Application of ion concentration polarization to water desalination and active control of analytes in paper

By

Pei Zhang

A thesis submitted in conformity with the requirements for the degree of Master of Applied Science
Graduate Department of Mechanical and Industrial Engineering
University of Toronto

© Copyright 2013 by Pei Zhang
Abstract

Application of ion concentration polarization to water desalination and active control of analytes in paper

Pei Zhang
Master of Applied Science
Graduate Department of Mechanical and Industrial Engineering
University of Toronto
2013

This thesis focuses on the development of two new applications using ion concentration polarization (ICP): an out-of-plane microfluidic approach for water desalination and a method for concentration and transportation of charged analytes in paper-based biomedical diagnostic device.

In the first work, we present an out-of-plane desalination approach using ICP. A depletion boundary separates salt ions and purified water into distinct vertically stacked layers. The out-of-plane design enables multiplexing in three dimensions, providing the functional density required for practical applications. The second work demonstrates an active control mechanism of target analytes in paper using ICP. Both external devices (with all functional units on one side of paper) and integrated paper microfluidic devices (by embedding all functional units in paper) were developed to concentrate and transport charged analyte molecules in the paper. We also demonstrate a new fabrication method of nanofluidic and hydrophobic barriers (nanoporous membrane patterning) in paper microfluidic device.
Acknowledgements

I would like to express my deep gratitude to several people who helped make this work possible. First of all, I thank my supervisor, Dr. David Sinton, for his guidance, enthusiasm and encouragement. His keen ability to manage people and projects helped keep things moving and on track, also showed me the way to ensure successful collaboration through communication with people in academia and industry. Secondly, I want to thank Dr. Brendan MacDonald and Max Gong for contributing their time and knowledge to my project and each member of the Sinton Lab for their helpful discussions. Thirdly, I would like to gratefully acknowledge funding from Grand Challenges Canada through the Stars in Global Health program and infrastructure funding from CFI. Last but not least, a great deal of recognition must also be given to my lovely mother and father who, through years of influence and support, have helped me learn to grow and peruse my goals.
# Table of Contents

Acknowledgements........................................................................................................................ iii

List of Figures........................................................................................................................................ vii

Chapter 1 Introduction.......................................................................................................................... 1

1.1 Background and Motivation ........................................................................................................ 1

1.2 Thesis overview ......................................................................................................................... 2

Chapter 2 Nanofluidic transport ........................................................................................................ 4

2.1 Surface charges ......................................................................................................................... 4

2.2 Electrical double layer ............................................................................................................. 4

2.3 Electro-osmosis ........................................................................................................................ 6

2.4 Surface conduction ................................................................................................................... 6

2.5 Electrical double layer overlap .............................................................................................. 8

2.6 Exclusion-enrichment effect .................................................................................................. 10

2.7 Perm-selectivity of the nanochannel ..................................................................................... 11

2.8 Ion concentration polarization ............................................................................................... 11

2.9 Integrated nanoporous membrane ......................................................................................... 14

2.10 Relevant applications ............................................................................................................ 15

Chapter 3 Out-of-plane ion concentration polarization for scalable water desalination ............. 18

3.1 Introduction ............................................................................................................................... 18

3.2 Experimental ............................................................................................................................ 20
3.3 Results and discussion ........................................................................................................ 21
  Effect of nanoporous membrane geometry ........................................................................... 21
  Effect of flow rate.................................................................................................................. 25
  Out-of-plane desalination device........................................................................................... 27
3.4 Conclusions....................................................................................................................... 29
3.5 Supplementary information ............................................................................................... 30
  Salt water mixtures ................................................................................................................ 30
  Material and device fabrication ............................................................................................ 30
  Data acquisition and analysis ............................................................................................... 30
  Dimensions of out-of-plane desalination device ................................................................... 31
  Supplementary figures .......................................................................................................... 32

Chapter 4 Life after wetting: Active transport and concentration in paper-based microfluidic
devices using ion concentration polarization........................................................................... 33
  4.1 Introduction......................................................................................................................... 33
  4.2 Experimental....................................................................................................................... 35
    Working principle.................................................................................................................. 35
    Materials and device fabrication ........................................................................................ 37
    Data acquisition and analysis ............................................................................................. 38
  4.3 Results and discussion .................................................................................................... 38
    Demonstration of analyte concentration/transportation with external stamp-like device..... 38
Demonstration of sample concentration and transportation with integrated paper microfluidic device

4.4 Conclusions

Chapter 5 Conclusions and Future Work

5.1 Conclusions

5.2 Future work

Potential optimization strategy for desalination device

Potential optimization strategy for external stamp-like device

Potential optimization strategy for integrated paper-based microfluidic device

Appendix A

References
List of Figures

Figure 1 Illustration of EDL model with inner Stern layer and the outer Diffusive layer. The electrostatic potential distribution within the EDL, the zeta potential $\zeta$ is indicated at the slip plane.............................................................. 5

Figure 2 Visualization of surface conduction in a flat electric double layer ......................... 7

Figure 3 EDL overlap occurs as channel size shrinks from micrometer to nanometer scale [19]. 9

Figure 4 Flux of anions $\Phi$ at higher ionic strength is higher than that at lower ionic strength due to exclusion enrichment effect inside nano-sized opening ................................................. 10

Figure 5 Schematic of ICP phenomena (a) before voltage applied (b) movement of counter-ions due to the electric field (c) movement of co-ions due to the electric field (d) formation of depletion and enrichment zone ........................................................................... 13

Figure 6 The general chemical structure of Nafion[32] ................................................................................. 14

Figure 7 Desalination process using ICP on planar device[8] .......................................................... 16

Figure 8 Different protein preconcentration schemes (a) radial preconcentration with an integrated planar nanoporous film (b) concentration device with a vertical nanoporous membrane connecting two microchannels (c) continuously flow concentration device (d) concentration device with nanoporous membrane embedded in a straight microchannel ........................................... 17

Figure 9 A schematic representation of the microfluidic device at the nanoporous membrane where ICP generates a depletion boundary. Charged species are deflected downwards by the depletion boundary into a separate layer, purifying the top stream. The purified, concentrated, and ground channels are located on distinct layers, which enables stacking for increased functional density ........................................................................................................ 20
Figure 10 (a) Schematic of the microfluidic device used for the characterization experiments to enable visualization of the depletion region. A single channel was fabricated on the bottom layer so an inverted microscope could capture the depletion effect. (b) Schematic of the microfluidic device used to illustrate out-of-plane ICP depletion and deflection of ions, and demonstrate the stackable functionality.

Figure 11 Experimental results for various nanoporous membrane widths at a flow rate of 0.5 µL/min and applied voltage of 5 V. (a) Ion depletion percentage is plotted versus nanoporous membrane width. The depletion percentage correlates to the depletion boundary depth, which is shown on the secondary y-axis. The results indicate that larger nanoporous membranes generate larger depletion boundaries. (b) Energy required per litre output is plotted versus nanoporous membrane width.

Figure 12 Experimental results for various flow rates with a nanoporous membrane width of 90 µm and applied voltage of 8 V. (a) Ion depletion percentage and depletion boundary depth is plotted versus flow rate. The results indicate that the depletion boundary depth decreased in a linear manner as the flow rate was increased. (b) Energy required per litre output is plotted versus flow rate. The energy efficiency was consistent for incoming flow rates due to the consistent flow rate of purified water.

Figure 13 Experimental demonstration of out-of-plane desalination. (a) Image of the experiment taken before the voltage was applied and (b) 20 seconds after 20 V was applied. The flow rate was 1.0 µL/min. The source water had a salt concentration of 500 mM and was separated into purified and concentrated layers.

Figure 14 Schematic of the procedure for producing the nanoporous membrane. The silicone sheet is cut with a CO₂ laser and the slice is filled with Nafion®. The result is a nanoporous
membrane between the fluid layers with a width ($w$) and length ($l$) that can be controlled by the laser settings.................................................................................................................................. 32

Figure 15 External paper-based devices. (a) Schematic of the external concentration device. (b) Schematic of the external transportation device. Nanoporous membrane layer on the bottom concentrates and transport charged analytes via application of electric fields as indicated (c) A magnified cross-section of the device where analyte concentration/transportation takes place. Depletion boundary forms at the interface of the saturated paper and nanoporous membrane, focusing analyte molecules towards the anodic side. .................................................................................................................................. 36

Figure 16 Active concentration using the external device. (a) Time evolution of peak concentration factor. The maximum value reaches 41-fold at $t = 190s$. (b) Contrast enhanced fluorescence image sequence of external concentration device. (c) Fluorescence intensity versus distance plot at selected time points as indicated. .................................................................................................................................. 41

Figure 17 Active transport using the external device. Fluorescein image sequence and corresponding intensity plot at 50V, showing active analyte transport in saturated paper strip. Sample “walking” time is ~5min.................................................................................................................................. 43

Figure 18 Integrated paper microfluidic devices. (a) Schematic of the integrated concentration device. (b) Schematic of the integrated transportation device. Nanoporous membrane is embedded in paper matrix to concentrates and transport charged analytes via application of electric fields as indicated (c) A magnified cross-section of the device where analyte concentration/transportation takes place. Depletion boundary forms at the interface of the saturated paper and in paper nanoporous membrane, focusing analyte molecules towards the anodic side. .................................................................................................................................. 45

Figure 19 Active concentration using the integrated paper-based device. (a) Time evolution of peak concentration factor. The maximum value reaches 22-fold at $t = 510s$. (b) Contrast
enhanced fluorescence image sequence of integrated concentration device. (c) Fluorescence intensity versus distance plot at selected time points as indicated. .................................................. 46

Figure 20 Active transport using the integrated paper-based device. Fluorescein image sequence and corresponding intensity plot at 50V, showing active analyte transport. ........................................... 47

Figure 21 Schematic diagram of possible scalable design ........................................................................ 53
Chapter 1 Introduction

1.1 Background and Motivation

What is microfluidics? Referring to Professor George M. Whitesides’ definition: it is the science and technology of systems that process or manipulate small (10⁻⁹ to 10⁻¹⁸ litres) amounts of fluids, using channels with dimensions of tens to hundreds of micrometres [1]. As fluid channel sizes are scaled down to the micro/nano-scale, the governing physics differ significantly from macro-scale fluid mechanics, particularly due to high surface to volume ratio and the dominance of surface effects at small scales. In analytical chemistry and biology, this regime offers several advantages, such as small sample volumes, low processing time, high throughput, small device size, high geometrical control, and potential for integration with electronics [2, 3]. This led to the advent of micro total analysis systems (μTAS) and lab-on-a-chip (LOC) devices, which aim at integrating all steps of biochemical analysis on one microchip [4].

Micro total analysis systems have gained further expansion and increased emphasis in micro/nanostructure based approaches by integrating with nanoporous membrane. This micro/nano hybrid system has been extensively studies for its potential application in analyte pre-concentration and separation, and can become a major tool for genomic and proteomic μTAS [5-11]. Recently, transferring those techniques into unconventional fields, such as environment and energy, has provided exciting opportunities such as desalination [8, 12]. Micro/nanofluidics as a tool-set not only can help us develop a deeper understanding of fundamental small scale science but also may revolutionize the way we do science.

The objective of this work is to apply micro/nanofluidic transport phenomena, specifically ion concentration polarization (ICP), to develop novel engineering approaches for seawater...
**desalination and paper-based point-of-care diagnostics.** In the first work, we present scalable and energy efficient desalination method utilizing a vertical nanoporous membranes within a multilayered microfluidic channel structure. In the second work, we demonstrate active control mechanism of charged analyte in paper using an external stamp-like device and an integrated paper microfluidic device. The external stamp-like device is a standalone device that can be used to transport and concentrate analytes by simply placing it on top of any paper-based microfluidic substrate. The integrated paper-based device is patterned with relevant ICP components (i.e. nanoporous membrane) such that no external parts are necessary except for a power source.

### 1.2 Thesis overview

In this work the concentration polarization phenomena resulting at the interface of micro and nano structures is explored and used to (1) desalinate seawater for potential application in remote areas and (2) active control of target analyte in paper for potential application in point-of-care diagnostics.

In the first project, a device using a vertical nanoporous membrane approach was developed with microchannels to define the active micro/nano interface and shown to be applicable to seawater desalination. In the second project, two portable external devices (with all functional units on one side of the paper) and two integrated paper microfluidic devices (by embedding all functional units in paper) were developed to concentrate and transport analyte molecules in paper. The thesis is broken down into 5 chapters as follows.

In Chapter 1 and chapter 2 the background and motivation were presented and a brief review of relevant nanofluidic transport phenomena is discussed.
Chapter 3 presents the work related to a submitted paper entitled “Out-of-plane ion concentration polarization for scalable water desalination”. The out-of-plane design enables multiplexing in three dimensions, providing the functional density required for practical application. We experimentally assess the impact of the geometry of the vertical nanoporous membrane and the effect of varying flow rates.

Chapter 4 demonstrates an active control mechanism of analyte molecules in saturated paper utilizing ion concentration polarization. The purpose is to design a tool for active concentration and transport of analytes for point-of-care analytical diagnostic device.

In Chapter 5 the thesis is concluded with a look to potential future work stemming from the work presented here.
Chapter 2 Nanofluidic transport

2.1 Surface charges

As the surface-to-volume ratio increases with miniaturization, surface effects dominate the liquid behavior in and around nanometer-sized objects. Generally, most substances (e.g., a fused-silica channel) will acquire a surface electric charge when brought into contact with a liquid electrolyte depending on the number and type of acid and basic groups present in solution. Some of the charging mechanisms include ionization of surface groups, ion adsorption on surface, and ion dissolution (e.g., Si – OH + H₂O ⇌ Si – O⁻ + H₃O⁺). The surface charge density changes with the pH value of the solution [13], its ionic strength [14], and the type of ions [15, 16].

2.2 Electrical double layer

Surface charges result in electrostatic forces which influence the distribution of nearby hydrated ions in the solution. Ions of opposite charge to that of the surface (counterions) are attracted toward the surface while ions of like charge (coions) are repelled from the surface (Figure 1). This attraction and repulsion, when combined with the mixing tendency resulting from diffusion of the ions, leads to the formation of an electric double layer (EDL) – a specific surface charge distribution at the liquid-solid interface.

In immediate proximity to the surface there exists a layer of ions which are relatively strongly fixed by electrostatic forces. This layer, called the Stern layer, forms the inner or compact region of the liquid-side part of the EDL, with a typical thickness of the order of one ion diameter. The outer Helmholtz plane (OHP) separates the Stern and diffusive layers, which together constitute the liquid-side part of the EDL. While ionic species in the diffusive layer undergo Brownian
motion, they are also influenced by the local electrostatic potential. At equilibrium their accumulation in this region can be described by the Boltzmann equation. The spatial dimension of the diffusive layer is typically between a few and a hundred nanometers. Evidently, charge neutrality is not applicable within the EDL since the number of counterions will be large compared with the number of coions. The plane separating the mobile ions from the immobile ions is known as the slip plane, the potential at this plane is characterized by the zeta potential $\zeta$.

Figure 1 Illustration of EDL model with inner Stern layer and the outer Diffusive layer. The electrostatic potential distribution within the EDL, the zeta potential $\zeta$ is indicated at the slip plane.
The EDL thickness is characterized by Debye screening length ($\lambda_D$):

$$\lambda_D = \left( \frac{\varepsilon \varepsilon_0 R T}{F^2 \sum_i Z_i^2 C_{i,\infty}} \right)^{1/2}$$

where $R$ is the gas constant, $T$ the absolute temperature, and $F$ the Faraday constant, $Z_i$ is the valence of ionic species $i$ and $C_{i,\infty}$ its molar concentration in the electro-neutral solution. Debye length corresponds to the thickness of the EDL and is typically on the scale of a few nanometers. This value increases with dilution of the bulkelectrolyte[17].

### 2.3 Electro-osmosis

As described in the previous section, EDL layer forms on the bulk solution side next to the charged surface, so application of an external DC electric field results in a net migration of ions inside EDL. Due to the viscous drag, bulk solution is drawn by the moving ions and causes flows through the channel. This flow motion is referred to as electro-osmosis.

### 2.4 Surface conduction

The surface conduction has long history before microfluidics, it is the excess ionic charges that take place on the solution side of the EDL and may move under the influence of electric fields applied tangentially to the surface as shown in Figure 2 [18]. This would result in tangential current in the EDL along the charged interfaces.
The surface conduction at the solid/liquid interface is quantified in terms of surface conductivity $k^\sigma$ and can be determined by its ratio to the bulk conductivity $k^L$. The fraction of the total conductivity that is governed by the surfaces is given by the dimensionless Dukhin number, $Du$,

$$Du = \frac{k^\sigma}{k^L h}$$

where $h$ is a geometrical length scale, such as the channel width or particle size. If $Du \gg 1$, conduction takes place mainly along the charged surface of the object. Besides, $Du$ increases with decreasing electrolyte concentration.

The total surface conductivity of a system $k^\sigma$ consists of the sum of the surface conductance components of the diffuse part of the double layer, $k^\sigma d$, and the stagnant layer behind the Helmholtz plane, $k^\sigma i$.
\[ k^\sigma = k^{\sigma_d} + k^{\sigma_i} \]

In the diffuse layer two ionic species contribute, their relative roles being determined by the ion transport number, is described as a fraction of the total current carried by an ion. Moreover, in this part the liquid as a whole can also move by electro-osmosis. In the Stern layer counterions dominate and co-ions are completely absent. In this layer there is no electro-osmosis (section 1.2.3).

2.5 Electrical double layer overlap

The size of the nano-sized opening becomes comparable to the thickness of the EDL (a few nanometers) and results in the local electrical potential (of the same sign as the surface potential) remains relatively high even at the center of the channel. This process is sometimes referred to as EDL overlap as shown in Figure 3.
Figure 3 EDL overlap occurs as channel size shrinks from micrometer to nanometer scale [19].

Figure 3 (A) shows the EDL in a microchannel has a Debye screening length much smaller than the channel dimensions. The ion concentration and electric potential are equal to the bulk solution concentration and zero at the center of the channel respectively. Figure 3 (B) shows in a nanochannel the solution is charged due to the overlapped EDL. The electric potential remains relatively high even at the center of the channel. The ion concentration distributions are asymmetric illustrating counter-ions (cations) are enriched and co-ions (anions) are excluded compared to that of the bulk solution. This affects the electrokinetic phenomena in the nanochannel and also results in permselectivity.
2.6 Exclusion-enrichment effect

The exclusion-enrichment effect (EEE) is a phenomenon of enrichment of counter-ions (cations) in and exclusion of co-ions (anions) from a nanometer-sized opening due to electrostatic interactions with the surface charges. This effect was systemically investigated by Plecis and his co-works [15, 16, 20] who developed a simple model of the nanochannel permeability for varying ionic strength.

![Diagram showing High Ionic Strength and Low Ionic Strength](image)

**Figure 4** Flux of anions $\Phi$ at higher ionic strength is higher than that at lower ionic strength due to exclusion enrichment effect inside nano-sized opening

The consequences of the exclusion-enrichment effect on passive transport of charged species through a nanochannel was investigated by measuring the quantitative changes in the nanochannel permeability when the Debye length increases. For anions species, the decrease in nanochannel permeability at low ionic strength can be schematically described as electrostatic “narrowing” of the nanometer-sized opening as shown in Figure 4. At low ionic strength where the EDL gets larger, the co-ions ability to diffuse is reduced, resulting in lower flux. For counter-
ion, the enrichment effect at low ionic strength increases the number of counter-ions in the nanochannel that can be transported by diffusion. This results in a higher flux.

### 2.7 Perm-selectivity of the nanochannel

As a result of the exclusion-enrichment effect, a nanochannel can exhibit perm-selectivity (preferentially conducting ions with one polarity over another) when its dimensions become comparable with the thickness of the Debye screening length as mentioned above. In such cases, counter-ions, which have much higher concentration than that of co-ions inside the nano-sized opening, can be transported through by diffusion, while co-ions cannot due to electrostatic repulsion. This is the fundamental basis of charge-selective nanoporous membranes as applied in reverse-osmosis water purification, and other applications.

### 2.8 Ion concentration polarization

When an external electric field is applied through a nanochannel, electrokinetic transport (migration) is superimposed on diffusion and causes considerable forces on electrolytes in nanometer-sized openings. Ionic concentration gradients and ion concentration polarization, will form in the electrolyte solution adjacent to the micro/nanochannel interface under these circumstances.

Ion concentration polarization (ICP) has been observed over 100 years at nanoporous membranes [21, 22]. The first experimental demonstration of ion depletion/enrichment using a nanochannel was conducted by Pu et al [23]. A microchannel and nanochannel geometry was used to demonstrate the ion concentration polarization effect as shown in Figure 5. Assuming the
surface of both the microchannel and nanochannel are negatively charged, the nanochannel becomes cation-selective due to overlapped EDL. When applying an external electric potential across the system, the counter-ions migrate toward the cathodic electrode due to EEE effect and applied field as shown in Figure 5 (b). However, the co-ions are similarly expelled at cathodic side of nanochannel due to the same reason Figure 5 (c). This results in countion-ion concentration decreases on the anodic side of nanochannel and increases on the cathodic side. In order to maintain local electroneutrality, the salt concentration is reduced near the anodic side of nanochannel and increased near the cathodic side. As a result, an ionic depletion zone is generated near the anodic side of the nanochannel and enrichment zone is formed near the cathodic side as shown in Figure 5 (d). The formation of the depletion zone, which acts as a large electrical resistance, reduces the local value of the electrical conductivity [24]. Therefore, local electric field is greatly amplified (in order to sustain the current) in this depleted region, near the anodic side of the nanochannel, and acts on the counterions screening the wall charge to drive surface conduction [25]. A recent study shows the surface conduction will eventually dominate bulk diffusion in carrying the current through the ion depletion zone [26]. Meanwhile, coions are driven to the left by the amplified electric field, thus further enhancing bulk depletion. Current lines are diverted from the electrolyte solution into the EDL, as they pass through the depletion boundary as shown in Figure 5 (d). The EDL thickness on the microchannel wall is increased inside the depletion zone as compared with that on the bulk side due to the reduced buffer concentration. Counterions move from the electrolyte solution into the EDL in order to carry current behind the depletion zone. Coions electromigrate ahead of the depletion boundary, and they become fully depleted behind it [27].
Figure 5 Schematic of ICP phenomena (a) before voltage applied (b) movement of counter-ions due to the electric field (c) movement of co-ions due to the electric field (d) formation of depletion and enrichment zone
There are many factors affects the performance of the ICP, such as ionic strength of buffer solution [23, 28, 29], pH value [30] and contact area between microchannel and nanochannel/nanoporous membrane [9]. It is also reported that a strong ICP can stop or even reverse the flow direction [31].

2.9 Integrated nanoporous membrane

In this work, a Nafion® membrane is employed as the nanoporous membrane (effectively nanochannels in parallel) and used to trigger ion concentration polarization. It is a widely used ion-selective membrane material with strongly negatively charged sulfonic groups. Nafion membrane was first developed by Dr. Walther Grot at Dupont in late 1960’s by modifying Teflon. Figure 6 shows the molecular structure of Nafion.

![Figure 6 The general chemical structure of Nafion](image)

Nafion combines ionic characteristics with the physical and chemical properties of the Teflon base material to give the final material some unique properties. For example, Nafion, like Teflon, is extremely resistant to chemical attack. Only metallic alkali metals like sodium can attack
Nafion directly under normal operating conditions. Therefore, Nafion doesn’t release degradation products into surroundings. Unlike Teflon, Nafion is highly ion-conductive. The sulfonic acid groups attached to Teflon backbone in Nafion function as an extremely strong proton donor. In addition, Nafion is highly permeable to water and can operate at higher temperatures than many other polymers.

According to the structural model of Gierke[33], the membrane can be considered as a capillary network, where all sulfonic groups are uniformly distributed at the pore wall with a constant negative surface charge density. Therefore, Nafion possesses cation selectivity property which more or less completely excludes negative ions because their electrical charge is identical to that of the pore wall. On the contrary, positive ions are allowed to pass through.

2.10 Relevant applications

Separation of charged species

The first demonstration of using ICP to separate charged species was reported by Han’s group[8]. As shown in Figure 7, a nanojunction, which is used to trigger ICP, is placed at the junction of branches. Under the influence of DC electric field as indicated, depletion zone forms at the junction, and the salts along with other charged species (including blood cells, viruses and microorganisms) in the incoming seawater were repelled away from depletion boundary to inclined channel and fresh water was obtained as product from horizontal channel.
Figure 7 Desalination process using ICP on planar device[8]

This method eliminates the potential for membrane fouling since salt ions are driven away from the membrane. It is a small scale technology, so has great potential to be used in remote areas if engineered properly. It is noteworthy that the high efficiencies achieved in Han’s work have very recently been found to be irreproducible, and an anomaly[34], however, the approach was pioneered by this group. The second chapter of this thesis will present a desalination device with embedded vertical nanoporous membrane approach to increase functional density.

**Protein preconcentration on a chip**

The ion concentration polarization phenomenon has been widely used as pre-concentration method on a chip since 2005. In a series of papers published by Han’s group used Nafion as a planar or vertical nano-junction connecting two microchannels to achieve easily fabricated preconcentration devices[5, 6, 9, 11, 35]. Usually, the DC field was applied across the nano-junction and concentration build-up could be observed at the anodic side of the microchannel. A radial pre-concentration scheme using ICP was also proposed by Scarff in which analyte was focused towards the centre of a micro-chamber and then analyzed in the upper level[36]. This
thesis looked into this scheme as well and presents a paper-based pre-concentration approach in chapter 4.

Figure 8 Different protein preconcentration schemes (a) radial preconcentration with an integrated planar nanoporous film (b) concentration device with a vertical nanoporous membrane connecting two microchannels (c) continuously flow concentration device (d) concentration device with nanoporous membrane embedded in a straight microchannel
Chapter 3 Out-of-plane ion concentration polarization for scalable water desalination

This chapter consists is the basis for a paper which has been submitted to Lab on a chip, and benefited from editing and direction from Dr. Brendan MacDonald and Prof. David Sinton. In this work, I prepared all 3D solid models and conducted experiments at different flow rates with single level unit cell and demonstrated water desalination by ICP with stackable device. Finally, I prepared all 3D solid models for publication.

3.1 Introduction

Access to fresh water is crucial for human health; however, there is a limited supply. Reduced precipitation and vanishing glaciers currently threaten the water supply of more than one-sixth of the Earth’s population [37, 38]. Many of the countries facing declining fresh water sources have ready access to seawater, which accounts for 97.5% of the water on the Earth [39].

Desalination is a capital- and energy-intensive process, which makes it challenging to implement in developing regions. Current technology, such as reverse osmosis, operates most efficiently at large scales, necessitating large centralized facilities. In addition to energy and cost aspects, centralized facilities have limited reach. In Vietnam for instance, ~70% of the population lives in rural communities (~60 million people). A smaller, scalable desalination technology with high energy efficiency could provide fresh water to rural populations on an individual, family, or village scale. A promising technology with the capacity to meet these criteria is desalination by ion concentration polarization (ICP) [8, 40].

ICP is a phenomenon that arises at the junction of microfluidic and ion selective nanofluidic channels when an electric field is applied [22]. A depletion zone forms at the junction, which
repels charged particles [23]. ICP has been largely applied as a method for concentrating analytes [5-7, 9, 10, 19]. Kim et al. demonstrated application of ICP for the inverse process – concentrating ions from salt water with pure water as the output [8]. The desalination device was a planar microfluidic device with a Nafion® nanoporous membrane. The energy efficiency of their device was comparable to large-scale reverse osmosis plants in spite of being a microscale device. Further studies have explored planar nanoscale pathways [7], slices in PDMS filled with a nanoporous membrane [9], and triangular nanochannels along the edge of planar nanoporous membranes [41].

All previous work on desalination using ICP has involved planar designs with the purified water, concentrated stream, and ground channel located in the same layer. While planar devices are well suited to microfabrication, visualization, and proof-of-concept demonstration, they are not suited to scale-up. Traditional planar lab-on-a-chip designs have been effective for analysis, but planar designs are not sufficiently volumetrically efficient when a fluid product is of interest. In short, traditional chips are mostly plastic by volume, whereas successful process technologies are mostly fluid. In order to be effective in applications where there is a fluid-product, as opposed to the more common information-product, a re-imagining of the planar design is required [42]. Transitioning lab-on-a-chip approaches, such as desalination by ICP, to practical technologies, will require effective utilization of the third dimension.

In this chapter a scalable, and energy efficient desalination device using ICP in the out-of-plane direction is presented. This approach enables stacking to yield cost-effective desalination units that can be customized for desired water output levels. The results of experiments with a single level unit cell are presented to quantify unit operation and assess the impact of the geometry of the vertical nanoporous membrane and the effect of varying flow rates. A desalination device, with three vertically stacked units is presented to demonstrate the scalability
of the design.

Figure 9 A schematic representation of the microfluidic device at the nanoporous membrane where ICP generates a depletion boundary. Charged species are deflected downwards by the depletion boundary into a separate layer, purifying the top stream. The purified, concentrated, and ground channels are located on distinct layers, which enables stacking for increased functional density.

3.2 Experimental

The out-of-plane ICP process is illustrated in Figure 9. Electric fields were applied through positive electrodes connected to the source water stream and grounded electrodes connected to a channel located above and perpendicular to the source channel. A vertical nanoporous membrane was used to provide a pathway for ion transport between the microfluidic layers. When an electric field was applied, the cation selectivity of the nanoporous membrane generated a depletion effect on the anodic side of the membrane, which redirected ions into the lower
channel and prevented ions from travelling into the top channel, as shown in Figure 9. The result of this ion depletion effect was to produce a purified water stream in the top channel, and a concentrated stream in the lower channel.

Experiments to characterize the effect of the size of the nanoporous membrane and the flow rate were performed using the device shown in Figure 10a. To visualize the depletion effect, the channel below the nanoporous membrane was constructed as a single level with the membrane opening covering a portion of the channel, so that ions were deflected around it and viewed with an inverted microscope. The experiments demonstrating the stackable functionality of the design were performed using the device shown in Figure 10b. The device incorporates a multilayer source channel, as shown in Figure 9, and the purified channel is deviated to allow for visualization of ion removal. Details regarding the materials and device fabrication are in the supporting information at the end of this chapter.

### 3.3 Results and discussion

**Effect of nanoporous membrane geometry**

The dimensions of the nanoporous membrane constrain the transport of cations through it, which can affect the size and effectiveness of the depletion boundary formed during ICP. There is a trade-off between the enhanced transport of ions through a larger nanoporous layer and the desire to keep the depletion effect localized and focused by using a smaller nanoporous membrane. The nanoporous membranes were cut to a length slightly smaller than the channel size (~1.8 mm), and the width values were varied. The experiments were run using the layout shown in Figure 10a with a channel depth of 200 µm and width of 2.0 mm, a flow rate of 0.5 µL/min, and an applied voltage of 5 V. Salt solutions of 20 mM and 200 mM were used for the
Figure 10 (a) Schematic of the microfluidic device used for the characterization experiments to enable visualization of the depletion region. A single channel was fabricated on the bottom layer so an inverted microscope could capture the depletion effect. (b) Schematic of the microfluidic device used to illustrate out-of-plane ICP depletion and deflection of ions, and demonstrate the stackable functionality.
The depth of the depletion boundary was measured using the intensity of the fluorescence signal to determine the percentage of the channel depth that was devoid of ions (in the portion of the channel directly downstream of the membrane). Larger nanoporous membranes resulted in deeper depletion boundaries as shown in Figure 11a. The increase in the depletion boundary size declined as the membrane width was increased, until the effect levelled off at ~125 µm. In these experiments the voltage was set at 5 V and the current was free to fluctuate. The current remained relatively steady for the various widths with an average value of 27 µA for the 20 mM solution and 80 µA for 200 mM. Since larger membranes enable more ion transport they offer a lower electrical resistance in the circuit. With lower resistance in the membrane, the depletion boundary grows in step, increasing the resistance, leading to a relatively constant current (and thus constant power) over a range of membrane widths. These results indicate that the membrane area should be made sufficiently large to achieve the minimum desired depletion boundary size. In the stackable configuration (Figure 10a) the desired size is the full extent of the opening to the purified water channel (Figure 9).

In terms of energy usage per volume of purified water, larger nanoporous membranes are more efficient, as shown in Figure 11b. This is due to larger depletion boundaries generated from larger nanoporous membranes, while the current (and power) remain relatively constant. The improvement in energy efficiency levelled off at ~125 µm for both salinities tested. Beyond that point the required energy was 4.6 Wh/L for the 20 mM solution, and 13.8 Wh/L for 200 mM. These results demonstrate the efficacy of the vertical nanoporous membrane (out-of-plane) approach and indicate the efficiency benefit of larger nanoporous membranes.
Figure 11 Experimental results for various nanoporous membrane widths at a flow rate of 0.5 μL/min and applied voltage of 5 V. (a) Ion depletion percentage is plotted versus nanoporous membrane width. The depletion percentage correlates to the depletion boundary depth, which is shown on the secondary y-axis. The results indicate that larger nanoporous membranes generate larger depletion boundaries. (b) Energy required per litre output is plotted versus nanoporous membrane width.
**Effect of flow rate**

The flow rate through the channel determines how many ions are being transported to the depletion boundary and consequently how many ions are deflected. Experiments were run using the same device as the nanoporous membrane geometry experiments, with a fixed membrane width of 90 µm and an applied voltage of 8 V. The nanoporous membrane was cut to a length shorter than the channel width (1.0 mm) to allow ions to be deflected to the side of the depletion boundary at higher flow rates.

The depth of the depletion boundary decreased as the flow rate was increased, as shown in Figure 12a. The decrease in depth was approximately linear with increasing flow rate such that at each flow rate approximately the same amount of ions were being deflected around the depletion boundary. This effect resulted in consistent outgoing flow rates of the purified water for all of the incoming flow rate values tested. The electrical power was also relatively consistent at all of the flow rates with an average value of 54 µA for the 20 mM solution and 89 µA for 200 mM. As a result of the consistent purified flow rates and electrical power values, Fig. 4b shows that the required energy was the same (within the error) for all flow rates. In summary, these results indicate that varying the flow rate has a negligible impact on the energy efficiency since the depletion boundary deflects the same amount of ions; therefore generating the same amount of outgoing purified water regardless of the incoming flow rate. In a functioning device, the critical aspect is that the depletion boundary is sized to cover the full extent of the purified water channel (Figure 9), by tuning flow rate, and/or membrane width.
Figure 12 Experimental results for various flow rates with a nanoporous membrane width of 90 µm and applied voltage of 8 V. (a) Ion depletion percentage and depletion boundary depth is plotted versus flow rate. The results indicate that the depletion boundary depth decreased in a linear manner as the flow rate was increased. (b) Energy required per litre output is plotted versus flow rate. The energy efficiency was consistent for incoming flow rates due to the consistent flow rate of purified water.
Out-of-plane desalination device

To demonstrate out-of-plane ICP in a functional desalination device experiments were run with the device shown in Figure 10b using a flow rate of 1.0 µL/min. The channel dimensions are listed in the ESI†. The width of the nanoporous membrane in these experiments was 90 µm, and the applied voltage was 20 V. A 500 mM salt solution was used for these experiments since it is a reasonable analog to seawater. As noted above, the multilayered nature of this device, while being necessary from a volumetric efficiency perspective, inherently complicates process visualization. In order to accommodate quantification of the purification process in the multilayered device, the one channel from the upper layer was deviated in-plane, as seen in Figure 13 (this would not be required or desired in an integrated device). As shown in Figure 13, the upper-level stream was visibly devoid of dye, and quantitative fluorescence measurements indicated 95% purity.

In order to demonstrate the scalability of the device the layout shown in Figure 10 with three stacked purification levels was used. Visualization of the fluorescent signal was only possible for the bottom level. Successful operation of all of the levels was confirmed by comparing the electrical current, since an approximately linear increase was measured with values of 60 µA, 110 µA, and 190 µA, for the single, double, and triple layer devices respectively. In all three cases visualization confirmed successful ion deflection for the bottom layer.
Figure 13 Experimental demonstration of out-of-plane desalination. (a) Image of the experiment taken before the voltage was applied and (b) 20 seconds after 20 V was applied. The flow rate was 1.0 µL/min. The source water had a salt concentration of 500 mM and was separated into purified and concentrated layers.

The vertical nanoporous membrane in the out-of-plane design permits an expansion of the channel width in a direction parallel to the length of the membrane without restricting the function of ICP. In previous planar devices, where ions were transported through the nanoporous membrane to a microfluidic channel beside the purification layer, there was a limit to the channel width based on the size of the depletion boundary. Improvements in functional density depend on the operating conditions and scale of the design. For the operating conditions and scale of the
three level device, ~45% of the unit device volume is fluid for the out-of-plane device, whereas previous planar devices were ~15% fluid. The out-of-plane device has a functional density that is three times greater; however, the previous devices were not optimized for functional density. These results indicate a ~50% greater functional density for out-of-plane devices if the planar devices are optimized. In summary, the scalability of the device was demonstrated and the layering enabled significantly greater functional density than planar devices.

3.4 Conclusions

In this work, an out-of-plane, scalable, and energy efficient desalination approach using ICP was demonstrated. Experimental results established that using larger nanoporous membranes for ICP yields larger depletion boundaries and higher energy efficiency. For larger membrane widths (125 – 200 µm), at an applied voltage of 5 V, the required energy was 4.6 Wh/L for the 20 mM solution, and 13.8 Wh/L for 200 mM. Flow rate variation was found to have a negligible impact on energy efficiency since the size of the depletion boundary adjusted to yield a consistent amount of purified water for a given applied voltage. Out-of-plane ICP desalination was demonstrated for 500 mM salt solution. A three level device was presented to demonstrate scalability, and was found to have a 3-fold improvement in functional density over current planar designs. Out-of-plane stacking of microfluidic layers and nanoporous membranes presents opportunities for practical low cost desalination devices.
3.5 Supplementary information

Salt water mixtures

Salt water mixtures were comprised of deionized water with sodium chloride (20, 200, and 500 mM concentrations, EMD Chemicals Inc., Germany), and 0.25× TAE buffer. Fluorescein (Sigma Aldrich, St Louis, MO) and green fluorescing polymer microspheres (9.9 µm diameter, Duke Scientific Corp., Palo Alto, CA) were added to the source water stream to enable visualization.

Material and device fabrication

The vertical nanoporous membrane was fabricated by cutting a slice through a piece of silicone rubber (McMaster Carr, Aurora, OH) using a CO₂ laser (Universal Laser Systems Inc., Scottsdale, AZ), (see Fig. S1). The slice was filled with a solution of 5% Nafion® 117 in a mixture of lower aliphatic alcohols (Sigma Aldrich, St Louis, MO), and placed in an oven to harden at 65 °C for at least 30 minutes.

The microfluidic device was fabricated by micromachining the channels and reservoirs into PMMA using a CO₂ laser. The nanoporous membrane layer was held between PMMA layers by bolts. Producing out-of-plane microfluidic layers and nanoporous membranes with the CO₂ laser permits a wide range of dimensions; therefore, low cost desalination devices can be produced for various applications and capacity demands.

Data acquisition and analysis

Ion depletion was imaged using an inverted fluorescence microscope (DMI 6000B, Leica, NJ). Image sequences were obtained using a CCD camera (Orca AG, Hamamatsu, NJ), and processed
in ImageJ. The fluorescence images were normalized by subtracting a dark field reference image that was created by flowing pure water through the device. Following the normalization procedure, the images were imported into Excel for processing and quantification. Average values were calculated over the region of interest, which encompassed the area downstream of the nanoporous membrane.

**Dimensions of out-of-plane desalination device**

The channels were approximately 500 µm wide, and the depth of the purified and concentrated channels was 230 µm, with a 270 µm layer between them. The incoming source channel had a depth equal to the sum of the two outgoing channels plus the layer between them for a total of 730 µm.
Supplementary figures

Figure 14 Schematic of the procedure for producing the nanoporous membrane. The silicone sheet is cut with a CO₂ laser and the slice is filled with Nafion®. The result is a nanoporous membrane between the fluid layers with a width \( w \) and length \( l \) that can be controlled by the laser settings.
Chapter 4 Life after wetting: Active transport and concentration in paper-based microfluidic devices using ion concentration polarization

This work was done through collaboration with Max Gong, Dr. Brendan MacDonald and Prof. David Sinton. In this work, I developed the four devices to demonstrate analyte transportation/concentration in paper and further analysed the image data and provided the first draft. Then, together with Max Gong, we refined the figures/plots and the text.

4.1 Introduction

Paper-based microfluidics has become a fast growing research field since its beginning in 2007 due to the fact that conventional analytical and clinical detection methods can be integrated in paper [43-46]. Paper is low cost, easily disposed (e.g, incineration), and flexible, which translates to devices that are simple to fabricate, easy-to-use, and portable. These characteristics have led to the use of paper-based devices in various industries, especially targeting the low-resource global health sector. With a growing healthcare burden in many developing countries, there is an increasing need for low cost and simple microfluidic point-of-care diagnostics devices [47-49].

Transport of fluid and analytes in paper-based devices is driven by passive capillary action. A number of methods have been developed to achieve transportation, mixing, separation, pre-concentration and detection of biochemical contents in paper-based devices using passive capillary action [50-56]. In general, liquid transport (and associated analytes transport) in paper depends on the texture and fibre alignment of the pre-manufactured paper, and is limited by
available wetting surface area. Once a paper-based device is saturated with fluid, analytes are fixed in position and further transport/manipulation is not possible.

Mechanisms for flow control during paper wetting have been developed [57]. Hwang et al. [58] recently demonstrated an active flow control mechanism by balancing the capillary force and centrifugal force in a spinning paper. In addition, via surface acoustic waves, uniform mixing along a Y-shaped paper channel can be achieved [59]. However, mechanisms for analyte concentration and transport after wetting have not yet been demonstrated. A novel method for active control of analyte transport and concentration in paper is the ion concentration polarization phenomenon. Ion concentration polarization (ICP) has been extensively used for analyte pre-concentration, separation and desalination [8, 11, 36]. ICP occurs at the interface of microfluidic and nanofluidic structures in the presence of an applied DC electric field. It causes the formation of an ion depletion region at the anodic side of the micro-nano interface against which charged analyte molecules can be efficiently trapped, concentrated and transported. This is mainly due to the development of a large electrical field gradient inside the depletion region and its propagation in the micro-structure [31, 60, 61].

In this chapter, an active transport and concentration of charged analytes in paper-based devices is presented using ion concentration polarization. This approach enables active manipulation of analytes after a paper-based device is saturated with fluid (i.e. after wetting). We present experimental results demonstrating the transport and concentration of charged analytes (e.g. Fluorescein) in saturated paper using ICP. Two types of devices are developed – an external stamp-like device and an integrated in-paper device. The external stamp-like device is a standalone unit that can be used to transport and concentrate analytes by simply placing it on top of any paper device. The integrated paper-based device is patterned with relevant ICP
components (i.e., Nafion nanoporous membrane) such that no external parts are necessary except for a power source.

4.2 Experimental

Working principle

Electric fields were applied via positive and grounded electrodes as indicated in Figure 15. The external stamp-like devices in Figure 15a and Figure 15b contain (1) electrode reservoirs on the top layer which are separated by a plastic insulator, and (2) a Nafion nanoporous membrane layer on the bottom that is used for analyte concentration and transportation. The paper-based devices in Figure 18a and Figure 18b contain (1) a wax boundary to define areas for fluid flow, and (2) a Nafion membrane boundary to concentrate and transport analytes, and to act as a hydrophobic barrier.

The concentration process for the external and integrated devices is illustrated in Figure 15c and Figure 18c. Under the influence of an applied DC electric field the cation selectivity of the Nafion membrane creates a depletion effect at the anodic membrane side and enrichment effect at the cathodic membrane side. Due to the highly charged nature of the membrane, the depletion layer does not reach steady state and its evolution is used to collect analyte molecules in the front of the depletion boundary, focusing them into the center. The concentration process stops when the depletion boundary merges with the enrichment boundary in the central region. Analyte transportation on paper is based on the same mechanism as the concentration process, where transport starts with the evolution of the depletion boundary and ends when the depletion boundary contacts with enrichment boundary.
Figure 15 External paper-based devices. (a) Schematic of the external concentration device. (b) Schematic of the external transportation device. Nanoporous membrane layer on the bottom concentrates and transport charged analytes via application of electric fields as indicated (c) A magnified cross-section of the device where analyte concentration/transportation takes place.
Depletion boundary forms at the interface of the saturated paper and nanoporous membrane, focusing analyte molecules towards the anodic side.

**Materials and device fabrication**

Background electrolyte of 0.25x TAE buffer (10 mM, pH = 8.2) was used for all experiments. Fluorescein (Invitrogen Inc., ON) was used as the model analyte.

The double nanoporous membrane layer for the external device was fabricated by cutting through a piece of silicone rubber (McMaster Carr, Aurora, OH) using a CO\textsubscript{2} laser (Universal Laser Systems Inc., Scottsdale, AZ). The gap was then filled with a solution of 5\% Nafion\textsuperscript{®} 117 in a mixture of lower aliphatic alcohols (Sigma Aldrich, St Louis, MO), and placed in an oven to harden at 65 °C for at least 30 minutes. The fluidic layer containing the reservoirs was micromachined into PMMA using a CO\textsubscript{2} laser. The nanoporous membrane layer was held between PMMA layers by bolts.

The integrated paper-based device was fabricated from 0.18mm thick no.1 Whatman chromatography paper. A Xerox Phaser 8560N color printer was used to pattern the wax boundary, and the patterned paper was heated at 150° C for 1~2 min to allow penetration of the wax though the thickness of the paper [62]. The Nafion membrane boundary was embedded in paper by carefully depositing Nafion\textsuperscript{®} 117 solution via a pipette. The entire device was then soaked in 10 mM TAE buffer for 20 min to ensure the Nafion membrane was fully saturated. Following Nafion membrane saturation the device was exposed to air and dried at room temperature.
Data acquisition and analysis

An inverted fluorescence microscope (DMI 6000B, Leica, NJ) was used to capture analyte concentration and transportation. Image sequences were taken every 5 s with a CCD camera (Orca AG, Hamamatsu, NJ), and processed in ImageJ. To normalize the fluorescence images, a dark field image was subtracted and brightfield normalization was performed. Following the normalization procedure images were imported into Excel for quantification.

4.3 Results and discussion

Demonstration of analyte concentration/transportation with external stamp-like device

Figure 16 shows the experimental results for the concentration of negatively charged fluorescence dye using the external concentration device. The diameter of the central Nafion hole and Nafion circle were fabricated to be 0.12 mm and 8 mm, respectively. Since the target analyte here is negatively charged, it will be subjected to an electrophoretic force towards the anode (i.e, the centre). Thus, to prevent the analyte from leaving the paper substrate and to improve the collection efficiency, the center hole is filled with negatively charged Nafion membrane.

Rectangular filter papers of 20 mm x 20 mm were prepared for the tests. A sample volume of 15uL with initial analyte concentration of 1.5uM in 10mM TAE buffer was deposited on the filter paper. The applied voltage was 50 V. Under these conditions, the time required to concentrate analyte is approximately 3~4 min as shown in Figure 16a. The diameter of the focused area is approximately 1.2mm from an initial diameter of 8 mm, which equates to a surface area concentration factor of ~44-fold (i.e. an upper limit on the concentration factor possible with this geometry). The measured concentration results indicatea peak fluorescence
intensity concentration factor of ~40-fold (90% of the upper limit achievable) as shown in Figure 3a.

The selection of applied voltage is based on previous experiments where we found at lower voltages the time required for concentration is relatively long and at higher voltages the local pH change that occurs at the Nafion hole can affect the fluorescence signal. Figure 16a shows normalized intensity profile obtained through fluorescence imaging at selected time during the concentration of fluorescein. At t = 15s the onset of enrichment is observed at the interface of the central Nafion hole. At t = 50s the onset of depletion is observed at the interface of the Nafion circle. The depletion zone advances somewhat faster from the bottom right side on the paper probably due to the fabrication imperfection of Nafion circle. One of the most notable distinctions between these in-paper results and previous in-channel concentration results [5, 36, 63-65] is the non-linearity of the concentration process. The peak concentration factor can be divided into three stages: (1) before t = 105s, the peak concentration factor is due to the enrichment effect under the Nafion hole and increases in a linear manner; (2) From t = 105s to t = 155s, peak concentration factor increases significantly; and (3) After t = 155, peak concentration factor starts to level off and no significant increase is observed. At t = 105s, the merging encounter of the depletion and enrichment boundaries squeezes analyte molecules together and results in the formation the concentrated ring as shown in Figure 16b. The formation of the concentrated ring is mainly due to non-uniform distribution of electric field gradient across those two boundaries. Inside the ring (i.e, enrichment zone), the electric field gradient is significantly smaller than the outside (i.e, depletion zone); therefore, it is more difficult to transport analyte molecules towards central region. The diameter of focused area is approximately 1.2mm and relatively constant for various initial sample concentrations. A larger
Nafion hole results in larger focused area and lower peak concentration factor because the larger Nafion hole generates larger enrichment zone which limits the propagation of the depletion zone as shown in Figure 16c. Over time due to migration and diffusion the size of focused area slightly shrinks. In this case, the maximum peak concentration factor of ~40-fold is achieved within the paper at \( t = 190s \).
Figure 16 Active concentration using the external device. (a) Time evolution of peak concentration factor. The maximum value reaches 41-fold at $t = 190s$. (b) Contrast enhanced fluorescence image sequence of external concentration device. (c) Fluorescence intensity versus distance plot at selected time points as indicated.
Another device with a 12mm focusing diameter was also fabricated and the results are very similar, indicating the robustness of this concentration approach. Electrokinetic instabilities (EKI) usually occur due to the mismatch of fields and velocities at large conductivity gradients, especially at the depletion boundary[36]. These instabilities occur less here, working within paper, due to the fiber structure inside the chromatography paper, which naturally suppresses flow instabilities, enabling the use of larger focusing diameters and corresponding larger concentration factors.
Figure 17 Active transport using the external device. Fluorescein image sequence and corresponding intensity plot at 50V, (a) showing analytes were concentrated to the right side via application of DC electric field as indicated; (b)-(c) concentrated analytes then was transported ~5mm to the anodic side via application of DC electric field as indicated.
Using the same approach, we developed device to demonstrate analyte transport in a paper strip, as shown in Figure 15b. Paper strip was cut to be 3mm in width and a sample volume of 5uL with initial analyte concentration of 45uM in 10mM TAE buffer was deposited. Target analyte was first concentrated to be a narrow band to increase the visibility, and then transported via charging fluid reservoirs as indicated. This approach provides an active means to manipulate sample on paper other than capillary effect and has potential to be integrated in paper-based analytical devices. As shown in Figure 17, target analyte starts with the initial position at around 1mm and moves to a final position of 5mm in 300 s.
Demonstration of sample concentration and transportation with integrated paper microfluidic device

Figure 18 Integrated paper microfluidic devices. (a) Schematic of the integrated concentration device. (b) Schematic of the integrated transportation device. Nanoporous membrane is embedded in paper matrix to concentrates and transport charged analytes via application of electric fields as indicated (c) A magnified cross-section of the device where analyte concentration/transportation takes place. Depletion boundary forms at the interface of the saturated paper and in paper nanoporous membrane, focusing analyte molecules towards the anodic side.
Figure 19 Active concentration using the integrated paper-based device. (a) Time evolution of peak concentration factor. The maximum value reaches 22-fold at $t = 510s$. (b) Contrast enhanced fluorescence image sequence of integrated concentration device. (c) Fluorescence intensity versus distance plot at selected time points as indicated.
Figure 20 Active transport using the integrated paper-based device. Fluorescence image sequence and corresponding intensity plot at 50V, (a) showing analytes were concentrated to the right side via application of DC electric field as indicated; (b) concentrated analytes then was transported to the left side by reverse the DC polarity.

Figure 19a shows the results for integrated paper concentration device of 1.5uM fluorescein trial. A semi-circle membrane boundary and a rectangular membrane boundary were patterned in paper to trigger ICP, focusing analyte molecules into the center region of the semi-circle. A contour plot of the concentration process at various stages is given in Figure 19b. In this case, the maximum peak concentration factor of 22-fold is achieve at t = 500s. The maximum peak is
lower than that in external device mainly due to the reduced area of focusing region (~32mm²).

Here the maximum expected concentration factor is 28-fold, as dictated by the area ratio of the initial area and the concentrated zone. The time needed to concentrate analyte molecules is two times longer here than with the external device. This is attributed to (1) patterning Nafion in paper resulting in an effective reduction in perm-selectivity of nanoporous membrane; and (2) fluid evaporation from paper hindering the ion transport and resulting in lower ionic current.

Both factors are relevant to the development of ICP for paper-based devices. Figure 19c shows the evolution of depletion region and enrichment region at selected time points. In this case, the enrichment boundary is not well defined and moves ~0.2mm to the right in axial direction. This is mainly due to incomplete filling of Nafion solution in paper matrix, allowing analyte molecules migrate into the paper fiber. Figure 20 demonstrates the analyte transportation using integrated paper-based device. Target analytes were transferred ~5mm in 8 min. Similar to the concentration results, the integrated in-paper device produces similar results to the external device, although a longer time is required when the Nafion is integrated into the paper. In summary, both external and integrated in-paper approaches have advantages and disadvantages. The external device can conveniently be applied to a number of paper-based devices or locations on the same device, while the Nafion in-paper approach requires less external infrastructure at the expense of speed and a more complicated diagnostic strip.

4.4 Conclusions

In this work, an active control mechanism of analytes in paper using ICP has been demonstrated. Both external and integrated in-paper approach behave similarly and both are easy to fabricate. However, external device requires extra external infrastructure to operate while
integrated in-paper device is relative time inefficient. Maximum peak concentration factor of 40-fold and 22-fold can be achieved with external stamp-like concentration device and integrated in-paper concentration device, respectively. Unique features of this concentration approach include (1) nonlinear field effects that confine the sample to a small central region; (2) the radial micro-nano interface has increased interfacial area and increased ionic currents. A new fabrication method for nanofluidics and hydrophobic barriers (Nafion nanoporous boundary) in paper was also demonstrated. Analyte transportation in paper strip offers possible applications in paper-based analytical device which can be further investigated in the future.

Finally, it is also noteworthy that not only is ICP applicable to paper-based microfluidics, paper media provides an advantage suppressing electrokinetic flow instabilities suffered in previous channel-based applications of ICP.
Chapter 5 Conclusions and Future Work

5.1 Conclusions

This thesis investigated two topics – seawater desalination and active control of analyte in paper using ion concentration polarization phenomenon and relevant microfluidic techniques. In the first project, an out-of-plane, scalable, and energy efficient desalination using ICP is demonstrated. Experimental results established that using larger nanoporous membranes for ICP yields larger depletion boundaries and higher energy efficiency. Flow rate variation was found to have a negligible impact on energy efficiency since the size of the depletion boundary adjusted to yield a consistent amount of purified water for a given applied voltage. Out-of-plane ICP desalination was demonstrated for 500 mM salt solution. In the second project, an active control mechanism of analytes in paper using ICP has been demonstrated. Maximum peak concentration factor of 40-fold and 22-fold can be achieved with external stamp-like concentration device and integrated paper concentration device, respectively. A new fabrication method for nanofluidics and hydrophobic barriers (Nafion nanoporous boundary) in paper was demonstrated.

The contributions of this work included: (1) development of a cross chip for vertical nanoporous membrane characterization, and fabrication of stackable water desalination device for proof-of-concept demonstration; (2) development of two external stamp-like devices and two integrated paper microfluidic devices to demonstrate active control mechanism of analyte in paper, post-wetting.
5.2 Future work

Potential optimization strategy for desalination device

1. Find a reliable flow control mechanism to achieve even flow rate in multiple branches, because the size and strength of depletion zone are flow rate dependent.

2. Use wider channel to increase water input, but use shallower channel to ensure the development of ICP.

3. Place electrodes close to the nanoporous membrane, this may further reduce the energy consumption.

4. Use hot embossing or other fabrication techniques to fabricate more smoother channels, this could improve visualization quality, and ultimately reduce costs for scale-up.

Potential optimization strategy for external stamp-like device

1. Fill the central hole with non-permselective membrane, but make sure the pore size is small enough to prevent sample from leaving.

Potential optimization strategy for integrated paper-based microfluidic device

1. Enclose entire device in airtight container to prevent buffer from evaporating.

2. Replace in-paper nanoporous membrane with commercial available membrane to improve perm-selectivity.

3. Integrate paper-based device with point-of-care diagnostic device.
Appendix A

In terms of applications, water usage varies greatly between regions, from 20L to 4654L per person per day. In Toronto, average usage is 566 L/person/day (including York region) [66]. Minimum water requirement estimate for human health and for economic and social development suggests that a country requires a minimum of 135 L per person per day [66, 67]. In developing regions such Africa people use only 20L per person per day [68].

Fouling and biofouling is an issue faced by all desalination technologies. Specifically, reverse osmosis suffers from surface fouling [69]. In the case of the present work, fouling is less of an issue because flow is not forced through the nanopores, rather diverted through microchannels. Besides, charged suspended particulates will be repelled away from nanoporous membrane, this also reduces the fouling possibility. These channels, however, as susceptible to clogging and fouling. While this needs to be tested, we anticipate that the end product will require pre-treatment procedure to remove all debris which could potentially cause clogging and fouling of microchannels. Suggested pre-treatment components are: (1) multimedia filters remove suspended solids; (2) activated carbon filter removes organic matter; (3) one micro filter makes water fit for drink; (4) five micro filters remove dirt and unwanted material.

In terms of scale-up it is important to note that both multiplexing in the vertical and horizontal (in plane) direction will be required. Figure 21 shows a picture of a micropatterned layer with possible optimized unit design. While this was not tested, it provides an indication of how the proposed method could scale, in plane, to achieve high effective volumetric operational density. A device with 850 units (achieved by stacking 100 such layers) would be approximately 70 mm x 90 mm x 200 mm, and have ~1.5 kg of PMMA, and 0.15 kg of Nafion. The total
capital cost would be $1173, which is 92.8% Nafion. Assembly/packaging costs would also be required.”

**Possible optimized unit**

While adding Nafion to the paper increases functionality, it also increases cost. Specifically, typical cost for paper employed in paper-based microfluidics, $0.0045, is $12.5/m². If 20% of that paper was to be impregnated with wax, the price increases to $13.25 /m². If 20% of the paper was to be impregnated with Nafion, the price increases to $99.60/m².

Figure 21 Schematic diagram of possible scalable design

~850 x units on 63mm x 88mm micropatterned layer
References


