

## Supplementary Information

### **The immune response effect of diverse vaccine antigen attachment ways based on the self-made nanoemulsion adjuvant in the systemic MRSA infection**

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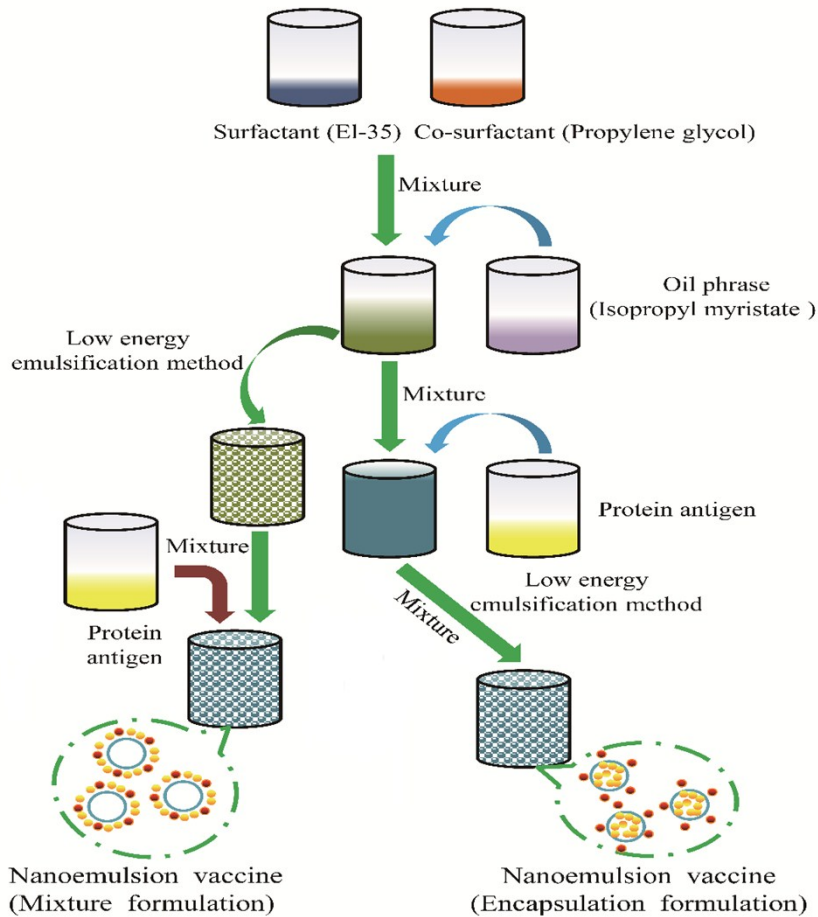
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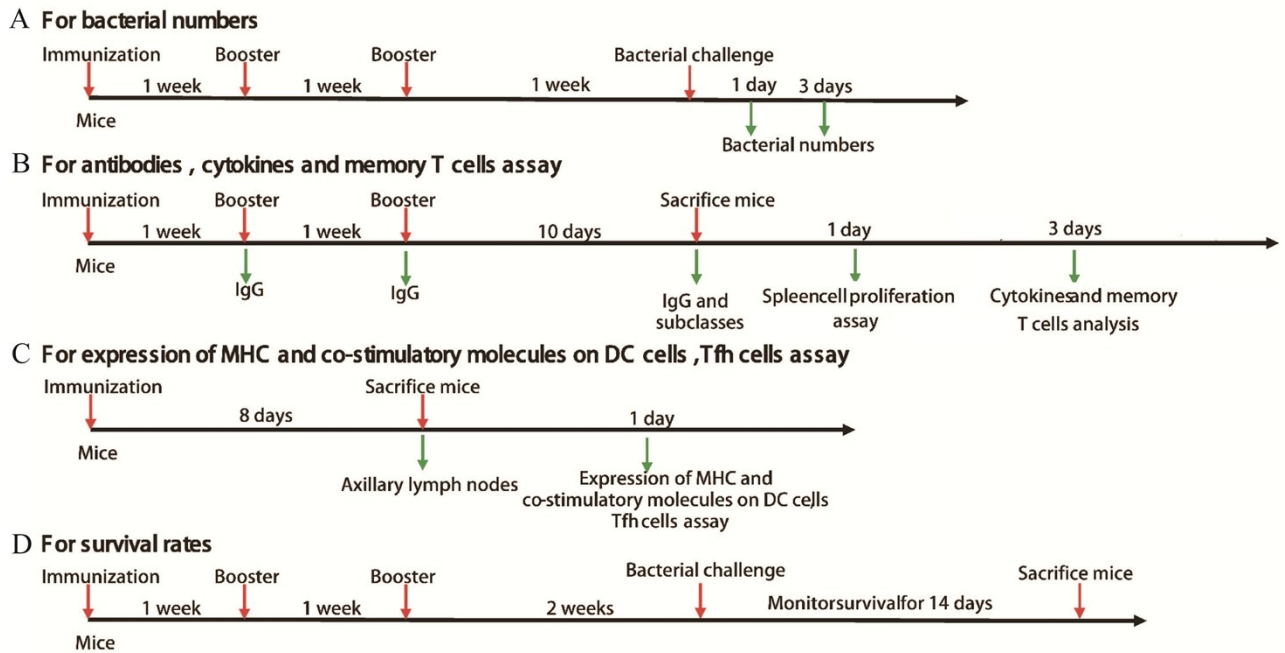
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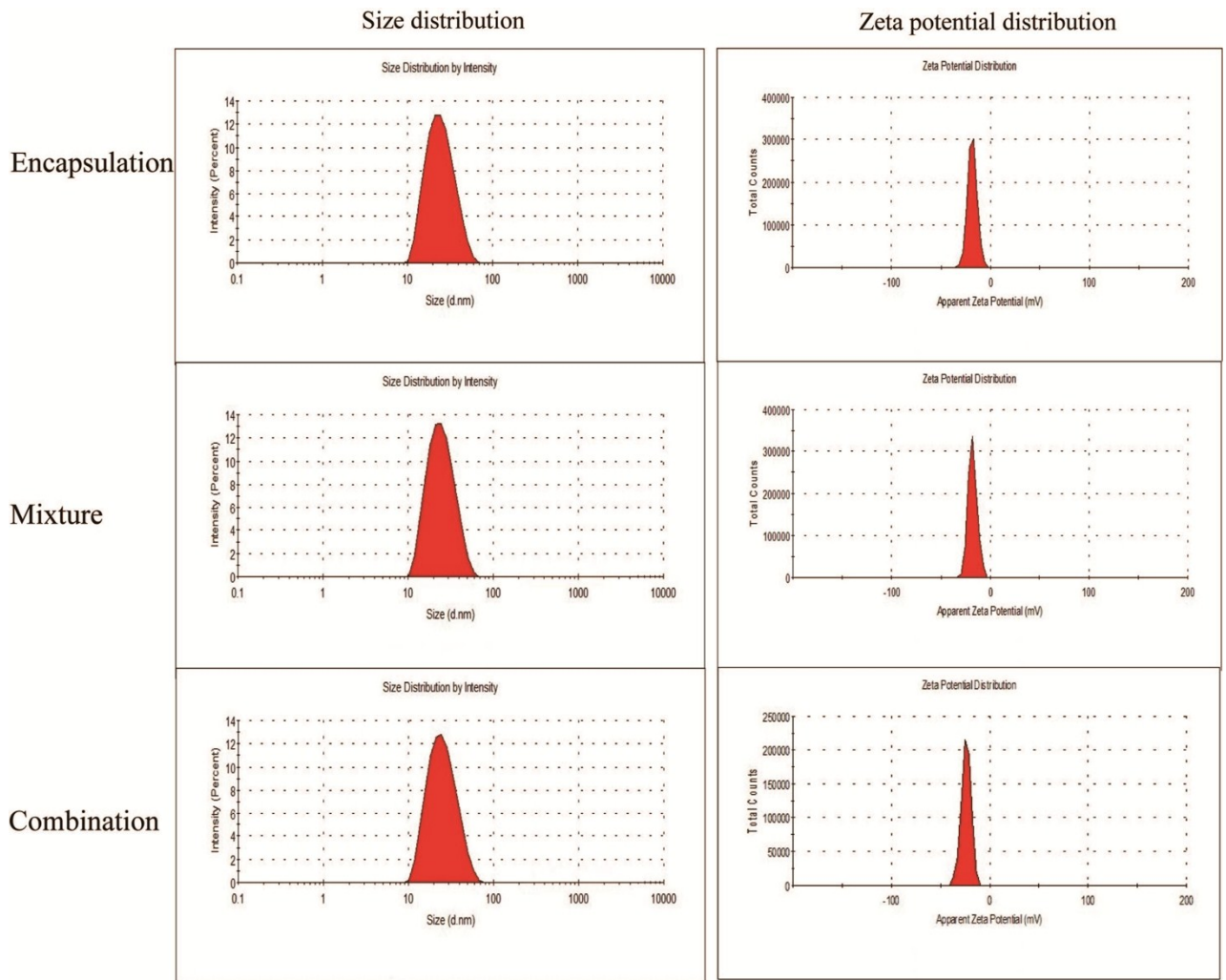
**Figure S1. Schematic illustration of three different vaccine antigen attachment ways by the low energy emulsification methods.**

The encapsulated formulation with 200 $\mu$ g/mL of the recombination antigen protein Hla<sub>H35L</sub>IsdB<sub>348-465</sub> was prepared. The mixture attachment way was stirred for 2h at 16 $^{\circ}$ C after adding the same concentration of Hla<sub>H35L</sub>IsdB<sub>348-465</sub> and the BNE. The combination attachment way was stirred for 2h at 16  $^{\circ}$ C after adding half volume encapsulation and half volume mixture way of nanoemulsion vaccine.



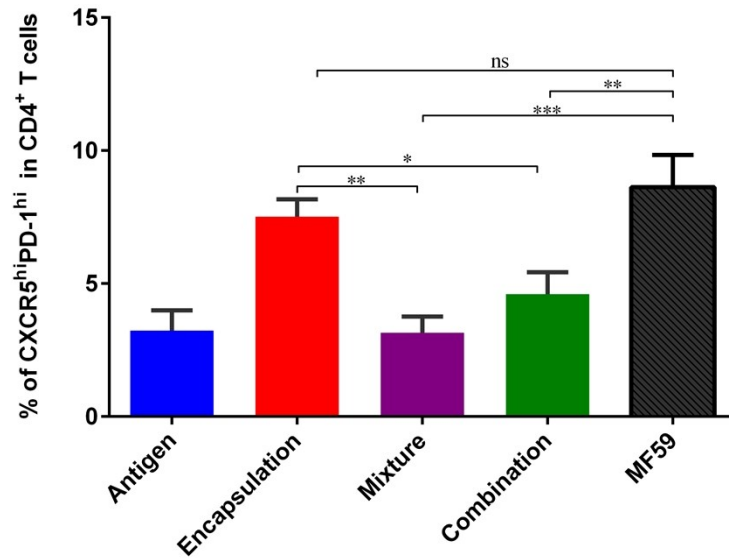
**Figure S2. The animal experiment scheme.**

Female blab/c mice (6–8 weeks old) were immunized by intramuscular injection into the upper quadriceps muscles three times on day 0, 7 and 14. (A) Bacterial burdens in organs were performed on 1 and 3 days post infection. (B) Antibody responses, spleen cell proliferation assay and memory T cell assay were performed on days 24, 25 and 27. (C) The expression of MHC and co-stimulatory molecules on DC cells and Tfh cells in draining lymph nodes were performed on 8 days after immunization. (D) Survival rates were performed on day 28 and monitored for 14 days.



**Figure S3. Size distribution and zeta potential distribution of three different nanoemulsion adjuvant vaccine (encapsulation, mixture and combination).**

The size distribution and zeta potential distribution of encapsulation, mixture and combination attachment ways.



**Figure S4. The frequency of follicular helper CD4<sup>+</sup> T cells in the draining lymph nodes of immunized mice.**

The popliteal lymph nodes of balb/c mice (n = 6) were isolated. The frequency of follicular helper CD4<sup>+</sup> T cells (CD4<sup>+</sup>CXCR5<sup>hi</sup>PD-1<sup>hi</sup>) was determined by flow cytometry.

Data are expressed as the mean  $\pm$  SD (n = 6).