

Supplementary Information

A study of Saponin-Encapsulated Ultrasound Microbubbles

Rb₃NPs@MBs for Atherosclerosis Targeted Treatment

Chunting Zhong ^a, Jianhua Bai ^a, Xiaoting Yang ^a, Yiran Ji ^a, Xiao Tan ^a, Jiabao Huang ^a, Xiaoyu Chen ^a, Bingxuan Xu ^a, Dianhuan Tan ^a, Yun Chen ^{a,*}, Tingting Zheng ^{a,*}

^a Shenzhen Key Laboratory for Drug Addiction and Medication Safety, Department of Ultrasound, Institute of Ultrasonic Medicine, Peking University Shenzhen Hospital, Shenzhen Peking University-The Hong Kong University of Science and Technology Medical Center, Shenzhen 518036, Guangdong, P. R. China.

*Corresponding authors: Yun Chen is yunchen@sphmc.org; Tingting Zheng is kzys_018@126.com

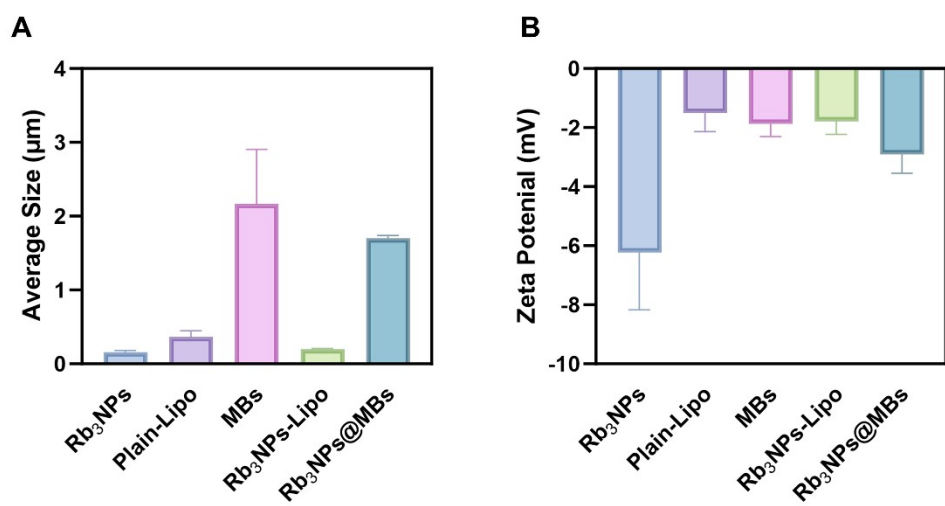


Fig.S1 Comparison of properties of different particles. **A** Comparison of particle sizes of different particles; **B** Comparison of Zeta potentials of different particles.

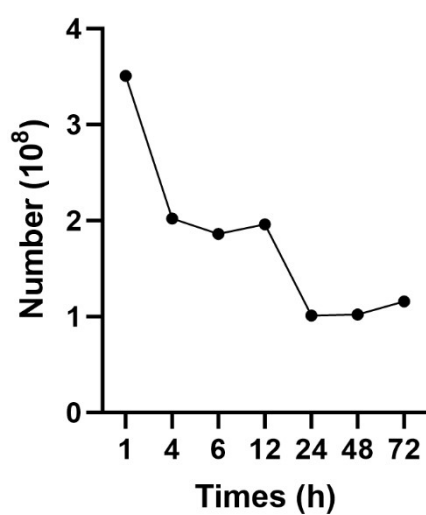


Fig.S2 Microbubble stability in 72h.

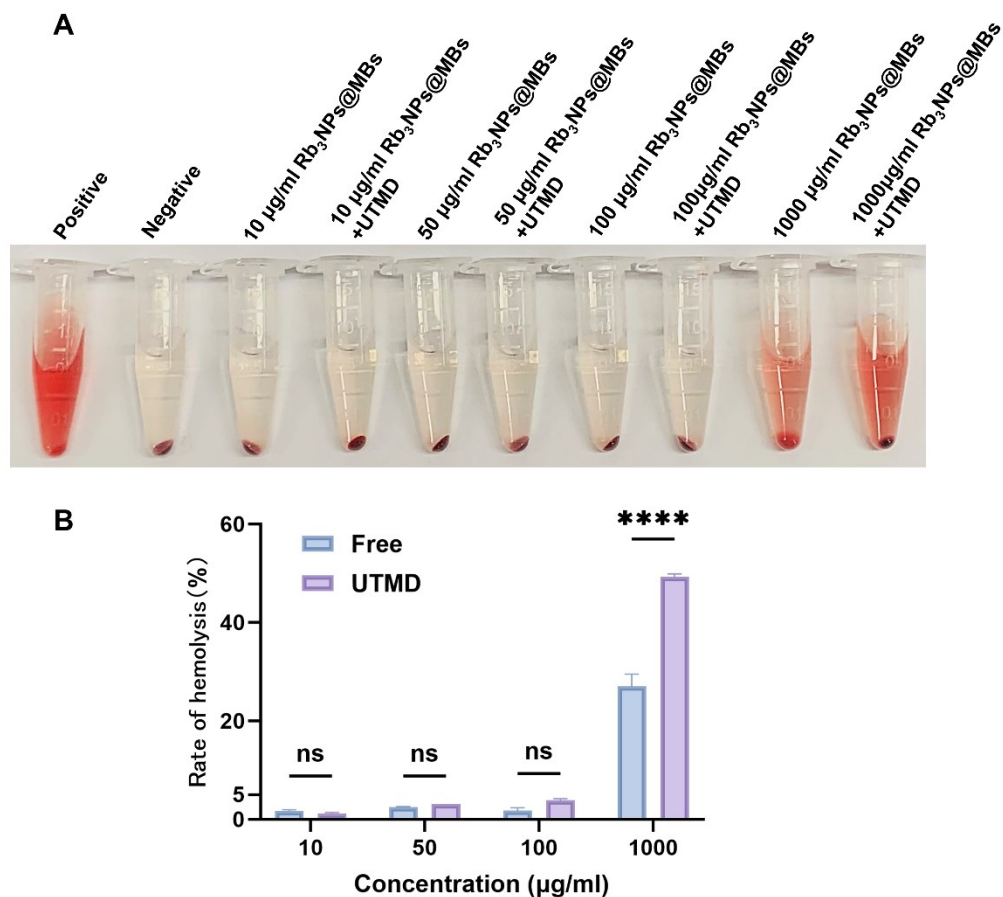


Fig.S3 Hemolysis experiments of $\text{Rb}_3\text{NPs@MBs}$. A) Comparison of $\text{Rb}_3\text{NPs@MBs}$ hemolysis supernatant with different concentrations and treatments; B) Statistical analysis plot of the OD values of the supernatant of each group at the specific wavelength of 541 nm of hemoglobin measured by microplate reader in panel A. Free: no ultrasound operation group; UTMD: ultrasound-operated group.

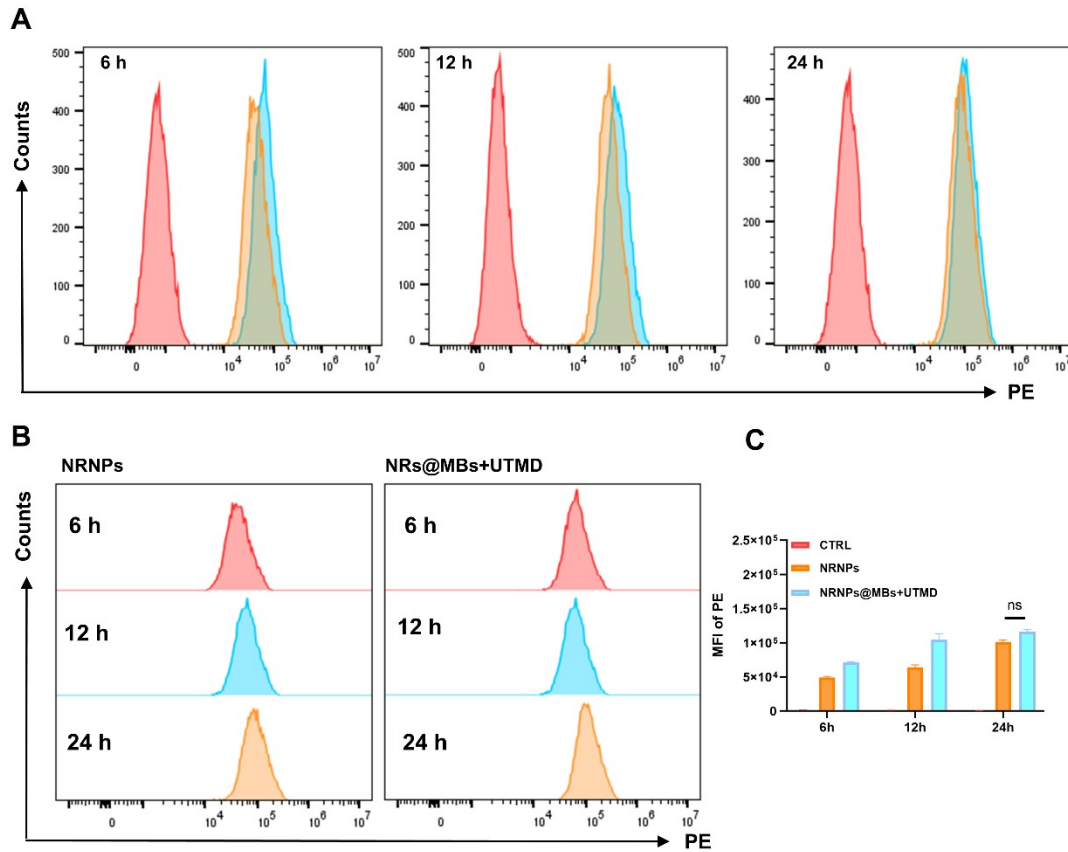


Fig.S4 Flow cytometry was used to detect the cell uptake of RAW264.7 cells for 24 h. A) Fluorescence flow diagram of RAW264.7 cells at 6 h, 12 h, and 24 h, and the grouping color corresponds to the legend color in figure C; B) Comparison of fluorescence flow images of RAW264.7 cells with NRNPs group and NRNPs@MBs+UTMD group at different time points; C) Quantitative analysis of flow fluorescence intensity in panel A (n=3).

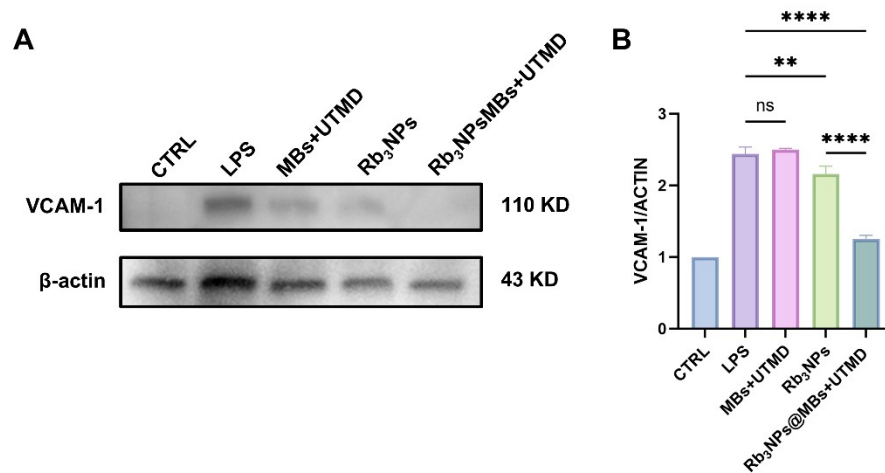


Fig.S5 Western Blot detected the expression of VCAM-1 protein in HUVEC cells. A) Western Blot images; B) Semi-quantitative analysis of Panel A (n=3).

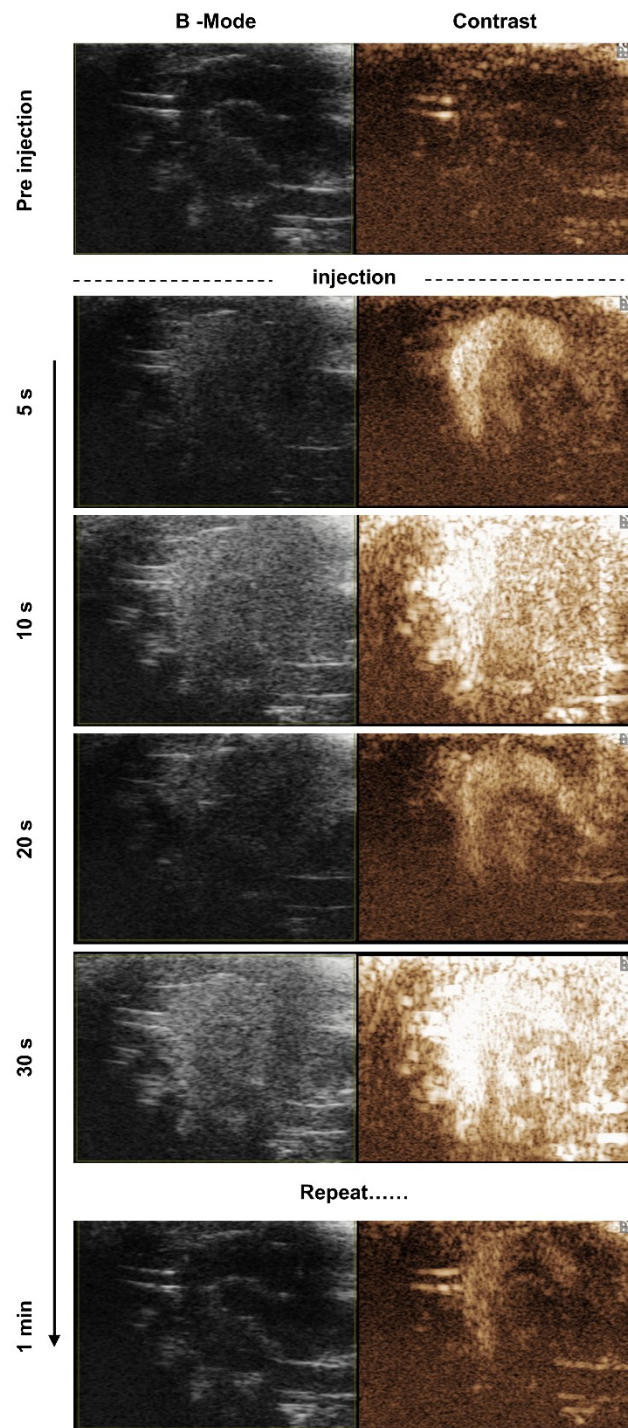


Fig.S6 The in vivo UTMD process of Rb₃NPs@MBs.

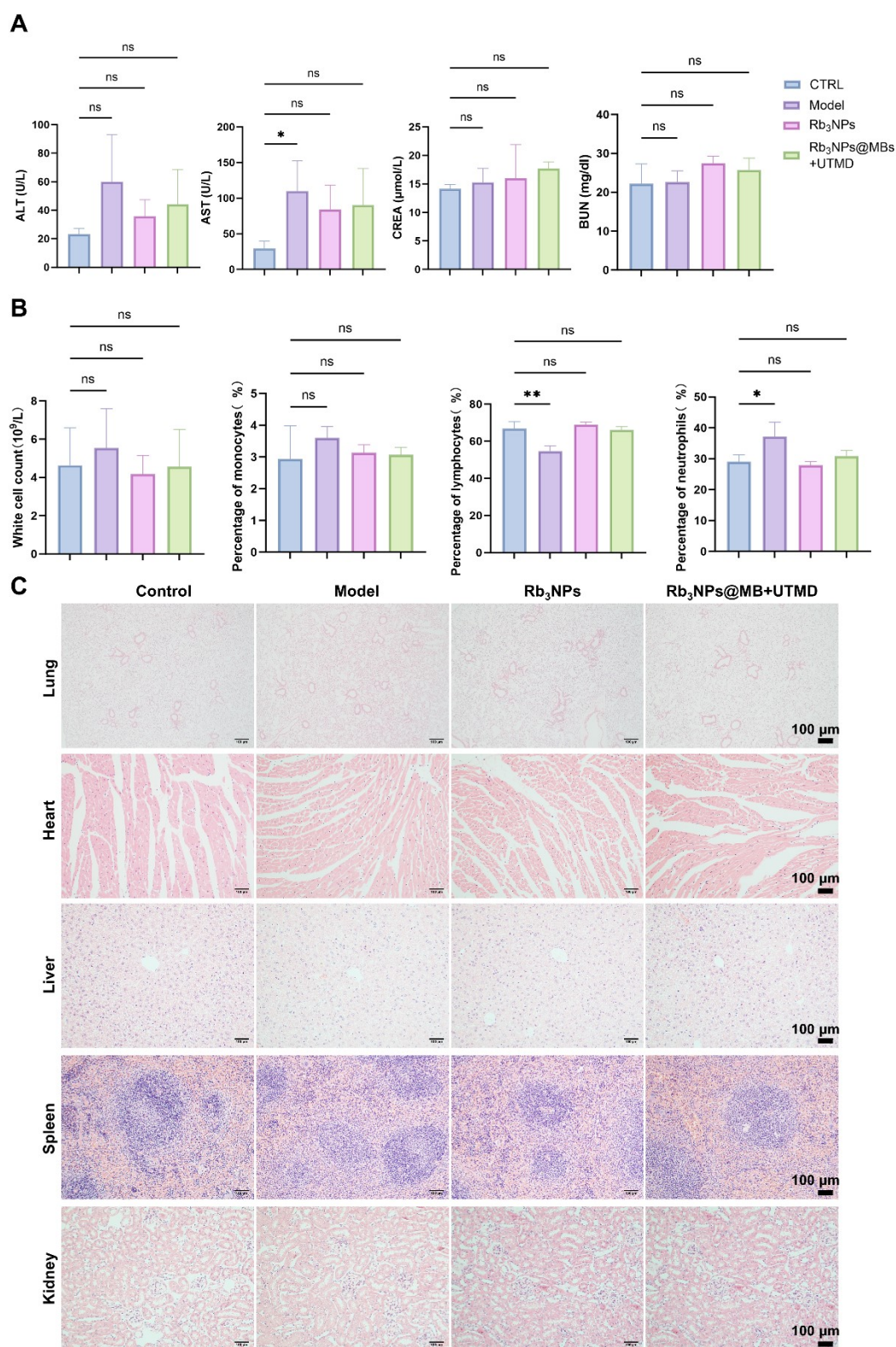


Fig.S7 Safety assessment (n=4 biologically independent mice). A) HE-stained sections of organs tissues from different groups of mice after 2w of treatment. B) Blood cell count. C) Markers of hepatic function (AST, ALT) and renal function (CREA, BUN) in different groups of mice.

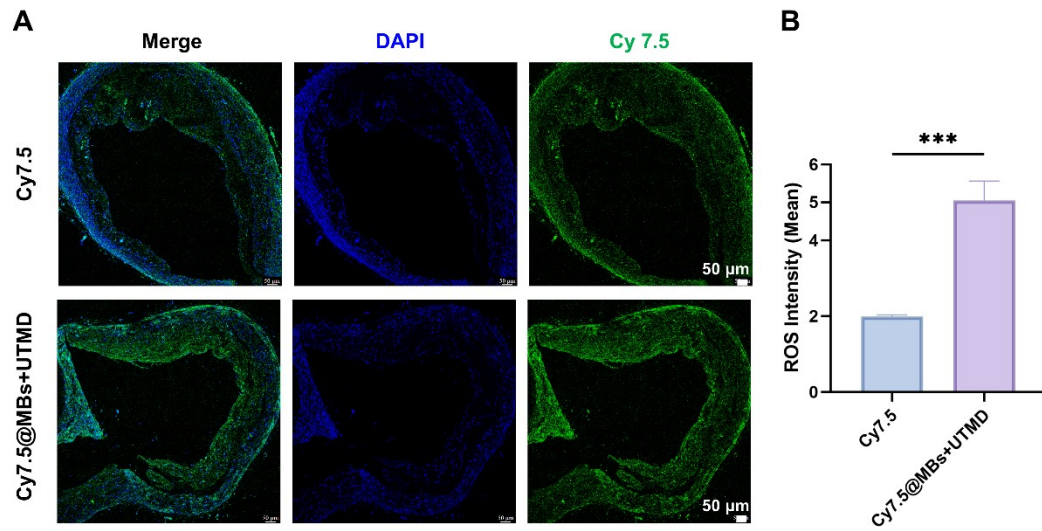


Fig.S8 Fluorescently stained sections with active arch. A) Cy7.5 fluorescence images of the aortic arch of Cy7.5 and Cy7.5@MBs+UTMD treated mice; B) Quantitative analysis of Panel A (n=3).