Published in final edited form as:

Biochemistry. 2019 October 08; 58(40): 4107–4111. doi:10.1021/acs.biochem.9b00707.

Anti-CD3 antibody for the prevention of type 1 diabetes - a story of perseverance

Jason Gaglia^{1,2}, Stephan Kissler^{1,2,*}

¹Section for Immunobiology, Joslin Diabetes Center, Boston, MA

²Department of Medicine, Harvard Medical School, Boston, MA

Abstract

Type 1 diabetes (T1D) is an autoimmune disease characterized by insulin deficiency. Ever since the discovery of insulin almost 100 years ago, patients with T1D have relied on multiple daily insulin injections to survive an otherwise deadly disease. Despite decades of research and clinical trials, no treatment exists yet to prevent or cure T1D. A recent prevention trial using the anti-CD3 antibody teplizumab in individuals at high risk of developing T1D has provided the first evidence that a safe and transient intervention may be able to delay disease. In this perspective, we review the 40-year long history of anti-CD3 and discuss how this antibody became a candidate for the treatment of autoimmune diabetes. The path that lead to its use in this latest clinical trial for T1D has been winding and strewn with set-backs. The molecular actions of anti-CD3 antibody that targets T lymphocytes are well understood, but its systemic effect on immune function have proven more difficult to unravel. Moreover, preclinical data suggested that the utility of anti-CD3 for the prevention of T1D may be limited. However, the latest clinical data are encouraging and exemplify how a basic discovery can, decades later and with much perseverance, become a promising candidate therapeutic.

In the late 1970s, Schlossman and colleagues generated a series of monoclonal antibodies against cell surface antigens of human T lymphocytes (Kung et al., 1979). These novel reagents became invaluable tools for the discovery of many fundamental principles of T cell development and function (Reinherz et al., 1980; Meuer et al., 1982). Notably, the antibody described in 1979 and termed Ortho Kung T3 (OKT3) or anti-T3 binds to a molecule on the surface of T cells that is involved in antigen recognition (Reinherz et al., 1980). This molecule, now known as CD3 epsilon, is part of the T cell receptor (TCR) complex that comprises the TCR alpha and beta chains and six CD3 molecules, including two CD3 epsilon chains anchored in the cell membrane. An initial report on the properties of OKT3 indicated that it could inhibit the cytotoxic activity of T cells (Reinherz et al., 1980). Subsequent experiments demonstrated that this anti-CD3 antibody was also a potent mitogen and could activate T cells at very low concentrations (Van Wauwe et al., 1980).

*Correspondence: stephan.kissler@joslin.harvard.edu.

Conflict of Interests

J.L.G. is an employee of Semma Therapeutics and has received support for performing clinical trials from the National Institutes of Health, Janssen Research & Development LLC, Caladrius Biosciences and Avotres Inc.

S.K. declares that he has no conflict of interests.

The mechanism underlying the effects of anti-CD3 relates to the function of the TCR complex. T cells recognize antigen using the TCR that interacts with peptide antigen embedded in a major histocompatibility complex (MHC) molecule on the surface of another cell. The productive interaction between a TCR and a specific peptide-MHC leads to sequential phosphorylation events along all six CD3 chains that are in close association with the TCR alpha and beta chains. The degree of CD3 phosphorylation depends on the clustering of multiple TCR complexes and on the duration of their interaction with peptide-MHC. The activation of multiple signaling pathways downstream of the TCR complex results in many changes in T cell behavior. TCR signaling alters cell metabolism, induces cell division and drives effector functions that include cytolytic activity and the secretion of signaling molecules such as interleukins, interferon (IFN)- γ , tumor necrosis factor (TNF)- α or transforming growth factor (TGF)- β .

The original demonstration that high concentrations of OKT3 inhibit the cytotoxic activity of T cells (Reinherz et al., 1980) can be explained by the antibody's allosteric hindrance of interactions between the TCR complex and peptide-MHC molecules. By abrogating TCR-peptide-MHC interactions, anti-CD3 prevents cytolytic T cells from interacting with their target cell. This inhibitory property led to the remarkably rapid use of OKT3 in the clinic. The first clinical trial with OKT3 came only two years after the original publication describing this antibody. Based on its ability to inhibit the lytic activity of T cells, OKT3 was administered to kidney transplant patients to prevent acute graft rejection that is driven by T cells (Cosimi et al., 1981).

Soon after this first clinical trial, another property of anti-CD3, namely its specificity for T cells, was taken advantage of in the setting of bone-marrow transplantation. OKT3 was used to deplete T cells from bone-marrow samples prior to their transplantation into patients. This approach reduced the risk of graft-versus-host-disease, which is caused by donor T cells attacking cells and organs in the transplant recipient (Prentice et al., 1982; Filipovich et al., 1982).

These earliest clinical results paved the way for a wider use of anti-CD3 to prevent acute graft rejection. The antibody was tested with success in liver and heart transplant patients (Cosimi et al., 1987; Gilbert et al., 1987). OKT3 (muromonab-CD3) was approved by the U.S. Food and Drug Administration (FDA) in 1985, the first monoclonal antibody to be approved as a drug in man. However, two problems emerged that prohibited the wider clinical adoption of OKT3. First, injection of high doses of anti-CD3 triggered a potentially life-threatening cytokine release syndrome (CRS) characterized by fatigue, headache, muscle and joint pain, rash, nausea, tachycardia, hypotension and fever (Cosimi et al., 1981; Ortho Multicenter Transplant Study Group, 1985; Chatenoud et al., 1989b). An anti-CD3 antibody similar to OKT3 was generated against mouse T cells (Leo et al., 1987), and its injection in mice replicated the syndrome observed in patients (Ferran et al., 1990). Having a mouse model of anti-CD3-induced CRS allowed investigators to better study and understand this adverse event. CRS was linked to the mitogenic activity of anti-CD3 and to a massive release of TNF- α . Both of these were found to depend on Fc-receptors on the surface of monocytes that bind the 'tail-end' of anti-CD3 in an antigen-non-specific manner (Vossen et al., 1995). Fc-receptor binding crosslinks the antibody and vastly increases its stimulatory

capacity. Blocking Fc-receptors at the time of anti-CD3 injection prevented CRS in mice. Importantly, inhibiting Fc-receptor binding did not impair the immunosuppressive effect of anti-CD3 (Vossen et al., 1995). These observations lead to the development of Fc-receptor-non-binding anti-CD3 (Cole et al., 1997). By mutating residues in the Fc region of the antibody, anti-CD3 could be prevented from causing acute T cell activation and TNF- α secretion without losing the antibody's therapeutic potential. The second problem with using an antibody of mouse origin in patients was the appearance of anti-OKT3 antibodies, a natural immune response to a protein of foreign (mouse) origin (Jaffers et al., 1986). Host antibodies against the therapeutic anti-CD3 greatly diminished its potency upon subsequent administrations. To counter this anti-drug antibody (ADA) response, investigators engineered the original mouse OKT3 to resemble a human antibody (Woodle et al., 1992). So-called humanized OKT3 was much better tolerated and did not elicit ADAs, permitting repeated dosing (Alegre et al., 1994; Woodle et al., 1999). Ultimately, these modifications gave rise to huOKT3 γ 1ala-ala, a humanized Fc-non-binding version of OKT3 now known as teplizumab (Xu et al., 2000).

The immunosuppressive properties of OKT3 prompted researchers to speculate that the antibody may be effective in treating autoimmunity. When an anti-mouse CD3 antibody that mimics OKT3 was tested in a mouse skin transplantation model (Hirsch et al., 1988), the results indicated that anti-CD3 treatment had a sustained immunosuppressive effect. Shortly thereafter, anti-CD3 was used to modify the course of autoimmune diabetes in the nonobese diabetic (NOD) mouse, a spontaneous animal model for T1D. Neonatal injection of anti-CD3 completely prevented diabetes in NOD mice (Hayward and Shreiber, 1989). Although the data were promising, Bluestone and colleagues cautioned against the use of anti-CD3 to treat human autoimmunity because of its potentially severe side effects (Hirsch et al., 1989). In response, Chatenoud and Bach, who went on to lead efforts to test anti-CD3 in T1D, argued that careful dosing of anti-CD3 may yet be of use to intervene in diabetes and even reported the first use of OKT3 in 7 patients with insulin-dependent diabetes (Chatenoud et al., 1989a). In a critically important study, Chatenoud and Bach demonstrated a few years later that anti-CD3 could reverse disease when administered shortly after the onset of diabetes in NOD mice (Chatenoud et al., 1994). The same investigators subsequently showed that anti-CD3 was only effective when given shortly before or after disease onset, but not in young pre-diabetic animals (Chatenoud et al., 1997). Together with a prior report that anti-CD3 could prevent diabetes in an drug-induced mouse model (Herold et al., 1992), these studies lay the foundation of clinical trials for anti-CD3 in T1D.

Of note, another similar anti-CD3 was developed in parallel to teplizumab with comparable properties. Waldmann and colleagues generated a rat antibody against human CD3 and engineered it as a hybrid human/rat molecule to make it less immunogenic in patients (Routledge et al., 1991). This antibody was later modified to prevent its glycosylation, which rendered it Fc-receptor non-binding and also abrogated complement binding (Friend et al., 1999). The resulting humanized antibody became known as otelexizumab (Hale et al., 2010). Both teplizumab and otelexizumab were tested in multiple randomized placebo-controlled clinical trials for the treatment of T1D.

The first trial with teplizumab yielded very promising data, suggesting preservation of insulin production two years after T1D onset when patients were treated within 6 weeks of diagnosis (Herold et al., 2002, 2005). A subsequent trial with repeat dosing attempted to improve on this positive outcome. However, the design deviated from the original protocol by leaving out a drug filter that caused a higher dose to be administered to patients. This led to serious adverse events and necessitated the trial to be terminated prematurely (Daifotis et al., 2013). Notwithstanding this set-back, further attempts at testing the efficacy of teplizumab in new onset T1D patients followed (Sherry et al., 2011; Herold et al., 2013b; a; Hagopian et al., 2013). The results of these later trials were suggestive of efficacy in specific subgroups of patients, particularly younger patients and those with better glucose control at recruitment. A confounding issue in these larger trials was the choice of a different primary endpoint that had not been used or validated previously to evaluate efficacy compared with earlier trials (Daifotis et al., 2013). Even though these trials could not be described as clear successes, the data were sufficiently encouraging for the lead investigators to plan additional improved trials.

Initial trials with otelexizumab provided additional support for anti-CD3 therapy. The first otelexizumab trial in new onset T1D patients indicated better preservation of insulin secretion 18 months after diagnosis (Keymeulen et al., 2005). Follow-up studies indicated that the positive effects of a short intervention with otelexizumab were durable up to 4 years (Keymeulen et al., 2010). However, the relatively high dose of antibody used in these trials elicited Epstein Barr virus reactivation in some patients (Keymeulen et al., 2005). Consequently, investigators decided to utilize a dose that was almost 20-fold lower in subsequent trials. Disappointingly, these larger phase III trials showed no efficacy and were terminated prematurely (Aronson et al., 2014; Ambery et al., 2014).

The consensus from more than a decade of clinical trials with anti-CD3 was that this intervention showed efficacy at higher doses, as measured by preservation of insulin secretion and a lesser requirement for exogenous insulin when patients were treated close to T1D onset. Significantly, results seemed best in patients that were younger, had better glucose control at the time of treatment and were generally at an earlier stage of disease (Sherry et al., 2011; Hagopian et al., 2013; Herold et al., 2013b). This is consistent with data from mouse models where anti-CD3 treatment was most effective shortly before or after diabetes onset (Chatenoud et al., 1997). An additional consideration is that high doses of anti-CD3 had substantial potential for adverse effects related to cytokine release, but the dose-escalation used in teplizumab trials showed that acute symptoms could be averted in > 90 % of patients (Sherry et al., 2011; Daifotis et al., 2013). These lessons were integrated into the design of the latest trial with teplizumab (Herold et al., 2019). This trial enrolled mostly children (72% of patients were 18 years of age or younger) and, critically, intervened before the diagnosis of T1D. These individuals were identified as having a very high risk of progressing to T1D, based on several criteria. First, all individuals had relatives with T1D and therefore already had a 5–10-fold increased risk of disease. Second, all of them were positive for two or more autoantibodies associated with progression to T1D (Ziegler et al., 2013). Third, these individuals already had evidence of abnormal glucose tolerance based on an oral glucose-tolerance test. In combination, these criteria provided near-certainty that most if not all of these individuals would develop T1D within a few years (Insel et al.,

2015). Choosing trial participants in this very select group ensured that 1) the treatment would be administered only to individuals predicted to develop T1D and 2) the intervention would be administered at an even earlier stage of disease than in prior trials. Including mostly children was also consistent with evidence that anti-CD3 was more efficacious in younger individuals (Sherry et al., 2011; Hagopian et al., 2013; Herold et al., 2013b). Finally, teplizumab was administered over 14 days with escalating doses to decrease adverse events related to cytokine release. Overall, the trial was successful in delaying the onset of T1D, with a median time to diagnosis that was 24 months longer in the treated group. The treatment was well tolerated with the expected transient adverse events of rash and lymphopenia being the most common. Treatment still had to be stopped in a small fraction of patients (7%) due to laboratory abnormalities. Results to date suggest that fewer individuals progressed to T1D after teplizumab treatment, though whether the treatment prevented disease altogether in some patients or merely delayed their disease onset will need to be confirmed in longer follow-up analyses. Of note, the treatment regimen that spans two weeks necessitated repeated testing for adverse events, making it a burdensome and costly intervention. Despite dose escalation, adverse events that forced some patients to stop treatment could not be avoided. Notwithstanding, the overall positive outcome of this latest trial, on balance, would justify the use of teplizumab for the prevention of type 1 diabetes, provided it is targeted to those individuals in whom efficacy is predicted to be highest based on HLA genotype and specific serological markers (Herold et al., 2019).

In conclusion, teplizumab may be the first treatment available to delay or even prevent T1D. The results of the latest trial are the culmination of 40 years of research and clinical trials with an antibody that was initially generated to unravel the basics of T cell biology. The remarkable evolution from research reagents to therapeutic was not without hurdles, but this slow and difficult progress is perhaps representative for the winding path that leads to most therapeutics. The new clinical data with teplizumab are promising. However, several aspects of this preventive treatment remain to be confirmed. First, a follow-up of these trial participants will reveal if the effects of teplizumab are long-lasting. Second, it will be important to test if teplizumab is similarly effective in individuals at lower risk of T1D, so that prevention could be expanded to a broader population. In this regard, efforts are underway to identify at-risk individuals in the general population using systematic genetic and serological screening (Winkler et al., 2019), and this may allow teplizumab to be administered more widely for the prevention of T1D in the future. Finally, an intervention that delays T1D would be most welcome, but much more research is needed to prevent disease altogether or to intervene in patients whose disease was not predicted in time for prevention.

Acknowledgments

The authors are supported by funds from the National Institutes of Health (R01DK120445 and UC4DK116280 to S.K.) the Harvard Stem Cell Institute (DP-0167017 to S.K.) and JDRF (2-SRA-2018-499-S-B to S.K. and 5-ECR-2016-186-A-N to J.L.G.), and work in the authors' laboratories is also supported in part by a Diabetes Research Center award to the Joslin Diabetes Center (P30DK036836).

References

- Alegre ML, Peterson LJ, Xu D, Sattar HA, Jeyarajah DR, Kowalkowski K, Thistlethwaite JR, Zivin RA, Jolliffe L, and Bluestone JA. 1994 A non-activating "humanized" anti-CD3 monoclonal antibody retains immunosuppressive properties in vivo. *Transplantation*. 57:1537–43. [PubMed: 8009586]
- Ambery P, Donner TW, Biswas N, Donaldson J, Parkin J, and Dayan CM. 2014 Efficacy and safety of low-dose oteelixumab anti-CD3 monoclonal antibody in preserving C-peptide secretion in adolescent type 1 diabetes: DEFEND-2, a randomized, placebo-controlled, double-blind, multi-centre study. *Diabet. Med* 31:399–402. doi:10.1111/dme.12361. [PubMed: 24236828]
- Aronson R, Gottlieb PA, Christiansen JS, Donner TW, Bosi E, Bode BW, Pozzilli P, and DEFEND Investigator Group. 2014 Low-Dose Oteelixumab Anti-CD3 Monoclonal Antibody DEFEND-1 Study: Results of the Randomized Phase III Study in Recent-Onset Human Type 1 Diabetes. *Diabetes Care*. 37:2746–2754. doi:10.2337/dc13-0327. [PubMed: 25011949]
- Chatenoud L, Ferran C, and Bach JF. 1989a In-vivo anti-CD3 treatment of autoimmune patients. *Lancet* (London, England). 2:164. doi:10.1016/s0140-6736(89)90226-2.
- Chatenoud L, Ferran C, Reuter A, Legendre C, Gevaert Y, Kreis H, Franchimont P, and Bach JF. 1989b Systemic Reaction to the Anti-T-Cell Monoclonal Antibody OKT3 in Relation to Serum Levels of Tumor Necrosis Factor and Interferon- α . *N. Engl. J. Med* 320:1420–1421. doi:10.1056/NEJM198905253202117. [PubMed: 2785642]
- Chatenoud L, Primo J, and Bach JF. 1997 CD3 antibody-induced dominant self tolerance in overtly diabetic NOD mice. *J. Immunol* 158:2947–54. [PubMed: 9058834]
- Chatenoud L, Thervet E, Primo J, and Bach JF. 1994 Anti-CD3 antibody induces long-term remission of overt autoimmunity in nonobese diabetic mice. *Proc. Natl. Acad. Sci* 91:123–127. doi:10.1073/pnas.91.1.123. [PubMed: 8278351]
- Cole MS, Anasetti C, and Tso JY. 1997 Human IgG2 variants of chimeric anti-CD3 are nonmitogenic to T cells. *J. Immunol* 159:3613–21. [PubMed: 9317161]
- Cosimi AB, BURTON RC, COLVIN RB, GOLDSTEIN G, DELMONICO FL, LAQUAGLIA MP, TOLKOFF-RUBIN N, RUBIN RH, HERRIN JT, and RUSSELL PS. 1981 TREATMENT OF ACUTE RENAL ALLOGRAFT REJECTION WITH OKT3 MONOCLONAL ANTIBODY. *Transplantation*. doi:10.1097/00007890-198112000-00018.
- Cosimi AB, CHO SI, DELMONICO FL, KAPLAN MM, ROHRER RJ, and JENKINS RL. 1987 A RANDOMIZED CLINICAL TRIAL COMPARING OKT3 AND STEROIDS FOR TREATMENT OF HEPATIC ALLOGRAFT REJECTION. *Transplantation*. 43:91–94. doi: 10.1097/00007890-198701000-00020. [PubMed: 3099442]
- Daifotis AG, Koenig S, Chatenoud L, and Herold KC. 2013 Anti-CD3 clinical trials in type 1 diabetes mellitus. *Clin. Immunol* 149:268–278. doi:10.1016/j.clim.2013.05.001. [PubMed: 23726024]
- Ferran C, Sheehan K, Dy M, Schreiber R, Merite S, Landais P, Noel LH, Grau G, Bluestone J, and Bach JF. 1990 Cytokine-related syndrome following injection of anti-CD3 monoclonal antibody: further evidence for transient in vivo T cell activation. *Eur. J. Immunol* 20:509–15. doi:10.1002/eji.1830200308. [PubMed: 2138557]
- Filipovich AH, Ramsay NKC, Warkentin PI, Mcglave PB, Goldstein G, and Kersey JH. 1982 PRETREATMENT OF DONOR BONE MARROW WITH MONOCLONAL ANTIBODY OKT3 FOR PREVENTION OF ACUTE GRAFT-VERSUS-HOST DISEASE IN ALLOGENEIC HISTOCOMPATIBLE BONE-MARROW TRANSPLANTATION. *Lancet*. doi:10.1016/S0140-6736(82)92840-9.
- Friend PJ, Hale G, Chatenoud L, Rebello P, Bradley J, Thiru S, Phillips JM, and Waldmann H. 1999 PHASE I STUDY OF AN ENGINEERED AGLYCOSYLATED HUMANIZED CD3 ANTIBODY IN RENAL TRANSPLANT REJECTION1. *Transplantation*. 68:1632–1637. doi: 10.1097/00007890-199912150-00005. [PubMed: 10609938]
- Gilbert EM, Dewitt CW, Eiswirth CC, Renlund DG, Menlove RL, Freedman LA, Herrick CM, Gay WA, and Bristow MR. 1987 Treatment of refractory cardiac allograft rejection with OKT3 monoclonal antibody. *Am. J. Med* 82:202–206. doi:10.1016/0002-9343(87)90056-8.

- Hagopian W, Ferry RJ, Sherry N, Carlin D, Bonvini E, Johnson S, Stein KE, Koenig S, Daifotis AG, Herold KC, and Ludvigsson J. 2013 Teplizumab Preserves C-Peptide in Recent-Onset Type 1 Diabetes: Two-Year Results From the Randomized, Placebo-Controlled Protege Trial. *Diabetes*. 62:3901–3908. doi:10.2337/db13-0236. [PubMed: 23801579]
- Hale G, Rebello P, Al Bakir I, Bolam E, Wiczling P, Jusko WJ, Vandemeulebroucke E, Keymeulen B, Mathieu C, Ziegler A-G, Chatenoud L, and Waldmann H. 2010 Pharmacokinetics and Antibody Responses to the CD3 Antibody Otelixizumab Used in the Treatment of Type 1 Diabetes. *J. Clin. Pharmacol* 50:1238–1248. doi:10.1177/0091270009356299. [PubMed: 20147616]
- Hayward AR, and Shreiber M. 1989 Neonatal injection of CD3 antibody into nonobese diabetic mice reduces the incidence of insulinitis and diabetes. *J. Immunol* 143:1555–9. [PubMed: 2527266]
- Herold KC, Bluestone JA, Montag AG, Parih A, Wiegner A, Gress RE, and Hirsch R. 1992 Prevention of Autoimmune Diabetes With Nonactivating Anti-CD3 Monoclonal Antibody. *Diabetes*. 41:385–391. doi:10.2337/diab.41.3.385. [PubMed: 1532369]
- Herold KC, Bundy BN, Long SA, Bluestone JA, DiMeglio LA, Dufort MJ, Gitelman SE, Gottlieb PA, Krischer JP, Linsley PS, Marks JB, Moore W, Moran A, Rodriguez H, Russell WE, Schatz D, Skyler JS, Tsalikian E, Wherrett DK, Ziegler A-G, and Greenbaum CJ. 2019 An Anti-CD3 Antibody, Teplizumab, in Relatives at Risk for Type 1 Diabetes. *N. Engl. J. Med* 10.1056/NEJMoa1902226.
- Herold KC, Gitelman SE, Ehlers MR, Gottlieb PA, Greenbaum CJ, Hagopian W, Boyle KD, Keyes-Elstein L, Aggarwal S, Phippard D, Sayre PH, McNamara J, Bluestone JA, and AbATE Study Team. 2013a Teplizumab (anti-CD3 mAb) treatment preserves C-peptide responses in patients with new-onset type 1 diabetes in a randomized controlled trial: metabolic and immunologic features at baseline identify a subgroup of responders. *Diabetes*. 62:3766–74. doi:10.2337/db13-0345. [PubMed: 23835333]
- Herold KC, Gitelman SE, Masharani U, Hagopian W, Bisikirska B, Donaldson D, Rother K, Diamond B, Harlan DM, and Bluestone JA. 2005 A single course of anti-CD3 monoclonal antibody hOKT3gamma1(Ala-Ala) results in improvement in C-peptide responses and clinical parameters for at least 2 years after onset of type 1 diabetes. *Diabetes*. 54:1763–9. doi:10.2337/diabetes.54.6.1763. [PubMed: 15919798]
- Herold KC, Gitelman SE, Willi SM, Gottlieb PA, Waldron-Lynch F, Devine L, Sherr J, Rosenthal SM, Adi S, Jalaludin MY, Michels AW, Dziura J, and Bluestone JA. 2013b Teplizumab treatment may improve C-peptide responses in participants with type 1 diabetes after the new-onset period: a randomised controlled trial. *Diabetologia*. 56:391–400. doi:10.1007/s00125-012-2753-4. [PubMed: 23086558]
- Herold KC, Hagopian W, Auger JA, Poumian-Ruiz E, Taylor L, Donaldson D, Gitelman SE, Harlan DM, Xu D, Zivin RA, and Bluestone JA. 2002 Anti-CD3 Monoclonal Antibody in New-Onset Type 1 Diabetes Mellitus. *N. Engl. J. Med* 346:1692–1698. doi:10.1056/NEJMoa012864. [PubMed: 12037148]
- Hirsch R, Eckhaus M, Auchincloss H, Sachs DH, and Bluestone JA. 1988 Effects of in vivo administration of anti-T3 monoclonal antibody on T cell function in mice. I. Immunosuppression of transplantation responses. *J. Immunol* 140:3766–72. [PubMed: 3286764]
- Hirsch R, Gress RE, and Bluestone JA. 1989 Anti-CD3 antibody for autoimmune disease, a cautionary note. *Lancet (London, England)*. 1:1390. doi:10.1016/s0140-6736(89)92835-3.
- Insel RA, Dunne JL, Atkinson MA, Chiang JL, Dabelea D, Gottlieb PA, Greenbaum CJ, Herold KC, Krischer JP, Lernmark Å, Ratner RE, Rewers MJ, Schatz DA, Skyler JS, Sosenko JM, and Ziegler A-G. 2015 Staging Presymptomatic Type 1 Diabetes: A Scientific Statement of JDRF, the Endocrine Society, and the American Diabetes Association. *Diabetes Care*. 38:1964–1974. doi:10.2337/dc15-1419. [PubMed: 26404926]
- Jaffers GJ, Fuller TC, Cosimi AB, Russell PS, Winn HJ, and Colvin RB. 1986 ANTI-IDIOTYPIC AND NON-ANTI-IDIOTYPIC ANTIBODIES TO OKT3 ARISING DESPITE INTENSE IMMUNOSUPPRESSION. *Transplantation*. 41:572–578. doi:10.1097/00007890-198605000-00004. [PubMed: 3085297]
- Keymeulen B, Vandemeulebroucke E, Ziegler AG, Mathieu C, Kaufman L, Hale G, Gorus F, Goldman M, Walter M, Candon S, Schandene L, Crenier L, De Block C, Seigneurin J-M, De Pauw P, Pierard D, Weets I, Rebello P, Bird P, Berrie E, Frewin M, Waldmann H, Bach J-F, Pipeleers D,

- and Chatenoud L. 2005 Insulin Needs after CD3-Antibody Therapy in New-Onset Type 1 Diabetes. *N. Engl. J. Med* 352:2598–2608. doi:10.1056/NEJMoa043980. [PubMed: 15972866]
- Keymeulen B, Walter M, Mathieu C, Kaufman L, Gorus F, Hilbrands R, Vandemeulebroucke E, Van de Velde U, Crenier L, De Block C, Candon S, Waldmann H, Ziegler AG, Chatenoud L, and Pipeleers D. 2010 Four-year metabolic outcome of a randomised controlled CD3-antibody trial in recent-onset type 1 diabetic patients depends on their age and baseline residual beta cell mass. *Diabetologia*. 53:614–23. [PubMed: 20225393]
- Kung PC, Goldstein G, Reinherz EL, and Schlossman SF. 1979 Monoclonal antibodies defining distinctive human T cell surface antigens. *Science* (80-.). doi:10.1126/science.314668.
- Leo O, Foo M, Sachs DH, Samelson LE, and Bluestone JA. 1987 Identification of a monoclonal antibody specific for a murine T3 polypeptide. *Proc. Natl. Acad. Sci* 84:1374–1378. doi:10.1073/pnas.84.5.1374. [PubMed: 2950524]
- Meuer SC, Hussey RE, Hodgdon JC, Hercend T, Schlossman SF, and Reinherz EL. 1982 Surface structures involved in target recognition by human cytotoxic T lymphocytes. *Science* (80-.). doi: 10.1126/science.6981845.
- Ortho Multicenter Transplant Study Group. 1985 A Randomized Clinical Trial of OKT3 Monoclonal Antibody for Acute Rejection of Cadaveric Renal Transplants. *N. Engl. J. Med* 313:337–342. doi: 10.1056/NEJM198508083130601. [PubMed: 2861567]
- Prentice HG, Janossy G, Skeggs D, Blacklock HA, Bradstock KF, Goldstein G, and Hoffbrand AV. 1982 USE OF ANTI-T-CELL MONOCLONAL ANTIBODY OKT3 TO PREVENT ACUTE GRAFT-VERSUS-HOST DISEASE IN ALLOGENEIC BONE-MARROW TRANSPLANTATION FOR ACUTE LEUKAEMIA. *Lancet*. doi:10.1016/S0140-6736(82)92619-8.
- Reinherz EL, Hussey RE, and Schlossman SF. 1980 A monoclonal antibody blocking human T cell function. *Eur. J. Immunol* doi:10.1002/eji.1830101006.
- Routledge EG, Lloyd I, Gorman SD, Clark M, and Waldmann H. 1991 A humanized monovalent CD3 antibody which can activate homologous complement. *Eur. J. Immunol* 21:2717–2725. doi: 10.1002/eji.1830211111. [PubMed: 1834468]
- Sherry N, Hagopian W, Ludvigsson J, Jain SM, Wahlen J, Ferry RJ, Bode B, Aronoff S, Holland C, Carlin D, King KL, Wilder RL, Pillemer S, Bonvini E, Johnson S, Stein KE, Koenig S, Herold KC, Daifotis AG, and Protégé Trial Investigators. 2011 Teplizumab for treatment of type 1 diabetes (Protégé study): 1-year results from a randomised, placebo-controlled trial. *Lancet*. 378:487–497. doi:10.1016/S0140-6736(11)60931-8. [PubMed: 21719095]
- Vossen ACTM, Tibbe GJM, Kroos MJ, van de Winkel JGJ, Benner R, and Savelkoul HFJ. 1995 Fc receptor binding of anti-CD3 monoclonal antibodies is not essential for immunosuppression, but triggers cytokine-related side effects. *Eur. J. Immunol* 25:1492–1496. doi:10.1002/eji.1830250603. [PubMed: 7614975]
- Van Wauwe JP, De Mey JR, and Goossens JG. 1980 OKT3: a monoclonal anti-human T lymphocyte antibody with potent mitogenic properties. *J. Immunol* 124:2708–13. [PubMed: 6966296]
- Winkler C, Haupt F, Heigermoser M, Zapardiel-Gonzalo J, Ohli J, Faure T, Kalideri E, Hommel A, Delivani P, Berner R, Kordonouri O, Roloff F, von dem Berge T, Lange K, Oltarzewski M, Glab R, Szypowska A, Snape MD, Vatish M, Todd JA, Larsson HE, Ramelius A, Kördel JÅ, Casteels K, Paulus J, Ziegler AG, and Bonifacio E. 2019 Identification of infants with increased type 1 diabetes genetic risk for enrollment into Primary Prevention Trials—GPPAD-02 study design and first results. *Pediatr. Diabetes* doi:10.1111/pedi.12870.
- Woodle ES, Thistlethwaite JR, Jolliffe LK, Zivin RA, Collins A, Adair JR, Bodmer M, Athwal D, Alegre ML, and Bluestone JA. 1992 Humanized OKT3 antibodies: successful transfer of immune modulating properties and idiotype expression. *J. Immunol* 148:2756–63. [PubMed: 1533410]
- Woodle ES, Xu D, Zivin RA, Auger J, Charette J, O’Laughlin R, Peace D, Jolliffe LK, Haverty T, Bluestone JA, and Thistlethwaite JR. 1999 PHASE I TRIAL OF A HUMANIZED, Fc RECEPTOR NONBINDING OKT3 ANTIBODY, huOKT3??1 (Ala-Ala) IN THE TREATMENT OF ACUTE RENAL ALLOGRAFT REJECTION. *Transplantation*. 68:608–616. doi: 10.1097/00007890-199909150-00003. [PubMed: 10507477]
- Xu D, Alegre M-L, Varga SS, Rothermel AL, Collins AM, Pulito VL, Hanna LS, Dolan KP, Parren PWI, Bluestone JA, Jolliffe LK, and Zivin RA. 2000 In Vitro Characterization of Five

Humanized OKT3 Effector Function Variant Antibodies. *Cell. Immunol* 200;16–26. doi:10.1006/cimm.2000.1617. [PubMed: 10716879]

Ziegler AG, Rewers M, Simell O, Simell T, Lempainen J, Steck A, Winkler C, Ilonen J, Veijola R, Knip M, Bonifacio E, and Eisenbarth GS. 2013 Seroconversion to Multiple Islet Autoantibodies and Risk of Progression to Diabetes in Children. *JAMA*. 309:2473. doi:10.1001/jama.2013.6285. [PubMed: 23780460]

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript