A.


Supplementary figure 1. Ovalbumine (OVA) peptide uptake by CD11c+MHCII+ DCs. A. Isolated dendritic cells (DCs) were incubated with FITC-labeled OVA peptide at $4^{\circ} \mathrm{C}$ and at $37^{\circ} \mathrm{C}$ simultaneously, as described in Materials and Methods. The percentage of OVA peptide uptake by $\mathrm{CD} 11 \mathrm{c}+\mathrm{MHCII}+\mathrm{DCs}$ was calculated by subtracting values of unspecific uptake observed at $4^{\circ} \mathrm{C}$. $\mathbf{B}$. Histogram plots of DC fluorescence after FITC-labeled OVA peptide uptake expressed as mean fluorescent intensities (MFI) in control samples (left histogram) and samples irradiated with 2 Gy (right histogram). Differences between samples incubated in $4^{\circ} \mathrm{C}$ (purple areas) and $37^{\circ} \mathrm{C}$ (green lines) are shown.
A.
$37^{\circ} \mathrm{C}$

0.25 Gy
2 Gy


$4^{\circ} \mathrm{C}$

B.


MHCII FITC
Supplementary figure 2. OVA peptide presentation on major histocompatibility complex ( MHCI ) molecules by CD11c+MHCII+ DCs. Isolated DCs were incubated in the presence of unlabeled SIINFEKL peptide. Cells were labeled with H2b-SIINFEKL-FITC antibody able to detect MHCI-bound SIINFEKL peptide. A. The percentage of OVA peptide presentation by DCs was calculated by subtracting values of unspecific binding observed at $4^{\circ} \mathrm{C}$. B. Gating strategy shown for $\mathrm{CD} 11 \mathrm{c}+\mathrm{MHCII}+$ cells.
A.


B.
isotype
OGy





IL-1 $\alpha$ PE
C.


2 Gy + LPS


IL-1 $\beta$ PE

Supplementary figure 3. A. Gating strategy of $\mathrm{CD} 11 \mathrm{c}+\mathrm{MHCII}+$ cells within splenocytes with lymphocyte population being excluded. Dot plots of (B.) IL1 $\alpha$ and (C.) IL1 $\beta$ levels. The plot shows the percentage of cytokine expression of control ( 0 Gy ) and irradiated ( 2 Gy ) and/or lipopolysaccharide (LPS)-stimulated CD11c+MHCII+ DCs. Mice were total-body irradiated, treated with intraperitoneal (i.p.) LPS injection 18 hours later, followed by i.p. Brefeldin A injection. Cytokine production was determined 6 hours after LPS treatment in the CD11c+MHCII+ splenic DCs by intracellular cytokine labelling as described in Materials and Methods. Data represent one typical experiment.

Supplementary table 1. Treg and Teff cells' proliferation measured in counts per minute (cpm). Either isolated Tregs or Teffs were cultured with DCs isolated from sham-irradiated or irradiated mice as described in Materials and Methods. T cell proliferation was measured by 3H-thymidine incorporation using a liquid-scintillation counter and expressed in cpm.

| Counts per minute (CPM) |  | 0 Gy DC |  | 2 Gy DC |  |
| :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | Teff | Treg | Teff | Treg |
|  | sample 1 | 985 | 1738 | 2260 | 846 |
|  | sample 2 | 1458 | 3578 | 2925 | 2505 |
|  | sample 3 | 1921 | 3578 |  | 2855 |
|  | sample 1 | 4382 | 5060 | 4593 | 3706 |
|  | sample 2 | 5481 | 6873 | 4549 | 4256 |
|  | sample 3 | 6518 | 6431 | 7122 | 3830 |
|  | sample 4 | 5921 | 6023 | 5179 | 2324 |
|  | sample 1 | 1880 | 2253 | 1791 | 2518 |
|  | sample 2 | 1651 | 2413 | 2676 | 2421 |
|  | sample 3 | 2432 | 3305 | 2459 | 2154 |
|  | sample 4 | 1888 | 2652 | 1693 | 1573 |
| Average |  | 3037.91 | 3991.27 | 3524.7 | 2635.27 |
| Standard deviation |  | 2024.85 | 1808.94 | 1765.21 | 999.20 |



Supplementary figure 4. Relative proliferation of Tregs and Teffs after coculture with 2 Gy DCs. Isolated Treg and Teff cells were incubated with 0 Gy and 2 Gy DCs. Proliferation was measured 5 days later by ${ }^{3} \mathrm{H}$-thymidine incorporation as described in Materials and Methods. Data represent the average of three independent experiments, with bars indicating mean $\pm$ SD. Significance was tested by Student's t-test with ${ }^{* *} \mathrm{p}<0.001$.


Supplementary figure 5. Dot plots showing the purity of DC-cell population. CD11c+ cells were purified by magnetic cell sorting using the Miltenyi Biotec's CD11c+ Dendritic Cell Isolation Kit, as described in Materials and Methods. Isolated CD11c+ cells were labelled with CD11c-PE and MHCIIFITC antibody. Data are from one representative experiment.
A.





Supplementary figure 6. (A.) Dot plot and (B.) histogram data showing the purity of isolated Treg cells. CD4+CD25+ Tregs were purified by magnetic cell sorting using the Miltenyi Biotec's CD4+CD25+ Regulatory T Cell Isolation Kit, as described in Materials and Methods. Isolated CD4+CD25+ cells were fixed, permeabilized, and labeled intracellularly with a Foxp3-FITC antibody. Percentage of Foxp3+ cells within CD4+CD25+ cell population represents purity of isolated Treg cells. Data are from one typical experiment.

