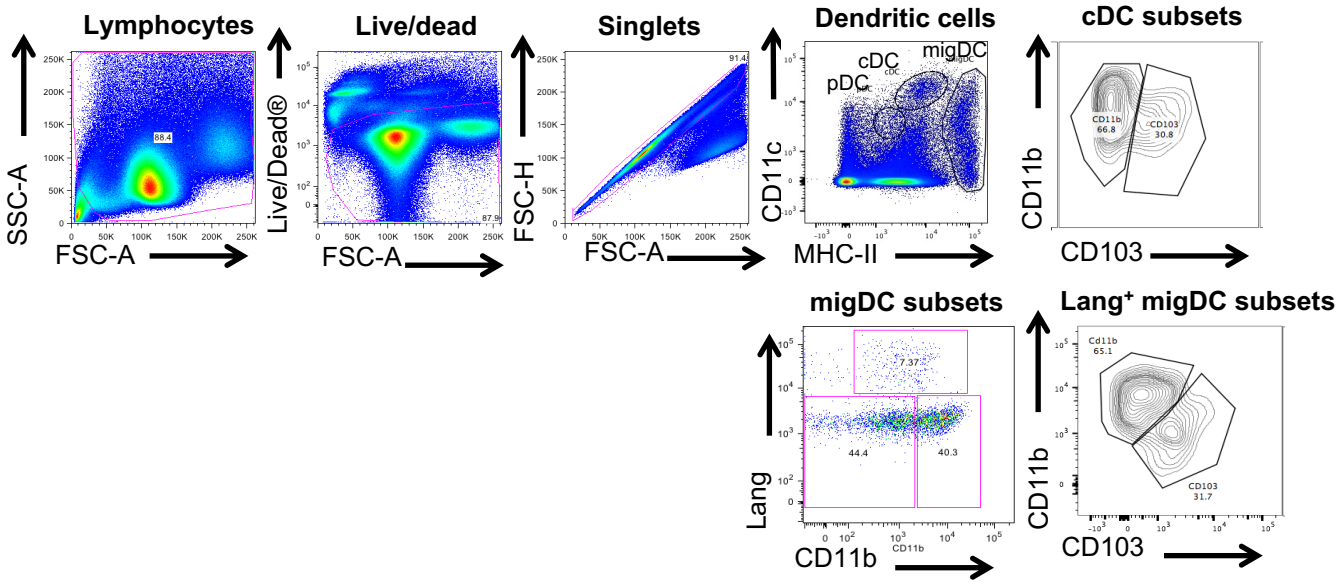


Supplementary Figure S1. Multi-parameter flow cytometry gating schematics for dendritic cell analysis.

Gating schematic to identify dendritic cell subsets within skin-draining lymph nodes. Dendritic cells (DCs) in skin-draining lymph nodes were processed and stained for multi-parameter flow cytometry 24 hours after vaccination with 40 µg Alexa Fluor-488-labeled ovalbumin (OVA) with or without one minute continuous wave (CW) or pulsed-wave (PW) 1064 nm NIR laser treatment.

Figure S1

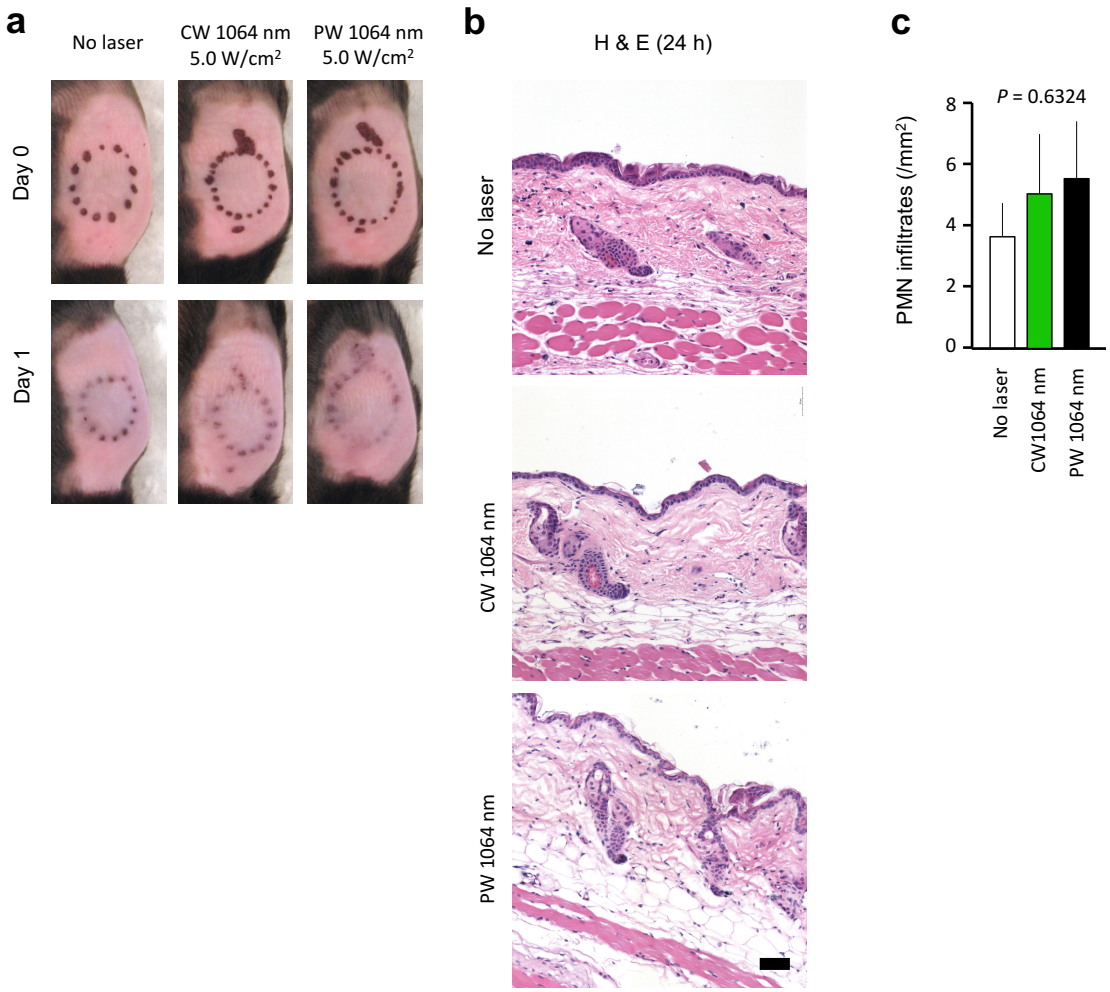
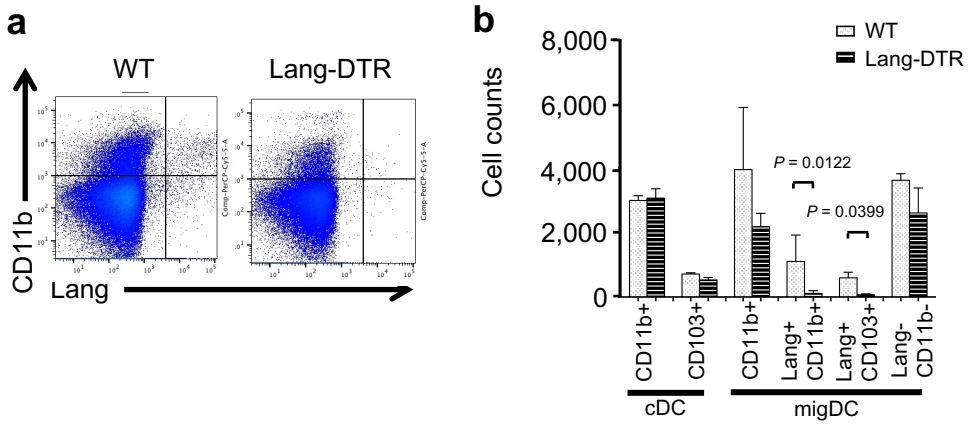


Figure S2. Effect of NIR laser on skin tissue.

(a) Images of the back of mice for visual inspection after the indicated CW or PW near-infrared (NIR) laser treatment. Note that no visual damage was observed at any time point. Representative images at 6 and 24 hours for each group are presented. (b) Microscopic assessment of skin damage and inflammatory infiltration after laser treatment. Note that no microscopic damage was observed at 2 hours (data not shown). Representative images of hematoxylin-eosin-stained skin tissue 24 hours after the NIR laser treatment are presented. The bar indicates 50 μm. (c) Quantification of polymorphonuclear leukocytes (PMN) 24 hours after the NIR laser treatment. Note that no significant increase in PMN infiltration was observed at 2 or 6 hours (data not shown). Error bars show means ± s.e.m. Results are pooled from two independent experiments and analyzed using one-way ANOVA with Tukey's correction. (a-c) n = 2-3 for each group.

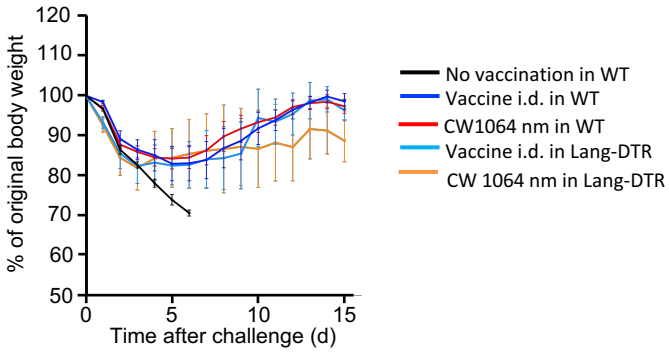
Figure S2



Supplementary Figure S3. Depletion of cells harboring DTR in Langerin-GFP/DTR animals.

In order to deplete cells of interest, 4 ng/g diphtheria toxin (DT) was intraperitoneally (i.p.) injected into wild-type (WT) and Lang-DTR mice. The following day, the DT treated mice were vaccinated intradermally with 40 µg OVA. DCs in skin-draining lymph nodes (skin-dLN) were processed and stained for multi-parameter flow cytometry 24 hours after intradermal vaccination. (a) Representative gates of Langerin (Lang)$^+$ migratory DC (migDC). (b) Cell counts of migDC and classical lymphoid tissue-resident DCs (cDCs) subsets in skin-dLN with or without DT treatment. Experimental and control groups: (a-b) n = 10, 6 for WT, Lang-DTR, respectively. Data are derived from three independent experiments.

Figure S3



Supplementary Figure S4. Effect of laser illumination on body weight following viral challenge.

In order to examine the role of Lang$^+$ cells, 4 ng/g diphtheria toxin (DT) was intraperitoneally (i.p.) injected into wild-type (WT) and Lang-DTR mice. Mice were then vaccinated intradermally with 1 µg inactivated influenza virus (A/PR/8/34) with or without CW 1064 nm NIR laser treatment 24 h after the DT treatment. 28 days later, the mice were intra-nasally challenged with homologous virus. Body weights were monitored daily for 15 days. Mean body weight ± s.e.m. of each experimental group was determined at each time point. Experimental and control groups: n=10, 15, 8, 5, 4 for no vaccine in WT, vaccine i.d. in WT, vaccine i.d. + CW 1064 nm in WT, vaccine i.d. in Lang/DTR + DT, vaccine i.d. + CW 1064 nm in Lang/DTR + DT, respectively.

Figure S4