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Microfluidic Mechanics and Applications: a Review

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Microfluidics involves the transportation, splitting and mixing of minute fluids to perform several chemical and biological reactions including drug screening, heating, cooling or dissolution of reagents. Efforts have been made to develop different microfluidic devices, droplets and valves that can stop and resume flow of liquids inside a microchannel. This paper provides the review related to the theory and mechanics of microfluidic devices and fluid flow. Different materials and techniques for fabricating microfluidic devices are discussed. Subsequently, the microfluidic components that are responsible for successful micrfluidic device formation are presented. Finally, recent applications related to the microfluidics are highlighted.

Keywords: Microfluidics; Reynold's number; Capillary; Surface tension; Fabrication.

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1. INTRODUCTION

Microfluidics is one of the potential areas of research and is at its initial step of developments to influence other research fields such as chemical, biological, physical, optics and information technology. Microfluidics, technically known as the science of fluid mechanics studied at micro-scale, provide a solution to overcome the problems related to development of an analytical method with high resolution and sensitivity to accomplish microanalysis purposes. Microfluidics deals with the study related to behavior of fluids, geometric manipulation of its micro domain and precise control of small volumes of fluid flow in microlitres (μL), nanolitres (nL), or even picolitres (pL). Microfluidics possesses the aptitude to carry out high resolution and sensitive separations to detect very small quantities of samples and reagents which makes this system relatively inexpensive and requires short time analysis [1]. Microfluidic detection includes some common fluids such as bacterial cell suspensions; blood samples; proteins or antibody solutions. Microanalysis of such fluids is successfully being used in several micro-scale applications and measurements that include fluid viscosity; capillary electrophoresis; DNA sequencing; diffusion coefficient detections; chemical binding; immunoassays; cell separation and many more [2, 3]. These factors are analyzed and measured using a microfluidic device that contains either one or more channel of small dimensions nearly less than 1 mm.

The focus of this paper is to present the recent research work on microfluidic mechanics and its applications that has been comprehensively investigated. The miniaturization and integration of microfluidic components has been exploited immensely resulting into development of several microfluidic devices. Different fabrication methods were reported by the researchers to show some specific application particularly keeping in view an idea to fit 'entire lab on a chip'. Microfluidic components like channels, valves and diaphragms can be made using different materials such as elastomers, PMMA, PDMS and solution gels [2].

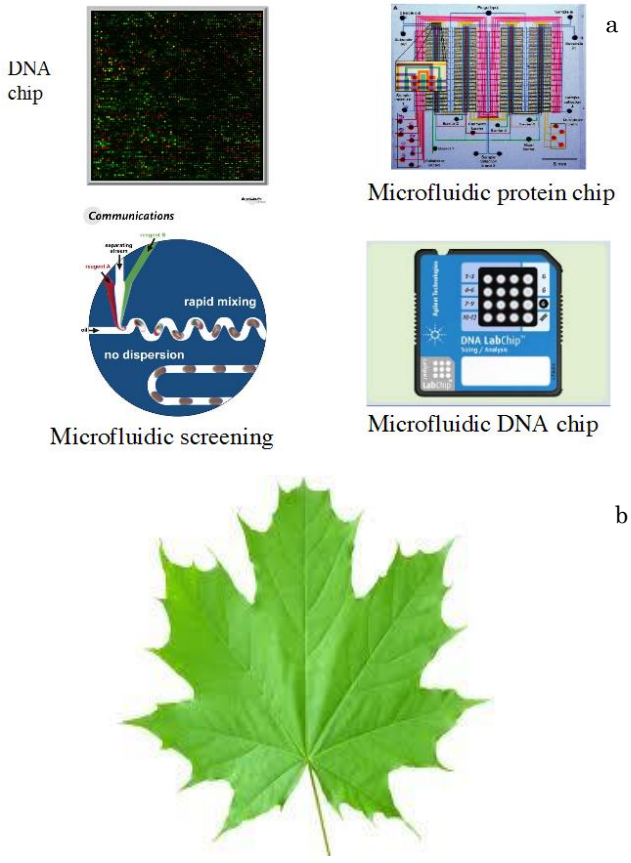

Fig. 1 – Fabricated microfluidics (a) and natural microfluidics (b)

2. MECHANICS OF MICROFLUIDICS

The numerical simulation of microfluidics is essentially valuable to know the behavior of a particular system which is difficult to predict at the outset. It can be analyzed and studied that further requires the incorporation of the complexities of different factors such as channel geometry, fluids flow rate, diffusion coefficients and possible chemical interactions altogether into a numerical model. This numerical modeling can be used to visualize the complex flow phenomenon

which otherwise is difficult to obtain experimentally. Several important parameters have to be studied to model it successfully. Some of them are mentioned in subsequent sections.

2.1 Reynolds Number

In fluid dynamics, when fluid flows through a micro channel, it is important to observe whether it is turbulent, that is, lesser orderly regime or laminar (stream lined flow). Table 1 shows the nature of flow for different Reynolds number [1]. The type of flow can be determined by a dimensionless parameter known as Reynolds number depending on its uncharacteristic flow geometry.

Table 1 – Fluid Flow on the basis of Reynolds Number

Range of Reynolds Number (Re)	Nature of fluid flow
Re < 2300	Laminar Flow
2300 < Re < 4000	Transient Flow
Re > 4000	Turbulent Flow

Reynolds number [7] is used to characterize the flow of a fluid through micro channel and is defined as

$$\mathrm{Re} = LV_{avg}\rho/\mu,$$

where μ is the viscosity, is the fluid density, V_{avg} is the average velocity of flow and 'L' is most relevant length scale, and

$$L = 4A/P,$$

A is crossectional area of the channel and P is wetted perimeter of the channel. Re depends on three factors, i.e. material properties (density, viscosity), boundary conditions and critical velocity.

Table 2 – Properties of different Fluid Flows [12]

Laminar	Turbulent	Periodic flow
Small flow velocities	Curling of field lines	3rd flow regime
Even sliding of adjacent layers	Mixing between adjacent layers	Surface waves
Field of velocity vectors constant in time	"Random" development of field of velocity vectors	Acoustic waves
Shear stress depends on viscosity (μ) and independent of density(ρ).	Flow patterns increasingly turbulent towards high velocities	
	Sometimes laminar flow preserved up to higher velocities	
	Shear stress :function of density (ρ) of density	

Table 2 shows different fluid flow and its properties [7]. The lower values of Reynolds numbers represents the laminar flow as the viscous forces are dominant and shows smooth fluid motion where as high Reynolds numbers represents the high inertial forces representing flow variations and the turbulence. This indicates that on the basis of Reynolds number value and channel crossectional geometry, the variation in critical transition point of fluid flow can be determined, i.e. whether the flow is laminar, transient or turbulent as tabulated below. It has also been reported that momentum based phenomenon's such as flow separation can be achieved at Reynolds numbers below 100 [8].

2.2 Flow Types in Microfluidics

The different types of flow [9] are as:

Bubbly flow: With a high focus of bubbles in the upper half of the tube the gas bubbles are isolated in the liquid due to their buoyancy. The bubbles tend to disperse uniformly in the tube when shear forces are dominant. The regime only occurs at high mass flow rates in horizontal flows.

Slug flow: The diameters of out such elongated bubbles can also be termed as large amplitude waves.

Stratified-wavy flow: Waves are formed on the interface when the gas velocity is increased which travel in the direction of flow and amplitude of the waves is noteworthy and depends stretched out bubbles become similar in size to the channel height at higher gas velocities.

Annular flow: A continuous annular film around the perimeter of the tube is formed by the liquid at large gas flow rates, same as in vertical flow but differs only in the liquid film which is thicker at the bottom than the top.

Mist flow: The liquid may be stripped from the wall and entrained as small droplets at very high gas velocities as in case of vertical flows in the now continuous gas phase.

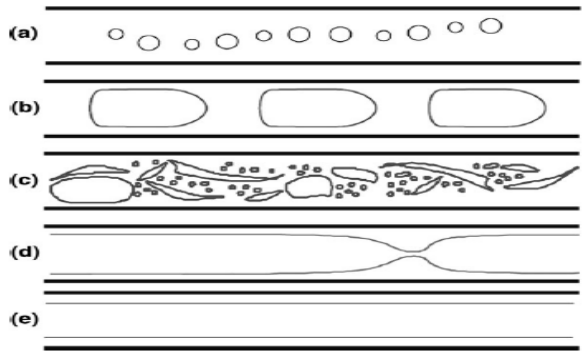

Fig 2 – Flow regimes in microchannel [9] [10]. a) bubbly flow, b) slug / taylor flow, c) churn flow, d) slug / annular flow, and e) annular flow

Sometimes, transition of fluid flow from laminar to turbulent can also occur due to its sensitivity to flow disturbances and channel imperfections. The extreme case of laminar flow is the Stokes flow which involves the creeping motion of fluid through channels at Reynolds number lesser than 1. This is due to greater effects of viscous forces acting relative to the inertial forces at low Reynolds number values.

2.3 Micromixing

Micromixing is a phenomenon in microfluids in which both stirring and diffusion occurs simultaneously. During micromixing, the molecules of different liquids exhibit different properties there by resulting in the random molecular motion at the interface which allows permeability of molecules from one liquid to

other liquid apparently. This permeation is called diffusion which is quite apparent and referred as final stage of micromixing. Micromixing is an important parameter in microfluidic applications especially for bio and chemical analysis that using lab on chips. The low diffusivity of fluids extends chemical reaction time without any chaotic advection. In micromixing, mixing quantities or mixing index, and residence time are used to evaluate the mixing performance and are developed from nonlinear dynamical systems. Since the microfluids deal with fluids flowing at low Reynolds numbers mostly less than 1, the main concern is confined to miscible liquids considering the diffusion as the final stage of micro mixing [13-16].

2.4 Diffusion

Diffusion implies the spreading of particles due to their Brownian motion. This occurs mostly due to thermal energy. The easiest and simplest way of modeling diffusion is in one dimension [17]. This can be represented as

$$X_{RMS} = \sqrt{2Dt}$$

where is the root mean square displacement of a particle in a time t. D is the diffusion coefficient of the particle in surrounding medium, typically 10^{-5} cm/s for a small molecule in water at room temperature. According to Einstein-Stokes equation [18], D is described as

$$\rho \frac{dU}{dt} = -\nabla P + g\rho + \eta \nabla^2 U + \frac{\eta}{3} \nabla \ \nabla . U$$

When two different fluid flows from different reservoirs across a micro channel that has no specific mixing element, the flow is parallel and there occurs no stirring and mixing is completely diffusive.

2.5 Poiseuille's Law

The Poiseuille's law helps in obtaining the flow rate, pressure drop as well as the effective resistance of viscous and incompressible fluids which has laminar flow nature. Mathematically, the Poiseuille's law [1] is given as

$$\Delta P = \frac{8\eta L Q}{\pi r^4}$$

where ΔP is the pressure drop, Q is volume flow rate, L is length of the channel, η is the dynamic fluid viscosity, and r is radius of the channel. The resistance to flow denoted as R and is given by,

$$R = \frac{8\eta \Delta x}{\pi r^4}$$

where x is the distance in direction of flow. Microfluidics is characterized by microchannels that may vary in diameter from 100 nm to 100 micron range or even from 10 nm to 10 microns. At these length scales of micro channels the mass transfer peclet number is large which in turn leads to microfluidic mixing regimes [7]. Peclet number is dimensionless and is used in calculations involving convective heat transfer. It is defines as the ratio of the thermal energy convected to the fluid to the thermal energy conducted within the fluid. Mathematically, Peclet number [19] is represented as

$$Pe' = \frac{heattransportbyconvection}{heattransportbyconduction}$$

$$Pe' = \mathrm{R_e}.\mathrm{P_r} = \frac{\rho.C_p.v_0.(T_1 - T_0)/l}{k.(T_1 - T_0)/l^2}$$

2.6 Other Mechanical Parameters

There are some other parameters which are equally important to understand the mechanics of microfluidics. Some of them are described in this sub-section.

2.6.1 Surface Tension

It is a contractive tendency of the liquid surface that allows it to resist an external acting force as the behavior of the liquid depends on cohesion forces acting between similar molecules. Its dimension is force per unit length or energy per unit area. Surface tension is due to imbalance of intermolecular attractive forces that one molecule experiences in vicinity of other molecules in a liquid. These attractive forces may be hydrogen bonds in case of polar molecules or vander waals forces for other molecules. There exist three system interfaces such as solid surface – gas interface; solid-liquid interface, liquid-gas interface that inturn give rise to surface tension forces in order to maintain a local equilibrium at specific contact angles forming a meniscus. Meniscus is a curve in the upper surface of a liquid close to the surface of the container or another object, caused by surface tension. The fluid attracts to surface due to maintenance of this local equilibrium carried by three system interfaces. This further propels the fluid along the surface [20] [21]. When a single layer of fluid molecules are absorbed by the channel, then dynamic contact angles can be observed which causes the flow through subsequent layers more easily. When the gas pressure of fluid nearly equals the atmospheric pressure acting on the open end of channel, the capillary action achieved is termed as perfect capillary action [20] [21]. Capillary action is defined as the movement of liquid through thin tubes, not a specific force. Capillary flow is due to the balance of surface energy and gravitational potential energy. Table 3 surface tension and thermal coefficient of Fluids at liquid-air interface at 200 °C. The increase in capillary forces with time depends on channel aspect ratio. Higher the aspect ratio of the channel, faster is the displacement and this leads to increase of capillary forces with time. The surface tension is determined by equation

$$\gamma = \frac{U}{2\delta^2}$$

where γ is surface tension, U is the average cohesive energy of a molecule, δ is characteristic dimension of a molecule and δ^2 represents the effective surface area of molecule. Total energy E stored in the interface is

$$E = \gamma S ,$$

where S is total surface area of interface [22]. Table 3 shows the values of surface tension and the thermal coefficients of some fluids used in the microchannels.

Table 3 – Surface Tension and Thermal Coefficient of Fluids at Liquid-Air Interface at 200 °C [22]

Liquid	Surface tension (γ)	Thermal coefficient (A)
Acetone	25.2	– 0.112
Benzene	28.9	– 0.129
Ethanol	22.1	– 0.0832
Glycerol	64.0	– 0.060
Mercury	425.4	– 0.205
Water	72.8	– 0.1514

2.6.2 Contact Angles

Capillary flow is required to maintain the surface tension equilibrium that can be achieved by varying the contact angles. Contact angles can be calculated using young's modulus and is an essential parameter in the process of slug formation. It is used to determine the feature when gas and liquid slugs interacts with the channel wall. The wall adhesion equation helps to determine the shape of fluid interface by specifying the value of static contact angle.

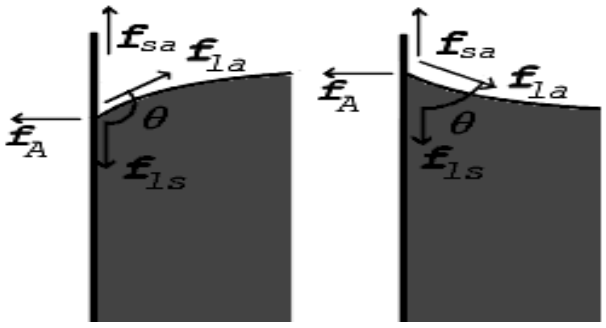

Fig. 3 – Different forces acting at contact angles (θ). From article [22]

The contact angle at any point of intersection is used to organize surfaces as wetting, hydrophilic, non wetting or hydrophobic. The wetting surfaces includes the contact angle range as $0 \le \theta < 900$ where as the range of $900 \le \theta \le 1800$ includes the non wetting surfaces [22]. Fig. 3 shows the formation of contact angle at the surface of a fluid. The contact angle is given as

$$\gamma_{LG}\cos\theta = \gamma_{SG} - \gamma_{SL}$$

The wall adhesion equation can be cast into the better known form

$$\left(p + \frac{n^2 a}{V^2}\right) V - nb \ = nRT$$

where, $a = N_A^2 a'$ is a measure of the attraction between the particles, and, is the volume excluded by a mole of particles. p is the pressure of the fluid, V is the total volume of the container containing the fluid, n is the number of moles, and T is the absolute temperature of the system [22].

3. DYNAMICS OF FLUIDIC FLOW

The dynamics of microfluids includes the parameters such as viscosity of fluids, resistance of fluids and capillary pressure. These parameters are illustrated below.

3.1 Viscosity of Fluids

The fluid – fluid interface gets affected due to variations in surface tensions that lead to marangoni flow instabilities. For example, surfactant laden flows exhibits surface tension variations at either gas-liquid or liquid- liquid interface causing instabilities due to accumulation of surfactants close to stagnation points [5] [27]. The marangoni effects for gas-liquid interface cause hardening of the gas bubbles that attains by surrogate no-shear boundary condition or with a no slip boundary condition. These effects alter the pressure drop and theoretical calculation based on no shear at new interfaces in microfluidic network which require intense care for its use in practical applications [28] [29]. The flows that are driven by surface tension gradients are Marangoni flows. Marangoni flow is generated by gradients in temperature or chemical concentration at an interface as it depends on surface tension. If the surface tension of a interface is varied, there would be disproportion of the forces which would result in flow. This flow is called the Marangoni effect.

3.2 Pressure Driven Micro-Flows

Navier stocks equations simplifies the flow in nano and microfluidic systems and is the ratio of the inertia terms to the viscous term that is characterized by Reynolds number Re attaining negligible value much lesser than one [30]. According to Navier Stockes equation

$$0 = -\nabla p + \eta \nabla^2 \vec{\mu}$$

where p is the pressure, u is the fluid velocity and μ is the dynamic viscosity of the fluid. This equation helps in determining various shapes of micro channels through which the fluid flows [31]. The relation between the pressure and flow rate of system is obtained by the Hagen-Poiseuille equation as described above [32].

3.3 Fluidic Resistance

The fluidic resistance comprises of fluidic analogy to electrical circuit. The fluidic resistance depends on the crossectional geometry and can be obtained by [33]

$$\frac{3\eta L}{h^3 w} > R_{fluid} > \frac{8\eta L}{\pi h^2 w^2}$$

where h is the smallest dimension of crossection of channels and w is the widest dimension of channel cross sections [31]. The average fluidic flow rate in a microchannel depends on pressure gradient imposed at capillary ends on both sides proportionally and it justifies haven Poiseuille's equation as classical ohms law given as

$$\Delta P = R_{fluid} Q$$

where R_{fluid} depends on geometry of the channel crossection. In any micro or nano networks, the fluidic

resistance can be calculated in same way as for electrical circuits i.e. by Kirchhoff's laws [33]. Fluidic resistance also helps to calculate the effective section S_{effect} that helps to calculate pressure drop in microchannels. The effective section is calculated as

$$S_{effect} \approx \sqrt{\frac{8\eta_0 L_0}{\pi R_{total}}}$$

3.4 Fluid Flow Control

Fluid flow can be controlled externally by three different ways. Hydrostatic generator is one such system by which flow is controlled using pressure difference by varying the altitude of fluid to atmosphere interface in different reservoirs [31]. Another system called pressure generator controls the flow rate by varying pressure drops. It comprises of a compressor, a static pressure regulator and a manometer to keep eye on pressure values with respect to atmospheric pressure. The compatibility of all these components must be essentially good to achieve the precise and robust results. Pressure control can also be achieved using a set of electro valves enslaved electronically to a pressure sensor. Syringe pumps are used to control the flow rate directly. The main advantage of such system is that the flow is independent of fluid resistance across the micro channels. The limitation of syringe pumps is that at low flow rates there occurs development of pulsate flow rates and the time required to stabilize them is in negligible. There are certain other pumps that are not perfect flow generators due to low flow rate by back pressure. This includes peristaltic, piezoelectric etc. In contrary Electro osmotic pumps are based on electrical pumping of fluid through nonporous materials which can bear the back pressures but it do not reveal flow fluctuation problems and requires low conductive fluids there by suffering from lack of reproducibility [33] [34].

3.5 Capillary Pressure

Capillary pressure of a flow depends on capillary action. Capillary action is the base of microfluidics. The fluid flow in microchannel keeps advancing due to the surface tension induced whenever fluid interacts with the hydrophilic micro capillary channel interface [35]. The adhesive intermolecular forces at liquid-material interface are much stronger than the cohesive intermolecular forces inside the liquid. Human eyes are best example of capillary actions which is used to clear tears on every eye blink passing the tear to canaliculars present in the inner corners of eyelids. Absorbent paper towels, candle wicks as well as thin layer chromatography presents capillary action in different ways. Due to capillary action the amount of material drawn is called the height h of liquid, and is calculated as

$$h = \frac{2\gamma \cos\theta}{gr\rho}$$

where γ is surface tension at liquid – air interface, θ is contact angle at the surface, ρ is density, g is Gravitational force and r is column radius [22]. Fluidic flow in capillaries with internal crossectional dimensions above 1 mm in a system named as milli fluidics. Milli fluidics has limitation when compared to microfluidics or other conventional fluidics that the transition to turbulence cannot be neglected once the Reynolds number value reaches 1 [36].

4. MATERIAL FOR CHANNEL FABRICATION

Some important materials that have been used by researchers for microfluidic device fabrication are shown in Fig. 4 and are discussed in this section.

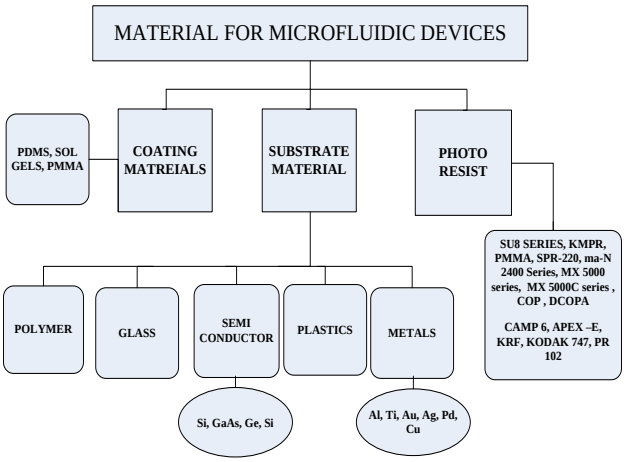

Fig. 4 – Materials used for microfluidic device fabrication

4.1 Substrate

Silicon and glass substrates are used for small channel fabrication in microfluidic system with high resolution and can be easily integrated into clean room environment. Metals like aluminium, titanium copper etc. are commonly used in MEMS fabrication depending on the required parameters such as mechanical, electrical, magnetic and chemical properties that also includes young's modulus, density, Poisson's ratio, fracture toughness etc [37] [38]. Sol Gels is another material and is silicate based compounds formed by condensation of $i(OH)_4$ by loss of water. It requires formation of Si-O network by thin film and should not contain OH as SOG is prone to shrinkage [40] [41].

4.2 Poly Dimethylsiloxane (PDMS)

Poly dimethylsiloxane (PDMS) alternately named as Sylgard 18 is commonly used in microfluidic applications. The empirical formula of PDMS is $(C_2H_6OSi)n$ where n is the number of monomer repetitions [42-43]. If the value of n is small then PDMS may be liquid and is semi solid for large value of n. It is a mixer of cross linker agent Siloxane in 1 : 10 ratio. Any increase in this ratio increases the rigidity of PDMS [26]. Mechanical properties can also be changed by altering the ratio of PDMS with other materials [44]. PDMS is optically transparent, nonflammable, cheap stable, glass fusible and inert when comes in contact with chemicals [45]. PDMS often becomes an elastic solid forming a hydrophobic surface when activated or cross linked [46]. This reason that PDMS surface never gets wet by polar molecules such as water which otherwise can lead to hydrophobic contaminant absorption [47]. When PDMS comes in contact with the flat substrate, there exists weak temporary spontaneous bonding [48]. For organic solvents, it may cause swelling and hence can't make

contact with PDMS. The elastomeric properties of PDMS also make it vulnerable to specific problems such as problems due to pressure gradients or gravitational deformations that cause closing of channels [49].

PDMS is poured into a mould and is placed in a furnace. After getting hard, mould can be taken out leaving behind the replica of microchannels in PDMS. Further the inputs and outputs of the microfluidic device are drilled with needle or punched to allow the injection of fluids to study future experiments. At last, the face of the PDMS block with microchannels is bonded to a glass slide by using plasma treatment that closes the microfluidic chip. This accomplishes the microfluidic device. PDMS are preferred due to transparency at optical frequencies (240 nm-1100 nm) that causes the observation of fluid in microchannels visually or under microscope possible [50]. However, PDMS also have certain limitations. Metal and dielectric depositions on PDMS are almost impossible; hence the integration of electrodes is limited [51]. Another limitation of PDMS is its aging property due to which the mechanical properties can be changed after few years. It is not feasible for biological studies. PDMS is also sensitive for exposure to some chemicals [52]. Besides Sylgard 184, PDMS RTV-615 is also preferred due to its strong and easy bilayer bonding but it is usually dirty and requires adjustment of certain bonding parameters on each purchase [37] [53-54].

4.3 Polymethyl Methacrylate (PMMA) and SU-8 Series

This is a polymeric material used for imaging sub 0.1μm and non imaging microelectronic applications. It is used as a high resolution positive resist for direct electron beam, X-ray or deep UV micro lithographic techniques. It is used as bonding adhesives that provides protective coating for wafer thinning and is available as MCC PMMA in package sizes varying between 500ml to 20 liters. It has fine submicron line width control and has broad range of molecular weight and dilutions. And has excellent adhesion to most of substrates there by making it compatible with multi layer processes [55].

SU-8 Series micromolds are used to produce microstructures in PDMS, PMMA and other materials. SU-8 is highly functional and easy integrable negative photo resist used in microfluidics as it contains excellent mechanical properties, etch resistances, high bond strength , low process temperature, thermal stabilities, high aspect ratio with faster drying and are chemically stable. SU-8 is an organic resin manufactured by micro chem. inc. with composition gamma butyrolactone, mixed triarylsulfonium hexafluoroantimonate salt, propylene carbonate and epoxy resin with elastic modulus 4.4pa, Poisons Coefficient 0.22, thermal conductivity of 0.2 W/mK and thermal expansion coefficient as 50 ppm/k. Further it has 200 C glass transition temperature and a refractive index of 1.8 at 100 GHz and 1.7 at 1.6 THz and Dielectric constant of 3 at 10 MHz [48]. It is used to coat thin layers or film on wafers with thickness varying from 1 micron to 2 mm. It is easy to process using simple mask aligners and mostly used near UV light at 365 nm as optimal absorption source to enhance crosslinking [56-58].

4.4 KMPR

KMPR is a high contrast, epoxy based photoresist that can coat 4-120 micron in a single coat using four standard viscosities. It is compatible to aqueous alkaline developers TMAH and KOH and has high aspect ratio imaging with vertical side walls and excellent dry etch resistance with film thickness > 100 micro m in a single coat. KMPR 1000 has excellent adhesion and plasma or chemical resistance and require conventional UV (350-400 nm) radiation. The normal process is Spin coat, soft bake, exposure, PEB followed by the developer [59].

5. FABRICATION TECHNIQUES

Several fabrication techniques are involved for microfluidic fabrication. In this section, some popular fabrication techniques are discussed that are mostly used by the researchers.

5.1 Micro-Machining and M4 Technique

Micromachining eliminates the wafer to wafer alignment steps and is derived from SCREAM process. In this, single sided silicon wafer is developed and is known as buried channel technology (BCT) that provides an alternate to bulk and surface micro machining using the substrate surface efficiently [60].

The BCT is based on 10 basic steps as follows:

1. Cover the bare substrate with a suitable mask material.
2. Patterning is done by lithographic techniques followed by etching.
3. Protect the trench coating.
4. Etch the trench in the substrate followed by suitable coating.
5. Remove the coating at the bottom off trench.
6. Etch the structure in the bulk of substrate.
7. Strip off the coating which is followed by filling the trench with suitable material there by sealing the structure.
8. If required the structure may be released partly.

The distance of the channel from the substrate surface is defined as the depth of the trench. Crystal orientation of silicon wafer and etching time gives the shape and dimension of the structure [61] [62].

M4 is another microfluidic channel fabrication technique which depends on the parameters like channel size, aspect ratio, surface roughness etc. It either includes the direct fabrication or uses microchannel master mold for replica. There are several M4 techniques such as micromilling, Micro Electrical Discharge Milling (micro EDM), Micro turning, etc. Micro and milling is direct microchannel fabrication technique on polymeric materials. It can be used for metals and nonmetals but it forms more burrs on metallic channels than plastic ones where as Micro ED milling is a cheaper and reproducible mass production technique of micro channels on polymeric materials. It is used for master mold fabrication to replicate microchannels on polymer by micro hot embossing .The aspect ratio and surface roughness of microchannels using micro ED milling is higher than the micro channels

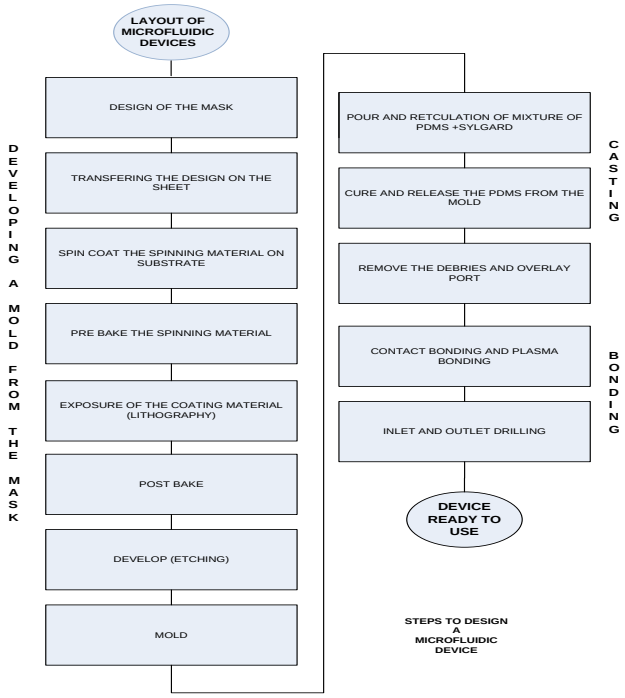

Fig. 6 – Flow-Chart showing microfluidic fabrication process

produced using micro end milling. When different M4 techniques are integrated together, a microfluidic device fabricated can be used for commercial purposes [63-65]. Microchannel surface can be inspected using SEM while surface roughness of fabricated microchannel can be measured using surface profilers.

5.2 Plasma Enhanced CVD

This technique is considered as one of the most important technology for amorphous thin film depositions [66]. It provides high deposition rate relatively and has lesser risk to particle contamination. PECVD was primarily used for masks diffusion and passivation and is useful in optical integration due to flexibility of depositing desired properties of thin films with high deposition rate. Deposition parameters such as temperature, pressure, RF power, and precursors gas mixture can be adjusted with respect to the desired chemical and physical properties.

5.3 Photolithography and X-Ray Lithography

In Photolithography technique, patterns are created when photosensitive material is exposed by UV light to generate the desired pattern. Material is deposited either using physical or chemical deposition processes such as vapor depositions, CVD, plating, epitaxy, etc [27]. Selective removal or creation of features on material is achieved by etching process that may be wet etching or dry etching method [27]. Substrates are sticked by bonding process which includes anodic fusion, thermal compression, or adhesive bonding.

X-ray lithographic technique uses the exposure energy source much shorter in wavelength than light wavelength and hence provides an increase in lateral resolution. The micro manufacturing is done using x-rays piercing deep into the photo resist. This technique is used to attain upto 1 mm deep patterns in microstructures as compared to optical lithography [67]. X-ray lithography is an expensive technique due to high operating cost of synchrotron upto hundreds of dollars per hour for its use. A new process was developed called Lithographie ,Galvanik and Abformung (LIGA) to overcome the expenditure of expensive material like synchrotron for fabrication of microstructures [68].

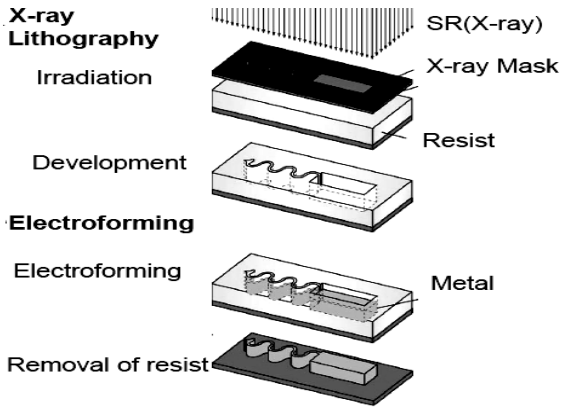

Fig. 7 – Steps in lithography

5.4 Lithographie, Galvanik and Abformung (LIGA); Lab on a CD

LIGA is a German acronym which means X-ray lithography, electroplating and molding respectively. The micro structures manufactured using this process is versatile with high aspect ratio, and stable. It has dimensional stability varying in range of some micro meters to some cms. The microstructures produces are feasible deep in range of about 50 nm. The processing steps for LIGA are shown in Fig. 7 [69].

Economical devices related to microfluidic and surface Plasmon resonance can also be produced by combining photolithographic and optical disc manufacturing techniques. The Lab on a CD is a high performance technique used for the automation of multiple micro reactor systems [70] [71]. This technique is efficient for mass production at low cost using optical discs such as CDs, DVDs, and blue-ray discs in which the track pitch can be designed at different values such as 1600 nm, 740 nm and 320 nm respectively.

The minimum individual pit marks vary in size from 830, 400 and 150 nm and the size of these pitches and pit marks recorded on optical discs lies at micro / nano scales which provides a promising technique for microstructure fabrication on the substrates [72]. This technique is it a good alternative to photolithography with direct laser writing on optical dics. The structure writing speed can be improved by adding the light emitting polymers (LEPs) to a conventional vinyl-based photo polymer system or customized poly functional aliphatic epoxy ether [73]. The writing speed can be increased by a factor of at least 100 using direct laser writing equipment. The use of Diffractive optical elements (DOEs) in the beam path of 325 nm permits the multiplexing of wave guiding in a single exposure [74]. The pitch of written waveguides can be determined by the following equation.

$$d = \frac{2,44M^2 f\lambda S}{D}$$

$$S = \frac{f\lambda}{T}$$

where f is focal length of lens, D is the diameter of incident beam, λ is the wavelength of the incident beam, T is period of fan-out DOE, and S is the sensitivity of photopolymer [75-76].

5.5 Embossing and Laser Ablation

It is another economical technique that requires an access to hydraulic press equipment and a patterned stamp to process the microfluidic design. It is a time consuming process that uses thermoplastic in the form of flat sheets and can be reshaped by heating near glass transition temperature of the material. Thermoplastic materials include PMMA, Polycarbonate, cyclic olefin copolymer, Polystyrene, PVC etc. [77]. For creating metal stamps, silicon or other metal and micromachining tools are used on silicon wafers. Electroplating is done using LIGA process [78-79]. The stamp after being created is placed into a hydraulic press and then heated. The pressure is applied for embossing the plastic against the stamp. The heating can also be replaced by applying more pressure while embossing. In order to eliminate air bubbles trapped between the substrate and stamp, embossing employs specialized vacuum presses to achieve precise replication [80].

It employs a high power pulse laser to remove material from a thermoplastic sheet with UV pulse rates of 10-104 Hz. Lasers with different intensities are used for removing different materials. For example, Lasers of intensity 193 nm are used to erode Polycarbonate, Polystyrene, Cellulose, Polyethyleneterephthalate while materials such as PMMA, PC, PVC etc. can be eroded using lasers with intensity 248 nm [81] [82]. The depth of channels along with pulse energy depends on the pulse rate and the substrate absorption characteristics. A metal or lithographic mask is used to protect the desired area before exposing it to a laser. A direct writing process for the pattern transfer can also be used. The desired channel system is obtained if the program translation of the stage is according to the specified pattern. The use of laser ablation for plastic microfluidic device fabrication are advantageous in prototype applications due to programming of microfluidic designs in to system easily but it has a disadvantage of direct laser writing [75] [83-84].

6. PHOTORESISTS FOR MICROFLUIDICS

Photoresists are viscous materials that are sensitive to light to form raised patterns in the microfluidics devices [45]. Photoresist is mainly used as a mask to transfer pattern of metals dielectrics or other materials. This mask is removed after adding or removing a layer. Epoxies and chemicals containing oxygen atom bonded other carbon atoms forms the basis of photoresists. Its principle is based on the transfer of reactants from a material with low molecular weight into a densely interconnected network [85]. Photoresists are of two types, i.e; positive photoresists and negative photoresists.

In positive photoresist, underlying material to be removed is coated with resist to be exposed to UV light. This exposure changes the chemical structure of the resist to make it soluble in the developer solution which is use to wash away the expose resist leaving behind the windows of bare underline material. The mask thus obtained is exact replica of the pattern desired to be imaged on wafer and is used as a template for further processing or fabrication. The commonly used positive tone resists includes PMMA Series. S1800 series require g-line exposure intensity, SPR-220 require i – line intensity and ma-P1200 Series comes under broad band intensity for exposure [85].

Negative photoresist get polymerized and cross linked when exposed to UV light and is difficult to dissolve in the developer solution. The negative photoresist masks usually contain inverse or negative photographic pattern that is to be transferred. Hence the exposed surface contains the negative resist while the unexposed areas are removed when dissolved in developer solution. Commonly used negative photo resists are SU-8, KMPR series, UVN-30 which require deep UV exposure etc [85].

7. MASKS FOR MICROFLUIDICS

Mask is a template to generate a desired pattern and resist the coated wafers. The pattern is made on mask using e beam lithography for high resolution patterns or CAD to make L edit or LASER plotter. The mask is kept in direct contact with the photoresist on exposure to UV light creating an image on wafer. The photolithographic masks are made up of three basic types of materials i.e. soda lime, quartz and polyester films. The polyester films have low resolution, economical and easy to microfluidic devices to 100 mm on a side. If the mold dissolves away while using once, then the topologies with multiple holes and 3d structures can be cast. The cast size must be able to accommodate the resolution limit of mask and illumination system [86]. Different materials can be used for mold formation to cast PDMS and different photo resists can be used for photolithography but mostly SU-8 is used. Mold fabrication method using SU-8 consist of following sequence.

1. SU-8 polymer photoresist after spin coating is poured onto a wafer. Ideally Si wafer is used with thickness of about 200 micro m.
2. Bubble trap, if any, is removed by degassing before use and heating it to 50-60 C.
3. Prebaking or soft bake of SU-8 is done to evaporate the solvent prepared for exposure.
4. Mount mask on top of SU-8 wafer and expose it with UV-rays of wavelength 350-400 nm. For thicker structure, the above steps are to be repeated.
5. Post exposure baking is done to aid the crosslinking of the exposed portion of SU-8 to be developed. Develop using the SU-8 developer which requires 16 minute for immersion development leaving the finished mold to be used in casting [88-89].

8. MOLDS

The mold formation requires a mask and light source in order to pattern a photosensitive resist polymer there by matching the features of the mask. The choice of mask depends on the required resolution by a simple laser printed with 1200 dpi or 250 micro m or

higher resolution print on thin polymer transparency film [87]. The high resolution masks are expensive and of several orders of magnitude thereby restricting the overall comes in contact with a glass or silicon overlays placed at top of poured PDMS with a weight placed at top of stack so that excess PDMS may be removed. Then the resultant is placed in an oven elevated at temperature of 650 °C for about 24 hours in order to cure the PDMS which is then peeled away from the mold and etched as desired. In some cases the solemnization of the mold is required in such cases the PDMS sticks to the SU-8 or Si [87].

9. MICROFLUIDIC APPLICATIONS

Microfluidic possesses several applications in almost every field of applied science and microelectronics and is shown in Fig. 8. Some important microfluidic applications reported by several researchers are discussed in this section.

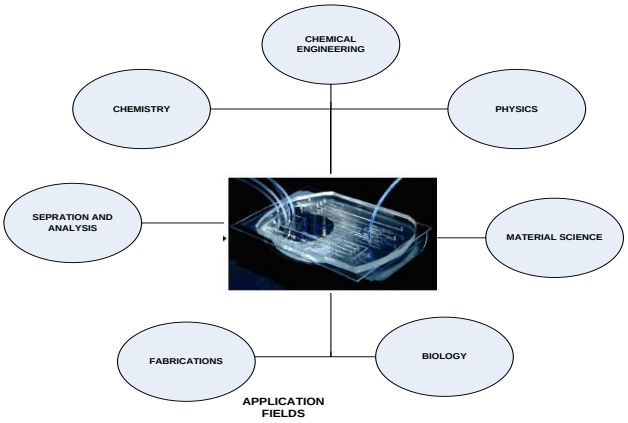

Fig. 8 – Different fields of applications of microfluids

9.1 Biomedicines

In biomedicines, the microfluidic technology is used for integrating many medical tests on a single chip employing lab on a chip application like blood tests, virus detection, separating detected virus and infections in blood [86]. These devices use the electrophoresis, DEP techniques to separate the different cell on the bases of their size and dielectric coefficients. Negative or positive DEP is used to separate the different micron particles, and cells. With decreasing assay times, reducing reagent requirements, and increasing sensitivity these also helps in enzyme assays. These systems are used to measure the functioning of liver transaminases [90] and reaction kinetics of enzyme galactosidase [91].

9.2 Protein Crystallization and Particle Tracking

Protein crystallization holds an important field in microfluidic applications that is used to generate various crystallization conditions such as temperature, humidity, pH etc. on a single chip. Some other interesting applications are drug screening, sugar testers, chemical micro reactor, micro fuel cells or microprocessor cooling systems [81]. Laminar flow is used to separate the fuel and its oxidant in fuel cells to control the interaction of the two fluids without a physical barrier as would be required in conventional fuel cells.

Particle detection and tracking is an efficient technique using microfluidics. Self initializing tracking tool for automated detection and tracking of particle trajectories from digital videos and video imaging in cell biology is major application that relies on low intensity fluorescence microscopy [86, 92-93].

9.3 Chemical Synthesis

Analyses of various chemical reactions in micro channel reactors make it applicable in the field of micro process engineering. The use of micro reactors increases the energy efficiency, reaction speed and improves the reliability, scalability and processing control thereby increasing the demand production [86], [94]. As we can do the mixing of the two fluids at micron level, we can be able to produce the new chemicals.

9.4 Separation and Analysis

The analysis of chemical reactions requires the ensuing steps that can separate and identify the individual species from the mixture [86], [95]. This is done by electrophoresis which tends to induce the behavioral differences between the charged species under the influence of applied electric field [92] [96-97]. Fig. 9 shows the process of particle separation in microfluidics using DEP [98].

9.5 Single Cell Biology / Cell Analysis

Microfluidic technology is employed for studying the cell behaviors from single to multicellular organism level that too with precisely localized application of experimental conditions at microscopic level such as the laminar flow effects are used to provide the spatial control of liquid composition at sub cellular resolution in addition to control in temperature changes etc [86] [99]. Microfluidics can be used for the cell analysis. These devices are used for cytometry (to analyse and count cells) and also capable of counting cells of different size [92-112]. This also find its use in cell-based assays [100], [107-108], cellular biosensors [109-112].

9.6 Micro Droplets

Microdroplets are single micro-reactors used as biodetector. The laminar flow enables the generation of monodisperse droplets, leading to multiphase fluid flow which is important application in microfluidics such as

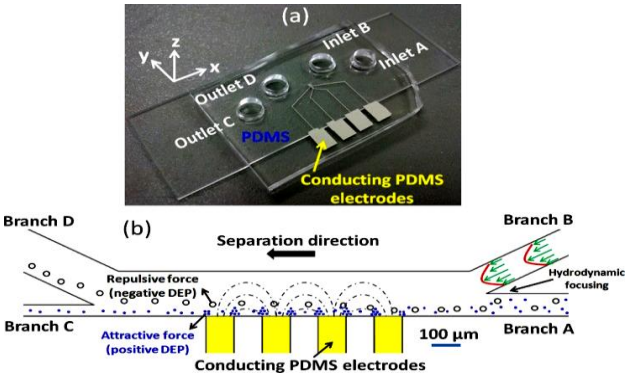

Fig. 9 – Separation of particles using DEP. From article [97]

emulsions for nano particle synthesis, drug micro encapsulation etc. Electro wetting can also be used to manipulate the discrete droplets on substrate that can be controlled independently in continuously flow system in digital microfluidics [86], [113]. The droplets can be generated easily by connecting submillimeter tubes to submillimeter T and cross junctions, thus providing an easy alternate way for production of droplets. Droplets can be made using a cross or *T* junction as chromatography tool employing a flow focusing or cross flowing methods [114]. The Micro Droplet Systems like optical twizzers enable users to produce more than 10,000 mono dispersed droplets per second ranging from 5μm to 250 μm in diameter. The different micro droplet chips arePressure-based Droplet Starter System, Syringe-based Droplet Starter System. Nanoparticles can be synthesized by microfluidics which plays a vital role from synthesis of silicon based fluorescent nanoparticles to label biomolecules for diagnostic assays, drug delivery, and cellular imaging as it leads to better efficiency and mixing [115-117].

9.7 Optofluidics

This technique involves the manipulation of light using fluids at micro or nano scale resulting in unique behavior of fluids. This is done by precisely controlling the optical properties of fluids to obtain the reconfigurable optical component [118-120].

9.8 Cell Deformation Using Microfluidic Chip

The shape of cell can be changed using microfluidic chip by forcing cell to enter in a small space. The factors are responsible for the growth of cells, gene expression or differentiation depends on the mechanical environment affected by the stress.

Thus their shape can be deformed and this deformation can be controlled by controlling shear stress to cells at single level which can be done by using microfluidic chip. This dynamically helps in cell squeezing with controlled pressure and force cells to grow in a peculiar geometry of the microchannel used. For example, yeast or bacteria when entered in a micro channel are allowed to grow in a controlled stress condition of microchannel that acquires the shape accordingly. Takeuchi et al used agarose and PDMS microchambers and forced E.coli cells to grow in circular or sinusoidal shape [121]. Mechanical stress on cells can also be changed by changing the flow that shear stress generated by fluid on adherent cell stuck on substrate which further subjects the cell to either laminar flow or the extensional flow [122] [123]. Fig. 10 shows the deformation of cells using PDMS quake valve [124].

The other fields where the microfluidics is used are Genomics; Drug screening, Electro-osmotic micro pumps, Clinical diagnostics, Drug Delivery, electrowetting, biochips, tissue engineering . Microfluidic devices are also used for the cell culturing. The yeast cells and many more cells are cultures in such devices [125-127]. These also used in concentration gradients [128] and Synthesis of Functional Reaction Networks.

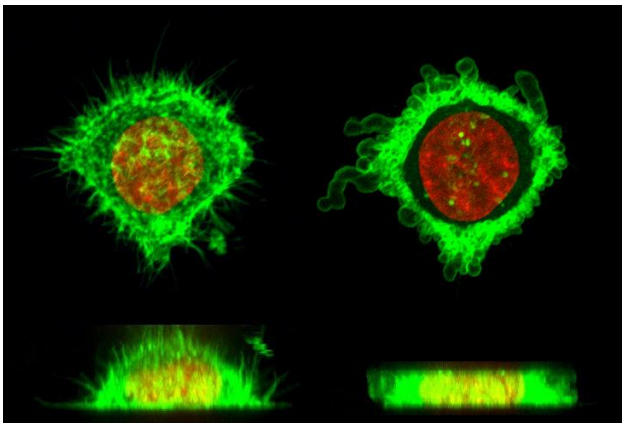

Fig. 10 – Hela cell deformation using PDMS quake valve. From article [124]

10. CONCLUSION

The microfluidics has taken the technology to the new scales. Several kinds of fluids are exploited to control their flow in the microchannels. The paper presents the review of the mechanics involved in the flow of the fluids in the channels. The devices are fabricated on the different substrates like silicon, glass, polymers or metals which are chemically inert, reusable, inert and biocompatible. The various techniques used for the fabrication of the devices are discussed and the type of material used also depends on the techniques used. The techniques of photolithography, micromaching, lab on CD make the prototyping of the devices easy and within reach of the researchers, biologists, and chemists. By knowing the basics of the fluid mechanics these devices are fabricated for different applications which has revolutionized the world of today with its reduced size, efficiency and portability. Upcoming era will be of microfluidics which provides many opportunities and challenges for the application of existing technologies in the micron scale and helps in solving many problems. It has already showed promising solutions in various fields and still has lot to offer, especially in the field of chemical biology. This will provide many opportunities in different areas of research and simulations. Innovations are awaited about the materials which can be used. As the devices discussed are feasible but there are some barriers in the field that have to be removed.

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