Control of Excited States and Photoinduced Ligand Substitution Reactions in Ru(II) Complexes for Photochemotherapy

DISSERTATION

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Abstract

Cisplatin currently remains one of the most effective ways to treat cancer. Its mode of action relies on thermal ligand dissociation upon entering the cell and then covalent binding to DNA, which interferes with transcription and cellular replication and ultimately causes cell death. However, due to poor selectivity and increased resistance, a new treatment method is in development known as photochemotherapy (PCT), in which light is used as an external source to trigger a photosensitizer that produces a species that is cytotoxic, but the drug remains inactive in the dark. PCT provides great spatial and temporal control and low systemic toxicity and could operate more effectively than current cisplatin treatment. The complex \([\text{Ru}(\text{bpy})_2(\text{CH}_3\text{CN})_2]^{2+}\) (bpy = 2,2’-bipyridine) has been thoroughly investigated in which irradiation with visible light in water results in efficient ligand dissociation forming the diaqua cisplatin analog, \([\text{Ru}(\text{bpy})_2(\text{H}_2\text{O})_2]^{2+}\). Furthermore, \([\text{Ru}(\text{bpy})_2(\text{CH}_3\text{CN})_2]^{2+}\) was found to covalently bind to DNA similar to cisplatin, but only upon irradiation (\(\lambda_{\text{irr}} > 455\) nm), making it a potential PCT agent. However, the optimum window for PCT is 600 – 850 nm, where light is absorbed by skin and tissue minimally and \([\text{Ru}(\text{bpy})_2(\text{CH}_3\text{CN})_2]^{2+}\) is only photoactive with 455 nm light.

To this notion the 2,2’-biquinoline (biq) ligand was incorporated into similar ligand exchange systems, specifically in the complex \([\text{Ru}(\text{biq})_2(\text{CH}_3\text{CN})_2]^{2+}\). Utilization of the biq ligand red shifts the lowest energy metal-to-ligand charge transfer (MLCT) transition and the complex is photoactive with relatively high efficiency just inside the therapeutic...
window (≥ 600 nm, Φ₅₅₀ = 0.15) and covalently binds to DNA while remaining inert in
the dark. However, ligand exchange is not observed for [Ru(biq)₂(CH₃CN)₂]²⁺ in
ultrafast laser experiments (λₑₓ = 310 nm, fwhm = 300 fs) in H₂O as in
[Ru(bpy)₂(CH₃CN)₂]²⁺.

Interestingly, the tris-heteroleptic complex [Ru(biq)(phen)(CH₃CN)₂]²⁺ (phen = 1,10-phenanthroline) undergoes selective ligand exchange of one CH₃CN preferential to
the other, which is not observed in the symmetric complexes [Ru(biq)₂(CH₃CN)₂]²⁺ and
[Ru(phen)₂(CH₃CN)₂]²⁺. Moreover, the Φ₅₅₀ measured for the first ligand exchange in
H₂O for the asymmetric complex is 0.14 and the second ligand exchange is 0.0014. This
unprecedented selective ligand exchange is enhanced based on irradiation wavelength
and could be useful for PCT in multiple drug delivery systems. The monosubstituted
product of [Ru(biq)(phen)(CH₃CN)₂]²⁺ irradiated in pyridine was isolated and
characterized by X-ray crystallography and it was found the CH₃CN trans to the phen
ligand is the more photolabile ligand.

In order to further understand the electronic and steric factors that govern selective
ligand exchange, asymmetric tris-heteroleptic complexes with the formula
[Ru(phen)(L)(CH₃CN)₂]²⁺, where L is a various dimethyl-phen or biquinoline ligand,
were synthesized and studied. It was found that complexes with L = 2,9-dimethyl-1,10-
phenanthroline and 1,1’-isobiquinoline also undergo selective ligand exchange in H₂O and
CD₃CN but complexes with L = 4,7-dimethyl-1,10-phenanthroline and 3,3’-
isobiquionline do not. Similar to the 2,2’-biq ligand, the 2,9-dimethyl-phen ligand
possesses steric bulk directed towards the metal center. This distorts the pseudo-
octahedral geometry and the e\textsubscript{g}-type orbitals involved in bonding to the CH\textsubscript{3}CN ligands. This distortion results in selective ligand exchange. Another commonality observed from X-ray crystallography and calculations in the complexes that undergo selective ligand exchange is tilting of the diimine ligand out of the normal octahedral plan. This tilting affects the overlap involved in $\pi$-backbonding to one CH\textsubscript{3}CN preferential to the other resulting in selective ligand dissociation. Furthermore, no significant distortions are observed for complexes that do not undergo selective ligand exchange.

The effects of steric strain imposed by the bulky 2,2'-biq ligand was further analyzed in the known complexes [Ru(biq)\textsubscript{2}(bpy)]\textsuperscript{2+} and [Ru(biq)\textsubscript{2}(phen)]\textsuperscript{2+} in which irradiation with light in the therapeutic window ($\geq$ 645 nm) in H\textsubscript{2}O results in photoejection of the biq ligand with $\Phi_{600} = 0.006$ and 0.002, respectively. In order to red shift the absorption further into the PCT window, the cyclometallated complex [Ru(biq)\textsubscript{2}(phpy)]\textsuperscript{+} was synthesized. Although it possesses similar steric strain imposed by the biq ligands, no photoinduced ligand dissociation is observed because the $^3$MLCT drops to low in energy making the $^3$LF state inaccessible. However the cyclometallated complex displayed successful cytotoxic results and was more potent when irradiated than both [Ru(biq)\textsubscript{2}(bpy)]\textsuperscript{2+} and [Ru(biq)\textsubscript{2}(phen)]\textsuperscript{2+}.

Not only does steric bulk contribute to bidentate ligand dissociation but it was found to induce efficient pyridine (py) ligand dissociation in the complex [Ru(tpy)(dmbpy)(py)]\textsuperscript{2+} (dmbpy = 6,6'-dimethyl-2,2'-bipyridine) relative to [Ru(tpy)(bpy)(py)]\textsuperscript{2+}, which have $\Phi_{500} = 0.16$ and $<10^{-4}$ in CH\textsubscript{3}CN, respectively. Ultrafast transient absorption studies in CH\textsubscript{3}CN ($\lambda_{ex} = 568$ nm, fwhm = 300 fs) reveal
excited state lifetimes of ~38 ps and ~470 ps for [Ru(tpy)(dmbpy)(py)]$^{2+}$ and [Ru(tpy)(dmbpy)(py)]$^{2+}$, respectively, supporting a lowering of the energy of the $^3$LF state below that of the $^3$MLCT in the former. The photoproduct [Ru(tpy)(dmbpy)(CH$_3$CN)]$^{2+}$ was also observed during the ultrafast experiment but due to the weak signal intensity relative to that of the starting material and the overlapping spectra, no specific kinetics of this formation could be discerned. Further understanding of pyridine ligand dissociation using steric bulk in Ru(II) complexes is useful for PCT in which pyridine based drugs can be “caged” and activated with light for selective treatment.
Dedication

This document is dedicated to Maryann and Eugene Albani.
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I would like to thank my advisor Claudia Turro for all of her help in my graduate research. She encouraged me to become an independent scientist while constantly pushing me and enabled me to think more critically and pay closer attention to detail. I am very grateful for former group members Alycia Palmer, Scott Burya, Nick Leed, and Robert Garner for training me to use instrumentation and teaching me important concepts to guide my research. I would also like to thank my current group members Jessica Knoll, Travis White, Regina Akhimie, Rachel Whitman, Suzanne Witt, Tyler Whittemore, TJ Rohrabaugh, Michele Folmar, and Lauren Loftus for their good comradery in the lab and a special thanks to those in the group I have publications with. Additionally, I appreciate Kim Dunbar of Texas A&M as a great collaborator and her students I have gotten to work with especially Bruno Peña, Zhanyong Li, and Amanda David.

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xxx
Chapter 1 : Introduction and Background

1.1 Light and Photochemistry

Upon absorption of a photon, a molecule or atom can be promoted from its ground state corresponding to its lowest energy electron configuration to higher energy excited states.\textsuperscript{1,2} These excited states can nonradiatively or radiatively decay through vibrational relaxation (heat) or emission of a photon typically at lower energy than the absorbed photon, respectively, as well as undergo photochemical reactions known as photochemistry.\textsuperscript{2} A photochemical reaction can be represented as $R + \hbar \nu \rightarrow R^* \rightarrow P$, where $R$ is a molecule that absorbs light, $R^*$ is the molecule in the excite state, and $P$ is the resulting photoproduct.\textsuperscript{2} Placing “$R$” in an excited state gives it sufficient energy to overcome activation barriers that were not possible in the lowest energy ground state. Photochemical reactions are utilized in nature in photosynthesis to convert CO$_2$ and H$_2$O into energy,\textsuperscript{3} and in bodily processes such as the production of Vitamin D.\textsuperscript{4} Moreover, photochemistry is essential for the advancement of many fields of research including photocatalysts,\textsuperscript{5,7} solar energy conversion,\textsuperscript{8,9} molecular switches,\textsuperscript{10} chemical sensors,\textsuperscript{11} and photoactivated drugs.\textsuperscript{12,13}
Figure 1.1. Jablonski diagram of a simple organic molecule.

In order to further understand specific photochemical systems for the above applications, a general Jablonski state diagram is shown in Figure 1.1 to illustrate the possible excited state pathways and reactions in a simplistic manner. Absorption of a photon by a molecule results in a vertical spin-allowed transition from the ground state, $S_0$, to the lowest energy excited singlet state, $S_1$ (1 in Figure 1.1). It should be noted that higher energy singlet states such as $S_2$, $S_3$, etc. could be included but are not included for simplicity. The molecule can relax back to the ground state by emission of a photon known as fluorescence (2 in Figure 1.1) or by nonradiative decay releasing heat known as internal conversion (IC, 3 in Figure 1.1). Under the proper conditions a photochemical reaction can occur from $S_1$ resulting in the corresponding photoproduct, $P_1$ (4 in Figure 1.1).
1.1), or a spin-forbidden nonradiative transition known as intersystem crossing (ISC) to the lowest energy triplet excited state, $T_1$, can occur corresponding to the spin flip of an electron (5 in Figure 1.1). Once again, other higher energy triplet excited states such as $T_2$, $T_3$ etc. could be included in the diagram if desired. From $T_1$, the molecule can return to $S_0$ through spin-forbidden transitions that emit a photon known as phosphorescence (6 in Figure 1.1) or heat by IC (7 in Figure 1.1). A photochemical reaction can also occur from $T_1$ resulting in the photoproduct, $P_2$ (8 in Figure 1.1). It is evident that the three important molecular states in photochemistry are $S_0$, $S_1$, and $T_1$, however, the state diagram in Figure 1.1 corresponds to organic molecules. Many of the photochemical applications listed earlier utilize inorganic complexes, which have been studied less extensively. The photochemical model in Figure 1.1 is the same for inorganic complexes, however, the excited state dynamics are different. The difference in the excited state dynamics can make understanding them more complicated than previously studied organic molecules. Furthermore, the ability to easily synthesize inorganic complexes with various metals or ligands provides a field of study with vast differences among molecules making inorganic photochemistry an important area of research.

1.2 Photochemistry of Ru(II) Complexes

1.2.1. [Ru(bpy)$_3$]$^{2+}$

The rich photophysical and photochemical properties of Ru(II) polypyridyl complexes have been extensively studied. Specifically, [Ru(bpy)$_3$]$^{2+}$ (bpy = 2,2’-bipyridine), shown in Figure 1.2a, has become the model complex to understand
inorganic photochemistry. It has been utilized in solar energy conversion schemes due to its good excited state oxidizing and reducing capabilities and many other examples of Ru(II) polypyridyl complexes have been previously reported.\textsuperscript{14,17}

![Molecular structure of [Ru(bpy)_3]^{2+}](image)

**Figure 1.2.** (a) Molecular structure (b) MO diagram, state diagram, and electron configuration of states and (c) Jablonski diagram for [Ru(bpy)_3]^{2+}.
A ligand coordination system in the presence of metals makes new transitions possible such as a metal-to-ligand charge transfer (MLCT), ligand-to-metal charge transfer (LMCT), and metal centered (MC) transitions. An example of a metal centered transition, also known as a ligand field (LF) or dd transition, involves electron density moving from one molecular d-orbital to another at higher energy.

A simplified molecular orbital (MO) diagram of the frontier orbitals of [Ru(bpy)$_3$]$^{2+}$, the state diagram, and the corresponding electron configuration of those states is shown in Figure 1.2b. The lowest energy transition is MLCT in nature corresponding to an electron that is promoted from the t$_{2g}$-type subshell to non-bonding unoccupied bpy($\pi^*$) molecular orbitals. The dd transitions occur at higher energy with low oscillator strength involving an electron that is promoted from the t$_{2g}$-type subshell to metal d orbitals that possess $\sigma^*$ character corresponding to the dd state and higher energy ligand centered transitions occur that involve the promotion of an electron from the filled bpy($\pi$) subshell to unoccupied bpy($\pi^*$) orbitals corresponding to a $\pi\pi^*$ state.

Upon excitation from the singlet ground state ($^1$GS) to the $^1$MLCT state, ISC is very fast for Ru(II) complexes (< 100 fs), and occurs with 100% quantum yield (Φ) due to enhanced spin orbit coupling. Moreover, due to fast ISC, the known excited state chemistry of Ru(II) polypyridyl complexes takes place from the triplet manifold schematically depicted in the state diagram in Figure 1.2b. In [Ru(bpy)$_3$]$^{2+}$ (and many other Ru(II) polypyridyl complexes) the $^3$MLCT excited state is emissive with a lifetime of ~600 ns in CH$_3$CN (Figure 1.2c). Also shown in Figure 1.2c is the $^3$dd state.
Population of this state can result in ligand dissociation decreasing the overall excited state lifetime and emission of the complex. A large portion of research in the field of Ru(II) photochemistry has been devoted to raise the energy of the $^3\text{dd}$ state to avoid this issue. In contrast, this dissertation focuses on complexes that possess low lying $^3\text{dd}$ states that are efficiently populated from the $^3\text{MLCT}$ state resulting in enhanced ligand substitution reactions for applications in photochemotherapy (PCT).

1.2.2. Photoinduced Ligand Substitution

The broadly accepted model for photoinduced ligand substitution in Ru(II) complexes is depicted in Figure 1.3. The mechanism relies on thermal population of the reactive $^3\text{dd}$ state(s) from the lower lying $^3\text{MLCT}$ state(s).$^{25-28}$ The population of the $^3\text{dd}$ states places electron density on the $e_g$-type orbitals with Ru – L $\sigma^*$ character, thus resulting in ligand dissociation (Figure 1.3a).$^{25-28}$ The energy gap between the $^3\text{MLCT}$ and $^3\text{dd}$ states greatly affects the quantum yield value of ligand exchange ($\Phi$),$^{26}$ and direct excitation of the $^1\text{dd}$ state(s) with higher energy light results in enhanced ligand exchanged yields.$^{27}$
Figure 1.3. (a) MO diagram with $\pi$ backbonding and $\sigma^*$ orbitals shown and (b) Jablonski diagram showing population of the $^3$dd state resulting in ligand dissociation.

Previous research showed that the complex $[\text{Ru(bpy)}_2(\text{CH}_3\text{CN})_2]^{2+}$ (1) undergoes sequential ligand substitution in $\text{H}_2\text{O}$ when irradiated with visible light forming the diaqua species $[\text{Ru(bpy)}_2(\text{H}_2\text{O})_2]^{2+}$ (Figure 1.4).$^{29}$ Moreover, 1 exhibits a relatively high quantum yield in $\text{H}_2\text{O}$ ($\Phi_{400} = 0.21$), a value that is significantly greater than those found with related Ru(II) complexes, such as $[\text{Ru(bpy)}_2(\text{NH}_3)_2]^{2+}$ and $[\text{Ru(bpy)}_2(\text{py})_2]^{2+}$ ($\text{py} = \text{pyridine}$).$^{12,29,30}$ In order to gain a better understanding of the ligand exchange dynamics in 1, ultrafast transient absorption (TA) experiments were carried out.

Figure 1.4. Photoinduced monodentate ligand dissociation of $[\text{Ru(bpy)}_2(\text{CH}_3\text{CN})_2]^{2+}$ in $\text{H}_2\text{O}$.
1.2.2. Ultrafast Ligand Exchange in [Ru(bpy)$_2$(CH$_3$CN)]$^{2+}$

Transient absorption studies allowed for the measurement of the kinetics of ligand exchange for 1 in CH$_3$CN and H$_2$O after ultrafast excitation ($\lambda_{ex} = 310$ nm, fwhm = 300 fs).$^{29}$ In CH$_3$CN, where no photoproduct is expected to form, typical features of the $^3$MLCT state were observed that include the signal at $\sim$365 nm corresponding to reduced bpy ligand, bleaching of the ground state at 425 nm, and a broad signal $>$ 500 nm.$^{23}$ The $^3$MLCT signal at 365 nm decays with $\tau = 50$ ps and the bleach recovers biexponentially with $\tau_1 = 28$ ps and $\tau_2 = 50$ ps, while the broad signal decays with a time constant $\tau = 28$ ps. The 50 ps component was assigned as the lifetime of the $^3$MLCT state and the 28 ps component was assigned as the regeneration of the initial species from the pentacoordinate intermediate (PCI), [Ru(bpy)$_2$(CH$_3$CN)]$^{2+}$. $^{29}$ It should also be noted that the excited state lifetime of 1 is much shorter than that observed for [Ru(bpy)$_3$]$^{2+}$ due to the efficient population of the $^3$dd state.

However, in H$_2$O the formation of the monoaqua photoproduct complicates the kinetics and the resulting TA spectra are shown in Figure 1.5. At early times (0 – 10 ps) the spectral features are identical to those observed in CH$_3$CN, but at later times (10 to 1000 ps) the growth of a band is observed at 458 nm corresponding to [Ru(bpy)$_2$(CH$_3$CN)(H$_2$O)]$^{2+}$. The decay of the signal at 365 nm is the same for that observed in CH$_3$CN with $\tau_1 = 50$ ps, corresponding to the decay of the $^3$MLCT state, but the longer biexponential decay of the ground state bleach is observed with $\tau_1 = 50$ ps and $\tau_2 = 77$ ps, which corresponds to the $^3$MLCT and formation of the monoaqua species, respectively. The broad absorption $>$ 500 nm was ultimately assigned as the PCI and an
important point from this experiment is that the PCI is formed within the 300 fs laser pulse indicating the ligand dissociation is fast and beckons the question as to whether the $^3\text{dd}$ state is directly populated from $^1\text{MLCT}$ state through ISC. This hypothesis is still an ongoing point of research in our group and some of the new CH$_3$CN/py ligand exchange complexes presented in this dissertation help elucidate this.

![Figure 1.5](image_url)

**Figure 1.5.** Ultrafast TA of [Ru(bpy)$_2$(CH$_3$CN)$_2$]$^{2+}$ in H$_2$O at early times (top) and later times (bottom) ($\lambda_{\text{ex}} = 310$ nm, fwhm = 300 fs). Reproduced from Reference 29.

### 1.3 Cisplatin and Photochemotherapy

#### 1.3.1. Cisplatin

Although it was approved for treatment by the FDA in 1977, the use of cisplatin, Pt(NH$_3$)$_2$Cl$_2$, and related platinum drugs currently remains one of the most effective ways
to treat cancer. The therapeutic action of cisplatin relies on the exchange of the chloride ligands with water molecules upon entering the cell, and the latter are easily displaced in the presence of DNA to form covalent bonds with adjacent guanine bases (Figure 1.6). This critical ligand exchange takes place thermally, simply activated through body heat; once formed, the metal-DNA adduct interferes with cellular replication and protein synthesis. If the DNA damage is too extensive to be repaired, then programmed cell death is induced, apoptosis (Figure 1.6).\textsuperscript{31-34}

\textbf{Figure 1.6.} Thermally activated ligand dissociation and covalent binding to DNA of cisplatin.

However, a drawback of cisplatin treatment is drug resistance, which led to the search for new platinum based drugs, such as oxaliplatin and carboplatin. These Pt based drugs possess different thermally labile ligands than the aforementioned chlorides in cisplatin and, in some cases display antitumor activity towards cisplatin-resistant cell lines in specific cancers.\textsuperscript{35,36} In addition, because all of these thermally activated drugs are active towards rapidly dividing cells, they are also effective towards healthy cells, resulting in detrimental side effects. One approach to combat the poor selectivity of current anticancer drugs is to develop a new treatment strategy where an external source
can be used to activate the drug, such as the use of light instead of body heat. The use of light for drug activation is known as photodynamic therapy (PDT).

1.3.2. Photodynamic Therapy and Photochemotherapy

The principle of PDT is to use light to activate a molecule that is then able to produce a reactive species that ultimately causes cell death. An important point for a successful PDT agent is that the molecule should be nontoxic in the dark, such that it is only activated by light. PDT provides low systemic toxicity, low levels of invasiveness, and increased selectivity, in some cases making it superior to conventional cancer therapies.\(^{37,38}\) To date, the field of PDT has focused nearly exclusively on organic molecules that produce $^1\text{O}_2$ as the reactive species when they absorb light. Because malignant tumors are often hypoxic (containing low levels of oxygen), there is a critical need to develop new drugs whose action is independent of oxygen. PDT agents that can potentially function in hypoxic environments is referred to as photochemotherapy (PCT). To this end, a molecule that forms metal-DNA bonds in a manner analogous to cisplatin, but only upon irradiation, would be ideal for PCT.

1.3.3. Ru(II) Complexes for PCT.

It is clear in Figure 1.6 that the active species involved in the therapeutic mechanism of cisplatin is the diaqua complex, $[\text{Pt(NH}_3)_2(H_2\text{O})_2]^{2+}$. Previously research showed that irradiation of $\text{I}$ ($[\text{Ru(bpy)}_2(\text{CH}_3\text{CN})_2]^{2+}$) in $\text{H}_2\text{O}$ results in ultrafast ligand exchange with high quantum efficiency forming the diaqua species $[\text{Ru(bpy)}_2(\text{H}_2\text{O})_2]^{2+}$,
and no ligand exchange is observed in the dark (Figure 1.4). The resulting diaqua species resembles the active form of cisplatin and can be considered a photogenerated cisplatin analog. In order to examine whether 1 could function as a PCT agent, preliminary DNA binding gel mobility assays were carried out and are shown in Figure 1.7. It is well documented that cisplatin thermally binds to linearized DNA and reduces its migration through an agarose gel in a concentrated dependent manner. The same trend is observed for 1 upon irradiation with visible light, but not in the dark. In Figure 1.7, lanes 1 and 8 contained 1 kb DNA ladder, lanes 2–7 were loaded with 50 mM pUC18 DNA, and lanes 3–6 contained increasing concentrations of 1 or cisplatin. It is evident in Figure 1.7a, that as the concentration of cisplatin is increased and incubated with DNA for 20 minutes the DNA mobility decreased. However, when 1 was incubated with DNA in the dark under the same experimental conditions no shift in mobility was observed (Figure 1.7b). The results are indicative of covalent binding of cisplatin to DNA in the dark but not for 1. In Figure 1.7c, complex 1 and DNA in lanes 3–6 were irradiated for 20 minutes with ≥ 420 nm light prior to loading. A similar pattern to cisplatin in Figure 1.7a was observed indicating that 1 only covalently binds to linearized DNA upon irradiation and could function as a PCT agent.
Figure 1.7. DNA binding gel mobility assays of (a) cisplatin incubated with DNA in the dark, (b) [Ru(bpy)$_2$(CH$_3$CN)$_2$]$^{2+}$ incubated with DNA in the dark, and (c) [Ru(bpy)$_2$(CH$_3$CN)$_2$]$^{2+}$ irradiated with ≥ 420 nm light in the presence of DNA.

Another area that has been explored with Ru(II) complexes as PCT agents is the release of bioactive molecules from the metal center. This was recently investigated by with cysteine protease inhibitors that were modified through their attachment to Ru(II) with nitrile groups in the complex [Ru(bpy)$_2$(L)$_2$]$^{2+}$ (2, L = cysteine protease inhibitor, Figure 1.8).$^{39}$ Cysteine cathepsins are overexpressed in a variety of cancers,$^{40}$ but are necessary for normal cell function.$^{41,42}$ Caging the inhibitor in 2 allows for selective enzyme inhibition locally in the tumor when irradiated with visible light. This process is depicted in Figure 1.8. A two-fold increase in enzyme inhibition was observed with 2 when irradiated with ≥ 395 nm light relative to the dark indicating successful caging of the inhibitor. Further exploration of the photorelease of other bioactive molecules for enzyme inhibition has also been conducted and is an ongoing area of research.$^{43-45}$ A criterion that is important in PCT, is that the complex absorbs light in the therapeutic
window (600-850 nm) to initiate the photoinduced ligand dissociation process. This window is where biological molecules and water do not absorb the least amount of light, therefore photons in this range can penetrate tissue optimally. Although 1 has been shown to covalently bind DNA and release bioactive molecules for enzyme inhibition when irradiated and is inactive in the dark, it requires irradiation with 455 nm light.

**Figure 1.8.** Molecular representation of the cysteine protease inhibitor and corresponding Ru(II) complex (top) and the proposed mechanism for enzyme inhibition upon irradiation (bottom).

### 1.3 Tuning Excited States for PCT.

This dissertation describes the development of new Ru(II) complexes capable of efficient ligand dissociation in the PCT window while gaining a better understanding of the factors that govern the overall excited state process. In order to red-shift the MLCT absorption maximum, ligands with greater aromaticity were utilized such as 2,2'-
biquinoline (biq), which possess stabilized $\pi^*$ orbitals relative to those of bpy. However, one needs to consider that lowering the energy of the $^3\text{MLCT}$ state while the energy of the $^3\text{dd}$ state remains the same increases the energy gap between the two states and most likely result in lower ligand exchange efficiency. It is possible to tune the excited states in Ru(II) complexes as shown in Figure 1.9. The diagram in the center of Figure 1.9 is of typical Ru(II) systems modeled after $[\text{Ru(bpy)}_3]^{2+}$ while the state diagram on the left represents a lowering of the $^3\pi\pi^*$ state energy below the $^3\text{MLCT}$ state and the state diagram on the right represents a lowering of the $^3\text{dd}$ state energy below the $^3\text{MLCT}$ state.

![State Diagram](image)

**Figure 1.9.** Possible state diagrams with lowest lying energy $^3\pi\pi^*$ or $^3\text{dd}$ states relative to $[\text{Ru(bpy)}_3]^{2+}$.

It has been shown that the addition of steric bulk distorts the pseudo-octahedral geometry about the Ru(II) metal center, thus elongating the Ru-L bonds and stabilizing the $^3\text{dd}$ state(s), and can lower them below the $^3\text{MLCT}$ state(s) resulting in more efficient ligand exchange. This phenomenon was observed in the complex $[\text{Ru(biq)}_2(\text{phen})]^{2+}$, which displayed cytoxicity with 650 nm light. The steric strain imposed by the bulky
biq ligands promotes bidentate ligand exchange with two solvent molecules upon irradiation (Figure 1.10). The concept of photoinduced bidentate ligand exchange is further analyzed in Chapter 5.

![Figure 1.10](image)

Figure 1.10. Photoinduced bidentate ligand dissociation of [Ru(biq)₂(phen)]²⁺ in H₂O.

Figure 1.9 also shows a state diagram where the $^3\pi\pi^*$ state is lower in energy than the $^3$MLCT state. This case is observed in the complex [Ru(bpy)₂(dppn)]²⁺ (dppn = dppn = benzo[i]dipyrido[3,2-a:2',3'-c]phenazine).⁴⁷ The long extended aromatic structure of the dppn ligand results in a low lying $^3\pi\pi^*$ state and a significantly greater excited state lifetime of $\tau = 33 \mu$s. It was found that this increased lifetime results in efficient $^1$O₂ production and DNA photocleavage upon excitation. Control of excited states was further tuned in the complex [Ru(bpy)(dppn)(CH₃CN)]²⁺, which is capable of both photoinduced ligand exchange and $^1$O₂ production through population of both the $^3$dd states and $^3\pi\pi^*$ state, and functions as a dual action PCT agent.⁴⁸ Furthermore, the dual
action complex was more cytotoxic than complexes that explicitly underwent ligand exchange or \( ^1\text{O}_2 \) production.

This dissertation gives a detailed analysis using steady-state and time resolved spectroscopy of new Ru(II) complexes with properties such as selective photoinduced ligand exchange, bidentate photoejection, photoinduced ligand exchange in the PCT window, as well as bimetallic photoactive Ru(II) complexes capable of releasing multiple drugs and new dual action complexes that produce \( ^1\text{O}_2 \) and undergo photoinduced ligand exchange. The results herein provide a better picture of the photoinduced ligand exchange process in Ru(II) complexes and show that the excited states can be tuned for by judicious choice of bidentate ligands for applications in PCT.

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Chapter 2: Experimental Methods

2.1 Materials

RuCl\(_3\)•3H\(_2\)O and 2,2´-biquinoline (biq) were purchased from CP Labs and Acros Organic, respectively, and were used without further purification. 2,3-naphtaldenediamine was purchased from Spectrum, 2-pyridinecarboxaldehyde from Acros Organics, and \(p\)-toluidine from TCI and were used as received. Ascorbic acid, 1,10-phenanthroline (phen), 4,7-dimethyl-1,10-phenanthroline (4,7-dmphen), 5,6-dimethyl-1,10-phenanthroline (5,6-dmphen), 2,9-dimethyl-1,10-phenanthroline (2,9-dmphen), 2,2´-bipyridine (bpy), 6,6´-dimethyl-2,2´-bipyridine (6,6´-dmbpy), 2,2´;6´;2´´-terpyridine (tpy), 2,2´-bipyrimidine (bpm), 2,2´-pyridil, potassium hexafluorophosphate, and ammonium hexafluorophosphate were purchased from Sigma Aldrich and used as received. All solvents used were purchased from commercial sources and used without further purification unless otherwise specified. The ligands 3,3´-isobiquinoline, 1,1´-isobiquinoline, and 1,12-diazaperylene (DAP) were prepared by the Thummel lab at the University of Houston. The ligands 2-\(p\)-tolylpyridinecarboxaldimine (PTPI),\(^1\) 2-(phenylazo)pyridine (PAP),\(^2\) 2,3-bis(2-pyridyl)benzoquinoxaline (dpb),\(^3\) and 1,4-bis(2-pyridylmethyleneamino)benzene (pbp)\(^4\) were prepared according to literature procedures. The complexes Ru(phen)Cl\(_4\),\(^5\) [Ru(phen)(CH\(_3\)CN)\(_4\)](PF\(_6\))\(_2\),\(^6\) Ru(phen)\(_2\)Cl\(_2\),\(^7\)
[Ru(biq)₂(bpy)][PF₆]₂, [Ru(biq)₂(phen)][PF₆]₂, Ru(PTPI)₂Cl₂, Ru(bpy)Cl₄, Ru(bpy)(DMSO)₂Cl₂, Ru(phen)(DMSO)₂Cl₂, Ru(bpy)(PTPI)₂Cl₂, and {[Ru(tpy)Cl₂(bpm)}[PF₆]₂ were also prepared according to literature procedures. The complexes [Ru(tpy)(bpy)(py)]²⁺ (py = pyridine), [Ru(tpy)(6,6'-dmbpy)(py)]²⁺, [Ru(tpy)(biq)(py)]²⁺, [Ru(tpy)(dppn)(py)]²⁺ (dppn = benzo[i]dipyrido[3,2-a;2',3'c]phenazine), and [Ru(tpy)(dmdppn)(py)]²⁺ (dmdppn = 3,6-dimethyl-dppn) that are discussed in Chapter 8 were prepared by Dr. Jessica Knoll according to reported procedures.

2.2 Synthesis

[Ru(biq)(phen)(CH₃CN)₂](PF₆)₂ (1). [Ru(phen)(CH₃CN)₄](PF₆)₂ (0.040 mg, 0.0544 mmol) and 2,2'-biquinoline (0.014 mg, 0.0542 mmol) were dissolved in 6 mL of DMF:CH₃CN (5:1). The yellow solution was stirred and purged for 5 minutes with N₂, and was then refluxed for 15 hours during which time a gradual color change from yellow to orange, then to light red was observed. The reddish orange solid was precipitated by the addition of 100 mL of H₂O and was filtered on a glass frit by vacuum filtration. The solid was dissolved in 20 mL of a CH₃CN:H₂O mixture (50:50) and heated under reflux for 4 hours, and 5 mL of a concentrated solution of NH₄PF₆ was added to the solution while still hot. The mixture was slowly cooled to room temperature then placed in an ice bath. A reddish orange solid precipitated and was filtered on a glass frit by vacuum filtration (0.015 mg, 31% yield). ¹H NMR (400 MHz, (CD₃)₂CO) δ 10.36 (dd, 1H, ³J = 5.2 Hz, ⁴J = 1.2 Hz) 9.25 (d, 1H, ³J = 8.9 Hz), 9.08 (m, 2H), 8.96 (dd, 1H, ³J =
8.2 Hz, $^4J = 1.2$ Hz), 8.79 (d, 1H, $^3J = 8.8$ Hz), 8.57 (m, 2H), 8.42 (m, 2H), 8.2 (m, 3H), 8.03 (t, 1H, $^3J = 7.4$ Hz), 7.87 Hz (m, 3H), 7.73 (m, 1H), 7.57 (t, 1H, $^3J = 3.5$ Hz), 7.34 (t, 1H, $^3J = 4.2$ Hz) 2.71 (s, 3H), 2.30 (s, 3H). Elem. anal. calcd. for [Ru(biq)(phen)(CH$_3$CN)$_2$](PF$_6$)$_2$·(C$_5$H$_5$)$_2$O·2H$_2$O: C, 48.4%; N, 9.34%; H, 3.70%. Found: C, 48.10%; N, 6.62%; H, 4.25%.

**Figure 2.1.** $^1$H NMR spectrum of 1 in (CD$_3$)$_2$CO. The resonance at ~2.84 ppm corresponds to residual water.

[Ru(phen)$_2$(CH$_3$CN)$_2$](PF$_6$)$_2$ (2). Ru(phen)$_2$Cl$_2$ (0.055g, 0.10 mmol) was dissolved in 20 mL of a mixture of CH$_3$CN and H$_2$O (50:50, v:v) and was refluxed for 4 hours. The solution was slowly cooled to room temperature and the solvent was evaporated to dryness by blowing with air. The remaining yellow solid was dissolved in 15 mL of H$_2$O washed with 5 aliquots of 20 mL CH$_2$Cl$_2$ until the organic layer was clear. CH$_3$CN (10 mL) was added to the aqueous layer and the mixture was refluxed for 1 hour. A saturated
solution of NH$_4$PF$_6$ in water (5 mL) was added to the solution while hot, the mixture was allowed to cool slowly to room temperature, and was then placed in an ice bath. A yellow solid precipitated and the powder was collected by vacuum filtrations and washed with 20 mL of diethyl ether (0.053 mg, 62% yield). $^1$H NMR (400 MHz, (CD$_3$)$_2$CO) $\delta$ 10.05 (dd, 2H, $^3$J = 5.3 Hz, $^4$J = 1.2 Hz), 9.05 (dd, 2H, $^3$J = 8.3 Hz, $^4$J = 1.3 Hz), 8.64 (dd, 2H, $^3$J = 7.1 Hz, $^4$J = 1.4 Hz), 8.46 (d, 2H, $^3$J = 9.1 Hz), 8.37 (m, 2H) 8.32 (d, 2H, $^3$J = 9.0 Hz), 8.09 (dd, 2H, $^3$J = 5.3 Hz, $^4$J = 1.2 Hz) 2.45 (s, 6H). Elem. anal. calcd. for [Ru(phen)$_2$(CH$_3$CN)$_2$](PF$_6$)$_2$: C, 40.3%; N, 10.1%; H, 2.66%. Found: C, 40.3%; N, 9.96%; H, 2.74%.

**Ru(biq)$_2$Cl$_2$.** Ru(biq)$_2$Cl$_2$ was prepared using a modification of the synthesis reported by Kubow et al.$^{15}$ RuCl$_3$.H$_2$O (0.18 g, 0.67 mmol), 2,2′-biquinoline (0.37 g, 1.5 mmol), and LiCl (0.087 g, 2.1 mmol) were dissolved in 7 mL of N,N-dimethylformamide. The solution was stirred until all solids dissolved, was purged with N$_2$ for 5 min, and was then refluxed for 6 hours turning a dark green color. The reaction mixture was slowly cooled to room temperature and pipetted dropwise into 500 mL of stirring H$_2$O, forming a green/blue precipitate that was collected by vacuum filtration. The solid was dissolved in CH$_2$Cl$_2$ forming a dark green solution, and then filtered to get rid of any remaining solid that did not dissolve. The green filtrate was washed 5 times with 20 mL of H$_2$O and then evaporated to a minimal amount of CH$_2$Cl$_2$. An excess of diethyl ether was added to the green CH$_2$Cl$_2$ solution, resulting in the formation of a green precipitate that was collected by vacuum filtration (0.22 g, 48% yield).
[Ru(biq)$_2$(CH$_3$CN)$_2$](PF$_6$)$_2$ (3). A procedure analogous to that for [Ru(phen)$_2$(CH$_3$CN)$_2$]$^{2+}$ was followed but using Ru(biq)$_2$Cl$_2$ (0.055 g, 0.080 mmol) as the starting material, which resulted in the isolation of a maroon powder (0.043 g, 54% yield). $^1$H NMR (400 MHz, (CD$_3$)$_2$CO) $\delta$ 8.75 (d, 2H, $^3$$J$ = 8.7 Hz), 8.42 (d, 2H, $^3$$J$ = 8.2 Hz), 8.34 (d, 2H, $^3$$J$ = 8.1 Hz), 8.20 (m, 4H), 8.01 (m, 4H), 7.92 (d, 2H, $^3$$J$ = 8.0 Hz), 7.46 (t, 2H, 7.1 Hz), 6.80 (m, 4H), 2.46 (s, 6H). Elem. anal. calcd. for [Ru(biq)$_2$(CH$_3$CN)$_2$](PF$_6$)$_2$·C$_2$H$_6$O: C, 48.7%; N, 8.52%; H, 3.07%. Found: C, 48.9%; N, 8.15%; H, 3.52%.

[Ru(4,7-dmphen)(phen)(CH$_3$CN)$_2$][PF$_6$]$_2$ (4). The complex Ru(4,7-dmphen)(phen)Cl$_2$ was synthesized according to a modified literature procedure.$^{16}$ Ru(phen)(DMSO)$_2$Cl$_2$ (0.050 g, 0.098 mmol) and 4,7-dimethylphenanthroline (0.020 g, 0.096 mmol) were dissolved in 8 mL of DMF. The reaction mixture was heated to reflux for 4 hours upon which a change from an orange to a purple color was observed. The mixture was cooled to room temperature and the solvent was evaporated under vacuum. The crude product was dissolved in ~30 mL of CH$_2$Cl$_2$ and washed with 20 mL aliquots of H$_2$O until no orange color was observed in the aqueous layer. The purple CH$_2$Cl$_2$ solution was concentrated and a purple solid was precipitated by the addition of excess ether. The solid was filtered and washed with ~50 mL of H$_2$O and ~50 mL of ether.

The resulting purple precipitate, Ru(4,7-dmphen)(phen)Cl$_2$ (0.020 g, 0.036 mmol), was suspended in a 10 mL mixture of CH$_3$CN and H$_2$O (50:50) and heated to reflux for
15 hours. A saturated aqueous solution of NH₄PF₆ (5 mL) was added to the resulting bright orange solution while hot. The mixture was cooled to room temperature and the solvent was reduced to half the original amount by blowing with air. The resulting orange/yellow precipitate was filtered and washed with excess ether (0.011 g, 55% yield).

$^1$H NMR (400 MHz, (CD$_3$)$_2$CO) δ 10.04 (dd, 1H), 9.88 (d, 1H), 9.05 (d, 1H), 8.65 (d, 1H), 8.59 (d, 1H), 8.46 (dd, 2H), 8.35 (m, 2H), 8.22 (d, 1H), 8.08 (d, 1H), 7.93 (d, 1H), 7.60 (m, 1H), 7.44 (d, 1H) 3.18 (s, 3H), 2.86 (s, 3H), 2.47 (s, 3H), 2.42 (s, 3H).

**Figure 2.2.** $^1$H NMR spectrum of 4 in (CD$_3$)$_2$CO.

[Ru(5,6-dmphen)(phen)(CH$_3$CN)$_2$][PF$_6$]$_2$ (5). A procedure analogous to that for [Ru(4,7-dmphen)(phen)(CH$_3$CN)$_2$][PF$_6$]$_2$ was followed but using 5,6-dimethylphenanthroline (0.008 g, 40% yield). $^1$H NMR (400 MHz, (CD$_3$)$_2$CO) δ 10.04 (d, 1H), 9.99 (d, 1H), 9.18 (d, 1H), 9.06 (d, 1H), 8.76 (d, 1H), 8.65 (d, 1H), 8.47 (d, 1H),
8.35 (m, 3H), 8.05 (m, 2H), 7.61 (m, 2H), 2.98 (s, 3H), 2.85 (s, 3H), 2.47 (s, 3H) 2.45 (s, 3H).

[Ru(2,9-dmphen)(phen)(CH$_3$CN)$_2$][PF$_6$]$_2$ (6). The complex was synthesized from the photochemical reaction of [Ru(2,9dmphen)(phen)]$^{2+}$ in CH$_3$CN. [Ru(2,9dmphen)$_2$(phen)]$^{2+}$ was synthesized by refluxing 1 equivalent of [Ru(phen)(CH$_3$CN)$_4$][PF$_6$]$_2$ (0.04 g, 0.054 mmol) with 2 equivalents of 2,9-dimethylphenanthroline (0.023 mg, 0.11 mmol) in DMF (5 mL) for one hour. The resulting orange solution was cooled to room temperature and added to 100 mL of H$_2$O while stirring. The orange solid that precipitated was filtered and washed with ether.

The orange powder, [Ru(2,9dmphen)$_2$(phen)][PF$_6$]$_2$ (0.020 g, 0.023 mmol), was dissolved in ~20 mL of CH$_3$CN and irradiated with ≥ 345 nm light while stirring for 14 hours in which a color change from orange to yellow was observed. The solution was evaporated to dryness by blowing with air and the product was dissolved in minimal acetone. Excess ether was added to precipitate a yellow/orange product which was filtered and washed with additional ether (0.009 g, 56% yield). $^1$H NMR (400 MHz, (CD$_3$)$_2$CO) δ 10.21 (d, 1H), 9.02 (d, 1H), 8.88 (d, 1H), 8.60 (d, 1H), 8.42 (m, 2H), 8.25 (m, 4H), 8.15 (d, 1H), 7.73 (d, 1H), 7.57 (m, 1H), 7.47 (d, 1H), 3.47 (s, 3H), 2.68 (s, 3H), 2.23 (s, 3H), 2.09 (s, 3H).

[Ru(3,3'-isobiq)(phen)(CH$_3$CN)$_2$][PF$_6$]$_2$ (7). A procedure analogous to that for [Ru(4,7-dmphen)(phen)(CH$_3$CN)$_2$][PF$_6$]$_2$ was followed but using the 3,3'-isobiq ligand (0.012 g,
60% yield). $^1$H NMR (400 MHz, (CD$_3$)$_2$CO) $\delta$ 10.38 (s, 1H), 10.04 (dd, 1H), 9.42 (s, 1H), 9.23 (s, 1H), 9.05 (dd, 1H), 8.65 (dd, 1H), 8.52 (s, 1H), 8.42 (m, 5H), 8.31, (d, 1H), 8.13 (m, 2H), 8.04 (m, 1H), 7.85 (m, 1H), 7.68 (m, 1H), 7.62 (m, 2H), 2.52 (s, 3H), 2.42 (s, 3H).

Figure 2.3. $^1$H NMR spectrum of 7 in (CD$_3$)$_2$CO. The resonances at ~2.74 ppm and 2.81 ppm correspond to residual water.

[Ru(1,1ʻ-isobiq)(phen)(CH$_3$CN)$_2$][PF$_6$]$_2$ (8). A procedure analogous to that for [Ru(4,7-dmphen)(phen)(CH$_3$CN)$_2$][PF$_6$]$_2$ was followed but using the 1,1ʻ-isobiq ligand (0.0090 g, 45% yield). $^1$H NMR (400 MHz, (CD$_3$)$_2$CO) $\delta$ 9.96 (d, 1H), 9.63 (d, 1H), 9.03 (m, 1H), 8.81 (d, 1H), 8.62 (m, 2H), 8.52 (d, 1H), 8.44 (m, 2H), 8.36 (m, 2H), 8.31, (m, 1H), 8.12 (m, 4H), 7.76 (m, 2H), 7.70 (m, 2H), 7.61 (m, 1H), 2.46 (s, 3H), 2.30 (s, 3H).
[Ru(DAP)(phen)(CH$_3$CN)$_2$][PF$_6$]$_2$ (9). A procedure analogous to that for [Ru(4,7-dmphen)(phen)(CH$_3$CN)$_2$][PF$_6$]$_2$ was followed but using the DAP ligand. However, in the purification of the chloride precursor Ru(DAP)(phen)Cl$_2$, the product was only partially soluble in CH$_2$Cl$_2$. Rather than saving the organic layer during washing with water, it was also discarded and only the undissolved solid was used to synthesize the desired CH$_3$CN complex (0.0060 g, 30%). $^1$H NMR (400 MHz, (CD$_3$)$_2$CO) $\delta$ 10.10 (dd, 1H), 9.62 (m, 1H), 9.10 (dd, 1H), 9.00 (m, 2H), 8.93 (m, 1H), 8.67 (dd, 1H), 8.55 (dd, 1H), 8.35 (dd, 1H), 8.24 (m, 2H), 8.08 (m, 2H), 7.91 (dd, 1H), 7.81 (dd, 1H), 7.76 (dd, 1H), 7.64 (m, 1H), 2.48 (s, 3H), 2.45 (s, 3H).

$\alpha$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$][PF$_6$]$_2$ (10). cis-Ru(PTPI)$_2$Cl$_2$ (0.050 g, 0.089 mmol) was suspended in a 50:50 mixture of CH$_3$CN and H$_2$O (10 mL) and heated to reflux for 15 hours. A saturated aqueous NH$_4$PF$_6$ (5 mL) solution was added to the resulting orange solution while hot. After cooling, the orange precipitate was collected by vacuum filtration and washed with diethyl ether. Purification was achieved by growing single crystals by slow vapor diffusion. The orange solid was dissolved in a solvent mixture of THF, CH$_3$CN, and acetone (1:1:1) to produce a nearly saturated solution and diethyl ether was then slowly diffused into the solvent mixture at ~ 0° C (0.0042 g, 5.5% yield). $^1$H NMR (400 MHz (CD$_3$)$_2$CO): $\delta$ 9.37 (d, 2H), 8.93 (s, 2H), 8.19 (m, 4H), 7.79 (m, 2H), 7.05 (d, 4H), 6.95 (d, 4H), 2.62 (s, 6H), 2.21 (s, 6H).
\[ \alpha-[\text{Ru}(\text{PTPI})_2(\text{CH}_3\text{CN})_2][\text{PF}_6]_2 \text{ (11)} \]. \textalpha-[\text{Ru}(\text{PTPI})_2(\text{CH}_3\text{CN})_2][\text{PF}_6]_2 \text{ (0.015 g, 0.017 mmol) was dissolved in ~15 mL of CH}_3\text{CN and the solution was irradiated with } \geq 395 \text{ nm light for 3 h and then evaporated to dryness under a stream of air. The resulting solid was re-dissolved in a minimal volume of acetone and excess diethyl ether was added to produce an orange precipitate which was collected by vacuum filtration. The solid was purified by the previously described method of growing single crystals used for the isolation of the } \alpha \text{ isomer (0.0034 g, 23% yield).} \text{ }^1\text{H NMR (400 MHz (CD}_3\text{)}_2\text{CO): } \delta 9.45 \text{ (s, 2H), 8.64 (d, 2H), 8.44 (d, 1H), 8.20 (m, 2H), 7.86 (d, 4H), 7.71 (m, 2H), 7.55 (d, 4H), 2.47 (s, 6H), 2.39 (s, 6H).} \]
γ-[Ru(PTPI)$_2$(CH$_3$CN)$_2$][PF$_6$]$_2$ (12). A procedure analogous to that of the α isomer was followed but using trans-Ru(PTPI)$_2$Cl$_2$ (0.030 g, 0.053 mmol) which resulted in an orange powder. Purification of the isomer was carried out by the same single crystal growth technique as described above which was isolated by selective harvesting with the use of a microscope as amber colored crystals (0.0015 g, 3.1% yield). $^1$H NMR (400 MHz (CD$_3$)$_2$CO): δ 9.70 (d, 2H), 9.08 (s, 2H), 8.48 (m, 4H), 8.08 (m, 2H), 6.97 (d, 4H), 6.86 (d, 4H), 2.39 (s, 6H), 2.35 (s, 6H).

δ-[Ru(PTPI)$_2$(CH$_3$CN)$_2$][PF$_6$]$_2$ (13). A procedure analogous to the γ isomer was used. Purification of the isomer was achieved as described above and then isolated with the use of a microscope as red crystals (0.0021 g, 4.6%). $^1$H NMR (400 MHz (CD$_3$)$_2$CO): δ 9.28 (s, 2H), 8.43 (d, 2H), 8.28 (m, 1H), 7.65 (d, 2H), 7.55 (m, 10H), 2.54 (s, 6H), 2.42 (s, 6H).

Figure 2.5. $^1$H NMR spectrum of 11 in (CD$_3$)$_2$CO.
**cis-[Ru(bpy)(PTPI)(CH$_3$CN)$_2$][PF$_6$]$_2$** (14). Ru(bpy)(PTPI)Cl$_2$ was prepared by a modification of the literature procedure.$^{16}$ Ru(bpy)(DMSO)$_2$Cl$_2$ (0.050 g, 0.10 mmol) and 1 eq of PTPI ligand (0.021 g, 0.11 mmol) were dissolved in 6 mL of DMF and heated to reflux for 3 hours. The resulting purple solution was evaporated to dryness *in vacuo* and dissolved in 20 mL of CH$_2$Cl$_2$. The CH$_2$Cl$_2$ solution was washed with successive 20 mL aliquots of water until no color was observed in the aqueous layer. The CH$_2$Cl$_2$ solution was evaporated to a minimal volume and a purple solid was precipitated by the addition of excess diethyl ether and collected by vacuum filtration. The purple solid (0.020 g, 0.038 mmol) was refluxed in 8 mL of a CH$_3$CN:H$_2$O mixture (4 mL:4 mL) for 15 hours. A saturated solution of aqueous NH$_4$PF$_6$ (5 mL) was added to the resulting hot orange solution. After cooling, an orange precipitate was collected by vacuum filtration and washed with diethyl ether (0.011 g, 31% yield). $^1$H NMR (400 MHz (CD$_3$)$_2$CO): $\delta$ 9.63 (d, 1H), 9.34 (d, 1H), 9.05 (s, 1H), 8.45 (m, 4H), 8.10 (m, 3H), 7.71 (d, 1H), 7.69 (m, 1H), 7.55 (m, 1H), 6.89 (d, 2H), 6.55 (d, 2H), 2.62 (s, 3H), 2.51 (s, 3H), 2.16 (s, 3H). Elem. Anal. Calcd. for [Ru(bpy)(PTPI)(CH$_3$CN)$_2$][PF$_6$]$_2$: C, 39.28%; N, 10.18%; H, 3.18%. Found: C, 39.01%; N, 10.10%; H, 3.10%.
cis-[Ru(bpy)(PAP)(CH$_3$CN)$_2$][PF$_6$]$_2$ (15). Prepared by Bruno Peña of Texas A&M University according to a published procedure.$^{17}$

[Ru(biq)$_2$(phpy)](PF$_6$) (16). Prepared by Bruno Peña of Texas A&M University according to a published procedure.$^{18}$

[Ru(biq)$_2$(dpb)](PF$_6$) (17). Ru(biq)$_2$Cl$_2$ (0.100 g, 0.15 mmol) and the dpb ligand (0.055 g, 0.16 mmol) were suspended in a 5:1 mixture of EtOH and H$_2$O (20 mL). Prior to heating 2 equivalents (0.075 g, 0.29 mmol) of silver triflate were added to the reaction mixture and the vessel was degassed with N$_2$. The mixture was then heated to reflux while stirring for 72 hours resulting in a deep purple color. The solution was cooled to

Figure 2.6. $^1$H NMR spectrum of 14 in (CD$_3$)$_2$CO.
room temperature and the solvent mixture was filtered through celite. A saturated aqueous solution (10 mL) of NH₄PF₆ was added to the filtrate resulting in a purple precipitate and collected by vacuum filtration. The solid was purified on a neutral alumina column using an acetone:toluene mixture (3:2) as the eluent. The free dpb ligand (yellow band) eluted first and the dark purple band was then collected. The solvent was evaporated to a minimal amount by blowing with air and a purple solid was precipitated by the addition of excess ether and collected by vacuum filtration. (0.057g, 31 % yield). ¹H NMR (400 MHz CD₂Cl₂): δ 8.85 (s, 1H), 8.76 (d, 1H), 8.68 (m, 3H) 8.51 (d, 1H), 8.00 (m, 13H), 7.64 (m, 5H) 7.40 (m, 8H), 7.01 (m, 4H), 6.69 (m, 1H), 6.47 (1H, d). MS: m/z 474. Elem. Anal. Calcd. for [Ru(biq)₂(dpdb)]PF₆₂·3H₂O: C, 53.91%; N, 8.67%; H, 3.43%. Found: C, 53.40%; N, 8.64%; H, 3.25%.

Figure 2.7. ¹H NMR spectrum of 17 in CD₂Cl₂.
\{[\text{Ru(CH}_3\text{CN)}_3]_2(tppz)}[\text{PF}_6]_4\) (18). Prepared by Bruno Peña of Texas A&M University according to a published procedure.\textsuperscript{19}

\{[\text{Ru(tpy)(CH}_3\text{CN)}_2(bpm)}][\text{PF}_6]_4\) (19). A mixture of \textit{cis} and \textit{trans} 
\{[\text{Ru(tpy)Cl}_2(bpm)}][\text{PF}_6]_2\ (0.100 \text{ g, 0.084 mmol}) was suspended in a CH\textsubscript{3}CN:H\textsubscript{2}O solvent mixture (50:50). While hot, 4 equivalents (0.085 \text{ g, 0.33 mmol}) of silver triflate were added to the dark stirring mixture and the solution was continued to reflux for 16 hours. While hot the solution was filtered through celite. Upon cooling, a brown precipitate formed and was collected by vacuum filtration. The solid was dissolved in minimal acetone, centrifuged, and the supernatant was subjected to excess ether dropwise to precipitate the solid. It was collected by vacuum filtration and was confirmed to be the \textit{cis} and \textit{trans} isomers of the desired product (0.054 \text{ g, 43\% yield}). The \textit{cis} isomer was isolated using single crystal growth by slow vapor diffusion of ether into CH\textsubscript{3}CN. \textsuperscript{1}H NMR (400 MHz (CD\textsubscript{3})\textsubscript{2}CO): \delta 10.51 (d, 2H), 8.85 (d, 4H), 8.70 (d, 4H), 8.57 (d, 4H), 8.51 (t, 2H), 8.19 (m, 4H), 8.03 (d, 2H), 7.66 (m, 4H), 7.12 (t, 2H), 2.40 (s, 6H).

\{[\text{Ru(bpy)(CH}_3\text{CN)}_2]_2(pbpp)}][\text{PF}_6]_4\) (20). Ru(bpy)((DMSO)\textsubscript{2}Cl\textsubscript{2} (0.045 \text{ g, 0.093 mmol}) and the pbp ligand (0.013 \text{ g, 0.0045 mmol}) were dissolved in approximately 6 \text{ mL} of DMF. The mixture was heated to reflux for 3 hours in which the solution color changed from orange to dark purple. After cooling to room temperature the solvent was removed
in vacuo. The resulting crude mixture was suspended in CH$_2$Cl$_2$ and a purple solid was collected by vacuum filtration. The solid was washed with several aliquots of water (5 x 15 mL) and ether (2 x 15 mL) yielding the precursor complex \{[Ru(bpy)Cl$_2$(pbp)]\}. \{[Ru(bpy)Cl$_2$(pbp)]\} (0.010 g, 0.011 mmol) was suspended in 8 mL of CH$_3$CN:H$_2$O (50:50) and heated to reflux for 16 hours in which the solution color changed from purple to orange. While hot, a saturated aqueous solution of NH$_4$PF$_6$ (5 mL) was added. Upon cooling an orange solid precipitated and was collected by vacuum filtration and washed with ether. The solid was purified using the same single crystal growth method as in \(\alpha\)-[Ru(PTPI)$_2$(CH$_3$CN)$_2$][PF$_6$]$_2$ (0.005 g, 31% yield). $^1$H NMR (400 MHz (CD$_3$)$_2$CO): $\delta$ 9.59 (d, 2H), 9.10 (d, 2H), 8.77 (s, 2H), 8.52 (d, 2H), 8.44 (td, 2H), 8.38 (d, 2H), 8.31 (d, 2H), 8.10 (m, 8H), 7.79 (d, 2H), 7.55 (m, 4H), 6.35 (s, 4H), 2.53 (s, 6H), 2.50 (s, 6H).

**Figure 2.8.** $^1$H NMR spectrum of 20 in (CD$_3$)$_2$CO; the resonances at ~2.76 ppm and 2.81 ppm correspond to residual water.
\([\{\text{Ru(phen)}(\text{CH}_3\text{CN})_2\text{2(pbpy)}\}\text{PF}_6\text{4}} (21). A procedure analogous to the complex \([\{\text{Ru(bpy)}(\text{CH}_3\text{CN})_2\text{2(pbpy)}\}\text{PF}_6\text{4}} was used but the starting material was Ru(phen)(DMSO)_2Cl_2 (0.003 g, 19% yield). \(^1\)H NMR (400 MHz (CD_3)_2CO): \(\delta\) 9.66 (d, 2H), 9.35 (dd, 2H), 8.72 (dd, 2H), 8.65 (dd, 2H), 8.60 (s, 2H), 8.56 (d, 2H), 8.47 (td, 2H), 8.20 (d, 4H), 8.11 (m, 4H), 7.84 (m, 4H), 5.93 (s, 4H), 2.55 (s, 6H), 2.36 (s, 6H).

2.3 Instrumentation

The \(^1\)H NMR spectra of all complexes were recorded using a Bruker 400 MHz DPX ultrashield system. Electrospray ionization (ESI) mass spectrometry data were collected on a Bruker micrOTOF instrument with methanol as the eluent. Electronic absorption spectroscopy was carried out using a Hewlett Packard 8453 diode array spectrometer, emission spectra were obtained on a Horiba Fluormax-4 spectrometer, and electrochemical studies were performed on a BAS CV-50W voltammetric analyzer. Steady state near-IR luminescence measurements at room temperature and 77 K were obtained on a home-built instrument utilizing an ultra-sensitive germanium detector (Edinburgh Instruments) powered by an Edinburgh PS-1 power supply and equipped with an Edinburgh MF-1 Muon Filter. The home built instrument was also equipped with a 101/102 monochromator (PTI) controlled by an SID-101 step motor (PTI). Excitation was achieved using 405 nm (RGBLase semiconductor laser, 45 mW, model FBB-4050450FS-1-0) or 658 nm (Newport, 65 mW, model LPM658-65C) that was optically
chopped (OC 4000, PTI) and a lock-in amplifier (Stanford Research Systems) was used to improve signal to noise ratio. Data were obtained using a home-built LabView program and was corrected using the provided manufacturer’s correction file. The home-built transient absorption instrument for measurements on the nanosecond timescales was previously described.20 Excitation was accomplished through the use of a frequency doubled or tripled Spectra Physics GCR-150 Nd:YAG laser ($\lambda_{ex} = 532$ or 355 nm, fwhm $\sim$ 8 ns) as the excitation source. Femtosecond transient absorption experiments were carried out using laser and detection systems that were previously described.21 The samples were excited with 310, 325, 350, or 568 nm light (1.5 mW at the sample) by the output of an optical parametric amplifier (OPA) with a sum frequency generation and ultraviolet visible harmonics attachment. Upon irradiation samples were kept in motion by use of a Harrick Scientific flow cell equipped with 1 mm CaF$_2$ windows (1 mm path length). A total volume of $\sim$5 mL was required for the flow cell to operate correctly. The polarization angle between the pump and probe beams was 54.7$^\circ$ to avoid rotational diffusion effects. The measurement at each time delay was repeated four times and the spectra were corrected for the chirp in the white light probe continuum.22 Photolysis and quantum yield experiments were carried out using a 150 W Xe short arc lamp (USHIO) in a Milliarci lamp housing unit (PTI) powered by a LPS-220 power supply (PTI) equipped with a LPS-221 igniter (PTI). The desired wavelength range was attained using bandpass filters (Thorlabs, fwhm$\sim$10 nm) or 3 mm thick (2 mm for 610 nm) long-pass filters (CVI Melles Griot). Gel imaging was performed with a Bio Rad Gel Doc 2000 transilluminator.
2.4 Methods

$^1$H NMR spectroscopy was performed in (CD$_3$)$_2$CO (acetone-$d_6$), CD$_3$CN, CD$_2$Cl$_2$, or C$_5$D$_5$N (py-$d_5$) and all resonances were referenced to the residual protonated solvent peak. In the photolysis experiments monitored by $^1$H NMR spectroscopy in CD$_3$CN or py-$d_5$, the intensities of the peaks were integrated relative to an internal standard of benzene (25 μL). The chloride salt of each complex was used for experiments performed in H$_2$O, which were obtained using an ion exchange column. The stationery phase was composed of Amberlite IRA-410 ion exchange resin prepared by soaking in 1 M HCl at 50 °C for 3 days and methanol was used as the eluent. Emission was measured at both room temperature and 77 K in CH$_3$CN in a 1x1 cm quartz cuvette using an excitation wavelength corresponding to the maximum of the MLCT absorption for each complex and 405 nm or 658 nm for complexes that emit in the near-IR. Cyclic voltammetry experiments were performed in a three-electrode cell with a Pt working electrode, a Pt wire auxiliary electrode, and a saturated Ag/AgCl reference electrode. The samples were dissolved in distilled CH$_3$CN or CH$_2$Cl$_2$ containing 0.1 M tetrabutylammonium hexafluorophosphate as the supporting electrolyte, and bubbled with N$_2$ for 10 minutes prior to each measurement. The cyclic voltammetry data was recorded at a scan rate of 100 mV/s, and ferrocene was added to each sample after the measurement as an internal standard (+0.40 V vs SCE in CH$_3$CN).$^{23}$ Elemental Analysis was performed by Atlantic Microlab Inc.

The quantum yields (Φ) for photoinduced ligand exchange of the first CH$_3$CN in H$_2$O were measured for the complexes using either 400 nm, 450 nm, 500 nm, 550 nm or
600 irradiation wavelengths using the appropriate bandpass filters. The moles of complex reacted was quantitated using electronic absorption spectroscopy by monitoring the decrease in MLCT absorption maximum of each complex as a function of irradiation time (moles reacted/s) at early irradiation times, and Reinecke’s salt or ferrioxalate was used as an actinometer to determine the intensity (Einstein/s) of the Xe arc lamp at the corresponding wavelengths. The Φ for the photoinduced ligand exchange of the second CH₃CN ligand was measured for 1 using 400 nm and 550 nm irradiation wavelengths using the appropriate bandpass filters monitoring the decrease in the MLCT absorption peak of the mono-aqua intermediate.

Crystals suitable for single X-ray diffraction were grown for 1, 4, 6, 7, 10 – 15, 20, and [Ru(biq)(phen)(CH₃CN)(py)]²⁺ by slow vapor diffusion. A ~ 2 mg sample was dissolved in a mixture of CH₃CN, THF, and acetone (0.25 mL, 0.10 mL, and 0.25 mL, respectively) in a small vial and inserted into a larger vial partially filled with ether, which was sealed and placed in the freezer resulting in red, yellow, or orange rods or platelets over a period of 2 weeks. Crystals suitable for single X-ray diffraction for complexes 3, 9, and 17 were grown in an analogous manner but were dissolved in CH₃CN (3 and 9) or a CH₃CN/toluene mixture (17) and slowly diffused with ether. X-ray diffraction experiments and data processing were performed by Christopher B. Durr.

Calculations were performed with density functional theory (DFT) using the Gaussian 09 program. The B3LYP functional along with the 6-31G* basis set for H, C, Cl, and N and the SDD energy consistent pseudopotentials were used for Ru. Model compounds were generated by replacing the methyl groups with hydrogen atoms.
on the acetonitrile groups. Optimization of full geometries were carried out with the respective programs and orbital analysis was performed in Gaussview version 3.09.\textsuperscript{32} Following optimization of the molecular structures, frequency analysis was performed to ensure the existence of local minima on the potential energy surfaces. Electronic absorption singlet to singlet transitions were calculated using time-dependent DFT (TD-DFT) methods with the polarizable continuum model (PCM) that mimicked the solvation effect of CH\textsubscript{3}CN in Gaussian 09.\textsuperscript{33} Singlet-triplet transitions were also calculated to generate difference density plots of the lowest energy triplet excited states.

The supercoiled pUC18 (Bayou Biolabs) used in the gel electrophoresis mobility shift assays was purified using a standard QIAprep\textsuperscript{®} Spin Miniprep Kit (QIAGEN). The DNA was bound to a miniprep spin column, washed with 500 μL PB buffer, with 750 μL PE buffer, and was extracted from the column with warm water. The purified DNA was then linearized using a QIAquick\textsuperscript{®} GelExtraction Kit (QIAGEN). For the linearization, 55 μL of water, 25 μL of pUC18, 10 μL of React 4 Buffer (Invitrogen), and 10 μL Sma1 enzyme (Invitrogen) were added to a small Eppendorf tube and heated at 30 °C for 1 hr, and the enzyme was then deactivated by incubating the sample at 65 °C for 10 minutes. QG buffer (300 μL) and isopropanol (100 μL) were added to the mixture, then bound to a spin column and washed with 750 μL of PE buffer, and was then extracted from the column with 40 μL of water. The agarose gel electrophoresis was carried out using 1 x TBE buffer (pH = 8.28) at room temperature for one hour at 95 V powered by an EC 105 voltmeter produced by E-C Apparatus Corporation. Gels were then stained in ethidium bromide solutions (0.5 μg/mL) for 30 minutes then soaked in water for 30 minutes.
For relative viscosity measurements calf-thymus (ct) DNA (Sigma) was purified by dialysis. Dialysis tubing was purchased from Sigma (D-9777) which retains DNA with molecular weight greater than 12,000. Ct-DNA (0.004 g) was dissolved in 2 mL of 5 mM Tris, 50 mM NaCl buffer adjusted to a pH of 7.2 by stirring vigorously in an Eppendorf tube for two hours. The 2 mL ct-DNA solution was added to the dialysis tubing and submerged in the buffer solution and stirred overnight. The solution of CT-DNA gave ratios of absorbance at 260 and 280 nm of 1.9:1, indicating it was sufficiently pure for further use. Measurements were carried out using a Cannon-Manning Semi-Micro viscometer (No. 50-N88) maintained at a constant temperature of 28 °C in a thermostatic bath. Flow time was measured with a digital stopwatch, and each sample was measured three times, and an average flow time was calculated. Data were presented as \(\eta/\eta_0^{1/3}\) vs [Ru]/[DNA], where \(\eta\) is the viscosity of DNA in the presence of the complex, and \(\eta_0\) is the viscosity of DNA alone.

Preliminary cell studies including uptake, dark toxicity, and phototoxicity with the human cervical adenocarcinoma cell line were performed by Christiane Pavani in the lab of Mauricio Baptista at Sao Paulo University in Brazil and are outlined in recent work. More specific conditions for these experiments are described in later chapters (excitation wavelength, concentration, etc.).

References


Chapter 3: Incorporation of the 2,2'-biquinoline Ligand in Ruthenium(II) Complexes with Photolabile CH$_3$CN Ligands


3.1 Introduction

Understanding the photochemistry of transition metal complexes is essential to the development of areas that include solar energy conversion, photocatalysis, and photochemotherapy (PCT).$^{1–8}$ These processes are initiated by the absorption of a photon by the molecule, placing it in an excited state that is able to undergo reactions that are not accessible from the ground state.$^{8–10}$ Ruthenium(II) complexes are of particular interest due to their success as sensitizers in dye sensitized solar cells,$^{2,8}$ as well as their potential as PCT agents.$^{11–14}$ In addition, many of these Ru(II) complexes have the ability to undergo excited state ligand substitution.$^{15}$

The broadly accepted model for the mechanism of photoinduced ligand exchange in complexes with lowest-energy $^3$MLCT (metal-to-ligand charge transfer) excited states relies on the thermal population of the reactive $^3$LF (ligand field) dd state(s) from the lower-lying $^3$MLCT state(s).$^{16–19}$ The population of the $^3$LF state(s) places electron density on the $e_g$-type orbitals with Ru–L $\sigma^*$ character, thus resulting in ligand
dissociation.\textsuperscript{16-19} Accordingly, the energy gap between the \textsuperscript{3}MLCT and the \textsuperscript{3}LF states has been shown to have a pronounced effect on the quantum yield of ligand exchange when the low-lying MLCT state is excited selectively.\textsuperscript{18} Moreover, direct excitation of the LF state(s) with higher energy light results in a significant increase in the photoreactivity.\textsuperscript{17}

The exploration of the photochemistry of Ru(II) complexes possessing monodentate CH\textsubscript{3}CN ligands such as [Ru(bpy)(CH\textsubscript{3}CN)\textsubscript{4}]\textsuperscript{2+} (bpy = 2,2$^\prime$-bipyridine) and [Ru(bpy)\textsubscript{2}(CH\textsubscript{3}CN)\textsubscript{2}]\textsuperscript{2+} revealed efficient exchange of the CH\textsubscript{3}CN ligands with coordinating solvent or with excess halide upon irradiation with visible light.\textsuperscript{20,21} It was shown that ligand exchange occurred in a stepwise manner and that the quantum yield for the exchange of the second CH\textsubscript{3}CN ligand is ~2-fold lower than that of the first.\textsuperscript{20,21} Moreover in [Ru(bpy)(CH\textsubscript{3}CN)\textsubscript{4}]\textsuperscript{2+}, which possesses four potential sites for exchange, only the axial CH\textsubscript{3}CN ligands undergo stepwise substitution with water upon irradiation.\textsuperscript{20} This reactivity provides an important synthetic tool for the preparation of new \textit{trans} tris-heteroleptic Ru(II) complexes, as well as PCT agents with photolabile ligands that can function in hypoxic environments without the need for oxygen.\textsuperscript{12-14}

As mentioned in Chapter 1, the therapeutic window for PCT is between 600 and 850 nm where biomolecules absorb weakly.\textsuperscript{22} The extension of the aromatic system of polypyridyl diimmine ligands bound to the Ru(II) metal by through the incorporation of the 2,2$^\prime$-biquinoline (biq) ligand has been shown to lower the energy of the lowest unoccupied molecular orbital (LUMO) resulting in a red shift of the \textsuperscript{1}MLCT absorption band.\textsuperscript{23} This red shift allows for photoinduced ligand exchange with lower energy
irradiation to occur, which would potentially make these types of complexes more favorable PCT agents.

In the present work, the asymmetric complex [Ru(biq)(phen)(CH$_3$CN)$_2$]$^{2+}$ (1, Figure 3.1) (phen = 1,10-phenanthroline, biq = 2,2’-biquinoline) was synthesized and characterized, and its photochemical properties were investigated. The results were compared to those of the symmetric complexes [Ru(phen)$_2$(CH$_3$CN)$_2$]$^{2+}$ (2, Figure 3.1) and [Ru(biq)$_2$(CH$_3$CN)$_2$]$^{2+}$ (3, Figure 3.1). Each of the complexes 1 – 3 possesses two potentially photolabile CH$_3$CN ligands, however, unlike 2 and 3, one CH$_3$CN ligand of 1 is preferentially substituted upon irradiation. The monosubstituted intermediate generated following photolysis in pyridine was isolated and characterized to ascertain which CH$_3$CN ligand was exchanging. Furthermore, the tail of the $^1$MLCT absorption band in 3 extends past 600 nm. Low energy irradiation (λ$_{irr}$ ≥ 610 nm) just inside the therapeutic window results in covalent binding to DNA. The work presented herein provides a greater understanding of photoinduced ligand exchange for the design of future systems with improved properties for PCT.

![Figure 3.1](image.png)

**Figure 3.1.** Schematic representation of the molecular structures of 1 – 3.
3.2 Results and Discussion

3.2.1 Photophysical Properties and Electrochemistry.

The electronic absorption spectra of 1, 2, and 3 are shown in Figure 3. 2. The $^1$MLCT absorption maxima of 1, 2, and 3 in CH$_3$CN are observed at 497 nm ($\varepsilon = 7,800$ M$^{-1}$cm$^{-1}$), 420 nm ($\varepsilon = 10,200$ M$^{-1}$cm$^{-1}$), and 535 nm ($\varepsilon = 7,900$ M$^{-1}$cm$^{-1}$), respectively. As expected, the sequential replacement of the phen ligands in 2 for biq in 1 and 3 results in a stepwise red shift in the lowest energy MLCT absorption maximum.$^{23}$ A similar trend is observed in the polypyridyl complexes [Ru(phen)$_3$]$^{2+}$, [Ru(phen)$_2$(biq)]$^{2+}$, and [Ru(phen)(biq)$_2$]$^{2+}$ with $^1$MLCT maxima at 450 nm, 523 nm, and 551 nm in methanol, respectively.$^{9,23}$ In addition to the Ru$\rightarrow$biq $^1$MLCT absorption in 1, a shoulder at ~410 nm arising from the Ru$\rightarrow$phen $^1$MLCT transition in the complex is apparent, which is at a position similar to that of 2.$^{23}$

![Figure 3.2. Electronic absorption spectra of 1 – 3 in CH$_3$CN.](image-url)
Very weak emission is observed for 1 ($\lambda_{\text{exc}} = 500 \text{ nm}$) and 3 ($\lambda_{\text{exc}} = 535 \text{ nm}$) at room temperature and 2 is not emissive ($\lambda_{\text{exc}} = 420 \text{ nm}$), however, relatively strong luminescence was detected for all three complexes at 77 K (Figures 3.3), as is typical for related systems.\textsuperscript{9,23} The emission maxima in CH$_3$CN at 77 K were measured to be 687, 582, and 696 nm for 1, 2, and 3, respectively. The similarity in the luminescence observed for 1 and 3 indicates a lowest energy Ru $\rightarrow$ biq $^3$MLCT excited state.

![Emission spectra of 1 – 3 in CH$_3$CN at 77 K using excitation wavelengths of 500 nm, 420 nm, and 535 nm, respectively.](image)

**Figure 3.3.** Emission spectra of 1 – 3 in CH$_3$CN at 77 K using excitation wavelengths of 500 nm, 420 nm, and 535 nm, respectively.

Cyclic voltammetry reveals quasi-reversible oxidation events, $E_{1/2}(\text{Ru}^{3+/2+})$, at +1.51 V, +1.45 V, and +1.55 V vs SCE for 1, 2, and 3, respectively, in CH$_3$CN. The similarity among the three potentials points at a metal-centered process, as is typical of Ru(II) polypyridyl complexes.\textsuperscript{9} Quasi-reversible reduction waves in 2 are observed at −1.39 V and −1.55 V vs SCE, at potentials similar to the ligand-centered reduction processes reported previously for the complex.\textsuperscript{24} As expected, these waves are shifted to −0.77 V and −1.04 V vs SCE in 3, since the electrons are localized on the biq ligands with a more
extend π-system as compared to phen. The reduction potentials measured for 3 are comparable to those published for related Ru(II) complexes containing the same ligand, including [Ru(biq)(bpy)_2](PF_6)_2 and [Ru(biq)_2(bpy)](PF_6)_2, with \( E_{1/2}(\text{Ru}^{2+/+}) = -0.97 \) V and \(-0.89 \) V vs SCE, respectively.\(^{25}\) In the asymmetric complex 1, the two reversible reduction waves are observed at \(-0.91 \) V and \(-1.45 \) V vs SCE, assigned to reduction of the biq ligand at the more positive potential, followed by reduction the phen ligand. The absorption \(^1\text{MLCT}\) maxima, emission maxima, and electrochemical data are compiled in Table 3.1.

Table 3.1. Photophysical and electrochemical data for 1 – 3.

<table>
<thead>
<tr>
<th>Complex</th>
<th>( \lambda_{\text{abs}}/\text{nm} ) ((\varepsilon/M^{-1}\ \text{cm}^{-1})^a)</th>
<th>( \lambda_{\text{em}}/\text{nm}^b)</th>
<th>( E_{1/2}/\text{V}^c)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>497 (7.8)</td>
<td>687</td>
<td>+1.51, -0.91, -1.45</td>
</tr>
<tr>
<td>2</td>
<td>420 (10.2)</td>
<td>582</td>
<td>+1.45, -1.39, -1.55</td>
</tr>
<tr>
<td>3</td>
<td>535 (7.9)</td>
<td>696</td>
<td>+1.55, -0.77, -1.04</td>
</tr>
</tbody>
</table>

\(^a\)In CH\(_3\)CN, at 298 K. \(^b\)In CH\(_3\)CN, at 77 K. \(^c\)In CH\(_3\)CN, vs. Ag/AgCl; 0.1 M[n-Bu\(_4\)N][PF\(_6\)].

Ultrafast transient absorption (TA) spectra of 1 – 3 in CH\(_3\)CN are shown in Figure 3.4 (\( \lambda_{\text{ex}} = 310 \) nm, fwhm = 300 fs). For 1 (Figure 3.4a), a typical bleach of the ground state absorption is observed at 500 nm along with a positive transient absorption feature spanning from \(-380\) to 470 nm assigned to reduced biq ligand in the lowest energy excited state, Ru\(\rightarrow\)biq \(^3\text{MLCT}\). Although the signal partially overlaps with the instrument response function, ns-transient absorption (\( \lambda_{\text{ex}} = 355 \) nm, fwhm = 8 ns)
reveals the same positive excited state feature at 380 nm and it can be fitted to a monoexponential decay with $\tau \sim 44$ ns. This state is tentatively assigned to the $^3$MLCT state after vibrational cooling and is much longer than the vibrationally cool Ru→bpy $^3$MLCT state of $[\text{Ru(bpy)}_2(\text{CH}_3\text{CN})_2]^{2+}$, which has a lifetime of 51 ps in CH$_3$CN.$^{21}$

**Figure 3.4.** Ultrafast transient absorption of complexes 1 – 3 in CH$_3$CN ($\lambda_{ex} = 310$ nm, fwhm = 300 fs).
Along with absorption and emission, 2 possesses similar excited state properties to [Ru(bpy)$_2$(CH$_3$CN)$_2$]$^{2+}$ with an overall excited state lifetime of 83 ps which was found by fitting the ground state bleach to a biexponential fit that contained a 3 ps component assigned as vibrational cooling of the $^3$MLCT state. In contrast, a relatively long lived ($\tau$ = 29 ns) positive transient absorption at 400 nm is observed in 3 with a shape similar to the spectrum measured for 2, which further supports our assignment as the reduced biq ligand for this feature. Although 3 undergoes relatively efficient photoinduced ligand exchange, no evidence of a monoaqua product or pentacoordinate intermediate is observed when performing ultrafast TA in H$_2$O as observed in [Ru(bpy)$_2$(CH$_3$CN)]$^{2+}$ under similar conditions.

3.2.2 Photochemistry

The photoreactivity of 1 – 3 was evaluated by monitoring the changes to the electronic absorption and $^1$H NMR spectra as a function of irradiation time. All three complexes possess photolabile CH$_3$CN ligands with photochemistry that can be accessed with $\lambda_{\text{irr}}$ $\geq$ 550 nm for 1, $\lambda_{\text{irr}}$ $\geq$ 455 nm for 2, and $\lambda_{\text{irr}}$ $\geq$ 610 nm for 3. Complexes 1 – 3 are inert to ligand substitution in the dark at room temperature under similar experimental conditions. For the symmetrical complexes 2 and 3, one resonance is observed in CD$_3$CN corresponding to the methyl protons of both bound acetonitrile ligands at 2.22 ppm and 2.28 ppm, respectively (Figure 3.5 and 3.6). During the photolysis of 2 in CD$_3$CN with $\lambda_{\text{irr}}$ $\geq$ 455 nm, the resonance at 2.22 ppm decreases with increasing irradiation time, while a peak at 1.96 ppm corresponding to free CH$_3$CN in CD$_3$CN
increases in intensity at the same rate. The photolysis of 3 in CD$_3$CN with $\lambda_{irr} \geq 610$ nm yields similar results with the decrease of the peak at 2.28 ppm with the concomitant increase of the free CH$_3$CN resonance at 1.96 ppm. The intensity of the peak corresponding to free CH$_3$CN at 1.96 ppm integrates to two ligands at the end of the photolysis for 2 and 3 in CD$_3$CN, indicative that both CH$_3$CN ligands in 2 and 3 are exchanged in each complex (Figures 3.5 and 3.6). It should be noted that because of the electronic equivalence of CH$_3$CN and CD$_3$CN, the $^1$H NMR resonance of bound CH$_3$CN ligands does not shift from those in 2 and 3 to that in the corresponding mono-substituted intermediate. Moreover, no shifts are observed in the aromatic region as the reaction progresses.

![Figure 3.5](image.jpg)

**Figure 3.5.** Photolysis of 2 monitored by $^1$H NMR spectroscopy in CD$_3$CN $\lambda_{irr} \geq 455$. The resonance at 2.13 ppm and 2.09 correspond to residual water and acetone, respectively, and the singlet growing in at 1.96 ppm corresponds to free CH$_3$CN in solution. An integrations of 2 were set relative to an internal standard of benzene.
Figure 3.6. Photolysis of 3 monitored by $^1$H NMR spectroscopy in CD$_3$CN $\lambda_{\text{irr}} \geq$ 610 nm. The resonance at 2.29 ppm corresponds to the bound CH$_3$CN, the resonances at 2.13 ppm and 2.09 ppm correspond to residual water and acetone, respectively, and the singlet growing in at 1.96 ppm corresponds to free CH$_3$CN in solution. An integrations of 2 were set relative to an internal standard of benzene.

The changes to the $^1$H NMR spectrum of 1 upon irradiation in CD$_3$CN using benzene as an internal integration standard are shown in Figure 3.7 ($\lambda_{\text{irr}} \geq$ 455 nm). The CH$_3$CN ligands of 1 are inequivalent, resulting in two resonances of equal integration at 2.53 ppm and 2.12 ppm, labeled CH$_3$CN$^1$ and CH$_3$CN$^2$ in Figure 3.7a, respectively. Figure 3.7b shows that the resonance corresponding to CH$_3$CN$^2$ decreases at a faster rate than that of CH$_3$CN$^1$ upon irradiation in CD$_3$CN; the former disappears within 20 min of photolysis while a significant (~75%) of the latter is still present after 60 min of irradiation. Because the disappearance of the peaks corresponding to bound CH$_3$CN ligands are concomitant with the increase of that associated with free CH$_3$CN at 1.96 ppm, the observed reactivity can be ascribed to the photoinduced ligand exchange with the CD$_3$CN solvent. The photoinduced ligand substitution of the two CH$_3$CN ligands in 2

![NMR Spectra](image)
and 3 is complete in 5 – 60 min. In contrast, the photosubstitution of one CH$_3$CN ligand for CD$_3$CN in 1 is accomplished in ≤ 5 min, but the second CH$_3$CN ligand does not exchange up to 180 min of irradiation.

![Molecular structure of 1 with labeled CH$_3$CN ligands](image)

**Figure 3.7.** (a) Molecular structure of 1 with labeled CH$_3$CN ligands and (b) changes to the $^1$H NMR spectrum of 1 in CD$_3$CN as a function of irradiation time ($\lambda_{\text{irr}} \geq 455$).

The photolysis of 1 in H$_2$O ($\lambda_{\text{irr}} \geq 550$ nm) results in a decrease of the $^1$MLCT band at 494 nm and the appearance of a peak with maximum at 518 nm (Figure 3.8a). The isosbestic points observed at 444 and 506 nm for the process are indicative of the formation of a single species, assigned as the mono-aqua complex, *cis-*
[Ru(bpy)(biq)(CH$_3$CN)(H$_2$O)]$^{2+}$, 4. The red shift of the MLCT maximum from 1 to 4 is similar to that previously reported between [Ru(bpy)$_2$(CH$_3$CN)$_2$]$^{2+}$ and [Ru(bpy)$_2$(CH$_3$CN)(H$_2$O)]$^{2+}$. Continuation irradiation with $\lambda_{\text{irr}} \geq 550$ nm results in negligible spectral changes and only at very long irradiation times, with the growth of a small shoulder at ~560 nm apparent after 3 hr associated with the formation of a small amount of [Ru(biq)(phen)(H$_2$O)$_2$]$^{2+}$ (5). In contrast, when 1 is irradiated in H$_2$O with higher energy light, $\lambda_{\text{irr}} \geq 420$ nm (Figure 3.8b), the formation of 5 is complete within 6 hr. The 2,192 cm$^{-1}$ red-shift of the MLCT band observed from 1 to 5 is of the same magnitude as that reported between [Ru(bpy)$_2$(CH$_3$CN)$_2$]$^{2+}$ and [Ru(bpy)$_2$(H$_2$O)$_2$]$^{2+}$, 3,121 cm$^{-1}$.21

Figure 3.8. (a) Changes to the electronic absorption spectrum of 1 (10 μM) in H$_2$O upon irradiation for 0 – 20 min ($\lambda_{\text{irr}} \geq 550$ nm) and (b) 40 μM for 0 – 4 hours ($\lambda_{\text{irr}} \geq 420$ nm)
It is evident in Figure 3.8 that the irradiation of 1 in H$_2$O for 20 min ($\lambda_{\text{irr}} \geq 550$ nm) results in a well-defined absorption peak with maximum at 518 nm, attributed to the mono-aqua intermediate 4, with negligible spectral changes with continued irradiation with this wavelength. However, when 4 is further irradiated with higher energy light ($\lambda_{\text{irr}} \geq 420$ nm), the bis-aqua species, 5, is formed after 3 hours. The mono-aqua intermediate 4, produced by the photolysis of 1 in H$_2$O ($\lambda_{\text{irr}} \geq 550$ nm, Figure 3.7a is stable in the dark for up to 6 h at room temperature.

![Absorbance vs. Wavelength](image)

Figure 3.9. Irradiation of (a) 50 $\mu$M 2 ($\lambda_{\text{irr}} \geq 420$ nm, 0 – 90 min) and (b) 60 $\mu$M 3 ($\lambda_{\text{irr}} \geq 610$ nm, 0 – 120 min) in H$_2$O.
In contrast to the results described with respect to 1, irradiation of 2 ($\lambda_{\text{irr}} \geq 455$ nm, Figure 3.9a) and 3 ($\lambda_{\text{irr}} \geq 610$ nm, Figure 3.9b) results in complete conversion to the corresponding bis-aqua complex. The intermediate peak corresponding to the mono-aqua species associated with each complex is nearly unidentifiable (Figure 3.9), in agreement with the $^1$H NMR photolysis data in CD$_3$CN. These results lead to the conclusion that in 2 and 3 both CH$_3$CN ligands exchange relatively easily and with similar rates, whereas in 1, one of the CH$_3$CN ligands is significantly more photolabile than the other.

The quantum yields for the first ligand exchange of 1 in H$_2$O to generate 4 with 500 nm and 550 nm irradiation, $\Phi_{500}^{1 \rightarrow 4}$ and $\Phi_{550}^{1 \rightarrow 4}$, were measured to be 0.26(1) and 0.140(5), respectively. These values are similar to those obtained for the formation of the mono-aqua species from 3, $\Phi_{500} = 0.24(1)$ and $\Phi_{550} = 0.150(8)$, as well as from 2, $\Phi_{400} = 0.22(1)$. However, the quantum yield measured for the exchange of the remaining CH$_3$CN ligand from 4 to generate the bis-aqua complex 5 were significantly lower, $\Phi_{400}^{4 \rightarrow 5} = 0.0045(1)$ and $\Phi_{550}^{4 \rightarrow 5} = 0.0014(5)$. These results differ from those for systems, such as [Ru(bpy)(CH$_3$CN)$_4$]$^{2+}$ and [Ru(bpy)$_2$(CH$_3$CN)$_2$]$^{2+}$, for which the quantum yield of the second photoinduced CH$_3$CN ligand exchange is approximately half the value of the first.$^{20,21}$ This comparison points to a selective ligand exchange in 1 that is not present in the symmetric complexes 2 and 3.
3.2.3 X-ray crystal structures

Structural analysis of complexes 1 – 3 can be used to aid in understanding the differences in ligand photodissociation among the complexes. The ORTEP diagrams resulting from X-ray diffraction of the single crystals of 1 and 3 are shown in Figure 3.10, along with the corresponding numbering schemes of the atoms of interest. The crystal structure of 2 was previously reported, and selected structural parameters are compared in Table 3.2. Not only does addition of the biq ligand cause a red shift in absorption, but the steric bulk provided by the ligand results in structural distortions about the metal center. Although 1 only contains one biq ligand, all Ru-N bond distances are similar to that of 3 and the Ru-N bond lengths in 1 and 3 are all approximately 0.02 Å longer than in 2. The elongation of these Ru-N bonds decreases overlap between the d orbitals of the metal and the σ bonding orbitals of the ligand which results in lowering of the energy of the $^3$dd state.

Other parameters that can decrease the overlap between the metal and ligand orbitals are bond angles and tilting of the diimmine ligand. When analyzing 2, the angles $N_2 – Ru – N_6$ and $N_1 – Ru – N_5$ are 93.13° and 94.28°, respectively. In complex 3, the same angles are widened to 98.2° and 98.51°, respectively. However, only the $N_4 – Ru – N_6$ angle is largely distorted in 1, which is measured to be 99.23°. The distortion of only one angle in the asymmetric complex may explain the selectivity observed in the photochemistry for 1 relative to 2 and 3. The biq ligands in complexes 1 and 3 also display tilting, with tilt angles calculated by drawing a plane that intersects with the Ru(II) metal center and the nitrogen atoms of the diimine ligand of interest (Ru-N$_4$-N$_5$ in
1, Ru-N2-N3 and Ru-N4-N5 in 3) and another plane that intersects with the same nitrogen atoms of the diimine ligand of interest and the C21 and C24 (1) and the C28 and C31 (3) atoms labeled in Figure 3.10 that are located furthest out of the original plane drawn relative to the metal center (N4-N5-C21-C24 in 1, N2-N3-C10-C13 and N4-N5-C28-C31 in 3). The angle between the two planes was then calculated. The tilt angle of the biq ligand in 2 was measured to be 22.4° while the tilt angles of the biq ligands in 3 were measured to be 26.9° and 29.1°. No significant tilt was observed for any of the phen ligands.

Figure 3.10. ORTEP plots of 1 and 3 drawn at 50% probability with selected atom numbers; solvent, hydrogen atoms, counteranions, omitted for clarity.
Table 3.2. Selected bond distances, bond angles, and dihedral angles in 1, 2, and 3.

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</table>

This tilting of the sterically demanding ligands was highlighted in our recent work in the complexes [Ru(tpy)(6,6′-dimethylbpy)(py)]²⁺ and [Ru(tpy)(biq)(py)]²⁺ (tpy = 2,2′:6,2′-terpyridine and py = pyridine). Similar tilt angles were observed in these complexes as those in 1 and 3, which resulted in geometric distortions about the metal center. The strain is hypothesized to be the reason for the unprecedented highly efficient photoinduced py ligand dissociation in the tpy complexes. The geometric distortions in
1 and 3 relative to 2 is further confirmed by analyzing specific dihedral angles, which are highlighted in Table 3.2. In 1, the torsional strain measured along the biq ligand (N₄-Ru-N₅-C₂₃ = +22.13°) is much larger than that along the phen ligand (N₂-Ru-N₃-C₁₀ = -0.87°), but is similar to that along both big ligands in 3 (N₂-Ru-N₃-C₁₂ = -28.88° and N₄-Ru-N₅-C₃₀ = +27.07°). No significant torsional strain along the phen ligands in 2 is apparent, as also the case in 1. This tilting and distortion of the biq ligands results in poorer overlap of the d orbitals involved in bonding to the various ligands, which can drastically affect the overall photoreactivity of the complexes, as previously reported for the tpy complexes discussed above.²⁸

3.2.4 Isolation of the Mono-substituted Intermediate of 1

In order to unequivocally assign which CH₃CN ligand of 1 is more photolabile, a photolysis intermediate was isolated and its structure was determined by X-ray crystallography. A mixture of pyridine (py) and CH₃CN (50:50, v:v) was used as the solvent rather than H₂O because py is a stronger coordinating ligand than H₂O, such that a more stable photolysis product was expected. In order to ensure the formation of the mono-substituted intermediate, a 550 nm bandpass filter was used, and the sample was irradiated until no additional changes in the absorption spectrum were apparent following the shift of the MLCT peak from 497 nm to 523 nm, which is accompanied by a change in color from orange to red. After this step in the photolysis was complete, the solution was concentrated and the [Ru(phen)(biq)(CH₃CN)(py)]²⁺ (6) product was precipitated by the addition of ether. Slow evaporation of a solvent mixture of acetone, THF, and
CH$_3$CN and diethyl ether resulted in red crystals suitable for X-ray diffraction, and the resulting ORTEP diagram is shown in Figure 3.11. It is evident from the two different views of the structure of 6 shown in Figure 3.11 that the py exchanged with the CH$_3$CN ligand positioned \textit{trans} to the phen ligand (CH$_3$CN$_2$ in Figure 3.7a) with 80\% occupancy, clearly indicating that this is more photolabile ligand in 1. For the remaining 20\% occupancy, both CH$_3$CN ligands were solved to be replaced by pyridine and the overall structure is a result of co-crystallization.

\textbf{Figure 3.11.} ORTEP plots of two different perspectives of PF$_6^-$ salt of the monosubstituted intermediate 6 (ellipsoids drawn at 50\% probability).

In order to confirm that the crystal structure of 6 accurately depicts the intermediate in solution, the photolysis ($\lambda_{\text{irr}} \geq 550$ nm) of complex 1 was carried out in deuterated pyridine (py-$d_5$) and was followed by $^1$H NMR spectroscopy (Figure 3.12). Upon irradiation up to 20 min, the two resonances corresponding each bound CH$_3$CN ligands at 2.35 and 2.75 ppm decrease in intensity, with the concomitant growth of resonances at 2.95 and 1.84 ppm, and each peak integrates to 3H. The former correspond to the
remaining bound CH$_3$CN ligand in the mono-substituted intermediate, [Ru(biq)(phen)(CH$_3$CN)(py-$d_3$)]$^{2+}$, and the latter to free CH$_3$CN in py-$d_5$. Further irradiation results in a decrease in the 2.95 ppm peak of the intermediate and an increase in free CH$_3$CN resonance, leading to the formation of the bis-substituted product, [Ru(biq)(phen)(py-$d_5$)$_2$]$^{2+}$. The appearance of only one resonance associated with the intermediate indicates that only one of the bound CH$_3$CN ligands exchanges first, followed by the second, making the crystal structure of 6 shown in Figure 3.11 an accurate representation of the mono-substituted species.

**Figure 3.12.** Photolysis of 1 monitored by $^1$H NMR spectroscopy in py-$d_5$ $\lambda_{irr} \geq 550$ nm at 0, 5, 20, and 60 minutes. Integrations relative to a benzene internal standard are listed next to the corresponding peaks.
3.2.5 Calculations

In order gain further understanding on the selective ligand photosubstitution of 1, density functional theory (DFT) calculations were performed. The highest occupied molecular orbitals (HOMOs) of all three complexes, 1 – 3, are calculated to be localized on the d-orbitals of the metal, as is typical of Ru(II) diimine complexes. The LUMO (lowest unoccupied molecular orbital) of 1 exhibits electron density on the biq ligand and the LUMO+1 is localized on phen (Figure 3.13), as expected from the ease of reduction of biq relative to phen. When analyzing the unoccupied orbitals in 2 and 3, the LUMOs are delocalized over both equivalent aromatic ligands of each complex, phen and biq, respectively.

![LUMO and LUMO+1](image)

**Figure 3.13.** Electronic density plots of the calculated LUMO and LUMO+1 of 1 (isovalue = 0.04).

Time-dependent DFT (TD-DFT) calculations reveal that the lowest vertical singlet excited states of 1 and 3 possess significant contribution, ~95%, from HOMO→LUMO transitions, but low oscillator strengths, with maxima at 476 nm (f = 0.0002) and 496 nm.
More intense absorption bands are predicted at 443 nm ($f = 0.0646$) for 1 and at 475 nm ($f = 0.1015$) for 3, calculated to possess 67% and 94% contribution from HOMO-1→LUMO transitions, respectively. The lowest energy vertical singlet excited states of 2 are calculated at 396 nm (81% HOMO→LUMO+1, $f = 0.0024$) and at 395 nm (91% HOMO→LUMO, $f = 0.0093$). It should be noted that the lowest energy electronic transitions predicted are slightly blue shifted relative to the experimental MLCT maxima, as is typical for DFT calculations. 

![Figure 3.14. Electronic density plots of the calculated HOMOs of 1, 2, and 3 (isovalue = 0.04).](image)

In order to understand the selective ligand exchange in 1, the electron density of the orbitals with greatest contribution to the lowest energy excited state need to be considered, the HOMO and the LUMO. In the lowest energy singlet excited state, electron density is removed from the HOMO, which possesses a bonding interaction between the metal d-orbital and the CH$_3$CN ligand positioned trans to the phen ligand.
(Figure 3.14). Therefore, the HOMO is involved in $\pi$-back bonding with CH$_3$CN, and removal of an electron from this MO is expected to weaken the bond. The LUMO of 1 is localized on the $\pi^*$ orbital of the biq ligand; placing electron density in this orbital is expected to strengthen the $\pi$-bond to the CH$_3$CN positioned $trans$ to biq. Moreover, it has been reported that placing additional electron density on the bidentate ligand in the MLCT state of Re(I) carbonyl complexes, such as in [Re(bpy)(CO)$_3$(PR$_3$)$_3$]$^+$ results in photoinduced ligand dissociation of the CO ligand positioned $cis$ to bpy.$^{30}$ Similarly, selective photoinduced ligand exchange of the axial ligands in [Ru(bpy)(CH$_3$CN)$_4$]$^{2+}$ and [Ru(tpy)(CH$_3$CN)$_3$]$^{2+}$ was observed, where the CH$_3$CN ligands $trans$ to the diimine, for which the $\pi$-backbonding is strengthened, do not exchange, but those positioned $cis$ to the bpy or tpy ligand, respectively, are photolabile.$^{12,20}$ Since the LUMOs of 2 and 3 are delocalized equally over both diimine ligands, this selectivity is not observed. Furthermore, a greater amount of energy is required to populate the Ru($t_{2g}$)$\rightarrow$phen($\pi^*$) MLCT singlet excited state in 1 than the Ru($t_{2g}$)$\rightarrow$biq($\pi^*$) MLCT singlet excited state (Table S2). This could be a factor in the enhanced selectivity using lower energy light in which only the Ru($t_{2g}$)$\rightarrow$biq($\pi^*$) state is accessed yielding ligand loss of the CH$_3$CN $trans$ to the phen ligand while higher energy light results in an increased rate for both CH$_3$CN ligands due to the direct population of the Ru($t_{2g}$)$\rightarrow$phen($\pi^*$) state. Moreover, the lowest energy triplet state of 1 resulting from the vertical transition from the minimized singlet ground state was calculated to be $^3$MLCT Ru$\rightarrow$biq in nature, similar to the lowest energy singlet excited state. It should also be noted that the ligand field orbitals play a critical
role in the ligand dissociation process and will be further discussed in 1 and 2 and similar complexes in greater detail in Chapter 4.

Additionally, the photoinduced ligand exchange is believed to occur via a dissociative mechanism, such that the ligand exchange in 1 is expected to proceed through a five-coordinate intermediate to produce the monosubstituted product, 4. Optimization of the five coordinate species, [Ru(biq)(phen)(HCN)]^{2+}, starting from a trigonal bipyramidal geometry results in a distorted square pyramidal geometry with an open site for coordination positioned \( \text{trans} \) to the phen ligand. This result is consistent with the observation of the intermediate 6 (Figure 3.11), where the CH\(_3\)CN ligand \( \text{trans} \) to phen was photosubstituted. In addition, both possible monosubstituted products were optimized, with py \( \text{trans} \) to phen and \( \text{trans} \) to biq. When the py replaced the CH\(_3\)CN \( \text{trans} \) to the phen ligand, the overall energy is more stable by 11 kJ/mol relative that \( \text{trans} \) to the biq, such that the former is thermodynamically favored.

### 3.2.6 DNA interactions of [Ru(biq)\(_2\)(CH\(_3\)CN)\(_2\)]^{2+}

Since complex 3 undergoes relatively efficient photoinduced ligand exchange in water with irradiation wavelength just inside the PCT window, preliminary DNA binding studies were conducted using gel electrophoresis mobility shift assays and viscosity measurements. It is well documented that cisplatin thermally binds to linearized DNA and reduces its migration through an agarose gel in a concentrated-dependent manner.\(^{31}\) A similar pattern is observed for 3 upon irradiation with low energy light, but to a lesser extent in the dark (Figure 3.15). In Figure 3.15, lanes 1 and 8 contain 1 kb DNA ladder,
lanes 2-7 were loaded with 50 μM pUC18 DNA, and lanes 3 – 6 contain increasing concentrations of 3. In Figure 2.19a, the samples in lanes 3 – 6 were irradiated for 20 minutes with λ_{irr} ≥ 610 nm light prior to loading. It is evident in Figure 3.15a that, as the concentration of 3 is increased, the DNA mobility decreases, whereas a much smaller shift that does not increase with increasing concentration is observed when the samples are incubated in the dark for 30 minutes under similar experimental conditions (Figure 3.15b). These results are indicative of covalent binding of 3 to DNA only upon irradiation.

Figure 3.15. Imaged ethidium bromide-stained agarose gels of 50 μM linearized pUC18 plasmid (10 mM phosphate buffer, pH = 8.78) in the presence of various concentrations of complex: lanes 1 and 8, 1 kb DNA molecular weight standard; lanes 2 and 7, linearized plasmid alone; lanes 3-6, 25, 50, 100, 200 μM of 3 (a) irradiated (λ_{irr} ≥ 610 nm, 20 min) and (b) incubated in the dark (30 min, 298 K).

The gel electrophoresis mobility shift assays of 3 and [Ru(bpy)_2(CH_3CN)_2]^{2+} (7) were then compared under analogous conditions (Figure 3.16). For both gels, lanes 1 and 8 contain 1 kb DNA ladder, lanes 2-7 were loaded with 50 μM pUC18 DNA, and lanes 3 – 6 contain increasing concentrations of 3. The samples in lanes 3 – 6 in both gels were
irradiated for five minutes with $\lambda_{irr} \geq 395$ nm light prior to loading. The same trend as seen in Figure 3.15a is observed for the two complexes in Figure 3.16, however, to more of an extent in complex 7. Both complexes undergo efficient ligand dissociation, but due to the bulky biq ligands, complex 3 cannot covalently bind to DNA as well and thus does not decrease the DNA migration at the same concentration ratios of Ru:DNA as 7.

![Figure 3.16](image_url)

**Figure 3.16.** Imaged ethidium bromide-stained agarose gels of 50 μM linearized pUC18 plasmid (10 mM phosphate buffer, pH = 87.8) in the presence of various concentrations of complex: lanes 1 and 8, 1 kb DNA molecular weight standard; lanes 2 and 7, linearized plasmid alone; lanes 3-6, 1, 2, 5, 10 μM of (a) 3 and (b) 7 irradiated with $\lambda_{irr} \geq 395$ nm for 5 min at 298 K.

In order to further clarify the binding interaction of 3 with DNA, relative viscosity measurements were conducted. Hydrodynamic measurements that sensitize the length of the DNA chain are regarded as the most unambiguous tests to provide a binding model in solution in the absence of a crystal structure. A covalent binding model involves kinking of the DNA helix, which essentially decreases the length of the overall DNA strand and causes the viscosity to decrease relative to DNA alone. In contrast, an intercalation interaction effectively lengthens the distance between base pairs increasing the overall length of the DNA strand and causes the viscosity to increase. The relative
viscosities of 3 (irradiated and in dark) are shown in Figure 3.17. As concentration of complex is increased when irradiated with $\geq 610$ nm, the relative viscosity decreases indicative of a covalent binding interaction. The same trend was observed for cisplatin but to a much greater extent. This again is most likely due to the bulky biq ligands that can interfere with covalently binding of adjacent guanine residues as is seen in the therapeutic mechanism of cisplatin. Also, no significant decrease of the relative DNA viscosity of 3 is observed when incubated in the dark with DNA for 20 minutes. This further suggests that 3 interacts with DNA by covalent binding only upon irradiation with low energy light and could function as an ideal candidate for PDT.

![Figure 3.17](image)

**Figure 3.17.** Relative viscosity changes of 200 mM ct-DNA in the presence of complex 3 irradiated and incubated in the dark in buffer (1.5 mM Na$_2$HPO$_4$, 0.5 mM NaH$_2$PO$_4$, pH = 7.0)
3.3 Conclusions

The series of Ru(II) complexes 1 – 3 possess two CH$_3$CN ligands in a cis-disposition and undergo photodinduced ligand exchange with solvent or coordinating molecules in solution when irradiated with visible light. Selective CH$_3$CN ligand exchange takes place in the asymmetric complex 1 with low energy irradiation ($\lambda_{\text{irr}} \geq 550$ nm), where only one of the ligands is photolabile. This selectivity is not observed in the symmetric complexes 2 and 3. A crystal structure of the PF$_6^-$ salt of the mono-substituted intermediate [Ru(biq)(phen)(CH$_3$CN)(py)]$^{2+}$ (6), was obtained as the product of the photolysis of 1 in a py:CH$_3$CN solvent mixture, showing the selective exchange of the CH$_3$CN ligand trans to phen. DFT calculations show that the lowest energy $^1$MLCT and $^3$MLCT states of 1 are characterized by a decrease of electron density in a Ru-CH$_3$CN $\pi$-bonding orbital, thus weakening the bond to the CH$_3$CN ligand trans to phen. The promoted electron is localized on the LUMO with biq($\pi^*$) character, strengthening the Ru-CH$_3$CN bond of the ligand positioned trans to biq. These results point at the direct role of the MLCT states in the photoinduced ligand exchange process. Our efforts could potentially serve as a route for the synthesis of heteroleptic inorganic complexes, as well as a new method for wavelength selective drug delivery in photochemotherapeutic applications. Furthermore, complex 3 was shown to covalently bind DNA when irradiated with $\lambda_{\text{irr}} \geq 610$ nm but remained inert in the dark. This irradiation wavelength just inside the therapeutic window suggesting that incorporation of biq ligands is useful in the design of future PCT complexes.
References


Chapter 4: Photoinduced Ligand Exchange in Tris-heteroleptic Ru(II) Complexes Possessing Various Dimethyl-1,10-Phenanthroline or Biquinoline Ligands

4.1 Introduction

The photochemistry of Ru(II) polypyridyl complexes has been investigated extensively due to their success in dyes sensitized solar cells (DSSCs) and use as luminescent biological probes.\textsuperscript{1-7} Some Ru(II) complexes possess low lying $^3$LF (ligand field, or $^3$dd) states that can become populated upon excitation resulting in ligand dissociation.\textsuperscript{8-11} Population of these states from excited triplet metal-to-ligand charge transfer ($^3$MLCT) states places an electron in an $e_g$ orbital with Ru – L ($\sigma^*$) character, thus weakening the bond and resulting in ligand dissociation.\textsuperscript{8-11} Moreover, the energy gap between the $^3$MLCT state and $^3$dd state can have a profound impact on the photoreactivity of the complex.\textsuperscript{12} Control of ligand dissociation reactions in Ru(II) complexes could be useful for light activated drug release involved in photochemotherapy (PCT).\textsuperscript{13-15}

Chapter 3 focused on extensive photochemical studies on the complex, $[\text{Ru}(2,2´-\text{biq})(\text{phen})(\text{CH}_3\text{CN})_2]^{2+}$ (1) (2,2´-biq = 2,2´-biquinoline, phen = 1,10-phenanthroline) in comparison to the complex $[\text{Ru}(\text{phen})_2(\text{CH}_3\text{CN})_2]^{2+}$ (2).\textsuperscript{16} Both complexes undergo dissociation of the CH$_3$CN ligands when irradiated with visible light in coordinating
solvents. However, 1 displayed selective photosubstitution of the CH₃CN trans to the phen ligand followed by the exchange of the second CH₃CN at a much slower rate. Although the analogous photochemical reaction of 2 must proceed through the mono-aqua intermediate [Ru(phen)₂(CH₃CN)(H₂O)]²⁺, experimental evidence presented in Chapter 3 suggests no selectivity in the exchange of the equivalent CH₃CN ligands with water.

Structurally, the presence of the bulky 2,2’-biq ligand results in geometric distortions about the metal center in 1. These distortions have been used to explain photochemical reactions where the 2,2’-biq ligand is exchanged photochemically for synthetic and chemotherapeutic applications, which will be described in greater detail in Chapter 5.¹⁷,¹⁸ No significant structural distortions were observed in 2 and it undergoes similar photochemistry, as reported for [Ru(bpy)₂(CH₃CN)]²⁺.¹⁹ Aside from the differences in geometry and asymmetry, complex 1 possesses a lowest energy MLCT state involving movement of an electron from a highest occupied molecular orbital (HOMO) on the Ru(II) metal to a lowest unoccupied molecular orbital (LUMO) localized on the biq ligand, while the lowest energy MLCT in 2 involves a HOMO on the metal and LUMO that is localized on one of the equivalent phen ligands. Since irradiation corresponding to this lowest energy transition was shown to cause photoinduced ligand exchange in 1 and 2, it was proposed that localization of the electron density on one bidentate ligand results in the initial labilization of the CH₃CN trans to the phen ligand, followed by photolabilization of the second bound CH₃CN molecule. This selective
photosubstitution reaction could be useful as a synthetic tool or for controlled release of multiple drugs.

However, recent research on Ru(II) complexes with bulky ligands that impart steric strain around the coordination sphere of the metal such as biq or derivatives of 6,6'-dimethyl-2,2'-bipyridine, has led to the conclusion that coordination of these ligands lowers the energy of the $^3$dd states decreasing its energy gap with the $^3$MLCT state. It has even been proposed that the energy of the $^3$dd state drops below that of the $^3$MLCT state in some cases, which is expected to increase the efficiency of photoinduced ligand exchange. In order to further investigate the effect of sterics and localized charge transfer transitions on selective ligand dissociation in Ru(II) complexes, a platform needs to be created with a series of complexes that possess these characteristics. The incorporation of methylated phen ligands such as 4,7-dimethyl-1,10-phenanthroline (4,7-dmphen), 5,6-dimethyl-1,10-phenanthroline (5,6-dmphen), and 2,9-dimethyl-1,10-phenanthroline (2,9-dmphen) and various biquinoline ligands, including 3,3'-isobiquinoline (3,3'-ibiq), 1,1'-isobiquinoline (1,1'-ibiq), and 1,12-diazaperylene (dap) in the coordination sphere was used to gain further insight into these effects. Herein, we compare the photophysical and structural properties of $[\text{Ru}(4,7$-dmphen)(phen)(CH$_3$CN)$_2$]$^{2+}$ (3), $[\text{Ru}(5,6$-dmphen)(phen)(CH$_3$CN)$_2$]$^{2+}$ (4), $[\text{Ru}(2,9$-dmphen)(phen)(CH$_3$CN)$_2$]$^{2+}$ (5), $[\text{Ru}(3,3$'-ibiq)(phen)(CH$_3$CN)$_2$]$^{2+}$ (6), $[\text{Ru}(1,1$'-ibiq)(phen)(CH$_3$CN)$_2$]$^{2+}$ (7), and $[\text{Ru}(\text{dap})(phen)(CH$_3$CN)$_2$]$^{2+}$ (8), as well as with those of 1 and 2. A structural representation of the tris-heteroleptic complexes are shown in Figure 4.1.
4.2 Results and Discussion

4.2.1 Synthesis.

The synthesis for the control complexes 1 and 2 was described in Chapter 2. Ru(phen)(DMSO)\(_2\)Cl\(_2\) proves to be a viable precursor in the preparation of the tris heteroleptic complexes 3, 4, 6, 7, and 8 via a building-block approach. In order to avoid contact between opposing hydrogen atoms on the 4 and 4′ positions on the fused benzene rings that make up the 1,1′-ibiq ligand, the ligand must orient itself in such a way to minimize these steric repulsions. It is evident in the \(^1\)H NMR spectrum that two isomers of complex 7 are present after synthesis, which are shown in Figure 4.2. Unfortunately no purification technique proved successful to separate them and the photophysical studies were conducted with the mixture of isomers. It should be noted that this synthetic route was unsuccessful in the preparation of 5 using 2,9-dimethyl-1,10-phenanthroline (2,9-
Refluxing Ru(phen)(DMSO)$_2$Cl$_2$ with 2,9-dmphen in DMF for 4 hours results in a brown solution. Purification of the crude product using the immiscible H$_2$O:CH$_2$Cl$_2$ system results in two layers of similar orange color indicating starting material in the organic CH$_2$Cl$_2$ layer and [Ru(phen)(2,9-dmphen)$_2$]$^{2+}$ in the aqueous layer. The typical purple color observed for Ru(phen)$_2$Cl$_2$ complexes in solution is not observed. The analogous experiment with longer reflux times in DMF (15 hours) leads to oxidation of the Ru(II) complex evidenced by the resulting brown color of the organic layer upon purification and broad absorption band that spans the visible region. Synthesis of Ru(2,9-dmphen)(DMSO)$_2$Cl$_2$ under the experimental conditions used to generate the corresponding unsubstituted phen complex was unable to be achieved, further confirming the synthetic difficulties of the incorporation of sterically demanding ligands in the Ru(II) coordination sphere.

An alternative route that proved successful in the synthesis of 5 was through extensive irradiation of [Ru(2,9dmphen)$_2$(phen)]$^{2+}$ with $\lambda_{irr} \geq 345$ nm light in CH$_3$CN. It
is well established that Ru(II) complexes with ligands that impart steric hinderance, specifically 2,9-dimethyl-1,10-phenanthroline and 6,6′-dimethyl-2,2′-bipyridine, are able to undergo photoinduced dissociation of one of the bulky bidentate ligand in coordinating solvents.\textsuperscript{17,20} Irradiation of \([\text{Ru}(\text{2,9dmphen})_{2}(\text{phen})]^{2+}\) in CH\(_3\)CN affords 5 as a pure product and no oxidation of the complex is observed. This photochemical route was also attempted for 3 and 4 using \([\text{Ru}(2,2′-\text{biq})(\text{phen})(\text{L})]^{2+}\) (L = methylated phen ligand). Extensive photolysis in CH\(_3\)CN does result in the exchange of the bulky biq ligand, but the reaction does not reach completion.

### 4.2.2 X-ray Crystal Structures

The crystal structures of 3 and 5 are depicted in Figure 4.3 with relevant crystallographic data compiled in Table 4.1. Single crystals of 4 were grown but were too small for successful data collection on our instrument. No significant differences in the six Ru-N bond lengths are found in 3 and these values are similar to those reported in \([\text{Ru}(\text{phen})_{2}(\text{CH}_{3}\text{CN})_{2}][\text{PF}_6]_2\), (2), previously discussed in Chapter 3.\textsuperscript{23} However, in 5, the Ru-N\(_3\) and Ru-N\(_4\) bond distances are 2.091 Å and 2.107 Å, respectively, and are slightly longer than the other Ru-N bonds in the complex and those found in 2 and 3.\textsuperscript{23} This difference can be explained by the steric strain caused by the methyl substituents directed towards the metal center. These bond distances are also greater than those observed in 1, possessing the 2,2′-biq ligand, which has Ru-N\(_3\) and Ru-N\(_4\) bond lengths of 2.076 Å and 2.087 Å, respectively (Chapter 3).\textsuperscript{16}
Figure 4.3. ORTEP plots of (a) 3 and (b) 5 drawn at 50% probability with selected atom numbers; solvent, hydrogen atoms, counteranions, omitted for clarity.
Table 4.1. Selected bond distances, bond angles, and dihedral angles in 3, 5, 6, and 8.

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<th>Bond distance (A)</th>
<th>3</th>
<th>5</th>
<th>6</th>
<th>8</th>
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<table>
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<th>5</th>
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<th>8</th>
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<td>+9.10</td>
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<td>-1.24</td>
<td>-0.34</td>
<td>+2.07</td>
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<tr>
<td>N₃-Ru-N₄-C</td>
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<td>-20.25 (C₁₆)</td>
<td>-3.55 (C₁₉)</td>
<td>+5.99 (C₁₆)</td>
</tr>
<tr>
<td>N₃-C-C-N₄</td>
<td>-0.632</td>
<td>+0.695</td>
<td>-0.57</td>
<td>+2.33</td>
</tr>
</tbody>
</table>

The presence of the methyl groups in 5 also causes a significant distortion of the N₄-Ru-N₆ angle measured to be 98.39°, which is 94.73° in 3, for which the methyl groups cause no steric strain and is similar to all angles around the central Ru atom reported in 2.²³ Moreover, the N₄-Ru-N₆ angle is also distorted in 1 and is measured to be 99.23° (Chapter 3). Another significant difference in the crystal structures of 3 and 5 is the tilt
of the 2,9-dmphen ligand in the latter, which is similar to that of complexes possessing
the 2,2′-biq ligand in Chapter 3. For the two crystal structures shown in Figure 4.2, a
plane was drawn through the Ru(II) metal center and the coordinating nitrogen atoms
(Ru-N3-N4) of the methyl-substituted phen ligand and another plane was then generated
such that it intersects with the coordinated nitrogen atoms as well as the carbon atoms at
the 5 and 6 positions in the methyl-substituted phen ligand (N3-N4-C18-C19). The angle
measured between these planes represents the tilt of the methyl-phen ligands in 3 and 5
from a planar bidentate ligand in an octahedral geometry. The tilt angles for 3 and 5 were
measured to be ~9° and ~28°, respectively, and the large deviation in the latter is due to
the steric hindrance provided by the methyl groups pointing towards the metal center in
5. The tilt also directs the methyl groups away from the other coordinated phen ligand in
5, which is not necessary in 3, and resulting in a smaller tilt angle. A similar trend is
observed in the crystal structure of 2, which has a tilt angle of ~22° imparted by the bulky
biq ligand.16 The tilt of the sterically demanding ligand is further highlighted by
comparing the torsional strain in complexes 3 and 5. The dihedral angles of the
methylated phen ligands (N3-Ru-N4-C16) were measured to be +4.53 and -20.25 in 3 and
5, respectively. Similar distortion angles, bond distances and tilts were reported in
complexes possessing the 2,2′-biq ligand or ligands with methyl substituents adjacent to
the coordinating nitrogen atoms.18,24,25 Clearly, steric demands result in distorted
structures being energetically favorable conformations in Ru(II) complexes and may play
a key role in the overlap of the d orbitals involved in bonding to the various ligands. The
reduced overlap caused by distortions can drastically affect the overall photophysical properties and photoreactivity of the complex.\textsuperscript{16,21,25}

The molecular structures of 6 and 8 are depicted in Figure 4.4 with relevant crystallographic data also compiled in Table 4.1. We were unable to grow single crystals of complex 7 most likely due to the mixture of isomers present after synthesis. The average Ru–N bond lengths in 6 and 8 are 2.049 Å and 2.053 Å, respectively, and are similar to 2, but are slightly shorter than those reported for [Ru(bpy)\textsubscript{3}]\textsuperscript{2+}, which are 2.044 Å and 2.057 Å, respectively.\textsuperscript{26} Furthermore, there are no bond angles that are significantly larger than those observed in typical Ru(II) complexes in 6 and 8 that were observed in complexes 1 and 5. In addition, the tilt angles measured in a manner analogous to that described for 3 and 5 were determined to be \(~3^\circ\) and \(~5^\circ\) for the 3,3′-ibiq and dap ligands in 6 and 8, respectively. Due to the lack of steric hindrance presented by the 3,3′-ibiq and dap ligands, no significant torsional strain is observed for 6 and 8, with dihedral angles measured along the 3,3′-ibiq ligand (N\textsubscript{3}-Ru-N\textsubscript{4}-C\textsubscript{19}) and along the dap ligand (N\textsubscript{3}-Ru-N\textsubscript{4}-C\textsubscript{16}) to be \(-3.55^\circ\) and \(+5.99^\circ\), respectively. It is clear that the addition of fused benzene rings pointing away from the metal center in 6 and 8 do not result in distortions about the Ru(II) metal center.
4.2.3 Electronic Absorption and Emission.

The electronic absorption spectra of 3 – 5 exhibit $^1$MLCT maxima at 420 nm (11100 M$^{-1}$ cm$^{-1}$), 423 nm (8700 M$^{-1}$ cm$^{-1}$), and 430 nm (10200 M$^{-1}$ cm$^{-1}$) in CH$_3$CN respectively, and are similar to that of 2 (Figure 4.5).$^{16}$ The slight red shift in absorption in 5 relative to 3 and 4 has been reported in related complexes possessing the 2,9-dmphen ligand.$^{27}$ Since the methyl substituents act as weakly electron donating groups to the aromatic phen ligand, the lowest energy $^1$MLCT transition can be assigned as Ru(t$_{2g}$) $\rightarrow$ phen-(π*) that overlaps with the slightly higher in energy Ru(t$_{2g}$) $\rightarrow$ dimethylphen (π*) $^1$MLCT transition.

No emission is observed for 2 – 5 at room temperature, a result that agrees with previously reported Ru(II) complexes possessing photolabile CH$_3$CN ligands, as well as Ru(II) complexes possessing bidentate ligands with methyl substituents adjacent to the coordinating nitrogen atoms, such as in [Ru(6-methyl-2,2’-bipyridine)$_3$]$^{2+}$ and [Ru(4,4’-
6,6'-tetramethyl-2,2'-bipyridine)\textsubscript{3}\textsuperscript{2+}.\textsuperscript{21} The lack of emission can be explained by the efficient population of low lying, non-luminescent \textsuperscript{3}dd states that results in ligand dissociation and thermal deactivation to the ground state. However, emission is observed for 2 – 5 at 77 K in CH\textsubscript{3}CN with maxima at \textasciitilde540 nm, \textasciitilde580 nm, and \textasciitilde630 nm corresponding to the vibrational structure of the ground state (Figure 4.6). Similar spectral features were reported for 2 in CH\textsubscript{3}CN,\textsuperscript{16} such that the absorption and emission data of 2 – 5 indicate similar \textsuperscript{1}MLCT and \textsuperscript{3}MLCT state manifolds for the complexes.

**Figure 4.5.** Electronic absorption spectra of complexes 2 – 5 in CH\textsubscript{3}CN.

**Figure 4.6.** Emission spectra of complexes 2 – 5 in CH\textsubscript{3}CN at 77 K.
In contrast, the biquinoline complexes, 1, 6 – 8, possess significantly different lowest energy \(^1\)MLCT transitions and the corresponding electronic absorption spectra measured in CH\(_3\)CN are shown in Figure 4.7. As stated in Chapter 3, complex 1 exhibits a peak at 497 nm (\(\varepsilon = 7,800\) M\(^{-1}\)cm\(^{-1}\)) assigned as a Ru\(\rightarrow\)biq \(^1\)MLCT transition. Addition of the 2,2\(^{\prime}\)-biq ligand significantly red-shifts the lowest energy \(^1\)MLCT absorption relative to 2 because the lowest unoccupied molecular orbital (LUMO) localized on the biq ligand is at significantly lower energy than that of phen, confirmed by confirmed electrochemistry, emission, and calculations in Chapter 3.\(^{1,6,28}\) A transition at similar energy is observed for 7 that contains the 1,1\(^{\prime}\)-ibiq ligand with a peak at 485 nm (\(\varepsilon = 7,500\) M\(^{-1}\)cm\(^{-1}\)) arising from Ru\(\rightarrow\)ibiq \(^1\)MLCT transitions. Incorporation of the dap ligand in 8 results in a red shift of the lowest energy absorption to 522 nm (\(\varepsilon = 11,500\) M\(^{-1}\)cm\(^{-1}\)), assigned as Ru\(\rightarrow\)dap \(^1\)MLCT. This red shift in absorption is typical when replacing the 2,2\(^{\prime}\)-biq ligand with dap in Ru(II) polypryidyl complexes and is observed in \([\text{Ru(biq)}_2(\text{bpy})]^2^+\) to \([\text{Ru(dap)}_2(\text{bpy})]^2^+\), with \(^1\)MLCT maxima at 549 nm and 576 nm in CH\(_3\)CN, respectively.\(^{29}\) Complex 8 also possesses peaks at 461 nm and 435 nm that are observed in \([\text{Ru(dap)}_2(\text{bpy})]^2^+\) at 468 nm and 441 nm and are assigned as \(\pi\rightarrow\pi^*\) transitions of the dap ligand.\(^{29}\) These low energy \(\pi\rightarrow\pi^*\) transitions are observed in other Ru(II) complexes containing ligands with extended \(\pi\) systems such as \([\text{Ru(bpy)}_2(\text{dppn})]^2^+\) (dppn = benzo[\(i\)]dipyrido-[3,2-\(a\);2',3'\(-c\)]phenazine).\(^{30}\) Although the spectral features of 6 are expected to be similar to those of 1 and 7, it displays a blue shift in absorption relative to them and 2 – 5, with a peak at 390 nm (\(\varepsilon = 7,800\) M\(^{-1}\)cm\(^{-1}\)) indicating that the lowest energy transition in the complex is Ru\(\rightarrow\)phen \(^1\)MLCT similar to that of 2. This finding is
consistent with complexes containing the 3,3′-ibiq ligand, where a stepwise blue shift of the $^1$MLCT maximum from 452 nm to 392 nm is observed in $\text{[Ru(bpy)}_3]\text{]}^{2+}$, $\text{[Ru(bpy)}_2(3,3′\text{-ibiq})]\text{]}^{2+}$, $\text{[Ru(bpy)} (3,3′\text{-ibiq)})_2]\text{]}^{2+}$, and $\text{[Ru(3,3′\text{-ibiq})}_3]\text{]}^{2+}$.31

Figure 4.7. Electronic absorption spectra of complexes 1 and 6 – 8 in CH$_3$CN.

Figure 4.8. Emission spectra of complexes 1 and 6 – 8 in CH$_3$CN at 77 K.
Similar to 1, very weak emission is observed for 7 and no luminescence is observed for 6 or 8 at room temperature. The emission spectra of 1, 6, and 7 at 77 K in CH$_3$CN are shown in Figure 4.8 and exhibit maxima at 687 nm, 596 nm, and 700 nm, respectively. No emission is observed for 8 at 77 K, consistent with a ligand-centered dap $^3\pi\pi^*$ excited state, similar to that of [Ru(bpy)$_2$(dppn)]$^{2+}$. Due to the nearly identical emission maxima measured for 1 and 7, the lowest energy excited state in 7 is assigned as arising from the lowest energy Ru→1,1′-ibiq $^3$MLCT excited state, which was reported to be Ru→2,2′-ibiq for 1.$^{16}$ The complex [Ru(3,3′-ibiq)$_3$]$^{2+}$ has an emission peak at 540 nm under analogous experimental conditions, which was determined to be ligand-centered phosphorescence.$^{32}$ The emission of 6 is similar to that of 2 indicating a lowest energy Ru→phen $^3$MLCT excited state similar to other mixed ligand systems possessing bpy and 3,3′-ibiq and further confirms the electronic absorption assignment. It is evident that simply moving the position of the fused ring of the biquinoline ligand can significantly impact the excited state properties of the complex, which would in turn affect photochemical reactivity.

4.2.4 Photochemistry.

As we have previously reported, 2 exchanges the monodentate CH$_3$CN ligands with solvent H$_2$O molecules when irradiated with ≥ 455 nm light to form the diaqua complex [Ru(phen)$_2$(H$_2$O)$_2$]$^{2+}$. The changes to the electronic absorption as a function of irradiation time for 3 are shown in Figure 4.9a. Photolysis of 3 in H$_2$O results in a decrease of the $^1$MLCT band at 420 nm and the growth of a broad absorption band with a
peak at 465 nm corresponding to the diaqua product ($\lambda_{\text{irr}} \geq 455 \text{ nm}$). This red shift in absorption of 2304 cm$^{-1}$ in 1 from the bis-CH$_3$CN the diaqua species is of the same order of magnitude to those reported for the corresponding ligand substitution in $[\text{Ru(phen)}_2(\text{CH}_3\text{CN})_2]^{2+}$ (2757 cm$^{-1}$) and $[\text{Ru(bpy)}_2(\text{CH}_3\text{CN})_2]^{2+}$ (3124 cm$^{-1}$).$^{16,19}$ Photolysis experiments with 4 results in similar spectral changes, suggesting analogous photoreactivity. No changes are observed to the electronic absorption for 3 and 4 in the dark (Figure 4.9b).

A similar trend is observed upon irradiation of 5 in H$_2$O ($\lambda_{\text{irr}} \geq 455 \text{ nm}$) in which an initial decrease of the $^1\text{MLCT}$ absorption band is observed (Figure 4.9c). However, over the same time span as in 3 and 4, the final product $[\text{Ru(2,9dmphen)(phen)(H}_2\text{O)}_2]^{2+}$ is not formed. Unlike 2 – 4, a distinguishable peak that appears at 465 nm in 3 and 4 and at 475 nm in 2 is not observed for 5.$^{16}$ It should be noted that 3 is slightly unstable in the dark, but the changes in absorption in the dark over an hour are significantly lower than under irradiation. In order to further assess the differences in photoinduced ligand exchange in the complexes, photolysis reactions were monitored by $^1\text{H}$ NMR spectroscopy.

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Figure 4.9. Changes to the electronic absorption spectrum in H$_2$O upon irradiation for 0 – 90 min of (a) 3 and (b) 4 and (c) for 0 – 210 min for 5 ($\lambda_{irr} \geq 455$ nm).
The changes to the $^1$H NMR spectra of 2 – 5 under photolysis in CD$_3$CN were performed in CD$_3$CN are shown in Figure 4.10. In the initial $^1$H NMR spectrum of the symmetric control complex 2 (Figure 4.10a), the resonance at 2.22 ppm corresponds to the methyl protons of both bound acetonitrile ligands. Upon irradiation of 2 in CD$_3$CN with $\lambda_{irr} \geq 455$ nm, the peak at 2.22 ppm decreases with increasing irradiation time, while a new resonance at 1.96 ppm known to correspond to free CH$_3$CN in CD$_3$CN increases in intensity at the same rate, as previously discussed in Chapter 3 and shown in Figure 3.5. After an hour of irradiation all bound CH$_3$CN ligands have exchanged with solvent CD$_3$CN, as evidenced by the equal integration of free CH$_3$CN and initial bound CH$_3$CN relative to the internal benzene standard. An analogous experiment was performed with 3. For which the resonances at 2.23 and 2.19 ppm correspond to the methyl protons on the bound CH$_3$CN ligands, which are inequivalent in this complex (Figure 4.10b). Both resonances decrease with a similar rate with increasing irradiation time, with a concomitant increase in intensity of the free CH$_3$CN peak resonance at 1.96 ppm. After an hour of irradiation, the resulting spectrum reveals all bound CH$_3$CN ligands exchange for CD$_3$CN in a manner similar to 2, and no selectivity in the exchange of these inequivalent bound CH$_3$CN ligands is observed. Complex 4 follows the same trend when irradiated in CD$_3$CN under similar experimental conditions.

In contrast, the photolysis of 5 reveals selective photoinduced ligand exchange of one of the bound CH$_3$CN ligands. In Figure 4.10c, the resonances at 2.09 and 1.98 ppm correspond to the methyl protons on the inequivalent bound CH$_3$CN ligands. Upon irradiation, both resonances decrease with the concomitant growth of free CH$_3$CN at 1.96
However, the resonance at 1.98 ppm decreases at a significantly faster rate than that at 2.09 ppm. After an hour of irradiation in CD$_3$CN, the peak originally at 1.98 ppm is no longer distinguishable from the baseline, while the resonance at 2.09 ppm remains present with 77% of the original intensity (using an internal benzene standard), indicating that a significant amount of bound CH$_3$CN is still present in solution. Extended photolysis results in a decrease of the resonance at 2.09 ppm but at a very slow rate. This result is consistent with the hypothesis that one of the bound CH$_3$CN ligands is significantly more photolabile than the other, such that when the monosubstituted intermediate [Ru(2,9-dmphen)(phen)(CH$_3$CN)(CD$_3$CN)]$^{2+}$ is irradiated, the CD$_3$CN (coordinated at the reactive position), exchanges selectively with another CD$_3$CN solvent molecule.

**Figure 4.10.** Changes to the $^1$H NMR spectrum (a) 2 (b) 3 and (c) 5 in CD$_3$CN after 60 minutes of irradiation ($\lambda_{\text{irr}} \geq 455$).
As stated previously in Chapter 3, this trend is also observed in the asymmetric complex, 1, that possesses the 2,2′-biq ligand, where the CH$_3$CN ligand bound \textit{trans} to the phen ligand is selectively substituted for a solvent molecule upon irradiation, followed by the exchange of the second CH$_3$CN ligand at a significantly slower rate.$^{16}$ The crystal structure of 1 was discussed, which shows significant tilting of the biq ligand that results in distortions about the metal center. This tilt directs the extended aromatic rings of the biq ligands toward the more photolabile CH$_3$CN in the complex. This bending of the 2,2′-biq ligand may result in poorer overlap of Ru($\pi$) t$_{2g}$-type orbitals and the empty $\pi^*$ orbitals of the CH$_3$CN ligand involved in $\pi$-backbonding, thus weakening this bond, which may account for its selective photosubstitution and thermal instability. The electronic properties of 3 – 5 are very similar, but the photoinduced ligand exchange properties are different for 5. Complex 5 displays similar photoreactivity as 1, such that the common features in the structures of both complexes may play a key role in the photochemistry. Furthermore, 5 displays a similar tilt of the 2,9-dmphen ligand as that observed in the 2,2′-biq ligand in 1, a feature that is not observed in the crystal structures of 2 or 3. The crystal structure of 5 reveals that methyl groups in the are directed towards the CH$_3$CN \textit{trans} to the unsubstituted phen ligand, which is also the more photolabile CH$_3$CN ligand, similar to complex 1. The proposed photochemical reaction is outlined in Figure 4.11. These results indicate that the structural distortion has a significant impact on the photoinduced ligand dissociation process in Ru(II) polypyridyl complexes and explains the specific steric features that give rise to selective ligand dissociation.
Figure 4.11. Proposed reaction upon irradiation in H₂O of 5.

Photolysis of 6 in CD₃CN monitored by ¹H NMR spectroscopy is shown in Figure 4.12 and results are similar to those observed for 3 and 4. The resonances at 2.10 ppm and 2.85 ppm correspond to the methyl protons on the two chemically inequivalent CH₃CN ligands and have equal integrations before irradiation, tᵢᵣᵢ = 0 min. Upon irradiation the resonances decrease at almost identical rates while that for free CH₃CN at 1.96 ppm appears and increases at the same rate (λᵢᵣᵢ ≥ 455 nm). Although 6 has two electronically independent ¹MLCT transitions, no selectivity is observed in the photoinduced ligand exchange process. Like 2 and 3, complex 6 does not display any structural distortions about the metal center or the 3,3'-ibiq ligand and the photoreactivity of the three complexes is comparable. This observation agrees with the hypothesis that complexes that do not have ligands that impart steric strain do not undergo selective photoinduced ligand substitution.
Figure 4.12. Changes to the $^1$H NMR spectrum of 6 in CD$_3$CN as a function of irradiation time ($\lambda_{irr} \geq 455$).

Figure 4.13. Changes to the $^1$H NMR spectrum of 7 in CD$_3$CN as a function of irradiation time ($\lambda_{irr} \geq 455$).
The analogous experiment was carried out for complex 7 (Figure 4.13). Due to the presence of two isomers for 7, there are four resonances that correspond to the protons of the CH$_3$CN ligands. The peaks at 2.21 and 2.19 ppm correspond to an isomer while those at 2.22 and 2.17 ppm correspond to the other. Upon irradiation the signals at 2.19 and 2.17 ppm decrease in intensity with concomitant growth of a resonance at corresponding to the free CH$_3$CN peak at 1.96 ppm. After an hour of irradiation, these resonances disappear, while very little change is observed in those at 2.22 and 2.21 ppm indicative of selective ligand exchange in 7. Therefore, the photochemistry of 7 parallels that of 1 and 5, which was further confirmed by monitoring the photochemistry using electronic absorption spectroscopy. The spectral changes of 7 upon irradiation in H$_2$O are shown in Figure 4.14 ($\lambda_{irr} \geq 455$ nm). At early irradiation times the initial peak at 485 nm decreases and the growth of a new peak at 514 nm is observed, resulting in an isosbestic point at 497 nm, indicative of the formation of the monoaqua intermediate [Ru(phen)(1,1´-ibiq)(CH$_3$CN)(H$_2$O)]$^{2+}$ after 30 minutes of irradiation. The shift of 1164 cm$^{-1}$ is similar to that observed for the formation of the monoaqua intermediate upon photolysis of 1 (938 cm$^{-1}$, Chapter 3). Continued irradiation results in a decrease of the band at 514 nm and the concomitant growth of a new peak at 551 nm with an isosbestic point at 529 nm; this process corresponds to the formation of the diaqua species, [Ru(phen)(1,1´-ibiq)(H$_2$O)$_2$]$^{2+}$, however, after much longer irradiation (6 hours) than needed to form the monoaqua species. This large difference in irradiation times required to form the monoaqua and diaqua species in 7 parallels the reactivity observed for 1 and is consistent with a stable monoaqua intermediate and selective ligand exchange. The
excited state properties of 7 are similar to that of 1 with a lowest energy MLCT to the biquinoline ligand and a higher energy MLCT to the phen ligand, however, the 1,1′-biq ligand should not provide any steric strain about the metal center, such that bulky substituents pointing towards the metal must not be the only factor that dictates selective ligand exchange, as was believed following the interpretation of the data of the photochemistry of 3 – 5. As stated previously and confirmed by $^1$H NMR, the 1,1′-ibiq ligand is forced to tilt when coordinating to Ru(II), which results in two distinct isomers. The tilting is observed for 1 and 5, which undergo selective photoinduced ligand exchange, compromises the interaction of the bidentate ligands with the metal and causes distortions that decrease the overlap of the ligand lone pairs with the metal’s $e_g$-type orbitals. This distortion may give rise to the unusual wavelength dependent selective ligand exchange observed in these complexes.

**Figure 4.14.** Electronic absorption spectral changes of 7 from (a) 0 to 30 min and (b) 30 min to 360 min in H$_2$O ($\lambda_{\text{irr}} \geq 455$ nm).
Photoinduced ligand exchange is not observed in 8, even when irradiated with higher energy light ($\lambda_{\text{irr}} \geq 395\ \text{nm}$) in H$_2$O or CD$_3$CN. This lack of reactivity can be explained by the different identity of the lowest energy excited state, which $^3\pi\pi^*$ centered on the dap ligand instead of $^3\text{MLCT}$. Following excitation and ultrafast intersystem crossing to the triplet manifold, the complex must selectively populate this lowest energy excited state and the dynamics must be such that the complex unable to populate the dissociative $^3\text{LF}$. [Ru(bpy)(dppn)(CH$_3$CN)$_2$]$^{2+}$ also possesses a $^3\pi\pi^*$ lowest energy excited state, but is able to undergo photoinduced ligand dissociation. The photoreactivity observed in the dppn complex can be explained if the $^3\text{MLCT}$ is higher in energy in this complex relative to that in 8, which places it either closer in energy to $^3\text{LF}$ states or further away from the $^3\pi\pi^*$ state, both of which are expected to result in increased population of $^3\text{LF}$ states. Incorporation of sterically hindering ligands into Ru(II) systems with the dap ligand are currently under further investigation to induce ligand substitution.

The introduction of sterically hindering ligands into Ru(II) complexes has been shown to lower the energy of the $^3\text{dd}$ states below that of the $^3\text{MLCT}$ state, which is believed to result in enhanced quantum yield ($\Phi$) for ligand exchange in complexes of the formula [Ru(tpy)(L)(LL')]$^{2+}$, where L is a bidentate ligand and LL' is a monodentate ligand. It was found that when L is a sterically bulky ligand such as 2,2'-biq or 6,6'-dimethyl-2,2'-bipyridine, then the ligand exchange of L' increases significantly. The $\Phi_{400}$ values for 3 – 5 measured using ferrioxolate as an actinometer were found to be 0.22(4), 0.15(6), and 0.24(19), respectively. These values are similar to the first ligand
exchange step reported for 2, for which the photosubstitution is not selective.\textsuperscript{16} Interestingly, strategic placement of the methyl groups to introduce steric bulk in 5 does not result in a significant enhancement of the quantum yield, however, it does result in selectivity. The quantum yield values for the first ligand exchange for 6 and 7 using ferrioxolcate as an actinometer were measured to be 0.12(3) and 0.03(7), respectively, and it is notable that the value for 6 is similar to that of the dimethylphen complexes, whereas that for 7 is significantly lower than all other complexes, including that reported for 1.\textsuperscript{16} Because both 1 and 7 have Ru-biq lowest energy $^3$MLCT states, these results suggest that steric bulk is critical in lowering $^3$LF states in order to maintain an efficient ligand substitution. Due to the close proximity in energy of the $^3$LF states and $^3$MLCT states in 2 – 6, steric bulk does not have a drastic effect on $\Phi$, but the distortions away from octahedral geometry about the metal center by tilting of the diimine ligands must affect the the overlap in the $e_g$ Ru – ($\sigma^*$) orbitals in a manner that results in the selective ligand dissociation observed in 1, 5, and 7.

4.2.5 Calculations

Density functional theory (DFT) calculations were employed to gain a better understanding of the differences among the complexes that result in selective ligand exchange. For all the complexes, DFT calculations reveal metal based highest molecular orbital (HOMO), HOMO-1, and HOMO-2 levels representing the $d_{xy}$, $d_{xz}$, and $d_{yz}$ orbitals, with the exception of 6, which has significant dap ligand character in the HOMO. The lowest unoccupied molecular orbitals (LUMOs) of 3 – 5 exhibit electron density on the
phen ligand and a LUMO+1 on the corresponding dimethyl phen ligand while 2 exhibits LUMO and LUMO+1 orbitals that are delocalized over both equivalent phen ligands. The calculated MO diagrams for 2 – 6 are shown in Figure 4.15. In Figure 4.15, the HOMO of 2 was arbitrarily set to 0.0 eV and energies calculated for the HOMOs of other complexes are plotted relative to 2. As expected, the HOMO and LUMO orbitals of 2 – 5 are calculated at nearly identical energies, which agrees with the absorption and emission results. Furthermore, the LUMO+6 and LUMO+7 (dashed lines in Figure 4.15), which correspond to the e_g-type orbitals (d_{x^2-y^2} and d_{z^2}), are at similar energies of ~10.30 eV for the d_{x^2-y^2} orbitals and ~11.00 eV for the d_{z^2} orbitals in 2 – 5. The fact that there is little variation among the complexes is not in agreement with the hypothesis that steric strain lowers the energy of these orbitals. However, further analysis of the e_g-type orbitals reveals that the d_{z^2} in 5 is directed along the bond of only one of the CH3CN ligands and is lower in energy than the d_{x^2-y^2} orbital, while the opposite is observed for 2 – 4, with an antibonding orbital that is directed along the bonds of both CH3CN ligands (Figure 4.16). This difference in the e_g orbitals among complexes 2 – 5 indicates that introducing steric bulk with the 2,9-dmphen ligand does distort the symmetry about the metal center and results in Ru–L(σ*) interactions that could result in selective ligand dissociation upon excitation.
Figure 4.15. Molecular orbital diagrams of \(2 - 5\), where dashed lines represent Ru–L (\(\sigma^*\)) orbitals.

Figure 4.16. Electronic density plots of the calculated LUMO+6 of \(2, 3,\) and \(5\) (isovalue = 0.04).

A similar analysis was used to probe the selective ligand exchange in the biquinoline complexes, \(1\) and \(6 - 8\). The MO diagrams are shown in Figure 4.17 and the HOMO of \(1\) was arbitrarily set to 0.0 eV. The LUMOs of \(1\) and \(7\) are localized on the corresponding biq ligands and the LUMO+1 is localized on the phen ligand at similar energies. The LUMO And LUMO+1 of \(8\) are localized on the dap and the phen ligands,
respectively, while the LUMO and LUMO+1 of 6 are localized on the phen ligand and 3,3'-ibiq ligands, respectively, agreeing with electronic absorption assignments. It is clear in Figure 4.16, the LUMOs corresponding to the Ru–L (σ*) orbitals do not follow a distinct pattern in which the more sterically demanding ligand results in lower energy e\textsubscript{g} orbitals. However, a similar trend correlating selective ligand exchange and the orientation of the e\textsubscript{g} orbitals that was discussed for 2 – 5 is also observed for 1 and 6 (Figure 4.18).

![Molecular orbital diagrams](image)

**Figure 4.17.** Molecular orbital diagrams of 1 and 6 – 8, where dashed lines represent Ru–L (σ*) orbitals.

The LUMO+6 of 1 corresponds to the d\textsubscript{z\textsuperscript{2}} orbital and is directed along the CH\textsubscript{3}CN bond *trans* to the biq ligand, while the LUMO+6 in 6 assigned as the d\textsubscript{x\textsuperscript{2} - y\textsuperscript{2}} is directed along both bonds between the metal center and the CH\textsubscript{3}CN ligands. Placing electron density in the LUMO+6 antibonding orbital in 1 should result in weakening of the Ru–NCCH\textsubscript{3}, such that the preferential ligand dissociation of the CH\textsubscript{3}CN *trans* to the biq ligand would be expected. However, our previous research reveals that the more
photolabile CH$_3$CN in 1 is that positioned *trans* to the phen ligand. In contrast, no selective ligand substitution is observed in 6, which agrees with the calculations. Moreover, the calculated LUMO+6 in 7 is oriented similar to that of 6, but experimental evidence suggests that the former undergoes selective photoinduced ligand substitution.

Time dependent DFT (TD-DFT) calculations were utilized to obtain the electronic transitions, energies, and orbitals involved in those transitions to better understand the photodissociation process.

**Figure 4.18.** Electronic density plots of the calculated LUMO+6 of 1, 6, and 7 (isovalue = 0.04).

TD-DFT calculations reveal that the lowest singlet vertical singlet excited states for 3 – 5 possess a significant contribution of the HOMO → LUMO transition with moderate oscillator strengths, and maxima at 401 (83%, $f = 0.0053$), 398 (75%, $f = 0.0058$), and 403 (91%, $f = 0.0046$), respectively. The lowest energy vertical singlet excited states of 2 are calculated at 396 nm (81% HOMO → LUMO+1, $f = 0.0024$) and at 395 nm (91% HOMO → LUMO, $f = 0.0093$). It should be noted that the lowest energy electronic
transitions predicted are slightly blue-shifted relative to the experimental MLCT maxima, as is typical for DFT calculations,\textsuperscript{1} but the slight calculated red shift that is observed in 5 is consistent with experimental results. TD-DFT calculations were also used to determine the energy of the lowest energy vertical transition with contribution of the LUMO+6 orbitals, which are HOMO \( \rightarrow \) LUMO+6 and were calculated at 3.81 eV, 3.82 eV, 3.83 eV, and 3.52 eV for 2–5, respectively. These results suggest that the \(^1\)dd state for 5 is slightly lower in energy; moreover, the \( \text{e}_g \) orbitals associated with this state are lower in energy by \( \sim 0.3 \) eV than in 2–4 resulting from the distortion imposed by the 2,9-dmphen ligand, also observed in related systems.\textsuperscript{21} The lower energy of the \(^3\)dd state and the preferential orientation of the \( \text{e}_{g}^{*} \)-\( \text{Ru} \)--\( \text{L}(\sigma^{*}) \) orbitals induced by the asymmetric steric bulk imposed on the metal center results in the phenomenon of selective ligand exchange in these complexes.

TD-DFT calculations on 1 and 6–8 reveal that the lowest singlet vertical singlet excited states for 1, 7, and 8 possess a significant HOMO \( \rightarrow \) LUMO contribution, with maxima at 476 nm (83\%, \( f = 0.0053 \)), 482 nm (75\%, \( f = 0.0058 \)), and 543 nm (91\%, \( f = 0.0046 \)), respectively. The lowest energy vertical singlet excited state of 6 is calculated at 402.43 nm (82\% HOMO \( \rightarrow \) LUMO, 42\% HOMO-1 \( \rightarrow \) LUMO \( f = 0.0024 \)). The calculated transition energies for the complexes are in good agreement with the experimental data with the exception of 1. To probe the vertical energy transition energies involving the \( \text{e}_g \) orbitals the HOMO \( \rightarrow \) LUMO+6 transitions were analyzed for 1 and 6, as well as the HOMO \( \rightarrow \) LUMO+7 transitions for 7 and 8. These transition were calculated to be at 3.51 eV, 3.84 eV, 3.76 eV, and 3.81 eV for 1 and 6–8, respectively,
showing lower energies of the $^1$dd and $^3$dd states of 1. This result is consistent with that for 5, and both 1 and 5 were shown to undergo selective photoinduced ligand dissociation and have similar structural distortion about the metal center. The calculated values for 6 and 8 are similar to those for 2 – 4, as expected due to the lack of steric strain observed in the crystal structure of 6 and 8. Since calculations indicate that the $^3$dd state of 8 is similar in energy to those of 2 – 4 and experimental data indicate that the $^3$MLCT is significantly lower in energy than 2 – 4, then the thermal energy required to populate the $^3$dd must be too great to overcome in order for ligand substitution to occur. Moreover, relaxation to the low lying $^3\pi^*$ state in 8 may also be a factor that results in lack of ligand dissociation. Interestingly, the value for the lowest energy transition involving the HOMO → LUMO+7 in 7, is between that of 1 and 6. This indicates that the dd states are not as accessible in 7 and explains the low quantum yield, however, the selective ligand exchange observed in this complex is still not completely understood and is under further investigation.

4.3 Conclusions

In Chapter 3, the selective photoinduced ligand exchange in [Ru(phen)(2,2′-biq)(CH$_3$CN)$_2$]$^{2+}$ (1) was discussed, in which the CH$_3$CN ligand trans to the phen ligand exchanges preferentially upon irradiation with visible light in coordinating solvents, while there is no evidence of preferential ligand substitution in the symmetric complex [Ru(phen)$_2$(CH$_3$CN)$_2$]$^{2+}$ (2). In order to investigate this phenomenon further, the complexes [Ru(4,7-dmphen)(phen)(CH$_3$CN)$_2$]$^{2+}$ (3), [Ru(5,6-dmphen)(phen)(CH$_3$CN)$_2$]$^{2+}$
(4), [Ru(2,9-dmphen)(phen)(CH$_3$CN)$_2$]$^{2+}$ (5), [Ru(3,3′-ibiq)(phen)(CH$_3$CN)$_2$]$^{2+}$ (6), [Ru(1,1′-ibiq)(phen)(CH$_3$CN)$_2$]$^{2+}$ (7), and [Ru(dap)(phen)(CH$_3$CN)$_2$]$^{2+}$ (8) were synthesized and photophysical properties compared to each other as well as to 1 and 2. Structural analysis of 1 – 3, 5, 6, and 8 reveal that the ligands 2,2′-ibiq and 2,9-dmphen, which possess bulky substituents adjacent to the coordinating nitrogen atoms, result in a tilting of the diimine ligand and significant torsional strain about the metal center that is not observed in 2, 3, 6, and 8. Although 5 possesses almost identical ground state photophysical properties to 2 – 4, it undergoes selective photoinduced ligand dissociation similar to that observed in 1. Although complex 6 possess separate charge transfer states, selectivity is not observed suggesting distortion caused by steric strain about the metal center is essential for the process to occur. Complex 8 does not undergo ligand substitution upon irradiation due to the presence of a low lying $^{3}\pi\pi^*$ state. Although there is no evidence of steric strain in complex 7, selective ligand exchange is observed albeit at a much less efficient rate. Analysis of the optimized structure by DFT calculations reveals that tilting of the 1,1′-ibiq ligand must occur when bound to the metal and is consistent with two isomers present in the $^1$H NMR spectrum. This tilting is most likely similar to that of 1 and 5 and is a commonality between the complexes that undergo selective ligand exchange.

Calculations of the complexes indicate that the presence of ligands that induce steric strain disrupts the symmetry about the metal center and orients the $e_g$ type orbitals involved in bonds along only one of the photolabile CH$_3$CN ligands, while no preferential orientation is observed in the complexes that are symmetric about the metal center.
These Ru–L (σ*) orbitals are generally accepted to be involved in the ligand substitution process and are preferentially directed along one bond in those that undergo selective ligand exchange with the exception of complex 7. A platform has been generated that provides chemically inequivalent photolabile CH₃CN ligands in Ru(II) complexes. Time resolved techniques will be employed to further elucidate the differences in selective ligand exchange among the complexes and to gain a better understanding of the effect that these ligands have on the energy of the ³dd states. Better control and understanding of selective ligand substitution will be useful in the design of complexes that can release multiple drugs for applications in photochemotherapy.

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Chapter 5: Incorporation of the 2,2’-biquinoline Ligand in Sterically Strained Ru(II) Complexes for Bidentate Ligand Dissociation for Photochemotherapy


5.1 Introduction

Ruthenium(II) polypyridyl complexes possessing aromatic ligands have been shown to interact with DNA as chemotherapeutic agents and molecular light switches through intercalation and electrostatic interactions.\(^1\)-\(^4\) More recent research shows that some Ru(II) complexes have the ability to undergo photoinduced ligand exchange forming covalent bonds with DNA in a manner akin to cisplatin, such that these lesions may result in cell death. Unlike traditional photodynamic therapy (PDT) agents that rely on the generation of singlet oxygen for action, these photo-cisplatin analogs achieve cell death via mechanisms that are independent of oxygen; in order to differentiate the two methods, the latter is referred to as photochemotherapy (PCT).\(^5\)-\(^7\)

PCT involving transition metal complexes has generally focused on the exchange of monodentate ligands upon irradiation.\(^5\),\(^8\),\(^9\) The photoinduced ligand exchange of bidentate ligands bound to ruthenium(II), however, is well documented for sterically strained
complexes including those with ligands such as 2,2′-biquinoline (biq).\textsuperscript{10} For example, the photoinduced exchange of a biq ligand in [Ru(biq)\textsubscript{2}(bpy)]\textsuperscript{2+} (bpy = 2,2′-bipyridine) in CH\textsubscript{3}CN results in the formation of the intermediate cis-[Ru(biq)(bpy)(CH\textsubscript{3}CN)\textsubscript{2}]\textsuperscript{2+}, which can be used in the synthesis of tris-heteroleptic Ru(II) complexes of the type [Ru(biq)(phen)(L)]\textsuperscript{2+} in the presence of a variety of bidentate ligands, L.\textsuperscript{10} It was not until decades later that [Ru(biq)(phen)\textsubscript{2}]\textsuperscript{2+} and [Ru(biq)\textsubscript{2}(phen)]\textsuperscript{2+} (phen = 1,10-phenanthroline) were shown to exhibit cytotoxicity upon irradiation with visible light, while being relatively non-toxic under similar conditions in the dark.\textsuperscript{11} Both Ru(II) complexes undergo ligand dissociation in water following the absorption of visible light to generate the corresponding bis-aqua complexes; the latter covalently bind to DNA \textit{in vitro} and these adducts are believed to result in cell death.\textsuperscript{6,11}

Photoinduced ligand exchange occurs in complexes with \textsuperscript{3}LF (ligand field) dd states that are thermally accessible from the lower-lying energy \textsuperscript{3}MLCT (metal-to-ligand charge transfer) state(s).\textsuperscript{12-15} The thermal population of the metal centered \textsuperscript{3}LF state results in electron density on the e\textsubscript{g}-type orbitals with Ru-L(σ*) character, thus resulting in ligand dissociation.\textsuperscript{12-15} The exchange of bidentate ligands, however, is unusual because both bonds need to be broken upon MLCT excitation. As previously stated in Chapter 4, complexes with bulky biq ligands that sterically strain the conventional octahedral geometry in Ru(II) complexes are believed to lower the energy of the \textsuperscript{3}LF state relative to the \textsuperscript{3}MLCT state, thus resulting in enhanced photochemistry.\textsuperscript{6,11}

In Chapter 3, it was shown that incorporation of the 2,2′-biq ligand in Ru(II) complexes in place of phen results in a significant red shift of the absorbance. The
maximum of the MLCT absorption of [Ru(biq)$_2$(phen)]$^{2+}$, at 550 nm in H$_2$O, is outside the optimal excitation range for PCT, 600–850 nm.\textsuperscript{11} Therefore, biquinoline complexes able to undergo photochemical ligand exchange, but with lower energy absorption are desirable. Cyclometallated Ru(II) complexes have been utilized as light harvesters in solar energy conversion schemes because their MLCT absorption bands are broader and at lower energy relative to the diimine analogs.\textsuperscript{16-18} Also the dpb ligand (dpb = 2,3-bis(2-pyridyl)benzoquinoxaline) was integrated into the biquinoline systems to try and red shift the absorption further into the red and induce even more geometric strain. The present work focuses on the synthesis and characterization of [Ru(biq)$_2$(phpy)](PF$_6$)$_2$ (1) (phpy$^-$ = deprotonated 2-phenylpyridine). Also, the photophysical properties and photochemistry of 1 were investigated and compared to those of [Ru(biq)$_2$(bpy)](PF$_6$)$_2$ (2), [Ru(biq)$_2$(phen)](PF$_6$)$_2$ (3), and [Ru(biq)$_2$(dpb)](PF$_6$)$_2$ (4). A schematic representation of the molecular structures of 1 – 4 is displayed in Figure 5.1.

\textbf{Figure 5.1.} Schematic representation of the molecular structures of 1 – 4.
5.2 Results and Discussion

5.2.1 Electronic Absorption, Emission, and Electrochemistry

The absorption profiles of 2 and 3 are in close agreement with previously published data (Figure 5.2). Complex 2 exhibits maxima at 549 nm ($\varepsilon = 6600 \text{ M}^{-1}\text{cm}^{-1}$), 482 nm ($\varepsilon = 4800 \text{ M}^{-1}\text{cm}^{-1}$), and 407 nm ($\varepsilon = 2800 \text{ M}^{-1}\text{cm}^{-1}$) in CH$_3$CN, and those for 3 are observed at 552 nm ($\varepsilon = 9600 \text{ M}^{-1}\text{cm}^{-1}$), 480 nm ($\varepsilon = 7100 \text{ M}^{-1}\text{cm}^{-1}$), and 409 nm ($\varepsilon = 3900 \text{ M}^{-1}\text{cm}^{-1}$) in the same solvent. The transitions at ~410 nm have been assigned as Ru(t$_{2g}$)$\rightarrow$L(\pi*) (L = bpy, phen) $^1$MLCT in 2 and 3, respectively, whereas those at ~480 nm and ~550 nm arise from Ru(t$_{2g}$)$\rightarrow$biq(\pi*) transitions. Three $^1$MLCT absorption peaks are also observed in 1, but are red-shifted as compared to the corresponding bands in 2 and 3 (Figure 5.4), with Ru(t$_{2g}$)$\rightarrow$phpy(\pi*) absorption maximum at 455 nm ($\varepsilon = 1700 \text{ M}^{-1}\text{cm}^{-1}$), and bands associated with Ru(t$_{2g}$)$\rightarrow$biq(\pi*) transitions at 545 ($\varepsilon = 2200 \text{ M}^{-1}\text{cm}^{-1}$) and 640 nm ($\varepsilon = 5200 \text{ M}^{-1}\text{cm}^{-1}$) in CH$_3$CN. The red shift in 1 can be attributed to the increase in energy of the highest occupied molecular orbital (HOMO) resulting from cyclometallation, since the energy of the biq(\pi*) orbitals is expected to remain relatively unchanged. The carbon metal bond provides significant ligand character to the HOMO, which is typically nearly solely metal in character in Ru(II) polypyridyl complexes. Complex 4 exhibits a broad absorption band from 500 to 600 nm in CH$_2$Cl$_2$ with maxima at 519 nm ($\varepsilon = 5,400 \text{ M}^{-1}\text{cm}^{-1}$) and 567 nm (5,200 M$^{-1}$ cm$^{-1}$) arising from Ru(t$_{2g}$)$\rightarrow$biq(\pi*) and Ru(t$_{2g}$)$\rightarrow$dpb(\pi*) $^1$MLCT transitions, respectively (Figure 5.2). This has a similar shape and absorption tail past 600 nm as the complex
[Ru(bpy)_2(dpz)]^{2+}, which has a lowest energy \(^1\)MLCT transition Ru(t_{2g}) \rightarrow dpz(\pi^*), further confirming our transition assignments.\(^{21}\) Clearly the \(^1\)MLCT transitions to the bq and dpz overlap, but the distinct difference in shape between 4 relative to either 2 or 3 indicates the HOMO \rightarrow LUMO transition in 4 is Ru(t_{2g}) \rightarrow dpz(\pi^*)).

**Figure 5.2.** Electronic absorption spectra of complexes 1 – 3 in CH\(_3\)CN and complex 4 in CH\(_2\)Cl\(_2\).

Emission in the near-IR spectral region is observed from complex 1 in CH\(_3\)CN with maxima at 1030 nm and 980 nm at 298 K and 77 K, respectively (\(\lambda_{\text{exc}} = 405\) nm) shown in Figure 5.3, which is significantly red shifted from that of 2 and 3 with maxima at 748 nm and 747 nm at room temperature, respectively, and at 740 nm and 735 at 77 K, respectively, in agreement with published results.\(^{19}\) Also, emission from 4 is observed in the near-IR spectral region with a maximum at 860 nm at 77 K in CH\(_3\)CN (Figure 5.3). Emission in CH\(_3\)CN at 77 K from the complex [Ru(bpy)_2(dpz)]^{2+} was observed at 863
nm, confirming that the lowest energy excited state is the dpb \(^3\)MLCT state rather than the biq \(^3\)MLCT state as in the other three complexes.\(^{21}\)

**Figure 5.3.** Emission spectra of complexes 1, 2, and 4 in CH\(_3\)CN at 77 K.

Cyclic Voltammetry reveals quasi-reversible metal-centered oxidation events, \(E_{1/2}(\text{Ru}^{3+/2+})\), at +1.44 V vs SCE for 2 and 3 in CH\(_3\)CN and +1.57 V vs SCE for 4, which is typical of similar Ru(II) polypyridyl complexes.\(^{22,23}\) In the case of the cyclometallated complex, 1, the observed Ru(III/II) couple is observed at a less positive potential, with \(E_{1/2}(\text{Ru}^{3+/2+}) = +0.65\) V vs SCE. This cathodic shift in oxidation potential is typical of cyclometallated complexes relative to bpy or phen and results from the increased electron density on the metal provided by the covalent bonding of the phpy\(^-\) ligand.\(^{20}\) The increased electron density on the metal also increases the \(\pi\)-backbonding to the pyridyl rings of the biq ligands resulting in a cathodic shift in the sequential ligand centered reductions for 1 relative to those of 2 and 3.\(^{20}\) Quasi-reversible reduction waves assigned
as reduction of the biq ligands are observed with $E_{1/2}(\text{Ru}^{2+/+}) = -1.06$ and $-1.31$ vs SCE for 1 in CH$_3$CN, while the analogous reduction events of the biq ligands occur at $-0.80$ V and $-1.03$ V vs SCE for 2, at $-0.80$ V and $-1.05$ V vs SCE for 3, and at $-0.70$ V and $-0.98$ V for 4 in the same solvent. A third reduction event is observed in 2 and 3 in CH$_3$CN with $E_{1/2}(\text{Ru}^{2+/+}) = -1.59$ V and $E_{1/2}(\text{Ru}^{2+/+}) = -1.56$ V vs SCE, respectively, assigned to the reduction of the bpy and phen ligands. A third reduction event is also observed in 4 in CH$_3$CN with $E_{1/2}(\text{Ru}^{2+/+}) = -0.43$ V vs SCE assigned to the reduction of the dpb ligand. This reduction at less negative potentials is observed in other complexes containing the dpb ligand and confirms the electronic absorption assignments. The reduction of the phpy$^-$ ligand in 1 is not observed and must occur outside the spectral analysis window for CH$_3$CN. The photophysical and electrochemical data are outlined in Table 5.1.

<table>
<thead>
<tr>
<th>Complex</th>
<th>$\lambda_{abs}$/nm ($e$/M$^{-1}$ cm$^{-1}$)$^a$</th>
<th>$\lambda_{em}$/nm$^c$</th>
<th>$E_{1/2}$/V$^d$</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>640 (5.2)</td>
<td>980</td>
<td>+0.65, −1.06, −1.31</td>
</tr>
<tr>
<td>2</td>
<td>549 (6.6)</td>
<td>740</td>
<td>+1.44, −0.80, −1.03, −1.59</td>
</tr>
<tr>
<td>3</td>
<td>552 (9.6)</td>
<td>735</td>
<td>+1.44, −0.80, −1.05, −1.56</td>
</tr>
<tr>
<td>4</td>
<td>567 (5.2)$^b$</td>
<td>860</td>
<td>+1.57, −0.43, −0.80, −0.98</td>
</tr>
</tbody>
</table>

$^a$ In CH$_3$CN, at 298 K. $^b$ In CH$_2$Cl$_2$. $^c$ In CH$_3$CN at 77 K. $^d$ In CH$_3$CN, vs. Ag/AgCl; 0.1 M[n-Bu$_4$N][PF$_6$].

### 5.2.2 Transient Absorption

The ultrafast transient absorption spectra in CH$_3$CN of complexes 1 – 4 were obtained with an excitation wavelength of 325 nm (fwhm = 300 fs) and are depicted in
Figure 5.4. The data pertaining to complexes 2 and 3 (Figures 5.4b and 5.4c) reveals a relatively long lived excited state with a transient absorption feature spanning the wavelength range of ~380 nm to 500 nm and a bleaching of the ground state at 370 nm and 550 nm. This same positive transient absorption feature is observed in the complexes [Ru(biq)(phen)(CH\textsubscript{3}CN)]\textsuperscript{2+} and [Ru(biq)\textsubscript{2}(CH\textsubscript{3}CN)\textsubscript{2}]\textsuperscript{2+} and further confirms the assignment in Chapter 3 as a Ru→biq \textsuperscript{3}MLCT state. Although slightly red shifted by approximately 70 nm, a similar broad transient absorption is observed in the spectrum of complex 1 (Figure 5.4a). This feature most likely corresponds to the reduced biq ligand similar to that of 2 and 3. The difference in shape is due to the superimposed bleaching caused by the ground state absorption at 400 nm.
The fs-TA spectrum for complex 4 possesses a transient absorption feature with two distinct features at 380 nm and 430 nm, which are assigned as a Ru→biq $^3$MLCT state and a Ru→dpb $^3$MLCT state, respectively. The peak at 380 nm decays with $\tau = 32$ ps assigned as interconversion to the Ru→dpb $^3$MLCT state, which was confirmed to be lower in energy than the Ru→biq $^3$MLCT state by emission. Furthermore, fs-TA of the free dpb ligand results in a positive feature centered at 440 nm verifying the assignment of the lower energy peak to the Ru→dpb $^3$MLCT state. No evidence of ligand exchange is observed in the time frame of the ultrafast experiment for complexes 2 – 4 and can be
attributed to the low quantum yield, which will be discussed later. Since the complexes do not decay within the 2 ns time performed.

The ns-TA spectra in CH₂Cl₂ of complexes 1 – 4 are shown in Figure 5.5 (λₑₓ = 532 nm, fwhm = 8 ns). The spectra were recorded in CH₂Cl₂ in order to prevent ligand dissociation due to the lack of a flow cell to flow the sample over the duration of the experiment. Also, the excited state features in the nanosecond experiments are in agreement with those on the ultrafast timescale. The transient absorption at 450 nm is observed for complex 1 (Figure 5.5a) and can be fitted to monoexponential time decay with τ = 14 ns, however, no signal is observed for the ground state bleach in 1 at 640 nm. Moreover, the 14 ns lifetime overlaps with the instrumental response function and the excited lifetime is approximated to be 2 – 20 ns. Complexes 2 and 3 (Figure 5.5b and 5.5c) possess much longer lived excited state lifetimes with τ ~200 ns and τ ~400 ns, respectively, for all features including the peak at 400 nm assigned to Ru→biq ³MLCT state, the ground state bleach at 550 nm, and the broad absorption at 650 nm assigned as a ligand-to-metal charge-transfer (LMCT) transition from one of the neutral ligands to the formally oxidized Ru³⁺ ion of the ³MLCT state. The lifetimes of 2 and 3 are much longer than those complexes containing the biq ligand in Chapter 3 that are also capable of undergoing photoinduced ligand dissociation. The shortened lifetime upon cyclometallation was also observed in the complexes [Ru(bpy)(phpy)(dpnn)]²⁺ and [Ru(phpy)(dpnn)]²⁺ (dpnn = benzo[i]dipyrido[3,2-a:2′,3′-c]phenazine), which were expected to have lifetimes in the microsecond time regime due to low lying ³ππ* states of the dpnn ligand but had lifetimes of 25 ps and 45 ps, respectively.²⁰ The relatively fast
nonradiative deactivation in 1 can be attributed to the decreased energy difference between the $^3$MLCT and the ground states in this complex as compared to 2, which was confirmed by emission and electrochemistry and follows the energy gap law.

In the ns-TA spectrum of 4 a typical bleach of the ground state absorption is observed between 500 nm and 650 nm, along with positive transient absorption signal from 400 nm to 500 nm, expected to correspond to the reduced dpb ligand in the lowest energy excited state, Ru($t_{2g}$)$\rightarrow$dpb($\pi^*$) $^3$MLCT. Although the signal partially overlaps with the instrument response function, the signals at 430 nm and 560 nm were fitted to a monoexponential decay with $\tau = 57$ ns. These absorption features are similar to those reported for [Ru(bpy)$_2$(dpb)]$^{2+}$, which has an excited state lifetime of 66 ns, and further supports the assignment as Ru($t_{2g}$)$\rightarrow$dpb($\pi^*$) $^3$MLCT.$^{21}$
5.2.3 Photochemistry

The photoreactivity of 1 – 4 was evaluated by monitoring the changes to the electronic absorption spectrum of each complex as a function of irradiation time. As expected based on prior work, the irradiation of 2 and 3 in H$_2$O and CH$_3$CN with visible light ($\lambda_{\text{irr}} \geq 530$ nm or $\lambda_{\text{irr}} \geq 630$) results in exchange of one biq ligand with two molecules of coordinating solvent, generating [Ru(biq)(L)(H$_2$O)$_2$]$^{2+}$ and [Ru(biq)(L)(CH$_3$CN)$_2$]$^{2+}$ (L = bpy, phen), respectively (Figures 5.6), but the complexes are not reactive when kept in the dark under similar experimental conditions.$^{10,11}$ In contrast, complex 1 is not photochemically active in CH$_3$CN ($\lambda_{\text{irr}} \geq 530$ nm, Figure 5.7) or H$_2$O based on absorption
changes or when monitored by mass spectrometry as a function of irradiation time ($\lambda_{\text{irr}} \geq 530$ nm).

**Figure 5.6.** Electronic absorption spectral changes of (a) 2 (40 μM) with increasing irradiation times, $t_{\text{irr}}$, 0, 1, 2, 3, 5, 10, 15, 20, 30, 45, 60, 90, and 180 min and (b) 3 (15 μM) at $t_{\text{irr}} = 0, 2, 5, 10, 20, 30,$ and 60 min in H$_2$O ($\lambda_{\text{irr}} \geq 630$ nm).
Figure 5.7. Electronic absorption spectral changes of 1 (50 μM) with increasing irradiation times, $t_{irr}$, 0, 1, 2, 5, 10, 20, 30 min in CH$_3$CN ($\lambda_{irr} \geq 530$ nm).

The photolysis of 4 in H$_2$O and CH$_3$CN are illustrated in Figure 5.8. Irradiation with red light ($\lambda_{irr} \geq 645$ nm) in H$_2$O results in a decrease in the $^1$MLCT band at 520 nm with the concomitant growth of a new peak at 586 with an isosbestic point at 546 nm, which indicates the formation of a single product upon irradiation. Photolysis under similar experimental conditions in CH$_3$CN results in the uniform decrease of the broad $^1$MLCT absorption band and the formation of a sharper peak at 535 nm. No changes are observed in the spectra when the complex is kept in the dark for over 4 hours in H$_2$O and CH$_3$CN. The peaks of the photoproduct in water at 585 nm and that in CH$_3$CN at 535 nm are in agreement with those reported for [Ru(biq)$_2$(H$_2$O)$_2$]$^{2+}$ and [Ru(biq)$_2$(CH$_3$CN)$_2$]$^{2+}$, respectively,$^{24}$ indicating that the dpb ligand is exchanging with the two solvent molecules.
Figure 5.8 Electronic absorption spectral changes of 4 in (a) H$_2$O with increasing irradiation times, $t_{irr}$, 0, 1, 5, 10, 20, 30, 60, 90, 120, 180, 240, 300, and 390 min and (b) CH$_3$CN at $t_{irr}$, 0, 0.5, 1, 2, 3, 5, 10, 20, and 30 min ($\lambda_{irr} \geq 645$ nm).

The finding that one of the biq ligands of 4 does not exchange upon irradiation is of particular interest because prior work on 2 and other [Ru(biq)$_2$(L)]$^{2+}$ (L = diimine ligand) complexes results in the photodissociation of a biq ligand.$^{10,11}$ The photodissociation of dpb in 4 was further confirmed by monitoring the progress of the photolysis by mass spectrometry. Initially, the only peak observed in the spectrum is at m/z = 474, which corresponds to the parent ion peak of 4. Irradiation ($\lambda_{irr} \geq 530$ nm) of 1 in CH$_3$CN for 1 h results in a decrease of the peak at m/z = 474 signal and the formation of new peaks that correspond to [Ru(biq)$_2$(CH$_3$CN)$_2$]$^{2+}$ at m/z = 348.0, [Ru(biq)$_2$(CH$_3$CN)]$^{2+}$ at m/z = 327.0, [Ru(biq)$_2$]$^{2+}$ at m/z = 307.0, and the free dpb ligand at m/z = 335.1 (Figure 5.9).
No changes to the mass spectrum are observed when the solution is kept in the dark for the same amount of time.

![Mass Spectrum](image)

**Figure 5.9.** Mass spectrum of 1 (a) before irradiation and (b) after irradiation in CH$_3$CN ($\lambda_{irr} \geq 530$ nm).

The quantum yield for the exchange of the biq ligand for 2 in H$_2$O with $\lambda_{irr} = 550$ nm and $\lambda_{irr} = 600$ nm irradiation were measured to be 0.068(2) and 0.053(3), respectively. These values are factors of 3.4 and 2.2 greater than those measured for 3, $\Phi_{550} = 0.020(3)$ and $\Phi_{600} = 0.024(2)$ at each wavelength, respectively. A similar trend was observed between the sterically hindered complexes [Ru(bpy)$_2$(dmbpy)]$^{2+}$ and [Ru(bpy)$_2$(dmdpq)]$^{2+}$ (dmbpy = 6,6’-dimethyl-2,2’-bipyridine, dmdpq = 7,10-dimethyl-pyrazino[2,3-f]-1,10-phenanthroline) in which the exchange of the dmdpq ligand possessing fused aromatic rings occurred less efficiently than the analogous dmbpy complex, suggesting less rigid bidentate ligands enhance the ligand exchange. The
quantum yield values of 2 and 3 are also in agreement with the shortened lifetime of 2 relative to 3. More accessible $^3$LF states in 2 result in enhanced nonradiative excited state deactivation that would decrease the overall excited state lifetime, which is observed in other Ru(II) polypyridyl complexes. The quantum yields for the dpb ligand exchange in 4 were measured to be $\Phi_{550} = 0.015(4)$ and $\Phi_{600} = 0.0094(8)$ in H$_2$O; these values are slightly lower than those reported for 2 and 3 under similar conditions. The lower quantum yields for 4 can be attributed to the poor solubility of the dpb ligand relative to the biq ligand in H$_2$O, which would make the escape from the solvent cage less favorable for the former and its subsequent recombination to regenerate 4 more likely. It should be noted that no photoinduced ligand exchange is observed for [Ru(bpy)$_2$(dpb)]$^{2+}$ ($\lambda_{irr} \geq 600$ nm), indicating that the steric bulk provided by the biq ligands play a critical role in the dissociation of the dpb ligand. It is hypothesized that the preferential photodissociation of the dpb ligand instead of biq in 4 occurs because the distortion caused by the addition of the bulky dpb ligand is greater than that caused by biq, making it more favorable to exchange the former; this point is further analyzed in the crystallography section. Another possible explanation of this observation is that the electronic properties introduced by the addition of the dpb ligand in 4 relative to the phen ligand in 3 results in excited state properties that favor dpb ligand dissociation.

5.2.4 X-ray crystal structures

The X-ray crystal structure of 1 is shown in Figure 5.10 and additional crystallographic data appear in Table 5.2. Compound 1 crystallizes in the monoclinic
space group $P2_1/n$ and there are two interstitial dichloromethane molecules in the asymmetric unit. The metal center is surrounded by five nitrogen atoms and one carbon atom in a distorted octahedral environment. The Ru1–C1 bond length of 2.095(4) Å in 1 is longer than the corresponding bond distances in [Ru(bpy)$_2$(phpy)]$^+$, 2.044(1) Å,\textsuperscript{26} and in [Ru(phen)$_2$(phpy)]$^+$, 2.036(7) Å,\textsuperscript{27} which may be attributed to the steric repulsion between the biq ligands. Three of the Ru–N bond lengths with the biq ligands, Ru1–N2, Ru1–N4 and Ru1–N5, are similar (~2.09 Å) and within the range of those found in the closely related compound 3 and [Ru(bpy)$_2$(biq)]$^{2+}$, 2.079(2) to 2.0112(3) Å.\textsuperscript{11} In contrast, the Ru1–N$^{\text{biq}}$ bond located $trans$ to the C donor atom of phpy$^-$ is the longest, 2.148(3) Å, reflecting the strong $trans$ influence of the phenyl ring of phpy$^-$.

The angle between adjacent biq ligands, N3–Ru1–N4 was determined to be 98.3(1)$^\circ$, which is greater than the angles formed between each biq and phpy$^-$, N1–Ru1–N3 and C1–Ru1–N4, measured to be 90.8(1) and 92.4(2)$^\circ$, respectively (Table 1). The greater N3-Ru1-N4 is likely necessary to reduce the steric repulsion between the two bulky biq ligands. The biq ligand $trans$ to the C1 atom of phpy$^-$ is twisted about the C–C bond, N2–C20–C21–N3, –9.9(6)$^\circ$, whereas such a distortion is not observed in the other biq ligand, N4–C38–C39–N5, 0.9(6)$^\circ$ (Table 5.2). In addition, the quinoline moieties of both biq ligands are bent by ~15$^\circ$ out of the plane formed with the metal center, a distortion that was also observed in 3.\textsuperscript{11} However, 1 is not photoactive, therefore, electronics play a crucial role in ligand dissociation in cyclometallated complexes.
Figure 5.10. Thermal ellipsoid plot of the [PF6]– salt of complex 1 (ellipsoids drawn at 50% probability).

Table 5.2. Selected bond lengths, angles, and dihedral angles for the cation \([\text{Ru(biq)}_2(\text{phpy})]^+\) (1).

<table>
<thead>
<tr>
<th>Bond Lengths (Å)</th>
<th>Bond Angles (°)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ru1–C1</td>
<td>2.095(4)</td>
</tr>
<tr>
<td>Ru1–N1</td>
<td>2.087(4)</td>
</tr>
<tr>
<td>Ru1–N2</td>
<td>2.092(3)</td>
</tr>
<tr>
<td>Ru1–N3</td>
<td>2.148(3)</td>
</tr>
<tr>
<td>Ru1–N4</td>
<td>2.091(4)</td>
</tr>
<tr>
<td>Ru1–N5</td>
<td>2.096(3)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Dihedral Angles (°)</th>
</tr>
</thead>
<tbody>
<tr>
<td>N2–C20–C21–N3</td>
</tr>
<tr>
<td>C19–C20–C21–C22</td>
</tr>
<tr>
<td>N4–C38–C39–N5</td>
</tr>
<tr>
<td>C37–C38–C39–C40</td>
</tr>
<tr>
<td>N2–Ru1–N3–C29</td>
</tr>
<tr>
<td>N4–Ru1–N5–C47</td>
</tr>
</tbody>
</table>
Structural analysis of complexes 3 and 4 can be used to aid in understanding the differences in ligand photodissociation among the complexes. The ORTEP diagram resulting from X-ray diffraction of the single crystal of 4 is shown in Figure 5.11, along with the corresponding numbering schemes of the atoms of interest. The two fused benzene rings on the quinoline portions of each biq ligand have been omitted from the structure of 4 for clarity. The crystal structure of 3 was previously reported, and selected structural parameters are compared to those of 4 in Table 5.3.

Figure 5.11. ORTEP plot of 4 drawn at 50% probability with selected atom numbers; solvent, hydrogen atoms, counteranions, omitted for clarity. The fused benzene rings on the quinoline portion of the biq ligands have been omitted for clarity.
Table 5.3. Selected experimental bond distances and dihedral angles, where L₂ is dpb in 4 and phen in 3.

<table>
<thead>
<tr>
<th>Bond distance (Å)</th>
<th>4</th>
<th>3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ru-N₁-biq(1)</td>
<td>2.077(2)</td>
<td>2.084(2)</td>
</tr>
<tr>
<td>Ru-N₂-biq(1)</td>
<td>2.127(2)</td>
<td>2.079(2)</td>
</tr>
<tr>
<td>Ru-N₃-biq(2)</td>
<td>2.096(2)</td>
<td>2.088(2)</td>
</tr>
<tr>
<td>Ru-N₄-biq(2)</td>
<td>2.117(2)</td>
<td>2.093(2)</td>
</tr>
<tr>
<td>Ru-N₅-L₂</td>
<td>2.072(2)</td>
<td>2.098(2)</td>
</tr>
<tr>
<td>Ru-N₆-L₂</td>
<td>2.143(2)</td>
<td>2.104(3)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Torsional Angle (°)</th>
<th>4</th>
<th>3</th>
</tr>
</thead>
<tbody>
<tr>
<td>N₁-Ru-N₂-C₉-biq(1)</td>
<td>8.51</td>
<td>-20.41</td>
</tr>
<tr>
<td>N₁-C-C-N₂-biq(1)</td>
<td>-12.67</td>
<td>2.7</td>
</tr>
<tr>
<td>N₃-Ru-N₂-C₉-biq(2)</td>
<td>-18.30</td>
<td>-19.5</td>
</tr>
<tr>
<td>N₃-C-C-N₄-biq(2)</td>
<td>9.58</td>
<td>12.1</td>
</tr>
</tbody>
</table>

Similar to 1, the coordination sphere around the Ru(II) metal center in 3 and 4 consists of six nitrogen atoms in a distorted octahedral geometry. The distortion results in lengthened Ru-N bonds in 4, which range from 2.072(2) to 2.143(2) Å (Table 5.3), as compared to the average bond lengths of [Ru(bpy)₃]²⁺, 2.057(3) Å.²⁸ The longest Ru–N bonds in 4 and 3 correspond to those to the dpb and phen ligands, respectively. Photodissociation of dpb is observed for 4, whereas the exchange of one of the biq ligands upon irradiation was reported for 3, attributed to the torsional angular strain displayed by the biq ligands. However, the torsional strain along one of the ruthenium-biq bonds is similar in 4 and 3, as much as 18° in 4 (N₂-Ru-N₄-C₂₇-biq(2)) and 20° in 3 (N₁-Ru-N₂-C₁₀-biq(2)). Furthermore, the strain between the quinolone rings in biq are both of similar magnitude for the most strained biq ligand in 4 (N₁-C₉-C₁₀-N₂-biq(1) = -12.67°) and
3 (N₃-C₂₈-C₂₇-N₄-biq(2) = +12.1%). Despite the similar magnitude of steric strain in 3 and 4, complex 4 displays preferential photosubstitution of the dpb ligand instead of one of the biq ligands, as is the case in 3. The photochemistry of 4 does not follow the previous crystallographic arguments, since the torsional strain about the ruthenium-dpb, N₅-Ru-N₆-C₄₁-dpb = –8.93°, is significantly smaller than that about the ruthenium-biq, N₃-Ru-N₄-C₂₇-biq(2) = –18.30°.

A possible explanation for the selective photoinduced dpb dissociation in 4 is the free rotation about the Ru-N₆-dpb bond once the Ru-N₅-dpb bond is broken. In contrast to biq, the pyrazine unit of dpb is expected to be better able to freely rotate away from the metal center, thus removing N₆-dpb from the vicinity of the metal. Such rotation in the η¹-dpb intermediate is expected to decrease its rate of re-association and favor the kinetics of solvent coordination to the open coordination site. In addition, the electronic structure must be considered when analyzing the ligand dissociation process in these complexes. In contrast to 3, the lowest energy ³MLCT state in 4 is Ru → dpb(π*) in character. The biq ligand has been shown to photodissociate when the lowest energy excited state is Ru → biq(π*) ³MLCT, placing electron density on one of the biq ligands,¹⁰,¹¹ but introducing a lower energy excited state in the complex, such that is the case in 4, may result in the dissociation of other bulky ligands such as dpb. This concept is currently under investigation.
5.2.5 Calculations

Density functional theory (DFT) and time dependent DFT (TD-DFT) calculations were performed to gain a better understanding of the electrochemistry, photophysical properties, and photochemistry in the four complexes. DFT calculations reveal a metal-based HOMO, HOMO-1, and HOMO-2 representing the $d_{xy}$, $d_{xz}$, and $d_{yz}$ orbitals for complexes 1 – 4. Also the HOMO of 4 possesses significant contribution from the dpb ligand. These HOMOs are calculated at nearly identical energies for 2, 3, and 4 but that of 1 is destabilized by $\sim0.9$ eV and exhibits significant phpy$^-$ ligand character and electron density on the metal-carbon bond and the MO diagrams are shown in Figure 5.12. The HOMO of 2 in Figure 5.12 was arbitrarily set at 0.0 eV. The differences in the calculated energies of the HOMOs agree with the cathodic shift in the experimental oxidation potential of 0.75 V between 1 and 2, 3, or 4 . The additional ease in oxidation by $\sim0.15$ V may be related to the overall +1 charge of 1 as compared to $+2$ of 2 and 3, making removal of an electron from the former more favorable. The LUMO and LUMO+1 orbitals of 1 – 3 are calculated to be localized on the biq ligands and to lie at similar energies in the three complexes (Figure 5.12). Although 1 is harder to reduce than 2 and 3 by $\sim0.2$ V, this difference may also be related to the difference in the overall charge of the complexes. The LUMO of 4 is calculated to be localized on the dpb ligand and is slightly lower in energy than the LUMOs of 1 by $\sim0.7$ V and of 2/3 by $\sim0.3$ eV. This agrees with the experimental electrochemical data that confirms the dpb ligand is reduced at a less negative potential than the biq ligand.
Figure 5.12. Calculated (a) MO diagrams showing the frontier orbitals and electrons densities of the (b) HOMOs and (c) LUMOS (isovalue = 0.04) of 1, 2, and 4. Hydrogen atoms were removed from the structures for clarity.

TD-DFT calculations reveal that the lowest vertical singlet excited states of 1, 2, and 3 possess significant contribution, ~88% for 1 and ~97% for 2 and 3, from HOMO→LUMO transitions, but low oscillator strengths, with maxima at 840 nm ($f = 0.0013$), 586 nm ($f = 0.0003$), and 587 nm ($f = 0.0004$), respectively (Tables S2–S4). However, the lowest vertical singlet excited state of 4 possesses significant contribution
from HOMO-1→LUMO (~61%) in addition to contribution from HOMO→LUMO (~34%) with a maximum at 635 nm ($f = 0.0007$). More intense absorption bands are predicted at 686 nm ($f = 0.027$) for 1, at 523 nm ($f = 0.076$) for 2, and at 527 nm ($f = 0.086$) for 3, calculated to possess 83%, 93%, and 96%, contribution from HOMO-1→LUMO transitions, respectively, and at 573 nm ($f = 0.0084$) for 4 that possesses 87% contribution HOMO-2→LUMO . These calculated absorption maxima are similar to the experimental values, 640 nm, 549 nm, 552 nm, and 585 nm for 1, 2, 3, and 4, respectively.

**Table 5.4.** 10 lowest energy singlet excited states obtained from DFT calculations, and the transitions associated with these states in CH$_3$CN (H = HOMO, L = LUMO) for complex 1.

<table>
<thead>
<tr>
<th>Wavelength (nm)</th>
<th>$f$</th>
<th>Calculated Transitions and Orbital Contributions$^a$</th>
</tr>
</thead>
<tbody>
<tr>
<td>839.53</td>
<td>0.0013</td>
<td>H→L(88%), H→L+1(8%)</td>
</tr>
<tr>
<td>765.85</td>
<td>0.0004</td>
<td>H→L+1(86%), H→L(9%), H-1→L+1(2%)</td>
</tr>
<tr>
<td>686.27</td>
<td>0.0266</td>
<td>H-1→L(83%), H-2→L(13%)</td>
</tr>
<tr>
<td>636.97</td>
<td>0.0288</td>
<td>H-2→L(46%), H-2→L+1(23%), H-1→L+1(18%), H-1→L(10%)</td>
</tr>
<tr>
<td>606.78</td>
<td>0.0397</td>
<td>H-2→L(39%), H-1→L+1(29%), H-2→L+1(26%)</td>
</tr>
<tr>
<td>537.81</td>
<td>0.0336</td>
<td>H-2→L+1(42%), H-1→L+1(41%), H→L+3(7%)</td>
</tr>
<tr>
<td>461.97</td>
<td>0.0068</td>
<td>H→L+2(52%), H→L+4(43%)</td>
</tr>
<tr>
<td>455.84</td>
<td>0.0122</td>
<td>H→L+4(46%), H→L+2(39%), H→L+3(10%)</td>
</tr>
<tr>
<td>438.86</td>
<td>0.0396</td>
<td>H→L+3(76%), H→L+2(5%)</td>
</tr>
<tr>
<td>418.32</td>
<td>0.0302</td>
<td>H-1→L+2(85%), H-1→L+4(5%)</td>
</tr>
</tbody>
</table>

$^a$Only contribution ≥5% are listed.
Table 5.5. 10 lowest energy singlet excited states obtained from DFT calculations, and the transitions associated with these states in CH$_3$CN (H = HOMO, L = LUMO) for complex 2.

<table>
<thead>
<tr>
<th>Wavelength (nm)</th>
<th>$f$</th>
<th>Calculated Transitions and Orbital Contributions$^a$</th>
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</thead>
<tbody>
<tr>
<td>586.3</td>
<td>0.0003</td>
<td>H$\rightarrow$L(97%)</td>
</tr>
<tr>
<td>560.87</td>
<td>0.00004</td>
<td>H$\rightarrow$L+1(90%), H-1$\rightarrow$L+1(4%)</td>
</tr>
<tr>
<td>522.69</td>
<td>0.0762</td>
<td>H-1$\rightarrow$L(93%)</td>
</tr>
<tr>
<td>514.61</td>
<td>0.0041</td>
<td>H-2$\rightarrow$L(96%)</td>
</tr>
<tr>
<td>491.68</td>
<td>0.0126</td>
<td>H-2$\rightarrow$L+1(90%), H-1$\rightarrow$L(5%)</td>
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<tr>
<td>476.43</td>
<td>0.0013</td>
<td>H$\rightarrow$L+2(97%)</td>
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<tr>
<td>467.09</td>
<td>0.0527</td>
<td>H-1$\rightarrow$L+1(76%), H-2$\rightarrow$L+2(6%), H$\rightarrow$L+1(6%)</td>
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<tr>
<td>439.47</td>
<td>0.0015</td>
<td>H-1$\rightarrow$L+2(96%)</td>
</tr>
<tr>
<td>404.76</td>
<td>0.0388</td>
<td>H-2$\rightarrow$L+2(85%)</td>
</tr>
<tr>
<td>389.55</td>
<td>0.005</td>
<td>H-3$\rightarrow$L(95%)</td>
</tr>
</tbody>
</table>

$^a$Only contribution $\geq$5% are listed.

Table 5.6. 10 lowest energy singlet excited states obtained from DFT calculations, and the transitions associated with these states in CH$_3$CN (H = HOMO, L = LUMO) for complex 3.

<table>
<thead>
<tr>
<th>Wavelength (nm)</th>
<th>$f$</th>
<th>Calculated Transitions and Orbital Contributions$^a$</th>
</tr>
</thead>
<tbody>
<tr>
<td>587.43</td>
<td>0.0004</td>
<td>H$\rightarrow$L(98%)</td>
</tr>
<tr>
<td>550.42</td>
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<tr>
<td>527.23</td>
<td>0.0026</td>
<td>H-2$\rightarrow$L(96%)</td>
</tr>
<tr>
<td>526.72</td>
<td>0.0862</td>
<td>H-1$\rightarrow$L(96%)</td>
</tr>
<tr>
<td>493.16</td>
<td>0.0037</td>
<td>H-2$\rightarrow$L+1(93%)</td>
</tr>
<tr>
<td>467.2</td>
<td>0.0008</td>
<td>H$\rightarrow$L+2(97%)</td>
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<tr>
<td>464.34</td>
<td>0.0567</td>
<td>H-1$\rightarrow$L+1(75%), H-2$\rightarrow$L+2(7%), H$\rightarrow$L+1(6%)</td>
</tr>
<tr>
<td>435.34</td>
<td>0.0006</td>
<td>H-2$\rightarrow$L+2(97%)</td>
</tr>
<tr>
<td>420.11</td>
<td>0.0005</td>
<td>H$\rightarrow$L+3(72%), H-2$\rightarrow$L+2(22%)</td>
</tr>
<tr>
<td>399.83</td>
<td>0.0208</td>
<td>H-1$\rightarrow$L+3(37%), H-3$\rightarrow$L+2(33%), H$\rightarrow$L+3(19%)</td>
</tr>
</tbody>
</table>

$^a$Only contribution $\geq$5% are listed.
In order to understand the lack of photodissociation of the biqu ligand in 1 and photodissociation of the dpb ligand in 4 as compared to 2 and 3, the orbitals and transitions involved in the process need to be considered. As stated previously, ligand dissociation is believed to occur via thermal population of the $^3$LF states from the low-lying $^3$MLCT state(s).\textsuperscript{12-15} Experimental data, calculations, and previous work indicate that the HOMO in 1 lies at a higher energy than those of 2 and 3, but the energies of the LUMOs remain relatively unchanged among the three complexes.\textsuperscript{16} It is expected that cyclometalation results in a larger gap between the $^3$MLCT state and $^3$LF states because the $e_g(\sigma^*)$ orbitals are destabilized due to the increased covalent interaction provided by the Ru–C bond, significantly raising the energy of the $e_g$ orbitals with Ru–L($\sigma^*$)
The Character of the e_g(σ*) orbitals in 2 are calculated as the LUMO+9 (d_2^2) and LUMO+10 (d_2^2, d_{x-y}^2), are relatively close in energy (ΔE = 0.26 eV). In contrast, the e_g(σ*) orbitals in 1 are calculated as the LUMO+10 (d_2^2) and LUMO+17 (d_{x-y}^2) with an energy difference of 3.22 eV.

The relative energies of the two e_g(σ*) orbitals is related to the energies of the 3^1LF states involving the population of each d-orbital, d_2^2 and d_{x-y}^2. In order for photodissociation of the bidentate ligand to take place, both Ru–N(biq) bonds must be broken. Because the 3^1LF state associated with the d_{x-y}^2 orbital lies at a very high energy in 1, it is likely not accessible upon irradiation with visible light. It is proposed that although one Ru–N(biq) bond may break upon irradiation, it quickly re-coordinates to the metal. This explanation also is consistent with the observed exchange of only one CH_3CN in the related cyclometallated complex [Ru(phpy)(bpy)(CH_3CN)_2]^+. Upon irradiation, one bond is weakened due to population of the lower lying energy 3^1LF state resulting in ligand dissociation and solvent coordination, but the remaining CH_3CN does not exchange upon extended photolysis because the higher-lying 3^1LF state is not populated.

In 4, the dpb ligand is photoejected rather than the biq ligand. To our knowledge this is the first instance of photoejection of a bidentate ligand in a bis-biq complex that is not one of the biq ligands. Calculations support our assignment of a lowest energy 1^1MLCT state localized on the dpb ligand rather than the biq ligand as observed in 2 and 3. Placing electron density on this ligand must cause geometric distortion resulting in
dissociation of the dpb ligand that alleviates the geometric strain in the complex and forms the more stable bis-biq complex.

5.2.6 Gel Mobility Assays

Gel electrophoresis mobility shift assays were carried out to compare the photoinduced DNA binding of 2, 3, and 4. It is well documented that cisplatin thermally binds to linearized DNA and reduces its migration through an agarose gel in a concentration-dependent manner. The same pattern is observed for 2 and 3 upon irradiation with low energy light, but not in the dark (Figure 5.13). In Figure 5.13, lanes 1 and 8 contain 1 kb DNA ladder, lanes 2 - 7 were loaded with 50 μM pUC18 DNA, and lanes 3 – 6 contain increasing concentrations of 2 or 3. In Figures 5a and 5c, the samples in lanes 3 – 6 were irradiated for 15 minutes with λ_{irr} ≥ 630 nm light prior to loading. It is evident in Figure 5.13a that, as the concentration of 2 is increased, the DNA mobility decreases, whereas no shift in mobility is observed when the samples are incubated in the dark for 20 min under similar experimental conditions (Figure 5.13b). These results are indicative of covalent binding of 2 to DNA only upon irradiation. Figures 5.13c and 5.13d display the results for complex 3 irradiated under the same conditions as 2 (Figure 5.13a) and in the dark, respectively. The reduced effect of 3 as compared to 2 on the DNA mobility can be attributed to the ~2-fold lower Φ_{600} value of the former, such that a smaller amount of the photoproduct able to bind to DNA is formed. Increased DNA binding was observed for 3 when the irradiation times were doubled, but it still remained lower than the effect observed for 2.
Figure 5.13. Imaged ethidium bromide-stained agarose gels of 50 μM linearized pUC18 plasmid (10 mM phosphate buffer, pH = 7.8) in the presence of various concentrations of complex: lanes 1 and 8, 1 kb DNA molecular weight standard; lanes 2 and 7, linearized plasmid alone; lanes 3-6, 25, 50, 75, 100 μM of 2 (a) irradiated (λ_{irr} ≥ 630 nm, 15 min), (b) incubated in the dark (20 min, 298 K) and 3 (c) irradiated (λ_{irr} ≥ 630 nm, 15 min), and (d) incubated in the dark (20 min, 298 K).

Gel electrophoresis mobility shift assays of complex 4 are shown in Figure 5.14. The DNA binding interactions were measured under identical conditions as 2 and 3. In Figure 5.14a, it can be seen that the migration of linearized DNA through the agarose gel is reduced in a concentration-dependent manner. This inhibition of the DNA migration through the gel when 4 is irradiated does not occur to the extent as in 2 and displays a similar amount of covalent binding as 3. This can be attributed to the quantum yield again. Since 4 has a similar quantum yield value of ~ 0.01 in H_2O and 3 has a quantum
yield that is \( \sim 0.02 \), then the extent of the DNA binding interaction of 4 should be similar under analogous conditions. Also, 4 does not display any covalent binding to DNA in the dark (Figure 5.14b).

**Figure 5.14.** Imaged ethidium bromide-stained agarose gels of 50 \( \mu \)M linearized pUC18 plasmid (10 mM phosphate buffer, pH = 7.8) in the presence of various concentrations of complex: lanes 1 and 8, 1 kb DNA molecular weight standard; lanes 2 and 7, linearized plasmid alone; lanes 3-6, 25, 50, 75, 100 \( \mu \)M of 4 (a) irradiated (\( \lambda_{irr} \geq 630 \) nm, 15 min), (b) incubated in the dark (20 min, 298 K).

### 5.2.7. Cytotoxicity

Percent cellular uptake values were measured to be 48 \( \pm \) 12 %, 8 \( \pm \) 5%, 4 \( \pm \) 2%, and 25\% \( \pm \) 5\% for complexes 1 – 4, respectively, after 24 hours of incubation in the dark with HeLa cells (Figure 5.15). Similar to cisplatin, the complexes must passively diffuse through the hydrophobic cell membrane environment to enter the cell. The poor cellular uptake of 2 and 3 is due to the over 2+ charge of the complex creating an unfavourable interaction with the hydrophobic cell membrane environment and is similar to related
compounds studied in recent work under similar conditions.\textsuperscript{31} Although 4 possesses a 2+ charge, its cellular uptake is significantly greater due to the presence of the large dpb ligand, which increases its overall hydrophobicity. Increased cellular uptake is typical of Ru(II) complexes with multiple large aromatic ligands.\textsuperscript{32} The relatively large cellular uptake measured for 1 is most likely due to the overall 1+ charge of the complex, which makes traversing the cell membrane more favourable.

\begin{figure}
\centering
\includegraphics[width=0.5\textwidth]{cellular_uptake.png}
\caption{Percent cellular uptake of complexes 1 – 4 (200 \textmu M, 24 hours) by HeLa cells cultured in Dulbecco’s modified eagle medium, supplemented with 10% fetal bovine serum and 1% penicillin/streptomycin at 37 °C in a humid incubator with 5% CO\textsubscript{2}.}
\end{figure}

Since a significant percent of complexes 1 and 4 enter the cell, dark toxicity and phototoxicity studies were conducted using HeLa cells and the results are shown in Figure 5.16. The IC\textsubscript{50} values upon irradiation for 1 (633 nm ± 20 nm) and 4 (522 nm ± 20 nm) were measured to be 1.04 \textmu M and 6.70 \textmu M, respectively. The important factor in PCT is the relative toxicity when the complex is kept in the dark versus when it is
irradiated, given by PI = IC_{50(dark)}/IC_{50(irr)}. Both complexes exhibit enhanced toxicity upon irradiation and although complex 1 was concluded to not be photoactive, its phototoxicity enhancement is greater than that of 4 with PI values equal to ~7 and ~2, respectively. Studies are currently in progress to further investigate the photocytoxic mechanism for 1 and 4 to potentially design more effective PCT agents.

![Cytotoxicity of (a) 1 and (b) 4 irradiated and in the dark.](image)

**Figure 5.16.** Cytotoxicity of (a) 1 and (b) 4 irradiated and in the dark.

### 5.3 Conclusions

The new cyclometallated complex [Ru(biq)_2(phpy)](PF_6) (1) and the complex [Ru(biq)_2(dpbi)](PF_6) (4) were synthesized and characterized by various methods. The photophysical properties, electrochemistry, and photochemistry of 1 and 4 were compared to those of known photoactive complexes [Ru(biq)_2(bpy)](PF_6)_2 (2) and [Ru(biq)_2(phen)](PF_6)_2 (3). Complexes 2 and 3 undergo exchange of one of the biq ligands when irradiated with \( \lambda_{irr} \geq 630 \) nm light in water to generate the corresponding complex, [Ru(biq)_2(bpy)(H_2O)](PF_6)_2 and [Ru(biq)_2(phen)(H_2O)](PF_6)_2. The resulting
bis-aqua photoproduct covalently binds to linearized ds-DNA. The quantum yield for ligand exchange for 2, $\Phi_{600} = 0.053(3)$, was measured to be 2.2-fold greater than that determined for relative to 3, $\Phi_{600} = 0.024(2)$, with the same irradiation wavelength, 600 nm, providing support to the hypothesis that less rigid ancillary ligands lead to more efficient biq ligand exchange. The differences in ligand exchange quantum yields of 2 and 3 were correlated to the DNA binding ability of the complexes.

Since [Ru(bpy)$_3$](PF$_6$)$_2$ and [Ru(phen)$_3$](PF$_6$)$_2$ are stable upon irradiation relative to the the biq complexes 2 and 3, steric bulk was thought to be a key criteria for photoinduced bidentate ligand exchange. Although the crystal structure of 1 reveals an elongated Ru–N(biq) bond, the complex does not display photoinduced ligand substitution in coordinating solvents under similar conditions as those used for 2 and 3. The difference in reactivity of the cyclometalled complex 1 may be ascribed to the increase in energy of the metal-centered $^3\text{LF}$ states resulting from the bonding of the strong $\sigma$-donor phpy$^-$ ligand, a finding supported by DFT calculations. Complexes 2 and 3 were thought to be good candidates as PCT agents owing to their covalent DNA binding upon irradiation with light in the photodynamic window but their potential may be limited to their lack of cellular uptake. Although the absorption maximum of 1 is red-shifted relative to those of 2 and 3, the lack of photochemistry of the former indicates that the use of cyclometallated Ru(II) complexes may not be a good strategy for the discovery of new PCT agents. However, cell toxicity studies revealed that 1 is more phototoxic than all other complexes studied and that cyclometalled complexes still may be promising.
candidates for PCT. Further studies that confirm the cytotoxic mechanism of 1 will aid in the further development of new PCT agents.

Interestingly, complex 4 undergoes photodissociation of a bidentate ligand, but the dpb ligand is released with red light rather than the biq ligand as observed in 2 and 3. Complex 4 possesses three bulky ligands (2 biq and 1 dpb) that cause steric strain of the octahedral geometry about the Ru(II) center rather than just two bulky biq ligands as in 2 and 3. Absorption, emission, electrochemistry, transient absorption, and calculations confirm the lowest energy excited state in 4 is localized on the dpb ligand rather than the biq ligands as in 2 and 3. This may play a key role in the photoinduced ligand dissociation of the dpb ligand because crystallographic data displays similar geometric distortions in 4 relative to 1 and 3. Also, 4 displays a covalent binding interaction with DNA upon irradiation, but not in the dark, and an enhanced toxicity towards HeLa cells upon irradiation indicating it can potentially function as a useful PCT agent. The open bidentate coordination site on the dpb ligand in 4 can be utilized for bimetallic systems that could possibly operate further in the PCT window.

References


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Chapter 6 : Isomerization Initiated by Photoinduced Ligand Dissociation in Ru(II) Complexes with the Ligand 2-p-tolylpyridinecarboxaldimine


6.1 Introduction

Photoinduced ligand dissociation in transition metal complexes is a phenomenon that can be controlled with visible light.\textsuperscript{1-12} This process, together with a more detailed understanding of the rich excited state dynamics and reactivity of ruthenium(II) complexes, is of interest due to their potential applications in medicine, in which controlled ligand dissociation can be used as a vehicle for drug delivery, as well as their widespread use for solar energy conversion schemes and as molecular switches.\textsuperscript{13-19} Recently, the ruthenium(II) complexes used as potential therapeutics triggered by light has deviated from traditional bidentate ligands, such as bpy (2,2'-bipyridine) and phen (phen = 1,10-phenanthroline) in favor of bulkier polypyridyl diimine ligands such as 6,6’-dimethyl-bpy or 2,2’-biquinoline (biq). The use of these and other sterically demanding ligand leads to distortions of the octahedral geometry around the metal and result in unusual photochemical properties, such as bidentate ligand photoejection and
selective ligand substitution upon irradiation. These concepts were thoroughly investigated and discussed in Chapters 3 – 5.

The less traditional bidentate ligands 2-<em>p</em>-tolylpyridinecarboxaldimine (PTPI) and 2-(phenylazo)pyridine (PAP), the latter of which contains an azo moiety that is known to undergo photoisomerization, have not been incorporated into Ru(II) photoinduced ligand exchange systems. The structures of PTPI and PAP are shown in Figure 6.1a. Photoisomerization of a bidentate ligand coordinated to a Ru(II) metal center is a rare occurrence. When Ru(II) photoisomerization does take place, the chemical behavior of the newly formed species can be drastically different than that of the precursor. An example of such behavior can be found in [Ru(tpy)(PAD)(OH<sub>2</sub>)]<sup>+</sup> (tpy = 2,2′;6′,2″-terpyridine, PAD = 2-(pyrid-2′-yl)acridine), wherein the photoisomerization of the bidentate PAD ligand results in a complex that electrocatalytically evolves O<sub>2</sub> gas less efficiently than the original isomer.<sup>24,25</sup>

In the present work, the preparation, characterization, and photochemistry of newly isolated isomers of a Ru(II) complex possessing two PTPI ligands and two photolabile monodentate CH<sub>3</sub>CN ligands, [Ru(PTPI)<sub>2</sub>(CH<sub>3</sub>CN)<sub>2</sub>]<sup>2+</sup> are reported. Owing to the asymmetry of PTPI, there are three possible <i>cis</i>-isomers (α, β, and ε) and two <i>trans</i>-isomers (γ and δ); the five structures are shown in Figure 6.1b. It should be noted that the naming scheme is based on previous work and that in Figure 6.1, N<sub>p</sub> and N<sub>i</sub> refer to the pyridine and imine nitrogen atoms, respectively.<sup>26</sup> In addition, the two related complexes <i>cis</i>-[Ru(bpy)(PTPI)(CH<sub>3</sub>CN)<sub>2</sub>]<sup>2+</sup> (1) and <i>cis</i>- [Ru(bpy)(PAP)(CH<sub>3</sub>CN)<sub>2</sub>]<sup>2+</sup> (2) were synthesized to gain a deeper understanding of the photochemistry observed for the
[Ru(PTPI)\(_2\)(CH\(_3\)CN)\(_2\)]^{2+}\) isomers and to compare the role of the PTPI and PAP ligands. Four of the five [Ru(PTPI)\(_2\)(CH\(_3\)CN)\(_2\)]^{2+}\) isomers, \(\alpha\), \(\beta\), \(\delta\), and \(\gamma\), were synthesized and structurally characterized, along with 1 and 2. The remaining isomer, \(\varepsilon\), was isolated from photolysis reactions, and its identity verified by its \(^{1}H\) NMR spectrum and crystal structure. The photophysical and photochemical properties of the complexes were investigated and found to involve ligand dissociation of the monodentate CH\(_3\)CN ligands in the compounds with the PTPI ligand but not for the PAP complex 2. In addition, the PTPI complexes display a novel ligand isomerization that accompanies ligand dissociation. Interestingly, the photolysis of each pure \(\alpha\)-, \(\beta\)-, \(\delta\)-, and \(\gamma\)-[Ru(PTPI)\(_2\)(CH\(_3\)CN)\(_2\)]^{2+}\) isomer results in the same mixture of products. Calculations were employed to aid in the understanding of the observed photochemistry.
Figure 6.1. (a) Structures of the PTPI and PAP ligands and (b) labeling scheme for the various isomers of Ru(PTPI)$_2$Cl$_2$ and [Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$, where L = Cl$^-$, CH$_3$CN.

### 6.2 Results and Discussion

#### 6.2.1 Synthesis and Characterization

The synthesis of cis- and trans-Ru(PTPI)Cl$_2$ reported by Goswami and coworkers involved the use of column chromatography to separate the isomers from the reaction mixture. In that work, only the δ isomer and one of the other two cis species, either the α or ε isomer, were characterized.$^9$ Unfortunately, we were unable to replicate their separation methods. In our hands, a green solid corresponding to the trans isomers precipitated from the reaction mixture, and a blue solid that was collected from the
reaction filtrate which was believed to contain the cis isomers based on the work by Goswami and coworkers.

An orange solid was isolated following the thermal exchange of chloride ligands of the green precipitate with CH$_3$CN, and the $^1$H NMR spectrum of the product revealed two distinct species with resonances associated with the proton on the imine carbon atom at 9.08 ppm and 9.28 ppm, which correspond to the $\gamma$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ and $\delta$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ complexes, respectively (Figure 6.1). The spectral assignments for the two isomers were possible by comparing the chemical shifts of the proton on the carbon of the pyridine ring adjacent (ortho) to N$_p$ in the PTPI ligand, which were characterized extensively in the corresponding PAP complexes using 2D $^1$H NMR spectroscopy. These specific protons correspond to the doublet at 9.71 ppm in $\gamma$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ and at 7.65 ppm in $\delta$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$. In the $\delta$ isomer, this proton is shifted upfield relative to the $\gamma$ isomer because it is pointing towards the phenyl ring on the other PTPI ligand, resulting in shielding provided by the ring current. This shielding does not occur in the $\gamma$ isomer because the ligands are positioned in a coplanar orientation, as expected from previous literature and confirmed crystallographically.

In order to ensure that these compounds were indeed the correct isomers, the mixture was subjected to slow vapor diffusion as described previously to obtain single crystals for X-ray crystallography. This method resulted in the growth of two clearly distinct types of crystals, one amber and the other red in color. The two sets of crystals were separated by hand under the microscope and the resulting structures are shown in
Figure 6. It is evident that the amber crystals correspond to the $\gamma$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ isomer whereas the red crystals are the $\delta$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ isomer. The ratio of the $\gamma$ to $\delta$ isomers in the synthetic mixture was found to be 28% and 72%, respectively by integration of the $^1$H NMR spectrum.

Similarly, the blue portion of the reaction mixture, corresponding to the cis-Ru(PTPI)$_2$Cl$_2$ isomers, was refluxed in CH$_3$CN:H$_2$O. Following the exchange of the chloride ions with CH$_3$CN, two distinct isomers were observed by $^1$H NMR spectroscopy. The number of resonances in the $^1$H NMR spectrum of the mixture in (CD$_3$)$_2$CO corresponds to an asymmetric complex, the $\beta$ isomer, and one symmetric complex, which is consistent with either the $\alpha$ or $\varepsilon$ isomer. Single crystals were grown from the mixture and the resulting structure reveals the presence of only the $\alpha$ isomer (Figure 6.2) in addition to $\beta$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$. Therefore, of the three possible cis-isomers, only the $\alpha$ and $\beta$ complexes were formed by this method in a ~60% to ~40% ratio, respectively as determined from NMR spectroscopy prior to crystallization.

It is evident from the structural characterization of the [Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ products resulting from the reaction of the chloride complexes with CH$_3$CN, that at least the $\delta$-, $\gamma$-, $\alpha$-, and $\beta$-Ru(PTPI)$_2$Cl$_2$ isomers were present as precursors. This finding is in disagreement with previous work, but instead parallels the findings established for Ru(II) complexes of the related PAP ligand. Crystals suitable for X-ray diffraction were not obtained for the $\beta$ isomer, but after the slow vapor diffusion process to isolate single crystals of $\alpha$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$, an oil in the vial wall remained assigned to the $\beta$ isomer with $^1$H NMR resonances in (CD$_3$)$_2$CO (chemical shifts reported in ppm) at 9.71
Following purification and single crystal growth, only small yields of each isomer were obtained, however. The mixture of cis-isomers resulted in a 52% yield, whereas that of the trans complexes corresponded to a 44% yield.

**Figure 6.2.** Thermal ellipsoid plots of the [PF$_6$]$^-$ salts of the isolated isomers and complexes studied (ellipsoids drawn at the 50% probability level, the [PF$_6$]$^-$ anions and the H atoms were removed for clarity).
It should be noted that we were only able to generate the $\varepsilon$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ isomer by photolysis in CH$_3$CN. When pure $\alpha$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ is irradiated for 120 min in CD$_3$CN ($\lambda_{\text{irr}} \geq 395$ nm), the disappearance of the singlet at 8.58 ppm in CD$_3$CN is observed, corresponding to the proton on the imine carbon of the reactant, with concomitant appearance of new resonances corresponding to two different products. One product is $\beta$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$, with resonances at 8.59 and 8.60 ppm in CD$_3$CN (~9.71 ppm in (CD$_3$)$_2$CO), and the other species is characterized by a singlet at 9.02 ppm in CD$_3$CN (9.45 ppm in (CD$_3$)$_2$CO). The X-ray crystallographic determination of the latter unknown product revealed it to be the $\varepsilon$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ isomer (Figure 6.2). After photolysis in CH$_3$CN for 3-4 h, the mixture contains 63% and 37% of the $\varepsilon$- and $\beta$-isomers, respectively, which will be further discussed in the photochemistry section.

When an equimolar mixture of the PTPI ligand and Ru(bpy)(DMSO)$_2$Cl$_2$ are refluxed in CH$_3$CN:H$_2$O, the cation [Ru(bpy)(PTPI)(CH$_3$CN)$_2$]$^{2+}$ (1) is formed. The $^1$H NMR spectrum of the product is consistent with the presence of only one isomer. The crystal structure of 1 shown in Fig. 2 reveals that this isomer has the pyridine moiety of PTPI positioned $\text{trans}$ to the bpy ligand and the imine moiety $\text{trans}$ to a CH$_3$CN ligand. The analogous structural isomer is observed for [Ru(bpy)(PAP)(CH$_3$CN)$_2$]$^{2+}$ (2) and agrees with previous reports (Figure 6.2).
6.2.2 Electronic Absorption and Emission

The singlet Ru→PTPI metal-to-ligand charge transfer ($^1$MLCT) maxima for trans isomers, δ- and γ-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ are observed at 471 nm in CH$_3$CN, with $\varepsilon$ = 10,800 M$^{-1}$ cm$^{-1}$ and 10,300 M$^{-1}$ cm$^{-1}$, respectively, and a shoulder at ~440 nm (Figure 6.3a). In addition, broad features attributed to the PTPI $^1\pi\pi^*$ transitions are observed in the 300-350 nm range for both isomers. A broad absorption is also observed between 350 and 400 nm in γ-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ in CH$_3$CN (Figure 6.3a).

![Figure 6.3](image)

**Figure 6.3.** Electronic absorption spectra of (a) α, ε, δ, and γ-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ and (b) 1 and 2 in CH$_3$CN.

The $^1$MLCT transition is slightly blue-shifted for the α- and ε-isomers, centered at 465 nm (10,900 M$^{-1}$cm$^{-1}$) and 461 nm (6,500 M$^{-1}$cm$^{-1}$), respectively, with a shoulder at
~410 nm (Figure 6.3a). In addition, their spectral profile in the 300-370 nm range is similar to that of δ-[Ru(PTPI)₂(CH₃CN)₂]²⁺, consistent with the PTPI ¹ππ* nature of this broad feature.

![Figure 6.4](image)

**Figure 6.4.** (a) Absorption, excitation, and emission spectra of 1 in CH₃CN at room temperature and (b) emission spectrum of 2 at 77 K.

For 1, the Ru→PTPI ¹MLCT maximum in CH₃CN is observed at 455 nm (8,100 M⁻¹ cm⁻¹) with a shoulder at 397 nm (Figure 6.3b). The shape of the ¹MLCT band in 1 is similar to that of the α-[Ru(PTPI)₂(CH₃CN)₂]²⁺. This similarity is expected since this isomer most resembles 1 in structure, whereas in the former case, the two pyridine units of the PTPI ligands are positioned *trans* to each other and the imine nitrogen atom is...
trans to one CH$_3$CN ligand. This arrangement is also apparent in 1 in which the pyridine unit of the PTPI ligand is positioned trans to the bpy ligand. The maximum of the Ru→PAP $^1$MLCT absorption band in 2 is red-shifted by 1486 cm$^{-1}$ relative to 1, observed at 488 nm (7,800 M$^{-1}$cm$^{-1}$), with no apparent shoulder (Figure 6.3b).

The $^1$MLCT absorption bands in the cis isomers are broadened as compared to those of the trans isomers, which are relatively sharp. Finally, the emission maxima for 1 at room temperature and for 2 at 77 K in CH$_3$CN are observed at 711 and 861 nm, respectively (Figure 6.4). The [Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ isomers are very weakly emissive at room temperature.

### 6.2.3 Photochemistry

The photoreactivities of the [Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ isomers, as well as that of 1 and 2, were evaluated by monitoring changes to the electronic absorption and $^1$H NMR spectra as a function of irradiation time. The changes to the electronic absorption spectrum of α-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ upon irradiation in water are shown in Figure 6.5a ($\lambda_{\text{irr}} \geq 495$ nm). It is evident from the data in Figure 6.4a that the absorption of the starting material decreases with concomitant growth of a peak at 488 nm that corresponds to the mono-aqua intermediate, [Ru(PTPI)$_2$(CH$_3$CN)(H$_2$O)]$^{2+}$, resulting in an isosbestic point at 475 nm between the irradiation times, $t_{\text{irr}}$, of 0 min to 30 min. Further irradiation results in a decrease of the absorption of the mono-aqua intermediate and the increase in intensity of a new peak at 515 nm with an isosbestic point at 503 nm, attributed to the bis-aqua product, [Ru(PTPI)(H$_2$O)$_2$]$^{2+}$ (Figure 6.5b). This shift in energy of
approximately 2,000 cm\(^{-1}\) between the bis-CH\(_3\)CN and bis-H\(_2\)O complexes is similar to other Ru(II) CH\(_3\)CN ligand exchange systems in water. It should be noted that no changes in the absorption spectrum are observed when \(\alpha\)-[Ru(PTPI)\(_2\)(CH\(_3\)CN)\(_2\)]\(^{2+}\) is stored in the dark in H\(_2\)O for 24 hours.\(^{22}\)

\[\text{Figure 6.5.} \text{ Electronic absorption spectral changes of } \alpha\text{-}[\text{Ru(PTPI)}\(_2\)(\text{CH}_3\text{CN})\(_2\)]^{2+} \text{ as a function of irradiation time in H}_2\text{O (10 } \mu\text{M) at (a) } t_{\text{irr}} = 0, 1, 2, 3, 5, 7, 10, 15, \text{ and } 30 \text{ min, (b) } t_{\text{irr}} = 30, 45, 60, 90, 120, 150, 180, 240, \text{ and } 300 \text{ min, and (c) in CH}_3\text{CN (30 } \mu\text{M) } t_{\text{irr}} = 0, 1, 2, 5, 10, 20, 30, 60, 120, 180, \text{ and } 300 \text{ min (} \lambda_{\text{irr}} \geq 495 \text{ nm).}\]
The analogous photolysis experiment was performed in CH$_3$CN and is shown in Figure 6.5c. Although the bound CH$_3$CN ligands are expected to exchange with solvent CH$_3$CN molecules upon irradiation, no net change in absorption is expected. As can been seen in Figure 6.5c, however, the initial absorption decreases and the growth of a more intense shoulder at 412 nm with an isosbestic point at 444 nm is observed upon irradiation for 5 hrs ($\lambda_{\text{irr}} \geq 495$ nm). Only minor spectral changes are observed when the complex is stored in the dark for 12 hrs. This result points to a photochemical reaction other than photoinduced ligand exchange. These same trends are also observed upon irradiation of the $\delta$- and $\gamma$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ isomers in water and CH$_3$CN (Figure 6.6), both of which are stable in the dark.

![Figure 6.6](https://example.com/figure6.6.png)

**Figure 6.6.** Changes to the electronic absorption spectrum of (a) $\delta$ and (b) $\gamma$ in CH$_3$CN upon irradiation ($\lambda_{\text{irr}} \geq 495$ nm).

In order to gain structural information, the photolysis of $\alpha$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ was carried out in CD$_3$CN and followed by $^1$H NMR spectroscopy ($\lambda_{\text{irr}} \geq 495$ nm, Figure
In this solvent, only a decrease in the resonance associated with bound CH$_3$CN at 2.45 ppm is expected if photoinduced ligand exchange is the sole process, together with the appearance of the free CH$_3$CN peak at 1.96 ppm. Although this ligand exchange does occur, the resonances corresponding to the methyl group and aromatic protons on the PTPI ligand also decrease with irradiation time, with concomitant appearance of new resonances in the aromatic region. The resonances corresponding to the $\alpha$ complex disappear with continued irradiation, and the final spectrum is identical to that of the products obtained from the photolysis of $\alpha$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ in CH$_3$CN, dried, and re-dissolved in (CD)$_3$CO (Figure 6.8b) or CD$_3$CN. For comparison, Figures 6.8a and 6.8c show the $^1$H NMR spectra of the pure $\alpha$- and $\varepsilon$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$, respectively, and the spectrum in Figure 6.8b shows that there are two different products present following irradiation of the $\alpha$ isomers for 3 hrs ($\lambda_{\text{irr}} \geq 495$ nm). Single crystal X-ray diffraction experiments confirmed that one of the photoproducts is $\varepsilon$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$, and the $^1$H NMR spectrum of the isolated $\varepsilon$ isomer is shown in Figure 6.8c. The resonances in the remaining product are indicative of the asymmetric $\beta$ isomer (Figure 6.8b). The relative amounts of the $\varepsilon$ and $\beta$ isomers after photolysis in CH$_3$CN were 63% and 37%, respectively.
Figure 6.7. Photolysis of α monitored by $^1$H NMR spectroscopy in CD$_3$CN $\lambda_{\text{irr}} \geq 495$ nm as a function of irradiation time. The singlet at 7.35 ppm corresponds to the benzene internal standard, the resonance at 2.45 ppm corresponds to bound CH$_3$CN, and the resonance growing in at 1.96 ppm corresponds to free CH$_3$CN.
A point of interest is that irradiation of pure solutions of each of the other isomers, 
$$\delta\gamma$$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ in CH$_3$CN, dried, then re-dissolved in (CD$_3$)$_2$CO and monitored by $^1$H NMR spectroscopy result in the same product distribution of the $\varepsilon$ (63%) and $\beta$ (37%) isomers. No additional changes in the $^1$H NMR spectrum are observed when the product mixture is irradiated further or kept in the dark for 3 hrs. In addition, the solutions containing each isomer are stable in the dark in CD$_3$CN for up to 3 h, indicating that the isomers do not interconvert in the dark at room temperature. Furthermore, irradiation of pure $\varepsilon$ in CD$_3$CN results in spectral changes consistent with formation of the $\beta$ isomer with a final ratio that corresponds to 63% $\varepsilon$- and 37% $\beta$-isomer after extensive irradiation. These results are consistent with the presence of a single high
energy photogenerated intermediate that is common to all the [Ru(PTPI)\(_2\)(CH\(_3\)CN)\(_2\)]\(^{2+}\) isomers, \textit{cis} and \textit{trans}, which is able to generate the same two products in identical ratios.

To gain additional understanding of the excited state processes, the photochemistry of [Ru(bpy)(PTPI)\(_2\)]\(^{2+}\) (3), which lacks CH\(_3\)CN ligands, was investigated. The irradiation of 3 in CH\(_3\)CN with \(\lambda_{irr} \geq 420\) nm for 60 min does not result in changes in the electronic absorption spectrum. Similar inactivity was observed for the mixture of the \textit{cis}-Ru(PTPI)\(_2\)Cl\(_2\) isomers, the precursor to the \textit{cis}-[Ru(PTPI)\(_2\)(CH\(_3\)CN)\(_2\)]\(^{2+}\) isomers, when photolyzed in CH\(_3\)CN (\(\lambda_{irr} 420\) nm, \(t_{irr} = 60\) min). On the basis of these results, it is concluded that the coordinated PTPI ligand, in the absence of photolabile ligands, does not photoisomerize when irradiated with visible light. Therefore, the PTPI isomerization observed for the \(\alpha\)-, \(\delta\)-, \(\varepsilon\)-, and \(\gamma\)-[Ru(PTPI)\(_2\)(CH\(_3\)CN)\(_2\)]\(^{2+}\) complexes must be preceded by photoinduced CH\(_3\)CN ligand dissociation. The inertness of Ru(PTPI)\(_2\)Cl\(_2\) to photochemical transformation, together with its thermal reactivity with CH\(_3\)CN, also indicates that there is only one pathway to generate the \(\varepsilon\)-[Ru(PTPI)\(_2\)(CH\(_3\)CN)\(_2\)]\(^{2+}\) isomer, the photolysis of each of the other isomers of the complex. A summary of these findings are depicted schematically in Figure 6.9a.
Figure 6.9. (a) Photochemical scheme showing the formation of the $\beta$- (37%) and $\varepsilon$- $[\text{Ru(PTPI)}_2(\text{CH}_3\text{CN})_2]^{2+}$ (63%) from the irradiation of each pure isomer in CH$_3$CN and (b) the calculated relative energies of the cis-$\text{Ru(PTPI)}_2\text{Cl}_2$ and cis-$[\text{Ru(PTPI)}_2(\text{CH}_3\text{CN})_2]^{2+}$ isomers (the calculated energies correspond to those of the model complexes where CH$_3$CN was replaced by HCN).

Complex 1 also possesses photolabile CH$_3$CN ligands and a red shift is observed upon irradiation with $\lambda_{\text{irr}} \geq 420$ nm in H$_2$O (Figure 6.10) but it remains stable in the dark. Also, the photolysis monitored by $^1$H NMR in CD$_3$CN reveals that one of the CH$_3$CN ligands is more photolabile than the other (Figure 6.11), which was also previously reported for the asymmetric complex $[\text{Ru(biq)(phen)(CH}_3\text{CN)}]^{2+}$. Moreover, the growth
of new aromatic resonances indicate that 1 also undergoes isomerization along with ligand exchange. Extensive photolysis of 1 (18 hours) does not complete the isomerization process and efforts were not undertaken to characterize the photoproduct. 

A downfield shift of the imine singlet of the PTPI ligand is observed from 9.05 ppm in 1, in which the imine nitrogen atom is trans to the CH$_3$CN ligand, to 9.42 ppm in the product. This shift is consistent with isomerization, such that a pyridine unit from the bpy ligand is positioned trans to the imine nitrogen atom in the product, based on the imine proton in the γ-isomer at 9.26 ppm. Such isomerization is similar to that observed in the photochemistry of α-, δ-, and γ-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$. Moreover, these results are consistent with the requirement of CH$_3$CN ligand dissociation prior to PTPI isomerization. Complex 2 possesses the related PAP ligand, which is known to undergo photoisomerization in solution afforded by the presence of the azo moiety. However, the Ru(II) complex does not display any photoinduced ligand exchange in H$_2$O ($\lambda_{irr} \geq 345$ nm) and very little exchange in CD$_3$CN even when irradiated with light in the near-ultraviolet range. Moreover, there is also no evidence in the $^1$H NMR spectrum of photoisomerization of 2 in CD$_3$CN ($\lambda_{irr} \geq 345$ nm).
**Figure 6.10.** Changes to the electronic absorption spectrum of 1 in H₂O upon irradiation ($\lambda_{\text{irr}} \geq 420$ nm).

![Absorption Spectrum](image)

**Figure 6.11.** $^1$H NMR spectra in (CD$_3$)$_2$CO of the aromatic region of a) 1 and b) 1 after photolysis in CH$_3$CN ($\lambda_{\text{irr}} \geq 420$, 15 hours).

![NMR Spectra](image)

It is generally accepted that photoinduced ligand dissociation in Ru(II) complexes with low-lying $^3$MLCT states is a result of thermal population of low lying $^3$LF (ligand field) dd states from the lowest energy $^3$MLCT excited state.$^{4-7}$ Although the $^1$MLCT
absorption maximum is only red-shifted by 1486 cm\(^{-1}\) in 2 relative to 1, the emission data indicate that the \(^3\)MLCT energy is approximately 2451 cm\(^{-1}\) lower in energy in the former case as compared to the latter. The resulting stokes shifts for the complexes are 7913 cm\(^{-1}\) and 8878 cm\(^{-1}\) for 1 and 2, respectively. Assuming that the energy of the \(^3\)dd states in 1 and 2 are similar, the \(^3\)MLCT-\(^3\)LF gap must be too large for thermal population of the dissociative \(^3\)LF from the \(^3\)MLCT state in 2, such that CH\(_3\)CN lability is not possible. Alternatively, the lower energy emission in 2 may be due to significant distortion in its \(^3\)MLCT state that is not present in 1. Such distortion may increase the activation barrier from the \(^3\)MLCT to the \(^3\)LF state. Although the reason for the lack of ligand dissociation in 2 remains to be fully understood, since CH\(_3\)CN dissociation through population of the \(^3\)LF state(s) is required prior to isomerization, the latter is not observed in 2.

6.2.4 Calculations

The relative energies of optimized geometric structures of \(\alpha\)-, \(\beta\)-, and \(\epsilon\)-[Ru(PTPI)\(_2\)(CH\(_3\)CN)\(_2\)]\(^{2+}\) were calculated using Density Functional Theory (DFT) methods and compared to those of the corresponding chloride precursors, \(\alpha\)-, \(\beta\)-, and \(\epsilon\)-Ru(PTPI)\(_2\)Cl\(_2\). For the chloride complexes, \(\alpha\)-Ru(PTPI)\(_2\)Cl\(_2\) isomer was calculated to be the most stable, followed by \(\beta\)-Ru(PTPI)\(_2\)Cl\(_2\) (+6.42 kJ/mol) and then \(\epsilon\)-Ru(PTPI)\(_2\)Cl\(_2\) (+28.41 kJ/mol), as shown in Figure 6.9b. The difference in energy among the isomers explains the observation of only the \(\alpha\)- and \(\beta\)-Ru(PTPI)\(_2\)Cl\(_2\) precursors in the initial cis-Ru(PTPI)\(_2\)Cl\(_2\) mixture. In addition, since these are the only two cis isomers present as
starting materials, their reaction with CH₃CN also results in the same two isomers, α- and β-[Ru(PTPI)₂(CH₃CN)₂]²⁺. The fact that the product mixture contains more of the α than the β isomer is consistent with the calculated relative stabilities of each bis-chloro precursor.

In order to simplify the calculations for the CH₃CN complexes, the related [Ru(PTPI)₂(NCH)₂]²⁺ model compounds were used. For these complexes, the opposite trend in the energies of the three isomers was observed from the calculations (Figure 6.9b), where the ε isomer is the most stable, followed by the β isomer (+0.866 kJ/mol). The energy of the α isomer is predicted to be at 9.27 kJ/mol above than that of ε-[Ru(PTPI)₂(NCH)₂]²⁺. This pattern explains the isomerization that occurs upon irradiation, resulting in the two most stable isomers. As shown in Figure 6.9a, ligand dissociation of one bound CH₃CN molecule allows for the complex to rearrange to an intermediate that goes on to form β- and ε-[Ru(PTPI)₂(CH₃CN)₂]²⁺ upon solvent coordination in a ratio of 37% and 63%, respectively, in CH₃CN. The experimental product ratio is remarkably close to that expected from the Boltzmann distribution at room temperature, 41% β- and 59% ε-isomer.

6.3 Conclusions

The α-, ε-, δ-, and γ- isomers of [Ru(PTPI)₂(CH₃CN)₂]²⁺ were isolated and characterized by ¹H NMR spectroscopy in solution and X-ray crystallography and shown to undergo photoinduced ligand exchange in H₂O and CD₃CN. When each pure isomer is irradiated in CH₃CN the result is the same product mixture containing 63% ε-
[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$ and 37% $\beta$-[Ru(PTPI)$_2$(CH$_3$CN)$_2$]$^{2+}$. It was also demonstrated that photoisomerization requires ligand dissociation, therefore, the product distribution can be explained by a common intermediate which generates the two products upon CH$_3$CN solvent coordination in a ratio dictated by the calculated relative stability of each isomer.

The asymmetric complex 1 with the PTPI ligand also undergoes photoinduced ligand exchange in H$_2$O and CD$_3$CN. Evidence of photoisomerization is present when 1 is irradiated in CH$_3$CN. In contrast, no evidence of photoinduced ligand exchange or photoisomerization is observed for the related PAP containing complex 2. The difference in reactivity is explained by the lower energy of the $^3$MLCT state in 2, such that it is not able to thermally populate the dissociative $^3$LF state(s). Ongoing work includes the preparation of different derivatives of the PTPI ligand to selectively stabilize one isomeric form, as well as investigation of the mechanism for photoinduced ligand exchange and isomerization phenomena. The work presented herein may be useful in the design of new complexes for solar energy conversion or photoswitching applications and that of new bidentate ligands in photoinduced ligand exchange systems results in interesting properties as was observed with sterically demanding ligands in previous chapters.

References


Chapter 7: Bimetallic Ru(II) Complexes for Photochemotherapy

7.1 Introduction

Current cancer treatment using cisplatin relies on thermal activation of the complex to generate the active species that results in DNA damage and ultimately cell death.\(^1\) The drawbacks of this thermally activated mechanism is poor selectivity and increased drug resistance.\(^1,2\) One approach to circumvent these deficiencies is through a process known as photochemotherapy (PCT). The principle of PCT is to use light to excite a molecule such that it becomes activated or generates a reactive species that ultimately causes cell death, but remains nontoxic in the dark. PCT provides low systemic toxicity, low levels of invasiveness, which in some cases makes it superior to conventional therapies such as cisplatin.\(^3-7\) Ruthenium(II) complexes capable of undergoing photoinduced ligand substitution reactions are of particular interest for PCT because they are able to covalently bind to DNA and other biomolecules and/or release bioactive molecules following excitation.\(^3-5\) Moreover, Ru(II) polypyridyl complexes provide a robust scaffold to design new molecules for facile tuning of the excited state properties.

It is broadly accepted that photoinduced ligand substitution occurs via population of low lying \(^3\)dd states, which can be thermally accessed from \(^3\)MLCT (metal-to-ligand charge transfer) excited states.\(^8-11\) Population of this state in Ru(II) polypyridyl
complexes results in placing electron density on e\textsubscript{g}–type orbitals with Ru – (\sigma\ast) character causing ligand dissociation.\textsuperscript{8-11}

Previous chapters focused on the photoinduced ligand dissociation of monodentate CH\textsubscript{3}CN ligands as well as the photoejection of sterically demanding diimine ligands in new Ru(II) complexes in an effort to gain a further understanding of the ligand dissociation process and to design complexes that are photoactive with lower energy light inside the therapeutic window (600-850 nm)\textsuperscript{6,7,12} for applications in PCT. Another strategy to create molecules that could function as superior PCT agents are bimetallic Ru(II) complexes capable of undergoing ligand exchange reactions at multiple metal centers upon excitation. Addition of another metal typically results in a shift of the absorption of the lowest energy excited state and increase molar absorptivity.\textsuperscript{13} Furthermore, complexes that have two sites that are photoactive could potentially form a stronger covalent binding interaction with DNA as well as release multiple drugs upon irradiation. To this end, the bimetallic Ru(II) complexes \{[Ru(CH\textsubscript{3}CN)\textsubscript{3}](tppz)}\textsuperscript{4+} (1, tppz = tetra-2-pyridylpyrazine), [Ru(bpy)(CH\textsubscript{3}CN)\textsubscript{2}](pbp)}\textsuperscript{4+} (2, bpy = 2,2’-bipyridine, pbp = 1,4-bis(2-pyridylmethyleneamino)benzene), [[Ru(phen)(CH\textsubscript{3}CN)\textsubscript{2}](pbp)]\textsuperscript{4+}, (3, phen = 1,10-phenanthroline), and \{[Ru(tpy)(CH\textsubscript{3}CN)]\textsubscript{2}(bpm)}\textsuperscript{4+} (4, tpy = 2,2’,6’,2”-terpyridine, bpm = 2,2’-bipyrimidine) were synthesized, which possess 6, 4, and 2 CH\textsubscript{3}CN ligands, respectively, that have the potential to be photolabile and the bridging ligands are shown in Figure 7.1. Complex 1 undergoes photoinduced ligand exchange of the 4 axial CH\textsubscript{3}CN ligands when irradiated with red light but does not display a covalent binding interaction with linearized DNA, photodissociation of all 4 CH\textsubscript{3}CN ligands
occurs in 2 and 3 and 2 covalently binds to linearized DNA, and no ligand exchange is observed in complex 4. Coordination of bioactive molecules such as 5-CNU and cysteine protease inhibitors possessing nitrile groups is currently in progress, which could result in enhanced cytotoxicity relative to previously reported complexes utilizing these bioactive molecules. 14–16

![Molecular structures of the relevant bridging ligands.](image)

**Figure 7.1.** Molecular structures of the relevant bridging ligands.

### 7.2 \{[Ru(CH₃CN)₃]₂(tppz)]PF₆\}_₄

#### 7.2.1 Photophysical Properties

The \(^1\)MLCT absorption maximum for 1 in CH₃CN is observed at 495 nm (\(\varepsilon = 18\) 000 M\(^{-1}\)cm\(^{-1}\)) with a broad tail that absorbs to 650 nm attributed to Ru(t\(_{2g}\)) → tppz(\(\pi^*\)) transitions and is similar to the related complex \{[Ru(tpy)]₂(tppz)]PF₆\}_₄ (tpy = 2,2’;6’,2”-terpyridine), which has a \(^1\)MLCT absorption maximum at 548 nm (\(\varepsilon = 36\)
Complex 1 also possesses intense peaks at 381 nm and 362 nm assigned as $1\pi\pi^*$ transitions and is emissive at room temperature and 77 K in CH$_3$CN. The $\lambda_{\text{max}}$ of emission in 1 is observed at 745 nm and 722 nm at room temperature and 77 K, respectively, with vibrational progressions of $\sim$1350 cm$^{-1}$ typical of Ru(II) polypyridyl complexes, and assigned as a Ru(t$_{2g}$) $\rightarrow$ tppz(π*) $^3$MLCT lowest energy excited state. The electronic absorption, emission at 77 K, and excitation spectra of 1 are shown in Figure 7.2. Since the excitation spectra when monitoring the emission at 725 nm and 800 nm matches the absorption spectrum, the emission results from excitation of 1 and not some impurity in the sample.

![Absorbance, Emission, Excitation](image)

**Figure 7.2.** Electronic absorption in CH$_3$CN, excitation spectra in CH$_3$CN at 77 K, and emission spectrum in CH$_3$CN at 77 K of complex 1.

The transient absorption spectra of 1 in CH$_3$CN on the ultrafast timescale ($\lambda_{\text{ex}} = 325$ nm, fwhm = 300 nm) is shown in Figure 7.3 (top). Typical ground state bleaching is
observed at 380 nm and spanning the wavelength range of 475 nm to 550 nm and a positive transient absorption feature is observed in the range of 390 nm to 475 nm tentatively assigned as reduced tppz in the lowest energy excited state, Ru(t$_{2g}$) $\rightarrow$ tppz($\pi^*$) $^3$MLCT. This feature associated with the $^3$MLCT state vibrationally cools after approximately 10 ps, which has a relatively long lived lifetime with $\tau = 570$ ns that was measured using nanosecond transient absorption ($\lambda_{ex} = 532$ nm, fwhm = 8 ns) in CH$_3$CN shown in Figure 7.3 (bottom). This lifetime is similar to that of [Ru(bpy)$_3$]$^{2+}$ under analogous conditions, and is much longer than [Ru(bpy)$_2$(CH$_3$CN)$_2$]$^{2+}$, which has a lifetime of 51 ps in CH$_3$CN and undergoes efficient photoinduced ligand dissociation. Similar transient absorption features are observed in {[Ru(tpy)$_2$(tppz)]$\text{PF}_6$)$_4$, under similar conditions, but with a much shorter lifetime of 82 ns. This is due the “bite” angles of the tpy ligands that result in geometric distortions about the metal center lowering the energy of deactivating $^3$dd states, which is typical of complexes possessing the tpy ligand.22
Figure 7.3. Ultrafast transient absorption spectrum in CH$_3$CN of 1 (top, $\lambda_{ex} = 325$ nm, fwhm = 300 fs) and nanosecond transient absorption in CH$_3$CN recorded 0.04 $\mu$s after the laser pulse (bottom, $\lambda_{ex} = 532$ nm, fwhm = 8 ns).

7.2.2 Photochemistry

The spectral changes in CD$_3$CN as a function of irradiation time monitored by $^1$H NMR spectroscopy of 1 are shown in Figure 7.4. Upon irradiation ($\lambda_{irr} \geq 435$ nm) the singlet at 2.03 ppm corresponding to the protons of the axial CH$_3$CN ligands on both metal centers decreases with the concomitant growth of a singlet at 1.96 ppm that corresponds to unbound CH$_3$CN. Integrations of 2 and 1 were set relative to a benzene internal standard and are listed next to the corresponding resonance in Figure 7.4. Extensive irradiation results in the resonance at 2.03 ppm to retrieve to baseline and the final integration of free CH$_3$CN is indicative of all axial CH$_3$CN ligands exchanging for deuterated solvent. No change is observed in the singlet at 2.88 ppm corresponding to
both equatorial CH\textsubscript{3}CN ligands revealing that only the axial CH\textsubscript{3}CN ligands are photolabile and the final product is \{[Ru(CD\textsubscript{3}CN)\textsubscript{2}(CH\textsubscript{3}CN)]\textsubscript{2}(tppz)}\textsuperscript{4+}. No changes are observed when the complex is kept in the dark in CD\textsubscript{3}CN over the same time period. Irradiation in D\textsubscript{2}O did not result in the growth of a distinguishable intermediate species corresponding to the exchange of two axial CH\textsubscript{3}CN ligands, therefore, the population of the excited state only results in the dissociation of one CH\textsubscript{3}CN ligand as expected.

\textbf{Figure 7.4.} Photolysis of 1 monitored by \textsuperscript{1}H NMR spectroscopy in CD\textsubscript{3}CN, λ\textsubscript{irr} ≥ 435 nm, as a function of irradiation time. The singlet at 2.03 ppm corresponds to the axial CH\textsubscript{3}CN protons, the singlet at 2.88 ppm corresponds to the equatorial CH\textsubscript{3}CN protons, and the resonance growing in at 1.96 ppm corresponds to free CH\textsubscript{3}CN.
Photolysis of 1 ($\lambda_{\text{irr}} \geq 435$ nm) monitored by electronic absorption spectroscopy was carried out in H$_2$O (Figure 7.5a). At early irradiation times the $^1$MLCT band at ~495 nm decreases with the concomitant growth of a broad absorption band centered at 543 nm after 20 minutes. This shift in absorption of 1786 cm$^{-1}$ is of a similar magnitude observed for the complexes studied in Chapter 3 after exchanging two CH$_3$CN ligands, therefore, this intermediate species is assigned as $\{[\text{Ru(H}_2\text{O})(\text{CH}_3\text{CN})_2]_2(\text{tppz})\}^{4+}$ where an axial CH$_3$CN has substituted for H$_2$O on each metal. Further irradiation results in the decrease of this intermediate and the growth of band at 600 nm assigned as the final product $\{[\text{Ru(H}_2\text{O})_2(\text{CH}_3\text{CN})_2(\text{tppz})\}^{4+}$ where all axial CH$_3$CN have exchanged for solvent molecules. The lack of isosbestic points observed in the photolysis data indicates that the dissociative axial CH$_3$CN ligands to form the disubstituted and tetrasubstituted products do not exchange simultaneously agreeing with the analysis of the photolysis data monitored by $^1$H NMR spectroscopy. Low energy irradiation ($\lambda_{\text{irr}} \geq 610$ nm) exciting the tail of the MLCT band and just inside the therapeutic window for PCT results in similar spectral changes albeit at a much slower rate (Figure 7.5b). Also, no spectral changes are observed when the complex is kept in H$_2$O in the dark for 24 hours.
Figure 7.5. Electronic absorption spectral changes of 1 as a function of irradiation time in H$_2$O with (a) ($\lambda_{irr} \geq 435$ nm), t$_{irr}$ = 0 to 360 min and (b) ($\lambda_{irr} \geq 610$ nm). t$_{irr}$ = 0 to 480 min.

7.2.3 Calculations

Density functional theory (DFT) calculations reveal an optimized structure with significant geometric distortion about the Ru(II) metal center (Figure 7.6a). This is due to the repulsion of the hydrogen atoms on opposing phenazine rings. The highest occupied molecular orbitals (HOMOs) are calculated to be localized d-orbitals of both metals, as is typical of Ru–diimine complexes.$^{23}$ The LUMO – LUMO+5 are delocalized over the bridging tppz ligand. In order to further understand the selective ligand exchange of strictly the axial CH$_3$CN ligands in 1 the orientation of the lowest energy orbitals that possess Ru–L ($\sigma^*$) character need to be considered as was done in Chapter 4. In 1 these correspond to the degenerate LUMO+7 and LUMO+8 orbitals (Figure 7.6b).
is clear in Figure 7.6b that these orbitals are directed along the bonds of the axial CH₃CN ligands and the tppz ligand. Placing electron density in these orbitals will result in weakening of the axial Ru–CH₃CN bond and cause ligand dissociation to occur. The Ru–L (σ*) orbitals directed along the equatorial Ru–CH₃CN bond must be significantly higher in energy and are not observed when analyzing the first 10 LUMOs. A similar explanation was used for the complex [Ru(bpy)(CH₃CN)₄]²⁺ in which only the axial CH₃CN ligands exchange upon irradiation.²⁴ Furthermore, in the monometallic complex, [Ru(tpy)(CH₃CN)₃]²⁺, only the axial CH₃CN ligands are photolabile and is most likely due to similar orientation and energies of the e_g type orbitals involved in ligand dissociation.¹⁵

Figure 7.6. (a) Calculated optimized structure of 1 and (b) Electronic density plots of the calculated LUMO+7 and LUMO+8 orbitals (isovalue = 0.04, hydrogens omitted for clarity).

Time dependent DFT (TD-DFT) calculations of 1 reveal the lowest vertical singlet excited state possess a significant contribution, ~95%, from a HOMO → LUMO
transition but with a low oscillator strength, with a maximum at 549 nm \( f = 0.0001 \). A more intense absorption band is predicted at 472 nm \( f = 0.0054 \) calculated to possess 95% contribution from a HOMO-1 \( \rightarrow \) LUMO+1 transition. The 10 lowest energy vertical singlet excited states and orbitals that make up those transition are in Table 7.1.

Table 7.1. 10 lowest energy singlet excited states obtained from DFT calculations, and the transitions associated with these states in CH$_3$CN (H = HOMO, L = LUMO) for complex 1.

<table>
<thead>
<tr>
<th>Wavelength (nm)</th>
<th>( f )</th>
<th>Calculated Transitions and Orbital Contributions$^a$</th>
</tr>
</thead>
<tbody>
<tr>
<td>548.99</td>
<td>0.0001</td>
<td>H( \rightarrow )L(95%)</td>
</tr>
<tr>
<td>521.89</td>
<td>0.00004</td>
<td>H-2( \rightarrow )L(88%), H-4( \rightarrow )L(7%),</td>
</tr>
<tr>
<td>508.94</td>
<td>0.0004</td>
<td>H-3( \rightarrow )L(95%)</td>
</tr>
<tr>
<td>503.00</td>
<td>0.00004</td>
<td>H-4( \rightarrow )L(88%), H-2( \rightarrow )L(8%),</td>
</tr>
<tr>
<td>493.94</td>
<td>0.004</td>
<td>H( \rightarrow )L+1(62%), H-1( \rightarrow )L(36%)</td>
</tr>
<tr>
<td>472.30</td>
<td>0.0054</td>
<td>H-1( \rightarrow )L+1(94%)</td>
</tr>
<tr>
<td>466.08</td>
<td>0.00004</td>
<td>H-2( \rightarrow )L+1(94%),</td>
</tr>
<tr>
<td>459.40</td>
<td>0.001</td>
<td>H-3( \rightarrow )L+1(98%)</td>
</tr>
<tr>
<td>453.54</td>
<td>0.00004</td>
<td>H-4( \rightarrow )L+1(99%)</td>
</tr>
<tr>
<td>446.67</td>
<td>0.3761</td>
<td>H-1( \rightarrow )L(60%), H( \rightarrow )L+1(34%),</td>
</tr>
</tbody>
</table>

$^a$Only contribution \( \geq 5\% \) are listed.

7.2.4 DNA Binding

Gel electrophoresis assays were carried out in order to identify whether 1 displays photoinduced covalent DNA binding (Figure 7.7). It is well documented that cisplatin thermally binds to linearized DNA and reduces its migration through an agarose gel in a concentration dependent manner.$^1$ Furthermore, the monometallic complex, [Ru(tpy)(CH$_3$CN)$_3$]$^{2+}$, was shown to inhibit the migration of linearized DNA through an agarose gel similar to cisplatin although the binding mode is most likely different than
cisplatin due to the fact the photolabile ligands are position \textit{trans} to one another in the complex.\textsuperscript{15} In Figure 7.7, lanes 1 and 8 contain 1 kb DNA ladder, lanes 2–7 were loaded with 50 μM pUC18 DNA, and lanes 3–6 contain increasing concentrations of 1. In Figure 7.7a, the samples in lanes 3–6 were irradiated for 15 minutes with $\lambda_{\text{irr}} \geq 435$ nm light prior to loading. It is clear in Figure 7.7a that irradiation of the complex in the presence of linearized DNA does not result in a reduction of the DNA migration through the gel and is further evidenced by the similarity of the control gel, which was incubated in the dark for 15 minutes under analogous experimental conditions (Figure 7.7b). These results are indicative of no covalent binding of 1 to DNA when irradiated or kept in the dark. Also, no covalent binding is observed when irradiation times of the samples are doubled. This lack of binding is most likely due to the long irradiation times needed for the exchange of the second CH$_3$CN to occur or the multiple metal centers sterically hinder one another from forming a stable binding interaction with DNA.

\textbf{Figure 7.7.} Imaged ethidium bromide-stained agarose gels of 50 μM linearized pUC18 plasmid (10 mM phosphate buffer, pH = 7.8) in the presence of various concentrations of complex: lanes 1 and 8, 1 kb DNA molecular weight standard; lanes 2 and 7, linearized plasmid alone; lanes 3-6, 50, 100, 150, 200 μM of 1 (a) irradiated ($\lambda_{\text{irr}} \geq 435$ nm, 15 min), (b) incubated in the dark (20 min, 298 K).
7.3 \{[\text{Ru(bpy)}(\text{CH}_3\text{CN})_2]_2(\text{pbp})\}[\text{PF}_6]_4 \text{ and } \{[\text{Ru(phen)}(\text{CH}_3\text{CN})_2]_2(\text{pbp})\}[\text{PF}_6]_4

7.3.1 Synthesis and Characterization

Complexes 2 and 3 were synthesized from the corresponding precursor molecules Ru(bpy)(DMSO)$_2$Cl$_2$ and Ru(phen)(DMSO)$_2$Cl$_2$, respectively, in a similar manner. Two equivalents of the precursor and 1 equivalent of the pbp ligand were refluxed in DMF resulting in a dark purple solution containing the bimetallic chloride complexes [Ru(L)Cl$_2$]$_2$(pbp) (L = bpy or phen). After drying the solution, unlike the complexes synthesized in a similar manner in Chapter 4, the crude purple mixture was only slightly soluble in CH$_2$Cl$_2$ and resulted in an orange solution with the desired solid in suspension. The solution was filtered, washed with water, resulting in the pure chloride precursor complex. Standard reflux techniques were used to exchange the Cl$^-$ ligands for CH$_3$CN yielding an orange solid product, however, $^1$H NMR, confirmed two symmetric isomers present in which the ancillary bpy or phen ligands were cis or trans to one another. The mixture was subjected to single crystal growth (detailed in Chapter 2) and resulted in the isolation of the cis isomer shown in Figure 7.8. The crystal structure of 2 reveals a $\pi$–stacking interaction between the benzene ring of the pbp ligand and the pyridine rings of the bpy ligands, which are separated by 3.38 Å each. This $\pi$ stacking interaction most likely explains the formation of single crystals of this isomer as oppose to the trans isomer. The same method was employed to isolate one isomer of 3, however, the single crystals that grew were too small for the X-ray diffraction experiment and a crystal structure could not be obtained. The isolated isomer is most likely the cis isomer similar
to 2. All photochemical experiments were performed using the isolated *cis* isomers of the complexes.

**Figure 7.8.** ORTEP plots of two different perspectives of PF$_6^-$ salt of the of 2 (ellipsoids drawn at 50% probability).

### 7.3.2 Electronic Absorption and Emission

The absorption profiles of 2 and 3 in CH$_3$CN are nearly identical with $^1$MLCT $\lambda_{\text{max}}$ at 459 nm ($\varepsilon = 11$ 600 M$^{-1}$ cm$^{-1}$) and 458 nm ($\varepsilon = 12$ 300 M$^{-1}$ cm$^{-1}$), respectively, assigned as Ru(t$_{2g}$) $\rightarrow$ pbp(\(\pi^*\)) transitions and less intense absorption bands from 350 nm to 400 nm assigned as Ru(t$_{2g}$) $\rightarrow$ bpy(\(\pi^*\)) and Ru(t$_{2g}$) $\rightarrow$ phen(\(\pi^*\)) $^1$MLCT transitions, respectively. Emission at 77 K in CH$_3$CN is observed at 687 nm and 684 nm for 2 and 3, respectively, and the excitation spectrum generated when monitoring the emission at these wavelengths matches the absorption spectrum indicating a pure sample (Figure 7.9). In Chapter 6 the absorption and emission spectra under similar conditions for the monometallic complex [Ru(bpy)(PTPI)(CH$_3$CN)$_2$]$^{2+}$ that basically is composed of half of the bridging pbp ligand were reported to be 455 nm and at 711 nm, respectively, and
assigned as Ru(t_{2g}) \rightarrow \text{PTPI}(\pi^*) 1\text{MLCT and } 3\text{MLCT transitions.}^{25} \text{ The lack of red shift observed in the bimetallic complexes relative to the monometallic complex indicates no involvement of the bridging benzene ring in the lowest energy excited states. Photophysical experiments and electrochemical studies of the complex } \{[\text{Ru(bpy)}_2](\text{pbp})\}^{4+} \text{ reveal no electronic communication between the two metal centers further supporting the excited state assignments of 2 and 3.}^{26}

**Figure 7.9.** Electronic absorption in CH\textsubscript{3}CN, excitation spectra in CH\textsubscript{3}CN at 77 K, and emission spectrum in CH\textsubscript{3}CN at 77 K of complex 2.

### 7.3.3 Photochemistry

The changes to the electronic absorption spectra of 2 and 3 in H\textsubscript{2}O when irradiated (\lambda_{irr} \geq 435 \text{ nm}) are shown in Figure 7.10. In 1, the peak at \sim 455 \text{ nm decreases while the growth of an absorption band centered at about 480 \text{ nm increases indicative of CH}_3\text{CN exchanging for H}_2\text{O molecules. The lack of distinct isosbestic points suggest no clear}
intermediate species similar to 1 and it is not clear whether the CH₃CNs are exchanging at both metal centers, but due to the lack of electronic communication between metal centers it is hypothesized that each Ru(II) subunit is photochemically reactive independent of one another and that both are undergoing ligand substitution, however, at a very slow rate relative to other CH₃CN ligand exchange complexes. Similar spectral changes are observed for 3 under analogous conditions (Figure 7.10b) and no ligand exchange is observed for the complexes when kept in the dark over the same time periods.

![Figure 7.10](image)

**Figure 7.10.** Electronic absorption spectral changes as a function of irradiation time in H₂O of (a) 2, tₐₐₐ = 0 to 490 min and (b) 3, tₐₐₐ = 0 to 240 min (λₐₐₐ ≥ 420 nm).

In order to gain better insight into the photochemistry of the complexes, the photolysis of 1 in CD₃CN was monitored by H NMR spectroscopy and is shown in Figure 7.11. The resonances at 2.27 ppm and 2.37 ppm correspond to both bound CH₃CN ligands at each metal and integrations of 1.00 were made using an internal standard of
benzene. Irradiation ($\lambda_{irr} \geq 435$ nm) for ~60 minutes results in the decrease of these resonances with the concomitant growth of a resonance at 1.96 ppm corresponding to free CH$_3$CN confirming that the CH$_3$CN ligands are photolabile. Moreover, both resonances decrease over the duration of the experiment indicating that both CH$_3$CN ligands are photolabile and most likely at both metal centers.

![Figure 7.11](image_url)

**Figure 7.11.** Photolysis of 2 monitored by $^1$H NMR spectroscopy in CD$_3$CN, $\lambda_{irr} \geq 420$ nm, after 60 minutes of irradiaiation. The singlets at 2.27 and 2.37 ppm correspond to the bound CH$_3$CN protons with integrations relative to a benzene internal standard, and the resonance growing in at 1.96 ppm corresponds to free CH$_3$CN. The resonances at 2.13 ppm and 2.08 ppm correspond to residual water and acetone.

An important point to take note of is that no change is observed in the aromatic region after irradiation of the complex in CD$_3$CN (Figure 7.12). Under similar conditions and described in Chapter 6, the monometallic complex [Ru(bpy)(PTPI)(CH$_3$CN)$_2$]$^{2+}$
undergoes photoinduced ligand dissociation accompanied by isomerization of the PTPI ligand. This resulted in the rise of new resonances corresponding to aromatic protons due to the new species forming in solution. Since no new aromatic resonance are observed when 2 is irradiated in CD$_3$CN, then no isomerization must be taking place suggesting that the coordination of two metal centers prevents the process from occurring.

![Figure 7.12](image)

**Figure 7.12.** $^1$H NMR spectrum of the aromatic region of 2 in CD$_3$CN after 60 min of irradiation ($\lambda_{irr} \geq 420$ nm). The singlet at ~7.3 ppm corresponds to the benzene internal standard.

### 7.3.4 DNA Binding

Gel electrophoresis assays were also carried out in order to identify whether 2 displays photoinduced covalent DNA binding (Figure 7.13). In Figure 7.13, lanes 1 and 8 contain 1 kb DNA ladder, lanes 2–7 were loaded with 50 μM pUC18 DNA, and lanes
3–6 contain increasing concentrations of 2. In Figure 7.13a, the samples in lanes 3–6 were irradiated for 15 minutes with $\lambda_{\text{irr}} \geq 435$ nm light prior to loading. It is clear in Figure 7.13a that, as the concentration of 2 is increased, DNA mobility decreases, whereas no shift in mobility is observed when the samples are incubated in the dark under similar experimental conditions (Figure 7.13b). These results are indicative of covalent binding of 2 to DNA only upon irradiation and 3 is expected to behave in the same way. Future experiments will investigate the cytotoxicity of the complexes.

**Figure 7.13.** Imaged ethidium bromide-stained agarose gels of 50 µM linearized pUC18 plasmid (10 mM phosphate buffer, pH = 7.8) in the presence of various concentrations of complex: lanes 1 and 8, 1 kb DNA molecular weight standard; lanes 2 and 7, linearized plasmid alone; lanes 3-6, 25, 50, 75, 100 µM of 1 (a) irradiated ($\lambda_{\text{irr}} \geq 435$ nm, 15 min), (b) incubated in the dark (20 min, 298 K).

7.4 $\{[\text{Ru(tpy)}(\text{CH}_3\text{CN})]_2(\text{bpm})\} \text{PF}_6_{14}$

**7.4.1. Synthesis**

Synthesis of the chloride precursor to generate 4 is straightforward but results in syn and anti isomers where the CH$_3$CN ligands are either trans to the nitrogen atoms on
the pyrimidine ring or opposing pyrimidine rings, respectively. Similar to complexes 2 and 3, single crystal growth results in the isolation of a single species characterized as the syn isomer by comparison of the $^1$H NMR spectra of it to that of the chloride precursor mixture, which was previously assigned.\textsuperscript{27}

7.4.2. Photophysical Data and Photochemistry

The $^1$MLCT $\lambda_{\text{max}}$ of 4 in CH$_3$CN is at approximately 600 nm with a tail that absorbs out to 700 nm making it a good candidate as a PCT agent (Figure 7.14). However, the complex does not undergo photoinduced ligand substitution in H$_2$O when irradiated with $\lambda \geq 395$ nm light. The lack of ligand exchange can be attributed to the large energy gap between the $^3$MLCT and $^3$dd states. Although the emission of $\{[\text{Ru(tpy)(bpy)}_2(bpm)]^{4+}\}$ emits at 820 nm at room temperature,\textsuperscript{28} no emission is observed in 4 up to 850 nm suggesting it emits in the near-IR range similar to the cyclometallated complex studied in Chapter 5,\textsuperscript{29} which also did not undergo ligand exchange. Cytotoxicity experiments should still be conducted on the complex due to the success of the cyclometallated complex discussed in Chapter 5.
7.5 Conclusions

In an effort to create molecules that could function as new PCT agents, the bimetallic complexes $\{\text{[Ru(CH$_3$CN)$_3$]}_2\text{(tppz)}\}^{4+}$ (1), $\{\text{[Ru(phen)(CH$_3$CN)$_2$]}_2\text{(pbp)}\}^{4+}$ (2), $\{\text{[Ru(phen)(CH$_3$CN)$_2$]}_2\text{(pbp)}\}^{4+}$ (3), and $\{\text{[Ru(tpy)(CH$_3$CN)]}_2\text{(bpm)}\}^{4+}$ (4) were synthesized and characterized. The advantages of the bimetallic systems include lower energy absorption towards the PCT window, the ability to release several drug equivalents, and the potential for multiple metal centers to bind to DNA. Complex 1 undergoes photoinduced ligand exchange of all axial CH$_3$CN ligands when irradiated with red light but does not covalently bind to DNA. However, the complex provides a platform to potentially release four nitrile based drugs upon irradiation, which is twice as many as previously reported complexes. All CH$_3$CN ligands are photolabile in complexes 2 and 3 when irradiated with visible light, and 2 displays a covalently binding interaction to DNA while remaining inactive in the dark. Cytotoxic studies for
complexes 2 and 3 are currently in progress. Although complex 4 absorbs into the therapeutic window, no photoinduced ligand exchange is observed when irradiated with visible light in H$_2$O. This is due to the large energy gap between the $^3$MLCT and $^3$dd states. These studies indicate that bimetallic complexes can be used to release multiple bioactive molecules upon irradiation and some have the ability to covalently bind to DNA, which could serve as dual functional PCT agents. Introducing steric bulk in 4 could lower the $^3$dd states enough to promote ligand dissociation when irradiated with red light.

References


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Chapter 8: Photophysical Study of [Ru(tpy)(L)(py) Complexes (L = bpy, 6,6′-dimethylbpy, biq, dppn, 6,3-dimethyldppn)]

8.1 Introduction

A deeper knowledge of the excited state dynamics and reactivity of Ru(II) complexes is of interest due to their widespread use in solar energy schemes, as molecular switches, and their potential in medicine, in which controlled ligand dissociation can be used as a vehicle for drug delivery. In previous chapters we discussed various Ru(II) complexes possessing monodentate CH$_3$CN ligands that are capable of undergoing relatively efficient ligand dissociation upon irradiation in coordinating solvents but are inert in the dark. The parent complex, [Ru(bpy)$_3$(CH$_3$CN)$_2$]$^{2+}$ (bpy = 2,2′-bipyridine), that led to the synthesis, characterization, and studies of the complexes in Chapters 3 – 7, undergoes sequential photoinduced ligand exchange of the CH$_3$CN ligands in H$_2$O resulting in the diaqua complex [Ru(bpy)$_3$(H$_2$O)$_2$]$^{2+}$ with $\Phi_{400} = 0.21$. This high efficiency of ligand dissociation has been translated to the idea of drug delivery in the complex [Ru(bpy)$_3$(L)$_2$]$^{2+}$ (L = nitrile containing cysteine protease inhibitor) for photochemotherapy (PCT). Irradiation of such drug release complexes with visible light results in enzyme inhibition corresponding to the exchange of both nitrile ligands; no decrease in enzyme activity is observed in the
dark at the same complex concentration indicating successful “caging” of bioactive molecules for PCT.\textsuperscript{14,17,18}

However, this dissertation has primarily focused on the photorelease of nitrile containing ligands due to the low quantum efficiency when using stronger $\sigma$-bonding monodentate ligands such as ammine ($\text{NH}_3$) or pyridine (py). For example, $[\text{Ru(bpy)}_2(\text{NH}_3)_2]^{2+}$ and $[\text{Ru(bpy)}_2(\text{py})_2]^{2+}$, exhibit ligand exchange quantum yield values, $\Phi$, of 0.018 ($\lambda_{\text{ex}} = 400$ nm, H$_2$O) and 0.0059 ($\lambda_{\text{ex}} = 436$ nm, CH$_3$CN and 1 mM Cl$^-$), respectively.\textsuperscript{15,19} Sadler and coworkers have explored Ru(II) arene complexes capable of releasing substituted pyridine ligands for PCT with visible light,\textsuperscript{20,21} but due to the inefficiency and exhaustive photolysis required, further advancement and applications involving the photodissociation of pyridine-containing ligands coordinated to Ru(II) has been largely impractical to date.

One way to increase the $\Phi$ value of pyridine ligand substitution is to introduce steric bulk,\textsuperscript{22} which was shown in Chapters 4 and 5 to also promote selective photoinduced ligand exchange and bidentate photoejection of diimine ligands.\textsuperscript{23,24} The model that is generally for photoinduced ligand dissociation in Ru(II) complexes involves thermal population of low lying $^3$LF (ligand field) states from the lowest energy $^3$MLCT (metal-to-ligand charge transfer) state.\textsuperscript{25-28} The population of the metal-based $^3$LF state in pseudo-octahedral low-spin d$^6$ complexes places electron density on the e$_g$-type orbitals with Ru–L($\sigma^*$) character, which weakens the bond and results in ligand dissociation.\textsuperscript{25-28} The addition of steric bulk to the ligand set in Ru(II) complexes provides a means to distort the pseudo octahedral geometry around the metal, which has been shown to
enhance the efficiency of the excited state ligand exchange.\textsuperscript{22,29} This observed enhancement is thought to be due to elongation of the Ru–N bond that results in lower energy \(^3\)LF states, making ligand dissociation more favorable upon excitation.\textsuperscript{22,29,30} This concept was utilized to promote efficient py ligand photodissociation, relative to the known complex [Ru(tpy)(bpy)(py)]\(^{2+}\) (1, tpy = 2,2′,6′,2′′-terpyridine), in [Ru(tpy)(dmbpy)(py)]\(^{2+}\) (2, dmbpy = 6,6′-dimethyl-2,2′-bipyridine) and [Ru(tpy)(biq)(py)]\(^{2+}\) (3, biq = 2,2′-biquinoline), with have \(\Phi_{500}\) values of \(<10^{-4}\), 0.16, and 0.03, respectively, in CH\(_3\)CN to generate the nitrile species (Figure 8.1 and Table 8.1).\textsuperscript{22} The methyl groups on 2 and fused benzene rings on 3 are positioned toward the center of the molecule to strain the pseudo-octahedral geometry around the metal center for efficient py exchange attributed to the lower energy of the \(^3\)LF states. The results support that incorporation of sterically demanding ligands in these types of systems provides a viable route for the release drugs containing pyridine units.

Another concept of interest is the investigation of complexes that possess dual photoreactivity for PCT, such that they undergo both ligand exchange and generate singlet oxygen (\(^1\)O\(_2\)) upon excitation.\textsuperscript{31} This phenomenon was observed in [Ru(bpy)(dppn)(CH\(_3\)CN)]\(^{2+}\) (dppn = benzo[\(i\)]dipyrido[3,2-a;2′,3′c]phenazine), where the quantum yields for ligand exchange in H\(_2\)O and for \(^1\)O\(_2\) production in methanol were measured to be \(\Phi_{400} = 0.002\) and \(\Phi_{460} = 0.72\), respectively.\textsuperscript{31} Transient absorption studies revealed a dppn-centered \(^3\)\(\pi\pi^*\) lowest energy excited state, typical of related Ru(II) complexes,\textsuperscript{32} with a lifetime of 20 \(\mu\)s. Further analysis of the transient absorption data revealed fast internal conversion from the \(^3\)MLCT state to the lower energy \(\pi\pi^*\).
However, for ligand substitution to take place, the $^3$LF states must be populated either through direct ISC from the $^1$MLCT state or from the vibrationally hot $^3$MLCT state; the former was proposed in the complex $[\text{Ru(bpy)}_2(\text{CH}_3\text{CN})_2]^{2+}$ and will be discussed later. To further investigate this concept, the complexes $[\text{Ru(tpy)}(\text{dppn})(\text{py})]^{2+}$ (4) and $[\text{Ru(tpy)}(\text{dmdppn})(\text{py})]^{2+}$ (5, 3,6-dimethyl-dppn) were synthesized and were expected to combine efficient pyridine photodissociation and $^1\text{O}_2$ generation in a single molecule.

The ligand exchange quantum yields of 4 and 5 in CH$_3$CN, were measured to be $<10^{-4}$ and 0.05 ($\lambda_{\text{irr}} = 500$ nm), respectively, and those for $^1\text{O}_2$ production in methanol were 0.98 and 0.69, respectively ($\lambda_{\text{irr}} = 460$ nm). Similar to 2 and 3, the steric bulk in 5 induces relatively efficient pyridine photodissociation, which is attributed to the lowering of the $^3$LF states. Also, complex 5 retains a relatively high quantum yield for $^1\text{O}_2$ production, indicative of the population of both the $^3$LF and $^3\pi^*$ states. In order to gain a better understanding of the excited state dynamics involved in photoinduced pyridine dissociation, the transient absorption (TA) and computational results of the new complexes 1 – 5, whose structures are depicted in Figure 1 and select photophysical data compiled in Table 8.1, will be analyzed and discussed.
Figure 8.1. Schematic representation of the molecular structures of 1 – 5.

Table 8.1. Photophysical data for 1 – 5.

<table>
<thead>
<tr>
<th>Complex</th>
<th>$\lambda_{\text{abs}}$/nm ($\varepsilon$/M$^{-1}$ cm$^{-1}$)$^a$</th>
<th>$\Phi$ ligand exchange$^b$</th>
<th>$\Phi^1$O$_2$$^c$</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>468 (8.12)</td>
<td>&lt;10$^{-4}$</td>
<td>NA</td>
</tr>
<tr>
<td>2</td>
<td>471 (8.02)</td>
<td>0.16</td>
<td>NA</td>
</tr>
<tr>
<td>3</td>
<td>530 (9.02)</td>
<td>0.03</td>
<td>NA</td>
</tr>
<tr>
<td>4</td>
<td>474 (12.9)</td>
<td>&lt;10$^{-4}$</td>
<td>0.98</td>
</tr>
<tr>
<td>5</td>
<td>486 (12.9)</td>
<td>0.05</td>
<td>0.69</td>
</tr>
</tbody>
</table>

$^a$ In acetone, at 298 K. $^b$ In CH$_3$CN, $\lambda_{\text{irr}}$ = 500 nm. $^c$ In MeOH, $\lambda_{\text{irr}}$ = 460 nm.
8.2 Results and Discussion

8.2.1. Transient Absorption of 1 – 3

The steady-state electronic absorption spectra of 1 – 3 were described in detail and are depicted in Figure 8.2. Complexes 1 and 2 exhibit nearly identical spectral features with lowest energy transitions centered at 468 nm ($\varepsilon = 8,120 \text{ M}^{-1} \text{ cm}^{-1}$) and 471 nm ($\varepsilon = 8,020 \text{ M}^{-1} \text{ cm}^{-1}$) in acetone, respectively, assigned to $^1$MLCT Ru(t$_{2g}$) $\rightarrow$ tpy($\pi^*$) transitions. The lowest energy absorption band of 3 is red shifted with respect to those 1 and 2, with $\lambda_{\text{max}} = 530$ nm ($\varepsilon = 9,020 \text{ M}^{-1} \text{ cm}^{-1}$) assigned to $^1$MLCT Ru(t$_{2g}$) $\rightarrow$ biq($\pi^*$) transitions. This red shift due to the stabilization of the biq($\pi^*$) orbitals relative to those of tpy($\pi^*$) or bpy($\pi^*$) orbitals, is typical in Ru(II) complexes, and was described in Chapters 3 – 5.

![Figure 8.2. Electronic absorption spectra of 1 – 3 in acetone.](image)

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The ultrafast TA spectra of 1 and 2 were measured in CH$_3$CN and are shown in Figure 8.3 ($\lambda_{ex} = 350$ nm, fwhm = 300 fs). A typical bleach of the ground state absorption is observed between 400 and 500 nm with a peak centered at ~470 nm in both complexes. A weaker positive transient absorption signal at ~380 nm is observed corresponding to the reduced tpy ligand in the lowest energy excited state, Ru($t_{2g}$)→tpy($\pi^*$) $^3$MLCT, as well as broad positive feature assigned as a ligand-to-metal charge-transfer (LMCT) transition from the neutral bpy/dmbpy ligand to the formally oxidized Ru$^{3+}$ ion of the $^3$MLCT state.$^{34,35}$ Our assignments are based on the similar spectral features observed in [Ru(tpy)$_2$]$^{2+}$. $^{36}$

Although the TA spectra of 1 and 2 display similar features, their excited state kinetics differ significantly. In 1 (Figure 8.3a), which does not undergo pyridine dissociation, the absorption changes of the bleach signal at 470 nm fits to a biexponential decay with $\tau_1 = 28$ ps (8%) and $\tau_2 = 544$ ps (92%). The absorption changes associated with the $^3$MLCT states in the 360–420 nm range display a maximum at ~370 nm at 1–10 ps delay times, but a red-shift to ~375 nm with a shoulder at ~390 nm at later times. These changes are accompanied by biexponential decays at 375 nm and 575 nm with $\tau_1$ ~3 ps and a long component with $\tau_2$ ~500 ps. The latter is slightly longer than the excited state lifetime reported for [Ru(tpy)$_2$]$^{2+}$ in CH$_3$CN, which has a lifetime of 124 ps,$^{36,37}$ but is significantly shorter than the overall excited lifetime of [Ru(bpy)$_3$]$^{2+}$, which is 700 ns in the same solvent.$^{38}$ The shorter lifetime observed in tpy complexes has been attributed to the bite angles associated with the tpy ligands, which distort the octahedral geometry and lower the energy of the $^3$LF states resulting in more accessible deactivation.
pathways.\textsuperscript{37,39} In 1, there is less strain from an octahedral geometry relative to [Ru(tpy)\textsubscript{2}]\textsuperscript{2+} resulting in higher energy \textsuperscript{3}LF states and a longer excited state lifetime.

\textbf{Figure 8.3.} Ultrafast transient absorption spectra in CH\textsubscript{3}CN of (a) 1 and (b) 2 with \(\lambda_{\text{ex}} = 350 \text{ nm} \) (fwhm = 300 fs) and of (c) 2 with \(\lambda_{\text{ex}} = 568 \text{ nm} \) (fwhm = 300 fs) collected 1, 5, 10, 20, 40, 60, 100, 200, 500, 1000, and 2000 ps following the laser pulse.
It is proposed that with 350 nm excitation, the Ru→bpy ¹MLCT is preferentially excited, resulting in fast ISC to the corresponding ³MLCT state with a maximum at ~370 nm, known to be associated with the reduced bpy ligand. This state decays to populate the Ru→tpy ³MLCT state within ~20 ps. Since the 28 ps component represents a minor fraction (8%) of the bleach recovery, it is proposed to correspond to changes in absorption that take place during the internal conversion (IC) process ³MLCT(bpy) → ...
$^3$MLCT(tpy), as shown schematically in Figure 8.4. The 23 ps IC from the $^3$MLCT state localized on bpy to that on tpy is consistent with the 26 ps value previously reported in CH$_3$CN for a related complex, [Ru(bpy)$_2$(dppp2)]$^{2+}$ (dppp2 = pyrido[2′,3′:5,6]pyrazino[2,3-f][1,10]phenanthroline), with two low-lying $^3$MLCT states.$^{40}

Excitation of 1 in CH$_3$CN with $\lambda_{ex} = 568$ nm selectively excites the Ru$\rightarrow$tpy $^1$MLCT absorption transition, such that the resulting kinetics are simplified. The bleach signal can now be fitted to $\tau_1 = 6$ ps (12%) and $\tau_2 = 437$ ps (88%), and similar kinetics are observed for the decay of the $^3$MLCT signal at 375 nm. Together these results can be interpreted as shown in Figure 4a, where the 6 ps component corresponds to the vibrational relaxation in the Ru$\rightarrow$tpy $^3$MLCT state, which then decays to regenerate the ground state with $\tau = 470$ ps. As expected, the 23-28 ps corresponding to IC from the higher energy $^3$MLCT state component is not present with 568 nm excitation.

It is evident in Figure 8.3 that 2, which undergoes efficient pyridine ligand dissociation, has a significantly shorter-lived excited state than 1; the TA spectra that results from 350 nm and 568 nm excitation of 2 are shown in Figures 8.3b and 8.3c, respectively (fwhm = 300 fs). Selective population of the Ru$\rightarrow$tpy $^1$MLCT state with 568 nm excitation reveals the absorption signals at $\sim$375 and $\sim$390 nm associated with the reduced tpy ligand in the Ru$\rightarrow$tpy $^3$MLCT state (Figure 8.3c). These features are also apparent with 350 nm excitation in Figure 8.3b, but the intensity of the peak at $\sim$370 nm relative to the shoulder at $\sim$390 nm is significantly greater, consistent with significant population of the Ru$\rightarrow$dmdbpy $^3$MLCT state. The biexponential fit to the bleach recovery
at 470 nm following 568 nm excitation results in $\tau_1 = 7$ ps (16%) and $\tau_2 = 38$ ps (84%) and a monoexponential decay of the signal at 375 nm with $\tau = 6$ ps. A long component is not apparent at 375 nm, such that the 6 ps decay can be attributed to IC of the Ru$\rightarrow$tpy $^3$MLCT to the $^3$LF state. The $^3$LF state then returns to the ground state in 38 ps evident in the bleach recovery signal; the 7 ps time constant with low amplitude changes are attributed to larger differences in extinction coefficient of the $^3$MLCT and $^3$LF states at 470 nm. The $^3$LF state repopulates the ground state with time constant of 38 ps; these processes are depicted in Figure 8.4b. Excitation of 2 with 350 nm results in similar kinetics, but with the population of the $^3$LF state from the Ru$\rightarrow$dmppy $^3$MLCT state within ~3 ps, as shown in Figure 8.4b. These experiments are consistent with the population of the $^3$LF state within 3-7 ps, which then deactivates via ligand dissociation and thermal decay to the ground state.

The steric strain imposed by the dmppy ligand in 2 induces additional distortions around the metal center resulting in lowering the energy of the $^3$LF state below the $^3$MLCT state and is consistent with our previous quantum yield data for ligand exchange in 2 relative to 1. It should be noted that these lifetimes are similar to those reported by Hauser for ruthenium(II) complexes with sterically bulky ligands, where the decay of the $^3$LF state to regenerate the ground state was reported to be 45 ps for [Ru(6-Mebpy)$_3$]$^{2+}$ and 7.5 ps for [Ru(4,4',6,6'-Me$_4$bpy)$_3$]$^{2+}$.30

Furthermore in Figure 8.3b, the ground state bleach at 470 nm does not completely return to baseline, indicative of the formation of a photoproduct on the timescale of the experiment, which is generally accepted to proceed through a dissociative mechanism.
and involving the formation of a pentacoordinate intermediate (PCI). Because the experiment is performed in CH$_3$CN solvent, a portion of the sample is expected to correspond to the [Ru(tpy)(dmbpy)(CH$_3$CN)]$^{2+}$ photoproduct. This point was further analyzed by calculating the difference spectrum of the initial species, 2, and the photoproduct that forms upon steady-state photolysis, [Ru(tpy)(dmbpy)(CH$_3$CN)]$^{2+}$. The resulting difference spectrum and the final 2 ns trace in Figure 8.3b are overlaid in Figure 8.5 and are in good agreement, consistent with the formation of the photoproduct on an ultrafast timescale. Due to the small shift in absorption between the initial species and the photoproduct, it is difficult to determine the kinetics of the or the dynamics associated with the formation and decay of the PCI. The detection of a PCI was hypothesized in ultrafast TA spectra of [Ru(bpy)$_2$(CH$_3$CN)$_2$]$^{2+}$ in H$_2$O when excited with 310 or 375 nm light, together with the formation of [Ru(bpy)$_2$(CH$_3$CN)(H$_2$O)]$^{2+}$ determined from the difference spectra of the starting material and mono-aqua product from the steady-state photolysis experiments.

![Graph](image)

**Figure 8.5.** Differential absorption spectrum of 2 and [Ru(tpy)(dmbpy)(CH$_3$CN)]$^{2+}$ (blue) and 2 ns time delay trace from transient absorption in CH$_3$CN (red).
A Comparison the ultrafast dynamics of [Ru(bpy)$_2$(CH$_3$CN)$_2$]$^{2+}$ in H$_2$O and CH$_3$CN led to the determination that the population of the $^3$MLCT and formation of the PCI occurs within the 300 fs laser pulse, suggesting that intersystem crossing (ISC) from the $^1$MLCT state directly populates both the $^3$MLCT and $^3$LF states, and the latter results in fast ligand dissociation.$^{41}$ The direct population from the $^1$MLCT state to two triplet states has been proposed by others; $^{42}$ one example is Cr(CO)$_4$(bpy), as determined by time resolved infrared measurements.$^{43}$ Furthermore, Stavros and coworkers have previously reported similar findings in a Ru(II) complex that undergoes ligand exchange, which involves the detection of a PCI and proposes that the $^3$LF state is populated from a vibrationally excited $^3$MLCT state.$^{44,45}$ It should be noted that given the recent time time-resolved studies by Hauser and those by Sadler and co-workers, it is possible that our previous assignments of the PCI may instead be due to the $^3$LF state, which is formed from the vibrationally hot $^3$MLCT state.$^{30}$ Direct ISC from the $^1$MLCT to the $^3$LF state cannot be ruled out at this time. Further ultrafast TA and vibrational experiments of 1 and 2 in pyridine will aid in a better understanding of the dynamics of ligand exchange dynamics in these complexes.

In order to further investigate the ligand substitution mechanism in 2, TA experiments were carried out in CH$_2$Cl$_2$ in the presence or absence of excess Cl$^-$ (500 mM) shown in Figure 8.6 ($\lambda_{ex} = 350$ nm, fwhm = 300 fs). In this system the photoproduc, [Ru(tpy)(dmbpy)(Cl)], absorbs lower energy light than the starting material, such that its spectral changes would be better resolved as a function of time. The resulting spectrum of 2 in the presence of 0.5 M Cl$^-$ possesses a ground state bleach
that does not completely decay indicating formation of the photoproduct, while in the absence of Cl\(^-\) the ground state beach fully recovers. However since the kinetics for both spectra result in \(\tau \sim 40\) ps for both the \(^3\)MLCT and ground state recovery, one interpretation is that ligand dissociation must occur instantaneously and the rate limiting step involves diffusion of the PCI and incoming Cl\(^-\) ligand. Alternatively, it is possible that because of low yield of product formation, absorption changes associated with this process are not large enough to provide a component in the lifetime decay fits.

**Figure 8.6.** Ultrafast transient absorption of 2 in CH\(_2\)Cl\(_2\) (a) in the presence (500 mM) and (b) absence of Cl\(^-\).

Ultrafast TA was also conducted on 3 in CH\(_3\)CN under similar conditions (Figure 8.7). The bleach of the ground state absorption is observed between 500 and 550 nm. A broad positive transient absorption signal in the ~380 - 500 nm ranges is also present
that corresponds to the reduced biq ligand in the lowest energy Ru(t\(_{2g}\) → biq(\(\pi^+\)) \(^3\)MLCT excited state, which was also observed in the complexes [Ru(biq)\(_2\)(CH\(_3\)CN)\(_2\)]\(^{2+}\) (Chapter 3) and [Ru(biq)\(_2\)(phen)]\(^{2+}\) (Chapter 5). The positive signal following 350 nm excitation exhibits a decay with \(\tau = 45\) ps, whereas this component is not present in the ground state bleach recovery. However, when using lower energy excitation (568 nm), the ground state bleach and the positive transient absorption vibrationally cool with \(\tau = 8\) ps. The difference when using 350 nm light excitation can be attributed to the population of the \(^1\)MLCT of the reduced tpy ligand that occurs to a lesser extent with 568 nm light excitation. Excitation to higher energy states results in great population of the higher energy tpy \(^3\)MLCT and then decaying to the lower energy biq \(^3\)MLCT state, which occurs in \(~45\) ps. Low energy excitation results in only population of the lower lying biq \(^3\)MLCT, therefore, the ultrafast kinetics of both \(^3\)MLCT and ground state recovery features are in agreement.

Figure 8.7. Ultrafast transient absorption of 3 in CH\(_3\)CN with (a) 350 nm and (b) 568 nm excitation (fwhm = 300 fs).
In order to determine the overall excited lifetime of 3, nanosecond TA experiments were performed in pyridine ($\lambda_{ex} = 355$ nm, fwhm = 8 ns, Figure 8.8). As expected the same positive and negative TA features are observed on the nanosecond timescale. Although the signal partially overlaps with the instrument response function, the signals at 390 nm, 520, and 650 nm were fitted to a monoexponential decay with $\tau \sim 50$ ns. This lifetime is similar to those measured for related ruthenium(II) complexes possessing the biq ligand, such as [Ru(biq)$_2$(CH$_3$CN)$_2$]$^{2+}$ and [Ru(biq)(phen)(CH$_3$CN)$_2$]$^{2+}$, with excited state lifetimes of $\sim$29 ns and $\sim$44 ns, respectively.

![Figure 8.8](image.png)

**Figure 8.8.** Nanosecond transient absorption of complex 3 in pyridine collected 12 ns after the laser pulse ($\lambda_{ex} = 355$ nm, fwhm = 8 ns).

### 8.2.2. Calculations of 1 – 3

Density functional theory (DFT) calculations of 1 – 3 were performed to better understand the differences in pyridine dissociation among the complexes. The highest occupied molecular orbitals (HOMOs) of 1 – 3 are calculated to be localized on the d-
orbits of the metal, as is typical of Ru(II)–diimine complexes. The lowest unoccupied molecular orbitals (LUMOs) of 1 and 2 are nearly identical with a LUMO and LUMO+1 localized on the tpy ligand and the LUMO+2 localized on the bpy/dmbpy ligand. The LUMO and LUMO+1 of 3 is equally delocalized on the tpy and biq ligand and a LUMO+2 and LUMO+3 localized on the tpy and biq ligands, respectively, which is unexpected from the ease of reduction of biq relative to tpy. The MO diagram for 1 and 2 is shown in Figure 8.9. The HOMOs of 1 and 2 were set to 0.0 eV given the nearly identical experimental oxidation potentials of the complexes; the orbitals possessing Ru–L(σ*) character are represented by dashed lines. The d_x^2-y^2 orbitals are calculated at significantly different energies of 12.34 eV and 11.52 eV for 1 and 2, respectively (Figure 8.9). Moreover, these orbitals are directed along the Ru–py bond and are associated with the Ru–L(σ*) LF states, therefore, placing electron density on this orbital is expected to weaken the Ru–py bond and result in ligand dissociation. The calculated lowered energy of the d_x^2-y^2 orbital in 2 relative to 1 supports the hypothesis of using steric bulk to enhance photoinduced ligand dissociation.

Time-dependent DFT (TD-DFT) calculations reveal that the lowest vertical singlet excited states of 1–3 possess significant contribution from HOMO → LUMO transitions with maxima at 523 nm (94%, f = 0.0073), 534 nm (95%, f = 0.0082), and 545 nm (77%, f = 0.0064). It should be noted that these lowest energy electronic transitions predicted are slightly red-shifted relative to the experimental MLCT maxima. The lowest vertical energy singlet excited state in 1–3 involving the d_x^2-y^2 orbitals arise from HOMO → LUMO+10 transitions corresponding to maxima at 359 nm (73%, f = 0.0011), 377 nm
(59\%, f = 0.0010), and 381 nm (41\%, f = 0.0077), respectively. This treatment further supports that the addition of steric bulk in 2 and 3 lower the energy of states with contribution from the e_g-type orbitals, expected to result in enhanced population of the LF states upon excitation in these systems. Therefore, it may be concluded that the addition of steric bulk can be utilized to tune complexes to efficiently dissociate monodentate pyridine based ligands.

\[ \text{Figure 8.9. Molecular orbital diagrams of 1 and 2, where dashed lines represent Ru–L (σ*) orbitals and the } d_{x^2-y^2} \text{ orbitals of 1 and 2.} \]
8.2.3. Transient Absorption of 4 and 5

The steady-state electronic absorption spectra of 4 and 5 in acetone are shown in Figure 8.10 and exhibit dppn-based \(^1\pi\pi^*\) transitions with maxima at 386 nm (\(\varepsilon = 12\ 200\ M^{-1}\ cm^{-1}\)) and 407 nm (\(\varepsilon = 13\ 400\ M^{-1}\ cm^{-1}\)) in the former and at 382 nm (\(\varepsilon = 11\ 400\ M^{-1}\ cm^{-1}\)) and 404 nm (\(\varepsilon = 12\ 400\ M^{-1}\ cm^{-1}\)). These spectral features are similar to those of the free dppn ligand in CH\(_3\)Cl, with maxima at 390 nm (\(\varepsilon = 9400\ M^{-1}\ cm^{-1}\)) and 414 nm (\(\varepsilon = 12\ 500\ M^{-1}\ cm^{-1}\)). The typical \(^1\text{MLCT}\) bands arising from Ru(d\(\pi\))→L(\(\pi^*\)) transitions are prominent in 4 and 5, centered at 474 nm (\(\varepsilon = 12\ 900\ M^{-1}\ cm^{-1}\)) and 486 nm (\(\varepsilon = 12\ 900\ M^{-1}\ cm^{-1}\)), respectively. It is clear from quantum yields of ligand exchange listed in Table 8.1 that the methyl groups at the 3 and 6 positions of the dppn ligand in 5 results in a large enhancement for photoinduced pyridine ligand dissociation relative to 4, suggesting a greater population of the \(^3\text{LF}\) states in the former, although the \(^1\text{O}_2\) production by 5 is still relatively efficient (Table 8.1). The known dual active complex, [Ru(bpy)(dppn)(CH\(_3\)CN)]\(^{2+}\), has a significantly lower quantum yield for ligand exchange (0.002) than 5 but a similar quantum yield for \(^1\text{O}_2\) production. As stated in Chapter 1, we recently reported time-resolved TA data on [Ru(bpy)(dppn)(CH\(_3\)CN)]\(^{2+}\). In order to gain a further understanding of the ultrafast dynamics in 4 and 5, similar TA experiments to those for 1 – 3 were performed, and compared the results to those reported for [Ru(bpy)(dppn)(CH\(_3\)CN)]\(^{2+}\).
Figure 8.10. Electronic absorption spectra of 4 and 5 in acetone.

Nanosecond TA spectra measured in deaerated pyridine reveal a strong absorption band at ~535 nm for 4 and 5 with τ = 47 µs and τ = 50 µs (λ_{ex} = 355 nm, fwhm ~8 ns), respectively, and the spectrum of 4 is shown in Figure 8.11. Similar features are observed for [Ru(bpy)(dppn)(CH_3CN)_2]^{2+} under the same experimental conditions in CH_3CN and for the free dppn ligand in CHCl_3, with τ = 20 µs and τ = 18 µs, respectively, both of which correspond to the dppn \( \pi\pi^* \) excited state. Therefore, the lowest energy excited states in 4 and 5 are assigned to the ligand-centered dppn \( \pi\pi^* \) state. In contrast and described earlier, 2 exhibits a significantly shorter \(^3\)MLCT lifetime, 38 ps in CH_3CN, owing to the competing ligand dissociation process and thermal population of the \(^3\)LF state from the \(^3\)MLCT state, expected to lie at similar or slightly lower energy. The different spectral profiles and short lifetimes of the \(^3\)MLCT states of 1 – 3 further support that the excited states of 4 and 5 is the low-lying dppn \( \pi\pi^* \) state.\(^{32}\)
Figure 8.11. Nanosecond transient absorption of complex 4 in pyridine collected 5 ns after the laser pulse ($\lambda_{ex} = 355$ nm, fwhm = 8 ns).

As previously described, the $^3$MLCT states of 1 – 3 are populated within the ~300 fs laser pulse, as expected from the known fast ISC rates typical of Ru(II) complexes.$^{46}$ A point of interest is that the population of both the $^3$MLCT and dppn centered $^3\pi\pi^*$ states is observed in 4 and 5 within the excitation with an ultrafast laser pulse (~300 fs, 350 nm, Figure 8.12). This behavior can be explained through the population of several states of 4 and 5 upon excitation, since both $^1$MLCT and $^1\pi\pi^*$ transitions have significant oscillator strength at 350 nm, including states with Ru$\rightarrow$dppn/dmppn and Ru$\rightarrow$tpy $^1$MLCT character. This is apparent by the positive absorption signals for the Ru$\rightarrow$dppn/dmdppn $^3$MLCT and dppn/dmdppn $^3\pi\pi^*$ states that are observable within the laser pulse ($\lambda_{exc} = 350$ nm, fwhm = 300 fs), as is evident in the 1 ps trace in Figure 8.12. It was found that the $^3$MLCT state of 5 decays with $\tau_1 = 0.7$ ps (42%) and $\tau_2 = 9$ ps (58%) and the $^3\pi\pi^*$ state grows with time constants of $\tau_1 = 0.4$ ps (46%) and $\tau_2 = 6$ ps (54%). Similar kinetics were measured for 4 with 350 nm excitation in CH$_3$CN, where the decay of the $^3$MLCT state at 415 nm can be fitted to a biexponential function with $\tau_1 = 0.6$ ps (48%).
and \( \tau_2 = 8 \) ps (52\%) and the growth of the \( ^3\pi\pi^* \) state at 535 nm takes place with \( \tau_1 = 0.4 \) ps (38\%) and \( \tau_2 = 4 \) ps (62\%). However, since there are many potential states that may be involved in the dynamics with overlapping absorption features, then it is not possible to assign specific kinetics.

Figure 8.12. Ultrafast transient absorption of complexes (a) 4 and (b) 5 in CH\(_3\)CN (\( \lambda_{ex} = 350 \) nm, fwhm = 300 fs).

The population of both the \( ^3\)MLCT and \( ^3\pi\pi^* \) states upon excitation was also observed in the dual action complex \([\text{Ru(bpy)}(\text{dppn})(\text{CH}_3\text{CN})_2]^{2+}\).\(^{31}\) It was found for this complex that the decay of the \( ^3\)MLCT state (bpy') could be fitted to a time constant of
720 fs, while the risetime of the $^3\pi\pi^*$ peak followed a bioexponential growth of $\tau_1 = 630$ fs and $\tau_2 = 22$ ps. Due to the similarity of the fast time constants, the growth of the signal corresponding to the $^3\pi\pi^*$ state is believed to arise from internal conversion from the $^3\text{MLCT}$ to the $^3\pi\pi^*$ state. The sharpening of the 535 nm signal occurs with a time constant of 22 ps, was attributed to vibrationally cooling. Ligand dissociation is hypothesized to occur through direct population of the $^3\text{LF}$ states from the Franck–Condon state but was not observed under the experimental conditions because of the low quantum yield for this process.

![Ultrafast transient absorption of complex 5 in CH$_3$CN (\(\lambda_{ex} = 568$ nm, fwhm = 300 fs).](image)

**Figure 8.13.** Ultrafast transient absorption of complex 5 in CH$_3$CN (\(\lambda_{ex} = 568$ nm, fwhm = 300 fs).

In order to avoid direct population of the $^1\pi\pi^*$ state, the ultrafast TA spectra of 4 and 5 were recorded following selective excitation of the Ru–tpy $^1\text{MLCT}$ state with 568 nm pulses (fwhm = 300 fs), and the resulting traces for the latter are shown in Figure
The Ru→tpy $^3$MLCT state of 5 is observed with features at ~390 nm and ~415 nm at the earliest time monitored, 0.3 ps, along with a strong ground state bleach centered at ~480 nm (Figure 8.13). Although the signal at 535 nm that corresponds to the dmdppn $^3\pi\pi^*$ state is not observed at early times, it grows in with $\tau_1 = 2$ ps (28%) and $\tau_2 = 17$ ps (72%), concomitant with the decay of the $^3$MLCT signals fitted to $\tau_1 = 3$ ps (13%) and $\tau_2 = 18$ ps (87%) at 415 nm. The intensity change in the bleach signal at 480 nm can be attributed to the superimposed growth in the broad peak at 535 nm, not ground state recovery, since it can be fitted to nearly identical lifetime components, $\tau_1 = 1$ ps (16%) and $\tau_2 = 18$ ps (84%). The ~2 ps component is assigned as arising from ISC, IC, and vibrational cooling, while the 18 ps component is assigned to the population of the $^3\pi\pi^*$ from the $^3$MLCT state. Similar spectral features and kinetics were measured for [Ru(tpy)(dppn)(py)]$^{2+}$ in CH$_3$CN under 568 nm excitation, for which ISC takes place within the laser pulse and the growth of the 540 nm peak and bleach recovery at 470 can be fitted to $\tau_1 = 1.1$ ps (21%) and $\tau_2 = 22$ ps (79%). The long component can be ascribed to IC from the Ru→tpy $^3$MLCT to the dppn $^3\pi\pi^*$ state, while the short component is related to ISC, IC, and vibrational cooling processes. The data obtained for 4 and 5 with low energy excitation reveals that the original assignment of vibrational cooling of the $^3\pi\pi^*$ state in [Ru(bpy)(dppn)(CH$_3$CN)$_2$]$^{2+}$ was incorrect and that the 22 ps time component corresponds to IC from the $^3$MLCT to the $^3\pi\pi^*$ state.

The Jablonski diagram for the excited state dynamics of 5 following 568 nm excitation is depicted in Figure 8.14. The major difference in the kinetics of the corresponding dppn complex is the IC from the Ru→tpy $^3$MLCT to the dppn $^3\pi\pi^*$ state.
takes place with time constant of 22 ps, instead of 18 ps in the dmdppn complex. Since photoinduced ligand exchange in 5 is observed with $\lambda_{irr} \geq 550$ nm, the dissociative $^3$LF state(s) must be populated with low energy light, however, it is unclear at this time whether this process takes place through ISC directly from the singlet manifold or from the Ru→tpy $^3$MLCT state, but clearly does not result from direct population of $^1$LF state.

![Jablonski diagram for the excited-state dynamics of 5 in CH$_3$CN with 568 nm excitation.]

**Figure 8.14.** Jablonski diagram for the excited-state dynamics of 5 in CH$_3$CN with 568 nm excitation.

### 8.3 Conclusions

The excited state properties of the complexes [Ru(tpy)(bpy)(py)]$^{2+}$ (1), [Ru(tpy)(dmbpy)(py)]$^{2+}$ (2), [Ru(tpy)(biq)(py)]$^{2+}$ (3), [Ru(tpy)(dppn)(py)]$^{2+}$ (4), and [Ru(tpy)(dmdppn)(py)]$^{2+}$ (5) were investigated using transient absorption (TA) spectroscopy and computational methods. Complexes 2 and 3 undergo photoinduced pyridine ligand dissociation in H$_2$O with $\Phi_{500} = 0.16$ and 0.03, respectively, while no photoreactivity is observed for 1. This enhancement was proposed to be due to the steric
bulk introduced by the dmbpy and biq ligands, which lowers the energy of the $^3$LF states resulting in more efficient population of this state and ligand dissociation. Ultrafast TA experiments in CH$_3$CN of 1 and 2 reveal overall excited state lifetimes of $\sim$500 and $\sim$40 ps, further suggesting a lowering of the $^3$LF states. Moreover, formation of the photoproduct, [Ru(tpy)(dmbpy)(CH$_3$CN)]$^{2+}$, upon irradiation of 2 in CH$_3$CN is observed during the ultrafast experiment similar to previous experiments involving [Ru(bpy)$_2$(CH$_3$CN)$_2$]$^{2+}$ in H$_2$O and no change in the overall excited state lifetime is observed in 2 when using 350 nm or 568 nm excitation. Nanosecond TA experiments of 3 in pyridine reveal a relatively longer lifetime of approximately 50 ns, which is similar to other ligand exchange complexes possessing the biq ligand (Chapter 3). Calculations reveal a lowering of the energy of the e$_g$-type orbitals in 2 relative to 1 supporting lower energy $^3$LF states. Also, the vertical energies involving these orbitals are lower for 2 further supporting the lowering of the $^3$LF states. The use of steric bulk to efficiently dissociate pyridine could be useful for controlled drug release in photochemotherapy (PCT).

Complexes 4 and 5 were designed to undergo ligand dissociation and generate singlet oxygen ($^1$O$_2$) upon irradiation for potential applications as dual therapeutics in PCT. Both complexes generate $^1$O$_2$ when irradiated, however, 5 also undergoes photoinduced pyridine dissociation due to the steric bulk directed towards the metal center that results in lower energy $^3$LF states. Ultrafast TA experiments with 350 nm excitation in CH$_3$CN of 4 and 5 reveal almost identical spectra and kinetics and population of both the $^3\pi\pi^*$ and $^3$MLCT state within the laser pulse (300 fs) revealing
direct population of both from the singlet manifold via inter system crossing (ISC). In order to avoid direct population of the $^1\pi\pi^*$ states, the same experiment was carried out for 4 and 5 with 568 nm excitation, which would only result in population of the $^1$MLCT state. Low energy excitation did not result in direct population of the $^3\pi\pi^*$ state from the singlet manifold, but the state was populated from the higher energy $^3$MLCT state with longer internal conversion kinetics to the $^3\pi\pi^*$ state. The overall excited state lifetimes of 4 and 5 are both $\sim$50 $\mu$s. This shows that higher energy excitation results in faster population of the $^3\pi\pi^*$ state and suggests the singlet and triplet manifolds are strongly vibrationally coupled in these complexes. The results support that the introduction of steric bulk into dual action complexes is promising for PCT and that ultrafast TA is a useful tool to better understand the excited state dynamics of these complexes to potentially design better therapeutics in the future.

References


Chapter 9 : Conclusion

9.1 Progress Toward Understanding Ligand Exchange for Photochemotherapy

Conventional chemotherapeutic treatments suffer from poor selectivity resulting in detrimental side effects and increased resistance,\(^1\) and PCT can be an effective alternative to circumvent these drawbacks by providing a method to locally irradiate the affected area only killing the tumor cells. To date, the field of PCT has focused nearly exclusively on organic molecules that produce \(^1\)O\(_2\) as the reactive species when they absorb light. The work described in this dissertation provides a deeper understanding of the photochemistry and excited state properties of new Ru(II) complexes capable of undergoing ligand substitution reactions only upon irradiation for potential applications in photochemotherapy (PCT).

In Chapter 3, the series of new Ru(II) complexes \([\text{Ru(biq)(phen)(CH}_3\text{CN)}_2]^{2+}\) (1) (phen = 1,10-phenanthroline, biq = 2,2’-biquinoline), \([\text{Ru(phen)}_2(\text{CH}_3\text{CN})_2]^{2+}\) (2), and \([\text{Ru(biq)(CH}_3\text{CN)}_2]^{2+}\) (3) were synthesized, characterized, and their photochemical properties studied. Incorporation of the biq ligand in Ru(II) complexes proved to be a viable route to red shift the absorbance closer to the optimum window for PCT relative to the previously studied complex \([\text{Ru(bpy)}_2(\text{CH}_3\text{CN})_2]^{2+}\) (bpy = 2,2’-bipyridine). Although addition, of the biq ligand lowers the energy of the \(^3\)MLCT state in 3, the steric bulk
provided by the fused benzene rings directed towards the metal center lowers the energy of the \( ^3 \)LF states so that ligand exchange still remains efficient, however, 3 possesses a much longer lifetime than \([\text{Ru(bpy)}_2(\text{CH}_3\text{CN})_2]^{2+}\) and ligand exchange is not observed on the ultrafast timescale. DNA gel mobility assays and viscosity measurements reveal that 3 binds to DNA when irradiated with red light (≥610 nm), but remains inert in the dark. Further studies are currently in progress to attach bioactive molecules to Ru(II) biquinoline systems that can be released upon irradiation acting as dual functional therapeutics.

Interestingly, the tris-heteroleptic complex 1 undergoes unprecedented selective ligand exchange of one \( \text{CH}_3\text{CN} \) ligand preferential to the other and was confirmed to be the \( \text{CH}_3\text{CN} \) ligand trans to the to the phen ligand by isolation of a monosubstituted species characterized by X-ray crystallography. Furthermore, it was confirmed that ligand exchange in 1 is wavelength dependent, where one \( \text{CH}_3\text{CN} \) is substituted in coordinating solvents with low energy light (≥ 550 nm) and an enhanced substitution rate is observed when switching to higher energy light (≥ 420 nm). This selectivity could be useful as a synthetic tool of various tris-heteroleptic Ru(II) complexes, as well as a new method for timed drug delivery in PCT applications, where one drug is released preferentially to the other. In order to further understand the electronic and steric factors that govern selective ligand exchange, more tris-heteroleptic complexes with various dimethylphen or biquinoline ligands were studied.

This was done in Chapter 4 by synthesizing the complexes \([\text{Ru}(4,7\text{-dmphen})(\text{phen})(\text{CH}_3\text{CN})_2]^{2+}\) (3, 4,7-dmphen = 4,7-dimethyl-1,10-phenanthroline), \([\text{Ru}(5,6\text{-dmphen})(\text{phen})(\text{CH}_3\text{CN})_2]^{2+}\) (4, 5,6dmphen = 5,6-dimethyl-1,10-phenanthroline),
[Ru(2,9-dmphen)(phen)(CH$_3$CN)$_2$]$^{2+}$ \hspace{1em} (5, \hspace{0.5em} 2,9-dmphen = 2,9-dimethyl-1,10-phenanthroline), \hspace{1em} [Ru(3,3’-ibiq)(phen)(CH$_3$CN)$_2$]$^{2+}$ \hspace{1em} (6, \hspace{0.5em} 3,3’-ibiq = 3,3’-isobiquinoline), \hspace{1em} [Ru(1,1’-ibiq)(phen)(CH$_3$CN)$_2$]$^{2+}$ \hspace{1em} (7, \hspace{0.5em} 1,1’-ibiq = 1,1’-isobiquinoline), \hspace{1em} and \hspace{1em} [Ru(dap)(phen)(CH$_3$CN)$_2$]$^{2+}$ \hspace{1em} (8, \hspace{0.5em} dap = 1,12-diazaperylene) and comparing their photophysical and structural properties to each other and 1 and 2. Structural analysis of 1 – 3, 5, 6, and 8 reveal that the ligands 2,2’-ibiq and 2,9-dmphen, which possess bulky substituents adjacent to the coordinating nitrogen atoms, result in a tilting of the diimine ligand and significant torsional strain about the metal center that is not observed in 2, 3, 6, and 8. Although 5 has almost identical steady-state photophysical properties to 2 – 4, it undergoes selective photoinduced ligand dissociation similar to that observed in 1. Although complex 6 posseses separate charge transfer states, selectivity is not observed suggesting distortion caused by steric strain about the metal center is essential for the process to occur. Complex 8 does not undergo ligand substitution upon irradiation due to the presence of a low lying $^3\pi^*$ state. Although there is no evidence of steric strain in complex 7, selective ligand exchange is observed albeit at a much less efficient rate. Analysis of the optimized structure by DFT calculations reveals that tilting of the 1,1’-ibiq ligand must occur when bound to the metal and is consistent with two isomers present in the $^1$H NMR spectrum. This tilting is most likely similar to that of 1 and 5 and is a commonality between the complexes that undergo selective ligand exchange.

Calculations of the complexes indicate that the presence of the sterically straining ligand disrupts the symmetry about the metal center and orients the $e_g$ type orbitals involved in bonds along only one of the photolabile CH$_3$CN ligands while no preferential
orientation is observed in the complexes that are symmetric about the metal center. These Ru–L (σ*) orbitals are generally accepted to be involved in the ligand substitution process and are preferentially directed along one bond in those that undergo selective ligand exchange with the exception of complex 7. A platform has been generated that provides chemically inequivalent photolabile CH₃CN ligands in Ru(II) complexes. Better control and understanding of selective ligand substitution will be useful in the design of complexes that can release multiple drugs for applications in photochemotherapy.

Not only has the incorporation of the biq ligand in certain complexes resulted in selective ligand exchange, but it has been utilized in sterically strained systems for bidentate photodissociation for PCT. This concept was further analyzed in Chapter 5 with the new cyclometallated complex [Ru(biq)₂(phpy)](PF₆) (9, phpy = deprotonated 2-phenylpyridine) and the complex [Ru(biq)₂(dpbc)](PF₆) (12, dpb = 2,3-bis(2-pyridyl)benzoquinoxaline). The photophysical properties, electrochemistry, and photochemistry of 9 and 12 were compared to those of known photoactive complexes [Ru(biq)₂(bpy)](PF₆)₂ (10) and [Ru(biq)₂(phen)](PF₆)₂ (11). Complexes 10 and 11 undergo exchange of one of the biq ligands when irradiated with λ_{irr} ≥ 630 nm light in water to generate the corresponding complex, [Ru(biq)₂(bpy)(H₂O)₂](PF₆)₂ and [Ru(biq)₂(phen)(H₂O)₂](PF₆)₂ and was previously explained by the lowering of the energy of the ³LF states due to elongation of the Ru-N bonds caused by the sterically cumbersome diimine ligands.
Since \([\text{Ru(bpy)}_3\text{(PF}_6\text{)}_2] \) and \([\text{Ru(phen)}_3\text{(PF}_6\text{)}_2] \) are stable upon irradiation relative to the the biq complexes 10 and 11, steric bulk was thought to be a key criteria for photoinduced bidentate ligand exchange. Although the crystal structure of 9 reveals an elongated Ru–N(biq) bond, the complex does not display photoinduced ligand substitution in coordinating solvents under similar conditions as those used for 10 and 11. The difference in reactivity of the cyclometallated complex 9 may be ascribed to the increase in energy of the metal-centered \(^3\)LF states resulting from the bonding of the strong \(\sigma\)-donor phpy\(^–\) ligand, a finding supported by DFT calculations. Complexes 10 and 11 were thought to be good candidates as PCT agents owing to their covalent DNA binding upon irradiation with light in the photodynamic window but their potential may be limited to their lack of cellular uptake. Although the absorption maximum of 9 is red-shifted relative to those of 10 and 11, the lack of photochemistry of the former indicates that the use of cyclometallated Ru(II) complexes may not be a good strategy for the discovery of new PCT agents. However, cell studies revealed that 9 is more phototoxic than all other complexes studied and that cyclometalled complexes still may be promising candidates for PCT.

Interestingly, complex 12 undergoes photodissociation of a bidentate ligand, but the dpb ligand is released with red light rather than the biq ligand as observed in 10 and 11. Complex 12 possesses three bulky ligands (2 biq and 1 dpb) that cause steric strain of the octahedral geometry about the Ru(II) center rather than just two bulky biq ligands as in 10 and 11. Absorption, emission, electrochemistry, transient absorption, and calculations confirm the lowest energy excited state in 12 is localized on the dpb ligand rather than the
biq ligands as in 10 and 11. This may play a key role in the photoinduced ligand dissociation of the dpb ligand because crystallographic data displays similar geometric distortions in 12 relative to 9 and 11. Also, 12 displays a covalent binding interaction with DNA upon irradiation, but not in the dark, and an enhanced toxicity towards HeLa cells upon irradiation indicating it can potentially function as a useful PCT agent.

It is clear that using the biq ligand results in complexes that are photoactive with light just inside the PCT window, however to our knowledge, no Ru(II) dinuclear complexes have been studied with photolabile CH$_3$CN ligands at both metal centers. To this end, in effort to design superior PCT agents that absorb lower energy light, the bimetallic complexes [{[Ru(CH$_3$CN)$_3$]$_2$(tppz)}$^{4+}$ (13, tppz = tetra-2-pyridylpyrazine) and [Ru(bpy)(CH$_3$CN)$_2$(pbp)]$^{4+}$ (14, pbp = 1,4-bis(2-pyridylmethyleneamino)benzene), were synthesized and characterized. Complex 13 undergoes photoinduced ligand exchange of all axial CH$_3$CN ligands when irradiated with red light but does not covalently bind to DNA. However, the complex provides a platform to potentially release four nitrile based drugs upon irradiation, which is twice as many as our previously reported complexes.$^3$ All CH$_3$CN ligands are photolabile in complex 14 when irradiated with visible light and displays a covalently binding to DNA while remaining inactive in the dark. Our studies indicate that bimetallic complexes can be used to release multiple bioactive molecules upon irradiation and some have the ability to covalently bind to DNA, which could serve as a dual functional PCT agent.

Steric bulk was shown to greatly influence selective ligand exchange and bidentate ligand dissociation and the majority of this dissertation focuses on ligand substitution
reactions in Ru(II) complexes involving photolabile CH$_3$CN ligands. Recently, it was shown that the introduction of steric bulk lowers the energy of the $^3$LF states such that unprecedented pyridine dissociation is observed upon irradiation. This allows for the use of pyridine containing bioactive molecules for PCT. In order to further understand this pyridine dissociation process, the excited state properties of the complexes [Ru(tpy)(bpy)(py)]$^{2+}$ (15, tpy = 2,2';6',2''-terpyridine), [Ru(tpy)(dmbpy)(py)]$^{2+}$ (16, dmbpy = 6,6'-dimethyl-2,2'-bipyridine), [Ru(tpy)(biq)(py)]$^{2+}$ (17), [Ru(tpy)(dppn)(py)]$^{2+}$ (18, dppn = benzo[i]dipyrido[3,2-$a$;2',3'$c$]phenazine), and [Ru(tpy)(dmdppn)(py)]$^{2+}$ (19 dmdppn = 3,6-dimethylldppn) were investigated using transient absorption (TA) and computational calculations. Ultrafast TA experiments in CH$_3$CN of 15 and 16 reveal overall excited state lifetimes of ~544 and ~30 ps, further suggesting a lowering of the $^3$LF states that would act as an excited state deactivating pathway. Moreover, formation of the photoproduct, [Ru(tpy)(dmbpy)(CH$_3$CN)]$^{2+}$, upon irradiation of 16 in CH$_3$CN is observed during the ultrafast experiment similar to previous experiments involving [Ru(bpy)$_2$(CH$_3$CN)$_2$]$^{2+}$ and no change in the kinetics is observed in 16 when using 350 nm or 568 nm excitation. Nanosecond TA experiments of 17 in pyridine reveal a relatively longer lifetime of approximately 50 ns, which is similar to other ligand exchange complexes possessing the biq ligand (Chapter 3). Calculations reveal a lowering of the energy of the $e_g$-type orbitals in 16 relative to 15 suggesting more accessible energy $^3$LF states. Also, the vertical energies involving these orbitals are lower for 16 further supporting the lowering of the $^3$LF states.
Recently the complex \([\text{Ru}(\text{bpy})(\text{dppn})(\text{CH}_3\text{CN})_2]^{2+}\) was reported,\(^5\) which is capable of undergoing ligand dissociation and produce \(^1\text{O}_2\) upon irradiation acting as dual action therapeutic. To this end, complexes 4 and 5 were also designed to undergo ligand dissociation and generate singlet oxygen (\(^1\text{O}_2\)) upon irradiation. Both complexes generate \(^1\text{O}_2\) when irradiated, however, 5 also undergoes photoinduced pyridine dissociation due to the steric bulk directed towards the metal center. Ultrafast TA experiments with 350 nm excitation in CH\(_3\)CN of 4 and 5 reveal almost identical spectra and kinetics and population of both the \(^3\pi\pi^*\) and \(^3\text{MLCT}\) within the laser pulse (300 fs) suggesting direct population of both from the singlet manifold via inter system crossing (ISC). In order to avoid direct population of the \(^1\pi\pi^*\) states, the same experiment was carried out for 4 and 5 with 568 nm excitation, which would only result in population of the \(^1\text{MLCT}\) state. Low energy excitation did not result in direct population of the \(^3\pi\pi^*\) state from the singlet manifold, but the state was populated from the higher energy \(^3\text{MLCT}\) state by internal conversion corresponding to longer time constants associated with the rise of the \(^3\pi\pi^*\) state. The overall excited state lifetimes of 4 and 5 are both \(\sim 50\ \mu\text{s}\). This shows that higher energy excitation results in faster population of the \(^3\pi\pi^*\) state and suggests the singlet and triplet manifolds are strongly vibrationally coupled in these complexes. The results support that the introduction of steric bulk into dual action complexes is promising for PCT.
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