Supporting Information

Conjugation of organic-metallic hybrid polymers and calf-thymus DNA

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Table of contents

1. Absorption spectra of MEPEs in the presence of calf-thymus DNA

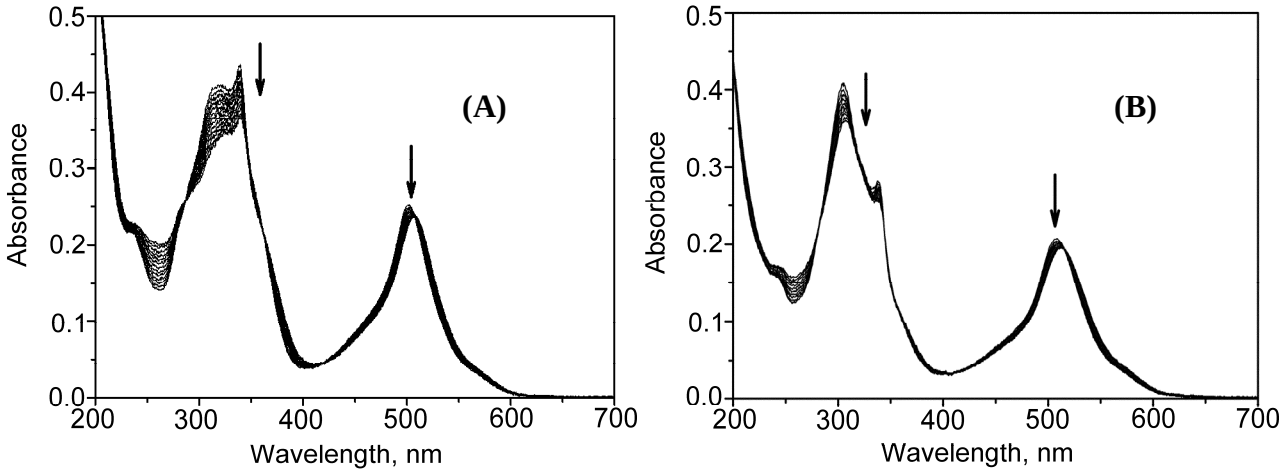

Figure S1. Absorption spectra of (A) RuL1-MEPE (7 μM) and (B) RuL2-MEPE (7 μM) in the absence (top curve) and presence (subsequent curves) of increasing concentrations of ct-DNA (up to 7 μM).

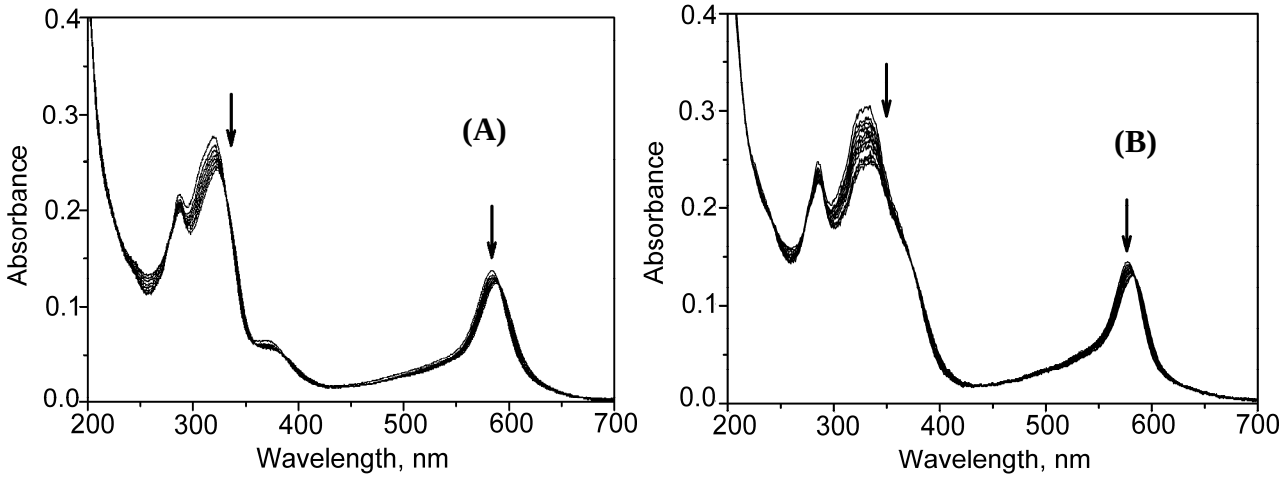

Figure S2. Absorption spectra of (A) FeL3-MEPE (7 μM) and (B) FeL4-MEPE (7 μM) in the absence (top curve) and presence (subsequent curves) of increasing concentrations of ct-DNA (up to 7 μM).

2. Determination of binding constant, K_b

Absorption spectra were recorded in H_2O at 30ºC by using a Shimadzu UV-2550 UV-visible spectrophotometer.

The concentration of ct-DNA per nucleotide was calculated from its known extinction coefficient at 260 nm (6600 $_M{}^{-1}cm^{-1}$, sheared, 200-500 base pairs) and the concentration of MEPE is expressed in repeating unit. The solution of ct-DNA in the buffer gave a ratio of UV absorbance of about 1.8-1.9:1 at 260 and 280 nm, indicating that the ct-DNA was sufficiently free of protein.

The binding constants of polymers to ct-DNA were determined by absorption titrations at room temperature in aqueous solution. The dilution of MEPEs at the end of each titration was negligible. The DNA binding constant, K_b, of polymers was determined from fit of the changes of the absorption of the polymer as a function of ct-DNA concentration of equation S1,

$$\frac{\varepsilon_a - \varepsilon_f}{\varepsilon_b - \varepsilon_f} = \frac{b-(b^2-2K_b{}^2C_t[DNA]_t / s)^{1/2}}{2K_bC_t} \qquad \text{(eqn S1)}$$

Where $b = 1+K_bC_t+K_b[DNA]_t / 2s$, K_b is binding constant between polymer complex and ct-DNA, C_t represents the total polymer concentration in repeating units, $[DNA]_t$ is the DNA concentration in nucleotides, s is the size of the binding site in nucleotide and ε_a, ε_f, ε_b represent the apparent, free and bound polymer molar extinction coefficients, respectively. The value of ε_b was determined from the plateau of DNA titration, where addition of DNA did not result in further changes to the absorption spectrum.

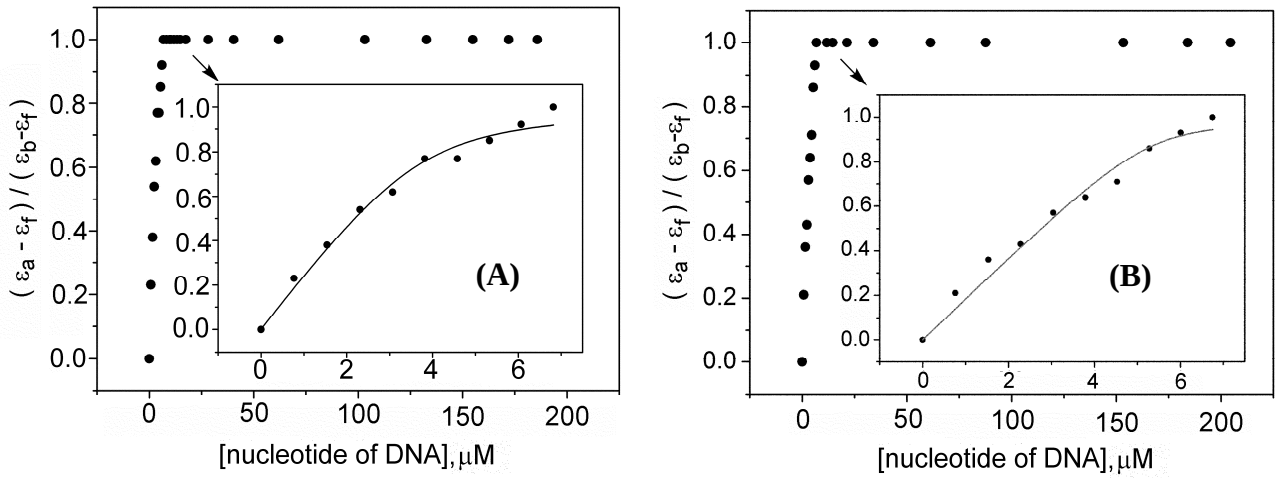

Figure S3. Plot of the titration of (A) RuL1-MEPE (K_b= 1.7×10^6 M^{-1}, s = 0.26) and (B) RuL2-MEPE (K_b= 7.3×10^6 M^{-1}, s = 0.38) with ct-DNA, monitoring the electronic transition of MLCT band at 508 nm and 502 nm, respectively.

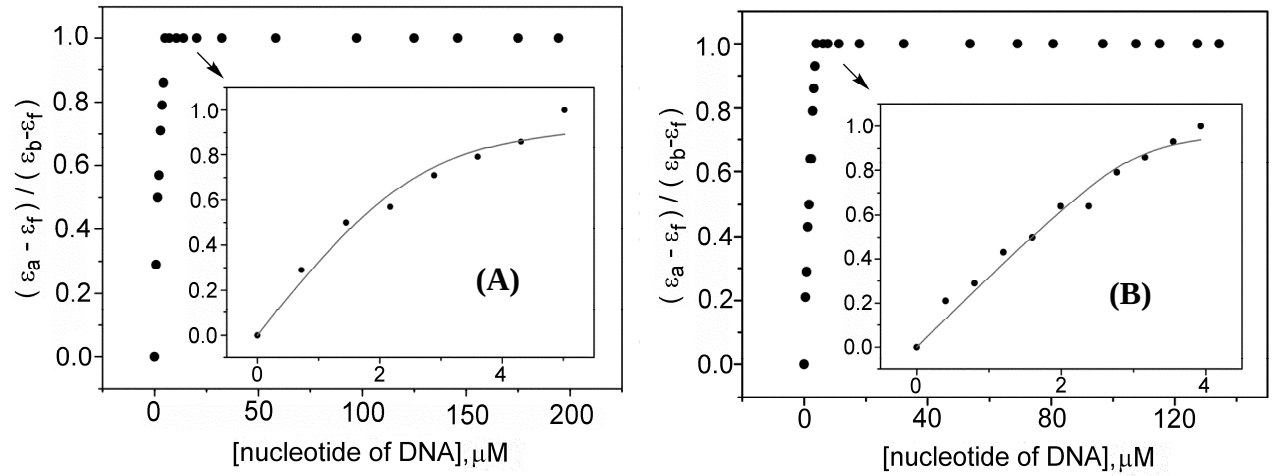

Figure S4. Plot of the titration of (A) FeL3-MEPE (K_b= 1.1×10^6 M^{-1}, s = 0.19) and (B) FeL4-MEPE (K_b= 6.1×10^6 M^{-1}, s = 0.22) with ct-DNA, monitoring the electronic transition of MLCT band at 582 nm and 577 nm, respectively.

Table S1. Absorption titration experiment

MEPEs	Hypochromicity (%)	Isobestic point (nm)	Red shift (nm)	K_b ($\times 10^6$ M^{-1})	S
RuL1-MEPE	6	283, 434, 514	6.5	1.72	0.26
RuL2-MEPE	6	286, 422, 507	6.5	7.26	0.38
RuL3-MEPE	14	275, 420, 516	9.5	4.05	0.43
FeL3-MEPE	10	277, 449, 590	7	1.14	0.19
FeL4-MEPE	9.7	271, 455, 584	6.5	6.11	0.22

3 Emission spectra of RuL3-MEPE in the presence of calf-thymus DNA

Luminescent experiments at 30ºC were recorded in H_2O using a Shimadzu RF-5300PC spectrofluorophotometer.

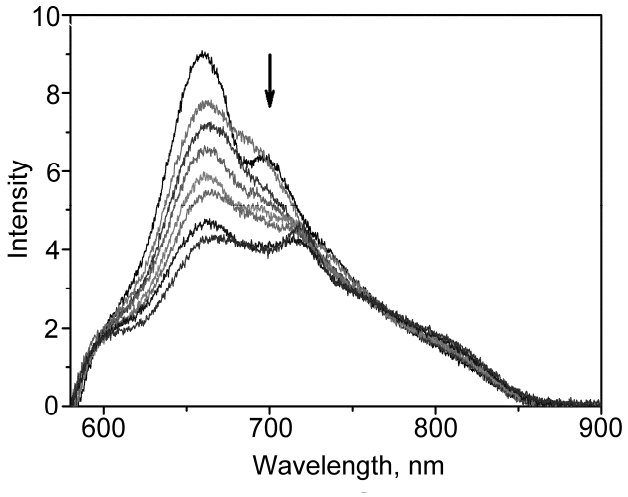

Figure S5. Emission spectra of RuL3-MEPE (7 μM) in the absence (top curve) and presence (subsequent curves) of increasing concentrations of ct-DNA (0-7.46 μM).

4. Circular dichroic spectra

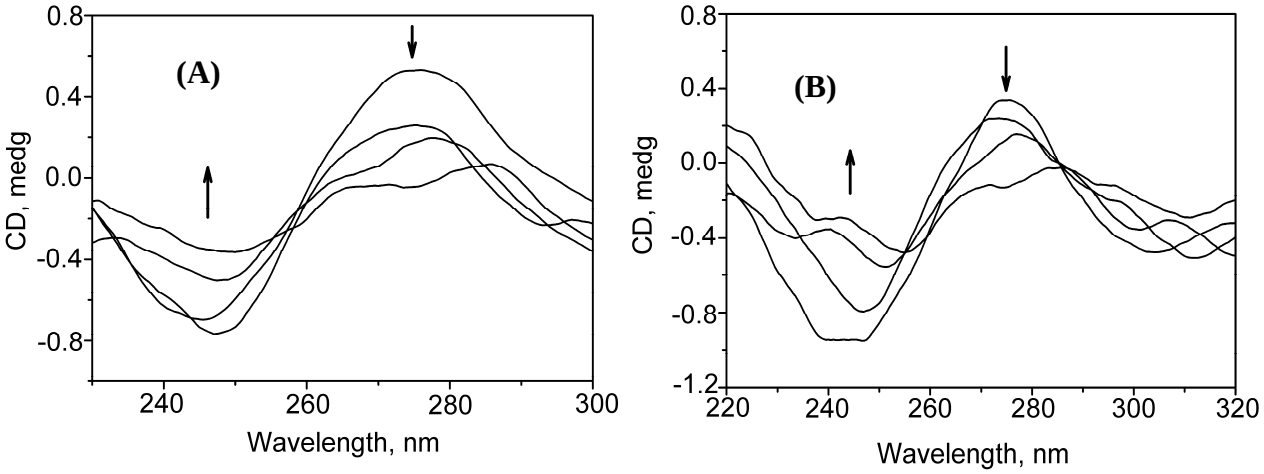

Figure S6. CD spectra of ct-DNA (7.5 μM) in the absence (top curve) and presence (subsequent curves) of increasing concentrations of (A) RuL1-MEPE and (B) RuL3-MEPE.

Circular dichroic spectrum of DNA in the absence and presence of polymers was recorded with a J-820 spectropolarimeter (Jasco) in H_2O at 30ºC in a quartz cuvette with 1 cm path length.

5. Cyclic voltammograms

Cyclic voltammetry (CV) studies were performed with an electrochemical analyzer, ALS/H CH Instruments, at room temperature at a scan rate of 100 mV s^{-1}. CV experiments were carried out in 30 mL three-electrode electrolytic cell in connection with a glassy carbon working electrode (GCE), Ag/AgCl electrode as reference electrode and a platinum wire as counter electrode. Supporting electrolytes were 0.1 M tetra-*n*-butylammonium perchlorate (TBAP) in argon saturated anhydrous acetonitrile.

CV can be used to calculate the corresponding equilibrium constant for the redox process. The oxidized and reduced forms are associated with a third species (DNA) in the solution using the following equation: $E_b^{o'} - E_f^{o'} = 0.059\log(K_{II} / K_{III})$ Where $E_b^{o'}$ and $E_f^{o'}$ are the formal potentials of M(III)/M(II) couple in the bound and free forms, respectively. K_{II} and K_{III} are the binding constants for the binding of the oxidized and reduced forms to DNA.

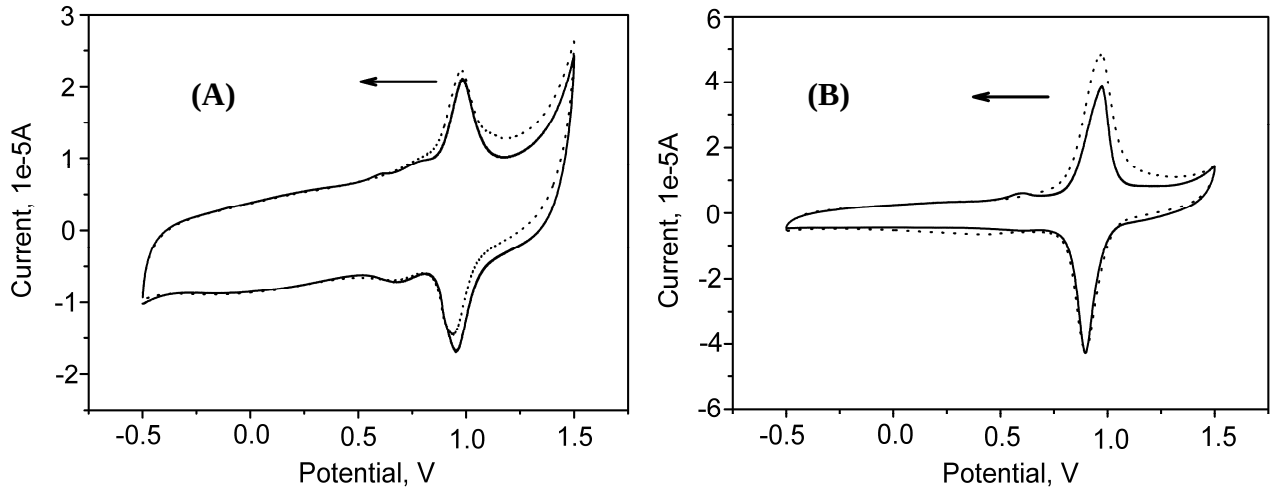

Figure S7. CV spectra of RuL1-MEPE and RuL3-MEPE in the absence (line curve) and presence (dot curve) of ct-DNA.

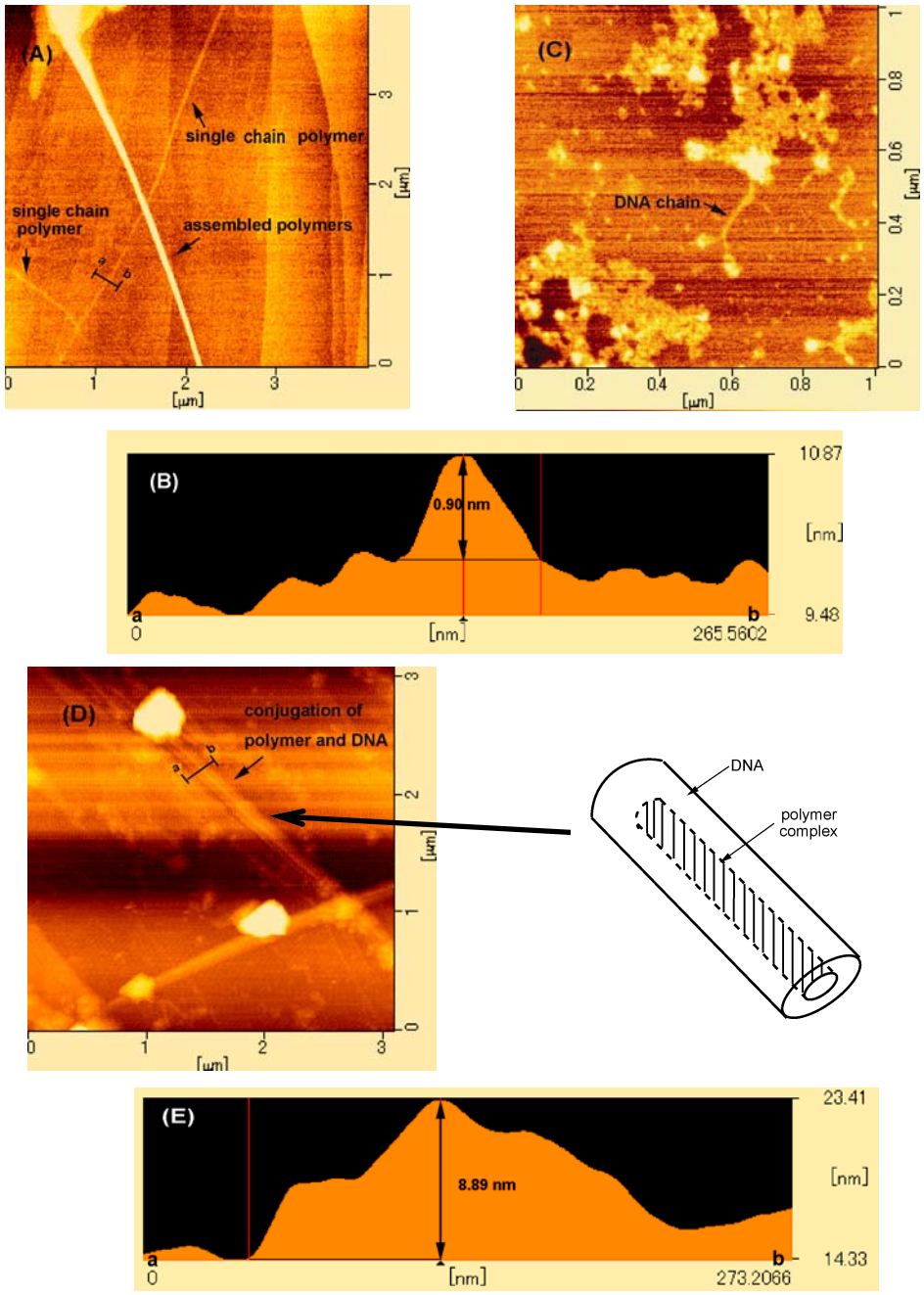

Figure S8. (A) AFM images of single-chain FeL3-MEPE. (B) Cross section of a – b in Figure S8A with an average height ca. 0.90 nm on HOPG in MeOH. (C) Calf-thymus DNA (ct-DNA, sheared, 200-500 base pairs) on mica. (D) FeL3-MEPE incubated with DNA. (E) Cross section of a – b in Figure S8D with an average height ca. 8.89 nm on HOPG.

6. Atomic force microscope images

AFM images were recorded on the DFM mode under air at room temperature (SII DNF L-Trace controlled with SI-DF40P, NanoNavi Technology Inc.). The DNA samples for free absorption were prepared by dropping a portion of 10 μl DNA solutions onto the fleshly-cleaved mica, leaving it static for 30 min for the absorption process. The residues of the solution were gently blown off, then rinsed with double distilled water to remove the remnants of the salts, and dried by the nitrogen before AFM imaging. The MEPE sample was prepared on the fleshly-cleaved high oriented pyrolytic graphite (HOPG) surface by spin coating. The sample substrate was also rinsed gently using distilled water before AFM observation.

7. Electronic structure calculations

Structure of DNA is represented by 10-base-pair oligonucleotide with 5'-TGAGCTCCAA-3' sequence of one strand, the latter one is complement according to Watson-Crick base-pairing. Standard parameters for B-DNA were used for creation of initial geometry and consequently the structure was relaxed by 500 ps MD run in water solution (TIP3P model for water molecules[11c] was used and DNA was parameterized by Amber FF96,[11a] simulation was performed at temperature 300 K). Na^+ cations were placed in vicinity of charged phosphate groups in order to have electrostatically neutral system.

Initial structures of ligand L1-L4 were optimized by DFT (B3LYP) / 6-31G (d) method and then parameterized by GAFF with RESP atom charges fitted from DFT (B3LYP) / 6-31++G (d, p) electron density. After connecting organic ligands by Ru^{2+} QM/MM methodology was used to avoid parameterization of Ru^{2+} and its interaction with nearest atoms. During all QM/MM calculations RI-DFT (BLYP) / def2-SV (P) method with pseudopotential for 28 core electrons on Ru^{2+} was used. QM part therefore consists of Ru^{2+} and surrounding six pyridine cycles, rest of the model was treated by MM. Link atom formalism was used for description of boundary region. Accuracy of this model was checked by geometry optimization of L3-Ru-L3 dimer and comparison of geometry parameters available. RMSD between calculated and measured distances is 0.031 Å and 0.40 deg for angles.

Several initial models were created for each type of ligand (L1-L4) interacting with DNA and their total energy evaluated. After that we selected three energetically lowest structures for each type of ligand so that they represent interaction with sugar-phosphate backbone both from major groove and minor groove of the helix. These models were heated to 300 K by QM/MM

MD and after that again re-optimized. During this procedure structure of polymer relaxed and fitted to geometry of DNA.

Electrostatic interaction of Ru^{2+} with two phosphate group in minor groove was found as most preferable in gas phase calculations without solution with association energies ca. 270 kcal/mol (varying about ±20 kcal/mol for different models) while for interaction with one phosphate only the value ca. 200 kcal/mol (±20 kcal/mol) was obtained. However, these values are overestimated because of lack of solvent screening of electrostatic interaction. In water solution the phosphate groups are surrounded by water molecules which also fill both grooves and therefore interaction of one Ru^{2+} with phosphates from both backbones is difficult and improbable.