Applications of Optical Spectroscopy in Studies on Energy & Electron Transfer and Solvation Effects in Nanoscale and Molecular Systems

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Abstract

This thesis describes three investigations, ranging in subject matters, all of which relating to systems capable of photoinduced reactions involving energy or electron transfer. The phenomenon and the effects of environment in the various systems are explored using different methodologies of optical spectroscopy. As the chapters progress, different investigations introduce and build on fundamental concepts encountered and in complexity of the methodologies used to explore the systems.

The first chapter introduces the preparation of water-soluble CdSe nanocrystal clusters. The clusters, created using a protein, are 3-D close-packed self-assemblies of nanocrystals. Due to this close-packed nature, electronic interactions between the nanocrystals allow for energy migration within the cluster. The structural and optical properties of the clusters were described. Then using steady-state spectroscopy, properties of the original nanocrystals were compared to that of the cluster to determine the consequence of nanocrystal coupling interactions and their potential use toward the development of artificial light-harvesting systems.

In the second chapter, CdSe nanocrystals are functionalized with a unique electro-active polymer, and the electron transfer between the nanocrystal and the electro-active polymer adsorbate is investigated. Using fluorescence decay measurements, the electron transfer reaction inherent to the system with respect to a comprehensive range of dielectric solvents was explored. The study illustrates the high complexity of seemingly typical nanocrystal-based systems and provides general awareness of what factors need to be considered when dealing with such systems.

The final chapter starts with an informal review of ultrafast nonlinear spectroscopy, focusing on two methods, three-pulse photon echo peak shift (3PEPS) and two-dimensional photon echo (2DPE) electronic spectroscopy, and how they are related. A straightforward approach for extracting 3PEPS data from 2DPE results is presented in a preliminary case study of a dye in two different solvents, one of which is electron-donating.
To my parents
Acknowledgments

I dreamt last night of flying pigs, only to awake this morning to find myself staring at a blank acknowledgment page of my Ph.D. thesis, trying desperately to find the right words to properly thank the handful of important individuals who have influenced my life and shaped my character.

Despite sounding like a cliché, this has definitely been a long and challenging journey. I started my undergraduate degree at the University of Toronto, many many years ago; me and tens of thousands of bright-eyed, freshly-enrolled science students. We all had very high expectations and big dreams. I remember my first day of class in Convocation Hall, sitting on the third floor balcony, staring at a giant screen at the front which held a live picture of my instructor. He started off by saying, “Look to the left of you… then look to the right. I can tell you that one of these people will not make it by the end of year.” After hearing these very first, less-than-encouraging words, I remember thinking I should have gone to McGill instead. Solely, due to the sheer number of students enrolled, it’s been tough growing up at UT.

I was reflecting back on how I ended up here today. I don’t think I ever thought about doing graduate work in physical chemistry, LIKE EVER. I guess it all started at the end of my first year undergrad, when I heard about a “Research Opportunity Program” called 299. I interviewed with a lot of different profs in different programs and decided to do a CHM299 with an old-school, hard-core inorganic guy, Dr. Douglas McIntosh. He turned out to be a really nice, smart guy and introduced me to the world of molecular symmetry, group theory, Gaussian structure calculations and simple computational chemistry. It was an interesting program and by the end, Doug had talked me into taking a few more inorganic and physical chemistry courses. Who would have thought chemistry would be interesting?

By the end of my undergraduate degree, I had taken many chemistry courses and found myself doing summer and 4th year research in various chemistry groups, the Farrar group, the Donaldson group, and the Kumacheva group. I learned many different things – from hard-core inorganic synthesis using schlenk line techniques to studying atmospheric adsorbates using a Knudsen cell and mass spectrometry to playing around with hydrogels and looking at drug delivery systems – from some really bright people, especially Dr. Edmond Lam, Dr. Mima Staikova, and Dr. Hong Zhang. I also had the opportunity of meeting some fantastic profs, Prof. James Donaldson and Prof. Eugenia Kumacheva. Jamie was one of the most encouraging and supportive profs I’ve ever met. Even though I had my doubts on atmospheric chemistry (I’ve never been a huge fan of open systems and giant error bars on data), he instilled an enthusiasm within me regarding research and physical chemistry and a desire to learn more. Eugenia was one out of a handful of female profs found in the chemistry department during my undergrad. I think chemistry had been a fairly male-dominated field (and to some extent, it still is) and the few female chemistry profs (who are very well-known in their fields) found at UT, in my opinion, have been truly remarkable and inspiring for all women professionals. Eugenia is very innovative, tenacious, driven, and a straightforward person and researcher. In a passing conversation with her regarding research and material science (as we were
coming out of a CHM220 exam invigilation), she told me, “all you need are some ideas, and if you are enthusiastic and are willing to try things out… you will do very well.” It seems that she has stood by these words and has done very well, and I think this is a good lesson for us all, not just in materials research but in life. Things don’t need to be complicated, we just need to have some ideas, passion, and determination, and we can do well. In addition, she has made me a huge fan of polymers and materials chemistry. After all these amazing experiences and meeting all these great people, I realized that I wanted to pursue graduate studies in chemistry.

So, I started my graduate work with Prof. Gregory Scholes. The first time I met Greg was actually during my third year phys chem and quantum courses. He was the lab coordinator. He had just started in the department and I remember thinking “what a very young, cool, laid-back prof.” I started in his group not knowing much about optical spectroscopy, but very interested in this cool material called “quantum dots.” I mainly wanted to work with quantum dots, create other new materials and make simple applications of these materials. However, Greg turned out to be a very hard-core, fundamental, theoretical, “let’s do some real research,” “what is the take home message” type of guy. Being in his group has made me face the harder questions and concepts that I generally tended to steer away from. He has made me appreciate the messy fundamental knowledge, chaotic disorder, and hard work that comes along with doing research. I remember one time when I was speaking to Greg and I was very frustrated with my project, he said to me, “Megan, it’s not supposed to be easy. If it was, everyone would be doing it.” I remember thinking, what a crazy bona fide statement. And it was so true that I found myself repeating it every time I became frustrated, and also saying, it’s probably not worth doing, if it is easy. It’s probably why he’s done so well and is such a brilliant, one-of-a-kind researcher. It’s also another lesson learned that I can apply not only to research or science but to life. Greg’s genuine love for science inspires us all, and it actually makes me very envious and makes me want to find something that I could be so passionate about as well. I’ve been fortunate to have him as my supervisor and I thank him for putting up with me for this long.

I would also like to say a special thank you to Prof. Mitch Winnik. I do not know him as well as I feel I should, but my encounters with him have always been very positive. His encouraging words, advice, and support have come at crucial times when I needed it the most, and has helped me to stay positive and motivated in important moments of my academic life. It almost makes me wonder if teachers and professors realize how much impact they have on students’ lives. The smallest of encouraging words and support can drastically alter our views and shift our course of life.

I would like to thank all past and current members of the Scholes group, Alexander, Mayrose, Tienieke, Karolina, Vanessa, Tihana, Cathy, Shun, Anna, Yaser, Sree, Sandeep, Jeongho, Carles, Marcus, Haizheng, Jun, Elisabetta, John, Inchian, Duffy, Evgeny, Francesca, Kyung-Koo, Michelle, Rayo, Yin, Hoda, Cathal, Yasser, Scott, Dan, Chantelle, Yoichi, Jessica, Paul, Ryan, Elsa, Aggie, Aurelia, and Kelly. We’ve gone through several generations now, so I hope I have them all. From the notorious Scholes Group Pub Crawls and boat races (never knew what it was until I joined the group), to lunches at “Yellow” (what was it? combo #46?) and Real Thai (best green curry ever!), to the crazy house parties, awkward Scholes group
moments and Darcy’s solo ramblings on pedagogy and the origins of the word “petrology” (that was a memorable party), to just hanging out at the office and being silly, to “party time,” text twister (the game that kept people from going home), Tetris (and Tihana’s quest for “infinite superiority”), the “digging game” (who the hell just digs straight down?.. that’s like cheating… the point of the game is to get the gems to upgrade your digging machine!), and Plants vs. Zombies (thanks Cathal for introducing my favourite game of all time), to Tienke’s influence on coupon clippings, Redflag deals, free Herbal Essences, and Dominion’s “Fresh or for Free,” to movie/pizza or sushi nights in Davenport East, to Chemistry/Scholes Group vball at the GSU and softball with CUPE, to rollerblading trips with Elisabetta (and the scary-ass bridge on Bathrust that blackened our hands from holding on for our dear lives, and #MegansRock), to the wonderful Wonderland trips and my favourite ride of all time (the Behemoth!), and to a plethora of awesome random times with various group members… the Scholes group has always been full of very memorable characters and it’s been a blast knowing them all. It’s a group that plays hard but also works even harder, and I wish everyone the best and I will miss them all.

I want to thank especially Mayrose and Tihana for all the extra good times I’ve had with them over the years. Whenever there IS a good time to be had, I know Mayrose is always close at hand and ready to party with you, even when she claims that she has no alcohol. Always a great friend to have around. Tihana has always been my partner in crime, or more like in being silly. From all of our energy draining arguments, coffee trips to SB and BS SB related chats, to introducing weird random acronyms to my daily vocabulary, chats on (nano)rods & balls and symmetry (or more like the horizontal rotational axis) of “poop” and “boob,” our more serious chats on how we think chemistry should be taught and our disapproval with Canada’s high school educational curriculum, our crazy bike ride from Etobicoke to the Beaches followed by massive brunch at Hot House and food coma to the point we could not ride our bikes back home (so very epic!), our random long walks to various places and Bambino nights, our trips to awesome Serbian restaurants (you with your rod platter and me with my mixed meat platter for two, and the amazed Serbian spectators), to late night phone conversations on the most random topics… we’ll always have our agreement over our beloved “crayons.” We argued a lot, but we also laughed a lot, and I’ve had a really fun time just hanging out with you. Hopefully, we’ll never grow up.

And to save the best for last, I would like to thank my amazing parents. Two of the most hardest working, sincerest, most loving parents in the world. Without them, none of this could have ever been possible. They are my source of hope, strength, and determination. I am who I am because of them. I love you very much! To my brother… hope you are well and we miss you lots.

And, I think “we’ve come full circle.”

“With one hand the past moves us forward, and with the other it holds us back.”
Lilith Sternin
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Charge Transfer in CdSe Nanocrystal Complexes with an Electro-Active Polymer

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Chapter 1

Energy Transfer in Water-Soluble CdSe Quantum Dot Clusters
1.1 Introduction

The most notable nanotechnology for energy conversion is found in the natural photosynthetic systems of plants and algae. Arrays of specialized pigments, arranged in light-harvesting pigment-protein complexes, absorb sunlight predominately in the visible region and transfer their excitations to collection units called reaction centers, where the energy is used to drive chemical reactions necessary for biological functioning. The efficiency at which the excitation energy is transferred in these networks usually exceeds 90% with excitation energy collection over hundreds of pigments and single-step hopping times in the sub-picosecond ranges.\textsuperscript{1-4}

Assembly of molecular-scale building blocks into functional nanoscale architectures, such as an artificial light-harvesting complex, currently belongs to a very popular and active area of research in materials chemistry.\textsuperscript{5-10} There are many studies reported of man-made multiple dye excitation energy transfer systems, some with fairly high efficiencies.\textsuperscript{11-15} Dye molecules are a popular subject of interest for fundamental research and applications in photochemistry due to their high quantum yield, small molecular size, reasonable photostability, and great biocompatibility. For every application, there exists a specific dye, in the vast catalog of dyes, suited for the situation. Despite these great advantages, many organic dyes suffer one main drawback which is that they cannot be efficiently used over a wide spectral range beyond their characteristically narrow absorption spectra, limiting their use in some important applications that require broader or extended spectral operation. In recent decades, researchers have introduced a contending material which overcomes this shortcoming with its convenient “one size fits all” approach to materials chemistry, the quantum dot.

Colloidal semiconductor nanocrystals (NCs) have been exhaustively studied due to their unique quantum confinement effects and size dependent characteristics. It now seems that the versatile optical and electronic properties of these nanocrystals seem to be reasonably well understood. However, many applications involve coupling of these nanocrystals to other species (i.e. fluorophores, electron donor/acceptors, or other semiconductor nanocrystals), with the trend now shifting from understanding the fundamental properties to creating and manipulating secondary structures of the nanocrystals; utilizing the unique size-dependent properties of the individual nanocrystals and tuning their collective properties in the larger, more complex, coupled systems. These coupled systems have attracted considerable attention due to their potential use toward the realization of artificial light-harvesting systems and other typical nanophotonic systems from a fundamental perspective.\textsuperscript{16-18}
In such systems, excitation energy transfer due to electronic coupling between the nanocrystals plays an essential role. However, more studies regarding the interactions of nanocrystals with other nanocrystals in close-packed systems are required to better understand these effects. The coupling observed in these systems may render the unique optical and electronic properties of the individual nanocrystals partially or entirely distorted, and studies that probe the consequences of such coupling interactions will prove to be useful and provide insights into improving and controlling many NC-based devices.

This chapter will focus on such a system as the one described above, the water-soluble CdSe quantum dot cluster. The following sections will describe some pertinent background information, preparation, characterization, and energy migration between the quantum dots observed in this model system.

1.2 Cadmium Selenide Semiconductor Nanocrystals (Quantum Dots)

What are quantum dots? Semiconductor nanocrystals (NCs), otherwise known as quantum dots (QDs), are semiconductor materials with physical dimensions typically ranging from a few to tens of nanometers, and involve roughly 100 to 10,000 atoms. Their size is an essential quality bestowing them their unique properties that deviate from those of bulk materials. Characterized by their widely tunable optical properties and photostability, semiconductor nanocrystals have motivated a large body of work based on their potential for various applications such as medical imaging (biolabeling), photovoltaics, light emitting devices, gain materials for lasers, and quantum computing. In particular, CdSe is a II-VI type semiconductor (categorized by its elemental groups) and is a binary compound made of cadmium and selenium. It is the prototypical quantum dot. Much of the fundamental research on semiconductor nanocrystals has been focused on CdSe nanoparticles, concentrating on developing controlled syntheses, understanding the properties, and accordingly applying these materials in useful and interesting ways. Since the topic of CdSe semiconductor nanocrystals will be the focus of the first two chapters of this thesis, the following subsections will delve further into some of their notable properties that are relevant to this work, and define terminologies that are used in succeeding sections and chapters.
1.2.1 Physical Properties

When the spatial dimensions of semiconductors become comparable to the de Broglie wavelength of the electron wave function, interesting things begin to happen. Normally, when bulk semiconductors are excited with a particular frequency of light, they form an electrostatically bound electron-hole pair, known as an exciton. The electron-hole pair behaves freely in the unrestricted dimensions of a seemingly infinite crystal structure and the energy states for the electrons and holes are continuous with a size-independent, material specific bandgap between the valence and conduction band. This picture changes, as the exciton is squeezed into a spatially confining dimension, like in a quantum dot. Consequently, semiconductor nanocrystals show properties which are intermediate, between those of bulk semiconductors and those of discrete molecules. As well, its electronic and optical properties can be manipulated by synthetically changing the size and shape of the nanocrystal, hence changing the depth of the potential and dimensions of confinement. This subsection will look more closely at some of these details and will attempt to explain the diverging distinctions that arise from the dimensional miniaturization of these materials.

1.2.1.1 Crystal Structure

A crystal structure is a particular repeating arrangement of atoms or molecules, which exhibit long-range order and symmetry, in a crystal. A periodic arrangement of an array of points (i.e. atoms or molecules) in space is known as a lattice. The smallest building unit in a lattice is a unit cell. The lengths of the edges and angles of a unit cell can vary giving the lattice distinct parameters. Bravais lattice are distinct lattice types, composed of distinct unit cells, from which all crystals can be constructed. The structure and symmetry of a crystal is significant in determining many of its properties, such as the electronic band structure and optical properties.

The crystal structure of semiconductor nanocrystals can vary depending on the temperature, pressure, monomer concentration, and type of ligands used during the colloidal synthesis. In the case of CdSe nanocrystals, there are three different structural forms that exist, zinc-blende/sphalerite (cubic), wurtzite (hexagonal), and rock-salt (cubic). The unstable zinc blende structure may be observed early, under ambient conditions, in the synthesis. However, upon further heating, above the melting temperature of Se (220°C), the nanocrystal transforms into the better ordered and stable wurtzite form. The rock-salt structure is only observed under high pressures. It is less common and will not be further discussed.
In many crystalline solids, the configuration of atoms, in relation to unit cells and lattices, can be explained in terms of hard spheres packing together in the closest possible arrangement. There are two simple regular lattices that allow for this highest average density, cubic close-packed (ccp) (also called face-centered cubic (fcc)) and hexagonal close-packed (hcp) structures, based on their symmetry. The arrangements are very close to one another in energy, which makes it difficult to predict which form will be favoured based on theory. Both arrangements are based upon sheets of spheres stacked on top of each other. The difference lies in how the sheets are stacked upon one another.

A single layer of spheres can be arranged in a close-packed lattice, where each sphere is in contact with six others (Figure 1a). A second layer of spheres can be stacked on top where the depressions or holes are located on the first layer. For the stacking of the third layer, there are two ways in which this can be accomplished. The spheres may fit into the depressions on the second layer, directly over the positions of the first layer of spheres. Alternatively, the spheres may fit into the depressions where there is no overlap in positions of any of the layers of spheres. The resulting difference in the arrangement of the third layer gives rise to hcp (ABA stacking) and ccp (ABC stacking) structures (Figure 1b). It is called cubic close-packed because the structure can be visualized as layers of hard spheres packed densely and stacked along the spatial diagonal of a cube. It is called hexagonal close-packed because the structure is made up of layers of hard spheres arranged in a hexagonal pattern.

The zinc-blende structure is derived from the ccp structure by stacking the two unit cells into one another, offset by a quarter of a spatial diagonal. One of these identical sublattices is populated by cations (i.e. Cd\(^{2+}\)), the other by anions (i.e. Se\(^{2-}\)) (Figure 1c, left). This results in a tetrahedral coordination for both species. The wurtzite structure is derived from the hcp structure where each of the two individual atom types forms a sublattice (Figure 1c, right). When viewed together, the positions are the same as in a hcp structure. Each atom in the wurtzite structure is also tetrahedrally coordinated.
Figure 1

Crystal Structure of CdSe Nanocrystals

a) Spherical particles can be packed into different arrangements. In closed-packed structures, the spheres are densely arranged in order to occupy space most efficiently (packing efficiency of 74%). Left figure shows the close packing of a single layer of spheres. Additional layers can be stacked on top of the holes from the previous layer. The right figure shows the arrangement of the third layer, giving rise to to 3D packing structures, hcp (ABA stacking) and ccp (ABC stacking).

b) ccp and hcp unit cells are shown to illustrate a different 3D view of the stacked layers. The ccp structure can be visualized as layers of hard spheres stacked along the apatial diagonal of a cube, while the hcp structure is made up of layers of spheres arranged in a hexagonal pattern.

c) Three different structural forms of CdSe nanocrystals exist, zinc-blende (cubic), wurtzite (hexagonal), and rock-salt (cubic) (not shown). The zinc-blende and wurtzite structures are derived from the ccp and hcp structures, respectively. Both structures are also tetrahedrally coordinated (indicated with light red tetrahedrals).
1.2.1.1 Electronic Structure: Bandgaps, Excitons & Quantum Confinement

To identify and understand the physical properties of materials, a good place to start would be to determine the electronic structure of the system. For a physicist or theorist, this can be obtained through quantum mechanics and solving the related Schrödinger equation, which will consequently lead to information on charge density, the potential, and hence the complete Hamiltonian through the calculation of the electronic structure. In the view of a chemist, we start by asking, “What is an electronic band structure?” The electronic band structure of a solid can be defined as a range of “allowed” energies an electron can access or possess. When a large number of atoms are periodically arranged into some crystal structure, the atomic orbitals occupied by electrons of individual atoms start to overlap with those of electrons confined to neighbouring atoms. This leads to a mass delocalization of the electrons extending across all the overlapping or shared orbitals, hence throughout the entire structure. It is from this delocalization and the various electronic interactions that can, more susceptibly, occur which gives rise to the material’s general and fine electronic structures. The electronic interactions are especially influenced by the specific crystal system and Bravais lattice, more specifically the symmetry properties arising from the crystal lattice.

An analysis of the electron waves in the periodic potentials (i.e. periodic/linear combinations of wave functions, known as Bloch functions), defined by the conditions set by the crystal structure, leads to a description of the energy levels of the electrons in the material. The resulting energy levels are found to be densely arranged over certain ranges, forming nearly continuous bands of energies (Figure 2). These energy bands are centred about the atomic energy levels and the width of the band is correlated to the amount of bonding or strength of the nearest-neighbour interactions (i.e. as the size of the system decreases, the nearest-neighbour interaction also decreases with a narrowing of the band widths). A single band is formed by the interaction of the same type of atomic orbitals from each of the neighbouring atoms composed of slightly varying degrees of bonding (i.e. assortment of bonding or anti-bonding characters), hence a multitude of bands can exist representing the different types of orbitals an atom can possess (i.e. s, p, d, f, etc.). These different bands can overlap (as in the case of a metal) or form distinct bands with a bandgap (as in the case of semiconductors and insulators). Bandgaps, no matter how many atoms are aggregated, are “forbidden” intervals of energies for electrons, which can be thought of as energy values where there are no orbitals. Electrons occupy energy levels starting from the lowest energies and filling upwards. In semiconductors, the highest occupied level, at the absolute zero temperature, is known as the
valence band (VB) and the lowest unoccupied level, above the valence band, separated by a bandgap, is known as the conduction band (CB). There are many different models for predicting band structures in crystals (i.e. the nearly free electron approximation, tight-binding model, KKR model, Kronig-Penney model, to name a few) which start from different basis functions and assumptions.60,61,69
The concept of the energy bandgap is crucial to the understanding of semiconductor materials and determines many of the important characteristics, in particular the material’s electronic and optical properties. In bulk semiconductors, the width of the bandgap is a material specific, fixed parameter. However, what are the consequences to the electronic structure of going nanoscale?

When a photon, of equivalent energy to a material bandgap, is absorbed by a semiconductor, an electron is excited from the valence band into the conduction band. This excitation process leaves behind a localized, positively-charged, unoccupied state, otherwise known as a hole. Attractive coulomb forces bind and stabilize the excited electron in the conduction band and the localized hole in the valence band. The formation of a bound, non-dissociated electron-hole pair is known as an exciton. The average separation distance between the electron-hole pair is called the exciton Bohr radius and it is a material dependent property. The strength of the coulomb interaction between the electron-hole pair, termed the exciton binding energy, is influenced by the electric field screening capabilities related to the dielectric constant of the material. Depending on the degree of this coulombic interaction, excitons may be classified into two limiting cases, Frenkel excitons and Wannier-Mott excitons (Figure 3a).58,59,68,70-73

Frenkel excitons are found in materials with small dielectric constants. The coulomb interaction binding the exciton can be considered strong, on the order of 0.1 to 1 eV, with a fairly small Bohr radius, on the same dimensional order as the unit cell. Wannier-Mott excitons are found in materials with generally large dielectric constants, such as in semiconductors. As a result, there is a reduction in the coulomb interaction between the electron and hole due to the screening of the charges. In this case, the binding energy of the exciton is considered relatively weak, on the order of 0.01 eV, thus the Bohr radius tends to be much larger and span beyond the lattice spacing.58,59,68,70-73

Semiconductor crystals with physical dimensions comparable or smaller than the exciton Bohr radius are said to be in the quantum confinement regime. In this regime, electronic excitations of a semiconductor crystal are confined by an infinite potential at the crystal boundaries. The effect of quantum confinement changes the density of the energy levels, as well, a size-dependent (on top of a material specific) bandgap is established. This phenomenon is called the quantum size effect and nanoscale particles that exhibit this behavior are called quantum dots. 74

In a simple analogy, the phenomenon can be described using a “particle-in-a-box” or particle in an infinite square well potential model, where if we take a spherical QD with radius R, the model predicts that the energy gap will be proportional to its size, which is 1/R². Hence, as the dimensions of a QD become
smaller, the bandgap becomes larger, which means higher energy is required to excite an electron across the bandgap and a blue shift in the optical transition (i.e. absorption or emission) wavelength can be observed. The phenomenon also leads to greater spacing between the intraband energy levels (with some of the energy levels at the edges no longer being allowed, causing the band edges to shift, resulting in a
larger bandgap) as the number of atoms decrease, as the size of the crystal reduces. This leads to an intermediate electronic structure where energy levels become discrete, especially near the band edges, in contrast to bulk crystals, while still maintaining a higher density and smaller spacing of energy states compared to atomic systems (Figure 3b). Because the edges of the bands determine the optical and electronic properties of the material and since the energy levels are now discernably separated near the edges, discrete atom-like transitions can be observed in the absorption spectrum of QDs, in contrast to the continuous spectrum seen in bulk semiconductors.74-76

The Brus equation is a popular approximation expression that can be used to relate particle size to the bandgap or lowest electronic transition energy of a spherical semiconductor nanocrystal:

\[
E_g(QD) = E_g(bulk) + \left( \frac{\hbar^2}{8R^2} \left( \frac{1}{m_e^*} + \frac{1}{m_h^*} \right) - \frac{1.8e^2}{4\pi\varepsilon_0 eR} \right) \text{ (Equation 1)}
\]

where \( E_g \) is the bandgap energy of the QD or bulk material, \( R \) is the QD radius, \( m_e^* \) and \( m_h^* \) are the effective masses of the electron and hole, respectively, and \( \varepsilon \) is the relative dielectric constant of the material. The energy increasing middle term in the equation represents a particle-in-a-box quantum localization energy term for the exciton and it has a \( 1/R^2 \) dependence. The energy stabilizing final term represents the electron-hole pair Coulombic attraction or exciton binding energy, which has a size dependence of \( 1/R \). In the limit of a large \( R \), the band gap approaches \( E_g(bulk) \), however for very small nanocrystals, the approach does not work well, resulting from the breakdown of the effective mass approximation.61,68,70,77

The exciton Bohr radius in bulk semiconductors is given by:

\[
a_B = \frac{4\pi\varepsilon_0 \varepsilon \hbar^2}{e^2} \left( \frac{1}{m_e^*} + \frac{1}{m_h^*} \right) \text{ (Equation 2)}
\]

In a more quantitative treatment of evaluating the extent of confinement in a low-dimensional structure, depending on the ratio between the nanocrystal radius (\( R \)) and the bulk exciton radius (\( a_B \)), three different size or confinement regimes can occur. In the strong confinement regime, where \( R < 2a_B \), the Coulomb electron-hole interaction is much smaller than the confinement energies, therefore the electron and hole can be considered as independent particles. In the intermediate confinement regime, where \( R \approx 2a_B \), the electronic structure depends on a complex interplay between quantum confinement and Coulomb interaction. In the weak confinement regime, where \( R > 2a_B \), as electron-hole interactions are significant in comparison with the confinement energies, the electron and hole motions are strongly correlated and the
QD energy spectra are determined by quantization of the motion of the exciton center of mass.\textsuperscript{50,54,61,62,68,70,77}

In the particle-in-a-box approach, if the Coulombic interaction between the electron and hole are included, analytical solutions for the Schrödinger equation cannot be obtained. However, in the strong confinement regime, even though the binding energy becomes stronger with a reduction in the nanocrystal size (since the relative dielectric constant of the material decreases), it is possible to disregard this interaction and apply the model, since the confinement term (scaling with $1/R^2$) overpowers the stabilizing attractive forces (scaling by $1/R$). So, in other words, in this particular size regime, the single particle properties dominate over the electron-hole interaction, imposing a quantum behaviour of the material, causing a size-dependent bandgap, and allowing for a reminiscent atom-like characteristic to emerge in the electronic properties of the strongly confined nanocrystal.

In practical terms, what do these consequences mean and why is it interesting? From a material chemist’s point of view, the very idea of being able to synthetically confine excitons in semiconductors and to have exquisite control over this theoretical confinement allowing for an accurate tuning of the electronic bandgap of a material through plausible experimental procedures, is a wonderful illustration of the marriage between theory and practice, not to mention the many useful and interesting implementations such a versatile building-block presents.
1.2.2 Synthesis

There are countless recipes for producing quantum dots. The two general approaches consist of a “top-down” and a “bottom-up” approach, and these approaches can also be further classified as physical or chemical methods. A “top-down” method involves taking a bulk slab of semiconductor substrate, then whittling it down to quantum dot size in the range of 1 to 10 nm, lithographically or electrochemically (i.e. through a series of plasma or wet chemical etching processes). A “bottom-up” method involves building up to the nanocrystal material “atom-by-atom”. The bottom-up approach comprises of several different physical or chemical methods ranging from molecular beam epitaxy (MBE) to solvent phase synthesis.78

As chemists, however, we are accustomed to making materials from a bottom-up approach and partial to using chemical means to attain this goal. The most successful and popular method in this genre for making CdE (E = S, Se, Te) semiconductor nanocrystals is through a pyrolytic, also known as the organometallic precursor route, brought to attention by Murray, Norris, and Bawendi.43 Large batches of quantum dots may be synthesized through this colloidal synthesis method. Furthermore, due to this scalability and the convenience of benchtop conditions, colloidal synthetic methods seem most promising for commercial applications. There are many variations that exists, however, in this particular method, molecular or ionic precursors are dissolved and brought together to instantaneously react at high temperatures in a coordinating solvent to produce colloidal inorganic semiconductor nanocrystals, overcoated with a layer of organic ligand molecules. Varying the reagent concentration and growth time controls the size of the particles, while the organic capping offers electronic and chemical passivation of dangling surface bonds, preventing uncontrolled growth and agglomeration, as well as allowing for chemical manipulation of the surface for increased solubility or reactivity of the nanoparticle with various environments. This section will detail the synthesis procedures for fabricating CdSe nanocrystals used throughout this work. Also, a discussion on the factors affecting nucleation and growth in the nanocrystal formation will be presented.
1.2.2.1 Organometallic Precursor Route and Size-Selective Precipitation & Purification

In regards to materials used, commercially available technical grade chemicals were purchased from Aldrich. Trioctylphosphine oxide, trioctylphosphine, and 100 mesh selenium powder were used without further purification. High purity dimethylcadmium was purchased from STREM and used as received.

A general procedure\textsuperscript{43} (Figure 4) involves first heating 60 g of metal-coordinating solvent, trioctylphosphine oxide ([CH\(_3\)(CH\(_2\))\(_7\)]\(_3\)PO) (TOPO), in a three-necked Schlenk flask to a relatively high temperature of approximately 300°C under an inert atmosphere. The actual temperature can range between 200 to 350°C depending on the experimental setup and the type of reagents used. In this particular case, dimethylcadmium (Cd(CH\(_3\))\(_2\)) is the source of the organometallic precursor ingredient used, which has a decomposition temperature of around 272°C, thus in order for any chemical reactions to occur, thorough decomposition must initially be achieved well above this temperature. Also, to ensure an inert atmosphere, the Schlenk flask, containing the TOPO, was first degassed at 150°C through repeated steps of vacuum pumping and inert gas (e.g. argon) flushing. Once the TOPO has fully melted into a clear, colourless liquid and reached the target temperature, colloid nucleation is initiated by a quick injection of 10.00 mmol of the organometallic precursor, Cd(CH\(_3\))\(_2\), and 13.35 mmol of the chalcogenide precursor, trioctylphosphine-selenium (TOPSe), into the reaction vessel. Since dimethylcadmium is a pyrophoric material, the precursor mixture was pre-prepared in a glove box, where solutions of Cd(CH\(_3\))\(_2\) diluted with 25 mL of trioctylphosphine (TOP) and selenium dissolved in 25 mL of TOP were thoroughly mixed together, then transferred to a syringe and taken out of the glove box just before immediate injection into the hot coordinating solvent. Then, time is allowed for growth, which can occur at a slightly lowered temperature around 275°C. The size and size distribution can be monitored by withdrawing aliquots of the solution and looking at its UV-Vis absorption spectrum every 5 minutes. The width, or more specifically the half width at half maximum (HWHM), of the first absorption peak, or correspondingly to the full width at half maximum (FWHM) of the emission spectrum, is an indication of the size distribution of the sample. A broad width signifies a broad distribution in the sizes. After a desired size has been reached, further reaction can be ceased by the complete removal of heat. Optimization of injection temperature, monomer concentrations, injection rates, growth temperature, along with other variables, can lead to better control over nanoparticle growth and achieving the target size, as well as better quality QDs with sample size distributions that are relatively monodisperse.
Once the QDs have been synthesized, size-selective precipitation is normally carried out to narrow the size distribution and clean the sample of any excess residues of coordinating solvent and contaminants.\(^4^3\) This is customarily accomplished in a qualitative manner by dissolving the freshly made QDs (in solid TOPO) in a minimal amount of toluene, then adding a small amount of non-solvent, in this case methanol, to mildly precipitate out the QDs. As minute amounts of non-solvent are added to the QD solution, there will come a point in time when the mixture will turn barely cloudy. The cloudiness indicates precipitation. At this point, the mixture is centrifuged just long enough to visually see the formation of a small amount of precipitate. The precipitate will contain a higher concentration of larger sized QDs that would have fallen out of the solution first, due to its heavier weight and increasing instability in the modified solvent and the effects of centrifugal acceleration. The supernatant, which has a tinge of colour, showing the presence of the smaller sized dots still dissolved in the solvent, is discarded (however, it could be kept separately for further size-selection and purification) and the precipitate is collected. This purification process is repeated twice to ensure a tighter narrowing of the size distribution and to maximize purification. However, it must be noted that excessive purification steps can lead to a destruction of the nanoparticle surface, decreasing the likelihood for redissolution of the nanoparticles in toluene. The final precipitate is
dried under nitrogen, then redispersed in toluene. The resulting batch of QDs are capped on the surface by a monolayer of organic ligands or surfactants, TOPO, which also played the role of the coordinating solvent, allowing for physical and chemical stability of the nanoparticles in a variety of organic environments. For prolonged storage of the QDs, it is best to store the sample in the solid TOPO form and purify only prior to usage.

1.2.2.2 Colloidal Synthesis: Nucleation and Growth

Colloidal synthesis of QDs is based on two essential stages of formation, nucleation and growth, which should occur at successive, discrete temporal periods in order for successful preparations of uniform nanocrystal samples to transpire. In the synthesis mentioned previously, there are several key variables which greatly influence the ultimate outcome of the product and these require careful attention when optimizing such procedures. The variables include, but are not exclusive to, temperature, time, and concentration of the reagents.

The concept of nucleation and growth in regards to colloidal aggregation adapted from classical nucleation theory were put forth by the work of La Mer and Dinegar on the preparation of monodisperse colloidal particles via a homogeneous precipitation reaction and their early theoretical model of “burst nucleation” (Figure 5a). Based on their study, it was found that the formation of monodisperse colloidal particles start with a short single burst of nucleation in a homogeneous solution. This is achieved by the quick injection of precursors into the flask of hot coordinating solvent. At this instant, the precursor reagents immediately undergo thermal decomposition releasing a critical concentration of monomers, the constituent species, into the reaction medium to form a supersaturated solution above the metastable limit. In this concentration regime, randomly diffuse monomers combine to form small aggregate embryos, which are preliminary formations of unstable nuclei. The fate of the embryo depends on its overall change in free energy (∆G), which is the sum of two components given by:

\[ \Delta G = -\frac{V}{\Omega} k_B T \ln \beta + S \gamma \]  

\[\text{Equation 3}\]

where \( V \) is the volume of the embryo, \( \Omega \) is the volume of the molecule inside the embryo, \( \beta \) is the ratio of actual concentration to the concentration at equilibrium used to define supersaturation, \( S \) is the surface area of the embryo, and \( \gamma \) is the interfacial free energy between the embryo and the solution. The first term is the volume term favouring the creation of the embryo and represents the energy gain in forming a new
volume. The second term is the surface term favouring the dissolution of the embryo and represents the energy cost in creating a new surface for the embryo. The competition between the two energy terms will ultimately determine the possible nucleation of an embryo of a particular dimension, as illustrated in the plot of the free energy for nucleation versus embryo size (Figure 5b)\textsuperscript{87}. The critical size (r*), denoted by the particle radius (r), is the minimum size a particle must attain to become stable. The corresponding critical activation energy for nucleation (ΔG*) at r* is the maximum value of ΔG the embryo must overcome in order to become a stable nucleus. Activation energy for nucleation decreases with increasing supersaturation and temperature, as well as with decreasing interfacial free energy. Embryos that have overcome this barrier (r ≥ r*) will attempt to further decrease their free energy by spontaneous growth transforming into macroscopic particles. Alternatively, embryos that have not attained the critical size (r < r*) will spontaneously dissolve and re-release the monomers back into the solution. Returning back to the time of injection, subsequent to reaching this critical supersaturation point, the system is partially relieved by the sudden appearance of nuclei which effectively reduces the monomer concentration below the nucleation threshold and initiates the growth stage where growth of the nanocrystal proceeds by the addition of monomers from the solution to the existing nuclei. The driving force for nucleation and growth is based on the chemical potential gradient of a molecule going from a saturated to a supersaturated solution. It is important to note that in this model, nucleation and growth stages must take place separately in time in order for monodisperse colloids to result. Since the rate of nucleation is sensitive to concentration, or more specifically to supersaturation, by controlling the initial amount of reagents injected into the flask (and/or by manipulating the temperature of the system), the rapid drop in monomer concentration observed after nucleation can be utilized to favourably restrict the nucleation stage in time, so that no new nuclei are formed after the initial burst. This will prevent any further repetitive nucleation processes to occur which would adversely overlap with the growth stage resulting in a non-uniform growth of particles, giving rise to a polydisperse colloidal dispersion.\textsuperscript{87-92}

The growth stage can be divided into two parts, “growth” and “ripening” (more specifically Ostwald ripening) phases, sometimes referred to as the “focusing” and “defocusing” regimes. Continuing on with the theory of classic crystal growth, similar to concepts applied to embryo nucleation, the self-focusing of particle size distributions depends, once again, on a critical radius (r*). Depending on the concentrations of monomers present in the system, a particular critical size will exist. The ratio of the average particle radius (\textlangle r\rangle) to this critical radius will determine whether a particle size distribution (Δr) will narrow or broaden (Figure 5d)\textsuperscript{93}.

Formation of CdSe Nanocrystals

a) Concentration of precursors/monomers and their evolution through nucleation and growth stages are shown.

b) The critical activation free energy ($\Delta G^*$) must be reached for creating a nucleus with a critical radius ($r^*$).

c) The addition of Cd and Se atoms to the nanocrystal and the association of TOPO and TOP are illustrated.

d) Depending on the average radius of the particle with respect to the critical sizes (and monomer concentration), corresponding growth rate will result and a focusing or defocusing of the size distribution can be achieved (modified from ref. 93).
The size focusing is optimal when the average particle size is always slightly larger than the critical size and conversely, distribution will broaden when the critical size becomes larger than the average size, as stated in the above equations. The first part of the growth stage is fairly rapid. At this stage, the monomer concentration has fallen below the critical concentration for nucleation, and is now within the “normal” metastable supersaturation limit, which means that monomers are still present in relatively high concentrations providing ample fuel for growth of the stable nuclei to occur. Growth proceeds, in a kinetically controlled manner, through the diffusion of monomers towards the surface, then their subsequent reaction onto the surface of the nuclei (Figure 5c). In this phase, smaller particles grow faster than larger ones, since larger crystals require more atoms to grow than small crystals. When monomer concentrations are high, the critical size is relatively small (where \( r \geq 2r^* \)) favouring growth of nearly all of the particles within the system; and since smaller particles are kinetically favoured to grow more easily (i.e. at a faster rate) than larger ones, this results in a focusing or narrowing of the size distribution down to one that is nearly monodisperse. The second part of the growth stage, Ostwald ripening, only occurs when nanocrystals are allowed to grow for longer periods of time until depletion of available monomers reach an eventual saturation point at which the nanocrystals stop growing. In this phase, growth kinetics are relaxed as the system heads toward equilibrium and the thermodynamically driven ripening process take effect. Ostwald ripening is based on the surface energy of particles and their solubility differences. Since small particles have larger surface area to volume ratio in comparison with large particles, they are said to have higher surface free energies and have higher solubilities making them fairly unstable in solution; in other words, larger particles are more energetically favoured than smaller particles. This is because molecules on the surface have a lower number of attractive, like-neighbouring interactions, since they are partially in contact with another medium (i.e. the have greater surface tension at the particle-solution interface). This makes them energetically less stable than the well-ordered, well-packed interior molecules. As the system tries to lower its overall energy, smaller particles sacrificially dissolve, increasing the monomer concentration once again to the supersaturation threshold, in favour of growth of larger particles. When monomer concentrations are depleted, the critical size shifts to a larger radius, and any particles below this critical size will have negative growth rates, while above \( r^* \) will have positive growth rates. The offset in growth rates will lead to an asymmetric, defocused size distribution. Sustained growth in this stage eventually results in fewer, larger, more energetically favoured particles left behind in
the system. To a certain degree, monodisperse dispersions can result from an initially polydisperse system through this ripening channel, however, it ultimately results in a rather broad particle size distribution, requiring additional post-preparative size fractionation. In general, Ostwald ripening provides a simple and logical way to achieve larger sized particles through the control of growth duration. This latter Ostwald ripening phase, largely developed by Lifshitz and co-workers, was added to the burst nucleation mechanism, since the original model was inadequate for rationalizing the discrepancies found in experimental results for the formation of larger particles greater than 100 nm. Further discussion on growth can be extended to include diffusion and reaction limited growth modes, however the general theory presented is adequate for a basic understanding of colloidal nanocrystal formation.

The above discussion has primarily been focused on the recurring theme of supersaturation and monomer concentrations, and their evolution through the nucleation and growth stages. However, besides reagent concentration and reaction time, formation of nanocrystals, with respect to achieving a target size (sometimes specific morphology) and distribution, can also be altered through another convenient experimental variable: temperature (and less practically through pressure). Careful regulation of temperature throughout the different steps in nucleation and growth is critical for developing optimal conditions for nanocrystal synthesis. Temperature affects the rate of reactions, it can aid in the diffusion and structural organization of the constituent species, as well its modulation can promote or limit solubility conditions for nucleation and growth. In many cases, temperature plays contrasting roles that can lead to inconsistent unpredictable results, and thus must be premeditated and suitably balanced for a desired outcome. For example, it helps to keep in mind that the injection temperature at the beginning of the synthesis should be high enough to induce an initial chemical reaction (i.e. thermal decomposition of precursors) for nucleation to occur, however a strategically reduced temperature immediately following, can avoid additional unwanted nucleation events. Relatively high temperatures must be established in order to allow for rearrangement and annealing of atoms, while still being low enough to allow for surface attachment and reaction to promote crystal growth. Also, studies have shown that lower nucleation temperatures can yield larger size nuclei. The reasoning behind this is not so intuitive, but this is because lower nucleation temperatures, lowers solubility of monomers, therefore supersaturation can be reached or supported at lower monomer concentrations. Thus, a combination of lower nucleation temperature and lower monomer concentration can result in a prolonged nucleation stage, prone to slower nucleation rates, leading to a small amount of nuclei formation (i.e. favourable for nuclei growth conditions), which can yield larger sized nuclei. In contrast, higher growth temperatures can generate larger particles due to the enhancement in the rate of monomer addition to the existing particles. As well, Ostwald ripening occurs more readily at higher temperatures. Based on the Gibbs-Thomson effect, where small crystals in a
solution are observed to melt more readily at lower temperatures than larger crystals in solutions, a higher temperature during Ostwald ripening will boost the dissolution of smaller particles which will consequently boost the growth of the larger particles. Seemingly simple, yet complex tips for nanocrystal recipes are found in many influential bodies of works, but with such complex interplay between these variables, sometimes the synthesis is still a bit of a black art. It is easier in some cases to fine tune procedures based on numerous experimental observations then work backwards to rationalize these findings. From a logical point of view, anything that can affect supersaturation (e.g. dilution with solvent, change in pH, addition and identity of the surfactant/chelating agent used, etc.) can be used to control nucleation and growth, and ultimately the size, shape, and distribution, experimentally. There are endless bodies of work devoted to nanocrystal synthesis with various emphasis on size and shape control, obtaining ideal size distributions, various specialized one-step or “green” synthesis procedures, and so forth. A discussion of all these factors would be a rather tall task, thus ends the rudimentary discussion on nucleation and growth.
1.2.3 Photophysical and Structural Characterization

Once the nanocrystals have been synthesized, various characterization methods can be employed to investigate the structure/property relationships of the nanocrystal. For a relatively efficient and effective assessment of their basic photophysical and structural properties, UV-Vis absorption and fluorescence spectroscopy and transmission electron microscopy (TEM) are utilized. The data obtained, along with the qualitative and quantitative information which can be extracted from the results, will be briefly discussed in this subsection.

1.2.3.1 UV-Vis Absorption and Fluorescence Spectroscopy

For group II-VI semiconductors, their bandgap energy falls in the UV-visible range (i.e. CdSe bulk bandgap is 1.74 eV or 712 nm), and covers a good portion of the solar spectrum, making them useful for many applications. Since it is possible to change the bandgap of the material by just changing the physical dimensions of the quantum dot, samples with different sizes can be custom made, which are capable of emitting a brilliant palette of rainbow colours (Figure 6a). This property makes characterization through the techniques of UV-Vis absorption and fluorescence spectroscopy very useful and interesting. Information regarding the size, bandgap energy, quantum yield, shape, quality (e.g. size dispersion and the presence of surface defects), for instance, can be obtained through their analysis.

A typical absorption and emission spectra of colloidal, spherical, CdSe nanocrystals are shown in Figure 6a. Let us start with the quantum dot absorption spectrum. The peaks seen in the absorption spectrum correspond to electronic transitions between the electron and hole quantized states. As previously discussed, quantum confinement leads to a collapse of the continuous energy bands of bulk semiconductors. This results in discrete energy states described by distinct quantum numbers (n,l,m), similar to electrons in orbitals of an atom, representing the various symmetry, orbital angular momentum, and its projection. Only certain transitions are possible between the various energy levels, due to selection rules, and these allowed transitions give rise to the distinct absorption features seen in the spectrum (Figure 6b,c). Depending on the quality of the nanocrystal samples, up to eight absorption features can be resolved in a single spectrum. The actual electronic structure and transitions are much more complex, and consideration of electronic interactions, such as spin-orbit coupling and crystal field splitting, and electron-
hole Coulomb and exchange interactions will provide a more complex picture of the energy levels (i.e. fine electronic structure).\(^{49}\)

An important and very useful feature in the quantum dot absorption spectrum is the peak for the first excited state or the first lowest energy absorption peak, also referred to as the first exciton peak. This is the minimum energy, hence maximum wavelength, required to excite an electron across the bandgap from the highest valence state to its lowest conduction state. This peak wavelength or energy is a strongly size-dependent parameter.\(^{101}\) Empirical relationships have been determined relating the diameter of nanocrystals (for CdSe, CdTe, CdS) to the wavelength of their first absorption peak by plotting and fitting sizing curves (size versus first absorption peak position) obtained from the absorption data of particles with known diameters. Through careful measurements of this first absorption peak and using the empirical relationship,\(^{102}\)

\[
D_{\text{CdSe}} = \left(1.6122 \times 10^{-9}\right)\lambda^4 - \left(2.6575 \times 10^{-6}\right)\lambda^3 + \left(1.6242 \times 10^{-3}\right)\lambda^2 - (0.4277)\lambda + (41.57)
\]

Equation 5

(where D is the diameter in nm and \(\lambda\) is the wavelength of the first excitonic peak in nm of the nanocrystal sample), a fairly accurate estimation of the nanocrystal size can be determined. The ease and accuracy of extracting the size through this peak position is uncontested compared to other direct methods for obtaining the QD size. It can be seen in Figure 6a (Size evolving absorption and emission spectra of CdSe nanocrystal) that quantum confinement clearly shifts the transitions to higher energies with decreasing size.

In the same manner as obtaining the nanocrystal size, it is possible to determine the concentration of a nanocrystal sample by simply using the absorbance of the first absorption peak and the Beer-Lambert law. By measuring the absorbance of nanocrystal solutions of known concentrations and fitting this data, an empirical relationship between the extinction coefficient per mole (or molar absorptivity), \(\varepsilon\), and the nanocrystal diameter can be found (Equation 6).\(^{102}\) Using the size-dependent extinction coefficient determined, the concentration of the nanocrystal sample can then be simply calculated using the Beer-Lambert law. Small QDs have \(\varepsilon\) values near 200,000 M\(^{-1}\) cm\(^{-1}\), similar to that of rhodamine 6G (R6G), while large QDs can have values as large as 2 \(\times\) 10\(^6\) M\(^{-1}\) cm\(^{-1}\), tenfold larger than R6G.

\[
\varepsilon_{\text{CdSe}} = 5857 \times \left(D_{\text{CdSe}}\right)^{2.65}
\]

Equation 6

Apart from the size and concentration of the quantum dots, qualitative information such as size distribution and shape of nanoparticles can be deduced from the absorption spectra. The absorption
Figure 6

Photophysical Properties of Nanocrystals

(a) Typical absorption and emission spectra for CdSe QDs in toluene. The different (coloured) sets of spectra correspond to different nanocrystal diameters.

(b) Absorption spectrum of CdSe QD. Marked positions correspond to well-resolved transitions.

(c) Quantum confinement leads to mixing of different valence subbands for CdSe QDs (leads to complex hole states - subscripts denote total hole angular momentum). The electron energy levels (CB) are more widely separated since the electron mass is smaller than the hole mass (m_e/m_h = 6 in CdSe). Arrows indicate allowed transitions (modified from ref. 49).

(d) Superposition of individual absorption transitions for QDs with varying radii.

(e) Illustration of the various fate of an exciton: (1) band-edge, (2) Auger, and (3) defect/surface recombinations.
spectrum for a collection of quantum dots with an arbitrary particle size distribution can be considered superpositions of individual absorption spectrum for a QD with a given radius (Figure 6d). The line shape of the individual QD absorption (i.e. for each of the transitions) is also assumed to be Gaussian to take into consideration the inhomogeneous width from the individual differences each nanoparticle can experiences in its environment (e.g. due to inconsistencies in shape or surface morphology, etc.). This will lead to an overall particle size distribution which is also assumed to be a Gaussian distribution. If the size distribution of a QD sample is narrow, the various absorption features seen in a typical QD absorption spectrum will be better resolved (i.e. sharper peaks), since there is less convolution due to a decrease in contribution from the differently sized QDs. In particular, the first exciton peak will look very prominent. Fitting this peak to a Gaussian curve and obtaining the full width at half-maxium (FWHM) value (easily obtained from the fluorescence peak without fitting), or through a rough estimation (with no fitting) obtaining the half width at half-maximum (HWHM) value, can be used as a convenient index for size distribution.103,104

It is also possible to qualitatively distinguish between nanocrystal shapes using the absorption spectrum. Although this thesis focuses solely on spherical quantum dots, for which the particle is quantum confined in every dimension, other shapes may only have certain dimensions that are in the quantum regime. For example, in nanorods or tetrapods, quantum effects are determined by the diameter of the rod or the arms of the tetrapod, and this is manifested as sharper absorption features compared to those seen in quantum dots. The absorption spectrum of different shapes can have slightly distinctive features, hence distinct looking spectra, representing the various types of quantum confinement that is inflicted in that particular nanocrystal. Additional information can be extracted from a meticulous analysis of the nanocrystal absorption spectrum (e.g. certain crystal lattice information, surface defects, etc.), however the aforementioned aspects cover the basic information commonly obtained through this routine measurement.105-110

Moving on to the quantum dot fluorescence spectrum, photoluminescence (PL) in nanocrystals arise from the radiative interband recombination of electron-hole pairs. After absorption, the electron in the excited state can relax back to its ground state, recombining with the hole, via two main channels, radiative decay and non-radiative decay (Figure 6e). Depending on the energy of the photon absorbed, an electron is excited to energy levels over the bandgap (i.e. higher electronic and/or vibrational states). Within a band (i.e. intraband), the electron, at this point, can quickly dissipate its excess energy through a rapid, non-radiative, relaxation pathway via lattice vibration (known as phonons), leaving the electron in the lowest vibrational level of the excited state. Across the bandgap (i.e. transitions between bands, interband),
several pathways are possible resulting in the relaxation of the electron back to the ground state for exciton recombination. These pathways include, radiative and non-radiative bandedge exciton recombination, radiative and non-radiative surface-assisted exciton recombination, and non-linear and non-radiative exciton-exciton annihilation (Auger recombination). Among these various transitions, the size-tunable fluorescence emission of QDs originate from the radiative exciton recombination at the CB and VB bandedges.\textsuperscript{111-118}

A typical CdSe QD emission spectrum is a Gaussian curve. As long as the excitation energy is greater than the absorption onset (or bandgap energy), the emission spectrum remains unchanged, irrespective of the excitation wavelength used. This follows Kasha's rule that photon emission in molecular systems occurs mainly from the lowest excited electronic and vibrational level ($S_1$) to some vibrational level of the ground state ($S_0$). Furthermore, since some of the excitation energy is lost through thermalization or non-radiative decay, the emission energy will be less than that of the absorption. This difference in energy is called the Stokes shift and it is the difference (or red shift) between the positions of the first absorption peak and the fluorescence peak of the nanocrystal. Besides this effect, in nanocrystals, further Stokes shifts can occur due to defects in the material, such as vacancies, impurities, or adsorbates at the surface. These defects cause the formation of "trap" or "surface" states\textsuperscript{103} for which an excited electron can fall, or in the case of a hole can float, and can radiatively recombine resulting in a photoluminescence spectrum that is even further Stokes shifted from the absorption, depending on the relative energies of the trap states versus the VB and CB bandedges.

Like the absorption spectrum, much of the same type of information discussed can also be extracted from the QD photoluminescence spectrum. Analogous to the first absorption peak, the emission peak can give details regarding the size, bandgap energy, extinction coefficient, and other size-dependent information. However, such information are normally taken from the absorption spectra, since fluorescence is much more sensitive to external factors and surface properties of the QD. As mentioned previously, surface defects and lattice imperfections in nanocrystals appear in the form of unsatisfied valencies, and like surface charges, these valencies present a sink for the charge carriers (electrons and holes), resulting in aberrant recombinations that can affect the intensity, peak shift, and even the line shape of the steady-state fluorescence spectra. The presence of surface ligands and adequate coverage over the nanoparticle surface can deter some of the effects of the surface defects (but not lattice imperfections), preserving the “original” optical features of the QD. External factors, such as the solvent polarity or local environment, can also have important consequences on the optical properties of nanocrystals. Solvent polarity can affect the dispersion of nanoparticles. When nanoparticles are well-dispersed in its solvent environment, the chances
for non-radiative decay events, which can occur when particles are in close proximity to each other, reduces. This will be reflected in the intensity of the PL peak.\textsuperscript{103,104}

Apart from the abstruse details that can be inferred from the PL spectrum, more concrete information regarding the size distribution and quantum yield of the sample can be determined. Size distribution information can be obtained through the FWHM value of the emission peak as previously described for the absorption spectrum. Since the emission peak is obtained in entirety, unlike the first absorption peak, where the peak position and the peak itself is sometimes difficult to distinguish from the rest of the spectrum, especially when the sample quality is not optimal, the fluorescence FWHM value is a more clear and convenient, as well as accurate, not to mention preferred, method of obtaining the size distribution. As a reference, the FWHM value is ideal if it is less than 30. Quantum yield (QY or \( \Phi \)) is a measure of the ratio of the number of photons emitted relative to the number of photons absorbed, with materials having the largest QY (e.g. rhodamine dyes), approaching unity, displaying the brightest, most efficient, emissions. The most reliable method for obtaining the QY of a sample is by using the comparative method\textsuperscript{119}. This method involves measuring the fluorescence and absorption spectra of a series of dilutions for the sample of interest and a carefully selected, well-characterized (i.e. with a known QY value) standard sample. After careful measurements, a plot of the integrated fluorescence intensity versus absorbance is made and the gradient of the resulting data is extracted. Absolute QY values are then calculated according to the following equation:

\[
\Phi_X = \Phi_{ST} \left( \frac{\text{gradient}_X}{\text{gradient}_{ST}} \right) \left( \frac{\eta_X^2}{\eta_{ST}^2} \right)
\]

where subscripts X and ST represent the test sample and the standard sample, and \( \eta \) is the refractive index of solvent used to measure the QY of the test and standard sample. CdSe QDs typically have QYs ranging from 1-10\%. There are many factors that can reduce the QY of a sample (i.e. reduce intensity of fluorescence). The main ones have been already discussed above. One way to eliminate trap states and increase QY drastically is to coat the QD with a shell of a higher bandgap material (e.g. CdSe(core)–ZnS(shell)). This will lead to light emission that is closer to the absorption energy and have QYs greater 50\%.\textsuperscript{120-122}
1.2.3.2 Transmission Electron Microscopy

If seeing is believing, transmission electron microscopy (TEM) and associated microscopy techniques are very powerful tools for visually characterizing nanocrystals and their assemblies. TEM is a unique technique allowing for direct, real-space imaging of nanocrystals, with spatial resolutions ranging from broad field views of NC ensembles down to finely focused atomic resolutions of 1 nm or better. Low-resolution TEM can be used to image nanocrystal ensembles for average size and shape information. High-resolution TEM (HRTEM) can be used to image individual nanocrystals and obtain a close-up of their internal structures (i.e. lattice fringes) for shape and crystallographic information, such as its phase and crystal axes. Many other forms of electron microscopy and extended analysis of TEM exists, such as scanning electron microscopy (SEM), high-resolution z-contrast imaging (STEM/HAADF/ZC), electron energy loss spectroscopy (EELS) and the complementary energy-dispersive X-ray spectroscopy (EDX, EDS, XEDS), et cetera, all having characterization advantages of their own.

For example, surveys of a nanocrystal sample, via a series of images, can be used to develop a statistical model of the size and shape, and description of the internal structure, of the NCs present in a particular sample. To obtain accurate measurements of size, the NC lattice planes or atom columns are counted for a quantitative, pedagogical determination of the size and calibration of the image magnification. Such rigorous analysis is not normally performed on TEM images, however this is to note that a formal internal standard exists for obtaining an initial set of NC structural parameters through this particular imaging technique. The rough statistics and limited observations are customarily contrasted with other techniques for more accurate and unified information. In general, TEM is a useful qualitative tool for obtaining NC sample quality information (i.e. size dispersion, morphology, purity) and it is an essential tool for microstructural investigations of NC assembly formations.
1.3 Water-Soluble CdSe QD Clusters

In this section, the preparation of self-assembled, water-soluble QD clusters (QD-P), composed of CdSe nanocrystals bound together by pepsin, a digestive enzyme found in mammals, is presented. Also, a structural model for the system is suggested using well known characterization techniques, comprised of TEM, dynamic light scattering (DLS), small angle X-ray scattering (SAXS), along with absorption and emission spectra.

1.3.1 Cluster Formation

For materials, commercially available technical grade chemicals were purchased from Aldrich. Porcine gastric mucosa pepsin powder and 8 kDa polyethylene glycol were used without further purification. Also, distilled, deionized water was employed in the preparation of the clusters.

For the preparation of water-soluble CdSe QD clusters, previously synthesized QDs, of any desired size, are first primed for use through size-selective precipitation using a solvent/non-solvent combination, in this case toluene/methanol, as mentioned in section 1.2.2.1. The resulting purified QDs dispersed in toluene are then precipitated out of solution using methanol, and the precipitant is collected through centrifugation with the supernatant discarded. Next, a pre-prepared 1 μM aqueous porcine pepsin solution is added to the precipitated QDs. Afterwards, the solid clumps of QDs in the pepsin solution are dispersed using sonication at 40 kHz, for a minimum of one hour. Then, the solution is centrifuged for another 80 min. at 13,000 rpm, and redispersed via sonication. The TOPO capped QDs are originally insoluble in water, however through the addition of a protein, pepsin, the nanocrystals are aided in their solubility in the polar media, as a result of QD cluster formation with the protein (Figure 7). The repeated steps of sonication and centrifugation facilitate the dissociation of any aggregated QDs for protein access, and allows for better dispersion in water. Finally, a 10 w/v % of 8 kDa polyethylene glycol (PEG), which is a common water-soluble, biocompatible, surface stabilizing polymer, is added to the solution. PEG is commonly used as an additive in protein formulations because of their ability to slow protein precipitation from the liquid state. They are known to impart water solubility to proteins by shielding hydrophobic domains exposed at the surface, as well, they help slow down formation of aggregates by inhibiting self-association, or protein-protein, contacts through a molecular spacer effect. Thus, in a similar manner, PEG addition was utilized, in this particular situation, to prevent further aggregation and stabilize the QD-
P clusters in water for a longer period of time. The structural and optical characterization of the prepared clusters will be presented in the following.

**Figure 7**

Water-Soluble CdSe QD-Pepsin Clusters

CdSe QD-pepsin clusters are prepared by dispersing precipitated QDs in a 1µM aqueous porcine pepsin solution, using sonication at 40 kHz for a minimum of one hour. The solution is then centrifuged for another 80 min. at 13,000 rpm and redispersed through sonication. The newly formed QD-pepsin clusters, through the addition of a protein, becomes soluble in polar media.
1.3.2 Cluster Characterization

For characterization of water-soluble QD-P clusters, scanning transmission electron microscopy (STEM) images were obtained in scanning (SE), transmission (TE), and high-resolution z-contrast/dark field (ZC) modes using a Hitachi HD-2000 instrument. Small angle X-ray scattering (SAXS) measurements were carried out on a Bruker Nanostar powder diffractometer with GADDS. The size and size distribution of the QD-P clusters were analyzed using dynamic light scattering (DLS), the DynaPro MS/X PCS. Absorption spectra were obtained on a CARY 100 BIO UV-Vis spectrophotometer. Photoluminescence spectra were measured using a Spex Fluorolog 3-22 spectrofluorometer fitted with center slits in both the excitation and emission double monochromators. The reported spectra were all collected at room temperature.

High resolution SE, TE, and ZC microscopy images of the QD-P clusters are shown in Figure 8a. From these images, it can be seen that the CdSe QDs have self-assembled to form highly symmetrical, spherical masses. In the ZC mode, the distribution of QD-P clusters of a typical week old sample is shown. The concentration of the QD-P clusters in a given sample is relatively sparse with ranging cluster sizes. Some of the clusters are found individually, while others are found in aggregates, which is a common sign of aging. The clusters eventually aggregate into giant masses that become unstable and settle out of the solution. In the SE mode (Figure 8a, left), a rather bumpy surface texture of the cluster can be seen. This bumpy texture can be attributed to the individual QDs forming the cluster or it could possibly be the texture of the pepsin itself surrounding the QDs, forming the outer layer of the cluster. In the TE mode (Figure 8a, right), individual QDs inside the cluster can be roughly differentiated. It would be difficult to precisely identify a discrete QD due to the overlap of different QD layers within the dense cluster. Since the size of the QD used (approximately 3 to 4 nm in diameter) is too small for sectioning with any currently known instruments, no other methods exist for imaging a single layer of QDs within the cluster.

DLS measurements provide quantitative size and size distribution information of a sample. DLS measurements of the clusters show that cluster sizes within a sample are relatively monodisperse, however average sizes vary from sample to sample, ranging from approximately 100-400 nm in diameter. Samples with and without PEG were also measured for comparison. It was found that samples containing PEG seemed to have larger diameters due to its surface associations with the cluster. Varying the concentration ratio of QDs to pepsin solution in the synthesis seemed to have no apparent effect on the cluster size. Neither did varying the length of time for sonication and centrifugation, nor temperature.
Figure 8

(a) Scanning Transmission Electron Micrographs of QD-Pepsin Clusters

Dark field (ZC) mode shows spatial distribution of clusters, scanning electron (SE) shows surface texture and transmission electrode (TE) shows the QDs inside the cluster.

(b) Absorption and Emission Spectra of CdSe QD (in toluene) and QD-P clusters (in water)

Debye scatter due to increased particle size is seen in the absorption of QD-P clusters. The scatter corrected spectrum is shown on the bottom. Otherwise, the absorption and PL peak positions and features are similar in both, QD and QD-P.
SAXS measurements provide structural, organizational information (i.e. lamella spacing, packing order of molecules, 3D superstructures, etc.) regarding a sample. Unlike wide angle X-ray scattering (WAXS), which gives information regarding crystal structures, SAXS works for amorphous materials that illustrate any degree of order (i.e. shows some periodic pattern). SAXS measurements on the QD-P clusters show a single diffraction pattern at around 35 to 40 Å. This suggests that there is some residual order in the packing of QDs within the QD-P cluster. The peak position does not seem to correspond to any structural spacing, but rather the actual size, or more specifically the centre-to-centre separation distance, of the QDs that form the clusters. This implies that there may be a close-packing assembly of the QDs. Then with the given information (the packing structure, the size of the QDs used and the size of clusters formed), the number of QDs per cluster can be deduced. It is estimated that ~40,000 to 300,000 QDs (3 to 4 nm in diameter) are contained in a typical cluster (with a diameter of 100 to 400 nm).

Optical properties remain similar in the transition from QD to cluster. Figure 8b shows the absorption and emission spectra of the original QDs and the QD-P clusters. There are slight shifts in peak positions owing to the different environments of the QDs in the original versus the clustered samples. In the original sample, QDs have a monolayer of TOPO and are dispersed in toluene, while the QDs in the clustered sample are in water and have an additional interaction with pepsin. The Stokes shifts of the QDs and clusters are 103.7 meV and 85.5 meV, respectively. Furthermore, due to the dramatic size increase upon cluster formation, the QD-P particles now have sizes that are comparable to the wavelength of visible light. As a result, Debye scattering becomes significant obscuring the absorption features of the cluster. Thus, a simple correction for the scatter needs to be made using a quadratic fit, which is subtracted from the baseline of the cluster absorption (Figure 8b, middle).

The quantum yields of the CdSe QDs used to make the various clusters were 1.5% (QD-3nm) and 3.1% (QD-4nm) in toluene. The QD-P clusters, made from the same samples, had a QY of 1.2% and 0.5% in water, respectively. The QD-P cluster samples used in this investigation were made from core CdSe QDs. These core QDs, although appearing quite photoluminescent, have fairly low QYs. For comparison, high quality core-shell CdSe-ZnS QDs (QY of 78% in toluene) with thiol-derived surface ligands, supplied by Evident Technologies, were also used to reproduce these QD-P clusters to see if there were any limitations or factors affecting the optical quality of these clusters, besides the quality of the QDs themselves. The QY of the QD-P clusters made from core-shell QDs was 62% in water. It seems as though the clusters in most cases retain the same degree of QY as the dots that form them. The core-shell QDs were originally passivated with amine ligands. This demonstrates that it is not essential to have TOPO-passivated QDs to enable the self-assembly process.
1.3.3 Role of Pepsin

There are many examples of biomaterials research involving protein-nanomaterial composites where the utilization of particular proteins lends many of their unique properties to the hybrid materials that they form.\textsuperscript{127-133} Proteins can assist in the self-assembly process of nanomaterials, impart biorecognition properties, catalyze useful electrochemical or cleavage reactions, allow materials to become biocompatible and water-soluble, and so forth. The precise role of the pepsin in this system is unclear. However, it is the pepsin that promotes the self-assembly of the QDs for cluster formation and ensures compatibility of the clusters with water. Hence, some contextual information on pepsin and the basics of proteins, and how proteins interact with inorganic materials, may shed some light in understanding its important function in water-soluble CdSe QD-P clusters, and the development of protein-nanomaterial composites.

So to begin with, pepsin is an aspartic protease, a protein-digesting (proteolytic) enzyme, found in the stomach of mammals. Enzymes are proteins that catalyze biological reactions, in this case, protein degradation. Proteins are composed of one or more polypeptides, where polypeptides are basically short polymers of amino acid monomers linked together by peptide bonds, or in the language of a chemist, amide bonds (Figure 9). Amino acids are the primary building blocks of proteins, which contain an amino group (-NH\textsubscript{2}), a carboxyl group (-COOH), and a side-chain (-R) that is specific to each amino acid. There are a variety of amino acids of which 21 are naturally found in proteins and are called proteinogenic or standard amino acids. The chemical composition of the distinctive R groups is responsible for the important characteristics of amino acids such as chemical reactivity, ionic charge, and hydrophobicity. The amino acids are usually classified by properties of these side-chains and are divided into four categories: weak acid (negatively charged), weak base (positively charged), hydrophilic (polar, uncharged), hydrophobic (non-polar, uncharged). The distinct function and properties of proteins originate from the unique structural organization and chemical composition of the amino acids. Each protein, or more specifically, the polypeptide chain, typically folds into a configuration (either water-soluble, globular or inert, fibrous forms) that is the most stable for its particular chemical structure and environment. Proteins must maintain this conformation in order to maintain their function. Four levels of organization are used to describe this overall conformation: primary (amino acid sequence), secondary (regular repeating structures formed by amino acids, stabilized by hydrogen-bonds – such as \(\alpha\)-helices, and \(\beta\)-sheets and -turns), tertiary (spatial relationship of secondary structures to one another stabilized by non-local interactions), and quaternary (structural organization of a protein containing more than one polypeptide chain) elements.\textsuperscript{134}
**Hierarchy of Protein Structure**

There are four distinct levels of protein structure. The primary structure has a polarity and the ends are labelled as N- (amino, positive charge) or C- (carboxylate, negative charge) terminus. Secondary structures consist of α-helices and β-sheets. The overall twists and turns (α-helix & β-sheets) of the protein are known as the tertiary structures (one polypeptide chain). Some proteins are composed of more than one polypeptide chain, which interact in specific ways to give the quaternary structure.
The aspartic proteases, a family of proteolytic enzymes, are composed of 323 to 340 amino acid residues with molecular weights around 35,000 amu (porcine pepsin consists of 327 amino acid residues with a mass of approximately 34.6 kDa or kilo-amu). Aspartic protease polypeptides consist of two domains that fold to produce a tertiary structure of two similar lobes. Each lobe consists of two $\beta$-sheets and two short $\alpha$-helices connected by an antiparallel $\beta$-sheet (Figure 10). The bridging site of the two domains forms a deep cleft, large enough to accommodate about seven amino acid residues, and the bottom of which contains the active site, where two asparatic acid residues reside. In the case of pepsin, these acid residues are responsible for fixation and degradation of proteins via hydrolysis (breaking of bonds through the addition of water) of the peptide bonds that link amino acids together in a polypeptide chain forming the protein. Pepsin displays a variety of substrate specificities, but normally it has a preference for peptides with hydrophobic, aromatic or carboxylic, amino acid residues. In most cases, the enzyme-substrate interactions are non-covalent, which include hydrogen bonding, ionic interactions, as well as hydrophobic interactions.134,135

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<thead>
<tr>
<th>Amino Acid</th>
<th>Residue per Molecule</th>
<th>Category</th>
<th>Residue Location</th>
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<tbody>
<tr>
<td>Lysine</td>
<td>1</td>
<td>Polar Basic</td>
<td>320 (in C-terminus 291-327)</td>
</tr>
<tr>
<td>Histidine</td>
<td>1</td>
<td>Polar Basic</td>
<td>53</td>
</tr>
<tr>
<td>Arginine</td>
<td>2</td>
<td>Polar Basic</td>
<td>308 &amp; 316 (in C-terminus 291-327, 316 near active site)</td>
</tr>
<tr>
<td>Aspartic Acid</td>
<td>30</td>
<td>Polar Acidic</td>
<td>(32, 215 near active site)</td>
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<td>Half-cystine</td>
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<tr>
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</table>

**Table 1**  Amino acid composition of porcine pepsin based on amino acid sequence.

- 4 polar basic residues
- 44 polar acidic residues
- 151 polar residues
- 128 non-polar residues
- (199 total polar residues)
Porcine pepsin has 44 polar acidic residues present throughout the entire chain, 4 polar basic residues, 151 polar residues (for an overall 199 total polar residues), and 128 non-polar residues (Figure 10 & Table 1). Dimensions of porcine pepsin, measured from reported structures of the Protein Data Bank (5PEP.pdb file), were found to be approximately 6.1 nm × 4.4 nm × 2.5 nm (length × width × thickness), and the opening of the active site measuring 1.9 nm with a depth of 2.8 nm. For the protein to be active, one of the two aspartate residues in the active site has to be protonated, and the other deprotonated. Thus, optimal activity occurs between pH 2 and 4, however it becomes inactive starting at pH 6.5, and above pH 7, pepsin becomes irreversibly inactive and found in a denatured state.

In the formation of water-soluble CdSe QD clusters, preparations take place in a reasonably pH neutral environment, as well, procedures entail repeated steps of prolonged sonication of the pepsin solution. Extensive sonication is known to disrupt the delicate balance of non-covalent interactions that regulate the functional native structure and activity of proteins, resulting in their denaturation and aggregation. Thus, pepsin is expected to be in a denatured state during the formation of the clusters. As for an elucidation on the pepsin-QD interaction, there are numerous metal detoxification or bioabsorption studies, looking at sequestration of toxic heavy metals, such as cadmium ions (Cd\(^{2+}\)), from the environment using peptides. The studies search for novel peptides with high affinity for metal ions, and look at the binding mechanism and formation of metal-amino acid complexes. From these studies, it has been found that cadmium (II) ions readily forms chelate complexes with most amino acids (e.g. cysteine, histidine, glycine, alanine, phenylalanine, leucine, valine), some capable of tridentate behaviours (from carboxyl, amino, thiol groups present in the amino acids). In QDs, studies have shown that there is a non-stoichiometric molar ratio of metal to chalcogenide counterions (for CdSe, a molar ratio of 60:40, Cd\(^{2+}\):Se\(^{2-}\), was reported). This is assumed to be due to the fact that the QD crystal structure at the surface is terminated by metal ions that can easily coordinate with ligand molecules, such as TOPO, stabilizing the system. Then, with an accompanied loss of the secondary and tertiary structures of pepsin, it is possible that ligand exchange takes place, whereby the multidentate amino acids of the denatured protein displace the TOPO ligands binding to the Cd\(^{2+}\) on the surface of the QDs through similar association found in the metal sequestration studies. This ligand exchange would provide a driving force for the co-assembly of the proteins and QDs that we observe. In addition, replacement of the hydrophobic TOPO ligands with the denatured pepsin, containing hydrophilic domains, allow for water compatibility of the clusters.
Aspartic Protease: Porcine Pepsin

Various representations of the protein structure of porcine pepsin is shown above. Cyan and red structures represent the two domains/lobes. Blue and pink structures represent the α-helices and β-sheets, respectively, of the protein’s secondary structure. The bluish-purple spheres represent the amino acids found in the active site. On the bottom, the 327 amino acids residues of the porcine pepsin sequence is shown with some of the special residues/regions labelled.
1.4 Mixed Size QD-P clusters & Energy Migration

Next, mixed QD-P clusters made from two different sized CdSe QD samples were prepared for studying energy migration within the system. Evidence for efficient resonance energy transfer from the smaller QDs, acting as donors (D), to the larger QDs, acting as acceptors (A), in the QD-P clusters is explored through steady-state absorption and emission spectroscopy. Relevant background on resonance energy transfer, preparation and characterization of mixed size QD-P clusters, and the result from the energy transfer study will be presented in this section.

1.4.1 Resonance Energy Transfer

Resonance energy transfer (RET), known by many other names, fluorescence resonance energy transfer (FRET), Förster resonance energy transfer (also called FRET), or electronic energy transfer (EET), refers to a process describing the transfer of electronic excitation energy between two electronically coupled molecules, a donor and an acceptor, in physical proximity. A donor fluorophore, in an electronically excited state, may transfer its excitation energy to a nearby acceptor chromophore, through electronic coupling in the form of non-radiative, dipole-dipole interaction (Figure 11). In order for this process to be observable, the fluorescence emission spectrum of the donor molecule must overlap with the absorption spectrum of the acceptor molecule (however the acceptor molecule need not be fluorescent), and both molecules must be within a minimal spatial radius (typically between 0.5 to 10 nm). Energy transfer occurs without the appearance of a photon, nor does it involve collisions or production of heat, and is the result of long-range dipole-dipole intermolecular interaction between the donor and acceptor. As well, this non-radiative energy transfer is achievable over much longer distances, due to the through-space interaction. When the transfer occurs, depending on the type of molecule, the acceptor molecule can either quench or emit the transferred excitation energy.150-156

Dipole-dipole interaction is a classical representation of coulombic (or electrostatic) interaction; hence a classical analogy describing the behaviour of electronically coupled oscillators can be applied to RET to better understand the underlying mechanism. The electronic charge distribution in an excited donor molecule will generate an electric field around the molecule, much like a field generated by a simple, classical oscillating dipole. The electrons in the ground-state acceptor molecule are assumed not to be oscillating. When RET takes place, the oscillating field of the donor stimulates or induces a dipole
oscillation, via electrostatic interactions, of a nearby (acceptor) electronic system which has a “matching” resonance frequency, and thus the energy is passed on. This transfer process is similar to a direct photon absorption process by the acceptor to generate an excited state. Through the coupling between the dipole oscillation of the acceptor and the oscillating electric field of the photon, energy is transferred resonantly from the photon to the acceptor molecule. The electrostatic interaction energy between the two dipoles is directly related to their magnitude and the distance between them. Theodor Förster was able to relate the dipole moments to the oscillator strengths of the corresponding transitions, and thus gave birth to Förster’s theory of resonance energy transfer.\textsuperscript{157}

Forster’s original theory, took the concept of long-range dipole-dipole interaction, frequency dispersion of the donor fluorescence and acceptor absorption spectra, and energy conservation (i.e. matching frequency or resonance), and united them using the Fermi Golden Rule (Equation 8a) for calculating rates in quantum mechanics to obtain an expression for the RET rate:

\[
k_{\text{RET}} = \frac{2\pi}{\hbar} \left| V_{DA} \right|^2 \rho_A \]

\[
= \frac{2\pi}{\hbar} \left| sV_{dd} \right|^2 \int_0^{\infty} J(E) dE
\]

where \( k_{\text{RET}} \) is the rate (or transition probability) of energy transfer, \( V_{DA} \) is the matrix element for interaction between the excited donor and ground-state acceptor, and \( \rho_A \) is the density of final acceptor states, \( s \) is the solvent screening factor for the electronic coupling (which equals \( 1/n^2 \), where \( n \) is the refractive index of the solvent), \( V_{dd} \) is the electronic coupling in the dipole-dipole approximation, \( J(E) \) is the area-normalized spectral overlap of the donor emission and acceptor absorption (energy conservation term). The theory was formulated for a model where the electronic coupling between the donor and acceptor molecule was assumed to be in the weak coupling limit (i.e. weaker than all the vibrational energies involved and weaker than the coupling energies to the solvent or neighbouring molecules – the donor and acceptor are at equilibrium with the surrounding medium during the time of energy transfer), such that there is no influence on their absorption and emission spectra, more specifically, there is no change in the emission lifetime, emission lineshape, absorption lineshape, and oscillator strength from the interaction. In other words, the coupling is present, allowing for the molecules to interact, but is weak enough in that the donor and acceptor still act as separate molecular species. Also, the dipole approximation and screening factor used in the theory are only accurate for systems where the donor and acceptor are far apart. For donor-acceptor pairs found in close proximity, the dipole-dipole approximation fails and a short-range electron exchange mechanism resulting from the overlapping of donor-acceptor orbitals, called the Dexter mechanism, becomes dominant.\textsuperscript{150-156}
Figure 11

Resonance Energy Transfer Jablonski Diagram

- **a** Resonance Energy Transfer Jablonski Diagram
  - The Jablonski diagram shows the coupled transitions (horizontal pink line connecting the dotted arrows/electronic transitions) between the donor emission and acceptor absorbance in a fluorescence resonance energy transfer system. The solid arrows represent absorption and emission transitions, while the wavy green arrows represent vibrational relaxation. It is important to note that energy is transferred without the emission of a photon.

- **b** Absorption and emission spectra of the donor and acceptor showing the donor acceptor spectral overlap required for resonance energy transfer.

- **c** Schematic representation of the angles used for obtaining the orientation factor between the donor and acceptor dipoles in Equations 14 a, b, c.
The unconvoluted form of the Förster equation, shown above, is usually the starting point for many energy transfer theoretical calculations. However, a general expression of the Förster theory, which has found countless applications in a variety of sub-disciplines in biology, chemistry and physics, is sufficient for universal purposes, and defines the distinct relationship between the transfer rate, interchromophore distance, extent of spectral overlap, the lifetime and quantum yield of the donor, and the relative orientations of the transition dipoles, as follows:

\[
k_{\text{RET}} = \left( \frac{1}{\tau_D} \right) \left( \frac{R_0}{r} \right)^6
\]

**Equation 9 a,b**

\[
k_{\text{RET}} = \left( \frac{9(\ln 10)}{128\pi^2N} \right) \frac{1}{r^6} \frac{\kappa^2 \Phi_D}{\tau_D} \int_0^\infty F_D(\lambda) \epsilon_A(\lambda) d\lambda
\]

**Equation 10 a,b**

\[
R_0 = \left[ \left( \frac{9(\ln 10)}{128\pi^2N} \right) \frac{\kappa^2 \Phi_D J(\lambda)}{n^4} \right]^{1/6}
\]

\[
J(\lambda) = \int_0^\infty F_D(\lambda) \epsilon_A(\lambda) d\lambda \text{ (in } M^{-1} \text{cm}^{-1} \text{nm}^{-4})
\]

**Equation 11**

where \(k_{\text{RET}}\) is the rate of energy transfer, \(\tau_D\) is the lifetime of the donor in the absence of acceptor, \(R_0\) is the Förster critical distance, \(r\) is the interchromophore or donor-to-acceptor separation distance, \(N\) is Avogadro's number, \(\kappa^2\) is the dipole orientation factor describing the relative orientation in space of the transition dipoles of the donor and acceptor (ranges from 0 to 4, but typically assumed to be 2/3), \(\Phi_D\) is the quantum yield of the donor in the absence of acceptor, \(J(\lambda)\) is the overlap integral which expresses the degree of spectral overlap between the donor emission and the acceptor absorption (in nm), \(n\) is the refractive index of the medium, \(F_D(\lambda)\) is the corrected and normalized fluorescence intensity of the donor (dimensionless), and \(\epsilon_A(\lambda)\) is the molar extinction coefficient (typically in \(M^{-1} \text{cm}^{-1}\)) of the acceptor at \(\lambda\) (in nm). The electronic coupling for this general expression is contained in the contributions from \((\kappa^2 \Phi_D)/(r^6 \tau_D)\) term and the acceptor dipole strength from the spectral overlap term.

As can be seen from the above equation, the transfer rate is strongly dependent on distance and is proportional to \(r^{-6}\). RET also depends on the donor lifetime, such that the average time the donor spends in the excited state must be of sufficient duration to allow for the event to occur. If the transfer rate is much faster than the excitation decay rate, then energy transfer will proceed efficiently. If the transfer rate is slower than the decay rate, then energy transfer will not proceed or proceed inefficiently. The efficiency of energy transfer (\(E\)) is a measure of the fraction of photons absorbed by the donor which are
transferred to the acceptor, and can be considered the quantum yield of the energy transfer transition. The efficiency is given by:

\[ E = \frac{k_{\text{RET}}}{\tau_D^{-1} + k_{\text{RET}}} = \frac{k_{\text{RET}}}{(k_{\text{rad}} + k_{\text{nr}}) + k_{\text{RET}}} \]  

Equation 12

which is the ratio of the energy transfer rate to the total decay rate of the donor in the presence of the acceptor, includes all forms of excitation deactivation pathways, radiative and non-radiative, as well as the RET process. The efficiency can be rewritten using the relation from Equation 9a, as given by the equation:

\[ E = \frac{R_0^6}{R_0^6 + r^6} = \frac{1}{1 + \left(\frac{r}{R_0}\right)^6} \]  

Equation 13

This equation emphasizes the strong dependence of \( E \) on the interchromophore distance with respect to \( R_0 \) (to the sixth power). As the term \( r/R_0 \) goes to zero (when \( r \ll R_0 \)), the efficiency will approach unity, while as the term goes to infinity (when \( r \gg R_0 \)), the efficiency will approach zero, and when \( r = R_0 \), the efficiency will be at 50%. The Förster critical distance defines a value for the donor-to-acceptor separation distance at which the energy transfer efficiency is 50% based on the optical properties of the system (Equation 10 a,b). At this separation distance, half of the donor excitation energy is transferred to the acceptor via RET, while the other half is dissipated through a combination of other available de-excitation processes. The Förster critical distance is to represent the donor-to-acceptor separation length under which RET is favoured to occur, and this critical distance typically falls within the range of 2 to 6 nm.156

The orientation factor is based on the relative orientations of the donor emission transition dipole and the acceptor absorption transition dipole and is given by the equation:

\[ \kappa = \hat{\mu}_D \cdot \hat{\mu}_A - 3(\hat{\mu}_D \cdot \hat{R})(\hat{\mu}_A \cdot \hat{R}) \]

\[ \kappa^2 = (\cos \theta_T - 3 \cos \theta_D \cos \theta_A)^2 \]

\[ = (\sin \theta_D \sin \theta_A \cos \phi - 2 \cos \theta_D \cos \theta_A \cos \phi)^2 \]

Equation 14 a,b,c

where \( \hat{\mu}_D \) and \( \hat{\mu}_A \) denote the transition dipole moment unit vectors of the donor and acceptor, \( \hat{R} \) is the unit vector of the centre-to-centre separation distance of the dipoles, \( \theta_T \) is the angle between the donor and acceptor dipoles, \( \theta_D \) and \( \theta_A \) are the angles between these dipoles and the vector, \( \hat{R}_r \) joining the donor and the acceptor, and \( \phi \) is the dihedral angle between the planes containing the dipoles (Figure 11c). As
mentioned, $\kappa^2$ can range from 0 to 4 depending on the relative orientation of the donor and acceptor. However, this term is usually taken to be equal to 2/3, which is an appropriate value for dynamic random averaging of the donor and acceptor orientation due to rotational diffusion (prior to energy transfer), applicable when both molecules are freely rotating and can be considered isotropically oriented during the excited state lifetime. In general, the uncertainty of $\kappa^2$ does not seem to result in major consequences in application to the Förster theory, due to the sixth root relationship to $R_0$.\textsuperscript{156,158}

1.4.2 Mixed QD-P Cluster Formation and Characterization

For the formation of mixed size QD-P clusters, QDs of two sizes, approximately 3 and 4 nm in diameter, were combined in a pre-determined ratio, then synthesis procedures for cluster formation, outlined in section 1.3.1, were followed. QDs with PL emission peaks separated by at least 40 nm were used in order to distinguish spectral features between the two different sizes of QDs.

Morphology wise, the mixed clusters look identical to the previous single size population QD-P clusters. However, spectral properties showed characteristics of mixing and interaction. The photophysical properties of the QDs and QD-P clusters are further illustrated in Figure 12. Figure 12a (top) shows the absorption spectra of 3 nm (donor), 4 nm (acceptor), and mixed (3 and 4 nm) QD-P clusters. As expected, the mixed cluster absorption spectrum shows two peaks, at approximately 495 and 546 nm, in the same first absorption peak positions as the individual, single size population clusters. Emission spectra of the single population 3nm, 4 nm, mixed (roughly 1:1) QD-P clusters in water is shown in Figure 12a (middle). Along with the QD-P cluster emission curves, plain mixed QDs (roughly 1:1) in toluene, without the addition of pepsin, is also shown to illustrate an uncoupled version of the mixing. Two distinct peaks in the emission can be noticed, which is attributed to the two differently sized QDs. The blue and red dotted curves represent the fluorescence spectra from the single population donor and acceptor QD-P clusters, shown for comparison of peak positions. For the emission spectrum of the mixed QD-P clusters in water, only one peak is observed. The mixed cluster peak position coincides with that of the acceptor cluster. Figure 12a (bottom) shows a significant spectral overlap of the donor emission and the acceptor absorption spectra. Taking everything into consideration, together with the spectral overlap and packing of the system (which results in a donor-acceptor centre-to-centre separation distance of approximately 4.5 nm), the observations are suggestive of a RET mechanism responsible for the quenching of the donor emission from the mixed QD-P cluster (while sensitizing the acceptor photoluminescence).\textsuperscript{159-163}
Figure 12

Resonance Energy Transfer in QD-P Clusters

(a) Absorption and PL spectra of mixed, 3nm, and 4 nm QD-P clusters. The PL spectrum of mixed QDs are shown for comparison (dashed purple line). The bottom plot shows the spectral overlap.

(b) As the donor:acceptor ratio is systematically increased (increase donors while acceptors held constant), energy transfer events reduce since there is less number of acceptors; thus donor quenching decreases.

(c) Emission spectra of a broad size distribution QD sample and QD-P clusters made from the same sample.
1.4.3 Evidence for Energy Transfer in Mixed QD-P Clusters

RET is potentially possible whenever the donor emission overlaps with the absorption spectrum of the acceptor, along with other requirements previously mentioned. The acceptor does not necessarily have to be fluorescent, but if it is, RET can be easily observed by monitoring the donor emission quenching along with the enhancement of the acceptor fluorescence (although a clear linear enhancement may not be the case, if there are more complicated de-excitation pathways available for the acceptor). In order to confirm the evidence for RET within the system and qualitatively look at efficiencies, the donor to acceptor ratio was systematically varied so that donor quenching and acceptor sensitization could be clearly monitored (Figure 12b). As the donor concentrations were increased, a second peak at the position of the donor emission emerged. At a ratio of almost 10:1, the intensities of the two peaks became equivalent. The results insinuate a high energy transfer efficiency within the system. Table 2 lists the energy transfer efficiencies at the various ratios. The energy transfer efficiency (ET) can be experimentally obtained using the following equation:

\[ E_{RET} = 1 - \frac{F_{DA}}{F_D} \]

where \( F \) stands for fluorescence intensity and the subscripts \( DA \) and \( D \) represent the donor in the presence and absence of the acceptor, respectively. The efficiency is approximately 100% for a donor to acceptor ratio of 1:1, whereas for a ratio of 9.75:1, the RET efficiency decreases to around 23%.

<table>
<thead>
<tr>
<th>D:A ratio</th>
<th>( \Phi_{DA}^a )</th>
<th>( \Phi_{tot}^b )</th>
<th>Efficiency %</th>
</tr>
</thead>
<tbody>
<tr>
<td>1:1</td>
<td>0.000</td>
<td>0.005</td>
<td>100.0</td>
</tr>
<tr>
<td>1.5:1</td>
<td>0.004</td>
<td>0.008</td>
<td>97.5</td>
</tr>
<tr>
<td>5:1</td>
<td>0.020</td>
<td>0.025</td>
<td>58.3</td>
</tr>
<tr>
<td>8:1</td>
<td>0.324</td>
<td>0.017</td>
<td>54.2</td>
</tr>
<tr>
<td>9.75:1</td>
<td>0.400</td>
<td>0.023</td>
<td>23.3</td>
</tr>
</tbody>
</table>

\( ^a \Phi_{DA} \) is the relative QY of the donor in the presence of the acceptor. 
\( ^b \Phi_{tot} \) is the total QY of the mixed cluster.

The absolute quantum yields of the mixed clusters (\( \Phi_{tot} \)) in Table X seem to have no observable trend and tend to fluctuate from synthesis to synthesis. Due to these varying QY values, relative donor (\( \Phi_{DA} \)) and acceptor QYs were used instead of absolute values. Additionally, the QYs listed are not averaged values, rather they are measurements taken from a single continuous experiment under consistent settings.
RET is also demonstrated in samples of single size population QD-P clusters created from QDs showing a large degree of polydispersity. Such polydispersity within a sample can be seen in the PL emission features, and manifests itself as broad peaks and far from normal Gaussian line shapes. The fluorescence spectra of a polydisperse QD sample and QD-P clusters made from the exact same sample are shown in Figure 12c. The polydisperse QD sample has a FWHM of 159 meV and shows a slight deviation from the Gaussian line shape, revealing a slight shoulder to the blue around 530 nm. The QD-P clusters made from these samples, however, show a narrower PL emission (FWHM of 125 meV) with a near perfect Gaussian shape and a red shift in peak position. The shift arises due to the sensitized emission of the larger QDs within the cluster resulting from energy migration. A red shift in similar systems composed of QDs with inhomogeneous size distribution has also been noted in other studies.

![Figure 13](image)

**Figure 13**

Schematic of energy migration process in a mixed QD-P cluster. Excitation energy hops down a QD energy gradient. The gradient is two-fold. First, energy will be transferred from small to large QDs. It is important to note that a distribution of sizes will exist within a single size population of QDs. Secondly, at the same time, energy will tend to hop towards dots that possess the closest corresponding transition dipole moment to their own. Hopping will eventually cease when excitation energy becomes trapped on a large dot.

A basic energy migration mechanism in this system is to picture the excitation energy hopping from one QD to another, through a sea of isotropically oriented transition dipole moments (Figure 13). An acceptor QD with an absorption transition dipole moment most favourably oriented (i.e. parallel and collinear) to the fluorescence transition dipole moment of the excited donor QD will have a higher chance of accepting this excitation energy. Hence, in this particular close-packed system, since the donor is able to selectively choose an acceptor from several other neighbouring QDs, \( \kappa^2 \) may not be a random average, and may take on values in between 2/3 and 1. A value of 1 represents a parallel orientation. A second factor to consider is that excitation energy will flow down a QD energy gradient, meaning excitation energy will
migrate from smaller to larger QDs. Energy hopping will cease when excitation energy becomes trapped in a larger dot, which will eventually fluoresce or decay non-radiatively back down to ground state. In a disordered 3-D antenna system, this happens with significantly greater efficiency than in quasi 1-D systems due to the greater number of hopping pathways available. Also, as the polydispersity study has illustrated, in a mixed QD-P cluster, the QD energy gradient not only exists between the two different sizes, but within the single size population. Hence, there are many processes to consider, not mentioning time scales, packing defects and so forth.

The Förster theory applies when a single donor interacts with a single acceptor with the result of energy transfer. The dynamics are much more complicated in a multiple, randomly dispersed donor–acceptor system such as the cluster reported here. These dynamics have not been theoretically modeled, but we have been able to observe them through the steady state fluorescence quenching experiments as a function of the systematic variations in the donor–acceptor ratios (Figure 12b). However, for a rough estimate, the overlap and energy transfer rate, which can be calculated within the Förster formalism (Equation 9), work out to $1.23 \times 10^{-4}$ cm and 6.9 ns$^{-1}$, respectively. A good spectral overlap is considered to be $<J> \approx 1.00 \times 10^{-4}$ cm. Hence, this particular donor–acceptor pair has a high degree of spectral overlap and a correspondingly high energy transfer rate.

The characteristic size distribution of QD samples results in a large inhomogeneous line broadening in optical properties. Thus, for simplification of a rather complicated treatment of the spectral overlap, the different inhomogeneous line broadening contributions in both the donor photoluminescence and acceptor absorption spectra should be summed over in the overlap calculations. All things considered, from these estimates it can be seen that energy migration within this artificial system seems to be efficient due to good spectral overlap and the 3-D close-packing nature of QDs in the cluster. The quenching experiments suggest that the excitation energy migrates to only a few QDs before trapping occurs, which is consistent with the calculated rate of energy transfer.
1.5 Conclusion

In this chapter, the preparation of water-soluble CdSe QD clusters was presented. The cluster was created using an enzymatic protein, pepsin, which provided the motivation for the self-assembly of the nanocrystals into a 3-D hierarchical structure and allowed for the protein-nanomaterial composite to be compatible with water. A general physical depiction of the protein-nanomaterial composite was constructed using information from various characterization techniques, comprising of various forms of STEM (SEM, TEM, ZC), DLS, SAXS, and steady-state optical spectroscopy (absorption, fluorescence). STEM images showed that these clusters form highly symmetrical, spherical masses that have dense, close-packed interiors. It was calculated that each cluster, formed from 3 or 4 nm QDs, range from 100 to 400 nm in diameter, and contain approximately 40,000 to 300,000 QDs.

It was found that pepsin, not only assists in the formation and water-solubilization of the clusters, but also allows for the electronic coupling of the QDs within the system. With this coupling in place, we were able to observe the coupling effects on the individual optical properties of the nanocrystals and also look at the excitation energy transfer interactions between the nanocrystals in the cluster. From the data, it was found that the optical features of the QD-P clusters remained similar to that of the QDs from which they were formed; with the exception of scatter due to increased particle size displayed in the absorption spectra, and slight peak shifts in both the absorption and photoluminescence spectra due to environmental factors. A slight decrease in QY, going from QD to cluster, was noted (e.g. core-shell CdSe–ZnS QD QY of 78% in toluene and cluster QY of 61.8% in water), with relatively little influence from the type of ligands used for surface passivation of the original QDs. It was found that clusters prepared from two different size populations of CdSe QD samples showed signs of resonance energy transfer. This phenomenon was further examined by looking at the donor PL quenching and acceptor emission sensitization for a series of clusters with the donor to acceptor ratios systematically varied. RET was also found to be present in single size QD population clusters. This means that energy migration in a mixed sized QD system depends, not only on the discrete QD sizes forming the system (i.e. energy flow from small to large QDs) but, on the continuous distribution of sizes available within the single size population (i.e. energy flow from smaller to larger QDs). This extra factor is assumed to allow for greater ease in acceptor selection, thus improved efficiency in energy migration. In conclusion, RET seems to be efficient in this system due to good spectral overlap of the donor-acceptor pair, as well as the 3-D close-packing arrangement of the QDs in the clusters.
1.6 References


Chapter 2

Charge Transfer in CdSe Nanocrystal Complexes with an Electro-Active Polymer
2.1 Introduction

Chapter 1 introduced and explored a popular building block of nanotechnology, the semiconductor nanocrystal, or more specifically, the CdSe quantum dot, in regards to properties of QD clusters and the fundamental phenomenon of resonance energy transfer. In this chapter, we look at another QD-based system, this time with respect to the QD interaction with a molecular dye and the fundamental phenomenon of electron transfer.

Photoinduced interfacial electron transfer between semiconductor nanocrystals and inorganic or organic substrates/adsorbates is a subject of great interest, not only for the purpose of pure scientific gain, but for applications in nanocrystal-based photovoltaic and optoelectronic devices, chemical- and bio-sensing probes, and photocatalysis. Many various composite systems have been created and investigated in recent years, including CdSe QD/TiO$_2$ nanoparticle systems by Kamat et al., various pairings of semiconductor nanoparticles and dyes by Lian and co-workers (e.g. CdSe QD/Re-bipyridyl complexes, TiO$_2$ nanoparticle/Re-bipyridyl complexes, CdS QD/Rhodamine B, etc.), CdSe QD/Ru-polypyridine complexes by Sykora et al., and many others, all of which provide excellent opportunities for improved understanding of charge transfer dynamics through their respectively unique systems. Many of the reports focus on matters ranging from tuning the QD bandgap for obtaining favourable energetics, extending spectral range of productivity through the use of various sensitizers, improving charge separation and suppressing charge recombination, improving donor-acceptor coupling to enhancing photocatalytic or photoelectrochemical efficiencies, all providing creative approaches for designing better QD-based electron transfer systems. However, despite these innovative ideas, the essential interaction of the QD with the substrate/adsorbates and surrounding media (i.e. the surface chemistry and solvation) and how it affects electron transfer, along with the consequences of working with semiconductor nanocrystal-based systems have not been fully explored.

So, in this chapter, we will focus on an uniquely fabricated CdSe QD-based electron transfer system using an electro-active polymer. The following sections will describe the preparation of the complex, characterize its basic properties, take a closer look at the charge transfer phenomenon found in the system, and attempt to understand what important factors need to be considered when dealing with such systems, in addition to some relevant background information.
Chapter 2: Charge Transfer in CdSe Nanocrystal Complexes with an Electro-Active Polymer

2.2 Electro-Active Block Copolymer (PDMAEMA-Ru(bpy)$_3^{2+}$-PDMAEMA)

Since much of the pertinent background information on QDs were covered in chapter one, we turn our attention to another necessary ingredient in our electron transfer system, the electro-active polymer. The electro-active polymer is actually a block copolymer composed of poly(N,N-dimethylaminoethyl methacrylate), a polymer, and ruthenium(II) tris(bipyridine), a dye. This section will discuss some of the interesting properties of these components and how they fit into our system.

2.2.1 Ruthenium Polypyridyl Photochemistry

Ruthenium polypyridine complexes are compounds in which a polypyridine, a multidentate organic ligand composed of multiple pyridine molecules, coordinates to a Ru metal ion. The bonds formed between the central metal ion and the stable polypyridine ligands (via electron donation from the nitrogens of the pyridine rings) are relatively strong and form stable complexes that have unique optical, electrochemical, and magnetic properties. They are one of the most researched family of compounds in inorganic photochemistry. A well-known example is the ruthenium tris(bipyridine), Ru(bpy)$_3^{2+}$, which is the main "electro-active" component in the polymer for this particular system, and have been used in numerous investigations as a photosensitizer due to many of its favourable properties. This bright reddish-orange complex exhibits strong UV and visible absorption (~452 nm in water with an extinction coefficient of 14,600 M$^{-1}$cm$^{-1}$), as well as intense emission (~620 nm) in the visible region. It has both oxidative and reductive capabilities and it is suitable for use in electron and energy transfer reactions due to its long excited state lifetime (650 ns in water).$^{28}$

The unique photophysical properties of ruthenium polypyridine complexes originate from the added layer of metal d-orbitals to the electronic structure of the organic polypyridine ligand. Focusing on just the central metal ion, a ruthenium complex in an oxidation state of II (Ru$^{2+}$) has 6 d-orbital valence electrons (d$^6$) and will have an octahedral coordination geometry (with D$_5$ symmetry). In an isolated atom the five d-orbitals will be degenerate, however in combination with the polypyridyl ligands, a splitting of the d-orbitals into two subsets (t$_{2g}$ and e$_g$ based on orbital symmetry) will result. The energy difference between the two subsets is the crystal field splitting, which defines the energy ordering and subsequently the orbital nature of the lowest lying excited state (for example, which can determine if the excited state can undergo radiative or non-radiative deactivation to the ground state), and plays an important role in the
photophysical properties of the complex. The splitting is dependent on the radius of the central metal ion, the charge on the metal ion, and the nature of the coordinating ligands (i.e. ligand field strength). For this particular complex, a low-spin \((\text{t}_{2g}^6)\) electron configuration is created by the strong-field polypyridyl ligands, causing a large separation between the orbital subsets. Crystal field or ligand field theory treats the electronic structure of the metal-ligand complex as a single entity, but it is convenient to represent an approximate electronic structure by distributing the electrons, involved in bonding or transitions, into orbitals that are localized on the central metal atom and the ligand. So, with this molecular orbital model in place, the filled triply degenerate \(\text{t}_{2g}\) and unoccupied doubly degenerate \(\text{e}_g\) metal orbitals are reclassified as \(\pi_M\) and \(\sigma_M^*\) of the complex molecular orbitals, respectively. As for the ligands, unlike the metal, the orbitals generally remain very similar to those of the free ligand. The polypyridine ligands possess \(\sigma\) (reclassified as \(\sigma_L\)) donor orbitals localized on the nitrogen atoms, and \(\pi\) (\(\pi_L\)) donor and \(\pi^*\) (\(\pi_L^*\)) acceptor orbitals mainly delocalized on the aromatic rings. The subscripts, \(M\) and \(L\), indicate whether the molecular orbital is predominantly metal or ligand in character, however it is important to note that in actuality the molecular orbitals are of mixed character.\(^{29,30}\)

Unifying the transition metal and ligand orbitals into a simplified molecular orbital diagram of the ruthenium (II) polypyridine complex, several different types of transitions open up: ligand-centred (LC) or ligand-ligand transition of the polypyridine molecule, metal-centered (MC) or d-d orbital metal transition, metal to ligand charge transfer (MLCT), and ligand to metal charge transfer (LMCT) transitions. The different types of transition are illustrated in Figure 1a in a molecular orbital diagram, together with the corresponding ground state absorption spectrum (Figure 1b). In the absorption spectrum, bands at 185 (not shown) and 285 nm correspond to spin-allowed LC \(\pi \rightarrow \pi^*\) transitions of the bipyridine, 240 and 450 nm correspond to spin-allowed MLTC \(d \rightarrow \pi^*\) transitions, and shoulders at 322 and 344 nm correspond to MC transitions, while the shoulder in the long-wavelength tail at \(\sim 550\) nm may be likely due to a spin-forbidden MLCT transition. The LMCT transitions are high in energy, thus are not visible in the absorption spectrum.\(^{31-35}\)

In general, it is believed that upon excitation at \(\sim 450\) nm of the \(\text{Ru(bpy)}_3^{2+}\), an electron from the ruthenium metal is promoted to one of the ligands, resulting in an excited MLCT singlet-state \((^1\text{MLCT})\). So, a charge has moved from one region of the complex to another, with the ruthenium metal oxidized while the electron is localized on one of the bipyridyl ligands, as shown in Equation 1.

\[
\text{Ru(bpy)}_3^{2+} \xrightarrow{\ h\nu\ } ^1\left[\text{Ru}^3^+\left(\text{bpy}\right)_2\left(\text{bpy}^*\right)\right]^{2+} \rightarrow ^3\left[\text{Ru}^3^+\left(\text{bpy}\right)_2\left(\text{bpy}^*\right)\right]^{2+}
\]

\text{Equation 1}
**Figure 1**

---

**Photophysical Properties of Ru(bpy)$_3^{2+}$**

a) Simplified molecular orbital diagram representing different transitions for a metal complex in octahedral symmetry ($d_6$).

b) Absorption spectrum of Ru(bpy)$_3^{2+}$ in alcoholic solution.

c) Shows the different deactivation paths possible after excitation in the visible.
Normally, singlet-triplet transitions are forbidden, thus occur with low probability and are often slow, however intersystem crossing (ISC) to the MLCT triplet-state (3MLCT), in this particular situation, becomes quite fast and efficient due to the heavy atom effect (causes high spin-orbit coupling, which mixes singlet and triplet manifolds of states, allowing for nominally spin-forbidden transitions to occur non-radiatively) on the subpicosecond timescale with the efficiency or quantum yield of formation (i.e. efficiency of ISC) of this lowest excited state at near unity. The $[^\text{[Ru(bpy)$_3$]}^2+]]$ has a substantial triplet character and a single ligand localized excitation. The 3MLCT state consists of three closely spaced energy levels in thermal equilibrium, known as the cluster of lowest excited states. These states are relatively long lived and give rise to the complex’s long excited state lifetime and unusual photochemistry.  

From the 3MLCT excited states, three main decay pathways to the ground state are possible (Figure 1c). Two pathways involve direct radiative ($k_r$) and non-radiative ($k_{nr}$) decay from the 3MLCT to the ground state, and the other pathway involves a crossover into the nearby thermally accessible MC excited state ($k_{d-a}$) followed by rapid radiationless decay to the ground state. Luminescence is caused by the radiative transition from the cluster of excited 3MLCT states to the ground state. The rate of non-radiative decay is dependent on the energy gap between the ground state and the 3MLCT states based on the energy gap law (a smaller energy gap between the lowest excited state and the ground state increases the non-radiative rate which makes the excited state lifetime shorter). Transition to the thermally activated MC state is mainly a temperature dependent process, and reduces the lifetime of the emission at higher temperatures. 

The physical separation of the charges causes a rather large dipole moment and makes the excited state molecule relatively sensitive to solvent polarity, hence changing the polarity of the solvent will affect the energy of the emitting state. Also, with the charges separated and paired with a fairly long lifetime (even at room temperature), the excited state simultaneously becomes a strong oxidizing and reducing agent, since the excited electron in the ligand becomes easily susceptible to ionization and the hole created at the ruthenium atom site increases its electron seeking abilities, which can be useful for promoting photochemical reactions.
2.2.2 PDMAEMA and Ligand Exchange

Poly(N,N-dimethylaminoethyl methacrylate) or PDMAEMA is a polymer which incorporates repeating tertiary amino groups that allow for binding of the polymer to a variety of materials, such as CdSe semiconductor nanocrystals. By introducing the ruthenium tris(bipyridine) constituent into the PDMAEMA polymer and forming block copolymers of [(PDMAEMA)-Ru(bpy)$_3^{2+}$-(PDMAEMA)] or RuPDMAEMA, a functional electro-active component of interest can be securely fixed onto the QD surface (Figure 2).44-46

As mentioned previously, CdSe nanocrystals, were prepared with TOPO,47-49 a monodentate organic ligand, which plays an important role in the nucleation, growth, and shape control of the nanocrystal during synthesis. Subsequent to its formation, the ligands, which are labile and in dynamic equilibrium with the surrounding medium, also prevent aggregation, determine the dispersion interactions of the particles, and passivate (electronically) the surface defects found on the nanocrystal. Thus, the surface of the nanocrystal and the type of ligands associated with the surface strongly influences many of the physical and chemical behavior and interaction of the nanocrystals.

Surface derivatization allows for tuning the surface properties of the nanocrystals and is very much a ligand specific process. Ligand exchange is one particular derivatization procedure that allows for the substitution of the existing surface associated ligands with another type, through prolonged exposure of the nanocrystals (in solution) to an excess amount of competing ligand of choice. Generally, as the new incoming ligand molecules bind more strongly to the nanoparticle surface, the existing ligands will be exchanged in a process driven by mass action. The degree of the exchange will strongly depend on the ligand properties and its affinity to the nanocrystal surface.

The QD-RuPDMAEMA complex, exploits this convenient ligand exchange procedure, utilizing the multiple amine binding sites of the PDMAEMA. Owing to the multidentate binding of PDMAEMA, the original TOPO ligands are displaced and the block copolymer binds strongly to the nanocrystal surface. Also, since PDMAEMA is soluble in an extensive selection of media, ranging from non-polar to polar aprotic and protic solvents, the modified QDs are able to form colloidally stable solutions in a wider range of solvents compared to the original TOPO passivated QDs.44-46
Figure 2

QD-RuPDMAEMA Complex Preparation via Ligand Exchange

QD-RuPDMAEMA complex preparation involves the addition of size precipitated QD stock solution in toluene to RuPDMAEMA polymer. The mixture is left stirring for two days at room temperature in the dark. A ligand exchange reaction takes place between the original TOPO ligands of the CdSe QD and the multidentate block copolymer. The newly established complex is now soluble in a wider range of solvents.
2.3 Electron Transfer & Marcus Theory

Electron or charge transfer (ET or CT) is the most fundamental of all chemical reactions. It refers to a process by which an electronic charge is transferred between two chemical species (intermolecular ET) or redistributed to different parts of one large molecular entity (intramolecular ET), where the oxidation states of both participating partners, the donor (D) and the acceptor (A), change. It also describes the mechanism underlying the thermodynamic changes associated with this redox reaction. ET reactions seem conceptually simple, but a barrage of systems and classes exist, ranging from basic metal to ligand ET reactions giving rise to the colourful chemistry in organometallics to complex and crucially life essential ion transporting processes found in biology to cleverly designed artificial energy sources and storage devices based on multi-part CT processes in material science. Depending on a variety of factors, such as the relative electronic states of the donor-acceptor pair, their structural connectivity, the nature of their electronic communication, the nature of the driving force for ET, different categories of electron transfer will have different mechanisms and interactions influencing the associated energies and kinetics of the process. There are many electron transfer theories that have been developed over the past 60 years and the cornerstone of these numerous models have been based on the Marcus theory, which is still widely applied today.

Marcus theory was originally developed by Rudolph A. Marcus to explain the relationship between the rate and the thermodynamics of an ET reaction, formulated to address “outer sphere” ET based on the transition state theory (TST). The general idea of the theory is that vibration-mediated nuclear reorganization of the system and surroundings create a geometrically favourable condition for ET, independent of and preceding the actual process. The theory separates the electronic and nuclear contributions of the process, revealing the importance of the solvent, uncovering the unexpected kinetic behavior of exergonicity, while presenting a way to calculate the Gibbs free energy of activation for the reaction and predicting the rate of ET for the given system. The original Marcus theory provides sufficient description of empirical data within certain limits, however the theory has been extended and developed to overcome limitations and provide a more accurate description and prediction of the mechanisms, energetics, and kinetics involved. The following outlines the basic evolution of the Marcus theory to a more common version presently used.

The essential framework of Marcus theory begins with the use of two parabolic free energy curves to describe the energy surface of a charge transfer reaction (Figure 3a). These plots express the Gibbs free
Figure 3

Energy Transfer and Marcus Theory

a) Marcus Theory curve for electron transfer (shows non-adiabatic and adiabatic transfers). The y-axis is the free energy and the x-axis is the reaction coordinate, which is a simplified axis representing the motion of all the atomic nuclei including solvent reorganization. In order for electron transfer to occur, an overlap of the donor populated orbital and acceptor empty orbital is required in the activated complex. This electronic interaction involves a split of electronic energy levels.

b) Three free energy regimes for electron theory exists (normal, ideal, and inverted) which can be seen from the rate constant versus free energy of reaction plot as predicted by Marcus Theory.
energy (ordinate) of the reactant (D—A) and product (’D—A’) with respect to a single reaction coordinate (abscissa) corresponding to the various summed up nuclear configurations of the system (all of the bonds and angles of the donor and acceptor, as well as the configuration of the surrounding solvent molecules). The variation in nuclear configuration gives a statistical distribution of the reactant and product energies, which can be converted to free energy. Assuming linear response and the central limit theorem, the free energy curves are Gaussian. This is analogous to the parabolic potential curve of the harmonic oscillator model based on the relationship between energy and bond length (and subsequently the vibrational state) of a simple diatomic molecule. It must be noted that this flattened two-dimensional representation of the ET is a gross simplification of the actual multi-dimensional process, and the abscissa and ordinate can take on many different meanings than the designations that they are given.67-71

Electron transfer from the reactant to the product occurs at a crossover point, where the parabolas “intersect.” The transfer occurs at this point mainly because of two reasons. First is the Franck-Condon Principle, which states that electron transfer is an instantaneous process. This means that since the transfer occurs so rapidly there is no change in nuclear configuration during the time of transfer. On the diagram of the Marcus theory curves, this condition would necessitate a vertical transition between the reactant and product curves at constant geometry (i.e. nuclear coordinates). The second is the First Law of Thermodynamics, which states that energy must be conserved. This means that electron transfer is required to be an isoenergetic process, and necessitates a horizontal transition on the diagram of the Marcus theory curves, holding the energy constant. Thus, the only place where both conditions are fulfilled is the crossover point, which represents the energy to which the reactant state must be raised before ET can occur to the product state. This point is known as the activation barrier of the ET reaction.

So, how does ET take place? In most cases, direct contact, more precisely, orbital overlap of the donor and acceptor is required for efficient ET, which is within the sum of their van der Waals radii. The overlap provides a direct, well-defined path for the electron to travel from the donor to the acceptor. However, transfer can occur over distances up to 30 Å, where direct donor-acceptor orbital overlap is negligible. This is achieved through indirect mechanisms of electron hopping or electron tunneling (e.g. superexchange) through the orbitals of the intervening medium (i.e. solvent molecules, bridging molecule).63

In the adiabatic ET case, a strong electronic coupling (|H_{DA}| ≫ k_BT) between the donor and acceptor is required, as in the situation of the direct overlapping orbitals. |H_{DA}| is an electronic coupling term that gives an approximate measure of strength of the orbital interactions between the donor and acceptor (takes
into account electronic interaction, distance, orientation dependence). If the orbitals are of the same symmetry, these electronic states will mix and the reactant and product will form an activated complex at the crossing point. The activated complex, which is in a quasi-equilibrium with the reactant, is a transitional state which is composed of the combined electronic states of the populated energy level of the donor and the unoccupied energy level of the acceptor, separated by an energy gap of $2|H_{DA}|$. With the reaction going through a gradual change, due to the addition of the activated complex, a continuous energy surface (saddle point) forms between the reactant and product with a second quasi-state energy surface just above it, correspondingly separated by an energy gap of $2|H_{DA}|$. This juncture is known as the “avoided crossing” (Figure 3a). The original adiabatic version of the Marcus theory predicts an activation-controlled (or nuclear reorganization-controlled) ET rate ($k_{ET}$), based on the Arrhenius-Eyring equation:

$$k_{ET} = A \exp \left( \frac{-\Delta G^\ddagger}{k_BT} \right)$$  \hspace{1cm} \text{Equation 2}

where $A$ is a pre-exponential or collision frequency factor and $\Delta G^\ddagger$ is the free energy of activation for ET. Marcus then realized that he could derive the activation energy term by solving for the intersection point between the two parabolas from the thermodynamic parameters of the system, arriving at:

$$\Delta G^\ddagger = \sqrt{\Delta G^0 \lambda}$$  \hspace{1cm} \text{Equation 3}

where $\Delta G^0$ is the total Gibbs free energy change for the redox reaction (i.e. the driving force) and $\lambda$ is the reorganization energy which represents the energy required to distort the nuclear configuration of the reactant (the geometry of the D-A including the orientation and polarization of the surrounding solvent) to reach the equilibrium nuclear configuration of the product without the occurrence of ET. Due to the quadratic dependence of the activation energy term, it was discovered that the ET rate which increases with the exergonicity of the reaction. In the normal region, $\lambda > |\Delta G^0|$, the ET rate increases reaching a maximum at the activationless optimal region, $\lambda = |\Delta G^0|$. As the driving force further increases, the rate drops as the reaction becomes further exergonic in what is known as the “Marcus inverted region” ($\lambda < |\Delta G^0|$) (Figure 3b). The idea of a falling reaction rate as the overall reaction free energy becomes more and more favourable turned out to be one of the most significant, counter-intuitive results of the theory.\textsuperscript{57,63,72,73}

The reorganization energy, $\lambda$ is an important aspect of the Marcus theory. It is defined in many different ways. In addition to the definition given above, it can also be thought of as the amount of energy required to push the reaction coordinate from the initial to final state of the reaction to enable ET, or as an actual graphical displacement of the reactant curve with respect to the product curve, or it can correspond to the system’s coupling to the environment. Regardless of the definition, through this parameter, the
significance of the solvent and its intricate role on the relationship between exergonicity and rate are revealed. It is commonly expressed as a sum of the inner sphere ($\lambda_i$) and outer sphere ($\lambda_o$) contributions (Equation 4, always $\lambda > 0$), however the original Marcus theory was designed to deal with outer sphere ET and was later extended to include inner sphere reactions.

$$\lambda = \lambda_i + \lambda_o$$  \hspace{1cm} \text{Equation 4}

So what are the inner and outer sphere contributions? Inner sphere contributions deal with the changes in the internal structure, bond lengths and angles, of the atoms close to the redox centre (i.e. the donor and acceptor structures) including any bridging ligands. Outer sphere contributions refer to changes involving the free solvent and surround media, otherwise known as the solvation shell or local environment. Outer sphere ET is the most common type of electron transfer reaction. It occurs in situations where the donor and acceptor exhibit weak to mild electronic coupling to each other and involves little to no structural changes, retaining the individuality of the donor and acceptor species. Since the internal coordinates remain more or less constant, the reaction coordinate becomes the solvent rearrangement coordinate. The reaction, starting with the system in an equilibriated state with the solvent, progresses as the local dielectric medium reorganizes to some unstable “transition state” nuclear configuration, obtained via thermal fluctuation, from which an ET reaction can then take place. Once the reaction has occurred, the solvent reorganizes once again to stabilize the charge redistribution in the system. Hence, the reorganization energy, for this particular reaction, is dominated by the contribution from the solvation shell ($\lambda_o$):

$$\lambda_o = \frac{e^2}{4\pi\varepsilon_0} \left( \frac{1}{2r_D} + \frac{1}{2r_A} - \frac{1}{r_{DA}} \right) \left( n^2 - \frac{1}{\varepsilon} \right)$$  \hspace{1cm} \text{Equation 5}

where $e$ is the electronic charge, $\varepsilon_0$ is the permittivity of free space, $r_D$ and $r_A$ are the donor and acceptor radii, $r_{DA}$ is the donor and acceptor separation distance, $n^2$ is the optical dielectric constant (square of the refractive index) representing the fast polarizability of the solvent, $\varepsilon$ is the static dielectric constant representing the slow time component of the solvent response. The expression is derived from the dielectric continuum solvent model and reflects the changes in the orientation polarization of the solvent molecules going from an equilibrium to a non-equilibrium state, with respect to the D-A system, preceding ET. The reaction rate is mainly determined by the reorganization of the solvent shell. Once the solvent molecules have acquired a suitable configuration, ET can take place with considerable ease. Inner sphere reactions deal with cases where the donor and acceptor exhibit strong electronic coupling, and involve changes in the internal reorganization of the reactant to form the transition state nuclear configuration, promoting ET and eventually forming the product. Since the reaction involves bond
rearrangement, the reaction rate tends to be much slower (at least four orders of magnitude) than outer sphere reactions. There is no simple expression for inner sphere reorganization energy, since it requires a knowledge of relevant vibrational states (i.e. force constants associated with all the molecular vibrations) from both the reactant and product, which can be difficult to calculate. However, resonance Raman spectroscopy can be used to obtain this value.\textsuperscript{63,72,73}

Despite the success of the theory in predicting many of the accumulated empirical results, the adiabatic ET theory overestimates many rates by a considerable degree, especially in the Marcus inverted region, and is not suitable for explaining ET reactions that take place over long distances, at low to intermediate temperatures, or involving nuclear reorganization through high frequency vibrational modes. It was found that in such situations, the donor and acceptor are usually weakly electronically coupled ($|H_{DA}| \leq 3k_BT$), and the ET rate is reduced and not governed by the formation of the activated state complex. This instance is known as the diabatic or non-adiabatic ET case. In this particular case, the reaction does not move on the lowest energy surface going from the reactant to the product. Since the coupling is weak, the splitting is small at the crossing point, and as the geometry of the system fluctuates, the activated complex geometry can be formed many times but does not necessarily lead to ET. For a productive ET event, the precursor complex must borrow some thermal energy from its surroundings to “jump” or “tunnel” from the reactant energy curve to the product curve at the point of crossing. The instantaneous jump prevents the system from gradually adapting its configuration during the transfer, curtailing the significance of the activated complex, and requires a semi-classical or quantum based modification to the ET expression. The general non-adiabatic ET rate expression is derived from Fermi’s golden rule:

$$k_{ET} = \frac{2\pi}{\hbar} |H_{DA}|^2 \rho(E_{product})$$

Equation 6

where $\rho(E_{product})$ is the density of states of the product, or for this particular derivation, the Franck-Condon weighted density (FCWD) of the acceptor states at the energy of the donor. This expression applies for all non-adiabatic processes and is based on the Born-Oppenheimer approximation, which separates the electronic ($|H_{DA}|$) and nuclear contributions (FCWD). As mentioned previously, $|H_{DA}|$ is proportional to the overlap of the electronic wavefunctions of the donor and acceptor, which reflects their electronic interaction. FCWD factor accounts for the effect of the nuclear configuration changes accompanying the ET reaction and is proportional to the vibrational overlap between the initial and final states.\textsuperscript{63,72,73}

The difference between the classical Marcus theory and the quantum based version lies in the interpretation of the pre-exponential factor, $A$. Whereby $A$, in the classical context, is defined as a product of the critical vibrational frequency (associated with the frequency of passage through the transition) and
the electronic transmission coefficient (describing the crossover probability), has been redefined to directly relate crossover probability with the magnitude of the electronic coupling (Equation 7), while the probability of reaching the crossing point by nuclear coordinates, which depend on the activation barrier (i.e. reorganization energy and standard Gibbs free energy difference) and the temperature of the system within the classical description, is reflected by the FCWD contribution (Equation 8).

\[
A_{\text{adiabatic}} = \frac{2\pi}{\hbar} |H_{DA}|^2
\]  
Equation 7

\[
\rho(E_{\text{product}}) = \langle \text{FCWD}\rangle_{\text{classical}} \propto \exp \left( \frac{-\Delta G^\dagger}{k_BT} \right) = \frac{1}{\sqrt{4\pi\lambda k_BT}} \exp \left( \frac{-\Delta G^\dagger}{k_BT} \right)
\]  
Equation 8

If the nuclear degrees of freedom are treated classically, where nuclear reorganization proceeds only through low frequency fluctuations, the general non-adiabatic ET rate is limited to the semi-classical expression:

\[
k_{ET} = \frac{2\pi}{\hbar} |H_{DA}|^2 \frac{1}{\sqrt{4\pi\lambda k_BT}} \exp \left( \frac{-\Delta G^\dagger}{k_BT} \right)
\]  
Equation 9

Further sophisticated developments to the Marcus theory have been made, but for general purposes Equation 9 serves adequately. Regardless of the model, the common theme among them reveals three important factors influencing the rate of reaction: electronic coupling of the donor and acceptor (incorporated either directly or indirectly), reorganization energy, and the driving force for the ET reaction. Understanding the ET reaction will require a knowledge of how these factors are affected.\textsuperscript{78-84}
2.4 Measuring Faster Time Scale Dynamics

In context to this thesis, solutions, steady-state spectroscopy, mainly absorption and fluorescence, plays an essential part in characterizing the photophysical properties of a sample. The measurements are usually performed with constant illumination while the sample response is also continuously detected and recorded. The spectra obtained gives insight into the fundamental electronic absorption and fluorescence transitions that are relevant to the system. It also allows for the retrieval of other interesting information, such as the verification of the mirror-image relation between the absorption and fluorescence spectra, determination of the 0-0 electronic transition and stokes-shift information, uncovering of vibronic transitions or interactions of the molecules with other solute or solvent molecules, and so forth. Any changes or unexpected irregularities found in the spectral features may be a preview of potentially interesting interactions that are occurring within the system and offers a starting point for more complex measurements that can delve further into these phenomena.

Due to the time scale of these transitions and interactions, steady-state measurements provide averaged information of the time-resolved response of the sample. While these measurements are fantastic for gaining an overview of the system’s photophysical properties, dynamics of molecular processes (i.e. rotational diffusion, resonance energy transfer, dynamic quenching) taking place cannot be resolved directly. So, we turn to time-resolved spectroscopy. Time-resolved spectroscopy is able to resolve some of the faster time scale responses through the use of ultrafast lasers and a high-speed detection system. The information is recorded at a series of time intervals after the system has been exposed to a small perturbation, a light pulse of a suitably short time duration (i.e. short pulse width) shorter than the decay time of the response. The information obtained, unlike steady-state spectroscopy, is more sophisticated and, in most cases, cannot be taken at visual value without additional analysis to extract and interpret the sample dynamics. However, despite the extravagance and the intricacies involved with the experimental setup and data analysis, the wealth of information obtained can be singular. In this chapter, we will make use of two time-resolved measurements, transient absorption and time-correlated single photon counting, to study the ET phenomena in our system through different perspectives. The following sub-sections will briefly explain the principles behind these measurements.
2.4.1 Transient Absorption (TA)

Transient-absorption spectroscopy is an extension to absorption spectroscopy suited for identifying transient species and exploring excited state dynamics in molecules. In this measurement, the data can be acquired in two ways (Figure 4). Either the sample change in absorbance at a selected wavelength can be measured as a function of time after excitation by a flash of light (lifetime/decay spectra) or the change in absorbance at a particular delay time after excitation can be measured as a function of wavelength (transient absorption spectra). The measurement is able to probe transient excited states of species, reaction intermediates, and formation of photoproducts, where the lifetime generally ranges from femtoseconds to kiloseconds.

TA is a type of pump-probe spectroscopy. In pump-probe spectroscopy, an initial “pump” laser pulse is used to perturb a given system creating a non-equilibrium situation (e.g. where some molecules are in an excited electronic state) which will evolve with time. After some time delay, a second “probe” laser pulse samples the effect of the perturbation, taking a snap shot of the system’s state at that particular delay time as the system relaxes back to its initial state before the perturbation. To monitor the time evolution of the perturbation, the delay time between the pump and probe pulse is varied, so that the response of the system can be sampled at various time delays. The time resolution is essentially given by the duration of the pump and probe pulses and not by the detector.

In context to TA, a strong pump pulse, generated by a pulsed laser, is used to create an excited state population, then a weak probe pulse, from either a continuous-wave (CW) or pulsed laser depending on the time resolution of the process being studied, measures the absorbance by monitoring the probe beam transmission intensity before and after excitation with respect to the time delay. The pump pulse needs to be at a wavelength where excitation of the sample will lead to ground state absorption. The probe pulse however can be of a broad spectrum (e.g. broadband light source in the UV-Vis-NIR spectral range) so that the entire TA spectrum can be obtained. Alternatively, if a narrow frequency range probe is used, single wavelength dynamics (i.e. kinetics of the processes involving the transient species) can be obtained. Depending on the time scale (from femtoseconds to seconds) and spectral range (from UV to infrared) of the measurement, TA techniques will vary and have very different experimental layouts.

How does TA work? The intensity of the transmitted probe pulse is influenced by the absence or presence of the pump pulse. In the absence of the pump pulse, the probe pulse will act like the pump pulse, which
**Figure 4**

Translating the transient absorption contributions with a diagram:

- **Pump Off**:
  - Probes: ground state
  - Reference: excited state

- **Pump On**:
  - Probes: ground state, excited state
  - GSB: ground state bleaching
  - SE: stimulated emission (excited state emission)
  - ESA: excited state absorption

**How Transient Absorption Works**

- **a)** TA is a form of pump-probe spectroscopy which is based on contrast mechanisms of ground state depletion/bleaching (GSB), stimulated emission (SE), and excited state absorption (ESA). The pump pulse excites the sample, then the subsequent probe pulse perturbs the created transient state giving rise to the different TA outcomes.

- **b)** The different TA contributions are illustrated in the differential absorption spectrum. TA can also be plotted as a kinetic trace at a specific absorption wavelength to obtain timescale information of a particular transient state.
will be absorbed by the sample and cause ground state absorption, generating an excited state population. This can be thought of as the reference scenario, or better yet, the reference transmitted intensity. In the presence of the pump pulse, an excited state population has already been generated, and the subsequent probe pulse applied to the system can cause a number of various outcomes for this excited state population. The various outcomes are correlated to the transmission intensity of the probe. Hence, by comparing the non-pumped versus the pumped scenarios, the change in sample absorbance ($\Delta A$) can be obtained.

So, what are the various outcomes? There are four main contributions to $\Delta A$. The first is called ground state bleach (GSB). The pump pulse will cause a depletion or “bleaching” of the ground state population through excitation. When the probe pulse is applied, most of the ground state population will be missing, thus the probe beam will be absorbed less in the presence of the pump pulse. If the transmitted intensity of the probe beam in the above reference scenario is compared with this current scenario, the transmission intensity with respect to GSB will be higher, and a negative $\Delta A$ signal will appear in the spectrum. In actuality, some of the probe beam will be absorbed and the magnitude of the GSB will be proportional to the population missing from the ground state. The dynamics obtained for GSB is referred to as the ground state recovery (GSR). Other processes depleting the ground state population (various quenching processes, such as RET, ET) will appear as a GSB in the TA spectra. The second contribution is stimulated emission (SE). When the pump is on and the excited state is generated, the incoming probe pulse can provoke or “stimulate” some of the excited state population to drop back down to the ground state emitting photons of the same phase, frequency, polarization, and direction as the photons of the incident wave. The transmitted probe intensity being detected will include this additional emission, increasing the transmitted intensity of the signal beam, and again showing up as a negative $\Delta A$ value. The magnitude of SE will be proportional to the population of the emitting state. The third contribution is known as excited state absorption (ESA). ESA will occur when the probe pulse is absorbed by the initially formed excited state population to transfer the population to higher excited states. This can only be observed if the probe frequency range corresponds to the ESA transition and has a sufficiently high absorption coefficient. The transmission intensity, in this case, will result in a decrease, and a positive $\Delta A$ signal will appear. If the initially pumped excited state population transfers to other states (e.g. triplet state, charge transfer state), aside from higher excited states, their absorption signature can appear in the TA spectrum as well. The dynamics obtained for both SE and ESA will be similar to that obtained by fluorescence decay measurements. Lastly, polarization anisotropy effects is known to affect the TA signal. The pump and probe beams resulting from the amplified output of laser systems are linearly polarized. In liquid samples, orientation anisotropy is induced by the pump excitation. The probe, which
is also polarized, will feel the polarization influences of the pump and its effects will emerge in the intensity of the $\Delta A$ signal (unless the pump and probe polarization are at a magic angle). Through time-resolved TA anisotropy measurements, information regarding reorientation and geometric changes can be obtained. The first three contributions to $\Delta A$ are illustrated in Figure 4a.

So, through these different contributions, a TA spectrum is generated (Figure 4b), containing the spectral signatures of populated transient electronic states, allowing us to identify the presence of short-lived, metastable species and monitor spectral evolution. And through the dynamics of the TA signal, it enables us to quantify time constants, providing us with kinetic information regarding the transformation of populations from one state to another. Some issues, such as weak signals, limited spectral window, and overlap of broad TA bands (in condensed phase), can arise, making the experiment and analysis of TA data difficult and quite complex. However, there is no doubt that TA spectroscopy is very diverse in its use and a great deal of valuable information can be obtained, making it a quintessential pump-probe technique which can solidly complement the data obtained from other measuring techniques.

### 2.4.2 Time-Correlated Single Photon Counting (TCSPC)

Time-resolved fluorescence spectroscopy is an extension to fluorescence spectroscopy which allows us to measure the time evolution of fluorescence signals. Similar to TA, in this measurement, the sample is excited with a flash of light and the fluorescence, at a selected wavelength, is detected by electronic or optical methods, as a function of time. The measurement is able to probe the excited state nature of molecules and obtain kinetic information regarding processes affecting the excited state dynamics of the system (i.e. RET, ET). There are various types of time-resolved fluorescence spectroscopy available, depending on the sensitivity and time resolution that is required for the investigation.

Time-correlated single photon counting is one of the most common time-resolved fluorescence techniques for measuring the fluorescence decay (lifetime) of a sample on a nanosecond time scale, using an electronic detection scheme. The idea behind TCSPC is a bit unique (Figure 5). In a nutshell, a pulse of light, from a repetitve nanosecond lamp (pulsed-laser diodes or light-emitting diodes) or a high repetition rate mode-locked picosecond or femtosecond laser, is used to excite a sample. When an emitted photon from the sample hits a detector, the time between the excitation pulse and the observed photon is recorded and stored in a histogram. The measurement is then repeated many times, for a large number of excitation
flashes, to build a good statistical data set of single decay kinetics (follows Poisson distribution, a discrete probability distribution that gives the average or probability of the number of times an event will occur within a given interval of time). The histogram represents the probability distribution, with respect to
time, for a photon emission after photoexcitation, generating the fluorescence decay profile of the sample (Figure 5b).

Here are the finer details (Figure 5a). Because the technique relies on analog signals and an electronic detection system, specialized hardware is required to measure the time between the excitation and emission signals. To start, the initial excitation pulse is detected and passes through a constant function discriminator (CFD), which transforms the light pulse into an electronic pulse that triggers the charging of a capacitor, starting a voltage ramp, in a time-to-amplitude converter (TAC). A voltage ramp is a voltage that increases linearly with time. When the light pulse of a single emitted photon from the sample is detected, another electronic pulse from a second CFD will stop the charging of the capacitor, hence the voltage ramp, in the TAC. The voltage of the TAC output is essentially seen as an analogue pulse of a particular amplitude corresponding to the time delay between the excitation and emission signals. This pulse is then amplified by a programmable gain amplifier (PGA), then measured and converted to a digital value by the analog-to-digital converter (ADC) or a multichannel analyzer (MCA) for older systems, which has a similar function. The ADC establishes how many discrete time values (i.e. channels) are possible and determines the time resolution of the measurement, usually given in picoseconds/channel or nanoseconds/channel. All measured TAC amplitudes are put into a corresponding channel in a memory bin location on a computer. Then after many repeated measurements, the memory bins get statistically averaged, by a program, to form a histogram of photon counts versus channel (or time) representing the waveform of the fluorescence lifetime. To minimize false readings or in the case where a photon was not emitted (no stop pulse for TAC), a window discriminator (WD) suppresses all TAC signals not falling within a given range of voltages, limiting the signals carrying on to the ADC.

The fluorescence decay profile obtained from the measured signals (i.e. histogram) is a convolution between the instrument response function (IRF), which is composed of the time profile of the excitation pulse and the response function of the detector, and the sample response (Figure 5c). Since the IRF has a finite temporal width, it will distort the intrinsic fluorescence response from the sample. In order to extract the exclusive sample fluorescence lifetime, two curves are measured in a standard experiment, the IRF (using a scatter solution) and the sample fluorescence decay. Then in the analysis, the IRF is convoluted with some model function (e.g. single exponential decay, multi-exponential decay, or some other function) until the resulting convolution matches the experimental sample decay. This matching is achieved through an iterative numerical procedure until a best agreement is found. Normally, a qualitative assessment regarding a particular mechanism (i.e. curve fitting function) can be made by examining the raw decay curve.
One of the most important issues surrounding TCSPC is the “pulse pileup,” which is related to the limitations on the electronics only being able to detect the first arriving pulse. Due to this reason, it is imperative to ensure that TCSPC conditions are adjusted to detect no more than one photon per laser pulse. Multi-photon events will result in nonlinear signal distortions (peal-up distortion) which will affect the histogram statistics, yielding underestimated lifetimes. So, in order to safeguard this condition, the detection rate is kept overcautiously low, typically 1 photon per 100 excitation pulses (count rate of 1%). This condition can lengthen the data collection time required for a reliable histogram.

Due to the high repetition rate of modern lasers, lifetime measurement can be performed much quicker compared to earlier generations. However, the maximum repetition rate of the laser is determined by the sample and cannot be shorter than the sample decay time, even if the laser is capable of pulsing at a higher frequency. And despite higher repetition rates being available, the “dead time” associated with the electronics preventing detection (reset time associated with TAC and overall system) and the “1%” count rate rule, the technique will still be limited and it will not be able to process an overflow of emitted photons generated by the high repetition rates. So, in order to utilize the idle electronic transient times when no photons are detected, TCSPC measurements are typically performed in “reverse mode”. The principle is identical to the above description, the only difference lies in the fact that initiation and termination of the TAC is carried out in reverse (i.e. the emission pulse is used to start the TAC and the excitation pulse is used to stop the TAC). In this mode, a shifting delay of the reference excitation pulses and an additional inversion of the histogram time scale (for photon events with long retardation times) is required, but the data collection time can be greatly reduced in comparison with the forward mode.86,87

Overall, TCSPC is a well-established and common technique for time-domain fluorescence measurements. Despite some of the issues associated with the measurement, it is a simpler, less expensive, and just as efficient technique compared to other time-resolved fluorescence measurements (i.e. streak cameras and fluorescence upconversion), albeit with less resolution.
2.5 CdSe Nanocrystal Complexes with Electro-Active Polymer (QD-RuPDMAEMA)

In this section, the synthesis of RuPDMAEMA block copolymer and the preparation of CdSe nanocrystal complexes with RuPDMAEMA is presented. The prepared complexes are then characterized through UV-Vis absorption and photoluminescence spectroscopy, scanning transmission electron microscopy, time-correlated single photon counting, and transient absorption spectroscopy.

For materials, commercially available technical grade chemical reagents and spectrophotometric grade solvents were purchased from Aldrich. Trioctylphosphine oxide, trioctylphosphine, and 100 mesh selenium powder were used without further purification. Acenaphthylene (Rütgers) was recrystallised three times from methanol. N-dimethylaminoethyl methacrylate (DMA) was purified by passing a short column of basic alumina oxide. High purity dimethylcadmium was purchased from STREM and used as received. 2,2-Azobis (2-methylbutyronitrile) (AMBN) was obtained from Wako Chemic Co. and used as received. Also, the molecular weights of the polymer were characterized by gel permeation chromatography (GPC) performed in tetrahydrofuran with 2% triethylamine (V/V, 0.6 mL/min) at 25°C with a Waters 515 HPLC pump, a Viscotek VE3580 Refractive Index Detector, a VE3210 UV/Vis Detector, and a Waters Styragel HR 4E column. The GPC was calibrated with a narrow polydispersity polystyrene standards (MW from 580 to 377400) and the molecular weights are reported as polystyrene equivalents. Synthesis of RuPDMAEMA block copolymer and related measurements in 2.5.1 were performed by Dr. Ming Chen from Dr. Gerald J. Wilson’s research group at CSIRO Molecular and Health Technologies in Australia, under the supervision of Professor Mitchell A. Winnik of the University of Toronto.

2.5.1 Synthesis of RuPDMAEMA samples

PDMAEMA samples (M_n = 27K, PDI = 3.9) are synthesized by a conventional solution polymerization of DMA in toluene at 95°C, initiated with an azo-type free-radical initiator, AMBN. PDMAEMA(18)-Ru(bpy)_3^{2+}-PDMAEMA(18) block copolymer with a functional Ru(II) tris-bipyridine complex is synthesized using RAFT (reversible addition-fragmentation chain transfer) polymerization. A Ru(bpy)_3^{2+} dye RAFT-acid, bis(dithiobenzoyl) functionalized Ru(II) complex (Ru-di-RAFT), is prepared as reported, forming the initial RAFT agent. Copolymerization of the DMA monomers with the RAFT
agent, followed by a RAFT end group termination procedure, produced the final desired polymer. Scheme 1 outlines the synthesis and the following describes the procedures in more detail.
For the synthesis of Ru(II) complex functionalized N-dimethylaminoethyl methacrylate polymer (RAFT-PDMAEMA-Ru(bpy)$_3^{2+}$-PDMAEMA-RAFT), a mixture of DMA (6.37 M), AMBN (0.021M), Ru-di-RAFT (0.106 M), and toluene in ampoules attached to a vacuum were degassed through three freeze-pump-thaw cycles, and heated in a 70°C oil bath for 8 hours. Polymerization is terminated by rapid cooling in cold water. The polymer was then precipitated into a rapidly stirred excess of $n$-hexane. The solid was purified twice by re-dissolving in toluene and re-precipitated in $n$-hexane. The precipitated polymer was finally dried under vacuum at room temperature to a constant mass (50% conversion).

In the removal of the dithiobenzoyl end group from polymer, a mixture of RAFT-PDMAEMA-Ru(bpy)$_3^{2+}$-PDMAEMA-RAFT, 20 molar equivalents of AMBN, and toluene were degassed through three freeze-pump-thaw cycles, sealed under vacuum, and heated at 80°C for 2h. The polymer was then precipitated three times by addition into a rapidly stirred excess of $n$-hexane. The precipitated polymer was finally dried under vacuum to a constant mass.

### 2.5.2 Complex Formation

The preparation of CdSe semiconductor nanocrystals by a well-established organometallic method is outlined in section 1.2.2.1. The synthesized QDs showed quantum yields of 0.5-1% and emission spectra FWHM of 35-40 nm. The surfaces of the QDs were capped by a monolayer of TOPO allowing solubility in various types of organic solvents.

QD-RuPDMAEMA complexes, and similarly QD-PDMAEMA complexes, are prepared by the addition of an aliquot of a size-precipitated QD stock solution in toluene (~7.17×10^{-7} mol/L) to 1 mg of RuPDMAEMA copolymer (or PDMAEMA polymer) to form a mixture of solution with a concentration of approximately 5.859×10^{-5} mol/L. The mixture is left stirring for two days at room temperature in the dark to allow the ligand exchange reaction between the block copolymer and the original TOPO ligands to take place. To transfer the complex into the various solvents, the newly formed complexes are precipitated using hexane (a non-solvent) and collected through centrifugation. Subsequently, the precipitant was redissolved in a solvent of choice via sonication. It is important to note that both the excess polymer and complex will be soluble and present in the solvents used.
2.5.3 Characterization

Scanning transmission electron microscopy (STEM) images were obtained in high-resolution annular darkfield (ZC) and transmission electron microscopy (TEM) modes using a Hitachi HD-2000 instrument. Absorption spectra were obtained on a CARY BIO UV-Vis spectrophotometer. Steady-state photoluminescence spectra were measured using a Cary Eclipse fluorescence spectrophotometer. Fluorescence decay profiles were recorded in solution using a nanosecond time-correlated single photon counting system with a 456 nm NanoLED from IBH as the excitation source. Femtosecond transient absorption measurements were conducted by Chi-Hung Chuang from Professor Clemens Burda’s research group at Case Western Reserve University in Cleveland, Ohio, using a Clark MXR 2001 femtosecond laser system producing 780 nm, 150 fs pulses from a regenerative amplifier. The laser pulse train was split to generate a white light continuum probe pulse in a sapphire crystal and a tunable pump pulse. The tunable pump pulse was generated using an optical parametric amplifier (TOPAS, Lightconversion). All femtosecond laser experiments were carried out in a 2 mm quartz cuvette at room temperature. The instrumental time resolution was determined to be ~150 fs via a pump-probe cross-correlation analysis. The reported spectra were all collected from fresh samples at room temperature.

2.5.3.1 STEM Images, UV-Vis Absorption and Fluorescence Spectroscopy

High-resolution annular darkfield images, from the STEM, were taken of significantly diluted QD-RuPDMAEMA samples, prepared in toluene (Figure 6a). The images show that the complexes are well dispersed and do not aggregate following ligand exchange, confirming solution stability. In an attempt to image the RuPDMAEMA polymer in association with the QDs, the sample preparation was drastically altered to reduce the amount of excess RuPDMAEMA in the solution, which interfered with the imaging. However, due to the changes in the sample recipe, the images obtained seem to show an incomplete ligand exchange, showing the QD in partial association with the RuPDMAEMA. The TEM images obtained, which do not resolve organic matter (i.e. PDMAEMA polymer), also confirms the decent dispersion of the complexes in the sample and show that there are no observable physical changes between the original QD-TOPO and newly formed QD-RuPDMAEMA samples.
Figure 6

Characterization of QD-RuPDMAEMA

a) High-resolution annular darkfield STEM images of QD-RuPDMAEMA.

b) Absorption and emission spectra for CdSe QD, RuPDMAEMA, QD-RuPDMAEMA, and QD-PDMAEMA (left). Emission spectra of QD-RuPDMAEMA complex formation, taken at increasing time intervals, from start of stock solution mixing to after two days of equilibration (right).
The absorption and emission spectra of the CdSe QDs, RuPDMAEMA polymer, QD-PDMAEMA complex, and QD-RuPDMAEMA complex are shown in Figure 6b (left). The QD samples used in the complex exhibit their first absorption peak at 530 nm (2.34 eV) and fluorescence peak at 542 nm (2.29 eV). A QD diameter of approximately 2.7 nm can be estimated from the energy of the lowest exciton band and can also be roughly confirmed by the STEM images. The RuPDMAEMA polymer shows an absorption maximum at 458 nm (2.71 eV) and emission peak at 627 nm (1.98 eV). The QD-RuPDMAEMA complex absorption and fluorescence spectra are comprised of the same peaks and peak locations as its constituents. Also, the QD-PDMAEMA complex, similar to the QD-RuPDMAEMA complex except without the electro-active (Ru(bpy)₃²⁺) ingredient, has an absorption spectrum that is virtually identical to the CdSe QD samples, within the visible wavelength range of interest. The minor shifts can be attributed to the differences in the contribution of the polymer or TOPO ligands on the QD surface passivation.

The QD-RuPDMAEMA complex formation was systematically monitored through its emission intensity (Figure 6b, right). Emission spectra were taken at increasing time intervals following the mixing of the initial components to approximately after two days of equilibration. The QD (~542 nm) and RuPDMAEMA (~630 nm) peaks of the complex were noted to have their features decreasing in intensity over time. The simultaneous quenching in both peaks did not seem to be in accordance with typical resonance energy transfer behavior, as seen in chapter 1, since there was no evidence of donor emission quenching followed by acceptor peak sensitization, although RET cannot be fully discounted due to a partial donor and acceptor spectral overlap present in the system. Sykora et al.²² had also reported similar quenching in their investigation with CdSe QDs and ruthenium polypyridine complexes. They explained that the quenching they observed occurred upon gradual adsorption of the complexes onto the QD surface and attributed their observations to electronic interactions between the two components, most likely a charge transfer reaction.

A closer look at the complex formation spectra reveals a faster quenching of the QD emission peak compared to the RuPDMAEMA polymer. The initial ratio of the QD:RuPDMAEMA peak intensity at the start of mixing is 0.28:1 and evolves to 0.17:1 after two days. The faster quenching of the QD peak suggests that more than one RuPDMAEMA polymer is adsorbed onto the QD surface over time. The adsorption of more than one polymer onto a single QD surface can allow for the possibility of multiple, concurrent charge transfer reactions to occur, which can result in a more rapid decrease in the emission intensity. Since the polymer was added in excess, the presence of the free polymer in the mixture is expected to contribute significantly and dominate the steady-state RuPDMAEMA emission peak intensity.
The quenching in the emission intensity of the RuPDMAEMA also seems to be in association with the polymer’s increasing fraction binding to the QD surface over time. The quenching in both of the emissions can be due to various possible reaction pathways, not just exclusive to charge transfer. These mechanisms are intricate and will require additional experiments to elucidate.

The information regarding the average number of polymer units per QD cannot be straightforwardly extracted from these ratios due to the presence of the large excess of polymer. However, an estimate can be obtained from previous work, which demonstrated a method of isolating a similar QD-polymer complex from the free polymer and quantifying this information using size-exclusion chromatography. It was found that on average, approximately 4 to 5 PDMAEMA polymer molecules (Mn = 6.7K, labeled at one end with a pyrene molecule) were bound to a single CdSe QD, approximately 2.5 nm in diameter. These approximate figures may be applied to our system given the similarities in the components (CdSe QD with d = 2.7 nm, Ru(bpy)$_3^{2+}$ labeled PDMAEMA polymer with Mn = 6.68K).

### 2.5.3.2 Fluorescence Decay

In order to investigate PL dynamics for the QD-RuPDMAEMA complex, the fluorescence decay profiles of the QD, RuPDMAEMA, QD-PDMAEMA and QD-RuPDMAEMA were qualitatively assessed from nanosecond TCSPC measurements. Figure 7, compares the decay curves of the QD, QD-PDMAEMA and QD-RuPDMAEMA samples with the detector set at 550 nm (QD steady-state PL maximum) and the decay curves of the RuPDMAEMA and QD-RuPDMAEMA samples set at 630 nm (Ru steady-state PL maximum). In Figure 7 (left), a faster decay of the QD sample can be seen in relation to the QD-RuPDMAEMA complex. This observation could possibly suggest a longer lived or delayed QD-RuPDMAEMA excited state fluorescing population owing to the addition of the electro-active polymer into the system, however, the decays of QD-PDMAEMA and QD-RuPDMAEMA look fairly similar at first glance. It has been reported previously that the QY of CdSe QDs are dependent on the type of ligands adsorbed onto the surface of the nanocrystal, changing the passivation, which affects the efficiency of the surface trapping of charge carriers (i.e. electrons or holes). Hence, the differences seen in the QD and complex decays are most likely attributed to and dominated by the surface states of the QDs due to the differences in surface defect passivating abilities of the TOPO and PDMAEMA ligands. In Figure 7 (right), the changes in the decay profiles of the polymer and polymer-QD complex also seem fairly
nominal. There may be some subtle differences in these similar decay profiles, however the distinctions, at this point, cannot be clearly determined in the time range and resolution of the measurement.

**Figure 7**

TCSPC Fluorescence Decay Profiles of QD-RuPDMAEMA
The left plot shows decay profiles of QD, QD-PDMAEMA, and QD-RuPDMAEMA with the detector set at 550 nm, the QD fluorescence peak maximum. The right plot shows decay profiles of RuPDMAEMA and QD-RuPDMAEMA at 630 nm, the RuPDMAEMA fluorescence peak maximum.

**2.5.3.3 Transient Absorption**

Femtosecond TA spectroscopy was also used to obtain further characterization of the QD-RuPDMAEMA complex and charge transfer dynamics found in the system. Figure 8a shows the TA spectra for the CdSe QD, RuPDMAEMA polymer, and QD-RuPDMAEMA complex following ~150 fs FWHM excitation at 458 nm. Starting with the characteristic features of the QD, transient bleach signals at ~530 nm (Figure 8a, green) corresponding to the nanocrystal exciton transition are evident. In the QD measurements, the excitation power was controlled to ensure the average number of excitations to be below one. In Figure 8a (yellow), the long-lived transient bleach signal of the RuPDMAEMA polymer is seen at ~455 nm. The location of the bleach band in the TA and the absorption maximum in the steady-state spectra are consistent. Figure 8a (red, 0.9 mW) shows the TA spectrum of the QD-RuPDMAEMA complex in toluene. The bleach signals near 530 and 455 nm correspond to the QD and RuPDMAEMA states,
respectively. For the QD-RuPDMAEMA complex, a high excitation power was required to obtain bleaching signals in both spectral positions of the CdSe and RuPDMAEMA transitions.

Figure 8b (left) and Table 1 shows the signal decays at 465 nm for the RuPDMAEMA polymer and QD-RuPDMAEMA complex. Based on the steady-state absorption spectra, the kinetic traces measured at this probe wavelength should represent a superposition of both the ground state (GSB) and excited state (SE) dynamics. Due to the strong ground state absorbance of Ru(bpy)$_3^{2+}$, hence RuPDMAEMA, the temporal signal profile typically seen would represent ground state recovery (i.e. the decay curve is formed as the transmittance of the probe beam decreases, hence absorption increases, caused by the repopulation of the ground state over time), however as the probe is shifted away from the main bleach, excited state dynamics can become dominant if SE features are intense enough to manifest themselves through the superimposed bleach (where the decay curve is formed as SE processes decrease and less emission is released as the excited state population decreases over time). Prior to the polymer’s association with the QD surface, the kinetic trace of the free RuPDMAEMA is long-lived with no decay in the first 100 ps. This indicates that the bleach is long-lived, and under isolated conditions (i.e. in the absence of any reactions), we can assume that GSR takes a long time to occur because the excited state is long-lived. In the QD-RuPDMAEMA complex, an extremely fast decay is observed. A numerical fit, using a biexponential function, was used to obtain two components 0.73 (82%) and 15.2 (18%) ps, yielding an average decay time constant of 3.33 ps. Here, the bleach is very short lived, however, at this point, it is difficult to distinguish whether GSR (i.e. repopulation of the ground state) or excited state (i.e. decay of the excited state population) dynamics is dominating the signal measured. But, there seems to be some unique process(es) present for the polymer-bound complex, not present in the free polymer, which is responsible for the exceptionally faster dynamics that are occurring on a picosecond time scale.

The availability of ultrafast techniques, in the last few decades, has allowed for the possibility of investigating the early-time excited-state dynamics of Ru(bpy)$_3^{2+}$. The mechanistic details of the earliest moments following photoexcitation are important in influencing processes associated with and subsequent to excited-state formation (i.e. excited-state reactivity). As previously mentioned in section 2.2.1, the general understanding is that the emissive state of Ru(bpy)$_3^{2+}$ is localized on a single ligand, however the early-time dynamics which lead to this emissive state is still in consideration. The largely accepted belief is that photoexcitation of Ru(bpy)$_3^{2+}$ in the $^1$MLCT manifold produces an initial Franck-Condon state that is delocalized (i.e. promoted electron is shared by all the ligands). Then, the promoted electron becomes localized on one of the ligands, due to local solvent dipole coupling, which occurs in tens of femtoseconds. ISC then takes place producing a localized triplet state within about 100 fs. Finally, the $^3$MLCT becomes
Figure 8

Transient Absorption of QD-RuPDMEMA

a) Transient absorption spectra of QD (green), RuPDMEMA (yellow), and QD-RuPDMEMA in toluene at various pump-probe delay times (taken at different excitation powers, $\Phi$, using a ~150 fs FWHM excitation pulse at 458 nm).

b) Transient absorption decays of RuPDMEMA and QD-RuPDMEMA collected at 465 nm (left), and for QD and QD-RuPDMEMA at 532 nm (right) (QD-RuPDMEMA was taken at an additional excitation power, 0.9 mW).
randomized through interligand hopping on a timescale of 10 ps (thermalized 3MLCT excited state). Since our work has been carried out with ~150 fs resolution, we are not able to gain insight into these initial processes, but based on literature, we assume that the data we have acquired largely reflect dynamics on the 3MLCT surface (since ISC is presumed to have already occurred within our excitation pulse).33,41,42,93-95

Table 1 Analysis of kinetic parameters of QD, RuPDMAEMA, and QD-RuPDMAEMA complexes in toluene.

<table>
<thead>
<tr>
<th></th>
<th>Excitation (\Phi) (mW)</th>
<th>Excitation (\lambda) (nm)</th>
<th>Probe (\lambda) (nm)</th>
<th>(\tau_1) (ps)</th>
<th>(\tau_2) (ps)</th>
<th>(\tau_{ave}) (ps)</th>
</tr>
</thead>
<tbody>
<tr>
<td>RuPDMAEMA</td>
<td>0.9</td>
<td>458</td>
<td>465</td>
<td>--</td>
<td>--</td>
<td>1207</td>
</tr>
<tr>
<td>QD</td>
<td>0.06</td>
<td>458</td>
<td>532</td>
<td>8.19%</td>
<td>267 44%</td>
<td>122</td>
</tr>
<tr>
<td>QD-RuPDMAEMA</td>
<td>0.06</td>
<td>458</td>
<td>532</td>
<td>4.79% 59%</td>
<td>223 41%</td>
<td>94.3</td>
</tr>
<tr>
<td></td>
<td>0.9</td>
<td>458</td>
<td>532</td>
<td>5.74 63%</td>
<td>157 37%</td>
<td>61.7</td>
</tr>
<tr>
<td></td>
<td></td>
<td>458</td>
<td>465</td>
<td>0.73 82%</td>
<td>15.2 18%</td>
<td>3.33</td>
</tr>
</tbody>
</table>

In Equation 1, \*[Ru(bpy)\(_3\)]\(^{2+}\) is depicted as an oxidized ruthenium metal (Ru\(^{3+}\)) with the excited electron localized on one of the bipyridyl ligands (bpy\(^{-}\)). The RuPDMAEMA TA spectrum obtained is similar to that of the difference absorption spectrum for Ru(bpy)\(_3\)\(^{2+}\) (in water) found in literature.96 The interpretation of the difference spectrum, based on the excited state absorption, reveals intense positive absorption bands in the 360-380 nm region and residual (low amplitude) positive absorption at wavelengths greater than 500 nm due to transitions localized on the bpy\(^{-}\) ligand, while a negative absorption band in the 400-450 nm region is representative of Ru(bpy)\(_3\)\(^{3+}\). If we take a closer look at the TA spectra for the RuPDMAEMA polymer and QD-RuPDMAEMA complex (Figure 8a, yellow and red, 0.9 mW), based on the above interpretation, we can state that in the original spectra for the polymer, both features, the bpy\(^{-}\) ligand (>550 nm, pronounced at 690 nm) and Ru(bpy)\(_3\)\(^{3+}\) (~455 nm), can be found and are relatively constant with respect to time, suggesting that these particular transient species of the excited state is long-lived. While in the QD-RuPDMAEMA complex, the Ru(bpy)\(_3\)\(^{3+}\) feature is seen diminishing over time and the bpy\(^{-}\) ligand feature seems to be almost nonexistent (near 690 nm \(\Delta A\) is near 0) for all times. Since our time resolution is only 150 fs and the lack of a bpy\(^{-}\) ligand absorption feature at time zero, leads to the possibility that the localized electron on one of the ligands may have relocated (possibly ET to the QD CB) within the 150 fs of the excitation pulse and the transient bpy\(^{-}\) feature remains absent for the duration of the measurement (1399 ps). The diminishing Ru(bpy)\(_3\)\(^{3+}\) feature shows the disappearance of the transient species over time, which suggests that Ru(bpy)\(_3\)\(^{3+}\) is being converted back to Ru(bpy)\(_3\)\(^{2+}\) (ground state recovery) through some other process. This other process may simply be a charge recombination process (where the transferred electron from the QD CB recombines with the Ru(bpy)\(_3\)\(^{3+}\)), subsequent to electron transfer. Or, since a high excitation power was used to obtain bleach signals in both the QD and polymer spectral positions for the QD-RuPDMAEMA complex, simultaneous
excitation of the QD and polymer species in the complex may likely have induced a side redox reaction between the Ru$^{3+}$ form and the excited QD (more specifically, the electron in the CB), causing the ruthenium species to recover to its Ru(bpy)$_3^{2+}$ state. The oxidized form of Ru(bpy)$_3^{2+}$, Ru(bpy)$_3^{3+}$, is an extremely strong oxidizing agent.

In addition to the RuPDMAEMA, the kinetic traces at 532 nm were obtained for the free QD and QD-polymer bound complex (Figure 8b, right). Due to the large near-degeneracy of VB edge states compared to the CB edge states in the CdSe QD (a difference of 0.05 eV or 20 nm red shift between the 1S absorption band and the PL maximum), it has been stated that TA dynamics measured at this transition provides information about the depopulation rate of the electrons from the CB band-edge states (1S excited state). The TA data for the QD samples were obtained using a low excitation power which made for a single-exciton scenario in the CdSe QDs. An average decay time of 122 ps was obtained using a biexponential fit (8.19 ps (56%) and 267 ps (44%)), which is a typical value assigned for electron relaxation from the conduction band to the defect surface states. In the QD-RuPDMAEMA complex, decays were obtained for two excitation powers (Table 1). On the lower power, the polymer-bound complex had an average decay time of 94.3 ps (4.79 ps (59%) and 223 ps (41%)), while on the higher power, the complex had an average decay time of 61.7 ps (5.74 ps (63%) and 157 ps (37%)). A higher power was used to observe both the QD and RuPDMAEMA bleaching, hence multiple excitations may have been generated in the QDs for the QD-RuPDMAEMA samples. Compared with the kinetic traces of the free QD, there is an overall faster decay rate measured for the complex, in either the low or high excitation power scenarios. However, due to the possibility of multi-exciton effects generated in the complex for the high power case, the faster decay may be partially accredited to this side-effect. Single-excitons mainly decay through slow radiative recombination which occurs on a nanosecond time scale, while multi-excitons mainly decay through fast non-radiative Auger recombination which occurs on a picosecond time scale. Also, the redox reaction resulting from the simultaneous excitation of the QD and polymer species, described above, will be an added contribution to the faster excited state decay observed.

If we consider the lower excitation power scenario (where multi-exciton and simultaneous excitation effects are less likely) and perhaps think of the decay in terms of GSR, we can state that there is some process that is repopulating the ground state at a faster rate compared to the free QD case. Similar conjugation systems consisting of ruthenium polypyridine dyes and CdSe QDs were also investigated by Schaller et al. using TA and time-resolved up-conversion experiments. They were able to determine that a hole transfer from the CdSe QD to the ruthenium polypyridine dye occurs (in ~5 ps) in their systems, with the QDs serving as sensitizers. This explanation would complement the faster GSR
dynamics that is observed for the QD-RuPDMAEMA samples. Since a hole transfer from the QD (VB) to the RuPDMAEMA (ground state) is the same as an electron transfer from the RuPDMAEMA to the QD. So, these transferred electrons are replenishing the bleached VB state of the QD, hence a faster GSR may result for the QD-polymer bound complex.

Based on these results, contrasting interpretations can be applied to build an argument for several different ET mechanisms for the system. Due to the difficulties involved with the measurement of this sample and various uncertainties and unavoidable side factors associated with the results, it would be difficult to be biased toward one mechanism at this point. In the next section, we take a look at the energetics of the system to see if further elucidations can be made.
2.5.4 Possible Processes of the Photoexcited QD-RuPDMAEMA Complex

Before we take a look at possible photoinduced processes of the QD-RuPDMAEMA complex, the relative positions of the VB/CB band-edge states for the QD and the reduction potentials of the RuPDMAEMA (i.e. Ru(bpy)$_3^{2+}$) must be considered. The energy of the CB edge for the QD was estimated from bulk CdSe values ($E_g = 1.74$ eV, $E_{CB} = 4.95$ eV, and effective electron and hole masses of $m_e^* = 0.13m_0$ and $m_h^* = 1.14m_0$, where $m_0$ is the electron rest mass)$^{9,104-106}$ and the absorption energy of the first absorption peak of the QD ($E_{1s}$) using Brus’ equation:

$$E_{CB}(QD) = E_{CB}(bulk) + \Delta E_{confinement}\left(\frac{m_h^*}{m_h^* + m_e^*}\right)$$  \hspace{1cm} \text{Equation 10}

The calculated CB edge is then put on a vacuum scale using the bulk CdSe electron affinity value ($\chi = -4.95$ eV) (Figure 9a). Due to the photoreactive flexibility of the Ru(bpy)$_3^{2+}$, the ground state and excited state redox potentials for RuPDMAEMA should be considered to determine the full range of processes which may be compatible, with the energies of the QD. The excited state redox potentials were also estimated using the ground state reduction potentials of Ru(bpy)$_3^{2+}$ (Figure 9b) obtained from literature (1.26 vs SCE for Ru(bpy)$_3^{3+/2+}$ and -1.28 vs SCE for Ru(bpy)$_3^{2+/+,}$ reported potentials are in aqueous solution)$^{22,33}$ along with the energy of the emission peak of the RuPDMAEMA ($E_{em}^{0-0}$):

$$E_{1/2}\left(Ru(bpy)_3^{3+/2+}\right) = E_{1/2}\left(Ru(bpy)_3^{2+/+}\right) - E_{em}^{0-0}$$  \hspace{1cm} \text{Equation 11a,b}

SCE, which stands for saturated calomel electrode (E = +0.244 V saturated), is a common reference electrode used in electrochemistry. Converting SCE literature potential values to NHE or SHE (normal or standard hydrogen electrode, E $\approx$ 0.000 or $= 0.000$ V) values and using the NHE or SHE value with respect to a vacuum level or an absolute scale ($E_{ref} = -4.50$ eV), we are able to put together the energies of the QD and redox potentials of the RuPDMAEMA into a relative redox potential energy diagram (Figure 9c). From here, we can assess which photoinduced processes are likely to occur for our particular system based on the relative energy information given above, as well as the spectroscopy data obtained.
Figure 9

Relative Positions of Band-Edge States of CdSe QD and Redox Potentials of RuPDMAEMA

a) Illustration of CdSe QD band-edge energy relative to the bulk. Bulk values taken from literature are shown in green, estimated CdSe QD band-edge values are in red, and the NHE value, $E_{\text{ref}}$, is in blue. All values are relative to vacuum.

b) Schematic diagram showing energy and electron transfer processes for Ru(bpy)$_3^{2+}$ (modified from ref. 31)

c) Combined diagram of the relative energies of CdSe QD band-edge states and RuPDMAEMA redox potential.
A host of various, energy and charge transfer, processes are potentially available to the QD-RuPDMAEMA system and Figure 10 illustrates these potential processes (resulting from single excitations of either the QD or the polymer). Starting with RET, given the spectral overlap of the donor emission and acceptor absorption spectra, energy transfer from the photoexcited QD to the RuPDMAEMA or the photoexcited RuPDMAEMA to the QD are both possible. However, given the small spectral overlap, in the case of RET from the photoexcited QD to the RuPDMAEMA, the process is less likely to occur. In the case of RET from the photoexcited RuPDMAEMA to the QD, there is a fairly good spectral overlap for the process to be plausible. However, RET from the RuPDMAEMA to the QD will result in a quenching of the polymer emission followed by a sensitization of the QD emission, which is not apparent in the steady-state complex formation emission spectra (shows quenching in both peaks). Hence, RET from the polymer to the QD cannot be the dominant process found in the system.

There are two single excitation, charge transfer processes which are likely candidates for this system. The first involves a hole transfer from the QD to the RuPDMAEMA polymer, following QD photoexcitation, and the second involves an electron transfer from the RuPDMAEMA to the QD, following RuPDMAEMA photoexcitation in the system. Based on the energetics of this system, both are just as likely, and despite the mechanism, the final charge transferred state will be the same in either case, with the extra electron residing on the QD CB (QD\(^-\)) and a deficit in electron density on the ruthenium metal ion (\(\text{Ru(bpy)}_3^{3+} \equiv [\text{Ru}^{III}(\text{bpy})_3]^{3+}\)). The charged QD\(^-\) is said to be in a dark non-emissive state because the additional electron in the CB decays through high-efficiency, non-radiative Auger recombination, while the oxidized \(\text{Ru(bpy)}_3^{2+}\) of the RuPDMAEMA polymer will also be in a non-emissive Ru(III) form.\(^{22}\)

Based on these predicted processes, looking at the energetics as well as the experimental data obtained, a clear dominant mechanism still cannot be determined and most likely a combination of these processes may exist for this very complex QD-RuPDMAEMA system. It is important to mention that different mechanisms have been proposed for similar nanocrystal-dye systems, and although the systems look seemingly identical in composition or in construct, the different mechanisms put forth may all be valid. A general mechanism may not be possible for such systems due to the versatility of the QD bandgap. Since every batch of QD synthesized will have slightly different properties from lab group to lab group, even person to person, depending on the recipe and techniques used. So, a slight difference in properties can shift the relative energies of the nanocrystal in relation to the dye, which can considerably change the likelihood of the processes available, hence altering the dominant mechanism of the system. So, it is imperative to consider the relative energies on a case by case scenario to determine which processes will be valid for each particular system.
**Figure 10**

Possible Processes for QD-RuPDMAEMA

Illustration of potential processes, resulting from single excitations of either the QD or RuPDMAEMA, which can take place for the QD-RuPDMAEMA complex. Green and yellow colours represent CdSe QD and RuPDMAEMA (or Ru(bpy)_3^{2+}), respectively.
2.5.5 Solvation & Charge Transfer

Generally, the solvent environment can have substantial effects on the ground and excited state energies of molecules and consequently their optical properties, especially with regards to the excited state and emissive characteristics (Figure 11). The size of these molecular systems are on the same scale as the solvent molecules. Hence, the locally surrounding solvent, provided that they are to some degree polar, can experience polarization effects from the molecule, which enables them to reorganize around the molecule, resulting in an overall energy stabilization of the system. This is one of the origins of the Stokes shift. As the solvent polarity increases, this energy stabilization effect becomes more pronounced and results are reflected in lower emission energies, as long as the fluorophores themselves also exhibit sensitivity to the solvent polarity.

Given the much larger size of QDs, in comparison with molecular systems, and their significantly large dipole moments (~41 D for QD with a diameter of 2.7 nm, compare with solvent dipole moments listed in Table 2)\textsuperscript{107-109}, QDs are expected to induce a very strong polarization on its environment. However, the surrounding solvent molecules may not be able to reciprocate a stabilization effect of a similar magnitude on the system. As a consequence, the bath reorganization energy is expected to be nominal, along with the energy stabilizing role of the solvent.

Analogous to solvent effects on emission properties of molecules, solvation plays a crucial role in charge transfer reactions (i.e. in determining rates of ET). A simple change in solvent polarity can affect the energetics of the donor and/or acceptor, their electronic coupling, and the activation barrier of the reaction, all of which affect the rate, as discussed in section 2.3. However, these effects in conjugate semiconductor nanoparticle and molecular/dye adsorbate systems and on CT processes in these systems have not been adequately addressed. Hence, exploring the scope of solvent dependence on our QD-RuPDMAEMA system will help us to gain general insight on QD-based CT systems.

In this section, we look at solvent contributions on the QD-RuPDMAEMA complex in four solvents, ranging in dielectric properties, using nanosecond TCSPC measurements. Reflecting back on the steady-state PL data obtained during the formation of the QD-RuPDMAEMA complex, we wanted take a closer look at the PL quenching produced from the complex formation. Since the PL decay, set at the RuPDMAEMA PL maximum, is relatively long lived, we turn our attention to the QD emission within the complex.
The photoluminescence decays of both, the QD-PDMAEMA and QD-RuPDMAEMA, complexes were measured in toluene (Tol), tetrahydrofuran (THF), dimethylformamide (DMF), and water at the QD emission (550 nm) using a 456 nm excitation source (Figure 12). The PL decays were fitted using a standard nonlinear, least-squares method with a multi-exponential decay function (Equation 12). The choice of the model for the fit is, to some extent, arbitrary, however it is a typical model used in decay kinetics of QD systems. And for convenience, in our analysis we compare average PL lifetimes ($<\tau>$), which is the average time a molecule spends in the excited state before emitting a photon and returning to the ground state, hence the mean time taken for a photon emission (Equation 13).
The two sets of decays show opposing trends in the calculated average lifetimes (Table 2). The average lifetimes of the QD-PDMAEMA complexes decrease with increasing solvent polarity, given in terms of dielectric constant, from toluene to water. Conversely, the average lifetimes of the QD-RuPDMAEMA complexes increase with the same increase in solvent polarity. 

The QD-PDMAEMA sample, in the absence of the Ru(bpy)$_2^{2+}$ group, is essentially a PDMAEMA passivated CdSe QD, similar to the TOPO passivated QD. Since the PL QY of CdSe QD samples are less than 100% (0.5 to 1%), the lifetime measured is not reflective of the natural radiative lifetimes of the
distribution of QD populations within the sample, but each time constant (τ) represent results from a competition between at least one radiative (k_r) and one non-radiative (k_{nr}) decay rate:

$$\tau = \frac{1}{k_r + k_{nr}}$$

Equation 14

And as mentioned in the first chapter, subsequent to excitation of the CdSe nanocrystal, many processes can be responsible for the relaxation of the innate QD excited state population, which includes radiative and non-radiative bandedge exciton recombination, radiative and non-radiative surface-assisted exciton recombination, and non-radiative Auger recombination. The rates of these radiative and non-radiative processes will affect the PL lifetime of the QD. Since all of the samples were prepared from the same batch of QDs and PDMA polymer, and the same experimental conditions were imposed during these measurements, the main factor for the variations that are observed in the QD-PDMAEMA lifetimes can be attributed to changes in surface trap states with respect to the changing dielectric environment.

<table>
<thead>
<tr>
<th>Solvents</th>
<th>μ (D)</th>
<th>n</th>
<th>ε</th>
<th>&lt;τ_{QD-PDMAEMA}&gt; (ns)</th>
<th>&lt;τ_{QD-RuPDMAEMA}&gt; (ns)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Toluene</td>
<td>0.36</td>
<td>1.496</td>
<td>2.38</td>
<td>25.09</td>
<td>40.20</td>
</tr>
<tr>
<td>THF</td>
<td>1.63</td>
<td>1.407</td>
<td>7.52</td>
<td>20.88</td>
<td>76.24</td>
</tr>
<tr>
<td>DMF</td>
<td>3.82</td>
<td>1.431</td>
<td>38.25</td>
<td>18.64</td>
<td>171.36</td>
</tr>
<tr>
<td>Water</td>
<td>1.85</td>
<td>1.333</td>
<td>80.10</td>
<td>9.39</td>
<td>230.56</td>
</tr>
<tr>
<td>CdSe QD</td>
<td>41</td>
<td>2.67</td>
<td>10.2</td>
<td>&lt;τ_{QD}&gt; (in toluene) = 12.06</td>
<td></td>
</tr>
</tbody>
</table>

Table 2 Calculated average lifetimes of QD-PDMAEMA and QD-RuPDMAEMA complexes in various solvents, along with selected solvent and CdSe QD physical properties.

μ is the permanent dipole moment in debyes, n is the refractive index, ε is the static dielectric constant

For the QD-PDMAEMA, it can be seen that samples in the more polar solvents have faster PL decay rates. It has been shown that surface trap states can quench photoluminescence on short time scales. Hole trappings in CdSe QDs are short-lived and occurs on a picosecond timescale (~3-50 ps), faster than Auger decay, and can result in both radiative and non-radiative recombination (although band edge electrons and trapped holes are known to radiatively recombine, and have a PL spectrum that overlaps the band edge states – thus, hole trapping does not directly quench the PL, but rather increases the radiative lifetime).

Electron trappings are long-lived and occurs on a ns timescale (~1-100 ns), and typically decay via non-radiative recombination (electron trapping processes are competitive with PL). This insinuates that as higher dielectric solvents are used, more surface traps seem to be available, trapping the excited electron or hole and reducing the excited state population, hence decreasing the average PL lifetimes. The question remaining is, what is causing the increase in availability of surface traps? This question directs us to take a closer look at surface passivation of the QD. During the experiment, it had been noted that PDMAEMA or RuPDMAEMA coated QDs seemed to be most stable in toluene and the least in water.
Figure 13

Photoluminescence Decay of QD-PDMAEMA and QD-RuPDMAEMA in Various Solvents

The PL decay profiles of QD-PDMAEMA (blue background with solid solvent lines) and QD-RuPDMAEMA (red background with dotted solvent lines) in toluene (yellow), THF (cyan), DMF (pink), and water (green) are shown. The top two plots are grouped by sample, while the bottom four plots are grouped by solvent. The inset is a close-up of the fast decay components.
Varying degrees of aggregation in the samples, in vacuum sealed tubes, were observed over time, in DMF and water. Sample aggregation in water occurred after approximately several weeks after sample preparation, while sedimentation was noticed for DMF approximately a few months after. From this observation, we propose that as higher dielectric solvents are used, the attractive polymer-solvent interaction diminishes, which can cause the polymer to shrink or collapse, reducing the number of available binding sites. With a reduction in the number of binding sites, the likelihood of adsorption onto the QD surface decreases and a poorer surface passivation results, hence exposing more surface traps (Figure 14).

The decay profiles of the QD-RuPDMAEMA sample interestingly show the presence of a fast and slow decay component. These components signify that at least two distinctive mechanisms are responsible for the deactivation of the excited state. The fast component (initial 50 ns) shows decay trends similar to the QD-PDMAEMA complex, and is likely resulting from higher exposure to surface traps with respect to the solvent polarity. Aside from the surface trap decay trend, these fast components show a subtly faster PL decay in relation to the increasing solvent polarity, compared to the QD-PDMAEMA samples (Figure 13, insets). The additional faster decay, or quenching, may be attributed to the supplementary charge transfer reaction, occurring between the QD and the polymer/dye in the system. Since, the nanosecond measurements cannot resolve these ultrafast dynamics (below ~22 ns), we are most likely capturing the aftermath of the CT reaction.

The additional slow component (above 100 ns), defining the tail of the decay profile, is quite interesting, and shows opposite solvent trends in comparison with the fast component. The slow component seems like it may be due to a delayed radiative recombination resulting from a back electron transfer (BET) reaction, which is the reversal of the ET process restoring the donor and acceptor back to their original oxidation states. Since our QD-RuPDMAEMA system is not designed for subsequent reactions after charge transfer, a BET process will inevitably occur in the system. However, to be able to observe BET at the QD emission, it would seem that a charge transfer processes involving a hole transfer from the QD to the RuPDMAEMA polymer (induced by QD photoexcitation in the system), followed by BET from the QD to the polymer, replacing the hole, hence restoring the exciton on the QD, allowing for subsequent radiative charge recombination (resulting in a delayed emission from the QD represented by the slow component in the PL decay profile), may be a possible mechanism that would result in this observation. And if the slow component does indeed represent the BET reaction, the solvent trend seen for this component would seem consistent with expectations. Once CT has occurred, polar solvent molecules are able to reorganize around the charge transferred (ionized) system to accommodate for the new charge
distribution, stabilizing the products of the ET reaction. In order for BET to occur, sufficient energy is required to overcome this solvent-formed stability. In cases of higher polarity solvents, a greater solvent stabilization will result, which will likely further impede the rate of BET.

In order to better understand the solvent trends, with respect to the CT reaction observed in the complex, and to tie in some of the concepts that have been discussed in this chapter, we turn to the Marcus theory to model our system. A slightly tailored version of the Marcus theory will allow us to obtain information regarding the energetics and kinetics of the photoinduced ET and BET reactions with respect to the various solvents utilized. For the purpose of obtaining $\Delta G^{\circ}_{\text{ET}}$, initial and final states of the ET reaction need to be defined. And, as stated above, an exciton dissociation pathway, involving a hole transfer from the QD to the Ru(bpy)$_3^{2+}$ group following QD photoexcitation will be used based on the interpretation of the QD-RuPDMAEMA PL decays.

Values for the free energy difference of the ET reaction ($\Delta G^{\circ}_{\text{ET}}$, $\Delta G^{\circ}_{\text{BET}}$) (also illustrated in Figure 3), reorganization energy ($\lambda$), barrier energies for ET and BET processes ($\Delta G^{\dagger}_{\text{ET}}$, $\Delta G^{\dagger}_{\text{BET}}$), as well as rate constants ($k_{\text{ET}}$, $k_{\text{BET}}$) have been calculated (Table 3) using Equations 15-19:

$$\Delta G^*_{\text{ET}} = e(E^*_{\text{OX}} - E^*_{\text{RED}}) - E_{00}$$  \hspace{1cm} \textit{Equation 15}
\[
\lambda_o = \frac{1}{4\pi\varepsilon_o} \times \frac{e^2}{2} \left[ \frac{1}{r_{Ru(bpy)}^3} \left( \frac{1}{n^2} - \frac{1}{\varepsilon} \right) - \frac{1}{2R} \left( \frac{n_{QD}^2 - n^2}{n_{QD}^2 + n^2} \times \frac{1}{\varepsilon_{QD} - \varepsilon} - \frac{1}{\varepsilon_{QD} + \varepsilon} \times \frac{1}{\varepsilon} \right) \right]
\]

Equation 16

\[
\Delta G_{ET}^+ = \frac{\left( \Delta G_{ET}^o + \lambda \right)^2}{4\lambda}
\]

Equation 17

\[
\Delta G_{BET}^+ = \Delta G_{BET}^o + \Delta G_{ET}^+ = \left( - \Delta G_{ET}^o \right) + \Delta G_{ET}^+
\]

Equation 18

\[
k_{ET(BET)} = \frac{2\pi}{\hbar} |H_{DA}|^2 \frac{1}{\sqrt{4\pi\lambda k_B T}} \exp \left( \frac{-\Delta G_{ET(BET)}^o}{k_B T} \right)
\]

Equation 19

where \(E^o_{OX}\) and \(E^o_{RED}\) are the oxidation and reduction energy of the electron donor and acceptor pair, and \(E_{00}\) is the excitation energy put into the system to form the initial photoexcited state of the QD which is equal to the energy from the absorption edge of the first exciton band. \(R\) and \(r_{Ru(bpy)}^3\) represent the donor-acceptor separation distance (or the mean ET distance) and the radius of the molecular adsorbate (0.58 nm for Ru(bpy)_3^{2+})\(^{116}\), respectively. The refractive index \(n\) and static dielectric constants \(\varepsilon\) refers to the respective solvents used, unless explicitly specified via subscript (i.e. QD). \(|H_{DA}|\), the electronic coupling between the initial and final states, is on the order of 0.05 eV\(^{117}\).

The ET process was modeled using non-adiabatic Marcus theory (weak donor-acceptor coupling). The solvent reorganization energy, at the planar semiconductor-liquid interface, is calculated using a dielectric continuum model given by the dielectric constant of the solvent and semiconductor.\(^{18,26,27}\) The inner sphere (\(\lambda_i\)) contributions to the reorganization energy is expected to remain relatively constant over the different solvents. Hence, the overall reorganization energy was calculated using the outer sphere (\(\lambda = \lambda_o\)) or structural solvent response contributions only (i.e. distortions of the solvent nuclei).

<table>
<thead>
<tr>
<th>Solvents</th>
<th>(\lambda_o) (eV)</th>
<th>(\Delta G_{ET}^o) (eV)</th>
<th>(k_{ET}) (1/s)</th>
<th>(\Delta G_{BET}^o) (eV)</th>
<th>(k_{BET}) (1/s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Toluene</td>
<td>0.04</td>
<td>3.3</td>
<td>(1.4 \times 10^{41})</td>
<td>0.75</td>
<td>(2.8 \times 10^{7})</td>
</tr>
<tr>
<td>THF</td>
<td>0.41</td>
<td>0.07</td>
<td>(1.3 \times 10^{13})</td>
<td>0.30</td>
<td>9.3 \times 10^{8}</td>
</tr>
<tr>
<td>DMF</td>
<td>0.52</td>
<td>0.02</td>
<td>(5.5 \times 10^{13})</td>
<td>0.32</td>
<td>4.0 \times 10^{8}</td>
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<tr>
<td>Water</td>
<td>0.62</td>
<td>0.01</td>
<td>(9.4 \times 10^{13})</td>
<td>0.34</td>
<td>1.7 \times 10^{8}</td>
</tr>
</tbody>
</table>

ET and BET activation energies and rate constants are calculated using \(\Delta G_{ET}^o = -0.75\) eV and \(\Delta G_{BET}^o = 0.3\) eV.

It can be seen from the calculated values that as solvent polarity increases, \(\lambda_o\) also increases. Since higher dielectric solvents exhibit greater sensitivity to the changes in charge distribution of a system which it surrounds, such as that resulting from an ET reaction, higher energies are associated with the structural reorientation of these solvent molecules. So, in comparison of the \(\lambda_o\) and \(\Delta G_{ET}^o (-0.75\) eV) values, this
particular ET reaction is considered to be in the Marcus inverted region \((\lambda < |\Delta G^\circ|)\) for all of the solvents. A greater difference between \(\lambda_o\) and \(|\Delta G^\circ_{ET}|\), in the inverted region, will cause the reaction rate to fall, which can be seen in our calculated \(k_{ET}\) values with water having the highest ET rate and toluene having a negligible rate (N.B. calculated rates will be overestimated, leading to faster rates, using this model in the inverted region), and this trend is also illustrated in the fast component of our QD-RuPDMAEMA PL decays. Water is a highly polar solvent, and it has a high \(\lambda_o\) very close to \(|\Delta G^\circ|\). As \(\lambda_o\) and \(|\Delta G^\circ_{ET}|\) get closer together in values, the reaction heads toward the Marcus optimal region, characterized by a barrierless \((\Delta G^\ddagger_{ET} = 0)\) transition, resulting in an optimal ET rate. Toluene, on the other hand, is a non-polar organic solvent, and thus has a very low \(\lambda_o\). So, it is expected that it will aid very little in the ET process (as can be seen in the relatively high \(\Delta G^\ddagger_{ET}\) and \(\Delta G^\ddagger_{BET}\) values), resulting in unusually slow calculated rates which do not seem to fit any trend (most likely since the model breaks down at this limit and cannot satisfactorily predict results). This is also reflected in the relatively similar PL decay curves obtained for the QD-RuPDMAEMA and QD-PDMAEMA in toluene.

For calculation of BET rates, Equation 18 was first employed to obtain values for the BET activation energy \((\Delta G^\ddagger_{BET})\), however with the reaction being in the Marcus inverted region, the calculated BET rates also came out with the same solvent trend as the forward ET rates, which is contrary to the slow component trend of the QD-RuPDMAEMA PL decays. The incongruence between the model and the experimental results seem to indicate that certain factors have been overlooked. As seen in the experimental results of the QD-PDMAEMA samples, solvent effects on QD passivation and surface states may play an important role in ET dynamics of the QD-RuPDMAEMA system. Since hole trapping events seem to occur relatively easily and rapidly in CdSe QDs,\(^{103}\) it seem to provide a rationale for surface-assisted hole transfer back from the RuPDMAEMA to the QD. By applying this rationale and decreasing the Gibbs free energy difference for the BET reaction, a suitable solvent trend could be produced which complemented our experimental data. Using a lower \(\Delta G^\circ_{BET}\) value, lowers the driving force and pushes the BET reaction into the Marcus normal region \((\lambda > |\Delta G^\circ|)\), except for toluene, which increases the BET rates for the lower dielectric solvents.

Putting things together, in the case of the forward ET reaction, which is in the Marcus inverted region, a faster rate is observed for higher dielectric solvents since \(\lambda_o\) is shifted closer to \(|\Delta G^\circ_{ET}|\), allowing for a better overlap of the reactant/product wavefunctions at the crossover point (where quantum mechanical contributions will come into play), resulting in an easier ET transition (seen in the \(\Delta G^\ddagger_{ET}\) values).\(^{118}\) In the case of the BET reaction, in the normal region, due to the high \(\lambda_o\) associated with the higher dielectric solvents, this factor will dominate over \(\Delta G^\circ_{BET}\) (in regards to \(\Delta G^\ddagger_{BET}\)), thus more energy is required to
reorganize the higher dielectric solvent molecules, especially once the ET reaction has taken place. The polar solvent molecules reach an equilibrium with the product state and are now relatively more stabilized compared to the reactant state, so the system resists BET and sufficient energy is required to overcome this stability (seen in the $\Delta G^{\ddagger}$BET values). This product stabilization effect becomes more pronounced as the solvent polarity increases, and slower BET rates result for the higher dielectric solvents. Obviously, the model is not a perfect fit and there are still many significant factors that have been overlooked in this simple model (i.e. conformation of adsorbate, coulombic attraction and solvation factors in the calculation of $\Delta G^\circ$), along with a potentially large margin of error associated with accurately predicting $\Delta G^\circ$ due to ambiguities in the actual energy levels and reorganization processes of the intrinsic surface states, and so forth. However, it manages to explain some of the basic trends that we have observed in our experimental data.

Through this solvation experiment we were able to demonstrate the complete ligand exchange and association of the PDMAEMA or RuPDMAEMA polymer with the QD through the solubility of the complex in an extensive range of dielectric solvents (which were not possible with the TOPO passivated QD). This allowed us to explore the changes in surface passivation of the QD, hence PL lifetime, with respect to the various solvents. With the addition of the electro-active component in the polymer, we were also able to see that forward and backward charge transfer processes, in combination with trap states, can alter the ideal kinetic scheme of QD photophysics, introducing intricate, alternative pathways, which can extend the overall excited state lifetime of the QD, especially with respect to various solvent polarities. And since QD-based CT systems are not normally soluble and have not been studied in such an extensive range of dielectric solvents, this gave us a unique opportunity to look at the degree of influence different dielectric media has on such systems.
2.6 Conclusion

In this chapter, we explored the properties of CdSe nanocrystal complexes passivated with a special electro-active block copolymer, RuPDMAEMA. The synthesis of the RuPDMAEMA polymer and the preparation of QD-RuPDMAEMA complexes, via a simple ligand-exchange procedure, were presented. Characterization of the QD-polymer complex was pieced together using STEM images, UV-Vis absorption and fluorescence spectroscopy, fluorescence decay, and transient absorption measurements. It was found that adsorption of the RuPDMAEMA polymer onto the QD surface produced PL quenching, at the respective emission sites of the QD and RuPDMAEMA, in the complex. It was deduced that the quenching was most likely due to a charge transfer reaction between the QD and polymer. Further characterization, and possible determination of the mechanism, of the CT reaction through fluorescence decay and TA measurements lead to fairly complex interpretations with mixed and opposing results. Through some refinement, the relative energy level positions of the QD and RuPDMAEMA were calculated to reflect upon which photoinduced processes of the QD-RuPDMAEMA complex were available. This lead to the conclusion that several mechanisms could possibly come into play for our system.

The effect of solvent polarity on the QD-RuPDMAEMA system was explored using nanosecond TSCPC measurements to gain further insight into QD-based CT systems. Firstly, we were able to verify the adsorption of the RuPDMAEMA polymer onto the QD surface through the QD solubilization in a wider range of solvents, than that was available to the TOPO-QD. It was found that QD-RuPDMAEMA PL decays showed the presence of two decay components, fast and slow. It was suggested that the fast component may have had contributions from surface trap and ET effects found in the system, while the unexpected slow component may have been a result of BET, resulting from the initial hole transfer from the QD to the RuPDMAEMA, a possible CT mechanism for the QD-RuPDMAEMA system. From this experiment, it was found that solvent polarity played an important role on surface passivation, hence exposing surface trap states, of the QD, ultimately influencing the PL lifetime of the complex. As well, it was found that with the addition of the ET and BET reactions, the overall excited state, or more specifically radiative, lifetime of the QD could be lengthened and also greatly affected by the polarity of the solvent. These results were put into perspective in conjunction with the Marcus theory of electron transfer, illustrating the high complexity and providing general insight on QD-based CT systems and the effects of solvent polarity.
2.7 References


Chapter 3

Photon Echo Spectroscopy: 2DPE Electronic Spectroscopy and How It Relates to 3PEPS
3.1 Introduction

From the previous chapters, it can be seen that spectroscopy is the quintessential tool for describing material properties and interactions. So far, with the exception of transient absorption, we have been dealing with mainly traditional spectroscopy, for which the only observables are wavelengths/frequencies, amplitudes, and lineshapes (and in some cases time). Even though a good amount of information can be obtained, especially with cleverly designed experiments, traditional spectroscopy can be somewhat limited and ambiguous when it comes to targeting specific states or interactions, and interpreting data extracted from the simple set of observables mentioned above. So, we are prompted to look for new ways of obtaining more control over the types of experiments we perform and the studies we undertake, which leads us to the world of nonlinear spectroscopy.

There are essentially two main reasons to use nonlinear optical spectroscopy. First, as mentioned, nonlinear optical spectroscopy enables us to measure and study material properties that cannot be addressed by linear optical spectroscopy. The use of varying number of pulses in specific sequences, wavelengths, polarizations, and geometries, typical in many nonlinear experiments, allow for more freedom in the choice of molecular states and interactions that can be probed, allowing for a wealth of diverse information. Also, nonlinear experimental techniques allow for true bulk interaction of the light with the material, and the sometimes unsolicited yet predominant surface effects can be minimized. Secondly, nonlinear spectroscopy allows us to obtain time-resolved information at a much higher resolution and sensitivity compared to linear spectroscopy. For example, due to the ultrafast nature (nanoseconds to femtoseconds) of the excitation laser pulses employed in nonlinear experiments, information that is normally masked under the lineshapes of absorption and fluorescence measurements can now be recovered.1,2

In this chapter, an informal review of photon echo spectroscopy is presented, focusing on two different methods, the three-pulse photon echo peak shift (3PEPS) and the two-dimensional photon echo (2DPE) optical spectroscopy, classic to this genre. A brief discussion is presented on system-bath interactions, how they relate to spectroscopy, and how photon echo spectroscopy, more specifically 3PEPS, can be used to obtain a valuable parameter called the transition frequency correlation function, which unlocks a fundamental element capable of complete description of a system’s optical response. The chapter is concluded with a demonstration on how 3PEPS data can be extracted from 2DPE measurements using the 2DPE data obtained for a model system, Rhodamine 6G.
3.2 Ultrafast Nonlinear Optical Spectroscopy for Dummies

From the viewpoint of materials science, occasionally acquainted terms and acronyms, such as ultrafast laser spectroscopy, nonlinear optical response, third-order polarization, four-wave mixing, photon echo spectroscopy, 3PEPS experiment, 2DPE electronic spectroscopy, can be pretty confusing. It is difficult to determine how these terms could relate to each other, if related at all, and where it all begins.

The field of ultrafast nonlinear optical spectroscopy is vast and characterized by a rather large number of highly complex and sensitive, yet powerful, experimental techniques. Different names and terminologies are used to describe similarly related or even identical spectroscopies, techniques, or processes – hierarchically related expressions which are sometimes interchangeably used – which can lead to a bit of confusion. For example, the widely-used transient absorption spectroscopy, discussed in chapter 2, could also be called “time-domain multiplexed two-beam heterodyne four-wave mixing spectroscopy”, which is a form of pump-probe, time-resolved, nonlinear and four-wave mixing spectroscopy. Because we are familiar with the basic principles of the experiment and aware of some of the terms, we can generally make sense of how things fit together, however it is common for a non-specialist to feel a sense of disconnection when stepping into this advanced realm of spectroscopy. Thus, in this section, we will breach this topic with a brief introduction to where it all begins and familiarize ourselves with some of the basic concepts and terms used in this chapter.

3.2.1 Nonlinear Optical Behaviour

To begin, we consider the interaction of light with matter. From a physicist’s perspective, this interaction essentially reduces to a dielectric medium being subject to an electric field, which is followed by phenomena such as light absorption, emission, refraction, reflection or scattering, diffusion, etc. All of which are characterized by material properties, which are dependent on the wavelength of the incident light, however under “normal” circumstances (i.e. weak light intensities), are independent of intensity. Most of these associations can be expressed as linear relationships. For example, Beer's Law is the most widely recognized linear relationship that deals with the absorption of light by a material.

With the invention of lasers, the previously unattainable concentrated light intensities can now be produced. These high intensity optical pulses have electric fields that are comparable to or larger
(typically $10^8$ V/m) than those existing between the charges (i.e. intra-atomic electric fields caused by nuclei and electrons) constituting the matter, such that they can modify the optical properties of matter. Hence, material properties begin to show a dependence on incident light intensity, and a nonlinear relationship is formed in the limit of strong electric fields.

I find that it is sometimes difficult to find everyday examples or analogies to better understand and explain (to family and friends at the dinner table, for instance) abstract or fundamental phenomena or concepts in physics, such as nonlinear behavior. So, I was delighted when I ran into a simple example describing the nonlinear phenomenon as the result of cranking up the volume of a sound system, almost to the point of blowing your speakers. It was stated that when the volume is raised to levels exceeding the capacity of the system’s audio speakers, sound distortions are created, where the response of the speakers no longer follow the driving voltage. The incoherently loud but muffled vocalizations emanating from cheap fast food drive-thru speakers or the irritatingly high-pitched screeches coming from an overloaded electric guitar amplifier are well-known examples of such distortions, which are created from new frequencies generated through the sum and difference of the original sound frequencies, as well as the changes in their amplitudes.

So, getting back to nonlinear optical behaviour, as light is introduced to a material, the electric field of the light induces an oscillating electric dipole moment, called a polarization, in the electrons/ions of the material. The induced polarization (i.e. the material response), depending on the intensity of the incident electric field, can be either linear or nonlinear in nature. Subsequently, this induced linear or nonlinear polarization then goes on to interact with the incident electric field giving rise to the various linear or nonlinear processes and effects. The behaviour of the electrons/ions in the material may be described using a harmonic oscillator model, starting with the electron/ion placed in a potential well at the equilibrium position. So, let us now consider the two scenarios.

For a relatively weak incident electric field, the resulting small oscillations or dipole displacement of the electron/ion will be harmonic about the equilibrium position. This, in turn, will cause a small induced polarization ($P_{\text{ind}}$) of the medium at the corresponding frequency of the incident light, and a linear relationship is formed between the polarization and the weak incident electric field ($E$):

$$P_{\text{ind}} = \varepsilon_0 \chi^{(1)} E(\omega)$$

Equation 1

where $E$ is a function of angular frequency ($\omega$), and $\chi^{(1)}$ is the linear susceptibility. This susceptibility parameter is related to the optical response of the medium, specifically the complex refractive index ($\tilde{n}$) or dielectric constant ($\varepsilon$), at the given frequency:
\[ E_{(\omega)} = \tilde{n}^2(\omega) = 1 + 4\chi^{(1)}(\omega) \]  \hspace{1cm} \text{Equation 2}

The induced oscillating polarization of the molecules in the medium will act as a source of radiation and emit a new electric field proportional to the oscillations. This electric field produced from the material will then go on to interact with the originally applied weak electric field, and yield the classic low power linear optical phenomena. Thus, all of the material’s optical properties and corresponding phenomena are associated with this polarization as defined in Equation 1 through the relationship of \( \chi^{(1)} \) and Equation 2 in the weak electric field limit.\(^1\)

For strong incident electric fields, the displacement is no longer harmonic and anharmonic oscillations begin to occur. In addition to the linear relation, the induced polarization now depends on higher powers of the electric field (i.e. nonlinear relationship), with higher order nonlinear susceptibilities also coming into play. The polarization expression can be expanded, to an infinite power/Taylor series of the electric field, to include the nonlinear factors:

\[
P_{\text{ind}} = P_L + P_{\text{NL}}
\]

\[
= P^{(1)} + \left( P^{(2)} + P^{(3)} + \ldots \right)
\]

\[
= E_0 \left[ \chi^{(1)} E_{(\omega)} + \left( \chi^{(2)} E_{(\omega)}^2 + \chi^{(3)} E_{(\omega)}^3 + \ldots \right) \right]
\]

\[
P_{(n)}^{(n)} = E_0 \chi^{(n)}(\omega_{\omega_1,\omega_2,\ldots,\omega_n}) E_{(\omega_1)} E_{(\omega_2)} \ldots E_{(\omega_n)}
\]  \hspace{1cm} \text{Equation 3}

\[ P_{(n)} = \chi^{(n)}(\omega) E_{(\omega_1)} E_{(\omega_2)} \ldots E_{(\omega_n)}
\]  \hspace{1cm} \text{Equation 4}

where \( P_L (= P^{(1)}) \) represents the linear (first-order) polarization and \( P_{\text{NL}} (= P^{(2)} + P^{(3)} + \ldots) \) represents the higher order nonlinear polarization given by the round bracketed terms in Equation 3, and \( P^{(n)} \) and \( \chi^{(n)} \) represent the nth-order polarization and susceptibility, respectively (where \( n = 1, 2, \ldots, \) an integer value). The additional higher order susceptibility terms (\( \chi^{(2)}, \chi^{(3)}, \ldots \)) account for higher order hyperpolarizability factors (\( P^{(2)}, P^{(3)}, \ldots \)), which give rise to nonlinear optical effects. The general form of the nth-order polarization is given in Equation 4, where the previous description of the polarization’s dependence on the nth-power of the electric field (\( E_{(\omega)}^n \)) in Equation 3, has been replaced by a dependence on the n number of material interactions with n distinct external electric fields (\( E_{(\omega_1)} E_{(\omega_2)} \ldots E_{(\omega_n)} \)). This is an important thing to note. Higher light intensities mean increased number of electric fields, thus when a strong intensity electric field interacts with matter, this can be thought of as multiple electric fields all interacting with the material.\(^4-7\)

The susceptibility terms are tensors which define the relationship between the polarization(s) of the applied electric field(s) and the induced polarization of the material, and reflect the symmetry of the material. For example, second-order nonlinear susceptibility of a centrosymmetric or isotropic material is
zero \( \chi^{(2)} = 0 \). This means that molecules with a centre of inversion or inversion symmetry (only found in certain crystals) or random orientation (i.e. liquids, glasses, polymers) have no second-order nonlinear susceptibility. Finding materials that are non-centrosymmetric and non-isotropic can be difficult, thus the application of second-order nonlinear optical spectroscopy is rather limited. On the other hand, all materials have a third-order nonlinear susceptibility term, hence show third-order nonlinear response, which is important for many applications.

Second-order nonlinear susceptibility \( \chi^{(2)} \) is responsible for second-order polarization and nonlinear effects such as second harmonic generation (SHG) (N.B. for a given single frequency of incident light, there will be a material response at the fundamental frequency and successive harmonics, and generation of light at these higher harmonic frequencies is known as harmonic generation) and the electro-optic effect (the modification of the refractive index of a material caused by an externally applied electric field). While the third-order nonlinear susceptibility \( \chi^{(3)} \) is responsible for third-order polarization and nonlinear effects such as third harmonic generation (THG) and the optical Kerr effect (another type of electro-optic effect, attributed to third-order susceptibility materials).

Whether it is second-order or third-order or higher order susceptibilities or combinations of, all materials have a nonlinear response, which only become evident at high light intensities (i.e. increasing number of electric field interactions with the material). The magnitude of the response, hence the size of the nonlinear susceptibility, will be determined by the microscopic structure of the material. The specific contributions to the nonlinear response will depend on the nature of the material, the parameters of the incident electric field, and the temporal evolution of the interactions.

There are a host of nonlinear optical processes and effects, defined by the various order nonlinear susceptibilities, resulting in a fairly large and complex field of nonlinear optical spectroscopy and associated experimental techniques. Since an electric field is characterized by many parameters (i.e. amplitude, frequency, wavevector, phase, polarization, arrival time, etc.), the number of different combinations of electric field parameters along with the multiple light-matter interactions, can lead to a rather large number of possible experiments, explaining the vast field. We will not be plunging into the depths of the various processes and intricate experiments, but this is a good starting point for understanding the differentiation of nonlinear optical spectroscopy.
3.2.2 Third-Order Nonlinear Polarization and Four-Wave Mixing

The third-order nonlinear polarization ($P^{(3)}$) of a material is given in Equation 5 and is dependent on the interaction of the three external electric fields ($E(\omega_1)E(\omega_2)E(\omega_3)$) with the material. The induced nonlinear polarization forms the fourth electric field, generated by the mixing (i.e. any combination of the sum and difference frequencies, $\omega = \omega_1 \pm \omega_2 \pm \omega_3$) of the first three incident fields (by which momentum and energy are conserved). Third-order nonlinear effects involve the interaction of all four electric fields, hence called four-wave mixing effects (or four-photon parametric interaction), leading to a large number of observations, such as stimulated fluorescence, amplification, Raman and Brillouin scattering, two-photon absorption, signal generation in pump-probe experiments, Kerr effect related phenomena (including self-modulation and self-focusing), etc. Since all materials show a third-order nonlinear response, four-wave mixing can take place in any media.4-7

$$P^{(3)}(\omega_1, \omega_2, \omega_3) = \varepsilon_0 \chi^{(3)}(\omega_1, \omega_2, \omega_3) E(\omega_1) E(\omega_2) E(\omega_3)$$  \hspace{1cm} \text{Equation 5}$$

Not only is four-wave mixing an effect of third-order polarization, it is also a type of third-order nonlinear spectroscopy, as well as a technique for nonlinear experiments. Four-wave mixing spectroscopy is concerned with investigations of various third-order nonlinear effects and phenomena specific to a given material, created by the interaction of three external electric fields in a material, generating a fourth signal field which “counter-interacts” with the applied three fields. However, this does not imply that three different laser beams are required for the experiment. For example, as mentioned earlier, transient absorption, which is a pump-probe experiment, is conceptually the simplest form of four-wave mixing nonlinear spectroscopy. It only uses two beams (a pump and probe) but involves a double interaction of the material and the pump/excitation beam, hence giving the effect of three applied electric fields. Pump-probe, transient grating, photon echo, and coherent simulated Raman are some common experiments (or sub-sub-genre spectroscopies) that belong to four-wave mixing spectroscopy.

This brings us to four-wave mixing techniques. Two beam techniques have a drawback, in that they do not allow for individual control of the polarization, frequency, direction, etc. of each wave. This limitation can be evaded with the use of three distinct beams and the general four-wave mixing technique, also known as the transient grating (TG) technique.8-12 Now, there are many ways to view concepts in nonlinear spectroscopy. Depending on the person you were seeking to gain advice or whichever journal paper you picked up for enlightenment, you may get very different technical, theoretical (physical or mathematical), and conceptually dense explanations of the same experiment. Although hardcore nonlinear
spectroscopists seem to love talking about the technical aspects of how their experiment was uniquely and brilliantly set up, how much power their laser is capable of generating, yielding unprecedented short pulse durations, and what awesome optics were employed in the making – and some may only speak in acronyms, numeric parameters, and mathematical functions – coming from a background in material science, with an obvious language barrier issue in place, an intuitively conceptual explanation, combining simple model examples from technical and theoretical aspects, not bogged down with equations, is preferred if such a thing exists, granted that it may not be the most accurate of depictions. Nonetheless, four-wave mixing technique can be explained in two ways.¹³

First is the grating formalism, which involves a two-step process for signal (fourth electric field) generation. When a material is excited with two laser pulses of the same wavelength ($\lambda_{\text{pump}}$), intersected at
an angle of 2θ, the two concentrated pump pulses will constructively and destructively interfere to generate a “transient” or temporary spatially distributed area (interference pattern) of low and high intensities/amplitudes of light on the sample (Figure 1), this is called a spatial intensity grating (analogous to the double slit diffraction experiment). The grating will have a fringe spacing of Λ (Equation 6) in the material. When a strong electric field (i.e. high intensity of light) strikes a material, reorientation of the material’s electric charges and even entire molecules (if it possesses a permanent or induced dipole moment) result along the electric field. This electronic and nuclear reorientation induced in the material will subsequently change the way light or radiation propagates through the material (i.e. the material’s refractive index, more specifically the complex refractive index, is altered). This is known as the optical Kerr effect (OKE).14-17 So, the interaction of the low and high intensities of light (i.e. the two pump pulses) on the sample will create areas of varying refractive indices within the sample, which is referred to as birefringence, since the material is no longer isotropic. A thing to note is that the real part of the complex refractive index is related to the absorbance, so the spatial distribution which is formed is essentially due to changes in both the material refractive index and the absorbance (which equates to a phase and amplitude grating). Once the grating has been established, a third laser pulse, at some probe wavelength (λ_{probe}), hits the grating at a Bragg angle (θ_B) (Equation 7). The Bragg angle allows for most of the light to be transmitted by the material, while a small portion of it propagates through the medium (i.e. the formed grating) and is diffracted (the diffracted pulse is phase shifted from the original probe pulse by π/2). In order for the diffraction efficiency to be as high as possible, it is important that all pulse angles are kept precise and for the third pulse to come in at this Bragg angle, so that reabsorption of the diffracted beam by the sample is kept to a minimum. The diffracted pulse, carrying the refractive index (hence phase and amplitude) information associated with the material, forms the signal, which is then collected. If the probe wavelength equals the pump wavelength, the technique is called degenerate four-wave mixing or TG.4-7

\[
\Lambda = \frac{\lambda_{\text{pump}}}{2 \sin \theta_{\text{pump}}} \quad \text{Equation 6}
\]

\[
\theta_B = \arcsin \left( \frac{\lambda_{\text{probe}}}{2\Lambda} \right) \quad \text{Equation 7}
\]

Since OKE is an effect displayed by χ^(3) materials, all materials show OKE to varying degrees. However, OKE is not the only factor which can contribute to changes in the optical properties of an isotropic material. There can be many other processes occurring in the material which can also lead to an intensity-dependent change in the refractive index, ultimately influencing the generated signal. The basic processes
\[ \Delta \tilde{n} \text{: When light passes through a material, a portion of it will always be absorbed, given by } \tilde{n} = n + i \kappa. n \text{ is the real refractive index associated with the phase speed of the propagating electric wave and } \kappa \text{ is the attenuation constant, forming the imaginary part, associated with the amount of light which will be absorbed by the material (} \kappa \alpha \text{ absorbance).} \]

\[ \Delta n \text{: The change in the refractive index has several origins, given by the expression } \Delta n = \Delta n_p + \Delta n_i + \Delta n_r. \]

\[ \Delta A \text{: The change in absorbance is basically due to changes in concentration, } \Delta C, \text{ hence population changes, } \Delta n_p. \]

\[ \Delta n_p \text{: The optical Kerr effect (OKE) is a type of electro-optic effect particular to } \chi^{(2)} \text{ materials, where a change in the refractive index of a material results from the interaction with a strong electric field (} \Delta n = \lambda K \text{, where } K \text{ is the Kerr constant). The change in refractive index due to the OKE has two contributions } (\Delta n_p = \Delta n_p^e + \Delta n_p^i). \]

\[ \Delta n_i \text{: The change in refractive index due to the density changes has two contributions, given by } \Delta n_i = \Delta n_i^p + \Delta n_i^t. \]

\[ \Delta n^p, \Delta n^i \text{: The propagation of an intense light beam in a material is followed by orientation of the electric charges along the applied electric field (i.e. polarization of electrons). In some cases, if the molecules have a permanent or an induced dipole, they can reorient along the field (i.e. nuclear response). After electronic and nuclear reorientation, the material becomes birefringent (no longer isotropic), since the refractive index near the applied electric field will be different from the rest of the material. Electronic polarization will occur instantaneously compared to slow nuclei reorganization, which also depend on temperature.} \]

\[ \Delta n^e \text{: Electrostriction is the process in which material density increases in the region of a high electric field. The electric field induces polarization of the molecules, resulting in intermolecular attraction, leading to a local increase in pressure, followed by compression of the material along the field direction. This effect is relatively slow and small.} \]

\[ \Delta n^t \text{: The thermo-optic response is due to absorption of the applied electric field by the material, in which a fraction of the excitation energy is dissipated in the form of heat. The warming of the material (i.e. changes in sample temperature) induces a series of processes (including a change in local density) leading to a spatial and temporal change in the refractive index of the material. The thermo-optic response is very slow, but the effect is cumulative, and thus it can become a dominant mechanism. The temperature dependence of a material’s refractive index is mainly due to } \Delta n^t. \]
responsible for intensity-dependent changes in the optical properties of an isotropic material are outlined in Figure 2. Changes in the phase and amplitude gratings can be broken down into various proportions and combinations of these elementary processes. What distinguishes these processes are essentially timescales. OKE effects can take anywhere between 1 to 100 fs (instantaneous electronic polarizations versus slow nuclei reorganization), electrostrictive processes occur on the timescale of nanoseconds, while thermo-optic effects can take up to several microseconds. Our entire discussion in this chapter, up to now, has focused on electronic nonlinearities and will continue to be the main focus for the remainder; however to gain a more comprehensive picture, it is useful to understand the basic factors affecting the generated signal.

The second explanation involves the nonlinear optics formalism. The technical aspects stay the same, however the signal generation is approached mathematically. Since conservation of energy and momentum are conditions that need to be strictly met in the mixing of the incident fields and generation of the signal, the frequency and wavevectors, parameters which correspond to energy and momentum of electric fields in light, need to be combined in ways where their sum and difference are cancelled out. This means that the frequency of all of the input electric fields must match the frequency of the output signal field (Figure 3a, \( \Delta \omega = \omega_{\text{output}} - \sum \omega_{\text{input}} = 0 \)), and the wavevector (k) of the generated output field must cancel all of the wavevectors of the input fields (Figure 3a, \( \Delta k = k_{\text{output}} - \sum k_{\text{input}} = 0 \)), which is called phase-matching. For this four-wave mixing technique, the interaction of the two pump fields imposes a strict phase-matching condition on the probe field’s angle of incidence (i.e. the Bragg angle). Figure 3b shows the wavevector diagram for the phase-matching of the four beams (the beams are drawn in the same plane of incidence for illustrative convenience). So, the interaction of the two pump pulses and the probe pulse will induce a nonlinear third-order polarization, determined by the susceptibility of the material, which will radiate a signal field in the phase-matched direction of the applied electric fields.

A good thing to note before we go any further is that, so far, the nonlinear polarization has been described in the frequency domain (Equation 5), but through the Fourier transform relation (a mathematical tool allowing a function to pass from the time domain to the frequency domain and vice versa), it can also be described in the time domain. Equation 8 shows the third-order nonlinear polarization as a function of time (where \( t_1, t_2, t_3 \) are the arbitrarily set time separations between the three applied electric fields and the observation time, Figure 4). This form of the equation may be more convenient when performing ultrafast laser experiments where the short applied pulses and interaction variables are typically described with respect to time. We know that susceptibility is a function of frequency of the applied field, however when a Fourier transform to the time domain is applied, the susceptibility is converted to the response function.
(R), still retaining the similar role. However, polarization and electric field in the time domain are observed quantities and are therefore real, which implies that the response function is also real. Susceptibility in the frequency domain is a complex quantity with real and imaginary parts.  

\[ P^{(3)}(t) = \mathcal{E}_0 \int_0^\infty dt_1 \int_0^\infty dt_2 \int_0^\infty dt_3 R^{(3)}(t_1, t_2, t_3) E(t_1-t_2-t_3) E(t_2-t_3) E(t_3-t_1) \]  

Equation 8

So, the general idea behind the four-wave mixing technique is to measure the diffraction/polarization signal intensity with respect to time, then extract the contributions responsible for the changes in the signal that was measured, and finally deduce information from the contributions regarding the dynamics of the system. The subject of basic contributions have already been breached, however we shall approach this from yet another angle. In order to extract the contributions, the signal is analyzed in conjunction with the susceptibility or response function. Since the complex refractive index and susceptibility are directly related parameters, the susceptibility can also be broken down into a sum of terms corresponding to the same basic processes given in Figure 2. However, the third-order susceptibility relating to the OKE electronic contribution is useful for probing various nonlinear interactions and can be further decomposed, using quantum mechanics and the time-dependent perturbation theory, details of which are beyond the

Figure 3

Nonlinear Optics Formalism

a) When the incident fields interact to generate a signal, energy and momentum needs to be conserved. This requires that the frequency and wavevectors of the input fields and output field must cancel out (e.g. \( \omega_s = \omega_1 + \omega_2 + \omega_3 + \omega_4, k_s = k_1 + k_2 + k_3 + k_4 + k_5 \)).

b) A diagram of the transient grating beam geometry and the corresponding wavevector diagram.
scope of this thesis. However, just to get a sense, in the view of quantum mechanics, everything virtually reduces to field interactions. So, the light-matter interaction we have been discussing, is depicted as follows: the material, represented by a field (i.e. a wave function, containing the information of the system), interacts (via dipole coupling) with light (i.e. the applied potential or perturbation), an external electric field, to give rise to an observable or measureable quantity (i.e. the expectation value of an operator), the (induced) dipole moment (microscopic quantity), hence polarization (macroscopic quantity). The susceptibility tensor or response function basically describes how the dipole moment of the system changes in response to changes in the applied electric field. In math language, the response function is obtained by differentiating the dipole moment with respect to the electric field. From here, the susceptibility tensor can be broken down into relevant tensor elements (based on the Cartesian components of the polarization and the incoming fields) or the response function can be broken down into a sum of correlation functions (based on the time-ordering of the incoming fields). For the sake of convenience and ease of explanation, we will switch to the time-domain and response function formalism for the remainder of the chapter. Continuing, the correlation function can be further broken down and be written as a sum of terms (each representing one individual light-matter interaction corresponding to the physical process of photons being absorbed or emitted by the system) over all possible intermediate states of the system resulting from the time-ordered interactions with a given number of applied fields. So, a higher order response function can be expressed as a sum of the correlation functions, each of which are also written as a sum of terms representing an individual light-matter interaction. Thus, the sum of all these terms will map out the different possible permutations of the combination of processes (i.e. transitions) and associated intermediate states of the system, revealing the origin of the polarization (i.e. signal).

All of this, as you can see, is quite complex and very mathematically rigorous. However, let us consider this quantum mechanics approach from a more user-friendly perspective. As we have already seen, there are many ways in which an incoming pulse can interact with a given material. If the material system has only two states (e.g. ground and excited state) and one external electric field is applied, the pulse can either cause absorption or stimulate emission depending on the previous state of the system. Now, if we consider a multi-leveled system and many more external electric fields were to be applied, a large number of different possible transitions and (intermediate) states become available. Response functions are mathematical expressions describing all possible arrangements of these light-matter interactions. Since it becomes difficult to keep track of the large number of transitions and intermediate states, it is useful to have a diagrammatic representation to help visualize and keep track of the interactions. Feynman and ladder diagrams, based on the density matrix formalism for representing response functions, are commonly used visualization tools for describing the various response pathways that contribute to the emitted signal.
Chapter 3: Photon Echo Spectroscopy: 2DPE Electronic Spectroscopy and How It Relates to 3PEPS

Figure 4

a) Ladder Diagram for an Example Experiment (Coherent Raman Scattering)

Ladder diagrams are useful for describing experiments with multiple frequencies or multistate systems, although the time evolutions of the interactions are not as easy to keep track of compared to Feynman diagrams. They are quite similar to Jablonski diagrams with an added time component, thus are more intuitively easier to understand.

1. Multiple states are arranged vertically by energy.
2. Time is running horizontally to the right.
3. Arrows connecting levels represent resonant interaction. Absorption is indicated by an upward arrow and emission with a downward arrow.

b) Double-Sided Feynman Diagrams for Various Four-Wave Mixing Experiments

Feynman diagrams are useful for tracking the state of a system with respect to time and for noting absorption and emission events. The nature of the population (or superposition) state resulting from each field-matter interaction is indicated.

1. Vertical lines represent the time evolution. Time is running upwards.
2. Interactions (of the system’s electric field) with the applied light field are presented by arrows. Absorption is represented through an inward pointing arrow, while emission or de-excitation is an outward pointing arrow. The last emissive interaction is different in character and is usually indicated using a different arrow and it always points away from the system.
3. “g” and “e” denote ground and excited states, respectively. Arrows pointing to the right side of the vertical double line represent the system’s electric field with $e^{i(\omega t-kz)}$, while arrows pointing to the left side represent an electric field with $e^{-i(\omega t+kz)}$. This is due to the fact that the real electric field $E(t) = 2E_0(t)\cos(\omega t)\cos(kz)$ can be separated into positive and negative frequencies $E(t) = E_0(t)(e^{i(\omega t-kz)} + e^{-i(\omega t+kz)})$. The last emitted interaction, has a frequency and wavevector which is the sum of the input frequencies and wavevectors.
4. The last interaction must end in a population state (i.e. eg or ge).

Since, we have not discussed the quantum Liouville equation (the alternative to the Schrödinger equation) and the evolution of density matrices, a full understanding and appreciation of these diagrams cannot be
attained, however for the sake of introduction, a brief overview of the rules of the diagrams are given in Figure 4.

Nonlinear methods, like four-wave mixing, are powerfully complex methods that allow us to select various light-matter interactions and probe specific states, hence dynamics, by controlling the main parameters – wavevector (i.e. beam geometry), frequency, and time-ordering (i.e. sequence and time separation of pulses) of the input fields – of the experiment. In the next section, we will look at experiments that probe these specific light-matter interactions and relate them to photophysical or chemical dynamics that occur in a system.
3.3 Photon Echo Spectroscopy

As we saw in chapter 2, solvent and the local environment can have important consequences on the behavior of materials, energies and rates of reactions, and to optical spectroscopy. For example, the broad, featureless absorption lineshape of tetrazine in an aqueous solution, the non-mirror-image absorption and emission spectra of conjugated polymers, the homogeneous and inhomogenous broadening contributions to the absorption spectrum of semiconductor nanocrystals, while seemingly different on the surface, all of these situations boil down to condensed phase systems subject to various types of fluctuating environments that influence energies, hence transition energy gaps probed in optical spectroscopy (Figure 5).

It appears that the nonlinear response function, describing the third-order polarization signal arising from certain types of four-wave mixing experiments, known as photon echo spectroscopy, probing the time-evolution of solute-solvent systems, contains information corresponding to the transition frequency correlation function related to solvent fluctuations. In particular, optical dephasing in a system is determined by its transition energy gap correlation function and the coupling of the system’s transition dipole to the bath. Dephasing is a process that occurs when synchronization or coherence in a material, caused by a perturbation, decays and eventually vanishes over times, and the system returns to its original state before the perturbation. Optical dephasing occurs when coherently excited molecules, due to interaction with themselves or with the environment, lose their state of coherence and return to their random or statistical nature, the effects of which are significant in condensed phase systems. Solvent fluctuations occur on timescales too short for traditional spectroscopies to probe. Experiments that are capable of resolving these fluctuations can be employed to capture the solvent effect on and response to the photophysical or chemical dynamics that occur in a system at real-time.

Photon echo spectroscopy, which is a subset of four-wave mixing spectroscopy, utilizes advance ultrafast laser techniques to measure the timescale of electronic dephasing in a system and the strengths of solute-solvent interactions, allowing for dynamics of a system to be investigated. There are many different types of photon echo measurements and they can be performed with either two or three optical pulses. This section will start with a discussion on system-bath interactions and spectral diffusion contributions influencing lineshapes in condensed phase systems, then an outline of the basic principles of photon echo spectroscopy will be given, and finally the section will be concluded with the evaluation of two specific
types of photon echo experiments, three-pulse photon echo peak shift (3PEPS) and two-dimensional photon echo (2DPE) electronic spectroscopy.

3.3.1 Solvent Dynamics and System-Bath Interactions: How Do Fluctuations Affect Spectroscopy?

Processes such as random fluctuations in the environment, solvent reorganization around new electronic or nuclear configurations, solvent induced changes of electronic states, are manifested as spectral diffusion, leading to the broadening of lineshapes in optical spectroscopy. Well, what exactly is spectral diffusion? It is given that when a system is placed in an environment, the system will interact with its environment. The degree and nature of the interaction will vary, however an interaction will exist. Since the environment is vast and surrounding, changes in the environment will indeed influence the system. Spectral diffusion is basically the fluctuations in frequency each molecule within a system exhibits, due to coupling with the environment, the effects of which add up to influence the “overall” (ensemble averaged) transition energy gap (being probed), ultimately broadening the spectral lineshape of the system (that is observed).
There are essentially two types of contributions to line broadening, homogeneous and inhomogenous broadening (Figure 6), both of which can be defined by the timescales of the processes involved in each of the broadening mechanisms. Homogeneous line broadening results in narrow Lorentzian line profiles. Homogeneous broadening mechanisms entail rapid dynamic processes involving randomized behavior where all members of the ensemble behave differently (e.g. collisions/pressure broadening) or rapid molecular processes involving population relaxation where all member of the ensemble behave identically (e.g. natural/lifetime broadening). Inhomogeneous line broadening results in broad Gaussian profiles, comprised of a distribution in frequency of homogeneously broadened profiles (each member of an ensemble has its centre frequency shifted by different amounts) (Figure 6d). Inhomogeneous broadening mechanisms entail static processes where molecules within an ensemble are influenced by different structural environmental factors, which can be molecular (e.g. surface defects in individual QDs) or macroscopic (e.g. identical chromophores embedded in non-equivalent sites in an individual protein complex, like in the Fenna-Matthews-Olson complex of green sulfur bacteria) in origin. In summary, fast random fluctuations in the environment will lead to homogeneous broadening and static or set environmental factors will lead to inhomogenous broadening of the linewidth. The environment effects a system experiences will most likely be a mixture of these contributions and spectral diffusion describes the amalgamation of these contributions defined by these two limits. Thus, by understanding these fluctuations we can predict the optical response of materials studied in spectroscopy, or conversely, the signals that we measure in spectroscopy can be traced back to their origin.28,29

The behavior of the environment is coupled to the behavior of the system, but how do we describe this coupled behavior in a quantitative way? As the bath fluctuates from one solvent configuration to another, the system is coupled to these solvent modes, and correspondingly react by changing its energy, hence its transition energy gap (Figure 6e). The correlation function that describes how the transition energy of the system and solvent fluctuations are related, as a function of time, is known as the electronic transition frequency correlation function (M(t)):

\[
M(t) = \frac{\langle \delta \omega_{eg}(0) \delta \omega_{eg}(t) \rangle}{\langle \delta \omega_{eg}^2 \rangle} \tag{9}
\]

\[
\omega'_{eg}(t) = \langle \omega_{eg} \rangle + \delta \omega_{eg}(t) + \varepsilon_i \tag{10}
\]

where \(\delta \omega_{eg}(t)\) is the time-dependent fluctuation of the transition frequency given by Equation 10, \(\omega'_{eg}(t)\) is the transition frequency of a solvated molecule \(i\), \(\varepsilon_i\) is a static offset from the average relating molecule \(i\) to its specific environment, and angle brackets \(<...>\) indicate a statistical average over the
**Figure 6**

**System-Bath Interactions, Spectral Diffusion & Broadening**

a) Generalized solvation coordinates represent solvent modes that couple to the solute (Brownian oscillator model).

b) Spectral dynamics following absorption: solvent relaxation, line broadening and dynamics Stokes shift (DSS).

c) Random fluctuations in transition frequency of an individual molecule.

d) Show the two limiting cases of line broadening. Every system lies between these limits. Spectral diffusion describes the process by which every molecule in a system has a different \( \omega_{eg} \) which evolves with time.

---

**Potential Energy Surfaces Resulting from Solute-Solvent Interactions**

- **Energy** vs. **Generalized Solvation Coordinates, Q**
  - Broadening
  - Absorption
  - Fluorescence
  - DSS

**Trajectory of Individual Molecule**

- **Energy** vs. **Time**
  - \( \hbar \omega_{eg}(t_1) \)
  - \( \hbar \omega_{eg}(t_2) \)
  - \( \hbar \omega'_{eg}(t_3) \)

---

**Homogeneous** vs. **Inhomogeneous** vs. **Spectral Diffusion**

- **Energy Distribution**
  - \( \omega_{eg} \)
  - \( <\omega_{eg}> \)
  - \( \omega'_{eg}(t) \)

---
ensemble. The denominator term $<\delta \omega_{eg}^2>$, also written as $\Delta^2$, represents the frequency dependent strength of the system-bath interaction (i.e. coupling strength). This expression contains all the information regarding homogeneous and inhomogeneous broadening, the system-bath coupling, and solvent (vibrational) dynamics for a given system.\textsuperscript{28,30-32}

Not only do fluctuations in the environment give rise to line broadening, they also give rise to the Stokes shift. The familiar red shift in the band maximum of a fluorescence spectrum relative to that of the absorption, accompanying a solvent reorganization and relaxation process is known as the time-dependent fluorescence Stokes shift, also known as the dynamic Stokes shift (DSS) (Figure 6b).\textsuperscript{33-38} Even though the environment is always in a state of fluctuation, it still allows the system to relax and equilibrate its free energy. This is because fluctuations in the environment are also influenced by the system (arising from the Bose-Einstein statistics of phonon occupation which provides the drive for the system to equilibrate). The DSS correlation function ($S(t)$), is given by:

$$S(t) = \frac{\nu(t) - \nu(\infty)}{\nu(0) - \nu(\infty)}$$

Equation 11

where $\nu(t)$ is the average frequency of an emission at time t. $S(t)$ and $M(t)$ are closely related (via the fluctuation dissipation theorem) and at ambient temperature, under the conditions of linear response (i.e. the high temperature classical limit, where the frequency distribution of the fluctuation is far less than the thermal energy), the two functions are equivalent ($S(t) = M(t)$). So, $M(t)$ and $S(t)$ make up for the dynamic influence the solvent and system inflict on each other.\textsuperscript{28,30-32}

$M(t)$ can also be written in terms of spectral density (Equation 12 and 13), which can be defined either as $\rho(\omega)$ or $\omega^2 \rho(\omega)$. Spectral density, which describes the density of states of the bath, is the fundamental quantity for which all other necessary quantities required to completely describe the optical response of a system can be derived. It can be further broken down into modes corresponding to low frequency continuous mode (over-damped solvent motions) and intermolecular vibrations (where the summation goes over all the vibrational modes of the system) (Equation 14).

$$M(t) = \frac{1}{\langle \Delta^2 \rangle} \int_0^\infty d\omega \omega^2 \rho(\omega) \coth\left(\frac{\hbar \omega}{2k_B T}\right) \cos(\omega t)$$

Equation 12

$$\omega^2 \rho(\omega) = \frac{2}{\pi} \langle \Delta^2 \rangle \tanh\left(\frac{\hbar \omega}{2k_B T}\right) \int_0^\infty dt M(t) \cos(\omega t)$$

Equation 13

$$\rho(\omega) = \rho_{\text{phonon}}(\omega) + \sum_j \rho_{\text{mode}}^j(\omega)$$

Equation 14
Spectral density, \( M(t) \), and \( S(t) \) can then be used to determine the lineshape function \( (g(t)) \) (Equation 15 and 16). In Equation 15, the real portion includes the coupling strength, the time-dependent (homogeneous) and static (inhomogeneous) line broadening contributions from \( M(t) \), while the imaginary portion includes \( \lambda \), the solvent reorganization, and \( S(t) \), both of which describes the shift of the centre transition frequency. In the high temperature limit, where \( S(t) = M(t) \), the imaginary part can also be described by \( M(t) \) (Equation 16). It can be seen in this equation that only the real part of \( g(t) \) is temperature dependent, which indicates that line broadening is sensitive to temperature while the shift is temperature independent. All of the other parameters \( S(t) \), \( \lambda \), and \( \langle \Delta^2 \rangle \) can also be written in terms of spectral density (Equation 17-19).

\[
g(t) = \langle \Delta^2 \rangle \int_0^t dt_1 \int_0^{t_1} dt_2 M(t_2) - i \lambda \int_0^t dt_1 S(t_1)
\]

Equation 15

\[
g(t) = \int_0^\infty d\omega \rho(\omega) \coth \left( \frac{\hbar \omega}{2k_B T} \right) \left( 1 - \cos(\omega t) \right) - \frac{i \lambda t}{\hbar} + i \int_0^\infty d\omega \rho(\omega) \sin(\omega t)
\]

Equation 16

\[
S(t) = \frac{1}{\lambda} \int_0^\infty d\omega \rho(\omega) \cos(\omega t)
\]

Equation 17

\[
\lambda = \int_0^\infty d\omega \rho(\omega)
\]

Equation 18

\[
\langle \Delta^2 \rangle = \int_0^\infty d\omega \rho^2(\omega) \coth \left( \frac{\hbar \omega}{2k_B T} \right)
\]

Equation 19

From \( g(t) \), the experimentally seen observables, the linear absorption and fluorescence lineshapes, as well as the nonlinear response functions can be obtained. The linear absorption (\( \sigma_A \)) and emission (\( \sigma_E \)) spectra can be reconstructed through a transformation of \( g(t) \) as shown:

\[
\sigma_A(\omega) = \int_\infty^\infty dt \exp \left[ -i (\omega - \omega_{eg}) t \right] \exp \left[ - g(t) \right]
\]

Equation 20

\[
\sigma_E(\omega) = \int_\infty^\infty dt \exp \left[ -i (\omega - \omega_{eg}) t \right] \exp \left[ - g^*(t) \right]
\]

Equation 21

where the asterisk signifies the complex conjugate.

In order to calculate, for example, the third-order nonlinear response functions from \( g(t) \), we first need to consider all of the possible four field-matter interactions for a two-level system contributing to the nonlinear response function. This can be done using a Feynman diagram. There are 8 diagrams or contributions that describe the pulse ordering and interactions contributing to the generation of the third-order polarization (Figure 7), four of which are degenerate cases (where the second and third interactions
Figure 7

Double-Sided Feynman Diagrams for Third-Order Optical Spectroscopies (i.e. Photon Echo) of a Two-Level System

These eight diagrams represent the correlation functions contributing to the response function for the third-order polarization signal emitted in the $k_1 + k_2 + k_3$ phase-matching direction. Time increases from bottom to top and $t$, $T$ represent time periods which can be controlled experimentally. The signal is typically recorded by integration over the period $t$. The eight diagrams can be grouped in the following ways:

1. Ones that involve rephaessing ($t > 0$) to produced echo signals ($R_{yy}$, $R_{yx}$, $R_{xy}$, $R_{xx}$) versus ones that do not rephaess ($t < 0$), termed non-rephasing, leading to free induction decay (FID) signals ($R_e$, $R_y$, $R_x$, $R_{xx}$). The ones that can rephaess have polarizations during time periods $|t|$ and $t$ which are complex conjugates of each other (i.e. superpositioned states are exchanged such that eg $\rightarrow$ ge). If they do not rephaess, such exchange does not take place (i.e. eg $\rightarrow$ eg or ge $\rightarrow$ ge).

2. Ones that go through the excited state (ee) ($R_e$ to $R_x$) versus ones that go through the ground state (gg) ($R_g$ to $R_g$) during the time period $T$. The ones that go through an excited state, indicates the presence of a population period (i.e. to form a population state). Experiments, such as 3PPE, which go through the ee state are capable of monitoring excited state dynamics or spectral diffusion (unlike 2PPE, which does not have a population period, hence is similar to experiments only going through the gg state). Note, for properly time-ordered 3PPE experiments under the impulsive limit (i.e. applied fields are assumed to be $\delta$ functions), only $R_e$ to $R_e$ contributions can be considered. However, for real experiments using laser pulses with finite durations, time-ordering becomes uncertain when pulses overlap, hence $R_e$ to $R_e$ contributions must be taken into account as well.

3. In the degenerate case, where the second and third pulses are equivalent (even in exchanged superpositioned states), diagrams for $R_e$, $R_y$, $R_x$, $R_{xx}$ are degenerate or equivalent to $R_e$, $R_y$, $R_x$, $R_{xx}$, respectively. Hence, only the four contributions need to be considered.
are equivalent and their exchange does not alter the time-evolution of the response). The response function for the third-order polarization signal emitted can actually be expressed as the sum of 48 correlation functions in all of the phase-matching directions \( \pm k_1 \pm k_2 \pm k_3 \), however utilizing the time-ordering of the pulses, the rotating wave approximation, and considering the \(-k_1 + k_2 +k_3 \) phase-matched direction only, the response function correlation functions can be reduced to 4, corresponding to each of
the 8 diagrams (with 4 degeneracies/complex conjugates), and then it can be expressed in terms of $g(t)$ utilizing a second order cumulant expansion:

$$R_1 = (R_4) = \exp\left[ -g^*(t_1) + g(t_2) - g^*(t_3) - g^*(t_1 + t_2) - g(t_2 + t_3) + g^*(t_1 + t_2 + t_3) \right]$$  \hspace{1cm} \text{Equation 22}

$$R_2 = (R_3) = \exp\left[ -g(t_1) - g^*(t_2) - g^*(t_3) + g(t_1 + t_2) + g^*(t_2 + t_3) - g(t_1 + t_2 + t_3) \right]$$  \hspace{1cm} \text{Equation 23}

$$R_3 = (R_8) = \exp\left[ -g^*(t_1) + g^*(t_2) - g(t_3) - g^*(t_1 + t_2) - g^*(t_2 + t_3) + g^*(t_1 + t_2 + t_3) \right]$$  \hspace{1cm} \text{Equation 24}

$$R_6 = (R_7) = \exp\left[ -g(t_1) - g(t_2) - g(t_3) + g(t_1 + t_2) + g(t_2 + t_3) - g(t_1 + t_2 + t_3) \right]$$  \hspace{1cm} \text{Equation 25}

The response function contributions can be summed to give the total third-order nonlinear response function ($R^{(3)}(t_1, t_2, t_3)$), which can be used with Equation 8 to obtain the third-order polarization and relate it to the signal. A summary chart of the theoretical framework relating the system-bath correlation function to spectroscopic observables obtained from different types of experiments is shown in Figure 8. 28,30-40

### 3.3.2 Principles of Photon Echo

Now that we have connected the dynamics found in a system to the fundamental theory to the experimental observable. We will look at a specific type of spectroscopy which ties in everything that we have discussed in this chapter thus far. Photon echo (PE) spectroscopy uses a variant form of four-wave mixing technique where the two pump pulses are not time-coincident (i.e. temporally overlapping). The time separation in the two pump pulses is what allows us to observe and measure the optical dephasing in a system. 41,42

When coherent light (such as a laser, which produces polarized, unidirectional waves at a single frequency whose phase is correlated over long distances) interacts with an ensemble of molecules, it can cause the system to exist in a superposition state between the ground and excited states (or in certain cases between excited states of different molecules). The formation of a superposition state is called a coherence. This coherent state possesses a fixed phase relationship (meaning a constant relative phase) between the wave function of the ground and excited state. As time evolves, the synchronized superposition of states created by the laser starts to decay or randomize (phases begin to shift and are no longer constant), due to interaction of the excited molecules with themselves or the environment (i.e. photophysical or chemical dynamics within the system) – a process called dephasing. And eventually, the coherence is lost and the
system is returned back to its original state before the interaction (provided that no irreversible chemical reaction has occurred).\textsuperscript{43}

In an actual photon echo experiment, typically performed with two or three optical pulses sequentially applied to a sample at a particular geometry, very similar in principle to the four-wave mixing technique described previously, optical dephasing is simply quantified by measuring the intensity of the generated third-order nonlinear polarization signal as it evolves in time. The intensity of the echo signal will be proportional to the time delay between the two excitation fields and decays through fluctuations of the transition frequencies (i.e. optical dephasing or decoherence). So, how do we do this if the two pump pulses are not time-coincident, thus an intensity grating cannot be formed? When the pump pulses arrive sequentially, with some time delay, the light fields will not be able to interfere directly. However, if we consider the case mentioned above, when a laser pulse interacts with a sample, a coherence state can be formed. If the formed coherence can be sustained until the arrival of a second pulse, interference between the polarization (created by the first pulse) and the second pulse will occur, forming a transient frequency or spectral grating (Figure 9a,b). If the electric fields associated with the pump pulses separated in time by τ in the time domain, were to undergo a Fourier transform to the frequency domain, a single Gaussian shaped intensity spectrum containing spectral fringes related to the time separation of the pulses (i.e. frequency grating) is produced. This technique is called spectral interferometry. It is important to note that a transient frequency grating can only be formed in the sample, if the sample retains “memory” of the first pump pulse when the second pulse arrives. As the two pump pulses are separated further in time (i.e. as τ increases), the fringe spacing of the frequency grating will correspondingly decrease (i.e. lose resolution). When the pulses are separated to the point where the sample has completely lost memory of the first pump field by the time the second pulse arrives (coherence has been lost), the fringes even off to form a smooth Gaussian curve (frequency grating has vanished), similar to the case when there is only one pulse applied to the sample (no frequency grating formed). The lifetime of the coherence corresponds to the electronic dephasing time. So, if the time delay, τ, between the first two pulses is greater than the dephasing time, the sample will have no memory of the first pulse and a grating will not be generated.\textsuperscript{5,7,13,30,31,44,45}

Now, let us consider a sample with an inhomogeneously broadened absorption spectrum. The frequency grating formed from the interaction of the two applied fields with the sample, basically represents a grating of excited and ground state molecules, which can be seen as a selective excitation of (homogeneously broadened) subpopulations within the sample. If we were to imagine the absorption spectrum immediately after selective excitation, we would most likely see empty portions or “holes” in the spectrum
Figure 9

**Frequency Grating and Photon Echo Technique**

a) Fourier transformed intensity spectra of an electric field and two electric fields separated in time at various intervals. Left diagram shows electric fields (in time domain) and the right shows intensity spectrum (in the frequency domain) associated with electric field(s) to the left. As separation time between the two electric fields (pulses) increases, the spectral fringe spacing decreases (or modulations increase) to the point where they get too close together to be resolved and eventually disappear, leaving a single Gaussian pulse (similar to the case where there is only one electric field).

b) Modulation of population in ground and excited states. The horizontal axis gives the detuning (i.e. frequency shift) from the center line. The diagrams do not indicate the absolute energy in either the ground or excited state.

c) A frequency grating formed by the interaction of two pump pulses selectively excites subpopulations of an inhomogeneously broadened sample (i.e. holes formed in the absorption spectrum). A probe pulse is applied to the sample. Sample acts like an optical filter and transmits probe frequencies corresponding to the hole. Signal emerges reproducing original frequency grating.
corresponding to the excited subpopulations that have been removed from the ground state (Figure 9c).

Such an experiment actually exists and it is called a spectral hole-burning experiment. Now, if a third field were to be applied to the sample, the sample will behave like an optical filter and transmit frequencies of the applied third field that correspond to the holes in its absorption spectrum, reproducing the original frequency grating that was formed by the first two fields (which were separated in time by \( \tau \)). This is how holography works. Also, please note that in the case of two-pulse photon echo (2PPE) experiments, where only two optical pulses are used, the sample interacts with the higher intensity second laser pulse twice (acting as a time-coincident pump and probe pulse), adding up to give the three light field-matter interactions.\textsuperscript{13,30,31,46,47}

Since the three light fields are applied to the sample at a particular geometry, a spatial grating will also form in addition to the frequency grating, like in the case of the general four-wave mixing technique. The spatial grating will transmit most of the light, while a small portion of it propagates through the medium, and through the frequency grating, which is then diffracted in the appropriate phase-matched direction, forming the signal. Unlike the four-wave mixing technique, the signal produced does not immediately appear with the application of the third field, but is uniquely delayed by \( \tau \) (the time separation of the initial two pulses) from the last pulse applied, reflecting the inverse-Fourier transform of the reproduced frequency grating. This delayed signal is what is known as a photon echo and it can only be formed in the presence of inhomogeneous broadening.

To appreciate the idea behind the photon echo, a well-known “foot race” analogy is be given for a 2PPE experiment (Figure 10).\textsuperscript{31,48-51} Let us consider a race between three runners, Jack, Dereke, and Scott (representing molecules in the sample). The three runners have different levels of fitness experience and will run at different speeds (inhomogeneous broadening). At the sound of a starting pistol (i.e. the first laser pulse), all three runners, lined up at the starting line, take off simultaneously (coherence induced). After some time delay, the once synchronized runners become randomly distributed on the track (dephasing), due to the different speed of each runner. With the sound of the second gunshot (the second laser pulse), the runners immediately stop, then start running in the reverse direction (rephasing initiated). If each runner maintains their respective speed, all of the runners will arrive at the start line in sync once again (rephased, echo signal). Despite the varying speed of the runners, everyone arrives at the start line at the same time.

Realistically speaking, the speed of the runners may fluctuate throughout the race. Dereke may get tired, Scott may stumble, and Jack may get a burst of energy nearing the end of the race (fluctuation in
Figure 10

**Photon Echo “Foot Race” Analogy**

Runners on a race track represent the molecules in a sample. A pulse is analogous to the sound of a starting pistol, initiating the race (coherence initiation) or a change in direction of the runners (rephasing initiation). The spread in position of the runners over time represents the amount of dephasing in the sample. If rephasing is successful, the end result is an “echo” indicating the re-coherence of the runners.

If a fluctuation in speed occurs, the runner will not be able to cross the starting line together, and there will be a spread in their positions (rephasing is imperfect, resulting in diminished echo signal). So, if the runners are allowed to run for a longer period before the second pistol is shot (more delay between first two pulses), there is a greater chance for the speed of the runners to fluctuate (irreversible dephasing), leading to a greater spread in positions (even after rephasing the inhomogeneity). Irreversible dephasing is what affects the maximum intensity (i.e. perfect rephasing) of the measured echo and the increased effects of irreversible dephasing is
what causes the eventual decay of the intensities over time (Figure 11a). Rephasing is what triggers the echo, but it basically only rephases the inhomogeneity in the sample. Thus, the effect of inhomogeneity is eliminated, at the same time we utilize it to produce an echo signal which carries the dephasing information of the collective runners (the system).

![Figure 11]

**Figure 11**

**Two-Pulse Photon Echo**

a) A typical integrated echo signal intensity versus delay time curve of a two-pulse photon echo experiment.

b) Pulse and signal sequence in a 2PPE experiment. The difference between an FID and an echo signal is illustrated.

Well, what happens in cases where the sample is not inhomogeneously broadened, so that rephasing does not really take place? If rephasing cannot take place, there will be no echo. However, this does not mean that a diffracted signal is not produced. This will produce a linear polarization situation, where the polarization built up in the sample will emit an electric field which will decay over a time period determined by the relaxation rate of the excited molecular states (i.e. dephasing time). This emission is called the free induction decay (FID). Since, FID is not an echo signal, its appearance will be time-coincident with the applied third field (i.e. there is no delay). So, technically we should be able to distinguish between the two signals (Figure 11b). However, temperature plays a big role in these measurements. At high temperatures (>100K), dephasing occurs rapidly in condensed phase systems due to an increase in thermal fluctuations. This becomes a bit of a nuisance when experiments are performed at room temperature. Since the coherence period is shortened as fluctuations increase, the time period between the third field and echo signal can reduce dramatically to the point where it becomes hard to distinguish between an echo signal and FID, thus a distinction between homogeneous and inhomogeneous broadening becomes unclear. A way to partially address this problem is to measure the time-integrated signal produced by the sample (since the intensity of the echo signal is expected to be slightly higher than
the FID signal). A common way to measure the time-integrated signal is using the echo-peak shift (EPS) method. The three-pulse photon echo peak shift (3PEPS) technique combines three-pulse photon echo (3PPE) measurements with EPS to provide photon echo information in a condensed and convenient form. The details of the experiment will be discussed in the following section.\textsuperscript{12,31}

3.3.3 Three-Pulse Photon Echo Peak Shift (3PEPS)

Before we jump into the details of 3PEPS, let us first briefly consider the idea behind the three-pulse photon echo. We have already discussed the general principles of photon echo in the previous section, which applies to all various forms of photon echo experiments, so only the variances will be highlighted. The three-pulse photon echo (3PPE) technique, also known as stimulated photon echo, utilizes three distinct optical pulses for generation of a photon echo signal which allows for more control of the experiment. The addition of a third pulse allows us to not only probe optical dephasing, but look at the flow of population within a system.

Starting with the technical details, the two typical beam geometries used for 3PPE are shown in Figure 12a. In the “box configuration”, the three beams are aligned to form three corners of a square/box before being focused onto the sample, and as usual, this configuration requires that Bragg diffraction conditions must be met in order to obtain a signal, in the phase-matching direction $-k_1 + k_2 + k_3$, which forms the fourth corner of the square. In the second configuration, the three beams form an equilateral triangle prior to being focused onto the sample, and allows for signals to be observed in two phase-matching directions, $-k_1 + k_2 + k_3$ (for pulse sequence 1-2-3) and $k_1 - k_2 + k_3$ (for pulse sequence 2-1-3), even though the beam angles depart from the phase-matching condition (phase-matching conditions can be partially eased, if the sample thickness is thin enough to be considered within the “thin grating” regime). The advantage of the second configuration is that two spatially isolated echo signals from the two different pulse sequences can be simultaneously measured and compared. The second geometry is used for 3PEPS experiments.\textsuperscript{52-58}

In 3PPE the three pulses are sequentially applied to generate an echo signal (Figure 12b). The time delay between the first and second pulse ($\tau$) is known as the “coherence period” and the delay between the second and third pulse ($T$) is known as the “population period”. A 3PPE experiment is basically conducted by measuring a series of 2PPE echo decays (echo intensity is recorded as a function of $\tau$) for various fixed values of $T$, except (mirror-image) echo signals in both phase-matching directions are
measured (which allows us to precisely determine the position of $\tau = 0$). This entails that pulse delays are scanned from negative $\tau$ (with pulse sequence 2-1-3) to positive $\tau$ (with pulse sequence 1-2-3), such that $T$ is fixed between pulses 1 and 3 at $\tau < 0$ and then between 2 and 3 at $\tau > 0$. Then, $T$ is incremented to a new value and $\tau$ is scanned once again. Eventually, after collecting many scans of $\tau$ at various $T$, a three-dimensional surface is effectively recorded (as a function of signal intensity, coherence time, and population time).
The main difference between 2PPE and 3PPE is the presence of a population period, due to the added control of a time delay for third pulse. Through the interaction of the first two pulses, a coherence is induced in the system and allowed to dephase in the time allotted by the coherence period. With the application of the second pulse, the dephasing process is “frozen” in time and the evolved dephased state during the coherence period is stored in sample (i.e. the sample keeps a memory of the generated frequency grating). During the population time (i.e. the time between the second and third pulse), the system is allowed to evolve a population on either the excited or ground state, before the third pulse is applied to initiate rephasing for echo generation (hence called stimulated photon echo). Please note that for $\tau < 0$ (with pulse sequence 2-1-3), due to the reverse order of the first two pulses, a coherence is not created, hence no rephasing will be initiated for echo generation. This situation will also produce an FID. Since two different behaviours are triggered for $\tau < 0$ and $\tau > 0$, we refer to these as nonrephasing and rephasing sides (Figure 7).

In 2PPE, the echo intensity can be affected by dephasing occurring during $\tau$ and $t$ (between the probe field of pulse 2 and the generated echo). However, in 3PPE, the echo intensity can be affected by dephasing occurring during $\tau$ and $t$ (between probe pulse 3 and the generated echo), as well as spectral diffusion which can occur during $T$. $\tau$ is the time period between the first two pulses, which defines the duration or amount of dephasing in the induced coherence, and affects the number of modulations in a frequency grating, which in turn determines when the echo is produced. As $\tau$ increases, dephasing sets in, reducing the coherence, and the frequency grating loses its resolutions (i.e. fringe spacing reduces as the number of modulations in the grating increase), resulting in a decrease in signal intensity. $T$ is the time period between the second and third pulse, which allows the population (either the excited state population created by the excitation pulses or the ground state population left behind) to explore and evolve (i.e diffusion). This exploration will manifest as changes in their average transition frequency (i.e. spectrum), undoing the memory of the frequency grating initially imprinted onto the population by the initial excitation pulses (i.e. diminishes the modulation depth of the grating). At short $T$ periods, the memory of the frequency grating is still “fresh”, since the population has not had much time to explore the range of transition frequencies available, so an echo will be produced, similar to the case of 2PPE (where pulse 2 and 3 are time-coincident, i.e. $T=0$). As $T$ increases, spectral diffusion takes place, resulting in a gradual loss of the memory. As memory of the frequency grating is lost, rephasing becomes less and less likely and the echo signal produced will start to resemble the FID signal on the nonrephasing side. However, an echo will form as long as $T$ is shorter than the lifetime of the frequency grating (i.e. lifetime of the hole or selective excitation produced by the two pump pulses). This could mean a few nanoseconds or even much
longer (i.e. if accompanied by chemical reactions). Being able to monitor spectral diffusion allows us to probe various dynamics that are lurking within the system and 3PPE becomes a multidimensional tool.\textsuperscript{12,31}

3PPE is normally measured as a time-integrated photon echo signal ($S_{\text{INT}}$), which is expressed as follows:

$$S_{\text{INT}}(T, \tau) = \int_0^\infty \left| P_{\text{3PPE}}^{(3)}(t, T, \tau) \right|^2 dt$$

\textit{Equation 26}

where $P_{\text{3PPE}}^{(3)}$ is the third-order polarization produced from the 3PPE experiment.\textsuperscript{30} However, it was found that the time-integrated photon echo signal could be better utilized if the information was presented as a peak shift ($\tau^*$) plotted as function of $T$ (which corresponds to the average of the coherence time when the time-integrated echo signals peak). Since each of the phase-matching 3PPE signals is approximately symmetric and its maximum value is shifted from the $\tau = 0$ position, the signals are fitted to a Gaussian, then the time interval between the two maxima divided by 2 (i.e. peak shift) is plotted against $T$ (Figure 13a).

If we consider the 2PPE case and the “foot race”, the echo intensity will be the highest at small $\tau$ values where irreversible dephasing (i.e. homogeneous broadening contributions) has not had a chance to set in, and echo is produced mainly from rephasing of inhomogeneity in the system (i.e. rephases to original coherence condition or has perfect memory of its initial state). As $\tau$ values increase, the intensity will decay mainly due to irreversible dephasing. So, for any 2PPE echo decay, the maximum echo peak position corresponds to mostly inhomogeneous contributions. 3PPE is very much like 2PPE except with another dimension, so we have to factor in another consideration, the population time. As $T$ increases, spectral diffusion sets in and the maximum echo peak position (of the 2PPE) shifts from $t = \tau > 0$ to $\tau = 0$ as it eventually converts to an FID signal (Figure 14). Thus, the time evolution of the peak shift ($\tau^*$) is able to separate inhomogeneous and homogeneous contributions and is said to closely follow the $M(t)$ correlation function, for population times longer than 50-100 fs:

$$M(T) = \frac{\Delta \sqrt{\pi} \tau^*(T)}{1 + 2\Delta^2 \tau^*(T)}$$

\textit{Equation 27}

$$\tau^*(T) \approx \frac{\hbar}{2\pi \lambda (k_B T)} M(T)$$

\textit{Equation 28}

where $T_k$ represents absolute temperature. Since real pulses are of finite duration, pulse overlap issues arise at short times, in the region of $\tau = T = 0$, and hinder the ability of 3PEPS to characterize the initial decay of $M(t)$. However, as long as pulse durations are short compared to the timescales of transition frequency fluctuations, 3PEPS observables and determination of $M(t)$ are not significantly affected and is a great match for $M(t)$ at longer times.\textsuperscript{28,30,52,59-61}
A sample 3PEPS plot is shown in Figure 13b. In order to extract information regarding the characteristic timescales of bath relaxation and system-bath coupling strength, computer simulations must be performed. However, some general qualitative interpretations can be made of the 3PEPS plot features. The initial peak shift is associated with the system-bath coupling strength. A higher initial peak shift implies a weaker coupling between the system and bath. Initial peak shift represents a time period (small τ) when inhomogeneous contributions dominate over homogeneous contributions. Let us consider a situation when inhomogeneity is high in the system. In this case, it will take a longer time to rephase this inhomogeneity,
and the echo will appear at longer $\tau$ (meaning a larger $\tau^*$). Conversely, if the inhomogeneity in the system is low, or to the point where the system has no inhomogeneity (which produces an FID with no delayed echo), then less rephasing is required and a small shifted echo will result (meaning a smaller $\tau^*$). So, a high initial peak shift may suggest that there is high inhomogeneity and also weak system-bath coupling. High inhomogeneity means longer rephasing time, and during this rephasing time, fluctuations can occur, but if fluctuations do not dominate and affect $\tau$, then this would mean that the system is weakly coupled to the bath. Also, it has been known in some situations, as $T$ increases, $\tau^*$ will not fully decay to zero, but to some nonzero asymptotic peak shift (offset). Various explanations have been proposed for this observation, and it seems that it is mainly related to the specific physical system under investigation.
However, many of these studies indicate that disorder in the system will manifest as an asymptotic shift, due to some memory retention of the initially prepared state.\textsuperscript{28,40} In addition, damped oscillatory components can also be clearly seen in 3PEPS data but are washed out in the integrated echo signal. These oscillatory components, known as “quantum beats”, are attributed to intramolecular vibrations (wavepacket dynamics in ground and excited states) in the system.

### 3.3.4 Two-Dimensional Photon Echo (2DPE) Electronic Spectroscopy

A newer generation of photon echo spectroscopy known as two-dimensional photon echo electronic spectroscopy (2DPE, 2D-PES or 2D-ES) has been under development for over a decade and has gained popularity in the past several years.\textsuperscript{62-67} The development of the technique was inspired by two-dimensional nuclear magnetic resonance (2D-NMR) experiments, which was later extended to infrared spectroscopy,\textsuperscript{68-71} and now many of the protocols have been extended and adapted for use in the visible wavelength regime to obtain ultrafast electronic dynamics and structural information of many diverse systems. Even though many difficulties regarding rigorous phase-stability requirements and interferometric signal detection had to be overcome, it has proven to be a powerful technique which has enhanced the field of nonlinear optical spectroscopy.

3PEPS and 2DPE are both from the same family of photon echo, more specifically 3PPE. Like 3PEPS, 2DPE measures the evolution of the third-order polarization signal, which contains dynamic population and coherence information about the ensemble. However, there are two main differences. First, the method of data collection is quite different. 3PEPS is considered a homodyne experiment, which means that the information on the phase of the induced polarization is lost. In order to obtain phase information of the induced polarization, a heterodyne-detected photon echo experiment needs to be performed. For heterodyne detection, a fourth field, called the local oscillator (LO), is used to cause interference with the signal field (i.e. spectral interferometry).\textsuperscript{30} This can be done in the time domain or the frequency domain. The technique in the frequency domain is much more convenient since the echo spectrum (i.e. reproduced frequency grating) can be taken in a single shot (using an optical multichannel analyser) without having to measure the signal as a function of delay time, and 2DPE utilizes this technique. Secondly, the measured data is also presented quite differently. In 2DPE, the information is provided in a two-dimensional (2D) plot, which allows for a direct visual mapping of the energetic and dynamic progression of the system’s
response to excitation. So, 2DPE offers the same nature of information as 3PEPS but in a perceptive graphical manner, making it the most versatile photon echo technique currently available.

In general, 2DPE uses a box configuration for the beam alignment and the signal is measured in only one phase-matched direction, \(-k_1 + k_2 + k_3\)\(^{62}\). Like 3PEPS, the photon echo signal is, once again, measured while a scan of the coherence time from \(-\tau\) to \(\tau\) is made for many fixed values of population times. A set of data can be collected for increasing values of \(T\) to obtain an evolution of the spectrum, hence mapping out the dynamics. Aside from the beam geometry, as mentioned, the main difference lies in signal detection.

While 3PEPS uses a diode detector to measure only the intensity of the homodyne signal, 2DPE makes use of an LO pulse, which is overlapped with the signal, to produce an interferogram (heterodyne signal) detected by a charge-couple device (CCD), recording both the amplitude and phase information of the signal field\(^{72,73}\). The interferometric signal picked up by the CCD camera can be displayed as a 2D plot of \(\tau\) versus pixel at a particular \(T\), where pixels directly correspond to the echo emission frequency (\(\omega_t\)). Through the Fourier relationship, the \(\tau\)-axis can also be transformed to form the conventional 2DPE spectrum, displayed as \(\omega_t\) versus \(\omega\tau\) (i.e. signal intensity versus \(\tau\) at a particular \(T\) in 3PEPS language). If we convert the frequency to energy and flip the axis, then we can have coherence energy (excitation energy put into the system) versus rephasing energy (energy of the signal emitted to bring the system back to original state), which can be thought of as an excitation versus emission plot, analogous to 2D fluorescence. The phase information obtained (in conjunction with a spectrally resolved pump-probe data set) is used to separate the real (absorptive) and the imaginary (refractive medium) contributions of the collected signal, for which only the real portion is used in creating the standard 2DPE spectrum.

Once the 2DPE contour plots are constructed at various \(T\), some of the important features to take note of are the diagonal peaks and cross peaks (or off-diagonal peak), the asymmetry of the spectrum, and also the shape of the peaks (Figure 15). Briefly stereotyping these features, diagonal peaks correspond to the linear absorption spectra; the position and development/formation of cross peaks and asymmetry correspond generally to dynamics of excitation, which can reveal information regarding the origin and evolution of the excitation (e.g. energy transfer, coherence, etc.). The shapes of the peaks correspond to the distribution of environments (e.g. homogeneous and inhomogeneous factors). Since the signal along the diagonal is produced when excitation and emission energies are identical (\(\omega_t = \omega_e\)), this indicates that the transition energy/frequency has not changed (similar to the initial peak shift in 3PEPS). Hence, broadening along
the diagonal is due to inhomogeneous broadening, while homogeneous broadening is observed in the direction perpendicular to the diagonal.

Figure 15

2D Photon Echo Spectroscopy and Its Features

In general, bleaching and stimulated emission appear as positive features, while excited state absorption appears negative. Diagonal peaks correspond to the linear absorption spectra. The position and development of any cross peaks and asymmetry correspond to various dynamics of excitation, which can reveal information regarding the origin and evolution of the excitation. Broadening along the diagonal is due to inhomogeneous broadening, while homogeneous broadening is observed in the direction perpendicular to the diagonal.

It has also been shown from experiments and simulations that solvent dynamics can be observed in both the real and imaginary contributions of the signals collected. In the real part of the signal, the peak shape will lose its diagonal structure at larger population times, as the solvent configuration changes in response to the new charge distribution in the excited solute. In the imaginary part of the signal, the slope of the nodal line between the positive and negative peaks of the refractive signal will change over population time, and the correlation function can also be directly obtained by this determination. A number of other methods have been suggested for the extraction of the correlation function from the 2DPE spectra. Next
section will describe a simple method that we have implemented for extraction of this important parameter.

3.4 2DPE Electronic Spectra of Rhodamine 6G Dye

2DPE electronic spectroscopy offers diverse and abundant amount of information on system-bath couplings, coherence dynamics, structural associations, to energy migration, for the most convoluted of systems. Information such as the linear absorption, pump-probe, transient grating, hole burning and various types of photon echoes, including 3PEPS data, are all contained within the single measurement, multidimensional data obtained, but a suitable form of data reduction must be applied in order to extract the information that is desired.

In this section, we take a glimpse into the ultrasfast optical behaviour of a common organic dye, Rhodamine 6G (R6G). 2DPE electronic spectra of R6G in two solvents, methanol and N,N-dimethylaniline, have been obtained using the technique of frequency heterodyned 3PPE. The following will provide a description of the 2DPE experimental setup, the steady-state and 3PEPS profile of R6G in the two solvents, an outline of a simple method for extracting 3PEPS data from the observables collected for 2DPE results, and the 2DPE electronic spectra of R6G obtained.

Rhodamine 6G (590) perchlorate laser dye was purchased from Exciton Co. and was used without further purification. Spectrophotometric grade or otherwise highest purity solvents, methanol (MeOH) and N,N-dimethylaniline (DMA), were obtained from Aldrich and used as received. Absorption spectra were obtained on a CARY BIO UV-Vis spectrophotometer. Steady-state photoluminescence spectra were measured using a Cary Eclipse fluorescence spectrophotometer. Details of the ultrafast photon echo measurements are outlined in section 3.4.1. The R6G 3PEPS data were provided by Dr. Mayrose Salvador and Professor Gregory Scholes, while the 2DPE results and analysis were acquired with the assistance of Dr. Cathy Wong and Dr. Kyung-Koo Lee. All ultrafast laser experiments on R6G were carried out at room temperature. R6G samples were solvated in methanol and DMA to an optical density of approximately 0.3 at the absorption maximum, in a 1mm path-length quartz cuvette, in order to avoid interchromophore interaction effects. For the R6G in DMA sample, a small amount of methanol (1%) was added to assist with solvation. The addition did not cause any change in the absorption shape and width of the sample.
3.4.1 Experimental Setup

In our laboratory, a titanium-sapphire regeneratively amplified laser system (Clark-MXR, CPA-2001) is used to generate 140 fs pulses at 775 nm, which has a repetition rate of 1 kHz at a power of ~780 mW. A non-collinear optical parametric amplifier (NOPA) is used to convert the wavelength to the visible region. The incoming beam from the laser is split into two beams by the NOPA. One beam is sent to a sapphire window to generate white light continuum. This beam will be used as a seed for the amplification process. The other beam is sent to a Type I BBO (β-Barium-Borate) crystal cut for second harmonic generation. This beam will be used as a pump for amplification. The two beams are then spatially and temporally overlapped in a second Type I BBO crystal cut to produce frequency-tunable pulses. The seed and the pump beams can be delayed using a translation stage to determine which spectral component of the white light continuum will enter the BBO crystal and be strongly amplified by the pump.

The resulting pulse from the NOPA will have some frequency dispersion or “chirp” (pulse stretching), and must be compressed in order for the entire spectrum of the pulse to interact with the sample at once. A pair of quartz prisms is used to accomplish this and also to compensate for any other dispersion that could occur in all of the transmissive optics used, leading up to the sample position. Also, two lenses are placed immediately after the NOPA and the prism pair to collimate and reduce the spatial profile of the beam. Note, the duration of the resulting pulse, measured by auto-correlation using a blank sample (such as methanol or toluene), is typically compressed to 25-30 fs. A beam splitter then divides the beam into a pump and probe beam. A population delay time (T) is introduced by sending the probe beam to a retroreflector, which is mounted on a stage of a stepper motor (Newport, UTM150PP.1), the movement of which controls the delay. A half-wave plate and polarizer are inserted in the path of both beams to enable power control.

At this stage, the two pulses enter the 2DPE setup (Figure 16), and are focused into a diffractive optic (DO) (Institut National d'Optique), a transmissive grating, which produces four beams. Two beams originate from the pump beam and two from the probe beam (probe and LO) to produce a box configuration beam geometry. The resulting pulse pairs are passively phase-locked, thus the resulting wavevectors are automatically aligned for Bragg angle phase-matching. Note, two glass windows are placed before (in the probe beam path) and after (in the pump beam paths) the DO to prevent the pump
and probe beams from overlapping, which can lead to unwanted pump-probe signals from the DO interfering with the pulses.

Next, the resulting four beams are collimated between two parabolic mirrors (PM), then focused into the sample. Between the two parabolic mirrors, a number of optics are placed in the beam paths while the beams are collimated. A half-wave plate (WP) is inserted into all four beam paths for independent control of the polarization, if needed. Two pairs of wedges are introduced into the paths of the pump beams to produce the coherence time delay ($\tau$) (where a wedge from each pair is mounted on a stage of a small stepper motor with a range of 10 cm, the movement of which controls the delay by adjusting the amount of glass the beam must pass through). Another pair of wedges is inserted into the LO beam to create another delay, so that the LO pulse can arrive (~500 fs) before the probe pulse. Additionally, a chopper is placed in the probe beam (set to ~3 Hz) to prevent any unwanted pump-probe signal contributions induced by the interaction of the LO and either of the pump beams.

![Figure 16](image)

2D Photon Echo Electronic Spectroscopy Experimental Setup

Light blue lines originate from the pump beam, while the dark blue lines originate from the probe beam. The dotted red line is the transient grating signal, which overlays with the LO beam (dotted cyan line). The pulse sequence is shown in the bottom right corner.
After careful alignment of the two pump pulses (i.e. wavevectors \( k_1 \) and \( k_2 \)), the probe pulse (\( k_3 \)), and the LO pulse (\( k_{LO} \)), the four beams are focused into the sample. The two pump and probe beam are blocked after the sample to decrease contributions from scattering. A signal field is radiated in the phase-matching direction \( k_S = -k_1 + k_2 + k_3 \), which is the same direction as the \( k_{LO} \). Finally, the spatially overlapped signal and LO fields are focused into a 0.63 m spectrograph with a 25 \( \mu \)m slit, then the interferogram is recorded using a 16 bit, 400 \( \times \) 1600 pixel, thermoelectrically cooled charged coupled device (CCD) detector.

### 3.4.2 Steady-State Measurements and 3PEPS Data

The molecular structure of the R6G dye and DMA are shown in Figure 17a. R6G, a member of the xanthene class of dyes, is a commonly used highly polar organic dye. It has many modes of vibration. The broad absorption and PL features observed for R6G in solution are due to strong molecular interactions with the solvent, broadening the vibrational structure of the transitions into unresolved bands (Figure 17b). In highly polar solvents, the electronic structure of R6G is strongly coupled to solvent fluctuations and the dephasing rate is expected to be fast. Due to the rigid structure of R6G, the lifetime of the excited state is long enough (and has a small intersystem crossing ratio) to allow for efficient emission of excitation energy. The PL quantum yield of R6G in methanol is nearly one (0.93) \(^78\), making it the most efficient lasing dye presently known.

N,N-dimethylaniline is a weakly polar, organic, substituted derivative of aniline. The lone pairs of electrons on the nitrogen feed into the adjacent \( \pi \) system, activating the aromatic benzene ring by increasing electron density on the ring through resonance effects, and making it more reactive toward electrophiles. These types of compounds are known as electron-donating solvents. Photoinduced intermolecular electron transfer between cationic dyes and electron-donating solvents, such as DMA, have been widely investigated. \(^79\)\(^\text{--}\)\(^82\) There is great interest in these systems since bimolecular ET in inert or unreactive solvents is mainly diffusion-limited and it becomes difficult to extract information on the true ET dynamics. \(^83\) However, with the use of reactive electron donating solvents, the electron donor (i.e. the solvent) and acceptor (i.e. the dye molecule) are in direct contact, thus diffusional motion is discounted, and ET can occur very fast after photoexcitation. \(^83\)\(^,\)\(^84\) It has been found that ET in these systems is nearly barrierless and occurs on a timescale much faster than that of diffusive solvation. \(^79\)\(^\text{--}\)\(^82\) Hence, we take a closer look at the photoinduced reactions of R6G dye in the unreactive and reactive solvents, methanol and DMA, respectively.
Absorption and emission spectra of R6G in methanol and DMA are shown along with the laser pulse spectra in Figure 17b. The absorption maximum in methanol (529 nm or 566 THz) is blue shifted compared to DMA (540 nm or 555 THz). The vibronic shoulder is also enhanced and the spectrum is broadened in DMA (512 nm or 585 THz) compared to methanol (497 nm or 602 THz). This is because the vibrational modes that are coupled to the reaction coordinate are likely to undergo larger displacements (i.e. larger coupling strength to the electronic transition of R6G) in the reactive solvent compared to methanol. The fluorescence of R6G in methanol (maximum 552 nm or 543 THz) was almost completely quenched in DMA (Figure 17b, dotted lines). The quenching indicates the removal of excited state...
population to a dark state through some process which we attribute to electron transfer. The following
reaction is thought to occur in the reactive solvent:

\[ \text{R6G}^+ + \text{DMA} \xrightarrow{hv} \text{R6G}^{*+} + \text{DMA} \xrightarrow{k_{ET}} \text{R6G}^* + \text{DMA}^+ \xrightarrow{k_{BET}} \text{R6G}^* + \text{DMA} \quad \text{Equation 29} \]

where R6G\(^+\) is R6G in its natural cationic state, \(k_{ET}\) and \(k_{BET}\) are the rate of photoinduced ET and back ET, respectively. The lifetime of R6G is measured to be \(~4\) ns in various alcohol solvents, while the excited state lifetime in DMA is reported to be under 6 ps.\(^8\)

3PEPS data for R6G in methanol and DMA are shown in Figure 17c. As mentioned in section 3.3.3, the initial peak shift value is largely influenced by the coupling strength between the electronic transition and the bath. A higher total coupling strength leads to lower initial peak shift values. As well, the asymptotic peak shift is associated with the long-time inhomogeneity. As can be seen from the 3PEPS plot, DMA has a higher initial peak shift, despite the larger coupling strength, and faster early time decay compared to the methanol. Xu et al.\(^8\) using a three-level model analysis of the 3PEPS data (incorporating an excited state absorption component to upper electronic states in the nonlinear response function), rather than the customary two-level system which typically reflects only solvation dynamics, revealed that in reactive systems, the initial peak shift value can be higher due to the ET reaction. The additional ET contribution to the analysis also accounts for the faster peak shift decay at early times, as well as the higher peak shift over the the intermediate population time region (200 to 600 fs). By combining the results of the transient grating and 3PEPS measurements, they were able to extract information on solvation dynamics and their entanglement with the ET reaction and concluded that for R6G in DMA, rapid photoinduced ET occurs on a time scale of \(~85\) fs. The use of the three-level model was able to provide information on the population sink between the excited and ground state, and indicated that the solvation process and the ET reaction could be considered essentially independent within the system.

### 3.4.3 3PEPS Data Extraction From 2DPE Electronic Spectra

Spectroscopic studies of condensed phase systems are convoluted by system-bath interactions occurring over a wide range of timescales. To better understand chemical reactivity, information regarding the system-bath interactions and dynamics are essential. 3PEPS spectroscopy allows us to investigate these environmental fluctuations, through the characterization of the M(t) or signal decay. The great potential of 2DPE electronic spectroscopy is that all of the possible nonlinear responses for a particular phase is
Chapter 3: Photon Echo Spectroscopy: 2DPE Electronic Spectroscopy and How It Relates to 3PEPS

3PEPS Extraction from 2DPE Data of R6G in Methanol

From the raw signal (a) picked up by the CCD camera, using the phase information obtained through a pump-probe measurement and Fourier transforming the $\tau$ axis, the imaginary (b) and real (c) contributions can be separated, for which the real component is shown in a typical 2DPE spectrum. From the same raw signal, it is possible to extract 3PEPS data by inverse Fourier transforming the pixel axis, then integrating over the positive portion of the results (d). Then, the integrated data is fit to a Gaussian (similar to time-integrated 3PPE curve), and $\tau^*$ is obtained. The steps are repeated for data collected at various $T$ to reconstruct the 3PEPS data set (e).
contained within the set of spectra measured. In this section, a procedure for extracting 3PEPS data from the measured 2DPE spectra of R6G in methanol is summarized.

Continuing from where we left off in the experimental setup section, raw interferograms produced by the signal and the LO field is detected by the CCD. The interferogram, picked up by the CCD, is essentially a 2D plot of pixels or rephasing frequency ($\omega_t$) versus coherence time ($\tau$) at a particular $T$. A set of complex algorithms (Fourier transform spectral interferometry) can be used to process the raw data and isolate the complex signal, containing both the real and imaginary components.\textsuperscript{85,86} Then, by Fourier transforming the $\tau$ axis (of the real component) a conventional 2DPE spectrum in frequency can be produced, which can be converted to energy. However, if the pixels or $\omega_t$ is inverse Fourier transformed, a plot of the time delay between the signal and LO pulse ($t_{LO} - t_s$) versus $\tau$ is produced, which is a 2D spectrum in time. By integrating over the $t_{LO} - t_s$ axis, we obtain a linear spectra showing the amount of signal (i.e. intensity) produced at each $\tau$. This yields the same information as a 3PPE measurement, which is simply the diode detected intensity of a homodyne PE signal with respect to $\tau$. Then, the shift of the peak (i.e. maximum $\tau$ value, $\tau^*$) from the 3PPE-like plot at at time $T$ can be collected for many $T$s and plotted as a conventional peak shift decay, $\tau^*$ versus $T$. So, by inverse Fourier transforming the pixels or rephasing frequency axis, the peak shift information can be obtained from 2DPE measurements.

In Figure 18, starting from the raw 2D photon echo data of R6G in methanol at $T = 200$ fs, a progression of the 3PEPS extraction process is illustrated and compared with the actual 3PEPS data for R6G. Furthermore, the imaginary and real (standard 2DPE plot) contributions of the collected 2D photon echo signal are shown.

3.4.4 2DPE Electronic Spectra of R6G in MeOH and DMA

Interpretation of 3PEPS data at first glance, beyond a few qualitative features, is an impossible feat without having modeled the system, nor having any supporting data from other complementary measurements. 2D optical spectroscopy allows us to make visual connections to many different phenomena related to the structure and organization of a system and its time dependence. Unlike conventional linear frequency or time decay profiles of optical spectroscopy, the increased dimensionality of the 2D spectra gives rise to diagonal and cross peaks with respect to $T$ (variously known as the population, waiting, or mixing time), the shapes of which are influenced by phenomena such as mode
coupling, energy transport, and internal energy reorganization. In this section we take a look at the 2D-ES of R6G in methanol and DMA, obtained from the frequency heterodyned 3PPE measurement, and see what qualitative information on this dye-electron donating solvent system can be gained.

The real, rephasing, and nonrephasing 2D-ES of R6G in methanol and DMA are shown in Figure 19. The real and imaginary spectra are produced by balancing the rephasing and nonrephasing coherence pathways. By looking at the deconstructed, rephasing and nonrephasing, contributions a more unambiguous assessment of the absorptive lineshapes in the real spectra can be made. For example, based on the phase and amplitude relationships of oscillations in the diagonal and cross peaks, their presence or absence in the rephasing and nonrephasing 2D maps may help to distinguish between vibrational or electronic coherences. Also, by looking at the amplitude differences of rephasing and nonrephasing signals, it may be possible to obtain information on the solvation dynamics of the system.

R6G excitation dynamics for a T time period up to 400 fs was obtained for both methanol and DMA (Figure 19). The 2D spectra for both solvents showed fairly consistent dynamics throughout the full T time period of the measurement. In the methanol sample, two features are notable. In the real spectra, a strong positive amplitude peak, at a coherence (or “absorption”) frequency value of 555 THz and a rephasing (or “emission”) frequency value of 555 THz, is centred about the diagonal. A second weaker positive amplitude feature appears as a vibrational cross peak just underneath the main diagonal peak. There is an initial inhomogeneity, seen at T = 0 fs, where the absorptive lineshape of the real spectra reveals a diagonal elongation. This is due to the varying solvent environment. The width of the diagonal signifies the instantaneous homogeneous distribution of solvent environments formed by the excitation. At longer T times, the rephasing and nonrephasing amplitudes equalize, and a rapid transition from inhomogeneous to homogeneous broadening occurs, as indicated by the decrease in diagonal elongation in the real spectra (T = 50 fs). This evolution of the initial diagonal elongation is attributed to inertial solvation. Solvent motion can have at least two major components, a fast inertial and a slower diffusive component, described via M(t) as a sum of a Gaussian and an exponential function, respectively. Figure 20 contains traces of selected diagonal and cross peak values as a function of T time periods. From the plot, it can be seen that the weak cross peak (in yellow) decays rapidly (~25 fs) and lingers as a small asymmetry to the main diagonal peak. There may also be some underlying oscillations seen in the R6G methanol traces (red and yellow), which are indicative of nuclear vibrational coherences, but the presence or absence of the oscillations is difficult to resolve due to the noticeable signal-to-noise in the measurement.
Figure 19

a  Real 2DPE Electronic Spectra of R6G in MeOH

<table>
<thead>
<tr>
<th>Rephasing Frequency (THz)</th>
<th>Coherence Frequency (THz)</th>
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<tbody>
<tr>
<td>0 fs</td>
<td>0 fs</td>
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<tr>
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<tr>
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<tr>
<td>400 fs</td>
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</tbody>
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Real 2DPE Electronic Spectra of R6G in DMA

<table>
<thead>
<tr>
<th>Rephasing Frequency (THz)</th>
<th>Coherence Frequency (THz)</th>
</tr>
</thead>
<tbody>
<tr>
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<td>400 fs</td>
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</tbody>
</table>
Figure 19 (continued)

Rephasing Spectra of R6G in MeOH

Rephasing Spectra of R6G in DMA
In the DMA sample, the 2D results seemed relatively similar to that of methanol. A strong diagonal peak with coordinates (548, 548)THz is observed. A similar transition from inhomogeneous to homogeneous broadening is seen at early times. There is less of a noticeable cross peak in the DMA, however there is a touch of asymmetry, to the left and bottom, of the main diagonal peak. Traces of the diagonal and cross peaks of DMA (Figure 20, blue and green) look relatively featureless. This may be due to interference from the somewhat significant nonresonant (background) response from the DMA. Indications of ET could not be revealed in the visual asessment of the 2D results.

Figure 20

Values Extracted from 2DPE Electronic Spectra of R6G

- MeOH diagonal peak @ (560,560)THz
- DMA diagonal peak @ (548,548)THz
- MeOH cross peak @ (560,520)THz
- DMA cross peak @ (550,540)THz

Population time, T (fs) vs. Intensity (a.u.)
3.5 Conclusion

In this chapter, a review of the essentials of nonlinear spectroscopy, system-bath interactions and how they relate to spectroscopy, principles of photon echo, 3PEPS and 2DPE, and a demonstration of how 3PEPS data can be extracted from 2DPE measurements were provided. The 2D visualization of information in 2DPE often allows for a more intuitive way to resolve complex dynamics occurring in systems and allows for the identification of phenomena that cannot be measured using other forms of spectroscopy. 3PEPS has been the preferred direct method for obtaining transition frequency correlation functions, which allows for the determination of spectral density, a fundamental quantity required to completely describe the optical response of a system. However, the direct physical interpretation of 3PEPS results is not always an intuitive process and requires careful consideration, in combination with modeling and other types of experiments. The extraction of this useful 3PEPS data from 2DPE results emphasizes the versatility of 2DPE spectroscopy and its power to characterize diverse and complex molecular systems.
3.6 References


