Electronic Supplementary Information for

Co-delivery of antigen and immunostimulant via a polymersome for improvement of antigen-specific immune response

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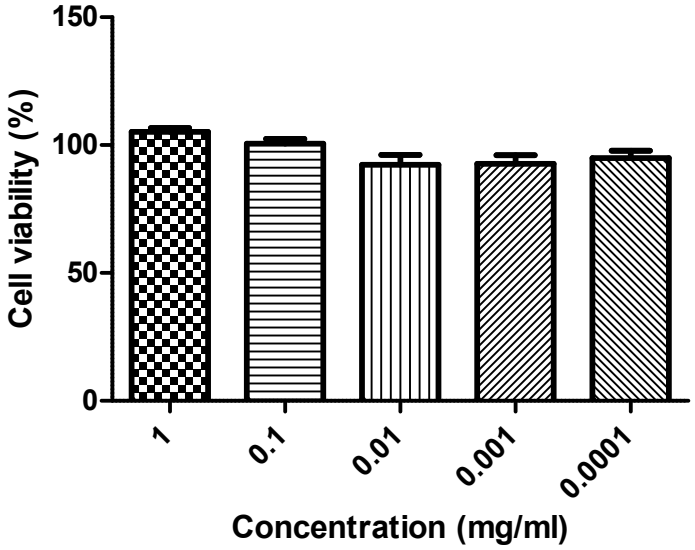

Figure S1. Cytotoxicity of mPEG-b-PLA by Ez-cytox cell viability test for R264.7 cells. Data are expressed as the mean ± S.E.M. (n=5).

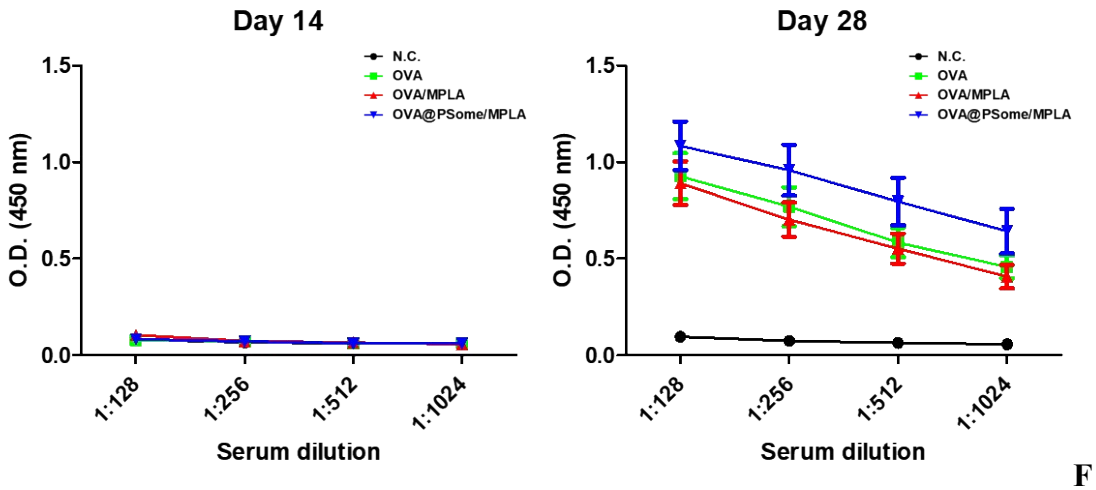

Figure S2. OVA-specific total IgG titer in the serum of mice vaccinated with OVA, OVA/MPLA, and OVA@PSome/MPLA 2 weeks after first and second vaccinations. Data are expressed as mean ± S.E.M. (n=5).

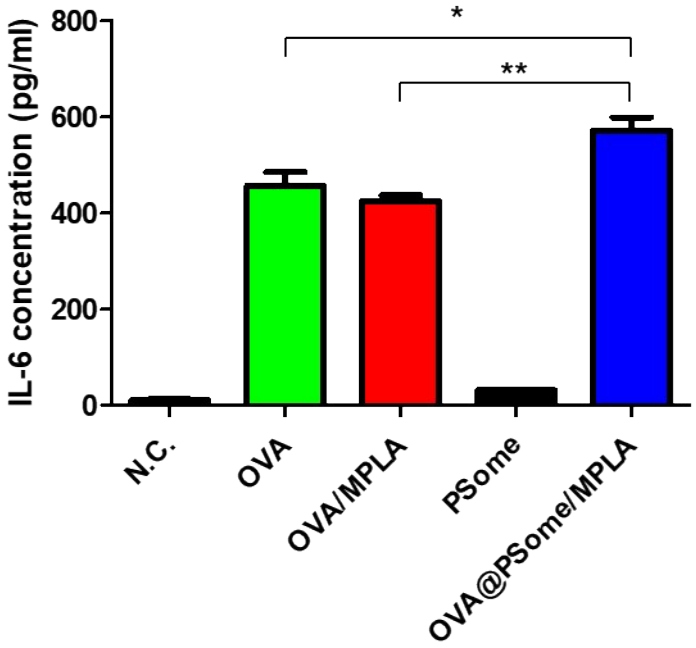

Figure S3. Quantitative comparison of IL-6 induction in xxx cells. Expression levels of IL-6 in macrophage was stimulated with OVA only (2.5 μg), OVA (2.5 μg)/MPLA (0.4 μg), PSome (83 μg) and OVA (2.5 μg)@PSome (83 μg) /MPLA (0.4 μg). The expression level of IL-6 in the supernatants was determined by ELISA. Student t-test was used for statistical analysis. Data are presented as the mean ± S.E.M. (n=3; *$p < 0.05$, **$p < 0.01$).

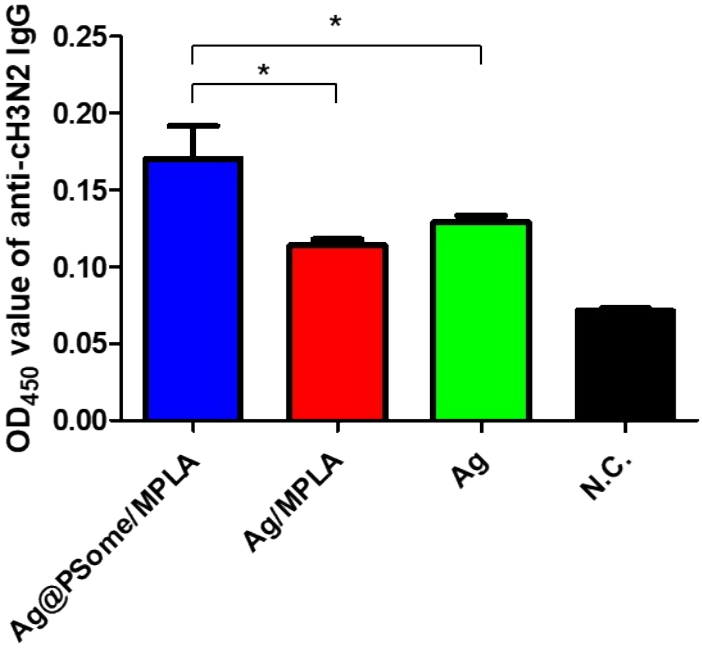

Figure S4. Quantitative comparison of cH3N2-specific Ab induction in mice group (n=5 for each group) at 2 wks after final vaccination. Induction of cH3N2-specific to (a) serum total IgG in mice vaccinated with inactivated Ag only, Ag/MPLA, and Ag@PSome/MPLA were determined using ELISA. Student t-test were used for statistical analysis. Data are presented as the mean ± S.E.M. (*$p < 0.05$).

Table S1. Characterization of synthetic mPEG-b-PLA copolymers

Polymer [a]	f_{mPEG} [b] (%)	M.W. [c] (g/mol)	f_{mPEG} [d] (%)	M.W. [e] (g/mol)
$mPEG_{44}$-b-PLA_{39}	0.385	5191.7	0.416	4803.2
$mPEG_{44}$-b-PLA_{47}	0.278	7179.6	0.371	5390.5

[a] Degree of PLA polymerization was determined by GPC.

[b, d] Calculated mass fraction of mPEG in block copolymers. $f_{mPEG} = Mass_{mPEG} / (Mass_{PLA}+Mass_{mPEG})$

[c] Determined from ^{1}H-NMR analysis by calculating the ratio of the methyl groups within PLA.

[e] Determined from GPC measurement analysis by calculating the standard curve of PEG series.

Table S2. Particle characterization of PSome nanoparticle constructs used in this study.

Method	Formulation	Hydrodynamic diameter (nm)	Zeta potential (mV)	Polydispersity index	Diameter after 14 days at 4°C (nm)
Thin film hydration	OVA@PSome#1/ MPLA	70.4±32.3	-12.38	0.234	92.0±40.3
	OVA@PSome#2/ MPLA	98.9±31.7	-14.73	0.093	88.8±19

Measured by dynamic light scattering. Hydrodynamic diameter values are given as mean ± S.D.