Type 1 diabetes mellitus as a disease of the β-cell (do not blame the immune system?)

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Abstract | Type 1 diabetes mellitus is believed to result from destruction of the insulin-producing β-cells in pancreatic islets that is mediated by autoimmune mechanisms. The classic view is that autoreactive T cells mistakenly destroy healthy (‘innocent’) β-cells. We propose an alternative view in which the β-cell is the key contributor to the disease. By their nature and function, β-cells are prone to biosynthetic stress with limited measures for self-defence. β-Cell stress provokes an immune attack that has considerable negative effects on the source of a vital hormone. This view would explain why immunotherapy at best delays progression of type 1 diabetes mellitus and points to opportunities to use therapies that revitalize β-cells, in combination with immune intervention strategies, to reverse the disease. We present the case that dysfunction occurs in both the immune system and β-cells, which provokes further dysfunction, and present the evidence leading to the consensus that islet autoimmunity is an essential component in the pathogenesis of type 1 diabetes mellitus. Next, we build the case for the β-cell as the trigger of an autoimmune response, supported by analogies in cancer and antitumour immunity. Finally, we synthesize a model (‘connecting the dots’) in which both β-cell stress and islet autoimmunity can be harnessed as targets for intervention strategies.

For several decades, type 1 diabetes mellitus (T1DM) was believed to be a T-cell-mediated autoimmune disease1–3. This notion still holds, but several observations in the past few years point to a role of β-cells that goes beyond being a non-provoking victim of an autoimmune attack4–6. The lack of durable effects of immune-suppressive intervention therapies, islet autoimmunity occurring without the development of T1DM, a remarkably low rate of insulitis at diagnosis and the unexpectedly high proportion of β-cells that persist (although they do not always function) after the diagnosis of T1DM prompted a revision of our take on the pathogenesis of type 1 diabetes mellitus. Next, we build the case for the β-cell as the trigger of an autoimmune response, supported by analogies in cancer and antitumour immunity. Finally, we synthesize a model (‘connecting the dots’) in which both β-cell stress and islet autoimmunity can be harnessed as targets for intervention strategies.

T1DM as an autoimmune disease

A connection between the immune system and T1DM was first suggested in 1973, when HLA antigens were found to be associated with insulin-dependent diabetes mellitus but not with insulin-independent diabetes mellitus10. Since then, genome-wide association studies have confirmed that HLA genes account for up to 50% of the genetic risk of T1DM (in particular HLA class II loci), which suggests that the selective presentation of specific autoantigen peptides is involved in the pathogenesis of T1DM1–3. Meta-analyses have also linked non-HLA high-risk polymorphisms within INS-VNTR (variable number of tandem repeats), PTPN22, CTLA4 and IL2RA with a reduction in central and peripheral immune tolerance to self and increased T-cell activation and proliferation14–17, which emphasizes the participation of the immune system in the development of T1DM18.

During the development of T1DM, seroconversion of islet autoantibodies to insulin, glutamate decarboxylase, insulinoma antigen 2 or zinc transporter 8 represents the first notable sign of autoimmunity and their combined presence in serum remains the best predictor for both loss of immune tolerance (that is, induction of autoimmunity) and clinical manifestation of T1DM, albeit that their role in β-cell destruction remains unclear6,20. During disease progression, immune cells that infiltrate the pancreas and target insulin-producing cells create an inflammatory environment characteristic of insulitis that triggers and accelerates T1DM development by increasing exposure of islet antigens presented by HLA class I molecules to the immune system21–23 (Box 1).
The presence of islet-specific autoreactive CD4+ and CD8+ T cells in peripheral blood, pancreatic draining lymph nodes and insulitic lesions provided evidence for T1DM as an autoimmune disease, where an impaired thymic education was responsible for the immune attack directed against self-proteins of insulin-producing cells. Yet, despite their importance in T1DM pathology, the frequency of these autoreactive cells in peripheral blood is low and quite similar between patients with T1DM and healthy individuals. Although the presence of naive autoreactive cells in healthy individuals indicates that these cells are part of the normal T cell repertoire and that ‘we are all autoimmune’, the increased frequency of CD8+ T cells (in particular resident memory cells) in the pancreata of patients with T1DM compared with those of control individuals implies a differential peripheral activation and/or regulation in patients with T1DM. Indeed, regulatory T (Treg) cells, which have an important role in repressing these autoreactive T cells in healthy individuals, show a similar frequency in control individuals and in patients with T1DM but with a reduced suppressive capacity in patients with T1DM.

Intriguingly, islet autoreactive T cells have unusual characteristics compared with T cells that protect us from cancer and infection, such as a fairly low epitope binding affinity for HLA, low T cell receptor (TCR) avidity for HLA–epitope complexes, tilted or even reversed docking of the TCR on the HLA–peptide complex, suboptimal synapse formation in the interface between T cells and antigen-presenting cells or target cells and abnormal expression of signalling molecules that might have contributed to incomplete thymic education and thymic selection. Consistently, patients with cancer who are treated with immune checkpoint inhibitors (that is, anti-PD1, anti-PDL1 or anti-CTLA4 therapies) aimed at reducing immune regulation and initiating an immune response against the tumour tissue are at risk of developing adverse effects, including autoimmunity, probably due to loss of immune regulation combined with activation of naive autoreactive T cells.

In the past couple of years, it has been argued that autoimmune diabetes mellitus induced by immune checkpoint inhibition and other types of T1DM relate to the acute manifestation and short prodromal phase of the former, leading to fairly frequent and severe ketoacidosis and paucity of autoantibodies at diagnosis. After all, islet autoantibodies take time to be generated, following T cell activation. The demonstration that T1DM can be transferred with bone marrow from a donor with T1DM to an immune-suppressed recipient who did not have T1DM only when T cells are not depleted, underscores the relevance of T cells in the immunopathogenesis of T1DM. Furthermore, pancreatitis rarely leads to T1DM, even in patients with an increased genetic risk of T1DM, which in turn emphasizes that loss of immune tolerance and induction of islet autoimmunity are a prerequisite for development of the disease. This finding is supported by the rapid recurrence of islet autoimmunity, selective β-cell destruction and T1DM following partial pancreas transplantation from non-diabetic donors to their monozygotic twins with T1DM, as well as islet autoimmunity predicting failure or poor prognosis of allogenic islet transplantation and autologous bone marrow transplantation. Finally, the fact that, until now, immunotherapeutic strategies have shown temporal efficacy in delaying disease progression implicates the immune system in T1DM pathology.

**Inconsistencies in the role of T cells**

A different stand on a role of T cells in the pathogenesis of T1DM can easily be defended (Box 2). Islet autoreactive T cells are common in the healthy population, and nine out of ten individuals with islet autoantibodies will never develop T1DM. Most patients with T1DM have immune regulation that is indistinguishable from that of healthy individuals, and over 99% of patients with cancer who are treated with immune checkpoint inhibitors do not develop T1DM. Furthermore, some patients with T1DM present with negligible T cell autoimmunity. Moreover, induction of autoimmune diabetes mellitus in mice by vaccination with islet autoantigens is very difficult, if not impossible. Even when transduction of human islet autoreactive TCRs in humanized mice leads to high frequencies of T cell autoimmunity to islets, no diabetes mellitus was induced. In addition, thus far, progression of T1DM has not been found to accelerate after patients with T1DM are injected with islet autoantigens. Of note, HLA upregulation as an early sign of islet distress frequently occurs without inflammation, even if β-cells are still present, while insulitis is a rare feature in individuals who have islet autoantibodies but not T1DM. Furthermore, immunotherapies in T1DM have not yet shown a durable effect on disease progression. These inconsistencies in our understanding of the critical role of islet autoimmunity, and T cells in particular, require reconciliation.

**T1DM as a disease of β-cells**

Given that autoreactive T cells are part of a normal T cell repertoire, it is implausible that the disease is entirely the result of dysfunctional immune cells; rather, peripheral activation of the immune system is required locally in the targeted tissue. A role for β-cells in their own...
Molecular fragility
The extreme sensitivity of β-cells to stress, inflammation and apoptosis.

demise was first proposed by Bottazzo. Different triggers that might lead β-cells to provoke an immune response have been proposed, ranging from the size of the pancreas and β-cell mass to viral infection and metabolic stress. Indeed, the pancreata of patients with T1DM are smaller than those from unrelated control individuals (Box 3). Yet, at-risk individuals and patients with T1DM have pancreata of similar sizes, and no data at this time suggest that the pancreas decreases in size with disease progression. Obviously, less β-cell mass might equal less β-cell functional capacity and increased pressure on β-cells to cope with glycaemic control. In addition to metabolic stress, viral infections or intestinal inflammatory agents ‘leaking’ into the pancreas might create a pro-inflammatory environment. β-Cells are exposed to viral infection as they express specific receptors and adhesion molecules. Indeed, the presence of a coxsackievirus and adenovirus receptor (CAR) that is unique to β-cells, found in the insulin-containing granules, might leave β-cells vulnerable to viral infection during insulin secretion, as illustrated by studies correlating enteroviral infection by coxsackievirus B4 with islet autoimmunity (but not T1DM). Viral infection might be a risk factor in, at best, a small minority of patients with T1DM. A viral contribution to the development of T1DM is certainly not limited to coxsackievirus; for example, rotavirus and cytomegalovirus have also been implicated.

Role of diet and microbiota. Similarly, dysbiosis of the gastrointestinal tract (a ‘gut storm’) provoked by changes in intestinal microbiota and an increased Bacteroidetes to Firmicutes ratio has been correlated with seroconversion and onset of T1DM (the pancreas being an intestinal organ). Microbiota shape peripheral immune tolerance, modulating both migration and differentiation of immune cells to maintain intestinal homeostasis; furthermore, local inflammation is limited through short-chain fatty acids (SCFAs) generated by resident gut bacteria from fermentation of non-digestible carbohydrates. SCFAs have a direct effect on T cell subsets via histone deacetylase inhibition and activation of mTOR and STAT3 signalling, leading to an increased proportion of regulatory T cells that produce IL-10 and express FOXP3. In addition, SCFAs can exert their anti-inflammatory effect on neutrophils, macrophages and plasmacytoid dendritic cells via antimicrobial peptides produced by innate lymphoid cells or by β-cells themselves. Strong evidence from studies in mice demonstrates the protective role of these cationic antimicrobial peptides against autoimmune diabetes mellitus, and SCFAs have been used to prevent cytokine-induced cell death of human islet cells and to improve β-cell function. Despite these positive findings, a first-in-human crossover clinical trial conducted in patients with longstanding T1DM (mean diabetes mellitus duration of 8 years) that aimed to restore epithelial integrity by short-term oral butyrate supplementation failed to show improvement in adaptive and innate immune system parameters.

Diet can also affect the microbiome favourably or unfavourably with regard to the predisposition for developing T1DM. A low gluten diet can induce favourable changes in the intestinal microbiome of healthy adults, while low maternal gluten intake during pregnancy shows a remarkable correlation with reduced development of T1DM in the offspring. While it remains to be established whether patients at risk of T1DM or patients newly diagnosed with T1DM would benefit from a low gluten diet, these data might also suggest that once initiated, local inflammation in the pancreas is sufficient to drive disease progression, given the inherent molecular fragility of β-cells (the ‘domino effect’).

Genetic risk. Genetic risk, determined by certain genetic variants in the gene encoding insulin (INS), might affect β-cell function and glycaemic control. Early studies suggest that protective variants of INS result in increased INS expression in the thymus, thereby increasing the probability that the immune system will be educated to avoid immune reactivity to insulin; however, differences in INS activity in pancreatic islets have also been linked to these genetic polymorphisms, as well as effects on β-cell function and resilience. Other genetic variants associated with increased risk of T1DM might affect β-cell health, vitality and self-defence. β-Cell mass and function might have been declining for more than 10 years before clinical manifestation of

Box 1 | Evidence supporting a role for T cells in T1DM pathogenesis

- Insulitis
- HLA association
- HLA class I upregulation in inflamed islets
- Autoreactive CD8+ T cells in insulitis
- Autoreactive CD4+ T cells in insulitis
- Recurrence of islet autoimmunity, insulitis and type 1 diabetes mellitus (T1DM) after twin pancreas graft into T1DM recipient
- Adoptive transfer of T1DM after bone marrow transplantation not depleted for T cells from a donor with T1DM
- T cell-dependent islet autoantibodies
- Recurrent islet autoimmunity and chronic progressive loss of islet allografts transplanted into patients with T1DM
- Recurrent islet autoimmunity in patients with T1DM who relapse after pancreas transplantation
- No islet autoimmunity in pancreas or T1DM in chronic pancreatitis (even with high-risk HLA)
- Higher islet autoreactivity of T cells in patients with T1DM than in healthy individuals
- Therapeutic effect of anti-T cell immune-suppressive therapy
- Complete and durable remission after autologous bone marrow transplantation in patients with new-onset T1DM
- Therapeutic effect of co-stimulation blockade of progression of T1DM
- Development of T1DM after co-stimulation blockade in cancer
- Development of insulitis and selective loss of β-cells in humanized mice carrying insulin-specific TCR
- Phenotypical and functional features in islet autoreactive T cells
- Functional abnormalities in regulatory T cells in T1DM
- Genetic risk associated with polymorphisms in genes involved in immune regulation (PTPN22, IL2R and CTLA4)
- Genetic defects of immune regulation genes causing T1DM (FOXP3 and AIRE)
- Genetic risk associated with INS gene associated with thymic education and central tolerance
Fig. 1 | Immunoregulation in health, and immune dysregulation in cancer, T1DM or immunotherapy. a | In healthy individuals, β-cells are protected from autoimmune β-cell destruction by immune regulation exerted by regulatory T (T<sub>reg</sub>) cells and PD1 ligation. b | While advantageous in preventing autoimmunity, T<sub>reg</sub> cells impede antitumour immunity. c | In type 1 diabetes mellitus (T1DM), insufficient immune regulation can result in an autoimmune response by autoreactive T cells, particularly if these cells are provoked by β-cells. d | The response in T1DM resembles effective antitumour immunity as a result of immunotherapeutic blockade of PD1 or its ligand PDL1 that otherwise keep autoimmune responses in check. In addition to resulting in antitumour immunity, other immune and autoimmune responses might be triggered, including those against pancreatic islets. T1DM is a serious adverse effect of tumour immunotherapy. GRZB, granzyme B; TCR, T cell receptor.
T1DM, adding to increasing metabolic stress in β-cells and vulnerability to autoimmune insults.\(^{101,102}\)

**Insights from human studies of insulitis.** Our understanding of the effect of insulitis on β-cells has exploded with the increased access to pancreata from donors with diabetes mellitus (Network for Pancreatic Organ Donors with Diabetes), even though the condition of the donors (factors such as cause of death, presence of brain death, stay and treatment in an intensive care unit, cold ischaemia, ketoacidosis, injury and stress) might influence some of the observations made on the pancreata, in terms of the effects of stress.\(^{20,21}\) An increased expression of markers specific for the unfolded protein response to stress in β-cells during insulitis suggests that adaptive mechanisms are engaged to help β-cells deal with the environmental pressure.\(^{22}\) Stressed β-cells have a reduced overall translation rate, initiate degradation of proteins accumulated in the endoplasmic reticulum (ER), increase the translation rate of chaperones and promote autophagy to return to cellular homeostasis.\(^{104–106}\) However, the extraordinary capacities of β-cells to produce up to 1 million insulin molecules per minute and to increase production in excess of 50-fold in response to glucose, combined with low expression of superoxide dismutase and anti-apoptotic factor BCL-2 make β-cells poorly equipped to survive the inflammatory milieu. β-Cells are more sensitive than α-cells to environmental stimuli, as illustrated by studies conducted on islets challenged by metabolic stress mimicking pathophysiological conditions in type 2 diabetes (T2DM).\(^{105,106}\) In addition to the cytotoxic function, activation of the ER stress sensors is known to lead to a cascade of events promoting direct apoptosis via activation of the IRF–STAT1 pathway, necroptosis via activation of TNFR1–RIP1 and necrosis by increased production of reactive oxygen species as well as induction of a form of β-cell senescence.\(^{106,108,111}\) These mechanisms might participate in the amplification of inflammation and destruction of β-cells by starting communication with other endocrine cells and resident immune cells. Stress-induced senescence drives β-cells to a senescence-associated secretory phenotype, which is correlated with intra-islet infiltration of CD45+ immune cells in patients with T1DM.\(^{103}\)

Studies of human insulitis have revealed that ‘danger signals’ from stressed β-cells might precede insulitis. Among these signals, hyper-expression of HLA class I (and possibly HLA class II) was noted across pancreata from patients with newly diagnosed T1DM. In addition, islets secrete the chemokine CXCL10, attracting leukocytes expressing its receptor CXCR3 to the lesion.\(^{113}\) This chemokine production by stressed β-cells might present a master switch of islet inflammation and has attracted interest from the pharmaceutical industry as an opportunity for intervention therapy.\(^{114}\) Other strategies include efforts to reduce β-cell stress with verapamil, where early studies have shown promise for delaying T1DM disease progression.\(^{115}\) Intriguingly, high levels of insulin-specific autoreactive human T cells only precipitated insulitis and selective β-cell destruction in humanized mice in vivo after the mice had been vaccinated with insulin peptide to prime an autoimmune response and subjected to low-dose streptozotocin to stress the β-cells. This finding underscores the need for β-cell perturbation and loss of autoimmune tolerance to β-cells to create a ‘perfect storm’ that causes their destruction.\(^{21}\)

**Role of the exocrine pancreas.** T1DM seems to affect both the endocrine and exocrine pancreas, as studies have shown inflammation and loss of exocrine parenchyma.\(^{22,23}\) This finding is a potentially important missing link to be discussed and incorporated in any hypothesis aiming to clarify the mechanisms that lead to T1DM. Yet, in spite of efforts to prove an actual decline in total pancreas mass longitudinally, no evidence indicates that pancreas mass decreases with time in patients with T1DM. Indeed, although patients with T1DM often have a small pancreas, the pancreata of first-degree relatives of patients with T1DM, with or without islet autoimmunity, tend to be smaller than those of the general population as well, possibly pointing to inherent small pancreas sizes in individuals prone to developing T1DM.\(^{24}\) It is tempting to speculate that a smaller pancreas and subsequent reduced endocrine mass would increase the burden on the reduced number of β-cells trying to cope with hyperglycaemia; that is, ‘size matters’. In terms of exocrine inflammation, the argument about whether this effect is secondary to the fatal condition of the pancreas donor and organ procurement has not been settled yet; however, pancreas tissue obtained from biopsy samples of living patients with newly diagnosed T1DM tends to show less pronounced or no exocrine involvement compared with samples obtained at autopsy.\(^{22,116}\) Importantly, insulitic lesions early after diagnosis of T1DM point to monoclonal or oligoclonal infiltration with islet autoreactive CD8+ T cells only, with little evidence of ‘bystander’ T cells or exocrine involvement, which underscores the central role of autoimmunity in pancreas immunopathology at that stage. In addition, islets depleted of β-cells no longer show insulitis,\(^{22}\) which suggest that β-cells are the driving force of this inflammatory process characteristic of T1DM and underscores the central role of β-cells in the disease process.
Box 3 | Evidence supporting a role for β-cells in T1DM pathogenesis

- Smaller size of pancreas and islet mass in patients with type 1 diabetes mellitus (T1DM) and individuals at risk.
- Genetic risk associated with INS gene polymorphism associated with β-cell function.
- Genetic risk associated with polymorphisms in genes with protein products involved in β-cell protection, health and vitality.
- β-Cell stress.
- Abnormal β-cell function preceding diagnosis of T1DM (in spite of sufficient β-cell mass).
- HLA class I upregulation on endocrine cells in inflamed islets.
- HLA class I upregulation preceding islet inflammation.
- Paucity of insulitis in individuals with islet autoantibodies.
- Development of post-translational modifications (such as deamidation, citrullination and transpeptidation).
- Stress-induced ribosomal errors; post-transcriptional modification.
- Alternative splicing of islet autoantigens.
- No development of insulitis or selective loss of β-cells in humanized mice transduced with islet antigen-specific T cell receptors unless β-cells are distressed.
- Histologically distinct lesion endotypes that correlate with age at diagnosis.
- Beneficial effects of verapamil on preservation of β-cell function in new-onset T1DM.

Islet-resident macrophages and inflammation. In the dialogue between β-cells and the immune cell compartment, islet-resident macrophages have a mediating role as they engulf, process and present catabolic products from insulin granules or products that are carried by exosome particles secreted by β-cells. The localization of islet-resident macrophages near blood vessels and in close contact with β-cells, forming synapse-like structures, emphasizes their role in the effector phase of T1DM as they secrete pro-inflammatory cytokines and free radicals, triggering NF-κB and STAT1 downstream signalling pathways and FAS-mediated apoptosis in β-cells. Conventionally, macrophages recognize pathogen-associated molecular patterns or damaged tissue-associated molecular patterns (DAMPs) via Toll-like receptors (TLRs). DAMPs derived from β-cell-specific antigens (such as insulin and islet amyloid polypeptide (IAPP, also known as amylin)) have been described. Interestingly, in mouse models susceptible to autoimmune diabetes mellitus, members of the TLR family in the presence of β-cell DAMPs trigger T1DM, while in the absence of the corresponding ligands, the same TLRs exert tolerance; this finding shows the importance of β-cells in the balance between tolerance and autoimmunity. In mice, therapies affecting macrophages limit T1DM progression, while immune cells producing pro-inflammatory cytokines are retained and bound to other N-terminal peptides in a process called peptide splicing (for example, cis-peptidation reaction within PTPRN and transpeptidation reactions between IAPP and PTPRN, SLC30A8 and PCSK2, and PIK3R3 and PIK3R1), before loading on HLA. Even though progress has been made, our knowledge of neoantigens in T1DM is still at an early stage and is limited. Furthermore, other potential factors and T1DM genetic predisposition loci, as shown in NOD mice and human islets after exposure to pro-inflammatory cytokines. Combining these results with other ‘omics’ studies indicates that genes responsive to interferon, protein degradation and HLA loading machinery processes are the main factors that are disturbed during inflammation, which suggests that insulins not only leads to β-cell dysfunction but also to increased β-cell visibility to immune surveillance. We have also described how inflammation induced by ER stress can shape β-cell immunogenicity and control cytotoxic destruction by miRNA-mediated regulation of ERAP1 and its effect on preproinsulin processing.

Peptide presentation by β-cells. The effect of cytokines on β-cells is not limited to an increased peptide–HLA density at the cell surface but also affects the nature of the peptides presented. Currently, several autoantigens have been identified and while many peptides are derived from native proteins, a new range of neoantigens (protein products from mutations, frameshifts, alternative mRNA splicing and post-translational modifications) originating from alternative splicing, translational mistakes, post-translational modifications, peptide fusion and possibly immunoproteasome activation has emerged that strongly activate the immune system response. In inflammatory conditions, the increased splicing events measured by RNA-seq in human islets combined with translation infidelity and increased activity of post-translational enzymes (such as protein arginine deiminases and tissue transglutaminase 2) contribute to the diversity of the islet proteome. A β-cell ligandome landscape was presented by combining HLA class I peptidomic and transcriptomic analyses after cytokine stimulation. While these results demonstrated that most of the presented (β-cell-specific) epitopes were derived from secretory granule components, which are hyper-immunogenic, most of the alternative epitopes were not detected, despite evidence that they were able to trigger a pro-inflammatory T cell response. The low expression rate of most of these neoantigens is probably close to the sensitivity limits of proteomic analyses, so they might not be detected. Alternatively, neoantigen synthesis might require chronic rather than acute exposure to cytokines, while immune cells producing pro-inflammatory cytokines might provide additional extra stress signals to the β-cells.

Interestingly, dendritic cells can convert native islet autoantigens into immunogenic neoantigens, revealing a role for islet-resident dendritic cells in the induction or expansion of islet autoimmunity. Yet, these results have shed light on new mechanisms implying that hybrid peptides are generated by β-cells during proteolysis in the proteasome, where some protein fragments can be retained and bound to other N-terminal peptides in a process called peptide splicing (for example, cis-peptidation reaction within PTPRN and transpeptidation reactions between IAPP and PTPRN, SLC30A8 and PCSK2, and PIK3R3 and PIK3R1), before loading on HLA. Even though progress has been made, our knowledge of neoantigens in T1DM is still at an early stage and is limited. Furthermore, other potential
a 'Non-stressed' β-cell

b β-Cell under 'stress'

- Native epitope
- HLA class I
- Proteasome
- Mitochondrion
- Endoplasmic reticulum
- Insulin granule
- K+ channel
- GLUT1
- Insulin
- K+ channel
- ATP
- HLA class I peptides
- BiP
- Ribosome
- mRNA
- Polypeptides
- Function
- PDX1
- GLUT2
- Insulin

- Stress
- ATF3
- NF-κB
- CHOP
- XBP
- BiP
- Protection
- PDL1
- IDO
- Alternative RNA splicing
- SASP
- CXCL10
- PDL1
- IL-1β
- N F-
- STAT1
- MyD88
- MyD88
- TLR2
- TLR4
- PRRs
- TLR3
- TLR9
- ROS
- Peptidylarginine deiminase
- Citrullination
- Deamidation
- Transglutaminase 2
- Misfolded proteins
- BiP (bound to misfolded proteins)
- NF-κB
- PRRs
- Coxsackievirus
- Lipopolysaccharides
- Lipopeptides
- Transglutaminase 2
- Peptidylarginine deiminase
- Citrullination
- Deamidation
- BiP
- Misfolded proteins
- ROS
- NF-κB
- PRRs
- SASP
- CXCL10
- IL-1β

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REVIEWS

156 | MARCH 2021 | VOLUME 17
mechanisms have just been presented (that is, hybrid peptides and defective ribosomal products) or have been overlooked (RNA editing). Mechanisms to create neoantigens in tumours should be investigated as additional potential sources of neoantigens in T1DM. For instance, the double-stranded RNA-specific adenosine deaminase ADAR1 switches adenosine to inositol, thereby changing aspartic acid into arginine. Expression of this enzyme in breast cancer correlates with high infiltration of T cells into tumours and immune reactivity to edited antigens. Increased expression of ADAR1 in patients with systemic lupus erythematosus is associated with increased RNA editing events, indicating the possible involvement of RNA editing in the autoimmune reaction.

All the aforementioned data highlight the importance of endogenous characteristics of β-cells and their response to exogenous inflammatory stimuli for disease progression and exacerbation. These findings also demonstrate the need for intensification of efforts to fully unravel β-cell physiology in health and autoimmunity.

Lessons from cancer

The dogma describing T1DM as a disease characterized by total destruction of the insulin-producing β-cells has been shaken by immunohistochemistry studies performed on pancreatic specimens from patients with longstanding T1DM showing the presence of β-cells and insulin microsecretion (C-peptide value of <30 pmol/l) in the majority of these patients, implying that some β-cells resist or escape the immune attack, or that new β-cells are formed. The lobularity of this feature (where β-cells in certain pancreatic lobules seem unaffected, while β-cells in other lobules are depleted) might imply formation of new pancreatic lobules with unaffected islets, which increases the sense of urgency to protect β-cells after a diagnosis of T1DM. Confirming these observations, the latest single-cell analysis methods (that is, transcriptomics, mass cytometry and imaging mass spectrometry) have revealed wide heterogeneity in the β-cell population in healthy pancreata but also during disease progression, which might contribute to different sensitivities of β-cells to immune responses.

Evidence of this concept is found in multiple sclerosis, where different oligodendrocyte phenotypes have different levels of autoimmune reactions, potentially driving self-destruction. Importantly, while the presence of insulin-positive cells and lack of insulitis in longstanding T1DM might suggest that ‘normal’ islets are present, the lack of detectable C-peptide and differential clustering from islets of non-diabetic donors in multidimensional mass cytometry analyses points to intrinsic differences in patient-derived islets that might reflect prodromal islet distress and prediabetic lesions. Intriguingly, studies of insulin and proinsulin in pancreata from patients with T1DM support the existence of aetiopathological endotypes of T1DM that are associated with age at diagnosis, and point to age-related intrinsic differences in distressed β-cells during insulitis that might lead to different autoimmune reactions.

A concept is emerging that the immune response seen in T1DM might be one with ‘good intentions’, where the immune response to distressed tissue resembles the immune response that has evolved to detect infected tissue or tumours. Indeed, people carrying T1DM risk gene variants have a hyper-inflammatory immune system. It can be argued that patients with T1DM have an immune system that might be beneficial in patients with cancer. A clear analogy in support of this provocative contention is presented by Lambert–Eaton myasthenic syndrome, which has two different aetiologies: one associated with immune hypersensitivity and autoimmunity (a phenotype shared with T1DM) and one where an antitumour immune response against the voltage-gated calcium channels expressed by small cell lung carcinoma cells and nerve endings causes cross reactivity in the neuromuscular synapse. Patients with small cell lung carcinoma who develop Lambert–Eaton myasthenic syndrome have a better prognosis for cancer survival than patients who do not develop this syndrome. In addition, in patients with cancer, immune responses that are initiated after antitumour immunotherapy tend to be directed to neoantigens rather than native autoantigens.

In a similar manner to tumour cells that evade immune responses to become more invasive, β-cells have developed active self-protective mechanisms to limit further autoimmune destruction; the upregulated expression of inhibitory receptors (such as PDL1) at their cell surface and the increased expression of IDO1...
after cytokine challenge illustrate these changes. A correlation between loss of IDO1 expression and β-cell destruction extends proof for the participation of these protective mechanisms in the maintenance of the β-cell integrity. In addition, several studies have suggested that increased degranulation and/or a loss of β-cell identity occurs under environmental pressure, which is supported by the defect in insulin production and the presence of polyhormonal cells in the pancreata of patients with T1DM. From these findings, a concept of a β-cell identity crisis has emerged where β-cells dedifferentiate into other endocrine cells (α-cells or δ-cells) as a defence mechanism. Along with this β-cell identity crisis, levels of ‘semi’ β-cells that only express chromogranin A (chromogranin-positive, hormone-negative (CPhN) cells) are increased in the pancreata from patients with T1DM and T2DM and they are scattered throughout the pancreas regardless of inflammation level. The origin of these cells is still unknown; however, the mere fact that CPhN cells express the autoantigenic chromogranin A without this leading to their destruction might suggest that insulin production and the inherent negative molecular effects are needed to drive autoimmune immunity. Similarly, not all T1DM autoantigens are β-cell-specific; chromogranin A and receptor-type tyrosine-protein phosphatase N2 are also expressed in other tissues not affected by an immune attack in patients with T1DM.

By comparing islet and tumour microenvironments, increasing evidence supports the notion that in autoimmune diseases, as in effective tumour immunity or following antitumour immunotherapy, the immune system is acting on dysfunctional cells or tissues that have accumulated aberrant or modified proteins.

Conclusions
The appreciation of a role for β-cells in their own demise, the importance of ER stress in T1DM pathology, the identification of residual β-cells in patients who have had T1DM for several decades and the presence of dormant (‘hibernating’) β-cells that evade immune attack suggest that β-cell dysfunction and destruction are driven by their metabolic activity, and might lie at the heart of the aetiologies of both T1DM and T2DM. Both types of diabetes mellitus are chronic inflammatory diseases, and both are β-cell diseases. Thus far, immunotherapy alone has proven insufficient to achieve lasting preservation of β-cell function, pointing to the need to combine this strategy with β-cell therapy. In T2DM, inflammatory cytokines (secreted by stressed adipocytes or stressed β-cells) and recruitment of macrophages, B cells and T cells have been found to participate in β-cell failure and pathology. Accordingly, several intervention strategies for T2DM aimed at alleviating pressure exerted on β-cells and improving glycaemic control have been evaluated in the context of T1DM: metformin, GLP1 analogues (lixisudine, exendin 4 or sitagliptin) and verapamil have shown some benefit when combined with insulin therapy in the treatment of patients with T1DM.

We favour the engagement of the immune system, rather than suppression of the immune system, to reverse the immunopathogenesis of T1DM, in combination with β-cell therapy to improve β-cell stamina and vitality and to protect these cells from metabolic and inflammatory assaults. At a time when the coronavirus disease 2019 (COVID-19) pandemic reminds us of the need for a fully functional immune system, we cannot afford to suppress it and put patients with inflammatory disorders in danger of infection or cancer. Novel therapies are already being assessed in the clinic that ‘negotiate’ with the immune system, rather than suppress it, including ‘inverse’ vaccination strategies that aim to induce selective immune tolerance to islet autoantigens, similar to desensitization when treating allergies.

This strategy, in combination with β-cell therapy, is an attractive strategy to achieve durable remission in T1DM.

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1. Gepts, W. Islet changes suggesting a possible immune aetiology of human diabetes mellitus. Acta Endocrinol Suppl. 205, 95–106 (1976).
2. Bottazzo, G. F. et al. In situ characterization of autoimmune phenomena and expression of HLA molecules in the pancreas in diabetic insulinoma. N. Engl. J. Med. 313, 353–360 (1985).
3. Roep, B. O. The role of FcεII in the pathogenesis of type 1 diabetes: from cause to cure. Diabetologia 46, 305–321 (2003).
4. Eizirik, L. D., Coll, M. L. & Orts, F. The role of inflammation in insulitis and β-cell loss in type 1 diabetes. Nat. Rev. Endocrinol. 5, 219–226 (2009).
5. Roep, B. O., Kracht, M. J., van Lummel, M. & Zalduaide, A. A roadmap of the generation of neoantigens as targets of the immune system in type 1 diabetes. Curr. Opin. Immunol. 43, 67–75 (2016).
6. Malloine, R. & Eizirik, L. D. Presumption of innocence for beta cells: why are they vulnerable autoimmune targets in type 1 diabetes? Diabetologia 63, 1959–2006 (2020).
7. Coppieries, K. T. et al. Demonstration of islet-autoreactive CD8 T cells in insulitic lesions from recent onset and long-term type 1 diabetes patients. J. Exp. Med. 209, 51–60 (2012).
8. Leete, P. et al. Studies of insulin and proinsulin in pancreas and serum support the existence of aepathological endotypes of type 1 diabetes associated with age at diagnosis. Diabetologia 63, 1258–1267 (2020).
9. Shields, B. M. et al. C-peptide decline in type 1 diabetes has two phases: an initial exponential fall and a subsequent stable phase. Diabetes Care 41, 1486–1492 (2018).
10. Nerup, J. et al. HLA antigens and diabetes mellitus. Lancet 2, 866–866 (1974).
11. Barrett, J. C. et al. Genome-wide association study and meta-analysis find that over 40 loci affect risk of type 1 diabetes. Nat. Genet. 41, 703–707 (2009).
12. van Lummel, M. et al. Dendritic cells guide islet autoreactivity through a restricted and uniquely processed peptide presented by high-risk HLADRB1. J. Immunol. 196, 3255–3263 (2016).
13. van Lummel, M. et al. Discovery of a selective islet peptide presented by the highest-risk HLADQA1trans molecule. Diabetes 65, 732–741 (2016).
14. Pugliese, A. et al. The insulin gene is transcribed in the human thymus and transcription levels correlated with allelic variation at the INS VNTR-IDDM2 susceptibility locus for type 1 diabetes. Nat. Genet. 15, 293–297 (1997).
15. Valadiis, P. et al. Insulin expression in human thymus is modulated by INS VNTR alleles at the IDDM2 locus. Nat. Genet. 15, 289–292 (1997).
16. Bottini, N. et al. A functional variant of lymphoid tyrosine phosphatase is associated with type I diabetes. Nat. Genet. 36, 337–338 (2004).
17. Vella, A. et al. Localization of a type 1 diabetes locus in the IL2RA/CDS2 region by use of tag single-nucleotide polymorphisms. Am. J. Hum. Genet. 76, 773–779 (2005).
18. Gebe, J. A., Swanson, E. & Risau, W. HLA class II peptide-binding and autoimmunity. Tissue Antigens 59, 78–87 (2002).
19. Bottazzo, G. F., Florin-Christensen, A. & Doniach, D. Islet cell antibodies in diabetes mellitus with autoimmune polyendocrine deficiencies. Lancet 2, 1279–1282 (1974).
20. Bloem, S. J. & Roep, B. O. The elusive role of B lymphocytes and islet autoantibodies in human type 1 diabetes. Diabetologia 60, 1185–1189 (2017).
21. Wilcox, A., Richardson, S. J., Bone, A. J., Fouillas, A. K. & Morgan, N. G. Analysis of islet inflammation in human type 1 diabetes. Clin. Exp. Immunol. 155, 173–181 (2009).
22. Campbell-Thompson, M. et al. Insulitis and β-cell mass in the natural history of type 1 diabetes. Diabetes 65, 719–731 (2015).
23. Gepts, W. Pathologic anatomy of the pancreas in juvenile diabetes mellitus. Diabetes 14, 619–663 (1965).
24. Velthuis, J. H. et al. Accumulation of autoreactive effector T cells and allo-specific regulatory T cells in the pancreas allograft of a type 1 diabetic recipient. Diabetologia 52, 494–503 (2009).
25. Michels, A. W. et al. Islet-derived CD4 T cells targeting proinsulin in human autoimmune diabetes. Diabetes 66, 722–734 (2017).
26. Babon, J. A. et al. Analysis of self-antigen specificity of islet-infiltrating T cells from human donors with type 1 diabetes. Nat. Med. 22, 1482–1487 (2016).
27. Roep, B. O. et al. Cell reactivity to 38 kDa insulin-secretory granule protein in patients with recent-onset type 1 diabetes. Lancet 357, 1439–1441 (1996).
28. Roep, B. O., Arden, S. D., De Vries, R. R. P. & Hutton, J. C. T cell responses from a type 1 diabetes patient respond to insulin secretory granule proteins. Nature 345, 652–654 (1990).
29. Tan, M. et al. Activation of pancreatitis induction in humanized mice. Proc. Natl Acad. Sci. USA 110, 10954–10959 (2013).
30. Skurnik, A. et al. CTls are targeted to kill β cells in patients with type 1 diabetes through recognition of a defective ribosomal insulin gene product in type 1 diabetes. Nat. Med. 23, 501–507 (2017).
31. Culina, S. et al. Specificity for CD8+ T cell frequencies in the pancreas, but not in blood, distinguishes type 1 diabetic patients from healthy donors. Sci. Immunol. 3, eaao4015 (2018).
32. Kuric, E. et al. Demonstration of tissue resident memory CD8+ T cells in insulinic lesions in adult patients with recent-onset type 1 diabetes. Am. J. Pathol. 187, 581–588 (2017).
33. Tree, T. I. et al. Naturally arising human CD4 T cells that recognize islet autoantigens and secreted interleukin-12 promote preclinical T cell responses via linked suppression. Diabetes 59, 1451–1460 (2010).
34. Lindsay, S. et al. Defective suppressor function in CD4+CD25+ T cells from patients with type 1 diabetes. Diabetes 54, 92–99 (2005).
35. Bucy, M. et al. Defective response of regulatory CD4+ T cells to CD8+ T cell frequencies in the pancreas, but not in blood, distinguishes type 1 diabetic patients from healthy donors. Sci. Immunol. 3, eaao4015 (2018).
36. Roep, B. O. et al. T cell reactivity to 38 kD insulin-preproinsulin epitopes and detection of autoreactive antigen class I binding affinity. Diabetes 63, 1532–1609 (2014).
37. Malme精, K. C. et al. Immunological balance is associated with clinical outcome after autologous hematopoietic stem cell transplantation in type 1 diabetes. Diabetes 62, 1107–1110 (2013).
38. Illbrun, R. et al. Differences in baseline lymphocyte counts and autoreactivity are associated with differences in outcome in the type 1 diabetes patients. Diabetes 58, 2267–2276 (2009).
39. Huurnman, V. A. et al. Cellular islet autoimmunity associates with clinical outcome of islet cell transplantation. PLoS ONE 5, e2435 (2008).
40. Pinck, G. G. et al. Autoreactive CD8+ T cells associated with β cell destruction in type 1 diabetes. Proc. Natl Acad. Sci. USA 101, 18245–18240 (2004).
41. Roep, B. O., Wheeler, D. C. S. & Peakman, M. Antigen-based immune modulation therapy for type 1 diabetes: the era of precision medicine. Lancet Diabetes Endocrinol. 7, 57–67 (2019).
42. Akison, M. A., Roep, B. O., Posaghi, A., Wheeler, D. C. S. & Peakman, M. The challenge of modulating β-cell autoimmunity in type 1 diabetes. Lancet Diabetes Endocrinol. T. 52–64 (2019).
43. Skog, O., Korsgren, S., Melhus, A. & Korsgren, O. Revisiting the type 1 diabetes beta cell autoimmune disease. Curr. Opin. Endocrinol. Diabetes Obes. 20, 118–123 (2015).
44. Roep, B. O. et al. T cell reactivity to β-cell membrane antigens associated with diabetes in IDDM. Diabetes 44, 278–285 (1995).
45. Roep, B. O. et al. Autoreactive T cell responses in insulin-dependent (type 1) diabetes mellitus. Report of the first international workshop for standardization of T cell assays. J. Autoimmun. 15, 267–282 (1999).
46. Long, S. A. et al. Defects in IL-2R signaling contributing to diminished maintenance of FOXP3 expression in CD4(+CD25+) regulatory T cells of type 1 diabetic subjects. Diabetes 59, 407–415 (2010).
47. Gibson, V. B. et al. Proinsulin multi-peptide immunotherapy induces antigen-specific regulatory T cells and limits autoimmunity in a humanized mouse model. Clin. Exp. Immunol. 185, 251–260 (2019).
48. Ludwigsson, J. et al. GAD treatment and insulin secretion in recent-onset type 1 diabetes. N. Engl. J. Med. 351, 2450–2458 (2004).
49. Huurnman, V. A., Decocoz, K., Mathieu, C., Cohen, I. R. & Roep, B. O. Therapy with the hsp70 peptide Danp27 peptide delays type 1 diabetes patients. Diabetes Metab. Res. Rev. 23, 269–275 (2007).
50. Pinter, G. et al. Short chain fatty acids stimulate insulin secretion and reduce apoptosis in mouse and human islets in vitro: role of free fatty acid receptor 2. Diabetes Obes. Metab. 21, 350–355 (2019).
51. de Groot, P. F. et al. Oral butyrate does not affect insulin secretion in healthy subjects. Diabetologia 56, 597–607 (2013).
52. Hansen, L. B. et al. A low-glutten diet induces changes on human metabolism. Diabetes Obes. Metab. 12, 10954–10959 (2018).
53. Nastasi, C. et al. Butyrate and propionate inhibit antigen-specific CD11c(+) dendritic cells favouring IL-12 production by antigen-presenting cells. Sci. Rep. 7, 14157 (2017).
54. Sun, J. et al. Pancreatic β cells limit autoimmune diabetes via an immunoregulatory antimineralcorticoid peptide expressed under the influence of the gut microbiota. Immunity 43, 304–317 (2015).
55. Mian, M. et al. Gut microbiota induces innate lymphoid cell secretion inducing a proinflammatory cytokine profile contributing to disease severity. Cell Metab. 28, 571–572 e6 (2018).
56. Foulis, A. K., Jackson, R. & Farquharson, M. A. The pancreas in idiopathic Addison's disease—a search for a prediabetic pancreas. Histopathology 12, 481–490 (1988).
57. Bannasch, D. L. et al. Remapping the insulin gene/IDDM2 gene- linked minisatelite locus. Diabetes 54, 18425–18430 (2015).
58. Donkor, O. Short Chain fatty acids regulate cytokines and Th1/Th2 cells in human peripheral blood mononuclear cells in vitro. Immunol. Invest. 45, 205–222 (2016).
59. Mathieu, C. Cytokine signalling in the innate immunity and islet autoimmunity in individuals with longstanding type 1 diabetes: a randomised controlled trial. Diabetologia 56, 597–610 (2013).
60. Hansen, L. B. et al. A low-glutten diet induces changes on human metabolism. Diabetes Obes. Metab. 12, 10954–10959 (2018).
61. Antvorskov, J. C. et al. Association between maternal glutten intake and type 1 diabetes: report of a first- in-man phase I safety study. Clin. Exp. Immunol. 182, 251–260 (2019).
62. Bennett, S. et al. Mapping the insulin gene/IDDM2 locus in type 1 diabetes. Diabetes 53, 1884–1889 (2004).
63. Nielsen, B. et al. Impact of IDDM2 on disease pathogenesis and progression in children with newly diagnosed type 1 diabetes: reduced insulin antibody titres and preserved beta cell function. Diabetologia 49, 71–76 (2006).
64. Durinovic-Bello, I. et al. Insulin gene VNTR genotype associates with frequency and phenotype of the autoimmune response to proinsulin. Genes Immun. 11, 188–193 (2010).
65. Durinovic-Bello, I. et al. Class III alleles at the insulin VNTR polymorphism are associated with T cell responses to proinsulin epitopes in HLA-DR4, DB8 individuals. Diabetes 54 (Suppl. 2), 18–24 (2005).
66. Bennett, S. et al. IDDM2-VNTR-encoded susceptibility to type 1 diabetes: dominant protection and parental transmission of alleles in the insulin gene-linked minisatelite locus. J. Autoimmun. 9, 415–421 (1996).
67. Vafadaz, P. et al. Imprinted and genotype-specific expression of genes in fetal liver precursor cells and leukocytes. J. Autoimmun. 9, 397–405 (1996).
68. Gysenens, C., Callwaert, H., Overbeek, L. & Mathieu, C. Cytochrome c oxidase: a dual role for IFNβ. Biochem. Soc. Trans. 36, 528–533 (2008).
69. Estiri, L. et al. The human pancreatic islet transcriptome: expression of candidate genes for type 1 diabetes and the impact of pro-inflammatory cytokines. PLoS Genet. 8, e1002552 (2012).
106. Davies, J. L. et al. A genome-wide search for human type 1 diabetes susceptibility genes. Nature 371, 130–136 (1994).
107. Ferrannini, E. et al. Progression to diabetes in relatives of type 1 diabetic patients: the Diabetes and mode of onset of diabetes. Nature 59, 679–685 (2010).
108. Sosenko, J. M. et al. The acceleration of the loss of pancreatic endocrine function in type 1 diabetes: discovery of NIDDM mice. Diabetologia 55, 1021–1029 (2012).
109. Rui, J. et al. Methylation of insulin DNA in response to proinflammatory cytokines during the progression of autoimmune diabetes in NOD mice. Diabetologia 55, 1021–1029 (2012).
110. Okazaki, K. et al. The activation of pancreatic β-secretase in the progression of type 1 diabetes. Nat. Med. 26, 1423–1432 (2020).
111. Thomaides, S. et al. β-Cell stress shapes CTL immune recognition of proinsulin signal peptide by posttranslational regulation of endoplasmic reticulum aminopeptidase 1. Diabetes 69, 670–680 (2020).
112. Marasco, M. R. & Linnemann, A. K. Cell-autophagy in diabetes pathogenesis. Endocrinology 159, 2127–2141 (2018).
113. Meyerovich, K., Ortis, F., Allagapt, F. & Cardozo, A. K. Endoplasmic reticulum stress and the unfolded protein response in pancreatic islet inflammation. J. Mol. Endocrinol. 57, R1–R17 (2016).
114. Stirling, J. et al. Posttranslational beta cell protein modifications trigger type 1 diabetes? Diabetesologia 56, 2354–2356 (2013).
115. Marroqui, L. et al. Pancreatic β-cells are resistant to metabolic stress-induced apoptosis in type 2 diabetes. ElBioMedicine 2, 378–385 (2015).
116. Moreno, P. et al. Overexpression of the endoplasmic reticulum chaperone Hsp90 is a master regulator of pancreatic β-cell apoptosis and islet inflammation. J. Biol. Chem. 286, 929–941 (2011).
117. Li, N. et al. Age-related stress induced β-cell senescence and its implication in diabetes development. Aging 11, 9947–9959 (2017).
118. Rojas, J. et al. Pancreatic β-cell death: novel potential mechanisms in diabetes therapy. J. Cell Res. 18, 9601801 (2018).
119. Thompson, P. J. et al. Targeted elimination of senescent beta cells prevents disease in diabetic mice. Cell 259, 1045–1060.e10 (2019).
120. Roep, B. O. et al. Islet inflammation and CXCL10 in recent-onset diabetes: a cross-sectional study. Clin. Exp. Immunol. 159, 338–343 (2014).
121. Bonvin, P. et al. Antibody neutralization of CXCL10 in vivo is dependent on binding to free and not endothelial-bound chemokine: implications for the design of a new generation of anti-chemokine therapeutic antibodies. J. Biol. Chem. 292, 4185–4197 (2017).
122. Ovalle, F. et al. Verapamil and beta cell function in adults with recent-onset type 1 diabetes. Nat. Med. 24, 1008–1010 (2019).
123. Krogvold, L. et al. Pancreatic biopsy by minimal tail resection in live adult patients at the onset of type 1 diabetes. Diabetologia 57, 841–843 (2014).
124. Wan, X. et al. Pancreatic islets communicate with lymphocytes to sequester the expression of insulin peptides. Nature 560, 107–111 (2018).
125. Carrero, J. A. et al. Resident macrophages of pancreatic islets have a seminal role in the initiation of autoimmune diabetes of NOD mice. Proc. Natl Acad. Sci. USA 114, E1048–E10627 (2017).
126. Kolb-Bachofen, V. & Kolb, H. A role for macrophages in the pathogenesis of type 1 diabetes. Autimmunity 3, 145–154 (1989).
127. Golden, E. & Wen, L. Toll-like receptor activation in islets and tolerance to autoimmunity in diabetes. Front. Immunol. 5, 119 (2014).
128. Carrero, J. A. et al. Depletion of islet resident macrophages protects mice from type 1 diabetes [abstract]. J. Immunol. 200 (Suppl. 1), 41 (2013).
129. Hutchings, P. et al. Transfer of diabetes in mice following the first positive β- cell response during the progression to type 1 diabetes in diabetes prevention trial-1 participants. Diabetes 62, 4179–4183 (2013).
130. Marflour, J. et al. Expression of endoplasmic reticulum stress markers in the islets of patients with type 1 diabetes. Diabetologia 55, 2417–2420 (2012).
131. Thomasaid, S. et al. β-Cell stress shapes CTL immune recognition of proinsulin signal peptide by posttranslational regulation of endoplasmic reticulum aminopeptidase 1. Diabetes 69, 670–680 (2020).
132. Lopes, M. et al. Temporal profiling of cytokine-induced genes in pancreatic β-cells by meta-analysis and network inference. Canones 105, 264–275 (2014).
133. Gonzalez-Duque, S. et al. Conventional and neo-antigens presented by β-cells are targeted by circulating naive CD8+ T cells in type 1 diabetic and healthy donors. Cell Metab. 28, 946–960 (2018).
134. Drez, J. et al. Differential splicing of the I-A2 mRNA in pancreas and lymphoid organs as a permissive genetic mechanism for autoimmunity against the IA-2 type 1 diabetes autoantigen. Diabetes 50, 895–901 (2001).
135. Remo, B. et al. T cells specific for post-translational modifications of both IA-2 and IA-2a contribute to autoimmunity in the NOD mouse. Nat. Commun. 3, 555 (2012).
136. McCaughan, R. J. et al. Human islets and dendritic cells can generate new specificities following modified islet autoantigens. Clin. Exp. Immunol. 183, 135–140 (2016).
137. Delong, T. et al. Pathogenic CD4 T cells in type 1 diabetes recognize epitopes formed by peptide fusion. Science 351, 711–714 (2016).
138. Thomaides, S., Zalutamide, A. & Roep, B. O. Islet stress, degradation and autoimmunity. Diabetes Obes. Metab. 20 (Suppl. 2), 88–94 (2018).
139. Hutton, J. C. & Davidson, H. W. Cytokine-induced dying and splicing of the i-A2β and i-A2α islet cell in the heterozygous β-cell response in type 1 diabetes. Diabetes 59, 355–356 (2010).
140. Alvise, M. J., Juan-Mateos, J., Colli, M. L., Turatsinze, V. J. & Eizirik, D. L. Why can some new and many–alternative splicing in β-cell function and failure. Diabetes Obes. Metab. 20 (Suppl. 2), 77–87 (2018).
141. Marre, M. L., James, E. A. & Piganelli, J. D. β-cell ER stress and the implications for immunogenicity in type 1 diabetes. Front. Cell Dev. Biol. 3, 67 (2015).
142. Zhang, M. et al. RNA editing derived epitopes function as autoreactive T cell targets for immune responses. Nat. Commun. 9, 3919 (2018).
143. Roth, S. et al. Increased RNA editing may provide a source for selection of the CD4+ CD8+ T cell receptor repertoire. Cell Res. 23, 50–57 (2013).
144. Morgan, N. G. & Richardson, S. J. Fifty years of pancreatic islet pathology in human type 1 diabetes: insights gained and progressed more. Diabetologia 61, 2499–2506 (2018).
145. Oram, R. A., Sims, E. K. & Evans-Molina, C. Beta cells in type 1 diabetes: mass and function; sleeping or dead? Diabetesologia 62, 567–577 (2019).
146. Muraro, M. J. et al. A single-cell transcriptome atlas of the human pancreas. Cell 32, 385–394.e5 (2016).
147. Wang, Y. J. et al. Multiplexed in situ imaging mass cytometry analysis of human islets and insulinomas. PNAS 115, 15460–15465 (2018).
148. Dammann, N. et al. Human type 1 diabetes progression by imaging mass cytometry. Cell Metab. 29, 755–768.e5 (2019).
149. Ananthan, D. et al. Cell death is not uniform after all–novel insights into molecular heterogeneity of insulin–secreting cells. Diabetologia 60, 1546–1557 (2018).
150. Orban, T. et al. Co- stimulation modulation with abatacept in patients with recent-onset type 1 diabetes: a randomized, double-blind, placebo-controlled phase 2 trial. Lancet Diabetes Endocrinol. 1, 284–294 (2013).
151. Schneider, A. et al. The combination of two T cell antigens in diabetes subjects are resistant to regulation via CD4+ FOXP3+ regulatory T cells. J. Immunol. 181, 7350–7355 (2013).
152. Anderson, M. S. et al. Projection of an immunological self shadow within the thymus by the AIRE protein. Science 298, 1595–1601 (2002).
173. Wildin, R. S. et al. X-linked neonatal diabetes mellitus, enteropathy and endocrinopathy syndrome is the human equivalent of mouse scurfy. *Nat. Genet.* **27**, 18–20 (2001).

174. Endl, J. et al. Identification of naturally processed T cell epitopes from glutamic acid decarboxylase presented in the context of HLA-DR alleles by T lymphocytes of recent onset IDDM patients. *J. Clin. Invest.* **99**, 2405–2415 (1997).

175. Roep, B. O. et al. Plasmid-encoded proinsulin preserves C-peptide while specifically reducing proinsulin-specific CD8\(^+\) T cells in type 1 diabetes. *Sci. Transl Med.* **5**, 191ra82 (2013).

176. Huurman, V. A. et al. Immunological efficacy of heat shock protein 60 peptide DiaPep277 therapy in clinical type I diabetes. *Clin. Exp. Immunol.* **152**, 488–497 (2008).

177. van Lummel, M. et al. Posttranslational modification of HLA-DQ binding islet autoantigens in type 1 diabetes. *Diabetes* **63**, 237–247 (2014).

178. de Jong, V. M. et al. Post-transcriptional control of candidate risk genes for type 1 diabetes by rare genetic variants. *Genes Immun.* **14**, 58–61 (2013).

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