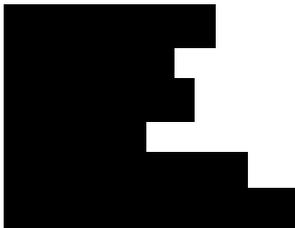




## CLINICAL STUDY PROTOCOL

**A SINGLE-ARM, OPEN-LABEL, MULTI-CENTRE, PHASE I/II STUDY EVALUATING THE SAFETY AND CLINICAL ACTIVITY OF AUTO3, A CAR T CELL TREATMENT TARGETING CD19 AND CD22 IN PAEDIATRIC AND ADULT PATIENTS WITH RELAPSED OR REFRACTORY B CELL ACUTE LYMPHOBLASTIC LEUKAEMIA.**

**Short Study Title:** AMELIA  
**Protocol Number:** AUTO3-PA1  
**Study Product:** AUTO3 for i.v. infusion  
**Development Phase:** I/II  
**Sponsor:** 

**Protocol Version:** Version 8.0  
**EudraCT Number:** 2016-004680-39  
**IND Number:** 18431

**Protocol Date:** 10 May 2019

**Compliance:** This study will be conducted in accordance with standards of Good Clinical Practice (as defined by the International Council on Harmonisation), ethical principles that have their origin in the Declaration of Helsinki and all applicable national and local regulations.

This protocol includes information and data that contain trade secrets and privileged or confidential information that is the property of the Sponsor (Autolus Limited). This information must not be made public without written permission from Autolus Limited. These restrictions on disclosure will apply equally to all future information supplied to you. This material may be disclosed to and used by your staff and associates as may be necessary to conduct the clinical study.

## ADMINISTRATIVE AND CONTACT INFORMATION

**Sponsor:** Autolus Limited  
[Redacted]  
Tel: [Redacted]

### Contact List

The List of Service Providers for the study will be maintained separately by Autolus Limited and maintained in the Trial Master File.

**Chief Investigator:** [Redacted]  
Tel: [Redacted]  
Email: [Redacted]

**Sponsor's Medical Monitor:** [Redacted]  
Tel: [Redacted]  
Mobile: [Redacted]  
Email: [Redacted]

**Sponsor's Project Manager:** [Redacted]  
Tel: [Redacted]  
Email: [Redacted]

**Notification of SAEs:** SAE Fax number: [Redacted]  
Email: [Redacted]

**24 Hour Safety Hotline:** [Redacted]  
Tel: [Redacted]

**SPONSOR SIGNATURE PAGE**

**Study Title:** A Single-Arm, Open-Label, Multi-Centre, Phase I/II Study Evaluating the Safety and Clinical Activity of AUTO3, a CAR T Cell Treatment Targeting CD19 and CD22 in Paediatric and Adult Patients with Relapsed or Refractory B Cell Acute Lymphoblastic Leukaemia.

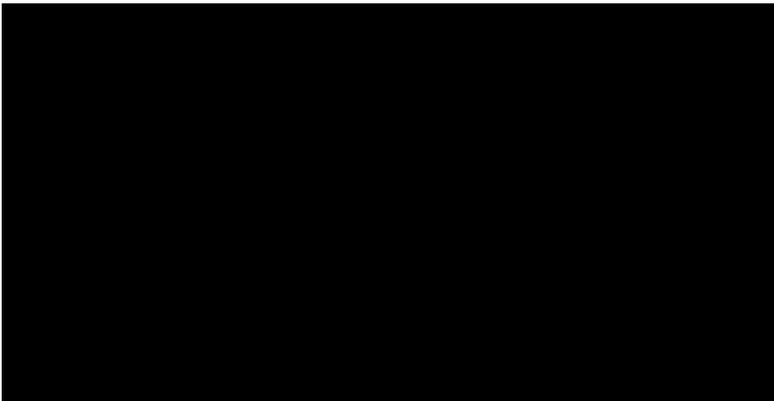
**Short Study Title:** AMELIA

**Protocol Number:** AUTO3-PA1

**Version Number:** Version 8.0

**Version Date:** 10 May 2019

I have read the protocol AUTO3-PA1 titled “A Single-Arm, Open-Label, Multi-Centre, Phase I/II Study Evaluating the Safety and Clinical Activity of AUTO3, a CAR T Cell Treatment Targeting CD19 and CD22 in Paediatric and Adult Patients with Relapsed Refractory B Cell Acute Lymphoblastic Leukaemia” and confirm that, to the best of my knowledge, the protocol accurately describes the design and conduct of the study.



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Date

(DD MMM YYYY)

## INVESTIGATOR SIGNATURE PAGE

**Study Title:** A Single-Arm, Open-Label, Multi-Centre, Phase I/II Study Evaluating the Safety and Clinical Activity of AUTO3, a CAR T Cell Treatment Targeting CD19 and CD22 in Paediatric and Adult Patients with Relapsed or Refractory B Cell Acute Lymphoblastic Leukaemia.

**Short Study Title:** AMELIA

**Protocol Number:** AUTO3-PA1

**Version Number:** Version 8.0

**Version Date:** 10 May 2019

I have read the protocol AUTO3-PA1 titled “A Single-Arm, Open-Label, Multi-Centre, Phase I/II Study Evaluating the Safety and Clinical Activity of AUTO3, a CAR T Cell Treatment Targeting CD19 and CD22 in Paediatric and Adult Patients with Relapsed Refractory B Cell Acute Lymphoblastic Leukaemia”. By signing this protocol, I agree to conduct the clinical study, after approval by an Institutional Review Board or Independent Ethics Committee (as appropriate), in accordance with the protocol, the principles of the Declaration of Helsinki (2008), the standards of Good Clinical Practice (as defined by the International Council on Harmonisation) and applicable regulatory requirements.

Changes to the protocol will only be implemented after written approval is received from Autolus Limited and the Institutional Review Board or Independent Ethics Committee (as appropriate), with the exception of medical emergencies. I will ensure that study staff fully understand and follow the protocol.

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Signature

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Date

Name and address:

(DD MMM YYYY)

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## PROTOCOL SYNOPSIS

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<b>Title</b>	A Single-Arm, Open-Label, Multi-Centre, Phase I/II Study Evaluating the Safety and Clinical Activity of AUTO3, a CAR T Cell Treatment Targeting CD19 and CD22 in Paediatric and Adult Patients with Relapsed or Refractory B Cell Acute Lymphoblastic Leukaemia
<b>Short Study Title</b>	AMELIA
<b>Protocol Number</b>	AUTO3-PA1
<b>Sponsor</b>	Autolus Limited
<b>Phase</b>	I/II
<b>Study Product</b>	AUTO3, an Advanced Therapy Investigational Medicinal Product, comprising of autologous enriched T cells retrovirally transduced to express two Chimeric Antigen Receptors (CAR), targeting cluster of differentiation (CD) 19 and CD22.
<b>Study Population</b>	Paediatric and adult patients aged $\geq 1$ year with B cell acute lymphoblastic leukaemia (ALL) who have relapsed or refractory disease.
<b>Study Duration</b>	The study will take approximately 5 years from recruitment to the last patient's last 24-month follow-up visit. The end of the trial will be 24 months after the last patient has received an AUTO3 infusion or the last patient last visit if this occurs earlier due to patient death or withdrawal.
<b>Overview</b>	<p>A single-arm, open-label, multi-centre, Phase I/II dose-escalation and expansion study to determine the safety and clinical activity of AUTO3 administered intravenously (i.v.) in paediatric and adult patients with relapsed or refractory B cell acute lymphoblastic leukaemia (ALL).</p> <p>AUTO3 is an autologous T cell product transduced to express two CARs targeting CD19 and CD22 using a retroviral vector. The CAR construct expresses endodomains of OX40 and CD3-<math>\zeta</math> on CD19 CAR, and 41BB-<math>\zeta</math> and CD3-<math>\zeta</math> on CD22 CAR. This first-in-human (FIH) study will assess the safety of AUTO3, evaluate the dose and determine dosing schedule for Phase II of the study, and evaluate the preliminary efficacy of the AUTO3 in paediatric and adult patients with relapsed or refractory B cell ALL.</p>

**Primary Objective(s) and Endpoints**

The primary objectives of the study are as follows:

Objectives	Endpoints
<b>Phase I</b>	
To assess the overall safety and tolerability of AUTO3 administration.	Incidence of Grade 3-5 toxicity occurring within the dose limiting toxicity (DLT) period (30 days after last dose) of AUTO3 infusion.
To confirm and evaluate the recommended Phase II dose (RP2D) and dosing schedule, and confirm maximum tolerated dose (MTD), if an MTD exists, of AUTO3 in both paediatric and adult patients.	Frequency of DLT of AUTO3.
<b>Phase II</b>	
To evaluate the anti-leukaemic effect of AUTO3 in paediatric and young adult patients (aged 1-24 years).	Proportion of patients achieving morphological remission (complete response [CR] or complete response with incomplete recovery of counts [CRi]) and minimal residual disease (MRD) negative response in bone marrow (BM) within 30 (±3) days post first AUTO3 infusion  In patients with isolated central nervous system (CNS) disease, anti-tumour effect will be assessed by whether patients clear their cerebrospinal fluid disease with ongoing BM CR, within 30 (±3) days post first AUTO3 infusion.

**Study Design**

This is a Phase I/II, open-label, multi-centre study to characterise the safety and clinical activity of AUTO3 when administered to paediatric and adult patients with relapsed or refractory B cell ALL. The study will consist of two parts: a Phase I, dose escalation in paediatric (>1 years) and adult patients followed by a Phase II, expansion in only paediatric and young adult patients (≥1-24 years). Both parts of the study will involve patients going through the following five sequential stages: screening, leukapheresis, pre-conditioning, treatment, and follow-up.

**Phase I (Dose Escalation):** To identify the optimal dose schedule (based on safety, tolerability, and anti-leukaemic activity) of AUTO3 based on disease burden, 36-60 patients with ALL will be treated in the dose escalation phase starting at 1.0 x 10<sup>6</sup>/kg CD19/CD22 CAR-positive T cells administered as a single or split dose based on disease burden. Then escalating to 3 x 10<sup>6</sup>/kg, and 5 x 10<sup>6</sup>/kg CD19/CD22 CAR-positive

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T cells administered in a similar manner.

Patients  $\geq 25$  years of age will enroll onto adult ALL dose cohorts. Adult ALL dose escalation may start at the highest paediatric dose considered safe after dosing 3 patients. Dosing will be based on a fixed dose which is equivalent to a dose in a 70 kg person.

**Phase II (Dose Expansion):** To further characterise the safety and assess the efficacy of AUTO3 at the recommended dose schedule confirmed in Phase I; up to 24 paediatric and young adult patients will be treated in Phase II.

Biomarkers relating to the CAR T cells and tumours will be evaluated in all patients. All patients enrolled in Phases I and II of the study will attend clinic visits for up to 24 months post AUTO3 infusion for study-specific assessments including adverse event (AE) assessments, physical examination, and laboratory and immunology tests.

After completion of the 24-month follow-up period, or following AUTO3 treatment and early withdrawal from this study, all patients will be eligible to be followed until death or for up to 15 years from the first AUTO3 infusion under a separate long-term follow-up study protocol.

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**Number of Patients**

Up to 100 patients in total are expected to be enrolled into Phase I and Phase II of the study and up to 84 patients in total are anticipated to be treated with AUTO3.

- **Phase I (Escalation):** Up to 33-46 patients in total (up to 3-6 patients per cohort)
  - 24-36 patients in the paediatric / young adult patient cohorts (age 1-24 years)
  - 12-24 patients in the adult patient cohorts ( $\geq 25$  years)

- **Phase II (Expansion):** Dose expansion: up to 24 evaluable patients (aged 1-24 years) in total, using a Simon's 2-stage optimal design.

Simon's 2-stage design (Simon 1989) will be used for Phase II. The null hypothesis that the true response rate is 25% will be tested against a 1-sided alternative. In the first stage, nine patients will be accrued. If there are two or fewer responses in these nine patients, the study will be stopped. Otherwise, 15 additional patients will be accrued for a total of 24. The null hypothesis will be rejected if 10 or more responses are observed in 24 patients. This design yields a 1-sided type I error rate of 5% and 80% power when the true response rate is 50%.

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**Criteria for Eligibility**

Only patients whose leukapheresate sample has been successful in generating AUTO3 in adequate quantity will be treated. The key criteria for patient eligibility are summarised as follows:

**INCLUSION CRITERIA**

**PAEDIATRIC AND YOUNG ADULT PATIENTS**

1. Male or female paediatric and young adult patients (aged 1-24 years) with high risk (HR) relapsed or refractory B-lineage ALL **and**
  - a. Any BM relapse or CNS relapse with detectable BM disease ( $>10^{-4}$ ) by flow cytometry/molecular MRD after allogeneic stem cell transplant (SCT) and must be  $\geq 6$  months from SCT at the time of AUTO3 infusion, OR
  - b. High Risk first relapse (as per International Study for Treatment of Childhood Relapsed ALL criteria [Appendix 6](#)), OR
  - c. Standard risk relapse patients with HR cytogenetics (HR defined as mixed linkage leukaemia gene rearrangement (KMT2A), intra-chromosomal amplification of chromosome 21 amplification, near-triploidy (60-78 chromosomes) or near-haploidy ( $<30$  chromosomes) and low hypodiploidy (30-39 chromosomes), OR
  - d. Second or greater relapse, OR
  - e. Bone marrow MRD  $\geq 10^{-3}$  prior to planned SCT, OR
  - f. Any on-treatment relapse in patients aged 16-24 years

**Phase II Only (Criteria in addition to those described above)**

  - g. Primary refractory disease defined as MRD  $\geq 5\%$  blasts in the BM by flow cytometry or molecular assay following frontline induction therapy. For good risk cytogenetics, MRD  $>0.1\%$  at Week 9 of consolidation chemotherapy is also required (*Note: For patients with good risk cytogenetics the leukapheresis may be done before starting consolidation therapy but manufacturing and AUTO3 infusion will only occur if MRD is  $>0.1\%$  at Week 9 of consolidation*), OR
  - h. Patients with Philadelphia chromosome-positive ALL are eligible if they are intolerant to or have failed 2 lines of tyrosine kinase inhibitor therapy, or if tyrosine kinase inhibitor therapy is contraindicated OR
  - i. Isolated CNS relapse (post-SCT or pre-SCT if high risk, age 16-24 years on therapy relapse or  $\geq 2^{\text{nd}}$  relapse) but with  $\leq$  CNS Grade 2 disease at time of enrolment.
2. Documentation of CD19 and or CD22 expression on leukaemic blasts in the BM, peripheral blood, or cerebrospinal fluid by flow cytometry within 3 months of screening.

3. Detectable disease in the BM at a level  $\geq 10^{-4}$  by molecular or flow cytometry based methods (Phase I only) at enrolment (patients developing  $\leq 10^{-4}$  BM disease due to bridging therapy may continue to receive AUTO3).
4. Absolute lymphocyte count  $\geq 0.5 \times 10^9/L$  at enrolment.
5. Adequate renal, hepatic, pulmonary, and cardiac function defined as:
  - Serum creatinine based on age/gender  $\leq 1.5x$  upper limit of normal (ULN).
  - Serum alanine aminotransferase/aspartate amino-transferase  $\leq 5x$  ULN.
  - Total bilirubin  $\leq 2x$  ULN, except in subjects with Gilbert's syndrome.
  - Left ventricular shortening fraction  $\geq 28\%$  confirmed by echocardiogram, or left ventricular ejection fraction  $\geq 45\%$  confirmed by echocardiogram.
  - Baseline oxygen saturation  $>92\%$  on room air.
6. Karnofsky (age 10-24 years) or Lansky (age  $<10$  years) score  $\geq 50\%$ .
7. Willing and able to give written, informed consent to the current study (patient and/or parent or legal guardian).

## **INCLUSION CRITERIA**

### **ADULT PATIENTS**

1. Age 25 or older.
2. Eastern cooperative oncology group (ECOG) performance status of 0 or 1.
3. Relapsed or refractory B-precursor ALL defined as one of the following:
  - a. Primary refractory disease
  - b. First relapse if first remission  $\leq 12$  months
  - c. Relapsed or refractory disease after 2 or more lines of systemic therapy
  - d. Relapsed or refractory disease after allogeneic transplant provided individual is at least 6 months from stem cell transplant at the time of infusion.
4. Patients with Philadelphia chromosome-positive ALL are eligible if they are intolerant to or have failed 2 lines of tyrosine kinase inhibitor therapy, or if tyrosine kinase inhibitor therapy is contraindicated,
5. Documentation of CD19 and/or CD22 expression on leukaemic blasts in the BM, peripheral blood, or CSF by flow cytometry within 3 months of screening.
6. For females of childbearing potential (defined as  $<24$  months after

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last menstruation or not surgically sterile), a negative serum or urine pregnancy test must be documented at screening, prior to pre-conditioning and confirmed before receiving the first dose of study treatment.

For females who are not postmenopausal (<24 months of amenorrhea) or who are not surgically sterile (absence of ovaries and/or uterus), two methods of contraception comprising of one highly effective method of contraception together with a barrier method must be used during the treatment period and for at least 12 months after the last dose of study treatment. They must agree not to donate eggs (ova, oocytes) for the purposes of assisted reproduction during the study and for 12 months after receiving the last dose of study drug (please refer to [Appendix 5](#)).

7. For males, it must be agreed that two acceptable methods of contraception are used (one by the patient – usually a barrier method, and one highly effective method by the patient’s partner as defined in [Appendix 5](#)) during the treatment period and for at least 12 months after the last dose of study treatment and that sperm will not be donated during the treatment period and for at least 12 months after the last dose of study treatment.
8. Absolute lymphocyte count  $\geq 0.5 \times 10^9/L$  at enrolment
9. Adequate renal, hepatic, pulmonary, and cardiac function defined as:
  - a. Serum alanine aminotransferase/aspartate aminotransferase  $\leq 2.5 \times ULN$ .
  - b. Creatinine clearance (as estimated by Cockcroft Gault)  $\geq 60$  cc/min.
  - c. Total bilirubin  $\leq 1.5$  mg/dl, except in subjects with Gilbert's syndrome.
  - d. Left ventricular ejection fraction (LVEF)  $\geq 45\%$  confirmed by ECHO or multigated acquisition (MUGA).
  - e. Baseline oxygen saturation  $>92\%$  on room air.

## **EXCLUSION CRITERIA**

Patients meeting any of the following exclusion criteria must not be enrolled into the study:

### **PAEDIATRIC AND YOUNG ADULT PATIENTS**

1. Isolated extra-medullary disease relapse (In Phase II of the study, patients with isolated CNS relapse post-SCT or pre-SCT if high risk, age 16-24 years on therapy relapse or  $\geq 2^{\text{nd}}$  relapse with  $\leq$  CNS Grade 2 disease at time of enrolment are eligible).
2. Active CNS involvement of ALL, defined by CNS Grade 3 per National Comprehensive Cancer Network guidelines. Patients

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- developing CNS Grade 3 disease at any time after enrolment will also be excluded.
3. Active infectious bacterial or viral disease (hepatitis B virus, hepatitis C virus, human immunodeficiency virus, human T-lymphotropic virus, syphilis, West Nile (US only) or Zika viruses (US only)) requiring i.v. anti-microbials for treatment.
  4. Females who are pregnant or lactating.
  5. Females of child-bearing potential (defined as all females physiologically capable of becoming pregnant) and post-pubertal male participants who are unwilling to use highly effective methods of contraception during the treatment period and for a period of 1 year after the AUTO3 infusion. *Note: Examples of highly effective contraception methods are described in [Appendix 5](#).*
  6. Inability to tolerate leukapheresis.
  7. Prior CD19 or CD22 targeted therapy with Grade 4 toxicity (except for haematological toxicity) or  $\geq$ Grade 3 cytokine release syndrome (CRS) or  $\geq$ Grade 3 drug-related CNS toxicity. CD22 targeted therapy such as inotuzumab ozogamicin in patients that are CD19 negative (unless it is demonstrated that this therapy had no effect on CD22 target expression). In Phase II, patients with prior CAR therapy will be excluded.
  8. Pre-existing significant neurological disorder (other than CNS involvement of underlying haematological malignancy).
  9. Stem Cell Transplant patients only: active significant (overall Grade  $\geq$ 2, Seattle criteria) acute graft versus host disease (GVHD) or moderate/severe chronic GVHD (National Institutes of Health consensus criteria) requiring systemic steroids or other immunosuppressant within 4 weeks of enrolment.
  10. The following medications are excluded:
    - a. Steroids: Therapeutic doses of steroids must be stopped  $>72$  hours prior to AUTO3 infusion and leukapheresis. However, physiological replacement doses of steroids are allowed:  $<12$  mg/m<sup>2</sup>/day hydrocortisone or equivalent.
    - b. Allogeneic cellular therapy: Any donor lymphocyte infusions must be completed  $>6$  weeks prior to AUTO3 infusion.
    - c. Graft versus host disease therapies: Any drug used for GVHD must be stopped  $>4$  weeks prior to AUTO3 infusion.
    - d. Chemotherapy: Should be stopped 1 week prior to leukapheresis and 2 days prior to starting pre-conditioning chemotherapy.
    - e. Intrathecal therapy: Methotrexate within 4 weeks and other intrathecal chemotherapy (e.g. Ara-C) within 2 weeks prior to

starting pre-conditioning chemotherapy.

- f. Live vaccine  $\leq 4$  weeks prior to enrolment.
11. Known allergy to albumin, dimethyl sulfoxide, cyclophosphamide or fludarabine.
  12. Any other condition which in the Investigator's opinion would prevent the patient from undergoing protocol-based therapy.

## **EXCLUSION CRITERIA**

### **ADULT PATIENTS**

1. Isolated extramedullary disease
2. Diagnosis of Burkitt's leukaemia/lymphoma according to World Health Organization (WHO) classification or chronic myelogenous leukaemia lymphoid blast crisis
3. Females who are pregnant or lactating.
4. History or presence of clinically relevant (CNS) pathology such as epilepsy, paresis, aphasia, stroke within 3 months prior to enrolment, severe brain injuries, dementia, Parkinson's disease, cerebellar disease, organic brain syndrome, uncontrolled mental illness, or psychosis.
5. Presence of CNS-3 disease or CNS-2 disease with neurological changes. Patients developing CNS Grade 3 disease or symptomatic CNS-2 disease at any time after enrolment will also be excluded.
6. Clinically significant, uncontrolled heart disease (New York Heart Association Class III or IV heart failure, uncontrolled angina, severe uncontrolled ventricular arrhythmias, sick-sinus syndrome, or electrocardiographic evidence of acute ischaemia or Grade 3 conduction system abnormalities unless the patient has a pacemaker) or a recent (within 12 months) cardiac event.
  - a. Uncontrolled cardiac arrhythmia (patients with rate-controlled atrial fibrillation are not excluded).
  - b. Evidence of pericardial effusion.
7. Patients with a history (within 3 months) or evidence of pulmonary embolism requiring ongoing therapeutic anticoagulation at the time of pre-conditioning.
8. Patients with active gastrointestinal (GI) bleeding.
9. Patients with any major surgical intervention in the last 3 months.
10. Active infectious bacterial or viral disease or fungal (hepatitis B virus, hepatitis C virus, HIV, HTLV, syphilis, West Nile virus [US only] or Zika virus [US only]) requiring treatment with i.v. antimicrobials.
11. History of autoimmune disease (e.g. Crohn's, rheumatoid arthritis, systemic lupus) resulting in end organ injury or requiring systemic

- immunosuppression/systemic disease modifying agents within the last 24 months. Any autoimmune disease with CNS involvement.
12. History of other malignant neoplasms unless disease free for at least 24 months (carcinoma *in situ*, non-melanoma skin cancer, breast or prostate cancer on hormonal therapy allowed).
  13. History of concomitant genetic syndrome such as Fanconi anemia, Shwachman-Diamond syndrome, Kostmann syndrome or any other known BM failure syndrome.
  14. Stem cell transplant patients only: Active significant (overall Grade  $\geq$ II, Seattle criteria) acute GVHD or moderate/severe chronic GVHD (NIH consensus criteria) requiring systemic steroids or other immunosuppressants within 4 weeks of enrolment.
  15. Prior CD19 or CD22 targeted therapy other than blinatumomab and inotuzumab ozogamicin.
  16. The following medications are excluded:
    - a. Steroids: Therapeutic doses of corticosteroids within 7 days of leukapheresis or 72 hours prior to AUTO3 administration. However, physiological replacement, topical, and inhaled steroids are permitted.
    - b. Immunosuppression: Immunosuppressive medication must be stopped  $\geq$ 2 weeks prior to leukapheresis or AUTO3 infusion
    - c. Allogeneic cellular therapy: Any donor lymphocyte infusions must be completed  $>$ 6 weeks prior to AUTO3 infusion.
    - d. Graft versus host disease therapies: Any drug used for GVHD must be stopped  $>$ 4 weeks prior to AUTO3 infusion.
    - e. Chemotherapy including TKIs for Philadelphia chromosome-positive ALL: Should be stopped 1 week prior to leukapheresis or 2 days prior to starting pre-conditioning chemotherapy.
    - f. Treatment with alemtuzumab within 6 months prior to leukapheresis, or treatment with clofarabine or cladribine within 3 months prior to leukapheresis.
    - g. Live vaccine  $\leq$ 4 weeks prior to enrolment.
    - h. Intrathecal therapy: Methotrexate within 4 weeks and other intrathecal chemotherapy (e.g. Ara-C) within 2 weeks prior to starting pre-conditioning chemotherapy.
    - i. Systemic inhibitory/stimulatory immune checkpoint - At least 2 half-lives must have elapsed from any prior systemic inhibitory/stimulatory immune checkpoint molecule therapy prior to enrolment.
  17. Presence of any indwelling line or drain (e.g., percutaneous

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nephrostomy tube, indwelling Foley catheter, biliary drain, or pleural/peritoneal/pericardial catheter). Ommaya reservoirs and dedicated central venous access catheters such as a Port-a-Cath or Hickman catheter are permitted.

18. Research participants receiving any other investigational agents, or concurrent biological, chemotherapy, or radiation therapy.
19. Inability to tolerate leukapheresis.
20. Patients, who in the opinion of the Investigator, may not be able to understand or comply with the safety monitoring requirements of the study or unlikely to complete all protocol-required study visits or procedures, including follow-up visits.

**Note:** Patients failing any of the eligibility criteria, after leukapheresis or AUTO3 product manufacture, such as those with progressive CNS disease and are discontinued may re-enter the study when they are considered to be eligible again. If AUTO3 product has been manufactured they can be treated at the current dose using the original product if it continues to meet release criteria.

**For Pre-conditioning or AUTO3 Infusion:** Patients meeting any of the following exclusion criteria must not be given pre-conditioning or treated with AUTO3 or treatment should be delayed until they no longer meet these criteria:

1. Severe intercurrent infection at the time of, or within 14 days of, scheduled AUTO3 infusion.
2. Requirement for supplementary oxygen at the time of scheduled pre-conditioning or AUTO3 infusion.
3. Allogeneic transplant recipients with active significant acute GVHD overall Grade  $\geq$ II or moderate/severe chronic GVHD requiring systemic steroids at the time of scheduled pre-conditioning or AUTO3 infusion.

**Note:** *Such patients will be excluded until the patient is GVHD free and off steroids.*

4. Significant deterioration of renal or hepatic function (Serum creatinine  $\geq$ 1.8 x ULN, Serum alanine aminotransferase or aspartate aminotransferase  $\geq$ 6 x ULN, Total bilirubin  $\geq$ 3 x ULN, except in subjects with Gilbert's syndrome) prior to pre-conditioning.

**Note:** *Patient with  $\geq$ Grade 3 CRS,  $\geq$ Grade 2 neurotoxicity or a DLT following the first split dose should not receive the second split dose.*

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<b>Study Product Dose, Dosing Regimen, and Administration</b>	<p>Eligible patients will receive a single or split dose i.v. infusion of AUTO3 following pre-conditioning treatment. The AUTO3 product contains both transduced (CD19/CD22 CAR-positive) and non-transduced cells. The dose is expressed as the number of CD19/CD22 CAR-positive T cells. The following dose levels are planned in Phase I:</p> <p>Paediatric and Young Adult Doses</p> <ul style="list-style-type: none"><li>• Dose Level 1: <math>1.0 \times 10^6</math>/kg CD19/CD22 CAR-positive T cells</li><li>• Dose Level 2: <math>3.0 \times 10^6</math>/kg CD19/CD22 CAR-positive T cells</li><li>• Dose Level 3: <math>5.0 \times 10^6</math>/kg CD19/CD22 CAR-positive T cells</li><li>• Dose Level 4: <math>10.0 \times 10^6</math>/kg CD19/CD22 CAR-positive T cells</li></ul> <p>Adult Doses</p> <ul style="list-style-type: none"><li>• <math>350 \times 10^6</math> CD19/CD22 CAR-positive T cells</li><li>• <math>700 \times 10^6</math> CD19/CD22 CAR-positive T cells</li></ul> <p>Each dose level (both age groups) will have 2 cohorts: one for patients with &lt;25% blasts who will receive a single dose and another for patients with <math>\geq 25\%</math> blasts (on Day -7) who will receive a split dose (approx. 1/3<sup>rd</sup>, 2/3<sup>rd</sup> split), second dose given between Days 3-10.</p>
<b>Pre-conditioning Treatment</b>	<p>All patients will receive a pre-conditioning regimen using fludarabine <math>30 \text{ mg/m}^2</math> i.v. over 30 minutes immediately followed by cyclophosphamide <math>500 \text{ mg/m}^2</math> i.v. over 30 minutes. Fludarabine will be given on Days -6, -5, -4, and -3 (total dose <math>120 \text{ mg/m}^2</math>) and cyclophosphamide will be given on Days -6 and -5 (total dose <math>1000 \text{ mg/m}^2</math>) before AUTO3 infusion.</p>
<b>Safety Evaluation</b>	<p>Safety will be assessed by physical examination, vital signs, laboratory tests, AE and serious AE monitoring, and concomitant medication usage. The severity of AEs will be assessed using the National Cancer Institute Common Terminology Criteria for Adverse Events (version 5.0), with the exception of CRS and CAR associated neurotoxicity, which will be graded according to the CRS grading.</p>
<b>Efficacy Evaluation</b>	<p>Efficacy will be evaluated by determining the proportion of patients achieving morphological CR, or CRi and/ or MRD-negative response in bone marrow (BM) within 30 (<math>\pm 3</math>) days post first AUTO3 infusion.</p>
<b>Biomarker Evaluation</b>	<p>Biomarker evaluation will include the following:</p> <ul style="list-style-type: none"><li>• Expansion and persistence of AUTO3 as determined by quantitative polymerase chain reaction and/or flow cytometry on the peripheral blood and BM.</li><li>• [REDACTED]</li></ul>

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<b>Special Study Procedures</b>	All patients will undergo an unstimulated leukapheresis for AUTO3 generation.
<b>Statistical Analysis</b>	<p>The data will be summarised using descriptive statistics. Continuous variables will be summarised using the number of observations, mean, standard deviation, median, and range as appropriate. Categorical values will be summarised using the number of observations and percentages as appropriate. Time-to-event endpoints will be estimated using Kaplan-Meier methodology.</p> <p>Data from all treated patients (safety analysis set) will be used for safety analyses and will be summarised by dose level. All treated patients (efficacy analysis set) and other subsets will be defined for the efficacy analyses.</p>
<b>Interim Analyses</b>	<p>An interim analysis on safety and preliminary efficacy will be performed when all Phase I patients have been followed for a minimum of 3 months. In Phase II of the study, an interim analysis will be performed when 9 patients have been evaluated for response. The study will be terminated if less than 3 responses are observed in 9 patients.</p>

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## LIST OF ABBREVIATIONS

<b>Abbreviation</b>	<b>Definition</b>
ACD-A	Anticoagulant citrate dextrose solution A
AE	Adverse event
ALL	Acute lymphoblastic leukaemia
BM	Bone marrow
CAPD	Cornell Assessment of Pediatric Delirium
CAR	Chimeric antigen receptor
CD3- $\zeta$ , -19, -20, -28, -134, -137	Cluster of differentiation 3, 19, 20, 28, 134, 137
CNS	Central nervous system
CR	Complete response
CRES	CAR-T-Cell-Related Encephalopathy Syndrome
CRi	Complete response with incomplete recovery of counts
CRS	Cytokine release syndrome
CSF	Cerebrospinal fluid
CT	Computerised tomography
CTCAE	Common Terminology Criteria for Adverse Events
CY	Cyclophosphamide
DFS	Disease-free survival
DIC	Disseminated intravascular coagulation
DLT	Dose limiting toxicity
DMSO	Dimethyl sulfoxide
DNA	Deoxyribonucleic acid
ECG	Electrocardiogram
ECHO	Echocardiogram
ECOG	Eastern Cooperative Oncology Group
eCRF	Electronic Case Report Form
EDC	Electronic data capture
EDTA	Ethylenediaminetetraacetic acid
EFS	Event-free survival
EU	European Union
FDA	Food and Drug Administration
FIH	First-in-human

<b>Abbreviation</b>	<b>Definition</b>
FLU	Fludarabine
FLU-CY	Fludarabine and cyclophosphamide
GCP	Good Clinical Practice
GMP	Good Manufacturing Practice
GVHD	Graft versus host disease
HIV	Human immunodeficiency virus
HR	High risk
HRQOL	Health-related quality of life
HSCT	Haematopoietic stem cell transplantation
HTLV	Human T-cell lymphotropic virus
i.v.	Intravenous(ly)
IB	Investigator's Brochure
ICANS	Immune effector Cell-Associated Neurotoxicity Syndrome
ICE	Immune effector Cell-associated Encephalopathy
ICF	Informed Consent Form
ICH	International Council on Harmonisation
ICP	Intracranial Pressure
ICU	Intensive Care Unit
IDMC	Independent Data Monitoring Committee
IEC	Independent Ethics Committee
IFN	Interferon
Ig	Immunoglobulin
IHC	Immunohistochemical
IL	Interleukin
irAE	Immune-related adverse event
IRB	Institutional review board
KM	Kaplan-Meier
LAIP	Leukaemia-associated immunophenotype
LFS	Leukaemia-free survival
LVEF	Left ventricular ejection fraction
MAS	Macrophage activation syndrome
MRD	Minimal residual disease
MRI	Magnetic resonance imaging

<b>Abbreviation</b>	<b>Definition</b>
MTD	Maximum tolerated dose
MUGA	Multigated acquisition scan
NCI	National Cancer Institute
NIH	National Institutes of Health
OS	Overall survival
OX40	Tumour necrosis factor receptor superfamily 4 [TNFRSF4] and cluster of differentiation 134 [CD134]) endodomain
PBMCs	Peripheral blood mononuclear cells
PCR	Polymerase chain reaction
PedsQL	Pediatric Quality of Life Inventory
PFS	Progression-free survival
PIS	Patient Information Sheet
PTT	Partial thromboplastin time
qPCR	Quantitative polymerase chain reaction
RCR	Replication competent retrovirus
RFS	Relapse free survival
RP2D	Recommended Phase II dose
SAE	Serious adverse event
ScFv	Single chain variable fragment
SCT	Stem cell transplant
SEC	Safety Evaluation Committee
SR	Standard risk
SST	Serum separator tube
SUSAR	Suspected unexpected serious adverse reaction
TLS	Tumour lysis syndrome
TNF	Tumour necrosis factor
UK	United Kingdom
ULN	Upper limit of normal
UPENN	University of Pennsylvania
WBC	White blood cell





























## 2 OBJECTIVES AND ENDPOINTS

### 2.1 PRIMARY OBJECTIVES AND ENDPOINTS

Primary objectives and endpoints for Phase I and Phase II of the study are presented in [Table 2](#) and [Table 3](#), respectively.

**Table 2: Phase I Primary Objectives and Endpoints**

Objectives	Endpoints
To assess the overall safety and tolerability of AUTO3 administration.	Incidence of Grade 3-5 toxicity occurring within the dose limiting toxicity (DLT) period (30 days after last dose) of AUTO3 infusion.
To confirm and evaluate the RP2D and dosing schedule, and confirm maximum tolerated dose (MTD, if an MTD exists, of AUTO3 in both paediatric and adult patients.	Frequency of DLT of AUTO3.

DLT = dose limiting toxicity; MTD = maximum tolerated dose; RP2D = recommended Phase II dose.

**Table 3: Phase II Primary Objectives and Endpoints**

Objectives	Endpoints
To evaluate the anti-leukaemic effect of AUTO3 in paediatric and young adult patients (aged 1-24 years).	Proportion of patients achieving morphological remission (CR or complete response with incomplete recovery of counts [CRi]) and MRD-negative response in bone marrow (BM) within 30 (-3/+5) days post first AUTO3 infusion.  In patients with isolated CNS disease, anti-tumour effect will be assessed by whether patients clear their cerebrospinal fluid (CSF) of disease with ongoing BM CR, within 30 (±3) days post first AUTO3 infusion.

BM = bone marrow; CNS = central nervous system; CR = complete response; CRi = complete response with incomplete recovery of counts; CSF = cerebrospinal fluid; MRD = minimal residual disease.

## 2.2 SECONDARY OBJECTIVES AND ENDPOINTS

Secondary objectives and endpoints are presented in [Table 4](#).

**Table 4: Secondary Objectives and Endpoints**

Objectives	Endpoints
To assess the safety and tolerability of AUTO3	Frequency and severity of adverse events (AEs) and serious adverse events (SAEs)
	Incidence and duration of severe hypogammaglobulinaemia
To evaluate the feasibility of generating AUTO3	Proportion of patients for whom an AUTO3 product can be generated (feasibility)
To evaluate the clinical efficacy of AUTO3	Proportion of patients (all and prior CD19 CAR T treatment naïve) achieving morphological remission (CR or CRi) and/or MRD-negative complete response in bone marrow (BM) within 30 ( $\pm$ 3) days post first AUTO3 infusion by qPCR and/or flow cytometry. Relapse Free Survival (RFS), Event Free Survival (EFS), Progression-Free Survival (PFS) and Overall Survival (OS) following first treatment with AUTO3 Proportion of patients in molecular remission without further therapy at 6 months, 1 & 2 years following treatment with AUTO3 Incidence of CD19 and/or CD22-negative relapse
To determine the expansion and persistence of AUTO3 following adoptive transfer	Quantitative polymerase chain reaction (qPCR) and/or flow cytometry at a range of time points in the peripheral blood and BM
Duration of B-cell aplasia	Depletion of circulating B cells assessed by flow cytometry at a range of time points in the peripheral blood

AE = adverse event; CD = cluster of differentiation; CR = complete response; CRi = complete response with incomplete recovery of counts; MRD = minimal residual disease; OS = overall survival; PFS = progression-free survival; qPCR = quantitative polymerase chain reaction; SAE = serious adverse event.

## 2.3

[REDACTED]

- [REDACTED]
- [REDACTED]
- [REDACTED]
- [REDACTED]
- [REDACTED]
- [REDACTED]
- [REDACTED]

### 3 STUDY DESIGN

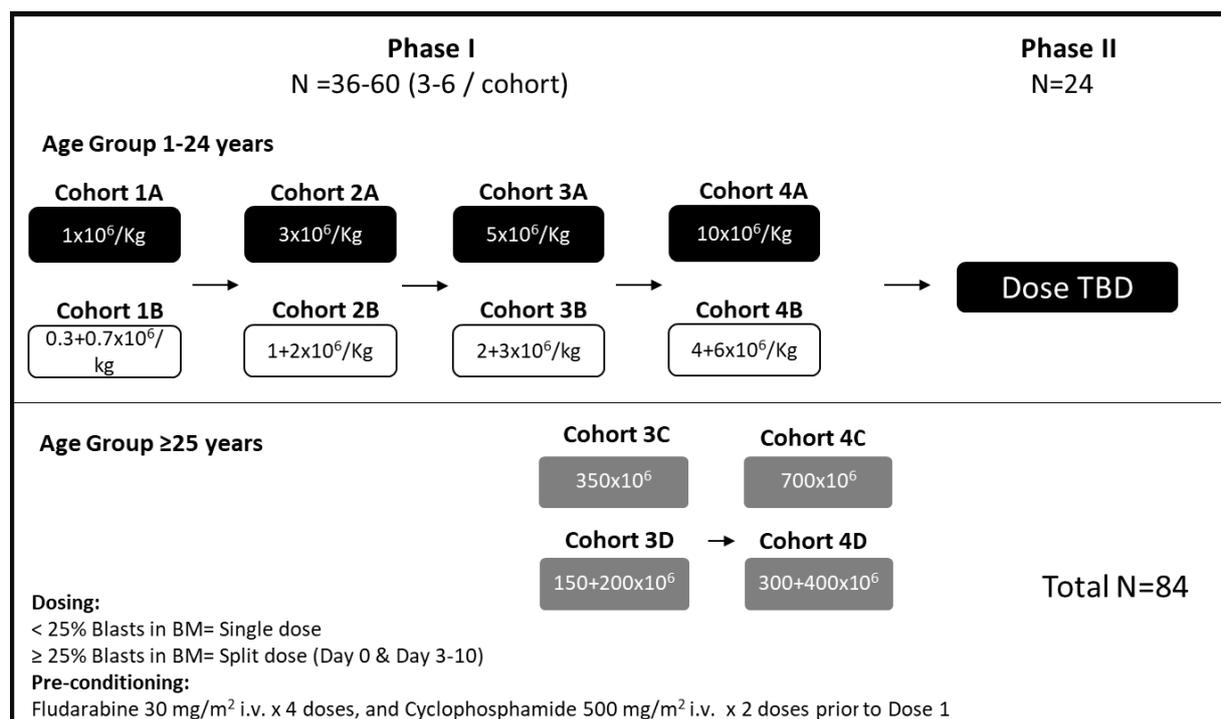
#### 3.1 STUDY OVERVIEW

This multi-centre, single-arm study will consist of two parts, a Phase I/dose escalation phase and a Phase II/expansion phase:

- **Phase I: Dose escalation**
  - To evaluate the optimal dose and schedule of AUTO3 in paediatric and young adults (age 1-24 years), starting at  $1.0 \times 10^6/\text{kg}$  CD19/CD22 CAR-positive T cells and then escalated to 3, 5 and  $10 \times 10^6/\text{kg}$  CD19/CD22 CAR-positive T cells as a single or split dose based on disease burden).
  - Adult ALL ( $\geq 25$  years) dose escalation may start at the highest paediatric/young adult dose declared safe. Dosing will be based on a fixed dose which is equivalent to a dose in a 70 kg patient.
- **Phase II: Dose expansion** to further assess safety and anti-leukaemic activity at the RP2D and dose schedule in paediatric and young adult patients only.

An overview of the study design is presented in [Figure 3](#) below.

**Figure 3: Dose Escalation and Dose Expansions Phases**



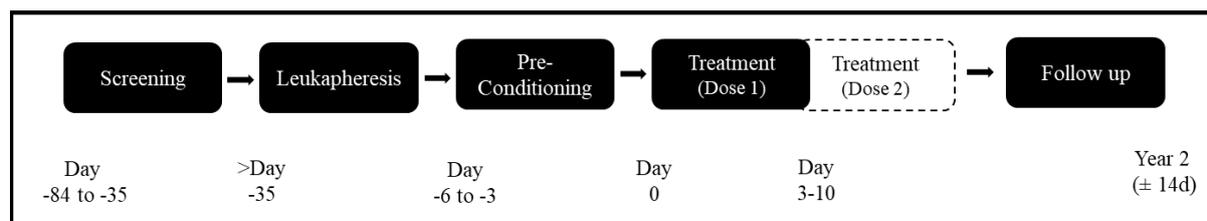
The study will consist of the following five stages:

- **Screening:** After providing written informed consent for study participation, all patients will be screened for study eligibility. Eligible patients will proceed to leukapheresis.
- **Leukapheresis:** Eligible patients will undergo leukapheresis to facilitate manufacture of AUTO3. If sufficient quantity of the cells (prescribed dose  $\pm 20\%$ ) are produced, the patient will proceed to the pre-conditioning phase.

- **Pre-conditioning:** If sufficient AUTO3 for the prescribed dose is successfully manufactured and the patients continue to meet eligibility requirements for the study, they will proceed to receive a prescribed lymphodepleting pre-conditioning treatment with CY and FLU before AUTO3 infusion.
- **Treatment:** AUTO3 for the prescribed dose will be administered i.v. as a single infusion on Day 0 or split into two, with the second split dose given between Day 3-10 in patients with higher blast count in the BM. The treatment phase will extend from Day 0 (infusion day) until the end of the DLT observation period, 30 days ( $\pm 3$  days) post last AUTO3 infusion. Phase I patients are expected to be admitted for a minimum of 30 days ( $\pm 3$  days), or longer if necessary for monitoring and management. The duration of admission of Phase II patients will be 30 days ( $\pm 3$  days) but may be reduced based on emerging data with a protocol amendment.
- **Follow-up:** The follow-up phase will begin after the treatment stage and end 24 months ( $\pm 14$  days) after first infusion with AUTO3 or at disease progression or withdrawal of consent, whichever ever happens first (End of Study visit performed for early withdrawal).

An overview of the five study stages is presented in [Figure 4](#).

**Figure 4: Overview of the Stages of the Study**



From signing of consent until the 24 month visit or the End of Study visit in the case of early withdrawals, information relating to AEs, laboratory abnormalities, biomarker changes, and anti-leukaemic activity will be collected according to the Schedule of Assessments.

All patients will be eligible for enrolment into a long-term follow-up protocol (AUTO-LT1) at the end of the study to be followed for safety evaluation and survival for 15 years from the first AUTO3 infusion or until death or withdrawal of consent, whichever ever happens first.

**Note:** Patients who have received AUTO3 and have discontinued or completed the study will continue to be monitored periodically for AUTO3 treatment-related SAEs, AEs of special interest and new malignancies until they enrol onto the long-term follow-up study (AUTO-LT1). Additionally, blood samples for CAR T persistence and replication competent retroviruses (RCRs) may also be collected every 3 months. In such cases, the End of Study visit may be delayed until enrolment onto the long-term follow-up study.

### 3.2 PHASE I DOSE ESCALATION

Phase I is designed to evaluate the optimum RP2D schedule of AUTO3 in paediatric and adult patients with B cell ALL. The paediatric/young adult (1-24 years) and adult ( $\geq 25$  years) cohorts will run in parallel. The paediatric/young adult group will start at a dose of  $1.0 \times 10^6/\text{kg}$  CD19/CD22 CAR-positive T cells. The adult patient ( $\geq 25$  years) cohort will open at or below the dose after at least 3 patients in the paediatric/young adult group have been treated at a dose level and no DLTs or more than one case of Grade 3 CRS or neurotoxicity has been observed.

Dose escalation will follow a rolling 6 design (Skolnik et al. 2008) (Table 5). Each cohort may treat up to six patients. Evaluation of a dose level with at least three patients treated at the planned dose level completing the DLT evaluation period is required prior to declaring the dose and schedule safe. If patients are treated below the planned dose level due to AUTO3 manufacturing or other reasons, then those patients will not be considered evaluable for making dose escalation decisions (additional patients will be enrolled to meet the minimum number needed to make the decision). Similarly if a patient receives a retreatment dose at the higher dose before its declared safe (lower dose considered sub-therapeutic) then that patient will not be evaluable for dose escalation decision making (will be considered evaluable for dose decision making if they develop a DLT with retreatment). However, dose escalation decisions will consider all available data, including biomarker data and the safety profile of all patients treated.

The trial is planned to comprise of up to 4 dose levels; eligible patients will be assigned to two subgroups based on disease burden concurrently. The inter-patient dosing interval for AUTO3 in Phase I will be at least **14 days**, for patients enrolled in a cohort and at least 7 days for patients enrolled to different cohorts until the dose schedule is declared safe, to allow for assessment of possible toxicity. Patients will be admitted to hospital (or ambulatory care setting) for a minimum of 30 days ( $\pm 3$  days), which can be extended as clinically indicated. The Safety Evaluation Committee (SEC) may increase the dosing interval based on emerging data. The DLT evaluation period will be **30 days ( $\pm 3$  days)** after the last dose of AUTO3.

The SEC will meet after the first patient in every cohort completes 14 days, to confirm continuation of enrolment to that cohort and thereafter meet again after the third or sixth patient in a cohort has completed the DLT assessment period.

In paediatric and young adult patients (aged 1-24 years), the planned starting dose level of  $1.0 \times 10^6$ /kg CD19/CD22 CAR-positive T cells will consist of two cohorts based on disease burden. Patients with  $<25\%$  blasts (cohort 1A) in the BM will receive the total dose of  $1.0 \times 10^6$ /kg CD19/CD22 CAR-positive T cells as a single dose, and those with  $\geq 25\%$  blasts (cohort 1B) in the BM will receive a split dose,  $0.3 \times 10^6$ /kg followed by  $0.7 \times 10^6$ /kg CD19/CD22 CAR-positive T cells (Table 6). The second split dose will be administered after 3-10 days and will be delayed or withheld if a patient experiences a severe toxicity with the first split dose (Table 6). If the emerging data indicates that the starting dose level and the next dose level ( $3 \times 10^6$ /kg) or a cohort in that dose has poor expansion, persistence or clinical response then SEC can recommend closure of that dose level or cohort after at least 2 patients (including cohorts A & B) have completed the DLT assessment period and can recommend escalation to the next dose level if no CAR related Grade 3 CRS or Grade 3 neurotoxicity has been noted. Dose level 2 is planned at  $3.0 \times 10^6$ /kg CD19/CD22 CAR-positive T cells. As with dose level 1 the dosing will be tailored to the disease burden. Patients with  $<25\%$  blasts (cohort 2A) will receive  $3.0 \times 10^6$ /kg CD19/CD22 CAR-positive T cells as a single dose and those with  $\geq 25\%$  blasts (cohort 2B) will receive  $1.0 \times 10^6$ /kg CD19/CD22 CAR-positive T cells followed by  $2.0 \times 10^6$ /kg CD19/CD22 CAR-positive T cells. Similarly, dose level 3 will evaluate a dose of  $5 \times 10^6$ /kg and dose level 4 will evaluate a dose of  $10 \times 10^6$ /kg CD19/CD22 CAR-positive T cells with the dosing schedule based on disease burden.

The adult patient ( $\geq 25$  years) cohort can open at  $350 \times 10^6$  CD19/CD22 CAR-positive T cell dose (fixed dose equivalent to  $5 \times 10^6$ /kg CD19/CD22 CAR-positive T cells in a 70 kg adult) if 3 patients have been safely treated at  $5 \times 10^6$ /kg CD19/CD22 CAR-positive T cells dose in

the paediatric/young adult group. The starting dose will be lower ( $210 \times 10^6$  CD19/CD22 CAR-positive T cells) if more than one case of Grade 3 CRS or neurotoxicity or a DLT has been observed in the paediatric group treated at  $5 \times 10^6/\text{kg}$  CD19/CD22 CAR-positive T cells. Patients will be dosed on a single or split dose schedule in the adult cohort (Schedules C & D) based on disease burden. Once initiated, dose escalation in the adult group will proceed independent of the paediatric/young adult group.

If the single dose schedule is found to be safe (Table 5), then that dose may be declared as a RP2D for an age group. If two or more DLTs are observed in the first 3-6 patients enrolled to cohort A then further enrolment to that cohort will stop (Table 5). However, patients with similar disease burden (<25% blasts) may be evaluated using the split dosing regimen used in cohort B if emerging data confirms that approach is likely to be safer. Similarly, if cohort B is considered safe, then it will also be declared a RP2D. Additionally, after cohort B is declared safe, if emerging data suggests that it may be appropriate to treat patients with this disease burden ( $\geq 25\%$  blasts) with a single dose, then they may be evaluated using the single dose regimen used in cohort A. Alternatively, if two or more DLTs are observed in the first 3-6 patients enrolled to cohort B then further enrolment to that cohort will stop. The SEC will consider totality of data before declaring a dose safe and as a RP2D or recommend further escalation or de-escalation. Dose escalation decision or RP2D for single and split dose can happen independent of the other cohort and Phase II can be opened for patients with similar disease burden. If a cohort eg 3A is declared as RP2D after dosing 6 patients then 3B can be declared as RP2D after 3 patients have been dosed and no DLTs observed. Alternatively, if 3 patients each are treated in cohort 3A and 3B then both cohorts can be declared as the Phase II dose. The decision for paediatric/young adults and older adult cohorts will be made independently.

If emerging data indicate that evaluation of a higher dose level is warranted, then a dose at or up to a total dose of  $10 \times 10^6/\text{kg}$  CD19/CD22 CAR-positive T cells may be opened, applying a similar dosing strategy based on disease burden as for the lower doses (Table 6). Adult patients ( $\geq 25$  years) will be treated at a fixed dose of up to  $700 \times 10^6$  CD19/CD22 CAR-positive T cells. Evaluation of a dose greater than  $10 \times 10^6/\text{kg}$  CD19/CD22 CAR-positive T cells will not happen without a protocol amendment.

**Table 5: Rolling 6 Dose Escalation Decision Rules**

Number of Patients with DLT at a Given Dose Level/Schedule	Escalation Decision Rules
0 out of 3	May declare dose safe and escalate (may continue to enrol additional patients for additional biomarker and safety data).
0 out of 6	Declare RP2D, may consider escalation to a higher dose level.
1 out of 3	Enrol 3 additional patients at the current dose level for a total of 6 patients.
1 out of 6	Declare RP2D, may consider escalation to a higher dose level.
2 or more patients in a dosing cohort (up to 6 patients)	The MTD has been exceeded, stop further enrolment. May evaluate a split dose or lower dose than the current dose.

DLT = dose limiting toxicity; MTD = maximum tolerated dose.

**Note:** The Investigators may override these guidelines if there are safety issues for which moving to a higher dose is not considered appropriate. Additionally, dose level 1 & 2 can be declared safe if there is poor expansion, persistence and efficacy after 2 patients (including cohort A & B) have been treated and no DLTs observed.

The dose decisions will be made by the SEC ([Section 13.1](#)) and the study could be stopped by the SEC or Independent Data Monitoring Committee (IDMC) upon occurrence of any of the events described in [Section 3.6](#).

### 3.3 PLANNED DOSE LEVELS

The planned dose schedules for Phase I are presented in [Table 6](#). These dose schedules were selected as they provide an optimum range for assessing safety, CAR T cell persistence and anti-tumour activity. This dose and dosing schedule is within the range assessed in other CAR T studies. Based on emerging data, intermediate dose levels may also be explored.

**Table 6: AUTO3 Treatment Cohorts & Schedules**

Dose Level	Treatment Cohorts	BM Blast %	Dosing Schedule Paediatric/Young Adults (1-24y) (Schedules A&B)		Dosing Schedule Adults (≥25y) (Schedules C&D)	
			Dose 1 Day 0	Dose 2 Day 3-10	Dose 1 Day 0	Dose 2 Day 3-10
Dose Level 1	Cohort 1A	<25% blasts	1.0 x 10 <sup>6</sup> /kg	None	n/a	n/a
	Cohort 1B	≥25% blasts	0.3 x 10 <sup>6</sup> /kg	0.7 x 10 <sup>6</sup> /kg	n/a	n/a
Dose Level 2	Cohort 2A	<25% blasts	3.0 x 10 <sup>6</sup> /kg	None	n/a	n/a
	Cohort 2B	≥25% blasts	1.0 x 10 <sup>6</sup> /kg	2.0 x 10 <sup>6</sup> /kg	n/a	n/a
Dose Level 3	Cohort 3A	<25% blasts	5.0 x 10 <sup>6</sup> /kg	None		
	Cohort 3B	≥25% blasts	2.0 x 10 <sup>6</sup> /kg	3.0 x 10 <sup>6</sup> /kg		
	Cohort 3C	<25% blasts			350 x 10 <sup>6</sup>	None
	Cohort 3D	≥25% blasts			150 x 10 <sup>6</sup>	200 x 10 <sup>6</sup>
Dose Level 4	Cohort 4A	<25% blasts	10.0 x 10 <sup>6</sup> /kg	None		
	Cohort 4B	≥25% blasts	4.0 x 10 <sup>6</sup> /kg	6.0 x 10 <sup>6</sup> /kg		
	Cohort 4C	<25% blasts			700 x 10 <sup>6</sup>	None
	Cohort 4D	≥25% blasts			300 x 10 <sup>6</sup>	400 x 10 <sup>6</sup>

Note: The adult starting dose will be lower (210 x 10<sup>6</sup> CD19/CD22 CAR-positive T cells) if a DLT has been observed in the paediatric group at 5 x 10<sup>6</sup>/kg CD19/CD22 CAR-positive T cells.

- Paediatric and young adult cohorts will enrol patients 1-24 years in Cohorts A & B and adult patients ≥25 years will be enrolled into adult Cohorts C & D.
- A patient may be eligible for a dose level if the dose is within 20% of the prescribed dose (±20%).
- Cohort will be decided based upon the results of the Day -7 BM blast count (can be local results) or the last blast count available during the screening period.
- On occasion, AUTO3 production may fail to generate sufficient cells for the current dose level. In this case, the patient can be treated on study but at a lower dose, however if a production fails to generate <0.2 x 10<sup>6</sup>/kg CD19/CD22 CAR-positive T cells (near the lowest known active dose of CD19 CAR-positive T cells, of 1.46 x 10<sup>5</sup> cells/kg, corresponding to approximately 10 x 10<sup>6</sup> total dose of CAR T-cells ([Porter et al. 2011](#)), then the patient will not be treated on study. Only patients treated at the planned dose

level will be evaluable for dose escalation decision making and primary efficacy analysis.

- In each dose level, the two sub-cohorts will be opened concurrently.
- In patients receiving a split dose the decision to administer the 2<sup>nd</sup> split dose will be made by the site PI or delegated individual in consultation with the Sponsor's Medical Monitor. Patients will be monitored for early onset of high grade fever that is suspected to be CAR related and evaluated for toxicity during the 3 days after infusion of the initial dose. If no  $\geq$ Grade 3 CRS or  $\geq$ Grade 2 neurotoxicity has occurred, the 2<sup>nd</sup> dose may be administered 3-10 days after the first if CRS has resolved to  $\leq$ Grade 1 and neurotoxicity has completely resolved. Dosing of the 2<sup>nd</sup> split dose can be further delayed (beyond 10 days) if clinically indicated. **Patients who develop AUTO3 related  $\geq$ Grade 3 CRS,  $\geq$ Grade 2 neurotoxicity or a DLT following the first split dose will not receive the second split dose.**
- Based on emerging data, patients with  $\geq$ 25% blasts in the BM may be treated with a single dose if both single and split doses are determined to be safe.
- Dose calculation should be made based on actual body weight, unless the patient is obese, in which case the dose calculation may be made based on ideal body weight for the age and height. Adult patients ( $\geq$ 25 years) will receive a fixed dose calculated based on a 70 kg adult weight.

### 3.4 PHASE II DOSE EXPANSION

Once the RP2D (for a disease burden) is determined for paediatric and young adult patients, the Phase II dose expansion part of the study will open. Individual patients may get a single or split infusion of the dose based on the disease burden. This part will treat up to 24 patients.

### 3.5 DOSE LIMITING TOXICITY

Toxicities will be graded for severity per the NCI Common Terminology Criteria for Adverse Events (CTCAE), version 5.0 and the CRS will be graded using Lee Criteria ([Lee et al. 2018](#)). The DLT evaluation period will be **30 days** ( $\pm$ 3 days) after the last dose of AUTO3.

**Dose limiting toxicity will be defined as:**

- Any new non-haematological AE of Grade 3 or higher toxicity using the NCI CTCAE (version 5.0), which is probably or definitely related to AUTO3 therapy, which occurs within the DLT evaluation period, and which fails to resolve to Grade 2 or better within 14 days, despite appropriate supportive measures.
- A Grade 4 CRS or neurotoxicity, cerebral oedema, or Grade 3 neurotoxicity (including cerebral oedema) that lasts  $>$ 72 hours.
- Grade  $>$ 3 disseminated intravascular coagulation (DIC).
- Grade  $>$ 2 infusion reaction.
- Any other fatal event (Grade 5) or life-threatening event (Grade 4) that cannot be managed with conventional supportive measures or which in the opinion of the SEC necessitates dose reduction or other modification to trial treatment to avoid a similar hazard in future patients.

- Effort should be made to perform an autopsy in the case of a fatal event where the aetiology is unclear.

### **Reporting Requirements for DLT**

All DLTs must be reported to the Sponsor as SAEs within 24 hours of site staff becoming aware of them (see [Section 12.3.2](#)). All DLTs will be notified to the SEC and IDMC by the Sponsor.

**Maximum Tolerated Dose:** The MTD is defined as the highest dose level of AUTO3 at which  $\leq$ one patient out of six patients experience a DLT during the DLT evaluation period. If two or more out of six patients at a dose level experience a DLT during the DLT evaluation period, the MTD has been exceeded. If the MTD is exceeded due to a specific toxicity that can be managed with supportive care, an additional three patients may be enrolled at the dose level that exceeded the MTD with establishment of supportive care measures after review and approval by the IDMC. A summary of available safety data and a description of the plans for supportive care measures with further enrolment at that dose level will be provided (as a notification) to Independent Ethics Committees/Institutional Review Boards (IECs/IRBs) prior to dosing.

**Recommended Phase II Dose:** The RP2D is either identical to the MTD or maximum administered dose or a lower dose level of AUTO3, selected based on a cumulative review of safety, persistence of the CAR T cells and clinical activity. Recommended Phase II dose schedules may be defined based on disease burden. A minimum of 6 patients need to be treated in a cohort before it can be declared as the Phase II dose. For example, if 6 patients are treated in cohort 3A then it can be declared as the RP2D for patients with lower disease burden. Alternatively, if 3 patients each are treated in cohort 3A and 3B then both cohorts/the dose level can be declared as the Phase II dose.

### **3.6 SAFETY STOPPING CRITERIA FOR THE CLINICAL TRIAL**

The study can be stopped by the SEC or IDMC upon occurrence of any of the following events:

- Unexpected and related SAEs that exposed patients participating in the study to unacceptable risk of harm. (*Class related toxicities such as CRS and neurotoxicities will not automatically result in stopping unless they are exposing patients to risk above what is expected for similar therapies*).
- Uncontrolled SAEs related to identified risks.
- The occurrence of Grade 4 non-haematological toxicity in 3 patients. Unless in the opinion of the IDMC (after review), it is likely manageable with the institution of appropriate supportive care.
- Death of a patient at any time after therapy that is definitely related to T cell therapy.
- The occurrence of a second malignancy at any point after therapy that is definitely related to the T cell therapy.

The study may be restarted after appropriate preventive or management guidelines have been instituted and a substantial protocol amendment has been approved by the regulatory authorities and ethics committees.

### 3.7 STUDY DURATION

The total study duration is estimated to be 5 years from first patient enrolled to the last patient, last visit (24-month visit). The end of the trial will be 24 months after the last patient has received a first AUTO3 infusion (or earlier if appropriate). Long-term follow up of patients for up to 15 years after AUTO3 treatment is described in [Section 8.5](#).

### 3.8 NUMBER OF PATIENTS

It is anticipated that approximately 100 patients will be enrolled (consented) into the study with up to 84 patients receiving the treatment as outlined below. The difference between the number of enrolled and treated patients accounts for manufacturing failures and inability of some patients to meet AUTO3 infusion criteria.

- **Phase I:** A dose escalation involving up to a total of 36-60 treated paediatric and adult patients (up to 3-6 patients per dose cohort), which will consist of:
  - 24-36 patients in the paediatric / young adult patient cohorts (age 1-24 years)
  - 12-24 patients in the adult patient cohorts ( $\geq 25$  years)
  - *Additional number accounts for the possibility of patient with higher disease burden being evaluated using a single dose*
- **Phase II:** Dose expansion involving up to a total of 24 treated patients in paediatric/young adults (aged 1-24 years).

Phase II of the study will follow Simon's 2-stage optimal design as described in [Section 14.1](#).

## 4 SCIENTIFIC RATIONALE FOR STUDY

### 4.1 DESIGN RATIONALE

This single-arm dose escalation and expansion Phase I/II study, is designed to assess safety, tolerability, and optimum RP2D schedule and MTD, if a MTD exists. Considering the experience the field has with targeting both CD19 and CD22 individually and the novelty of targeting CD19 and CD22 with a combined CAR, a starting dose of  $1.0 \times 10^6/\text{kg}$  CD19/CD22 CAR-positive T cells encompassing splitting of dose based on disease burden is considered appropriate. Additionally, escalation to a higher dose of 3, 5 and then  $10 \times 10^6/\text{kg}$  CD19/CD22 CAR-positive T cells is incorporated as other CD19 41BB- $\zeta$  CARs in a similar patient population have indicated tolerability of these doses ( $2.0\text{-}5.0 \times 10^6$  CAR T cells/kg for  $\leq 50$  kg and  $1.0\text{-}2.5 \times 10^8$  cells for  $>50$  kg) (Lee et al. 2014, Maude et al. 2014b) and our initial data indicate improved CAR T cell expansion/persistence at doses of  $3 \times 10^6/\text{kg}$  and above. Adult patients ( $\geq 25$  yrs) will be treated at a fixed dose (as per Schedule C and D) starting at  $350 \times 10^6$  CD19/CD22 CAR-positive T cells if an equivalent dose has been administered to 3 pediatric/young adult patients and found to be safe; if not, treatment will start at a lower dose. The subsequent dose will be  $700 \times 10^6$  CD19/CD22 CAR-positive T cells. This starting dose is in the range being evaluated with other CAR T cells in adult ALL, such as tisagenlecleucel (CTL019, Kymriah) of  $100\text{-}500 \times 10^6$  CAR T cells which was dosed with a split dose schedule (Frey et al. 2016), escalation to higher doses will be based on emerging data from lower dose cohort.

The rationale for our dose level and dosing schedule is outlined in detail in Sections 4.4 and 4.5. In Phase I of the study, a starting dose level ( $1.0 \times 10^6/\text{kg}$  CD19/CD22 CAR-positive T cells/kg), which is similar to the MTD of autologous CD19 CAR T cells in the National Institutes of Health (NIH) study in paediatric ALL, will be evaluated (Lee et al. 2015). Based on emerging data this dose could be declared RP2D or the dose further escalated as described above.

During Phase I of the study, a minimum interpatient dosing interval of 2 weeks within a cohort and 1 week between cohorts is incorporated to reduce the risk of inducing severe adverse effects in more than one patient until a dose schedule is declared safe. Following evaluation of the safety of AUTO3 and determination of the RP2D, safety and efficacy will be further evaluated in a Phase II expansion cohort. There is no suggested interval between patients in the Phase II cohort as we anticipate to have generated safety data in at least 15-18 patients by then, including a minimum of six patients treated at the Phase II dose/schedule.

The number of patients in Phase I of the study is designed for dosing an adequate number of patients prior to evaluation of safety and biological activity such as serum cytokine levels and CAR T cell persistence and to facilitate better decision making. Phase II of the study is based on a Simon's 2-stage optimal design; the rationale for dosing up to 24 patients is to detect early signs of efficacy in addition to generating additional safety data at the RP2D.

### 4.2 STUDY POPULATION RATIONALE

AUTO3 is designed to specifically bind to CD19 and or CD22, which are only expressed on normal and malignant B lymphocytes. The study patient population for this study is paediatric and adult patients with B lineage ALL.

An age range of 1-24 years has been set for the paediatric and young adult patients because patients in this age range are treated with frontline and relapse protocols that differ from older patients with this disease. In addition, since this is an experimental therapy, the study patient population will be restricted to patients with HR relapsed or refractory B lineage ALL who are predicted to have a poor outcome even with intensified chemotherapy and SCT. As outlined in [Section 1.1](#), the long-term survival of paediatric/young adult patients with HR relapse in large UK and European studies is 20-30%. Similarly, the outcomes of patients relapsing post allogeneic SCT are also very poor. Long-term survival rates are of about 10-20% in the population receiving a second allogeneic SCT (but with considerably increased transplant-related morbidity and mortality), and negligible in those unable to proceed to a second transplant ([Borgmann et al. 2003](#), [Roy et al. 2005](#), [Saarinen-Pihkala et al. 2006](#)). These patients are appropriate candidates for clinical trials and data from ongoing trials using second-generation CD19 CAR T cells show unprecedented outcomes in patients with relapsed or refractory paediatric ALL (see [Section 1.3](#)). Since the recently approved CD19 CAR therapy is not yet widely accessible, enrolling patients on clinical trials with a CAR therapy that is designed to prevent CD19 negative relapse is appropriate.

The prognosis of adult ALL patients is worse than for paediatric/young adult patients and has not changed significantly during the last two to three decades, with long-term remission rates limited to 30–40%. Approximately 50% of all adult ALL patients will relapse, and UKALL12 data shows that 5-year OS in adults who relapse following standard multi-agent chemotherapy is 7%. Significant number of patients treated with CD19 or CD22 targeted therapy such as blinatumomab and inotuzumab ozogamicin relapse, these therapies are largely utilized as a bridge to allo-HSCT. A standalone targeted therapy is in need and CAR T cell therapy is likely to have significant therapeutic potential in these patients as a standalone therapy ([Wierda et al. 2018](#)). Considering the emerging safety and tolerability profile of AUTO3 ([Section 1.6](#)) it is appropriate to evaluate AUTO3 in these patients.

Dual targeting of CD19 and CD22 is likely to improve outcomes further by preventing antigen negative escape and immune mediated rejection of CAR T cells.

#### **4.3 CYCLOPHOSPHAMIDE AND FLUDARABINE PRE-CONDITIONING RATIONALE**

Pre-conditioning strategies that deplete host lymphocytes have been shown to enhance clinical responses to some adoptive T cell therapies ([Muranski et al. 2006](#), [Spear et al. 2013](#)). Lymphodepletion prior to adoptive transfer of tumour specific T-lymphocytes is thought to enhance treatment efficacy by eliminating regulatory T cells and increasing access of the transfused CAR T cells to activating cytokines ([Klebanoff et al. 2005](#), [Wrzesinski and Restifo 2005](#)). Cyclophosphamide (CY) has an established history in lymphodepleting regimens used prior to adoptive cell immunotherapy ([Sporn et al. 1993](#), [Curti et al. 1998](#), [Brentjens et al. 2011](#), [Chu et al. 2012](#)). It is used alone or often used in combination with other agents ([Dudley et al. 2008](#), [Laurent et al. 2010](#), [Geller et al. 2011](#)). Fludarabine (FLU), either as a single agent or in combination with other cytotoxic agents, has been used as a lymphodepleting preparative regimen to reduce transplant-related toxicities and allow SCT in elderly and medically infirm patients, as well as in adoptive T cell therapies, including CAR T cell therapy ([Louis et al. 2011](#)). Fludarabine is also commonly used in combination with CY in the treatment of patients with chronic lymphocytic leukaemia ([Hallek 2013](#)) where it is well tolerated.

Cyclophosphamide and FLU based pre-conditioning have become the preferred regimen for CAR T cell therapies and have been used in multiple studies ([Kochenderfer et al. 2015](#), [Lee et al. 2015](#), [Ali et al. 2016](#)). The combination is also considered to be superior to CY alone. In a CD19 CAR T cell study (JCAR014) in adult patients with ALL, those who received FLU-CY had a significantly greater expansion of CAR T cells at Day 28 and a lower rate of relapse ([Turtle et al. 2016](#)). Most relapses in the CY alone group were due to immune rejection of CAR T cells and it is likely that the addition of FLU to lymphodepletion may reduce the risk of this. Similarly, in the NIH study of CD19 CAR T cells in paediatric ALL, use of a FLU-CY regimen correlated with a higher response rate (29/44 patients, 66%) and LFS (53% at 18 months) than alternative regimens (2/8 patients, 25% responders  $P < 0.05$  and LFS of 0% at 18 months) ([Lee et al. 2016b](#)). We believe these data establish FLU-CY as the standard of care for lymphodepletion prior to CAR T cell therapy for ALL.

The exact dose regimens used for lymphodepletion prior to CAR T cell therapy are variable. However, since the purpose of pre-conditioning chemotherapy is lymphodepletion and prolonged myelosuppression (particularly in post-transplant patients) is undesirable, higher doses of CY (2 to 4 g/m<sup>2</sup> for 1 to 2 days) are probably unnecessary, especially when given with FLU. The regimen utilised in this study (FLU 30 mg/m<sup>2</sup> for 4 days + CY 500 mg/m<sup>2</sup> for 2 days) is similar to that evaluated in studies with tisagenlecleucel (Kymriah) in multiple clinical studies including the multi-centre ELIANA study and appears both safe and active and is part of the Food and Drug Administration (FDA) and European Medicines Agency approved label ([Lee et al. 2015](#), [Ali et al. 2016](#), [Fry 2016](#), [Kochenderfer et al. 2016](#), [Maude et al. 2016b](#), [Neelapu et al. 2016](#), [Turtle et al. 2016](#), [FDA 2018a](#)).

Cyclophosphamide can cause haemorrhagic cystitis and cardiotoxicity but these are extremely rare at the proposed dose. The likely side effects of CY at this dosage are transient nausea and cytopenia. Patients will be given anti-emetics, prophylactic anti-microbials and transfusion support as per standard institutional policy. Fludarabine is generally well tolerated: the most common side effects are lymphopenia and infection. Neurotoxicity can occur but generally at higher doses, and at the dose proposed, neurotoxicity is rarely seen. However, severe neurotoxicity leading to death was reported recently in the JCAR015 ROCKET trial ([NCT02535364 2016](#)) that used a FLU based conditioning regimen. On 07 July 2016, the FDA suspended recruitment into the ROCKET trial following the deaths of three young adult patients with ALL due to cerebral oedema, although no detail of the neurological toxicity has yet been published. The three deaths occurred amongst approximately seven patients treated with FLU-CY conditioning prior to JCAR15 treatment, whereas this had not previously been seen with CY alone. The investigators subsequently re-opened the trial with CY alone as lymphodepletion. This study was placed on hold again on 23 November 2016 due to two further deaths because of cerebral oedema which happened in the absence of FLU based conditioning, suggesting that FLU was not primarily responsible for the observed neurotoxicity. Observations suggest that the severe neurotoxicity observed in that study may reflect the use of a relatively high dose of CD19 CAR T cells with a CD28 costimulatory domain which appears to cause stronger early proliferation and cytokine secretion compared to CD19 CAR T cells with 41BB- $\zeta$  costimulatory domains.

.In all the studies conducted at the University of Pennsylvania (UPENN) in the last 3 years, most of which utilised FLU-CY lymphodepletion and a CD19-41BB- $\zeta$  CAR (CTL019), only one neurotoxicity related death has been reported. In a larger multi-centric Phase II study, the same CAR was used (CTL019) in paediatric patients using the similar dose FLU-CY condition regimen to that proposed in this study, of the 75 patients (median age 11 years,

61% with prior HSCT) infused with a single dose of CTL019 at a median weight-adjusted dose of  $3.1 \times 10^6$  transduced viable T cells per kilogram of body weight (range,  $0.2 \times 10^6$  to  $5.4 \times 10^6$  cells per kilogram), 13% of patients experienced transient Grade 3 or 4 neurotoxicity including confusion, delirium, encephalopathy, agitation and seizure but no cerebral oedema or deaths due to neurotoxicity was reported (Maude et al. 2018).

Neurotoxicity is the most challenging clinical problem associated with CD19 CAR since it does not easily correlate with T cell dose, underlying disease or the neurological involvement of leukaemia/lymphoma or CRS. However, it is clear that this is not a direct effect of FLU. Data from multiple studies also suggest that a lower dose/intensity of pre-conditioning (Grupp et al. 2016, Kochenderfer et al. 2016, Neelapu et al. 2016), with CY (300 to 500 mg/m<sup>2</sup> for 2-3 days or 900 mg/m<sup>2</sup> for 1 day) and FLU (30 mg/m<sup>2</sup> for 3-4 days), compared to the ROCKET study (1 to 3 mg/m<sup>2</sup> CY + 25 to 30 mg/m<sup>2</sup> of FLU for 5 days which is repeated 2 to 4 weeks later) is safe and appropriate.

In the current study, we propose to use FLU-CY (FLU [30 mg/m<sup>2</sup> for 4 days] and CY [500 mg/m<sup>2</sup> for 2 days]) based pre-conditioning regimen. A 3±1 day washout period prior to infusion of AUTO3 is also incorporated, taking into consideration the 20-hour half-life of FLU and the need to infuse early to maximize exposure to the transient IL-15 surge following lymphodepletion (Rossi et al. 2015). This would result in elimination of over 3 half-lives of FLU and will potentially decrease any risk of cumulative toxicity with AUTO3. Given the nature of our patient cohort being treated and the data cited above demonstrating that lymphodepletion with FLU-CY enhances the clinical efficacy of CAR T cells, we believe the proposed dose and regimen is justified.

#### 4.4 STARTING DOSE RATIONALE

The primary consideration for selection of a starting dose of  $1.0 \times 10^6$ /kg CD19/CD22 CAR-positive T cells for the proposed trial in the paediatric/young adult population is the safety of the patient. Considerations for CAR T cell dosing are challenging. Chimeric antigen receptor T cells are a “living therapy” i.e. they can engraft within the study subject, they can expand and they have no half-life in the standard conception of the term (Ghorashian et al. 2015). Efficacy and toxicity appears related to the magnitude and tempo of engraftment (Maude et al. 2014b). A wide range of doses have been utilised in existing studies of CD19 CAR T cell therapy ( $5 \times 10^5$ - $1.7 \times 10^7$  CAR T cells/kg), with no clear correlation between cell dose administered and efficacy, reflecting the fact that infused CAR T cells can expand by several logs after transfer to a lymphodepleted host. Consequently, the correlation between CAR T cell dose and toxicity is much less clear than the correlation with standard small molecule or protein based therapeutics.

In the only dose-finding study of CD19 CAR T cells, a Phase I dose escalation study CD19 of CAR T cells transduced with a second-generation FMC63-derived CAR containing a CD28 co-stimulatory endodomain, a dose of  $1 \times 10^6$ /kg was defined as the MTD. Four patients were treated at a dose of  $1 \times 10^6$ /kg and none developed DLT. The subsequent four patients were then treated with  $3 \times 10^6$ /kg CAR T cells, but two of these four then developed DLT (Grade 4 and 3 CRS). These findings are obscured by differences in CAR design and CAR T cell production processes. For example, in a study in children and young adults treated with a CD19 41BB-ζ CAR, a higher dose ( $2.0$  to  $5.0 \times 10^6$  cells/kg for  $\leq 50$  kg and  $1.0$  to  $2.5 \times 10^8$  cells for  $>50$  kg) was considered tolerable (Maude et al. 2014b, Lee et al. 2015). However, a dose in the order of millions of CAR T cells/kg seems generally optimal.

The starting dose for adult ALL patients takes into consideration the current dose and safety data from the paediatric and young adult patients ([Section 1.6](#)). The adult dose escalation may start at a dose equivalent to the highest paediatric dose considered safe after at least 3 patients in that age group have been treated at a dose level and no DLTs or more than one case of Grade 3 CRS or neurotoxicity has been observed. Adult dosing will be based on a fixed dose which is equivalent to a dose in a 70 kg person, e.g. if  $5 \times 10^6/\text{kg}$  CD19/CD22 CAR-positive T cells dose is considered safe in paediatric patients then the adult starting dose ( $\geq 25$  years) will be  $350 \times 10^6$  CD19/CD22 CAR-positive T cells and the subsequent dose will be  $700 \times 10^6$  CD19/CD22 CAR-positive T cells. Patients will be dosed with a single or split dose based on disease burden as in the paediatric patients. Since the starting dose is based on the emerging data of AUTO3 in paediatric and young adult patients it is likely to be safe in the older adults that meet the study eligibility criteria. Additionally, this dose is in the range being evaluated with other CAR T cells therapies in adult ALL, such as tisagenlecleucel (CTL019, Kymriah) of  $500 \times 10^6$  CAR T cells ([Frey and Porter 2016](#)).

Dual targeting of CD19 and CD22 is not anticipated to result in more activity or toxicity over a CD19 or a CD22 CAR alone for several reasons: CD22 is significantly less densely expressed than CD19 on ALL blasts so the cumulative targetable antigen density will not be significantly increased ([Lee et al. 2015](#), [Shah et al. 2015](#)). Further, little if any synergy is anticipated from signalling through both TNF-family receptor endodomains, as a signal through either OX40 or 41BB- $\zeta$  is likely to saturate the common signalling pathway. In the absence of a CD28 co-stimulatory domain less rapid proliferation is expected. Furthermore, no increases in cytotoxicity are observed between CD19 and CD19/CD22 CAR constructs with CD19+CD22+ target cells. Only in situations where the targets lose expression of CD19 do we expect enhanced killing with the CD19/CD22 CAR.

Other CD19 CAR studies, such as the CARPALL study in paediatric and young adult ALL ([NCT02443831 2016](#)), have used a fixed starting dose of  $1 \times 10^6$  CAR T cells/kg and found it to be safe with no Grade 3 CRS in the 14 patients treated (P. Amrolia, personal communication). Recent experience with the NCI's anti-CD22 41BB- $\zeta$  CAR also indicated a tolerable safety profile, no Grade 3 CRS or neurotoxicity were noted in the study that treated 16 children and young adult patients at doses up to  $3 \times 10^6$  CAR T cells/kg ([Shah et al. 2016](#)).

Considering the experience in the field with CD19 and CD22 targeted CARs, for this ALL study, the proposed starting dose of  $1 \times 10^6$  CD19/CD22 CAR-positive T cells/kg, in paediatric and young adult patients, administered as a single or split dose based on disease burden (which is expected to add to the safety) is justified. The proposed starting dose will consist of two sub-cohorts, patients with  $<25\%$  blasts in the BM will receive the total  $1.0 \times 10^6/\text{kg}$  CD19/CD22 CAR-positive T cells dose as a single dose and those with  $\geq 25\%$  blasts in the BM will receive a split dose,  $0.3 \times 10^6/\text{kg}$  followed by  $0.7 \times 10^6/\text{kg}$  CD19/CD22 CAR-positive T cells. The second split dose will be administered after 3-10 days and will be delayed or withheld if any patient experiences severe toxicity with the first split dose. In adult ALL, other CD19 CAR studies have demonstrated that the recommended dose of CD19 CAR T cells in adults is approximately twice that of paediatric patients. For example, the dose of axicabtagene ciloleucel (Yescarta) in paediatric patients is  $0.5 \times 10^6$  CAR T cells/kg and in adults it is  $1 \times 10^6$  CAR T cells/kg ([Wierda et al. 2018](#)); the dose of tisagenlecleucel (Kymriah) in paediatric patients is 2 to  $5 \times 10^6$  cells/kg for  $\leq 50$  kg and 1 to  $2.5 \times 10^8$  CAR T cells for  $>50$  kg ([Maude et al. 2014b](#), [Lee et al. 2015](#)), and in adults it is  $500 \times 10^6$  CAR T cells ([Frey and Porter 2016](#)). Therefore, the proposed starting dose in adult ALL based on

emerging data from AUTO3 in paediatric and young adult patients is likely to be safe and appropriate.

#### 4.5 DOSING SCHEDULE RATIONALE

Chimeric antigen receptor T cell therapies are generally administered once, undergo significant expansion *in vivo* upon contact with the target antigen expressed on tumour cells and, particularly where a 41BB- $\zeta$  costimulatory domain is incorporated into the CAR, persist long-term in a proportion of patients (Maude et al. 2014b). It is anticipated that AUTO3 will have similar expansion and persistence *in vivo* to the CD19 CAR T cells utilised in the UPENN studies, rendering the need for re-dosing unnecessary. On occasion, a re-treatment dose may be given if the patient meets the criteria for re-treatment (Section 8.4.7).

There is now strong data from a number of different CD19 CAR T cell trials in ALL indicating that higher disease burden is predictive of more severe CRS (Davila et al. 2014, Lee et al. 2015). This has led some groups to mitigate this toxicity by either administering a lower dose of CAR T cells to patients with higher disease burden (Lee et al. 2015, Turtle et al. 2016), or have tried splitting the total dose.

Data presented at the American Society of Clinical Oncology 2016 meeting (Frey and Porter 2016), provide support for a split dosing schedule in patients with higher leukaemic burden. In that study in adult patients with ALL there was a high incidence (three out of six patients) of fatal, refractory CRS when patients were treated with a single dose of  $5 \times 10^8$ /kg CD19 CAR T cells. However, when the dose was split over 3 days (10%, 30% and 60%), no deaths occurred and nine out of 12 patients had manageable Grade 3 to 4 CRS with an objective response rate of 83% (9 of 12 patients reached CR; 1 of 12 patients reached PR). Split dosing gave an opportunity to hold or delay the subsequent dose when signs of early CRS were observed. Fractionation may reduce the extent and rate of expansion, and thereby the severity of CRS.

To optimise safety and overcome the problem of T cell exhaustion in patients with high leukaemic burden, we propose to combine these two approaches. Patients treated at the planned starting dose of  $1.0 \times 10^6$ /kg CD19/CD22 CAR-positive T cells, will be stratified per leukaemia burden. Patients with <25% BM blasts (M1, M2 disease) at the start of lymphodepletion will receive the full dose of  $1.0 \times 10^6$ /kg CD19/CD22 CAR-positive T cells as a single dose. In those with  $\geq 25\%$  BM blasts (M3 disease), AUTO3 will be administered as a split dose  $0.3 \times 10^6$ /kg at Day 0 followed 3-10 days with a second dose of  $0.7 \times 10^6$ /kg CD19/CD22 CAR-positive T cells. This approach allows for safer administration of doses that are more likely to be effective, the idea being that the initial lower dose “debulks” the tumour load allowing for safer administration of a subsequent dose. Similarly if the dose is escalated to the higher dose levels of 3, 5 or  $10 \times 10^6$ /kg CD19/CD22 CAR-positive T cells, the dose will be similarly split (Table 6).

Additionally, cytokine surge, especially the IL-15 surge, observed during post-conditioning chemotherapy with FLU-CY happens in the first few days and returns rapidly to baseline around 10 days later (Rossi et al. 2015). The experience in the first few patients in this study also indicates that significant IL-15 surge lasts only for a few days and returns to baseline in 10-14 days similar to what has been reported in literature (Rossi et al. 2015). It is important to dose the CAR T cells early within this window as IL-15 is necessary for CAR T cell expansion, engraftment and resulting efficacy (Xu et al. 2014). Moreover, availability of antigen (tumour cells) at the time of administration of the second split dose is also necessary for expansion and engraftment. The flexibility in timing the second split dose i.e. 3-10 days,

will allow the use of available biomarker data and clinical judgement in administering the second dose after the peak signs and symptoms related to CRS have resolved or hold for severe toxicity.

A recent analysis of 133 patients with relapsed or refractory B cell malignancies (B-ALL, n=47; NHL, n=62; CLL, n=24) indicated that early onset of high grade fever  $\geq 38.9^{\circ}\text{C}$  within the first 36 hours after CAR-T cell infusion was noted in all patients who subsequently developed Grade  $\geq 4$  CRS (Hay et al., 2017). Fever also occurred earlier after CART cell infusion in patients who subsequently developed Grade  $\geq 3$  neurotoxicity compared with those who developed Grade 1–2 neurotoxicity ( $P = 0.0007$ ). Fever  $\geq 38.9^{\circ}\text{C}$  occurring within 36 hours of CAR T cell infusion had a 100% sensitivity for subsequent Grade  $\geq 4$  neurotoxicity, the specificity was only 82%, in part due to other causes of fever in these patients with chemotherapy-induced neutropenia (Gust et al., 2017). In a paediatric ALL study, the median start of CRS was 4 days (range: 1-7 days) following CD19 CAR T cell infusion and lasted a mean of 4.8 days (range: 1-9 days) (Lee et al. 2015) and the median time to onset of Grade  $\geq 1$  neurologic AEs was 4 days after CAR T cell infusion (Gust et al., 2017). The anticipated time to peak expansion of the infused CAR T cells is between 7 and 14 days (Davila et al. 2014). The safety and the cellular kinetic data from ongoing clinical trials (Section 1.6) also support the early dosing of the 2<sup>nd</sup> split dose by Day 3 similar to the data from the UPENN study in adult ALL patients (Frey and Porter 2016).

Based on these data, in patients receiving a split dose the decision to administer the 2<sup>nd</sup> split dose will be made by the site PI or delegated individual in consultation with the Sponsor's Medical Monitor. Patients will be monitored for early onset of high grade fever that is suspected to be CAR related and evaluated for toxicity during the 3 days after infusion of the initial dose. If no  $\geq$ Grade 3 CRS or  $\geq$ Grade 2 neurotoxicity has occurred, the 2<sup>nd</sup> dose may be administered 3-10 days after the first dose if CRS has resolved to  $\leq$ Grade 1 and neurotoxicity has completely resolved. The dosing of 2<sup>nd</sup> split dose can be further delayed (beyond 10 days) if clinically indicated. **Patients who develop AUTO3 related  $\geq$ Grade 3 CRS,  $\geq$ Grade 2 neurotoxicity or a DLT following the first split dose will not receive the second split dose.**

After treatment with AUTO3, safety will be monitored closely (both in hospital and as an outpatient) and efficacy will be assessed periodically as described in the Schedule of Assessments. The timing of assessments is designed to capture early signs of toxicity and efficacy.

#### 4.6

Current experience with CAR T cell therapies indicate that the safety depends on the disease

## 5 RISKS AND MITIGATION STRATEGY

This exploratory study is designed to assess the safety and biological activity of different dose schedules of AUTO3 in a limited number of patients. Given the treatment novelty, this study will involve patients whose disease has progressed after treatment with the main approved classes of anti-leukaemia agents. Such patients are considered to have poor prognosis and are candidates for promising experimental therapies.

**Leukapheresis:** This is necessary for enrolling onto this study as it is a prerequisite for preparing AUTO3. This is generally performed in specialist units, where such procedures are relatively standard. Cell production efficiency of at least 80% is anticipated and is dependent on the quality of the leukapheresate. AUTO3 infusion may or may not provide clinical benefit to these patients. Risks of leukapheresis are summarised in [Table 7](#).

**Table 7: Leukapheresis – Risks and Mitigation Strategy**

Risks	Mitigation Strategy
<b>Pain and bruising</b> due to insertion of cannula/ central venous access.	Experienced clinicians/nurses performing the procedure, analgesics to be used as needed for pain
<b>Bacterial bloodstream infections</b> associated with the insertion of access and return venous access devices.	The procedure will be carried out by trained and experienced personnel and risks will be minimised by strict adherence to aseptic measure.
<b>Symptoms of hypocalcaemia</b> e.g. muscle cramps due to chelation by anticoagulants used to prevent clotting.	Patients will be monitored for symptoms of hypocalcaemia, the rate of citrate infusion to the patient and duration of the procedure will be controlled by experienced personnel. Calcium supplements will be given as needed

**Pre-conditioning chemotherapy:** Lymphodepletion with FLU and CY prior to adoptive transfer of CAR T cells enhances treatment efficacy by eliminating regulatory T cells and increasing access of the transfused CAR T cells to activating cytokines. Pre-conditioning is expected to increase the survival of AUTO3 and therefore the chance of anti-tumour efficacy. Pre-conditioning will only be undertaken after confirmation of the successful production of AUTO3. Risks of pre-conditioning with chemotherapy are summarised in [Table 8](#). Please refer to the Summary of Product Characteristics for more details.

**Table 8: Pre-conditioning Chemotherapy – Risks and Mitigation Strategy**

Risks	Mitigation Strategy
<p><b>Myelosuppression</b> and immunosuppression resulting in anaemia, neutropenia, thrombocytopenia, and lymphopenia, are the most common toxicities. In addition to being a manifestation of ALL, myelosuppression can be a toxicity of chemotherapy and also of CAR T cells treatment. Transient moderate to severe myelosuppression lasting up to 30 days is anticipated in this patient cohort. Bleeding, neutropenic fever, infections, and septic shock may occur and may sometimes be fatal. Herpes zoster and other viral reactivations may occur.</p>	<p>The FLU and CY chemotherapy given is milder than most chemotherapy treatment regimens for patients with relapsed or refractory ALL and will be given only once (1 cycle, given over 3 days). Anti-microbial prophylaxis (including Pneumocystis prophylaxis and acyclovir) will be given to prevent infections and patients will be monitored for cytomegalovirus, adenovirus and Epstein-Barr virus viraemia weekly for the first month. If infections arise they will be treated as per institutional guidelines. Blood, platelet and fresh frozen plasma transfusions will be given as per standard institutional guidelines. All sites have extensive expertise in managing these complications in paediatric/adult patients with ALL and have on site intensive care unit support.</p>
<p><b>Cyclophosphamide</b> associated toxicities (as per approved label), including but not limited to nausea/vomiting, haemorrhagic cystitis, myocarditis and myopericarditis and tamponade, pneumonitis and pulmonary fibrosis, and veno-occlusive liver disease may occur.</p>	<p>Given the low dose and short duration of treatment, these toxicities are unlikely. Patients will be given anti-emetic prophylaxis and hydration during lymphodepletion as per institutional policy. If haemorrhagic cystitis occurs, i.v. fluids and mesna will be given. Other toxicities will be managed as per the standard institutional policy and by trained personnel.</p>
<p><b>Fludarabine</b> is generally well tolerated: the most common side effects are lymphopaenia and infection (see above). Neurotoxicity can occur but generally at higher doses. Other associated toxicities (as per the label) include, but are not limited to, autoimmune disorders, hepatic impairment, neurotoxicity, and renal impairment.</p>	<p>Given the low dose and short duration of treatment, these toxicities are unlikely. Toxicities will be managed as per the standard institutional policy and by trained personnel.</p>

ALL = acute lymphoblastic leukaemia; CAR = chimeric antigen receptor; CY = cyclophosphamide; FLU = fludarabine; i.v. = intravenous.

## AUTO3 infusion

Risks associated with the infusion of AUTO3 are presented in [Table 9](#) below and detailed management guidelines are presented in [Section 9.6](#).

**Table 9: AUTO3 Infusion – Risks and Mitigation Strategy**

Risks	Mitigation Strategy
<b>Infusion reactions</b> may occur with the infusion of AUTO3.	The product is autologous and the risk is likely to be low. Patients will be pre-medicated with chlorpheniramine and paracetamol.
<b>Cytokine-release syndrome</b> is a key toxicity expected for any CAR T cell treatment. Cytokine release syndrome may present as fevers, myalgia, hypoxia, hypotension and potentially life-threatening cardiac dysfunction, acute respiratory distress syndrome, neurologic toxicity, renal and/or hepatic failure, and disseminated intravascular coagulation. The clinical features may overlap with macrophage activation syndrome. In studies with second generation CD19 CAR T cells, the majority of patients have developed CRS but the severity appears to correlate with leukaemic burden.	The risk will be mitigated by adherence to CRS management guidelines as per (Lee et al. 2014), (Section 10.4) formulated based on prior CAR T cell-gene therapy experience at various institutions. Splitting of the CAR T cell dose at the higher dose level may reduce risk of severe CRS in patients with higher disease burden.
<b>Neurotoxicity</b> has been seen with CD19 CARs in patients with leukaemia and lymphoma. Clinical presentation can be variable and manifest as mild to severe encephalopathy, focal neurologic deficits particularly in speech, hallucinations, generalised seizures, severe irreversible neurologic deficits, cerebral oedema and occasionally death. The cause of neurotoxicity is not well-understood, although it is reported to be fully reversible in most cases.	The patient will be closely monitored for neurological signs and symptoms, neuroimaging will be performed as appropriate. Patients will be managed with early supportive care (e.g. anti-convulsants) as appropriate. In patients who develop severe neurotoxicity or cerebral oedema, dexamethasone will be given (Section 10.6).
<b>Off-tumour toxicity</b> could be due to either on-target (due to expression of the antigen on non-tumour cells) or off-target (recognition of a molecular target other than CD19 or CD22) interactions. Historically, there have been reports of on-target/off-tumour toxicity with CAR therapy as well as T cell receptor engineered T cells, the details are described in the IB.	Preclinical toxicology indicates the risk to be low as CD19 and CD22 expression is largely limited to the B cell lineage. Ongoing CD19 and or CD22 CAR studies have not shown significant off-target toxicity. Considering the body of knowledge with regards to targeting CD19 and CD22, this risk is likely to be low.
<b>Cardiac toxicity:</b> One cardiac-related fatality has been reported with CD19 CAR T cells, although the causality was unclear.	The protocol excludes patients with underlying/prior cardiac history. Additionally, patients will be required to undergo echocardiogram (ECHO) or MUGA to demonstrate normal cardiac function prior to study enrolment and at the onset of CRS.
<b>Hypoxia:</b> Severe hypoxia has been reported in patients treated with a CD22 CAR, JCAR018. The causality was unclear but in at least 1 case it was associated with CRS. Hypoxia is a recognised manifestation of severe CRS. It is unlikely to be a consequence of off-target toxicity specific for CD22 as there is no expression of CD22 in lung except on the resident B-cells (as CD19).	All patients will be evaluated for oxygen saturation at baseline, and through the duration of the study. Patients with a baseline oxygen saturation of <92% on room air and those requiring supplemental oxygen at the time of AUTO3 infusion will not be eligible to receive AUTO3. Patients with an oxygen requirement of >40% (or lower based on emerging data) in setting of CRS will receive treatment with tocilizumab. Patients with hypoxia will also be investigated for pulmonary infections and treated promptly.

Risks	Mitigation Strategy
<p><b>Tumour lysis syndrome (TLS)</b> may occur on treatment with AUTO3 due to rapid killing of malignant cells in the context of a high tumour burden, but this is rarely seen after CD19 CAR T cell therapy.</p>	<p>In patients with high leukaemic burden (<math>\geq 25\%</math> blasts in the BM) allopurinol and i.v. fluids will be given. If TLS occurs, all sites have extensive experience in managing this complication and supportive care will be initiated rapidly as per standard institutional protocols (<a href="#">Section 10.5</a>).</p>
<p><b>Hypogammaglobulinaemia</b> may occur because of depletion of normal B cells by AUTO3. This may potentially increase the risk of infections. The degree of hypogammaglobulinaemia and its duration are variable depending on the persistence of CAR T cells.</p>	<p>B cell recovery and Ig levels will be monitored regularly following AUTO3 infusion. Patients with recurrent and persistent severe hypogammaglobulinaemia will receive i.v. Ig replacement (<a href="#">Section 10.7</a>).</p>
<p><b>Insertional mutagenesis:</b> Activation of proto-oncogenes by retroviral-mediated insertional mutagenesis following gene therapy into haemopoietic stem cells has resulted in leukaemogenesis. However, this has not been seen after transduction of terminally differentiated T cells. There are no reported cases of insertional mutagenesis in <math>&gt;200</math> patients treated with genetically modified T cells (including CAR T cells) over an extended period of follow-up.</p>	<p>Patients will be monitored for secondary malignancy and survival in a long-term follow-up protocol for a total of 15 years.</p>
<p><b>Risks associated with a RCR:</b> There is a risk that a recombination event may occur during vector production that results in a RCR, which may be pathogenic in humans.</p>	<p>All vector lots are tested for RCRs prior to release to sites. The risks of transmission of RCRs are extremely low and to date, no patient has developed a RCR following retroviral based CAR T cell therapy. Patients will be monitored for a RCR by polymerase chain reaction (PCR) during their scheduled follow-up visits. If a positive signal is confirmed, additional testing will be performed and medical and research experts will be consulted for the optimal treatment approach should any complication arise.</p>
<p><b>Dimethyl sulfoxide (DMSO)</b> which is part of cryopreservative buffer may cause an abnormal taste at the time of infusion and body odour lasting 1-2 days afterwards. At high doses, DMSO may cause nausea, vomiting, abdominal pain, headache and haemolysis. Rarely, patients may experience mild or severe cardiac, pulmonary, renal, or neurological symptoms.</p>	<p>Most patients are likely to be exposed to a small dose of DMSO. Even at the highest proposed dose of <math>10 \times 10^6</math> CAR-positive T cells/kg (or <math>700 \times 10^6</math> CAR-positive T cells in a 70 kg adult), the maximum exposure to DMSO would be 52.5 g (assuming a 70 kg patient and a minimum transduction of 5% and formulation of product at <math>20 \times 10^6</math> viable cells per mL) which is lower than the standard institutional maximum dose of 70 g (1 g/kg). At these dose levels the side effects are likely to be mild and short lasting.</p>
<p><b>Graft versus host disease (GVHD):</b> Some subjects, will be recruited to the study after allogeneic HSCT and there is a theoretical risk of alloreactivity of AUTO3 and consequent GVHD. However the risk is low, several studies have reported little or no evidence of GVHD following CD19 CAR T cell therapy in patients treated post allogeneic HSCT.</p>	<p>Patients who have developed severe GVHD will be excluded from the study effectively screening out potential for alloreactivity. T cells will be collected from the recipient, these harvested donor T cells will have been subject to tolerization within the recipient including thymic education, reducing alloreactivity. In case of GVHD, study subjects will be treated as per institutional standard of care and at the investigator's discretion.</p>

BM = bone marrow; CD = cluster of differentiation; CAR = chimeric antigen receptor; CRS = cytokine release syndrome; DMSO = dimethyl sulfoxide; ECHO = echocardiogram; GVHD = graft versus host disease; IB = Investigator's Brochure; Ig = immunoglobulin; i.v. = intravenous; MUGA = multigated acquisition; PCR = polymerase chain reaction; RCR = replication competent retrovirus; TLS = tumour lysis syndrome.

**Unknown long-term risks of gene therapy:** Following completion of this study, all patients will be enrolled and monitored for SAEs, AEs of interest and survival in a long-term follow-up study.

## 6 PATIENT POPULATION

Patients will be eligible for the trial if all the inclusion criteria are met and none of the exclusion criteria applies. There will be no exception to the eligibility requirements at the time of registration. Ensuring patient eligibility is the responsibility of the Principal Investigator or other delegated Investigator(s).

### 6.1 INCLUSION CRITERIA

#### 6.1.1 Paediatric and Young Adult Patients

Patients must meet all the following criteria for study entry:

1. Male or female paediatric and young adult patients (aged 1-24 years) with HR relapsed or refractory B-lineage ALL **and**
  - a. Any BM relapse or CNS relapse with detectable BM disease ( $>10^{-4}$ ) by flow cytometry/molecular MRD after allogeneic SCT and must be  $\geq 6$  months from SCT at the time of AUTO3 infusion, OR
  - b. High Risk first relapse (as per International Study for Treatment of Childhood Relapsed ALL criteria [Appendix 6](#)), OR
  - c. Standard risk relapse patients with HR cytogenetics (HR defined as mixed linkage leukaemia gene rearrangement (KMT2A), intrachromosomal amplification of chromosome 21 amplification, near-triploidy (60-78 chromosomes) or near-haploidy ( $<30$  chromosomes) and low hypodiploidy (30-39 chromosomes), OR
  - d. Second or greater relapse, OR
  - e. Bone marrow MRD  $\geq 10^{-3}$  prior to planned SCT, OR
  - f. Any on-treatment relapse in patients aged 16-24 years.

#### **Phase II Only (criteria in addition to those described above)**

- g. Primary refractory disease defined as MRD  $\geq 5\%$  blasts in the BM by flow cytometry or molecular assay following frontline induction therapy. For good risk cytogenetics, MRD  $>0.1\%$  at Week 9 of consolidation chemotherapy is also required. (*Note: For patients with good risk cytogenetics the leukapheresis may be done before starting consolidation therapy but manufacturing and AUTO3 infusion will only occur if MRD is  $>0.1\%$  at Week 9 of consolidation*), OR
    - h. Patients with Philadelphia chromosome-positive ALL are eligible if they are intolerant to or have failed 2 lines of tyrosine kinase inhibitor therapy, or if tyrosine kinase inhibitor therapy is contraindicated, OR
    - i. Isolated CNS relapse (post-SCT or pre-SCT if high risk, aged 16-24 years on therapy relapse or  $\geq 2^{\text{nd}}$  relapse) but with  $\leq$  CNS Grade 2 disease at time of enrolment.
2. Documentation of CD19 and or CD22 expression on leukaemic blasts in the BM, peripheral blood, or CSF by flow cytometry within 3 months of screening.
3. Detectable disease in the BM at a level  $\geq 10^{-4}$  by molecular or flow cytometry based methods (Phase I only) at enrolment (patients developing  $\leq 10^{-4}$  BM disease due to bridging therapy may continue to receive AUTO3).

4. Absolute lymphocyte count  $\geq 0.5 \times 10^9/L$  at enrolment.
5. Adequate renal, hepatic, pulmonary, and cardiac function defined as:
  - Serum creatinine based on age/gender  $\leq 1.5 \times$  upper limit of normal (ULN).
  - Serum alanine aminotransferase/aspartate aminotransferase  $\leq 5 \times$  ULN.
  - Total bilirubin  $\leq 2 \times$  ULN, except in subjects with Gilbert's syndrome.
  - Left ventricular shortening fraction  $\geq 28\%$  confirmed by ECHO, or left ventricular ejection fraction (LVEF)  $\geq 45\%$  confirmed by ECHO.
  - Baseline oxygen saturation  $>92\%$  on room air.
6. Karnofsky (age 10-24 years) or Lansky (age  $<10$  years) score  $\geq 50\%$ .
7. Willing and able to give written, informed consent to the current study (patient and/or parent or legal guardian).

### 6.1.2 Adult Patients

1. Age 25 or older.
2. Eastern cooperative oncology group (ECOG) performance status of 0 or 1.
3. Relapsed or refractory B-precursor ALL defined as one of the following:
  - a. Primary refractory disease
  - b. First relapse if first remission  $\leq 12$  months
  - c. Relapsed or refractory disease after 2 or more lines of systemic therapy
  - d. Relapsed or refractory disease after allogeneic transplant provided individual is at least 6 months from stem cell transplant at the time of infusion.
4. Patients with Philadelphia chromosome-positive ALL are eligible if they are intolerant to or have failed 2 lines of tyrosine kinase inhibitor therapy, or if tyrosine kinase inhibitor therapy is contraindicated.
5. Documentation of CD19 and/or CD22 expression on leukaemic blasts in the BM, peripheral blood, or CSF by flow cytometry within 3 months of screening.
6. For females of childbearing potential (defined as  $<24$  months after last menstruation or not surgically sterile), a negative serum or urine pregnancy test must be documented at screening, prior to pre-conditioning and confirmed before receiving the first dose of study treatment.

For females who are not postmenopausal ( $<24$  months of amenorrhea) or who are not surgically sterile (absence of ovaries and/or uterus), two methods of contraception comprising of one highly effective method of contraception together with a barrier method must be used during the treatment period and for at least 12 months after the last dose of study treatment. They must agree not to donate eggs (ova, oocytes) for the purposes of assisted reproduction during the study and for 12 months after receiving the last dose of study drug (please refer to [Appendix 5](#)).
7. For males, it must be agreed that two acceptable methods of contraception are used (one by the patient – usually a barrier method, and one highly effective method by the patient's partner as defined in [Appendix 5](#)) during the treatment period and for at least

12 months after the last dose of study treatment and that sperm will not be donated during the treatment period and for at least 12 months after the last dose of study treatment.

8. Absolute lymphocyte count  $\geq 0.5 \times 10^9/L$  at enrolment.
9. Adequate renal, hepatic, pulmonary, and cardiac function defined as:
  - a. Serum alanine aminotransferase/aspartate aminotransferase  $\leq 2.5 \times ULN$ .
  - b. Creatinine clearance (as estimated by Cockcroft Gault)  $\geq 60$  cc/min.
  - c. Total bilirubin  $\leq 1.5$  mg/dl, except in subjects with Gilbert's syndrome.
  - d. Left ventricular ejection fraction (LVEF)  $\geq 45\%$  confirmed by ECHO or MUGA.
  - e. Baseline oxygen saturation  $> 92\%$  on room air.

## 6.2 EXCLUSION CRITERIA

### 6.2.1 Paediatric and Young Adult Patients

Patients meeting any of the following exclusion criteria must not be enrolled into the study:

1. Isolated extra-medullary disease relapse (in Phase II of the study, patients with isolated CNS relapse post-SCT or pre-SCT if high risk, aged 16-24 years on therapy relapse or  $\geq 2^{\text{nd}}$  relapse with  $\leq$  CNS Grade 2 disease at time of enrolment are eligible).
2. Active CNS involvement of ALL, defined by CNS Grade 3 per National Comprehensive Cancer Network guidelines. Patients developing CNS Grade 3 disease at any time after enrolment will also be excluded.
3. Active infectious bacterial or viral disease (hepatitis B virus, hepatitis C virus, human immunodeficiency virus (HIV), human T-cell lymphotropic virus [HTLV], syphilis, West Nile (US only) or Zika viruses (US only)) requiring i.v. anti-microbials for treatment.
4. Females who are pregnant or lactating.
5. Females of child-bearing potential (defined as all females physiologically capable of becoming pregnant) and post-pubertal male participants who are unwilling to use highly effective methods of contraception during the treatment period and for a period of 1 year after the AUTO3 infusion. *Note: Examples of highly effective contraception methods are described in [Appendix 5](#).*
6. Inability to tolerate leukapheresis.
7. Prior CD19 or CD22 targeted therapy with Grade 4 toxicity (except for haematological toxicity) or  $\geq$  Grade 3 CRS or  $\geq$  Grade 3 drug-related CNS toxicity. CD22 targeted therapy such as inotuzumab ozogamicin in patients that are CD19 negative (unless it is demonstrated that this therapy had no effect on CD22 target expression). In Phase II, patients with prior CAR therapy will be excluded.
8. Pre-existing significant neurological disorder (other than CNS involvement of underlying haematological malignancy).
9. Stem cell transplant patients only: Active significant (overall Grade  $\geq$  II, Seattle criteria) acute graft versus host disease (GVHD) or moderate/severe chronic GVHD (NIH

consensus criteria) requiring systemic steroids or other immunosuppressants within 4 weeks of enrolment.

10. The following medications are excluded:
  - a. Steroids: Therapeutic doses of steroids must be stopped >72 hours prior to AUTO3 infusion and leukapheresis. However, physiological replacement doses of steroids are allowed: <12 mg/m<sup>2</sup>/day hydrocortisone or equivalent.
  - b. Allogeneic cellular therapy: Any donor lymphocyte infusions must be completed >6 weeks prior to AUTO3 infusion.
  - c. Graft versus host disease therapies: Any drug used for GVHD must be stopped >4 weeks prior to AUTO3 infusion.
  - d. Chemotherapy: Should be stopped 1 week prior to leukapheresis and 2 days prior to starting pre-conditioning chemotherapy.
  - e. Intrathecal therapy: Methotrexate within 4 weeks and other intrathecal chemotherapy (e.g. Ara-C) within 2 weeks prior to starting pre-conditioning chemotherapy.
  - f. Live vaccine ≤4 weeks prior to enrolment.
11. Known allergy to albumin, DMSO, CY or FLU.
12. Any other condition which in the Investigator's opinion would prevent the patient from undergoing protocol-based therapy.

### **6.2.2 Adult Patients**

1. Isolated extramedullary disease.
2. Diagnosis of Burkitt's leukaemia/lymphoma according to World Health Organization (WHO) classification or chronic myelogenous leukaemia lymphoid blast crisis.
3. Females who are pregnant or lactating.
4. History or presence of clinically relevant CNS pathology such as epilepsy, paresis, aphasia, stroke within 3 months prior to enrolment, severe brain injuries, dementia, Parkinson's disease, cerebellar disease, organic brain syndrome, uncontrolled mental illness, or psychosis.
5. Presence of CNS-3 disease or CNS-2 disease with neurological changes. Patients developing CNS Grade 3 disease or symptomatic CNS-2 disease at any time after enrolment will also be excluded.
6. Clinically significant, uncontrolled heart disease (New York Heart Association Class III or IV heart failure, uncontrolled angina, severe uncontrolled ventricular arrhythmias, sick-sinus syndrome, or electrocardiographic evidence of acute ischaemia or Grade 3 conduction system abnormalities unless the patient has a pacemaker) or a recent (within 12 months) cardiac event.
  - a. Uncontrolled cardiac arrhythmia (patients with rate-controlled atrial fibrillation are not excluded).
  - b. Evidence of pericardial effusion.
7. Patients with a history (within 3 months) or evidence of pulmonary embolism requiring ongoing therapeutic anticoagulation at the time of pre-conditioning.

8. Patients with active gastrointestinal (GI) bleeding.
9. Patients with any major surgical intervention in the last 3 months.
10. Active infectious bacterial or viral disease or fungal (hepatitis B virus, hepatitis C virus, HIV, HTLV, syphilis, West Nile virus [US only] or Zika virus [US only]) requiring treatment with i.v. antimicrobials.
11. History of autoimmune disease (e.g. Crohn's, rheumatoid arthritis, systemic lupus) resulting in end organ injury or requiring systemic immunosuppression/systemic disease modifying agents within the last 24 months. Any autoimmune disease with CNS involvement.
12. History of other malignant neoplasms unless disease free for at least 24 months (carcinoma *in situ*, non-melanoma skin cancer, breast or prostate cancer on hormonal therapy allowed).
13. History of concomitant genetic syndrome such as Fanconi anaemia, Shwachman-Diamond syndrome, Kostmann syndrome or any other known BM failure syndrome.
14. Stem cell transplant patients only: Active significant (overall Grade  $\geq$ II, Seattle criteria) acute GVHD or moderate/severe chronic GVHD (NIH consensus criteria) requiring systemic steroids or other immunosuppressants within 4 weeks of enrolment.
15. Prior CD19 or CD22 targeted therapy other than blinatumomab and inotuzumab ozogamicin.
16. The following medications are excluded:
  - a. Steroids: Therapeutic doses of corticosteroids within 7 days of leukapheresis or 72 hours prior to AUTO3 administration. However, physiological replacement, topical, and inhaled steroids are permitted.
  - b. Immunosuppression: Immunosuppressive medication must be stopped  $\geq$ 2 weeks prior to leukapheresis or AUTO3 infusion.
  - c. Allogeneic cellular therapy: Any donor lymphocyte infusions must be completed  $>$ 6 weeks prior to AUTO3 infusion.
  - d. Graft versus host disease therapies: Any drug used for GVHD must be stopped  $>$ 4 weeks prior to AUTO3 infusion.
  - e. Chemotherapy including TKIs for Philadelphia chromosome-positive ALL: Should be stopped 1 week prior to leukapheresis or 2 days prior to starting pre-conditioning chemotherapy.
  - f. Treatment with alemtuzumab within 6 months prior to leukapheresis, or treatment with clofarabine or cladribine within 3 months prior to leukapheresis.
  - g. Live vaccine  $\leq$ 4 weeks prior to enrolment.
  - h. Intrathecal therapy: Methotrexate within 4 weeks and other intrathecal chemotherapy (e.g. Ara-C) within 2 weeks prior to starting pre-conditioning chemotherapy.
  - i. Systemic inhibitory/stimulatory immune checkpoint - At least 2 half-lives must have elapsed from any prior systemic inhibitory/stimulatory immune checkpoint molecule therapy prior to enrolment.

17. Presence of any indwelling line or drain (e.g., percutaneous nephrostomy tube, indwelling Foley catheter, biliary drain, or pleural/peritoneal/pericardial catheter). Ommaya reservoirs and dedicated central venous access catheters such as a Port-a-Cath or Hickman catheter are permitted.
18. Research participants receiving any other investigational agents, or concurrent biological, chemotherapy, or radiation therapy.
19. Inability to tolerate leukapheresis.
20. Patients, who in the opinion of the Investigator, may not be able to understand or comply with the safety monitoring requirements of the study or unlikely to complete all protocol-required study visits or procedures, including follow-up visits.

**Note:** Patients failing any of the eligibility criteria, after leukapheresis or AUTO3 product manufacture, such as those with progressive CNS disease and are discontinued may re-enter the study when they are considered to be eligible again. If AUTO3 product has been manufactured they can be treated at the current dose using the original product if it continues to meet release criteria.

**For Pre-conditioning or AUTO3 Infusion:** Patients meeting any of the following exclusion criteria must not be given pre-conditioning or treated with AUTO3 or treatment should be delayed until they no longer meet these criteria:

1. Severe intercurrent infection at the time of, or within 14 days of, scheduled AUTO3 infusion.
2. Requirement for supplementary oxygen at the time of scheduled pre-conditioning or AUTO3 infusion.
3. Allogeneic transplant recipients with active significant acute GVHD overall Grade  $\geq$ II or moderate/severe chronic GVHD requiring systemic steroids at the time of scheduled pre-conditioning or AUTO3 infusion.

*Note: Such patients will be excluded until the patient is GVHD free and off steroids.*

4. Significant deterioration of renal or hepatic function (Serum creatinine  $\geq$ 1.8 x ULN, Serum alanine aminotransferase or aspartate aminotransferase  $\geq$ 6 x ULN, Total bilirubin  $\geq$ 3 x ULN, except in subjects with Gilbert's syndrome) prior to pre-conditioning.

*Note: Patient with  $\geq$ Grade 3 CRS,  $\geq$ Grade 2 neurotoxicity or a DLT following the first split dose should not receive the second split dose.*





## 8 STUDY PROCEDURES AND TREATMENT

### 8.1 CONSENT, SCREENING, AND REGISTRATION

**Consent:** Patients, or their parent/legal guardian where appropriate, must sign an Informed Consent Form (ICF) prior to the conduct of any study-related procedures. The trial includes both paediatric and adult patients. Adults ( $\geq 16$  or 18 years of age) will be informed of all aspects of the trial and will be asked for written consent. For patients aged  $< 16$  (or 18 as per local regulations) years, the person with parental responsibility or legal guardianship of the child must be informed of all aspects of the trial and written consent will be obtained from this person. Additionally, the clinician will explain the study to patients of appropriate age and assent will be obtained from the patient whenever it is possible to do so. The child must be informed about the trial to the extent compatible with their understanding.

Information sheets are available for different age groups as well as for the parent/legal guardian. Sites are responsible for assessing a patient's (and/or parent/legal guardian's) capacity to give informed consent (or assent).

Sites must ensure that all patients, and/or parents/legal guardians, have been given the current approved version of the Patient Information Sheets (PIS), are fully informed about the trial, and have confirmed their willingness to take part in the trial by signing the current approved consent (or assent) form. Sites must assess a patient's (and/or parent/legal guardian's) ability to understand verbal and written information in English or the local language, and whether an interpreter will be required to ensure fully informed consent. If a patient (and/or parent/legal guardian) requires an interpreter and none is available, the patient should not be considered for the trial.

The Investigator, or, where delegated by the Investigator, other appropriately trained site staff, are required to provide a full explanation of the trial and all relevant treatment options to each patient (and/or parent/legal guardian) prior to trial entry. During these discussions, the current approved PIS for the trial should be discussed with the patient (and/or parent/legal guardian). Sufficient time must be allowed for the patient (and/or parent/legal guardian) to consider and discuss participation in the trial. Written informed consent/assent on the current approved version of the consent/assent form for the trial must be obtained (according to national requirements and guidance) before any trial-specific procedures are conducted. The discussion and consent/assent process must be documented in the patient notes.

**Patient ID number Assignment:** After providing written informed consent, the patient will be issued with a unique patient identification number. The patient identification number will be used to identify the patient for the duration of the study. Patient identification numbers will not be reassigned or reused.

**Screening:** Following informed consent, patients will be screened for eligibility for the study (Day -84 to Day -35). During the screening phase, eligibility criteria will be reviewed, a complete clinical evaluation and the investigations below will be performed. If an assessment was performed as part of the patient's routine clinical evaluation and not specifically for this study, it does not need to be repeated after signed informed consent has been obtained if the assessments fulfil the study requirements and are performed within the specified timeframe prior to AUTO3 administration. Retesting of abnormal screening values that lead to exclusion is allowed during the screening phase (to reassess eligibility). The patient should continue to meet protocol specified renal, hepatic, and pulmonary function requirements prior to initiating pre-conditioning chemotherapy. Adverse events associated with screening

procedures will be collected. Baseline data will be collected during screening as described in the Schedule of Assessments and will include:

- A full medical history and physical examination including weight, vital signs and oxygen saturations
- Clinical assessment to exclude active significant (overall Grade  $\geq$ II, Seattle criteria) acute GVHD or moderate/severe chronic (NIH consensus criteria) GVHD (SCT patients only)
- Karnofsky (age  $\geq$ 10-24 years), Lansky (age <10 years), or ECOG (age  $\geq$ 25 years) score
- Assessment of patient notes to undergo leukapheresis
- pregnancy test in female patients of child-bearing potential
- Full blood count and coagulation
- Biochemical assessment
- Infectious disease screening (see below)
- Peripheral blood chimerism in T cell and myeloid lineage (post-BM transplant patients only)
- Electrocardiogram (ECG) and ECHO
- Bone marrow sample for morphology, immunophenotyping, cytogenetics, molecular and flow-based MRD
- Lumbar puncture
- Blood for serum cytokines

*Infectious disease screening:* Patients will be tested by the site (in accordance with the site Human Tissue Authority license for human application) for infectious diseases as outlined below prior to (within 30 days of) leukapheresis. [Table 10](#) lists the tests that must be performed as a minimum requirement:

**Table 10: Infectious Disease Screening**

Pathogen	Test
HIV 1 and 2	Anti-HIV-1 and 2
Hepatitis B	Hepatitis B surface antigen, anti-Hepatitis B core antibody
Hepatitis C	Anti-hepatitis C virus-antibody
Syphilis	Syphilis serology (chemiluminescent microparticle immunoassay)*
HTLV 1 and 2	Anti-HTLV-1 and 2
West Nile and Zika (both US only)	As per local testing

HIV = human immunodeficiency virus; HTLV = human T-cell lymphotropic virus.

\* A validated testing algorithm must be applied to exclude the presence of active infection with *Treponema pallidum*. A non-reactive test, specific or non-specific, can allow tissues and cells to be released. When a non-specific test is performed, a reactive result will not prevent procurement or release if a specific *Treponema* confirmatory test is non-reactive. A patient whose specimen tests reactive on a *Treponema*-specific test will require a thorough risk assessment to determine eligibility for clinical use.

A screening log must be maintained by the site and filed in the Investigator Site File.

**Patient Registration:** Patient registration will be performed prior to commencement of any trial treatment/intervention. Following screening evaluations, confirmation of eligibility and consent of a patient or parent/legal guardian at a site, the registration form must be fully completed and then sent to Autolus. The eligibility information will be reviewed, and registration approved by the medical monitor.

After registration of patients, the site will coordinate with Autolus on timing of leukapheresis to generate AUTO3. If the patient continues to meet the eligibility criteria for entry to the study (as outlined in [Section 6](#)), the site will liaise with the manufacturer to arrange transfer of the AUTO3 to the participating site. Further details are provided in the Cell Handling Instruction Manual.

## 8.2 LEUKAPHERESIS

Following informed consent, confirmation of eligibility and registration to the study, patients will undergo an unstimulated leukapheresis for the generation of AUTO3.

Prior to leukapheresis, the following medications are not permitted:

- Steroids: therapeutic doses must be stopped at least 72 hours before leukapheresis. Physiological replacement doses are permitted: <12 mg/m<sup>2</sup>/day hydrocortisone or equivalent.
- Chemotherapy: should be stopped 1 week prior to leukapheresis.

Leukapheresis must be performed within 30 days of infectious disease testing and will be carried out following the standard institutional process. An additional infectious disease test will be done on the day of leukapheresis or within 7 days after. In general, leukapheresis should be performed approximately 35 days before the planned AUTO3 dosing date, based on emerging data this window can be changed. This may require insertion of a central venous access device and is a day case procedure. Typically, a double volume leukapheresis will be performed. Should there be any issues with the procedure, such as inadequate cell collection for primary dose or for re-treatment or due to contamination of the apheresate etc then the patient may undergo a repeat leukapheresis if clinically appropriate.

The leukapheresate is the starting material for the manufacture of AUTO3. The total cell number that is required for successful manufacture varies according to the dose level. The target collection is  $1 \times 10^9$  PBMCs. If the collection is insufficient, the Sponsor will advise as to the feasibility of successful manufacture. If collection is determined to be inadequate, then collected cells may be used for research purposes as per the consent. The leukapheresate will be transported to the Sponsor for generation of AUTO3 at a temperature of 2 to 8°C within 48 hours, ideally within 24 hours. US centres will transport the leukapheresate to a freezing facility for onward frozen shipment to the manufacturing facility. Further details regarding this process can be found in the Cell Handling Instruction Manual.

Each leukapheresate will be identified by a unique patient identification number plus any additional patient identifiers as allowed per local regulations (typically initials and date of birth).

**Note:** For patients who have relapsed following SCT, AUTO3 will be generated from the patient (not the stem cell donor).

**Note:** Patients failing any of the eligibility criteria after apheresis or AUTO3 product manufacture such as those with progressive CNS disease should hold further study related

activity until they are considered to be eligible again even if it extends the overall screening window beyond 84 days. If stored apheresate from a prior leukapheresis (on the current study) is available or if AUTO3 product has been manufactured they can be treated using the original product (or manufactured using the available apheresate) if it continues to meet release criteria. These patients do not need to undergo leukapheresis again, but may do so if necessary and clinically appropriate. Patients should undergo evaluation and monitoring as prescribed from Day -7 onwards or earlier as appropriate. Additional investigations and assessments should be performed as clinically indicated (eg ECHO or ECG).

### 8.3 PRE-CONDITIONING CHEMOTHERAPY

Patients that still meet eligibility requirements for the study and in whom AUTO3 meets the release criteria will be admitted at the study site from the day before starting pre-conditioning chemotherapy until a minimum of 30 days ( $\pm 3$  days) post first infusion of AUTO3 (longer if necessary for monitoring and management). The pre-conditioning phase will begin with administration of pre-conditioning chemotherapy and will end with the beginning of treatment with AUTO3 infusion.

Prior to administration of pre-conditioning chemotherapy, patients will undergo clinical and laboratory assessments and the site investigator, or his designee will determine if the patient is fit to receive pre-conditioning chemotherapy. Assessment will include:

- Clinical history and physical examination (including neurological examination, see below) including weight, vitals and oxygen saturations
- Clinical assessment to exclude active significant (overall Grade  $\geq$ II, Seattle criteria) acute GVHD or moderate/severe chronic (NIH consensus criteria) GVHD (SCT patients only)
- Karnofsky (paediatric/young adult cohort age  $\geq$ 10-24 years) or Lansky (age <10 years) or ECOG (adult cohort age  $\geq$ 25 years)
- Repeat pregnancy test in female patients of child-bearing potential
- Full blood count
- Biochemical assessment
- Lymphocyte subsets and Igs
- Baseline assessment of serum cytokine levels
- Bone marrow sample for morphology, immunophenotyping, cytogenetics, molecular and/or flow-based MRD
- Lumbar puncture if clinically indicated
- Blood for RCR testing and insertional mutagenesis

See Schedule of Assessments for details.

*Neurological examination:* In any patient experiencing new CNS signs or symptoms at any time after enrolment, a detailed neurological assessment should be performed by a neurologist including appropriate imaging and diagnostics.

Following these assessments, patients will proceed to receive a lymphodepleting pre-conditioning treatment with CY for 2 days and FLU for 4 days (starting Day -6), timed to end 3 ( $\pm$ 1) days before AUTO3 infusion.

### 8.3.1 Pre-conditioning Chemotherapy Dose and Regimen

Cyclophosphamide and fludarabine dosing is described below. Fludarabine will be administered first.

- Fludarabine 30 mg/m<sup>2</sup> followed by cyclophosphamide 500 mg/m<sup>2</sup> day 1 (Day -6)
- Fludarabine 30 mg/m<sup>2</sup> followed by cyclophosphamide 500 mg/m<sup>2</sup> day 2 (Day -5)
- Fludarabine 30 mg/m<sup>2</sup> (Day -4)
- Fludarabine 30 mg/m<sup>2</sup> (Day -3)

The pre-conditioning chemotherapy should be completed a minimum of 3 days ( $\pm$ 1 day) prior to AUTO3 infusion.

**Fludarabine (FLU)** will be given by i.v. infusion over 30 minutes in sodium chloride 0.9%. For patients with renal impairment (glomerular filtration rate 30-60 mL/min/1.73 m<sup>2</sup>, or those who weigh <10 kg, the dose of FLU should be reduced according to standard institutional practice. When patients receive both cyclophosphamide (CY) and FLU, FLU will be given first. Patients that have received a cumulative fludarabine dose of >300 mg/m<sup>2</sup> and those who have received prior cranial irradiation (not total body irradiation) may receive only 90 mg/m<sup>2</sup> pre-conditioning.

**Cyclophosphamide** will be given by i.v. infusion over 30 minutes. Adequate pre- and post-hydration for up to 4 to 6 hours (or as per institutional practice) should be given post infusion to induce diuresis. Use of Mesna may be considered per institutional practice.

The dose of both drugs may be adjusted for weight of the patient as clinically appropriate. The anticipated minimum weight is approximately 10 kg, but may be slightly lower in some patients.

Anti-emetic prophylaxis will be given as per standard institutional policy. Prophylaxis for TLS with allopurinol and i.v. fluids may be given in patients with high leukaemic burden ( $\geq$ 25% blasts)

For additional information regarding both the drugs, please refer to the Summary of Product Characteristics.

### 8.3.2 Supply of Cyclophosphamide and Fludarabine

Cyclophosphamide and FLU are Non-Investigational Medicinal Products as they are not being tested or used as a comparator in this trial. Cyclophosphamide and FLU will be used to induce lymphodepletion in the current study as a pre-conditioning treatment, prior to AUTO3 treatment. Both drugs are authorised and commercially available in the UK, US and EU. Investigators will be responsible for their own supply of CY and FLU. Sufficient quantities of CY and FLU will be dispensed to cover the prescribed dose and will be prepared as per site Standard Operating Procedures and accordingly to manufacturer recommendations. Cyclophosphamide and FLU are cytotoxics and must be handled with care in accordance with local policy. Good aseptic practice must be employed when preparing CY and FLU solutions for infusion.

### 8.3.3 Accountability of Cyclophosphamide and Fludarabine

Pharmacy records should be kept of the CY and FLU dispensed to trial patients. The expiry date, batch number used, and manufacturer (if known) should be recorded as well as details of vials dispensed.

## 8.4 AUTO3 TREATMENT AND PATIENT MONITORING

The treatment phase will involve infusion of AUTO3 on Day 0 and will extend until the completion of the DLT period or until start of a new ALL therapy, whichever happens first.

### 8.4.1 Assessment Prior to AUTO3 Infusion

Prior to administration of AUTO3, patients will undergo clinical and laboratory assessments and the site Investigator or his designee will determine if the patient is eligible to receive AUTO3 as per [Section 6](#). These assessments should be performed prior to the first infusion of AUTO3 and will include:

- Clinical history and physical examination including vitals and oxygen saturations
- Clinical assessment to exclude active significant (overall Grade  $\geq$ II, Seattle criteria) acute GVHD or moderate/severe chronic (NIH consensus criteria) GVHD (SCT patients only)
- Full blood count
- Pregnancy test in female patients of child-bearing potential
- Biochemical assessment
- Coagulation
- Ferritin
- Baseline assessment of serum cytokine levels
- Bone marrow sample for morphology, immunophenotyping, cytogenetics, molecular and flow-based MRD if clinically indicated
- Lumbar puncture if clinically indicated

See Schedule of Assessments for details.

**Note:** Patients will not be eligible for infusion of AUTO3 if they have, or had, severe intercurrent infection within 14 days of infusion, have active significant ( $\geq$ Grade II) acute GVHD or moderate/severe chronic GVHD requiring systemic steroids, supplemental oxygen or active pulmonary infiltrates ([Section 6.2](#)). In this situation, AUTO3 infusion will be delayed and infusion may be performed as per dose delay guidelines ([Section 8.4.4](#)).

In addition, where patients are to receive a split dose, if after the first dose they experienced:

- Grade 4 or refractory Grade 3 CRS, Grade 4 neurotoxicity or a DLT: the second dose should not be given.

### 8.4.2 AUTO3 Administration

AUTO3 will be administered as a single rapid i.v. infusion within 30 minutes (as quickly as possible) from AUTO3 being thawed on Day 0 in an in-patient setting. Full details will be

provided in the Cell Handling Instruction Manual. Only the Investigator or Investigator's designee will dispense the study product.

Premedication with chlorpheniramine and paracetamol may be given prior to infusion of AUTO3 as per standard institutional practice (see [Section 10.2](#)), but steroids should not be given as part of premedication.

The Investigator or Investigator's designee or a study research nurse experienced in the administration of cellular blood products as per the trial-specific procedure will verify the patient details (unique patient identification number, initials, and date of birth) on the AUTO3 label matches the recipient, check the dose of cells, volume and number of cryobags to be infused and prescribe AUTO3 on a blood product chart. Details of the exact dose, volume, time of completion of thawing and time of completion of infusion will be documented in the applicable study records.

AUTO3 will be infused as described in the Cell Handling Instruction Manual. In brief, AUTO3 will be thawed in a 37°C water bath under sterile conditions the bag will be gently massaged until the cells have just thawed. There should be no frozen clumps left in the bag.

For patients  $\leq 50$  kg, the entire contents of the bag may be drawn up into a syringe, a normal saline rinse of the bag should be performed to ensure all cells have been drawn. AUTO3 will then be given as an i.v. infusion through a central venous line over up to **5-10 minutes** by the Investigator, Investigator's designee, or a study research nurse experienced in the administration of cellular blood products as per the Cell Handling Instruction Manual and the line flushed with normal saline. Aseptic techniques will be applied during the syringe filling process and administration.

For patients  $>50$  kg, following thawing of the bag in a 37°C water bath under sterile conditions, the entire contents of the bag will be given through an appropriate gauge blood set as an i.v. infusion through a central venous line over **5-15 minutes** by the Investigator, Investigator's designee, or a study research nurse experienced in the administration of cellular blood products as per the Cell Handling Instruction Manual and the line flushed with normal saline. **NO FILTER should be used in the line used for the infusion of the T cell product.** Alternatively, the administration may be performed using a syringe as described above for patients weighing  $\leq 50$  kg. If the AUTO3 cell product appears to have a damaged or leaking bag, or otherwise appears to be compromised, it should not be infused.

**The time between completion of thawing and completion of infusion should not exceed 30 minutes.**

#### **8.4.3 Monitoring During and After AUTO3 Administration**

All patients will be monitored closely with temperature, pulse, blood pressure, respiratory rate and oxygen saturations observed immediately prior to, and every 30 minutes ( $\pm 10$  minutes) for 4 hours after, AUTO3 infusion. In the event of allergic adverse reactions, anti-histamines may be administered, as well as oxygen and salbutamol in the event of respiratory distress.

Patients will be observed as an in-patient (in hospital or ambulatory care as clinically appropriate [ambulatory care setting is a facility close to hospital where a patient is seen by physician/nursing staff at least once a day]) for a minimum of 30 days ( $\pm 3$  days) post-AUTO3 infusion or longer if necessary for monitoring and management. Patients may be re-admitted based on emerging safety findings. During hospitalisation/ambulatory care, patients will be monitored clinically as per clinical practice and will undergo appropriate blood tests

(see Schedule of Assessments) for signs of toxicity, in particular for CRS, TLS and neurological disturbance and managed as detailed in [Section 9.6](#). Transfusions of blood products, antibiotics, analgesics, and intensive care will also be provided as clinically indicated. Urate levels will be monitored and treatment started if indicated.

Patients may be discharged from hospital/ambulatory care when clinically appropriate after a minimum of 30 days ( $\pm 3$  days) after AUTO3 infusion if deemed appropriate by the treating physician. The in-patient stay may be extended if this is felt to be necessary by the treating physician. The duration of admission of Phase II patients will be 30 days ( $\pm 3$  days) but may be reduced based on emerging data with a protocol amendment. The required study procedures and assessments to be conducted during the Treatment Stage are outlined in the Schedule of Assessments. Clinical evaluations and laboratory studies may be repeated more frequently, if clinically indicated.

Data regarding intensity of care will be collected on a periodic basis e.g. duration of in-patient stay or ambulatory care admission, ICU/ITU stay, duration of vasopressor support, mechanical ventilation, dialysis etc.

#### **8.4.4 Dose Delay**

If the pre-conditioning regimen is interrupted for intercurrent illness or other reasons, the patient may complete or recommence the pre-conditioning regimen after recovery, according to the Investigator's judgment after consultation with the Sponsor. Patients will be closely monitored during and after the pre-conditioning regimen.

If the patient has completed the pre-conditioning regimen but is unable to receive AUTO3 on Day 0 for any reason, clinical judgement will be used to decide whether it is appropriate to delay the administration of AUTO3 or to wait for the patient's safety blood results to recover sufficiently before repeating the pre-conditioning regimen and administering AUTO3.

If the patient is deemed unsuitable to receive the AUTO3, they will be discontinued from the clinical trial (see [Section 15.3](#)) and replaced. Each such case will be discussed with the Sponsor.

#### **8.4.5 Interruption of Infusion**

In the event of severe infusion reaction, the FLU/CY/AUTO3 infusion should be stopped and the patient treated as clinically indicated. When the patient has recovered, the infusion may be restarted.

Interruption of AUTO3 should not be greater than 30 minutes after thawing of AUTO3. If an infusion is interrupted for mechanical, technical or any other reason, then this should be dealt with according to local practice and the infusion restarted as soon as possible. In case of uncertainty, individual cases should be discussed with the Sponsor and documented.

#### **8.4.6 Duration of Treatment**

In most patients, it is expected that AUTO3 will be given once, or twice as a split dose. However, if a patient has sufficient AUTO3 remaining from the original manufacture and meets the re-treatment criteria, a second treatment may be given (see Re-treatment of Patients; [Section 8.4.7](#)).

#### **8.4.7 Re-treatment of Patients**

It is expected that most patients will receive a single or split dose infusion of AUTO3, given 3-10 days apart as part of their treatment. However, some patients may be considered

appropriate to receive a re-treatment (as a single or split infusion) of their own stored AUTO3. Prior to re-treatment, the patient needs to meet the following criteria:

■ [REDACTED]

OR

■ [REDACTED]

OR

■ [REDACTED]

**AND all of the following:**

■ [REDACTED]

■ [REDACTED]

■ [REDACTED]

■ [REDACTED]

Patients undergoing a second AUTO3 infusion should receive the same pre-conditioning. The dose of AUTO3 can be at (or up to) the highest dose that has been declared safe after a minimum of 3 patients have completed the DLT period and evidence of efficacy ([Appendix 1](#)) has been shown. These patients will not be considered evaluable for dose escalation decision making but in the event of a DLT in these patients, it will be considered in dose escalation decision making. A minimum of 7 days will be maintained between any patients treated at this dose level. Depending on the number of cells available, an intermediate dose may also be administered if considered appropriate. The decision to re-treat a patient will be made by the treating Investigator and Sponsor in consultation with the SEC. The retreatment will occur only after CRS or neurotoxicity has resolved. Patients re-treated will be monitored in a similar way to patients being treated for the first time, in that they would start evaluation and management as defined in the protocol, starting from the pre-conditioning stage.

Patients treated in the US will not have an option of retreatment until it is agreed upon after discussions with the FDA.

## **8.5 FOLLOW-UP PHASE**

Upon completion of the treatment phase (30 days [±3 days] post last AUTO3 infusion), patients will enter the follow-up phase where they will continue to be followed monthly for the first 4 months (relative to the first dose) and then every other month until the end of the

year and then every 3 months until 24 months for the assessment of anti-tumour response. Responding patients will undergo clinical assessment, laboratory tests and biomarker evaluations as described in the Schedule of Assessments. Grade 3 or higher AEs that are considered related to the study medication will be recorded/reported as detailed in [Section 12.3](#).

Patients that have disease progression or those withdrawn from the study earlier than 24 months will complete the End of Study visit and transition into a long-term follow-up study (AUTO-LT1 – see [Section 8.6](#)). The End of study visit can be delayed until start of a new treatment, and additional biomarkers related to disease relapse or CAR persistence may be collected.

**Note:** Patients who have received AUTO3 and have discontinued or completed the study may continue to be periodically monitored for AUTO3 treatment-related SAEs, AEs of special interest, and new malignancy until they enrol onto the long-term follow-up study (AUTO-LT1), and the End of Study visit will be delayed until time of such enrolment. Additionally, blood samples for CAR T cell persistence and RCRs may also be collected every 3 months.

## **8.6 LONG-TERM FOLLOW-UP**

Provided that patients, or parents/legal guardian(s), are willing, and sign a new informed consent/assent form, they will be enrolled onto a long-term follow-up protocol (AUTO-LT1) after completion of AUTO3-PA1. They will be monitored for SAEs considered related to the study treatment, AEs of special interest and any new malignancy for a period of up to 15 years following treatment with AUTO3.

## **9 STUDY ASSESSMENTS**

### **9.1 DEMOGRAPHIC AND BASELINE ASSESSMENTS**

The following will be collected at screening to determine eligibility and baseline status of the patient.

#### **9.1.1 Demographic Data and Baseline Variables**

Demographic data will include self-reported race/ethnicity, age, gender, and height and will be recorded at the screening visit prior to leukapheresis.

#### **9.1.2 Medical/Leukaemia History**

Medical history includes clinically significant diseases, surgeries, leukaemia history (including highest white blood cell [WBC] count cytogenetics/CNS involvement at presentation, timing, and sites of relapse, immunophenotype including CD19 and CD22 expression at relapse, all prior therapy for leukaemia including transplant, and prior GVHD and current disease status), all medications used by the patient and known allergies. Histological/cytological confirmation of disease diagnosis will be obtained (pathology report).

#### **9.1.3 Pregnancy Test**

Serum ( $\beta$ -human chorionic gonadotropin) or urine pregnancy testing will be performed for females of childbearing potential at the screening visit and will be repeated when the patient is admitted prior to starting lymphodepletion as described in the Schedule of Assessments.

### **9.2 SAFETY EVALUATIONS**

All patients who receive AUTO3 will be considered evaluable for toxicity assessment. Any clinically relevant changes occurring during the study must be recorded in the AE section of the Electronic Case Report Form (eCRF). Safety assessments will be based on medical review of AE reports and the results of vital sign measurements, ECGs, physical examinations, and clinical laboratory tests performance status assessment at specified time points as described in the Schedule of Assessments. Any clinically significant abnormalities persisting at the end of the study/early withdrawal will be followed by the Investigator until resolution, or until a clinically stable endpoint is reached. The study will be monitored by the SEC and IDMC; details regarding the SEC and IDMC are provided in [Section 13](#). The study will include the following evaluations of safety and tolerability according to the time points provided in the Schedule of Assessments.

#### **9.2.1 Adverse Events and Toxicity**

Adverse events will be noted by clinic staff or reported by the patient (or, when appropriate, by a caregiver, surrogate, or the patient's legally acceptable representative) for the duration of the study. Adverse event recording and reporting is described in detail in [Section 12.3](#).

Toxicity will be graded using the NCI CTCAE version 5.0 criteria. To assess the effect of AUTO3 on normal B cell immunity, circulating B cell counts and Ig levels will be assessed as outlined in the Schedule of Assessments and the incidence and duration of hypogammaglobulinaemia determined.

### 9.2.2 Clinical Laboratory Tests

Blood samples for haematology, coagulation and biochemistry will be collected at each visit as specified in the Schedule of Assessments. Where appropriate, tests must be performed prior to receiving pre-conditioning treatment or AUTO3 infusion. More frequent clinical laboratory tests may be performed if indicated by the overall clinical condition of the patient or by abnormalities that warrant more frequent monitoring. Assessments for Flu/Cy chemotherapy drug levels may be undertaken if clinically indicated. Screening laboratory results must be available to the Investigator for evaluation before AUTO3 infusion and subsequent laboratory results should be available at the time of the patient's evaluation by the treating physician. The Investigator must review the laboratory reports, document this review, and ensure that any clinically relevant changes occurring during the study are recorded in the AE section of the eCRF. The laboratory reports must be filed with the source documents. A summary of the tests that will be performed by the local laboratory is presented in [Table 11](#).

**Table 11: Clinical Laboratory Tests**

Assessment	Description
Haematology	Haemoglobin, platelet count, and WBC count with differential (neutrophils, lymphocytes).
Coagulation	Prothrombin time, international normalised ratio, activated partial thromboplastin time, and fibrinogen.
Biochemistry	Sodium, phosphate, potassium, magnesium, chloride, bicarbonate or total CO <sub>2</sub> , alanine aminotransferase, serum uric acid/urate, blood urea nitrogen/urea, creatinine, serum creatine phosphokinase, total bilirubin, calcium, C-reactive protein, and albumin. All tests must be performed prior to AUTO3 infusion on Day 0.
Limited Panel	Ferritin weekly until Day 21 or until resolution of CRS.
Pregnancy test	Serum (β-human chorionic gonadotropin) or urine pregnancy testing for females of childbearing potential.
Serology (at screening only)	<ul style="list-style-type: none"> <li>• HIV antibody.</li> <li>• Hepatitis B core antibody: if positive, further testing (deoxyribonucleic acid [DNA] by PCR) to rule out active disease or chronic carrier. Must be confirmed negative prior to screening.</li> <li>• Hepatitis C virus antibody: if positive for hepatitis C virus, further testing (by ribonucleic acid PCR) should be performed to rule out active infection.</li> <li>• Anti-HTLV-1.</li> <li>• Anti-HTLV-2.</li> <li>• Syphilis Serology.</li> <li>• West Nile and Zika viruses (both US only)</li> </ul>

CRS = cytokine release syndrome; DNA = deoxyribonucleic acid; HIV = human immunodeficiency virus; HTLV = human T-cell lymphocyte virus; PCR = polymerase chain reaction; WBC = white blood cell.

### 9.2.3 12-lead Electrocardiogram

A 12-Lead ECG will be obtained using an ECG machine that automatically calculates the heart rate and measures PR, RR, QRS, QT, and corrected QT intervals. Refer to the Schedule of Assessments for details regarding the frequency of ECG assessments. At each time point, a single 12-lead ECG will be performed by qualified site personnel. The clinical Investigator or designee will review the printout, including ECG morphology. The ECG should be repeated in triplicate if motion artefacts or clinically relevant abnormalities are noted. Additional cardiovascular assessments should be performed as clinically appropriate to ensure patient

safety. If blood sampling or vital sign measurement is scheduled for the same time point as ECG recording, the procedures should be performed in the following order: ECG(s), vital signs, blood draw.

#### **9.2.4 Echocardiogram and Multigated Acquisition**

Echocardiogram is the preferred method to assess if left ventricular shortening fraction is  $\geq 28\%$  or LVEF is  $\geq 45\%$  at screening; MUGA is an acceptable alternative for adult patients. Additional ECHO or MUGA assessments should be performed at the onset of CRS and when clinically indicated.

#### **9.2.5 Vital Signs**

Vital signs will include temperature, pulse/heart rate, respiratory rate, blood pressure (systolic and diastolic), oxygen saturation and weight. Blood pressure and pulse/heart rate measurements should be recorded with the patient in a seated or lying position. Multiple time points (minimum of 3) will be collected prior to treatment to establish a good baseline blood pressure for the patient. Blood pressure and pulse/heart rate measurements will be assessed with an automated device or manual techniques.

#### **9.2.6 Physical Examination**

A complete physical examination including detailed neurological assessment will be conducted at screening and on Day -7 as per the institutional standard practice. Thereafter, a symptom-directed physical examination will be conducted at subsequent visits. The schedule for physical examinations is provided in the Schedule of Assessments. In any patient experiencing new CNS signs or symptoms at any time after enrolment, a detailed neurological assessment should be performed by a neurologist including appropriate imaging and diagnostics.

#### **9.2.7 Karnofsky or Lansky or ECOG Performance Status**

The Karnofsky and Lansky scales provided in [Appendix 2](#) and [Appendix 3](#) will be used to grade changes in the patient's daily activities for patients aged <10 years and 10-24 years, respectively. For patients aged  $\geq 25$  years, the ECOG scale provided in [Appendix 4](#) will be used to grade changes in the patient's daily living activities. The frequency of assessments is provided in the Schedule of Assessments.

### **9.3 PHARMACODYNAMICS AND BIOMARKER EVALUATION**

Blood-based pharmacodynamic biomarkers will be evaluated in all patients as described in the Schedule of Assessments. Peripheral and BM biomarkers may be assessed pre- and post-treatment with AUTO3. Assessment at additional or fewer time points may be performed based on emerging data. Details regarding sample collection and processing are provided in the laboratory manual.

#### **9.3.1 Evaluation of AUTO3 Persistence in Peripheral Blood**

Blood samples (9-18 mL) will be used to measure the expansion/persistence of CD19/CD22 T cells at time points indicated in the Schedule of Assessments. Flow cytometry will be used to measure the frequency of CD19/CD22 CAR-positive T cells per microliter of whole blood and/or a qPCR assay/digital PCR assay will be used to quantify the number of copies of the CD19/CD22 CAR transgene per microgram of genomic DNA and/or per cell in peripheral blood. Please refer to the AUTO3-PA1 laboratory manual for the handling and storage of samples.

### 9.3.2 Evaluation of RCR in Peripheral Blood

As per health authorities' guidelines, tests will be performed to evaluate and monitor the presence of RCR by qPCR; 5 mL blood will be collected for this analysis. Confirmatory assays may be performed if the initial results are positive. Please refer to the AUTO3-PA1 laboratory manual for the handling and storages of samples.

### 9.3.3 Insertional Mutagenesis

Insertional mutagenesis leading to oncogenesis is a recognised safety concern of vector-based gene therapy. Samples will be stored and archived before and after treatment (per the Schedule of Assessments) with the intent to be analysed for insertional mutagenesis, should a patient develop a new malignancy. The result will allow establishing a potential relationship between AUTO3 treatment and the development of any new malignancy.

### 9.3.4 [REDACTED]

**Serum cytokine profile:** (minimum dataset of TNF- $\alpha$ , IFN- $\gamma$ , and IL-6) will be measured

[REDACTED]

[REDACTED]

[REDACTED]

[REDACTED]

[REDACTED]

[REDACTED]

### 9.3.5 [REDACTED]

[REDACTED]

## 9.4 EFFICACY EVALUATION

Response evaluations for the primary endpoint/final analysis will be based on the response criteria for ALL according to the National Comprehensive Cancer Network guidelines version 2.2014, listed in [Appendix 1](#). Efficacy will be assessed by determining morphological CR/CRi as well as molecular remission based on MRD in the BM aspirate using IgH qPCR and/or next generation sequencing and flow cytometry as outlined in the Schedule of Assessments in all patients. The proportion of patients achieving molecular remission at 1 month post AUTO3 infusion will be determined. The duration of response and EFS and RFS at 6, 12 and 24 months will be assessed. Relapse rate and proportion of patients in molecular remission without further therapy at 6 months, 1 and 2 years will be determined.

It is important that instances and evidence of disease progression be reported to the Sponsor as soon as possible. The Medical Monitor will review the data to confirm that the criteria for disease progression have been met. If the Medical Monitor concurs, the Investigator will be notified and the patient will be withdrawn from the study

### 9.4.1 Bone Marrow Aspiration and Biopsy

Bone marrow will be used for haematological assessment for morphological remission and for evaluation of MRD by IgH qPCR and/or next generation sequencing and flow cytometry. Trephine will be performed if the patient is pancytopenic according to Investigator clinical judgment. Morphological and cytogenetic analysis of the BM will be done by the local laboratory, but MRD analysis will be performed at Bristol Genetics Laboratory or other designated laboratories. Bone marrow aspirates will be performed (usually under general anaesthetic) as outlined in the Schedule of Assessments. Details regarding sample collection and processing are provided in the laboratory manual. The first screening (morphology and cytogenetics) assessment can be performed at the referring institution and reports should be available. In general, BM samples will be analysed as below and may be stored for later analysis.

**Morphology:** Bone marrow smears (slides), will be analysed to determine the degree of leukaemic infiltration (% blasts) and the recovery of normal haemopoiesis.

**Leukaemia Immunophenotype:** The B-precursor phenotype should be confirmed at screening by flow cytometry and should include CD19 and CD22 markers as well as development of a leukaemia-associated immunophenotype (LAIP) for subsequent assessment. In patients who relapse post AUTO3, the expression of CD19 and CD22 on the leukaemic blasts will also be assessed.

**Minimal Residual Disease (by qPCR & Flow cytometry):** A sample of BM aspirate should be sent for IgH qPCR and/or next generation sequencing at the Molecular Haematology laboratory at Bristol Genetics Laboratory or other designated laboratories. Flow cytometric MRD assessment will also be done.

**Cytogenetics:** Known leukaemia associated cytogenetic and molecular aberrations will be documented in the eCRF at baseline and at disease progression. Where an abnormality is detected, this may be optionally assessed on follow-up BM samples post AUTO3 infusion.

#### 9.4.2 Treatment Response

The treatment is defined to be efficacious, when the patient is stated to be in morphological remission (CR or in CRi) and has MRD-negative response. Responses will be assessed within 30 days ( $\pm 3$  days) post first AUTO3 infusion. The morphological treatment response criteria are defined in [Appendix 1](#). The MRD will be defined as:

##### **Minimal Residual Disease Negative Response:**

Minimal residual disease negative response is achieved if MRD is  $<10^{-4}$  (0.01%) by PCR amplification of individual rearrangements of Ig genes and/or flow cytometry MRD testing (next generation sequencing may also be carried out).

##### **Minimal Residual Disease CR:**

Minimal residual disease CR is achieved if no PCR amplification of individual rearrangements of Ig genes can be detected using an appropriately sensitive assay (generally  $<5 \times 10^{-5}$  copies). Minimal residual disease CR is achieved if blast count is  $<0.01\%$  by flow cytometry or qPCR.

##### **Progressive Disease:**

Progressive disease is defined as an increase of  $\geq 25\%$  in the absolute number of circulating or BM blasts or development of extramedullary disease.

##### **Disease Relapse:**

Morphological relapse is defined as patients who achieved a CR or CRi and who have reappearance of blasts in the blood ( $\geq 1\%$ ), or reappearance of blasts in BM ( $\geq 5\%$ ), or reappearance of any extramedullary disease after CR or CRi ([Appendix 1](#)).

Molecular/flow MRD relapse is defined as patients who achieved a MRD negative CR by PCR or flow and who now have disease detectable by qPCR and flow cytometry ( $>0.01\%$ ). Patients with qPCR only relapse may be followed until they have flow cytometry and morphological relapse if clinically appropriate. This will help evaluate the reasons for the relapse such as loss of CD19 and or CD22 or PD-L1 expression. The proportion of patients in morphological and molecular CR without further treatment at 6 months and 1 and 2 years post AUTO3 infusion will be determined.

#### 9.4.3 Documentation of CNS/Extramedullary Disease

All patients will be assessed for CNS disease by lumbar puncture prior to lymphodepletion, on Day 0 and Day 30 and 2 years ( $\pm 14$  days) after AUTO3 infusion or if clinically indicated. In patients with extramedullary disease without detectable BM disease at the time of AUTO3 infusion (chiefly patients with isolated CNS relapse in Phase II of the study), other restaging investigations e.g. additional lumbar punctures and imaging will be performed as clinically indicated.

Complete remission is achieved when no lymphoblasts are detected in CSF by cytopspin or flow cytometry ([Appendix 1](#)).

## **9.5 PATIENT REPORTED OUTCOMES**

Health-related quality of life (HRQOL) measures may be collected at periodic intervals (paper or electronic) prior to AUTO3 infusion and then at 1, 3, 6, 8, 12 and 24 months, and as necessary. The instruments may include the Pediatric Quality of Life Inventory (PedsQL) which is a brief, standardised, generic assessment instrument that systematically assesses patients' and parents' perceptions of HRQOL in pediatric patients. Additional instruments such as the EQ-5D, which is a standardised instrument developed by the EuroQol Group as a measure of health-related quality of life, will also be used, as age appropriate.

## **9.6 BLOOD VOLUME COLLECTIONS**

In general, blood will be taken from a Hickman line or other central venous access device for the first year after AUTO3 infusion so that venepuncture will not be needed. If blood is taken peripherally at later time points, patients will be offered local anaesthetic cream/spray. The maximum volume of blood collected for study related assessments on any 1 day will not exceed 80 mL, or 2 mL/kg in patients less than 30 kg. In patients under 40 kg, the blood collection may be spread over 2-3 days. Additional samples may be collected as required to ensure the safety of the patient. Refer to the laboratory manual for the handling and storage of samples.

## 10 GUIDELINES FOR PREVENTION, MONITORING, AND MANAGEMENT OF ADVERSE EVENTS

The guidelines detailed in [Table 12](#) are suggested ways of managing key toxicities, however deviations from suggested recommendations are allowed per the Investigator’s judgement and local institutional practice.

### 10.1 GENERAL SUPPORTIVE CARE GUIDELINES FOR PATIENTS RECEIVING CAR T CELL THERAPY

The recommendations should be modified appropriately to suit patients’ age and weight.

**Table 12: General Supportive Care Guidelines for Patients Receiving CAR T Cell Therapy**

Toxicity	Preventive and Supportive Care Interventions
Prophylaxis	<p>Patients should receive pneumocystis prophylaxis with trimethoprim-sulfamethoxazole or suitable alternative agents, and either acyclovir or valacyclovir for herpes virus prophylaxis from the start of conditioning chemotherapy until 6 months post AUTO3 infusion.</p> <p>All patients should be monitored for CMV by PCR weekly during admission or as necessary.</p> <p>Additional anti-microbial (e.g. ciprofloxacin) and anti-fungal prophylaxis to be given as per institutional practice.</p> <p>All patients with fevers while neutropenic must have blood cultures drawn and receive broad-spectrum antibiotics as per institutional practice.</p>
Constitutional	Administer paracetamol for symptomatic management of fevers in patients;
	Avoid corticosteroids and if thrombocytopenic non-steroidal anti-inflammatory drugs.
Respiratory	Monitor for oxygen saturation at every visit, a significant decrease in oxygen saturation at room air should be investigated and managed with supportive care including supplemental oxygen, anti-microbials and ventilator support as appropriate. Patients with an oxygen requirement of >40% (or lower based on emerging data) in setting of CRS should receive treatment with Tocilizumab
Cardiovascular	Stop or taper antihypertensive medications prior to cell infusion;
	Monitor vital signs at least every 4-8 hours on an inpatient unit during hospital stay;
	Monitor vital signs every 2 hours in patients with fevers and tachycardia;
	Initiate replacement i.v. fluids for patients with poor oral intake or high insensible losses to maintain net even fluid balance;
	Administer i.v. fluid boluses for patients with systolic blood pressure less than 80% of their pre-infusion baseline and poor peripheral perfusion. A 20 mL/kg normal saline bolus (500 mL if age ≥16) should be given initially and the patient reassessed after 30 minutes. Consider a further bolus of 20 mL/kg (500 mL if age ≥16) if blood pressure remains low and ongoing poor perfusion.
	In patients receiving >2 i.v. fluid bolus (or as age appropriate) for hypotension, consider transfer to the intensive care unit, an ECG and an ECHO/MUGA should be performed to evaluate for cardiac toxicity.
	Noradrenaline is the preferred first-line vasopressor for patients with hypotension initiated on inotropic support.

Toxicity	Preventive and Supportive Care Interventions
Hematologic	Initiate allopurinol and i.v. fluid prophylaxis for TLS prophylaxis at start of lymphodepletion in patients with high leukaemic burden ( $\geq 25\%$ BM blasts); (see <a href="#">Section 10.5</a> )
	Monitor complete blood count with differential at least daily.
	Maintain haemoglobin of $\geq 80$ g/L;
	Maintain platelets $\geq 20,000/\mu\text{L}$ ;
	If absolute neutrophil count $< 500/\mu\text{L}$ at 30 days ( $\pm 3$ days) post AUTO3 infusion (or earlier if clinically indicated), initiate granulocyte colony stimulating factor support. Continue until absolute neutrophil count increases to $\geq 1000 \mu\text{L}$ ;
	Transfuse fresh frozen plasma with a goal of normalisation of partial thromboplastin time (PTT) in patients with a PTT $> 1.5$ -fold above the ULN; and
	Transfuse cryoprecipitate to maintain fibrinogen of $\geq 1$ g/L. If patient is bleeding, a higher level of fibrinogen should be maintained.
Neurologic	The nursing staff conducts focused neurologic examinations every 4-6 hours in patients experiencing neurologic toxicity;
	Perform brain magnetic resonance imaging (MRI) in any patient experiencing neurologic toxicity;
	Perform lumbar puncture to evaluate for infectious pathogens, cytokine levels, and CAR T cell levels in patients experiencing neurologic toxicity whenever feasible;
	Consider a neurology consultation for any patient experiencing neurologic toxicity; and
	Standard antiepileptic medications for patients with active seizures. Prophylactic antiepileptic medications are not required.

BM = bone marrow; CAR = chimeric antigen receptor; ECG = electrocardiogram; ECHO = echocardiogram; i.v. = intravenous; MRI = magnetic resonance imaging; PTT = partial thromboplastin time; TLS = tumour lysis syndrome; ULN = upper limit of normal.  
Taken from ([Brudno and Kochenderfer 2016](#)).

## 10.2 PRE- AND POST-INFUSION SUPPORTIVE THERAPY

AUTO3 is an autologous product, with fully human CAR constructs and is less likely to be immunogenic and induce an infusion or hypersensitivity reaction. However, the following medications should be given 30 minutes before the study drug infusion: paracetamol orally and an antihistamine (chlorpheniramine or equivalent) i.v. These medications may be discontinued based on emerging data. In addition, pre-infusion medications listed in [Table 13](#) may also be administered if necessary.

Post-infusion medication listed in [Table 13](#) may be considered following AUTO3 infusion if necessary. Post-infusion medication(s) may be continued for up to 48 hours after the infusion. Use of additional supportive care measures may be instituted as clinically necessary at the discretion of the Investigator.

**Table 13: Pre- and Post-infusion Medications (Dose Adjusted as Age Appropriate)**

Medication	Dose	Administration	Pre-infusion	Post-infusion
Antihistamine	Chlorpheniramine (age <5 years, 2.5 mg; age 6-11, 5 mg; age ≥12, 10 mg, 6 hourly)	I.v. – administer at least 30 minutes prior to study drug	X	Optional
Antipyretic	Paracetamol (age ≥12 years, 1000 mg; age ≤11 years, 15 mg/kg, 6 hourly)	Oral - administer at least 30 minutes prior to study drug	X	Optional as clinically indicated
Antiemetic	Ondansetron (age ≤17 years, 150 µg/kg maximum 8 mg; age ≥18 years, 8 mg or equivalent)	I.v. - start infusion 30 minutes prior to study drug	Optional	N/A
	Ondansetron (weight <11 kg, 2 mg; weight 11-40 kg, 4 mg; weight >40 kg, 8 mg, twice daily) or equivalent	Oral - as clinically indicated	N/A	Optional

i.v. = intravenous; N/A = not appropriate

**Note:** Steroids should in general be avoided but may be used in case of severe reactions not controlled by other measures.

### 10.3 TREATMENT OF INFUSION RELATED REACTIONS

Patients who experience infusion-related reactions (to pre-conditioning or AUTO3) that manifest as wheezing, flushing, hypoxemia, fever, chills, rigors, bronchospasm, headache, rash, pruritus, arthralgia, hypo- or hypertension or other symptoms, should have the symptoms managed according to the recommendations provided in [Table 14](#) or as per institutional practice. All NCI CTCAE Grade 3 or 4 infusion-related reactions should be reported within 24 hours to the Sponsor. If the event meets the criteria of an SAE, follow SAE reporting criteria in [Section 12.3.2](#).

**Table 14: Guidelines for the Management of Infusion-related Reactions (Dose Adjusted as Age Appropriate)**

NCI CTCAE Grade	Treatment/Intervention	Premedication at Subsequent Dosing (if patient receiving split dosing)
<p><b>Grade 1</b> Mild reaction; infusion interruption not indicated; intervention not indicated</p>	<p><b>No intervention indicated</b> Monitor patient as medically indicated until recovery from symptoms.</p>	<p>May consider chlorpheniramine (age &lt;5 years, 2.5 mg; age 6-11, 5 mg; age ≥12, 10 mg 6 hourly) or equivalent and/or paracetamol (age ≥12 years, 1000 mg; age ≤11 years, 15 mg/kg 6 hourly) orally.</p>
<p><b>Grade 2</b> Moderate reaction; requires therapy or infusion interruption but responds promptly to symptomatic treatment</p>	<p><b>Interrupt infusion</b> Start i.v. fluids; give chlorpheniramine (age &lt;5 years, 2.5 mg; age 6-11, 5 mg; age ≥12, 10 mg 6 hourly) (or equivalent) i.v. and/or paracetamol; consider bronchodilator therapy; may also consider corticosteroids if necessary; monitor patient closely until recovery from symptoms.  Restart infusion if AUTO3 dose has not been fully administered.  <b>Symptoms recur:</b> Stop and discontinue further infusion.  The amount of AUTO3 infused must be recorded on the eCRF.</p>	<p>Chlorpheniramine (age &lt;5 years, 2.5 mg; age 6-11, 5 mg; age ≥12, 10 mg 6 hourly) or equivalent and/or paracetamol (age ≥12 years, 1000 mg; age ≤11 years, 15 mg/kg 6 hourly) orally.</p>
<p><b>Grade 3 or 4</b> Severe reaction;</p> <p><b>Grade 3:</b> prolonged (i.e. not rapidly responsive to symptomatic medication and/or brief interruption of infusion); recurrence of symptoms following initial improvement; hospitalisation indicated for other clinical sequelae (e.g. renal impairment, pulmonary infiltrates)</p> <p><b>Grade 4:</b> life-threatening; pressor or ventilator support indicated</p>	<p><b>Stop infusion</b> Start i.v. saline infusion; recommend bronchodilators and chlorpheniramine (age &lt;5 years, 2.5 mg; age 6-11, 5 mg; age ≥12, 10 mg 6 hourly) i.v. +/- adrenaline 1 µg/kg for patients &lt;50 kg and 50 µg for patients over 50 kg, injected slowly for i.v. administration or administer intramuscularly. If unresponsive, give with methylprednisolone 1 mg/kg (max 80 mg) i.v. (or equivalent), as needed, and other drugs as appropriate.  Patients should be monitored until the Investigator is comfortable that the symptoms will not recur. Investigators should follow their institutional guidelines for the treatment of anaphylaxis. In the case of late-occurring hypersensitivity symptoms (e.g. appearance of a localised or generalised pruritus within 1 week after treatment), symptomatic treatment may be given (e.g. oral antihistamine or corticosteroids), as appropriate.</p>	<p><b>Grade 3:</b> Discuss with Sponsor's Medical Monitor about merits of completing AUTO3 infusion if full dose has not been administered.</p> <p><b>Grade 4:</b> Permanently discontinue any remaining AUTO3.</p>
<p><b>General</b></p>	<p>Appropriate personnel and appropriate resuscitation equipment should be available in or near the infusion room and a physician should be readily available during the infusion of study drug.</p>	

CTCAE = Common Terminology Criteria for Adverse Events; eCRF = Electronic Case Report Form; i.v. = intravenous; NCI = National Cancer Institute.

#### **10.4 GRADING AND MANAGEMENT OF CYTOKINE RELEASE SYNDROME**

Cytokine release syndrome is a known toxicity with CAR T cell therapies and can be severe in 20 to 30% of patients, particularly those with high leukaemic burden. Clinical symptoms indicative of CRS may include, but are not limited to, culture negative fever (with or without rigors), myalgia, nausea/vomiting, tachycardia, hypoxia, hypotension, headache, confusion, tremor, and delirium. Potentially life-threatening complications of CRS may include cardiac dysfunction, acute respiratory distress syndrome, renal and/or hepatic failure, and disseminated intravascular coagulation (DIC). DIC, macrophage activation syndrome (MAS) and hemophagocytic lymphohistiocytosis may occur in some for whom CAR-mediated inflammatory responses continue to evolve. The clinical syndrome of MAS is characterised by high grade non-remitting fever, cytopenias affecting at least two of three lineages, and hepatosplenomegaly. It is associated with biochemical abnormalities, such as high circulating levels of serum ferritin, soluble interleukin-2 receptor (sCD25), and triglycerides, together with a decrease of circulating NK activity. Other findings include variable levels of transaminases up to signs of acute liver failure and coagulopathy with findings consistent with DIC.

Patients will be admitted as an inpatient for a minimum of 30 days ( $\pm 3$  days) (longer if necessary for monitoring or management) after first AUTO3 infusion and will be closely monitored for early signs and symptoms indicative of CRS with clinical review and blood tests including C-reactive protein/clotting as well as 3 times weekly monitoring of serum cytokines for the first 14 days or longer if clinically indicated. Clinical personnel will be trained in the management of CRS. Resources necessary for resuscitation and supportive care including intensive care should be readily available. In the event of severe CRS, as major paediatric/adolescent haematology centres, all sites have extensive experience in resuscitation and an intensive care unit on site. Recommendations for the clinical management of CRS are provided in [Table 15](#).

For the purposes of capture in the eCRF, as CRS can present with multiple signs and symptoms, it is important to assess the character and nature of these occurrences. Therefore, avoid entering events of 'Cytokine Release Syndrome' in the eCRF. Please add the associated adverse events and indicate their association with CRS, as indicated in the CRF.

**Table 15A: Severity Grading of Cytokine Release Syndrome (ASBMT CRS Consensus Grading)**

CRS Parameter	Grade 1	Grade 2	Grade 3	Grade 4
<b>Fever<sup>†</sup></b>	Temperature $\geq 38^{\circ}\text{C}$	Temperature $\geq 38^{\circ}\text{C}$	Temperature $\geq 38^{\circ}\text{C}$	Temperature $\geq 38^{\circ}\text{C}$
<b>With either:</b>				
<b>Hypotension</b>	None	Not requiring vasopressors	Requiring one vasopressor with or without vasopressin	Requiring multiple vasopressors (excluding vasopressin)
<b>And/or<sup>‡</sup></b>				
<b>Hypoxia</b>	None	Requiring low-flow nasal cannula <sup>^</sup> or blow-by	Requiring high-flow nasal cannula <sup>^</sup> , facemask, non-rebreather mask, or Venturi mask	Requiring positive pressure (eg: CPAP, BiPAP, intubation and mechanical ventilation)

Organ toxicities associated with CRS may be graded according to CTCAE v5.0 but they do not influence CRS grading.

CPAP: Continuous positive airway pressure; BiPAP: Bilevel positive airway pressure

<sup>†</sup> Fever is defined as temperature  $\geq 38^{\circ}\text{C}$  not attributable to any other cause. In patients who have CRS then receive antipyretics or anti-cytokine therapy such as tocilizumab or steroids, fever is no longer required to grade subsequent CRS severity. In this case, CRS grading is driven by hypotension and/or hypoxia.

<sup>‡</sup> CRS grade is determined by the more severe event: hypotension or hypoxia not attributable to any other cause. For example, a patient with temperature of  $39.5^{\circ}\text{C}$ , hypotension requiring one vasopressor and hypoxia requiring low-flow nasal cannula is classified as having Grade 3 CRS.

<sup>^</sup> Low-flow nasal cannula is defined as oxygen delivered at  $\leq 6$  L/minute. Low flow also includes blow-by oxygen delivery, sometimes used in pediatrics. High-flow nasal cannula is defined as oxygen delivered at  $> 6$  L/minute.

Taken from (Lee et al. 2018).

**Table 15B: Management of Cytokine Release Syndrome (Dose Adjusted as Age Appropriate)**

CRS Grade	Treatment
<p><b>Grade 1</b> Symptoms are not life-threatening and require symptomatic treatment only, e.g. fever, nausea, fatigue, headache, myalgia, and malaise or organ toxicity.</p>	<p>Supportive care per institutional standards including analgesics and antipyretics, assess and treat for neutropenic infections. Consider tocilizumab or siltuximab for persistent (lasting &gt;3 days) and refractory fever.</p>
<p><b>Grade 2</b> Symptoms require and respond to moderate intervention</p>	<p>Supportive care including fluid substitution is recommended. Use tocilizumab (or siltuximab) early if persistent fever of <math>\geq 39^{\circ}\text{C}</math> despite antipyretics for 10 hours, persistent/ recurrent hypotension after initial fluid bolus, and initiation of oxygen supplementation. Low-flow-oxygen (nasal cannula or blowby). Use dexamethasone if hypotension persists after 1 to 2 doses of anti-IL-6 therapy.</p>
<p><b>Grade 3</b> Symptoms require and respond to aggressive intervention</p>	<p>Intensive care should be considered. Immunotherapy with tocilizumab (or siltuximab). Repeat dose of tocilizumab at 6-12 hours if no clinical improvement. Add siltuximab as necessary if not previously administered. Add steroids if unresponsive within 24 hours. Also, consider anti-TNF antibodies as clinically appropriate. Low-dose vasopressor therapy. Oxygen (flow <math>\geq 40\%</math> fraction of inspired oxygen) (high flow oxygen through nasal cannula oxygen or facemask, continuous positive airway pressure or bilevel positive airway pressure). Uncontrolled MAS or haemophagocytosis may also be treated with anakinra, an IL-1 receptor antagonist, as a steroid-sparing agent at a dose of 100 mg by subcutaneous injection, to be repeated daily as clinically indicated (<a href="#">Shah et al. 2017</a>).</p>
<p><b>Grade 4</b> Life-threatening symptoms</p>	<p>Intensive care. Ventilatory support requiring positive pressure (CPAP/BIPAP/mechanical ventilation) Multiple vasopressors or high-dose vasopressors. Immunotherapy with tocilizumab, <math>\pm</math> siltuximab, and high dose steroids. Consider alternative agents such as anti-TNF, and other agents as appropriate (<a href="#">Kenderian et al. , Gargett et al. 2015, Ruella et al. 2016</a>). Uncontrolled MAS or haemophagocytosis may also be treated with anakinra, an IL-1 receptor antagonist, as a steroid-sparing agent at a dose of 100 mg by subcutaneous injection, to be repeated daily as clinically indicated (<a href="#">Shah et al. 2017</a>).</p>
<p><b>Grade 5</b> Death</p>	

CRS = cytokine release syndrome; CTCAE = Common Terminology Criteria for Adverse Events; NCI = National Cancer Institute; TNF = tumour necrosis factor.  
Taken from ([Lee et al. 2018](#)).

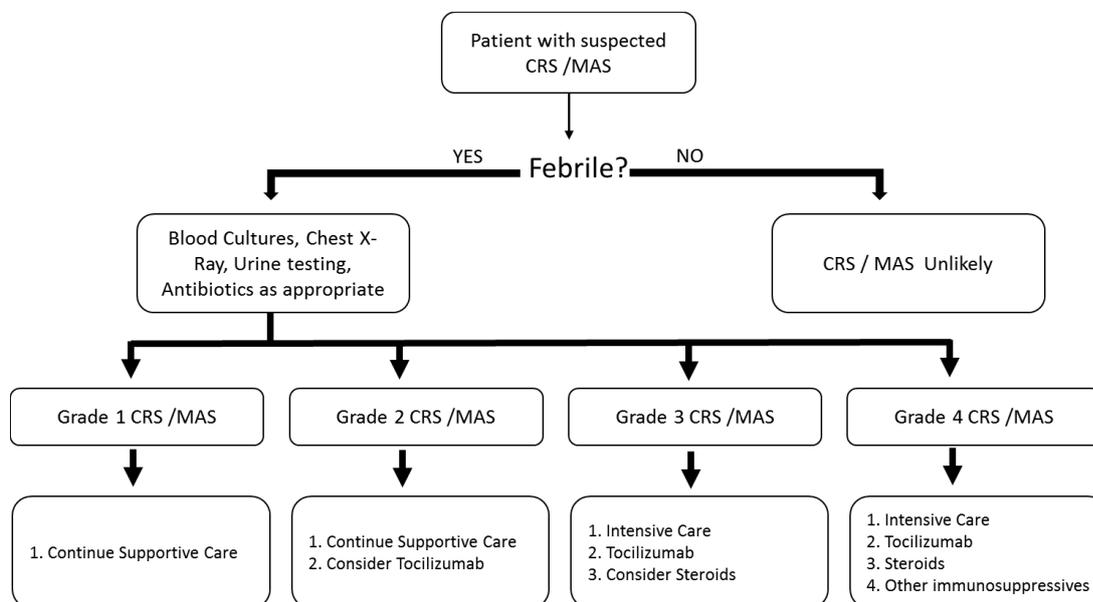
Details of supportive medication and steroid doses are presented in [Table 16](#) and [Table 17](#).

**Table 16: Definitions and Doses of Vasopressors (Dose Adjusted as Age Appropriate)**

Definition of High-Dose Vasopressors	
Vasopressor	Dose for $\geq 3$ hours
Noradrenaline monotherapy	$\geq 0.2$ mcg/kg/minute
Dopamine monotherapy	$\geq 10$ mcg/kg/minute
Adrenaline monotherapy	$\geq 0.1$ mcg/kg/minute
If on vasopressin	High dose of vasopressin + noradrenaline equivalent of $\geq 0.1$ mcg/kg/minute (using Vasopressin and Septic Shock Trial formula)
If on combination vasopressors (not vasopressin)	Noradrenaline equivalent of $\geq 20$ mcg/minute

An overview of the CRS management is presented in [Figure 5](#).

**Figure 5: CRS Management Overview**



The pharmacologic management of CRS is presented in [Table 17](#).

**Table 17: Pharmacologic Management of CRS/MAS (Dose Adjusted as Age Appropriate)**

Drug	Indication	Dose
<p>Tocilizumab</p> <p>Siltuximab Siltuximab is an alternative to tocilizumab and may be used according to local institutional guidance. Siltuximab has been approved to treat Castleman's disease (<a href="#">Grupp et al. 2013</a>, <a href="#">Calabrese and Rose-John 2014</a>, <a href="#">Maude et al. 2014a</a>, <a href="#">Teachey et al. 2018</a>), but not currently for CRS.</p> <p>Siltuximab may be added if there is no response to tocilizumab+/- corticosteroids.</p>	<p>LVEF &lt;40% by ECHO; Creatinine &gt;2.5-fold higher than the most recent level prior to CAR T cell infusion; Norepinephrine requirement at a dose &gt;2 µg/minute for 48 hours since the first administration of norepinephrine, even if administration is not continuous; Systolic blood pressure of 90 mm Hg that cannot be maintained with norepinephrine; Oxygen requirement of fraction of inspired oxygen &gt;50% or more for more than 2 hours continuously; Dyspnoea that is severe enough to potentially require mechanical ventilation; Activated PTT &gt;2 × the ULN; Clinically-significant bleeding; and Creatine kinase &gt;5 × the ULN for longer than 2 days.</p> <p><b>Early use criteria:</b> May consider using for persistent fever of ≥39°C despite antipyretics for 10 hours, persistent/ recurrent hypotension after initial fluid bolus, and initiation of oxygen supplementation</p>	<p>Tocilizumab Weight &gt;30 kg, 8 mg/kg; weight &lt;30 kg, 12 mg/kg given i.v. over 1 hour.</p> <p>NB: organ dysfunction secondary to CRS and cytopenias due to disease/chemotherapy will not constitute a contraindication to tocilizumab.</p> <p>Repeat dose if no response within 8 hours and consider steroids as below if unresponsive at 24 hours. Refer to local prescribing information and institutional guidance.</p> <p>Siltuximab 11mg/kg i.v. over 1 hour as an intravenous infusion as a single dose (<a href="#">Grupp et al. 2013</a>, <a href="#">Shah et al. 2017</a>).</p>
Methylprednisolone	CRS toxicity refractory to tocilizumab.	2 mg/kg i.v. every 12 hours weaned over 5 days.
Anakinra	Uncontrolled CRS or haemophagocytosis or Macrophage activation syndrome (MAS) and or Disseminated intravascular coagulation (DIC) refractory to standard measures ( <a href="#">Brudno and Kochenderfer 2016</a> , <a href="#">Shah et al. 2017</a> , <a href="#">Liu and Zhao 2018</a> ).	100 mg (or as age appropriate) by subcutaneous injection, to be repeated daily as clinically indicated.

CAR = chimeric antigen receptor; CRS = cytokine release syndrome; ECHO = echocardiogram; i.v. = intravenous; LVEF = left ventricular ejection fraction; PTT = partial thromboplastin time; ULN = upper limit of normal.  
Reference: ([Brudno and Kochenderfer 2016](#)); Tocilizumab (Actemra) label ([FDA 2018b](#)).

## 10.5 MANAGEMENT OF TUMOUR LYSIS SYNDROME

Treatment with CAR T cells, such as AUTO3, can lead to rapid killing of malignant B cells, which can occasionally result in TLS associated with a release of intracellular ions and metabolic by-products into the systemic circulation in patients with a high leukaemic burden. Clinically, TLS can be characterised by rapid development of hyperuricemia, hyperkalaemia, hyperphosphatemia, hypocalcaemia, and potentially acute renal failure.

Patients with a high leukaemic burden ( $\geq 25\%$  BM blasts) will be given allopurinol and i.v. fluids from the start of lymphodepletion to prevent TLS. Patients will be monitored vigilantly for early signs and symptoms of TLS with daily clinical review and blood biochemistry. Should TLS occur, supportive care will be given as per standard institutional protocols. Management of TLS will include i.v. fluids, aggressive correction of abnormal laboratory test results such as hyperkalaemia, hyperuricemia, and hypocalcaemia, together with rasburicase to increase urate excretion if needed.

## 10.6 GRADING AND MANAGEMENT OF NEUROTOXICITY (ICANS)

Neurotoxicity has been seen in patients with leukaemia and lymphoma after treatment with CD19 CAR T cells and is now referred to as ‘Immune Effector Cell-associated Neurotoxicity Syndrome’ (ICANS). The cause of neurotoxicity is not well-understood, although it is generally reported to be fully reversible. Transient neurological complications have also been reported with CD19 bispecific T cell engagers, suggesting that the target may have some relevance. In neither case do these toxicities correlate with CRS/macrophage activation syndrome. However, close attention should be paid to neurological signs and symptoms.

Although symptoms can vary, the early manifestations of ICANS are often tremor, dysgraphia, mild difficulty with expressive speech especially naming objects, impaired attention, apraxia, and mild lethargy. Other symptoms can include confusion, depressed level of consciousness/ encephalopathy, hallucinations, dysphasia, ataxia, apraxia, cranial nerve palsies, and seizures. Headache is a non-specific symptom, frequently occurring during fever or after chemotherapy, thus, headache alone is not a useful marker of ICANS. Expressive aphasia, on the other hand, appears to be a very specific symptom of ICANS.

There appears to be no correlation between ICANS and CRS/macrophage activation syndrome and both can occur together or separately.

Patients will be admitted as an inpatient for a minimum of 30 days ( $\pm 3$  days) (longer if necessary for monitoring or management) after first AUTO3 infusion and closely monitored for neurological signs and symptoms of neurotoxicity. If neurotoxicity is observed, a neurology opinion should be sought, MRI imaging performed and patients will receive supportive care (e.g. anti-convulsants), as appropriate. In general, neurotoxicity is transient and resolves spontaneously with no long-term sequelae so that supportive care alone is sufficient in most patients. Steroids may be given for Grade 3 or 4 neurotoxicity or in the case of cerebral oedema or generalised seizures but otherwise treatment with steroids should be avoided as this may be deleterious to the persistence of AUTO3.

Neurotoxicity may also be caused by FLU but generally at higher doses (Helton et al. 2013) than those being administered in this protocol. Symptoms including objective weakness, agitation, confusion, seizures, visual disturbances, optic neuritis, optic neuropathy, blindness, and coma have been reported in CLL patients treated with multiple cycles of FLU. As described in Section 4.3, death of three young adult ALL patients due to cerebral oedema was partly attributed to FLU; however, two additional deaths due to cerebral oedema in the absence of FLU indicates that was not the cause. Moreover, it now appears that cerebral oedema is likely a consequence of very rapid CAR T cell proliferation, driven by the CD28 co-stimulatory domain, this event has not been reported with CARs containing a 41BB- $\zeta$  co-stimulatory domain which results in a slower proliferation. Patients should be closely monitored and managed as appropriate. Please see Table 18 for suggested assessment and grading of neurotoxicity as per the ASBMT guidelines for Immune effector Cell-Associated Neurotoxicity Syndrome (ICANS) (Lee et al. 2018). Patients may be managed as per the

suggested guidelines below but may also be managed as per institutional management guidelines.

**Table 18: Assessment & Grading of Neurological AEs (ASBMT ICANS Consensus Grading for Adults (Lee et al. 2018))**

Neurotoxicity Domain	Grade 1	Grade 2	Grade 3	Grade 4
<b>ICE Score for children &gt;12 years<sup>^</sup></b> (see scale below)	7-9	3-6	0-2	0 (patient is unarousable and unable to perform ICE)
<b>CAPD score for children ≤12 years</b> (see scale below)	<9	<9	≥9	Unable to perform CAPD
<b>Depressed level of consciousness*</b>	Awakens spontaneously	Awakens to voice	Awakens only to tactile stimulus	Patient is unarousable or requires vigorous or repetitive tactile stimuli to arouse; stupor or coma
<b>Seizure (any age)</b>	N/A	N/A	Any clinical seizure focal or generalized that resolves rapidly; or Non-convulsive seizures on EEG that resolve with intervention	Life-threatening prolonged seizure (>5 min); or Repetitive clinical or electrical seizures without return to baseline in between.
<b>Motor weakness (any age)<sup>§</sup></b>	N/A	N/A	N/A	Deep focal motor weakness such as hemiparesis or paraparesis
<b>Raised ICP / Cerebral Edema (any age)</b>			Focal/local edema on neuroimaging <sup>#</sup>	Decerebrate or decorticate posturing; or Cranial nerve VI palsy; or Papilledema; or Cushing's triad; or Signs of diffuse cerebral edema on neuroimaging

ICANS grade is determined by the most severe event (ICE or CAPD score, level of consciousness, seizure, motor findings, raised ICP/cerebral edema) not attributable to any other cause.

<sup>^</sup>A patient with an ICE score of 0 may be classified as having Grade 3 ICANS if the patient is awake with global aphasia. But a patient with an ICE score of 0 may be classified as having Grade 4 ICANS if the patient is unarousable.

\*Depressed level of consciousness should be attributable to no other cause (e.g. no sedating medication)

<sup>§</sup> Tremors and myoclonus associated with immune effector cell therapies may be graded according to CTCAE v5.0 but they do not influence ICANS grading.

<sup>#</sup> Intracranial hemorrhage with or without associated edema is not considered a neurotoxicity feature and is excluded from ICANS grading. It may be graded according to CTCAE v5.0.

ICE: Immune effector Cell-associated Encephalopathy; CAPD: Cornell Assessment of Pediatric Delirium; ICP: Intracranial pressure; EEG: electroencephalogram

Taken from (Lee et al. 2018)

### **Encephalopathy Assessment for Children ≥12 (Encephalopathy Assessment Tools for Grading of Immune effector Cell-Associated Neurotoxicity Syndrome (ICANS))**

#### **Immune effector Cell-associated Encephalopathy (ICE)**

Orientation: orientation to year, month, city, hospital: 4 points

Naming: ability to name 3 objects (eg, point to clock, pen, button): 3 points

Following commands: ability to follow simple commands (eg, “Show me 2 fingers” or “Close your eyes and stick out your tongue”): 1 point

Writing: ability to write a standard sentence (eg, “Our national bird is the bald eagle”): 1 point

Attention: ability to count backwards from 100 by 10: 1 point

**Scoring**

10, no impairment;

7-9, grade 1 ICANS;

3-6, grade 2 ICANS;

0-2, grade 3 ICANS;

0 due to patient unarousable and unable to perform ICE assessment, grade 4 ICANS

**Encephalopathy Assessment for Children <12 years using Cornell Assessment of Pediatric Delirium (CAPD) (Traube et al. 2014).**

Answer the following based on interactions with the child over the course of the shift	Never 4	Rarely 3	Sometimes 2	Often 1	Always 0
1. Does the child make eye contact with the caregiver?					
2. Are the child’s actions purposeful?					
3. Is the child aware of his/her surroundings?					
4. Does the child communicate needs and wants?					
5. Is the child restless?					
6. Is the child inconsolable?					
7. Is the child underactive – very little movement while awake?					
8. Does it take the child a long time to respond to interactions?					

For patients age 1-2 years, the following serve as guidelines to the corresponding questions:

1. Holds gaze. Prefers primary parent. Looks at speaker.
2. Reaches and manipulates objects, tries to change position, if mobile may try to get up
3. Prefers primary parent, upset when separated from preferred caregivers. Comforted by familiar objects (i.e., blanket or stuffed animal)
4. Uses single words or signs
5. No sustained calm state
6. Not soothed by usual comforting actions, for example, singing, holding, talking, and reading
7. Little if any play, efforts to sit up, pull up, and if mobile crawl or walk around
8. Not following simple directions. If verbal, not engaging in simple dialogue with words or jargon

**Table 18A: Management and Follow-up of Neurological AEs and CRES**

<b>Management and Follow-up of Neurological AEs.</b>	
<b>Grade 1</b>	<p>Monitor closely. Treat symptomatically.</p> <p>Vigilant supportive care; aspiration precautions; IV hydration</p> <p>Withhold oral intake of food, medicines, and fluids, and assess swallowing</p> <p>Convert all oral medications and/or nutrition to IV if swallowing is impaired</p> <p>Avoid medications that cause central nervous system depression</p> <p>Low doses of lorazepam (0.25 to 0.5 mg IV every 8 hours or as age appropriate) or haloperidol (0.5 mg IV every 6 hours as age appropriate) can be used, with careful monitoring, for agitated patients</p> <p>Neurology consultation</p> <p>Fundoscopy exam to assess for papilloedema</p> <p>MRI of the brain with and without contrast; diagnostic lumbar puncture with measurement of opening pressure; MRI spine if the patient has focal peripheral neurological deficits; CT scan of the brain can be performed if MRI of the brain is not feasible</p> <p>Daily 30 min EEG until toxicity symptoms resolve; if no seizures are detected on EEG.</p> <p>Consider levetiracetam 750 mg (as age appropriate) every 12 hours (oral or IV) for a month.</p> <p>If EEG shows non-convulsive status epilepticus, treat as per algorithm in <a href="#">Table 18B</a>.</p> <p>Consider anti-IL-6 therapy with tocilizumab or siltuximab, if neurotoxicity is associated with concurrent CRS (see <a href="#">Section 10.4</a>).</p> <p><b>Worsening:</b> treat as <math>\geq</math>Grade 2</p>
<b>Grade 2</b>	<p>Monitor closely. Supportive care and neurological work-up as indicated for Grade 1.</p> <p>Tocilizumab or siltuximab if associated with concurrent CRS.</p> <p>Dexamethasone 10 mg IV every 6 hours or methylprednisolone 1 mg/kg IV (as age appropriate) every 12 hours if refractory to anti-IL-6 therapy, or for neurotoxicity without concurrent CRS.</p> <p>Consider transferring patient to ICU if neurotoxicity is associated with Grade <math>\geq</math>2 CRS.</p> <p><b>Worsening:</b> treat as Grade 3 to 4</p>
<b>Grade 3</b>	<p>Supportive care and neurological work-up as indicated for Grade 1 if not done already.</p> <p>ICU transfer is recommended</p> <p>Anti-IL-6 therapy if associated with concurrent CRS, as described for Grade 2 neurotoxicity and if not administered previously.</p> <p>Dexamethasone 10 mg IV every 6 hours or methylprednisolone 1 mg/kg IV (as age appropriate) every 12 hours if refractory to anti-IL-6 therapy, or for neurotoxicity without concurrent CRS; continue corticosteroids until improvement to Grade 1 neurotoxicity and then taper.</p> <p>Anakinra, at a dose of 100 mg by subcutaneous injection, to be repeated daily as clinically indicated (<a href="#">Brudno and Kochenderfer 2016</a>, <a href="#">Norelli et al. 2016</a>, <a href="#">Liu and Zhao 2018</a>).</p> <p>Stage 1 or 2 papilloedema with CSF opening pressure <math>&lt;</math>20 mmHg should be treated as per algorithm presented in <a href="#">Table 18C</a>.</p> <p>Consider repeat neuroimaging (CT or MRI) every 2 to 3 days if patient has persistent Grade <math>\geq</math>3 neurotoxicity.</p>
<b>Grade 4</b>	<p>Supportive care and neurological work-up as outlined for Grade 1 neurotoxicity.</p> <p>ICU monitoring; consider mechanical ventilation for airway protection.</p> <p>Anti-IL-6 therapy and repeat neuroimaging as described for Grade 3 neurotoxicity.</p>

<b>Management and Follow-up of Neurological AEs.</b>	
	<p>High-dose corticosteroids continued until improvement to Grade 1 neurotoxicity and then taper; for example, methylprednisolone IV 1 g/day for 3 days, followed by rapid taper at 250 mg every 12 hours for 2 days, 125 mg every 12 hours for 2 days, and 60 mg every 12 hours for 2 days.</p> <p>Anakinra, at a dose of 100 mg by subcutaneous injection, to be repeated daily as clinically indicated (<a href="#">Brudno and Kochenderfer 2016</a>, <a href="#">Norelli et al. 2016</a>, <a href="#">Liu and Zhao 2018</a>).</p> <p>For convulsive status epilepticus, treat as per algorithm in <a href="#">Table 18B</a>.</p> <p>Stage <math>\geq 3</math> papilloedema, with a CSF opening pressure <math>\geq 20</math> mmHg or cerebral oedema, should be treated as per algorithm in <a href="#">Table 18C</a>.</p> <p><b>Worsening:</b> May consider use of lymphodepleting drugs such as cyclophosphamide (<a href="#">Garfall et al. 2015</a>) or other drugs (<a href="#">Klinger et al. 2016</a>) if unresponsive to standard immunosuppressive therapies.</p>
<b>General</b>	<p>Patients on IV steroids may be switched to an equivalent dose of oral corticosteroids (e.g. prednisone) at start of tapering or earlier, once sustained clinical improvement is observed.</p> <p>Prophylactic antibiotics or other antimicrobials as clinically appropriate.</p> <p>Rigorous control of blood pressure and electrolytes (particularly calcium and magnesium).</p>

AE=adverse event; CRS=cytokine release syndrome; CSF=cerebrospinal fluid; CT=computerised tomography; EEG=electroencephalogram; ICU=intensive-care unit; IV=intravenous; MRI=magnetic resonance imaging.

**Table 18B: Recommendations for the Management of Status Epilepticus After CAR T Cell Therapy**

<b>Event</b>	<b>Management</b>
Non-convulsive status epilepticus	<p>Assess airway, breathing, and circulation; check blood glucose.</p> <p>Lorazepam 0.5 mg IV, with additional 0.5 mg IV every 5 min, as needed, up to a total of 2 mg (as age appropriate) to control electrographical seizures.</p> <p>Levetiracetam 500 mg IV bolus (as age appropriate), as well as maintenance doses.</p> <p>If seizures persist, transfer to ICU and treat with phenobarbital loading dose of 60 mg IV (as age appropriate).</p> <p>Maintenance doses after resolution of non-convulsive status epilepticus are as follows: lorazepam 0.5 mg IV every 8 hours for three doses; levetiracetam 1000 mg IV every 12 hours; phenobarbital 30 mg IV every 12 hours (as age appropriate).</p>
Convulsive status epilepticus	<p>Assess airway, breathing, and circulation; check blood glucose.</p> <p>Transfer to ICU.</p> <p>Lorazepam 2 mg IV, with additional 2 mg IV to a total of 4 mg to control seizures (as age appropriate).</p> <p>Levetiracetam 500 mg IV bolus, as well as maintenance doses (as age appropriate).</p> <p>If seizures persist, add phenobarbital treatment at a loading dose of 15 mg/kg IV (as age appropriate).</p> <p>Maintenance doses after resolution of convulsive status epilepticus are: lorazepam 0.5 mg IV every 8 hours for three doses; levetiracetam 1000 mg IV every 12 hours; phenobarbital 1–3 mg/kg IV every 12 hours (as age appropriate).</p> <p>Continuous electroencephalogram monitoring should be performed, if seizures are refractory to treatment.</p>

ICU=intensive-care unit; IV=intravenous.

**Table 18C: Recommendation for Management of Raised Intracranial Pressure (ICP) After CAR T Cell Therapy**

Condition	Management
Stage 1 or 2 papilloedema with CSF opening pressure of <20 mmHg without cerebral oedema	Acetazolamide 1000 mg IV, followed by 250 to 1000 mg IV every 12 hours (adjust dose based on renal function and acid–base balance, monitored 1 to 2 times daily) (dose as age appropriate).
Stage 3, 4, or 5 papilloedema, with any sign of cerebral oedema on imaging studies, or a CSF opening pressure of ≥20 mmHg	<p>Use high-dose corticosteroids with methylprednisolone IV 1 g/day (as age appropriate), as recommended for Grade 4 CAR-T-cell-related encephalopathy syndrome (Table 18A).</p> <p>Elevate head end of the patient’s bed to an angle of 30 degrees.</p> <p>Hyperventilation to achieve target partial pressure of arterial carbon dioxide (PaCO<sub>2</sub>) of 28 to 30 mmHg, but maintained for no longer than 24 hours.</p> <p>Hyperosmolar therapy with either mannitol (20 g/dl solution) or hypertonic saline (3% or 23.4%, as detailed below) Mannitol: initial dose 0.5 to 1 g/kg; maintenance at 0.25 to 1 g/kg every 6 hours while monitoring metabolic profile and serum osmolality every 6 hours, and withhold mannitol if serum osmolality is ≥320 mOsm/kg, or the osmolality gap is ≥40.</p> <p>Hypertonic saline: initial 250 mL of 3% hypertonic saline; maintenance at 50 to 75 mL/h while monitoring electrolytes every 4 hours, and withhold infusion if serum Na levels reach ≥155 mEq/l.</p> <p>For patients with imminent herniation: initial 30 mL of 23.4% hypertonic saline; repeat after 15 minutes, if needed.</p> <p>If patient has ommaya reservoir, drain CSF to target opening pressure of &lt;20 mmHg.</p> <p>Consider neurosurgery consultation and IV anaesthetics for burst-suppression pattern on electroencephalography.</p> <p>Metabolic profiling every 6 hours and daily CT scan of head, with adjustments in usage of the aforementioned medications to prevent rebound cerebral oedema, renal failure, electrolyte abnormalities, hypovolemia, and hypotension.</p>

CSF=cerebrospinal fluid; CT=computerised tomography; IV=intravenous.

## 10.7 HYPOGAMMAGLOBULINAEMIA

Hypogammaglobulinaemia may occur as a consequence of depletion of normal B cells by AUTO3. However, the degree and duration of this is likely to depend on the persistence of CAR T cells in the body and could last for months to years. At worst, the clinical consequences of therapeutic B cell elimination will resemble X-linked agammaglobulinaemia and patients with this disorder have a normal life expectancy when treated with regular i.v. Ig replacement. Immunoglobulin levels will be monitored regularly as in the Schedule of Assessments and patients with severe hypogammaglobulinaemia (serum IgG level is <4 g/L) persisting longer than 6 months will be treated with i.v. Ig administration until recovery of B cells and normal IgM levels, or lifelong if this does not occur. Management of hypogammaglobulinaemia associated with AUTO3 will be per the suggested guidance or as per the local institutional standards.

## **10.8           MANAGEMENT OF IMMUNE-RELATED ADVERSE EVENTS DUE TO ON-TARGET BUT OFF-TUMOUR TOXICITY**

Though it is unlikely that AUTO3 will cause immune-related AEs (irAEs) due to on-target but off-tumour toxicity, the patients will be closely monitored for signs and symptoms indicative of irAEs, which may allow for an early recognition of these events. Special attention will be paid to vital organs and irAEs of any grade involving vital organs (e.g. lung, brain, and eyes), more aggressive monitoring and rapid institution of appropriate supportive care including systemic steroids should be administered. In case of severe irAE, not successfully managed by general supportive care, treatment with steroids and other agents may be considered.

## 11 CONCOMITANT MEDICATIONS AND THERAPIES

Any medication the patient is receiving on Day -7 must be recorded. Thereafter only those that are related to an AE or are study related prophylaxis must be recorded along with:

- Reason for use
- Dates of administration including start and end dates
- Dosage information including dose and frequency

Concomitant medication may be given as medically indicated. Details (including doses, frequency, route and start and stop dates) of the concomitant medication/treatment given must be recorded in the patient's medical records and details entered into the eCRF. Standard drugs required by the patient may be administered alongside the trial protocol.

All safety management guidelines are only recommendations and deviations from this scheme are allowed according to the Investigator's judgement and local institutional practice.

### 11.1 ALLOWED CONCOMITANT MEDICATIONS/THERAPIES

**Palliative radiotherapy:** Palliative radiotherapy may be given concomitantly as clinically appropriate.

**Other permitted therapies:** The following medications and supportive therapies are examples of support therapies that may be used during the study:

- Anti-microbial prophylaxis may be given
- Colony stimulating factors, erythropoietin, and transfusion of platelets and red cells.

#### **Pre-and Post-AUTO3 Infusion Supportive Therapies:**

Please refer to [Section 10.2](#).

### 11.2 PROHIBITED AND CAUTIONARY THERAPIES

#### **Herbal, Homeopathic agents:**

No herbal, homeopathic agents should be used between Day -10 and Day 30 ( $\pm 3$  days) following last AUTO3 infusion, unless recommended by the Principal Investigator.

#### **Corticosteroids and Immunosuppressant (except for managing treatment related toxicity):**

The use of immunosuppressants such as high dose corticosteroids should be avoided where possible, as these are likely to influence the efficacy and possibly safety of AUTO3. Therapeutic doses (doses of  $>5$  mg prednisolone or equivalent) of steroids must be stopped at least 72 hours prior to leukapheresis and AUTO3 infusion. However, physiological replacement doses of steroids are allowed:  $<12$  mg/m<sup>2</sup>/day hydrocortisone or equivalent. Corticosteroids should also be avoided post-AUTO3 infusion if possible, although it is recognised that their use may be required in the context of development of CRS or infusion reactions.

Physicians may use any medication as clinically appropriate and necessary to manage emerging AEs. The use of other immunosuppressants should be discussed with the Sponsor's Medical Monitor.

Investigators can use any medication based on their clinical judgement and local institutional practice to optimise patient safety.

### **11.3 BRIDGING THERAPY**

Patients may receive bridging therapy while the product is being manufactured. The dates of bridging therapy, the chemotherapy agents and the doses given must be recorded in the eCRF. Additionally, the intent of bridging therapy such as prevention of disease progression or induction of remission in rapidly progressing disease should be documented in the eCRF.

Chemotherapy must be stopped 1 week before leukapheresis and 2 days prior to starting pre-conditioning chemotherapy.

Any donor lymphocyte infusion must be completed at least 6 weeks prior to AUTO3 infusion.

Any drug used for GVHD must be stopped at least 4 weeks prior to AUTO3 infusion.

### **11.4 OVER-DOSAGE**

There is currently no experience of overdose of AUTO3 as no clinical studies have been performed to date. There is no specific treatment for an overdose of AUTO3. In event of overdose, any adverse reactions could be treated symptomatically. In the event of unmanageable toxicity, steroids may be used to deplete AUTO3.

AUTO3 cells will be provided in patient specific dose aliquots and will be administered by trained staff in a hospital setting; therefore, the chance of overdose is unlikely.

### **11.5 DIETARY AND LIFESTYLE RESTRICTIONS**

No dietary restrictions are recommended. A normal balanced diet is recommended; the patient may also continue his/her normal diet as appropriate.

Donating blood products and organs:

Patients should not donate blood, organs, tissue or cells to others if they receive AUTO3.

## 12 SAFETY AND PHARMACOVIGILANCE

### 12.1 DEFINITIONS

#### 12.1.1 Definition of an Adverse Event

An AE is defined as any untoward medical occurrence in a patient administered a medicinal product which does not necessarily have a causal relationship with the treatment.

An AE can therefore be any unfavourable and unintended sign (including an abnormal laboratory finding, for example), symptom or disease temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product.

#### 12.1.2 Definition of an Adverse Reaction

An adverse reaction is any untoward and unintended responses to a medicinal product related to any dose administered. A causal relationship between a medicinal product and an AE is at least a reasonable possibility, e.g. the relationship cannot be ruled out.

An unexpected adverse reaction is an adverse reaction in which the nature or severity of which is not consistent with the Reference Safety Information section outlined in the IB for AUTO3.

#### 12.1.3 Definition of a Serious Adverse Event

An SAE is defined as an AE that meets any of the following criteria:

- **Results in death** (death due to disease progression will not be considered as an SAE).
- **Life-threatening** (the term ‘life-threatening’ refers to an event in which the patient was at risk of death at the time of the event. It does not include any AE that, had it occurred in a more severe form, might have caused death).
- **Requires in-patient hospitalisation or prolonged existing hospitalisation.**
- **Results in persistent or significant disability/incapacity.**
- **Congenital anomaly/birth defect.**
- **Medically significant** (e.g. important medical events that may not be immediately life-threatening or result in death or hospitalisation but may jeopardise the patient or may require intervention to prevent one of the other outcomes listed above).

If the event was initially non-serious, the onset of the SAE should be considered the time when the AE met the above seriousness criteria. Resolution of the SAE should also be considered when the event no longer is serious and is either a non-serious event or has resolved.

#### **Protocol-specific SAEs are as follows:**

- Grade 4 CRS and neurotoxicity/ICANS.
- Any new primary cancers.
- Significant cardiac dysfunction such as Grade 3 or higher decrease in LVEF.
- Grade 4 non-haematological laboratory abnormalities (not disease related) when considered clinically significant.

Serious adverse events must be reported by the Investigator to the Sponsor within 24 hours of being made aware of their existence (see [Section 12.3.2](#) for reporting instructions).

**Events NOT considered as SAEs:**

- A procedure requiring hospitalisation for protocol/disease-related investigations (e.g. surgery, scans, endoscopy, sampling for laboratory tests, and BM sampling). However, hospitalisation or prolonged hospitalisation for a complication of such procedures remains a reportable SAE.
- Extension of routine in-patient hospital stay beyond the recommended 30 days ( $\pm 3$  days) for general management purposes or if it is standard process for the management of underlying disease.
- Routine treatment or monitoring of the indication not associated with any deterioration in condition.
- The administration of blood or platelet transfusion as routine treatment of studied indication. However, hospitalisation or prolonged hospitalisation for a complication of such transfusion remains a reportable SAE.
  - Prolonged Grade 4 cytopenia lasting more than 60 days or those considered medically significant or requiring significant management in addition to transfusions and growth factor support (e.g. stem cell top up or transplant) should be reported as SAEs.
- Hospitalisation or prolongation of hospitalisation for technical, practical, or social reasons, in absence of an AE.
- Hospitalisations not intended to treat an acute illness or AE (e.g. social reasons such as pending placement in a long-term care facility).
- A procedure that is planned (i.e. planned prior to starting of treatment on study) must be documented in the source document and the eCRF. Hospitalisation or prolonged hospitalisation for a complication remains a reportable SAE.
- An elective treatment of a pre-existing condition unrelated to the studied indication.
- Emergency outpatient treatment or observation that does not result in admission, unless fulfilling other seriousness criteria above.
- An event which is part of the natural course of the disease under study (i.e. disease progression or hospitalisation due to disease progression, or pain control, stabilisation of fractures) does not need to be reported as an SAE. Death due to the disease under study is to be recorded on the Death Case Report Form. However, if the underlying disease (i.e. progression) is greater than that which would normally be expected for the patient, or if the Investigator considers that there was a causal relationship between treatment with study medication(s) or protocol design/procedures and the disease progression, then this must be reported as an SAE.

**12.1.4 Suspected Unexpected Serious Adverse Reactions**

A suspected unexpected serious adverse reaction (SUSAR) is any suspected adverse reaction related to the study treatment that is both unexpected and serious (see [Section 12.3.3](#)).

### 12.1.5 Adverse Events of Special Interest

The following are AEs of special interest:

- Grade 2-5 CRS.
- Grade 2-5 neurotoxicity (including depressed level of consciousness, dysphagia, ataxia, seizures and cerebral oedema).

Adverse events of special interest  $\geq$ Grade 3 will be reported on an AESI/SAE form to the Sponsor within 24 hours of becoming aware of the event. Events can be both an AESI and a SAE.

## 12.2 ASSESSMENT OF ADVERSE EVENTS

Adverse events will be elicited at each study visit as indicated in the Schedule of Assessments and as clinically necessary. Patients will be instructed to report any AEs occurring between study visits to the study site. Adverse events will be assessed by the Investigator, or appropriately qualified designee, for severity, relationship to study treatment, action taken, outcome and whether the event meets criteria as an SAE according to the guidelines presented in [Section 12.1.3](#).

### 12.2.1 Severity of Adverse Events

The severity of AEs will be graded according to the NCI CTCAE (version 5.0).

Adverse events that are not defined in the NCI CTCAE should be evaluated for severity according to the following scale:

**Table 19: Severity Grading of AEs Not Listed on the NCI CTCAE Grading System**

Grade	Severity	
1	Mild	Transient or mild discomfort; no limitation in activity; no medical intervention/therapy required.
2	Moderate	Mild to moderate limitation in activity, some assistance may be needed; no or minimal medical intervention/therapy required.
3	Severe	Marked limitation in activity, some assistance usually required; medical intervention/therapy required, hospitalisation is possible.
4	Life-threatening	Extreme limitation in activity, significant assistance required; significant medical intervention/therapy required, hospitalisation or hospice care probable.
5	Fatal	Death as a result of this AE.

AE = adverse event.

It is important to distinguish between serious and severe AEs. Severity is a measure of intensity whereas seriousness is defined by the criteria in [Section 12.1.3](#).

### 12.2.2 Relationship of Adverse Events to Treatment

The Investigator must determine the relationship between the administration of the study drug and the occurrence of an AE/SAE as defined below:

#### Relationship assessments classified as “Not Related” to study treatment:

Not Related: The AE is not related to the investigational product. The patient either did not receive the investigational product or the event is related to an

aetiology other than the investigational product (the alternative aetiology must be documented in the study patient's medical record).

Unlikely Related: The AE is doubtfully related to the investigational product.

The event is not clearly related to an identified aetiology other than the investigational product; but there is no plausible mechanism for the event to be related to the investigational product and/or there is no clear association between the event and the administration of the investigational product.

**Relationship assessments classified as “Related” to study treatment:**

Possibly Related: The AE may reasonably be related to the investigational product.

Probably Related: The AE is likely to be related to the investigational product.

There is an association between the event and the administration of the investigational product, there is a plausible mechanism for the event to be related to the investigational product, the event is less likely to be explained by known characteristics of the patient's clinical status, and an alternative aetiology is not apparent.

Definitely Related: The AE is clearly related to the investigational product.

If an event is assessed as related to a drug other than the investigational product which has not been provided by the Sponsor, the name of the manufacturer must be provided when reporting the event.

## **12.3 REPORTING PROCEDURES**

### **12.3.1 All Adverse Events**

All AEs will be recorded in the eCRF using medical terminology that accurately reflects the event. A diagnosis should be recorded when signs and symptoms are due to a common aetiology. The relationship of the AE to study therapy must be recorded in the eCRF by the Investigator or designee. All measures required to manage an AE must be recorded in the patient's medical notes and reported accordingly in the eCRF.

Anaemia, neutropenia, lymphopenia, thrombocytopenia requiring transfusion are expected (due to disease and pre-conditioning) and are not subject to reporting unless they persist beyond Day 30, are considered clinically significant and/or require advanced intervention above blood product and GCSF support, or meet the criteria as an SAE.

Similarly, Grade 1-2 laboratory abnormalities are extremely frequent in this patient population and are not subjected to reporting unless felt to be clinically significant and/or require advanced intervention above blood product and GCSF support.

Laboratory related AEs should be entered along with laboratory values in the laboratory section of CRF. Whenever possible, AEs associated with laboratory values, should have these values included in the CRF. Reporting of events such as febrile neutropenia should associated with temperature reading entry in vitals and ANC in laboratory section. Non laboratory related AEs should be entered in the AE section.

AEs/SAEs related to leukaemia or due to holding chemotherapy administered after enrolment but before pre-conditioning should not be reported as AEs/SAEs for this study. Significant events should be added to the patient’s medical history.

The reporting period for all AEs is described in [Table 20](#).

**Table 20: Reporting Period for All AEs**

<b>From:</b> ICF signature <b>Until:</b> Day -7	<b>From:</b> Day -7 <b>Until:</b> Day 60 post last AUTO3 infusion*	<b>From:</b> Day 60 post last AUTO3 infusion <b>Until:</b> End of study *
<ul style="list-style-type: none"> <li>AEs/SAEs related to study procedures (leukapheresis, bone marrow assessments, lumbar punctures etc).</li> <li>Note: AEs/SAEs associated with holding/bridging therapy prior to admission for conditioning chemotherapy, are not associated with study procedures and therefore do not need to be reported as AEs.</li> </ul>	<ul style="list-style-type: none"> <li>All AEs</li> <li>All SAEs</li> </ul>	<ul style="list-style-type: none"> <li>Treatment related (to AUTO-3) Grade 3-5 AEs</li> <li>Treatment related (to AUTO-3) SAEs</li> <li>AEs of special interest (<a href="#">Section 12.1.5</a>)</li> <li>AEs related to study procedure (e.g. BM aspirate, lumbar puncture)</li> </ul>

AE = adverse event; SAE = serious adverse event.

\* or until withdrawal whichever occurs first.

\*\* as patients may be getting intervening non-study related treatments. Adverse events occurring out of this window may be reported if the Investigator considers them to be related to this study.

The Investigator should follow each AE until 1 of the following occurs:

- Adverse event has resolved to baseline or Grade 1
- Adverse event is assessed as stable by the Investigator
- Patient is lost to follow-up or
- Patient withdraws consent

If a drug related AE and/or any SAE is ongoing at the time of study completion (2 years after dosing or disease progression, whichever happens first) the event will be followed until resolved or stabilised for up to a maximum of 6 months, or earlier if one of the following apply:

- Death
- Withdrawal of consent
- Start of new treatment or
- Patient lost to follow-up

### 12.3.2 Serious Adverse Event Reporting

All SAEs occurring during the study must be reported to the Sponsor within 24 hours of becoming aware of the event. Additional or follow-up SAE reports should be submitted with relevant information promptly. Should the regulatory authority require the Sponsor to submit additional data on the event, the Investigator will be requested to provide additional data to the Sponsor promptly.

**Information to be Provided for an SAE:**

Although much information will have been provided in the eCRF, this information may not always be readily available at the time of evaluating each Serious Adverse Event, or when reporting to the regulatory authorities, so it is essential that as much relevant information is included on the AESI/SAE form. At a minimum, the following is required.

- An identifiable patient (patient number)
- Reporter details
- The AUTO product (if not clear on the AESI/SAE form) if administered
- The Protocol (if not clear on the AESI/SAE form)
- The Adverse Event, with a description of the event and a causality assessment

Please also ensure that this information is accurate, at the time of reporting. Missing information, or information that is inconsistent with the eCRF will automatically generate queries. The initial SAE report will be provided to the Sponsor (or designee) using the AEI/SAE form. The Sponsor or designee will require the following information related to the event:

The SAE form completion and reporting must not be delayed, even if all of the information is not available at the time of the initial contact.

The SAE report should be submitted to the following by fax or email:

- SAE Fax number: +44 (0) 1252 842277
- Email: Autolus@tmcpharma.com

**The 24-Hour Safety Hotline:**

TMC Pharma Services Ltd  
Hartley Wintney  
Hampshire  
RG27 8AS, UK  
Tel: +44 (0)1252 842255

Fax numbers and email address are listed on the SAE form and in the SAE form completion guidelines.

**SAE Follow-up Reporting:**

After the initial SAE report, the Investigator is required to provide additional follow-up information on the SAE by submitting an updated SAE report form to the Sponsor (or designee). New significant information includes, but is not limited to, the following:

- New signs or symptoms or a change in the diagnosis
- Significant new diagnostic test results
- Change in causality based on new information
- Change in the event's outcome, including recovery
- Additional narrative information on the clinical course of the event

Serious adverse events must be followed through to resolution by the Investigator. Resolution is defined as a return to baseline status, Grade 1, or stabilisation of the condition with the expectation that it will remain chronic. For all SAEs, the Investigator may be requested to obtain additional information in an expedited manner. This additional information will allow for a complete medical assessment of the case and independent determination of possible causality. Information on other possible causes such as concomitant medication and illnesses must be provided.

The Medical Monitor may specify a longer period of time, if required, to assure the safety of the patient.

At the end of the study, or following discontinuation, the events listed below will be reported under the long-term follow-up protocol:

- Adverse events of special interest
- Serious adverse events related to the study drug
- New malignancies

### **12.3.3 Suspected Unexpected Serious Adverse Reaction Reporting**

If the event is evaluated as a SUSAR, i.e. unexpected events that are related (reasonable possibility) to AUTO3, the Sponsor will submit the SUSAR to the regulatory authorities and the Research Ethics Committee within 7 calendar days for initial reports of fatal/life-threatening events (with a follow-up report within a further 8 calendar days) and 15 calendar days for all other events, or in accordance with local regulatory requirements. Where there are conflicting evaluations of causal relationship, the more conservative will be used for reporting purposes.

All reporting to regulatory authorities will be by Autolus, or through an Autolus designated vendor.

The Sponsor (or designee) will notify Investigators of all SUSARs, in accordance with local regulations. The Investigator must immediately review with the Investigator's site team and retain the documentation in the Investigator Site File.

### **12.3.4 Adverse Events of Special Interests Reporting**

Adverse events of special interest that are  $\geq$ Grade 3 will follow the same reporting procedure and timelines as described in [Section 12.3.2](#) for SAEs. The report form should be completed and faxed or emailed to the sponsor or designee (fax numbers and email address are listed on the bottom of the form and in the SAE form completion guidelines) within 24 hours of becoming aware of the event.

### **12.3.5 Pregnancy Reporting and Management**

#### **Pregnancies**

Female patients of childbearing potential will be instructed to immediately inform the Investigator if they become pregnant during the study or within 12 months after the last dose of study treatment. Any pregnancies during this period must be reported to the Sponsor (or designee) within 24 hours of knowledge of the event using the pregnancy report form (fax numbers and email address are listed on the bottom of the form).

While pregnancy itself is not considered to be an AE or SAE, any pregnancy complication or elective termination of a pregnancy for medical reasons will be recorded as an AE or SAE.

Abnormal pregnancy outcomes (e.g. spontaneous abortion, foetal death, stillbirth, congenital anomalies, and ectopic pregnancy) are considered SAEs and must be reported to the Sponsor immediately using the SAE form (i.e. no more than 24 hours after learning of the event).

The Investigator must counsel the patient, discussing the risks of the pregnancy and the possible effects on the foetus. Monitoring of the patient should continue until conclusion of the pregnancy.

### **Pregnancies in Female Partners of Male Patients**

Male patients will be instructed to immediately inform the Investigator if their partner becomes pregnant during the study or within 12 months after the last dose of study treatment. Any pregnancies during this period must be reported to the Sponsor (or designee) within 24 hours of knowledge of the event using the pregnancy report form (fax numbers and email address are listed on the bottom of the form).

Attempts should be made to collect and report details of the course and outcome of any pregnancy in the partner of a male patient exposed to study treatment. The pregnant partner will be asked to sign an authorisation for use and disclosure of pregnancy health information. The Investigator may provide information on the risks of the pregnancy and the possible effects on the foetus, to support an informed decision in cooperation with the treating physician and/or obstetrician.

### **Outcome**

Additional information on the course and outcome of the pregnancy should be provided to the Sponsor (or designee) as soon as becoming available (i.e. no more than 24 hours after obtaining the information) using the paper pregnancy report form. The following pregnancy outcomes will be considered to be SAEs and should be reported according to the procedure in [Section 12.3.2](#).

- Spontaneous abortion (as the Sponsor considers spontaneous abortions to be medically significant events).
- Any congenital anomaly/birth defect in a child born to a female patient or female partner of a male patient.
- All neonatal deaths occurring within 30 days of birth should be reported as SAEs, without regard to causality.

Follow-up information regarding the outcome of the pregnancy and any postnatal sequelae in the infant will be required.

### **12.3.6 Overdose Reporting and Dosing Errors**

Safety information on the study drug may require expedited reporting and/or safety evaluation. This will include:

- Overdose of a study drug
- Suspected abuse/misuse of a study drug
- Inadvertent or accidental exposure to a study drug
- Medication error involving a study drug

A study drug overdose is the accidental or intentional use of AUTO3 in an amount higher than the dose being studied. An overdose or incorrect administration of study drug is not an

AE unless it results in untoward medical effects. Any AUTO3 overdose or incorrect administration of AUTO3 should be noted on the corresponding eCRF page. All AEs associated with an overdose or incorrect administration of AUTO3 should be recorded on the AE eCRF page. If the associated AE fulfils the criteria for an SAE, the event should be reported immediately (i.e. no more than 24 hours after learning of the event; see [Section 12.3.2](#)).

## 13 OVERSIGHT COMMITTEES

### 13.1 SAFETY EVALUATION COMMITTEE

Patient safety will be monitored throughout all parts of the study by a SEC established by the Sponsor. This internal committee will monitor treatment-emergent data on an ongoing basis throughout study conduct for the purpose of ensuring the continued safety of patients enrolled in this study.

The SEC will be chaired by the Sponsor's Medical Monitor and membership will include Study Investigators, statistician, safety/pharmacovigilance and biomarker representative, along with additional Sponsor staff as appropriate. The team will meet at regular frequencies throughout study conduct;

During Phase I (dose escalation), the SEC will meet:

1. After the first patient in each cohort completes 2 weeks
2. After the third patient in each cohort completes the DLT evaluation period of 30 days ( $\pm 3$  days) (after the last dose of AUTO3 in the case of split dosing). In case of DLT, will meet again after the sixth patient has been treated to make the dose escalation and schedule decision
3. Additional ad hoc meetings if safety stopping criteria is met
4. When clinically necessary based on emerging data.

During Phase II (dose expansion), the SEC will meet:

1. On a periodic basis not exceeding 3 to 6 months to assess cumulative safety data
2. After 10 patients have completed 30 days following AUTO3 infusion
3. After 20 patients have completed 30 days following AUTO3 infusion

The Sponsor will maintain documentation of meeting outcomes. Decisions with the potential to impact patients' safety (e.g. unfavourable change in risk/benefit assessment) will be promptly communicated to Investigators, ethics committees and regulatory authorities as appropriate. Throughout the trial, information regarding all SAEs and potential DLTs will be sent to the SEC members. Dose limiting toxicities will be monitored centrally and the decision to assign the optimal dose will be taken by the SEC.

Dose escalation decisions in Phase I of the study will be made by the SEC. The schedule of dose escalation meetings will depend on the time to completion of a cohort frequency of DLT and when a RP2D is determined. The SEC may stop further enrolment into one or more of the cohorts if treatment-emergent toxicity is determined to result in an unfavourable change in patient benefit/risk. Decisions and/or recommendations made by the SEC will be communicated to the Principal Investigators at all active study centres and to the Sponsor.

Mandatory SEC processes will be included in a SEC Charter.

### 13.2 INDEPENDENT DATA MONITORING COMMITTEE

An IDMC, consisting of 2 independent physicians and 1 statistician, will be established by the Sponsor and they will review serious safety events. The IDMC will meet up on occurrence at the following; the decision of the IDMC will supersede that of SEC:

- When any safety stopping, criteria ([Section 3.6](#)) is met

- Prior to opening Phase II
- Every 6 months during Phase II to review cumulative safety data

Throughout the trial, information regarding all SAEs and DLTs will be sent to the IDMC members. IDMC can ask for and will be provided with any additional information relevant to the SAEs and DLTs. The IDMC will receive all dose evaluation/escalation meeting minutes (as soon as possible following the meeting) and will have the opportunity to review and can over-rule the SEC decision if clinically warranted and necessary.

When a RP2D decision is made by the SEC, the decision will need to be reviewed and endorsed by the IDMC prior to commencing Phase II.

Upon occurrence of any events as defined above, detailed event summaries and cumulative safety data will be sent to the IDMC. A decision to continue or to hold or modify the study will be made by the IDMC. If and when a study is stopped, it will be re-started after a protocol amendment has been approved by the regulatory authorities and ethics committees.

Decisions and/or recommendations made by the IDMC will be communicated to the Principal Investigators at all active study centres and to the Sponsor.

Mandatory IDMC processes will be included in the IDMC Charter.

## 14 STATISTICS

Further details of the statistical analysis of all the endpoints will be included in a separate Statistical Analysis Plan. Any analysis that deviates from the Statistical Analysis Plan will be documented and justified in the Clinical Study Report.

### 14.1 SAMPLE SIZE ESTIMATION

Up to 100 patients in total are expected to be enrolled into both the dose escalation and expansion parts of the study, and up to 84 patients in total are anticipated to be treated with AUTO3 therapy. The difference between the number of enrolled and treated patients accounts for manufacturing failures and inability of some patients to meet AUTO3 infusion criteria.

- **Phase I (Escalation):** Up to 36-60 patients treated in total (up to 3-6 per dose cohort, following a Rolling 6 design (Skolnik et al. 2008), which will consist of:
  - 24-36 patients in the paediatric / young adult patient cohorts (age 1-24 years)
  - 12-24 patients in the adult patient cohorts ( $\geq 25$  years)
  - *Additional number accounts for the possibility of patient with higher disease burden being evaluated using a single dose*
- **Phase II (Expansion):** Dose expansion: up to 24 evaluable paediatric/young adult patients (aged 1-24 years) in total, using a Simon's 2-stage optimal design.

Simon's 2-stage design (Simon 1989) will be used for Phase II. The null hypothesis that the true response rate is 25% will be tested against a 1-sided alternative. In the first stage, nine patients will be accrued. If there are two or fewer responses in these nine patients, the study will be stopped. Otherwise, 15 additional patients will be accrued for a total of 24. The null hypothesis will be rejected if 10 or more responses are observed in 24 patients. This design yields a 1-sided type I error rate of 5% and 80% power when the true response rate is 50%.

### 14.2 DESCRIPTION OF ANALYSIS DATASETS

#### 14.2.1 Full Analysis Set

The full analysis set will consist of all patients enrolled into the study.

#### 14.2.2 Safety Analysis Set for the Study and for Study Treatment

All patients who have started pre-conditioning therapy of FLU and CY will be included in the safety analysis set for the study. A patient who has pre-conditioning therapy but not AUTO3 therapy will not be excluded from the safety analysis set for the study.

All patients who receive at least one dose (complete or partial dose) of AUTO3 therapy will be included in the safety analysis set for the study treatment.

#### 14.2.3 Efficacy Analysis Set

All patients who receive at least one dose (complete or partial dose) of AUTO3 therapy will be included in the efficacy analysis set.

### 14.3 STATISTICAL ANALYSES AND METHODS

Continuous data will be summarised using the mean, median, standard deviation, minimum and maximum, while frequency counts and percentages will be presented for discrete variables. Time to event endpoints will be summarised using the Kaplan-Meier (KM) method. Summary statistics will be presented for baseline characteristics.

The paediatric/young adult patient cohorts (age 1-24 years) will be analysed separately from the adult patient cohorts ( $\geq 25$  years) in Phase I.

### 14.4 PRIMARY ENDPOINTS

The primary endpoints of the study are as follows:

#### **Phase I: Safety and RP2D**

- Incidence of Grade 3-5 toxicity occurring within the DLT period (30 days  $[\pm 3]$  days] after last dose) following AUTO3 infusion.
- Frequency of DLT.

#### **Phase II: Anti-Leukaemic Effect and Safety**

- Proportion of patients achieving morphological remission (CR/CRi) and MRD-negative response in BM within 30 days ( $\pm 3$  days) post AUTO3 administration.
- For patients with isolated CNS disease, anti-tumour effect will be assessed by the proportion of patients achieving clearance of their CSF from CNS disease with ongoing BM CR 30 ( $\pm 3$ ) days post AUTO3 infusion (or first post-infusion CNS assessment).

#### 14.4.1 Phase I: Safety

Safety associated with AUTO3 administration (only those who received AUTO3).

Summary statistics and analyses will be provided by dose level and overall. The safety analysis set for the study and study treatment will be used for the analysis of safety data.

Safety evaluations will be based on the incidence, severity and type of AEs, and changes in the patient's vital signs and clinical laboratory results.

Adverse events will be coded using the Medical Dictionary for Regulatory Activities AE coding system for purposes of summarisation. All AEs occurring on study will be listed in by-patient data listings. Treatment-emergent events will be tabulated, where treatment-emergent is defined as any AE that occurs during or after administration of AUTO3 up to 60 days after the last infusion, any  $\geq$ Grade 3 event that is considered drug-related regardless of the start date of the event. Events that are considered related to treatment (possibly, probably, or definitely related) will also be tabulated. Adverse events by the NCI CTCAE toxicity grade will also be summarised. Deaths and SAEs will be tabulated in data listings including additional relevant information on the patient.

Laboratory toxicity grades will be calculated for the appropriate laboratory parameters per NCI CTCAE version 5.0.

Adverse events of special interest will be analysed in greater depth, including the time to onset and time to resolution where appropriate.

#### 14.4.2 Phase I: Recommended Phase II Dose and Maximum Tolerated Dose

At the end of the Phase I dose escalation part of the study, the RP2D and dose schedule will be identified based on the safety data and evaluation of the activity data collected during Phase I.

#### 14.4.3 Phase II: Anti-Leukaemic Effect

**The primary endpoints for Phase II (efficacy) will be assessed as follows:**

- Proportion of patients achieving morphological remission (CR or CRi) and MRD-negative complete response in the first post-infusion BM assessment within 30 days ( $\pm 3$  days) post first AUTO3 administration.
- For patients with isolated CNS disease anti-tumour effect will be assessed by whether patients clear their CSF disease with ongoing BM remission within 30 days ( $\pm 3$  days) post first AUTO3 administration

The response rate calculations will be based on the entire Phase II patient populations.

#### 14.5 SECONDARY ENDPOINTS

The secondary endpoints of the study are as follows:

##### **Safety of AUTO3**

- Frequency and severity of AEs and SAEs.
- Incidence and duration of severe hypogammaglobulinaemia (serum IgG level is  $< 4$  g/L).

##### **Feasibility of AUTO3 Manufacture in this Patient Population (All Patients)**

- Feasibility of product generation will be examined by assessing the number of AUTO3 successfully manufactured as a fraction of the number of patients undergoing leukapheresis (and all patients registered).

##### **Clinical Efficacy of AUTO3**

- Proportion of patients (all and prior CD19 CAR T treatment naïve) achieving morphological remission (CR/CRi) and MRD-negative response within 30 days ( $\pm 3$  days) post first AUTO3 administration based on flow or PCR. Evaluate clinical outcomes including RFS, EFS, PFS, and OS.

**Relapse free survival (RFS):** Relapse free survival is measured by the time from achievement of CR or CRi (whichever occurs first) to relapse or death due to any cause during CR or CRi. In case a patient does not have relapse or death due to any cause prior to data cut-off, RFS will be censored at the date of the last adequate disease assessment.

In the main analysis of RFS, patients who proceed to SCT after AUTO3 infusion will be censored at the time of SCT. In addition, a sensitivity analysis of RFS will be performed without taking time of SCT into account.

RFS will be assessed only in patients with the best overall response of CR or CRi. The distribution function of RFS will be estimated using the KM method. The median RFS along with 95% confidence intervals will be presented if appropriate. The 6, 12, and 24 months EFS will also be assessed.

- **Event free survival (EFS):** Event free survival is the time from date of first AUTO3 infusion to the earliest of the following:

- Death from any cause
- Morphological relapse
- Treatment failure: Defined as no response in the study and discontinuation from the study due to any of the following reasons:
  - Adverse event
  - Lack of efficacy or progressive disease
  - New anticancer therapy

In case of treatment failure, the event date will be set to study Day 0. In case a patient does not have relapse, death due to any cause or treatment failure (e.g. discontinuation because of withdrawal of consent, lost to follow-up, protocol violation or administrative problems or transplant prior to relapse) prior to data cut off, EFS is censored at the date of last adequate disease assessment. Patients who undergo SCT or other therapy prior to morphological relapse will also be censored at the date of last adequate disease assessment.

EFS will be assessed in the efficacy analysis set. The distribution function of EFS will be estimated using the KM method. The median EFS along with 95% confidence intervals will be presented if appropriate. The 6, 12, and 24 months EFS will also be assessed.

- Progression-free survival will be calculated from the date of AUTO3 treatment to the date of disease relapse or disease progression. Patients who have not progressed, relapsed or died from disease will be censored at the last adequate disease assessment.
- Overall survival will be calculated from the date of AUTO3 treatment to the date of death. Patients who have not died will be censored at the date of last contact (clinic visit or telephone contact).
- Proportion of patients in molecular remission without further therapy at 6 months, 1 and 2 years ( $\pm 14$  days) following treatment with AUTO3.
- Incidence of CD19 and or CD22-negative relapse.

### **Biomarker, Pharmacokinetic and Pharmacodynamic Effects of AUTO3**

The following analyses on the pharmacokinetics and pharmacodynamic effects of AUTO3 will be performed for patients who received AUTO3:

- Expansion and persistence of CD19/CD22 CAR-positive T cells as determined by qPCR and/or flow cytometry will be summarised using the appropriate statistical methods.
  - Expansion is defined as the maximum level of CD19/CD22 CAR expression in both qPCR (copies/ $\mu\text{g}$  genomic DNA) and/or flow cytometry (cells/ $\mu\text{L}$ ) assays during follow-up.
  - Engraftment is defined as detection of any level of CD19/CD22 CAR expression in circulating T cells (i.e. PBMC) by qPCR (viral insertions per genome) and/or flow cytometry following infusion (cells/ $\mu\text{L}$ ) above baseline controls.
  - Persistence is defined as the duration of detectability, from infusion to the first negative result (less than detection limit).
- The kinetics of CD19/CD22 CAR-positive T will be documented over time for each patient and the area under the curve calculated, then summarised for all patients.

- Duration of B cell aplasia as determined by flow cytometry in the peripheral blood.

#### 14.6

- [REDACTED]
- [REDACTED]
- [REDACTED]
- [REDACTED]
- [REDACTED]

#### 14.7 IMMUNOGENICITY DATA

Analyses of immunogenicity data will be documented in the Statistical Analysis Plan.

#### 14.8 PHARMACOKINETICS

See above biomarker evaluation for cellular kinetics ([Section 14.5](#)).

#### 14.9 OTHER ANALYSES

Not applicable.

#### 14.10 INTERIM ANALYSIS

An interim analysis on safety and preliminary efficacy will be performed for the Phase I paediatric and adult cohorts separately when all Phase I patients have been followed for a minimum of 3 months. The start of the Phase II part of the study is not dependent on this interim analysis.

In Phase II of the study, an interim analysis will be performed when 9 paediatric/young adult (1-24 years old) patients have been evaluated for response. The response from the patients will be monitored on an ongoing basis:

- If 3 or more responses are observed before the 9<sup>th</sup> patient is evaluated for response, the study will continue without stopping recruitment.
- Otherwise recruitment will halt temporarily until all 9 patients have been assessed for response. Recruitment will re-start when 3 or more responses are observed in these 9 patients; otherwise the study will terminate.

## 15 STUDY COMPLETION/DISCONTINUATION AND WITHDRAWAL OF PATIENTS

### 15.1 COMPLETION

A patient will be considered to have completed the study if he or she has completed assessments up to and including the End of Study visit or has experienced a clinical endpoint that precludes further continuation in the study (e.g. death, unacceptable toxicity, disease progression).

### 15.2 DISCONTINUATION IN STUDY

A patient should be discontinued if:

- Disease progression as assessed by the Investigator.
  - Patients with frank morphologic relapse per Response Criteria for Acute Lymphoblastic Leukaemia ([Appendix 1](#)) will come off study.
  - Patients with molecular evidence of disease, defined as BM MRD  $>10^{-4}$  within the first 2 years after infusion by qPCR and/or flow cytometry, will have the BM MRD re-assessed 2-4 weeks later. If MRD  $>10^{-4}$  is confirmed, the patient may come off study to start a subsequent therapy. However, where possible, data regarding disease progression by morphological relapse should be collected to facilitate evaluations of aetiology of relapse.
- Clinical progression as assessed by the Investigator.
- The patient received concurrent (non-protocol) anticancer treatment.
- Intercurrent illness that prevents administration of pre-conditioning chemotherapy and or AUTO3 administration.
- Intercurrent illness that prevents further follow-up.
- Patient refuses further follow-up.
- Non-compliance with study procedures.

If a patient achieves a transient disease response but then shows evidence of recurrent disease associated with loss of detectable circulating CD19/CD22 CAR-positive T cells, further doses of AUTO3 may be given if available but the patient will be censored from the primary efficacy assessment at that time point but not discontinued from the study.

If a patient is discontinued after apheresis or AUTO3 product manufacture they may re-enter the study at the current dose level if and when they are considered to be eligible again.

If a patient discontinues the study, this will not result in automatic halt of data collection for the study. Following discontinuation, the patient should complete the End of Study visit as described in the Schedule of Assessments. The primary reason for treatment discontinuation will be documented in the eCRF. Once a patient discontinues, they will be eligible for entry into a long-term follow-up study if they have received AUTO3. If a patient has ongoing toxicity related to the study their exit from the study may be delayed until the event resolves or stabilizes.

### 15.3 PATIENT WITHDRAWAL FROM THE STUDY

A patient, or their parents/legal guardian, may withdraw consent and/or may be discontinued by the Investigator for any reason at any time.

A patient may withdraw or be withdrawn from the study for any of the following reasons:

- Lost to follow-up
- Withdrawal of consent<sup>1</sup>
- The Sponsor discontinues the study.

If a patient is lost to follow-up, every reasonable effort must be made by the study site personnel to contact the patient, or their parents/legal guardian, and determine the reason for discontinuation/withdrawal. The measures taken to follow-up must be documented.

When a patient withdraws before completing the study, the reason for withdrawal is to be documented in the eCRF and in the source document. Patients who withdraw may be replaced at the discretion of the Sponsor.

A patient who withdraws from the study will be eligible for entry into the long-term follow-up study. Additionally, research/biomarker samples collected will be retained and used in accordance with the original separate informed consent for research samples.

**Future Data Collection and Withdrawal of Consent:** Informed consent is a continuous and dynamic process. Patients, or their parents/legal guardian, are free to withdraw their consent at any point in the clinical study and without detriment to their future medical care. However, patients, or their parents/legal guardian, who wish to withdraw consent from further intervention/study visits may be willing to be followed in the clinical trial so as to enable the collection of key safety information. In this case, the Investigator will discuss this possibility with the patient, or their parents/legal guardian, to continue in the study and be contacted by a suitable means of communication agreeable to the patient, or their parents/legal guardian, to enable collection of information (survival status and evaluation of safety). In this event, the details should be recorded in the patient's hospital records and only key information entered into the eCRF thereafter. The above information will be presented in the main PIS and ICF to ensure the patient or their parent/legal guardian understands the available options.

#### 15.3.1 Procedures for Handling Withdrawals

A patient, or their parents/legal guardian, may, of their own volition, withdraw their consent at any time during the course of the study without any resulting detriment. All data collected up to the point of withdrawal will be maintained in the study database and included in subsequent analyses, as appropriate. Where a patient is withdrawn from the trial at their own request, or the request of their parents/legal guardian, or based on a decision of the Investigator, the follow-up should be maintained for safety review, subject to the continuing consent of the patient, or their parents/legal guardian. The Investigator will discuss the arrangements for withdrawing from any further study interventions and continuing to be followed for safety purposes.

If a patient is lost to follow-up at a site, every effort should be made to contact the patient's family doctor/general practitioner to obtain information on the patient's status.

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<sup>1</sup> Following withdrawal, no further protocol procedures can be performed. However, patients may be followed in the long-term follow-up protocol based on consent.

In the case AUTO3 does not meet the specifications of the release criteria, the case will be discussed with the Principal Investigator. It may be necessary to generate AUTO3 again if time permits (and biological screening repeated if necessary), otherwise the patient will be withdrawn from the study.

#### **15.4 REPLACEMENT POLICY**

If a patient is unable to be dosed on the planned day, they may undergo delayed dosing and be re-pre-conditioned (if appropriate) as long as they continue to meet the study enrolment criteria. Bone marrow sampling and imaging studies may not be repeated if the patient has not received any other anti-leukaemia therapy in the interim (excluding steroids and pre-conditioning chemotherapy). Patients undergoing delayed dosing may be evaluable for dose escalation decision making if the SEC so concludes.

Patients that have disease progression prior to completion of the DLT evaluation period or withdrawn from the study for reasons other than toxicity may be replaced, unless the SEC concludes that the patient is evaluable for dose escalation decision making.

## 16 END OF TRIAL/TERMINATION

For clinical trials located in the EU, a declaration of the end of the clinical trial will be made according to the procedures outlined in Directive 2001/20/EC, Article 10(c) and for those countries outside the EU, local regulations will be followed.

The study will be considered complete when the last patient completes their final follow-up visit. A final Clinical Study Report will be provided to the relevant authorities within 6 months after the end of the study.

The Sponsor reserves the right to terminate this study at any time for reasonable medical or administrative reasons. Any premature discontinuation will be appropriately documented per local requirements (e.g. IEC/IRB, regulatory authorities). In addition, the Investigator or the Sponsor has the right to discontinue a single site at any time during the study for medical or administrative reasons such as:

- Significant non-compliance with contractual enrolment timelines and targets
- Serious or continued Good Clinical Practice (GCP) non-compliance
- Inaccurate, incomplete, or delayed data collection
- Failure to adhere to the study protocol
- Failure to provide requested follow-up information for data queries.

## **17 ETHICAL AND REGULATORY CONSIDERATIONS**

### **17.1 ETHICS COMMITTEE REVIEW AND APPROVAL**

The final Clinical Study Protocol, including the final version of the written PIS, ICF and any other relevant patient facing material, must be approved, or given a favourable opinion in writing by an IEC/IRB as appropriate.

If it is necessary to amend the protocol or the PIS/ICF during the study, an IEC/IRB approval of the amended protocol and/or PIS/ICF must be obtained prior to implementation of the amended procedures and before new patients are consented to participate in the study using the amended version of the PIS/ICF.

### **17.2 REGULATORY AUTHORITY REVIEW AND APPROVAL**

The study will not commence before approval from the regulatory authority has been granted according to local requirements. The Sponsor (or designee) will be responsible for the preparation, submission, and confirmation of receipt of any regulatory authority approvals required prior to release of study treatment for shipment to the study site.

During the study, the Sponsor (or designee) is also responsible for submitting subsequent amendments and notifications to the regulatory authority according to local requirements.

### **17.3 INVESTIGATOR RESPONSIBILITIES**

#### **17.3.1 Overall Responsibilities**

The Investigator is responsible for conducting the study in full accordance with the Clinical Study Protocol, the latest revision of the Declaration of Helsinki, the GCP Consolidated Guideline, and all applicable national and local laws and regulations for clinical research. Information regarding any investigational sites participating in this study that cannot comply with these standards will be documented and appropriate actions taken. For studies conducted in the EU/European Economic Area countries, the Investigator will ensure compliance with the EU Clinical Trial Directive (2001/20/EC). For studies conducted in the United States or under a United States Investigational New Drug, the Investigator will additionally ensure adherence to the basic principles of “Good Clinical Practice” as outlined in the current version of 21 Code of Federal Regulations, subchapter D, part 312, “Responsibilities of Sponsor and Investigators”, part 50, “Protection of Human Subjects”, and part 56, “Institutional Review Boards”.

#### **17.3.2 Site Review**

Prior to the study start, the Investigator is required to sign a protocol signature page confirming his/her agreement to conduct the study in accordance with these documents and all the instructions and procedures found in this protocol, and to give access to all relevant data and records to Clinical Research Associates, auditors and regulatory authorities as required. Investigators ascertain they will apply due diligence to avoid protocol deviations.

The Investigator will make appropriate reports on the progress of this study to the Sponsor or its designee in accordance with applicable government regulations and their agreement with the Sponsor/Contract Research Organisation.

### **17.3.3 Informed Consent**

It is the responsibility of the Investigator to obtain written informed consent/assent from patients, or their parents/legal guardian, prior to conducting any study-related procedures. All consent documentation must be in accordance with applicable regulations and International Council on Harmonisation (ICH) GCP. Each patient, or their parents/legal guardian, is requested to sign and date the informed consent/assent form after she/he has received and read the PIS and received an explanation of what the study involves, including but not limited to the objectives, potential benefits and risk, inconveniences and the patient's rights and responsibilities.

Patients, or their parents/legal guardian, will be given adequate time to evaluate the information given to them before signing and dating the informed consent/assent form. It will also be explained to the patients, or their parents/legal guardian, that patients are free to refuse entry into the study at any time and without prejudice to future treatment. The informed consent/assent form must also be signed and dated by the person obtaining consent. The original signed informed consent/assent form for each patient, or their parents/legal guardian, will be retained on file by the Investigator, a copy will be given to the patient, or their parents/legal guardian and a copy will be kept in the patient's hospital notes.

Informed consent/assent forms must be retained for all screened patients and must be available for verification by the Study Monitor at any time.

In the event of changes to the informed consent/assent form during the study, the Investigator must always use the most current IEC/IRB approved form for documenting written informed consent.

### **17.3.4 Delegation of Investigator Duties**

The Investigator should ensure that all persons involved in the clinical study are adequately qualified, informed about the protocol, any amendment to the protocol, the study treatments, and their study related duties and functions before any involvement takes place. Delegation of any study related duties and documentation of training performed will be recorded in the signature and delegation log.

### **17.3.5 Communication with Independent Ethics Committee/Institutional Review Board**

A list of IEC/IRB members should be obtained by the Investigator or qualified designee and provided to the Sponsor/representative.

The Sponsor, or its designee, is responsible to assist the Investigator with applicable documentation for communication with IECs/IRBs. Before initiating a trial, the Investigator/institution should have written and dated approval of the study protocol, the patient ICF, any written information to the patients, patient recruitment procedures (e.g. advertisement if applicable), IB, Investigational Medicinal Product labelling (if applicable) and the coordinating investigator's curriculum vitae to the relevant IEC/IRB for evaluation before the study start. The IEC/IRB's unconditional approval statement will be transmitted by the Investigator to the Sponsor or a designee prior to shipment of AUTO3 to the site. This approval must refer to the study by exact protocol title and number, and should identify the documents reviewed and the date of review.

During the trial, the Investigator (or designee) is responsible for forwarding the applicable documents for approval and/or review including protocol deviations, protocol amendments,

ICF changes or revisions of other documents originally submitted for review including serious and/or unexpected adverse experiences occurring during the study, new information that may affect adversely the safety of the patients or the conduct of the study, an annual update and/or request for re-approval, and when the study has been completed. The Investigator or designee will follow national and local requirements.

The Investigator must supply the Sponsor with a copy of the list of members of the IEC/IRB and the letter(s) of approval(s) defining the version of each document approved.

### **17.3.6 Confidentiality of Trial Documents and Patient Records**

The Investigator must ensure that patients' anonymity will be maintained and that their identities are protected from unauthorised parties. The Sponsor will maintain confidentiality standards by assigning a unique coded identification number to each patient included in the study. Patient names will never be included in data sets that are transmitted to the Sponsor or their representatives, or to third parties as permitted by the ICF.

On eCRFs or other documents submitted to the Sponsor, patients will not be identified by their names, but by an identification code. The Investigator will keep a patient enrolment log relating codes to the names of patients. The Investigator will maintain documents not for submission to the Sponsor, e.g. patients' written consent forms, in strict confidence. Records will not be destroyed without giving the Sponsor prior written notice and the opportunity to further store such records, at the Sponsor's cost and expense.

After obtaining a patient's consent, the Investigator will permit the study monitor, independent auditor, or regulatory agency personnel to review the portion of the patient's medical record directly related to the study. This shall include all study relevant documentation (e.g. patient medical history to verify eligibility, laboratory tests result, admission/discharge summaries for hospital admissions occurring while the patient is on study and autopsy reports for deaths occurring during the study).

Patient medical information obtained by this study is confidential and may only be disclosed to third parties as permitted by the ICF signed by the patient, unless permitted or required by law.

### **17.4 LONG-TERM RETENTION OF SAMPLES FOR ADDITIONAL FUTURE RESEARCH**

Samples collected in this study may be stored by Autolus Ltd or an authorised designee for up to 15 years (or according to local regulations) for additional research. Samples will only be used to further understand AUTO3, to understand advanced cancers, to understand differential drug responders, and to develop tests/assays related to AUTO3 and advanced cancer. The research may begin at any time during the study or the post-study storage period.

Stored samples will be coded throughout the sample storage and analysis process and will not be labelled with personal identifiers.

## **18 ADMINISTRATIVE REQUIREMENTS**

### **18.1 DATA QUALITY CONTROL AND ASSURANCE**

#### **18.1.1 Compliance with the Protocol and Protocol Revisions**

The study shall be conducted as described in the approved protocol. All revisions to the protocol must be discussed with and be prepared by the Sponsor. The Investigator should not implement any deviation or change to the protocol without prior review and documented approval/favourable opinion from the IEC/IRB of an amendment, except where necessary to eliminate an immediate hazard(s) to the study patients.

If a deviation or change to a protocol is implemented to eliminate an immediate hazard(s) prior to obtaining IEC/IRB approval such deviation or change will be submitted for approval as soon as possible to:

- Independent Ethics Committee/IRB for review and approval/favourable opinion
- The Sponsor
- Regulatory authority(ies) if required by local regulations

Documentation of approval signed by the chairperson or designee of the IEC(s)/IRB(s) must be sent to the Sponsor.

If an amendment substantially alters the study design or increases the potential risk to the patient (1) the ICF must be revised and submitted to the IEC(s)/IRB(s) for review and approval/favourable opinion; (2) the revised form must be used to obtain consent from patients currently enrolled in the study if they are affected by the amendment; and (3) the new form must be used to obtain consent from new patients prior to enrolment.

If the revision is an administrative letter, investigators must inform their IEC(s)/IRB(s).

#### **18.1.2 Protocol Violations and Deviations**

All patients who are enrolled into the study, irrespective of whether or not they receive any treatment, will be followed according to the protocol regardless of the number of treatments received, unless consent for follow-up is withdrawn. Minor protocol deviations that do not result in harm to the study patients or significantly affect the scientific value of the reported results of the study will be recorded. In case of a major protocol deviation occurring (i.e. a deviation that could have a significant effect on the patient's safety, rights, or welfare and/or on the integrity of the study data), the Investigator must notify the Sponsor and the appropriate IEC/IRB as soon as possible or as per local requirements. Major protocol deviations that meet the criteria for a serious breach of GCP (e.g. a protocol violation, or non-reporting of critical safety information potentially jeopardising patients' safety) should be reported within 24 hours to the Sponsor. The Sponsor is required to report a serious GCP breach within 7 days to the applicable Health Authorities. Protocol deviations will be recorded on the source documents with an explanation for the deviation. No deviation from the inclusion/exclusion criteria will be permitted.

#### **18.1.3 Monitoring**

Before the trial can be initiated at a site, the prerequisites for conducting the trial must be clarified and approved by the Sponsor.

Representatives of the Sponsor (or designee) must be allowed to visit all study site locations periodically to assess the data quality and study integrity according to FDA, EU directives and ICH GCP. On-site they will review study records and directly compare them with source documents, discuss the conduct of the study with the Investigator and verify that the facilities remain acceptable.

Electronic Case Report Form completion and accuracy will be checked by performing source data verification that is a direct comparison of the entries made against the appropriate source documentation. Any resulting discrepancies will be reviewed with the Investigator(s) and/or his/her site staff and resolved. Monitoring procedures require that patients' informed consents, adherence to the inclusion/exclusion criteria and SAE documentation be verified. Additional monitoring activities maybe outlined in the study specific monitoring plan. The Sponsor may request adequately redacted source documents and hospital records for review by medical monitor (e.g. pathology reports, lab or investigations reports, consultation notes, cancer treatment history), this may help to better understand patient's general condition, emerging safety issues, effectiveness of their management and the efficacy.

The Sponsor must be informed immediately of any change in the personnel involved in the conduct of the trial.

#### **18.1.4 Audits and Site Inspections**

Authorised personnel from domestic and foreign regulatory authorities and the Sponsor's Quality Assurance specialist (or designee) may carry out inspections and audits respectively. The purpose of an audit/inspection is to ensure that ethical, regulatory and quality requirements are fulfilled in the Sponsor studies.

The Investigator will permit international, national and local health authorities, the Sponsor monitors, representatives, and collaborators, and the IECs/IRBs to inspect facilities and records relevant to this study. The Investigator should promptly notify the Sponsor or their authorised representative of any inspections scheduled by any regulatory authorities and promptly forward to the Sponsor or their authorised representative copies of any audit reports received.

### **18.2 DATA HANDLING AND RECORD KEEPING**

#### **18.2.1 Data Management**

Data will be entered in a timely manner, for each patient into the (Sponsor approved) clinical database via electronic data capture (EDC). The EDC application uses system controls to ensure that unauthorised users cannot access or modify data. All users must have successfully undergone EDC application training prior to entering data into the EDC system. Electronic Case Report Forms should be reviewed and electronically signed and dated by the Investigator or a designee. The eCRF system will be compliant with the FDA Code of Federal Regulations 21 Part 11 and EU Clinical Trial Directive (EC) No. 2001/20/EC.

It is the responsibility of the Investigator to ensure that the data included in the eCRF is accurate, complete and electronically signed where appropriate.

The data will be electronically verified through use of on-line checks during data entry and through programmed edit checks specified by the clinical team. Discrepancies in the data will be brought to the attention of the investigational site personnel. Data entered into the eCRF will be validated as defined in the data validation specifications. All updates to queried data will be made by authorised study site personnel only and all modifications to the database

will be recorded in an audit trail. Once all the queries have been resolved, eCRFs will be locked by password protection. Any changes to locked eCRFs will be approved by the Investigator. Once the full set of eCRFs have been completed and locked, the Sponsor will authorise database lock and all electronic data will be sent to the designated statistician for analysis. Subsequent changes to the database will then only be made by written agreement of the Sponsor.

Adverse events and medical/cancer history terms will be coded from the verbatim description (Investigator term) using the Medical Dictionary for Regulatory Activities. Prior and concomitant medications and therapies will be coded according to the World Health Organisation Drug Dictionary. Coding review will be performed by the Sponsor (or designee) prior to database lock.

At the end of the study, the clinical data will be transferred to the Sponsor and the investigative site will receive patient data for their site in a readable format that must be kept with the study records.

### **18.2.2 Study Documentation and Retention of Records**

The Investigator must maintain records of all study documents and supporting information relating to the conduct of the study. This documentation should include but is not limited to, protocols, eCRF data, SAE reports, patient source data, correspondence with health authorities and IEC/IRBs, ICFs, Investigator(s) and study team members' curricula vitae, monitor visit logs, laboratory reference ranges, laboratory certification or quality control procedures and laboratory director curriculum vitae (Essential documentation listed in ICH GCP [CPMP/ICH/135/95]). Patient files and other source data must be kept for the maximum period of time required by applicable regulations and guidelines or institution procedures or for the period specified by the Sponsor, whichever is longer. The Sponsor must be consulted if the Investigator(s) wishes to assign the study files to someone else, remove them to another location or is unable to retain them for the specified period.

The patient medical records must contain the product name/code, the trial reference code, trial subject code and administration dates and dose in order to ensure that a link can be made back to the identity of the product and the further traceability records of the Investigator and Sponsor.

### **18.3 CLINICAL TRIAL AGREEMENT, FINANCE AND INSURANCE**

This study will be conducted under a Clinical Trial Agreement between Autolus ("Sponsor") and the institution(s) representing the investigational study site(s) ("Investigator"). Financial support to the investigational site(s) will be detailed in the Clinical Trial Agreement. The Clinical Trial Agreement must be signed before the commencement of the study and will clearly delineate the responsibilities and obligations of the Investigator and the Sponsor, and will form the contractual basis under which the clinical study will be conducted.

A Certificate of Clinical Trials Insurance will be provided to the study centres by the Sponsor, where required. Details of the Sponsor's arrangement for clinical study insurance to provide for compensation to patients for any claim for bodily injury or death arising from participation in the clinical study are provided in the Patient Information Sheet.

## **19 PUBLICATION OF DATA AND PROTECTION OF TRADE SECRETS**

Both the EU Data Protection Regulations and the FDA Amendments Act mandates the registration with ClinicalTrials.gov of certain clinical trials of drugs (including biological products) and medical devices subject to the European Medicines Agency and FDA regulations for any disease or condition. The International Committee of Medical Journal Editors requires study registration as a condition for publication of research results generated by a clinical study (<http://www.icmje.org>).

All information supplied by the Sponsor in connection with this study and not previously published to the public, is considered confidential information (“Confidential Information”). This confidential information includes, but is not limited to, the Investigators’ Brochure, Clinical Study Protocol, eCRFs and other scientific data. Any data collected during the study are also considered confidential information. This confidential information shall remain the sole and exclusive property of the Sponsor, shall not be disclosed to others without prior written consent of the Sponsor, and shall not be used except in the performance of this study.

The information developed during the conduct of this study is also considered confidential information, and will be used by the Sponsor in connection with the development of AUTO3. The confidential information may be disclosed as deemed necessary by the Sponsor. To allow the use of the confidential information derived from this study, the Investigator is obliged to provide the Sponsor with complete test results and all data developed in this study.

The Sponsor has full ownership of the original eCRFs completed as part of the study.

By signing the Clinical Study Protocol, the Investigator agrees that the results of the study may be used for the purposes of national and international registration, publication, and information for medical and pharmaceutical professionals. The authorities will be notified of the Investigator’s name, address, qualifications, and extent of involvement.

It is agreed that, consistent with scientific standards, publication of the results of the study shall be made only as part of a publication of the results obtained by all sites performing the protocol.

Should the Investigator wish to publish the results of this study, the Investigator agrees to provide the Sponsor with a manuscript for review 60 days prior to submission for publication.

The Sponsor retains the right to delete from the manuscript, information that is confidential or proprietary and to object the suggested publication and/or its timing (at the Sponsor’s discretion).

In addition, if requested by the Sponsor, the Investigator shall withhold publication for an additional 6 months to allow for filing a patent application or taking other such measures as the Sponsor deems appropriate to establish and preserve its proprietary rights.

In the event that the Sponsor chooses to publish the data from this study, a copy will be provided to the Investigator at least 30 days prior to the expected date of submission to the intended publisher.

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## 21 APPENDICES

### Appendix 1: Response Criteria for Acute Lymphoblastic Leukaemia (National Comprehensive Cancer Network Guidelines)

Patients with acute lymphoblastic leukaemia (ALL) will be evaluated using Response Criteria for Acute Lymphoblastic Leukaemia (National Comprehensive Cancer Network Guidelines, version 2.2014) for documenting disease response as shown below.

<b>Response Criteria for Blood and Bone Marrow</b>	
Complete response (CR)	<ul style="list-style-type: none"> <li>No circulating blasts or extramedullary disease</li> <li>No lymphadenopathy, splenomegaly, skin/gum infiltration, testicular mass, or central nervous system (CNS) involvement</li> <li>Trilineage haematopoiesis and &lt;5% blasts</li> <li>Absolute neutrophil count &gt;1000/<math>\mu</math>L</li> <li>Platelet count &gt;100,000/<math>\mu</math>L</li> <li>No recurrence for 4 weeks</li> </ul>
CR with incomplete recovery of counts (CRi)	<ul style="list-style-type: none"> <li>Recovery of platelets to &lt;100,000/<math>\mu</math>L</li> <li>Recovery of absolute neutrophil count to &lt;1000/<math>\mu</math>L</li> </ul>
Overall response rate	Sum of CR and CR with incomplete recovery of counts.
Refractory ALL	Failure to achieve CR at the end of induction therapy.
Progressive ALL	Increase $\geq$ 25% in the absolute number of circulating or bone marrow blasts or development of extramedullary disease.
Relapsed ALL	Reappearance of blasts in the blood or bone marrow (>5%) or any extramedullary site after CR.
<b>Response Criteria for CNS Disease</b>	
CNS remission	Achievement of no lymphoblast in cerebrospinal fluid (CSF) (cytospin or flow) regardless of white blood cell (WBC) count in patients with WBC count <5/mcL or $\geq$ 5/mcL and presence of lymphoblasts in CSF at diagnosis.
CNS relapse	New development of WBC count $\geq$ 5/mcL and presence of lymphoblasts in CSF or clinical signs of CNS leukaemia such as facial nerve palsy, brain/eye involvement, or hypothalamic syndrome.
<b>Response Criteria for Mediastinal Disease</b>	
CR	Complete resolution of mediastinal enlargement by CT.
CR unconfirmed	Residual mediastinal enlargement that has regressed by >75% in sum of the products of the greatest perpendicular diameters.
Partial response	>50% decrease in the sum of the products of the greatest perpendicular diameters.
Progressive disease	>25% increase in the sum of the products of the greatest perpendicular diameters.
No response	Failure to qualify for partial response or progressive disease.
Relapse	Recurrence of mediastinal enlargement after achieving CR or CR unconfirmed.

ALL = acute lymphoblastic leukaemia; CNS = central nervous system; CR = complete response; CSF = cerebrospinal fluid; WBC = white blood cell.

## Appendix 2: Karnofsky Score

For assessment of eligibility in patients  $\geq 10$  years

100%	Normal, no complaints, no signs of disease
90%	Capable of normal activity, few symptoms, or signs of disease
80%	Normal activity with some difficulty, some symptoms, or signs
70%	Caring for self, not capable of normal activity or work
60%	Requiring some help, can take care of most personal requirements
50%	Requires help often, requires frequent medical care
40%	Disabled, requires special care and help
30%	Severely disabled, hospital admission indicated but no risk of death
20%	Very ill, urgently requiring admission, requires supportive measures or treatment
10%	Moribund, rapidly progressive fatal disease processes
0%	Death

### Appendix 3: Lansky Score

For assessment of eligibility in patients <10 years

100%	Fully active, normal
90%	Minor restrictions in strenuous physical activity
80%	Restricted in strenuous play, tires more easily, otherwise active
70%	Greater restriction of play <i>and</i> less time spent in play activity
60%	Up and around, but active play minimal; keeps busy by being involved in quieter activities
50%	Lying around much of the day, but gets dressed; no active playing participates in all quiet play and activities
40%	Mainly in bed; participates in quiet activities
30%	Bedbound; needing assistance even for quiet play
20%	Sleeping often; play entirely limited to very passive activities
10%	Doesn't play; does not get out of bed
0%	Unresponsive

**Appendix 4: Eastern Cooperative Oncology Group Performance Status Score**

<b>Grade</b>	<b>Eastern Cooperative Oncology Group Performance Status</b>
0	Fully active, able to carry on all pre-disease performance without restriction
1	Restricted in physically strenuous activity but ambulatory and able to carry out work of a light or sedentary nature, e.g. light house work, office work
2	Ambulatory and capable of all self-care but unable to carry out any work activities. Up and about more than 50% of waking hours
3	Capable of only limited self-care, confined to bed or chair more than 50% of waking hours
4	Completely disabled. Cannot carry on any self-care. Totally confined to bed or chair
5	Dead

Source: Eastern Cooperative Oncology Group, Robert Comis M.D., Group Chair (Oken et al. 1982).

## **Appendix 5: Highly Effective Methods of Birth Control**

For females of childbearing potential (defined as <2 years after last menstruation or not surgically sterile), a negative serum or urine pregnancy test must be documented at screening, prior to pre-conditioning and confirmed before receiving the first dose of study treatment.

For females who are not post-menopausal (24 months of amenorrhea) or surgically sterile (absence of ovaries and/or uterus), highly effective methods of contraception must be used during the treatment period and for at least 12 months after the last dose of study treatment. They must agree not to donate eggs (ova, oocytes) for the purposes of assisted reproduction during the study and for 12 months after receiving the last dose of study drug.

For males, it must be agreed that a barrier method of contraception should be used and that sperm will not be donated during the treatment period and for at least 12 months after the last dose of study treatment.

### **Birth Control Methods Considered Highly Effective:**

For the purpose of this guidance, methods that can achieve a failure rate of less than 1% per year when used consistently and correctly are considered as highly effective birth control methods. Such methods include:

1. Total abstinence (when this is in line with the preferred and usual lifestyle of the patient. Periodic abstinence (e.g. calendar, ovulation, symptothermal, post-ovulation methods) and withdrawal are NOT acceptable methods of contraception.
2. Female sterilisation (have had surgical bilateral oophorectomy with or without hysterectomy) or tubal ligation at least 6 weeks before taking study treatment. In case of oophorectomy alone, only when the reproductive status of the woman has been confirmed by follow-up hormone level assessment.
3. Male sterilisation (at least 6 months prior to screening). For female patients on the study the vasectomised male partner should be the sole partner for that patient.
4. BOTH of the following forms of contraception must be utilised:
  - a. Use of oral, injected or implanted hormonal methods of contraception or other forms of hormonal contraception that have comparable efficacy (failure rate <1%), for example hormone vaginal ring or transdermal hormone contraception.
  - b. Barrier methods of contraception: Condom or Occlusive cap (diaphragm or cervical/vault caps) with spermicidal foam/gel/film/cream/vaginal suppository.
5. Use of intrauterine devices are excluded due to increased risks of infection and bleeding in this population.
6. In case of use of oral contraception, females must be stable on the same pill for a minimum of 3 months before taking study treatment.

### **Birth Control Methods Which May NOT Be Considered as Highly Effective**

Acceptable birth control methods that result in a failure rate of more than 1% per year include:

- Progestogen-only oral hormonal contraception, where inhibition of ovulation is not the primary mode of action.
- Male or female condom with or without spermicide.<sup>2</sup>
- Cap, diaphragm, or sponge with spermicide.<sup>2</sup>

### **Birth Control Methods Which are Considered UNACCEPTABLE in Clinical Trials**

Periodic abstinence (calendar, symptothermal, post-ovulation methods), withdrawal (coitus interruptus), spermicides only, and lactational amenorrhoea method are not acceptable methods of contraception. A female condom and male condom should not be used together.

### **Paediatric Patients Acceptable Methods Include**

Females who are not of reproductive potential (defined as either <11 years of age, Tanner Stage 1, post-menopausal for at least 24 consecutive months or have undergone hysterectomy, salpingotomy, and/or bilateral oophorectomy) are eligible without requiring the use of contraception. Acceptable documentation includes written or oral documentation communicated by the clinician or clinician's staff of 1 of the following:

- a. Demographics show age <11.
- b. Physical examination indicates Tanner Stage 1.
- c. Physician report/letter.
- d. Operative report or other source documentation in the patient record.
- e. Discharge summary.
- f. Follicle stimulating hormone measurement elevated into the menopausal range.

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<sup>2</sup> A combination of male condom with either cap, diaphragm, or sponge with spermicide (double barrier methods) are also considered acceptable, but NOT highly effective, birth control methods.

## Appendix 6: International Study for Treatment of Childhood Relapsed Acute Lymphoblastic Leukaemia Definition of Risk Groups in Relapsed Paediatric Acute Lymphoblastic Leukaemia

The time-point of relapse is defined in relation to the date of primary diagnosis and the date of the end of frontline therapy. Completion of primary therapy is defined as the end of anti-leukaemic therapy according to the frontline protocol. In most cases, it is the end of maintenance therapy, but may also be the last treatment after interruption of the intensive treatment or of an inadequately short primary therapy. Since the duration of maintenance therapy varies between different protocols and individual patients (in most patients and protocols duration of treatment is 24 months), completion of primary therapy is a flexible time-point in contrast to time-point definitions referring to the date of primary diagnosis.

In the rare case of inadequately short frontline therapy (e.g. according to B cell Non-Hodgkin lymphoma/acute lymphoblastic leukaemia [ALL] protocols) the interval after completion of primary therapy is the decisive one for time-point classification. The site of relapse is classified based on conventional light microscopy using the French-American-British criteria. In morphologically undetermined situations such as bone marrow (BM) involvement of around 5% in extramedullary relapse, minimal residual disease quantification may be regarded as secondary criterion to determine the extent of BM involvement.

### Definition of Time-Point of Relapse

Time-point	After primary diagnosis	and	After completion of primary therapy
Very early	<18 months		<6 months
Early	≥18 months		<6 months
Late			≥6 months

### Definition of Site of Relapse

Bone marrow		M1 (<5% blasts)	M2 (≥5% and <25% blasts)	M3 (≥25% blasts)
Extramedullary relapse	No	No ALL relapse	Requires follow-up control	Isolated BM relapse
	Yes	Isolated extramedullary relapse	Combined BM/extramedullary relapse	

ALL = acute lymphoblastic leukaemia; BM = bone marrow.

### Definition of International Study for Treatment of Childhood Relapsed ALL Standard Risk (SR)/High Risk (HR) 2010 Risk Groups

Site Time-point	Immunophenotype: B cell precursor		
	Extramedullary isolated	BM combined	BM isolated
Very early	HR	HR	HR
Early	SR	SR	HR
Late	SR	SR	SR

BM = bone marrow; HR = high risk; SR = standard risk.