CHAPTER 1
INTRODUCTION

This chapter introduces different actuation mechanisms for microscale liquid flows, with particular emphasis on compact disk based microfluidic systems. Different operations and applications are extensively reviewed in context of microfluidic transport on rotational platforms. Based on the important conclusions drawn from literature review, the aim of the thesis is defined. Finally, outline of the thesis is delineated at the end of this chapter.

1.1 MINIATURISATION AND MICROFLUIDICS

Abilities of manipulating structures and patterns over small scales have motivated a wide range of scientific innovations, resulting in the development of miniaturized fluidic devices and systems (often referred to as lab on a chip) for handling small volumes of fluids with ultra-tunable precision. The underlying applications are many-fold because of their inherent advantages such as high transfer coefficients (on account of large surface area to volume ratios), low sample volume consumption, portability, versatility for rapidly performing complex syntheses, measurements and analysis, and addressability of biophysically-compatible length scales. The emergence of microfluidics, primarily triggered by these phenomenal advancements in generating small-scale geometrical features through micro- and nano-fabrication technologies, has simultaneously rekindled the interests in several classical areas of fluid dynamics, including creeping flows, with a fusion and agglomeration of concepts from molecular physics, surface chemistry and life sciences. Traditionally, silicon micromachining methods have been used to fabricate microfluidic channels from silicon and glass. Of late, other types of materials such as Polydimethyl siloxane (PDMS) and Polymethyl methacrylate (PMMA) have been successfully employed for generating microfluidic structures because of their advantages in terms of faster design times, low cost, the ability to seamlessly impregnate micro/nanoscale features, and the possibilities of obtaining flexibly deformable shapes.

Primary advantage that microfluidics offers is the consumption of small quantities of the sample and reagent for carrying out separation and detection with high resolution, sensitivity, and specificity with reduction in time of analysis and cost, and possibility of developing compact and portable systems that might ease the use of

bio/chemical handling and analysis systems. It also addresses new capabilities in control of concentration in space and time. It has gained its applications in truly diverse fields, ranging from multifarious facets of biotechnology and biomedical engineering, (commonly classified under the broad theme of biomicrofluidics or BioMEMS), biological weapon detection on one side to inkjet printing and thermal management of electronic devices on the other. Developments in many of these applications have often been facilitated by the technological advancements in optical and other detection technologies, thereby enabling the researchers to probe the device functionalities with unprecedented accuracies, precisions and sensitivities (Whitesides, 2006). With the successes of the existing micro-analytical methods, it has become an obvious challenge to develop new, more compact and more versatile sensing, actuating and diagnostic protocols, and to look for expanded applications of microfluidics in areas beyond traditional chemistry and life sciences. Microfluidics, in this respect, has truly emerged as an interdisciplinary science and holds the potential in influencing subject areas from interfacial physics, surface chemistry, chemical synthesis and biological analysis to optics and information technology. Nevertheless, the heart and soul of microfluidics appears to be revolving around the transport phenomena and flow physics in micro and nano scale systems.

1.2 DISTINCTIVE MICROSCALE FEATURES

Is microfluidics a science which only translates the ideas from conventional fluid mechanics? To address this issue, it is imperative to emphasize on some of the key demarcating features of microscale fluid mechanics, as compared to its macroscale counterparts. These distinctive issues are critical, and in many respects determine the functionalities of microfluidic devices to a significant extent, as indicated in our subsequent discussions.

i) As the size of a system is reduced, its surface area to volume ratio increases. If we consider the length scale as L, the surface force scales as L^2 and the volumetric force scales as L^3. Hence, for a micro-device with characteristic length scale $L \sim O(10^{-6}\text{m})$, this ratio turns out to be of the order of 10^6, resulting in the dominance of surface effects over volumetric effects. Thus, over microscopic length scales, inertia forces may often turn out to be negligible in comparison to viscous forces, electrostatics/electrodynamics forces, or surface tension forces. Effects of many of the

surface forces, which are not otherwise felt very prominently over macroscopic scales, may thus play decisive roles towards regulating the functionalities of microfluidic devices.

In order to adjudge the relative importance of different competing forces/interactions in microfluidics, various dimensionless numbers are commonly invoked. These numbers are typically indicatives of the relative strengths of the various forces acting on a particular system. For example, Reynolds number, *Re*, often relates the ratio of the inertial forces to viscous forces. In most cases of liquid microflows, *Re* is typically less than 10, so that viscous forces play significant roles towards dictating the fluid flow characteristics. Further, because of characteristic low values of *Re*, momentum transport is essentially diffusion dominated, which renders efficient mixing in microfluidics an ever-threatening potential challenge, particularly considering the exploitation of turbulence as a prohibitively difficult proposition over reduced length scales. Apart from *Re*, several other dimensionless numbers are also commonly invoked in the context of micro-scale transport, as summarized in Table 1.

Table 1.1: Important dimensionless numbers in microfluidics

Symbol	Name	Ratio
Re	Reynolds Number	Inertial/Viscous
Pe	Peclet Number	Convection/Diffusion
Pr	Prandtl Number	Momentum Diffusivity/thermal diffusivity
Ca	Capillary Number	Viscous/Surface Tension
Bo	Bond Number	Gravity/Surface Tension
Kn	Knudsen Number	Molecular free path/ system length scale

ii) Definition of fluid properties over micron or sub-micron scales is not often free from ambiguities. Such ambiguities stem from the fact that traditional fluid mechanics is normally concerned with the behaviour of matter over dimensions that are significantly larger as compared to the molecular length scales (may be characterized loosely by average intermolecular distances, or more rigorously, by the molecular mean free path). The behaviour of fluids, under such conditions, may be idealized as the same as if the fluid was a continuous medium (continuum description). The local fluid property at a point may then be defined as the average

property of all the molecules occupying a 'sensitive' elemental volume chosen in the neighbourhood of the point under concern (Batchelor, 2000). The sensitive volume should be small enough for the measurement to be local enough; so that further reduction in its size does not change the value of the property. Considering a large elemental volume would invariably include the variations associated with the spatial distribution of the property, as demonstrated in Fig. 1.1, preventing the analyzer in capturing the trends in variations of properties over the system scale.

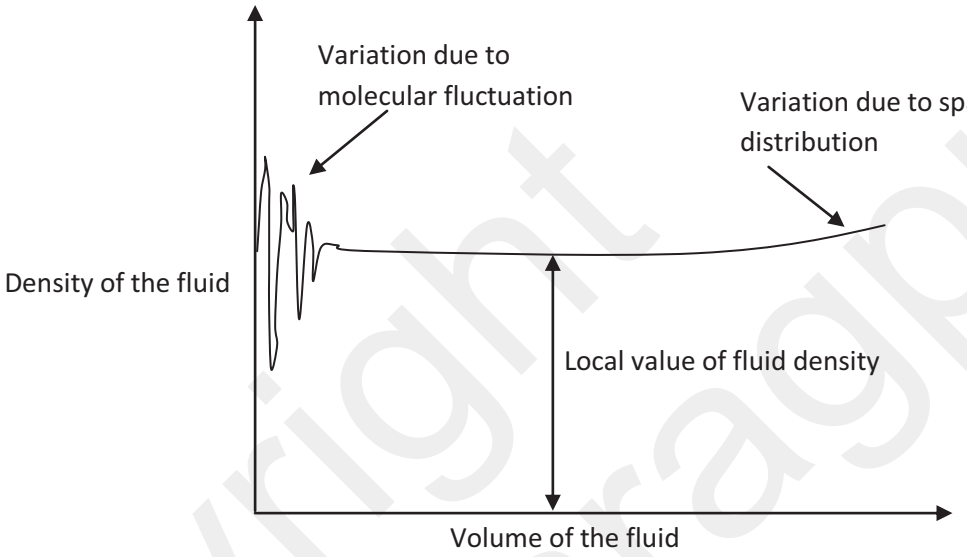

Figure 1.1: Variation of the density of the fluid on the volume considered

On the other hand, if the volume is considered to be too small to contain only a few numbers of molecules, then statistical fluctuations with regard to the relative occupancies of the molecules in the elemental volume may give rise to locally oscillating natures in the predicted fluid properties (see Fig. 1.1). If the system length scale is itself of comparable extent as that of the characteristic length scales of these local oscillations, continuum considerations may not be applicable altogether. Issues of microfluidics concerning the flow behaviour over such regimes are generally addressed by molecular modelling considerations. However, at this stage we would simply reiterate the fundamental consideration that fluid is said to be in a continuum when the measured fluid property is constant for sensitive elemental volumes that are small as compared to the system scale but large as compared to the local scale, and would essentially consider the suitability of this consideration for all our foregoing discussions.

iii) Microfluidic transport is often characterized by sharply demarcating variations in the fluid properties over the interfacial regions as compared to those in the bulk. As the confinement size becomes narrower, interactions between the wall and fluid atoms (typically van der Waals, electrostatic, structuration and solvation forces) tend to play more prominent and decisive roles towards dictating the interfacial phenomena, primarily by giving rise to strong fluctuations in the near-wall number densities. Such strong local density fluctuations may be observed for several reasons, for example due to layering of fluid atoms parallel to the atomic layers adjacent to the solid boundary. On the contrary, there may also be an appreciable depletion of a denser fluid phase close to the walls in preference to a less dense one, under the influences of hydrophobic interactions in narrow confinements. Accordingly, the effective transport properties as well as diffusion coefficients of the fluid may be remarkably different in the near-wall region, as compared to the far stream. In several instances, anomalous transport close to the wall may render the validity of the classical notion or paradigm of no-slip boundary condition (i.e., zero relative velocity between the fluid and the solid at their points of contact) somewhat questionable. The criticality of this issue is immense; without an appropriate boundary condition no accurate estimates of the flow field may become possible, despite the employment of correct governing equations.

1.3 MICROFLOW ACTUATION MECHANISMS

Various flow actuation mechanisms have been discussed in the literature in the context of microfluidic transport. These mechanisms have their own potentials and drawbacks. Here, we briefly describe and critically asses some of the common mechanisms involved in actuating microscale flows.

1.3.1 Pressure Driven Flow

Pressure driven flows are the easiest and common means to drive microscale transport. An important characteristic of the concerned flow profiles is that the axial velocity varies quadratically across the channel height, with the maximum velocity at the centerline. This implies a significant dispersion due to considerable variations between wall-adjacent and centerline velocities. For cases in which the end objective is to transport chemical or biological samples for wall-bounded reactions, the above may be a serious deterring factor, since the sample may have a tendency to be ‘swept’

away along the centerline instead of converging onto wall-bounded reactive sites. Clearly, more uniform velocity profiles would perform better in cases of such specific requirements. The wall shear force (and hence the pressure drop) increases perpetually as the cross-sectional dimensions of the channel are progressively reduced. This, in turn, implies the requirements of huge pumping power in driving the flow for reduced cross-sectional dimensions, and acts as one of the limiting constraints for pressure-driven flow actuation in microfluidic applications. The product of friction factor and Reynolds number is a constant for fully developed laminar flows, from a classical perspective. However, micro-scale flows are commonly featured with surface roughness elements having characteristic length scales not of trivially negligible order as that of the system length scales. Consequently, interesting interfacial phenomena may occur, overruling the satisfaction of the no-slip boundary condition, either in a true sense or in an apparent sense (Chakraborty, 2007a). Under such circumstances, surface roughness dependent frictional characteristics may be observed in microchannels, even for fully developed laminar transport (Chakraborty, 2007c).

1.3.2 Surface Tension Driven Flow

Surface tension driven flows refer to the actuation and control of fluid transport through a manipulation of the surface tension forces. The manipulation may be hydrodynamic, thermal, chemical, electrical or optical in nature. Since surface tension forces scale with the linear dimensions, these become progressively more dominant with reduction of system length scale from macro to micro and further to nano. In a liquid-gas system, for example, molecules in the bulk of the liquid are pulled equally in all directions by the neighboring liquid molecules, resulting in no net force. At the interface, however, the molecules experience a net attractive force by other molecules inside the liquid, since these are not attracted as intensely by the molecules in the gaseous phase located on the other side, because of a denser molecular packing in the liquid phase than in the gas phase and a consequent stronger intermolecular force of attraction offered by the liquid molecules. To maintain interfacial equilibrium, molecules at the interface rearrange themselves to diminish the surface area (in order to minimize the surface energy), and a meniscus is formed in the form of a surface resembling a stretched elastic membrane. The pressure difference on either sides of the meniscus leads to development of net normal force (pressure difference times the projected surface area). This normal force acting on the meniscus

is balanced by the surface tension force in equilibrium, leading to a curved meniscus. Curvature of a meniscus essentially implicates a pressure jump across the same, which can act as a forcing parameter. In several cases, gradients of surface tension, as induced by gradients of temperature, concentration, or electrical voltage, may be utilized to realize differentials in a net driving force to manipulate microscale flows.

1.3.3 Electrokinetic Actuation

Electrical forces may be used to actuate microflows for various lab-on-a-chip based applications, including pumping, mixing, thermal cycling, dispensing and separating. In addition to the fluid mechanical advantages (such as a virtually uniform velocity profile that may be obtained under certain circumstances), electrical actuation and control of microflows exploit several advantageous features of micro- and nano-fabrication technology. In fact, with rapid advancements in miniaturized fabrication technology, integration of micro or nanoscale electrodes in fluidic device has become a simple procedure.

Electrical actuation and control in microfluidics may be achieved by several means. Most of these techniques strongly depend on the phenomenon of Electrical Double Layer (EDL) formation adhering to an electrically charged substrate. Clubbed altogether, fluid flows influenced by EDL effects are also known as electrokinetic flows. One classical example of electrokinetic effect is electroosmosis, in which fluid elements in the diffuse (mobile) part of the EDL are dragged along with the surplus ionic species, by virtue of an axially applied electric field, so that a net fluid flow may be induced. For various types of electrokinetically actuated flows, one may refer to the classical text of Hunter (1989).

1.3.4 Acoustic Streaming

Acoustic streaming originates from the word *acoustics* or actuation based on sound, although, it is a misnomer. The interaction caused by an external high-frequency oscillation driven by sound or any other obstacles present in the fluid flow leads to this phenomenon. Acoustic streaming usually refers to both of these effects. When the fluid experiences a high-frequency oscillation given by an ultrasound source, a progressive wave is established in the air. Due to the attenuation of the wave, a nonzero time averaged Reynolds stress is built in the region close to the sound source, and this stress pushes the fluid in the direction of wave propagation. The

resultant wind is called a quartz wind. Eulerian streaming refers to the flow driven by the time averaged Reynolds stress term. When a fluid within a duct receives a standing wave, a nonzero time average of the Reynolds stress is built inside the duct. Due to the interaction between the air and the duct wall, a steady recirculating flow takes place within the duct. The net effect is that dust or particles accumulate at nodes. This is commonly referred to as Kundt's dust pattern. The Stokes drift flow is purely kinematic and so it is basically different from the Eulerian streaming flow. However, the mass-transport effect given by the Stokes drift flow is not weaker than the Eulerian streaming. For a progressive wave, the Stokes drift flow plays a dominant role (e.g., application of flexural plate waves is pumping and mixing in microfluidics).

1.3.5 Rotationally Actuated Microflows

Microflows may also be actuated by rotational (centrifugal and Coriolis) forces, by spinning a disc containing microfluidic networks. The disc in many ways may resemble the Compact Discs (CD) used for external data storage, and hence this type of flow actuation is also known as CD-based microfluidics (Madou et al. 2006). CD based microfluidics has gained considerable attention owing to its utility in bio-microfluidic analysis. It can act as a relatively inexpensive platform for chemical analysis and biomedical (pathological) diagnostics, exploiting the advantageous features of portability and rapidity of the analytic platform. Its prime advantages lie in handling wide variety of sample types, the ability to gate the flow of liquids, simple rotational motor requirements, economized fabrication methods, large ranges of flow rates attainable, and the possibility of performing simultaneous and identical fluidic operations.

A number of research investigations have been reported in the literature on several distinctive aspects of rotationally-actuated microchannel flows on CD-based platforms, leading to the common consensus that rotational effects induce an artificial gravity to pump the fluid in the radial direction without pulsation. These advantages render the CD an attractive platform for multiple parallel assays, despite the apparent constraint that the rotational force is essentially a volumetric force that scales with the cube of a characteristic length scale (which may otherwise not sound to be so attractive over the micro-domain, as compared to the surface forces that scale more favourably with linear dimensions). There are two important forces which become important because of the system rotation viz. centrifugal force acting in a radially

outward direction and the Coriolis force acting in a transverse direction. Moreover, use of varying rotation speed results in another force due to angular acceleration. In addition, the fluid also experience viscous force which acts to resist its motion. At low rotation speeds, Coriolis force has less contribution over centrifugal force and the flow behaves similar to pressure driven flow. As Coriolis force dominates, a transverse component is introduced into the flow. In multiphase flow, surface tension force also plays a key role in tailoring the fluid flow. All these forces are tuned to achieve desired flow patterns in the microchannels present in a CD-based microdevice. Importantly, forces in the rotating platform are the functions of the rotation speed and the geometry of the channel, which may be altered dynamically with real time monitoring of the system.

CD-based microfluidic platforms have two major components– a polymeric CD and a driving motor. The polymeric CD is fabricated using a lamination technique, which looks very similar to the commercially available data storage disks but contains engraved network of microchannels in it. Different fluidic samples are loaded through the vents into the reservoirs present inside the CD. These are transported to different other reservoirs containing reagents for analysis via the microchannels connecting different reservoirs. Different fluidic operations (like valving, mixing) are also performed in these microchannels. The second major component is the driving motor. The CD is driven by a rotating AC servo motor. The rotor may be programmed at several rotating speeds and tuned to obtain the desired flow characteristics. After the reactions are complete, the readout is taken using a camera to analyse the results of the diagnosis. This uses the forces in rotating platform to drive and manipulate fluid flow.

1.4 RELATIVE ADVANTAGES OF ROTATING PLATFORM OVER OTHER FLOW ACTUATION MECHANISMS

CD-based microfluidic platforms have several advantages over other traditional microfluidic platforms. Rotationally-actuated pumping is insensitive to the physicochemical properties of the fluid like ionic concentration or pH, unlike the case with electrokinetically actuated microflows. It can successfully eliminate the macroscopic to microscopic interconnects thereby implying “contact free” pumping to induce pressure gradient, which becomes an important task in most of the other lab-on-a-chip devices. Hence, this eliminates the use of tubing, sample loading ports, external valves, splitters and/or syringe connectors, centrifuge tube, glass slides to

reduce the cost of different consumables. Although in the limit of constant low rotation speed, the flow behaviour is analogous to the pressure driven flows, but it can be used efficiently to multiplex several operations on a single platform. The flow rates induced in a channel by centrifugal forces depend mostly on the channel dimensions and its relative position and orientation to the center of rotation, which varies from 1 nl/sec to100 μl/sec, a range of volume flow rates cannot be achieved by any other flow actuation mechanism. The drive of the CD is inexpensive and involves the use of an AC motor. These CDs are made from polycarbonate materials which are found to be biocompatible, making it apt for clinical diagnosis. The complete fluidic network can be contained in a single device (which can be disposable), be it a chip that can be embedded in a CD or the whole disk.

1.5 DIFFERENT OPERATIONS WITH CD BASED MICROFLUIDICS

As CD based microfluidics holds the potential of replacing other microfludic platforms, we shall first discuss on the state of the art developments of different fundamental operations on a CD, most of which are already standardised on other platforms.

1.5.1 Valving

Valving is an operation which enables delivering precise volumes of fluid gated to different chambers. CD based platforms may also be cleverly designed to act as valves. Capillary valves are based on the principle of the balance between the surface tension and the centrifugal pressure at a junction (Ducrée *et al.*, 2007; Haeberle and Zengerle, 2007c). Beyond a critical rotational frequency (also known as the bursting frequency), centrifugal force acting on a fluid may overcome resistive surface tension and viscous forces, so that the fluid starts moving, whereas below the threshold limit the fluid is designed to be restricted at its position relative to the CD. This arrangement, thus, acts like a smart centrifugal valve. The capillary valves work by utilising the pressure induced by the centrifugal force $P_\omega = \rho\omega_c^2 \bar{r} \Delta r$ overcoming the capillary pressure given by $P_s = 4a\left(\sigma / D_h\right) + b$ for typical channel valve of hydraulic diameter (D_h) with surface tension coefficient (σ) and b representing the pressure required to wet the chamber past the capillary valve and ω_c as the burst frequency. The calculation of the burst frequency is experimentally verified by Badr

et al. (2002). Hydrophobic valves are designed by balancing the surface tension and centrifugal force to obtain the burst frequency (*f*) as (Madou *et al.*, 2006): $f \geq \left(\gamma \cos\theta / \rho\pi^2 \bar{r} \Delta r D_h\right)^{1/2}$ where θ is the contact angle at the junction of the valve, $\bar{r}$ is the average distance of the liquid element as measured from the centre of the CD and Δr is the radial length of the fluid in microchannel. The surface tension should resist the centrifugal pumping force, which is directed in radial direction and hence, the surface should be rendered hydrophobic with contact angle greater than 90^0.

Applications of the valves based on the balance of forces on the capillary are restricted to liquids, while non-existence of physical barriers for passive valves cannot prevent the passage of gases. In order to have a physical barrier which acts like a sacrificial valve, Park *et al.* (2007a, 2007b) reported the use of Laser Irradiated Ferrowax Microvalves (LIFM). These valves are made of iron oxide nanoparticles dispersed in paraffin wax. Under the exposure of laser diodes, the nanoparticles get heated up acting as heating elements for the wax, causing the wax to melt. Unlike the previous capillary based valves, LIFM is not dependant on the rotation speed or the specific design of the capillary networks.

1.5.2 Volume Metering

Distribution of liquid of specified volumes to different chambers (also known as aliquoting) is an integral operation essential for automation of different steps in multiplexing in a single device. It involves splitting of liquid volumes into several chambers with different sub-volumes subjected to different analyses. Precise manipulation of fluid volume is important in order to avoid cross-contamination between neighbouring chambers. This is achieved using either a two step or one step process. In a one step process, the liquid volume is directly transferred to the final reaction chambers by the overflow of channels connected to the fluid reservoir. As the chambers are filled to a radial distance of the overflow channel, additional fluid is routed to the next chambers and after filling all the chambers, the remaining fluid is directed to the waste chamber. Although this method is easy to implement, the connectivity of the reservoirs may pose a severe problem of cross contamination of reagents. As an alternative, in a two step process, liquid is first metered into sub-volumes into different channels and a valve is present which opens only when the rotation speed increases beyond the burst frequency. This has several advantages with

respect to the one step process - the metering is uninfluenced by the pre-stored reagent, cross contamination is effectively prevented, and as the volume metered is simultaneous in all the channels so that the reactions for the assay start all at a time. This two step metering has been modified using a centrifugal pneumatic valve to enable efficient metering in this platform (Mark *et al.*, 2009; Mark *et al.*, 2011). A disk based metering structure was used to define nanolitre sample volumes at a coefficient of variation below 5% as a part of integrated colorimetric assay (Steigert *et al.*, 2006). A three-dimensional structure was constructed to perform non-contact based addition and distribution to a centrifugal platform (Bouchard *et al.*, 2010). The structure enabled liquid to be transferred directly into a rotating microfluidic platform while simultaneously distributing the liquid into a series of reservoirs with 9% relative standard deviation of the volume.

1.5.3 Mixing

Mixing is one of the essential steps to achieve homogeneous sample pertinent for any biochemical and biomedical applications. In microfluidic applications, mixing is one of the key issues owing to the low Reynolds number in such systems. Diffusion based mixing is slow and requires long channel requirement beyond the standard design considerations of typical microscale devices. In order to overcome these issues, there have been several studies in CD based platforms to enhance mixing. Mixing under laminar conditions has been demonstrated in centrifugal flows through straight microchannels (with low aspect ratio) using inhomogeneous distribution of the velocity-dependent Coriolis force (Ducree *et al.*, 2006). It has been shown that transversal flow components induce stirring and flipping of concurrent liquid streams through a radial channel and result in flow patterning. Further, enhancement has been demonstrated using switching the rotation speeds at clockwise and anticlockwise direction at regular intervals (Grumann *et al.*, 2005; Haeberle *et al.*, 2005; Steigert *et al.*, 2005). Magnetic stirring with introduction of paramagnetic particles in the liquid and triggering these particles by positioning permanent magnets in non-symmetrical fashion underneath the mixing chamber lead to further reduction in mixing time (Grumann *et al.*, 2005; Brenner *et al.*, 2005; Kido *et al.*, 2007).

1.6 BIOLOGICAL APPLICATIONS OF CD BASED MICROFLUIDICS

CD based microfluidics has been used to automate several fundamental biological operations like blood separation, cell lysis, protein purification, nucleic acid amplification, DNA hybridisation, immunoassays and colorimetric analyte detection. One of the pivotal steps for clinical diagnosis with blood involves separation of different components form the blood plasma, so that these cellular components do not interfere with amplification of Nucleic Acids. Inspired from the commonly used centrifuges in laboratories, CD based platforms use the centrifugal force to separate plasma from the whole blood owing to the density differences in the components of blood. Several devices have been developed to cater the needs of different volumes of blood extraction. A small volume (2 μ*l*) of plasma is extracted from 5 μl of blood sample using rotating device (Haeberle *et al.*, 2006). A large-volume CD-based device was developed combining microfluidics structures with large-volume samples and the implications for sample-driven microfluidics systems (Amasia and Madou, 2010). Single step centrifugal hematocrit determination was performed using spun by a macroscopic drive unit containing a downstream blind channel to sediment the blood with centrifugally assisted capillary filling along sloped hydrophilic sidewall along with an upstream metering structure (Riegger *et al.*, 2007). The level of hematocrit is indicated at the sharp phase boundary between the plasma and the segregated cellular pellet in the channels inside the rotating device where a calibrated scale is imprinted. The cost of the device was estimated to be very low with a high degree of linearity while the processing of the blood can be done within 5 mins. A multiplexed centrifugal device was developed by Schembri *et al.* (1995), capable of separating the plasma from 90μL of blood sample and subsequently diluting it into different testing chambers.

An important step for any molecular diagnostic assays is cell lysis, which enables us to extract the intracellular components for studying subsequent downstream processes. It is pursued using different means like – mechanical or physical methods, chemical methods or osmotic imbalance. The method of chemical lysis of cells has been extensively used using surfactants (detergents) or enzymes used to break the cell membranes. Physical lysis methods involve different methods like grinding, sonication or mechanical disruption. A comparative study, however, reveals that mechanical cell lysis is an effective method over chemical lysis to rupture the cell membrane especially for cells with thick cell walls like Gram-positive microbes in

extracting DNA for subsequent analysis (Jarrell et al., 1992; Gorkin *et al.*, 2010). Impacting beads over the cells is a mechanical lysis, alternative to the chemical methods, which utilises collision and shear to break the cell walls. Interactions between beads and cells were demonstrated inside a partially filled annular chamber in the CD, with a programmed alternate rotating speeds, to cause disruption of mammalian (CHO-K1), bacterial (*Escherichia coli*), and yeast (*Saccharomyces cerevisiae*) cells (Kim *et al.*, 2004). Magnetic forces were used to further enhance bead impaction by integrating permanent magnet below the CD (Kido *et al.*, 2007). Further, magnetic blades were placed inside the CD along with grinding-bead media which helps the lysis process using non-uniform magnetic field. This was modified by taking the advantage of both the bead collision and magnetic force, which was validated using Bacillus subtilise spores and nasophargyngeal aspirates (Siegrist et al., 2010). Laser based cell lysis was shown by Cho *et al.* (2007a), utilising localised heat shocks and mechanical shock due to collision from the heated beads and the cells.

An integral step towards analysis of the nucleic acid extracted after cell lysis is the amplification of the target which is performed using enzymatic amplification in the form of polymerase chain reaction (PCR). Thermocycling operation, used for amplification of DNA, has been integrated in a disk based PCR for analysis of E. Coli, with a Peltier thermoelectric device and a thermistor for temperature measurement and control (Kellog *et al.*, 2000). As this operation involves heating cycles with temperatures as high as 95ºC, evaporation loss of samples at such enhanced temperature was compensated by condensation in an upstream chamber and spinning back to the original PCR chamber. In order to achieve homogeneity of the samples, Martensson *et al.* (2006) developed CD based PCR system which utilised the Coriolis force induced mixing with heating performed by infrared radiations. Amplification of Plasmid DNA was performed within the CD based platform and analysed in 50 min with 58-1000 wells containing template (Sundberg *et al.*, 2010). CCD camera was used to acquire fluorescent images of the disk following the PCR steps, and the well intensity frequency distribution and Poisson distribution statistics were used to count the positive wells on the disk to determine the number of template molecules amplified. Target concentrations measured were 3 times less than that predicted by absorbance measurements. CD based platform reduces cost, complexity of the instrumentation, and thermocycling time compared to other current digital PCR platforms.

Immunoassays are used to evaluate drug targets for detection of specific antigens or antibody using biomarkers to aid clinical diagnosis. The test is based on the binding reactions of antigen or antibody, with the labelling of the antigen or antibody with fluorescent or enzymatic labels, gold or radioisotopes. Although immunoassays have been successfully used for detection and quantification of proteins, the tests are mostly associated with labour intensive procedures and time consuming protocols. It involves a series of mixing, incubation and washing steps which may require hours or even to days to complete. Further, long incubation times and lengthy manually operated protocols make it tedious and error prone. Immunoassays integrated into the microfluidic devices allow precise control of fluids throughout the assay process while also reducing reagent consumption. This is also associated with reduction in channel dimensions and shorter incubation times. Enzyme – Linked ImmunoSorbent Assays (ELISA) has been successfully implemented in CD by functionalising the disk surface, spotting directly on it and subsequent reactions to obtain detectable signals from the antigen-antibody complex (Lai *et al.*, 2004). The CD was prepared in PMMA and series of passive valves which released fluids depending on the rotation speed. Fully integrated multiplexed ELISA was demonstrated for rat IgG, running 24 simultaneous assays. Several other immunoassays were reported following this study, which involved targets like Hepatitis A, Tetanus (Riegger *et al.*, 2006), secretory IgA for mental stress (Nagai *et al.*, 2007) and Hepatitis B virus (Lee *et al.*, 2009). Lee *et al.* (2009) utilized LIFM valves (as described in section 1.5.1) to hold reagents and transfer fluids. The sample preparation steps were integrated into the CD with isolating Hepatitis B virus antibodies in plasma from whole blood. The sample was passed into a reaction chamber that contained a bed of functionalized polystyrene beads functionalized with capture antigens and separate antigens labelled with enzyme. The bead based assays have improved capture efficiency, increasing surface area and improving mass transport efficiency.

DNA-hybridization in microfluidic system was developed for a compact disc (CD) platform by Jia *et al.* (2006). CD was created using polydimethylsiloxane (PDMS) using a SU-8 master mold fabricated by a two level lithography process which were developed specially for the microfluidic structures. Self-assembled DNA oligonucleotide monolayers were prepared over gold pads patterned on glass slides to serve as the capture probes. The PDMS stamps were aligned and bonded with the

glass slides to form the DNA hybridization units which were detected using an enzymatic-labelled fluorescence technique. DNA samples (25-mers) of various concentrations down to 100pM were used to demonstrate the hybridisation in this platform and passive assays (no flow) using samples of the same concentrations were performed for comparison. Fluorescence intensity was increased three time compared to passive hybridisation in identical concentration and time of hybridization time (3 min).

Spinning disk interferometry uses changes in the refractive index of beams of light to identify the presence of bound proteins. The change in refractive index is directly proportional to the amount of mass on the surface. Zhao *et al.* (2006) demonstrated label-free detection using this interferometery to measure the presence of bound protein, although the sample preparation steps were carried out on a separate platform. Li *et al.* (2008) integrated bioassays on a standard CDs and readouts from a standard CD player by attaching biorecognition molecules to the CD surface which can bind to a wide variety of medically important target molecules. It involves oxidizing the polymeric surface of the disc to form chemical groups which bind specifically to streptavidin proteins, which can subsequently be used to immobilize various biorecognition molecules. Light reflects through a surface-bound complex with large silver particles grown around gold nanoparticles, which can generate an optical signal. The data on every pre-recorded audio CD are encoded in such a way that errors can be detected and corrected by CD players, using a standard algorithm. The silver particles that are associated with the formation of target complexes system obscure data pre-recorded on the CD, and so can be detected as errors. Each block of data on a CD is associated with a physical position on the disc; these errors can be tracked to their locations. By plotting the rate of error detection against the distance from the centre of the CD, the formation of target complexes at all points on the CD's surface can be quantified.

CD based system has the capacity to change gravity levels by altering centrifugal force which offers an opportunity to investigate genetic geotatic responses in a living system. CDs were used for automated cultivating and monitoring Caenorhabditis elegans (C. Elegans), proficient in automated feeding, waste removal and live-animal microscopy (Kim *et al.*, 2007a). It contains cultivation, nutrient and waste chamber, channels connecting the chambers and venting holes to create such automated cultivation system. The hypergravity induced by such rotating platform is

converted into biological signalling using a genetic pathway involving mechanosensory channels of touch receptor neurons, the neurotransmitter serotonin, and transcriptional factors to regulate development, energy metabolism, stress responses, and aging (Kim *et al.*, 2007b). It was interestingly observed that even after prolonged exposure to hypergravity (for 3 hrs), the worms preserved their muscular and neuronal functions, but showed an amassing of excess fat.

Integration of the dielectrophoresis (DEP)-assisted filter with a compact disk (CD)-based platform was shown by Martinez-Duarte *et al.* (2010). 3D carbon electrodes were fabricated using the C-MEMS technique which was used to implement a DEP-enabled active filter to trap particles with enhanced filtering efficiency in CD based platforms. This work was the first to illustrate the combination of the electrical and the centrifugal forces on a single platform, for demonstrating trapping and selective filtering of yeast cells from a mix of latex and yeast cells at flow rates up to 35 μl/min.

1.7 SOME PERTINENT ISSUES OF TRANPORT PHENOMENA: A REVIEW OF LITERATURE

Rapid advancements in biomedical, bio-technological and micro-electronics applications have resulted in growing demands for more comprehensive understanding of interfacial phenomena over microscopic length scales, rotationally actuated micro-devices being of no exception. In such miniaturized devices, interplay between various forces at different length scales may lead to interesting and non-trivial interfacial dynamics, which are not only rich in fundamental physics and chemistry but are also characterized with major technological implications. In the following discussion, we review four important issues of transport phenomena in microfluidic devices, bearing significant consequences to the central theme of this dissertation.

1.7.1 Capillary Dynamics

The physics of interfacial interactions in narrow fluidic confinements has given rise to many seemingly unresolved anomalies, as attributable to a complex interplay between interfacial phenomena over small scales and topographical features of the confining system boundaries. Dynamical evolution of contact angle in microchannels and nanochannels offers with one such complicated scenario, which essentially stems from the non-integrable stress (Dussan, 1974; Hocking 1977) arising

at the three phase contact line. Interestingly, the linearity in the stress-strain relationship for a non-deformable solid with no-slip condition at the three phase contact line would ensure infinite viscous dissipation (Hocking, 1977), which is physically impossible. Not only that, the physical fact that a capillary front moves over a solid substrate despite no-slip boundary condition at the fluid-solid interface had also appeared to be a mathematical paradox for long. To resolve such apparently anomalous trends, researchers in many cases had abstracted the underlying details of interfacial interactions with the notion of slip-based hydrodynamic boundary conditions (Cox, 1986; Shikhmurzaev, 1997) for non-wetting fluids, with an intention of mimicking the concerned molecular level interactions in an up-scaled continuum limit. Such considerations, nevertheless, are not physically compatible in nature for perfectly wetting fluids (i.e., for limiting small contact angles), for which a thin film typically runs ahead of the meniscus in the form of a precursor films (Leger *et al.*, 1988). Interestingly, the effects of surface roughness on the resultant contact line dynamics do not remain far from being non-trivial. This may be attributed to the fact that irrespective of the nature of wetting from a global perspective, the local contact line dynamics is significantly dictated by small-scale variations in the surface roughness characteristics, which cannot be merely described as sole function of the averaged roughness heights of the surface elements. Such issues are further complicated by the existence of disparate physical scales governing the problem, which have been addressed by the research community primarily through mesoscopic formalisms (Qian *et al.*, 2006; Huang *et al.*, 2007). From a continuum viewpoint, however, several aspects of contact line dynamics over rough surfaces still remain to be poorly understood, particularly within the purview of experimentally realizable physical scales. This deficit stems from the complexities in describing the underlying roughness-wettability coupling over physical scales that are substantially larger than those addressed in related molecular dynamics simulations (Koplik *et al.*, 1988; Thompson & Robbins, 1989; Qian *et al.*, 2003; Koplik and Banavar, 2006).

Comprehensive efforts have been devoted in the literature to analyze the contact line from a continuum perspective (Ren *et al.*, 2010). The contact line, in essence, may be described from a continuum viewpoint by decomposing the contact region into various sub-domains, considering the relative dominance of various forces over different length scales (Cox, 1986; Park and Homsy, 1984; Kalliadasis and Chang, 1994). For example, close to the substrate viscous forces, surface tension

forces and intermolecular forces govern the evolution of a thin film, while deeper in the bulk the substrate-fluid intermolecular forces cease to be important. One may obtain the dynamic evolution of the interface through an asymptotic matching of two regions (Park and Homsy, 1984), namely, an 'outer region' far from the contact line and an 'inner region' formed by a thin lubricating film adhering to the channel walls. Different kinds of inner regions may be formed on the basis of wetting behaviour of the films. In purely wetting limit, the inner region may be described by the evolution of the precursor film (Kalliadasis and Chang, 1994), while for partially wetting surfaces the same may be described by a wedge shaped region with slip at the contact line (Eggers, 2004a). An effective or apparent contact angle may be defined as an extrapolation of the outer profile of the interface towards the solid substrate (Kalliadasis and Chang, 1994). Since this description of the contact angle depends on the contact line velocity, the contact angle also varies dynamically as a meniscus evolves.

1.7.2 Microbubble Generation

Microbubbles have numerous applications, ranging from medical applications such as contrast agent for ultrasound imaging (Stone et al., 2004), targeted drug and gene delivery (Ferrara *et al.*, 2007; Prentice *et al.*, 2005), tumor detection and destruction (Lindner, 2004), to non-medical applications such as heavy metal removal during mineral processing, bubble-based logic circuits (Prakash and Gershenfeld, 2007), reducing drag in ships and pipelines (Murai *et al.*, 2003) and even lowering water temperatures in the sea by enhancing its effective reflectivity. Several approaches of microbubble generation have accordingly been investigated in the literature (Chen *et al.*, 2009; Cubaud and Ho, 2004; Cubaud *et al.*, 2005; Ganan Calvo and Gordillo, 2001; Garstecki *et al.*, 2004; Garstecki *et al.*, 2006; Gordillo *et al.*, 2004; Hettiarachchi *et al.*, 2007; Xu *et al.*, 2006). Many of these strategies have been guided by the fundamental principle that in the low Reynolds number based hydrodynamic regime typical to microfluidic devices, the flow is essentially dominated by a delicate balance of the viscous forces and the driving flow actuating influences, precluding any significant role played by the inertial effects (Ganan Calvo, 2004). Considering such constraints, several innovative microbubble-generation strategies have been suggested in the literature in the recent past. For example, air may be released through specially designed nozzle system (e.g. T-junctions), so as to generate small scale bubbles (Xu *et*

al., 2006) by rupture of cross-flowing streams. The mechanism of the formation of droplets in a T-junction for small values of the capillary number is governed by the dominant effect of balance of hydrostatic pressures in the two immiscible fluids. The 'squeezing' mechanism, specific to microsystems is associated by blockage of the channel by a liquid and or gaseous plug and subsequent breakup (pinching). It has been demonstrated that the break-up is not dominated by shear stresses but from the pressure drop across the emerging droplet or bubble (Garstecki *et al.*, 2006). Power ultrasound may also be utilized to induce locally cavitated small bubbles at high rarefaction points in standing ultrasonic waves (Azuma *et al.*, 2005). Emerging of droplets from nozzles exhibits finite time singularities. Apparently, it may seem that dynamics of the formation of the droplets differ from one case to another. However, a detailed analysis reveals that the dynamics near pinch-off ought to exhibit self-similar behavior due to the orders of magnitude difference between local length and time scales and corresponding global scales (Basaran, 2002). The dynamics near the pinch point should be identical in every situation irrespective of the global experimental conditions.

1.7.3 Microfluidic Mixing

Several strategies have been adopted to achieve fast mixing in low Reynolds number situations, which are classified as either active or passive strategies. Active strategies involve external energy input in the form of electromagnetic field, pressure modulation or acoustic field, whereas passive approaches mostly utilise alteration of the geometric features of the microchannel. Typically, in both of these approaches, a secondary flow is induced in the microchannel in a direction perpendicular to the axial flow. Some of the techniques utilize mechanical pulsation (Glasgow and Aubry, 2003) of fluids to create a change in flow pattern to cause stretching and folding of fluid elements inside the microchannel, thereby inducing chaotic flows. Different chaotic micromixers have been designed by the use of lithographically defined structures (Niu and Lee, 2003) on the surface of the channel, or by temporal active variation of the pressure through multiple side channels (Bottausci, 2004). Spatial flow control has also been demonstrated by modulating electroosmotic flows (Qian and Bau, 2002; Ng *et al.*, 2009; Lin *et al.*, 2004; Song *et al.*, 2010) with varying the zeta potential or the external field along the channel.

In some of the flow situations, transverse vortex arises naturally from specific geometries. Flow in curved channels is one such example where centrifugal force acts in the radial direction because of the change in direction of motion along a curve. This results in a radial pressure gradient whose magnitude can become sufficient to generate a transverse flow field (the so-called Dean Flow) (Berger and Talbot, 1983). This, in turn, may disrupt inherent symmetry of the streamlines to yield chaotic flow trajectory. Flow in curved channels is one such example where centrifugal force acts in the radial direction because of the change in direction of motion along a curve. These concepts have been translated in microscale with the use of 3D non coplanar geometries in the form of helical and twisted microchannels (Liu *et al.,* 2000; Jiang *et al.,* 2004; Scho and Hardt 2004; Sudarsan and Ugaz, 2006). The transverse Dean flows can also be achieved by a planar split-and-recombine (P-SAR) arrangement capable of generating multiple alternating lamellae of individual fluid species, and an asymmetric serpentine micromixer (ASM) configuration coupling vertical transverse Dean flow effects with the action of expansion vortices in the horizontal plane (Sudarshan and Ugaz, 2006).

1.7.4 Microfluidic Separation

Different microfluidic separation methods are classified on the basis of its mode of operation principles. Field induced separation refers to the use of external field – electric, magnetic, temperature or flow which alters the velocity depending upon the property of the analyte e.g. size, charge or density. The second strategy refers to the use of geometric features of the separation chamber, which helps in inducing velocities at different regions for the analytes to experience different forces at different locations. Some of the techniques judiciously couple above two strategies, so as to achieve an efficient separation. Several studies have been devoted to optimise different operations and conditions, to augment the migration of particles, using different flow fields, or by modifying pertinent geometric parameters in order to maximise the separation efficiency (Pamme, 2007). Electric field has been one of the popular strategies of separation in microfluidic devices. Huang *et al.* (2002) developed a 'DNA prism' using an array of posts with application of electric field by continuously switching between parallel or diagonal directions to the posts. Chen and Chauhan (2005) used electric field flow fractionation (EFFF) for the size-based separation of DNA strands in a microchannel. As the electric mobility of the strands is

independent of the length of DNA, an axial electric field cannot separate DNA strands in solution. This necessitates the use of lateral electric fields coupled with an axial Poiseuille flow for size based separation of DNA strands. Das and Chakraborty (2008) theoretically investigated nonlinear electrophoretic effects on the transport and size-based separation of charged macromolecules in nanoscale confinements. Paul and Chakraborty (2007) investigated the influences of the near-wall interaction potentials and the consequent migrative fluxes on the size-based separation of macromolecules in microchannels subject to combined electromagnetohydrodynamic influences. Size based separation processes have also shown to be influenced by alterations in the channel geometry. For example, one may refer to the pinched flow fractionational technique (Yamada et al., 2004. In this technique, two laminar streams – one with the sample containing particle suspension and a carrier stream containing buffer, are pumped through a narrow channel neck before entering a channel with greater cross-section. The flow rate of the carrier stream is set higher than that of the sample stream which results in pushing the microparticles against the wall in the pinched channel segment. Small particles find themselves in a flow stream close to the wall, while the larger particles have their centre of mass drifted further away from the wall. When the particles enter the wider channel, particles get separated along different streamlines and finally get separated along the width of the channel. Deterministic lateral displacement (Huang *et al.*, 2004; Inglis *et al.*, 2006) has been applied to the size based separation of particles by pumping through an array of obstacles used as filters to prevent or block access. Zweifach–Fung effect have been utilised to efficiently separate blood plasma from whole blood with 100% plasma selectivity. The working principle behind this effect is that when a particle approaches a bifurcating region within microfluidic channels, they have a tendency to move toward a daughter channel, which has a higher flow rate compared with the other daughter channel (Yen and Fung, 1978; Fung, 1973). When the flow rate ratio between the two daughter channels is higher than 6: 1, 100% of the particles will flow into the daughter channel with the higher flow rate. It was demonstrated that flow separation of particles and biological cells can also be performed with acoustic forces generated from ultrasonic waves (Petersson *et al.*, 2007; Bhat and Chakraborty, 2010). Flow in curved channel also has been utilised for particle separation which have been demonstrated in spiral microchannel where the bends induce centrifugal force (Bhagat *et al.,* 2008). Hydrodynamic filtration is one of the techniques which utilises both the geometric

design and flow control. It consists of a microfluidic main channel with multiple narrow channels branching to the sides (Yamada and Seki, 2005; Yamada and Seki, 2006). As the fluid is pumped along the main channel, some liquid escapes through the branched channels. When a mixture of particles is pushed through the main channel, smaller particles occupy flow streams much closer to the channel wall than that of the larger particles, because the centre of mass of the particle is located far from the wall. The small particles are mostly located within a stream line that enters the side channel, whereas the centre of a larger particle is still too far away from the wall. This results in the larger particles to flow through the straight channel while the smaller particles escape through the branching channels. However, if a particle is located initially far away from the channel wall, it would eventually get closer to the wall after they pass an outlet branch, to get finally aligned along the wall.

1.8 IMPORTANT INFERENCES DRAWN FROM LITERATURE REVIEW

1.8.1 Capillary Dynamics

Although dynamics of capillary motion has been extensively studied in the literature, many of the fundamental issues pertinent to the same in small-scale systems, especially those related to the combined effects of roughness and wetting phenomenon, remain far from being well resolved. In general, whenever a liquid-vapor interface is formed inside a capillary, its topographical evolution is effectively governed by the combined dynamics of an 'outer region' far from the contact line as well as that of an 'inner region' that is formed by a thin lubricating film adhering to the capillary walls. The dynamic contact angle corresponds to an extrapolation of the outer profile of the capillary meniscus towards the solid substrate. Therefore, the contact line motion is merely an illusion of the dynamical behaviour of the outer region. In reality, it is rather difficult to simulate the entire interface from a thin film to a macroscopic meniscus from a continuum perspective. This difficulty stems from the fact that the microscopic and macroscopic regions are characterized with length scales that are widely disparate in order. In the literature, this issue has typically been handled by decomposing the problem into different length scales and formulating a matched asymptotic analysis that links the outer region to the precursor film in front of the meniscus, through an intermediate lubricating film. However, investigation of the dynamics of moving contact lines in microfluidic confinements under the

combined effects of different wetting conditions of the surface and the presence of surface heterogeneties have not been yet studied. In particular, the effects of slip, correlation length and other roughness parameters on the dynamic contact angle are yet to be systematically explored. Further, no studies have yet been reported in the literature depicting the influence of rotational effects on the contact line dynamics of an immiscible two-fluid system, considering the above-mentioned complexities aptly into account.

1.8.2 Microbubble Generation

Microbubbles are commonly generated by mechano-fluidic action. Traditionally, these are obtained by compressing air into liquid with subsequent dissolution of nucleating bubbles from supersaturated liquid. An alternative strategy is to utilize the phenomenon of "flow-focusing", in an effort to force a central stream of gas to impinge onto a liquid flow at a narrow orifice. Different pneumatic actuators have been utilized towards the formation of droplets/bubbles as well. Importantly, the generation of microbubbles using the above traditional methods necessitates complex and delicate external pressure balance and flow control. Achieving such control, in turn, may demand somewhat sophisticated and complex platforms with limited capability of establishing an integrated environment for microbubble generation and manipulation. On the other hand, it may be more appropriate to regiment fundamental operations for bubble generation and manipulation on a CD in an integrated fashion, which is an issue that remains to be emphasized in the literature.

1.8.3 Microfluidic Mixing

Most of the mixing processes implemented through rotational platforms, as reported in the literature, involve mixing in large scale chambers, but do not commonly involve microfluidic channels. The only aspect of microscale mixing that has been studied on a rotating platform has essentially been based on a diffusion dominated regime. However, different regimes of flow mixing over microscopic scales may exist on a rotating platform, as characterized by the relative dominance of different forces influencing the mixing process. Each of the regimes, in turn, may be characterized by different flow behaviour distinctly different from that of the diffusion based mixing process. However, any comprehensive quantitative study on different micromixing regimes on a rotating platform is yet to be reported in the literature.

1.8.4 Microfluidic Separation

Centrifugal force has been used traditionally to separate analytes from a mixture of particles based on density. Macro-scale chambers have been integrated into the CD platform to separate different components from the blood. What remains to be studied in this perspective is the consequence of miniaturization of separation processes in rotational platforms, which may bear an important advantage in terms of the reduction of the sample volume. No studies, however, have been reported in the literature illustrating the capabilities of both the centrifugal and Coriolis force for size-based separation of analytes in microchannels. Nevertheless, it is imperative to study microscale separation process in order to achieve true integration of the different operations in a CD based microfluidic system.

1.9 AIM AND SCOPE OF THE DISSERTATION

Considering the shortcomings in the reported literature delineated as above, the present study focuses on four important and inter-related facets of interfacial transport in rotationally actuated microfluidic devices, as discussed below.

First, we characterize capillary filling dynamics on a CD as a simultaneous function of surface roughness and wettability conditions, as well as the rotational speed of the fluidic platform. It is important to note in this context that during the capillary filling of a microchannel, the fluid enters into a microchannel by the effects of surface tension. However, capillary filling in rotating microdevice, further advancement of the capillary front is observed with an additional driving influence of the centrifugal effects. The fluid motion is opposed by the viscous resistances, as determined by the different flow regimes instantaneously prevailing within the liquid in the capillary. A critical assessment of the underlying consequences would effectively demand a comprehensive analysis of the complicated interplay between various forces dictating the interface evolution. The effects of surface wetting condition also play a crucial role in altering the contact line dynamics, bearing particular non-trivial interactions with the topological features of the solid boundaries. In an effort to assess the underlying consequences, we aim to study different surface wetting conditions, namely, completely wetting substrate and partially wetting substrate. The former entails the presence of a precursor film running ahead of the meniscus while the later results in a film terminating to form a wedge like thin film. Our studies reveal that the consequent wetting characteristics are strongly influenced

by action of intermolecular forces in presence of surface roughness features, which helps in increasing the area of contact. We also observe the effect of slip, surface correlation length and roughness parameters on the dynamic contact angle. A reduced order model is developed to describe the capillary filling of purely wetting fluid which needs to be validated using full scale numerical model and experimental results for low and high rotation speeds.

Secondly, we exploit the rotational forces for microbubble generation on a CD. We demonstrate explicit control over the frequency and dimensions of the bubbles generated, by programming the rotational speeds in a dynamical environment. We further study different regimes of two phase flows in the form of the multiphase jets and their stabilities, in an effort to gain control over the size and shape of the bubbles generated within a broad domain of rotational speeds. Further, it may be necessary to break the larger bubbles into numbers of small bubbles, as per the requirements of the downstream process. Accordingly, we aim to demonstrate a mechanism of splitting of the larger bubbles in a rotational environment, and transportation of these bubbles into a designated chamber.

Thirdly, we exemplify the use of CD as an efficient micro-mixing platform. As mentioned earlier, several strategies have been reported in the literature to increase mixing efficiency in microdevices. Intuitively, it is expected that mixing may be facilitated by augmenting transverse components of the flow velocities. However, contrary to common intuition, here, for the first time, we aim to demonstrate that mixing performance may not be trivially improved with enhancements in transverse velocity components, signifying a criticality as well as optimality in designing of mixing devices for energy-efficient performance. We delineate this principle through implementation on a rotationally actuated microfluidic device (Lab-on-a-CD). In effect, we demonstrate that there exists a critical regime of the rotational speeds in which the mixing performance deteriorates instead of improving, with further increase in rotation speed, bearing far-ranging scientific and technological consequences. We also unveil the mechanism responsible for such non-trivial mixing characteristics. Understanding such mechanisms may prove to be an essential step towards the development of integrated bio-microfluidic devices in the Lab-on-a-CD framework, with improved mixing characteristics achieved in an energy efficient paradigm through operation in the regime of optimal rotational speeds.

Finally, we investigate the deployment of CD as a particle separation platform and delineate the effects of Coriolis force on the same. In this regard, it may be emphasized that microscale size-based separation may act as a precursor towards analyses of raw samples in a Lab-on-a-CD platform. Aim of our analysis, accordingly, is to get an insight on the implications of rotational forces towards particle separation in a micro-confined environment. In a perturbation analysis based paradigm, we effectively characterize the band velocity as a function of two different parameters – the particle size and rotation speed. Based on these considerations, we obtain an important measure of the separation efficiency, namely, the Resolution of separation, as a function of the pertinent parameters.

1.10 CONTRIBUTIONS FROM THE PRESENT THESIS

Primary contributions from the present Thesis may be summarized as follows:

- We have developed a theoretical model using matched asymptotic analysis to study the combined effects of random surface roughness and substrate wettability on the dynamics of the contact lines in microchannels. It has been utilised to develop a reduced order model to study the dynamics of the centrifugally actuated capillary filling with dynamically evolving contact line and has been validated using full scale numerical and experimental analysis.
- We have utilised rotating platform for controlled generation of microbubbles by dynamically tuning the rotational speeds through a simplistic design. We have demonstrated that body forces in rotating platform may alter the stability of two phase jets, which may be utilised for alteration the morphology of the microbubbles.
- Pertaining to mixing in CD, we have identified three distinctive regimes for mixing, namely, diffusion based mixing, Coriolis based mixing and instability based mixing. Our studies have revealed that an intuitive prediction of trivially improved mixing with increased rotational speeds may not necessarily turn out to be true in practice.
- We have utilised CD as an efficient size-based separation tool for particles. We have identified an optimum rotation speed to achieve most efficient particle separation and characterised it using a parameter called resolution, corresponding to a given geometric dimension and particle size combination.

1.11 OUTLINE OF THE THESIS

The remaining part of the thesis is organized as follows:

- In Chapter 2, the fabrication process of compact disk platform is outlined along with integration of different components required for the platform
- In Chapter 3, the dynamics of the capillary filling in microchannels embedded on a rotating platform is studied, considering combined influences of substrate roughness and wettability conditions.
- In Chapter 4, development of a rotationally-actuated fluidic device is discussed in the context of controlled generation of microbubbles with tunable spatio-temporal frequencies and size distributions, as controlled by simply tailoring the rotational speeds, corresponding to given channel dimensions and fluid-substrate combinations.
- In Chapter 5, micromixing in rotating platforms is analysed to identify three different regimes – diffusion based mixing, Coriolis force based mixing and instability based mixing. Alternative strategies of mixing are outlined by varying rotation speed and introducing slug and bubbly flow on CD.
- In Chapter 6, particle separation in rotating platform devices is studied theoretically for separating particles based on size in a microfluidic environment. Particular emphasis is laid to investigate the implications of the rotational speeds on the separation characteristics.
- In Chapter 7, important inferences drawn from the studies in the present thesis and possible extensions of these works are indicated.