Supplementary Materials

Endogenous programmed death ligand-1 restrains the development and onset of Sjögren's syndrome in non-obese diabetic mice

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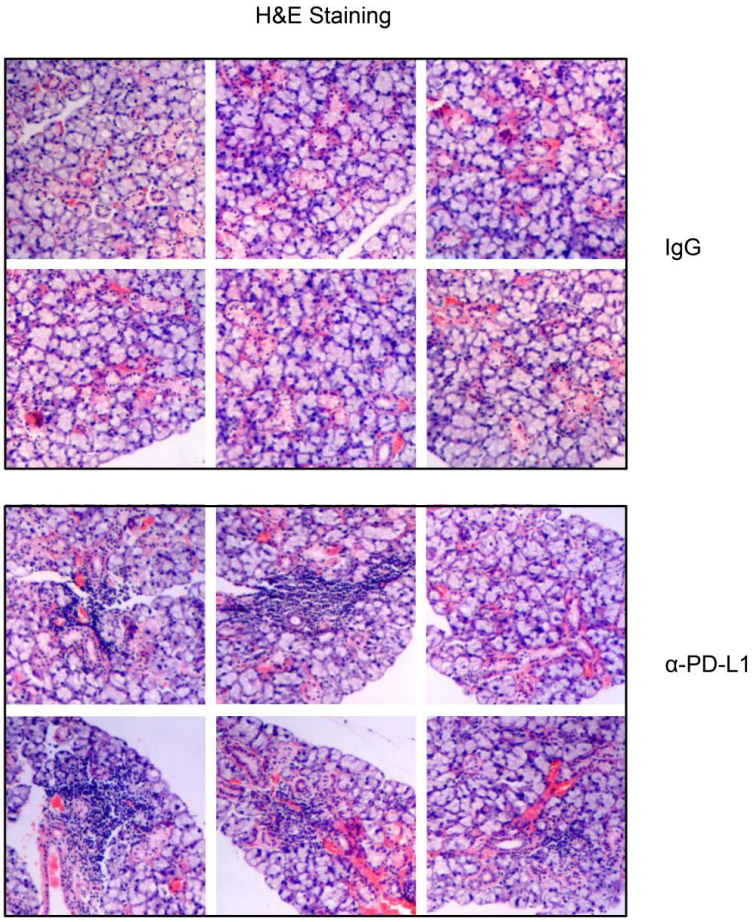
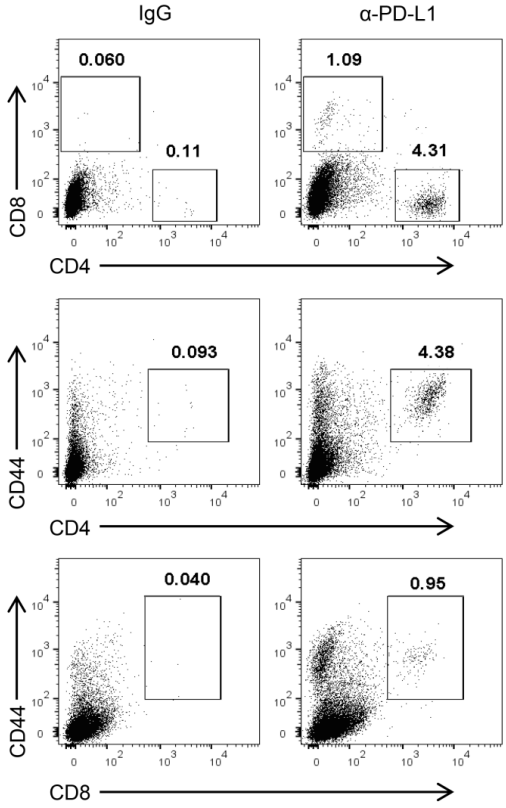


Supplementary Figure S1. Blockade of PD-L1 accelerates the development of salivary gland inflammation. NOD/ShiLtJ mice received injections of rat anti-mouse PD-L1 antibody and the isotype control rat IgG2b. Each group of tile image shows multiple areas of a representative H&E stained SMG section from the IgG-treated group (upper panel) or anti-PD-L1-treated group (lower panel). Original magnification × 200. Data are representative of analyses of 18 - 20 mice for each group.

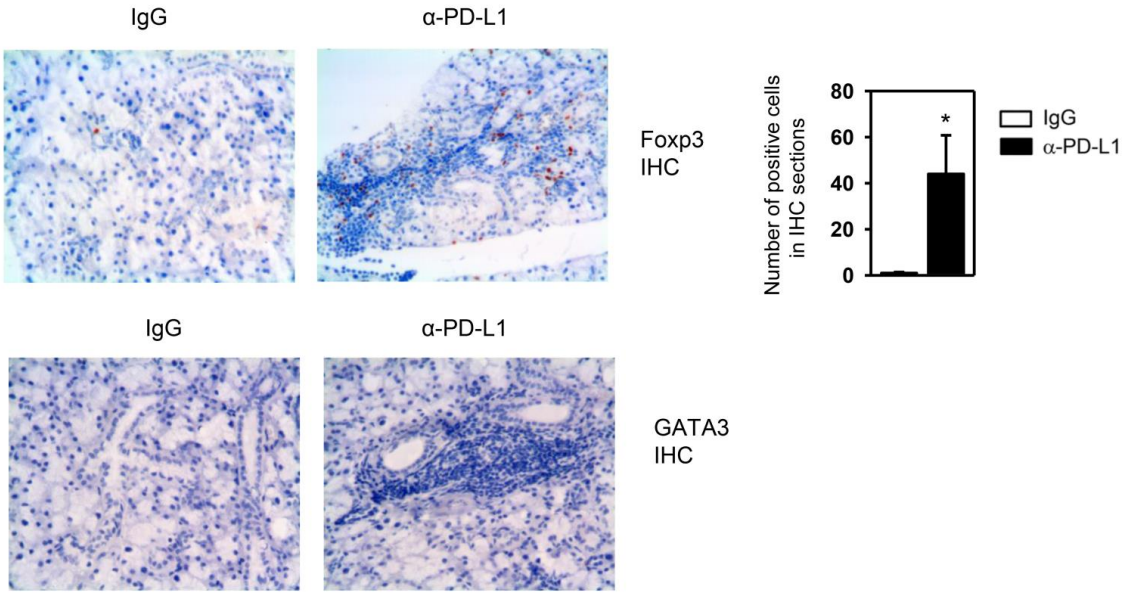
Table S1. Blockade of PD-L1 accelerates leukocyte infiltration of the SMG

Treatment / Mouse	IgG	anti-PD-L1
1	0	0
2	0	12
3	0	0
4	0	6
5	0	6
6	0	1
7	1	0
8	0	0
9	0	4
10	0	0
11	0	0
12	0	0
13	0	1
14	0	7
15	0	0
16	0	0
17	0	4
18	0	5
19		1
20		2
% Mice with focus (foci)	5.5	55
Mean number of foci	0.055	2.45 (P<0.05)

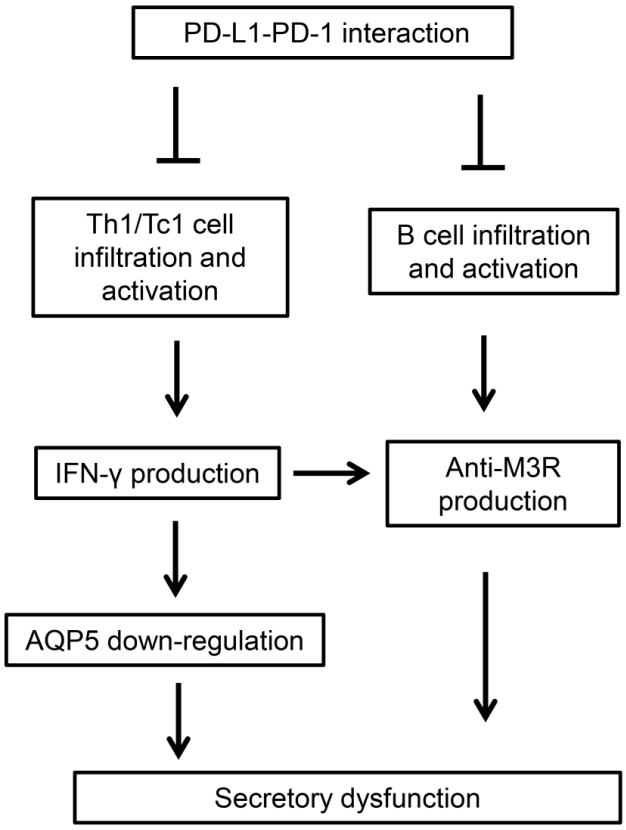
Supplementary Table S1. Blockade of PD-L1 accelerates the appearance of leukocyte foci in the SMG. The number of leukocyte foci, defined as leukocyte aggregates containing at least 50 cells in a 4 mm^2 area of SMG sections, in NOD/ShiLtJ mice treated with anti-PD-L1 or the isotype control IgG.



Supplementary Figure S2. PD-L1 blockade increases the number of CD4 and CD8 T cells in the SMG. Representative flow cytometry of T lymphocyte populations in SMG-infiltrating mononuclear cells. Data are representative of 6-7 mice for each group.



Supplementary Figure S3. PD-L1 blockade increases the number of Tregs in the SMG. Immunohistochemical staining of Foxp3 (upper panels) and GATA3 (lower panels) in the SMG sections of anti-PD-L1- and IgG-treated NOD/ShiLtJ mice. Original magnification: × 400. The number of $Foxp3^+$ cells in the immunohisto-chemistry sections were quantified and shown (upper right panel). Data are representative of or the average of the analyses of 6-7 mice for each group. Error bars represent the SEM. *$P < 0.05$



Supplementary Figure S4. Proposed mechanisms of the regulatory effect of PD-L1 on SS development. The interaction between PD-L1 and PD-1 impedes Th1 and Tc1 recruitment to the salivary glands in part by down-regulating CXCL9. Diminshed IFN-γ production resulting from impaired Th1 and Tc1 responses in turn increases AQP5 expression and reduces anti-M3R autoantibody production to hinder the development of salivary gland hypofunction. In addition, PD-L1-PD-1 interaction also impedes B cell recruitment in part by down-regulating CXCL13, thereby curtailing B cell activation and autoantibody production. Hence, endogenous PD-L1, which is up-regulated during early phase of SS development, impedes the development and delays the onset of this disease in a negative feedback manner.