Title: Rapid Microfluidic Mixing via Rotating Magnetic Microbeads

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Presented here is a novel method for mixing in a microfluidic channel via an array of rotating magnetic microbeads. The microbeads rotate around circular permalloy (NiFe) features magnetized by a rotating external magnetic field. This system demonstrates rapid mixing in short channel lengths. The effectiveness of this system is quantified by analyzing the degree of mixing of two streams, one with fluorescence and one without. Our experiments and numerical simulations reveal that beads orbiting at a high velocity as compared to flow down the channel cause the fluid to form circular coronae that stretch across the microchannel leading to fluid mixing. Under these conditions, rotating magnetic microbeads were able to fully mix the streams in a 270 μm mixing region.

Keywords

Microfluidic mixing, continuous-flow, magnetic microbeads, lab-on-a-chip

1 Introduction

Microfluidics and lab-on-a-chip techniques have made significant impacts in fields from medical diagnostics to chemical processing to reduce reagent cost, decrease operation time and increase portability [1-3]. However, mixing is much more difficult in the low Reynolds number flow conditions experienced in microfluidic systems than in a typical larger-scale chemical laboratory setting, since in these flows inertial effects are essentially nonexistent. Thus, in order to avoid the need to rely on molecular diffusion for microfluidic mixing, a variety of passive and active mixers have been proposed.

Passive mixers use geometrical features to enhance mixing by stretching and folding fluid interfaces to decrease the distance over which diffusion acts. However, this stretching and folding can require extensive channel lengths. Stroock et al. demonstrated a device that utilized staggered herringbone-shaped ridges patterned on the floor of a microfluidic channel to create chaotic flow patterns that fold fluid into thin laminae and thus effectively mixed fluid streams [4]. However, such a device required a length of 500 mm or more to achieve uniform mixing [4]. Various other systems make use of bending the channel either in two or three dimensions, but these systems still require footprints of a few millimeters to achieve substantial mixing [5, 6]. Other systems make use of side channels that remove a small portion of the fluid and reintroduce it downstream for redistribution back into the main channel [7]. All of these passive mixers rely upon fluid flow through the channel to generate mixing, and thus mixing degree and flow rate cannot be controlled independently in such devices.

Active mixers expend energy into the system to perturb the fluid and to accelerate mixing. Lin et al. demonstrated mixing in a 1 mm-long channel by utilizing a switching pumping system operating at 1Hz between two separate reservoirs in a T-shaped microfluidic channel [8]. While technically an active mixer, this system is unable to actuate mixing independently from flow rate. Additionally, the stop and start nature of the flows makes it difficult to integrate into a continuous-flow system. A similar concept was presented by Niu and Lee using side channels to alternate pumping into a single main channel also containing two fluids [9]. This system required at least 3 mm of channel length to achieve strong mixing of small proteins. Campisi et al. demonstrated an electrokinetic micromixer utilizing AC osmosis and dielectrophoretic (DEP) force to induce chaotic flows in the a microfluidic channel [10]. While this system does allow for independent control of flow rate and mixing, the use of chaotic flows requires 5 mm of channel length to achieve mixing. Further, the use of DEP precludes the use of many common buffers used in biological studies, due to their high salt concentration.

Several studies have investigated the use of sonic and ultrasonic transducers which can be actuated externally to the microfluidic channel. Mixing has also been achieved in very short channel lengths using ultrasonic actuation. However, this method increased the fluid temperature by 15 °C, and is thus incompatible with many biological applications where heating can be deleterious to sample

integrity [11-13]. More recent work has demonstrated mixing induced by LED illumination which overcomes heating problems but still requires a few millimeters to achieve even 50% mixing [14]. To summarize, microfluidic mixing falls into two groups: passive mixing and active mixing. Passive mixing folds the fluid over long channel lengths, and in such devices, mixing cannot be actuated independent of fluid flow. Active mixing creates effective mixing in shorter channel lengths, but often has difficulty integrating with continuous-flow systems or is incompatible with biological applications and requires more complex fluid systems.

An alternative approach relies on the use of magnetic beads to perform microfluidic mixing. Grumann et al. described a batch mixing system that utilizes large magnetic beads whose trajectories on a rotating disk are deflected by permanent magnets, such that they traverse through the mixing chamber and provide for mixing [15]. However, this system does not lend itself to continuous-flow mixing applications. Lee et al. [16] used ferromagnetic microbeads that were driven by a rotating external magnetic field such that they rotated in columns or clumps of beads through the mixing section of a continuous-flow channel. They demonstrated good mixing over a length of 800 μm . However, the use of agglomerations of ferromagnetic microbeads, which continue to agglomerate even after removal of the external magnetic field, limits the ability to control the mixing and to extract beads for further analysis, as superparamagnetic beads should be used if the latter functionality is desired [17].

This paper demonstrates and quantifies the mixing effectiveness of a periodic array of rotating magnetic microbeads that actively perturb the fluid in a continuous-flow microfluidic system, such as we have previously proposed and investigated through numerical simulations [18-21]. We demonstrate that this mixing device, actuated simply by a rotating external magnetic field, can produce rapid and controllable mixing over a short distance in a microfluidic channel. This device operates by actuating microbeads (Dynabeads M-280, 2.8 μm diameter) in controlled orbits through a microfluidic channel. The rotation of these microbeads is induced by microfabricated paramagnetic features that are magnetized by a rotating external magnetic field. When in the presence of a permanent magnetic field, the magnetic features develop magnetic poles, where the external magnetic field lines are concentrated. The magnetic microbeads are superparamagnetic and become magnetized in the presence of the magnetic field, and are attracted to the poles of the magnetic features (Figure 1). When the external magnetic field is rotated, the locations of these poles in the magnetic features rotate along with the external magnetic field. This rotation then pulls individual magnetic beads through precisely controlled orbits around the magnetic features, as the force of magnetic attraction pulls the bead continuously to the new pole location. This array of orbiting beads acts as an active mixer and is easily integrated into a continuous-flow system. The mixing can be actuated independently of the fluid flow rate through control of the rotation rate of the external magnetic field. This system uses low magnetic hysteresis materials, so there is minimal heating

inside the channel and this method is compatible with biological applications. Unlike the mixing applications of magnetic beads described above [15, 16], this device precisely controls the motion of individual microbeads, which are held in place purely by magnetic forces, and does not require the use of size exclusion to keep beads from exiting the system. Additionally, magnetic microbeads can be functionalized to capture specific biological particles from the mixed fluid. The captured sample could then be readily extracted for analysis by removing the external magnetic field, thereby de-magnetizing the magnetic beads and features, and flowing away the magnetic beads with their bound targets for further examination. Thus, our approach can potentially yield a multi-functional microfluidic system for mixing, capture, and analysis of biological and other samples.

2 Fabrication and Methodology

2.1 Microfabrication

Nickel-iron (NiFe) features were created by evaporating permalloy (80% Ni, 20% Fe pellets from Kurt J Lesker) to a thickness of 350 nm onto a bare soda lime glass wafer (100 mm diameter, 550 μm thick) via e-beam evaporation. During evaporation, the chamber was heated to 250 $^{\circ}\text{C}$ to improve adhesion of the film to the substrate, such that an adhesion layer was not required. An etch mask was created using positive photoresist (Microposit SC1827), protecting regions of the NiFe. The unprotected metal was then removed via wet etching in Al Etch A (Transene Company Inc.). After removing the positive resist etch mask, 150 nm of silicon dioxide (SiO_2) was deposited over the entire wafer via standard plasma-enhanced chemical vapor deposition (PECVD) at 250 $^{\circ}\text{C}$. This SiO_2 layer protects the NiFe features from oxidation in aqueous solutions and provides a uniform layer for surface passivation. These steps are outlined in Figure 2. Completed wafers were then annealed at 300 $^{\circ}\text{C}$ for 30 minutes (2 $^{\circ}\text{C}/\text{min}$ ramp rate) to improve PECVD SiO_2 film quality.

2.2 M-280 Denaturing

Dynabeads M-280 from Life Technologies were used as magnetic beads for experiments. The M-280 beads exhibit superparamagnetic properties and are pre-functionalized with streptavidin. To reduce the interaction between the M-280 functionalized protein and the fluorescent nanoparticles used in these experiments, the M-280 beads were placed in a 0.6 mL polystyrene conical tube and incubated in boiling water for 30 minutes.

2.3 PDMS Channel Attachment and Passivation

A 10:1 polydimethylsiloxane (PDMS) microfluidic channel (6.7 μm height, 150 μm wide) was made via standard molding using an SU-8 mold (Figure 3). After removing the PDMS microfluidic

channel, holes were punched into the PDMS, creating four fluid reservoirs. The four reservoirs are: the M-280 bead loading reservoir, reservoirs for the fluorescent and non-fluorescent streams used in mixing experiments, and the outlet reservoir. The PDMS channel is then manually placed onto a glass wafer with NiFe features creating the enclosed array. For the series of experiments presented in this paper, the geometry of the array is shown in Figure 4. To limit non-specific adsorption, a solution of 0.1% (weight to volume) bovine serum albumin (BSA) in deionized (DI) water is loaded into the reservoirs and channels, making sure to avoid air bubbles, and allowed 1 hour of incubation to ensure adequate channel passivation.

2.4 Loading of Magnetic Beads

After passivation, the glass wafer with attached PDMS channel is placed onto an inverted microscope (Nikon TES2000) stage. The M-280 beads are diluted from stock concentrations into DI water with 0.1% BSA at a ratio of 1:25. A small volume (1-3 μL) of the diluted M-280s is loaded into the bead loading reservoir and then pumped to the outlet reservoir via electro-osmotic flow (EOF). A permanent magnet (NdFeB, Grade N52, 6.35 mm by 6.35 mm by 6.35 mm, from K&J Magnetics, Inc.) affixed to the spindle of a DC motor is suspended over the magnetic features while rotating. As shown in Figure 5, M-280 beads can enter the region of NiFe features as clumps or strings of beads, but they are effectively distributed across the array of NiFe features in a relatively uniform fashion, such that on average NiFe features control a single microbead at each of its magnetic poles. We have previously demonstrated that the beads remain synchronized with the magnet rotation up to rotational speeds of 9,000 revolutions per minute (RPM) [18] and that they can be controlled with flow velocities at least as high as 12 mm/s [21].

2.5 Mixing Experiments

Two streams were pumped via EOF. A 90V potential was applied, so as to produce linear flow rates of 10-30 microns/second, measured from captured images. One stream contained DI water with 0.1% BSA and the other contained the same but with a 0.1% concentration of fluorescent nanoparticles (Fluospheres Yellow-Green, 40nm, from Life Technologies). The speed of the orbit of the magnetic beads is controlled by the speed of the suspended DC motor and the effect on mixing of the two streams was recorded via Photometrics CCD camera (Cool Snap HQ).

2.6 Image Analysis

Images and videos were captured via an inverted microscope (Nikon Eclipse) with a CCD camera (Photometrics Cool Snap HQ). Since the device is viewed from below, the NiFe features appear as dark

spots in a field of fluorescence. Under each set of flow conditions, multiple images were taken of the section of the channel containing beads. To calculate mixing at the channel outlet, each image was cropped down to a 66 pixels wide by 225 pixels tall section representing the 20 μ m section of channel immediately after the array, and was imported into MATLAB for analysis. Each row of 66 pixels was averaged into a single intensity value to create an intensity profile across the width of the channel. This averaging helps to smooth out outlier intensity values that can result from clusters of nanoparticles or other non-uniformities in the flow.

2.7 Numerical Simulation

In order to simulate microfluidic mixing using orbiting magnetic microbeads, a computational method for fluid-structure interactions [22-24] was employed based on the lattice Boltzmann method [25-27] to simulate a viscous incompressible fluid in a microfluidic channel. Surfaces of the solid superparamagnetic beads were triangulated to impose no-slip and no-penetration boundary conditions with surrounding viscous fluid [26, 28, 29]. Trajectories of the superparamagnetic beads were determined as described previously [18, 21]. This model was coupled to a stochastic Brownian dynamics (BD) model [30, 31] that calculates the trajectories of 15 million diffusive particles and calculates the particle concentration field, so as to solve the convection-diffusion equation and model microfluidic mixing.

3 Results and Discussion

3.1 Mixing of Stationary Fluid

Two streams were pumped over the mixing array with the magnetic microbeads held stationary. The pumping was stopped, leading to rapid deceleration of the fluid. After the deceleration, the magnetic microbeads were started rotating at 4000 RPM as a video recorded the results. Still images from the video are shown in Figure 6. The magnetic beads are spinning clockwise in the image. The video was recorded with an inverted microscope and thus the NiFe features appear as black circles surrounded by fluorescence. Orbiting beads are not visible, since their movement is faster than the camera exposure time.

The first noticeable effects in the images of Figure 6 are coronae surrounding the NiFe features and stretching across the microchannel, which coronae are visible at $t = 1$ s and more pronounced at $t = 2$ s. These columns of coronae form due to fluid moving along circulatory and cross-channel patterns due to the rotating microbeads dragging fluid in their orbits and passing fluid to the next orbiting bead, as will be addressed in more detail in Section 3.3. This fluid motion transports fluorescent particles across the channel, resulting in mixing. In this condition with no net flow down the channel, the transported fluorescence travels down a single column of NiFe features with little discernible transport to the left or

right in the image. In some regions the fluorescence is transported across the channel in less than 2 seconds while some regions fill in more slowly. By the end of the image sequence, the coronae have disappeared, replaced with a uniform diffuse fluorescence across the entire array.

The fastest regions of transport are at the outer edges of the array. At the left edge of the array, the rapid transport effect is due to the beads orbiting on the left edge of the mixing array always moving upward when they are on the portion of their trajectories that is external to the array. This results in the magnetic microbeads pulling the fluid external to the array across the channel (upward in Figure 6). The beads on the right edge of the array pull fluid across the channel in the opposite direction (downward in Figure 6) due to the same mechanism. Over time, this accounts for the accumulation of fluorescence in the bottom right corner as fluorescent fluid is pulled to the bottom of the channel. The void of fluorescence formed in the top left corner is the result of non-fluorescent fluid being pumped from the bottom to the top. This effect is minimal in a channel with a net axial flow, since in this case fluid does not stay at the edge of the array long enough to be moved across the channel by this localized effect. A similar effect of rotating beads on fluid pumping due to asymmetric channel geometry was recently reported [32]. Within the array itself, the advancement of fluorescence across the channel occurs in pockets. This is likely due to irregularities in bead coverage across the array of NiFe features.

3.2 Mixing under Flow

Since a major application of our micromixer is the use in continuous-flow systems, the mixing effectiveness of the rotating beads was investigated under flow. The system presented here contains two independent velocities of note. The first is the linear flow velocity derived from the volumetric flow rate of the bulk channel fluid. In the experiments presented here, the linear flow velocity U ranged from 30 $\mu\text{m/s}$ to 110 $\mu\text{m/s}$. The second velocity of note is the linear velocity of the magnetic microbeads as they rotate around the NiFe features. The velocity of the magnetic microbeads, V_b , can be calculated from the path of the magnetic microbeads, approximated by the perimeter of the NiFe features, and the rotation rate of the external magnetic field. In our experiments, the linear velocity of the beads varied from 20 $\mu\text{m/s}$ to 1400 $\mu\text{m/s}$. Since our system operates at a low Reynolds number, the ratio of the flow and bead velocities U/V_b is more pertinent to system behavior than the actual magnitude of either velocity [21]. This is provided that the flow rate and channel length are such that diffusion alone does not lead to a noticeable mixing over the length of the mixing region, which was the case for our system. Indeed, we observed through experiments and simulations with varied U and V_b that system behavior was defined by the ratio U/V_b and not by magnitudes of U or V_b . Thus, we use this velocity ratio to describe the flow inside the array of NiFe features with rotating magnetic microbeads.

Figure 7 contains still images recorded under various values of U/V_b . Little mixing is visibly evident in the range of moderate to large U/V_b . However, strong mixing was observed for very low values of U/V_b (Figure 7c), as is evident by the separated fluorescent streams at the inlet to the mixing section (at left) becoming nearly uniform as they pass through the mixing array, such that the area of the channel at the outlet (at right) exhibits a diffuse fluorescence across the entire channel width. Thus, effective mixing occurs under flow conditions in which the velocity of the magnetic microbeads is much faster than that of the bulk fluid. Similar to the effects seen in the stationary flow experiment, corone of fluorescence surround the NiFe features and reach across the channel width as orbiting beads transport fluid further across the channel. Whereas under stationary fluid the fluorescence travels along a single column of NiFe features, under a net flow the fluid and fluorescence is pushed continually downstream and thus does not travel along a single column. Instead, the fluorescent particles are pushed to the neighboring column where it is caught in the corona around the neighboring feature. As the fluorescence reaches each neighboring column, some of it is transferred further across the width of the channel until it reaches the far channel wall. The physical mechanisms behind this motion are explored in greater detail for the limit of low U/V_b in Section 3.3.

The regions immediately past the microbead array were isolated for analysis, and profiles of the fluorescent intensity as a function of the distance across the channel were plotted in Figure 8. The plots confirm the observed mixing, with the fluorescence profile at the mixing section outlet region becoming more uniform with decreasing values of U/V_b . At high values of U/V_b , there is a sharp gradient in the fluorescent profile across the channel, indicating separated streams of fluid. From Figure 8, it is evident that for velocity ratios below approximately $U/V_b = 0.1$, the gradients in the fluorescence profile become more gradual, such that by $U/V_b = 0.04$ the fluorescence is fairly uniform across the channel.

Fluorescence intensity profiles were used to calculate a mixing degree, σ , using the relation $\sigma = 1 - 2 \left(\sqrt{\frac{1}{N} \sum_{i=1}^N (\theta_i - \bar{\theta})^2} \right)$, where θ_i is the normalized fluorescent intensity at a point i and $\bar{\theta}$ is the average normalized fluorescent intensity across the entire channel. The normalized fluorescent intensity in each image was found by normalizing the pixel values in each image by the maximum fluorescence intensity in that image. This puts all fluorescent intensity profiles on a scale from 0 to 1. With this definition of the mixing degree, an increasing level of σ corresponds to an increasingly mixed fluid region. When $\sigma = 0$, the fluid streams are perfectly separated, and when $\sigma = 1$ they are perfectly mixed. Figure 9 shows the mixing level σ after the 270 μm mixing section for test runs using various values of U/V_b . As shown by these data, there is a stark increase in mixing for $U/V_b < 0.1$. Above this threshold value of U/V_b , orbiting microbeads did not provide for a significant mixing improvement over the short mixing distance used in these experiments.

As a point of comparison, we performed experiments in which the bead rotation rate was slow, yielding a velocity ratio of $U/V_b \approx 5$. In this case, the fluid streams enter the mixing section with $\sigma = 0.267$ and exit at $\sigma = 0.261$ showing minor effect of either the beads or diffusion on mixing over this short mixing distance. This mixing level is close to the mixing level achieved by beads orbiting at all velocity ratios in the range of $U/V_b > 0.1$. This result indicates that that our device should be used within the velocity ratio range of $U/V_b < 0.1$ in order for to efficiently mix fluids in a short mixing region.

3.3 Numerical Simulations

Numerical simulations were used to determine the flow fields generated by the beads, so as to understand the physical mechanism of mixing and to explain why mixing drops off rapidly with increasing velocity ratio. In Figure 9, we compared our simulation results with the experimental data. In the simulations, we used inlet boundary conditions which approximately matched the inlet intensity profile shown in Figure 8. We found that the predicted mixing levels were close to those measured in the experiments. Specifically, the numerical simulations confirmed the critical threshold of approximately $U/V_b \sim 0.1$ below which significant mixing was obtained. Some discrepancy in the results in Figure 9 between the experiments and simulations can be attributed to clustering of fluorescent particles, non-uniform bead coverage, and non-uniformities in the inlet flow.

To understand the physical mechanism of mixing by orbiting microbeads, we examined the flow structures forming in the microchannel. Figure 10 shows the streamlines of the period averaged velocity fields generated by the beads at various velocity ratios. As seen in Figure 10a, orbiting microbeads in an otherwise stagnant fluid ($U/V_b = 0$) create circulatory flow cells that are linked by streamlines that cross the channel, thus advecting fluid across the channel and stretching the fluid interface, leading to greatly enhanced mixing. This is evidenced by the circular patterns of florescence leading across the channel in the experiments of Figures 6-7. As flow down the channel is increased, the circulatory cells gradually shrink, and the streamlines linking cells and crossing the channel begin to instead turn downstream and eventually do not cross the channel, thus stopping the bulk advection of fluid across the channel and decreasing mixing.

Physically, the streamlines crossing the channel are due to the orbiting beads dragging fluid, some of which is left in their wake and picked up by adjacent beads and dragged further across the channel. Indeed we have recently showed that the mixing effectiveness of orbiting magnetic microbeads increases monotonically with increasing bead coverage density leading to reduced distances between adjacent beads [21]. When there is a net flow down the channel, some of the fluid left in the wake of orbiting beads is pulled downstream out of the path of oncoming beads, and thus the streamlines turn downstream rather than crossing the channel and linking transverse cells, thereby reducing the mixing efficiency [21].

These findings on the decay of cross-channel streamlines explain the drop in mixing by orbiting beads with increasing U/V_b beyond approximately 0.1 as shown in Figure 9. This is also consistent with the fluorescence intensity profiles in Figure 8, where relatively uniform profile curves are shown for $U/V_b < 0.1$, whereas profiles for greater velocity ratios indicate separated fluid streams. Thus, this device can best be used with beads orbiting at a sufficient rate such that $U/V_b < 0.1$. For an in-depth numerical study on the effect of various geometrical alterations to this system over a large range of velocity ratios, we refer the reader to the recent work of Ballard et al [21].

4 Conclusion

This paper presents findings on a novel method for mixing inside a microfluidic channel using a regular array of orbiting magnetic microbeads which are actuated by an external magnetic field. Microfluidic mixing in a 270 μm -long channel is demonstrated. The time required to mix fluid in our experiments was between 2.5 and 9 seconds depending on the flow velocity. We note that this time can be further reduced by increasing the rotation rate of the beads, as we did not approach the upper limits on the velocity [18, 21].

Effective mixing has been shown through both experiment and numerical simulation. In this paper we focused on mixing in a relatively short mixing section, although a longer array with more columns of features and more magnetic beads could produce even greater mixing by having a longer residence time. Alternatively, U/V_b could be further lowered to achieve equal or greater mixing in even shorter mixing sections.

In addition to the ability to provide for mixing over a distance much shorter than is done in most existing microfluidic mixers, this system also has the capacity to actuate mixing independent of fluid flow, and is compatible with conditions required for analysis of biological samples. Furthermore, our system enables simple distribution across the array of magnetic beads that can be held in place and precisely controlled in orbits by magnetic forces without additional control systems. Finally, functionalized magnetic beads can be used to capture specific targets from the mixed fluid sample and, after the magnetic field is removed, can be released for consequent analysis in the following sections of the microfluidic channel, thus giving the system potential for multistage use in complex microfluidic lab-on-a-chip systems.

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Figure 1. The soft magnetic NiFe features are magnetized by an external magnetic field creating magnetic poles on the feature. Superparamagnetic microbeads are attracted to the poles of the NiFe feature. Rotating the external magnetic field changes the position of the magnetic poles and pulls the magnetic beads along with it, such that they are driven in controlled orbits around the NiFe feature.

Figure 2. MEMS Process flow for creation of NiFe features. A key feature in this process is that the elevated chamber temperature during NiFe evaporation allows for deposition and further processing without the use of an adhesion layer.

Figure 3. PDMS microchannel structure with reservoirs (overall dimensions 4 cm x 2.5 cm). An SU-8 mold is used to create PDMS microchannels. After molding, four holes are punched into the PDMS to create reservoirs. Finally the PDMS microchannel is manually aligned and placed over the array of NiFe features.

Figure 4. Dimensions of a microfluidic channel with its floor patterned with NiFe features. All dimensions are in microns (μm). The channel height is $6.7\mu\text{m}$. The total length down the channel occupied by the NiFe features is $270\mu\text{m}$.

Figure 5. a-d) Time-lapse images of self-dispersal of magnetic microbeads which reached the NiFe feature array in the form of a large clump (dark spot at left). e) Image of the magnetic beads after they have self-dispersed, resulting in a relatively uniform coverage of magnetic beads across the array, with each NiFe feature on average controlling a single magnetic bead at each of its magnetic poles.

Figure 6. Time lapse fluorescence images of a microchannel with beads rotating at 4000 RPM, resulting in mixing in an otherwise stagnant fluid.

Figure 7. Fluorescence images showing mixing due to rotating magnetic beads in a continuous-flow microfluidic channel with net flow and bead velocities leading to varied velocity ratios, a) $U/V_b = 4.8$, b) $U/V_b = 0.15$, c) $U/V_b = 0.04$.

Figure 8. Normalized fluorescence intensity profiles at the inlet and after a 270 μm mixing section with beads orbiting at various velocity ratios. The profiles shown here represent averaging over 6 images. The flattening of the curve represents the mixing effect achieved by the rotating magnetic beads.

Figure 9. Mixing degree after a 270 μm mixing section, for various flow conditions from measured experiments and numerical simulations. In both experiments and numerical simulations, a significant increase in mixing was observed for $U/V_b < 0.1$.

Figure 10. Numerically calculated streamline plots of the period-averaged velocity of the fluid at the half-height of the orbiting microbeads in a representative periodic section of the microchannel, shown for the following velocity ratios: a) $U/V_b = 0$, b) $U/V_b = 0.025$, c) $U/V_b = 0.05$, d) $U/V_b = 0.1$, e) $U/V_b = 0.15$, f) $U/V_b = 0.2$.