

## Supplementary material

### **Study on the Therapeutic Effect and Some Immune factors by Methotrexate Modified Superparamagnetic Nanoparticles in Rat Mammary Tumors**

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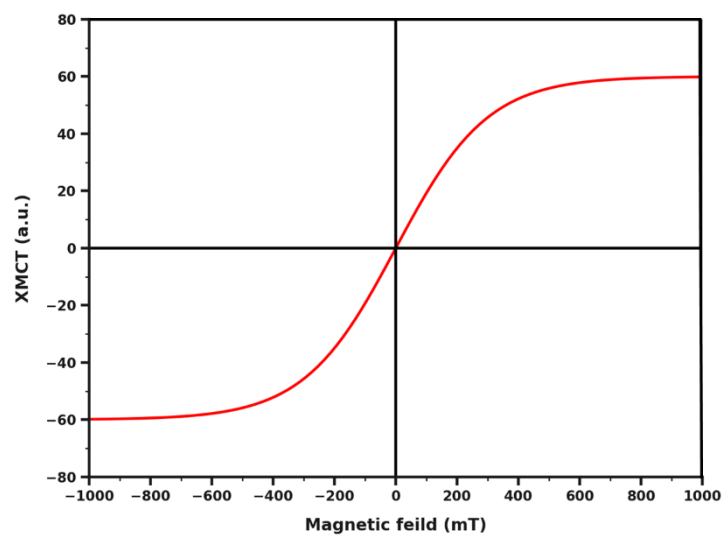
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## Materials

Ferric chloride hexahydrate ( $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$  > 99%), ferrous chloride tetrahydrate ( $\text{FeCl}_2 \cdot 4\text{H}_2\text{O}$ ), polyethylenimine (PEI, Mw: 25,000), MTX, 1-ethyl-3-[3-(dimethylamino)propyl] carbodiimide (EDC), N-hydroxysuccinimide (NHS) and ammonium hydroxide (25 wt.%  $\text{NH}_3$  in water) obtained from Sigma.



**Figure S1** The magnetization curve of MTX-modified magnetic nanoparticles.

**Table S1. Grouping Information for 201 Rats**

Groups	Quantity	Magnetic Particles	Whether to enter the magnetic field	Number of times for entering the magnetic field	Thermotherapy temperature (°C)	Thermotherapy time(min)
47TC	24	MTX	Yes	1	47	30
47T	24	PEI	Yes	1	47	30
42TC	21	MTX	Yes	1	42	60
42T	21	PEI	Yes	1	42	60
Repeated 42TC	12	MTX	Yes	2-5 times, interval of 72 h	42	60
Repeated 42T	12	PEI	Yes	2-5 times, interval of 72 h	42	60
MFC	16	MTX	No	0		
C	21	None	No	0		
M	12	None	Yes	1		30
MF	12	PEI	No	0		
MTX	16	20 mg/kg free MTX	No	0		
Normal	10	None	No	0		