Electronic Supplementary Information (ESI)

Zn(II), Cd(II) and Hg(II) saccharinate complexes with 2,6-bis(2-benzimidazolyl)pyridine as promising anticancer agents in breast and lung cancer cell lines via ROS-induced apoptosis

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Table S1 Crystallographic data and structure refinement for the metal complexes.

	Zn	**Cd**	**Hg**
empirical formula	$C_{52}H_{38}N_{12}O_8S_2Zn$	$C_{33}H_{21}CdN_7O_6S_2$	$C_{33}H_{21}HgN_7O_6S_2$
formula weight	1088.43	788.09	876.28
crystal system	triclinic	monoclinic	monoclinic
space group	$P\bar{1}$	*I*2/*a*	*I*2/*a*
a, Å	10.1938(5)	10.1600(7)	10.2014(6)
b, Å	12.5434(9)	14.1135(9)	14.2651(7)
c, Å	19.5581(14)	21.9573(14)	21.9691(12)
α, deg	88.570(6)	90	90
β, deg	82.144(5)	97.560(6)	97.393(6)
γ, deg	76.167(5)	90	90
V, Å^3	2405.4(3)	3121.2(4)	3170.5(3)
T, K	150(10)	293(2)	293(2)
Z	2	4	4
ρ_{calc} (g cm^{-3})	1.503	1.677	1.836
μ (mm^{-1})	0.669	0.893	5.045
F(000)	1120	1584	1712
θ (°)	3.042-25.681	2.946-25.678	3.148-25.676
collected refls	13899	5916	5075
data/ parameters	9069 /692	2968/227	3001/223
goodness-of-fit	1.011	1.074	1.031
R_1 [$I>2\sigma$]	0.0472	0.0349	0.0337
wR_2	0.1000	0.0702	0.0686

Table S2 Selected bond lengths (Å) and angles (°) for the metal complexes.

	Zn	**Cd**	**Hg**
M1−N1	2.208(2)	2.359(4)	2.420(5)
M1−N2	2.139(2)	2.390(2)	2.437(3)
M1−N3	2.146(2)	-	-
M1−N4	-	2.222(3)	2.215(4)
M1−N6	2.151(3)	-	-
M1−N7	2.136(2)	-	-
M1−N8	2.181(3)	-	-
N1−M1−N2	74.01(8)	69.16(6)	68.02(9)
N1−M1−N3	149.08(9)	-	-
N1−M1−N4	-	117.01(8)	118.69(11)
N2−M1−N4	-	104.39(9)	104.34(14)
N2−M1−N2^{i}	-	138.32(13)	136.04(18)
N2−M1−N4^{i}	-	94.28(9)	96.41(13)
N4−Cd1−N4^{i}	-	125.97(15)	122.6(2)
N1−M1−N6	96.67(9)	-	-
N1−M1−N7	93.66(8)	-	-
N1−M1−N8	89.12(9)	-	-
N2−M1−N3	75.16(8)	-	-
N2−M1−N6	114.40(9)	-	-
N2−M1−N7	164.78(9)	-	-
N2−M1−N8	95.91(9)	-	-
N3−Zn1−N6	93.85(9)	-	-
N3−Zn1−N7	117.16(8)	-	-
N3−Zn1−N8	96.31(9)	-	-
N6−Zn1−N7	75.24(9)	-	-
N6−Zn1−N8	149.59(9)	-	-
N7−Zn1−N8	74.61(9)	-	-

Symmetry code (*i*): −*x*+1/2, *y*, −*z*+1.

Table S3 Intermolecular interactions of the metal complexes with DNA obtained from molecular docking models.

	(D–H)	Hydrogen bonds (H···A)	(H···A, Å)	Binding free energy (kJ mol^{-1})
		PDB : 1BNA		
Zn	bzimpy-N5H	DA5:OP2	2.42	–34.73
	bzimpy-N10H	DT19:O4	2.51	
	DG4: OP2 *electrostatic*	π (bzimpy)	3.69	
Cd	DG4:H21	O2-sac (sulfonyl)	2.03	–37.66
	DC23:H4'	O1-sac (carbonyl)	2.37	
	sac (sulfonyl-S) *electrostatic*	DA6: OP1	3.81	
Hg	DG4:H21	O3-sac (sulfonyl)	1.97	–38.49
	DC23:H4'	O1-sac (carbonyl)	2.36	
	DA5:H5'2	O2-sac (sulfonyl)	2.53	
	sac (sulfonyl-S) *electrostatic*	DA6: OP1	3.84	
		PDB : 3CO3		
Zn	bzimpy-N10H	DG21:O6	2.12	–33.89
	bzimpy-N4H	DC2:OP2	2.61	
	DA20: OP2 *electrostatic*	π (bzimpy)	3.89	
Cd	DC11:H5'1	O2-sac (sulfonyl)	2.10	–37.24
	DA17:H4'	O2-sac (sulfonyl)	2.59	
	bzimpy-N3H	DT10:O2	2.65	
	bzimpy-N3H	DC12:O5'	2.88	
	DC12:OP1 *electrostatic*	π (bzimpy)	3.32	
	sac (sulfonyl-S) *electrostatic*	DC12: OP1	3.84	
	DA17:OP1 *electrostatic*	π (sac)	3.88	
Hg	DG16:H21	O3-sac (sulfonyl)	2.19	–38.07
	DC11:H5'1	O1-sac (carbonyl)	2.43	
	DC11:H5'2	O2-sac (sulfonyl)	2.64	
	DC12:OP1 *electrostatic*	π (sac)	3.64	
	sac (sulfonyl-S) *electrostatic*	DC12:OP1	3.96	
		PDB : 1LU5		
Zn	bzimpy-N10H	DG6:OP2	2.01	–37.66
	bzimpy-N4H	DA15:N7	2.50	
	DA15: OP2 *electrostatic*	π (bzimpy)	3.35	
	DT5: OP2 *electrostatic*	π (bzimpy)	3.66	
Cd	DG16:H21	O2-sac (sulfonyl)	2.28	–37.24
	DA17:H5'2	O3-sac (sulfonyl)	2.71	
	DA17:H5'2	O1-sac (carbonyl)	2.75	
Hg	DG16:H21	O3-sac (sulfonyl)	2.06	–38.49
	DC11:H4'1	O3-sac (sulfonyl)	2.58	
	DT10:H1'	O3-sac (sulfonyl)	2.80	
	DT10:H1'	O2-sac (sulfonyl)	2.84	
	DC11:OP1 *electrostatic*	π (sac)	3.92	

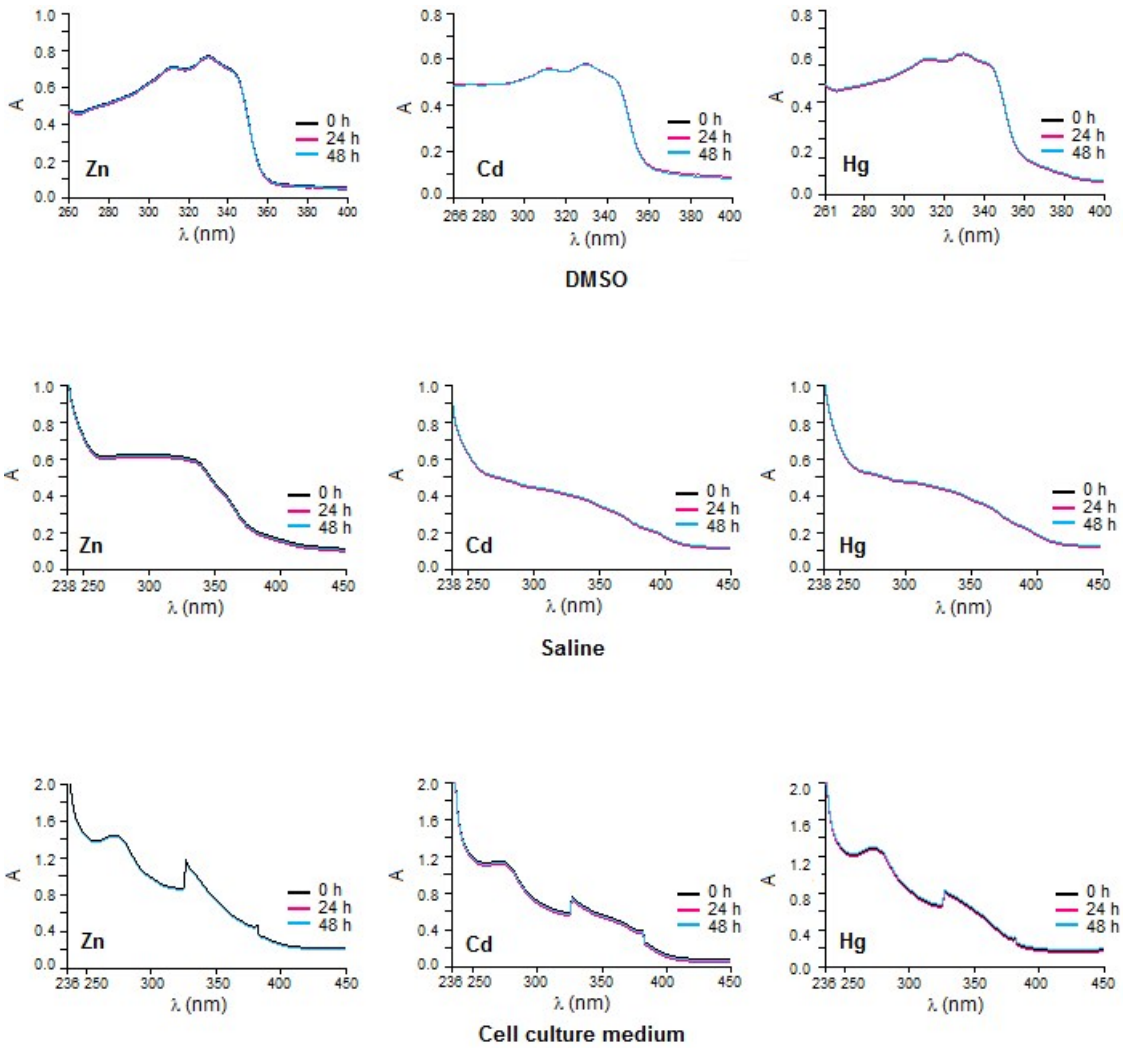

Fig. S1 UV-Vis spectra of the metal complexes (10 µM) showing their time-dependent stability in DMSO, saline and cell culture medium.

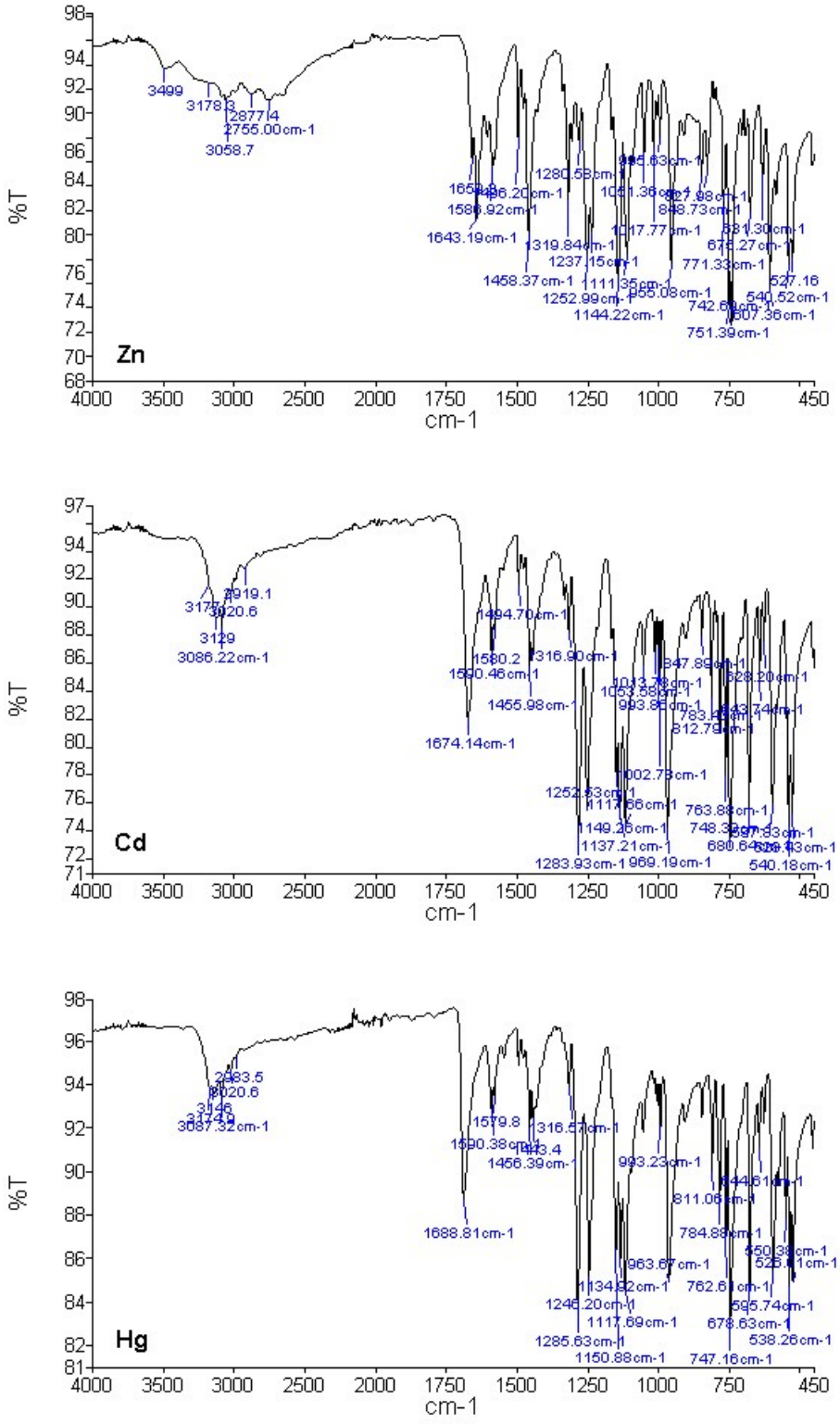

Fig. S2 IR spectra of the metal complexes.

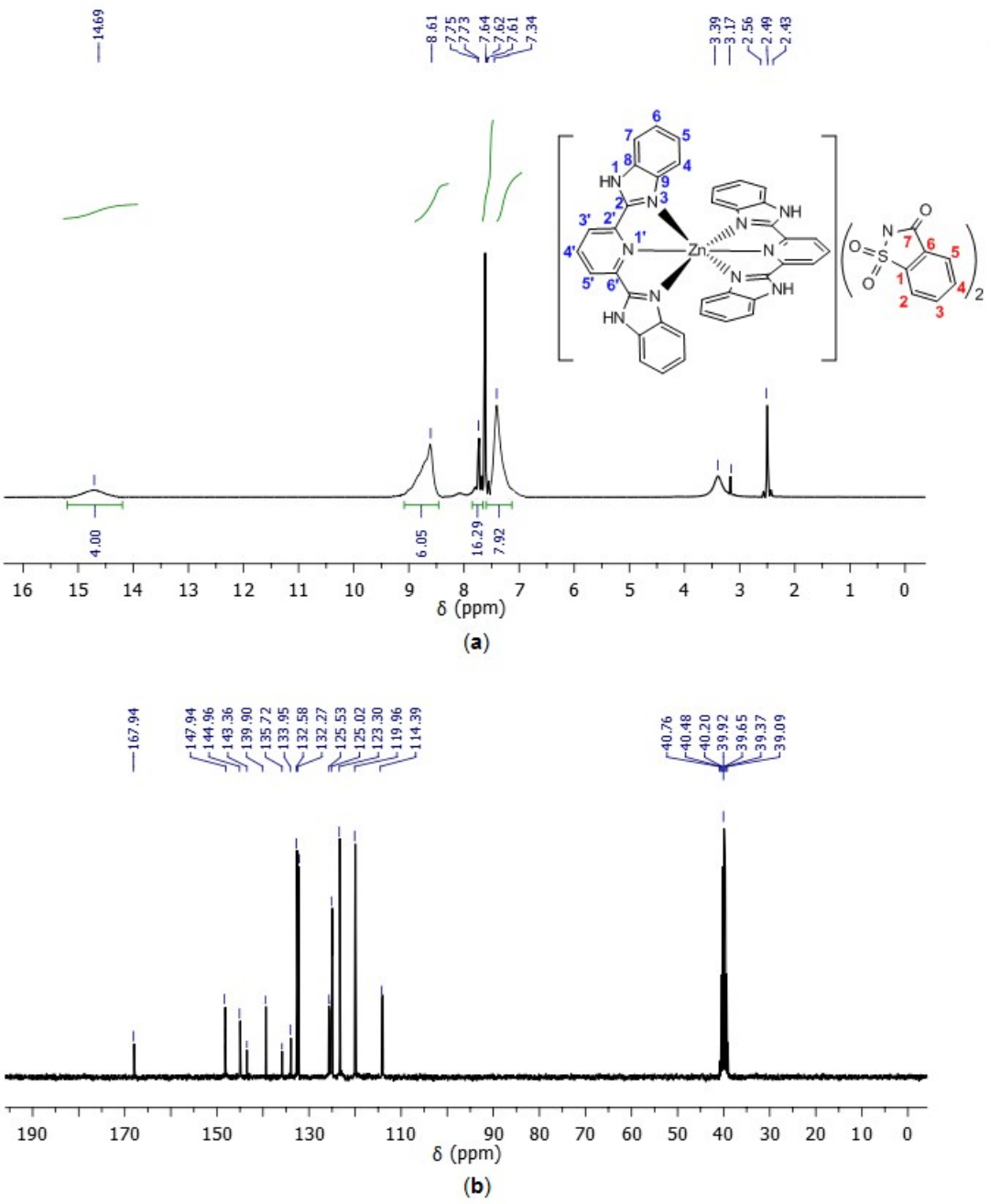

Fig. S3a ^{1}H- (**a**) and ^{13}C- (**b**) spectra of **Zn**.

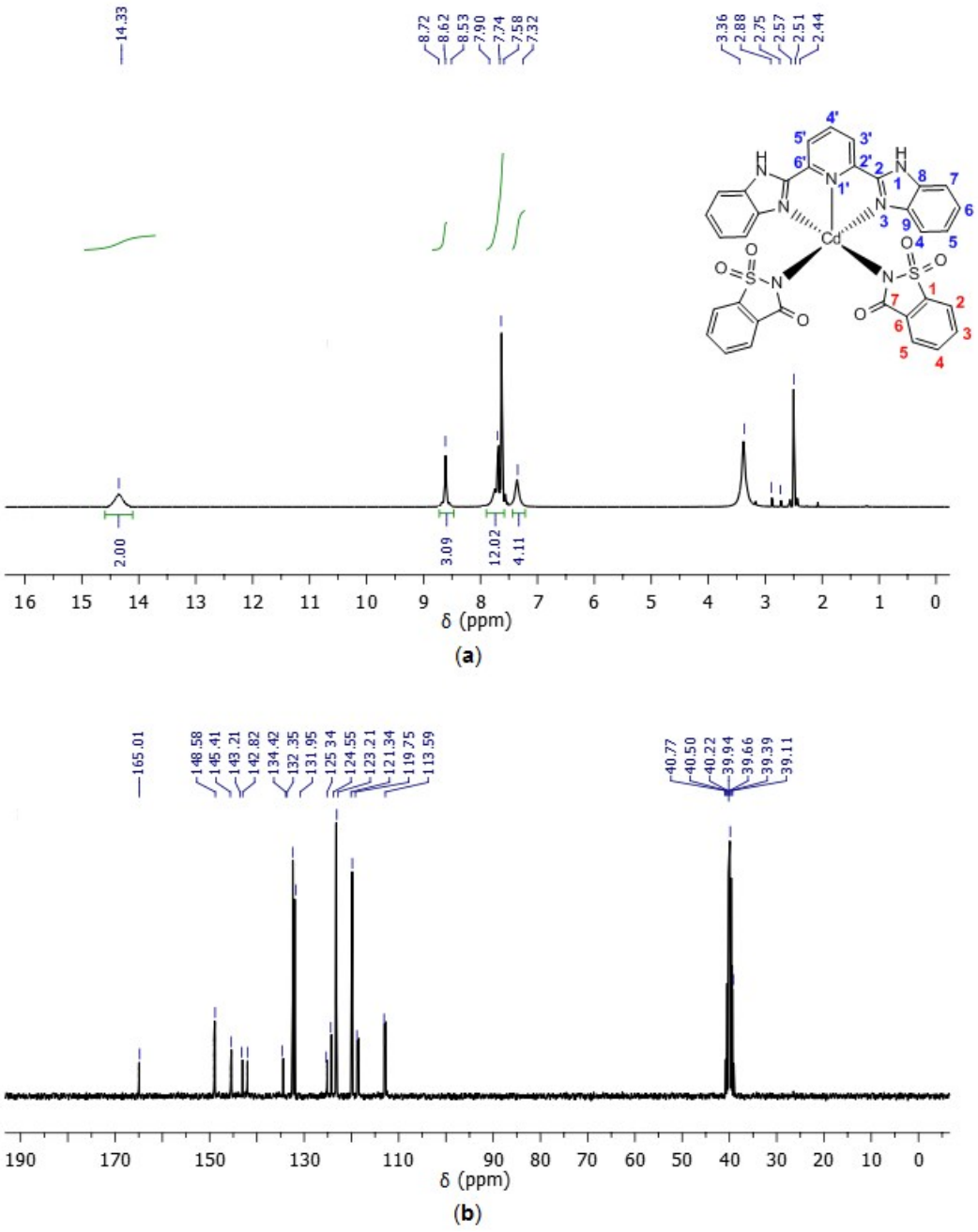

Fig. S3b ^{1}H- (**a**) and ^{13}C- (**b**) spectra of **Cd**.

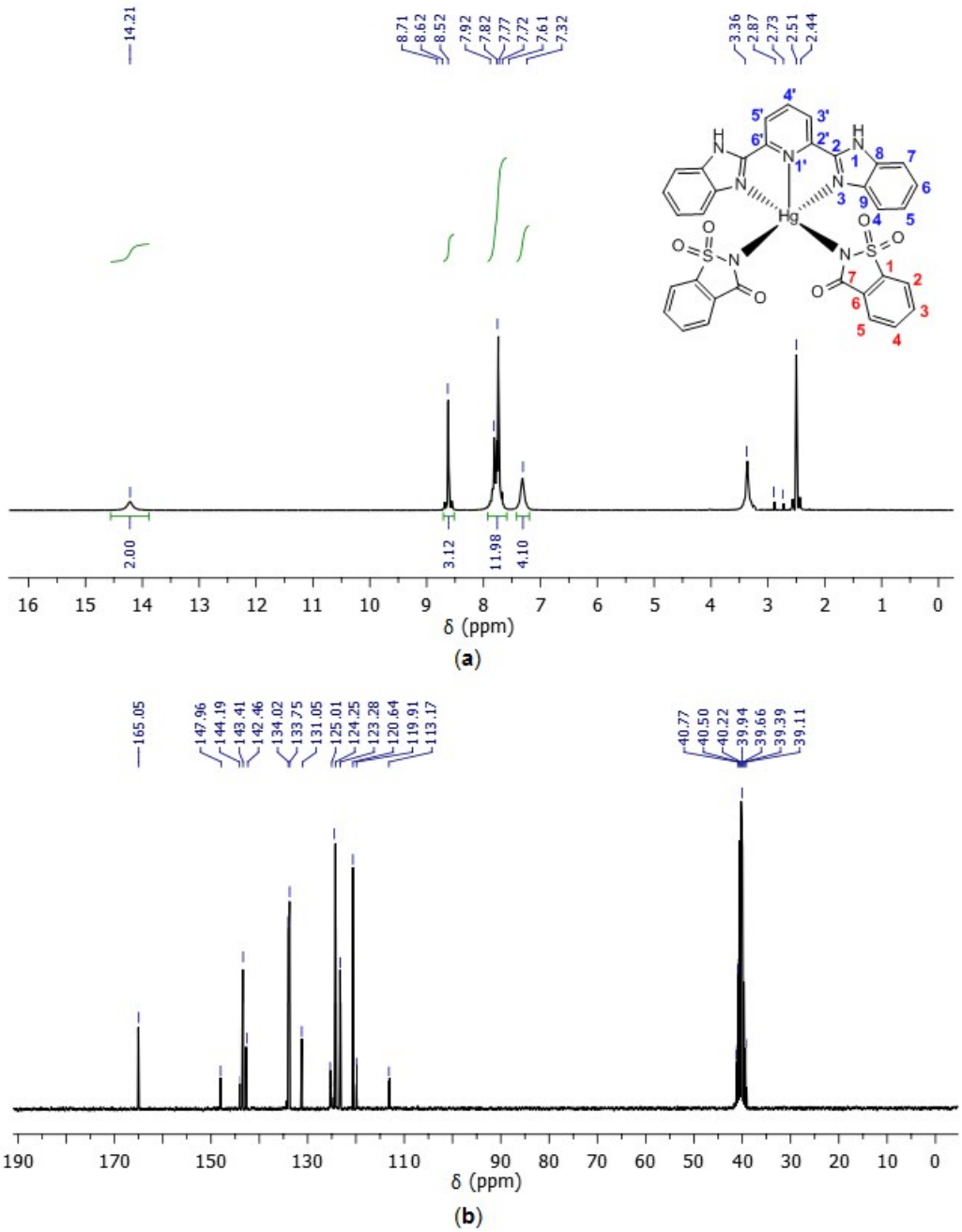

Fig. S3c ^{1}H- (**a**) and ^{13}C- (**b**) spectra of **Hg**.

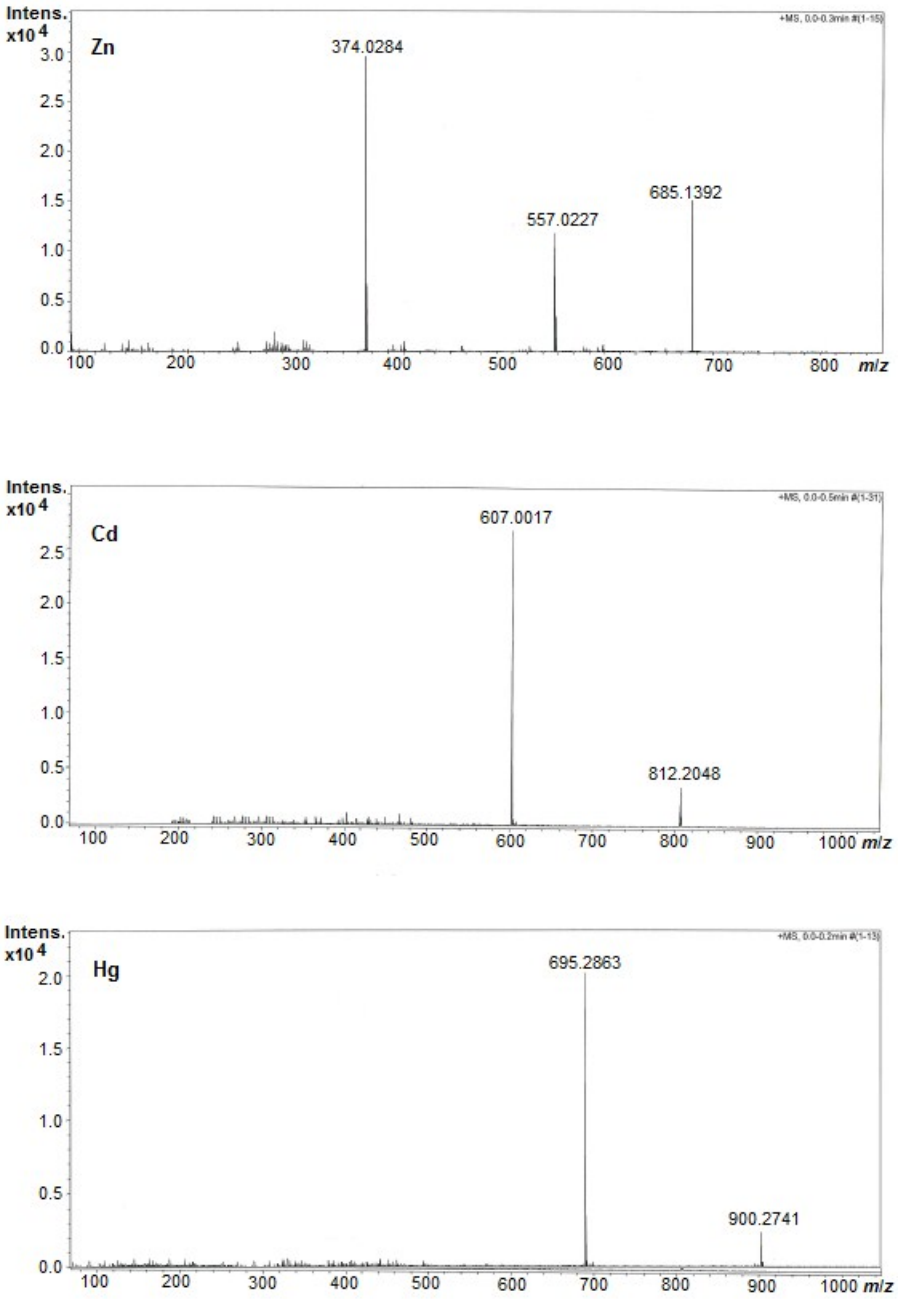

Fig. S4 ESI-MS spectra of the metal complexes.

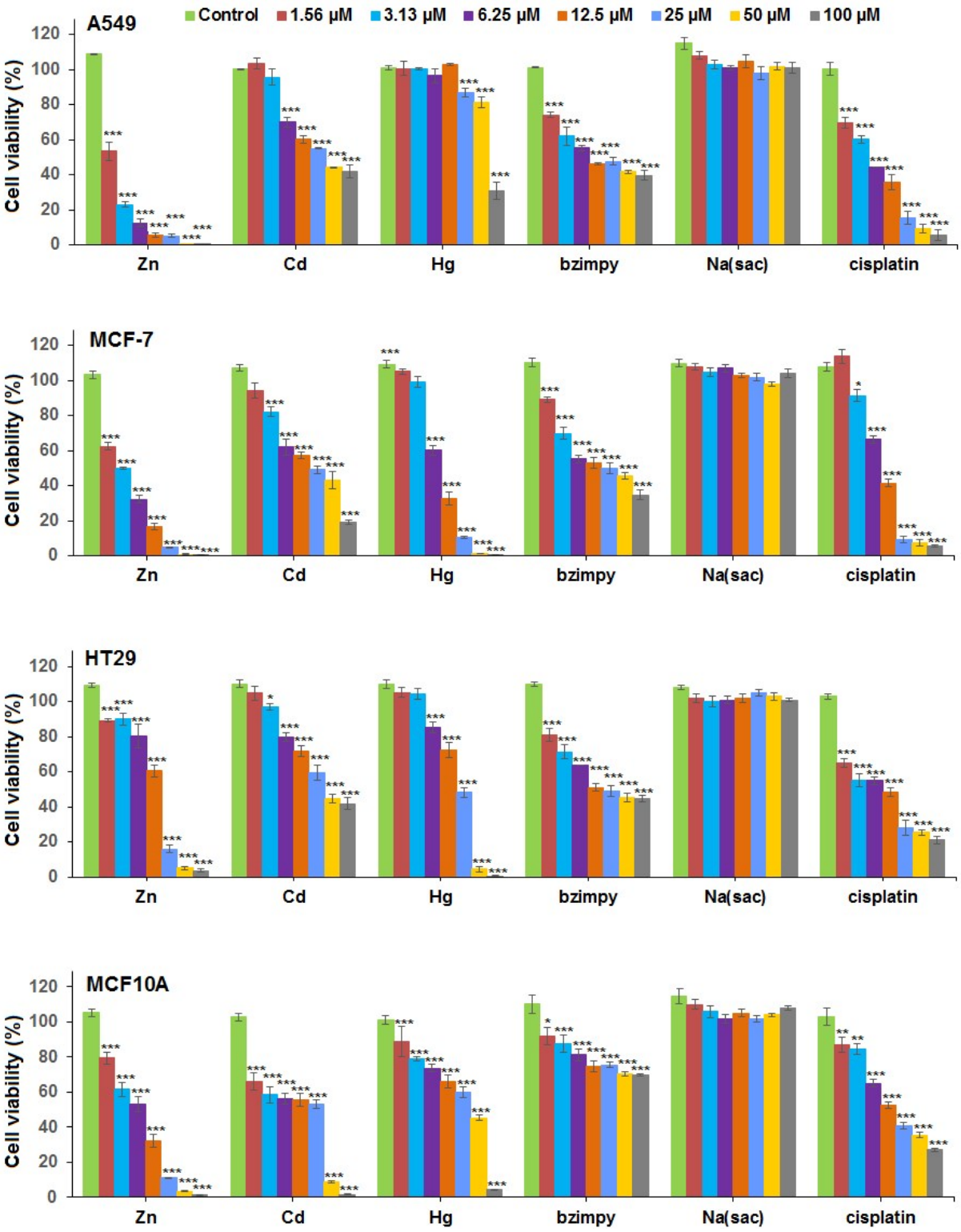

Fig. S5 The dose-response graphics for the metal complexes, bzimpy, Na(sac) and cisplatin obtained from ATP assay, showing the effect of the complexes on the growth of the cell lines after 48 h of treatment. Results are represented as mean ± standard deviation (n = 3). The asterisks denote statistically significant differences in comparison with the control group (*p < 0.05, **p < 0.01 and ***p < 0.001).

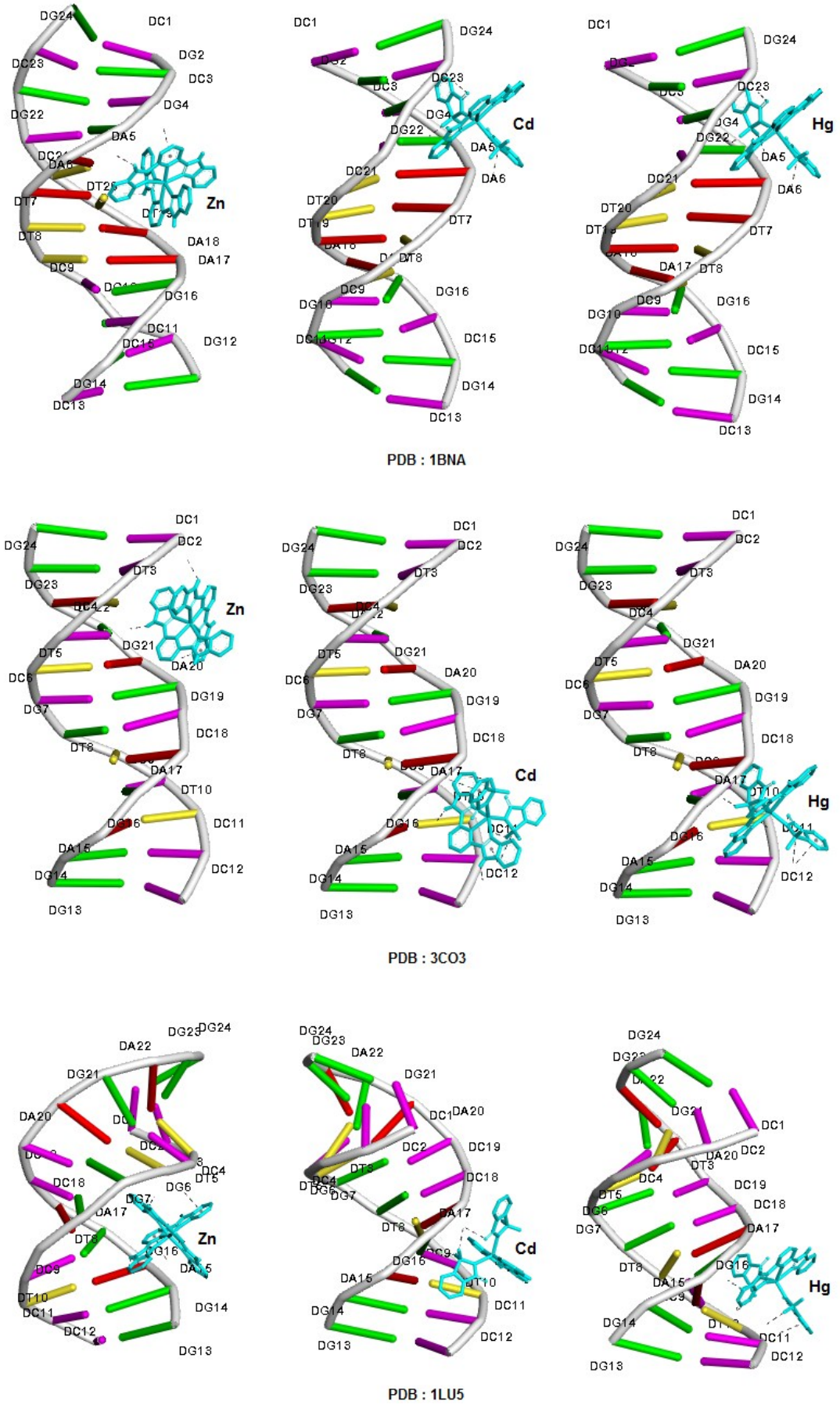

Fig. S6 DNA docking models of the metal complexes with 1BNA, 3CO3 and 1LU5.

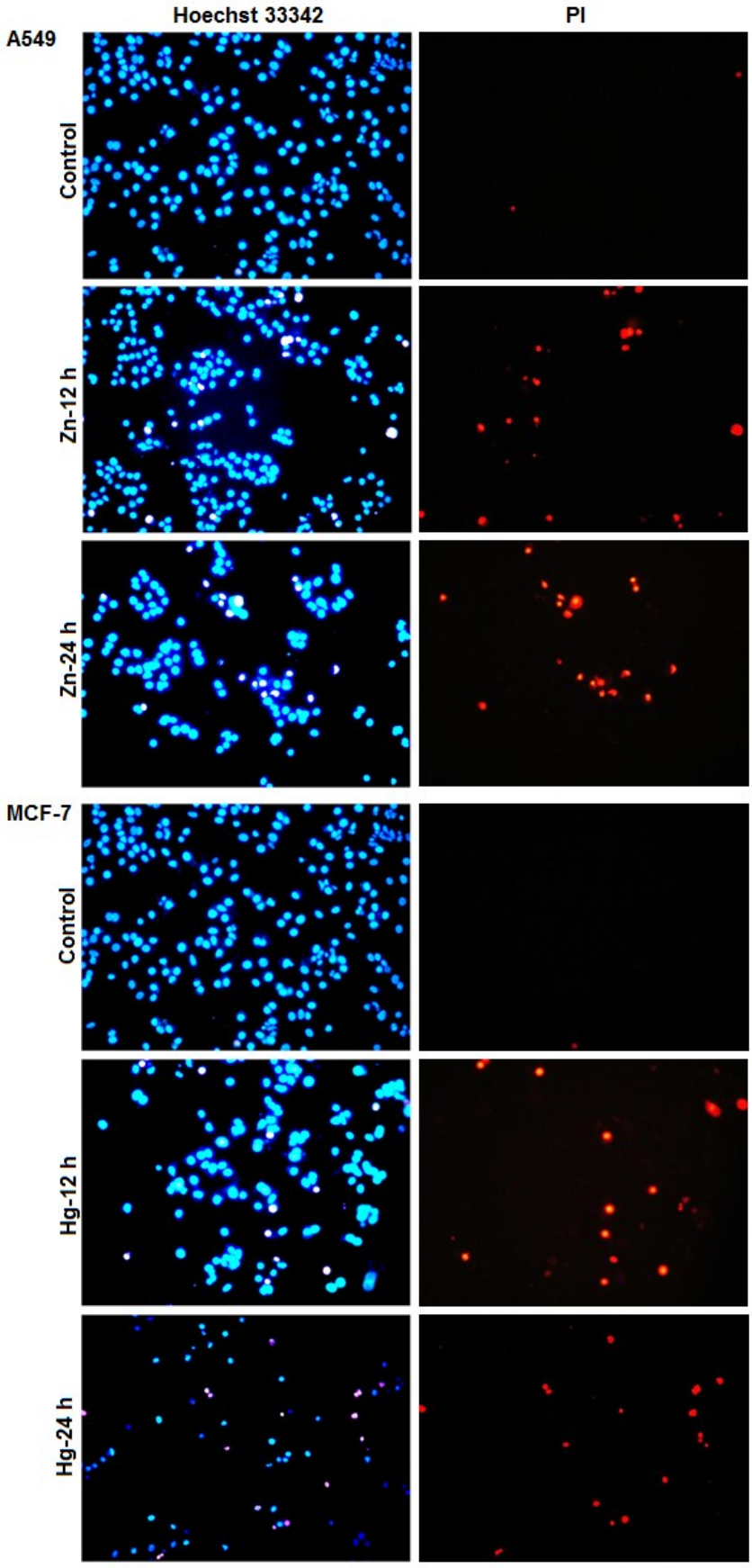

Fig. S7 Morphological changes in the nuclei of A549 and MCF-7 cells treated with IC_{90} doses of **Zn** and **Hg**, respectively, for 12 and 24 h. Cells were stained with Hoechst 33342/PI followed by detection using a fluorescence microscope. Magnification: 20×.

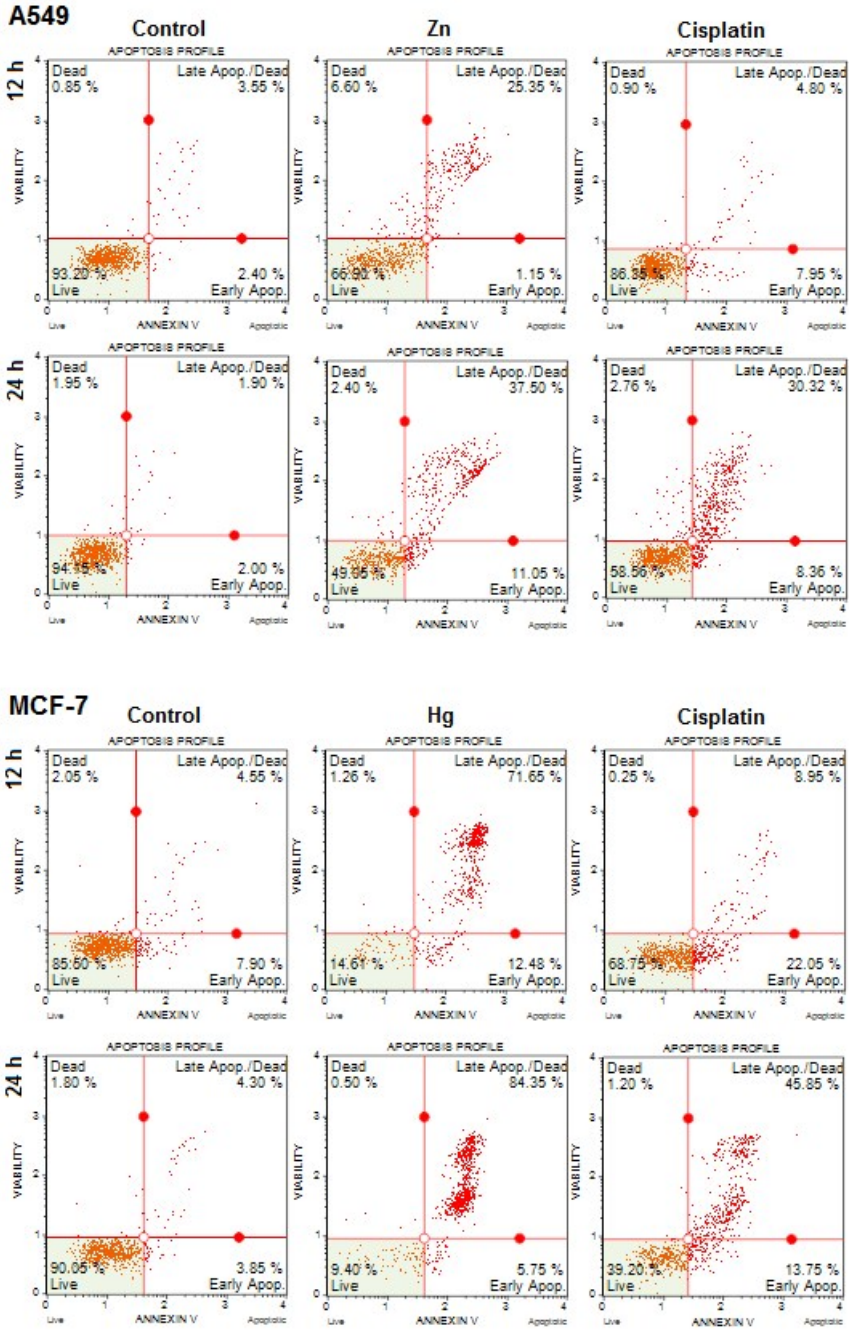

Fig. S8 Annexin-V/7-AAD staining assay. A549 and MCF-7 cells treated with IC_{90} doses of **Zn** and **Hg**, respectively, for 12 and 24 h. Cisplatin was used as a positive control.

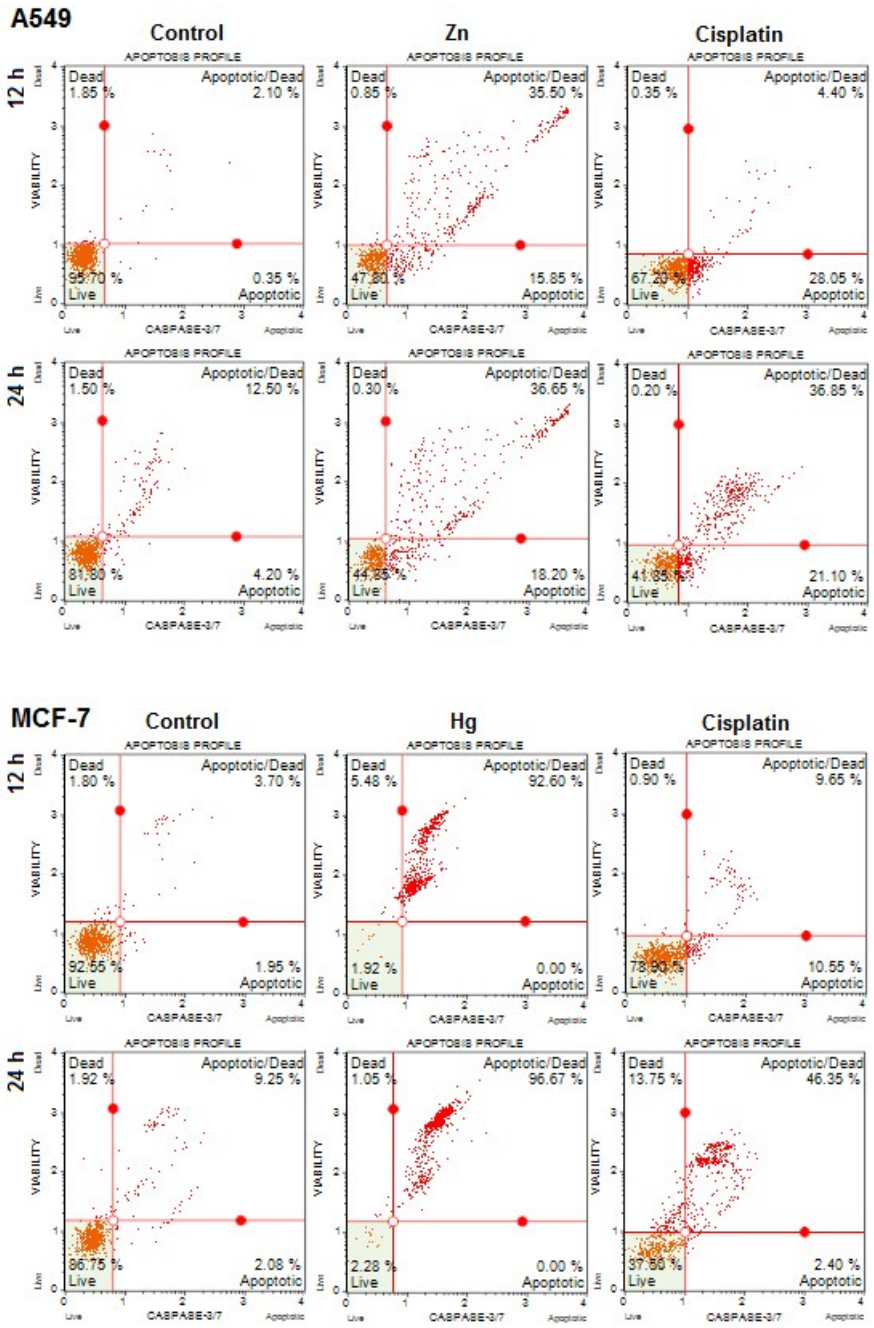

Fig. S9 Caspase 3/7 activity in A549 and MCF-7 cells treated with IC_{90} doses of **Zn** and **Hg**, respectively, for 12 and 24 h. Cisplatin was used as a positive control.

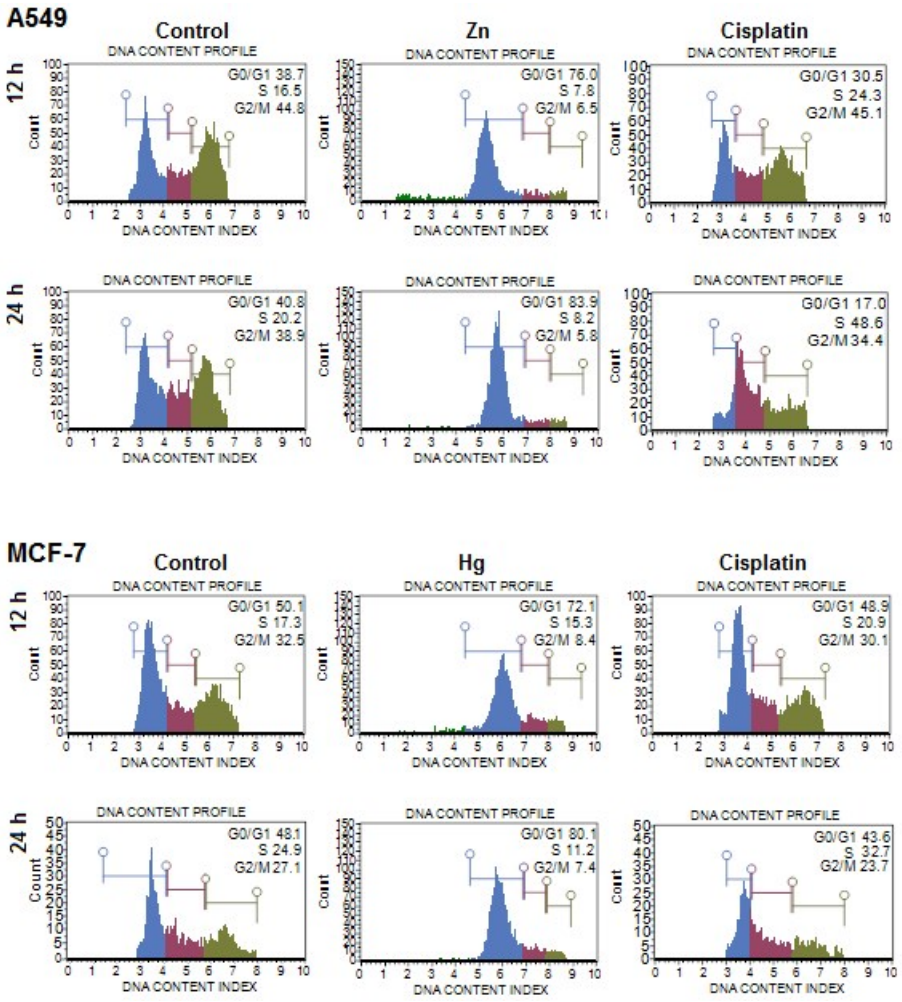

Fig. S10 Cell cycle progression of A549 and MCF-7 cells treated with IC_{90} doses of **Zn** and **Hg**, respectively, for 12 and 24 h. Cisplatin was used as a positive control.

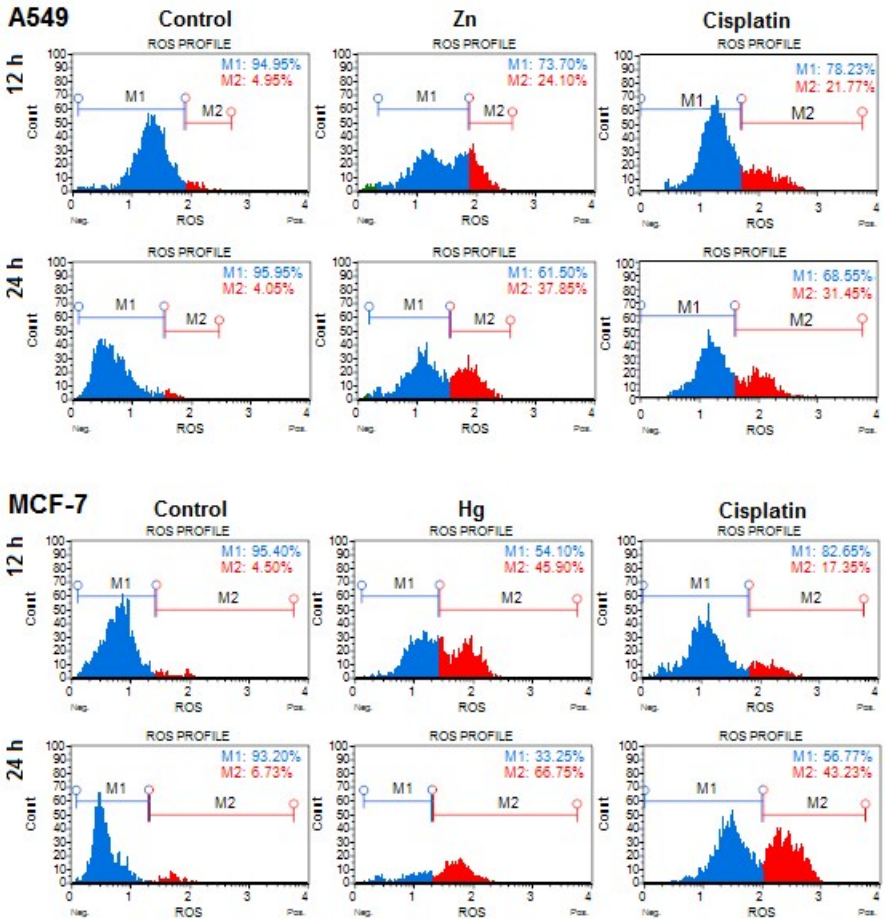

Fig. S11 ROS generation in A549 and MCF-7 cells treated with IC_{90} doses of **Zn** and **Hg**, respectively, for 12 and 24 h. Cisplatin was used as a positive control.

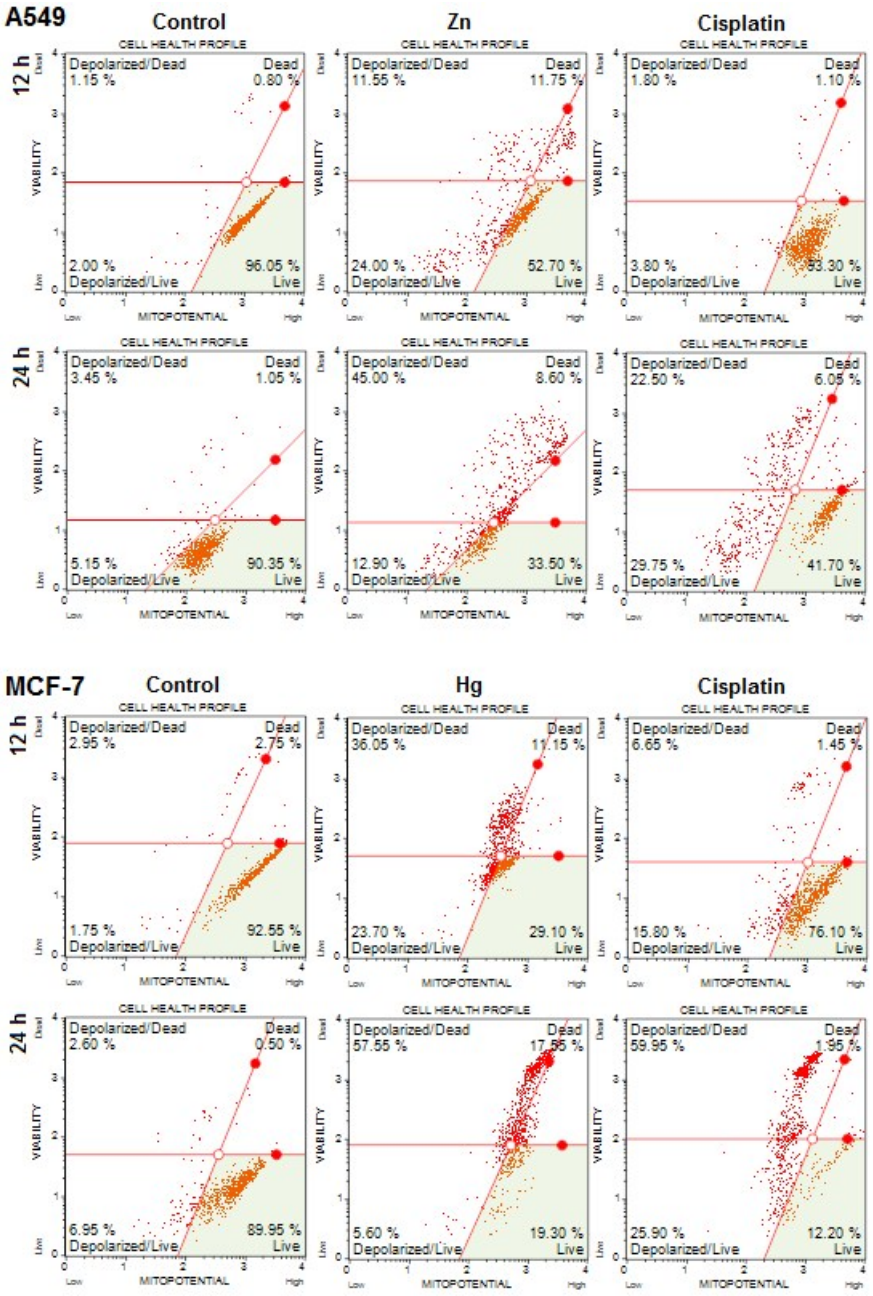

Fig. S12 Mitochondrial membrane depolarization in A549 and MCF-7 cells treated with IC_{90} doses of **Zn** and **Hg**, respectively, for 12 and 24 h. Cisplatin was used as a positive control.

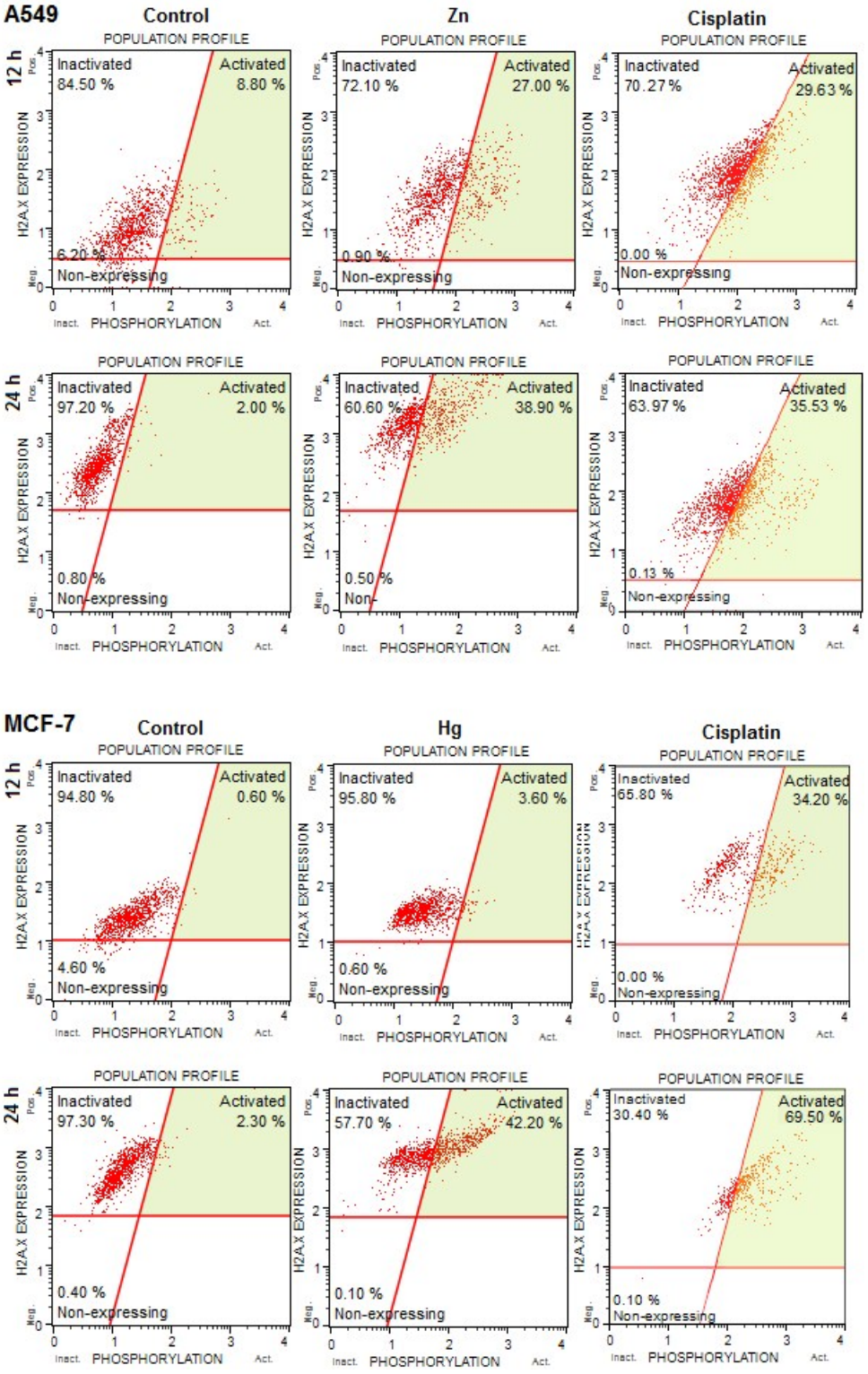

Fig. S13 Formation of DNA double-strand breaks in A549 and MCF-7 cells treated with IC_{90} doses of **Zn** and **Hg**, respectively, for 12 and 24 h. Cisplatin was used as a positive control.