UV source, and the absorption of that

light induces bond-breakage in the polymer backbone [49]. The exact mechanism of polymer decomposition in the ablation process is still under investigation, but it is widely accepted that it is due to either photodegradation or thermal degradation, or a combination of the two. Local temperatures on the polymer surface during ablation can be very high during the ablation process (i.e. 427 °C for PMMA) [50]. In this high-energy process, a shock wave is produced and particles are ejected from the substrate thus creating a void. Small particulates can be seen on the surface of the substrate material after laser ablation, but other decomposition products include gases such as carbon monoxide and carbon dioxide. Excimer lasers with emissions of 193 (ArF) or 248 nm (KrF) are both effective in polymer ablation but to varying degrees depending on the absorption spectra of the polymer. Polymers that have significant absorption at the emission wavelength of the laser are the most effectively ablated.

Micromachining using laser ablation may be achieved by exposing the polymer substrate to the laser through a mask that defines the area to be ablated. In this case, the mask is made from a material that does not have significant absorption at the laser wavelength, e.g. some metals. Alternatively, channels and other structures can be defined and micromachined using a direct-write, maskless process. In this process, the polymer substrate material is placed on a moveable (preferably programmable) stage, and the substrate is moved under the focused laser beam to create the desired structure. The laser beam may be further defined by passing it through an aperture of appropriate dimension and shape prior to focusing. The direct-write micromachining process is advantageous in that a mask does not have to be created to change the design of the microchannel network; therefore, channel design can be changed rapidly during the prototyping process. The disadvantage to this approach is that parts are made in a sequential manner thereby limiting the ability to mass produce devices for commercial applications.

The excimer laser provides a pulsed output; therefore, depending on the pulse rate and the rate of movement of the stage, laser ablated channels can have a rippled appearance Fig. 2A. Generally, laser ablated channels have greater surface roughness than imprinted, hot embossed, or injection molded channels. The degree of roughness is highly dependent on the absorption of the polymer at the excimer wavelength. For instance, PMMA channels ablated at 248 nm have a very rough appearance with significant porosity as shown in Fig. 2B [10]. The depth of laser ablated channels is dependent on many parameters including polymer absorption, laser power, pulse rate, and number of passes made across the channel. Channels are generally square or rectangular shaped with straight walls,

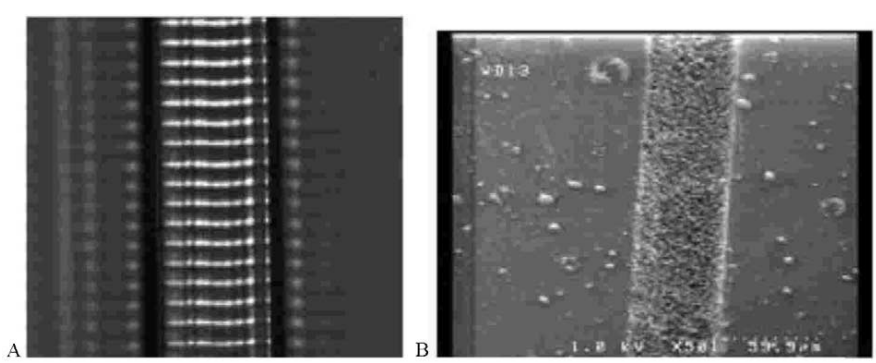


Fig. 2. A. Brightfield microscopic image (20 ×) of polystyrene microchannel ablated using a 248 nm eximer laser. B. SEM image of PMMA channel ablated using a 248 nm eximer laser.

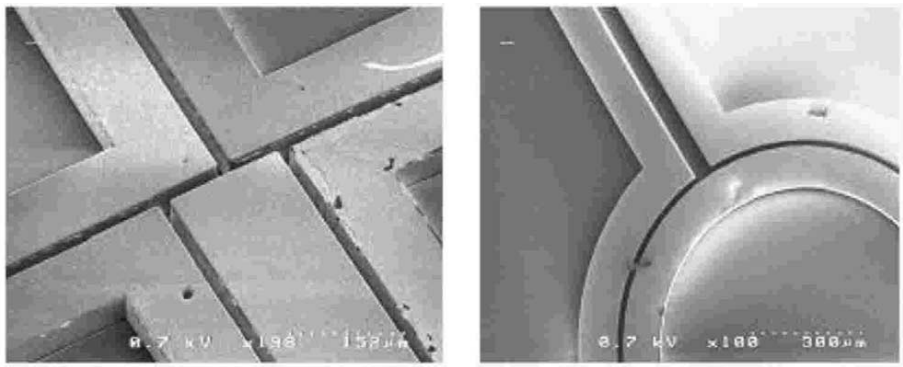


Fig. 3. SEM images of PMMA microchannels fabricated using LIGA process. Images courtesy of S. Soper, Louisiana State University.

however, in very deep channels, walls can become slanted due to laser defocusing effects with each successive pass. The smallest channel width that is achievable is defined either by the mask or by the focusing optics and aperture. Using a direct-write laser ablation system at NIST with an aperture to define the beam, channels with 1.5 μm features may be fabricated.

Channels have been fabricated in a wide variety of commercially available plastics using laser ablation. Those that have been reported in the literature using the ArF eximer laser (193 nm) include polycarbonate, polystyrene, cellulose acetate, and poly(ethylene terephthalate) [7,51–53]. In our work, microchannels were fabricated using a KrF eximer laser (248 nm) in PMMA, PETG, PVC, polycarbonate, polystyrene, and polyimide [10].

2.5. X-ray lithography

Recently, X-ray lithography has been adapted [32,54,55] for fabricating polymer microchannels. The most common substrate material used in this process is PMMA because it exhibits high X-ray absorption (soft X-rays of 0.7–0.8 nm) and is sensitive to X-ray degradation. To fabricate structures using X-ray lithography, a quartz–chrome mask is first generated to define the pattern. To create a reuseable gold/Kapton™ mask for the LIGA process, a Kapton film coated with a very thin film of gold is placed in contact with the PMMA substrate. This Kapton layer is transparent to X-rays. The Kapton layer is then coated with photoresist and the image from the quartz–chrome mask is transferred photolithographically to the photoresist over the Kapton layer. Once the photoresist is developed, a thick layer of gold is deposited onto the Kapton surface in the open areas in the photoresist. The thick gold layer absorbs X-rays thereby protecting the polymer substrate that lies beneath it. Sections of the Kapton without the thick gold layer are transparent to the X-rays. The photoresist is removed, and the polymer substrate material is irradiated with X-rays through the gold/Kapton mask to degrade the exposed polymer. For PMMA substrates, the X-ray exposure induces a number of scission reactions resulting in a variety of soluble oligomers [56]. The exposed, degraded polymer is finally dissolved in a developing solvent that solubilizes the reaction products, thus forming the microstructure. This process can yield high aspect ratio structures with straight, smooth walls as shown in Fig. 3. The channel depth depends on the X-ray energy and on the exposure time.

This process can be used to fabricate microchannels, or may also be used to generate stamps (similar to silicon stamps) for imprinting or injection molding. Since a polymer substrate material would not be robust enough for direct use as a template, features can be transferred to a nickel electroform as described previously.

2.6. Other methods

Several other methods have been reported recently in the literature for the fabrication of microfluidic channels. Lee et al. described the use of low energy ion beam etching (IBE) with Ar^+ ions (500 eV, 0.5 mA cm^{-2}) to fabricate high aspect ratio microchannels in a variety of fluoropolymers, including polytetrafluoroethylene (PTFE), Tefzel™, fluoroethylenepropylene (FEP) and Teflon AF™ [57]. Rossier et al. described the use of plasma etching as a method for mass-producing microfluidic devices in polymer substrates [58]. Several groups reported UV-patterning of photoresists for microfluidics and MEMS applications. SU-8, a negative photoresist, has been the most widely used photoresist for this application due to its excellent chemical resistance and the fact that it can be coated in thick layers [59–61]. Lastly, Mastrangelo et al. [62] described a sacrificial etch process to fabricate polymer microchannels in parylene-C.

As discussed, many of these fabrication techniques require the use of a master. Table 3 shows a comparison of the different technologies available for fabricating masters for use in the various methods. It is important to consider that many of the fabrication processes described are suitable for only a limited number of polymers and plastics for the reasons stated previously.

2.7. Sealing

All fabrication methods described thus far besides the sacrificial etching method require post-fabrication sealing of the microchannel to form an enclosed structure. Sealing of polymer microchannels is generally much simpler than with silicon or glass channels and can often be accomplished using low temperature thermal annealing [8,34]. The same polymer substrate can be used to form the seal or alternatively, a polymer with a lower glass transition temperature can be used to ensure that there is no deformation of the microchannel during the sealing process [7]. Elastomeric polymers such as PDMS have excellent adhesion to a wide variety of substrate materials and can be used to enclose microchannels with a non-permanent seal [63,64]. To form a permanent seal with PDMS, Whitesides et al. [14] describe plasma oxidation of PDMS surfaces to bond the material to itself or to other substrates including glass, silicon, silicon oxide, quartz, silicon nitride, polyethylene, polystyrene, and glassy carbon. It was hypothesized that the permanent bond between two PDMS pieces is a result of a condensation reaction to form a covalent siloxane bond.

3. Applications

3.1. Theory

To understand the motivation for miniaturizing applications in chemistry and the life sciences, it is of the utmost importance to look at the behavior of physical parameters when a system is scaled down in size. These scaling laws have been investigated in the groundbreaking paper by Manz et al. [65]. The underlying assumption for all miniaturized systems is that their transport phenomena are controlled by diffusion. For fluidic systems, this means that the flow regime is strictly laminar [66]. The transport phenomena relevant for the applications at hand cover two aspects, on the one hand the transport of individual molecules, on the other hand the transport of heat. Diffusion is described by Fick's Laws or the diffusion equation:

$$\dot{n} = D\Delta n, \tag{1}$$

where n is the particle density or concentration, D is the diffusion coefficient and Δ is the Laplace operator. The scaling behavior becomes clearer if Fick's Law is rewritten in terms of diffusion time t_D, i.e. the time a molecule needs to travel the distance l by diffusive processes (or alternatively in the case of heat diffusion the time needed for a thermal gradient to equalize):

$$t_D = D^{-1}l^2. \tag{2}$$

It becomes clear that the biggest advantage in miniaturization lies in the quadratic decrease of equilibration time with a downscaling of the linear dimension of a system. To illustrate this, some diffusion times for a small and a large molecule

Table 3
Overview on existing master fabrication methods

Technology	Choice of geometry	Minimum feature size	Height	Total surface area	Aspect ratio	Lifetime	Cost	Availability
Wet silicon etching	–	+	0	++	–	+	+	++
Dry silicon etching	+	++	+	++	+	–	0	+
Optical lithography and electroforming	+	++	+	++	0	+	0	0
Laser ablation and electroforming	++	+	+	–	+	+	–	–
LIGA	+	++	++	–	++	+	– –	– –
Mechanical micromachining	+	0	+	+	0	++	–	–
μ-EDM	–	0	+	–	+	++	–	–

for a range of distances are listed in Table 4. It is important to note that the utilization of microsystem technologies reduces typical time scales from hours to seconds.

While the simple equations for diffusion processes explain well the behavior of molecules in static reaction vessels (e.g. nanowell plates), the theoretical advantages of performing electrophoresis in a miniaturized capillary have to be evaluated by looking at some simple equations for the separation efficiency [67], the so called 'number of theoretical plates' N. This value is defined by:

$$N = \frac{L^2}{\sigma_x^2}, \tag{3}$$

where L is the length of the capillary and σ_x^2 is the variance of the migration zone width, i.e. the diffusion limited spread in the peak of a single component.σ_x^2 is given by

$$\sigma_x = \sqrt{2D_i t} = \left(\frac{2D_i L}{\mu_{CE} E}\right)^{1/2}, \tag{4}$$

with D_i the diffusion coefficient of component i, E the electric field along the capillary and μ_{CE} the electrophoretic mobility of this particular species. Inserting Eq. (4) into Eq. (3) we obtain

$$N = \frac{\mu_{CE}}{2D} U, \quad \text{i.e. } N \propto U. \tag{5}$$

The number of theoretical plates is therefore proportional to the applied voltage U along the capillary. The maximum permissible value of U is due to the generated Joule heat in the capillary proportional to the geometry factor L/d, where d is the capillary diameter. Therefore, recalling Eq. (5) we get

$$N \propto \frac{L}{d}. \tag{6}$$

The second important variable is the analysis time t, i.e. the time a component i needs to travel the distance L, which is given by

$$t = \frac{L}{v_i} = \frac{L^2}{\mu_{CE} U}, \tag{7}$$

where v_i is the velocity of species i.Recalling Eq. (5), inserting Eq. (6), and using Eq. (7), we obtain

$$t \propto Ld \tag{8}$$

We now have the two important results motivating the miniaturization of capillary electrophoresis, which can be combined to a single equation (by combining Eqs. (6) and (8)):

$$\frac{N}{t} \propto \frac{1}{d^2}. \tag{9}$$

This means that the separation efficiency per unit time is inversely proportional to the square of the capillary diameter, indicating the potential of microsystem technologies to realize great improvements in performance. The typical performance increase is approximately two orders of magnitude in analysis speed.

3.2. Example devices

The following sections list several applications of polymer-based microfluidic systems that have been fabricated with methods described previously.

Table 4
Diffusion times of molecules vs. typical length scales

Distance	Object	Small molecule, e.g. O_2 ($D = 2\times10^{-9}$ m^2 s^{-1})	Large molecule, e.g. protein ($D = 2\times10^{-11}$ m^2 s^{-1})
8 nm	Cell membrane	32 ns	3.2 μs
8 μm	Erythrocyte/microchannel	32 ms	3.2 s
80 μm	Cell/microchannel	3.2 s	320 s
0.8 mm	Amoeba/nanowell	320 s	8.9 h
8 mm	Leech/microwell	8.9 h	890 h
80 mm	Hand	890 h	10 years

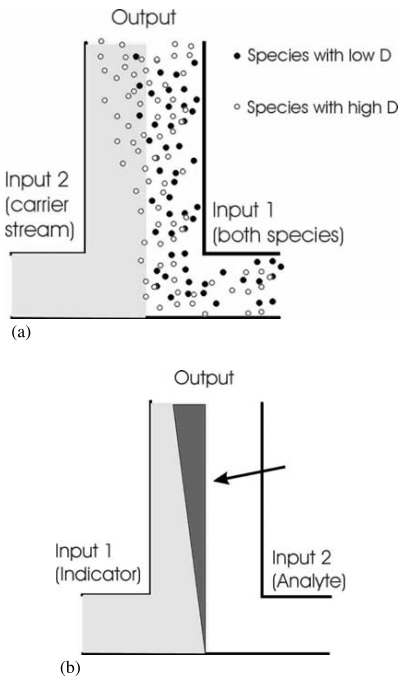


Fig. 4. A. Extraction of chemical species with high diffusion coefficients in a T-cell. B. Measurement of a sample concentration due to diffusive mixing with an indicator stream.

3.2.1. Flow cells

Even geometrically simple microchannel configurations can be very successfully utilized in microfluidics. Examples for such devices are T-junctions or double T-junctions as shown in Fig. 4 [68,69]. Such devices solely make use of the diffusion properties of the substances involved and can be used, for example to extract a species with a high diffusion coefficient from a sample stream due to the diffusion of molecules from one carrier stream to another (Fig. 4A). Other applications include the measurement of a sample concentration due to diffusive mixing with an indicator stream [70] (Fig. 4B) where the color change of the indicator can be optically detected. This same concept can be applied in a diffusion-based immunoassay. In this latter case, the difference in diffusion coefficients between smaller antibodies and larger antigens is utilized to create a specific response.

More or less complicated microchannel networks or manifolds, which are mostly manufactured with an injection molding process, are used to encapsulate other functional elements, namely DNA-arrays or biosensors. In these cases, the microfluidic network fulfills metering, dosing and distribution functions only [71,72] as can be seen in Fig. 5. Typical dimensions of these distribution channels are of the order of 100 μm, which allows the use of conventional machining methods for mold preparation.

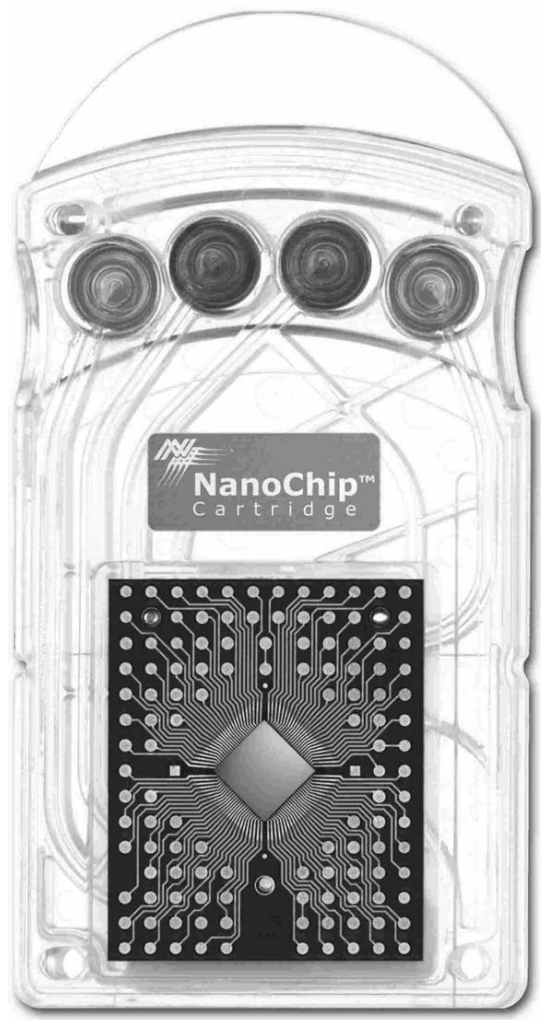


Fig. 5. Nanochip™ Cartridge with microfluidic channels for sample manipulation. Figure courtesy of Nanogen Inc., San Diego, CA.

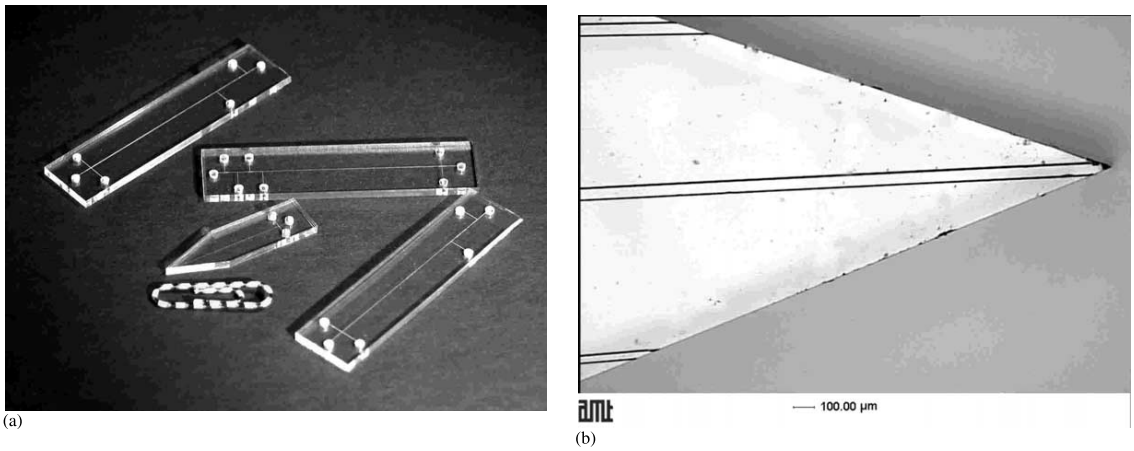
3.2.2. Capillary electrophoresis

Historically, the first widely published application of μTAS devices was in capillary electrophoresis [65,2] and this technology was also the first to be commercially available. While the first devices were all on glass or quartz substrates [2,73–75], first applications of polymer devices were reported in the patent literature in 1990. A massive use of polymers as substrate materials for CE-applications started in the last few years, being driven on the one hand by the extremely simple and cheap manufacturing of silicone elastomer based microfluidic devices, which allows, particularly, academic groups to prototype new devices rapidly. For industrial applications, however, it is the use of thermoplastic polymers that is attractive for a number of reasons:

- *Cost of substrate material*. As many microfluidic devices have a comparatively large footprint (typically several cm^2) to achieve either long separation channel length or a high parallelization of their functions, the cost of the substrate material plays an important factor for high volume production. Polymers like PMMA are of the order of 0.2–2 cent cm^{-2}, while boro-float glasses (e.g. Corning Pyrex) are of the order of 10–20 cents cm^{-2}, boro-silicate glasses (e.g. Schott B270) approximately 5–15 cents cm^{-2}, and photostructurable glasses (e.g. Schott Foturan) approximately 20–40 cents cm^{-2}.
- *Process complexity*. Many process steps (cleaning, resist coating, photolithography, development, wet etching) and harmful wet chemistry (e.g. hydrofluoric acid) are involved in fabricating glass devices. In addition, process costs are significant due to the reagents involved as well as their waste disposal.
- *Limitations in geometrical design*. Due to the isotropic nature of the etching process, only shallow, mainly semicircular channel cross-sections are possible in glass substrates. For many applications, however, other channel cross-sections are desirable including high aspect ratio square channels, channels with a defined but arbitrary wall angle, or channels with different heights.

Polymer-based devices for application in capillary electrophoresis have been fabricated using all of the methods previously described including hot embossing [23,24,21,55,76–81], injection molding [30,82], laser ablation [7,58,83] and direct X-ray exposure [84,85] in thermoplastic polymers as well as in elastomers, namely polydimethylsiloxane (PDMS) [38,14,86–89]. The achievable separation speed and resolution is comparable with devices made out of glass. As a detection method laser induced fluorescence is mostly used [23,30,38], but also electrochemical detection methods with on-chip electrodes have been reported [81,58]. In case of fluorescence detection, the generally higher autofluorescence of polymer materials in comparison to glass has to be taken into account. PMMA and special grades of PC, however, show an autofluorescence not much higher than standard borofloat glass. For electrochemical detection, the manufacturing of electrodes normally is realized with shadow-masks, where the metal is deposited by evaporation or sputtering through a mask containing the electrode outline. Lithographic processes are also possible but difficult due to the incompatibility of many polymers with most chemicals (e.g. developer) used in photolithography.

A recent development is the coupling [90] of CE-chips with mass spectrometry [91,92]. The CE-chips have been used in this format either for sample preparation in a separation mode or for sample preconcentration by isoelectric focussing [93]). The low surface charge of most polymers proves advantageous in this application since, with glass chips, coatings are often necessary to reduce the electroosmotic flow [94]. In such a CE-MS device, the microchannel must end in a sharp tip to provide a sufficiently high electrical field for producing an electrospray injection into the MS. Any geometrical deformation leads to a non-optimal Taylor-cone configuration, therefore the fabrication method is crucial. Fig. 6A shows the layout of such a device and Fig. 6B shows an actual photograph of the tip. The device was fabricated at amt Jena by hot embossing in PMMA and the tip was cut mechanically with a wafer saw.



(a) (b)

Fig. 6. A. Hot embossed PMMA devices for CE and CE-MS. B. Photograph of tip on microchannel device that couples with the electrospray mass spectrometer.

3.2.3. Miniaturized PCR

One of the most widely used processes in biotechnology is the polymerase chain reaction (PCR) for the amplification of specific DNA fragments. While the macroscopic devices for PCR are mainly thin-walled tubes made out of polystyrene, most microscopic devices to date have been made out of silicon or silicon/Pyrex. Miniaturized devices for batch PCR [95] as well as continuous flow PCR have been reported [96,97]. As the PCR process involves three temperatures (47–55, 70–75, 95 °C), only polymers with a higher temperature stability can be utilized for this application, namely polycarbonate and cyclo-olefin copolymer (COC) as thermoplastic materials and PDMS as elastomer. In Fig. 7, polymer chips for continuous PCR [97] together with a nickel master for the replication is shown. DNA-amplification has been performed with the pyrosequencing technique, which manages without the need for high temperature cycling [98], and has also been performed using conventional thermal PCR cycling as reported by Eckersten et al. [99] in a molded CD-platform.

3.2.4. Clinical chemistry and diagnostics

An application field particularly suited for polymer devices is diagnostics. In this application, disposable devices are needed to avoid contamination and, therefore, inexpensive polymer microfabrication technologies offer great potential for their commercial viability. An example of a device already on the market is the portable blood analyzer system from i-STAT [100]. While the measurements themselves are performed on silicon-based sensors, the liquid handling, dosing and sampling is done in an injection molded polymer cartridge. Several other diagnostic systems are currently under development, notably CD-based systems like the Lab-on-a-disk [101].

3.2.5. Cell handling

In many biological applications, the handling of (living) cells is of great interest. Furthermore, as typical cell sizes and the geometrical dimensions of microchannels are of the same order of magnitude, handling and manipulation of single cells are greatly simplified. A typical task performed in a microfluidic structure is cell counting and flow cytometry [102]. Cell culturing on a microstructured polycarbonate CD has been reported by Thomas et al. [103], with all necessary process steps like cell transport, introduction of culturing and assay reagents, and incubation performed in the microstructure. PDMS-based microfluidic systems have also been reported for cell handling and cytometry tasks [104,105].

Other complex planar microfluidic networks have been used for cell manipulation [106,107]. These devices make use of the electrokinetic trans-

port phenomena of dielectrophoresis, travelling wave dielectrophoresis, or electrorotation to move, separate and characterize individual cells or cell clusters. Such devices have been fabricated using UV sensitive dry laminate negative resist substrates with microchannel structures produced by laser ablation. A multilayer network of electrodes allows the creation of a travelling electric field, having four sets of electrodes with a phase difference of the electrical field of 90° between each electrode. Such a configuration can act as a 'conveyer-belt' for cells or other dielectric particles.

3.2.6. Micro-reactors and containers

While the previously described devices all used flow systems, many applications can also take place in a static environment in miniaturized reaction vessels. The classical example of such devices is the miniaturized versions of the open microtiter plate, the nanowell plate. These open systems can be filled with pipetting devices and are easily fabricated in polymers. Fig. 8 shows two examples of such nanowell plates: (A) wells fabricated by laser ablation [108] in PMMA with a well volume of 11 nl [109], (B) wells made by hot embossing from a wet etched silicon master with a well volume of 700 pl [110]. Other examples are PDMS nanowell plates [111], PDMS bioreactor vessels [112] with immobilized enzymes for biocatalysis, and injection molded polycarbonate PCR well arrays [113].

4. Markets

The markets for the above-mentioned microfluidic devices are strongly driven by the expanding biotechnology market with its demand for fast analytical processes for applications like high-throughput screening, gene expression analysis and pharmacogenomics. Almost any laboratory for molecular biology is therefore likely to be equipped with instrumentation containing miniaturized devices in the near future. In addition, new and much larger markets will open up if microfluidic devices can enter the arena of clinical and point-of-care or even the home diagnostics market. Caliper Technologies and Aclara BioSciences have been in existence for several years and lead the way in commercialization of microfluidic devices by teaming up with large partners. However, a great number of small start-up companies have been formed in this field and the

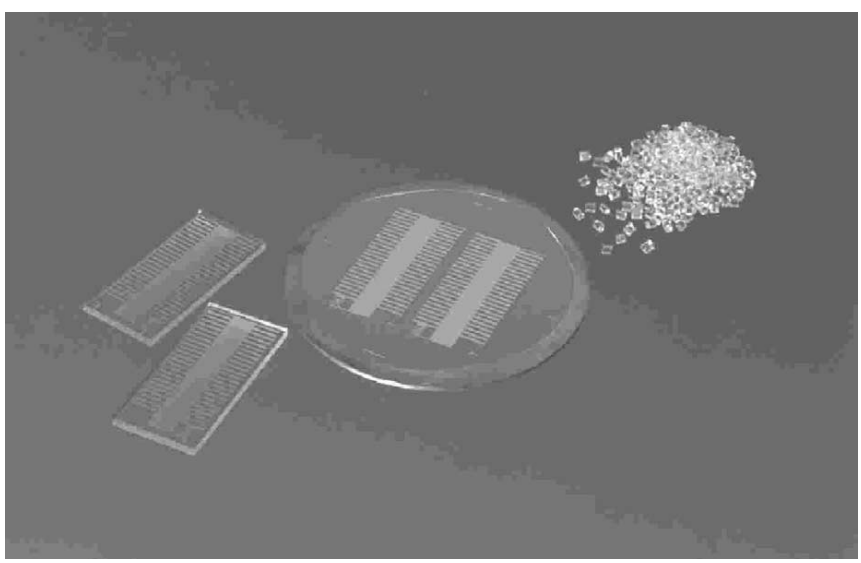
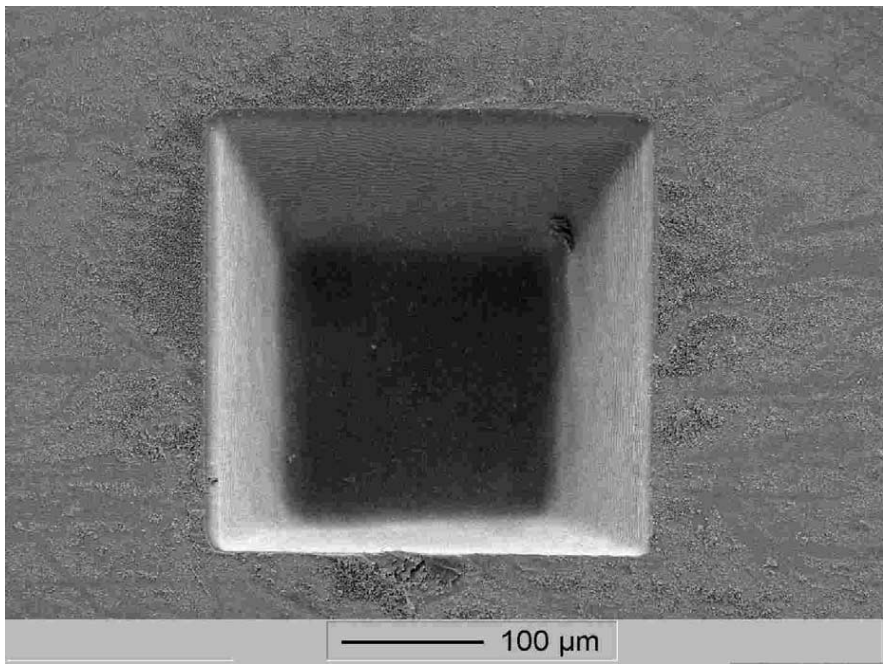
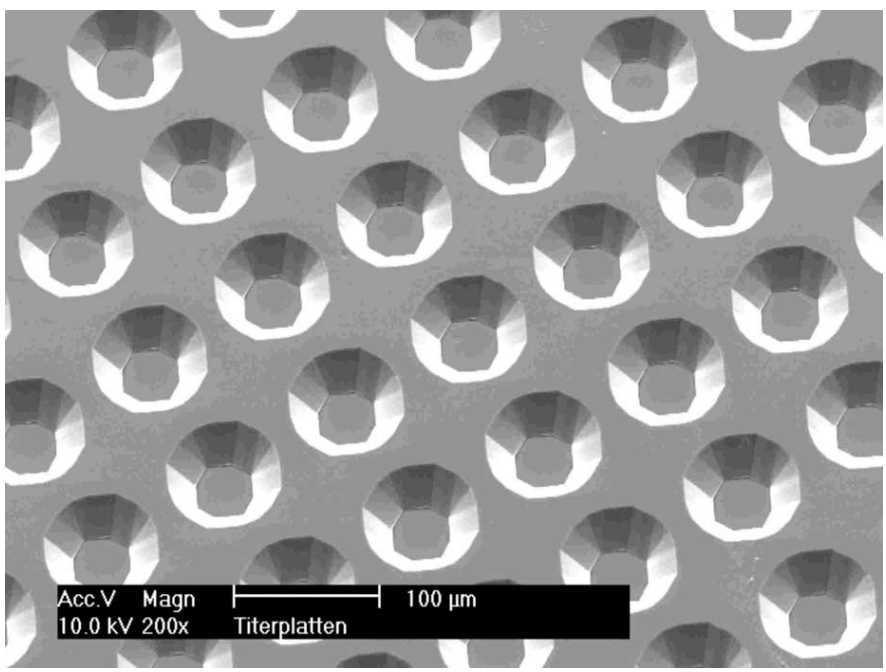


Fig. 7. Polymer devices fabricated for continuos-flow PCR with Ni master used for fabrication.



(a)



(b)

Fig. 8. A. Laser ablated nanowell in PMMA with well volume of 11 nl. B. Hot embossed nanowell plate with a well volume of 700 pl.

Table 5
Overview on market data

Source	Market	Market size today/ time of study (Mio USD)	Future market size (Mio USD)	Compound Av. growth rate (%)
Lehmann Brothers 1997	BioMEMS		23.000 (2004)	
NEXUS Market Study 1998	Lab-on-a-chip	0 (1998)	1.000 (2002)	
BioInsight 1999	Biochips (data shown here for microfluidic devices)	11 (1999)	157 (2005)	33
Frost and Sullivan 1999	Biochips	40 Mio USD [3]	6.300 (2005)	48
MarkIntel 1999	Blood analysis + genomics		540 (2003)	
System Planning Corporation 1999	Microfluidics		2.600–4.600 (2003)	25–30
Biotechcoverage.com 2000	Biochips (array + microfluidics)	2.150 (2001)	10.000 (2004)	

number is steadily increasing. Regardless of this fact, there is still a distinct lack of microfluidic systems that are commercially available, as most systems are either custom-developed for a specific purpose or are still in experimental stages. Furthermore, very few companies in the field generate revenue from product sales. An estimation of the potential market size remains difficult. While there have been several market studies published by consulting companies, the reported market figures differ vastly. One explanation for this trend is the often-lacking differentiation between microfluidic devices and array devices. Another explanation is that there is such an extensive range of potential applications for these devices. Table 5 compiles an overview of some of the current market data.

Topics that will influence the market performance of microfluidic devices in the future include the following:

- *Standardization*, *modularization*, *and platform technologies*. Thus far, all published devices feature their own technical solution to a given application. No standards have to this date been developed and devices from different suppliers are mutually incompatible. This is a notable difference, for example, to the open high-throughput screening systems based on microtiterplates where all hardware can handle a common external format (SBS-standard 96-well plate), independently of supplier or number and shape of the wells. It is very likely that such an approach could greatly extend the user base of microfluidic systems.
- *Production infrastructure*. No technology can be commercially successful without a dedicated production infrastructure. The advent of polymer foundry services in analogy to semiconductor foundries will allow the development and manufacturing of microfluidic devices without large capital investment for in-house microfabrication capabilities.
- *Business models and success-stories*. In novel markets, time is required to establish working business models. In the microfluidic sector a trend towards certain teambuilding strategies can be observed: either a dedicated microfluidics company in collaboration with a system supplier with distribution channels (e.g. Caliper and Agilent), or a biotech or pharmaceutical company in collaboration with a microfabrication company (foundry service).

Despite the insecurities of commercializing new technologies, a long-term success of microfluidic devices and systems is almost assured due to the broad technological base and the wide range of applications.

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References

[1] A. Manz, D.J. Harrison, E.M.J. Verpoorte, J.C. Fettinger, A. Paulus, H. Ludi, H.M. Widmer, J. Chromatogr. 593 (1992) 253–258.
[2] D.J. Harrison, K. Fluri, K. Seiler, Z.H. Fan, C.S. Effenhauser, A. Manz, Science 261 (1993) 895–897.
[3] W.A. Kaplan, Modern Plastics Encyclopedia '99, McGraw-Hill, New York, 1998.
[4] C.A. Harper, Modern Plastics Handbook, McGraw-Hill, New York, 2000, pp. C.1–C.60.
[5] Certain commercial equipment, instruments or materials are identified in this report to specify adequately the experimental procedure. Such identification does not imply the recommendation or endorsement by the National Institute of Standards and Technology, nor does it imply that the materials or equipment identified are necessarily the best available for the purpose.
[6] R.C. Weast, M.J. Astle, CRC Handbook of Chemistry and Physics, 60th ed., CRC Press, Boca Raton, FL, 1979, p. F-80.
[7] M.A. Roberts, J.S. Rossier, P. Bercier, H. Girault, Anal. Chem. 69 (1997) 2035–2042.
[8] L.E. Locascio, C.E. Perso, C.S. Lee, J. Chromatogr., A 857 (1999) 275–284.
[9] A.C. Henry, T.J. Tutt, M. Galloway, Y.Y. Davidson, C.S. Mcwhorter, S.A. Soper, R.L. McCarley, Anal. Chem. 72 (2000) 5331–5337.
[10] T. Johnson, E.A.Waddell, G.W. Kramer, L.E. Locascio (2001), submitted for publication.
[11] M.L. Branham, W.A. MacCrehan, L.E. Locascio, J. Cap. Electrophoresis 6 (1999) 43–50.
[12] S.-C. Wang, C.E. Perso, M.D. Morris, Anal. Chem. 72 (2000) 1704–1706.
[13] S.L.R. Barker, M.J. Tarlov, H. Canavan, J.J. Hickman, L.E. Locascio, Anal. Chem. 72 (2000) 4899–4903.
[14] D.C. Duffy, J.C. McDonald, O.J.A. Schueller, G.M. Whitesides, Anal. Chem. 70 (1998) 4974–4984.
[15] R.C. Weast, M.J. Astle, CRC Handbook of Chemistry and Physics, 60th ed., CRC Press, Boca Raton, FL, 1979, pp. C781–C790.
[16] S.L.R. Barker, D. Ross, M.J. Tarlov, M. Gaitan, L.E. Locascio, Anal. Chem. 72 (2000) 5925–5929.
[17] J.S. Rossier, G. Gokulrangan, H.H. Girault, S. Svojanovsky, G.S. Wilson, Langmuir 16 (2000) 8489–8494.
[18] B. Ekström, G. Jacobsson, O. Öhman, H. Sjödin, Patent WO 91/16966, 1990.
[19] D. VerLee, A. Alcock, G. Clark, T.M. Huang, S. Kantor, T. Nemcek, J. Norlie, J. Pan, F. Walsworth, S.T. Wong, Proc. Solid State Sensor Actuator Workshop, Hilton Head, 1996, pp. 9–14.
[20] L. Lin, C.-J. Chiu, W. Bacher, M. Heckele, Seventh International Symposium on MicroMachine and Human Science, 1996, pp. 67–71.
[21] H. Becker, W. Dietz, P. Dannberg, Microfluidic manifolds by polymer hot embossing for micro-TAS applications, in: D.J. Harrison, A. van den Berg (Eds.), MicroTotal Analysis Systems '98, Kluwer, Dordrecht, 1998, pp. 253–256.
[22] H. Becker, U. Heim, Sens. Mater. 11 (1999) 297–304.
[23] L. Martynova, L. Locascio, M. Gaitan, G.W. Kramer, R.G. Christensen, W.A. MacCrehan, Anal. Chem. 69 (1997) 4783–4789.
[24] L.E. Locascio, M. Gaitan, J. Hong, M. Eldefrawi, Plastic microfluid devices for clinical measurements, in: D.J. Harrison, A. van den Berg (Eds.), MicroTotal Analysis Systems '98, Kluwer, Dordrecht, 1998, pp. 367–370.
[25] L.E. Locascio, C.E. Perso, C.S. Lee, J. Chromatogr., A 857 (1999) 275–284.
[26] D.-C. Chen, F.-L. Hsu, D.-Z. Zhan, C.-H. Chen, Anal. Chem. 73 (2001) 758–762.
[27] S.C. Wang, C.E. Perso, M.D. Morris, Anal. Chem. 72 (2000) 1704–1706.
[28] Y.H. Chen, S.H. Chen, Electrophoresis 21 (2000) 165–170.
[29] J. Xu, L. Locascio, M. Gaitan, C. Lee, Anal. Chem. 72 (2000) 1930–1933.
[30] R.M. McCormick, R.J. Nelson, M.G. AlonsoAmigo, J. Benvegnu, H.H. Hooper, Anal. Chem. 69 (1997) 2626–2630.
[31] H. Becker, U. Heim, Sens. Actuators, A 83 (2000) 130–135.
[32] K. Ueno, F. Kitagawa, H.B. Kim, T. Tokunaga, S. Matsuo, H. Misawa, N. Kitamura, Chem. Lett. 8 (2000) 858–859.
[33] K. Uchiyama, W. Xu, M. Yamamoto, T. Shimosaka, T. Hobo, Anal. Sci. 15 (1999) 825–826.
[34] S.A. Soper, S.M. Ford, S. Qi, R.L. McCarley, K. Kelly, M.C. Murphy, Anal. Chem. A 72 (2000) 642A–651.
[35] G. Ekstrand, C. Holmquist, A.E. Orlefors, B. Hellman, A. Larson, P. Andersson, Integrated cell based assays in microfabricated disposable CD devices, in: A. van den Berg, W. Olthius, P. Bergveld (Eds.), MicroTotal Analysis Systems 2000, Kluwer, Dordrecht, 2000, pp. 249–252.
[36] N. Thomas, A. Ocklind, I. Blikstad, S. Griffiths, M.Kenrick, H. Derand, G. Ekstrand, C. Ellstrom, A. Larsson, P. Andersson, Proc. Microtas 2000, Enschede, pp. 249–252.

[37] G.J. Kellogg, T.E. Arnold, B.L. Carvalho, D.C. Duffy, N.F. Sheppard, Centrifugal microfluidics: applications, in: A. van den Berg, W. Olthius, P. Bergveld (Eds.), MicroTotal Analysis Systems 2000, Kluwer, Dordrecht, 2000, pp. 239–242.
[38] C.S. Effenhauser, G.J.M. Bruin, A. Paulus, M. Ehrat, Anal. Chem. 69 (1997) 3451–3457.
[39] J.C. McDonald, D.C. Duffy, J.R. Anderson, D.T. Chiu, H.K. Wu, O.J.A. Schueller, G.M. Whitesides, Electrophoresis 21 (2000) 27–40.
[40] K. Hosokawa, T. Fujii, I. Endo, Hydrophobic microcapillary vent for pneumatic manipulation of liquid in microTAS, in: D.J. Harrison, A. van den Berg (Eds.), MicroTotal Analysis Systems '98, Kluwer, Dordrecht, 1998, pp. 307–310.
[41] R.J. Jackman, S.T. Brittain, A. Adams, M.G. Prentiss, G.M. Whitesides, Science 280 (1998) 2089–2091.
[42] J.R. Anderson, D.T. Chiu, R.J. Jackman, O. Cherniavskaya, J.C. McDonald, H.K. Wu, S.H. Whitesides, G.M. Whitesides, Anal. Chem. 72 (2000) 3158–3164.
[43] B.H. Jo, L.M. Van Lerberghe, K.M. Motsegood, D.J. Beebe, J. MEMS (2000) 76–81.
[44] E. Delamarche, A. Bernard, H. Schmid, B. Michel, H. Biebuyck, Science 276 (1997) 779.
[45] E. Delamarche, A. Bernard, H. Schmid, A. Bietsch, B. Michel, H. Biebuyck, J Am. Chem. Soc. 120 (1998) 500.
[46] R.S. Martin, A.J. Gawron, S.M. Lunte, C.S. Henry, Anal. Chem. 72 (2000) 3196–3202.
[47] K. Hosokawa, T. Fujii, I. Endo, Anal. Chem. 71 (1999) 4781–4785.
[48] T. Yang, S.-Y. Jung, H. Mao, P.S. Cremer, Anal. Chem. 73 (2001) 165–169.
[49] R. Srinivasan, Polym. Degrad. Stab. 17 (1987) 193–203.
[50] H. Schmidt, J. Ihlemann, K. Luther, J. Troe, Appl. Surf. Sci. 138–139 (1999) 102–106.
[51] J.S. Rossier, P. Bercier, A. Schwarz, S. Loridant, H.H. Girault, Langmuir 15 (1999) 5173–5178.
[52] J.S. Rossier, G. Gokulrangan, H.H. Girault, S. Svojanovsky, G.S. Wilson, Langmuir 16 (2000) 8489–8494.
[53] N. Xu, Y. Lin, S.A. Hofstadler, D. Matson, C.J. Call, R.D. Smith, Anal. Chem. 70 (1998) 3553–3556.
[54] S.M. Ford, J. Davies, B. Kar, S.D. Qi, S. Mcwhorter, S.A. Soper, C.K. Malek, J. Biomech. Eng. ASME 121 (1999) 13–21.
[55] S.M. Ford, B. Kar, S. McWhorter, J. Davies, S.A. Soper, M. Klopf, G. Calderon, V. Saile, J. Microcolumn Sep. 10 (1998) 413.
[56] J.A. Moore, J.O. Choi, Radiation Effects on Polymers, in: R.L. Clough, S.W. Shaleby (Eds.), ACS Symposium Series, vol. 475, American Chemical Society, Washington, DC, 1991, p. 156.
[57] L.P. Lee, S.A. Berger, L. Pruitt, D. Liepmann, Key elements of a transparent Teflon microfluidic system, in: D.J. Harrison, A. van den Berg (Eds.), MicroTotal Analysis Systems '98, Kluwer, Dordrecht, 1998, pp. 245–248.
[58] J.S. Rossier, A. Schwarz, F. Bianchi, F. Reymond, R. Ferrigno, H.H. Girault, Polymer microstructures: prototyping, low-cost mass fabrication and analytical applications, in: A. van den Berg, W. Olthius, P. Bergveld (Eds.), MicroTotal Analysis Systems 2000, Kluwer, Dordrecht, 2000, pp. 159–162.
[59] H. Lorentz, M. Despont, N. Fahrni, J. Brugger, P. Vettiger, P. Renaud, Sens. Actuators, A—Physical 64 (1998) 33–39.
[60] P. Renaud, H. van Lintel, M. Heuschkel, L. Guerin, Photo-polymer microchannel technologies and applications, in: D.J. Harrison, A. van den Berg (Eds.), MicroTotal Analysis Systems '98, Kluwer, Dordrecht, 1998, pp. 17–22.
[61] K.Y. Lee, N. LaBianca, S.A. Rishton, S. Zolgharnain, J.D. Gelorme, J. Shaw, T.H.P. Chang, J. Vac. Sci. Technol., B 13 (1995) 3012–3016.
[62] J.R. Webster, M.A. Burns, D.T. Burke, C.H. Mastrangelo, An inexpensive plastic technology for microfabricated capillary electrophoresis chips, in: D.J. Harrison, A. van den Berg (Eds.), MicroTotal Analysis Systems '98, Kluwer, Dordrecht, 1998, pp. 249–267.
[63] Y.C. Kwok, N.T. Jeffery, A. Manz, Anal. Chem. 73 (2001) 1748–1753.
[64] S.L.R. Barker, M.J. Tarlov, H. Canavan, J.J. Hickman, L.E. Locascio, Anal. Chem. 72 (2000) 4899–4903.
[65] A. Manz, N. Graber, H.M. Widmer, Sens. Actuators, B 1 (1990) 244–248.
[66] P. Gravesen, J. Branebjerg, O.S. Jensen, J. Micromech. Microeng. 3 (1993) 168–182.
[67] H. Becker, A. Manz, in: H. Baltes, W. Göpel, J. Hesse (Eds.), Sensors Update, vol. 3, VCH, Weinheim, 1998, pp. 208–238.
[68] B.H. Weigl, P. Yager, Science 283 (1999) 346–347.
[69] A.E. Kamholz, B.H. Weigl, B.A. Finlayson, P. Yager, Anal. Chem. 71 (1999) 5340–5347.
[70] P. Yager, C. Cabrera, A. Hatch, K. Hawkins, M. Holl, A. Kamholz, K. Macounova, B.H. Weigl, Analytical devices based on transverse transport in microchannels, in: A. van den Berg, W. Olthius, P. Bergveld (Eds.), MicroTotal Analysis Systems 2000, Kluwer, Dordrecht, 2000, pp. 15–18.
[71] C. Harrington, C. Rosenow, J. Retief, Curr. Opin. Microbiol. 3 (2000) 285–291.
[72] R.C. Anderson, G.J. Bogdan, A. Puski, X. Su, Advances in integrated genetic analysis, in: D.J. Harrison, A. van den Berg (Eds.), MicroTotal Analysis Systems '98, Kluwer, Dordrecht, 1998, pp. 11–15.
[73] Z. Fan, D.J. Harrison, Anal. Chem. 66 (1994) 177–184.
[74] C.S. Effenhauser, A. Manz, H.M. Widmer, Anal. Chem. 65 (1993) 2637–2642.
[75] S.C. Jacobson, R. Hergenröder, L.B. Koutny, J.M. Ramsey, Anal. Chem. 66 (1994) 2369–2373.
[76] H. Becker, W. Dietz, Proc. SPIE 3515 Micro Fluidic Devices and Systems, Santa Clara, 1998, pp. 177–182.
[77] R. Konrad, W. Ehrfeld, H.J. Hartmann, P. Jacob, R. Pommersheim, I. Sommer, Proc. Third Int. Conf. Microreaction Technol., Frankfurt, 1999, pp. 420–429.

[78] Y.H. Chen, S.H. Chen, Electrophoresis 21 (2000) 165–170.
[79] S.H. Chen, G.B. Lee, W.C. Sung, G.R. Huang, Y.H. Chen, K.C. Young, T.T. Chang, Plastic microchip electrophoresis for clinical applications of DNA analysis, in: A. van den Berg, W. Olthius, P. Bergveld (Eds.), MicroTotal Analysis Systems 2000, Kluwer, Dordrecht, 2000, pp. 497–500.
[80] G.B. Lee, S.H. Chen, G.R. Huang, Y.H. Lin, W.C. Sung, Proc. SPIE 4177, Microfluidic Devices and Systems III, Santa Clara, 2000, pp. 112–121.
[81] G. Weber, M. Jöhnck, D. Siepe, A. Neyer, R. Hergenröder, Capillary electrophoresis with direct and contactless conductivity detection on a polymer microchip, in: A. van den Berg, W. Olthius, P. Bergveld (Eds.), MicroTotal Analysis Systems 2000, Kluwer, Dordrecht, 2000, pp. 383–386.
[82] A. Paulus, S.J. Williams, A.P. Sassi, P.K. Kao, H. Tan, H.H. Hooper, Proc. SPIE 3515, Microfluidic Devices and Systems, Santa Clara, 1998, pp. 94–103.
[83] A. Schwarz, J.S. Rossier, F. Bianchi, F. Reymond, R. Ferrigno, H.H. Girault, Micro-TAS on polymer substrates micromachined by laser photoablation, in: D.J. Harrison, A. van den Berg (Eds.), MicroTotal Analysis Systems '98, Kluwer, Dordrecht, 1998, pp. 241–244.
[84] Y. Baba, Nanochip technology for single DNA molecule analysis and human genomic polymorphism analysis, in: A. van den Berg, W. Olthius, P. Bergveld (Eds.), MicroTotal Analysis Systems 2000, Kluwer, Dordrecht, 2000, pp. 467–472.
[85] M. Ueda, Y. Kiba, H. Abe, A. Arai, H. Nakanishi, Y. Baba, Electrophoresis 21 (2000) 176–180.
[86] C.S. Effenhauser, G.I. Bruin, A. Paulus, M. Ehrat, Proc. μTAS'96, Basel, Anal. Methods Instrum. Special Issue, 1996, pp. 124–125.
[87] P.R. Fielden, S.J. Baldock, N.J. Goddard, L.W. Pickering, J.E. Prest, R.D. Snook, B.J. Treves Brown, D.I. Vaireanu, Proc. Micro-TAS'98, Banff, 1998, pp. 323–326.
[88] S.J. Baldock, N. Bektas, P.R. Fielden, N.J. Goddard, L.W. Pickering, J.E. Prest, R.D. Snook, B.J. Treves Brown, D.I. Vaireanu, Isotachophoresis on planar polymeric substrates, in: D.J. Harrison, A. van den Berg (Eds.), MicroTotal Analysis Systems '98, Kluwer, Dordrecht, 1998, pp. 359–362.
[89] G. Ocvirk, M. Munroe, T. Tang, R. Oleschuk, K. Westra, D.J. Harrison, Electrophoresis 21 (2000) 107–115.
[90] Q. Xue, F. Foret, Y.M. Duanyevskiy, P.M. Zavracky, N.E. McGruer, B.L. Karger, Anal. Chem. 69 (1997) 426–430.
[91] C.H. Yuan, J. Shiea, Anal. Chem. 73 (2001) 1080–1083.
[92] Z. Meng, S Qi, S.A. Soper, P.A. Limbach, Anal. Chem. 73 (2001) 1286–1291.
[93] J. Wen, Y. Lin, F. Xiang, D.W. Matson, H.R. Udseth, R.D. Smith, Electrophoresis 21 (2000) 191–197.
[94] R.S. Ramsey, J.M. Ramsey, Anal. Chem. 69 (1997) 1174–1178.
[95] A.T. Woolley, D. Hadley, P. Landre, A.J. deMello, R.A. Mathies, M.A. Northrup, Anal. Chem. 68 (1996) 4081–4086.
[96] M.U. Kopp, A.J. deMello, A. Manz, Science 180 (1998) 1946–1948.
[97] J.M. Köhler, U. Dillner, A. Mokansky, S. Poser, T. Schulz, Proc. Sec. Int. Conf. Microreaction Technol. (IMRET II), New Orleans, 1998, pp. 241–247.
[98] M. Ronaghi, M. Uhlen, P. Nyren, Science 281 (1998) 363–365.
[99] A. Eckersten, et al., High throughput SNP scoring in a dispoable microfabricated CD device, in: A. van den Berg, W. Olthius, P. Bergveld (Eds.), MicroTotal Analysis Systems 2000, Kluwer, Dordrecht, 2000, pp. 521–524.
[100] www.i-stat.com.
[101] J. Virtanen, US Patent 6030581, 2000.
[102] E. Alterndorf, D. Zebert, M. Holl, A. Vannelli, C. Wu, T. Schulte, Results obtained using a prototype microfluidics-based hematology analyzer, in: D.J. Harrison, A. van den Berg (Eds.), MicroTotal Analysis Systems '98, Kluwer, Dordrecht, 1998, pp. 73–76.
[103] N. Thomas, A. Ocklind, I. Blikstad, S. Griffiths, M. Kendrick, H. Derand, G. Ekstrand, C. Ellstrom, A. Larsson, P. Andersson, Integrated cell based assays in microfabricated disposable CD devices, in: A. van den Berg, W. Olthius, P. Bergveld (Eds.), MicroTotal Analysis Systems 2000, Kluwer, Dordrecht, 2000, pp. 249–252.
[104] S. Takayama, J.C. McDonald, E. Ostuni, M.N. Liang, P.J.A. Kenis, R.F. Ismagilov, G.M. Whitesides, Proc. Natl Acad. Sci. USA 96 (1999) 5545–5548.
[105] P.B. Monagham, A. Manz, W.W. Nichols, Microbiology on-a-chip, in: A. van den Berg, W. Olthius, P. Bergveld (Eds.), MicroTotal Analysis Systems 2000, Kluwer, Dordrecht, 2000, pp. 111–114.
[106] R. Pethig, J.P.H. Burt, A. Parton, N. Rizvi, M.S. Talary, J.A. Tame, J. Micromech. Microeng. 8 (1998) 57–63.
[107] X.F. Zhou, J.P.H. Burt, M.S. Talary, A.D. Goater, R. Pethig, Proc. SPIE 4177, Microfluidic Devices and Systems III, Santa Clara, 2000, pp. 241–250.
[108] R.A. Clark, A.G. Ewing, Anal. Chem. 70 (1998) 119–125.
[109] H. Becker, T. Klotzbücher, Proc. Third Int. Conf. Microreaction Technol. 1999, Frankfurt, pp. 102–112.
[110] C. Gärtner, amt Jena, private communication.
[111] R.J. Jackman, D.C. Duffy, E. Ostuni, N.D. Willmore, G.M. Whitesides, Anal. Chem. 70 (1998) 2280–2287.
[112] C.K. Dickey, B.B. Elmore, F. Jones, Proc SPIE 4177, Microfluidic Devices and Systems III, Santa Clara, 2000, pp. 56–64.
[113] T.B. Taylor, P.M. St. John, M. Albin, Micro-genetic analysis systems, in: D.J. Harrison, A. van den Berg (Eds.), MicroTotal Analysis Systems '98, Kluwer, Dordrecht, 1998, pp. 261–266.