Preparation and characterization of cellulose-based nanomaterials

by

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Abstract

The interest in cellulose for the purposes of nanocomposite engineering lies in its natural abundance and renewable nature, but also in the truly impressive range of diverse properties which can be accessed from the raw resource. In cellulosic nanocomposite materials, the cellulose component may provide the polymeric matrix for nanoparticles and/or the nanometer-scale constituent. This dissertation presents unique cellulose-based nanocomposites and examines the properties of these materials. Fluorescent cellulose triacetate films are obtained by solvent casting suspensions of CdSe/ZnS quantum dots in polymer solution. The films possess properties characteristic to each distinct component: for instance, the optical absorbance and fluorescence are defined by the quantum dots, and the optical clarity and mechanical properties by the polymer. Surface hydrolysis of the hydrophobic films does not substantially alter bulk film properties but does impart aqueous compatibility, allowing film pieces to be introduced into wet paper-making suspensions to produce novel fluorescent papers. The mixture of quantum dots in cellulose triacetate solution is also employed to electrospin sub-micron diameter, birefringent, fluorescent fibers. A different cellulosic-quantum dot system is explored with the asymmetric, reducing end tagging of cellulose nanocrystals. The target fluorescently-labeled cellulose nanocrystal is achieved but the presence of substantial unreacted starting material seems unavoidable, apparently regardless of reaction conditions. Finally, a cellulosic-nanocomposite hydrogel is prepared by incorporating cellulose nanocrystals into polyvinyl alcohol hydrogels with the aim of reinforcement. The cellulose nanocrystal-loaded hydrogels exhibit distinct morphologies and microstructures, and improved elastic strengths. The thesis discusses the rationale and promise of employing cellulose particles and polymers in nanocomposite materials.
Résumé

L’intérêt d’utiliser la cellulose dans l’ingénierie des nanocomposites repose sur son abondance naturelle, son aspect renouvelable et sa grande variété de propriétés qui sont accessibles à partir de sa source brute. Dans les matériaux nanocomposites, la cellulose peut fournir une matrice polymérique aux nanoparticules ou peut être utilisé comme constituants nanométriques. Cette thèse présente de nouveaux matériaux nanocomposites faits à partir de cellulose et en examine les propriétés uniques. Des films fluorescents de triacétate de cellulose ont été obtenu par coulée-évaporation de suspension de points quantiques de CdSe/ZnS dans une solution de polymère. Ces films possèdent des propriétés caractéristiques distinctives. Par exemple, l’absorbance optique et la fluorescence sont définis par les points quantiques, tandis que la clarté optique et la plasticité sont définis par le polymère. L’hydrolyse à la surface des films hydrophobiques n’a pas altéré de manière significative les propriétés générales du film, mais les a rendu compatibles en milieux aqueux, ce qui a permis l’introduction de morceaux de ces films dans le procédé de fabrication du papier pour produire du papier fluorescent. Le mélange de points quantiques dans une solution de triacétate de cellulose a aussi été employé pour l’électrofilage de fibres fluorescentes ayant des diamètres inférieurs à un micron. Un système différent de points quantiques et de cellulose a aussi été exploré. Dans ce dernier système, le marquage asymétrique d’une des extrémité des nanocristaux de cellulose a été réalisé avec succès. Cependant, même en modifiant les conditions expérimentales, la présence d’une quantité substantielle de matériaux réactifs non-réagis n’a pu être évitée. De plus, un nanocomposite hydrogel à base de cellulose a été préparé par incorporation de nanocristaux de cellulose dans un hydrogel d’alcool de polyvinyle, à des fins de renforcement. L’hydrogel ainsi formé possède une microstructure distincte, une intégrité structurale améliorée, un module plus élevé ainsi qu’une structure résiliente au gonflement d’eau. Cette thèse aborde les principes et applications prometteuses de l’emploi des particules de cellulose et polymères dans la fabrication de matériaux nanocomposites.
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Contribution of Authors

All the manuscripts presented in this dissertation were entirely written by Tiffany Abitbol and were co-authored with Professor Derek G. Gray (Department of Chemistry, McGill University), who acted as research advisor. All of the research presented in this thesis was planned, executed and analyzed by Tiffany Abitbol, with guidance from Professor Derek G. Gray. The manuscripts presented in Chapters 4 and 6 were co-authored with Jordan Wilson and Timothy Johnstone, respectively, summer students who assisted with experiments. Professor Thomas M. Quinn (Department of Chemical Engineering, McGill University) co-authored Chapter 6. Dr. Quinn provided training and access to the mechanical testing apparatus, and assisted with manuscript editing.

Chapter 1 presents a review of pertinent cellulosics and their usage in nanocomposite materials. The chapter was researched and written by the author, with editorial assistance from Professor Derek G. Gray. Chapter 1 is unpublished.

Chapter 2 describes a novel fluorescent film system comprised of quantum dots dispersed in cellulose triacetate. Experiments and result interpretation were performed by the author with project inspiration and guidance from Professor Derek G. Gray. Dr. N. Ulkem is acknowledged for help with Soxhlet extraction of blank cellulose sample for XPS analysis, A. Lejeune (UQTR) for XPS data acquisition, and Dr. Xui Dong Liu for TEM. The work was published in Chemistry of Materials in 2007.

Chapter 3 explores the potential application of fluorescent cellulosic films for use as optical markers in paper. The experiments and interpretation were performed under the supervision and direction of Professor Derek G. Gray. The results were published in Cellulose in 2009.

Chapter 4 employs the now established quantum dot - cellulose triacetate system to prepare fluorescent fibers using electrospinning. The experiments and interpretation of results were conducted by the author with guidance from Professor Derek G. Gray. Under the supervision of the author, Jordan T. Wilson assisted with the fabrication of the electrospinning apparatus, solution preparation and electrospinning experiments. This study was published in the Journal of Applied Polymer Science in 2011.
Chapter 5 looks at the possibility of tagging cellulose nanocrystals with quantum dots. The experimental procedure and interpretation were performed by the author, under the supervision of Professor Derek G. Gray. Dr. Xue Dong Liu is acknowledged for TEM imaging and Wayne Mah for a fresh take on some of the experiments. This chapter is unpublished.

Chapter 6 describes a novel cellulosic hydrogel nanocomposite material. The experiments and interpretation were performed by the author, with supervisory assistance from Professor Derek G. Gray. Timothy Johnstone executed preliminary experiments under the supervision of the author, and his work and insights are acknowledged. The work was done in collaboration with Professor Thomas M. Quinn, who is acknowledged for training the author to use the compression apparatus, describing the theoretical basis of the experiment, and general editorial guidance. This study was published in *Soft Matter* in 2011.
I hereby give copyright clearance for the inclusion of the following unpublished chapters, of which I am a co-author, in the dissertation of Tiffany Abitbol.

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And

Chapter 5: “Fluorescent labeling of cellulose nanocrystals with quantum dots”
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<tr>
<td>AFM</td>
<td>atomic force microscopy</td>
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<tr>
<td>AGU</td>
<td>anhydroglucose units</td>
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<td>ATR - IR</td>
<td>attenuated total reflectance – infrared spectroscopy</td>
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<tr>
<td>C.I.</td>
<td>crystallinity index</td>
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<tr>
<td>CA</td>
<td>cellulose acetate</td>
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<tr>
<td>CMC</td>
<td>carboxymethyl cellulose</td>
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<tr>
<td>CNC</td>
<td>cellulose nanocrystal</td>
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<td>CTA</td>
<td>cellulose triacetate</td>
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<tr>
<td>DMF</td>
<td>N,N-dimethylformamide</td>
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<tr>
<td>DMSO</td>
<td>dimethyl sulfoxide</td>
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<tr>
<td>DP</td>
<td>degree of polymerization</td>
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<td>dp</td>
<td>depth of penetration</td>
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<td>DS</td>
<td>degree of substitution</td>
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<td>DSC</td>
<td>differential scanning calorimetry</td>
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<td>e</td>
<td>electron charge</td>
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<tr>
<td>EDC</td>
<td>1-ethyl-3-(3-dimethylaminopropyl)carbodiimide</td>
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<tr>
<td>( E_g )</td>
<td>band gap</td>
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<td>EtOH</td>
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<td>FTIR</td>
<td>Fourier transform infrared spectroscopy</td>
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<td>h</td>
<td>Planck’s constant</td>
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<tr>
<td>LC</td>
<td>liquid crystalline</td>
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<td>LCST</td>
<td>lower critical solution temperature</td>
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<td>MC</td>
<td>methylene chloride</td>
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<td>microcrystalline cellulose</td>
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<tr>
<td>( m_e )</td>
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<td>methanol</td>
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<tr>
<td>( m_h )</td>
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<td>P</td>
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<td>polyethylene glycol</td>
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<td>QD</td>
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<td>r</td>
<td>Bohr radius</td>
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<td>R</td>
<td>radius</td>
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<td>SEM</td>
<td>scanning electron microscopy</td>
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<tr>
<td>Sulfo-NHS</td>
<td>N-hydroxysulfosuccinimide</td>
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<tr>
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<td>transmission electron microscopy</td>
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<tr>
<td>TOPO</td>
<td>n-trioctyl phosphine oxide</td>
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<tr>
<td>XPS</td>
<td>X-ray photoelectron spectroscopy</td>
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Greek symbols:

<table>
<thead>
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<th>Symbol</th>
<th>Description</th>
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<td>$\varepsilon$</td>
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<td>$\varepsilon$</td>
<td>strain (Chapter 6)</td>
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<tr>
<td>$\varepsilon_o$</td>
<td>vacuum permittivity</td>
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<td>$\sigma$</td>
<td>stress</td>
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Chapter 1
Introduction

This chapter introduces nanocomposite materials comprised of at least one cellulosic component, either in the role of polymer matrix or nanoparticle filler. It also provides background on quantum dots, and the electrospinning technique.
Introduction
1.1. Cellulose

Cellulose has very long been an indispensable raw material and it is no great leap to credit the evolution of mankind, in some part, to its effective manipulation. The relationship between human industriousness and cellulose is ancient, perhaps most significantly beginning in the Stone Age with the burning of woods and grasses and progressing into current times to help meet the needs of industrialized man. While commercial interest in cellulose may have waned somewhat during the second half of the 1900s due to the pervasiveness of cheap plastics from fossil fuels and the widespread use of digital media, it has recently been picked up again. Being the most abundant, truly renewable, biodegradable natural polymer, it is none too surprising, that science and engineering continue to look to cellulose as a fundamental building block for a huge range of endeavors. Cellulose is arguably the original “advanced material” with a wide variety of pre- and post-processing applications, including shelter, fuel, building materials, textiles, paper and more recently in high technology plastics and fibers.

Cellulose was first identified as the main constituent of the plant cell wall by the French chemist Anselme Payen in 1838. It is isolated mainly from higher plants but also from marine plants (e.g. algae) and animals (e.g. tunicate), and bacterial sources (e.g. *Acetobacter xylinum*), with annual global production estimated to be as high as $10^{12}$ tons. The repeat unit of the polymer is cellobiose, which consists of two D-anhydroglucose units (AGUs) in chair conformation linked in an equatorial or $\beta$ configuration at the C1 and C4 positions (Figure 1.1). Each cellobiose unit is rotated 180 degrees with respect to its neighbors, resulting in a sterically stable, linear configuration. The chain is directionally asymmetric, with an alcohol at one terminus and a hemiacetal at the other, referred to respectively as the non-reducing and reducing ends. The biogenesis of cellulose produces chain aggregates or bundles of elementary fibrils called microfibrils, which vary in diameter depending upon source, from ~4-35 nm. The chair conformation of the sugar units and the equatorial positioning of the hydroxyl groups promote the formation of hydrogen bonds along the chain lengths. The complex network of intra- and inter- molecular hydrogen bonds and van der Waal interactions stabilize the chains and microfibrils. The associative nature of cellulose chains gives rise to a highly
crystalline extended structure which, though hydrophilic and moisture responsive, is entirely insoluble in water and most other common solvents.

![Chemical structure of cellulose.](image)

**Figure 1.1**: Chemical structure of cellulose.

The molecular weight, purity and crystallinity of cellulose are origin dependent. Cellulose is polydisperse, with a degree of polymerization (DP) based upon anhydroglucose units (AGUs) of up to 12,000 for cotton but significantly lower for wood pulps (DP = 600-1200) and man-made fibers (DP = 250-500). Cotton is almost entirely composed of pure cellulose (~95%) whereas the cellulose in woody plants is closely associated with lignin and hemicelluloses (i.e. polysaccharides containing other sugars and sugar derivatives) and only makes up ~42% of the cell wall. Cellulose from wood can however be isolated and purified by pulping, a process which selectively targets the removal of lignin, without incurring substantial chain scission. The degree of crystallinity varies widely, for example the crystallinity index (C.I.) from X-ray diffraction studies was found to be ~0.95 for cellulose from *Valonia ventricosa*, ~0.83 for ramie cellulose and ~0.67 for wood cellulose.

The currently known and accepted crystalline allomorphs of cellulose are I, III$_I$ and IV$_1$ with parallel chain packing, II, III$_{II}$ and IV$_{II}$ with anti-parallel packing. The crystal structure of native celluloses is cellulose I, whereas non-native celluloses (i.e. regenerated or derivatized) have different H-bonding patterns and the potential for anti-parallel chain orientation. Solid-state $^{13}$C-NMR and X-ray diffraction studies have identified two distinct crystalline modifications within the general native cellulose I
family, referred to as Iα and Iβ: a monoclinic unit cell (Iα) and a triclinic unit cell (Iβ). The Iα/Iβ ratio is origin dependent, for example bacterial and Valonia celluloses are rich in Iα, whereas celluloses from ramie and cotton are dominated by the Iβ allomorph. Iβ is more stable than Iα, with the transformation of Iα to Iβ occurring under certain conditions, such as swelling, acetylation followed by saponification and heat annealing.

1.1.1 Cellulose derivatization

Cellulose is amenable to chemical derivatization through hydroxyl substitution reactions, and is the raw material for a number of interesting and useful cellulosic polymers. Chemically, cellulose behaves as an alcohol and can therefore undergo esterification, etherification, grafting, cross-linking, oxidation and chain degradation reactions. The limited solubility of cellulose may however require an approach to modification which differs from conventional homogeneous chemistries.

The literature tackles cellulose reactivity as a balance between accessibility and susceptibility, where accessible cellulose typically refers to amorphous regions but also to exposed crystallites, and susceptibility is a measure of the propensity of individual cellulose elements toward reaction. The morphology of the starting cellulose, specifically in relation to concentration and size of crystallites, is a huge factor in determining ease of reaction, for example, the lattice transformation from cellulose I to cellulose II improves accessibility because of a decrease in overall crystallinity. In addition to morphological considerations, the hydroxyl groups of cellulose, if equally accessible to reagents, will exhibit different susceptibilities reflective of the usual pecking order. Chemical history and moisture content also influence the reactions of cellulose, for instance, drying cellulose decreases reactivity due to ‘hornification’, the hydrogen-bond strengthening which occurs when the swollen structure is collapsed. In order to improve the accessibility of cellulose to solvent and reagents, the standard approaches to derivatization either involve swelling the lattice structure (e.g. in water, ethylamine or aliphatic diamines) or fully dissolving the cellulose in an appropriate solvent (e.g. N,N-dimethylacetamide/LiCl).
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Cellulose derivatives possess a range of distinct physical and material properties: cellulose acetate (CA) is a transparent, flexible plastic which is soluble in acetone, hydroxypropyl cellulose is water soluble and can be cast into highly colored, chiral nematic, plastic films and exhibits a lower critical solution temperature (LCST) of ~40 °C, and cellulose nanocrystals are highly crystalline particles which may be colloidally dispersed in water. Successful derivatization, to obtain a product with the desired properties and quality, requires careful consideration of both the starting cellulose and reaction conditions.

1.1.2 Cellulose triacetate

The cellulosic polymer featured in this thesis is cellulose triacetate (CTA) (Figure 1.2), which is defined by a degree of substitution (DS) of 2.8-3.0 acetyl groups per AGU. The generic term cellulose acetate may be used to refer to derivatives with DS values anywhere from 0.1-3, whereas the DS of cellulose mono- and di-acetates range from 0.6-1.4 and 2.2-2.7, respectively. The DS is an average value since the starting material is in itself heterogeneous in terms of crystallinity, purity and chain length, and therefore a range of substitutions is obtained in the final acetylated product.

Figure 1.2: Chemical structure of cellulose triacetate.

Cellulose esters are derived from the acetylation of cellulose, typically from wood sources but also sometimes from cotton linters. Two polymorphs of CTA are possible: CTA I, with parallel chains, is derived from the heterogeneous acetylation of...
native celluloses, and the anti-parallel CTA II allomorph is obtained from the reaction of regenerated or mercerized celluloses. Deacetylation of CTA by heterogeneous saponification yields cellulose with the same chain packing arrangement (i.e. anti-parallel or parallel) as the starting CTA.

In contrast to the more common, homogeneous, ‘solution process’, where the acetylated product is solubilized, the heterogeneous ‘fibrous process’ retains the gross and fine morphologies of the initial cellulose through the addition of a non-solvent (e.g. toluene, benzene or amyl acetate), which prevents dissolution of the esterified product. The ‘solution process’ and the ‘fibrous process’ both utilize a dry mixture of acetic anhydride and acetic acid in the presence of a catalytic quantity of either sulfuric or perchloric acids. Cellulose esterification typically involves an initial activation step, where the cellulose is treated mechanically to obtain smaller fragments and the pore structure is swollen in water or aqueous acetic acid. Prior to reaction, the water may be exchanged for a mix of dry acetic acid and acetic anhydride or consumed by addition of equimolar acetic anhydride. The competing acid-catalyzed hydrolysis reaction which cleaves the glycosidic bond is an issue, and reaction conditions are optimized to control chain length while achieving the desired degree of acetylation. The temperature also needs to be controlled by some method of cooling since both acetylation and acetic anhydride hydrolysis are exothermic. The reaction is quenched by the addition of water, which consumes unreacted acetic anhydride through hydrolysis and promotes acid catalyzed desulfation. At this stage, the degree of acetylation may be reasonably well controlled by acid catalyzed hydrolysis to less substituted ester products, such as mono- or di-acetates.

The exact mechanism of acetylation remains unclear but most likely proceeds through the formation of intermediate cellulose sulfates (Cell-O-SO$_3$H). It is however accepted that acetylation first targets the amorphous regions of the cellulose (i.e. accessibility) and next tackles the crystallites through an ‘erosion’ mechanism which peels back the crystallites, chain by chain, working from the exterior toward the interior (i.e. susceptibility). Sassi and Chanzy validated the ‘erosion’ hypothesis by monitoring the acetylation of *Valonia* and tunicin crystallites using transmission electron microscopy (TEM) and X-ray diffraction. They observed a decrease in the lateral order
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and dimensions of crystallites which had undergone homogeneous acetylation, consistent with surface acetylation and crystallite erosion.\textsuperscript{24} The ‘erosion’ mechanism has since been confirmed by Kim et al.\textsuperscript{25} who studied the fibrous acetylation of bacterial cellulose and obtained surface acetylated microfibrils with crystalline cellulose I cores. The chain packing of the crystallites also seems to play a role; according to Sassi et al.\textsuperscript{27} the I\textsubscript{α} phase is more susceptible to acetylation compared to the I\textsubscript{β} phase.

Cellulose acetates are employed in numerous applications, such as photographic films, liquid crystal displays (LCDs), separation membranes, rigid plastics (e.g. eyeglass frames and handles), textiles, cigarette filters (i.e. Filter Tow), etc.\textsuperscript{22} CTA is soluble in organic solvent, typically in solvent mixtures dominated by chlorinated hydrocarbons such as dichloromethane, and is most commonly used in the form of films and fibers. CTA films were first commercialized over 50 years ago as an alternative to the highly flammable photographic films based upon cellulose nitrate and have since been used as protective films for polarizers (FUJI-TACTM) and optical compensation films to widen the viewing angle in LCDs (WV FilmTM).\textsuperscript{28} CTA films are characterized by very low birefringence, high transparency (≈93%), reasonable mechanical strength, low flammability, and good surface smoothness, dimensional stability (e.g. curl recovery) and chemical resistance.\textsuperscript{28}

In contrast to the currently profitable CTA films, the market for CTA fibers has largely been replaced by less expensive synthetic polyesters. However, as fossil fuels become scarcer, it may be reasonable to expect a new surge in the production of cellulose-based fibers.\textsuperscript{20} (In fact, Lenzing Fiber Inc. is currently marketing a cellulose fiber called Tencel® which is touted for its natural source, eco-friendly and economical production, and attractive fabric properties.) CTA fibers, which are prepared from either melt or solution spinning processes, exhibit resistance to wrinkling, good dimensional stability, high tolerance to elevated temperatures and ease of care.\textsuperscript{29} Heat treatment may be employed to improve fiber crystallinity, and surface saponification to impart desirable anti-static properties.\textsuperscript{29}
1.1.3 Cellulose nanocrystals

In native cellulose, dislocations or defects disrupt the crystalline ordering of the microfibril chains, resulting in the presence of disordered (i.e. amorphous) regions. Colloidal cellulose crystallites are produced by acidic treatment which selectively targets the accessible, amorphous regions of the microfibrils, leaving behind highly crystalline, rod-shaped, nm-sized particles (Figure 1.3). Cellulose nanocrystals are also sometimes referred to as nanocrystalline cellulose (i.e. NCC), whiskers, needles, microcrystals, monocrystals or crystallites, with the chosen term often a question of author preference or trend. It has recently been argued that CNCs are “the most dominant fundamental building block of the biosphere”\textsuperscript{4}, a statement which highlights both the readily available natural abundance and the structural significance of the crystalline material in plant tissues. The preparation of colloidal cellulose particles by treatment of wood or cotton with sulfuric acid was first reported by Rånby in 1949.\textsuperscript{30} Cellulose nanocrystals produced by hydrochloric acid (HCl) hydrolysis are uncharged and colloidally unstable, whereas sulfuric acid hydrolysis results in electrostatically stabilized particles due to the charged sulfate ester groups introduced onto the surface of the crystallites during reaction.\textsuperscript{31}

![Figure 1.3: Schematic representation of acid hydrolysis of cellulose microfibrils, which targets amorphous regions preferentially.](image)

CNCs retain the crystal structure of the native cellulose from which they are derived (i.e. cellulose I).\textsuperscript{31} The particles are polydisperse and the size distribution of crystallites depends upon both the source of cellulose and the reaction conditions employed. In general, it seems that the more crystalline the starting material the greater
the particle dimensions, for example, CNCs from *Valonia Ventricosa* have lengths in excess of 1000 nm and cross-sectional widths of 10-20 nm\textsuperscript{32,33}, and CNCs from cotton (Figure 1.4) and wood have similar dimensions of 100-350 nm in length and ~5 nm in cross section\textsuperscript{34}. Beck-Candanedo et al.\textsuperscript{34} showed that, for CNCs derived from black spruce pulps, longer reaction times produced shorter particles with greater surface charge densities. Indeed, excessive reaction conditions will simply break down the cellulose entirely into its constituent simple sugars. To produce colloidal CNCs, conditions of temperature, acid concentration, reaction time and ratio of acid to cellulose source must be carefully controlled. The commonly employed recipes were developed here in the Gray lab and use temperatures of 45 °C, 64 weight % sulfuric acid, 25-45 minute reaction times and acid to cellulose ratios of 8.75-17.5 mL/g.\textsuperscript{34,35}

![Figure 1.4: Transmission electron micrograph of CNCs produced from the sulfuric acid hydrolysis of cotton. (Copied from Ref.\textsuperscript{36} and reprinted with permission from ACS, © 1996.)](image)

Research into CNCs is exploding at the moment as a direct consequence of the impressive physical properties and “green” nature of the material (i.e. bio-sourced, renewable, sustainable, biodegradable, low apparent toxicity). One of the most interesting properties of CNCs is the spontaneous phase separation of sufficiently concentrated
Chapter 1

suspensions into a lower chiral nematic (cholesteric) liquid crystalline (LC) phase and an upper isotropic phase.\cite{36,37} This interesting optical phenomenon was first noted in 1959 by Marchessault et al.\cite{38} who observed birefringent ‘tactoids’ upon viewing a concentrated cellulose suspension between crossed polarizers. Within the anisotropic phase, the rods form a chiral nematic liquid crystal, which corresponds to stacked nematic planes of CNCs, with the orientation of the crystallites in each layer slightly offset with respect to adjacent layers, such that the LC director rotates through the planes helicoidally (Figure 1.5). The birefringent chiral nematic phase is characterized by a left-handed, cholesteric pitch which can be obtained by sample observation between crossed polarizers, where the spacing between lines within the characteristic fingerprint texture gives half the pitch.\cite{37} The remarkable phase separation behavior, native crystallinity, and high strength (i.e. Young’s modulus in excess of 100 GPa\cite{39}) have spurred the exploration of CNCs in iridescent films\cite{37,40,41}, as alignment media for proteins in NMR studies\cite{42,43}, in model cellulose surfaces\cite{44-47}, and as reinforcement agents in polymer nanocomposite materials\cite{48,49}.

![Figure 1.5: Schematic diagram of isotropic and chiral nematic phases of rod-shaped particles. The cholesteric phase is shown over the distance $P/2$ which is half the chiral nematic pitch ($P$).](image)

1.2. Cellulose-based polymer nanocomposites

The term ‘nanocomposite’ refers to a material made by combining two or more components, at least one of which has nanometer-scale dimensions.\cite{50} The aim of
nanocomposite engineering is to create novel materials whose properties are somehow improved in comparison to those of the individual components, with the beauty of the material often derived from the combination of dissimilar entities. The greatest challenge to the successful preparation of nanocomposites is the requirement of intimate mixing of components. Various approaches exist in order promote miscibility, such as using common solvents, high energy sonication treatments, surfactants, chemical modification, etc. In the case of cellulose-based polymer nanocomposites, the cellulosic component may occupy the role of either nanoparticle filler or polymer host, with the method used to produce a homogeneous material dependent upon the chemical nature of the different material elements.

1.2.1 Nanoparticles incorporated into cellulosics

For a variety of reasons, including the natural abundance and biodegradability of cellulose, researchers have and continue to use cellulose and cellulose polymers as a matrix for nanoparticles. The use of cellulose as a polymeric matrix typically requires either the dispersal or the dissolution of cellulose and either the concurrent or subsequent addition of nanoparticles/nanoparticle pre-cursors. For instance, clay particles have been added to NMMO-based solutions of cellulose\textsuperscript{52, 53}, microcrystalline cellulose (MCC)-hydroxyapatite nanocomposites were prepared by a microwave assisted, one-step reaction where CaCl\textsubscript{2}, NaH\textsubscript{2}PO\textsubscript{4} and MCC were added to \textit{N,N}-dimethylacetamide solvent\textsuperscript{54}, CdS particles were prepared in NaOH/urea cellulose solutions and the regenerated cellulose films cast from the dispersion exhibited the optical properties of CdS\textsuperscript{55}, and all-cellulose nanocomposites were prepared by the selective surface dissolution of bacterial cellulose sheets\textsuperscript{56} and by the electrospinning of core-shell fibrous mats in which CNCs were sheathed by a shell composed of regenerated cellulose\textsuperscript{57}.

Another approach involves the \textit{in-situ} synthesis of nanoparticles within a previously prepared solid cellulose scaffold: Zhou et al.\textsuperscript{58} prepared superparamagnetic nanocomposite films by synthesizing iron oxide (Fe\textsubscript{2}O\textsubscript{3}) nanoparticles within the pores of regenerated cellulose films, Liu et al.\textsuperscript{59} wet spun cellulose fibers from solution in NaOH/urea/H\textsubscript{2}O, followed by treatment in FeCl\textsubscript{3} (0.01, 0.1, 0.5 M) and NaOH (2 M) in order to generate iron oxide particles \textit{in-situ}, and Vilela et al.\textsuperscript{60} prepared CaCO\textsubscript{3}-cellulose
nanocomposites by the synthesizing CaCO₃ nanoparticles in the presence of hardwood bleached Kraft pulp and carboxymethylated cellulose fibers.

Despite the successes achieved using cellulose to prepare interesting and functional nanocomposites, the limited solubility of the polymer somewhat curbs its applicability. In this regard, cellulose derivatives, which are soluble in solvents ranging from water to non-polar organics, are more versatile and have the potential for improved compatibility with nanoparticles. Cellulose acetate is a transparent, flexible, easily processible plastic which has been used as the matrix in nanocomposite materials: Hassan-Nejad et al.⁶¹ used polymer melt intercalation and Wibowo et al.⁶² used extrusion followed by either injection or compression molding to create cellulose acetate-clay nanocomposites, and Jang et al.⁶³ incorporated TiO₂ particles into CA films in order to promote the enzymatic biodegradation of CA by cellulase. Carboxymethyl cellulose (CMC) is a water soluble cellulose derivative, which has been used to stabilize ZnO nanoparticle dispersions in glycerol plasticized-pea starch⁶⁴. CMC has also been employed as the matrix in pH sensitive superabsorbent nanocomposites containing attapulgites⁶⁵, in nanocomposites containing metals (Cu, Ag, In and Fe)⁶⁶ and in hydrogels cross-linked with poly(N-isopropylacrylamide) which contained clay⁶⁷.

1.2.2 Cellulose nanocrystals incorporated into polymeric materials

The high strength, aspect ratio (i.e. L/d ~20-70 for CNCs derived from cotton, greater for CNCs derived from tunicin or MC) and surface area of CNCs impart a potential to significantly improve the mechanical properties of nanocomposites at low CNC filler loadings.⁴⁹, ⁶⁸, ⁶⁹ Percolation theory has been used to describe the surprising reinforcement effects observed at low loadings of fibrous elements. Mechanical properties, such as strength and modulus, are optimal at or above the percolation threshold, where each CNC is, on average, in contact with two others and a rigid, 3-D, hydrogen-bonded network is formed within the polymeric matrix.⁴⁸, ⁷⁰ As with all nanocomposite materials, the challenge resides in the uniform dispersal of CNCs and in achieving good interfacial adhesion between CNCs and matrix, particularly if hydrophobic.⁷⁰ In general, CNC-nanocomposites are either processed into films, by solvent casting, or fibers/fibrous mats, by electrospinning⁵⁷, ⁷¹-⁷³. Electrospinning is very
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promising because the alignment of CNCs in the fibers may enhance axisymmetric properties.69

The most straightforward method for the preparation of polymeric nanocomposites which contain CNCs is the direct addition of CNCs into either a pre-polymer or polymeric solution. To proceed successfully in this approach, the CNCs must be dispersible in the polymeric phase. CNCs prepared from sulfuric acid hydrolysis are colloidally stable in water and it is therefore possible to simply add the crystallites directly to an aqueous system. Some recent examples of CNC-based nanocomposites which rely upon the aqueous stability inherent to sulfuric acid hydrolyzed samples include: (1) films cast from mixtures of CNCs and aqueous poly(oxyethylene) solutions74, (2) films of oriented CNCs in a polyvinyl alcohol (PVA) matrix prepared by application of a 7 T magnetic field during solvent evaporation75, (3) fibrous mats electrospun from CNCs dispersed in aqueous PVA72 and (4) films cast from CNCs were dispersed in aqueous polyurethane solutions76.

In order to disperse CNCs in non-aqueous media (i.e. hydrophobic solvents and polymeric matrices), the approach may involve (1) vigorous sonication to disperse freeze and/or vacuum dried CNC powders in solvents, such as ethanol71, furfuryl alcohol77, formic acid78, N,N-dimethylformamide (DMF)79,80,81 or dimethyl sulfoxide (DMSO)81, which are compatible with a pre-polymeric/polymeric matrix, (2) the use of a surfactant82,83,84,85, (3) solvent exchange of water for organic media such as toluene86 or DMF73 or (4) surface grafting reactions to improve CNC compatibility with hydrophobic polymers86–89. Researchers have achieved some degree of success with each of these methods however, drying CNCs results in aggregation which is never completely reversed upon re-dispersal and it remains uncertain whether graded solvent exchanges are truly able to produce stable CNC dispersions in organic media. In fact, Viet et al.81 attributed the stability of freeze-dried CNCs re-dispersed in polar organic solvents (e.g. DMF, DMSO) to the presence of residual water (~0.1%). In addition, surface modification of the CNCs is challenging since the chemistry may be limited by the dispersibility of CNCs in organic reaction media and must truly be surface specific in order to retain the colloidal nature of the particles. A further difficulty is characterization of the modified product because the usual techniques may not apply to colloidal particles
or else may not possess the degree of sensitivity required to detect changes in surface groups which represent a small fraction of total atoms. For these reasons, direct observation of surface modified CNCs is rarely noted in the literature but is instead inferred from observed changes in fundamental properties (e.g. stability, dispersibility, charge, the presence or absence of LC ordering). This area of research is still quite new and it is hoped that, in the future, approaches which reliably produce stable dispersions at the percolation threshold will be discovered, as they will be necessary for the production of homogeneous CNC-reinforced hydrophobic nanomaterials.

1.3. Background of quantum dots

Semiconductors (Si, Ge, groups III-V and II-VI materials) are characterized by a filled valence band (VB) and an empty conduction band (CB), a concept analogous to the HOMO and LUMO energy states from molecular-orbital theory.\textsuperscript{90, 91} The difference in energy between the valence and conduction bands, called the band gap ($E_g$), is a characteristic property of the bulk solid and varies from 0.5-3.5 eV depending upon elemental composition.\textsuperscript{91} Absorption of a quanta of light will excite an electron from the VB to the CB, leaving behind a positively charged hole in the VB (Figure 1.6).\textsuperscript{92} The electron and hole, referred to collectively as an exciton, are bound together via Coulombic interactions. Equation 1.1 gives the characteristic Bohr radius of a semiconductor, defined as the radius $r$ of the sphere encompassed by the 3-D separation of the electron-hole pair, where $\varepsilon$ is the dielectric constant of the semiconductor, $h$ is Planck’s constant, $m_r$ is the reduced mass of the exciton and $e$ is the electron charge.
Quantum dots (QDs) are nm-sized semiconductor crystals (Figure 1.7). In QDs, the Bohr exciton diameter exceeds the physical confines of the particle leading to quantum size-effects.\textsuperscript{90, 94, 95} For CdSe, this translates to particle sizes which are smaller than 112 Å, the bulk Bohr radius value.\textsuperscript{94} The most straightforward and familiar analogue to a QD is the \textit{\textquoteleft particle-in-a-box\textquoteright}, whose energy states are discrete and inversely dependent upon the size of the box. The properties of QDs are strongly size-dependent and differ dramatically from those of bulk phase semiconductors.\textsuperscript{93} The small size of QDs has two major implications: (1) the phase behaviors are dictated by the high fraction surface atoms (e.g., melting temperature is depressed and pressure needed to induce solid-state phase transformations is increased) and (2) the electronic and optical properties are transformed due to quantum confinement effects.\textsuperscript{93} The electronic properties of QDs are intermediate between the discrete energies of molecules and the
continuous bands of bulk semiconductors (Figure 1.8).\textsuperscript{90, 96} Within a given semiconductor composition, the band gap of QDs occurs at shorter wavelengths compared to the bulk material and red-shifts toward the bulk value as particle size increases.\textsuperscript{90} Quantum size-effects, i.e. the shift in band gap to higher energies with decreasing particle size and the discrete energetic transitions observed in QDs, are a direct consequence of exciton confinement in the nm-scale particles.\textsuperscript{92}

![Image](image_url)

Figure 1.7: Transmission electron micrograph of CdSe/ZnS quantum dots dried down from suspension in toluene onto a Formvar® treated carbon-coated copper substrate.
Figure 1.8: Energy level diagram comparing the $E_g$ of quantum dots to bulk semiconductors and molecules. (Figure adapted from Refs.⁹¹, ⁹³) Within a fixed elemental composition, the smaller the QD, the more blue-shifted $E_g$ will be compared to the bulk material and *vice-versa*.

Equation 1.2 is an approximate calculation for the bandgap energy of a quantum dot formulated by Brus⁹⁵, where $R$ is the radius of the QD, $m_e$ and $m_h$ are the effective masses of the charge carriers in the bulk solid and $\varepsilon_0$ is the vacuum permittivity. The second term in the equation is a *particle-in-a-box*-like term, with an energy-dependence of $1/R^2$, and the third term takes into account the Coulombic attraction between the electron and hole. This formulation makes some assumptions (i.e. spherical shape, size-independent effective mass and dielectric constant) but works well for larger particles where the exciton Bohr radius does not hugely exceed QD dimensions.⁹¹

$$E_g(QD) = E_g(bulk) + \left( \frac{\hbar^2}{8R^2} \right) \left( \frac{1}{m_e} + \frac{1}{m_h} \right) - \frac{1.8e^2}{4\pi\varepsilon_0\varepsilon R} \quad (1.2)$$
The fluorescence of QDs arises from the radiative recombination of electron-hole pairs across the band gap. The absorbance onset and the emission wavelengths are dependent upon particle size and shift to higher energies as the size decreases. It is therefore possible, by tuning the QD size, to access different and potentially non-overlapping emission wavelengths. The broad absorbance of QDs (Figure 1.9) means that fluorescence can be excited by wavelengths greater than or equal to the band gap (i.e. the absorption onset) and, regardless of excitation wavelength, the fluorescence peak will still more or less occur at the characteristic wavelength.\textsuperscript{97} This is important for multicolor applications, where a mixture of different QD sizes can be combined and all emissions can be excited using a single wavelength.

CdSe quantum dots have band edge absorptions and emissions within the visible range of the spectrum, emitting from blue to red with increasing particle size.\textsuperscript{94} The quantum dots employed in this work were CdSe with a ZnS “overcoat”: passivation of uncoordinated CdSe surface sites with a higher band gap material such as ZnS prevents leakage of charge carriers from the CdSe core and removes surface trap states, i.e. energetic states which lie within the band gap of the semiconductor. In QDs, where a large fraction of total atoms are surface atoms, emission from these lower energy states may dominate over band-edge recombination (Figure 1.10).\textsuperscript{98} The addition of a ZnS shell does not substantially alter the CdSe emission position or spectral bandwidth (slight red-shift and peak broadening), but improves quantum efficiency by 35-50%, resistance to photobleaching and photochemical stability.\textsuperscript{98-100} Coating CdSe cores with ZnS has also been shown to decrease the fluorescence intermittency, i.e. the on/off behavior observed in single particle photoluminescence experiments.\textsuperscript{94}
Figure 1.9: Absorbance spectrum of CdSe/ZnS quantum dots in toluene as a function of QD concentration. Note the broad and continuous absorbance. QDs were purchased from Evident Technologies, Inc. with 2.1 nm nominal core sizes.
Figure 1.10: Comparison of the fluorescence of bare CdSe QDs (dashed lines) and CdSe/ZnS QDs (solid lines) having different core diameters: (a) 2.3 nm, (b) 4.2 nm, (c) 4.8 nm and (d) 5.5 nm. Note the intensity increase for QDs with a ZnS shell and the strong size-dependence of emission wavelength. (Copied from Ref.\textsuperscript{98} and reprinted with permission from ACS, © 1997.)

Colloidal CdSe/ZnS quantum dots are prepared in the general fashion outlined by Hines and Guyot-Sionnest\textsuperscript{100} and Dabbousi et al.\textsuperscript{101}, however the highly toxic Cd(CH\textsubscript{3})\textsubscript{2} precursor has been replaced by CdO as described by Peng and Peng\textsuperscript{102}. The colloidal stability of the particles in organic solvent (e.g. toluene, hexane) is imparted by the hot surfactant reaction medium (e.g. \textit{n}-trioctylphosphine oxide).\textsuperscript{93} CdSe/ZnS QDs which are water dispersible have been prepared by surface functionalization with polar ligands\textsuperscript{103,104}, silanization\textsuperscript{97,105}, encapsulation within polymeric micelles\textsuperscript{106} or amphiphilic polymer\textsuperscript{107}, and PEGylation\textsuperscript{108-111}. The quantum dots employed in this work were purchased commercially from Evident Technologies, Inc and eBioscience, Inc, either as dispersions in toluene or in water.

Scientific interest in quantum dots ranges from probing fundamental electronic states, to simply using the particles in lieu of organic fluorescent dyes, for example as fluorescent biological probes. The advantages of QDs compared to conventional dyes include (1) a unique optical profile consisting of broad and continuous absorbance and
narrow emission, (2) good photostability, (3) long fluorescence lifetimes and (4) the potential to disperse the particles in a range of solvents by tailoring surface functionality.\textsuperscript{97} In the research presented here, the optical properties of CdSe/ZnS quantum dots are exploited in order create polymer films and fibers which fluoresce in the visible spectral range.

1.4. Background of electrospinning

Electrospinning is a technique where sub-micron diameter fibers (Figure 1.11) are drawn out of a metallic capillary by application of an electric field to a fiber-forming fluid.\textsuperscript{112-116} The focus in this work is electrospinning fibers from polymeric solution. Figure 1.12 depicts a typical electrospinning set-up, the basic components being (1) a high voltage supply (5-30 kV) needed to produce an electric field, (2) a reservoir of polymer solution in a capillary which can be charged in some way (e.g. through insertion of a metallic electrode, or though a hollow metallic needle) and (3) a grounded collector of some sort, composition and geometry being variable. Electrospinning is a variant of the electrospraying technique, which is quite similar but produces polymer droplets as opposed to fibers. The difference between the techniques lies in the processing parameters, with higher concentrations/viscosities and voltages resulting in fibers.
Figure 1.11: Scanning electron micrograph of nanocomposite fibers electrospun from dispersions of CNCs in polyvinyl alcohol solution. These fibers were spun using the following experimental conditions: polymer concentration = 21 wt. %, CNC loading = 2 wt. % relative to polymer mass, voltage = 20 kV, capillary tip-collector distance = 10 cm, flow rate = 1 mL/min.

Figure 1.12: Diagram of electrospinning set-up. Application of a high voltage to the polymer solution causes a charged jet to be ejected from the capillary. As the jet travels through space it is dried and elongated, finally reaching the collector where it is deposited as solid fibers.

The way electrospinning works is more or less straightforward. The application of an electric field induces charges onto the surface of the polymer droplet located at the tip of the capillary. The charges repel one another in opposition to the surface tension of the polymer solution, which acts to maintain the droplet. As the strength of the electric field is increased, the repulsion causes the droplet to distort in shape from hemispherical to
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conical (i.e. a Taylor cone or some near approximation of one). At sufficiently high electric fields, repulsion outweighs surface tension and a charged polymer jet is elongated from the apex of the cone, traveling through the air toward a grounded collector, with jet diameter decreasing and solvent evaporating along the trajectory.\textsuperscript{117} High-speed photography has allowed researchers to directly visualize the electrified polymeric jet and has clarified certain misconceptions regarding fiber formation. It was previously thought that electrostatic instability caused the splaying or branching out of a linear jet, resulting in the deposition of multiple fibers\textsuperscript{118}, but in fact, photographic evidence has shown that, in general, fibers are formed from a primary jet which undergoes rapid whipping motions and bending instabilities as it travels through space, from capillary tip to collector.\textsuperscript{119-121} Secondary jets which emanate from the primary jet have also been observed.\textsuperscript{119, 122}

Numerous studies have been conducted in order to elucidate the relationship, if any, which exists between processing parameters and resultant fiber diameters and morphologies. For a given polymer, experimental conditions are crucial but the conditions which work well for one system may not translate to another. It is therefore important when electrospinning a polymer which has yet to be processed in this manner to spend some time tweaking parameters in order to determine optimal fiber forming conditions. This may be a complicated and time consuming task due to the large number of variables inherent to the method. Doshi and Reneker\textsuperscript{117} have broken down the important parameters into three basic groups: (1) solution properties (e.g. concentration, conductivity, viscosity, surface tension, molecular weight, solvent volatility), (2) controllable experimental variables (e.g. electric field strength, polymer flow rate, distance between capillary and collector, mode of collection) and (3) ambient parameters which may not always be readily controllable (e.g. temperature, air flow, humidity). With regard to solution properties, there is something to be said about concentration/viscosity which is applicable to all systems, namely that for a given fiber-forming voltage, fibers are formed within a boundary defined by upper and lower concentration limits.\textsuperscript{117} Below the lower limit, droplets or beads are formed, and above the upper limit, the viscosity is too great and the droplet dries out at the capillary tip before electrospinning can be initiated.\textsuperscript{117} Very generally speaking and not universally applicable, fiber diameter decreases with increasing solution conductivity (i.e. by adding salt), and with decreasing
flow rate, voltage, and concentration. Increasing polymer molecular weight shifts the minimum concentration required to electrospin fibers (c*) to lower values, and the distance from capillary tip to collector is mostly important in the sense that sufficient distance must be traversed by the fibers to allow for drying.

The nature of the collector (i.e. material and geometry) determines fiber density and whether aligned fibers or random non-woven fabrics are obtained. A collector can basically be anything, ranging from a human hand, to a piece of metal, to more elaborate assemblies like rotating drums. Fibers collected on a flat surface are laid down continuously and somewhat randomly to form a mat of non-woven fabric, whereas a rotating metallic drum will produce a threadlike-spindle of fibers. The fiber deposition density is related to collector geometry and material, for instance a conductive collector allows dissipation of charge from the fibers and consequently, fibers are able to pack closer, and Doshi and Reneker observed improved collection using a curved metallic screen. Two types of collectors were utilized in this work (Figure 1.13): (1) a parallel electrode collector described in detail by Li et al. to achieve individualized fibers, and (2) an Al foil collector to obtain denser mats. The advantages and disadvantages of each collection type are dependent upon end-purpose: both are fairly simple in design but a foil sheet results in a thick layer of fibers (i.e. comparable to a thin tissue) which are randomly oriented, whereas the parallel electrode collector results in a low deposition density of shorter fibers which are aligned across the electrode gap and are easily transferable to any type of solid substrate.
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Figure 1.13: Diagram of collectors used in electrospinning experiments: A) fibers collect randomly and thickly on Al foil collector and B) fibers align between the metallic electrodes in parallel bar collector.

The use of electrospinning to produce nm-scale diameter fibers is a tremendously useful technique owing to the versatility and simplicity of the method. Fibers may be prepared from virtually any polymer, polymer blend, or polymer nanocomposite (e.g. nanoparticles incorporated into polymer solution). Depending upon conditions and polymer employed, as well as desired application, it may be possible to tailor deposition density, fiber diameter and morphology (e.g. porous, smooth, ribbon-like, bundled, helicoidal). The strong extensional flow results in orientation of macromolecular chains along the long-axis of the fibers (Figure 1.14). Electrospun fibers have been proposed for many different applications, for example as artificial tissue samples or as optical waveguides.
Figure 1.14: Polarized electron micrograph of electrospun fibers which are birefringent as a result of strong elongational forces. These fibers were spun using the following experimental conditions: polymer concentration = 21 wt. %, CNC loading = 2 wt. % relative to polymer mass, voltage = 20 kV, capillary tip-collector distance = 10 cm, flow rate = 1 mL/min.

1.5. Outline of thesis research

Significant research has been and is currently underway in the area of novel cellulose-based materials, with inspirations deriving from the inherent attributes of the cellulosics.

The aim of the research presented in this thesis was to prepare interesting and relevant cellulose-based nanomaterials. In chapters 2-5, fluorescent materials using cellulosics and quantum dots were prepared with the aim of incorporating desirable aspects of each component into the finished product. Chapter 6 is a slight departure from the previous studies of fluorescent materials, with a study of a polyvinyl alcohol (PVA) hydrogels which were reinforced with CNCs but had the same general aim achieving some degree of symbiosis between disparate components, one of which being a cellulosic.

Chapter 2 describes the preparation and characterization of cellulose triacetate films embedded with CdSe/ZnS quantum dots. The bulk properties of the films were largely defined by cellulose triacetate and consequently, the films were hydrophobic,
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pliable, robust and highly transparent. The quantum dots imparted a unique optical signature to the films which was directly derived from the size-dependent properties of the semiconductor nanoparticles. The surfaces of the fluorescent films were converted to cellulose using a saponification reaction in order to make them hydrophilic and compatible with wet-papermaking.

In Chapter 3, the concept of using the fluorescent films as optical taggants in paper sheets was successfully achieved on a laboratory scale. The alkaline treatment used to convert the film surfaces was refined somewhat compared to the hydrolysis conditions reported in the previous chapter. The work established the CTA/QD film system as highly suited for the preparation of specialty or security papers with distinct and controllable fluorescence profiles.

As an extension of the film system, chapter 4 describes fluorescent CTA fibers which were prepared by electrospinning dispersions of QDs in CTA solution. The properties of the fluorescent fibers were similar to the films introduced in Chapters 2 and 3. The fibers are suggested as fluorescent additives in papermaking.

Chapter 5 takes a different approach to fluorescent cellulosics and describes an attempt at covalent bonding of QDs to the reducing ends of cellulose nanocrystals. Working with particles in the same size regime posed some difficulties but also allowed direct observation of the localized attachment using electron beam imaging.

Chapter 6 describes the preparation and characterization of PVA hydrogels which were reinforced with CNCs. PVA hydrogels have water contents similar to biological tissues (~90%) and can be prepared by temperature cycling treatments, without the addition of chemical crosslinking agents. The gel microstructures and morphologies were found to be distinct and dependent upon CNC loading. The mechanical properties of the gels were studied under equilibrium swelling conditions and a reinforcement effect was observed.

The final chapter describes potential future work and applications for the cellulose based nanomaterials presented in this thesis.
1.6. References


Introduction


76. Cao, X.; Dong, H.; Ming Li, C., *Biomacromolecules* 2007, 8 (899-904).


Introduction
Chapter 2
CdSe/ZnS QDs Embedded in Cellulose Triacetate Films with Hydrophilic Surfaces

In this chapter, the incorporation of semiconductor nanoparticles into cellulose triacetate films is discussed. Fluorescent films are prepared which are stable for indefinitely long periods of time at ambient conditions, and mechanically robust. The optical properties of the films and quantum dots are found to be more or less retained after hydrolysis of the film surfaces to cellulose. This study is a first step towards preparing fluorescent films which are compatible with wet papermaking. The work has been published and is reproduced with permission from:

CdSe/ZnS QDs in CTA films
2.1. Abstract

Characterization results are presented for quantum dot (QD)/polymer composite films prepared by solvent casting suspensions of CdSe/ZnS semiconductor nanoparticles in cellulose triacetate (CTA) solution. Direct addition of QDs into CTA film casting solution was possible because of the common solubilities of the QD organic capping ligands and the polymer. The films were robust, with typical thicknesses of ~ 0.05 mm, and possessed optical properties characteristic to the QDs. The peak fluorescence wavelength of the QD/CTA films shifted with time but eventually reverted toward the initial value. This effect is attributed to changes in humidity and solvent content of the films. As inferred from transmission electron micrographs (TEM), the QDs appeared to be well-dispersed within the CTA film matrix. Alkaline hydrolysis of QD/CTA films in 0.1 M NaOH over 24 hours resulted in the surface selective conversion of CTA to regenerated cellulose. The fluorescence of the films was found to be generally unaltered by the hydrolysis treatment. The incorporation of the fluorescent, alkaline treated films into paper sheets is proposed.

2.2. Introduction

The incorporation of semiconductor nanoparticles into polymer matrices is an active field of research motivated in part by the development of novel optical devices. Here we describe the preparation of transparent polymer films embedded with semiconductor nanoparticles. The films exhibit the size-quantized optical properties characteristic of semiconductor nanoparticles.

Semiconductor nanoparticle/polymer composites have been achieved by two general methods: (1) the in-situ synthesis of nanoparticles in either polymer solution or film and (2) the ex-situ synthesis of nanoparticles and their subsequent incorporation into either monomer or polymer solution or film. Experimental approaches to the ex-situ method include the covalent linkage of nanoparticle surface ligands to polymer molecules, layer by layer (LBL) assembly of polymer and nanoparticles and the addition of colloidal nanoparticles directly into polymer solution. Both the in-situ and ex-situ approaches highlight the importance of the organic ligands tethered to
CdSe/ZnS QDs in CTA films

the surfaces of the nanoparticles. The ligands provide the interface between the colloidal nanoparticles and surrounding environment and mediate the nature of the interaction between nanoparticles and a given macromolecular matrix. For example, nanoparticles passivated with hydrophilic ligands will generally be compatible with aqueous systems.

Cellulose is the most abundant and readily available biopolymer.\textsuperscript{22} The solubility of cellulose is limited by its strongly hydrogen-bonded and highly crystalline structure, and as such, in some instances, it may be preferable to work with cellulose derivatives, such as cellulose esters soluble in common organics, or to work with aqueous suspensions of colloidal cellulose nanocrystals.\textsuperscript{23} The use of cellulose or of cellulose derivatives as a matrix for semiconductor nanocrystals has been previously investigated: Ruan et al.\textsuperscript{8} reported the \textit{in situ} synthesis of CdS nanoparticles in cellulose solution and the subsequent casting of CdS/regenerated cellulose films and Yuan et al.\textsuperscript{11, 12} have incorporated CdS nanoparticles into polymer blend membranes (PBMs) where one of the polymeric components of the PBM was cellulose acetate. The rationale behind the current selection of CTA was two-fold: (1) CTA has very good material properties, including high transparency, and (2) CTA may be converted to regenerated cellulose by saponification, if so desired. The solubility of CTA in common organic solvents, such as dichloromethane, allowed for the facile incorporation of appropriately passivated nanoparticles directly into the polymer solution. CTA provided a stable and inert environment for the nanoparticles, encapsulating the particles within a robust, transparent polymer matrix. In order to make hydrophobic QD/CTA films compatible with aqueous systems, the film surfaces were chemically modified to regenerated cellulose. Surface hydrolysis to regenerated cellulose modified the surface properties of the films, while allowing the hydrophobic character of the film bulk to be retained. The fluorescent, hydrolyzed films may potentially be incorporated into paper products by hydrogen-bonding to other cellulose surfaces.
2.3. Experimental

2.3.1 Materials

Cellulose triacetate (43% acetyl content), Congo red (MW 697 g/mol), methanol (spectrophotometric grade) and dichloromethane were purchased from Sigma-Aldrich. Commercial suspensions of CdSe/ZnS QDs in toluene with nominal particle sizes ranging from 1.9-4.0 nm and nominal concentrations of 0.5-1.18 mg/mL were purchased from Evident Technologies. The QDs were capped with either tri-octyl phosphine oxide (TOPO) or with a proprietary 16 carbon linear chain.

2.3.2 Film preparation

A 40 g/L CTA solution was prepared by dissolving CTA in a 9:1 mixture of dichloromethane and methanol. Small volumes (0.1-0.2 mL) of either one or several different sized QDs were added to a 20-25 mL volume of CTA solution. A homogeneous distribution of QDs in the casting solution was achieved by vigorously stirring the mixture for a minimum of 1 hour using a vortex mixer. To cast the film, the mixture was poured into a glass Petri dish, lightly covered with Al foil and the solvent was left to evaporate for approximately 24 hours under ambient conditions. Transfer to the Petri dish was not quantitative due to the high viscosity of the polymer solution. Finally, once the majority of solvent had evaporated, the films were carefully peeled off the glass surface of the dish. The films were robust, transparent, and had thicknesses on the order of 0.05 mm, depending on the amount of casting mixture used and effectively transferred.

2.3.3 Transmission electron microscopy (TEM)

TEM images were obtained of QD/CTA films dried onto carbon coated TEM grids using a Philips CM200 TEM, operated at 200kV, with a point-to-point resolution of 0.24 nm and line resolution of 0.17 nm.

2.3.4 Alkaline hydrolysis

Four hydrolysis conditions were studied: (1) 24 hours in 0.1 M NaOH, (2) 48 hours in 0.1 M NaOH, (3) 24 hours in 2.4 M NH₄OH and (4) 48 hours in 2.4 M NH₄OH.
CdSe/ZnS QDs in CTA films

Film samples were submerged in the alkaline medium, with continuous stirring. Once the treatment was completed, the films were removed from the alkaline bath and rinsed thoroughly and repeatedly under a flow of distilled water to remove any excess base. Drying was performed under ambient conditions.

2.3.5 **Optical characterization**

UV-VIS spectra were obtained using a Cary 300 BIO UV-Vis spectrometer (Varian). Fluorescence spectra were obtained using a FluoroMax-2 fluorimeter (Jobin Yvon-Spex), with excitation wavelengths of 350 nm, and emission and excitation monochromator slit widths of 1 mm and 3 mm, for solutions and films, respectively.

2.3.6 **QD content**

To determine the weight % of QDs in the films, a weighed piece of the QD/CTA film was dissolved in a known volume of 9:1 dichloromethane: methanol. The mass of QDs in the volume of dissolved film was inferred from Beer-Lambert curves of suspensions of QDs in 10g/L solutions of CTA.

2.3.7 **Bulk compositional analysis**

To study the bulk composition of the films, transmission mode Fourier transform infrared spectra of both untreated and alkaline treated films were recorded with a Spectrum BX FTIR spectrometer (PerkinElmer). The FTIR spectrum was an average of 8 scans obtained at a resolution of 4 cm⁻¹.

2.3.8 **Surface compositional analysis**

Congo red film staining, attenuated total reflectance spectroscopy (ATR-FTIR) and X-ray photoelectron spectroscopy (XPS) were performed to compare the surface compositions of untreated and hydrolyzed films. (i) Alkaline treated films were submerged in 4.8 × 10⁻⁴ M solutions of Congo red dissolved in equal parts of water and ethanol. After 15 minutes, the films were removed from the dye solution and rinsed with distilled water in order to wash away surplus stain. (ii) ATR-FTIR spectra were obtained using the MIRacle™ ATR accessory (Pike technologies) in conjunction with a Spectrum
BX FTIR spectrometer (PerkinElmer). All ATR-FTIR spectra were recorded using a
diamond crystal plate and each spectrum was an average of 8 scans with 4 cm\(^{-1}\)
resolution. A correction was applied in order to take into account the higher penetration
depth of the IR beam which occurs at lower frequencies. The depth of penetration \((d_p)\) is
expressed in terms of the wavelength of light \((\lambda)\), the angle of incidence of the IR beam
\((\theta)\) and by the refractive indices of the crystal \((n_1)\) and sample \((n_2)\).\(^{24}\)

\[
d_p = \frac{\lambda}{2\pi(n_1^2 \sin^2 \theta_1 - n_2^2)^{1/2}}
\]  

(2.1)

As an example, given an angle of incidence of 45° and approximate refractive indices of
1.5 for CTA\(^{25}\) and 2.4 for diamond\(^{26}\), the depth of penetration of the IR beam into a CTA
film at 1730 cm\(^{-1}\) is 1.2 \(\mu\)m. (iii) XPS spectra were recorded of the front and back of five
film samples using an AXIS Ultra electron spectrometer (Kratos Analytical), under UHV
conditions and using an Al K\(_\alpha\) source (12.5 kV, 8 mA). Low resolution 0.33 eV or 1 eV
step survey scans provided qualitative information regarding the overall elemental
composition of the film surfaces. The high resolution 0.1 eV scans were used to resolve
the O\(_{1s}\) and C\(_{1s}\) peaks and to quantify the C\(_{1s}\) peak components. Atomic concentration
percentages for the oxygen and carbon components were obtained by applying the
appropriate sensitivity factors \((S = 0.278\) for C\(_{1s}\) and \(S = 0.780\) for O\(_{1s}\)) to the raw peak
areas \((I)\) according to the following equation:

\[
\text{Atomic Concentration percent}_A = \frac{I_A}{S_A} \times \left(\frac{I_B}{S_B} + \frac{I_A}{S_A}\right)^{-1}
\]  

(2.2)

In addition to the film samples, XPS spectra were obtained for a piece of Whatman no.
541 filter paper which had been extracted in acetone using a Soxhlet apparatus. The filter
paper was intended as a pure cellulose reference.

2.4. Results and Discussion

2.4.1 Stability and dispersal of QDs embedded in CTA film

The UV-VIS spectrum of a CTA film embedded with 1.9 nm QDs is presented
in Figure 2.1. The spectra of the 1.9 nm QDs in toluene and in a 10 g/L solution of CTA
are included for comparison. While the characteristic first excitonic peak located at ~ 474 nm was present in all three curves, the absorbance spectrum of the film was comparatively broadened and structureless and the higher energy transition at ~ 430 nm was not at all apparent. Broadening and red-shifting of the QD absorbance spectrum can be understood in terms of the transition from isolated QDs, with localized electronic states, to the delocalized electron-hole states characteristic of nanocrystal clusters or aggregates.\(^{27-29}\) The experimental observation of spectral broadening in the film samples may therefore be reflective of some QD aggregation in the films. However, this mechanism requires the overlap of QD electronic states and is unlikely to be a dominant process in the current system where the presence of polymer and surface ligands will hinder contact between QDs. In fact, polymer is sometimes added to dilute QD suspensions in order to observe the localized states associated with isolated nanoparticles. The noise associated with the solid spectrum may also contribute to the peak broadening, making it somewhat difficult to discern precise peak position and to conclusively state whether or not the absorbance peaks were in fact red-shifted.

Figure 2.1: UV-VIS spectra of 1.9 nm QDs in three environments: (1) in toluene, (2) in a 10 g/L CTA solution and (3) embedded in a CTA film. The concentration of QDs in the solution spectra is 0.0057 mg/mL and the weight percentage of QDs in the film is 0.010 ± 0.002 %.
In general, TEM images showed relatively homogenous distributions of the QDs/small QD clusters within the CTA films. Consistent with the presence of polymer, no evidence of a regular QD packing arrangement or significant aggregation was observed. A representative TEM image of a QD/CTA film is presented in Figure 2.2. The film appeared densely crowded with QDs, which were more or less uniformly dispersed throughout.

Figure 2.2: TEM image of CTA film embedded with 1.9 nm QDs. Concentration of CTA solution is 1 g/L and concentration of QDs in polymer solution is 0.0006 mg/mL.
2.4.2 Fluorescence of films and stability over time

The fluorescence spectra of 1.9 nm QDs in a 10 g/L CTA solution and in a CTA film are presented in Figure 2.3.

![Fluorescence spectra](image)

Figure 2.3: Emission spectra of 1.9 nm QDs in (1) 10 g/L CTA solution and (2) CTA film. The concentration of QDs in the solution spectrum is 0.0133 mg/mL and the weight percentage of QDs in the film is 0.005 ± 0.003%. The inset shows an emission spectrum of a CTA film embedded with 3 different sized QDs: 2.1 nm, 2.6 nm and 4.0 nm with weight percents of 0.0171 ± 0.0004%, 0.017 ± 0.003% and 0.01 ± 0.01%, respectively.

As might be expected from the solution absorbance curves presented in Figure 2.1, the fluorescence of QDs in 10 g/L CTA solution and in toluene overlapped. The emission spectrum of the QD/CTA film had a broad, high energy tail, centered at ~430 nm, which is attributed to the crystallinity of CTA polymer. In general, the CTA emission was found to be negligible in comparison to the highly luminescent QDs but obviously, at sufficiently low QD concentrations, this statement will no longer hold true. QD optical transitions are excited at energies greater than or equal to the band gap, which makes it possible to excite QDs of different sizes using a single wavelength. The inset of Figure 2.3 presents the emission of a CTA film embedded with three different sized QDs, and establishes the QD/CTA system as appropriate for multiplexing purposes. In the
multiplexed films presented in the current work, QDs with different core diameters were incorporated into single films without any size segregation. In this type of highly fluorescent and densely populated film, some light emitted from the smaller QDs will be absorbed by the larger QDs\textsuperscript{3} and Förster resonance energy transfer (FRET) is expected to occur between proximal QDs\textsuperscript{32-35}. In general, the overlap between absorbance and emission dictates the likelihood of both radiative and non-radiative (FRET) energy transfer between QDs. Radiative and non-radiative energy transfer red-shift the emission wavelengths and result in an overrepresentation of the larger sized, redder QDs and an under representation of the smaller sized, bluer QDs in the emission profile. In contrast, segregation of the different QD sizes into separate layers will minimize FRET and allow for greater control of the color observed under UV irradiation.\textsuperscript{3} However, radiative energy transfers through the layers and even within a layer containing a single size distribution of QDs is still possible, as is FRET within a given layer.

The stability of the QD/ CTA films, stored under ambient conditions in covered plastic Petri dishes, was assessed from fluorescence measurements. Fluorescence spectra of a CTA film embedded with 2.1 nm QDs are presented in Figure 2.4: curve 1 was obtained shortly after the film was cast and curves 2 and 3 were taken 60 days and 187 days later, respectively. The fluorescence of the 2.1 nm QDs in toluene and in a 10 g/L solution of CTA are included for comparison. The characteristic, narrow QD emission peak was retained over time, a good indication that the CTA polymer matrix provides a stable and appropriate environment for the QDs. A relatively large overall blue-shift in the peak emission wavelength was observed over time and in comparison to the QD emission in solution: the emission wavelengths of curves 1, 2 and 3 were \textasciitilde 518 nm, \textasciitilde 509 nm and \textasciitilde 495 nm, compared to \textasciitilde 515 nm for the QDs in CTA solution and in toluene. As previously discussed, the initial red-shift of \textasciitilde 5 nm is most likely due to radiative and non-radiative energy transfer between the QDs, which may be further intensified by fixed proximity of the QDs in the films. We propose that the blue-shift in emission position of the film over time can be attributed to evaporation of residual solvent in the film samples. Trapping QDs in non-fluid, rigid media may inhibit relaxation of the excited state, resulting in emission from a higher energy excited state.\textsuperscript{36} Over time, as the solvent gradually evaporates from within the films, the environment becomes more condensed
and the molecules (residual solvent, polymer and ligand) surrounding the QDs and the QD crystal lattice may be less able to undergo the conformational changes necessary to lower the energy of the excited state. It is expected that the emission wavelength will more or less stabilize once solvent evaporation reaches equilibrium, however, for films stored under ambient conditions, humidity will likely continuously affect film environment and consequently emission position. In fact, a fluorescence spectrum of this sample obtained at \( t_{1+260} \) days exhibited a fluorescence peak of \( \sim 513 \) nm, reflecting further minor fluctuations in the film environment. Improved surface passivation of the QDs by CTA may also explain the observed blue-shift in emission wavelength. Finally on this topic, van Sark et al.\textsuperscript{37} studied the emission of continuously irradiated single QDs under ambient conditions and attributed the resulting irreversible blue-shift and bleaching to photooxidation of the CdSe core. Oxidation of the CdSe core may seem more likely compared to emission from a higher energy excited state, but it does not address the apparent reversibility of the trend. Admittedly though, the conclusion of reversibility hinges on the consistent measurement of the same area of a given film, and while this was attempted, it is possible that the results may be attributed to a small shift in the emission area. Other films were of course studied but over shorter time periods. Most films exhibited a blue-shift of around 5-10 nm after a month and \( \sim 30 \) nm after 6 months, but some exhibited a smaller red-shift over a similar time frame.
Figure 2.4: Effect of time on the emission of a 2.1 nm QD/CTA film with 0.006 ± 0.003% QD content by weight. Curve (1) was obtained at time \( t \), (2) at \( t + 60 \) days and (3) at \( t + 187 \) days. The inset depicts the overlapping emission spectra of 2.1 nm QDs dispersed in toluene and in 10 g/L CTA at a concentration of 0.021 g/L.

### 2.4.3 Hydrolysis of fluorescent films

A saponification reaction, depicted in Figure 2.5, was utilized to hydrolyze the film surfaces to cellulose. Four alkaline hydrolysis conditions were studied: 24 hour and 48 hours in 0.1 M NaOH, and 24 and 48 in 2.4 M NH₄OH.

Figure 2.5: Alkaline hydrolysis of cellulose triacetate to cellulose. Four room temperature conditions were studied: (1) 24 hours in 2.4 M NH₄OH, (2) 48 hours in 2.4 M NH₄OH, (3) 24 hours in 0.1 M NaOH and (4) 48 hours in 0.1 M NaOH.
2.4.4 Staining of hydrolyzed films with Congo Red

Congo red dye has a high affinity for cellulose and can be used as a qualitative measure for the degree of acetyl desubstitution.\(^{38, 39}\) The azo moieties of Congo red hydrogen bond with the hydroxyl groups of cellulose, staining cellulose film surfaces red, while no analogous interaction exists between Congo red and CTA. The Congo red absorbance peak at \(\sim 515\) nm was used as a marker for the hydrolysis reaction; the more stained a film surface appears, the greater the extent of CTA hydrolysis. From the curves presented in Figure 2.6, it was apparent that all treatment conditions resulted in some degree of CTA deacetylation, with the CTA films treated in \(0.1\) M NaOH experiencing the most significant conversion to cellulose. However, little information was provided from the Congo red results regarding the depth of hydrolysis.

![Absorbance Spectra](image)

Figure 2.6: UV-VIS absorbance spectra of CTA films which have been treated in alkaline and stained with Congo red dye. Hydrolysis conditions: (1) 24 hours in \(2.4\) M NH\(_4\)OH, (2) 48 hours in \(2.4\) M NH\(_4\)OH, (3) 24 hours in \(0.1\) M NaOH and (4) 48 hours in \(0.1\) M NaOH.

2.4.5 Infrared analysis

To better assess the depth to which the samples were deacetylated, ATR-FTIR analysis was performed on QD/CTA films which had undergone the previously described
hydrolysis treatments. Infrared absorbance can be used to differentiate between cellulose acetates and regenerated cellulose \textsuperscript{38-41} since the carbonyl stretch at \( \sim 1730 \text{ cm}^{-1} \) is absent in the cellulose spectrum whereas the hydroxyl group absorbance at \( \sim 3300-3450 \text{ cm}^{-1} \) is increased. IR spectra of films embedded with QDs appeared identical to spectra of CTA films which did not incorporate the QDs. A diamond crystal plate was used in the ATR-FTIR experiments. The depth of penetration \( (d_p) \) of the IR beam into the sample is wavelength dependent and varies from 0.5 \( \mu \text{m} \) to 4 \( \mu \text{m} \) for the given system, in the spectral range of 4000 \text{ cm}^{-1} \) to 565 \text{ cm}^{-1}. The spectrum is therefore representative of the average composition of the sample at surface depths ranging from 0.5 \( \mu \text{m} \) to 4 \( \mu \text{m} \). From the IR results presented in Figure 2.7, in particular as indicated by carbonyl stretch at 1730 \text{ cm}^{-1}, we conclude the samples treated with 0.1 M NaOH were on average composed of cellulose to a depth of at least 1.2 \( \mu \text{m} \) into the film surfaces, and the condition of 2.4 M NH\(_4\)OH was insufficient for deacetylation, even at the relatively shallow depth of 1.2 \( \mu \text{m} \). We reported\textsuperscript{39} the successful deacetylation of CTA films in 2.6 M NH\(_4\)OH, but the previous work described shear-cast CTA films that were much thinner than the solvent cast films of the current study. The Congo red and ATR-FTIR results were complimentary; treatment in 0.1 M NaOH over 24 hours was sufficient for surface deacetylation with little gained by an additional 24 hours of treatment and, in comparison to the NaOH conditions, reaction in NH\(_4\)OH was not extensive. Tables 2.1 and 2.2 include a detailed assignment of the infrared absorption peaks for the CTA and regenerated cellulose films.\textsuperscript{8, 40-44}
Figure 2.7: ATR-FTIR spectra of alkaline treated CTA film embedded with 1.9 nm QDs (0.010 ± 0.002 wt. %). Hydrolysis conditions: (1) untreated film, (2) 24 hours in 2.4 M NH₄OH, (3) 48 hours in 2.4 M NH₄OH, (4) 24 hours in 0.1 M NaOH and (4) 48 hours in 0.1 M NaOH. The spectra of the samples treated in NaOH are characteristic of cellulose.

Table 2.1: Important infrared absorption bands for CTA films.

<table>
<thead>
<tr>
<th>Wavenumber (cm⁻¹)</th>
<th>Band assignment</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>890</td>
<td>δC-H</td>
<td>41</td>
</tr>
<tr>
<td>1024</td>
<td>νC-O-C (pyranose)</td>
<td>40, 41, 44</td>
</tr>
<tr>
<td>1210</td>
<td>νC-O-C (ester)</td>
<td>40, 41, 43</td>
</tr>
<tr>
<td>1365</td>
<td>δC-H</td>
<td>41, 44</td>
</tr>
<tr>
<td>1420</td>
<td>δCH₃ (α)</td>
<td>41, 44</td>
</tr>
<tr>
<td>1730</td>
<td>νC=O</td>
<td>40, 41, 43</td>
</tr>
<tr>
<td>2872 &amp; 2932</td>
<td>νC-H</td>
<td>40, 41, 43, 45</td>
</tr>
<tr>
<td>3460</td>
<td>νO-H</td>
<td>41, 44</td>
</tr>
</tbody>
</table>
Table 2.2: Important infrared absorption bands for regenerated cellulose films.

<table>
<thead>
<tr>
<th>Wavenumber (cm(^{-1}))</th>
<th>Band assignment</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>600</td>
<td>δ(_{\text{O-H}})</td>
<td>42</td>
</tr>
<tr>
<td>890</td>
<td>δ(_{\text{C-H}})</td>
<td>8, 41, 42</td>
</tr>
<tr>
<td>988</td>
<td>ν(_{\text{C-O-C}}) (pyranose)</td>
<td>8, 40, 42, 43</td>
</tr>
<tr>
<td>2850</td>
<td>ν(_{\text{C-H}})</td>
<td>42, 43</td>
</tr>
<tr>
<td>3300</td>
<td>ν(_{\text{O-H}})</td>
<td>8, 42, 43</td>
</tr>
</tbody>
</table>

The FTIR transmission spectra presented in Figure 2.8 show that the bulk of the deacetylated samples remained cellulose triacetate, as indicated by the persistence of the carbonyl absorbance at all reaction conditions. Thus, the hydrolysis reaction, as inferred from the IR results, occurred only in the surface region and did not significantly penetrate the films. The spectral interpretation presented in Tables 2.1 and 2.2 still applies, although the peaks located at frequencies lower than \(\sim 1500\) cm\(^{-1}\) were not very well resolved.

Figure 2.8: FTIR spectra of alkaline treated CTA film embedded with 1.9 nm QDs (0.010 ± 0.002 wt. %). Hydrolysis conditions: (1) untreated film, (2) 24 hours in 2.4 M NH\(_4\)OH, (3) 48 hours in 2.4 M NH\(_4\)OH, (4) 24 hours in 0.1 M NH\(_4\)OH.
CdSe/ZnS QDs in CTA films

NaOH and (5) 48 hours in 0.1 M NaOH. Regardless of treatment conditions, all samples exhibit a spectrum characteristic of cellulose triacetate.

2.4.6 Fluorescence of films post hydrolysis

In Figure 2.9 the emission of a film which has been hydrolyzed in 0.1 M NaOH for 24 hours is compared to the emission of the same film prior to alkaline treatment. The apparent increase in intensity after NaOH treatment was not significant; the variation in the emission curves is attributed to the uneven thicknesses of the solvent cast films. For example, when the thickness of a single film embedded with 1.9 nm QDs was measured 46 times at random film locations, it was found to vary from 0.13 mm to 0.036 mm, with an average thickness of 0.071 ± 0.007 mm. In general, an increase in the emission intensity and peak area with film thickness was observed. Average film thickness decreased slightly upon 0.1 M NaOH hydrolysis, but the change in emission, if any, was difficult to quantify due to the large variation associated with the fluorescence measurement of a small film area (approximately 1 cm²) of a given average thickness. Interestingly, the emission wavelength post alkaline treatment was blue-shifted compared to the wavelength prior to hydrolysis; the aforementioned film incorporating 1.9 nm QDs experienced a pronounced decrease in average film thickness to 0.019 ± 0.003 mm and a shift in emission wavelength from ~501 nm to ~483 nm. As previously discussed, the blue-shift may be due to emission from a higher energy excited state as a consequence of the reduction in film thickness upon hydrolysis (i.e. the film is densified) or to some degree of core degradation.
Figure 2.9: Emission of film embedded with QDs before alkaline treatment, (1), and post treatment in 0.1 M NaOH, (2). The film is embedded with 3 different sized QDs: 2.1 nm, 2.6 nm and 4.0 nm with weight percents of 0.017 ± 0.004%, 0.017 ± 0.003% and 0.01 ± 0.01%, respectively.

Quenching of QD emission was observed for the films treated in 2.4 M NH₄OH. To further assess the effect of alkaline on QD emission, small amounts of QD suspensions were added to 0.1 M NaOH and to 2.4 M NH₄OH solutions, and a film embedded with 2.1 nm QDs was suspended above a concentrated bath of NH₄OH. As indicated by observation under UV-light, the fluorescence of all three samples was entirely quenched within a 24 hour period. It therefore seems reasonable to conclude that exposure to alkali quenches QD emission, and that the CTA polymer matrix adequately shields the embedded QDs from quenching by NaOH. However, the fluorescence of films exposed to aqueous NH₄OH and to ammonia vapor was quenched, indicating that the penetration of NH₄OH/ammonia into the films is significant compared to NaOH treatment. Hence, all subsequent deacetylation reactions were performed with 0.1 M NaOH over a period of 24 hours.

2.4.7 XPS analysis

XPS was used to estimate the surface conversion of CTA to cellulose. The fronts

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CdSe/ZnS QDs in CTA films

and backs of six different samples were analyzed: a CTA film, a CTA film embedded with 1.9 nm QDs (QD content estimated at 0.02 weight %) referred to as film C, a deacetylated CTA film, a sheet of Whatman no.1 filter paper and two deacetylated films embedded with 2.1 nm QDs, designated deacetylated films A and B. Deacetylated films A and B were cast from the same suspension of QDs in polymer and therefore had an identical weight % of QDs in CTA, 0.023 ± 0.009%. From the survey scan results of all six samples, the only elements present in significant concentrations were carbon and oxygen. Trace amounts of silicon were detected in most samples, excluding the front surface of the hydrolyzed CTA film, the filter paper and the back surface of the CTA film. Interestingly, XPS did not detect the inorganic elements Cd, Se, Zn, and S which comprise the QDs, which may be taken as evidence for the successful incorporation of QDs well within the film bulk or may indicate that the low concentration of QDs within the photoelectron escape depth is beyond detection. The survey results may however include a contribution from the hydrocarbon QD ligands to the C(1s) signal.

To attempt a quantitative differentiation of samples, the contributions of the molecular species to the total oxygen O(1s) and carbon C(1s) XPS signals, and to the component C1, C2, C3 and C4 carbons were considered. The labels C1, C2, C3 and C4 refer to a carbon with no oxygen bonds, a carbon with a single oxygen bond, a carbon with two oxygen bonds and a carbon with three oxygen bonds, respectively. The atom concentrations and binding energies of the C1, C2, C3 and C4 peaks are obtained by resolving the overall carbon 1s peak into its component carbons. In the high resolution scans of the cellulose samples, a small tail (in all cases ≤1.95 atomic concentration percent) was observed at a slightly up-field bonding energy from the main C3 peak. The small tail was considered to be part of the C3 peak and not a minor C4 peak. Similarly, for the CTA samples, two nearly overlapping peaks were combined to obtain the C1 atomic concentration percentage.

The ratios O/C and C1/(C2+C3+C4) are characteristic of either cellulose or CTA. Cellulose has 5 oxygens and 6 carbons per repeat unit (O/C = 0.83) and, with three acetyl groups per unit, CTA has 8 oxygens and 12 carbons per repeat unit (O/C = 0.67). In addition, the C1/(C2+C3+C4) ratio is 0 for cellulose with no C1 carbons, but 0.33 for CTA with a degree of acetyl substitution of 3. In our commercial CTA sample with a
percent acetyl content of 43.38%, the actual number of acetyl groups and hydroxyl
groups per repeat unit is 2.906 and 0.094, respectively. The corresponding numbers of
total carbons, total oxygens and C1 carbons per repeat unit are 11.81, 8.0 and 2.906,
respectively, giving a theoretical O/C ratio of 0.68 and a C1/(C2+C3+C4) ratio of 0.326.

Surface contamination by hydrocarbon impurities often causes the measured O/C
values to be lower (and C1 values to be higher) than expected.\(^{47}\) The XPS results are
presented in Table 2.3. Results for the filter paper and CTA were in reasonable agreement
with literature values\(^{46, 48}\), but deviated from the theoretical values in the manner
expected for the presence of carbon-rich material at the surface.

Table 2.3: Atomic concentration percentages obtained from XPS of 5 cellulosic samples.
The theoretical values for CTA were calculated for commercial CTA with an acetyl
content of 43.38%. Samples A and B are alkaline treated CTA films embedded with 2.1
nm QDs at a weight percent of 0.023 \(\pm\) 0.009%. The hydrolyzed films were all treated in
0.1 M NaOH over 24 hours. Film C is an untreated CTA film which has 1.9 nm QDs
dispersed within at a weight percent of 0.02%.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Atomic concentration percents</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C</td>
</tr>
<tr>
<td>Filter paper</td>
<td>57.4 ± 0.4</td>
</tr>
<tr>
<td>Hydrolyzed CTA</td>
<td>61 ± 1</td>
</tr>
<tr>
<td>Hydrolyzed A</td>
<td>63.4 ± 0.7</td>
</tr>
<tr>
<td>Hydrolyzed B</td>
<td>64 ± 1</td>
</tr>
<tr>
<td>CTA</td>
<td>63.8 ± 0.4</td>
</tr>
<tr>
<td>Untreated C</td>
<td>63.7 ± 0.9</td>
</tr>
<tr>
<td>Theoretical C(_6)O(_5)</td>
<td>54.55</td>
</tr>
<tr>
<td>Theoretical C(_{12})O(_8)</td>
<td>59.62</td>
</tr>
</tbody>
</table>

The XPS evidence for surface carbon-rich contaminants was more marked for the
hydrolyzed CTA films and particularly for the samples containing quantum dots. The
ATR-FTIR evidence discussed above suggests that the surfaces of the hydrolyzed films
CdSe/ZnS QDs in CTA films

were essentially pure cellulose, but the observed O/C ratios (0.57-0.65) for the much thinner surface layer sampled by XPS indicated the presence of carbon rich-material. The nature of the material is not known in the case of the hydrolyzed cellulose acetate, but we speculate that some of the TOPO or hydrocarbon surfactants used to stabilize the QD suspensions may leach to the cellulose surface. However, regardless of QD content, the hydrolyzed films experienced the most significant surface contamination because of the increased manipulation of these films during alkaline treatment.

2.5. Conclusions

Stabilized CdSe/ZnS semi-conductor nanoparticles may be incorporated in cellulose triacetate films with essentially unaltered fluorescence characteristics. The solvent-cast films protect and facilitate handling of the quantum dots, and surface hydrolysis of the CTA should facilitate utilization of the films in aqueous media.

2.6. Acknowledgements

We thank NSERC Canada and Paprican for financial support, the Centre for Self-Assembled Chemical Structures (CSACS) for laboratory access, A. Lejeune (UQTR) for XPS data acquisition and Xue Dong Liu (McGill) for TEM imaging. Professional insight from P. Kambhampati is acknowledged.

2.7. References

Chapter 2

CdSe/ZnS QDs in CTA films

44. Kondo, T.; Sawatari, C., Polymer 1996, 37 (3), 393-399.
CdSe/ZnS QDs in CTA films
The fluorescent films which were developed in the previous chapter are incorporated into paper with the aim of creating an optical security feature for paper and packaging products. The alkaline hydrolysis of the films is improved, converting only a very thin surface layer of the films to cellulose. The quantum yields of the QDs in polymer solution are found to be diminished compared to the initial colloidal quantum dots. The hydrolyzed films are shown to be compatible with wet papermaking and adhere to pulp fibers, likely through hydrogen bonding interactions. Paper sheets speckled with fluorescent film pieces are successfully prepared. The work has been published and is reproduced with permission from:

Fluorescent CTA films in paper
3.1. Abstract

CdSe/ZnS quantum dots (QDs) were embedded in films of cellulose triacetate (CTA) to give clear films with the broad absorbance and well-defined, size-tunable fluorescence characteristic of QDs. A decrease in quantum yield upon dispersal of QDs in CTA solution was observed. Alkaline hydrolysis of the film surfaces to regenerated cellulose converted the previously hydrophobic CTA film surfaces hydrophilic and compatible with aqueous papermaking. Films containing combinations of different sized QDs gave more complex emission profiles, with peak areas somewhat dependent upon excitation wavelength. Small pieces of fluorescent films were added to pulp slurries and incorporated into laboratory paper sheets through hydrogen bonding interactions between the regenerated cellulose film surfaces and cellulosic pulp fibers. The film system (cellulose ester bulk/ cellulose surface) can be used to incorporate hydrophobic particles or molecules compatible with solutions of cellulosic polymers into paper products at both high and low loadings. QDs in paper may prove useful for security applications, such as in sheets which possess unique optical signatures.

3.2. Introduction

The optical properties of quantum dots include broad absorbance, size-tunable fluorescence and relative insensitivity to photobleaching.\(^1\), \(^2\) These characteristics contrast with standard organic dyes which often have narrow absorbance bands, broad, asymmetric fluorescence and may be susceptible to photobleaching. QDs are inherently suited for use in systems which incorporate a mixture of different sized QDs since a single wavelength may be used to excite the emission bands of each QD size, resulting in a spectrum with multiple fluorescent peaks. Multiplexed quantum dot/polymeric systems have been explored in the literature, often in regard to light emitting diodes (LEDs) where different sized quantum dots are combined in order to achieve a specific colored or white light.\(^3\), \(^4\) Similar systems have also been proposed for spectral coding technologies where sets of inks made from ratios of different sized QDs in polymeric matrices are used to encode data.\(^5\)
The commercial semiconductor nanocrystals used in this study were composed of a CdSe core and a ZnS outer layer(s), with nominal core diameters ranging from 2.1-.2 nm and corresponding fluorescence wavelengths of approximately 505-610 nm. Cellulose triacetate (CTA), a cellulose ester where the hydroxyl groups of cellulose are replaced by acetyl groups, in this case with a degree of acetyl substitution of 2.9 groups per anhydroglucose unit, was employed as the film matrix for the QDs. Cellulose esters are derived from cellulose, a ubiquitous, renewable, plant-based resource. Cellulose esters are non-toxic, biodegradable under certain conditions, and highly versatile, with applications ranging from drug delivery systems to optical films. Replacement of hydroxyl groups with acetyl groups disrupts the H-bonded structure of cellulose and significantly alters the properties of the material: for example, while the highly H-bonded and crystalline structure of cellulose requires strongly-interacting solvents such as LiCl/DMAc for dissolution, cellulose esters readily dissolve in common organic solvents and are hydrophobic. CTA is easily cast into robust films which possess high clarity, transparency in the visible spectral range, good surface smoothness and film uniformity, and are insensitive to changes in relative humidity. CTA films are therefore ideal for optical film and liquid crystal display (LCD) applications. The characteristics of CTA films also make them appropriate as matrices for semiconductor nanocrystals, for example, chemical compatibility aside, the films are highly transparent in the visible spectral region where the QDs emit. The functional QD/CTA film system is inherently interesting by virtue of the complimentary and desirable optical properties of each component. It is possible to imagine many useful applications stemming from the QD/CTA system and in this paper, we put forward a simple method for the incorporation of QD/CTA films into paper products.

For use as optical taggants in paper sheets, the optical properties of the QDs must be retained throughout the papermaking process and the lifetime of the finished paper products. Embedding QDs within a polymer matrix may shield the nanoparticles from potential degradation but for this approach to work, the polymer itself must not adversely affect the properties of interest. We recently reported the preparation, optical and compositional characterization of cellulose films embedded with CdSe/ZnS semiconductor nanoparticles. Briefly, film preparation involved the addition of...
hydrophobic colloidal nanoparticles into a cellulose triacetate (CTA) solution and the subsequent casting of high clarity, robust films which possessed the optical characteristics of the quantum dots. Selective alkaline hydrolysis of the cellulose triacetate films resulted in the surface conversion of cellulose triacetate to cellulose, producing an interesting film system, where the film bulk is hydrophobic, the film surfaces are hydrophilic and hydrophobically-passivated fluorescent nanoparticles are embedded throughout. Here we present the incorporation of these films into paper sheets through hydrogen bonding interactions between film surfaces and cellulose pulp fibers. The indirect addition of quantum dot taggants into paper sheets using a cellulosic carrier does not significantly diminish the optical integrity of the taggant and may therefore be useful in the preparation of novel types of fluorescent papers.

3.3. Experimental

3.3.1 Materials

Cellulose triacetate (43% acetyl content, 103, 000 g/mol) was purchased from Sigma-Aldrich. Suspensions in toluene of CdSe/ZnS QDs capped with tri-octylphosphine oxide (TOPO), with nominal sizes of 2.1-6.0 nm, concentrations of 1.17-1.77 mg/mL and approximate quantum yields of 30-50% were purchased from Evident Technologies, Inc. Rhodamine B from Chroma-Gesellschaft (Schmid & Co.) was used. Handsheets were prepared from non-fluorescent, photo-grade Nexfor low-yield sulphite pulp sheets obtained from Fraser Papers, Thurso, QC.

3.3.2 Film preparation

QD suspensions in toluene were directly added to 40 g/L CTA solution (9:1 methylene chloride: methanol). The mixture was shaken for at least one hour prior to film casting using a Vortex mixer. Films incorporated into paper sheets were surface deacetylated in 0.05 N NaOH for 24 hours. Films were also spin-coated from 0.5 g/L CTA in methylene chloride onto glass substrates.
3.3.3 Spectroscopy

Fluorescence spectra were recorded on a FluoroMax-2 fluorimeter (Jobin Yvon Spex). Film spectra were obtained at 450 nm excitation, unless otherwise noted, and solution spectra with 514 nm excitation. UV-VIS spectra were obtained with a Cary 300 BIO UV-Vis spectrometer (Varian). ATR-FTIR spectra were obtained using the MIRacle™ ATR accessory (Pike technologies) in conjunction with a Spectrum BX FTIR spectrometer (PerkinElmer). All ATR-FTIR spectra were recorded using a diamond crystal plate and each spectrum was an average of 16 scans with 4 cm⁻¹ resolution.

3.3.4 Relative quantum yield

We attempted to quantify the change in quantum yield (QY), if any, which occurs when the QDs, which are initially suspended in toluene, are dispersed in polymer solution. To do this, integrated areas under the fluorescence curves (520-675 nm) were plotted against absorbance at excitation wavelength (514 nm) and the change in QY, represented as a percent decrease, was determined from equation 1:

\[
\frac{\Phi_{QD}(CTA)}{\Phi_{QD}(toluene)} = \frac{m_{QD}(CTA)}{m_{QD}(toluene)} \cdot \frac{n_{CTA}^2}{n_{toluene}^2}
\]

where, \( \Phi \) is the QY of the QDs, dispersed in CTA solution or toluene, \( m \) is the slope of plots of the fluorescence areas against absorbance at 514 nm, and \( n \) is the refractive index of CTA solution or toluene. The refractive indices were not measured but given a literature range of 1.45 -1.5 for CTA, and a value of 1.496 for toluene, the ratio of indices was assumed to be ~1. Rhodamine B, a standard organic dye, was employed as a general check (e.g. for lamp power) and the 514 nm wavelength was selected to excite the fluorescence of both Rhodamine B and the QDs. The experiment was repeated three times using fresh QD suspensions, dispersed in toluene and in 10 g/L CTA solution, and with Rhodamine B dissolved in absolute ethanol. Associated errors were derived from regression statistics. The final relative quantum yields are reported as a range of the three trials.
3.3.5 **Fluorescent paper**

Handsheets were prepared from alkaline treated films, which were either coarsely ground using a Wiley mill (~0.5 mm fragments), cut into small pieces (triangles with ~2 mm sides) or cut into slivers (length ~10 cm, width ~0.5 mm) with a blade. The weight percent of film to fiber in the sheets was varied from 1% to 60%. Sheets were prepared by adding either the coarsely ground films or the small cut pieces to a pulp slurry and stirring until the mixture was well dispersed. The dispersion was then filtered through a fine grid to yield sheets of 5 cm in diameter. Higher basis-weight handsheets which incorporated slivers of film were made using a laboratory-scale former to give 15 cm diameter sheets.

3.4. **Results and Discussion**

3.4.1 **Quantum yields**

In our previous work, the optical efficiency of the quantum dots dispersed in toluene and in CTA polymer was not addressed. We attempt to quantify the change in quantum efficiency which occurs when the QDs are dispersed in CTA compared to toluene, relative to the performance of a standard dye, Rhodamine B. One point in the quantum yield determination for the 5.2 nm QDs is presented in Figure 3.1: a decrease of ~20% in relative efficiency for the QD suspension in CTA solution compared to toluene was observed.
Figure 3.1: Fluorescence ($\lambda_{\text{EXC}} = 514$ nm) of Rhodamine B in ethanol, 5.2 nm QDs dispersed in toluene and 5.2 nm QDs in 10 g/L CTA. The absorbance of each sample at the excitation wavelength was matched at ~0.02.

The percent decreases in quantum yield for three different sized QDs upon dispersal in 10 g/L CTA are presented in Table 3.1. The QDs experienced a decrease in efficiency ranging from ~5-50% when dispersed in 10 g/L CTA solution. The large variation may be an indicator of the inherent difficulty of accurate quantum yield measurement, especially in this case where a polymer solution is employed, but regardless, it seems that the optical performance of the QDs is to some degree adversely affected by dispersal in polymer solution.

Table 3.1: Percent decrease of quantum yields (relative to Rhodamine B in absolute ethanol) on dispersal of QDs in 10 g/L CTA.

<table>
<thead>
<tr>
<th>QD diameter (nm)</th>
<th>% decrease in quantum yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>2.6</td>
<td>6-29</td>
</tr>
<tr>
<td>4.0</td>
<td>10-38</td>
</tr>
<tr>
<td>5.2</td>
<td>37-50</td>
</tr>
</tbody>
</table>
3.4.2 Fluorescence and absorbance

QD/CTA films are clear, robust, with film thicknesses of ~0.05 mm, and appear fluorescent under UV illumination (Figure 3.2).

Figure 3.2: QD/CTA films incorporating QDs with core diameters of (from left to right) 2.1 nm, 2.4 nm, 5.2 nm and 6.0 nm under UV illumination.

The fluorescence and absorbance are determined by the size of the QDs embedded within the films (Figure 3.3) (Note that the nominal QD size is actually an average of some size distribution).

Figure 3.3: Normalized fluorescence ($\lambda_{\text{exc}} = 350$ nm) of QD/CTA films incorporating QDs with core diameters of 2.1 nm, 2.4 nm, 5.2 nm, and 6.0 nm. Figure inset shows absorbance spectrum of 2.1 nm QDs. The concentration of QDs in the casting solution was approximately 0.01 mg/mL for all films.
We previously observed a blue shift in the emission wavelength of the QDs in CTA film compared to in solution. This effect is attributed to the density of the polymer environment surrounding the QDs. The encapsulating CTA may improve the surface passivation of the QDs by removing surface trap states which would otherwise red-shift the emission wavelength. The blue-shift might also be due to oxidation of the CdSe core or perhaps to emission from a higher energy excited state. To create multiplexed systems, either different sized QDs were added to a single film casting solution or layered structures were made by stacking or by spin-coating films incorporating a single size of QDs. In layered or sandwich type structures, distance dependent energy transfer from the smaller to the larger QDs is minimized but reabsorption of the light emitted from the layers incorporating the smaller QDs by the QDs in other layers is still possible. The order of the layers is non-trivial and stacking a film embedded with smaller sized QDs either above or below a film containing larger sized QDs will obviously give different results. Practically, it may be easier to control the color of the sandwich type films under UV illumination.

3.4.3 Homogeneity of multiplexed films

Some interesting information may be inferred from the ratio of emission peaks observed at different film locations in a given multiplexed film. The solvent casting technique results in films having non-uniform thicknesses and consequently non-uniform optical properties. For example, when three films cast using the same volume of a single casting solution, were each cut into four pieces, the thicknesses of the 12 resulting pieces of film were found to vary from 0.018 ± 0.005 mm to 0.065 ± 0.006 mm, and the ratios of the emission peak areas of the larger QDs to the smaller QDs ranged from approximately 1:1 to 2:1. Our previous TEM studies suggest that the intimate mixing of the nanoparticles and the polymer is indeed occurring, and therefore we expect that the ratio between emission peaks is expected to be more or less constant for films of low optical density. The result of an experiment using a typical solvent cast film is presented in Figure 3.4. Variations in emission wavelength (i.e. red-shifting at some film positions) are attributed to varying degrees of radiative energy transfer caused by the non-uniform film thickness. In general, the % deviation from the average peak ratios ranged from ~10-
75% with most films lying in the lower end of the range. Not surprisingly, the most consistent peak ratios were observed in films with the lowest optical densities either because the films had lower overall thicknesses and/or lower concentrations of QDs. To minimize the effects of non-radiative and radiative energy transfer between different size distributions of QDs, experiments were performed with spin-coated films where the QD sizes are segregated into different film layers. This type of film is made by 5 successive spin-coating steps: (1) 5 layers of CTA, (2) 25 layers of CTA incorporating QDs, (3) 5 layers of CTA, (4) 25 layers of CTA incorporating a second size of QD, and (5) 5 CTA layers. The spin-coated films are approximately an order of magnitude thinner than films prepared by solvent casting and the intercalating CTA layers prevent the mixing of nanoparticles at the interface between layers. The spin-coated films had a spread in peak ratio of ~20% (e.g. 1.3 ± 0.3), a value which falls in the lower range observed for solvent cast films. This value was higher than expected and is attributed to non-uniform film deposition; under UV observation it appeared that the films were in fact thicker at the center compared to the edges. In general, the suggestion we are making is that a large spread in peak ratios observed at multiple locations on a given film is not indicative of nanometer scale compositional inhomogeneities but rather of non-uniform optical densities due to non-uniform thicknesses. It should be noted that solid state fluorescence is often non-linear and that scattering will be more pronounced in the solid state compared to dilute solution fluorescence, in particular because of the presence of dust particulates which are difficult to filter from a viscous polymer solution.
Figure 3.4: Fluorescence peaks for a film containing both 2.1 and 4.0 nm QDs at 5 different positions. The concentrations of QDs in the film casting solution were 0.004 mg/mL for the 2.1 nm QDs and 0.006 mg/mL for the 4.0 nm QDs. The ratios of integrated peak areas (area under each curve from 476 to 545 nm divided by area from 546 to 674 nm) are indicated in the key.

3.4.4 Fluorescence of multiplexed films

The broad absorption of QDs means that changing the excitation wavelength will affect film emission since different wavelengths will be absorbed to varying extents. In order to study this effect, the fluorescence of multiplexed films was recorded at a single film location while sweeping the excitation wavelengths. The changes in the ratio of emission peak areas which occurred with varying excitation were calculated (Figure 3.5). The results indicate that the fluorescent color of multiplexed films is not solely determined by the film composition but also by the wavelength of exciting light.
Figure 3.5: Effect of excitation wavelength on fluorescence from a single point on a film containing both 2.1 and 5.2 nm QDs. Shown are ratios of integrated peak areas corresponding to each size of QDs (area under each curve from 476 to 545 nm divided by area from 546 to 674 nm). The inset shows typical fluorescence intensity curves for two excitation wavelengths.

3.4.5 Hydrolysis of films

The general motivation for this study was to develop a vehicle capable of depositing functional, hydrophobic particles onto pulp fibers for their eventual incorporation into paper sheets. The QDs used in this study are solubilized by organic surfactants which are tethered to their surfaces. The hydrophobic nature imparted to the QDs by the surfactants makes their direct use in aqueous papermaking problematic. Embedding hydrophobic particles, such as organically-passivated QDs, into a hydrophobic cellulosic matrix followed by the surface conversion of the matrix to cellulose provides a simple route for QD incorporation into paper sheets. To convert the CTA film surfaces to regenerated cellulose, the films were treated in 0.05 M NaOH over a 24 hour period. Hydrolysis to cellulose is confirmed by the disappearance of the carbonyl stretch at ~1730 cm\(^{-1}\) in the infrared spectrum. The concentration of alkali was half that which was used in our previous work but produced similar results, with the depth of hydrolysis penetrating at least 1.2 µm into the films surfaces (Figure 3.6). The carbonyl stretch persisted in measurements where the IR beam penetrated the breadth of
the film, indicating that the hydrolysis reaction is indeed limited to a shallow surface depth.

![Graph](image.png)

Figure 3.6: Attenuated total reflectance (ATR)-Fourier transform infrared spectra of CTA films treated with different concentrations of aqueous NaOH.

### 3.4.6 Incorporation of films into paper

Surface hydrolysis to cellulose does not seem to have a large effect upon the optical properties of the films but significantly, does to some degree impart the insolubility characteristic of cellulose. The cellulose surfaces of the films will hydrogen bond with pulp fibers and allow the films to be drawn into the wet web as the sheet forms. Experimentally, particles of the fluorescent film, with sizes ranging from approximately 0.1-1 mm produced by grinding the initial film, were added to an aqueous suspension of cellulose pulp fibers. The film particles adhered to the fibers, covering sections of the fibers with the fluorescent films. Figures 3.7 and 3.8 show UV illuminated photographs of sheets which incorporate QD/cellulosic film pieces. The multicolored fluorescent response of the QD films in the sheet is clearly evident. Many combinations can be made working with a size series of QDs which emit at discrete, non-overlapping
wavelengths. Films can be made which incorporate one or more different sizes of QDs in different ratios, and sheets can be made using varying amounts of one or more of these films. The relatively simple and versatile system of cellulosic polymer embedded with QDs permits the straightforward preparation of a range of optical taggants compatible with the manufacture of paper and board products. In the course of our work, wet-laid sheets have been prepared having a wide range of film loadings, anywhere from ~1-60% by weight. The film pieces seem to be uniformly dispersed within the sheet. The fluorescent properties of the films containing embedded QDs are unusually stable, with shelf lives under ambient conditions currently in excess of three years. Importantly, it is the CTA matrix which seems to preserve the stability of the QDs.

Figure 3.7: Photographs of paper sheet incorporating cellulose surface/CTA bulk film pieces (~2 mm sides) embedded with 2.1 nm, 2.6 nm, 5.2 nm or 6.0 nm QDs under ambient visible (left) and UV (right) lighting conditions. Weight percent of film in sheet was ~60%. (Scale bar = 5mm).
3.5. Conclusions

Cellulose triacetate films incorporating quantum dots can be easily prepared. The surface conversion of the films to cellulose renders the otherwise hydrophobic films hydrophilic and facilitates their usage in aqueous processes such as papermaking. The films were incorporated into paper sheets through hydrogen bonds between cellulose surfaces. The resultant paper appeared highly fluorescent under UV illumination in the regions where the films were present.

3.6. Acknowledgements

We thank NSERC Canada and Paprican for financial support and the Centre for Self Assembled Chemical Structures (CSACS) for use of laboratory equipment. T. A. thanks Dr. N. Ulkem and J.M. Berry for helpful discussion.
3.7. References

Fluorescent CTA films in paper
Chapter 4
Electrospun cellulose triacetate fibers containing CdSe/ZnS quantum dots

Similar to the fluorescent films described in the previous chapters which indicated a degree of compatibility between CTA and QDs, in this chapter we prepare fluorescent CTA fibers using the electrospinning technique. The work has been published and is reproduced with permission from:

Electrospun fluorescent CTA fibers
4.1. Abstract

Fluorescent cellulose triacetate (CTA) fibers containing CdSe/ZnS quantum dots (QDs) were prepared by electrospinning solutions of CTA dissolved in an 8:2 v/v co-solvent system of methylene chloride (MC) and methanol (MeOH). The relatively low loading of colloidal nanoparticles was sufficient to impart fluorescence to the fibers but did not significantly alter fiber morphologies, which tended toward smooth surfaces with the occasional longitudinal feature. The fibers were birefringent due to the alignment of the polymer chains which occurred during electrospinning and had widths on the order of a hundred nanometers.

4.2. Introduction

Electrospinning is an established and experimentally straightforward technique which has the potential to produce virtually continuous lengths of sub-micron width fibers, from a wide range of materials, including many polymers and blends.\textsuperscript{1-5} Electrospun fibers have been proposed for many different types of applications including filtration, textiles\textsuperscript{6}, tissue engineering\textsuperscript{7-10}, optical and electronic devices\textsuperscript{11, 12}, and sensing\textsuperscript{13, 14}. The properties of electrospun polymer fibers can be further enhanced by the incorporation of functional materials such as polymer (i.e. to make a blend)\textsuperscript{15, 16}, metal complexes\textsuperscript{17-19}, nanoparticles\textsuperscript{12,20-23}, carbon nanotubes \textsuperscript{17, 18}, dye molecules\textsuperscript{12}, dye loaded zeolite crystals\textsuperscript{19}, or proteins\textsuperscript{14, 20}.

In a typical experiment, a high voltage (kV) is applied to a metallic capillary through which the polymeric solution is fed. The charges induced on the surface of the pendant polymer droplet act in opposition to the surface tension of the fluid. Above a critical voltage, repulsive Coulombic interactions overcome surface tension, causing an electrified polymer jet to be accelerated from the apex of the droplet towards a grounded collector, located some fixed distance away. The polymer jet is elongated into a long, thin filament as the solvent evaporates and is deposited onto the collector in the form of fibers. Depending upon the nature of collection, fibers may be collected in random mats or in more ordered assemblies. The elongational flow of the polymer to some degree results in orientation of the macromolecular chains in the fibers and to interesting
Electrospun fluorescent CTA fibers

uniaxial properties such as birefringence.\textsuperscript{2,4,28} The morphologies and dimensions of electrospun fibers are dependent upon the often complex interrelationship between intrinsic polymeric and solution properties, processing parameters and ambient conditions.\textsuperscript{4}

In this paper, we describe the electrospinning of CTA fibers containing CdSe/ZnS quantum dots from a mixed solvent composed of methylene chloride and methanol. The fluorescence of the fibers was derived from the colloidal QDs which were incorporated into the electrospinning solutions. The quantum confinement of excitons in semiconductor nanoparticles results in unique electronic properties.\textsuperscript{29-31} Briefly, the energetic spacing between the valence and conduction bands increases with decreasing particle size giving rise to size dependent properties which include broad excitation and sharp fluorescence. For fluorescent applications this translates into smaller nanoparticles emitting bluer wavelengths compared to larger nanoparticles and a potential for creating single systems which incorporate QDs having discrete fluorescent wavelengths. The QDs used in this study are core-shell nanoparticles with a CdSe core and a ZnS shell. The CdSe core has a lower band-gap compared to the ZnS outer layer(s) and largely defines the optical characteristics of the particles, whereas the ZnS shell improves the optical efficiency and stability of the particles.\textsuperscript{21} In comparison to organic fluorophores, surface passivated QDs possess comparable quantum yields but are less susceptible to photobleaching.\textsuperscript{22}

CTA is a commercially important cellulose ester and is a key component of photographic films and liquid crystal display (LCD) screens. CTA is a β-(1-4) glycosidic polymer derived from the acetylation of cellulose, with acetyl groups in place of the hydroxyl groups of cellulose.\textsuperscript{23} Unlike native cellulose which is highly crystalline, hydrophilic and insoluble in most solvents, cellulose triacetate is semi-crystalline, hydrophobic and readily soluble in common organic solvents, making it more amenable to solution processing. Cellulose can be regenerated by the base-catalyzed de-esterification of CTA. The conversion to cellulose can be limited to the surfaces of CTA fibers and films, improving aqueous dispersibility while retaining the bulk properties of the polymeric material. In previous work\textsuperscript{24, 25}, we have shown that CTA is an appropriate polymeric matrix for CdSe/ZnS QDs and that surface hydrolysis to cellulose made the
fluorescent materials water dispersible and able to adhere to other cellulose surfaces. The electrospinning conditions described in the current paper were similar to those employed by Han et al.\textsuperscript{26} who made CTA fibers from mixtures of ethanol and methylene chloride, and established the practicability of preparing CTA fibers using the electrospinning technique. The desirable properties of CTA, including good spinnability and compatibility with QDs, made it a highly suitable polymer for the current application. We found it relatively straightforward to prepare fluorescent CTA fibers with reasonably reproducible morphologies and dimensions from solutions of CTA in MeOH and methylene chloride mixed solvent.

A parallel electrode collector was employed which resulted in the spatial alignment of fibers across the gap between electrodes.\textsuperscript{27, 28} The fluorescent fibers were characterized by a high density arrangement of QDs, and the method of collection provided some degree of control over the 2-D fiber architecture. The ability to produce polymeric fibers incorporating QDs and ordered arrangements of these fibers may be important for photonic and electronic devices such as polymeric lasers and light-emitting diodes.\textsuperscript{29, 30} Electrospinning seems to provide a potentially straightforward route toward the preparation of such systems.\textsuperscript{11,20,21}

4.3. Experimental

4.3.1 Materials

Cellulose triacetate (43% acetyl content by weight, 103 kDa), and HPLC grade methanol (MeOH) and methylene chloride were purchased from Sigma-Aldrich. Commercial suspensions of tri-octylphosphine oxide (TOPO) capped CdSe/ZnS quantum dots in toluene with nominal sizes of 2.1-5.2 nm and approximate quantum yields of 30-50\% were obtained from Evident Technologies. All materials were used as received.

4.3.2 Preparation of electrospinning solutions

CTA solutions in methylene chloride and alcohol mixed solvent (65 g/L) were prepared in the following ratios: 10\% and 20\% v/v solutions of methanol in methylene chloride. The spinning solutions incorporating QDs were prepared from CTA dissolved
in 20% by volume methanol in methylene chloride. The fluorescent solutions had a first excitation peak absorbance of approximately 0.01 units, corresponding to an approximate QD loading of less than 1% by weight.

### 4.3.3 Electrospinning

A horizontal set-up was used. The voltage was fixed at 15 kV, the solution flow rate at 1 mL/hr and the distance between capillary and collector at 10 cm. Depending upon the subsequent characterization, the grounded collector consisted of either a piece of metal foil or parallel metallic electrodes held in place by a plastic clamp which allowed the distance between electrodes to be adjusted. The parallel electrode collection allowed the fibers to be directly transferred onto a substrate for subsequent analysis. The experiments were repeated at least twice with fresh solutions at ambient temperature (ca. 20 °C) and relative humidity (ca. 30-50%).

### 4.3.4 Scanning electron microscopy (SEM)

Morphologies and fiber dimensions were determined from SEM images obtained using a Hitachi S-4700 cold field emission scanning electron microscope (FE-SEM). Prior to imaging the samples were sputter coated with Au-Pd using a Technics Hummer IV Sputter/Coater System.

### 4.3.5 Polarized optical microscopy

A Nikon Eclipse LV100POL microscope was used to observe the fibers under polarized light.

### 4.3.6 Differential scanning calorimetry (DSC)

The thermal properties of the fibers were studied using a TA Instruments Q2000 DSC with a heating rate of 10 °C/minute and sample masses of 5-10 mg.
4.3.7 Fluorescence and UV-VIS spectroscopy

Fluorescence spectra were recorded on a FluoroMax-2 fluorimeter (Jobin Yvon-Spex) using 400 nm excitation. UV-VIS spectra were obtained using a Cary 300 BIO UV-Vis spectrometer (Varian). Fiber spectra were obtained by transferring fiber mats onto quartz slides.

4.3.8 Confocal microscopy

Fluorescent images of the fibers containing QDs were obtained using a Zeiss 510 confocal microscope. The optical configuration of the microscope was optimized for the different QD emission wavelengths. Fibers incorporating 525 nm QDs (green) were visualized using a 505-550 nm band pass filter and fibers incorporating 615 nm (red) QDs were visualized using a 560 nm long pass filter. A 405 nm laser excitation was employed.

4.4. Results and Discussion

4.4.1 Effect of alcohol content upon morphologies of CTA fibers

Prior to preparing fluorescent fibers, the properties CTA fibers which did not contain QDs were studied in order to better assess the affects, if any, resulting from the incorporation of nanoparticles into the polymer fiber. CTA fibers were prepared from mixed solvent comprised by volume of either (1) 20% MeOH and 80% MC or (2) 10% MeOH and 90% MC. As mentioned in the introduction section, CTA fibers have been successfully electrospun by Han et al.\textsuperscript{26} who dissolved the cellulosic in mixtures of ethanol and MC. We employed MeOH as the alcohol component of our solvent since our past work\textsuperscript{24} indicated some compatibility between QDs and this solvent mixture, although this does not exclude possible compatibility with EtOH and MC mixtures.

In general, the SEM observations presented in Figure 4.1 indicated that an increase in the volume percentage of alcohol altered the fiber morphologies from porous to non-porous. As the volatility of the solvent system decreased with added alcohol, the topology of the fibers tended to smoother surfaces. The use of volatile solvents, such as MC, has been shown to produce porous fibers.\textsuperscript{26, 31, 32} Replacing methylene chloride
Electrospun fluorescent CTA fibers

(boiling point = 40 °C) with lower vapor pressure alcohols will reduce the tendency to pore formation but at the 10% by volume methanol content (Figure 4.1a) porous morphologies were still apparent. The fibers prepared from 10% by volume MeOH solvent had widths ranging from ~300 nm to ~3 μm, whereas the widths of the fibers electrospun from the higher MeOH content solvent had a narrower range of ~400 nm to ~700 nm. The relatively large thicknesses are not uncommon for fibers electrospun from cellulose derivatives 29, 36-39 and may be related to the relative stiffness of cellulosic chains. Dimensions were not entirely consistent across single fibers.

The porous fibers were characterized by longitudinal pores having approximately 100 nm long-axis widths, with some of the larger pores seemingly formed from coalescence of smaller ones, and by smaller pores with cross-sections approaching circular. Pore formation in electrospinning is generally attributed to phase separation induced by rapid solvent evaporation.31 For polymer solutions exhibiting an upper critical solution temperature (UCST), such as CTA, the sudden cooling caused by rapid solvent evaporation may be sufficient to quench the system into the biphasic regions of the phase diagram.33, 34 The solvent rich phase evaporates to form pores and the concentrated phase solidifies into the fiber. The dominant mechanism for the formation of pore structures in electrospun fibers, particularly when interconnected pores are observed, is generally considered to be phase separation by spinodal decomposition.31 The rapid rates of fiber formation and solvent evaporation, characteristic of electrospinning from volatile solvent, are compatible with the fast quench needed to bring the system into the unstable spinodal region of the phase diagram.18 Porous structures have also been attributed to vapor induced phase separation34, 35, the effects of humidity32, 34, 35 and solvent evaporation through a polymer skin32. In the current system, where highly volatile methylene chloride was employed under relatively high ambient humidity and where polymer skins were formed, it is difficult to conclusively pinpoint a single pore forming process.

The fibers spun from the higher alcohol solvent compositions (Figure 4.1b) were generally smooth with occasional longitudinal ridges. This type of feature has been previously observed by Han et al.26 for electrospinning CTA in ethanol and methylene chloride co-solvents and by Park et al.36 who prepared ethyl cellulose fibers by electrospinning from mixtures of DMAc and THF. The corrugations, particularly when
they extended in a linear fashion along the fiber lengths or disengaged from the main fiber axis, seem to indicate fiber bundles, which occur when fibers are still wet as they hit the collector. The addition of alcohol, from 10% to 20% by volume of MeOH, decreased the overall volatility of the system and, with the electrospinning conditions employed in the current experiments (i.e. 10 cm between capillary and collector), resulted in wetter fibers which were more likely to stick to each other. In addition, the fibers were sometimes ribbon-like, a morphology which results from the collapse of a polymer skin. Polymer skin collapse patterns have been previously described by Koobhongse et al., include ribbons, branches and wrinkles, and may be the source of the occasional textured morphologies which were observed in the fibers prepared from the higher percentage of alcohol.

Figure 4.1: SEM of CTA fibers electrospun from MC and MeOH mixed solvent: (A) 10% MeOH by volume and (B) 20% MeOH by volume.
4.4.2 Birefringence of electrospun CTA fibers

Polarized microscope images of the CTA fibers are presented in Figure 4.2. Due to the nature of collection, the fibers were more or less aligned with the direction of alignment indicated with arrows. Samples which are oriented or crystalline appear bright when viewed between crossed polarizers, and conversely, amorphous materials appear dark. All the electrospinning conditions resulted in fibers which appeared bright between crossed polarizers, indicating birefringence and polymer chain orientation within the fibers. Birefringent wet spun CTA fibers were prepared by Bheda et al.\textsuperscript{39}, but in that case the initial polymer solution was an ordered liquid crystalline phase. Han et al.\textsuperscript{26} did not address the birefringence of the CTA fibers which they prepared. The observation of birefringence does not seem altogether surprising for fibers prepared from semi-crystalline polymer in a mesogenic solvent, subjected to the strong elongational forces characteristic to electrospinning.
Figure 4.2: Polarized optical micrographs of CTA fibers electrospun from MC and MeOH mixed solvent: (A) 10% MeOH by volume and (B) 20% MeOH by volume. Arrows indicate direction of fiber alignment.

4.4.3 Properties of fluorescent electrospinning solutions

QDs were incorporated into the fibers electrospun from the higher alcohol content solvent mixture because the fibers prepared from this condition were most uniform. QDs have been previously incorporated into polymeric electrospun fibers: PLLA and PS fibers embedded with ZnSe quantum dots were prepared by Schlect et al.\textsuperscript{11}, PMMA fibers embedded with CdSe/ZnS quantum dots were electrospun by Tomczak et al.\textsuperscript{12}, Liu et al.\textsuperscript{20} spun fibers from mixtures of CdSe/ZnS QDs and light guiding polymer, Wang et al.\textsuperscript{40} prepared fibers from cadmium acetate and polyethylene oxide (PEO) solution and treated the resultant fibers with H\textsubscript{2}S to generate CdS nanoparticles in-situ and most
recently, Li et al.\textsuperscript{41} made nanotubes by electrospinning mixtures of colloidal ZnO and polyvinylpyrrolidone (PVP) solution. In the current approach, small volumes (~0.1 mL) of CdSe/ZnS QDs in toluene (either single particle sizes or a mixture) were added to the CTA solution and fibers were obtained by electrospinning the mixture. The fluorescence and absorbance spectra of the QDs and a photograph of the spinning solutions taken under UV illumination are presented in Figures 4.3 and 4.4, respectively.

Figure 4.3: Absorbance (dotted lines) and fluorescence (solid lines) of QDs used in electrospinning experiments. (From left to right, an increasing QD size series in toluene).

Figure 4.4: Fluorescent electrospinning solutions containing QDs with fluorescence peaks at (A) 525 nm QDs, (B) 550 nm, (C) 590 nm, (D) 615 nm and (E) 525 and 615 nm, photographed in UV-light.
4.4.4 Morphology of CTA-QD fibers

In Figure 4.5, SEM images are presented of two representative fluorescent samples. The morphologies and dimensions of the fibers were found to be generally unaffected by the relatively small addition of QDs, and there did not seem to be any measurable correlation between the size of the QDs and the size of the fibers. The widths of the fibers ranged from 100 nm-2 μm.

Figure 4.5: SEM of CTA fibers containing quantum dots with fluorescence peaks at (A) 550 nm and (B) 590 nm.

4.4.5 Fluorescence of CTA-QD fibers

The fluorescence of the fibers was visualized using UV-illumination (Figure 4.6), fluorescence spectroscopy (Figure 4.7) and confocal microscopy (Figure 4.8). Figure 4.6
Electrospun fluorescent CTA fibers

illustrates the macroscopic fluorescence of a mat of unaligned fluorescent fibers collected on a sheet of foil and Figure 4.7 looks at that same mat using fluorescence spectroscopy. The peak wavelength was found to be 611 nm, slightly blue-shifted compared to the fluorescence of the initial colloidal QDs. The shift to higher energy emissions has been previously observed in CTA films embedded with commercial CdSe/ZnS QDs. Figure 4.8 shows confocal microscopy of the fibers which allowed direct visualization of the fluorescence of single fibers. The fibers in 4.8a contained QDs with 525 nm peak fluorescence and in 4.8b, two types of fibers, containing QDs which fluoresced at either 525 nm or 615 nm, were collected onto a single substrate.

Figure 4.6: Macroscopic fluorescence of a mat of CTA fibers containing QDs with fluorescence at 615 nm.

Figure 4.7: Fluorescence of CTA fiber mat containing QDs.
Figure 4.8: Confocal microscopy of fluorescent fibers: (A) fibers containing QDs with 525 nm fluorescence peaks and (B) fibers containing two QD sizes with either 525 and 615 nm fluorescence peaks. (Scale bars = 20 mm).
4.4.6 Birefringence of CTA-QD fibers

Polarized microscopy images of CTA fibers which contain QDs are shown in Figure 4.9. The fibers are birefringent and the addition of QDs does not seem to significantly alter the ordering of polymer chains which occurs during the electrospinning process.

![Polarized optical micrographs of CTA fibers containing quantum dots with fluorescence peaks at (A) 525 nm and (B) 615 nm.](image)

Figure 4.9: Polarized optical micrographs of CTA fibers containing quantum dots with fluorescence peaks at (A) 525 nm and (B) 615 nm.

4.4.7 Thermal properties of electrospun fibers

Differential scanning calorimetry (DSC) was used to probe the thermal signature of the electrospun fibers, and to determine if crystalline order played a role in the observed birefringence. In Figure 4.10, the DSC curves for first heating are presented for commercial CTA melt-processed pellets, for CTA fibers electrospun from 20% by
volume alcohol content and for a CTA film cast from the same solvent mixture. The thermograms for fibers containing QDs are not shown since they were very similar to those of the blank CTA fibers. The endothermic hump observed between 50 °C and 100 °C in the CTA fiber and film thermograms is attributed to the evaporation of solvent.\textsuperscript{42} The thermal properties of CTA have been studied extensively, and the absence of a crystallization temperature (T$_c$) and of a well-defined glass transition (T$_g$) in the commercial sample is typical.\textsuperscript{42} In contrast, the film and fiber samples exhibited glass transition regions at approximately 190 °C, and exothermic peaks at around 218 °C, indicative of some crystallization upon heating. All samples finally melted at around 290 °C. The transition of amorphous regions to crystalline domains, observed upon heating the film and fiber samples, is related to the processing of the samples. For the CTA film, the slow timescale of evaporation (ca. 24 hours) may have allowed the chains to achieve some degree of orientation before being frozen into the film structure. The electrospun fibers were subjected to extremely rapid solvent evaporation, but also to strong stretching forces that may be sufficient to produce significant polymer chain alignment. The chain alignment induced during processing likely facilitates the crystalline transition observed upon heating.

The DSC data can be used to determine the degree of crystallinity by evaluating the heats of fusion of the samples (\(\Delta H_M\)) and comparing those values to the heat of fusion for a 100% crystalline CTA sample (\(\Delta H^\circ_M\)). In these calculations the heat of fusion of 100% crystalline CTA was taken to be 58.8 J/g.\textsuperscript{43} The data is summarized in Table 4.1. The percent crystallinity of all three samples was quite similar (26-28%) but was greatest for the fiber sample and least for the commercial pellets.
Electrospun fluorescent CTA fibers

Figure 4.10: DSC thermograms of commercial CTA pellets (a), CTA film and electrospun fibers from 8:2 v/v MC:MeOH solvent, (b) and (c), respectively.

Table 4.1: DSC data for CTA film, fiber and commercial pellets.

<table>
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<th>$T_m$ (°C)</th>
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<td>Fiber</td>
<td>217.7</td>
<td>288.6</td>
<td>3.2</td>
<td>19.8</td>
<td>28.2</td>
</tr>
</tbody>
</table>

$^1$ % C = (H$M$ - $\Delta H_C/\Delta H_M^*$) × 100%

4.5. Conclusions

CTA fibers of micron and submicron widths were successfully electrospun from CTA dissolved in mixtures of methanol and methylene chloride. The fibers were macroscopically aligned by the use of a parallel electrode collector. By varying the alcohol component of the solvent it was possible to obtain fibers with either porous
morphologies (lower alcohol content) or smoother morphologies (higher alcohol content). Fluorescent fibers were prepared by incorporating QDs into the CTA spinning solution and the fibers possessed morphologies similar to fibers spun from the initial polymer solution. The fibers were all found to be birefringent due to the alignment of the polymer chains which occurs during the electrospinning process.

4.6. Acknowledgements

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4.7. References

Electrospun fluorescent CTA fibers


Electrospun fluorescent CTA fibers
Appendix A

Solvent effects in electrospinning experiments

A.1. Comparison of ethanol and methanol as alcoholic solvent components

CTA fibers were prepared from MC solvent with the following alcohol contents by volume: (1) 20 % ethanol, (2) 10 % ethanol, (3) 20 % methanol and (4) 10 % methanol. In general, the SEM observations presented in Figures A.1 and A.2 indicated that an increase in the volume percentage of alcohol altered the fiber morphologies from porous to non-porous. As the volatility of the solvent system decreased with added alcohol, the topology of the fibers tended towards smoother surfaces. The use of volatile solvents, such as MC, has been shown to produce porous fibers.1-3 Replacing methylene chloride (boiling point = 40 °C) with lower vapor pressure alcohols will reduce the tendency to pore formation. By similar reasoning, as compared to methanol, higher boiling ethanol is expected to hinder pore formation to a greater extent. Indeed, the fibers spun from 10% v/v methanol presented more regular and pronounced pore structures (Figures A.2a & A.2b) compared to those prepared using the same volume of ethanol (Figures A.1a & A.1b).

Figure A.1: SEM of CTA fibers in ethanol and methylene chloride mixed solvent: (A) and (B) 9:1 v/v MC:EtOH, (C) and (D) 8:2 v/v MC:EtOH.
Solvent effects in electrospinning

The general trend in the surface morphologies of the fibers was similar to those observed by Han et al.\textsuperscript{1} for CTA fibers spun from mixtures of methylene chloride and ethanol. However, instead of the interconnected pores reported for the fibers spun from 10\% v/v ethanol, we observed both porous and non-porous morphologies on single fibers (Figures A.1a & A.1b). The fibers spun from 10\% v/v methanol were generally characterized by longitudinal pores having approximately 100 nm long-axis diameters, with some of the larger pores perhaps formed by the coalescence of smaller ones (Figures A.2a & A.2b). Very few of the 10\% v/v methanol fibers showed the porous/non-porous mixed morphology (Figure A.2a). The fibers spun from the higher alcohol solvent compositions were smoothest but sometimes presented corrugated and wrinkled textures.

The observation of both porous and smooth morphologies on single fibers spun from the lower percentage alcohol solvent, especially when ethanol was used (Figures A.1a & A.1b), is perplexing. These fibers seemed to posses all possible morphologies: pores, necking, twists, long ridges, shorter ridges perhaps formed from the merging and collapse of adjacent pores, cup shapes associated with skin collapse and smooth regions. The observation of mixed morphologies seemed to indicate local compositional inhomogeneities but since ethanol and methylene chloride are miscible and the solutions...
were vigorously mixed prior to each experiment it is unclear why this would be the case. More likely, fluctuations in ambient conditions along the spin line were responsible for the mixed morphologies. Also, post experiment the fibers were swiped onto substrates and it is possible that the flat regions in the porous fibers were caused by fiber regions drying against the substrate surface.

A.2. References

Solvent effects in electrospinning
Chapter 5
Fluorescent-labeling of CNCs with QDs

A different approach to fluorescent cellulosic nanocomposite materials involving the covalent linkage of cellulose nanocrystals to quantum dots is presented in this chapter. In this study, the reducing ends of cellulose nanocrystals are modified to carboxylic acid groups and using a standard biochemical reaction are labeled with the fluorescent nanoparticles. This work was extremely challenging, largely due to the similar size regimes of the two constituent particles.
Fluorescent labeling of CNCs
5.1. Abstract  

Cellulose nanocrystals (CNCs) were fluorescently labeled at one end with quantum dots (QDs). The asymmetric, covalent linkage of QDs to CNCs was achieved through reaction of the reducing ends of the CNCs with the surface ligands associated with the QDs. The resultant assemblies were analyzed using fluorescence spectroscopy, atomic force microscopy (AFM) and transmission electron microscopy (TEM). The TEM images provided direct visual evidence of positive coupling reactions. Fluorescently labeled CNCs may potentially be used in toxicity studies to assist in the localization of CNCs in tissue samples, or in the preparation of novel fluorescent cellulosics.

5.2. Introduction

Cellulose is a ubiquitous biopolymer which consists of D-glucose units, linked in a \( \beta-1, 4 \) conformation. The native sources of cellulose may be algal, bacterial or plant-based. Depending upon source and chemical history, several crystalline modifications of cellulose are possible: I, II, III\(_{l}\), IV\(_{I}\) and IV\(_{II}\).\(^1\) Native celluloses exist in either I\(_{\alpha}\) (triclinic unit cell) or I\(_{\beta}\) (monoclinic unit cell) crystalline phases\(^2\), the ratio of which is dependent upon the source, with cellulose from cotton predominantly in the one-chain triclinic I\(_{\beta}\) phase.\(^3\) The cellulose chain is characterized by distinct end groups, and has a reactive hemiacetal at the C1 reducing end position and a less reactive acetal at the C4 non-reducing end position. The nature of the molecule is therefore inherently suited to asymmetric end group modification.

The polarity of cellulose chains (i.e. end group directionality) within different crystalline polymorphs has, in the past, been an issue of some contention which has been addressed experimentally both by X-ray and neutron diffraction studies, and by the chemical labeling of reducing end groups. Labeling cellulose crystalline domains with nanometer scale colloidal particles, such as gold or silver, coupled with electron microscopy helped to provide further insight into the molecular directionality of cellulose chains. Chain packing is designated as parallel when reducing ends are present only on one side of a crystalline domain and anti-parallel when reducing ends alternate on either side. Hieta et al.\(^4\) were the first to address the polarity of cellulose chains in crystalline...
Fluorescent labeling of CNCs

native celluloses by a reducing end labeling approach. Using chemistry which targeted reducing end groups only, the silver staining of Valonia microfibrils occurred exclusively at one end of the microcrystallites. Similarly, the reducing ends of bacterial cellulose were stained with silver by Kuga and Brown⁵, who confirmed that cellulose chains from higher native sources are oriented in parallel. Maurer and Fengel⁶ used the silver labeling method of Kuga and Brown⁵ to demonstrate the parallel packing in cellulose I derived from plant sources (cotton linters). More recently, Kim et al.⁷ studied the molecular directionality of different cellulose polymorphs, and the solid state transition from one crystalline modification to another, by the gold nanoparticle labeling of reducing ends.

Cellulose nanocrystals (CNCs) are produced by the acid hydrolysis of the amorphous or accessible regions of native celluloses, resulting in highly crystalline rod-shaped particles, with typical dimensions of 5-10×150-300 nm. The reducing end labeling of CNCs has not been previously studied, perhaps since it is wholly accepted that the chains of cellulose I crystallites are packed in parallel orientation. However, the non-specific labeling of cellulose nanocrystals with FITC fluorescent dye for bio-imaging applications has recently been explored in the literature.⁸ Here we describe the reducing end labeling of CNCs with quantum dots. Quantum dots are semiconductor nanoparticles with typical diameters of ca. 2-10 nm and size-dependent optical properties, such as sharp, discrete fluorescence peaks and broad absorbance. The aim of this study was to functionalize CNCs with a fluorescent particle using a reducing end labeling approach. This approach is attractive because it is compatible with the reaction asymmetry inherent to cellulose I crystallites and also, since the chemistry only targets end groups, we hoped it would not alter the properties or stability of the CNCs significantly. In addition, QDs have improved photo-stability compared to organic dyes and excellent electron beam contrast which should provide visual evidence of covalent attachment. Fluorescently labeled CNCs may potentially be used to monitor the location and lifetime of CNCs in tissue samples, or perhaps to prepare fluorescent liquid crystalline materials.
5.3. Experimental

5.3.1 Materials

1-ethyl-3-(3-dimethylaminopropyl) carbodiimide (EDC), N-hydroxysulfosuccinimide (sulfo-NHS) and ion-exchange resin were purchased from Sigma-Aldrich. Aqueous, colloidal suspensions of amine functionalized CdSe/ZnS QDs were purchased from Evident Technologies, Inc. and eBioscience, Inc. A variety of different emission wavelengths are available, but in the complete experiment described below, QDs from Evident Technologies, Inc. which fluoresced at 590 nm were employed.

5.3.2 Cellulose nanocrystal preparation

Aqueous suspensions of CNCs were prepared by sulfuric acid hydrolysis of cotton. Briefly, Whatman ashless filter aid (40 g) was reacted with sulfuric acid (64% by weight, 700 mL) at 45 °C for 45 minutes, at which time the reaction was quenched by 10-fold dilution. The CNCs were concentrated and washed using repeated centrifugation cycles (6,000 rpm, 10 minutes) followed by extensive dialysis against distilled water. Next, the suspension was sonicated, treated with mixed bed ion-exchange resin and filtered using glass microfiber filters. The concentration of the final suspension was 2.0 ± 0.1 % by weight and the dimensions of the nanocrystals from AFM were approximately 5 nm × 200 nm. Finally, the suspension was neutralized by the addition of NaOH, the amount of which was determined from conductometric titration (%S = 0.23±0.02).

5.3.3 Reducing end carboxylation of cellulose nanocrystals

The reducing ends of the cellulose chains which comprise the CNCs were carboxylated using a method similar to that described by Hieta et al.4 Acetic acid (2.36 mL, 99.7%), CNC suspension (2% by weight, 50 mL), deionized water (10 mL) and sodium chlorite (1.1645 g) were combined. After approximately 48 hours with stirring at room temperature, the yellowed reaction mixture was quenched by the addition of deionized water (50 mL). The modified CNCs were dialyzed against distilled water until the pH inside the dialysis bags approached the pH of the water, sonicated and filtered through a glass microfiber filter. The bleached CNCs were treated with mixed bed ion-
Fluorescent labeling of CNCs

exchange resin, in order to complex stray ions. The CNCs were filtered prior to titration and reaction.

5.3.4 Tagging reaction

In a typical experiment, reducing end carboxylated CNCs (~0.6 g, 0.6 weight %) were combined with amine functionalized QDs (100 µL, 12 nmol/mL). EDC and sulfo-NHS (65 µL of 10 mM stock solutions) were added, the pH was adjusted to ~7.5 with the addition of 10 mM N NaOH. The volume of the reaction was approximately 1 mL. The reaction was left stirring, overnight at room temperature. The mixture was then separated into four fractions by a series of 15 minute centrifugation cycles: (1) 3,000 rpm pellet, (2) 4,000 rpm pellet, (3) 5,000 rpm pellet and (4) supernatant remaining after centrifugation cycles. The loose pellets were re-dispersed in 0.5 mL deionized water by vigorous mixing.

5.3.5 Fluorescence

The fluorescence spectrum of each fraction was measured with a microplate reader (Synergy 4 Hybrid Multi-Mode Microplate Reader) using a 400 nm excitation wavelength.

5.3.6 Imaging

Prior to imaging, the samples were placed in a gentle sonication bath for approximately 30 minutes in order to promote dispersal. Atomic force microscopy (AFM) tapping-mode images were obtained using an MFP-3D™ from Asylum Research. Samples for AFM were prepared by drying down drops of diluted sample onto glass or mica substrates. For TEM, diluted samples were dried onto carbon coated grids and imaged using a Philips CM200 TEM, operated at 80-200 kV, with a point-to-point resolution of 0.24 nm and line resolution of 0.17 nm. The samples were unstained.
5.4. Results and discussion

5.4.1 Oxidation of cellulose nanocrystals

Aqueous, electrostatically stabilized suspensions of CNCs were prepared by the sulfuric acid hydrolysis of cotton. The CNCs possess a negative surface charge due to the presence of anionic sulfate ester groups introduced during hydrolysis. The hemiacetal reducing ends of the CNCs were oxidized to carboxylic acid groups using sodium hypochlorite in a quantitative reaction. The modified CNCs were also characterized by \( \xi \)-potential measurements which indicated similar values to the starting material (-35 to -45 mV).

5.4.2 Fluorescent labeling of cellulose nanocrystals

Following carboxylation, a 1-pot labeling reaction which involved the formation of an amide linkage between the carboxylated reducing ends of the CNCs and the amine groups of the QD surfactants was performed. The linkage reaction is a standard biochemical protocol and uses EDC and sulfo-NHS as the coupling agents. Similar chemistry has been employed previously both in the functionalization of QDs and in the modification of cellulose microcrystals, most notably by Araki et al., who attempted the steric stabilization of cellulose microcrystals using polyethylene glycol (PEG). Significantly, concentrations of EDC greater than ~10 mM were found to quench QD emission and therefore, reaction conditions had to be tailored appropriately. Characterization of the reaction between colloidal-sized particles is non-trivial and in this case we relied heavily on electron and atomic force imaging. It is difficult, for example, to detect the amide bond, which represents a very small fraction of total surface and bulk bonds, and to separate unreacted starting materials because of the similar size regimes of the particles.

5.4.3 TEM analysis of blank solutions

Figure 5.1 presents transmission electron microscopy (TEM) images prepared from the following four suspensions: (1) original CNC material, (2) reducing end carboxylated CNC, (3) QDs alone and (4) QDs combined with reducing end modified CNCs in the absence of linker chemicals. The TEM results indicated that the original
CNC and oxidized CNC were similar (5.1a and 5.1b, respectively) and that the mixture of QDs and oxidized CNC did not result in any specific localization of the QDs (5.2d).

Figure 5.1: TEM of (A) unmodified CNCs, (B) oxidized CNCs, (C) QDs and (D) a mixture of unreacted QDs and oxidized CNCs.

5.4.4 TEM and AFM analysis of reducing end tagged CNCs

Centrifugation was employed in order to achieve some degree of segregation between unreacted particles (CNCs and QDs) and reacted products. We had previously observed that at low centrifugation speeds (i.e. below 6,000 rpm) CNCs would sediment preferentially. It was therefore hoped that centrifugation at relatively low speeds could be
used to separate, albeit crudely, the different species present in the reaction mixture. We attempted other separation and purification techniques (i.e. gel electrophoresis, Sephadex™ columns, and dialysis) with little success, for example, the use of a Sephadex™ column resulted in the retention of all the cellulosic material, with only unreacted QDs eluted.

In Figure 5.2, AFM images are presented of the four fractions isolated by centrifugation and described in the experimental section. Here, the resolution was not sufficient to pinpoint any linkage between QDs and CNC but the images were useful in identifying gross differences between fractions. In general, rod-like particles were visible in each fraction. However, the supernatant predominately consisted of aggregates characteristic of high QD concentrations (5.2d).

Figure 5.2: AFM images of the redispersed ‘loose’ pellets collected by centrifugation and of the final supernatant: (A) pellet after centrifugation at 3,000 rpm, (B) pellet after centrifugation at 4,000 rpm, (C) pellet after centrifugation at 5,000 rpm and (D) supernatant.
TEM images of the four fractions are presented in Figures 5.3, 5.4, 5.5 and 5.6. Figure 5.3 showed the presence of large aggregates, as well as QD-labeled CNCs. Figure 5.3b shows a CNC with a large quantity of QDs emanating from one end of the particle: images of this kind were fairly common and may be a result of successful reaction coupled with the packing of unreacted QDs about the reacted QD as the sample is dried. The aggregates in the sample isolated at 4,000 rpm (Figure 5.4) were smaller sized, and both asymmetrically labeled CNCs and some free particles were apparent. The fraction isolated at 5,000 rpm (Figure 5.5) seemed to have the greatest proportion of the target product, but unreacted particles were visible in many images. The final supernatant (Figure 5.6) consisted mainly of free QDs, but asymmetrically reacted CNCs were also seen.

Figure 5.3: TEM images of fraction isolated at 3,000 rpm: (A) large µm-scale aggregate, (B) CNC with multiple QDs clustered around one end and (C) CNC with QDs at one end. Scale bars from left to right = 500 nm, 100 nm and 100 nm.
Chapter 5

Figure 5.4: TEM images of fraction isolated at 4,000 rpm. Evidence of a successful asymmetric reaction is present in each image, as are clusters of unreacted particles. Scale bars = 100 nm.

Figure 5.5: TEM images of fraction isolated at 5,000 rpm. The target, asymmetric product is present in each image. Scale bars = 100 nm.

Figure 5.6: TEM images of fraction isolated at 5,000 rpm. Unreacted QDs are present in each image. Scale bars = 100 nm.
Fluorescent labeling of CNCs

The TEM imaging places the method of separation into question since all fractions contain CNCs which are apparently asymmetrically labeled, and unreacted material seems omnipresent. Additionally, whether by action of drying or loss in colloidal stability due to reaction, significant particle aggregation is observed. Instead of separation of reacted product from starting materials, the TEM images suggest a segregation of species based upon aggregate size, with the largest aggregates observed in the loose pellet isolated at 3,000 rpm. With large aggregates, it was difficult to pinpoint the exact location of the linkage, if any, between QDs and CNCs due to the close proximity of crystallites and the possibility of the adsorption of free QDs onto the CNCs during sample drying. The sample fractionated at 5,000 rpm seemed to be the most promising and, in retrospect, perhaps we should have attempted some additional purification of this fraction, for example, filtration through a commercial 100 nm pore-sized membrane may have further isolated unreacted QDs.

5.4.5 General discussion of results from AFM and TEM

The overall picture from TEM, in general, is that of a reasonably successful reaction, with QDs localized at one end of the CNCs, and with some degree of separation of different sized aggregates achieved through successive centrifugation cycles at relatively low speeds (5,000 rpm top speed). The nature of TEM imaging, where the small area to be imaged is selected from a quick visual scan of the entire grid, favors the observation of larger particles or aggregates, and therefore may not be entirely reflective of the true sample composition. Although the resolution is not as good with the AFM imaging, the larger areas scanned may actually provide a more representative description of the different fractions. When considering the AFM and TEM results together, it seems reasonable to conclude that (1) in general, the asymmetric linkage was successful, (2) the CNCs and CNC-assemblies have diminished stability compared to the free QDs, (3) significant unreacted material was present, indicating less than optimal reaction conditions (i.e. in this case, approximately 1:1 QD:CNC ratio) and 4) the isolation of a fraction consisting of dispersed reacted material was perhaps an unrealistic expectation.
5.4.6 General discussion of reaction conditions

Here we would like to briefly address the issue of reaction conditions. The ratio of reactants was determined from estimates of the molecular weight of a nanocrystal (i.e. we obtained values on the order ~10^6 g/mol). In all reactions the EDC and sulfo-NHS reagents were always present in large excess. While we assumed, perhaps incorrectly, that steric constraints would limit the number of QDs linked to a given CNC, it seemed possible that several CNCs could be attached to a single QD, particularly due to the spacers provided by the amine-functionalized PEG surface ligands. Therefore, although each particle contained multiple reactive sites, we considered the CNCs to be sterically limited to approximately one site and each QD to possess several potential sites, related to the accessibility of the amine groups and the geometry of the particle. As such, the reaction condition with a 1:1 QD: CNC ratio, translates to an excess of amine sites compared to carboxylic acid sites. We also explored different reaction conditions, such as a large excess of CNCs compared QDs, and vice versa, with surprisingly little difference in results, i.e. the reaction (1:1) seemed to occur to some extent as long as there were plenty of unreacted particles of each type present.

5.4.7 Fluorescence of reducing end tagged CNCs

In Figure 5.7 the fluorescence spectrum of each fraction is presented. The fractions were all fluorescent and the emission position, ca. 590 nm, was unchanged compared to a blank solution of QDs in water. Since all the fractions contained free QDs, the observation of fluorescence was not very surprising. Without reading too much into the spectra, the supernatant seemed most fluorescent, a result which coincided well with the AFM and TEM images of this fraction, presented in Figures 5.2d and 5.6, respectively.
5.5. Conclusions

The reducing ends of CNCs were successfully labeled with QDs using a two-step process: (1) CNC reducing ends were converted to carboxylic acid groups using a chlorite treatment and (2) the carboxylic acid groups were reacted with amine functionalized QDs using carbodiimide chemistry. Four fractions were isolated by centrifugation, and in general, the size of any aggregates or assemblies decreased with increasing centrifugation speeds. The supernatant fraction was most stable and contained mostly unreacted QDs and some tagged product. In all fractions, evidence of successful labeling reaction (i.e. QDs localized on one end of the CNCs) was present, although sometimes the occurrence of larger assemblies complicated the analysis. The separation of unreacted particles from product was considered largely unsuccessful.
5.6. Acknowledgements

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5.7. References

Fluorescent labeling of CNCs
Chapter 6
Reinforcement with Cellulose Nanocrystals of Polyvinyl Alcohol Hydrogels Prepared by Cyclic Freezing and Thawing

In the previous chapter, CNCs were tagged with QDs for potential fluorescent marker applications. In this chapter, we explore the use of CNCs as reinforcing agents within a polymeric matrix. CNCs are incorporated into polyvinyl alcohol hydrogels and the properties of the CNC-loaded hydrogels are compared with those of a pure polyvinyl alcohol system.

PVA gels reinforced with CNCs
6.1. Abstract

Cellulose nanocrystals (CNCs) were incorporated into polyvinyl alcohol (PVA) hydrogels prepared by repeated freeze-thaw processing. The CNC-loaded hydrogels had improved structural stabilities and distinct microstructures, characterized by ordered domains of CNCs. The water sorption of the gels increased with CNC content due to the hydrophilic nature of the cellulosic and the decrease in PVA crystallinity. A reinforcement effect was observed in the CNC-loaded samples upon the application of uniaxial, confined compression, with the elastic moduli of the PVA-CNC samples increased relative to pure PVA hydrogels. Hydraulic permeability values were derived from the stress transients: at strains of ~15-20% and greater, the permeability of all samples approached a plateau value reflective of the hindered flow in soft gels which have been compressed, densified and dehydrated.

6.2. Introduction

The gelation of aqueous solutions of polyvinyl alcohol (PVA) may be achieved by irradiation\(^1\), \(^2\), chemical cross-linking\(^3\)-\(^5\) and cyclic freezing-thawing processes\(^3\), \(^4\). The freeze-thaw method has the advantages of being experimentally straightforward and not requiring the use of potentially undesirable chemical crosslinking agents. Gelation of PVA by the freeze-thaw mechanism is driven by the phase separation which occurs as the solution freezes and polymer is rejected from the growing ice crystallites, a process which is refined with repeated cycling.\(^3\), \(^4\) The size of the ice crystallites increases with cycling and the resultant gels are composed of water filled pores where the ice has melted surrounded by a polymer skeleton.\(^6\) Physical crosslinks in the form of hydrogen bonds and crystalline polymeric regions reinforce the gel structure.\(^3\), \(^4\) The properties of PVA hydrogels prepared by cyclic freezing and thawing have been studied extensively and depend upon a variety of factors, most important of which are molecular weight, concentration and number of cycles.\(^5\) Typically, an increase in any of these factors results in more crystalline gels which swell to a lesser extent in water and exhibit improved elastic strength. The properties of PVA hydrogels prepared by cyclic freezing and
PVA gels reinforced with CNCs

Thawing include biocompatibility, good stability at RT, water contents in the range of biological tissues (~90%) and potentially high elasticities.

The incorporation of functional materials into PVA hydrogels has been studied with the general aims of either reinforcement for artificial tissue applications or for drug delivery. Here we describe the reinforcement of PVA hydrogels with cellulose nanocrystals (CNCs). CNCs are needle-shaped, high aspect ratio particles produced from the mineral acid hydrolysis of native celluloses, such as cotton or wood. The amorphous regions of cellulose microfibrils are accessible to acidic degradation and controlled hydrolysis results in a relatively narrow size distribution (L ~100-300 nm and d ~5-10 nm) of highly crystalline residual particles. Unlike HCl hydrolyzed CNCs, sulfuric acid hydrolysis produces stable colloidal suspensions of CNCs in water. The stabilization is electrostatic in nature and arises from charged sulfate ester groups which are introduced onto the CNC surfaces during acidic treatment. CNCs are attractive for the reinforcement of nanocomposite materials because they are derived from an affordable, renewable, ubiquitous bio-resource and because of the high crystallinity, aspect ratio and apparent biocompatibility characteristic to the particles. The combination cellulose and PVA was first studied over 20 years ago by Nishio and Manley, who prepared polymer blends by combining solutions of cellulose and PVA dissolved in N,N-dimethylacetamide-lithium chloride. They reported a decrease in PVA crystallinity with increased cellulose content and attributed the result to an improvement in the miscibility of the blend components, perhaps due to mutual H-bonding interactions.

Microfibre, microfibrillated (MFC) and bacterial celluloses (BC) have been introduced to PVA films and hydrogels for many of the same reasons outlined for CNC reinforcement, with some important differences. Unmodified microfibre, microfibrillated and bacterial celluloses are uncharged and significantly longer than CNCs, with lengths in the micron scale. In addition, unlike CNCs and BC, microfibre cellulose and MFC contain substantial amorphous regions, which perhaps make these types of cellulose less than ideal for reinforcement applications. Although cellulose is hydrophilic and therefore inherently compatible with aqueous systems, uniform dispersal of long and uncharged strands is challenging. CNCs produced from sulfuric acid hydrolysis have the advantage of electrostatically-derived aqueous stability and relatively easy dispersal in aqueous...
media, including polymeric solution, without requiring high energy sonication. PVA nanocomposite materials have been previously studied: Kvien and Oksman\textsuperscript{15} aligned CNCs in PVA films using a strong magnetic field, Roohani et al.\textsuperscript{16} prepared films of CNCs dispersed in PVA copolymers, Peresin et al.\textsuperscript{17} electrospun fibers from mixtures of CNCs in PVA solution, and most recently, Wang et al.\textsuperscript{18} prepared PVA/starch sponges reinforced with cellulose nanowhiskers.

To the best of our knowledge, we are the first to look at the properties of CNCs dispersed in pure PVA hydrogels prepared by cyclic freezing and thawing. PVA and CNCs have complimentary chemical natures: PVA is water-soluble, but only at temperatures in excess of ca. 80 °C and CNCs are insoluble but hydrophilic. It was hoped that the incorporation of nm-scale, rigid colloidal particles into PVA hydrogels would lead to an enhancement of gel mechanical properties. However, the interactions of hydrophilic rods, hydrophilic, crystallizable polymer chains and water are likely to be complex. A few potential scenarios may be envisaged. (1) PVA and CNCs show little mutual attraction, and entropically-driven phase separation is possible. (2) CNCs and PVA are mutually attractive, leading to polymer adsorption and bridging interactions. (3) Interactions with CNCs may interfere with the crystallization of PVA. Any one or a combination of these scenarios may be occurring during the formation of the PVA-CNC hydrogels. In this paper, we explore the properties and structures of PVA hydrogels containing CNCs relative to pure PVA hydrogels and attempt to understand the nature of the interactions which give rise to these differences.

6.3. Experimental

6.3.1 Materials

Polyvinyl alcohol (25,000 g/mol, 98.5 mol % hydrolyzed, atactic) was purchased from Polysciences, Inc. and used as received. Aqueous suspensions of CNCs (4.4 wt. %, 0.59 ± 0.01% S) from softwood Kraft pulp were kindly provided by FPInnovations. The CNCs were in acidic form with H\textsuperscript+ counterions associated with the surface sulfate ester groups introduced during the mineral acid hydrolysis. The CNCs were filtered through glass microfibre filters prior to use.
6.3.2 Atomic force microscopy (AFM)

Tapping-mode AFM images of the CNCs were obtained using an MFP-3D™ from Asylum Research. Samples for AFM were prepared by placing a drop of dilute suspension on a poly-(L)-lysine treated mica substrate, followed by rinsing in deionized water and drying under a gentle flow of argon.

6.3.3 Preparation of PVA solutions containing CNCs

In all solutions, the concentration of PVA was 15 g PVA/100 g solvent and the CNC content varied from 0, 5, 10 or 20% of the PVA mass. The aqueous CNC suspension was added to a flask containing PVA, followed by dilution with deionized water. The mixtures were left stirring in sealed flasks for a minimum of 24 hours to facilitate dispersal of polymer and CNCs, followed by heating in a 90 °C oven for 6 hours until complete dissolution of polymer. The mixtures were then cooled and stirred at room temperature for at least 24 hours.

6.3.4 Preparation of CNC-loaded hydrogel

The aqueous PVA-CNC mixtures were poured into a rectangular Plexiglas® mold (outer dimensions = 9.5×6×0.6 cm, inner dimensions = 8.2×4.8×0.15 cm) and covered with a flat Plexiglas® sheet (9.5×6×0.6 cm) flush against the mixture. The assemblies were placed in the freezer and weighted with a metal plate (~0.2 kg) in order to hold everything in place and to ensure that any surplus mixture would be squeezed out of the mold. The mixtures were subjected to 5 successive freeze (-20 °C, 18 hour) and thaw (RT, 4 hour) cycles. For the final cycle, the freeze typically exceeded 18 hours and the thaw was defined in terms of the subsequent experiment.

6.3.5 Polarized optical microscopy

A Nikon Eclipse LV100POL microscope was used with a 530 nm waveplate to observe initial solutions and to compare different cycle hydrogels, prepared with varying amounts of CNCs, under polarized light. The solutions were placed in hollow rectangular
glass capillaries which had been sealed at both ends with Parafilm®. Capillaries which cracked due to sample expansion upon thawing were discarded.

6.3.6 Attenuated Total Reflectance (ATR) – Infrared Spectroscopy

Spectra were obtained using the MIRacle™ ATR accessory (Pike technologies) in conjunction with a Spectrum BX FTIR spectrometer (PerkinElmer). Spectra of freeze-dried samples were recorded using a diamond crystal plate and each spectrum was an average of 16 scans with 4 cm⁻¹ resolution.

6.3.7 Scanning electron microscopy (SEM)

Xerogels for SEM were prepared as described by Trieu and Qutubuddin¹⁹. The gels were sliced to expose cross-sectional areas, dehydrated in ethanol and by critical point drying (CPD). This method prevents shrinkage and collapse of the pore structure with drying. Prior to imaging with a Hitachi S-4700 cold field emission scanning electron microscope, the hydrogels were coated with Au-Pd using a Technics Hummer IV sputter coater. Gel surfaces were imaged by coating freeze-dried samples with Au-Pd.

6.3.8 Differential scanning calorimetry (DSC)

The crystallinity of the hydrogels was studied using a TA Instruments Q2000 DSC with a heating rate of 5 °C per minute and sample masses of 5-10 mg. The hydrogels were freeze-dried prior to measurement in order to prevent water evaporation from overwhelming the PVA thermal events.

6.3.9 Swelling

Using a circular bore (diameter = 5 mm), frozen samples were cut from the gels at the end of the final freezing step and quickly weighed while still frozen. The samples were then submerged in deionized water and weighed at specific time intervals for a minimum of 420 minutes. (The experiments were repeated with a 1.2 cm diameter bore to assess size related effects).
6.3.10 Compression experiments

A complete description of the apparatus and theoretical background are presented by Quinn and Grodzinsky. Prior to the experiment, the gel sample which had been previously thawed and equilibrated in deionized water, was cut into a cylindrical slice (diameter = 12 mm, thickness ~1.5-2 mm) using a circular bore. The gel was sandwiched between a porous, rigid barrier and a non-porous, rigid barrier, and was constrained laterally in order to prevent outward expansion during the uniaxial compression. The porous barrier was in contact with an external reservoir containing deionized water, permitting fluid flow between the sample and bath. The assembly, including reservoir, was mounted between a displacement actuator and a load cell. The displacement actuator controlled gel thickness ($d$) and was used to apply precise compressive strains ($\varepsilon$), defined relative to free-swelling gel thickness ($d_0$) according to equation 1. The load was transmitted through a loading pin to a load cell (1000 g maximum load). In a typical experiment, the samples were compressed in increments of 5% strain, with the thickness of the gel held constant after each compression, up to a total of 25% strain. The stress transients were recorded at each compressive step and consisted of a sharp increase in stress followed by relaxation to a new equilibrium value (~2-3 hours). Equilibrium stress was defined as a stress value which did not fluctuate by more than 2 g over the span of 1000 seconds. The equilibrium stress-strain behavior of the gels provided the confined compression elastic moduli as a function of strain. Stress relaxation transients were exponential in character when close to equilibrium with a time constant ($\tau$), defined in equation 2, which depended upon the product of modulus ($H_A$) and permeability ($\kappa$). The evolution of strain through the tissue is described by a mechanical diffusion equation (equation 3), where $D_M$ (m$^2$/s) is the mechanical diffusivity and describes the ability of strain to diffuse through the soft tissue. Reasonably, $D_M$ increases with permeability and elasticity since the greater either of these factors, the easier it is to displace the fluid with compression (i.e. to dehydrate the gel) and for the strain to dissipate through the solid components of the gel.

$$\varepsilon = \frac{d_0 - d}{d_0} \quad (6.1)$$
\[
\tau = \frac{d^2}{\pi^2 H \kappa (1 - \varepsilon)}
\]

(6.2)

\[
\frac{\partial \varepsilon}{\partial t} = D_M \frac{\partial^2 \varepsilon}{\partial \varepsilon^2}; \quad D_M = \left[ H \kappa (1 - \varepsilon) \right]_{\text{eq}}
\]

(6.3)

6.4. Results and Discussion

6.4.1 Preparation of PVA hydrogel samples

PVA hydrogels (25 kDa, 15 wt. %) were prepared with varying CNC loadings (0, 0.75, 1.5 and 3.0 wt. %) corresponding to 0, 5, 10 and 20% of the dry weight of polymer. The molecular weight of PVA was in the low range compared to other PVA hydrogel studies but was sufficient to produce reasonably solid gels after 2-3 freeze-thaw cycles. Complete dissolution of PVA was confirmed by inspection of the polymer solutions and mixtures using polarized light microscopy, where undissolved PVA crystallites appear bright and chunky. There was no evidence of undissolved PVA crystallites in the final solutions and mixtures after heat treatment (90 °C, ~6 hours).

6.4.2 Effect of heat treatment upon CNC sulfate content

The dimensions of the CNCs used in this study were determined from AFM height images (Figure 6.1): the lengths were between 100-300 nm and the widths were on the order of 10 nm. In order to determine whether the heat treatment diminished the CNC sulfate contents, CNC suspensions (0.75, 1.5 and 3.0 wt. %) were subjected to identical heat treatments, followed by routine clean-up (dialysis, treatment with mixed bed and cationic ion exchange resins) and conductometric titration. The titration results (Table 6.1) indicated that sulfate groups were diminished but still present to a significant extent on the surfaces of the heat treated CNCs and it seems reasonable to conclude that desulfation does not play a major role in the gelation of the PVA-CNC mixtures. An increase in desulfation with CNC concentration was observed and is due to the greater number of acidic groups in the more concentrated samples (i.e. desulfation is an acid-catalyzed reaction). Preliminary gelation experiments performed using the more
thermally stable Na-form CNCs, in this case derived from the sulfuric acid treatment of cotton, showed no significant counterion effect.

Figure 6.1: AFM image of a dilute suspension of CNCs. (Scale bar = 1 µm)

Table 6.1: Sulfur content (\(\%S = \text{g sulfur/g cellulose} \times 100\%\)) of CNC blank solutions (i.e. aqueous CNC suspensions) before and after heat treatment (6 hours, 90 °C).

<table>
<thead>
<tr>
<th></th>
<th>Initial % S</th>
<th>Final % S</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.75 wt. % CNC</td>
<td>0.59 ± 0.01</td>
<td>0.40 ± 0.07</td>
</tr>
<tr>
<td>1.5 wt. % CNC</td>
<td>0.59 ± 0.01</td>
<td>0.36 ± 0.04</td>
</tr>
<tr>
<td>3.0 wt. % CNC</td>
<td>0.59 ± 0.01</td>
<td>0.27 ± 0.04</td>
</tr>
</tbody>
</table>

6.4.3 Polarized optical micrographs

In Figure 6.2 polarized light micrographs obtained using a 530 nm red waveplate are presented for pure PVA and a 1.5 wt. % CNC hydrogels at three different stages: a) initial solution prior to freezing, b) 3 cycle hydrogel and c) 5 cycle hydrogel. In these images isotropic regions are pink and anisotropic areas, in the plane of the microslide, generally show up as either blue or yellow, the difference in color simply a 90 ° in plane rotation. The pure PVA hydrogel remained isotropic with increased cycling (although the
gel surface appeared rough and textured after 3 cycles), whereas relatively large microdomains of aligned CNCs emerged in the freeze-thaw processing of the PVA-CNC hydrogels. Interestingly, liquid crystalline ordering was apparent in the solution prior to thermal cycling (Figure 6.2d) but became more distinct with processing. In the absence of PVA, CNC suspensions with concentrations of 0.75 to 3.0 wt. % appear isotropic.

Figure 6.2: Polarized optical micrographs of a pure PVA hydrogel and 1.5 wt. % CNC-loaded hydrogel obtained at different stages of freeze-thaw processing: (a) pure PVA pre-cycling, (b) pure PVA after 3 cycles, (c) pure PVA after 5 cycles, (d) CNC-loaded mixture pre-cycling, (e) CNC-loaded hydrogel after 3 cycles and (f) CNC-loaded hydrogel after 5 cycles.

After 5 processing cycles, the translucent gels had a rubbery consistency and were solid enough to be handled (Figure 6.3), although the pure PVA hydrogel was noticeably more fragile compared to the CNC-loaded samples. Figure 6.4 depicts the four gel samples at the end of 5 freeze-thaw cycles and the increase in anisotropy with CNC loading. The 0.75 wt. % hydrogel was characterized by distinct stretches of isotropic and anisotropic regions which seem indicative of CNC rich domains within the hydrogel, possibly caused by phase separation. The 1.5 wt. % sample appeared entirely anisotropic with relatively large micro-regions of oriented CNCs, whereas the 3 wt. % hydrogel was more fragmented and showed some interference colors possibly due to overlapping CNC domains. The absence of isotropic regions may be indicative of improved interactions between CNCs and PVA.
Figure 6.3: CNC-loaded PVA hydrogel after 5 freeze-thaw cycles (thickness ~2 mm).

Figure 6.4: Polarized optical micrographs of hydrogel samples after 5 freeze-thaw cycles: (a) pure PVA, (b) 0.75 wt. % CNC, (c) 1.5 wt. % CNC and (d) 3.0 wt. % CNC. Scale bars are 100 µm for (a), and 500 µm for (b), (c) and (d).
6.4.4 Infrared analysis

IR analysis of a pure PVA hydrogel, CNC-loaded hydrogels and a CNC film are presented in Figure 6.5. The CNC-PVA hydrogels possessed peaks characteristic and unique to both PVA and CNCs. CNC absorptions at ~1050 and 1027 cm\(^{-1}\) (weak) arose from C-O and O-C-O stretching vibrations, and PVA absorptions at ~1700 cm\(^{-1}\) from residual C=O groups (i.e. polyvinyl acetate starting material) and 1080 cm\(^{-1}\) from C-O stretches. The O-H stretching band in the gel samples was strong and broad, relative to the CNC film, indicative of significant hydrogen bonding.

![Image of IR analysis](image_url)

Figure 6.5: IR analysis of CNC-loaded hydrogels, a pure PVA hydrogel and an evaporated CNC film.

6.4.5 Scanning electron micrographs

Scanning electron micrographs of the gel samples are presented in Figure 6.6 and the interior morphologies (i.e. perpendicular to the gel surfaces) were found to be highly dependent upon CNC loading. The general morphological trend observed was an apparent shift of the pore size distribution to smaller pores as the CNC loading increased.
from 0 to 1.5 wt. %, followed by the re-emergence of larger pores in the 3.0 wt. % sample. Pore sizes are thought to reflect the dimensions of the ice regions formed during the freeze cycles, which in turn will be influenced by the polymer and CNC concentrations. The observation of larger pores in the 3.0 wt. % sample may thus be due to the decrease in free PVA volume fraction in the presence of higher CNC concentrations. For instance, Yokoyama et al.\(^3\) noted an increase in pore size with decreasing PVA concentration. The surfaces of the hydrogels were also imaged by SEM and were found in all cases to be relatively smooth and unremarkable.

**Figure 6.6**: Scanning electron micrographs of hydrogel samples: (a) pure PVA, (b) 0.75 wt. % CNC, (c) 1.5 wt. % CNC and (d) 3.0 wt. % CNC. Scale bars are 1 \(\mu\)m.

### 6.4.6 DSC thermograms

Differential scanning calorimetry (DSC) was used to explore the effect of CNCs upon the crystallization of PVA in the hydrogels. The first heating curves for a pure PVA hydrogel and for a CNC-loaded sample are presented in Figure 6.7 and the full set of thermal data in Table 6.2. The melting point \(T_m\) of the hydrogels decreased with added...
CNCs, from 224 °C for the pure PVA hydrogel to ~218 °C for the hydrogels containing CNCs. The percent crystallinity was obtained by dividing the heat of melting of the hydrogels by the heat of melting of a 100% crystalline sample of PVA with a value of 138.6 J·g⁻¹. The depressions in melting point and percent crystallinity are good indications that the introduction of stiff, rod-like, nm-scale particles interferes with the ordering of the polymer chains. The decrease in PVA crystallinity with CNC loading may also be indicative of a growing preference toward mutual interaction as opposed to self-association. By this reasoning, the expected decrease in gel strength due to the diminished polymer crystallization may be more than compensated by improved interaction and miscibility of the PVA and CNC phases.

Figure 6.7: First heat DSC curves for a pure PVA hydrogel and a CNC-loaded hydrogel.
PVA gels reinforced with CNCs

Table 6.2: DSC data of hydrogel samples.

<table>
<thead>
<tr>
<th></th>
<th>$T_m$</th>
<th>$\Delta H_m$ (J/g)</th>
<th>% C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PVA gel</td>
<td>224</td>
<td>55</td>
<td>39</td>
</tr>
<tr>
<td>0.75 wt. % CNC</td>
<td>219</td>
<td>50</td>
<td>36</td>
</tr>
<tr>
<td>1.5 wt. % CNC</td>
<td>218</td>
<td>~42</td>
<td>~30</td>
</tr>
<tr>
<td>3.0 wt. % CNC</td>
<td>218</td>
<td>36-44</td>
<td>26-32</td>
</tr>
</tbody>
</table>

6.4.7 Swelling in water

The swelling of the hydrogels in water over time was demonstrated by the water sorption curves presented in Figure 6.8. The swelling ratio is defined relative to the frozen sample weight since thawing is considered a swelling event. The swelling behavior is dominated by the porosity and crystallinity of the gels, with increased water uptake expected for more porous and/or amorphous samples, and by the hydrophilic nature imparted to the gels by the CNCs. The water uptake of the hydrogels at equilibrium (i.e. the plateau region of the sorption curves) increased with CNC content. The hydrogels containing CNCs, particularly the 1.5 and 3.0 wt. % samples, maintained their integrity on swelling (up to at least 1 year for samples stored in deionized water) in contrast to the pure PVA sample which immediately began to disintegrate along the cut edges when submerged in water. Results from the swelling experiments are presented in Table 6.3. The time required to achieve equilibrium water saturation increased with CNC content and the mass increase at equilibrium relative to the initial gel mass nearly trebled as the CNC loading was increased to 3.0 wt. %. The experiment was repeated with a 1.2 cm circular bore (results not shown) with a similar result of increased swelling in water with CNC loading.
Figure 6.8: Swelling ratio, $W/W_f$ (swollen sample weight relative to frozen sample weight), plotted against time for hydrogel samples with varying CNC contents.

Table 6.3: Swelling results for hydrogel samples, where $t_{eq}$ is the minimum time required for equilibrium saturation, $\% W_{gain}$ is the increase in sample mass at equilibrium relative to the initial weight and $t_{degr}$ is the time where sample degradation was first observed.

<table>
<thead>
<tr>
<th></th>
<th>$t_{eq}$ (hr)</th>
<th>$% W_{gain}$</th>
<th>$t_{degr}$ (hr)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PVA gel</td>
<td>~2</td>
<td>8</td>
<td>&lt;1</td>
</tr>
<tr>
<td>0.75 wt. % CNC</td>
<td>~3</td>
<td>12</td>
<td>&gt;6</td>
</tr>
<tr>
<td>1.5 wt. % CNC</td>
<td>~4</td>
<td>19</td>
<td>&gt;6</td>
</tr>
<tr>
<td>3.0 wt. % CNC</td>
<td>~5</td>
<td>23</td>
<td>&gt;6</td>
</tr>
</tbody>
</table>

6.4.8 Mechanical properties of hydrogels

The mechanical properties of the hydrogels were studied using radially-confined compressions. The crux of the experiment was to study the stress response of the gels to a successive series of applied compressive strains ($\varepsilon$), while maintaining constant thickness and allowing the gels to relax back to an equilibrium stress value ($\sigma_{eq}$) after
PVA gels reinforced with CNCs

each compressive step. A representative set of data is presented in Figure 6.9. As the gel
is compressed and dehydrated (i.e. water is squeezed out through the porous, rigid
surface), strain gradients are established within the gel which over time smooth out to a
uniform strain at the new equilibrium state. The $\sigma_{eq}$ versus $\varepsilon$ plots are presented in Figure
6.10. The confined compression moduli ($H_A$) of the gel samples as a function of strain are
obtained from the slopes of the $\sigma_{eq}$ versus $\varepsilon$ plots and, in general, were found to increase
with applied strain (Figure 6.10 inset). From the equilibrium stress versus strain curves, it
is evident that the moduli of CNC-loaded gels are increased compared to the pure PVA
hydrogel. The trend in chord moduli seems to be as follows, 1.5 wt. % > 0.75 wt. % ~ 3.0
wt. % > pure PVA hydrogel. This result may indicate that at loadings greater than 1.5 wt.
%, the CNCs disturb rather than reinforce the hydrogel structure. It may also simply
highlight experimental uncertainty, where while it is clear that CNC-loaded samples have
increased mechanical properties compared to pure PVA gels, the trend with CNC content
may be more difficult to interpret. It may be important to note that while it is necessary to
pre-equilibrate the gel samples in order to prevent expansion during the experiment, the
gel structure is most likely somewhat weakened by swelling (i.e. H-bond rupture).
Consequently, the observed reinforcement effect may be undermined, particularly in the
case of the most swollen 3.0 wt. % sample since equilibrium swelling increased with
CNC content.
Figure 6.9: Schematic data of stress (solid lines) and sample thickness (dotted lines) transients over 4 compressive steps, with equilibrium stress values ($\sigma_{eq}$) and sample thicknesses ($d$) shown at each step.
PVA gels reinforced with CNCs

Figure 6.10: Plots of equilibrium stress versus strain and chord modulus versus strain (insert). Duplicate points were measured for each composition, to give some indication of experimental reproducibility: pure PVA (♦), 0.75 wt. % CNC (◊), 1.5 wt. % CNC (■) and 3.0 wt. % CNC (□).

In Figure 6.11, $\kappa$ versus $\varepsilon$ plots for the four gel samples are presented. In units of m$^2$/Pa·s, $\kappa$ takes into account the resistance to flow due to the viscosity of the fluid. At low strains, the trend in $\kappa$ is not a clear function of CNC content, possibly due to the first compressive step being too large. Indeed, a similar experiment using 3% strains (data not presented) indicated an initial trend in $\kappa$ values of pure PVA ~ 3.0 wt. % CNC > 0.75 wt. % CNC > 1.5 wt. % CNC, possibly related to a trend in sample pore sizes. At sufficiently high strains ($\varepsilon > 15\%$), the $\kappa$ values of the hydrogel samples all approach $\sim 10^{-14}$ m$^2$/Pa·s, a fairly typical value for polymeric gels. The leveling off with strain can be understood in terms of the morphology of the hydrogel samples. The gels are quite soft and the increase in sample density due to compression hinders flow and causes the permeability value to decrease.
Figure 6.11: Hydraulic permeability plotted on a logarithmic scale versus equilibrium strain, with two curves presented for each sample in order to address experimental reproducibility.

6.5. Conclusions

PVA hydrogels reinforced with CNCs have been successfully prepared. All loadings resulted in a decrease in the percent crystallinity of the hydrogel samples perhaps indicative of improved interaction between the CNCs and PVA. The water uptake of the samples increased with increased CNC loading, with the hydrogels containing CNCs exhibiting improved structural integrity upon swelling, evidence further supporting a conclusion of structural reinforcement. Hydraulic permeabilities were related to gross morphology of the hydrogels.

6.6. Acknowledgements

We thank NSERC Canada and FPInnovations/Paprican for financial support. T.A. thanks the Centre for Self-Assembled Chemical Structures (CSACS), H.C. Chin and G.
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6.7. References


PVA gels reinforced with CNCs
Appendix B
Confined-compression experiments

B.1. Background of compression experiments

The theoretical basis of these experiments was outlined by Quinn and Grodzinsky\(^1\) who described in detail the first generation of the instrument utilized in this work. Here the experimental rationale and basic equations relevant to fluid flow in a porous medium (i.e. soft tissue) are reproduced.

Darcy’s law (Equation 1) relates fluid velocity (m\(^3\)/m\(^2\)/s) to gradient in fluid pressure for laminar flow:

\[
U = -\kappa \nabla p
\]

where, \(\kappa\) in units of m\(^2\)/Pa·s is the hydraulic permeability and represents the ease of fluid flow through a porous medium.

Local variations in solid strain are driven by fluid content in the tissue (equation 2):

\[
\frac{d\varepsilon}{dt} = (1 - \varepsilon) \frac{dU}{dx}
\]

where, a positive velocity gradient indicates a net out-flux of fluid from the volume element and vice versa, and the \((1-\varepsilon)\) term takes into account the decrease in volume with strain since,

\[
\varepsilon = \frac{V_o - V}{V_o}
\]
Compression experiments

For small changes in fluid volume fraction ($\varphi$), the force balance equation for soft tissues, which relates the gradients in solid stress and fluid pressure, is as follows:

$$\frac{d\sigma}{dx} = -\frac{dp}{dx}$$

For elastic materials, the bulk elastic modulus ($H_A$) is:

$$H_A = \frac{d\sigma}{d\varepsilon}$$

Equation 2 can be re-written in a form analogous to the diffusion equation, by combining (1), (2), (4) and (5) to describe the diffusion of strain through a soft tissue as follows:

$$\frac{\partial \varepsilon}{\partial t} = (1 - \varepsilon) \kappa H_A \frac{\partial^2 \varepsilon}{\partial x^2}$$

(2a)

The mechanical diffusivity ($D_M$), in units of m$^2$/s, describes how fast or slow the strain is able to diffuse through the tissue. At equilibrium it is given by the pre-factor in equation 2a:

$$D_M = (1 - \varepsilon) \kappa H_A$$

(6)

The solution to the diffusion equation, for confined compression with the condition of $U = 0$ at both boundaries (porous and non-porous rigid interfaces) is:

$$\varepsilon(x,t) = \varepsilon_{eq} + \sum_{n=1,2,3...} A_n \cos \frac{n \pi x}{d} e^{-\left(\frac{n \pi}{d} D_M t\right)}$$

(7)

where, $\varepsilon_{eq}$ is the equilibrium uniform strain, $A_n$ is a constant and $d$ is the tissue thickness.
Near or at equilibrium equation 7 reduces to:

$$\varepsilon(x,t) = \varepsilon_{eq} + A_1 e^{-\frac{(x^2)^2}{d^2}d_{eq}}$$  \hspace{1cm} (8)

Since, stress transients are actually being measured in the experiment:

$$\sigma(x,t) = \sigma_{eq} + A e^{-\frac{(x^2)^2}{d^2}d_{eq}}$$  \hspace{1cm} (9)

where, $A$ is $A_1/H_A$.

**B.2. Data analysis**

In the constrained compression experiments, the measured sample load is converted to stress using the following relationship:

$$\sigma(Pa) = \frac{\text{Load} \times g_o}{A}$$  \hspace{1cm} (10)

where, the load is measured in grams, $g_o$ is the acceleration due to gravity, and $A$ is the area of the sample (i.e. $A = \pi r^2$).

Stress strain curves were simply produced by plotting the equilibrium stress values as a function of equilibrium strain, defined as:

$$\varepsilon = \frac{d_o - d}{d_o}$$  \hspace{1cm} (11)

To obtain, hydraulic permeability ($\kappa$), equation 10 was re-written as a natural logarithm:
Compression experiments

\[
\ln\left[\sigma - \sigma_{eq}\right] = \ln A - \left(\frac{\pi}{d}\right)^2 D_M t \quad (10a)
\]

Finally, from a plot of \( \ln[\sigma - \sigma_{eq}] \) against time, a straight line with a slope of \(-\left(\pi/d\right)^2 D_M\) is obtained. From this slope, values of \( \kappa \) and \( D_M \) can be determined at each equilibrium strain studied, using \( H_A \) values obtained from the slope of the equilibrium stress versus strain curves.

B.3. References

Chapter 7
Conclusions, Current State and Future Work

7.1. Conclusions

From the work presented in this thesis, we may conclude that cellulose polymers and particles show great promise as the constituents of interesting nanocomposite materials. Depending upon source and subsequent chemical treatment, cellulosics are ideal candidates for a vast number of diverse applications, (e.g., fibers, films, fillers, plastics, gels, powders). The impressive variety of properties which can be accessed from a relatively cheap, green and readily available bio-resource, make cellulosics exciting and highly relevant for the preparation of functional nanocomposites.

Films prepared by incorporating quantum dots into cellulose triacetate displayed properties which were additive; the films were fluorescent, flexible and largely transparent. The addition of quantum dots into optically transparent plastics has been previously explored by others, for example there is some history between PMMA and QDs\textsuperscript{1, 2}. Our contribution to the field has been the establishment of the very good compatibility, stability and promise exhibited by quantum dot-cellulose triacetate hybrid materials, and of a polymeric alternative to PMMA or other polymers.

The selection of CTA was well-considered, since the conversion of CTA to cellulose allowed the possibility of water-dispersible applications. By using some simple and well-known chemistry, we were able to improve the adhesion between cellulose pulp fibers and the films, and to prepare fluorescently marked papers using a wet-process. Perhaps most importantly, we converted a hydrophobic film into a hydrophilic-compatible system, without significant sacrifice to bulk properties. The application of fluorescent film fragments as security markers in papers is, to the best of our knowledge, entirely unique.

Fluorescent, sub-micron diameter fibers were also prepared from the QD-CTA system by electrospinning. Cellulose polymers, including CTA,\textsuperscript{3} have previously been electrospun into fibers, and we relied upon the existing knowledge in order to prepare isolated fibers and non-woven fibrous mats based upon our novel fluorescent system. Electrospinning has also been applied to other polymer-QD mixtures\textsuperscript{2, 4-6}, however the
Conclusions

advantage of our approach lies in the use of a bio-based polymer which has well-established scientific applications and compatibility with topo-capped CdSe/ZnS QDs.

Additionally, we showed that it was possible to chemically link quantum dots to the reducing end of cellulose nanocrystals. During the course of our work, some success was achieved by other researchers using FITC to label CNCs\(^7,8\), but we had hoped that the advantages of QDs over conventional fluorophores would make our system more robust. While the separation of desired product was problematic, we have demonstrated the feasibility of this approach.

The use of cellulose nanocrystals as reinforcing fillers in polymeric materials, in this case polyvinyl alcohol hydrogels, was also successfully demonstrated. The work clearly shows dramatic changes in nanocomposite properties which are accessed by very small changes in CNC contents. This type of behavior, where the incorporation of relatively low loadings of nanoparticle fillers (i.e. at or above the percolation threshold) leads to an enhancement in material properties due to the formation of a network structure of “communicating” particles, has been observed for other cellulose nanoparticle-polymeric materials.\(^9\) Our work is unique in the combination of nanoparticle, polymer, nanocomposite preparation and mechanical characterization. The mechanical testing method has previously been applied to pure polymer gels\(^10\), but not to nanocomposites.

7.2. Current status

In general, interest in cellulosics, particularly CNCs, has never been greater. Several excellent reviews looking at the properties and applications of CNCs have recently been published in quality journals.\(^11-15\) Additionally, the recent announcement of a CNC pilot plant to be built in Windsor, Quebec, jointly run by Domtar and FPInnovations, and backed by substantial government investment, has garnered media attention and is testament to the current mainstream buzz surrounding CNCs.

Interest in the QD-CTA system established in the Gray lab has been increasing. Recently, our work was cited in a 2010 paper\(^16\) describing the incorporation of CdSe/CdS nanorods into CTA polymer films. This study further established the compatibility of CTA with semiconductor nanoparticles by showing that the fluorescent lifetimes of the
particles is unchanged by dispersal in CTA. It was demonstrated that the quantum efficiency of the composites was improved compared to the nanorods in solution, suggesting some polymeric stabilization. However, aligned agglomerates of nanorods were observed, possibly indicating some colloidal instability. The general interest in cellulosic-QD nanocomposites has been sustained, some recent examples include a study by Luna-Martinez et al. \(^{17}\) which described the preparation of ZnS-carboxy methyl cellulose nanocomposites and a study by Niu et al. \(^{18}\) which looked at the layered deposition of CdSe particles alternating with stearic acid on titania coated cellulose nanofibers. In our own laboratory, we have moved forward with the granting of a U.S. patent for the use of surface-hydrolyzed QD-CTA films in security papers.\(^{19}\)

CNC-PVA nanocomposites films have been previously prepared by other researchers\(^{20, 21}\) and, during the course of this work, CNC-PVA nanocomposites were prepared by the Rojas group at NC State University\(^{22}\) and by a group at Wuhan University\(^{23}\). All of these studies concluded that the combination of PVA and CNCs held promise, and observed a reinforcement effect with the addition of CNCs to the matrix. Kvien and Oksman\(^{20}\) showed that the alignment of CNCs within a PVA matrix improved the dynamic modulus of the nanocomposite by \(\sim 2\) GPa in the aligned direction, compared to the transverse properties. Roohani et al.\(^{21}\) looked at CNC-reinforced PVA films, and found that CNC-polymer interactions became stronger with PVA samples which had a higher degree of hydrolysis. Peresin et al.\(^{22}\) confirmed the previous conclusion (i.e. better interactions with highly hydrolyzed PVA), and found an increase in elastic modulus with increased CNC content. They also observed a decrease in PVA crystallization in the presence of CNCs when compared to fibers electrospun from neat PVA. Most significantly, while our work was already well underway, a paper\(^{23}\) which looked at the reinforcement of PVA hydrogels (i.e. sponges) with CNCs was published. However, the methods and conclusions, though complimentary, were different enough to warrant the publication of our work. The main difference between our work and the work presented by Wang et al.\(^{23}\) was the addition of starch into the hydrogel-system at contents equal to PVA (2.5 wt. %). The experiments and conclusions presented by Wang et al.\(^{23}\) were very well-considered and illustrated the merit of using starch as a cheap filler material. Our
interest was to attempt to better understand the fundamental interactions existing in the CNC-PVA-water system, and we believe complete understanding is still wanting.

7.3. Future Work

With regard to the fluorescent QD-cellulosic system, the possibility and conditions of metal leaching from the films needs to be explored and may impinge upon commercialization of the technology. It may also be of interest to optimize the deacetylation conditions for the fluorescent CTA fibers and to prepare papers using these fibers, or to explore other water-compatible applications. Perhaps the real promise in the QD-CTA fibers lies in photonic applications, which were not explored in this work. The optical properties of the films should be understood in greater detail in order to better assess whether the observed blueing is indicative of core degradation, which may have serious implications regarding the lifetime of the material. For the covalent attachment of QDs to cellulose nanocrystals, several different avenues may be explored in the future: (1) improved separation of the target complex from unreacted materials, perhaps using size exclusion chromatography, (2) the use of a stronger oxidant (i.e. TEMPO) to oxidize a greater number of alcohol groups, and perhaps allowing the surfaces of the CNCs to be entirely decorated by QDs, and (3) the exploration of completely different chemistries, e.g., targeting cellulose binding domains, introducing streptavidin and biotin functionalities.

Aside from the obvious tweaking of experimental parameters (e.g., MW, degree of hydrolysis, number of cycles), further understanding of the fundamental behavior of the CNC-loaded PVA hydrogel system is needed. Stepping back, it may prove useful for researchers to gain a better comprehension of the effect of freezing on CNCs, and how the presence of CNCs alters the crystallization of ice. It is anecdotally known that freezing results in the sometimes irreversible aggregation of CNCs, perhaps due to phase separation similar to that observed for pure PVA hydrogels (i.e. as the ice crystals form, “impurities” are rejected from the growing crystal structure, resulting in “impurity”-rich and “impurity”-poor regions). It may be useful to study freezing-induced CNC aggregation by dynamic light scattering or viscosity measurements. Additionally, the freezing/melting DSC thermograms of ice in the presence of CNCs may help us better
understand whether CNCs affect the ability of ice to crystallize. DSC and NMR can also provide information regarding the structures of water in the system, for example these techniques can be used to differentiate between bound water which may be unable to freeze due to limited mobility, and unbound water which can freeze and melt. This knowledge may be helpful toward a fuller understanding of the interactions between PVA, water and CNCs in the hydrogel system, and in general, may improve our understanding of aqueous CNC systems.

7.4. References

Conclusions


