<table>
<thead>
<tr>
<th>Abstract</th>
<th>Poster Title</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>AES-1</td>
<td>Rapid Detection of Bacteria in Blood for the Early Diagnosis of Sepsis</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td><em>by Sachidevi Puttaswamy and Shramik Sengupta</em></td>
<td></td>
</tr>
<tr>
<td>AES-2</td>
<td>Electrokinetic Differentiation of Microorganisms in Glass Microchannels</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td><em>by Lucia D. Garza-García, Victor H. Pérez-González, Oscar A. Pérez-Sánchez</em></td>
<td></td>
</tr>
<tr>
<td></td>
<td>and Blanca H. Lapizco-Encinas</td>
<td></td>
</tr>
<tr>
<td>AES-3</td>
<td>Dielectrophoretic Characterization of Microorganisms Employing Three Dimensional</td>
<td>4-5</td>
</tr>
<tr>
<td></td>
<td>Carbon Electrodes</td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>by Héctor Moncada-Hernández, Victor H. Pérez-González, Rodrigo Martinez-Duarte,</em></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Sergio O. Martínez-Chapa, Marc J. Madou and Blanca H. Lapizco-Encinas</em></td>
<td></td>
</tr>
<tr>
<td>AES-4</td>
<td>Assessing Particle Selectivity of An Insulator-Based Dielectrophoretic Microdevice</td>
<td>6</td>
</tr>
<tr>
<td></td>
<td><em>by Ana V. Chávez-Santoscoy, Javier L. Baylon-Cardiel, Héctor Moncada-Hernández</em></td>
<td></td>
</tr>
<tr>
<td></td>
<td>and Blanca H. Lapizco-Encinas</td>
<td></td>
</tr>
<tr>
<td>AES-5</td>
<td>Size, Charge, and Affinity Fractionation of Hemolyzed Sera From a Neonatal Repository</td>
<td>7</td>
</tr>
<tr>
<td></td>
<td><em>by Gary B. Smejkal, Camilia R. Martin, Steven Freedman and Winston P. Kuo</em></td>
<td></td>
</tr>
<tr>
<td>AES-6</td>
<td>Spermatogenic Cells Manipulation Employing Dielectrophoresis</td>
<td>8</td>
</tr>
<tr>
<td></td>
<td><em>by Elizabeth Rosales-Cruzaley, Perla A. Cota-Elizondo, Daniel P. Sánchez-Herrera</em></td>
<td></td>
</tr>
<tr>
<td></td>
<td>and Blanca H. Lapizco-Encinas</td>
<td></td>
</tr>
<tr>
<td>AES-7</td>
<td>Particle Manipulation In a Multi-Section Insulator-Based Dielectrophoresis</td>
<td>9-10</td>
</tr>
<tr>
<td></td>
<td><em>by Roberto C. Gallo-Villanueva and Blanca H. Lapizco-Encinas</em></td>
<td></td>
</tr>
<tr>
<td>AES-8</td>
<td>Electro-Poiseuille Flow Modeling in Annular Geometry</td>
<td>11</td>
</tr>
<tr>
<td></td>
<td><em>by Jeffery W. Thompson, Seth Wynne, Holly Stretz, Mario Oyanader and Pedro Arce</em></td>
<td></td>
</tr>
<tr>
<td>AES-9</td>
<td>Temperature Distribution in Electrochromatography with an Oscillatory Transverse Electric Field</td>
<td>12</td>
</tr>
<tr>
<td></td>
<td><em>by Wei Yuan, David R. Nielsen and Yan Sun</em></td>
<td></td>
</tr>
<tr>
<td>AES-10</td>
<td>Monitoring Algae Species in Bio-Fuel Production by CE-SSCP</td>
<td>13</td>
</tr>
<tr>
<td></td>
<td><em>by Alice C. Jernigan, Lauren Woods, Robert Beitle, Jamie Hestekin and Christa Hestekin</em></td>
<td></td>
</tr>
<tr>
<td>AES-11</td>
<td>Role of Channel Morphology In Microfluidic Applications: Impact on the Behavior of the Electrostatic Potential For An Idealized Case</td>
<td>14</td>
</tr>
<tr>
<td></td>
<td><em>by Parvin Golbayani, Kevin T. Scale, Robert Sanders and Pedro Arce</em></td>
<td></td>
</tr>
<tr>
<td>AES-12</td>
<td>Gradient Dielectrophoresis and Separations-Based Arrays</td>
<td>15</td>
</tr>
<tr>
<td></td>
<td><em>by Sarah J.R. Staton, Michelle M. Meighan, Noah G. Weiss, Stacy Kenyon, Paul V. Jones, Prasun Mahanti, Thomas J. Taylor, Kang P. Chen and Mark Hayes</em></td>
<td></td>
</tr>
<tr>
<td>AES-13</td>
<td>Mobility and Diffusion Regimes In Field Inversion Gel Electrophoresis of DNA In the Sub-35 Kilobase Size Range</td>
<td>16</td>
</tr>
<tr>
<td></td>
<td><em>by Airong Li and Victor M. Ugaz</em></td>
<td></td>
</tr>
<tr>
<td>AES-14</td>
<td>Enhancing Resolution In Microchip DNA Electrophoresis by Tailoring Hydrogel Nanostructure to Exploit Entropic Trapping</td>
<td>17</td>
</tr>
<tr>
<td></td>
<td><em>by Nan Shi and Victor M. Ugaz</em></td>
<td></td>
</tr>
<tr>
<td>AES-15</td>
<td>Blood Cell Capture In a Gradient Dielectrophoretic Microchannel</td>
<td>18</td>
</tr>
<tr>
<td></td>
<td><em>by Paul V. Jones, Sarah J.R. Staton and Mark Hayes</em></td>
<td></td>
</tr>
<tr>
<td>AES-16</td>
<td>A Membraneless Continuous-Flow Filter for High-Throughput Separation and Enrichment of Particles and Cells</td>
<td>19</td>
</tr>
<tr>
<td></td>
<td><em>by Jen-Huang Huang and Victor M. Ugaz</em></td>
<td></td>
</tr>
</tbody>
</table>
AES-1

Rapid Detection of Bacteria in Blood for the Early Diagnosis of Sepsis

Sachidevi Puttaswamy and Shramik Sengupta

Department of Biological Engineering
University of Missouri
Columbia, MO

Sepsis, (systemic inflammation caused by bacterial infection of blood) is a leading cause of death in the US with a mortality rate of ~ 30%. The clinical symptoms displayed by the patients are subtle and non-specific. At the time clinical symptoms begin to be displayed, the concentration of bacteria present in their blood is often as low as 1-10 CFU/ml. If allowed to progress, the disease leads to multiple organ failure and ultimately death. Multiple studies have shown that the quicker one can detect the presence of bacteria, and start effective antibiotic therapy, the greater is the likelihood of survival for the patient. The current technologies for this purpose are Automated Blood Culture Systems such as the BACTEC, BacT/Alert and VITEK, which take 1-5 days to obtain the result.

Unlike the current culture systems that detect the presence of bacteria in the suspensions indirectly via the effects of bacterial metabolism (change in pH of levels of O₂/CO₂), we use a novel technique that relies on the fact that bacteria can store large amount of charge and thus increase the bulk capacitance of the liquid in which they are suspended. An increase in the number of bacteria leads to an increase in the bulk capacitance of the suspension. Usually changes in the bulk capacitance are masked by the electrochemical interfacial (double layer) capacitance. But by leveraging microfluidic geometry, and taking measurements at multiple frequencies, we are able to accurately monitor changes in the bulk capacitance. Doing so enables us to pick up signatures of bacterial presence in the suspension at concentrations of ~ 1000 cfu/ml (as opposed to > 10⁶ cfu/ml for current technologies), and detect the presence of ~10 E coli /ml within 4 hours, as opposed to ~20 hours required by current technologies.
Microscale electrokinetic techniques have a great potential for the separation and sorting of microorganisms, and could solve the need for rapid and early detection of pathogens in medical diagnostics and microbial contamination of food items and water. Presented here is the application of micro particle image velocimetry (µPIV) for the characterization of electrokinetic transport of three types of microorganisms in a glass microchannel: Escherichia coli, Ankistrodesmus sp., and Saccharomyces cerevisiae. The electrokinetic behavior of these three types of microorganisms was characterized employing a straight glass microchannel and direct current electric fields (50-300 V/cm). The effects of microorganism type and size, electric field magnitude, pH (5-8) and conductivity (25-100 µS/cm) of the suspending medium were analyzed. Additionally, electrokinetic differentiation was achieved when a sample containing a mixture of the three types of microorganisms was analyzed by performing fluorescence measurements at the outlet of the microchannel and generating electropherograms in 1-2 minutes. It was demonstrated that this simple system allowed identifying that three different species were present. These results demonstrate that fast and effective differentiation of intact microorganisms can be achieved employing microscale electrokinetic techniques.

**Figure 1.** Schematic representation of the experimental set-up, showing the interrogation window employed for µPIV.
Dielectrophoretic Characterization of Microorganisms Employing Three Dimensional Carbon Electrodes

Héctor Moncada-Hernández¹, Victor H. Pérez-González¹, Rodrigo Martinez-Duarte², Sergio O. Martínez-Chapa¹, Marc J. Madou² and Blanca H. Lapizco-Encinas³

¹BioMEMS Research Chair
Tecnologico de Monterrey
Monterrey, Mexico

²Department of Mechanical & Aerospace Engineering
University of California, Irvine
Irvine, CA

³Microscale Bioseparations Laboratory
CINVESTAV-Monterrey
Apodaca, Mexico

Dielectrophoresis (DEP) refers to the movement of a particle immersed in a polar media exposed to a non uniform AC or DC electric field. As the particle interacts with the applied electric field gradient a dipole moment is induced. The magnitude of this force is mainly determined by the magnitude of the applied electric field while the direction is given by the relative difference on dielectric properties between the particles and their surrounding media. DEP has been successfully applied for the manipulation and concentration of a wide array of bioparticles. Several applications have been demonstrated using metal or insulator-based electrodes which until now had been the two main trends on the material choice for electrodes. The traditional method to induce DEP has been based on the use of metal electrodes. Advantage of this approach is the obtainment of high electric field gradients employing low applied voltages. However the use of metal electrodes must obey a limit on the magnitude of the voltage applied in order to prevent sample electrolysis. An alternative to metal electrodes is the use of insulator-based DEP (iDEP). In this technique a uniform electric field is applied across an array of insulating structures. The presence of these structures will then distort the electric field rendering it non uniform around the insulating structures. However, since only two electrodes are used, large potential are required and only low-conductivity samples can be used to avoid Joule heating. In general the use of 3D electrodes in DEP applications allows for higher separation throughputs than when using 2D electrodes. Carbon-based-electrode Dielectrophoresis (CarbonDEP) is a technique that employs carbon structures or surfaces as electrodes. The use of carbon as electrode material offers several advantages which make it a more suitable material than those traditionally used in metal-electrode DEP. One of the most current and promising techniques for carbon microfabrication is that of Carbon MicroElectroMechanical Systems (C-MEMS). Presented here is the implementation of a bioparticle characterization platform based on CarbonDEP. A microdevice containing arrays of
3D carbon electrodes was employed to manipulate bacterial and yeast cells in order to characterize cell dielectric properties. Different cells responses were obtained by varying the magnitude and frequency of the applied AC potential as well as the characteristics of the suspending medium. Simulation work allowed to build a model to predict cells responses. These results have the potential to be used as guidelines for the design and operation of CarbonDEP-based systems. Potential applications include clinical analyses, environmental screening for water contamination, and optimization of cell culture techniques, improvement of clean energy production methods and food safety methodologies and procedures.

Experimental Set-up. a) representation of microchannel and carbon cylindrical electrodes. b) top view of microdevice. c) Communication and control between the inverted microscope and the multichannel generator with a personal computer.
Assessing Particle Selectivity of An Insulator-Based Dielectrophoretic Microdevice

Ana V. Chávez-Santoscoy¹, Javier L. Baylon-Cardiel¹, Héctor Moncada-Hernández¹ and Blanca H. Lapizco-Encinas²

¹BioMEMS Research Chair
Tecnológico de Monterrey
Monterrey, Mexico

²Microscale Bioseparations Laboratory
CINVESTAV-Monterrey
Apodaca, Mexico

Miniaturation has brought important advantages to separation technologies; such as reduced sample and reagent consumption; higher resolution and sensitivity, low cost and shorter processing time. Dielectrophoresis (DEP) is an extensively employed micro analytical technique with great potential for the manipulation of a wide array of bioparticles. Insulator-based DEP (iDEP) is a dielectrophoretic mode where nonuniform electric fields are created employing insulating structures. In this work, an evaluation of the selectivity of an iDEP microdevice by employing mixtures of polystyrene nano and microparticles is presented. Experimental and mathematical modeling work was carried in order to assess the selectivity of an iDEP glass microdevice to immobilize only the large particles from a binary mixture. The effects of particle concentration, concentration ratio, size ratio, and magnitude of the applied electric potential (200 to 600 V) on the microdevice selectivity were studied. The results demonstrated that high selectivity can be obtained with iDEP.
Size, Charge, and Affinity Fractionation of Hemolyzed Sera From a Neonatal Repository

Gary B. Smejkal\textsuperscript{1}, Camilia R. Martin\textsuperscript{2}, Steven Freedman\textsuperscript{2} and Winston P. Kuo\textsuperscript{1}

\textsuperscript{1}Harvard Catalyst, Laboratory for Innovative Translational Technologies
Harvard Medical School
Boston, MA

\textsuperscript{2}Department of Neonatology
Beth Israel Deaconess Medical Center
Boston, MA

Hemolysis of serum or plasma can exclude very valuable samples from longitudinal studies, particular over time course, and risks the loss of valuable data. In human sera, the concentration of albumin is nearly ten billion times greater than that of cell-signaling proteins like the interleukins. The relative proportions of such rare, but extremely important biomarkers is further decreased in hemolyzed samples in which hemoglobin may nearly double the total protein concentration. Further, hemoglobin removal alone does not significantly decrease sample complexity. Sample prefractionation ideally partitions high abundance proteins like albumin, IgG, and hemoglobin into fractions separate from those containing the protein(s) of interest, potentially enabling enrichment and increasing the sensitivity of downstream microassays attempting to quantify these critical molecules. Samples from a neonatal depository are hoped to provide valuable information on pro-inflammatory response during the first days of life. However, such samples are extremely rare and frequently hemolyzed. To derive meaningful data from hemolyzed whole bloods and sera, fractionation schemes based of protein molecular mass, charge, or affinity were investigated as means of hemoglobin depletion with concomitant biomarker enrichment. Size separations were performed by sequential ultrafiltration with decreasing molecular weight cutoffs (MWCOs) to produce distinct size fractions, or by electrophoresis in the GELFREE System. Charge fractions were evaluated in the OFFGEL Fractionator or a Digital Protein Chip (dPC) capable of producing fractions of 0.05 pl unit intervals as well as pH specific column binding and elution. For affinity based separations, The ProteoMiner immobilized hexapeptide ligand library was used to isolate low abundance proteins without requiring prior hemoglobin depletion.
Spermatogenic Cells Manipulation Employing Dielectrophoresis

Elizabeth Rosales-Cruzaley, Perla A. Cota-Elizondo, Daniel P. Sánchez-Herrera and Blanca H. Lapizco-Encinas

Microscale Bioseparations Laboratory
CINVESTAV-Monterrey
Apodaca, Mexico

Dielectrophoresis (DEP) is an electrokinetic transport mechanism that occurs when particles are exposed to a nonuniform electric field. DEP has great potential for the manipulation of a wide array of bioparticles; it has been successfully used to concentrate, detect and separate virus, bacteria, yeast and parasites. Clinical applications of DEP have been also explored, demonstrating the dielectrophoretic detection of tumor cells and the sorting of red blood cells. Traditionally, DEP has been applied using arrays of microelectrodes and alternating current electric fields. However, electrode-based DEP has some drawbacks, such as the complex fabrication steps required to create microelectrode arrays and loss of efficiency due to fouling which is common when handling biological samples. An alternative is to employ insulator-based DEP, a technique, where nonuniform electric field are created by using “obstacles” or insulting structures that distort electric field distribution. This novel DEP mode requires simples and inexpensive microdevices. In this work we report the manipulation of mice spermatogenic cells employing direct current electric fields applied across a microchannel containing an array of insulating structures. Sperm cells were stained with a DNA-intercalating dye to allow visualization. Rapid concentration and sorting of sperm cells was achieved by employing an electric field gradient. The response of the cells was analyzed by varying operating conditions: magnitude of the applied field and suspending medium characteristics. It is expected that the application of DEP for sperm cell manipulation could be used to speed up selection of sperm cell in to be used in reproductive technologies.
Particle Manipulation In a Multi-Section Insulator-Based Dielectrophoretosis Microdevice

Roberto C. Gallo-Villanueva¹ and Blanca H. Lapizco-Encinas²

¹BioMEMS Research Chair
Tecnologico de Monterrey
Monterrey, Mexico

²Microscale Bioseparations Laboratory
CINVESTAV-Monterrey
Apodaca, Mexico

Particle separation and concentration has become very important as an analytical method; however, performing such operations is not simple due to similarities in physical and chemical properties of the analytes on complex samples. Many techniques have been developed with the aim of purifying samples, but there is still a wide area of opportunity in terms of resolution and efficiency. Driven by advances in electronics, miniaturization offers an alternative with minimal usage of substances and samples, decrease of wastes, shorter process times, greater sensitivity and portability. Nowadays fabrication techniques allow for novel designs to improve separation technologies.

One of such technologies is dielectrophoresis (DEP), an electrokinetic transport mechanism of particles due to polarization effects in the presence of non-uniform electric fields [1]. The most common approach to apply DEP is with AC electric fields employing arrays of microelectrodes [2]. However, manufacture of microdevices with electrode arrays can be expensive and device performance is affected by electrode degradation [3]. An alternative to produce nonuniform electric field is to employ insulating structures between two electrodes, i.e., insulator-based dielectrophoresis (iDEP).

Despite of being a nascent technique, there have been many successful microdevice designs used to separate mixtures of particles according to their dielectrophoretic response. Kang, et al. [4], developed a microchannel with an insulating hurdle that allowed separation of particles by deflecting their electrokinetic path according to their size. The particles experience negative DEP at the corners of the block which magnitude depends on their volume. With their design the authors could continuously separate a mixture of two different polystyrene microspheres. Using the same principle but with a circular channel with electrodes on its extremes, Zhang, et al. [5], suggested continuous sorting of particles by their size. Pysher and Hayes [6] developed a saw-tooth microchannel which tooth dimensions change along the channel. In this design the width of the channel gradually decreased in the direction of the flow, leading to a gradual increase of the electric field gradient, and therefore the increasing the DEP force. They reported the separation of live and dead samples of Bacillus subtilis, Escherichia coli and Staphylococcus epidermis in different regions of the same microdevice.
In the present study, insulating cylindrical posts are used to produce a nonuniform electric field and dielectrophoretically trap polystyrene microspheres of different sizes, employing DC electric fields. The microdevice employed was manufactured from glass arrays of cylindrical post of different diameters, which creates regions with different magnitudes of dielectrophoretic forces. In this way, with a specific electric field, a mixture of particles can be sorted by size and trapped in the region where negative dielectrophoretic force overcomes electroosmotic flow. Each section of the microdevice has its own outlet, allowing for the concentrated and separated sample to be recovered. Results show that the spheres exhibited negative DEP under direct current electric fields, where the larger particles showed stronger response. A complex mixture of particles could then be fractionated and simultaneously concentrated, demonstrating the great potential of the technique to handle complex samples.

**References**


![Figure 1. Schematic representation of the microdevice employed. Insulator posts are arranged in three sections with different cylindrical insulator posts in each one (D = diameter of posts). Sample is loaded in the inlet reservoir and electric fields are applied between the inlet and outlet reservoirs. Lateral reservoirs were sealed and not used.](image)
The effect of geometry on the separation of ions in electrolytic solution is studied. The rectangular and cylindrical cases have already been derived as the result of previous work. For the case of steady-state flow in the annular region between two fixed, charged, concentric cylinders, the profiles of concentration, electric potential, and total mixture velocity have been obtained assuming the electrolytic fluid to be dilute in ion concentration. A decoupling technique is employed to handle the separation of the hydrodynamic problem from the electrostatics problem. For the electrostatics, the Debye-Huckel approximation is used, however a numerical method for improving this linearization of the Poisson-Boltzmann equation is discussed and shown to provide excellent convergence characteristics. Finally, the species continuity equation is stated (in terms of the electro-migration flux) and solved.
Temperature Distribution in Electrochromatography with an Oscillatory Transverse Electric Field

Wei Yuan¹, David R. Nielsen² and Yan Sun¹

¹Department of Chemical Engineering
Tianjin University
Tianjin, China

²Department of Chemical Engineering
Arizona State University
Tempe, AZ

A three-compartment column of ion-exchange electrochromatography with an oscillatory electric field perpendicular to mobile-phase flow driven by pressure (pIEEC) was verified for its high throughput purification of proteins. Electroosmotic flow was proved to be major contributor to its mass transfer acceleration. By modeling work, protein distribution in adsorbents was found to present excursion along the electric field direction. The electro-kinetic convection in porous particle was predicted as exponentially decreased function of protein adsorption amount. So that, at the beginning of the pIEEC, intraparticle convection caused by the electric field contributed more to the enhancement of dynamic binding capacity. For utilization of the new design, an experimental system for in-column temperature measurements was constructed, and the dynamic processes of the in-column temperature in electrochromatography were examined. With the experimental system, the effect of electric current strength and mobile-phase ionic strength on the in-column temperature was investigated. Then, a heat transfer model for the pIEEC was established for the dynamic process modeling. The mathematical model concerns force and free convection heat transfer, as well as Darcy modified wall effect in packed bed column. It was confirmed that the model was in good agreement with the measurements. By the model calculations, we could also obtain the in-column voltage distribution, effective voltage applied to the central compartment and the efficiency of energy consumption. Moreover, the model was used for scale-up analysis and for investigation of temperature distribution under various conditions.
With its ability to use carbon dioxide from the atmosphere as a carbon source, and to clean nitrates and phosphorous from waste water, algae has the potential to be a significantly eco-friendly bio-fuel source. One of the problems faced in using algae as a bio-fuel is identifying and characterizing the best species for the purpose, and keeping them as the dominate species. The identification of algal communities can be accomplished by isolating DNA from water samples. These DNA samples can then be characterized by a technique called single-strand conformational polymorphism (SSCP) analysis of the 18S gene. The 18S gene is highly conserved among eukaryotes, but has unique small variable regions that can be used to differentiate between species. These regions can be amplified by polymerase chain reaction (PCR) giving small fragments of the gene sequence. SSCP, a method that is normally used to detect mutations, is the heat denaturing of these fragments resulting in single strand DNA. The refolding of the single strand DNA will form different secondary structures (conformations) depending on the nucleotide sequence. These different conformations can be separated due to their different electrophoretic mobility, even if they are the same size. The increased speed and automation of capillary electrophoresis (CE)-SSCP will allow for more rapid and reliable results allowing a quick and inexpensive method for monitoring the different algae species.
Role of Channel Morphology In Microfluidic Applications: Impact on the Behavior of the Electrostatic Potential for An Idealized Case

Parvin Golbayani¹, Kevin T. Seale³, Robert Sanders² and Pedro Arce¹

¹Department of Chemical Engineering
²Office of Research and Graduate Studies
Tennessee Technological University
Cookeville, TN

³Department of Biomedical Engineering
Vanderbilt University
Nashville, TN

Microfluidic concepts have become more widely known and helpful to advance technological applications in, for example, health informatics and environmental proteomics just to mention two leading ones. The systematic investigation of the role of both, channel geometry and channel morphology are far to be complete. In particular, their impact on the key variable for electrokinetic applications, i.e., the electrostatic potential has not been addressed. This is of crucial importance since the electric fields can induce electro-convective flow of an electrolyte inside the microfluidic devices that controls, for example, the overall motion of the analyte. The understanding of this motion can help in assessing the role of the electrostatic potential on the transport aspects of the analyte and, therefore, understand the characteristics and performance of microfluidic devices.

The complexities of analyzing channel geometry and channel morphology are not trivial and, in this project, we have proposed a relative simple (but effective) selection of two geometries, i.e. rectangular and cylindrical, and one type of morphology: A convergent and divergent section of the channel. As a by-product of the investigation, one can assess also the behavior of the electrostatic potential inside of a convergent-divergent section. Three key parameters have been identified to describe the electrostatic potential behavior: The angle (α) of the convergent (or divergent) section (related to the walls of the channels) that handles the “magnitude” of the deviation with respect to the regular channel; the ratio of the wall potentials, R, which handles the symmetrical/non-symmetrical aspects of the electrostatic potential, and the ratio of the width to the length (γ) that controls the “shape” of the channel section.

A study of the electrostatic potential in divergent and convergent symmetrical channels has been performed based on the use of the 2D Poisson-Boltzmann Equation (PBE); this equation has been solved for the cased of the Debye –Huckel approximation. Results of this study will be shown by using a series of portraits that capture the key behaviors of the electrostatic potential with respect to the three parameters described above. Suggestions for integrating these results into flow and transport studies will be also offered.
So called ‘dirty' and messy complex samples from biological and environmental sources require significant processing to present a simplified and clean fraction to detection elements. In many cases, it is desirable to provide for analysis of multiple analytes from a single sample, such that specific targets are isolated and concentrated away from background and other analytes. Further, there is a significant push to miniaturize and create one step processing based on microfluidic systems. Here we present two interrelated techniques to take real world biological and environmental samples, remove unwanted background particulate debris, and then isolated and concentrate targets in a highly parallelized format. The two techniques are DC insulator-based gradient dielectrophoresis and parallel electrophoretic capture. Preliminary results show the ability to differentially isolated particles ranging from 20 nm to 1 micron using various physical parameters, increasing their local concentration by up to one million times. For molecular targets, small molecules to proteins have been differentially isolated and concentrated in specific sub-microliter volumes demonstrating the utility of the approach across the entire range of targets of interest. We envision being able to uniquely isolate and concentrate particulates (cells, viruses, bacteria, spores, organelles, etc.), process them and execute highly parallelized molecular isolation and concentration on a large number of targets reaching maximum detection limits and dynamic range.
Mobility and Diffusion Regimes In Field Inversion Gel Electrophoresis of DNA In the Sub-35 Kilobase Size Range

Airong Li and Victor M. Ugaz

Artie McFerrin Department of Chemical Engineering
Texas A&M University
College Station, TX

Pulsed field gel electrophoresis (PFGE) methods have become standard tools in a wide range of DNA analysis applications, but many aspects of DNA migration phenomena under these conditions are not well understood. One of the main reasons for this deficiency is that PFGE experiments are cumbersome to perform due to extremely long separation times (~ 10 - 15 hours) and the need to perform gel analysis by post-staining after completion of the run. We have developed an easy to build miniaturized slab gel apparatus that addresses these issues by enabling large DNA fragments up to 35 kb in length to be separated using field inversion gel electrophoresis (FIGE) in 60 - 90 min. The compact size of the device combined with the use of quartz as the substrate material permit the gel to be continuously illuminated with UV light so that the separation processes can be recorded in real time using a CCD camera.

These capabilities allow us to probe size dependence of fundamental physical parameters associated with DNA migration (mobility, diffusion, and separation resolution). These data reveal a surprising regime where separation resolution increases with DNA fragment size owing to a favorable interplay between mobility and diffusion scalings, and highlight the important role of diffusion (a seldom quantified parameter). In addition to the practical benefit of separation times that are an order of magnitude faster than conventional instruments, the results of these studies provide a previously unavailable rational basis to identify optimal separation conditions and contribute new insights toward understanding the underlying physical processes that govern DNA electrophoresis in pulsed fields.

Evolution of a typical separation run in the miniaturized FIGE system shows the ability to resolve DNA fragments ranging from 2.5 to 35 kb in 90 min.
Electrophoretic DNA transport through nanoporous hydrogels is characterized by an entropic trapping (ET) mechanism when the DNA size is close to the gel pore size. This mode of transport can yield highly desirable transport properties (e.g., enhanced size dependences of mobility and diffusion for improved separations). But hydrogels have largely been ignored in efforts to understand and exploit ET effects, where the focus has instead centered on simpler idealized “nanofilter” architectures based on arrays of alternating deep wells and narrow slits (i.e., describable by only two length scales).

We have recently developed a new model that captures the inherently heterogeneous pore morphology comprising realistic hydrogel matrices, enabling key features of macromolecular transport in the ET regime to be successfully predicted. When combined with material characterization methods we have developed that enable both the mean pore size and pore size distribution of the gel to be quantified, we are able to establish a direct link between sieving matrix morphology and DNA migration in photopolymerized crosslinked polyacrylamide gels cast under different UV intensities and concentrations. We can then directly apply these insights using a microfluidic platform that allows continuous whole channel scanning of DNA separation progress so that the size dependence of these parameters can be quickly and accurately measured. A photocurable hydrogel matrix allows the pore architecture to be tailored by adjusting the UV light intensity. In this way, we are able to select polymerization conditions that produce pore morphologies favorable for ET, and harness these effects to achieve improved separation performance.

We have applied this approach to examine electrophoresis of double-stranded DNA in the 100 to 1000 bp size range, and have identified gel polymerization conditions that induce a transition in the physical mechanism of DNA migration from reptation to entropic trapping. Furthermore, we have identified a range of conditions where a favorable interplay exists between these migration mechanisms that results in improved separation performance (e.g., resolving power that increases with DNA size, the opposite of what is conventionally observed). These new insights have also led to an unexpected prediction that oscillating the applied electric field at a specific frequency, in a specific gel morphology where ET effects dominate, can establish a resonance phenomenon that greatly improves separation resolution by reducing diffusion. Here the electric field is generated in an “on-off” manner to synchronize discrete hops between neighboring pores in a very specific gel morphology, in contrast to conventional pulsed field methods. Our microchip electrophoresis experiments support this prediction, with a doubling in separation resolution observed at DNA fragment sizes below 500 bp.
Blood Cell Capture In a Gradient Dielectrophoretic Microchannel

Paul V. Jones, Sarah J.R. Staton and Mark Hayes

Department of Chemistry and Biochemistry
Arizona State University
Tempe, AZ

Biological fluids can be considered to contain information-rich mixtures of biochemicals and particles that enable clinicians to accurately diagnose a wide range of pathologies. Rapid and inexpensive analysis of blood and other bodily fluids is a topic gaining substantial attention in both science and medicine. Current limitations to these analyses include long culture times, expensive reagents, and the need for specialized laboratory facilities and personnel. Improving these tests and overcoming their limitations would allow faster and more widespread testing for disease and pathogens, potentially providing a significant impact for healthcare, especially in developing nations. One line of development involves microfluidic approaches that provide unique advantages over entrenched technologies, including rapid analysis times, microliter sample and reagent volumes, and portability. The present study focuses on the isolation and concentration of human blood cells from small-volume samples of diluted whole blood. Separation of cells from the matrix of whole blood was accomplished using DC insulator-based gradient dielectrophoresis in a converging, sawtooth-patterned microchannel. The channel design enabled high-resolution capture by exploiting variations in the characteristic physical properties of cells and other bioparticles. Reproducible capture occurred at specific locales within the channel, over a global applied voltage range of 200 to 700 V. Separation was achieved in isotonic buffers, allowing capture of whole cells.
A Membraneless Continuous-Flow Filter for High-Throughput Separation and Enrichment of Particles and Cells

Jen-Huang Huang and Victor M. Ugaz

Artie McFerrin Department of Chemical Engineering
Texas A&M University
College Station, TX

There is a critical need for advanced filtration methods adaptable for separation of particles, cells, and cell-sized components from complex fluid mixtures—specifically those offering the capability to rapidly process large sample volumes (> mL/min flow rates). Microfluidic technologies provide a natural platform to address these challenges, but most of these efforts have yet to advance past the proof of concept stage.

Here we describe a new microfluidic-based filtration method capable of performing simultaneous size-based isolation and enrichment of particles and cells. Instead of forcing a particle- or cell-laden suspension to flow through tiny pores in a membrane filter, we are able to construct a filter oriented along the centerline of the microchannel so that it creates a barrier between the left and right hand sides. When this geometry is incorporated into a curved flow path, the resulting centrifugal forces that arise due to fluid motion act to push the suspended components across the centerline barrier from the inside wall to the outside wall, with only those smaller than the barrier gap able to pass across. Consequently, this filtration method does not impose an excessive pressure drop because the barrier is oriented parallel to the flow direction rather than perpendicular to it. Moreover, this approach is most effective at high flow rates because the magnitude of the curvature-induced transverse flow is maximized under these conditions, making it ideally suited for high-throughput analysis of large sample volumes.

To construct microchannel networks incorporating embedded centerline barriers, we have developed a new microfabrication approach that capitalizes on the properties of biodegradable poly(lactic acid) (PLA). The process works by perfusing the surface of a PLA substrate with an enzymatic agent capable of cleaving lysine-lysine bonds (e.g. proteinase-k). By manipulating parameters associated with the laminar flow enzymatic degradation process (e.g., temperature, degradation time, enzyme concentration, etc.), complex microchannel topologies can be precisely etched in PLA sheets without the multiple lithography steps that would otherwise be needed.

Filtration capability was evaluated by injecting a mixture of fluorescent polystyrene beads of diameter 3 and 10 µm into the inner inlet at a flow rate of 1 mL/min. Samples were then collected at outlets positioned at the inner and outer sides of the centerline barrier and analyzed by flow cytometry. The inner outlet contained the 10 µm beads that were unable to pass across the barrier, while the outer outlet contained only 3 µm beads. In addition to demonstrating extremely high selectivity, no clogging effects were observed because the primary flow acts to sweep aggregates downstream.