

MIXED-METAL RUTHENIUM-PLATINUM POLYAZINE SUPERMOLECULES:
SYNTHESIS, CHARACTERIZATION AND EXPLORATION OF DNA BINDING

by

Matthew Milkevitch

Dissertation submitted to the faculty of the Virginia Polytechnic Institute and State
University in partial fulfillment of the requirements for the degree of

DOCTOR OF PHILOSOPHY

in

CHEMISTRY

Dr. Karen J. Brewer, Chair

Dr. Brenda S.J. Winkel, Co-Chair

Dr. Gary L. Long

Dr. Brian E. Hanson

Dr. Mark R. Anderson

June 22, 2001
Blacksburg, Virginia

Keywords: Supermolecules, Electrochemistry, Spectroscopy, DNA Binding, Cisplatin,
1,1/t,t, Gel Electrophoresis, Gel Densitometry

Copyright 2001, Matthew Milkevitch

MIXED-METAL RUTHENIUM-PLATINUM POLYAZINE SUPERMOLECULES: SYNTHESIS, CHARACTERIZATION AND DNA BINDING

by

Matthew Milkevitch

(ABSTRACT)

The goal of this research was to design, prepare and study a new class of supermolecules coupling ruthenium and platinum, which would display covalent binding to DNA. These supermolecules were modular in design, facilitating the use of coordination chemistry to vary individual components within the design. Drawing upon the well-established efficacy of *cis*-diamminedichloroplatinum(II) (cisplatin) and the DNA-binding properties of select ruthenium polyazine complexes, the approach was to bind the *cis*-Pt^{II}Cl₂ active site of cisplatin to ruthenium light absorbers using the dpq and dpb bridging ligands (where dpq = 2,3-bis(2-pyridyl)quinoxaline, dpb = 2,3-bis(2-pyridyl)benzoquinoxaline). These complexes are potentially bifunctional, capable of DNA intercalation through the bridging ligand and covalent binding to DNA through the *cis*-PtCl₂ site. The use of the ruthenium(II) center allows these complexes to be cationic and provides for enhanced water-solubility relative to the neutral cisplatin. These ruthenium centers function as chromophores leading to efficient light absorption by the Ru-Pt systems throughout the ultraviolet & visible region of the electromagnetic spectrum.

Synthetic methods were developed to prepare the mixed-metal, bimetallic complexes $[(bpy)_2Ru(BL)PtCl_2](CF_3SO_3)_2$ and $[(phen)_2Ru(BL)PtCl_2](CF_3SO_3)_2$ (where $bpy = 2,2'$ -bipyridine, $phen = 1,10$ -phenanthroline) in high purity and good overall yields. These new systems were characterized using electronic absorption spectroscopy, electrochemistry, and FAB-MS. The DNA-binding ability of these complexes was probed by reaction with linearized plasmid DNA and subsequent analysis by native and denaturing gel electrophoresis. The known DNA binders, cisplatin and *trans*- $\{[PtCl(NH_3)_2]_2(\mu-H_2N(CH_2)_6NH_2)\}^{2+}$ (1,1/t,t), were examined under equivalent conditions and used as positive controls. Native gel electrophoresis was used to show that these complexes strongly bind DNA, retarding the migration of DNA through the gel in a fashion inversely proportional to the ratio of DNA base pairs (bp) to metal complex (mc). Equivalent studies using the ruthenium monometallic synthons, $[(bpy)_2Ru(BL)](CF_3SO_3)_2$ and $[(phen)_2Ru(BL)](CF_3SO_3)_2$, had no effect on DNA migration, suggesting that the Ru-Pt complexes bind to DNA through the $PtCl_2$ site. Analysis by denaturing gel electrophoresis determined that the Ru-Pt complexes bind to DNA in a fashion similar to cisplatin, forming primarily intrastrand adducts. However, these systems also appear to form interstrand adducts at a 10-fold lower metal concentration than cisplatin.

In addition to affecting the migration rate, the bimetallic complexes also significantly reduced the fluorescence of DNA-intercalated ethidium bromide for the Ru-Pt reacted samples at low-DNA bp: mc ratios. This was not observed for the cisplatin and 1,1/t,t treated samples. This observation was quantitated by gel densitometry. Precipitation of the DNA by cisplatin, 1,1/t,t and all four Ru-Pt complexes was determined not to be the cause of reduced ethidium bromide fluorescence intensity. Homogenous solution fluorescence quenching studies have revealed that the Ru-Pt complexes quench the emission of ethidium bromide even in the absence of DNA, whereas cisplatin and 1,1/t,t do not.

In order to compare the effects on DNA migration produced by cisplatin, 1,1/t,t and the Ru-Pt complexes, R_f values were calculated. This analysis has revealed that all

four Ru-Pt complexes retard DNA migration to approximately the same degree. Calculation of theoretical DNA migration distances, based upon the molecular weight change of DNA caused by metal-complex binding, have revealed that the observed affect on DNA migration cannot be accounted for by an increase in molecular weight alone. This indicates that changes in charge and three-dimensional shape of the DNA upon binding of the Ru-Pt complexes may also contribute.

The Ru-Pt complexes developed in this work represent a successful attempt to design and produce a new class of supermolecules that will covalently bind to DNA. The design of this molecule, coupling a ruthenium light-absorbing unit to a *cis*-PtCl₂ site, has proven to be synthetically achievable and the complexes prepared display easily tunable light absorbing properties and reversible metal and ligand based redox processes. These molecules have been shown to avidly bind DNA through the PtCl₂ site. A group of *in vitro* biological assays have been developed to explore and characterize the binding of these molecules to DNA. These molecules therefore have met the goals of this project, and represent a new class of compounds that may find diverse applications in medicine, chemistry, and molecular biology.

Table of Contents

Table of Contents.....	v
List of Figures.....	x
List of Tables.....	xv
Abbreviations.....	xvii
Acknowledgments.....	xviii
Chapter 1 Introduction.....	1
Cancer: history, incidence, causes.....	1
Cancer treatment: focus on chemotherapy.....	2
Survey of chemotherapeutic agents.....	3
Problems with chemotherapy and their consequences.....	3
Cisplatin: anticancer agent targeting DNA.....	5
Cisplatin: mechanism of action.....	6
DNA structure.....	7
Cisplatin: DNA adducts.....	7
Gel electrophoresis: studying drug-DNA adducts.....	11
Effects of cisplatin-DNA binding.....	13
Problems with cisplatin.....	14
Cisplatin analog research.....	14
Other metal-based anticancer agents.....	16
DNA intercalators.....	17
Ru(II) based DNA intercalators.....	19
[Ru(phen) ₃] ²⁺ : chiral DNA intercalator.....	23

[(bpy) ₂ Ru(dppz)] ²⁺ and [(phen) ₂ Ru(dppz)] ²⁺ “molecular light switches for DNA”	25
Ru(II) bimetallic complexes as DNA intercalators	27
Project description	28
Chapter 2: Experimental	34
Chapter 3: Results and Discussion	49
Synthesis: Ru-Pt complexes containing the bridging bidentate ligands dpq and dpb	49
Electrochemistry	50
Electronic absorption spectroscopy	58
The interaction of Ru-Pt bimetallic complexes with DNA	64
Native gel studies: concentration-dependent interaction with DNA	64
Determination of R _f values	74
Theoretical modeling of DNA migration, molecular weight effect	80
Gel densitometry studies: concentration dependent interaction with DNA	83
DNA precipitation studies	97
Ethidium bromide fluorescence quenching by Ru-Pt complexes	98
Denaturing gel studies: concentration-dependent interaction with DNA	111
Chapter 4: Conclusions and Future Work	126
Conclusions	126
Future Work	129
References	133
Appendix A-1 Cyclic voltammogram for [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂	143

Appendix A-2 Electronic absorption spectrum of [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ and [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂	144
Appendix A-3 Electronic absorption spectrum of [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂ and [(phen) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	144
Appendix A-4 Spectroscopic and electrochemical data used to construct the plot of the energies of the lowest energy absorption band (eV) vs. E _{1/2} (V) shown in Figure 3.12.....	145
Appendix B-1 Distance traveled by DNA bands (in mm) in a <i>cis</i> -[Pt(NH ₃) ₂ Cl ₂] (cisplatin) – DNA interaction study.....	146
Appendix B-2 R _f value determination for a <i>cis</i> -[Pt(NH ₃) ₂ Cl ₂] (cisplatin) – DNA interaction study.....	146
Appendix B-3 Distance traveled by DNA bands (in mm) in a <i>trans</i> -{[PtCl(NH ₃) ₂] ₂ (μ-H ₂ N(CH ₂) ₆ NH ₂)}(NO ₃) ₂ (1,1/ t,t), – DNA interaction study.....	147
Appendix B-4 R _f value determination for a <i>trans</i> -{[PtCl(NH ₃) ₂] ₂ (μ-H ₂ N(CH ₂) ₆ NH ₂)}(NO ₃) ₂ (1,1/ t,t), – DNA interaction study.....	147
Appendix B-5 Distance traveled by DNA bands (in mm) in a [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ , – DNA interaction study.....	148
Appendix B-6 R _f value determination for a [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ , – DNA interaction study.....	148
Appendix B-7 Distance traveled by DNA bands (in mm) in a [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂ , – DNA interaction study.....	149
Appendix B-8 R _f value determination for a [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂ , – DNA interaction study.....	149
Appendix B-9 Distance traveled by DNA Bands (in mm) in a [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ , – DNA interaction study.....	150
Appendix B-10 R _f value determination for a [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ , – DNA interaction study.....	150
Appendix B-11 Distance traveled by DNA bands (in mm) in a [(phen) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂ , – DNA interaction study.....	151

Appendix B-12 R _f value determination for a [(phen) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂ , – DNA interaction study.....	151
Appendix C-1 Gel densitometry data for <i>cis</i> -[Pt(NH ₃) ₂ Cl ₂] (cisplatin).....	152
Appendix C-2 Gel densitometry data for <i>trans</i> -{[PtCl(NH ₃) ₂] ₂ (μ-H ₂ N(CH ₂) ₆ NH ₂)}(NO ₃) ₂ (1,1/ t,t).....	153
Appendix C-3 Gel densitometry data for [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂	154
Appendix C-4 Gel densitometry data for [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	155
Appendix C-5 Gel densitometry data for [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂	156
Appendix C-6 Gel densitometry data for [(phen) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	157
Appendix C-7 Average IDV values for control samples obtained from metal-DNA experiments.....	158
Appendix C-8 Average IDV values from metal-DNA experiments shown in appendices C-1 through C-6, 5: 1 DNA bp: mc ratio.....	159
Appendix C-9 Average IDV values from metal-DNA experiments shown in appendices C-1 through C-6, 10: 1 DNA bp: mc ratio.....	160
Appendix C-10 Average IDV values from metal-DNA experiments shown in appendices C-1 through C-6 20: 1 DNA bp: mc ratio.....	161
Appendix C-11 Average IDV values from metal-DNA experiments shown in appendices C-1 through C-6, 100: 1 DNA bp: mc ratio.....	162
Appendix C-12 Average IDV values from metal-DNA experiments shown in appendices C-1 through C-6, 200: 1 DNA bp: mc ratio.....	163
Appendix C-13 Average IDV values from metal-DNA experiments shown in appendices C-1 through C-6, 300: 1 DNA bp: mc ratio.....	164
Appendix D-1 Emission data for ethidium bromide in the presence of cisplatin and 1,1/ t,t.....	165
Appendix D-2 Emission data for ethidium bromide in the presence of [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ and [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	166
Appendix D-3 Emission data for ethidium bromide in the presence of [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ and [(phen) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	167

Appendix D-4 Ethidium bromide quenching study for <i>cis</i> -[Pt(NH ₃) ₂ Cl ₂] (cisplatin) and <i>trans</i> -{[PtCl(NH ₃) ₂] ₂ (μ-H ₂ N(CH ₂) ₆ NH ₂)}(NO ₃) ₂ (1,1/ t,t).....	169
Appendix D-5 Ethidium bromide quenching study for [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ and [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	171
Appendix D-6 Ethidium bromide quenching study for [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ , and [(phen) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	173
Appendix E-1 Fast atom bombardment mass spectrum of [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂	174
Appendix E-2 Fast atom bombardment mass spectrum of [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	175
Appendix E-3 Fast atom bombardment mass spectrum of [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂	176
Appendix E-4 Fast atom bombardment mass spectrum of [(phen) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	177

List of Figures

Figure 1.1 <i>cis</i> -diamminedichloroplatinum(II) (cisplatin).....	5
Figure 1.2 <i>trans</i> -diamminedichloroplatinum(II) (transplatin).....	7
Figure 1.3 Structure of DNA Nucleotides.....	8
Figure 1.4 Structure of DNA double helix showing major and minor grooves.....	8
Figure 1.5 Schematic of cisplatin entering the cell and interacting with DNA, RNA and cellular thiols.....	10
Figure 1.6 Gel electrophoresis of DNA.....	12
Figure 1.7 Structures of cisplatin analogs presently used in the clinic.....	15
Figure 1.8 Structure of 1,1/t,t.....	16
Figure 1.9 Chemical structure of BBR 3464.....	16
Figure 1.10 Chemical structures of DNA intercalating dyes.....	18
Figure 1.11 Structure of 2,2',2''-terpyridine (terpy).....	18
Figure 1.12 Molecular orbital diagram for a d^6 octahedral complex, LCAO based.....	20
Figure 1.13 Major electronic transitions for Ru(II) polypyridyl compounds.....	22
Figure 1.14 Structure of $[(bpy)_2Ru(dppz)]^{2+}$	26
Figure 1.15 Structure of $[(phen)_2Ru(dppz)]^{2+}$	26
Figure 1.16 Structure of $[L_2Ru\{dppz(11-11')dppz\}RuL_2]$	28

Figure 1.17 Structures of $[(bpy)_2Ru(dpq)PtCl_2](CF_3SO_3)_2$ and $[(bpy)_2Ru(dpb)PtCl_2](CF_3SO_3)_2$	30
Figure 1.18 Structure of $(phen)_2Ru(dpq)PtCl_2(CF_3SO_3)_2$ and $[(phen)_2Ru(dpb)PtCl_2](CF_3SO_3)_2$	31
Figure 1.19 Terminal and bridging ligands used to construct Ru-Pt complexes.....	32
Figure 3.1 Synthetic scheme for $[(bpy)_2Ru(dpq)PtCl_2](CF_3SO_3)_2$	49
Figure 3.2 Electrochemical mechanism for complexes of the type $[(bpy)_2Ru(BL)]^{2+}$	53
Figure 3.3 Cyclic voltammogram of $[(bpy)_2Ru(dpq)PtCl_2](CF_3SO_3)_2$	55
Figure 3.4 Cyclic voltammogram of $[(bpy)_2Ru(dpb)PtCl_2](CF_3SO_3)_2$	55
Figure 3.5 Cyclic voltammogram of $[(phen)_2Ru(dpq)PtCl_2](CF_3SO_3)_2$	56
Figure 3.6 Cyclic voltammogram of $[(phen)_2Ru(dpb)PtCl_2](CF_3SO_3)_2$	56
Figure 3.7 Electrochemical mechanism for complexes of the type $[(bpy)_2Ru(BL)PtCl_2](CF_3SO_3)_2$	57
Figure 3.8 Electronic absorption spectrum for $[(bpy)_2Ru(dpq)](CF_3SO_3)_2$ and $[(bpy)_2Ru(dpb)](CF_3SO_3)_2$	59
Figure 3.9 Electronic absorption spectrum of $[(phen)_2Ru(dpq)](CF_3SO_3)_2$ and $[(phen)_2Ru(dpb)](CF_3SO_3)_2$	59
Figure 3.10 Electronic absorption spectrum of $[(bpy)_2Ru(dpq)PtCl_2](CF_3SO_3)_2$ and $[(bpy)_2Ru(dpb)PtCl_2](CF_3SO_3)_2$	60
Figure 3.11 Electronic absorption spectrum of $[(phen)_2Ru(dpq)PtCl_2](CF_3SO_3)_2$ and $[(phen)_2Ru(dpb)PtCl_2](CF_3SO_3)_2$	60

Figure 3.12 Plot of energies of the lowest energy absorption band (eV) vs. $E_{1/2}(V)$	63
Figure 3.13 Native agarose gel electrophoresis of interaction of <i>cis</i> -[Pt(NH ₃) ₂ Cl ₂] (cisplatin) and <i>trans</i> -{[PtCl(NH ₃) ₂] ₂ (μ -H ₂ N(CH ₂) ₆ NH ₂)(NO ₃) ₂ (1,1/t,t).....	65
Figure 3.14 Native agarose gel electrophoresis of interaction of [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ and [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	67
Figure 3.15 Native agarose gel electrophoresis of interaction of [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ and [(phen) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	69
Figure 3.16 Native agarose gel electrophoresis of interaction of [(bpy) ₂ Ru(dpq)](CF ₃ SO ₃) ₂ and [(bpy) ₂ Ru(dpb)](CF ₃ SO ₃) ₂	70
Figure 3.17 Native agarose gel electrophoresis of interaction of [(phen) ₂ Ru(dpq)](CF ₃ SO ₃) ₂ and [(phen) ₂ Ru(dpb)](CF ₃ SO ₃) ₂	71
Figure 3.18 Comparison of R _f values for the untreated control.....	77
Figure 3.19 Comparison of R _f values for <i>cis</i> -[Pt(NH ₃) ₂ Cl ₂] (cisplatin), <i>trans</i> -{[PtCl(NH ₃) ₂] ₂ (μ -H ₂ N(CH ₂) ₆ NH ₂)}(NO ₃) ₂ (1,1/t,t), [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ , [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂ , [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ , and [(phen) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	79
Figure 3.20 Standard curve relating the relationship between the molecular weight of DNA and distance traveled through a native gel.....	82
Figure 3.21 Comparison of gel densitometry results for the untreated control (no metal added), included with each respective metal-DNA interaction study.....	91
Figure 3.22 Comparison of gel densitometry results for [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ , [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂ , [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ , [(phen) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂ , <i>cis</i> -[Pt(NH ₃) ₂ Cl ₂] (cisplatin), and <i>trans</i> -{[PtCl(NH ₃) ₂] ₂ (μ -H ₂ N(CH ₂) ₆ NH ₂)}(NO ₃) ₂ (1,1 t,t).....	93
Figure 3.23 DNA precipitation study for <i>cis</i> -[Pt(NH ₃) ₂ Cl ₂] (cisplatin).....	95

Figure 3.24 DNA precipitation study for <i>trans</i> -{[PtCl(NH ₃) ₂] ₂ (μ-H ₂ N(CH ₂) ₆ NH ₂)}(NO ₃) ₂ (1,1/t,t).....	100
Figure 3.25 DNA precipitation study for [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂	101
Figure 3.26 DNA precipitation study for [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	102
Figure 3.27 DNA precipitation study for [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂	103
Figure 3.28 DNA precipitation study for [(phen) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	104
Figure 3.29 Ethidium bromide quenching study for <i>cis</i> -[Pt(NH ₃) ₂ Cl ₂] (cisplatin) and <i>trans</i> -{[PtCl(NH ₃) ₂] ₂ (μ-H ₂ N(CH ₂) ₆ NH ₂)}(NO ₃) ₂ (1,1/t,t).....	108
Figure 3.30 Ethidium bromide quenching study for [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ and [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	110
Figure 3.31 Ethidium bromide quenching study for [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ and [(phen) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	113
Figure 3.32 Schematic representation of intrastrand and interstrand crosslinking.....	114
Figure 3.33 Denaturing agarose gel electrophoresis of the interaction of <i>cis</i> -[Pt(NH ₃) ₂ Cl ₂] (cisplatin) with linearized plasmid DNA.....	116
Figure 3.34 Denaturing agarose gel electrophoresis of the interaction of [{ <i>trans</i> - PtCl(NH ₃) ₂ } ₂ (μ-H ₂ N(CH ₂) ₄ NH ₂)](NO ₃) ₂ (1,1/t,t) with linearized plasmid DNA.....	117
Figure 3.35 Schematic representing the affects of a small, intermediate and large number of interstrand crosslinks on the size and shape of a DNA molecule.....	119
Figure 3.36 Denaturing agarose gel electrophoresis of the interaction of [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ and [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	121

Figure 3.37 Denaturing agarose gel electrophoresis of the interaction of
[(phen)₂Ru(dpq)PtCl₂](CF₃SO₃)₂ and [(phen)₂Ru(dpb)PtCl₂](CF₃SO₃)₂.....123

List of Tables

Table 1.1 Risk factors for the development of cancer.....	2
Table 1.2 Survey of chemotherapeutic drug classes.....	4
Table 1.3 Absorption and electrochemical data for a series of Ru(II) bridging ligand and related complexes.....	24
Table 2.1 FAB mass spectral data for [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂	41
Table 2.2 FAB mass spectral data for [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	41
Table 2.3 FAB mass spectral data for [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂	42
Table 2.4 FAB mass spectral data for [(phen) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	43
Table 3.1 Electrochemical data for a series of ruthenium and ruthenium-platinum complexes incorporating the bidentate bridging ligands dpq and dpb.....	57
Table 3.2 Summary of UV-visible data for the Ru-Pt bimetallics.....	62
Table 3.3 Molecular weights of <i>cis</i> -[Pt(NH ₃) ₂ Cl ₂] (cisplatin), <i>trans</i> -{[PtCl(NH ₃) ₂] ₂ (μ-H ₂ N(CH ₂) ₆ NH ₂)}(NO ₃) ₂ (1,1/t,t), [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ , [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂ , [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ , [(phen) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	73
Table 3.4. R _f values for <i>cis</i> -[Pt(NH ₃) ₂ Cl ₂] (cisplatin), <i>trans</i> -{[PtCl(NH ₃) ₂] ₂ (μ-H ₂ N(CH ₂) ₆ NH ₂)}(NO ₃) ₂ (1,1/ t,t), [(bpy) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ , [(bpy) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂ , [(phen) ₂ Ru(dpq)PtCl ₂](CF ₃ SO ₃) ₂ , and [(phen) ₂ Ru(dpb)PtCl ₂](CF ₃ SO ₃) ₂	76
Table 3.5 Migration distance data for the 4361, 2322 and 2027 kb bands of the lambda DNA molecular weight standards.....	81
Table 3.6 Molecular weight and average migration distance data for 4361, 2322, 2027 kb fragments of lambda molecular weight standards.....	81
Table 3.7 Experimental and theoretical DNA migration distances for the interaction of <i>cis</i> -[Pt(NH ₃) ₂ Cl ₂] (cisplatin) with linearized plasmid DNA.....	84
Table 3.8 Experimental and theoretical DNA migration distances for the interaction of <i>trans</i> -{[PtCl(NH ₃) ₂] ₂ (μ-H ₂ N(CH ₂) ₆ NH ₂)}(NO ₃) ₂ (1,1/ t,t) with linearized plasmid DNA.....	85

Table 3.9 Experimental and theoretical DNA migration distances for the interaction of $[(bpy)_2Ru(dpq)PtCl_2](CF_3SO_3)_2$	86
Table 3.10 Experimental and theoretical DNA migration distance calculations for the interaction of $[(bpy)_2Ru(dpb)PtCl_2](CF_3SO_3)_2$ with cut plasmid DNA.....	87
Table 3.11 Experimental and theoretical DNA migration distances for the interaction of $[(phen)_2Ru(dpq)PtCl_2](CF_3SO_3)_2$ with linearized plasmid DNA.....	88
Table 3.12 Experimental and theoretical DNA migration distances for the interaction of $[(phen)_2Ru(dpb)PtCl_2](CF_3SO_3)_2$ with cut plasmid DNA.....	89
Table 3.13 Percent differences between initial and final absorbance recorded at 260 nm, for each metal-DNA precipitation study.....	105

Abbreviations

bpy	2,2'-bipyridine
bp	base pair
CT	charge transfer
C	control
DNA	deoxyribose nucleic acid
dpb	2,3-bis(2-pyridyl)benzoquinoxaline
dppz	dipyrido[3,2- <i>a</i> : 2', 3'- <i>c</i>]phenazine
dpq	2,3-bis(2-pyridyl)quinoxaline
HOMO	highest occupied molecular orbital
Kb	kilobase pairs
LC	ligand centered
LF	ligand field
LUMO	lowest unoccupied molecular orbital
mc	metal complex
MLCT	metal to ligand charge transfer
nt	nucleotides
phen	1,10-phenanthroline
py	pyridine
Ru-Pt	ruthenium-platinum complex
S	molecular weight standard

Acknowledgements

This research accomplishment was made possible only through the support and inspiration of numerous people. I owe a debt of gratitude to all of them:

to Almighty God, who endowed me with the strength, dedication and perseverance necessary to see this project through, and to whose glory this work is dedicated;

to my parents, Joe and Carole, and my sisters, Joann and Karen, for always being there for me with support, advice, and prayers;

to my wife Gail, for her unending and immeasurable support, understanding, patience, example, prayers, and above all, love;

to the Reed family, Frank, Lois, Beth, Anne, Allen, Joel, and Mark, for their interest in my work, unwavering support for its completion, and many prayers;

to my research advisor, Dr. Karen J. Brewer, for guidance, advice, and the willingness to work with me in completing this dissertation off-site;

to my co-advisor, Dr. Brenda S.J. Winkel, for her never-ending enthusiasm, advice, and inspiration;

to Dr. Gary L. Long, for sitting for my defense and becoming part of my advising committee at the very last minute;

to Dr. Mark R. Anderson, for advice, discussions, and the willingness to make the long trip back to Blacksburg for my defense;

to Dr. Brian E. Hanson, for advice and encouragement over the years;

to the Brewer research group, past and present, for being my friends, companions and instructors over many years;

to the Winkel research group, most notably Dr. Ian Burbulis, for useful advice, ideas and training;

to Mr. Jeff Clark, for useful conversations, advice, lodging on my many trips back to Blacksburg, and above all, friendship;

to Dr. N.P. Farrell of Virginia Commonwealth University, for his kind gift of the compound, 1,1/t,t;

and to the various funding agencies, among them Virginia Tech, The Jeffress Memorial Trust, and the National Science Foundation (CHE – 9632713) for financial support.