Electronic Supplementary Material (ESI) for RSC Advances.

An Efficient Route to Asymmetrically Diconjugated tris(heteroleptic) Complexes of Ru(II)

Christopher S. Burke[a] and Tia E. Keyes[a]

[a] School of Chemical Sciences, Dublin City University, Dublin 9, Co. Dublin, Ireland.

Supplementary Information

Structural characterisation of the Ru(II) complexes and conjugates

[Ru(dppz)(DMSO)$_2$Cl$_2$] (1)

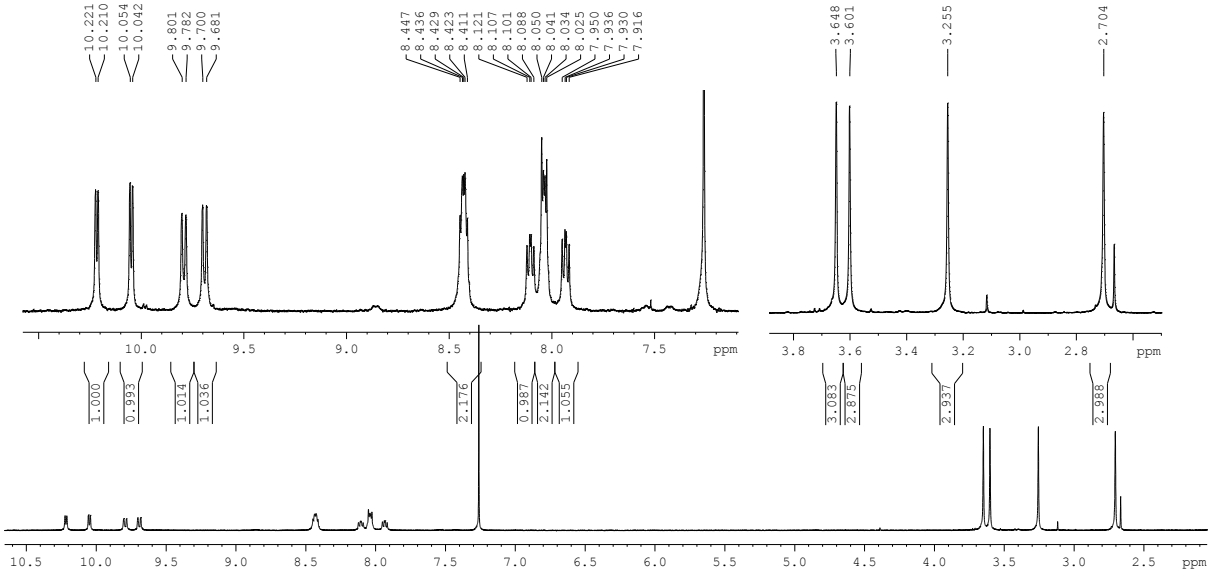


Figure S1 – ^{1}H NMR Spectrum (400 MHz) of **(1)** in $CDCl_3$.

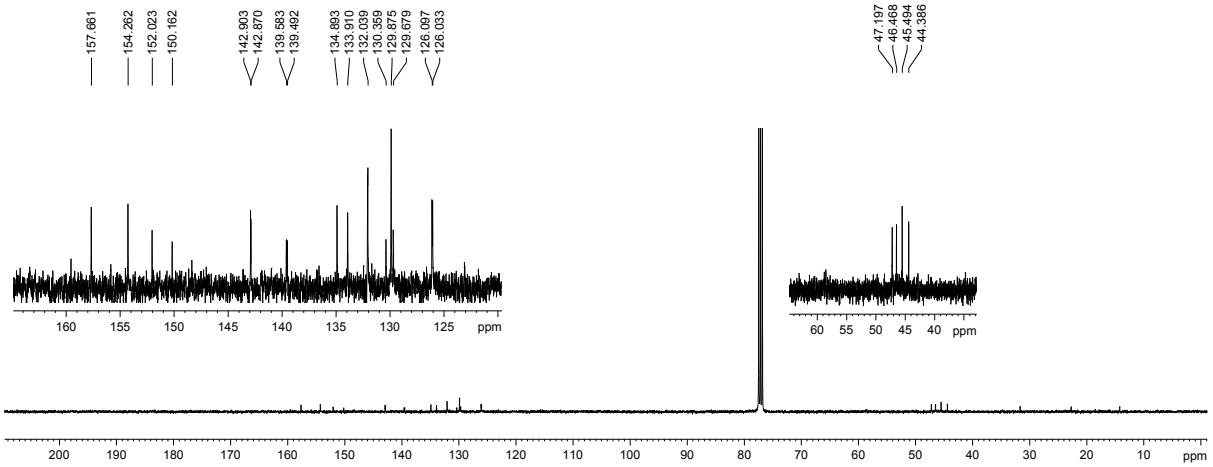


Figure S2 – ^{13}C NMR Spectrum of **(1)** in $CDCl_3$.

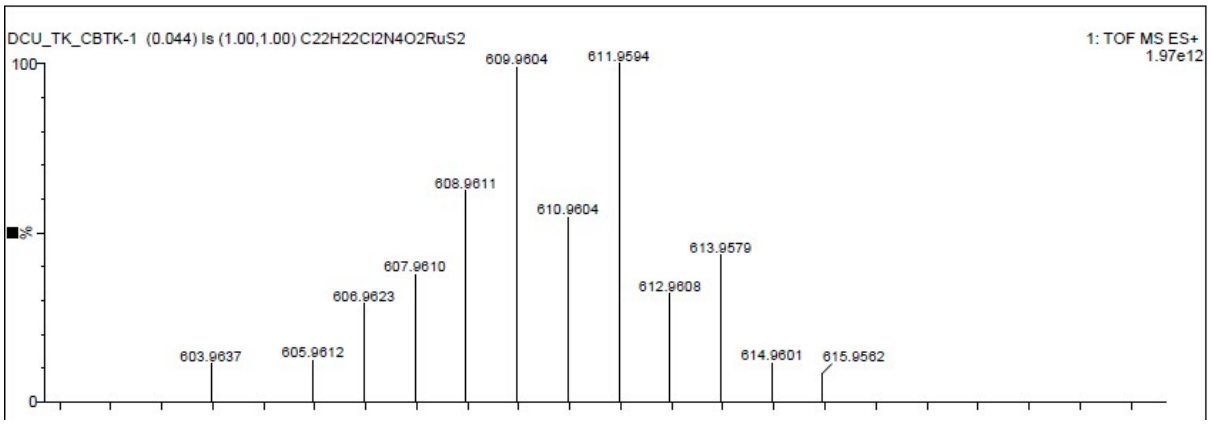


Figure S3 – HR-MS (ESI-QTOF): Single Mass Analysis of **(1)** indicating $[M]^+$.

[Ru(dppz)(bpy)(ox)] (2)

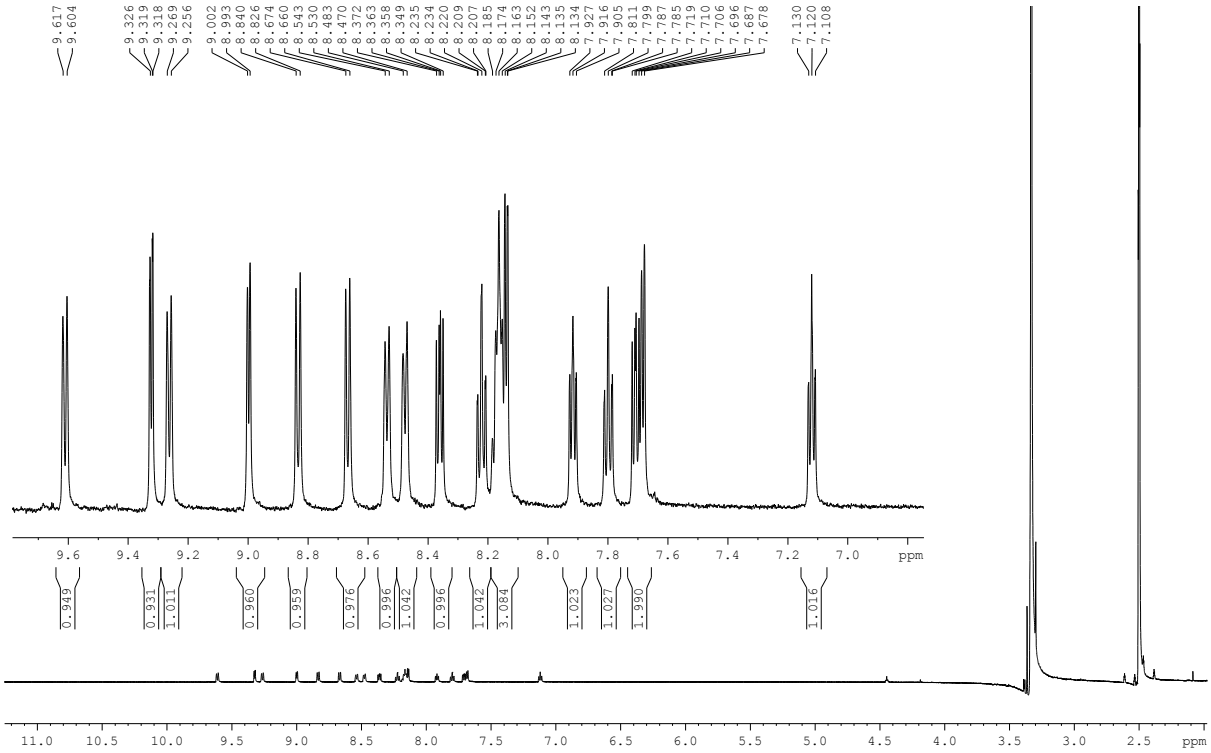
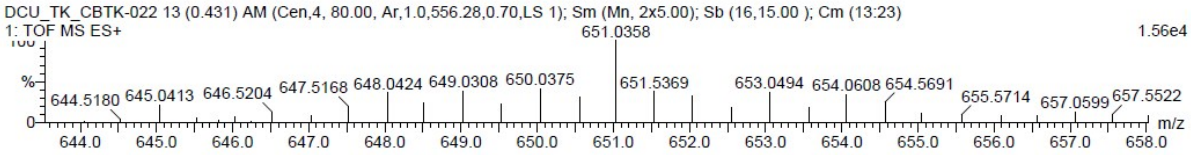


Figure S4 – ^{1}H NMR Spectrum (400 MHz) of **(2)** in DMSO-d_6.

Single Mass Analysis
Tolerance = 5.0 PPM / DBE: min = -1.5, max = 1000.0
Isotope cluster parameters: Separation = 1.0 Abundance = 1.0%

Monoisotopic Mass, Odd and Even Electron Ions
106 formula(e) evaluated with 1 results within limits (all results (up to 1000) for each mass)



Minimum:				-1.5		
Maximum:		200.0	5.0	1000.0		
Mass	Calc. Mass	mDa	PPM	DBE	Score	Formula
651.0358	651.0331	2.7	4.2	24.5	1	C30 H18 N6 O4 Na Ru

Figure S5 – HR-MS (ESI-QTOF): Single Mass Analysis of **(2)** indicating $[M + Na]^+$.

[Ru(dppz)(bpyArCOOH)(ox)] (3)

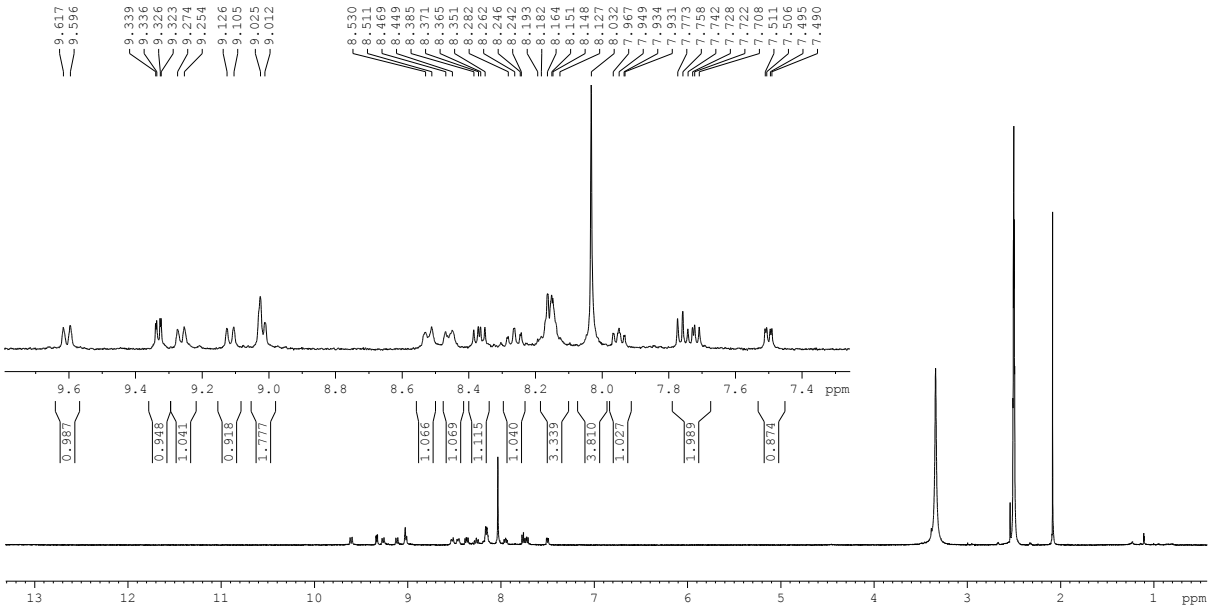


Figure S6 – ^1H NMR Spectrum (400 MHz) of **(3)** in DMSO-d_6.

[Ru(dppz)(bpy)(bpyArCOOEt)](PF$_6$)$_2$ (4)

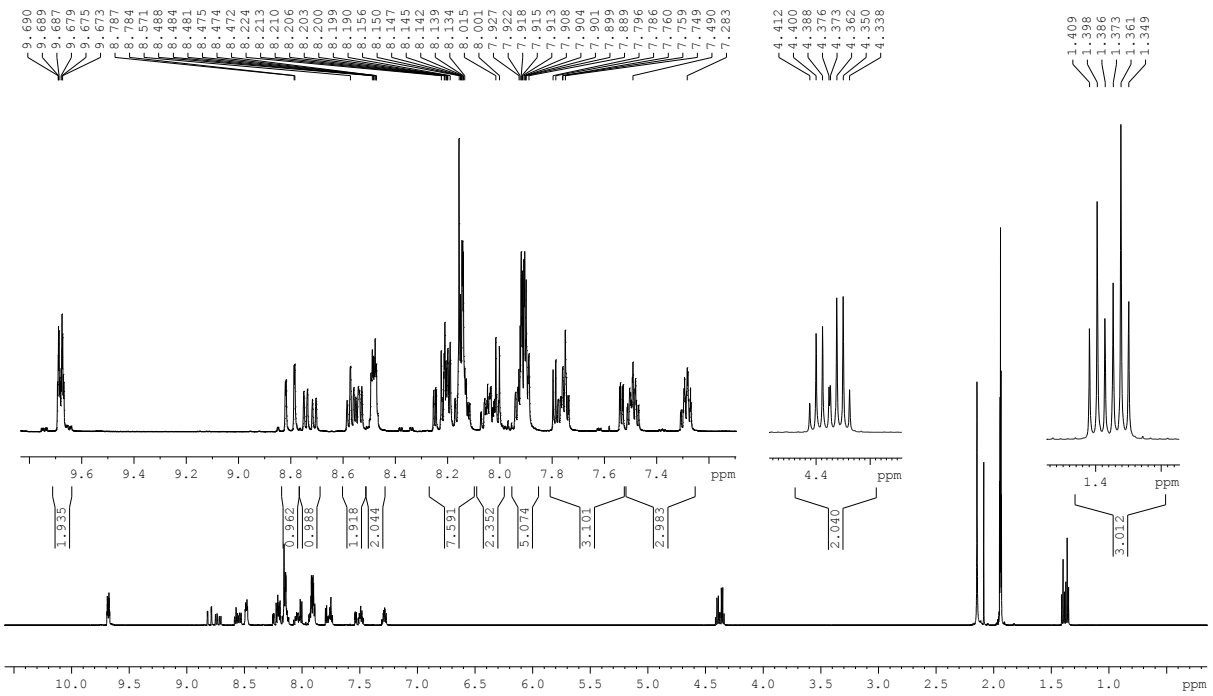


Figure S7 – ^{1}H NMR Spectrum (600 MHz) of **(4)** in CD_3CN.

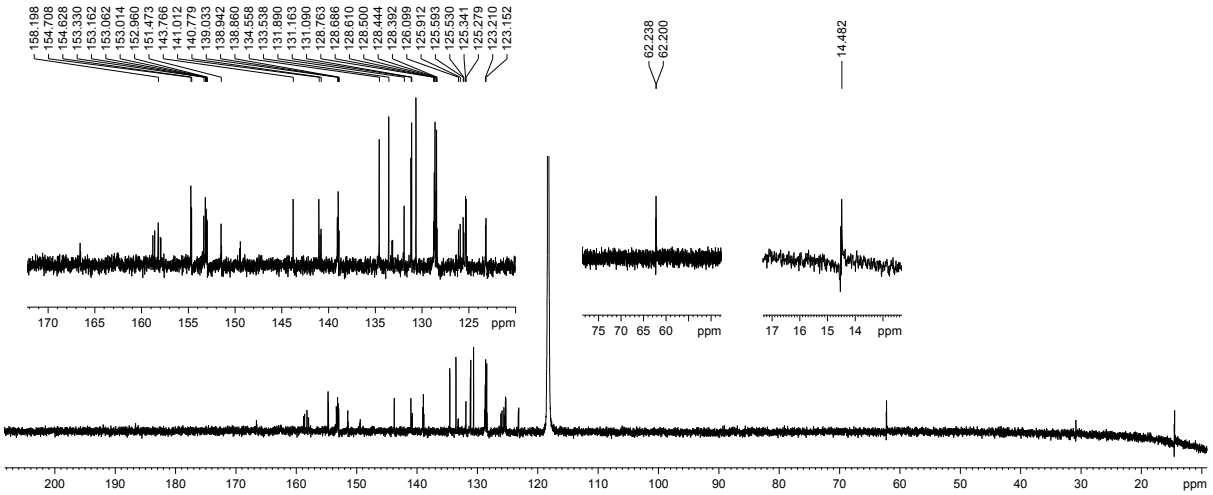


Figure S8 – ^{13}C NMR Spectrum of **(4)** in CD_3CN.

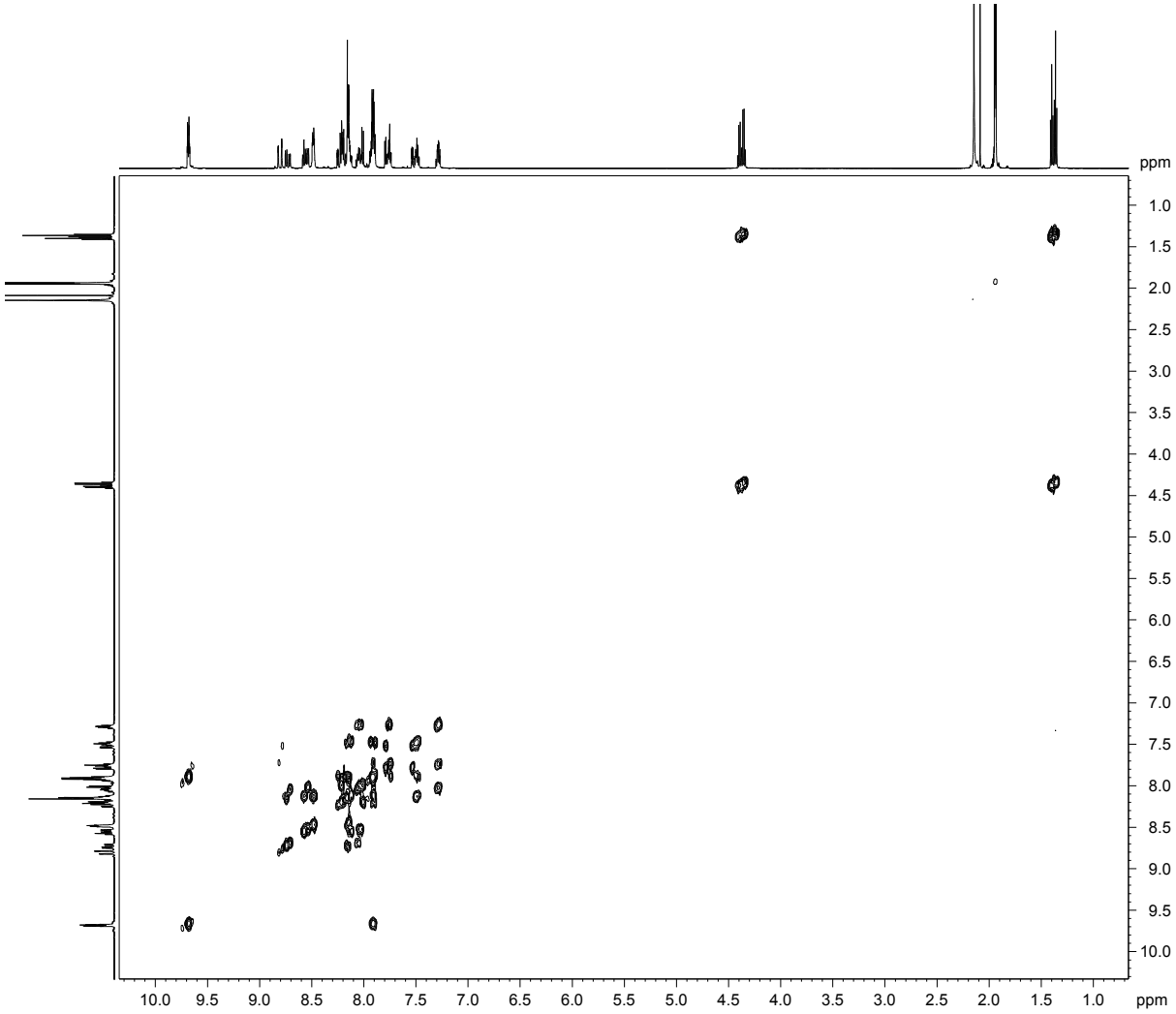
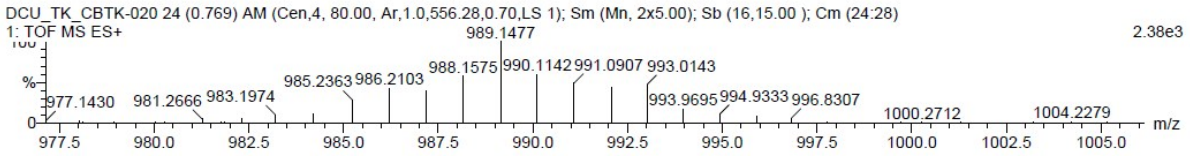


Figure S9 – COSY Spectrum (600 MHz) of **(4)** in CD_3CN.

Single Mass Analysis
Tolerance = 5.0 PPM / DBE: min = -1.5, max = 1000.0
Isotope cluster parameters: Separation = 1.0 Abundance = 1.0%

Monoisotopic Mass, Odd and Even Electron Ions
392 formula(e) evaluated with 1 results within limits (all results (up to 1000) for each mass)



Minimum:				-1.5		
Maximum:		200.0	5.0	1000.0		
Mass	Calc. Mass	mDa	PPM	DBE	Score	Formula
989.1477	989.1490	-1.3	-1.3	32.5	1	C47 H34 N8 O2 Ru F6 P

Figure S10 – HRMS (ESI-QTOF): Single Mass Analysis of **(4)** indicating $[M - PF_6^-]^+$.

[Ru(dppz)(bpy)(bpyArCOOH)]$(PF_6)_2$ (5)

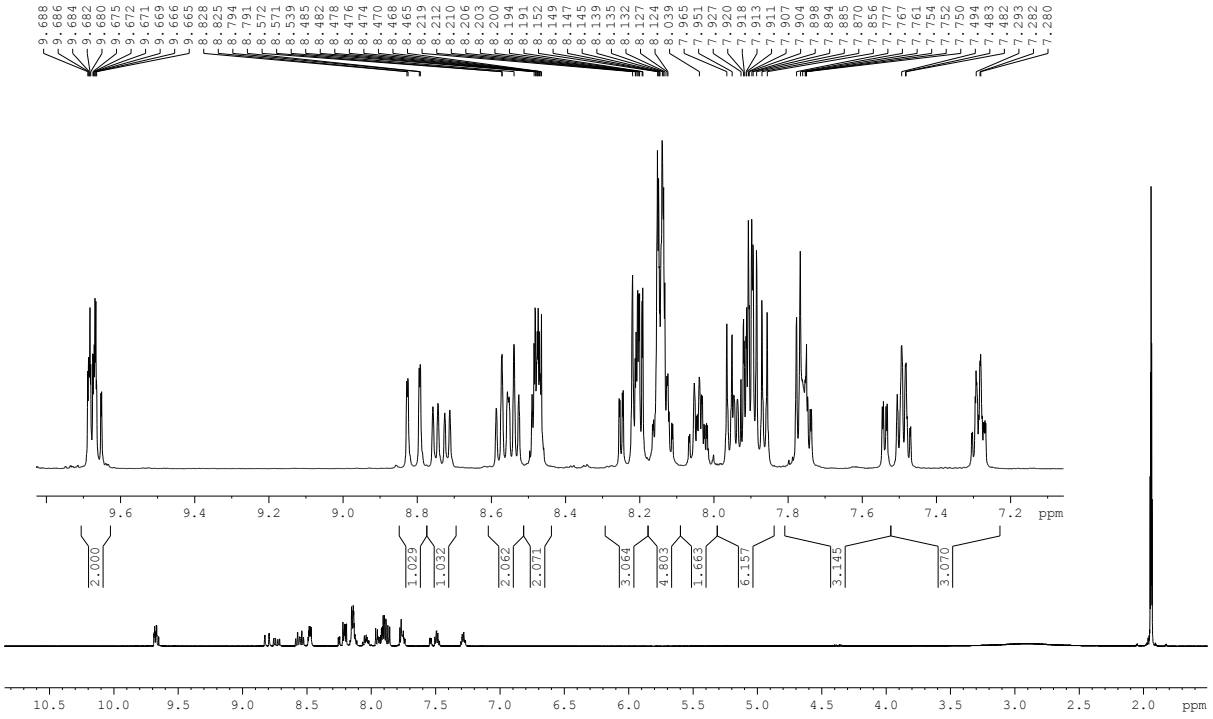


Figure S11 – ^{1}H NMR Spectrum (600 MHz) of **(5)** in CD_3CN.

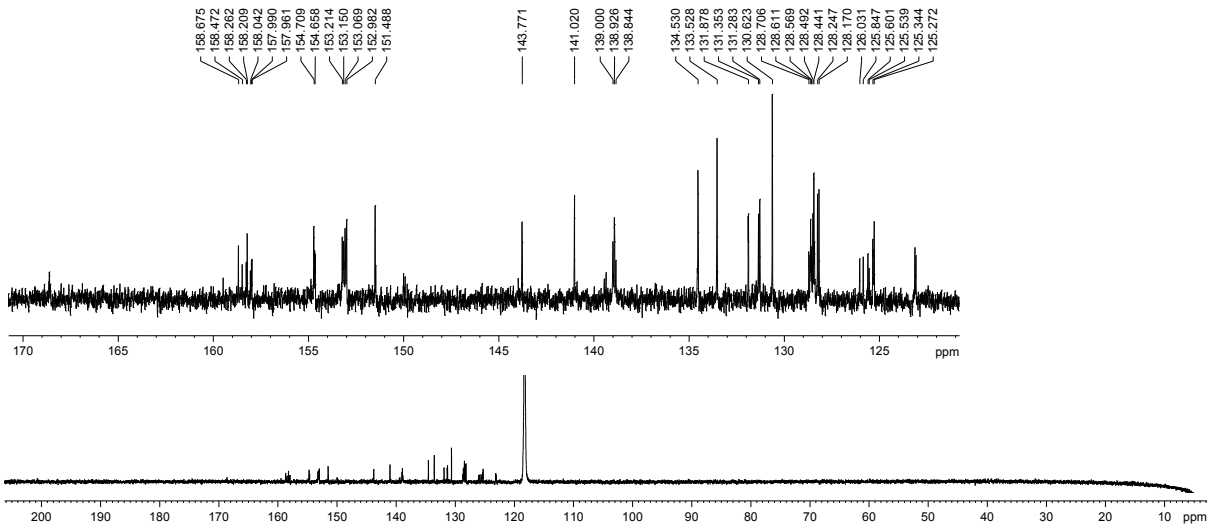


Figure S12 – ^{13}C NMR Spectrum of **(5)** in CD_3CN.

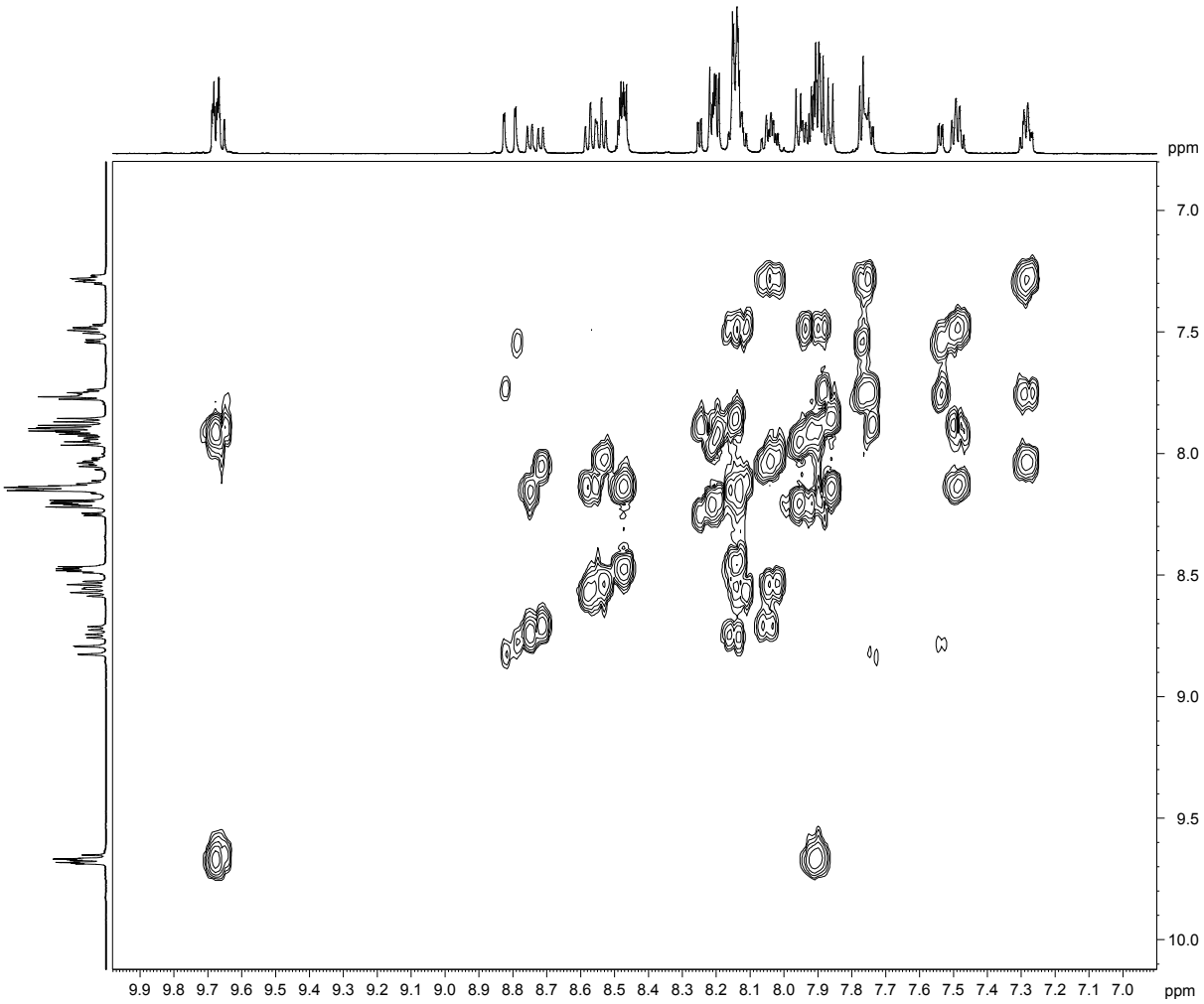


Figure S13 – COSY NMR Spectrum of **(5)** in CD_3CN (aromatic region).

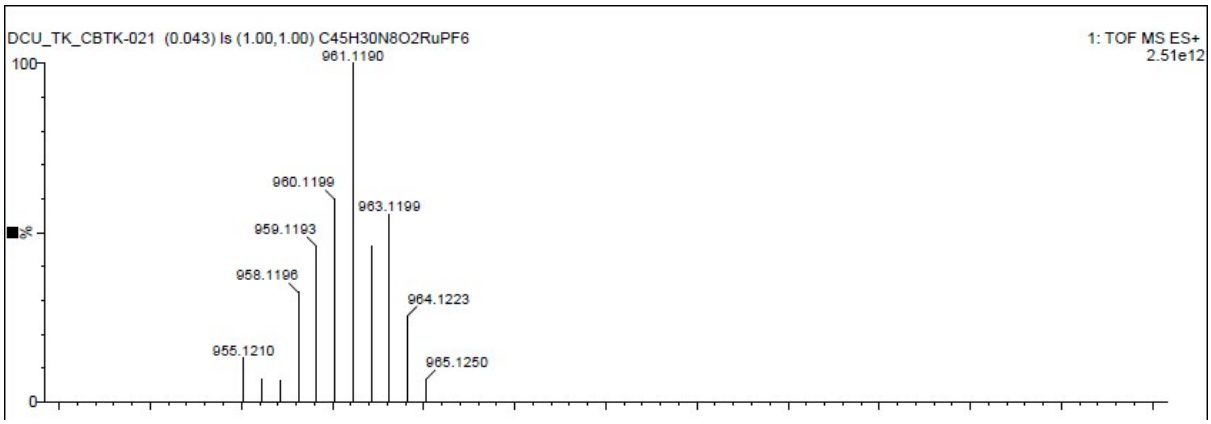


Figure S14 – HRMS (ESI-QTOF): Mass spectrum of **(5)** indicating $[M - PF_6^-]^+$.

[Ru(dppz)(bpyArCOOH)(bpyArCOOEt)]$(PF_6)_2$ (6)

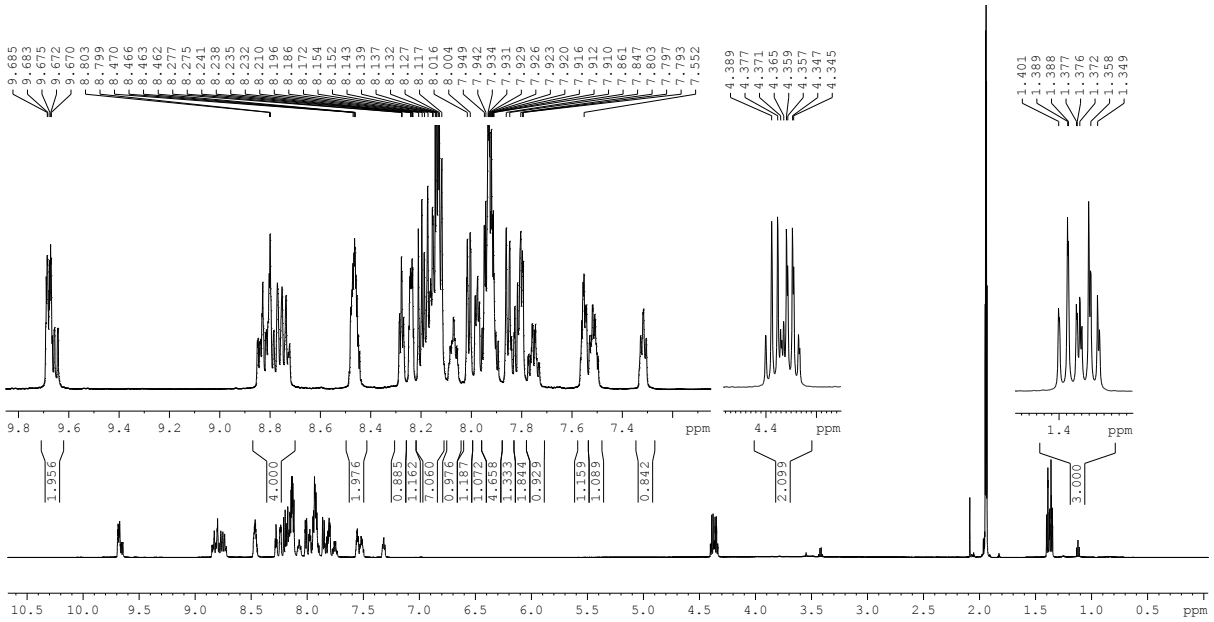


Figure S15 – ^{1}H NMR Spectrum (600 MHz) of **(6)** in CD_3CN. Peaks at 1.1 and 3.4 are assigned to residual diethyl ether.

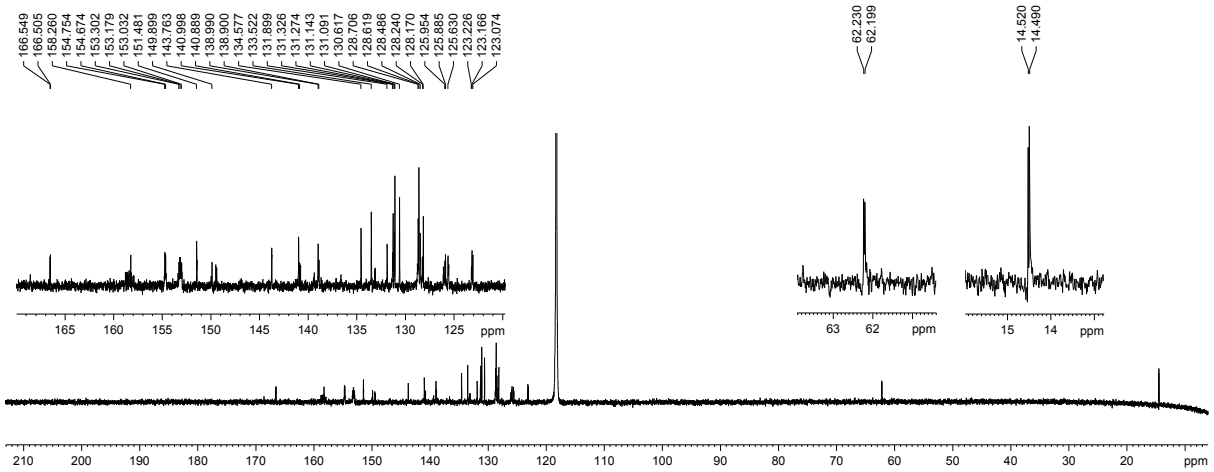


Figure S16 – ^{13}C NMR Spectrum of **(6)** in CD_3CN.

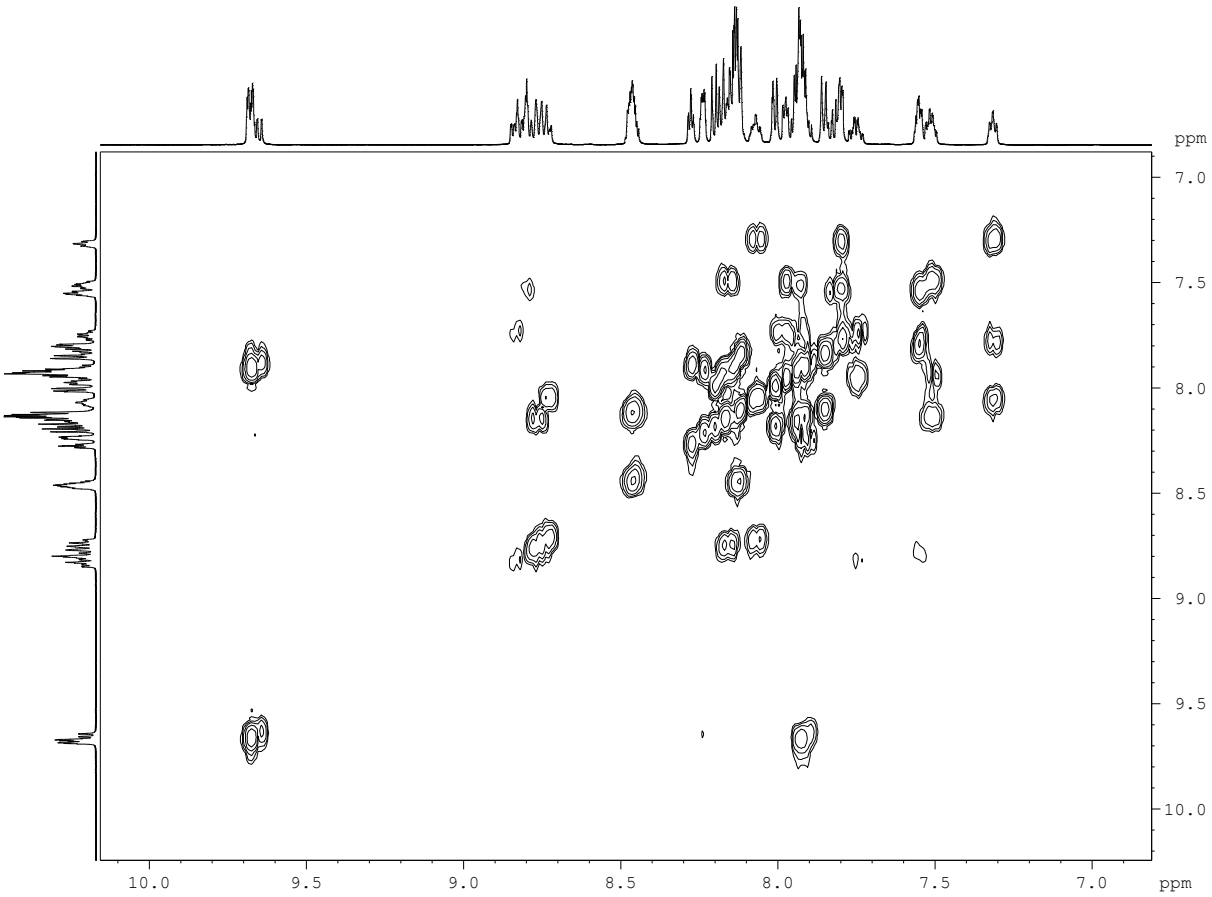
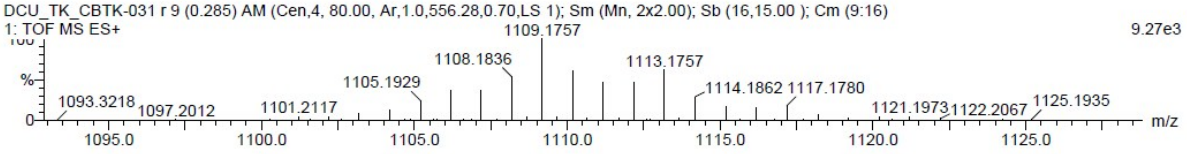


Figure S17 – COSY NMR Spectrum (600 MHz) of **(6)** in CD_3CN (aromatic region).

Single Mass Analysis
Tolerance = 6.0 PPM / DBE: min = -1.5, max = 1000.0
Isotope cluster parameters: Separation = 1.0 Abundance = 1.0%

Monoisotopic Mass, Odd and Even Electron Ions
459 formula(e) evaluated with 1 results within limits (all results (up to 1000) for each mass)

DCU_TK_CBTK-031 r 9 (0.285) AM (Cen,4, 80.00, Ar,1.0,556.28,0.70,LS 1); Sm (Mn, 2x2.00); Sb (16,15.00); Cm (9:16)
1: TOF MS ES+ 9.27e3



Minimum:				-1.5		
Maximum:		200.0	6.0	1000.0		
Mass	Calc. Mass	mDa	PPM	DBE	Score	Formula
1109.1757	1109.1701	5.6	5.0	37.5	1	C54 H38 N8 O4 F6 P Ru

Figure S18 – HR-MS (ESI-QTOF): Single Mass Analysis of **(6)** indicating $[M - PF_6^-]^+$.

[Ru(dppz)(bpy-PEG)(bpyArCOOEt)]$(PF_6)_2$ (7)

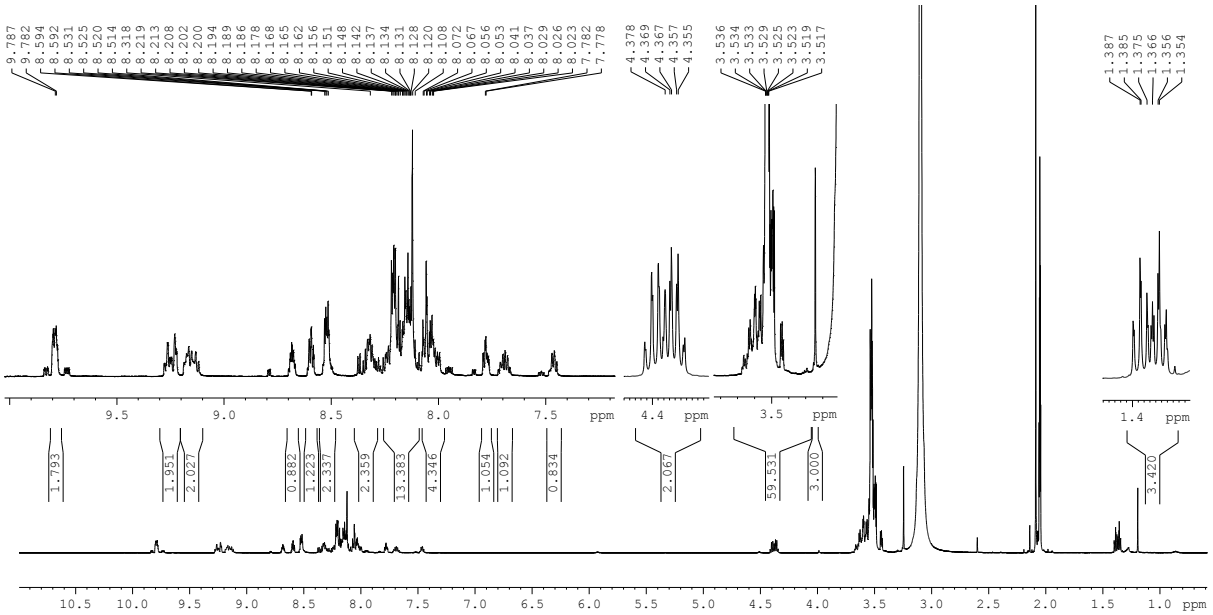
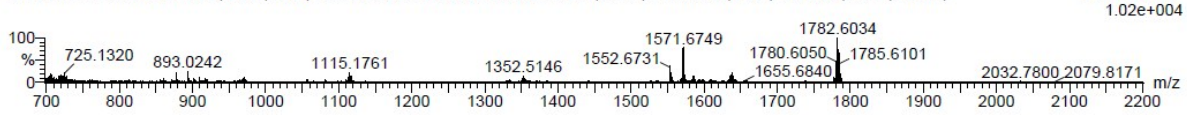


Figure S19 – ^{1}H NMR Spectrum (600 MHz) of **(7)** in acetone-d_6. The peaks at 2.09 and 3.10 are assigned to residual acetone and water respectively.

Single Mass Analysis
Tolerance = 100.0 PPM / DBE: min = -1.5, max = 500.0
Element prediction: Off
Number of isotope peaks used for i-FIT = 3

Monoisotopic Mass, Odd and Even Electron Ions
5250 formula(e) evaluated with 1 results within limits (all results (up to 1000) for each mass)
Elements Used:
C: 0-85 H: 0-101 N: 0-9 O: 0-18 F: 0-6 P: 0-1 Ru: 0-1

Christopher Burke (TK), CBTK-032
Q-TOF20160108MF001 103 (2.067) AM (Cen,4, 80.00, Ar,10000.0,1570.68,0.70); Sm (SG, 1x5.00); Sb (15,10.00); Cm (46:125) TOF MS LD+ 1.02e+004



Minimum:				-1.5			
Maximum:		5.0	100.0	500.0			
Mass	Calc. Mass	mDa	PPM	DBE	i-FIT	i-FIT (Norm)	Formula
1782.6034	1782.5950	8.4	4.7	37.5	105.1	0.0	C85 H101 N9 O18 F6 P Ru

Figure S20 – HR-MS (MALDI-QTOF): Single Mass Analysis of **(7)** indicating $[M - PF_6^-]^+$.

[Ru(dppz)(bpyArCONH-PEG$_{15}$-OMe)(bpyArCOOH)](PF$_6$)$_2$ (8)

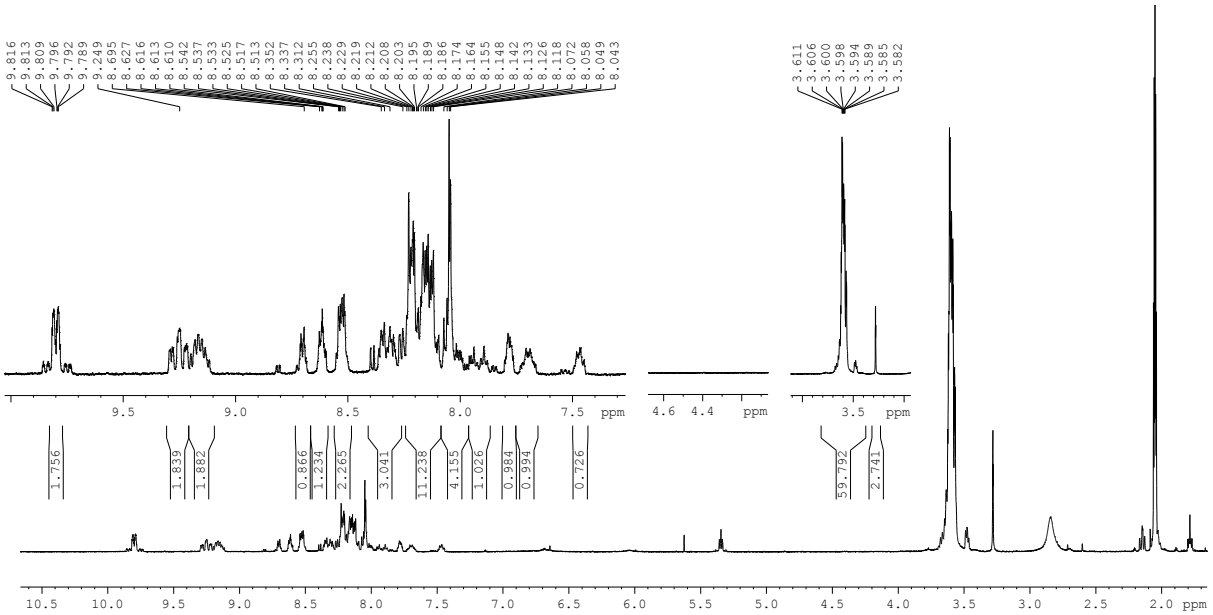
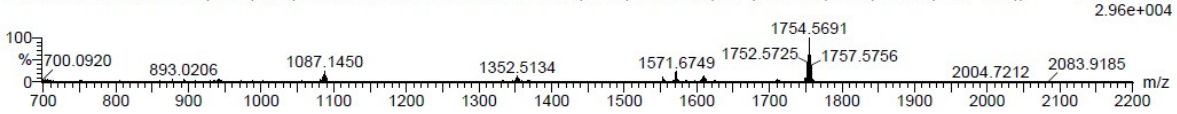


Figure S21 – ^{1}H NMR Spectrum (600 MHz) of **(8)** in acetone-d$_6$. The peaks at 5.63, 2.84 and 1.79 are assigned to dichloromethane, water and THF residual solvents from reaction work-up.

Single Mass Analysis
Tolerance = 100.0 PPM / DBE: min = -1.5, max = 500.0
Element prediction: Off
Number of isotope peaks used for i-FIT = 3

Monoisotopic Mass, Odd and Even Electron Ions
5250 formula(e) evaluated with 1 results within limits (all results (up to 1000) for each mass)
Elements Used:
C: 0-83 H: 0-97 N: 0-9 O: 0-18 F: 0-6 P: 0-1 Ru: 0-1

Christopher Burke (TK), CBTK-033
Q-TOF20160108MF002 95 (1.925) AM (Cen,4, 80.00, Ar,10000.0,1570.68,0.70); Sm (SG, 1x5.00); Sb (15,10.00); Cm (27:122-(95:97+119)) TOF MS LD+
2.96e+004



Minimum:				-1.5			
Maximum:		5.0	100.0	500.0			
Mass	Calc. Mass	mDa	PPM	DBE	i-FIT	i-FIT (Norm)	Formula
1754.5691	1754.5637	5.4	3.1	37.5	122.0	0.0	C83 H97 N9 O18 F6 P Ru

Figure S22 – HR-MS (MALDI-QTOF): Single Mass Analysis of **(8)** indicating [M – PF$_6^-$]$^+$.

[Ru(dppz)(bpy-PEG)(bpy-NFkB)]$(PF_6)_6$ (9)

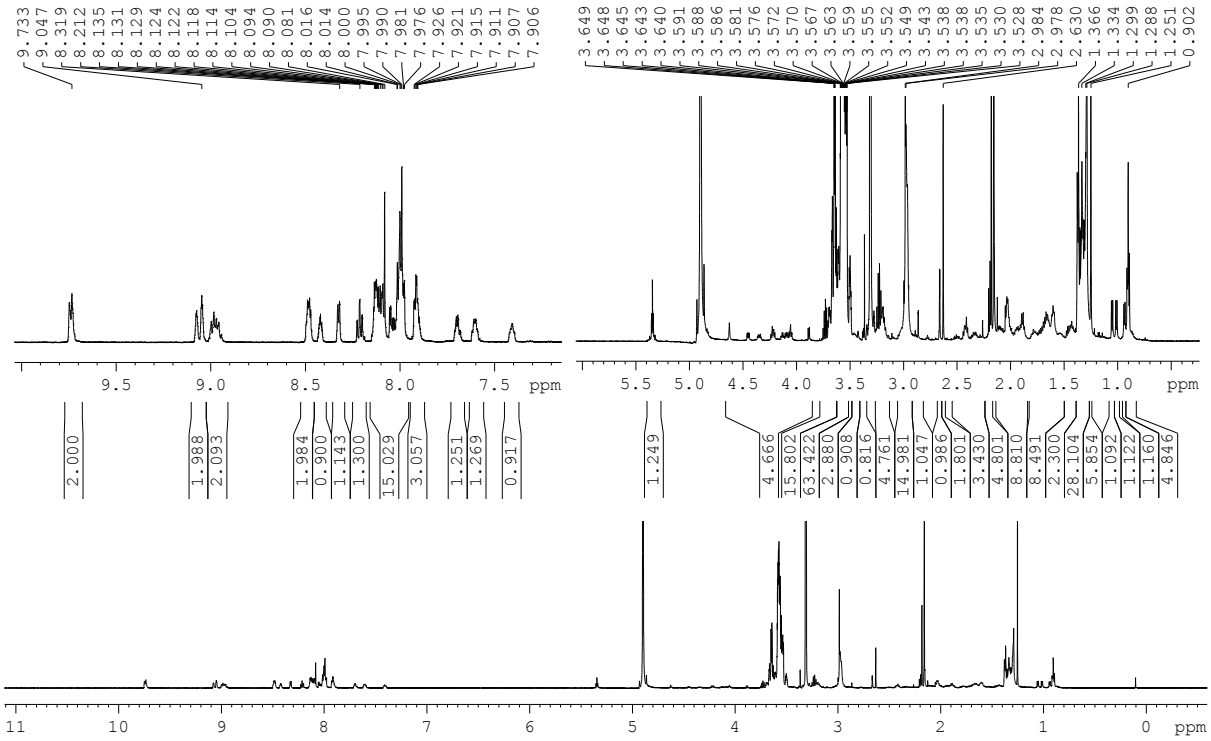


Figure S23 – ^{1}H NMR Spectrum (600 MHz) of **(9)** in methanol-d_4. The peaks at 4.79 and 2.15 ppm are due to residual water and acetone solvent from reaction work-up.

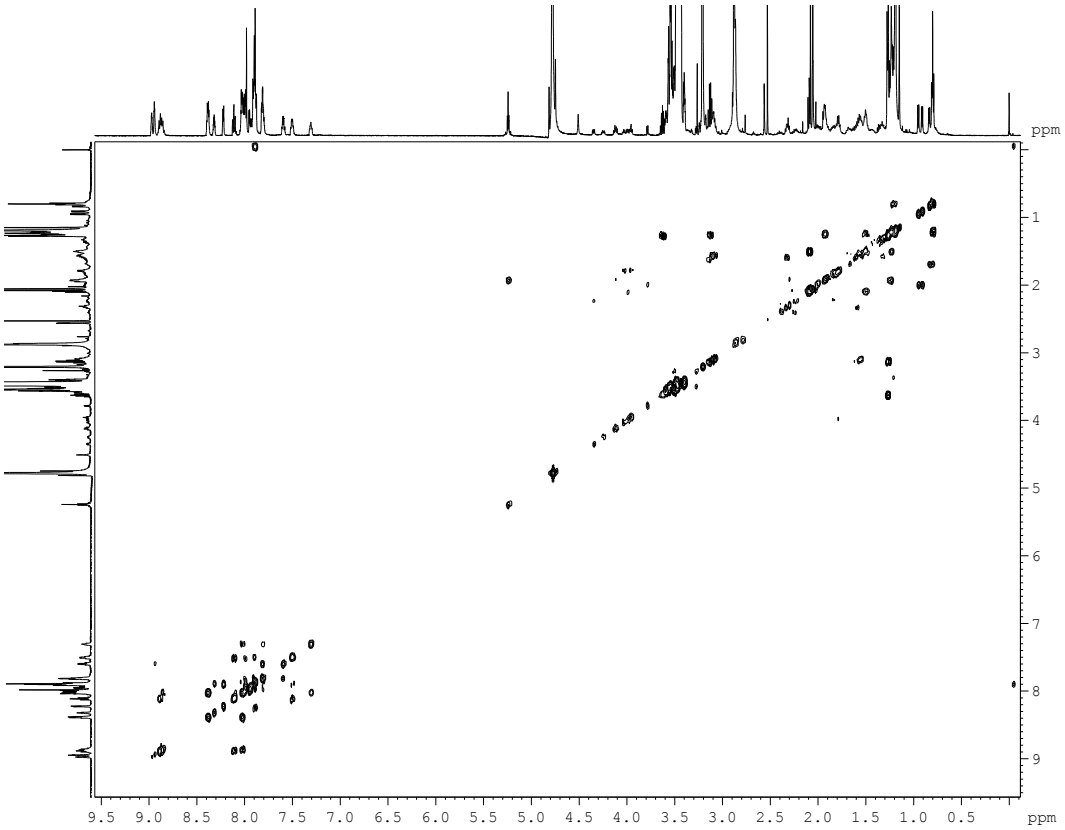


Figure S24 – COSY NMR Spectrum of **(9)** in methanol-d_4.

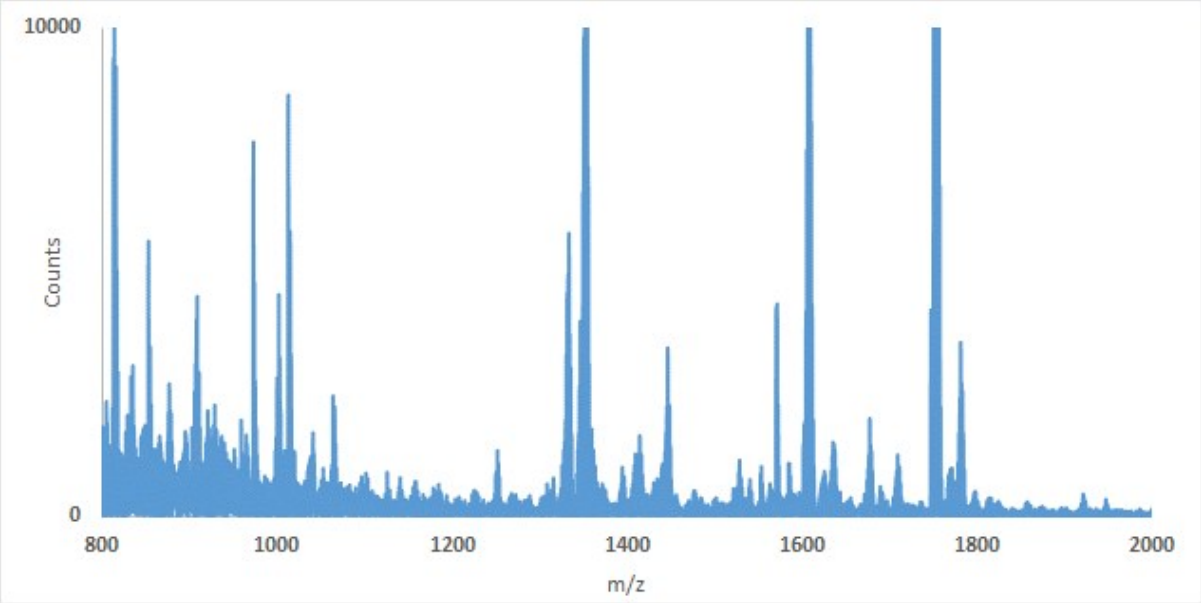


Figure S25 – HR-MS (MALDI-QTOF): Mass spectrum of **(9)**.

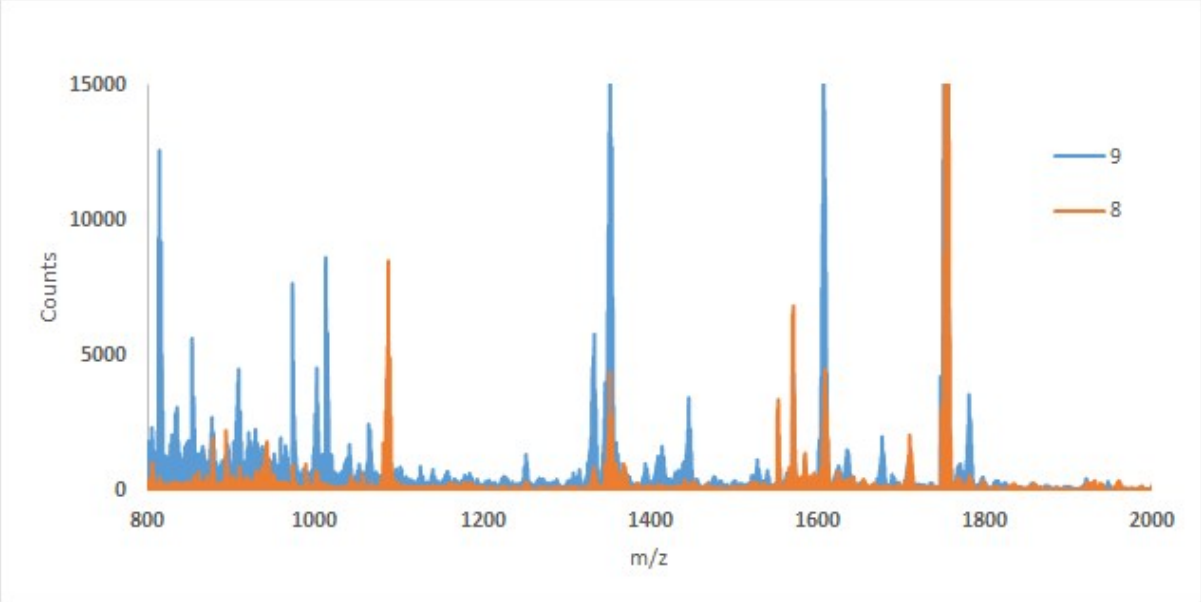
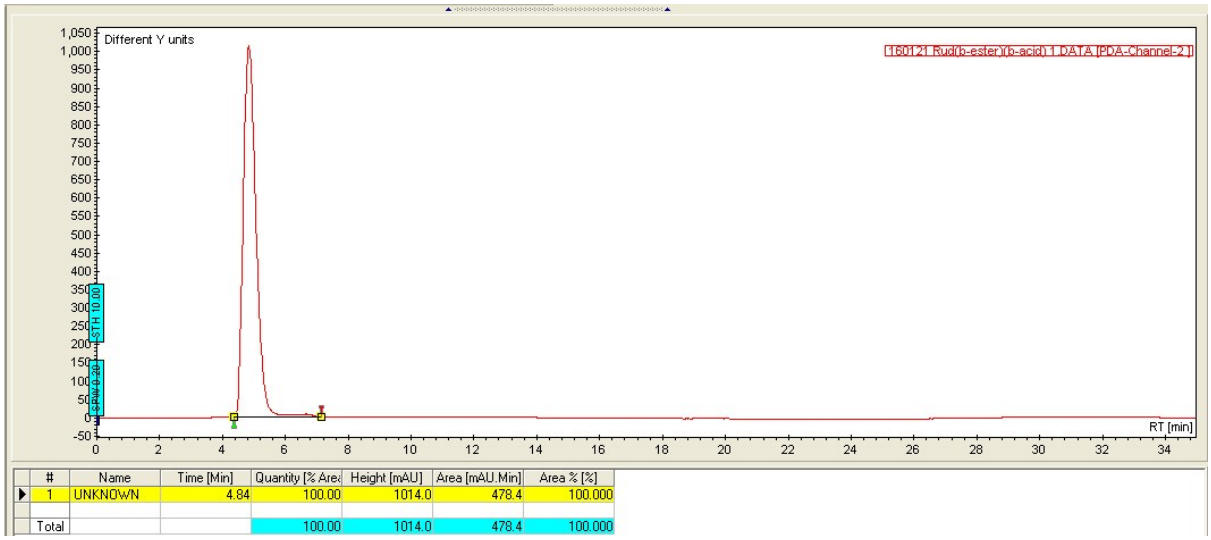


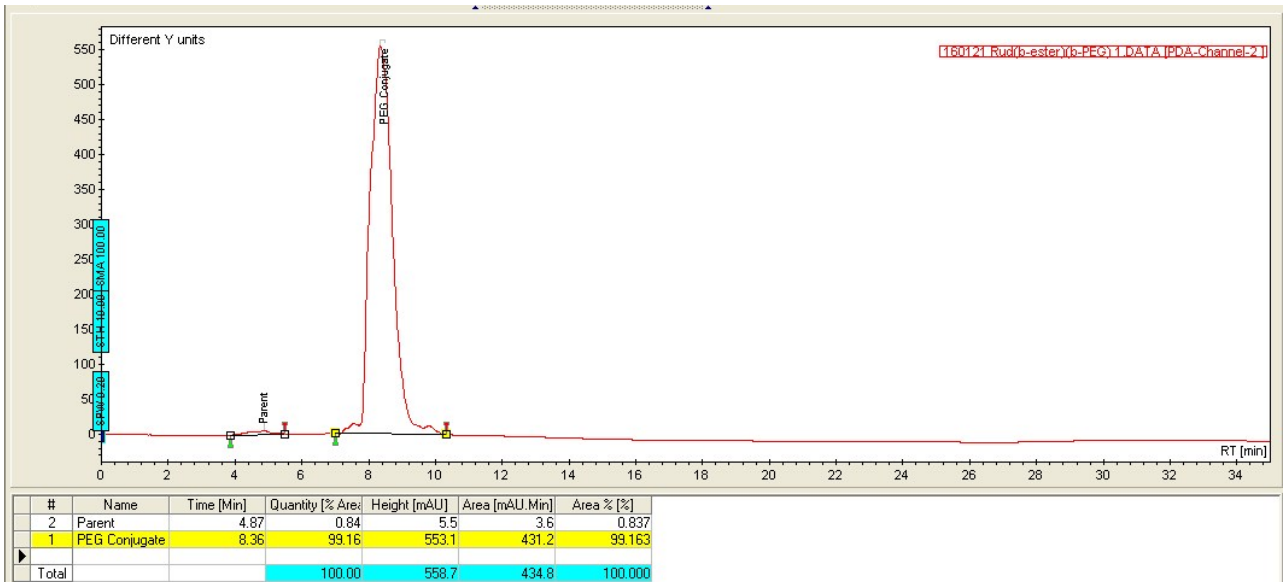
Figure S26 –Spectral overlay of the mass spectra of **(8)** and **(9)**.

HPLC Analysis of the Conjugates



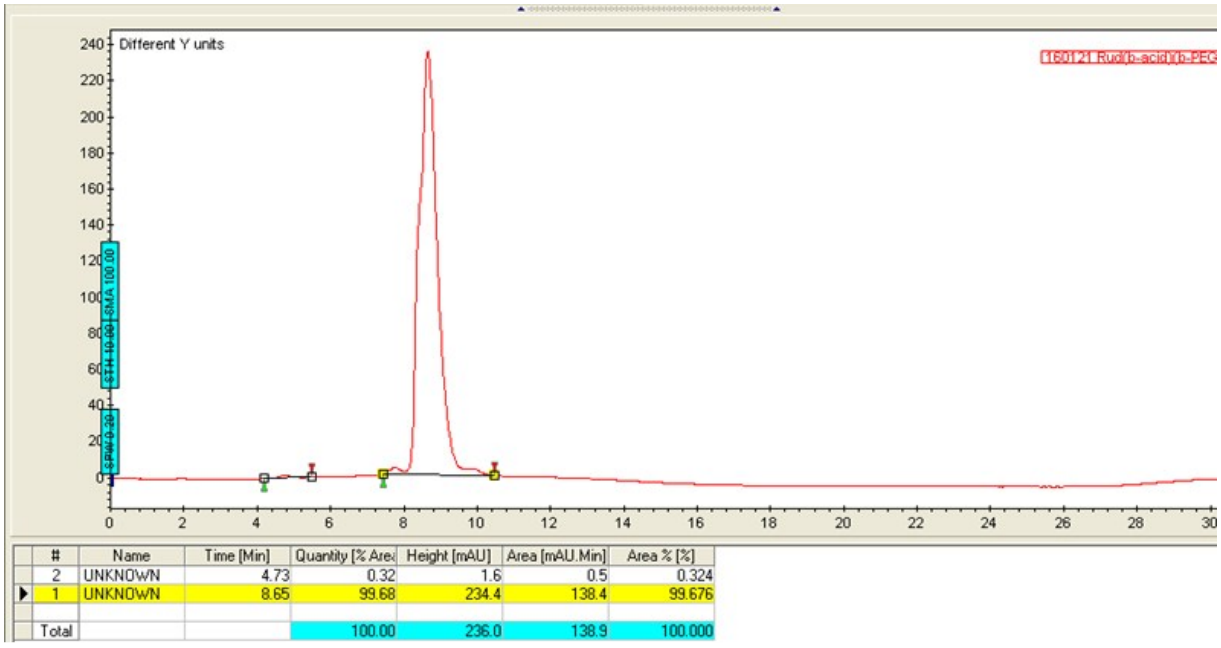
#	Name	Time [Min]	Quantity [% Area]	Height [mAU]	Area [mAU.Min]	Area % [%]
1	UNKNOWN	4.84	100.00	1014.0	478.4	100.000
Total			100.00	1014.0	478.4	100.000

Figure S27 – HPLC trace (RP-C18, MeCN/Water (0.1 % TFA) Gradient; PDAD) for **(6)**. Indicative Purity (vs Ru(II) precursors; 450 nm) = 100 %.



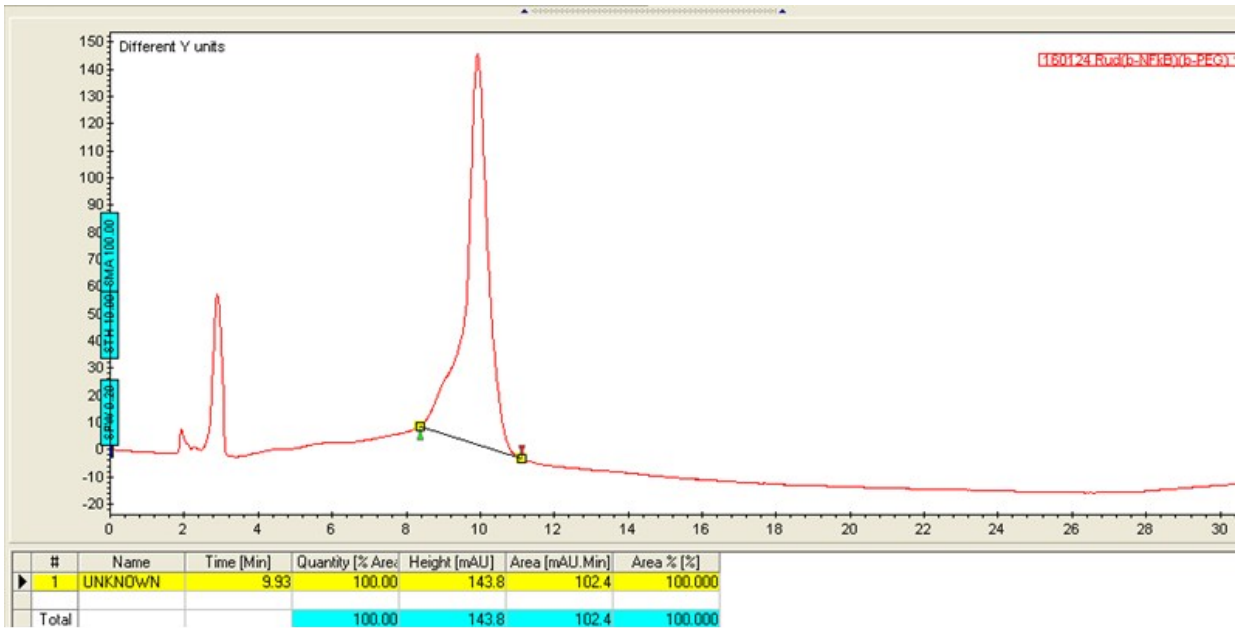
#	Name	Time [Min]	Quantity [% Area]	Height [mAU]	Area [mAU.Min]	Area % [%]
2	Parent	4.87	0.84	5.5	3.6	0.837
1	PEG Conjugate	8.36	99.16	553.1	431.2	99.163
Total			100.00	558.7	434.8	100.000

Figure S28 – HPLC trace (RP-C18, MeCN/Water (0.1 % TFA) Gradient; PDAD) for **(7)**. Indicative Purity (vs Ru(II) precursors; 450 nm) = 99.2 %.



#	Name	Time [Min]	Quantity [% Area]	Height [mAU]	Area [mAU.Min]	Area % [%]
2	UNKNOWN	4.73	0.32	1.6	0.5	0.324
1	UNKNOWN	8.65	99.68	234.4	138.4	99.676
Total			100.00	236.0	138.9	100.000

Figure S29 – HPLC trace (RP-C18, MeCN/Water (0.1 % TFA) Gradient; PDAD) for **(8)**. Indicative Purity (vs Ru(II) precursors; 450 nm) = 99.7 %.



#	Name	Time [Min]	Quantity [% Area]	Height [mAU]	Area [mAU.Min]	Area % [%]
1	UNKNOWN	9.93	100.00	143.8	102.4	100.000
Total			100.00	143.8	102.4	100.000

Figure S30 – HPLC trace (RP-C18, MeCN/Water (0.1 % TFA) Gradient; PDAD) for **(9)**. Early eluting peaks are due to sample solvent.

Additional Photophysical Data

General Information

For compounds **(4)** – **(6)**, solutions were prepared in MeCN using the PF_6^- salt of the complex whereas the Cl^- salt was preferred for the aqueous samples. Samples **(7)** and **(9)** were prepared from their PF_6^- salts and required *ca*. 0.5 % v/v DMSO as an initial solubilising agent. All scans and lifetime measurements were performed using 10 µM solutions. The extinction coefficients were calculated from standard curves (5 – 30 µM). Standard deviations are calculated from triplicate analyses. All lifetime curve fitting conformed to chi-squared tail-fit criteria of $0.9 < \chi^2 < 1.10$. Slit widths set to 5 nm for emission and excitation runs. Deaeration was performed by bubbling N_2 through the analytical sample for 15 mins minutes.

Charts

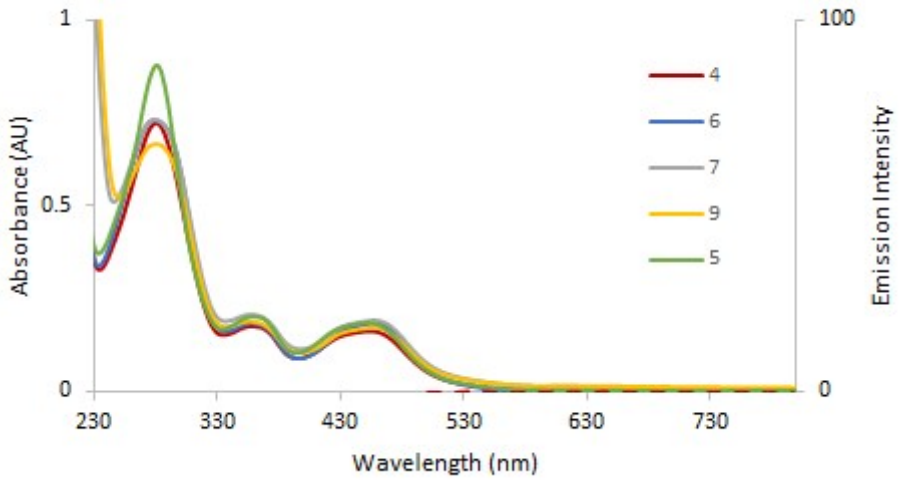


Figure S31 – Absorbance (full lines) and emission (dashed lines) spectra in water for the complexes **(4) – (9).**