Electronic Supplementary Material (ESI) for New Journal of Chemistry.

Supporting Information

First heterobimetallic Cu−dppf complexes designed for anticancer applications: synthesis, structural characterization and cytotoxicity

Catarina Bravo[a,b], M. Paula Robalo[c,d], Fernanda Marques[e], Alexandra R. Fernandes[f], Diogo A. Sequeira[f], M. Fátima M. Piedade[b,c], M. Helena Garcia[a,b], Maria J. Villa de Brito[a,b*], Tânia S. Morais[a,b*]

[a] Centro de Química Estrutural, Faculdade de Ciências, Universidade de Lisboa, Portugal.

[b] Centro de Química Estrutural, Instituto Superior Técnico, Universidade de Lisboa, Portugal.

[c] Departamento de Química e Bioquímica, Faculdade de Ciências, Universidade de Lisboa, Portugal.

[d] Centro de Ciências e Tecnologias Nucleares, Instituto Superior Técnico, Universidade de Lisboa, Portugal.

[2] UCBIO, Departamento de Ciências da Vida, Faculdade de Ciências e Tecnologia, Universidade Nova de Lisboa, Portugal.

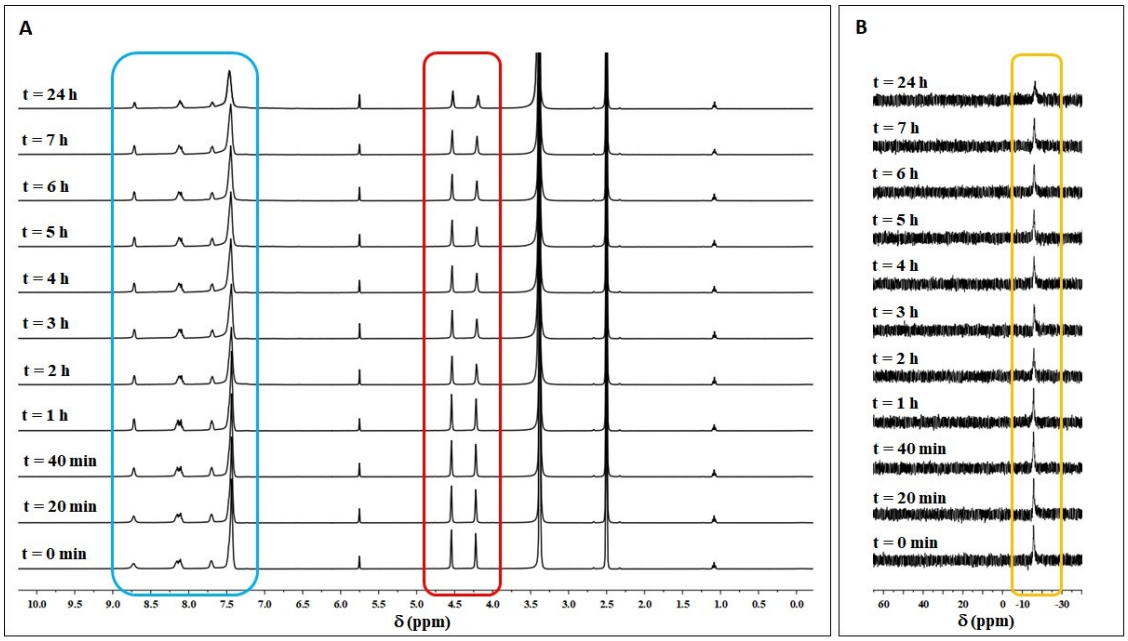


Figure S1 - Evaluation of complex **4** stability by ^{1}H- (**A**) and ^{31}P-NMR (**B**) over 24 h.

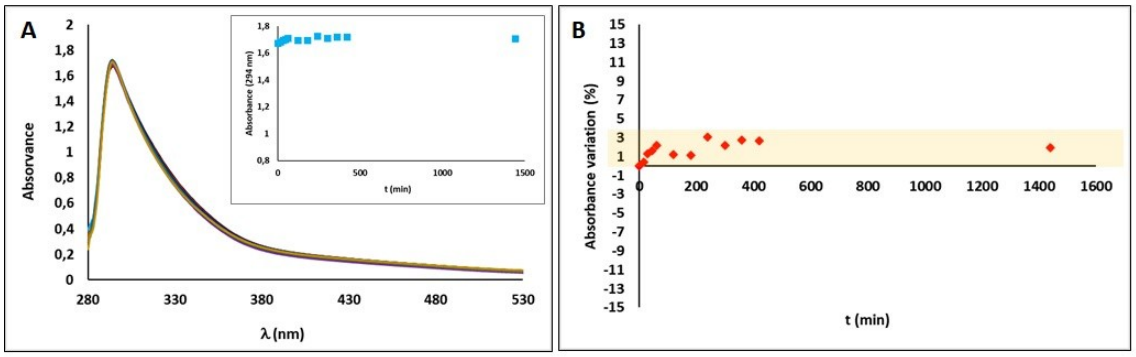


Figure S2 - Evaluation of the stability of complex **4** by UV-vis spectroscopy over 24 hours in 5%DMSO/ DMEM+GlutaMAX-I™ solution. (**A**) UV–visible spectra recorded over time, from 0 min up to 1440 min=24 h, for a 43 µM solution; inset: changes in absorbance observed at 294 nm (■); (**B**) absorbance variation percentage along time (0 min to 1440 min) (♦).

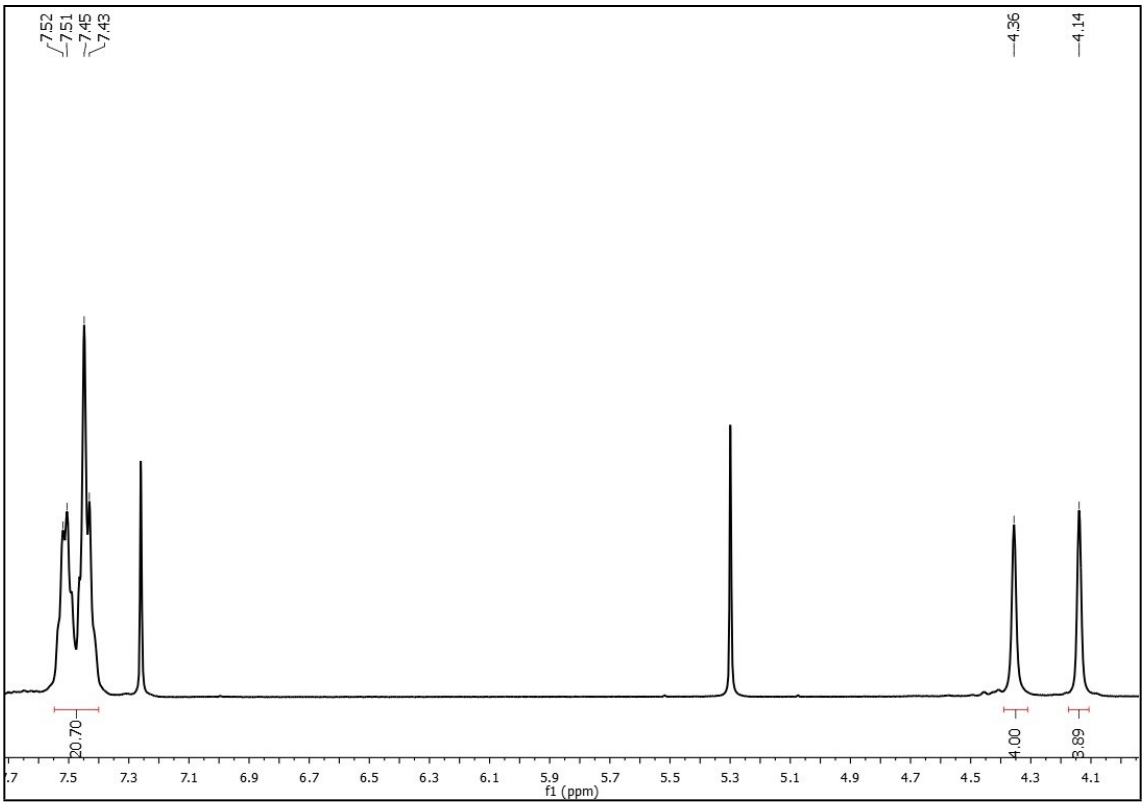


Figure S3 - ^{1}H-NMR spectrum of $[Cu(dppf)(NCMe)_2][BF_4]$ in CDCl3.

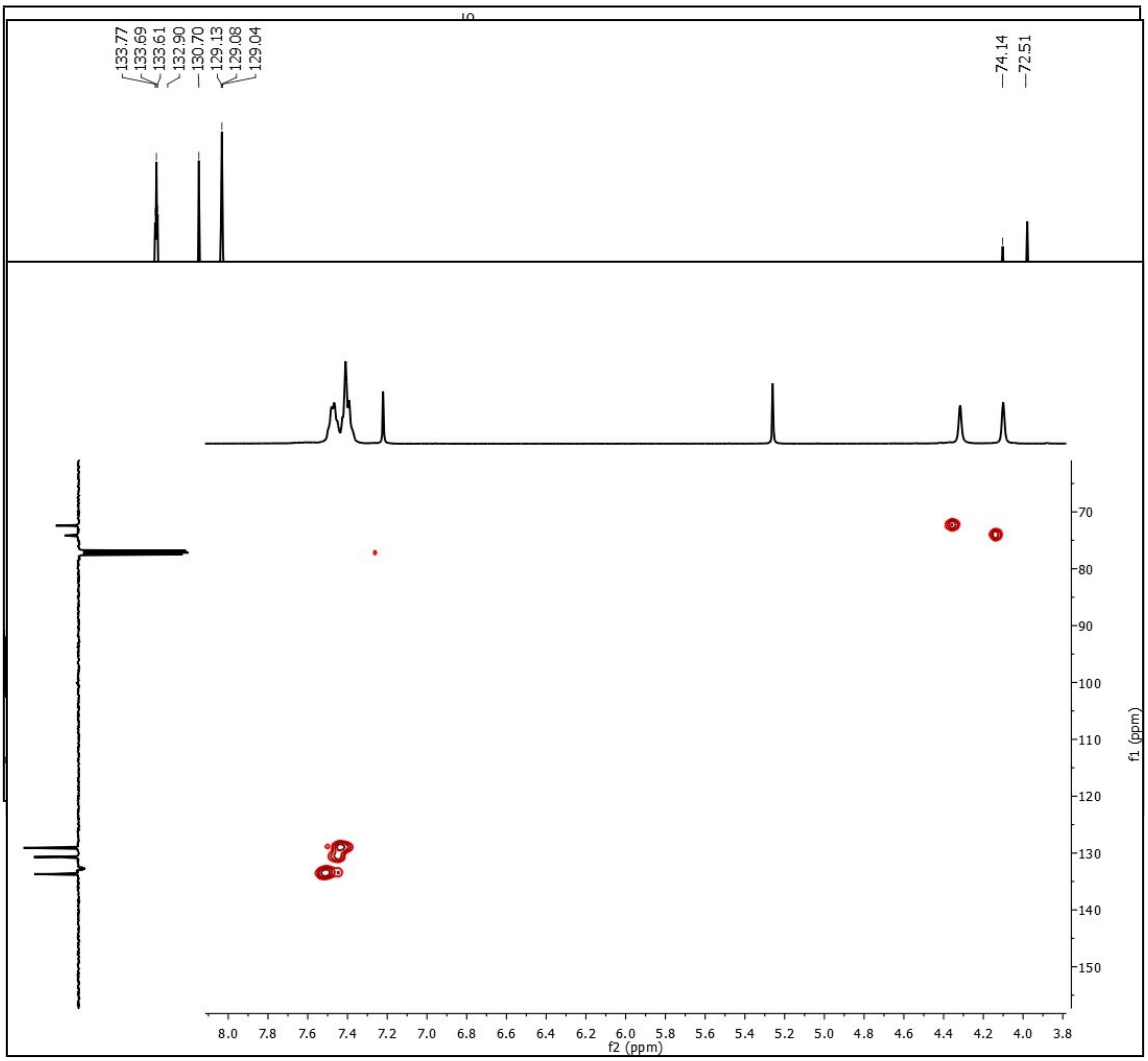


Figure S6 - HSQC-NMR spectrum of $[Cu(dppf)(NCMe)_2][BF_4]$ in CDCl3.

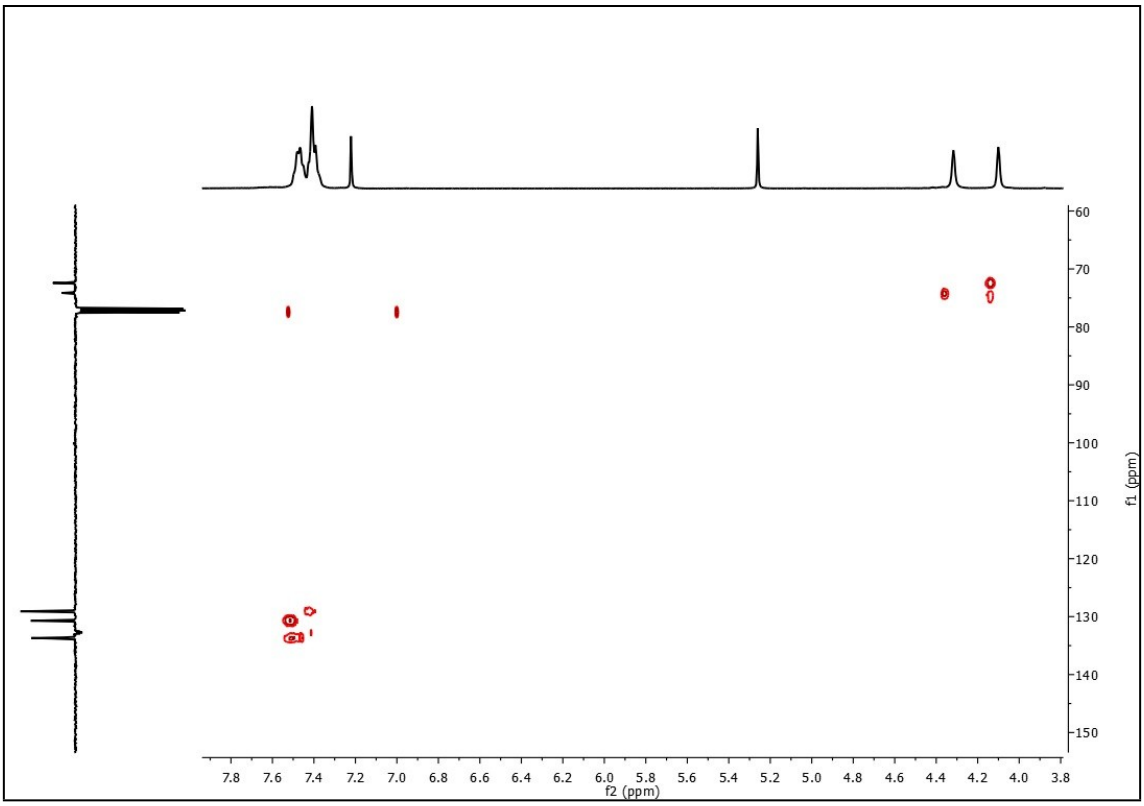


Figure S7 - HMBC-NMR spectrum of $[Cu(dppf)(NCMe)_2][BF_4]$ in CDCl3.

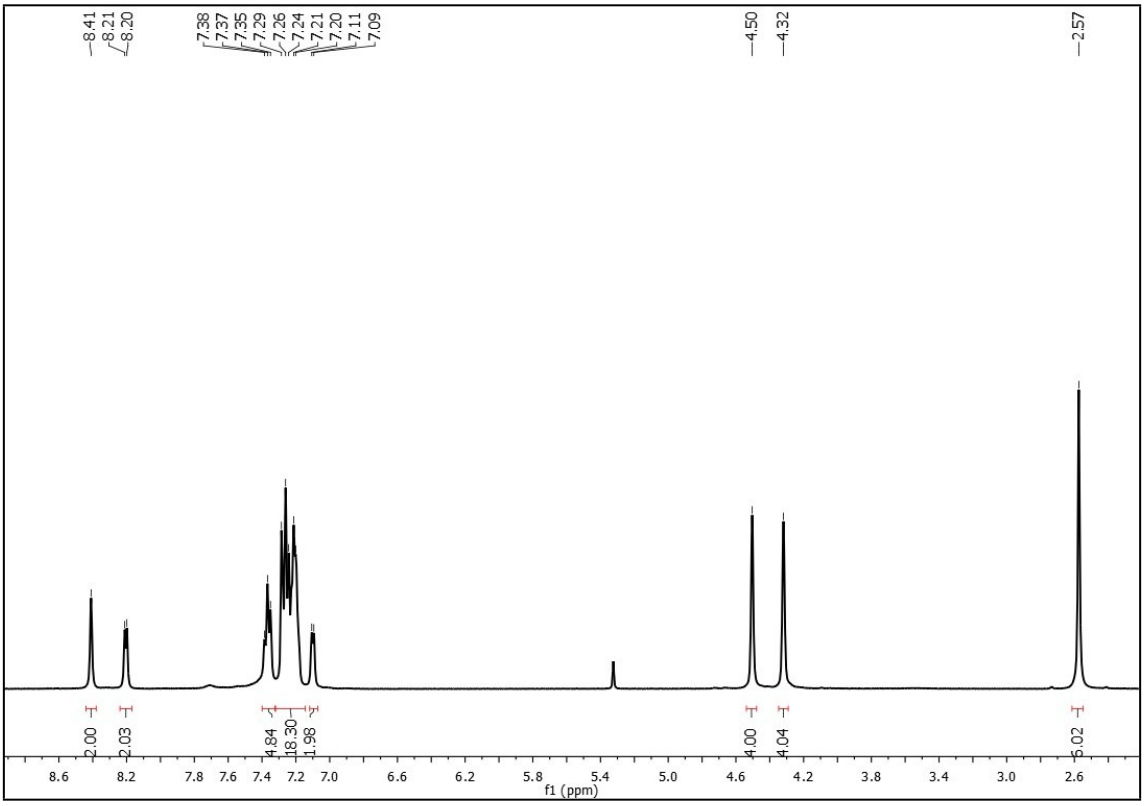


Figure S8 - ^{1}H-NMR spectrum of $[Cu(dppf)(Me_2bpy)][BF_4]$(**1**) in CDCl3.

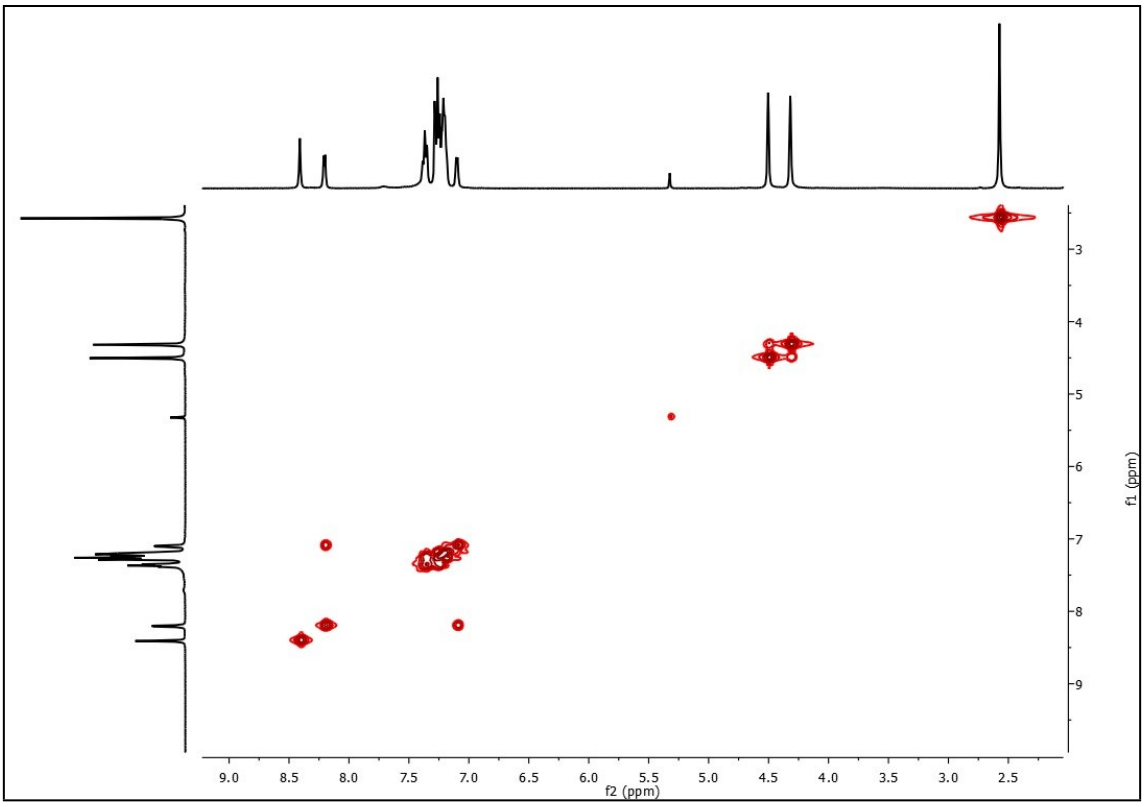


Figure S9 – cosy-NMR spectrum of [Cu(dppf)(Me_2bpy)][BF_4](**1**) in CDCl3.

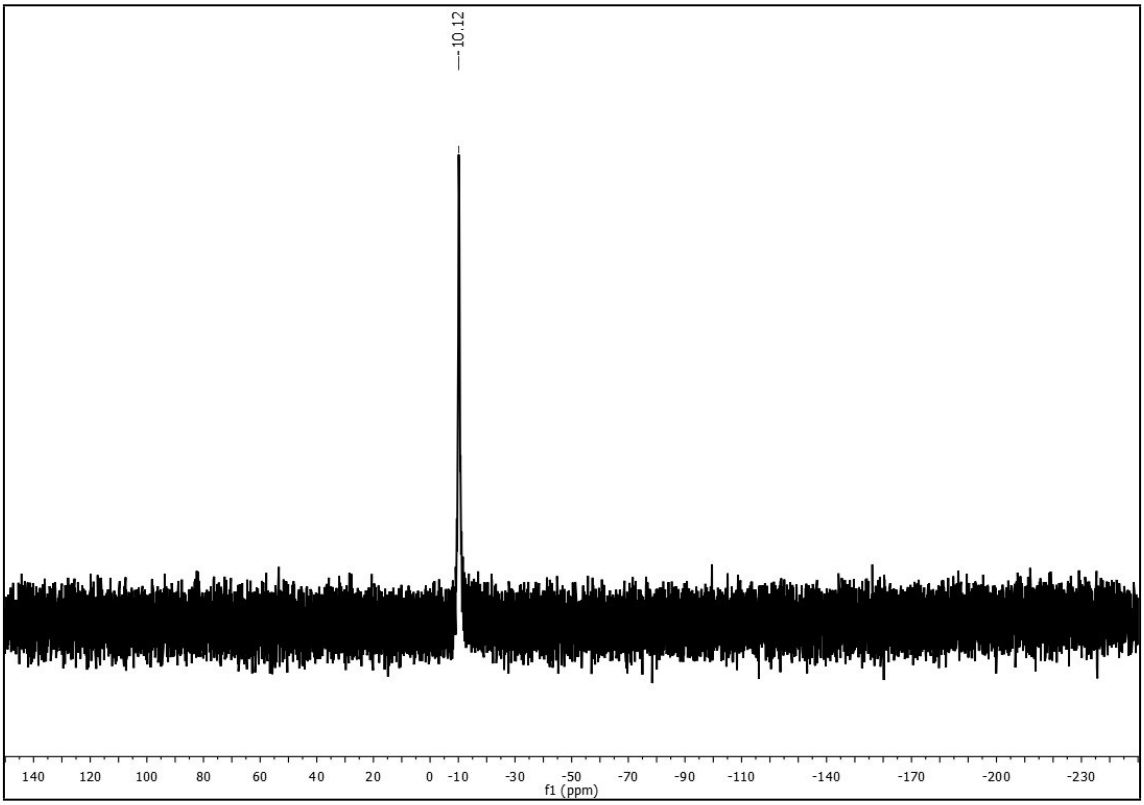


Figure S10 – ^{31}P-NMR spectrum of [Cu(dppf)(Me_2bpy)][BF_4](**1**) in CDCl3.

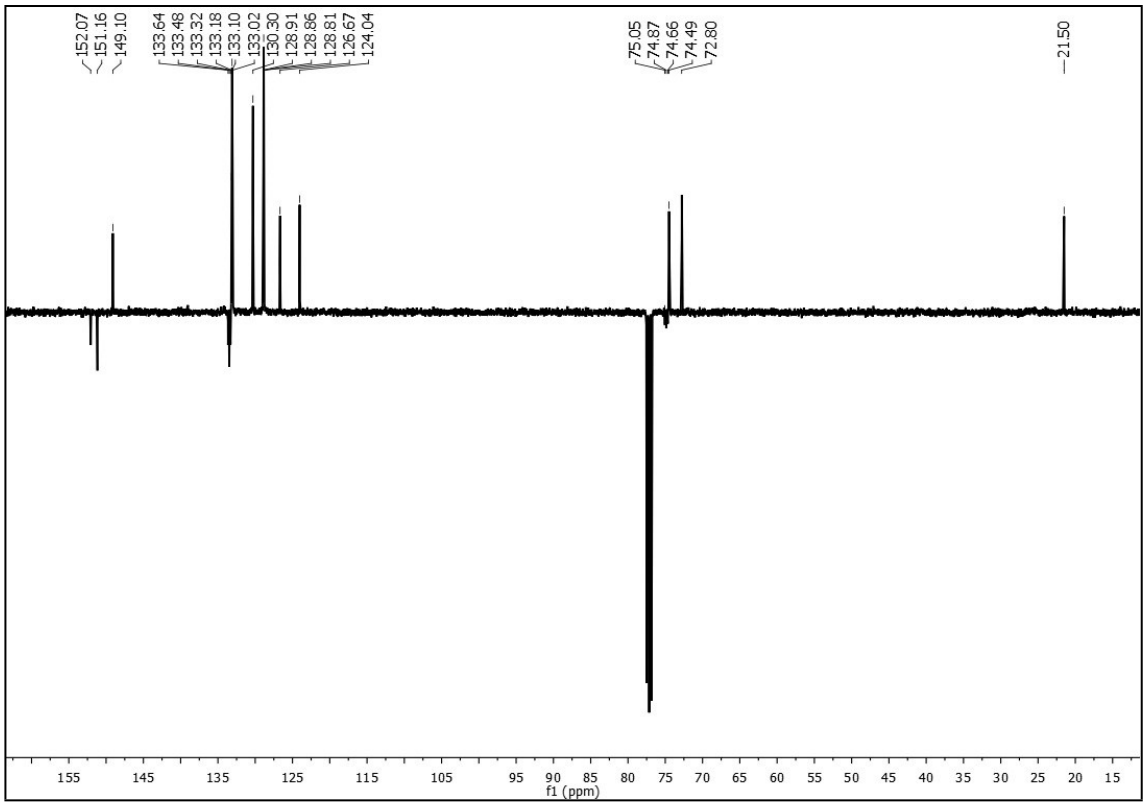


Figure S11 – ^{13}C-NMR spectrum of [Cu(dppf)(Me$_2$bpy)][BF$_4$](**1**) in CDCl3.

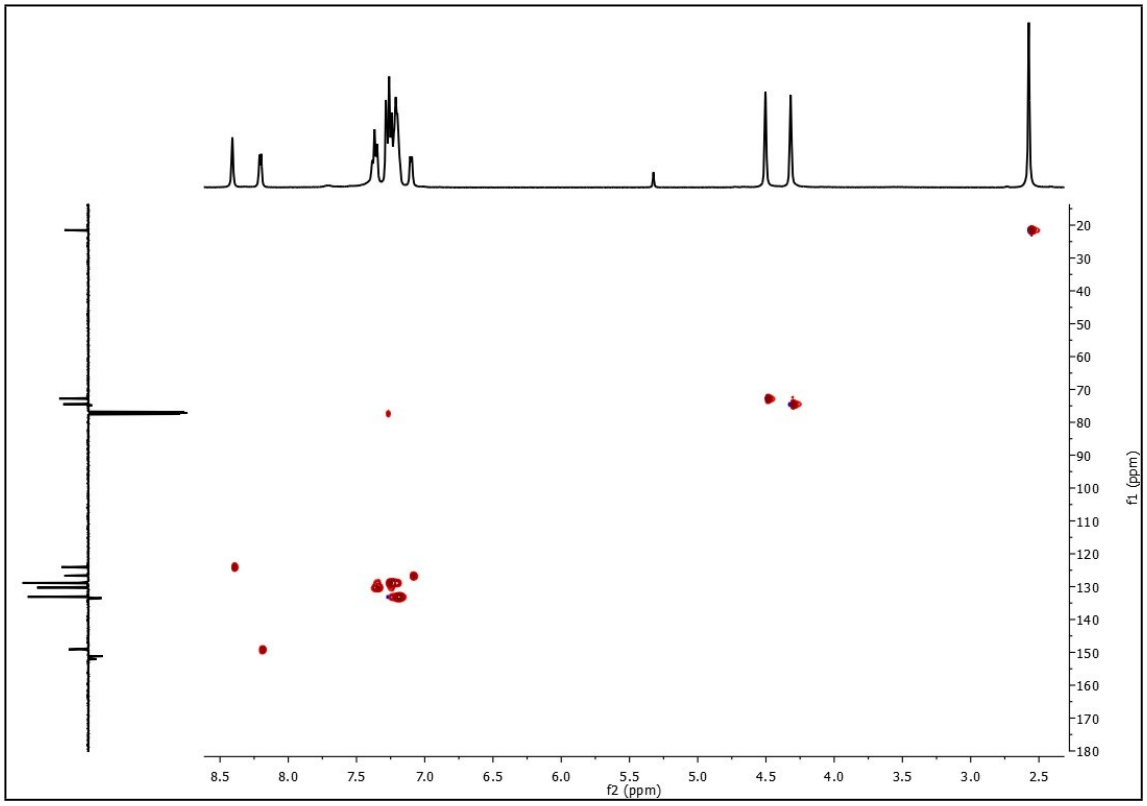


Figure S12 - HSQC-NMR spectrum of [Cu(dppf)(Me$_2$bpy)][BF$_4$](**1**) in CDCl3.

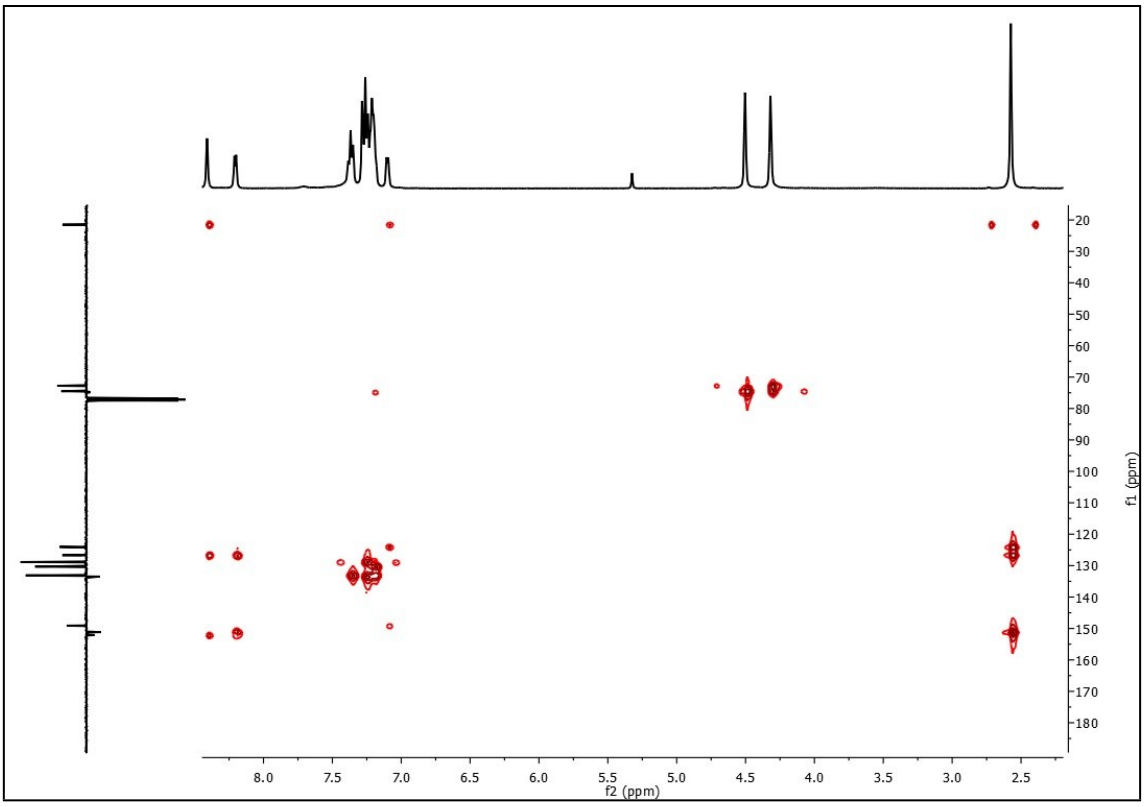


Figure S13 - HMBC-NMR spectrum of [Cu(dppf)(Me_2bpy)][BF_4](**1**) in CDCl3.

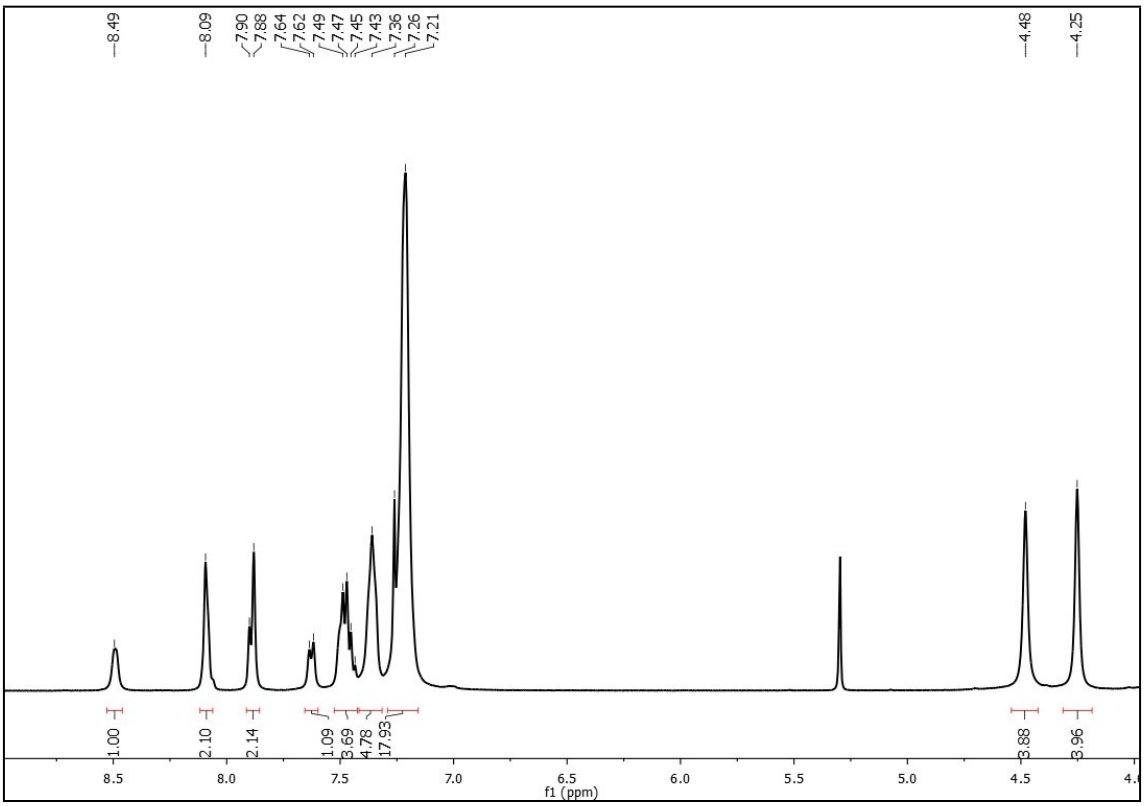


Figure S14 - ^{1}H-NMR spectrum of [Cu(dppf)(pbt)][BF_4](**2**) in CDCl3.

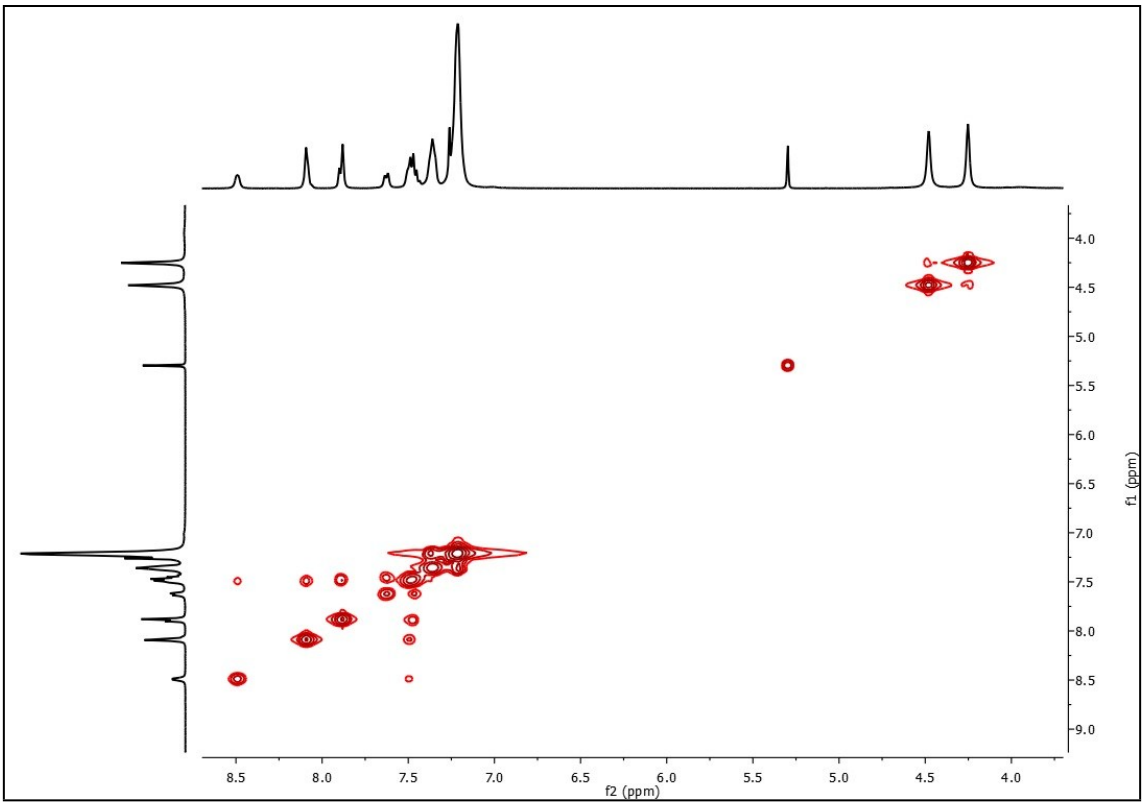


Figure S15 – cosy-NMR spectrum of [Cu(dppf)(pbt)][BF_4](**2**) in CDCl3.

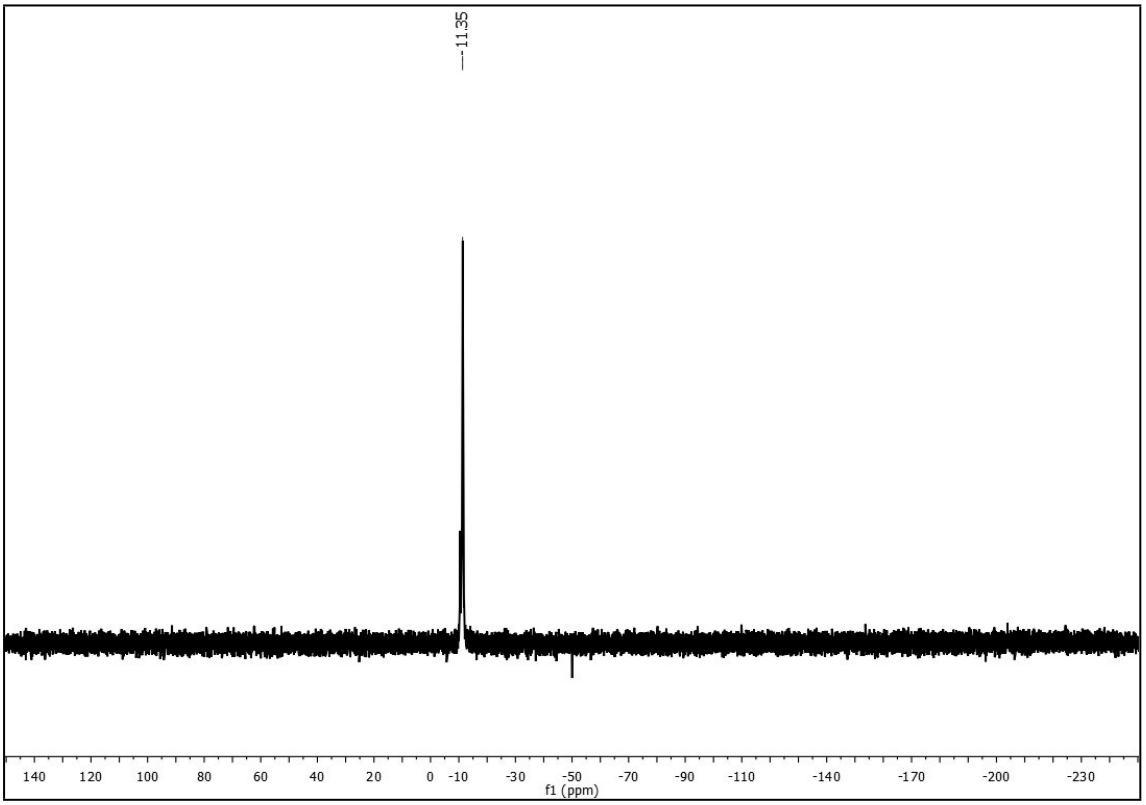


Figure S16 – ^{31}P-NMR spectrum of [Cu(dppf)(pbt)][BF_4](**2**) in CDCl3.

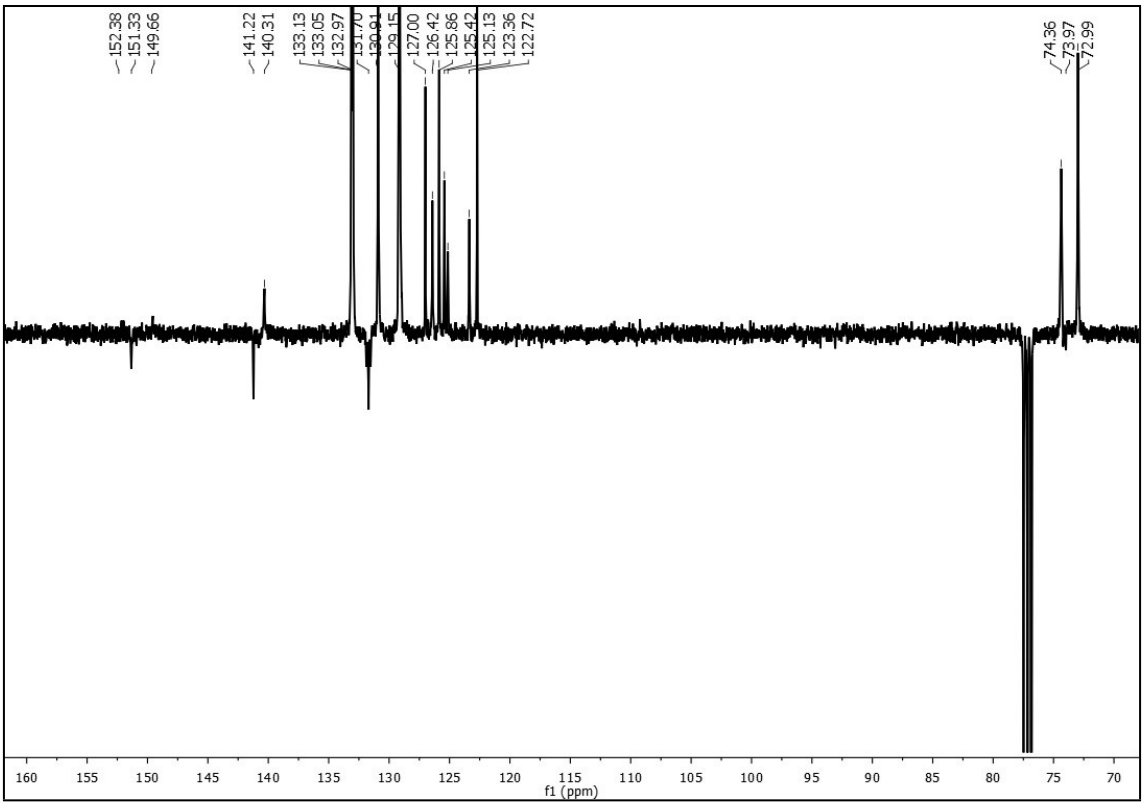


Figure S17 – ^{13}C-NMR spectrum of [Cu(dppf)(pbt)][BF_4](**2**) in CDCl3.

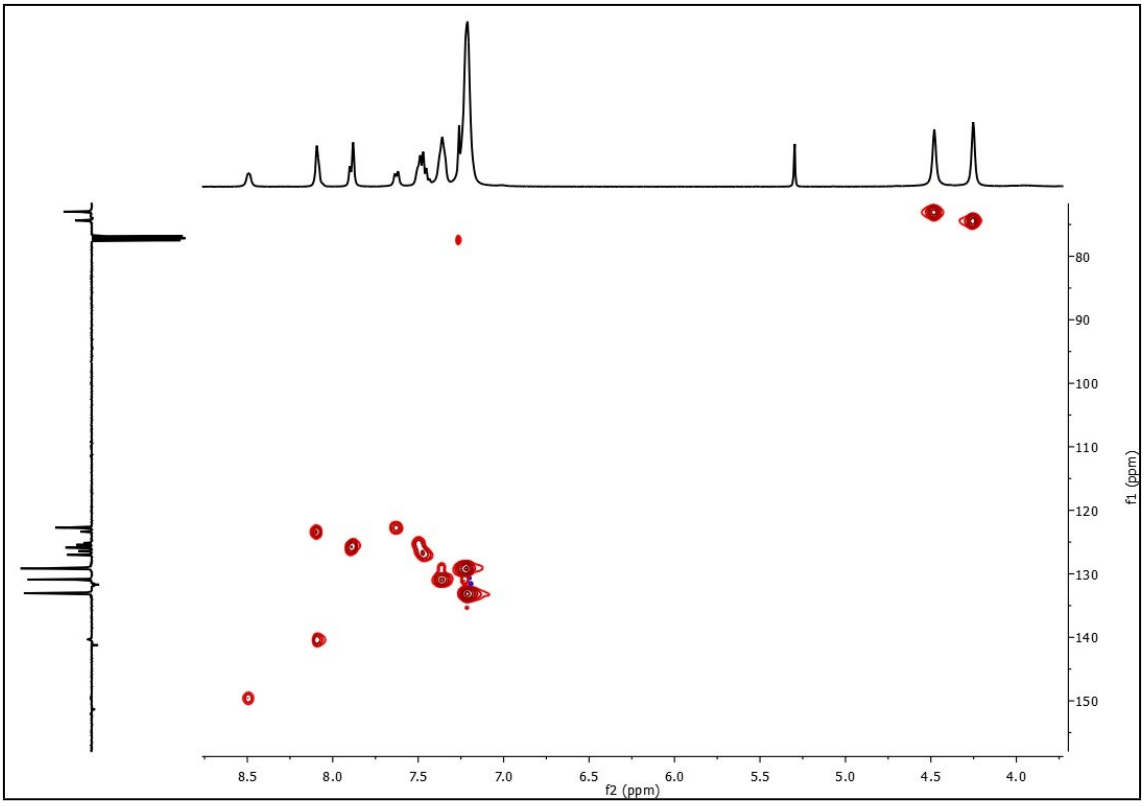


Figure S18 - HSQC-NMR spectrum of [Cu(dppf)(pbt)][BF_4](**2**) in CDCl3.

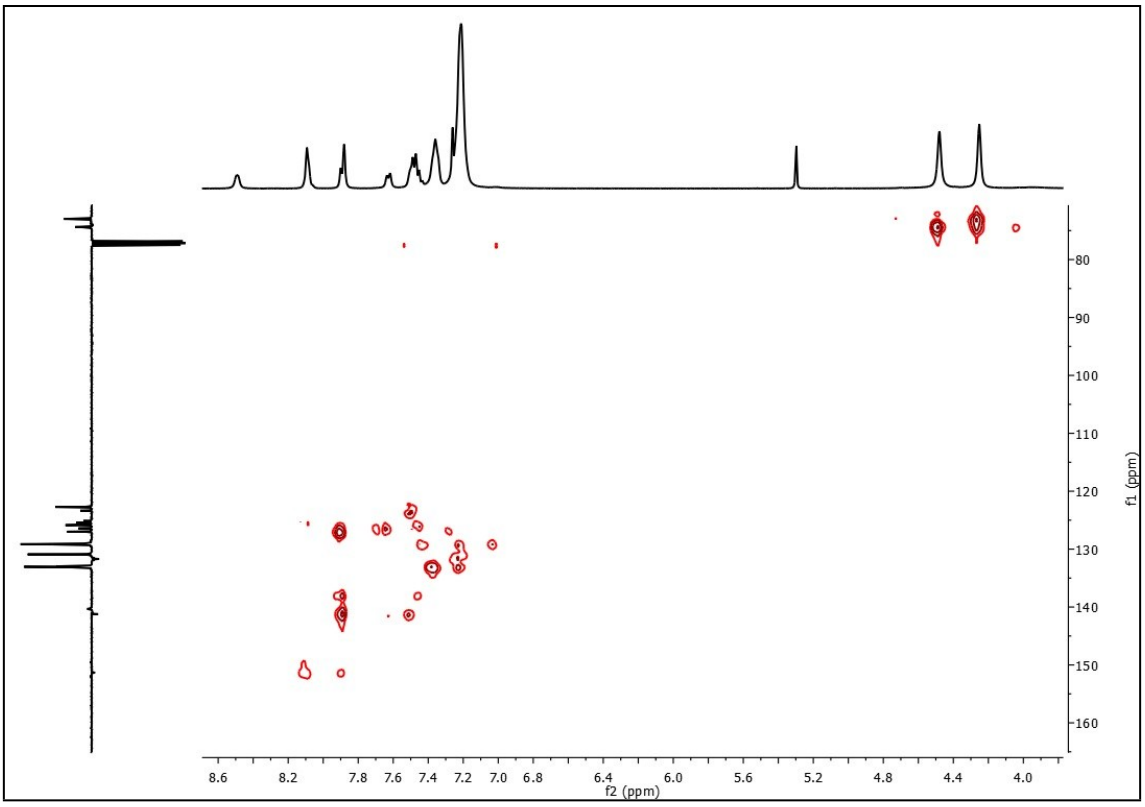


Figure S19 - HMBC-NMR spectrum of [Cu(dppf)(pbt)][BF_4](**2**) in CDCl3.

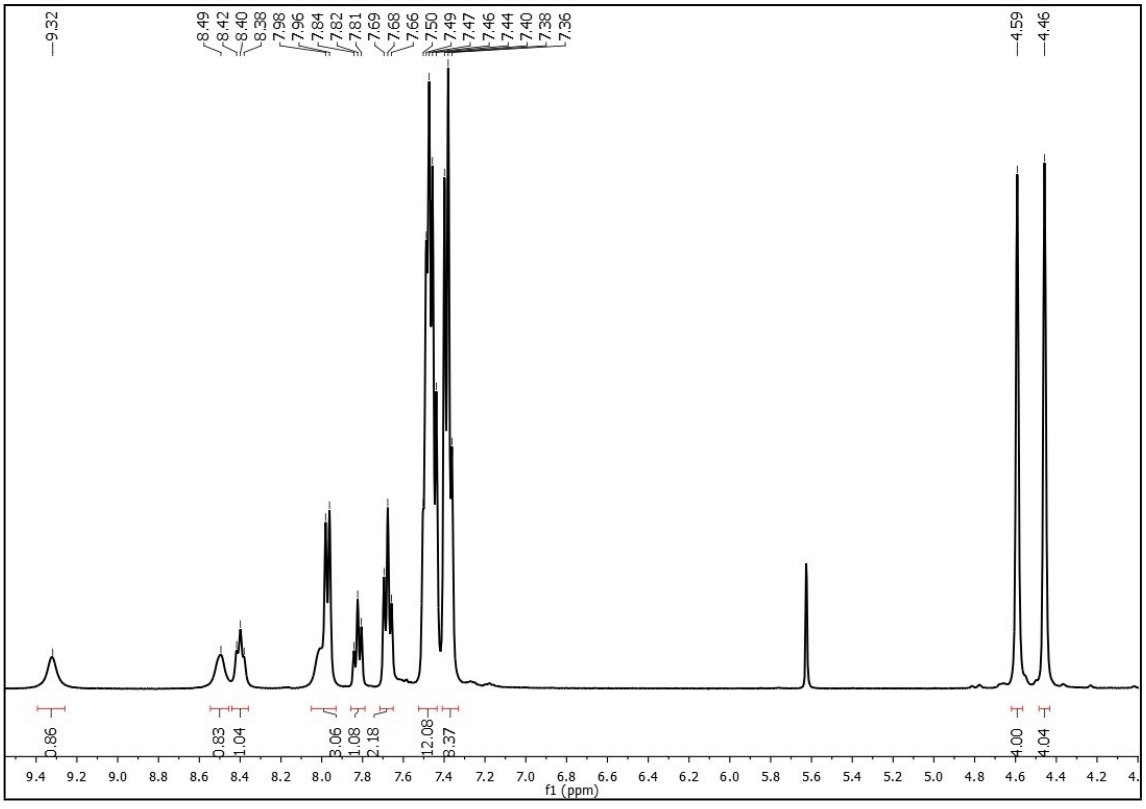


Figure S20 - ^{1}H-NMR spectrum of [Cu(dppf)(bopy)][BF_4](**3**) in $(CD_3)_2CO$.

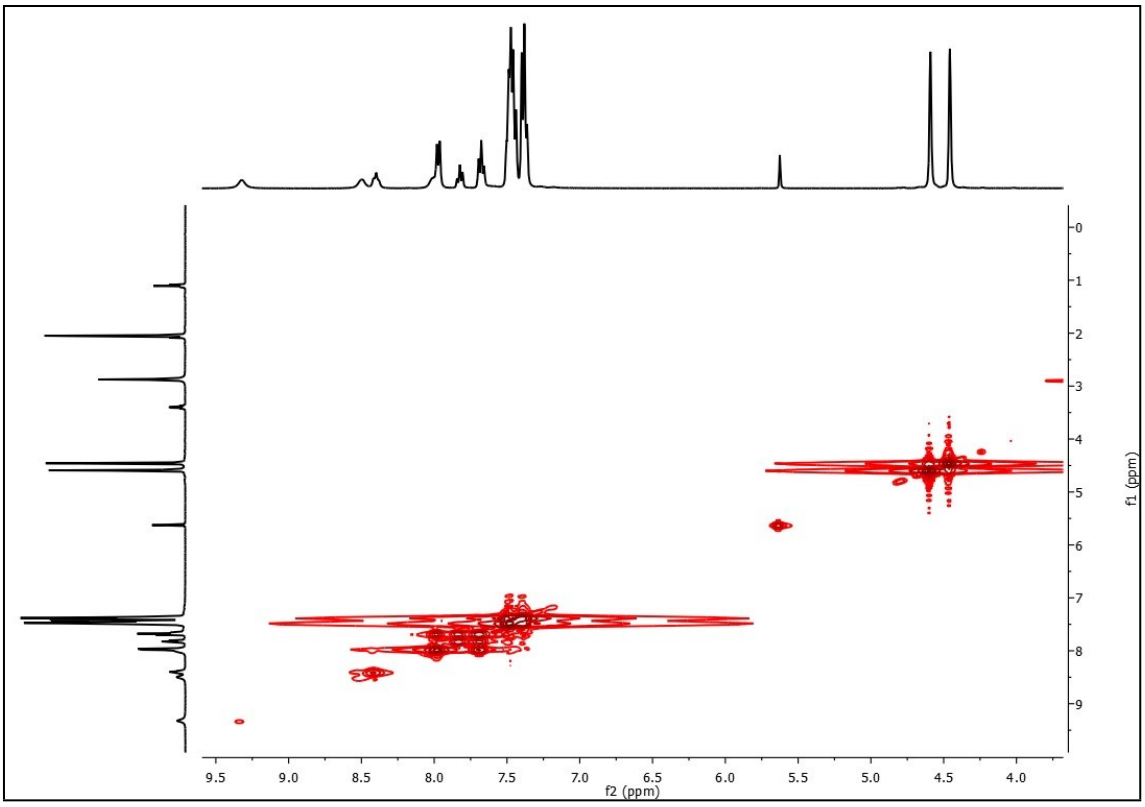


Figure S21 – cosy-NMR spectrum of [Cu(dppf)(bopy)][BF_4](**3**) in $(CD_3)_2CO$.

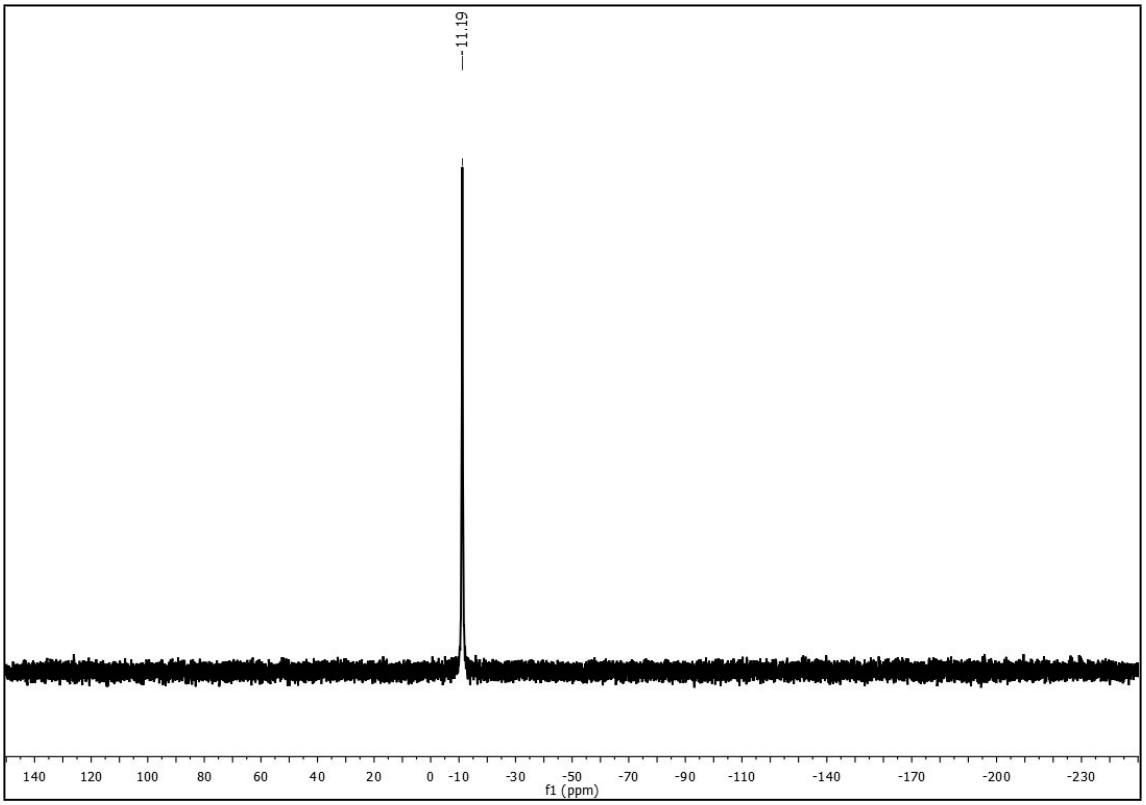


Figure S22 – ^{31}P-NMR spectrum of [Cu(dppf)(bopy)][BF_4](**3**) in $(CD_3)_2CO$.

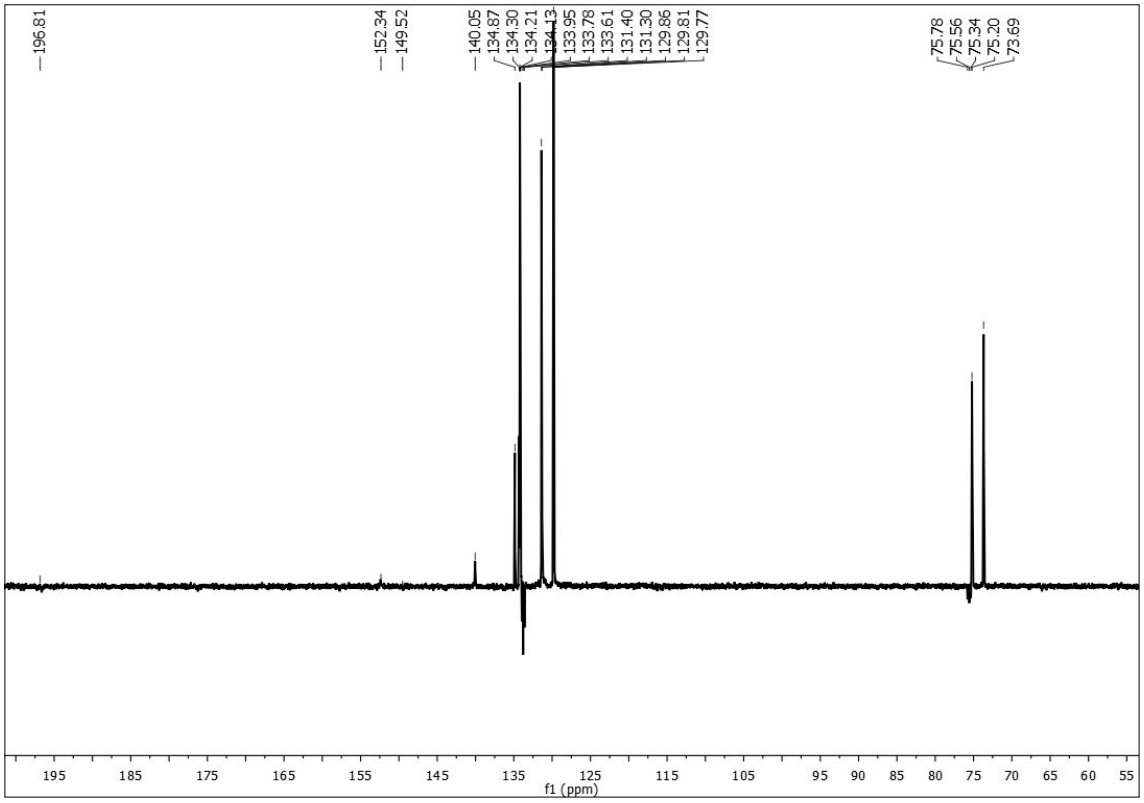


Figure S23 – ^{13}C-NMR spectrum of [Cu(dppf)(bopy)][BF_4](**3**) in $(CD_3)_2CO$.

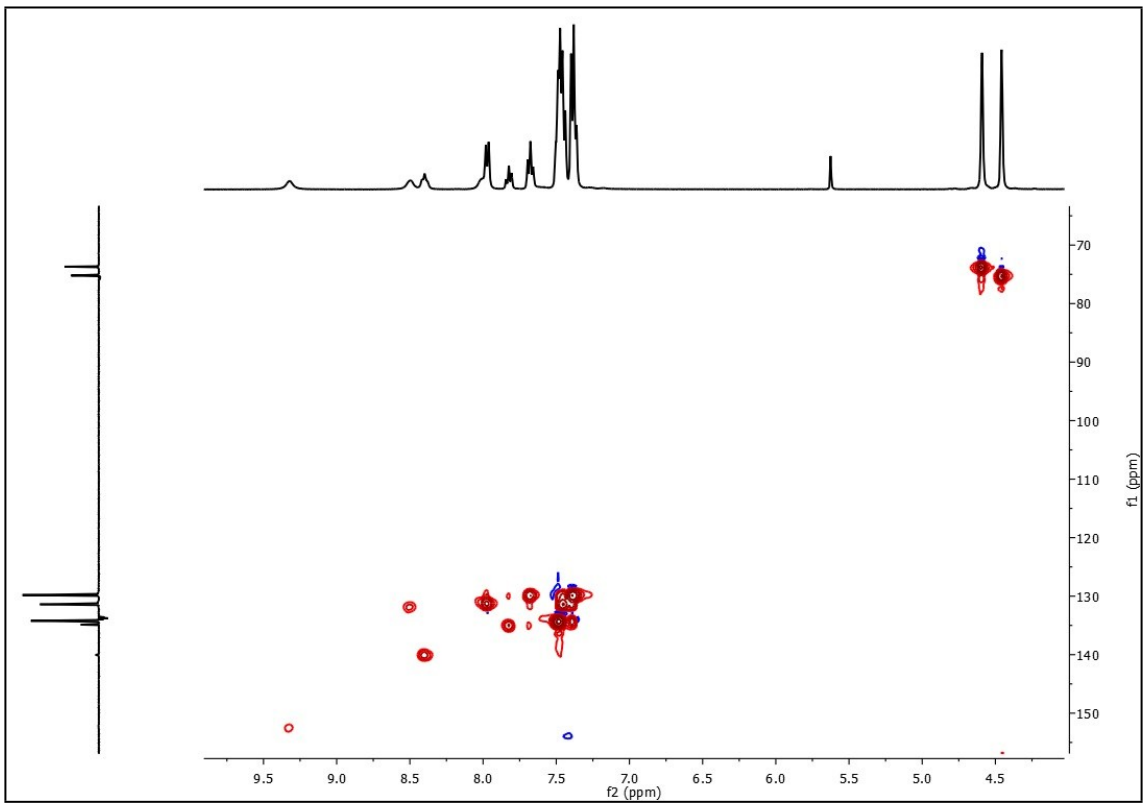


Figure S24 - HSQC-NMR spectrum of [Cu(dppf)(bopy)][BF_4](**3**) in $(CD_3)_2CO$.

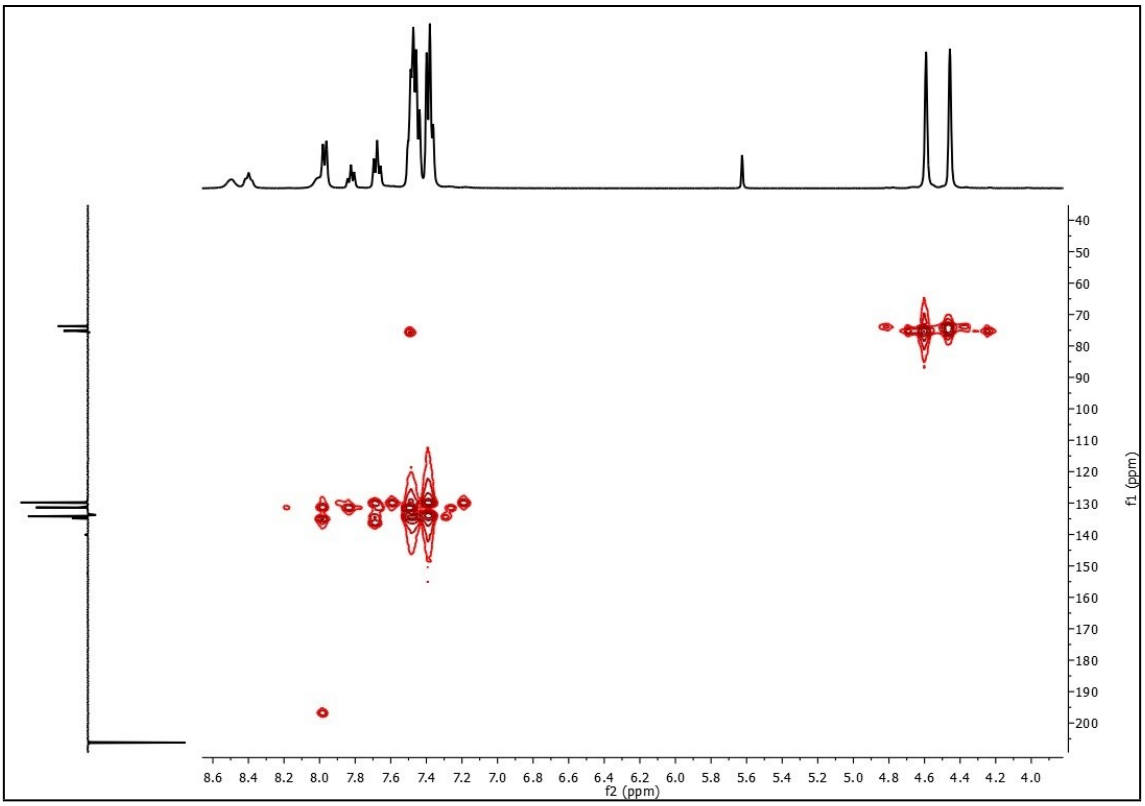


Figure S25 - HMBC-NMR spectrum of [Cu(dppf)(bopy)][BF_4](**3**) in $(CD_3)_2CO$.

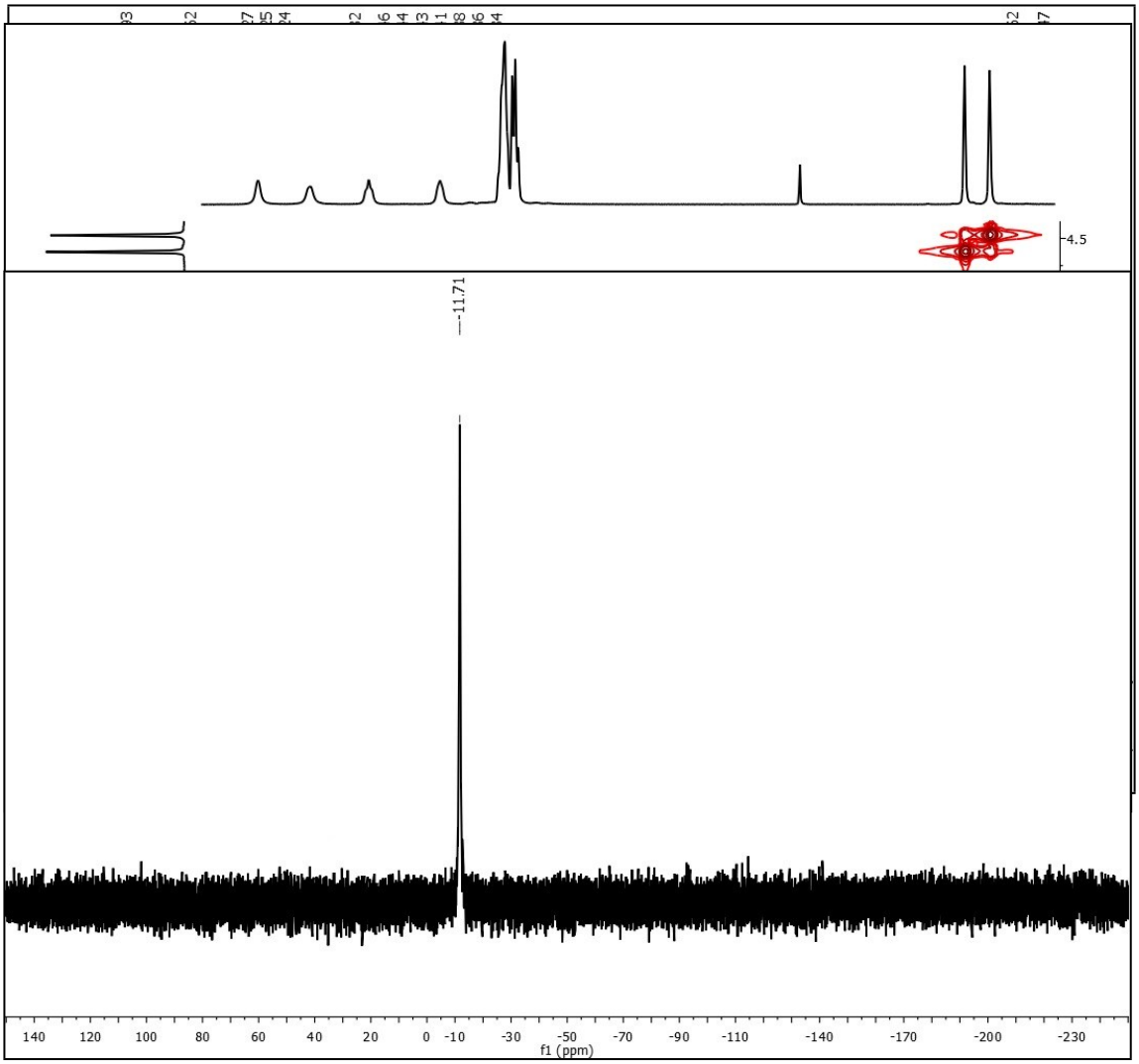


Figure S28 – ^{31}P-NMR spectrum of [Cu(dppf)(dpk)][BF_4](**4**) in $(CD_3)_2CO$.

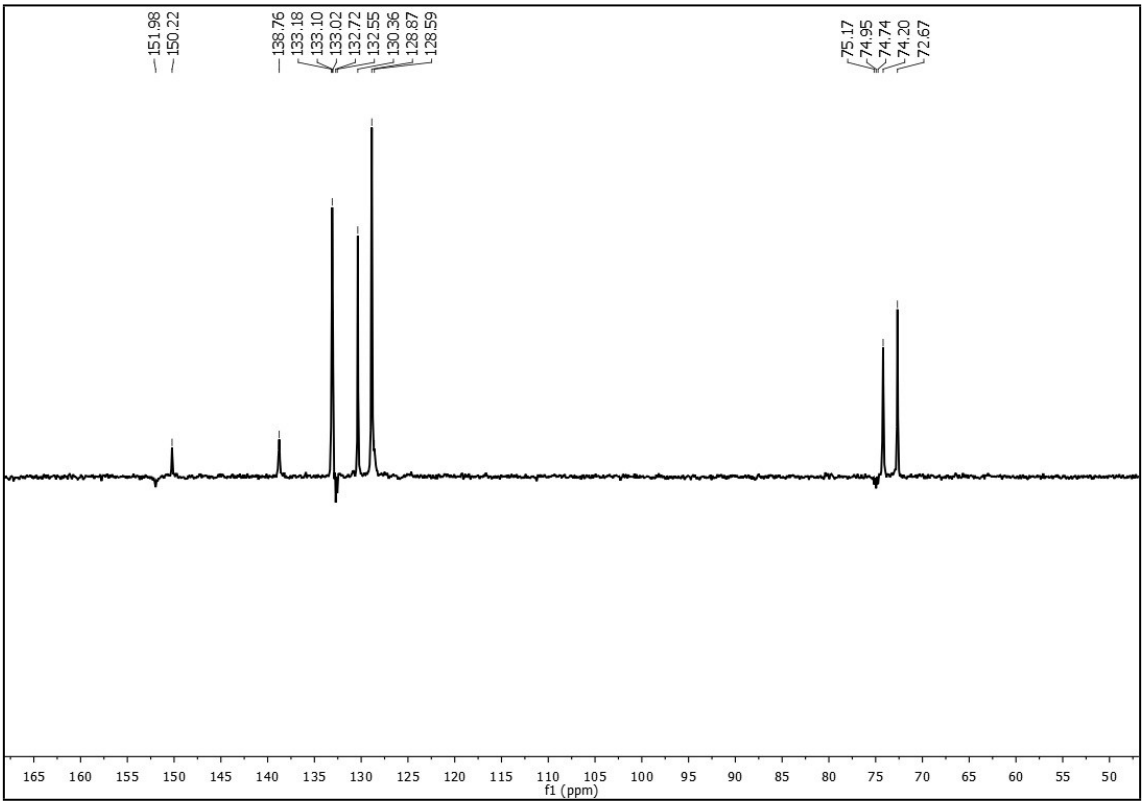


Figure S29 – ^{13}C-NMR spectrum of [Cu(dppf)(dpk)][BF_4](**4**) in $(CD_3)_2CO$.

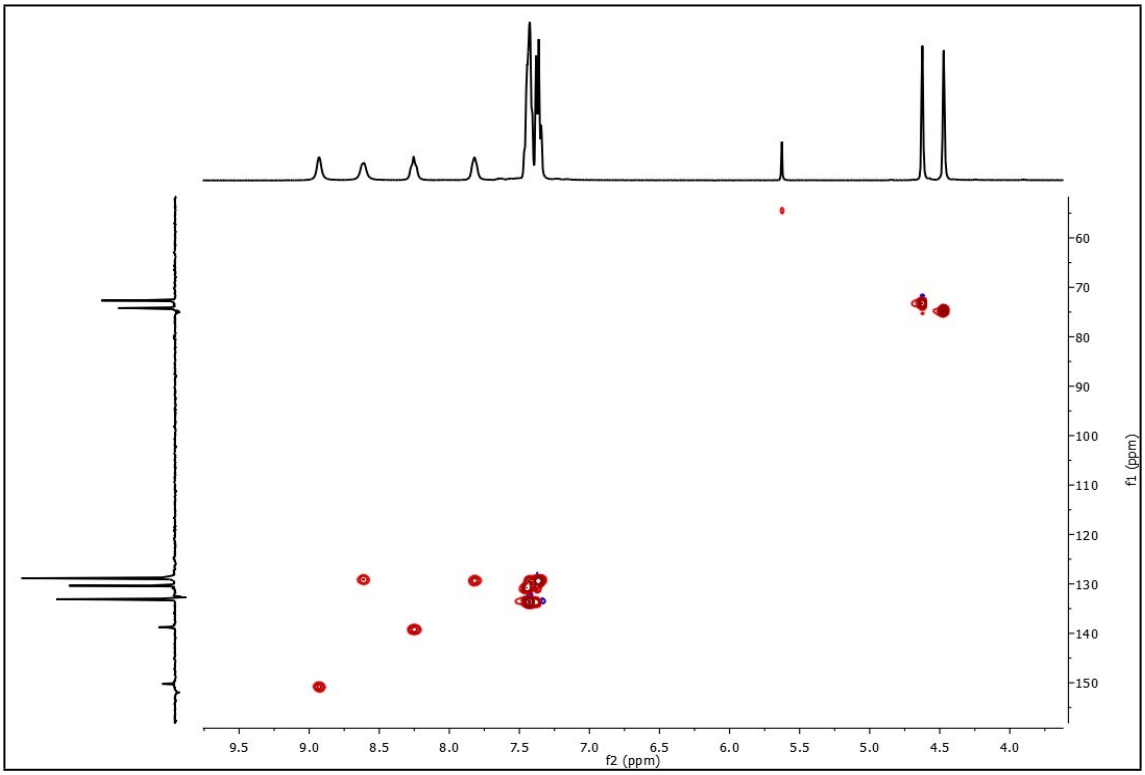


Figure S30 - HSQC-NMR spectrum of [Cu(dppf)(dpk)][BF_4](**4**) in $(CD_3)_2CO$.

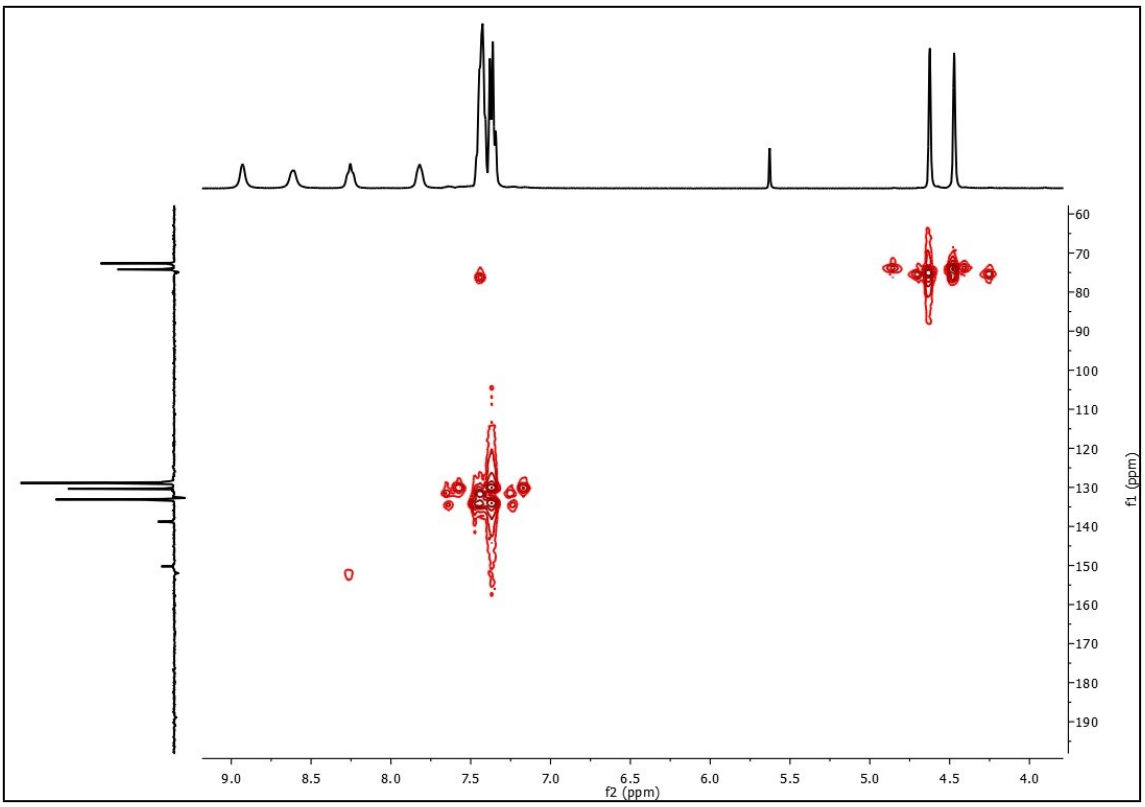


Figure S31 - HMBC-NMR spectrum of [Cu(dppf)(dpk)][BF_4](**4**) in $(CD_3)_2CO$.

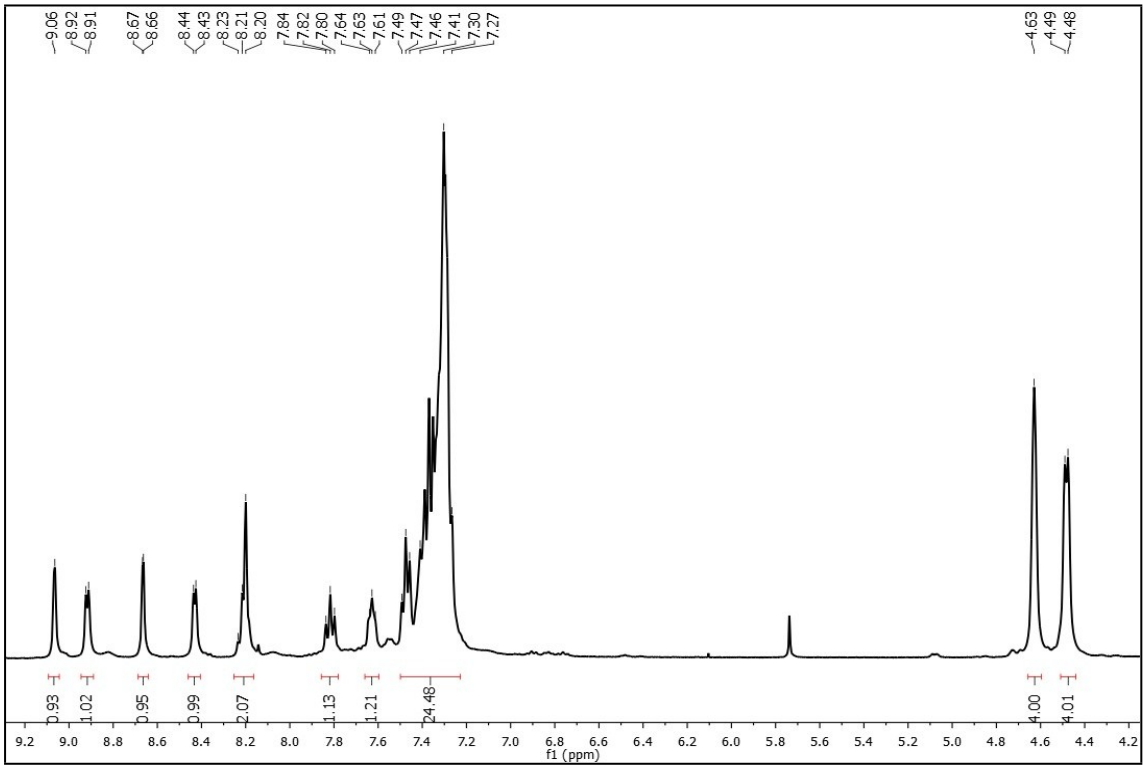


Figure S32 - ^{1}H-NMR spectrum of [Cu(dppf)(dpp)][BF_4](**5**) in $(CD_3)_2CO$, T = -40°C.

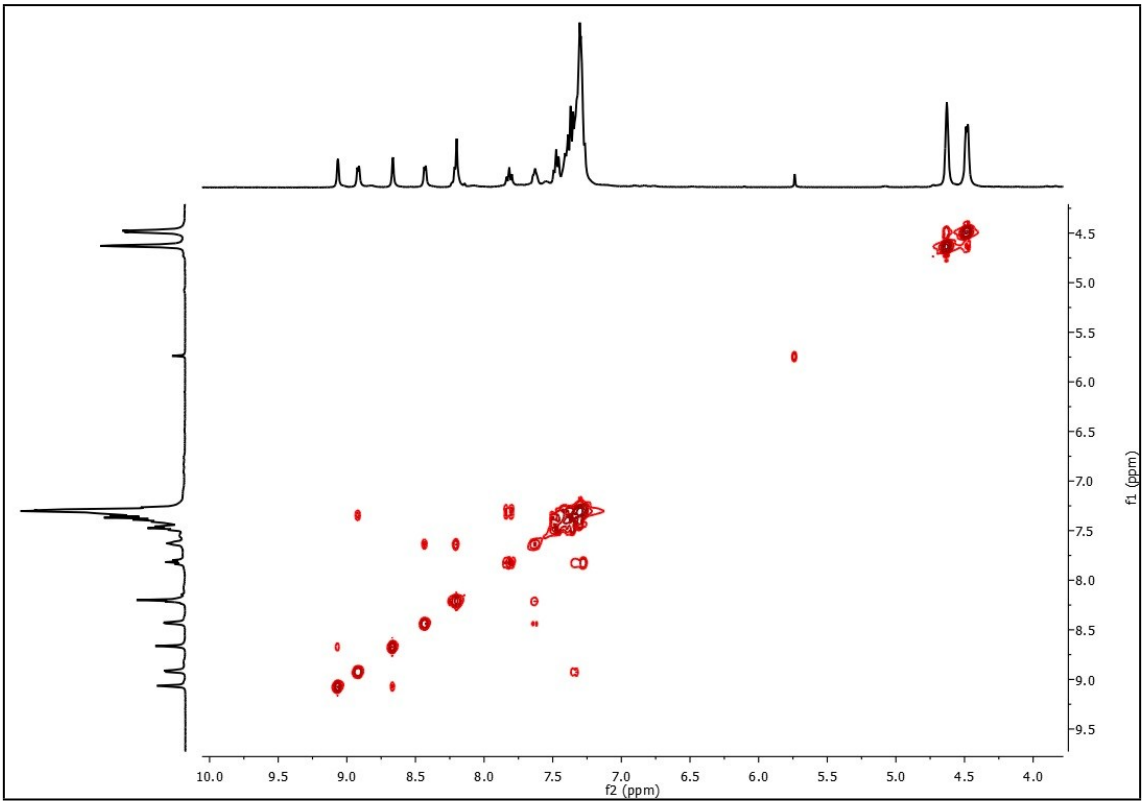


Figure S33 – cosy-NMR spectrum of [Cu(dppf)(dpp)][BF_4](**5**) in $(CD_3)_2CO$, T = -40°C.

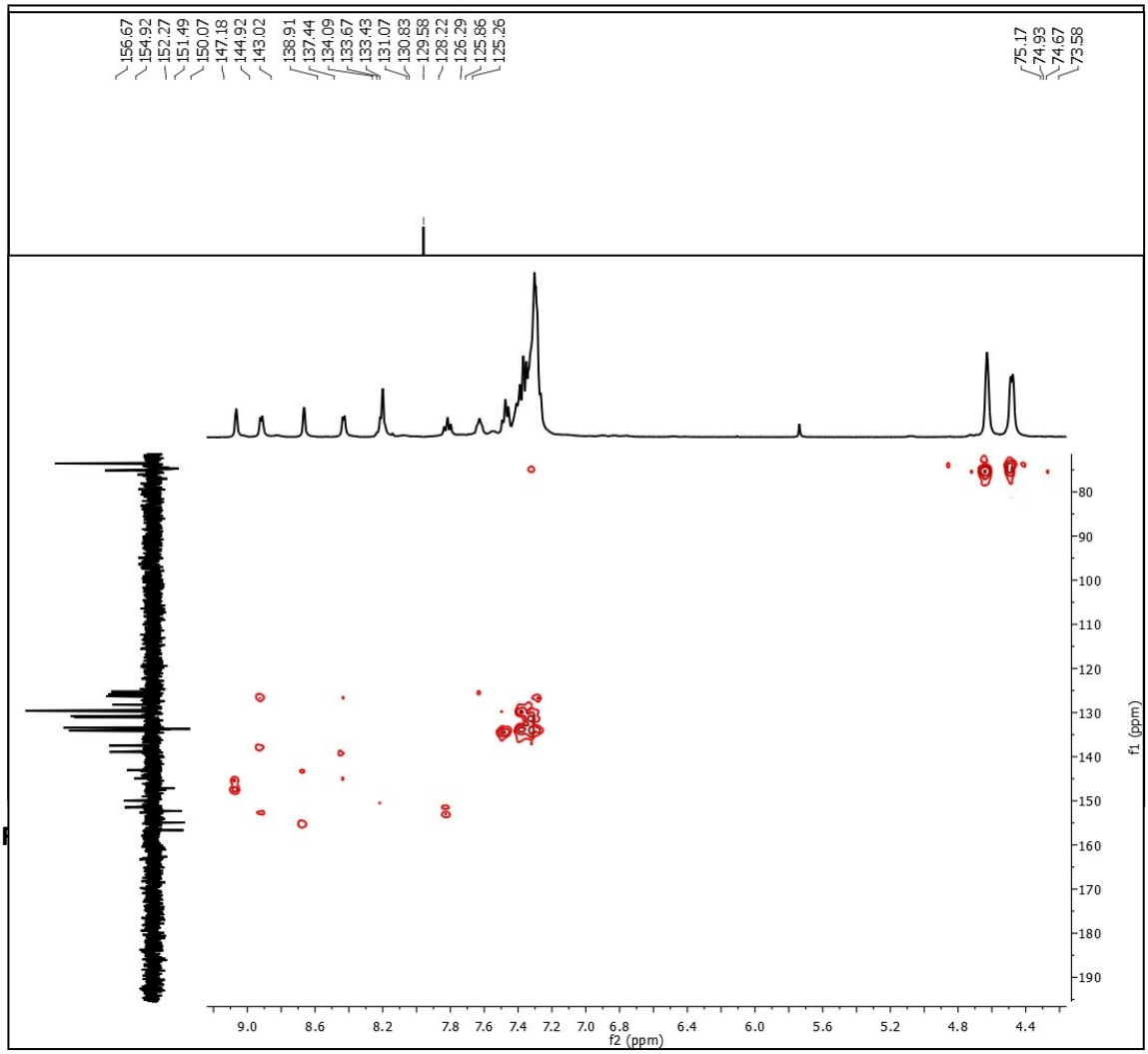


Figure S36 - HSQC-NMR spectrum of [Cu(dppf)(dpp)][BF_4](**5**) in $(CD_3)_2CO$, T = -40°C.

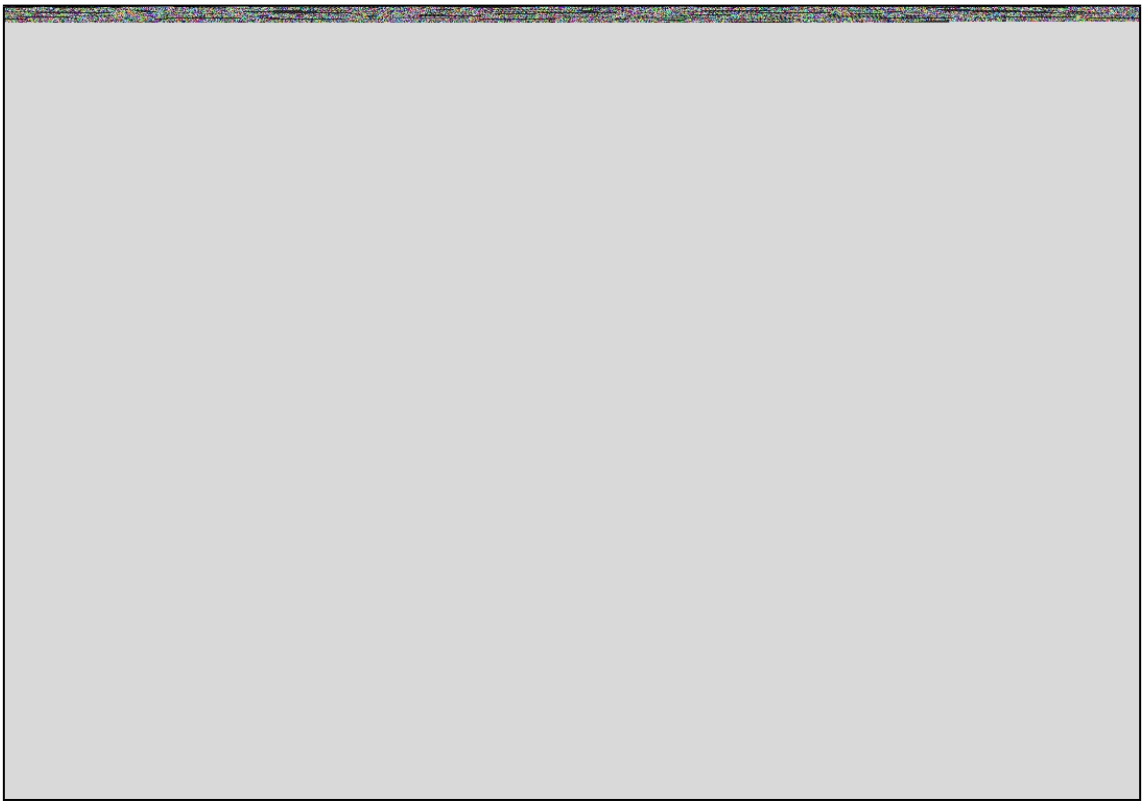


Figure S37 - HMBC-NMR spectrum of [Cu(dppf)(dpp)][BF_4](**5**) in $(CD_3)_2CO$, T = -40°C.

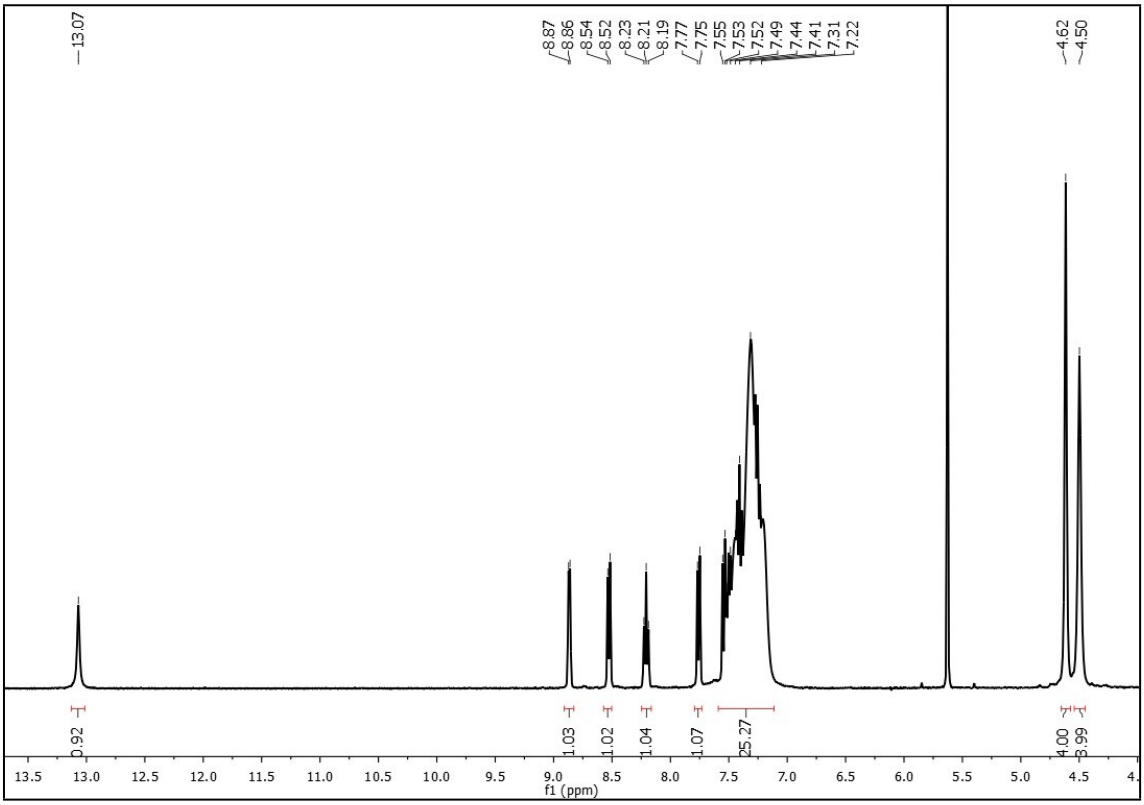


Figure S38 - ^{1}H-NMR spectrum of [Cu(dppf)(pBI)][BF_4](**6**) in $(CD_3)_2CO$.

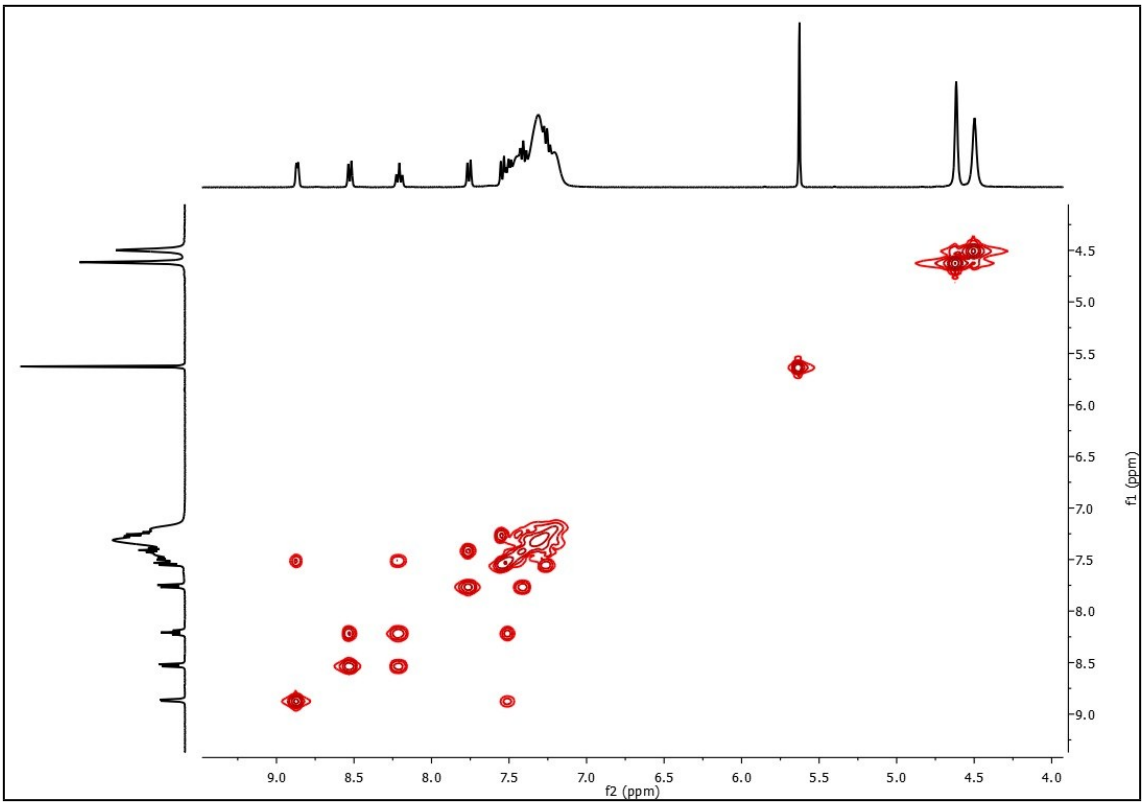


Figure S39 – cosy-NMR spectrum of [Cu(dppf)(pBI)][BF_4](**6**) in $(CD_3)_2CO$.

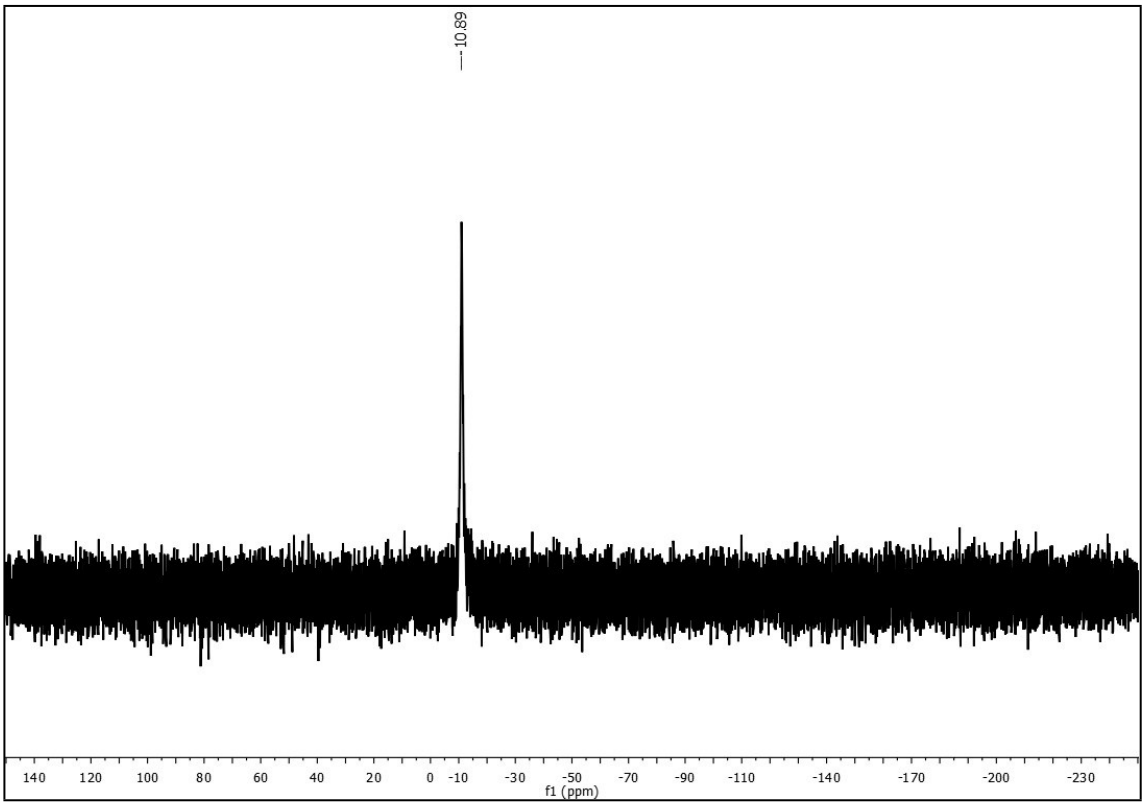


Figure S40 – ^{31}P-NMR spectrum of [Cu(dppf)(pBI)][BF_4](**6**) in $(CD_3)_2CO$.

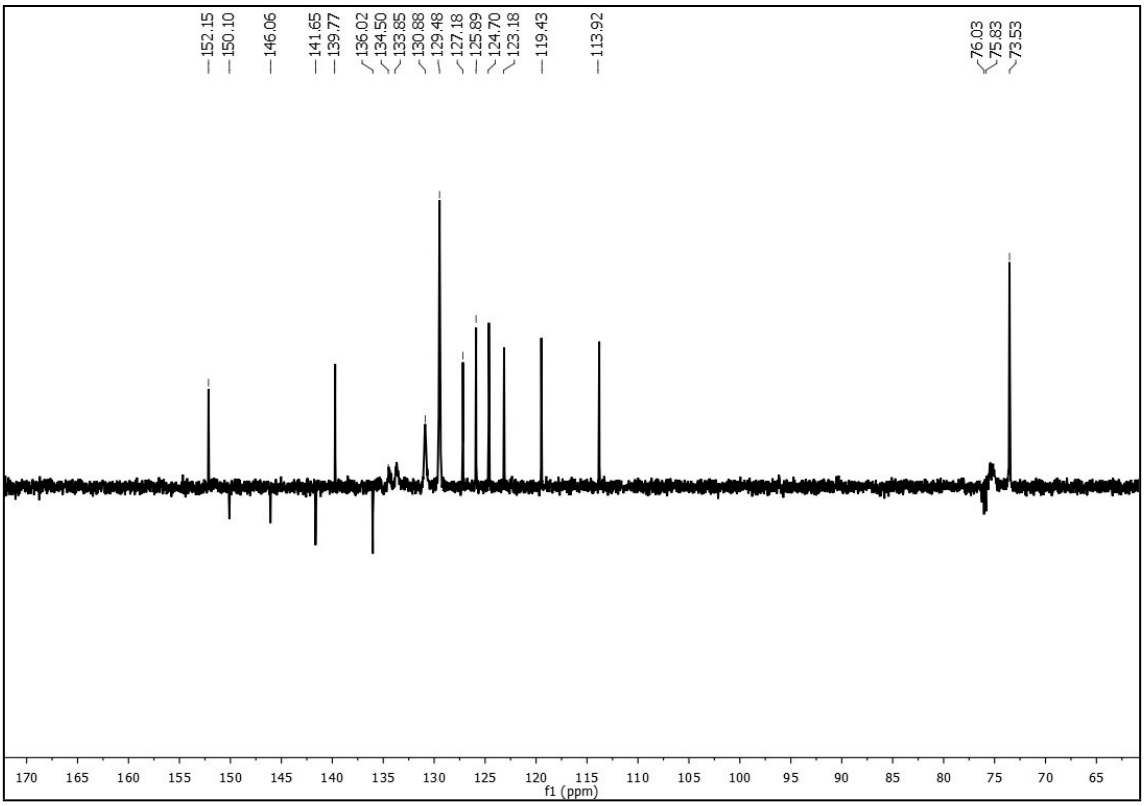


Figure S41 – ^{13}C-NMR spectrum of [Cu(dppf)(pBI)][BF_4](**6**) in $(CD_3)_2CO$.

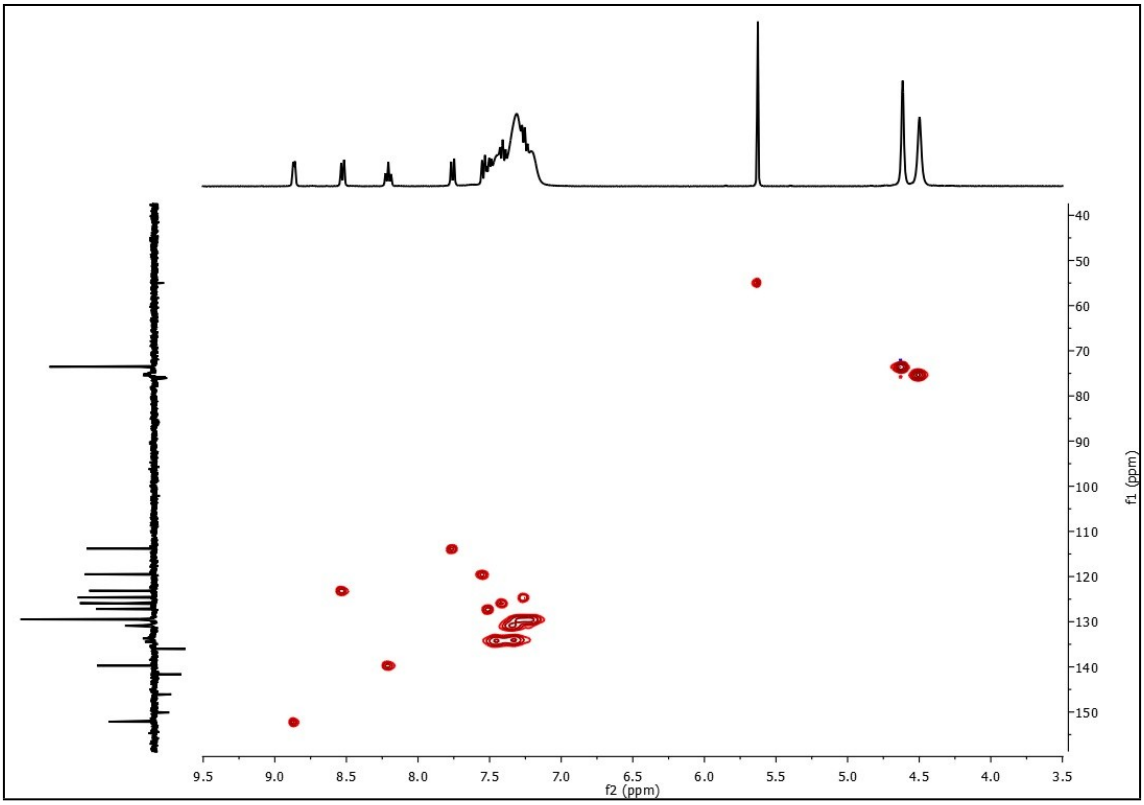


Figure S42 - HSQC-NMR spectrum of [Cu(dppf)(pBI)][BF_4](**6**) in $(CD_3)_2CO$.

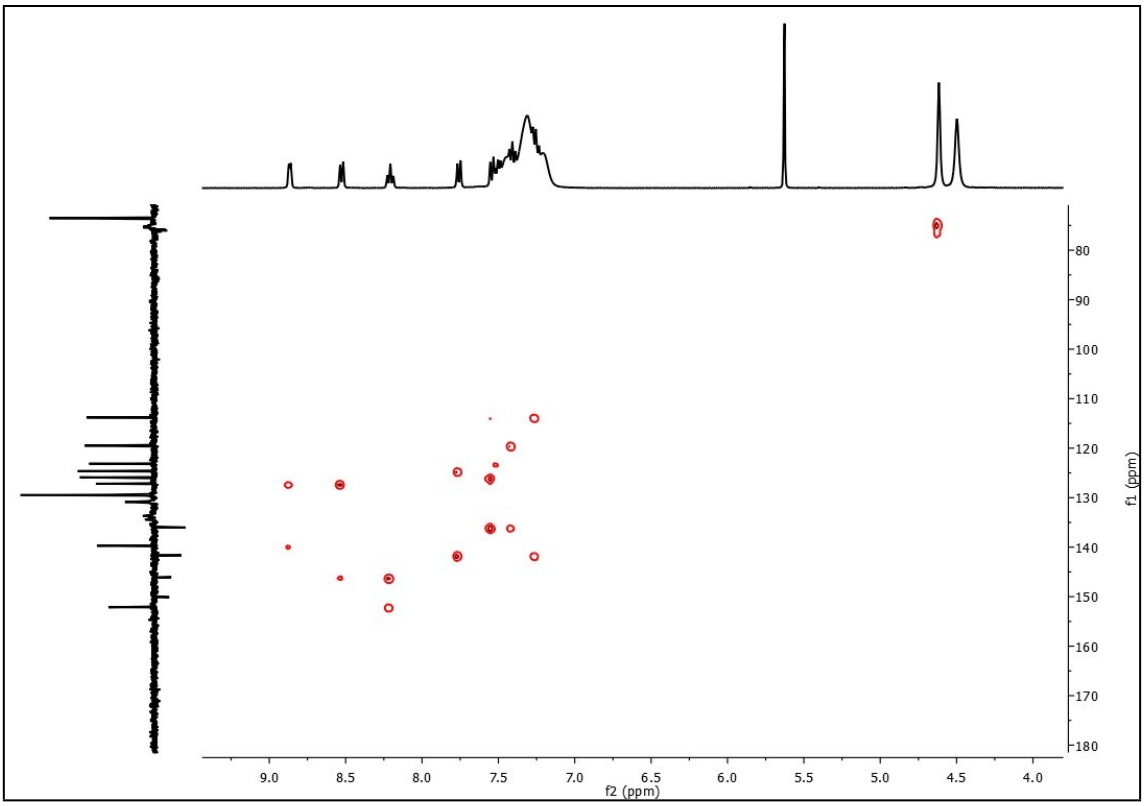


Figure S43 - HMBC-NMR spectrum of [Cu(dppf)(pBI)][BF_4](**6**) in $(CD_3)_2CO$.

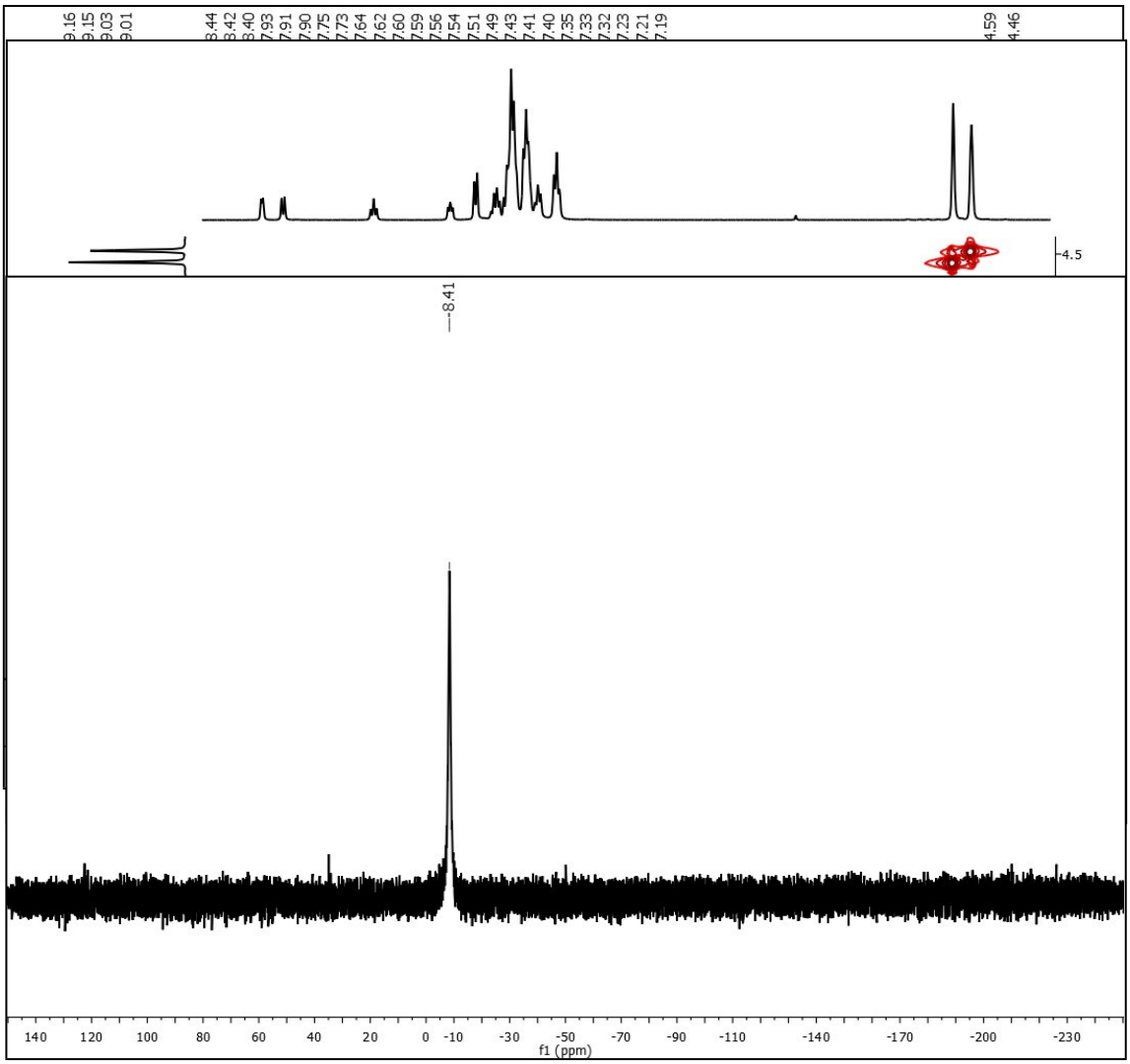


Figure S46 – ^{31}P-NMR spectrum of [Cu(dppf)(dpytz)][BF_4](**7**) in $(CD_3)_2CO$.

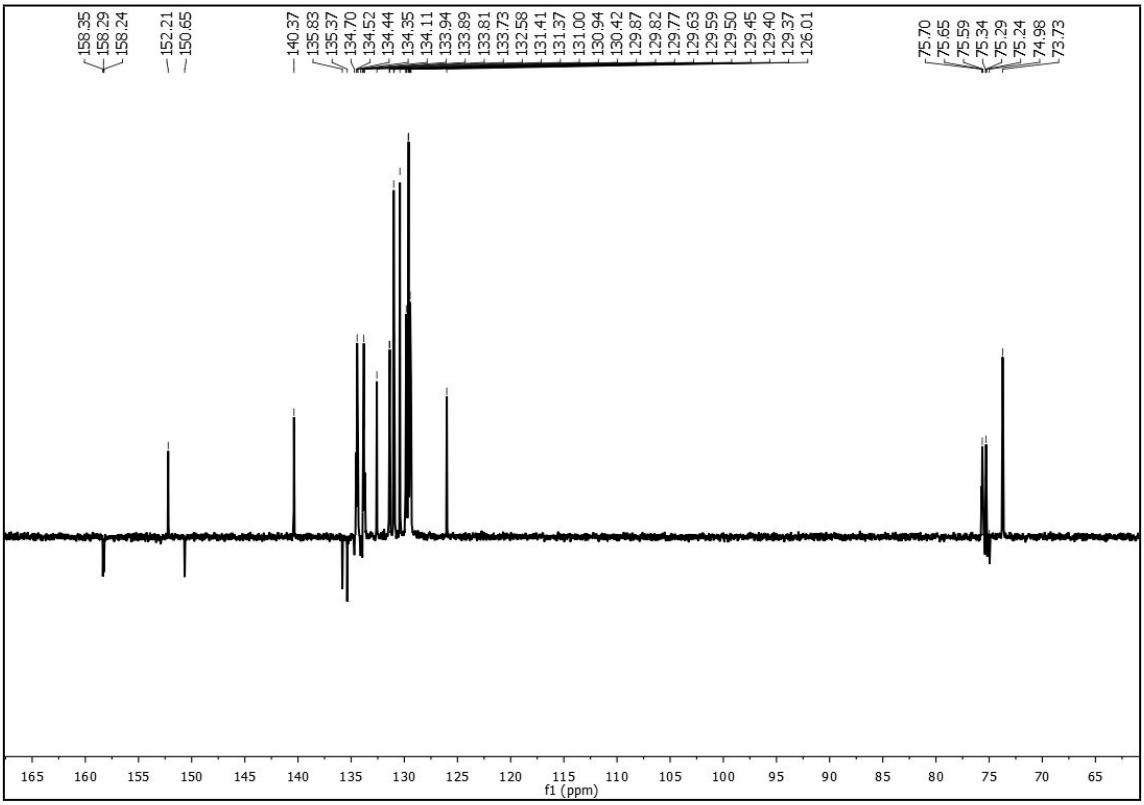


Figure S47 – ^{13}C-NMR spectrum of [Cu(dppf)(dpytz)][BF_4](**7**) in $(CD_3)_2CO$.

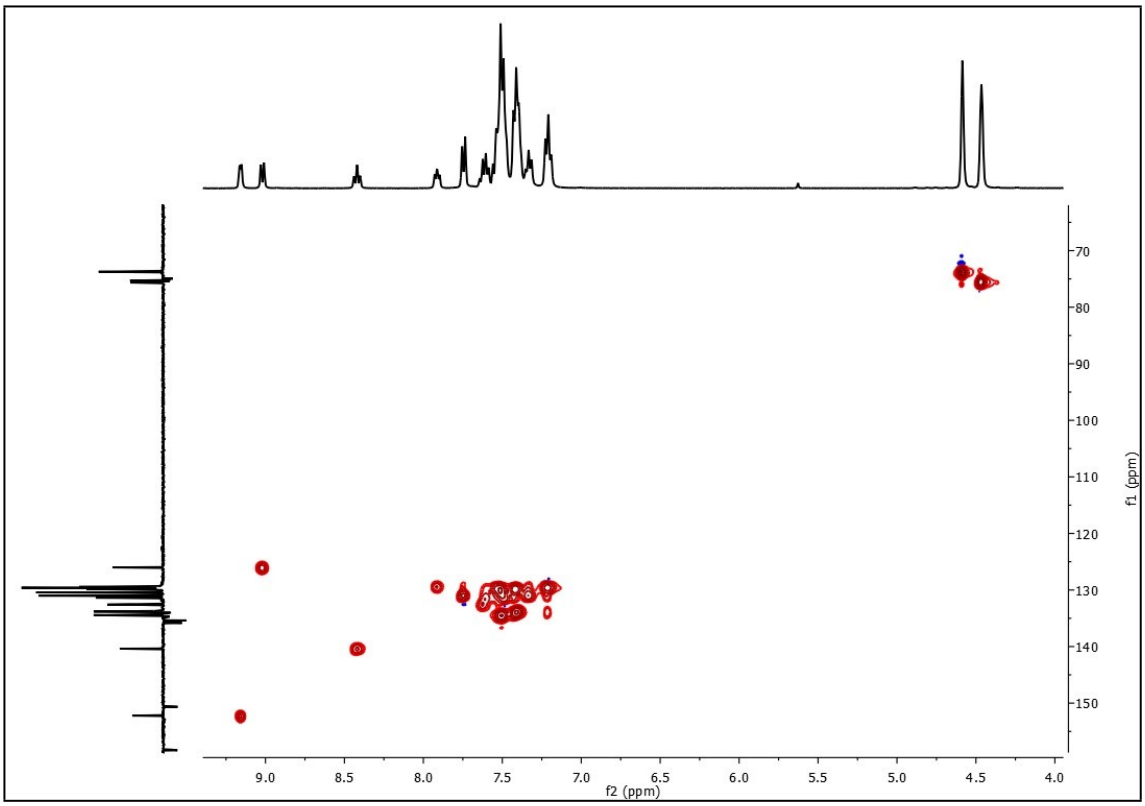


Figure S48 - HSQC-NMR spectrum of [Cu(dppf)(dpytz)][BF_4](**7**) in $(CD_3)_2CO$.

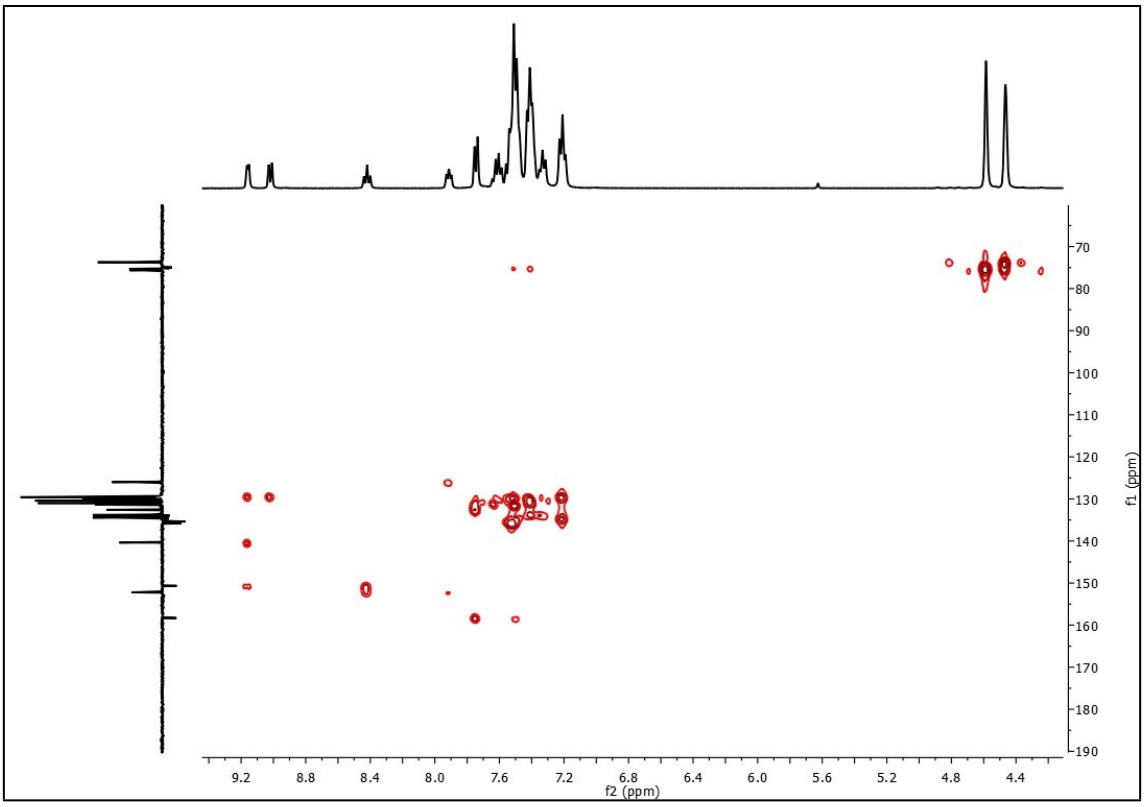


Figure S49 - HMBC-NMR spectrum of [Cu(dppf)(dpytz)][BF_4](**7**) in $(CD_3)_2CO$.

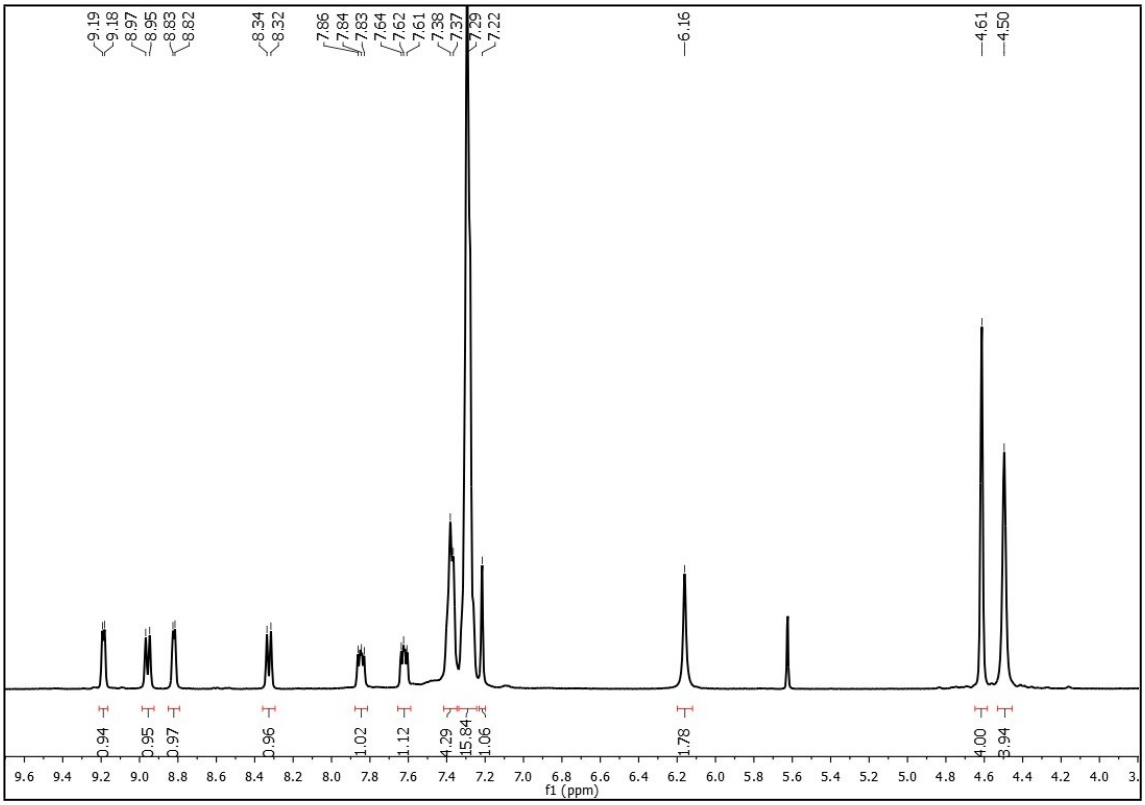


Figure S50 - ^{1}H-NMR spectrum of [Cu(dppf)(5-Aphen)][BF_4](**8**) in $(CD_3)_2CO$.

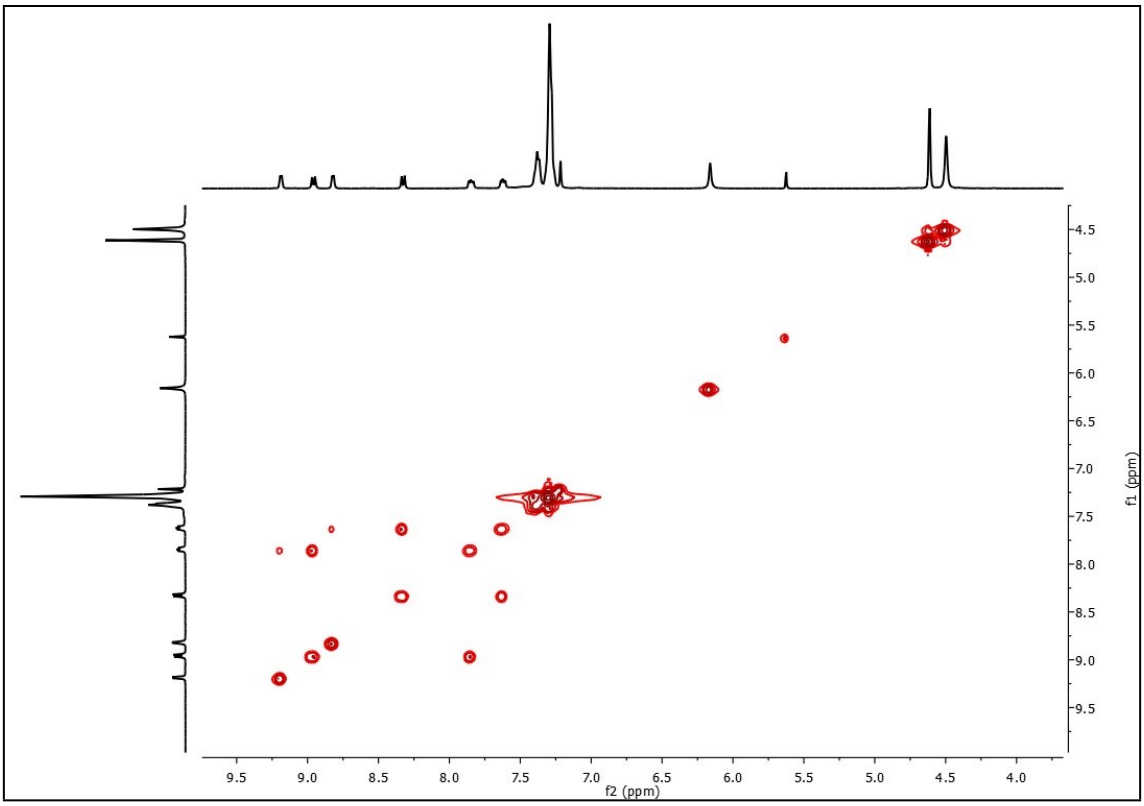


Figure S51 – cosy-NMR spectrum of [Cu(dppf)(5-Aphen)][BF_4](**8**) in $(CD_3)_2CO$.

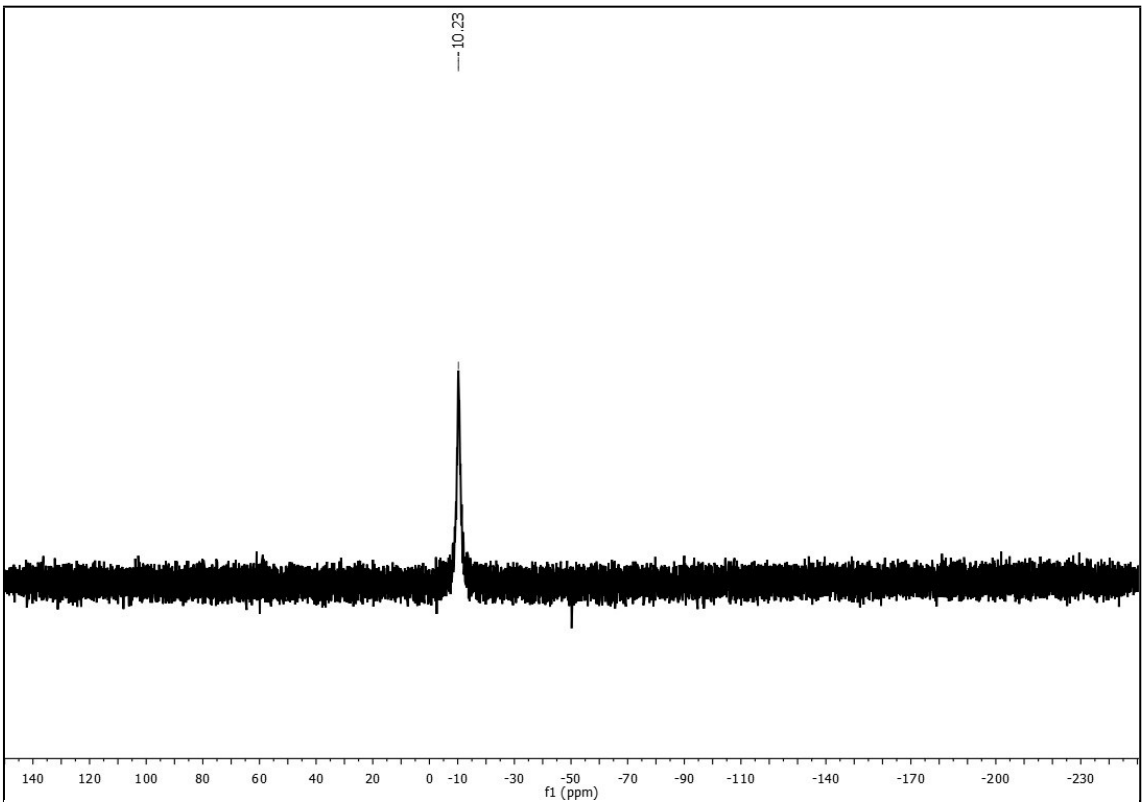


Figure S52 – ^{31}P-NMR spectrum of [Cu(dppf)(5-Aphen)][BF_4](**8**) in $(CD_3)_2CO$.

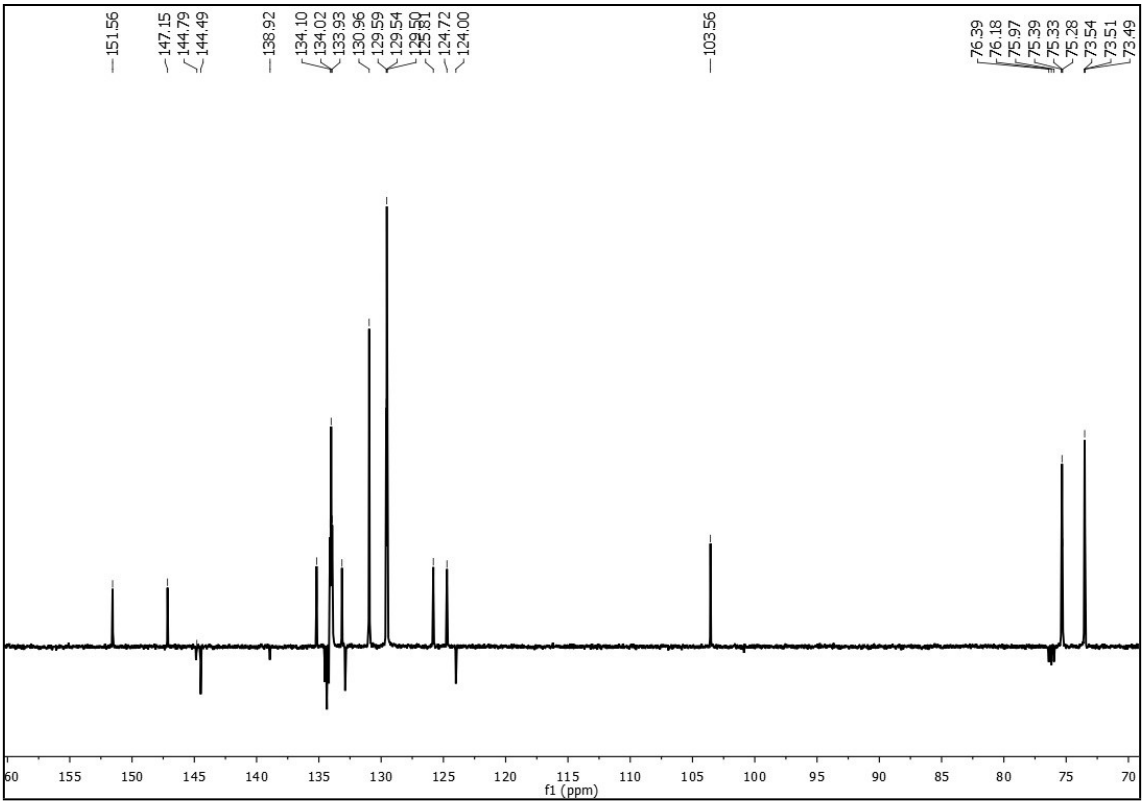


Figure S53 - ^{13}HC-NMR spectrum of [Cu(dppf)(5-Aphen)][BF_4](**8**) in $(CD_3)_2CO$.

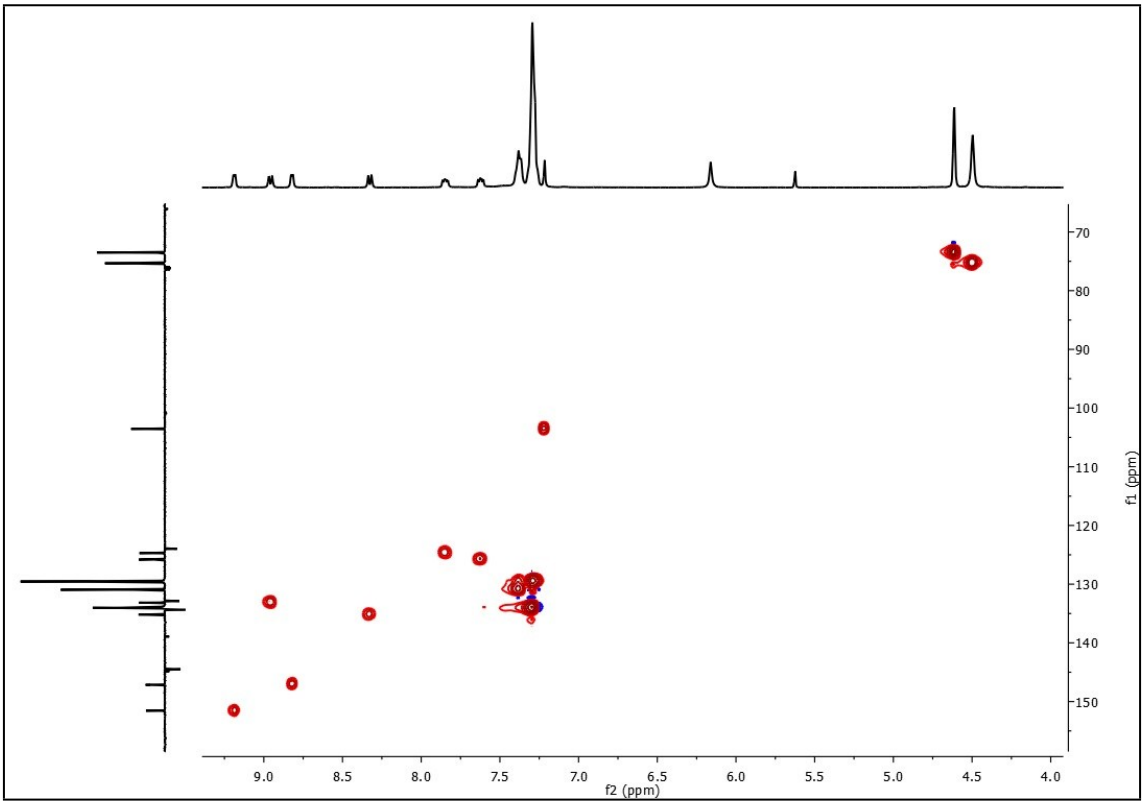


Figure S54 - HSQC-NMR spectrum of [Cu(dppf)(5-Aphen)][BF_4](**8**) in $(CD_3)_2CO$.

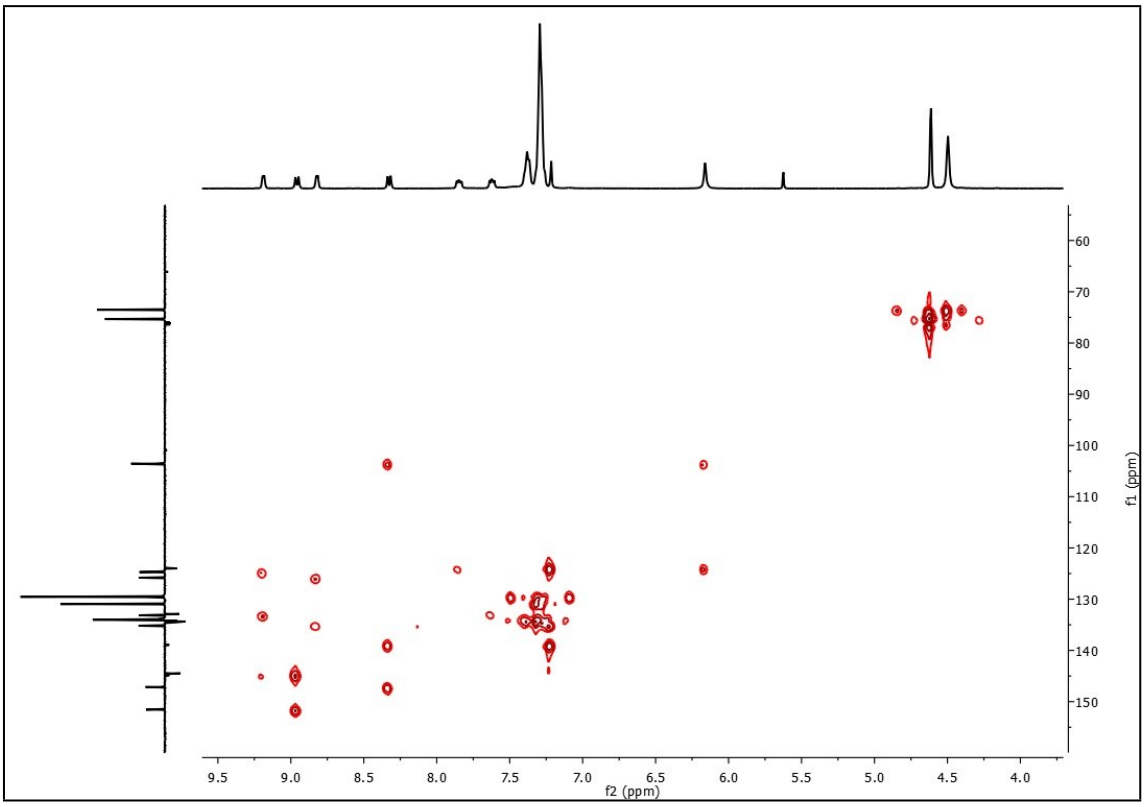


Figure S55 - HMBC-NMR spectrum of [Cu(dppf)(5-Aphen)][BF_4](**8**) in $(CD_3)_2CO$.

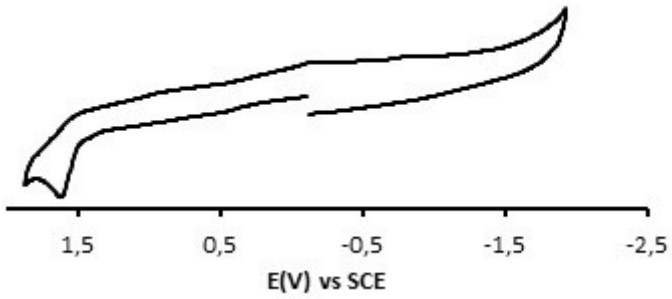


Figure S56 - Cyclic voltammogram of ligand pbt (L2) in acetonitrile (scan rate:

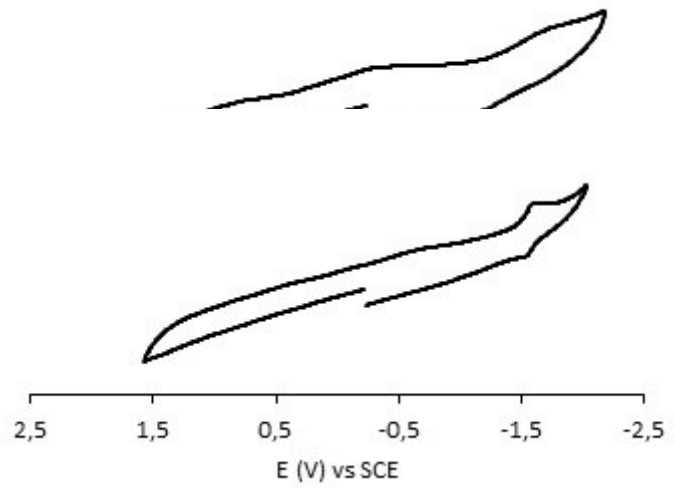


Figure 'scan rate: 0

Figure S58 - Cyclic voltammogram of ligand dpytz (L7) in acetonitrile (scan rate: 0.2 $V.s^{-1}$).

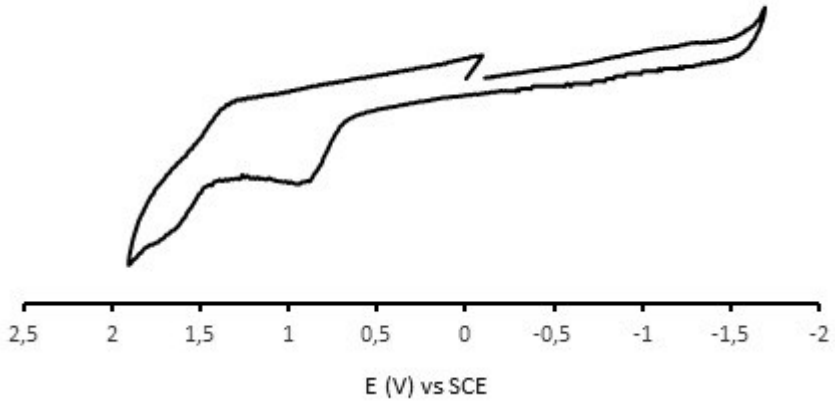


Figure S59 - Cyclic voltammogram of ligand 5-Aphen (L8) in acetonitrile (scan rate: 0.2 $V.s^{-1}$).

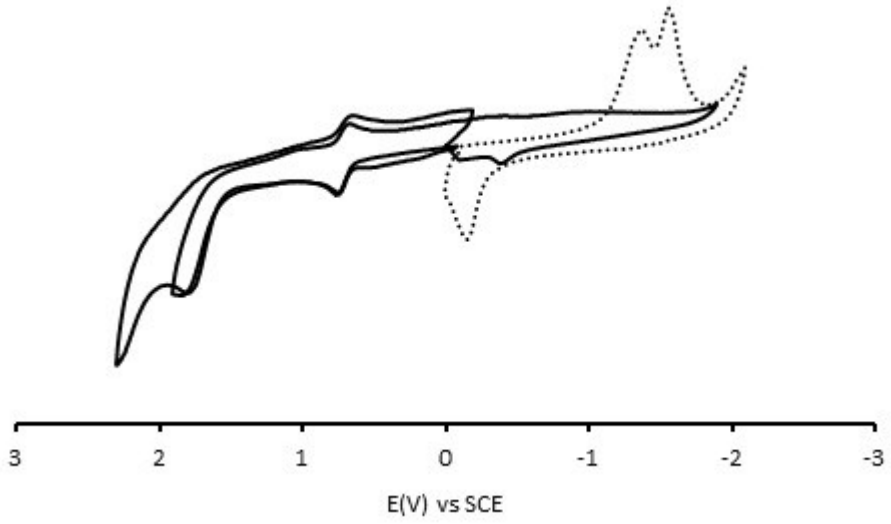


Figure S60 - Cyclic voltammogram of complex $[Cu(dppf)(NCMe)_2][BF_4]$ in acetonitrile (scan rate: 0.2 V.s-1), showing the redutive processes present in the first scan (dashed line).

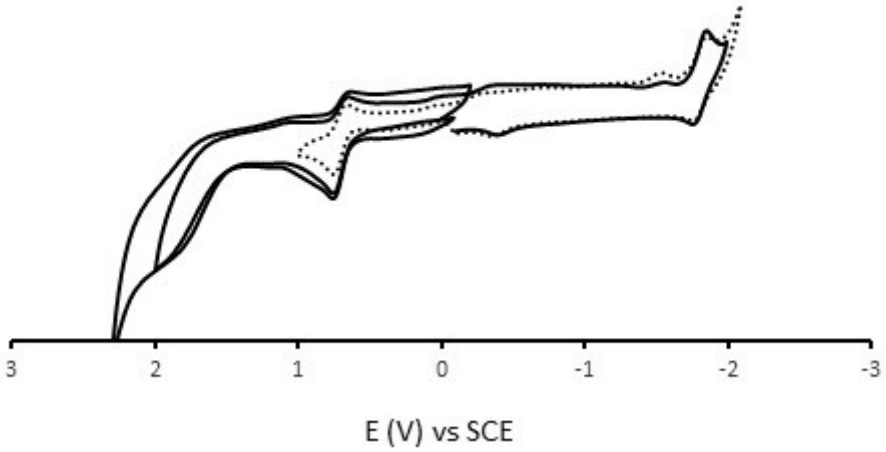


Figure S61 - Cyclic voltammogram of complex [Cu(dppf)(Me_2bpy)][BF_4], **<u>1</u>** in acetonitrile (scan rate: 0.2 V.s^{-1}), showing the isolated Fe(II)/Fe(III) and Me2bpy processes (dashed line).

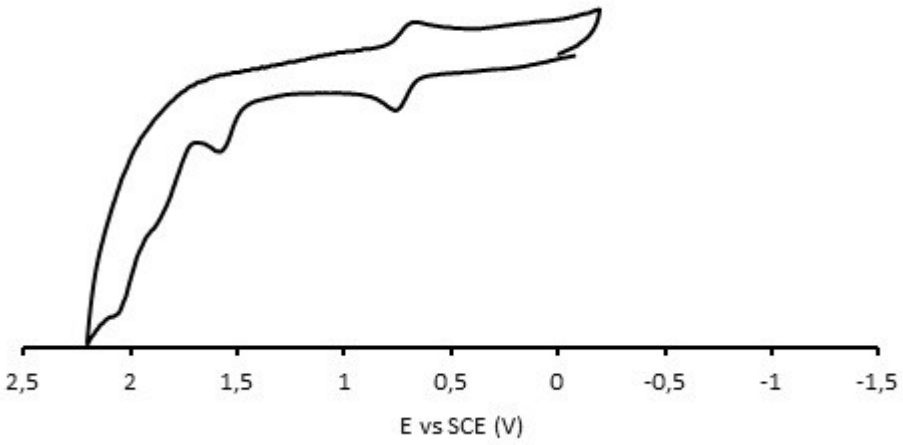
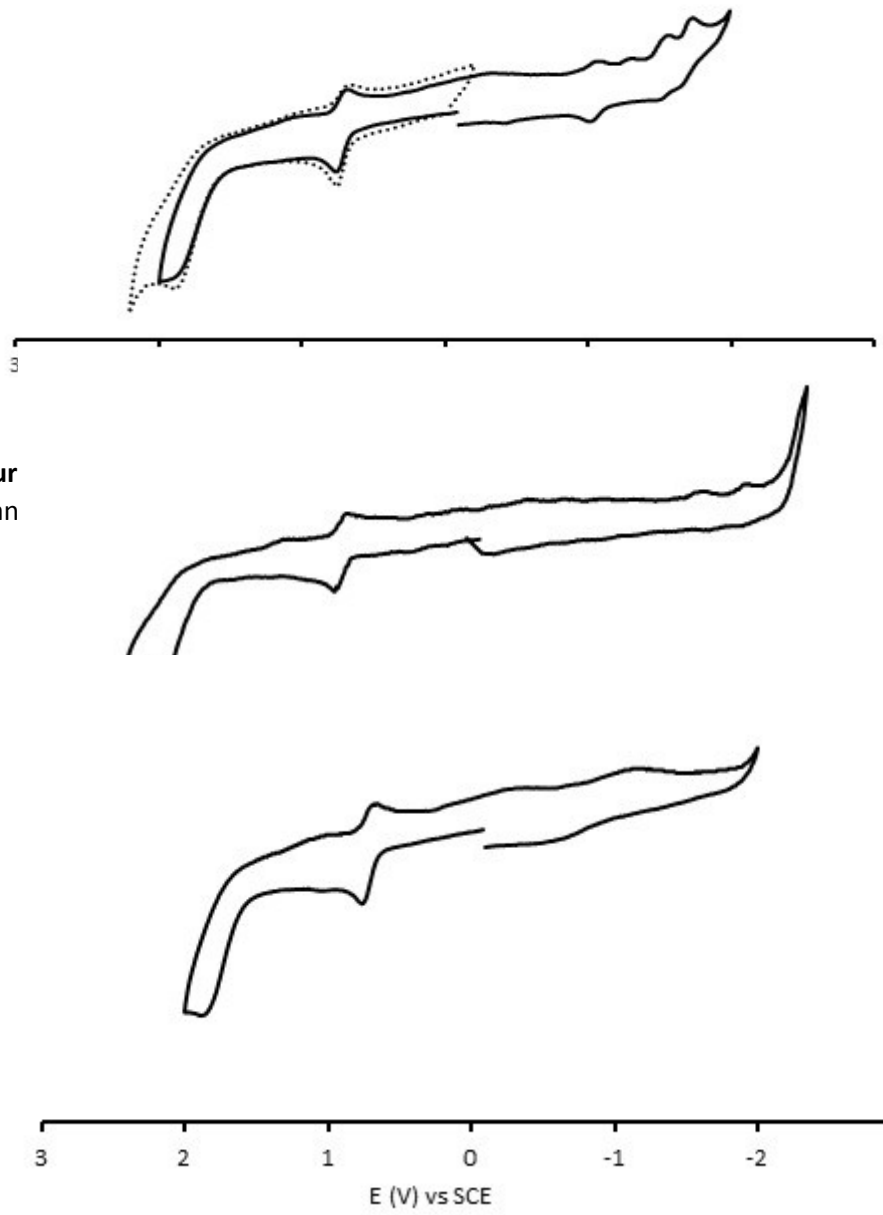


Figure S62 - Cyclic voltammogram of complex [Cu(dppf)(pbt)][BF_4], **2** in acetonitrile (scan rate: 0.2 $V.s^{-1}$).



Figur
(scan

$V.s^{-1}$).

Figure S65 - Cyclic voltammogram of complex [Cu(dppf)(pBI)][BF_4], **6** in acetonitrile (scan rate: 0.2 $V.s^{-1}$).

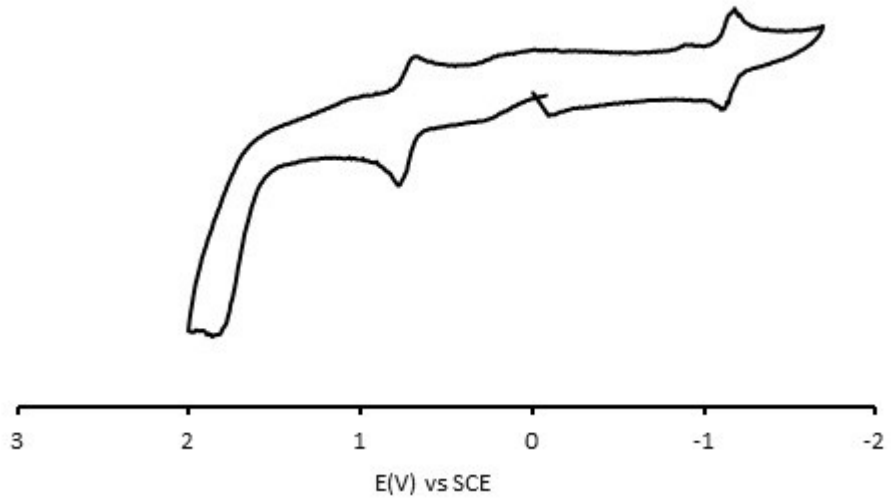


Figure S66 - Cyclic voltammogram of complex [Cu(dppf)(dpytz)][BF_4], **7** in acetonitrile (scan rate: 0.2 $V.s^{-1}$).

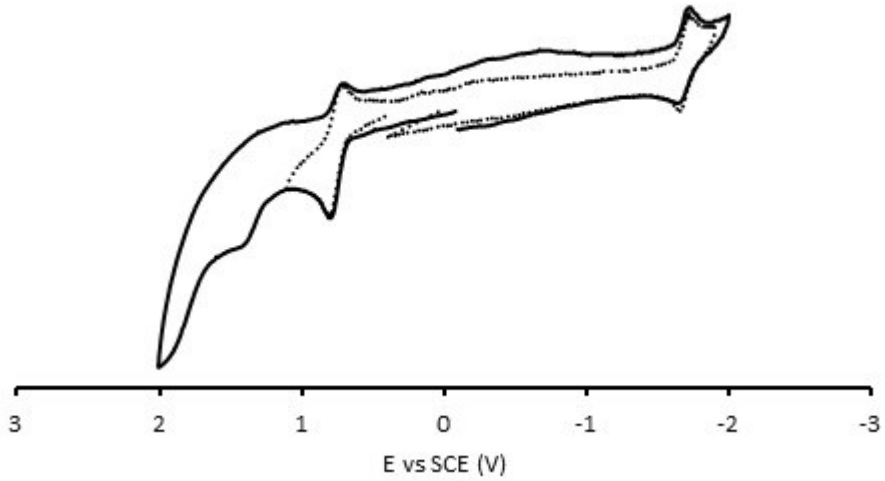


Figure S67 - Cyclic voltammogram of complex [Cu(dppf)(5-Aphen)][BF_4], **8** in acetonitrile (scan rate: 0.2 $V.s^{-1}$), showing the isolated Fe(II)/Fe(III) and phenanthroline based processes (dashed line).

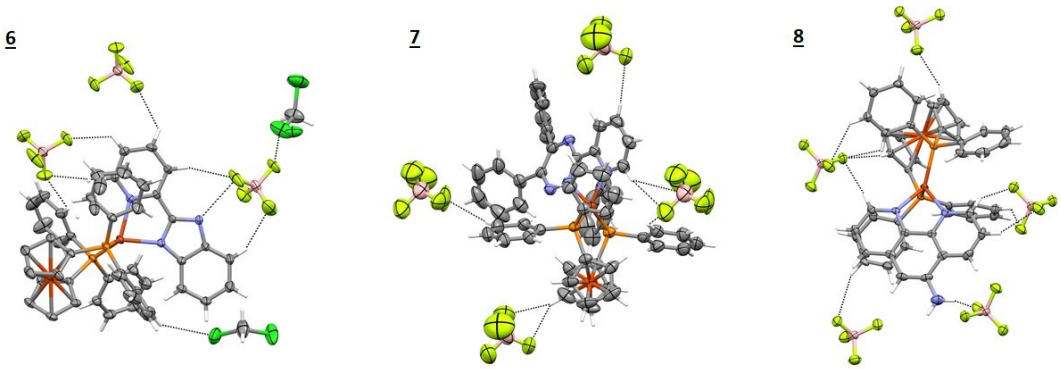


Figure S68 - Intermolecular interactions for complexes $[Cu(dppf)(pBI)]^+$ **6**, $[Cu(dppf)(dpytz)]^+$ **7** and $[Cu(dppf)(5\text{-}Aphen)]^+$ **8**.

Table S1 - Hydrogen bonds for complexes $[Cu(dppf)(pBI)]^+$ **6**, $[Cu(dppf)(dpytz)]^+$ **7** and $[Cu(dppf)(5\text{-}Aphen)]^+$ **8**.

D --- H....A	[ARU]	D - H	H...A	D...A	D - H...A
Complex 6					
C3 --H3 ..F4	1-x, -1/2+y, 1/2-z	0.95	2.45	3.1437(1)	130
C8 --H8 ..F4	x, y, z	0.95	2.39	3.2507(1)	150
C19--H19 ..Cl2	1-x, 1-y, 1-z	0.95	2.82	3.5607(1)	135
C19--H19 ..Cl2A	1-x, 1-y, 1-z	0.95	2.82	3.4672(1)	126
C22 --H22 ..F1	x, 1/2-y, 1/2+z	0.95	2.47	3.3209(1)	149
C47 --H47A ..F3	x, y, z	0.99	2.42	3.0124(1)	118
Complex 7					
C28--H28 ..F2	1/2+x, 1/2-y, -1/2+z	0.93	2.54	3.4465(2)	165
Complex 8					
N3 --H3B ..F1	1-x, -y, -z	0.88	2.21	2.9420(2)	140
C1 --H1 ..F4	x, y, z	0.95	2.31	3.1019(2)	140
C10 --H10 ..F2	-1+x, y, z	0.95	2.30	3.2416(2)	169
C16 --H16 ..F3	1/2-x, 1/2+y, 1/2-z	0.95	2.46	3.3456(2)	155
C22 --H22 ..F2	-1+x, y, z	0.95	2.45	3.2691(2)	145
C33 --H33 ..F1	-1+x, y, z	0.95	2.55	3.2856(2)	135

Table S2 - IC_{50} values found for compounds **1**–**8**, precursor, free ligands and cisplatin in the breast adenocarcinoma MCF7 and MDAMB231 and primary healthy fibroblasts cells (24 h, 37 °C).

Compound	IC_{50} (μM)		
	MDAMB231	**MCF7**	**Fibroblasts**
$[Cu(dppf)(NCMe)_2][BF_4]$	34.1 ± 7.0	18.6 ± 4.1	
$[Cu(dppf)(Me2bpy)][BF_4]$ **1**	2.92 ± 1.5	1.39 ± 0.4	1.5 ± 0.05
$[Cu(dppf)(pbt)][BF_4]$ **2**	35.5 ± 5.5	8.78 ± 1.5	20 ± 0.5
$[Cu(dppf)(bopy)][BF_4]$ **3**	19.1 ± 4.6	7.53 ± 1.6	15 ± 0.2
$[Cu(dppf)(dpk)][BF_4]$ **4**	14.3 ± 2.5	6.36 ± 1.4	10 ± 0.1
$[Cu(dppf)(dpp)][BF_4]$ **5**	3.2 ± 1.4	1.80 ± 0.5	10 ± 0.2
$[Cu(dppf)(pBI)][BF_4]$ **6**	2.61 ± 0.9	2.51 ± 0.9	8 ± 0.1
$[Cu(dppf)(dpytz)][BF_4]$ **7**	1.72 ± 0.4	1.48 ± 0.3	5 ± 0.08
$[Cu(dppf)(5\text{-}Aphen)][BF_4]$ **8**	1.21 ± 0.2	0.77 ± 0.1	1 ± 0.05
Cisplatin		59 ± 12	
Dppf	> 100	51.6 ± 10	
Me2bpy		> 100	
pbt		55.1±7.5	
bopy		> 100	

dpk	> 100
dpp	> 100
pBI	> 100
dpytz	> 100
5-Aphen	> 100
$[Cu(dppe)(NCMe)_2][BF_4]$[27]	1.38 ± 0.71
$[Cu(PPh_3)_2(NCMe)_2][BF_4]$[26]	13.4 ± 5.3
$[Cu(dppe)(Me2bpy)][BF_4]$[27]	0.46 ± 0.14
$[Cu(dppe)(dpp)][BF_4]$[27]	0.80 ± 0.34
$Cu(dppe)(dpytz)][BF_4]$[27]	0.51 ± 0.08
$[Cu(PPh_3)_2(pbt)][BF_4]$[26]	6.6± 2.2
$[Cu(PPh_3)_2(bopy)][BF_4]$[26]	11.6 ± 3.9
$[Cu(PPh_3)_2(dpk)][BF_4]$[26]	7.6 ± 3.2
$[Cu(PPh_3)_2(dpp)][BF_4]$[26]	17.7 ± 5.5