J. Chem. Sci., Vol. 117, No. 2, March 2005, pp. 153–159.

Mixed-ligand complexes of ruthenium(II) incorporating a diazo ligand: Synthesis, characterization and DNA binding

MEGHA S DESHPANDE and AVINASH S KUMBHAR*
Department of Chemistry, University of Pune, Pune 411 007, India
e-mail: askum@chem.unipune.ernet.in

Abstract. Mixed-ligand complexes of the type $[Ru(N\text{-}N)_2(dzdf)]Cl_2$, where N–N is 2,2′-bipyridine (bpy), 1,10-phenanthroline (phen) and 9-diazo-4,5-diazafluorene (dzdf), have been synthesized and characterized by elemental analysis, UV–Vis, IR and NMR spectroscopy. Binding of these complexes with calf thymus DNA (CT-DNA) has been investigated by absorption spectroscopy, steady-state emission spectroscopy and viscosity measurements. The experimental results indicate that the size and shape of the intercalating ligands have marked effect on the binding affinity of the complexes to CT-DNA. The complex $[Ru(phen)_2(dzdf)]Cl_2$ binds with CT-DNA through an intercalative binding mode, while the complex $[Ru(bpy)_2(dzdf)]Cl_2$ binds electrostatically.

Keywords. Ruthenium(II) polypyridyl complexes; 9-diazo-4,5-diazafluorene ligand; DNA binding.

1. Introduction

The interaction of transition metal complexes with nucleic acids is a major area of research due to the utility of these complexes in the design and development of synthetic restriction enzymes, chemotherapeutic agents, footprinting agents, spectroscopic probes, site-specific cleavers and molecular photoswitches.[1] Ruthenium polypyridyl complexes have been extensively studied in this context as their luminescence and photochemical reactivity make them exceptionally versatile as probes of DNA structures.[2–8] These complexes bind to DNA by non-covalent interactions such as electrostatic binding, groove binding, intercalative binding and partial intercalative binding.[9] Barton *et al* have pioneered the application of chiral transition-metal polypyridyl complexes to probe local variations in double-helical DNA structures and their role in gene expression,[9–13] and have recently reported the mismatch recognition agent $[Rh(bpy)_2(chrysi)]^{3+}$ (chrysi = 5,6-chrysenequinone diimine) that binds mismatch sites in DNA specifically and upon photoactivation cleaves the DNA backbone neighbouring the mismatch site.[14] Andree Kirsch-De Mesmaeker and coworkers[15] have developed bifunctional Ru(II) complex $[Ru(TAP)_2POQ\text{-}Nmet]^{2+}$ composed of a $[Ru(TAP)_2(phen)]^{2+}$ unit linked to an N-methyl-aminoquinoline moiety by a seven-atom chain (TAP = 1,4,5,8-tetraazaphenanthrene), the emission of which is dependent on the guanine content of the polynucleotide, thus developing a novel DNA sensor for guanine content.[15] They have also synthesized complexes with extended aromatic ligands such as $[Ru(phen)_2(dppz)]^{2+}$[16] (dppz = dipyrido [3,2-a: 2′,3′-c]phenazine), $[Ru(bpy)_2(hat)]^{2+}$[17] (hat = 1,4,5,8,9,12-hexaazatriphenylene), $[Ru(phen)_2(phehat)]^{2+}$[18] (phehat = 1,10-phenanthrolino[5,5-b]1, 4,5,8,9,12-hexaazatripheny-lene) and $[Ru(phen)_2(dpq)]^{2+}$[19] (dpq = dipyrido[3,2-d: 2′,3′-*f*]quinoxaline) which facilitate strong non-covalent binding interactions especially through complete or partial intercalation of one of the ligands. Complexes such as $[Ru(phen)_2(dppz)]^{2+}$ or $[Ru(phen)_2(phehat)]^{2+}$ are not luminescent in aqueous solution, but their emission is switched on when they intercalate a portion of their extended aromatic ligand into the stacking of the DNA bases.[16a,18,20] It has also been shown that the complexes of the type $[Ru(TAP)_2(dppz)]^{2+}$ undergo photo-induced proton-coupled electron transfer with guanosine-5′-monophosphate.[21] Liang Nian Ji *et al* have shown simple modifications on the ancillary ligands create interesting differences in space configurations and electron-density distribution of Ru(II) polypyridyl complexes which result in different DNA binding behaviours.[22] In another report, a series of Ru(II) polypyridyl complexes containing phenylhydrazones derived from 4,5-diazafluorene-9-one were synthesized which bind to CT DNA through variety of modes.[22e] Maiya *et al*[23] reported "molecu-

Dedicated to the memory of the late Professor Bhaskar G Maiya
*For correspondence

lar light switch" and "electro-photo switch" effects for complexes containing either a quinone-fused (qdppz = naphtha [2,3-a]dipyrido[3,2-h: 2′,3′-*f*] phenazine-5,18-dione) or a dicyano aromatic subunit (6,7-dicyanodipyridoquinoxaline) dppz based ligand and recently studied the redox chemistry of Ru(II) complexes of 6,7-dicyanodipyridoquinoxaline by pulse radiolysis techniques.[23d] They have also extended studies with a new modified phenanthroline ligand (ptzo = 1,10-phenanthrolino[5,6-*e*]1,2,4-triazine-3-one) showing moderate affinity for binding with CT DNA.[23e] Many useful applications of all the above complexes require that the complexes bind to DNA by an intercalative mode with a planar aromatic ligand that has been modified extensively, while the role of ancillary ligand has not been investigated in detail. Zaleski *et al*[24] have recently reported that 9-diazo-4,5-diazafluorene a nitrogen chelate-containing an exocyclic diazo group complexes with copper and effectively photocleaves DNA under anaerobic conditions using visible light. However, the interactions of other transition metal complexes containing the dzdf ligand and DNA have not been investigated. In this paper, we have synthesized and characterized Ru(II) polypyridyl complexes of the type $[Ru(N\text{-}N)_2dzdf]Cl_2$ where N-N is bpy, phen and dzdf is 9-diazo-4,5-diazafluorene ancillary ligand. The DNA binding of these complexes has been examined by absorption titration, luminescence quenching of ethidium bromide (EB) bound to DNA and viscosity measurements.

2. Experimental

2.1 *Materials*

All reagents and solvents were purchased commercially and were used as received. $RuCl_3.nH_2O$ was obtained from SD Fine Chemicals (India). Calf thymus DNA was purchased from SRL (India). Double-distilled water was used to prepare phosphate buffer. DNA concentration per nucleotide was determined by absorption spectroscopy using the molar absorption coefficient (6600 M^{-1} cm^{-1}) at 260 nm. Solutions of calf thymus DNA in phosphate buffer gave a ratio of absorbance at 260 and 280 nm of 1·8–1·9 : 1, indicating that the DNA was sufficiently free of protein.[25]

4,5-Diazafluoren-9-one (dafone),[26] 4,5-diazafluorenone 9-hydrazone (dzdfh)[27] and 9-diazo 4,5-diazafluorene (dzdf)[27] were synthesized by the reported procedures modified as shown below. $[Ru(bpy)_2Cl_2].2H_2O$ and $[Ru(phen)_2Cl_2].2H_2O$ were synthesized by the literature procedures.[28] Structures of the various polypyridyl ligands and the Ru(II) complexes used in the present study are given in figure 1.

2.2 *Synthesis of ligands*

2.2a *4,5-Diazafluoren-9-one (dafone):* 1,10-Phenanthroline (9 g, 0·05 M) and KOH (5 g, 0·09 M) were added to 850 ml of water and brought to reflux. $KMnO_4$ (25·30 g, 0·16 M) in 400 ml water was added dropwise to the refluxing mixture. After addition the solution was refluxed for 1 h and filtered to remove MnO_2; when the solution was cooled, crude 4,5-diazafluoren-9-one precipitated as yellow needles. Yield: 40%, m.p. = 212°C.

2.2b *4,5-Diazafluorenone-9-hydrazone (dzdfh):* 4,5-Diazafluoren-9-one (5 g, 0·027 mmol) and excess hydrazine hydrate in the presence of glacial acetic acid was refluxed in 100 ml methanol for 4 h resulting in the formation of 4,5-diazafluorenone-9-hydrazone. Yield = 70%, m.p. = 198–205°C.

2.2c *9-Diazo 4,5-diazafluorene (dzdf):* 4,5-Diazafluorenone-9-hydrazone (1·0 g, 5·1 mmol) was oxidized using a basic solution (8 drops of saturated KOH in water) of yellow HgO (1·2 g, 5·5 mmol) in benzene (100 ml). The solution was stirred overnight at room temperature and then filtered through glass wool or filter paper to remove the insoluble product of Hg waste. The resulting orange solution was concentrated to a solid and then recrystallised from a mixture of dichloromethane and pentane at –20°C. Yield: 60%, m.p. = 160°C.

2.3 *Synthesis of ruthenium(II) polypyridyl complexes*

2.3a *$[Ru(bpy)_2(dzdf)]Cl_2$*: A mixture containing $[Ru(bpy)_2Cl_2]$ (0·100 g, 0.19 mmol) and dzdf (0·037 g, 0·19 mmol) was refluxed in methanol/water (1 : 1) for 5 h to give a dark red solution. The solution was cooled to room temperature. After evaporation of the solvent, the solid was collected, washed with small amounts of methanol and diethyl ether, and dried under suction. The product was purified by column chromatography on alumina using acetone and methanol as eluent. Yield = 55%.

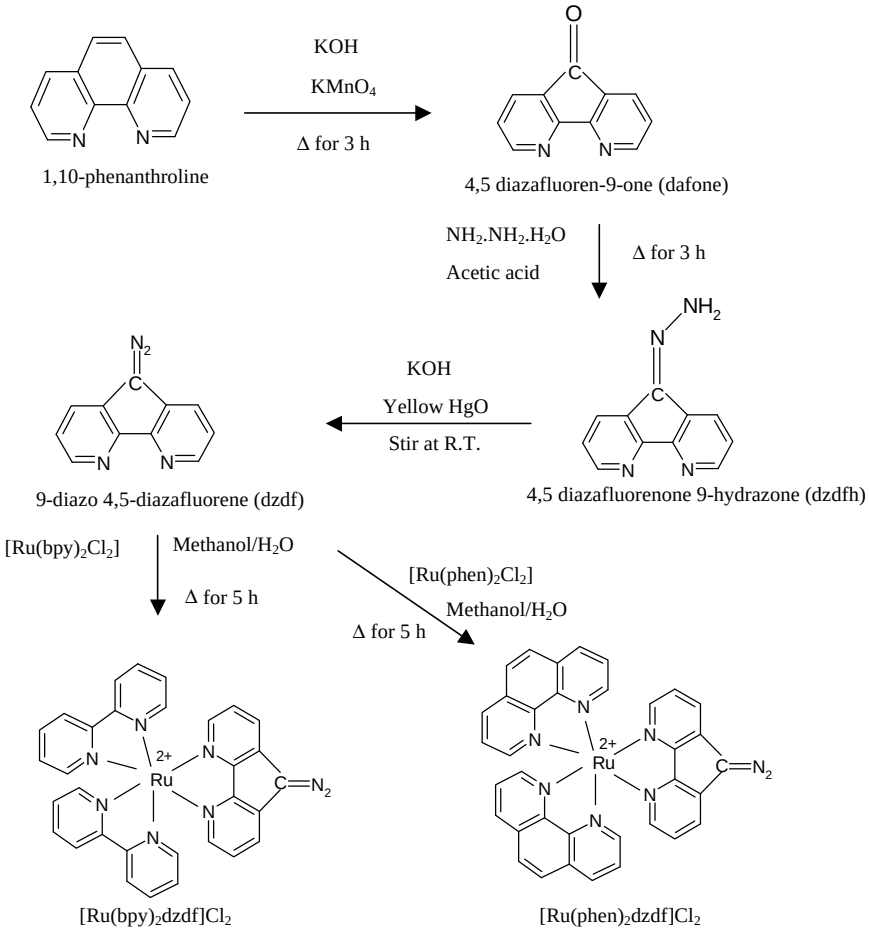

Figure 1. Scheme leading to synthesis of various polypyridyl ligands and Ru(II) polypyridyl complexes.

(Analysis – Found: C, 49·30; H, 4·27; N, 14·58%. Calcd. for $C_{31}H_{30}N_8O_4Cl_2Ru$: C, 49·59; H, 4·03; N, 14·93%.)

2.3b *[Ru(phen)$_2$(dzdf)]Cl$_2$:* A mixture containing [Ru(phen)$_2$Cl$_2$] (0·100 g, 0·16 mmol) and dzdf (0·032 g, 0·16 mmol) was refluxed in methanol/water (1 : 1) for 5 h to give a dark red solution. The solution was cooled to room temperature. After evaporation of the solvent, solid was collected, washed with small amounts of methanol and diethyl ether and dried under suction. The product was purified by column chromatography on alumina using acetone and methanol as eluent. Yield = 60%.

(Analysis – Found: C, 51·90; H, 3·50; N, 14·01%. Calcd. for $C_{35}H_{30}N_8O_4Cl_2Ru$: C, 52·62; H, 3·78; N, 14·03%.)

2.4 *Physical methods*

Microanalyses (C, H and N) were carried out on a Perkin–Elmer 240 Q elemental analyser at the National Chemical Laboratory, Pune. UV–Vis spectra were recorded on a Shimadzu UV-1601 spectrophotometer. ^{1}H NMR spectra were measured on a Varian-Mercury 300 MHz spectrometer at room temperature and all chemical shifts are given relative to TMS. The infrared spectra were recorded on a Shimadzu FTIR-8400 spectrophotometer. Spectra of solid samples were recorded by dispersing the samples in KBr pellets. Steady-state emission titrations were carried out on a Shimadzu RF-5301 spectrofluorometer at room temperature.

2.5 *DNA binding experiments*

2.5a *Absorption titration:* Absorption titrations of Ru(II) complexes in buffer (phosphate, pH 7·2) were done using a fixed ruthenium concentration to which increments of the DNA stock solution were added. Ruthenium solutions employed were 0·020 mM in concentration and calf thymus DNA was added to a ratio of 0–30 [DNA]/[Ru]. Ruthenium–DNA solutions

Table 1. IR and ^{1}H NMR spectral data.

Compound	IR (cm^{-1})[c]				^{1}H NMR (*d*, ppm)
	C=C	C=N	C=O	N=N	
dafone[a]	1403	1593	1718		8·81 (*d*, 2H), 8·01 (*d*, 2H), 7·37 (*dd*, 2H)
dzdfh[a]	1402	1562	–		8·56 (*dd*, 2H), 8·29 (*dd*, 2H), 7·98 (*dd*, 2H), 5·37 (NH_2, *s*)
dzdf[a]	1410	1593	–	2060	8·72 (*dd*, 2H), 7·91 (*dd*, 2H), 7·39 (*dd*, 2H)
$[Ru(bpy)_2(dzdf)]Cl_2$[b]	1421	1602	–	2104	8·47 (*m*, 5H), 8·05 (*m*, 9H), 7·73 (*dd*, 2H), 7·46 (*m*, 6H)
$[Ru(phen)_2(dzdf)]Cl_2$[b]	1421	1633	–	2088	8·42 (*m*, 6H), 8·07 (*m*, 8H), 7·55 (*m*, 6H), 7·27 (*d*, 1H), 7·17 (*d*, 1H)

^{1}H NMR spectra were recorded in [a]$CDCl_3$; [b]D_2O; [c]IR spectra were measured as KBr pellets

were allowed to incubate for 10 min before the absorption spectra were recorded. The intrinsic binding constant of the complex with CT-DNA was determined from the equation,

$$[DNA]/[e_a–e_f] = [DNA]/[e_b–e_f] + 1/K_b\,[e_b–e_f]. \quad (1)$$

through a plot of $[DNA]/[e_a–e_f]$ versus [DNA], where [DNA] is the concentration of DNA in the base pairs. The apparent absorption coefficients e_a, e_f and e_b correspond to A_{obsd}/[Ru], the extinction coefficient for the free ruthenium complex and the extinction coefficient for the ruthenium complex in the fully bound form respectively. The slope and *Y*-intercept of the linear fit of $[DNA]/[e_a–e_f]$ versus [DNA] give $1/[e_a–e_f]$ and $1/K_b\,[e_b–e_f]$ respectively. The intrinsic binding constant K_b can be obtained from the ratio of the slope to the *Y*-intercept.[29]

2.5b *Luminescence titration in the presence of ethidium bromide (EB):* Luminescence titration quenching experiments were conducted by adding small aliquots of 0–60 *m*M solutions of the Ru(II) complexes to samples containing 20 *m*M EB and 20 *m*M DNA in buffer. The resulting solution was allowed to equilibrate for 5–10 min at room temperature. The Stern–Volmer quenching constant was calculated according to the classical Stern–Volmer equation,[34]

$$I_0/I = 1 + Kr, \quad (2)$$

where I_0 and I are the fluorescence intensities in the absence and presence of complex respectively. K is a linear Stern–Volmer quenching constant and r is the ratio of the total concentration of complex to that of DNA.

2.5c *Viscosity measurements:* Viscosity experiments were carried out using a semi-micro viscometer maintained at 28°C in a thermostatic water bath. Flow time was recorded three times for each sample and average flow time was calculated. Data were presented as $(h/h°)^{1/3}$ versus binding ratio, where h is the viscosity of DNA in the presence of complex and $h°$ is the viscosity of DNA alone.[30]

3. Results and discussion

3.1 *Spectral characterization*

All the compounds synthesized in this study have been characterized by elemental analysis, UV–Vis, IR and ^{1}H NMR spectroscopic methods. Electronic absorption spectra of the complexes are characterized by metal-to-ligand charge transfer (MLCT) transitions in the visible region. The low energy bands at 446 nm, 443 nm for compounds $[Ru(bpy)_2(dzdf)]Cl_2$ and $[Ru(phen)_2(dzdf)]Cl_2$ respectively are assigned to the metal-to-ligand charge transfer transition. No luminescence was observed for the complexes $[Ru(bpy)_2(dzdf)]Cl_2$ and $[Ru(phen)_2(dzdf)]Cl_2$ upon excitation in the MLCT bands either in aqueous solution or in the presence of CT DNA.The important stretching frequencies observed in the infrared spectra are listed in table 1. ^{1}H NMR spectral data for the ligands and compounds synthesized in this study are summarized in table 1 which shows the expected peaks in the aromatic region.

3.2 *DNA binding*

DNA binding of Ru(II) complexes synthesized in this study with CT DNA has been monitored by absorption titration, luminescence quenching of ethidium bromide (EB) bound to DNA by the metal complexes and viscosity measurements.

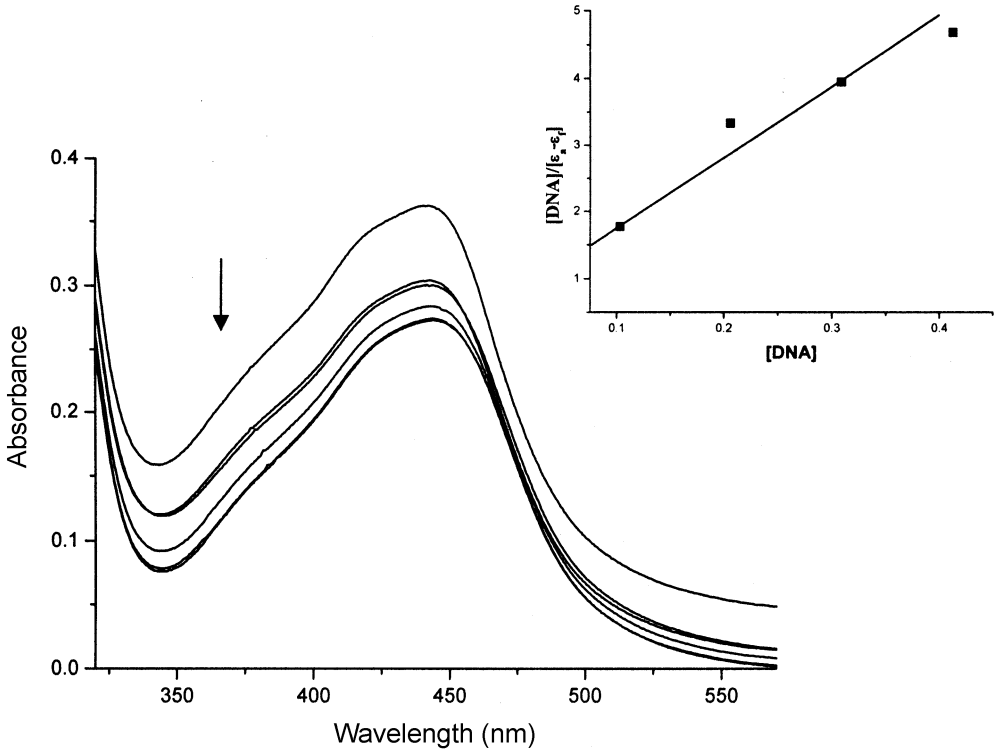

Figure 2. Absorption spectra of $[Ru(phen)_2dzdf]Cl_2$ in the presence of increasing amounts of CT DNA. [Ru] = 0·020 mM, [DNA]/[Ru] = 0–30. Inset illustrates the best fit of the binding data to (1) (see text for details).

Table 2. Spectroscopic properties on binding to CT DNA.

	Absorption[a]			
Compound	Free (e M^{-1} cm^{-1})	Bound[b]	l_{max} (nm) Δl	K_b
$[Ru(bpy)_2(dzdf)]Cl_2$	446 ($1{\cdot}73 \times 10^3$)	447	1	–
$[Ru(phen)_2(dzdf)]Cl_2$	440 ($2{\cdot}97 \times 10^3$)	443	3	$6{\cdot}83 \times 10^3$
$[Ru(bpy)_3]Cl_2$	452	452	0	–
$[Ru(phen)_2(dppz)]Cl_2$[c]	437	440	3	$>10^6$

[a][Ru] = 0·020 mM; all solutions in phosphate buffer (7·2); [b][DNA]/[Ru] = 30; [c]data from ref. [22a]

3.2a *Absorption titration:* Absorption titration can be used to observe the interaction of the complex with DNA. In general, hypochromism and red-shift are associated with the binding of the complex to the helix by an intercalative mode involving strong stacking interaction of the aromatic chromophore of the complexes between the DNA base pairs.[31] The absorption spectra of complex $[Ru(phen)_2(dzdf)]Cl_2$ in the absence and presence of CT DNA is given in figure 2 and the data tabulated in table 2. Only very weak hypochromism and spectral shift were found after complex $[Ru(bpy)_2(dzdf)]Cl_2$ was mixed with CT DNA. These optical changes are unlike those observed for proven intercalators (e.g. $[Ru(phen)_2dppz]^{2+}$), but very similar to that of $[Ru(bpy)_3]^{2+}$.[13,32] However, for complex $[Ru(phen)_2(dzdf)]Cl_2$, hypochromism and red shift are observed and binding constant estimated using (1) is of the order of 10^3 (see figure 2 inset).

3.2b *Luminescence titration in the presence of ethidium bromide (EB):* No luminescence was observed for the complexes $[Ru(bpy)_2(dzdf)]Cl_2$ and $[Ru(phen)_2(dzdf)]Cl_2$ upon excitation at the MLCT

bands either in aqueous solution or in the presence of CT DNA. Hence, competitive binding studies using ethidium bromide (EB) bound to DNA was carried out for these complexes. The quenching extent of fluorescence of EB bound to DNA is used to determine the binding of the complex and DNA. Binding of the complex results in the displacement of bound EB molecule with a reduction of emission intensity due to fluorescence quenching of free EB by water. The addition of the complex $[Ru(phen)_2(dzdf)]Cl_2$ to DNA pretreated with EB causes appreciable reduction in the emission intensity, indicating that the displacement of the EB fluorophore by the complex $[Ru(phen)_2(dzdf)]Cl_2$ results in a decrease of the binding of the ethidium to the DNA. However, very small decrease in emission intensity is observed for the complex $[Ru(bpy)_2(dzdf)]Cl_2$ indicating very weak binding affinity of this complex with CT DNA.

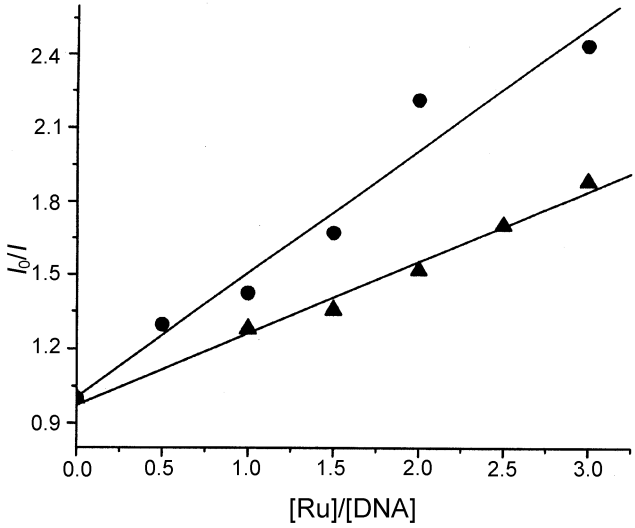

Figure 3. Fluorescence quenching curves of ethidium bromide bound to DNA in the presence of (▲) $[Ru(bpy)_2dzdf]Cl_2$ and (●) $[Ru(phen)_2dzdf]Cl_2$ ([EB] = 20 *m*M, [DNA] = 20 *m*M, [Ru] = 0–60 *m*M).

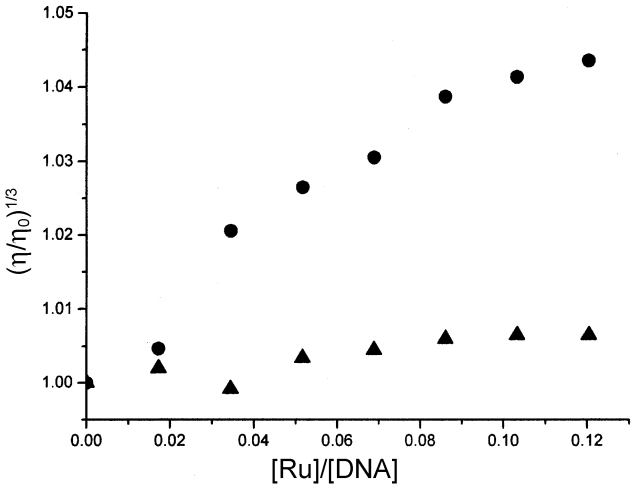

Figure 4. Effect of increasing amounts of (▲) $[Ru(bpy)_2(dzdf)]Cl_2$ and (●) $[Ru(phen)_2(dzdf)]Cl_2$ on the relative specific viscosity of CT DNA. ([DNA] = 0·2 mM, [Ru]/[DNA] = 0–0·12).

The fluorescence quenching curves of EB bound to DNA by the Ru(II) complexes are shown in figure 3. In the plot of I_0/I versus [Ru]/[DNA], K is given by the ratio of the slope to the intercept. The K values for $[Ru(bpy)_2(dzdf)]Cl_2$ and $[Ru(phen)_2(dzdf)]Cl_2$ estimated using (2) are 0·28 and 0·50 respectively, and suggest that the interaction of the complex $[Ru(phen)_2(dzdf)]Cl_2$ with DNA is strong as compared to that with $[Ru(bpy)_2(dzdf)]Cl_2$ which is consistent with the above absorption spectral results.

3.2c *Viscosity measurements:* Viscosity measurements were carried out, which are sensitive to change in length and are regarded as the most critical tests for the binding mode of complexes with DNA. The effect of the complexes $[Ru(bpy)_2(dzdf)]Cl_2$ and $[Ru(phen)_2(dzdf)]Cl_2$ on the viscosity of CT DNA is shown in figure 4. Two different kinds of behaviour can be distinguished by this experiment. Complex $[Ru(bpy)_2(dzdf)]Cl_2$ which exhibits a decrease in viscosity with increase in metal complex concentration indicates an electrostatic association of this complex with CT DNA. For complex $[Ru(phen)_2(dzdf)]Cl_2$, the viscosity of DNA increases with the increase of the concentration of the complex indicating intercalative binding mode.

4. Conclusions

Weak hypochromism and spectral shift and decrease in viscosity with increasing metal complex concentration indicate that the complex $[Ru(bpy)_2(dzdf)]Cl_2$ may bind to DNA by an electrostatic binding mode. Pronounced hypochromism and red-shift, and increase in viscosity with increasing metal complex concentration observed for the complex $[Ru(phen)_2(dzdf)]Cl_2$ indicates an intercalative binding mode. Stern–Volmer quenching constants for $[Ru(bpy)_2(dzdf)]Cl_2$ and $[Ru(phen)_2(dzdf)]Cl_2$ are 0·28 and 0·50 respectively, suggesting that the interaction of the complex $[Ru(phen)_2(dzdf)]Cl_2$ with CT DNA is the strongest, which is consistent with the above absorption spectral results and viscosity measurements.

Acknowledgements

ASK acknowledges the financial assistance from the Council of Scientific and Industrial Research, New Delhi. MSD thanks the Bhabha Atomic Research Centre, Mumbai, India for a research fellowship.

References

1. (a) Tullius T D 1989 In *Metal–DNA chemistry* (ed.) T D Tullius, ACS Symp. Ser. No. 402 (Washington, DC: Am. Chem. Soc.) pp 1–23; (b) Kelly S O and Barton J K 1999 In *Metal ions in biological systems* (eds) A Sigel and H Sigel (New York: Marcel Dekker) **39** 211
2. Erkkila K E, Odom D T and Barton J K 1999 *Chem. Rev*. **99** 2777
3. Sigman D S, Mazumder A and Perrin D M 1993 *Chem. Rev*. **93** 2295
4. Eriksson M, Leijon M, Hiort C, Norden B and Graslund A 1994 *Biochemistry* **33** 5031
5. Hudson B P, Dupureur C M and Barton J K 1995 *J. Am. Chem. Soc*. **117** 9379
6. Terbrueggen R H and Barton J K 1995 *Biochemistry* **34** 8227
7. Lecomte J P, Kirsch-De Mesmaeker A and Kelly J M 1996 *Top. Curr. Chem.* **177** 25
8. Lincoln P and Norden B 1998 *J. Phys. Chem.* **B102** 9583
9. Barton J K 1986 *Science* **233** 727
10. Barton J K, Dannenberg J J and Raphael A L 1982 *J. Am. Chem. Soc*. **104** 4967
11. Barton J K, Danishefsky A T and Goldberg J M 1984 *J. Am. Chem. Soc*. **106** 2172
12. Barton J K and Raphael A L 1985 *Proc. Acad. Sci. USA* **82** 6460
13. Kumar C V, Barton J K and Turro N J 1985 *J. Am. Chem. Soc*. **107** 5518
14. (a) Jackson B A and Barton J K 1997 *J. Am. Chem. Soc*. **119** 12986; (b) Jackson B A, Alekseyev V Y and Barton J K 1999 *Biochemistry* **38** 4655; (c) Jackson B A and Barton J K 2000 *Biochemistry* **39** 6176.
15. (a) Feeney M M, Kelly J M, Tossi A, Kirsch-De Mesmaeker A and Lecomte J P 1994 *J. Photochem. Photobiol.* **B23** 69; (b) Kelly J M, Feeney M M, Tossi A, Lecomte J P and Kirsch-De Mesmaeker A 1990 *Anti-Cancer Drug Design* **5** 69; (c) Jacquet L, Davies R J H, Kirsch-De Mesmaeker A and Kelly J M 1997 *J. Am. Chem. Soc*. **119** 11763
16. (a) Friedman A E, Chambron J C, Sauvage J P, Turro N J and Barton J K 1990 *J. Am. Chem. Soc*. **112** 4960; (b) Hartshorn R M and Barton J K 1992 *J. Am. Chem. Soc*. **114** 5919; (c) Jenkins T, Friedman A E, Turro N and Barton J K 1992 *Biochemistry* **31** 10809; (d) Coates C G, Jacquet L, McGarvey J J, Bell S E J, Obaidi A H R and Kelly J M 1997 *J. Am. Chem. Soc*. **119** 7130; (e) Tuite E, Lincoln P and Norden B 1997 *J. Am. Chem. Soc*. **119** 239; (f) Haq I, Lincoln P, Suh D, Norden B, Chowdhry B Z and Chaires J B 1995 *J. Am. Chem. Soc*. **117** 4788
17. (a) De Buyl F, Kirsch-De Mesmaeker A, Tossi A and Kelly J M 1991 *J. Photochem. Photobiol.* **A60** 27; (b) Lecomte J P, Kirsch-De Mesmaeker A, Feeney M M and Kelly J M 1995 *Inorg. Chem*. **34** 6481
18. Moucheron C, Kirsch-De Mesmaeker A and Choua S 1997 *Inorg. Chem*. **36** 584.
19. Collins J G, Sleeman A D, Aldrich-Wright J R, Greguric L and Hambley T W 1998 *Inorg. Chem*. **37** 3133
20. (a) Turro C, Bossmann S H, Jenkins Y, Barton J K and Turro N J 1995 *J. Am. Chem. Soc*. **117** 9026; (b) Hiort C, Lincoln P and Norden B 1993 *J. Am. Chem. Soc*. **115** 3448.
21. Ortmans I, Benjamin E, Kelly J M, Moucheron C and Kirsch-De Mesmaeker A 2004 *J. Chem. Soc., Dalton Trans*. 668
22. (a) Zhen O-X, Zhang O-L, Liu J-G, Ye B-H and Ji L-N 2000 *J. Inorg. Biochem*. **78** 293; (b) Liu J-G, Zang Q-L and Ji L-N 2001 *Transition Met. Chem.* **26** 733; (c) Chao H, Mei W-J, Huang O-W and Ji L-N 2002 *J. Inorg. Biochem*. **92** 165; (d) Yang G, Wang L and Ji L-N 1997 *J. Inorg. Biochem*. **67** 289; (e) Yang G, Wu J-Z, Wang L, Ji L-N and Tian X 1997 *J. Inorg. Biochem*. **66** 141; (f) Wu J-Z, Li L, Tian X, Ji L-N, Zhou J-Y and Li R-H 1997 *Polyhedron* **16** 103.
23. (a) Arounaguiri S and Maiya B G 1999 *Inorg. Chem*. **38** 842; (b) Ambroise A and Maiya B G 2000 *Inorg. Chem*. **39** 4256; (c) Ambroise A and Maiya B G 2000 *Inorg. Chem*. **39** 4264; (d) Kulkarni M S, Rao B S M, Sastri C V, Maiya B G, Mohan H and Mittal J P 2004 *J. Photochem. Photobiol.* **A167** 101; (e) Murali S, Sastri C V and Maiya B G 2002 *Proc. Indian Acad. Sci.* (*Chem. Sci*.) **114** 403
24. Eppley H J, Lato S M, Ellington A D and Zaleski J M 1999 *Chem. Commun*. 2405
25. Marmur J 1961 *J. Mol. Biol*. **3** 208
26. Henderson L J, Fronczek F R and Cherry W R 1984 *J. Am. Chem. Soc*. **106** 5876
27. Kraft B J, Eppley H J, Huffman J C and Zaleski J M 2002 *J. Am. Chem. Soc*. **124** 272
28. Sullivan B P, Salmon D and Meyer T J 1978 *Inorg. Chem*. **17** 3334
29. Wolfe A, Shimer G H and Meehan T 1987 *Biochemistry* **26** 6392
30. Cohen G and Eisenberg H 1969 *Biopolymers* **8** 45
31. Norden B, Lincoln P, Akerman B and Tuite E 1996 In *Metal ions in biological systems* (eds) A Sigel and H Sigel (New York: Marcel Dekker) **33** 177
32. Friedman A E, Kumar C V, Turro N J and Barton J K 1991 *Nucleic Acids Res.* **19** 2595
33. Baguley B C and LeBret M 1984 *Biochemistry* **23** 937
34. Lakowicz J R and Webber G 1973 *Biochemistry* **12** 4161