Supporting information

Decomposable STING nanoagonist-amplified oncolytic virotherapy through remodeling the immunosuppressive microenvironment of triple-negative breast cancer

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Fig. S1 WB analysis of STING pathway after treated with different dose of OVs for 24 h.



Fig. S2 WB analysis of STING pathway after treated with OV, Mn^{2+} , $OV+Mn^{2+}$ for 24 h.



Fig. S3. The size distribution of OV, OV-MnO₂, and OV-MnO₂/HE nanoagonist by DLS.



Fig. S4. The stability of OV-MnO_2 and OV-MnO_2/HE at 4 $^{\circ}\mathrm{C}.$



Fig. S5. Degradation behavior of OV-MnO₂ with GSH. (A) Absorption spectra of OV-MnO₂ treated with GSH at different time. (B) The degradation behavior of OV-MnO₂ dispersed water with GSH determined by the absorbance of MnO_2 at 400 nm.



Fig. S6. FTIR spectra of OV-MnO $_2$ and OV-MnO $_2/HE.$



Fig. S7. OVs release profile of OV- MnO_2/HE in PBS buffer (pH 7.4) with 10 mM GSH.



Fig. S8 Optical imaging of OV and OV-MnO_2/HE infected-4T1 cells for 48 h. Scale bar: 100 μm



Fig. S9. The fluorescence imaging of 4T1 cells staining with H2DFCDA after different treatments. G1: NS; G2: OV; G3: MnO_2 ; G4: OV- MnO_2 ; G5: OV- MnO_2 /HE. Scale bar: 100 μ m



Fig. S10. (A) Absorption spectra of OV- MnO_2/HE treated pH 6.5 buffer with H_2O_2 . (B) The O₂ generation ability of OV- MnO_2/HE treated pH 6.5 buffer with H_2O_2 .



Fig. S11. The O₂ generation of 4T1 cells via [Ru(ddp)3]Cl₂ after various treatment.G1: NS; G2: OV; G3: MnO₂; G4: OV-MnO₂; G5: OV-MnO₂/HE. Scale bar: 100 μm



Figure S12. WB analysis for the expressions of proteins in cell apoptosis pathway in 4T1 cells. G1: NS; G2: OV; G3: MnO₂; G4: OV-MnO₂; G5: OV-MnO₂/HE.



Fig. S13 The photos of 3D tumor spheroids after different treatments for 48 h. G1: NS; G2: OV; G3: MnO₂; G4: OV-MnO₂; G5: OV-MnO₂/HE



Fig. S14 WB and quantitative analysis of STING pathway proteins after incubation of 4T1/DC2.4 co-culture system with OV, MnO₂, OV-MnO₂, and OV-MnO₂/HE. G1: NS; G2: OV; G3: MnO₂; G4: OV-MnO₂; G5: OV-MnO₂/HE



Fig. S15. Neutralization of OV and OV- MnO_2/HE in 4T1 cells was performed using anti-Ad5 serum at the specified concentrations. Scale bar: 100 μ m



Fig. S16 The photographs (A) and weight (B) of the tumors in each group after treatment with different formulations. G1: NS; G2: OV; G3: MnO₂; G4: OV-MnO₂; G5: OV-MnO₂/HE



Fig. S17. The semi-quantitative fluorescence of TUNEL data.



Fig. S18 Relative body weights of the mice during the treatment. G1: NS; G2: OV; G3: MnO₂; G4: OV-MnO₂; G5: OV-MnO₂/HE



Fig. S19. HE staining of Heart, Liver, Spleen, Lung, and Kidney. G1: NS; G2: OV;G3: MnO₂; G4: OV-MnO₂; G5: OV-MnO₂/HE. Scale bar: 50 μm



Fig. S20. Blood biochemical values of blood aspartate aminotransferase (AST), ALT, ALP, CRE, BUN, and RBC at 21 d post-injection (n = 5).



Fig. S21. The hemolysis of OV-MnO $_2$ /HE at varied concentrations.