THE PENNSYLVANIA STATE UNIVERSITY
SCHREYER HONORS COLLEGE

DEPARTMENT OF BIOCHEMISTRY AND MOLECULAR BIOLOGY

NOVEL FORWARD OSMOSIS DESALINATION TECHNIQUE COMPRISING AN

AQUAPORIN MEMBRANE AND A POLYETHYLENE-GLYCOL-FUNCTIONALIZED

IRON OXIDE NANOPARTICLE DRAW SOLUTE

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ABSTRACT

Improving access to freshwater is a global challenge. Seawater desalination can solve water shortages, but faces significant obstacles as a method of efficient freshwater generation. Here, we construct a novel forward osmosis (FO) system that utilizes polyethylene glycol coated magnetic nanoparticles (PEG-MNPs) as a draw solute for desalination across an aquaporin membrane. Osmotic pressure produced by the draw solute pulls water across the selectively-permeable aquaporin membrane. A magnet can then be used to remove the draw solute from solution, yielding potable water.

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Chapter 1

Introduction

Within the next ten years, half the world's population will live in areas without dependable access to clean water [1]. Towards the objective of avoiding this developing global crisis, it is essential that new means of water distribution and generation are developed with all possible urgency. This study seeks to contribute to the second of these objectives. One of the most common methods currently used for fresh water generation is sea water desalination. There are 18,000 seawater desalination plants worldwide, capable of generating 22.9 billion gallons of water a day. Currently, 300 million individuals "rely on desalinated water for some or all of their daily needs [2]." Thus, seawater desalination presents a potentially feasible, though currently underutilized, means of fresh water generation.

Among the most pressing concerns presented by the growth of the desalination industry is the energy demand of the most commonly utilized desalination technologies, multi-stage flash (MSF) distillation and reverse osmosis (RO) filtration, which, as of 1999, represented 78% and 10% of the world's desalination capacity, respectively [3]. MSF distillation separates salt from water by evaporating water under low pressure and high temperature conditions through consecutive "stages" [3]. Heating is usually accomplished through the use of "low pressure steam externally supplied by a cogeneration power plant." RO filters water by forcing it through a selectively permeable membrane against its concentration gradient at high pressures. Current MSF distillation plants consume power at a rate of approximately 4 kWh/m^3[4], while RO plants consume power at rates of approximately 6 to 8 kWh/m^3[5]. A number of technologies are

currently being developed to decrease the energy demands of MSF and RO plants. Despite these advances, the high energy demand of the FO and MSF desalination techniques may make desalination prohibitively costly in the developing world, and environmentally unsustainable. This thesis seeks to address this challenge by designing a novel means of *forward osmosis* (FO) desalination - a less commonly used filtration technique with potentially lower energy demands.

Commercially available forward osmosis technology generally operates through the use of draw solutes that change phases at temperatures well separate from the melting and boiling points of water. A commonly used salt with these characteristics is ammonium carbonate, which decomposes to ammonia and carbon dioxide gas when heated to 60 $^{\circ}$C [6]. High concentrations of aqueous ammonium carbonate can be used to generate osmotic pressure sufficient to pull fresh water from a feed solution of seawater across a semipermeable membrane. The previously described heating process occurs in a second chamber, yielding liquid water and gaseous ammonia and carbon dioxide. Ventilation can be used to remove the ammonia and carbon dioxide gas produced from this process and recondense it within the draw solution, producing fresh water. A schematic of this process can be found below (Figure 1).
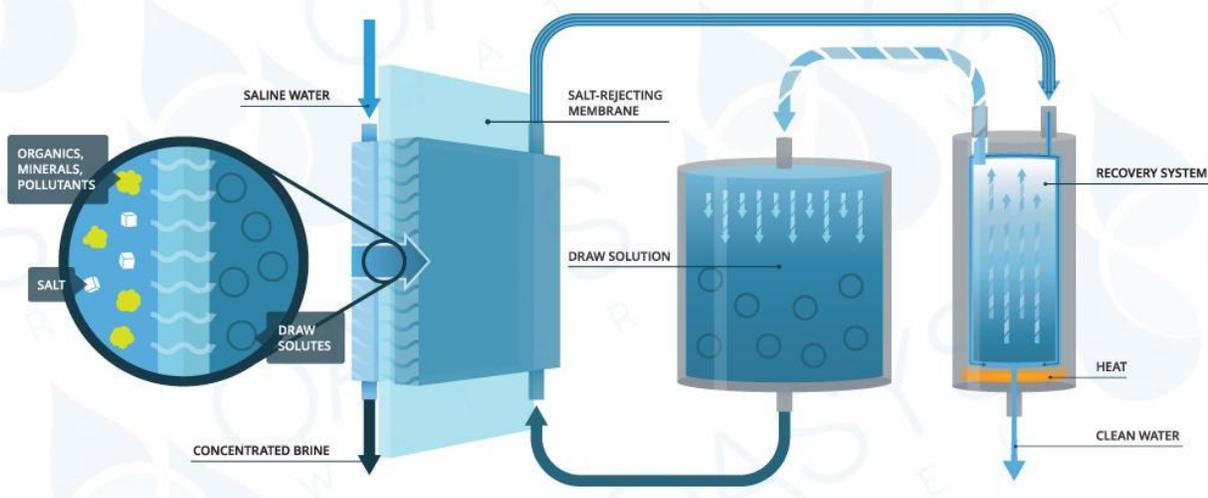

Figure 1 Forward Osmosis Desalination [7].

FO does not have the same energy demands as MSF or RO, because it does not have to boil water as required by MSF, or mechanically generate the pressure gradient required by RO. This does not mean that FO is necessarily a better alternative to either MSF or RO. FO is uniquely vulnerable to some phenomena, such as membrane fouling, that prevent it from achieving the sort of wide-reaching commercial dominance currently held by MSF and RO [13]. However, because of its low energy demand, RO remains an interesting potential source of study.

This thesis specifically seeks to innovate on existing RO technology by determining whether ferromagnetic iron oxide nanoparticle bonded to the carbohydrate polyethylene glycol could viably serve as a draw solute for RO. Ferromagnetic nanoparticles bonded to a carbohydrate would be highly soluble, allowing them to generate osmotic pressure across a semipermeable membrane. Magnetic force could then be used to remove them from solution in the same way heat removes ammonium carbonate salt from solution, generating fresh water. Following the production of fresh water, they could be re-dissolved in the draw solution in order to once again generate osmotic pressure. Thus, in principle, these nanoparticles fulfill the three previously discussed requisite attributes of a FO draw solute - (1) high solubility, (2) easily removable from solution through the use of an external force, and (3) reusable.

There is a demonstrated precedent in the literature for this research - magnetic nanoparticles have been designed in laboratory settings for the expressed purpose of serving as a draw solute for RO [14] [15]. The laboratory of Dr. Tai-Shung Chung in the Department of Chemical and Biomolecular Engineering at the National University of Singapore has been especially productive in this field, publishing the two most highly cited articles on the subject.

However, despite being published in 2010 and 2011, respectively, and receiving hundreds of citations between them, neither of these studies have led to the use of magnetic nanoparticle draw solutes on the commercial level. One potential concern not addressed in the two most influential papers by Chung et al. is the safety of their draw solutes. Ling et al. (2010) designed nanoparticles comprising iron bound to 2-pyrrolidine, triethylene glycol, and polyacrylic acid, and were able to generate flux across a membrane using the osmotic pressure of all three nanoparticle types [14]. Unfortunately, there are safety concerns associated with all three iron-bound chemicals, with pyrrolidine being the most hazardous to human health [16] [17] [18]. As draw solutes degrade over time, it is of utmost importance that they are designed with chemicals that do not negatively impact human health. Thus, although innovative from a technical standpoint, no magnetic draw solute has yet been designed that could viably be used in commercial settings.

Magnetic nanoparticle-like substances have been designed for biomedical applications (for example, for use as "magnetic resonance contrast agents, magnetic drug targeting, or cancer hyperthermia treatment") [12] [19]. Such work became the focal point of this thesis because it demonstrated that soluble iron nanoparticles could be designed that did not negatively affect human health. Garcia-Jimeno et al (2013) published an article that synthesized iron oxide nanoparticles bound to the polyether compound polyethylene glycol (PEG). This process was chosen to serve as the foundation for the nanoparticle synthesis used in this thesis, because of the simplicity of the synthesis described in the article, and because of the widely-established use of PEG in biomedical and biotechnological applications [20]. Thus, based on this background, nanoparticles were synthesized comprising iron oxide bound to PEG. The chemical attributes of

these nanoparticles were analyzed, and their capacity as a draw solute was evaluated. A

description of this process is detailed in the subsequent sections of this thesis.

Chapter 2

Materials and Methods

I.   *Reagents*

Chemicals were used without purification and were reagent grade. Ferric chloride

hexahydrate (FeCl3·6H2O), Ferrous chloride tetrahydrate (FeCl2·4H2O), Polyethylene glycol

(PEG), mw = 6,000 and 10,000, and Ammonium hydroxide (NH4OH, 25%) were purchased

from Sigma-Aldrich (St. Louis, MO). Deionized Millipore Water was used in all experiments.

II.   *Glassware/Setup*

A 250 ml Three-neck round bottom flask, angled (center and side joint size 24/40), a Flow

control adapter stopcock (inner joint size 24/40), and a Pennyhead Stopper (inner joint size

24/40), 2x were purchased from CTechGlass (River Edge, NJ). A 40 ml Mineral oil bubbler was

purchased from Sigma-Aldrich (St. Louis, MO). Nylon Tubing was purchased from McMaster-

Carr (Robbinsville, NJ). A Neodymium-iron-boron (Nd2Fe12B) magnet was purchased from

CMS Magnetics, Inc. (Garland, TX).

III.   *Synthesis of Polyethylene Glycol Coated Iron Oxide Nanoparticles*

PEG (4.5 g, mw = 6000 or 10000) was dissolved in 45°C water (5 mL) under magnetic

stirring. The three-neck round bottom flask was degassed. Pressurized nitrogen gas was run into

the three-neck round bottom flask through nylon tubing and a flow control adapter stopcock, and

back out of the three-neck round bottom flask through a second flow control adapter stopcock,

nylon tubing, and a 40 mol mineral oil bubbler. The water was stirred vigorously for 30 minutes.

Once the PEG was dissolved, $FeCl_3 \cdot 6H_2O$ (0.435 g) and $FeCl_2 \cdot 4H_2O$ (0.16 g) (1:2 molar ratio of

$FeCl_2/FeCl_3$) were added to the three-neck round bottom flask. The water was stirred until the

added $FeCl_3 \cdot 6H_2O$ and $FeCl_2 \cdot 4H_2O$ dissolved. Once the iron salts were well dissolved, 0.75 M

NH$_4$OH (10 mL) was added at a rate of 0.6 mL/min under vigorous mechanical stirring. The stock solution of Ammonium Hydroxide used in this synthesis was calculated to have a concentration of 14.007 M, based on a density of 0.9 g/ml, a formula weight of 35.05 g/mol, and a concentration of 54.55% w/w. To make a 0.75 M solution, 2.142 ml of the stock solution was slowly added to 10 ml deionized water. The solution was adjusted to a final volume of 40 ml with deionized water. 10 mL of NH$_4$OH was manually withdrawn from a 50 ml conical containing 0.75 M NH$_4$OH with an appropriate syringe. Plastic tubing was added to the syringe tip and the plunger was compressed until fluid reached the tip of the plastic tubing. The syringe was fastened to the pump and the pump was set to dispense the 0.75 M NH$_4$OH at a rate of 0.6 mL/min. The suspension was stirred for another 30 minutes at a temperature of 45°C. The ferrofluid was removed from the beaker and washed four times with Deionized Millipore water via magnetic decantation. The nanoparticles were resuspended in 20 mL of Deionized Millipore water, and the solution was sonicated for 12 minutes.

 IV.   *Nanoparticle Characterization*

   Nanoparticle size distribution was determined using a Malvern Zetasizer Nano ZS. Nanoparticles were diluted into solutions with a concentration of 0.05 g/ml. Information about the chemical bonds present in the synthesized nanoparticles were determined using fourier-transform infrared (FT-IR) spectroscopy. A Bruker Vertex V70 FT-IR spectrometer with an optical objective capable of collecting information about the mid-IR range (4,000 - 400 cm-1) was used in this study.
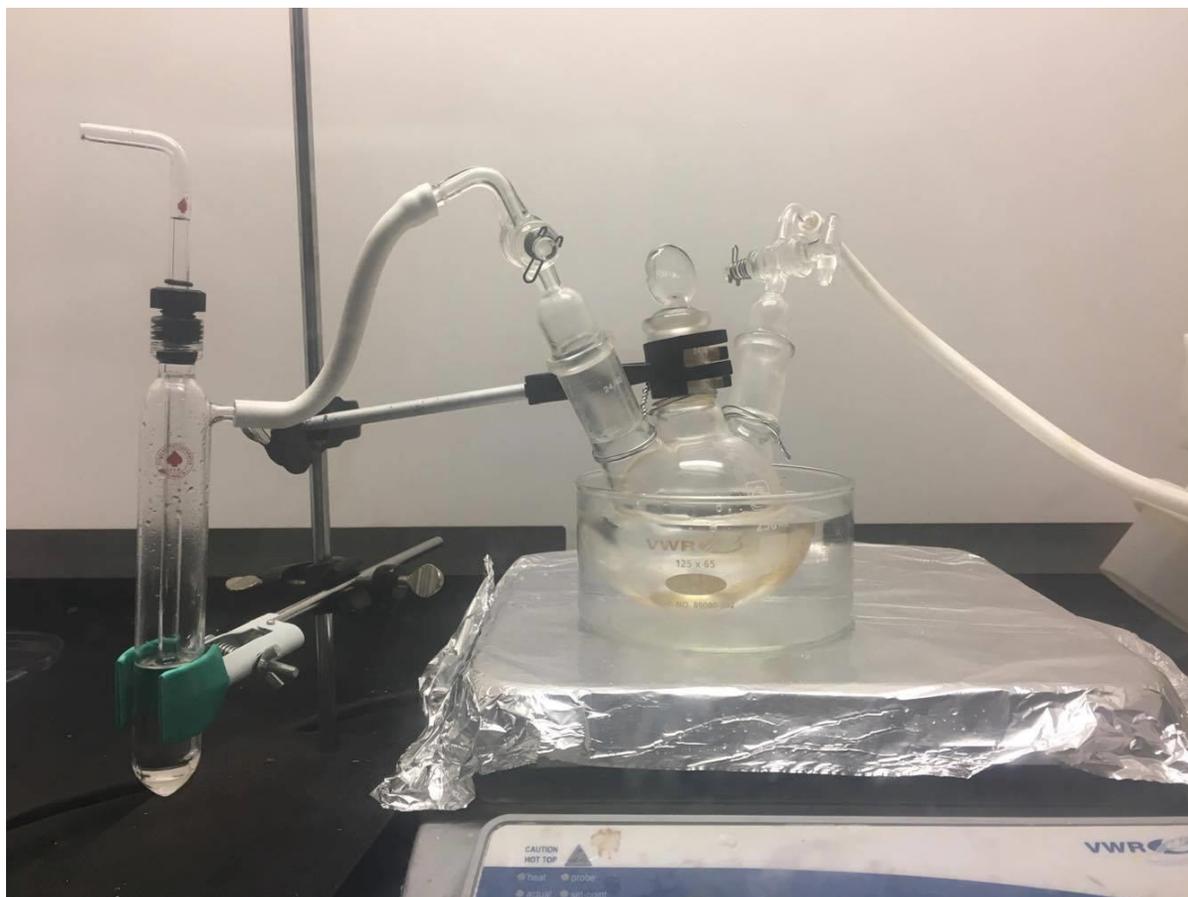
Figure 2 Laboratory Setup

The iron/PEG nanoparticles discussed in this study were synthesized using the following experimental setup. Components from left to right: 40 mL mineral oil bubbler, nylon tubing, flow control adapter stopcock (inner joint size 24/40), 250 ml three-neck round bottom flask (angled, center and side joint size 24/40) in a 500 ml water bath on a heat plate, flow control adapter stopcock (inner joint size 24/40), nylon tubing to pressurized nitrogen gas line.

Chapter 3

Results

I.   **Nanoparticle Synthesis and Analysis**

The first objective following the completed nanoparticle synthesis was to qualitatively

validate the characteristics of the synthesized nanoparticles. Namely, it was essential to

determine whether they could be removed from solution, where they existed as a soluble

ferrofluid, through the use of a magnetic force. A Neodymium-iron-boron ($Nd_2Fe_{12}B$) magnet

was placed on the side of a 50 ml conical containing 5.0 g of nanoparticles dissolved in 20.0 ml

of distilled water. The nanoparticles separated from solution over the course of approximately 40

seconds and stuck to the side of the conical proximal to the magnet (Figure 3). The nanoparticles

also demonstrated the ability to be repeatedly removed from and then dissolved back into

solution. Thus, from a qualitative standpoint, the synthesis was able to generate nanoparticles

with the three most essential characteristics of a FO draw solute. Namely, the synthesized

nanoparticles were (1) highly soluble in water, (2) removable from solution through the exertion

of an external force, and (3) reusable.

The second objective was to evaluate the size distribution of the synthesized

nanoparticles using a zetasizer. The zetasizer functions based on the principle of laser diffraction

[11]. A laser is transmitted through the sample, and the scattering pattern of the laser after it has

passed through the sample is recorded by the instrument. The angular variation in the intensity of

the light scattered is a function of the size of the particles in the sample, and the instrument can

analyze the scattering pattern to calculate the range of particle sizes in the sample indirectly.

A Malvern Zetasizer was used to measure the size of the nanoparticles. Six measurements were taken (Figure 4). Records 1, 2, and 3 were taken from a sample of nanoparticles synthesized using the protocol described in the Materials and Methods Section. The average nanoparticle diameter in Record 1 was 523 nm, in Record 2 was 564.6 nm, and in Record 3 was 556.8 nm. These nanoparticles were approximately an order of magnitude larger than those described by Garcia-Jimeno et al. [12], who measured the hydrodynamic diameter of their PEG-coated iron nanoparticles to be 56 nm using dynamic scattering. In the interest of decreasing the size of the nanoparticles synthesized in this study, the temperature of the synthesis was manipulated, and the effect of temperature on nanoparticle diameter was examined. Decreasing the synthesis temperature from 45 $^\circ$C to 35 $^\circ$C produced nanoparticles that failed to separate from solution under the influence of a magnet. These nanoparticles were thus not analyzed using a zetasizer. Records 4, 5, and 6 were taken from a sample of nanoparticles synthesized at a temperature of 55 $^\circ$C rather than 45 $^\circ$C (Figure 4). The average nanoparticle diameter in Record 1 was 463 nm, in Record 2 was 450.2 nm, and in Record 3 was 455.7 nm. To compare, the average nanoparticle diameter in Records 1 to 3 was 548 nm, while the average nanoparticle diameter in Records 4 to 6 was 456 nm. An unpaired t-test revealed that there was a statistically significant difference (p = 0.0023 < 0.05) between these two values. However, increasing the temperature of the synthesis to 55 $^\circ$C still failed to yield nanoparticles comparable in size to those described by Garcia-Jimeno et al. [12].

The third objective was to determine the bonds present in the synthesized nanoparticles. This information was determined through the use of IR-spectroscopy. An absorbance spectrum between 4000 cm^{-1} and 400 cm^{-1} was obtained for a sample of nanoparticles synthesized using the protocol described in the Materials and Methods section (Figure 5, b). This absorbance

spectrum was then compared to an absorbance spectrum of pure PEG, courtesy of the American

Chemical Society (Figure 5, a). In analyzing their synthesized nanoparticles with IR-

spectroscopy, Garcia-Jimeno et al. (2013) observed a number of bands corresponding to the

bonds present in PEG. Notably, they observed bands at 950 cm^{-1}, corresponding to a C-C stretch,

at 1080 cm^{-1}, corresponding to a C-O-C stretch, at 1132 cm^{-1}, corresponding to a -CH$_2$ twist, and

between 3000 and 3500 cm^{-1}, corresponding to an O-H stretch. Corresponding bands that likely

represented the C-C stretch, the C-O-C stretch, and the -CH$_2$ twist were observed in the present

study at 958 cm^{-1} 1095 cm^{-1} 1147 cm^{-1}. Notably, no band was observed in the current study

between 3000 and 3500 cm^{-1}. Though not entirely consistent with the results published in the

literature, these spectroscopy data strongly suggested that PEG had been successfully bonded to

the iron oxide cores, thus contributing to the solubility of the synthesized nanoparticles.

Figure 3 Soluble and Magnetic Properties of Synthesized Nanoparticles

The nanoparticles were suspended in Deionized Millipore water and then removed from solution with a Neodymium-iron-boron (Nd2Fe12B) magnet. Approximately 5.0 g of nanoparticles suspended in 20.0 mL of Deionized Millipore water were removed from solution over the course of approximately 40 seconds. A video detailing the above process can be found at the following link: https://youtu.be/x8fCjbrkFRM.
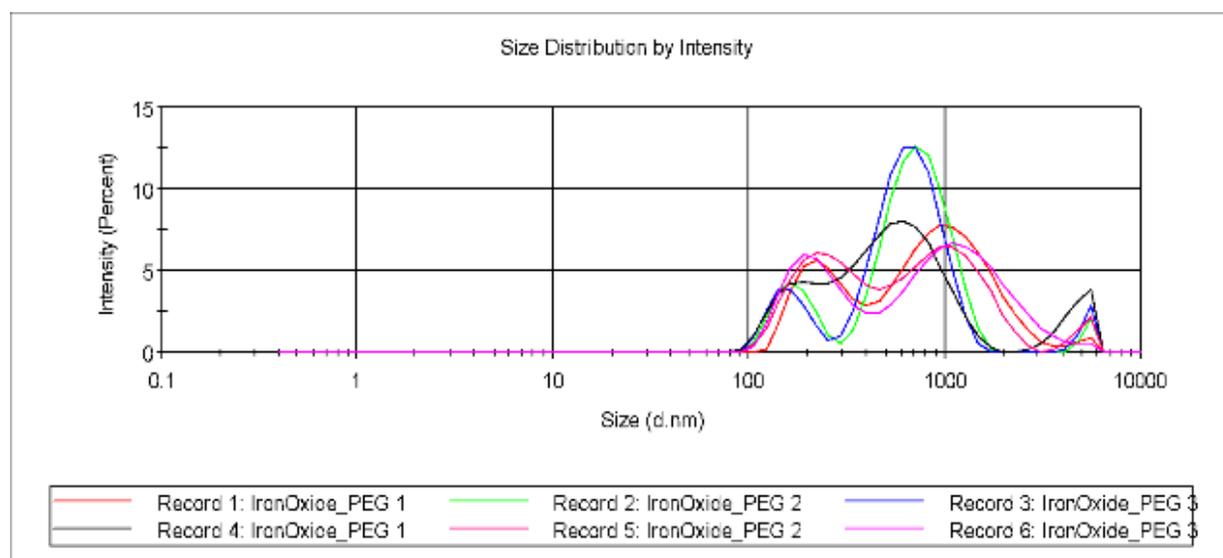
Figure 4 Size Distribution of Synthesized Nanoparticles.

	Reco rd 1	Reco rd 2	Reco rd 3	Reco rd 4	Reco rd 5	Recor d 6
Aver age Size (d.nm)	523	564.6	556.8	463	450.2	455.7

A Malvern Zetasizer Nano ZS was used to determine the size distribution of the synthesized nanoparticles at 45 ºC (Records 1, 2, and 3) and 55 ºC (Records 4, 5, 6). The nanoparticles synthesized at 45 ºC had an average diameter of 548 nm, while the nanoparticles synthesized at 55 ºC had an average diameter of 456 ºC. The full data set displayed in this figure can be found in Appendix I.
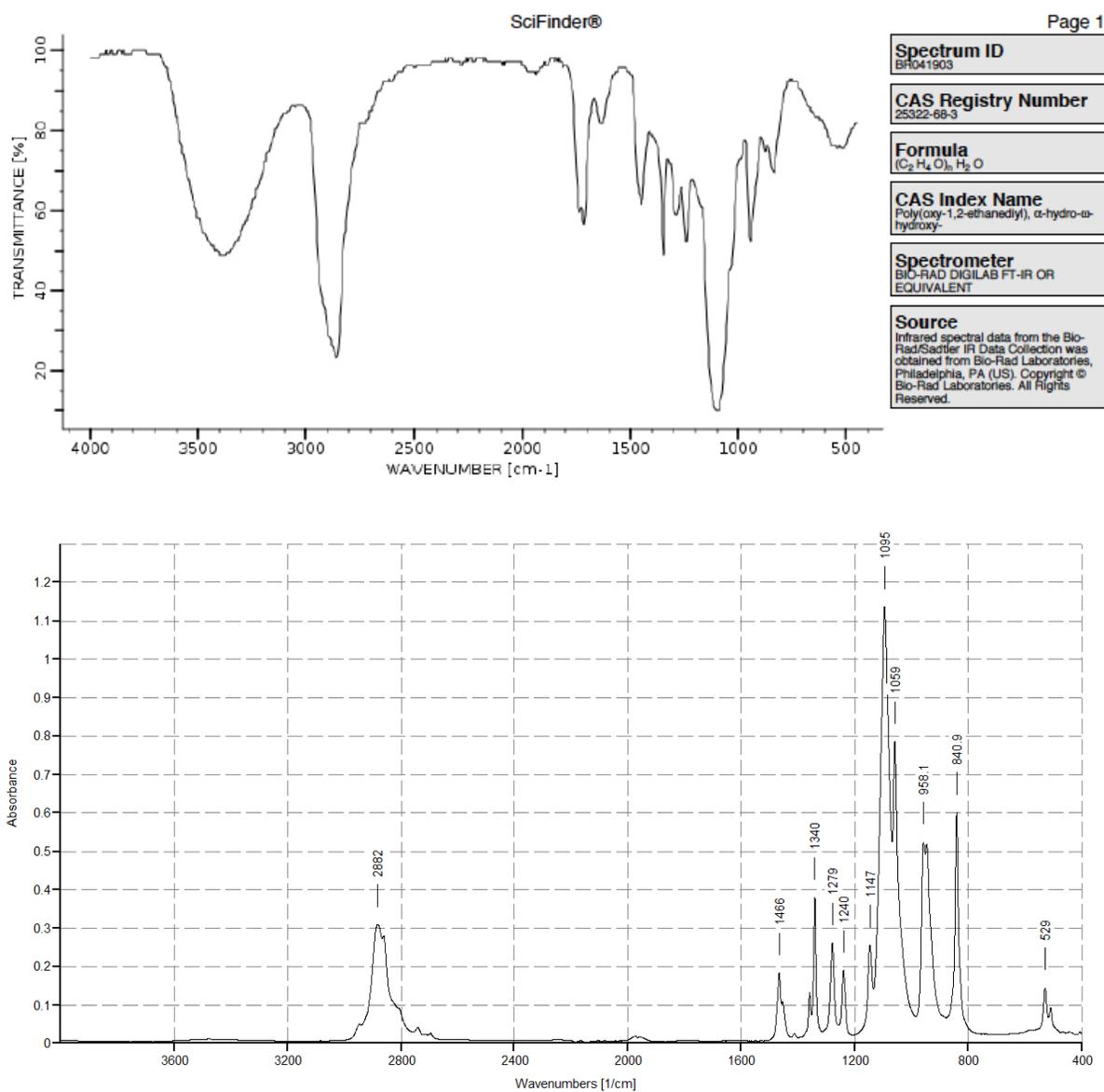
Figure 5 Bonds Present in Synthesized Nanoparticles

An infrared transmittance spectrum of pure polyethylene glycol is displayed above, courtesy of the American Chemical Society. An absorbance spectrum of the nanoparticles synthesized in the current study is displayed below, generated by a Bruker Vertex V70 FT-IR spectrometer.

II.   Aquaporin Membrane Analysis

A FO Flat Sheet Membrane comprising a polyethersulfone base embedded with aquaporin proteins was purchased from Sterlitech Corporation (Kent, WA). There are no known papers or patents in the existing body of scientific literature that discuss a FO method comprising a ferrofluid draw solute and an embedded aquaporin membrane. Aquaporin proteins were chosen for use in this study because of the rapidity with which they can transfer water across a membrane, and also their selectivity for water. Water can flow through a single aquaporin protein at a rate of $3 * 10^9$ molecules per second [23] [24]. This corresponds to a rate of about 18.02 mL of water in 2.5 seconds. Additionally, aquaporins are selectively permeable to water, and capable of rejecting the passage of molecules as small as hydrogen ions [25]. Thus, the ultimate objective of this thesis was to determine whether a membrane with embedded aquaporin proteins could be used with an iron/PEG nanoparticle draw solute as a novel means of FO desalination.

A waterproof vessel comprising two chambers of equal volume, along with a removable frame to separate the two chambers and hold a semipermeable membrane in place, was designed to evaluate the flow of water across the Sterlitech aquaporin membrane (Figure 6, a). The two chambers were filled with equal volumes of Deionized Millipore water and a solution of 1 M NaCl in Deionized Millipore water, respectively. Volume changes between the two chambers were observed on an hourly basis to determine flux across the membrane. The specifications accompanying the membrane list it as capable of achieving flux greater than 7 L/m^2/hr when using the membrane under FO conditions between two chambers containing Deionized Millipore water and a solution of Deionized Millipore water and NaCl at a concentration of 1 M.

Water was permitted to flow across the membrane for 16 hours under these conditions. The experiment was repeated three times and an average height difference of 0.1875 inches between the water levels in the two chambers was observed at the end of the time periods. Average flux across the membrane was then calculated in the following manner:

$$(0.1875 \text{ in} / 16 \text{ hr}) (5.5 \text{ in} / 5 \text{ in}) (0.0164 \text{ L} / 1 \text{ in}^3) = 0.00529 \text{ L/hr}$$

$$(0.00529 \text{ L/hr}) / ((16 \text{ in}^2 * (0.000645 \text{ m}^2) / 1 \text{ in}^2)) = 0.512 \text{ L} / \text{m}^2 / \text{hr}$$

Thus, the membrane achieved a flux of approximately 7% its minimum expected performance under these conditions. If the membrane had achieved a flux consistent with its minimum expected performance, a height change of 0.16 in would have been observed over a time interval of 16 hours. The rationale for this height change is based on the following calculations:

$$(7 \text{ L} / \text{m}^2 / \text{hr}) (16 \text{ in}^2) (0.000645 \text{ m}^2 / 1 \text{ in}^2) = 0.07224 \text{ (L/hr)}$$

$$(0.07224 \text{ L}) (1 \text{ in}^3 / 0.0164 \text{ L}) = 5 \text{ in} * 5.5 \text{ in} * dH$$

Solving for dH yields a dH value of 0.160 in.

Sterlitech Corporation was contacted following the completion of this experiment. A replacement membrane was received.

The flux experiment was repeated using Vessel I with comparable results, and so a new vessel was designed to determine if the membrane was achieving a flux significantly lower than its minimum expected performance due to the design of the first experiment (Figure 7). The flux experiment was then performed with Vessel II (Figure 8). Because the active membrane surface area in Vessel II was 0.01 m^2, a flux of 70 mL / hr was expected, assuming the membrane was capable of achieving a flux consistent with its minimum expected performance.

A flux of 2.22 mL/hr was achieved (3.3% the minimum expected value) with the active surface of the membrane facing the draw solution. In a FO system, the active surface of a membrane should face the draw solution for optimal performance. The flux experiment was repeated with the active surface of the aquaporin membrane facing the feed solution to determine whether the active surface of the membrane had been misidentified. Under these conditions, a flux of 2.6 mL/hr was achieved (3.7 % the minimum expected value). Finally, the reverse flux of salt was measured under both conditions to determine whether the membrane was permeable to NaCl. With the active membrane surface facing the draw solution, a salinity increase of 5 ppt / hr was observed in the feed solution. With the active membrane surface facing the feed solution, a salinity increase of 7 ppt / hr was observed in the feed solution. This result in particular suggests that the Sterlitech membrane did not have the selectivity of pure aquaporin proteins. Thus, it is possible that a lower than expected flux was observed under these conditions because the draw solution was not able to generate the osmotic pressure required to draw 7 L / m^2 / hr across the membrane.

FO Flat Sheet Membrane Filter Specifications

General	
Base Membrane Material	Thin-Film Composite
Membrane thickness	110 micron (±15 micron)
Water flux	>7 L/m²/hr (H_2O vs 1 M NaCl; FO mode)
NaCl reverse flux	<2 g/m²/hr (H_2O vs 1 M NaCl; FO mode)
Boron rejection	>70%
Arsenic rejection	>95%
Operating conditions	Temperature range: 5-50 °C Short term exposure maximum: 65 °C pH range: 2-11 (short term exposure)
Packaging	Stored wet and sealed in a watertight bag.
Storage*	Can be stored at room temperature, but preferably stored at 4 °C. *Do not allow membrane to dry as this will compromise performance
Shelf life	Minimum 6 months

Figure 6 Vessel I used to evaluate flux across a Sterlitech aquaporin FO membrane

(Upper panel) The Vessel I was designed from manually cut plexiglass. The active

membrane area was 16 in², while the dimensions of each of the two chambers was 5.5 x 8.0 x 5.0

in. The membrane contains one active surface, which was oriented towards the draw solution of

Deionized Millipore water and NaCl at a concentration of 1 M. (Bottom panel) The

specifications of the FO Flat Sheet Membrane purchased from Sterlitech Corporation.
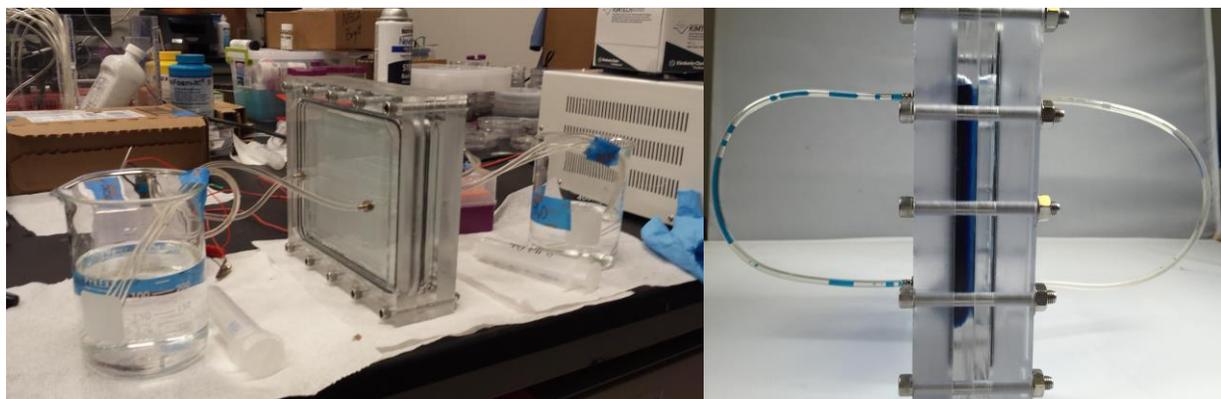
Figure 7 Vessel II used to evaluate flux across a Sterlitech aquaporin FO membrane.

(left) Vessel II was a 3D printed acrylic slab. The active membrane surface area was 10

cm x 10 cm, or 0.1 m^2. Each side of the membrane was connected to 1000 mL beakers

containing initially equal volumes of Deionized Millipore water or a solution of Deionized

Millipore water and NaCl at a concentration of 1 M. Water was circulated across the membrane

from the beakers through the use of a Micro Pump, purchased from Servo Flo (Lexington, MA).

(right) Deionized Millipore water containing blue dye was pumped across one side of the

membrane. The dye remained on one side of the membrane after 24 hours, demonstrating that

Vessel II was watertight.

Active membrane surface facing draw solution, unidirectional flow, 2.0 V in parallel, with massing								0.9975 = water density at 23 C	
	Draw Solution (1 M NaCl)			Feed Solution (Deionized Millipore Water)					
Time (hr)	Approx. Volume (mL)	Weight (g)	Salinity (ppt)	Approx. Volume (mL)	Weight (g)	Salinity (ppt)		Volume Change (mL)	2.2174425
0	300	481.72	54	300	466.71	0			
1	302	483.943	47	295	460.479	5			
Active membrane surface facing feed solution, unidirectional flow, 2.0 V in parallel, with massing								0.9975 = water density at 23 C	
	Draw Solution (1 M NaCl)			Feed Solution (Deionized Millipore Water)					
Time (hr)	Approx. Volume (mL)	Weight (g)	Salinity (ppt)	Approx. Volume (mL)	Weight (g)	Salinity (ppt)		Volume Change (mL)	2.6334
0	300	479.74	53	300	465.404	0			
1	303	482.38	47	295	459.348	7			

Figure 8 Flux observed through Vessel II

The flux of water through Vessel II was measured over the course of 1 hour with the active side facing the draw solution and the feed solution, respectively. Water was pumped across both sides of the membrane at a rate of 93 mL/min. The reverse flux of salt was measured using a refractometer (ppt). The flux was determined by measuring the change in mass of the draw solution over the course of an hour and then converting this value to a volume using the density of water at the temperature at which the experiment was performed (23 ºC).

Chapter 4

Discussion

Discussion

I.   **Nanoparticle Synthesis and Analysis**

The results of this study demonstrated that nanoparticles could be synthesized with the

three requisite attributes of a FO draw solute. Namely, the synthesized nanoparticles were, as

discussed in the Introduction, (1) soluble in water, (2) easily removable from solution through

the use of an external force (in this case through the use of a Neodymium-iron-boron

($Nd_2Fe_{12}B$) magnet), and (3) reusable. Regarding reusability, the synthesized nanoparticles

could be continuously removed from and resuspended in solution through the use of the

Neodymium-iron-boron ($Nd_2Fe_{12}B$) magnet. These observations align with the results of Ling

et al. (2011) [14]. This study demonstrated that a synthesized nanoparticle draw solute was

capable of performing FO across a membrane at 5 distinct time points without losing its soluble

or magnetic properties. Further experiments could be performed to verify the reusability of the

nanoparticles synthesized in this study. Garcia-Jimeno et al. (2013) stored synthesized

nanoparticles at various temperatures and evaluated their properties at the end of a two-year

period; a similar longitudinal experiment could be conducted with the nanoparticles synthesized

in this experiment [12].

The characterization performed on the nanoparticles demonstrated that the synthesis did

not work entirely as intended. The FT-IR analysis performed in this study identified the likely

presence of PEG on the surface of the synthesized nanoparticles. The results of the FT-IR

analysis were supported by the observation that the nanoparticles readily dissolved in solution in

the absence of a magnetic field. However, the synthesized nanoparticles were between 420 nm

and 520 nm larger than comparable iron nanoparticles synthesized by Garcia-Jimeno et al. (2013). The present study demonstrated that increasing the temperature at which the synthesis was performed significantly decreased the size of the synthesized nanoparticles, although not to the desired size of approximately 20 to 30 nm. Further alterations of the nanoparticle synthesis protocol will need to be made in order to determine how to synthesize smaller nanoparticles. Hyeon (2003) suggests that rapid injection of reagents into a synthesis vessel can decrease the size of synthesized iron nanoparticles [12]. Increasing the rate of injection of the reagents used in this experiment into the three-neck flask, as well as increasing the temperature at which they are injected, may decrease the size of the nanoparticles synthesized in this study.

II.   Aquaporin Membrane Analysis

This study was unable to replicate the minimum expected flux of $7 \, L / m^2 / hr$ across a Sterlitech FO aquaporin membrane using a 1 M draw solute. Across two separate experiments, the maximum flux observed under these conditions was just 7% of the minimum expected flux. observed. As a result, the objective of designing a novel FO system comprising a membrane with embedded aquaporin proteins could be used with an iron/PEG nanoparticle draw solute. It is entirely possible that the flux values observed in this study were an artifact of the experimental setup. Further experiments will need to be performed to determine whether the FO aquaporin membranes developed by Sterlitech Corporation are indeed incapable of achieving the claimed minimum expected flux of $7 \, L / m^2 / hr$ in the presence of a 1 M concentration gradient.

Chapter 5

Future Directions

I.   Nanoparticle Synthesis and Analysis

A number of further experiments should be performed in order to evaluate the efficacy of the synthesized nanoparticles as a draw solute. A qualitative experiment was performed in order to verify that the ferrofluid could generate osmotic pressure across a semipermeable membrane and produce flow of water across the membrane, but these results were not quantified. A few experiments would need to be performed in order to quantify these results. First, the molecular weight of the synthesized nanoparticles would need to be determined in order to find the molarity of aqueous solutions containing varying masses of nanoparticles. Currently, static light scattering is one method available at Penn State that can be used to evaluate the masses of nanoparticles. Static light scattering uses the interaction between light and the zeta potential (the surface charge acquired by a molecule when it is dissolved in solution) in order to calculate the molecular weight of microscopic substances. It can be used to measure the molecular weight of polymers, proteins, and nanoparticles, among other types of particles [21].

Once the molecular weight of the synthesized nanoparticles was determined, solutions with known molarities could be created, and placed inside dialysis membranes. These dialysis membranes could then be placed in beakers containing volumes of distilled water. Measuring the volume of water in the beaker periodically could be used to evaluate the flux of water (ml/min) generated by solutions of the ferrofluid with varying molarities. Given the surface area of the dialysis membrane, the flux of water across the membrane (in units of volume per unit area per unit time) could also be calculated. In this way, the efficacy of the synthesized nanoparticles as a draw solute could be validated quantitatively.

II.   Aquaporin Membrane Analysis

It is also possible that the aquaporin membranes designed by Sterlitech are synthesized in a manner that compromises the conformation or filtration capacity of the embedded aquaporin proteins. A number of advances have been made to the deposition of channel proteins into polymer membranes, such as the development of computational models [26], protein-stabilizing polymer scaffolds [28], and protein-stabilizing synthetic vesicles [29], since Sterlitech Corporation began marketing their FO aquaporin membranes in January of 2015 [26] [27]. Notably, most of these advances focus on synthesis methods that emulate wherever possible the environmental conditions of aquaporin proteins in vivo, a recommendation made by the first and most influential research article on synthetic membranes with embedded aquaporin proteins. [30]. As Sterlitech does not make their membrane synthesis protocols publicly available, one essential future experiment is to determine whether the aquaporin proteins present in their commercially available membranes have a conformation consistent with the proteins in vivo. Ultimately, it is hoped that these future directions will help realize the initial objective of a novel FO technique comprising an aquaporin membrane and a polyethylene-glycol functionalized iron oxide nanoparticle draw solute.

Chapter 6

Conclusions

To conclude, iron nanoparticles functionalized with polyethylene-glycol were successfully synthesized. They possess (1) high solubility, (2) easy removability from solution through the use of an external force, and (3) reusability, the three necessary attributes of a forward osmosis draw solute. Further experiments will need to be performed to determine their maximum possible osmotic pressure in solution. Further improvements will also need to be made to the synthesis in order to reduce the size of the synthesized nanoparticles. A Sterlitech FO membrane with embedded aquaporin proteins was also analyzed to determine the flux of water across it in the presence of a 1 M concentration gradient. A maximum flux of between 3% and 7% the minimum expected performance of 7 $L/m^2/hr$ was observed. Further improvements will need to be made to the experimental setup used in this study to determine whether the minimum expected performance can be achieved. Further analysis of the Sterlitech FO membrane should also be performed to determine whether the method by which it is synthesized has an adverse effect of the aquaporin proteins embedded in it.

Appendix

Size Distribution of Synthesized Nanoparticles (Additional Data)

X Intensity	Record 1	Record 2	Record 3	Record 4	Record 5	Record 6
58.8	0	0	0	0	0	0
68.1	0	0	0	0	0	0
78.8	0	0	0	0	0	0
91.3	0	0	0	0.0837	0	0
106	0	0.599	0.926	1.05	0.44	0.359
122	0.0988	2.19	2.56	2.42	1.49	1.76
142	1.62	3.74	3.76	3.56	2.94	3.62
164	3.68	4.32	3.85	4.15	4.43	5.17
190	5.19	3.7	2.91	4.25	5.57	5.9
220	5.63	2.33	1.6	4.15	6.1	5.74
255	5.12	1.02	0.741	4.18	6.01	4.92
295	4.13	0.532	0.972	4.54	5.46	3.83
342	3.22	1.33	2.52	5.27	4.72	2.88
396	2.81	3.41	5.13	6.23	4.11	2.35
459	3.06	6.31	8.18	7.17	3.83	2.36
531	3.9	9.32	10.9	7.84	3.99	2.87
615	5.08	11.6	12.5	8.02	4.53	3.74
712	6.29	12.6	12.5	7.59	5.26	4.75
825	7.25	12	11	6.61	5.96	5.67
955	7.74	9.95	8.22	5.21	6.39	6.33
1.11E+03	7.67	6.96	5	3.63	6.38	6.61
1.28E+03	7.06	3.84	2.2	2.14	5.87	6.47
1.48E+03	6.04	1.39	0.48	0.975	4.91	5.95
1.72E+03	4.76	0.134	0	0.262	3.65	5.14
1.99E+03	3.43	0	0	0	2.32	4.17

2.30E+03	2.22	0	0	0	1.15	3.16
2.67E+03	1.27	0	0	0.0809	0.347	2.23
3.09E+03	0.648	0	0	0.46	0	1.48
3.58E+03	0.365	0	0	1.17	0.088	0.942
4.15E+03	0.366	0	0.0821	2.11	0.587	0.625
4.80E+03	0.553	0.606	1.09	3.06	1.35	0.491
6.44E+03	0	0	0	0	0	0
7.46E+03	0	0	0	0	0	0
8.63E+03	0	0	0	0	0	0

BIBLIOGRAPHY

1. World Health Organization, Drinking-water fact sheet, 2016

2. International Desalination Association, Annual Report, 2015

3. Khawaji, A. D, Kutubkhanah, I. K., Wie, J. Advances in seawater desalination technologies. Desalination, 47-69, 221(1-3), 2008.

4. Awerbuch, L. A. Vision for Desalination-Challenges and Opportunities. Proceedings of the IDA World Congress and Water Reuse, Manama, Bahrain. 2002

5. Moch, I. A 21st Century Study of Global Seawater Reverse Osmosis Operating and Capital Costs. Proceedings of the IDA World Congress on Desalination and Water Reuse, Manama, Bahrain. 2002

6. McCutcheon, J. R., McGinnis, R. L., Elimelaech, M. Desalination by ammonia-carbon dioxide forward osmosis: Influence of draw and feed solution concentrations on process performance. Journal of Membrane Science. 114-123, 278(1-2), 2006.

7. Oasys Water, 2017

8. Murata, K., Mitsuoka, K., Hirai, T., Walz, T., Agre, P., Heymann, J. B., Engel, A., Fujiyoshi, Y. Structural determinants of water permeation through aquaporin-1. Nature. 599-605, 407, 2000.

9. Kumar, M., Grzelakowski, M., Zilles, J., Clark, M., Meier, W. Highly permeable polymeric membranes based on the incorporation of the functional water channel protein Aquaporin Z. PNAS. 20719-20724, 104(52), 2007.

10. Wang, H. Chung, T., Tong, Y. W., Jeyaseelan, K., Armugam, A., Chen, Z., Hong, M., Meier, W. Highly Permeable and Selective Pore-Spanning Biomimetic Membrane Embedded with Aquaporin Z. Small. 1185-1190, 8, 2012.

11. Eshel, G., Levy, G. B., Mingelgrin, U., Singer, M. J. Critical Evaluation of the Use of Laser Diffraction for Particle-Size Distribution Analysis. Soil Science Society for America Journal. 736-743, 68, 3. 2003.

12. Garcia-Jimeno, S., Estelrich, J. Ferrofluid based on polyethylene glycol-coated iron oxide nanoparticles: Characterization and properties. 74-81, 420(5), 2013.

13. Cornelissen, E. R., Harmsen, D., de Korte, K. F., Ruiken, C. J., Qin, J. J., Oo, H., Wessels, L. P. Membrane fouling and process performance of forward osmosis membranes on activated sludge. Journal of Membrane Science. 158-168, 319(1-2), 2008.

14. Ling, M. M., Wang, K. Y., Chung, T. S. Highly Water-Soluble Magnetic Nanoparticles as Novel Draw Solutes in Forward Osmosis for Water Reuse. Industrial & Engineering Chemistry Research. 5869-5876, 49(12), 2010.

15. Ling, M. M. Chung, T. S., Lu, X. Facile synthesis of thermosensitive magnetic nanoparticles as "smart" draw solutes in forward osmosis. Chemical Communications 10788-10790, 47(38), 2011.

16. National Center for Biotechnology Information. PubChem Substance Database; SID=329769283

17. National Center for Biotechnology Information. PubChem Substance Database; SID=24900333

18. National Center for Biotechnology Information. PubChem Substance Database; SID=24850924

19. Brullot, W., Reddy, N. K., Wouters, J., Valev, V. K., Goderis, B., Vermant, J., Verbiest, T. Versatile ferrofluids based on polyethylene glycol coated iron oxide nanoparticles. Journal of Magnetism and Magnetic Materials. 1919-1925, 324(11), 2012.

20. Inada, Y., Matsushima, A., Kodera, Y., Nishimura, H. Polyethylene Glycol (PEG)-Protein Conjugates: Application to Biomedical and Biotechnological Processes. Journal of Bioactive and Compatible Polymers. 343-364, 5(3), 1990.

21. Burchard, W. Static and dynamic light scattering from branched polymers and biopolymers. Light Scattering from Polymers. Advances in Polymer Sciences, vol. 48. Springer, Berlin, Heidelberg. 1983.

22. Hyeon, T. Chemical synthesis of magnetic nanoparticles. The Royal Society of Chemistry. 927-934, 2003.

23. Zeidel, M. L., Ambudkar, S. V., Smith, B. L. Agre, P. Reconstitution of functional water channels in liposomes containing purified red cell CHIP28 protein. Biochemistry. 7436 - 7340, 31(33), 1992.

24. Van Hoek, A. N., Verkman, A. S. Functional reconstitution of the isolated erythrocyte water channel CHIP28. Journal of Biological Chemistry. 18267 - 18269, 267, 1992.

25. Ilan, Boaz, Tajkhorshid, E., Schulten, K., Voth, G. A. The mechanism of proton exclusion in aquaporin channels. Proteins: Structure, Function, and Bioinformatics. 223 - 228, 55(2), 2004.

26. Jefferys, E., Sands, Z., Shi, J., Sansom, M. S. P., Fowler, P. Alchemid: A Computational Method for Incorporating Multiple Proteins into Complex Lipid Geometries. Journal of Chemical Theory and Computation. 2743 - 2754, 11(6), 2015.

27. Sterlitech distributes Aquaporin FO membrane. Membrane Technology. 5, 2015(1), 2015.

28. Hu, Z., Ho, J. C. S., Nallani, Madhavan. Synthetic (polymer) biology (membrane):

    functionalization of polymer scaffolds for membrane proteins. Current Opinion in

    Biotechnology. 51 - 56, 46, 2017.

29. Beales, P. A., Khan, S., Muench, S. P., Jeuken, J. C. Durable vesicles for reconstitution

    of membrane proteins in biotechnology. Biochemical Society Transactions. 15 - 26,

    45(1), 2017.

30. Kumar, M., Grzelakowski, M., Zilles, J., Clark, M., Meier, W. Highly permeable

    polymeric membranes based on the incorporation of the functional water channel protein

    Aquaporin Z. Proceedings of the National Academy of Sciences. 20719 - 20724. 104(52),

    2007.

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EDUCATION

Pennsylvania State University, University Park, PA

Schreyer Honors College   May 2017
  ● Bachelor of Science in Biochemistry and Molecular Biology

HONORS AND AWARDS

Phi Beta Kappa Society   2016
          Induction into the oldest honors society for the liberal arts and the sciences in the United
States
          Presidential Leadership Academy Research Scholarship   2016
          Funding to support undergraduate research in the lab of Dr. Tak-Sing Wong
Undergraduate Research Exhibition Award   2016
          Award presented to best presentations at the Penn State Undergraduate Research
          Exhibition; presented for poster entitled "Urinalysis Screening Test Strips for Diabetes," later
          published as "Arnett, N. et al. (2016)"
Undergraduate Research Travel Grant   2016
          Funding provided by the Schreyer Honors College and the Eberly College of Science to
          support attendance at the 2016 Global Humanitarian Technologies Conference in Seattle,
          Washington
Morrow Family Endowed Prize   2015
          Academic scholarship provided by the department of Biochemistry & Molecular Biology
          on the basis of excellence in the Eberly College of Science
The President Sparks Award   2015
          Award presented to sophomores who have earned a 4.00 cumulative grade-point average
Student Leadership Scholarship   2015
          Academic scholarship provided by the Pennsylvania State University for significant
          leadership as founder of the Undergraduate Research Society, and for the organization's
          contribution to the university community
Eberly College of Science Undergraduate Research Award   2014 - 2016
          Funding to support research in the lab of Dr. Moriah Szpara
          Schreyer Honors College Travel Grant   2014 - 2016
          Academic scholarship for students in the Schreyer Honors College incorporating
          international study into their undergraduate degree
Eberly College of Science Travel Grant   2014
          Academic scholarship for students in the Eberly College of Science incorporating
          international study into their undergraduate degree
The President's Freshman Award   2014
          Award presented to freshman who have earned a 4.00 cumulative grade-point average
Schreyer Ambassador Travel Grant   2014
          Academic scholarship provided by the Schreyer Honors College incorporating
          international study into their undergraduate degree
Braddock Scholarship   2013 - Present
          Academic scholarship given to the most exceptional freshmen entering the Eberly
          College of Science

Schreyer Scholarship for Academic Excellence   2013 - Present

Academic scholarship on the basis of maintaining at least a cumulative 3.4 GPA and meeting all honors requirements of the Schreyer Honors College

National Merit Scholarship   2013

Academic scholarship given yearly to approximately 8,000 students nationally on the basis of academic record, written recommendation, leadership outside the classroom, and an essay

RESEARCH EXPERIENCE   Pennsylvania State University, University Park, PA

Research Assistant, Dr. Tak-Sing Wong   Aug. 2015 - Present
- Polyethylene Glycol Coated Magnetic Iron Oxide Nanoparticles for Forward Osmosis Desalination Across an Aquaporin Membrane

Research Assistant, Khanjan Mehta, M.S.   Aug. 2015 - Present
- Arnett, N., Vergani, A., Winkler, A., Ritter, S., Stapleton, J., Mehta, K. Inexpensive Urinalysis Test Strips to Test for Diabetes. *GHTC*. 2016.
- Arnett, N., Bseiso, A. Opalinski, N. Mehta, K. An Analytical Framework for Selecting Desalination Technologies for Use in Low Resource Settings. (submitted to *Water Research*)
- Vergani, A., Arnett, N., Sen, S., Obeysekare, E. Recommended distribution Methods for Screening Test Strips in Low Resource Settings. (in review)

Research Assistant, Dr. Moriah Szpara   Apr. 2014 - Aug. 2015
- Collaborate with technicians and graduate students to discern the mechanism of Herpes Simplex Virus infection and virulence in the nervous system
- Proficient in many wet lab techniques, including assays and titers, as well as the appropriate navigation of a Biosafety Level 2 laboratory

RESEARCH INTERNSHIPS

Research Assistant, Dr. Ian Maze   May - Aug. 2016
- Funded by the Icahn School of Medicine at Mount Sinai Summer Undergraduate Research Program
- Identified a neurotransmission-independent role for serotonin as a histone modification

Research Assistant, Dr. Donald Defranco   May - Aug. 2015
- Funded by the University of Pittsburgh School of Medicine Summer Undergraduate Research Program
- Developed a novel in vitro model for the blood brain barrier for use in pharmacology research

STUDY ABROAD EXPERIENCE

BIOL 499A: Tropical Field Ecology (Costa Rica)   Dec. 2015 - Jan. 2016
Dr. James Marden, Dr. Tomas Carlo
- Conducted faculty-led research projects on the interactions of organisms and their environments

THEA 490: London Study Tour (United Kingdom)   Dec. 2014 - Jan. 2015
Dr. Beverly Patton, Dr. Raymond Sage
- Studied the ways various genres of musical theatre and drama examine areas of diversity such as gender, race, sexuality, politics, and religion

BIOL 498 A: Biology of Eco-Health (Tanzania)   May - June 2014
Dr. Douglas Cavener
- Studied the intersection of ecology, conservation policy, and human health under the guidance of Penn State faculty member Dr. Douglas Cavener

TEACHING EXPERIENCE   Pennsylvania State University, University Park, PA

Learning Assistant, BMB 402, General Biochemistry II   Jan. - May 2017
Dr. Timothy Meredith

Teaching Assistant, BMB 464, Molecular Medicine   Aug. - Dec. 2015
Dr. James Howell

Grader, CHEM 110H, Honors General Chemistry   Aug. - Dec. 2014
Dr. Philip Bevilacqua

Grader, ECON 302, Intermediate Microeconomic Analysis   Jan. - May 2016
Dr. Mark McLeod

LEADERSHIP EXPERIENCE   Pennsylvania State University, University Park, PA

The Undergraduate Research Society
Founder and President   Sept. 2013 - Present
- Created community for undergraduate research at Penn State with over 100 active members
- Founded U-RISE program to teach undergraduates the foundations of research and connect undergraduates to permanent positions in research labs
- Host weekly lectures for undergraduates on faculty research and professional development

The Presidential Leadership Academy
Zeta Class Member   Mar. 2014 - Present
- Cultivate leadership fundamentals through discussion, writing, and field experiences
- Led panel of students and university officials to review and revise campus sexual assault policy
- Complete three year program endowed by Ed Hintz, President of Hintz Capital Management, Inc., taught by Dr. Christian Brady, Dean of the Schreyer Honors College, and Penn State President Dr. Eric Barron

The Penn State Club Squash Team
Treasurer and Traveling Member   Sept. 2013 - Present
- Competed in matches against Bucknell University, Lehigh University, Cornell University, Siena College, Ithaca College, and Johns Hopkins University
- Managed the expenses of the Club Team, petitioned Penn State University for funding

Science LionPride
Member and Tour Guide   Sept. 2013 - Present
- Student ambassador for the Eberly College of Science
- Tour guide for families of prospective students of the Eberly College of Science

SHO TIME 2014, 2015
Team Leader and SHO TIME Mentor   Feb. 2014 - Aug. 2015
- Plan new student orientation for the Schreyer Honors College
- Organize Spirit Dinner, Late Night With The Dean, and Final Bow events for new student orientation
- Coordinate the roles of 13 SHO TIME Mentors

The Penn State Debate Society
Treasurer   Sept. 2013 - May 2014
- Hosted and debated the British National Debate Team
- Responsible for petitioning Penn State University for funding

SERVICE EXPERIENCE   Pennsylvania State University, University Park, PA

Volunteer at the Mount Nittany Medical Center, State College, PA
Patient Floor and Emergency Department Volunteer   Aug. 2010 - Present
- Train new volunteers on duties necessary for effective volunteer work
- Comfort, direct, and escort patients, family, and visitors
- Transport confidential health documents and ensure patient privacy
- Maintain well-stocked, sanitary, comfortable environment for patients and hospital staff
- 400+ hours on duty

Volunteer through The Penn State Dance Marathon, University Park, PA   Sept. 2014 - Feb. 2015
Rules and Regulations Committee
- Organized event schedule and safety procedures for a 46 hour dance marathon as part of the largest student-run philanthropic organization in the world, raising over $13 million in 2015
- 200+ volunteer hours

Volunteer through The Penn State Urban Service Experience, Washington, D.C.
Volunteer   Nov. 2013
- Prepared a Thanksgiving dinner for the homeless shelter at The Church of the Epiphany in the Episcopal Diocese of Washington, D.C.
- Assisted the local community by stocking the Capital Area Food Bank and planting trees in Marvin Gaye Park
- 25+ volunteer hours

Volunteer through Habitat for Humanity, White Sulfur Springs, W.V.
Volunteer   July 2012; July 2013
- Constructed roofing, tiling, siding, and electrical infrastructure for local, affordable housing
- 100+ hours of volunteer work