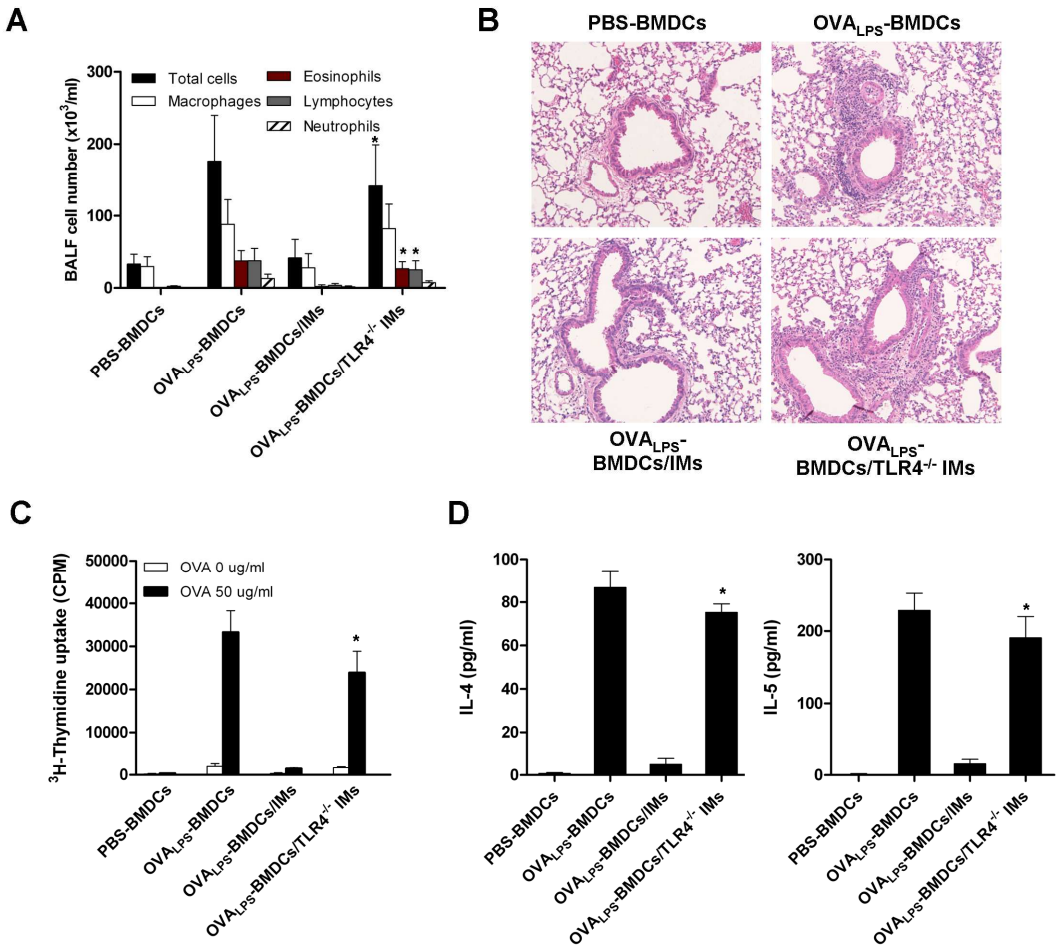
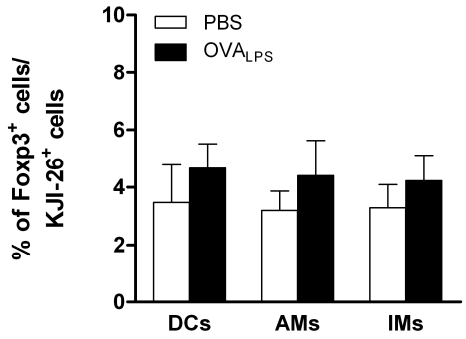
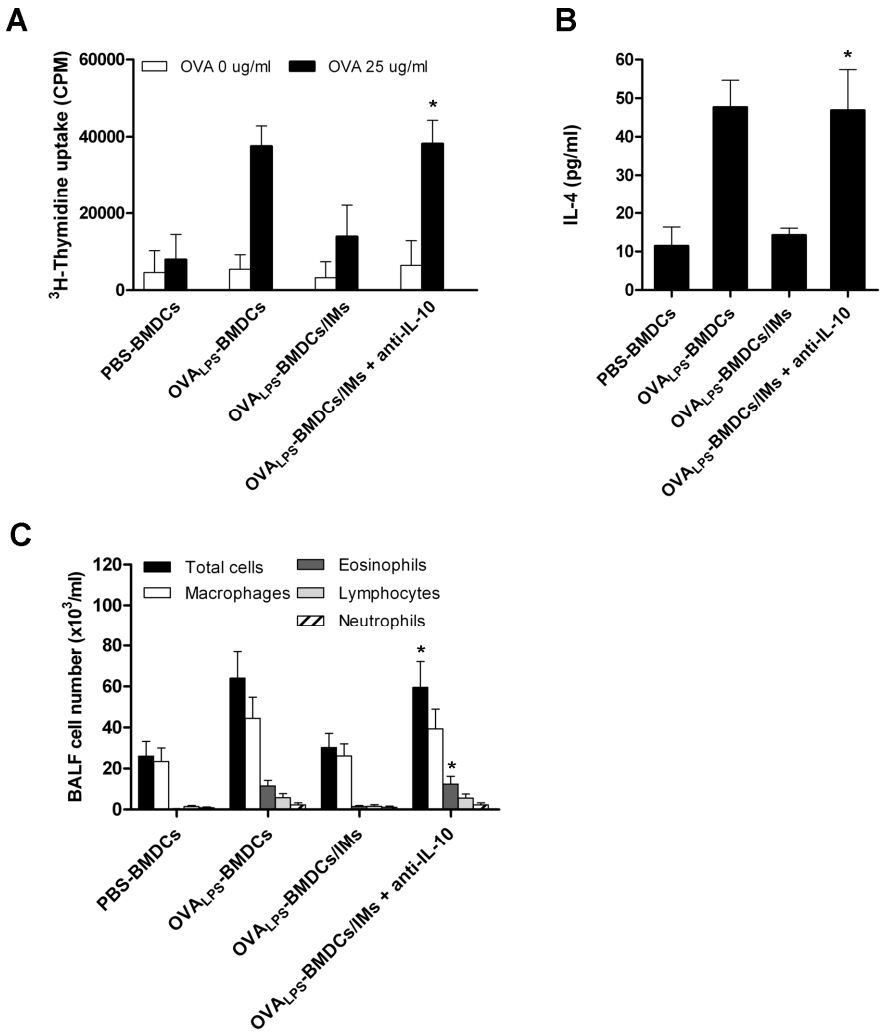


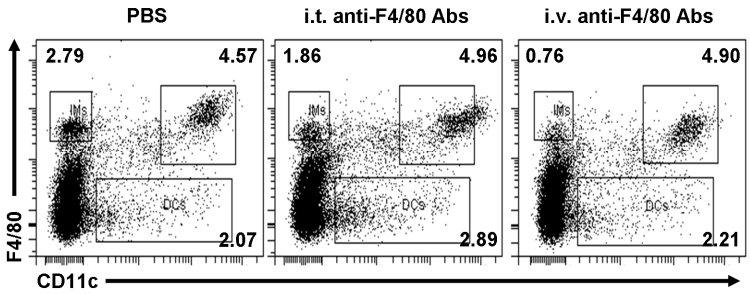
Supplemental Figure 1. TLR4 is required for inducing the immunosuppressive activity of IMs. (A-D) Naive C57BL/6 mice were injected i.t. with either PBS-BMDCs, OVA_{LPS}-BMDCs, OVA_{LPS}-BMDCs/IMs, or OVA_{LPS}-BMDCs/$TLR4^{-/-}$ IMs. From day 10 to day 14, mice were exposed to OVA aerosols. Twenty-four hours after the last challenge, the severity of airway allergy was evaluated. (A) BALF was subjected to total and differential cell counts. (B) Lung sections were stained with hematoxylin and eosin. (C and D) MLN cells were restimulated in vitro for 3 days with 50 μg/ml OVA. (C) The proliferation was measured as ^{3}H-thymidine incorporation during the last 16 h. (D) Culture supernatants were assayed for IL-4 and IL-5 by ELISA. (A, C, and D) *$P < 0.05$ versus OVA_{LPS}-BMDCs/IMs.



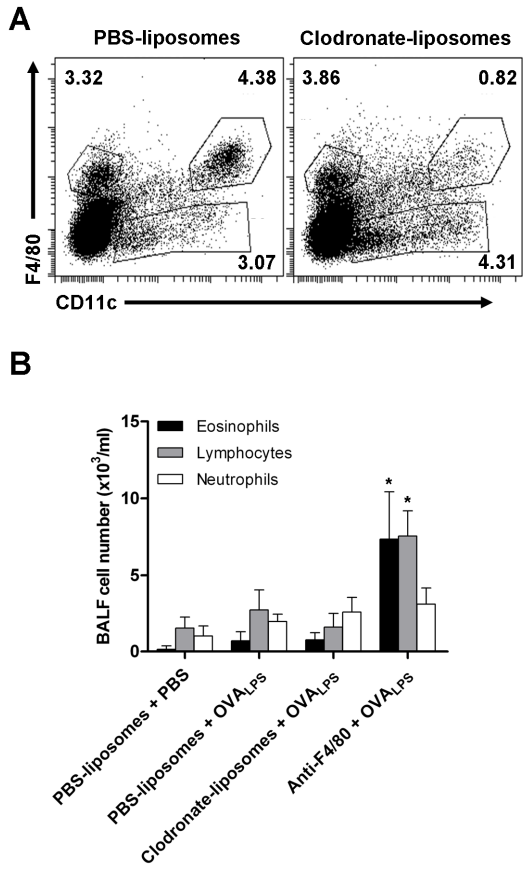
Supplemental Figure 2. IMs are unable to induce the differentiation of $Foxp3^+$ regulatory T cells. FACS-sorted lung DCs, AMs or IMs (2 x 10^4 cells), stimulated or not with OVA_{LPS} (OVA: 100 µg/mouse; LPS: 10 ng/mouse), were cultured for 4 days in the presence of freshly isolated DO11.10 $CD4^+$ T cells (2 x 10^5 cells). The percentage of $Foxp3^+$ T cells among the $CD4^+KJI\text{-}26^+$ DO11.10 T cell population was then assessed by intracellular staining of Foxp3 and flow cytometry analyses.



Supplemental Figure 3. Neutralizing anti-IL-10 antibodies abrogate the immunosuppressive effects of IMs. (A-C) On day 0, naive BALB/c mice received either PBS-BMDCs, OVA_{LPS}-BMDCs, OVA_{LPS}-BMDCs/IMs or OVA_{LPS}-BMDCs/IMs that were cultured in the presence of 1µg/ml neutralizing anti-IL-10 antibodies (OVA_{LPS}-BMDCs/IMs + anti-IL-10). (A and B) On day 4, MLN cells were restimulated in vitro for 3 days with 25 µg/ml OVA. The proliferation was measured (A) and culture supernatants were assayed for IL-4 by ELISA (B). (C) From days 10 to 14, mice were exposed to OVA aerosols. On day 15, BALF was subjected to total and differential cell counts. (A-C) *, $P < 0.05$ versus PBS-BMDCs and OVA_{LPS}-BMDCs/IMs.



Supplemental Figure 4. Intratracheal administration of anti-F4/80 antibodies do not result in depletion of AMs. Naive BALB/c mice were injected i.t. or i.v. on 3 consecutive days with 250 μg of depleting anti-F4/80 or control isotype antibodies. On day 4, lungs were digested and stained for F4/80 and CD11c. Percentages of lung DCs ($F4/80^{-}CD11c^{+}$; lower right quadrant), AMs ($F4/80^{+}CD11c^{+}$; upper right quadrant) and IMs ($F4/80^{+}CD11c^{-}$; upper left quadrant) among living cells are provided.



Supplemental Figure 5. AMs are not required for protection against allergic sensitization in the airways. (A) Naive BALB/c mice were injected i.t. on day 1 with either 100 µl PBS-encapsulated liposomes or 100 µl clodronate-encapsulated liposomes. On day 3, lungs were digested and stained for F4/80 and CD11c. Percentages of lung DCs ($F4/80^{-}CD11c^{+}$; lower right quadrant), AMs ($F4/80^{+}CD11c^{+}$; upper right quadrant) and IMs ($F4/80^{+}CD11c^{-}$; upper left quadrant) among living cells are provided. (B) Naive BALB/c mice were injected i.t. on day 1 with either 100 µl PBS-encapsulated liposomes or 100 µl clodronate-encapsulated liposomes. On day 3, mice received an i.t. injection of OVA_{LPS} (OVA: 100 µg/mouse; LPS: 10 ng/mouse). Control mice received an i.t. injection of PBS rather than OVA_{LPS}. From day 10 to day 13, mice were challenged intranasally with 25 µg OVA (grade V, Sigma) in 50 µl PBS. On day 15, BALF was subjected to differential cell counts. Differential BALF cell counts from mice that received anti-F4/80 antibodies rather than liposomal clodronate prior to sensitization and challenge are given to compare the effects of AM depletion to those of IM depletion. *$P < 0.05$ versus results obtained in the three other groups.