

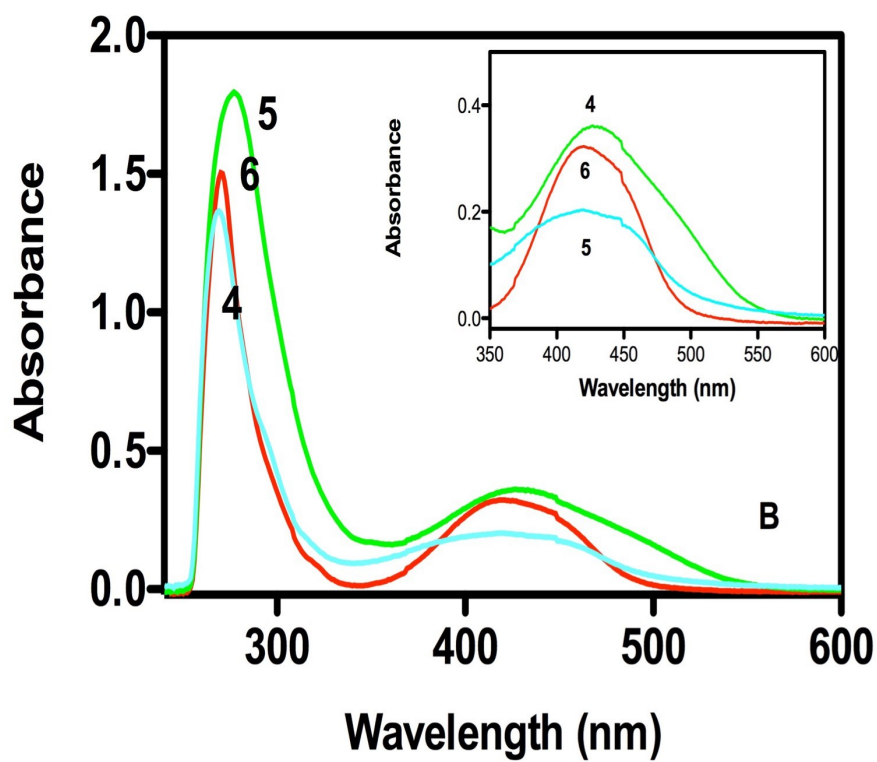
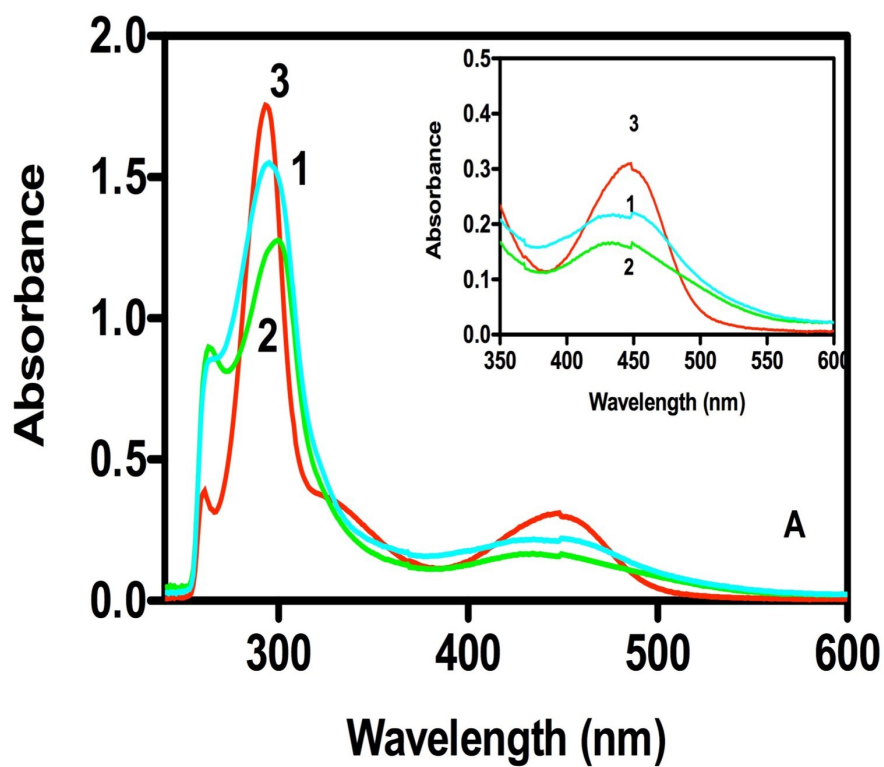
## SUPPLEMENTARY MATERIAL

### **Ru(II)-(PTA) and –mPTA complexes with N<sub>2</sub>-donor Ligands Bipyridyl and Phenanthroline and their Antiproliferative Activities on Human Multiple Myeloma Cell Lines.**

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KEYWORDS. Ruthenium, PTA, mPTA, Metal-based drugs.



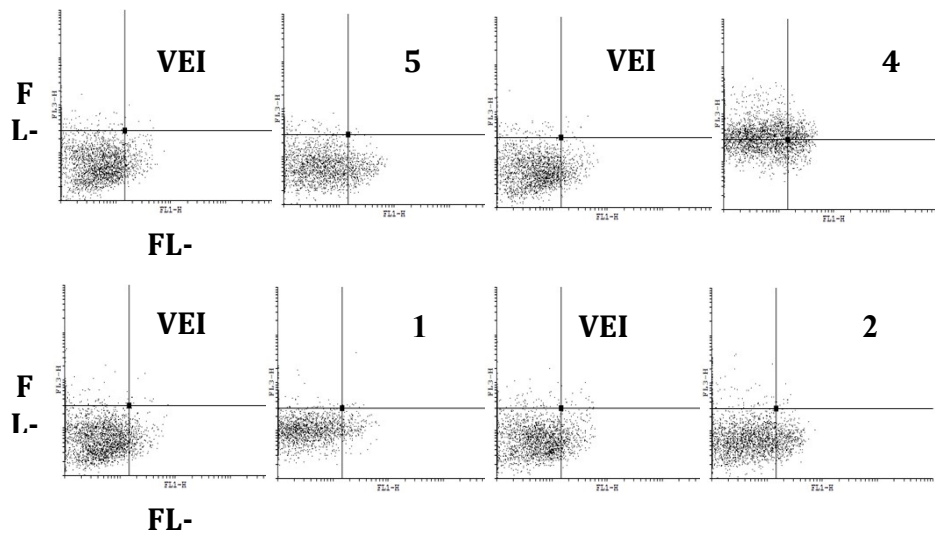
**Figure 1S.** Absorption spectral traces of complexes 1-6 in DMSO. Panel A, complex 1(95  $\mu$ M), complex 2(40  $\mu$ M), complex 3(45  $\mu$ M). Panel B, complex 4(30  $\mu$ M), complex 5(60  $\mu$ M), complex 6(70  $\mu$ M).

**Table 1S.** Wavelength ( $\lambda_{\max}$ ) and molar absorption values ( $\epsilon$ ) of different complexes in DMSO.

Complex	$\lambda_{\max}$ (nm)	$\epsilon$ ( $M^{-1} \text{ cm}^{-1}$ )
<b>1</b>	296	23306
	434	1709
<b>2</b>	301	18543
	437	3729
<b>3</b>	294	24146
	443	4269
<b>4</b>	270	26100
	422	6283
<b>5</b>	271	21750
	422	2483
<b>6</b>	273	28702
	404	6164

**RPMI**

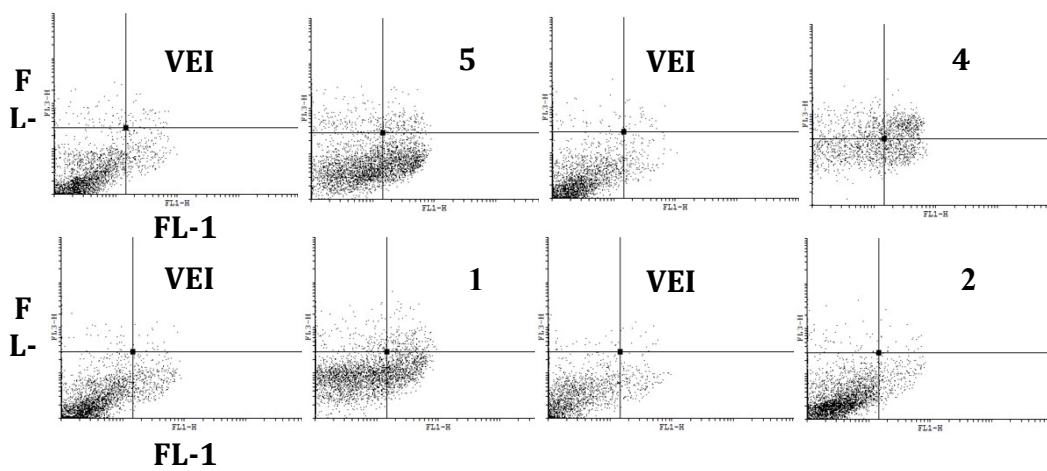
COMPOUND ( $\mu\text{M}$ )	AnnV <sup>-</sup> PI <sup>+</sup>	AnnV <sup>+</sup> PI <sup>+</sup>	AnnV <sup>-</sup> PI <sup>-</sup>	Annv <sup>+</sup> PI <sup>-</sup>
Vehicle 5	2.78 $\pm$ 0.4	0.03 $\pm$ 0.0	94.1 $\pm$ 5.4	3.1 $\pm$ 0.2
5, (80)	1.9 $\pm$ 0.7	0 $\pm$ 0.0	92.3 $\pm$ 6.3	5.9 $\pm$ 0.4
Vehicle 4	2.4 $\pm$ 1.1	0.1 $\pm$ 0.0	93.7 $\pm$ 4.3	3.7 $\pm$ 0.2
4, (50)	24.9 $\pm$ 2.4*	1.6 $\pm$ 0.2	66.7 $\pm$ 3.5*	6.7 $\pm$ 0.3
Vehicle 1	2 $\pm$ 0.1	0.06 $\pm$ 0.0	95 $\pm$ 6.2	2.8 $\pm$ 0.1
1, (120)	1.6 $\pm$ 0.2	0 $\pm$ 0.0	95.7 $\pm$ 6.5	2.6 $\pm$ 0.2
Vehicle 2	2.2 $\pm$ 0.2	0.2 $\pm$ 0.0	92.5 $\pm$ 5.8	5.1 $\pm$ 0.3
2, (140)	2.7 $\pm$ 0.1	0.05 $\pm$ 0.0	90.6 $\pm$ 7.1	6.6 $\pm$ 0.2



**Figure 2S.** Complexes inducing cell death. RPMI treated with the appropriate dose for each compound, were analysed for the percentage of PI<sup>+</sup> and Ann-V<sup>+</sup> cells by FACS analysis. Dot Plot are representative of one of three independent experiments. The values in the table represent the percentage of cells in each quadrant and are expressed as mean  $\pm$  SD. \* p < 0.05 vs vehicle treated cells.

**U266**

COMPOUND ( $\mu\text{M}$ )	AnnV <sup>-</sup> PI <sup>+</sup>	AnnV <sup>+</sup> PI <sup>+</sup>	AnnV <sup>-</sup> PI <sup>-</sup>	Annv <sup>+</sup> PI <sup>-</sup>
Vehicle 5	1.3 $\pm$ 0.1	0.4 $\pm$ 0.0	94.9 $\pm$ 3.4	3.36 $\pm$ 0.2
5, (120)	6.3 $\pm$ 0.3*	1.5 $\pm$ 0.1	70.1 $\pm$ 4.2*	22.05 $\pm$ 1.3*
Vehicle 4	1.7 $\pm$ 0.1	0.3 $\pm$ 0.0	96.3 $\pm$ 5.3	1.65 $\pm$ 0.3
4, (100)	17.7 $\pm$ 0.2*	9.6 $\pm$ 0.2	60.7 $\pm$ 4.2	11.9 $\pm$ 1.1
Vehicle 1	0.7 $\pm$ 0.0	0.15 $\pm$ 0.0	94.2 $\pm$ 5.3	4.9 $\pm$ 0.3
1, (180)	4.5 $\pm$ 0.3*	1.9 $\pm$ 0.1	78.3 $\pm$ 5.2*	15.3 $\pm$ 0.9*
Vehicle 2	1.3 $\pm$ 0.2	0.1 $\pm$ 0.0	96.4 $\pm$ 6.1	2.1 $\pm$ 0.1
2, (180)	1.3 $\pm$ 0.1	0.1 $\pm$ 0.0	95.4 $\pm$ 5.8	3.04 $\pm$ 0.1



**Figure 3S.** Complexes inducing cell death. U266 treated with the appropriate dose for each compound, were analysed for the percentage of PI<sup>+</sup> and Ann-V<sup>+</sup> cells by FACS analysis. Dot Plot are representative of one of three independent experiments. The values in the table represent the percentage of cells in each quadrant and are expressed as mean  $\pm$  SD. \* p < 0.05 vs vehicle treated cells.

**CELL CYCLE ANALYSIS**

**TABLE S2**

**RPMI**

<b>COMPLEX n°, (μM)</b>	<b>Sub-G0 % of cells</b>	<b>G0/G1 % of cells</b>	<b>S % of cells</b>	<b>G2/M % of cells</b>
<b>Vehicle 5</b>	4.6 ±1	34.5±2	37.2±2	23.8±2
<b>5, (80)</b>	26.9±2*	26.2±2	11.3±1	34.6±3
<b>Vehicle 4</b>	3.94±0	36.41±2	34.02±2	27.6±2
<b>4, (50)</b>	64.8±5*	36.4±1	0.0±0	0.0±0
<b>Vehicle 1</b>	5.51±0	29.2±2	20.6±2	44.6±3
<b>1, (120)</b>	9.4±6*	36.6±1*	22.0±0	30.0±0
<b>Vehicle 2</b>	6.0±0	24.2±2	28.6±1	40.1±4
<b>2, (140)</b>	22.6±1*	37.9±1*	24.1±1	34.7±4

**TABLE S3**

**U266**

<b>COMPLEX (μM)</b>	<b>Sub-G0 % of cells</b>	<b>G0/G1 % of cells</b>	<b>S % of cells</b>	<b>G2/M % of cells</b>
<b>Vehicle 5</b>	0	60.2±6	21.3±1	18.4±1
<b>5, (120)</b>	0	84.8±6*	3.6±0	11.4±1
<b>Vehicle 4</b>	0	59.2±4	15.9±1	24.9±2
<b>4, (100)</b>	0	76.1±4*	10.2±1	12.8±1
<b>Vehicle 1</b>	0	53.7±5	19.6±1	26.7±2
<b>1, (180)</b>	0	65.3±4*	9.9±1	24.7±3
<b>Vehicle 2</b>	0	54.7±4	23.3±1	21.9±2
<b>2, (180)</b>	0	68.2±5*	14.3±1	29.4±2

**Table S2, S3.** Ru complexes in regulating cell cycle. A, B) Cell cycle analysis in RPMI and U266 cell lines treated with the appropriate dose of Ru complexed (complex number in the table). Cell cycle was performed by PI incorporation assay and FACS analysis, after 48 h post-treatments. Histograms are representative of one of three independent experiments. The dose for each compound was described in the table. The values represent the percentage of cells in each phase and are expressed as mean ± SD. \* p < 0.05 vs vehicle treated cells.