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SUPPLEMENTARY MATERIAL

Homo- and heterometallic complexes of Zn(II), {Zn(II)Au(I)}, and {Zn(II)Ag(I)} with pentadentate Schiff base ligands as promising anticancer agents

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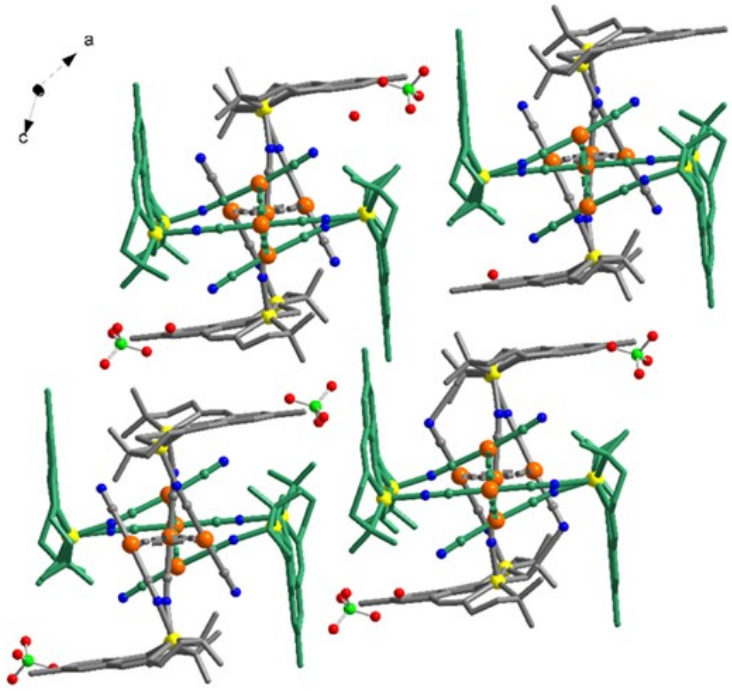

Figure S1. Detail of the crystal packing diagram in compound **ZndmenAu** (the two crystallographic independent heptanuclear units are colored in grey and green).

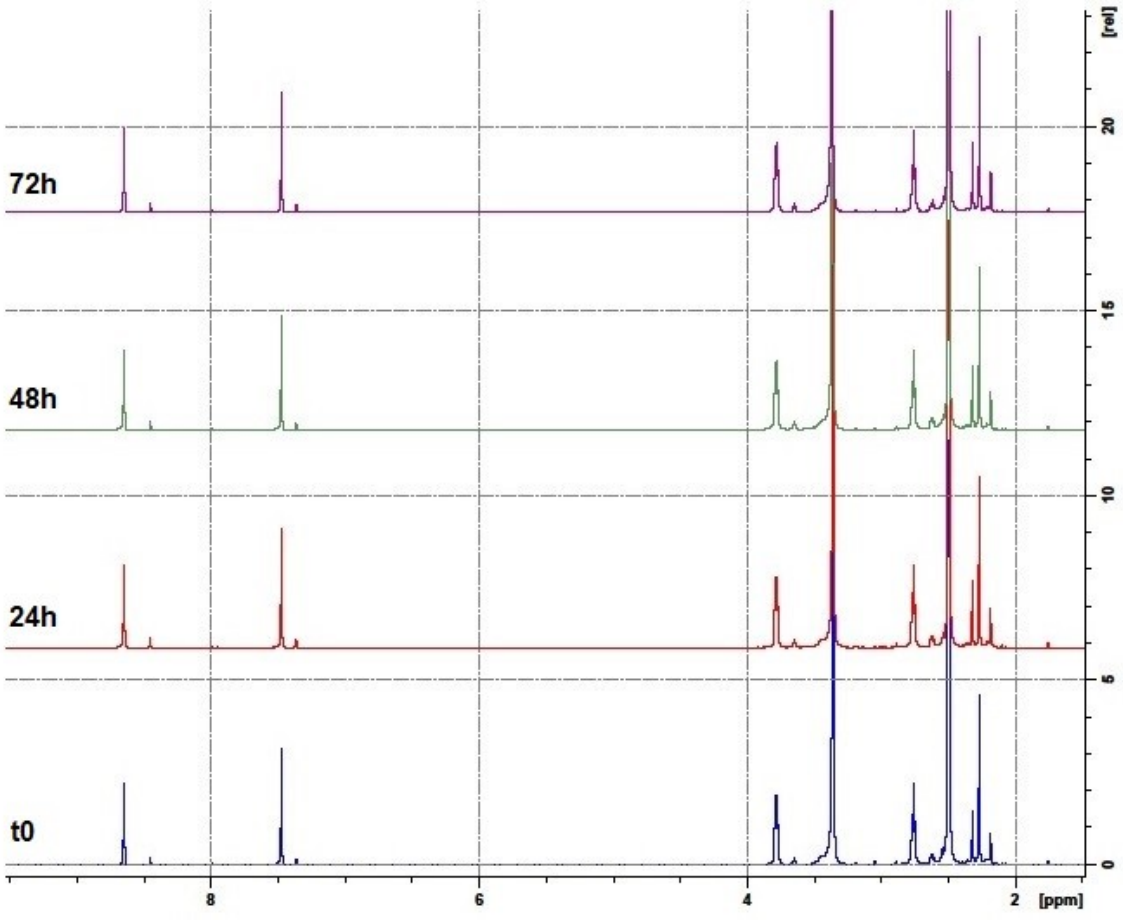

Figure S2. ^{1}H NMR (DMSO-d6) spectra of **ZndmenAu** at t0, 24, 48, and 72h.

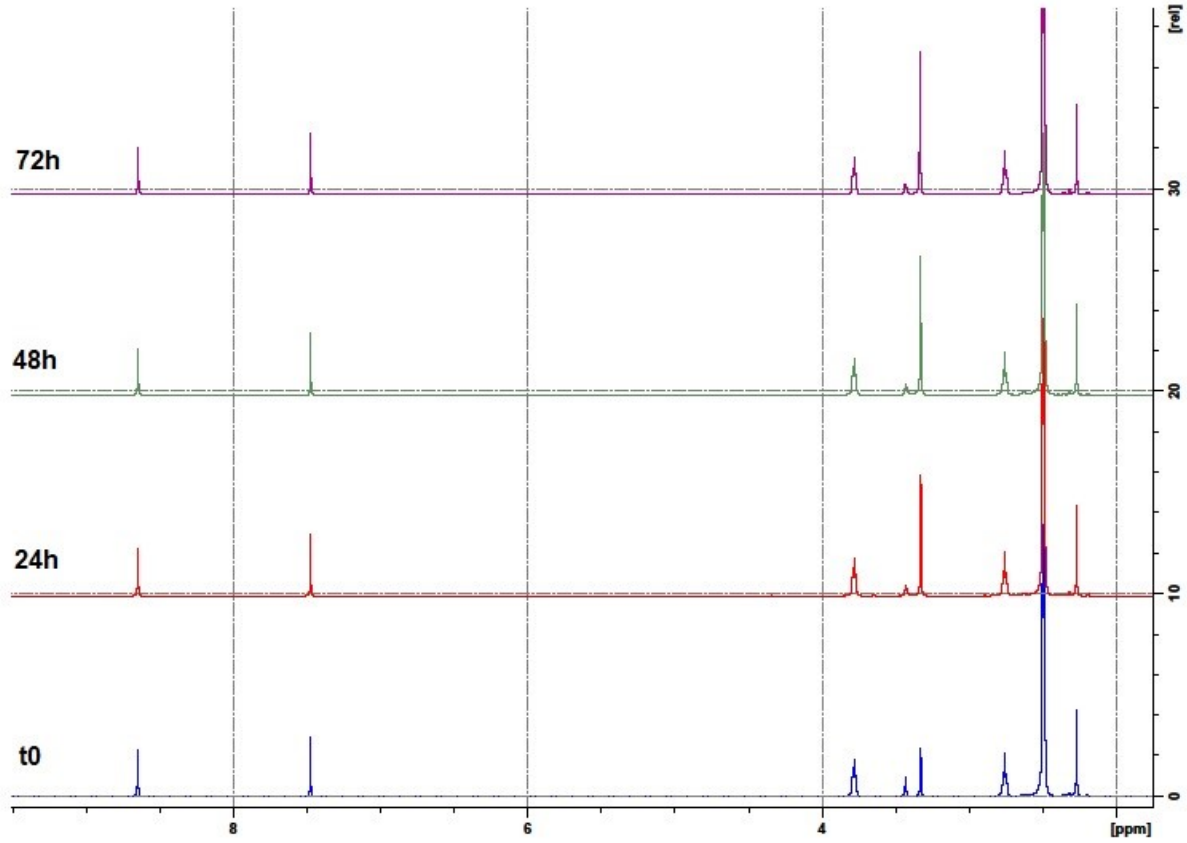

Figure S3. ^{1}H NMR (DMSO-d6) spectra of **ZndmenAg** at t0, 24, 48, and 72h.

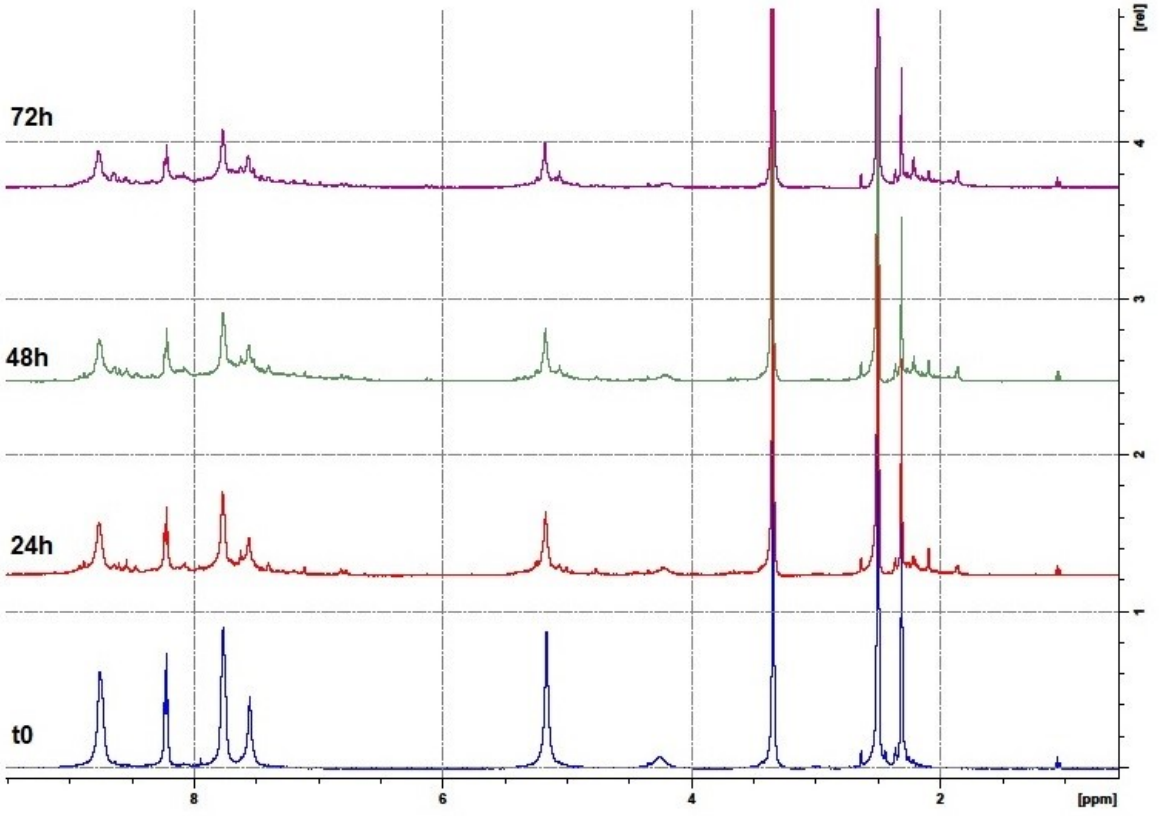

Figure S4. ^{1}H NMR (DMSO-d6) spectra of **ZnampyAu** at t0, 24, 48, and 72h.

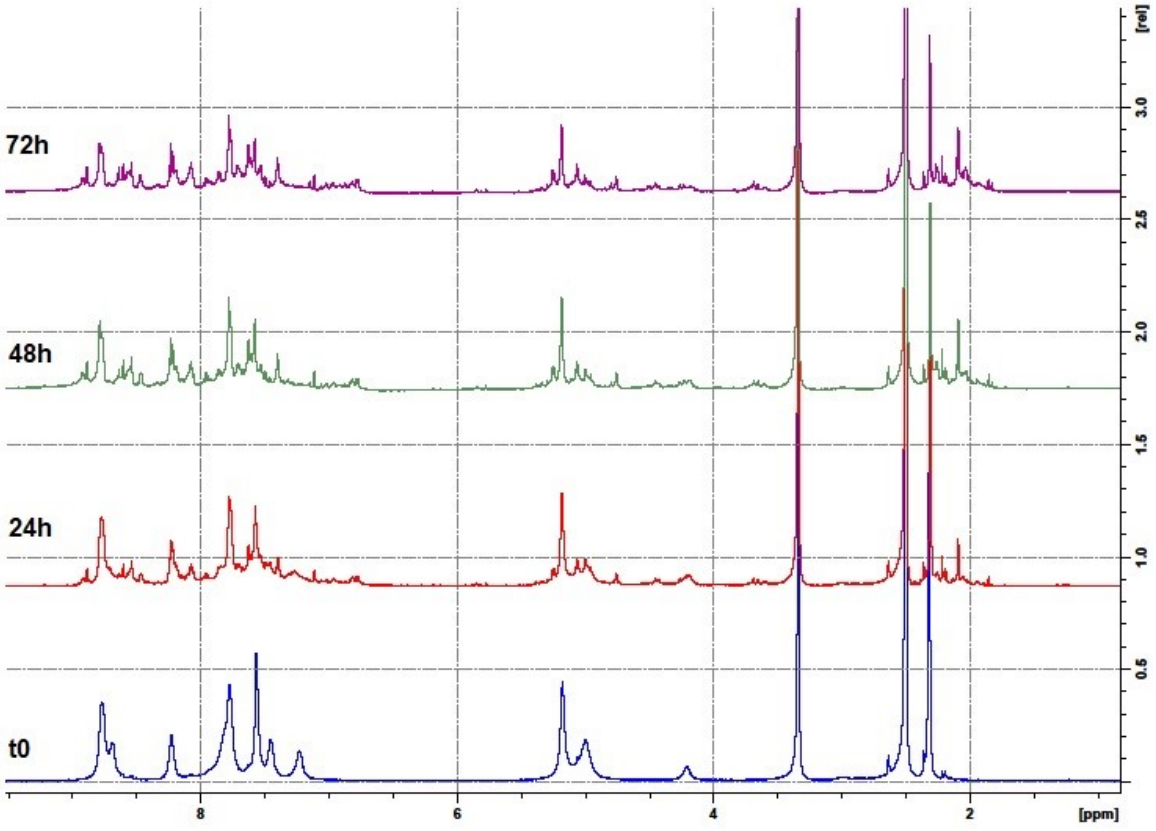

Figure S5. ^{1}H NMR (DMSO-d6) spectra of **Znampy** at t0, 24, 48, and 72h.

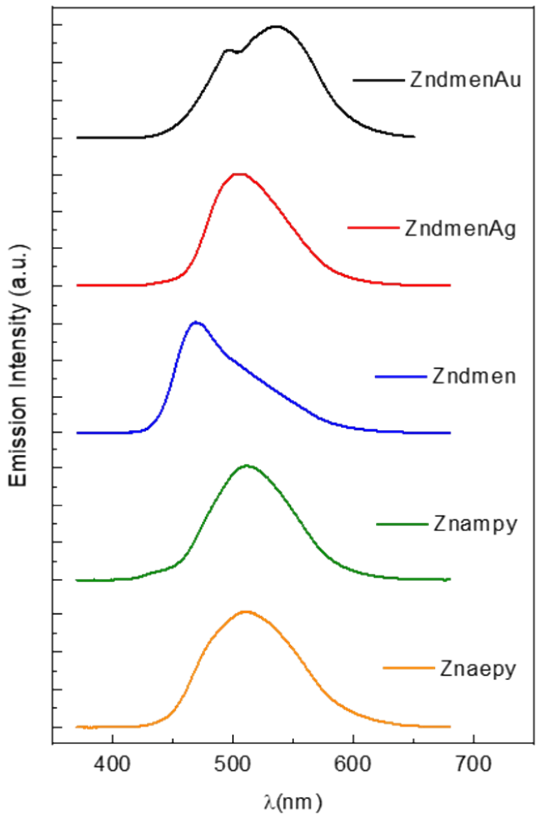

Figure S6. Solid-state emission spectra of **Zndmen**, **Znampy**, **Znaepy**, **ZndmenAg**, and **ZndmenAu** at room temperature (λ_{exc} = 350 nm).

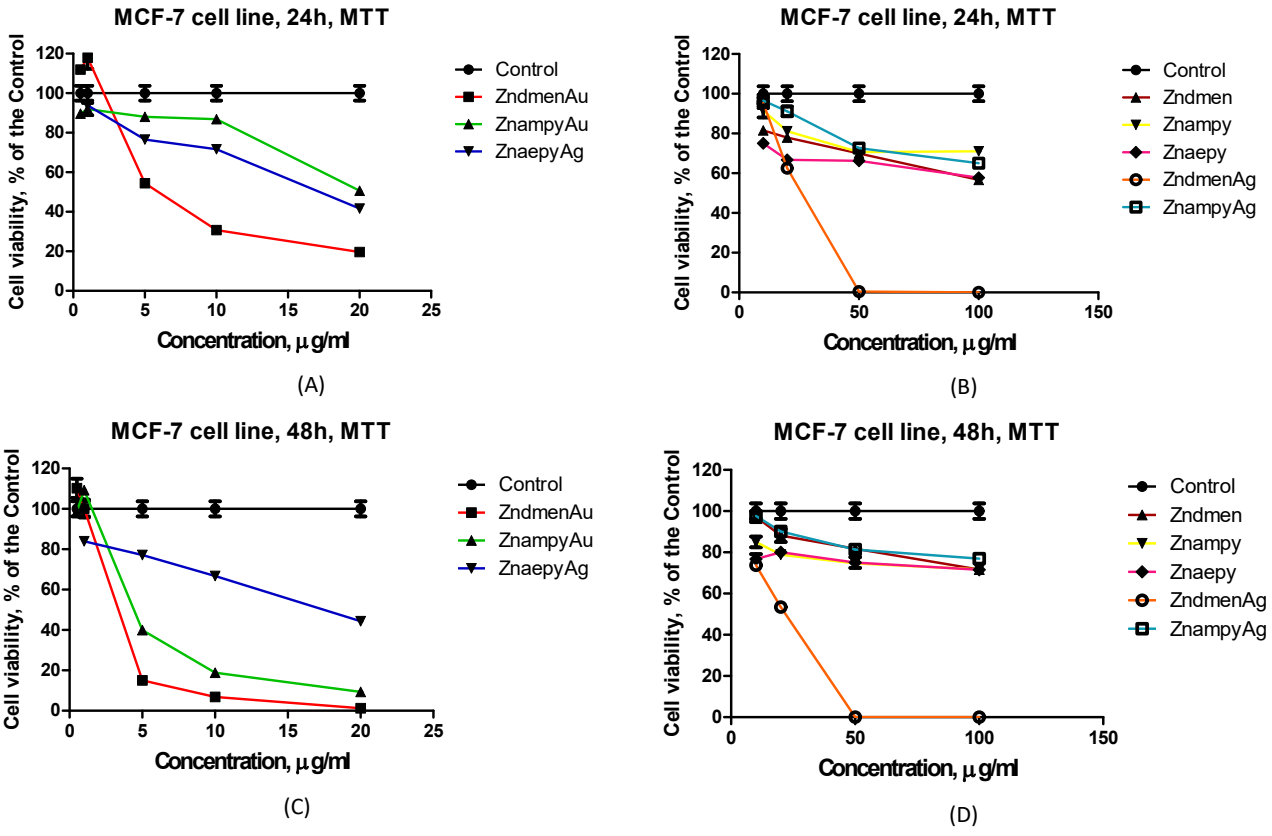

Figure S7. Concentration-response curves of investigated complexes for luminal A breast cancer MCF-7 cells evaluated by MTT test after 24 h (A, B) and 48 h (C, D) treatment period.

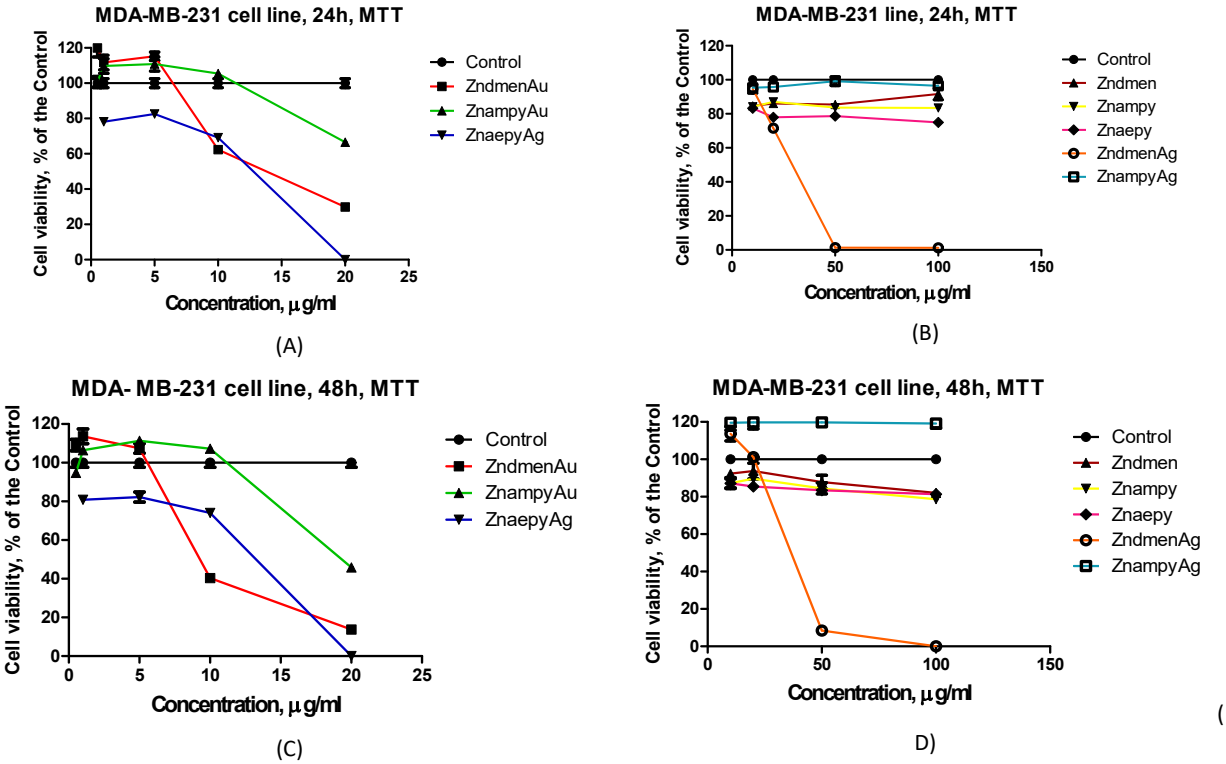

Figure S8. Concentration-response curves of investigated complexes for triple negative breast cancer MDA-MB-231 cells evaluated by MTT test after 24 h (A, B) and 48 h (C, D) treatment period.

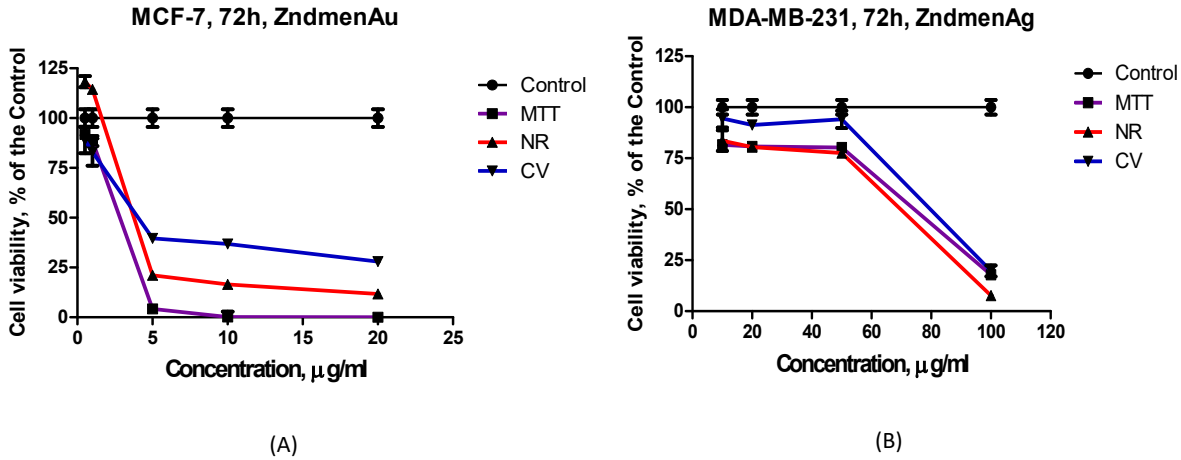

Figure S9. Comparison between three different cytotoxicity assays (MTT, NR, and CV) after 72h of treatment with (0.1-20 µg/ml) **ZndmenAu** on MCF-7 (A); **ZndmenAg** (10 - 100 µg/ml) on MDA-MB-231 cells (B): representative curves.

Table S1. Selected geometric parameters: bonds (Å) and angles (°) in compounds **ZndmenAg** and **ZndmenAu**.

ZndmenAg		**ZndmenAu**	
Bonds length (Å)			
Zn1-N1 = 2.108(3)	Zn2-O1 = 2.103(2)	Zn1-O1 = 2.090(3)	Zn4-O4 = 1.965(4)
Zn1-N2 = 2.122(3)	Zn2-O2 = 1.944(3)	Zn1-O2 = 1.945(4)	Zn4-O3 = 2.092(4)
Zn1-N3 = 2.017(3)	Ag1-C18 = 2.047(4)	Zn1-N5 = 2.049(6)	Zn4-N14 = 2.025(5)
Zn1-O1 = 2.024(3)	Ag1-C18’ = 2.047(4)	Zn1-N1 = 2.068(4)	Zn4-N11 = 2.178(4)
Zn1-O2 = 1.997(2)	Ag2-C19 = 2.058(4)	Zn1-N2 = 2.154(4)	Zn4-N10 = 2.059(5)
Zn2-N4 = 2.079(3)	Ag2-C19” = 2.058(4)	Zn2-O1 = 2.059(3)	Au1-C18 = 1.983(7)
Zn2-N5 = 2.169(3)	N3-C18 = 1.125(5)	Zn2-O2 = 1.973(4)	Au1-C19 = 1.984(8)
Zn2-N6 = 2.023(3)	N6-C19 = 1.120(5)	Zn2-N7 = 2.032(5)	Au2-C20’ = 1.985(6)
		Zn2-N4 = 2.124(5)	Au2-C20 = 1.985(6)
		Zn2-N3 = 2.074(5)	Au3-C62 = 1.990(7)
		Zn3-O4 = 1.952(4)	Au3-C73 = 1.985(7)
		Zn3-O3 = 2.088(4)	Au4-C64 =1.996(6)
		Zn3-N12 = 2.023(5)	Au4-C64’ = 1.996(6)
		Zn3-N8 = 2.067(6)	Au2-Au1’ = 3.2496(9)
		Zn3-N9 = 2.143(5)	Au2-Au1 = 3.2496(9)
			Au4-Au3’ = 3.2813(5
			Au4-Au3 = 3.2814(6)
Angles (°)			
O2-Zn1-O1 = 80.7(1)	O1-Zn2-N5 = 160.0(2)	O2-Zn1-O1 = 78.7(2)	O4-Zn3-O3 = 77.7(2)
O2-Zn1-N1 = 151.1(2)	N4-Zn2-O1 = 82.1(1)	O2-Zn1-N1 = 146.1(2)	O4-Zn3-N8 = 145.6(2)
O2-Zn1-N2 = 100.8(2)	N4-Zn2-N5 = 81.3(2)	O2-Zn1-N2 = 104.8(3)	O4-Zn3-N9 = 104.3(2)
O2-Zn1-N3 = 104.2(2)	N6-Zn2-O1 = 95.5(2)	O2-Zn1-N5 = 109.2(3)	O4-Zn3-N12 = 114.2(2)
O1-Zn1-N1 = 83.6(2)	N6-Zn2-N4 = 114.8(2)	O1-Zn1-N1 = 84.1(2)	O3-Zn3-N8 = 83.7(2)
O1-Zn1-N2 = 148.6(1)	N6-Zn2-N5 = 101.4(1)	O1-Zn1-N2 = 159.0(2)	O3-Zn3-N9 = 156.2(2)
N1-Zn1-N2 = 80.4(2)	Zn2-O2-Zn1 = 102.1(1)	N1-Zn1-N2 = 81.8(2)	N8-Zn3-N9 = 82.0(3)
N3-Zn1-O1 = 107.1(2)	Zn1-O1-Zn2 = 95.9(1)	N5-Zn1-O1 = 95.7(2)	N12-Zn3-O3 = 99.3(3)
N3-Zn1-N1 = 103.4(2)	N3-C18-Ag1 = 173.6(4)	N5-Zn1-N1 = 101.4(2)	N12-Zn3-N8 = 97.0(2)
N3-Zn1-N2 = 102.7(3)	N6-C19-Ag2 = 178.2(5)	N5-Zn1-N2 = 102.3(2)	N12-Zn3-N9 = 101.1(2)
O2-Zn2-O1 = 80.0(2)	C19-N6-Zn2 = 165.4(4)	O2-Zn2-O1 = 78.9(2)	O3-Zn4-O4 = 77.3(2)
O2-Zn2-N4 = 141.8(2)	C18-N3-Zn1 = 161.4(4)	O2-Zn2-N3 = 153.5(2)	O3-Zn4-N10 = 83.6(3)
O2-Zn2-N5 = 106.7(1)	C18’-Ag1-C18 = 180.0	O2-Zn2-N4 = 103.5(2)	O3-Zn4-N11 = 150.2(2)
O2-Zn2-N6 = 100.3(1)	C19”-Ag2-C19 =180.0	O2-Zn2-N7 = 104.8(2)	O3-Zn4-N14 = 105.8(2)
		O1-Zn2-N4 = 147.4(2)	O4-Zn4-N14 = 105.6(2)
		N3-Zn2-O1 = 83.7(2)	N11-Zn4-O3 = 150.2(2)
		N3-Zn2-N4 = 80.9(2)	N11-Zn4-N10 = 81.7(2)
		N7-Zn2-O1 = 111.8(3)	N14-Zn4-O3 = 105.8(2)
		N7-Zn2-N3 = 100.1(2)	N14-Zn4-N10 = 102.6(2)
		N7-Zn2-N4 = 98.9(3)	N14-Zn4-N11 = 102.6(2)
		Zn2-O2-Zn1 = 100.6(2)	Zn3-O4-Zn4 = 100.9(3)
		Zn1-O1-Zn2 = 93.1(2)	Zn3-O3-Zn4 = 92.6(2)
		N5-C18-Au1 = 179.2(6)	N12-C37-Au3 = 177.9(5)
		N6-C19-Au1 = 178.9(8)	N13-C38-Au3 = 177.3(7)
		N7-C20-Au2 = 175.0(5)	N14-C39-Au4 = 178.3(5)
		C20-N7-Zn2 = 165.9(5)	C39-N14-Zn4 = 171.7(4)
		C18-N5-Zn1 = 165.4(4)	C37-N12-Zn3 = 168.1(5)
		C18-Au1-C19 = 177.4(2)	C37-Au3-C38 = 175.7(3)
		C20’-Au2-C20 =177.0(3)	C39’-Au4-C39 =178.5(3)
‘= -x,1-y,1-z; “= 1-x,1-y,-z		*’ = 1-x, y, 1/2-z*	

Table S2. Cytotoxic activity (CC_{50} and CC_{90}, μM) of the investigated complexes on cultured MCF-7 cells.

Cell line	MCF-7			
	MTT		NR	CV
	24 h	48 h	72 h	72 h
Znampy	n.d.	n.d.	(-)	n.d.
ZnampyAg	n.d.	n.d.	n.d.	n.d.
ZnampyAu	30.5 ± 0.2 (n.d.)	2.7 ± 0.9 (11.5 ± 0.7)	3.6 ± 2.1 (6.1 ± 1.8)	5.3 ± 1.7
Zndmen	n.d.	n.d.	(-)	n.d.
ZndmenAg	36.8 ± 3.5 (63.8 ± 5.9)	30.7 ± 4.8 (62.3 ± 4.9)	n.d.	123.7 ± 5.6
ZndmenAu	3.4 ± 1.5 (n.d.)	2.2 ± 1.8	2.1 ± 2.1 (n.d.)	2.3 ± 2.0
Znaepy	n.d.	n.d.	(-)	n.d.
ZnaepyAg	20.1 ± 4.4	20.4 ± 3.1	(-)	7.8 ± 1.0

CC_{50} and CC_{90} concentrations (in brackets); results are obtained by MTT test (MTT), NR uptake cytotoxicity assay (NR) and CV staining technique (CV) after 24, 48, and 72 h of treatment; n.d. (not determined) marked the cases in which CC_{50} and CC_{90} were not calculated because the viability of the cells is >50% and respectively >10%; (-) = no data.

Table S3. Cytotoxic activity (CC_{50} and CC_{90}, μM) of the investigated complexes on cultured MDA-MB-231 cells.

Cell line	MDA-MB-231			
	MTT		NR	CV
	24 h	48 h	72 h	72 h
Znampy	n.d.	n.d.	(-)	256.6 ± 3.5
ZnampyAg	n.d.	n.d.	n.d.	n.d.
ZnampyAu	n.d.	n.d.	6.1 ± 2.2 (n.d.)	8.2 ±3 .7
Zndmen	n.d.	n.d.	(-)	235 ± 2.7
ZndmenAg	41.1 ± 3.2 (65.2 ± 1.7)	51.0 ± 5.9 (69.2 ± 4.9)	n.d.	112.4 ± 5.9
ZndmenAu	7.8 ± 1.4	5.2 ± 1.5 (n.d.)	2.7 ± 1.7 (5.2 ± 1.6)	1.8 ± 1.9
Znaepy	n.d.	n.d.	(-)	162.8 ± 7.1
ZnaepyAg	14.8 ± 5.9 (21.3 ± 5.3)	15.3 ± 4.2	(-)	8.2 ± 5.6 (18.8 ± 4.2)

CC_{50} and CC_{90} concentrations (in brackets); results are obtained by MTT test (MTT), NR uptake cytotoxicity assay (NR) and CV staining technique (CV) after 24, 48, and 72 h of treatment; n.d. (not determined) marked the cases in which CC_{50} and CC_{90} were not calculated because the viability of the cells is >50% and respectively >10%; (-) = no data.

Table S4. Cytotoxic activity (CC_{50} and CC_{90}, μM) of the investigated complexes on cultured HeLa cells.

Cell line	HeLa			
	MTT		NR	CV
	24 h	48 h	72 h	72 h
Znampy	157.9	98.7 ± 2.2 (145.1 ± 5.2)	86.2 ± 2.6 (137.3 ± 2.5)	112.6 ± 3.6
ZnampyAg	43.6 ± 0.3 (121 ± 0.8)	16.0 ± 1.5 (43.6 ± 1.8)	7.9 ± 0.8 (22.3 ± 1.5)	9.8 ± 2.0 (19.3 ± 2.6)
ZnampyAu	3.0 ± 0.9	0.6 ± 0.3 (0.9 ± 0.5)	0.3 ± 1.3 (2.9 ± 1.4)	1.0 ± 0.4 (2.4 ± 0.7)
Zndmen	n.d.	n.d.	n.d.	n.d.
ZndmenAg	15.9 ± 1.7 (26.8 ± 1.8)	13.2 ± 3.5 (25.4 ± 2.5)	11.8 ± 3.0 (23.9 ± 3.9)	12.3 ± 2.4 (25.5 ± 2.8)
ZndmenAu	2.6 ± 1.6 (20.3 ± 2.3)	0.7 ± 1.7 (2.7 ± 1.4)	0.6 ± 1.4 (2.4 ± 1.3)	1.2 ± 0.9 (8.2 ± 1.4)
Znaepy	n.d.	n.d.	n.d.	n.d.
ZnaepyAg	6.9 ± 0.8 (18.8 ± 1.4)	4.2 ± 2.4 (8.2 ± 2.9)	5.2 ± 3.7 (10.1 ± 3.2)	7.2 ± 3.1 (10.8 ± 3.5)

CC_{50} and CC_{90} concentrations (in brackets); results are obtained by MTT test (MTT), NR uptake cytotoxicity assay (NR) and CV staining technique (CV) after 24, 48, and 72h of treatment; n.d. (not determined) marked the cases in which CC_{50} and CC_{90} were not calculated because the viability of the cells is >50% and respectively >10%.

Table S5. Cytotoxic activity (CC_{50} and CC_{90}, μM) of the investigated complexes on cultured Lep-3 cells.

Cell line	Lep-3	
	NR	CV
	72 h	72 h
Znampy	45.0 ± 4.9 (66.5 ± 5.8)	36.1 ± 5.4 (66.0 ± 3.9)
ZnampyAg	9.0 ± 1.7 (12.0 ± 1.3)	9.0 ± 2.0 (12.0 ± 2.6)
ZnampyAu	0.4 ± 0.8 (0.5 ± 1.3)	0.3 ± 1.8 (0.5 ± 1.3)
Zndmen	23.4 ± 2.9 (62.1 ± 1.8)	20.5 ± 4.6 (29.6 ± 3.9)
ZndmenAg	8.3 ± 2.6 (12.7 ± 2.9)	7.9 ± 0.7 (13.1 ± 3.2)
ZndmenAu	0.9 ± 1.1 (2.7 ± 1.8)	1.9 ± 0.8 (2.7 ± 0.9)
Znaepy	28.7 ± 3.7 (61.5 ± 3.3)	20.9 ± 2.5 (29.0 ± 2.0)
ZnaepyAg	3.3 ± 1.9 (5.6 ± 2.4)	3.3 ± 3.0 (5.6 ± 2.2)

CC_{50} and CC_{90} concentrations (in brackets); results are obtained by NR uptake cytotoxicity assay (NR) and CV staining technique (CV) after 72 h of treatment.

Table S6. Cytotoxic activity (CC_{50} and CC_{90}, µM) of cisplatin on viability and proliferation of treated tumor and non-tumor cells.

Cell line	Cisplatin					
	MTT	**NR**	**MTT**	**NR**	**MTT**	**NR**
	24 h		48 h		72 h	
MCF-7	n.d.	n.d.	154.1 (n.d.)	166.0 (n.d.)	92.0 (n.d.)	99.6 (n.d.)
MDA-MB-231	n.d.	n.d.	54.3 (n.d.)	82.6 (n.d.)	51.6 (132.9)	60.0 (152.1)
HeLa	99.6 (-)	85.9 (-)	21.9 (55.5)	38.5 (61.0)	28.0 (62.9)	19.9 (56.6)
Lep-3	n.d.	70.0	27.5 (93.2)	17.6 (32.2)	1.6 (29.5)	(-)

CC_{50} and CC_{90} concentrations (in brackets) are obtained by MTT test (MTT) and NR uptake cytotoxicity assay (NR) after 24, 48, and 72 h of treatment; n.d. (not determined) marked the cases in which CC_{50} and CC_{90} were not calculated because the viability of the cells is >50% and respectively >10%; (-) = no data.

Table S7. Cytotoxic activity (CC_{50} and CC_{90}, µM) of oxaliplatin and epirubicin on viability and proliferation of treated tumor and non-tumor cells.

Cell line	Oxaliplatin			Epirubicin		
	24 h	**48 h**	**72 h**	**24 h**	**48 h**	**72 h**
MCF-7	n.d.	251.7 (n.d.)	111.3 (n.d.)	n.d.	52.4 (n.d.)	28.7 (n.d.)
MDA-MB-231	n.d.	43.7 (226.6)	25.1 (238.4)	131.8 (n.d.)	16.9 (184.0)	16.0 (132.3)
HeLa	170.0 (-)	38.9 (190.7)	15.4 (117.3)	113.3 (-)	35.6 (128.4)	31.9 (85.7)
Lep-3	174.4 (n.d.)	76.8 (251.6)	2.4 (125.8)	n.d.	45.8 (n.d.)	1.4 (33.7)

CC_{50} and CC_{90} concentrations (in brackets) are obtained by MTT test (MTT) after 24, 48, and 72 h of treatment; n.d. (not determined) marked the cases in which CC_{50} and CC_{90} were not calculated because the viability of the cells is >50% and respectively >10%; (-) = no data.