

Supporting Information

Supermagnetic niosomal nanocarrier as a new approach for treatment of breast cancer: A case study on SK-BR-3 and MDA-MB-231 cell lines

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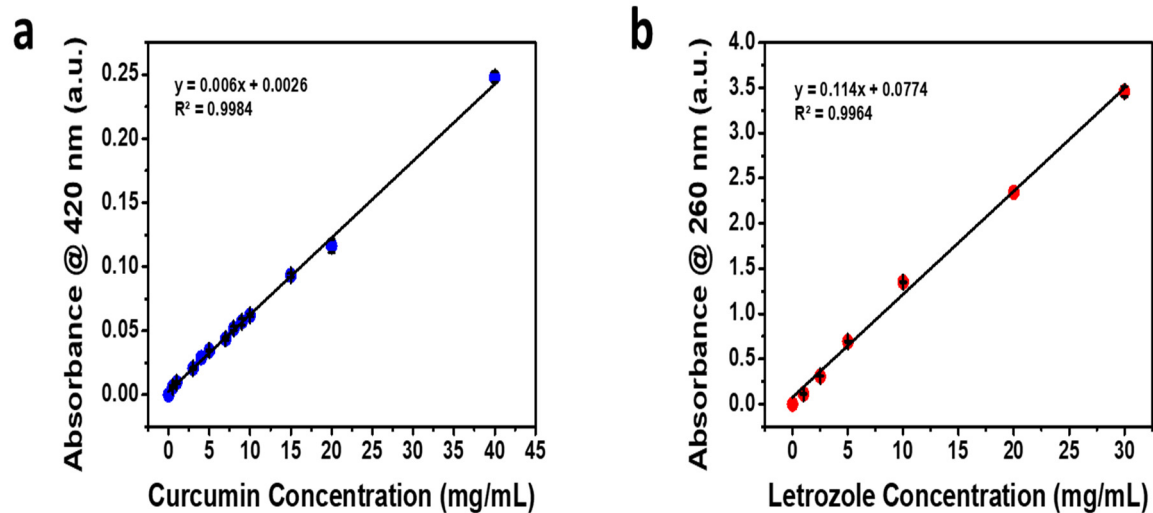


Figure S1. The calibration curve for absorbance of curcumin (a) and letrozole (b) versus their concentrations

Table S1. The sequence of primers used in real-time PCR analysis.	
Gene	Sequence of Primers
<i>Cyclin D</i>	Forward: 5'- CAGATCATCCGCAAACACGC-3' Revers: 5'- AAGTTGTTGGGGCTCCTCAG-3'
<i>b-actin</i>	Forward: 5'- TCCTCCTGAGCGCAAGTAC-3' Revers: 5'- CCTGCTTGCTGATCCACATCT-3'
<i>Caspase 3</i>	Forward: 5'- CATACTCCACAGCACCTGGTTA-3' Revers: 5'- ACTCAAATTCTGTTGCCACCTT-3'
<i>Caspase 9</i>	Forward: 5'-CATATGATCGAGGACATCCAG-3 Revers: 5'-TTAGTTCGCAGAAACGAAGC-3'
<i>Cyclin E</i>	Forward: 5'- CTCCAGGAAGAGGAAGGCAA-3' Revers: 5'- TTGGGTAAACCCGGTCATCA-3'
<i>Bax</i>	Forward: 5'-CGGCAACTTCAACTGGGG-3' Revers: 5'- TCCAGCCCAACAGCCG-3'
<i>Bcl2</i>	Forward: 5'- GGTGCCGGTTCAGGTACTCA-3' Revers: 5'- TTGTGGCCTTCTTTGAGTTCG-3'
<i>P53</i>	Forward: 5'- CATCTACAAGCAGTCACAGCACAT-3' Revers: 5'- CAACCTCAGGCGGCTCATAG-3'

Table S2. Magnetic properties of NiCoFe ₂ O ₄ and NiCoFe ₂ O ₄ @Silica nanoparticles by VSM			
Sample	Saturation magnetization (Ms) (emu/g)	Remnant field (Mr) (emu/g)	Coercivity (H _c) (gauss)
NiCoFe ₂ O ₄	17.58	5.55	805
NiCoFe ₂ O ₄ @Silica	11.71	4.06	1013

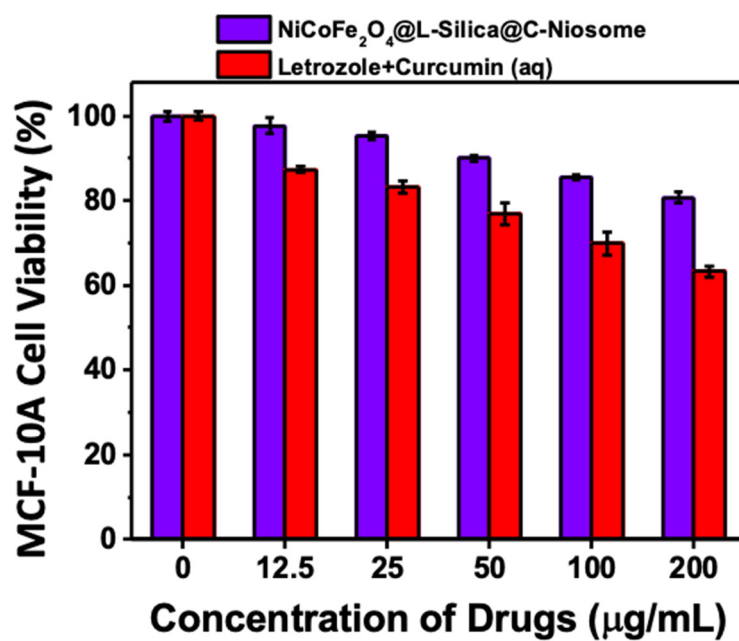


Figure S2. cytotoxicity effect of the as-synthesized nanocarriers against MCF-10A cell line

Table S3. A comparison for change in the IC50 values for different drugs before/after loading to different nanocarriers against and after incubation with different cell lines

Drug	Cell line	Time (hr)	Form	IC50	Ref
Curcumin	SK-BR-3	72	Free drug	127.39 µg/ml	[5]
			Loaded on niosomal formulation	69.77 µg/ml	
Curcumin	MDA-MB-231	72	Free drug	175.91 µg/ml	[5]
			Loaded on niosomal formulation	83.68 µg/ml	
Melittin	SK-BR-3	48	Free drug	87.87 µg/ml	[6]
			Nano	47.65 µg/ml	
		72	Free drug	50.56 µg/ml	
			Loaded on niosomal formulation	31.05 µg/ml	
Letrozole Curcumin	MDA-MB-231	48	Free Letrozole	354.81 µg/ml	This Study
			Free Curcumin	414.15 µg/ml	
			NiCoFe ₂ O ₄ @L-Silica@C-Niosome	93.47 µg/ml	
Letrozole Curcumin	SK-BR-3	48	Free Letrozole	337.82 µg/ml	This Study
			Free Curcumin	431.62 µg/ml	
			NiCoFe ₂ O ₄ @L-Silica@C-Niosome	39.48 µg/ml	

Table S4. apoptotic rate in presence of samples.

MDA-MB-231										
	Q1		Q2		Q3		Q2+Q3		Q4	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Control	0	0	0.012	0	0.47	0.005	0.34	0.40	99.9	1.1
S1	1.06	0.1	5.81	0.35	0	0	7.10	1.82	93.1	0.85
S2	0.46	0.08	4.7	0.21	5.05	0.42	9.43	0.46	89.79	1.34
S3	1.39	0.19	5.69	0.29	11.5	0.57	15.54	2.33	81.4	0.58
S4	2.22	0.24	7.05	0.08	4.28	0.21	12.80	2.07	86.5	0.72
S5	3.98	0.35	5.4	0.12	13.8	0.66	19.65	0.64	76.8	1.58
S6	2.37	0.07	21.3	1.34	14.8	0.18	37.70	2.26	61.5	0.37
SKBR3										
	Q1		Q2		Q3		Q2+Q3		Q4	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Control	0.684	0.05	0.289	0.041	0.289	0.01	0.81	0.32	98.7	1.24
S1	2.45	0.19	4.28	0.22	6.09	0.28	10.94	0.80	87.2	1.92
S2	1.44	0.031	3.95	0.74	10.2	0.79	13.13	1.45	84.4	2.21
S3	1.96	0.25	6.96	0.66	9.87	1.21	17.32	0.69	78.21	0.79
S4	1.94	0.07	9.29	0.19	11.2	0.97	21.85	1.92	77.5	0.68
S5	0.838	0.1	24.1	1.25	4.56	0.74	29.96	1.83	70.5	1.25
S6	0.963	0.02	28.1	1.34	21.9	1.34	48.63	1.94	49	1.47

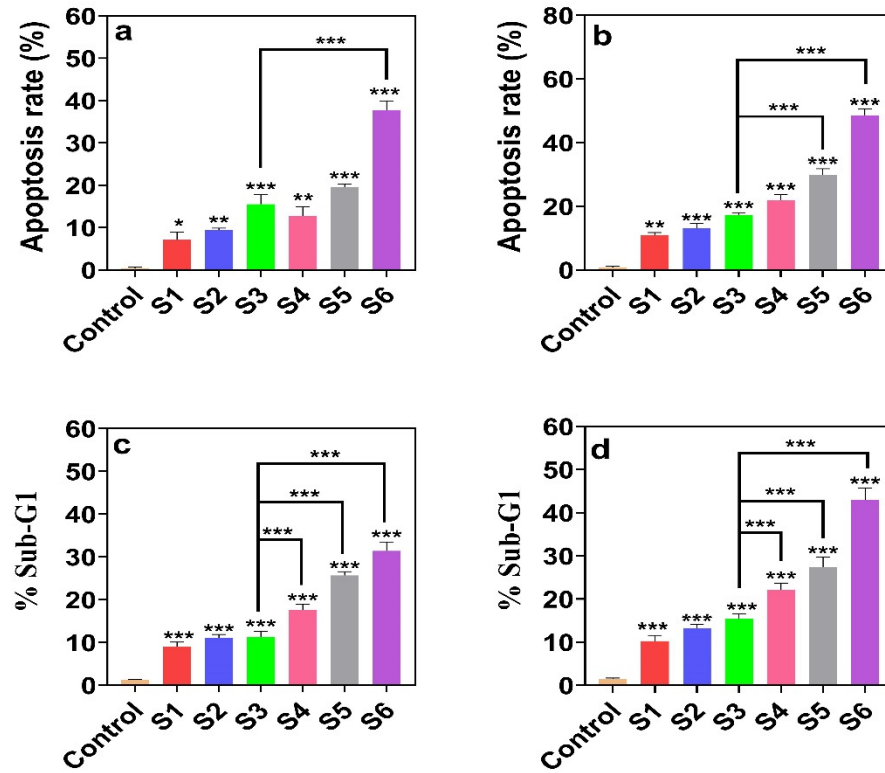


Figure S3. Statistical analysis for the apoptosis rate and Sub-G1% (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$) in different cell lines after treatment with different samples. **a** and **c**) MDA-MB-231 cell line, and **b** and **d**) SK-BR-3 cell line.

Table S5. Cell cycle rate of samples

MDA-MB-231								
	SubG1		G1		S		G2	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Control	1.2	0.12	49.24	0.67	24.46	1.68	25.1	1.68
S1	8.98	1.1	61.34	1.14	21.46	1.22	8.22	0.94
S2	11.11	0.78	53.07	2.57	16.45	0.39	19.37	1.1
S3	11.3	1.35	37.71	0.84	30.5	0.73	20.49	2.42
S4	17.56	1.42	57.3	0.57	19.2	0.29	5.94	1.33
S5	25.67	0.91	44.37	1.55	8.67	1.1	21.29	1.21
S6	31.4	2.13	48.13	1.02	5.37	0.58	15.1	1.48
SKBR3								
	SubG1		G1		S		G2	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Control	1.4	0.28	39.21	1.44	43.64	1.88	15.75	0.3
S1	10.25	1.34	62.21	0.67	22.35	0.92	5.19	0.75
S2	13.24	0.92	50.1	0.33	25.09	1.34	11.57	1.25
S3	15.42	1.1	42.33	0.88	28.04	1.12	14.21	1.34
S4	22.19	1.54	41.37	0.49	12.12	0.64	24.32	0.92
S5	27.44	2.33	60.24	0.23	2.19	0.42	10.13	1.13
S6	42.94	2.75	26.95	0.68	13.54	0.92	16.57	0.68