SUPPLEMENTARY DATA

Supplementary Table 1. Primers used for the synthesis of fluorescently-tagged human ADORA1 and mouse Adora1 fusion proteins.

Fusion protein	**Plasmid**	**Enzyme sites**	**Primer Sequence (5' to 3')**
Human *ADORA1-VAR* -EGFP or –tRFP (cloning)	pEGFP-N1 pTurbo-RFP-N	EcoR1 SacII	For: 5'ATATAGGAATTCGCCACCATGGTGGTGACCCCCCGGAGGGCG 3' Rev: 5' CTATATCCGCGGGTCATCAGGCCTCTCTTC 3'
Mouse *Adora1-Var* -EGFP or –tRFP (cloning)	pEGFP-N1 pTurbo-RFP-N	EcoR1 BamHI	For: 5' ATATAGGAATTCGCCACCATGTTTGGCTGGAACAACCTGAGT3' Rev: 5' CTATATGGATCCTGGTCATCAGCTTTCTCCTC 3'

Supplementary Table 2. Primary and secondary antibodies used for immunohistochemistry.

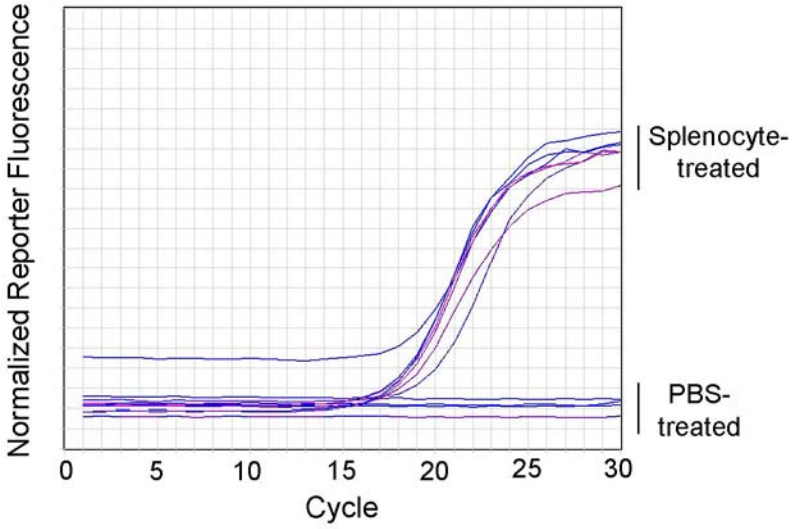
Primary Antibody	**Supplier**	**Dilution**	**Secondary Antibody for detection**
Rabbit anti-Adora1	Abcam #ab82477	1:750	donkey anti-rabbit IgG conjugated to Alexa594 (1:500, Invitrogen)
Goat Anti-Adora2a	Santa Cruz sc-7504	1:100	donkey anti-goat IgG conjugated to Alexa594 (1:500, Invitrogen)
Mouse Anti-Adora2a	Santa Cruz Clone 7F6-G5-A2	1:100	donkey anti-mouse IgG conjugated to Alexa594 (1:500, Invitrogen)
Rabbit Anti-Adora2a	Abcam ab3461	1:100	donkey anti-rabbit IgG conjugated to Alexa594 (1:500, Invitrogen)
Mouse anti-glucagon	R & D systems #MAB1249	1:50	donkey anti-mouse IgG conjugated to Alexa488 (1:500, Invitrogen)
Rabbit Anti-glucagon	Zymed 18-0064	1:100	donkey anti-rabbit IgG conjugated to Alexa488(1:500, Invitrogen)
Rat anti-insulin	R & D systems #MAB1417	1:50	donkey anti-rat IgG conjugated to Alexa488 (1:500, Invitrogen)
Rat anti-CD3	eBiosciences clone 17A2	1:100	donkey anti-rat IgG conjugated to Alexa488 (1:500, Invitrogen)

Supplementary Table 3. Cytokines and chemokines present in the supernatant of activated and non-activated NOD.B10 splenocytes.

	Concentration (pg/ml)	
Sample	Control supernatant	Activated supernatant
IL1A	3.3	150.5
IL1B	<1.22	82.2
IL2	<0.84	42.7
IL3	1.1	80.7
IL4	0.9	33.1
IL5	<1.22	153.5
IL6	1.3	426.5
IL10	1.1	163.3
IL12P40	33.7	27.3
IL12P70	5.8	77.5
IL17	<0.4	175.1
IL13	0.9	169.8
IL23	0.2	4.5
CCL2	7.9	62.9
CCL3	24.4	3230.2
CCL5	581.4	1991.0
CCL7	12.8	85.4
CCL11	<1.22	9.2
CXCL1	1.7	32.1
CXCL10	8.4	451.1
IFNG	36.5	>5835.59
TGFB	1.3	7.8
TNFA	16.4	422.2
GCSF	<1.22	24.7
GMCSF	1.8	314.6
VEGF	3.0	52.1

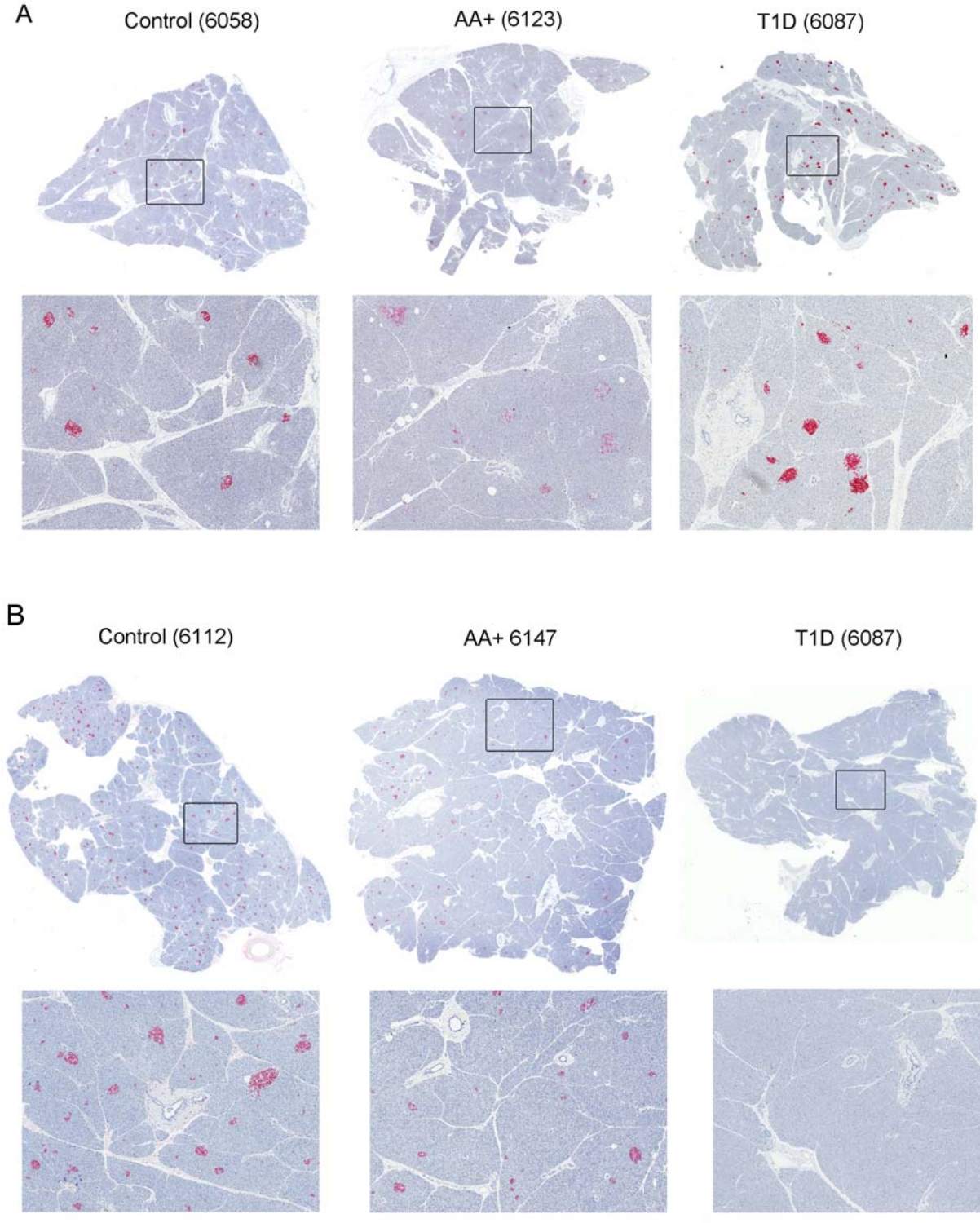
SUPPLEMENTARY DATA

Supplementary Figure 1. PCR Amplification plot showing the expression of the BDC2.5 transgene in pancreas samples of NOD.SCID mice treated with activated splenocytes of NOD.BDC.2.5 mice but not in NOD.SCID mice treated with PBS.



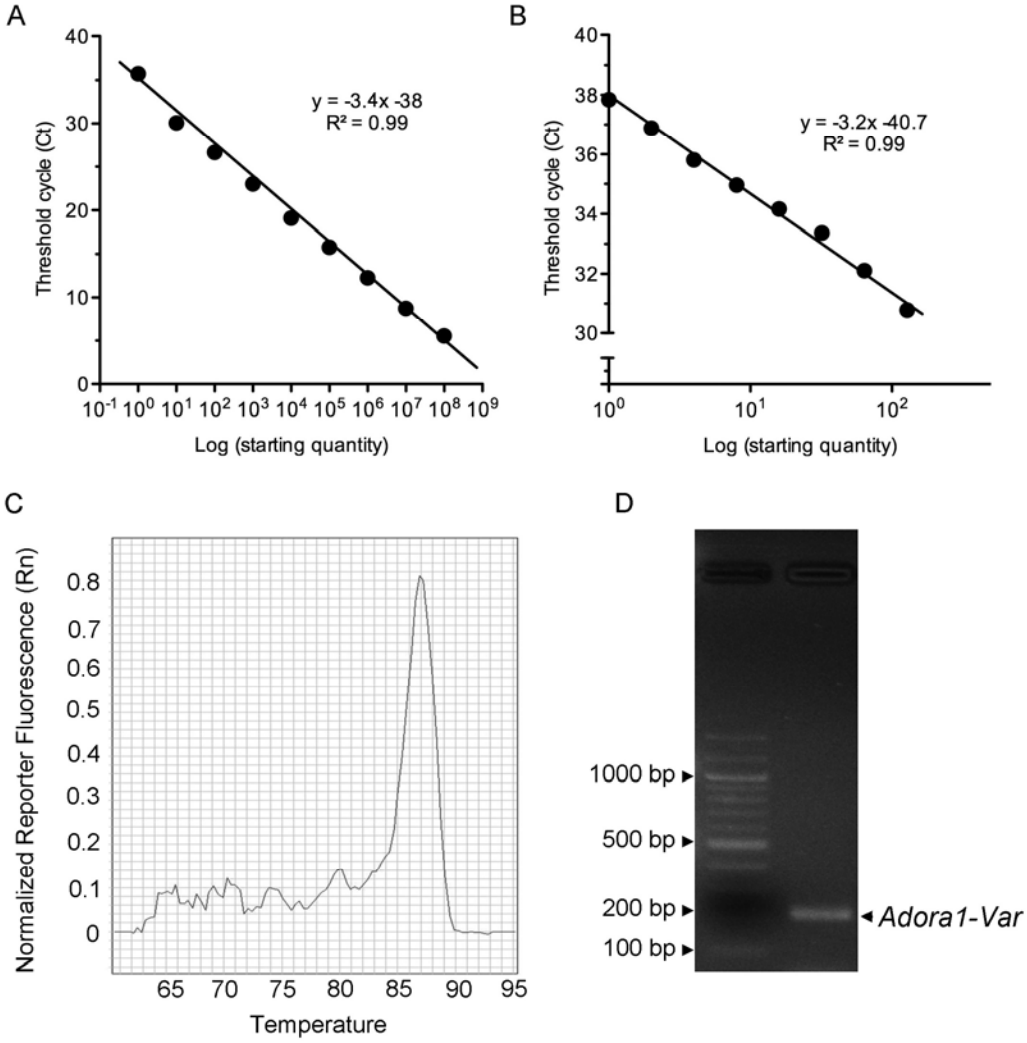
SUPPLEMENTARY DATA

Supplementary Figure 2. Glucagon (A) and insulin (B) staining of pancreata sections representing control, AA+ and T1D patients. High magnification images of marked areas are shown underneath the stained sections. Additional images are available from nPOD (http://www.jdrfnpod.org/online-pathology.php).



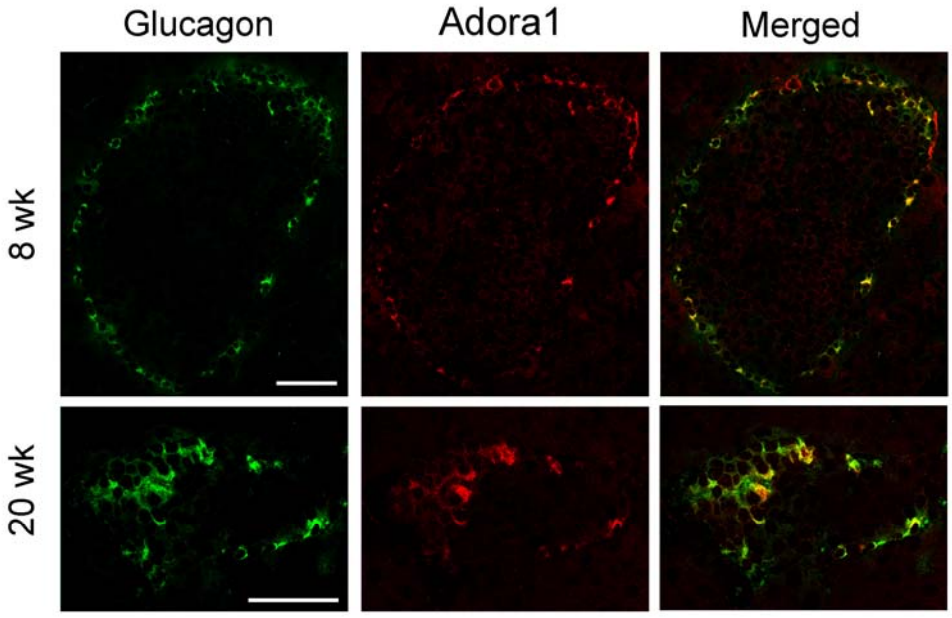
SUPPLEMENTARY DATA

Supplementary Figure 3. Design and assessment of the *Adora1-Var* QPCR SYBR assay. A QPCR assay was developed to amplify only the *Adora1-Var* isoform using an upper primer that spans exon 1 and exon 3 and a lower primer in exon 3. Standard curves were performed and the assay was found to amplify with an efficiency of > 95%, with a linear range of 9 log scale, and to a threshold cycle of 38 (A and B). Melting curve dissociation of the QPCR product (C) and RT-PCR (D) using pooled 12 wk old NOD pancreas tissue as the template show that only one specific product of the expected size is produced by this QPCR assay.



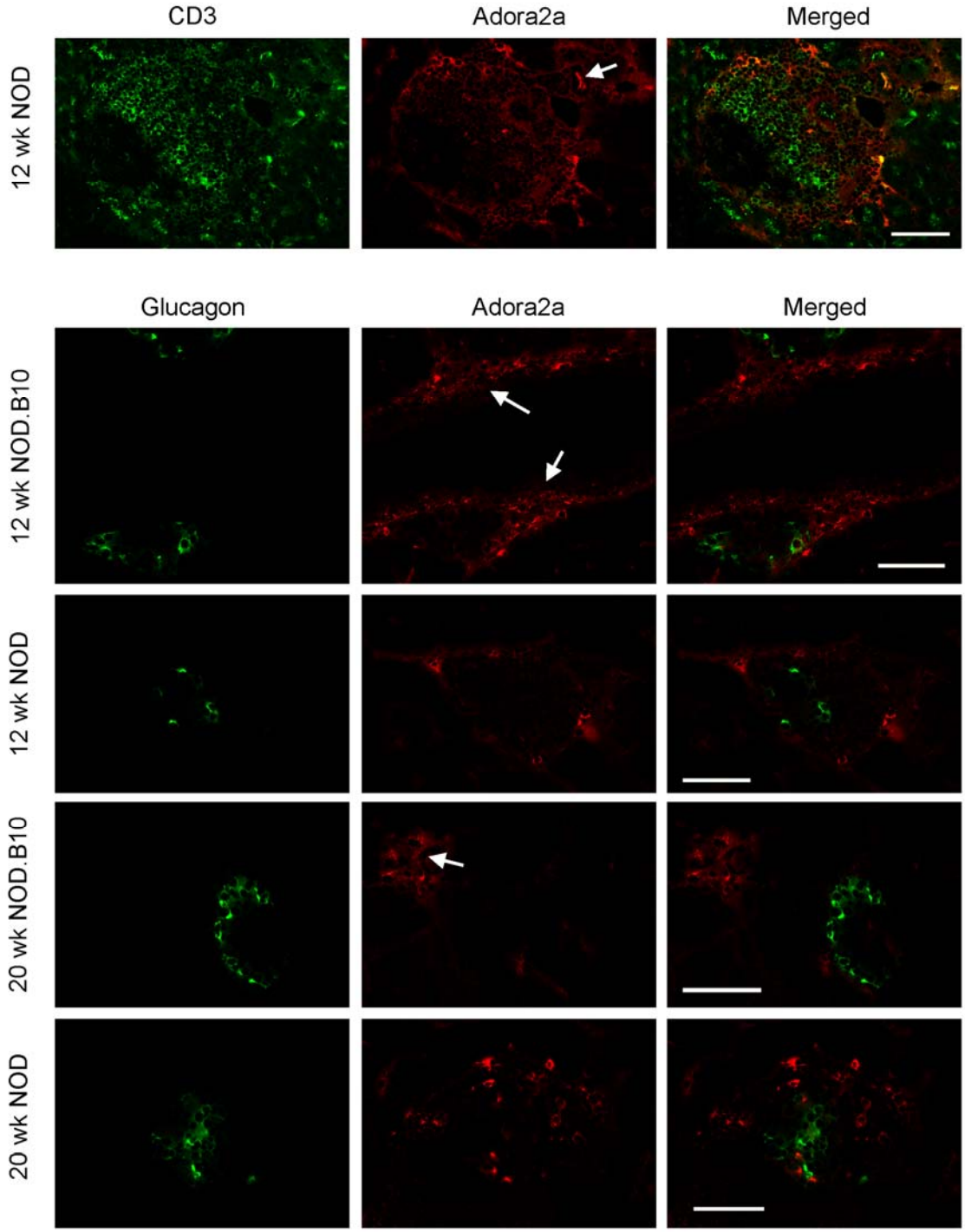
SUPPLEMENTARY DATA

Supplementary Figure 4. Confocal microscopy images showing the expression of Adora1 (red) in the islets of 8 wk and 20 wk old NOD.B10 mice. Adora1 (red) was found to co-localize with glucagon (green) in all alpha cells observed. Scale bars represent 50 μm.



SUPPLEMENTARY DATA

Supplementary Figure 5. Confocal microscopy images showing the expression of Adora2a (red) with CD3 (green) or glucagon (green) in the islets of 12 wk and 20 wk old NOD and NOD.B10 mice. Adora2a was stained using a goat primary antibody from Santa Cruz (sc-7504). Adora2a was expressed in pancreatic ducts (arrows) and in the infiltrate surrounding NOD islets, but not alpha cells. Scale bars represent 50 μm.



SUPPLEMENTARY DATA

Supplementary Figure 6. Confocal microscopy images showing the absence of Adora2a (red) staining in alpha cells. 8 wk old NOD.B10 pancreas tissue sections were stained with Adora2a antibodies purchased from Santa Cruz (clone 7F6-G5-A2, red, top row) or Abcam (ab3461, red, bottom row), and with antibodies against glucagon (green). Scale bars represent 50 μm.