Compressed-air flow control system†

Ki Wan Bong,‡[a] Stephen C. Chapin,‡[a] Daniel C. Pregibon,[b] David Baah,[c] Tamara M. Floyd-Smith[d] and Patrick S. Doyle*[a]

Received 17th August 2010, Accepted 4th November 2010
DOI: 10.1039/c0lc00303d

We present the construction and operation of a compressed-air driven flow system that can be used for a variety of microfluidic applications that require rapid dynamic response and precise control of multiple inlet streams. With the use of inexpensive and readily available parts, we describe how to assemble this versatile control system and further explore its utility in continuous- and pulsed-flow microfluidic procedures for the synthesis and analysis of microparticles.

Introduction

Despite the multitude of innovations in the field of microfluidics over the past decade, there remains a need for simple setups that can be quickly and inexpensively implemented for the generation and manipulation of flows in microchannels. Existing setups typically rely on expensive syringe or peristaltic pumps that exhibit poor temporal response and instability in fluid delivery, thus limiting their utility in procedures that require precise multi-port fluid injection and periodic flow stoppage.[1,2] Due to the compression of fluid in the tubing leading into the device, such displacement-driven flow can lead to transients lasting longer than a minute in micron-scale flow systems. Meanwhile, methods that generate complex flows with microfabricated valves and pumps are difficult to implement and are not always compatible with the transported solutions.[3,4] Fluid transport *via* electroosmosis requires strong electric fields ($\sim$kV cm^{-1}) that can lead to local heating of reagents and is difficult to realize for biological samples capable of disrupting the surface charge density of device walls.[2,5] Seeking to move beyond the limitations of these approaches, we developed a pressure-driven control system that enables rapid and precise manipulation of complex microflows.

In this note we describe the fabrication and operation of a simple and inexpensive flow control apparatus that has been used extensively in past work for high-throughput microparticle synthesis,[6–8] as well as high-velocity flow-through microparticle analysis.[9,10] The compressed-air driven flow that we illustrate provides an economical and scalable solution to generating structured microflows with tunable properties for a range of applications. With the details provided, it should be possible for experimentalists with even minimal microfluidic experience to replicate and use this versatile setup.

Experimental

Apparatus construction

The flow control system consists of parts and supplies that can be easily obtained from commercial sources. A compressed air source ($\sim$40 psi) was connected to a high-pressure regulator (150 psi maximum outlet, Dayton, Niles, IL), which in turn was connected to a low-pressure regulator (0.5–25 psi outlet range, Controlair, Inc., Amherst, NH) equipped with a digital gauge (Omega, Stamford, CT). PVC tubing (1/4″ ID, 3/8″ OD, VWR Cat. No. 89068-502) was then used to link this regulator with a three-way solenoid valve (Model 6014, Burkert, Germany), with one outlet left open as a vent to the atmosphere and the other connected *via* PVC tubing to the pressure distribution manifold (Fig. 1). All connections within the manifold consisted of the aforementioned PVC tubing, while junctions were untapered, serrated two- or three-way polypropylene pieces (size 8, VWR Cat. No. 46600-126 and -148, respectively).

Depending on the number of streams to be addressed, a series of control channels were branched off the main supply line, each with its own independent brass needle relief valve (1/8″, Swagelok, Part No. B-ORS2, Billerica, MA) for venting purposes. In previous experiments, up to ten independent control channels were used with no loss in performance at a total driving pressure of 5 psi. The ability to scale the system up for the manipulation of many streams without adding additional pieces of expensive control equipment was highly advantageous.

The thread of the connection point of each valve was tightly wrapped with PTFE tape (Swagelok) to ensure the tightest fit possible with the PVC tubing. Between the main supply line and each relief valve, a three-way junction with a modified outlet (sample arm) was inserted to interface the manifold with the actual microfluidic device. These sample arm outlets consisted of a 1 mL plastic syringe (Becton Dickinson, Franklin Lakes, NJ) cut at the 0.3 mL level and inserted into the PVC tubing, thus exposing the loading tip to the atmosphere. In addition, a modified pipette tip (ART 200, Molecular BioProducts, Inc., San Diego, CA) was inserted between the supply line and each sample arm (Fig. 1) to increase resistance and thus provide

[a]*Department of Chemical Engineering, MIT, 66-053, 77 Massachusetts Ave., Cambridge, MA, 02139, USA. E-mail: pdoyle@mit.edu*
[b]*Firefly BioWorks, Inc., Cambridge, MA, 02139, USA*
[c]*Materials Science and Engineering Program, Tuskegee University, Tuskegee, AL, 36088, USA*
[d]*Department of Chemical Engineering, Tuskegee University, Tuskegee, AL, 36088, USA*

† Electronic supplementary information (ESI) available: Real-time movies showing regulation of stream size and generation of droplets of various sizes; circuit diagram and script for control of solenoid valve. See DOI: 10.1039/c0lc00303d

‡ Authors contributed equally to this work.

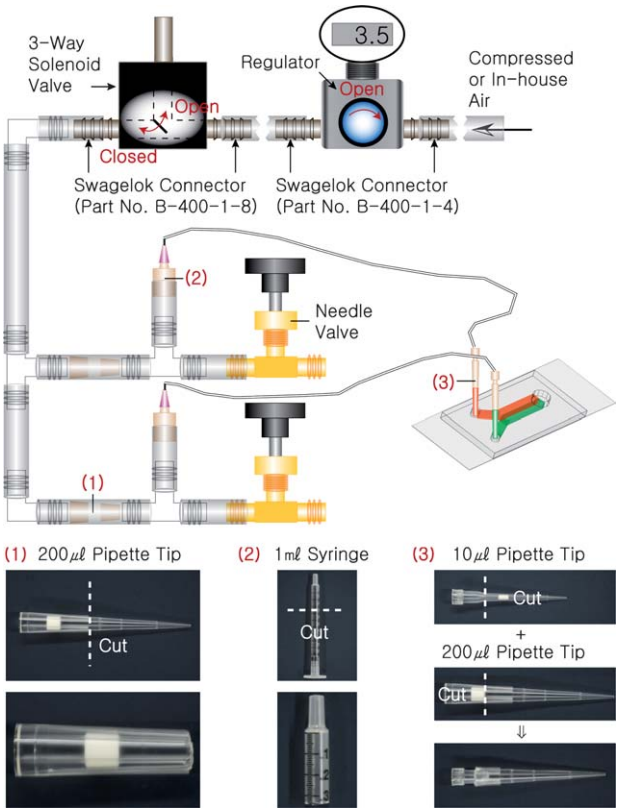

Fig. 1 Schematic of the pressure manifold and its attachment to a two-inlet microfluidic device. Compressed air is downregulated and then passed through a three-way solenoid valve that serves to either pressurize the manifold (open) or vent to the atmosphere (closed). Two control channels, each with its own sample arm and relief needle valve, are pictured branching off the main supply line. Bottom images depict (1) the shortened pipette tip used in the control channel to increase resistance and evenly distribute pressure, (2) the shortened plastic syringe incorporated into the sample arm to enable luer stub connection, and (3) the modified pipette tips utilized for sample loading of PDMS device. In typical pressure ranges (0–15 psi), all connection parts can be sealed by fitting without the use of adhesives.

a more even pressure distribution to all control channels, regardless of position on the supply line.

Sample loading and injection

To interface the pressure-driven flow system with a poly(dimethylsiloxane) (PDMS) microfluidic device, pipette tips (ART 10 Reach and ART 200, Molecular BioProducts, Inc.) were cut and then pushed together to form a sample loading chamber (Fig. 1). Tygon tubing (S-50-HL formulation, 1/32″ ID, 3/32″ OD, VWR Cat. No. 63010-231) was forced into the wide end of the modified pipette tip and capped on the other end with a luer stub adapter (18 gauge, Becton Dickinson). A standard 1 mL plastic syringe was then connected to the stub for drawing sample from an Eppendorf tube. Once the desired amount of fluid had been taken up (5–125 μL range), the pipette tip was inserted into an inlet port punched out of the PDMS with a 15 gauge luer stub adapter (Becton Dickinson), and the loading syringe was removed. This process was repeated for the various streams involved in the flow experiment, with negligible changes in the levels of input fluid over 5–10 min preparation times.

Generally, the loading volumes (5–125 μL) provide one to four hours of synthesis as the typical volumetric flow rate for the system is ~0.01 μL s^{-1}. Under conditions where the running time is longer, or the volumetric flow rate is larger (*e.g.* low viscosity solutions), the loading samples can be refilled. The reloading can be repeated several times as the junction between the pipette tip and the inlet port is reversibly sealed. To reduce the number of reloading operations, the modified pipette tip can be replaced with a 1.7 mL Eppendorf tube. Two holes are punched in the cap of the tube with a scalpel; a Tygon tube is attached to one and provides air pressure control. A 4 cm length of aluminium tubing (1/16″ K&S, Part No. K + S1008) is used to connect the Eppendorf to the PDMS. The aluminium tube is bent in the middle to ~60° and inserted into the other cap hole so that the end of the tube nearly reaches the bottom of the Eppendorf tube. The cap connections are sealed with epoxy to prevent air or liquid leakage.

Upon the completion of sample loading, each stub adapter was connected to a corresponding syringe tip on the manifold. The PDMS device was mounted on an inverted microscope (Zeiss Axio Observer) equipped with a Zeiss Plan Neofluar 20× objective (NA 0.50), and a simple Python script was initialized on a PC for the automated control of the three-way solenoid valve *via* the parallel port connection (see ESI†). Depending on the microflow application, this script could be used to provide a continuous driving pressure (valve open) or to deliver regularly spaced driving pulses (alternation between valve open and closed).

It should be noted that this basic form of the system can be further modified to suit a variety of experimental needs. For example, for equal forcing of two streams, a single control channel may be used by replacing the three-way polypropylene junction within a control channel with a four-way junction connected to two separate sample arms. In this way, the driving pressure of two streams can be simultaneously regulated by one relief valve. Furthermore, to directly monitor the driving pressure for a given stream in real-time, an in-line pressure gauge (Sigma Aldrich, Cat. No. 20469) may be inserted in the sample arm.

Results and discussion

The compressed-air flow control system has been used extensively for the development of structured microflows of polymeric liquids in the synthesis of droplets and complex hydrogel microparticles.

Continuous-flow operation

During continuous-flow operation, streams were injected into PDMS devices at pressures ranging from 0.5–15 psi. As demonstrated in Fig. 2, there was a linear relationship between driving pressure and tracking-bead velocity over the operational range that could be accurately analyzed using our imaging system (KP-M1A, Hitachi, 30 frames s^{-1}).

Food coloring (2–70% (v/v), Durkee) was added to certain streams to provide contrast differences and thus assist in the

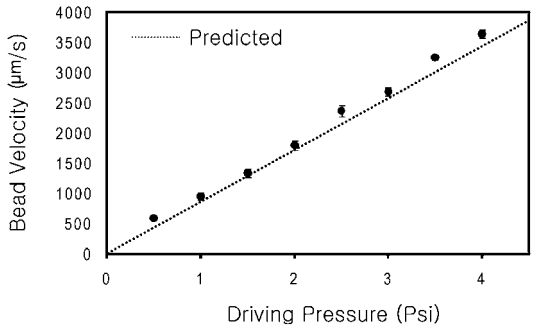

Fig. 2 Demonstration of the linear relationship between forcing pressure and tracking-bead velocity for a one-inlet PDMS channel. A 0.02% solution of 1.6 μm polystyrene beads in poly(ethylene)glycol diacrylate (PEG-DA) 700 was used. The velocity of beads traveling along the channel centerline was measured for a range of inlet pressures and compared to the predicted maximum fluid velocity (see ESI†). Error bars represent standard deviation over 12 measurements for each pressure.

visualization of fluid–fluid interfaces. The relative widths of the streams were precisely and reversibly controlled in real-time by simply adjusting the needle valves (Fig. 3 and Video S1†). Reduction of a particular stream's width was achieved by opening the appropriate valve and relieving a certain percentage of the driving pressure, providing a minimum stable width of ~5 μm. Typical lag time between valve motion and stream change was <1 s. In-line pressure gauges were used to monitor the forcing pressure of each fluid without disrupting ongoing experiments. This robust setup formed the basis for the continuous-flow lithography (CFL) process,[6] in which one or more photopolymerizable streams were injected into a PDMS device at various steady flow rates to generate laminar co-flows (Fig. 3). Polymerization across the moving streams with brief pulses of mask-defined UV light enabled the formation of chemically and

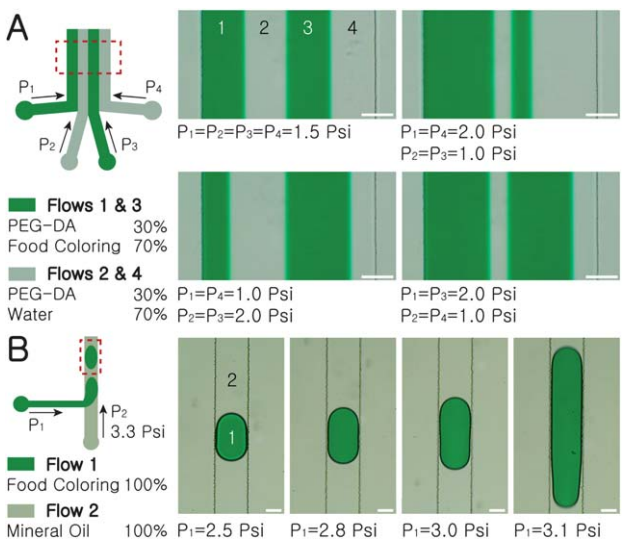

Fig. 3 Dynamic microflow control using relief valves. (A) Widths of co-flowing laminar streams are modulated by adjusting driving pressure of each stream. (B) Droplet size modulation using the compressed-air flow control system. Images taken downstream from a T-junction demonstrate the size range that can be achieved by simple adjustment of the dispersed phase driving pressure. All scale bars are 50 μm.

geometrically complex gel particles that would freely advect with the flow following synthesis. Recent applications of CFL have produced hydrogel microparticles that can be organized into intricate structures for directed assembly,[11] as well as microgels that can be folded into novel drug delivery capsules.[12]

The control system was further utilized to produce monodisperse droplets with flow-focusing channels. Generally, in droplet-based microfluidics, syringe pumps have been used as a driving source to squeeze droplets into immiscible flows using channel geometry.[13] Our system can be implemented for this application for only 20% of the cost of the syringe approach (see ESI†). In addition, as syringes use a rotary motor to control volumetric flow rates, the size variation of droplets can be substantial at low flow rates.[13] Our system exhibited high monodispersity at these reduced rates, and the rapid response of the pressure-driven setup was used to quickly change droplet size during experiments (Fig. 3 and Video S2†).

The system was also used in a continuous-flow mode to control core and sheath streams for high-throughput, two-dimensional flow focusing and alignment of graphically encoded gel particles.[9] In this application, the two streams were independently forced at pressures between 1 and 10 psi to reliably achieve single-file processions of precisely oriented microparticles at rates up to 40 particles s^{-1}. Typical particle velocities were ~50 cm s^{-1}.

Pulsed-flow operation

The rapid pulsing of microflows was explored with this system in order to synthesize particles in a stationary fluid for higher feature resolution and improved throughput. Pulsed flow has proven to be a valuable operational regime for a variety of on-chip applications, including photothermal control of microscale reactions,[14] protein coating of microchannels,[15] and single-molecule analysis.[16] Although syringe pumps have been adapted to produce such flows, their long stabilization time and fluctuation at low flow rates have imposed a number of inconveniences,[16–18] including the need to reverse pumping direction in order to accelerate stoppage,[16] the need to account for the mechanical properties of the pump's thread screw,[16,17] and the need to select materials and components (such as rigid tubing) that will minimize the hysteresis and instabilities introduced by the pump.[18]

We demonstrate here the superiority of our pressure-driven system for the generation of pulsed microflows. Exploiting the quick dynamic response of the compressed-air driving force, regularly spaced flow pulses were easily generated within our PDMS devices using standard elastic tubing and the script-controlled solenoid valve. A simple GUI (see ESI†) was constructed using Python to allow the user to cycle this process automatically through the specification of a flow duration and a stoppage duration. With the channel geometries and PDMS formulations studied, it was found that the rate-determining step in the stoppage of flow was the retraction of the PDMS from its bulged (flow) state to its rectangular cross-section (stop), a process which took ~200 ms.[7]

Rapid alternation between the flow and stoppage states was achieved with this control system. When a pulsing frequency of 0.5 Hz was established, the flow reacted quickly (Fig. 4 and Video S3†). In an analogous experiment with a syringe pump, the same

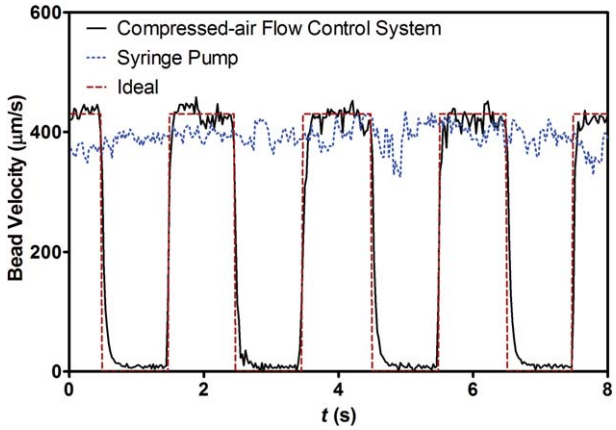

Fig. 4 Comparison of compressed-air system with syringe pump in pulsed-flow operation. Pulsing frequency was fixed at 0.5 Hz for both systems. The compressed-air system exhibited a rapid reaction to the driving force, while the syringe setup largely failed to respond on the time scale investigated.

Table 1 Summary of flow stoppage time

Bead velocity	430 μm s^{-1}	860 μm s^{-1}	1720 μm s^{-1}
Compressed-air flow control system	0.2 s	0.2 s	0.2 s
Syringe pump	350 s	480 s	620 s

periodic variation in the flow velocity was not observed upon automated pulsing (Fig. 4 and Video S3†), demonstrating a clear advantage of using pressure-driven flow. Further experiments investigating the time-to-stop for several initial flow velocities revealed dramatically longer stabilization times for the syringe setup (Table 1). For these trials, flow was considered to be "stopped" when the bead velocity fell below 10 μm s^{-1}. The large lag time of the syringe, particularly at higher flow velocities, precludes the use of such an instrument for high-throughput particle synthesis and other applications that demand rapid alternation between the flow and stoppage states. The observation of a prolonged response time is in agreement with a previous study which found that it takes ~1 h for traditional syringe-based setups to achieve a stable doubling of the flow rate.[18] More sophisticated on-chip "microsyringes" would also be unsuitable for rapid pulsing, as they exhibit delay times on the order of minutes.[19] Although the delay in our pressure-driven system is several orders of magnitude smaller, performance could be further enhanced by modifying the microfluidic device itself. For example, stiffer PDMS formulations and inlet channels with smaller cross-sectional areas could be introduced to reduce the lag time for even greater temporal precision.

Additional experiments synchronized the pulsed flow control with brief, shutter-mediated UV exposures to achieve stop-flow lithography (SFL) of complex, multifunctional microparticles.[7,20,21] Following stoppage, laminar co-flow of up to seven streams was reestablished after 500 ms of pressure-driven flow at 5 psi. Sharp interfaces between adjacent chemistries on the resultant gel particles indicated minimal diffusion between streams during the brief stoppage period in which polymerization took place.[8]

In recent work pulsed-flow operation was utilized to generate highly structured microflows for the creation of particles with complex three-dimensional morphology. With the control afforded by the pressure manifold, rapid fluid exchanges (<1 s) were achieved within microfluidic channels for the synthesis of composite particles with spatially configurable chemistries.[22] In work by Lee *et al.*, an unspecified flow-control system was used to exchange fluids in a microchannel for the creation of highly complex, color-barcoded microparticles *via* lithographic patterning.[23] Additional trials in our group used the control system for three-dimensional flow focusing to create stacked co-flows in two-layered PDMS channels.[24] Particles synthesized from these structured flows featured chemical anisotropy in two separate dimensions. Finally, the control system was implemented for the simultaneous manipulation of synthesis streams and an integrated perfusion/washing stream to generate magnetically addressable gel particles with precise localization of magnetic material.[25]

Acknowledgements

This research was supported by, or supported in part by, the US Army through the Institute for Soldier Nanotechnologies, under Contract DAAD-19-02-D-0002 with the US Army Research Office, Grant R21EB008814 from the National Institute of Biomedical Imaging and Bioengineering, NIH, Kwanjeong Educational Foundation, and National Science Foundation Grant DMR-10066.

References

1 H. A. Stone, A. D. Stroock and A. Ajdari, *Annu. Rev. Fluid Mech.*, 2004, **36**, 381–411.
2 C. Futterer, N. Minc, V. Bormuth, J.-H. Codarbox, P. Laval, J. Rossier and J.-L. Viovy, *Lab Chip*, 2004, **4**, 351–356.
3 M. A. Unger, H.-P. Chou, T. Thorsen, A. Scherer and S. R. Quake, *Science*, 2000, **288**, 113–116.
4 A. Y. Fu, H.-P. Chou, C. Spence, F. H. Arnold and S. R. Quake, *Anal. Chem.*, 2002, **74**, 2451–2457.
5 S. L. R. Barker, D. Ross, M. J. Tarlov, M. Gaitan and L. E. Locascio, *Anal. Chem.*, 2000, **72**, 5925–5929.
6 D. Dendukuri, D. C. Pregibon, J. Collins, T. A. Hatton and P. S. Doyle, *Nat. Mater.*, 2006, **5**, 365–369.
7 D. Dendukuri, S. S. Gu, D. C. Pregibon, T. A. Hatton and P. S. Doyle, *Lab Chip*, 2007, **7**, 818–828.
8 D. C. Pregibon, M. Toner and P. S. Doyle, *Science*, 2007, **315**, 1393–1396.
9 S. C. Chapin, D. C. Pregibon and P. S. Doyle, *Lab Chip*, 2009, **9**, 3100–3109.
10 S. C. Chapin, D. C. Appleyard, D. C. Pregibon and P. S. Doyle, 2010, submitted.
11 S. E. Chung, W. Park, S. Shin, S. A. Lee and S. Kwon, *Nat. Mater.*, 2008, **7**, 581–587.
12 T. S. Shim, S. H. Kim, C. J. Heo, J. H. Choi and S. M. Yang, *MicroTAS Conference Proceedings*, 2010, pp. 1850–1852.
13 T. Ward, M. Faivre, M. Abkarian and H. A. Stone, *Electrophoresis*, 2005, **26**, 3716–3724.
14 Y. Tanaka, M. N. Slyadnev, A. Hibara, M. Tokeshi and T. Kitamori, *J. Chromatogr., A*, 2000, **894**, 45–51.
15 A. Lionello, J. Josserand, H. Jensen and H. H. Girault, *Lab Chip*, 2005, **5**, 1096–1103.
16 K. Jo, Y. L. Chen, J. J. de Pablo and D. C. Schwartz, *Lab Chip*, 2009, **9**, 2348–2355.
17 D. Kim, N. C. Chesler and D. J. Beebe, *Lab Chip*, 2006, **6**, 639–644.
18 J. Goulpeau, V. Taniga and C. A. Kieffer, *Mater. Res. Soc. Symp. Proc.*, 2009, **1191**, 67–72.

19 R. Yokokawa, T. Saika, T. Nakayama, H. Fujita and S. Konishi, *Lab Chip*, 2006, **6**, 1062–1066.
20 P. Panda, S. Ali, E. Lo, B. G. Chung, T. A. Hatton, A. Khademhosseini and P. S. Doyle, *Lab Chip*, 2008, **8**, 1056–1061.
21 R. F. Shepherd, P. Panda, Z. Bao, K. H. Sandhage, T. A. Hatton, J. A. Lewis and P. S. Doyle, *Adv. Mater.*, 2008, **20**, 4734–4739.
22 K. W. Bong, D. C. Pregibon and P. S. Doyle, *Lab Chip*, 2009, **9**, 863–866.
23 H. Lee, J. Kim, H. Kim, J. Kim and S. Kwon, *Nat. Mater.*, 2010, **9**, 745–749.
24 K. W. Bong, K. T. Bong, D. C. Pregibon and P. S. Doyle, *Angew. Chem., Int. Ed.*, 2010, **49**, 87–90.
25 K. W. Bong, S. C. Chapin and P. S. Doyle, *Langmuir*, 2010, **46**, 8008–8014.