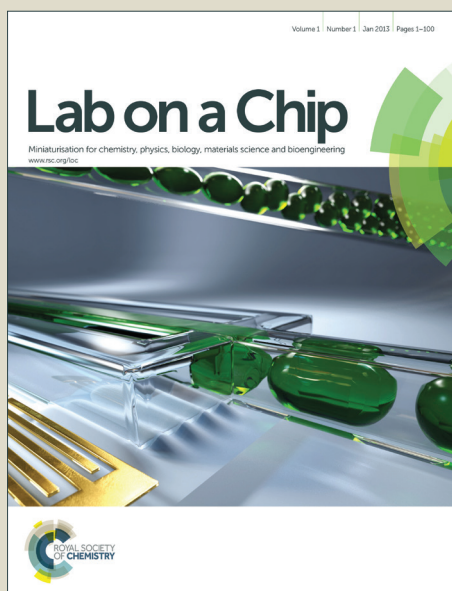


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## ARTICLE TYPE

## Three-dimensional Flash Flow Microreactor for Scale-up Production of Monodisperse PEG-PLGA Nanoparticles

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We present pressure tolerant 3D parallel polyimide (PI) film microreactor of up to ~160 bars with direct 3D flow focusing geometry for mass production of PEG-PLGA nanoparticles in a ~10<sup>1</sup> gram-scale (g/h).

## Introduction

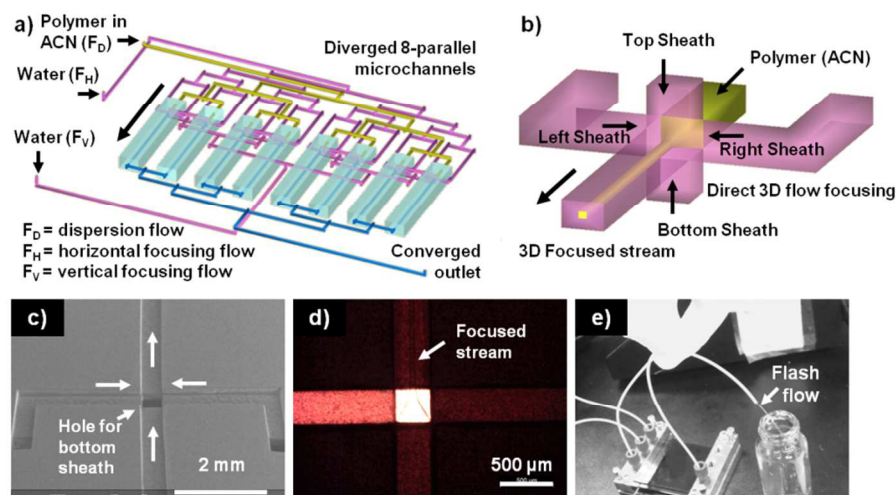
Biodegradable and biocompatible polymeric nanoparticles have received immense attention of researchers for applications like drug delivery, as physicochemical properties of polymer can be easily tailored for it to function as a drug carrier.<sup>[1,2]</sup> In particular, PEG-PLGA (Poly(ethylene glycol)-b-Poly(lactide-co-glycolide)) nanoparticles as drug carriers showed in vivo results for treatment of cancer with stealth function for immune evasion, and have successfully completed the phase 1 clinical trial.<sup>[3]</sup> Therefore it is a major issue to develop the reliable manufacturing process without batch-to-batch variability for high-throughput production. The PEG-PLGA nanoparticles have been routinely prepared by precipitation in bulk, which involves drop wise addition of polymer solution into large amount of water.<sup>[4]</sup> However, the procedure always results in poor control over the essential features of nanoparticles such as size and polydispersity (PDI), moreover additional difficulty arises in terms of low reproducibility even at small production scale. Recently, microfluidic approaches with short diffusive length in microscale level have provided better control over nanoparticle size and their PDI by simply manipulating the flow rate of the polymer solution and solvent (water) with resultant excellent reproducibility.<sup>[5-9]</sup> Despite the excellent performance of microfluidic approaches for producing nanoparticles, lack of industrial interest towards this field is evident due to extremely low production profile (e.g., ~10<sup>2</sup> mg/h). An ideal microfluidic system for mass production of nanoparticles is one in which high contents of polymer can be employed without any clogging and aggregation in the channels. Therefore, as previously reported, high influx of concentrated polymer at flash flow rate is exceedingly desirable for scale up and minimizing re-dissolution of the formed nanoparticles, which is mainly due to reduced retention in organic solvents.<sup>[7]</sup> Recently, Langer's group reported a controllable microvortex platform for the synthesis of lipid-polymer hybrid nanoparticles with improved productivity up to 0.3 g/h.<sup>[7]</sup> However, the study employed highly diluted polymer (5 mg/ml) to avoid aggregation problem. Karnik's group demonstrated parallelization of nanoparticle synthesis by 3D hydrodynamic flow focusing using

a PDMS microfluidic system with a limited production rate of only up to 84 mg/h.<sup>[9]</sup> Besides, the multilayer PDMS microfluidic system required sophisticated fabrication step; moreover, low elastic modulus of PDMS could not endure high flow rate for long-term use. Consequently, high-throughput nanoparticle production in gram-scale per hour is still a challenging task and it is proposed that novel microfluidic platform harboring continuous and flash flow approaches will be highly feasible to facilitate industrial applications of versatile drug carrier candidates.

Here in, we present for the first time the production of PEG-PLGA nanoparticles in a ~10<sup>1</sup> gram-scale (g/h) using novel and durable polyimide (PI) film microreactor with direct 3D flow focusing geometry that enhanced the productivity by performing at such a flash flow (11 ms of retention time in a unit microchannel). The 3D flash flow microreactor (3D-FFM) system fabricated by stacking seven layers of the resistant PI polymer film and subsequent one-step adhesive bonding process consisted of 8 sets of microreaction units in parallel with the diverged inlets and single converged outlet. In addition, the hydrophobic and non-sticky fluoropolymer coated PI polymer channel and direct 3D hydrodynamic flow focusing geometry led to zero adsorption on the channel surface and no particle aggregation, thus rendering it suitable for long-term use even under challenging conditions of high polymer influx at flash flow rate of concentrated polymer, which is promising for industrial mass production.

## Results and Discussion

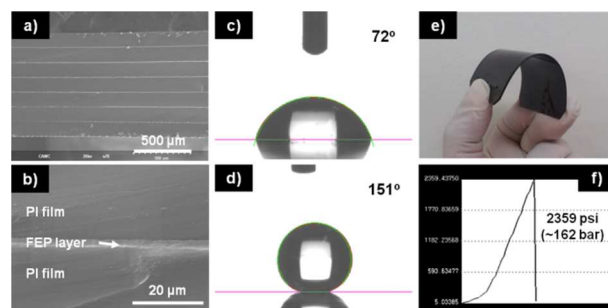
We recently reported a double-layered PI film microreactor which had excellent chemical resistance for organic syntheses under harsh conditions.<sup>[10]</sup> Unfortunately, the double-layered PI film stacking method is not compatible to construct 3D multilayer system because the bonding between patterned layers by inserting extra adhesive polyimide films caused difficulties in alignment and reproducible fidelity. In this work, we aimed to construct a novel and pressure tolerable 3D-FFM device for scale-up production of polymer nanoparticles through inexpensive route, which could sustain high flow rate of polymer for producing ~10<sup>1</sup> gram-scale of PEG-PLGA nanoparticles per hour. The parallel 8 sets of microchannels were designed to attain exact symmetrical structure, which ultimately allowed accomplishment of identical conditions in each microchannel for homogeneous and high-throughput production (Fig. 1).



**Fig. 1.** a) Illustration of 3D flash flow microreactor (3D-FFM) system for mass production of PEG-PLGA nanoparticles in a parallel manner. b) Schematic magnified view of direct 3D flow focusing. The polymer in acetonitrile ( $F_D$ ) infuses into yellow colored microchannel as a dispersion phase while two water flows ( $F_V$  and  $F_H$ ) introduce into purple colored microchannels for direct 3D flow focusing, respectively. All input flow diverge to 8 sets of unit microchannels (aquamarine) with symmetric dimension and then dispersion phases are focused to center of microchannel by 4 directional sheath flow of water phase, finally all streams come out to converge into an outlet (blue). c) Top-view SEM image of 3D flow focusing unit (unsealed microchannel). Optical images for (d) 3D flow focused stream in a semi-transparent and sealed microchannel and (e) formation of PEG-PLGA nanoparticles at flash flow condition ( $0.9 \times 8 = 7.2$  mL/min of polymer solution ( $F_D$ ), and  $2.1 \times 8 = 16.8$  mL/min of water ( $F_{H+V}$ )).

In proposed 3D-FFM system, every input flow ( $F_D$ ,  $F_H$  and  $F_V$ ) diverged into 8 sets of unit microchannels and subsequently polymer phase was focused into center of unit microchannel by 4 directional sheath flow of water phase, and finally all streams moved out to converge into an outlet. The PI film based 3D-FFM system was fabricated by UV laser ablation technique as previously reported by our group (for detailed fabrication method see Supporting Information).<sup>[10]</sup> Each layer of PI film (95 mm x 70 mm) was ablated by laser to form desired pattern along the edge of microchannel (300  $\mu$ m width and 125  $\mu$ m depth) and holes (1 mm diameter circle for in/outlet, 300  $\mu$ m square shape for junction between microchannels) as shown in Fig. S1, and the end of microchannel overlapped with other microchannel or square hole (300  $\mu$ m x 300  $\mu$ m) whilst assembling all films through simple alignment. Finally, the dimensions of main microchannel were 300  $\mu$ m of width, 125  $\mu$ m of height and 1.5 cm of length, respectively. The multilayer stacking of seven PI films was precisely aligned by inserting 4 metal pins of 1 mm in diameter into 4 corner holes of each PI film layer. For fabrication of pressure resistant 3D microfluidic device with durable bonding strength, 3  $\mu$ m thick layer of hydrophobic thermal-adhesive fluoroethylene-propylene (FEP, ND-110, Neoflon, DAIKIN, Japan, 60 % solids fluorothermoplastic aqueous dispersion) with a softening point at 260–280  $^{\circ}$ C was coated on each PI layer. The nano-powdery FEP layer reached molten state upon annealing at 300  $^{\circ}$ C and was evenly distributed between the PI films under pressure for bonding and tight sealing of the microchannels with no distortion upon solidification on cooling (Fig. 2). It is well known that hydrophobic FEP polymer acts as a thermal adhesive with chemical inertness.<sup>[11]</sup> The contact angle of FEP coated PI film was 151 $^{\circ}$  while original PI film was 72 $^{\circ}$  (Fig. 2c, d). This superhydrophobic FEP coating as a family of Teflon renders antifouling effects at the surface to result in no channel clogging. It is desirable to emphasize at this point that self-adhering FEP in

the multilayers facilitated the multiple layered bonding, rather than the inserting extra adhesive PI films between patterned layers as reported,<sup>[10]</sup> and readily sealed into complex shape of microchannel with high reproducibility; moreover, the thin FEP adhesive layer retained flexibility of whole film device that enabled bending with no mechanical crack. Furthermore, the 3D-FFM system endured the conditions of continuous flow rate of 30 mL/min for 3 hours with no delamination; however, the channel ruptured under the applied pressure of  $\sim$ 160 bars (2359 Psi).<sup>[12]</sup>


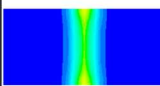
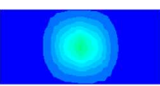
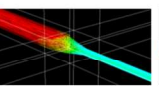
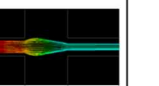
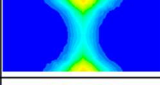
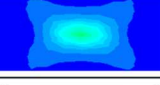
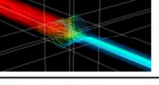
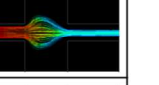
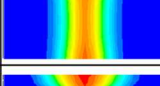
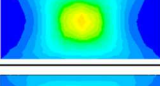
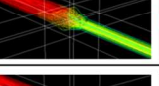
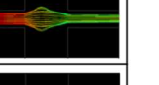
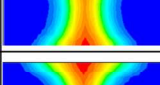
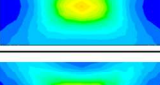
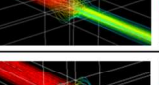
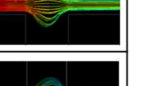
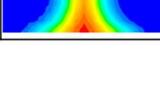
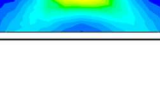
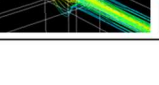



**Fig. 2.** Cross-sectional SEM images of (a) seven-layered PI films with FEP layer after one step bonding and (b) magnified view. The thin FEP layer is well distributed and tightly sealed between PI films. Contact angle of polyimide film (c) before and (d) after FEP coating. (e) Optical image showing flexible property of fabricated 3D-FFM system by one-step bonding process using thin FEP adhesive layer. (f) Recorded curve of burst pressure while rapidly pumping water at 10 mL/min using HPLC through 3D-FFM PI microfluidic device with closed outlet.

It is noteworthy that semi-transparent thin 3D-FFM system as a novel platform for high-throughput production of PEG-PLGA nanoparticles was fabricated by rapid one-step bonding process of brown-colored PI film layers with a simple metal pin alignment, rather than by following conventional and laborious repeated layer-up process. Furthermore, the striking pressure tolerance allowed application of continuous flash flow rates to 8 sets of

compactly arranged microreactor.

**Table 1.** Summarized experimental conditions and results of simulation for both 2D and 3D hydrodynamic flow focusing pattern.

Ratio	Total Flow rate (mL/min)	Re	Flow Condition (μl/min) F <sub>D</sub> -dispersion flow F <sub>H</sub> -horizontal / F <sub>V</sub> -vertical flow	Expected Flow pattern 0  1		Pathline profile (3D)	
				2D flow focusing	3D flow focusing	3D -view	Side-view
1 : 9	0.08	1.33	F <sub>H</sub> : F <sub>V</sub> = 7 : 3 F <sub>D</sub> = 8 / F <sub>H</sub> = 50.4 / F <sub>V</sub> = 21.6 Unit microchannel : F <sub>D</sub> = 1 / F <sub>H+V</sub> = 9				
	8	133	F <sub>H</sub> : F <sub>V</sub> = 4 : 6 F <sub>D</sub> = 800 / F <sub>H</sub> = 2880 / F <sub>V</sub> = 4320 Unit microchannel : F <sub>D</sub> = 100 / F <sub>H+V</sub> = 900				
3 : 7	0.08	1.33	F <sub>H</sub> : F <sub>V</sub> = 7 : 3 F <sub>D</sub> = 24 / F <sub>H</sub> = 39.2 / F <sub>V</sub> = 16.8 Unit microchannel : F <sub>D</sub> = 3 / F <sub>H+V</sub> = 7				
	8	133	F <sub>H</sub> : F <sub>V</sub> = 5 : 5 F <sub>D</sub> = 2400 / F <sub>H</sub> = 2800 / F <sub>V</sub> = 2800 Unit microchannel : F <sub>D</sub> = 300 / F <sub>H+V</sub> = 700				
	24	400	F <sub>H</sub> : F <sub>V</sub> = 5 : 5 F <sub>D</sub> = 7200 / F <sub>H</sub> = 8400 / F <sub>V</sub> = 8400 Unit microchannel : F <sub>D</sub> = 900 / F <sub>H+V</sub> = 2100				

Prior to actual experiment on scale-up production of PEG-PLGA nanoparticles, simulation using computational fluid dynamics (CFD) was carried out to elucidate direct 3D hydrodynamic flow focusing pattern under conditions of continuous and flash flow rates; the study proved to be useful for visualization of flow focusing behavior in translucent microchannels. The flow pattern was simulated at 0.01 ~ 3 mL/min of total flow rates in a unit microchannel, which corresponds to Reynolds number 1.33 ~ 400, with two different flow ratios of polymer solution to water as 1:9 (0.11 of polymer contents) and 3:7 (0.43 of polymer contents), respectively. (Table 1 and see Supporting Information Table S1).

Initially, every flow ratio between vertical and horizontal flow was unable to create homogeneous isolation of inner flow into the center of microchannel because of asymmetric dimension of microchannel (300 μm width and 125 μm depth) and parabolic velocity distribution within the microchannel in which middle of stream was much faster than flow near the wall. Thus, we optimized flow condition by controlling the ratio of vertical and horizontal flow rate at given flow rate as shown in Table S1.

In general, direct 3D hydrodynamic flow focusing was very effective to isolate dispersion phase into inner core flow from the channel walls regardless of flow rate even at high polymer influx flow ratio (3 : 7). Moreover, even an increased flow rate of upto Re = 400 enhanced convective mixing where time scale for mixing is 300 times faster than Re = 1.33. It is hypothesized that short mixing time in such a flash flow (Re = 133 and 400) might dominate particle creation to produce smallest sizes as possible because of mixing time being much faster than aggregation time as compared to slow diffusive mixing.<sup>[6a, 7]</sup> In contrast, 2D flow focusing revealed the existence of initial concentration of the dispersion phase near the walls without undergoing mixing under high flow rate. These simulations revealed high desirability of direct 3D hydrodynamic flow focusing under flash flow condition for reproducible mass production of polymer nanoparticles

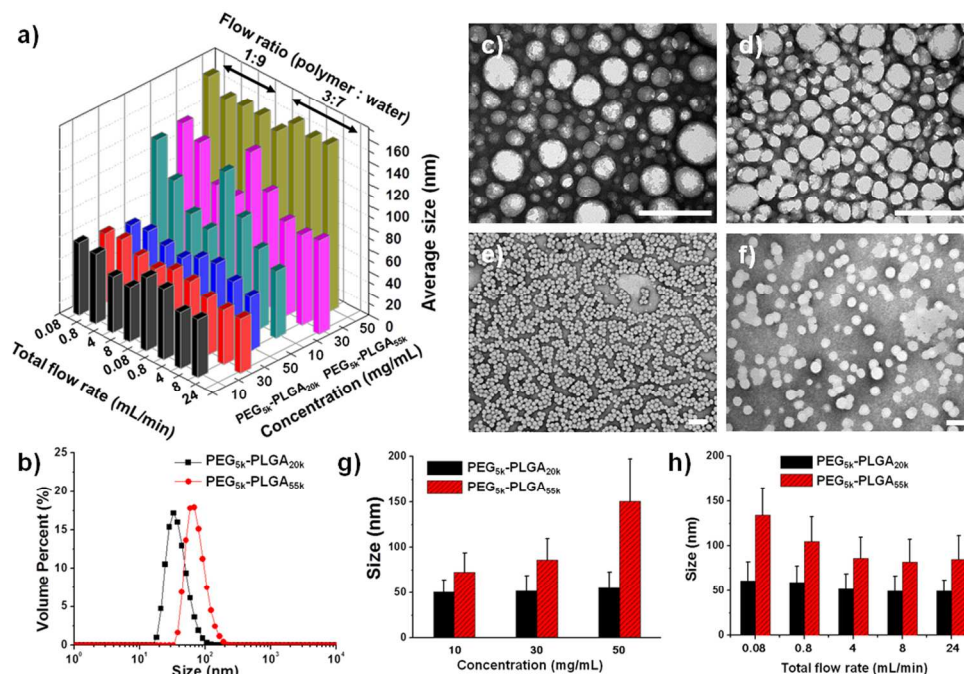
without aggregation.

Inspired by preliminary simulation result, we focused our attention towards production of polymer nanoparticles to prove simulation hypothesis of our parallel 3D-FFM system as a function of flow rate, flow ratio, concentration and molecular weight of PEG-PLGA polymer at conditions as shown in Table S1 (for detailed experimental procedure see Supporting Information). For self-assembly of nanoparticles by solvent exchange, PEG-PLGA polymer in acetonitrile (ACN) were infused and focused both vertically and horizontally by four water sheath flows under desired flow rates. Dynamic light scattering (DLS) and transmission electron microscopy (TEM) were employed for characterization of nanoparticles. Fig. 3a shows the size of PEG-PLGA nanoparticles produced from 3D-FFM system at various conditions; two different molecular weight (PEG<sub>5k</sub>-PLGA<sub>20k</sub> and PEG<sub>5k</sub>-PLGA<sub>55k</sub>), and polymer concentrations in ACN (10, 30 and 50 mg/mL) under 0.08 ~ 24 mL/min range of total flow rates, which corresponds to Re 1.33 ~ 400 in a unit microchannel, with different flow ratio (1:9 and 3:7). By simply varying the above parameters, the size of produced nanoparticles can be reproducibly controlled in a range of 50 nm to 150 nm, which could be an ideal range for stable and biologically relevant nanoparticles.<sup>[2]</sup> The smaller size is feasible with lower PLGA molecular weight.<sup>[6c, 9, 13]</sup> We observed that the results of size distribution for all experimental conditions in 3D flow focusing were quite narrow without exhibiting any aggregation except in two cases where 50 mg/mL of PEG<sub>5k</sub>-PLGA<sub>55k</sub> was infused into device at a flow ratio of 3:7 and low flow rate (for all DLS results, see Supporting Information). Polymer with high molecular weight or high concentration frequently exhibit phenomenon of aggregation. Because diffusive mixing at low Reynolds number is not efficient enough to fully manipulate high contents of polymer, it is possible that non-precipitated polymer could aggregate into larger particles in a range ~10<sup>4</sup> nm (see Supporting Information DLS result of 50



mg/mL PEG<sub>5k</sub>-PLGA<sub>55k</sub>). In contrast, such a flash flow induced fast convective mixing could avoid generation of larger particles because of rapid mixing time than the aggregation time as compared to slow diffusive mixing even in the presence of high contents of PEG<sub>5k</sub>-PLGA<sub>55k</sub>.<sup>[7, 14]</sup> Fig. 3b shows highly monodisperse size distribution of PEG-PLGA nanoparticles obtained at fastest condition in 24 mL/min of total flow rate (Re =

400) from 30 mg/mL of polymer; and the average fluid velocity and residence time in the flow focusing main unit microchannel for precipitation were approximately 1.33 m/s and 11 ms, respectively. The achievement of 3D flow focusing consistently produced smaller nanoparticles in 50 nm and 85 nm size from PEG<sub>5k</sub>-PLGA<sub>20k</sub> and PEG<sub>5k</sub>-PLGA<sub>55k</sub>, respectively (Fig. 3e, f).



**Fig. 3.** a) 3D map of nanoparticle size produced from 3D-FFM system as a function of flow rate, flow ratio, molecular weight and concentration of polymer. b) The size distribution by volume fraction of nanoparticles prepared using 30 mg/mL of PEG<sub>5k</sub>-PLGA<sub>20k</sub> and PEG<sub>5k</sub>-PLGA<sub>55k</sub> at Re = 400 (0.9 x 8 = 7.2 mL/min of polymer solution (F<sub>D</sub>), 2.1 x 8 = 16.8 mL/min of water (F<sub>H+V</sub>)). TEM images of nanoparticles prepared from c) bulk method, d) 2D flow focusing and e, f) 3D-FFM. The scale bars are 200 nm. For bulk method (c), 0.9 mL of 30 mg/mL of PEG<sub>5k</sub>-PLGA<sub>20k</sub> was added to 2.1 mL of water phase by dropwise addition. For 2D flow focusing (d), 30 mg/mL of PEG<sub>5k</sub>-PLGA<sub>20k</sub> and water were infused into 2D flow focusing device at 0.9 mL/min and 2.1 mL/min, respectively. TEM images of (e) and (f) were obtained from PEG<sub>5k</sub>-PLGA<sub>20k</sub> and PEG<sub>5k</sub>-PLGA<sub>55k</sub> at same conditions with (b), and the average sizes are 50 nm and 85 nm, respectively. g) Effect of PEG-PLGA polymer concentration on the produced nanoparticle size at Re = 66 (0.15 x 8 = 1.2 mL/min of polymer (F<sub>D</sub>), 0.35 x 8 = 2.8 mL/min of water (F<sub>H+V</sub>)). h) Effect of flow rate on the produced nanoparticle size prepared from 30 mg/mL of PEG-PLGA (Re = 1.3 ~ 400).

Under these conditions, the uniformly produced PEG-PLGA nanoparticles were surprisingly spewed out from outlet tube as can be seen in Fig. 1e (see Supporting Movie). In contrast, the 2D flow focusing and bulk method produced highly polydisperse nanoparticles with larger size under similar conditions (Fig. 3c, d, see Supporting Information), which are not efficient for complete mixing of high contents of polymer within short time (~ ms range) and the remained polymer is prone to aggregation of particles (Table 1).<sup>[14]</sup> In the microfluidics such as 2D flow focusing, low quantity of polymer influx (e.g. 0.1 ratio of polymer to water) has been widely used to avoid aggregation and channel clogging, rather than high quantity of polymer influx.<sup>[6-9]</sup> At here, it is worthy to note that the developed 3D-FFM system enables not only high throughput production of polymer nanoparticles, but also uniform control of the physicochemical quality even under high influx of polymer.

As shown in Fig. 3g, the average size of nanoparticles produced at Re = 66 was increased from 50 nm to 55 nm in the case of PEG<sub>5k</sub>-PLGA<sub>20k</sub> and from 72 nm to 150 nm in the case of PEG<sub>5k</sub>-PLGA<sub>55k</sub> with an increase in polymer concentration, respectively.

This observation indicates concentration dependant variance in size of nanoparticles from PEG<sub>5k</sub>-PLGA<sub>55k</sub> when compared to PEG<sub>5k</sub>-PLGA<sub>20k</sub>. It is generally known that the length of polymer block plays an important role in the critical association concentration (CAC) to form micelle-like polymer aggregates.<sup>[6c, 13]</sup> And, the larger hydrophobic PLGA block lowers CAC to produce larger nanoparticles. In the present study, the increase in flow rates showed gradual decrease in the size of nanoparticles at the given conditions because of mixing time being faster than the aggregation time under condition of increased flow rate (Fig. 3h and see Supporting Information Fig. S4). In general, larger particles can reflect more light, the solution containing larger particles obtained from high polymer ratio looks milky with low transparency (see Supporting Information Fig. S5). Interestingly, sizes of nanoparticles produced at Re = 400 were 50 nm for PEG<sub>5k</sub>-PLGA<sub>20k</sub> and 85 nm for PEG<sub>5k</sub>-PLGA<sub>55k</sub>, because flash flow induced fast convective mixing results kinetically locked nanoparticles that are smallest sizes as possible (Fig. 3e, f).<sup>[6a, 7, 14]</sup> It is commendable to state that 3D-FFM system can produce homogeneous polymer nanoparticles of smallest size on a flash

production at a given polymer composition.

**Table 2.** Comparison of production rate of our 3D Flash Flow Microreactor against the reported literature works at various experimental conditions.

	Ref. 9	Ref. 8	Ref. 7	3D-FFM
$F_D^{[a]}$	28 $\mu\text{L}/\text{min}$	2 $\text{mL}/\text{h}$	1 $\text{mL}/\text{min}$	7.2 $\text{mL}/\text{min}$
$F_C^{[b]}$	400 $\mu\text{L}/\text{min}$	10 $\text{mL}/\text{h}$	10 $\text{mL}/\text{min}$	16.8 $\text{mL}/\text{min}$
Concentration <sup>[c]</sup>	50 $\text{mg}/\text{mL}$	50 $\text{mg}/\text{mL}$	5 $\text{mg}/\text{mL}$	30 $\text{mg}/\text{mL}$
Ratio ( $F_D/F_C$ ) <sup>[d]</sup>	0.1	0.2	0.1	0.43
Production rate	0.084 $\text{g}/\text{h}$	0.1 $\text{g}/\text{h}$	0.33 $\text{g}/\text{h}$	12.9 $\text{g}/\text{h}$

<sup>a</sup> Flow rate of polymer solution; <sup>b</sup> Flow rate of water; <sup>c</sup> Concentration of polymer; <sup>d</sup> Flow ratio of polymer solution to water.

Moreover, ideal mass production of nanoparticles in microfluidic system is required to operate device under flash flow using high contents of polymer. In comparison of production rate in the previously reported works, our 3D-FFM system demonstrates highest production scale as shown in Table 2.<sup>[7-9]</sup> At this point it can be remarkably stated that 3D-FFM system could produce uniform nanoparticles without channel clogging and aggregation and with 12.9 g/h of production rate at  $Re = 400$  using 30 mg/mL of polymer. It means that only 1 min of processing time could produce amount equivalent to 216 mg nanoparticles that is sufficient for ca. 8 sets of *in vivo* mice experiments under estimated 25 mg per 5 mice in a set. Practically, the developed 3D-FFM system even at short processing time is useful for high throughput and cost effective continuous production (300g/day).

### Conclusions

In summary, we present pressure tolerant 3D parallel microfluidic system of up to ~160 bars, and termed as a 3D flash flow microreactor (3D-FFM) system, for mass production of monodisperse PEG-PLGA polymeric nanoparticles. The multilayered PI film microreactor fabricated by simple one-step multilayer bonding process consists of not only 8 sets of microchannels that could work under high flow rate (e.g., 30 mL/min) to enhance productivity but also 3D hydrodynamic flow focusing to avoid aggregation in case of long period usage. The 3D-FFM system enables reproducible fabrication of polymer nanoparticles without aggregation under fast flow rate with higher ratio of polymer to solvent (~0.43), yielding 12.9 g/h production rate of monodisperse PEG-PLGA nanoparticles with average diameter of 50 nm and 85 nm, respectively. To the best of our knowledge this is the highest production rate of nanoparticles and highest polymer ratio under microreactor system reported until date. It is plausible that this system with the enhanced productivity contributes to bridge the gap between academic research and industrial mass production in the area of pharmaceuticals and drug delivery.

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### Notes and references

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- a) R. Gref, Y. Minamitake, M. T. Peracchia, V. Trubetskoy, V. Torchilin, R. Langer, *Science* 1994, **263**, 1600; b) F. Gu, L. Zhang, B. A. Teply, N. Mann, A. Wang, A. F. Radovic-Moreno, R. Langer, O. C. Farokhzad, *Proc. Natl. Acad. Sci. U S A* 2008, **105**, 2586; c) L. F. Zhang, J. M. Chan, F. X. Gu, J. W. Rhee, A. Z. Wang, A. F. Radovic-Moreno, F. Alexis, R. Langer, O. C. Farokhzad, *ACS Nano* 2008, **2**, 1696.
  - a) K. S. Soppimath, T. M. Aminabhavi, A. R. Kulkarni, W. E. Rudzinski, *J Control Release* 2001, **70**, 1; b) D. N. Nguyen, J. J. Green, J. M. Chan, R. Longer, D. G. Anderson, *Adv Mater* 2009, **21**, 847; c) N. Kamaly, Z. Y. Xiao, P. M. Valencia, A. F. Radovic-Moreno, O. C. Farokhzad, *Chem Soc Rev* 2012, **41**, 2971; d) J. Nicolas, S. Mura, D. Brambilla, N. Mackiewicz, P. Couvreur, *Chem Soc Rev* 2013, **42**, 1147.
  - J. Hrkach, D. Von Hoff, M. M. Ali, E. Andrianova, J. Auer, T. Campbell, D. De Witt, M. Figa, M. Figueiredo, A. Horhota, S. Low, K. McDonnell, E. Peeke, B. Retnarajan, A. Sabnis, E. Schnipper, J. J. Song, Y. H. Song, J. Summa, D. Tompsett, G. Troiano, T. V. Hoven, J. Wright, P. LoRusso, P. W. Kantoff, N. H. Bander, C. Sweeney, O. C. Farokhzad, R. Langer, S. Zale, *Sci Transl Med* 2012, **4**, 128ra39.
  - S. Schubert, J. T. Delaney, U. S. Schubert, *Soft Matter* 2011, **7**, 1581.
  - a) S. Marre, K. F. Jensen, *Chem Soc Rev* 2010, **39**, 1183; b) C. A. Serra, B. Cortese, I. U. Khan, N. Anton, M. H. J. M. de Croon, V. Hessel, T. Ono, T. Vandamme, *Macromol React Eng* 2013, **7**, 414.
  - a) R. Karnik, F. Gu, P. Basto, C. Cannizzaro, L. Dean, W. Kyei-Manu, R. Langer, O. C. Farokhzad, *Nano Lett* 2008, **8**, 2906; b) N. Kolishetti, S. Dhar, P. M. Valencia, L. Q. Lin, R. Karnik, S. J. Lippard, R. Langer, O. C. Farokhzad, *Proc Natl Acad Sci U S A* 2010, **107**, 17939; c) M. Rhee, P. M. Valencia, M. I. Rodriguez, R. Langer, O. C. Farokhzad, R. Karnik, *Adv Mater* 2011, **23**, H79.
  - a) Y. T. Kim, B. L. Chung, M. M. Ma, W. J. M. Mulder, Z. A. Fayad, O. C. Farokhzad, R. Langer, *Nano Lett* 2012, **12**, 3587; b) Y. Kim, B. L. Chung, M. M. Ma, W. J. M. Mulder, Z. A. Fayad, O. C. Farokhzad, R. Langer, *Nano Lett* 2013, **13**, 4997; c) The expected production rate (P) is determined by polymer concentration (C) and flow rate of polymer solution (F). The equation is  $P (\text{mg}/\text{min}) = C (\text{mg}/\text{mL}) \times F (\text{mL}/\text{min})$ ; Thus their productivity of PLGA nanoparticles is 0.3 g/h by following equation;  $(5 \text{ mg}/\text{mL} \text{ of PLGA}) \times (1 \text{ mL}/\text{min} \text{ of flow rate})$ . Here, lipid is negligible due to its relatively small amount;  $(0.075 \text{ mg}/\text{mL} \text{ of lipid}) \times (10 \text{ mL}/\text{min} \text{ of flow rate}) = 0.045 \text{ g}/\text{h}$ .
  - X. J. Kang, Q. X. Luo, Q. Wei, C. Y. Xiong, Q. Chen, Y. Chen, Q. Ouyang, *Microfluid Nanofluid* 2013, **15**, 337.
  - J. M. Lim, N. Bertrand, P. M. Valencia, M. Rhee, R. Langer, S. Jon, O. C. Farokhzad, R. Karnik, *Nanomed-Nanotechnol Biol Med* 2014, **10**, 401.
  - a) K. I. Min, T. H. Lee, C. P. Park, Z. Y. Wu, H. H. Girault, I. Ryu, T. Fukuyama, Y. Mukai, D. P. Kim, *Angew Chem Int Ed* 2010, **49**, 7063; b) S. Sharma, R. A. Maurya, K. I. Min, G. Y. Jeong, D. P. Kim, *Angew Chem Int Ed* 2013, **52**, 7564.
  - a) W. H. Grover, M. G. von Muhlen, S. R. Manalis, *Lab Chip* 2008, **8**, 913; b) K. N. Ren, W. Dai, J. H. Zhou, J. Su, H. K. Wu, *Proc Natl Acad Sci U S A* 2011, **108**, 8162.
  - a) J. B. You, K. I. Min, B. Lee, D. P. Kim, S. G. Im, *Lab Chip* 2013, **13**, 1266; b) V. Sunkara, D. K. Park, H. Hwang, R. Chantivas, S. A. Soper, Y. K. Cho, *Lab Chip* 2011, **11**, 962; c) D. A. Mair, E. Geiger, A. P. Pisano, J. M. J. Frechet, F. Svec, *Lab Chip* 2006, **6**, 1346.
  - Y. S. Nam, H. S. Kang, J. Y. Park, T. G. Park, S. H. Han, I. S. Chang, *Biomaterials* 2003, **24**, 2053.
  - B. K. Johnson, R. K. Prud'homme, *Phys Rev Lett* 2003, **91**, 118302.