**Materials**

dGTP was obtained from Pharmacia and Roche and used as received. dGMP was purchased from Sigma. Highly polymerized calf-thymus and herring-testes DNA were purchased from Sigma and sheared by repeated sonication and passage through a 22-gauge needle. Synthetic oligonucleotides were purchased from the Nucleic Acid Core Facility at the Lineberger Comprehensive Cancer Center at the University of North Carolina at Chapel Hill and purified by double ethanol precipitation. Water was purified with a MilliQ purification system (Millipore), and $D_2O$ (99.9 atom %) was purchased from Aldrich. $Na_2HPO_4$, $NaH_2PO_4$, and NaCl were purchased from Mallinckrodt and used without further purification. $Na_2DPO_4$ and $NaD_2PO_4$ were prepared by dissolving $Na_2HPO_4$ and $NaH_2PO_4$ in $D_2O$, followed by evaporation of the solvent. The pH of the sodium phosphate solutions was measured with a standard pH meter, calibrated with $H_2O$ buffers, and for $D_2O$ solutions the relationship pD = pH + 0.4 was used (Perrin, D.D.; Dempsey, B. *Buffers for pH and Metal Ion Control*; Science Paperbacks, Chapman and Hall: London, U.K., 1974.). $RuCl_3 \cdot xH_2O$, 2,2'-bipyridine, and 4,4'-dimethyl-2,2'-bipyridine (dmb) were purchased from Aldrich. $(NH_4)_2OsCl_6$ and $FeSO_4 \cdot 7H_2O$ were obtained from Alfa Aesar. 4,4'-dichloro-2,2'-bipyridine was prepared according to a literature procedure (Wenkert, D.; Woodward, R. B. *J. Org. Chem.* **1983**, *48*, 285-289).

**Metal Complexes**

Literature procedures were followed to prepare polypyridyl complexes of ruthenium(II) (Mabrouk; P. A.; Wrighton, M. S. *Inorg. Chem.* **1986**, *25*, 526-531), iron(II) (DeSimone, R. E.; Drago, R. S. *J. Am. Chem. Soc.* **1970**, *92*, 2343-2352), and osmium(II) (Constable, E. C.; Raithby, P. R., Smit, D. N. *Polyhedron*, **1989**, *8*, 367-369;

Yang, J. C.; Meyer, T. J., unpublished results). Products were characterized by UV/Vis spectrophotometry and electrochemistry, and their purity was checked by thin-layer chromatography (TLC) on aluminum oxide plates. Metal(III) complexes were synthesized by oxidation of the corresponding metal(II) complexes according to a published protocol (DeSimone and Drago, 1970).

**Nucleic Acids**

Nucleic acid concentrations were determined spectrophotometrically using a Hewlett-Packard HP 8452 diode array spectrophotometer. The extinction coefficient used for calf-thymus and herring-testes DNA was $\varepsilon_{260} = 6600\ M^{-1}cm^{-1}$ where concentration of nucleic acid is measured in nucleotide phosphate. The extinction coefficient for dGMP was $\varepsilon_{260} = 13{,}700\ M^{-1}\ cm^{-1}$. Extinction coefficients for oligonucleotides were calculated using the nearest neighbor equation, giving the concentration of nucleic acid in strand concentration.

Solutions of double-stranded oligonucleotide were prepared by mixing two oligonucleotides in the 1:1 ratio in 100 mM sodium phosphate, pH 8 with 800 mM NaCl, heating at 90°C for 5 min, and cooling the mixture to room temperature over a period of 3 h.

**Stopped-flow Spectrophotometry and Global Fitting**

Kinetic experiments were carried out using an On Line Instrument Systems OLIS RSM-1000 stopped-flow system. The reaction was monitored spectrophotometrically at $\lambda_{max}$ of the metal complex in the reduced form($M^{2+}$) ± 230 nm. Solutions were maintained at 25 ± 1°C. The oxidant and DNA were dissolved in ~10 mM $H_2SO_4$ or DCl with 800 mM NaCl (pH 2) and pH 8 phosphate buffer with 800 mM NaCl, respectively,

to give a solution that is 50 mM sodium phosphate, pH 7 with 800 mM NaCl after mixing. Concentrations of $M^{2+}$ and $M^{3+}$ in each run were obtained from $A_{min}$ and $A_{max}$-$A_{min}$, respectively, at the $\lambda_{max}$ for $M^{2+}$. Maximum wavelengths and the extinction coefficients of $M^{2+}$ are given in Table S1, and the substrate and oxidant concentrations along with the length of the run are summarized in Table S2. Second-order oxidation rate constants were determined by global analysis of all the data using the SPECFIT software (Spectrum Software Associates, Chapel Hill). The rate constants for the faster and slower components for each metal complex are given in Tables S3 and S4.

**Cyclic Voltammetry**

Electrochemistry experiments were performed at 25 mV/s with an ITO working electrode, Ag/AgCl reference electrode and a Pt counter electrode. ITO electrodes were washed by sonicating 10 minutes in isopropanol followed by two washes in water (10 minutes each wash). Electrodes were air dried. The samples consisted of 1 mM herring testes DNA or 50 μM dGMP and 50 μM $Ru(bpy)_3^{2+}$ in 50 mM sodium phosphate, 800 mM NaCl (pH, pD = 7.0). ITO electrodes were conditioned for 6 cycles at 25 mV/s in 50 mM sodium phosphate, 800 mM NaCl. A background cyclic voltammogram was obtained of the buffer alone. This background was subtracted from the subsequent Ru only cyclic voltammogram and Ru + DNA cyclic voltammogram. The cyclic voltammograms were fit to a three-step oxidation mechanism using DigiSim software.

**Table S1. Properties of ruthenium, iron, and osmium polypyridyl complexes used in the study.**

| Metal Complex | $E_{1/2}$ (III/II), V | $\lambda_{max}$(II), nm | $\varepsilon_{max}$(II), $M^{-1}cm^{-1}$ |
|---|---|---|---|
| $Os(dmb)_3^{3+/2+}$ | 0.47 | 496 | 13 000 |
| $Os(dmb)(bpy)_2^{3+/2+}$ | 0.55 | 494 | 13 000 |
| $Os(bpy)_3^{3+/2+}$ | 0.61 | 490 | 12 900 |
| $Fe(dmb)_3^{3+/2+}$ | 0.66 | 529 | 8 400 |
| $Fe(bpy)_3^{3+/2+}$ | 0.83 | 522 | 8 600 |
| $Ru(dmb)_3^{3+/2+}$ | 0.86 | 458 | 17 000 |
| $Ru(dmb)_2(bpy)^{3+/2+}$ | 0.93 | 456 | 14 000 |
| $Ru(dmb)(bpy)_2^{3+/2+}$ | 0.98 | 454 | 16 000 |
| $Ru(bpy)_3^{3+/2+}$ | 1.05 | 452 | 14 600 |
| $Ru(bpy)_2(Cl_2\text{-}bpy)^{3+/2+}$ | 1.13 | 448 | 12 600 |

**Table S2. Conditions for stopped-flow spectrophotometry experiments used to monitor oxidation of various DNA substrates by ruthenium, iron, and osmium polypyridyls.**

| Substrate | [Substrate], μM | Oxidant | [Oxidant], μM | Time, sec |
|---|---|---|---|---|
| dGTP | 100 | $Fe(bpy)_3^{3+}$ | 27-34 | 240 |
| dGTP | 50 | $Ru(dmb)_3^{3+}$ | 3-11 | 120 |
| dGTP | 25, 50 | $Ru(dmb)_2(bpy)^{3+}$ | 8-22 | 10-20 |
| dGTP | 25, 50 | $Ru(dmb)(bpy)_2^{3+}$ | 16-23 | 1-2 |
| dGTP | 25, 50 | $Ru(bpy)_3^{3+}$ | 13-35 | 1-2 |
| dGTP | 13 | $Ru(bpy)_2(Cl_2\text{-}bpy)^{3+}$ | 7-16 | 0.1 |
| ss oligo[a] | 200[c] | $Fe(bpy)_3^{3+}$ | 30-58 | 300 |
| ss oligo[a] | 200[c] | $Ru(dmb)_3^{3+}$ | 5-15 | 300 |
| ss oligo[a] | 80[c] | $Ru(dmb)_2(bpy)^{3+}$ | 10-17 | 45-60 |
| ss oligo[a] | 80[c] | $Ru(dmb)(bpy)_2^{3+}$ | 8-20 | 10 |
| ss oligo[a] | 80[c] | $Ru(bpy)_3^{3+}$ | 20-36 | 0.5-1 |
| ds oligo[b] | 300[c] | $Fe(bpy)_3^{3+}$ | 18-25 | 300 |
| ds oligo[b] | 300[c] | $Ru(dmb)_3^{3+}$ | 7-10 | 60 |
| ds oligo[b] | 120[c] | $Ru(dmb)_2(bpy)^{3+}$ | 11-22 | 120 |
| ds oligo[b] | 120[c] | $Ru(dmb)(bpy)_2^{3+}$ | 25-31 | 20 |
| ds oligo[b] | 120[c] | $Ru(bpy)_3^{3+}$ | 28-35 | 1 |
| ht DNA | 750[c] | $Fe(bpy)_3^{3+}$ | 27-31 | 900 |
| ht DNA | 750[c] | $Ru(dmb)_3^{3+}$ | 7-19 | 900 |
| ht DNA | 750[c] | $Ru(dmb)_2(bpy)^{3+}$ | 14-28 | 180 |
| ht DNA | 750[c] | $Ru(dmb)(bpy)_2^{3+}$ | 18-30 | 30 |

**Table S2.** (continued)

| | | | | |
|---|---|---|---|---|
| ht DNA | 500, 750[c] | $Ru(bpy)_3^{3+}$ | 20-46 | 1-2 |
| ct DNA | 400, 660[c] | $Fe(bpy)_3^{3+}$ | 14-45 | 600-900 |
| ct DNA | 520[c] | $Ru(dmb)_3^{3+}$ | 2-6 | 30 |
| ct DNA | 340[c] | $Ru(dmb)_2(bpy)^{3+}$ | 12-21 | 180 |
| ct DNA | 340[c] | $Ru(dmb)(bpy)_2^{3+}$ | 9-25 | 30-45 |
| ct DNA | 400, 600[c] | $Ru(bpy)_3^{3+}$ | 22-35 | 1-2 |

[a] Sequence: 5'-GCA GTA GCA TGT GAC GAG TCG-3'
[b] Sequence: 5'-GCA GTA GCA TGT GAC GAG TCG hybridized to 5'-CGA CTC GTC ACA TGC TAC TGC-3'.
[c] Guanine concentration.

**Table S3. Second-order rate constants for the faster reaction component for guanine oxidation in different environments.**

| Oxidant | Rate constant, $M^{-1}sec^{-1}$ | | | | |
|---|---|---|---|---|---|
| | dGTP | ss oligo | ds oligo | ht DNA | ct DNA |
| $Fe(bpy)_3^{3+}$ | $(3.6 \pm 0.30)\ 10^3$ | $(1.0 \pm 0.050)\ 10^3$ | $460 \pm 11$ | $62 \pm 5.1$ | $160 \pm 24$ |
| $Ru(dmb)_3^{3+}$ | $(8.2 \pm 0.60)\ 10^3$ | $(1.3 \pm 0.058)\ 10^3$ | $390 \pm 40$ | $61 \pm 9.1$ | $(1.8 \pm 0.13)\ 10^3$ |
| $Ru(dmb)_2(bpy)^{3+}$ | $(2.2 \pm 0.40)\ 10^5$ | $(1.5 \pm 0.20)\ 10^4$ | $(3.4 \pm 0.20)\ 10^3$ | $400 \pm 28$ | $(1.0 \pm 0.29)\ 10^3$ |
| $Ru(dmb)(bpy)_2^{3+}$ | $(1.3 \pm 0.20)\ 10^6$ | $(6.7 \pm 1.2)\ 10^4$ | $(3.6 \pm 0.20)\ 10^4$ | $(2.3 \pm 0.082)\ 10^3$ | $(5.5 \pm 0.89)\ 10^3$ |
| $Ru(bpy)_3^{3+}$ | $(5.4 \pm 0.30)\ 10^6$ | $(8.0 \pm 0.50)\ 10^5$ | $(2.5 \pm 0.90)\ 10^5$ | $(3.4 \pm 0.080)\ 10^4$ | $(2.6 \pm 0.7)\ 10^4$ |
| $Ru(bpy)_2(Cl_2\text{-}bpy)^{3+}$ | $(3.2 \pm 0.30)\ 10^7$ | -[a] | -[a] | -[a] | -[a] |

[a] Rate not measured.

**Table S4. Second-order rate constants for the slower reaction component for oxidation of guanine in different environments.**

| Oxidant | Rate constant, $M^{-1}sec^{-1}$ | | | | |
|---|---|---|---|---|---|
| | dGTP | ss oligo | ds oligo | ht DNA | ct DNA |
| $Fe(bpy)_3^{3+}$ | 320 ± 40 | 110 ± 3.5 | 41 ± 0.60 | 7.9 ± 0.32 | 14 ± 0.70 |
| $Ru(dmb)_3^{3+}$ | 720 ± 50 | 130 ± 9.6 | -[a] | 7.2 ± 1.3 | 140 ± 40 |
| $Ru(dmb)_2(bpy)^{3+}$ | (2.2 ± 0.50) $10^4$ | (1.8 ± 0.20) $10^3$ | 370 ± 10 | 42 ± 4.9 | 69 ± 17 |
| $Ru(dmb)(bpy)_2^{3+}$ | (2.0 ± 0.7) $10^5$ | (5.3 ± 1.2) $10^3$ | (3.0 ± 0.30) $10^3$ | 180 ± 34 | 410 ± 45 |
| $Ru(bpy)_3^{3+}$ | (3.1±0.4) $10^5$ | (2.5 ± 0.40) $10^5$ | (4.6 ± 1.4) $10^4$ | (5.5 ± 0.30) $10^3$ | (3.3 ± 0.60) $10^3$ |
| $Ru(bpy)_2(Cl_2\text{-}bpy)^{3+}$ | (8.7 ± 0.30) $10^6$ | -[b] | -[b] | -[b] | -[b] |

[a] Fit to one-population model.
[b] Rate not measured.