BAKING-POWDER DRIVEN CENTRIPETAL PUMPING CONTROLLED BY EVENT-TRIGGERING OF FUNCTIONAL LIQUIDS

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ABSTRACT

This paper reports radially inbound pumping by the event-triggered addition of water to on-board stored baking powder in combination with valving by an immiscible, high-specific weight liquid on a centrifugal microfluidic platform. This technology allows making efficient use of precious real estate near the center of rotation by enabling the placement of early sample preparation steps as well as reagent reservoirs at the spacious, high-field region on the perimeter of the disc-shaped rotor. This way the number of process steps and assays that can be integrated on these of this "Lab-on-a-Disc" (LoaD) cartridge can be significantly enhanced while maintaining minimum requirements on the intrinsically simple, spindle-motor based instrumentation.

INTRODUCTION

By now LoaD platforms have shown great benefit for sample-to-automation of bioanalytical assays for a manifold of applications such as point-of-care diagnostics [1-3]. The underlying centrifugal microfluidic liquid handling scheme allows actuation by a simple, low-cost spindle motor. An inherent feature of this paradigm is the unidirectional nature of the radially outbound centrifugal field which severely constricts the number of assay steps such as metering and mixing that can be integrated on the limited real estate of a typically CD-sized LoaD cartridge. Furthermore, upstream sample preparation processes, such as blood centrifugation, and reagent reservoirs must be placed near the centre of the disc where space is most precious and the centrifugal field lowest.

To mitigate these limitations, a number of methods for pumping against the centrifugal field have been developed. These can include the addition of energy from external sources, transfer of energy to the liquid through the spindle motor and the storage of energy on the disc. External energy sources include connection to air bottles [4], the use of a thermal heat source to expand trapped gas (and thus displace liquid radially inwards) [5] and the use of external electrical energy to electrolytically displace liquid [6].

These methods tend to increase the complexity of instrumentation with respect to the system-innate spindle motor. Centrifugo-pneumatic pumping has been used to transiently story centrifugal energy in a compressed gas volume on the disc; at a reduced spin rate, the gas expands to centripetally pump liquid without need of any additional instrumentation [7]. While conceptually simple and elegant, this mechanism relies on highly 'dynamic' pumping implemented by powerful spindle motors for rapid changes of the spin rate during compression and expansion of the enclosed gas volume. Similarly, this pumping mechanism is largely enabled through use of high-resistance microchannels and, similarly, its efficiency is largely based on ratio of the flow resistances between inlet and outlet microchannels.

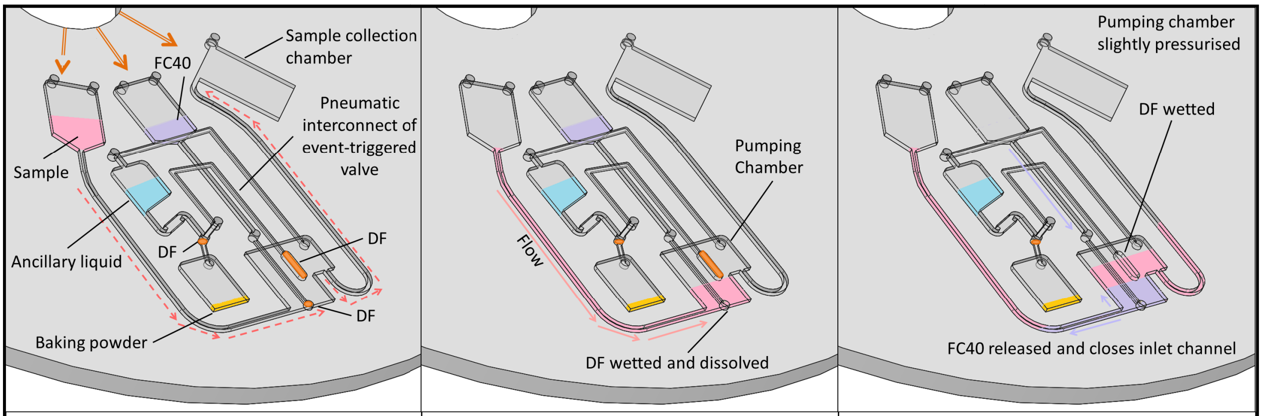
Potential energy has also been stored on-disc. Positive [8] displacement pumping uses an ancillary liquid to push a sample radially inwards. This can be implemented using an intermediary air pocket to ensure that the liquids do not come in contact. In a similar approach, negative [9] liquid displacement-based pumping, an ancillary liquid is generates an underpressure which draws a sample radially inwards. While promising, these pneumatic methods are linked to rather sensitive sample loading procedures. Recently, variations on this scheme have been introduced where relatively heavy immiscible ancillary liquids, such as oils [10] and fluorocarbons 11], have been used to displace samples radially inwards based through directly contacting the samples. This approach can also be used to accurately meter the samples during the pumping operations. However, a drawback to each 'potential energy' storage mechanism is that the ancillary liquid consumes valuable on-disc real-estate; while the most efficient pumping will be enabled by the ancillary liquid being located radially inwards.

In this work we present a pumping mechanism based on storage of chemical energy in ubiquitous, low-cost baking powder [12]. This pumping mechanism is governed by event-triggered control flow [13] and thus making it broadly independent of spin rate. Therefore the mechanism does not impact upstream and downstream Laboratory Unit Operation (LUOs) such as mixing and metering. Additionally, the pumping structure occupies minimal space and, connected via pneumatic channels, can be located at arbitrary locations on the disc.

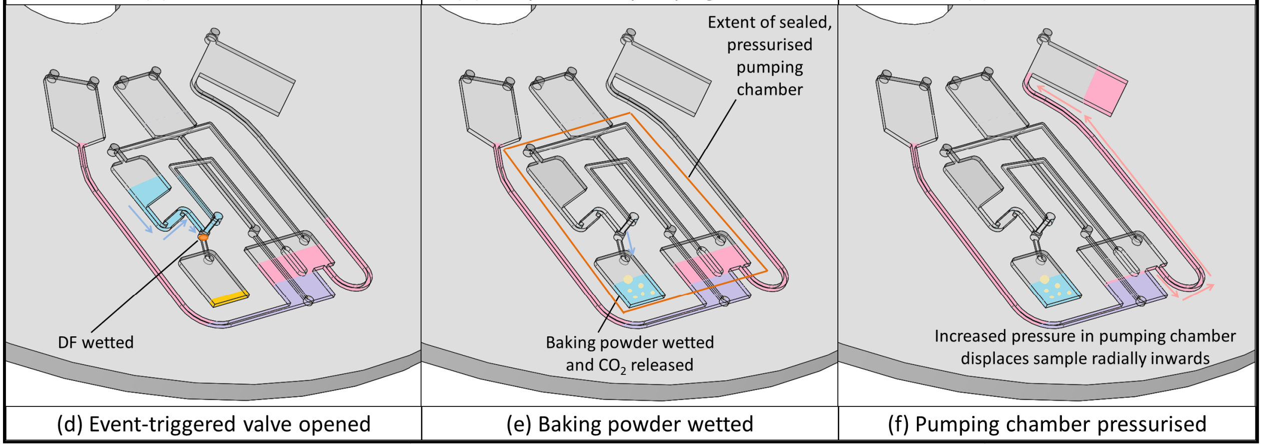
OPERATION AND METHODS

Operating Principle

In our pumping concept (Fig. 1), the release of CO_2 from on-board stored baking powder [10] pressurizes a chamber to drive centripetal pumping. The pumping chamber is composed of four compartments; one containing the baking powder, one the ancillary liquid (water), the third contains a heavy immiscible liquid (Fluorocarbon FC-40; specific gravity ~1.85) while the fourth chamber transiently stores the sample. The FC-40 compartment is sealed by a water dissolvable film (DF). The other chambers are in pneumatic communication without allowing the interchange of liquids. When the sample enters the pumping chamber, the DF (called the VF)



(a) Disc loaded | (b) Sample enters pumping chamber | (c) FC40 released



(d) Event-triggered valve opened | (e) Baking powder wetted | (f) Pumping chamber pressurised

Figure 1 - Centripetal pumping structure. Note in the multilayer disc shown in Figure 2 some connecting channels are hidden as opaque materials are used during manufacture. (a) Disc-stored reagents. The orange arrows indicate the direction of centrifugal force and the dashed red line shows the nominal path of the sample. (b) Upon spinning the sample is centrifugally driven into the pumping chamber. Note the loading chamber is open to atmosphere and the pumping mechanism is triggered by the presence of the sample. Thus the centripetal pump is independent of spin-protocol. (c) FC-40 is released and closes the inlet channel. (d) The pumping liquid is displaced upwards and the event triggered valve is actuated. (e) The release the ancillary liquid (DI Water) to wet the baking powder and trigger the release of CO_2. Note the solid orange triangle represents the extent of the pressurised pumping chamber. (f) The arrival and centrifugal stabilization of the high-density FC-40 effectively seals the inlet channel so the emerging gas expansion displaces the sample through the only outlet radially inwards.

(a) Sample loaded | (b) FC-40 released | (c) Ancillary liquid released | (d) Centripetal pumping | (e) Pumping complete

Figure 2: Centripetal Pumping. (a) Sample loaded and flowing radially outwards. (b) Clear liquid at the base of the pumping chamber is FC-40 has a higher specific weight than water so it is layered by the centrifugal field at the bottom where it thus effectively seal the inlet channel. (c) Ancillary liquid is released, activates the baking powder to generate CO_2. (d-e) Gas expands to centripetally pump the sample through the radially inbound channel.

dissolves and releases the FC-40. This liquid flows underneath the sample to fill the inlet channel. Through dissolving the control film (CF), the sample then opens the event-triggered valve and liquid is released to activate the baking powder. The subsequent release of CO_2 increases the gas pressure in the pumping chamber. However, due to the differing densities between the FC-40 (filling the inlet chamber) and sample (filling the outlet chamber), only the sample is pumped radially inwards. Thus the FC-40 acts akin to a check valve, preventing backflow of the sample during pumping.

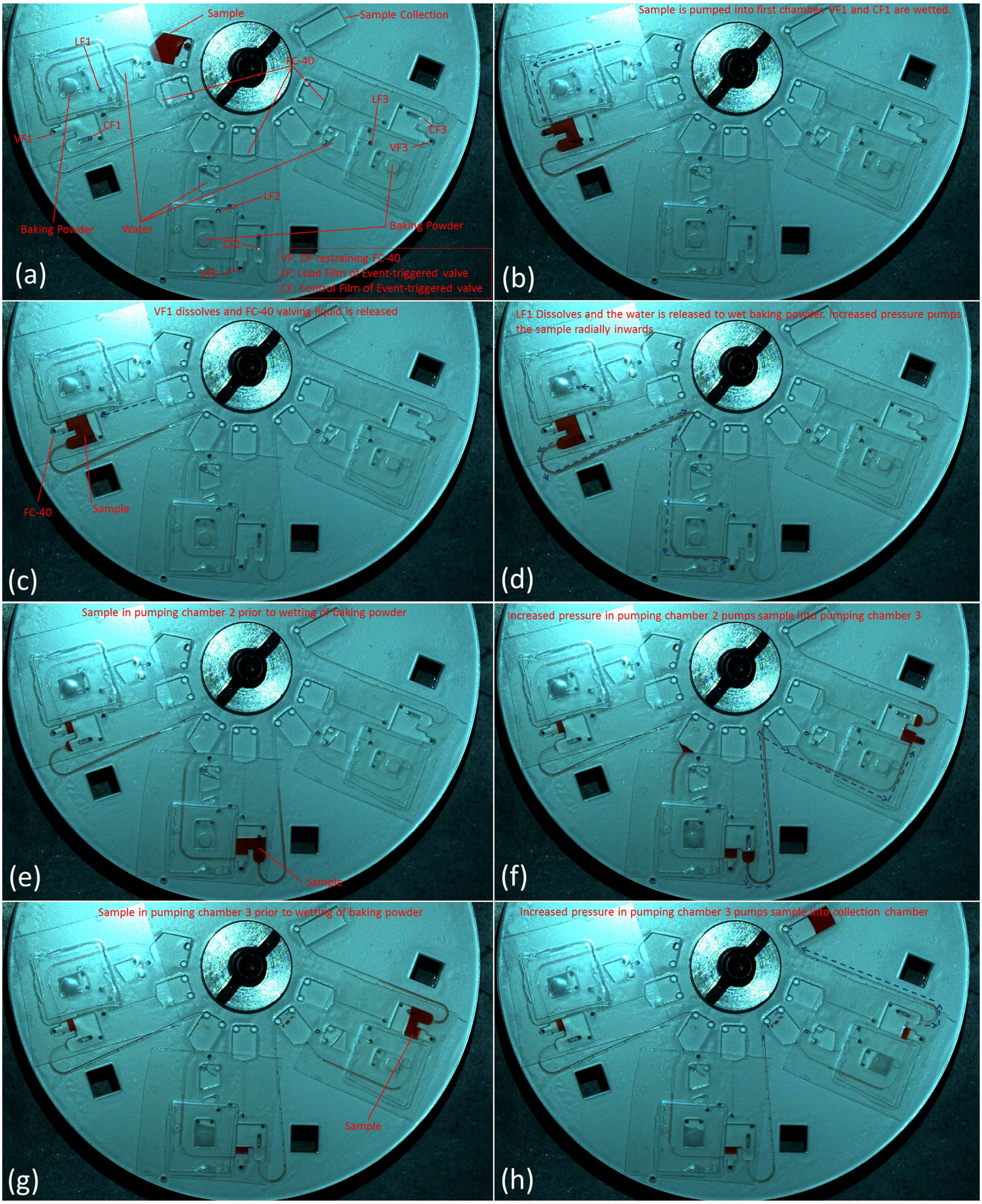


Figure 3: Repeated Centripetal Pumping of a Sample (a) Sample is loaded and flowing radially outwards. (b) Sample enters the first pumping chamber and wets DFs (VF restraining the FC-40 valving liquid and CF of an event-triggered valve). (c) the FC-40 (Clear liquid) is released and blocks the inlet channel. (d) Ancillary liquid (water) is released, activates the baking powder to generate CO_2 and pumps the sample radially inwards and into pumping chamber 2 (e-h) Process is repeated through pumping chamber 2 and pumping chamber 3 until the sample enters a collection chamber located radially inwards. Note this pumping occurs at a constant disc spin rate and is triggered only by the entry of the liquid into the pumping chambers.

Fabrication

The disc is prototyped by stacking multiple, specifically designed adhesive and structural layers as previously described [11]. Briefly, the disc is manufactured from laminates of PMMA bonded with pressure sensitive adhesive (PSA). DFs are mounted to block vertical vias where required. Note also that 'lower level' microchannels (not visible in the images here) provide liquid and pneumatic vias.

CONCLUSIONS AND OUTLOOK

As shown in Figure 2, this pumping mechanism can be used to pump a liquid, initially located at the centre of the disc, to the periphery and back to a radially central location. Figure 3 demonstrates the implementation of this mechanism in a series of three pumping chambers. Here, with the disc rotating at a constant spin rate, the sample is pumped inwards and outwards three times.

The pumping mechanism presented here expands the capabilities of the centrifugal platform in a number of ways. As the pump is only triggered by the presence of the sample, the structure operates widely independent of the spin rate. Thus, and unlike most previous implementations of centripetal pumping, we represent a module which can readily be inserted at any point of an on-disc workflow. Additionally, this pump takes up a comparatively little of space and, due to pneumatic connecting channels, the ancillary liquid and baking powder can be located at remote, arbitrary locations on the disc cartridge. Similarly, unlike displacement based (potential energy) pumping methods, the module saves valuable real-estate near the centre of the disc. While some sample is lost during pumping (representing a lack of efficiency), this is primarily owed to our present system design and prototyping methods.

These pumps have further application towards storage of reagents on the periphery of the disc. Here, the ancillary liquid could be released to wet baking powder at a pre-determined rotational frequency. This variant significantly increases the on-disc real-estate available for reagent storage and permits the spin rate controlled release of reagents.

REFERENCES

[1] Gorkin, R., Park, J., Siegrist, J., Amasia, M., Lee, B. S., *et al.,* "Centrifugal microfluidics for biomedical applications" *Lab on a Chip*, vol. 10, pp. 1758-1773, 2011.

[2] Ducrée, J., Haeberle, S., Lutz, S., Pausch, S., von Stetten, F. *et al* "The centrifugal microfluidic Bio-Disk platform", *Journal of Micromechanics and Microengineering*, vol. 17 pp 103, 2007.

[3] Madou, M., Zoval, J., Jia, G., Kido, H., Kim, J *et al.,* "Lab on a CD", *Annu. Rev. Biomed. Eng.*, vol. 8, pp 601-628, 2006.

[4] Kong, M. C., and Salin, E. D. "Pneumatically pumping fluids radially inward on centrifugal microfluidic platforms in motion", *Analytical chemistry*, vol. 82, pp. 8039-8041, 2010.

[5] Abi-Samra, K., Clime, L., Kong, L., Gorkin III, R., Kim, *et al.,* "Thermo-pneumatic pumping in centrifugal microfluidic platforms", *Microfluidics and nanofluidics*, vol. 11, pp. 643-652, 2011.

[6] Noroozi, Z., Kido, H., and Madou, M. J. "Electrolysis-induced pneumatic pressure for control of liquids in a centrifugal system", *Journal of The Electrochemical Society*, vol. 158, pp. 130-P135, 2011.

[7] Zehnle, S., Schwemmer, F., Roth, G., von Stetten, F., Zengerle, R. *et al.,* "Centrifugo-dynamic inward pumping of liquids on a centrifugal microfluidic platform", *Lab on a Chip*, vol. 12, pp. 5142-5145, 2012.

[8] Kong, M. C., Bouchard, A. P., & Salin, E. D. "Displacement pumping of liquids radially inward on centrifugal microfluidic platforms in motion", *Micromachines*, vol. 3, pp.1-9, 2011.

[9] Soroori, S., Kulinsky, L., Kido, H., and Madou, M. "Design and implementation of fluidic micro-pulleys for flow control on centrifugal microfluidic platforms" *Microfluid Nanofluid,* vol. 16, pp. 1117-1129, 2014.

[10] Kilcawley, N., Kinahan, D., Nwankire, C., Glynn, M., and Ducrée, J. "Buoyancy-driven centripetal pumping for nested sample preparation in bioassays" In *Proceedings of the 18th International Conference on Miniaturized Systems for Chemistry and Life Sciences, October 26-30, 2014,* San Antonio, Texas, USA.

[11] Kim, T.H, Park, J. and Cho, Y.K., "Fully Integrated Molecular Diagnostics of Pathogenic Microorganisms on a Disc" in *Proceedings of the 18th International Conference on Miniaturized Systems for Chemistry and Life Sciences,* October 26-30, 2014, San Antonio, Texas, USA.

[12] Ahn, Chong H. "Disposable Polymer "Smart" Lab-on-a-Chip for Point-of-Care Testing (PocT) in Clinical Diagnostics", in *Proceedings of The 13th International Conference on Solid-State Sensors, Actuators and Microsystems*, Seoul, Korea, June 5-9, 2005.

[13] Kinahan, D. J., Kearney, S. M., Dimov, N., Glynn, M. T., and Ducrée, J. "Event-triggered logical flow control for comprehensive process integration of multi-step assays on centrifugal microfluidic platforms", *Lab on a Chip*, vol. 14, pp. 2249-2258, 2014.

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