Design, Fabrication, and Actuation of Biomimetic Cilia

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This thesis presents the details behind the production of a novel biomimetic cilia system which will be useful in exploring the detailed mechanisms of fluid flow generation in biological cilia systems. The biomimetic system consists of an array of flexible, high-aspect-ratio magnetic structures of the same size as biological cilia, and is actuated by magnetic fields. First, I will describe the array of materials which are available for use in this system, including a couple of novel materials which I have designed for this work. Next, I will describe the steps necessary to fabricate large arrays of microscale biomimetic cilia with these materials. I will then formulate a mathematical model to predict the bending of these structures in an applied magnetic field, and demonstrate how this model is helpful in designing a biomimetic cilia system. Finally, I will describe several modes of actuation which are predicted by the model and have been verified by experiment in the biomimetic system, and discuss the biological relevance of these modes of actuation.
ACKNOWLEDGEMENTS

No good work is ever performed in isolation, and this work in particular is a product of intense collaboration and represents the work of many talented researchers, whose contributions are inextricable from my own. The single biggest factor in the success of these efforts has been the tireless work of my Right-Hand Man in Cilia, Adam Shields. It’s a rare privilege for a graduate student to have such a close and constant collaborator. Since Adam came on the project, shortly after I started work on it myself, he has complemented my every weakness with an unbelievable suite of strengths, which I believe will make him a much stronger experimentalist than I could ever manage to be. I’ve learned much from Adam about perseverance, about experimental precision, about pushing on and taking the next step when I’m too often content to pencil it in for “tomorrow.” The work in this dissertation would not have happened if not for Adam, and I expect his attitude towards research and his enthusiasm for his work will remain an inspiration to me as I move on to my next big adventure on my own – a prospect which, I’ll be honest, scares the hell out of me. Thanks, buddy.

As I move away from UNC toward whatever the future may bring, I’m very happy to trust the future of the cilia project to Briana Carstens, who in the short time I have had to work with her has proven to be extremely competent, motivated, and inspired. I’m confident that I’m leaving the cilia project in better hands than it’s ever been in.

There have been a variety of undergraduate students who have lent their time and talents to the cilia project as well, most notably Sarita Prakash, who has been working closely
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To my fellow NSRG’ers who have been constant workday companions: thanks, and best of luck to you all. You’ve made coming to work fun. Jerome, for your constant cheer and almost-as-constant reminders that the universe is still weird, after all these years, and we really don’t know squat about it. Jeremy, for being my MatLab muse and chemistry confidant; Vinay, bringer-of-coffee and singer-of-songs; Lamar, faithful optimist; and many others, too numerous to name. You’re the bizarre potpourri that make NSRG what it is, and the reason I don’t want to leave.

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<table>
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<th>Abbreviation</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>A/P</td>
<td>anterior / posterior</td>
</tr>
<tr>
<td>AAO</td>
<td>anodized aluminum oxide</td>
</tr>
<tr>
<td>AIBN</td>
<td>azobisisobutyronitrile</td>
</tr>
<tr>
<td>AM</td>
<td>acrylamide monomer</td>
</tr>
<tr>
<td>APMS</td>
<td>aminopropylmethyisiloxane</td>
</tr>
<tr>
<td>APS</td>
<td>ammonium persulfate</td>
</tr>
<tr>
<td>ATP</td>
<td>adenosine triphosphate</td>
</tr>
<tr>
<td>BA</td>
<td>N, N methylene bisacrylamide</td>
</tr>
<tr>
<td>Bis-CA</td>
<td>bisacryloyleystamine</td>
</tr>
<tr>
<td>CF</td>
<td>cystic fibrosis</td>
</tr>
<tr>
<td>citFF</td>
<td>citrate-stabilized ferrofluid</td>
</tr>
<tr>
<td>COOH-PEG</td>
<td>carboxy-terminated polyethylene glycol</td>
</tr>
<tr>
<td>CTAB</td>
<td>cetyl trimethylammonium bromide</td>
</tr>
<tr>
<td>DCM</td>
<td>dichloromethane</td>
</tr>
<tr>
<td>DEE</td>
<td>diethyl ether</td>
</tr>
<tr>
<td>EDAC</td>
<td>1-ethyl-3-(3-dimethylaminopropyl) carbodiimide</td>
</tr>
<tr>
<td>FFAAm</td>
<td>ferrofluid - polyacrylamide</td>
</tr>
<tr>
<td>FFPDMS</td>
<td>ferrofluid - polydimethylsiloxane</td>
</tr>
<tr>
<td>FFPDMS-C16</td>
<td>hexadecane-stabilized FFPDMS</td>
</tr>
<tr>
<td>FFPDMS-NH2</td>
<td>ferrofluid - PDMS-NH2</td>
</tr>
<tr>
<td>FFPVMS-NH2</td>
<td>ferrofluid - PDMS-NH2</td>
</tr>
<tr>
<td>Acronym</td>
<td>Full Name</td>
</tr>
<tr>
<td>-----------</td>
<td>-----------------------------------------------</td>
</tr>
<tr>
<td>GMA</td>
<td>glycidyl methacrylate</td>
</tr>
<tr>
<td>MEA</td>
<td>mercaptoethylamine</td>
</tr>
<tr>
<td>MEK</td>
<td>methyl ethyl ketone</td>
</tr>
<tr>
<td>MES</td>
<td>2-(N-morpholino)ethanesulfonic acid</td>
</tr>
<tr>
<td>NIB</td>
<td>neodymium iron boride</td>
</tr>
<tr>
<td>NMP</td>
<td>N-methylpyrrolidone</td>
</tr>
<tr>
<td>PAAm</td>
<td>polyacrylamide</td>
</tr>
<tr>
<td>PCL</td>
<td>periciliary liquid</td>
</tr>
<tr>
<td>PCTE</td>
<td>polycarbonate track-etched (membrane)</td>
</tr>
<tr>
<td>PDMS</td>
<td>polydimethylsiloxane</td>
</tr>
<tr>
<td>PDMS-NH2</td>
<td>amine-functionalized polydimethylsiloxane</td>
</tr>
<tr>
<td>PEG</td>
<td>polyethylene glycol</td>
</tr>
<tr>
<td>PES</td>
<td>poly(ether sulfone)</td>
</tr>
<tr>
<td>PGMA</td>
<td>poly(glycidyl methacrylate)</td>
</tr>
<tr>
<td>PVMS</td>
<td>polyvinylmethylsiloxane</td>
</tr>
<tr>
<td>PVMS-NH2</td>
<td>amine-functionalized polyvinylmethylsiloxane</td>
</tr>
<tr>
<td>SDS</td>
<td>sodium dodecyl sulfate</td>
</tr>
<tr>
<td>SEM</td>
<td>scanning electron microscope</td>
</tr>
<tr>
<td>SH-PEG</td>
<td>thiol-terminated polyethylene glycol</td>
</tr>
<tr>
<td>SQUID</td>
<td>superconducting quantum interference device</td>
</tr>
<tr>
<td>TBAF</td>
<td>tetrabutyl ammonium fluoride</td>
</tr>
<tr>
<td>TCEP</td>
<td>tris(2-carboxyethyl)phosphine</td>
</tr>
<tr>
<td>Abbreviation</td>
<td>Full Name</td>
</tr>
<tr>
<td>--------------</td>
<td>-----------</td>
</tr>
<tr>
<td>TEMED</td>
<td>tetramethylethylenediamine</td>
</tr>
<tr>
<td>TMAOH</td>
<td>tetramethylammonium hydroxide</td>
</tr>
<tr>
<td>TRIS</td>
<td>trishydroxymethylaminomethane</td>
</tr>
<tr>
<td>V/D</td>
<td>ventral / dorsal</td>
</tr>
</tbody>
</table>
LIST OF COMMON SYMBOLS

\( M \)  
magnetization per unit volume (A/m)

\( M_{\text{sat}} \)  
saturation magnetization per unit volume (A/m)

\( m \)  
magnetic moment (A m\(^2\))

\( E \)  
Young’s modulus (Pa)

\( I \)  
bending moment of inertia (m\(^4\))

\( f \)  
volume fraction of magnetic material

\( \mu_0 \)  
permeability of free space (units)

\( V \)  
volume of microrod (m\(^3\))

\( L \)  
length of microrod (m)

\( B \)  
magnetic field magnitude (T)

\( \nabla B \)  
magnetic field gradient magnitude (T/m)

\( \phi \)  
angle of the rod with respect to the vertical

\( \alpha \)  
angle of the magnetic moment with respect to the vertical

\( \psi \)  
angle of the magnetic field with respect to the vertical

\( \psi' \)  
angle of the magnetic field gradient with respect to vertical
Chapter I

INTRODUCTION

From sea urchin sperm to bacterial locomotion to immune defense, cilia and flagella are ubiquitous in biology. They are present in nearly every cell in the human body and perform a variety of functions within their dual roles of sensor and actuator. For example, cilia are present in the kidney, where they act as flow sensors, and in the lungs, where they transport mucus. In the reproductive system, cilia are responsible for transporting an unfertilized ovum along the fallopian tube toward the uterus, and their close cousins, flagella, provide essential locomotion for sperm. In fact, cilia are present in so many of the body’s organ systems that many researchers are turning their focus towards this ubiquitous feature of biology, and our understanding of the role of cilia in health and biology is rapidly expanding.

It has only been in recent years that scientists have discovered that cilia may play a role in transporting neurons in the brain\(^1\) and are present in the nodal plate of a developing embryo, where they help to establish the symmetries of the fetus.\(^2\)

Scientists and medical professionals have come far in understanding the critical roles that cilia play in their various manifestations, and have pinpointed cilia dysfunction as the cause of a wide variety of symptoms, including polycystic kidney disease, infertility, and situs inversus. However, despite the pervasiveness of cilia throughout biology, very little is known about the fundamental mechanisms by which cilia interact with the complex fluids which make up their environment.
Due to their microscale nature, cilia exist in a world in which viscous fluid forces outweigh inertial forces by orders of magnitude. Fluids at this scale exhibit no turbulence, and fluid flows are generally reversible. Under these conditions, it is often unclear how cilia perform the functions that they do, such as locomotion and transport. For example, cilia in the lung perform a vital function by continuously transporting a layer of protective mucus out of the lung via the trachea, preventing inhaled contaminants from penetrating the lung tissue and invading the body. However, it is unclear exactly what mechanisms the cilia employ to achieve such transport. Understanding the mechanisms behind mucociliary transport in the human lung not only stands to answer fundamental questions of microscale hydrodynamics, but may also aid in the development of better treatments for diseases caused by dysfunctional transport, such as cystic fibrosis.

Studies of biological samples, however, have proven difficult to control – it is extremely difficult to manipulate variables of interest independently due to the robustness built into the biological system. Manipulation of one variable may lead to unpredictable compensation in other areas of the parameter space of the system, as the ciliated tissue adapts to perform its evolutionarily mandated task. A biomimetic model of the system, however, can allow for straightforward exploration of the full parameter space – beat shape, beat frequency, cilia spacing, fluid properties, and other parameters can all be manipulated independently.

In this work, I present techniques for developing a biomimetic cilia model which is capable of replicating all of the key features of biological cilia. The biomimetic model consists of an array rod-like magnetic structures, which are similar in size to biological cilia
(Figure 1). They are flexible, and therefore may be actuated by an applied external magnetic field.

Aside from their role as biomimetic cilia, these structures are of interest in their own right as high-aspect-ratio flexible structures. High-aspect-ratio micro- and nano-structures have attracted increasing attention in the MEMS and microfluidics communities due to their potential applications as sensors and actuators, and the effect of their presence on the surface properties of a material, such as adhesion and wetting. High-aspect-ratio polymer rods have been produced with materials of high elastic moduli (on the order of 100 MPa), but these are generally unsuitable as actuating mechanisms due to their stiffness,
and microstructures made of softer materials such as polydimethylsiloxane have been reported to be unstable at aspect ratios as low as three to five.\textsuperscript{16,17}

We have been able to produce for the first time flexible, actuable templated microstructures with aspect ratios as high as 125. In addition, we have proven that actuated arrays of silia have the ability to pump and mix fluids at the microscale. In their passive state they may act as sensors of fluid flow or acoustic vibrations – an array of miniature microphones.

In the first chapter, “Materials,” I will describe the synthesis of the ferroelastomers which we have used in the production of these biomimetic arrays, including a novel material which I have produced specifically for this work. In the second chapter, “Fabrication,” I will describe the details of the procedures by which we produce nanorod arrays made of the ferroelastomer materials.

In addition to fabricating arrays of synthetic cilia, (‘silia’), it is essential that we are able to reliably and predictably actuate the silia arrays with applied magnetic fields. To this end, I have developed a model which describes the interaction of a silium with an external magnetic field, and this effort is described in the third chapter, “Modeling.” This model is the first of its kind in that it considers the effect of not only the magnetic field, but the magnetic field gradient as well, which we have found to be essential in understanding some of the modes of actuation of our silia. I have also produced some simple expressions which are helpful in designing high aspect ratio flexible magnetic structures for various purposes.

We have successfully actuated fields of silia in accordance with the model, and I will describe a couple interesting modes of actuation which we are able to produce experimentally in the fourth chapter, “Actuation.” This chapter will also describe the
potential of several field generating mechanisms, as well as a couple novel features of silia actuation which are predicted for the first time by my model.

Finally, we have been able to generate biologically relevant fluid flows with actuating silia arrays. These flows will be described in the fifth chapter, “Fluid Flow.”
2.1 Introduction

The ideal material for use in a magnetically actuable microstructure such as the silia described in this work must be both magnetic and flexible. In any homogenous material, these features are generally mutually exclusive. We can, however, combine the magnetic properties of a ferromagnetic metal with the flexibility of a polymer by carefully constructing a composite of the two materials: a ferroelastomer. A ferroelastomer is a composite of a magnetic material, such as magnetic nanoparticles, with an elastic polymer. (In discussing the properties of candidate ferroelastomers, I will assume a familiarity with magnetic quantities and magnetic materials; for background, the reader may refer to a general reference on magnetic theory.\textsuperscript{18,19})

There is a virtually unlimited number of polymers which could potentially be used in the construction of a ferroelastomer. The candidate material, however, should have the following properties:

- Low Young’s modulus, for flexibility
- Chemical compatibility with magnetic nanoparticles
- Chemical compatibility with liquid environment
- Ease of processing
In this chapter, I will discuss two main classes of polymer, each of which has its own set of advantages over the other: silicones and hydrogels. Silicones consist of a siloxane backbone with attached organic groups, and are generally hydrophobic. Their hydrophobicity prevents unwanted interaction with an aqueous environment, such as osmotic swelling, and allows the fabricated silia to be stable under a wide range of pH and ion concentration. Silicones are also widely available in pre-mixed kits, aiding in processibility. However, most magnetic nanoparticle suspensions are chemically incompatible with silicones. I will discuss strategies for overcoming this chemical incompatibility in this chapter.

Hydrogels are polymer matrices which are swollen with water, generally more than 90% by weight. They generally offer lower Young’s moduli than silicones, and are easily compatible with standard aqueous ferrofluids, enabling high magnetic loading with little effort. They may interact unpredictably with an aqueous environment, however, and processing is not as simple as with a silicone.

The available options for the magnetic component of the ferroelastomer are much more limited. In order to circumvent the high modulus associated with most magnetic materials, we are confined to using nanoparticle suspensions. A candidate nanoparticle should have the following properties:

- High saturation magnetization
- High magnetic permeability
- Ease of synthesis
Nanoparticles have been synthesized of nickel, cobalt, and iron (among a few more exotic alloys). Cobalt and iron nanoparticles offer very similar magnetic properties, while nickel particles have a saturation magnetization and permeability about an order of magnitude lower.\textsuperscript{19,20} Iron oxide nanoparticles are by far the most prevalent in the literature, due to their applications in magnetic recording media and the relatively simple fabrication methods available, and so all magnetic nanoparticles used in this work are iron oxide; namely magnetite (Fe\textsubscript{3}O\textsubscript{4}) or maghemite (\(\gamma\)-Fe\textsubscript{2}O\textsubscript{3}).

In general, the synthesis of a ferroelastomer proceeds by three steps. First, we make a suspension of magnetic nanoparticles – a ferrofluid – which will be compatible with the chosen polymer. Next, the polymer and ferrofluid are combined to form a composite material, and then the composite material is cured to form an elastic solid.

Ultimately, the suitability of a particular material for this application is determined by both its magnetic content and its modulus. Specifically, the ability of a silium to bend in an applied magnetic field is directly proportional to the quantity \(\frac{\mu_0 M^2 f^2}{E}\), where \(M\) is the magnetization of the magnetic material within the composite, \(f\) is the loading fraction of this material per unit volume, and \(E\) is the Young’s modulus of the composite. While the magnetization \(M\) is in general dependent on the magnitude of the applied field, for uniformity we will take \(M\) to be the value at saturation. This unitless ratio can be taken as a figure of merit for the material. The ratio is described in Chapter 4, “Modeling,” and will be referred to in this text as the magnetoelastic ratio. As a benchmark, a silium with an aspect ratio of 100 will bend through an angle of 10° with a magnetoelastic ratio of \(7 \times 10^{-5}\).
I present in this chapter the synthesis of three candidate materials: two silicone materials of differing magnetic loading and a hydrogel. These materials are presented in the table below with their respective magnetoelastic ratios.

Table 1: Summary of Candidate Materials

<table>
<thead>
<tr>
<th>Material</th>
<th>Young’s Modulus</th>
<th>Magnetic Loading</th>
<th>Magnetoelastic Ratio (x 10^5)</th>
</tr>
</thead>
<tbody>
<tr>
<td>FFPDMS-C16 (silicone)</td>
<td>2-3 MPa</td>
<td>1-4 %v.</td>
<td>0.7 - 16</td>
</tr>
<tr>
<td>FFPDMS-NH2 (silicone)</td>
<td>2-3 MPa</td>
<td>15-23 %v.</td>
<td>150 - 530</td>
</tr>
<tr>
<td>FFAAm (hydrogel)</td>
<td>0.1-1 MPa</td>
<td>10-18 %v.</td>
<td>200 - 6500</td>
</tr>
</tbody>
</table>

2.2 Ferrofluids

2.2.1 Introduction to Ferrofluids

In order to be actuable by an applied magnetic field, the nanorods must contain a large concentration of magnetic material, such as iron, cobalt, or nickel. Such metals, however, necessarily have very high Young’s moduli and are not easily bent by magnetic fields. A simple solution is to distribute magnetic material throughout the bulk of the rod in very small particles (much smaller than the rod diameter), in order to provide a high magnetization which is uniform on length scales on the order of the rod diameter. While distributing hard spheres of magnetic material throughout the matrix material does increase the modulus of the composite, reasonable concentrations of magnetic particles can be expected to increase the modulus by significantly less than an order of magnitude. Since biological cilia are on the order of 200 nm in diameter, it is necessary to have magnetic
particles which are at least an order of magnitude smaller, or less than 20 nm. Magnetic nanoparticles – which can be produced with sizes ranging from about 2 to 10 nm – fit the bill perfectly.

Magnetic nanoparticles also have the advantage of being paramagnetic in colloidal suspensions. While bulk iron and iron oxide are ferromagnetic by nature – that is, they maintain a permanent magnetization upon the removal of an applied magnetic field – iron oxide nanoparticles do not. Below a certain size (generally 20-30 nm), thermal energy causes a randomization of the orientation of the atomic dipole moments within each particle, resulting in an absence of a net dipole moment.\textsuperscript{19,22} The absence of a remanence field in the nanoparticle suspension obviates the need for any demagnetization apparatus, and the lack of magnetic history dependence simplifies both calculations and experimentation.

Magnetic nanoparticles were first synthesized by the coprecipitation of iron salts in the mid 1800’s by LeFort\textsuperscript{23}, to form magnetite. The first stable colloidal suspension of such particles was not achieved, however, until Elmore stabilized magnetite in an acidic medium with a surfactant in 1938.\textsuperscript{24} The same basic recipe that Elmore used – the coprecipitation of iron II chloride and iron III chloride with a base – has become the standard for the production of iron oxide nanoparticles, and has been refined by many other researchers for various means\textsuperscript{19,25,26}. Colloidal suspensions of magnetite nanoparticles have since been formed in both acidic and alkaline media\textsuperscript{27}, aqueous solutions of neutral pH\textsuperscript{28,29}, and in organic media\textsuperscript{29,30}. Magnetite particles have been further oxidized to form maghemite – which has a higher saturation magnetization than magnetite – and stable colloidal solutions of maghemite have been formed as well\textsuperscript{29,31}. For both magnetite and maghemite, solutions of very narrow size distribution have been produced\textsuperscript{32}, and the conditions for colloidal stability\textsuperscript{33,34} as well
as the magnetic properties of iron oxide nanoparticle suspensions are very well understood.

Although nickel and cobalt nanoparticles have also been synthesized by chemical means, iron oxide particles were used in this work due to their relative ease of production and their prevalence in the literature.

2.2.2 Making an Aqueous Ferrofluid

Following the work of Massart and van Ewijk, the iron oxide nanoparticles used in the material presented here were precipitated from ferric chloride (FeCl₃) and ferrous chloride (FeCl₂) salts in a 2:1 molar concentration. The methods for synthesizing this ferrofluid are detailed at the end of this chapter.

This ferrofluid has a pH of about 2 and contains a theoretical yield of 8.2 grams of maghemite (Fe₂O₃). The maghemite will form a stable suspension in water of up to 5% nanoparticles by weight; higher concentrations result in flocculation of the particles. Van Ewijk et al. have found maghemite nanoparticles produced by these methods to have an average size of 9 ± 0.4 nm. This material is only stable at a low pH and is limited in concentration. This is not ideal for use in many hydrogels, which are generally crosslinked under conditions of controlled pH. Also, the maximum concentration of only 5% wt. makes this material less than ideal for use without further concentration in hydrogels. However, this material has been incorporated in a silicone elastomer, PDMS (polydimethylsiloxane) to produce the majority of the devices which are described in this dissertation. This material, a composite of the maghemite ferrofluid and PDMS will be referred to as FFPDMS, and the
methods by which it is produced are described in the “Methods” section at the end of this chapter.

2.2.3 Stabilizing the Aqueous Ferrofluid with Citrate

In order to make a ferrofluid which is more appropriate for incorporation in hydrogels, the aqueous maghemite ferrofluid can be made stable at neutral pH with sodium citrate (Na$_3$C$_6$H$_5$O$_7$) and tetramethylammonium hydroxide (TMAOH, (CH$_3$)$_4$NOH)$^{29,30}$. A 1 M citric acid solution (0.2 g citric acid in 10 mL water) is added dropwise to the maghemite ferrofluid until the maghemite is completely flocculated. The solution will turn cloudy upon flocculation, and the maghemite can then be sedimented with a permanent magnet. After decanting the supernatant, a small amount of dry TMAOH is added to the sediment. The TMAOH dissolves quickly and will resuspend the flocculated particles. This citrate-stabilized ferrofluid, “citFF,” is stable at pH 7 and can be concentrated by evaporation. In this work, citFF solutions have been concentrated to as much as 50% wt. before becoming too solid to be workable. Even at these concentrations, they show no sign of flocculation. The citFF is ideal for use in a variety of hydrogels, as described in later in this chapter.

2.3 Silicone Composite Materials

2.3.1 Introduction to Silicones

A hydrophobic material such as a silicone polymer is ideal for application in an aqueous environment, since it ensures that the nanorod material will not interact unpredictably with the surrounding environment. Swelling of the polymer matrix and
leeching of nanoparticles, catalysts, or uncrosslinked polymer segments are all concerns when dealing with a water permeable matrix material. Swelling of the nanorod can unpredictably change the Young’s modulus of the material, affecting its response to the applied magnetic field. Leeching of particulates into the surrounding fluid may at best affect the chemical composition or pH of the water, and at worst significantly reduce the magnetic content of the nanorod, should the nanoparticles themselves leech out. Using a hydrophobic material such as a silicone eliminates any interaction of the surrounding water with the interior of the rod, preventing all of these problems. However, a hydrophobic material comes with its own problems, such as adhesion of the rods to one another due to their relatively high surface energy in water. Adhesion is discussed in detail in Chapter 3, “Fabrication.”

The simplest of the silicones is polydimethylsiloxane (PDMS), which consists of methyl groups attached to a siloxane backbone (Figure 2). Ferrofluid-PDMS composite materials have been described previously in the literature, with both cobalt\textsuperscript{37-39} and iron oxide\textsuperscript{40-42} nanoparticles. All have been produced by the same group (the Riffle group at Virginia Tech)\textsuperscript{43} and involve relatively complicated chemical synthesis. The materials presented in this chapter are synthesized by simpler means.
Presented in this chapter are two classes of ferrofluid-PDMS (FFPDMS) composite materials: the first is a simple mixture, which uses a surfactant to aid in mixing, and the second is a complexed material, in which nanoparticles are chemically bound to the PDMS. In general, the complexed materials have greater magnetic loading. However, there is still work to be done on the fabrication of rods made of complexed materials, and so all of the actuation and fluid flow experiments presented in this work are performed with the former class of FFPDMS composite.

The first of these mixed FFPDMS materials is a composite of maghemite nanoparticles mixed with a widely available PDMS, produced by Dow Corning: Sylgard 184. Efficient mixing in this composite material is encouraged by the addition of hexadecane as a surfactant, and will be referred to as FFPDMS-C16.
Although its exact composition is proprietary, Sylgard 184 consists of a vinyl-terminated PDMS and a filler material. It is available in a two-part kit, the second part of which contains a platinum catalyst and lower-molecular-weight vinyl-terminated PDMS. The fillers and the catalyst are formulated in such a way as to produce a product with desirable mechanical properties (in the absence of fillers, many crosslinked polydimethylsiloxanes become brittle) and predictable cure times, and so the Sylgard kit presented a convenient starting point for experimentation.

2.3.2 Preparation of FFPDM S-C16

Producing a composite material of the maghemite nanoparticles and Sylgard 184 PDMS, however, is not a simple matter of mixing the two together; the hydrophilicity of the maghemite prevents it from mixing well into the hydrophobic matrix of the PDMS. The nanoparticles must first be made stable in an organic solvent. Methods for doing so are described in the literature.\textsuperscript{29, 30} After suspending the nanoparticles in an organic solvent, they must be mixed in a stable suspension in the PDMS. The methods of successfully mixing the composite to produce the novel material FFPDMS-C16 are described in the “Methods” section at the end of this chapter.

While this material is relatively simple to make, it presents several difficulties that keep it from being an ideal material for making actuable nanorods in an aqueous environment. First, PDMS has a very high surface energy with water, resulting in a contact angle of about 100°. This property causes the thin, flexible PDMS nanorods to aggregate with each other when placed in an aqueous environment, in an effort to reduce total surface energy. This problem is described in detail in Chapter 3, “Fabrication.” Functionalizing the
surface of the nanorods after their fabrication might be a means to reduce their surface energy in water; however, PDMS is extremely difficult to functionalize due to the chemical inertness of the methyl groups, which line its backbone. Efforts to functionalize PDMS are detailed in Chapter 3.

Figure 3. Magnetization curves of pure maghemite nanoparticles and FFPDMS-C16, as measured by SQUID. The difference between magnetization per unit mass and magnetization per unit volume is a factor of density; here, we used the density of maghemite to determine the left axis. The left axis is therefore invalid for FFPDMS-C16.

Another difficulty that FFPDMS presents is that current processing procedures make it difficult to control the relative concentrations of PDMS, hexadecane, and maghemite in the finished composite material. Samples of FFPDMS produced by the methods described here have topped out at about 18% maghemite loading by wt. (4% v.), as measured by SQUID on bulk samples of FFPDMS-C16 (Figure 3), and leave little opportunity for adjustment.
Measurements of maghemite loading via fits to the model described in Chapter 4 indicate an even lower loading of about 7\% wt. (1.4\% v.) Combined with a relatively high modulus of 2.6 MPa, this material has a magnetoelastic ratio of less than 1 x 10^{-4}, which is low compared to other materials (See Table 1.)

For these reasons, other materials may be more attractive for use in the actuable nanorods. For example, PDMS terminated with functional groups would provide a chemical handle for functionalization of the nanorod surface after fabrication, as well as allow for fundamentally different methods of producing the nanoparticle-polymer composite, resulting in more controllable materials with higher concentrations of magnetic components.

2.3.3 Silicone-Nanoparticle Complexes

2.3.3.1 FFPDMS-COOH

One such functionalized PDMS has been used by Riffle\textsuperscript{40-42} to produce a magnetite-PDMS composite material with as high as 60\% wt. (23\% v.) particle loading. This group has synthesized a tri-vinyl functional PDMS, which is then reacted with mercaptoacetic acid to produce a PDMS terminated with three carboxyl groups. The carboxyl groups are then adsorbed onto the surface of magnetite nanoparticles to form a complex.
This reaction proceeds in a two-phase solution; one phase is aqueous and contains the magnetite nanoparticles, and the other phase is organic and contains the functionalized PDMS. Lowering the pH of the aqueous phase floods the solution with $\text{H}^+$ ions, which are adsorbed onto the surface of the magnetite nanoparticles and give them a positive surface charge\(^{44}\) (Figure 4). (While this plot is for maghemite rather than magnetite, magnetite behaves similarly and has a point of zero charge at a pH of 6.55)\(^{45}\) At the same time, many of the COOH groups in the PDMS phase become dissociated when in contact with the water phase, becoming COO\(^{-}\). The pKa of a carboxyl group is around 2-3, so for pH above this value, a significant fraction of the carboxy groups remain dissociated. By adjusting the pH of the aqueous phase to be greater than the pKa of the carboxyl and less than the isoelectric point of the magnetite, one can ensure that there are both negatively charged carboxyl groups
in the organic phase and positive surface charge on the magnetite. When these conditions occur, the carboxy-terminated PDMS bonds adsorbs onto the magnetite, and the magnetite is transferred into the organic phase, forming a complex (Figure 5). Vigorous mixing increases the surface area between the two phases, speeding the reaction.

![Figure 5. Schematic representation of the complexation of magnetite nanoparticles with a carboxyl-terminated PDMS. As the pH is lowered to 3, the nanoparticles take on a positive surface charge. Since pH 3 is above the pKa of the carboxyl, many of the carboxyl groups remain dissociated and therefore negatively charged. Over time, the positively charged magnetite is attracted to the negatively charged carboxyl and transfers to the organic phase.](image)

The complex produced by this procedure has a magnetoelastic ratio of as much as $4 \times 10^{-3}$, which makes it a good candidate for use in our biomimetic system. However, it
requires lengthy chemical synthesis which makes it a cumbersome material for those not
directly involved in the chemical sciences. For this reason I have developed a similar
material which can be made with easily with commercially available materials and has
similar material properties and magnetoelastic ratio. This material uses amines as the
adsorbing agent, and is therefore labeled FFPDMS-NH2.

2.3.3.2 FFPDMS-NH2

While it is difficult to find heterobifunctional PDMS – such as that synthesized by the
Riffle group – commercially, PDMS copolymers are much more common. If chosen
carefully, they can provide the same single-ended linkage as a heterobifunctional polymer.
(Note that a homobifunctional polymer is likely to link to multiple nanoparticles, potentially
crosslinking the complex prematurely. They are therefore unsuitable for this application.) In
this case, the polymer I used is a copolymer of aminopropylmethysiloxane (APMS) with
dimethylsiloxane (DMS), containing 6-7 percent APMS mole % (Gelest, AMS-161 figure).
For simplicity, I will refer to this material as PDMS-NH2. The reaction proceeds similar to
that described for FFPDMS-COOH, except that the magnetite transitions to the organic phase
at high pH rather than low.

The pKa of a primary amine is about 10. At pH less than its pKa, there is a
significant concentration of protonated amines, \( \text{NH}_3^+ \). Additionally, at pH greater than the
isoelectric point of magnetite, the magnetite adsorbs \( \text{OH}^- \) ions and carries a negative surface
charge. By tuning the pH to be greater than the isoelectric point of magnetite (pH 6.8) and
less than the pKa of the secondary amines (pH 10), conditions can be created which will
cause the amine functionality of the PDMS to bond to the magnetite particle, transferring the
magnetite from the aqueous phase to the organic phase and producing a PDMS-magnetite complex. A detailed description of the synthesis and crosslinking of FFPDMS-NH2 is included in the “Methods” section at the end of this chapter.

This is the first time that this material – a composite of PDMS bonded to magnetite nanoparticles via an amine functionality – has been produced.

2.3.3.3 FFPVMS-NH2

As I will discuss in the following chapter, it may become imperative in the future to functionalize the surface of our FFPDMS silia. However, polydimethylsiloxane (PDMS) itself is very difficult to functionalize, due to its abundance of relatively unreactive methyl groups, and it may be convenient to have a material which provides other chemical handles for functionalization. Polyvinylmethylsiloxane (PVMS) has mechanical properties similar to PDMS, but offers multiple mechanisms for functionalization through the vinyl groups along the siloxane backbone. A chemical diagram of PVMS is shown in Figure 2. In the “Methods” section at the end of this chapter, I present a method of producing PVMS complexed with magnetite nanoparticles, which will proceed by affixing an amine functionality to a fraction of the vinyl groups; the amines may then be adsorbed onto the nanoparticle surface in a manner identical to that for FFPDMS-NH2. This material is called PVMS-NH2.

One method of functionalization is to react the vinyl groups with a thiol, to form sulfur bonds. This reaction happens at room temperature and without any catalyst. In order to prepare the PVMS for complexation with magnetite nanoparticles, the PVMS was functionalized with an amine, via thiol-ene addition of mercaptoethylamine (MEA,
SHCH₂CH₂NH₂). This process is recorded in detail in the “Methods” section at the end of this chapter.

2.4 Hydrogel Composite Materials

2.4.1 Introduction to Hydrogels

Besides the silicone elastomers, another likely candidate material for the fabrication of flexible magnetic rods is a hydrogel. Hydrogels are, by definition, any polymeric material which is swollen to a great degree with water (generally > 90% water) to achieve a gelatinous state, without dissolving. They may be crosslinked, such as in an acrylamide gel or most polyvinyl alcohol hydrogels or uncrosslinked, as in agarose. While hydrogels are susceptible to osmotic effects while in an aqueous environment and are therefore likely to swell or shrink unpredictably, they offer several advantages over silicon elastomers which make them attractive. Most intriguing is their affinity for water, which gives them an extremely low surface energy in an aqueous medium. Contact angle measurements of water performed on hydrogels have shown contact angles indistinguishable from zero degrees. This property has the potential to significantly reduce the adhesion issues prevalent in silicone rods, possibly eliminating the need to further functionalize the surface of the rods. In addition, water-based ferrofluids such as the citrate-stabilized maghemite ferrofluid described earlier in this chapter mix readily into many hydrogels, simplifying synthesis and allowing for, in some cases, very high concentrations of magnetic material. Finally, most hydrogels have very low elastic moduli (compared to silicones), making very flexible nanorods. In most cases, the modulus is easily tuned simply by adjusting the ratio of polymer to water.
A significant amount of work has been done on magnetic nanoparticle–hydrogel composites, or ‘ferrogels.’ Generally, the hydrogels of choice are polyvinyl alcohol or polyacrylamide, and iron oxide nanoparticles are either mixed with the gel prior to crosslinking or precipitated directly within the gel network.

2.4.2 FFPAAm

One of the most promising hydrogels for application in flexible, magnetic nanorods is polyacrylamide (PAAm). Acrylamide is commonly used in gel electrophoresis, and so details regarding its synthesis are prevalent in the literature. In addition, examples of ferrofluid-polyacrylamide gels (FFPAAm) are also available in the literature – researchers have made magnetite-loaded acrylamide hydrogels with magnetite concentrations as high as 18% v. (50% wt.), and moduli as low as $10^4$ Pa. In the fabrication of the acrylamide hydrogel, the maghemite ferrofluid is first mixed in an aqueous solution of the acrylamide monomer. The very low viscosity and absence of long-chain polymers at this stage allows the ferrofluid to be mixed easily in very high concentrations; in fact, the presence of the monomer had no effect on the maximum concentration of maghemite which could be attained. As with the citrate stabilized ferrofluid itself, the monomer-ferrofluid solution would solidify beyond workability before showing any signs of flocculation. After mixing, polymerization and crosslinking are performed simultaneously, forming a gel in which the maghemite particles are trapped.

A typical recipe for an acrylamide gel includes the acrylamide monomer (AM), a crosslinker (N, N methylene bisacrylamide, or BA), a catalyst (ammonium persulfate, APS), and an initiator (tetramethylethylenediamine, TEMED). Polymerization and crosslinking
necessarily occur simultaneously, as one side of the symmetric crosslinker is polymerized within each of two different chains of the polymerizing acrylamide, forming crosslinks. The APS catalyzes the polymerization of the monomer, and the TEMED speeds the breakdown of APS, allowing the reaction to occur within a reasonable amount of time at room temperature. The free radical breakdown products of the APS, which react with the double bond in the acrylamide monomer to promote polymerization, will also react with atmospheric oxygen, so the reaction is carried out in an oxygen-free environment. A detailed recipe for producing FFAAm is in the “Materials” section at the end of this chapter.

Using this method, we have been able to make FFAAm gels with as much as 50% wt. magnetic loading (18% v.). Higher magnetic loading seems to be possible without flocculation of the ferrofluid, but the uncrosslinked FFAAm begins to take on a pasty consistency at beyond 50% wt. loading, indicating that it would be difficult to work with a higher-concentration ferrogel. At 50% wt. loading, these gels may have magnetoelastic ratios as high as about 0.1, making them ideal candidates for silica. Techniques for producing microrods of this FFAAm material will be presented in the following chapter.

2.5 Conclusion

In this chapter, I have described the materials available for producing magnetic, flexible structures, and have selected candidate materials based on a figure of merit, the magnetoelastic ratio, which is presented here for the first time. I have also described the synthesis of these candidate materials, including two novel FFPDMS materials which will be of use to the scientific community. In the following chapter, “Fabrication,” I will describe in
detail the processes by which we are able to generate arrays of high-aspect-ratio microstructures – silia – from these materials.

2.6 Methods

2.6.1 Preparation of an Aqueous Ferrofluid

In a typical sample, 3.97 grams of iron(II) chloride tetrahydrate and 6.49 grams of iron (III) chloride, anhydrous, were added to 500 mL of deionized water and mixed thoroughly with a magnetic stir bar. The salts were precipitated with 40 mL of concentrated ammonium hydroxide (NH₄OH, 25% in water) while stirring vigorously. The formation of the nanoparticles is manifested in a dramatic change in color from the clear orange of the iron salt mixture to a dark, opaque black. The flocculated nanoparticles can be sedimented with a permanent magnet, and the supernatant decanted. The theoretical yield of this recipe is 4.7 grams of magnetite (Fe₃O₄). Massart has found magnetite nanoparticles produced by these methods to have a diameter of 7 ± 0.4 nm.

The particles in this state are flocculated and will not form a stable suspension in water. In order to make a stable aqueous suspension, the particles are further oxidized to form maghemite by heating in an acidic solution in the presence of iron nitrate (Fe(NO₃)₃), following the methods of vanEwijk. Generally, 80 mL of 1 M nitric acid (7.5 mL 68% aqueous HNO₃ diluted to 80 mL with deionized water) is added to the sedimented magnetite and stirred for 5 minutes. 120 mL of 0.35 M iron nitrate (17 g Fe(NO₃)₃ + 9 H₂O diluted to 120 mL with deionized water) is then added, and the mixture is stirred at boiling for 30 minutes. After boiling, the maghemite nanoparticles can be sedimented with a magnet and
the supernatant decanted. The sediment is rinsed with an additional 80 mL of 1 M nitric acid, sedimented and decanted. The resulting sludge will form a stable suspension of magnetic nanoparticles – a ferrofluid – upon the addition of deionized water.

2.6.2 Preparation of FFPDM S-C16

Following the method of vanEwijk,29 1 mL of the aqueous maghemite ferrofluid (at 5% wt. maghemite) (before stabilization with sodium citrate) is diluted into 20 mL deionized water. Ammonium hydroxide is added dropwise until the solution becomes completely flocculated (as described earlier, the solution will turn orange and cloudy upon flocculation.) The flocculated maghemite can be sedimented with a magnet, and the supernatant decanted. The sediment is resuspended in 20 mL deionized water (it will not form a stable suspension, but remain flocculated), and a couple of drops of oleic acid are added to the solution. Upon gentle mechanical stirring, the maghemite will transfer from the water phase into the oil phase, leaving very dark droplets of oil in a clear, aqueous background. The oil phase is separated by sedimentation with a magnet and rinsed twice with ethanol to remove excess oleic acid. The maghemite may then be resuspended to form a stable ferrofluid in an organic solvent, such as toluene or cyclohexane.

Hexadecane is then added in a 1:1 v/v ratio to the oleic acid nanoparticle suspension and is further diluted in toluene (5:1 v/v toluene to product.) The Sylgard 184 is also diluted 5:1 by volume with toluene. The two solutions are then combined over the course of several minutes under ultrasonication by a Fisher Scientific Sonic Dismembrator 550 operating at 150 W. The toluene is removed by evaporation, leaving behind the less-volatile hexadecane, which stabilizes the maghemite particles within the PDMS matrix. Upon resting for several
days, excess hexadecane and PDMS separates from the composite, leaving behind a stable suspension of maghemite nanoparticles in a PDMS-hexadecane matrix. This ferrofluid-PDMS composite material will be henceforth referred to as “FFPDMS-C16.”

2.6.3 Preparation of FFPDMS-NH2

In order to synthesize FFPDMS-NH2, magnetite is first produced in a manner similar to that described earlier in this chapter: 1.2 grams of ferric chloride (FeCl$_3$), and 0.74 grams of ferrous chloride tetrahydrate (FeCl$_2$ + 4 H$_2$O) are each dissolved in 20 mL deionized water, and then mixed together in a beaker with a magnetic stir bar. After stirring for several minutes, 20 mL of concentrated (29%) ammonium hydroxide (NH$_4$OH) is added, while stirring vigorously, to precipitate the salts and form nanoparticles. The organic phase is prepared by mixing 2 grams of PDMS-NH2 in 60 mL of dichloromethane (DCM). The phases are combined and stirred, and the pH of the solution at this point is about 10. The mixture is stirred for about 8 hours while maintaining the volume of dichloromethane, and then overnight in order to allow the reaction to complete and to evaporate the dichloromethane.

After stirring overnight, the mixture separates upon resting into two phases: a dark aqueous phase, which still contains some magnetite, and a dark PDMS phase, which ranges from thick and gummy to thin and oily depending on the molecular weight of the PDMS used. The PDMS phase is separated from the mixture and rinsed extensively with water to remove salts and with methanol to remove uncomplexed PDMS (five times each, sedimenting with a magnet to decant.) Finally, the material is diluted in chloroform, ultrasonicated for several minutes, and placed on a magnet to sediment overnight. Any
uncomplexed magnetite will fall out of solution, and the supernatant is recovered and dried of solvent. This final product is a complex of PDMS and magnetite, and is referred to as “FFPDMS-NH2.”

The resulting material has a thick, pasty consistency. It is still malleable enough for use in silia fabrication (described in the next chapter), but may be diluted with excess PDMS-NH2 without any loss of homogeneity, should a liquid be preferred.

This material lacks the terminal vinyl groups of Sylgard 184 PDMS, and therefore must be crosslinked by a peroxide-based cure. A peroxide cure proceeds by the formation of free radicals upon the breakdown of a peroxide, generally due to heat. The free radicals then cause a number of functional groups to bind, including the amines present in FFPDMS-NH2. In this case, the FFPDMS-NH2 is diluted by a factor of 100 in chloroform or dichloromethane, and 2% dicumylperoxide (wt. %, relative to FFPDMS-NH2) is added to the solution and mixed thoroughly with ultrasonication. The mixture is then dried of solvent in a rotovapor. The mixture will be stable at room temperature for several months, and will cure in about 1 hour at 200º C.

2.6.4 Preparation of FFPVMS-NH2

The reaction of the thiol functionality of the MEA with the vinyl group of the PVMS closely followed the work of Wilson et al.40:

First, a solution of 0.4 grams MEA, 1 gram PVMS, and 0.2 grams azobisisobutyronitrile (AIBN) in 50 mL of toluene was prepared. This provides about 0.4 equiv. MEA to vinyl, or enough MEA to functionalize 40% of the available vinyl groups. The free radical initiator, AIBN, is supplied in a 0.1 equiv. concentration to vinyl groups.
The mixture was placed in a flask equipped with a magnetic stir bar and a condenser. The solution was bubbled with dry nitrogen and heated to 70º C in an oil bath, and allowed to react over the course of 40 hours (Figure 6).

![Reaction mechanism](image)

**Figure 6.** Addition of mercaptoethylamine to poly(vinylmethyl siloxane).

Upon completion of the reaction, the mixture was allowed to rest for several hours, and a small amount of light-colored particulates was allowed to sediment. A small amount of the supernatant was removed to an 18 mm square glass coverslip and allowed to dry, leaving a thin film of reacted PVMS on the coverslip. Similarly, a small amount of fresh, unreacted PVMS was diluted in toluene, placed on a coverslip, and allowed to dry. A small amount of water was placed on each of the thin films, and contact angles were observed. The reacted PVMS showed a dramatically smaller contact angle with water than the unreacted PVMS, indicating an abundance of hydrophilic amines.
2.6.5 Preparation of FFPAAm

Performing a procedure similar to that described by Galicia, synthesis of a representative sample of a ferrofluid-acrylamide material (FFPAAm) which contains 6% acrylamide in water is as follows:

The initiator, APS, is mixed 10% by weight into deionized water (0.2 grams APS into 2 mL H$_2$O), and set aside. If not used immediately, this solution should be stored at 4º C. The monomer, AM, and crosslinker, BA, are then mixed in a ratio of 20 monomers per crosslinker, in a 20% wt. solution in deionized water (2.22 grams of AM and 0.11 grams of BA into 10 mL H$_2$O). This solution is diluted with citrate-stabilized ferrofluid (citFF) of any maghemite concentration to produce a maghemite-AAm solution of 6% wt. polymer (0.54 mL AM/BA mixture to 1.46 mL citFF). The solution is degassed by bubbling argon through it for about 10 minutes, and then 40 microliters of the 10% APS solution are added. Finally, 4 microliters of TEMED are added, the solution is mixed quickly and allowed to cure in an argon-purged environment. Polymerization occurs within a couple of minutes. The acrylamide monomer is extremely toxic and should be handled with care. The toxic effects are eliminated upon polymerization, but unpolymerized monomer may remain in small quantities in a crosslinked gel. A finished gel may be rinsed by soaking in water to eliminate unreacted monomers.
Chapter III

FABRICATION

3.1 Introduction

Having produced several materials which are good candidates for the production of flexible magnetic biomimetic cilia, it is necessary to devise a method for molding those materials into particular geometries of interest. In this chapter, I will describe the templates which we have chosen as a mold for our silia arrays, as well as the fabrication steps which allow us to produce a viable sample which can be simultaneously observed via optical microscopy and actuated with an external magnetic field.

One major challenge in fabricating successful silia arrays has proven to be lateral collapse of the silia due to adhesion with one another. I will discuss this issue in both silicone and hydrogel silia samples, and present strategies for overcoming adhesion in each.

3.2 Choosing a Template

Biological cilia range in length from about 3 to 10 microns and are about 250 nm in diameter. They are found in various densities in the body, ranging from a low of about 2 x 10^6 cilia/cm^2 in the embryonic nodal plate to as high as 1 x 10^8 cilia/cm^2 in airway epithelial tissue. In order to produce nanorods in these geometries, it is necessary to have a template which is tunable over a wide range of density, able to produce nanorods of various lengths, and releases easily from the fragile finished product.
There are several methods available for producing high-aspect ratio structures on the nanoscale. Most common in the literature is photolithography, which is used to pattern anything from nanoscale circuitry to microscale machinery. However, while photolithographic methods are capable of producing structures of very high aspect ratio in a plane parallel to the substrate (length $\gg$ width), they are not well-suited to producing high-aspect-ratio structures in a plane perpendicular to the substrate (depth $\gg$ width). Furthermore, with soft materials such as silicones and hydrogels, photographic lift-off procedures may lead to structure collapse.\textsuperscript{16,17}

Another common method for producing high-aspect-ratio nanostructures uses anodized aluminum oxide (AAO) as a template.\textsuperscript{55,56} AAO has the advantage of being cheap enough to be disposable, and so it can be gently removed from the molded structures by dissolution. In addition, AAO membranes can be grown to a wide variety of thicknesses, allowing for the production of nanorods of various length. However, AAO imposes severe limits on the diameter and spacing of the pores, and so is unsuitable as a template for biomimetic cilia.

Track-etched membranes have also been used in the synthesis of nanostructures\textsuperscript{57,58} – most notably metallic nanowires\textsuperscript{59,60} – and they present a third possibility for a template for the production of biomimetic cilia. These membranes generally consist of a very thin layer of polymer (most commonly polycarbonate) which is exposed to high-energy ions from a nuclear source or ion beam. As the ions travel through the polymer, they leave behind a track of broken bonds (about 10 nm diameter) surrounded by a halo of additional damage (10 – 100 nm).\textsuperscript{61} The particle tracks etch preferentially in a solution of sodium hydroxide – at a rate as high as 1000 times the bulk etch rate, albeit only through the radius of the halo\textsuperscript{61} – to
produce pores of a uniform and tunable diameter. Polycarbonate track etched (PCTE) membranes are available in a variety of thicknesses and pore densities, allowing for the production of nanorods of different length over a wide range of density. They are also soluble in organic solvents such as dichloromethane (DCM) and N-methylpyrrolidone (NMP), among others, allowing for the gentle removal of the membrane from the molded structures. As such, PCTE membranes make a very convenient template for biomimetic cilia, and all of the templated nanorods presented in this work are templated in these membranes.

PCTE membranes are available commercially for water filtration applications with pore diameters ranging from 10 nm to 15 microns. However, since they are usually intended for use as filters, pore density is generally a function of pore diameter (in order for membranes of different pore size to function at similar flow rates) and rarely specified separately. Also, a primary concern of a filter membrane is to have no pores larger than those specified. Since the techniques used to produce the particle tracks produce a random distribution of tracks across the surface of a membrane, manufacturers vary the angle of entry of the particles as a function of time to prevent two parallel tracks from overlapping. This reduces the possibility of producing pores larger than specifications, but results in a less-than-ideal template for biomimetic cilia. At best, the resulting nanorods are tilted through a range of angles ranging to as high as $45^\circ$ from the vertical; at worst, intersecting particle tracks result in a network of interlocked and immobile nanorods. In order to avoid these difficulties, the membranes used in this work were ordered to specification from a company which produces track-etched membranes for research purposes. The pores are parallel to
each other and can be specified to be produced at any angle to the membrane surface. In addition, the pore size and pore density can be specified independently.

3.2.1 Etching the Membrane

The membranes are purchased from it4ip with a minimal pore diameter of 200 nm, and in a variety of thicknesses and pore densities. The pores can then be etched to any desired size using a 4M solution of sodium hydroxide (NaOH). The membranes are submerged in the 4M NaOH and placed in a convection oven set to 80\( ^\circ \) C. Figure 7 demonstrates the relationship between etch time and pore diameter under these conditions.

![Polycarbonate Track Etched Membrane Etched with 4 M NaOH](image)

**Figure 7.** Polycarbonate membrane pore diameter as a function of etch time in 4 M sodium hydroxide. All membranes began with 200 nm pore diameters (nominal), and all etching took place at 80\( ^\circ \) C. The line is a linear least-squares fit to the data.
3.3 Design Considerations

Once fabricated, the rod arrays will be observed under optical microscopy and actuated with external magnetic fields. For high-magnification optical microscopy to be feasible, it is necessary that the silia array be mounted on a glass coverslip and be close enough to the coverslip surface to be within the working distance of a 100x objective, generally a few hundred microns. In order for the array to be effectively actuated with an external magnetic field, it must be possible to position the magnetic system as close as possible to the silia as well, without blocking the light path of the microscope. Figure 8 depicts a geometry which accomplishes these goals.
Figure 8. Schematic representation of the setup for actuation of magnetic rod arrays. Imaging is done in reflection to allow the magnet to be placed as close as possible to the sample (this position would otherwise block the light path.) Reflection imaging is made possible by the reflective gold coating on the lower coverslip.

The silia are imaged in reflection in order to allow the insertion of a magnet (or any magnetic actuation system) immediately below the sample without affecting the light path. A thin layer of gold, thermally evaporated onto a glass coverslip, provides a reflective surface. The sample must be entirely enclosed in order to prevent the evaporation of the liquid in which the rods are submerged, and so it is sandwiched between two #0 coverslips. In order to be as close as possible to both the objective from above and the magnetics system from below, the sample must be as thin as possible. Current procedures produce samples
which are less than 500 microns in total thickness. A typical view of silia in this geometry is shown in Figure 9, below.

It is also essential that the silia array be firmly anchored to the glass surface. This is accomplished in various ways for different materials; these techniques are described in the appropriate sections below.

Figure 9. A silia array as viewed via optical microscopy from above, in the geometry shown in the previous figure. This particular array consists of 2 million silia per square centimeter, and each silium is 20 microns long and 800 nm in diameter. They are fabricated of FFPDMS-C16.
3.4 Fabrication of Silicone Silia Arrays

While dry silicone elastomers readily wet and adhere to a glass surface, submersion in a liquid will cause them to quickly release. Adhesives would be useful in fixing the rod arrays to the glass substrate, but very few adhesives are able to withstand the solvents which are used to dissolve the PCTE membrane, making it is necessary to devise an alternative means of attaching the silicone rod array to the substrate. Fortunately, this can be accomplished by activating the surfaces of both the silicone and the glass with an oxygen plasma, and then allowing them to contact and form a permanent, covalent chemical bond.66

Two things are necessary to activate the surface of the silicone: energy and monatomic oxygen. In an oxygen plasma, the high energy oxygen collides with the methyl groups attached to the surface, breaking them free. The oxygen then binds to the surface in its place, forming a hydroxyl group. The glass is functionalized with hydroxyl in a similar manner. When the two materials come in contact with each other, the hydroxyls react via a condensation reaction to produce a covalent bond (Figure 10).
Figure 10. Covalent bonding of plasma-treated PDMS and glass. A) The PDMS and glass in their native state. B) An oxygen plasma bonds OH groups to both the glass and PDMS. C) The OH groups bond via a condensation reaction, binding the PDMS and glass monolithically.

In the fabrication process described below, a thin PDMS ring is first permanently attached to an 18 mm square coverslip by plasma treatment, to form a small well. Later, the PDMS which forms the substrate of the nanorod array will cure monolithically with this well, firmly anchoring the rods to the glass.

Once the PDMS well is in place, a small piece of membrane is cut to an appropriate size. The membrane pores are impregnated with FFPDMS by immersion (Figure 11A), and the outer surfaces of the membrane are gently cleaned of excess FFPDMS. The impregnated membrane is then submerged in a small drop of uncured Sylgard 184 PDMS mixed 10:1 with curing agent and placed directly on a #1.5 glass coverslip in the center of the ~100 µm thick
PDMS well that has been permanently sealed to the glass as described above (Figure 11B). The sample is then placed in a convection oven at 80 °C for several hours to cure.

Figure 11. A cross-sectional view of the fabrication steps used to template magnetically actuated composite polymer rod arrays. Details in the text.

Upon removal from the oven the upper layer of encapsulating PDMS is carefully peeled away to expose the polycarbonate membrane (Figure 11C). Once exposed, the membrane is dissolved in a 70 °C bath of dichloromethane (DCM) for about ten minutes. The sample is then carefully transferred to another bath of clean DCM for rinsing. The thickness of the PDMS well maintains a layer of DCM around the rods, preventing damage due to drying or excessive flow. Since the sample is intended for an aqueous environment and dichloromethane is immiscible with water, the sample is next transferred to an ethanol
bath with 1% Triton-X 100 to replace the DCM. (The Triton-X 100 acts as a surfactant; it is described in detail in section 3.5.1.) The DCM is significantly more dense than ethanol, however, and will remain in the PDMS well; it must be poured out carefully while the sample remains submerged in the ethanol bath. Finally, the sample is transferred to a second ethanol/Triton X-100 bath to eliminate traces of dichloromethane.

At this point, the ethanol can be exchanged for water by similar means. When the sample is removed from its final bath, a #0 coverslip is placed on top of the PDMS spacer to prevent evaporation (Figure 11D). The coverslip can be sealed to the PDMS well with Norland Optical adhesive to provide a more durable moisture barrier.

Using this method, rods have been produced which range in size from that of biological cilia (~200 nm diameter and ~10 micron length) to much larger (1 micron diameter and 25 micron length), and with aspect ratios as high as 125 (200 nm diameter and 25 micron length). These structures are unparalleled in aspect ratio for a soft polymeric material – similar structures fabricated by other groups have aspect ratios of only three to five and, in fact, these groups predict collapse for structures of higher aspect ratio.\textsuperscript{16, 67} A scanning electron micrograph of a large array of silia fabricated by these methods is shown in Figure 12, below.
Figure 12. A scanning electron micrograph of an array of FFPDMS-C16 silia produced by the methods detailed in this chapter. These silia are about 20 microns tall by 1 micron in diameter.

3.4.1 Process Modifications for FFPDMS-NH2

Because FFPDMS-NH2 must be crosslinked by a peroxide cure, rather than the platinum-catalyzed cure used in Sylgard 184 PDMS, fabrication processes vary slightly from those for FFPDMS-C16 silia. As discussed in Chapter 2, a peroxide cure proceeds via the formation of free radicals upon the breakdown of the peroxide. This happens at significantly higher temperatures than the platinum-catalyzed cure, and may be hindered by reactions with atmospheric oxygen. For these reasons, I have found that a two-step curing procedure works best for peroxide cured silia: they are assembled just like the FFPDMS-C16 silia described in the previous section, but are placed in an oven at 80º C for one hour and then transferred to
a second nitrogen flushed oven at 200 °C for an hour. The 80°C oven crosslinks the Sylgard 184 substrate via a conventional platinum cure, and the 200°C oven crosslinks the FFPDMS-NH2 via a peroxide cure. In addition, a second piece of polycarbonate membrane, without pores, is placed on top of the sample after the first curing step in order to exclude atmospheric oxygen from the PDMS during the second cure, as shown in Figure 13 below.

![Figure 13. Sample geometry for peroxide-cured FFPDMS. Here, the second membrane has been placed on top of the sample to exclude atmospheric oxygen during the 200°C cure. The hash marks above the porous membrane indicate the portion of the platinum catalyzed PDMS which remains uncured during the 80°C cure.](image)

After the first curing step, the thin layer of Sylgard 184 PDMS which lies on top of the membrane remains uncured (hash marks in Figure 13). We suspect this is due to interference from peroxide diffusing into this material from the FFPDMS-NH2 in the pores of the membrane. This layer cures later in the 200°C cure, provided oxygen is excluded from the layer with the second membrane and nitrogen-purged oven.

We have found that covering the sample with the second membrane prior to the first curing step results in no improvement in the final product – it seems that atmospheric oxygen
does not begin to react with the peroxide until it is placed in the higher temperature oven. In addition, applying the second membrane prematurely makes it much more difficult to remove later, as it tends to settle into the uncured Sylgard 184.

After the sample is fully cured, the upper membrane is removed, and the underlying layer of PDMS is peeled carefully from the FFPDMS-NH2-impregnated membrane. In these peroxide-cured samples, this often leaves a residual film of PDMS on top of the membrane; this may be removed with a PDMS etchant, such as tetrabutyl ammonium fluoride (TBAF) mixed 1:3 in N-methyl pyrrolidone (NMP). The etchant is placed on the sample for about one minute, after which the surface is rinsed with NMP and then isopropanol. Samples prepared with and without this etching procedure are shown in Figure 14 below.

Figure 14. FFPDMS-NH2 silia arrays prepared by the procedure outlined above. The sample in A was not etched with TBAF, and the residual PDMS film on top of the rods is clearly visible. The sample in B has been etched, and shows a remarkable reduction in the presence of this layer. Etching procedures are still being optimized.
3.5 Adhesion Issues in Silicone Silia

One difficulty in fabricating successful PDMS silia arrays is the high surface energy of the hydrophobic PDMS in water – water has a contact angle of about 95° on bulk PDMS. The silia have a much lower surface energy when in contact with each other, and so they are likely to aggregate in an aqueous environment.

Hui et al. and Roca-Cusachs et al. have developed models for lateral and ground collapse of high-aspect-ratio structures, respectively, based on energy considerations.\textsuperscript{16, 67} In short, the decrease in surface energy gained from a structure being in contact with another structure (lateral collapse) or the substrate (ground collapse) is balanced with the increase in elastic energy that such a movement would entail. The conditions become, for lateral collapse:

$$\frac{L}{d} = \left( \frac{\pi^4 3^3}{2^{11/3} (1 - \nu^2)} \right)^{1/12} \sqrt[4]{\frac{s}{d}} \left( \frac{E}{W} \right)^{1/3} (d)^{1/3}$$  \hspace{1cm} (1)

and ground collapse:

$$\frac{L}{d} = \frac{\pi^{5/3}}{2^{11/3} 3^{1/2}} (1 - \nu^2)^{-1/6} \left( \frac{E}{W} \right)^{2/3} (d)^{2/3}$$  \hspace{1cm} (2)

where $L$ is the height of the structure, $d$ is the diameter, $\nu$ is the Poisson ratio of the material, $s$ is the lateral distance between structures, $E$ is the Young’s modulus, and $W$ is the work of adhesion of the material to itself. Figure 15 illustrates the two types of collapse and the geometrical quantities under consideration.
Figure 15. Illustration of modes of collapse in high-aspect-ratio microstructures. A) uncollapsed structures. B) Structures exhibiting lateral collapse. C) Structure exhibiting ground collapse.

Since lateral collapse is the limiting condition in our samples (over ground collapse), I have calculated the maximum aspect ratios which are predicted to be stable for our materials. We will assume a Young’s modulus of 2.6 MPa for Sylgard 184 PDMS, and the following values for the work of adhesion of PDMS to itself in various media, given by Roca-Cusachs. I have listed values for the lateral distance between structures, s, based on the extremes of biologically-relevant densities: about $10^6$ per cm$^2$ for nodal cilia and $10^8$ per cm$^2$ for airway epithelial cilia.
Table 2: Critical Aspect Ratios of PDMS Structures

<table>
<thead>
<tr>
<th>Medium</th>
<th>Work of Adhesion</th>
<th>Critical Aspect Ratio</th>
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<tr>
<td></td>
<td></td>
<td>$10^6$ silia/cm²</td>
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<tr>
<td></td>
<td></td>
<td>(s ~ 10 µm)</td>
</tr>
<tr>
<td>Air</td>
<td>44 mN/m</td>
<td>13</td>
</tr>
<tr>
<td>Water</td>
<td>86 mN/m</td>
<td>10</td>
</tr>
<tr>
<td>Ethanol</td>
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<td>16</td>
</tr>
</tbody>
</table>

We have been able to produce uncollapsed PDMS silia with aspect ratios much higher than these models predict, and we believe this is due to the care taken in fabrication. Specifically, our fabrication methods allow us to gently remove the template by dissolution, thereby preventing the rods from every coming in contact with each other or the ground. We do, however, notice that silia often exhibit lateral or ground collapse upon actuation with magnetic fields (which often causes silia to come into contact with each other) or fluid forces. Also, samples tend to exhibit more collapse over time, giving them a limited shelf life. Finally, the highest-aspect-ratio structures we have produced (aspect ratio 125) are difficult to fabricate without collapse, and we find that we are limited to low-density silia arrays in order to avoid collapse. Figure 16 shows FFPDMS-C16 silia of aspect ratio 125 which have succumbed to lateral collapse upon exposure to air. (Samples of this geometry have been successfully fabricated and actuated without collapse in ethanol.)
Figure 16. High-aspect-ratio FFPDMS-C16 silia exhibiting lateral collapse. In this case, collapse has been induced upon exposure to air upon preparation for the SEM rather than water, however the mechanics of the collapse are the same.

Currently, lateral collapse limits the density of our samples to about $2 \times 10^6$ silia/cm$^2$, which is about a factor of 100 less than the density of human airway cilia. If we are to be able to fabricate arrays of biological densities, we must overcome the issue of lateral collapse. Figure 17 illustrates the difference in density between human airway cilia and our biomimetic silia.
Figure 17. Scanning electron micrographs of human airway cilia (left) and biomimetic cilia (right). The 100-fold difference in density is apparent in the two images. Currently, adhesion issues prevent us from fabricating silia of higher density (not the two silia exhibiting lateral collapse in the center of the frame.)

To overcome lateral collapse without negatively impacting the flexibility of the silia, it is necessary to decrease the surface energy of the material in water. There are two available strategies for doing so: we can decrease the surface energy of the PDMS by surfactants or surface functionalization, or we can replace the PDMS with a material with a lower surface energy.
3.5.1 Surfactants for Silicones

Introducing a surfactant to the fluid is the simplest solution, and provides an incremental improvement over pure water. We have tested contact angles of water containing various surfactants – Triton-X 100, cetyl trimethylammonium bromide (CTAB), sodium dodecyl sulfate (SDS) and Pluoronic S-127 – on bulk samples of PDMS. In addition, we have found that ethanol has a lower contact angle on PDMS than water, and so – while water is the more biologically relevant material – we have often used ethanol as the liquid medium in our experiments. We therefore tested contact angles of ethanol containing each of the surfactants on bulk PDMS as well. All surfactants were mixed 1% wt. in the solvent. The only surfactants which showed a contact angle significantly smaller than the surfactant-free controls were Triton-X 100 and Pluronic S-127. Triton-X 100 showed significantly reduced contact angle in both water and ethanol, and Pluronic showed a reduced contact angle in water only. Triton-X 100 is therefore used in a 1% concentration in all liquid media following the dissolution of the template in silicone silia fabrication processes, as noted in the previous section. Results of these experiments are shown in Table 3, below.

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>Triton-X 100</th>
<th>CTAB</th>
<th>SDS</th>
<th>Pluronic</th>
</tr>
</thead>
<tbody>
<tr>
<td>water</td>
<td>100°</td>
<td>50°</td>
<td>100°</td>
<td>100°</td>
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<td>40°</td>
<td>20°</td>
<td>40°</td>
<td>40°</td>
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3.5.2 Surface Functionalization of Silicones

Another method of lowering the surface energy is to functionalize the surface of the PDMS. While functionalizing the surface after fabrication may provide a longer shelf life for low-density samples, if it is to be of any aid in producing viable high-density samples, the functionalization must be applied before the silia have the opportunity to contact one another – either within the template or during the dissolution of the template.

This limits the available strategies for functionalization to three: the functionalizing polymer may be applied to the PDMS during dissolution; the functionalizing polymer may be applied to the interior of the template before fabrication; or the functionalizing polymer may be mixed with the PDMS prior to crosslinking. These strategies are represented pictorially in Figure 18.
Three proposed schemes for functionalizing the silia either within the membrane (2 and 3), or during the dissolution of the membrane (1). In 1, the functionalizing agent is carried by the solvent and deposited on the silia as the membrane is dissolved. In 2, the membrane is coated with the functionalizing agent before it is impregnated with the ferroelastomer. In 3, the functionalizing agent is dispersed within the uncured ferroelastomer with which the pores of the membrane are impregnated. The agent will migrate to the surface of the elastomer due to a chemical affinity, after which the silia is cured, and the membrane dissolved.

The first two strategies require a chemical group in the PDMS to which the functionalizing polymer may be bound. Unfortunately, pure PDMS contains nothing but methyl groups bound to silicon along its backbone; these groups are extremely inert, and chemically useless (Figure 2). Established methods of functionalizing PDMS require that the surface be activated with either via plasma (as shown in Figure 10) or ultraviolet radiation in an oxygen or nitrogen environment. Each of these methods, however, requires that the
sample be exposed to a gas, which is not possible in many of our samples without inducing lateral collapse. However, other silicones such as PVMS or PVMS-NH2, described in Chapter 2, may provide the necessary chemical groups for effective functionalization to occur without requiring activation. Unfortunately, fabrication processes for these materials are not yet completed, and so these methods of functionalizing silicone silia have been put on hold.

The third method does not require a chemical bond between the silicone and the functionalizing material. Rather, a copolymer with both hydrophobic and hydrophilic components may be used. The hydrophobic portions remain in the silicone, while the hydrophilic portions migrate to the surface. This strategy has been used by Wang et al. with the amphiphilic copolymer Pluronic 127 in hydrophobic poly(ether sulfone) (PES) to produce a hydrophilic, antibiofouling membrane.71

We have attempted a similar strategy with a copolymer of PDMS and polyethylene glycol (PEG). While we were unable to successfully mix the copolymer with PDMS, we have attempted to treat the template with the copolymer (as in Figure 18, strategy 2) in the hopes that the hydrophobic portions of the copolymer will migrate into the PDMS while curing, leaving a hydrophobic exterior upon removal of the template. In this set of experiments, a polycarbonate membrane without pores was treated by submersion in a solution of 1% wt. PDMS-PEG (25% PEG, molar ratio) in hexane for one minute. The membranes were either treated with plasma for 20 seconds prior to the PDMS-PEG treatment, or left in their native state. The results of these experiments are shown in Figure 19. The data suggest that the amphiphilic PDMS-PEG copolymer adsorbs differently on raw and plasma-treated membranes: on a raw membrane, which is hydrophobic, the PDMS
portion of the copolymer adsorbs onto the membrane, leaving a hydrophilic PEG surface; on a plasma treated membrane, which is hydrophilic, the PEG portion of the copolymer adsorbs onto the membrane, leaving a somewhat hydrophobic PDMS surface.

Figure 19. Contact angle measurements of water on treated polycarbonate membranes. Details are in the text.

Unfortunately, the copolymer failed to transfer to bulk PDMS cured on the surface of the plasma-treated / PDMS-PEG treated membrane. All samples of bulk PDMS cured on the treated membrane showed contact angles of about 90° with water upon removal of the membrane – whether the membrane was mechanically removed or dissolved in dichloromethane. This angle is indistinguishable from untreated bulk PDMS.

Finally, a material with an inherently lower surface energy with water that a silicone may be used. A hydrogel such as FFAAm is just such a material, and contact angle
measurement of water on bulk samples of AAm have shown a contact angle indistinguishable from 0\(^\circ\). The following section will detail the fabrication of silia arrays of FFAAm.

3.6 Fabrication of Hydrogel Silia Arrays

Rod arrays made of ferrofluid-polyacrylamide (FFAAm) are fabricated in much the same way as the silicone arrays. However, polyacrylamide (AAm) will not bond to the permanently-attached PDMS well, and so other means must be devised of attaching the gel to the glass substrate. Sidorenko et al. have developed a method of bonding the polymer to the glass, by activating the glass surface with plasma and using the available OH groups to attach poly(glycidyl methacrylate) (PGMA) to the surface\(^{72}\) as in Figure 20 A and B. The epoxy groups of the PGMA are then functionalized with acrylic acid (Figure 20 C). The acrylic functionality will then become incorporated in the AAm backbone, upon polymerization of the polymer, firmly attaching the AAm to the glass substrate, as shown in Figure 20 D.
Figure 20. Functionalization of glass substrate with PGMA and acrylic acid to allow the bonding of polyacrylamide to the surface. In A and B, PGMA is placed on the surface and forms covalent bonds with the activated glass upon annealing. The PGMA is then functionalized with acrylic acid (C). Finally, when acrylamide is polymerized on this surface, it will incorporate the acrylic groups from the acrylic acid into its backbone, bonding it to the surface. This figure is from Sidorenko, 2007.

Poly(glycidyl methacrylate) is not readily available commercially, and must be polymerized in the lab. The method described here follows the work of Sidorenko. First, a solution of 30% glycidyl methacrylate (monomer, GMA) and 1% azo-bis-isobutyronitrile (AIBN) is mixed in methyl ethyl ketone (MEK) (3 mL GMA and 0.1 g AIBN in 10 mL MEK). The AIBN acts as a free radical initiator for the polymerization of the GMA. The
solution is degassed by bubbling with argon for about 10 minutes, then reacted at 60º overnight. Upon completion of the reaction, 8 mL of diethyl ether (DEE) is added to precipitate the PGMA from solution. The DEE is replaced 3-4 times, decanted, and the precipitate is dried in under vacuum overnight. The resulting PGMA forms a hard, white pellet.

Before functionalizing the glass surface with PGMA, a PDMS well is first attached to facilitate dissolution steps later in the fabrication process. The coverslip is then plasma treated for two minutes to ensure that the glass surface is activated. A mixture of 1% PGMA in MEK (0.1 g PGMA in 10 mL MEK) is ultrasonicated for 1 hour to mix, and then a few drops are added to fill the PDMS well of the sample. After 10 minutes, the sample is rinsed with ethanol and dried with nitrogen. The sample is placed in an oven at 110º C for 10 minutes to encourage bonding of the epoxy groups of the PGMA to the surface of the glass. The well is then filled with acrylic acid, and the sample is left in the oven at 110º C for an additional 15 minutes for the reaction to take place. Upon removal, the sample is rinsed with ethanol and dried with nitrogen. The surface is now prepared for the acrylamide gel.

The ferrofluid-acrylamide (FFAAm) rod arrays are cured in a manner very similar to that of bulk acrylamide, as described in Chapter 2, with a few exceptions. Uncured AAm and FFAAm have such low viscosities (similar to that of water) that simply filling the pores with FFAAm and then submerging the sample in AAm (similar to the method used for curing arrays of FFPDMS rods) is not effective. The FFAAm is likely to flow out of the pores as soon as the membrane is submerged in AAm, to be replaced by non-magnetic polymer. In order to overcome this difficulty, curing proceeds in two steps: first, the FFAAm rods are
cured in the membrane, then the membrane is placed on top of an AAm substrate, which is subsequently cured in the presence of the FFAAm nanorods.

To accomplish this, an appropriate PCTE membrane is cut and submerged in a solution of uncured FFAAm, before the addition of catalyst or initiator (APS and TEMED). The solution is ultrasonicated or placed under vacuum to eliminate air bubbles and ensure that the FFAAm solution has permeated the pores. The solution is bubbled with argon for 10 minutes, after which APS and TEMED are added, as described in Chapter 2. After several minutes in an argon-purged environment, the FFAAm will have polymerized and crosslinked. Encapsulated in the FFAAm gel, the membrane may be stored in a sealed container indefinitely. To prepare a membrane for use, it is removed from the FFAAm gel, and the surface is cleaned of excess FFAAm, leaving only the pores filled.

Next, a solution of 6% acrylamide monomer and 0.3% bisacrylamide in water (0.54 mL of the 20:1 AM:BA solution described in Chapter 2 in 1.46 mL deionized water) is prepared, and deoxygenated with argon for 10 minutes. 40 microliters of 10% APS is added to the AAm. A small amount of this solution is removed and beaded on the surface of a Petri dish in an argon-purged environment. Next, a couple of microliters of TEMED are added to the bead of AAm solution; at this point, the AAm will begin to cure rapidly. Before the AAm has cured, the cleaned membrane, impregnated with FFAAm, is touched to the bead of AAm to coat one side with uncured polymer mixture. The membrane is then quickly set, wet-side-down, on the functionalized glass coverslip, within the PDMS well. The thin layer of AAm will cure in an argon-purged environment within a couple of minutes, permanently adhering the FFAAm rod array to the glass surface. This sample geometry is very similar to
that of silicone silia, shown in Figure 11, except that the membrane is not covered with a polymer layer on top.

The membrane is dissolved in a manner identical to that for FFPDMS rod arrays, with the exception that the upper face of the membrane need not be peeled prior to dissolution since it is already exposed. The cured sample should not be left exposed to ambient air for long periods of time before dissolution, since the hydrogel is prone to drying out. However, it may be stored underwater indefinitely.

Since the maghemite ferrofluid is water-based, there is some concern over whether the nanoparticles are prone to diffusing out of the polymer network during storage (or regular use, depending on the time scale of the diffusion). Galicia et al. have observed significant nanoparticle leeching in polyacrylamide gels with a 1% crosslinker to monomer ratio in BA crosslinked PAAm. However, Mayer et al. have reported that nanoparticles do not leech with higher concentrations of crosslinker, and this result is consistent with our own experiments on bulk materials.

Using these techniques, we have produced arrays of FFPAAm silia with lengths of 20 microns and diameters ranging from 200 – 700 nm. Since the silia are about 90% water, they are not suitable for imaging via SEM, which must occur in vacuum. Additionally, optical images of successful samples are taken from above, showing only spots to indicate the presence of rods. The best image of FFPAAm silia, then, is an optical image of ground collapsed rods, shown in Figure 21.
3.7 Adhesion Issues in FFAAm Silia

While FFAAm has an extremely low surface energy in water and therefore would not be expected to aggregate in an aqueous suspension, the vast majority of FFAAm samples we have made have been unactuable due to lateral or ground collapse. At this point, the nature of this collapse remains a mystery. Efforts are underway, however, to eliminate collapse by
surface functionalization of the FFAAm. We have successfully employed two strategies for functionalizing bulk acrylamide. Both methods serve to bind polyethylene glycol (PEG) to the surface of the acrylamide, in hopes of forming a polymer brush. PEG is widely regarded as an all-purpose anti-adhesion coating, and polymer brushes in particular are noted for their self-repelling properties.73

3.7.1 Functionalization with COOH-Terminated PEG

The acrylamide gel has an amine group at every repeat unit, which is an ideal handle for functionalization with a variety of reagents. One standard reaction which links amines with carboxyl groups uses 1-ethyl-3-(3-dimethylaminopropyl) carbodiimide hydrochloride (EDAC) as a linker and is referred to as an EDAC reaction. This reaction might be useful in functionalizing the amine-containing polyacrylamide with a carboxy-terminated PEG.

In testing the efficacy of the reaction, it would be difficult to determine whether the COOH-PEG were bound to the surface as we expect, and so it is useful to first test the reaction by linking a visible marker to the polyacrylamide substrate. In this case, we chose to use a fluorescent bead with a carboxy functionality.

To perform the reaction, a bulk sample of polyacrylamide was submerged in a 0.1 M 2-(N-morpholino)ethanesulfonic acid (MES) buffer of pH 5.4 for fifteen to twenty minutes. In a separate test tube a solution of buffer and carboxy-functionalized beads was sonicated for several minutes to ensure that the beads were freely floating in the solution and able to attach to the surface easily. EDAC was added to this solution in a 10-fold excess to the number of available carboxy functionalities. The solution of beads was added to the bulk acrylamide and gently mixed for 15 minutes. Finally, a 0.2 M MES buffer with a pH of 12-
13 was added and mixed gently for one hour, after which the sample was rinsed twice with a trishydroxymethylaminomethane (TRIS) buffer.

The bulk acrylamide was then removed from the solution, rinsed with deionized water, and observed with fluorescence microscopy. A control was prepared in exactly the same manner, but without the addition of EDAC. The reacted sample exhibited a brightness of 180 ± 100, as measured by average grayscale value, while the control sample exhibited a brightness of only 20 ± 30. This is a clear indication that the reaction has succeeded.

### 3.7.2 Functionalization with Thiol-Terminated PEG

Another method of functionalizing polyacrylamide with a PEG brush takes advantage of a thiol-gold bond to bind a polymer to the substrate. In this method, a portion of the bis-acrylamide crosslinkers (BA) are replaced with a similar crosslinker containing a dithiol bond, bisacryloylcystamine (Bis-CA). After polymerization and crosslinking, some of the dithiol bonds of the Bis-CA are broken with tris(2-carboxyethyl)phosphine) (TCEP), leaving thiol groups exposed on the surface of the polymer. The surface is then exposed to colloidal gold particles in water, which bind to the thiols on the surface. Finally, after rinsing, a thiol-terminated PEG (SH-PEG) is allowed to bind to the gold particles, resulting in a pegylated acrylamide surface.

We tested this reaction on a bulk sample by bonding a thiol-biotin and subsequently a fluorescent streptavidin to bound gold particles, and found a significant increase in fluorescence relative to controls prepared without TCEP, indicating that the reaction proceeded as expected.
These methods show promise in functionalizing the surface of bulk FFAAm. However, they are unlikely to be of use in preventing adhesion in silia arrays. As we noted earlier, in order to be useful the surface functionalization must be applied before the silia have had a chance to interact with each other – either within the template or during dissolution of the template. The EDAC reaction described above occurs in water and must therefore be performed after the dissolution of the membrane. The dithiol linkage has the complication of being an A-A reaction, and unlinked thiol groups on the silia surface are as likely to bind to other thiols on another silia as to the thiol-terminated PEG.

An A-B reaction (in which each reactant is unable to bond to those of the same kind) would be ideal, and again, performing the reaction within the template before dissolution would avoid premature losses due to lateral collapse. One strategy which conforms to these requirements is outlined below.

3.7.3 Functionalization with Template-Bound Mucins

It is generally thought that biological cilia are covered in polymer brushes made of glycoproteins which are bound to the cell membrane via a hydrophobic region. The sugars in the glycoproteins are charged, and so the proteins, bound side-by-side on the cilium surface form a highly charged polymer brush which repels the polymer brushes of neighboring cilia. It would be interesting to reproduce these same brushes on artificial silia made of FFAAm. Fortunately, work by Schnaar and others\textsuperscript{74-76} has resulted in a method for immobilizing proteins on the surface of a polyacrylamide gel, and Pless et al. have used these techniques specifically for bonding glycoproteins to polyacrylamide.\textsuperscript{77} This work opens the door to
creating polyacrylamide silia which have a glycoprotein bush similar to that of their biological cousins.

To bind proteins to the gel, the acrylamide monomer would first be copolymerized with a small amount of N-acryloxsuccinimide, which becomes a part of the polymer backbone via its acrylic functionality. This leaves active esters in the crosslinked polymer, which are able to bind to amine functionalities in a protein. This reaction is attractive because it occurs at room temperature with no catalysts, and is an A-B type reaction. In addition, the glycoproteins with which we would like to functionalize the silia are hydrophilic, and may adsorb from solution onto a plasma-treated polycarbonate membrane template. This would allow for the functionalization of the silia within the bounds of the template, during polymerization.

3.8 Conclusion

In this chapter, I have presented fabrication processes for making large arrays of high-aspect-ratio silia of the various materials described in Chapter 2. We have used these fabrication processes to produce soft, actuable structures of unprecedented aspect ratio. The density of the arrays is limited, however, by lateral collapse, and I have described strategies which may be employed to alleviate this problem in the future.

In the next chapter I will turn to the issue of actuating these silia arrays, and I will detail the mathematical model which I have produced to predict how the silia will bend in prescribed magnetic fields.
4.1 Introduction

Our goal is to be able to design rod arrays and magnetic field geometries to generate a variety of spatial and temporal beat patterns. To this end I have developed an energy minimization model of a flexible magnetic rod in the presence of a defined magnetic field and field gradient. A model for the energy of a flexible magnetic rod in a constant magnetic field has been proposed by multiple authors.\textsuperscript{4, 78-80} However, because our experimental results are not consistent with a constant, gradient-free field, we have developed a model which is able to account for the effects of both a magnetic field and a magnetic field gradient on a cantilevered, flexible magnetic rod. We conclude that the effect of a torque applied by a magnetic field may under certain conditions be dominated by the effect of a force caused by a magnetic field gradient. One very relevant case under which this gradient dominance may occur is under actuation due to the field generated by a current carrying wire. This will be discussed in detail in Chapter V, “Actuation.”

In this chapter, I will begin with a simple analytical expression for the energy of a cantilevered magnetic rod in an applied magnetic field of constant gradient – the “Continuous Model.” This expression will be used to explore some of the parameter space, offering simple analytical expressions for special cases of bending, such as field-dominated bending and gradient-dominated bending. Expressions derived in the continuous model are
useful in instructing the design of biomimetic silia geometries and actuating field geometries, and will be put to use in the next chapter, “Actuation.”

The continuous model, however, is only an approximation, and has the added limitation of predicting only bend angle, and not bend shape. In order to remedy both of these constraints, I have developed a refinement of the continuous model in which the silium is subdivided into an arbitrary number of segments. I derive an expression for the sum of the energy of each of these segments in an applied field, which can be minimized to predict bend shape. This model also more accurately predicts bend angle, since it does not incorporate many of the assumptions of the continuous model. I refer to this model as the “Discrete Model.”

Finally, I derive the torque applied to each element of the silium in the discrete model, which leads to a much faster and more robust method of finding the minimum energy state of the silium than an energy minimization routine. This is described in the last section of this chapter.

4.2 The Continuous Model

In general, the energy of a bent, cantilevered rod in a magnetic field consists of two components: the elastic energy of the rod bent from equilibrium, and the magnetic energy. The magnetic energy can be further subdivided into two elements: the energy associated with the misalignment of the axis of the rod with an applied magnetic field, and the energy associated with the tendency to move along the gradient toward areas of higher field.

To derive a simple analytical expression for the energy of a cantilevered magnetic rod in a magnetic field, it is necessary to make a few simplifying approximations. This model
attempts to achieve a balance between simplicity (for utility in drawing useful generalizations about the parameter space) and accuracy. Specifically, this model assumes a constant curvature with respect to elastic energy, and a straight, rigid rod with respect to magnetic energies. These assumptions lead to a model which is simple enough for utility, yet generates results that agree well with calculations from the more complete discrete model.

4.2.1 Elastic Energy

The total energy of the magnetic rod system is given by $U_T = U_E + U_M$, where $U_E$ is the elastic energy of the bent rod and $U_M$ is the magnetic energy of the rod in an applied magnetic field. The elastic energy is described by the Kirchhoff model of a uniform elastic rod and has the form

$$U_E = \frac{1}{2} EI \int_0^L \frac{1}{R(s)^2} ds$$

where $E$ is the Young’s modulus of the rod, $l$ is the bending moment ($l = \frac{\pi}{4} t^4$ for a rod of circular cross section), and $R(s)$ is the radius of curvature of the bent rod. In this model we do not solve for the shape of the rod in an applied field, but assume a constant curvature, and hence set $R(s) = R$. We therefore set $R = L/2\phi$ (see Figure 22), and the elastic energy can be written as

$$U_E = \frac{\pi}{2} \frac{E}{L} \frac{t^4}{\phi^2}$$
4.2.2 Magnetic Energy

In general, the magnetic energy contained in a rod can be described by the following integral,

$$ U_M = -A \int_0^l \mathbf{B}_I(s) \cdot \mathbf{M}(s) \, ds $$

(5)

where $A$ is the cross-sectional area of the rod, $\mathbf{M}(s)$ is the magnetization at a point along the length of the rod and the total field $\mathbf{B}_I = \mathbf{B}_N + \mathbf{B}_i + \mathbf{B}_A$, where $\mathbf{B}_N$, $\mathbf{B}_i$, and $\mathbf{B}_A$ are the magnetic field from neighboring rods, the internal magnetic field, and the applied
external magnetic field, respectively. In this sense, the total magnetic energy can be thought of as a sum of three energies, one for each of the component fields.

\[
U_M = U_N(B_N) + U_I(B_I) + U_A(B_A)
\]  \hspace{1cm} (6)

4.2.2.1 Field from nearest neighbor

We can show that the field produced by nearest neighbor rods is negligible compared to the magnitude of the applied field. The magnetic field due to a magnetic dipole at any position \( \hat{x} \) is

\[
B(r) = \frac{\mu_0}{4\pi} \frac{\mathbf{m} \cdot \hat{x}}{r^3} (3(\mathbf{r} \cdot \hat{x})\hat{x} - \mathbf{r})
\]  \hspace{1cm} (7)

The greatest magnitude occurs for \( \mathbf{m} \parallel \hat{x} \), and is

\[
B_{\text{max}} = \frac{\mu_0}{2\pi} \frac{m}{x^3}
\]  \hspace{1cm} (8)

Taking the value of the magnetic moment \( m \) to be the moment at magnetic saturation, \( m = M_{\text{sat}} \pi^2 L f \), where \( M_{\text{sat}} \) is the saturation magnetization of the magnetic material. For maghemite, this is approximately 300,000 A/m. The largest rods used in experiments in this work have radii of about 500 nm and length of about 25 microns. Finally, magnetic loading content is generally limited to < 20% by volume. (In fact, nearly all the data presented for actuated nanorod arrays in this dissertation come from rods made of FFPDMS-C16-maghemite, which has a volume fraction of only 4% maghemite.) The separation between two rods is on the order of 10 microns. These numbers yield a maximum magnetic field from a neighboring rod of
As shown in (this section), typical actuating fields are on the order of a few hundred Gauss, or a few hundredths of a Tesla. This is two orders of magnitude larger than the maximum field from a neighboring rod, so these neighboring fields will be ignored. (This is, of course, an extreme example as well; more typical values of a 250 nm radius and a 4% volume fraction of maghemite yield a truly negligible neighboring field strength of $10^{-5}$ Tesla.)

It should be noted, however, that the effect of the field from a neighboring rod is highly dependent on rod spacing, and if future experiments approach significantly smaller rod spacing, this effect may become important. For example, for a geometry similar to that of human airway cilia, rod spacing may be on the order of 0.5 microns. Given a length of about 10 microns and a cilia radius of about 100 nm, and assuming a large maghemite loading of 20%, the field due to a neighboring rod at saturation becomes 0.03 T, which is on the same order as the applied field.

### 4.2.2.2 Internal energy

The second term, the internal magnetic energy of the rod, has been described by Stoner-Wohlfarth for an ellipsoid of revolution.\(^82\)

\[
U_1 = \frac{1}{2} \mu_0 \frac{m^2}{V} (N_a - N_b) \cos^2(\alpha - \phi) \tag{10}
\]

Here $\alpha$ and $\phi$ are the angular positions of the net magnetic moment of the rod, $m$, and the long axis of the rod, respectively, from the vertical (Figure 6). In the Stoner-
Wolfarth model, $N_a$ and $N_b$ are the demagnetization factors along the long axis of the rod and along the radius of the rod, respectively, and $V$ is the volume of the rod. For a rod of aspect ratio greater than 20, as is satisfied by our experiments, $N_a \sim 0$ and $N_b \sim 0.50$. In addition, it can be shown that under the condition $4B \mu_0 M(B)f \gg 1$, which is satisfied by our experiments, the magnetic moment of the rod aligns with the applied magnetic field. (See Appendix A for details) We can therefore replace $\alpha$ with $\psi$, the angular position of the applied field with respect to the vertical. The internal magnetic energy for a straight rod then becomes

$$U_I = -\frac{1}{2} \mu_0 \frac{m^2}{V} \cos^2 (\psi - \phi)$$  \hspace{5em} (11)

### 4.2.2.3 Energy due to Applied Field

The third magnetic energy term, the energy due to the applied field, may be written as follows (see Appendix A for derivation and other details.)

$$U_A = -\frac{1}{2} m \nabla B \cos (\psi - \phi)$$  \hspace{5em} (12)

where $\psi'$ is the angular position of the magnetic field gradient with respect to the vertical (Figure 6). We can now write down the total energy of the rod, elastic and magnetic.

$$U_T = \frac{\pi}{2} Er^4 - \frac{1}{4} \mu_0 \frac{m^2}{V} \cos^2(\phi - \phi) - \frac{1}{2} m \nabla B \cos (\psi - \phi)$$  \hspace{5em} (13)

In general, the magnetic moment $m = M(B)f$, where $M(B)$ is the magnetization of the magnetic particles in the rod and $f$ is the volume fraction of magnetic material. In our model, the magnetization curve $M(B)$ is determined by SQUID measurements of the maghemite, allowing us to determine the magnetic moment $m$ for any applied field.
At this point it is evident that the third term in the total energy does not depend on the magnetic field directly, but only on the magnetic field gradient. This term represents the effect of the gradient pulling on the center of mass of the bent rod, which we will call “gradient energy.” The second term in the total energy depends on the direction of the magnetic field and will be called “field energy.” The field energy term represents the misalignment of the long axis of the rod from the direction of the applied field.

4.2.3 Special Cases

The continuous model is very useful in gaining an understanding of the interaction between the magnetic field geometry and the magnetic rods. The model, however, represents an enormous parameter space with many independent variables: radius, length, magnetic loading, Young’s modulus, magnetic field, magnetic field gradient, and bend angle. It is impossible to plot the entire parameter space on any one set of axes. However, slices through this parameter space can yield very useful insights and information.

This complete continuous model is extremely useful for determining the degree of bending of a given rod in a given field geometry, and such analysis is useful in very specific geometries, such as in the experiment detailed in “Comparison with Experimental Results,” below. In designing future experiments, however, it is more useful to make generalizations about how rods of given physical parameters may behave in a wide range of field and gradient magnitudes. The complete energy expression in equation 13 is not very useful for doing so; it is much more instructive to consider the effects of the field and gradient separately.
Examining each of these effects independently provides simple and useful analytical expressions. By taking the derivative of the energy function with respect to the angle $\phi$, we find that

$$\frac{\partial U_T}{\partial \phi} = \pi \frac{Er^4}{L} \phi - \frac{1}{2} \mu_0 \frac{m^2}{V} \cos(\theta - \phi) \sin(\psi - \phi) - \frac{1}{2} m \nabla B L \sin(\psi - \phi)$$  \hspace{1cm} (14)

This function represents the torque on the rod, or, by setting $\frac{\partial U_T}{\partial \phi} = 0$, yields the minima of the energy function.

4.2.3.1 Pure Gradient Bending

To consider the gradient effect only, we can require that the field be aligned with the rod (i.e. $\psi = \phi$), in which position it will exert no torque. This allows the field to effectively be turned off without eliminating the magnetization of the rod, $m$, which is required for the gradient effect to occur. Under this condition, the minimum of the energy function can be found by

$$\pi \frac{Er^4}{L} \phi - \frac{1}{2} m \nabla B L \sin(\psi - \phi) = 0$$  \hspace{1cm} (15)

or

$$\nabla B = \frac{2Er^2}{Mf L^3} \frac{\phi}{\sin(\psi - \phi)}$$  \hspace{1cm} (16)

where the magnetization $m$ has been replaced by $m = M f \pi^2 L$. Considering an optimal alignment of the gradient direction with the rod position, such that $\sin(\psi - \phi) \rightarrow 1$, the minimum gradient required to bend a rod through an angle $\phi$ is then given by this simple expression:
\[ \nabla B_{\text{min}} = \frac{2E}{Mf} \frac{r^2}{L} \phi \]  \hspace{1cm}(17)

This expression can provide very useful information as to what gradient is necessary to effectively bend rods with a wide range of physical parameters, as shown in Figure 23. Here we interested in the minimum gradient required to bend a rod of given magnetic loading and Young’s modulus through a representative angle of 10 degrees. We have further limited the parameter space by fixing the length and radius – in this case they have been selected to allow the maximum amount of bending (i.e. large \( L \) and small \( r \)).

Figure 23. The gradient required (boxed values in units of T/m located on the contour lines) to bend a rod of radius 125 nm, length 25 \( \mu \)m, and a range of magnetic loading (\( f \)) and Young’s moduli (\( E \)), given the condition of pure gradient bending represented by Equation 14. The blue and green boxes
represent reasonable ranges of material properties for hydrogel-based microrods and silicone-based microrods, respectively, and the red spot indicates the material properties of FFPDMS-C16.

The plot clearly provides benchmark values for magnetic field gradient magnitudes which must be attained in order to actuate a given microrod. In addition, it is relatively simple to extrapolate to rods of different radius or length, by recognizing that the required gradient is proportional to $r^2$ and $L^{-3}$, as shown in equation 17. This plot will be used to predict the efficacy of field generating mechanisms in Chapter 5: Actuation.

4.2.3.2 Pure Field Bending

Similarly, the effect of the magnetic field can be isolated by setting the magnetic field gradient in equation 14 to zero. In this case, setting the torque to zero then yields

$$\pi \frac{Er^4}{L} \phi - \frac{1}{2} \mu_0 \frac{m^2}{V} \cos(\psi - \phi) \sin(\psi - \phi) = 0$$

(18)

and solving for $M$ (where, again, $m = M f \pi^2 L$) gives the magnetization per unit volume necessary to bend a rod through an angle $\phi$:

$$M = \frac{1}{f} \sqrt{\frac{2E}{\mu_0 \cos(\psi - \phi) \sin(\psi - \phi) L}} \phi^{1/2}$$

(19)

Considering an optimal alignment of the field direction with the rod position, $\cos(\psi - \phi) \sin(\psi - \phi) \to \frac{1}{2}$, and

$$M_{\text{min}} = \frac{2}{f} \sqrt{\frac{E}{\mu_0 L}} \phi^{1/2}$$

(20)
The magnetization per unit volume is a function of the applied magnetic field, and an appropriate mapping may be used to restate this relationship in terms of applied field, which is a more easily controlled variable. Ideally, the mapping would be a full hysteresis curve for the magnetic material, but in special cases such as very small or very large applied fields, a linear relationship is sufficient.

Figure 24 makes use of Equation 20 to demonstrate what field is necessary to bend microrods with a wide range of physical properties through an angle of 10 degrees. The full hysteresis curve of maghemite (as shown in Figure 3) has been chosen as the mapping between magnetization and field.

![Figure 24. The field required to bend a rod of radius 125 nm, length 25 µm, and a range of magnetic loading (f) and Young's moduli (E), given the condition of pure field bending represented by Equation 17. The blue and](image)
green boxes represent reasonable ranges of material properties for hydrogel-based microrods and silicone-based microrods, respectively, and the red spot indicates the material properties of FFPDMS-C16.

It is interesting to note that the extreme upper-right corner of the plot (at low magnetic loading and high Young’s modulus) represents an area of the parameter space in which it is impossible to bend a rod through ten degrees via pure field bending. This occurs because the applied magnetic field results in the saturation of the rod at values above about 2 Tesla (see Figure 3), resulting in no further increase in the magnetization $M$, and therefore no increase in bend angle $\phi$.

By solving Equation 20 for the bend angle $\phi$, we find that

$$\phi = \frac{\mu_0 M \frac{L}{2r}}{E \left( \frac{L}{2r} \right)^2}$$

(21)

The second fraction contains the geometrical parameters of the silium, and the first fraction contains only material properties. This first term is the ratio of magnetic to elastic energies, and is referred to in this text at the magnetoelastic ratio. This ratio tells us how the material properties of the silium affect field bending, and is therefore used as a figure of merit in selecting candidate materials. A table of candidate materials with their respective magnetoelastic ratios is in Chapter 2, “Materials.” (Table 1)

4.2.3.3 Gradient Bending vs. Field Bending

Finally, it may be useful to determine which method of bending, field or gradient, will be most dominant under particular circumstances, in order to inform the design of useful actuation fields. We can find the ratio $R$ of pure gradient bending to pure field bending by
dividing the gradient torque (the third term of Equation 14) by the field torque (the second term of Equation 14):

\[
R = \frac{2\nabla B L}{M(B) \mu_0 f}
\]  

(22)

Here again we have assumed that the field and the gradient are applied in their respective optimal directions, in order that each provides maximum torque. We have also used the fact that \( m = M(B) f \pi r^2 L \). It is interesting to note that the ratio of the torques is independent of both the radius \( r \) and the Young’s modulus \( E \). This fact is helpful in reducing the dimension of the parameter space, and we can obtain a very useful plot by fixing the magnetic loading \( f \) and length \( L \), and plotting the ratio \( R \) as a function of both applied field and applied field gradient.
Figure 25. The ratio $R$ of gradient bending to field bending for the case of a 25 µm rod made of FFPDMS-C16. Areas to the lower right of the $R = 1$ contour represent gradient-dominated bending; areas to the upper left represent field-dominated bending.

Figure 25 and Figure 26 show the ratio of gradient bending to field bending across reasonable ranges of magnetic field and magnetic field gradient. The rod parameters ($f$ and $L$) in Figure 25 best represent the current formulation of FFPDMS rods, while those in Figure 26 represent expected parameters for a hydrogel rod. From these plots it is clear that bending in hydrogel rods will be almost entirely dominated by field bending. In fact, the FFPDMS rod represented in Figure 25 represents both the lowest magnetic loading $f$ and the longest length $L$ likely in any future silia formulations, and therefore boasts the highest values of $R$. It is likely, therefore, that bending with future materials will occur almost exclusively in the field regime.
4.2.4 Comparison with Experimental Results

The full energy function, with both field and gradient effects, can be applied to a specific magnetic field geometry to determine the angular position of the rods at any point in space. We use a finite element modeling software (COMSOL Multiphysics) to generate a model of the magnetic field geometry created by the conical actuating magnet, allowing us to determine the field and field gradient at any position with respect to the actuating magnet. We adjust the physical parameters of the COMSOL model to match measured values of the magnetic field along the axis of the cone tip (measured by a F.W. Bell Model 5080...
The magnetic field, $B$, magnetic field gradient, $\nabla B$, and the angles of each with respect to the vertical, $\psi$ and $\psi'$, at a particular rod position are then passed to MatLab 7.1, which minimizes the total energy function for the given field and gradient with respect to $\phi$. The $\phi$ which minimizes the total energy is output as the final angular position of the rod.

Figure 27. (A) Diagram of magnetic field lines from the steel-tipped magnet used in these experiments. (B) Definition of rod and field geometries. (C) and (D) Graphs of rod bending angle (top) and magnetic energies (bottom) vs. magnet tip position ($X$) at a constant $z$. For both (C) and (D), in the upper plot the open circles are the experimentally observed average bending and the solid line is the theoretically predicted bending. When the magnet’s tip is within $z \sim 0.5$ mm (D) there is a rapid switching of the rod orientation as it passes over the tip. Our theoretical calculations suggest that at distances closer to the magnet the gradient energy (dashed lines) becomes larger than the field energy (solid lines) causing the rods to bend away from the field lines.

We performed an actuation experiment in which a magnet was moved beneath the sample while under observation by optical microscopy (as described above and pictured in Figure 8.) The magnet was comprised of two cylindrical neodymium iron boride (NIB) magnets (25 mm diameter, 6 mm height) topped with a cone of 12L14 steel. This produced a field in the region above the magnet (see Figure 27A) in which the magnetic field and the
field gradient point in opposing directions. In addition, the steel tip focuses the field lines and produces a particularly large gradient near the tip of the cone.

This steel-tipped magnet was translated beneath the rod array at a distance \( z \) below the rods. The plots in Figure 27 show the average angle through which the rods are bent as a function of the rod position relative to the magnet tip for two different heights above the magnet. To obtain the theoretical fit displayed in each plot, we used an algorithm written in MatLab to minimize the root-mean-square deviations between the theoretical and experimental curves with respect to the physical parameters \( f, E, r, \) and \( L \). Since the experimental data in the following two plots represent the same rods at different heights above the magnet, the root-mean-square deviations of both sets of curves were minimized simultaneously with respect to the same set of parameters.

According to the minimization routine, a Young’s modulus of 2.6 MPa best fit the experimental data shown in Figure 27. The literature suggests a Young’s modulus of 2.3-2.7 MPa for Sylgard 184 PDMS with a 1:10 curing agent to prepolymer ratio.\(^8\) Work by Guild and Young\(^2\) suggests that the small amount of maghemite nanoparticles in our FFPDMS rods (~ 4% by volume) should have little effect on the Young’s modulus of the material.

The upper limit for the volume fraction of maghemite loading in our rods was taken to be 4%, as indicated by SQUID measurements on a bulk sample of FFPDMS. However, the actual fraction of maghemite in the templated rods may be significantly lower, due to dilution of the material with hexadecane during processing, possible exclusion of maghemite aggregates from the pores, or dilution of the FFPDMS due to diffusion of PDMS and curing agent into the pores just before curing. The minimization routine returned a value of 1.4%, which is consistent with our expectations.
We have measured the radii and lengths of multiple rods in SEM micrographs of rods produced by the templates used in our experiments and have found the radii to be 125 ± 25 nm and the lengths to be 22.5 ± 2.5 microns. The 21 micron length and 136 nm radius returned by the minimization routine fall well within these ranges.

The optimized model fit and the experimental bending data are shown in Figure 27. A novel feature of these experiments which cannot be predicted by a constant field model is exhibited in Figure 27D. In this example, the positive angles exhibited at large distances to the right of the magnet (and negative values exhibited at large distances to the left) indicate an alignment of the rods with the magnetic field of the conical tip, and therefore a tendency to point away from the magnet. This behavior occurs in areas of large field energy and small gradient energy. Closer to the magnet, however, the field gradient becomes large enough to overwhelm the field alignment effect, and the rods are pulled by the gradient toward the magnet tip (shown in the large negative angles immediately to the right of X = 0, and large positive angles to the left.) We have seen this behavior in planes very close to the tip of the magnet (less than ~500 μm) where the gradient is able to dominate, and not at greater heights above the magnet.

4.3 The Discretized Model

While the model presented above is very useful in gaining insight into the mechanisms by which uniformly magnetized nanorods bend, and in predicting field strengths and geometries which are likely to produce intended bending, it has one major failing: it cannot predict the shape that the rod will take in a particular field. Biological cilia bend in a particular manner which may be essential to the production of net fluid flow in the cilia-
mucus system; if we are to reproduce these bend shapes, it is essential that we have a predictive model which will allow us to design materials and magnetic field geometries that will do so.

The previous model makes certain assumptions about the bend shape, rather than finding the minimum-energy shape that a nanorod will naturally take. In the case of elastic energy, it assumes constant curvature, and in the case of magnetic energy, it assumes that the rod is straight. As described previously (earlier in this chapter and in Appendix A, respectively), these assumptions result in relatively small errors; the previous model does not suffer much for accuracy, but lacks the ability to predict bending shapes. In order to remedy this, a new model has been generated which treats a nanorod as a series of segments and allows each segment to position itself to achieve a minimum energy. This is referred to as the “discretized model.” The difference between the continuous and discrete models is graphically represented in Figure 28.
Figure 28. A schematic representation of the difference between the continuous model and the discretized model. In the continuous model, the silicon is treated as a single element. For magnetic energies, it is assumed to be straight; for elastic energies it is assumed to have a constant curvature. In the discretized model, the rod is taken to be composed of many segments, each of which has a magnetic energy and is coupled via elastic energy to the neighboring segments.

4.3.1 Elastic Energy

Beginning as before with the expression for elastic energy of a thin cylinder from Landau and Lifshitz, \(^{85}\)

\[
U_E = \frac{1}{2} EI \int_0^L \frac{1}{R(s)^2} ds \tag{23}
\]

where \(E\) is the Young’s modulus and \(I\) is the bending moment of inertia. As before, we need only find the value of the radius of curvature, \(R\). Rewriting this in terms of a summation over \(N\) segments of a discretized rod, rather than an integral along the length \(s\),
Here the Young’s modulus $E_n$ has been specified independently for each segment of the rod, allowing for greater generality. $R_n$ represents the radius of curvature between the $n^{th}$ and $(n+1)^{th}$ segments of the rod. Figure 29 depicts this radius of curvature between two neighboring rod segments, each of length $\frac{L}{N}$, where $L$ is the total length of the rod and $N$ is the number of segments into which the rod is broken.

\[
U_E = \frac{1}{2} \sum_{0}^{N} E_n \frac{1}{R_n^2} \frac{L}{N}
\] (24)

Figure 29. Geometrical definitions for the derivation of the radius of curvature of two consecutive elements of a silium. The dark grey curve represents two neighboring segments; the dark dots are at the endpoints.
Since it is evident that $c = R - a$, then by the Pythagorean theorem,

$$(R - a)^2 + b^2 = R^2,$$

and therefore $R = \frac{a^2 + b^2}{2a}$. Furthermore, since the line segment $a$ necessarily bisects the angle $\theta$, it can be shown from simple trigonometry that $a = \frac{L}{N} \cos \frac{\theta}{2}$ and $b = \frac{L}{N} \sin \frac{\theta}{2}$. Plugging this into the equation for $R$ presented above and simplifying,

$$R = \frac{L}{N} \frac{1}{2 \cos \frac{\theta}{2}}$$

(25)

From geometrical considerations represented in the figure above, one can show that $\theta = \phi_n + \pi - \phi_{n+1}$, where $\phi_m$ represents the absolute angle (relative to the perpendicular to the substrate) of the $m^{th}$ segment of the rod. Substituting this into the equation for $R$ above, one can find that

$$\frac{1}{R^2} = 4 \left( \frac{N}{L} \right)^2 \cos^2 \left( \frac{\pi}{2} + \frac{\phi_n - \phi_{n+1}}{2} \right)$$

(26)

Plugging this into equation 24 and simplifying, the final expression for the elastic energy of the bent rod, summed over its $N$ segments, is

$$U_e = \frac{\pi N}{2L} \sum_{n}^{N} E_n \cos^2 \left( \frac{\pi}{2} + \frac{\phi_n - \phi_{n+1}}{2} \right)$$

(27)

where the bending moment of inertia has been taken to be $I = \frac{\pi}{4} r^4$, as in the continuous model.
4.3.2 Magnetic Energy

As in the continuous model, the magnetic energy can be split into three parts: the energy due to the applied external field, $U_A$, the energy due to internal magnetic fields, $U_I$, and the energy due to a field from a neighboring rod, $U_N$. As shown in (this section), $U_N$ is negligible, and will be ignored.

4.3.2.1 Energy due to applied field

The energy $U_A$ will be written as a sum of the energies associated with each of the $N$ segments which make up the rod,

$$U_{A,n} = \sum_{n=1}^{N} m_n \cdot B_n = m_n B_n \cos(\alpha_n - \psi)$$

(28)

Here, $\alpha_n$ is the angle of the magnetic moment of the $n^{th}$ rod segment with respect to the perpendicular, as shown in Figure 22. The magnetic moment of the $n^{th}$ rod segment, $m_n$, is equal to the magnetization of the segment per unit volume, $M$, multiplied by the volume of the segment, which has a length of $\frac{L}{N}$.

$$m_n = M (B_n) f_n \pi r^2 \frac{L}{N}$$

(29)

where $f_n$ is the volume fraction of magnetic material in the rod segment (which is allowed to vary from segment to segment, for generality), and $B_n$ is the field magnitude at the location of the $n^{th}$ segment of the rod.

To find the value of $B_n$, we assume that the gradient of the applied magnetic field is constant over the volume occupied by the rod. Knowing the magnetic field at the base of the rod, $B_0$, and the gradient at the rod, $\nabla B$, the local field at the $n^{th}$ segment can be written as

$$B_n = B_0 + l_n \nabla B \cos(\psi' - \beta_n)$$

(30)
(see Appendix B for details) where $l_n$ is the vector pointing from the base of the rod to the $n^{th}$ segment, and $\beta_n$ is the angle of $l_n$ with respect to the vertical, as shown in Figure 30. $\psi$ is the angle of the field gradient with respect to the vertical.

Figure 30. Geometrical definition of the angle $\beta_n$ and the vector $l_n$.

Since the model will be parametrized in terms of the angles of each of the $N$ rod segments relative to the vertical, $\phi_n$, it is necessary to rewrite $l_n$ and $\beta_n$ in terms of these quantities. The length of the vector $l_n$ can be written in terms of its vertical and horizontal components:

$$l_n = \sqrt{l_{n,x}^2 + l_{n,y}^2}$$  \hspace{1cm} (31)$$

which, in turn, can be written in terms of the angles of each of the segments preceding the $n^{th}$. 
\[
\sum_{m=0}^{n} \frac{L}{N} \sin \phi_m = l_{n,x}
\]

\[
\sum_{m=0}^{n} \frac{L}{N} \cos \phi_m = l_{n,y}
\]

Finally, the angles \( \beta_n \) must also be rewritten in terms of the \( \phi_n \). This can be accomplished by noting that, by a trigonometric identity,

\[
\cos(\psi' - \beta_n) = \cos \psi' \cos \beta_n + \sin \psi' \sin \beta_n
\]

(34)

The terms containing \( \beta_n \) can be rewritten in terms of \( l_n \) and its components, (see Figure 30)

\[
\cos \beta_n = \frac{l_{n,y}}{l_n}
\]

(35)

\[
\sin \beta_n = \frac{l_{n,x}}{l_n}
\]

(36)

which may, in turn, be rewritten in terms of the angles \( \phi_n \), as shown above.

Finally, putting it all together, we can write down an expression for the energy of a magnetic rod of arbitrary shape (as defined by the \( \phi_n \)) in an applied magnetic field with a constant field gradient:

\[
U_A = \frac{\pi r^2}{N} \sum_{m=0}^{N} M (B_n) f_n B_n \cos(\alpha_n - \psi)
\]

(37)

where \( B_n = B_0 + \nabla B \left( \cos \psi \sum_{m=0}^{n} \frac{L}{N} \cos \phi_m + \sin \psi \sum_{m=0}^{n} \frac{L}{N} \sin \phi_m \right) \)

(38)
4.3.2.2 Internal Energy

In the continuous model, we found that the energy associated with each magnetic
dipole of the rod in the magnetic field of the remaining dipoles, the internal energy, could be
written as follows:

\[ U_I = -\frac{1}{4} \mu_0 \frac{m^2}{V} \cos^2(\alpha - \phi) \quad (39) \]

where we have reinstated the angle of the magnetic moment, \( \alpha \), in place of the angle
of the magnetic field, \( \psi \), for generality. (See Appendix A for details on the conditions under
which \( \alpha = \psi \).) Restructuring this as a summation over \( N \) segments of a rod, each with a
magnetic moment \( m_n \), and oriented at an angle \( \phi_n \) from the vertical, the energy term becomes

\[ U_I = -\frac{\mu_0}{4V} \sum_{n=0}^{N} m_n^2 \cos^2(\alpha_n - \phi_n) \quad (40) \]

Recalling that \( m_n = M (B_n) f_n V \), the energy term becomes

\[ U_I = -\frac{\mu_0}{4} \pi r^2 L \sum_{n=0}^{N} M^2 (B_n) f_n^2 \cos^2(\alpha_n - \phi_n) \quad (41) \]

Where \( B_n \) is as described above.

The total energy of the rod in the discretized model is then

\[ U = \frac{\pi}{2} N r^4 \sum_{n=0}^{N} E_n \cos^2\left(\frac{\pi}{2} + \phi_n - \phi_{n+1}\right) + \frac{\pi r^2 L}{N} \sum_{n=0}^{N} M (B_n) f_n B_n \cos(\alpha_n - \psi)L \]

\[ -\frac{\mu_0}{4} \pi r^2 L \sum_{n=0}^{N} M^2 (B_n) f_n^2 \cos^2(\alpha_n - \phi_n) \quad (42) \]

where

\[ B_n = B_0 + \nabla B \left( \cos \psi \sum_{m=0}^{n} \frac{L}{N} \cos \phi_m + \sin \psi \sum_{m=0}^{n} \frac{L}{N} \sin \phi_m \right) \quad (43) \]
The energy can then be minimized with respect to the angles $\phi_n$, using a minimization routine in a numerical package such as MatLab.

4.3.3 Torque Minimization

It is possible to run a minimization routine on the total energy expression in order to find the position of the rod (the vector $\phi_n$) which minimizes the total energy. This requires, however, that the expression be minimized with respect to $N$ variables, where $N$ is often in the range of tens to hundreds of segments. (In addition, if the magnetization vectors $\alpha_n$ are not assumed to align with the magnetic field (see Appendix A for more on this), the number of variables becomes $2N$.) This is computationally intensive, it is difficult to ensure that the solution represents a true minimum, rather than a metastable state. A much faster technique is to find the torques applied to the rod in the energy space of Equation 42, and allow the rod to move in order to minimize torque. In either case – energy minimization or torque minimization – the rod position is changed ultimately to minimize the quantity in question. However, this movement occurs randomly for each segment of the rod in the energy minimization routine; in torque minimization, the movement of the rod is directed explicitly towards lower torque, allowing the segments of the rod to move quickly and in concert towards a solution. Implementing a torque minimization routine in MatLab shortened solution times by an order of magnitude.
4.3.3.1 Elastic Torque

The elastic torque acting on the \( n \text{th} \) segment of the rod is just the negative of the derivative of the \( n \text{th} \) term of the elastic energy (Equation 27 or the first term of Equation 42) with respect to the angle \( \phi_n \), or

\[
\tau_{E(n,n+1)} = -\frac{dU_{E,n}}{d\phi_n} = \frac{\pi r^4}{4} \frac{N}{L} E_n \sin(\phi_{n+1} - \phi_n)
\]  

(44)

where we have used the double-angle identity \( \sin(2A) = 2\sin(A)\cos(A) \). This is the elastic torque that the \((n+1)\)\textsuperscript{th} segment of the rod exerts on the \( n \text{th} \). It follows that the \((n-1)\)\textsuperscript{th} segment also exerts a torque on the \( n \text{th} \). This torque can be found by making the transformations \( n \rightarrow n - 1 \) and \( n + 1 \rightarrow n \), and takes this form:

\[
\tau_{E(n,n-1)} = \frac{\pi r^4}{4} \frac{N}{L} E_n \sin(\phi_{n-1} - \phi_n)
\]  

(45)

(note that reordering the subscripts on \( \tau \) introduces an additional factor of -1. We have also assumed for simplicity that the Young’s modulus varies slowly, and have left the subscript on \( E \).) The total torque on each segment due to elasticity is then

\[
\tau_{E,n} = \frac{\pi r^4}{4} \frac{N}{L} E_n \left[ \sin(\phi_{n+1} - \phi_n) + \sin(\phi_{n-1} - \phi_n) \right]
\]  

(46)

4.3.3.2 Torque due to Applied Field

Similarly, the torque due to the applied field is the negative of the derivative of the energy due to the applied field (Equation 37 or the second term of Equation 42) with respect to \( \phi_n \). In this case, the dependence on \( \phi_n \) is through the magnetic field at the position of the \( n \text{th} \) segment, \( B_n \). The torque on the \( n \text{th} \) element becomes:
\[ \tau_{An} = -\frac{dU_{An}}{d\phi_n} = \frac{\pi L r^2}{N} \left[ \frac{dM(B_n)}{d\phi_n} B_n + \frac{dB_n}{d\phi_n} M(B_n) \right] f_n \cos(\alpha_n - \psi) \]  

(47)

Recognizing that

\[ \frac{dM(B)}{d\phi} = \frac{dM(B)}{dB} \frac{dB}{d\phi} \]  

(48)

the torque becomes

\[ \tau_{An} = \frac{\pi L r^2}{N} \left[ \frac{dM(B_n)}{dB_n} B_n + M(B_n) \right] \frac{dB_n}{d\phi_n} f_n \cos(\alpha_n - \psi) \]  

(49)

In order to find the derivative of \( B_n \) with respect to \( \phi_n \), it is helpful to rewrite Equation 43 as follows

\[ B_n = B_0 + \nabla B \left[ \cos \psi' \sum_{0}^{n-1} \frac{L}{N} \cos \phi_m + \cos \psi' \cos \phi_n + \sin \psi' \sum_{0}^{n-1} \frac{L}{N} \sin \phi_m + \sin \psi' \sin \phi_n \right] \]  

(50)

It is then evident that the derivative is

\[ \frac{dB_n}{d\phi_n} = \nabla B \frac{L}{N} \left[ \sin \psi' \cos \phi_n - \cos \psi' \sin \phi_n \right] \]  

(51)

or, simplifying:

\[ \frac{dB_n}{d\phi_n} = \nabla B \frac{L}{N} \sin (\psi' - \phi_n) \]  

(52)

Recognizing that the torque represents the change in energy with respect to the rotation of the \( n^{th} \) element, and that a rotation of the \( n^{th} \) element necessarily translates each of the following elements to an area of lower field, we replace the length \( L \) with the effective length equal to the portion of the rod above the \( n^{th} \) element, or \( L \left( 1 - n/N \right) \).

Finally, the torque due to the applied field can be written as
The derivative of the magnetization $M(B_n)$ with respect to field $B_n$ can be calculated numerically from the experimental magnetization curve.

4.3.3.3 Torque due to Internal Energy

Finally, the torque due to the internal magnetic energy of the rod can be found by taking the negative of the derivative of the internal energy (Equation 41 or the third term of Equation 42) with respect to $\phi_n$. As before, there is a dependence on $\phi_n$ through the magnetic field $B_n$. Performing the derivative in a manner similar to that in the previous section, the torque on the $n^{th}$ segment of the rod due to the internal magnetic energy becomes

$$\tau_{in} = \frac{\mu_0}{2} \frac{\pi r^2}{N} f_n \int \left[ M(B_n) \nabla B - \frac{L}{N} \frac{dM(B_n)}{dB_n} \sin(\phi_n - \psi) \cos(\alpha_n - \psi) + L \right] M^2(B_n) \cos(\alpha_n - \phi_n) \sin(\alpha_n - \phi_n)$$  \hspace{1cm} (54)

Here, the second term represents the torque caused by the misalignment of the rod from the magnetic field. The first term represents the potential decrease in internal magnetic energy should bending of the rod segment introduce the segment to a region of lesser field magnitude (i.e. in a direction opposite the gradient). This is a second order term, and can be ignored as long as the ratio of the coefficients of the first term to the second is significantly less than one (Equation 55)

$$\frac{L}{N} \frac{\nabla B}{M(B_n)} \frac{dM(B_n)}{dB_n} \ll 1$$  \hspace{1cm} (55)
Generally, this condition would not hold only for a very large magnetic field gradient or a very long rod. In the case of experiments presented in this work, $\frac{L}{N}$ is on the order of one micron. An upper bound for the magnetic field gradient is about 1000 T/m, the magnetization of the magnetic material in the rods (magnetite or maghemite) is on the order of 100,000 A/m, and the slope of the magnetization curve at its greatest is on the order of $10^6$ A/mT (see Figure 3). These values yield a value of 0.01, which is sufficiently less than one.

Finally, the angles of the magnetic moments of each section of the rod, $\alpha_n$, can be replaced with the angle of the magnetic field, $\psi$, under the condition that

$$4B/[\mu_0 M(B)f] \gg 1,$$

which is satisfied for the experiments described in this work. (See Appendix A for details) The total torque on the $n^{th}$ segment of the rod can then be written as the sum of Equations 46, 53, and 54:

$$\tau_n = \frac{\pi r^4 N}{4L} E_n \left[ \sin(\phi_{n+1} - \phi_n) + \sin(\phi_{n-1} - \phi_n) \right]$$

$$+ \nabla B \frac{\pi L^2 r^2}{N^2} f_n \left( 1 - \frac{n}{N} \right) \left[ B_n \frac{dM(B_n)}{dB_n} + M(B_n) \right] \sin(\psi - \phi_n)$$

$$+ \frac{\mu_0 \pi r^2 L}{2N} f_n^2 M(B_n) \cos(\psi - \phi_n) \sin(\psi - \phi_n)$$

(56)

In this final form it is easy to see that the first term represents the elastic torque, the second represents the torque due to the gradient, and the third represents the torque due to the field. The angle definitions from Figure 22 are reproduced below for convenience (Figure 31).
Figure 31. Angle and vector definitions used in deriving the rod bending theory. \( B \) is the applied magnetic field, \( \nabla B \) is the applied field gradient, and \( m \) is the rod’s magnetic moment.

4.3.3.4 Implementation of Torque Minimization

As mentioned previously, the great advantage of torque minimization over energy minimization is the directionality implicit in the former – knowing the energy of a rod in a given position tells only the energy; knowing the torque on a rod in a given position tells which direction the rod should move to find a position of lower energy. In order to achieve movement towards lower energy, we will let the rod move in a roughly physical manner in response to the calculated torque. Knowing the torque on the \( n^{\text{th}} \) element, we can calculate a rotation given a small time step \( t \), using simple Newtonian physics.
\[ \phi_{n,1} = \phi_{n,0} + \frac{1}{2} \frac{\tau_n}{I_n} t^2 \]  

(57)

Here \( I_n \) is the moment of inertia of the \( n^{th} \) segment, which is

\[ I_n = \frac{1}{4} m_n r^2 + \frac{1}{12} m_n \frac{L^2}{N^2}, \]

where \( m_n \) is the mass of the \( n^{th} \) segment. I have written a routine in MatLab which, beginning with an initial rod position vector, \( \phi_{n,0} \), will calculate the torque \( \tau_n \) and subsequently the new position vector \( \phi_{n,1} \) which represents the position of the rod after the time step \( t \). The position vector \( \phi_{n,1} \) then becomes the initial position for the second iteration, and so on, until the total torque on the rod falls below some threshold value which represents a zero torque.

It is important to note that the time evolution of the rod as represented above assumes that each segment of the rod is allowed to rotate freely about its centroid in a given torque. (It also does not carry any momentum from one iteration to another!) In reality, the movement of each segment is coupled to the movement of every other segment, representing a difficult and computationally intensive dynamic. The model presented in this chapter does not intend to accurately represent the dynamics of a bending rod, but only to quickly and accurately find the final, relaxed, energy minimum. The use of a physical relationship to calculate rod movement between iterations (Equation 57) is simply a convenience; in reality any appropriately small movement in the direction of the applied torque would suffice.
4.4 Conclusions

I have presented in this chapter two models for determining the bending of a cantilevered magnetic rod in a magnetic field. The first provides very useful expressions for drawing conclusions to inform the design of biomimetic cilia and their actuating field geometries, and the second serves to quickly and accurately calculate bend shapes. This will be useful in the future as we develop more advanced field generation mechanisms such as those described in the next chapter, which may allow us to come closer to replicating the complex bend shapes of certain biological cilia, such as human airway epithelial cilia.

Since the continuous model assumes a bend shape while the discrete model predicts bend shape, one would expect the differences in predictions between the two models to be the greatest when the difference in the shape would result in the greatest error in calculating energies. This would occur for field geometries with very large spatial variation in field strength – geometries with small fields and large gradients. In the next chapter, I will discuss the case of the field produced by a current-carrying wire, which is one such case in which the change in field over the length of the cilia due to the gradient is relatively large compared to the field magnitude. The predictions of the discrete model for this case are represented below, in Figure 32, along with predictions from the continuous model. In addition, I have included plots of the absolute and relative differences in angle predictions from the two models.

It is clear that both models show the same gross behavior, but the continuous model does incur significant errors in certain areas of the parameter space. This is one geometry, therefore, in which it is advantageous to use the more accurate discrete model.
Figure 32. Comparison between the discrete and continuous models for the case of a hydrogel silium \((f = 0.17, E = 0.4 \text{ MPa}, L = 10 \text{ microns})\) a distance above a wire carrying 1 A. This is a case of extreme gradient, when the error of the continuous model would be expected to be at its greatest. All contour labels are in degrees, except for in the lower right figure, where they indicate relative magnitudes.
Chapter V

ACTUATION

5.1 Introduction

The models presented in the previous chapter detail the bending we can expect from a given applied magnetic field. It says nothing, however, about what sorts of magnetic fields we can reasonably generate. This chapter will describe the types of magnetic fields which are experimentally feasible, and will explore possibilities for using these fields to actuate arrays of silia. For the first time, we will have a comprehensive view of both the range of materials available and the fields available to actuate them, tied together with the predictive model described in Chapter IV. This will allow us to design complete actuation systems to obtain the bend geometries we would like to produce.

Whether cilium or silium, any type of actuation results from a delicate balance of energies. In the case of biomimetic silia, the actuating energy is provided by a magnetic field; in the case of biological cilia, by the hydrolysis of ATP. This chapter will begin by exploring the energy considerations of both biological cilia and biomimetic silia, and observe how they differ. In addition, we will show how a simple energy model may be used to predict a novel feature of a biological cilium beating under a load, which has been experimentally observed by other members of the group.\textsuperscript{86}

Since the actuating energy of biomimetic silia comes from an applied magnetic field, we will then explore the various options for producing magnetic fields in the lab: a current-
carrying wire, an electromagnet, and a permanent magnet. I will explore the possibilities each holds for actuating silia. Finally, I will describe some biologically-relevant beat patterns which are predicted by the model, and show experimental evidence of these patterns in our biomimetic system. I will also discuss a novel feature of one of these patterns – the snap beat – which is predicted by the model and verified by experiment, and may have application in producing fluid flows in viscoelastic materials such as mucus.

5.2 Energy Considerations for Silia and Cilia

Both biological cilia and biomimetic silia actuate by way of a careful balance of energy input and energy output. Output energies are similar in the two types: each gives up energy to elastic stresses within the structure and to viscous drag forces in the fluid. The energy input is of an entirely different nature, however. In the case of a biological cilium, it comes in the form of chemical energy carried by adenosine triphosphate (ATP), and in the case of the biomimetic cilium, it comes in the form of a magnetic field. We will begin by exploring the output energies of both types of cilium, elastic and drag, and then take a careful look at the energy supply.

5.2.1 Elastic Energy

The elastic energy of a bent beam, as shown in Equation 3, is given by

$$ U_e = \frac{1}{2} EI \int_0^L \frac{1}{R(s)^2} ds $$

where $EI$ is the flexural rigidity and $R$ is the radius of curvature, which in general is variable along the length of the beam. The flexural rigidity of a cilium is well known, and three-
point bending experiments\textsuperscript{87} show it to be about $7 \times 10^{-22}$ N m$^2$. Assuming a constant curvature, we can rewrite this energy in terms of the amplitude of the ciliary beat, $A$:

$$U_\varepsilon = \frac{E I}{2L^3} A^2$$

(59)

A typical amplitude for a 7 µm long airway epithelial cilia is about 7 µm, yielding a total elastic energy in a cilium at the end of its effective stroke of about $1 \times 10^{-16}$ J. This value is in good agreement with values given for cilia of \textit{sabellaria} and \textit{mytilus}.

In contrast, the elastic energy of a hydrogel silia of the same proportions of a human airway cilia (7 microns in length, 200 nm in diameter) will have an elastic energy of about $1 \times 10^{-17}$ J when bent through a similar angle, and a silicone silia will have an energy of about 6 $\times 10^{-17}$ J. These results come directly from Equation 4, where we have assumed values for the modulus $E$ of the hydrogel and silicone silia of 0.5 MPa and 2.5 MPa, respectively. It is interesting to note here that, while the Young’s modulus of the hydrogel is four orders of magnitude less than that of a single microtubule (~2 GPa)\textsuperscript{87} and the cilium and the hydrogel rod are identical in size, the complex inner structure of the cilium results in a small enough bending moment of inertia, $I$, to nearly make up the difference.

5.2.2 Drag Energy

According to work by Sleigh and Holwill,\textsuperscript{88} the energy lost to viscous drag during the effective stroke of a cilium is

$$U_D = \frac{1}{3} C_D \omega^2 L^3 t$$

(60)
where \( \omega \) is the angular velocity of the cilium, \( L \) is the length of the cilium, \( t \) is the
time over which it actuates, and \( C_N \) is the drag coefficient per unit length for flow normal to a
cylinder, given by\(^{89}\)

\[
C_N = \frac{4 \pi \eta}{\ln \left( \frac{2L}{d} \right) - \frac{1}{2}}
\]  

(61)

For a 7 micron cilia of diameter 200 nm in water \((\eta = 8.9 \times 10^{-4} \text{ Pa s})\), the drag
coefficient is 0.0015. The period of the effective stroke is about 33 ms\(^{90}\) and the angular
velocity is can be found by dividing the angle subtended by the full stroke by the 33 ms: \( \omega = 60 \text{ rad/s} \). The drag energy is therefore \( U_D = 2 \times 10^{-17} \text{ J} \) – an order of magnitude less than the
elastic energy of the cilium.

We will take the energy lost to viscous drag to be the same for the hydrogel and
silicone silia, since they are of the same dimensions and describes a similar stroke.

5.2.3 Available Energy

The question remains of how much energy is available to either of the actuators. In
the case of the airway cilia, this energy comes in the form of chemical energy from the
hydrolysis of ATP; in the case of the hydrogel silia, it comes directly from the magnetic field.

While there is a certain amount of chemical energy available to a cilium via the
hydrolysis of ATP, it may be that only a fraction of this energy is converted into mechanical
energy. Given a stall force of a single dynein of 6 pN\(^{91}\) and a step size of 8 nm\(^{90}\) the
mechanical energy of a single dynein step cannot be more than \( 4.8 \times 10^{-20} \text{ J} \), which is about
half of the chemical energy available from the hydrolysis of a single ATP. We can assume,
therefore, an efficiency in converting chemical energy to mechanical energy of about one
half. The total mechanical energy expected from a single dynein during the effective stroke of a cillum is then \( U_D = e_{\text{ATP}} E_{\text{ATP}} k_{\text{ATP}} t_B \), where \( e_{\text{ATP}} = 0.5 \) is the conversion efficiency, \( E_{\text{ATP}} = 1 \times 10^{-19} \text{J} \) is the chemical energy per ATP (ref), \( k_{\text{ATP}} \) is the rate of hydrolysis of ATP per dynein, which may be as high as \( 120 \text{s}^{-1} \), and \( t_B \) is the duration of the effective stroke (about 33 ms).

The number of dynein which participate in the effective stroke of the cillum is a topic of discussion. However, most sources cite approximately 2000 outer arm dynein as the responsible motors in a 7 micron cillum, or 2.9 x 10^8 dynein per unit length. Not all of these dynein are oriented to apply a maximum force in a ciliary stroke and many sources assume that about \( \frac{1}{2} \) are ‘active’ in a particular event. By geometrical considerations, we can refine this ratio of active dynein to 0.64. The number of dynein involved in the ciliary beat is then \( N_D = R_D n_0 L \), where \( R_D = 0.64 \) is the fraction of dynein which are active, \( n_0 = 2.9 \times 10^8 \) is the number of dynein per unit length, and \( L = 7 \) microns is the length of the cillum. Then the total mechanical energy available to the cillum via hydrolysis of ATP is then the number of dynein multiplied by the energy per dynein:

\[
U_M = R_D n_D L e_{\text{ATP}} E_{\text{ATP}} k_{\text{ATP}} t_B \tag{62}
\]

Inserting the values quoted above, the total amount of chemical energy which is available for conversion to mechanical energy is about 2.6 x 10^{-16} J. This is significantly larger than the sum of elastic and drag energies.

\[\text{Given that the microtubule doublets are spaced every 40º (or } 2\pi/9 \text{ radians) around the cillum, and their component in the direction of the effective stroke is proportional to the cosine of their angle, the effective number of dynein can be written as } \bar{N}_D = \frac{N_D}{9} \sum_{n=0}^{8} \cos \left( \frac{2\pi n}{9} \right) = 0.64N_D\]
The energy available to a hydrogel silia is in the form of magnetic energy, stored in the magnetic field surrounding the silium. Assuming that the bending of the silium is due to only magnetic field (and not field gradient), then Equation 20 gives the magnetization per unit volume (A/m) to bend the rod through an angle $\phi$. In general, the mapping from magnetization $M$ to field $B$ is given by the magnetization curve for the material. However, for a hydrogel of dimensions similar to those of airway cilia, bent through an angle of 60 degrees (as is maximal for epithelial cilia), the magnetization required falls well within the linear range of the curve, where the slope $\chi = 2.8 \times 10^6$ A/Tm. Therefore we can say that $M = \chi B$.

The energy contained in a magnetic field, per unit volume, is $u_B = \frac{1}{2\mu_0} B^2$. The energy per unit volume in the magnetic field required to actuate the silium is therefore

$$u_M = 2 \frac{r^2 E}{L^2 \chi f^2 \mu_0^2} \phi^4$$  \hspace{1cm} (63)

For a hydrogel silia of typical Young’s modulus (0.5 MPa) and a reasonable loading of 10% maghemite by volume, the energy density is about 1,600 J/m$^3$. Assuming the same density (silia/area) as biological cilia, the spacing between silia is on the order of about 1 micron. The volume surrounding each individual silium can therefore be taken to be that of a box 1 micron by 1 micron by 7 microns, or $7 \times 10^{-18}$ m$^3$, yielding a total magnetic energy per silium of about $1 \times 10^{-14}$ J. This value is three orders of magnitude larger than the elastic energy. Assuming the same magnetic loading (10% v., which is reasonable for complexed materials such as FFPDMS-NH2) for a silicone silia, yields a value of $5 \times 10^{-14}$ J.

These energies are summarized in the following table:
Table 4: Energy Summary for Cilia and Silia

<table>
<thead>
<tr>
<th></th>
<th>$U_{\text{INPUT}}$ ($10^{-17}$ J)</th>
<th>$U_{\text{ELASTIC}}$ ($10^{-17}$ J)</th>
<th>$U_{\text{DRAG}}$ ($10^{-17}$ J)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cilia</td>
<td>26</td>
<td>10</td>
<td>2</td>
</tr>
<tr>
<td>Hydrogel Silia</td>
<td>1000</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Silicone Silia</td>
<td>5000</td>
<td>6</td>
<td>2</td>
</tr>
</tbody>
</table>

5.2.4 Discussion

One very interesting thing about this is that there is about $2 \times 10^{-16}$ J of available chemical energy that is unused in a typical stroke of a biological cilium. This is energy which may be available to do extra work, such as apply an additional force on the tip, in order to overcome an additional load. If such a load were applied to the tip of a biological cilium, it would be reasonable to assume that the cilium is capable of applying all of the available chemical energy to the elastic energy of the bent cilium, $U_E$, and work done on the applied load, $W_L$. Here, we are assuming that the drag force is negligible.

\[ U_M = U_E + W_L \]  

(64)

The work done on the load, $W_L$, is simply the magnitude of the force exerted by the load, $F_L$, multiplied by the amplitude of the beat, or $W_L = F_L A$. Inserting this expression and the expression for elastic energy (Equation 59) into Equation 64 and solving for the amplitude $A$ yields,

\[ A = \frac{L^3}{E I} \left[ \sqrt{F_L^2 + \frac{2}{L} E I U_M} - F_L \right] \]  

(65)

where $U_M = R_D \eta_D L e_{\text{ATP}} E_{\text{ATP}} k_{\text{ATP}} t_b$
Figure 33 shows the expected amplitude as a function of the applied load. The amplitude has been normalized, assuming an original, unloaded, amplitude of 7 microns.

![Cilium with a Force Applied at Tip](image)

Figure 33. Cilia beat amplitude as a function of force applied at the tip. The amplitude has been normalized, assuming an initial unloaded amplitude of 7 microns. During the flat portion of the curve, the cilium does not use all available energy; at the turning point (about 30 pN), the cilium has used all available energy, and further work done on the load must come at the expense of elastic energy, and hence amplitude. The grey box represents the amount of initially unused chemical energy, to within a normalization factor.

The curved line (continued in dashes in the upper left) indicates the amplitude expected if all of the energy available from ATP is used. However, under normal operation the cilia work below capacity, as shown by the flat line at a relative amplitude of one. This allows some reserve capacity for handling additional loads. In the plot, the reserve energy is tapped at about 30 pN, and additional efforts come at the expense of elastic energy, and hence, amplitude. Aside from the normalization of the beat amplitude, the total excess chemical energy is the area underneath the flat portion of the curve.
Unpublished work by Swaminathan and Hill has shown a similar change in ciliary beat amplitude upon loading with a magnetic bead.\textsuperscript{86} Their data is shown in Figure 34 below, with the previous figure (Figure 33) overlaid.

![Figure 34. Ciliary beat amplitude as a function of force applied at the tip. In the experimental data, the force was applied via a magnetic bead attached to the cilium.](image)

5.2.5 Conclusions

A couple of useful pieces of information can be deduced from this plot. First, experiments by Swaminathan and Hill\textsuperscript{86} have indicated that the terminus of the excursion of the cilium in the direction of the magnetic force remains relatively constant as force is increased; the decrease in the amplitude occurs on the opposite side of the beat. When the
cilium reaches $\frac{1}{2}$ of its original amplitude, then, it is upright and elastic forces are negligible. At this point all of the force exerted by the cilium goes to balance the force exerted by the magnetic bead. The maximum force that the cilium can generate can therefore be taken as the applied force which causes the cilium to beat at $\frac{1}{2}$ its original amplitude. From the model, the point at which the relative amplitude is 0.5 corresponds to a maximum force of about 69 pN. This is in good agreement with the value of 62 pN given by Hill.\textsuperscript{86} In more general terms, we can find an expression for the maximum force generated by a cilium by setting $A = 0.5 A_0$ (where $A_0$ is the initial, unloaded amplitude) and solving Equation 64 for the force, $F_L$. This gives a value of

$$F_{\text{MAX}} = 2 \frac{U_M}{A_0} - \frac{1}{4} \frac{A_0EI}{L^3}$$

(66)

for the maximum force a cilium can exert. For a cilium in which the available mechanical energy $U_M$ is significantly greater than the elastic energy at full deflection – which we would expect to be the case for all biological cilia – the second term is much less than the first, and the maximum force may be written as:

$$F_{\text{MAX}} \approx 2 \frac{U_M}{A_0}$$

(67)

where $U_M$ is as described in Equation 62.

In addition, the turn-around point at which a cilium begins to decrease amplitude (about 30 pN in Figure 33) tells us something about the total chemical energy which is able to be converted to mechanical energy. Values quoted for essential components of this energy, such as the rate of ATP hydrolysis, $k_{\text{ATP}}$ and the number of dynein which participate in the effective stroke per unit length, $n_D$, vary widely in the literature. Experimental
determination of this turn around point may be able to put bounds on the product of these critical values.

5.3 Field Generating Mechanisms

Biomimetic silia are actuated by magnetic fields, and there are as many possibilities for magnetically actuating these nanorod arrays as there are ways to produce magnetic fields: three. Magnetic field may be generated directly from a current-carrying wire, from a permanent magnet, or from an electromagnet (Figure 35). Each of these methods holds its own set of advantages and disadvantages for use in the actuation of our biomimetic cilia samples.

![Figure 35. Methods of Producing Magnetic Fields. A: a permanent magnet; B: a current-carrying wire; C: an electromagnet.](image)
5.3.1 Current-Carrying Wires

The magnetic field generated by an infinite, straight current-carrying wire is oriented tangential to a circle centered about the wire (Figure 35, B) and is given by

$$ B = B_\theta = \frac{\mu_0 I}{2\pi r} $$

where $I$ is the current through the wire, $\mu_0$ is the permeability of free space, and $r$ is the radial distance from the wire. The closest to the nanorod array that a wire could be placed is a distance on the order of the rod length, or about 10 microns, and the largest current which could feasibly be carried by such a wire is on the order of one ampere (see Appendix C). Under these most favorable conditions, the wire would produce a field of 0.02 Tesla at the rod array. As shown in Figure 36, this field is far too small to actuate the current 1.4% v. FFPDMS, represented by the red spot, but might easily actuate lower-modulus silicone rods with magnetic loadings of about 10% v. – such as FFPDMS-NH2, or a typical hydrogel rod.
Figure 36. The field required to bend a rod of radius 125 nm, length 25 µm, and a range of magnetic loading (f) and Young's moduli (E), given the condition of pure field bending represented by Equation 17. The blue and green boxes represent reasonable ranges of material properties for hydrogel-based microrods and silicone-based microrods, respectively, and the red spot indicates the material properties of FFPDMS-C16.

The magnetic field magnitude is only half the picture, however – the magnetic field gradient also plays a strong role, and calculations of the gradient in this field generating geometry show that the current-carrying wire places us in a very interesting piece of the parameter space. The magnetic field gradient from a current-carrying wire would be oriented along the radius of the wire, and is found by taking the derivative of the scalar field, with respect to \( r \):

\[
\nabla_B = -\frac{\mu_0 |I|}{4\pi r^2}
\]
At one ampere and a distance of 10 microns, the gradient produced by the current-carrying wire is on the order of 1000 T/m. A magnetic field of 0.02 T and a gradient of 1000 T/m places the FFPDMS silia firmly within the gradient-dominated area of the parameter space, as shown in Figure 25, and Figure 23 indicates that a gradient of 1000 T/m is more than enough to actuate a FFPDMS silia. However, the gradient in this case is not optimally oriented – in fact, it is oriented in such a way as to provide almost no torque to the silium: directly downward along the length of the structure. The only way the gradient can actuate the silium is if the field is able to bend it far enough from equilibrium for the gradient to exert a non-negligible torque. The field, however, is far too weak, and calculations of the bending of a 1.4% v. FFPDMS silia, 20 microns in length and over a wide range of radii, show negligible bending in this geometry. In all cases the bending is significantly less than one degree.

For a softer structure, however, actuation is very different. With a magnetic field of 0.02 T and a field gradient of 1000 T/m, Figure 26 indicates that a hydrogel silium is in a region of the parameter space in which the field and gradient effects are of the same order. In this case, the field is able to bend the silium far enough for the gradient to apply a significant torque, resulting in an increased angle from the combined efforts of field and field gradient.
Figure 37. Predicted bending angle of a typical hydrogel silium over a range of distances from a wire carrying 1 A. Angles are in degrees. The silium has a length of 10 microns, a Young's modulus of 0.4 MPa, and a magnetic loading of 17% v. This plot was calculated using the discretized torque model.

This is a particularly interesting case of bending because it illustrates the utility of the gradient term in the bending model. A model without a gradient term under-predicts the bend in this case by as much as 40-50%. Figure 37 shows the bend angle expected from a hydrogel silium of 17% v. loading, Young’s modulus 0.4 MPa, and length 10 microns over a range of radii and distance from the current carrying wire. Differences between the complete model and a constant field model over this same parameter space are shown in Figure 38.
Figure 38. The number of degrees by which a constant field model underpredicts the bending shown in the previous figure, in degrees. In the extreme cases, the constant-field model errs by nearly 50%.

In summary, it is unlikely we will ever achieve actuation of the current 1.4% v. FFPDMS-C16 silia with a current carrying wire. However, materials such as FFAAm and FFPDMS-NH2 show great potential in such a system.

5.3.2 Electromagnets

Another option for applying a magnetic field to a silia array is with an electromagnet. This would consist of a wire winding on a paramagnetic material, and as with the current-carrying wire, offers the advantage of varying field strength over time. Specially-engineered
paramagnetic materials such as mu-metal or permalloy are designed with extremely high magnetic susceptibilities, providing a large response to a relatively small actuating field. It is this material response, which amplifies the field applied to the material by orders of magnitude, that gives an electromagnet its greatest advantage over a current-carrying wire.

It is useful to think of the magnetic field produced by an electromagnet as existing within a magnetic circuit – in fact, there is a sense in which a magnetic circuit acts much like an electrical circuit (see Figure 39). A source of magnetic field – such as a permanent magnet, an electromagnet, or a current-carrying wire – is analogous to a voltage supply in an electric circuit. The material through which the magnetic field ‘travels’ is much like the wire conductor in an electrical circuit, with a ‘resistance’ inversely proportional to its permeability. An air gap, for example, has a very high ‘resistance’ due to its extremely low permeability; a superparamagnetic material has a very low ‘resistance’ due to its high permeability. The ‘resistance’ in a magnetic circuit is referred to as reluctance, and is defined as $\mathcal{R} = \frac{l}{\mu A}$, where $l$ and $A$ are the length and cross-sectional area of the material, respectively, and $\mu$ is its permeability.
To actuate a nanorod array via a magnetic field from an electromagnet, it is advantageous to have the highest field possible in the space occupied by the array. This can best be accomplished by providing a very small air gap (and therefore a low reluctance and a large flux) just underneath the array. Fortunately, this is the same goal for which commercial read/write heads have been designed, and so an electromagnet design which maximizes magnetic field in a small area has been years in the making. While it is true that current read/write heads use more exotic phenomena to communicate with the recording media, such as magnetoresistance, I refer here to older, inductive read write heads, such as the Karlqvist head, pictured below. When ‘writing’ to a magnetic tape or hard drive element, a write head must provide a relatively large magnetic field (fields may be as high as on the order of a
Tesla between the pole tips)\textsuperscript{94} to a very small area on the disk or tape (on the order of tens of microns on a side). Commercial read/write heads are therefore produced with a geometry similar to that described above, and pictured in Figure 40.

Figure 40. A Karlqvist read/write head. At right, a zoom-in of the tip of the head, showing the magnetic tips of the head and the magnetic recording media. Figure from Karlqvist, 1954.

The magnetic field produced by such a geometry is given by Karlqvist\textsuperscript{95}

\[
B_x = \frac{1}{\pi} B_0 \left( \arctan \left( \frac{N + x}{y} \right) + \arctan \left( \frac{N - x}{y} \right) \right), \quad B_y = \frac{1}{2\pi} B_0 \log \frac{y^2 + (N + x)^2}{y^2 + (N - x)^2},
\]

where \(B_0\) is the magnetic field in the pole gap and \(N\) is one-half the horizontal distance between the poles. Since the \(y\)-component of the field is much smaller than the \(x\)-component and, furthermore, is necessarily oriented such as to provide a minimal torque on the nanorods, the \(y\)-component will be ignored.

Letting the distance between the poles, \(2N\), equal an arbitrary value of 2, and setting \(B_0\) to 1, the relative magnitude of the magnetic field decays with distance from the pole tip as shown below. (Figure 41)
Figure 41. The relative magnitude of the magnetic field as a function of horizontal distance. Each curve represents a different vertical position with respect to the pole tip; the highest curve is at the surface of the pole, and the successively lower peaks represent distances of 0.5, 1, 2, 4, 6, 8, and 10 times the pole separation, respectively.

The field from the head falls off to about 10% of its maximum value at a distance of 10 times the pole separation. Our own attempts to actuate fields of siila with commercially available read/write heads have not met with success, and this is likely due to the thickness of the supporting coverslip in our sample geometry relative to the pole separation of the read/write heads (shown in Figure 11). Read/write heads may be a possibility for actuating in the future, should the restriction of the coverslip be circumvented.
5.3.3 Permanent Magnets

To date, permanent magnets show the most promise as an actuation mechanism for nanorod arrays. Neodymium iron boride (NIB) magnets are commercially available with field strengths of as much as 0.5 T on the pole face and, due to their relatively large size, the field falls off little over the distance by which they are separated from the nanorod array. Measurements of the magnetic field and magnetic field gradient as a function of distance were taken from several permanent NIB magnets, and the results are shown in Figure 42 and Figure 43.

Figure 42. Magnetic field from permanent neodymium iron boride magnets, in the symmetry plane perpendicular to the axis of the magnet. The dotted line represents the field from a large disk magnet (diameter 50 mm, thickness 6 mm), and the solid line represents the field from a small disk magnet (diameter 25 mm, thickness 6 mm) Zero distance indicates the edge of the magnet.
Magnetic Field Gradient from Permanent NIB Magnets

Figure 43. Magnetic field gradient from permanent neodymium iron boride magnets, in the symmetry plane perpendicular to the axis of the magnet. The dotted line represents the gradient from a large disk magnet (diameter 50 mm, thickness 6 mm), and the solid line represents the gradient from a small disk magnet (diameter 25 mm, thickness 6 mm). Zero distance indicates the edge of the magnet.

While the magnetic field gradient is large enough to actuate hydrogel silia and, potentially, future formulations of silicone silia, it is too small to effectively actuate the current FFPDMS-C16. However, the field magnitudes from the permanent magnets are more than enough to actuate most silicone silia, including FFPDMS-C16. We have experimentally demonstrated actuation of FFPDMS-C16 microrod arrays with these fields, and have been able to achieve a couple of interesting modes of actuation.
5.4 Modes of Actuation

5.4.1 Conical Orbit

By orienting a permanent magnet relative to a rod array as shown in Figure 44, the silia experience a field perpendicular to their relaxed vertical orientation. They are bent towards alignment with the field. When the magnet is rotated about its axis, the field rotates in the horizontal plane and the rod rotates with it (since the rod position which minimizes energy is always constant with respect to the magnet position) producing a conical orbit, as shown in Figure 45B. This figure also shows an experimental conical beat, which was produced by the magnet orientation shown in Figure 44.

Figure 44. Sample geometry for conical actuation of a silia array. At left, the silia array is positioned for optical observation via the overhead objective and magnetic actuation via the magnet underneath. The magnet is oriented in order to produce a field parallel to the array substrate, as shown at right.
Figure 45. Two prominent beat patterns for biomimetic silia. The first is linear (A), and is similar to that produced by airway epithelial cilia. The second is conical (B) and is similar to that produced by embryonic nodal cilia.

Figure 46, below, shows a larger-scale view of the actuation of an array.
Figure 46. Actuation of an array of 500 nm diameter silia with aspect ratios of 50. In (A), the silia array is pointing straight up, while in (B), (C), and (D), the arrows represent the direction of the applied magnetic field.

By laterally offsetting the magnet a few millimeters from the base of the silium, it is possible to cause the silium to beat in a tilted cone. The geometry of the magnetic field is similar to the condition in which the magnet is centered on the silium – it is still largely in the plane of the substrate – but the magnitude varies as the magnet rotates. This results in a variable bend angle in phase with the rotation of the magnet: a tilted cone. The tilted cone is of particular interest from a biological point of view, since it is understood to be the manner in which nodal cilia beat in the prechordal plate of the vertebrate embryo. This will be
discussed in detail in Chapter 6, “Fluid Flow.” Images of an array undergoing tilted-cone actuation are shown in Figure 47, below.

![Figure 47. A minimum intensity projection (in essence, a time-lapse image) of a movie of silia undergoing tilted-cone actuation. Cones are tilted toward the lower left of the figure. The inset shows overlaid minimum intensity projections from actuations in fields of different magnitudes, which result in a different overall bend angle.](image)

### 5.4.2 Planar Orbit

A second beat pattern can be realized by passing a permanent magnet just above the silia array, as shown in Figure 48. As the magnet approaches the microrod, the silia begins to tilt away the magnet in order to align with its dipolar field (toward the right in Figure 48).
The silia retains a rightward deflection shortly after the magnet passes directly above the rod position, at which point the field is positioned to favor alignment in the opposite direction of the initial bend (toward the left). At some point, it becomes energetically favorable for the silium to rapidly reorient itself to align with the current local orientation of the magnetic field, producing a quick beat in the leftward direction. All silia positions shown in Figure 48 are calculated with the Discretized Model, via torque minimization, described in detail in Chapter 4: Modeling. The end result is a beat located entirely within the x-z plane and consisting of half of a slow forward beat, a rapid backwards beat, and another slow half beat forwards. In the experiments described in this work, this pattern is repeated indefinitely by passing a series of magnets attached to a spinning disk across the silia array. One half of this linear beat pattern is shown experimentally in Figure 45A.

5.4.3 Snap Beat

One particularly fascinating feature of this planar orbit is the existence of the rapid return stroke shown in Figure 48. This feature is interesting both from a mechanical point of view, since this is the first report of such a phenomenon in flexible magnetic filaments, but also from a biological point of view: the temporal asymmetry in this beat pattern is very similar to that shown by beating human airway cilia, which show a quick active stroke and a slower return stroke. Understanding the mechanics behind the rapid return stroke (the ‘snap’) in the biomimetic model might be the first step toward accurately reproducing the beat patterns observed in biological cilia.
Figure 48. Quasi-static evolution of microrod position in the field from a passing magnetic dipole. As the dipole passes to the right, the rod bends to align with the magnetic field. A snap is visible just to the left of center, just after the magnet passes overhead the microrod. Details are in the text.

To understand what causes the snap beat, it is instructive to look at the energy space of the rod in the field produced by the permanent magnet shown in Figure 48. The Continuous Model described in Chapter 4 provides a convenient analytical expression for the energy of the rod (Equation 13). This can be further simplified by neglecting the gradient term (the third term of Equation 13) and assuming that the rod is magnetically saturated. For typical values of magnetic field and magnetic field gradient from the permanent magnets (about 0.1 T and 10 T/m, respectively), Figure 25 and Figure 26 clearly show that bending will be field dominated, justifying the neglect of the gradient energy term; the magnetization curve of maghemite in Figure 3 indicates that at these field values the maghemite will be largely saturated. Applying these assumptions yields
\[ U = \frac{\pi E r^4}{2 L} \phi^2 - \frac{1}{4} \mu_0 M_0 s^2 f^2 \pi r^2 L \cos^2 (\psi - \phi) \]  \hspace{1cm} (69)

where \( M_0 \) is the saturation magnetization of the magnetic material in the rod.

Figure 49 shows the energy and torque as functions of the angle of the rod for various magnetic field orientations. In the first row, the magnetic field is vertical, just as in the rightmost position in Figure 48. The energy space clearly and predictably shows a minimum at 0°, and the torque plot also indicates that this is a stable equilibrium, with restoring torques to the left and right of center. As the field rotates to 45° from the vertical, as in the right third of Figure 48, the energy minimum moves to the right, causing the rod to bend. At a field angle of 90° (the center of Figure 48), a second minimum is visible in a symmetric position to the left of the rod. The rod, however, will remain in the original minimum. Finally, when the field angle increases to 135° (left third of Figure 48, fourth row of Figure 49), the original minimum has disappeared, and the rod moves to the position indicated by the second minimum.
Figure 49. Energy space and torque for a microrod in a rotating magnetic field. The field begins upright ($\psi = 0^\circ$) and rotates clockwise to $\psi = 135^\circ$. In each case, the rod position is calculated from the Discretized Model, using the previous rod position as the initial position. A snap is clearly evident between the third and fourth rows.

In the quasi-static representation of the model, the transition from a rightward to a leftward bend is truly instantaneous – that is, there are no stable rod positions available within the jump. This is shown in Figure 50A, which plots the angle of the rod as a permanent magnet passes overhead; data in this plot is calculated with the Discretized Model, using a true dipolar field oriented as shown in Figure 48. The plot consists of 121 data points, and illustrates well the instantaneous jump from a right bend to a left bend. In experimentation, of course, the jump will never be instantaneous due to the presence of inertia and viscous drag. However, we have seen evidence of a snap-type beat pattern in
experiments with FFPDMS-C16 microrod arrays. Figure 50B shows the angle as a function of position relative to an overhead magnet for a FFPDMS-C16 microrod. The data clearly indicate a slow forward stroke followed by a quick return stroke, just as in the model.

Figure 50. A) the angle of the rod depicted in Figure 48 as a function of magnet displacement, as computed by the discretized model. This data is much higher in resolution than is indicated in Figure 48, exhibiting 120 data points over the same range of motion. B) depicts experimental measurements of rod angle as a function of magnet displacement, and shows a snapping behavior similar to that in the model.

Not every rod will produce a snap – the phenomenon is restricted only to rods with particular physical parameters. In fact, the snap can only happen in situations where there are two local minima in the energy space, as in the third line of Figure 49. (Recall that the snap occurs when the rod is ‘passed’ from one minimum to another.) An energy space with multiple minima requires that the derivative of the energy – the torque – have more than one zero. We can be sure that the torque has only one zero (and therefore eliminate snap) by requiring that it is monotonic. Finally, the torque is monotonic if the derivative of the torque has no zero. To within a sign, the derivative of the torque (or the second derivative of the energy, Equation 69) is
\[
\frac{d^2U}{d\phi^2} = \pi \frac{r^4E}{L} + \frac{1}{2} \mu_0 M_s^2 f^2 \pi r^2 L \cos[2(\phi - \psi)]
\]  

(70)

The expression is capable of multiple zeros through periodicity of the cosine term. However, the smallest the cosine can be is -1, and therefore the expression will have no zeros under the condition that

\[
\pi \frac{r^4E}{L} - \frac{1}{2} \mu_0 M_s^2 f^2 \pi r^2 L > 0
\]

(71)
or

\[
\frac{\mu_0 f^2 M_s^2(B)}{2E} \left(\frac{L}{r}\right)^2 < 1
\]

(72)

where we have replaced \(M_s\) with \(M(B)\) for generality. For lack of a better term, we will call this the “snap number” and denote it “S.” Equation 73 below summarizes this result.

\[
S = \frac{\mu_0 f^2 M_s^2(B)}{2E} \left(\frac{L}{r}\right)^2
\]

\[
S > 1 \Rightarrow \text{Snap}
\]

\[
S < 1 \Rightarrow \text{No Snap}
\]

(73)

Assuming complete magnetic saturation and a reasonable value for the Young’s modulus of the FFPDMS-C16 nanorods, Figure 51 below depicts the snap number over a range of possible aspect ratios and magnetic loadings. The magnetic loading fraction is allowed to range from the value of \(f = 0.0143\) determined from the fit to the continuous model in Chapter 4 up to \(f = 0.040\) as determined by SQUID measurement of bulk material.
Figure 51. Snap number for microrods with a Young's modulus of 2.6 MPa, over a range of magnetic loading $f$ and aspect ratio $L/r$. Microrods with $S > 1$ are predicted to exhibit a snap; those with $S < 1$ are not. The glyphs on the plot represent calculations from the discretized model, which are in good agreement with the snap number derivation from the continuous model. Open circles indicate a no-snap condition, open diamonds a snap condition.

The snapping behavior of our silia arrays is biologically relevant in that it mimics the quick effective stroke followed by the slower recovery stroke in biological cilia. This may be an essential component of inducing flow in a viscoelastic fluid, as I will discuss in the following chapter, “Fluid Flow.”
Chapter VI
FLUID FLOW

6.1 Introduction

While the novel, high-aspect-ratio microstructures – the silia – presented in this work may find many applications in many fields, the work is at heart a biomimetic endeavor. One of the driving goals of the project is to reproduce the beat patterns of biological cilia in order to gain a better understanding of the mechanisms behind the fluid flow that such patterns induce. I have presented two distinct beat patterns which we are able to replicate with our biomimetic system, and each of these patterns corresponds to a biological analog. The conical beat pattern is very similar to that found in cilia beating in the node of a developing embryo, which have been found to be responsible for establishing critical features of the embryo’s development. I will discuss nodal flow in the first half of this chapter. The planar beat pattern is similar to that found in cilia in the lung, which are a critical component of the body’s first line of defense against airborne pathogens. These cilia, responsible for mucociliary clearance in the human lung, will be discussed in the second half of the chapter.

6.2 Nodal Flow

Any coordinate system in three-space is made up of three axes: x, y, and z. However, as every good student knows, to simply place the axes at random orthogonal angles is to risk
defying convention and producing a left-handed coordinate system. Any two of the axes may be placed at random, but the third must be carefully oriented relative to the first two.

Just as a coordinate system in three space has \(x, y,\) and \(z,\) every vertebrate life form has three relevant axes – ventral/dorsal, anterior/posterior, and left/right. During embryogenesis, the orientations of the first two of these axes (V/D and A/P) are established first, and as in our mathematical coordinate system, they may be established randomly. The addition of the third axis, however, must be correctly oriented relative to the first two, or an embryo will develop with an inverted symmetry. This condition of inverted symmetry is called \(\text{situs inversus},\) and manifests itself in a left/right mirroring of the body’s organ systems. While much of our exterior is symmetric, and so \(\text{situs inversus}\) often goes undiagnosed, the inner workings of the body are not so: our heart is canted to one side, our lungs have three lobes on the right and two on the left, our liver is to the right, our stomach to the left, and so on.

While \(\text{situs inversus}\) by itself is may be asymptomatic, it is often associated with diseases of ciliary dysfunction. This led researchers to suspect a link between cilia and \(\text{situs inversus},\) and to explore its causes.

After the establishment of V/D and A/P axes, the vertebrate embryo contains a prechordal plate, which is a depressed region on the ventral side of the embryo (Figure 52 2A and D, indicated by the arrow.) The node is filled with ciliated cells, as shown in Figure 52 E. For many years, researchers believed these cilia to be immotile. Kathy Sulik, at UNC Chapel Hill, was the first to show by video microscopy that the cilia were motile, and to suggest that they may play a role in determining left/right asymmetry in the developing embryo.\(^2\)
Figure 52. Notochordal plate of a murine embryo. Each image is an enlargement of the boxed area of the previous image. Image E clearly shows ciliated cells, which are also present in the bowl of the node, indicated by the arrows in Image 2A and Image D. Images are from Sulik, 1994.

Since her work, Nonaka et al. have demonstrated that nodal cilia produce a leftward flow in the node, and demonstrated that knockout mice with dysfunctional nodal cilia exhibited randomized left-right asymmetry, strongly suggesting that a directed nodal flow is responsible for normal situs.\textsuperscript{96} Images of the nodal flow from the Nonaka paper are shown in Figure 53. Later, Okada et al. performed work which suggested that the leftward flow generated by nodal cilia is responsibly for establishing a chemical gradient which results in asymmetric gene expression, and leads to left-right asymmetry.\textsuperscript{97} Finally, Nonaka et al. showed that they could induce situs inversus in mouse embryos by artificially establishing a rightward flow across the node during development.\textsuperscript{98} This firmly established the leftward
flow generated by nodal cilia as the mechanism by which left-right asymmetry is broken in vertebrate embryos.

Figure 53. Fluid flow in the murine node. A shows traces of fluorescent bead aggregates from right to left across the node. Nodal boundaries are indicated by the dashed lines. B shows the path of a single tracer bead as it becomes trapped in vortices around individual cilia. C: Paths of individual tracer beads in a wild-type mouse, indicating leftward flow. D: Paths of individual tracer beads in a knockout mouse, with immotile cilia. Only Brownian motion is observed. Images are from Nonaka, 1998.

The question remained, however, as to how the nodal cilia generated flow at all. An array of cilia simply beating in a conical orbit may produce large scale vortices, but is
unlikely to produce large scale directional flow. In 2004, Cartwright et al. constructed a numerical model of the fluid flow generated by an array of cilia beating in a conical orbit, and showed that if the cones were upright, there would be no net flow across the field. However, they showed that tilted cones would produce a flow, and suggested that nodal cilia were beat about a tilted axis. The tilt of nodal cilia was later confirmed experimentally, and Cartwright’s model has since become the accepted description of the mechanism behind nodal flow.

In Cartwright’s model, all the cilia in the node are tilted toward the posterior, and beat with a clockwise beat as viewed from above. As each cilium beats toward the left along the highest part of the orbit (furthest from the floor), it moves fluid to the left. As it returns to the starting position, it passes toward the right very near the nodal floor. Boundary conditions restrict flow on this return stroke, resulting in an overall flow to the left.

Since we are able to reproduce the tilted cone orbit of nodal cilia with our biomimetic silia (as demonstrated in Chapter 5), this system serves as a perfect testbed for our biomimetic system, before exploring the more difficult issue of mucociliary clearance in the human lung. The nodal system is simpler than the human lung system for a few reasons: the fluid flow occurs in a low Reynolds number fluid without a significant elastic component, the beat pattern of the cilia is relatively simple, and the mechanism behind the generated flow is well understood. We have been able to reproduce the beat pattern in a purely viscous fluid, and have demonstrated that our system generates flows similar to those in biological systems. In addition, we have been able to reverse the direction of the flow by reversing the direction in which the silia beat (from clockwise to counterclockwise), something that cannot be done in a biological experiment. Some of this data is shown in Figure 54, and indicates a driven
flow of about 5 microns/second in the biomimetic sample at a driving frequency of 30 Hz. The beat frequencies and flow rates in biological nodes varies from species to species, but are generally in the range of 5-50 Hz and 1-10 microns/second, respectively.\textsuperscript{100}

\textbf{Figure 54.} Velocities of tracer particles in a FFPDMS-C16 silia sample. The array was actuated with an offset rotating magnet, producing a tilted cone orbit similar to that found in biological nodal cilia. The actuating frequency is about 30 Hz; the cones are tilted about 30° from the vertical. All bead traces are drift-subtracted.

While the global properties of fluid flow generated by nodal cilia are well understood, the smaller-scale properties of the flow are of interest as well. For example, heterogeneity of the flows at length scales on the order of the interciliary spacing may be an indication of chaotic mixing flows, which would be of great interest to the microfluidics community. Such
heterogeneity is suggested by our data; the wide divergence of bead traces in the 35-40 second timeframe of Figure 54 is a prime example. Efforts are underway to understand and quantify this chaotic property of the flow in these biomimetic experiments.

6.3 Mucociliary Clearance

The human lung is capable of adsorbing huge amounts of oxygen from the atmosphere – about 3.5 liters per minute for an average male during aerobic activity. This requires that they expose a very large surface area to air; in fact, the total surface area of an average set of human lungs is estimated to be about 75 m$^2$, or roughly the area of a tennis court. This large surface area provides ample opportunity for invasion by airborne pathogens and other debris, and so the lung has developed a defense mechanism in the form of cilia-mediated mucus clearance, or mucociliary clearance. Pathogens and debris are caught in a layer of viscoelastic mucus which lines the entirety of the lung, and the mucus is continuously moved up out of the lung to be ingested. The normal process of mucociliary clearance is depicted on the left in Figure 55, from Boucher, 2004. In this image, cilia are surrounded by a layer of liquid with minimal elasticity, the periciliary liquid (PCL). Their tips extend into the layer of viscoelastic mucus above, and as they beat they transport the mucus layer at a speed of about 60 microns/second.

Certain genetic defects can cause the dysfunction of this defense system. For example, a mutation in the gene responsible for a particular transmembrane protein can cause improper regulation of the thickness periciliary liquid, which results in a breakdown of mucociliary transport, as shown at right in Figure 55. This results in an increase in
bacterial growth within the lung and leads to lung disease and dramatically shortened lifespan. Manifestation of this genotype is referred to as cystic fibrosis (CF).

Figure 55. Depiction of mucociliary clearance in the lung. At left, in health lung tissue cilia move mucus along above a layer of periciliary liquid (PCL) at a rate of about 60 microns/second. At right, the PCL is absent due to a genetic defect and the mucus is not cleared. Image from Boucher, 2004.

While researchers and medical professionals have come a long way in understanding the cellular mechanics of cystic fibrosis – the mechanisms behind the regulation of the PCL, for example – very little is understood about the interaction between the cilia and the mucus layer itself. To understand the physical mechanisms behind the fluid flow produced in a healthy patient, and to understand why those mechanisms fail in a patient with CF would be invaluable to research on the disease.

The cilia-mucus system in epithelial lung tissue has several interesting characteristics which may or may not be directly involved in producing fluid flow. The cilia beat in a
pattern which is largely planar, which eliminates the flow-inducing mechanism of nodal cilia as a consideration. In this low-Reynolds regime, where flows are generally reversible, it is unclear how a planar beat is able to generate a net flow. However, the rheology of the mucus layer is also more complex than that of the fluid in the embryonic node: the mucus is viscoelastic, and many viscoelastic materials exhibit shear thinning behavior at certain strain rates. Practically speaking, this may mean that the viscosity of the fluid is dependent on the speed of the cilium.

While planar, the shape and speed of the beat is asymmetric in time. The cilia have a stiff, quick forward stroke in the direction of mucus flow, and a slower, whip-like return stroke. Combined with the possibility of a shear thinning fluid, this may result in a net fluid flow. We have taken some preliminary data consisting of tracer particle tracks in a viscoelastic fluid (5% polyvinyl alcohol) in which our silia perform the planar beat pattern described in Chapter 5 which suggests that this may be a viable mechanism. However, the role of the periciliary layer in the healthy patient, and the lack of mucociliary clearance in the absence of this layer are not well understood.

Our biomimetic model is perfectly poised to begin to answer fundamental questions of microscale fluid flow in a viscoelastic fluid, and we can begin to explore the full parameter space of the mucociliary clearance system in a way that has never been possible before. We have a fabrication procedure that will enable us to replicate the size and shape of biological cilia, we have materials and a mathematical model which will allow us to engineer beat patterns across a wide range of shapes and frequencies, and we have the ability and techniques to measure flow profiles in a variety of fluid environments. By fully exploring the physical mechanisms behind mucociliary clearance in the human lung we can add an
essential component to the body of knowledge regarding cystic fibrosis, which may lead to better and more advanced treatment options in the future.
APPENDIX A: Applied Field Calculation for Continuous Model

Taking a look for a moment at the case of a rigid rod in a constant magnetic field, we could quickly write down the total magnetic energy as the sum of this internal energy and the energy from an applied field \((\hat{m} \cdot \vec{B})\) as follows:

\[
U' = -\frac{1}{4} \mu_0 \frac{m^2}{V} \cos^2(\alpha - \phi) - mB \cos(\psi - \alpha) \tag{74}
\]

The second term dominates if \(4B / [\mu_0 M(B)f] >> 1\), (where we have used the fact that \(m = M(B)f\), where \(M(B)\) is the magnetization curve of the magnetic material in the rod and \(f\) is the volume fraction of magnetic material in the composite), and the energy is therefore minimized when the magnetic moment lies along the field line \(\psi = \alpha\). This condition depends only on the material and not the geometry of the rod. For our material, where \(M(B)\) is the magnetization curve of the maghemite as determined by SQUID measurements, \(4B / [\mu_0 M(B)f] \sim 100\). We may therefore assume that the magnetic moment of the rod aligns at every point along the length of the rod with the applied magnetic field, i.e. that \(\hat{B}_A = M(B)\) are parallel.

With this in mind, the third term (the energy of the rod in an applied external field) can be written as follows

\[
U_A = -A \int_0^L \hat{B}_A M(B) dl \tag{75}
\]

Since \(\hat{B}_A\) is non-uniform, we can approximate it with the first couple terms of a Maclaurin series as follows:

\[
B(l) = B_0 + \nabla B l \cos(\psi - \phi) + 2l^2 \frac{\partial}{\partial l} (\nabla B) + L \tag{76}
\]
For our experiments, $l \sim 25$ microns $\nabla B \sim 300$ T/m, and $\frac{\partial}{\partial l}(\nabla B) \sim 3 \times 10^3$ T/m$^2$.

Therefore the third term of the expansion is approximately 20 times smaller than the second and may be ignored.

The magnetization of the material, $M(B)$ depends on the applied magnetic field in a rather complicated manner, as described by the hysteresis curve of the material. For fields that are small enough that they do not saturate the material, however, we can approximate the magnetization as linear with respect to magnetic field, $M(B) = \chi B(l)$. Inserting these relationships into the applied field energy term from above,

$$U_A = -A \chi \int_0^l [B_0 + \nabla B | \cos(\psi' - \phi)] dl$$  \hspace{1cm} (77)

Due to the extremely small size of the nanorods in our experiments, $B >> \nabla B$ (for our experimental geometries, $\frac{B}{\nabla B} \sim 100$) and upon squaring, the term of order $\nabla B|^2$ can be ignored. Thus, after integrating,

$$U_A = A L \chi B_0 [B_0 + \frac{1}{2} \nabla B L \cos(\psi' - \phi)]$$ \hspace{1cm} (78)

The first term is constant with respect to the motion of the rod and may be dropped. Recognizing that $AL \chi B_0 = m$, the magnetic moment of the rod, the applied field term becomes

$$U_A = \frac{1}{2} m \nabla B L \cos(\psi' - \phi)$$ \hspace{1cm} (79)

For larger fields we can arrive at the same result by assuming that as the metal approaches saturation we can approximate the magnetization as constant with respect to field, $M(B) = M$. Inserting a constant magnetization and the first two terms of the Maclaurin series (Equation 10) into Equation 9, we again obtain the energy $U_A$ derived above.
This term can be thought of as the effect of the magnetic field gradient pulling on a magnetic moment of magnitude $m$ located at the centroid of the rod. Under the straight-rod assumption of the Maclaurin series, the centroid is located on the long axis of the rod, half way along its length. For a rod of constant curvature, however, the centroid does not lie on the axis of the rod. By geometrical arguments, we can show that for a rod bent through an angle $\leq 90^\circ$, the error in the position of the centroid generated by the straight rod assumption is $\sim 33\%$. Given that the field gradient is approximately linear on the scale of the rods, this results in an error of $\leq 33\%$ in this term.
APPENDIX B: Taylor Expansion for the B-Field in the Discretized Model

Taking the magnetic field to be a function of $x$ and $y$, and expanding it via a Taylor expansion,

$$B(x, y) = B(0,0) + \left[ \frac{\partial B}{\partial x} \bigg|_{0,0} x + \frac{\partial B}{\partial y} \bigg|_{0,0} y \right] + \frac{1}{2} \left[ \frac{\partial^2 B}{\partial x^2} \bigg|_{0,0} x^2 + \frac{\partial^2 B}{\partial y^2} \bigg|_{0,0} y^2 + 2\frac{\partial^2 B}{\partial x \partial y} \bigg|_{0,0} xy \right] + \mathcal{O}(3)$$

(80)

Recall also that, by definition,

$$\nabla B = \left( \frac{\partial B}{\partial x} + \frac{\partial B}{\partial y} \right)$$

(81)

Figure 56. Geometrical considerations for the Taylor expansion.

Then, following Figure 56, we can rewrite partial derivatives of $B$ and the coordinates $x$ and $y$ in terms of the angles $\phi$ and $\psi$, the field gradient $\nabla B$, and $l$: 

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\[
\cos \psi' = \frac{\partial B}{\partial y} \frac{1}{|\nabla B|} \quad \Rightarrow \quad \frac{\partial B}{\partial y} = |\nabla B| \cos \psi' \tag{82}
\]

\[
\sin \psi' = \frac{\partial B}{\partial x} \frac{1}{|\nabla B|} \quad \Rightarrow \quad \frac{\partial B}{\partial x} = |\nabla B| \sin \psi' \tag{83}
\]

\[
\cos \phi = \frac{y}{l} \quad \Rightarrow \quad y = l \cos \phi \tag{84}
\]

\[
\sin \phi = \frac{x}{l} \quad \Rightarrow \quad x = l \sin \phi \tag{85}
\]

Plugging these into the Taylor expansion, the first two terms become

\[
B(x, y) = B(0, 0) + |\nabla B| \left[ \sin \psi' \sin \phi + \cos \psi' \cos \phi \right] + L \tag{86}
\]

which, by a trigonometric identity, is

\[
B(x, y) = B(0, 0) + |\nabla B| \cos (\psi' \phi) + L \tag{87}
\]

Using the same method to rewrite the third term of the Taylor expansion in terms of the relevant quantities, the third term, \(B_3\), becomes

\[
B_3 = \frac{l^2}{2} \left[ \sin^2 \phi \sin \psi' \frac{\partial \nabla B}{\partial x} + \cos^2 \phi \cos \psi' \frac{\partial \nabla B}{\partial y} + 2 \cos \phi \sin \phi \cos \psi' \frac{\partial \nabla B}{\partial x} \right] \tag{88}
\]

Since the quantity in brackets is necessarily less than or equal to four,

\[
B_3 \leq 2l^2 \frac{\partial \nabla B}{\partial l} \tag{89}
\]
Therefore for small length scales or for fields with nearly constant gradient, the third
term is negligible. In the experiments described in this dissertation, a typical value of \( l \) is 10
microns, and the greatest measured change in gradient over distance (from the pole of a
permanent cylindrical magnet) is about \( 3 \times 10^5 \) T/m\(^2\). The average field gradient over this
range is 3600 T/m. Using these conservative values, the magnitude of the third term of the
Taylor expansion is \( 1/60^{\text{th}} \) the second. Therefore, the magnetic field term may be written as
just the sum of the first two terms of the Taylor expansion:

\[
B = B_0 + \nabla l \cos(\psi - \phi)
\]  

(90)
APPENDIX C: Current-Carrying Capacity of a Gold Microwire

The current capacity of any wire is limited by the physical damage caused by the thermal energy dissipated in the wire due to resistive heating. As an upper bound on current capacity on a gold wire, we will assume that the wire will continue to function until it reaches its melting temperature. The wire is then gains thermal energy by resistive heating and loses thermal energy due to convection, conduction, or radiation. In the case of a gold wire lithographed on a silicon wafer and encased in PDMS (Figure 57), there is no convection and so all losses will be due to conduction or radiation.

![Diagram of a cross-section of a lithographed gold wire on a silicon substrate.](image)

**Figure 57.** Cross-section of a lithographed gold wire on a silicon substrate. In this model, thermal energy flows from the gold wire (the hot bath, $T_H$) to the cold bath, $T_C$ on the opposite side of the silicon. We assume no thermal
energy is dissipated through the PDMS due to its low thermal conductivity. Thermal conductivities and electrical resistivities depicted in the figure are at 300 K.

Thermal Losses Due to Conduction

The thermal conductivity of PDMS is very low compared to that of the silicon, so the PDMS may be reasonably approximated a perfectly insulating material, and conductive losses through the three sides of the wire in contact with the PDMS will be ignored. Conductive losses through the fourth side, in contact with the silicon, may be calculated by Fourier’s Conduction Law as follows:

$$p_{\text{cond}} = \frac{dQ}{dt} = -\frac{A}{R_T} \nabla T$$  \hspace{1cm} (91)

where $A$ is the area of contact between the hot bath (the gold) and the thermal conductor (the silicon), $R_T$ is the thermal resistance of the silicon, and $\nabla T$ is the temperature difference between the hot bath and the cold bath. For $T_H$ equal to the melting point of gold, $1337$ K, and $T_C = 300$ K, the thermal resistance of a 500 micron layer of silicon is about $1\times10^5$ (as calculated below). The area $A$ is equal to $2\pi L$, where $L$ is the length of the wire. For a radius of 5 microns, the power dissipated via conduction per unit length is:

$$\left(\frac{P}{L}\right)_{\text{cond}} = 990 \text{ W/m}$$  \hspace{1cm} (92)

Thermal Losses Due to Radiation

Thermal losses due to radiation can be calculated with the Stefan-Boltzmann law, as follows
\[ P_{\text{rad}} = \sigma \varepsilon A (T_H - T_C)^4 \]  \hspace{1cm} (93)

where \( \sigma = 5.67 \times 10^{-8} \text{ W/m}^2\text{K}^4 \) is the Stefan-Boltzmann constant, \( A \) is the surface area of the wire, and \( \varepsilon \) is the emissivity of the material. For gold, \( \varepsilon \approx 0.1 \). For a square wire with \( r = 5 \) microns and setting \( T_H \) as the melting point of the gold, 1337 K, and \( T_C = 300 \) K, the power dissipated per unit length due to radiation is

\[ \left( \frac{P}{L} \right)_{\text{rad}} = 0.7 \text{ mW/m} \]  \hspace{1cm} (94)

Since this is much less than the power lost due to conduction, radiative losses will subsequently be ignored.

**Thermal Gains due to Resistive Heating**

The thermal power gained by resistive heating is

\[ P_{\text{elect.}} = I^2 R = I^2 \rho \frac{L}{A} \]  \hspace{1cm} (95)

where \( A \) is the cross-sectional area of the wire, \( 4r^2 \), \( L \) is the length of the wire, and \( \rho \) is the resistivity of the material.

**Calculating the Current**

In the steady state, the thermal gains due to resistive heating must be offset by the thermal losses due to thermal conduction. Therefore,

\[ P_{\text{elect.}} = P_{\text{cond.}} \]  \hspace{1cm} (96)

\[ I^2 \rho \frac{L}{8r^2} = \frac{2rL}{R_T} (T_H - T_C) \]  \hspace{1cm} (97)
\[ I = \sqrt{\frac{8\rho^3}{R_T \rho}} (T_H - T_C) \quad (98) \]

represents the current through the wire in the steady state. Both the conductivity of the wire and the thermal resistance may depend on temperature. In the case of a gold wire, \( \rho \) is linear with respect to temperature over the range \( 50 \leq T \leq 1000 \) K, and takes the form \[^{103}\]

\[ \rho_{Au}(T) = 8.7 \times 10^{-11} T \ \Omega \ m \quad (99) \]

The dependence of the thermal resistance on temperature is described below.

**Thermal resistance with temperature dependent \( k \)**

For a material of constant thermal conductivity, the thermal resistance is given by

\[ R_T = \frac{d}{k} \], where \( d \) is the thickness of the thermal conductor and \( k \) is its thermal conductivity.

However, the thermal conductivity of the silicon varies significantly with temperature. Since thermal resistances add linearly, the total thermal resistance may be generalized as

\[ R_T = \int_0^d \frac{dx}{k(T(x))} \quad (100) \]

According to data from Glassbrenner and Slack\[^{104}\], the thermal conductivity of silicon varies with temperature as

\[ k_{Si}(T) = 6.83 \times 10^5 T^{-1.45} \ \text{W m}^{-1} \text{K}^{-1} \quad (101) \]

at temperatures greater than about 30 K.

Assuming that in the steady state there will be a constant thermal gradient between \( T_H \) and \( T_C \), of the form \( T(x) = (T_H - T_C) \frac{x}{d} + T_C \), then the thermal conductivity as a function of \( x \) is
\[ k_{si}(x) = 6.83 \times 10^5 \left[ (T_h - T_c) \frac{x}{d} + T_c \right]^{-1.45} \] (102)

Upon integrating, the thermal resistance, \( R \), then becomes

\[ R_T = \frac{d}{6.83 \times 10^5} \left[ 0.44(T_h - T_c)^{1.45} + T_c^{1.45} \right] \] (103)

Putting it All Together

Inserting the thermal resistance from Equation 103 and the temperature dependent conductivity from Equation 99 into the steady state current equation (Equation 98), the steady state current is as plotted in Figure 58. Here we have assumed that the silicon substrate is 500 microns thick, which is at the thinner end of what is commercially available, and have plotted curves for both a room temperature bath (300 K) and a liquid nitrogen bath (77 K). In either case, the current capacity of the wire is on the order of one ampere.
Steady-State for a Current-Carrying 10 micron Gold Wire on a 500 micron Silicon Substrate

Figure 58. Current-carrying capacity of a gold microwire. Solid lines represent the steady-state current; dashed lines represent the electric field in the wire.
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