# Pico Liter Dispensing Using Liquid Bridge Breakup in Immiscible Fluid

Dustin Moon, Do Jin Im, Taewoong Uhm, and In Seok Kang

***Abstract*—A simple and robust way is devised to generate picoliter droplets out of a single microliter drop for the use of generating monodisperse droplets in droplet-based microfluidics. A single aqueous drop is placed between two glass substrates and immersed in silicone oil, to form a liquid bridge. Then one substrate is moved with a constant velocity ranging from 50 to 1000 μm/s. As the distance between two glass plates increases, the liquid bridge breaks up and smaller droplets or satellites are formed. It is found that, for the case of fixed outer fluid, the droplets of nearly the same size are generated over several orders of moving velocity. However, the size of a satellite droplet increases as the mother drop size increases, and it decreases as the outer fluid viscosity increases. Based on this result, a picoliter-droplet was successfully dispensed repeatedly for 100 times within 2% relative standard deviation, on-demand. To confirm its feasibility of single particle encapsulation, a single polystyrene microparticle has been captured successfully using this method without complex control.**

***Index Terms*—Satellite formation, drop formation, drop-on-demand, picoliter dispensing.**

## I. Introduction

Droplet-based microfluidics offers a wider range of application over conventional or continuous analytical techniques [1]. By encapsulating reagents in drops, advanced control of reaction kinetics can be obtained. Physical and chemical isolation of droplets eliminate the risk of cross-contamination. Droplets of reagent can be transported faster, or more precisely than conventional microfluidic devices with controlled contents [2]. These merits result in the increase of areas of applications. However, in order to further utilize droplet microfluidics, stable generation of ultrasmall droplets should be first established [3].

There have been many researches on the conventional droplet generating methods. Monodisperse droplets can be generated using T-junctions, flow focusing, or electro hydrodynamic method [4, 5]. These methods can generate thousands of uniform droplets in seconds. To generate droplets, only when needed with definite number of droplets, or on-demand, electric pulse, piezo-eletric or thermal methods are commonly used [6-8]. Both of these methods generate droplets with the size of the nozzle i.e., smaller drops can be made using smaller nozzle, limiting its use for more general purposes including cell or particle dispensing. Among number of papers for cell printing based on these methods, only few can stably generate single-cell-encapsulated droplets, without cell rupture [9].

For most cases mentioned above, additional devices such as syringe pump, high-voltage power supply or additional processes are required to make droplets inside a channel. For mobile use of microfluidic devices, smaller and simpler device for generating only few droplets on-demand without need of additional equipments are required.

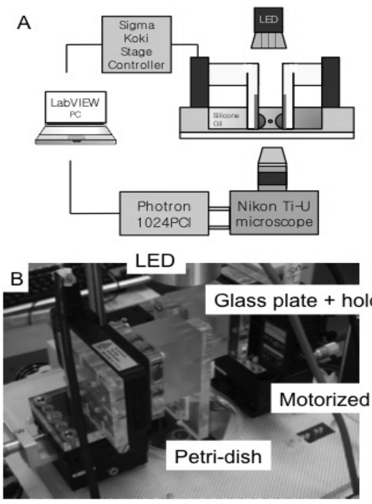


Fig. 1. (A) Schematic diagram of the dispensing system. (B) Dispensing system on inverted microscope for observation

Development of a dispensing method using a single drop - only one or less microliter amount of unknown fluids to generate smaller volume in a simple device would utilize the advantages of droplet microfluidics for portable use. In this paper, we suggest a new method of using satellites formed during liquid bridge breakup to generate picoliter droplets of aqueous solution in oil phase.

The primary purpose of this work is to investigate the formation of satellite and to see the effects of the outside fluid viscosity, the mother drop size, and the elongation rate of liquid bridge for further use of encapsulation

## II. Experimental Section

### *A. Experimental Setup*

The satellite formation is a multi-scale problem in both the time scale and the length scale [10]. To fulfill the above conditions, the experimental setup was on an inverted microscope (Nikon Eclipse Ti-U) with a high-speed imaging device (Photron 1024 PCI), as shown in Fig. 1 (A) and (B). Microslide glasses (Corning, single frosted, pre-cleaned)

Manuscript received December 25, 2011; revised February 9, 2012. This work was supported by the grant R01-2010-0027653 from National Research Foundation (NRF) of Korea, and by the BK21 program of the Ministry of Education, Science and Technology (MEST) of Korea.

The authors are with the Department of Chemical Engineering, POSTECH, San 31 Hyojadong Namgu Pohang, Republic of Korea (e-mail: heavymix@ postech.ac.kr).

were cut into 5 mm by 25mm strips, each bonded to a transparent holder, as hydrophilic plate to hold the mother drop and split the liquid bridge. All the glass plates were cleaned with isopropyl alcohol and air-dried.

Velocity controllable motorized stages (Sigma Koki, SGSP2020) were used with LabVIEW via RS-232 protocol. Due to the high magnification microscope on a high-speed camera, a 5 W white light emitting diode (LED) light source was used to provide enough backlight without heating instead of the front-side mercury light source. All the experiments were done in room temperature, and mechanical movement was slow enough not to heat up the oil by viscous dissipation.

In droplet microfluidics, silicone oil is commonly used due to its chemical stability and various controllable viscosities and transparency for optical observation. Therefore, silicone oil (Shin Etsu KF-96, 10, 50, 100 cSt, Dow Corning DC200F, 6 cSt) was used as outer fluid in this experiment. Silicone oil with very low viscosity (kinematic viscosity $\nu$< 5 cSt) was also available but due to its low vapor pressure, alkane hydrocarbon was used instead. N-dodecane (Alfa Aesar) has relatively low viscosity ($\nu$= 1.79 cSt), with high boiling point (Tbp > 489 K) and thus suitable for the use in digital microfluidics.

## B. Dispensing Procedure

The droplet dispensing procedure is as follows. On one side of the glass plate, a single drop of given volume is placed using a micropipette (Eppendorf, single channel). Using a motorized stage, either plate approaches to the other plate or a drop. A drop contacts the other plate, forming a liquid bridge. Depending on the viscosity of the outer fluid, few seconds of stabilization time is needed to form a liquid bridge. After the liquid bridge is stabilized, plates move apart with constant velocity ranging from 50 to 1000 μm/s. After it reaches critical length depending on the surface tension, liquid bridge breaks up, and a satellite is formed leaving two mother drops on each plate. Then the plate stops, and repeats the whole process for further dispensing.

Velocity can be applied in different profiles, i.e., constant velocity, constant elongation rate, constant acceleration, but since the breaking and collapsing takes in O(10-3) seconds, different profiles of velocity can approximated as constant velocity for a short time. Therefore in this experiment, plates were moved with constant velocity.

High-speed images were captured using Photron PCI 1024X, which is capable of capturing up to 109,500 frames-per-second (fps). The full-frame resolution was possible only for the speed up to 1000 fps. Higher capturing frequency reduces the size of the region-of-interest (ROI) and also requires more light. To capture the whole system 1000 fps with full frame were used and for detailed dynamics up to 18,000 fps were used.

The obtained images were analyzed using LabVIEW with Vision Assistant and ImageJ. For accurate analysis, droplet volume and moving distance were crosschecked using both programs. Average size of satellites is O(1) ~ O(10) pixels in radius, resulting in few pixels of measurement error. All the measurements were averaged over 3 times of measurement.

## III. Results and Discussions

The liquid bridge breakup and satellite formation is a process balanced between viscous force and surface tension force. The overall dynamics can be characterized by two nondimensional numbers - Capillary number and Ohnesorge number [10],

$$Ca = \frac{\mu V}{\sigma} \tag{1}$$

$$Oh = \frac{\mu}{\sqrt{\rho \sigma D}} \tag{2}$$

where ν, V, σ, D and ρ is dynamic viscosity, velocity, interfacial tension, drop diameter and density, respectively.

With the characteristic length D = 1 mm, the interfacial tension between silicone oil and water = 40 mN/m, the kinematic viscosity 1 cSt < $\nu$< 100 cSt, and the glass plate moving velocity 50 m/s < V < 1000 μm/s, Ca ranges from O(10-4) to O(1) and Oh ranges from O(10-3) to O(1). This shows that the surface tension effect is dominating.

In this regime, the capillary time scale is determined only with its fluidic properties, not by the mechanical movement and is the characteristic time scale of the overall process and this is less than O (10-3) s. This capillary time scale could be experimentally confirmed via high-speed imaging. Both in the 6 cSt and 100 cSt outer fluid cases, overall process takes place within milliseconds, confirming the overall time scale. Within this range of outer fluid viscosity range, the droplet formation process is irrelevant to the plate elongation rate, but relevant to its fluidic properties – surface tension and viscosity.

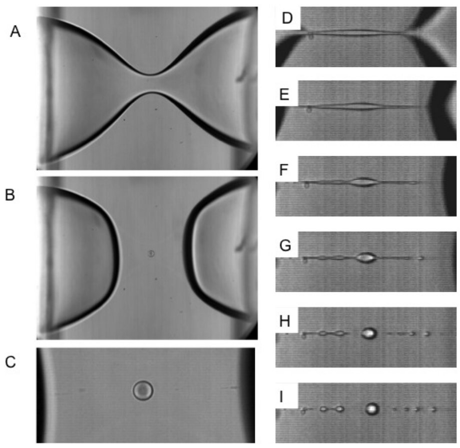


Fig. 2. (A)-(C) Satellite formation in 6 cSt silicone oil. time step between (A) and (B) is 1/1000 s. (D)-(I) Thread necking and collapsing in 100 cSt silicone oil. time step between images is 1/2250 s.

As shown in Fig. 2, the detailed dynamics of the liquid bridge breakup and satellite formation can be divided into four stages - bridge elongation stage, unstable thread formation stage, breakup stage, and collapsing stage, As the liquid bridge elongates, the minimum radius is at the center of the liquid bridge and it decreases. The triple contact line moves inward so that the liquid bridge can have a more stable shape. At a certain point, it moves to the second stage, where the minimum radius decreases spontaneously, i.e., without liquid bridge elongation. In this stage the minimum radius is not at the center but near the mother drop at each end [11-13].

From these points, a drop is divided into three parts of two mother drops and a middle thread. The last step and the only different step among various viscosities is the collapsing step. After the thread is detached from the two mother drops, it relaxes back to its equilibrium shape. Depending on the Oh and viscosity ratio between drop and external fluid, the thread can either shrink into a relatively larger drop and several smaller droplets or divide into several equal size droplets. Thread shape is irrelevant to the flow or the plate velocity but closely related to the free surface and the boundary of the liquid bridge. Therefore, once the Oh number is fixed, the detached thread volume as well as the resulting satellite volume is determined, regardless of plate velocity.

To observe the effect of mother drop size and the effect of viscosity, three different sizes of drops – 0.5, 1, 2 μL – were initially positioned on the glass plate. The generated droplet radii were 12.3, 24.2, and 34.6 μm respectively in 50 cSt silicone oil, 33.8, 40.8, and 70.3 μm respectively in 6 cSt silicone oil. As the mother drop size increases, the satellite size also increases. However, it is difficult to find a general law on this since drops larger than 2 μL are too large to be held on glass plate without other treatment, and mother drops smaller than 0.5 μLgenerate too small satellite which is hard to observe. Still, on each case in this regime, droplets were generated within 9% relative standard deviation in radius and except for the 2 μL in 50 cSt case, all the error were with in 1,2 pixels, mainly caused by measurement error. On 2 μL in 50 cSt case, triple contact line movement was retarded due to the viscous outer oil, causing larger error than other cases which triple contact line doesn't move.

Effect of plate velocity was also observed. From 50 μm/s to 1000 μm/s, satellites were stably generated on average 40.64 μm and 24.1 μm with 2.01% and 3.17% relative standard deviation. It is clearly shown that over several orders of magnitude of elongation rate, nearly uniform sizes of primary satellites are generated.

Droplets generated from higher viscosity external fluid tends to be smaller than those from lower viscosity oil. This is mainly because threads formed in higher viscosity external fluid tend to breakup into many droplets while in threads in lower viscosity oil tend to relax back to a spherical shape before capillary breakup.

Also, the thread volume itself is different depending on the viscosity ratio between drop and external fluid. In high viscosity external fluid case, due to the high viscosity of the external fluid, it took longer time to refill, or push out the fluids inside the liquid bridge, retarding the transition from single minimum radius to double minimum radius points. As the result, drops in high viscosity external fluid generated a longer aspect ration thread with smaller average radius, which is eventually divided into smaller droplets. In the 6 cSt external fluid case, thread breaks into one main satellite and few smaller droplets.

Since using lower external fluid viscosity generates larger, and fewer satellites, by using even lower viscosity fluid single droplet generation could be achieved. N-dodecane was used instead of low viscosity silicone oil. Fig. 3 shows the droplet size for 100 droplets in n-dodecane at constant plate velocity of 1000 μm/s.

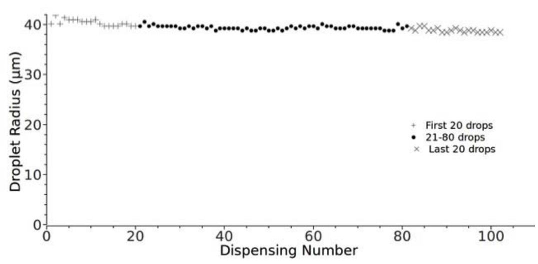


Fig. 3. Droplet size during 100-drop-dispening out of a single 1μL droplet.

All droplets were generated sequentially from a single mother drop, without any other extra control than given back and forth movement by the motorized stage. Average radius of generated droplets is 39.46 μm with 1.73% relative standard deviation. During the 100-droplet-dispensing only single satellites were observed. This result shows the potential of this method as a picoliter dispensing method on-demand, using only 1 μL without any complex fabrication techniques or active feedback control. Most other drop-on-demand methods besides this method requires either complicated fabrication technique for the nozzle, precise measurement or control on the dispensing sample or additional devices such as laser or high voltage power supply. Slight decrease of satellite volume during the dispensing occurs due to the decrease of mother drop volume. Average droplet radius of the first 20 droplets was 40.37 μm while the average radius of last 20 droplets was 38.79 μm. Since one satellite volume is 257 pL, during the 100-droplet-dispensing, the decreased volume (25.7 nL, 7%) is eventually affecting the satellite volume. During the dispensing, motorized stage was moving without additional feedback control. After 100-droplet-dispensing, due to the volume decrease, motorized stage had to be repositioned for stable coalescence.

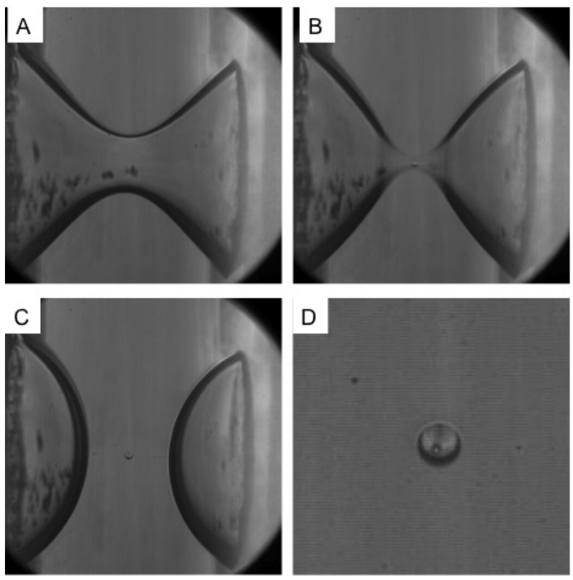


Fig. 4. Single encapsulation of 11 μm polystyrene microparticle.

To confirm its viability toward single cell encapsulation, polystyrene microparticles were dispensed using the same method, in 6 cSt silicone oil. As shown in Fig. 4, a single particle, initially positioned within the thread while necking, resulted in encapsulated inside the primary satellite droplet. (Fig. 4. (D)), confirming its applicability of single cell encapsulation.

## IV. Conclusion

A new simple droplet generation method based on satellite formation is suggested and investigated. Most importantly, under given condition, stable generation of single or multiple droplets has been confirmed over a large number of dispensing. This method requires only a single drop of sample to dispense, a micropipette, and a hydrophilic plate to move. With this simple structure, it can be easily implemented into a lab-on-a-chip system without additional devices to dispense few droplets into oil phase.

Effects of the outer viscosity and the elongation rate were observed, and droplets were generated constantly with weak dependency on the elongation rate over several orders under given outer fluid. Larger droplets could be obtained using larger mother drop or less viscous external fluid. This can be a good alternative to a conventional continuous method to dispense a drop on-demand method with small amount of sample without preprocessing. This method has also a great potential for bio-medical applications including single cell encapsulation. By dispensing polystyrene microparticle suspension, we confirmed it's the potential use for single cell encapsulation without cell rupture.

## References


[1] M. Abdelgawad and A. R. Wheeler, "The Digital Revolution: A New Paradigm for Microfluidics," *Adv Mater*, vol. 21, no. 8, pp. 920-925, 2009.

[2] S.-Y. Teh, R. Lin, L.-H. Hung and A. P. Lee, "Droplet microfluidics," *Lab Chip*, vol. 8, no. 2, pp. 198, 2008.

[3] D. T. Chiu, R. M. Lorenz, and G. D. M. Jeffries, "Droplets for ultrasmall-volume analysis," *Analytical Chemistry*, vol. 81, no. 13, pp. 5111-5118, 2009.

[4] S. L. Anna, N. Bontoux, and H. A. Stone, "Formation of dispersions using "flow focusing" in microchannels," *Appl Phys Lett*, vol. 82, no. 3, pp. 364-366, 2003.

[5] S. N. Jayasinghe, A. N. Qureshi, and P. A. M. Eagles, "Electrohydrodynamic jet processing: An advanced electric-field-driven jetting phenomenon for processing living cells", *Small*, vol. 2, no. 2, pp. 216-219, 2006.

[6] E. Brouzes, M. Medkova, N. Savenelli, D. Marran, M. Twardowski, J. B. Hutchison, J. M. Rothberg, D. R. Link, N. Perrimon, and M. L. Samuels, "Droplet microfluidic technology for single-cell high-throughput screening", in *Proc. of the National Academy of Sciences of the United States of America*, vol. 106, no. 34, pp. 14195-14200, 2009.

[7] N. R. Beer, B. J. Hindson, E. K. Wheeler, S. B. Hall, K. A. Rose, I. M. Kennedy and B. W. Colston, "On-chip, real-time, single-copy polymerase chain reaction in picoliter droplets", *Analytical Chemistry*, vol. 79, no. 22, pp. 8471-8475, 2007.

[8] M. Hameed and J. F. Morris, "Breakup of a Liquid Jet Containing Solid Particles: A Singularity Approach," *SIAM Journal on Applied Mathematics*, vol. 70, no. 3, pp. 885-900, 2009.

[9] M. He, J. S. Edgar, G. D. M. Jeffries, R. M. Lorenz, J. P. Shelby, and D. T. Chiu, "Selective encapsulation of single cells and subcellular organelles into picoliter- and femtoliter-volume droplets," *Analytical Chemistry*, vol. 77, no. 6, pp. 1539-1544, 2005.

[10] O. A. Basaran, "Small-scale free surface flows with breakup: Drop formation and emerging applications," *AIChE Journal*, vol. 48, no. 9, pp. 1842-1848, 2002.

[11] K. C. Chaudhary and T. Maxworthy, "Nonlinear capillary instability of a liquid jet - 3. Experiments on satellite drop formation and control," *J Fluid Mech*, vol. 96, no. pt 2, pp. 287-297, 1980.

[12] I. Cohen, M. P. Brenner, J. Eggers, and S. R. Nagel, "Two fluid drop snap-off problem: Experiments and theory," *Physical Review Letters*, vol. 83, no. 6, pp. 1147-1150, 1999.

[13] J. Eggers, "Universal pinching of 3D axisymmetric free-surface flow," *Physical Review Letters*, vol. 71, no. 21, pp. 3458-3464, 1993.