

Supplementary Figure 1. Characterisation of WT and *Ptpn22^{-/-}* **BMDC co-culture with OT-II T-cells.** WT and *Ptpn22^{-/-}* GM-CSF derived bone marrow derived dendritic cells (BMDC) were harvested at day 6 and the proportion (**A**) and number (**B**) of CD11c⁺ BMDC per 24 well plate determined by cell counting with trypan blue exclusion and flow cytometry. (**A-B**) Data are of >7 independent experiments. Bars represent mean \pm s.d. (**C-E**) WT and *Ptpn22^{-/-}* BMDC were pulsed overnight with OVA₃₂₃₋₃₃₉ (50nM) in the presence or absence of curdlan (100 µg/ml) and co-cultured with CTV labelled OT-II T-cells. (**C**) T-cells co-cultured for 6 days with WT or *Ptpn22^{-/-}* BMDC were harvested and replated in IL-2 and IL-23 for a further 4 days. Cell-free supernatants were assessed for IL-17, IFNy, and TNF α production by immunoassay. Each point represents independent WT (closed symbols) or *Ptpn22^{-/-}* (open symbols) BMDC preparations, each paired with OT-II T-cells; NS = not significant, *p<0.05, **p<0.01 by two-way ANOVA, applying Sidak's multiple comparisons test. At day 6 T-cells were either assessed for viability (**D**) and CTV dilution (**E**) by flow cytometry. (**D**) Data are of 5 independent experiments bars represent mean \pm s.d. (**E**) data are mean \pm s.e.m of 10 independent experiments; NS = not significant.



Supplementary Figure 2. PTPN22 regulates T cell dependent IL-17 responses induced by curdlan stimulated BMDC *in vivo*. Wild type (WT) and *Ptpn22^{-/-}* derived bone marrow derived dendritic cells (BMDC) were pulsed overnight with $OVA_{323-339}$ (50nM) in the presence or absence of curdlan (100μ g/ml). BMDC were harvested and injected into the left footpad of OT-II mice. 7 days post immunisation the non-draining and draining popliteal lymph nodes were isolated and the (A) number of cells within the non-draining lymph nodes determined. (B) Total non-draining lymph node T-cells were stimulated with immobilised anti-CD3 for 48 hours and cell-free supernatant assayed for IL-17, IFNy and TNF α by immunoassay. Data are representative of three independent experiments, each data point representing an individual OT-II mouse lymph node. Bars represent the mean \pm s.d. NS = not significant.



Supplementary Figure 3. WT and *Ptpn22^{-/-}* BMDC activation with curdlan, HKCA and LPS. (A) WT and *Ptpn22^{-/-}* bone marrow derived dendritic cells (BMDC) (1x10⁶ c/ml) were stimulated for 24 hours in the presence or absence of curdlan (400, 100, or 25 µg/ml) and cell-free supernatants were assessed for IL-1 β . Data are representative of 3 independent experiments, bars represent mean ± s.d. (B, C) WT and *Ptpn22^{-/-}* BMDC (1x10⁶ c/ml) were stimulated for 24 hours in the presence or absence of (B) HKCA (6.25x10⁵ c/ml) or (C) LPS (100 ng/ml). Cell-free supernatants were assessed for IL-1 β , IL-6, IL-12/23p40, and TNF α by immunoassay. Data are of >10 independent experiments, bars represent mean ± s.d; *p<0.05, **p<0.01, ***p<0.001 compared with two-way ANOVA applying Sidak's multiple comparisons test.



Supplementary Figure 4. Characterisation of WT and *Ptpn22^{-/-}* BMDC activation. (A, B) Day 6 WT and *Ptpn22^{-/-}* BMDC were surface staiged witboantibodies towards CD11c and dectin-1 and exp20 ssion40 eterms bed by flow cytometry. (A) Representative histogram of 1 of 6 independent experiments; solid black investor and *Ptpn22^{-/-}* FMO (B) Dectin-1 geometric mean fluorescence intensity (GMFI) data are of 6 independent experiments; line represents the mean \pm s.d. (C-E) Day 6 WT and *Ptpn22^{-/-}* BMDC were stimulated for 24 hours in the presence or absence of curdlan (100µg/ml) and stained for (C) viability dye and (D) surface markers CD11c, MHCcII IA^b, CD40 and CD86. Cell debris was excluded and the % of viability dye negative cells calculated. Data are of 7-9 independent experiments. Bars represent mean \pm s.d. (E) WT and *Ptpn22^{-/-}* BMDC or WT BMDC pre-treated with anti-dectin-1 (100µg/ml) were incubated at 37°C with UV labeled HKCA for 0-60 minutes and the percentage of CD11c⁺UV⁺ cells determined by flow cytometry. WT and *Ptpn22^{-/-}* data are of 7 independent experiments and WT + α -dectin-1 are of 3 independent experiments. Bars represent mean \pm s.d.



Supplementary Figure 5. Induction of IL-17 by curdlan activated BMDC is IL-1 dependent. (A) Day 6 bone marrow derived dendritic cells (BMDC) were pulsed overnight with OVA₃₂₃₋₃₃₉ in the presence or absence of curdlan (100µg/ml). BMDC were then co-cultured with OT-II T-cells for 6 days in the presence or absence of IL-1RA (0.2µg/ml). Cell-free supernatants were assessed for IL-17, IFNγ, and TNFα by immunoassay. Data show three independent experiments, bars represent mean \pm s.d; *p<0.05 compared with one-way ANOVA, with Holm-Sidak's multiple comparisons test. **(B)** WT and *Ptpn22^{-/-}* derived bone marrow derived dendritic cells (BMDC) were pulsed overnight with OVA₃₂₃₋₃₃₉ (50 nM) in the presence or absence of curdlan (100 µg/ml). BMDC were harvested and 5x10⁵ cells were injected into the left footpad of OT-II mice in the presence or absence of 0.5mg IL-1RA. 7 days post immunisation the non-draining (right) popliteal lymph nodes were isolated. Total non-draining lymph node T-cells were stimulated with immobilised anti-CD3 for 48 hours and cell-free supernatants assayed for IL-17, IFNγ and TNFα by immunoassay. Data are representative of two independent experiments (N = 3-5 mice per group), each point representing an individual OT-II mouse lymph node. Data represent mean ± s.d. NS = not significant, * p<0.05, **p<0.01, ***p<0.001 by two-way ANOVA, applying Sidak's multiple comparisons test.



Supplementary Figure 6. Dectin-1 induced WT and Ptpn22-/- BMDC signalling Bone marrow derived dendritic cells (BMDC) were pretreated for 30 minutes with (A, B) Syk inhibitor SykII (2 μ M) or (C) MEK1/2 inhibitor U0126 (10 μ M) or DMSO and stimulated for 24 hours in the presence or absence of (A, C) curdlan (100 µg/ml) (B) LPS 100ng/ml. Cell-free supernatants were assessed for expression of IL-1β by immunoassay. Data are representative of 4-5 independent experiments, presented as mean \pm s.d; NS = not significant, **p<0.01; ***p<0.001 applying one-way ANOVA. (D) WT bone marrow derived dendritic cells (BMDC) (3x10⁶ c/ml) were pretreated with anti-dectin-1 (5µg/ml) and stimulated for 10 minutes at 37°C in the presence of HKCA (2.5 x10⁶ c/ml). Whole cell lysates were blotted for total and pSyk (E-F) WT and Ptpn22^{-/-} BMDC (3x10⁶ c/ml) were stimulated for 0-20 minutes at 37°C in the presence of HKCA (6.25 x10⁵ c/ml). Whole cell lysates were blotted for total and Pp38 and total and plkB α . (E) Representative blots of 3 independent experiments (F) ImageJ quantification of band intensity. Phosphorylated protein values were normalised to total protein and the fold change to 0 min calculated. Bars represent the mean of 3 independent experiments ± s.e.m. (G-H) WT and Ptpn22^{-/-} BMDC were stimulated for 0-20 minutes at 37°C in the presence of curdlan (100µg/ml). Whole cell lysates were blotted for total and pErk (G) Representative blots of 3 independent experiments (H) ImageJ quantification of band intensity. Phosphorylated protein values were normalised to total protein and the fold change to 0 min calculated. Bars represent the mean of 3 independent experiments ± s.e.m.; *p<0.05 compared with two-way ANOVA, applying Sidak's multiple comparison test.



Supplementary Figure 7. Curdlan induced IL-1ß trancription. (A) WT and *Ptpn22^{-/-}* bone marrow derived dendritic cells (BMDC) were stimulated for 0-24h at 37°C in the presence of curdlan (10 μ g/ml). Expression of IL-1 β was determined by real-time PCR and normalised to expression of 18S and IL-1 β expression at 0h. Bars represent the mean of triplicate values representative of 3 independent experiments ± s.d.