Supporting Information

One-pot generating subunit vaccine with high encapsulating efficiency and fast lysosome escape for potent cellular immune response

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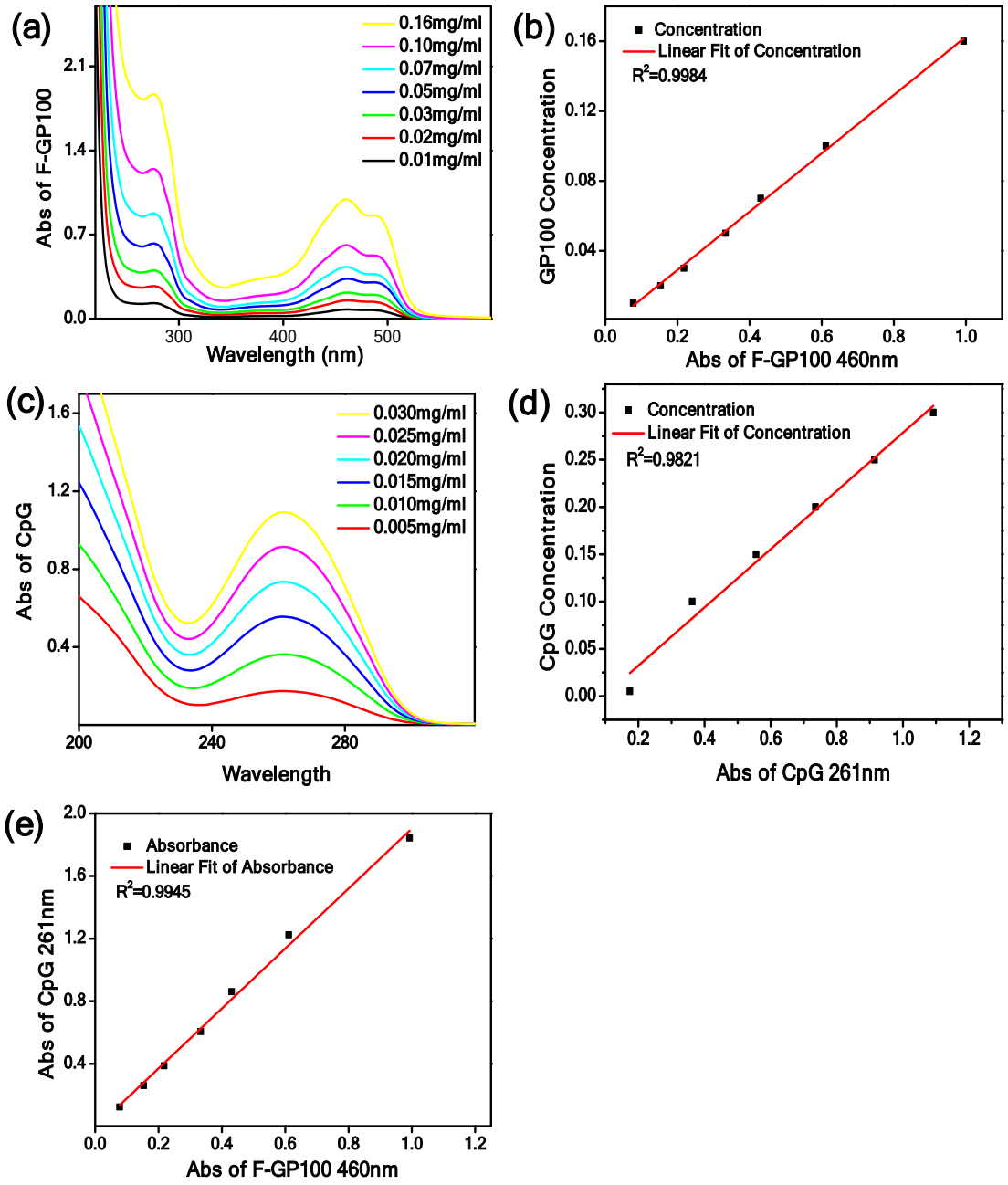


Figure S1. The absorption spectra of F-GP100 (a) and CpG (c) with different concentrations were detected by UV-vis spectrometer. The standard curve of F-GP100 at 460 nm (b) and CpG at 261 nm (d) were detected by UV-vis spectrometer. (e) The related standard curves of F-GP100 at 460 nm and 261 nm excitation wavelengths.

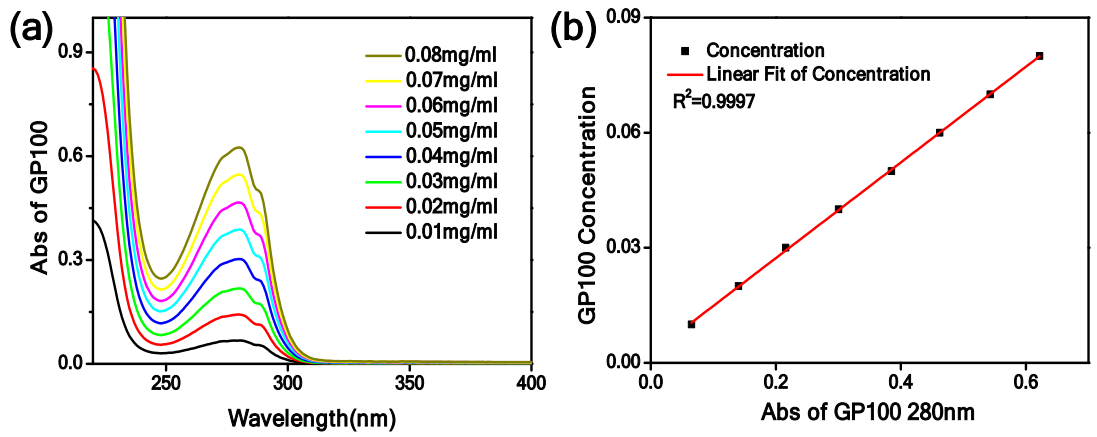


Figure S2. (a) The absorption spectra of GP100 with different concentrations was detected by UV-vis spectrometer. (b) The standard curve of GP100 at 280 nm was detected by UV-vis spectrometer.

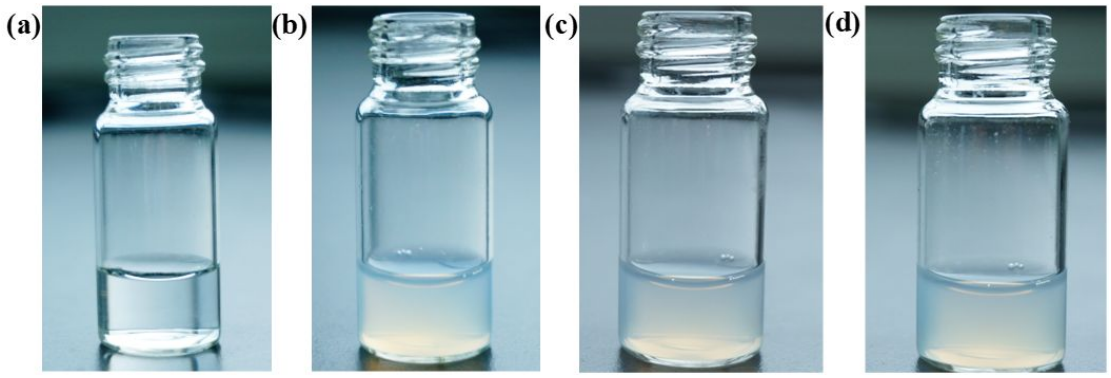


Figure S3. The representative picture of the solutions (His_6, His_6+GP100 and His_6+GP100+CpG) (a) before the addition of zinc ions, (b) after adding zinc ions to form HmA, (c) GP100@HmA and (d) (GP100+CpG)@HmA .

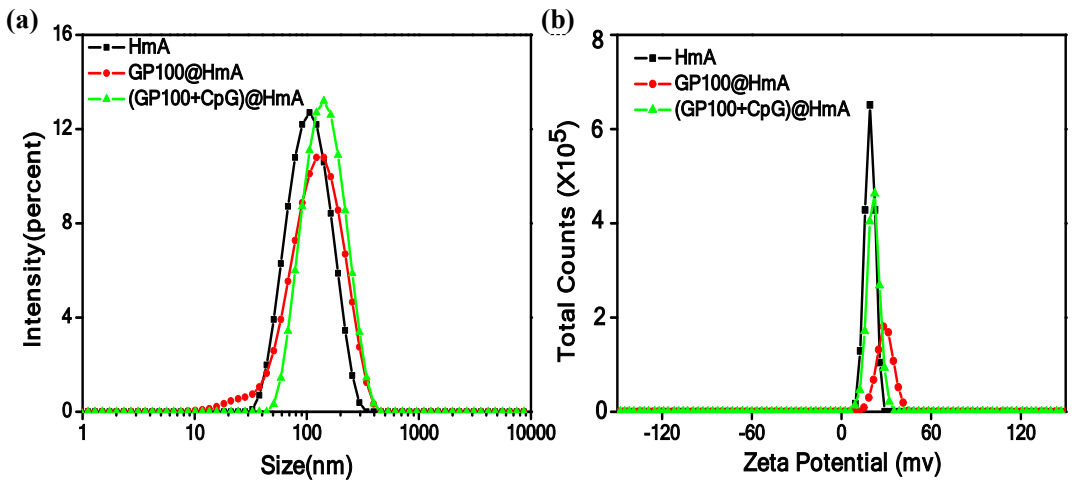


Figure S4. The original graphs of (a) size distributions and (b) zeta potential of HmA, GP100@HmA and (GP100+CpG)@HmA.

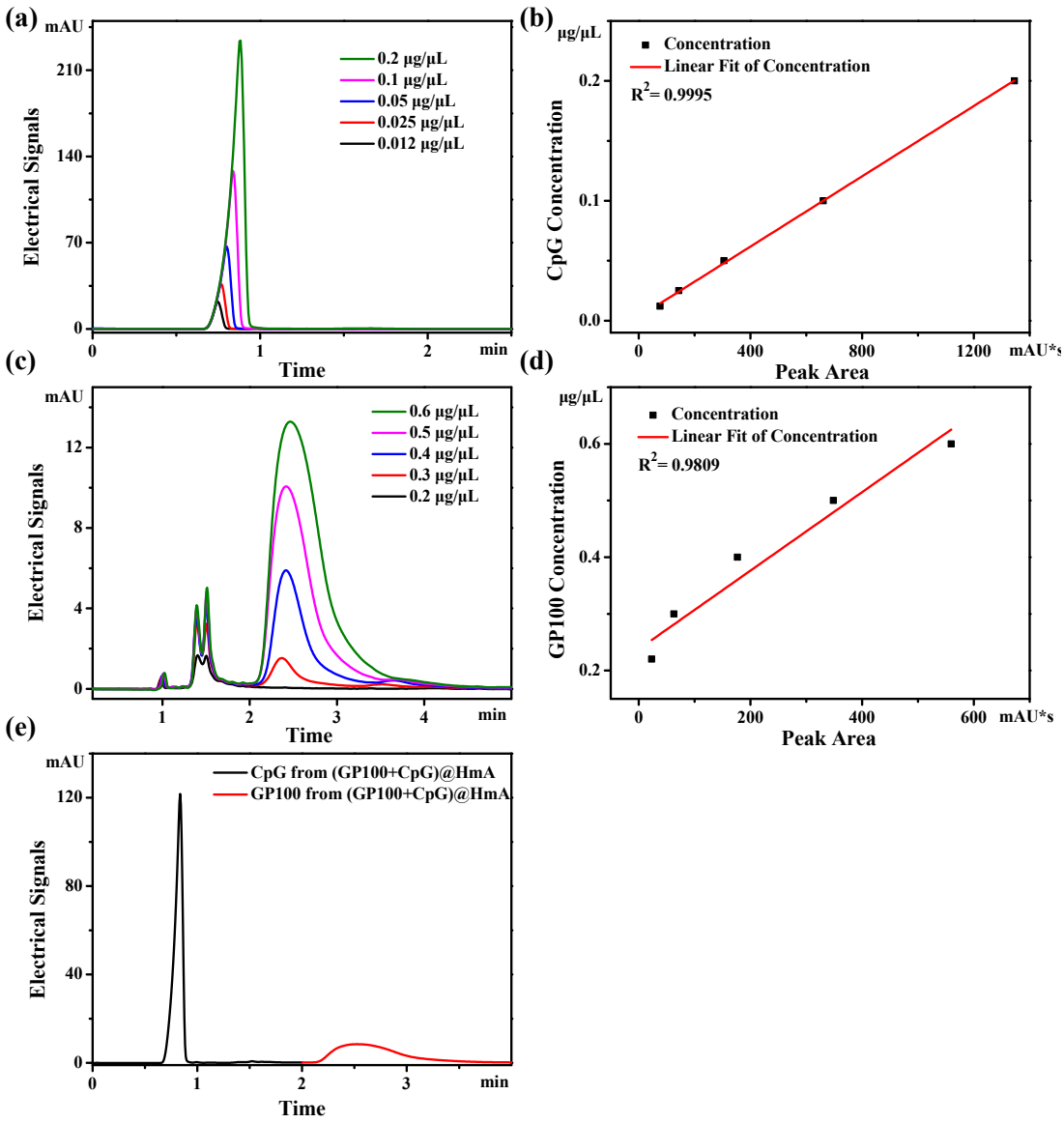
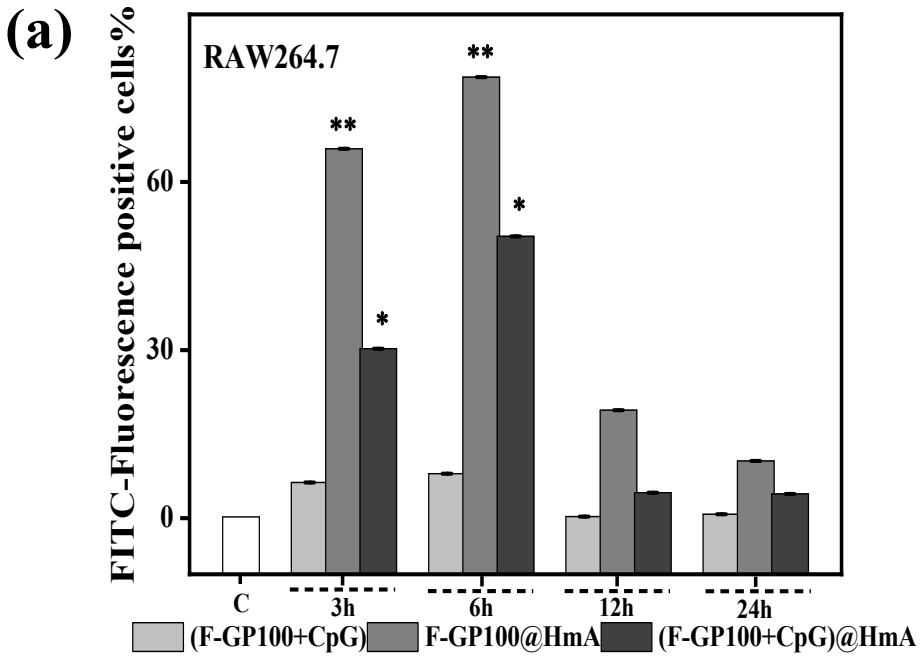


Figure S5. The absorption spectra of GP100 (a) and CpG (c) with different concentrations were detected by HPLC. The standard curve of GP100 at 280 nm (b) and CpG at 261 nm (d) were detected by HPLC. (e) High performance liquid chromatogram of nanometer vaccine preparation (GP100+CpG)@HmA.


(a)
RAW264.7
FITC-Fluorescence positive cells%
60
30
0
**
*
**
*
C
3h
6h
12h
24h
(F-GP100+CpG)
F-GP100@HmA
(F-GP100+CpG)@HmA

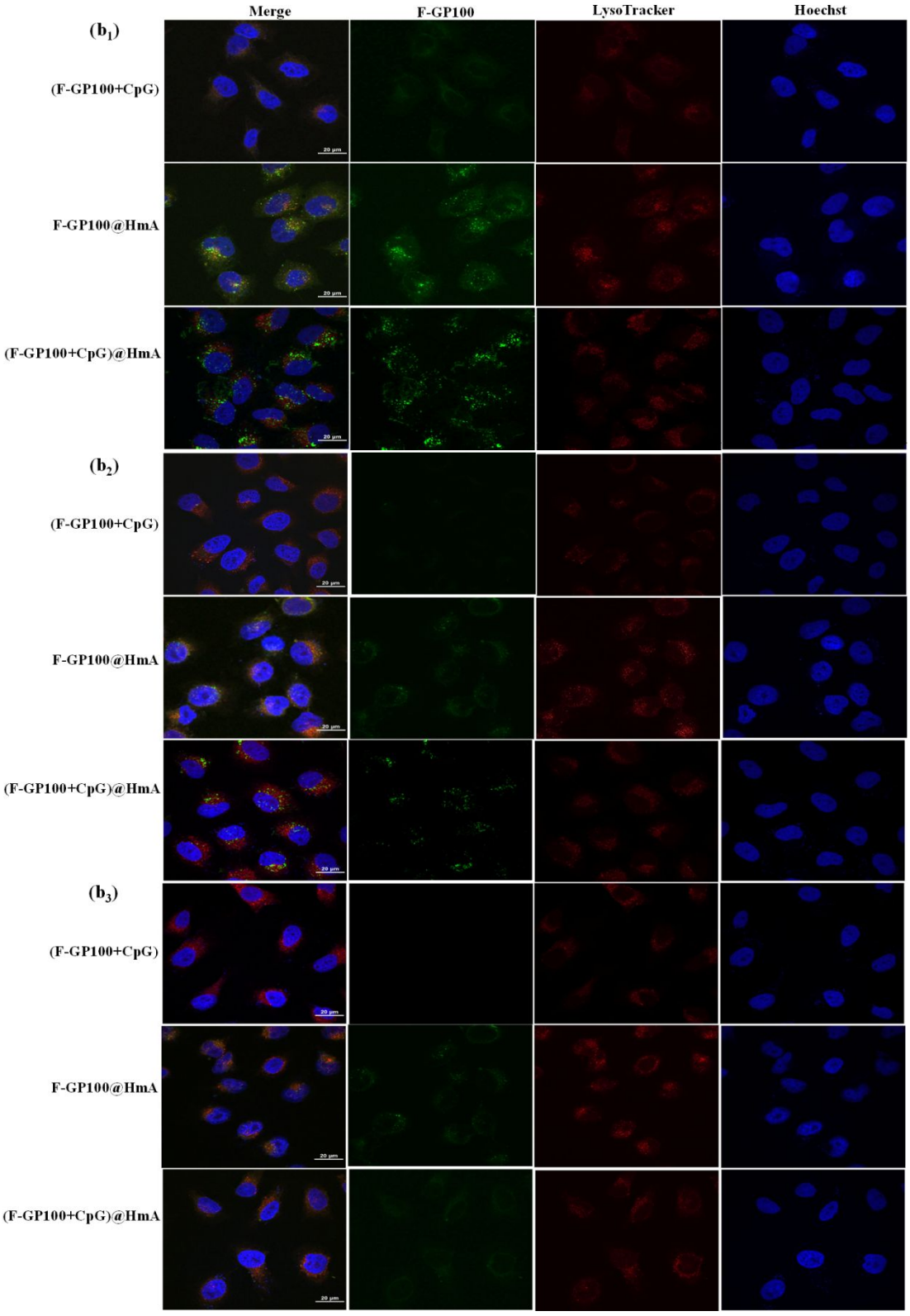


Figure S6. (a) Percentage of positive RAW264.7 treated by (F-GP100+CpG), F-GP100@HmA, and (F-GP100+CpG)@HmA with different incubation time. (b) The merged CLSM images localization of (F-GP100+CpG), F-GP100@HmA and (F-GP100+CpG)@HmA in DC2.4 observed under confocal scanning laser

microscopy, and CLSM images were incubated for 6h (b_1), 24h (b_2) and 48 h (b_3). The cells were stained blue by Hoechst, Lysotracker stained the lysosomes red, and F-GP100 was used for nanovaccine preparation and fluorescence detection. (scale bar = 20 μm)

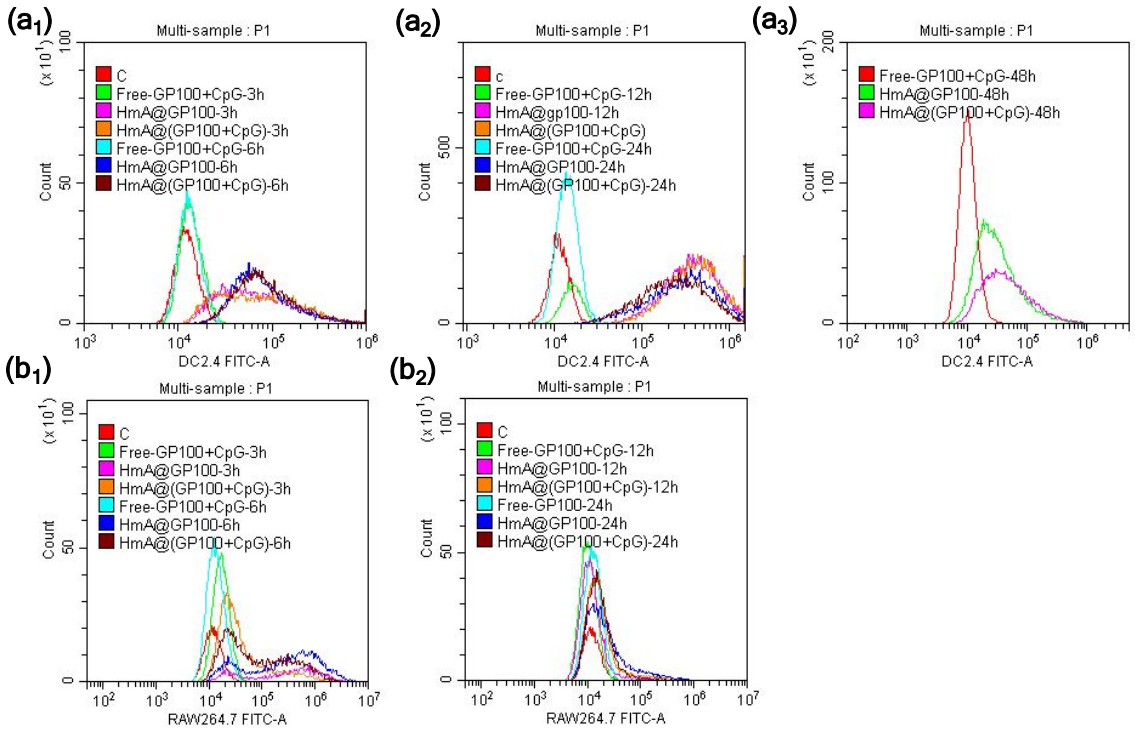


Figure S7. The representative scatter diagrams tested by flow cytometry of (a.) DC2.4 and (b) RAW264.7 treated by (F-GP100+CpG), F-GP100@HmA, and (F-GP100+CpG)@HmA with different incubation time.

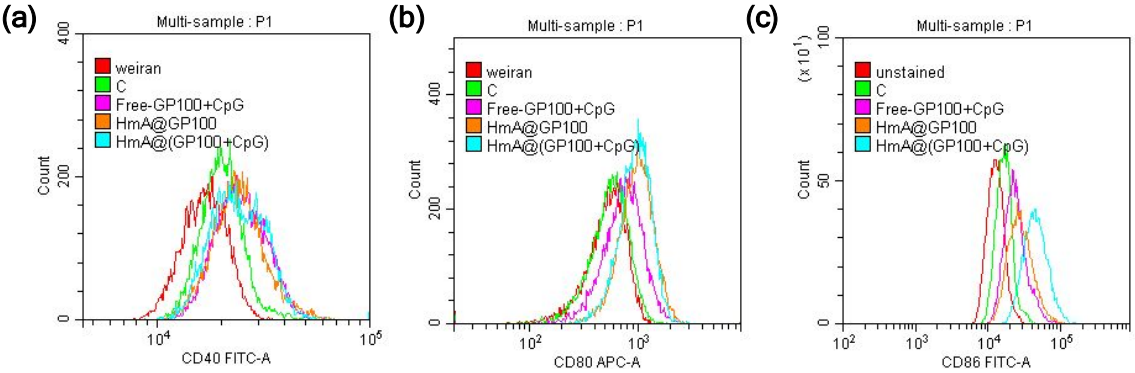


Figure S8. The representative scatter diagrams tested by flow cytometry of the expression of co-stimulatory factors (a) CD40, (b) CD80 and (c) CD86 on the surface of DC2.4.