A new structure for microchip capillary electrophoresis with high velocity and resolution and efficiency

Seyyedeh Sepideh. Taheri Otaghsara[1], Bahram. Azizollah Ganji[2]

Morteza. Fathipur[3]

1- Department of Electrical & Computer Engineering, Babol noshirvani university of technology,Babol,Iran.
Email: sepidehtaheri@stu.nit.ac.ir
2- Department of Electrical & Computer Engineering, Babol noshirvani university of technology,Babol,Iran.
Email: baganji@nit.ac.ir(Corresponding author)
3- Department of Electrical & Computer Engineering, University of Tehran, Tehran, Iran.
Email:mfathi@ut.ac.ir

ABSTRACT:
Separation of charged particles is a major problem in biological and chemical researches. There are several techniques for separate charged particles, one of them is electrophoresis. electrophoresis separates electrically charged particles by applying external uniform electric field. In recent years, conventional cross-form of geometry has been used for microchip electrophoresis. this paper represents new cross-form geometry of microchip capillary electrophoresis with low separation time and high resolution and efficiency and are respectively equal to 100s,3.1,1166.5 while these values for conventional geometries are respectively equal to 120s,2.85,1004.8. these geometries are simulated by comsol.

1. INTRODUCTION

In the last two decades, the use of microfluidic platforms for chemical and biochemical applications has achieved remarkable progress [1-4]. separating species is an application of these devices. one method of separation is electrophoresis. electrophoresis is motion of electrically charged particle in a medium by applying external electric field [5]. Electrophoresis phenomenon first discovered by a Reuss in 1807. The capillary electrophoresis was performed in a capillary tube, but from 2001 expanded microchip capillary electrophoresis [6,7]. rapid separation, high efficiency, low electrolyte and sample consumption and low cost, are the main advantages of this method compared to others techniques. different geometries represent for microchip capillary electrophoresis, In 2005, chien hisung tsia and et al tested different geometries of micro channel and found cross-form of geometry better than other geometry because of less leakage during injection therefore it has high resolution[8]. Since then, the usual geometry for micro channel has been cross-form. this paper represents new cross-form geometry of microchip capillary

electrophoresis with low separation time and high resolution and efficiency.

2. THEORY OF ELECTROPHORESIS

Two major electro kinetic effects contribute in electrophoretic separation; electro-osmosis and electrophoresis. this section explains these two effects. electro-osmosis, the movement of liquid relative to a stationary charged surface induced by an applied electric field. Electrophoresis, the movement of suspended charged molecules relative to a stationary liquid induced by an applied electric force. Electrophoretic and electro-osmotic velocity are obtained respectively, according to Fig. 1 (a) and (c) according to Eq. 1 and 2[9].
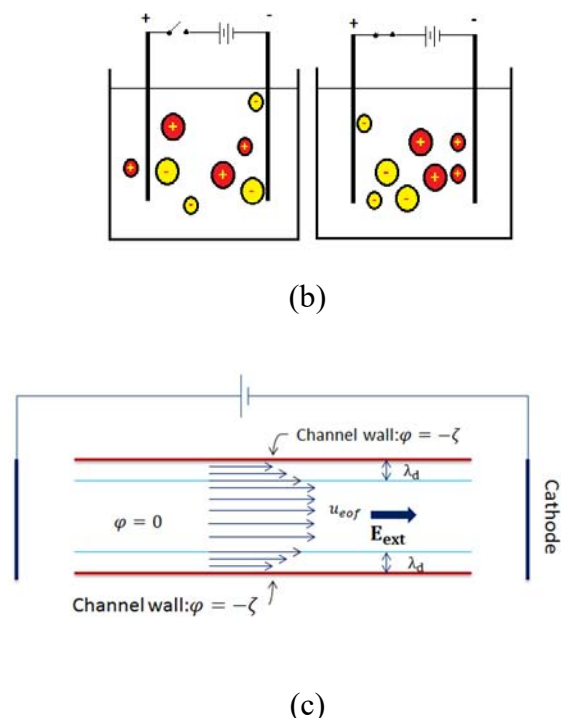

(a)

(b)

(c)
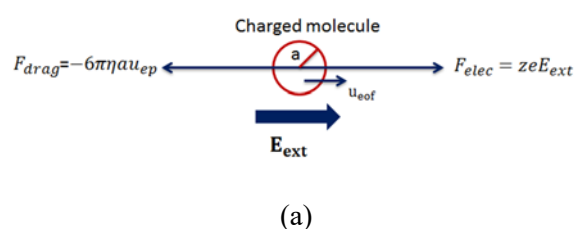
Fig. 5.(a) Force diagram of electrophoresis, **(b)**concept of relationship between electrophoretic velocity and size of charged particles, **(c)**electro-osmotic velocity[9].

For simplicity, a system of a spherical charged molecule of radius a and electric charge ze placed in a stationary liquid is considered. In order to involve only the electrophoresis, we assume that the electrical conductivity of the liquid is very low, so that the liquid does not flow while the external electric field is applied. The charged molecule could perceive an electric force, which causes it to move, and simultaneously experience a drag force as shown in Figure1(a) and obtain the electrophoretic velocity relation:

$$F_{total} = F_{elec} + F_{drag} = 0$$

$$zeE_{ext} = 6\pi\eta a u_{ep}$$

$$u_{ep} = \frac{ze}{6\pi\eta a}E_{ext} = \mu_{ep}E_{ext} \tag{1}$$

In Eq.1 u_{ep} , ze , a , η, E_{ext} and μ_{ep} respectively is electrophoretic velocity of the charged molecule

, electric charge of the molecule, molecule radius , viscosity of liquid, external electric field and absolute electrophoretic mobility of molecule.

according to Figures 1 (c), the electro-osmotic velocity can be defined as

$$U_{eo} = -\frac{\varepsilon\zeta}{\eta}E = \mu_{eo}E \tag{2}$$

ζ represents zeta potential, η denoted liquid viscosity, ε is dielectric coefficient of liquid.

The total velocity for charged particle in a medium by applying external electric field is obtained by Eq.3.

$$u_T = u_{ep} + u_{eof} = (\mu_{ep} + \mu_{eof})E_{ext} \tag{3}$$

As mentioned, microchips electrophoresis has several geometries, the most common one is cross-form. microchips electrophoresis have different methods for injection such as pressure, vacuum, electrokinetic and siphoning [10]. this paper uses gated injection of electrokinetic because This kind of injection used lower sample and easier to controlled [11].figure 2 illustrated gated injection.
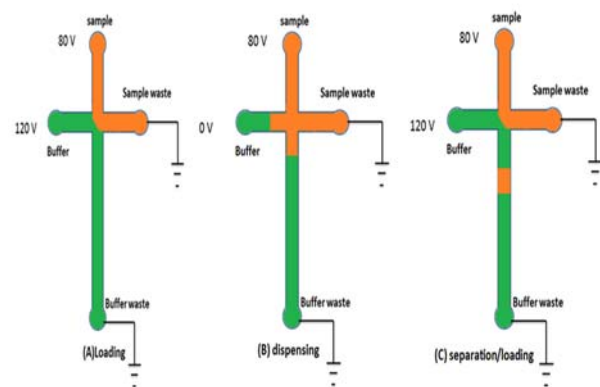

Fig. 2.the injection sequence for the gated injection

The gated injection has a loading/separation mode where the sample flows from sample reservoir to sample waste reservoir while the buffer flows from buffer reservoir to buffer waste reservoir to prevent sample leakage and provide continuous buffer

supply into the separation channel (Fig. 2,A). To make an injection, the field in buffer reservoir is set to zero allowing a plug of sample to move into the separation channel (Fig. 2, B). In the subsequent separation step, the field is switched back to the loading/separation step. The buffer flow cuts the sample plug, and the injection valve returns to its original state. explains the slight sample penetration into the buffer channel (Fig. 2,B) and nonsymmetric sample plug tails after dispensing (Fig. 2,C). The gated valve is capable of dispensing sample plugs of variable volume and provides unidirectional flow in the separation channel.

3. EQUATIONS OF MICROCHIP ELECTROPHORESIS

Fig.3 shows separation channel. In this model, H and L denote the width and length of the separation channel and E represents an applied electric field. according to fig. 3 equation of microchip electrophoresis is (i) the Poisson equation for the electric potential and zeta potential for the fluid-solid boundary, (ii) the Nernst-Planck equations for the positive and negative ionic concentrations, (iii) the full Navier-Stokes equations modified to include the effects of the body force due to the electric and charge density, and (iv) a sample concentration equation for the sample plug distribution.
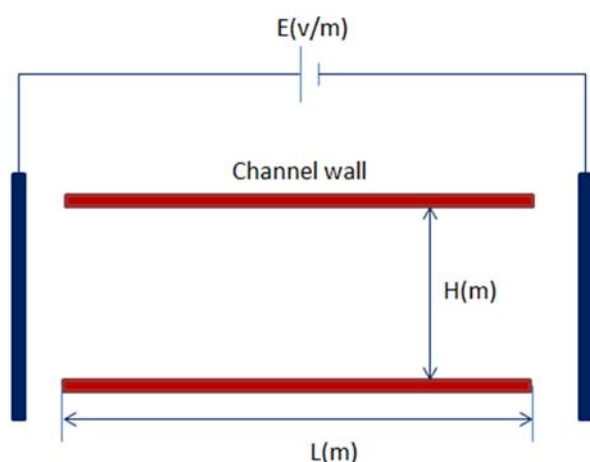

Fig. 3.sepration channel to describe equation[8-10]

3.1. Poisson equation

According to electrostatics theory, the relationship between the electric potential and the net charge density per unit volume is described at any point in the solution by the dimensionless Poisson equation [12-14]:

$$\nabla^2(\psi + \phi) = \frac{K^2}{2}(n^+ - n^-) \tag{4}$$

ψ represents Electric potential of electrical double layer, ϕ denote external electric potential of the electrodes , n^+, n^- are local concentration of positive and negative ions.
where k = K=KH, in which H is the width of the channel and $k^{-1} = (\frac{\varepsilon k_b T}{2n_0 z^2 e^2})^{0.5}$ is the Debye-Hückel parameter. The term '1/K' represents the characteristic thickness of the net charge density, n_0 denotes the bulk concentration of the ions, k_b is the Boltzmann constant ,z,e,ε are respectively valence of ions, electron charge, dielectric constant and T indicates the absolute temperature.

3.2. Nernst-Planck equation

Nernst-Planck equation describes the ionic concentration distributions, n^+ and n^-. Under the assumptions of no chemical reactions, incompressible flow and a constant diffusion coefficient, this equation can be expressed in the following dimensionless form[12-14]:

$$\vec{\nabla}.\left(\vec{V}n^+\right) = \frac{\{\nabla^2 n^+ \vec{\nabla}.[n^+(\vec{\nabla}\psi + A\vec{\nabla}\phi)]\}}{Re\ Sc^+} \tag{5}$$

$$\vec{\nabla}.\left(\vec{V}n^-\right) = \frac{\{\nabla^2 n^- \vec{\nabla}.[n^-(\vec{\nabla}\psi + A\vec{\nabla}\phi)]\}}{Re\ Sc^-} \tag{6}$$

Where v is velocity $Sc^{\pm} = \frac{\mu}{\rho D^{\pm}}$ indicates the Schmidt number ,and $Re = \frac{\rho U_{ref} L}{\mu}$ denotes the Reynolds number, in which D is diffusion coefficient of ions μ represents the liquid viscosity, ρ is the density of the fluid, and $U_{ref} = \frac{\varepsilon E_{ref}\zeta}{\mu}$, where $E_{ref} = \frac{\phi_{inlet}}{L}$ and ϕ_{inlet} expresses the activated potential at the inlet and ζ is the surface zeta potential.

$A = \frac{E_{ref}H}{\frac{k_bT}{Z}}$ Is a dimensionless parameter, which represents the ratio of the applied voltage and basis voltage.

3.3. Navier-Stokes equations

The Navier-Stokes equations describe the fluid flow in general cases. However, in the case of micro channel flows, these equations must be modified to take account of the electrical force generated by the interaction between the net charge density and the electric potential field. The equations of movement for an incompressible liquid can be expressed in the following dimensionless form:

$$\vec{\nabla}.(\vec{V}.\vec{V}) = -\vec{\nabla}P + \frac{1}{Re}\nabla^2\vec{V} - B\rho e(\vec{\nabla}\psi + A\vec{\nabla}\phi) \quad (7)$$

Where $B = n_0 k_b T/\rho U_{ref}^2$ Is a dimensionless parameter, which represents the ratio of the ion pressure and the dynamic pressure. v is velocity and Re denotes the Reynolds number.

3.4. Equation of Concentration

To ease a comprehensive understanding of the sample flow distribution in a micro channel, it is essential to solve the sample diffusion equation through a process of numerical simulation. The flow of the sample distribution is gained by solving the following nondimensional diffusion equation[12]:

$$\vec{\nabla}.\vec{\nabla}C = \frac{1}{ReSc}\nabla^2 C \quad (8)$$

In which C is the sample concentration and *SC* indicates the Schmidt number ,and Re denotes the Reynolds number.

4. IMPORTANT FACTORS IN SEPARATION

Three major factors in separation is (1)separation time, (2)resolution, (3)efficiency. Separation time refers to the time at which the concentration of ions are separated to defined other factors see fig.4. Resolution can be represents as the ratio of the distance between sample peaks and the standard deviation of the sample zones[15].

$$R = \frac{\Delta L}{\sigma} \quad (9)$$

Plate models for electrophoretic separations are used very frequently for overall system-to-system comparisons of separation efficiency. Plate models have historically been used for describing stage-wise distillation columns[16].
NTP called the number of theoretical plates, can be defined

$$NTP = (\frac{L_d}{\sigma})^2 \quad (10)$$

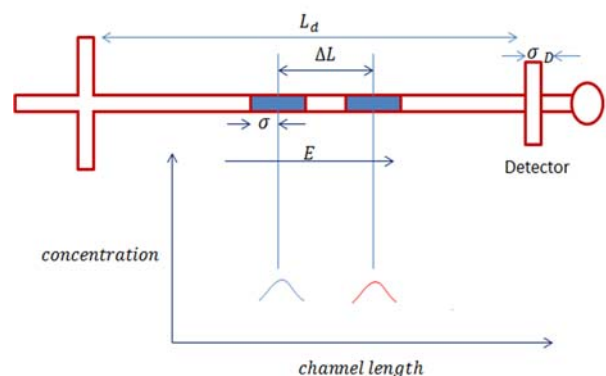The efficiency of separation is typically said to be higher for systems with higher plate number.

Fig.4. separation *channel* to defined resolution and efficiency[15].

According to fig.4 $\Delta L, \sigma$ and L_d are respectively the distance between sample peaks , the standard deviation of the sample zones and distance between cross section and detection point.

5. MICROCHIPS DESIGN

In this paper represents new cross-form geometry of microchannel and compared with conventional cross-form geometry of microchannel in the same conditions : microchip and electrode length are respectively 45mm and 50 μm,buffer is mes/his ,sample is k^+ and Na^+ and Li^+(200 μmol) with ionic radius that are respectively 152, 116 ,90 pm, gated injection ,applied voltage between buffer and buffer waste reservoir is 120 v and applied voltage between sample and sample waste reservoir is 80 v(shown in fig.2).
According to fig.5.(a) conventional cross-form geometry of microchannel ,all channel width are 50

μm and in fig.5.(b) new cross-form geometry of microchannel assume separation channel width is 100 μm and other channels are 50 μm .according to eq.5 and eq.7 when assume larger channel ,sample and buffer distribution move quickly. So separation time decrease and resolution and efficiency increase.
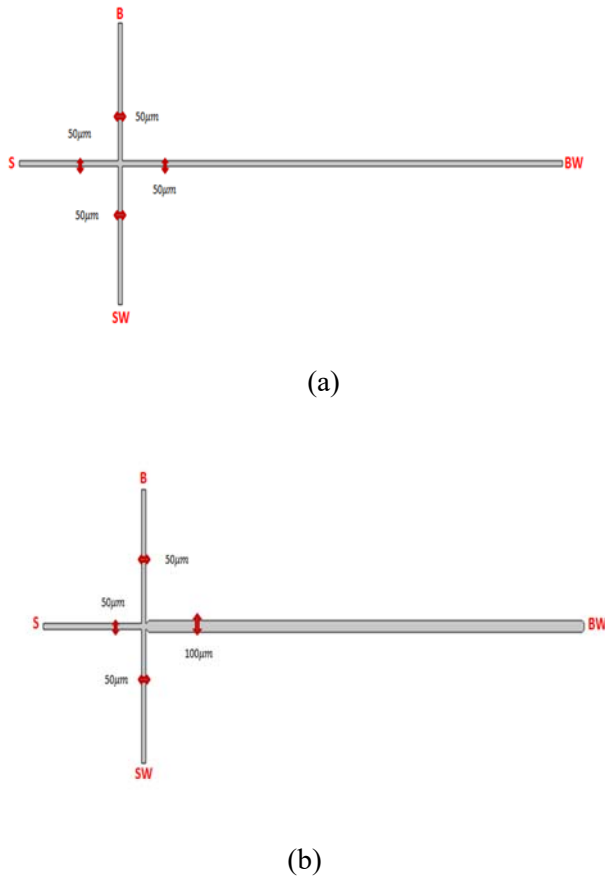

(a)

(b)

Fig.5.(a) conventional cross-form geometry **(b)** new cross-form geometry of microchannel
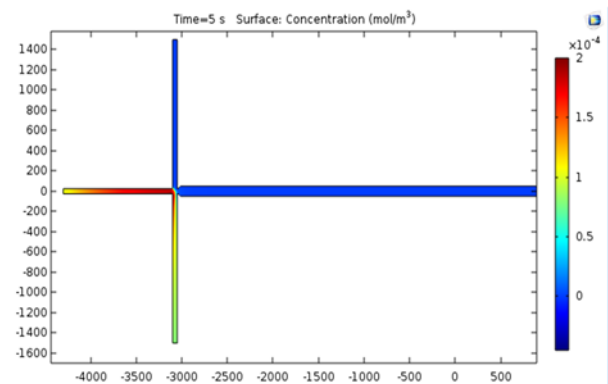
Fig.5 represents geometry of microchannel where S, Sw, B, BW are respectively sample, sample waste, buffer and buffer waste reservoir. injection sequence shown in fig.2.

6. RESULT AND DISCUSSION
These microchips are simulated by Comsol and the Eqs.(4-8) are used in modules of this software.Fig.6 is shown the result of injection method in two different geometries :
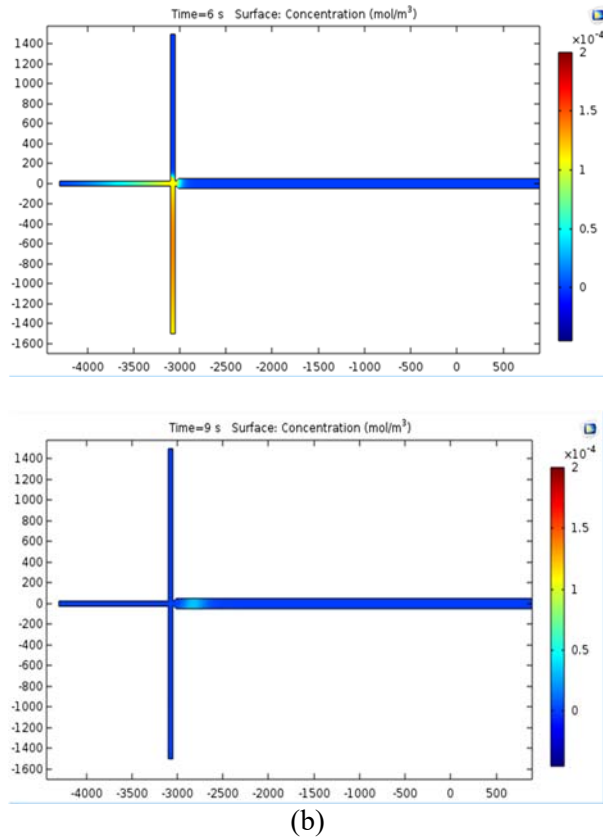

(a)

(a)

(b)
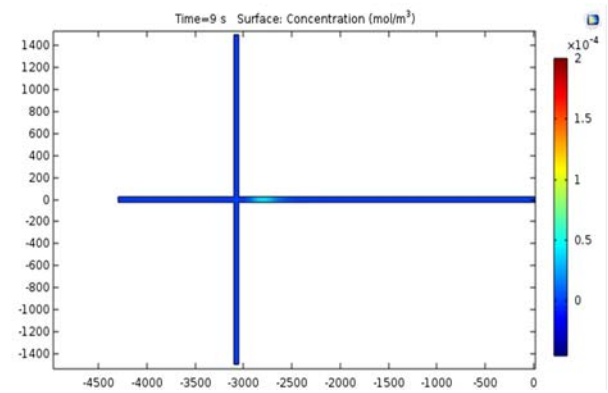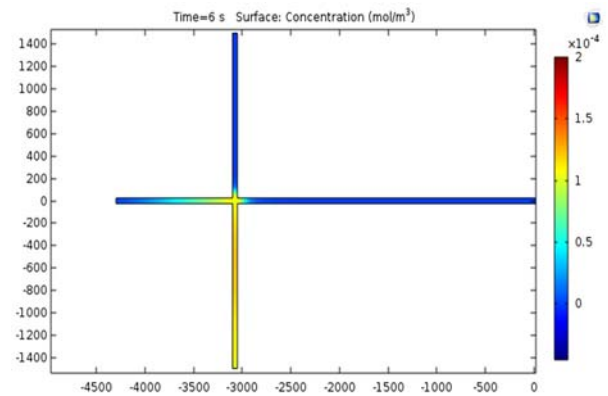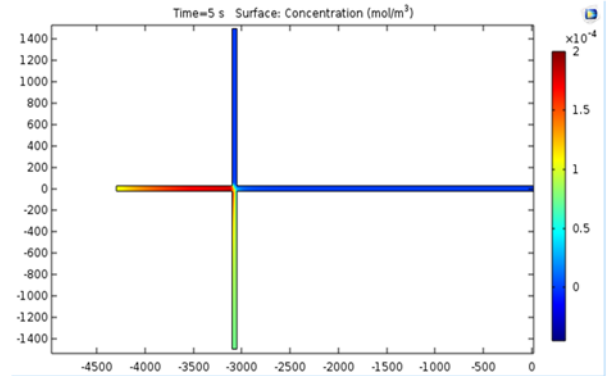
Fig.6. gated injection in **(a)** conventional cross-form geometry **(b)** new proposed cross-form geometry.

According to Fig. 6 gated injection process is performed as shown in Fig. 2. Fig. 6 (a) represents the concentration of the sample is injected into the channel in the conventional geometry and Fig. 6 (b) represents the concentration of the sample is injected into the channel in the proposed geometry and also is shown when samples inject into separation channel ,because of the larger separation channel, it faced less resistance and flow quicker.

Separation time refers to the time at which the concentration of ions are separated.Fig.7shows separation time of each studied geometry.
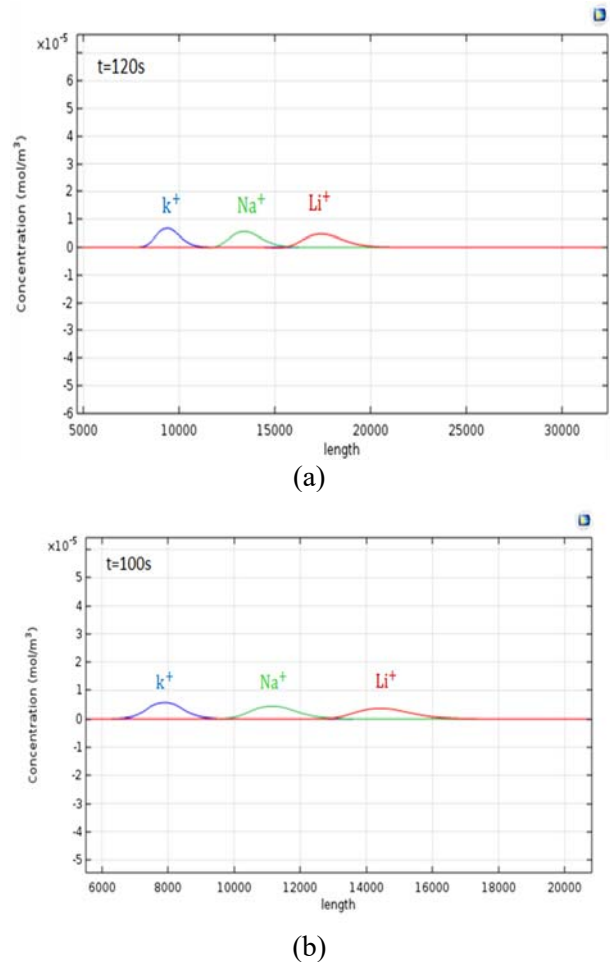

(a)

(b)

Fig.7.concentration as function of channel length at different time in the**(a)** conventional cross-form geometry **(b)** new proposed cross-form geometry.

According to fig.7 separation time for conventional cross-form geometry is 120s and for new proposed cross-form geometry is 100s. new proposed cross-form geometry separated samples in lower time than conventional cross-form geometry. because of the larger separation channel and according to eq.5and eq.7 sample and buffer distribution move quickly . also this figure shows that electrophoretic velocity of Li^+ is greater than $Na^+$and Na^+ is greater than k^+ because according to table1 ionic radius of Li^+ is smaller than $Na^+$and Na^+ is smaller than k^+,so according to eq.1 electrophoretic velocity of Li^+ is greater than $Na^+$and Na^+ is greater than k^+.

To obtain resolution and efficiency ,consider concentration as function of channel length at the same time (120s) for each geometry and according

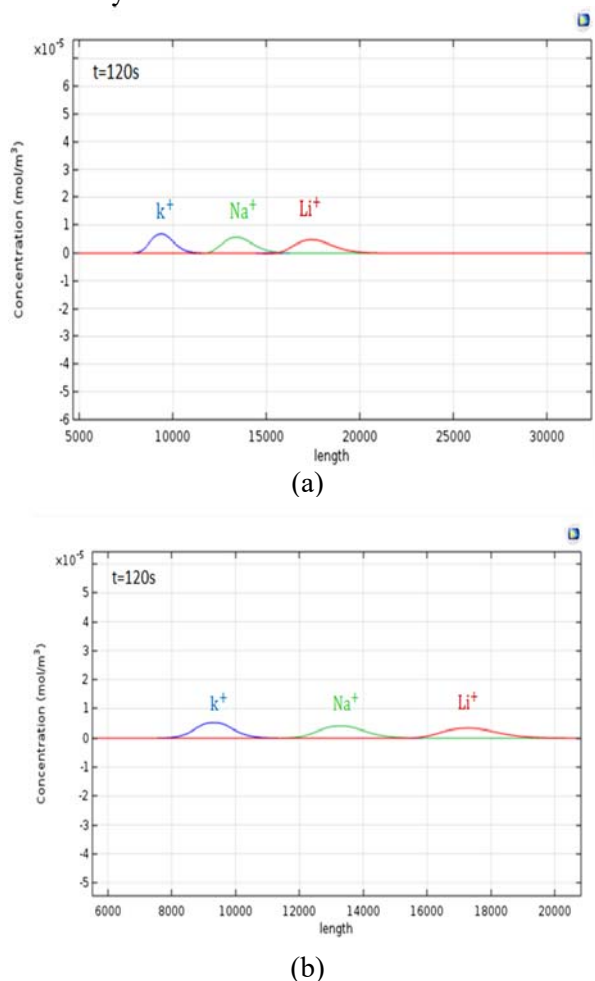to fig.8 and eq.9 and 10 ,calculate resolution and efficiency.

(a)

(b)

Fig.8. concentration as function of channel length at 120s in the **(a)** conventional cross-form geometry **(b)** new proposed cross-form geometry.

according to fig.8 and eq.9 and 10 , resolution and efficiency for conventional cross-form geometry is equal to 2.85 and1004.8 and new proposed cross-form geometry is equal to 3.1 and1166.5.so resolution and efficiency for new proposed cross-form geometry is greater than conventional cross-form geometry. Because in the same conditions of applied voltage and channel length when separation time decreases, resolution and efficiency increases.

Table 1. Comparison of results

Geometry of channel	conventional cross-form	new proposed cross-form
Separation time	120s	100s
Resolution	2.85	3.1
efficiency	1004.8	1166.5

According to table 1 in new proposed cross-form geometry, separation time decreases and resolution and efficiency increases.

CONCLUSION

electrophoresis separate electrically charged particles by applying external uniform electric field. in biological and chemical researches speed and resolution of separation is very important.so this paper represented new cross-form geometry witch has higher speed and resolution than conventional cross-form geometry .in proposed geometry ,separation time and resolution and efficiency is respectively equal to 100s,3.1 and 1166.5.

REFERENCES

[1] P. S. Dittrich, K. Tachikawa and A. Manz, **"Micro total analysis systems. Latest advancements and trends, "** *Analytical Chemistry*, vol.78,pp.3887,jun. 2006.

[2] W. K. T. Coltro, E. Piccin, E. Carrilho, D. P. de Jesus, J. A. F. da Silva, H. D. T. da Silva and C. L. do Lago, Quim, **"A toner-mediated lithographic technology for rapid prototyping of glass microchannels, "** *lab on chip*, vol.30, pp.1986, Nova. 2007.

[3] J. West, M. Becker, S. Tombrink and A. Manz, **" Micro total analysis systems. latest achievements, "** *Analytical Chemistry*, vol.80,pp.4403 , 2008.

[4] A. Arora, G. Simone, G. B. Salieb-Beugelaar, J. T. Kim and A. Manz, **" Latest developments in micro total analysis systems,** *Analytical Chemistry*, vol. 82, pp.4830, 2010.

[5]DongqingLi, **"Electrokinetics in Microfluidics, "**, *ELSEVIER* ,pp. 28, 2004.

[6]Laugere, F., Guijt, R.M., Bastemeijer, J., Van Der Steen, G.,Berthold, A., Baltussen, E., Sarro, P., Van Dedem, G.W.K.,Vellekoop, M.J., and Bossche, **"On-chip contactless four-electrode conductivity detection for capillary electrophoresis devices, "** *Analytical Chemistry*,vol. 75,pp.306–312, Jan .2003.

[7]Berthold, A., Laugere, F., Schellevis, H., de Boer, C.R.,Laros, M., Guijt, R.M., Sarro, P.M., and Vellekoop, M.J **"Capillary electrophoresis and microchip capillary electrophoresis "**, *Electrophoresis* ,vol.22,pp. 3511–3519,2001.

[8]Chien-HsiungTsai , Ruey-Jen Yang,Chang-Hsien Tai , Lung-Ming Fu, **"Numerical simulation of electrokinetic injection techniques in capillary electrophoresis microchips, "**, *electrophoresis*,vol.26,pp.674-686, 2005.

[9]chiang , **"principle of electrophoresis, "**, mai university,pp.13-17,1994.

[10]Marko stojkovic, Peter C. Hauser, Edwin C. Constable , **"Further Development and Applications of Capillary Electrophoresis with Capacitively Coupled ContactlessConductivity Detection and Sequential Injection Analysis in Analytical Chemistry, "**,pp.13,2013.

[11]Sergey V. Ermakov, Stephen C. Jacobson, and J. Michael Ramsey, **"Computer Simulations of Electrokinetic Injection Techniques in Microfluidic Devices, "**, *Analytical Chemistry*, pp.3512-3517,2000.

[12]H. Niazmand, J. Jamaati, M. Renlsizbulut, **" Investigation of electrokinetic mixing in 3D non-homogenous microchannels, "** *Journal Of Computational And Applied Research In*

Mechanical Engineering,Vol. 3, No. 1, pp. 41-52, 2013.

[13]J.Jamaati, A. R. Farahinia, H. Niazmand, **" Numerical Investigate of Electroosmotic Flow in Heterogeneous Microchannels, "**, *ModaresMechanical Engineering*,Vol. 15, No. 3, pp. 260-270,2015.

[14]J. H. Masliyah , **"Electrokinetik transport phenomena, "** *Alberta Oil Sands Tecnology and Research Authority*, 1994.

[15]Rajiv Bharadwaj, Juan G. Santiago,Bijan Mohammadi, **"Design and optimization of on-chip capillary electrophoresis, "** *Chemical Engineering Department*, *electrophoresis*,pp.2729-2731, 2002.

[16] McCabe, L. W., Smith, J. C., Harriott, P., **" Unit Operations of Chemical Engineering, "**, *McGraw-Hill Book Co., Singapore*, pp.521–587, 1993.