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THERAPIES AND NEW TECHNOLOGIES IN THE TREATMENT OF DIABETES (M PIETROPAOLO, SECTION EDITOR)

Current Status on Immunological Therapies for Type 1 Diabetes Mellitus

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Abstract

Purpose of Review Type 1 diabetes (T1D) occurs when there is destruction of beta cells within the islets of Langerhans in the pancreas due to autoimmunity. It is considered a complex disease, and different complications can surface and worsen the condition if T1D is not managed well. Since it is an incurable disease, numerous treatments and therapies have been postulated in order to control T1D by balancing hyperglycemia control while minimizing hypoglycemic episodes. The purpose of this review is to primarily look into the current state of the available immunological therapies and their advantages for the treatment of T1D.

Recent Findings Over the years, immunological therapy has become the center of attraction to treat T1D. Immunomodulatory approaches on non-antigens involving agents such as cyclosporine A, mycophenolate mofetil, anti-CD20, cytotoxic T cells, anti-TNF, anti-CD3, and anti-thymocyte globulin as well as immunomodulative approaches on antigens such as insulin, glutamic acid decarboxylase, and heat shock protein 60 have been studied. Aside from these two approaches, studies and trials have also been conducted on regulatory T cells, dendritic cells, interleukin 2, interleukin 4, M2 macrophages, and rapamycin/interleukin 2 combination therapy to test their effects on patients with T1D. Many of these agents have successfully suppressed T1D in non-obese diabetic (NOD) mice and in human trials. However, some have shown negative results.

Summary To date, the insights into the management of the immune system have been increasing rapidly to search for potential therapies and treatments for T1D. Nevertheless, some of the challenges are still inevitable. A lot of work and effort need to be put into the investigation on T1D through immunological therapy, particularly to reduce complications to improve and enhance clinical outcomes.

Keywords T1D · Immunotherapies · Non-antigenic agents · Antigenic agents

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Introduction

Diabetes mellitus composes a group of metabolic defects due to deformities in the action of insulin, the secretion of insulin, or both. It is defined by hyperglycemia [1, 2].

Type 1 diabetes (T1D) arises from induced beta-cell autoimmunity, which involves a process that is marked by the production of islet-reactive autoantibodies [3]. These autoantibodies play a major role in the development of autoreactive T cells and are capable of destroying insulin-producing beta cells. Overtime, there will be a progressive loss of beta-cell function [4, 5] (Fig. 1). Various treatments and therapies have been invented in order to control T1D by balancing hyperglycemia control while minimizing hypoglycemic episodes. However, glycemic control alone does not cure or reverse the autoimmune process [6]. Michels and colleagues have reported on an update about the successful prediction and prevention of type 1 diabetes. Autoantigens found in the islets are thought to play a significant role in the pathogenesis of T1D. In addition, genetic predisposition and several other environmental factors too are responsible for the development of the disease [7].

Several new approaches have been designed and tested to contain the disease. Immunological targets and related therapy has gained much significance lately. Although immune interventions have been studied in much detail, the effectiveness of such strategies in curing T1D seems disappointing. Scientists have, ever since, started to rethink the immunotherapeutic strategies in the management of this condition. Several studies involving laboratory animals have shown a large success. However, this could not be translated into human studies. Our review, in this direction, attempts to look into the current status of the various reported immunological therapies for T1D.

Methodology

This review on immunological therapies for T1D was carried out from April 2018 to September 2018. Pubmed and Google Scholar were the main resources used to perform online searches. Keywords such as T1D, immunological therapies, and immunomodulative approaches were used. Only papers in English language were chosen. Among over 50,000 hits, 85 of the most relevant papers were considered for this review. All the authors were involved in the selection of papers. Publications and reports that had the primary keywords, type 1 diabetes, immunology, and immunological therapy, were selected for inclusion. In addition, reports and publications that had the above keywords in the abstract or/and main text were also included. Reference lists of selected publications were also checked for additional articles. All relevant articles, irrespective of the type of the study were included. These included clinical studies, preclinical studies, opinions, reviews, and commentaries. Non-immunological therapies, foreign language publications, unsolicited opinions, general comments, and related papers were omitted. The titles, as well as the abstracts, were screened thoroughly to assess the relevance of each study for this review. Several meetings were conducted to segregate the papers based on sub-clusters. When there arose a conflict of opinion between two co-authors, a third co-author was included.

Non-antigen Specific Immunomodulative Approaches

In the 1980s and early 1990s, several clinical intervention trials were performed to investigate the use of general immunosuppressive agents (non-antigen-specific) on T1D [8], with

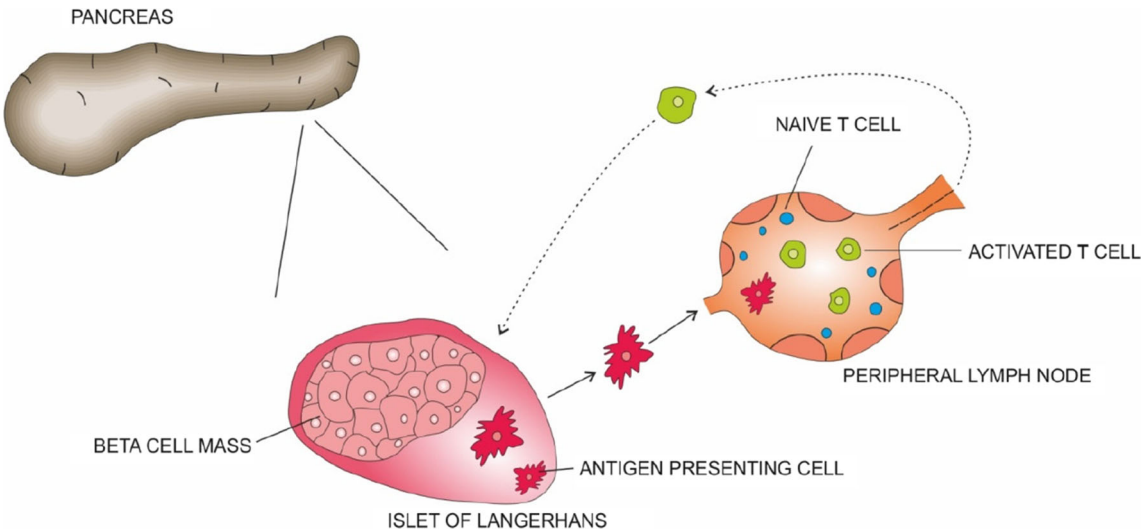

Fig. 1 Pathophysiology of T1D

the hope that they would interrupt the autoreactive process and halt the development of T1D. Some of these approaches delayed the progression of the condition. However, the benefits disappeared when the immunosuppressive agents were removed [9]. A list of non-antigen specific immunomodulatory approaches is presented in Table 1.

Cyclosporin A (CsA)

Due to its powerful immunosuppressive capability, CsA is one of the earliest immunosuppressive drugs used in patients who were newly diagnosed with T1D [10]. CsA is a calcineurin inhibitor which interferes with signal transduction mediated by T cell receptors (TCR), causing the inhibition of T cell activation as well as the secretion of interleukin 2 (IL-2) by helper T cells [11]. Its drawbacks include potential toxicity towards pancreatic beta cells and the high cost of this drug [9].

Mycophenolate Mofetil (MMF)

MMF suppresses T cell responses towards allogeneic cells and many antigens [12]. It inhibits inosine monophosphate dehydrogenase (IMPDH), the rate-limiting enzyme in the de novo production of guanosine nucleotides. It suppresses the proliferation of both T cells and B cells, thus reducing the formation of antibodies and cell-mediated responses. Moreover, MMF diminishes the recruitment of lymphocytes to inflammatory sites and elevates their apoptosis rate [13]. A multicenter trial conducted by the Type 1 Diabetes TrialNet at 13 sites in Europe as well as North America aimed to determine whether MMF alone or in combination with daclizumab (DZB) was able to delay the destruction of beta cells. Subjects with recent onset T1D were randomly allocated to MMF alone, the combination of MMF and DZB, or placebo. Over 2 years of follow-up, the mean of C-peptide was not affected by MMF alone nor by the combination of MMF and DZB as compared with placebo [14]. Regardless of its negative outcome in T1D, the trial showed that not all immunosuppressive procedures can treat T1D effectively. One potential explanation is that DZB can remove cell subsets that help to maintain self-tolerance in T1D [9].

Table 1 Non-antigen specific immunomodulative approaches

Non-antigen specific immunomodulative approaches	Cyclosporin A (CsA) Mycophenolate mofetil (MMF) Rituximab (anti-CD20) Cytotoxic T cells-associated protein 4 (CTLA-4) • Abatacept • Belatacept Anti-TNF • Etranercept Anti-CD3 • hOKT3gamma1(Ala-Ala) • Aglycosylated human IgG-1 antibody directed against CD3 (ChAglyCD3) • Teplizumab Anti-thymocyte globulin (ATG)

Rituximab

Rituximab is a monoclonal antibody that targets the surface marker B lymphocyte antigen-CD20 (CD20), which is expressed only by pre-B cells and mature B cells [15]. The expression of CD20 begins on the surface of the B cells from the early phase of the life cycle and then progresses until maturity [16].

In 2009, a phase 2 trial was conducted to determine if rituximab can preserve beta cell function in T1D. Eighty-seven patients with newly diagnosed TID were randomly allocated to receive four weekly doses of rituximab or placebo. One year after the first infusion, the mean C-peptide area under the curve (AUC) was higher in the rituximab group than in placebo, but unfortunately, this response was not sustained long-term [17].

Cytotoxic T Cells-Associated Protein 4 Immunoglobulin

Cytotoxic T cell-associated protein 4 (CTLA-4) is an antigen expressed by activated T cells that aids in the mediation of a T cell inhibitory signal to limit excessive T cell stimulation. CTLA-4 immunoglobulins (CTLA4-Ig) such as abatacept and belatacept are fusion proteins engineered pharmacologically to interfere with T cell immunity [18]. CTLA4-Ig connects the extracellular binding domain of CTLA-4 to an Fc domain of immunoglobulin G (IgG) and binds both CD80 and CD86 with high affinity [19, 20]. When non-obese diabetic (NOD) mice were treated with CTLA4-Ig, the protein was shown to be ineffective against diabetes [21]. A phase 2 trial was performed in which subjects received abatacept every month for 2 years or placebo. Over 2 years, it was shown that abatacept delayed the deterioration of beta cells function [22].

Anti-TNF

Anti-TNF drugs are used to treat chronic pro-inflammatory autoimmune diseases such as RA [23]. However, a soluble recombinant TNF receptor fusion protein that binds TNF was unable to arrest the development or the progression of T1D [24]. In contrast, a double-blinded study whereby etanercept was used to treat children with recent onset T1D showed that etanercept helped lower the insulin dose required by the children given etanercept as compared with the placebo group, suggesting that it might help preserve beta cell function [25].

Anti-CD3

Anti-CD3 is a monoclonal antibody that binds to CD3 regions on the surface of T cells and. is commonly used in transplantation and autoimmunity [26]. A study used non-activating humanized anti-CD3 (hOKT3gamma1(Ala-Ala), subsequently named teplizumab) to evaluate its reaction on insulin production in patients with T1D. Twelve randomly selected participants were given hOKT3gamma1(Ala-Ala) for 14 days whereas 12 others were not. hOKT3gamma1(Ala-Ala) maintained insulin production in the subjects who were given the monoclonal antibody [27]. Furthermore, in a multicenter, phase 2 study, 80 subjects with recent T1D onset were assigned randomly to receive anti-CD3 in the form of a glycosylated human IgG-1 antibody directed against CD3 (ChAglyCD3) or placebo. Beta cell function was maintained and the required insulin dose was lowered in the group receiving ChAglyCD3 [28]. Thus a 2 year, double-blinded trial was conducted to evaluate the efficiency and safety of teplizumab [29]. Teplizumab was injected intravenously daily for a period of 14 days at baseline and then after 26 weeks in patients with recent-onset T1D. Among 516 subjects, 513 were treated and only 462 completed their 2-year follow-up. Upon 2 weeks of a full dose of teplizumab, the mean C-peptide AUC loss was reduced in the treated group as compared with the placebo group, as was the required insulin dose. Therefore, teplizumab appeared to be safe and effective in reducing the loss of C-peptide in patients with T1D even 2 years after the diagnosis [30]. However, the phase 3 clinical trial conducted by MacroGenics on teplizumab failed to meet the primary endpoint.

Anti-thymocyte Globulin

Anti-thymocyte globulin (ATG) is a polyclonal antibody that can deplete T cells. It is often used in the setting of transplantation rejection to avoid graft-versus-host disease (GVHD) [31]. In an early study, ATG appeared to prevent C-peptide loss [32]. In a more recent study, a 2-year randomized trial was conducted in patients with new-onset T1D. In a post hoc analysis, the group of subjects who were given ATG showed higher C-peptide AUC values as compared with the group that was given a placebo. However, ATG can only considerably reduce T cell subsets when given within a short period of time after disease onset, and in most subjects, it does not preserve islet function after 24 months [33•].

Antigen-Specific Immunomodulative Approaches

Antigen-specific therapeutic approaches have been successfully applied to various autoimmune diseases [34]. Antigen-specific immune modulation can inactivate autoreactive T cells, which has been leveraged in numerous trials on animals and subjects with T1D [32]. A list of antigen-specific immunomodulatory approaches is presented in Table 2.

Insulin

Insulin is activated via enzymatic cleavage to produce proinsulin and C-peptide from its preproinsulin precursor. In NOD mice, insulin autoantibodies appear at the 8th week of age [35]. When NOD mice were given 1 mg of insulin via oral administration twice a week for 5 weeks, diabetes onset was delayed, and diabetes incidence was lowered [36]. Moreover, the development of diabetes was blocked when NOD mice were given insulin intranasally [37]. However, in humans, oral insulin administration did not stop the occurrence of T1D in high-risk patients or delay its progression in newly diagnosed patients. In a double-masked trial, first- and second-degree relatives of individuals with T1D were screened, and 97,273 samples were analyzed. Three thousand four hundred eighty-three individuals had positive anti-islet cell antibodies, and 372 were randomly allocated to receive 7.5 mg/day of oral insulin or placebo. Forty-four patients who were given insulin and 53 who received placebo were diagnosed with diabetes, showing that oral insulin did not slow down or block the progression of T1D [38]. In a 2-year trial, 52 subjects with new-onset T1D were randomly assigned to nasal insulin or placebo. Within this period, serum C-peptide, fasting blood glucose, serum antibodies to islet antigens, and glucagon-stimulated serum C-peptide levels were carefully monitored. Overall, the β cell function dropped by about 35% and 23 of 52 participants (44%) progressed to insulin treatment. Most of the metabolic parameters remained the same between nasal insulin and placebo groups, but the insulin antibody response to injected insulin was significantly blunted in a sustained manner in those who had received nasal insulin. Therefore, it can be concluded that inhalation insulin did not prevent T1D [39]. Despite the negativities, evidence shows that immune tolerance to induced insulin contributes a theory for its approach to inhibit T1D. A significant study conducted by the TrialNet Oral Insulin Group revealed crucial information regarding insulin based intervention in T1D. The findings suggest that oral insulin did not delay or prevent the development

Table 2 Antigen specific immunomodulative approaches

Antigen specific immunomodulative approaches	Insulin Glutamic acid decarboxylase (GAD) • Glutamic acid decarboxylase 65 (GAD65) • Alum-formulated GAD (GAD-alum) Heat shock protein 60 (Hsp60) • Heat shock protein 65 (Hsp65) • DiaPep277

of type 1 diabetes. The study recruited a total of randomly selected 560 autoantibody-positive participants. Diabetes was diagnosed in 28.5% and 33% subjects in the oral insulin and the placebo groups respectively. However, there was no significant difference in the time to diabetes between the two groups [40].

Glutamic acid decarboxylase 65 (GAD 65)

In preclinical studies in NOD mice, GAD65 can induce immunotolerance and halt diabetes development before the onset of the disease. The immune responses of T-helper 2 cells (Th2) to GAD65 were studied to determine if inhibition of the cascade of T-helper 1 cells (Th1) which leads to T1D can be arrested before the onset of diabetes. In 2 to 3-week-old NOD mice, the administration of GAD65 induced IgG-1 antibodies at high levels. GAD65 effectively lowered insulitis as well as T1D [41].

In clinical trials, the ability of alum-formulated GAD (GAD-alum) to overturn recently diagnosed T1D was tested in individuals of 10 to 18 years of age. Seventy participants with fasting C-peptide levels of more than 0.3 ng/mL were randomly allocated to receive 20 μg of GAD-alum or placebo (alum alone). In both the GAD-alum and the placebo group, there was a gradual decrease in the level of insulin secretion. However, after 15 months, no changes were shown in the fasting C-peptide level. As compared with the placebo group, the fasting C-peptide level of the GAD-alum group decreased less over 30 months. Thus it seemed that GAD-alum contributed to the preservation of insulin secretion but failed to reduce insulin requirements in patients with new-onset T1D [42]. To further support this finding, a double-masked, randomized trial was performed in 145 individuals with T1D. In this study, GAD-alum did not change the loss of insulin secretion in patients with newly diagnosed T1D [43]. In another phase 2 study designed to evaluate the safety and adequacy of GAD-alum, 70 subjects of 10 to 18 years of age were enrolled to receive either 20 μg of GAD-alum subcutaneously or placebo. After 15 months, insulin secretion as measured by C-peptide was significantly preserved in the GAD-alum group as compared with the placebo group, with a lower reduction in fasting C-peptide level between day 1 and month 15 in the GAD-alum group [44].

Heat Shock Protein 60

Heat shock protein 60 (Hsp60) is a protein expressed in mitochondria that aids in the folding of small and soluble proteins that takes place at the mitochondrial matrix [45]. Under conditions of mitochondrial stress, its expression is upregulated. Hsp60 is also found to be elevated in T1D [46•]. In NOD mice, the antigen that targets beta cells was found to cross-react with the heat shock protein 65 (Hsp65) of the bacteria *Mycobacterium tuberculosis*. Beta cell destruction was linked to the spontaneous growth of anti-Hsp65 T cells. The Hsp65 cross-reactive antigen was also detectable in the blood of pre-diabetic mice but declines with overt T1D development. Clones of anti-Hsp65 T cells had the ability to cause insulitis as well as hyperglycemia in young NOD mice [47]. DiaPep277 is a synthetic peptide derived from Hsp60 [26]. According to a phase 2 trial, 35 male patients with C-peptide levels higher than 0.1 nmol/L were treated periodically with 1 mg of DiaPep277 or placebo for 12 months and observed for 18 months. At the 18th month, the level of C-peptide was sustained in the DiaPep277 group, whereas the level of C-peptide was decreased in the placebo group. Additionally, the exogenous insulin requirement was lower in the DiaPep277 group than the placebo group [48]. However, a phase 2 study of DiaPep277 in children failed to show preservation of beta-cell function: 30 children with T1D aged 7 to 14 years received DiaPep277 (1 mg) subcutaneously or mannitol (40 mg) at study entry and at the 1st, 6th, and 12th months. The C-peptide levels in both DiaPep277 and mannitol groups decreased similarly over time and there was no significant change in the insulin dose in both groups [49].

Other Immunomodulative Approaches

Regulatory T Cells

In the mid-1990s, CD4 was found to be expressed on a subpopulation of suppressor T cells, named regulatory T cells (Tregs). Tregs aid in the maintenance of immune tolerance, suppress allergies and asthma, and prevent autoimmune diseases [50, 51]. Tregs can also induce resistance against dietary antigens [52]. In 2003, the forkhead box transcription factor (FOXP3) was found to assist Tregs in inducing immune tolerance [53]. Tonkin and colleagues used NOD mice to evaluate the influence of Tregs in suppressing T1D. Their findings showed that Tregs is capable of suppressing T1D only in the presence of their Ag. The suppression was not observed in mice that lacked Treg Ag in their islets [54]. Several cell surface markers are commonly employed in the identification of Tregs. These are CD4(+)CD25(+)Foxp3(+) and CD45 isoforms RA/RO [55]. It is now known that T1D patients show a Tregs activation defect, who have increased resting Tregs (rTregs) and reduced activated Tregs (aTregs) [56]. Despite several attempts made to study, the expression of Tregs in type 1 diabetic patients to evaluate its function in immune tolerance, the effect of Tregs in subjects with T1D still remains greatly unknown.

Dendritic Cells

Dendritic cells are antigen-presenting cells derived from the bone marrow. The interface between both adaptive and innate immunities are needed for the induction of both humoral and cellular immunities [57], including the activation of T cells [58]. They help maintain immune tolerance by clearing apoptotic cells rapidly. To determine if dendritic cells can prevent T1D by reestablishing peripheral tolerance via the use of dendritic cells primed with apoptotic bodies from beta cells, NOD rat insulin promoter interferon-β mice were injected and tested with dendritic cells that had been cultured with nitrilase-1 apoptotic bodies. The treatment group had lower disease incidence than the control group [59]. Though favorable outcomes were seen in NOD mice, unsatisfactory results were achieved in humans with T1D [60]. A randomized phase 1 study of 10 subjects with T1D who were injected with dendritic cells into the abdomen intradermally and monitored for 12 months showed that the therapy was safe [61]. However, several other human studies resulted in unsatisfactory results. There are several factors contributing to this. In contrast to homogenous mice strains, human subjects are naturally heterogeneous, which is one of the important factors contributing to unsuccessful outcomes. In addition, there is the factor of differential environmental exposures that exist in subjects with T1D [60].

Interleukin 2

IL-2 is produced by activated $CD4^+$ T-helper cells and enhances T cell proliferation and differentiation [62, 63]. In addition, IL-2 allows the formation of $CD8^+$ memory T cells and full secondary expansion [64], raising the possibility that it might be used to develop vaccines or as a therapeutic strategy in autoimmune diseases. In early studies [65], immunotherapies involving IL-2 reversed diabetes in NOD mice. Deficiency of IL-2 resulted in the loss of regulatory T cell function in the NOD model. A potential improvement can be made by using antibody-bound IL-2 (Ab/IL-2). NOD mice with recent onset of diabetes were injected with Ab/IL-2 intraperitoneally for 21 days. After 3 weeks of Ab/IL-2 treatment, 6 out of 13 NOD mice became normoglycemic, and NOD mice injected with Ab/IL-2 survived even when insulin therapy was not provided. However, Ab/IL-2 treatment could not reverse the onset of hyperglycemia if it was delayed for 4 weeks after the onset of diabetes [66].

Interleukin 4

. In NOD mice, IL-4 has been shown to block T cell responses and protect against diabetes [67, 68]; IL-4 also prevented insulitis in NOD mice [69]. IL-4 immunotherapy has been attempted in NOD mice by employing adenovirus vectors to

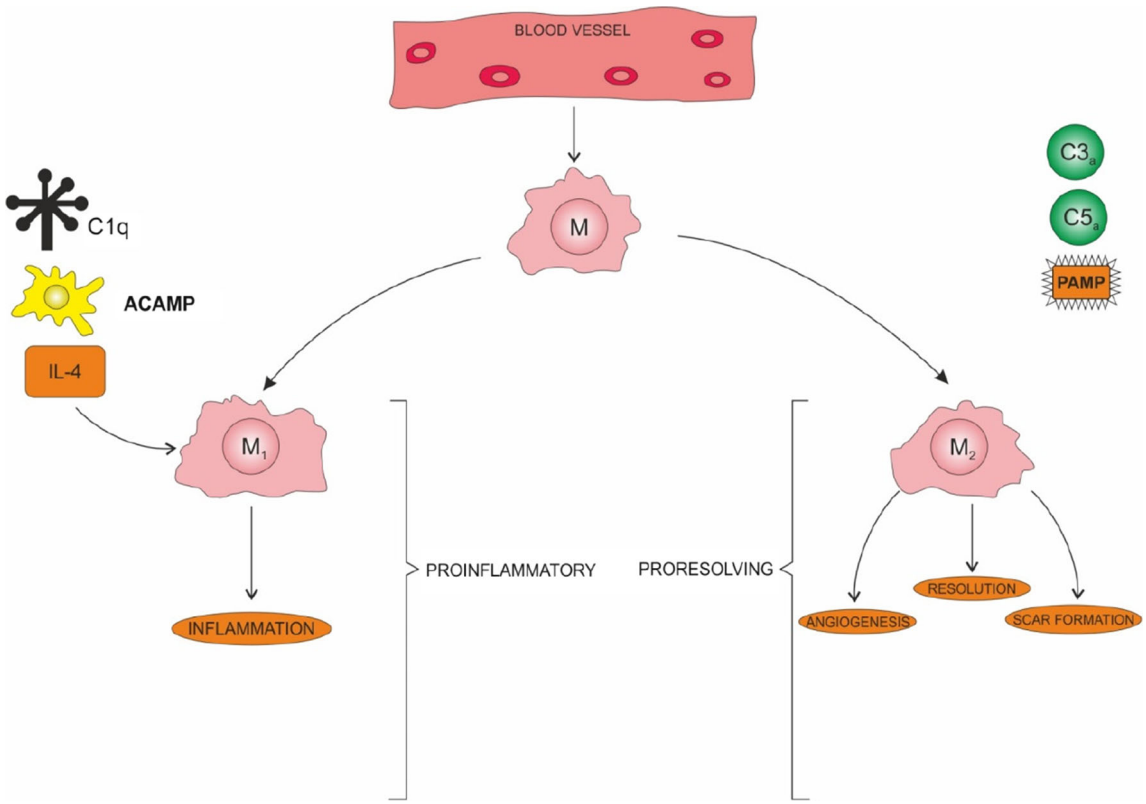

Fig. 2 Polarization of macrophages

mediate gene transfer, which lowered diabetes incidence from 80 in the control group to 20% in the treated group [70].

M2 Macrophages

Macrophages play a vital role in immune functions of the body [71, 72]. Macrophage polarization and the overall roles of M1 and M2 macrophages are simplified in Fig. 2. A study conducted [73] on NOD mice to determine the effect of TGF beta/IL-4/ IL-10 on the immunosuppression of M2 macrophage phenotype (M2r) revealed that more than 80% of the NOD mice were protected against T1D for at least 3 months, and NOD mice which were treated with M2r had an increased amount of beta cells in the pancreas [74].

Combination Therapies

To date, clinical trials on the evaluation of combination therapies for T1D are very limited. The success of combination therapies of immunomodulatory agents in NOD mice might not have the same outcome in humans [75]. In NOD mice, the combination of rapamycin and IL-2 combination have proven to be effective against T1D. To prove the effectiveness and safety of this combination therapy in humans, a phase 1 trial was conducted on nine subjects. Within the first month of therapy, the levels of Tregs were increased in parallel to the IL-2 treatment, but there was a transient metabolic worsening indicating that IL-2 therapy can enhance the level of Tregs in patients with T1D [76] while promoting beta cell dysfunction. Given the dearth of data, it is necessary to perform more trials by using combinations of different immunosuppressive agents in patients with T1D to seek promising drug combinations [77].

GNbAC1

A newly established therapeutic strategy uses the human endogenous retrovirus-W (HERV-W) envelope [78, 79]. Numerous experiments and studies suggest that this genome is involved in T1D pathogenesis. HERV-W envelope was detected in 70% of the sera of patients with T1D, whereas it was present in only 12% of patients without diabetes [80]. Similarly, its RNA was detected in 57% of individuals with T1D but only in 12% of those without diabetes [81••]. GNbAC1 is an IgG-4 kappa recombinant antibody that was developed to target the genome specifically and to cause neutralization of the effects of HERV-W envelope both in vitro and in vivo. Currently, the safety aspects of GNbAC1 and its effect on insulin production in patients with recent onset T1D is being studied [82]. If the outcomes are positive, treatment with GNbAC1 might open a door for the immunomodulation of T1D [83, 84].

Challenges and Conclusions

Insights into the immune system have been increasing at a great rate. A major hurdle in T1D immunotherapy is that the interventions are typically deployed when the immune responses have already peaked. Therefore, the timing of intervention is considered very significant if one is to interfere with the progression of T1D. Other challenges include the lack of adequate endpoints to evaluate outcomes and the difficulties in predicting the outcome of these therapies from animal models to human subjects. Most immunotherapies focus on ways to suppress autoimmunity to restore and preserve the function of insulin-secreting beta cells in the pancreas. Immunotherapy remains a promising approach in search of a cure for T1D.

Compliance with Ethical Standards

Conflict of Interest The authors declare that they have no conflict of interest.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

References

Papers of particular interest, published recently, have been highlighted as:
- • Of importance
- •• Of major importance

1. American Diabetes Association. Diagnosis and classification of diabetes mellitus. Diabetes Care. 2010;33 Suppl 1:S62–9. https://doi.org/10.2337/dc10-S062.
2. Gupta G, de Jesus Andreoli Pinto T, Chellappan DK, Mishra A, Malipeddi H, Dua K. A clinical update on metformin and lung cancer in diabetic patients. Panminerva Med. 2018;60(2):70–5. https://doi.org/10.23736/s0031-0808.18.03394-3.
3. American Diabetes A. Diagnosis and classification of diabetes mellitus. Diabetes Care. 2010;33(Suppl 1):S62–S9. https://doi.org/10.2337/dc10-S062.
4. Atkinson MA. The pathogenesis and natural history of type 1 diabetes. Cold Spring Harb Perspect Med. 2012;2(11). doi:https://doi.org/10.1101/cshperspect.a007641.
5. Eisenbarth GS. Update in type 1 diabetes. J Clin Endocrinol Metab. 2007;92(7):2403–7. https://doi.org/10.1210/jc.2007-0339.
6. Morales J, Schneider D. Hypoglycemia. Am J Med. 2014;127(10 Suppl):S17–24. https://doi.org/10.1016/j.amjmed.2014.07.004.
7. Michels A, Zhang L, Khadra A, Kushner JA, Redondo MJ, Pietropaolo M. Prediction and prevention of type 1 diabetes: update on success of prediction and struggles at prevention. Pediatr Diabetes. 2015;16(7):465–84.
8. Rigby MR, Ehlers MR. Targeted immune interventions for type 1 diabetes: not as easy as it looks! Curr Opin Endocrinol Diabetes Obes. 2014;21(4):271–8. https://doi.org/10.1097/med.0000000000000075.

9. Weigmann B, Franke RK, Daniel C. Immunotherapy in autoimmune type 1 diabetes. Rev Diabet Stud. 2012;9(2–3):68–81. https://doi.org/10.1900/rds.2012.9.68.
10. Durnian JM, Stewart RM, Tatham R, Batterbury M, Kaye SB. Cyclosporin-A associated malignancy. Clin Ophthalmol. 2007;1(4):421–30.
11. Chatenoud L, Warncke K, Ziegler AG. Clinical immunologic interventions for the treatment of type 1 diabetes. Cold Spring Harbor Perspectives in Medicine. 2012;2(8). doi:https://doi.org/10.1101/cshperspect.a007716.
12. Allison AC, Eugui EM. Mycophenolate mofetil and its mechanisms of action. Immunopharmacology. 2000;47(2):85–118. https://doi.org/10.1016/S0162-3109(00)00188-0.
13. Heatwole C, Ciafaloni E. Mycophenolate mofetil for myasthenia gravis: a clear and present controversy. Neuropsychiatr Dis Treat. 2008;4(6):1203–9.
14. Gottlieb PA, Quinlan S, Krause-Steinrauf H, Greenbaum CJ, Wilson DM, Rodriguez H, et al. Failure to preserve beta-cell function with mycophenolate mofetil and daclizumab combined therapy in patients with new-onset type 1 diabetes. Diabetes Care. 2010;33(4):826–32. https://doi.org/10.2337/dc09-1349.
15. Emer JJ, Claire W. Rituximab: a review of dermatological applications. J Clin Aesthet Dermatol. 2009;2(5):29–37.
16. Randall KL. Rituximab in autoimmune diseases. Aust Prescr. 2016;39(4):131–4. https://doi.org/10.18773/austprescr.2016.053.
17. Pescovitz MD, Greenbaum CJ, Bundy B, Becker DJ, Gitelman SE, Goland R, et al. B-lymphocyte depletion with rituximab and β-cell function: two-year results. Diabetes Care. 2014;37(2):453–9. https://doi.org/10.2337/dc13-0626.
18. Mayer E, Holzl M, Ahmadi S, Dillinger B, Pilat N, Fuchs D, et al. CTLA4-Ig immunosuppressive activity at the level of dendritic cell/T cell crosstalk. Int Immunopharmacol. 2013;15(3):638–45. https://doi.org/10.1016/j.intimp.2013.02.007.
19. Vital EM, Emery P. Abatacept in the treatment of rheumatoid arthritis. Ther Clin Risk Manag. 2006;2(4):365–75.
20. Melvin G, Sandhiya S, Subraja K. Belatacept: a worthy alternative to cyclosporine? J Pharmacol Pharmacother. 2012;3(1):90–2. https://doi.org/10.4103/0976-500x.92499.
21. Tooley JE, Waldron-Lynch F, Herold KC. New and future immunomodulatory therapy in type 1 diabetes. Trends Mol Med. 2012;18(3):173–81. https://doi.org/10.1016/j.molmed.2012.01.001.
22. Orban T, Bundy B, Becker DJ, DiMeglio LA, Gitelman SE, Goland R, et al. Co-stimulation modulation with abatacept in patients with recent-onset type 1 diabetes: a randomised, double-blind, placebo-controlled trial. Lancet. 2011;378(9789):412–9. https://doi.org/10.1016/s0140-6736(11)60886-6.
23. Monaco C, Nanchahal J, Taylor P, Feldmann M. Anti-TNF therapy: past, present and future. Int Immunol. 2015;27(1):55–62. https://doi.org/10.1093/intimm/dxu102.
24. Tack CJ, Kleijwegt FS, Van Riel PL, Roep BO. Development of type 1 diabetes in a patient treated with anti-TNF-alpha therapy for active rheumatoid arthritis. Diabetologia. 2009;52(7):1442–4. https://doi.org/10.1007/s00125-009-1381-0.
25. Mastrandrea L, Yu J, Behrens T, Buchlis J, Albini C, Fourtner S, et al. Etanercept treatment in children with new-onset type 1 diabetes: pilot randomized, placebo-controlled, double-blind study. Diabetes Care. 2009;32(7):1244–9. https://doi.org/10.2337/dc09-0054.
26. Penaranda C, Tang Q, Bluestone JA. Anti-CD3 therapy promotes tolerance by selectively depleting pathogenic cells while preserving regulatory T cells. J Immunol (Baltimore, Md : 1950). 2011;187(4):2015–22. https://doi.org/10.4049/jimmunol.1100713.
27. Herold KC, Hagopian W, Auger JA, Poumian-Ruiz E, Taylor L, Donaldson D, et al. Anti-CD3 monoclonal antibody in new-onset type 1 diabetes mellitus. N Engl J Med. 2002;346(22):1692–8. https://doi.org/10.1056/NEJMoa012864.
28. Keymeulen B, Vandemeulebroucke E, Ziegler AG, Mathieu C, Kaufman L, Hale G, et al. Insulin needs after CD3-antibody therapy in new-onset type 1 diabetes. N Engl J Med. 2005;352(25):2598–608. https://doi.org/10.1056/NEJMoa043980.
29. Herold KC, Gitelman S, Greenbaum C, Puck J, Hagopian W, Gottlieb P, et al. Treatment of patients with new onset type 1 diabetes with a single course of anti-CD3 mAb Teplizumab preserves insulin production for up to 5 years. Clin Immunol. 2009;132(2):166–73. https://doi.org/10.1016/j.clim.2009.04.007.
30. Hagopian W, Ferry RJ Jr, Sherry N, Carlin D, Bonvini E, Johnson S, et al. Teplizumab preserves C-peptide in recent-onset type 1 diabetes: two-year results from the randomized, placebo-controlled Protege trial. Diabetes. 2013;62(11):3901–8. https://doi.org/10.2337/db13-0236.
31. Mohty M. Mechanisms of action of antithymocyte globulin: T-cell depletion and beyond. Leukemia. 2007;21(7):1387–94. https://doi.org/10.1038/sj.leu.2404683.
32. Luo X, Herold KC, Miller SD. Immunotherapy of type 1 diabetes: where are we and where should we be going? Immunity. 2010;32(4):488–99. https://doi.org/10.1016/j.immuni.2010.04.002.
33.• Gitelman SE, Gottlieb PA, Felner EI, Willi SM, Fisher LK, Moran A, et al. Antithymocyte globulin therapy for patients with recent-onset type 1 diabetes: 2 year results of a randomised trial. Diabetologia. 2016;59(6):1153–61. https://doi.org/10.1007/s00125-016-3917-4. **This study discusses the findings observed from a 2-year randomised trial on antithymocyte globulin in new-onset type 1 diabetes.**
34. Coppieters KT, Harrison LC, von Herrath MG. Trials in type 1 diabetes: antigen-specific therapies. Clin Immunol. 2013;149(3):345–55. https://doi.org/10.1016/j.clim.2013.02.002.
35. Clemente-Casares X, Tsai S, Huang C, Santamaria P. Antigen-specific therapeutic approaches in type 1 diabetes. Cold Spring Harb Perspect Med. 2012;2(2):a007773. https://doi.org/10.1101/cshperspect.a007773.
36. Zhang ZJ, Davidson L, Eisenbarth G, Weiner HL. Suppression of diabetes in nonobese diabetic mice by oral administration of porcine insulin. Proc Natl Acad Sci U S A. 1991;88(22):10252–6.
37. Harrison LC, Dempsey-Collier M, Kramer DR, Takahashi K. Aerosol insulin induces regulatory CD8 gamma delta T cells that prevent murine insulin-dependent diabetes. J Exp Med. 1996;184(6):2167–74.
38. Skyler JS, Krischer JP, Wolfsdorf J, Cowie C, Palmer JP, Greenbaum C, et al. Effects of oral insulin in relatives of patients with type 1 diabetes: the diabetes prevention trial–type 1. Diabetes Care. 2005;28(5):1068–76.
39. Fourlanos S, Perry C, Gellert SA, Martinuzzi E, Mallone R, Butler J, et al. Evidence that nasal insulin induces immune tolerance to insulin in adults with autoimmune diabetes. Diabetes. 2011;60(4):1237–45. https://doi.org/10.2337/db10-1360.
40. Krischer JP, Schatz DA, Bundy B, Skyler JS, Greenbaum CJ. Writing committee for the type 1 diabetes TrialNet Oral insulin study group, effect of oral insulin on prevention of diabetes in relatives of patients with type 1 diabetes: a randomized clinical trial. JAMA. 2017;318(19):1891–902.
41. Tian J, Atkinson MA, Clare-Salzler M, Herschenfeld A, Forsthuber T, Lehmann PV, et al. Nasal administration of glutamate decarboxylase (GAD65) peptides induces Th2 responses and prevents murine insulin-dependent diabetes. J Exp Med. 1996;183(4):1561–7.
42. Ludvigsson J, Cheramy M, Axelsson S, Pihl M, Akerman L, Casas R. GAD-treatment of children and adolescents with recent-onset type 1 diabetes preserves residual insulin secretion after 30 months. Diabetes Metab Res Rev. 2014;30(5):405–14. https://doi.org/10.1002/dmrr.2503.

43. Wherrett DK, Bundy B, Becker DJ, DiMeglio LA, Gitelman SE, Goland R, et al. Antigen-based therapy with glutamic acid decarboxylase (GAD) vaccine in patients with recent-onset type 1 diabetes: a randomised double-blind trial. Lancet. 2011;378(9788):319–27. https://doi.org/10.1016/s0140-6736(11)60895-7.
44. Ludvigsson J, Hjorth M, Cheramy M, Axelsson S, Pihl M, Forsander G, et al. Extended evaluation of the safety and efficacy of GAD treatment of children and adolescents with recent-onset type 1 diabetes: a randomised controlled trial. Diabetologia. 2011;54(3):634–40. https://doi.org/10.1007/s00125-010-1988-1.
45. Pellegrino MW, Nargund AM, Haynes CM. Signaling the mitochondrial unfolded protein response. Biochim Biophys Acta. 2013;1833(2):410–6. https://doi.org/10.1016/j.bbamcr.2012.02.019.
46.• Juwono J, Martinus RD. Does Hsp60 provide a link between mitochondrial stress and inflammation in diabetes mellitus. J Diabetes Res. 2016;2016:8017571. https://doi.org/10.1155/2016/8017571. **This review summarises the known relationships between the expression of heat shock protein 60 and type 1 diabetes.**
47. Elias D, Markovits D, Reshef T, van der Zee R, Cohen IR. Induction and therapy of autoimmune diabetes in the non-obese diabetic (NOD/Lt) mouse by a 65-kDa heat shock protein. Proc Natl Acad Sci U S A. 1990;87(4):1576–80.
48. Raz I, Avron A, Tamir M, Metzger M, Symer L, Eldor R, et al. Treatment of new-onset type 1 diabetes with peptide DiaPep277 is safe and associated with preserved beta-cell function: extension of a randomized, double-blind, phase II trial. Diabetes Metab Res Rev. 2007;23(4):292–8. https://doi.org/10.1002/dmrr.712.
49. Lazar L, Ofan R, Weintrob N, Avron A, Tamir M, Elias D, et al. Heat-shock protein peptide DiaPep277 treatment in children with newly diagnosed type 1 diabetes: a randomised, double-blind phase II study. Diabetes Metab Res Rev. 2007;23(4):286–91. https://doi.org/10.1002/dmrr.711.
50. Corthay A. How do regulatory T cells work? Scand J Immunol. 2009;70(4):326–36. https://doi.org/10.1111/j.1365-3083.2009.02308.x.
51. Curotto de Lafaille MA, Muriglan S, Sunshine MJ, Lei Y, Kutchukhidze N, Furtado GC, et al. Hyper immunoglobulin E response in mice with monoclonal populations of B and T lymphocytes. J Exp Med. 2001;194(9):1349–59.
52. Karlsson MR, Rugtveit J, Brandtzaeg P. Allergen-responsive CD4+ CD25+ regulatory T cells in children who have outgrown cow's milk allergy. J Exp Med. 2004;199(12):1679–88. https://doi.org/10.1084/jem.20032121.
53. Li Z, Li D, Tsun A, Li B. FOXP3+ regulatory T cells and their functional regulation. Cell Mol Immunol. 2015;12(5):558–65. https://doi.org/10.1038/cmi.2015.10.
54. Tonkin DR, He J, Barbour G, Haskins K. Regulatory T cells prevent transfer of type 1 diabetes in NOD mice only when their antigen is present in vivo. J Immunol (Baltimore, Md : 1950). 2008;181(7):4516–22.
55. Cabrera SM, Rigby MR, Mirmira RG. Targeting regulatory T cells in the treatment of type 1 diabetes mellitus. Curr Mol Med. 2012;12(10):1261–72.
56. Okubo Y, Torrey H, Butterworth J, Zheng H, Faustman DL. Treg activation defect in type 1 diabetes: correction with TNFR2 agonism. Clin Transl Immunol. 2016;5(1):e56. https://doi.org/10.1038/cti.2015.43.
57. Wu L, Dakic A. Development of dendritic cell system. Cell Mol Immunol. 2004;1(2):112–8.
58. Shang N, Figini M, Shangguan J, Wang B, Sun C, Pan L, et al. Dendritic cells based immunotherapy. Am J Cancer Res. 2017;7(10):2091–102.
59. Marin-Gallen S, Clemente-Casares X, Planas R, Pujol-Autonell I, Carrascal J, Carrillo J, et al. Dendritic cells pulsed with antigen-specific apoptotic bodies prevent experimental type 1 diabetes. Clin Exp Immunol. 2010;160(2):207–14. https://doi.org/10.1111/j.1365-2249.2009.04082.x.
60. Creusot RJ, Giannoukakis N, Trucco M, Clare-Salzler MJ, Fathman CG. It's time to bring dendritic cell therapy to type 1 diabetes. Diabetes. 2014;63(1):20–30. https://doi.org/10.2337/db13-0886.
61. Giannoukakis N, Phillips B, Finegold D, Harnaha J, Trucco M. Phase I (safety) study of autologous tolerogenic dendritic cells in type 1 diabetic patients. Diabetes Care. 2011;34(9):2026–32. https://doi.org/10.2337/dc11-0472.
62. Furtado GC, Curotto de Lafaille MA, Kutchukhidze N, Lafaille JJ. Interleukin 2 signaling is required for CD4(+) regulatory T cell function. J Exp Med. 2002;196(6):851–7.
63. Bachmann MF, Oxenius A. Interleukin 2: from immunostimulation to immunoregulation and back again. EMBO Rep. 2007;8(12):1142–8. https://doi.org/10.1038/sj.embor.7401099.
64. Williams MA, Tyznik AJ, Bevan MJ. Interleukin-2 signals during priming are required for secondary expansion of CD8+ memory T cells. Nature. 2006;441(7095):890–3. https://doi.org/10.1038/nature04790.
65. Grinberg-Bleyer Y, Baeyens A, You S, Elhage R, Fourcade G, Gregoire S, et al. IL-2 reverses established type 1 diabetes in NOD mice by a local effect on pancreatic regulatory T cells. J Exp Med. 2010;207(9):1871–8.
66. Diaz-de-Durana Y, Lau J, Knee D, Filippi C, Londei M, McNamara P, et al. IL-2 immunotherapy reveals potential for innate beta cell regeneration in the non-obese diabetic mouse model of autoimmune diabetes. PLoS One. 2013;8(10):e78483. https://doi.org/10.1371/journal.pone.0078483.
67. Luzina IG, Keegan AD, Heller NM, Rook GA, Shea-Donohue T, Atamas SP. Regulation of inflammation by interleukin-4: a review of "alternatives". J Leukoc Biol. 2012;92(4):753–64. https://doi.org/10.1189/jlb.0412214.
68. Rapoport MJ, Jaramillo A, Zipris D, Lazarus AH, Serreze DV, Leiter EH, et al. Interleukin 4 reverses T cell proliferative unresponsiveness and prevents the onset of diabetes in nonobese diabetic mice. J Exp Med. 1993;178(1):87–99.
69. Cameron MJ, Arreaza GA, Zucker P, Chensue SW, Strieter RM, Chakrabarti S, et al. IL-4 prevents insulitis and insulin-dependent diabetes mellitus in nonobese diabetic mice by potentiation of regulatory T helper-2 cell function. J Immunol (Baltimore, Md : 1950). 1997;159(10):4686–92.
70. Cameron MJ, Arreaza GA, Waldhauser L, Gauldie J, Delovitch TL. Immunotherapy of spontaneous type 1 diabetes in nonobese diabetic mice by systemic interleukin-4 treatment employing adenovirus vector-mediated gene transfer. Gene Ther. 2000;7(21):1840–6. https://doi.org/10.1038/sj.gt.3301309.
71. Elhelu MA. The role of macrophages in immunology. J Natl Med Assoc. 1983;75(3):314–7.
72. Gensel JC, Kopper TJ, Zhang B, Orr MB, Bailey WM. Predictive screening of M1 and M2 macrophages reveals the immunomodulatory effectiveness of post spinal cord injury azithromycin treatment. Sci Rep. 2017;7:40144. https://doi.org/10.1038/srep40144.
73. Martinez FO, Gordon S. The M1 and M2 paradigm of macrophage activation: time for reassessment. F1000 Prime Rep. 2014;6:13. https://doi.org/10.12703/p6-13.
74. Parsa R, Andresen P, Gillett A, Mia S, Zhang XM, Mayans S, et al. Adoptive transfer of immunomodulatory M2 macrophages prevents type 1 diabetes in NOD mice. Diabetes. 2012;61(11):2881–92. https://doi.org/10.2337/db11-1635.
75. Culina S, Boitard C, Mallone R. Antigen-based immune therapeutics for type 1 diabetes: magic bullets or ordinary blanks? Clin Dev Immunol. 2011;2011:286248–15. https://doi.org/10.1155/2011/286248.
76. Long SA, Rieck M, Sanda S, Bollyky JB, Samuels PL, Goland R, et al. Rapamycin/IL-2 combination therapy in patients with type 1

diabetes augments Tregs yet transiently impairs beta-cell function. Diabetes. 2012;61(9):2340–8. https://doi.org/10.2337/db12-0049.

77. Matthews JB, Staeva TP, Bernstein PL, Peakman M, von Herrath M. Developing combination immunotherapies for type 1 diabetes: recommendations from the ITN-JDRF type 1 diabetes combination therapy assessment group. Clin Exp Immunol. 2010;160(2):176–84. https://doi.org/10.1111/j.1365-2249.2010.04153.x.
78. Lord S, Greenbaum CJ. Disease modifying therapies in type 1 diabetes: where have we been, and where are we going? Pharmacol Res. 2015;98:3–8. https://doi.org/10.1016/j.phrs.2015.02.002.
79. Perron H, Germi R, Bernard C, Garcia-Montojo M, Deluen C, Farinelli L, et al. Human endogenous retrovirus type W envelope expression in blood and brain cells provides new insights into multiple sclerosis disease. Mult Scler. 2012;18(12):1721–36. https://doi.org/10.1177/1352458512441381.
80. Faucard R, Madeira A, Gehin N, Authier FJ, Panaite PA, Lesage C, et al. Human endogenous retrovirus and neuroinflammation in chronic inflammatory demyelinating polyradiculoneuropathy. EBioMedicine. 2016;6:190–8. https://doi.org/10.1016/j.ebiom.2016.03.001.
81.•• Levet S, Medina J, Joanou J, Demolder A, Queruel N, Reant K et al. An ancestral retroviral protein identified as a therapeutic target in type-1 diabetes. JCI Insight. 2017;2(17). doi:https://doi.org/10.1172/jci.insight.94387. **The study reports on the potential of human endogenous retroviruses as therapeutic targets in the potential cure of type 1 diabetes.**
82. Curtin F, Bernard C, Levet S, Perron H, Porchet H, Medina J, et al. A new therapeutic approach for type 1 diabetes: rationale for GNbAC1, an anti-HERV-W-Env monoclonal antibody. Diabetes Obes Metab. 2018;20(9):2075–84. https://doi.org/10.1111/dom.13357.
83. Rewers M, Gottlieb P. Immunotherapy for the prevention and treatment of type 1 diabetes: human trials and a look into the future. Diabetes Care. 2009;32(10):1769–82. https://doi.org/10.2337/dc09-0374.
84. von Herrath M, Peakman M, Roep B. Progress in immune-based therapies for type 1 diabetes. Clin Exp Immunol. 2013;172(2):186–202. https://doi.org/10.1111/cei.12085.

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