SUPPLEMENTARY DATA

Supplementary Table S1. Demographics and identifiers of donor samples. Isolated β–cells from donors used for transcriptome analyses and tissue sections from donors without diabetes (w/o diabetes) and donors with type 1 diabetes used for immunohistochemical analyses (immunohistochemical and/or immunofluorescence, IHC/IF), RNA-Seq, or flow cytometry (FC) are listed from the EADB, DiViD, VU/UMass (VU/UMass cohort were obtained through a collaborative agreement with Vanderbilt University), or from UMass (from either Prodo Labs or IIDP, RRID codes) cohorts. Partial data previously described for the samples 13226 and 13255 in (23) and for T1D.6 in (3). N/A: not applicable. N/R: not reported. w, m, y: weeks, months, or years of disease duration.

A- Donors without diabetes

Case Code	RRID Source: IIDP	Case Type	Cohort	Use of islets	Age (years)	Sex
PM65/71		w/o diabetes	EADB	IHC/IF	40	M
8579		w/o diabetes	EADB	IHC/IF	7	N/R
330/71		w/o diabetes	EADB	IHC/IF	47	M
184/90		w/o diabetes	EADB	IHC/IF	5	M
17188		w/o diabetes	VU/UMass	RNAseq	10	F
14223		w/o diabetes	VU/UMass	RNAseq, FC	11	M
16149		w/o diabetes	VU/UMass	RNAseq	11	M
16181		w/o diabetes	VU/UMass	RNAseq	12	F
16191		w/o diabetes	VU/UMass	RNAseq	12	F
16278	SAMN0876 9124	w/o diabetes	UMass	RNAseq	15	M
16029		w/o diabetes	VU/UMass	RNAseq	16	M
16338		w/o diabetes	VU/UMass	RNAseq	16	F
17181		w/o diabetes	VU/UMass	RNAseq	16	M
13226		w/o diabetes	UMass	RNAseq	18	M
17103		w/o diabetes	VU/UMass	RNAseq	19	M
15049		w/o diabetes	UMass	RNAseq	23	M
13255	SAMN0878 3913	w/o diabetes	UMass	RNAseq	30	M
18076		w/o diabetes	UMass	FC	33	F
14128	SAMN0877 5087	w/o diabetes	VU/UMass	FC	45	M
14149	SAMN0877 5077	w/o diabetes	VU/UMass	FC	49	F
14219	SAMN0877 5025	w/o diabetes	UMass	FC	52	F

B- Donors with type 1 diabetes

Case Code	Case Type	Cohort	Use of islets	Age (years)	Sex	Time from diagnosis
E560	T1D	EADB	IHC/IF	42	F	18m
E124B	T1D	EADB	IHC/IF	17	M	1w
E375	T1D	EADB	IHC/IF	11	F	1w
11746	T1D	EADB	IHC/IF	6	M	<1w
11713	T1D	EADB	IHC/IF	3	M	3m
Sc76	T1D	EADB	IHC/IF	21	M	3w
Sc115	T1D	EADB	IHC/IF	1	F	<1w
Sc109	T1D	EADB	IHC/IF	20	M	6y
9256	T1D	EADB	IHC/IF	2	M	2w
8566	T1D	EADB	IHC/IF	12	F	3w
DiViD1	T1D	DiViD	IHC/IF	25	F	4w
DiViD2	T1D	DiViD	IHC/IF	24	M	3w
DiViD3	T1D	DiViD	IHC/IF	34	F	9w
DiViD4	T1D	DiViD	IHC/IF	31	M	5w
DiViD5	T1D	DiViD	IHC/IF	24	F	5w
DiViD6	T1D	DiViD	IHC/IF	35	M	5w
nPOD6414	T1D	nPOD	RNAseq IHC/IF/FC	23	M	5m
nPOD6480	T1D	nPOD	FC	17	M	2y
nPOD6367	T1D	nPOD	RNAseq IHC/IF/FC	24	M	2y
nPOD6268	T1D	nPOD	RNAseq	12	F	3y
nPOD6323	T1D	nPOD	FC	22	F	6y
T1D.6	T1D	VU/UMass	RNAseq	20	M	7y
T1D.18	T1D	VU/UMass	FC	24	F	18y
T1D.19	T1D	VU/UMass	FC	54	F	14y
T1D.13	T1D	VU/UMass	FC	56	M	25y

SUPPLEMENTARY DATA

Supplementary Table S2. Antibody details and immunocytochemistry conditions used for staining human pancreas specimens are detailed.
IHC; immunohistochemistry, IF; immunofluorescence.

Primary Antibody	Manufacturer and clone	Antigen Retrieval	Antibody Dilution	Incubation Time with Primary Antibody	Secondary Detection System
CIITA	Santa Cruz C#sc-13556 Mouse monoclonal	10mM citrate pH6.0	1/100	IHC 1h RT IF o/n 4°C	Immunohistochemistry using Dako REAL™ Envision™ Detection System or immunofluorescence using tyramine signal amplification kit (HRP conjugated secondary antibody: 1/100 for 1h, fluorescent tyramide substrate; 1/100 10mins)
HLA-DR	Santa Cruz C#sc-25614 Rabbit polyclonal	10mM citrate pH6.0	1/400	IHC 1h RT IF o/n 4°C	Immunohistochemistry using Dako REAL™ Envision™ Detection System or immunofluorescence using tyramine signal amplification kit (HRP conjugated secondary antibody: 1/100 for 1h, fluorescent tyramide substrate; 1/100 10mins)
HLA-ABC	Abcam C#ab70328 Mouse monoclonal	10mM citrate pH6.0	1/1000	1h at RT	Immunofluorescence staining using anti-mouse IgG (H+L) Alexa Fluor™-conjugated secondary antibodies (1/400 for 1hr)
Insulin	Dako C#A0564 Guinea-pig polyclonal	10mM citrate pH6.0	1/700	1h at RT	Immunofluorescence staining using anti-guinea-pig IgG (H+L) Alexa Fluor™ -conjugated secondary antibodies (1/400 for 1hr)
CD45	Dako C#M0701 Mouse monoclonal	10mM citrate pH6.0	1/1000	1h at RT	Immunofluorescence using tyramine signal amplification kit (HRP conjugated secondary antibody: 1/100 for 1h, fluorescent tyramide substrate; 1/100 10mins)
CD68	Dako C#M0876 Mouse monoclonal	10mM citrate pH6.0	1/400	1h at RT	Immunofluorescence staining using anti-mouse IgG (H+L) Alexa Fluor™-conjugated secondary antibodies (1/400 for 1hr)
CD80	Abcam C#ab86473 Mouse monoclonal	10mM citrate pH6.0	1/500	o/n at 4°C	Immunohistochemistry using Dako REAL™ Envision™ Detection System or immunofluorescence staining using anti-mouse IgG (H+L) Alexa Fluor™-conjugated secondary antibodies (1/400 for 1hr)
CD86	Abcam C#ab53004 Rabbit monoclonal	10mM Tris, 1mM EDTA pH9.0	1/300	o/n at 4°C	Immunohistochemistry using Dako REAL™ Envision™ Detection System
VP1	Dako C#M7064 Mouse monoclonal	10mM citrate pH6.0	1/2000	o/n at 4°C	Immunofluorescence staining using anti-mouse IgG (H+L) Alexa Fluor™-conjugated secondary antibodies (1/400 for 1hr)

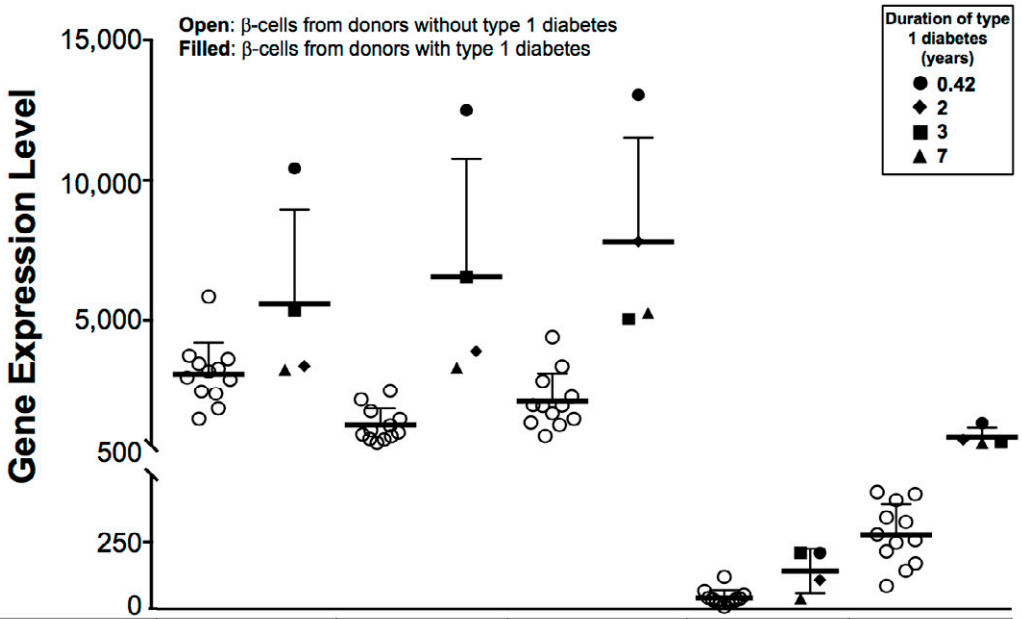
SUPPLEMENTARY DATA

Supplementary Table S3. Case by case examination of CIITA and Class II staining. Green colour indicates cases in which CIITA was absent, whereas red colour demarks cases containing CIITA positive islets. ND: not determined.

Case Code	Case Type	Islets + or – for CIITA	Number of Class II$^+$ islets
PM65/71	without diabetes	-	0
8579	without diabetes	-	0
330/71	without diabetes	-	0
184/90	without diabetes	-	0
Sc109	with type 1 diabetes	-	0
9256	with type 1 diabetes	-	0
8566	with type 1 diabetes	-	1
E375	with type 1 diabetes	-	ND
E560	with type 1 diabetes	+	16
E124B	with type 1 diabetes	+	84
Sc76	with type 1 diabetes	+	113
SC115	with type 1 diabetes	+	ND
11746	with type 1 diabetes	+	22
11713	with type 1 diabetes	+	16
DiViD3	with type 1 diabetes	+	26
DiViD6	with type 1 diabetes	+	2

SUPPLEMENTARY DATA

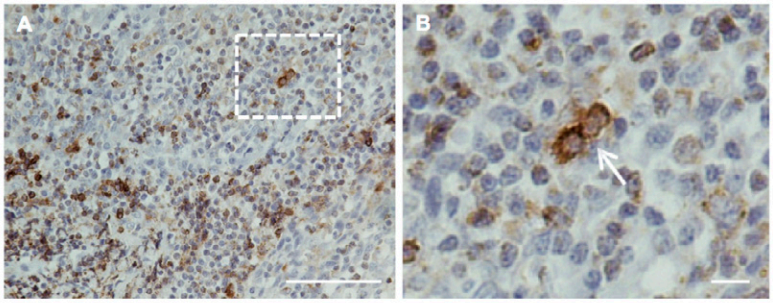
Supplementary Figure S1. Bulk β cell population, as sorted by intracellular insulin staining, isolated from donors with type 1 diabetes shows increased HLA Class I mRNA expression. β cell gene expression (shown as expected counts of raw values calculated using RSEM) in donors without diabetes (N=12, open circles) and with type 1 diabetes (N=4, filled shapes, circle: nPOD 6414, diamond: nPOD 6367, square: nPOD 6268, triangle: T1D.6) are shown on the ordinate for 4 HLA Class I mRNA species (*HLA-A, HLA-B, HLA-C, and HLA-F*) and the HLA Class I transactivator, *NLRC5*. The fold change increase in expression from donors with type 1 diabetes and the adjusted *p* value are shown below the abscissa. Bars indicate the mean values +SEM.



Gene Name	HLA-A	HLA-B	HLA-C	HLA-F	NLRC5
T1D/ND fold Δ	1.9	5.6	4.0	3.5	3.1
Adj. *p* value	0.09	6.3E-08	1.3E-05	0.02	9.4E-05

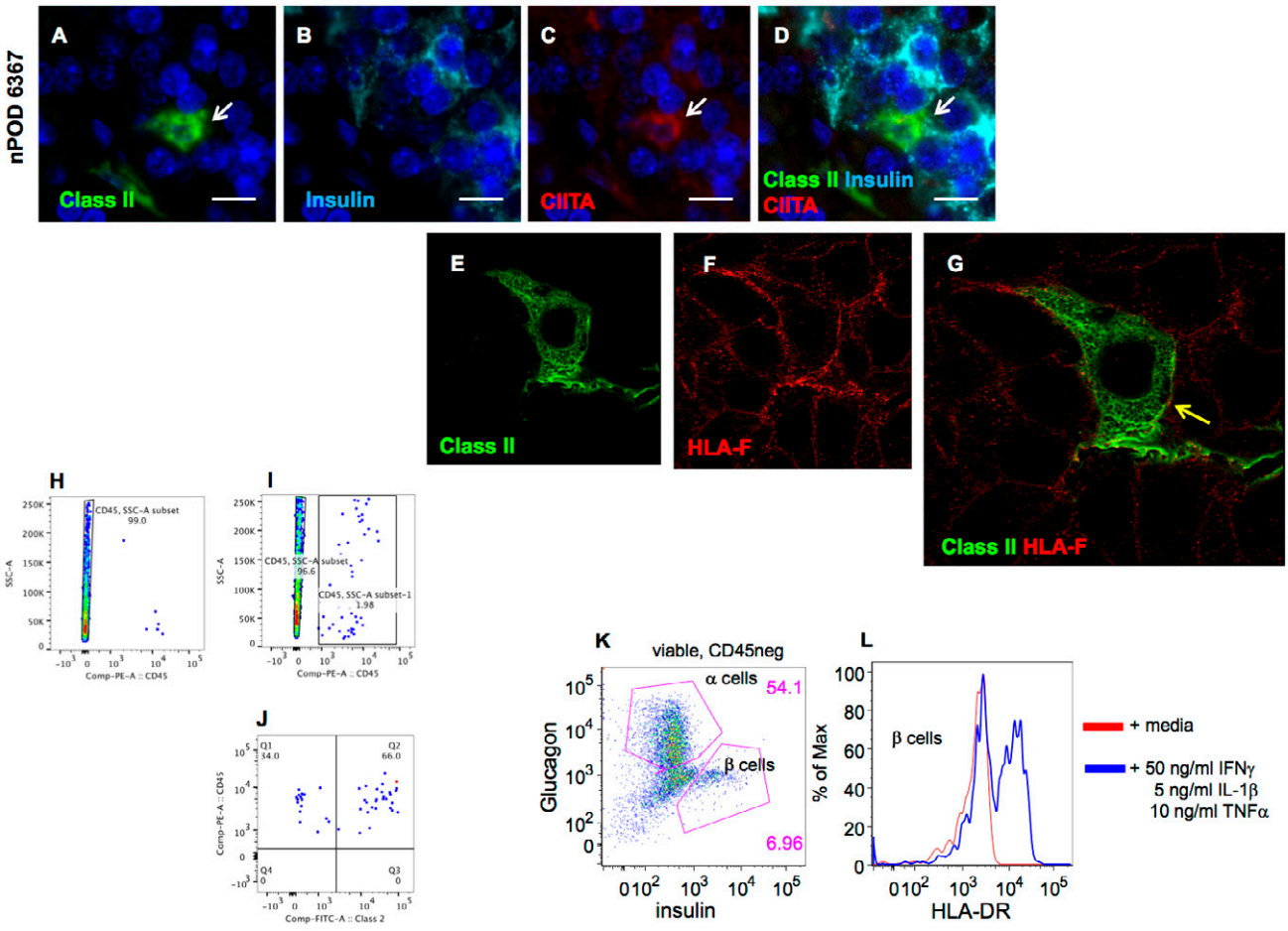
SUPPLEMENTARY DATA

Supplementary Figure S2. CIITA is expressed in the tonsil, and has predominantly a cytosolic localisation. (A) Representative image of tonsil tissue from a single donor stained using an immunoperoxidase approach probed with an antibody raised against CIITA. An enlargement of the highlighted area (dotted white box) is presented (B). The white arrow indicates cells with high levels of cytoplasmic, but low nuclear CIITA. Images were captured at x400 magnification; scale bar (A) 50µm, (B) 20µm.



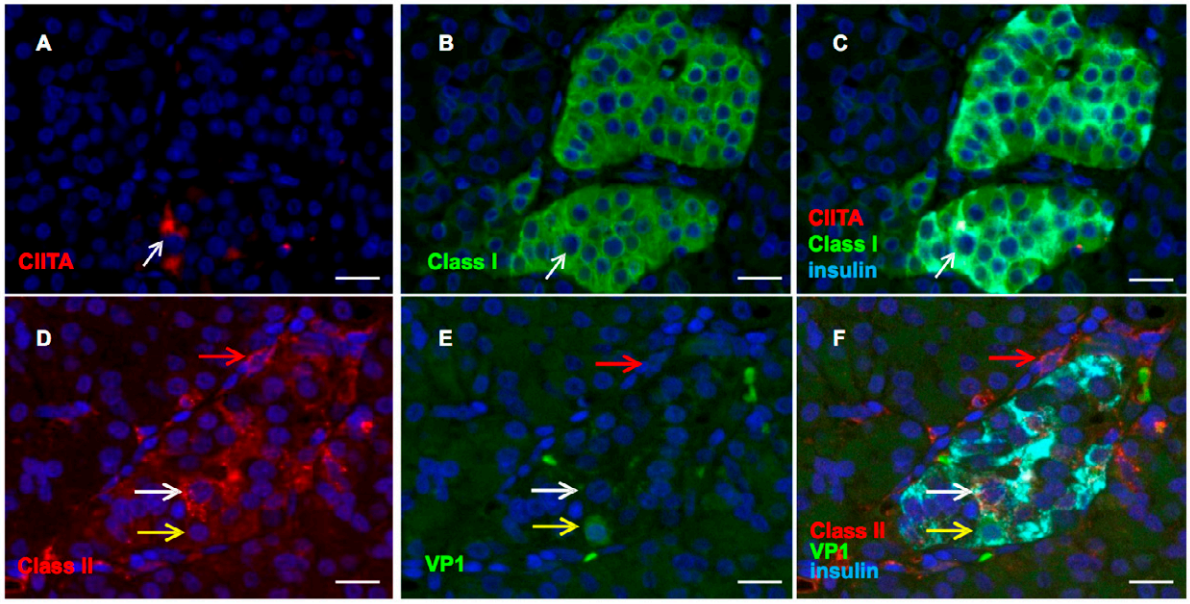
SUPPLEMENTARY DATA

Supplementary Figure S3. Surface expression of HLA Class II on β-cells. From a single donor with T1D (nPOD 6367) from which RNA transcriptome data is shown in Figs. 1 and 2 and Supplementary Fig. 1, pancreas tissue was stained with antibodies to HLA Class II (green, A, D), insulin (light blue, B, D), and CIITA (red, C, D). Merged images are shown (D). White arrows denote a Class II^{+}insulin^{-} $CIITA^{+}$ cell. Images captured at x400 magnification; scale bar, 10µm. A high-resolution image of the same cell shown in Fig. 5 (Fig. 5 A-E) was captured using deconvolution software at x630 magnification with optical zoom (E-G). The yellow arrows indicate areas in which Class II and HLA-F co-localise. An entire *z*-stack of this region is presented in the **Supplementary Video**. $CD45^{+}$ cells from islets are detected by flow cytometry. As described in Fig. 5, islets were dispersed and surface stained for CD45 and HLA-DR. Examples of the $CD45^{-}$ and $CD45^{+}$ populations within two islet samples [nPOD6367 (an islet sample analyzed for transcriptome, Figs. 1 and 2) (H) and T1D.19 (I)] are shown. A representative profile of surface $CD45^{+}$ cells for HLA-DR staining is shown for T1D.19 (J). $CD45^{-}$ cells were then analyzed for surface HLA-DR and intracellular insulin as shown in Fig. 5. Representative plots are shown for islets for induction of surface HLA Class II expression on islet β-cells by pro-inflammatory cytokines (K and L). 2000 IEQ from a control, non-diabetic donor (18076) were treated +/- proinflammatory cytokines as indicated in the figure for 48 hours. Islets were collected, dispersed with enzyme, stained with viability dye, blocked, surface stained for CD45 and HLA-DR (L243), washed, fixed/permeabilized and stained intracellularly for glucagon and insulin as described. Approximately 50% of $insulin^{+}$ β-cells upregulated surface HLA-DR upon treatment with proinflammatory cytokines. Similar results were seen with 3 other islet isolations.



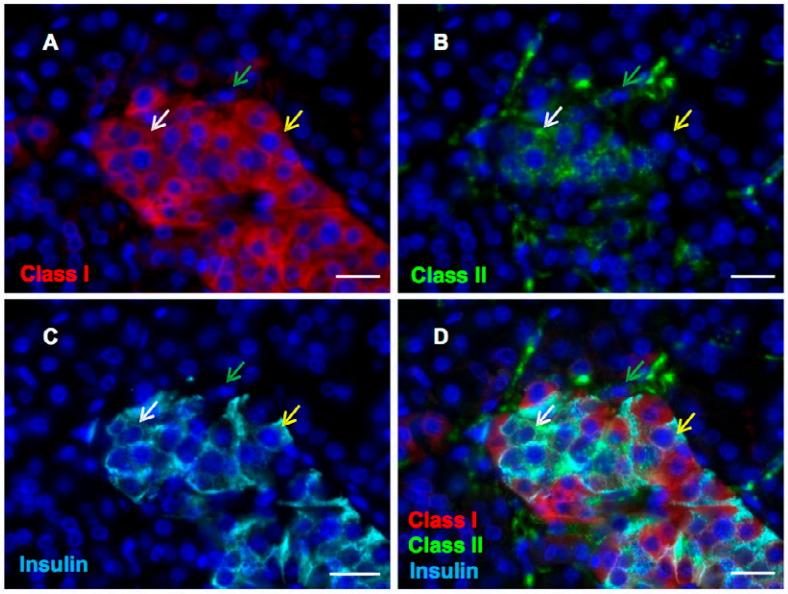
SUPPLEMENTARY DATA

Supplementary Figure S4. CIITA expression does not correlate with the presence of Class I or enterovirus VP1, in the islets of donors with type 1 diabetes. Representative images from pancreas sections of individuals with type 1 diabetes stained with antisera raised against CIITA (A, red), Class I (B, green), Class II (D, red), VP1 (E, green) and insulin (C, F, light blue). Merged images are presented (C, F). White arrows in the upper panel (A-C) point to examples of CIITA immunopositive cells. White arrows (D-F, lower panels) indicate Class II positive cells that are negative for VP1. Yellow arrows show a VP1 positive cell which is immunonegative for Class II. Red arrows indicate an example of an immune cell that stains positive for HLA Class II in the islet periphery. Images captured at x400 magnification; scale bar 20μm.



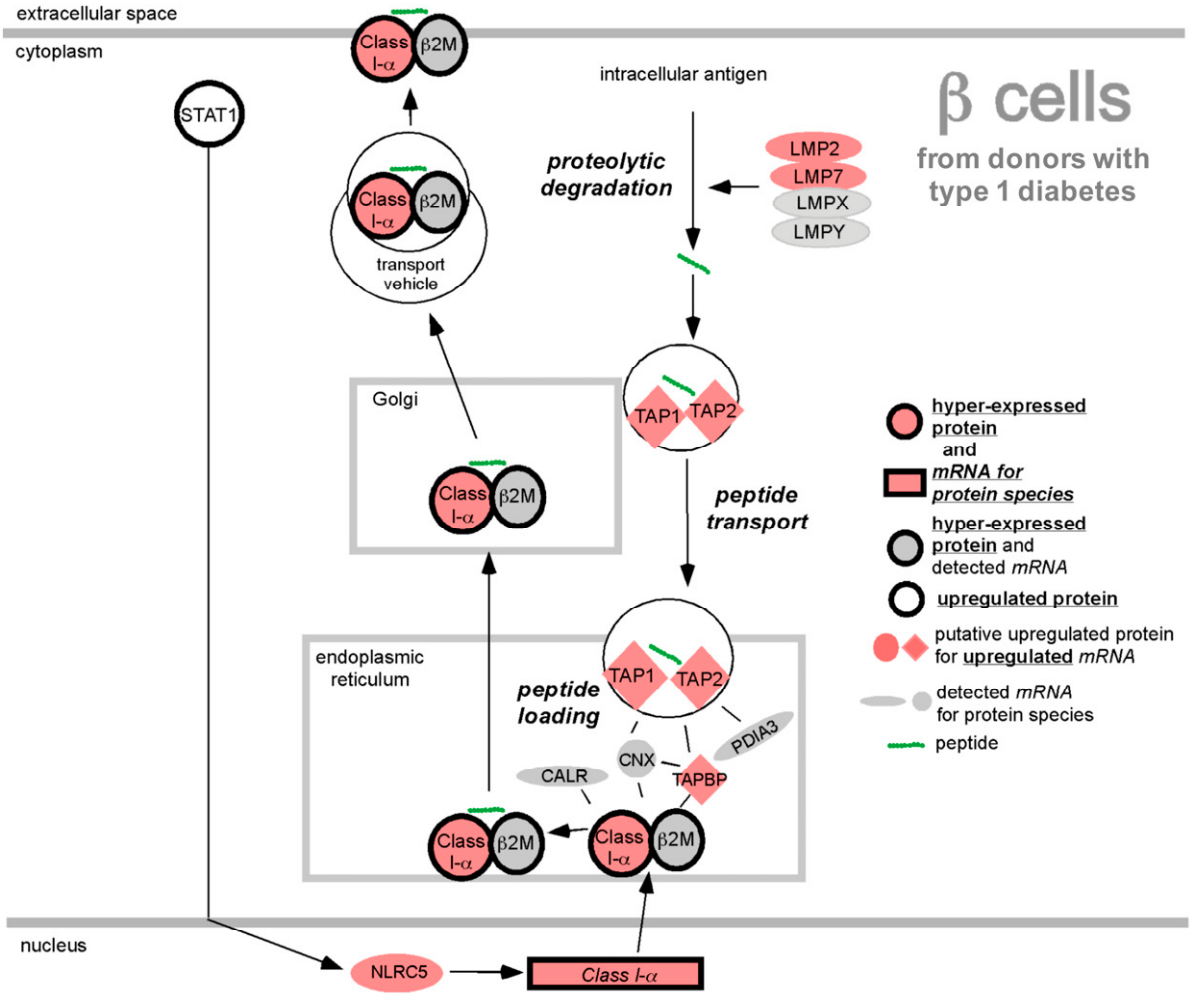
SUPPLEMENTARY DATA

Supplementary Figure S5. Class I and Class II expression do not correlate within islets. Representative immunofluorescence image of a pancreas section from a donor with type 1 diabetes stained for Class I (A, D: red), Class II (B, D: green) and insulin (C, D: light blue). The white arrow indicates an islet cell co-expressing Class I and Class II, whereas the yellow arrow marks a cell within the islet that is positive for Class I, but negative for Class II. Islet associated endothelial cells expressing Class II were also observed (green arrow). Images captured at x400 magnification; scale bars, 20µm.



SUPPLEMENTARY DATA

Supplementary Figure S6. Upregulated protein and mRNA species in β cells from donors with type 1 diabetes from the HLA Class I processing and presentation pathway. The observed upregulated proteins and mRNA species from the HLA Class I processing and presentation pathway in β cells from donors with type 1 diabetes are displayed according to their intracellular locations and with their functions. Depicted by symbols with black borders are upregulated proteins (red circles) and mRNA species (red squares), while putative upregulated proteins identified by upregulated mRNA species are shown with red ovals, diamonds or elongated ovals, and gray ovals depict mRNA species that were detected, but not upregulated in the islets of donors with type 1 diabetes. A detected, upregulated protein, STAT1 (9), but without mRNA detection, is indicated by an empty circle with a black border.



SUPPLEMENTARY DATA

Supplementary Video. *z*-stack of HLA Class II and HLA -F co-localise on the surface of an islet cell in a donor with type 1 diabetes. A high-resolution z-stack of the islet shown in Fig. 5 A-E and Supplementary Fig. 3A-D is presented. Sections from a donor with type 1 diabetes (E560) were stained with antisera against Class II (green) and HLA-F (red), with areas of co-localisation appearing yellow. This islet was imaged using deconvolution software at x630 magnification with additional optical zoom.