

effectively separated. In this exemplary system, close to 100% separation of 10- $\mu\text{m}$  beads is provided, as shown in FIG. 35F, at a flow rate of 3.5 ml/min.

#### Example 19

[0246] FIG. 36 illustrates utilizing inertial focusing for particle separation. An input solution of 0.1% w/v of mean diameter 3.87  $\mu\text{m}$  (4) and 7.32  $\mu\text{m}$  (7), uniformly distributed, was introduced into a single asymmetric device 100 (narrow)-160 (wide)  $\mu\text{m}$  in width, 50  $\mu\text{m}$  tall, and 3 cm in length. At the outlet the channel was split into 5 exit channels with equivalent resistance and fractions were collected for a flow at  $Re \sim 8$ . Flow of a total of 1 mL of solution over 10 minutes allowed ample sample for analysis by coulter counter. Histograms of particle sizes are shown for each of these fractions (numbers 1-5 and indicated in the image). The volumetric ratio between 4 (3-5  $\mu\text{m}$ ) and 7 (6-8.5  $\mu\text{m}$ ) micrometer particles is shown above each histogram. Notably in fraction 2 the larger particles are enhanced two-fold while in fraction 5 the larger particles are depleted  $\sim 200$  times.

#### Example 20

[0247] FIG. 37 illustrates focusing behavior in symmetric curving channels. As  $Re$  number increases from 0.5 to 5 a transition to two focused streams is observed. As  $Re$  is increased further stable but more complex behavior is observed. Scale bar is 50  $\mu\text{m}$ .

#### Example 21

[0248] FIG. 38 illustrates the dependence of particle focusing on  $a/D_h$ . Streak images at the outlet are shown 3 cm downstream of the inlet for a flow at  $Re=100$ . The image is shown at the recombination of two branches to illustrate the uniformity of the flow profile from channel to channel.

#### Example 22

[0249] FIG. 39 illustrates focusing behavior for channels of 35  $\mu\text{m}$  to 65  $\mu\text{m}$  width. The average radius of curvature for the small dimension is 32.5  $\mu\text{m}$ . Focusing to a single stream is observed  $\sim Re$  of 5 while focusing to two streams is observed at higher  $Re$  and at lower  $Re$  number. The particle diameter is 10  $\mu\text{m}$ . Scale bar is 50  $\mu\text{m}$ .

#### Example 23

[0250] FIG. 40 illustrates focusing behavior for channels of 50  $\mu\text{m}$  to 80  $\mu\text{m}$  width. The average radius of curvature for the small dimension is 40  $\mu\text{m}$ . Focusing to a single stream is observed  $\sim Re$  of 2.5 while focusing to two streams is observed at higher  $Re$ . Above  $Re=25$  more complex but stable behavior is observed. The particle diameter is 10  $\mu\text{m}$ . Scale bar is 50  $\mu\text{m}$ .

#### Example 24

[0251] FIG. 41 illustrates focusing behavior for channels of 100  $\mu\text{m}$  to 160  $\mu\text{m}$  width. The average radius of curvature for the small dimension is 80  $\mu\text{m}$ . Focusing to a single stream is observed  $\sim Re$  of 12 while more complex but stable behavior is observed for higher  $Re$ . The particle diameter is 10  $\mu\text{m}$ . Scale bar is 100  $\mu\text{m}$ .

#### Example 25

[0252] FIG. 42 illustrates focusing behavior for channels of 350  $\mu\text{m}$  to 650  $\mu\text{m}$  width. The average radius of curvature for the small dimension is 325  $\mu\text{m}$ . Focusing to a single stream is observed  $\sim Re$  of 90 while more complex but stable behavior is observed for higher  $Re$ . The particle diameter is 10  $\mu\text{m}$ . Scale bar is 100  $\mu\text{m}$ .

#### Example 26

[0253] FIG. 43 illustrates particle dependent focusing for separation. A uniform mixture 10  $\mu\text{m}$  and 2  $\mu\text{m}$  particles was input at the inlet and fluorescent streak images were observed at the outlet for (A) the green fluorescent 10  $\mu\text{m}$  particles and (B) the red fluorescent 2  $\mu\text{m}$  particles. The flow is at a  $Re$  of 5. There is a distinct separation across streamlines for the different size particles with no externally applied forces. The scale bar is 50  $\mu\text{m}$ .

#### Example 27

[0254] FIG. 44 illustrates focusing of blood cells in the same manner as rigid particles. Five percent whole blood diluted in PBS is run through rectangular channels of 50  $\mu\text{m}$  width. At the outlet, 3 cm downstream, streak images of cells are observed in phase contrast. These appear as dark streams in the gray channel. The channel edges are also dark. As in the case with rigid particles 3 streaks are observed which correspond to four focus points on the rectangular channel faces.

#### Example 28

[0255] FIGS. 45A and 45B illustrate focusing of cultured cell lines. As with particles, deformable cells are focused to single streams. FIG. 43A shows streak images of cells focusing for various  $Re$  numbers are shown. The inlet of each focusing area is shown on the left. Focusing to a single lane starts to occur for  $Re \sim 2$  after 3 cm of travel. In FIG. 43B, intensity cross sections at various turns and at the outlet are shown. Note that at the outlet the width of the focused stream is comparable to the diameter of a single cell ( $\sim 15 \mu\text{m}$ ).

[0256] Experimental Conditions and Apparatus

[0257] While many experimental conditions can be used to create and utilize the exemplary systems described herein, some conditions used to achieve the results discussed above are presented below.

[0258] Materials

[0259] Fluorescent polystyrene microparticles (density  $\sim 1.05 \text{ g/ml}$ ) were either purchased from Bangs Laboratories (Fishers, Ind.) or Duke Scientific (Fremont, Calif.). For 4 (3.87)  $\mu\text{m}$  and 7 (7.32)  $\mu\text{m}$  particles the Bangs Labs product codes were FS05F/7772 and FS06F/6316 respectively. For 2 (2.0)  $\mu\text{m}$ , 9  $\mu\text{m}$ , 10 (9.9)  $\mu\text{m}$  and 17  $\mu\text{m}$  the Duke Scientific product numbers were R0200, 36-3, G1000 and 35-4. Particles were mixed to desired weight fractions by dilution in Phosphate buffered saline (PBS) and stabilized by addition of 0.1% Tween 20. Particles were mixed to desired weight fractions by dilution in PBS and stabilized by addition of 0.1% Tween 20. In the various described experiments particle wt/vol % varied between 0.1% and 1%. Silicone oil droplets were formed from 10% wt/vol DC 200 (10 centistokes, Dow Corning) stabilized with 2% wt/vol polyethylene glycol monooleate (molecular weight 860, SigmaAldrich). The mixture was shaken vigorously and allowed to settle for 20 min. Solution was taken from the bottom 1 cm of the vial to ensure