Suppl. Fig. 1. Gating strategies for flow cytometry. Lymphocytes were isolated from ILN 14 d.p.v. and analyzed for the frequency of follicular CD8+ and CD4+ T cells (Figure 9A and 9B, Suppl. Fig. 3A,B). Samples were acquired on a LSR II flow cytometer (BD Biosciences) and data were analyzed with FlowJo (Tree Star). Cells were gated for singlets (A) first and then lymphocytes (B). CD8+ follicular T cells were determined to be CD4- (PerCP-Cy5.5-) CD3ε+ (APC+) (C) and CXCR5+ (PE-Cy7+) PD-1+ (APC-Cy7+) (Da). CD4+ follicular T cells were determined to be CD4+ (PerCP-Cy5.5+) CD3ε+ (APC+) (C) and CXCR5+ (PE-Cy7+) PD-1+ (APC-Cy7+) (Db).
Suppl. Fig. 2. Activation of T cells in inguinal lymph nodes (ILN) and spleen.

Lymphocytes were isolated from the ILN and the spleen 7 and 14 days post-MN vaccination. Gating strategy for flow cytometry is described in Suppl. Figure 2 (A) CD4+ follicular cells (CD4+CXCR5+PD-1+) were quantified via flow cytometry, and (B) vaccine-specific IL-4-secreting cells were isolated from the spleen at day 7, and enumerated in ELISPOT plates following stimulation with A/Christchurch/16/2010 subunit vaccine. Values are expressed as mean ± SEM.
Suppl. Fig. 3. Cross-reactive antibody titers against H1N1 and H3N2 viruses. Serum was collected from mice 90 days post-A/Christchurch/16/2010 vaccination with dissolving MN. IgG reactivity with monovalent subunit vaccines for (A) H1N1 A/California/10/1978 (p=0.2), (B) H1N1 A/FM/1/1947 (p=0.3), (C) H3N2 A/Victoria/210/2009 (p=0.055), and (D) H3N2 A/Aichi/2/1968 (p=0.055) was measured with ELISA. Statistics performed in Mann-Whitney test for comparison of 1 μg HA/GM-CSF vs. 1 μg HA groups.