

Figure S1 The size distribution of ZnO NPs in cell culture medium.

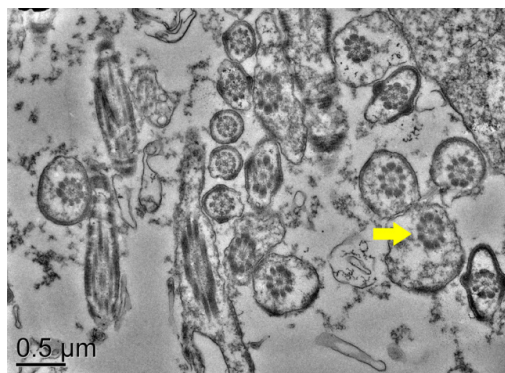


Figure S2 The TEM image of sperm cell in testis tissue of 350 mg/kg SD rats. The yellow arrow indicates the degraded mitochondrial sheath of sperm cell.

Table S1 EDX analysis results of particles in the testis

Element	Apparent concentration (KeV)	K ratio	Wt%	Wt% Sigma	Standard sample label	Manufacturer standard
O	34.15	0.11491	28.41	0.14	SiO ₂	Yes
Mg	0.17	0.00115	0.29	0.04	MgO	Yes
Al	6.07	0.04363	8.44	0.06	Al ₂ O ₃	Yes
Si	6.24	0.04948	7.86	0.05	SiO ₂	Yes
P	0.55	0.00307	0.44	0.02	GaP	Yes
Ti	0.29	0.00293	0.29	0.02	Ti	Yes
Ni	8.32	0.08323	7.44	0.07	Ni	Yes
Cu	26.84	0.26839	25.01	0.12	Cu	Yes
Zn	0.26	0.00259	0.24	0.06	Zn	Yes
Pb	4.48	0.04171	5.02	0.11	PbTe	Yes
C	15.11	0.15110	16.55	0.12	C	Yes
Total			100.00			

Table S2 The effects of ZnO NPs exposure on the number of sperm cells in the male rat seminiferous tubules at VII stage

Test Items	ZnO NPs (mg/kg bw/d)			
	0	7	50	350
Spermatogonia/tube	2.27±0.46	2.24±0.77	1.87±0.64	2.20±0.68
Preleptotene primary spermatocytes/tube	56.99±3.59	58.74±4.46	57.63±4.39	54.87±4.60
Pachytene primary spermatocytes/tube	67.04±4.12	66.27±5.21	66.37±4.61	66.47±3.10
Round sperm cells/tube	210.68±10.44	210.92±8.62	211.98±6.90	213.24±7.88
Spermatogonia/100 supporting cells	10.72±1.90	10.55±3.37	9.20±3.17	10.74±3.14
Preleptotene primary spermatocytes/100 supporting cells	271.53±30.70	283.77±30.75	283.71±21.29	269.71±27.61
Pachytene primary spermatocytes/100 supporting cells	318.86±30.22	320.99±33.14	326.87±24.31	326.78±24.49
Round sperm cells/100 supporting cells	1002.13±83.85	1020.97±75.28	1044.98±64.62	1047.57±61.36

The data are mean ± SD in the table.

Table S3 DLS determination results of ZnO NPs in cell culture medium

Concentration	1 day		7 days	
	Particle (nm)	Zeta potential	Particle (nm)	Zeta potential
20 µg/mL	243.3±44.1	-5.8±0.8	267.1±48.1	-6.2±2.9

The data are mean ± SD in the table.

Table S4 Effects of ZnO NPs on cell nuclear parameters

Concentration (µg/mL)	24 h		48 h	
	P2A	LWR	P2A	LWR
0	1.23±0.16	1.38±0.45	1.23±0.13	1.36±0.34
0.2	1.23±0.16	1.38±0.47	1.22±0.13	1.36±0.36
0.5	1.23±0.16	1.38±0.48	1.22±0.13	1.35±0.33
0.75	1.23±0.16	1.37±0.46	1.22±0.12	1.34±0.32
1	1.22±0.17	1.36±0.50	1.22±0.14	1.34±0.38
1.25	1.24±0.22	1.42±0.65	1.23±0.16	1.35±0.42

The data are mean ± SD in the table.