Supplement

Figure S1. CVB3 or LCMV infection of prediabetic NOD mice causes PD-L1 up-regulation on all subsets of lymphoid cells in the pancreatic LN and spleen. Percentage of PD-L1^{hi} cells over time in CD4^{+} T cells, CD8^{+} T cells, B cells and DCs from the pancreatic LN (left) and spleen (right) of NOD mice after infection with CVB3 (top) or LCMV (bottom) at 9 weeks old, measured by flow cytometry. Shown is that average percentage of PD-L1^{hi} cells ± standard deviation obtained using 2 to 11 individual mice per group/time point.

Figure S2. Inhibition of LCMV-induced up-regulation of PD-L1 on lymphoid cells using PD-L1 siRNA. (A) Percentage of PD-L1^{hi} cells in NOD splenocytes cultured for 24 h *in vitro* with media alone or LCMV (MOI=1) after electroporation in the absence of RNA (None) or transfection with a control, scrambled-sequence siRNA (Control), measured by flow cytometry. Shown is the average percentage of PD-L1^{hi} cells ± standard deviation for each group in duplicate. (B) Representative flow cytometry histogram plots showing PD-L1 expression in the "None" (black dotted lines) and "Control" (green solid lines) groups. Numbers indicate the average percentage of PD-L1^{hi} cells for each group in duplicate. Gray histograms represent isotype control staining. (C) PD-1 and CD44 expression by IGRP-specific CD8^{+} T cells in the pancreatic LN and spleen of 12-week-old NOD mice infected 21 days prior with LCMV and simultaneously injected with a cationic vehicle alone (LCMV, green bars) or containing PD-L1 siRNA #76238 (LCMV+PD-L1si, white bars), measured by flow cytometry. Shown is the average percentage of PD-1^{+} or CD44^{hi} gated tetramer^{+} cells ± standard deviation for 6 individual mice per group.

Figure S3. Proposed model for prevention of T1D by acute viral infection. Upon infection by β-cell non-lytic viruses during the prediabetic phase, release of inflammatory cytokines causes a "wave" of PD-L1 up-regulation in lymphoid organs, which curbs diabetogenic $CD8^+$ T cells and delays the onset of overt diabetes. In parallel, natural Tregs stimulated (by e.g. DCs) under inflammatory conditions expand and acquire enhanced suppressor function, notably via TGF-β production. Such invigorated, "inflammation-specific" Tregs take additional advantage of PD-L1/PD-1-mediated demise of autoreactive effectors to efficiently control the remaining diabetogenic T cells, and thus prevent T1D with high efficacy.

Figure S1

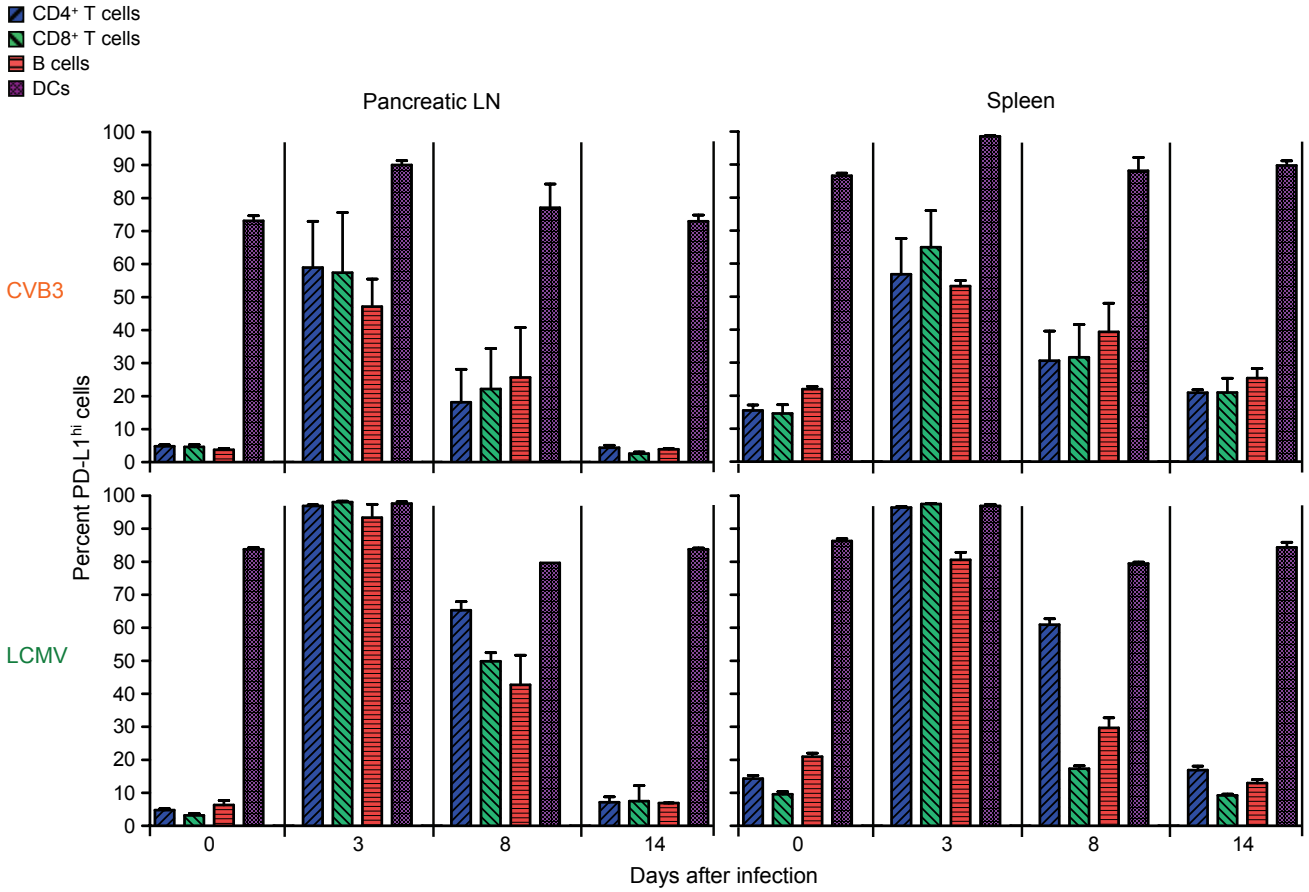

CD4+ T cells
CD8+ T cells
B cells
DCs
Pancreatic LN
Spleen
CVB3
LCMV
Percent PD-L1hi cells
0
10
20
30
40
50
60
70
80
90
100
0
3
8
14
Days after infection

Figure S2

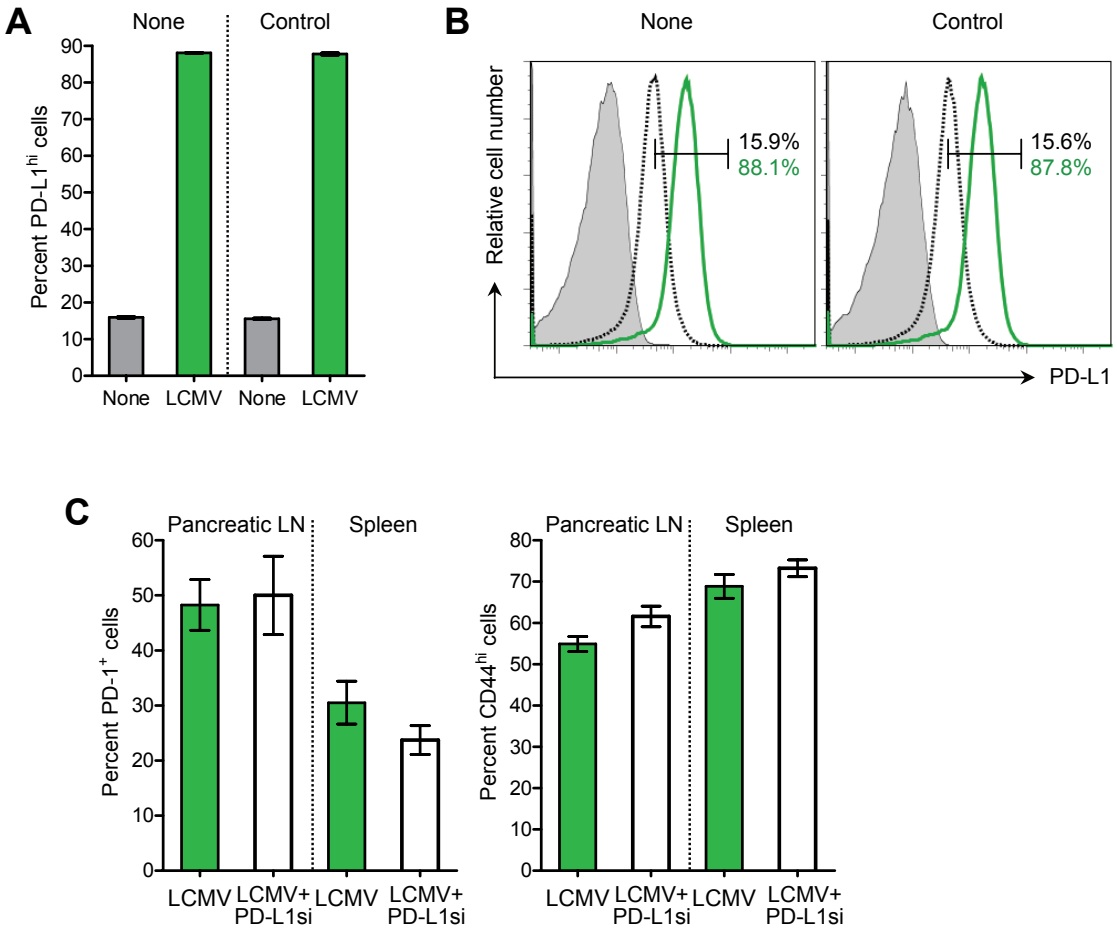
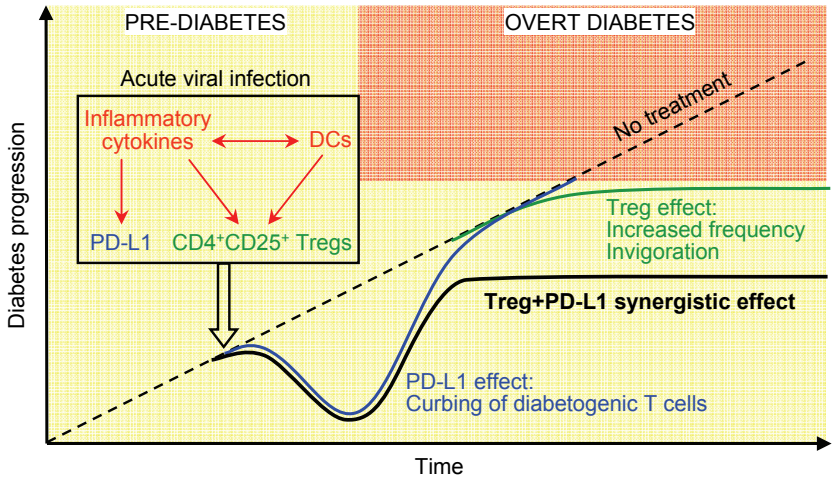

A
None
Control
Percent PD-L1hi cells
90
80
70
60
50
40
30
20
10
0
None
LCMV
None
LCMV
B
None
Control
Relative cell number
15.9%
88.1%
15.6%
87.8%
PD-L1
C
Pancreatic LN
Spleen
Percent PD-1+ cells
60
50
40
30
20
10
0
LCMV
LCMV+ PD-L1si
LCMV
LCMV+ PD-L1si
Pancreatic LN
Spleen
Percent CD44hi cells
80
70
60
50
40
30
20
10
0
LCMV
LCMV+ PD-L1si
LCMV
LCMV+ PD-L1si

Figure S3


PRE-DIABETES
OVERT DIABETES
Acute viral infection
Inflammatory cytokines
DCs
PD-L1
CD4+CD25+ Tregs
No treatment
Treg effect:
Increased frequency
Invigoration
Treg+PD-L1 synergistic effect
PD-L1 effect:
Curbing of diabetogenic T cells
Diabetes progression
Time