Enhancement of surface adsorption-desorption rates in microarrays invoking surface charge heterogeneity

Mojtaba Abdollahzadeh^a, Mohammad Said Saidi^a, Arman Sadeghi^{b,1}

^aCenter of Excellence in Energy Conversion (CEEC), School of Mechanical Engineering, Sharif University of Technology, Tehran

11155-9567, Iran

^bDepartment of Mechanical Engineering, University of Kurdistan, Sanandaj 66177-15175, Iran

ABSTRACT

This investigation is devoted to the influences of non-uniform wall characteristics on the surface adsorption-desorption rates in an electrokinetic microarray. Utilizing already explored electroosmotic and electrophoretic velocities, the species transport equations are solved by a finite-volume-based numerical approach. Uniform, sinusoidal, and pulse-like distributions of the zeta potential are considered in the analysis. The developed model is validated by comparing the results with those of two analytical solutions that are derived for limiting conditions. The results reveal that, in some cases, the surface charge heterogeneity can reduce the saturation time by more than 60%. The efficacy of the surface charge heterogeneity is limited to high Damkohler numbers and its effects are negligible for small values of this parameter. Whereas the impact of non-uniform surface characteristics is amplified by increasing the Peclet number, it is reduced by increasing either of the channel length to radius ratio or the electrophoretic mobility of the analytes.

Keywords: Electroosmotic flow; Microarray; Mass transport; Surface reaction; Numerical modeling

---

¹ Corresponding author

E-mail addresses: abdollahzadehm@alum.sharif.edu (M. Abdollahzadeh), mssaidi@sharif.edu (M. S. Saidi), a.sadeghi@eng.uok.ac.ir (A. Sadeghi)

1. Introduction

Recent advances in microfabrication technology have led to the wide development of different microfluidic-based instruments such as the lab-on-a-chip (LOC) devices. LOCs are instruments that integrate several laboratory functions on a single chip of only millimeters to a few square centimeters in size [1]. Broadly regarded as the biological laboratories of the future, LOCs provide several advantages over the present technologies comprising low sample and reagent consumption, eliminating human errors, performing multiple tests simultaneously, and faster analyses. Among the various LOCs being developed are different microarrays including DNA microarrays, protein microarrays, peptide microarrays, antibody microarrays, etc. A typical microarray is a 2D array on a solid substrate that is capable of assaying large quantities of biological samples using high-throughput screening, processing, and detection techniques. As it is usually the case, a carrier fluid is used to bring the samples close to the binding sites immobilized on the surface.

Microarrays were first introduced by Chang in 1983 [2] and started to grow significantly after the 1995 paper by Schena et al. [3]. The first modeling of surface reactions in microarrays was conducted by Glaser [4]. He numerically investigated the adsorption and desorption kinetics between a soluble analyte and immobilized binding sites on a surface. A contemporary study by Yarmush et al. [5] dealt with both theoretical and experimental analyses of biospecific interactions between one species in a flowing solution and another species immobilized in a thin hydrogel instrument using a BIAcore instrument. Shortly thereafter, Myska et al. [6] extended the range of rate constants available from BIAcore by making use of simulated data sets. Mason et al. [7] were able to investigate both the theoretical basis and numerical accuracy of the already developed models for determining reaction rates of biomolecules in biosensors.

More recently, Erickson et al. [8] proposed a general kinetic model of heterogeneous hybridization and developed a technique for estimating the kinetic constants for spatially resolved biochips. The experimentally verified model was then used to numerically model the dynamic hybridization on a biochip surface in the presence of a temperature gradient. Subsequently, a diffusion-reaction model was suggested by Gadgil et al. [9] for DNA microarray assays. Kim et al. [10] experimentally investigated the influence of volumetric flow rate, flow velocity, complementary DNA concentration, channel height, and time on DNA hybridization kinetics. Gervais and Jensen [11] considered different regimes of diffusion and laminar flow convection coupled with surface reactions relevant to biochemical assays. They compared analytic solutions of concentration field with the predictions of 2D numerical simulations

under various operation regimes. Shortly afterward, Parsa et al. [12] studied the effect of volume- and time-based constraints on the capture of analytes in microfluidic heterogeneous immunoassays. Contemporarily, Squires et al. [13] developed a physically intuitive and practical understanding of analyte transport in biosensors. There are also more recent works dealing with different aspects of mass transport and surface reactions in microarrays [14-21].

The research works mentioned above are all pertinent to pressure-driven flow biochemical assays. Nevertheless, the electroosmotic-based microarrays are becoming more and more common. Electroosmosis refers to the movement of ionic liquids with respect to electrically charged surfaces under the application of external electric fields. The contact of the charged surface with the liquid leads to the creation of a region of net electric charge, termed electric double layer (EDL), adjacent to the surface. EDL includes an immobile layer stuck to the wall and an outer diffuse layer. The Coulomb force exerted to the ions within the diffuse layer makes them move and owing to the viscous drag a net flow is created. The pioneering studies on mass transport in electroosmotically actuated microfluidic biosensors were performed by Das et al. who conducted both analytical [22] and numerical [23] analyses of DNA hybridization in microchannels. Subsequently, the reaction kinetics of antigen-antibody binding in electrokinetic-based heterogeneous immunoassays was numerically investigated by Hu et al. [24]. The effects of protonation of histidine buffer in promoting DNA hybridization in electronically active microarrays were explored by Kassegne [25]. More recently, analytical solutions were presented by Sharma et al. [26] for species transport in microreactors in the presence of electroosmotic flow. The same analysis, but this time for a mixed electroosmotic/pressure-driven flow, was done by Subramaniam and Chakraborty [27]. In two recent studies, Sadeghi et al. conducted numerical modelings of analyte transport and their hybridization with the binding sites in electrokinetically actuated microfluidic biosensors considering both Newtonian [28] and shear-rate dependent [29] carrier fluids.

As mentioned before, one of the main advantages of the LOC devices is their shorter detection times. In this respect, several methods have been proposed for improving different LOC functions. Liu et al. [30] proposed a micromixing technique based on cavitation microstreaming principle in order to enhance the hybridization process in DNA microarrays. A contemporary research by Pappaert et al. [31] demonstrated that DNA microarray analysis may be enhanced using a shear-driven microchannel flow system. Utilizing microfluidic planetary centrifugal mixing, Bynum and Gordon [32] were able to increase the sensitivity and dynamic ranges of microarrays by a factor of ten. Among the more recent techniques being developed for the improvement of hybridization are utilizing the

convective transport [33], using piezoelectric microagitation[34], and taking advantage of isotachophoresis [35]. Some methods have also been proposed for a faster mixing of the samples in micromixers such as utilizing time-interleaved segmentation[36] and surface heterogeneity[37]. The latter included the introduction of oppositely charged surface heterogeneities to micromixer walls by which the authors reported up to 70% reduction in the mixing length. This technique here is invoked to increase the rate of hybridization in an electrokinetic-based microarray. Along this line, a circular microchannel is considered for which the electroosmotic velocity components are available for an arbitrary but axisymmetric distribution of the zeta potential under thin EDL limit [38]. Both sinusoidal and pulse-like axial distributions of the zeta potential are considered along with the uniform case. The surface adsorption-desorption mechanism is modeled using the first order Langmuir model and the analyte transport equation is solved utilizing a finite-volume-based numerical method. Analytical solutions are also presented for the concentration wave speed assuming a uniform zeta potential distribution. A complete parametric study is then performed revealing that the surface charge heterogeneity can significantly decrease the saturation time under certain conditions.

2. Mathematical model

We examine the mass transport and surface reactions in electrokinetic-based microarrays of uniform and nonuniform surface potential. The geometry considered, shown schematically in Fig. 1, is a circular microchannel of radius R and length L . The carrier fluid is assumed to be a liquid with constant physicochemical properties. In addition, although the zeta potential is allowed to vary along the channel axis, it is supposed not to change with time, implying that possible alterations of the surface properties due to the adsorption-desorption mechanisms are considered negligible. The probable variations of the surface reaction constants due to the surface charge heterogeneity are also neglected. Finally, the last assumption is that the EDL thickness is very small as compared to the channel radius.

2.1. Species velocity

The velocity of particles being dispersed into ionic liquids in the presence of an external electric field is generally composed of two components: electroosmotic velocity of the solution and the particle electrophoretic velocity relative to the liquid. Therefore, the solute velocity vector $\mathbf{u}$ is obtained from

$$\mathbf{u} = \mathbf{u}_{eo} + \mathbf{u}_{ep} \quad (1)$$

wherein $\mathbf{u}_{eo}$ and $\mathbf{u}_{ep}$ are the electroosmotic and electrophoretic parts of the velocity, respectively. In the problem considered, for which the applied electric field is along the channel and EDL is vanishingly small, the particle electrophoretic velocity is in the axial direction only and is given as [39]

$$u_{ep} = \frac{\varepsilon \zeta_p}{\mu} E \quad (2)$$

where ε stands for the permittivity constant of the solution, ζ_p represents the zeta potential of the particle, μ shows the dynamic viscosity, and E is the applied electric field. In order to evaluate the electroosmotic velocity field, attention should be given toward the fact that the solution is electroneutral and the EDL thickness has been neglected; as a result, no electrical force is exerted on the fluid outside of this layer. Moreover, the electroosmotic microflows are of typically very low Reynolds numbers, so the Stokes flow equations can be used to obtain the velocity distribution. Therefore, the equations governing the flow field including the momentum conservation and continuity equations are given as

$$\mu \nabla^2 \mathbf{u}_{eo} - \nabla p = 0 \quad (3)$$

$$\nabla \cdot \mathbf{u}_{eo} = 0 \quad (4)$$

wherein p represents the pressure. By neglecting the EDL thickness, the fluid at the wall can no longer be considered stationary. In this case, the velocity at the edge of EDL can be found from Eq. (5):

$$u_{eo,w} = -\frac{\varepsilon \zeta}{\mu} E \quad (5)$$

where ζ denotes the zeta potential of the surface. In 1985, Anderson and Idol [38] solved Eqs. (3) and (4) subject to the boundary condition (5) analytically by using the Fourier transform. They showed that, for an arbitrary axisymmetric distribution of the zeta potential, the radial and axial velocities become, respectively

$$u_{eo,r} = 2\pi(1 - r^2/R^2) \frac{r}{L} \sum_{m=1}^{\infty} m \left[-\bar{u}_c(m) \sin\left(\frac{2m\pi z}{L}\right) + \bar{u}_s(m) \cos\left(\frac{2m\pi z}{L}\right) \right] \quad (6)$$

$$u_{eo,z} = -\frac{\varepsilon\zeta_{av}}{\mu}E - 2(1 - 2r^2/R^2) \sum_{m=1}^{\infty} \left[\bar{u}_c(m) \cos\left(\frac{2m\pi z}{L}\right) + \bar{u}_s(m) \sin\left(\frac{2m\pi z}{L}\right) \right] \quad (7)$$

ζ_{av} in equation (7) represents the zeta potential averaged over the surface and $z = z - L/2$. To obtain $\bar{u}_s(m)$ and $\bar{u}_c(m)$, the ζ distribution function should be expanded by Fourier series as

$$\zeta = \sum_{m=1}^{\infty} \zeta_c(m) \cos\left(\frac{2m\pi z}{L}\right) + \zeta_s(m) \sin\left(\frac{2m\pi z}{L}\right) \quad (8)$$

Then, $\bar{u}_s(m)$ and $\bar{u}_c(m)$ will be found as

$$\bar{u}_c(m) = -\frac{\varepsilon\zeta_c(m)}{\mu}E \quad (9)$$

$$\bar{u}_s(m) = -\frac{\varepsilon\zeta_s(m)}{\mu}E \quad (10)$$

2.2. Mass transport

In a typical microarray, three components are involved in the reaction: analytes or targets, surface bound analytes, and the binding sites. The analytes, which are being injected into the channel and are transferred by the flow, are actually the only component in the buffer fluid and have a number concentration c expressed in units of m^{-3} . The binding sites are immobilized on the surface and their concentration, shown here by c_{s0} , is in m^{-2} . The analytes arriving near the surface by the diffusion and advection mechanisms may be captured by the binding sites after which are called surface bound analytes with a concentration of c_s . Under the problem assumptions, the equation governing the transport of analytes may be written as

$$\frac{\partial c}{\partial t} + \mathbf{u} \cdot \nabla c = D \nabla^2 c = D \left[\frac{1}{r} \frac{\partial}{\partial r} \left(r \frac{\partial c}{\partial r} \right) + \frac{\partial^2 c}{\partial z^2} \right] \quad (11)$$

in which D is the diffusion coefficient of the analytes. Among the four required boundary conditions of Eq. (11) three of which include: constancy of the axial concentration gradient at the outlet, symmetry conditions at the centerline, and finally the inlet concentration being equal to the injection concentration c_{in} . The remaining one is

the equality of the mass flux of the analytes at the surface and the rate of increase in the concentration of the surface bound analytes, that is

$$\frac{\partial c_s}{\partial t} = -D \frac{\partial c}{\partial r} \quad (12)$$

Besides the initial condition, which is usually considered as $c|_{t=0} = 0$, an understanding of $\partial c_s / \partial t$ is required for Eq. (11) to be solved. This quantity may be evaluated using the first order Langmuir adsorption model, given as

$$\frac{\partial c_s}{\partial t} = k_a c_w (c_{s0} - c_s) - k_d c_s \quad (13)$$

In Eq. (13), k_a and k_d are the association and dissociation rates, respectively, and c_w denotes the analyte concentration in the channel near the reactive wall. For generalization of the findings, the analyte transport equation is made dimensionless as

$$\frac{\partial \theta}{\partial t^*} + u_r^* \frac{\partial \theta}{\partial r^*} + \frac{1}{\alpha} (u_z^* - \beta) \frac{\partial \theta}{\partial z^*} = \frac{1}{Pe} \left[\frac{1}{r^*} \frac{\partial}{\partial r^*} \left(r^* \frac{\partial \theta}{\partial r^*} \right) + \frac{1}{\alpha^2} \frac{\partial^2 \theta}{\partial z^{*2}} \right] \quad (14)$$

which is subject to the following initial and boundary conditions

$$\theta|_{t^*=0} = \frac{\partial \theta}{\partial r^*} |_{r^*=0} = \frac{\partial^2 \theta}{\partial z^{*2}} |_{z^*=1} = 0 \quad (15)$$

$$\theta|_{z^*=0, t^*>0} = 1 \quad (16)$$

$$\frac{\partial \theta}{\partial r^*} |_{r^*=1} = -Pe \frac{\partial \theta_s}{\partial t^*} \quad (17)$$

The dimensionless parameters appeared in Eqs.(14) to (17) are of the following forms

$$\theta = \frac{c}{c_{in}}, \quad \theta_s = \frac{c}{c_{in} R}, \quad u^* = \frac{u_{eo}}{U}, \quad t^* = \frac{tU}{R}, \quad z^* = \frac{z}{L}, \quad r^* = \frac{r}{R}, \quad \alpha = \frac{L}{R}, \quad Pe = \frac{UR}{D}, \quad \beta = \frac{\zeta_p}{\zeta_{av}} \quad (18)$$

where Pe is the Peclet number and $U = -\varepsilon \zeta_{av} E / \mu$ is the average boundary velocity. The equation governing the rate of increase in the concentration of the surface bound analytes, that is Eq. (13), can also be expressed in dimensionless form as

$$\frac{\partial \theta_s}{\partial t^*} = \frac{\epsilon Da}{Pe} [\theta_w (\epsilon^{-1} - \theta_s) - K_D \theta_s] \quad (19)$$

where the Damkohler number Da , the relative adsorption capacity ϵ , and the kinetic equilibrium constant K_D are of the following forms

$$Da = \frac{k_a c_{s0} R}{D}, \quad K_D = \frac{k_d}{k_a c_{in}}, \quad \epsilon = \frac{c_{in} R}{c_{s0}} \quad (20)$$

Assuming no captured analytes at time zero, the following initial condition holds for the dimensionless rate equation

$$\theta_s|_{t^*=0} = 0 \quad (21)$$

Because of the complex nature of the mass transport equations, especially the coupling between the analyte conservation equation and the rate equation, analytical equations are generally not possible and we should invoke numerical methods. The next section discusses the elements of the numerical method considered.

3. Numerical procedure

We make use of a finite-volume-based numerical method for handling the mass transport mechanism. The grid system is considered uniform and composed of square meshes since, due to the chaotic nature of the flow, large gradients may exist everywhere, even away from the wall. The discretized form of Eq. (14), after being integrated over the control volume and using the first-order upwind differencing scheme for the advection terms along with the central differencing scheme for the diffusion terms, may be written as

$$a_p \theta_{i,j}^{t+1,k+1} = \sum_{nb} a_{nb} \theta_{nb}^{t+1,k} + S_i^{t+1,k+1} + a_p^0 \theta_{i,j}^{t,k} \quad (22)$$

where the superscript t denotes the time step and k represents the iteration number in each time step, while indices i and j stand for the grid numbers in the z and r directions, respectively. Furthermore, $S_i^{t+1,k+1}$ is the sink term appeared due to the surface reactions and, therefore, appears only for the control volumes located at the wall. By relating the mass diffusion at the north side of the boundary cell $j_n^* = -\frac{1}{Pe} \frac{\partial \theta}{\partial r^*} |_{r^*=1}$ to the surface reaction rate using Eq. (17), $S_i^{t+1,k+1}$ is obtained as

$$S_i^{t+1,k+1} = -\frac{\theta_{s,i}^{t+1,k+1} - \theta_{s,i}^t r_n^* \Delta z^*}{\Delta t^* \alpha} \quad (23)$$

For evaluation of the sink term, $\theta_{s,i}^{t+1,k+1}$ should be calculated first. From the discretized form of Eq.(19), $\theta_{s,i}^{t+1,k+1}$ becomes

$$\theta_{s,i}^{t+1,k+1} = \frac{Pe\theta_{s,i}^t + Da\Delta t^* \theta_{w,i}^{t+1,k}}{Pe + \epsilon Da K_D \Delta t^* + \epsilon Da \Delta t^* \theta_{w,i}^{t+1,k}} \quad (24)$$

Since we make use of an implicit time-differencing method, an iterative mechanism should be invoked to handle the computations. Through this line, first $\theta_{s,i}^{t+1,k+1}$ should be obtained from Eq. (24) utilizing the data of the previous iteration (guessed data for the first iteration). Afterward, $\theta_{s,i}^{t+1,k+1}$ is substituted into Eq. (23) for $S_i^{t+1,k+1}$ to be evaluated. Having the values of the sink term in hand, the set of equations (22) is solved by taking advantage of the Tri-Diagonal Matrix Algorithm (TDMA). The computations are continued until the maximum relative difference between the results of two successive iterations becomes less than 10^{-6} .

4. Method validation

To ensure the validity of the simulations conducted, both the grid and time step dependency analyses should be performed. In this respect, by keeping $\alpha = 2$, three different grid systems composed of 50×100 , 100×200 , and 200×400 grid points along with three time steps of 10^{-6} , $\frac{1}{2} \times 10^{-6}$, and $\frac{1}{4} \times 10^{-6}$ were examined. It was observed that the results obtained utilizing a 100×200 grid system along with a 10^{-6} time step are nearly the same as those predicted utilizing finer meshes and time steps. Hence, all the forthcoming data are obtained by making use of the above mentioned time step and a grid system of $100 \times (100\alpha)$. Besides the grid and time step dependency, a comparison with available experimental or theoretical data is required for validation of the simulations. Since such data are not available for the present physical problem, we here try to derive a simple approximate solution under limiting conditions. As is well-known, the parameter Da shows the ratio of the diffusion time scale to the chemical reaction timescale. Therefore, a system is reaction-limited if $Da \ll 1$ and is diffusion-limited if $Da \gg 1$. Accordingly, in the limit $Da \rightarrow 0$, there is no resistance against the transport of analytes and we may assume they

instantaneously reach the surface. So the species concentration near the wall can be approximated by that at the entrance, that is $\theta_w \cong 1$. Adopting this approximation, Eq. (19) reduces to

$$\frac{d\theta_{s,rl}}{dt^*} = \frac{\epsilon Da}{Pe} [\epsilon^{-1} - (1 + K_D)\theta_{s,rl}] \quad (25)$$

As such, the surface reaction rate equation is no longer coupled with the analyte transport equations and may be easily solved to yield

$$\theta_{s,rl} = \frac{1 - e^{-\frac{\epsilon Da}{Pe}(1+K_D)t^*}}{\epsilon(1 + K_D)} \quad (26)$$

The numerically computed values of θ_s considering a very small Damkohler number of 10^{-10} and the predictions of Eq. (26) are compared in Fig. 2. As seen in this figure, the two sets of results are in excellent agreement, revealing the validity of the numerical solution methodology. In addition to Eq. (26), another simplified solution may be derived for the mass transfer problem that can also be used for comparison with numerical results. This matter will be discussed more thoroughly in the results and discussion section.

5. Results and discussion

It was found in the previous analyses that six dimensionless factors including Da , K_D , α , ϵ , Pe , β as well as the zeta potential distribution govern the mass transport and surface reaction in an electrokinetic-based microarray. Here, a complete parametric study is performed to determine the extent to which each factor affects the surface reaction rates. Special consideration is given toward the differences between nonuniform and uniformly distributed zeta potentials. To this end, two heterogeneous surfaces with pulse-like and sinusoidal distributions of the zeta potential are considered. The sinusoidal distribution, which is usually considered in modelings of the surface inhomogeneities caused by the microfabrication techniques [40, 41], is given as

$$\zeta^* = \begin{cases} 1 + d \sin\left(\frac{n\pi z}{L}\right) & \text{for } z < 0.85L \\ 1 & \text{for } z \geq 0.85L \end{cases} \quad \text{where } d = 0.5, n = 40 \quad (27)$$

The pulse-like distribution also is considered to be of the same period and amplification as the sinusoidal distribution. This type of surface charge heterogeneity is the most-used pattern in laboratory investigations and may

be obtained by different methods such as a standard photolithography coupled by simple surface charge patterning techniques[42]. Both of the aforementioned distributions are illustrated in Fig. 3 so as to provide the readers with a better insight into the surface charge heterogeneity patterns considered. As observed, the last 15% of the channel length has been assumed to have a uniform zeta potential; this is essential since otherwise the problem physics do not allow the vanishing of $\partial^2\theta/\partial z^{*2}$ at the outlet, which is required as per Eq. (15).

When the zeta potential distribution at the surface is not uniform, the flow patterns will distort and will no longer be uniform. Fig.4 shows the flow patterns resulted from a sinusoidal distribution of the zeta potential. The chaotic flow observed in this figure results in a higher dispersion of samples. In particular, such flow patterns will increase the mass transport flux toward the wall and, consequently, decrease the time needed for the samples to reach the surface.

As the target molecules are captured by the binding sites, a depleted zone is formed adjacent to the surface. The analyte concentration within this zone increases as the distance from the wall increases, until the edge of the depletion layer where the concentration variation is negligible. The collection rate or the mass flux through this zone is inversely proportional to its thickness. When the zeta potential distribution is non-uniform, the chaotic flow will increase the reactants flux toward the wall, making the depletion zone thinner. Fig. 5 shows the analyte concentration contours for both the uniform and sinusoidal distributions of the zeta potential at $time^* = 3$. At this moment, a portion of the channel surface is saturated; therefore, the depletion zone is not started from the channel entrance. As is clear, the distance from the channel entrance to the starting point of the depletion zone is larger for the non-uniform distribution, implying a higher portion of the surface being saturated. Furthermore, one can see the effect of the chaotic flow caused by the surface charge heterogeneity on the depletion layer shape. The enhanced mixing of flow has decreased the depletion zone thickness in most of the channel. To better demonstrate this result, we have plotted the dimensionless depletion zone thickness, denoted by δ^* , along the channel surface for both of the zeta potential distributions in Fig. 6.

We now turn our attention to the graphs of θ_s versus z^* at different equidistant amounts of t^* , depicted in Fig. 7. Due to the more availability of the analytes at smaller axial positions, θ_s is a decreasing function of z^* . Because of the large channel length to radius ratio considered, the entrance region reaches saturation at a time that the reaction is not actually started at the channel outlet. Under these conditions, a concentration wave is established which moves in a fairly constant speed, saturating any point of the surface it reaches[11]. The speed of this wave can be obtained

analytically when the zeta potential distribution is uniform. For a uniform zeta potential, there will be a unidirectional plug-flow inside the channel,so

$$u_r^*(z^*, r^*) = 0 \quad , \quad u_z^*(z^*, r^*) = 1 \quad (28)$$

Adopting the above velocity components, Eq. (14) reduces to

$$\frac{\partial \theta}{\partial t^*} + \frac{1}{\alpha}(1 - \beta) \frac{\partial \theta}{\partial z^*} = \frac{1}{Pe} \left[\frac{1}{r^*} \frac{\partial}{\partial r^*} \left(r^* \frac{\partial \theta}{\partial r^*} \right) + \frac{1}{\alpha^2} \frac{\partial^2 \theta}{\partial z^{*2}} \right] \quad (29)$$

which upon integration over the channel cross-sectional area yields

$$\frac{\partial \theta_{csa}}{\partial t^*} + \frac{1}{\alpha}(1 - \beta) \frac{\partial \theta_{csa}}{\partial z^*} = \frac{1}{Pe} \left[2 \int_0^1 \frac{\partial}{\partial r^*} \left(r^* \frac{\partial \theta}{\partial r^*} \right) dr^* + \frac{1}{\alpha^2} \frac{\partial^2 \theta_{csa}}{\partial z^{*2}} \right] \quad (30)$$

The parameter θ_{csa} in Eq. (30) is the dimensionless analyte concentration averaged over the channel area and is, hence, given as

$$\theta_{csa} = 2 \int_0^1 \theta r^* dr^* \quad (31)$$

Taking advantage of the boundary conditions(15) and (17), the integral term in Eq. (30)can be rewritten as

$$\int_0^1 \frac{\partial}{\partial r^*} \left(r^* \frac{\partial \theta}{\partial r^*} \right) dr^* = \frac{\partial \theta}{\partial r^*} \Big|_{r^*=1} = -Pe \frac{\partial \theta_s}{\partial t^*} \quad (32)$$

Eq. (30), therefore, is modified into the following form

$$\frac{\partial \theta_{csa}}{\partial t^*} + \frac{1}{\alpha}(1 - \beta) \frac{\partial \theta_{csa}}{\partial z^*} = \frac{1}{Pe} \left(-2Pe \frac{\partial \theta_s}{\partial t^*} + \frac{1}{\alpha^2} \frac{\partial^2 \theta_{csa}}{\partial z^{*2}} \right) \quad (33)$$

It is possible to make both θ_{csa} and θ_s a function of one spatial parameter only by fixing the coordinate system to the wave front; this may be performed invoking a variable change of the form $Z = z^* - u_{eff}^* t^*/\alpha$ in which $u_{eff}^* = u_{eff}/U$ is the dimensionless form of the concentration wave velocity u_{eff} . The transformed form of Eq. (33) and the associated limiting conditions then become

$$-u_{eff}^* \frac{d\theta_{csa}}{dZ} + (1 - \beta) \frac{d\theta_{csa}}{dZ} = \frac{1}{Pe} \left(2Pe u_{eff}^* \frac{d\theta_s}{dZ} + \frac{1}{\alpha} \frac{d^2\theta_{csa}}{dZ^2} \right) \quad (34)$$

$$\theta_{csa}|_{Z^* \rightarrow +\infty} = \frac{d\theta_{csa}}{dZ}|_{Z^* \rightarrow +\infty} = \frac{d\theta_{csa}}{dZ}|_{Z^* \rightarrow -\infty} = \theta_s|_{Z^* \rightarrow +\infty} = 0 \quad (35)$$

$$\theta_{csa}|_{Z^* \rightarrow -\infty} = 1, \quad \theta_s|_{Z^* \rightarrow -\infty} = \frac{\epsilon^{-1}}{1 + K_D}$$

Integrating Eq.(34) with respect to Z from $-\infty$ to $+\infty$ with the consideration of the limiting conditions (35), the concentration wave velocity is obtained to be

$$u_{eff}^* = (1 - \beta) \left(1 + \frac{2\epsilon^{-1}}{1 + K_D} \right)^{-1} \quad (36)$$

Fig. 8 sketches u_{eff}^* as a function of the solute to surface zeta potential ratio β . The numerically computed values of the wave concentration speed are also shown by symbols, revealing a fairly good agreement between the two data sets.

In order to evaluate the extent to which a chaotic flow may influence the surface reaction kinetics, the relative difference between the saturation times of the uniform and non-uniform distributions of the zeta potential τ is defined as

$$\tau = \frac{t_{sat,U}^* - t_{sat,NU}^*}{t_{sat,U}^*} \quad (37)$$

wherein the dimensionless saturation time t_{sat}^* is considered to be the moment at which the relative difference between θ_s of two successive time steps for all the grid points becomes less than 10^{-6} . In Fig. 9, the relative saturation time difference for the sinusoidal distribution of the zeta potential is depicted as a function of Da at different Peclet numbers. The positive values of τ_{sin} reflect the fact that a chaotic flow reduces the saturation time. Furthermore, it is obvious that when Da is small the saturation time is not much dependent upon the zeta potential distribution; but for large values of this parameter the surface charge heterogeneity has a significant impact on the saturation time. This can be explained by noticing the system behavior for limiting cases of Da . As mentioned earlier, if $Da \ll 1$ the system is reaction-limited and the reaction timescale determines the saturation time. As a result, any improvement in analyte transport has no influences on the time required to reach equilibrium.

Nonetheless, when $Da \gg 1$ the system is diffusion-limited and the saturation time is determined by the timescale reactants need to reach the surface. Another point taken from Fig. 9 is that the surface charge heterogeneity effects are amplified by increasing Pe . This is not surprising by referring to the physical interpretation of the Peclet number which is the ratio of the advection to the diffusion of mass. As such, a higher Peclet number is accompanied by a pronounced influence of the velocity field on the analyte transport.

A higher α may be thought of as a larger channel length for a fixed radius. As a result, not only the area of the surface requiring hybridization will increase, but also the targets need more time to reach the points near the channel end. These, ultimately, give rise to a larger saturation time. In Fig. 10, this trend is noticeable for both uniform and non-uniform distributions of the zeta potential.

Fig. 10 reveals a slightly better efficiency for the pulse-like distribution as compared to the sinusoidal function. This may be attributed to the discontinuous nature of the pulse-like distribution which can create more chaotic flows. Fig. 10 also shows that the surface charge heterogeneity decreases the saturation time to a great extent when the length to radius ratio is small. For instance, when $\alpha = 2$, the reduction in the saturation time is more than 60%. It should be pointed out that whereas the difference between the uniform and non-uniform zeta potential distributions is an increasing function of α , quite the opposite is true for τ , mainly because t_{sat}^* is longer for larger channel length to radius ratios.

The last illustration, that is Fig. 11, shows the saturation time versus the zeta potential ratio β . As seen in this figure, a positive β leads to smaller reaction rates whereas the opposite holds for a negative value of this parameter. This occurs since a positive β corresponds to an electrophoretic velocity in the opposite direction of the mean electroosmotic velocity. In other words, electrophoresis leads to a decrease in the net velocity of the analytes. Accordingly, it will take longer for the analytes to get to the surface, leading to an increase in the saturation time. The reverse of the reasoning made is true for a negative β since the electrophoretic velocity in this case is favorable. Fig. 11 also illustrates that although the reduction in the saturation time due to the surface charge heterogeneity is achieved irrespective of β , it is maximal when $\beta = 0$, that is when the analytes are neutral or their electrophoretic mobility is negligible as compared to the electroosmotic mobility. For $\beta < 0$, the electrophoretic velocity is added to the electroosmotic part and, hence, reduces the chaotic nature of the flow since the radial velocity component remains unchanged. For a positive β , the reduction of the surface charge heterogeneity effects may be attributed to the longer reaction times due to the lower availability of the analytes in the channel which itself

is an outcome of lower axial velocities. Accordingly, despite the fact that a non-uniform zeta potential distribution still provides the same reduction in t_{sat}^* as for $\beta = 0$, the relative saturation time difference noticeably decreases.

At the end of the results section, it is worth noting that the influences of the parameters K_D and ϵ on the surface charge heterogeneity effects were found to be below 2% as long as the values within practical ranges were considered. Accordingly, the pertinent graphs are not shown here so as to save space. Clearly, these conclusions are pertinent to the case in which the only source of heterogeneity is the surface charge and may change to some extent by including other forms of surface heterogeneity such as position-dependent reaction constants.

6. Conclusions

The influences of surface charge heterogeneity on the mass transfer and surface reaction kinetics in an electrokinetic-based microarray were studied in this paper. Finite-volume-based numerical simulations of the species transport equations were conducted utilizing the already available electroosmotic and electrophoretic velocity components for the vanishing EDL limit. Two different non-uniform distributions of the zeta potential including the pulse-like and sinusoidal functions were considered along with a uniformly distributed zeta potential. The method considered was validated by comparing the concentration of the surface bound analytes with the predictions of the reaction-limited solution. The results showed that a chaotic flow caused by the surface charge heterogeneity may significantly increase the reaction speed by amplifying the analyte mass flux toward the binding sites. It was observed that the importance of the surface charge heterogeneity effects is severe for high Damkohler numbers; on the other hand, at small values of this parameter, for which the system is reaction-limited, creating a chaotic flow has no impact on the saturation time. Moreover, the influences of the surface charge heterogeneity are pronounced by decreasing the channel length to radius ratio and increasing the Peclet number. In addition, it was observed that the surface charge heterogeneity effects are maximal when the analytes are neutral and are not noticeably changed by varying the values of the relative adsorption capacity and the kinetic equilibrium constant. Last but not least, a concentration wave of constant speed was shown to be created at large channel length to radius ratios; an analytical solution was developed for the wave velocity by fixing the coordinate system to the wave front and was found to be consistent with the numerical results.

Acknowledgment

The corresponding author sincerely thanks the Iran National Science Foundation (INSF) for their supports during the course of this work.

References

- [1] Y.H. Ghallab, W. Badawy, Lab-on-a-Chip; Techniques, Circuits, and Biomedical Applications, Artech House, Norwood, MA, 2010.
- [2] T.-W. Chang, Binding of cells to matrixes of distinct antibodies coated on solid surface, *Journal of Immunological Methods*, 65 (1983) 217-223.
- [3] M. Schena, D. Shalon, R.W. Davis, P.O. Brown, Quantitative Monitoring of Gene Expression Patterns with a Complementary DNA Microarray, *Science*, 270(5235) (1995) 467-470.
- [4] R.W. Glaser, Antigen-antibody binding and mass transport by convection and diffusion to a surface: A two-dimensional computer model of binding and dissociation kinetics, *Anal. Biochem.*, 213(1) (1993) 152-161.
- [5] M.L. Yarmush, D.B. Patankar, D.M. Yarmush, An analysis of transport resistance in the operation of BIAcore(TM); Implications for kinetic studies of biospecific interactions, *Molecular Immunology*, 33(15) (1996) 1203-1214.
- [6] D.G. Myszk, X. He, M. Dembo, T.A. Morton, B. Goldstein, Extending the Range of Rate Constants Available from BIAcore: Interpreting Mass Transport-Influenced Binding Data, *Biophys. J.*, 75(2) (1998) 583-594.
- [7] T. Mason, A.R. Pineda, C. Wofsy, B. Goldstein, Effective rate models for the analysis of transport-dependent biosensor data, *Math. Biosci.*, 159(2) (1999) 123-144.
- [8] D. Erickson, D. Li, U.J. Krull, Modeling of DNA hybridization kinetics for spatially resolved biochips, *Anal. Biochem.*, 317(2) (2003) 186-200.
- [9] C. Gadgil, A. Yeckel, J.J. Derby, W.-S. Hu, A diffusion-reaction model for DNA microarray assays, *J. Biotechnol.*, 114(1-2) (2004) 31-45.
- [10] J.H.-S. Kim, A. Marafie, X.-Y. Jia, J.V. Zoval, M.J. Madou, Characterization of DNA hybridization kinetics in a microfluidic flow channel, *Sensor Actuat. B-Chem.*, 113(1) (2006) 281-289.
- [11] T. Gervais, K.F. Jensen, Mass transport and surface reactions in microfluidic systems, *Chem. Eng. Sci.*, 61(4) (2006) 1102-1121.
- [12] H. Parsa, C.D. Chin, P. Mongkolwisetwara, B.W. Lee, J.J. Wang, S.K. Sia, Effect of volume- and time-based constraints on capture of analytes in microfluidic heterogeneous immunoassays, *Lab Chip*, 8(12) (2008) 2062-2070.
- [13] T.M. Squires, R.J. Messinger, S.R. Manalis, Making it stick: Convection, reaction and diffusion in surface-based biosensors, *Nat. Biotechnol.*, 26(4) (2008) 417-426.
- [14] D. Mocanu, A. Kolesnychenko, S. Aarts, A. Troost-Dejong, A. Pierik, E. Vossenaar, H. Stapert, Mass transfer effects on DNA hybridization in a flow-through microarray, *J. Biotechnol.*, 139(2) (2009) 179-185.
- [15] M. Rober, J. Walter, E. Vlach, F. Stahl, C. Kasper, T. Tennykova, New 3-D microarray platform based on macroporous polymer monoliths, *Anal. Chim. Acta*, 644(1-2) (2009) 95-103.
- [16] S. Jomeh, M. Hoorfar, Numerical modeling of mass transport in microfluidic biomolecule-capturing devices equipped with reactive surfaces, *Chem. Eng. J.*, 165(2) (2010) 668-677.
- [17] B. Roy, T. Das, T.K. Maiti, S. Chakraborty, Effect of fluidic transport on the reaction kinetics in lectin microarrays, *Anal. Chim. Acta*, 701(1) (2011) 6-14.
- [18] R. Hansen, H. Bruus, T.H. Callisen, O. Hassager, Transient convection, diffusion, and adsorption in surface-based biosensors, *Langmuir*, 28(19) (2012) 7557-7563.
- [19] R.M. Winz, W. Wiechert, E. von Lieres, Surface bound adsorption in a microfluidic T-sensor: Numerical comparison and optimization of 2D and 3D models and of sensor designs, *Sensor Actuat. B-Chem.*, 170 (2012) 75-81.
- [20] Y. Liu, Y. Zhang, Z. Lu, C.M. Li, 3-D microarray and its microfabrication-free fluidic immunoassay device, *Anal. Chim. Acta*, 889 (2015) 187-193.
- [21] D. Rath, S. Panda, Correlation of Capture Efficiency with the Geometry, Transport, and Reaction Parameters in Heterogeneous Immunosensors, *Langmuir*, 32(5) (2016) 1410-1418.
- [22] S. Das, T. Das, S. Chakraborty, Analytical solutions for the rate of DNA hybridization in a microchannel in the presence of pressure-driven and electroosmotic flows, *Sensor Actuat. B-Chem.*, 114(2) (2006) 957-963.

- [23] S. Das, T. Das, S. Chakraborty, Modeling of coupled momentum, heat and solute transport during DNA hybridization in a microchannel in the presence of electro-osmotic effects and axial pressure gradients, *Microfluid Nanofluid*, 2(1) (2006) 37-49.
- [24] G. Hu, Y. Gao, D. Li, Modeling micropatterned antigen-antibody binding kinetics in a microfluidic chip, *Biosens. Bioelectron.*, 22(7) (2007) 1403-1409.
- [25] S. Kassegne, B. Arya, N. Yadav, Numerical modeling of the effect of histidine protonation on pH distribution and DNA hybridization in electronically active microarrays, *Sensor Actuat. B-Chem.*, 143(2) (2010) 470-481.
- [26] H. Sharma, N. Vasu, S. De, Mass transfer during catalytic reaction in electroosmotically driven flow in a channel microreactor, *Heat Mass Transfer*, 47(5) (2011) 541-550.
- [27] K. Subramaniam, S. Chakraborty, A semi-analytical model for species transport in combined electroosmotic and pressure driven microflows with surface adsorption-desorption reactions, *Microfluid Nanofluid*, 10(4) (2011) 821-829.
- [28] A. Sadeghi, Y. Amini, M.H. Saidi, S. Chakraborty, Numerical modeling of surface reaction kinetics in electrokinetically actuated microfluidic devices, *Anal. Chim. Acta*, 838 (2014) 64-75.
- [29] A. Sadeghi, Y. Amini, M.H. Saidi, H. Yavari, Shear-rate-dependent rheology effects on mass transport and surface reactions in biomicrofluidic devices, *AIChE J.*, 61 (2015) 1912-1924.
- [30] R.H. Liu, R. Lenigk, R.L. Druyor-Sanchez, J. Yang, P. Grodzinski, Hybridization Enhancement Using Cavitation Microstreaming, *Anal. Chem.*, 75(8) (2003) 1911-1917.
- [31] K. Pappaert, J. Vanderhoeven, P. Van Hummelen, B. Dutta, D. Clicq, G.V. Baron, G. Desmet, Enhancement of DNA micro-array analysis using a shear-driven micro-channel flow system, *J. Chromatogr. A*, 1014(1-2) (2003) 1-9.
- [32] M.A. Bynum, G.B. Gordon, Hybridization Enhancement Using Microfluidic Planetary Centrifugal Mixing, *Anal. Chem.*, 76(23) (2004) 7039-7044.
- [33] J.R. Pascault, H.S. Zhou, Enhancement of DNA hybridization kinetics in microarrays by convective transport, *Chem. Eng. Commun.*, 195(2) (2008) 167-186.
- [34] K. Rodaree, T. Maturos, S. Chaotheing, T. Pogfay, N. Suwanakitti, C. Wongsombat, K. Jaruwongrungee, A. Wisitsoraat, S. Kamchonwongpaisan, T. Lomas, A. Tuantranont, DNA hybridization enhancement using piezoelectric microagitation through a liquid coupling medium, *Lab Chip*, 11(6) (2011) 1059-1064.
- [35] C.M. Han, E. Katilius, J.G. Santiago, Increasing hybridization rate and sensitivity of DNA microarrays using isotachopheresis, *Lab Chip*, 14(16) (2014) 2958-2967.
- [36] N.-T. Nguyen, X. Huang, Mixing in microchannels based on hydrodynamic focusing and time-interleaved segmentation: modelling and experiment, *Lab Chip*, 5(11) (2005) 1320-1326.
- [37] D. Erickson, D. Li, Influence of surface heterogeneity on electrokinetically driven microfluidic mixing, *Langmuir*, 18(5) (2002) 1883-1892.
- [38] J.L. Anderson, W. Keith Idol, Electroosmosis through pores with nonuniformly charged walls, *Chem. Eng. Commun.*, 38(3-6) (1985) 93-106.
- [39] J.H. Masliyah, S. Bhattacharjee, *Electrokinetic and Colloid Transport Phenomena*, First ed., Wiley, New Jersey, 2006.
- [40] H. Keramati, A. Sadeghi, M.H. Saidi, S. Chakraborty, Analytical solutions for thermo-fluidic transport in electroosmotic flow through rough microtubes, *Int. J. Heat Mass Transfer*, 92 (2016) 244-251.
- [41] L. Chang, Y. Jian, M. Buren, Y. Sun, Electroosmotic flow through a microparallel channel with 3D wall roughness, *ELECTROPHORESIS*, 37(3) (2016) 482-492.
- [42] L.W.H. Winky, W.T. Dieter, J.S. Nikolaus, W. Man, Z. Yitshak, Surface-chemistry technology for microfluidics, *J. Micromech. Microeng.*, 13(2) (2003) 272.

Fig. 1. Schematic representation of the geometry considered along with the coordinate system and the reacting components.

Fig. 2. Comparison between the numerically computed values of θ_s and the predictions of Eq. (26) from injection to saturation time for reaction-limited conditions.

Fig. 3. Two non-uniform distributions of ζ^* considered in the present study.

Fig. 4. Flow patterns in the channel considering a sinusoidal distribution of zeta potential.

Fig. 5. Analyte concentration contours for (a) uniform and (b) sinusoidal distributions of zeta potential.

Fig. 6. Dimensionless depletion zone thickness δ^* for both uniform and sinusoidal distributions of zeta potential.

Fig.7. θ_s versus z^* at different equidistant values of t^* between 0.45 and 14.4 for uniform surface characteristics.

Fig. 8. Comparison between numerically computed values of u_{eff}^* with those predicted by Eq. (36).

Fig. 9. Relative difference between the saturation time of uniform and sinusoidal distributions of the zeta potential

τ_{sin} versus Da at different values of Pe .

Fig.10. t_{sat}^* versus α for uniform and non-uniform distributions of the zeta potential; the relative saturation time difference is also shown on the right ordinate.

Fig. 11. t_{sat}^* versus β for uniform and non-uniform distributions of the zeta potential; the relative saturation time difference is also shown on the right ordinate.

Biographies

Mojtaba Abdollahzadeh received his B.Sc. in mechanical engineering from Shiraz University, Shiraz, Iran in 2013 and his M.Sc. in mechanical engineering, energy conversion from Sharif University of Technology, Tehran, Iran in 2016. His research interest is microfluidics, especially using CFD for enhancing the performance of microdevices.

Mohammad Said Saidi received his Ph.D. in Nuclear Engineering from Massachusetts Institute of Technology, USA. He is now Professor at the School of Mechanical Engineering at Sharif University of Technology, Tehran. His research concentrates on Computational Fluid Dynamics and Simulation of micro- and macro-multiphase flows in human body.

Arman Sadeghi received his B.Sc. in mechanical engineering from Razi University in 2006. He then continued his studies in Sharif University of Technology from which he obtained his M.Sc. and Ph.D. degrees in 2009 and 2013, respectively. Since 2014 he is an assistant professor of Mechanical engineering at the University of Kurdistan. His research interests include electrokinetic phenomena and micro/nanofluidics.