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Supporting Information

An Organometallic Gold(I) Bis-N-Heterocyclic Carbene Complex with Multimodal Activity in Ovarian Cancer Cells

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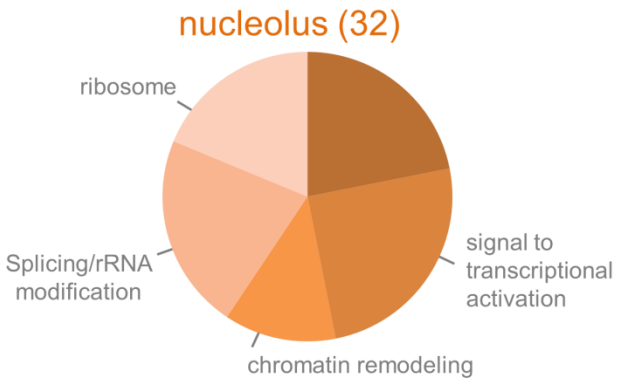
Supplementary Tables

Table S1. Groups of significantly regulated proteins associated with the nucleolus obtained from the proteomic response profiling experiments. The LFQ intensity values of a selection of regulated proteins are reported in cytoplasmic (CYT) and nuclear (NE) fraction. # = multi-parameter significant protein regulation (FDR = 0.05, S0 = 0.1).



Group	Fraction	FC	Gene	Accession
Stress-signal to transcriptional activation	CYT	5,26	HMOX1	P09601
	CYT	5,07	KDM2A	Q9Y2K7
	CYT	1,91	TXNRD1	Q16881
	CYT	1,20	STAT1	P42224
	CYT	-2,47	SMARCA4	P51532
	NE	6,67	HMOX1	P09601
	NE	1,49	PML	P29590
	NE	2,80	STAT1	P42224
chromatin remodeling	CYT	-1,25	FEN1	P39748
	NE	-1,22	TOP2A	P11388
	NE	1,12	XRCC5	P13010
	NE	1,32	CDCA8	Q53HL2
splicing/rRNA processing	CYT	-1,26	DDX5	P17844
	NE	1,27	DDX18	Q9NVP1
	NE	-1,24	DDX6	P26196
	NE	1,21	DIMT1	Q9UNQ2
	NE	1,66	IMP4	Q96G21
	NE	1,48	NSUN2	Q08J23
	NE	1,30	RNMT	O43148
ribosome	CYT	1,16	EIF6	P56537
	NE	-1,21	RPL13A	P40429
	NE	-1,19	RPL4	P36578
	NE	-1,33	RPS19	P39019
	NE	-1,22	SBDS	Q9Y3A5

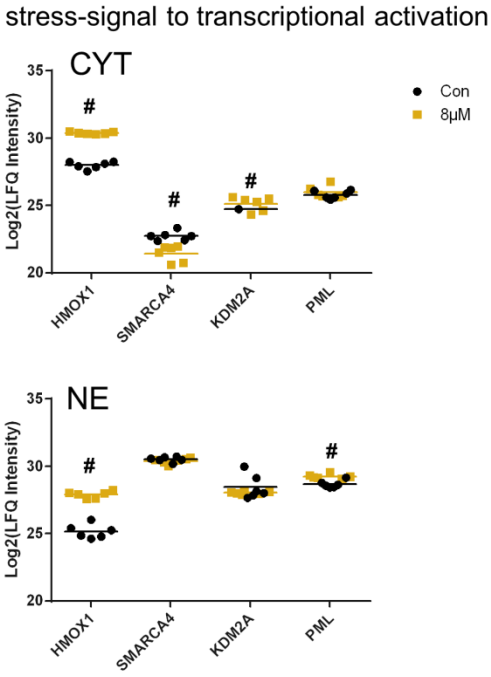


Table S2. Groups of significantly regulated proteins associated with telomeres obtained from the proteomic response profiling experiments. The LFQ intensity values of a selection of regulated proteins are reported in cytoplasmic (CYT) and nuclear (NE) fraction. # = multi-parameter significant protein regulation (FDR = 0.05, S0 = 0.1).

Telomere-associated proteins (14)

Group	Fraction	FC	Gene	Accession
telomere region	CYT	-1,25	FEN1	P39748
	CYT	-1,15	MCM6	Q14566
	CYT	-1,30	MSH2	P43246
	CYT	-1,26	POLD1	P28340
	NE	-1,44	CBX1	P83916
	NE	-1,42	CBX3	Q13185
	NE	2,49	HAT1	O14929
	NE	1,49	PML	P29590
	NE	1,30	RPA1	P27694
	NE	1,29	RPA2	P15927
	NE	1,12	XRCC5	P13010
telomere maintenance	NE	1,40	CCT6A	P40227
	NE	1,51	CCT8	P50990
	NE	1,48	TCP1	P17987

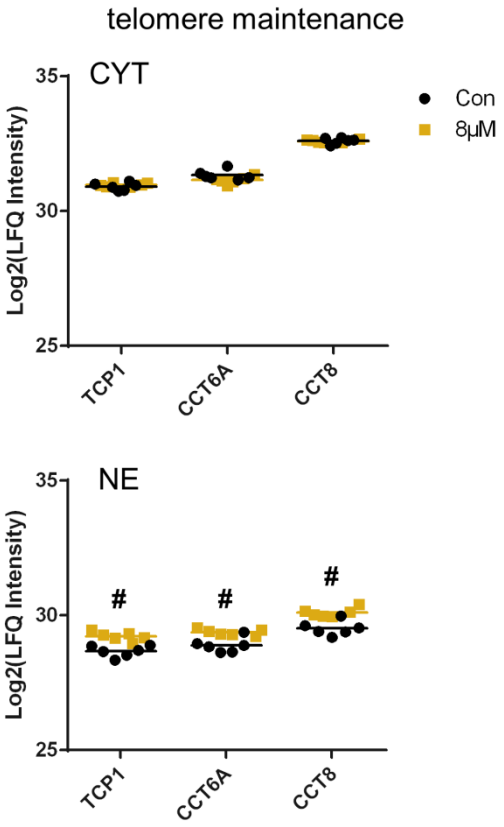


Table S3. Groups of significantly regulated proteins associated with actin stress fibers obtained from the proteomic response profiling experiments. The LFQ intensity values of a selection of regulated proteins are reported in cytoplasmic (CYT) and nuclear (NE) fraction. # = multi-parameter significant protein regulation (FDR = 0.05, S0 = 0.1).

Actin stress fibers (18)

Group	Fraction	FC	Gene	Accession
Actin stress fiber	CYT	1,31	FLNB	O75369
	NE	3,44	RRAS	P10301
	NE	-2,07	ACTN1	P12814
	NE	-2,04	ACTN4	O43707
	NE	-2,02	CFL2	Q9Y281
	NE	-1,38	CORO1C	Q9ULV4
	NE	-1,55	FSCN1	Q16658
	NE	-1,68	FLNA	P21333
	NE	-1,49	LIMA1	Q9UHB6
	NE	-1,88	PDLIM5	Q96HC4
	NE	-1,77	PDLIM7	Q9NR12
	NE	-1,70	ENAH	Q8N8S7
	NE	-1,60	RDX	P35241
	NE	-1,45	Sep 02	Q15019
	NE	-1,51	Sep 06	Q14141
	NE	-1,47	Sep 07	Q16181
	NE	-1,56	Sep 09	Q9UHD8
	NE	-1,56	Sep 11	Q9NVA2

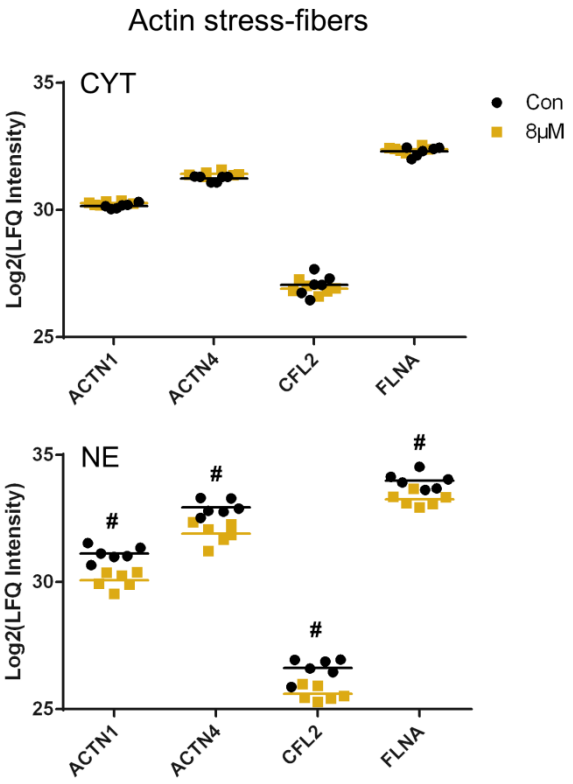
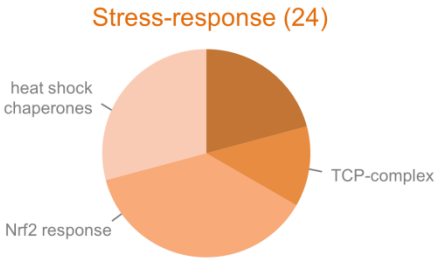


Table S4. Groups of significantly regulated proteins associated with stress-response obtained from the proteomic response profiling experiments. The LFQ intensity values of a selection of regulated proteins are reported in cytoplasmic (CYT) and nuclear (NE) fraction. # = multi-parameter significant protein regulation (FDR = 0.05, S0 = 0.1).



Group	Fraction	FC	Gene	Accession
TCP-complex	NE	1,40	CCT6A	P40227
	NE	1,51	CCT8	P50990
	NE	1,48	TCP1	P17987
Nrf2 response	CYT	1,52	GCLC	P48506
	CYT	2,09	GCLM	P48507
	CYT	1,30	GSR	P00390
	CYT	5,26	HMOX1	P09601
	CYT	-1,29	PRDX2	P32119
	CYT	-2,59	TXNL1	O43396
	CYT	1,91	TXNRD1	Q16881
	NE	4,35	GSTM5	P46439
	NE	6,67	HMOX1	P09601
heat shock chaperones	CYT	2,10	HSPA1B	P0DMV9
	CYT	1,59	HSPB1	P04792
	NE	1,35	DNAJB1	P25685
	NE	-2,53	DNAJB11	Q9UBS4
	NE	3,14	DNAJC7	Q99615
	NE	-1,23	HSPA5	P11021
	NE	4,51	HSPA6	P17066

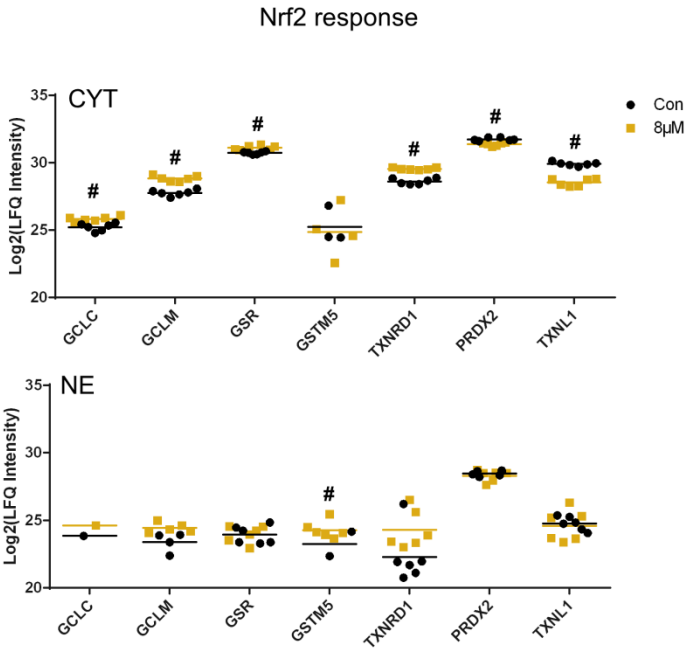
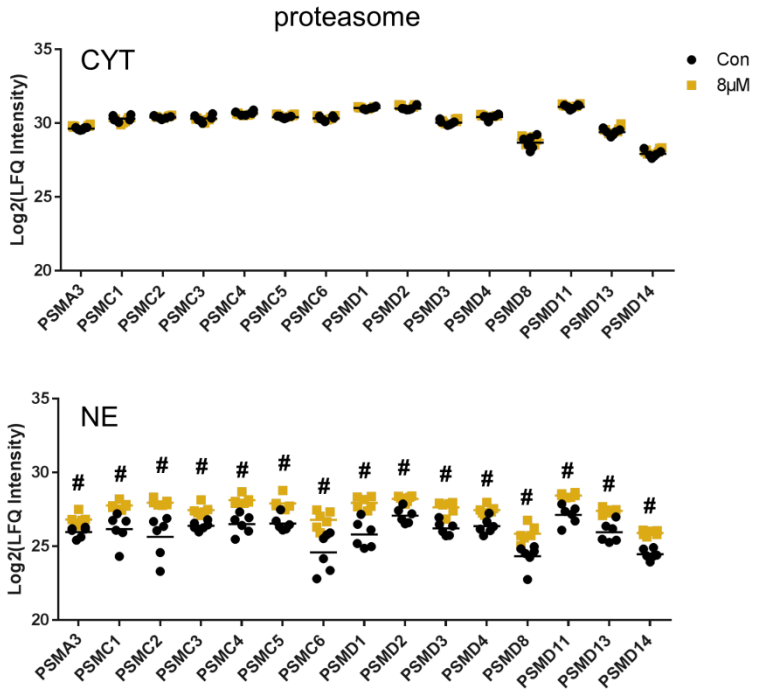


Table S5. Groups of significantly regulated proteins associated with the proteasome obtained from the proteomic response profiling experiments. The LFQ intensity values of a selection of regulated proteins are reported in cytoplasmic (CYT) and nuclear (NE) fraction. # = multi-parameter significant protein regulation (FDR = 0.05, S0 = 0.1).

Proteasome (19)

Group	Fraction	FC	Gene	Accession
proteasome	CYT	2,36	PSMB4	P28070
	CYT	-2,59	TXNL1	O43396
	NE	2,22	ADRM1	Q16186
	NE	1,80	PSMA3	P25788
	NE	3,03	PSMC1	P62191
	NE	4,97	PSMC2	P35998
	NE	2,09	PSMC3	P17980
	NE	3,08	PSMC4	P43686
	NE	2,56	PSMC5	P62195
	NE	4,58	PSMC6	P62333
	NE	4,42	PSMD1	Q99460
	NE	2,20	PSMD2	Q13200
	NE	2,69	PSMD3	O43242
	NE	2,12	PSMD4	P55036
	NE	2,86	PSMD8	P48556
	NE	2,50	PSMD11	O00231
	NE	2,73	PSMD13	Q9UNM6
	NE	2,71	PSMD14	O00487



Supplementary Figures

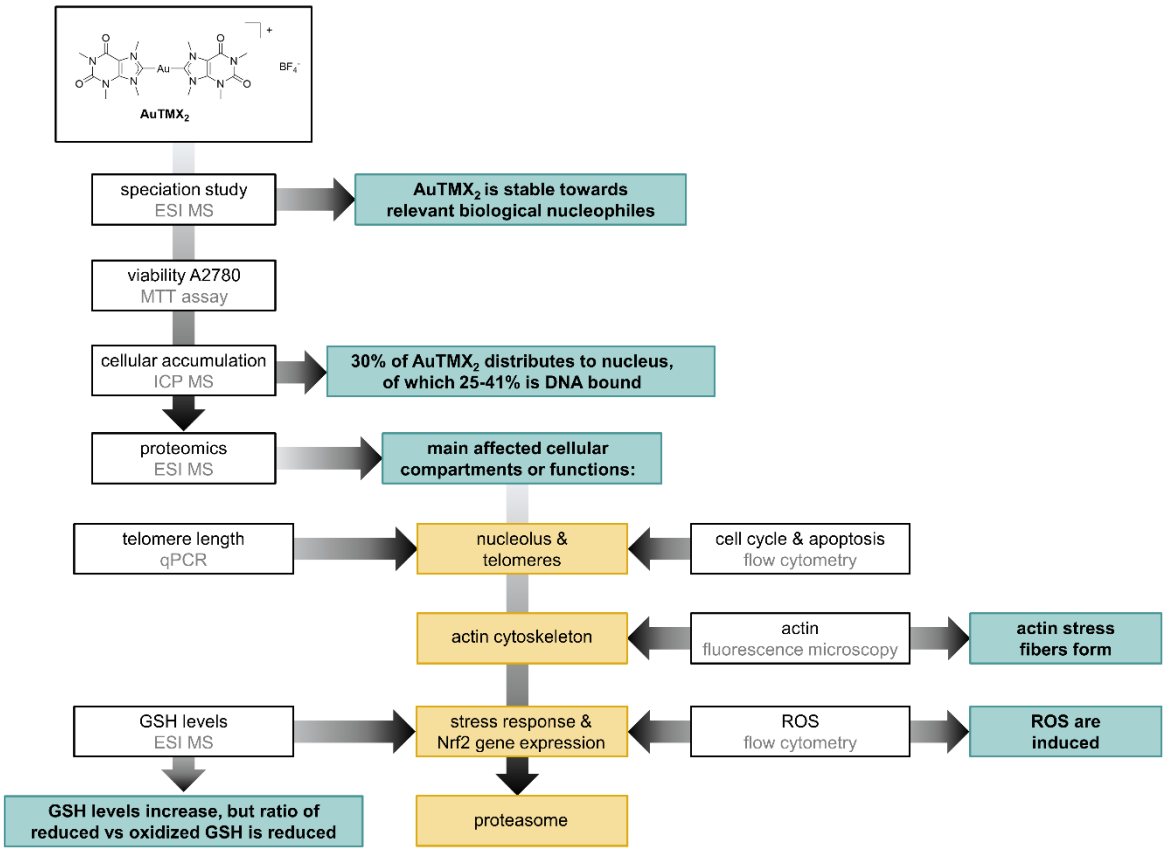


Figure S1. The figure summarizes the experimental workflow of this study and highlights the main results.

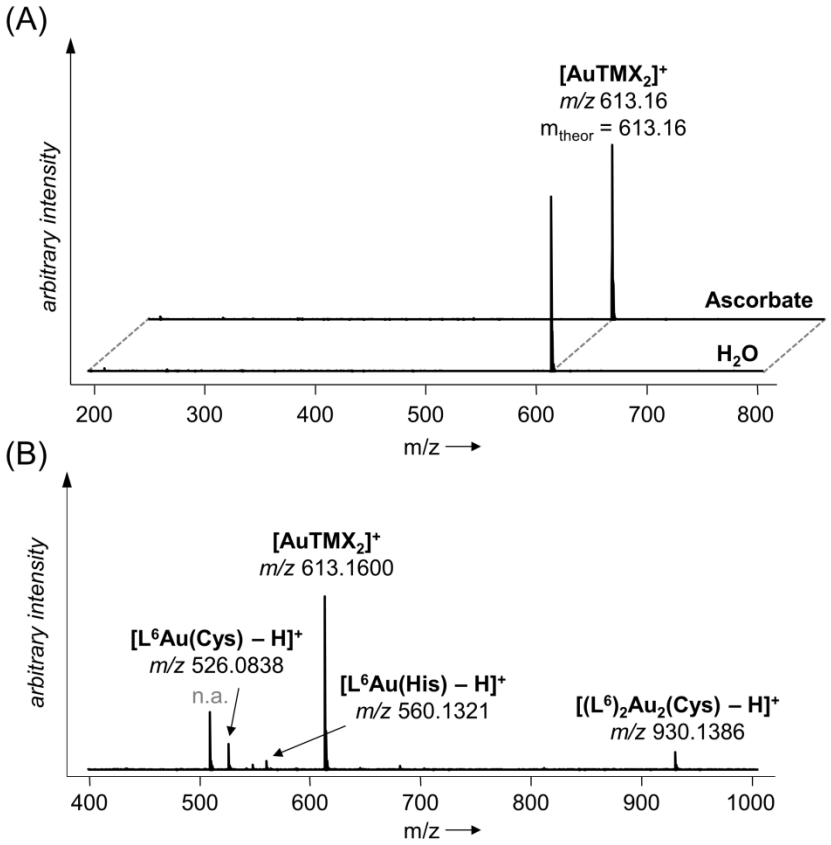


Figure S2. (**A**) ESI-ion trap mass spectrum of $AuTMX_2$ dissolved in aqueous solution or in the presence of ascorbate for 24 h. $AuTMX_2$ remains stable over the entire incubation period. (**B**) ESI-TOF mass spectrum of $AuTMX_2$ in the presence of His, Cys, Met, Glu and ^{Me}Se-Cys after 24 h. Adduct formation occurs exclusively with Cys and His after release of one TMX ligand.

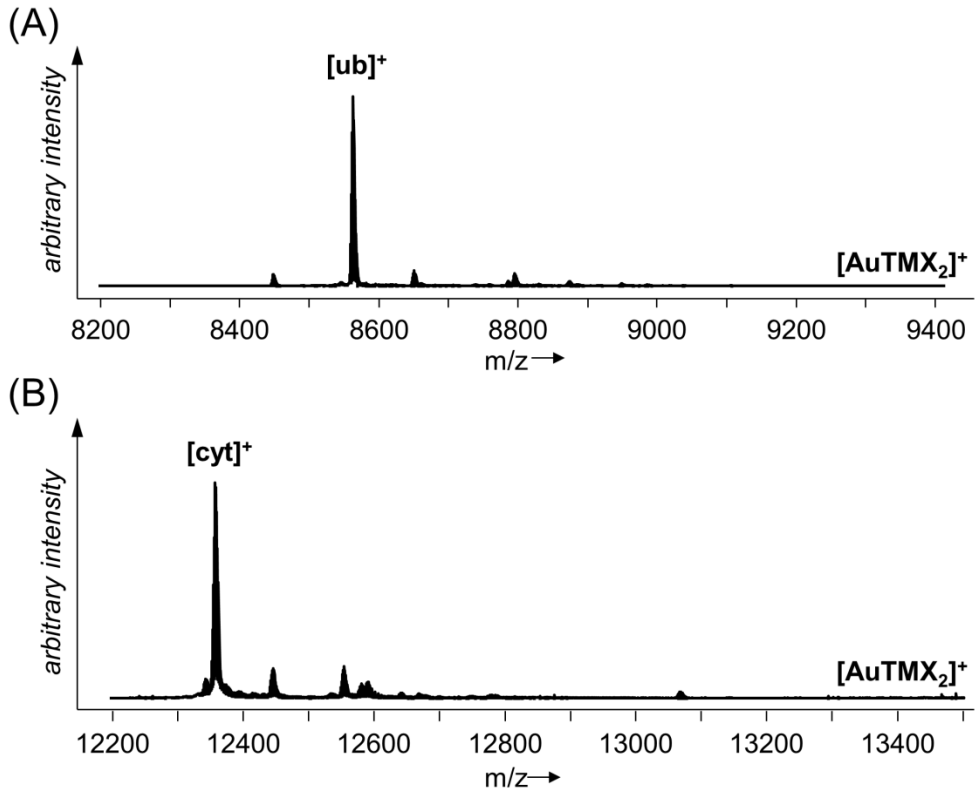


Figure S3. (**A**) Deconvoluted ESI-TOF mass spectra of ubiquitin (**A**) and cytochrome C (**B**) exposed to $AuTMX_2$ for 24 h. No adducts were observed.

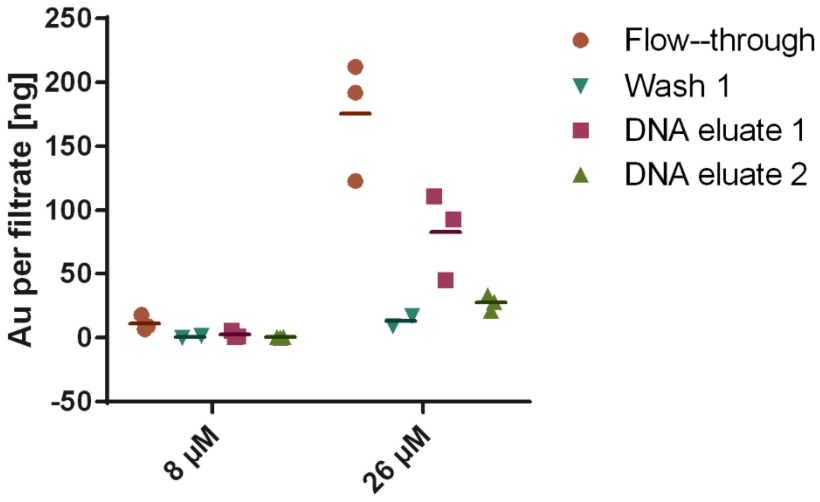


Figure S4. Amount of Au found during the DNA isolation process of A2780 (cancer) cells in filtrates after loading the lysate to the spin column (flow-through), after the first washing step (wash 1) and in both eluates. In samples treated for 24 h with 8 µM (2/3 EC_{50}) **AuTMX$_2$**, DNA concentration of the eluate 1 and 2 were 105.6 ± 18.9 ng μL^{-1} and 55.0 ± 8.6 ng μL^{-1}, respectively. After 26 µM (2×EC_{50}) treatment, DNA concentration of the eluate 1 and 2 were 110.7 ± 17.9 ng μL^{-1} and 51.4 ± 12.6 ng μL^{-1}, respectively.

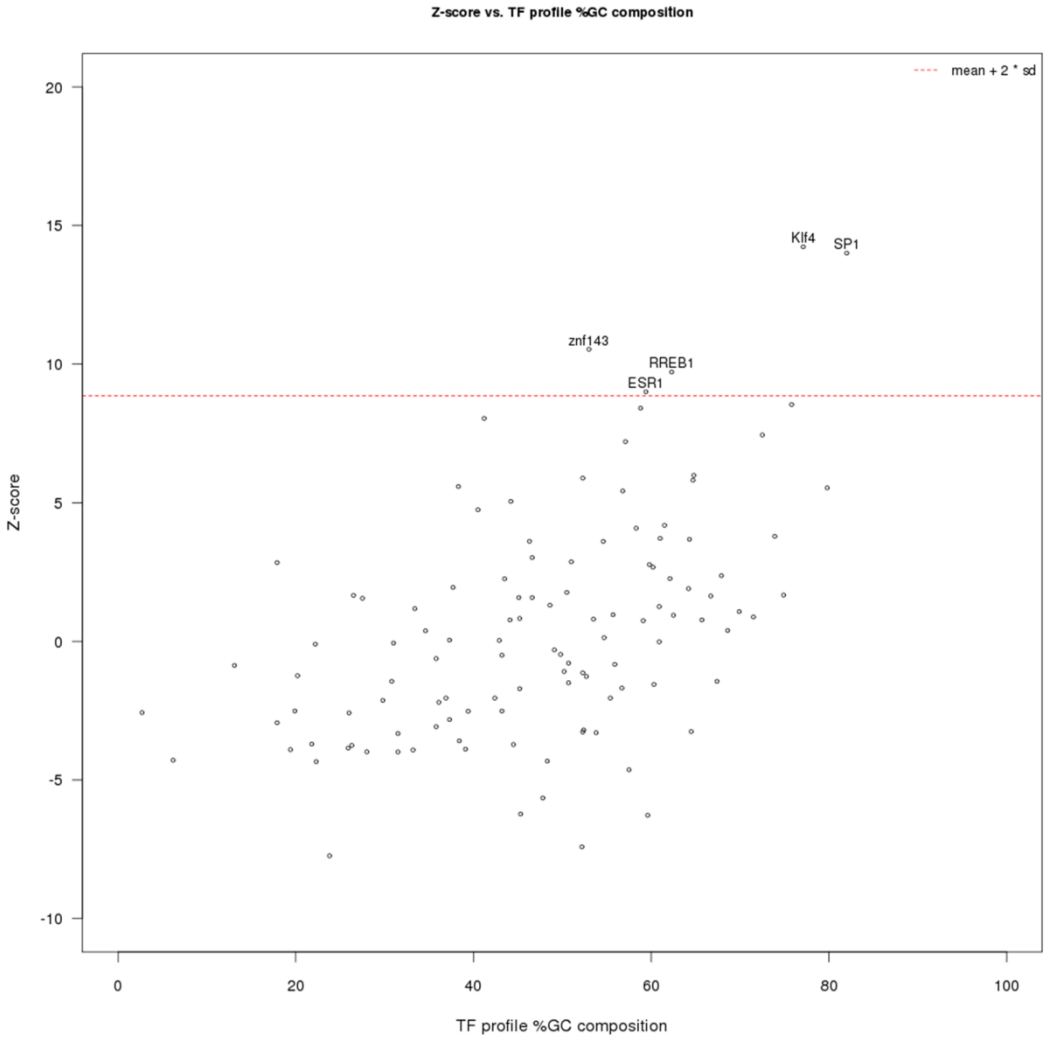


Figure S5. oPOSSUM search of enriched transcription factor activity from the set of significantly up-regulated proteins of **AuTMX$_2$**-treated A2780 cancer cells.

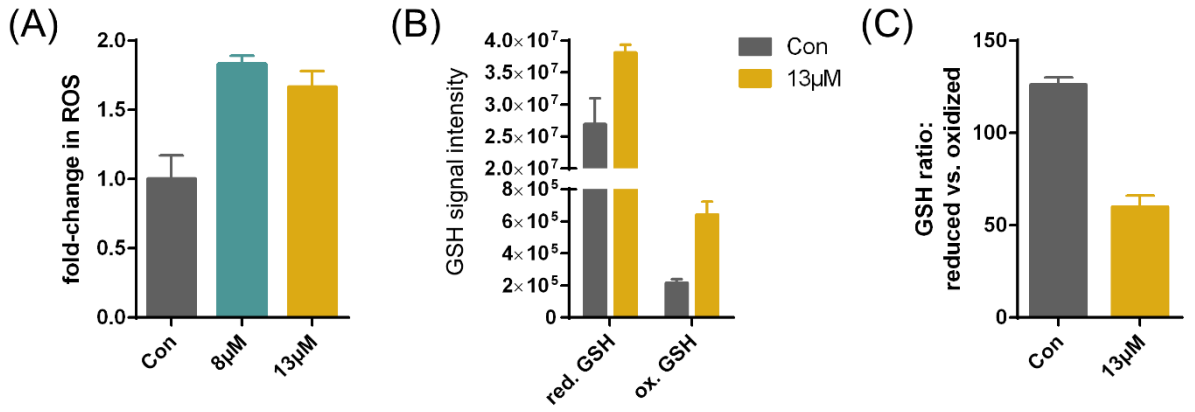


Figure S6. (**A**) Induction of reactive oxygen species (ROS) in A2780 cancer cells as measured by an increase in mean fluorescence intensity of the 2',7'-dichlorofluorescein (DCF) fluorophore upon treatment with **AuTMX$_2$** for 24 h. Data were set relative to control cells. p-value (one-way ANOVA) = 0.0004. (**B**) Signal intensity of GSH stemming from reduced and oxidized glutathione (GSH) after treating A2780 cancer cells with **AuTMX$_2$** at the indicated concentration for 24 h. (**C**) The ratio of reduced *vs* oxidized GSH of the signals in (B) as measured in untreated A2780 cancer cells and treated cells with **AuTMX$_2$** (13 μM) for 24 h. p-value (unpaired, two-tailed) = 0.0001.