Supplementary Appendix

Supplement to: Reichman TW, Markmann JF, Odorico J, et al. Stem cell–derived, fully differentiated islets for type 1 diabetes. N Engl J Med. DOI: 10.1056/NEJMoa2506549

This appendix has been provided by the authors to give readers additional information about the work.

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Supplementary Methods

Clinical Development

The trial was conducted in accordance with the Declaration of Helsinki, local applicable laws and regulations, and current Good Clinical Practice Guidelines as described by the International Council for Harmonization.

Inclusion Criteria

1. Participant will sign and date an informed consent form (ICF).

2. Willing and able to comply with scheduled visits, treatment plan, trial restrictions, laboratory tests, contraceptive guidelines, and other trial procedures, and able to understand the risks and the requirements for participation in the trial.

   This includes:

   - absence of clinically significant cognitive impairment assessed by the investigator using a local site-standard cognitive screening tool, and

   - willingness and ability to travel to the clinical trial site or local transplant center, if needed for evaluation or treatment.

3. Participants (male and female) between the ages of 18 and 65 years (inclusive) on the date of informed consent.

4. Body mass index of 18 to 30 kg/m^2, and minimum body weight ≥45 kg, at Screening.

5. Compatible blood group (A or AB)

6. Clinical history and laboratory evidence of Type 1 diabetes (T1D) with:

    - onset of T1D at <40 years of age;
    - insulin dependence for ≥5 years at time of Screening; and
    - peak stimulated C-peptide level <50 pmol/L during mixed-meal tolerance testing (MMTT), and
    - prior or current presence of at least one T1D autoantibody, with appropriate documentation; in the absence of at least one T1D autoantibody, the diagnosis of T1D must be confirmed by an independent endocrinologist.

      Note: Historical T1D autoantibodies may include insulin autoantibody if drawn prior to or within 10 days of having started exogenous insulin. Current T1D autoantibodies (i.e., antibodies against glutamic acid decarboxylase 65 [GAD65], islet antigen 2 [IA2], and zinc transporter 8 [ZnT8]) will not include antibodies against insulin for the purpose of trial inclusion.

7. Consistent use of continuous glucose monitor (CGM) for at least 3 months before Screening and willingness to use only the trial-provided CGM for the duration of the trial.

8. History of intensive diabetes management, requiring multiple daily insulin injections or insulin pump use, under the direction of an endocrinologist, diabetologist, or diabetes specialist with ≥3 documented clinical evaluations during the 12 months before signing of informed consent at Screening and no change in insulin delivery method (continuous subcutaneous insulin infusion [CSII] versus multiple daily injections [MDI]) for at least 3 months before Screening.

9. At least 2 episodes of severe hypoglycemia (confirmed by independent adjudication for participants in Parts B and C) in the 12 months before signing of informed consent at Screening. An episode of severe hypoglycemia is defined as an event with one of the following symptoms: cognitive impairment (memory loss or confusion); changes in behavior (uncontrollable or irrational behavior); unusual difficulty in awakening; suspected seizure (convulsions)/seizure; loss of consciousness; or visual symptoms, in which the participant was unable to treat him/herself and which was associated with either a blood glucose level <54 mg/dL (3.0 mmol/L) or prompt recovery after oral carbohydrate, intravenous (IV) glucose, or glucagon administration.

   Note: For Parts B and C, all severe hypoglycemic events (SHEs) that occur within the 12 months before signing of the Screening ICF will be adjudicated by independent adjudication and used to assess eligibility.

10. Metabolically unstable, as defined by the presence of at least one of the following conditions:

    - Reduced awareness of hypoglycemia at Screening as defined by a Clarke score of 4 or more;
    - Clinically significant hypoglycemia defined at Screening by a composite hypoglycemic (HYPO) score $\geq 90^{th}$ percentile ($\geq$1,047);
    - Marked glycemic lability characterized by wide swings in blood glucose and defined at Screening by a liability index (LI) score $\geq 90^{th}$ percentile ($\geq$433 $mmol/L^2/h \cdot week^{-1}$)

11. Up to date on routine cancer screening per US Preventive Service Task Force guidelines for screening before solid organ transplantation with no disqualifying findings.

Exclusion Criteria

1. Prior islet cell transplant, organ transplant, or cell therapy

2. Advanced complications associated with diabetes including untreated proliferative retinopathy, skin ulcers, or amputations attributable to diabetes.

3. Participants who have any 1 of the following criteria:

    - Insulin requirements: >0.8 U/(kg*day), >55 U/day, or <10 U/day;

    - Glycosylated hemoglobin (HbA1c): <7.0% or >9.5%

4. Clinically significant active infection or chronic infection such as hepatitis B, hepatitis C, human immunodeficiency virus (HIV), and/or tuberculosis (TB); or invasive aspergillus, histoplasmosis, or coccidioidomycosis infection within one year prior to signing of informed consent at Screening.

5. Negative screen for Epstein-Barr virus (EBV) by immunoglobulin G (IgG) determination

6. Positive for coronavirus disease (COVID)-19 by polymerase chain reaction (PCR) or evidence of active infection per local institutional standards

7. Panel-reactive antibodies (PRA) to human leukocyte antigen (HLA) > 0% or positive donor-specific antibody (DSA)

8. Blood transfusion within 9 months before Screening or known hemoglobinopathy, chronic anemia, or any condition known to interfere with the measurement of HbA1c

9. Administration of live attenuated vaccine(s) within 2 months before infusion.

10. Hematological and biochemical indices within the following ranges:

- Hemoglobin (Hb) below the lower limit of normal (LLN):
  - Males: 13.2 g/dL
  - Females: 11.5 g/dL
- Neutropenia (<1,500/µL) or thrombocytopenia (platelets <100,000/ µL)
- Lymphocytes <1,000/µL
- Estimated glomerular filtration rate (eGFR) of <80 mL/min/1.73m^2 calculated using the participant's measured serum creatinine and the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equation
- Aspartate transaminase (AST), alanine transaminase (ALT), alkaline phosphatase or total bilirubin >1.5 × the upper limit of normal (ULN)
- Untreated or inadequately treated hyperlipidemia at Screening: fasting low-density lipoprotein (LDL) cholesterol >130 mg/dL (3.4 mmol/L) and/or fasting triglycerides >200 mg/dL (2.3 mmol/L)

  Note: If deemed appropriate by the investigator, participants meeting this exclusion criterion may have their medication adjusted (either started or modified) during the Screening Period, and may be re-evaluated after ≥2 weeks on a stable medication

regimen. Re-evaluation of this criterion must be performed at least 2 weeks before the Pre-infusion Safety Assessment Visit.

- Presence of macroalbuminuria >300 mg/g creatinine on a spot urine test (first morning sample preferred) or 24-hour urine collection.

  Note: If deemed appropriate by the investigator, participants with macroalbuminuria (albumin/creatinine ratio of >300 mg/g) may have relevant medication(s) started, discontinued, or adjusted during the Screening Period, and may be re-evaluated for eligibility after ≥2 weeks on a stable medication regimen. Re-evaluation of this eligibility criterion must be performed at least 2 weeks before Pre-infusion Safety Assessment Visit.

11. 5-hydroxyindoleacetic acid (5-HIAA) measured using 24-hour urine collection >ULN and/or serum fasting somatostatin level >ULN.

    Note: A 1-time repeat of the 5-HIAA and somatostatin assessment may be allowed, if in the opinion of the investigator, the initial value is confounded by non-adherence to dietary restrictions, inappropriate sample collection, or other confounding factors or is not consistent with the participant's current medical condition. If the repeat value still meets the exclusion criterion, then the participant is excluded from continuing in the trial.

12. Participant's with poorly controlled hypertension as defined by systolic blood pressure >140 mmHg or diastolic blood pressure >90 mmHg.

    Note: If deemed appropriate by the investigator, participants meeting this exclusion criterion may have their blood pressure medication adjusted (either started or modified) during the Pre-

screening or Screening Period, and may be re-evaluated after ≥2 weeks on a stable antihypertensive medication regimen. Re-evaluation of this criterion must be performed at least 2 weeks before the Pre-infusion Safety Assessment Visit.

13. Participants with any history of cardiac disease, as well as any clinically significant abnormality identified on cardiac stress test, angiogram evaluation, or echocardiogram performed at Screening.

14. Participants with liver disease, portal hypertension, any coagulopathy (including history of Factor V deficiency) or long-term anti-coagulant therapy (except low-dose aspirin). Other hepatic conditions including hepatic anatomic abnormalities or variants that would place the individual at increased risk in the judgment of the investigator are also considered exclusionary.

15. Symptomatic cholecystolithiasis; acute or chronic pancreatitis; or current symptomatic peptic ulcer disease.

16. Any condition possibly affecting drug absorption (e.g., severe diarrhea, vomiting, or gastrectomy, etc.)

17. Any medical condition requiring chronic use of systemic steroids greater than 5 mg of prednisone or equivalent per day; treatment with anti-hyperglycemic agents **(other than insulin)** or treatment with any immunosuppressive regimen during the 30-day period before Screening.

18. Unable to reliably take daily medications

19. History of any illness or any clinical condition that, in the opinion of the investigator, might confound the results of the trial or pose an additional risk in administering study drug to the participant. This may include, but is not limited to, history of relevant drug or food allergies; bronchoconstrictive or bronchospastic disease; adrenal insufficiency; uncontrolled thyroid disease; history or presence of clinically significant pathology; and history of cancer, except for squamous cell skin cancer, basal cell skin cancer, and Stage 0 cervical carcinoma in situ (all 3 with no recurrence for 5 years before enrollment).

20. Known active alcohol or substance abuse; or tobacco use during the 6-month period before Screening.

21. Known allergy or hypersensitivity to milk, milk products, soy products, adenosine, or dextran, or to any study drug administered in the protocol (e.g., heparin, sirolimus, etanercept).

22. Prior serious or severe adverse reactions to any study drug administered in the protocol that in the opinion of the investigator, would place the participant at risk with repeat exposure (e.g., heparin, sirolimus, etanercept).

23. Use of the substances, activities, or devices as indicated in the protocol, during the specified times.

24. For female participants: Pregnant, nursing, or planning to become pregnant during the trial or within 90 days after discontinuation of immunosuppression therapy or unwillingness to comply with contraceptive requirements.

For male participants: Male participants with a female partner who is planning to become pregnant with the male participant during the trial or within 90 days after discontinuation of immunosuppression therapy or unwillingness to comply with contraceptive requirements.

25. Participants with 5 or more adjudicated episodes of severe hypoglycemia in the 12 months before signing of informed consent at Screening or 2 or more adjudicated episodes of severe hypoglycemia in the time period from signing of informed consent at Screening to zimislecel infusion.

26. Prior participation in the active dosing phase of a clinical trial using an investigational product within 30 days of signing of informed consent. If a participant has received any prior treatment with an investigational therapy, the specific product, duration of use, and last use will be documented.

27. Participant, or close relative of the participant, is the investigator or a sub-investigator, research assistant, pharmacist, study coordinator, or other staff directly involved with the conduct of the trial at that site.

28. Karnofsky performance status of <80%.

Informed Consent Process

All potentially interested participants underwent the informed consent process before participating in the trial. Informed consent forms were reviewed and approved by institutional review boards (IRB), ethics committees (EC), or research ethics boards (REB). Investigators obtained consent through discussion with potentially interested participants by disclosing all the critical information about the trial (e.g., purpose, procedures, potential risks and benefits of participation, alternatives to participation and right to not participate or to withdraw from the trial at any time) and addressing questions or concerns the participant had about the trial before the start of screening activities. In these discussions, investigators ensured that the potentially interested participant had the ability to understand the information provided and to make a reasoned, voluntary, decision about participation in the trial. All participants consented to participate in the trial, which is documented on the trial specific informed consent forms.

Immunosuppression and Zimislecel dosing

Induction

Rabbit anti-thymocyte globulin (Thymoglobulin®; ATG) was used for the first infusion of zimislecel. The total dose of ATG was 6 mg/kg given as an IV infusion starting on Day -2 through Day 3. The first dose was 0.5 mg/kg infused over 6 to 12 hours, the second dose was 1.0 mg/kg infused over ≥6 hours, and the third, fourth, and fifth doses were 1.5 mg/kg infused over ≥6 hours. The first 3 doses were administered before the zimislecel infusion procedure, which occurred approximately 12 hours after the completion of the third dose. The fourth and fifth doses of ATG were administered on Day 2 and Day 3, respectively. No ATG was administered on Day 1.

The following pre-medications were administered before infusion of ATG:

1. Acetaminophen: 650 mg PO/PR (taken by mouth/taken by rectum), 30 minutes before and 650 mid-way through ATG infusion
2. Diphenhydramine: 50 mg PO, 30 minutes before and 50 mg midway through ATG infusion
3. Methylprednisolone: 1 mg/kg IV, 1 hour before and as needed during first ATG infusion only on Day -24.
4. Pentoxyfilline: 400 mg PO 3 times a day (tid) 1 hour before the first ATG infusion on Day -2 and continued through Day 8.

Etanercept was administered peri-transplant 50 mg IV on Day 1 (approximately 1 hour before zimislecel infusion), then 25 mg subcutaneously on Days 4, 8, and 11. If a site was unable to administer etanercept IV due to local guidelines, then subcutaneous dosing may have been used for all time points as described in the study reference manual.

Zimislecel Production and Infusion

Human pluripotent stem cells were differentiated into stem cell-derived islets with growth factors and small molecules using an advanced protocol evolved from the work previously reported by Pagliuca[1] and Veres.[2]

Zimislecel is composed of allogeneic human embryonic stem cell (hESC)-derived, fully differentiated, islets (stem-cell derived islets). For the clinical manufacturing process of zimislecel, hESC line SEM-01 was thawed, expanded, adapted to 3D cultures, and fully differentiated into stem-cell derived islets. Generation of zimislecel follows a multistage differentiation protocol using key small molecules and growth factors as follows: Stage 1, DE definitive endoderm (CHIR, Activin A), Stage 2 primitive gut tube (KGF), Stage 3 pancreatic progenitor PP1 (KGF, PDBU,

Sant-1, Retinoic Acid, Activin A, Thiazovinin, DMH-1), Stage 4 pancreatic progenitor PP2 (KGF, Sant-1, Thiazovinin, Activin A, Retinoic Acid, PDBU, XXI, AS1842856), Stage 5 pancreatic endocrine cells (NVP-TNKS656, PDBU, Sant-1, Betacellulin, XXI, Alk5i, GC-1, LDN-193189, Thiazovinin, SSP, DZNEP, Retinoic Acid). The final product consists of insulin-producing β-cells, glucagon-expressing α-cells, somatostatin-expressing δ-cells, and cells expressing genes similar to enterochromaffin cells. The proportion of β- and non-β islet cell types for all released drug product lots were within the published range of naturally occurring islets.[3]

Stem-cell derived islets were formulated (resuspended in a shipping media) and filled into an infusion bag (zimislecel) for shipment to the clinic as a cold (2°C to 8°C) shipping solution. The product was characterized for cell dose, composition, viability, purity, and potency, and must meet regulatory (e.g., FDA) release criteria as defined in the product specifications before shipment to the site and formal product release. The product was tested for endotoxin and mycoplasma, and a gram stain is performed for sterility assessment that is confirmed to be negative prior to release for infusion. Upon arrival at the clinical site, the product was examined for container integrity, maintenance of temperature, and other labeling requirements. Zimislecel was administered as a single infusion.

All participants received an induction and maintenance immunosuppressive regimen before and after zimislecel infusion. Two days before zimislecel infusion, participants began a standard steroid free immunosuppressive regimen consistent with the islet transplantation protocol established by the Clinical Islet Transplantation Consortium.[4]

Peri-infusion of Zimislecel

Sirolimus was started at a dose of 0.05 to 0.2 mg/kg PO on Day -2, followed by 0.05 to 0.1 mg/kg once each morning. The sirolimus dose was adjusted as tolerated to whole blood 24-hour trough target of 10 to 15 ng/mL for the first 3 months and then 8 to 12 ng/mL thereafter.

Tacrolimus was started at an oral dose of 0.015 mg/kg PO every 12 hours on Day 2, targeting a whole blood 12-hour trough of 3 to 6 ng/mL.

Mycophenolate mofetil (500 to 1000 mg PO twice per day [bid]) or mycophenolate sodium (360 to 720 mg orally PO bid) may have been used as a replacement for either tacrolimus or sirolimus if not tolerated.

For participants who discontinued sirolimus and converted to mycophenolate mofetil or mycophenolate sodium, tacrolimus was administered to target whole blood trough levels of 10 to 12 ng/mL for the first 3 months after zimislecel infusion, 8 to 10 ng/mL from 3 to 6 months after zimislecel infusion, and 6 to 8 ng/mL thereafter.

Zimislecel Infusion

Zimislecel was delivered via gravity drainage infusion through a catheter in the portal vein over an approximately 30-to-60-minutes time-period.

Access to the portal vein and islet infusion followed the procedures established for deceased donor islet transplantation. An interventional radiologist or surgeon obtained access to the hepatic portal vein (site experience and expertise determined the approach). In the radiologically guided procedure, the portal vein was accessed percutaneously through a transhepatic approach using a needle and guidewire and a catheter was placed in the portal vein for infusion. In the surgical

approach a mini-laparotomy was performed to directly visualize a mesenteric vein and place a catheter by venotomy.

At a minimum, portal vein pressure was measured before infusion of the first bag of the islet product, following infusion of the first half of the total planned dose for the procedure, and after delivery of the total dose is completed. This may be performed under anaesthesia with direct surgical visualization or with conscious sedation and percutaneous administration with ultrasound guidance.

Participants received IV insulin before infusion through ≥24 hours after zimislecel infusion to achieve target glucose levels of 81 to 115 mg/dL (4.5 to 6.4 mmol/L). Once transitioned to subcutaneous insulin, target glucose levels were 100 to 140 mg/dL (5.6 to 7.8 mmol/L) fasting and 100 to 180 mg/dL (5.6 to 10.0 mmol/L) post-prandial for approximately six weeks after zimislecel infusion. Insulin withdrawal was at the discretion of investigators who were asked to regularly assess insulin requirements and adjust exogenous insulin dosing to maintain target glucose levels.

Management of Participants

Participants were clinically managed by the trial investigators, which included transplant surgeons, endocrinologists, and additional transplant specialists if needed. Insulin dose adjustment was guided by protocol-specified fasting and prandial glucose targets (see above). Trial investigators monitored glucose levels throughout the trial by means of trial-provided CGM and glucometers. Insulin dosing was decreased and, ultimately, discontinued in participants who were able to meet protocol specified target glucose levels with less or no exogenous insulin. Participants who discontinued exogenous insulin continued to use the study-provided CGM for monitoring of glycemic control throughout the trial.

Definition of Severe Hypoglycemic Event and Insulin Independence

Definition of Severe Hypoglycemic Event

An episode of severe hypoglycemia is defined as an event with at least 1 of the following symptoms: cognitive impairment (memory loss or confusion); changes in behavior (uncontrollable or irrational behavior); unusual difficulty in awakening; suspected seizure (convulsions)/ seizure; loss of consciousness; or visual symptoms, in which the participant was unable to treat him/herself and which was associated with either a blood glucose level <54 mg/dL (3.0 mmol/L) or prompt recovery after oral carbohydrate, IV glucose, or glucagon administration. Each severe hypoglycemic event in the one year before screening and after enrollment is reviewed and adjudicated by an independence endpoint adjudication committee according to these criteria.

Definition of Insulin Independence in the Phase 1/2 Trial

Participants were considered to be insulin-independent if they met all the following five criteria at a visit where an MMTT assessment was conducted: able to titrate off insulin therapy for at least 1 week; HbA1c ≤ 7%; and a post prandial serum glucose ≤180 mg/dL (10.0 mmol/L) at 90 minutes; fasting serum glucose level ≤126 mg/dL (7.0 mmol/L) and at least one fasting or stimulated C-peptide ≥166 pmol/L during the MMTT.

Safety and Efficacy Assessments

All participants were monitored for safety and tolerability through adverse event reporting, clinical laboratory and vital signs assessments, physical examinations, cardiac monitoring (echocardiogram and electrocardiogram), and liver imaging.

Efficacy was assessed using serum C-peptide and glucose, HbA1c, CGM, and exogenous insulin dose requirements.

Measurements of C-Peptide and Mixed-Meal Tolerance Test (MMTT)

Standardized four-hour MMTTs were conducted at Day 90, 180, 270, 365 and every six months thereafter to measure fasting and stimulated serum C-peptide and glucose concentrations.

C-peptide is co-secreted with insulin and is used as a marker of β-cell function. Evidence of islet cell function is defined as peak C-peptide ≥100 pmol per liter during MMTT. Conversely, insufficient islet cell function is defined as peak C-peptide <100 pmol per liter during MMTT.

Basal (fasting) C-peptide was measured to assess islet cell function. Basal (fasting) and stimulated glucose and C-peptide levels was determined during 4-hour MMTTs. Each MMTT was performed after an overnight fast. Participants were instructed not to inject or deliver additional rapid-acting (bolus) insulin within 4 hours of the start of test. Evening or bedtime administration of long-acting insulin was permitted, as was consumption of water. Participants receiving CSII (insulin "pump" therapy) could have continued their normal basal rate of insulin but could not receive a bolus for 4 hours before start of the test. Participants who used a closed loop insulin delivery system were required to change to manual mode prior to and for the duration of the MMTT.

Each blood sample collected for C-peptide and glucose determination was drawn and shipped frozen for laboratory evaluation.

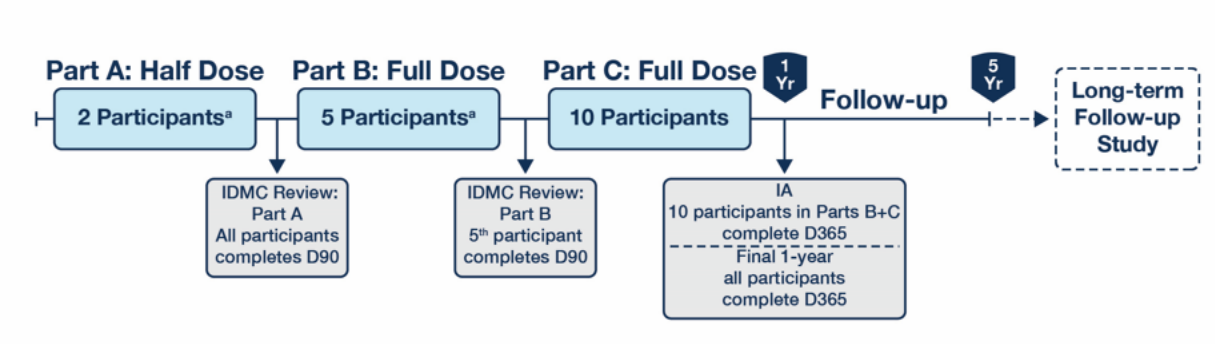
Results

Evaluation of Half Dose of Zimislecel (Part A)

Two participants received half the target dose of zimislecel. Following the infusion, both participants in Part A had detectable fasting and stimulated levels of C-peptide on MMTT on Day 90 along with reductions in the extent of glucose excursions, indicating engraftment of zimislecel islet cells and evidence of meaningful glucose-responsive endogenous insulin production. The first participant was free from exogenous insulin by Day 210, and the participant maintained an HbA1c of <5.5% (**Figure S3**) and >90% time-in-range thereafter through 24 months of follow-up (**Figure S4**). The second participant had improvement in HbA1c from 7.5% to 6.7% at Day 90, with a concurrent reduction in insulin use by ~30% (**Figure S3**). The second participant received a second half dose, per protocol, approximately 9 months after the first infusion of zimislecel but withdrew consent, not related to an adverse event, approximately 3 months after the second infusion of zimislecel.

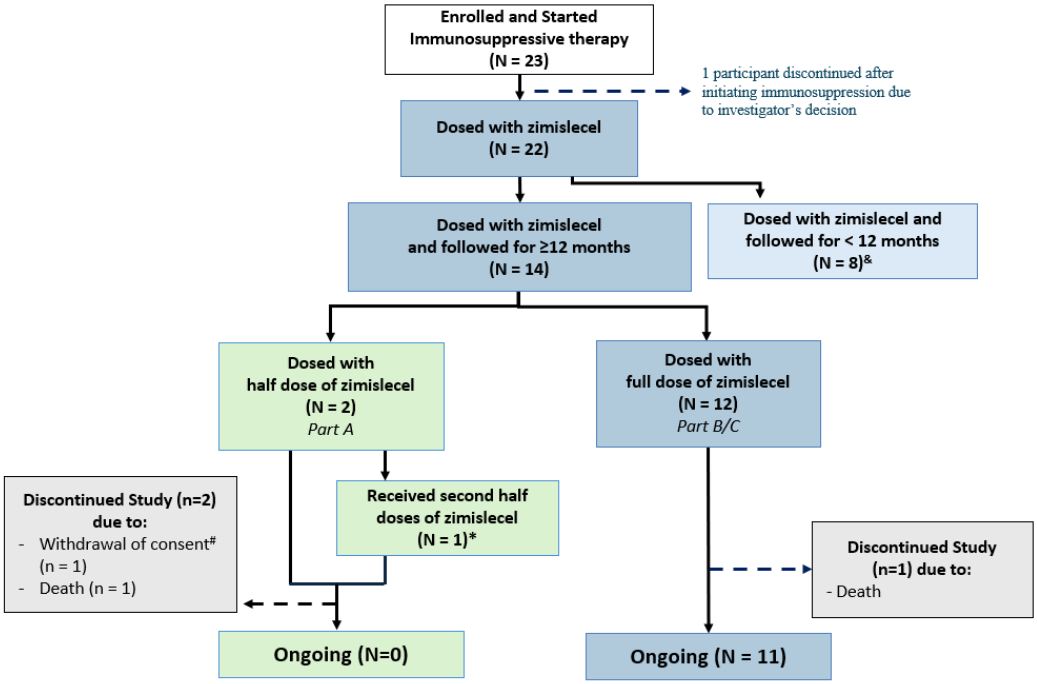
Supplementary Figure

Figure S1. Study Design



Note: Enrollment and dosing of the phase 1/2 portion of the trial is complete
D: day; IA: interim analysis; IDMC: independent data monitoring committee
[a] Participants in Parts A and B were dosed in a staggered manner. Part A and B enrolled and dosed 2 and 5 participants, respectively. One participant enrolled and received zimislecel as two half-doses, per protocol, and is thus counted in both Parts A and B (once for receiving a half dose in Part A and once for receiving a full dose in Part B).

Figure S2. CONSORT Diagram (as of October 2024)



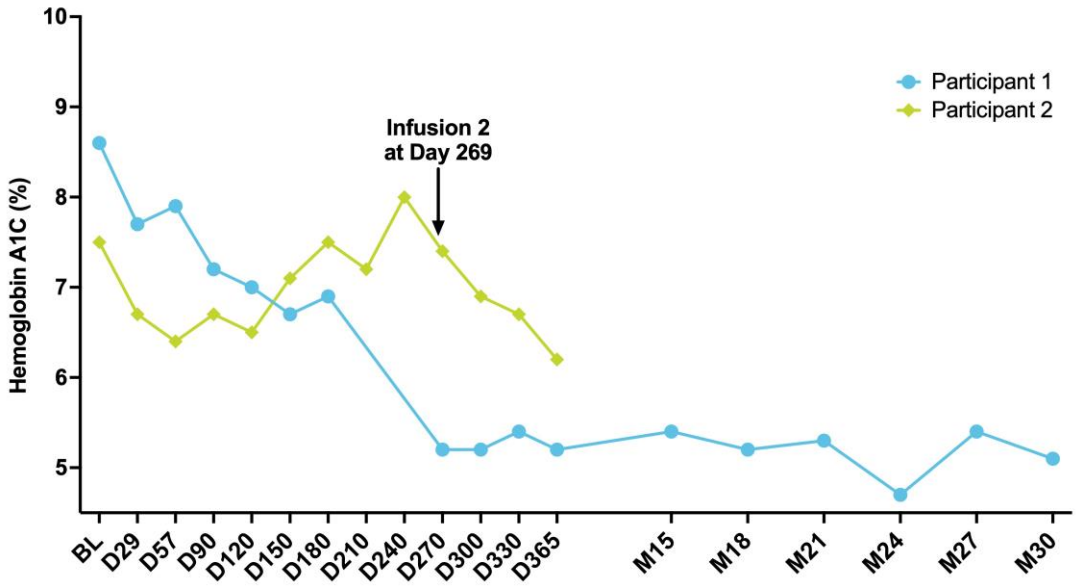
* One participant enrolled and received a full-dose of zimislecel as two infusions and, per protocol, is included in Part A for receiving a half dose and in Part B for receiving a full dose.
& Subjects followed for less than 12 months had completed between 10 days and approximately 4 months of follow-up
# not due to an AE

Figure S3. HbA1c (Panel A) and Total Daily Insulin Use (Panel B) Following Zimislecel Over Time

Data shown is for the 2 participants in Part A: one who received a half dose of zimislecel and the second one who received a half dose of zimislecel and then a second half dose nine months after the first dose. Panel A shows HbA1c over time. Each solid line shows the HbA1c for an individual participant over time. Panel B shows total daily insulin use over time. Each solid line shows total daily insulin use for an individual participant over time.

ADA: American Diabetes Association; BL: baseline; D: Day; EASD: European Association for the Study of Diabetes; T1D: Type 1 diabetes.

A. HbA1c Following Zimislecel Over Time



B. Total Daily Insulin Use Following Zimislecel Over Time

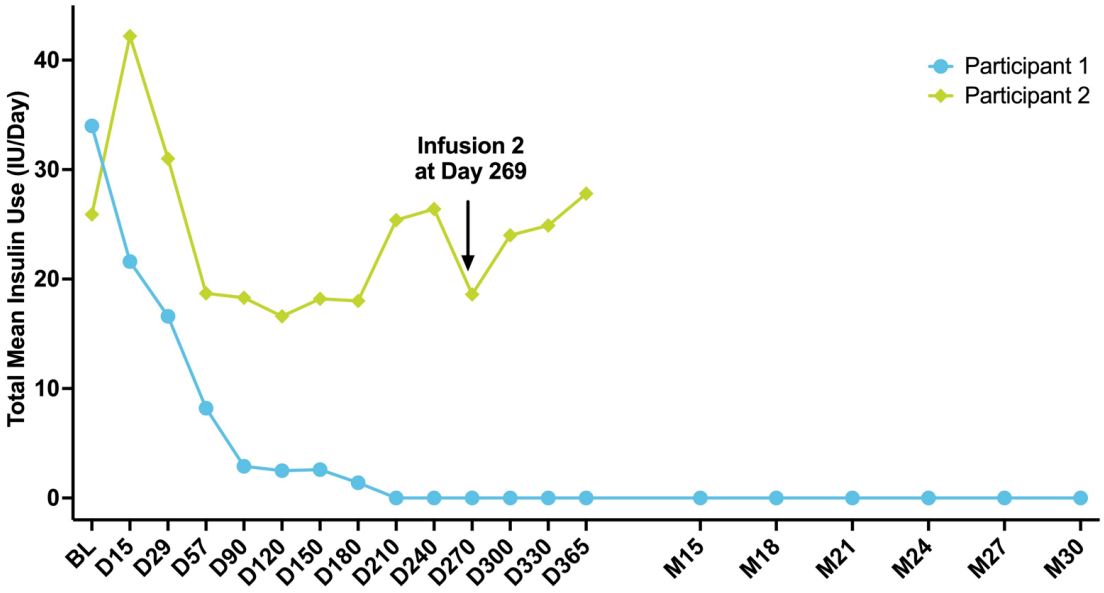
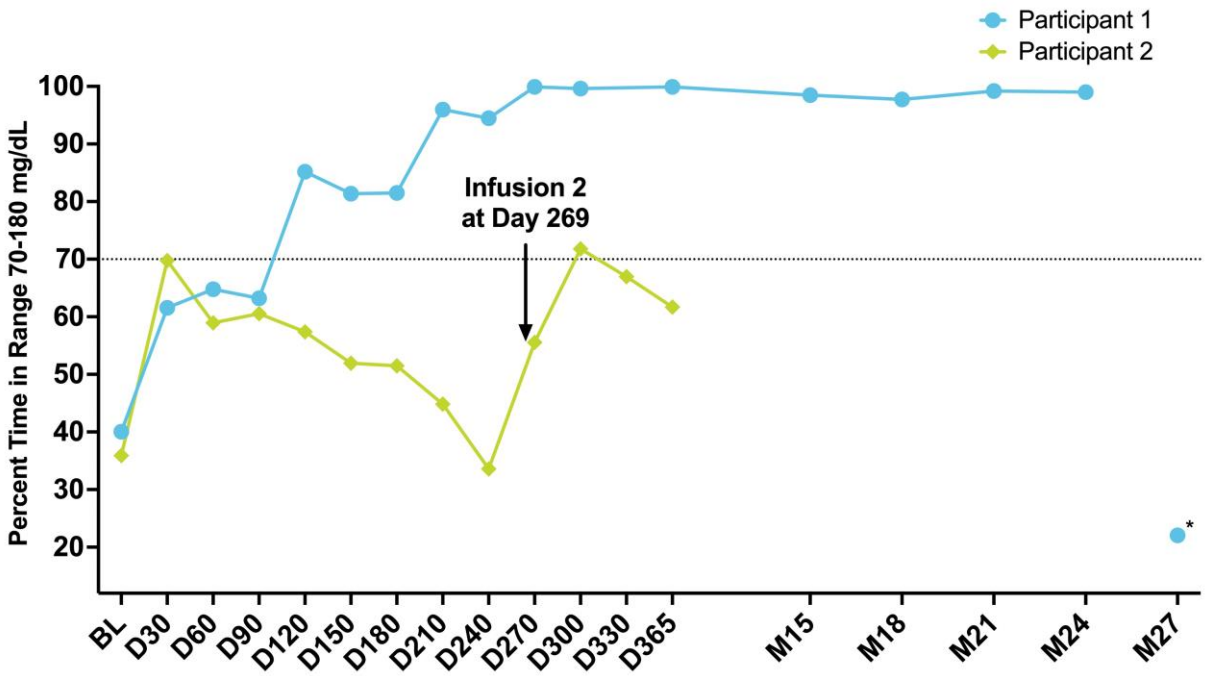


Figure S4. Percent Time Spent in the Recommended Target Glucose Range Following Zimislecel

Data shown is for the 2 participants in Part A: one who received a half dose of zimislecel and the second one who received a half dose of zimislecel and then a second half dose nine months after the first dose. Figure shows the percent time spent in the ADA/EASD target range over time. Each solid line shows the percent glucose time in range for an individual participant over time.

*At the M27 assessment, the time in range for Participant 1 was temporarily impacted by intercurrent COVID-19 infection and subsequent recovered.

ADA: American Diabetes Association; D: Day; EASD: European Association for the Study of Diabetes; M: Month



Supplementary Tables

Table S1. Demographics and Clinical Characteristics at Baseline for All Participants Dosed with Zimislecel with ≥12 Months of Follow-up

Characteristic	Total N=14
Age at screening (years), mean (min, max)	43.6 (24, 64)
Sex (Female/Male), n/n	5/9
Race, n (%)	
White	14 (100)
Weight (kg), mean (min, max)	73.3 (56.0, 97.5)
Body-mass index (kg/m^2), mean (min, max)	24.5 (21.3, 28.5)
Duration of diabetes (years), mean (min, max)	22.8 (7.8, 47.4)
HbA1c (%), mean (min, max)	7.8 (7.1, 9.9)
Glucose time-in-range (%), mean (min, max)	47.8 (19.0, 66.2)
Fasting C-peptide (pmol/L), mean (min, max)	Undetectable
Total daily insulin (unit/day), mean (min, max)	39.3 (19.8, 52.0)
Insulin requirements (unit/kg/day) , mean (min, max)	0.54 (0.35, 0.64)
Severe hypoglycemia events (SHEs) in year prior to screening, mean (min, max)	2.7 (2, 4)
Use of insulin pump at pre-screening or screening, n (%)	9 (64.3)
Use of Hybrid Closed Loop Systems at pre-screening or screening, n (%)	6 (42.9)

Data shown are for 14 participants who received zimislecel with ≥12 months of follow-up (2 participants in Part A, 4 participants in Part B, and 8 participants in Part C). Participants in Part A received half the dose of zimislecel (one participant in Part A received a second half dose after the first dose) and participants in Parts B and C received full dose of zimislecel in a single infusion.

Table S2. Representativeness of Population of Participants

Category Special considerations related to:	Type 1 Diabetes
Sex and gender	Men and women are equally affected by T1D, however in populations with high incidence of T1D there is excess in males in both childhood and adult onset T1D.[5-8]
Age	Globally more than 80% of diagnosed T1D patients today are over age 20, and approximately 64% of all people diagnosed with T1D are 20 to 59 years of age.[9, 10]
Race or ethnic group	It is unknown the exact impact race/ethnicity may have in the development of T1D; however, significant variability exists in the incidence and prevalence of T1D globally. Incidence rates for T1D are generally seen to be the highest in Nordic countries and lowest in Asian countries.[5, 9, 10]
Overall representativeness of this trial	Participants were 24 to 64 years of age and both male (n=9) and female (n=5), and all participants were White. Overall, the participants of the study are representative of T1D patients.

Table S3. Summary of Peak C-peptide (pmol/L) from MMTT

Visit Statistics	Zimislecel N = 12
Baseline	
N	12
Mean (SD)	8 (3)
Median	7
Min, max	7, 13
Day 90	
N	12
Mean (SD)	471 (165)
Median	424
Min, max	262, 762
Day 180	
N	12
Mean (SD)	1163 (372)
Median	1235
Min, max	636, 1980
Day 270	
N	12
Mean (SD)	1290 (400)
Median	1223
Min, max	626, 1947
Day 365	
N	12
Mean (SD)	1525 (415)
Median	1576
Min, max	775, 2126

Data are presented for 12 participants who received the full dose of zimislecel in a single infusion with ≥12 months of follow-up (4 participants in Part B, and 8 participants in Part C).

Table S4. Improvement in Percent Glucose Time-in-range Per Day

Glycemic Outcomes Based on Continuous Glucose Monitoring					
Outcome	**Baseline N = 12**	**Day 90 N = 12**	**Day 180 N = 12**	**Day 270 N = 12**	**Day 365 N = 11**
Percent of time spent in specified glucose range, mean (SE)					
Above range, >250 mg/dL	18.08 (3.47)	2.32 (0.58)	0.47 (0.20)	0.28 (0.10)	0.32 (0.18)
Above range, 181-250 mg/dL	30.12 (1.41)	20.68 (2.34)	10.93 (1.74)	6.52 (1.64)	6.03 (1.53)
In range, 70-180 mg/dL	49.46 (3.43)	76.10 (2.61)	88.34 (1.80)	92.69 (1.66)	93.32 (1.52)
Below range, 54-69 mg/dL	1.66 (0.49)	0.80 (0.29)	0.24 (0.10)	0.43 (0.15)	0.30 (0.10)
Below range, <54 mg/dL	0.68 (0.43)	0.10 (0.03)	0.03 (0.02)	0.08 (0.03)	0.04 (0.02)
Average Glucose, mg/dL, mean(SE)	**186.0 (7.3)**	**151.1 (3.6)**	**141.4 (3.4)**	**131.2 (3.6)**	**134.4 (3.5)**
Glycemic variability (CV%) , mean(SE)	**36.3 (1.4)**	**27.6 (1.0)**	**21.2 (0.9)**	**21.1 (0.6)**	**20.0 (1.0)**

Data are presented for 12 participants who received the full dose of zimislecel in a single infusion with ≥12 months of follow-up (4 participants in Part B, and 8 participants in Part C).

Table S5. Summary of Adverse Events for All Participants Dosed with Zimislecel with ≥12 months of Follow-up

Category	Participants With AEs N=14 n (%)
Any adverse event	14 (100)
Adverse events by maximum severity	
Grade 1/Mild	1 (7.1)
Grade 2/Moderate	6 (42.9)
Grade 3/Severe	5 (35.7)
Grade 4/Life threatening	0
Grade 5/Death	2 (14.3)
Adverse events related to zimislecel	**7 (50.0)**
Adverse events related to surgical procedure	**8 (57.1)**
Adverse events related to immunosuppression	**14 (100.0)**
Adverse events related to ancillary drug	**13 (92.9)**
Discontinuation of trial due to adverse event	**0**

Data shown are for 14 participants who received zimislecel with ≥12 months of follow-up (2 participants in Part A, 4 participants in Part B, and 8 participants in Part C). Participants in Part A received half the dose of zimislecel (one participant in Part A received a second half dose after the first dose) and participants in Parts B and C received full dose of zimislecel in a single infusion.

Table S6. Summary of Serious Adverse Events by Preferred Term for All Participants Dosed with Zimislecel with ≥12 months of Follow-up

Preferred Term	Participants With AEs N=14 n (%)
Any serious AEs	**7 (50.0)**
Neutropenia	3 (21.4)
Acute kidney injury	2 (14.3)
COVID-19	1 (7.1)
Cellulitis	1 (7.1)
Confusional state	1 (7.1)
Dehydration	1 (7.1)
Dementia	1 (7.1)
Electrolyte imbalance	1 (7.1)
Headache	1 (7.1)
Hypokalaemia	1 (7.1)
Malnutrition	1 (7.1)
Meningitis cryptococcal	1 (7.1)
Mental status changes	1 (7.1)
Migraine	1 (7.1)
Rash	1 (7.1)
Small intestinal obstruction	1 (7.1)
Tenosynovitis	1 (7.1)
Urinary tract infection	1 (7.1)

Data shown are for 14 participants who received zimislecel with ≥12 months of follow-up (2 participants in Part A, 4 participants in Part B, and 8 participants in Part C). Participants in Part A received half the dose of zimislecel (one participant in Part A received a second half dose after the first dose) and participants in Parts B and C received full dose of zimislecel in a single infusion.

Safety Data for all 22 Participants Dosed with Zimislecel

Table S7. Summary of Adverse Events

Category	Participants With AEs N=22 n (%)
Any adverse event	22 (100)
Adverse events by maximum severity	
Grade 1/Mild	3 (13.6)
Grade 2/Moderate	10 (45.5)
Grade 3/Severe	7 (31.8)
Grade 4/Life threatening	0
Grade 5/Death	2 (9.1)
Adverse events related to zimislecel	**10 (45.5)**
Adverse events related to surgical procedure	**11 (50.0)**
Adverse events related to immunosuppression	**22 (100.0)**
Adverse events related to ancillary drug	**18 (81.8)**
Discontinuation of trial due to adverse event	**0**

Data shown are for 22 participants who received zimislecel (2 participants in Part A, 4 participants in Part B, and 16 participants in Part C), including participants with <12 months of follow-up. Participants in Part A received half the dose of zimislecel (one participant in Part A received a second half dose after the first dose) and participants in Parts B and C received full dose of zimislecel in a single infusion.

Table S8. Summary of Most Common (≥6 Participants) Adverse Events

Preferred Term	Participants with AEs N=22 n (%)
Diarrhea	14 (63.6)
Headache	12 (54.5)
Nausea	11 (50.0)
Aspartate aminotransferase increased	9 (40.9)
Leukopenia	9 (40.9)
Rash	9 (40.9)
Alanine aminotransferase increased	8 (36.4)
Mouth ulceration	8 (36.4)
Neutropenia	8 (36.4)
Anemia	7 (31.8)
COVID-19	7 (31.8)
Tremor	7 (31.8)
Constipation	6 (27.3)
Pyrexia	6 (27.3)

Data shown are for 22 participants who received zimislecel (2 participants in Part A, 4 participants in Part B, and 16 participants in Part C), including participants with <12 months of follow-up. Participants in Part A received half the dose of zimislecel (one participant in Part A received a second half dose after the first dose) and participants in Parts B and C received full dose of zimislecel in a single infusion.

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Table S9. Summary of Serious Adverse Events by Preferred Term

Preferred Term	Participants With AEs N=22 n (%)
Any serious AEs	**9 (40.9)**
Neutropenia	3 (13.6)
Acute kidney injury	2 (9.1)
Cellulitis	2 (9.1)
COVID-19	1 (4.5)
Confusional state	1 (4.5)
Dehydration	1 (4.5)
Dementia	1 (4.5)
Electrolyte imbalance	1 (4.5)
Headache	1 (4.5)
Hypokalemia	1 (4.5)
Infusion-related hypersensitivity reaction	1 (4.5)
Malnutrition	1 (4.5)
Meningitis cryptococcal	1 (4.5)
Mental status changes	1 (4.5)
Migraine	1 (4.5)
Rash	1 (4.5)
Serum sickness	1 (4.5)
Small intestinal obstruction	1 (4.5)
Tenosynovitis	1 (4.5)
Urinary tract infection	1 (4.5)

Data shown are for 22 participants who received zimislecel. (2 participants in Part A, 4 participants in Part B, and 16 participants in Part C), including participants with <12 months of follow-up. Participants in Part A received half the dose of zimislecel (one participant in Part A received a second half dose after the first dose) and participants in Parts B and C received full dose of zimislecel in a single infusion.

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Data Sharing Statement

Vertex is committed to advancing medical science and improving patient health. This includes the responsible sharing of clinical trial data with qualified researchers. Proposals for the use of these data will be reviewed by a scientific board. Approvals are at the discretion of Vertex and will be dependent on the nature of the request, the merit of the research proposed, and the intended use of the data. Please contact CTDS@vrtx.com if you would like to submit a proposal or need more information.