Electronic Supplementary Material (ESI) for RSC Advances.

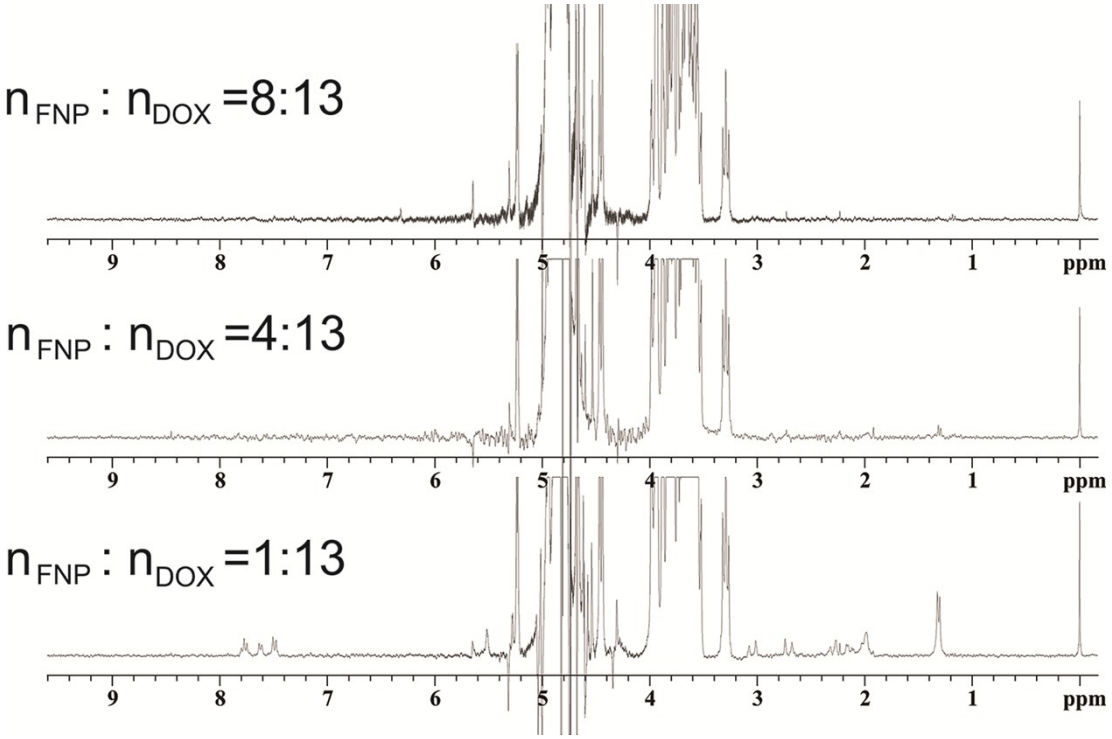

Figure S1. ^{1}H NMR spectra of FNP/DOX nanocomposite. The effect of different molar ratio of FNP and DOX on the chemical shift of the aromatic protons and methyl group of doxorubicin

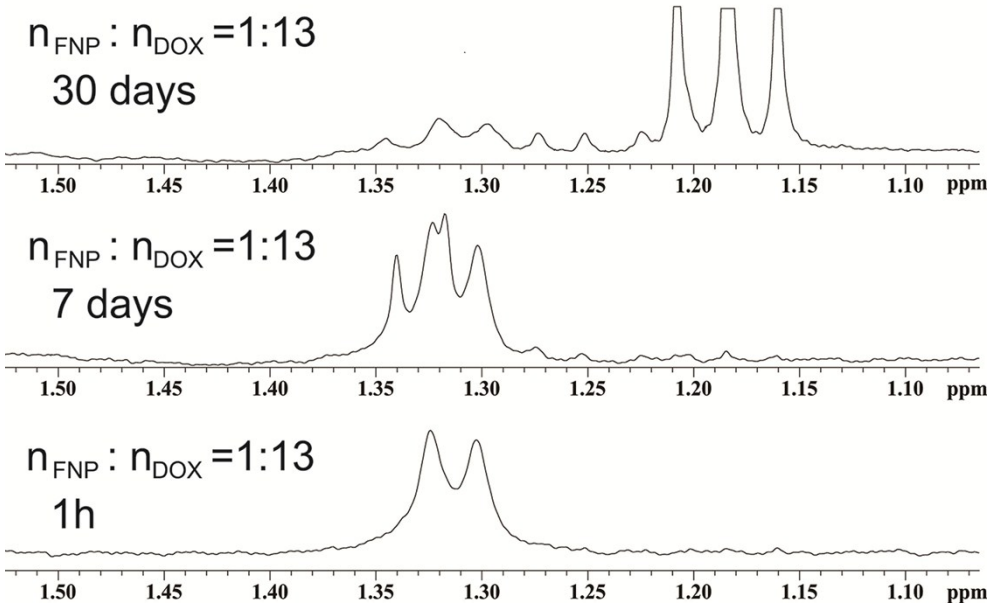

Figure S2. ^{1}H NMR spectra of FNP/DOX nanocomposite. Chemical shift of methyl group at molar ratio nFNP : nDOX = 1 : 13 after 30 days, after seven days, and after 1 h of preparation

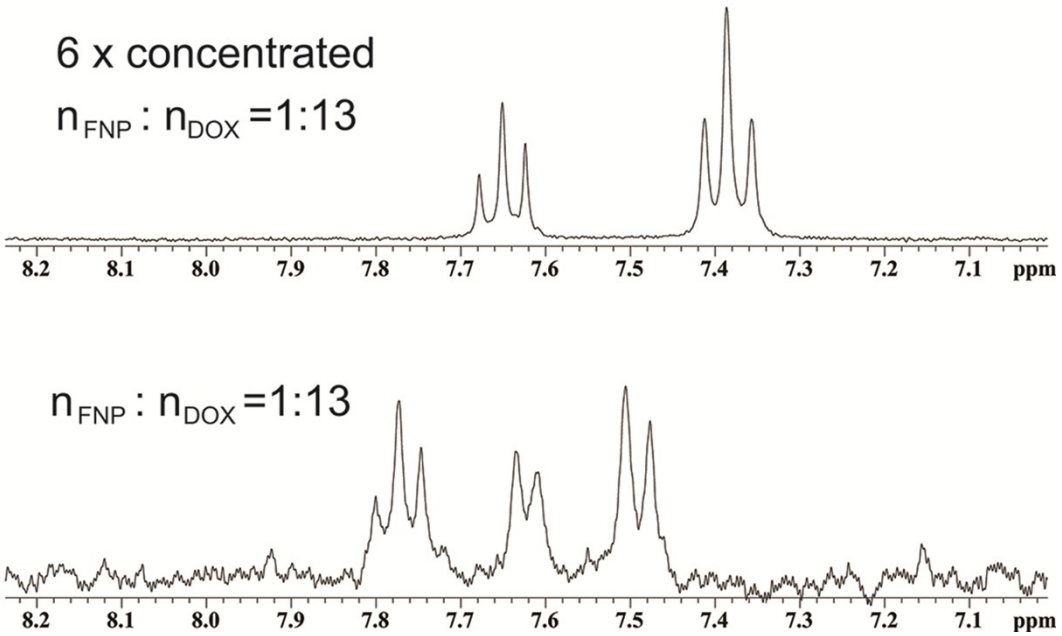

Figure S3. ^{1}H NMR spectra of FNP/DOX nanocomposite. Chemical shift of aromatic protons in the nanocomposite of molar ratio nFNP : nDOX = 1 : 13 in concentrated sample (1 : 6), and unconcentrated sample

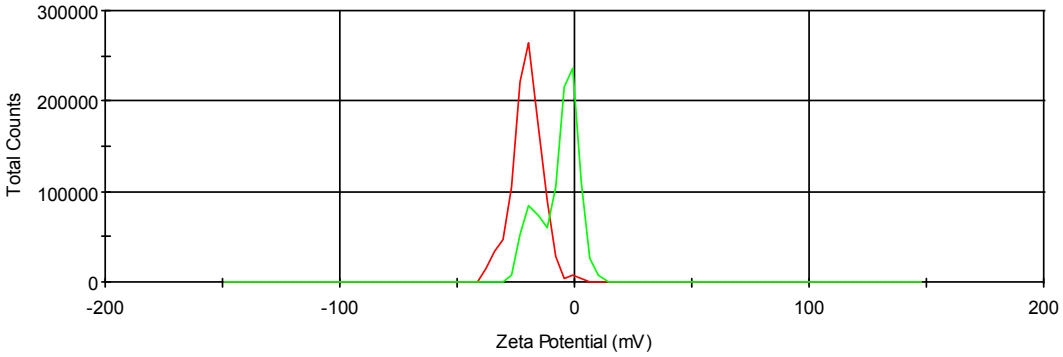

Figure S4. ζ potential of FNP and FNP/DOX in aqueous solutions after 48 h. Red line referes to FNP and green line referes to FNP/DOX. Red line refers to FNP and green refers to FNP/DOX. Three independent measurements were performed and one representative is displayed. Measurements were done by Zetasizer Nano based on method of dynamic light scattering.

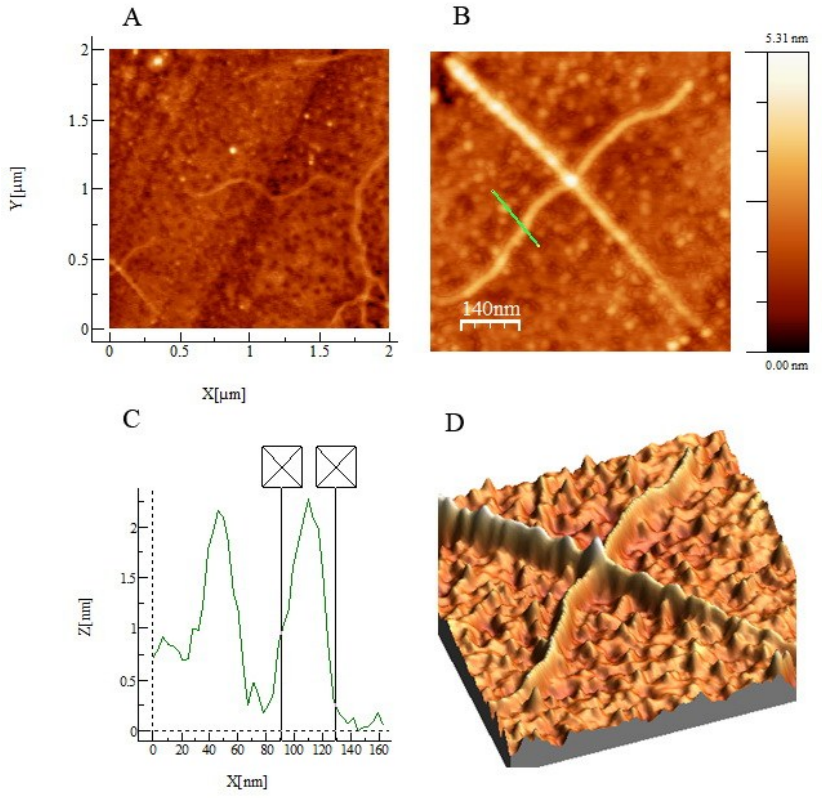

Figure S5. AFM measurement of FNP/DOX nanocomposite, on HOPG surface, after 30-day storage in dark at 22 °C a) scale 2000 x 2000 nm^2 b) scale 670 x 670 nm^2 c) corresponding cross-section d) 3D image

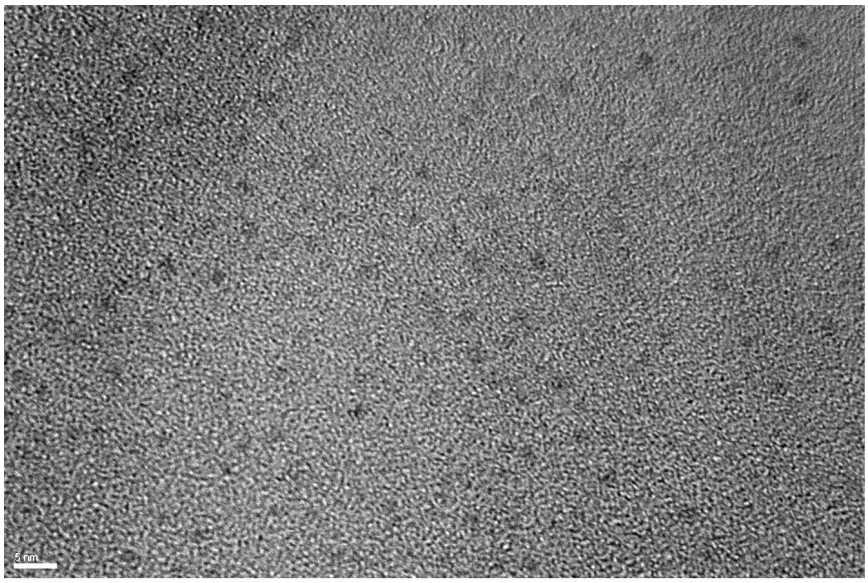

Figure S6. TEM measurement of aqueous solution of FNP immediately after sonication. The size of FNP nanoparticles is around 2 nm. Measurements were performed on copper grid 300 mesh.

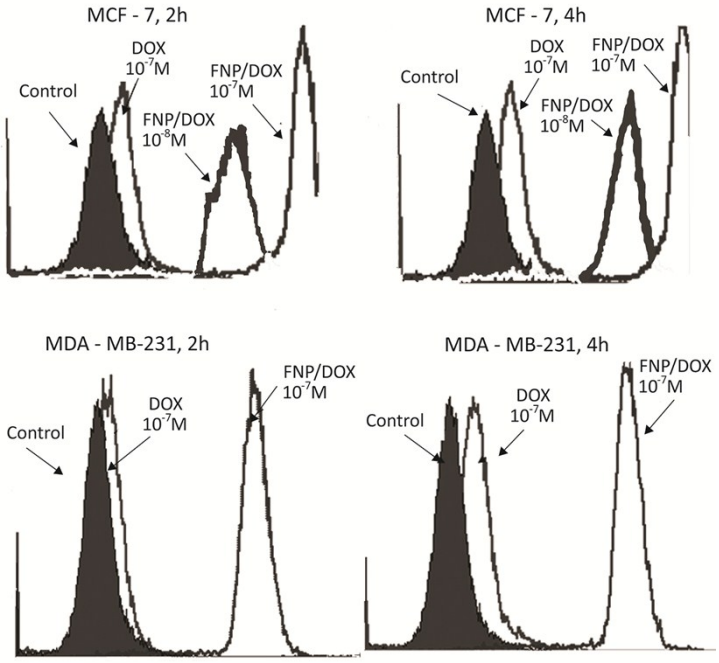

Figure S7. Uptake of DOX and FNP/DOX by human breast cancer cells (MCF-7, ER^+; MDA-MB-231, ER^-). Control cells and cells treated with DOX and FNP/DOX were incubated at 37 °C for 2 and 4 hours. Fluorescent data were achieved using flow cytometry analysis. Results are presented as overlaid histograms of one representative experiment from three independent experiments with similar results.

Table S1. Distribution of MCF-7 cells between the cell cycle phases (%) in regard to treatments in four different time points

Control		G0G1	S	G2M	subG1
	2h	54.63	26.32	17.69	2.01
	4h	nr	nr	nr	nr
	24h	65.19	17.29	15.39	2.93
	48h	69.93	17.27	11.69	1.97

FNP		G0G1	S	G2M	subG1
	2h	nr	nr	nr	nr
	4h	nr	nr	nr	nr
	24h	69.09	18.46	9.19	4.34
	48h	62.44	29.35	8.57	0.60

DOX		G0G1	S	G2M	subG1
	2h	54.19	24.05	20.37	1.27
	4h	51.66	25.74	20.25	3.57
	24h	18.78	6.83	73.26	1.71
	48h	63.25	13.65	21.46	2.34

FNP/DOX 0.1µM		G0G1	S	G2M	subG1
	2h	50.77	28.48	18.87	2.88
	4h	45.56	35.75	14.41	5.62
	24h	53.32	24.96	15.03	7.70
	48h	53.67	34.84	8.88	3.35

FNP/DOX 0.01µM		G0G1	S	G2M	subG1
	2h	53.97	25.43	19.00	2.91
	4h	53.39	24.76	18.93	6.68
	24h	61.88	21.26	15.52	2.11
	48h	50.90	35.76	6.41	8.27

Results present one representative experiment from three independent experiments with similar results. nr means that data were not recorded at specified time points.

Table S2. Distribution of MDA-MB-231 cells between the cell cycle phases (%) in regard to treatments in four different time points

Control		G0G1	S	G2M	subG1
	2h	nr	nr	nr	nr
	4h	nr	nr	nr	nr
	24h	53.65	34.64	9.84	1.23
	48h	nr	nr	nr	nr

FNP		G0G1	S	G2M	subG1
	2h	nr	nr	nr	nr
	4h	nr	nr	nr	nr
	24h	56.96	30.95	11.12	1.62
	48h	58.89	25.49	13.34	1.37

DOX		G0G1	S	G2M	subG1
	2h	55.86	31.04	13.03	0.77
	4h	55.77	32.45	11.49	0.92
	24h	50.71	28.25	20.5	1.36
	48h	60.15	26.2	11.43	1.16

FNP/DOX 0.1μM		G0G1	S	G2M	subG1
	2h	56.09	31.95	11.08	1.63
	4h	50.72	35.09	9.84	5.68
	24h	55.84	32.1	8.58	4.95
	48h	59.3	28.02	11.8	0.13

Results present one representative experiment from three independent experiments with similar results. nr means that data were not recorded at specified time points.

Table S3. Lethal and teratogenic effects observed in zebrafish (*Danio rerio*) embryos at different hours post fertilization (hpf).

Category	Developmental endpoints	Exposure time (hpf)			
		24	48	72	96
Lethal effect	Egg coagulation[a]	•	•	•	•
	No somite formation	•	•	•	•
	Tail not detached	•	•	•	•
	No heart-beat		•	•	•
Teratogenic effect	Malformation of head	•	•	•	•
	Malformation of eyes[b]	•	•	•	•
	Malformation of sacculi/otoliths[c]	•	•	•	•
	Malformation of chorda	•	•	•	•
	Malformation of tail[d]	•	•	•	•
	Scoliosis	•	•	•	•
	Heart beat frequency		•	•	•
	Blood circulation		•	•	•
	Pericardial edema	•	•	•	•
	Yolk edema	•	•	•	•
	Yolk deformation	•	•	•	•
	Growth retardation[e]	•	•	•	•

[a] No clear organs structure are recognized

[b] Malformation of eyes was recorded for the retardation in eye development and abnormality in shape and size.

[c] Presence of no, one or more than two otoliths per sacculus, as well as reduction and enlargement of otoliths and/or sacculi (otic vesicles).

[d] Tail malformation was recorded when the tail was bent, twisted or shorter than to control embryos as assessed by optical comparation.

[e] Growth retardation was recorded by comparing with the control embryos in development or size (before hatching, at 24 hpf and 48 hpf) or in a body length (after hatching, at and onwards 72 hpf) using by optical comparation using a inverted microscope (CKX41; Olympus, Tokyo, Japan).

Table S4. Effect percentages for abnormal morphological characteristics evaluated in the zebrafish toxicity assay after 96 h exposure to fullerenol (FNP).

FNP	Lethal embryos[a]	Teratogenic embryos[a]	Normal embryos[a]	Growth retardation[b]	Notochord[b]	Eyes[b]	Otoliths[b]	Somites[b]	Tail detachment[b]	Heart beat[b]	Pericardial edema[b]	Blood circulation[b]	Yolk edema[b]	Hatched[b]	Head deformity[c]	Skeletal deformities[c]	Jaw deformity[c]	Tail deformity[c]
24 hpf																		
92 µM	1.11	0.00	98.89	0.00	0.00	0.00	0.00	0.00	0.00	0.00	-	-	-	-	-	-	-	-
46 µM	0.00	0.00	100.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	-	-	-	-	-	-	-	-
9.2 µM	0.00	0.00	100.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	-	-	-	-	-	-	-	-
4.6 µM	0.00	0.00	100.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	-	-	-	-	-	-	-	-
48 hpf																		
92 µM	2.22	0.00	97.78	0.00	0.00	0.00	0.00	0.00	0.00	95.56	0.00	0.00	0.00	-	-	-	-	-
46 µM	0.00	0.00	100.00	0.00	0.00	0.00	0.00	0.00	0.00	89.56	0.00	0.00	0.00	-	-	-	-	-
9.2 µM	0.00	0.00	100.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	-	-	-	-	-
4.6 µM	0.00	0.00	100.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	-	-	-	-	-
72 hpf																		
92 µM	4.44	6.67	88.89	0.00	0.00	0.00	0.00	0.00	-	100.00	3.49	0.00	0.00	100.00	0.00	6.98	0.00	0.00
46 µM	0.00	0.00	100.00	0.00	0.00	0.00	0.00	0.00	-	100.00	0.00	0.00	0.00	100.00	0.00	0.00	0.00	0.00
9.2 µM	0.00	0.00	100.00	0.00	0.00	0.00	0.00	0.00	-	0.00	0.00	0.00	0.00	100.00	0.00	0.00	0.00	0.00
4.6 µM	0.00	0.00	100.00	0.00	0.00	0.00	0.00	0.00	-	0.00	0.00	0.00	0.00	100.00	0.00	0.00	0.00	0.00
96 hpf																		
92 µM	4.44	6.67	88.89	100.00	0.00	0.00	0.00	0.00	-	100.00	3.49	0.00	0.00	100.00	0.00	6.98	0.00	0.00
46 µM	0.00	0.00	100.00	0.00	0.00	0.00	0.00	0.00	-	0.00	0.00	0.00	0.00	100.00	0.00	0.00	0.00	0.00
9.2 µM	0.00	0.00	100.00	0.00	0.00	0.00	0.00	0.00	-	0.00	0.00	0.00	0.00	100.00	0.00	0.00	0.00	0.00
4.6 µM	0.00	0.00	100.00	0.00	0.00	0.00	0.00	0.00	-	0.00	0.00	0.00	0.00	100.00	0.00	0.00	0.00	0.00

Table S5. Effect percentages for abnormal morphological characteristics evaluated in the zebrafish toxicity assay after exposure to doxorubicin (DOX).

DOX	Normal embryos[a]	Lethal embryos[a]	Teratogenic embryos[a]	Growth retardation[b]	Notochord[b]	Eyes[b]	Otoliths[b]	Somites[b]	Tail detachment[b]	Pericardial oedema[b]	Yolk oedema[b]	Heart beat[b]	Blood circulation[b]	Hatching[b]	Head[c]	Scoliosis[c]	Tail[c]
24 hpf																	
50 µg/ml	74.0	26.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0						
5 µg/ml	74.7	25.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0						
2.5 µg/ml	77.1	22.9	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0						
1 µg/ml	81.3	18.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0						
0.5 µg/ml	87.5	12.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0						
0.1 µg/ml	87.5	12.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0						
48 hpf																	
50 µg/ml	0.0	34.0	66.0	0.0	69.7	0.0	100.0	0.0	0.0	100.0	100.0	100.0	100.0	6.1	100.0	100.0	100.0
5 µg/ml	70.7	25.3	4.0	0.0	0.0	0.0	0.0	0.0	0.0	1.8	1.8	1.8	1.8	50.9	0.0	7.1	3.6
2.5 µg/ml	77.1	22.9	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	78.4	0.0	0.0	0.0
1 µg/ml	81.3	18.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	64.1	0.0	0.0	0.0
0.5 µg/ml	87.5	12.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	38.1	0.0	0.0	0.0
0.1 µg/ml	87.5	12.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	28.6	0.0	0.0	0.0
72 hpf																	
50 µg/ml	0.0	56.0	44.0	9.1	100.0	0.0	90.9	0.0	0.0	100.0	100.0	100.0	100.0	81.8	100.0	100.0	100.0
5 µg/ml	68.0	25.3	6.7	0.0	3.6	0.0	0.0	0.0	0.0	5.5	5.5	5.5	5.5	98.2	0.0	5.6	0.0
2.5 µg/ml	75.0	22.9	2.1	0.0	2.7	0.0	0.0	0.0	0.0	2.7	0.0	0.0	2.7	100.0	0.0	0.0	0.0
1 µg/ml	81.3	18.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	100.0	0.0	0.0	0.0
0.5 µg/ml	87.5	12.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	100.0	0.0	0.0	0.0
0.1 µg/ml	87.5	12.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	100.0	0.0	0.0	0.0
96 hpf																	
50 µg/ml	0.0	96.0	4.0	0.0	100.0	0.0	0.0	0.0	0.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0
5 µg/ml	69.3	26.7	4.0	0.0	1.8	0.0	0.0	0.0	0.0	1.8	1.8	1.8	1.8	100.0	0.0	3.6	0.0
2.5 µg/ml	75.0	22.9	2.1	0.0	2.7	0.0	0.0	0.0	0.0	2.7	0.0	0.0	2.7	100.0	0.0	2.7	0.0
1 µg/ml	81.3	18.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	100.0	0.0	0.0	0.0
0.5 µg/ml	87.5	12.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	100.0	0.0	0.0	0.0
0.1 µg/ml	87.5	12.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	100.0	0.0	0.0	0.0

Abrevitation used: (hpf) hours post fertilisation.
[a] Percentage of mortality and teratogenicity based on all eggs.
[b] Percentage of teratogenic effect based on all alive embryos at the time of assessment.
[c] Percentage of teratogenic effect based on all hatched embryos at the time of assessment.

Table S6. Effect percentages for abnormal morphological characteristics evaluated in the zebrafish toxicity assay after exposure to a nanocomposite (FNP/DOX).

FNP/DOX	Normal embryos[a]	Lethal embryos[a]	Teratogenic embryos[a]	Growth retardation[b]	Notochord[b]	Eyes[b]	Otoliths[b]	Somites[b]	Tail detachment[b]	Pericardial oedema[b]	Yolk oedema[b]	Heart beat[b]	Blood circulation[b]	Hatching[b]	Head[c]	Scoliosis[c]	Tail[c]
24 hpf																	
50 µg/ml	54.2	27.1	18.8	0.0	0.0	0.0	17.1	0.0	0.0	2.9	5.7						
5 µg/ml	64.0	32.0	4.0	0.0	0.0	0.0	0.0	0.0	0.0	2.0	0.0						
2.5 µg/ml	68.8	31.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0						
1 µg/ml	79.2	20.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0						
0.5 µg/ml	84.0	16.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0						
0.1 µg/ml	83.3	16.7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0						
48 hpf																	
50 µg/ml	0.0	31.3	68.8	0.0	0.0	0.0	18.2	0.0	0.0	42.4	36.4	93.9	42.4	33.3	0.0	45.5	9.1
5 µg/ml	61.3	32.0	6.7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	60.8	0.0	3.2	6.5
2.5 µg/ml	68.8	31.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	72.7	0.0	0.0	0.0
1 µg/ml	79.2	20.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	92.1	0.0	0.0	0.0
0.5 µg/ml	84.0	16.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	38.1	0.0	0.0	0.0
0.1 µg/ml	83.3	16.7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	40.0	0.0	0.0	0.0
72 hpf																	
50 µg/ml	0.0	33.3	66.7	0.0	100.0	0.0	78.1	0.0	0.0	93.8	93.8	93.8	100.0	93.8	36.7	36.7	36.7
5 µg/ml	60.0	33.3	6.7	0.0	2.0	0.0	0.0	0.0	0.0	4.0	4.0	4.0	4.0	94.0	2.1	4.3	2.1
2.5 µg/ml	68.8	31.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	100.0	0.0	3.0	3.0
1 µg/ml	79.2	20.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	97.4	0.0	0.0	0.0
0.5 µg/ml	84.0	16.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	100.0	0.0	0.0	0.0
0.1 µg/ml	83.3	16.7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	100.0	0.0	0.0	0.0
96 hpf																	
50 µg/ml	0.0	68.8	31.3	0.0	86.7	0.0	86.7	0.0	0.0	93.3	93.3	86.7	93.3	93.3	100.0	85.7	78.6
5 µg/ml	53.3	34.7	12.0	0.0	4.1	0.0	0.0	0.0	0.0	4.1	6.1	0.0	4.1	98.0	4.2	14.6	2.1
2.5 µg/ml	66.7	31.3	2.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	100.0	0.0	3.0	3.0
1 µg/ml	79.2	20.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	97.4	0.0	0.0	0.0
0.5 µg/ml	84.0	16.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	100.0	0.0	0.0	0.0
0.1 µg/ml	83.3	16.7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	100.0	0.0	0.0	0.0

Abrevitation used: (hpf) hours post fertilisation.
[a] Percentage of mortality and teratogenicity based on all eggs.
[b] Percentage of teratogenic effect based on all alive embryos at the time of assessment.
[c] Percentage of teratogenic effect based on all hatched embryos at the time of assessment.