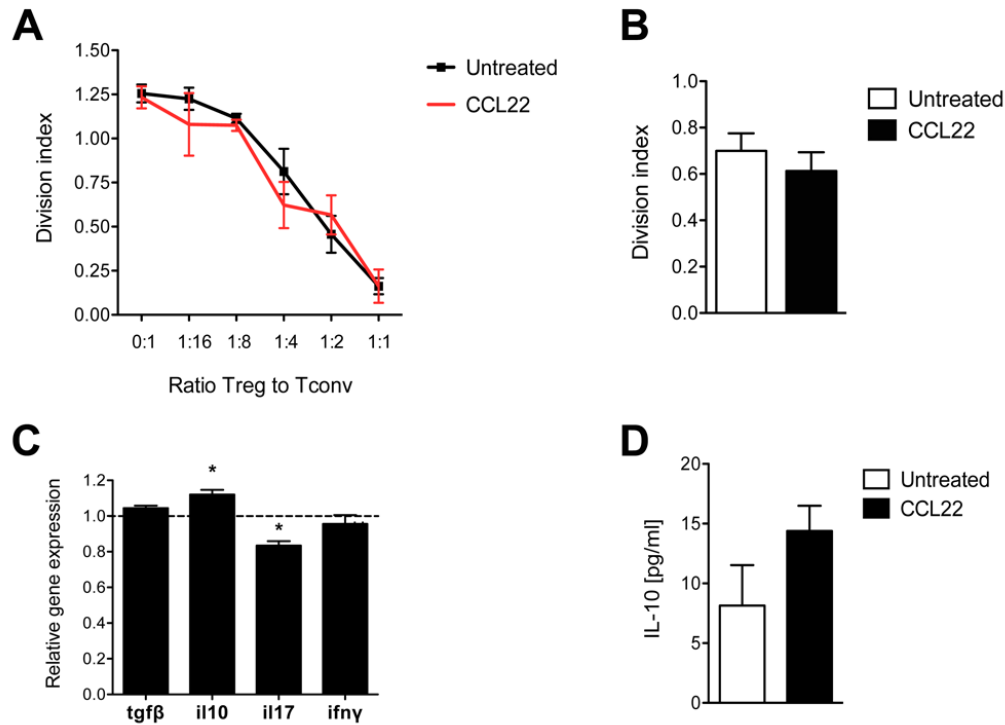


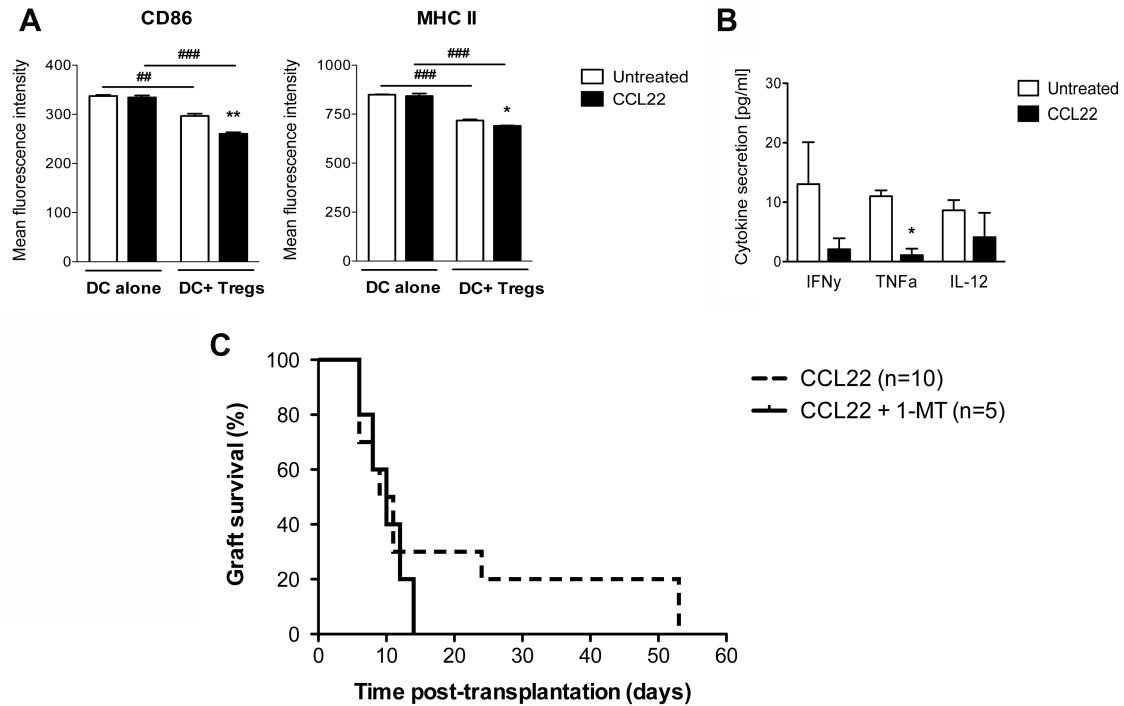
### Supplementary Figure 1: Responsivity of immune subsets to CCL22

**A.** CCR4 expression profile on splenocytes before and after activation with ConA. Data are representative of 2 independent experiments, each with n=4. \*\* $p < 0.01$  and \*\*\* $p < 0.001$  versus control of the same subset. **B.** Mean fluorescence intensity of CCR4 expression on indicated cell subsets compared to Tregs. Data are representative of 2 independent experiments, each with n=4. \* $p < 0.05$ , \*\* $p < 0.01$  and \*\*\* $p < 0.001$  versus Tregs. **C.** Migration of Tregs, iNKT cells and pDCs towards recombinant CCL22 determined at 1, 2 and 4 hours. Migration index represents the ratio of cells that migrate towards CCL22 to cells that migrate to medium only. Data are representative of 2 independent experiments performed in triplicate. \* $p < 0.05$ , \*\* $p < 0.01$  and \*\*\* $p < 0.001$  versus medium alone. **D.** Chemotaxis assay showing migration of CCR4<sup>-/-</sup> or wildtype (WT) leukocytes towards 500 pg/ml of recombinant CCL22. Data are representative of 2 independent experiments, each performed in triplicate. **E.** Chemotaxis assay showing migration of CCR4<sup>-/-</sup> or wildtype (WT) leukocytes towards CCL22-expressing islets. Data are representative of 2 independent experiments, each performed in triplicate. \* $p < 0.05$  and \*\* $p < 0.01$  versus medium alone or as indicated by bar.



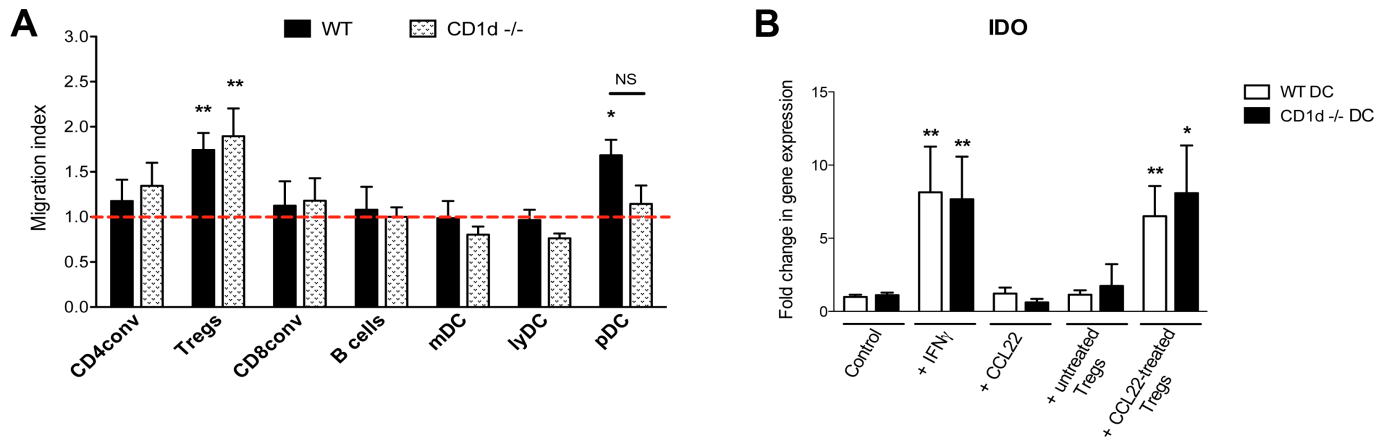
### Supplementary Figure 2: Impact of CCL22 on other Treg functions

**A.** Treg suppression assay of CD4conv proliferation after 4 days of culture at indicated cell ratios in the presence or absence of CCL22. Data are representative of 2 independent experiments, each with 4 replicates.  $p=NS$  at each cell ratio. **B.** Proliferation of isolated Tregs cultured with 500 pg/ml CCL22 for 4 days. Data are representative of 2 independent experiments, each performed in triplicate.  $p=NS$ . **C.** Gene expression profile in isolated Tregs after 24h incubation with 500 pg/ml CCL22. Data are shown as gene expression relative to untreated Tregs. Data are representative of 2 independent experiments, each performed in triplicate.  $*p<0.05$ . **D.** IL-10 protein levels in supernatants of Tregs cultured for 24 h with 500 pg/ml CCL22. Data are representative of 2 independent experiments, each performed in triplicate.  $p=NS$ .



### Supplementary Figure 3: CCL22 impact on DCs and role of IDO in CCL22-mediated protection of islet grafts

**A, B.** DCs were cocultured with Tregs at a ratio of 1:2 for 48h in the presence or absence of CCL22. Data are representative of 2 independent experiments, each performed in triplicate. **A.** Surface expression of CD86 and MHC class II on DCs measured by flow cytometry.  $*p < 0.05$  and  $**p < 0.01$  versus untreated DC+Tregs cocultures.  $###p < 0.01$  and  $####p < 0.001$  as indicated by bars. **B.** Cytokine production by DCs cultured with untreated or CCL22-treated Tregs was measured by Luminex assay.  $*p < 0.05$  versus untreated condition. **C.** To determine the role of IDO in CCL22-mediated protection of islet grafts, diabetic NOD mice were transplanted with Ad-CCL22-transduced islets and given 1-methyl-tryptophan in drinking water or normal water. Survival curves were compared using the log-rank test where  $p = \text{NS}$ .



### Supplementary Figure 4:

**A.** Migration assay towards recombinant CCL22 with splenocytes from WT or CD1d<sup>-/-</sup> mice. Data are representative of 2 independent experiments, each with 4 replicates.

\* $p < 0.05$  and \*\* $p < 0.01$  versus medium alone. **B.** DCs were isolated from WT or CD1d<sup>-/-</sup> mice and cultured with or without Tregs at a ratio 1:2 (DC:Tregs) in the presence or absence of CCL22. IFN $\gamma$  was used a control for IDO stimulation in DCs. IDO gene expression was analyzed after 48 h and expressed as fold change compared to control DCs from the same genotype. Data are representative of 2 independent experiments, each performed in triplicate. \* $p < 0.05$ , \*\* $p < 0.01$  and \*\*\* $p < 0.001$ .