SUPPLEMENTAL MATERIAL

Willart et al., http://www.jem.org/cgi/content/full/jem.20082401/DC1

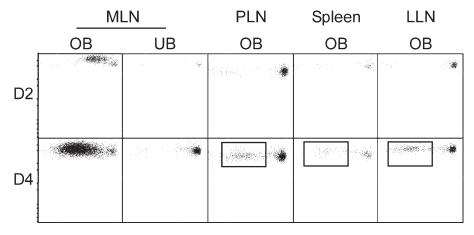


Figure S1. Mice were injected with CFSE-labeled OVA-specific T cells and, 2 d later, injected with either UB or OB i.v. At 2 and 4 d after bead injection, lung draining LNs (MLNs), PLNs, liver LNs (LLNs), and spleen were analyzed by FACS for T cell proliferation. This experiment was performed twice.

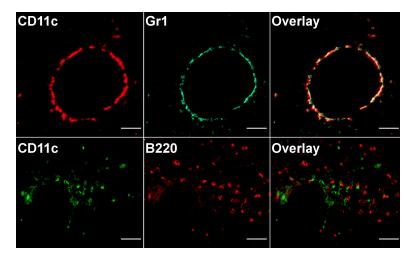


Figure S2. Confocal microscopy of frozen lung sections taken 24 h after injection of OBs in mice that received a cohort of DO11.10 T cells. (Top) Slides were double stained with anti-CD11c PE (red) and anti-Gr1 FITC (green). (Bottom) To discriminate pDCs, slides were also stained for CD11c⁺ cells (FITC, green) in combination with anti-B220 PE (red). This experiment was performed twice. Bars, 10 μm.

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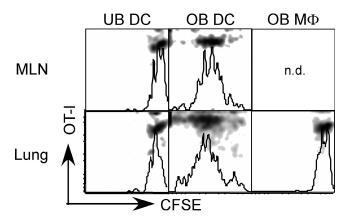


Figure S3. DCs and macrophages were sorted from MLN and lung from mice injected with UB or OB. These APCs were subsequently put in coculture with CFSE-labeled OT-l cells for 4 d. Autofluorescent alveolar macrophages were not detected (n.d.) in the MLNs. This experiment is performed twice.

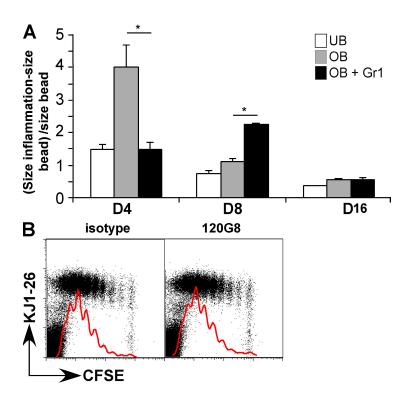


Figure S4. Depletion of monocytes and pDCs by using GR1 depleting antibody around the time of bead injection showed a reduction of inflammatory infiltrate around beads at day 4, whereas treatment with pDC-depleting antibody (120G8) did not reveal differences in antigen presentation. (A) Measurement of the size the inflammatory infiltrate at day 4, 8 and 16 around beads in the lungs of mice receiving either UBs (white bars), OBs plus isotype (OB, gray bars), or OBs plus α Gr-1 (OB Gr1, black bars). Data are shown as mean (n = 4 mice/group) \pm SEM *, P < 0.05. (B) Effect of depletion of 120G8 positive pDCs. Comparison of CFSE labeled OVA specific CD4+ D011.10 division profile in draining MLNs, in mice treated with 120G8 (for pDC depletion) or isotype i.p. at day 0 and injected with OB, measured at day 4. Both experiments were performed twice.