COVER PAGE

Official Study Title: Efficacy and safety of a cryopreserved formulation of autologous CD34+ hematopoietic stem cells transduced *ex vivo* with EFS lentiviral vector encoding for human ADA gene in subjects with Severe Combined Immunodeficiency due to Adenosine Deaminase deficiency.

Statistical Analysis Plan Version 1.0, 25 January 2018

NCT02999984

Orchard therapeutics

Statistical Analysis Plan

Study Name:	**Efficacy and safety of a cryopreserved formulation of autologous CD34+ hematopoietic stem cells transduced ex vivo with EFS lentiviral vector encoding for human ADA gene in subjects with severe combined immunodeficiency (SCID) due to adenosine deaminase deficiency**
Protocol Number:	**OTL-101-4**
Protocol Version:	**4.0**
Protocol Date:	**08 September 2017**
SAP Version 1.0:	**25 January 2018**

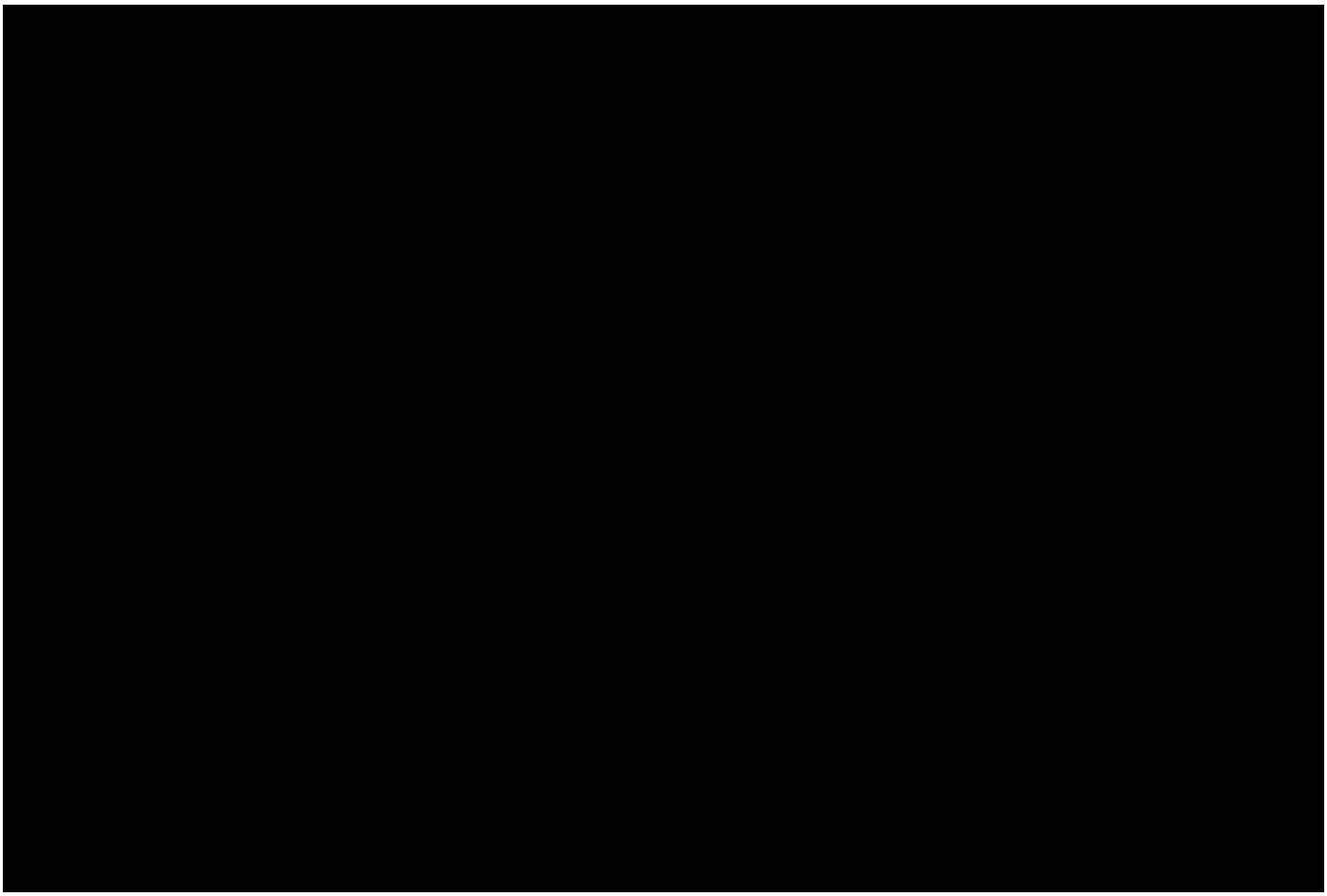

Table of Contents

List of Abbreviations

ADA	Adenosine deaminase
ADA-SCID	Adenosine deaminase deficient severe combined immunodeficiency
AE	Adverse event
ALP	Alkaline phosphatase
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
ANC	Absolute neutrophil count
AUC	Area under the curve
CI	Confidence interval
CMC	Chemistry, manufacturing and control
CT	Computer tomography
dAXP	Adenine deoxyribonucleotides
dPCR	Digital polymerase chain reaction
ECG	Electrocardiogram
EFS	Elongation Factor 1α Short form
ERT	Enzyme replacement therapy
FACS	Fluorescence-activated cell sorting
GTMP	Gene therapy medicinal product
Hb	Hemoglobin
HLA	Human leukocyte antigen
HSC	Hematopoietic stem cell
HSCT	Hematopoietic stem cell transplant
IgA	Immunoglobulin A
IgG	Immunoglobulin G
IgM	Immunoglobulin M
IgRT	Immunoglobulin replacement therapy
INR	International normalized ratio
LV	Lentiviral vector
MedDRA	Medical Dictionary for Regulatory Activities
MCH	Mean corpuscular hemoglobin
MCHC	Mean corpuscular haemoglobin concentration
MCV	Mean corpuscular volume
NK	Natural Killer
PBMC	Peripheral blood mononuclear cells
PCR	Polymerase chain reaction
PEG	Polyethylene-glycol
PHA	Phytohemagglutinin
PK	Pharmacokinetic
PRBC	Packed red blood cells
PT	Preferred term
PT	Prothrombin time
PTT	Partial thromboplastin time
qPCR	Quantitative polymerase chain reaction
RBC	Red blood cell(s)
RCL	Replication competent lentivirus
SAE	Serious Adverse Event

SAS®	Statistical Analysis System®
SCID	Severe combined immunodeficiency
SOC	System Organ Class
TCR	T-cell receptor
TREC	T-cell receptor excision circle
UCLA	University of California Los Angeles

1 Introduction

The primary objective of this clinical study is to further explore the safety and efficacy of OTL-101, an autologous, genetically modified CD34+ HSC based gene therapy for the treatment of ADA-SCID. The study will also be supportive of the analytical CMC comparability studies between the fresh and cryopreserved OTL-101 formulations. The following clinical outcomes, which are known to be good predictors of survival and event free survival: RBC ADA >0 Units, absolute CD3+ T-cell counts ≥ 200/mm3, and peripheral blood samples positive for vector sequences by qPCR (>1/10,000 cells), will be assessed as part of an interim analysis following completion of the 6 month follow-up visit.

2 Study Design

This is a prospective, non-randomized, single-cohort, longitudinal, single-center, clinical study designed to assess the efficacy and safety of OTL-101 cryopreserved formulation administered in ADA-SCID subjects aged between ≥30 days to <18 years of age, who are not eligible for an HLA-matched sibling/family donor and meeting the inclusion/exclusion criteria. This study aims to recruit 10 evaluable subjects.

The aim of this clinical study is also to assess the success of treatment at the subject level ("responder analysis") 6 months post GTMP administration, using predictive criteria for overall survival and event free survival and to compare data obtained from clinical studies using the fresh formulation of OTL-101.

Eligible subjects will be hospitalized to undergo the harvesting of autologous CD34+ cells. A backup harvest of non-transduced CD34+cells will be obtained during the harvesting procedure to be used in the event of i) product damage during the thawing of the GTMP that would prevent infusion of the GTMP although the patient has already been conditioned or ii) lack of engraftment/hematopoietic reconstitution 42 days post infusion of the protocol treatment. A failure of hematologic reconstitution is defined as at least two of the following: absolute neutrophil count (ANC) <200/mm3, platelets <20,000/mm3 without transfusions, haemoglobin (Hb) <8.0 g/dl without transfusions on three independent and consecutive determinations over at least 10 days beyond Day 42 from initial GTMP infusion.

The protocol treatment is a cryopreserved formulation of autologous CD34+ hematopoietic stem cells transduced ex vivo with EFS LV encoding for the human ADA gene (OTL-101). To enable the release of the GTMP for infusion, the cryopreserved OTL-101 product must meet various quality control criteria for safety, identity, viability, purity and potency. If OTL-101 meets the acceptance criteria and is released, the subjects will be readmitted for conditioning with busulfan. The OTL-101 product will be infused after a minimal interval of at least 24 hours following the completion of busulfan administration. Subjects may remain as an in-patient following autologous transplantation for approximately three to 35 days.
For subjects who have successfully received the OTL-101 product, PEG-ADA ERT will be discontinued at Day+30 (+/-3) after the transplant. After their discharge from hospital, the subjects will be seen at regular intervals to review their history, perform examinations and draw blood samples at Months 1, 3, 6, 9, 12, 18, and 24. Any medically-indicated interventions will be determined at these visits. Hematopoietic reconstitution will be assessed at Day 42, and in the event of no reconstitution the backup HSC sample will be administered. Hematopoietic reconstitution will be reassessed at the Month 6 visit and if there is still no evidence of cellular recovery, the subject will be given ERT and/or HSCT, if available, and deemed appropriate by

the Investigator. After Month 24 visit, the subjects will have completed the study and may enter a long-term follow-up study.

3 Study Schedule

Table 1: Schedule of Events – Screening to Day 24

Study Period	**Screening**	**Pre-harvest Assessment**	**Bone Marrow Harvest**	**Baseline**	**Conditioning**		**OTL-101 Treatment**	**Short-term Follow-up**						
Visit number	1	2[1]	2	3[1]	3	3	3	3	3	3	3	3	3	3
Assessment number		A	B	C	D	E	F	G	H	I	J	K	L	M
Visit window								+/-2 days						
Day	Within 60 days of Visit 2B	Up to 5 days prior to Visit 2B	Approx 30 day prior to Visit 3F	Day-10 to -4	Day -4 to -3	Day -2 to -1	Day 0	Day 1	Day 4	Day 8	Day 11	Day 15	Day19	Day 24
Informed Consent/Assent	X													
Eligibility check	X													
Demographic Data	X													
Medical History	X	X												
Physical examination	X	X		X										
Vital Signs	X			X			X[2]							
Weight (kg)	X	X			X									
Height (cm)	X													
Biochemistry sample[3]	X	X		X				X	X	X	X	X	X	X
Hematology sample[4]	X	X		X				X	X	X	X	X	X	X
PT or PTT/INR	X	X		X										
PB or BM cytogenetics[5]	X													
HIV-1/Hep B/Parvo B19	X													
Serum pregnancy test[6]	X	X		X			X							
Urinalysis[7]	X	X		X										
ECG	X													
Echocardiogram	X													
Chest X-ray[8]	X													
Pulse oxymetry	X			X										
Biopsy of skin lesions	X													
ADA-SCID diagnosis confirmation	X													
Questionnaire/QofL scale	X													
Infectious episodes	X			X										
Adverse events		X	X	X	X	X	X	X	X	X	X	X	X	X
Concomitant Medication[9]		X	X	X	X	X	X	X	X	X	X	X	X	X
Type and cross PRBC		X												
Bone marrow harvest			X											

Study Period	**Screening**	**Pre-harvest Assessment**	**Bone Marrow Harvest**	**Baseline**	**Conditioning**		**OTL-101 Treatment**	**Short-term Follow-up**						
Visit number	1	2[1]	2	3[1]	3	3	3	3	3	3	3	3	3	3
Assessment number		A	B	C	D	E	F	G	H	I	J	K	L	M
Visit window								+/-2 days						
Day	Within 60 days of Visit 2B	Up to 5 days prior to Visit 2B	Approx 30 day prior to Visit 3F	Day-10 to -4	Day -4 to -3	Day -2 to -1	Day 0	Day 1	Day 4	Day 8	Day 11	Day 15	Day19	Day 24
PRBC transfusion			X											
Immune function[10]				X[11]										
PBMC banking for RCL				X[12]										
Serum banking for RCL				X[12]										
Erythrocyte ADA enzymatic activity				X[12]										
Erythrocyte deoxyadenine nucleotide levels				X[12]										
Busulfan administration					X	X								
PK samples					X[13]	X[14]								
OTL-101 administration							X							

1. Subjects will be admitted to the hospital for Visits 2 and 3.
2. During the infusion of OTL-101 vital signs will be monitored according to standard institute protocols.
3. Biochemistry samples will include the assessment of parameters listed in Table 3. Albumin ALT, AST, ALP will be measured at all time points indicated in this table. Total bilirubin, total protein, total protein, creatinine, blood urea, sodium, potassium, chloride, and calcium will be measured at screening, baseline, Day 1 and all follow-up visits (Month 1 to Month 24). Magnesium and phosphate will be measured only at pre-harvest and baseline time points.
4. Hematology samples will include the assessment of parameters listed in Table 3 and will be measured at all time points indicated in this table.
5. If cytogenetic testing was not performed on cells from amniocentesis, assessment should be by karyotype, CGH, and WES
6. Serum pregnancy tests are required in females of child-bearing potential at screening, prior to bone marrow harvest, prior to conditioning (Busulfan) treatment, and before the infusion of OTL-101. Any positive test will immediately result in the withdrawal of the subject from the study and should be reported as per guidance in Section **Error! Reference source not found.**
7. Urinalysis will include the assessment of parameters listed in Table 3 and will be measured at all time points indicated in this table. Any abnormalities detected on dipstick at Screening, Pre-harvest and Baseline will require repeat dipstick and microscopic examinations.
8. If there is any evidence of severe lung disease on the screening chest X-ray, a CT scan is required in order to establish if the subject meets any of the specified pulmonary exclusion criteria defined in Section **Error! Reference source not found.**.
9. Standard post-transplant medication will be administered from Day 0 and recorded in the eCRF.
10. Immune function will be assessed absolute numbers of CD3+, CD4+ and CD8+ T-lymphocytes, CD19+ B-lymphocytes and CD56+/CD16+ (or CD56+/CD3-) NK cells, CD4+/CD45RA+ (naïve) and CD4+/CD45RO+ (memory) T cells, TREC levels, TCR Vβ usage, lymphocyte response to PHA (if sufficient cells), and serum immunoglobulin (IgG, IgA, IgM).
11. The immune function tests required for Visit 3, Assessment C can be performed at any point after provision of consent/assent and prior to Busulfan administration in younger subjects if deemed appropriate by the Investigator.
12. These test can be performed at anytime following BM harvest (Visit 2: Assessment B) and prior to OTL-101 administration (Visit 3: Assessment F) in younger subjects if deemed appropriate by the Investigator.

Study Period	**Screening**	**Pre-harvest Assessment**	**Bone Marrow Harvest**	**Baseline**	**Conditioning**		**OTL-101 Treatment**	**Short-term Follow-up**						
Visit number	1	2[1]	2	3[1]	3	3	3	3	3	3	3	3	3	3
Assessment number		A	B	C	D	E	F	G	H	I	J	K	L	M
Visit window								+/-2 days						
Day	Within 60 days of Visit 2B	Up to 5 days prior to Visit 2B	Approx 30 day prior to Visit 3F	Day-10 to -4	Day -4 to -3	Day -2 to -1	Day 0	Day 1	Day 4	Day 8	Day 11	Day 15	Day19	Day 24

13. PK blood samples are required at the end of Busulfan infusion, +1, 2, 4, 8, and 13 hours post-infusion (+/- 15 minutes). These values will be used to determine the second dose of Busulfan.
14. One PK blood sample is required immediately before the second dose of Busulfan is administered, and then at the end of Busulfan infusion, +1, 2, 4, 8, and 13 hours post-infusion (+/- 15 minutes).

Abbreviations: ADA=adenosine deaminase, ALP=alkaline phosphatase, ALT=alanine aminotransferase, AST=aspartate aminotransferase, BM=bone marrow, CGH= Comparative Genome Hybridization, CT=computer tomography, ECG=electrocardiogram, eCRF=electronic case report form, HIV=human immunodeficiency virus, IgA=immunoglobulin A, IgG=immunoglobulin G, IgM=immunoglobulin M, INR=international normalized ratio, NK=natural killer, PB=peripheral blood, PBMC=peripheral blood mononuclear cells, PHA= Phytohemagglutinin, PK=pharmacokinetic, PRBC=packed red blood cells, PT=prothrombin, PTT=prothrombin time, QofL=quality of life, RCL= Replication Competent Lentivirus, SCID=severe combined immunodeficiency, TCR= T-cell receptor, TREC= T-cell receptor excision circle, WES= whole exome sequencing.

Table 2: Schedule of Events – Month 1 to Month 24

Follow-up Period	**Month 1 (Day 30)**	**Day 42[1]**	**Month 3**	**Month 6**	**Month 9**	**Month 12**	**Month 18**	**Month 24**	**ET**
Visit number	4	5	6	7	8	9	10	11	N/A
Visit window	+/- 1 week	+/- 1 week	+/- 2 weeks	+/- 2 weeks	+/- 2 weeks	+/- 4 weeks	+/- 4 weeks	+/- 4 weeks	
Physical examination	X		X	X	X	X	X	X	X
Vital Signs	X		X	X	X	X	X	X	X
Weight (kg)	X		X	X	X	X	X	X	X
Height (cm)	X		X	X	X	X	X	X	X
Biochemistry sample[2]	X	X	X	X	X	X	X	X	X
Hematology sample[3]	X	X	X	X	X	X	X	X	X
Questionnaire/QofL scale				X		X	X	X	X
Infectious episodes	X		X	X	X	X	X	X	X
Adverse events	X	X	X	X	X	X	X	X	X
Concomitant Medication[4]	X	X	X	X	X	X	X	X	X
Record IgG and PEG-ADA therapy received	X	X	X	X	X	X	X	X	X
Immune function[5,6]	X		X	X	X	X	X	X	X
Serum banking for RCL			X	X		X		X	X
Vector integration analysis			X	X		X	X	X	
Leukaemia assessment[7]				X		X	X	X	X
T-cell proliferation to PHA				X	X	X	X	X	X
Antibodies to tetanus toxoid						X	X	X	X
Presence of ADA gene in PBMC/granulocytes and/or lineage-sorted cells			X	X		X		X	X
RCL assay in PBMC			X	X		X		X	X
ADA enzymatic assay	X		X	X	X	X	X	X	X
Deoxyadenine nucleotides in RBC	X		X	X	X	X	X	X	X
Responder assessment[8]				X					

1. Day 42 assessment will use laboratory parameters to confirm if hematologic reconstitution has occurred. If this has not happened by Day 42 rescue medication, as per standard institute procedure, will be implemented. Any treatment administered to the subject will be recorded in the eCRF.
2. Biochemistry samples will include the assessment of parameters listed in Table 3. Albumin ALT, AST, ALP will be measured at all time points indicated in this table. Total bilirubin, total protein, total protein, creatinine, blood urea, sodium, potassium, chloride, and calcium will be measured at screening, baseline, Day 1 and all follow-up visits (Month 1 to Month 24). Magnesium and phosphate will be measured only at baseline and the conditioning time points.
3. Hematology samples will include the assessment of parameters listed in Table 3 and will be measured at all time points indicated in this table.
4. Standard post-transplant medication will be administered from Day 0 and recorded in the eCRF.

5. Immune function will be assessed by absolute numbers of CD3+, CD4+ and CD8+ T-lymphocytes, CD19+ B-lymphocytes and CD56+/CD16+ (or CD56+/CD3-) NK cells, CD4+/CD45RA+ (naïve) and CD4+/CD45RO+ (memory) T cells, TREC levels, TCR Vβ usage, lymphocyte response to PHA (if sufficient cells), and serum immunoglobulin (IgG, IgA, IgM).
6. If IgG treatment is stopped, immunoglobulin levels will be required every month for the first 3 month after discontinuation. This is to ensure IgG levels hold before commencing vaccinations as per standard institute protocols.
7. A blood sample will be taken unless clinical/hematologic findings indicate potential leukoproliferation, whereby a bone marrow sample will be required. In addition to the time points specified in this table, this assessment should be performed if the nrLAM-PCR criteria is fulfilled (see Section **Error! Reference source not found.**).
8. Responder assessment includes: erythrocyte ADA enzyme activity, immune reconstitution, and gene-marked granulocytes as described in Section **Error! Reference source not found.**.

Abbreviations: ADA=adenosine deaminase, ALP=alkaline phosphatase, ALT=alanine aminotransferase, AST=aspartate aminotransferase, eCRF=electronic case report form, ET=early termination, IgA=immunoglobulin A, IgG=immunoglobulin G, IgM=immunoglobulin M, N/A=not applicable, NK=natural killer, PBMC=peripheral blood mononuclear cells, PCR=polymerase chain reaction, PEG= polyethylene-glycol, PHA= Phytohemagglutinin, QofL=quality of life, RBC=red blood cell, RCL= Replication Competent Lentivirus, TCR= T-cell receptor, TREC= T-cell receptor excision circle.

4 Study Objectives

4.1 Primary Objectives

There are two primary objectives for this study:

- The success of treatment at the subject level ("responder analysis") 6 months post OTL-101 infusion, defined as:
    - Evidence of erythrocyte ADA enzyme activity above baseline/pre-treatment level (>0 Units),
    - Evidence of immune reconstitution (absolute number of CD3 cells ≥200/mm3),
    - Detectable gene-marked granulocytes by differential polymerase chain reaction (dPCR)/qPCR (≥1/10,000 cells).

  This 6 month data will be used to compare the proportion of responders among subjects treated with the cryopreserved population with a comparable population obtained from one of the ongoing Phase I/II studies using the fresh formulation.
- Overall survival and event free survival at 12 months among ADA-SCID subjects treated with a cryopreserved formulation of OTL-101.
    - Overall survival is defined as the proportion of subjects alive.
    - Event-free survival is defined as the proportion of subjects with no "event", an "event" being:
        - the resumption of PEG-ADA ERT
        - the need for a rescue allogenic HSCT, or
        - death.

4.2 Secondary Objectives

The secondary objectives to be evaluated among ADA-SCID subjects treated with a cryopreserved formulation of OTL-101 include:

- confirmation of the overall survival and event free survival at 24 months,
- safety evaluation including infection rates,
- quality of life, and
- immune response.

4.3 Exploratory Objectives

Exploratory objectives include the measurement of biological correlates of efficacy including:

- percentage of gene marking in peripheral blood granulocytes,
- vector integration analysis,
- quantification of clonal diversity of vector integrants,
- T-cell receptor excision circle (TREC) and fluorescence-activated cell sorting (FACS) for T-cell receptor (TCR) V-β family use,
- ADA enzyme activity in erythrocytes, total adenine nucleotides in erythrocytes, and
- immune reconstitution.

5 Endpoints

5.1 Primary Endpoints

Primary endpoints for this study are:

- Response to OTL-101 treatment at 6 months post infusion based on the following parameters and their thresholds:
    - Erythrocyte ADA enzyme activity above baseline/pre-treatment level (>0 Units),
    - Absolute CD3+ T-cell counts ≥200/mm3, and
    - Peripheral blood samples positive for vector sequences by qPCR (≥1/10,000 cells).

  Subjects must meet all three criteria. Subjects not meeting one or more of these criteria will be designated a failure (non-responder) and will be withdrawn from the study. This data will be used for comparison with subject data from the ongoing Phase I/II study using the fresh formulation.
- Overall survival and event free survival at 12 months post GTMP administration.
    - Overall survival is defined as the proportion of subjects alive.
    - Event-free survival is defined as the proportion of subjects with no "event", an "event" being:
        - resumption of PEG-ADA ERT, or
        - the need for a rescue allogenic HSCT, or
        - death.

5.2 Secondary Endpoints

Secondary endpoints for this study are:

- Overall survival and event free survival 24 months post GTMP administration
- Immunoglobulin replacement therapies prior to and after gene therapy.
- Safety and tolerability.
- Performance outcomes and quality of life measured by the Karnofsky/Lansky scale and questions relevant to general well-being, school attendance and ability to practice sports, respectively.
- Severe infections or opportunistic infectious episodes, defined as infections or severe infections requiring hospitalization or prolonging hospitalization and/or documented infections by opportunistic pathogens (i.e. interstitial pneumonia, intractable diarrhoea).
- Response to tetanus vaccination.
- Immune reconstitution: T and B cell reconstitution.

5.3 Exploratory Endpoints

Exploratory endpoints for this study are laboratory correlates of efficacy that will be used to assess the level of gene correction, engraftment and immune reconstitution.

6 Timing of Analyses

The statistical analyses of efficacy data from this study will be performed in three stages:

- The first analysis will be carried out when a minimum of 5 subjects have completed 6-months follow-up post OTL-101 infusion. At this analysis, the success/failure of the OTL-101 cryopreserved infusion will be defined by the three criteria listed in the primary

objectives. This data will be compared with available data from UCLA ongoing Phase I/II study, treated with the OTL-101 fresh formulation. This will support the CMC comparability data between OTL-101 cryopreserved and fresh formulations. Secondary and exploratory endpoints will also be described and compared with the data from the fresh study.

- The second analysis will be carried out when all subjects have completed 12-months follow-up post GTMP administration. At this analysis, overall survival and event free survival will be evaluated. Secondary and exploratory endpoints, as well as baseline characteristics, subject disposition, and safety data will also be described.
- The third and final analysis will be performed to determine the overall survival and event free survival for all subjects 24 months post GTMP administration. Secondary and exploratory endpoints, as well as baseline characteristics, subject disposition, safety and busulfan pharmacokinetic (PK) data will also be described.

7 Analysis Populations

7.1 Efficacy Population

The efficacy population will be a modified intent-to-treat population including all subjects treated with OTL-101 within this protocol.

7.2 Safety Population

The safety population will consist of all subjects treated with OTL-101 within this protocol.

8 Statistical Methods

In general, continuous variables will be summarized using the mean, standard deviation, minimum, median, maximum and number of subjects. Categorical variables will be summarized using number of subjects and percentages.

8.1 Subject Disposition

8.1.1 Summary of Populations

Number and percentage (%) of subjects enrolled, treated, and in efficacy and safety populations will be summarized. The denominator for the percentages will be the number of subjects enrolled.

8.1.2 Summary of Withdrawals

Number of subjects enrolled, number treated, number (%) of subjects who completed the study period will be summarized along with the reason for withdrawal. The denominator for the percentages will be the number of subjects treated.

For subjects in whom the treatment failed, the number (%) who had a second gene therapy, a rescue HSCT or other therapy will be summarized, the denominator will be the number of subjects who failed treatment.

8.2 Demography and Baseline Characteristics

Demography and baseline characteristics will be summarized for the safety population.

8.2.1 Demography

Age in months will be calculated as:

12*(date informed consent signed – date of birth)/365.25.

Age, weight and height at screening will be summarized using descriptive statistics.

Sex, race, ethnicity and location will be summarized using the number (%) of subjects in each category.

8.2.2 Medical History

The number (%) of subjects reporting medical history will be summarized overall and by category.

8.2.3 Diagnosis of ADA-SCID

First symptoms and primary diagnosis will be summarized using the number (%) of subjects with each symptom/diagnosis.

Age at symptom onset and age at diagnosis in months will be calculated as

12*(date of onset/diagnosis – date of birth)/365.25.

Age at symptom onset and age at diagnosis in months will be summarized using descriptive statistics.

8.2.4 Disease Treatment History

The number (%) of subjects previously receiving PEG-ADA ERT, immunoglobulin therapy, prophylactic agents and other therapies will be summarized.

The number (%) of subjects with a previous transplant, type of transplant cells, donor type and outcome will be summarized.

Time since transplant in months will be calculated as

12*(date informed consent signed – date of transplant)/365.25.

8.2.5 Cytogenetics

Cytogenetics will be assessed at screening and categorised as normal or abnormal.

The number (%) of subjects falling into each category for type of test, source and result (normal/abnormal) will be summarized.

8.2.6 Coagulation Tests

Coagulation tests (prothrombin time (PT), partial thromboplastin time (PTT) and internal normalised ratio (INR)) will be evaluated over time prior to the OTL-101 infusion.

For each parameter, the actual value will be summarized at each visit using descriptive statistics.

Coagulation tests will be flagged against the normal range as low/normal/high. For each parameter, the number (%) of subjects with evaluations that were low/normal/high relative to the normal range will be summarized by visit. Out of range values will be assessed for their clinical significance. For each parameter, the number (%) of subjects with clinically significant evaluations will be summarized by visit.

8.2.7 Urinalysis

Urinalysis tests (pH, glucose, ketones, protein, bilirubin, blood, urobilinogen, nitrites, leukocytes, specific gravity) will be evaluated over time prior to the OTL-101 infusion.

The urinalysis evaluations are assessed as 'Normal/Negative' or 'Abnormal/Positive'. If any urine cultures are taken, the results will be categorized as 'positive', 'negative' or 'mixes'. These data will be listed.

8.2.8 ECG

An ECG will be taken at screening.

RR interval, PR interval, QRS interval and QT interval will be summarized using descriptive statistics.

The ECG will be categorised as 'normal'/ 'abnormal, not clinically significant'/ 'abnormal, clinically significant'. The number (%) of subjects with evaluations falling into each category will be summarized along with the number (%) with each type of abnormal finding.

8.2.9 Echocardiogram

An echocardiogram will be taken at screening.

Left ventricular ejection fraction will be summarized using descriptive statistics.

The echocardiogram will be categorised as 'normal'/ 'abnormal, not clinically significant'/ 'abnormal, clinically significant'. The number (%) of subjects with evaluations falling into each category will be summarized.

8.2.10 Chest X-ray and Computed Tomography (CT) Scan

A chest x-ray will be taken at screening, in addition a CT scan will be performed if the x-ray shows evidence of severe lung disease.

Both the chest x-ray and the CT scan will be categorised as 'normal'/ 'abnormal, not clinically significant'/ 'abnormal, clinically significant'. The number (%) of subjects with evaluations falling into each category will be summarized.

8.2.11 Skin Lesions

The number (%) of subjects with any suspicious skin lesions at screening, having a biopsy and falling into each category of result ('normal'/ 'abnormal, not clinically significant'/ 'abnormal, clinically significant') will be summarized.

8.2.12 Bone Marrow Harvest

The number (%) of subjects with each blood type, having a transfusion of PRBC and with each reason for a lack of back-up stem cells (if appropriate) will be summarized. If there is a repeated harvest, these data will be summarized for each evaluation.

Total number of nucleated cells and total CD34+ cell count in the bone marrow harvest will be summarized using descriptive statistics along with the number of mononuclear and total nucleated cells stored for back-up.

8.2.13 Busulfan Administration

For each administration of Busulfan, the duration of administration, dose given and AUC will be summarized using descriptive statistics. In addition, for Day -4/-3, the calculated Busulfan dose for Day -2/-1 will be summarized.

8.2.14 Busulfan Pharmacokinetics

For each administration of Busulfan, the PK parameters (C_{max}, T_{max}, $AUC_{0\text{-}t}$, $AUC_{0\text{-}13}$, $AUC_{0\text{-}\infty}$) will be summarized using descriptive statistics.

8.2.15 OTL-101 Administration

The number (%) of subjects who received OTL-101 will be summarized along with any reasons for non-administration.

The number (%) of subjects who were pre-medicated with acetaminophen or diphenhydramine will be summarized.

For each bag of product, the volume, number of CD34+ cells and the planned volume to be infused, the duration of infusion, the volume administered and the % cell viability will be summarized using descriptive statistics.

For each additional infusion, the number of CD34+ cells, the number per kg, the final product volume, the total cells in the final product, the %CD34+, the CD34+ cells per mL and the final volume to be infused will be summarized using descriptive statistics.

8.2.16 Exposure

Duration of follow-up in months will be calculated as:

12*(date of final evaluation – date of infusion + 1)/365.25.

Duration of follow-up will be summarized using descriptive statistics. In addition, the total subject-years of follow-up will be presented.

8.3 Efficacy Evaluations

8.3.1 Primary Efficacy Endpoints

8.3.1.1 Response to OTL-101 Treatment at 6 Months

Response to OTL-101 treatment at 6 months post administration will be based on the following criteria:

- ADA activity in red blood cells: change from baseline >0 Units,
- Absolute CD3+ T-cell count ≥200/mm3, and
- Detectable gene-marked granulocytes by differential polymerase chain reaction (dPCR)/qPCR ≥1/10,000 cells.

Subjects who meet all three criteria will be said to have mounted a response.

For any subjects with missing values at 6 months, the earliest assessments after month 6 will be used. In addition, if ADA activity at baseline is not available, any positive value will be considered as a positive response.

The number (%) of subjects mounting a response will be summarized and descriptively compared with the proportion from the UCLA "Fresh" study. In addition, the number (%) of subjects with a response to each of the individual criteria will be summarized.

8.3.1.2 Overall Survival at 12 Months

Overall survival will be evaluated using the time in months from transplant to either the subject's death or their last evaluation calculated as:

12*(date of death/last evaluation – date of transplant)/365.25.

The number (%) of subjects who were still alive at 12 months post-transplant will be summarized. For the purpose of this analysis, any subject who withdrew from the study prior to 12 months, who was known to be alive at the last evaluation, will have a status of "unknown". The proportion and exact 95% confidence interval (CI) for survivors at 12 months post-transplant will be calculated.

Overall survival will also be represented using a Kaplan-Meier curve for time to death. For the purpose of this analysis, any subject who withdrew from the study prior to 12 months and was known to be alive at the last evaluation will be censored at the date of their last evaluation. If appropriate, the proportion alive at 12 months and the associated 95% CI will be estimated from the Kaplan-Meier curve along with the median survival time (and interquartile range), if they can be estimated from the data.

8.3.1.3 Event Free Survival at 12 Months

Event-free survival will be evaluated using the time in months from transplant to either the first event or their last evaluation calculated as:

12*(date of event/last evaluation – date of transplant)/365.25.

For the purpose of this analysis, the events of interest are:

- death;
- reinstitution of PEG-ADA ERT;
- requirement for a second transplant.

Event-free survival will be summarized in the same way as overall survival described in Section 8.3.1.2.

8.3.2 Secondary Efficacy Endpoints

8.3.2.1 Overall and Event Free Survival at 24 Months

Overall and event-free survival at 24 months will be summarized in the same way as for the analyses at 12 months described in Sections 8.3.1.2 and 8.3.1.3.

8.3.2.2 Use of Immunoglobulin Replacement Therapy

The number (%) of subjects receiving immunoglobulin replacement therapy will be summarized by visit.

The time to cessation of Ig RT will be calculated (in months), using the stop date on the concomitant medication form, as:

12*(stop date of Ig RT – date of infusion) /365.25.

Time to cessation of IgRT will be summarized using descriptive statistics and, if appropriate, estimated from a Kaplan-Meier curve in which subjects who are still receiving IgRT will be censored at the time of their last visit.

The number (%) of subjects who have stopped treatment with IgRT at 12, 18 and 24 months without restarting at any subsequent time point will be summarized.

In addition, serum immunoglobulin levels (IgG, IgA and IgM) will be measured at each visit. Levels of IgG, IgA and IgM and changes from baseline will be summarized by visit using descriptive statistics. In addition, plots of the median value and range over time on a logarithmic scale will be produced for each parameter along with individual plots for each subject which will include age-dependent normal ranges provided by the study site.

8.3.2.3 Performance Outcomes and Quality of Life

The baseline evaluation for the quality of life outcomes will be the final evaluation prior to the OTL-101 infusion.

The number (%) of subjects choosing each answer to the questions “How have you been feeling in general?” and “Can the patient practice sports?” will be summarized at each visit. Changes from baseline to each subsequent visit will be summarized using the number (%) of subjects whose status improved/no change/deteriorated.

The number of days absent from school and the change from baseline will be summarized by visit.

Scores on the Lansky and Karnofsky performance scales and the change from baseline will be summarized by visit.

8.3.2.4 Rate of Severe Infections

Infections will be recorded as adverse events. The infections of interest in this study are severe infections or opportunistic infectious episodes, defined as infections requiring hospitalization or prolonging hospitalization and/or documented infections by opportunistic pathogens (e.g. interstitial pneumonia, intractable diarrhoea).

The number (%) of subjects reporting severe infections or opportunistic infectious episodes along with the number of events reported will be summarized. In addition, the infection rate will be calculated as the number of infections/the total follow-up time. The rate will be calculated both for the entire group and for each individual subject. The subject infection rate will be summarized using descriptive statistics. In addition to the rate for the entire follow-up period, the rate will be calculated for both the first 12 months following transplant and for the remaining study period.

8.3.2.5 Response to Tetanus Vaccination

Tetanus antibodies will be measured in subjects who have stopped immunoglobulin replacement and have received a tetanus vaccination.

Pre-vaccination and post-vaccination titres will be summarized by vaccination number.

Subjects will be to considered to have mounted a protective antibody response to tetanus if they have a titre of anti-tetanus antibodies >0.15 IU/ml. The number (%) of subjects who mounted a protective antibody response to tetanus will be summarized.

8.3.2.6 Immune Reconstitution

Immune reconstitution will be assessed using lymphocyte subsets. Absolute numbers and percentage of $CD3^+$, $CD4^+$, $CD8^+$ and $CD4^+/CD45RO^+$ T-lymphocytes, $CD19^+$ B-lymphocytes and $CD56^+/CD16^+$ NK cells will be measured.

For each of the lymphocyte subsets, the actual value and change from baseline will be summarized at each visit. The median value and range will be plotted over time on a logarithmic scale and individual counts will be plotted along with the age-dependent normal ranges provided by the study site.

8.3.3 Exploratory Efficacy Endpoints

8.3.3.1 Use of PEG-ADA Enzyme Replacement Therapy

The time to withdrawal of PEG-ADA ERT will be calculated (in months), using the stop date on the concomitant medication form, as:

12*(stop of PEG-ADA ERT – date of infusion) /365.25.

Time to withdrawal of PEG-ADA ERT will be summarized using descriptive statistics and, if appropriate, estimated from a Kaplan-Meier curve in which subjects who are still receiving PEG-ADA ERT will be censored at the time of their last visit.

8.3.3.2 T Cell Receptor Excision Circles (TREC)

TREC levels will be summarized at each visit using descriptive statistics. Plots of the median and range over time on a logarithmic scale will be produced. In addition, individual subject plots of TREC levels over time will be produced.

8.3.3.3 T Cell Receptor Vβ Panel Results

TCR V-β usage, categorized as normal/abnormal, will be summarized by visit and overall using the number (%) with an abnormal evaluation at any post-infusion visit.

8.3.3.4 T Lymphocyte Response to Phytohemagglutinin (PHA)

T lymphocyte response to PHA, categorized as normal/abnormal, will be summarized by visit and overall using the number (%) with an abnormal evaluation at any visit.

8.3.3.5 Engraftment

Engraftment of transduced cells will be assessed over time using vector gene marking in peripheral blood cells (PBMC, granulocytes, T-cells, B-cells, NK cells, myeloid cells). The vector copy numbers in each type of cell will be summarized using descriptive statistics. In addition, for each cell type, plots of the median and range over time on a logarithmic scale will be produced along with individual subject plots.

8.3.3.6 ADA Expression, Enzyme Activity and Detoxification

RBC ADA activity, deoxyadenine nucleotides, AXP, dAXP and %dAXP will be summarized by visit. The median value and range will be plotted over time on a logarithmic scale as will the values for individual subjects. Deoxyadenine nucleotide evaluations that are 'undetectable' will use the limit of quantitation in the summary.

ADA activity and deoxyadenine nucleotides in erythrocytes will also be categorized as normal/abnormal, not clinically significant/abnormal clinically significant. The number (%) of subjects with evaluations in each category will be summarized by visit.

8.3.3.7 Vector Integration Analysis

The number (%) of subjects with one integration site representing > 30% of total integration will be summarized by visit. The number (%) of subjects for whom one integration site contributed > 30% of total integration at two or more time points will also be summarized. In addition, the total number of integration sites will be summarized by visit.

8.4 Safety Evaluations

8.4.1 Adverse Events

All adverse events will be coded using MedDRA version 20.0 or higher.

8.4.1.1 Definition of Treatment-emergent Adverse Events

Adverse events (AEs) will be summarised on the basis of treatment emergence. An adverse event is considered to be treatment emergent if it occurred on or after the date and time of the

start of the IMP infusion. If the start time of the event is not present, any event starting on the same day as the infusion will be considered to be treatment emergent. If the start date of the adverse event is partial or missing, it will be considered to be treatment emergent if:

- the day is missing, the month and year are present and are the same as or after the month and year of the infusion and the end date is missing or is not before the date of infusion,
- the day and month are missing, the year is present and is the same as or after the year of the infusion and the end date is missing or is not before the date of infusion,
- the entire date is missing and the end date is missing or is not before the date of infusion.

8.4.1.2 Adverse Events

Treatment emergent adverse events will be summarized in an overall summary presenting the number (%) of subjects with:

- any AE,
- AE grade 2 or higher (at least moderate in severity),
- AE grade 3 or higher (at least severe in severity),
- treatment related AE,
- AE related to autoimmunity,
- AE leading to study discontinuation,
- any serious adverse event (SAE),
- treatment related SAE,
- SAE related to autoimmunity,
- SAE leading to death.

Incidence and frequency of treatment emergent AEs will be summarised by system organ class (SOC) and preferred term (PT):

- overall,
- by maximum severity,
- treatment related AEs,
- SAEs (if sufficient events),
- AEs leading to study discontinuation (if sufficient events).

In addition, a summary table for the most frequently reported treatment emergent adverse events (those reported in 2 or more subjects) will be presented by PT in descending order of incidence and frequency.

In each table, if the same SOC or PT is reported on multiple occasions for a single subject, it will only be included once in the summary. In the table by maximum severity, only the most severe occurrence for each SOC and PT for each subject will be included in the summary. In the table by maximum relationship, only the most related occurrence for each SOC and PT for each subject will be included in the summary.

Any adverse events that are not treatment emergent (occurred prior to the infusion) will be listed.

8.4.2 Concomitant Medications

Concomitant medications taken during the study will be listed.

8.4.3 Infectious Episodes

Number (%) of subjects with an active infection will be summarized at each visit along with the number (%) with each type/location of infection. In addition, an overall summary of the number (%) of subjects with an active infection and each type/location of infection at any time post-infusion will be presented.

8.4.4 Replication Competent Lentivirus (RCL) Testing

Number (%) of subjects testing positive for RCL will be summarized by visit and at any time.

8.4.5 Leukaemia Assessment

Blood samples for leukaemia assessments will be taken over time. The number (%) of subjects with potential leukoproliferation will be summarized by visit and at any time post-infusion.

The number (%) with a bone marrow sample taken, with morphology assessed, with an abnormal morphology assessment, tested for integration analysis and with an abnormal assessment for integration analysis will summarized by visit and at any time post-infusion.

8.4.6 Laboratory Evaluations

8.4.6.1 Haematology

Haematology parameters (red blood cell (RBC) count, hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), white blood cell count (WBC), neutrophils, lymphocytes, monocytes, eosinophils, basophils and platelet count) will be evaluated over time. The baseline evaluation for each parameter will be the final evaluation prior to the OTL-101 infusion.

For each parameter, the actual value and change from baseline will be summarized at each visit using descriptive statistics.

Hematology evaluations will be flagged against the normal range as low/normal/high. For each parameter, the number (%) of subjects with evaluations that were low/normal/high relative to the normal range will be summarized by visit. In addition, shift tables will be constructed comparing the flags at the baseline evaluation with the minimum and maximum evaluation post-infusion. Out of range values will be assessed for their clinical significance. For each parameter, the number (%) of subjects with clinically significant evaluations will be summarized by visit and at any time post-infusion.

8.4.6.2 Biochemistry

Biochemistry parameters (blood urea nitrogen, creatinine, total bilirubin, sodium, potassium, chloride, calcium, aspartate transaminase (AST), alanine transaminase (ALT), alkaline phosphatase (ALP), albumin, total protein, glucose, magnesium, phosphates) will be evaluated over time. The baseline evaluation for each parameter will be the final evaluation prior to the OTL-101 infusion.

For each parameter, the actual value and change from baseline will be summarized at each visit using descriptive statistics.

Biochemistry evaluations will be flagged against the normal range as low/normal/high. For each parameter, the number (%) of subjects with evaluations that were low/normal/high relative to the normal range will be summarized by visit. In addition, shift tables will be constructed comparing the flags at the baseline evaluation with the minimum and maximum evaluation post-infusion. Out of range values will be assessed for their clinical significance. For each parameter, the number (%) of subjects with clinically significant evaluations will be summarized by visit and at any time post-infusion.

8.4.7 Vital Signs, Height and Weight

Vital signs (temperature, pulse rate, respiratory rate, systolic and diastolic blood pressure and oxygen saturation), height and weight are measured over time.

The baseline evaluation for each of the vital signs, height and weight will be the final evaluation prior to the OTL-101 infusion.

For each parameter, the actual value and change from baseline will be summarized at each visit using descriptive statistics.

Any vital signs found to be out of range will be categorized as not clinically significant or clinically significant. For each vital sign, the number (%) of subjects with clinically significant evaluations will be summarized by visit and at any time post-infusion.

8.4.8 Physical Examination

Physical examinations will be performed over time. The number (%) of subjects with abnormal evaluations will be summarized by body system at each visit. An overall summary will also be produced for each body system showing the number (%) of subjects with normal evaluations at all pre-treatment evaluations who had one or more abnormal evaluations post-treatment.

9 Analysis Software

All summaries and analyses will be carried out using SAS® version 9.4.