Supplementary Information

An inflammation-targeted nanoparticle with bacteria forced release of polymyxin B for pneumonia therapy

Peisen Zhang[a,#], Qiuhong Ouyang[a,#], Tianshu Zhai[b,#], Jing Sun[c], Jun Wu[a], Feng Qin[d], Ni Zhang[d], Saisai Yue[a], Xinchen Yang[a], Hanyi Zhang[a], Yi Hou[a], Li Deng[a], Fang Wang[a], Qingyuan Zhan[b]，Qingsong Yu[a*], Meng Qin[a*], and Zhihua Gan[a]*

[a]Beijing Advanced Innovation Centre for Soft Matter Science and Engineering, College of Life Science and Technology, Beijing University of Chemical Technology, Beijing 100029, PR China

[b]Department of Pulmonary and Critical Care Medicine, Center of Respiratory Medicine, China-Japan Friendship Hospital, Beijing 100029, PR China

[c]Institute of Chinese Materia Medica, China Academy of Chinese Medical Sciences, Chaoyang District, Beijing 100029, PR China

[d]National Chengdu Center for Safety Evaluation of Drugs, State Key Laboratory of Biotherapy/Collaborative Innovation Center for Biotherapy, Department of Psychiatry, West China Hospital of Sichuan University, Chengdu, Sichuan, 610041, PR China
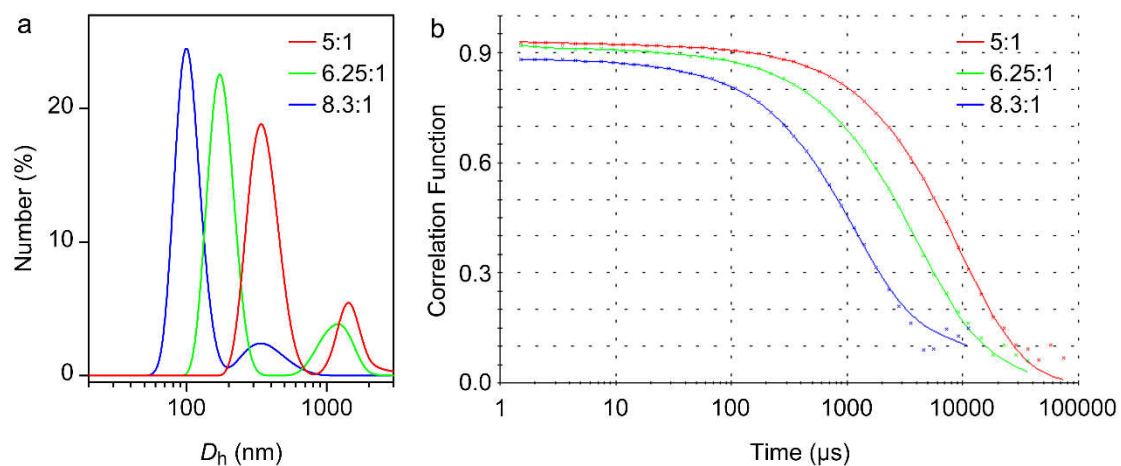
Fig. S1 (a) Number-weighted DLS and (b) the self correlation functions of the PMB-HA with different HA to PMB molar ratio.
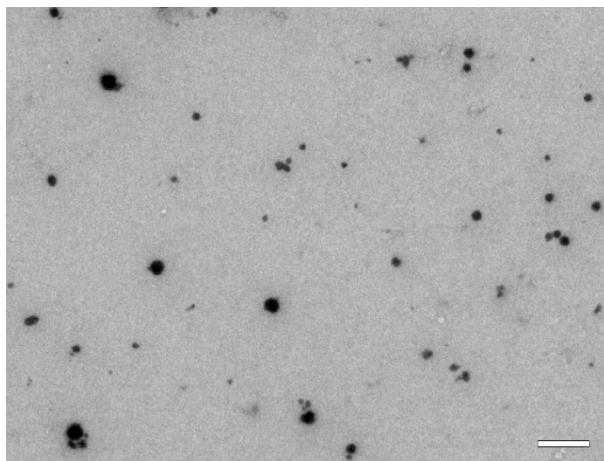

Fig. S2 TEM images of PMB-HA nanoparticles with the molar ratios of HA to PMB of 8.3:1. The embedded scale bar corresponds to 500 nm.
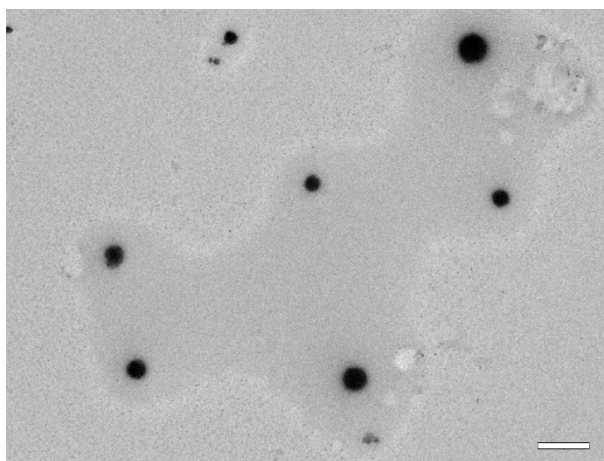

Fig. S3 TEM images of PMB-HA nanoparticles with the molar ratios of HA to PMB of 5:1. The embedded scale bar corresponds to 500 nm.
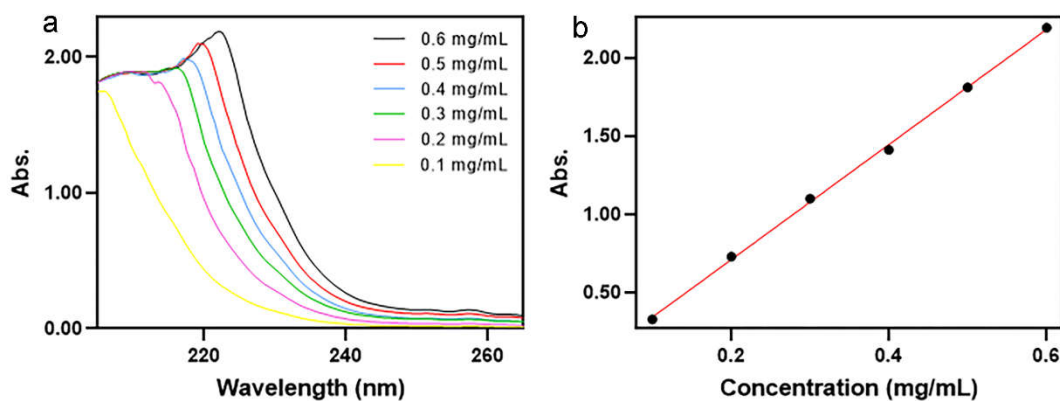
Fig. S4 Absorption spectra of 1× PBS buffer solutions containing different concentrations of PMB (left) and a standard curve at 222 nm wavelength (right).
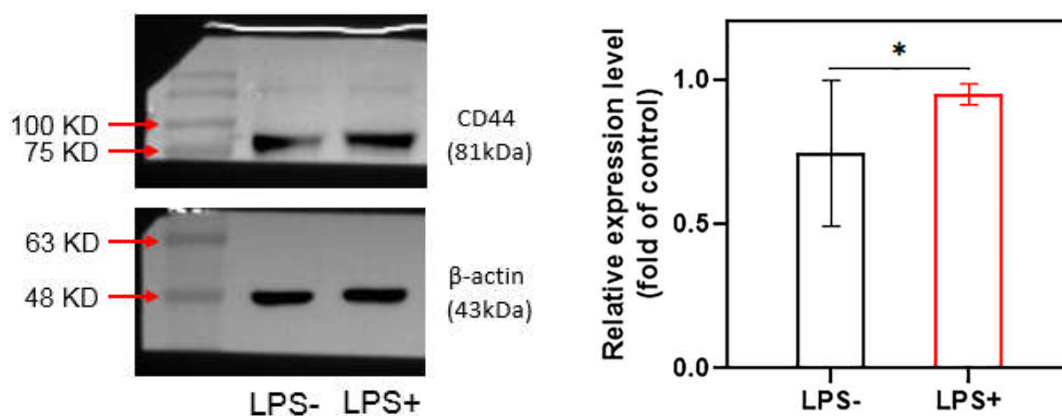

Fig. S5 Raw data of the western blot analysis of the CD44 proteins expressin on HUVECs (left) and quantitatively statistical analysis (right).
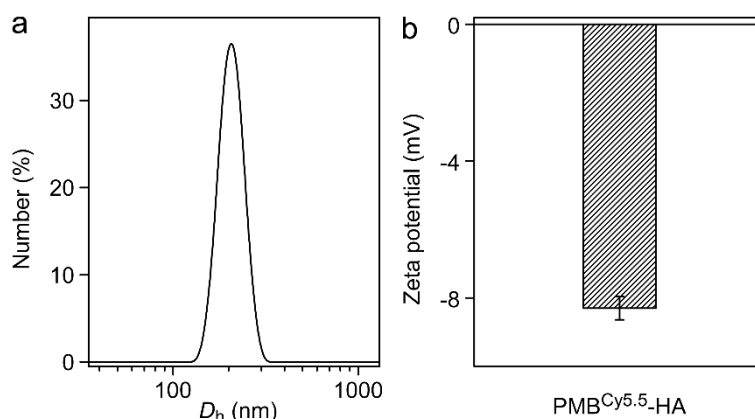

Fig. S6 (a) DLS and (b) zeta potential of the PMB$^{Cy5.5}$-HA nanoparticles. As a result, the PMB$^{Cy5.5}$-HA nanoparticles with 207.4 nm of D_h and -8.3 mV of surface charge are similar with the original PMB-HA nanoparticles.
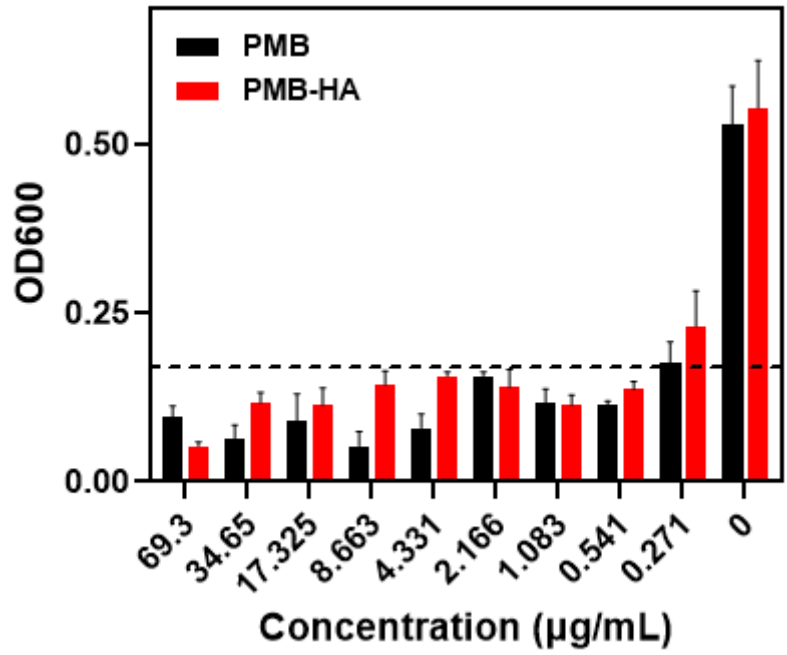
Fig. S7 The *in vitro* antibacterial effect of PMB-HA nanoparticles and free PMB on *P. aeruginosa* bacterial strain.
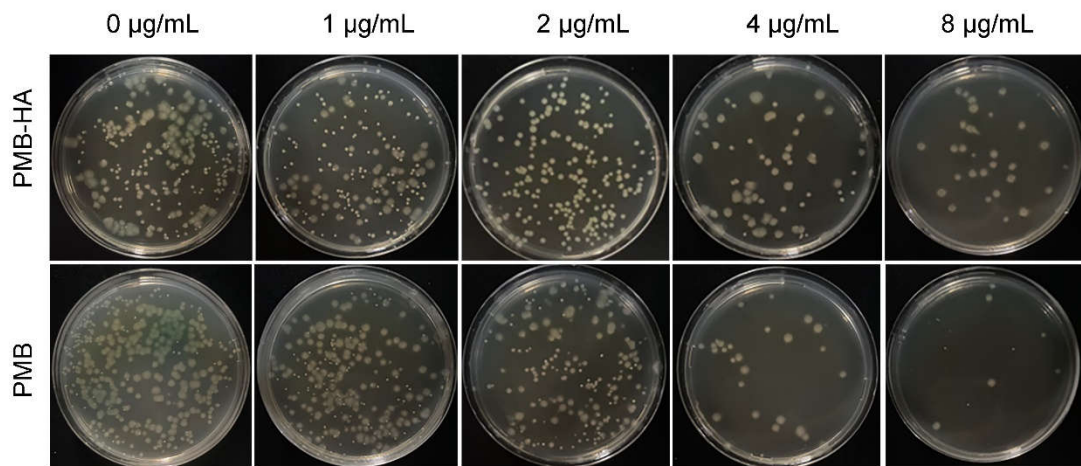

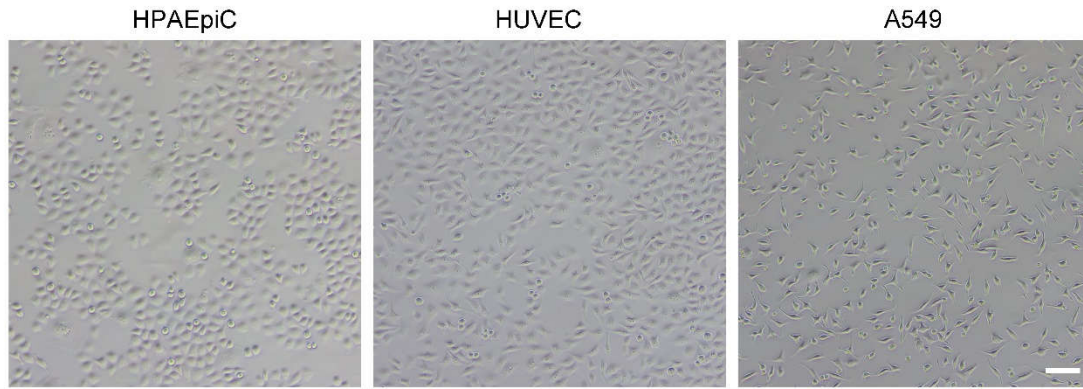Fig. S8 Representative images of bacterial colonies after treatment of PMB-HA nanoparticles or free PMB.

Fig. S9 The morphology of HPAEpiC, HUVEC, and A549 cells observed with the optical microscopy. The embedded scale bar corresponds to 100 μm.
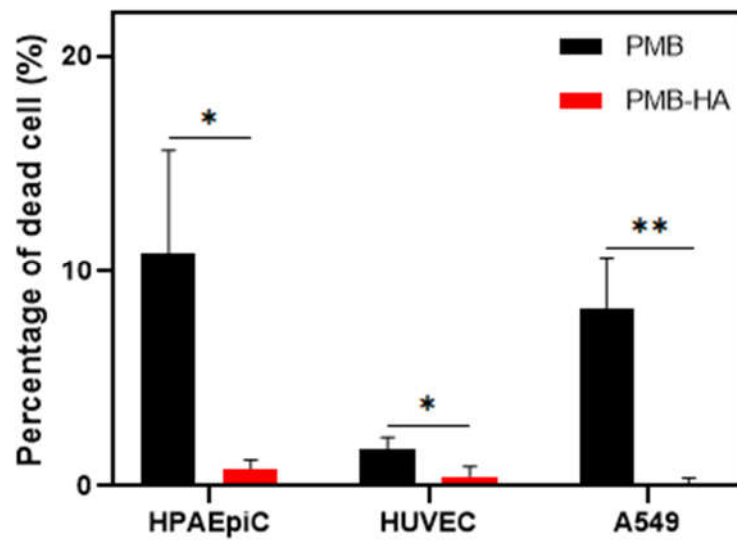

Fig. S10 The percentages of dead cells after coincubating with PMB or PMB-HA nanoparticles in the same conditions. Statistical significances were determined by one-way ANOVA (*$p < 0.05$; ** $p < 0.01$).
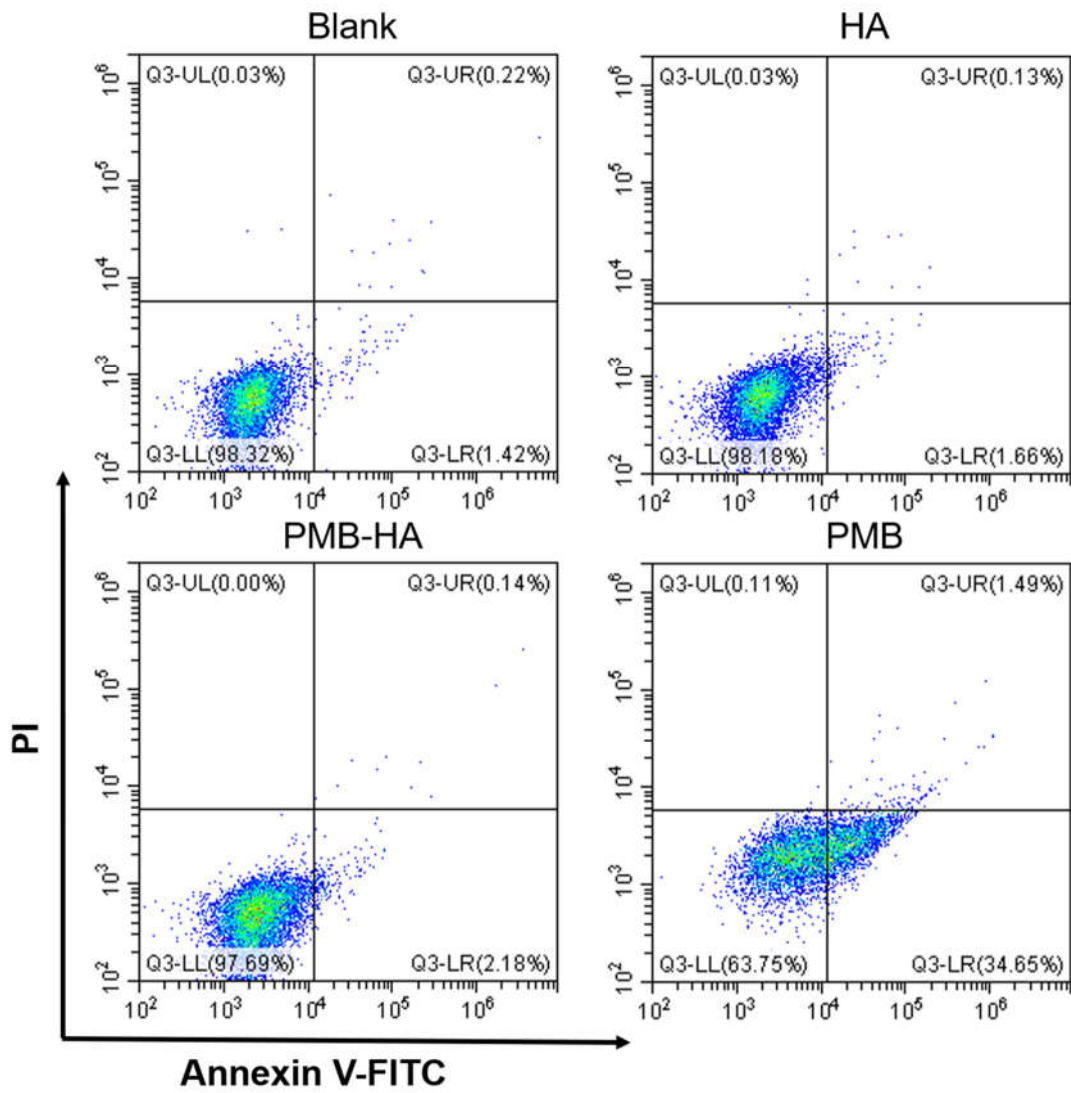
Fig. S11 The Annexin V-FITC/PI double stained flow cytometry of HPAEpiC, for showing the early apoptotic cells (Annexin V+/PI-, bottom right of each frame), and the late apoptotic cells (Annexin V+/PI+, top right of each frame).
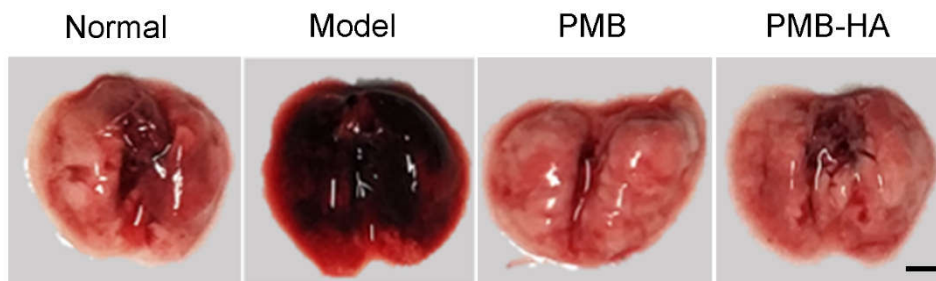

Fig. S12 The representative photographs of the lungs after receiving different treatments. The embedded scale bar corresponds to 1 mm.
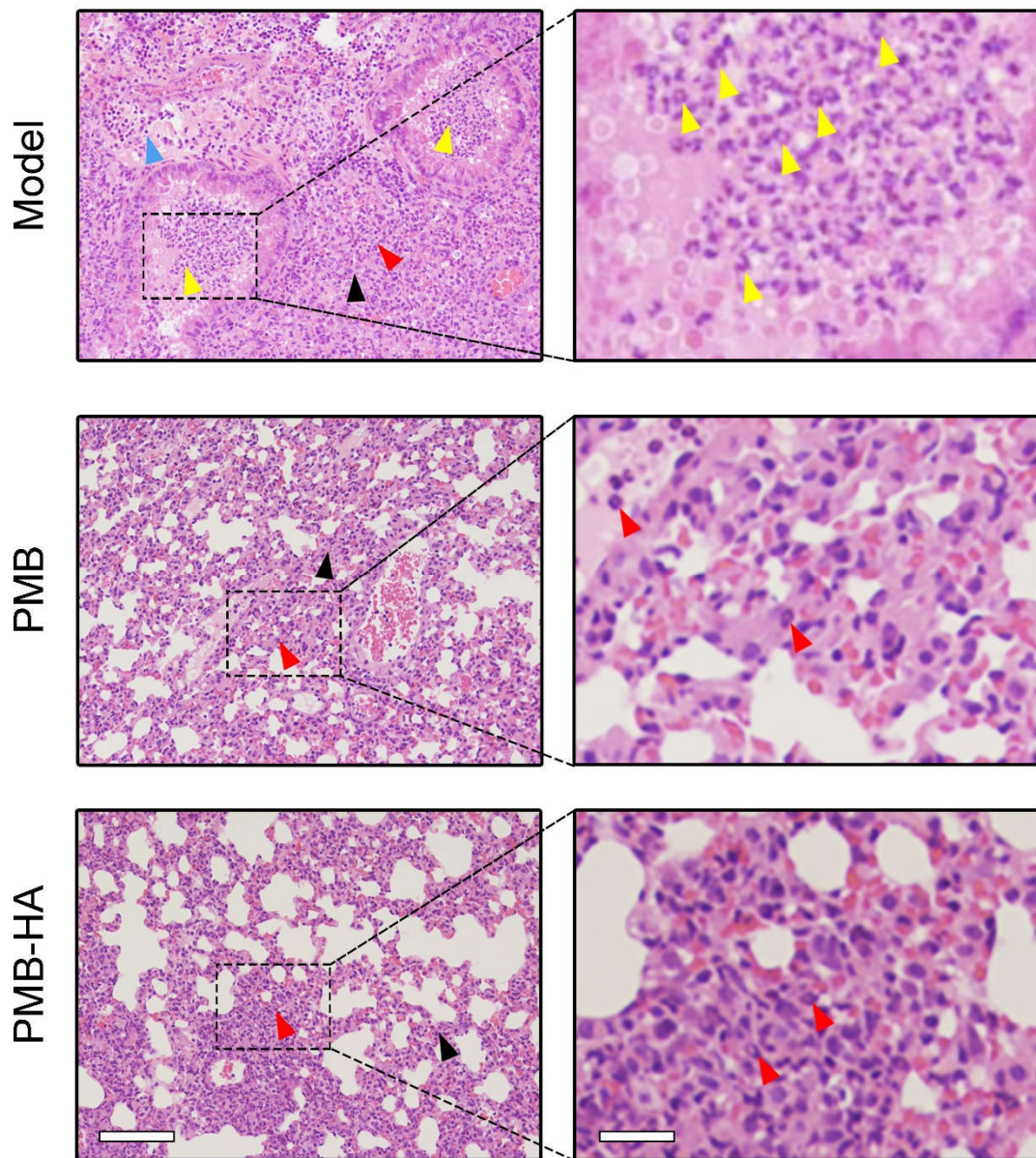
Fig. S13 H&E stained slices of lungs receiving different treatments (left), and the locally enlarged views for clearly showing the abnormal regions (right). The embedded scale bars correspond to 100 μm (left) and 25 μm (right).
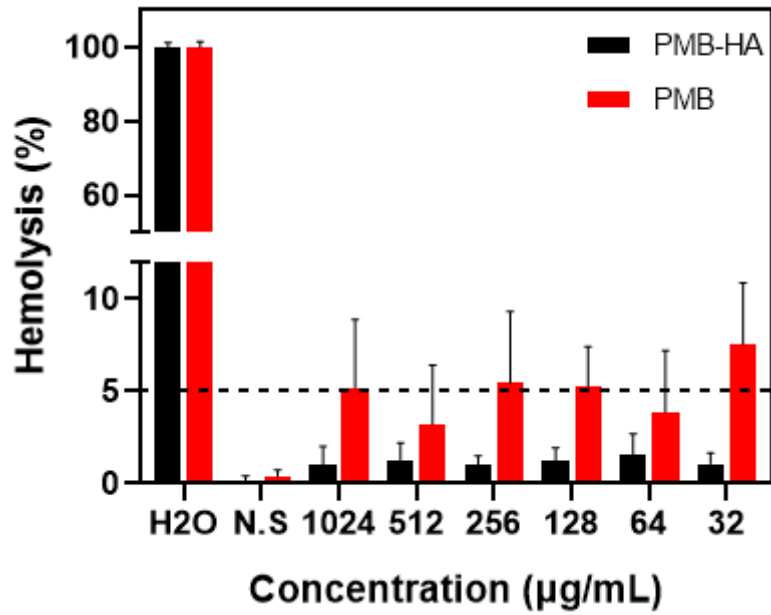
Fig. S14 The hemolysis rate of PMB-HA nanoparticles and free PMB with different concentrations.
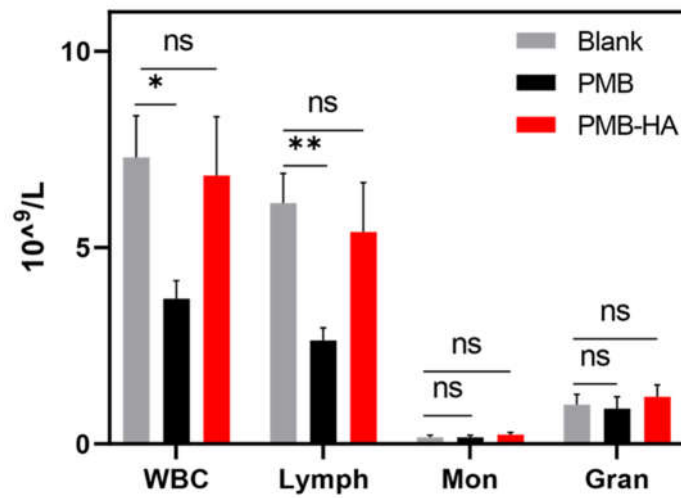

Fig. S15 The Blood routine and blood biochemical test results of mice treated with PMB-HA nanoparticles. Abbreviation: WBC, white blood cell; Lymph, lymphocyte; Mon, monocyte; Gran, granulocyte. Statistical significances were determined by one-way ANOVA (*$p < 0.05$; ** $p < 0.01$; NS: Not Statistically Significant, $p > 0.05$).
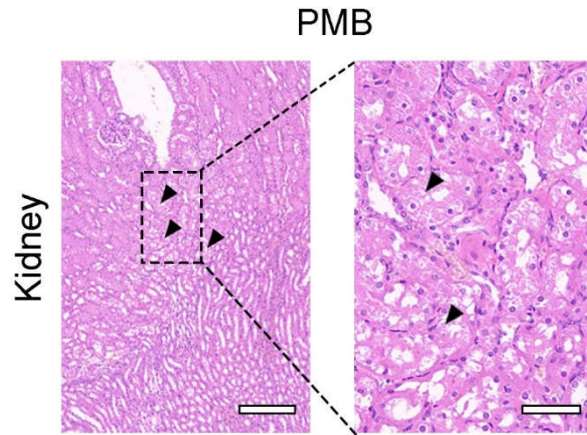
Fig. S16 H&E staining of kidney tissue slice after receiving free PMB treatment (left) and the locally enlarged view (right). The embedded scale bars correspond to 100 μm (left) and 50 μm (right).
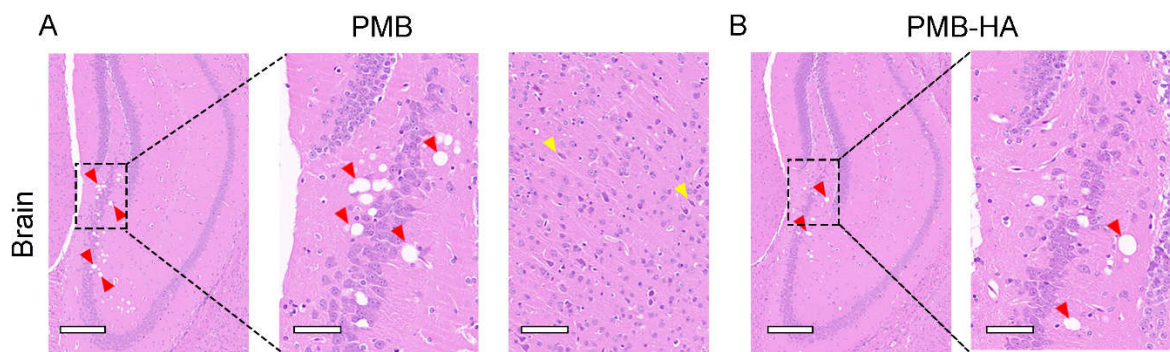

Fig. S17 (A) H&E staining of brain tissue slice after receiving free PMB treatment (left) and the locally enlarged views (middle and right). (B) H&E staining of brain tissue slice after receiving free PMB-HA treatment (left) and the locally enlarged view (right). The embedded scale bars correspond to 100 μm for the original views, and 50 μm for the enlarged views.
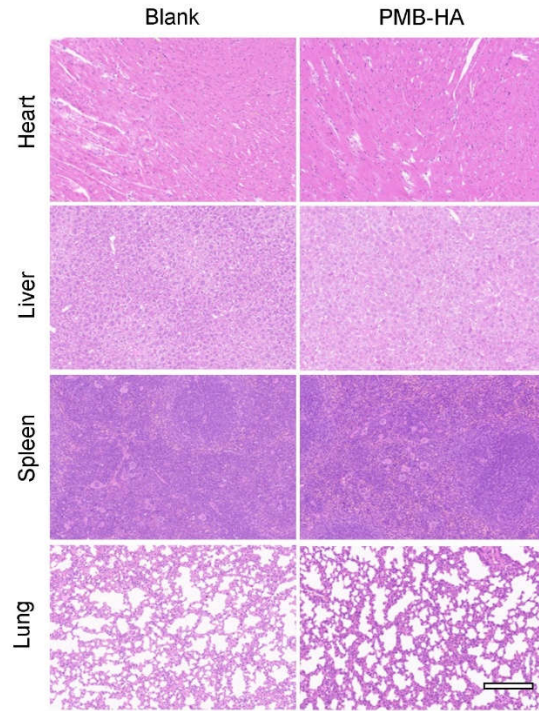
Fig. S18 H&E staining of tissue slices from major organs of mice after the long-term toxicity evaluation. The embedded scale bar corresponds to 200 μm.

Table S1 The value of the average lung index and lung index inhibition rate of each treatment group.

Group	Number of mice	Lung index (x±s)	Lung index inhibition rate (%)
Healthy	10	0.64±0.07	—
Model	10	1.02±0.02	—
PMB	10	0.86±0.11	41.87
PMB-HA	10	0.78±0.07	64.80