The diffusion of divalent nickel (Ni$^{2+}$) from wet silica gels was investigated. Silica gel is gaining interest as an encapsulation matrix for biological components. The transport of biologically relevant species within the gel is determined by the structural characteristics of the gel, which are in turn governed by synthesis parameters. Gels were synthesized by an acid-base two step process from tetraethoxysilane (TEOS) precursors. Organically modified siloxane precursors, including methyltriethoxysilane (MTES), dimethyldiethoxysilane (DMDES), trimethylethoxysilane (TMES), and ethyltriethoxysilane (ETES) were also used for some samples at a concentration of 10 molar % of silicon. PEG200 was used as an additive in some samples. Sample space covered a full factorial design of three water ratios during hydrolysis of 4:1, 10:1 and 20:1, three acid catalyst concentrations as a ratio of silicon to acid, including 1:0.005, 1:0.01, and 1:0.02, and four dilution ratios during gelation to yield gels with a final silica content of 40:1, 60:1, 80:1, and 100:1, moles of water to moles of silicon. This processing space was selected due to its relevance to applications in the encapsulation of biological components.

Using Ni$^{2+}$ as a tracer due to its strong absorbance peak at 395 nm, diffusion coefficients were calculated for all samples using both an analytical solution to Fick’s Law, appropriate for one-dimensional diffusion, and an exponential empirical approximation. Estimates were calculated using Microsoft Solver and ANOVA in SAS. It was found that the diffusion coefficient in TEOS gels ranged from approximately $1.4 \times 10^{-10}$ m$^2$s$^{-1}$ to $6.3 \times 10^{-10}$ m$^2$s$^{-1}$, with a mean of approximately $2.5 \times 10^{-10}$ m$^2$s$^{-1}$.
corresponding to approximately 14% to 63% of D for Ni$^{2+}$ in unconfined aqueous solution, estimated to be approximately $1 \times 10^{-9}$ m$^2$s$^{-1}$. The addition of 10 mol% ORMOSILS was found to have a small effect on the predicted value of the diffusion coefficient depending on silicon content. In samples with a final silicon content of 80:1, D was slightly decreased to approximately $2.0 \times 10^{-10}$ m$^2$s$^{-1}$, but in samples with a silicon content of 100:1, D was slightly increased to approximately $3.5 \times 10^{-10}$ m$^2$s$^{-1}$. Variations in hydrolysis ratio, acid catalyst content, and dilution ratio had relatively weak effects on overall diffusion rates of Ni$^{2+}$ with the exception of a few anomalous samples which were either unstable or displayed some syneresis. It can be concluded that over this broad processing space, gels can be tailored to best suit the particular bioencapsulation application, altering the chemical environment for optimal performance with minimal variation in the diffusion transport of small cationic ions such as Ni$^{2+}$. 
©Copyright by David J. Dickson
June 9th, 2010
All Rights Reserved
Influence of Processing Parameters on Diffusion of Divalent Nickel in Wet Silica Sol-Gel Monoliths

by
David J. Dickson

A THESIS
Submitted to
Oregon State University
in partial fulfillment of
the requirements for the
degree of
Master of Science

Presented June 9, 2010
Commencement June 2011

APPROVED:

_________________________
Major Professor, Representing Materials Science

_________________________
Director of the Materials Science Program

_________________________
Dean of the Graduate School

I understand that my thesis will become part of the permanent collection of Oregon State University libraries. My signature below authorizes release of my thesis to any reader upon request.

_________________________
David J. Dickson, Author
ACKNOWLEDGEMENTS

I would like to express my sincere thanks to my committee members, especially Professors Alex Yokochi and Roger Ely. First, for indulging me in what may have been a foolish request to concurrently pursue two graduate degrees. I'm still alive, and it looks like I might actually survive the PhD defense in the coming year! Secondly, for helping me develop ideas and experimental work plans, meeting when asked, providing thoughtful feedback, and then giving me room to run without overbearing supervision. I learn best this way, and I very much appreciate the trust.

My sincere thanks to Professor Catherine Page at University of Oregon, who, although not officially part of my committee, may as well have been. As a collaborator on this sol-gel research, she has facilitated many productive conversations and interesting electron microscopy work.

Many thanks to Tia Gabalita for her incredibly generous help with the statistical analysis, and for running me into the ground in the Mac Forest!

I would like to thank my lab mates, past and present, including Jed Eberly, Mark Luterra, Elizabeth Burrows, and Paul Schrader. These are brilliant wonderful people who are a pleasure to work with, which makes spending time in the lab quite easy. The lab staff, including Kelsey Ward, Kelsey Baker, Mark McGuire, Siri Erikson, Ann Wynn, and Matt Galvin, were also incredibly helpful with lab upkeep, experimental set-up, and data collection.

The Material Science Department always made me feel welcome, even though it was a second home after Biological Engineering. Professors Brady Gibbons, David Cann, and Bill Warnes were particularly welcoming and helpful with my pursuits of a Materials Science degree.

Lastly, many thanks to those who helped distract me and maintain my mental health, especially my partner, Erica McKenzie, my friends, Ram Ravichandran, Matt Schmidt, Andrea Fideler, and Keir Thomas, the Triathlon Club, both the Men’s and Women’s Rugby Clubs, and the Corvallis running and triathlon communities.
## TABLE OF CONTENTS

<table>
<thead>
<tr>
<th>Chapter 1 – Introduction and Project Objectives</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.1 Introduction</td>
<td>1</td>
</tr>
<tr>
<td>1.2 Objectives</td>
<td>5</td>
</tr>
</tbody>
</table>

| Chapter 2 – Literature Review                  |      |
| 2.1 Introduction                               | 7    |
| 2.2 Chemistry of Silicon and Silica           | 8    |
| 2.3 Sol-gel Chemistry                          | 10   |
| 2.4 Aqueous Precursors                         | 11   |
| 2.5 Alkoxide Precursors                        | 13   |
| 2.5.1 Acid-Catalyzed Hydrolysis                | 13   |
| 2.5.1 Base Catalyzed Hydrolysis                | 16   |
| 2.5.2 Acid-Catalyzed Condensation              | 18   |
| 2.5.3 Base-Catalyzed Condensation              | 18   |
| 2.5.4 Gelation                                 | 20   |
| 2.5.5 Summary of Relevant Silica Sol-Gel Investigations | 21   |
| 2.5.6 Kinetics and Structural Modeling         | 26   |
| 2.6 Gel Aging                                  | 30   |
| 2.6.1 Ostwald Ripening & Coarsening            | 30   |
| 2.6.2 Capillary Forces                         | 33   |
| 2.7 Organically Modified Silicates             | 35   |
| 2.8 Additives                                  | 36   |
| 2.9 Diffusion                                  | 37   |
| 2.9.1 Mathematical Treatment                  | 37   |
## LIST OF FIGURES

<table>
<thead>
<tr>
<th>Figure</th>
<th>Description</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>Figure 1-1</td>
<td>Ternary phase diagram of silica sol-gel processing space.</td>
<td>3</td>
</tr>
<tr>
<td>Figure 2-1</td>
<td>Silica tetrahedron.</td>
<td>9</td>
</tr>
<tr>
<td>Figure 2-2</td>
<td>Mechanism of acid catalyzed hydrolysis (adapted from [4]).</td>
<td>16</td>
</tr>
<tr>
<td>Figure 2-3</td>
<td>Mechanism of base catalyzed hydrolysis (from [4]).</td>
<td>17</td>
</tr>
<tr>
<td>Figure 2-4</td>
<td>Electron donating characteristics of various silicon side groups.</td>
<td>19</td>
</tr>
<tr>
<td>Figure 2-5</td>
<td>Ternary phase diagram (A).</td>
<td>22</td>
</tr>
<tr>
<td>Figure 2-6</td>
<td>Ternary phase diagram (B).</td>
<td>23</td>
</tr>
<tr>
<td>Figure 2-7</td>
<td>Matrix representation of next nearest neighbor speciation [4].</td>
<td>27</td>
</tr>
<tr>
<td>Figure 2-8</td>
<td>(A) Polymeric network, and (B), a particulate network.</td>
<td>29</td>
</tr>
<tr>
<td>Figure 2-9</td>
<td>Ostwald ripening.</td>
<td>31</td>
</tr>
<tr>
<td>Figure 2-10</td>
<td>Coarsening through necking.</td>
<td>33</td>
</tr>
<tr>
<td>Figure 2-11</td>
<td>Schematic of idealized 1-D diffusion.</td>
<td>40</td>
</tr>
<tr>
<td>Figure 3-1</td>
<td>Common alkoxide precursors.</td>
<td>49</td>
</tr>
<tr>
<td>Figure 3-2</td>
<td>An illustration of a typical gel sample.</td>
<td>53</td>
</tr>
<tr>
<td>Figure 3-3</td>
<td>Ni(^{2+}) absorbance calibration curve.</td>
<td>57</td>
</tr>
<tr>
<td>Figure 3-4</td>
<td>Diffusion analysis experimental set-up.</td>
<td>58</td>
</tr>
<tr>
<td>Figure 3-5</td>
<td>Illustration of a typical color change.</td>
<td>59</td>
</tr>
<tr>
<td>Figure 3-6</td>
<td>Semi-quantitative illustration of Ni(^{2+}) concentration profile.</td>
<td>61</td>
</tr>
<tr>
<td>Figure 4-1</td>
<td>Diffusion data from 100:1 TEOS gels at 0.005 acid ratio, comparing different water ratios</td>
<td>62</td>
</tr>
<tr>
<td>Figure 4-2</td>
<td>Diffusion data from TEOS gels prepared with a 10:1 water ratio.</td>
<td>64</td>
</tr>
<tr>
<td>Figure 4-3</td>
<td>The first 60 minutes of the same data shown in Figure 4-2(B).</td>
<td>65</td>
</tr>
<tr>
<td>Figure 4-4</td>
<td>Increased Ni(^{2+}) transport in low water ratio, high silica gels.</td>
<td>66</td>
</tr>
<tr>
<td>Figure 4-5</td>
<td>Increased Ni(^{2+}) transport from low water ratio, low silica gels.</td>
<td>67</td>
</tr>
<tr>
<td>Figure 4-6</td>
<td>Solver and ANOVA estimates of D from analytical solution.</td>
<td>70</td>
</tr>
<tr>
<td>Figure 4-7</td>
<td>Estimates of D predicted by Solver for gels containing ORMOSIL or PEG200.</td>
<td>73</td>
</tr>
<tr>
<td>Figure 4-8</td>
<td>Linearized experimental data with analytical and exponential models for gel #28</td>
<td>75</td>
</tr>
</tbody>
</table>
Figure 4-9: Solver and ANOVA estimates of D from exponential approximation...... 75
Figure 4-10: Solver estimates of D, ORMOSIL and PEG-containing gels from both analytical and exponential expressions.................................................................78
Figure 4-11: Solver and ANOVA estimates of D using diffusion exponent n = 0.45. . 79
Figure 4-12: ANOVA estimates of D using analytical solution and exponential expression with n = 0.50 and n = 0.45. ............................................................. 80
Figure 5-1: Comparison of D estimates for TEOS gels arranged by water ratio and acid content. (A): 4:1 water ratio gels; (B): 10:1 water ratio gels; and (C): 20:1 water ratio gels.................................................. 84
Figure 5-2: Schematic of ideal 1-D diffusion geometry............................................. 88
Figure 5-3: Schematic of actual diffusion geometry, as used in this investigation. .....89
Figure 5-4: Estimated diffusion coefficients of TEOS gels containing 10 wt% and 25 wt% PEG200................................................................. 91
Figure 5-5: Contact angles for water on a surface coated in TEOS gel (A), and a surface coated in a gel containing 10 mol% MTES (B). ............................................. 93
Figure 5-6: Estimated diffusion coefficients of gels containing 10 mol% various ORMOSILs and 10wt% PEG200. ................................................................. 94
### LIST OF TABLES

<table>
<thead>
<tr>
<th>Table</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>Table 3-1: Primary tetraethoxysilane sample set.</td>
<td>51</td>
</tr>
<tr>
<td>Table 3-2: ORMOSIL and PEG200 secondary sample set.</td>
<td>52</td>
</tr>
<tr>
<td>Table 4-1: Analytical solution estimated values of the diffusion coefficient for TEOS gel samples, calculated by Solver and ANOVA.</td>
<td>69</td>
</tr>
<tr>
<td>Table 4-2: TEOS gel sample composition and ID numbers.</td>
<td>70</td>
</tr>
<tr>
<td>Table 4-3: Calculated values of D from analytical solution using Solver for ORMOSIL and PEG-containing gels.</td>
<td>72</td>
</tr>
<tr>
<td>Table 4-4: Sample composition and ID for gels containing ORMOSIL and PEG200.</td>
<td>73</td>
</tr>
<tr>
<td>Table 4-5: Exponential solution estimated values of the diffusion coefficient for TEOS gel samples, calculated by Solver and ANOVA.</td>
<td>76</td>
</tr>
<tr>
<td>Table 4-6: Estimates of D for ORMOSIL and PEG-containing gels, by Solver with exponential approximation.</td>
<td>77</td>
</tr>
</tbody>
</table>
Chapter 1 – Introduction and Project Objectives

1.1 Introduction

Sol-gel processing is a broad and extensively studied field in the area of materials science. The term ‘sol-gel’ generally refers to liquid chemistry routes of fabricating metal oxide gels at room temperature, as opposed to extraction from melts or deposition techniques. The process requires less energy since it occurs at room temperature and ambient conditions, and also enables the production of some materials not possible through conventional melt procedures due to crystallization, phase separation, and other phenomena. Once the ‘gel’ is formed from the ‘sol,’ heat treatments, such as pyrolysis and/or annealing, can then be used to fully condense the material into a stable amorphous glass or ceramic with microcrystalline structure, depending on the nature of the heat treatment. Sol-gel derived materials can be used in optical and electrical devices, coatings, functional matrices for adsorption, separation membranes, and mixed phase composite materials of significance in sensing, catalytic processes, and biomedical applications.

Most transition and semi-metals through the middle of the periodic table are amenable to sol-gel fabrication routes, the most commonly utilized including titanium, aluminum, copper, zinc, boron, and, perhaps most importantly, silicon, among many others. The common aspect to all sol-gel processing techniques is to first produce a ‘sol,’ or solution of hydrolyzed molecules containing active metal centers in the form of monomers or small oligomers of varying sizes and geometries. The metal atoms are coordinated by covalently bonded hydroxyl groups, with negative charge shifted toward the oxygen atoms. The number of hydroxyl groups on any given metal atom depends on the valence and orbital structure of the metal. Silicon, the subject of this investigation, forms a well described sp$^3$ hybridized orbital structure, resulting in four hydroxyl groups oriented in a tetrahedron when fully hydrolyzed to silicic acid, Si(OH)$_4$.

Depending on the material, conditions, and chemistry, the sol may or may not be stable, and is therefore primed for ‘gelation,’ resulting from the condensation of
hydroxyl groups, forming metal-oxygen bridges and ultimately a solid structure. The particles in a sol prior to gelation are at the small end of the colloidal range or even smaller, on the order of nanometers to Å, hence the ‘sol’ is on the cusp between a homogenous solution and colloidal suspension. The gel is formed once the metal oxide oligomers bridge the geometry of the solution’s entire volume. At that moment, the solid and liquid phases are both continuous across the geometry, each phase interwoven with the other. Heat treatments can remove the liquid phase and fully condense the gel into a glass that is structurally indistinguishable from glasses extruded from a melt. Again, depending on the material, if the temperature is high enough and/or the exposure time long enough, the glass may crystallize, developing from amorphous glass to a ceramic.

Sol-gel chemistry is well studied in the context of creating condensed functional materials, including optical lenses, sensing devices, coatings, electrical components, etc. In all these cases, the material is condensed through heat treatment, turning the gel, which contains a significant portion of solvent and possibly other components, into a dense stable solid, containing only the metal oxide. The functionality of these materials is highly sensitive to heat treatments and the final microstructure of the material. Typically, important properties are defined by whether the material is amorphous or crystalline, having gone through a crystallization phase transition. If it is crystalline, the grain size, grain boundaries, and pore structure, if present, are also extremely important.

What is less well understood, largely because there has been no relevant application until very recently, is how processing parameters govern the structure and resulting transport within the wet gel prior to being subjected to any drying, extensive condensation, or heat treatments. As mentioned, the gel is technically a two-phase material with a continuous solid phase containing a continuous liquid phase. For such a material to exist, the solid must be extremely porous, akin to a sponge, which is obviously quite different than a condensed glass or ceramic. The structure of that metal oxide ‘sponge’ is dependent on the processing parameters under which it was created, including the nature of the sol prior to gelation and how the sol was catalyzed toward gelation. Whether the micro structure more resembles a loosely
bound system of colloidal particles or a highly cross-linked network of linear polymers is entirely dependent on the synthesis route. Controlling that structure has been explored only recently as new applications for the gel, which take advantage of that ‘empty’ space within the structure to encapsulate other components, have been devised, leaving much to be learned in this regime of sol-gel processing. The figure below, a ternary phase diagram, illustrates the novelty of this investigation compared to many other investigations in the literature, each discussed at length in Chapter 2, §2.5.5.

Figure 1-1: Ternary phase diagram of silica sol-gel processing space.

Silica sol-gel, by far the most extensively studied sol-gel system, has received much recent attention as an encapsulation matrix for functional biological components, such as pigments, proteins, enzymes, and viable cells. This application opens opportunities for the development of biosensors, drug delivery systems, tissue therapy applications, and bioreactors for the production of value-added secondary metabolites. This was first demonstrated in 1989 by Carturan et al., who
encapsulated *Saccharomyces cerevisiae*, common yeast, and has been explored extensively since [1]. However, this is a challenging process with extensive empirical development but still no concrete theoretical description. The processing space for silica sol-gel chemistry is enormous, encompassing a broad range of silica content, precursor materials available, acid and base chemistry for catalysis, and the use of additives to alter the gelation process and ultimately influence the final structure and properties of the gel. Due to the broad range over which parameters can be manipulated, the differing chemistries that occur through that experimental space, and the vast number of reactions taking place, there is no complete theoretical description for silica sol-gel chemistry or the encapsulation process. As a result, in the field of biological encapsulation, each new system must undergo some degree of iterative experimentation to refine and improve the system for the target application. Design of an optimal encapsulation matrix *a priori* is not currently possible.

Despite these challenges, using silica sol-gel as an encapsulation matrix offers many advantages. Silica is inert and biologically compatible. The matrix is virtually invulnerable to biological processes that may digest, breakdown, or otherwise compromise other encapsulation matrices such as organic polymers. Also, the precursors are relatively cheap and available since silicon is one of the most abundant elements on earth. Most importantly, the matrix is highly porous, or at least it can be if designed properly, and relatively stable over time scales comparable to the stability of viable cells.

The high porosity enables a high diffusion rate of dissolved gases and dissolved materials through the matrix, an essential characteristic for encapsulating functional biological components. This diffusion is necessary to support high enzymatic turnover, rapid response in a biosensor, and the exchange of fresh nutrients and metabolic waste products, essential to supporting viable cells. Ultimately, the higher the diffusion rate, the better the matrix will support functional biological components, as diffusion approaches that in an unconfined aqueous solution.
However, since the matrix must have a certain minimum amount of silica to form a solid gel, there will be an upper limit to diffusion that will be well below that in an unconfined solution. If the gel structure is lost, then all the benefits of the gel are also lost, including the ability to stabilize and protect biological components over time, contain engineered organisms, and prevent contaminants from penetrating the gel. The target gel synthesis space is that area where the bare minimum silica is used to form a rigid gel, and it is processed in a way to maximize biocompatibility with the target encapsulant and maximize diffusion within the constraints imposed at that silica content. The objectives of this project, stated below, are designed to explore this processing space, and understand the influence of processing parameters on diffusion within it.

1.2 Objectives

There are two primary objectives to this project:

1. Estimate the diffusion coefficient, $D$, for gel samples within a parameter space relevant to encapsulation of biological components. This includes very low silicon content, excess water, low acid and base content used for catalysis, minimal residual or co-solvent alcohol in the sol, and maintaining a fully hydrated gel with aging.

2. Establish a correlation between processing parameters and $D$ in order to understand how those parameters influence $D$, better understand the diffusion processes taking place in this system, and improve a priori design of silica gel encapsulation matrices for optimal diffusion.

Objective 1 is intended simply to understand how diffusion behavior changes over a broad range of processing space. Objective 2 is aimed at understanding the causes of variation in diffusion, or at least understanding the influence of processing parameters on diffusion. Modeling of diffusion will be accomplished through two parallel approaches. First, an analytical solution to Fick’s Second Law, taking the form of a trigonometric series, will be applied to estimate the effective diffusion coefficient, $D$. Next, an empirical approximation to diffusion, in the form of an exponential
expression, will also be used to estimate $D$ and the diffusion exponent (indicative of the type of diffusion occurring and other effects related to experimental geometry). Subsequent analysis and comparison of the two modeling approaches are intended to identify differences, if any, between the two approaches and correlate processing parameters with changes in diffusion behavior.

An improved understanding of the relationship between processing parameters and diffusion in a wet gel will enable improved design and synthesis of encapsulation matrices for biological components. The results of this work may only be relevant to silica sol-gel systems within this processing space, but the range is broad and silica is considered the most promising material for sol-gel encapsulation, so there is obvious utility in examining this system.
Chapter 2 – Literature Review

2.1 Introduction

Sol-gel chemistry was first described in the 19th century and is perhaps one of the easiest and most versatile fabrication methods in all of materials science. The term “sol-gel” derives from abbreviating and combining the names of the two primary steps of the process. The first creates a solution of hydrolyzed metal, in the form of a true solution of monomers, or a colloidal suspension of partially condensed oligomers and particles. This is called the “sol” and it may or may not be stable. Next, either through natural equilibrium or a catalyzed process, the condensation reaction accelerates and the sol eventually undergoes gelation, or turns into a “gel” fully bridging the geometry of the solution. The variety of metal compounds amenable to sol-gel processing are quite broad, and the variety of structures achieved, depending on processing parameters during gelation and afterward, are equally broad. This approach has been used to fabricate materials for electrical devices, membranes, coatings, composite materials, and many other applications. In addition to the ease of processing, one significant advantage offered by sol-gel chemistry is the ability to produce monolithic glasses not otherwise possible to fabricate from a conventional melt due to effects of phase separation or crystallization [2].

The following review focuses exclusively on silicon and silica sol-gel as this is arguably the best and most common inorganic material for bio-encapsulation. The chemistry of silicon and the silica sol-gel system is also among the most extensively studied, including two thorough treatises by Iler [3] and Brinker [4], concise summaries [5-7], and countless peer reviewed articles focused on various aspects of the process. This review will briefly summarize each step of the silica sol-gel process prior to conventional heat treatment, provide a description of how changes in processing can influence structure, summarize the treatment of diffusion in silica sol-gel, and conclude with a brief discussion on encapsulating biological components. Although this investigation did not directly examine the encapsulation of biological components, the intent is to provide a better understanding of the processes that will facilitate improved transport to better support encapsulated biological components.
2.2 Chemistry of Silicon and Silica

Silicon is the second most abundant element in the earth’s crust, accounting for approximately 27% of the crust by mass. Silicon occurs commonly as quartz, crystalline SiO$_2$, the stable phase under approximately 870°C, and in a variety of oxide and silicate minerals, including potassium silicate, sodium silicate, and many others. In fact, the origin of many silicate minerals is super saturated suspensions in prehistoric seas, which formed gels and then condensed into mineral [3]. Other phases of silica, aside from amorphous glass and quartz, stable at ambient pressure over various temperatures include tridymite (870°C up to 1470°C), and cristobalite (stable to 1700°C, then vitreous silica forms). At elevated temperature and pressure, keatite, coesite, and stishovite may form [3]. Silicon also occurs in aqueous solution as orthosilicic acid, Si(OH)$_4$, which is ubiquitous in the world’s oceans at varying concentrations, averaging approximately 17 µM. Although the solubility of silicic acid is low, this is still below the predicted equilibrium concentration at 25°C of approximately 2 mM, largely because of the complex interactions with its environment, especially with certain metal species in solution, particularly aluminum and iron [3]. Silicon is also found in living organisms, playing an important role in the connective tissues and structural fibers of animals and plants alike. The variety of forms observed in the silica skeletons of diatoms is probably the most well known instance of biological utilization of silica.

Dissolution of silica in water is given by the hydrolysis reaction:

$$SiO_2 + 2H_2O \rightarrow Si(OH)_4$$

The above reaction is promoted by alkaline environments and elevated pH. However, under most conditions this reaction will produce very low concentrations of silicic acid as its solubility in water is quite low, as mentioned above. Silicic acid may also enter the aqueous environment through chemical erosion of minerals containing silicon. The concentration of silicon in surface fresh water is often significantly higher than in marine waters, but the concentration varies widely depending on local mineral and water chemistry [3]. From a dilute solution, it is not possible to isolate pure orthosilicic acid, it will polymerize and condense to silica long before it becomes anhydrous.
In most compounds, including silica, or silicon dioxide, silicon occurs overwhelmingly in the tetravalent form. In some minerals, like stishovite or thaumasite, silicon is coordinated by six bonds, but this is an extremely rare occurrence [3]. The tetrahedral coordination is due to sp\(^3\) hybridized orbitals. In ideal crystalline silica, or \(\alpha\)-quartz, the silicon – oxygen bonds assume a bond angle of 109.5° and Si – O – Si bonds are 144°. The Si – O bond length is 1.61Å and the distance between adjacent oxygen atoms can vary from 2.60 to 2.67Å. A silica tetrahedron is shown in Figure 2-1, below.

Figure 2-1: Silica tetrahedron.

In amorphous silica, which is the form of all silica gels, the basic tetrahedral structural subunit remains largely intact. However, long range order is lost as the arrangement between adjacent tetrahedra introduces some flexibility between second and third nearest neighbors and beyond. The angle between oxygen atoms on adjacent silicon atoms, fixed in \(\alpha\)-quartz, may vary as much as 25% and the bond lengths may vary as much as 10%, but despite these variations, the optical properties remain largely intact [8-10]. This may be of interest in encapsulation of light sensitive biological components, including phototrophs, since the silica gel must remain transparent to the visible spectrum.
In condensed amorphous silica, or common glass, these variations in bond angle and length, which distort any long range order, are the primary distinguishing feature from crystalline quartz. However, in an uncondensed silica gel, there remains a large number of hydroxyl groups on the gel surface within the micropores. These hydroxyl groups are readily available for hydrogen bonding with water or any other polar material that may be available in the liquid phase. Furthermore, although it is beyond the scope of this investigation, uncondensed hydroxyl groups are reactive, allowing for surface treatments to covalently attached moieties with varying functionalities.

2.3 Sol-gel Chemistry

The term “sol-gel” refers generally to the two steps of the process: preparation of the liquid “sol,” and gelation to form a two phase “gel,” a continuous solid containing a continuous liquid. Both steps can be carried out at ambient temperatures and pressures. A “sol” is technically defined as a colloidal suspension of a solid phase within a liquid phase. The term “silica sol” has a slightly broader definition, encompassing both the colloidal suspension and solutions of polysilicic acid, or oligomers of silica, that vary in size and degree of condensation, within or slightly smaller than the colloidal size regime (10’s to 100’s of Angstroms). Condensation reactions then occur, either due to sol instability, or through catalysis, which cause the colloidal particles and oligomers to condense and coalesce into a gel, spanning the whole solution geometry. It is this initial preparation of a sol followed by the formation of the gel that gives the overall process the generic “sol-gel” name.

A sol is usually prepared by hydrolyzing a metallic compound into an appropriate solvent with either an acid or base to catalyze hydrolysis. The metal is usually a transition metal, including elements like aluminum, copper, zinc, or titanium, or a semi-metal, like silicon, boron, or germanium. Fully hydrolyzed acids of these compounds, in the form of X(OH)$_n$, where X is the metal and n is the number of hydroxyl groups bound to the metal, usually corresponding to the valence of its stable cation, often have limited stability and solubility in an aqueous solution. Various alcohols are typically used as co-solvents, and other compounds may also be used
either as co-solvents or to improve sol stability through chelating active metal compounds.

In all sols, condensation reactions are in competition with hydrolysis reactions. Stability is achieved if the two reactions are in equilibrium and the stable particle size at that equilibrium is something less than or equal to colloidal size. Once condensation accelerates past the rate of hydrolysis, which can happen for many reasons, condensation may proceed unabated until the solid phase becomes fixed, spanning the geometry of the solution. Once this occurs, the sol is said to have “gelled,” having gone through a gel transition. However, once the gel forms, condensation and hydrolysis are by no means complete. There is a continuous liquid phase within the solid phase, and that liquid phase will still contain unreacted monomers or oligomers of the sol precursor (metallic acid). Again, depending on the chemistry, the gel will continue to age in differing ways depending on how condensation and hydrolysis proceed after gelation. Each of these steps is explained in more thorough detail in subsequent sections below, with the discussion being split between the two major systems: aqueous precursors and alkoxide precursors. There is extensive literature in both areas. However, this investigation used alkoxide precursors exclusively, as it is the preferred system for biological encapsulation of phototrophs, so the discussion of the aqueous route is extremely brief with a more thorough discussion of the alkoxide route following.

2.4 Aqueous Precursors

One very common method of silica sol-gel synthesis is to use a solution of sodium silicate, Na$_2$SiO$_3$, or soda glass (more accurately a solution of Na$_2$O and SiO$_2$ in equilibrium), in the preparation of a sol precursor. This is often referred to as an “aqueous precursor,” as opposed to alkoxide precursors, discussed below. The dissolution of sodium silicate with acid catalysis of hydrolysis is shown by the following reaction:

$$Na_2SiO_3 + H_2O + 2HCl \xrightarrow{\text{yields}} Si(OH)_4 + 2NaCl$$
This type of reaction may occur with any soluble silicate. However, this system, which usually results in a supersaturated solution of silicic acid monomers, is unstable and will undergo some degree of condensation into small silica particles [3]. The condensation of monomers into a particle containing \( n \) silicon atoms can be described by the following expression (Iler, page 5):

\[
Si_nO_{2n-(nx/2)}(OH)_{nx} + mSi(OH)_4 = Si_{n+m}O_{2n-(nx/2)+2m(2-p)}(OH)_{nx+4(m-p)} + 2pmH_2O
\]

Where \( n \) = number of silicon atoms present in the particle,

\( x \) = number of OH groups per silicon atom, not to exceed 4

\( m \) = silicic acid monomers

\( p \) = fraction of labile hydroxyl groups on silicic acid monomers that undergo condensation during the polymerization [3].

Aqueous precursors have been used extensively in encapsulation of biological materials, primarily because there is no alcohol present in this process. Many biological components, from proteins to whole cells, are extremely sensitive to alcohols, which may be denaturing or cytotoxic. However, while this process removes alcohol stress, it introduces a significant osmotic stress through high sodium ion concentrations. These sodium ions may be removed through the use of a strongly acidic ion exchange resin, replacing sodium ions with protons, but this then introduces the need for a strong neutralizing base. The salts that result may also reduce biocompatibility in certain systems.

The reaction kinetics in this system also favor condensation, promoting the formation of larger particles prior to gelation. Although the gel can remain clear as with the alkoxide system, it is easy for the gel to transition to opaque when the particle size reaches an order of 100’s of nanometers, large enough to scatter visible light. The aqueous system is also not as flexible as the alkoxide system in terms of altering gel structure through the use of varied precursors. The aqueous system can be combined with other polymers, like polyacrylic acid, for example, but the
application is often chromatography and separations [11], not bioencapsulation. The results of this investigation are intended to improve the encapsulation process for light sensitive biological components, so these two factors excluded any extensive use of aqueous precursors.

2.5 Alkoxide Precursors

Alkoxide precursors are a general name for any hydrolysable organometallic compound [5], including siloxanes, typically composed of a central silicon atom bound to four organic ligands via ester linkages. The most commonly used compounds of this category are tetramethoxysilane (TMOS), with a formula of Si(OCH₃)₄, and tetraethoxysilane (TEOS), with a formula of Si(OCH₂CH₃)₄, which are synthesized by the alcoholysis of silicon tetrachloride [4]. Siloxanes can be hydrolyzed by acid or base catalysis, in aqueous solution, with the generation of alcohol, to prepare a sol of silicic acid of varying degrees of condensation. The sol can then be catalyzed toward gelation through either base or acid catalysis. Acid catalyzed hydrolysis followed by base catalyzed condensation was the synthesis route used exclusively during this investigation, warranting additional attention in the discussion below. Each process is discussed separately, concluded by a summary of relevant investigations of the sol-gel process with alkoxide precursors.

2.5.1 Acid-Catalyzed Hydrolysis

Hydrolysis is a water-consuming reaction that displaces an alcohol ligand with a hydroxyl group, liberating alcohol into solution. Hydrolysis of silicon alkoxides is expected to occur by a nucleophilic attack of a water oxygen on the silicon atom. The oxygen is electronegative while the silicon is weakly electropositive, contributing to the relatively slow kinetics of hydrolysis in the silicon system as compared to other, more strongly electropositive metals [4]. In an acidic environment, the alkoxide group likely becomes protonated, which draws electron density away from the silicon atom, making it more susceptible to nucleophilic attack by a water molecule. Thus, hydrolysis will occur in the absence of catalyst, but an acid catalyst greatly accelerates the kinetics, also making hydrolysis more complete.
Once in solution, the isoelectric point for silicic acid occurs approximately at pH 2, where its solubility and stability are maximum. Solubility is expected to be a minimum at approximately pH 7, increasing slightly at acidic pH before reaching a maximum plateau at approximately pH 2, and quite significantly at alkaline pH, increasing through pH 10 [3]. This apparent lack of correlation between isoelectric point, deprotonization and solubility is not well understood [3].

Without an acid catalyst, the pH in a sol may be as high as 5 and hydrolysis is very slow as a result, so gelation will take on the order of many days (>1000 hours) [12]. Strong acids, including hydrochloric, sulfuric, and nitric (used in this study), all reduce the pH below the isoelectric point at low mole fractions. The system must be below the isoelectric point for acid catalysis of hydrolysis to be effective. It is in this region that proton concentration is high enough to influence the charge distribution of the alkoxide bond. It should also be noted that throughout the entire range of pH under consideration in this study, silicic acid is fully protonated during the preparation of the sol. The first $pK_a$ is about 9.8, meaning Si(OH)$_4$ does not lose its first proton until nearly pH 10, and the second deprotonation is expected to occur beyond pH 12. The species Si(OH)O$_3$ and SiO$_4$ are typically not observed. However, it must remain clear that this refers strictly to silicic acid. Subsequent condensation reactions will alter the electronic environment on each silicon atom, and thereby inductively alter the relative acidity of each remaining hydroxyl group (see Figure 2-4 and associated discussion, § 2.5.3).

Initial hydrolysis in a TEOS system is slower than in a TMOS system due to steric effects from the larger alkoxide group. The relative rate of reaction for hydrolysis increases as the monomer becomes increasingly hydrolyzed, the rate of the initial hydrolysis being $1/144$ the rate of the last hydrolysis [13]. In the acid system with TMOS or TEOS, hydrolysis can be nearly complete prior to gelation, whereas in the base-catalyzed system, as much as 20% of the alkoxy groups may remain at the onset of gelation [13]. What unhydrolyzed groups remain in the acid catalyzed system will tend to be distributed from completely unhydrolyzed monomers to fully hydrolyzed silicic acid, with the concentration of each increasing significantly with increased level of hydrolysis (i.e. very little completely unhydrolyzed monomer is expected). In
contrast, the distribution of the alkoxy groups in a base catalyzed sol tend to be bimodal. There will generally be condensed colloidal particles of silica with hydroxyl groups on the surface and completely unhydrolyzed tetra alkoxy in solution, with little in between. This is not preferable in the context of biological encapsulation since later hydrolysis after gelation would produce potentially cytotoxic alcohol with no available means to prevent this prior to encapsulation. The exact mechanisms at play in this chemistry and how they determine the composition of the sol as it proceeds toward and through gelation is still an area of active inquiry.

The hydrolysis reaction is shown below, with the corresponding reverse reaction, a replacement of the hydroxyl group with alcohol in a reesterification reaction.

\[
\text{hydrolysis} \rightarrow \quad \equiv S\!i - O\!R + H_2O \quad \overset{\text{esterification}}{\leftrightarrow} \equiv S\!i - O\!H + R\!O\!H
\]

Stoichiometrically, the complete hydrolysis of a tetra alkoxy silane requires four moles of water per mole of siloxane, shown below. Condensation releases water, one molecule per oxygen bridge formed, so net, formation of anhydrous silica from alkoxy precursors has a stoichiometric requirement of two moles of water per mole silica. Temporally separating hydrolysis and condensation also separates water consumption and production, necessitating the intermediate excess of water in the sol solution to improve stability without additional co-solvents.

\[
S\!i(OR)_4 + 4H_2O \rightarrow S\!i(OH)_4 + 4ROH
\]

As mentioned, the hydrolysis mechanism is reported to be a bimolecular nucleophilic displacement, whereby an alkoxy is rapidly protonated, stabilizing reduced electron density on the silicon atom, enabling nucleophilic attack by a water molecule on the opposite side of the tetrahedron. The alcohol becomes a better leaving group, losing its partial positive charge to the water, so it is displaced and the tetrahedron inverts with a hydroxyl group in its place [14]. This mechanism is shown schematically in Figure 2-2, below.
Alkoxide compounds have virtually no solubility in water, so an alcohol co-solvent may be used to facilitate miscibility and stabilize the mixture. If the hydrolysis ratio, water to silicon is particularly low, approaching or even below the stoichiometric minimum of 4 to 1, then supplemental alcohol solvent may be necessary to promote as complete hydrolysis as possible and stabilize the sol. However, silicic acid does have limited solubility in water, and the hydrolysis process may produce enough alcohol to facilitate a completely homogeneous phase, especially as the molar ratio of water to silicon increases, diluting the concentration of silicic acid. Throughout this investigation, for example, all sols eventually achieved homogeneity through vigorous mixing without the use of any supplemental alcohol co-solvent.

2.5.1 Base Catalyzed Hydrolysis

A basic compound, such as ammonium hydroxide or potassium hydroxide can also be used to catalyze hydrolysis. The reaction mechanism is only slightly different than the acid catalyzed route, but the kinetics vary and the competition with condensation are different, leading to very different structures in the wet gel, despite no visible sign of synthesis history left in an annealed and condensed monolith. Base catalysis was not used in this investigation, therefore its description here is brief and the reasons for not employing this route in this system will become self evident.

Base catalyzed hydrolysis, often with ammonium hydroxide, occurs by a nucleophilic substitution of hydroxyl ions for the alkyl group, as follows [12], and shown in Figure 2-3 below:
As with the proton in acid catalysis, the hydroxyl ion alters the charge distribution of the alkoxide bonds enabling the reaction and is regenerated in the reaction by the deprotonated alcohol group which promptly reacts with water. Steric and inductive effects will influence kinetics. However, the shift in charge distribution around the silicon atom is expected to be minimal, suggesting steric effects have a more pronounced influence on reaction kinetics [4].

The kinetics are expected to be first order with regard to OH$^-$ concentration, and are expected to accelerate with increased substitution of hydroxyl groups on the silicon atom [4]. This is because the substitution of hydroxyl for alkoxide is enabled by more electron-withdrawing side groups, such as –OH or a condensed –O-Si bridge. Due to steric effects and the difficulty in forming the reaction intermediate, the first hydrolysis is expected to be slowest, and then accelerates with increased substitution as both steric and inductive hindrances are reduced. The result is a bimodal distribution of species in a base catalyzed sol, including completely unhydrolyzed alkoxides and highly condensed particles, with low concentrations of intermediate species. The first hydrolysis is slow enough that upon gelation, completely unhydrolyzed monomers of the alkoxide precursor may still be present [15]. The resulting gel is usually a more coarse structure, with larger particle size and larger pore size with reduced surface area and cross linking. The base catalyzed system was not selected for this investigation primarily because the coarse structure of the resulting gel and the possibility of unreacted monomers being present. Although this approach can be used for bioencapsulation, the acid catalyzed approach is preferable for many applications.
2.5.2 Acid-Catalyzed Condensation

Condensation is the reaction to form an oxygen bridge between two silicon atoms with the concomitant release of water. As shown in the following reactions, this can occur by condensation of two hydroxyl groups, or by hydroxyl and alkyl groups:

\[
\begin{align*}
\text{water condensation} & \rightarrow \quad \text{hydrolysis} \\
\equiv Si - OH + HO - Si & \equiv Si - O - Si \equiv +H_2O \\
\text{alcohol condensation} & \rightarrow \quad \text{alcoholysis} \\
\equiv Si - OR + HO - Si & \equiv Si - O - Si \equiv +ROH
\end{align*}
\]

In systems with excess alcohol solvent and limiting water, the alcohol condensation reaction may be significant. However, in systems with excess water, limiting alcohol, and low silicon concentrations, such as all gels prepared for this investigation, alcohol condensation is considered quite negligible. Under the prevailing conditions of this investigation, hydrolysis is expected to be near completion prior to catalyzing condensation in a second step.

At acidic pH, all silanol groups are expected to be protonated. Protonated silanol groups affect withdrawal of electron density from the central silicon, making it more electrophilic and therefore vulnerable to nucleophilic attack. Additional condensation with other side groups will further increase the acidity of the remaining silanol groups (see Figure 2-4 below). In this environment, condensation will preferentially occur with the most basic silanol groups (presuming the concentration of remaining unhydrolyzed alkoxide groups is negligible), which tend to be fully hydrolyzed monomers or end groups of linear oligomers with minimal branching. The result is a network of linear polymers that condenses more slowly as condensation proceeds.

2.5.3 Base-Catalyzed Condensation

Base catalyzed condensation is probably the most common catalysis route used in silica sol-gel processing. The most widely accepted reaction mechanism is the nucleophilic attack of a deprotonated silanol group upon a neutral (i.e. protonated) silanol. Recall that below pH 9.8, silicic acid is expected to be mostly protonated. At a
pH in the 7 to 8 range, based on a pKₐ of 9.8, there should be a mix of approximately 90% protonated species with 10% deprotonated species. However, the acidity of silanol groups depends on the chemical environment of the silicon atom, including its coordination with other groups. Side groups that are more electron withdrawing increase the acidity of the silanol, making it more likely to be deprotonated and therefore primed for nucleophilic attack. As a result, in the pH range of 7 to 8, any degree of condensation increases the acidity of remaining silanol groups, meaning more are likely to be deprotonated (see Figure 2-4 below). Comparable concentrations of both protonated and deprotonated silanol groups strongly accelerates the condensation reaction.

![Figure 2-4: Electron donating characteristics of various silicon side groups.](image)

Siloxane bonds are the most electron withdrawing, followed by hydroxyl, then alkoxide, and finally alkyl side groups, as shown in figure 2-4, above (used with permission from [4]). The result is that as condensation proceeds in the base catalyzed system, it actually accelerates to quickly proceed toward full condensation, consuming all available monomers. The rapid condensation usually results in a highly cross-linked network with fine structure and high porosity. The amount of base required to achieve catalysis is minimal, the pH only need be neutralized, not actually made alkaline. Combined with acid hydrolysis, this is an approach well suited to bioencapsulation because the first step hydrolyzes nearly all of the alkoxide groups to alcohol, enabling alcohol removal as necessary. The second step leads to rapid
gelation, is at neutral or slightly basic pH, and quickly consumes reactive silicate species, limiting cytotoxic effects.

2.5.4 Gelation

Gelation occurs when condensed polymers bridge the geometry of the sol and no longer move freely in suspension. There is a distinct and dramatic increase in viscosity at the gel point, although a quantitative description of that change in rheological behavior depends on the composition of the gel. At this point, the matrix has formed a continuous porous structure, yet also contains a continuous liquid phase within that pore structure. Prior to gelation, condensation is governed by diffusion limited encounters of adjacent monomers and oligomers, in addition to electrostatic interactions, meaning that sols with higher silica content will have larger oligomers, consistent with a higher encounter rate [16]. Once the gel has formed, any subsequent condensation is diffusion limited as the bulk of silicon atoms are fixed within the gel structure, and surface groups available for condensation only react upon diffusion limited encounters with unreacted species in the liquid phase. After gelation, there are also aging processes occurring, discussed separately, where monomers may dissolve from the matrix and re-condense elsewhere, depending on the local chemical and physical environment.

The structure of the gel is dependent on hydrolysis history and the competing kinetics of hydrolysis and condensation. Although gelation has occurred, it certainly does not mean that hydrolysis has ceased, only that the condensation rate is significantly faster under those conditions. As discussed, acid catalyzed hydrolysis followed by base catalyzed condensation leads to a highly cross-linked gel with high porosity and small pore size. Through base catalysis, condensation is quite rapid, as is hydrolysis under acidic conditions. These two sequential steps effectively separate two regimes where each reaction is significantly faster than the other, hydrolysis followed by condensation. Increased acid content has been shown to increase the hydrolysis rate and lead to gels with high porosity upon gelation, confirmed experimentally through small-angle x-ray scattering (SAXS) [17]. Processed correctly, it is even possible to produce gels with pore volume inaccessible to nitrogen via
porosimetry, suggesting a closely packed structure of linear polymers that are highly condensed [18].

Temporal separation of hydrolysis and condensation is necessary to create a fine structure. If hydrolysis and condensation have similar rates, or condensation strongly accelerates as the acidity of silanol groups increases, the sol prior to gelation is composed of a mix of highly condensed oligomers and predominantly monomers or small polymers in solution. This mix of highly condensed particulate oligomers and poorly hydrolyzed monomers yields a structure that is coarse and particulate.

In reality, the above two scenarios represent ideal situations at the opposite ends of the processing spectrum. Even with an acid-base two step process, the gel will tend to have some particulate structure with particles that are small, on the order of a few to 10’s of nanometers. The kinetics can be manipulated to alter the size of these particles to some degree. With increased particle size comes increased pore size, but slightly decreased porosity and substantially decreased surface area.

2.5.5  Summary of Relevant Silica Sol-Gel Investigations

The three primary components in the silica sol-gel system, silicon alkoxide, water, and alcohol, are the primary determinants of final gel structure and the fundamental processing parameters exercising the most control over the system. Figure 2-5, a ternary phase diagram, illustrates the common processing space of silica sol-gel in terms of those components: silicon, water, and alcohol. As can be seen from the figure, supplemental alcohol solvent is more common than not, facilitating improved solubility of phases. Since alcohol is cytotoxic, which is of the utmost concern for encapsulation of live cells, this study focuses on the area of minimal alcohol content, only that which is produced by the hydrolysis of the alkoxide precursor, as shown in the figure in the lower left corner.
Figure 2-5: Ternary phase diagram (A).

Figure 2-5 also illustrates regions of processing space typically utilized in silica sol-gel processing (adapted from Brinker & Scherer [4]). It should be noted that while the current investigation lies approximately within the phase space identified as relevant to biological encapsulation, and this space is also immiscible at 25°C, sols can become homogeneous. As discussed below, sols were allowed to mix at 60°C, which promoted both hydrolysis and the evaporation of some alcohol. The diagram is interpreted to reflect solubility limits of purely uncondensed silicic acid, but some degree of condensation improves solubility outside the limits indicated in the figure.
Figure 2-6: Ternary phase diagram (B).

Figure 2-6 shows a similar phase diagram, adapted with permission from Brinker et al. [19], illustrating the relative positions of numerous investigations, including this one, in the ternary phase space of silica sol-gel preparation from alkoxide precursors. As can be seen from the figure, most investigations focus on regions of stoichiometric to slight excess water, with supplemental alcohol solvent. Also suggested by the figure is that the investigation of silica systems using a large excess of water with no supplemental alcohol co-solvent is a relatively new area of investigation. The relevant findings of each study identified in Figure 2-6 are discussed below in ascending chronological order.

Aelion provided an early investigation of hydrolysis kinetics in 1950 [20]. It was concluded that increasing excess of water does promote more complete hydrolysis in an acid catalyzed system. Elevated temperature also improves hydrolysis rates, but only to a certain extent, believed to reach at a peak at approximately 45°C [20]. This set of experiments used excess alcohol in the examination of kinetics, and as a
result, hydrolysis never approached completion. In fact, in the most acidic system, with the most complete hydrolysis, only about 2.3 moles of water were consumed per mole of silicon alkoxide, much less than the 4 moles expected to be consumed for complete hydrolysis. The kinetics observed under these conditions, aided by increased acid concentration and elevated temperature, are relevant to this investigation. Increased water will improve the extent of hydrolysis and the lack of excess alcohol should minimize the reesterification reaction. However, the lack of alcohol co-solvent means the initial preparation is immiscible and the homogeneous sol, once produced, will be less stability.

Bechtold et al. investigated TEOS hydrolysis and polymerization and verified the formation of small linear polymers under acid catalyzed conditions [21]. This supported the hypothesis of nucleophilic water attack on an electrophilic silicon, as well as increased inductive effects and decreased steric restrictions on subsequent hydrolysis.

The investigation by Brinker et al. was among the most comprehensive examinations to date of the hydrolysis and condensation reactions in a silicon alkoxide system [19]. Many observations from this investigation supported prevailing mechanisms for hydrolysis and condensation. The relevant observation for the current investigation was the confirmation of completely hydrolyzed silicate species from acid hydrolysis leading to highly cross-linked gels with high porosity. By comparison, base catalyzed systems were observed to reach less complete hydrolysis, shown by the presence of unhydrolyzed alkoxides remaining in the sol at the time of gelation, leading to less condensed gel. A subsequent investigation by Brinker et al. confirmed these observations and went on to further observe that polymers in the acid catalyzed system, prior to gelation, were more linear and elongated, with weak branching. Base catalyzed gels, by contrast, were more clustered or particulate, although not large enough to reach the colloidal range and scatter visible light [22].

Pouxviel et al. illustrated through $^{29}$Si-NMR that under excess water conditions, acid catalysis of TEOS hydrolysis can produce mostly complete
hydrolysis. Although some condensation occurs to form cyclic or short linear oligomers, very few unhydrolyzed alkoxide groups remained in the sol prior to gelation [23]. Higher water content also helps temporally separated hydrolysis and condensation reactions, as observed in a system with a water to TEOS molar ratio of 10:1 [24]. This investigation also found that in a system where \( r = 10 \), molar ratio of water to silicon, there is no unhydrolyzed monomer remaining after approximately 1 hour of reaction [23]. Furthermore, the dominant silanol species in this excess-water system were fully hydrolyzed and triply hydrolyzed [23]. This is not consistent with the expected kinetics resulting from inductive changes to the hydrolysis reaction as it proceeds toward completion. Recall that replacing an alkoxy group with an alky group increases the electron density on the silicon atom, reducing acidity, and likewise, hydrolysis and condensation both decrease electron density on the silicon, resulting in an inductive increase of acidity [4]. In acidic conditions, the rate of hydrolysis is predicted to decrease with each subsequent hydrolysis reaction, whereas in basic conditions, the rate of hydrolysis is predicted to increase with each subsequent hydrolysis step. These experimental data suggest that despite presumed inductive restraints on hydrolysis, the reaction will proceed very near to completion in an acid catalyzed sol with excess water.

Kelts & Armstrong also used \(^{29}\text{Si-NMR}\) to examine the relative completion of hydrolysis in TEOS and TMOS systems with limited up to stoichiometric water. They echoed the findings of Pouxviel et al. and found that at stoichiometric water, hydrolysis exceeded 85% relative completion, and the limited condensation that took place prior to gelation resulted in the formation of many cyclic or short linear structures, consistent with other investigations [25].

The investigation of Elferink et al. began to help define the parameter scope of this study. The authors showed that increased water and acid for catalysis both increase the relative completeness of hydrolysis. The authors were investigating the influence of processing over pore structure with the intent of controlling pore size distribution, relevant for membrane applications. The acid ratio varied from 0.00085 to 0.34 and the water ratio varied from 1.6 to 12.8. It was found that the combination of lower acid content, 0.021 in this case, and higher water resulted in mesoporous gels.
It was also determined that acid content and water content both have an effect on final gel structure and porosity, although the effect of acid content is presumed to be much stronger [18]. These gel samples were heat treated into microporous membranes on a mesoporous alumina support membrane, designed for gas separation, so although the findings are relevant, they do not translate directly to a system examining hydrated wet gels.

The investigation by Nair et al. examined an acid catalyzed system as it is the preferred route to achieve a microporous structure. The main focus was SAXS examination of pore structure, which reiterated that accelerated reaction rates are the predicted path to increased porosity [17]. This is presumed to include both hydrolysis and condensation, assuming the two can be temporally separated in a two-step process, and each one catalyzed sequentially.

Peeters et al. observed through $^{17}$O-NMR of hydrolysis of TEOS and TMOS that hydrolysis did not go to completion, particularly in conditions of limiting water, which is not a surprising result and is not inconsistent with previously mentioned investigations. With limited water, all water was not consumed, and an equilibrium concentration between hydrolyzed and unhydrolyzed species was established. Most interesting was the verification that reactivity toward hydrolysis also depended on the alkyl group. A hydrolyzed TMOS system was more reactive toward hydrolysis than a comparable TEOS system, but less reactive toward condensation and gelation, presumably due more to steric effects rather than inductive effects [26]. Despite these kinetics, TEOS was still observed to hydrolyze, just not as quickly as TMOS.

2.5.6 Kinetics and Structural Modeling

Manipulation of sol-gel processing for a desired gel structure ultimately depends on managing the competing hydrolysis and condensation kinetics. However, due to the number of reactions involved, it is impossible to predict how these two conceptually very simple reactions will actually proceed. The tendency for any given side group on a silicon atom to react is controlled by the electron density at the central silicon, which is in turn dictated by the other three side groups. The chemistry of these side groups are also influenced by the electronic environment of their
nearest neighbor. The result is that at the next nearest neighbor level, there are 1,365 distinct local silicon environments, the quantitative description of which would require 199,290 rate coefficients [27]. The figure below illustrates this concept in matrix form (used with permission from [4]).

Figure 2-7: Matrix representation of next nearest neighbor speciation [4].

The consequence of these effects is that a truly quantitative description of any significant number of silicon monomers, even a small fraction of a mole, requires solutions to an astronomical number of rate expressions. This is highly computationally intensive, and the utility is questionable. Researchers have taken a number of alternative approaches to modeling the gelation process and the growth of the silica gel matrix. Among the simplest models is the Bethe lattice, constructed of numerous branches representing an expanding polymer network. The structure resembles a tree diagram, and was first used to describe amorphous silica networks by Laughlin et al. [8]. This approach yields reasonable results to describe the electrical and optical properties of dense amorphous silica, but gives very little realistic information on the structure of the network.
More recently, increasingly sophisticated and computationally intensive approaches, such as continuous random network (CRN)\cite{9, 28}, cluster-cluster aggregation \cite{29} and molecular dynamics (MD) simulations \cite{30, 31} have attempted to model silica network growth at the scale of single monomers polymerizing. The CRN approach is highly empirical and provides no kinetic information, so has largely been superseded by the MD approach. Results are providing some limited ability to predict gel microstructure at the Angstrom scale, a priori. Condensation and polymerization predictions are based on probabilistic estimates of reactions, determined by parameters such ionic potentials, encounter rates, interaction potential, thermal energy, etc \cite{32}. Results have been promising and improving \cite{31, 33, 34}, although model sizes remain very constrained. For example, a system of $N$ atoms will require solutions to $N$ second-order differential equations with non-linear dependence on each other \cite{35}. This often limits modeling runs to sample sizes vastly smaller than 1 mole of atoms, in geometries at the Angstrom to nanometer scale. While there is certainly value in attempting to model the microstructure of a silica gel at the atomic level, extrapolating the results to a meaningful description of diffusion or success as an bioencapsulation matrix is challenging. A full discussion of these modeling approaches is beyond the scope of the current investigation.

The final microstructure of a silica gel depends on each and every step of preparation, including hydrolysis of the sol, condensation to form the gel, aging, and any post-gelation treatments. As mentioned, acid catalyzed hydrolysis, below the isoelectric point, tends to favor more complete hydrolysis. Followed by acid condensation, where silica remains largely insoluble, one would expect a highly condensed polymeric gel with fine structure (shown schematically in Figure 2-8(A), below). Base catalyzed hydrolysis tends to lead to more granular structures in the gel (illustrated schematically in Figure 2-8(B), below). Also, the amount of water present plays a large role in the gelation process. Water is an active reactant for the hydrolysis reaction, so the availability of water can govern the hydrolysis rate and degree of completion. Even more important may be the role of water as a solvent, stabilizing the sol. Solubility of silica is limited, so under water limiting conditions, larger, more condensed particles tend to form. More water favors a higher degree of
hydrolysis and improved solubility, reducing the size of silica monomers present in the sol. Ultimately, the catalysts used and the order or combination in which they are used as well as the water content in the system can largely dictate the structure of the gel upon gelation.

Meixner et al. found that xerogels prepared with a high water:TEOS ratio, at a pH of 2-3, had a mean micropore size of approximately 10 Å. Furthermore, it was found that microstructure was only weakly dependent on hydrolysis water ratio when it exceeded 10:1, water to silicon. Synthesized at pH 3 and a water hydrolysis ratio of 83, the structure was predicted to resemble close packed spheres [36]. This group generally focused on a water ratio greater than 10 and acidic pH, which avoided Ostwald ripening, expected to be a stronger effect at higher pH where silica solubility is increased dramatically.

Relative humidity and temperature during gelation may also have an effect on final gel structure, but this is predicted to be a result of influences on the evaporation rate of solvent and not due to any direct alteration to the gelation process [37]. Rapid evaporation, encouraged by higher temperatures and lower relative humidity, promotes more rapid gelation and increased pore contraction, resulting in a more closely packed structure compared to gels formed under higher relative humidity and lower temperatures.

Figure 2-8: (A) Polymeric network, and (B), a particulate network.
Fast hydrolysis paired with slow condensation produces a more coarsely grained structure, like (B) in Figure 2-8. Rapid condensation with comparatively slower hydrolysis, as occurs in an acid-base two step process, should resemble the more polymeric network [19], as shown in (A) of Figure 2-8. However, both cases represent idealizations, so in reality, gel structures will often have some degree of granular structure, what changes is the mean size of those grains and how completely condensed they may be. In the current system, the acidic hydrolysis followed by neutral to slightly alkaline condensation would be expected to promote a gel with grains on the order of 10’s of nanometers and some degree of necking due to Ostwald ripening and coarsening.

2.6 Gel Aging

2.6.1 Ostwald Ripening & Coarsening

After gelation, the hydrolysis and condensation reactions continue until a final equilibrium is achieved. As a result, the structure of the gel continues to evolve until it is stable. What determines a stable gel depends on the processing conditions and numerous factors may influence equilibrium. Like all reactions, hydrolysis and condensation will continue until there is no driving force to perpetuate on-going reactions. In the case of condensation, this may occur when all free monomers or oligomers in solution have bound to the surface of the gel, drastically slowing additional condensation due to diffusion limitations. Hydrolysis will cease when the surface energy of the gel is minimized.

Solubility of silica is dependent on particle size, with solubility generally increasing with decreased particle size. It is also related to the curvature on the surface of the particle, or the surface of a solid or gel. A small particle has a small positive radius of curvature, which promotes higher solubility compared to the surfaces of larger particles which have a more broad radius of curvature. Areas with tight negative curvature, such as a crevice in a surface or seam between two particles in contact, are actually less soluble. The results are twofold: First, in solutions of mixed particle size, smaller particles tend to dissolve and deposit upon larger particles, making the particle size distribution more narrow and the total number of
particles smaller. This process is known as Ostwald ripening, shown schematically in
Figure 2-9 below. Above pH 7, this is expected to occur for particles with a diameter
smaller than approximately 5 to 10 nm in a silica system. However, under more acidic
conditions where silica is less soluble, particles are expected to remain stable to
approximately 2 nm in diameter [4], below which dissolution and ripening is expected
to occur.

![Figure 2-9: Ostwald ripening.](image)

The solubility of these small particles can be described by the Ostwald-
Freundlich equation:

\[ S = S_o \exp \left( \frac{2 \gamma_{SL} V_m}{R_g T r} \right) \]

which gives the solubility of a particle with radius \( r \), where \( S_o \) is the solubility of a flat
plate, \( \gamma_{SL} \) is the solid-liquid interfacial energy between the silica gel and mother liquor
phases, \( V_m \) is the molar volume of silica (the solid phase), \( R_g \) is the ideal gas
constant, and \( T \) is the absolute temperature [4]. Qualitatively, it can be seen from this
equation that as \( r \) decreases, solubility increases asymptotically toward infinite
solubility as \( r \) approaches zero. Practically, the radius of a particle of course cannot
reach zero. However, a critical radius, \( R_c \), above which the particle is considered
stable and below which it will likely dissolve, can be described by a formulation of the
Gibbs-Thomson equation:

\[ R_c = \frac{2 \gamma \Omega}{k_B T} \]
where \( \gamma \) is the surface tension, \( \Omega \) is the atomic volume, \( k_B \) is the Boltzmann constant, and \( T \) is the absolute temperature. As mentioned above, for silica, \( R_c \) is on the order of approximately 5 nm in alkaline systems reducing to as little as 1 nm in acidic systems. This is based on an equilibrium between an increase in free energy due to adding surface area to the particle and a decrease in free energy due to adding volume of condensed silica to the particle.

Globally, the driving force for this process is a reduction in system free energy. There is an interfacial energy between the solid and liquid phases, and a free energy associated with the volume of the two phases. Intuitively, smaller particles have a higher surface to volume ratio, and would need to be more numerous in order to accommodate the same mass of silica. Furthermore, the smaller particles are less condensed, with as much as 50% of the silicon atoms located at the surface. These surfaces contain reactive hydroxyl groups as opposed to stable Si – O – Si bridges (an exothermic reaction) contained within the volume of the particle. These phenomena can be described in terms of chemical potential, \( \mu \), which is equivalent to the contribution of free energy per atom, \( \partial G/\partial n_i \), in the particle. The chemical potential is also related to the curvature of the surface, as this influences both the number of atoms per unit surface area and the accessibility of those atoms for hydrolysis reactions (i.e. relative chemical activity). The chemical potential at the surface can be described in terms of surface curvature by the Herring-Mullins equation:

\[
\mu = \Omega (\gamma \theta_x) \kappa_x + \gamma (\theta_y) \kappa_y
\]

where \( \Omega \) is atomic volume, \( \gamma \) is the surface tension, \( \theta \) is the contact angle between the particle and the liquid phase and \( \kappa \) is the mean curvature [38]. Assuming local equilibrium, this expression can be used to relate particle radius, \( R \), and chemical potential through a generalization of the Gibbs-Thomson equation [38]:

\[
\mu = \frac{2 \Omega \gamma}{R}
\]
Taken together, the above expressions illustrate that atoms on the surface of smaller particles will have larger chemical potentials compared to the larger particles. These atoms will therefore have a tendency to hydrolyze, or dissolve, creating a local increase in monomer concentration. Meanwhile, condensation is occurring on the surface of larger particles, where chemical potential is lower, creating a local decrease in the concentration of silica monomers. As a result, the chemical potential gradient is driving solubility and subsequent hydrolysis and condensation reactions are creating a concentration gradient, both driving mass transport from smaller particles to larger particles.

The second effect, also driven by a net reduction in free energy, occurs when particles come in contact and suddenly form an area of tight negative curvature. Silica deposits at this crevice, forming a broader connection between particles (i.e. necking) [3]. The source of silica deposited at these crevices is presumed to be hydrolyzed from surfaces of positive curvature, which are comparably more soluble. This process, called coarsening, helps strengthen the gel as it ages, increasing the connective strength between particles. The effect on transport is difficult to predict. Necking can reduce pore connectivity, but the hydrolysis of silica from elsewhere can open up pore volume and broaden channels between pores.

Figure 2-10: Coarsening through necking.

2.6.2 Capillary Forces

Due to the extremely small pore size within the gel matrix and the surface energies between the liquid and solid phases, very high capillary forces may act to
collapse the gel, to force it to contract and expel “mother liquor,” as the residual liquid is sometimes called. Pore size exerts strong influence over capillary forces, particularly as pore size shrinks below 100 nm mean radius. At a pore radius of 10nm, capillary pressure can exceed 15 MPa, or ~2,200 psi. At a radius of 5 nm, pressure exceeds 25 MPa (3,600 psi) and increases geometrically as pore radius decreases [4]. Without adequate structural integrity, the gel will shrink under this pressure, being literally pulled in upon itself. While the gel remains hydrated, capillary forces are expected to be minimal although some on-going condensation can result in some gel contraction, often called syneresis. Once the gel starts to dry, capillary forces become dramatically more pronounced as the liquid phase recedes from the solid phase.

As mentioned above, coarsening is the result of dissolution of silica from smaller particles or areas of positive curvature followed by deposition in crevices of tight negative curvature. This does not result in shrinkage, but it does serve to reduce surface area and increase bulk modulus. As a result, after gelation, there is a competition between coarsening, which serves to strengthen and stiffen the gel, and syneresis, which shrinks the gel under internal capillary pressures [39]. Shrinkage will only continue so long as the bulk modulus cannot resist the capillary forces. This increases the density and cross linking of the gel, which also increase modulus, so at some point after gelation, an equilibrium will be established between bulk modulus and capillary forces, arresting shrinkage.

Shrinkage is a dominant phenomenon in gels that are dried, as the interaction between liquid-solid, vapor-solid, and liquid-vapor interfaces amplify capillary forces. This is an extensively studied area, but as the gels in this investigation are never dried, it is considered beyond scope. However, hydrated gels, such as those investigated here, can also shrink to varying degrees with aging. The amount of shrinkage often depends on solvent type because the presence of alcohol and the pH can influence on-going hydrolysis, re-esterification, and condensation reactions. In a purely water system, such as this, shrinkage is actually expected to be minimal, less than 5% [39]. Furthermore, in neutral or alkaline solutions, where silica has higher solubility, coarsening is expected to be a dominant phenomena, further strengthening
the gel and reducing shrinkage as a result. In this investigation, where the pH was steady at approximately 7 and the solvent was pure water, shrinkage was not apparent in most samples. Where it did occur, it was considered minimal (discussed below).

Aging is discussed at length in the texts of Iler [3] and Brinker [4], and also in a concise series of four articles by Davis et al. [40-43].

### 2.7 Organically Modified Silicates

Organically modified silicates, typically referred to as ORMOSIL's, come in the general form of $R_x(R'O)_{4-x}Si$. $R$ denotes an alkyl group covalently attached to the central silicon without a bridging oxygen, and $R'O$ denotes an alkoxy group, an identical or different alkyl group attached to the silicon by an oxygen bridge. From a purely reaction kinetics standpoint, increasing substitution (increasing $x$) increases the hydrolysis rate of the alkoxy groups in an acid catalyzed system due to the stabilization of the reaction intermediate by the electron-providing alkyl groups [4]. This exact same effect slows hydrolysis in a base catalyzed system. However, steric effects actually slow the hydrolysis process by obstructing access to adjacent alkoxide bonds. Single substituted ORMOSILs, such methyltrimethoxysilane (MTMOS) or methyltriethoxysilane (MTES), are used commonly. The single methyl group is not expected to sterically inhibit hydrolysis since it is actually smaller than the alkoxide side groups.

ORMOSIL's allow for altering both the physical and chemical properties of the gel. Alkyl groups occupy bridging sites, altering the structure of the gel, usually introducing some flexibility and reducing brittleness, and they also alter bridging through steric interference, depending on the size and chemical nature of the organic group. In dried gels, it has been suggested that reactivity of the precursors toward hydrolysis, regardless of initial conditions, will be 96 to 98% in flexible gels, but falls to 87% in brittle gels composed of pure TEOS, so ORMOSILs can facilitate the complete reaction of silicon species [44]. They also alter the surface chemistry, both through simple electrostatic interactions and via the introduction of labile moieties...
vulnerable to additional reactions to covalently attach functional groups or entire proteins to the gel surface [45]. This is particularly useful in applications as broad as separations, sensors, ion-exchange, and encapsulation, among others.

A blend of TEOS and ORMOSIL, like MTES, can provide control over the number of hydrophobic methyl groups introduced into the structure, altering both its surface chemistry and structural properties. One investigation found that gels containing increasing amounts of MTES were increasingly microporous, initially thought to be attributable to pore collapse due to reduced cross linking, but the same was true in wet gels [46]. The observation of this effect in wet gels suggests the steric and inductive effects of alkyl groups will reduce cross linking, making the gel susceptible to shrinkage as condensation proceeds even in the absence of strong capillary forces. The effect of MTES increasing the porosity of the gel reached a maximum at about 20% ORMOSIL. As MTES increased, porosity began to decrease slowly, then precipitously above 70% ORMOSIL, suggesting dramatic structural changes. The gels containing as little as 10% MTES also showed a dramatic drop in the mass of water adsorbed on the surface as well as differences in nitrogen, methane, and carbon dioxide adsorption, confirming the change in surface chemistry by the introduction of non-polar, hydrophobic methyl groups [46].

The competing effects of reducing cross-linking leading to more open pore structure while simultaneously making the gel more vulnerable to collapse obviously suggest ORMOSILs are most appropriately used in blends with other materials to tune the chemistry of the gel. The variety of functional side groups available allow for the development of large monoliths useful for sensing, chromatography, ion exchange, and filtration, as well as potentially improving the surface interaction with encapsulated biological components.

2.8 Additives

A large variety of additives can be used in all sol-gel processing systems to alter a number of properties of the gel. Additives can change the viscosity of the sol which can change the hydrolysis kinetics by hindering molecular rearrangements that
may be necessary for a hydrolysis reaction to take place. This influence over the hydrolysis and condensation reactions alter the gelation process and final structure of the gel. Additives may also change surface chemistries and affect templating of the gel, providing some control over the final properties and structure of the material. The effect of any given additive is determined by both its chemistry and how it is used in processing. Researchers have explored the usage of organic compounds, including glycerol [47-49], polyethylene glycol (PEG)[50-52], sugars [48], surfactants [53], crown ethers [54], gelatin [55, 56], alginate [57], collagen [58, 59], formamide [60], organic acids [61], and many others. Metals, ions, and inorganic additives can also be used to alter electrical and optical properties of silica gel.

Of interest to this investigation was the use of PEG. This additive has been primarily used to stabilize the sol [50, 51], but can also reduce the surface tension of the liquid phase, reducing the interfacial energy to minimize shrinkage [3]. PEG is composed of repeated units of ethylene monomers joined by ether bonds, or repeating \((C_2H_4O)_n\) subunits, where \(n\) can vary from single digits to 10’s of thousands. In this investigation, PEG with a formula weight of 200 was used, which is relatively light, existing as a viscous liquid at room temperature. At a formula weight of 200, the PEG molecules are expected to contain 4 or 5 monomers. PEG adsorbs to the silica surface, attracted by hydroxyl groups and bonding at a density of approximately seven \((-C_2H_4O-)\) groups per square nanometer [3]. The presence of PEG will provide steric interference to reduce cross-linking through condensation, and also electrostatic interference by altering hydrogen bonding between neighboring uncondensed hydroxyl groups and/or silica in solution. Both effects should work to promote increased pore size and porosity. However, what is unclear is the effect PEG will have on the transport of divalent nickel, a cation that may interact electrostatically with the PEG monomers.

### 2.9 Diffusion

#### 2.9.1 Mathematical Treatment

Diffusion is defined as the transport of material due to the random motion of atoms and molecules [62]. This random motion is the result of thermal vibrations and
collisions between atoms, molecules, and particles. Although diffusion is analogous to conductive heat transport, it is not the result of other transport processes, including convection or other forms of active mass transport. The driving force of diffusion is a concentration gradient. The first formal statement of this process was formulated by Adolf Fick in 1855, applying a similar quantitative description developed for heat transfer by Fourier in 1822 [62]. The intent was to describe passive mass transport in physiological phenomena [63].

Qualitatively, when a concentration gradient exists, mass transfer will occur such that molecules will move from the region of higher concentration to the region of lower concentration. The rate will depend on the magnitude of the concentration gradient and other properties of the system, including the mobility of the diffusing material, which itself is dependent on many characteristics of the system. Stated generally, this can be written as:

\[ J = -D \frac{dC}{dx} \]

Where \( J \) is the flux of the diffusing material per unit surface area, \( \frac{dC}{dx} \) is the concentration gradient, \( D \) is the diffusion coefficient, and the sign is negative because the flux occurs away from the higher concentration, in the opposite direction of the gradient. This is known as Fick's First Law and its power lies in its ability to provide a quantitative description of a complex diffusion process by only the concentration gradient and one empirical proportionality constant, \( D \), the diffusion coefficient. The magnitude of this coefficient captures many of the important characteristics of the system controlling diffusion that are otherwise difficult or impossible to measure directly.

The above expression can be manipulated to describe the rate of concentration change per unit time in terms of the concentration curvature, or rate of change of the gradient, at that same point in space. Assuming the element is isotropic, \( D \) does not vary with direction, the expression can be restated as (in one dimension):

\[ J = -D \frac{dC}{dx} \]
This is commonly known as Fick’s Second Law, a second order partial differential equation, and it is where solutions to describe diffusion in a given system begin. Qualitatively, it means that the rate of concentration change at some point \( x \) is equal to the product of the diffusion coefficient and the second derivative of the concentration at that same point [63]. Since the second derivative is inversely proportional to curvature, or proportional to the reciprocal of the radius of curvature, the tighter the curvature, the larger the rate of change of the concentration gradient and the faster the diffusion process.

The above expression describes diffusion in one dimension of Cartesian coordinates. The expression can easily be transformed into three dimensions as well as cylindrical and spherical coordinate systems. See Crank’s thorough treatise *The Mathematics of Diffusion* for a complete treatment of these various forms, which are beyond the scope of the current investigation.

Describing the geometry used in this investigation, consider a fixed slab of thickness \( l \) with an initial uniform concentration of some material. In one dimension, the boundaries of this slab are fixed at \( x = 0 \), an impermeable surface, and \( x = l \), where the diffusing material will enter a finite volume of a well-stirred solution, making concentration in that solution a function of time only, not position. The initial concentration of the solute in the larger volume is initially zero, but as soon as the slab and solution are in contact, diffusion will take place, the concentration in the slab will decrease while the concentration in the solution will increase. The geometry is illustrated in the schematic figure below:
The analytical solution to Fick’s Second Law, describing the behavior of the above system is given as [62]:

Equation 2-1: Analytical solution to 1-D diffusion from a finite slab.

\[
\frac{M_t}{M_\infty} = 1 - \sum_{n=0}^{\infty} \frac{8}{(2n+1)^2\pi^2} \exp\left[\frac{-D(2n+1)^2\pi^2}{l^2} t\right]
\]

Where \(M_t\) is the mass that has entered the volume at some time \(t\), and \(M_\infty\) is the mass that will have entered at some equilibrium time, which is presumed to be calculable as the difference of the total mass of solute in the system and the mass of solute remaining in the gel slab at equilibrium. The form of the solution is due to a trigonometric series (Crank, Chapter 3). The above expression has been used in the literature to describe diffusion in a similar system of slab geometry [64]. However, it should be noted that the above solution works for planar slabs losing mass from both sides, as in the Ritger et al. investigation, and for planar slabs diffusing in one direction due to an impermeable barrier on one side, as in the current study. The difference is the characteristic length, \(l\), which is actually the half thickness of the slab.
in the two sided system, and the full thickness in the one sided system [62]. Due to superposition, the form of the solution for both systems is identical, only the characteristic length has changed.

For short time periods, characterized by times corresponding to $\frac{M_t}{M_\infty} \leq 0.60$, or less than approximately 60% of the total diffusion-driven transport, the following error function expression may also be used as an approximate solution [62, 64]:

Equation 2-2: Error function solution for 1-D diffusion from a finite slab.

$$\frac{M_t}{M_\infty} = 4 \left( \frac{Dt}{\ell^2} \right)^{1/2} \left[ \frac{1}{\pi^{1/2}} + 2 \sum_{n=1}^{\infty} \frac{(-1)^n \text{erf} \left( \frac{n\ell}{2\sqrt{Dt}} \right)}{\pi^{1/2}} \right]$$

Lastly, again for short times early in the diffusion process, the simplest approach to provide an estimate of $D$ uses the following exponential expression, an empirical approach rather than an analytical solution [62]:

Equation 2-3: Exponential approximation to analytical solutions.

$$\frac{M_t}{M_\infty} = 4 \left( \frac{D}{\pi \ell^2} \right)^{1/2} t^{1/2}$$

The exponential expression can be used to approximate $D$ in a variety of geometries, but since analytical solutions are readily available and easy to manipulate for the geometry used in this study, a comparison between the two to validate the less cumbersome exponential expression is possible. Assuming $D$ does not vary with concentration or time during the initial ~60% of the diffusion process, $D$ can readily be extracted from the data, which is explained further in Chapter 3.

Depending on the exact system employed and the geometry of that system, the solutions to Fick’s Second Law often require numerical methods or cumbersome transformations. The experimental set-up selected for this investigation is among the simplest to describe mathematically. For further discussion on more complex forms and their derivations, see Crank.
2.9.2 Types of Diffusion

There are three classes of diffusion processes, as classified by Alfrey, Gurnee, and Lloyd in 1966 [62]:

1. Case I, or ideal Fickian diffusion, characterized by a diffusion process that is much slower than relaxation processes or other structural changes in the material in which diffusion is taking place.
2. Case II diffusion, characterized by a diffusion process that is much faster than relaxation processes, meaning diffusion is not the rate limiting step of mass transport.
3. Case III diffusion, often called anomalous diffusion, characterized by diffusion and relaxation taking place on comparable time scales.

Case I is the classic or ideal model of diffusion which is practically useful in a number of real applications, providing a meaningful description of the diffusion process. The diffusion of atoms in a metal sample or ions in an aqueous solution, for example, both generally adhere to Fickian diffusion. Case II describes the situation where diffusion is not the rate limiting component of transport. For example, a hydrophilic organic polymer undergoing a slow glass transition to a swelled state under the advancing front of a material of differing composition is an example of Case II diffusion, presuming the swelling and resulting structural changes take much longer than the diffusion process.

Case III, or anomalous diffusion, takes place in the region between Case I and Case II. This occurs when diffusion and relaxation occur on similar time scales. A common example of a system that displays anomalous non-Fickian diffusion is diffusion of material into or from a swelling gel. If the swelling (or contraction) process occurs at a similar rate as diffusion, non-Fickian behavior may arise due to structural changes in the material altering the mobility of the diffusing species.

2.9.3 Diffusion in Silica Gels

Once formed, if kept hydrated, dilute silica gels will not swell and are actually more likely to undergo some amount of contraction and shrinkage. Contraction of the
gel, explained above, results from strong capillary forces and syneresis which will indeed alter the microstructure of the gel even if there is no macroscopic evidence of this structural rearrangement. More dilute gels may contract little because once fixed, there may not be a next-nearest-neighbor with a labile hydroxyl group available for condensation. In a more silica-dense gel, nearest neighbors may be close enough to condense. While it is obvious that such structural changes will change the mobility of diffusing species, it is less clear over what time scales these changes occur relative to the diffusion process and whether the cumulative effect constitutes Case III anomalous diffusion.

The gels of this investigation, particularly at lower silica concentrations, should be reasonably stable against such effects. Presumably, diffusion behavior in this system should be closer to Case I, or true Fickian diffusion. The diffusion process nears equilibrium at 24 hours, whereas gelation occurs on the order of minutes. Although the gel certainly evolves as it ages, the vast majority of structural arrangements are presumed to occur within a relatively short time during and immediately following gelation, putting approximately an order of magnitude difference in time scale between gel kinetics (minutes) and diffusion kinetics (hours), meaning the diffusion process is the rate limiting step of transport, suggesting predominantly Fickian behavior.

Polar solvents have been found to diffuse faster through porous silica as compared to nonpolar solvents [65], with bulk water having a diffusion coefficient on the order of $2.2 \times 10^{-9} \text{ m}^2\text{s}^{-1}$ and $D$ for trivalent Nd$^{3+}$ in water on the order of 3.0 to $5.1 \times 10^{-10} \text{ m}^2\text{s}^{-1}$ [66]. The differences in diffusion behavior are attributed to differences in surface interaction with the gel, as polar solvents participate in hydrogen bonding and non-polar solvents, like hexane, do not. Paradoxically, one would expect molecules that participate in hydrogen bonding to display slower diffusion, not faster, since molecules adsorbed to the surface are effectively immobilized which has the dual effects of constraining their transport and restricting the effective pore volume [65]. As a result, it remains unclear what mechanisms may be causing this observed behavior.
The above observations were also made in a microporous, condensed membrane that went through heat treatment, so D for water in a wet gel should be higher. Diffusion is expected to be the dominant form of transport within the gel because other bulk movements, like convection, have been eliminated [67]. This should remain true in the current investigation since the gels are exposed to the water solvent on only one side, shrinkage is considered to be minimal, and ion transport is predicted to be dominated by a diffusion mediated process, not bulk transport.

2.9.4  Diffusion of Divalent Nickel through Silica Gel

NiCl$_2$ is a metal salt that readily dissolves into Ni$^{2+}$ and Cl$^{-}$ in aqueous solution. Ni$^{2+}$ is expected to exist in octahedral coordination with six water molecules below at least 150°C, and its absorption spectrum is so characteristic that its concentration can actually be used to create tuned optical filters [68]. Nickel is not expected to participate in the silica sol-gel chemistry, so ions present in the sol prior to and throughout gelation are not expected to strongly influence or alter the process. As a result, Ni$^{2+}$ is expected to remain in solution and readily diffuse from gel samples, enabling easy concentration measurements through UV-vis spectrophotometry. In unconfined aqueous solution, octahedrally coordinated Ni$^{2+}$ is expected to have a diffusion coefficient on the order of approximately 1x10$^{-9}$ m$^2$s$^{-1}$ [69].

NiCl$_2$ has actually been used as a nickel source in the synthesis of a variety of Ni/NiO/SiO$_2$ composite materials. However, in the preparation of these materials, divalent nickel is presumed to remain in the pore volume of the silica gel and remain in solution until heat treatment induces a reduction reaction. Dextrose is typically added to the sol to support reduction and the heat treatment is done in an inert nitrogen atmosphere at temperatures exceeding 500°C [70, 71]. Another route for actively reacting divalent nickel into the composite material is by including a bifunctional amine-substituted siloxane into the sol precursor, such as 3-(2-aminoethylamino) propyltrimethoxysilane [72] or 3-(2-aminoethylamino) propyltriethoxysilane, among others [73]. In these cases, the amino-substituted siloxane is a covalent part of the gel network and also an active chelating agent for the divalent metal. By contrast, in the current investigation neither heat treatments nor amino-substituted siloxanes have been used, so the aqueous Ni$^{2+}$, aside from
possible alterations to the ionic environment, was not expected to participate in the gelation process of this system.

2.10 Silica Sol-Gel for the Encapsulation of Biological Components

Historically, sol-gel processing has been used to fabricate novel metal oxides, most often used for optical and electrical components, adsorption matrices, membranes, catalytic surfaces, and many other applications usually requiring robust thermal stability. More recently, beginning in 1989, sol-gel, and specifically silica sol-gel, has been used to encapsulate viable biological components, yeast being the first example [1]. The attraction of silica sol-gel is, as mentioned, that monoliths can be made at ambient temperatures with biologically compatible chemistry. Silica gel may also be more stable against swelling and biological attack, unlike many organic polymers which are commonly used for encapsulating biological components. This stability lends the material robustness against thermal and chemical degradation, which may prolong cell activity, even after cell senescence, and prevent contamination by foreign cells [50, 74].

A broad spectrum of applications is being explored with sol-gel encapsulation, including:

- Biosensors
- Bioreactors
- Drug release systems
- Tissue therapy systems
- Biological catalysts

Perhaps among the most powerful applications are those for drug release and tissue therapy. Silica sol-gel is porous enough to allow necessary gas and nutrient exchange to support living cells, yet the pores are too small to let larger objects, such as the cells themselves escape. This also means other cells, including contaminants, or white blood cells responsible for immune response, also cannot penetrate the gel. As a result, researchers have been exploring ways to encapsulate viable tissue cells, like pancreatic islets, to treat diabetes while avoiding the complications of immune
response and ultimate tissue rejection. Pancreatic islets have been shown to survive and successfully treat diabetic rats for numerous weeks, and the rats neither mounted an immune response against the encapsulated cells nor were they given immunosuppressant drugs [75].

Encapsulation also shows promise in the development of biosensors, biocatalysts, and bioreactors. Encapsulated cells still interact with their environment, and very quickly due to high diffusion rates. This allows sensor cells to provide a biological response to a product or contaminant in an effluent stream in real-time or something approaching real time, which has certain advantages in sensitive biological systems, like wastewater treatment, over time delayed laboratory analysis. Encapsulated cells may also be used to produce valuable enzymes, like invertase [76] or secondary metabolites, like hydrogen [52]. Encapsulation of biological components in silica gel and the variety of possible applications have been extensively reviewed elsewhere [47, 77-79].
Chapter 3 – Materials and Methods

This investigation was designed to explore diffusion in silica sol gels across a broad processing space relevant to applications in the encapsulation of biological components. As a result, a simple geometry with highly reproducible sample preparation and measurement protocol was used to screen through more than 150 samples. Silicon alkoxide precursors were used exclusively for two-step, acid-base catalyzed gel sample preparation, and the diffusing material was divalent Ni$^{2+}$, provided as NiCl$_2$ dissolved in BG-11 media for all samples. Each experimental step is described below.

3.1 Experimental Design

All gel samples were prepared in the same general manner, a two step process beginning with acid-catalyzed hydrolysis followed by base-catalyzed condensation, resulting in gelation (see additional information in §3.2). Silicon alkoxide was hydrolyzed with varying amounts of nitric acid and water in the first step, and then buffered media containing a small amount of strong base, potassium hydroxide, was added in step two to cause gelation. Silicon alkoxide precursors were used exclusively, including tetraethoxysilane (TEOS) (Alfa Aesar, Ward Hill, MA), methyltriethoxysilane (MTES) (Sigma Aldrich, St. Louis, MO), dimethyldiethoxysilane (DMDES) (Sigma Aldrich, St. Louis, MO), trimethylethoxysilane (TMES) (Tokyo Chemical Industry Co., Tokyo, Japan), and ethyltriethoxysilane (ETES) (Alfa Aesar, Ward Hill, MA). The structures of each precursor are shown below in Figure 3-1. Although the bulk of samples were produced with pure TEOS alone, the other precursors were used in smaller sample sets to explore the influence of alkyl groups present in the final gel on diffusion. The difference between the precursors listed is the number and type of alkoxide ligands bound to the central silicon atom, as shown. TEOS is a fully hydrolysable alkoxide while the other compounds contain alkyl ligands directly bound by an unhydrolysable silicon-carbon bond. These precursors are collectively known as organically modified silicates, or ORMOSILs.

Three parameters were varied over a broad range of processing space:
1. The molar ratio of water to alkoxide precursor in the initial hydrolysis (step 1), called the “water ratio,” was varied at 4:1, 10:1, and 20:1, water to silicon;

2. The molar ratio of nitric acid used to catalyze hydrolysis in step 1, or the “acid ratio,” varied from 1:0.005 to 1:0.02, silicon to acid, at three increments, 0.005, 0.01, and 0.02; and

3. The final concentration of silicon during gelation (step 2), or the “dilution ratio,” again expressed as a molar ratio of water to silicon, varied at 40:1, 60:1, 80:1, and 100:1.

The expectation was that these three processing parameters would exercise the strongest influence over the reactions determining the gel structure, namely hydrolysis and condensation. The range of these parameters were determined based on values that would be relevant to designing gels for the encapsulation of biological materials. A consequence of fabricating samples in this parameter space is that the silicon content is extremely low, approaching the point where forming a gel becomes tenuous, and the condition of the solution upon inducing gelation must be at approximately neutral pH and ambient temperature. Furthermore, depending on the sensitivity of the biological component, alcohol content must be minimized, and no other potentially cytotoxic solvents may be used. Despite these constraints, alkoxide precursors were the method of choice due to the flexibility of processing, the ability to minimize alcohol content, the expectation of a more porous structure, and the avoidance of excessive osmotic stress often associated with aqueous precursors like sodium silicate.
Figure 3-1: Common alkoxide precursors.

The water ratio took on three values, 4:1, 10:1, and 20:1. As discussed above, 4:1 is the minimum stoichiometric requirement for complete hydrolysis. However, hydrolysis is always in competition with condensation, so hydrolysis does not proceed to completion. In fact, in the 4:1 system, hydrolysis is expected to be far
from complete, leaving a significant percentage of partially hydrolyzed species and clusters with varying degrees of condensation. This has been verified experimentally through $^{29}\text{Si}$ NMR of sols prepared in similar manner [13, 23, 25]. The 10:1 and 20:1 systems provide excess water, which serves two purposes. First, and most importantly, excess water promotes more complete hydrolysis [19, 23]. Second, in conjunction with additional alcohol released through more complete hydrolysis, the additional water provides more solvent, diluting the concentration of the various silicon-containing species, reducing the size of oligomers formed at equilibrium, and stabilizing the sol. Without supplemental ethanol as a co-solvent, this system is not truly miscible, so stability is limited. Gelation will eventually occur if the sol is stored, however the kinetics are very slow without subsequent basic catalysis, requiring 100’s to 1000’s of hours for gelation to occur.

The acid ratio was varied at 1:0.005, 1:0.01, and 1:0.02, moles of silicon to moles of acid, or simply 0.005, 0.01, and 0.02. The acid is required to catalyze the hydrolysis reaction via a nucleophilic attack, as previously discussed [19, 80]. In this system, the acid truly acts as a catalyst and is regenerated during each hydrolysis reaction, making the amount of acid required relatively small. However, acid does accelerate the hydrolysis process, so this parameter was varied in order to determine its effect on the final gel structure through changes in the relative completion of hydrolysis. A more completely hydrolyzed sol should yield a more highly condensed gel since more labile hydroxyl groups will be available for condensation during the second step of sample preparation. Hydrolysis without acid catalyst is possible, only the kinetics are extremely slow, requiring 10’s to 100’s of hours to run to completion. This system was expected to complete hydrolysis in less than 2 hours.

The last parameter varied was the dilution ratio, describing the final silicon content of the gel, in moles of water per mole of silicon. Values of 40:1, 60:1, 80:1, and 100:1 were used. The content of silicon in the gel determines its density and influences its structure, so this parameter was examined in order to look at the effect of reduced silicon content on diffusion as the gels approach the point where there is not enough silicon present to form a gel, which is estimated to be at a ratio approaching 120:1 in this system, depending on initial sol preparation. Other
Researchers have verified the inverse relationship between diffusion rate and silica content [81]. Gel samples were prepared at a dilution ratio of 108:1 but were extremely soft and unstable once submerged in water, which is why 100:1 was the most dilute sample used in this investigation.

A full factorial experimental design was employed, covering all three parameters over the ranges mentioned above. All samples were prepared in triplicate, from the same precursor batch for each group of triplicates. Table 3-1, below, illustrates the sample space.

Table 3-1: Primary tetraethoxysilane sample set.

<table>
<thead>
<tr>
<th>Water Ratio (water:Si; [moles])</th>
<th>Acid Ratio (moles acid per mole Si)</th>
<th>40:1</th>
<th>60:1</th>
<th>80:1</th>
<th>100:1</th>
</tr>
</thead>
<tbody>
<tr>
<td>4:1</td>
<td>0.005</td>
<td>•</td>
<td>•</td>
<td>•</td>
<td>•</td>
</tr>
<tr>
<td></td>
<td>0.01</td>
<td>•</td>
<td>•</td>
<td>•</td>
<td>•</td>
</tr>
<tr>
<td></td>
<td>0.02</td>
<td>•</td>
<td>•</td>
<td>•</td>
<td>•</td>
</tr>
<tr>
<td>10:1</td>
<td>0.005</td>
<td>•</td>
<td>•</td>
<td>•</td>
<td>•</td>
</tr>
<tr>
<td></td>
<td>0.01</td>
<td>•</td>
<td>•</td>
<td>•</td>
<td>•</td>
</tr>
<tr>
<td></td>
<td>0.02</td>
<td>•</td>
<td>•</td>
<td>•</td>
<td>•</td>
</tr>
<tr>
<td>20:1</td>
<td>0.005</td>
<td>•</td>
<td>•</td>
<td>•</td>
<td>•</td>
</tr>
<tr>
<td></td>
<td>0.01</td>
<td>•</td>
<td>•</td>
<td>•</td>
<td>•</td>
</tr>
<tr>
<td></td>
<td>0.02</td>
<td>•</td>
<td>•</td>
<td>•</td>
<td>•</td>
</tr>
<tr>
<td></td>
<td>0.005 + 10% PEG200 →</td>
<td>•</td>
<td>•</td>
<td>•</td>
<td>•</td>
</tr>
<tr>
<td></td>
<td>0.005 + 25% PEG200 →</td>
<td>•</td>
<td>•</td>
<td>•</td>
<td>•</td>
</tr>
</tbody>
</table>

* = one gel sample, 120 samples total.

A smaller sample set was used to examine the effects of ORMOSIL’s and PEG200 on the diffusion of Ni\(^{2+}\). As discussed, covalently bound alkyl groups occupy bridging sites, reduce cross linking, and alter the electrostatic interactions between adjacent silicon-containing moieties and between the gel and the liquid phase. PEG200 can also influence the gel structure by altering surface chemistry and reducing capillary forces. Due to the relevance to encapsulation of biological components, the ORMOSIL gel sample set was truncated as compared to the pure TEOS sample set described above, including only the most dilute samples with the least amount of acid. All samples were prepared with a water ratio of 20, an acid ratio
of 0.005, and a dilution ratio of 80 or 100. Samples contained 10% on a molar basis of MTES, DMDES, TMES, or ETES, and 10% by weight of sol PEG200 in samples with a dilution ratio of 100. The sample set is illustrated in Table 3-2, below.

Table 3-2: ORMOSIL and PEG200 secondary sample set.

<table>
<thead>
<tr>
<th>ORMOSIL</th>
<th>Dilution Ratio</th>
<th>100:1</th>
<th>100:1 + 10% PEG200</th>
</tr>
</thead>
<tbody>
<tr>
<td>MTES</td>
<td>• • • • • • • • • • •</td>
<td>• • • • • • • • • • •</td>
<td>• • • • • • • • • • •</td>
</tr>
<tr>
<td>DMDES</td>
<td>• • • • • • • • • • •</td>
<td>• • • • • • • • • • •</td>
<td>• • • • • • • • • • •</td>
</tr>
<tr>
<td>TMES</td>
<td>• • • • • • • • • • •</td>
<td>• • • • • • • • • • •</td>
<td>• • • • • • • • • • •</td>
</tr>
<tr>
<td>ETES</td>
<td>• • • • • • • • • • •</td>
<td>• • • • • • • • • • •</td>
<td>• • • • • • • • • • •</td>
</tr>
</tbody>
</table>

* = one gel sample, 36 samples total.

### 3.2 Sample Preparation

#### 3.2.1 Hydrolysis

Hydrolysis was performed similarly for all sol precursor batches. Components, including alkoxide precursors, water, and acid were combined in a 50 mL Erlenmeyer with a small magnetic stir bar. A typical sol precursor sample with a molar ratio of water to silicon to acid of 20:1:0.005, respectively, contained 8.08 mL deionized water, 5.00 mL TEOS, and 112 µL of 1.0M nitric acid. Other samples, as discussed above, contained varying amounts of water and/or acid, depending on the sample formulation being prepared. The unhydrolyzed alkoxide precursor is not miscible in water, so initially the solution is phase separated.

The flask was then placed on a magnetic stir plate that contained a heating element. The liquid was continuously and vigorously stirred for approximately 90 minutes over mild heat of approximately 60°C. This accelerated the hydrolysis reaction and allowed for the evaporation of some ethanol as it evolved during hydrolysis. Silicic acid has limited solubility in water and the hydrolysis reaction produces ethanol, which is an effective co-solvent for both water and unhydrolyzed or partially hydrolyzed alkoxide species. Therefore, within approximately 30 minutes, the solution became homogenous, allowing hydrolysis to proceed more readily with all relevant compounds in a single phase. After approximately one hour, the heating
element was turned off and stirring continued for approximately 30 minutes to allow the sol to cool to room temperature, \(~25\degree C\). Once cooled, the sol was immediately used in the preparation of gel samples, described below.

### 3.2.2 Gelation

The second step of fabricating the gel samples after acid-catalyzed hydrolysis was base-catalyzed condensation, which resulted in gelation. This was achieved by the addition of BG-11 media containing 35 mM HEPES organic acid buffer, 500 mM NiCl$_2$, and an amount of 1.0 M KOH stoichiometrically equivalent to the amount of acid present in the sol being used to form the sample. This would neutralize the sol and bring the final solution to a pH of approximately 7.5, the same as the BG-11 media as originally prepared. The volume of 500 mM NiCl$_2$ BG-11 was held constant so all samples contained the same final concentration of Ni$^{2+}$ at 300 mM. As mentioned, the Ni$^{2+}$ was not expected to play any part in the sol-gel chemistry, but whatever ionic effects it may have exerted on the gelation process were uniform throughout and considered to be background. The ratio of silicon in the final gel was altered by using different volumes of sol precursor and BG-11 without NiCl$_2$. The final volume of all samples was 8.0 mL, which filled a 35 mm Petri dish nearly to the brim, for a mean gel thickness of 8.3 mm. A schematic of a typical sample is shown below.

![Schematic of gel sample](image)

**Figure 3-2: An illustration of a typical gel sample.**

NiCl$_2$ has limited solubility in alkaline solutions, so in order to prevent precipitation, successful preparation of translucent gels was sequence dependent. The sol was initially added to a 35 mm Petri dish. In a separate beaker, the specified
amount of BG-11 without NiCl$_2$ was mixed with the appropriate amount of 1.0 M KOH. After the BG-11 and KOH were mixed, then the BG-11 containing NiCl$_2$ was added to the beaker. For example, a typical 100:1 gel beginning with a sol containing a ratio of 0.005 acid would contain 1.6 mL sol, 1.6 mL BG-11, 12 µL 1.0 M KOH, and 4.8 mL BG-11 with 500 mM NiCl$_2$. The non-sol components are combined in that order, BG-11 then base followed by BG-11 with NiCl$_2$. If the KOH is added directly to the solution containing NiCl$_2$ prior to being diluted, the pH is locally too alkaline and a nickel hydroxide precipitate forms. This procedure avoids the formation of a precipitate. After all the components aside from the sol are well mixed, the contents of the beaker are poured into the Petri dish containing the sol, and a gel forms within a few minutes. Depending on the formulation, the gel may form anywhere on the order of 1 minute to 15 minutes. The most dilute gels with a poorly hydrolyzed sol may take as long as 30 minutes to fully form into a gel. Approximately 30 minutes after pouring, the lids were placed on the Petri dishes and they were wrapped in parafilm in order to minimize evaporation and any shrinkage of the gel that may result from the evaporation of the mother liquor. The experimental trial began approximately 24 hours later.

### 3.3 Diffusion Analysis

#### 3.3.1 Mathematical Treatment

The diffusion coefficient can be estimated with concentration or mass flux data over a time series, provided an appropriate solution to Fick’s Second Law is determined. The geometry of the sample and the method of monitoring diffusion is critically important to correctly determining the diffusion coefficient, D, of any sample, yet, in principle, the experimental procedure is relatively simple. In this analysis, the concentration of Ni$^{2+}$ in the bulk water solution was measured as it increased over time, a result of Ni$^{2+}$ diffusing out of the gel sample due to the concentration gradient between the gel and the bulk. The concentration of Ni$^{2+}$ in the gel correspondingly decreased, although it was not directly measured, and mass conservation was maintained throughout the system as gel and solution approached the same equilibrium Ni$^{2+}$ concentration of 10 mM throughout the entire volume.
Equation 2-1, given as:

\[ \frac{M_t}{M_\infty} = 1 - \sum_{n=0}^{\infty} \frac{8}{(2n + 1)^2 \pi^2} \exp \left[ \frac{-D(2n + 1)^2 \pi^2}{l^2} t \right] \]

was rewritten as:

Equation 2-1b

\[ \frac{C_t}{C_\infty} = 1 - \sum_{n=0}^{\infty} \frac{8}{(2n + 1)^2 \pi^2} \exp \left[ \frac{-D(2n + 1)^2 \pi^2}{l^2} t \right] \]

because concentration was being observed through absorbance measurements. Although mass transported could have been explicitly determined as well, a unitless ratio of concentration as a function of time over the equilibrium concentration is conceptually and mathematically equivalent to a unitless ratio of mass transported as a function of time to mass transported at equilibrium.

Similarly, Equation 2-3, given as:

\[ \frac{M_t}{M_\infty} = 4 \left( \frac{D}{\pi \ell^2} \right)^{1/2} t^{1/2} \]

was rewritten as:

Equation 2-3b

\[ \frac{C_t}{C_\infty} = 4 \left( \frac{D}{\pi \ell^2} \right)^{1/2} t^{1/2} \]

Lastly, Equation 2-3b can be written as:

\[ \frac{C_t}{C_\infty} = 4 \left( \frac{D}{\pi \ell^2} \right)^{1/2} t^n \]

Where \( n \) is the “diffusion exponent,” describing the time dependence of the diffusion process in question. For ideal Fickian diffusion (Case I), \( n = 0.5 \), but \( n \) may deviate
from 0.5 to empirically capture some aspects of the system geometry that deviate from ideal behavior (see Discussion) [64].

3.3.2 Experimental

The concentration of Ni$^{2+}$ was monitored spectrophotometrically by a UV-Vis spectrophotometer (Spectronic Genesys 10 Bio, Thermo Scientific, Waltham, MA). This spectrophotometer has a dual light source composed of tungsten and deuterium lamps with a monochromator capable of measuring absorbance in the range of 190 nm to 1100 nm. Ni$^{2+}$ has a strong absorbance peak at 395 nm, in the near UV/blue region of the spectrum. This absorption peak has been used in a similar manner by other investigators to monitor the diffusion of Ni$^{2+}$ through various matrices and solvents, including silica sol-gels [69, 81]. The absorbance peak results from a characteristic excitation energy and decay process from outer shell electrons, which contributes to the distinguishing blue-green color of Ni$^{2+}$. The absorbance can then be used to calculate concentration via Beer’s Law, $A = \log \frac{P_0}{P} = \varepsilon bc$, where $A$ is the absorbance (equivalently expressed as optical density at the specified wavelength, or OD$_{\lambda}$), $P_0$ is the power of the incident light, $P$ is the power of transmitted light, $b$ is the path length, $c$ is the concentration, and $\varepsilon$ is the molar absorptivity, a property of the dissolved species. In this system, $\varepsilon$ was not previously known, but the creation of a calibration curve provided an effective value for $\varepsilon$, a constant of proportionality, equivalent to the slope of the linear calibration curve. It should be noted that in regions of high absorbance, where $A$ exceeds a value of approximately 1.0, measurements may depart from linearity. However, the range of absorbance readings used in this investigation were well below that value, and verified to be linear over the entire experimental range, discussed below. Samples were measured in polystyrene cuvettes with a deionized water blank.

Before beginning diffusion experiments, the concentration of Ni$^{2+}$ was calibrated over a concentration range of 0 to 500 mM. It was found that the calibration curve was linear over that entire range of concentration and highly repeatable. Specific attention was given to a narrow lower range of 0 to 10 mM and it was found that this method could reliably distinguish differences in concentration of
approximately 0.3 mM with 95% confidence, effectively allowing resolution of small differences in concentration. Due to the nature of this experimental set-up, high resolution over the time series between slight differences in concentration was critically important. The calibration curve is shown below for the range of 0 to 10 mM.

![OD₃₉₅ Calibration for Ni²⁺](image)

**Figure 3-3:** Ni²⁺ absorbance calibration curve.

Samples were prepared so that the final concentration of Ni²⁺ in all samples was 300 mM. The volume of water in which the samples were placed was also constant at 240 mL across all experimental trials, meaning the final equilibrium concentration of Ni²⁺ through the whole volume, if equilibrium were achieved, would hypothetically be 10 mM, with an absorbance of approximately 0.054±0.002. A schematic of the experimental set-up is shown below in Figure 3-4.
Gel samples were affixed to the bottom of a dry glass vessel using two-sided adhesive tape. The water, previously decanted into a flask, was slowly poured into the vessel with the stir bar already active, and sample collection began immediately. Samples of approximately 1.0 mL were withdrawn through a rubber septum using a syringe, placed in a polystyrene cuvette, and absorbance was promptly measured in a spectrophotometer (water blank). After measurement, the sample was returned to the reaction vessel. The top of the vessel and the access ports, where the sample was returned, were kept closed to minimize water losses to evaporation.

Below (Figure 3-5) is an illustration of how the experiment proceeds, with the gel sample losing Ni$^{2+}$ and the bulk solution gaining it, acquiring a blue-green color.
Samples were collected at identical time points through all trials. The frequency was highest during the beginning of the experiment, when diffusion was presumed to be fastest due to the magnitude of the concentration gradient, and then sampling frequency slowed as diffusion slowed. During a typical experiment, samples were collected and measured immediately upon the addition of water and subsequently at intervals of 2, 5, 10, 20, 40, 60, 90, 120, 180, 240, 300, and 360 minutes. A final sample was usually collected at 24 hours to verify samples were approaching the expected equilibrium value, although these data were not used in subsequent analysis.

### 3.3.3 Calculating the Diffusion Coefficient

Diffusion coefficients were calculated for all sample types using the following procedure:

1. Samples were analyzed in triplicate, one absorbance reading per sample per time point. Three absorbance measurements at each time point were averaged, standard deviations were calculated, and the calibration curve was used to convert mean absorbance and standard deviation to units of concentration. These data were then normalized by dividing through by the absorbance predicted at the equilibrium concentration to give all data as a proportion of equilibrium concentration reached in the water solvent at a given time.
2. The analytical solution, Equation 1b, was first used to calculate \( D \) for the data covering time points 2 minutes through 4 hours. The SOLVER add-in of Microsoft Excel was used to calculate \( D \) through minimizing the sum of squared variance, minimizing the difference between the data and the analytical solution. SAS v9.2 software (SAS Institute, Inc., Cary, NC) was also used to calculate \( D \) through analysis of variance (ANOVA) from the analytical solution for all TEOS gels (samples 1 through 36) as a comparison to the SOLVER results.

3. The exponential approximation given in Equation 2b was used to calculate \( D \), again via minimizing the sum of squared variance using SOLVER and ANOVA, assuming a diffusion exponent of \( n = 0.50 \). This was also applied to data from 2 minutes through 4 hours, with the exception of the few samples where \( C(t)/C_\infty \) exceeded 0.60 in less than 4 hours. In those instances, all points exceeding 0.60 were excluded.

4. Next, ANOVA was used to predict a different value of the diffusion exponent, \( n \), as used in the exponential expression. Prior to this point, \( D \) was the only parameter being calculated by all modeling approaches. This step added a second parameter. A different estimate of \( n \) was presumed to give a better description of this system, addressing deviations from ideal behavior not addressed in the previously employed expressions (see Discussion).

5. Lastly, the exponential expression was again used to predict a third estimate of \( D \), via SOLVER and least-sum of squared variance, using the value of the diffusion exponent predicted through the ANOVA analysis. This step was performed to verify whether there was any significant non-ideal behavior occurring in this experimental system that may not be captured by analytical solutions assuming ideal one dimensional diffusion.

The above routine was followed for all gels composed of only TEOS. Subsequent samples in the second round of experiments, containing either ORMOSIL and/or PEG200 were analyzed with only SOLVER given there was
insignificant differences between the estimates of D generated by SOLVER and the estimates generated by ANOVA (see Results and Discussion).

Figure 3-6, below, illustrates quantitatively in concentration and qualitatively in time approximately how each experimental trial proceeds. The concentration of Ni$^{2+}$ in the gel source sample decreases from 300 mM toward the equilibrium concentration of 10 mM as the concentration in the solution increases from zero initially toward the equilibrium concentration. The bottom curve represents the data used to estimate D, and the top curve was back calculated from the bottom curve data using a mass balance. Absorbance of the gel samples was not directly measured at any time during this experimental procedure. In fact, the concentration of the gel sample shown in the top curve represents a mean concentration in the entire volume of the gel. In reality, there would be a concentration profile, dependent on depth in the gel, with increasing concentration toward the bottom, impermeable side of the gel sample.

Figure 3-6: Semi-quantitative illustration of Ni$^{2+}$ concentration profile.

Concluding the analysis was a comparison of the multiple estimates of D determined from the various methods to assess any variation and correlate that variation with differences in processing parameters (see Results and Discussion).
Chapter 4 – Results

4.1 Diffusion Measurements

As discussed previously, all diffusion measurements were conducted in the same manner. Samples were loaded into dry reaction vessels, 240 mL of DI water was added while being actively stirred, and sampling began immediately upon addition of water. OD\textsubscript{395} was measured, and the 1 mL sample was promptly returned to the reaction vessel from which it came. The generated calibration curve was then used to convert all OD\textsubscript{395} measurements to normalized concentration of Ni\textsuperscript{2+} by dividing through by the OD\textsubscript{395} of the equilibrium concentration, 10 mM, giving a series of normalized concentration versus time. The figure below illustrates example data from one group of samples. Error bars equal one standard deviation and lines have been added simply for clarity, not to suggest trends. Plots of all experimental data covering the full combination of processing parameters are included in Appendix A.

![Figure 4-1: Diffusion data from 100:1 TEOS gels at 0.005 acid ratio, comparing different water ratios.](image-url)
As shown in Figure 4-1, the data look as expected, an asymptotic approach to a normalized concentration of 1.0, or complete equilibrium, with the steepest slopes at the beginning of the diffusion process when the concentration gradient is greatest. The error bars indicate one standard deviation as calculated from the triplicate samples. Also of note is the time at which data points exceed a normalized concentration of 0.60. This is predicted to be the approximate value where the exponential expression can no longer be used to calculate $D$. In most trials, data up to 240 minutes, or 4 hours, was usable, remaining under 0.60. In the few instances where data points exceeded 0.60 by 4 hours, those points were retained in using the analytical solution to estimate $D$, but excluded when applying the exponential solution.

In preliminary analysis of these graphical representations of the data, very few significant differences in diffusion behavior were readily apparent. The plots below, for example, compare all acid and dilution ratios for gels prepared with a water ratio of 10:1. As can be seen in the plots, error bars (one standard deviation) generally overlap and there is little discernable difference between samples.

(A)
Figure 4-2: Diffusion data from TEOS gels prepared with a 10:1 water ratio.
By design, the data were clustered in the earlier time points to describe diffusion at its fastest. However, even truncating the data to the first 60 provided little improvement in resolution, as shown below.

Figure 4-3: The first 60 minutes of the same data shown in Figure 4-2(B).

There was one instance where a significant difference in diffusion behavior was readily apparent. In the case of samples prepared with a low acid ratio (0.005) and low dilution ratio, 40:1 (high silica content), there was a clear difference resulting from water ratios during hydrolysis (see Figure 4-4, below). This can be explained by a combination of nearly complete hydrolysis of the precursor and relatively high silica content upon gelation. Under these conditions, there is enough silica to form a reasonably dense gel, in fact the most dense gels examined in this study. Furthermore, condensation will proceed more rapidly toward completion as compared to less well hydrolyzed samples, as would be expected certainly with the 4:1 water ratio gel and less so with the 10:1 gel. At this higher silica content, condensation would then be expected to produce increased syneresis, or expulsion of liquid phase as the gel contracts, which was observed (~10% linear shrinkage). The other gels in the same group, with lower water ratios during sol preparation, would be expected to
have less complete hydrolysis upon gelation, slowing condensation, and reducing syneresis over the time scale of these experiments. Indeed, these 40:1 samples were the only samples that displayed visible shrinkage, contracting slightly from the edges of the Petri dishes within which they were cast (the diameter contracted from an as-cast 35 mm to approximately 32 mm). The conclusion is that although this appears to be the result of improved diffusion, it is actually the result of syneresis, a completely different transport process. Gels with a strong tendency toward syneresis are not preferable for bioencapsulation because the increase of capillary pressure and resulting contraction of the gel could place lethal stress upon the encapsulated material.

![Effect of Hydrolysis Water Ratio, 40:1 gels](image)

**Figure 4-4**: Increased Ni$^{2+}$ transport in low water ratio, high silica gels.

In the same large group of gels made with low acid content, a similar, although less pronounced effect was observed at the opposite end of the dilution ratio spectrum, 100:1, where the gels contained the least amount of silica examined in this study. Diffusion appeared to be higher from the gels prepared with a 4:1 hydrolysis ratio, but this is not evidence of a true diffusion process. Under these conditions (poor hydrolysis, high dilution ratio of 100:1), the gels were very tenuous, on the cusp of not
forming solid gels at all. Indeed, at the end of experimental trials, these gels fell apart at the slightest mechanical agitation. Therefore, the apparent increase in Ni\(^{2+}\) transport is a result of some contribution from gel dissolution. Although hydrolysis and condensation are continuing after gelation, the silica content is low enough that next-nearest neighbors are separated by too great a distance to allow adequate condensation to strengthen the gel matrix. The gels prepared with 10:1 and 20:1 water ratios during hydrolysis, by contrast, were more completely hydrolyzed and therefore able to form stable gels at the 100:1 dilution ratio, which was observed experimentally, presumably a result of more condensation and better cross-linking throughout the matrix.

![Figure 4-5: Increased Ni\(^{2+}\) transport from low water ratio, low silica gels.](image)

Qualitatively, although there were few dramatic differences, these diffusion data appeared as expected. Estimates of the diffusion coefficients, D, for each sample type followed by statistical estimates of confidence intervals on those estimates of D were then performed to determine if significant differences existed between sample types.
4.2 Analytical Solution Estimate of Diffusion Coefficient

The diffusion coefficient, $D$, was calculated for all samples using an analytical solution of Fick’s Second Law taking the form of a trigonometric expansion, as shown in Equation 2-1b. Values were calculated in Microsoft Excel using the Solver function to vary $D$ in order to minimize the sum of squared variance between the mean of the experimental data and values calculated from the analytical solution. ANOVA was also used to produce the same estimate for all TEOS samples. Values of $D$ varied surprisingly little over this broad range of processing space, covering a range from approximately $1.4 \times 10^{-10}$ m$^2$s$^{-1}$ to $6.3 \times 10^{-10}$ m$^2$s$^{-1}$, as estimated with the analytical solution by SOLVER. This corresponds to approximately 14% to 63% of $D$ for Ni$^{2+}$ in unconfined aqueous solution, estimated to be approximately $1 \times 10^{-9}$ m$^2$s$^{-1}$. Results are shown in Table 4-1 below.

The analytical solution used was expanded to five terms for these calculations. Crank had suggested that expansion of the expression beyond this number of terms in order to achieve increased accuracy would imply that an alternative solution should be employed, like the error function solution [62]. The solution was expanded to sixteen terms for a small subset of samples and the results were unaltered to three significant figures (data not shown), so it was assumed that five terms, in addition to being a practical limit to applying the solution, also provides as accurate a result as can be expected from this expression. The error function solution was also applied to the same subset, and the results were nearly identical, varying slightly in the third significant figure for some samples (data not shown).

In calculating the estimates of $D$ with the analytical solution, the initial data point was excluded to minimize sampling error introduced with time “zero” and to ensure comparison between analytical and exponential solutions was from the exact same data set, providing a better comparison. Although every effort was made to collect initial samples at a true time zero, there were small variations in time, on the order of 10’s of seconds, and varying degrees of mother liquor above the gel sample, due to syneresis, providing a small but non-trivial sudden flux of Ni$^{2+}$ to the reaction volume.
Table 4-1: Analytical solution estimated values of the diffusion coefficient for TEOS gel samples, calculated by Solver and ANOVA.

<table>
<thead>
<tr>
<th>Gel type</th>
<th>Water Ratio</th>
<th>Acid Ratio</th>
<th>Dilution Ratio</th>
<th>D Analytical Solver</th>
<th>D Analytical ANOVA</th>
</tr>
</thead>
<tbody>
<tr>
<td>69</td>
<td>0.005</td>
<td>4</td>
<td>40</td>
<td>2.17E-10</td>
<td>2.42E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.72E-10</td>
<td>2.95E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>2.53E-10</td>
<td>2.77E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>4.02E-10</td>
<td>4.53E-10</td>
</tr>
<tr>
<td>69</td>
<td>0.01</td>
<td>4</td>
<td>40</td>
<td>2.00E-10</td>
<td>2.33E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.09E-10</td>
<td>2.31E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>1.81E-10</td>
<td>2.11E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>3.32E-10</td>
<td>3.59E-10</td>
</tr>
<tr>
<td>69</td>
<td>0.02</td>
<td>4</td>
<td>40</td>
<td>3.06E-10</td>
<td>3.02E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.06E-10</td>
<td>2.06E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>1.55E-10</td>
<td>2.00E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>2.35E-10</td>
<td>2.62E-10</td>
</tr>
<tr>
<td>69</td>
<td>0.005</td>
<td>10</td>
<td>40</td>
<td>2.23E-10</td>
<td>2.46E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.18E-10</td>
<td>2.39E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>2.20E-10</td>
<td>2.33E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>1.94E-10</td>
<td>2.18E-10</td>
</tr>
<tr>
<td>69</td>
<td>0.01</td>
<td>10</td>
<td>40</td>
<td>3.78E-10</td>
<td>4.21E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.79E-10</td>
<td>2.92E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>2.80E-10</td>
<td>3.04E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>1.74E-10</td>
<td>2.09E-10</td>
</tr>
<tr>
<td>69</td>
<td>0.02</td>
<td>10</td>
<td>40</td>
<td>2.66E-10</td>
<td>2.90E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.47E-10</td>
<td>2.34E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>1.44E-10</td>
<td>1.79E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>2.63E-10</td>
<td>2.76E-10</td>
</tr>
<tr>
<td>69</td>
<td>0.005</td>
<td>20</td>
<td>40</td>
<td>6.27E-10</td>
<td>7.53E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.32E-10</td>
<td>2.59E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>2.39E-10</td>
<td>2.66E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>2.27E-10</td>
<td>2.50E-10</td>
</tr>
<tr>
<td>69</td>
<td>0.01</td>
<td>20</td>
<td>40</td>
<td>2.55E-10</td>
<td>2.78E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.12E-10</td>
<td>2.31E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>1.74E-10</td>
<td>2.01E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>2.06E-10</td>
<td>2.23E-10</td>
</tr>
<tr>
<td>69</td>
<td>0.02</td>
<td>20</td>
<td>40</td>
<td>4.60E-10</td>
<td>4.77E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.08E-10</td>
<td>2.40E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>3.04E-10</td>
<td>2.77E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>3.09E-10</td>
<td>3.32E-10</td>
</tr>
</tbody>
</table>

* All values of D are in units of m²s⁻¹
Confidence intervals (95%) on the calculated values of D were estimated in SAS for the above TEOS samples. The plot below shows estimates of D as tabulated above with the corresponding confidence intervals on the values predicted via ANOVA. The x axis is sample ID for the 36 sample types, shown below in Table 4-2.

![Global Results from Analytical Solution, TEOS gels](image)

Figure 4-6: Solver and ANOVA estimates of D from analytical solution.

Table 4-2: TEOS gel sample composition and ID numbers.

<table>
<thead>
<tr>
<th>Sample Type (water/acid/dilution)</th>
<th>ID</th>
<th>Sample Type (water/acid/dilution)</th>
<th>ID</th>
<th>Sample Type (water/acid/dilution)</th>
<th>ID</th>
</tr>
</thead>
<tbody>
<tr>
<td>4/0.005/40</td>
<td>1</td>
<td>10/0.005/40</td>
<td>13</td>
<td>20/0.005/40</td>
<td>25</td>
</tr>
<tr>
<td>4/0.005/60</td>
<td>2</td>
<td>10/0.005/60</td>
<td>14</td>
<td>20/0.005/60</td>
<td>26</td>
</tr>
<tr>
<td>4/0.005/80</td>
<td>3</td>
<td>10/0.005/80</td>
<td>15</td>
<td>20/0.005/80</td>
<td>27</td>
</tr>
<tr>
<td>4/0.005/100</td>
<td>4</td>
<td>10/0.005/100</td>
<td>16</td>
<td>20/0.005/100</td>
<td>28</td>
</tr>
<tr>
<td>4/0.01/40</td>
<td>5</td>
<td>10/0.01/40</td>
<td>17</td>
<td>20/0.01/40</td>
<td>29</td>
</tr>
<tr>
<td>4/0.01/60</td>
<td>6</td>
<td>10/0.01/60</td>
<td>18</td>
<td>20/0.01/60</td>
<td>30</td>
</tr>
<tr>
<td>4/0.01/80</td>
<td>7</td>
<td>10/0.01/80</td>
<td>19</td>
<td>20/0.01/80</td>
<td>31</td>
</tr>
<tr>
<td>4/0.01/100</td>
<td>8</td>
<td>10/0.01/100</td>
<td>20</td>
<td>20/0.01/100</td>
<td>32</td>
</tr>
<tr>
<td>4/0.02/40</td>
<td>9</td>
<td>10/0.02/40</td>
<td>21</td>
<td>20/0.02/40</td>
<td>33</td>
</tr>
<tr>
<td>4/0.02/60</td>
<td>10</td>
<td>10/0.02/60</td>
<td>22</td>
<td>20/0.02/60</td>
<td>34</td>
</tr>
<tr>
<td>4/0.02/80</td>
<td>11</td>
<td>10/0.02/80</td>
<td>23</td>
<td>20/0.02/80</td>
<td>35</td>
</tr>
<tr>
<td>4/0.02/100</td>
<td>12</td>
<td>10/0.02/100</td>
<td>24</td>
<td>20/0.02/100</td>
<td>36</td>
</tr>
</tbody>
</table>
As can be seen from Figure 4-6 above, although not precisely identical, there is not a significant difference between the results generated by Solver and by ANOVA in SAS. With very few exceptions, the Solver estimate falls within the 95% confidence interval of the ANOVA estimate. This also highlights the difference between most of the samples and sample 25, the sample discussed above, prepared at the highest water ratio and silica content with the lowest acid catalyst. As mentioned, the apparent increase in diffusion is an artifact of syneresis.

The majority of the estimates of D fall around a mean of approximately $2.5 \times 10^{-10} \text{ m}^2\text{s}^{-1}$ with overlapping confidence intervals (this mean excludes sample 25). Samples 4 and 33 are significantly above this mean, while samples 8, 9, 17, 18, 19, and 36 appear to have slightly larger values of D, although with confidence intervals that overlap the bulk of samples centered around the mean. There is no readily apparent common factor, such as high dilution ratio, that accounts for these higher estimates of D across all samples.

4.2.1 Estimates of D in ORMOSIL & PEG-Containing Gels

As shown above, Solver and ANOVA did not generate significantly different predictions for D across the entire set of TEOS samples. Therefore, Solver was used exclusively to generate estimates of D for the second series of samples, containing various kinds of ORMOSIL and PEG200. These results are tabulated below in Table 4-3 and plotted in Figure 4-7. Calculated values of D ranged from approximately $1.9 \times 10^{-10} \text{ m}^2\text{s}^{-1}$ to $4.5 \times 10^{-10} \text{ m}^2\text{s}^{-1}$, with a mean of approximately $2.7 \times 10^{-10} \text{ m}^2\text{s}^{-1}$, or approximately 19% to 45% of D in unconfined solution.

The ORMOSIL samples displayed more variability across replicates, resulting in wider 95% confidence intervals. The reason for this behavior is unclear. However, despite this, two observations are immediately apparent. First, surprisingly, the addition of PEG200 has a small and inconsistent effect on overall diffusion, slowing it down as often as it improves the process. Secondly, the presence of 10% ETES, which incorporates some relatively large ethyl side groups into the gel matrix, causes a small but statistically significant increase in D at the 100:1 dilution. Sample 51 contains 10% ETES and no PEG and has an estimated D of $3.6 \times 10^{-10} \text{ m}^2\text{s}^{-1}$, which is
slightly above the background mean of approximately $2.7 \times 10^{-10}$ m$^2$s$^{-1}$, a mean nearly identical to the mean of the pure TEOS gels examined in the first series of samples.

Table 4-3: Calculated values of D from analytical solution using Solver for ORMOSIL and PEG-containing gels.

<table>
<thead>
<tr>
<th>Gel type</th>
<th>Hydrolysis Ratio</th>
<th>Acid Ratio</th>
<th>Dilution Ratio</th>
<th>D Analytical Solution</th>
</tr>
</thead>
<tbody>
<tr>
<td>TEOS</td>
<td>20</td>
<td>0.005, 10% PEG</td>
<td>80</td>
<td>2.41E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>2.46E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.005, 25% PEG</td>
<td>80</td>
<td>3.24E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>2.63E-10</td>
</tr>
<tr>
<td>ORMOSIL's**</td>
<td>10% MTES</td>
<td></td>
<td>80</td>
<td>1.91E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>4.47E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100 (10% PEG)</td>
<td>2.97E-10</td>
</tr>
<tr>
<td>ORMOSIL's**</td>
<td>10% DMDES</td>
<td></td>
<td>80</td>
<td>1.72E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>3.51E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100 (10% PEG)</td>
<td>1.86E-10</td>
</tr>
<tr>
<td>ORMOSIL's**</td>
<td>10% TMES</td>
<td></td>
<td>80</td>
<td>2.79E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>3.42E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100 (10% PEG)</td>
<td>3.42E-10</td>
</tr>
<tr>
<td>ORMOSIL's**</td>
<td>10% ETES</td>
<td></td>
<td>80</td>
<td>2.03E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>3.56E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100 (10% PEG)</td>
<td>2.50E-10</td>
</tr>
</tbody>
</table>

*All values of D are in units of m$^2$s$^{-1}$

** Hydrolysis ratio = 20 and acid ratio = 0.005 for all ORMOSIL samples.
Figure 4-7: Estimates of $D$ predicted by Solver for gels containing ORMOSIL or PEG200.

Table 4-4: Sample composition and ID for gels containing ORMOSIL and PEG200.

<table>
<thead>
<tr>
<th>Sample Type (water/acid/dilution)</th>
<th>ID</th>
<th>Sample Type (water/acid/dilution)</th>
<th>ID</th>
</tr>
</thead>
<tbody>
<tr>
<td>20/0.005/80 + 10% PEG</td>
<td>37</td>
<td>20/0.005/100 w/ 10% DMDES</td>
<td>45</td>
</tr>
<tr>
<td>20/0.005/100 + 10% PEG</td>
<td>38</td>
<td>20/0.005/100 w/ 10% DMDES + 10% PEG</td>
<td>46</td>
</tr>
<tr>
<td>20/0.005/80 + 25% PEG</td>
<td>39</td>
<td>20/0.005/80 w/ 10% TMES</td>
<td>47</td>
</tr>
<tr>
<td>20/0.005/100 + 25% PEG</td>
<td>40</td>
<td>20/0.005/100 w/ 10% TMES + 10% PEG</td>
<td>48</td>
</tr>
<tr>
<td>20/0.005/80 w/ 10% TMES</td>
<td>41</td>
<td>20/0.005/100 w/ 10% TMES + 10% PEG</td>
<td>49</td>
</tr>
<tr>
<td>20/0.005/100 w/ 10% MTES</td>
<td>42</td>
<td>20/0.005/80 w/ 10% ETES</td>
<td>50</td>
</tr>
<tr>
<td>20/0.005/100 w/ 10% MTES + 10% PEG</td>
<td>43</td>
<td>20/0.005/100 w/ 10% ETES + 10% PEG</td>
<td>51</td>
</tr>
<tr>
<td>20/0.005/80 w/ 10% DMDES</td>
<td>44</td>
<td>20/0.005/100 w/ 10% ETES + 10% PEG</td>
<td>52</td>
</tr>
</tbody>
</table>
4.3 Calculation of D from Exponential Approximation

The analytical solutions to the diffusion equation can take many potentially cumbersome forms depending on geometry, including trigonometric series, error functions, or series containing Bessel functions, among others. An exponential approximation, as given in equation 2-3b, can be used to estimate D for a variety of geometries, avoiding the complexity of the analytical solutions. This experimental set-up allows for a direct comparison between this exponential approximation and a reasonably simple analytical solution.

4.3.1 Calculation of D with Ideal Fickian Diffusion Exponent

The exponential approach is only considered a valid estimator of D for approximately the first 60% of the diffusion process [64]. As a result, the data set was slightly truncated for this analysis. As with the analytical solution, points at time zero were excluded, for the same reason (excessive sampling error in time). Furthermore, up to 4 hours, although most samples did not exceed a normalized concentration of 0.60, there were a few instances where 0.60 was exceeded and these data points were dropped. In the few instances where this occurred, only the 4-hour time point was eliminated. There were no samples where a normalized concentration of 0.60 was exceeded by 3 hours. Figure 4-8, below, illustrates linearized data as well as analytical and exponential predictions for gel 28, with a composition of 20:1 water ratio, 0.005 acid ratio, and 100:1 dilution ratio. As can be seen from the plot, the exponential prediction remains linear and deviates from the data and the analytical solution at a normalized concentration of approximately 0.60.

As above, estimates of D using the exponential expression were calculated using Solver and ANOVA. The results are shown in Figure 4-9 and Table 4-5, below, again by the same sample ID. This first iteration of calculations used a diffusion exponent of n = 0.50, assuming ideal Fickian diffusion behavior.
Figure 4-8: Linearized experimental data with analytical and exponential models for gel #28.

Figure 4-9: Solver and ANOVA estimates of D from exponential approximation.
Table 4-5: Exponential solution estimated values of the diffusion coefficient for TEOS gel samples, calculated by Solver and ANOVA.

<table>
<thead>
<tr>
<th>Gel type</th>
<th>Water Ratio</th>
<th>Acid Ratio</th>
<th>Dilution Ratio</th>
<th>( D ) Exponential Solver</th>
<th>( D ) Exponential ANOVA</th>
</tr>
</thead>
<tbody>
<tr>
<td>4</td>
<td>0.005</td>
<td></td>
<td>40</td>
<td>2.43E-10</td>
<td>2.42E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.96E-10</td>
<td>2.96E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>2.78E-10</td>
<td>2.78E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>4.51E-10</td>
<td>4.51E-10</td>
</tr>
<tr>
<td></td>
<td>0.01</td>
<td></td>
<td>40</td>
<td>2.26E-10</td>
<td>2.32E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.35E-10</td>
<td>2.31E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>2.10E-10</td>
<td>2.12E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>3.57E-10</td>
<td>3.57E-10</td>
</tr>
<tr>
<td></td>
<td>0.02</td>
<td></td>
<td>40</td>
<td>3.30E-10</td>
<td>2.99E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.05E-10</td>
<td>2.07E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>1.91E-10</td>
<td>2.00E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>2.61E-10</td>
<td>2.62E-10</td>
</tr>
<tr>
<td>10</td>
<td>0.005</td>
<td></td>
<td>40</td>
<td>2.49E-10</td>
<td>2.46E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.47E-10</td>
<td>2.38E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>2.46E-10</td>
<td>2.32E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>2.22E-10</td>
<td>2.18E-10</td>
</tr>
<tr>
<td></td>
<td>0.01</td>
<td></td>
<td>40</td>
<td>4.20E-10</td>
<td>4.20E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>3.00E-10</td>
<td>2.90E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>3.04E-10</td>
<td>3.04E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>2.01E-10</td>
<td>2.09E-10</td>
</tr>
<tr>
<td></td>
<td>0.02</td>
<td></td>
<td>40</td>
<td>2.91E-10</td>
<td>2.91E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.32E-10</td>
<td>2.34E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>1.74E-10</td>
<td>1.79E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>2.87E-10</td>
<td>2.74E-10</td>
</tr>
<tr>
<td>20</td>
<td>0.005</td>
<td></td>
<td>40</td>
<td>7.44E-10</td>
<td>7.44E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.59E-10</td>
<td>2.59E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>2.66E-10</td>
<td>2.66E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>2.53E-10</td>
<td>2.50E-10</td>
</tr>
<tr>
<td></td>
<td>0.01</td>
<td></td>
<td>40</td>
<td>2.92E-10</td>
<td>2.76E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.39E-10</td>
<td>2.30E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>2.02E-10</td>
<td>2.01E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>2.34E-10</td>
<td>2.22E-10</td>
</tr>
<tr>
<td></td>
<td>0.02</td>
<td></td>
<td>40</td>
<td>4.75E-10</td>
<td>4.75E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>60</td>
<td>2.35E-10</td>
<td>2.40E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>80</td>
<td>3.29E-10</td>
<td>2.72E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>3.32E-10</td>
<td>3.32E-10</td>
</tr>
</tbody>
</table>

* All values of \( D \) are in units of \( m^2 s^{-1} \)
As with the analytical solution, the calculated values of D from Solver and ANOVA, although not identical, were statistically the same. The values of D predicted by Solver consistently fell with the 95% confidence intervals predicted by ANOVA. The results from the exponential expression also very closely matched the results from the analytical solution, suggesting it is a valid approximation for this system (discussed more below).

Solver was then used to estimate D for the second series of samples containing ORMOSIL and PEG200. The results are tabulated and graphed below in Table 4-6 and Figure 4-9, respectively. As with the pure TEOS gels from the first series, there was excellent agreement between the analytical and exponential estimates of D, with the exponential estimates falling within the 95% confidence intervals of the analytical estimates, as shown in Figure 4-9.

Table 4-6: Estimates of D for ORMOSIL and PEG-containing gels, by Solver with exponential approximation.

<table>
<thead>
<tr>
<th>Gel type</th>
<th>Hydrolysis Ratio</th>
<th>Acid Ratio</th>
<th>Dilution Ratio</th>
<th>D, Exponential</th>
</tr>
</thead>
<tbody>
<tr>
<td>TEOS</td>
<td>20</td>
<td>0.005, 10% PEG</td>
<td>80</td>
<td>2.42E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.005, 25% PEG</td>
<td>80</td>
<td>3.50E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>2.68E-10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>100</td>
<td>2.90E-10</td>
</tr>
<tr>
<td>10% MTES</td>
<td></td>
<td>80</td>
<td>2.18E-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>100</td>
<td>5.12E-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>100 (10% PEG)</td>
<td>3.24E-10</td>
<td></td>
</tr>
<tr>
<td>ORMOSIL's**</td>
<td>10% DMDES</td>
<td>80</td>
<td>2.00E-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>100</td>
<td>3.87E-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>100 (10% PEG)</td>
<td>2.29E-10</td>
<td></td>
</tr>
<tr>
<td>10% TMES</td>
<td></td>
<td>80</td>
<td>3.03E-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>100</td>
<td>2.20E-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>100 (10% PEG)</td>
<td>3.67E-10</td>
<td></td>
</tr>
<tr>
<td>10% ETES</td>
<td></td>
<td>80</td>
<td>2.43E-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>100</td>
<td>3.82E-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>100 (10% PEG)</td>
<td>2.77E-10</td>
<td></td>
</tr>
</tbody>
</table>

*All values of D are in units of m²s⁻¹.
** Hydrolysis ratio = 20 and acid ratio = 0.005 for all ORMOSIL samples.
4.3.2 Estimate of D with Adjusted Diffusion Exponent

The diffusion coefficient, n, in equation 2-3b, is 0.50 for ideal Fickian diffusion. Small deviations from 0.50 can empirically account for geometric variations in the system not described by D. For example, a value of \( n = 0.45 \) has been shown to be appropriate for slab or cylindrical geometries, with slight variation depending on aspect ratio [64]. Although this system is intended to be an example of one dimensional diffusion, where n should equal 0.50, the surface area of the gel was significantly smaller than the bottom surface area of the reaction vessel, creating the possibility of non-ideal behavior through imperfect mixing, vortices, or other anomalies of the geometry. Therefore, ANOVA was used in an attempt to predict a more descriptive value of n for this system. The diffusion exponent was estimated as \( n = 0.45 \pm 0.02 \).

With this value of n, Solver and ANOVA were again used to predict D, shown in the figure below (Figure 4-11). Similar to previous results, Solver and ANOVA
predicted nearly identical estimates of D, with the estimates from Solver falling within
the 95% confidence intervals of the estimates from ANOVA.

The figure below, Figure 4-12, plots results generated by ANOVA for
estimates of D from the analytical solution, the exponential approximation with n =
0.50, and with n = 0.45, with 95% confidence intervals for all three series. Four
observations are readily apparent from this figure. First, the estimates of D generated
by the analytical solution and the exponential approximation using a diffusion
coefficient of n = 0.50 are indistinguishable. Second, the estimates of D with the
adjusted diffusion exponent follow the same pattern as the other estimates
qualitatively. Third, and a serious fault of this approach, is the magnitude of the
estimates is consistently higher. In some instances, the estimate of D exceeds the
documented diffusion coefficient of Ni²⁺ in an unconfined aqueous solution, 1x10⁻⁹
m²s⁻¹, which does not make physical sense in this system. Lastly, and this is more an
artifact of the mathematical approach, the confidence intervals tend to be much more
broad. See Chapter 5 for additional discussion.

Figure 4-11: Solver and ANOVA estimates of D using diffusion exponent n = 0.45.
Figure 4-12: ANOVA estimates of D using analytical solution and exponential expression with $n = 0.50$ and $n = 0.45$.

The adjusted diffusion exponent was used to estimate D for the second series of samples containing ORMOSIL and PEG. However, the same trend was clear: results were qualitatively similar but the magnitude of the estimates of D were consistently too large, exceeding D for an unconfined solution in one instance. Therefore, these results with the adjusted diffusion exponent were not considered to yield a meaningful description of the system. As a result, subsequent analysis focused on results generated from the analytical solution and the exponential approximation with an ideal diffusion exponent of $n = 0.50$. 
Chapter 5 – Discussion

5.1 Water Hydrolysis Ratio

The hydrolysis ratio, or the water present during the first step of acid-catalyzed hydrolysis, was varied at 4:1, 10:1, and 20:1, molar ratio water to silicon. 4:1 provides the stoichiometric amount of water required for complete hydrolysis, while 10:1 and 20:1 provide substantial excess. It is known that in the presence of stoichiometric water, hydrolysis will not proceed to completion. The 4:1 gels were used as this baseline gel composition to contrast with 10:1 and 20:1 gels where hydrolysis was expected to be mostly complete. The rationale for including these samples was to examine gels where cross linking, and therefore over all density, might be reduced due to potential bonding sites being occupied by unhydrolyzed alkoxy groups. Of course, this also alters the aging process as hydrolysis will continue as water is generated through condensation. The final structure would be predicted to be more granular, with less surface area, but potentially higher porosity and larger pore size. The synthesis of 2:1 gels, with half the stoichiometrically required water, was attempted for additional comparison to the excess water systems, but the sol was not stable prior to gelation, preventing the synthesis of usable gels by these protocols.

The effect of restricted water during hydrolysis appeared to only manifest in the most dilute samples, with a dilution ratio of 100:1 and the lowest silica content of any samples included in this study. Samples 4 and 8 (both 4:1 hydrolysis ratio and 100:1 dilution ratio, 0.005 and 0.01 acid ratios, respectively) in particular had estimated values of D above the other samples in the group, at 4.0 and 3.3 x10^{-10} m^2s^{-1}, respectively. However, as mentioned above, this was not interpreted to be an actual improvement in diffusion through some structural change in an otherwise stable gel, but was likely the result of some limited dissolution of the gel. Although bulk modulus was not measured, these gels were extremely soft and fragile, falling apart upon the slightest mechanical manipulation. Some preliminary samples at this composition, not included in this study, actually did fully dissolve during the reaction process.
The differences between the 10:1 and 20:1 gels were more subtle. Under both conditions, hydrolysis is expected to go virtually to completion under these reaction conditions [19, 23, 24]. However, the difference in water volume should affect sol stability during hydrolysis and ultimately regulate the size of oligomers or particles that form in the sol prior to gelation. It is expected that in the 10:1 system, particles would be larger because the sol is twice as concentrated. Since solubility is limited, some condensation would be expected prior to gelation. A gel created from the higher water ratio would, therefore, be expected to have a finer structure, higher surface area, and higher porosity. Since extensive structural characterization was not conducted as part of this study, this hypothesis was not verified. However, what is certain is that there are no strong differences in the diffusion behavior of Ni\(^{2+}\) that can be easily and clearly linked to differences in water ratio used to prepare the sol. Additional structural characterization would be required to verify differences between the two sample groups.

In terms of compatibility with biological encapsulation, both the 10:1 and 20:1 systems remain viable options. Hydrolysis is expected to be complete, allowing for the removal of generated alcohol, and diffusion behavior is similar. The 4:1 system, however, is not expected to be a good candidate for biological encapsulation because hydrolysis is incomplete upon gelation. Subsequent hydrolysis within the gel will then yield potentially cytotoxic concentrations of alcohol.

### 5.2 Acid Content

Of the three processing parameters explored, the amount of acid catalyst probably had the weakest effect on overall diffusion behavior as compared to the other factors examined. Figure 5-1, A through C, provide a side by side comparison of estimated diffusion coefficients, arranged by water ratio groups, respectively. There are a few instances where D varies considerably, for example the 100:1 sample at low acid in 5-1 (A) and the 40:1 sample at low acid in 5-1 (C). As discussed previously, these large predictions of D are expected to be the result of dissolution and syneresis, respectively, not a diffusion transport process.
Estimated $D$ for 4:1 Water Ratio Gels, Grouped by Dilution Ratio

Estimated $D$ for 10:1 Water Ratio Gels, Grouped by Dilution Ratio
Aside from the mentioned outliers, all other factors being equal, the estimates of $D$ for samples with differing acid ratios typically have overlapping or very nearly overlapping confidence intervals. There are two possible explanations for this lack of a dramatic effect. The first is that there truly were few differences between the sols prior to gelation that manifested in structural changes in the final gels and the hydrolysis was simply a function of pH. Even at an acid ratio of 0.005, the pH of the sol was approximately 1 and remained approximately 1 for 0.01 and 0.02 acid ratio sols (data not shown). The isoelectric point for silicic acid is pH 2, below which hydrolysis of alkoxide precursors is expected to be favored [3]. Therefore, it would appear that the acid catalyst has the dual effect of dropping the pH below 2 and catalyzing the hydrolysis reaction. Once there is enough acid present to cause that change in pH, additional acid may not further accelerate hydrolysis significantly.
The other, more plausible explanation, is that an examination of diffusion simply did not provide the resolution to identify differences between the gels prepared with differing amounts of acid catalyst. This investigation was limited to a macroscopic process from which inferences about the microstructure of the gel are being made. Subtle differences in the microstructure likely exist, and differences in the sol prior to gelation certainly exist. The strong influence of the acid catalyst on hydrolysis rate has been well documented in the literature [4, 5, 12, 13], it simply did not appear to have a strong effect on the diffusion of Ni\(^{2+}\) in this investigation. Although it was not a part of this investigation, differences in the size regimes of particles present in the sols could be explored with optical techniques examining diffraction and scattering of light from the sol.

Although hydrolysis is possible without any catalyst, the time required is so long as to be impracticable for most applications. For biological encapsulation, it is therefore advisable to use the minimal amount of acid that will catalyze complete hydrolysis in a reasonable time-frame. This will minimize additional stresses on the biological component caused by the acid or the salts formed upon its neutralization.

### 5.3 Dilution Ratio & Silica Content

The final content of silica in the formed gel, controlled by the dilution ratio, had a surprisingly small effect on the diffusion rate of Ni\(^{2+}\). The hypothesis, which was not confirmed by these data, was that lower silica content would create a more open, less dense structure that would promote faster diffusion. As shown in Figures 5-1 (A), (B), and (C), the confidence intervals for most samples, all other factors being constant, overlapped or very nearly overlapped. The few exceptions, as already discussed, are in general cases where either hydrolysis was poor and silica content was low (Figure 5-1 (A)), or hydrolysis was nearly complete and silica content was high (Figure 5-1 (C)). The former is due to a soft unstable gel that partially dissolved over the course of the experiment, and the latter is due to syneresis, not a true diffusion transport process.
The gels containing less silica are certainly less dense. By conservation of mass, this must be true in order for a smaller amount of silica to form a stable gel of equivalent volume. The total silica content, from gels of a dilution ratio of 40:1 to gels of 100:1, covers a factor of 2.5, from approximately 3 to 8 wt% silica in the final gels, respectively. Despite the more dense structure, the diffusion of a divalent metal ion, like Ni\(^{2+}\), was not strongly altered. It is possible the gels of lower silica content have a finer structure of smaller mean particle size, producing higher porosity and higher surface area. The more tortuous path travelled by diffusing species may compensate for the increased porosity that might otherwise be expected to promote diffusion.

Gels of different dilution ratios were prepared from identical sols, however, suggesting that the mean particle size prior to dilution and gelation should have been identical. This would suggest the particulate structure of the gels should have been similar. However, at lower silica content, aggregation could have been diffusion limited, allowing some particles to remain in suspension longer and eventually dissolve, promoting ripening of the sol and coarsening of the gel. This would be a very different situation from that described above, but the result could be the same: increased diffusion from a more open structure being offset by necking and coarsening, which confines pore structure and reduces pore connectivity. Structural investigations would be required to resolve this question. The observation that diffusion of a small ionic species does not vary over such a broad range of silica content implies that silica content can be adjusted widely to best suit the encapsulation application with minimal change in transport of these cationic species.

### 5.4 Diffusion Models & Mathematical Treatment

#### 5.4.1 Analytical & Exponential Solutions

The values of D estimated by the analytical expression and the exponential expression with an ideal diffusion coefficient of \( n = 0.50 \) did not vary significantly, as shown in Chapter 4. This illustrates that the exponential approximation is a valid estimator of D for this system, prior to 60% of equilibrium concentration. The exponential expression is easy to linearize and very simple to use as a predictor of D, making it an attractive alternative to the analytical solution.
5.4.2 Solver versus ANOVA

The results generated by the Solver add-in of Microsoft Excel and ANOVA analysis performed by SAS were nearly identical. Although not precisely identical, the Solver estimates fell within the 95% confidence intervals of the ANOVA estimates in nearly all cases. The implications merely suggest an investigator may use either approach effectively depending on familiarity which each software package. The author used Solver to calculate values of D while receiving assistance from a statistician for estimates generated by ANOVA in SAS. The advantage of Solver is that it is easy to use and readily available with Microsoft Excel. Solver can also easily handle models with many more parameters, meaning this particular investigation underutilized the full capabilities of the program. The disadvantage is that Solver must be run for every single prediction made, it does not readily process an entire data set. It also does not generate useful estimate statistics, such as confidence intervals. These must be calculated separately, as was done for the second series of samples containing ORMOSIL and PEG200. The advantage of SAS is that it can process an entire data set at once. For example, what took 36 separate executions of Solver was done with one run in SAS, once the code was written. SAS also generates data set statistics such as confidence intervals. The disadvantage to SAS is the learning curve required to use the software and the availability. Ultimately, Solver is suitable for quick results from small data sets. As the number of samples and required calculations increases, Solver quickly becomes cumbersome and the use of SAS is warranted.

5.4.3 Geometric Effects

The experimental apparatus were designed to mimic one-dimensional diffusion from a sheet or slab. The analytical solution used to estimate D is specific to this geometry, so would be invalid if the geometry did not properly conform. Figure 5-2, below, illustrates a schematic of ideal one-dimensional diffusion from a slab into solution. The diffusing species can only go in one direction, into a perfectly mixed solvent above the slab. The actual geometry of this study is shown schematically in Figure 5-3, below. The gel slabs are in 35 mm Petri dishes, filled just to the brim with a slight negative meniscus. These sample are completely immersed in solvent,
deionized water, which is well stirred by a magnetic stir bar. However, the stir bar is beside the gel sample.

Although diffusion of Ni$^{2+}$ out of the surface of the gel is one-dimensional, three-dimensional phenomena were expected to be occurring throughout the volume of the solvent. Non-ideal behavior, such as flow of the solvent across the top of the gel sample or vortices or eddies forming around the edges of the Petri dish, could be altering the diffusion process.

![Figure 5-2: Schematic of ideal 1-D diffusion geometry.](image-url)
Use of the exponential expression and ANOVA to predict a different value of $n$, the diffusion exponent, was an attempt to verify whether non-ideal behavior was having a strong influence on diffusion and if this process could be better captured empirically by a geometry specific diffusion exponent. As mentioned, for ideal one-dimensional diffusion, the diffusion exponent is 0.50. For other geometries with three dimensional character, such as discs, spheres, and cylinders, the diffusion exponent is predicted to be some value less than 0.50, often in the range of 0.42 to 0.48, determined empirically [64]. Radial effects, dispersing the diffusing species in three dimensions, causes the exponent to be slightly smaller. Since non-ideal behavior in this system could potentially be having the same effect, it was expected that an exponent some value slightly smaller than 0.50 would better describe the system.

ANOVA predicted a value of 0.45 for the diffusion exponent, as mentioned previously, based on analysis of the first set of pure TEOS samples. The estimates of $D$ qualitatively followed the same pattern as estimates generated by the analytical solution and exponential approximation. However, the estimates were consistently higher by a margin of approximately of $4.0$ to $5.0 \times 10^{-10} \text{ m}^2\text{s}^{-1}$, which resulted in a few
estimates of $D$ which exceed the estimated $D$ of $\text{Ni}^{2+}$ in unconfined aqueous solution. This suggests that altering the diffusion exponent is not a valid approach in this system and that the diffusion in this geometry is best described as truly one-dimensional in nature. The altered diffusion exponent predicted by ANOVA is likely an artifact of the algorithm making the estimate. It may very well provide an exponential approximation that better describes the data empirically, but the predicted diffusion coefficients do not conform to the physical reality of the process. Furthermore, the fact that the results from the analytical solution and the exponential approximation with $n = 0.50$ were nearly identical further supports the conclusion that this system is most accurately described as one dimensional.

### 5.5 PEG200

Polyethylene glycol, or PEG, with a formula weight of 200 (PEG200) was used as an additive in an attempt to maintain pore structure. The PEG200 was primarily expected to reduce syneresis. A low formula weight PEG was selected in order to improve surface coverage of the additive. At a formula weight of 200, each molecule contained approximately four or five monomers. PEG is also expected to alter the surface tension of the liquid phase and the interfacial energy of the solid and liquid phases, resulting in reduced capillary forces upon drying. However, as these gels were not dried, this effect was not observed.

PEG is hydrophilic with some polar character due to carbon-oxygen bonds. As a result, any reduction in syneresis was likely due to steric obstruction of additional condensation reactions rather than electrostatic effects repelling nearest neighbor hydroxyl groups. However, as illustrated in Figure 5-4 below, addition of PEG200 in TEOS gels, up to a concentration of 25 wt% of sol precursor, did not significantly alter the diffusion behavior of $\text{Ni}^{2+}$. This could be due to offsetting effects of reduced syneresis and electrostatic interactions with $\text{Ni}^{2+}$, promoting and slowing diffusion, respectively. Or, it is possible that the effects of PEG do not strongly manifest until the gels are dried.
Figure 5-4: Estimated diffusion coefficients of TEOS gels containing 10 wt% and 25 wt% PEG200.

Although PEG did not strongly influence the diffusion of Ni$^{2+}$ in this investigation, PEG can be used to alter surface interactions with biological components, improving the biocompatibility of sol-gel materials for bioencapsulation. Therefore, it has been shown that PEG can be used for this purpose without strongly altering transport properties of divalent ions like Ni$^{2+}$.

5.6 ORMOSILs

Organically modified silicates, ORMOSILs, contain unhydrolysable carbon-silicon bonds which, as a result of their presence, incorporate organic functional groups into the gel matrix. This investigation included varying degrees of simple alkyl side groups, including methyl alkoxysilanes MTES, DMDES, and TMES, as well as a singly substituted ethyl alkoxysilane, ETES. The effects of using such additives in this context are threefold. First, the alkyl groups cause inductive and steric hindrance to acid catalyzed hydrolysis. The carbon-silicon bond is not as polarized as the oxygen-silicon bond, meaning a silicon with one or more alkyl groups is increasingly less
susceptible to nucleophilic attack. The large alkyl group close to the silicon simply obstructs hydrolysis on other side groups as well. However, for methyl and even ethyl alkyl groups, the steric effect is expected to be minimal.

Secondly, these alkyl groups occupy bonds that would otherwise be occupied by oxygen bridges between silicon atoms. Their presence is therefore expected to loosen the structure of the gel, and even provide some flexibility to gels that are typically brittle [39]. There is an optimal amount of ORMOSIL that can be added, however, as the lack of bridging will eventually degrade a flexible gel to a viscous liquid through the increased presence of methyl groups [44].

Lastly, alkyl groups are non-polar and hydrophobic, altering the interfacial energy between the solid matrix and liquid phase. This should serve to reduce capillary pressure and maintain pore size. The images below (Figure 5-5) illustrate the effect of 10 mol% MTES, replacing 2.5% of the total available bonding sites with a hydrophobic methyl group. This small increase in hydrophobicity has a clear and profound effect on interfacial energy between the solid and liquid phases. After gelation, these same effects may also slow syneresis through the occupation of potential bridging sites and electrostatically repelling nearby polarized hydroxyl groups. Steric and electrostatic effects on the surface may also slow coarsening of the gel structure. The gels in this investigation were not dried, therefore the latter two effects may be occurring while the former was not expected to exert strong influence.

(A)
Figure 5-5: Contact angles for water on a surface coated in TEOS gel (A), and a surface coated in a gel containing 10 mol% MTES (B).

Figure 5-6, below, shows the estimated diffusion coefficients of gels containing up to 10 mol% ORMOSIL. At the same 10 mol%, these ORMOSILs contributed one, two, or three methyl groups or one ethyl group per silicon, respectively, as shown in the figure. These methyl groups occupy 2.5%, 5%, and 7.5% of potential bonding sites, respectively. In this investigation, since drying was not performed, the dominant effect was predicted to be electrostatic repulsion of nearby hydroxyl groups, minimizing syneresis. No gels containing OROMSILs were observed to shrink during this investigation. However, the effect on diffusion was minimal. In most cases, the additional of ORMOSIL had the effect of slightly slowing diffusion in gels with a 80:1 dilution ratio, reducing diffusion coefficients from approximately $2.5 \times 10^{-10} \text{ m}^2\text{s}^{-1}$ in TEOS gels to approximately $2.0 \times 10^{-10} \text{ m}^2\text{s}^{-1}$ in similar gels containing 10 mol% ORMOSIL (TMES being the one exception). This is a puzzling result. Most likely, any increase in porosity caused by the presence of the ORMOSIL could be offset by a corresponding increase in tortuosity, neutralizing any improvement in diffusion.
Figure 5-6: Estimated diffusion coefficients of gels containing 10 mol% various ORMOSILs and 10wt% PEG200.

In contrast, gels prepared with a 100:1 dilution ratio appeared to have a slight increase in diffusion coefficient, to approximately $3.5 \times 10^{-10}$ m$^2$s$^{-1}$, TMES excepted. This is without the presence of PEG200, which then had the effect of slowing diffusion back into the range of comparable gels composed purely of TEOS, again TMES being the one exception.

These results suggest that at high dilution, some amount of ORMOSIL does provide a slight improvement of diffusion of $\text{Ni}^{2+}$. That improvement can then be neutralized by the addition of PEG. It is unclear why there are offsetting effects with these two alterations to the gels, although there is clearly an interaction between the alkyl surface moieties and the PEG molecules. One likely possibility is that the PEG monomers do not adhere smoothly to the solid surface, but are obstructed by the alkyl groups. This could significantly increase surface roughness and force PEG monomers to protrude into the pore volume, reducing the effective pore volume and slowing diffusion. In either case, the changes in diffusion coefficients are relatively small, most 95% confidence intervals overlap, suggesting yet again that the use of
hydrophobic alkyl ORMOSIL and/or PEG can be tuned to best suit the properties of the biological component without severe effects on the transport of ions like divalent nickel.

5.7 Divalent Nickel and Ionic Effects

The gels used in this investigation were prepared identically with 300 mM NiCl$_2$, added during the gelation step, so the nickel salt was present during gelation. Ionic compounds, like salt, can potentially have strong effects on the chemistry of soluble silica and the gelation process. Iler mentions interaction with hydroxides of aluminum, iron, manganese, and magnesium, along with other interactions with compounds of calcium, lanthanum, gadolinium, molybdenum, and potassium. However, strong interactions with compounds of nickel are not expected. Aluminum seems to have the strongest influence by far of any of the compounds explored to this point [3]. In fact, Ni$^{2+}$ has been added to silica gels during gelation with other investigations and was predicted to remain in solution, not reacting with the silicon species in any significant way [68, 70].

In slight contrast to the results presented, Takahashi et al. looked at transport of Ni(NO$_3$)$_2$ through wet silica gels and found that transport was relatively fast, with a $D$ estimated at approximately 6x10$^{-10}$ m$^2$s$^{-1}$, or approximately 60% of that in aqueous solution [69]. The gel samples were of similar density, approximately 6 wt% silica, but the nickel was added afterward by soaking the wet gel in 1.0 M Ni(NO$_3$)$_2$ for 24 hours to allow solvent exchange and penetration of Ni$^{2+}$ into the gel sample. The likely explanation for the difference in results is that in the current investigation, nickel was cast within the gel during gelation, possibly confining some nickel to inaccessible pore volume. In the Takahashi study, nickel was added after gelation, penetrating only the readily accessible pore volume, which would then allow more rapid diffusion when the process was reversed. The difference in diffusion coefficient estimates is not expected to be the result of any side reactions taking place between nickel and silicon species in the current investigation. As mentioned, although divalent nickel can be reacted with silica sol-gels, this generally requires amino substituted ligands, a redox partner (e.g. dextrose), and/or heat treatment of at least 500°C[70-73].
One limiting factor of the choice of divalent nickel for diffusion investigations is its relatively small size. This may be an asset for probing very small pore sizes, but once the mean pore size reaches a certain radius, diffusion of nickel may not be strongly altered and therefore loses its resolution. Hydrated Ni\textsuperscript{2+} in solution, in octahedral coordination, is estimated to have an effective diameter on the order of 1 nm. It has been demonstrated that diffusion is not restricted by the structure of the gel when the mean pore diameter is \( \geq 5 \) nm. In the pore size range of 3 to 5 nm, the diffusion coefficient decreases rapidly and smaller pore features strongly restrict diffusion [81]. From this evidence, it could be inferred that the mean pore size of all gels included in this investigation was at least 5 nm.
Chapter 6 – Conclusion

The diffusion of Ni$^{2+}$ from silica sol-gel monoliths prepared in a two step acid-base catalyzed process from alkoxide precursors was investigated. The processing space included molar ratios of water to silicon during hydrolysis of 4:1, 10:1, and 20:1, with acid molar ratios of 0.005, 0.01, and 0.02. The dilution ratio, or final silica content in the gel as a molar ratio of water to final silicon content, included values of 40:1, 60:1, 80:1, and 100:1, water to silicon, and the experimental space was a factorial design covering all combinations of these three parameters. This was the first series of samples. In addition, the second series of samples contained 10% ORMOSIL’s in the form of MTES, DMDES, TMES, and ETE S, prepared at a water ratio of 20:1, an acid ratio of 0.005, and dilution ratios of 80:1 and 100:1, included to examine the effect of hydrophobic alkyl groups within the gel. Lastly, PEG 200 was used as an additive in some samples, including all the gels containing ORMOSIL at a concentration of 10 wt% of sol, and concentrations of 10 and 25 wt% for TEOS gels at 20:1 water ratio, 0.005 acid ratio, and 80:1 and 100:1 dilution ratios. The diffusion coefficient, D, was calculated using an appropriate analytical solution to Fick’s Law and an exponential approximation, applicable for the first 60% of the diffusion process. Values of D did not vary broadly across this large processing space. D, as estimated by the analytical solution, ranged from 1.4x10^{-10} m^2 s^{-1} to 4.5x10^{-10} m^2 s^{-1} with a mean of approximately 2.5x10^{-10} m^2 s^{-1} for all samples within the study, excluding some samples with a dilution ratio of 40:1 which displayed pronounced syneresis (i.e. visible shrinkage). The estimated values of D range from approximately 15 to 45% of D estimated for Ni$^{2+}$ in unconfined aqueous solution, ~1x10^{-9} m^2 s^{-1}. Differences in D between sample types were small, but significant in some cases.

The processing space for this investigation was intended to capture the space relevant to sol-gel encapsulation of biological components. As such, gels were kept hydrated and never subjected to heat treatments, alcohol content was minimized, and the use of strong acids and bases was also kept to a minimum. No potentially cytotoxic solvents or additives were used in the process, which, in other circumstances, provide a great deal of flexibility in sol-gel processing. Given these constraints, it was surprising how little diffusion actually varied across this processing
space. This is probably attributable to three factors. First, the diffusing species, \( \text{Ni}^{2+} \), even in octahedral coordination, has an effective diameter much smaller than the expected pore size. Although the mean pore size and pore size distribution were not directly measured, both were expected to fall in a size regime approximately one order of magnitude larger than the radius of the coordinated nickel ion. As a result, variations in pore size or pore size distribution would not be readily apparent by observing \( \text{Ni}^{2+} \) diffusion, although variations in pore connectivity were expected to be observable through changes in diffusion. Second, maximum diffusion from a gel will never approach that observed in an unconfined aqueous system because there is a minimum threshold silica content required to form a gel. Once a stable gel has formed, the liquid phase is continuous yet somewhat confined, meaning there will be some immediate and distinct impediment to diffusion. Finally, the three processing parameters explored in this study are known to have a profound effect on the microstructure of silica gels. However, those differences are often not readily apparent until the gels are dried to form xerogels. These types of gels can have structural differences at the scale of single nanometers, which can be probed by the diffusion and adsorption of gaseous species, but cannot be effectively examined by the diffusion of small aqueous species.

The use of ORMOSILs also did not have a dramatic effect on diffusion. The presence of alkyl groups covalently attached to silicon should have the effect of opening structure by both occupying bridging sites and providing steric hindrance for condensation to occur at other bridging sites on the same silicon. However, they also alter the hydrophobicity of the gel surface. These two effects could be synergistic or opposed, depending on the material diffusing in the gel. In this system, the two effects were either insignificant or off-set each other, so the net rates of observed diffusion were not significantly different. The estimated diffusion coefficients for gels containing 10\% MTES, DMDES, and ETES (100:1 dilution ratio) were 4.5 \( \times 10^{-10} \), 3.5 \( \times 10^{-10} \), and 3.6\( \times 10^{-10} \) m\(^2\)s\(^{-1}\), respectively, slightly above the mean of 2.7 \( \times 10^{-10} \) m\(^2\)s\(^{-1}\) in that sample group. This supports the conclusion that, at least when transport of cations is a concern, attention in tailoring the gel to a given biological component can
focus on altering other aspects of the gel without drastic changes in transport properties.

One significant limitation of this investigation is that only divalent nickel was examined as the diffusing species. These results may extrapolate well to other cationic species with similar effective radii. However, diffusion of anionic species as well as larger molecules, including polar and non-polar compounds of biological significance, may be impossible to predict from these results. The expectation is that diffusion of these compounds would vary little as well, provided the effective ionic or molecular size is smaller than the pore size of the wet gel. The range over which D is estimated and varies could be significantly different than for Ni$^{2+}$ due to ionic interactions with the gel surface, but these effects are impossible to predict from first principles. Continuing research in this area by the author and collaborators is examining the diffusion behavior of other ionic species with differing valences as well as larger organic molecules, like methyl orange and other organic pH indicators.

The implication of this investigation for biological encapsulation is that attention can be focused on tailoring the material toward optimal performance of the biological component without significant changes in transport behavior, at least in the case of relatively small cations. Biological components require the diffusion of numerous species in order to survive, including dissolved gases, trace minerals, ions, and nutrients such as carbohydrates, volatile fatty acids, and others. This investigation illustrates that the diffusion of smaller species, particularly cations, is not strongly altered by these processing parameters in the phase space of very low silica and alcohol content. Formulations with the least amount of residual alcohol and the minimal amount of silica to maintain a structurally sound gel are therefore advisable. Both objectives can be facilitated by fully hydrolyzing the precursors with an acid catalyst in excess water.

Further investigation is required to determine how well this would hold for anions or uncharged small species, like dissolved gases. As the diffusing species become larger, as in the case of numerous organic compounds, it should be possible
to develop a correlation between the physical constraints imposed by the gel, the processing parameters, and the size of the diffusing species.
Chapter 7 – Future Work

This investigation has demonstrated that the diffusion behavior of Ni\(^{2+}\) is not strongly altered by variations in processing parameters in a range relevant to biological encapsulation. However, as mentioned, this observation may not hold for other compounds of biological importance. Future work will explore the diffusion behavior of compounds that are larger and/or have a different valence. For example, permanganate, MnO\(_4^{2-}\), is a divalent anion with a strong absorbance peak at 525 nm. It would be expected that MnO\(_4^{2-}\) will not diffuse in a similar manner as Ni\(^{2+}\) due to its negative charge and resulting, presumably repulsive, interaction with the hydroxyl groups on the surface of the silica gel. Methyl orange, a colorimetric pH indicator, is significantly larger than either permanganate or nickel and can be used to explore the diffusion of larger compounds.

In a biological system, ions and dissolved gases are metabolically important, but larger compounds, including organic acids, fermentation products, carbohydrates, proteins, etc., may be important both for the survival of the biological components and optimal performance of the composite system. Therefore, future work should also explore the diffusion of these compounds through silica sol-gel monoliths. It is expected that once the compounds approach a size regime on the order of the pore size, diffusion will begin to be significantly hindered.

This study did not explore any structural characterization of the gels which may be a fruitful area of additional investigation. Although structural features will almost certainly change upon drying, some optical techniques can provide information on structural features without altering the gels. Ellipsometry, for example, could potentially provide information about the dominant size scale of the gel features by analyzing how the gel scatters light, from the ultraviolet to infrared range. This technique will not provide fine resolution of the gel structure, but will give a coarse estimate of the size of its features, enabling a correlation between the size and diffusion behavior of a given analyte and the size of the structural features of the gel.
Lastly, the use of ORMOSIL’s in this investigation was extremely limited. Future work should explore a broader array of ORMOSIL’s, including those that contain larger side groups and side groups of differing chemical characteristics. This investigation examined the use of simple methyl and ethyl groups, whereas the variety available is enormous. Larger alkyl groups, including propyl, butyl, and even benzyl could have differing effects on the gel structure and diffusion. Other functional groups, including amino or saccharide side groups, are also available. The possible effects on gel structure and diffusion are difficult to predict, and the interaction with biological components is virtually impossible to predict. Therefore, use of these ORMOSIL’s must be explored through subsequent investigation.
Bibliography


<table>
<thead>
<tr>
<th></th>
<th>Reference</th>
</tr>
</thead>
</table>
Appendix A – Experimental Data
The following plots illustrate all experimental data in plots with a full factorial coverage of all samples. They are arranged first by water ratio (4:1 followed by 10:1 followed by 20:1) displaying data for dilution ratios at fixed acid content, and then by acid content at fixed dilution ratios. Finally, data for samples containing ORMOSIL’s and PEG200 are arranged in similar fashion.

4:1 Water Ratio Gels, Samples 1 – 12
Effect of Silica content, 0.01 Acid

Effect of Silica content, 0.02 Acid
10:1 Water Ratio Gels, Samples 13 – 24

Effect of Silica Content, 0.005 Acid

Effect of Silica Content, 0.01 Acid
Acid Effect, 60:1 Silica

Acid Effect, 80:1 Silica
20:1 Water Ratio Gels, Samples 25 – 36

Effect of Silica Content, 0.005 Acid, 20:1 Water Ratio

Acid Effect, 100:1 Silica
Effect of Acid, 40:1 Silica

Effect of Acid, 60:1 Silica
Effect of Acid, 80:1 Silica

Effect of Acid, 100:1 Silica
Comparison of Water Ratios at Low Acid Content

Effect of Hydrolysis Water Ratio, 40:1 gels
0.005 Acid

Effect of Hydrolysis Water Ratio, 60:1 gels
0.005 Acid
Effect of Hydrolysis Water Ratio, 80:1 gels
0.005 Acid

- 4:1
- 10:1
- 20:1

Effect of Hydrolysis Water Ratio, 100:1 gels
0.005 Acid

- 4:1
- 10:1
- 20:1
Comparison of Water Ratios at Medium Acid Content

Effect of Hydrolysis Water Ratio, 40:1 gels

0.01 Acid

Normalized Concentration

Time (min)

Effect of Hydrolysis Water Ratio, 60:1 gels

0.01 Acid

Normalized Concentration

Time (min)
Comparison of Water Ratios at High Acid Content

Effect of Hydrolysis Water Ratio, 40:1 gels
0.02 Acid

Effect of Hydrolysis Water Ratio, 60:1 gels
0.02 Acid
Effect of Hydrolysis Water Ratio, 80:1 gels
0.02 Acid

Effect of Hydrolysis Water Ratio, 100:1 gels
TEOS Gels with PEG200, Samples 37 – 40

Effect of PEG200 Content, TEOS gels, 20:1 water, 0.005 acid

Effect of ORMOSIL Content without and with PEG200, Samples 41 – 52

Effect of 10% ORMOSIL, 0.005 Acid, 80:1 gels
Effect of 10% ORMOSIL, 0.005 Acid, 100:1 gels

Effect of 10% ORMOSIL + 10%PEG200, 0.005 Acid, 100:1 gels