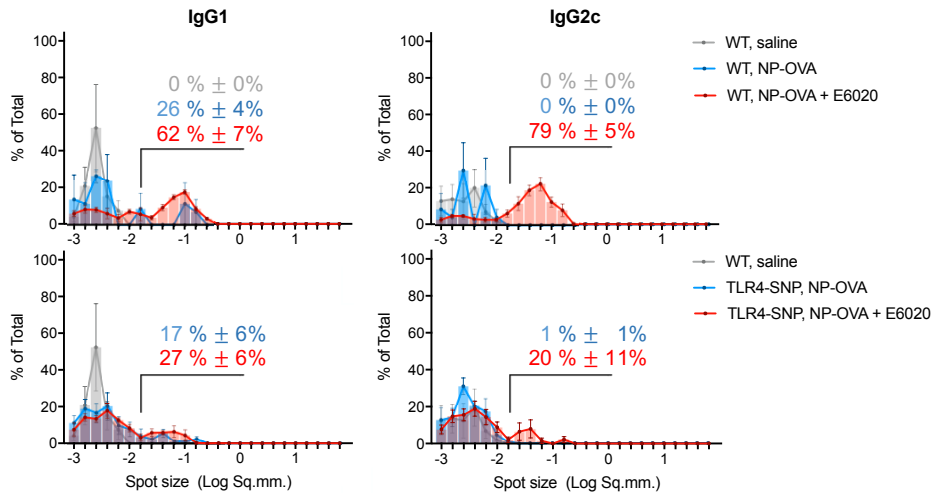


A. ELISA titer and ELISpot frequencies on day 80 after immunization.

Genotype and Treatment Group	ELISA: IgG1 Titer ($\times 10^3$) Mean \pm SEM	ELISA: IgG2c Titer ($\times 10^3$) Mean \pm SEM	ELISpot: IgG1 Frequency (per 10^6 splenocytes) Mean \pm SEM	ELISpot: IgG2c Frequency (per 10^6 splenocytes) Mean \pm SEM
C57BL/6J NP-OVA (n=3)	3.0 \pm 0.8	undetected	4.0 \pm 1.0	2.9 \pm 1.4
C57BL/6J NP-OVA + E6020 (n=3)	45. \pm 8.7 **	13. \pm 2.2 ***, #	54. \pm 6.4 **	23. \pm 4.2 ~
TLR4-SNP NP-OVA (n=4)	4.0 \pm 1.9	undetected	18. \pm 3.4	13. \pm 1.8
TLR4-SNP NP-OVA + E6020 (n=4)	16.0 \pm 1.3 *	1.7 \pm 0.8 *	25. \pm 12	16. \pm 3.4

B.



Supplementary Figure 1: E6020 sustained enhanced immune responses to NP-OVA in C57BL/6J mice, but not TLR4-SNP mice. Age-matched C57BL/6J WT and TLR4-SNP mice were immunized on days 0 and 21 as described in Figure 3. A: Sera were harvested from blood collected from individual mice on day 80 and titers of IgG1 and IgG2c anti-NP antibody were measured by ELISA. Splenocytes from the same mice were incubated overnight on ELISpot plates and developed as described in Materials and Methods. Each data set was analyzed by Two-Way ANOVA with Tukey's multiple comparison post-tests: for NP-OVA alone vs. NP-OVA + E6020 within the same genotype: ***, $p < 0.001$; **, $p < 0.01$; *, $p < 0.05$; ~, $p = 0.0571$; NP-OVA + E6020 in C57BL/6J vs. TLR4-SNP: #, $p < 0.05$. B: Spot size distribution histograms (Immunospot® version 5.0 spot recognition software) of IgG1 (left) and IgG2c (right); grey—unimmunized control mice, blue—NP-OVA immunized mice, and red—NP-OVA+E6020-immunized mice; upper panels C57BL/6J, lower panels TLR4-SNP; single experiment with 3-4 mice/group, shown are arithmetic mean and SEM of the biological replicates.