



**DESIGN OF MICROMIXER FOR PRECISION MILLED MICROFLUIDIC
SYSTEMS**

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**DESIGN OF MICROMIXER FOR PRECISION MILLED MICROFLUIDIC
SYSTEMS**

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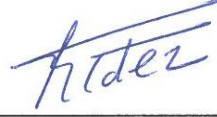
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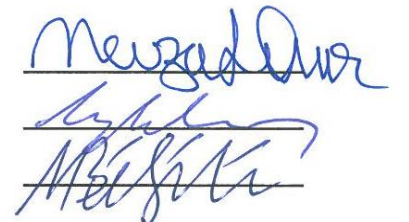
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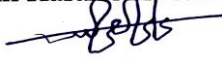
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ABSTRACT

DESIGN OF MICROMIXER FOR PRECISION MILLED MICROFLUIDIC SYSTEMS

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Mixing in microfluidic systems is a challenging problem since the flow is almost always laminar hence the transport of species depends only on diffusion. In this thesis, a passive micromixer is designed for mixing two miscible liquids in a micro channel. The mixer utilizes series of throttles, which reduce the diffusion length, placed along the mixing channel. Although there are many fabrication techniques to manufacture microfluidics, the throttles constituting the micromixer are specially designed to be fabricated by micromilling. Here, micromilling is particularly chosen because of its flexibility and productivity compared to other microfluidic fabrication methods.

For design purposes, firstly the parameters affecting the mixing performance are determined. These parameters are defined as the Reynolds number (Re), non-dimensional throttle size and the number of throttles. Then, micromixers are simulated using computational fluid dynamics tools available in COMSOL Multiphysics at different parameter levels. As a result, it is found that micromixers with 15 to 20 throttles can achieve a mixing efficiency greater than 80% in very low Re flows. After optimizing the design, the micromixer is improved by shifting the

throttles off the axis of the mixing channel. It is seen that the improvement increases the mixing efficiency to 84%.

Keywords: Micromixing, Micromilling, Microfluidics, Computational Fluid Dynamics

ÖZ

HASSAS FREZELEME İLE ÜRETİLMİŞ MİKRO AKIŞKAN SİSTEMLER İÇİN MİKRO KARIŞTIRICI TASARIMI

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Mikro akışkan sistemlerde karıştırma, akışın neredeyse her zaman laminer olması ve buna bağlı olarak taşınımın sadece difüzyona bağlı olması nedeniyle zor bir problemdir. Bu tezde, birbiriyle karışabilen iki sıvının bir mikro kanal içerisinde karıştırılması için pasif bir mikro karıştırıcı tasarlanmıştır. Karıştırıcı, difüzyon mesafesini kısaltan ve karıştırma kanalı boyunca yerleştirilmiş olan bir dizi boğumdan faydalanmaktadır. Mikro akışkan sistemlerin imalatı için birçok üretim tekniği olsa da, mikro karıştırıcıyı oluşturan boğumlar özellikle mikro frezeleme ile üretilecek şekilde tasarlanmıştır. Burada mikro frezeleme, diğer mikro akışkan üretim yöntemlerine göre esnekliği ve üretkenliği sebebiyle özellikle seçilmiştir.

Tasarım amacıyla, ilk olarak karıştırma performansını etkileyen parametreler belirlenmiştir. Bu parametreler Reynolds sayısı (Re), boyutsuz boğum büyüklüğü ve boğum sayısıdır. Sonrasında mikro karıştırıcıların, COMSOL Multiphysics içerisinde yer alan hesaplamalı akışkanlar dinamiği araçları kullanılarak farklı parametre seviyelerinde simülasyonları yapılmıştır. Sonuç olarak, 15-20 boğumlu mikro karıştırıcıların çok düşük Re akışlarında %80'in üzerinde karıştırma verimine ulaşabildikleri bulunmuştur. Tasarım eniyilendikten sonra, mikro karıştırıcı,

boğumlar kanal eksenine göre kaydırılarak iyileştirilmiştir. Bu iyileştirmenin karıştırma verimini %84'e yükselttiği görülmüştür.

Anahtar Kelimeler: Mikro Karıştırma, Mikro Frezeleme, Mikro Akışkanlar, Hesaplamalı Akışkanlar Dinamiği

*To the memory
of my dear father*

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Thanks to God the most compassionate and the most merciful. May Allah's mercy and peace be upon our prophet Mohammed, who invites us to science and wisdom, and his household.

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CHAPTER 1

INTRODUCTION

Recently, there have been an increasing interest in academy and industry in microfluidics. Microfluidics is the field or system dealing with small amounts of fluids, generally with microliter scale or less. The minimization arises many applications in biomedical and biochemical fields. Due to that minimization, the possibility of establishing a laboratory on a single chip was appeared, which would be disposable and portable. Some applications which are enabled by the use of these lab-on-a-chip devices include separation of reagents, DNA analysis, cell culturing, drugs testing, and electrophoresis for DNA sequencing [1]. Besides, there are more applications such as, DNA synthesis, interaction of enzymes, folding proteins, cancer screening, observation of water pollution, and monitoring of changes in the environment [2– 4]. For these applications many components were developed to regulate fluid flow such as micromixers, microvalves, micropumps, and other devices. For example in many of the aforementioned applications, good mixing of components is important in order to get accurate and fast analysis. Hence, micromixers play an important role in microfluidics because they reduce interaction time, and increase interaction efficiency.

In microfluidic systems Reynolds number (Re) is quite small since Re is a function of channel width and fluid velocity, which are quite small in microfluidics. Therefore, turbulent flow does not occur. Thus, the flow is always laminar. Mixing process in laminar flow range is slow because of low convection of species and diffusion perpendicular to flow is low. Therefore, the necessity of including a special design, micromixer, to enhance mixing performance is emerging.

Micromixers can be classified into two types: active and passive micromixers. As it is discussed in following sections, an active micromixer needs external power to improve mixing process in contrast to the passive micromixer, which does not require an external power but needs modification of the channel geometry to enhance mixing. By comparing passive micromixers with active micromixers, active micromixers are almost more efficient compared to the passive micromixers. On the other hand, passive micromixers require no external power, do not include any moving parts, and are easier to integrate with other microfluidic components. Consequently, the researchers widely prefer passive micromixers for many microfluidic applications.

The major objective of micromixers is to get a perfect mixing of two or more components in micro-volume scale within short time. The complete mixing in passive micromixers is difficult to achieve and it needs a quite long diffusion length, since the mixing process depends on diffusion only. In addition to diffusion effect in mixing process, the mass transport by convection in same direction of diffusion can improve the mixing performance. For that, enhancing mixing performance needs modulation in mixing channel.

In this study, simple throttles are utilized along the mixing channel to improve mixing efficiency. The presented micromixer is designed particularly for micromilling, which is a suitable, rapid, and efficient microfluidic fabrication method to obtain complex geometries [5]. The micromixer presented in this thesis is a simple passive micromixer. Besides, the present micromixer is easy to create because it does not consist of complex geometry and it has an acceptable mixing efficiency. In addition, the presented micromixer works at low Reynolds numbers, less than 1.

The expected function of the throttles is to work as barriers on the side walls to reduce the width of the mixing channel. Decreasing the channel width confines two streams into narrower stream, which decreases the diffusion length (z) [6]. This, in turn decreases the mixing length (x) required to achieve a complete mixing in accordance with the following equation:

$$x = Uz^2 / D \quad (1.1)$$

where x is the mixing length, U is the fluids velocity, z is the stream width, and D is the diffusion constant for the species of interest within the working fluid. Reducing mixing length improves the mixing performance. Furthermore, the throttle utilized in our micromixer reduces the stream width and speeds up the mixing process [6, 7]. Furthermore, the throttle size also affects the mixing performance. In this study, we optimized the throttles by considering the throttle size and number of throttles to maximize the mixing efficiency. Finally, we presented a possible improvement, which utilized throttles offset from the axis of the mixing channel to make a zigzag series of throttles. The benefit of this zigzag series is, in addition to reduce the stream width, to trigger chaotic advection for species. The chaotic advection increases mass transport by convection between the two streams, which enhances mixing efficiency [8, 9].

1.1. Thesis Organization

The thesis is organized in five chapters. Where, Chapter 2 contains the literature review. The first part of Chapter 2 presents a brief information on micromixers and its applications, micromixer types, microfluidic fabrication methods, and design of the proposed micromixer. The second part focuses on literature review of design and simulation of passive micromixers. Chapter 3 explains the details of the proposed micromixer, simulation and optimization the micromixer by choosing a set of input parameters that gives the highest mixing efficiency. Chapter 4 presented an improvement of the proposed micromixer by offsetting the throttles. Finally, Chapter 5 concludes the study and presents an outlook for the future research.

CHAPTER 2

LITERATURE REVIEW

2.1. Review of Micromixers

Micromixer is an important component, which integrates with other micro components to produce a microfluidic system. The valuable impact of the micromixer is shown in many fields of science such as drug development, biomedical diagnostics, life sciences and it has been extensively applied in the food and chemical industries [10, 11]. The reason of the micromixer importance is that many of these applications need a rapid and good mixing of the reagents and working fluids involved. However, good and rapid mixing is difficult to achieve because of the laminar flow, which is a result of low Re. Hence the mixing process depends only on diffusion of the species. On the other hand, the micro scale involved increases the surface to volume ratio, which is desirable for the micromixers.

Micromixer applications are almost apparent in chemical, biological and medical analysis fields. Nguyen in his study [2, 12] mentions many of the micromixer applications.

2.1.1. Types of Micromixers

The main goal of micromixers is to obtain a perfect mixing of two or more fluids within a short time. However, it is difficult to achieve a perfect mixing in laminar flow regimes, which is almost always the case in microfluidics. The flow regime is characterized by the Reynolds number (Re), such that, for Re smaller than 2000 the flow is laminar. When Re is between 2000 and 4000 flow is transitional, but when Re is greater than 4000 the flow is turbulent.

Reynolds number is calculated by

$$Re = \rho U w / \mu \quad (2.1)$$

where ρ is density of the fluid, μ is the dynamic viscosity, U is the flow velocity and w is the characteristic dimension of the flow, which is often the channel width in microfluidics. For instance, in a microfluidic channel of 100 μm width, which involves water flow at a velocity equal to 1 mm/s, assuming that the water density (ρ) is 1000 kg/m^3 and the viscosity (μ) is 0.001 Pa.s, then Re comes out to be in the order of 0.1. As a conclusion, the Reynolds number in microfluidic systems is very small. Hence, diffusion plays the main role in mixing process instead of convection. Fig. 1 illustrates the effect of laminar flow in microfluidics. Fluids with different colors streaming through 8 branches continue flowing as separate lanes through the main channel. It should be noted that there is no permeation between these laminar streams. Since the aim of micromixers is to improve the mixing efficiency, therefore, the diffusion effect must be enhanced between the different flowing species [13]. To improve mixing performance for any micromixer, external forces may be needed to cause turmoil for the species, to increase the contact area, or the contact time between the species. Generally, the micromixers are classified into two types: active and passive micromixers.

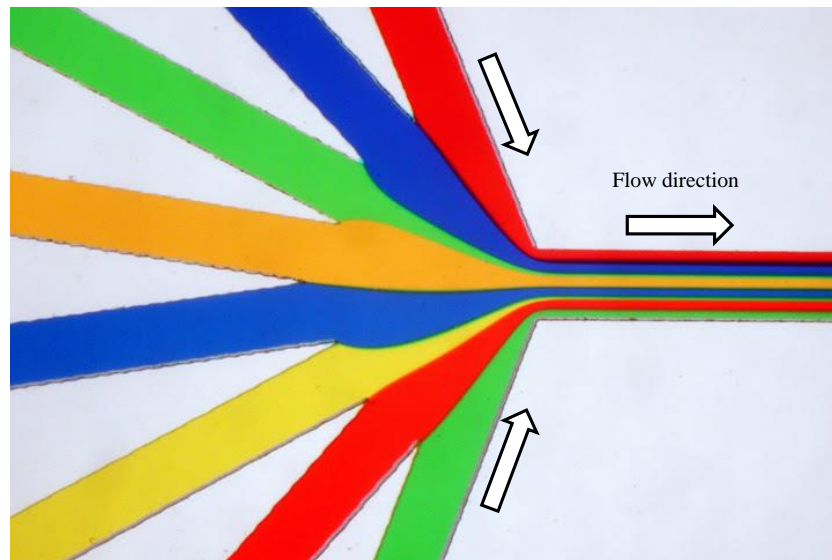


Figure 1: Example on microfluidic device with multiple inlets. The laminar flow in such microfluidic devices is clear in this figure (Source: Folch Lab Gallery, University of Washington) [14].

2.1.1.1. Active Micromixers

This type of micromixers need an external effect to operate the mixing process, which be achieved by adding micropumps, or by generating a thermal, electric or magnetic field to manipulate the streams. Active micromixers generally reduce the mixing time significantly. However, their production is not easy because of the fact that the active micromixer consists of active components (moving parts, electrodes, etc.) [15].

In active micromixers, the necessity of external power emerges to improve the mixing performance through stirring the flowing fluids. There are different various schemes used in active micromixers such as electro-hydrodynamic, thermal acoustic/ultrasonic, electrokinetic, pressure perturbation, magnetic, or dielectrophoretic (Figure 2) [13]. Lee et al. [13] summarize the most common external powers that the designers use in their micromixers.

Figure 3 illustrates an active micromixer, which uses electromagnets to stir the fluids to enhance mixing performance.

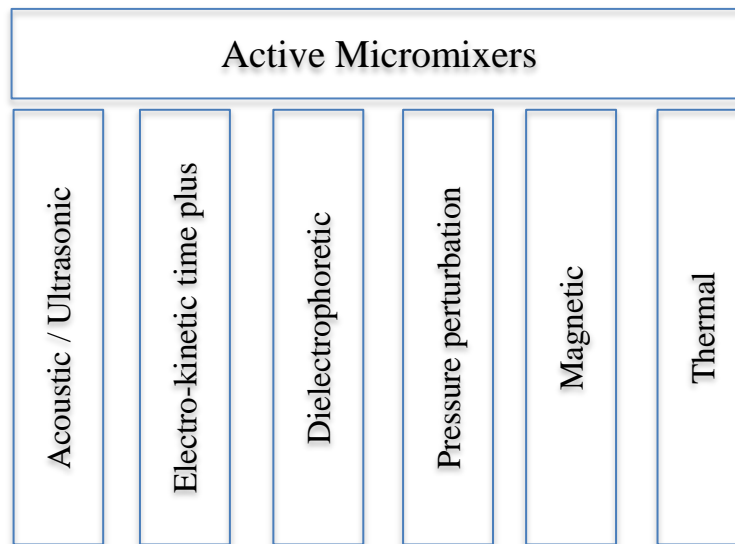


Figure 2: Widely used active micromixers [13].

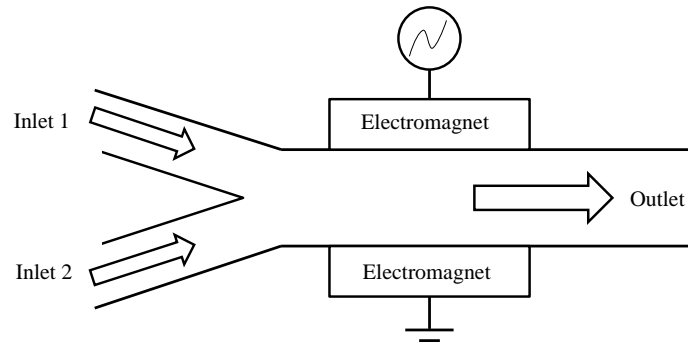


Figure 3: An active micromixer which uses electromagnets to improve mixing efficiency [16].

2.1.1.2. Passive Micromixers

In this type of micromixers there is no external force applied on the fluids to facilitate mixing. As stated before, diffusion between working fluids plays the main role in mixing process. In this case, to improve the mixing performance the contact interface area or the contact time should be increased. For instance, increasing interface between the fluids can be done by splitting the flow into streams and recombining them again [17]. Another scheme can be using obstacles in mixing channel to facilitate a rapid mixing [15].

Mixing in passive micromixers can also be enhanced by utilizing chaotic advection [18], where vortices are induced within the channel by changing the channel geometry. Figure 4 shows a particular passive micromixer, which has Tesla structure [19] to improve mixing performance by utilizing splitting and recombining streams. Lee et al. [13] summarize the most common passive micromixers (Figure 5).



Figure 4: Schematic drawing of one of Tesla type micromixer [19].

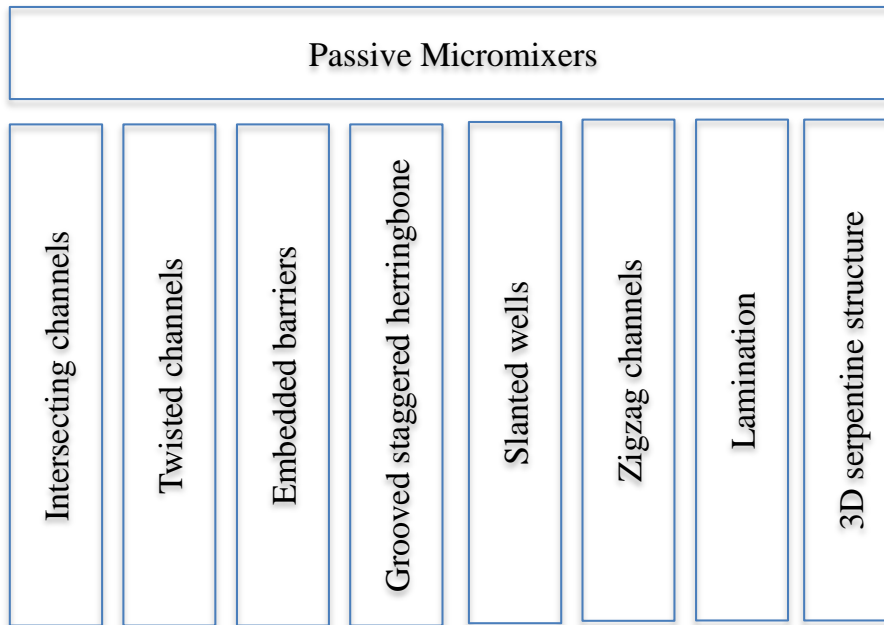


Figure 5: Explanation the common types of passive micromixers [13].

2.1.2. Microfluidic Fabrication Techniques

There are many methods used to fabricate microfluidic devices, hence micromixers. Each technique has its own advantages and disadvantages. The choice of suitable technique depends on the geometry and the dimension of the micromixer. In addition, it relies on the accuracy that the designer needs and the budget of the project. There are several methods, which are widely used for microfluidic fabrication, such as PDMS molding, micromilling, hot embossing, and microinjection molding.

In PDMS molding, an elastomeric polymer, polydimethylsiloxane (PDMS) mixed with a curing agent is poured on a micromold and cured by applying heat [20]. The micromold (or the master) is often fabricated using standard microfabrication techniques such as photolithography. Figure 6 illustrates the steps in microfluidic fabrication by PDMS molding. PDMS molding is currently the most common microfluidic fabrication technique due to its robustness. However the process can be used only with PDMS and often requires a long time.

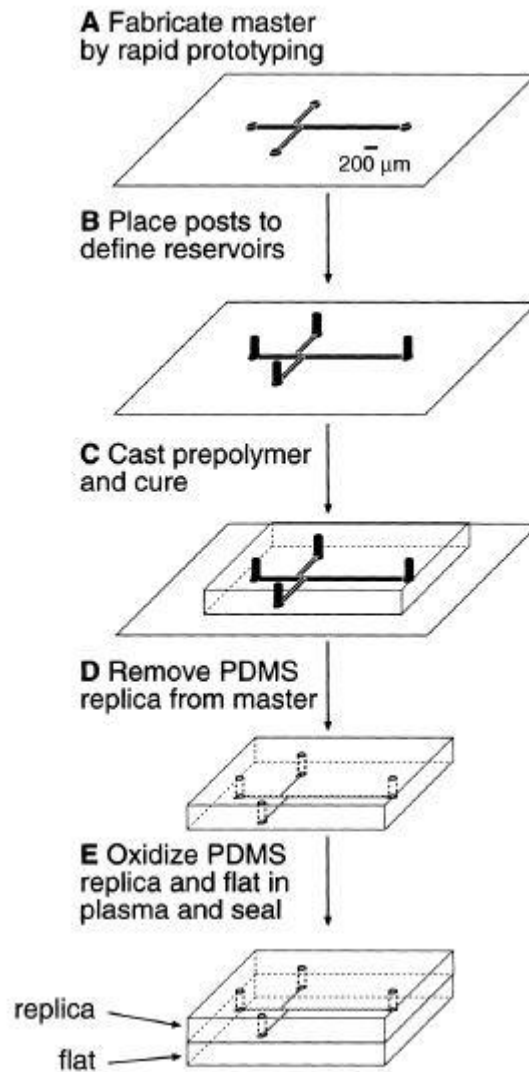


Figure 6 Illustration for PDMS molding [20].

The second method, used for microfluidic fabrication is micromilling. Micromilling is modification of conventional milling. Micromilling can be considered as a micro-scale milling. In this method CNC machining centers are used to mill the microchannels on plastic substrates. In this method, milling tools are commonly end mills and have diameters less than 1 mm. Micromilling is a suitable technique to fabricate microchannels with high aspect ratio, complex geometries and internal structures. However, the size of the concave features are limited with the diameter of the end mill, which should not be smaller than 50 μm the smallest end mill [21]. However, the convex features do not have the same limitation, but the milling tool must resist the cutting forces without elastic deformation and the dimensions around these convex features. The micromilling technique is fast, cost effective, and flexible

method with respect to other fabrication methods [21]. The disadvantages of this method are due to the low structural stiffness of the milling tool, and the deflection that was caused by the cutting forces may not be negligible. This results in variations between design dimensions and actual dimensions after milling. The important drawback of micromilling method is the limitation of the resolution and the deformation of material problems such as the presence of burrs and cutting tool marks [21]. Figure 7 illustrates micromilling process and a CNC machine tool [5].

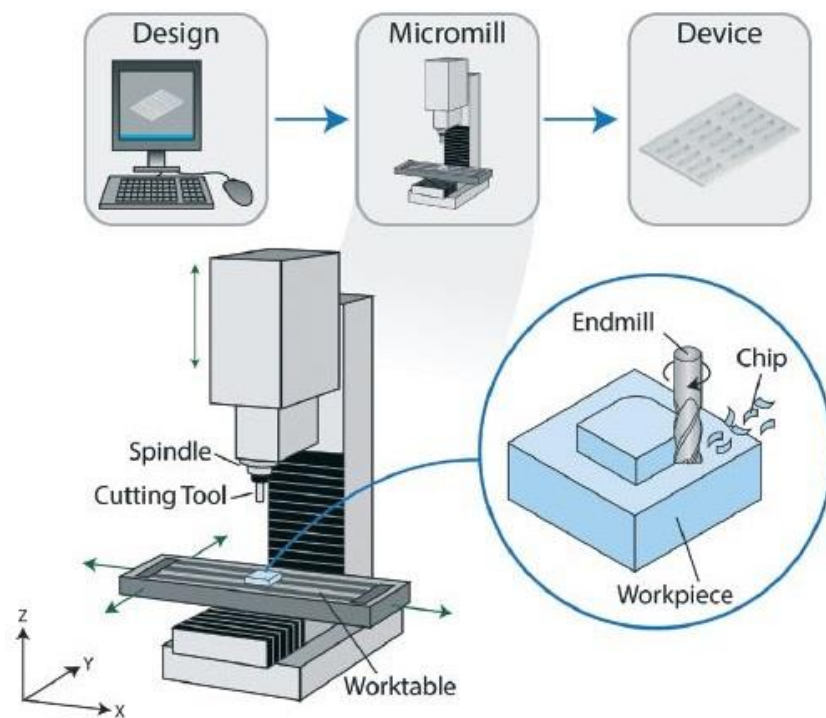


Figure 7 Illustration of micromilling process [5].

Another method is hot embossing. It is a technique for the replication of a boss mold that has a permanent pattern on the side that faces the substrate in thermoplastic sheets. Hot embossing is done in four steps (Figure 8):

1. Heating the substrate above glass-transition temperature between lower and upper micro-mold plates.
2. Moving the upper mold down and embossing the substrate with an imposed pressure at the hot embossing temperature.
3. Cooling down the substrate.

4. Opening the mold to obtain substrate with micro cavities that represent the microchannels [22].

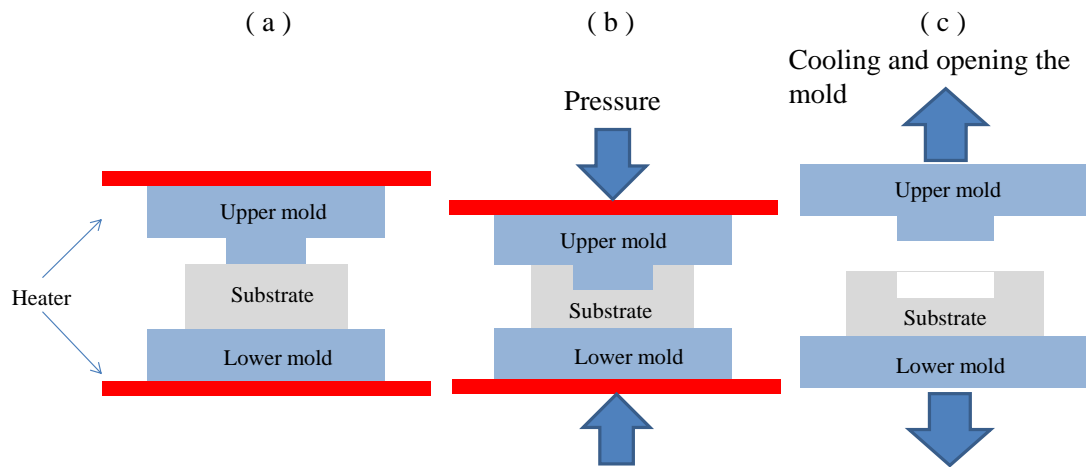


Figure 8 Hot embossing process steps. a) Heating the substrate. b) Embossing the mold with substrate. c) Cooling the substrate and mold and opening the mold to take out the substrate.

In hot embossing cost per part is often low and the mold can be used for several times. However, the process is not suitable for prototyping since it requires fabrication of mold. Generally the mold is fabricated by micromilling [22].

Microinjection molding is another widely used microfluidic fabrication technique. The manufacturing steps of this technique are as follows. A thermoplastic polymer is melted and injected under high pressure into a mold. Then mold is rapidly cooled below solidification temperature of the polymer. After that the mold is opened and the device is ejected. Cycle time per part is the lowest among all microfluidic fabrication techniques due to efficient thermal cycling. However, the main disadvantage is that it is an expensive process due to the cost of the mold itself. In addition, similar to other techniques, producing separate layers is required for the construction of closed microfluidic devices [23]. Figure 9 illustrates microinjection molding.

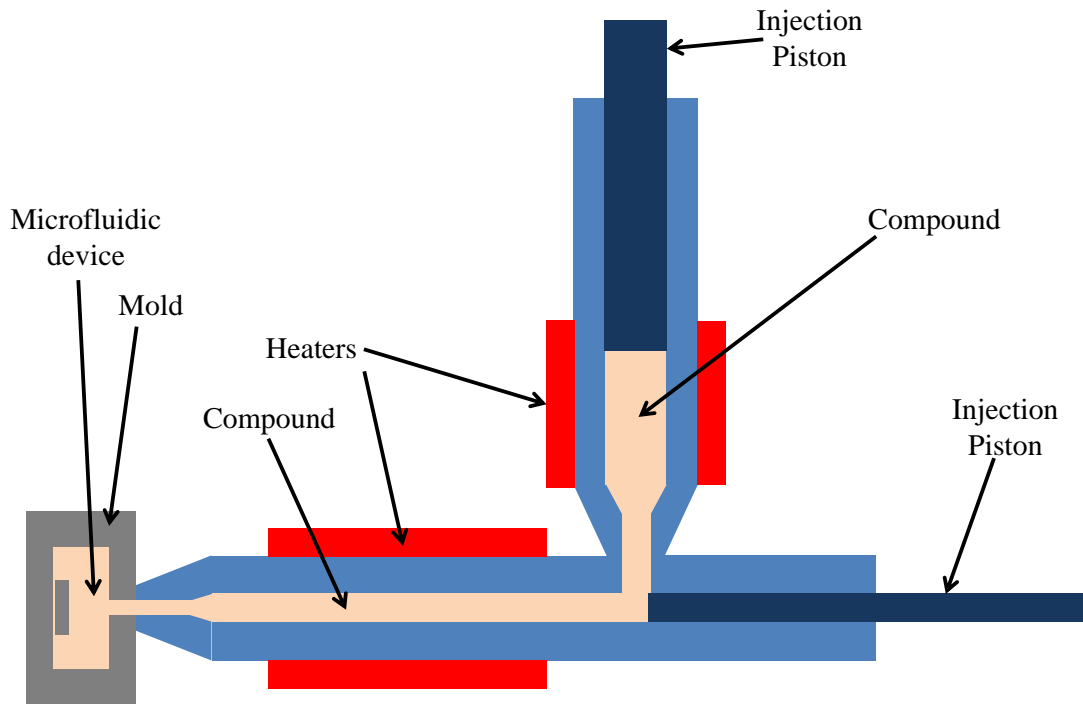


Figure 9 Illustration of microinjection molding.

In all of the aforementioned methods, if there is a complex geometry like a 3D channel, the fabrication will be in multi layers, which must be combined to get the final design. Yet, this process is often costly and takes a long time. In cases of PDMS molding, microinjection molding, and hot embossing, the channel depth is limited. In all of the methods except micromilling, any small change in the microchannel design required production of a new mask or mold, which is costly. In micromilling method, it is difficult to fabricate complex shapes that need multiple tools with different diameters. On the other hand, although the cycle time is not the lowest among the microfluidic fabrication techniques, micromilling still offers an acceptable process time (1 min-1 hr) and it does not require a prior fabrication of mold or mask, which makes it a proper tool for prototyping and low to medium production. Figure 10 presents a table comparing microfluidic fabrication techniques.

2.1.3. The Micromixer Design

In this thesis we considered a micromixer specially designed for micromilled microfluidic systems. In accordance with the micromilling process, we assumed 200 μm diameter end mill. Normally in micromilling, it is not possible to produce cut

features less than the diameter of the mill. However, by using the approach illustrated in Figure 11 it is possible to obtain a throttle of a width less than the tool diameter.

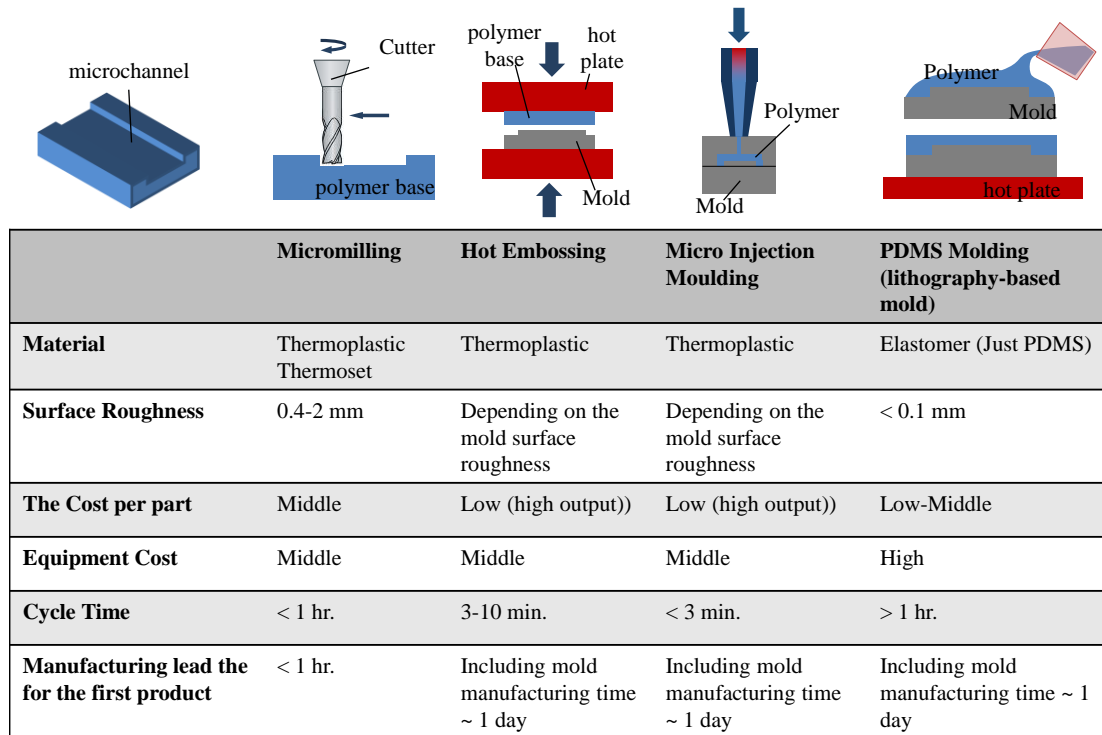
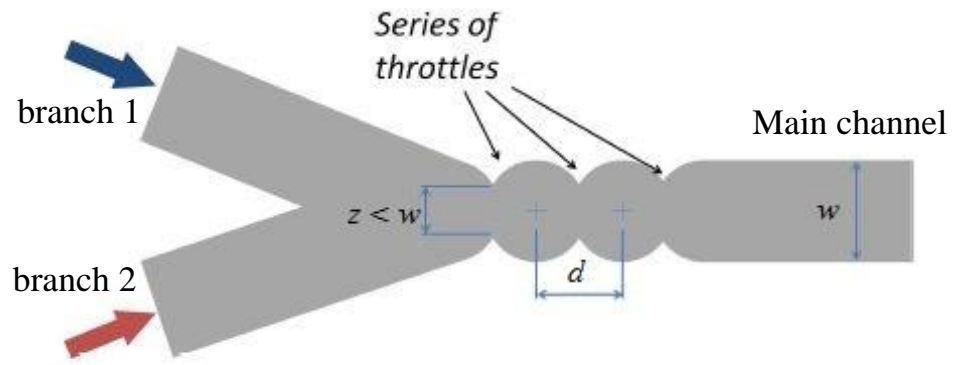


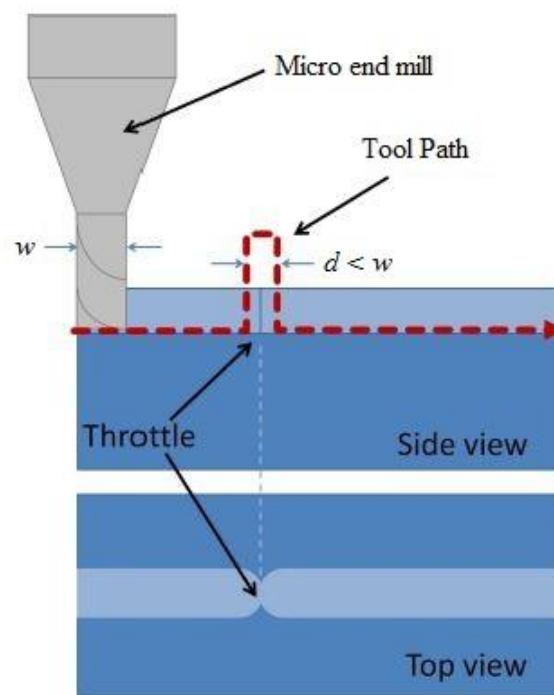
Figure 10 Comparison between microfluidics fabrication processes.

The design is composed of two branches meeting at a main channel of 200 μm width, which involves at least one throttle. Figure 11.a illustrates the design. The fluids in the branch start mixing in main channel, where the mixing is facilitated by decreasing the diffusion length through the use of the throttle.

The effect of the size of the throttle and number of throttles, in mixing process and possible improvement are examined within this thesis. An optimum design is obtained based on the numerical analysis carried out by using COMSOL Multiphysics.



(a)



(b)

Figure 11 (a) The micromixer design, (b) use of micromilling to produce the throttle.

2.2. A Survey on Analysis of Micromixers

First, all parameters that have an effect on mixing performance should be taken into consideration in the design process. These parameters are basically the inlet velocity of fluids and the fluid properties such as concentration of the fluids (c), density (ρ), viscosity (μ), and the diffusivity (D).

Peclet number (Pe) represents the ratio of convective mass transport to the diffusion constant in flow direction

$$Pe = Uw/D \quad (2.2)$$

here U is the flow velocity, w is the characteristic dimension, D is the diffusivity.

It is common to set Reynolds number, Peclet number or both as non-dimensional input parameters and study the effect of them on mixing performance. For instance, Chen et al. took into consideration the effect of the Peclet number in mixing efficiency [11]. The reason of studying the impact of Pe in mixing process is to show the effect of the diffusion and the input velocity in mixing process.

Soleymani et al. [24] showed in their study the effect of Re number as an index of inlet velocity that has an impact in mixing process. Muhammad Virk et al. [25] and Abraham Stroock et al. [26] studied the effect of both Pe and Re on mixing efficiency separately. Shun-Jyh Wu et al. [27] presented the influence of Re in mixing process.

Another factor the designers care of is the geometry of the micromixer such as the angle between the inlet channels or aspect ratio. Soleymani et al. [24] studied the effect of many factors on mixing efficiency. The angle between the inlet channels was among these factors. They found that if the angle is between 90° and 105° , the performance of mixing increases. On the other hand, Muhammad Virk et al. [25] found that the angle has no clear effect on mixing performance. Valery Rudyak et al. [28] also took into consideration the effect of the angle. They found that the change in inlet angle has a slight impact in mixing process.

Another geometry factor is the aspect ratio, which is the ratio of width to height of the mixing channel. Again Soleymani et al. [24], Muhammad Virk et al. [25] and Valery Rudyak et al. [28] studied the influence of aspect ratio on mixing efficiency. They found that decreasing the channel's width improves mixing efficiency. On the other hand, the depth of the mixing channel has no considerable effect in mixing performance.

In addition, complexity of the channel geometries is also investigated. Ali Asgar et al. [29] utilized obstructions with rectangular shape in the main channel (Figure 12) to enhance mixing efficiency. Again, Ali Asgar et al. [30] presented a new micromixer with diamond obstacles to improve the mixing performance by splitting and recombining the fluid streams (Figure 13).

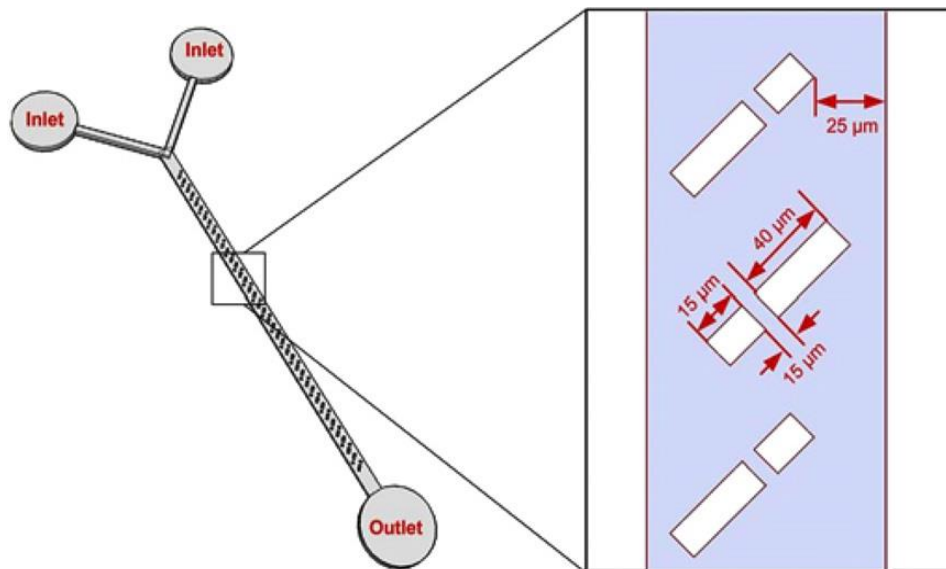


Figure 12 Illustration of a micromixer with rectangular obstructions in mixing channel. The function of these obstructions is to improve the mixing efficiency [29].

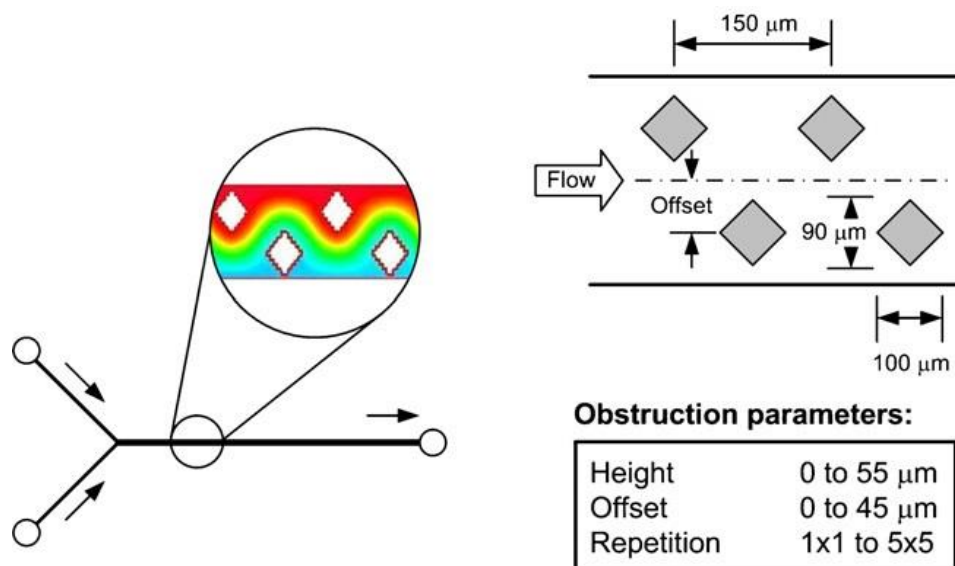


Figure 13 Illustration of a micromixer with diamonds obstacles in mixing channel to enhance the mixing efficiency [30].

Arshad Afzal et al. [31] reported a split and recombine type micromixer (Figure 14).

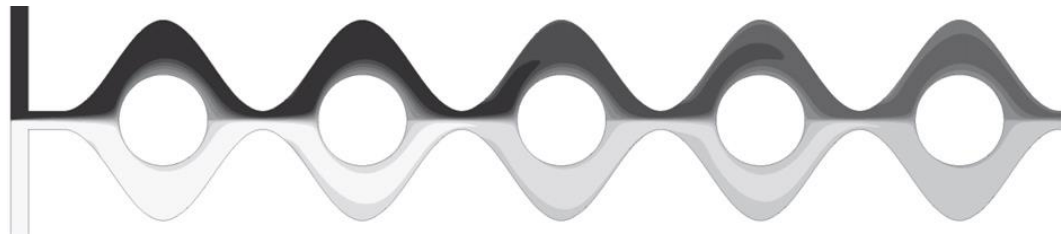
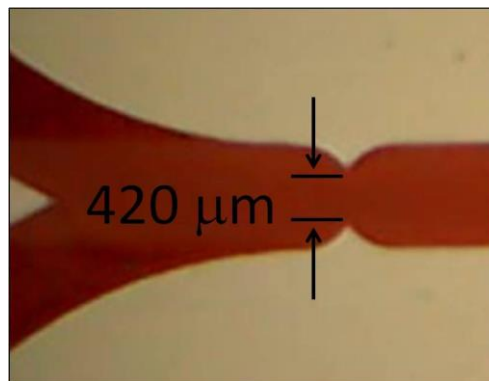
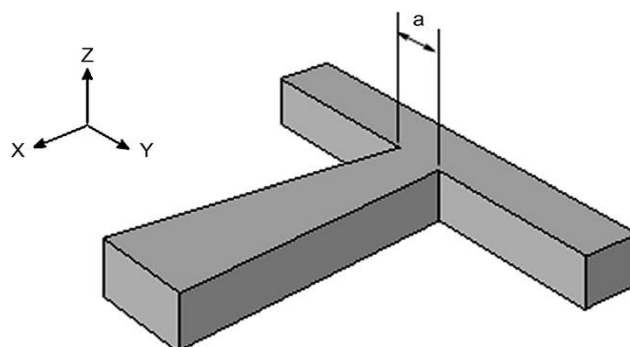


Figure 14 A split and recombine type micromixer [31].

Soleymani et al. [24] and Yıldırım [32] studied mixing by taking effect of throttles in the mixing channel into consideration. They found that adding throttle has a clear impact in the mixing process that develops the mixing efficiency. Moreover, they reported that reducing the throttle width improves the mixing efficiency. Furthermore, Yıldırım [32] tested the effect of adding series of throttles in mixing process. He found that this improves the mixing performance as well.



(a)



(b)

Figure 15 (a) The micromixer presented by Yıldırım [32].

(b) Micromixer proposed by Soleymani et al. [24].

Yong-Jun Ko et al. [15] combined three designs of micromixers. They utilized splitting and recombining by implementing circular obstacles and small channels branching from the main channel. In addition, they added a nozzle at meeting point of branched channels with the main channel (Figure 16). Hence, the chaotic movement of the fluids made the micromixer more efficient. However, a micromixer with a complex shape is difficult to fabricate. Photolithography based methods such as PDMS molding are suitable to fabricate such micromixers regardless of the disadvantages of these methods as discussed earlier.

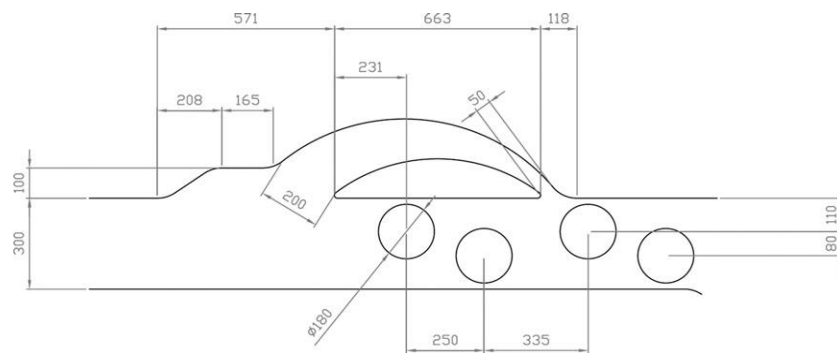


Figure 16 Illustration of the micromixer presented by Yong et al., which combine three types of passive micromixers. These types are intersecting channels, throttles at the end of branched channels and obstacles in mixing channel [15].

C. K. Chung et al. [33] presented a rhombic micromixer with flat angles at the top and a nozzle at the end of the micromixer (Figure 17). They explained that the rhombic shapes of the mixing channels and the nozzle could improve mixing performance.

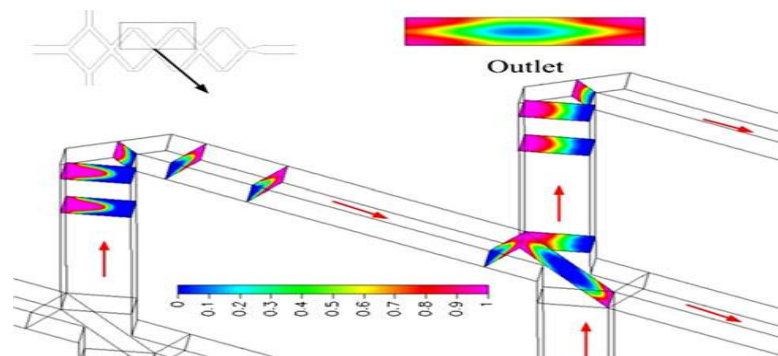


Figure 17 Micromixer with rhombic mixing channels and nozzles at the end of mixing channels [33].

Stroock et al. [26] proposed a herringbone type micromixer which involves grooves at the channel bottom. This micromixer is probably the most famous one as it is commonly used by many researchers [34 – 36]. In this design the grooves distort the streamlines to cause a circulation within the channel that facilitates the mixing.

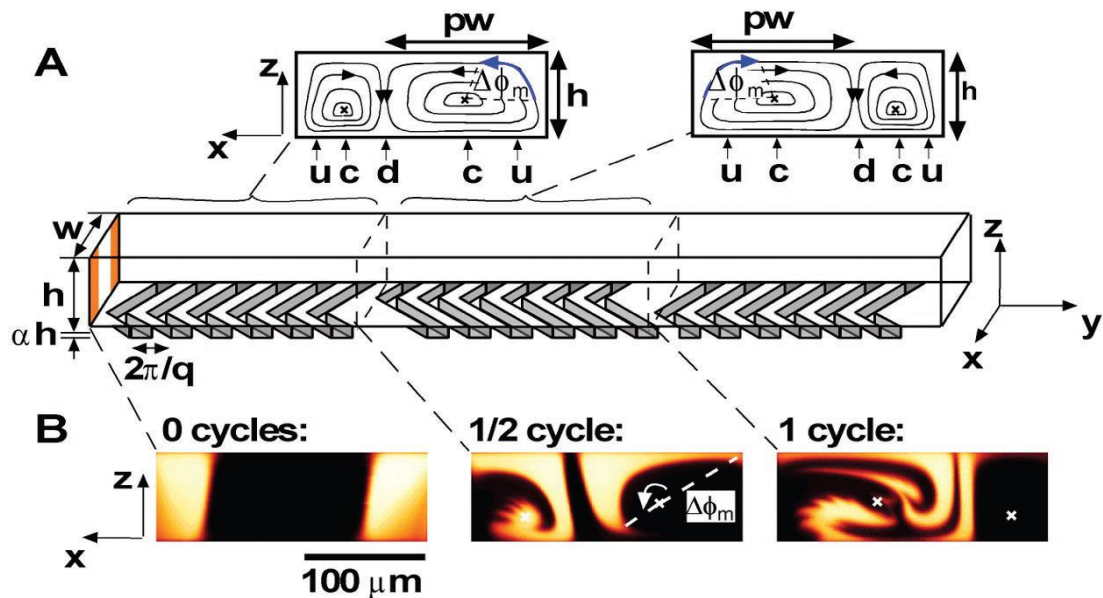


Figure 18 Illustration of the herringbone mixer and its mixing performance [26].

On the other hand, many researchers prefer simple micromixers with extended mixing channels, which increase the interface area. Since the designs are limited by the small size of the microfluidic chip, they commonly utilized serpentine channels. Shakhawat Hossain et al. [37] and Nan Kuo et al. [38] used these serpentine channels to enhance the mixing performance of their micromixers. They tested many shapes of these serpentine channels such as square wave, zigzag and curved to choose the best one that improves the mixing in their micromixers (Figure 19).

2.3. Computational Fluid Dynamics (CFD)

CFD, which can be defined roughly as numerical analysis of flow related problem by using computer algorithm, is widely used to simulate micromixers and analyze their mixing performance [8, 18, 28, 36, 41].

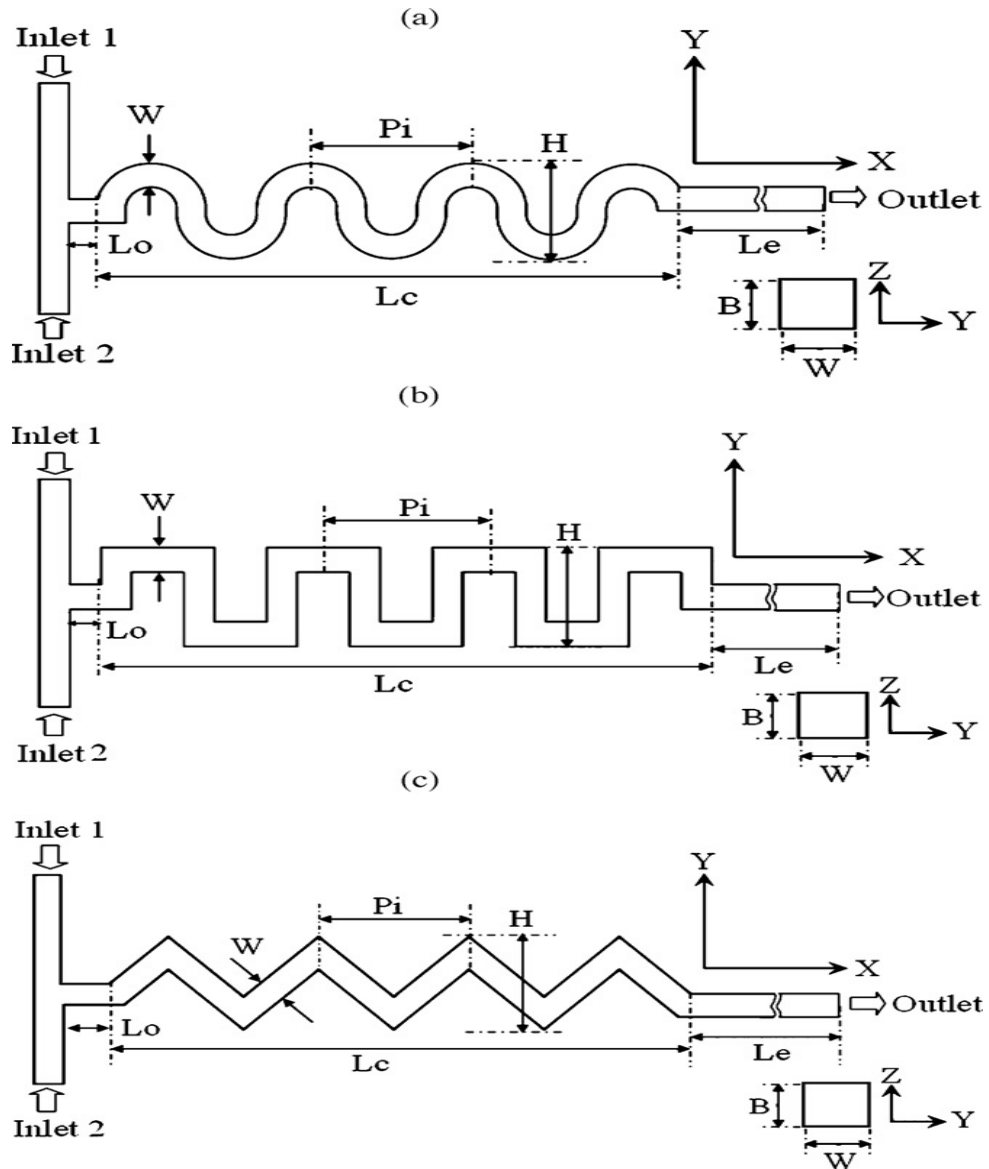


Figure 19 Micromixers with serpentine channels [37].

There are many CFD programs such as ANSYS CFX, ANSYS FLUENT, and COMSOL Multiphysics and others. Hossain et al. [37], Afzal and Kim[31] and other researchers used ANSYS CFX in their studies to simulate the micromixers and calculate the mixing performance. Other designers such as Wu and Lee [27] simulated the micromixers and computed the mixing efficiency in ANSYS FLUENT. While Kuo and Jiang [38] Virk and Hold [25], Yan Du et al. [34] and other designers used COMSOL Multiphysics to design their micromixers and check mixing performance. Hence we preferred COMSOL Multiphysics to analyze the micromixer due to its wide use in microfluidics.

2.4. Description of the Mixing Problem

Firstly, it should be recalled that the flow of the working fluids in the micromixer is always laminar. In addition, we can make the assumption that the working fluids are Newtonian. Additional common assumptions are incompressible flow, smooth channel's walls by neglecting the roughness and manufacturing imperfections, neglecting the surface tension force, no slip boundary, and the continuum [29, 2]. We also neglected transient effects and assumed a steady flow and diffusion.

Since the fluids are Newtonian and the flow is incompressible and laminar, the mixing profile will be analyzed by using finite elements methods to solve 2D Navier-Stokes, or finite volume approximation to solve 3D Navier-Stokes and continuity equations [31].

The first equation is the steady Navier-Stokes equation

$$(\vec{V} \cdot \nabla) \vec{V} = -\frac{1}{\rho} \nabla P + \nu \nabla^2 \vec{V} \quad (2.3)$$

and the second is the continuity equation

$$\nabla \cdot \vec{V} = 0 \quad (2.4)$$

where \vec{V} is the velocity vector, P is the pressure, ρ is the fluid's density, and ν is the kinematic viscosity of the fluid.

The numerical analysis is conducted by solving the steady Navier–Stokes and continuity equations. Since the concentration profile is the main parameter affecting the mixing performance, the third equation to be solved is the diffusion convection equation (Equation 2.5). By this equation, the distribution of the species concentration will be detected.

Steady diffusion- convection equation is given by:

$$\vec{V} \cdot \nabla c = D \nabla^2 c \quad (2.5)$$

here c is the concentration of the species and D is the diffusion coefficient.

Firstly, velocity vector \vec{V} is calculated by solving equations (2.3) and (2.4) considering the boundary conditions and assumptions mentioned before. Then these values are used to determine the concentration profile c by substituting the velocity in diffusion convection equation (Equation 2.5).

The concentration profile is an indicator of mixing performance. Therefore, by using this profile, the efficiency of the micromixer can be detected. Mixing efficiency at node i in a meshed domain is defined as the ratio of the concentration at the i th node to the mean concentration. The designers who have worked on micromixers use different formulas in their numerical calculation to calculate overall mixing efficiency. Kuo and Jiang [38], Virginie Mengeaud et al. [40], and Afzal and Kim [31] utilized:

$$\eta = \left\{ 1 - \sqrt{\frac{1}{N} \sum_{i=1}^N \left(\frac{c_i - \bar{c}}{\bar{c}} \right)^2} \right\} \times 100\% \quad (2.6)$$

where \bar{c} is the average of concentration of the species at inlet channels, which is 0.5 (assuming 100% concentration at one inlet channel and 0 at the other), N is the number of sampling nodes along the cut line, and c_i is the concentration of species at node i . These results of concentration profile (c_i) are acquired from Equation (2.5). Then, they are used in Equation (2.6) to calculate the mixing efficiency of the micromixer.

Afzal and Kim [36] and Shakhawat Hossain et al. [37] used Equations (2.7) and (2.8) to evaluate the efficiency.

$$\sigma = \sqrt{\frac{1}{N} \sum_{i=1}^N (c_i - \bar{c}_m)^2} \quad (2.7)$$

$$\eta = 1 - \sqrt{\frac{\sigma^2}{\sigma_{\max}^2}} \quad (2.8)$$

where σ is the variance of the concentration. Indeed, the Equation (2.6) is compact form of Equations (2.7) and (2.8) for that the similarity is clear between these equations.

Rahim Shamsoddini et al. [41] used Equation (2.9) for time averaged mixing efficiency.

$$\eta = \frac{1}{T} \int_T \sqrt{\frac{1}{N} \sum_{i=1}^N \left(\frac{c_i - c_{mean}}{c_{mean}} \right)^2} dt \quad (2.9)$$

where T , in his case, is the time of the stirring period. Therefore to calculate the mixing efficiency in time dependent problem, the integration of mixing efficiency equation with time is needed. However, we neglected transient effects in our simulation.

2.5. Mesh Sensitivity

The starting point for solving mixing problems is analyzing the geometry. Naturally, CFD divides the mixing domain into small cells, which is called meshing. The first benefit of meshing is that subdividing the geometry into small cells makes it easy to write a set of equations to find the solution. Second, the mesh makes the solution suitable to the physics of the problem that is dealt with. Furthermore, the mesh has a clear impact on rate of convergence and solution accuracy.

There are a lot of factors that should be taken in consideration in mesh quality, for instance, mesh type, elements shape, and mesh density.

2.5.1. Mesh Type

The mesh can be classified into structured and unstructured depending on the arrangement of cells. The differences between these types of meshes are as follows. On one hand is structured type, the cells are arranged in an orderly fashion where i , j and k coordinates are used to locate neighboring cell. Moreover, the grid lines must pass through the entire domain. Actually, this type of mesh cannot be used for complex geometries. On the other hand, the cells of the unstructured mesh are arranged in a disorganized fashion. Indeed, there is no i , j and k indices. Furthermore, there are no restrictions on cell layout. Unstructured type is used for complex geometries. However, in case of unstructured mesh, need for a fast CPU and a large

RAM is emerged in order to solve the problem. Figure 18 explains these types of meshes. Shun-Jyh Wu et al. [27], Muhammad Virk and Arne Hold [25], Shakhawat Hossain et al. [19], Shakhawat Hossain and Kwang-Yong Kim [17] and others used unstructured mesh type in their simulations because of complexity of their micromixers design.

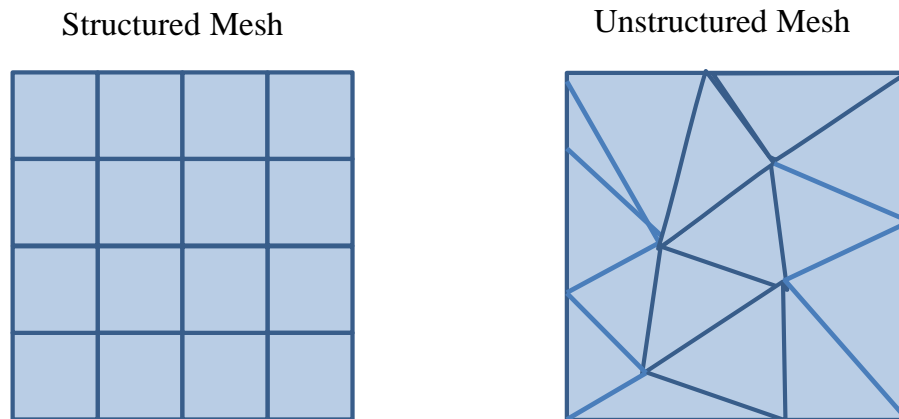


Figure 20 Structured and unstructured mesh types. The figures show the arrangement of elements in each type. The organized fashion for structure mesh and disorganized fashion for unstructured one are clearly seen.

2.5.2. Shapes of Elements

The geometry can be divided into cells with different shapes. The choice of the suitable shape depends on the problem and the solving capabilities.

In 2D simulation, the geometry can be divided into only two types of elements. These cells are in triangular (tri) or quadrilateral (quad). Figure 21 shows these types.

In 3D geometries, body can be divided into four shapes: tetrahedron (tet), pyramid, prism with quadrilateral base (hexahedron) (hex) and prism with triangular base (wedge) (Figure 22).

Hai Le The et al. [18] and Yan Du et al. [34] presented a 3D micromixer and used combination of triangular and tetrahedral cells. Also, Muhammad and Arne Hold [25] used triangular mesh in their simulation. Hossain and Kim [17], Afzal and Kim

[31], again Afzal and Kim [36], Shakhawat Hossain et al. [19] and others used tetrahedral mesh in their designs. Hexahedral mesh was used in [35, 37].

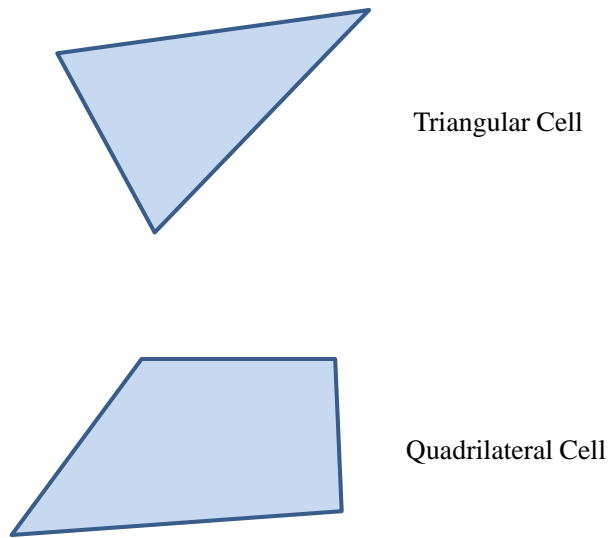


Figure 21 Shape of the cells in case of 2D geometry.

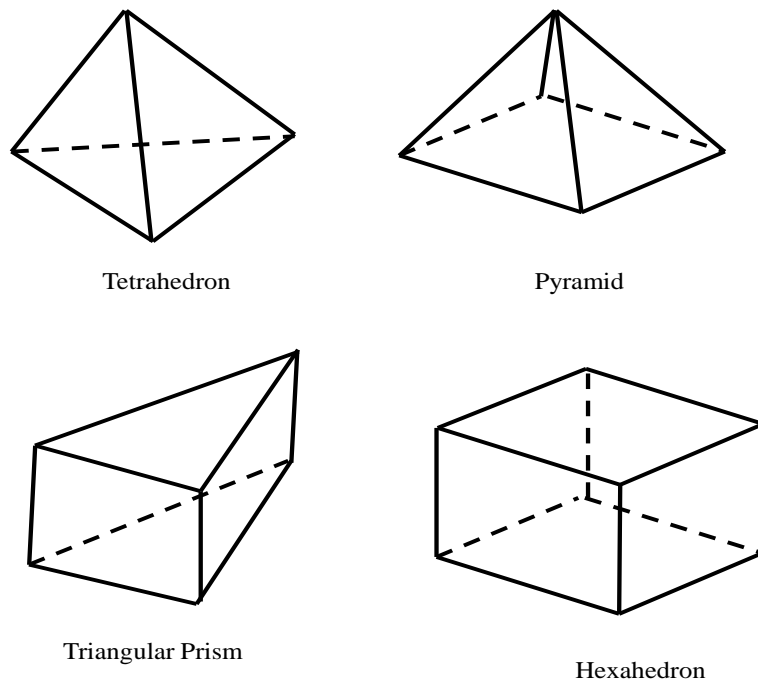


Figure 22 Shape of the cells in case of 3D geometry.

In general, the quadrilateral, in 2D mesh, and brick mesh (hexahedron and triangular prism) present higher order elements. Normally, solving the problems in higher order

elements gives better results. Nevertheless, there are two exceptions, which are the problems including chemical species transport and flowing fluid field. Since the governing equations in those kinds of problems are convection dominated, it is recommended to use first-order elements, namely triangular or tetrahedral cells [42].

2.5.3. Mesh Density

The most important factor that affects the mesh quality is the mesh density. Therefore, during the simulations mesh density is continually increased and convergence of the solution is checked to ensure the accuracy of the solution [29]. Examining and modifying the quality of the mesh is necessary to obtain accurate simulations for the micromixer [37]. A good quality of mesh is required to decrease the numerical errors in calculation of the mixing efficiency.

Afzal and Kim [36] presented mesh sensitivity test conducted for the mixing index along the channel in order to decide which mesh is the more suitable for micromixer problem. They tested five different meshes in their study to find the mixing efficiency in each mesh. The smallest difference in the mixing index between any two meshes indicates an accurate solution. Again, Afzal and Kim [31] presented a grid sensitivity test to determine the optimum grid for their micromixer by way of using five different grid systems. They found out that any mesh giving an error of 1% or less in the mixing index is an acceptable mesh. Hai Le The et al. [18] said that to obtain high accuracy of mixing performance between the two working fluids, a mesh sensitivity test must be done before running the simulation for the micromixer. In mesh sensitivity test, they implemented four different sizes of meshes and calculated the mixing efficiency in each case. Then, they compared the results and found very small difference between the case of finer mesh and the case of extra fine mesh indicating that the results of mixing profile, starting from the case of finer mesh, is independent from mesh size. The mesh must be suitably refined to be sure that the simulation results are independent from mesh size. Thus, the simulation can be achieved within a reasonable computational time, minimum numerical iteration, and the more accurate results.

CHAPTER 3

A PASSIVE MICROMIXER DESIGN FOR MICROMILLED MICROCHANNELS

Here we investigate the micromixer presented in Chapter 1, which utilizes throttles at the beginning of the mixing channel. The purpose of the throttle is to squeeze the streams on each side such as to reduce the diffusion length. The micromixer is designed specially for micromilled microfluidics devices as specified before.

In this chapter the simulation of this specific micromixer with throttles is presented to examine the effect of throttle size, and the number of throttles, and hence to optimize the micromixer.

3.1. Input Parameters for the Micromixer

First of all, in examining the micromixer in Figure 23, the input parameters should be determined. These parameters are summarized as follows.

1. Since the micromixer is fabricated by micromilling process, the width of channels is controlled by the diameter of the end mill. The micromixer is optimized assuming 200 μm diameter end mill. Therefore, the width of the channels is 200 μm .
2. Concentration of the fluids at the inlet (c), the density of fluids (ρ), the viscosity (μ), and, diffusivity (D) also effect the mixing. Furthermore, velocities at the inlet have an important effect in mixing process. We defined constant and equal flow velocities at the inlets during simulation of the presented micromixer. In determining the values we referred to the velocities in microfluidics ranging between 10^{-4} - 10^{-1} m/s [12, 26].

3. The size of throttle should also be taken into consideration. Throttle size is described by center-to-center distance (d) and the width of the channel (w), which is $200\ \mu\text{m}$ (Figure 23).
4. The number of throttles also acts on the mixing performance. The effect of adding multiple throttles in series is examined in this study.
5. In microfluidics often aqueous solutions with biological materials (cells, protein, etc.) are used. For these cases, the diffusivity is often in scale of $1 \times 10^{-9}\ \text{m}^2/\text{s}$ [43].

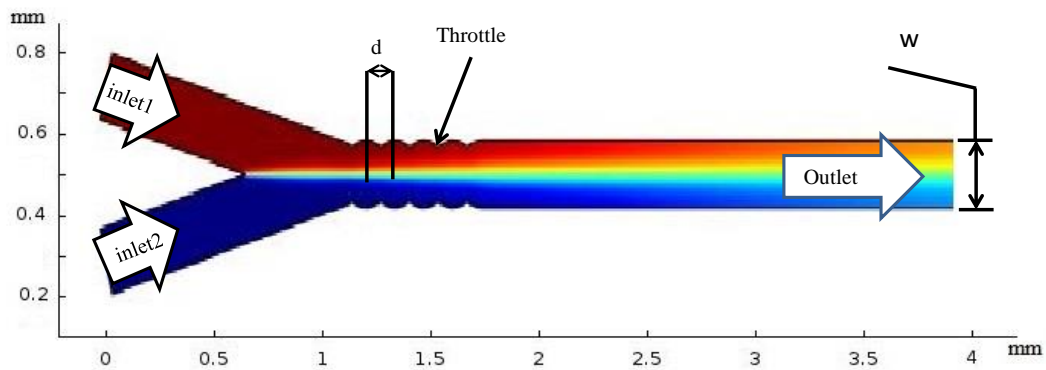


Figure 23 Clarification of the proposed micromixer.

The effect of the channel depth is neglected and the micromixer is simulated in 2D, since depth has no considerable effect in mixing performance [12, 26, 32].

As it is clear, there are many parameters, which effect on mixing process. The simulation process for the presented micromixer with such number of parameters is not feasible. Therefore, reducing the number of the parameters is required. This is done by carrying out a dimensional analysis, utilizing the Buckingham Pi Theorem.

3.2. Dimensional Analysis

The benefit of dimensional analysis is creating non-dimensional parameters, which include more than one parameter to reduce the number of initial parameters. The steps followed in carrying out the dimensional analysis are explained below.

First step is to set the matrix of all input parameters with their respective dimensions and units [44] (Table 3.1).

Table 3.1 Setting input parameters with their units and dimensions.

Parameter	Unit	Dimension
Width (w)	m	L
Velocity (U)	m/s	L/T
Density (ρ)	kg/m ³	M/L ³
Viscosity (μ)	Pa.s	M/LT
Center-to-center distance(d)	m	L
Diffusion constant (D)	m ² /s	L ² /T

The second step is to choose some parameters as repeated factors. In this study the parameters (ρ , U , and w) are chosen as repeated factors.

The third step is to write a new equation that includes these repeated parameters and one extra as follows.

$$\Pi'_1 = D \rho^a U^b w^c \quad (3.1)$$

The fourth step is to substitute the parameters with their respective dimensions in Equation (3.1).

$$\Pi'_1 = (L^2 / T) (M / LT)^a (L / T)^b (L)^c \quad (3.2)$$

The fifth step is making calculations to find the values of the exponents a , b , and c . Then equation (3.1) can be rewritten as

$$\Pi'_1 = D / Uw \quad (3.3)$$

Recall that $Pe = Uw/D$, therefore

$$\Pi'_1 = 1 / Pe \quad (3.4)$$

$$\Pi_1 = Pe \quad (3.5)$$

From Equation (3.5) Pe comes out to be the first non-dimensional parameter.

Similarly, other non-dimensional parameters can be found as:

$$\Pi_2 = Re \quad (3.6)$$

$$\Pi_3 = d/w \quad (3.7)$$

$$\Pi_4 = N \quad (3.8)$$

The new non-dimensional input parameters are (Pe , Re , d/w , and N). Hence, mixing efficiency depends on these four parameters

$$\eta = \phi(Pe, Re, d/w, N) \quad (3.9)$$

Note that here Re and Pe are interdependent. Pe depends on the diffusion constant, inlet velocity and channel width, while Re includes density, viscosity, velocity, and channel width. To resolve this interdependency the effect of Pe on mixing efficiency is ignored in simulation process and only Re is taken into consideration in the simulations of the presented micromixer [24].

$$\eta = \phi(Re, d/w, N) \quad (3.10)$$

3.3. Input Parameters Matrix

The channel width is selected as 200 μm . The throttles are placed at the junction where the inlet channels meet. We chose Re as ranging between 0.1 and 10, which is typical to microfluidics.

The size of the throttle is characterized by the non-dimensional value (d/w). When d/w is smaller than 0.5 the throttle will be too large and there is no throttling. Also, if d/w is equal to or bigger than 0.9 it is difficult to fabricate. Therefore, (d/w) is chosen between (0.5 - 0.8).

Finally, the number of throttle (N) is added in series to enhance mixing. We start the simulations with five throttles to make mesh sensitivity test (Figure 24). After that, a series of 10, 15, and 20 throttles are added to see how that improves mixing efficiency. The matrix of non-dimensional input parameters, which are used in the simulations, are listed in Table 3.2.

Table 3.3 shows the values of the dimensional parameters corresponding to non-dimensional ones listed in Table 3.2.

Table3.2 The set of non-dimensional input parameters levels.

Non-dimensional parameter	Level 1	Level 2	Level 3	Level 4
Re	0.1	1	10	
d/w	0.5	0.6	0.7	0.8
N	5	10	15	20

Table3.3 Values of the dimensional parameters.

Non-dimensional parameter	Dimensional parameter	Level 1	Level 2	Level 3	Level 4
Re	Inlet velocity (m/s)	5×10^{-4}	5×10^{-3}	5×10^{-2}	
d/w	d (mm)	0.1	0.12	0.14	0.16

3.4. Simulations

First of all, to check the mixing efficiency a case with two fluids with same viscosity, density, and velocity flowing through the inlet channels is considered. The concentration of arbitrary species in the fluids was assumed to be 0 and 1 mol/m³ at the inlets 1 and 2 respectively. The simulation is done using COMSOL Multiphysics. It was assumed that the velocity profile is uniform at the inlets. The pressure at the exit is set to atmospheric pressure (0 gauges). No-slip boundary condition was assumed at the sidewalls. The input parameter and the boundary conditions for the initial simulation with five throttles are shown in Figure 24.

The solution of mixing profile is done in two steps. The first step is solving the continuity and Navier-Stokes equations by finite elements method to obtain the velocity profile along the mixing channel. Following equations are solved for incompressible laminar flow. We assumed a steady flow and assumed that the temperature is constant at all boundaries. This assumption is based on the good thermal isolation properties of polymers commonly used in microfluidics.

$$(\vec{V} \cdot \nabla) \vec{V} = -\frac{1}{\rho} \nabla P + \nu \nabla^2 \vec{V} \quad (3.11)$$

$$\nabla \cdot \vec{V} = 0 \quad (3.12)$$

The second step is using the velocity profile obtained in first step in the convection-diffusion equation (Equation 3.13) to solve the concentration profile.

$$\vec{V} \cdot \nabla c_i = D \nabla^2 c_i \quad (3.13)$$

Equation (3.13) gives the concentration distribution along the mixing channel. After that, by using the efficiency equation, which is rewritten below in Equation 3.14 for convenience, mixing efficiency at specified cross-section after the throttles is calculated.

$$\eta = \left\{ 1 - \sqrt{\frac{1}{N} \sum_{i=1}^N \left(\frac{c_i - \bar{c}}{\bar{c}} \right)^2} \right\} \times 100\% \quad (3.14)$$

3.5. Mesh Sensitivity Test

An important step in micromixer simulation is mesh sensitivity test. Mesh sensitivity test is done to ensure that the solution is independent from the mesh size. Hence, it ensures an accurate solution for the mixing efficiency.

In mesh sensitivity test, initial micromixer (Figure 24) (200 μm channel width, five throttles, $d/w = 0.6$, and $\text{Re} = 0.1$) is simulated. The mesh is unstructured triangular mesh because that type is reported to be convenient for problems, which involve chemical species transport and fluid flow [42].

We started simulation with an extra coarse triangular mesh yielding 436 cells (Figure 25). After that, the mesh is refined gradually and at each refinement level mixing efficiency is calculated. Figure 26 shows the efficiency with respect to number of elements at each refinement level.

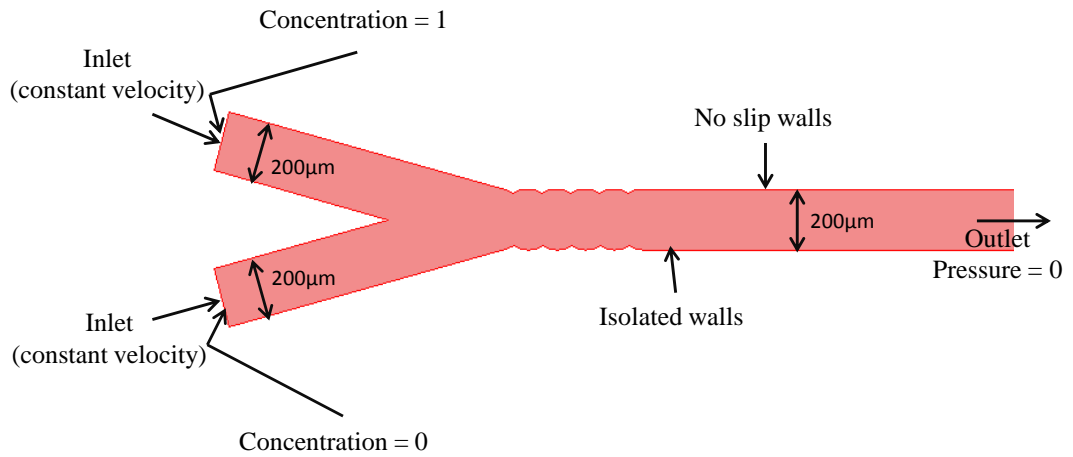


Figure 24 Boundary conditions. The velocities at the inlets are the same and equal to 5×10^{-4} m/s. Mixing efficiency is calculated at the cross-section at 3.5 mm distance from the junction in downstream direction.

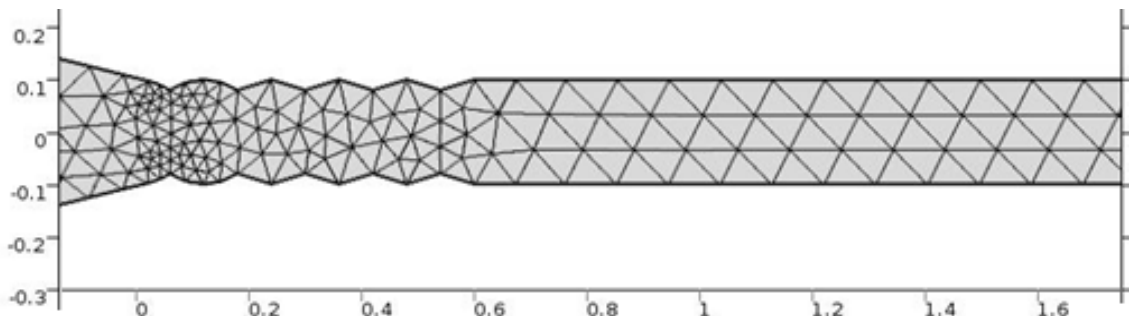


Figure 25 Extra coarse mesh with 436 cells.

Researchers who analyzed micromixers using CFD, commonly carry out mesh sensitivity test by checking the convergence of the efficiency [19, 30, 32, 37, 38]. Accordingly we analyzed the change in efficiency with respect to the number of elements (Figure 26).

As it is clear in Figure 26, refining the mesh results in convenience of the efficiency. Figure 26 also shows that for extra fine meshes (generally roughly 25000 or more cells), the solution is independent of the mesh. Therefore the simulations were done at extra fine refinement level in this study.

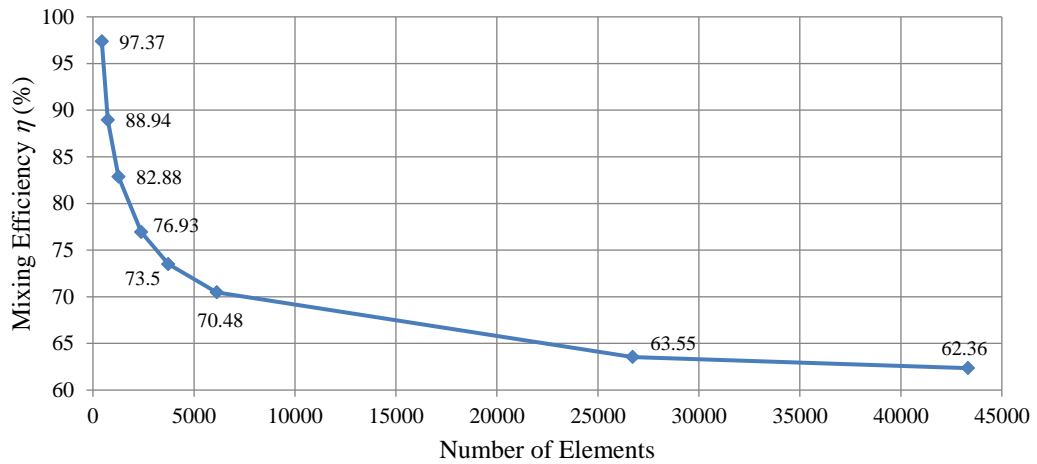


Figure 26 Mixing efficiency with respect to number of cells. It is clear that the solution converges after 26724 cells, which means dividing the micromixer to cells equal to or more than roughly 25000 cells, the solution become independent from the mesh size.

Figure 27 shows the concentration profiles in the micromixer at the same position on mixing channel (3.5 mm) for different number of elements. Note that the profiles are almost the same for 26724 and 43324 elements as expected (Figure 27).

Consequently through the mesh sensitivity test, we concluded that the appropriate mesh size for the simulation is 26724 cells or more. This means that the suitable mesh is unstructured, triangular, and extra fine (Figure 28).

3.6. Effect of Re on Mixing Efficiency

First of all, as it was mentioned previously that the levels of Re was determined as 0.1, 1, and 10. We tested three levels of Re to see the impact of Re in mixing efficiency. Figure 29 shows the results. Number of throttles (N) and non-dimensional throttle size (d/w) are set to 5 and 0.6 respectively.

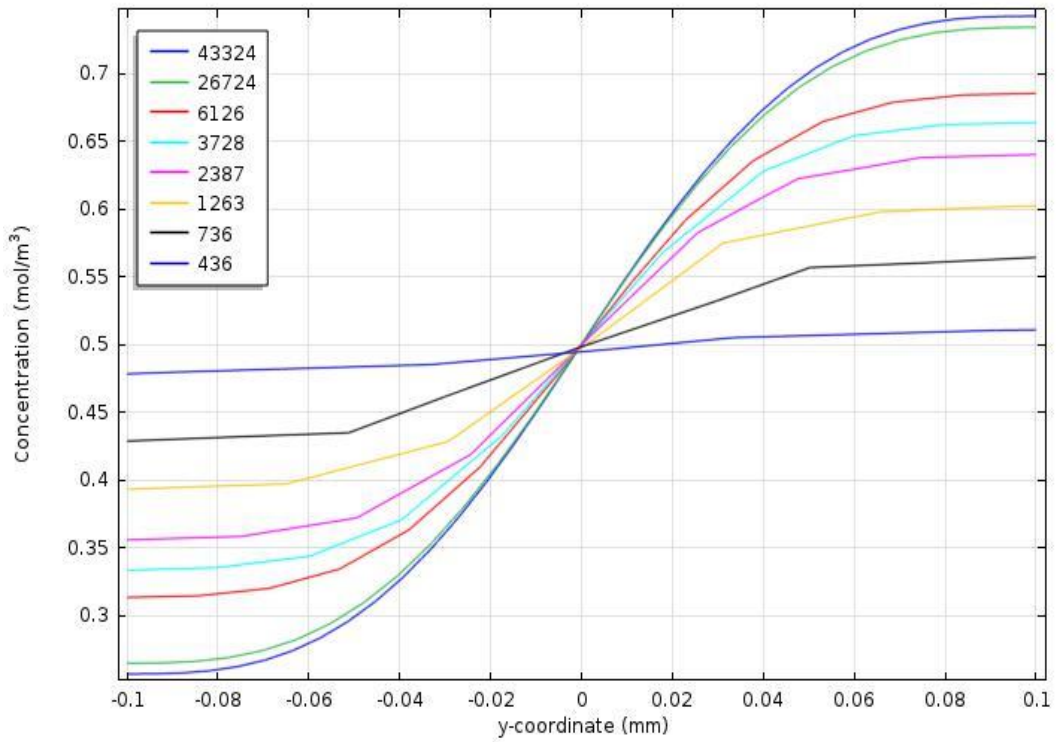


Figure 27 The concentration profile at different mesh refinement levels at the same cross-section (3.5mm).

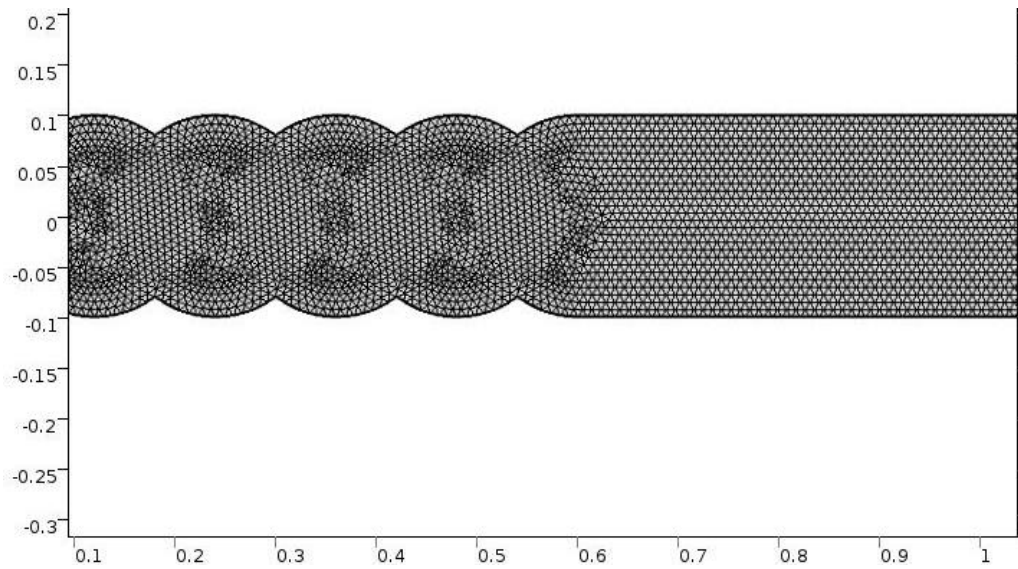


Figure 28 Extra fine mesh that is used in simulations. The figure shows the mesh for the initial design only.

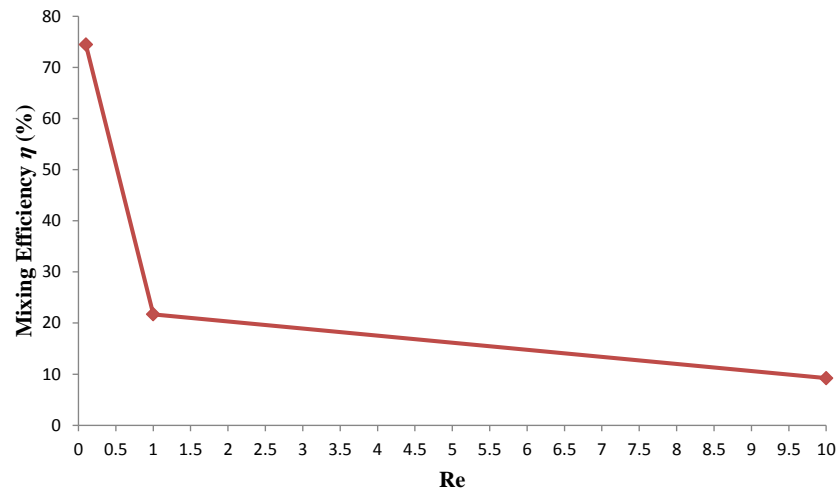


Figure 29 Mixing efficiency various Re. Considerable effect of Re is clearly seen on the plot.

The plot in Figure 29, which explains the relation between Re and the efficiency of the micromixer, shows that mixing process at low Re is more efficient than the process at high Re. Also, when Re increases, the mixing efficiency decreases. The considerable effect of increasing Re is clear from the step change in the efficiency from Re = 0.1 and 1. Figure 30, shows the concentration profile at 3.5 mm from the junction at Re = 0.1, 1 and 10. The results agree with the ones in Figure 29 that there is no significant difference between the profiles for Re =1 and Re = 10.

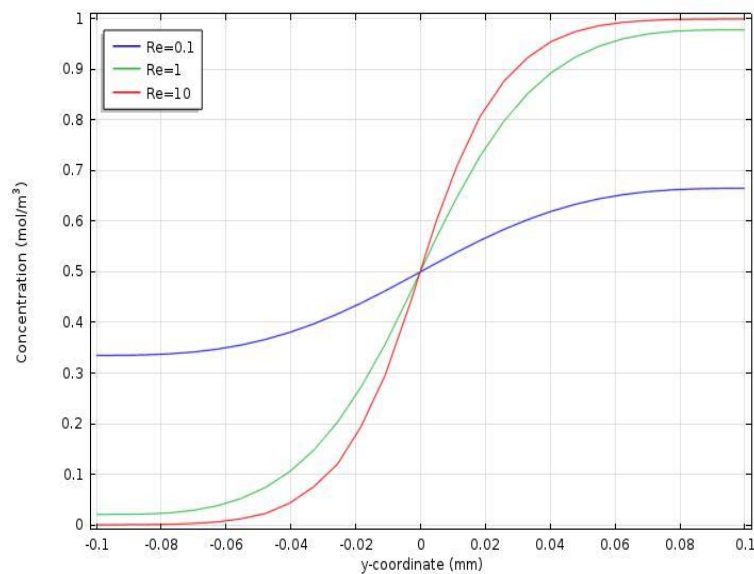


Figure 30 Concentration profile for three levels of Re.

The results shows that the micromixer is suitable for very low Re flow regimes. From this point on we set $Re = 0.1$ in the simulations. Figure 31 summarizes the results of the simulations for three levels of Re.

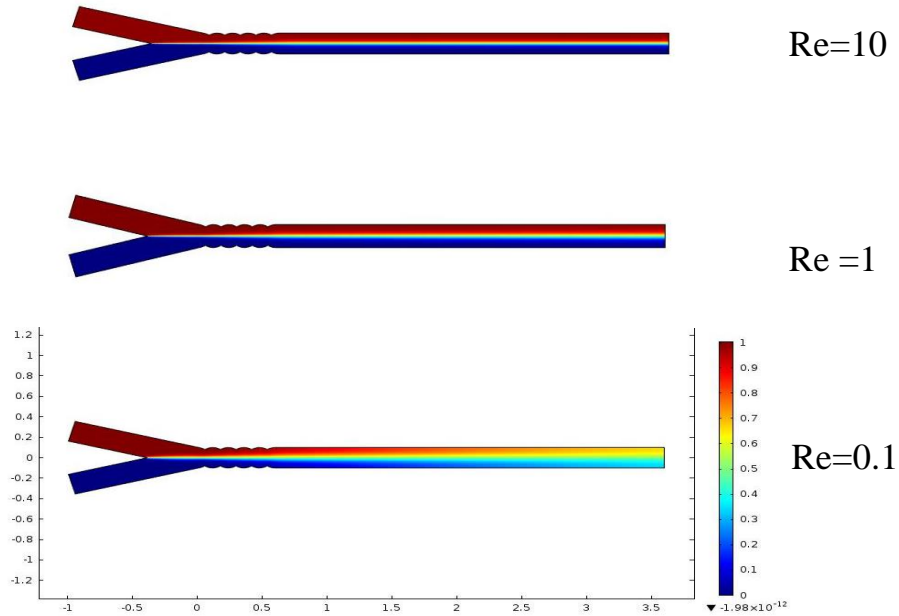


Figure 31 Results of the simulations for three different Re levels.

3.7. Optimization of the Micromixer

For optimization purposes Re is considered to be equal to 0.1. Remaining two factors namely the non-dimensional throttle size (d/w) and the number of throttles are varied between 0.5 - 0.8 and 5 – 20 respectively in 4 discrete levels (Table 3.4).

Table 3.4 Levels of the input parameters.

Parameters	Level 1	Level 2	Level 3	Level 4
d/w	0.5	0.6	0.7	0.8
N	5	10	15	20

As indicated by Table 3.4, total of $4 \times 4 = 16$ simulations were run to sweep all input parameters. Combination of parameter levels yielding the maximum mixing efficiency was chosen as the optimum micromixer design. Table 3.5 shows the

mixing efficiencies for 16 runs. Figure 32 presents a 3D surface plot in accordance with Table 3.5.

Table 3.5 Mixing efficiency for 16 runs.

d/w ratio	No. of throttles			
	5	10	15	20
0.5	73.95%	74.61%	75.12%	75.61%
0.6	74.44%	75.51%	76.39%	77.32%
0.7	75.11%	76.61%	77.98%	79.28%
0.8	76.23%	78.65%	80.84%	82.79%

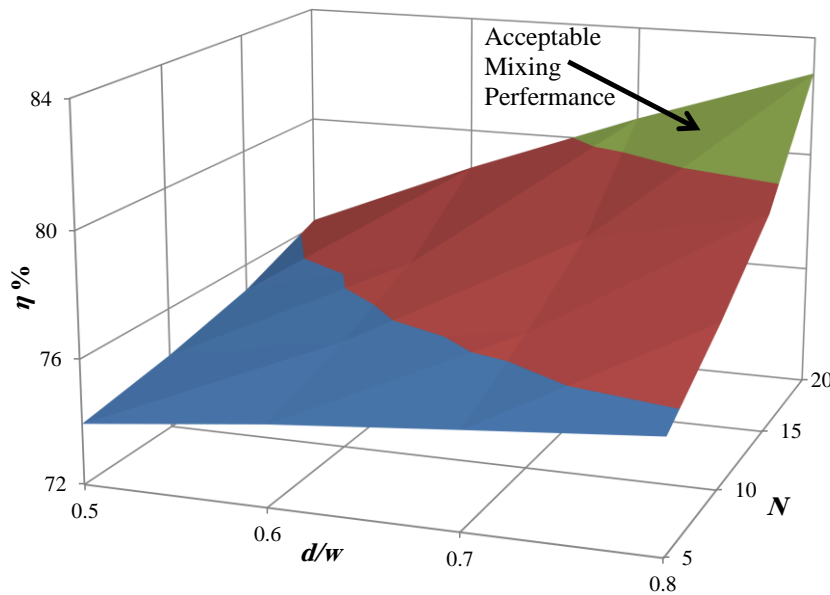


Figure 32 3D surface plot showing the mixing efficiency for the micromixer with respect to d/w and N .

The results show that mixing performance improves if the throttle size decreases and number of throttles increases. It can be seen that the relation between d/w and the efficiency is almost linear. On the other hand, efficiency increases with an exponent of number of throttles. According to the references [10, 46] a micromixer is acceptable if its mixing efficiency is greater than equal to 80%. This acceptable

region is indicated on Figure 32 with the light shaded area. Accordingly, this can be obtained when d/w is 0.8 and N is between 15 and 20.

As a result, the optimum micromixer operates at $Re = 0.1$ with 15 to 20 throttles, where non-dimensional throttle size d/w is 0.8. Increasing the number of throttles improves the mixing efficiency. Figure 33 illustrates the simulation result for the micromixer with $d/w = 0.8$ and $N = 20$.

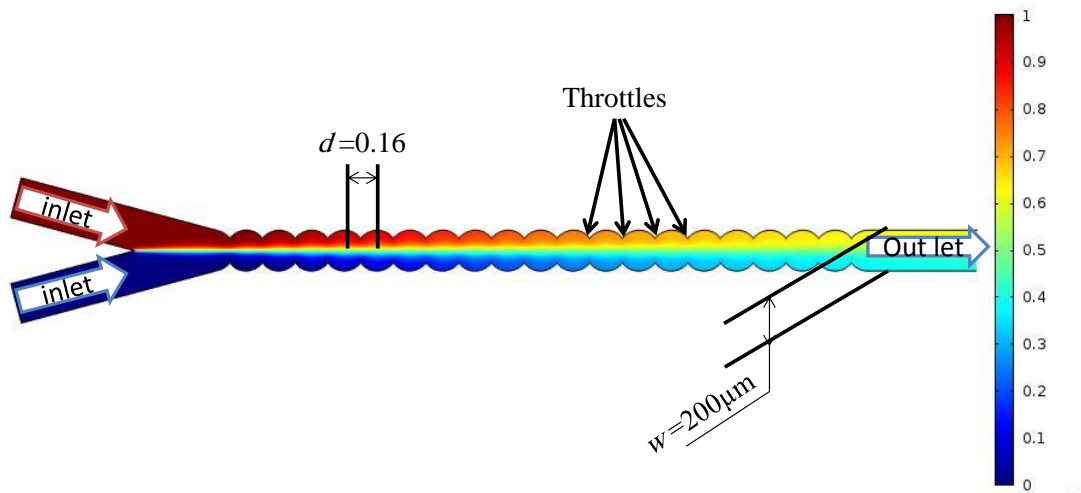


Figure 33 Simulation result for the micromixer with $d/w = 0.8$ and $N = 20$, which yields the highest mixing efficiency. The color scale shows the concentration.

CHAPTER 4

IMPROVEMENTS RELATED TO THE MICROMIXER DESIGN

In this part of the study, we investigated possible ways to improve the mixing efficiency. The previous design was composed of a number of throttles along the mixing channel aligned with channel axis. Here we propose that the mixing efficiency can be improved further by shifting the throttles off the channel axis as indicated in Figure 34.

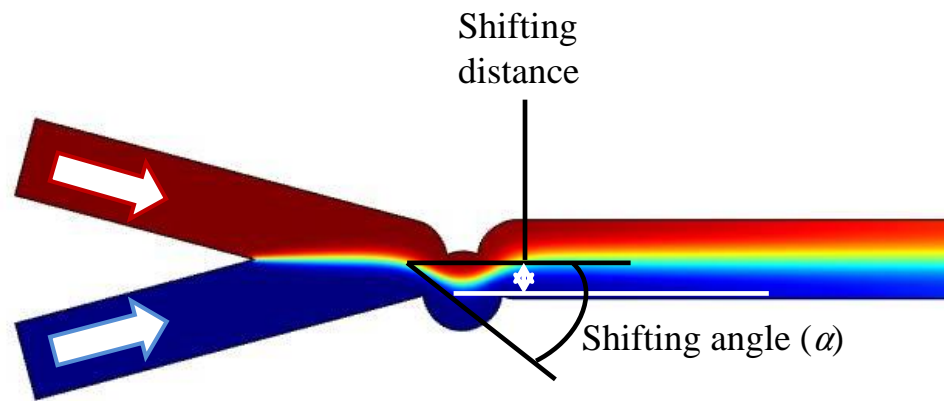


Figure 34 Proposed micromixer design. The throttles are shifted off the mixing channel axis by a shifting angle (α).

4.1. Simulation of the Proposed Micromixer

For the previous design it was found that an acceptable mixing could be achieved when $d/w = 0.8$ and $N = 15 - 20$. Here while keeping these parameters, we tested the effect of shifting angle on mixing efficiency. For this purpose we defined 3 levels of shifting angles: 30° , 45° , and 60° . Hence, we obtained a simulation table as presented in Table 4.1. In the simulation we kept $d/w = 0.8$ and varied N as 15 and 20. During the simulation we set the mesh refinement level to extra fine in accordance with the results in Chapter 3.

Table 4.1 Input parameters for simulation of the proposed micromixer.

d/w is kept as 0.8.

Number of Throttles (N)	15	20	
Shifting angle (α)	30°	45°	60°

The results of the simulations are plotted as a 3D surface as shown in Figure 35.

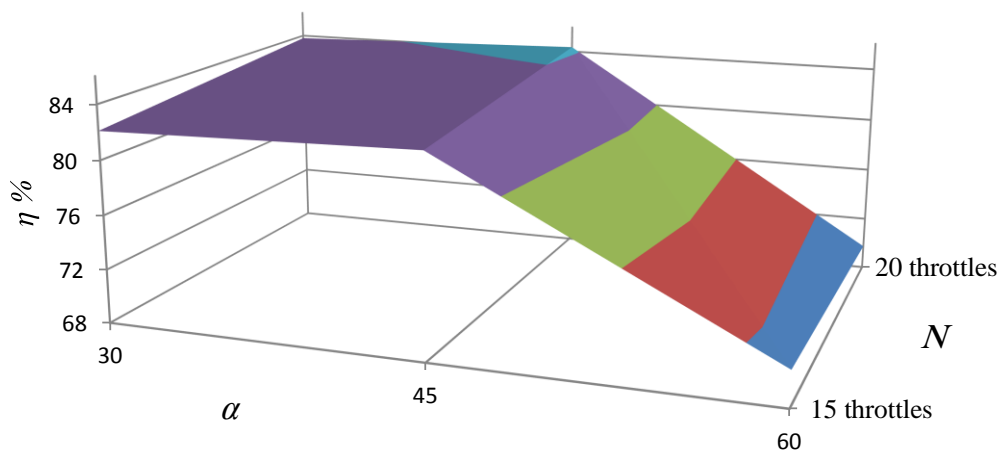


Figure 35 3D surface plot showing the change in mixing efficiency with respect to shifting angle (α) and the number of throttles (N).

The results show that a non-linear relation exists between the mixing efficiency and the shifting angle. A relatively small shifting angle causes a slight increase in the mixing efficiency. Mixing efficiency becomes maximum when the shifting angle is 45°. However increasing the shifting angle beyond 45° decreases the mixing efficiency hence the mixing performance starts to decrease.

Figure 36 shows the simulation results for the improved micromixer with $d/w = 0.8$, $N = 20$, and $\alpha = 45^\circ$ ($Re = 0.1$).

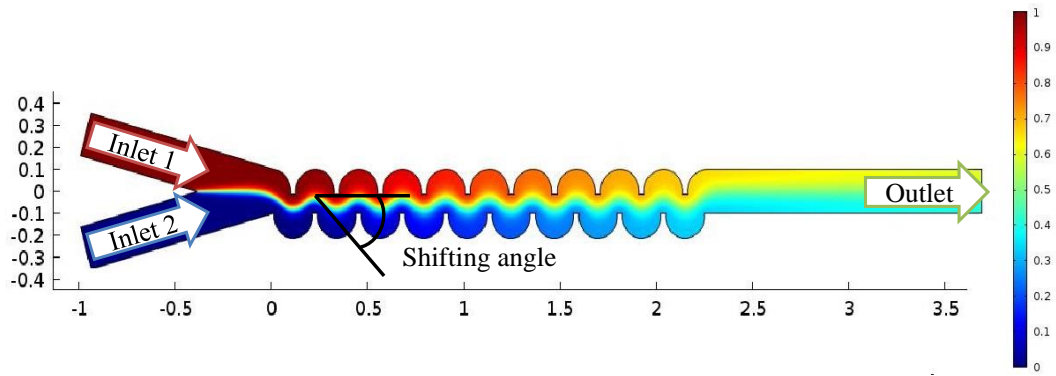


Figure 36 Improved micromixer design. $N = 20$, $d/w = 0.8$, $\alpha = 45^\circ$ and $Re = 0.1$.

CHAPTER 5

CONCLUSION

The major focus of this thesis is designing a simple micromixer with an acceptable efficiency, greater than or equal to 80% [10, 46]. The micromixer designed in this study is a passive micromixer. This kind of micromixers does not need external energy source to operate. Considering the possible fabrication methods, we decided to design a passive micromixer for precision milled (micromilled) microfluidics.

The initial design was composed of two inlet channels merging at the junction and a mixing channel, on which a series of throttles were placed. To optimize this design we utilized CFD and implemented the model by using COMSOL Multiphysics. Before implementing the model a dimensional analysis was carried out to find non-dimensional parameters affecting the mixing performance as Reynolds number (Re), Peclet number (Pe), non-dimensional throttle size (d/w), (where d is the center-to-center distance and w is the channel width), and the number of throttles (N).

The results showed that the micromixer is suitable for very low Re flows ($Re = 0.1$). Hence, we optimized the micromixer for this flow regime. The simulations yielded that we could obtain an acceptable micromixer when $N = 15$ to 20 and $d/w = 0.8$. For the micromixer with 20 throttles, the mixing efficiency goes up to 82.79% .

After optimizing the design, we proposed an improvement for the micromixer. We proposed that shifting consecutive throttles off the channel axis could improve the mixing performance. By again utilizing CFD we found that a 45° angle shift slightly increases the mixing efficiency to 84.36% . We presume that this is caused by a little chaos induced in flow. However this presumption is needed to be verified through proper testing, which we leave it as a future work.

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